Alternative protein sources for organic poultry

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Published in:
Proceedings of the XV's European Poultry Conference

Publication date:
2018

Document Version
Publisher's PDF, also known as Version of record

WORLD'S POULTRY SCIENCE JOURNAL

education
organization
research

THE XVth EUROPEAN POULTRY CONFERENCE

DUBROVNIK, CROATIA
17th to 21st September 2018

Conference Information and Proceedings

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Editors: Estella Prukner-Radovčić, Helga Medić

Hosted by the Croatian Branch of Worlds Poultry Science Association,
and organized under the auspices of the WPSA
and the European Federation of the WPSA

WORLD'S POULTRY
SCIENCE ASSOCIATION
CROATIAN BRANCH

The XVth European Poultry Conference
Dubrovnik, Croatia
17th to 21th September 2018
Conference Information and Proceedings

Publisher:
Croatian Branch of the World's Poultry Science Association
under the hospice of the World's Poultry Science Journal.
Udruga za znanosti o peradi,
Heinzelova 55, 10000 Zagreb, Croatia

Production:
Acting as agents for the Croatian Branch of the WPSA;
the layout of the supplement of the World's Poultry Science Journal
has been built by Linear Studio d.o.o., Dubrovnik

Printed by: Alfa-2, Dubrovnik, Croatia

Printed in September, 2018.

Edition: 1600 copies

ISBN/EAN: 978-90-829157-0-9

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Welcome

Chairperson of the 15th European Poultry Conference
Estella Prukner-Radovčić

Dear Colleagues and Friends,
On behalf of the Croatian Branch of the Worlds Poultry Science association, I would like to welcome all the participants to Dubrovnik and to the 15th European Poultry Conference which is held for the first time in Croatia, but also in this part of Europe.

We were pleasantly surprised by the response from participants, they are coming from 71 countries located on all continents. A large number of poultry scientists and specialists gathered at this Conference to study and discuss the most recent advance in poultry science. The conference program carefully prepared in cooperation with the chairman of the Working groups and the scientific committee will be able to get an overview of various relevant developments in the area of poultry science. We believe that we have assembled an excellent scientific programme emphasising the latest advancements, appropriately interspersed with up-to-date information. It will be focused on the main issues currently related to poultry research and production in order to add improve knowledge in a world where information and innovations are spread rapidly.

This conference would not have been possible without the tenacious persistence and long-term hard work of all of the committee chair and members. The organisers are indebted to many companies, institutes, and universities that have contributed towards the conference during its preparation. Without the generous contributions of our sponsors and dedicated members of the committees, it would not have been possible to organise this event. I thank PCO Adria Congress for their input to the staging of the conference. In particular, I express my gratitude to the president of the European Federation of the WPSA Prof Birger Svihus, the secretary and treasurer Prof Michael Grashorn, and the general secretary of the WPSA Dr Roel Mulder for advice and support with the preparation of this conference. Their suggestions and recommendations, based on a huge experience, allowed me to set up the EPC2018 for you to enjoy and remember. Lastly, I would like to express my deep appreciation to all who supported me in the organisation of this conference.

I hope that scientific presentations and the printed Proceedings will serve as a valuable source of additional information and will also remind the participants of the impressive days in Dubrovnik as a place of meeting old and getting new friends. Dubrovnik one of the most beautiful towns in the Mediterranean will be, I hope, enough challenging even for those who are just wanted to hear what’s new in poultry science.

I’d like to thank each of your for attending our conference and bringing your expertise to our gathering.

Welcome

President of the European Federation of the WPSA Branches (2014–2018)
Birger Svihus

Since the first meeting in Utrecht in 1960, the European Federation of World's Poultry Science Association (WPSA) has initiated a vast number of meetings within poultry science in the European and Mediterranean area. In addition to the multidisciplinary European poultry conferences, a number of more specialised meetings organized under the auspices of our many working groups, have been organized.

The aim of WPSA is to contribute to the dissemination of knowledge in poultry science. The World's Poultry Science Journal, skilfully edited by Dr Lucy Waldron, is one major contribution to this aim. However, even more important is meetings like this one to achieving our goal. Our scientific symposia and conferences allows for the presentation and critical discussion of new insights in poultry science, and equally important, allows for the mutual exchange of experiences and knowledge between academia and industry. I do not think it is too bold to claim that this unique cooperative spirit between academia and industry has played a key role in the ascent of poultry production to the pinnacle of animal production, as the largest animal industry in the world in regards to the production of (solid) food. The current European Poultry Conference, the 15th of a long series of successful conferences, has been skilfully planned and organized by the Croatian branch of WPSA. I hereby would like to use the opportunity to thank the organizers, not the least chairperson of the Conference prof. Estella Prukner-Radovčić and chairperson of the Scientific Committee prof. Helga Medić. I know that they have worked very hard over a long time to make sure every participant will have an enjoyable time, both professionally and socially. Without such volunteer activity of key persons in the local branches, WPSA would not have been able to succeed in its aim of contributing to insights and knowledge within poultry science and production.

Our meetings are not only a place for exchange of knowledge, but also serves an important role in forging bonds and mutual understanding and respect between countries, disciplines and people with different roles in the sustainable and efficient production of healthy poultry products. As such, the venue, with its beautiful surroundings and ancient history of a great civilization, will provide the perfect surroundings to forge these bonds.

I wish you all a great conference, full of memorable and rewarding encounters with poultry people from all over the world, and filled with inspired moments in regards to both science, culture and nature.

Welcome

Chairperson of the Scientific Committee
Helga Medić

Dear colleagues,

On behalf of the scientific committee I would like to welcome you all to the 15th European Poultry Conference.
The conference is dedicated to challenges and opportunities in poultry production with an emphasis on productivity, welfare and sustainability.
The multidisciplinary approach will bring scientists and experts together to present, discuss and highlight the latest achievements in the poultry science and their possible practical application in the field. We have received a large number of abstracts from all over the world and the members of the scientific committee had a difficult task to choose some of them as oral presentations.
The conference programme consists of six plenary and 22 parallel sessions, out of which 238 oral presentations and 284 posters. The plenary lectures will be given by the outstanding scientists whose topics cover the most important areas of poultry science and production.
In addition to an abundant scientific and social programme, the participants can discover the beautiful city of Dubrovnik, which is known as the pearl of the Adriatic. Due to the moderate Mediterranean climate, proximity of the sea and nature surroundings, September in Dubrovnik is the ideal time and place for this conference.
Dubrovnik, the capital of historic Dubrovnik Republic is a late medieval planned city which kept its extraordinary urban character defined by his famous city walls. As such, Dubrovnik is a UNESCO World Heritage site.
The uniqueness of Dubrovnik manifests in the skill of successful diplomatic art of preserving its freedom and independence through centuries. In addition to the Dubrovnik diplomacy, its cultural and scientific heritage are also unbeatable. From scientific achievements of Ruđer Bošković and Marin Getaldić, medical scientist Đuro Baglivi, literary works of Marin Držić who is often called Croatian Shakespeare, international influence of painter Vlaho Bukovac, to economist Benedikt Kotruljević who invented double bookkeeping which leads entire world economy today.

I would like to take this opportunity to thank our invited speakers and chairpersons for accepting our invitation to serve their sessions. I am very grateful to the members of the scientific and organizing committee, Adria Congress Ltd. and others who have worked very hard on the preparation of this conference. A special word of thanks goes to Dr. Birger Svihus for his sustained help and valuable suggestions.
At the end, I would like to express my personal thanks to all participants, especially the speakers and poster presenters, who have contributed to this conference. It wouldn't be such a success without them.

XVth European Poultry Conference

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Prof. Dr. Estella Prukner-Radovčić

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2. Nutrition, Dr Sanna Steenfeldt (Denmark)
3. Breeding and Genetics, Dr Steffen Weigend (Germany)
4. Egg Quality, Dr M. M. Bain (United Kingdom)
5. Poultry Meat Quality, Dr M. Petracci (Italy)
6. Reproduction, Dr Marleen Boerjan (The Netherlands)
7. Hygiene and Pathology (covered by WVPA)
8. Waterfowl (transferred to the Asia Pacific Federation)
9. Poultry Welfare and Management, Mrs Virginie Michel (France)
10. Turkeys, Prof Dr Hafez M. Hafez (Germany)
11. Education and Information, Dr Eva Sossidou (Greece)
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Working group 1 - Poultry Economics and Marketing

The working group for Poultry Economics and Marketing had their 6th round table in The Hague, the Netherlands in October 2017. The meeting was organised by the Dutch members of the working group: Peter van Horne (chairman of wg1), Gemma Tacken en Izak Vermeij. On the first day of the round table wg1 had a general session on 'economics of bio-security' with invited speaker Mr Musa Fresiji (Egypt) and Barbara Grabkowsky (Germany). On Thursday morning several members gave a country report on the actual situation of the poultry sector in their country. On Thursday afternoon wg1 had a joint meeting with some members of the Dutch en Belgian WPSA branch. Speakers were Maro Ibaruru-Blanc of Iowa State University in the USA, Sean Wennerlind, Canada, Francois Cadual, ITAVI France and Peter van Horne, Wageningen Economic Research. On Friday the group had country report and other short communication and afterwards a visit to broiler farm with slow growing broilers (production according to the better life one star system).
The next meeting of working group will be in Germany in october 2019.
Working group 1 has 25 members in 12 countries.

Chairman Peter van Horne
peter.vanhorne@wur.nl
Wageningen, July 2018

Working Group 2 - Nutrition

The WG2 has a long history since the first activities were initiated almost 45 years ago and from 1965 and until 1977 the WG2 held their scientific meetings on a regular basis, often in connection with the European Conferences. The first European Symposia on Poultry Nutrition was held in Denmark in 1977 with 73 delegates and since then the European Symposia on Poultry Nutrition (ESPN) have been organised bi-annually at different locations in Europe.
The first chairperson was Prof. J. Baelum, Denmark (1965-1966), followed by Prof. Reyntens, Belgium (1966-1973), Dr. W. Bolton, UK (1973-1978), Prof. H. Vogt, Germany (1978-1986), Dr. C. Fisher, Scotland (1986-1990), Dr. N. Rand, Israel (1990-1997), Dr. M. Larbier, France (1997-2000), Dr. C. C. Whitehead, Scotland (2000-2009), Dr. Steenfeldt, Denmark (2009-).

The ESPN is a popular and scientific event, which is an important part of the activities carried out in WG2, and the members of WG2 has always been active in the planning of these Symposia in close co-operation with the local branches and organising committees in order to assemble an interesting scientific programme. The ESPN has attracted an increasing number of delegates being close to 600-700 during the last symposia held in Edinburgh in 2009, in Cesme in 2011 and in Potsdam in 2013. The 21th ESPN in Spain in 2017 was attended by over 1500 delegates from 62 countries. The structure of the ESPN are organised in a special way in order to stimulate the delegates to be active in the discussions of the scientific topics presented by invited speakers. There are no parallel sessions and in each session 3-4 speakers are covering different aspects of a topic, where after the audience is divided into a number of discussion groups that will discuss topics raised

during the speaker session.
The WG2 meets regularly in connection with the ESPN or during the European conferences and world poultry congresses. At present, the WG2 has 31 members representing 18 European countries. During the years, several sub-committees have been generated under the WG2 in order to solve specific problems, resulting in publication as "European Tables of Energy Values for Poultry Feedstuffs" and "the European amino acid tables". The phosphorus sub-committee was established during the ESPN in 2009, and their first WG report, "Determination of phosphorus availability in Poultry" was published in the WPSJ in 2013. The "Results of an international P digestibility ring test with broiler chickens", was published in Poultry Science in 2017, where several members of the P-sub-group were involved.
The next symposia will be held in Gdansk, Poland in June 2019 and WG2 had a meeting in June 2018 in Gdansk to plan and discuss the scientific program and the challenges regarding organising, which has increased due to the high number of delegates expected.
Additional information about the WG2 can be obtained from Sanna Steenfeldt, Aarhus University, Denmark, sanna.steenfeldt@anis.au.dk

Working Group 3 - Breeding and Genetics

Historically, WG3 had three main activities: networking, summary of random sample tests, and scientific meetings. Networking activity of WG3 included the preparation of a diary of on-going research projects between 1992 and 1999 (more than 100 projects were listed from different countries). Random sample tests (RST) for laying hens were organized in many countries to support egg producers with independent information on the performance profiles of different strains under comparable environmental conditions. Over the years, RST for laying hens was repeatedly on the agenda of WPSA Working Group 3 "Breeding and Genetics" (WG3). However, only few tests were remaining at the turn of the new century. Therefore, members of WG3 decided at their business meeting in 2003 to suspend activities to prepare summary reports for European testing stations until more information was available.
For many years, an important activity of WG3 has been to organize Poultry Genetics Symposia. In former times Working Group 3 'Breeding and Genetics' organized meetings or sessions during European poultry conferences. Initially, these meetings were organised on specific issues, with the example of the Symposium on Genotype-Environment Interactions in Poultry Production organised in 1989 in France. At this time, there was another regular flow of poultry genetics meetings in Western and Eastern Europe since the early 1960s which were attended by WG3 members but operated independently from WPSA. The British Poultry Breeders' Round Table (BPBRT) started in 1959 as a platform to exchange information and novel ideas related to quantitative genetics theory and application in poultry breeding among scientists from the breeding industry and academia. Annual meetings attracted a large number of participants from Europe and North America until the end of the 1980s. As a result of globalization of the poultry breeding industry, with fewer potential sponsors and active poultry geneticists in the UK, it was decided to change the name to "European Poultry Breeders' Roundtable" (EPBRT) and to meet only biannually starting with Oxford/UK in 1993, followed by Foulum/DK in 1995. During the 1970s and 1980s when the different political systems prevented regular communication among colleagues from Eastern and Western Europe, poultry genetics meetings were organized in Eastern Europe under the name "AVIAGEN - Current Problems in Avian Genetics". After 1989, colleagues from East and West attended both types of meetings and began to think of a possible merger. The 12th AVIAGEN Symposium and the 3rd EPBRT were organised consecutively in Pruhonice near Prague (Czech Republic) in the same week, and it was then decided to merge the organizations under the umbrella of "Working Group 3" of WPSA. It was also agreed that the Symposia should be biannual, with venues alternating between "East" and "West", giving the opportunity to discover several research institutions across Europe.

So far the following symposia were held:
- 1st Symposium 1999 in Mariensee, Germany
- 2nd Symposium 2001 in Gödöllö, Hungary
- 3rd Symposium 2003 in Wageningen, The Netherlands
- 4th Symposium 2005 in Dubrovnik, Croatia
- 5th Symposium 2007 in Braedstrup, Denmark
- 6th Symposium 2009 in Bedlewo, Poland
- 7th Symposium 2011 in Peebles, United Kingdom
- 8th Symposium 2013 in Venice, Italy
- 9th Symposium 2015 in Tuusula, Finland
- 10th Symposium 2017 in Saint Malo, France

These meetings provide a forum to exchange recent results and current research activities, encourage interaction between junior and senior scientists, and between colleagues from industry and research institutes. The interaction of the genetics symposium with other disciplines will be continued and academic scientists and scientists from commercial companies will continue to be involved in preparing the scientific programme to ensure attractiveness and industrial relevance of the symposium. The next symposium will be held in the beautiful town Prague, Czech Republic, in 2019.

Additional information about the WG3 and its coming activities can be obtained from the Chairman:
Dr. Steffen Weigend, Germany
steffen.weigend@fli.de.

Working Group 4 - Quality of Egg and Egg Products

The aim of WG4 is to provide a platform that stimulates discussion between European scientists and those working in the egg and egg products industries. Our aim is to exchange knowledge and work towards developing new collaborative projects that advance knowledge and provide solutions to enhance the quality and safety of egg and egg products. A full synopsis detailing the work of WG4 can be found on the WPSA website: http://www.wpsa.com/index.php/federations/european-federation/2014-03-08-15-52-25/quality-of-eggs-and-egg-products-wg-4 .

WG4 currently has representatives from 21 countries and meets every 2 years at a key WPSA event. Every other year the group holds its own joint 'EGGMEAT' symposia with

WG5 (Poultry Meat quality). The aim of these symposia is to bring likeminded people together to discuss hot topics in the field of egg and meat quality. Topics range from the structure and properties of the egg to meat quality issues, from the microbiological safety of meat and egg products to economics and marketing. Our last symposia (EGGMEAT2017) took place in Edinburgh, UK in September 2017 and was attended by 170 delegates from Europe and North America. You can read more about EGGMEAT2017 on the WPSA website: http://www.wpsa.com/index.php/57-news-latest/news-archive/277-report-eggmeat-2017

EGGMEAT2019: The Turkish Branch of WPSA will host our next EGGMEAT symposia in Çeşme-Izmir, Turkey from the 23rd -25th June 2019. Çeşme is a coastal town located at the end of Western Anatolia and is surrounded on three sides by the Aegean Sea. It is a prominent centre of international tourism in Turkey, famous for its excellent holiday accommodations, restaurants and entertainment facilities. Further details about EGGMEAT2019 are available on the conference website http://www.eggmeat2019.com/index.html#divdavet.

EPC2018: Members of WG4 attending EPC2018 are cordially invited to attend the next meeting of the group which will take place on Thursday 20th September 2018. Key items for our agenda will be the scientific program for EGGMEAT2019 and Horizon 2020 funding opportunities under work program 2018-2020 (http://ec.europa.eu/research/participants/data/ref/h2020/wp/2018-2020/main/h2020-wp1820-food_en.pdf) . Details of the venue and time of our meeting will be made available during the conference.

For further information about WG4 membership and associated activities please contact Prof Maureen Bain (maureen.Bain@glasgow.ac.uk)

Working Group 5 - Poultry Meat Quality

This Working Group looks back on a long history. As the first Symposium was organized in 1973 in Roskilde (DK) the establishment of the group was around in the early 1970s. The main objectives of this group are to provide the different research teams in the branches of European Federation with basic knowledge in the whole field of poultry carcass and meat quality. Members contribute to exchange of information between the different research institutes and to the development of new research topics for international cooperation. Working group activities are presented to a broader audience during Working Group Symposia and specialized sessions of the European Poultry Conferences and World's Poultry Congresses.

The combined expertise of the Working Group members resulted in the several recommendations for the assessment of carcass and meat quality traits in the past. The first recommendation dealt with standardizing terms for poultry carcass parts (Terms used for parts of poultry in different languages; WPSJ 39, 64-73, 1983) and the second one with a recommendation for the sensory assessment of poultry meat (Recommendation for a standardized method of sensory analysis for broilers. WPSJ 43, 64-68, 1987). Several attempts have also been made to harmonize the methodologies for the evaluation of poultry meat quality and finally a document for the assessment of chemical and physical criteria of poultry meat quality has been recently published (Harmonization of methodologies for the assessment of poultry meat quality features. WPSJ, 67, 137-151, 2011).
Since foundation of the group, Working Group Symposia took place regularly in biannual intervals:

Edition	Year	Location	Edition	Year	Location
1st	1973	Roskilde (DK)	13th	1997	Poznan (PL)
2nd	1975	Oosterbeek (NL)	14th	1999	Bologna (I)
3rd	1977	Grub (D)	15th	2001	Kuşadasi (TR)
4th	1979	Norwich (UK)	16th	2003	St. Brieuc (F)
5th	1981	Apeldoorn (NL)	17th	2005	Doorwerth (NL)
6th	1983	Ploufragran (F)	18th	2007	Prague (CZ)
7th	1985	Vejle (DK)	19th	2009	Turku (FIN)
8th	1987	Budapest (H)	20th	2011	Leipzig (D)
9th	1989	Stuttgart (D)	21th	2013	Bergamo (I)
10th	1991	Doorwerth (NL)	22th	2015	Nantes (F)
11th	1993	Tours (F)	23th	2017	Edinburgh (UK)
12th	1995	Zaragoza (E)			

Since 1989, the Symposium is run in parallel with the European Symposium on "Quality of Egg and Egg products" with an overall attendance of 200-250 participants coming from all over the World.

In its history, the working group was chaired by D.H. Shrimpton (UK), S. Scholtyssek (D), J. Fris Jensen (DK), G.C. Mead (UK), T.G. Uijttenboogaart (NL), P. Colin (F), R.W.A.W Mulder (NL), M. Grashorn (D), M.J. Duclos (F) and M. Petracci (I). Currently, the Working Group included 16 delegates from 12 Countries of the European Federation. The next EggMeat Symposium will be held in Çeşme (TR) from 23 to 26 June, 2019 (www.eggmeat2019.com).

Further details about Working Group 5 can be obtained from the current Chairperson:
Prof. Massimiliano Petracci
m.petracci@unibo.it

Working Group 6 - Reproduction

Sub-committee No. 6 ("Physiological Aspects of Incubation") of the European Federation of Branches of the World's Poultry Science Association met on the 15th May 1975 at the Poultry Research Centre at Roslin, Midlothian, Scotland. Mr Harry Lundy, who was Head of the Environmental Physiology section at Roslin, organised a highly successful scientific programme which attracted a large audience. During the meeting a number of people expressed an interest in forming an informal Incubation Research Group. The first meeting of this group took place on 15 and 16 July 1976 at the University of Bath, and in April 1978, a meeting was held in conjunction with the British Ornithological Union at Royal Holloway College, London. The Group held another meeting in 1978 at the Wildfowl Trust at Martin Mere. In 1980 the Group met at the University of Bath, and at this meeting the Incubation Research Group became more formal; it was agreed to hold annual meetings, which have been held ever since (almost). Occasionally , there have also been joint meetings with other organisations: with the Immunology Society in 1976, with the Society of Applied Biology in 1982, and in 1983 with the British Veterinary Poultry Association.

The Group has remained active with well-attended meetings, its longevity may be due to the informal atmosphere of its meetings and the wide range of topics covered. Nowadays, the meetings are truly international attended by incubationists from all continents and participants from research, commerce, industry and the academic world. The name of the group was the Incubation Research Group (IRG) up to 1995 when it was decided to expand the remit of the group to include fertility and the name was changed to the Incubation and Fertility Research Group (IFRG). In 1998, an association was made with the "World's Poultry Science Association" when the IFRG adopted the European Branches' Working Group 6 - Reproduction.

The Chairs of Working Group 6 - Reproduction have been,
Dr Glenn Baggott 1998-2009
Dr Nick French 2009-2012
Marleen Boerjan 2012-

Meetings are annually, and have been held the following places:
1975: Poultry Research Centre, Edinburgh, UK
1976: University of Bath, UK
1977: University of Edinburgh, UK
1978: Royal Holloway College, London, UK
1978: Wildfowl Trust, Martin Mere, Lancashire, UK
1980: University of Bath, UK
1981: Poultry Research Centre, Edinburgh, UK
1982: Royal Veterinary College, London, UK
1983: University of Bath, UK
1984: Tarporley, Cheshire, UK
1985: Wildfowl Trust, Slimbridge, Gloucestershire, UK
1986: London Zoo, UK
1987: University of Reading, UK
1988: Game Conservancy, UK
1990: Harper Adams Agricultural College, Shropshire, UKJ
1991: Wildfowl Trust, Slimbridge, UK
1992: Birkbeck College, London, UK
1993: Scottish Agricultural College, Ayr, UK
1994: University of Warwick, UK
1995: University of Warwick, UK
1996: University of Warwick, UK
1997: Scottish Agricultural College, UK
1998: University of Warwick, UK
1999: Vinci Conference Center, Tours, France
2000 : St Edmund's Hall, University of Oxford, UK

2001 : St Edmund's Hall, University of Oxford, UK
2002 : St Edmund's Hall, University of Oxford, UK
2003 : University of Lincoln, Lincoln, UK
2004 : University of Lincoln, Lincoln, UK
2005 : University of Lincoln, Lincoln, UK
2006: University of Lincoln, Lincoln, UK
2007: University of Edinburgh, Edinburgh, UK
2008: Moorfields Hospital, London, UK
2009: University of East Anglia, Norwich, UK
2010: Tours, France
2011: Ede, The Netherlands
2012: Pisa, Italy
2013: Gottingen, Germany
2014: Lunteren, The Netherlands
2015: Berlin, Germany (Combined with Working Group 12)
2016, Brugge, Belgium
2017: Wageningen, The Netherlands
2018: Edinburgh, Scotland

Working Group 9 - Poultry Welfare and Management

Working group IX "Poultry Welfare and management" has actually 33 members and 4 guests.

The last Poultry Welfare Symposium was organized in France by the WG IX and the French branch of WPSA (Ploufragan, 2017 locally organized by Virginie Michel and coll) and was a full success with the highest number of participants ever registered (201 participants from 27 countries). Fifty three oral communications and sixty posters had been presented on the following topics:

- Broiler breeders welfare and effect of early age experience on welfare
- Cognition: another way to investigate welfare?
- Feather pecking : origins and prevention
- New methods of on farm killing and of slaughter
- Welfare at depopulation and during transport
- New contributions of PLF for behavioural assessment
- On field Welfare Monitoring
- Poultry production sustainability and consumer perception

A satellite workshop on the Broiler Breeder paradox had been organized with 37 participants (organized by Sabine Gebhardt and coll) as well as 3 different technical tours in poultry farms with 50 participants. At this symposium the chairmanship of the WG IX changed from Valentina Ferrante to Virginie Michel.

The annual meeting of WG IX was held in Dublin in April 2018 and it was decided that the next European Symposium will certainly be organized in Brno (Czech Republic, locally organized by Martina Lichovnikova and coll). During the annual meeting, others topics discussed were: members country reports, keel bone damage problematic, Hennovation project, Business Benchmark on Farm Animal Welfare, welfare indicators in broiler chicken and an EFSA report on low atmosphere pressure stunning.

The next meeting in April 2019 will be organized in Brno to visit the facilities and assess the proposal for the next Symposium.
Virginie Michel **virginie.michel@anses.fr**, 28/08/2018

Work Group 10 - Turkeys

9th "Hafez" International Symposium on Turkey production and Health: Challenges and opportunities, Berlin, Germany, 18th - 20th May 2017

To strengthen the dialog between the persons that are involved in the different production stages, especially veterinarians and agriculturists, but also economists, and to exchange new results of scientific works and practical experiences we organized in Berlin since 2000 on the behalf of the World Poultry Science Association - Federation of European Branches (WPSA), the international symposiums for the working group 10 (Turkey).

The aim of the symposium is to stimulate the exchange of current information and ideas related to turkey production and health as well as to focus the attention on specific problems and encouraging new approaches to solve them.
2017 symposium, which held between 18th - 20th May 2017 with the title **"Turkey Production and Health: Challenges and opportunities**" was attended by 92 scientists and experts from 19 countries (Algeria, Austria, Brazil, Canada, Egypt, Finland, France, Germany, Israel, Italy, Morocco, Nigeria, Syria, Sweden, The Netherlands, Tunis, Turkey, United Kingdom and United States of America).

Currently several factors and problems face and influence the turkey production worldwide. Today's intensive turkey production, located in an area of conflict between consumer protection, animal welfare and economics, requires a lot of knowledge about breeding, husbandry, nutrition, diseases, slaughter and marketing as well as the governmental regulations and legislations related to turkey production and health. Many human foodborne bacterial infections were linked to poultry. Control and/or elimination of these organisms present a great challenge. The development of antibiotic resistant bacteria will also be a continuous public health hazard. On the other hand, only a few authorised pharmaceutical veterinary products will be available for the treatment of turkeys as food producing animals. Vaccination today and in future is regard as one of the most beneficial interventions to prevent diseases. The use of future progressive vaccine production can significantly reduce the cost of vaccines, ensure better efficacy, and allow easy and rapid intervention to face the steady mutation of the microorganisms. Furthermore, the development of efficient vaccines against bacterial infections will lead to a reduction of the use of antibiotics and subsequently will reduce the development of resistant bacteria. In general, consumer expectations for high quality products will strongly influence future production methods. This means that farmers, veterinarians, stockholders and all other partners involved in the production chain need to share more responsibilities.
At the symposium, 29 oral presentations in several sections were held. Topics related to

the current and future challenges of turkey production and health in several countries were given. Furthermore, papers covered exist problems and solution through the entire production chain from hatchery to slaughterhouse. These includes, the effect of stress, light, rearing management as well as stocking density on animal welfare aspects and performances. In addition, several papers dealt with the effect of feeding quality on performances, litter quality, footpad health were presented. Papers dealt with current health problems and their diagnostic, problem related antibiotic resistance and control methods, with special attention to histomoniasis and avian influenza (H5N8) and problem related trade with turkey and turkey products were held.
The proceedings (181 pages) with the complete articles in English was published by the Mensch & Buch Verlag (ISBN 3978-3-86387-884-9)

Last but not least, I would like to express my gratitude to all individuals who worked in the planning and co-ordination of that event, especially the staff of the Institute of Poultry Diseases at the Free University Berlin.

The 10th "Hafez" International Symposium on Turkey Production will be hold in Berlin, Germany; May/June 2019. I hope to see you in Berlin.

Prof. Dr H. M. Hafez, Chair of the Working group 10
World's Poultry Science Association, Federation of European Branches
Institute of Poultry Diseases, Free University Berlin
Königsweg 63, 14163 Berlin, Germany

Working Group 11 Report (2017-2018)

Chair: Evangelia N. Sossidou, Veterinarian-Senior Research Scientist
Monday, 06 August 2018

1. Events

6th Mediterranean Poultry Summit
Torino, Italy, June 18-20, 2018
All information available at: http://www.mpn-wpsa.org/main/

Mediterranean Poultry Network (MPN) of World's Poultry Science Association (WPSA) was established in 2008. The MPN of the WPSA presently operates directly under the umbrella of Working Group (WG) 11 (Education and Information) of WPSA's European Federation. The 6th Mediterranean Poultry Summit was held on the grounds of the University of Torino, which is one of the oldest universities in Europe with excellent facilities for the size of this event.

2. Meetings

WG11 members who participated in the Summit have been invited to a Group Meeting to take decisions about the next MPS but also for the WG11 further activities. It was decided to organize a special WG11 meeting in Greece during the next months (winter 2018 or spring 2019) in order to discuss the future of WG11 (with or without the MPN) and also to put educational, research and management priorities.

In the meanwhile,
a WG11 was arranged within the EPC in Dubrovnik, 17 September 2018.

3. New Member

At the end of 2016, following the official procedure, Dr Emily Burton from UK Branch, has been nominated as new member of WG11 (find attached the letter sent to Dr Burton).

4. Official Communications with WG11 Members

The last official communication with the Members of WG11 was at the end of year 2017 (find attached). Official communications between Chair and Members of WG11 occur at a maximum of a 6 month period.

Working Group 12 - Physiology (2017-2018)

Main topic of WG 12 is the physiology of developing embryo, its programming by parental and environmental incubation factors and the long-lasting effects on performance, health and welfare. In 2017, from 30th August to 1st September, the WPSA Working Group 12 (Physiology) organized together with the Working Group 6 (Incubation and Fertility) a meeting on "Perinatal Development and Physiology in Poultry" (PDP) in conjunction with the meeting of the "Incubation and Fertility Research Group" (IFRG; www.ifrg.org) in Wageningen, the Netherlands. It was the fourth combined scientific meeting of both WPSA working groups 6 and 12. Altogether, 56 delegates from Australia, North America, Africa and Eastern and Western Europe were present during two and a half conference days. The delegates, speakers, and poster presenters came from different scientific disciplines and companies. The latest developments in basic and applied poultry physiology, focused on poultry incubation and long-lasting parental and environmental effects on poultry welfare and performance, as well as on incubation practice and technology, were presented under the following topics:

- Influence of breeder on chick health and performance
- Egg storage and handling
- Impact of parental effects
- Adaptation and epigenetic alteration in birds
- Incubation parameters and chick vitality
- Control and programming of energy balance

Keynote speakers were Mylene Mariette, postdoctoral fellow at the Centre of Integrative Ecology, Deakin University, Victoria, Australia (avian adaptation to heat by prenatal acoustic stimulation), Bence Lácár, Phd student from the Doctoral School of Animal Husbandry Science, Szent István University, Gödöllö, Hungary (combined effect of parental thermal stress and thermal treatment of the offsprings on early embryonic development and on primordial germ cells), and Prof. Dr. A.G.G. (Ton) Groothuis from the Department of Behavioural Biology, Centre for Behaviour and Neuroscience, University of Groningen, the Netherlands (avian yolk hormones and the state of the art of understanding the mechanisms and their functions). The impressive talks of the keynote speakers as well as the 29 oral and poster presentations formed the excellent scientific workshop fundament.

Further details are available in the workshop proceedings, which were published online in the European Poultry Science (EPS) journal. Preface and abstract collection of oral and poster presentations related to the respective session can be downloaded under the following link: http://www.ulmer.de/preview/preview.dll/EPS?AID=5595966
In 2019, the next combined meeting of the Working Group 12 and the Working Group 6 will be organized in Tours (France). First information will be presented at the group meeting during the EPC2018 in Dubrovnik.

PD Dr. Barbara Tzschentke, Berlin, June 29, 2018
Humboldt-Universität zu Berlin
Faculty of Life Science, Institute of Biology
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Chair WG Physiology, European Branches WPSA

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General information

Venue

Dubrovnik, Croatia

Dubrovnik is situated at the most southern part of Croatia, on the Adriatic coast. Unique for its impressive medieval forts, churches, monuments and palaces, Dubrovnik is often called the pearl of the Adriatic. More than a thousand-year old history of Dubrovnik made it a cultural centre of Europe. History is present in the entire city, which is both a museum and a picturesque stage where cultural heritage and contemporary life meet. All houses and monuments have a unique value. The Old City is encompassed by medieval walls, which have been preserved in their original form and opened for visitors as Dubrovnik's major attraction. In 1979 the City was included in UNESCO World Heritage Site list.

Conference hall

The XVth European Poultry Conference will take place in Valamar Lacroma hotel.
https://www.valamar.com/hr/hoteli-dubrovnik/valamar-lacroma-dubrovnik-hotel
Valamar hotels and resorts in Dubrovnik are located on a narrow peninsula of Dubrovnik called Lapad, on its narrow west side known, and usually called the peninsula of Babin kuk. Hotel Valamar Lacroma Dubrovnik is the largest conference venue in Dubrovnik. Its main conference hall has capacity of 1300 participants, with additional 7 smaller conference halls capacity from 12 to 60 participants. With up mentioned, Valamar Lacroma offers as well 1000m2 space for exhibitors and poster sessions.

Registration desk working hours

Lacroma Hotel

16th September 2018 / 16-19h
17th September 2018 / 10-20h
18th September 2018 / 8-18h
19th September 2018 / 8-11h

Social Events

Welcome reception

17th September 8PM-10PM / Lacroma hotel terrace
Valamar Lacroma hotel (conference venue) beautiful terrace will be the location of the Welcome cocktail on Monday, 17th September 2018. Throughout the terrace, there are breath-taking views across the sea and Elaphite Islands.
For this evening we prepared a special musical program as a gift to the participants! All delegates are cordially invited to join the Welcome ceremony.

Sponsored by: MSD Animal Health

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Gala dinner

19th September 8PM - 10:30PM / the fortress Revelin/ Dubrovnik old city centre
Gala dinner will take place in one of the most interesting Dubrovnik city centre sites – fortress Revelin. Enjoy nice and tasty dishes accompanied with great music and have some fun with your colleagues and friends.
Sponsored by: ew nutrition

Technical visit

21st September 8AM – cca 4 or 5PM / the Neretva valley and city Metković
Technical visit is planned as full day tour incl. transfer and lunch. Visiting poultry farm near the Metković city, you will be able to get a "wow effect" by getting the view to the wonderful Neretva Valley.

Youth Programme

The goal of this programme is to give the students possibility to attend the XVth EPC 2018 in Dubrovnik, Croatia and to present their paper. The students will be awarded with full registration to EPC2018 and four-day post-conference journey, visiting poultry facilities in Croatia, as well as cultural and natural sites. Applicants are students (undergraduate, graduate, master or PhD student). Maximum age of the applicant is 35 at the time of application. Applicants should submit an abstract for an oral or poster presentations and be the first author.
Sponsored by: Dechra Veterinary Products

Internet connection

Wi-Fi free access for all conference participants will be available during the whole conference in congress centre. Many Dubrovnik hotels as well offer free Wi-Fi in their hotels.

Language

Official language at conference is English with no possibility of simultaneous interpreting.

PROGRAMME

SUNDAY, 16th September 2018

16:00-19:00	**REGISTRATION**

MONDAY, 17th September 2018

10:00-20:00	**REGISTRATION**
13:00-14:30	**FRAmelco Symposium: Viral challenges in the poultry industry** Dr. Maarten de Gussem (Vetworks) & Ir. Olga Dansen (FRAmelco)
15:00-16:30	**MSD Animal Health Symposium: Innovax ND-IBD and Exzolt Interactive Symposium**
17:00-18:30	**EW Nutrition Symposium: AMR-Lessons learned and future perspectives** Guest Speakers: Dr. Adela Maghear (HCWH) and Dr. Suwit Chotinan (Faculty of Veterinary Medicine, Chiang Mai University)
17:00-19:00	**Executive committee meeting**
20:00-22:00	**Welcome Cocktail**

TUESDAY, 18th September 2018

08:00-18:00	**REGISTRATION**
08:30-09:00	**Opening Ceremony - Hall: Elafiti**
09:00-12:30	**Plenary Sessions -** Hall: Elafiti
09:00-10:30	PLENARY SESSION 1 / Chair: S. Weigend
09:00-09:40	Are there limits to selection in poultry: theoretical, biological, ethical, enviromental? - M. T. Boichard / France
	Discussion
09:45-10:25	How Scientific Innovation may improve egg production and quality or the development of non-food uses of eggs - Y. Nys / France
	Discussion
10:30 - 11:00	**COFFEE BREAK AND VIEWING POSTERS**
11:00-12:30	PLENARY SESSION 2 / Chair: H. Medić
11:00 - 11:40	Preincubation and incubation conditions, hatching time and broiler growth - S. Yalcin / Turkey
	Discussion
11:45 - 12:25	New insights on breast myopathies - S. Barbut / Canada
	Discussion

12:30 - 13:30	**LUNCH BREAK**	
13:30-15:30	**Parallel Oral Sessions**	
Session: Nutrition 1 / Minerals in Poultry Nutrition - Hall: Elafiti 1 Chairs: Z. Uni, Z. Kralik		
Time	*Abstract ID*	*10-minutes oral presentations*
13:30-13:43	265	Effects of dietary supplementation of different sources of selenium and different levels of selenium yeast on laying hens - G. Qi, Q. Sun, H. Yue, S. Wu, J. Wang, H. Zhang/China
13:43-13:56	297	Hydroxy-selenomethionine improves feed conversion of heat stressed finisher broilers associated with enhanced Se bioavailability and antioxidant response - J. Michiels, M. Briens, T. Guillou, M. Majdeddin, J. Pincemail/Belgium
13:56-14:09	638	Influence of use of organic minerals in laying hen diets on productive performance, egg quality and bone strength - Z. Janjecic, D. Bedekovic, I. Matanic, M. Madjeruh, I. Kovacev / Croatia
14:09-14:22	312	Micro-Mineral composition and deposition efficiency in the broiler hatching egg - D. Eytan, Z. Uni / Israel
14:22-14:35	351	Effects of two limestone sources differing in particle size in broiler diets with or without phytase - C. Kwakernaak, R. Davin, Y. Djersant-Li / United Kingdom
14:35-14:48	333	Comparison between inorganic and hydroxychloride zinc on performance and carcass characteristics in broiler chickens: a meta-analysis - S. van Kuijk, M. Jacobs, C. Smits, Y. Han / NETHERLANDS
14:48-15:01	555	Calcium level in pre-starter diets of broilers: too high? - C. Araujo Torres, A. Bonilla, A. Dijkslag, A. Isabel Garcia / Spain
15:01-15:14	444	Influence of calcium solubility on egg production and egg quality in 68-week-old laying hens – L. van Eck, D. Lamot, H. Enting, S. Powell / Netherlands
15:14-15:27	401	Phytogenic premix effects on gene expression of intestinal antioxidant enzymes and broiler meat antioxidant capacity - K. Mountzouris, V. Paraskeuas, E. Griela, G. Papadomichelakis, K. Fegeros / Greece
Session: Poultry Health 1 / Vaccines - Hall: Elafiti 2 Chairs: H. Mohamed Hafez; V. Savić		
Time	*Abstract ID*	*Keynote Lecture*
13:30-13:50	502	New insights on application of IBV vaccines - J. (Sjaak) de Wit / Netherlands
		Discussion
Time	*Abstract ID*	*8-minutes oral presentations*
13:55-14:04	300	Gel vs spray administration for infectious bronchitis virus: which is better? - B. Jordan, A. Reith, L. Newman/USA
14:04-14:13	83	Comparison two type of inactived avian infectious bronchitis vaccines (M41 and M41/Dutch variants) in protection against Variant-2 A. Ghalyanchilangeroudi, V. Karimi, R. KH Farahani, H. Hosseini, T. Zabihi, M. Hossein Fallah/Iran
14:13-14:22	260	Adenoviral gizzard erosion in pullet and layer flocks and potential means of prophylaxis due to live vaccination with an apathogenic FAdV-A (CELO) - B. Grafl, D. Liebhart, A. Schachner, M. Hess/Austria

14:22-14:31	355	Effect of age of vaccination on antibody titers, clinical signs occurrence and performances after challenge with H9N2 on broilers Y. Bensassi/Morocco
14:31-14:40	52	DNA construct vaccine applied in hatchery as a tool to combat ILT virus circulating in the field D. Furmanek, P. Stachów/Poland
14:40-14:49	85	Assessment of innate immune response by applying mono or polyvalent live Newcastle disease and Infectious bronchitis vaccinations A. Ghalyanchilangeroudi, P. Hesari, R. KH Farahani, S. Ali Ghafouri, H. Hosseini, N. Sadri, A. Homayounmehr, R. Ehsan, H. Abdollahi, V. Karimi, M. Hossein Fallah, M. Jabbarifakhr/Iran
14:49-14:58	534	Investigation of the effect of live attenuated *Escherichia coli* vaccination on experimentally induced salpingitis in layers - I. Thøfner, J. Peter Christensen/Denmark
14:58-15:07	627	Autogenous vaccine against *Escherichia coli* and *Gallibacterium anatis* reduces losses and improves production on layer farms - Ž. Gottstein, L. Lozica, D. Horvatek Tomić, G. Nedeljković, M. Lukač, E. Prukner-Radovčić /Croatia
15:07-15:16	276	Comparison of Two European Gel-Type Diluents vs. Water - L. Newman/USA
15:16-15:25	356	Efficacy of different vaccination programs against colibacillosis after intratracheal challenge at the beginning of laying period in commercial layers - D. Koutsianos, H. Gantelet, L. V.Athanasiou, D. Mossialos, E. Thibault, K. Koutoulis/Greece
15:25-15:34	49	Comparision of commercial broilers flocks vaccinated with a novel DNA construct vaccine applied in hatchery and reared in different field conditions - D. Furmanek, M. Mamczur/ Poland

Session: Poultry Welfare 1 - Laying Hens- Hal: Elafiti 3
Chairs: V. Ferante, T. Norton

Time	*Abstract ID*	*8-minutes oral presentations*
13:30-13:40	484	Influence of different environmental enrichment programmes on behaviour and utilisation of floor space of pullets and laying hens kept on commercial farms - S. Freytag, B. Spindler, N. Kemper/Germany
13:40-13:50	422	Assessing the Welfare of end of lay hens during the catching and packing process of depopulation - C. Gerpe, M. Toscano, A. Stratmann/ Switzerland
13:50-14:00	474	Feeding black soldier fly larvae to laying hens: effects on production performance - M. A.W. Ruis, L. Star, J.L.T. Heerkens, H. J. Kuipers, F. Kromhout, E. Beitler, J. Katoele/ The Netherlands
14:00-14:10	342	Effect of keel bone fractures on laying hen behaviour in a non-cage housing system - A. K. Rentsch, C. B. Rufener, C. Spadavecchia, A. Stratmann, M. J. Toscano/Switzerland
14:10-14:20	217	Welfare assessment of laying hens in four housing systems in Slovenia - O. Zorman Rojs, A. Dovč, M. Červek, M. Zupan/Slovenia
14:20-14:30	240	Being fearful or calm individual predisposes different development of keel bone in laying hens - N. Rokavec, I. Dimitrov, M. Zupan/Slovenia
14:30-14:40	466	Comparing stress levels and behavior in two lines of layer hens transported with and without break - H. Sprafke, R. Palme, J. Reinhard, M. Erhard, S. Bergmann/Germany
14:40-14:50	200	The use of pheromones in laying hens in aviary housing system - N. Sleeckx, I. Kempen, K. De Baere, S. Cardinaels, J. Zoons/Belgium

14:50-15:00	593	Effects of hybrid and ramps status on health parameters and behaviour of laying hens: An on-farm study - N. Mackie, J. Tarlton, S. Buijs, S. De Knibber, B. Ampe, F. Tuyttens/ United Kingdom
15:00-15:10	283	Effects of a treatment against *Dermanyssus gallinae* with fluralaner on welfare parameters in laying hens - D. Temple, D. Escribano, M. Salas, E. Mainau, X. Manteca, I. Petersen, E. Thomas, R. Dolz, C. Escoda, A. Flochlay-Sigognaul/Spain
15:10-15:20	156	Implementation of project results for the rearing of pullets under practical conditions - H. Louton, A. Schwarzer, M. Erhard/Germany
15:20-15:30	198	Economic supporting model to determine optimal age of layers flock Replacement - H. Arazi, Y. Malka, A. Regev/Israel

Session: Antibiotic resistance - Hall: Elafiti 4
Chairs: I. Rychlik, Ž. Gottstein

Time	*Abstract ID*	*Keynote Lecture*
13:30-13:50	316	Retrospective study on antimicrobial resistance profiles from *E. coli* isolated from diagnostic samples - **C. Hess**, B. Grafl, D. Jandreski-Cvetkovics, A. Georgi, M.Hess/Austria
Time	*Abstract ID*	*9-minutes oral presentations*
13:50-14:01	41	Microfloral rehabilitation: Normalisation of Gut Function - R. Murphy /Ireland
14:01-14:12	558	The role of the poultry veterinarian in reduction of antibiotic use, intervention study in broiler farms in different EU countries - H. Van Meirhaeghe, M. De Gussem, P. Sanders, C. Chauvin, J. David, N. Ongena/ Belgium
14:12-14:23	148	Ecology from farm to fork of microbial drug resistance and transmission" Interventions to reduce antibiotic use - N. Ongena, M. De Gussem, H. Van Meirhaeghe, P. Sanders, M. Van Leuven/Belgium
14:23-14:34	345	Good management and poultry welfare - perfect combination for a responsible use of antimicrobials - I. Ajuda, E. Bianco, B. Bertolina, A. Costa/United Kingdom
14:34-14:45	583	Effective replacement of in-feed antibiotics with a blend of free and buffered organic acids on performance and gut health in broilers P. Roubos-van den Hil, N. de Groot, Y. Wu, Y. Ding, K. Zhang/ The Netherlands
14:45-14:56	512	Effect of disinfectant use on antibiotic susceptibility of *E. coli* isolates from broiler houses - H. Maertens, E. Van Coillie, E. Meyer, J. Dewulf, K. De Reu/ Belgium
14:56-15:07	493	Practical evidence-based approaches towards reduction of antimicrobial use and antimicrobial resistance - T. van Gerwe, A. Awati, M. Cabellero/ Germany
15:07-15:18	140	Should enzymes be part of your antibiotic reduction strategy? - A. Awati, T. van Gerwe, M. Caballero/Germany
15:18-15:29	519	Dietary nucleosides improved performance and gut health lately in broilers - M. Gopi, J. Rokade Jaywant, T. S Shyamkumar, G. Kolluri, G. Khillare, J. S Tyagi/India
15:30 - 16:00		**COFFEE BREAK AND VIEWING POSTERS**

16:00-18:00 Parallel Oral Sessions

Session: Nutrition 2 / Perspectives on Feeding strategies - Hall: Elafiti 1
Chairs: W. Siegert, Z. Janječić

Time	*Abstract ID*	*Keynote lecture*
16:00-16:20	193	In-ovo fed nutrients accelerate intestinal brush border maturation towards hatch - N. Reicher, J. Dayan, **Z. Uni**/Israel
		Discussion
Time	*Abstract ID*	*10-minutes oral presentations*
16:25-16:37	471	Digestibility of conventional and organic feedstuffs in laying hens L. Star, M. M. van Krimpen, A. J.W. Mens/The Netherlands
16:37-16:49	414	Effect of split feeding on performance and eggshell quality in laying hens E. Delezie, A. Molnar, I. Kempen, N. Sleeckx, L. Maertens, J. Zoons, J. Buyse/Belgium
16:49-17:01	582	Identification of candidate genes for calcium absorption along the small intestine of the laying hen - A. Gloux, N. Leroy, A. Brionne, E. Bonin, A. Juanchich, G. Benzoni, Y. Nys, J. Gautron, A. Narcy, M. J Duclos/France
17:01-17:13	86	Digestibility coefficients of pea, faba bean and lupin seeds following application of protease in broiler chickens - S. Kaczmarek, M. Kubiś, S. Peris, S. Budnik, M. Hejdysz, A. Rutkowski/Poland
17:13-17:25	309	Effects of an organic acid combination with polyphenols via drinking water supplementation on broiler growth performance and ascites incidence under heat stress conditions - A. Sozcu, A. Ipek, K. C. Kjeldsen/ Denmark
17:25-17:37	237	Relationship between variations in particle size distribution of mash diets and the occurrence of performance and health issues in German hen flocks - M. Lieboldt, L. Borgelt, P. Wolf/Germany
17:37-17:49	229	The effect of feed restriction on performance, organ development and blood picture of broiler chickens - V. Machander, E. Tůmová, D. Chodová/Czech Republic
17:49-18:00	203	Glycine plus serine requirements of broilers fed low-protein diets J. van Harn, M. Dijkslag, M. van Krimpen/The Netherlands

Session: Poultry Health 2 / Microorganisms - Elafiti 1
Chairs: A. Franchini, E. Prukner-Radovčić

Time	*Abstract ID*	*8-minutes oral presentations*
16:00-16:10	535	Estimates on the significance of chronic *E.coli* infections of the reproductive tract in laying hens - I. Thøfner, J. Peter Christensen/ Denmark
16:10-16:20	305	Expressional analysis of few immune relavant genes and bio-molecule at nasal mucosal surface against bacterial pathogen *Avibacterium paragallinarum* - D. Aagza, S. Deshmukh, D. Narang, D. Deka, H. Singh Banga/India
16:20-16:30	623	Prevalence of *Gallibacterium anatis* isolated from layer poultry farms in Croatia - L. Lozica, D. Horvatek Tomić, E. Prukner-Radovčić, Ž. Gottstein/Croatia
16:30-16:40	244	Effect of selected yeast fraction on the growth of *Clostridium perfringens*: quantitative determination of growth inhibition and adsorption capacity E. Santovito, D. Greco, V. Marquis, R. Raspoet, V. D'Ascanio, G. Avantaggiato / Italy
16:40-16:50	304	Different strains of *Clostridium perfringens* cause different levels of severity of necrotic enteritis in broiler chickens - K. Gharib Naseri, R. Swick, M. Choct, N. Morgan, C. Keerqin, S. Wu/Australia
16:50-17:00	165	Efficacy of specific compositions of 1-monoglycerides of short- and medium chain fatty acids in controlling *Salmonella* Typhimurium and other serotypes of *Salmonella* spp. in broiler chickens and in vitro conditions- M. Parini, A. Paoli, A. Buccioni, M. Antongiovanni, P. Massi, G. Tosi, L. Fiorentini/Italy
17:00-17:10	443	Delivery of oligodeoxynucleotides containing CpG motifs (CpG-ODN) by the intrapulmonary route against bacterial septicemia in neonatal broiler chickens - S. Gomis, K.B. Goonewardene, S. Popovich/Canada
17:10-17:20	353	Amino acid substitutions in the Receptor Binding Domain (RBD) of avian influenza viruses subtype H9N2 in Israeli isolates, with human health implications - A. Lublin, I. Shkoda, A. Biton/Israel
17:20-17:30	328	Avian botulism: recent investigations from diagnosis to management (in France) - C. Le Maréchal, R. Souillard, S. Rouxel, T. Poezevara, E. Houard, P. Fach, S. Le Bouquin, M. Chemaly/France
17:30-17:40	284	Effects of feeding Diamond V fermentation metabolites on avian pathogenic *E. coli* prevalence and antibiotic resistance of *E. coli* in ceca samples taken from commercial broilers and turkeys - W. Abdelrahman, J. A. Byrd, D. R. McIntyre, H. O. Pavlidis, J. P. Carroll, S. Carlson/USA
17:40-17:50	151	Development and application of a quantitative real-time PCR for the detection of *Enterococcus cecorum* - A. Jung, H. Petersen, L. Mohr, S. Rautenschlein/Germany
17:50-18:00	84	Comparative study on primers for molecular detection of different infectious bronchitis virus genotypes - A. Ghalyanchilangeroudi, R. Kh Farahani, N. Sedighi, H. Hosseini, M. Jabbarifakhr, F. Mousavi, H. Maghsousdloo/Iran

Session: Breeding and Genetics - Hall: Elafiti 3
Chairs: S. Weigend, M.T. Boichard

Time	*Abstract ID*	*10-minutes oral presentations*
16:00-16:10	548	The epigenetic effects of feeding increased methyl donors to Japanese quail - C. Phillips, R. Angel, C. Ashwell/USA
16:10-16:20	437	Broiler body composition is transgenerationally affected by reduced dietary protein levels in breeder hens - S. Schallier, C. Li, J. Lesuisse, N. Everaert, J. Buyse/Belgium
16:20-16:30	294	Visceral fat of broiler and layer chickens and its apparent endocrine role in female reproduction - M. Friedman-Einat, S. Yosefi, D. Shinder, M. Rozal, E. Seroussi/Israel
16:30-16:40	372	Exploiting extreme phenotypes to investigate haplotype structure and detect signatures of selection for body weight in broilers - E. Tarsani, A. Kominakis, G. Theodorou, I. Palamidi/Greece
16:40-16:50	494	Effect of long-term heat stress on production, egg quality and physiological traits in four experimental lines of layers differing in heat tolerance and feed efficiency - A. Tholance, D. Nyuiadzi, V. M. Darras, D. Laloë, F. Jaffrezic, S. Lagarrigue, A. Rau, A. Colli, T. Zerjal/France
16:50-17:00	625	Genome-wide association study for eggshell crystal structures in chicken - C. Sun, Z. Duan, G. Xu, N. Yang/China
17:00-17:10	509	The use of axial-transmission ultrasound to determine bone quality in the laying hen - H. McCormack, E. Sanchez-Rodriguez, C. Benavides-Reyes, A. B. Rodríguez-Navarro, B. Andersson, W. Icken, N. Sparks, D. de Koning, I. C. Dunn/United Kingdom

17:10-17:20	526	Genetic relationship between the keel bone palpation score and performance traits in white layers - B. Andersson, W. Icken, F. Kaufmann/Germany
17:20-17:30	644	Exploiting linkage disequilibrium in GWAS analysis - F. Ramzan, M. Gültas, H. Simianer, D. Cavero, A. O. Schmitt/Germany
17:30-17:40	551	Gene-based mapping and pathway analysis of feather pecking in laying hens - J. Tetens, J. Beier, C. Falker-Gieske, S. Preuß, W. Bessei, J. Bennewitz/Germany
17:40-17:50	207	Different evolutionary dynamics revealed by functional SNP classes in global chicken groups - D. Kholofelo Malomane, H. Simianer, C. Reimer, A. Weigend, S. Weigend/Germany
17:50-18:00	417	Gene pool preservation in poultry- actuality and solutions S. Cherepanov, O. Stanishevskaya/Russian Federation
18:00-18:10	364	Male germ line transplantation as an efficient technique of transgenesis in chicken - P. Trefil, J. Mucksova, A. Koslova, J. Kalina, B. Benesova, J. Hejnar/Czech Republic
18:10-18:20	609	Evaluation of growth and egg production performance of Iranian improved indigenous hens in rural areas - A. Gheisari, J. Amini Jebel Kandi, a. Hesabi Nameghi, H. Norolahi, A. Haghnazar Kocheksaraei/Iran
18:20-18:30		*Discussion*

Session: Poultry Meat Quality and Safety - Hall: Elafiti 4
Chairs: M. Grashorn, S. Barbut

Time	*Abstract ID*	*Keynote lecture*
16:00-16:15	507	European union approaches toward safe poultry meat Hafez Mohamed Hafez/Germany
Time	*Abstract ID*	*8-minutes oral presentations*
16:20-16:29	220	Evolution of macroscopic and microscopic broiler breast alteration related to white striping - E. Russo, V. Felice, M. Drigo, C. Lupini, C. Longoni, E. Catelli, A. Dalle Zotte, M. Cecchinato/Italy
16:29-16:38	628	White striping prevalence and its effect on meat quality of broiler chicken breast fillets under commercial conditions S. Golzar Adabi, E. Demirok Soncu, N. Ceylan, N. Kolsarıcı/Turkey
16:38-16:47	408	Comparison of protein profile by SDS-PAGE of broiler breast meat affected by white striping, wooden breast and spaghetti meat myopathies F. Soglia, G. Baldi, E. Babini, C. Cavani, M. Petracci/Italy
16:47-16:56	264	Rearing factors affecting myopathy rate and meat quality in broiler chickens - A. Trocino, A. Pascual, F. Gratta, M. Birolo, C. Zomeño, G. Xiccato/ITaly
16:56-17:05	607	Characterization of productive traits and incidence of breast myopathies in a broiler chicken crossbred proposed for alternative production systems - M. Zampiga, M. Petracci, A. Meluzzi, F. Sirri/Italy
17:05-17:14	352	Raw and cooked meat texture as affected by white striping, wooden breast and spaghetti meat myopathies - G. Baldi, F. Soglia, C. Cavani, M. Petracci/Italy
17:14-17:23	277	Effect of myopathy occurrence on quality of broiler chicken breasts during storage - F. Gratta, L. Fasolato, M. Birolo, E. Novelli, M. Petracci, C. Zomeño, A. Piccirillo, A. Pascual Guzmán, G. Xiccato, A. Trocino/Italy
17:23-17:32	238	Occurrence and characterization of biofilms in drinking water systems of broiler houses - S. Maes, T. Vackier, M. Heyndrickx, H. Steenackers, I. Sampers, K. Raes, A. Verplaetse, K. De Reu/Belgium
17:32-17:41	337	Is meat of turkey breeders really different from that of standard turkeys? - P. Chartrin, T. Bordeau, E. Godet, K. Meteau, J. Gicquel, E. Drosnet, S. Briere, M. Bourin, E. Baeza/France
17:41-17:49	529	Volatile fingerprints of raw and cooked meat of different commercial chicken genotypes A. Cartoni Mancinelli, E. Siletti, S. Mattioli, A. Dal Bosco, B. Sebastiani, L. Menchetti, A. Koot, S. Van Ruth, C. Castellini/Italy

09:00-18:00	**Poster Sessions Meeting Place - Business Center**
Antibiotic resistance	
Breeding and Genetics	
Poultry Welfare	
Nutrition 1	

WEDNESDAY, 19th September 2018

08:00-11:00	**REGISTRATION**
09:00-12:30	**Plenary Sessions - Hall: Elafiti**
09:00-10:30	PLENARY SESSION 3 / Chair: B. Svihus
09:00-09:40	Alternative protein sources for poultry nutrition - Steenfeldt S. / Denmark
	Discussion
09:45-10:25	Non-essential amino acids – the forgotten nutrients? - Siegert W. / Germany
	Discussion
10:30-11:00	**COFFEE BREAK**
11:00-12:30	PLENARY SESSION 4 / Chair: D. Horvatek Tomić
11:00-11:40	Application of Precision Livestock Farming technologies in the poultry sector - T. Norton / Germany
	Discussion
11:45-12:25	Managing on-farm health and welfare risks to promote sustainability in poultry industry - Sossidou N. Evangelia / Greece
	Discussion
12:30-13:30	**LUNCH BREAK**

13:40-15:40 Parallel Oral Sessions

Session: Nutrition 3 / Additives in Poultry Nutrition - Hall: Elafiti 1
Chairs: S. Steenfeld, D. Bedeković

Time	Abstract ID	*10-minutes oral presentations*
13:40-13:51	520	Nanoparticles as feed additives in poultry nutrition - A. Hable, R. Hude, S. C. Jagdale/India
13:51-14:02	492	Performance and intestinal microflora of broilers fed the probiotic *Bacillus amyloliquefaciens* H57 -W. Bryden, Yadav.Bajagai, D. Zhang, X. Li, P. Dart, A. Klieve, P. Hugenholz/Australia
14:02-14:13	211	A probiotic containing viable spores of *Bacillus licheniformis* reduces caecal colonization of Campylobacter - V. Hautekiet, A. Kanora/Belgium
14:13-14:24	210	A probiotic containing viable spores of *Bacillus licheniformis* prevents outbreak of necrotic enteritis - V. Hautekiet, A. Kanora/Belgium
14:24-14:35	459	Effect of pH-manipulation of a wheat-based diet on phytase-induced phytate degradation in an in vitro simulation of crop retention - S. Kristoffersen, N. Tao, B. Svihus/Norway
14:35-14:46	138	Phytate disappearance and myo-inositol release in gnotobiotic broiler chickens - V. Sommerfeld, A. Van Kessel, H. L. Classen, M. Schollenberger, I. Kühn, M. Rodehutscord/Germany
14:46-15:57	81	Dried yeast and its enzymatically hydrolyzed derivatives can improve performance and organ development of broiler chickens challenged with Salmonella lipopolysaccharide - E. U. Ahiwe, M. Abdallh, E. Chang'a, M. Al-Qahtani, A. Omede, H. Graham, P. Iji/Nigeria, Australia
15:57-15:08	392	Efficacy of a novel silica supplement fed to broilers - S. E. Prentice, D. J. Belton, D. V. Scholey, C. C. Perry, E. J. Burton/United Kingdom
15:08-15:19	178	Barley in broiler diets: Optimum inclusion and response to carbohydrase enzyme - R. Abdollahi, K. Perera, R. Ravindarn, F. Zaefarian/New Zealand
15:19-15:30	48	Effect of licorice essential oils on meat quality and cecal microbial population of broiler chickens - A. Yaghobfar, A. Hossein Alizadeh-Ghamsari, S. Abdoullah Hosseini, M. Garavand, S. Davood Sharifi/Iran
15:30-15:41	642	Poultry meat quality & functionally food - F. Rutz, E. Xavier, D. Lopes, V. Roll, J. Nunes, A. Roll/Brazil

Session: Poultry Health 3 / Parasites - Hall: Elafiti 2
Chairs: O. Sparagano, D. Horvatek Tomić

Time	Abstract ID	*8-minutes oral presentations*
13:40-13:49	269	Comprehensive epidemiological investigations support the role of *Histomonas meleagridis* for systemic translocation of *Escherichia coli* in chickens - S. Paudel, B. Stessl, C. Fürst, D. Jandreski-Cvetkovic, C. Hess, M. Hess/Austria
13:49-13:58	318	Influence of alternative husbandry systems on post-mortem findings and prevalence of important bacteria and parasites in layers - determined from end of rearing until slaughter - A. Zloch, S. Kuchling, M. Hess, C. Hess/Austria
13:58-14:07	282	Treatments of gastrointestinal problems in broiler breeders and pullets after coccidiosis vaccination – a retrospective study - M. Vereecken, K. De Gussem, M. De Gussem, P. Mitsch/Belgium
14:07-14:16	272	Monitoring after coccidiosis vaccination- sample collection and dynamics in different housing systems - M. Vereecken, P. Mitsch, K. De Gussem/Belgium
14:16-14:25	212	A probiotic containing viable spores of *Bacillus licheniformis* improves gut health after coccidiosis vaccination - V. Hautekiet, M. Vereecken/Belgium
14:25-14:34	379	Overview on the efficacy of paromomycine as a treatment for commercial turkeys experiencing outbreaks with *Histomonas meleagridis* - M. Vereecken, D. Wunderl, K. De Gussem, W. Depondt, B. Mägdefrau-Pollan/Austria
14:34-14:43	261	Hens in mixed meat turkey flocks separated by wire fence from toms showing high mortality due to histomonosis are infected based upon laboratory diagnostics - B. Grafl, T. Sulejmanovic, B. Jaskulska, I. Bilic, D. Liebhart, M. Hess/Austria
14:43-14:52	510	Effects of diet dilution on bone development and mineralization of coccidiosis-infected broilers - I. Oikeh, P. Sakkas, J. Taylor, D. Blake, I. Kyriazakis/United Kingdom
14:52-15:01	508	Phytogenics improve resilience against coccidiosis and secondary intestinal infections in broilers - J. Dirk van der Klis, L. Jungbauer, A. Mueller/Austria
15:01-15:10	298	Coccidiosis control with farm management to prepare for "No antibiotics ever" production - L. Newman, A. Montoya/USA
15:10-15:19	251	Efficacy of a monensin/nicarbazin combination in the control of coccidiosis in turkeys under floor pen conditions using a seeder model M. Marien, B. Dehaeck, M. Vereecken, M. Geerinckx, K. De Gussem/Belgium
15:19-15:28	246	Comparison of different anticoccidial programs on coccidiosis control of broilers under floorpen conditions: shuttle versus full - M. Marien, B. Dehaeck, M. Vereecken, M. Geerinckx, K. De Gussem/Belgium
15:28-15:37	28	Efficacy of Gallifen® 40 mg/ g premix against natural infections of *Ascaridia galli* and *Heterakis gallinarum* in layer chickens W. Depondt, A. Kanora, M. Vereecken, L. Claerhout/Belgium

Session: Poultry Welfare 2 / Broilers – Hall: Elafiti 3
Chairs: W. Bessei, V. Michel

Time	Abstract ID	*10-minutes oral presentations*
13:40-13:50	33	Evaluating environmental enrichment for broiler chickens using the transect method - N. Ben Sassi, J. Vas, G. Vasdal, X. Averos, I. Estevez, R. Newberry/Spain
13:50-14:00	45	To catch or not to catch? That is not the only question in an educational program for catchers - K. Kittelsen, E. Georg Granquist, R. Oppermann Moe, E. Tolo/Norway
14:00-14:10	102	Identification of risk factors and prevalence of injuries at different stages of the broiler slaughter process - I. de Jong, H. Reimert, T. Lohman, M. Gerritzen/The Netherlands
14:10-14:20	577	Development of a user-friendly protocol and webtool for monitoring and benchmarking broiler chicken welfare during the pre-slaughter phase - F. Tuyttens, L. Jacobs, B. Ampe, L. Duchateau, E. Delezie/Belgium
14:20-14:30	130	Environmental enrichment increases positive activity in broiler chickens - G. Vasdal, J. Vas, R. Newberry, R. Moe/Norway

14:30-14:40	194	The effect of different eggshell temperature patterns during incubation on broiler chicken behavior determined by an automatic tracking system - R. Molenaar, E. de Haas, T. Rodenburg, L. Olde Bolhaar, H. Wijnen, H. van den Brand/The Netherlands
14:40-14:50	209	A probiotic containing viable spores of *Bacillus licheniformis* reduces lameness in broilers - V. Hautekiet, A. Kanora/Belgium
14:50-15:00	340	'European Broiler Ask' – The new era for poultry welfare - I. Ajuda/United Kingdom
15:00-15:10	404	Production performance of two dual-purpose chicken breeds in a mobile stable system - F. Kaufmann, U. Nehrenhaus, R. Andersson/Germany
15:10-15:20	409	Value creation by a novel ceiling-based mobile broiler system ("ChickenBoy") for improving animal health, wefare and productivity J. Hartung, H. Lehr, D. Rosés, M. Mergeay, J. van den Bossche/Germany
15:20-15:30	528	Effect of feed color on growth performance and tonic immobility in broiler chicks - M. Toghyani, M. Ali Mesmarian/Iran

Session: Poultry Housing and Management – Hall: Elafiti 4
Chairs: R. Akbay, R. Mulder

Time	*Abstract ID*	*10-minutes oral presentations*
13:40-13:52	530	Innovative agroforestry models: chickens in olive orchards and geese in vineyards - S. Mattioli, A. Cartoni Mancinelli, E. Cotozzolo, A. Rosati, A. dal Bosco, M. Guarino Amato, C. Castellini/Italy
13:52-14:04	69	Microbial-mineral manure additive reducing odours - S. Opalinski, M. Korczynski, K. Kalus, R. Kolacz, Z. Dobrzanski, B. Gutarowska/Poland
14:04-14:16	195	Improving external egg quality in enriched cages - I. Kempen, N. Sleeckx, S. Cardinaels, K. De Baere, J. Zoons/Belgium
14:16-14:28	201	On farm hatching in broilers – Effect on production, welfare and profitability - I. Kempen, K. De Baere, S. Cardinaels, N. Sleeckx, J. Zoons/Belgium
14:28-14:40	228	Influence of the housing system on mating behavior in two broiler breeder hybrids - S. G. Gebhardt-Henrich, A. Jordan, M. J. Toscano, H. Wuerbel/Switzerland
14:40-14:52	313	Providing ramps during rearing improves bone strength in laying hen pullets - A. Stratmann, D. Guggisberg, J. Siegford, M. Toscano/Switzerland
14:52-15:04	375	The role of farms of Galliformes birds during 2016-2017 avian influenza epizootic in Europe - V. Tsiouris, V. Tarantili, A. Mitsi, E. Sossidou, I. Georgopoulou/Greece
15:04-15:16	434	Effect of time period between hatching and the onset of feeding on the physiology and growth of broiler chicks - R. Cepero, M. Mar Campo, M. Bataller, A. Fernández/Spain
15:16-15:28	482	Perch use by broiler breeders in commercial flocks - B. Spindler, A. Brandes, M. Franziska Giersberg, N. Kemper/Germany
15:28-15:40	208	Qualification and continuing education for people working in the poultry sector -F. Kaufmann, L. Klambeck, I. Angela Goy, H. Grygo, R. Andersson/Germany

Session: Egg Safety and Quality – Hall: Nocturno
Chairs: Y. Nys, H. Medić

Time	*Abstract ID*	*Keynote lecture*
13:40-14:00	30	Effect of yolk on functional properties of chicken eggs **M. A. Grashorn**, F. Hüeber, N. Kretzschmar/Germany
		Discussion
Time	*Abstract ID*	*10-minutes oral presentations*
14:05-14:17	117	Production and characteristics of functional egg based products with high biological and nutritive value - V. Mazo, I. Stefanova, A. Kavtarashvili, I. Mokshantseva/Russian Federation
14:17-14:29	247	Effect of dietary microalgae oil supplementation on fatty acid composition and sensory profile of table eggs in laying hens - J. Wang, G. Qi, S. Wu, H. Zhang/China
14:29-14:41	343	Influence of eggshell quality and egg white protein composition on *Salmonella* spp. Contamination - N. Domínguez Gasca, M. Hincke, A. Muñoz, A. Rodríguez-Navarro/Spain
14:41-14:53	307	Evaluation of antioxidative capacity of egg by using the ORAC and SOAC assay - Y. Wang, Y. Tanaka, H. Hatta/Japan
14:53-15:05	403	EDIL3 and MFGE8: key proteins in the biomineralization of the hen eggshell - L. Stapane, N. Le Roy, J. Ezagal, J. Poirier, V. Labas, L. Combes-Soia, J. Gautron/France
15:05-15:17	405	Proteomics and ultrastructural study of the Guinea fowl eggshell at key stages of the biomineralization - N. Le Roy, A. Brionne, L. Combes-Soia, V. Labas, A. Rodriguez-Navarro, C. Rivard, Y. Nys, J. Gautron/France
15:30-16:00		**COFFEE BREAK AND VIEWING POSTERS**
16:00-18:00		**Parallel Oral Sessions**

Session: Nutrition 4 / Alternative Sources of Nutrients - Hall: Elafiti 1
Chairs: B. Svihus, C. Garces Narro

Time	*Abstract ID*	*10-minutes oral presentations*
16:00-16:13	546	Effect of novel soya bean meal processing technologies on broiler performance and digestibility - P. Sakkas, E. Royer, S. Smith, P. Carré, A. Quinsac, I. Oikeh, I. Kyriazakis/United Kingdom
16:13-16:26	249	Dietary Schizochytrium oil supplementation affect n-3 LC-PUFA enrichment in egg yolk, plasma lipid metabolism and egg yolk quality in laying hens - S. Wu, G. Qi, H. Zhang, H. Wang/China
16:26-16:39	105	Effects of supplementing microalgae in laying hen diets on productive performance, color and content of carotenoids and fatty-acid profile of yolks - Z. Janjecic, D. Bedekovic, K. Kljak, M. Gorupic, M. Musulin/Croatia
16:39-16:52	567	Effect of microencapsulated dry chestnut wood extract and salts of butyric acid on production parameters of broiler chickens and laying hens - H. Valpotić, N. Mas, Ž. Mikulec, M. Đurić Jarić, D. Brozić, Ž. Gottstein/Croatia
16:52-17:05	131	Effect of pea extrusion and probiotic supplementation on performance, microbiota activity and biofilm formation in the gastrointestinal tract of broilers - P. Konieczka, K. Nowicka, S. Smulikowska/Poland

17:05-17:18	116	The effect of pelleting on starch digestion rate of soft and hard wheat in broiler chickens - Y. Khanfas/Canada
17:18-17:31	77	Emulsifier and multi-carbohydrase in a maize, SBM, rapeseed meal and palm oil diet for broiler chickens - S. Kaczmarek, M. Kubiś, M. Hejdysz, P. Konieczka, P. Górka, J. Flaga/Poland
17:31-17:44	461	Preference tests in laying hens; the choice between insect type and treatment - J. Zandvliet, E. Beitler, C.M. Verwer/The Netherland
17:44-17:57	590	Influence of insect (*Hermetia illucens*) or algae meal (*Spirulina platensis*) in broiler diets on growth performance, precaecal digestibility and intestinal microbiota of meat type chickens C. Neumann, F. Liebert, S. Velten/Germany

Session: Nutrition 5 / Nutrition and Poultry Health - Hall: Elafiti 2

Chairs: P. Trefil, H. Valpotić

Time	Abstract ID	*12-minutes oral presentations*
16:00-16:15	400	Impact of fumonisins and deoxynivalenol on laying hens and the effect of a counteracting strategy - B. Doupovec, G.Bichl, S. Masching, D. Schatzmayr/Austria
16:15-16:30	129	A study of coccidiostat and phytase supplementation effects on precaecal phytate breakdown and P digestibility in broiler chickens - S. Künzel, V. Sommerfeld, I. Kühn, M. Rodehutscord/Germany
16:30-16:45	481	Performance enhancing effect of a natural resin acid composition in broiler chickens under a variety of challenge conditions - E. Valkonen, H. Kettunen, J. Kivinen, J. Vuorenmaa/Finland
16:45-17:00	325	Dose-effect relationship and safe use of a matrix-encapsulated phytogenic product based on carvacrol, cinnamaldehyde, capsaicin and cineol in diets for broiler and laying hens - T. Borchardt, K. Maenner, J. Zentek/Germany
17:00-17:15	562	Effect of a multi-strains yeast fractions product plus anticoccidial on performance and gut health of broiler chickens under Eimeria challenge - I. Giannenas, V. Tsiouris, E. Bonos, V. Demey, G. Filliousis, I. Georgopoulou, I. Stylianaki, P. Florou-Paneri, E. Christaki/Greece
17:15-17:30	257	Formulation of Bacillus-based probiotics is key to product performance - K. Sidelmann Brinch, A. Nelson, G. Lafitte, R. Plowman/Denmark
17:30-17:45	299	Dietary arabinoxylan-oligosaccharides (AXOS) kick start arabinoxylan degradation in the ageing broiler - A. Bautil, J. Verspreet, J. Buyse, M. R. Bedford, C. M. Courtin/Denmark
17:45-18:00	285	The effect of feeding Diamond V fermentation metabolites on reducing Salmonella prevalence, numbers and NARMS panel antibiotic resistance in samples taken from commercial broiler breeder hens - W. Abdelrahman, J. P. McGinnis, S. A. Carlson, D. R. McIntyre, H. O. Pavlidis/USA

Session: Poultry Health 4 / Parasites - Hall: Elafiti 3

Chairs: O. Zorman Rojs, J. de Wit

Time	Abstract ID	*Keynote lecture*
16:00-16:20	396	Impact of the poultry red mite, *Dermanyssus gallinae* on the European poultry production systems - **O. Sparagano**, F. Tomley, R. Finn, K. Tiligada, L. Roy, D. Horvatek Tomic, A. Giangaspero/United Kingdom
Time	*Abstract ID*	*10-minutes oral presentations*
16:25-16:37	196	The effects of treatment with fluralaner on poultry red mite infestation and on production of laying hens housed in enriched cages and aviaries N. Sleeckx, K. Van Hoye, I. Kempen, P. De Herdt, S. Cardinaels, K. De Baere, R. Koopman, S. Van Gorp, J. Zoons/Belgium
16:37-16:49	303	Effect of fluralaner treatment on the production results at commercial layers farms in Poland - S. Doner, D. Furmanek/Poland
16:49-17:01	145	Efficacy of fluralaner administered through drinking water with or without brilliant blue in layer hens suffering from poultry red mite infestation and effect of treatments on performance - P. De Herdt, K. Van Hoye, R. Koopman, S. Van Gorp, J. Van Erum/Belgium
16:01-17:13	115	Field trials and experiences with fluralaner against poultry red mite in the UK - T. Cserep, K. Pitman/United Kingdom
17:13-17:25	603	*Dermanyssus gallinae* as a vector of selected bacterial and viral diseases in flocks of hens in Poland - L. Gawel, J. Urbanowicz, P. Falkowski, K. Bobrek, A. Gaweł/Poland

Session: Poultry Welfare 3 / Other Topics - Hall: Elafiti 4

Chairs: E. Sossidou, M. Petracci

Time	Abstract ID	*8-minutes oral presentations*
16:00-16:10	64	Temperature training in the hatcher – effects on robustness, performance and production efficiency in dual- and laying-type cockerels - B. Tzschentke, I. Halle, M. Lieboldt, M. Henning/Germany
16:10-16:20	39	Evaluation of development of foot pad dermatitis in Pekin ducks under commercial conditions - L. Klambeck, F. Kaufmann, N. Kemper, R. Andersson/Germany
16:20-16:30	42	Organic fattening of turkeys: investigations on animal welfare M. Krautwald-Junghanns, M. Huchler, T. Bartels, D. Freihold, S. Thieme, R. Müller, M. Coenen, F. Deerberg, H. M. Hafez/Germany
16:30-16:40	179	Effect of a chemical litter amendment on animal-based welfare indicators and litter quality in a European commercial chicken husbandry - K. Toppel, F. Kaufmann, H. Schön, M. Gauly, R. Andersson/Germany
16:40-16:50	334	Electrical stunning of poultry: impact factors of stunning efficiency and meat quality - M. Bourin, E. Baeza, C. Souchet, K. Anger, C. Le Bourhs, L. Bignon/France
16:50-17:00	344	Genetic diversity – Potential for diversity in adaption and productive quality among chicken breeds - I. Tiemann, S. Hillemacher, M. Wittmann, K. Schellander/Germany
17:00-17:10	359	Use of perches and grids during light and dark period by growing chickens differing in growth intensity - J. Malchow, J. Berk, L. Schrader/Germany

17:10-17:20	374	An integrated farming concept with the use of dual-purpose chickens: investigations of aspects related to animal health and immunity M. Auerbach, M. Dobner, C. Sürie, E. Mundt, W. Icken, S. Rautenschlein/ Germany
17:20-17:30	427	Spontaneous intake of essential oils after a negative postnatal experience in chicks and long-term effects on blood transcriptome L. A. Guilloteau, A. Collin, A. Foury, J. Helbling, A. Koch, S. Crochet, E. Cailleau-Audouin, P. Constantin, S. Lagarrigue, C. Désert, P. Chartrin, M. Moisan, C. Leterrier/France
17:30-17:40	441	Walking on tiptoes: Description of alterations of the digital pads as one indicator of foot pad dermatitis in turkeys - J. Stracke, B. Spindler, N. Kemper/Germany
17:40-17:50	499	Specification for light sources in poultry - J. Daniel Kämmerling, S. Döhring, F. Kaufmann, R. Andersson/Germany
17:50-18:00	22	The point and purpose of rearing male chicks - C. von der Crone/ Germany

Session: Physiology - Hall: Nocturno
Chairs: B. Tzschentke, C. Hess

Time	Abstract ID	12-minutes oral presentations
16:00-16:15	483	Comparative assessment of renal plasma flow and tubular secretion in five different poultry species by determining the para-aminohippuric acid clearance - L. Stroobant, S. Croubels, L. Dhondt, J. Millecam, S. De Baere, G. Antonissen/Belgium
16:15-16:30	98	The postprandial activity of digestive enzymes in pancreatic juice and blood serum in chicken - A. A. Grozina, V. G. Vertiprakhov, V. I. Fisinin/ Russian Federation
16:30-16:45	336	Monitoring of turkey breeders growth and composition through rearing to photostimulation as influenced by body weight category at 16 weeks of age - M. Dewez, S. Brière, P. Froment, P. Etourneau, F. Lecompte/ France
16:45-17:00	113	Effects of organic minerals, fish oil and hydrolysed collagen in broiler diets on growth performance and tibia characteristics - B. Can Güz, R. Molenaar, I. de Jong, B. Kemp, H. van den Brand, M. van Krimpen/The Netherlands
17:00-17:15	162	Impact of thermal manipulation during embryogenesis on hepatic metabolism in ducks - W. Massimino, S. Panserat, S. Davail, M. Houssier/France
17:15-17:30	230	Body weight is affected by early life feeding strategy and hatch moment in broiler chickens - M. S. Hollemans, M. W. Noorloos, S. de Vries, A. Lammers/The Netherlands

09:00-18:00	Poster Sessions Meeting Place - Business Center
Egg safety and quality	
Physiology	
Poultry housing and management	
Poultry meat quality and safety	
Nutrition 2	

18:00-19:30	**Council Meeting - Hall: Elafiti**
20:00-22:30	**Gala Dinner - Revelin Fortress**

THURSDAY, 20th September 2018

09:00-12:30	**Plenary Sessions - Hall: Elafiti**
09:00-10:45	PLENARY SESSION 5 / Chair: E. Prukner-Radovčić
09:00-09:40	Campylobacter infection compromises broiler health, welfare and performance - Humphrey T. / United Kingdom
	Discussion
09:45-10:25	Viral infections of poultry – the globally challenging situation - Savic V. / Croatia
	Discussion
10:30 - 11:00	**COFFEE BREAK AND VIEWING POSTERS**
11:00-12:30	PLENARY SESSION 6 / Chair: H. Mohamed Hafez
11:00 - 11:40	Metagenomic insights into the dynamics of microbial communities in poultry and poultry products: current challenges and future opportunities - De Cesare A. / Italy
	Discussion
11:45 - 12:25	Microbiota, chickens gut health and antibiotic reduction/resistance - Rychlik I. / Czech Republic
	Discussion
12:30 - 13:30	**LUNCH BREAK**
13:40-15:40	**Parallel Oral Sessions**

Session: Nutrition 6 / Proteins in Poultry Nutrition - Hall: Elafiti 1
Chairs: A. Cahaner, M. Cassandro

Time	Abstract ID	10-minutes oral presentations
13:40-13:52	385	Feeding broiler breeders with guanidinoacetic Acid (GAA) supplementation affect creatine egg resources, elevates laying percent and improves progeny's performance - T. Epstein, D. Gravitz, A. Cahaner, Z. Uni/Israel
13:52-14:04	21	Response of broiler chicken fed diets supplemented with dry powdered gluco- and manno-protein yeast cell wall extracts - E. U. Ahiwe, M. Al-Qahtani, E. Chang'a, M. Abdallh, H. Gausi, H. Graham, P. Iji/Nigeria, Australia
14:04-14:16	630	Effect of low protein diets on nitrogen utilization and litter quality of broilers - O. Cirot, W. Lambert, M. Létourneau-Montminy/France
14:16-14:28	464	Valine requirement under low crude protein diets in Ross PM3 broiler chickens - M. Lessire, H. Juin, W. Lambert/France
14:28-14:40	263	OH-Methionine is as efficacious as DL-Methionine to sustain growth performance in ducks and improves their antioxidant capacity - D. Batonon-Alavo, L. Zhao, N. Zhang, N. Zhang, L. Zhu, L. Ma, M. Mohamed Khali, D. Qi, L. Sun, Y. Mercier/France

14:40-14:52	135	Isocaloric and isonitrogenous replacement of soybean meal with other legumes influenced growth performance and ileal amino acid digestibility in young broiler chickens - O. Olukosi, J. G M Houdijk/ United Kingdom
14:52-15:04	421	Whole wheat supplemented with butyric acid influence growth performance, gut development and apparent ileal digestibility of protein and amino acids in broilers fed different protein sources S. Nawaz Qaisrani, A. I. Hussain, M. Salman, Saima, T. N. Pasha, J. A. Bhatti, F. Azam/Pakistan
15:04-15:16	592	Effect of sexing and dietary incorporation of sugar cane molasses on broiler performance and carcass characteristics - M. Saeed Babiker Mahmoud, S. Abdalla Abd Elrheem Abdalla, H. Osman Abdalla/Sudan
15:16-15:28	594	Chemical analysis and tannin content of some raw and germinated sudanese local varieties of sorghum and their potential as poultry energy feed source for food security in rural areas M. Saeed Babiker Mahmoud, E. Osman Eltayib/Sudan

Session: Poultry Health 5 / Gut Health - Hall: Elafiti 2
Chairs: A. de Cesare, M. Hess

Time	*Abstract ID*	*Keynote lecture*
16:00-16:20	326	Comparison of the gut microflora composition between different commercial layer farms: feasibility and limitations - S. Rautenschlein, H. Kathy Scholtes, J. Kamphues, I. Rychlik, F. J. Hoerr
		Discussion
Time	*Abstract ID*	*10-minutes oral presentations*
14:05-14:15	323	Interaction of *Campylobacter jejuni* with the gut barrier of broiler chickens: *Campylobacter jejuni* has diametral effects on broiler gut health - W. Awad, C. Hess, K. Dublecz, J. Aschenbach, M. Hess/Austria
14:15-14:25	584	Fungal fermented feed ingredient for improved gut health in broiler chickens - P. Roubos-van den Hil, C. Silva, B. D'heer, J. Allaart, C. Smits/The Netherlands
14:25-14:35	581	Similar longitudinal development of cecal microbiota diversity within four broiler houses at two different farms - J. G Kers, F. C. Velkers, E. A.J. Fischer, P. Konstanti, J. E. de Oliveira, J. Arjan Stegeman, H. Smidt/ Belgium
14:35-14:45	413	Identification of faecal protein biomarkers for intestinal health in broilers - F. De Meyer, V. Eeckhaut, A. Dedeurwaerder, R. Ducatelle, M. De Gussem, F. Van Immerseel/Belgium
14:45-14:55	624	Assessment of the efficacy of fecal sampling in representing gastrointestinal microbiota in chickens - W. Yan, J. Zheng, C. Wen, C. Ji, D. Zhang, Y. Chen, C. Sun, N. Yang/China
14:55-15:05	429	Beyond effects on the microbiota, *Bacillus subtilis* 29784 shows direct effects on the host - D. Prévéraud, V. Jacquier, A. Nelson, L. Rhayat, E. Devillard/France
15:05-15:15	419	Bacillus subtilis 29784 prevents a pro-inflammatory response in an induced inflammation condition using the Caco-2 cells model D. Prévéraud, L. Rhayat, E. Devillard, E. Eckhardt, M. Maresca/France
15:15-15:25	252	Comparison of preventive ionophore supplementation on gut health and performance in broilers: evaluation in a necrotic enteritis model M. Marien, B. Dehaeck, M. Vereecken, M. Geerinckx, K. De Gussem/ Belgium

15:25-15:35	143	Establishment of the cecal microbiota of broiler chickens supplemented with protected sodium butyrate alone or in combination with essential oils and challenged with coccidia and *Clostridium perfringens* C. Bortoluzzi, M. Rothrock, B. S Vieira, J. Jose Mallo, M. Puyalto, C. Hofacre, T. Applegate /USA

Session: Reproduction and Incubation - Hall: Elafiti 3
Chairs: S. Yalcin, F. Sirri

Time	*Abstract ID*	*10-minutes oral presentations*
13:40-13:50	371	Ultrastructural development of the yolk sac tissue during incubation J. Dayan, N. Reicher, Z. Uni/Israel
13:50-14:00	93	Influence of incubation temperature on the development of the Bursa of Fabricius in the chicken (*Gallus gallus*) embryo - R. Noiva, A. Menezes, H. L. Shivaprasad, M. C Peleteiro/Portugal
14:00-14:10	94	Changes in gizzard development caused by high incubation temperatures in the chicken (*Gallus gallus*) embryo - R. Noiva, A. Menezes, H. L. Shivaprasad, M. C Peleteiro/Portugal
14:10-14:20	100	Both the rooster and incubation temperature affect embryonic metabolism and day-old chicken quality in laying hens - H. van den Brand, S. van de Kraats, A. Sözcü, R. Jöerissen, M. Heetkamp, I. van den Anker, M. Ooms, B. Kemp/The Netherlands
14:20-14:30	187	The in ovo critical period for somatotrophic axis elevation by green light photostimulation - I. Dishon, N. Avital-Cohen, J. Bartman, S. Zaguri, I. Rozenboim/Israel
14:30-14:40	233	Embryonic thermal manipulation of the Japanese quail: impacts on physiology and hypothalamic methylome and transcriptome A. Vitorino Carvalho, N. Couroussé, F. Gataud, C. Gimmonet, S. Crochet, T. Bordeau, M. Mersch, B. Piégu, C. Hennequet-Antier, A. Brionne, C. Noirot, Y. Bigot, F. Pitel, A. Collin, V. Coustham/France
14:40-14:50	245	Chick yolk mineral levels during their sojourn in the hatcher R. Hopcroft, W. Muir, P. Groves/Australia
14:50-15:00	255	Effects of in ovo injection of N-Carbamylglutamate (NCG) on hatchability, quality of chickling, growth performance and carcass composition in broiler chickens - G. Qi, H. Zhang, J. Wang, S. Wu, Y. Ma, F. Zhang/China
15:00-15:10	324	Applying cold incubation profiles during the last week of incubation in a commercial incubator: effects on broiler embryonic mortality, hatchability, and chick quality - I. van Roovert, M. van Eijk-Priester, J. Wijnen, C. van der Pol/The Netherlands
15:10-15:20	384	The effect of combined monochromatic photostimulation on reproductive activities of broiler breeders - J. Bartman, S. Zaguri, L. Dishon, N. Avital Cohen, I. Rozenboim/Israel
15:20-15:30	420	Temperature variations during incubation and postnatal period affect performance, metabolism, health and gene expression in the blood of fast-growing chickens - A. Collin, V. Coustham, N. Couroussé, S. Crochet, C. Praud, T. Bordeau, E. Godet, E. Cailleau-Audouin, P. Chartrin, I. Gabriel, C. Schouler, T. Larcher, C. Le Bourhis, K. Germain, O. Zemb, A. Travel, C. Berri, L. A. Guilloteau/France
15:30-15:40	591	Assessment of GnRH analogues efficacy in enhancing the reproductive functionality in broiler breeders - G. Kolluri, G. Marappan, J. Mohan, H. Ali, J. Singh Tyagi/India

Session: Poultry Health 6 / Other Topics - Hall: Elafiti 4

Chairs: S. Ozkan, M. Toscano

Time	Abstract ID	Keynote lecture
13:40-14:00	357	An innovative approach for analysing and evaluating poultry farms odour related to animal health and welfare - **V. Ferrante,** *G. Grilli, F. Borgonovo, L. Susanna, E. Tullo, M. Guarino*/Italy
		Discussion
Time	*Abstract ID*	*8-minutes oral presentations*
14:05-14:15	480	Bio-monitoring of mycotoxins in blood serum and feed to assess exposure of broiler chickens in Belgium G. Antonissen, M. Lauwers, N. Caekebeke, M. Ringenier, F. De Meyer, J. Dewulf, S. Croubels/Belgium
14:15-14:25	34	Biomarkers as a tool to better understand the effect of mycotoxins on gut barrier functioning and immunosuppression - A. Koppenol, L. Caron/Brazil
14:25-14:35	311	Transcriptomics approaches in enteric inflammation models for the assessment of dietary supplements - B. Grenier, S. Fibi-Smetana, S. Ilic, R. Berrios, C. Emsenhuber, V. Nagl, G. Schatzmayr, N. Reisinger/ Austria
14:35-14:45	289	Supplementation of Diamond V fermentation metabolites in feed or drinking water to reduce stress in broilers - W. Abdelrahman, A. Kovács, H. O. Pavlidis, D. R. McIntyre, G. Archer/USA
14:45-14:55	78	A retrospective analysis to identify the contribution of the breeder on health and performance of broiler chickens - I. de Jong, J. van Riel/The Netherlands
14:55-15:05	132	Released-active drugs enhance poultry production performance K. Ganina, E. Karelina, P. Kudryavtsev, S. Tarasov/Russian Federation
15:05-15:15	301	Effects of a dry hydrogen peroxide (DHP) air sanitation system used in an egg cooler on hatchability and chick quality - B. Jordan, E. Melo, J. McElreath, R. Stephens, L. Lara, N. Cox, J. Wilson/ USA
15:15-15:25	369	Effects of various housing conditions and genotypes on health and performance of tom turkeys - J. Berk/Germany
15:25-15:35	24	Prevention of MG-MS infection using live vaccines - L. Kőrösi, J. Povazsán/ Hungary
15:35-15:45	370	Insights on avian metapneumovirus subtype B circulation in Europe - C. Lupini, G. Mescolini, G. Franzo, A. Blanco, M. Biarnes, M. Cecchinato, C. Maria Tucciarone, V. Felice, F. Silveira, E. Catelli/Italy

09:00-18:00	**Poster Sessions Meeting Place - Business Center**
	Economics and marketing
	Education and information
	Family poultry production
	Poultry health
	Reproduction and incubation
	Turkeys
	Nutrition 3
	Other Topics
15:45-16:30	**Coffee break and viewing posters**
16:30-17:30	**Closing ceremony (Hall Elafiti)**

FRIDAY, 21th September 2018

08:00-17:00	**TECHNICAL VISIT - Meeting place: Hotel Lacroma**
09:00-	**YOUTH PROGRAMME - Meeting place: Hotel Lacroma**

Disclaimer: The information in the XVth European Poultry Conference 2018 programme is correct at the time of providing, however, the organizers reserve the right to change the information where necessary without notice.

PLENARY LECTURES

Contents

Are there limits to selection in poultry: theoretical, biological, ethical, environmental?

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Running title: Selection limits in poultry

Summary: In theory, selection may reach a plateau when all favorable alleles have reached fixation. Yet, current data in poultry show that selection response can still take place after 50 generations of selection or more. The mechanisms maintaining selection response in closed populations may involve recombination, mutation and epistatic interactions. Furthermore, the continuous addition of new selection criteria can delay the possible limit associated to single trait selection. Thus, selection response is mainly threatened by inbreeding which occurs as a consequence of a narrow genetic base and/or a poor management of genetic variability within the population. Biological limits are encountered when selection is degrading fitness traits to a point that the survival of individuals is affected. Biological limits induced by extreme performance can be by-passed by adapting the breeding program, introducing new selection criteria, changing the management or developing remedial technologies. Extreme situations affecting bird welfare raise ethical issues. Lameness in broilers or spontaneous bone breakage in layers are painful and one may question whether such pain is justified by the human need for protein consumption. Regulations or market requirements may be set up to limit the performance at a level which is compatible with animal welfare, resulting in a voluntary limit to selection. Furthermore, highly performing animals need a very well controlled environment with high quality diets, which may divert food resources from humans and may not be sustainable. Breeding objectives have to integrate environmental impact and robustness towards the use of alternative feed sources, in addition to production level, product quality, health and welfare status. Overall, poultry production systems can be viewed as special ecosystems, delivering services but also impacts to human societies, that need to be balanced to improve robustness of the food system.

Keywords: *Selection, limits, welfare, ethics, environment, services, sustainability*

Introduction

Large scale selection of poultry lines has now been in operation since almost 70 years, which is about 70 generations for chickens, and has greatly modified poultry performance. At that point, one may question whether this can go on forever, in which conditions, for which food system? This paper provides an overview of the different mechanisms or factors that may limit selection response, and examine strategies to handle such limits.

1. Selection theory and possible limits

1. Principles

Selection theory has been established only a century ago and has been applied at a large scale in poultry breeding. The power of this theory is that it relies on a simple and generalist model, the infinitesimal model, which sets the hypothesis that a large number of genes, each of them having a small additive effect, is controlling the variation of a quantitative trait. Each gene is generally considered to have two alleles, and the allelic substitution effect is measured by the change in trait value when one allele is replaced by the other one. Furthermore, allelic effects are considered to be independent from one gene to another and assumed to be normally distributed, so that the genetic value of an individual for a trait is simply the sum of allelic effects at all genes. The genetic value cannot be measured directly and is estimated by comparing the performance of related animals, the higher the similarity between related animals, the higher the contribution of additive genes. Successful selection is thus expected to progressively increase the frequency of favorable alleles, so that selection should reach a plateau when all favorable alleles have been fixed. At that stage, variation of performance will just reflect environmental variation and there will be no more response to selection: the limit will be reached. The smaller the initial population size, the faster the limit will be reached. The history of the concepts underlying animal breeding has been revisited by Hill and Kirkpatrick, (2010), from the viewpoint of evolutionary biology. Referring to Robertson, (1960), they reminded that "*half of the ultimate (selection) response occurs by $1.4N_e$ generations*" where N_e is the effective population size. Thus, for a population with a N_e of 40, which is rather small, selection response should still take place after 56 generations. Since poultry lines generally have a N_e value higher than 40, no selection limit is expected at present. Thus, theory predicts that limits will occur. Yet, real selection differs from theory for several reasons:

- Selection plateau can be expected if the selection objective is always the same, but as soon as a new trait is added in the selection objective, different genotypes may perform better and the set of most favorable alleles is changing;
- Selection plateau is tightly associated with the additive model, which is an oversimplification of genetic reality, ignoring dominance and epistasis; as soon as interactions between genes take place, the ranking of genotypes will change, and these interactions will contribute to maintain genetic variation;
- Furthermore, if allelic effects of a given gene are influenced by alleles of the neighboring genes, the genetic value of an individual will be modified when recombination occurs and modifies the 'neighbor alleles'. This would have no effect in animals which would be homozygous for genes flanking the recombination, so that identical alleles will remain neighbors;
- New alleles may arise by mutation, but this phenomenon is rare.
- Allelic effects may depend on the environment where the trait is measured, genotype x environment interactions are disturbing prediction of breeding values and may prevent fixation of a 'best genotype'.

Thus, the lack of any selection response would correspond to a fully inbred line or to cloned individuals where all candidates to selection are homozygous for the same alleles. This is one of the reasons why inbreeding is a major concern in all breeding programs. Inbreeding induces allelic fixation at random: some inbred lines may perform better than others by chance, and most of them are likely to exhibit genetic defects. Although the occurrence of genetic abnormalities may not limit selection per se, it generally decreases the number of candidates to selection and decreases selection intensity.

In conclusion, there are many factors that postpone or even avoid genetic limits to selection. A time frame of 60 or even 70 generations of selection, although it represents an impressive amount of work and genetic progress, is still very short as compared to the scale of natural selection. The main limit to selection could come from inbreeding, either because of a narrow genetic base or because of a poor management of genetic variability in the breeding plan.

2. Evidence from poultry populations

Commercial lines

A selection plateau does not seem to have been encountered in commercial poultry species, but estimates of genetic trends are not available. Inbreeding levels can be estimated by pedigree analysis and more recently by the use of molecular markers. All studies generally show that observed heterozygosity is lower in white-egg layers than in brown-egg layers and lower in egg layer lines than in broiler lines (Granewitz *et al.*, 2007), with a reverse order for inbreeding. Some additional considerations may explain this difference:

- layers and particularly white-egg layers originate from a single breed (White Leghorn) or a small number of breeds, which results in a narrower gene pool than in broilers;
- early inbreeding may have been used in layer breeding more often than in broiler breeding;
- layers have been selected for a larger number of traits than broilers which may have led to allelic fixation at a higher number of loci.

As long as this phenomenon is slow, no visible consequence is observed. However, a high inbreeding level may often lead to difficulties such as low reproductive ability. This could be related to an increased frequency of deleterious alleles, such as recessive lethals. This has been recently studied in layers, ans a higher ratio of deleterious variants, as compared to synonymous variants, has been observed in the most inbred of 3 layer lines (Derks *et al.*, 2018). Segregation of deleterious variants is observed in other selected populations (cattle, pigs) and may be a source of limitation of response to selection.

Experimental lines

Selection experiments are generally performed on populations with a limited size, for a variable number of generations. There are a few very long term selection experiments in poultry that would make possible to observe selection limits. The longest selection experiment for a domestic bird has been achieved by Henry Marks who selected quails on 4 wk body weight for 97 generations (Marks, 1996). Two lines were selected, one being fed on an optimal diet, the P line, the other being fed on a low protein diet, the T line. Selection response was regularly observed in the P line until generation 97, with some transient plateau, whereas longer plateau were observed in the T line, but a selection response was still observed after generation 90. Several trials were made during the experiment by deriving sublines selected with a different diet composition: it was concluded that changing the selection environment boosted selection response.

In chickens, the longest selection experiment is most probably the divergent selection experiment on 8wk body weight, conducted by Paul Siegel and collaborators for 54 generations (Dunnington *et al*., 2013). No selection limit was yet encountered for the high body weight line after 54 generations whereas a limit was observed in the low line, starting earlier in females than in males, with no selection response after generation 50. A similar asymmetry in selection response was observed in White Leghorn lines divergently selected for antibody response to sheep red blood cells: at generation 36, selection response was still taking place in the high line whereas a selection limit occurred after generation 12 for the low line (Zhao *et al*., 2012).

These selection experiments showed little evidence of selection limits for high value of the trait even in a population of limited size (8 sires for each of the body weight lines of Paul Siegel). The mechanism maintaining selection response in such closed populations has not been established clearly: mutations were generally thought to have taken place, but the impact of recombination breaking critical haplotypes should also be considered. The situation was different in lines selected for low value of the trait, where selection plateau was more frequently observed, probably as a result of a physiological limit. Indeed, these experiments also illustrated the importance of unfavorable correlated selection responses which are creating limits to selection response for biological reasons.

2. Biological limits to selection

Biological limits to selection may be encountered either when the trait(s) under selection cannot be measured or is/are not accessible because of a biological limit, or when selection is degrading fitness traits to a point that the survival of individuals is affected, selection intensity is decreased and, ultimately, the population itself may be lost, suppressing any possible selection. Thus, biological limits are very much related to unfavorable correlated selection responses. Furthermore, some of the long-term selection experiments have shown the importance of a balanced use of resources to maintain the physiological functions such as growth, immunity and reproduction: when such a balance is disrupted, the fitness of selected birds will decrease. This has been confirmed in commercial broilers by Cheema et al (2003) who observed a change in the immune function of fast-growing modern broilers as compared to slow-growing broilers (a control population from 1957) with a higher inflammatory response and a lower antibody response in modern broilers.

1. Broilers

Broiler selection has put a major emphasis on early growth rate since many generations. Unfavorable correlated responses have been observed on fitness traits:

- Skeletal development and walkability: selection has been applied on breast muscle, with little consideration of the mechanical balance of the bird. The center of gravity of broilers with heavy breast is different from that of a slow-growing bird, and this puts pressure on legs and bone strength to maintain walkability and leg integrity. Several leg disorders have been described (tibial dyschondroplasia, bone deformities, valgus-varus deformities.) which are now considered to be a consequence of extremely rapid growth as reviewed by Angel *et al.,* (2014).

- Cardio-vascular insufficiency: selection has been applied on breast muscle, with little consideration to the supply organs, such as the cardio-vascular system needed to provide oxygen to fast growing muscle. The increase in muscular mass has not been accompanied by increased vascularisation, which has resulted in large diffusion distances for oxygen and metabolites, as reviewed by Petracci *et al.*, (2014). This has two major consequences for selection: an increased incidence of myopathies with meat quality defects and the occurrence of a cardiac insufficiency syndrome with ascites.

Two strategies can be adopted to overcome this type of limit: add a new selection criteria targeting the biological pathway affected by the correlated selection response, or modify the environment to decrease the negative impact of the correlated change in the impacted trait. In the case of ascites, both strategies have been tried and the addition of oxygen partial pressure in the blood has proven to be very efficient to avoid cardio-vascular insufficiency. Up to now, the consequence of such biological limits has been an increased complexity of breeding programs, with new traits being added into the program, which does not constitute a real limit *sensu stricto*, but is decreasing the selection pressure on the main trait. However, the accumulation of metabolic disorders is not a good sign for long-term selection and revising the selection objectives with the aim to select more robust animals is recommended.

Another major biological issue which may really limit selection in broilers is the antagonism between early growth and reproduction: at present, broiler breeders need to be feed restricted by up to 90% of their voluntary intake in order to maintain a sufficient reproductive capacity. Feed restriction is here a management solution to an unfavorable correlated response, but obviously there is a zero limit to feed restriction ! As pointed out by M. Vaarst (2014), there are situations in nature where a species needs to grow very fast for a short period, with little impact on adult body weight which remains rather constant for the reproductive period. In chickens, this would mean selecting on the shape of the growth curve, which was shown to be possible by Fernand Ricard in France, who selected a line for a high juvenile body weight and a moderate adult body weight (Mignon-Grasteau *et al*., 2001). This breeding approach would be worthwhile in order to avoid the biological collapse of heavy birds not being able to reproduce.

2. Layers

In layers also, unfavorable trends have affected the biological balance of the bird.

- Osteoporosis is a consequence of the major pressure put on calcium metabolism and bone remodeling by eggshell synthesis. It may be all the more an issue that the length of the laying period has now increased As in broilers, selection programs have been adapted to overcome this effect, while maintaining the improvement of egg production, and management solutions have been set up, particularly at the level of housing to maintain a physical activity of hens.

- Behavioural problems with feather pecking and cannibalism are more and more often encountered at the commercial stage, but the link with past selection is not so obvious. It had been suggested that selecting laying hens on egg number in individual cages was at the same time selecting for a higher social dominance, the most dominant bird being the one producing more eggs and thus more progeny for the next generation. Under this hypothesis, the development of aggressive behavior would represent a strong limit to individual selection for egg production. Again, management solutions have been proposed, such as debeaking or having the hens wearing preventive devices, but cannibalism and mortality still occur in free-range systems, as reviewed by Elson (2014), although these systems are expected to provide better welfare which is not the case. A change in the breeding program is another approach, such as group-selection or incorporating social relationships in the prediction of the genetic value. However, the genetics of behavioral traits is more complex than the genetics of growth or egg production, because behavior is a matter of interactions, and this represents a methodological challenge for selection, as reviewed by Wade *et al.,* (2010).

Layers also provide a beautiful example of a biologically limited trait: the laying cycle results from the coincidence between the external circadian rhythm of 24h and the endogenous ovulatory rhythm which is more variable. Selection has resulted in having the majority of layers tuned on an internal rhythm of 24h so that the maximal egg production is reached with one egg per day (i.e. the external and endogenous rhythm coincide). Thus, the biological rhythm of ovulation is limiting the laying intensity at one egg a day. Interestingly, a selection experiment conducted by B. Sheldon in the 80s showed that hens could lay one normal egg at shorter intervals than 24h if they were acclimated to constant day light and were not subjected to any external rhythm (Yoo *et al.*, 1986). In this condition, the genetic variability of the endogenous ovulatory rhythm was accessible to selection and hens laying normal eggs every 22h or even 20h were obtained. However, such a management system was complex, probably not cost-effective and the current solution adopted by breeders to overcome the biological limitation is to work on the persistency of lay, where the 'one egg a day' rhythm is prolonged on a longer production period.

3. Ethical limits to selection

Ethical concerns come into perspective for various reasons. In the case of poultry, they have been raised by issues of pain and survival in highly producing birds: lameness in broilers or spontaneous bone breakage in layers are painful and one may question whether such pain is justified by the human need for protein consumption. Considering the extreme feed restriction imposed to broiler breeders, these birds are constantly feeling hungry. The absence of hunger is one of the component of animal welfare, recognized by the European Union. In other words, for the sake of economic efficiency, a chronic state of suffering is imposed to chickens during most of their life. A major ethical issue for laying lines is the culling of male day-old chicks, which results from the high specialization of these lines and the lack of marketing value for very slow growing males. Proposing dual-purpose crosses is a way to overcome it, but coming back to dual-purpose breeds would represent a major reorientation of selection and correspond actually to a selection limit for highly specialized layers.

At first, this looks to be a very emotional motivation, but it raises more philosophical questions: what is the right of humans to cause suffering in other living organisms? This is quite different from the acceptance of killing animals for food. The main issue is that selecting for continuously higher performance should not induce a chronic state of pain for the animals. Is this the responsibility of breeders? The general answer is no, because they need to answer the market demand. Are the slaughterhouses, the distributors of processed food, in other words the leaders of the market demand, aware of the welfare issues occurring upstream in the production chain? Not so much. So nobody feels really responsible for the state of suffering of broiler breeders, there is always a good reason to say that we cannot do differently because of the market demand. That is the reason why NGOs have started to act at the market level, by increasing awareness of the consumers on such issues. The most efficient action in this respect is probably the one conducted in The Netherlands at the level of supermarkets since 2013, called the 'Chicken of Tomorrow': supermarkets modified their requirements to their providers with a limit imposed to the growth rate of broilers (=<50g/day) together with density and environmental conditions. Consequently, the market share of slow-growing broilers significantly increased in the Netherlands up to 50% in 2016.

Another ethical concern comes from the fact that highly performing animals generally need a very well controlled environment, including a preventive use of antibiotics. This practice has raised the growing issue of microbial resistance to antibiotics, which in turn may affect human health. There seems to be a contradiction here: health monitoring in farm animals with antibiotics raise a health hazard for the human population, but using antibiotics for intensive animal production is generally justified by the need to supply a growing human population with animal proteins. So, a compromise is needed. A one-health approach needs to be adopted and research has to be strengthened regarding the epidemiology and ecology of chicken pathogens in large-scale chicken farms.

4. Environmental limits to selection

1. Environmental factors as a limit to performance: temperature

Among livestock systems, poultry production is probably the one where a strict environmental control has been set up with high requirements, regarding feeding, lighting, temperature, and health. Temperature deserves special consideration for two reasons: poultry production is particularly growing in countries with tropical climatic conditions and heat waves are affecting production in temperature countries, leading to mortality. High economic losses due to heat stress has been documented since many years for livestock production (St Pierre *et al.,* 2003). Highly producing animals are more sensitive to high temperature conditions mainly because they need to dissipate metabolic heat associated to protein synthesis. Of course, performance of selected lines remain higher than that of local unselected populations, but this corresponds to what is called 'yield gap' in plant production, where the observed performance does not match the expected genetic gain. In plants, climate change has been recognized as a main cause of the yield gap. Similarly, high ambient temperature is likely to have the same effect on animal productions, including poultry, by setting a limit to the expression of response to selection.

To overcome this limit, management solutions are predominating, with cooling devices and nutritional strategies. Air conditioning is raising an issue of power supply, and even an ethical issue, since in some countries chickens would be housed in better temperature conditions than humans... in order to feed humans, from the same country or other countries. Tailored nutritional approaches have also been proposed for broilers (Gous and Morris, 2005), but limitation of performance due to heat stress remains an issue. Decreasing ambient temperature to a low value (14°C) in order to facilitate heat dissipation of broilers did not change body weight and meat quality, as compared to broilers raised in a standard ambient temperature of 21°C. This suggested that meat quality defects were not due to inability to dissipate heat, but rather to correlated responses to selection (Zahoor et al., 2016).

Genetic solutions to improve heat tolerance have been proposed with major genes decreasing feather cover, such as the Naked neck gene and more recently the scaleless gene (Cahaner *et al.*, 2008). Yet, there is little adoption of these solutions until now, probably because introducing a major gene or replacing a line by another one is much more complex than changing a selection coefficient or introducing a new trait. Furthermore, the fully naked scaleless chickens was not appealing for consumers. Thus, research is still active in looking for selection criteria that would improve heat tolerance in chickens, but there may be a biological limit in dissipating metabolic heat.

2. Environmental factors as a limit to poultry production

As compared to ruminants, poultry can considered to be on the safe side from the viewpoint of Green-House-Gas production, thanks to the high feed efficiency of poultry commercial lines. Another currently active debate relates to the use of high quality feed for animals, and poultry in particular. Indeed, poultry can have a very good feed efficiency when eating high quality nutrients such as soybean and corn. However, these feed resources can also be used directly for human nutrition, or are grown on arable lands that could be used to produce vegetal proteins for human nutrition. Thus, a potential competition between poultry feed and human food can be seen here, and a land-use approach has been proposed to analyze the contribution of animal products to sustainable diets (Van Zanten *et al.*, 2016). With such an approach, animal proteins from grazing systems are favored. A model has been proposed to define the optimal % of animal protein in human diets, depending on human population size and on the relative share of land unsuitable for crop production (Van Kernebeek *et al.*, 2016). It showed that land was used more efficiently when the % of dietary proteins of animal origin ranged from 15% to 45% for large human populations. Unfortunately, the case of poultry production was not tested in these studies.

Whereas the land use approach of sustainable food systems does not directly set a limit to selection response, it sets a new framework to define selection objectives in poultry. Since breeders and producers are already aware of this debate, diversification of feed resources has been considered. As an example, genetic variability of digestibility has been studied in the case of sup-optimal diets made of wheat, and selection for apparent metabolizable energy corrected for zero nitrogen balance (AMEn) has been shown to decrease environmental impact in broilers (De Verdal *et al.*, 2013). Yet, suboptimal diets may be used at the production level that differ from the diet used for the selection nucleus. This may lead to an environmental limitation to selection response, on the same model as the yield gap, where observed performance stays behind genetic trend.

Discussion

This overview of potential limits to selection on poultry shows that important factors to consider are ethical and environmental. The occurrence of a true selection plateau has not been observed yet, which could be due to the relatively short duration of selection, from an evolutionary viewpoint. Yet, the selection of extreme phenotypes lead to biological limits due to unfavorable correlated responses, as illustrated by the impact of a very high growth rate of broilers on key biological functions (skeleton, cardio-vascular system, reproduction, heat tolerance).

Which strategies to adopt? Whereas management or technological changes provide short-term solutions to compensate for a biological limit to selection, changes in breeding programs have the advantage of being cumulative. This may be achieved at the level of selection criteria, selection methods or selection objectives.

Improving our understanding of the regulatory pathways connecting various traits could help to anticipate correlated selection responses and lead to define selection criteria in order to reach a better balance of resource allocation in selected birds. Such an understanding requires large-scale phenotyping to monitor a range of correlated traits.

A major change in breeding methods has occurred with the introduction of genomic selection which takes advantage of using a high number of SNP markers over the whole genome in order to obtain a genomic prediction of the breeding value, more accurate than the one based on pedigree relationships. It requires genotyping and phenotyping a reference population and can be particularly interesting to estimate breeding values of males for female traits as well as breeding values of a young animal for an adult trait. It can also greatly improve the genetic connection between crossbred performance at the production level and grand-parental lines in the selection nucleus. Poultry breeders have now adopted this method and it has been suggested to be a suitable approach to improve welfare by applying group selection to select against cannibalistic behavior (Alemu *et al.*, 2016). A more efficient selection method opens new opportunities to implement a revision of selection objectives. Yet, the most powerful the breeding methods become, the most responsible the breeders must be regarding society. Defining selection objectives must take into account the contribution of poultry to the whole agricultural and food systems. The challenge is to combine production with welfare and low environmental footprint.

FAO is promoting a transition to sustainable food and agricultural systems on the basis of agroecology (http://www.fao.org/about/meetings/second-international-agroecology-symposium). Maintaining a choice of genotypes for adaptation to a variety of production environments and market needs is a breeding strategy suited to agroecology (Phocas *et al.*, 2016). The choice may include different commercial lines, but also alternative genotypes such as the slow-growing broiler, and different crossbreeding combinations. The main change here is to recognize that there is not a one-for-all genotype.

Maintaining genetic variation is a key for adapting breeding strategies. To avoid the limitation due to low genetic variation, maintaining genetic reserve, either *in vivo* or *in vitro* (i.e. gene banks) offers the possibility to renew the genetic basis of a breeding program by using crossbreeding and/or deriving new composite populations. The overall gene pool of chickens is still high if we consider all local populations, and the wild ancestor species is still existing, which should be viewed as a great opportunity to enlarge the choice of genotypes.

Thus, the limits that can be encountered by selection are not so much on the genetic side but rather consist in the difficulty to establish balanced selection objectives, suited to different populations of consumers, in order to meet the expectation of sustainable food systems for human societies. Whereas poultry breeding has always been very reactive to market trends and already has complex breeding objectives, the new challenge is to promote a sustainable use of feed and energy resources globally.

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How Scientific Innovation is developing tools to improve egg quality, and support development of innovative therapeutic molecules.

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Short title: novel proteins, egg formation and use

Abstract: In laying hens, selection has resulted in a very productive hybrid for a variety of traits associated with egg production, in particular a high persistency of lay. The increased duration of production is however associated with difficulties in terms of egg quality. Such changes in laying hens' management require additional consideration of the physiology of birds, their nutrition and rearing strategies to benefit from the improved genetics. An additional challenge concerns the microbiological safety of eggs produced in alternative systems, in which birds are subjected to higher challenges due to changing climatic and microbiological environments. The egg possesses efficient physical protection (eggshell), as well as many antimicrobial molecules, which can be reinforced for improving egg safety and for increasing the diversity of egg-derived products to promote consumer health. Research in nutrition and rearing systems has led to some innovation for improving hen performance and egg quality by testing the effect on egg quality of novel rearing strategies or nutritional additives (black box approach). Recent proteomic analyses of egg compartments and transcriptomic studies of gene expression in the various segments of the oviduct, in which egg is sequentially formed, have led to the identification of a large range of effectors that participate in egg formation and of novel proteins involved in antimicrobial protection of the egg. Bioinformatic analyses of protein sequences enable the characterization of their putative functions, leading to the identification of uterine ion transporters and eggshell matrix proteins contributing to eggshell formation, by supplying mineral precursors and by controlling its crystal growth, respectively. Numerous in vitro and in vivo evidences have demonstrated the involvement of these proteins in eggshell fabric and biomechanical properties. It is therefore possible to analyze the origin of an eggshell defect and to explore how effectors of eggshell formation are modified by hen physiology or nutrition. These genes can therefore be used to study the regulation of the mineralization process and to define novel markers for genetic selection. Similarly, this high-throughput technology has contributed to the identification of numerous novel antimicrobial proteins distributed in the various egg compartments. This information has been used to understand the origin of the variability in antimicrobial activity of egg white due to hen physiology or during egg storage, and to explore the feasibility of stimulating natural antimicrobial activities of the egg by the hen microbial environment or with live vaccines. Purified antimicrobial proteins from egg white have provided alternative anti-infective agents for human and animal health. This review will summarize current information on the identification of novel egg and uterine proteins and will analyze how this knowledge can be used to define genetic, nutritional or physiological factors controlling variability in egg quality to develop innovative therapeutic molecules

Keywords: *egg, eggshell, novel protein, therapeutic molecule*

Introduction

The first priority for producers, breeding and feed companies is to increase egg production and quality. Continuous improvement in egg number has been initially obtained by selecting hens for early sexual maturity and for improved peak of production. Recently, selection for increasing persistency in lay while maintaining egg quality in older hens (Bain et al., 2016) has permitted the egg production to be extended to more than 80 weeks of age with the objective of keeping commercial flocks up to 100 weeks to get more than 500 eggs in a single cycle. This achievement challenges the metabolism and homeostasis of the hen and of some organs in particular: the liver where yolk protein and lipid syntheses take place and the oviduct where the egg white, eggshell membranes and eggshell are formed. Indeed, the genetic progress on persistency and egg quality is used to postpone the end of the laying cycle and therefore does not solve the problem but just delays the impact of hen age on egg quality. Arrest of egg production is not due to reduced production but to defects in egg shell quality and at a lower magnitude, to defects in Haugh units and the vitelline membrane that deeply compromise the selling of eggs as egg product. Therefore, we need to innovate to find ways to improve egg quality in older hens but also to better analyse the physiological origin of these defects. Hen rearing practices and nutrition are crucial for optimising the genetic potential of modern hens in terms of productivity and high egg quality, to reduce the incidence of bone demineralisation or fatty liver syndrome in aged hens but alleviate rather than solve the problems (Nys, 2017a; 2017b). Alternatively, sorting eggs by analysing their quality allows commercial diversification for different markets. The development of non-invasive techniques to evaluate egg quality as a basis for sorting improves egg safety for consumer and innovation in this area is promising (De Ketelaere et al., 2017). Another approach will be to improve the storage of the egg to avoid further degradation of its lower quality due to hen age. Attempts to store eggs under controlled atmosphere might be an interesting approach but remain poorly studied (Berardinelli et al, 2011). It is well established for example that egg storage under carbon dioxide improves gel properties and Haugh unit of eggs. Egg washing ((Sexton, 2017) and alternative techniques of egg contamination (Berardinelli et al, 2011) have been proposed to improve egg safety by decreasing its initial degree of contamination. An alternative would be to reinforce the natural antibacterial defences of egg by investigating new storage conditions (Guyot et al, 2017). Most of ways to control egg quality are empirical, and experimental solutions exploring the effect of one nutritional or genetic factor on the egg qualitative parameter are a black box in which the physiological or metabolic origin of the positive effect remains unknown. Since the sequencing of the laying hen genome, a large number of genes and proteins have been biochemically and functionally characterised. During the last fifteen years, the development of high-throughput methods used in combination with the available chicken genomic sequence and the development of bioinformatics tools to predict functions have generated new insights for the characterisation of new and minor egg components (Gautron et al, 2011; Guyot et al., 2017) and of the cellular and extra-cellular proteins from organs contributing to egg formation (Jonchére et al., 2012; Sun et al., 2013; Brionne et al., 2014; Marie et al., 2015 Du et al., 2015). This novel information allows us on one hand to better understand the mechanisms of egg formation and its regulation and therefore has the potential to analyse the origin of the effect of an external factor on egg quality. On the other hand, the prediction of putative function of egg proteins by the bioinformatics analysis of their sequence favours the identification of novel and active molecules with specific properties that may be beneficial to human and animal health. The aims of this review are to summarize the novel information derived from the development of proteomic, transcriptomic and bioinformatics analyses of egg and uterine proteins and to underline how this information might contribute to understand the process of egg formation, its regulation and to identify innovative uses of egg components.

Identification of proteins as novel egg molecules or contributing to its formation

The chicken genome sequence (International Chicken Genome sequencing Consortium, 2004; Gal5), the existence of a large number of ESTs (Expressed Sequence Tags representing specific mRNA sequences expressed in the chicken) and the use of high-throughput technologies (proteomics and transcriptomics) have led to the identification of a large number of proteins present in the egg or involved in its formation (Gautron et al;, 2011). In 2006, the total number of identified proteins in the egg was about 50 but has been enlarged to more than 1000 in a very short period of time using these new technology and tools. The egg proteome referring to the complete set of proteins present in the egg has been explored in specific egg compartments by mass spectrometry. These approaches were augmented by studying genes that are specifically expressed in the oviduct segments responsible for the deposition of each specific egg compartments. Egg formation occurs in a precise temporal and spatial sequence as the forming jsynthesis and secretion of the vitelline membrane, egg white, eggshell membrane or eggshell at different times of egg formation. Mann (2007) by using LC-MS/MS and MS3 of peptide mixtures prepared by in solution cleavage of egg white proteins and D'Ambrosio et al. (2008) by using combinatorial peptide ligand libraries identified a total of 148 proteins in the egg white. In parallel, cDNA microarrays were used to determine global gene expression profiling of magnum tissue involved in egg white deposition and allowed the identification of more than 800 genes (Gautron et al., 2011). Proteomic analysis revealed 137 proteins in the vitelline membrane (Mann, 2008) and 119 yolk proteins in the yolk (Mann and Mann, 2008). Similarly, these proteomic or transcriptomic approaches have revealed more than 500 eggshell matrix proteins, including those present at very low amounts (Mann and Siedler 2006; Mann et al. 2007; Brionne et al. 2014; Marie et al. 2015a; Marie et al. 2015b) and 135 specific proteins in the isthmus, which secretes the eggshell membrane (Du et al., 2015). A very large number of proteins has been therefore identified in the egg compartment during the decade, including of course the major proteins that were previously identified by classic biochemistry. It is worth pointing out that some of these proteins are present at very low abundance and could derive from epithelium and glandular cells lining the oviduct. The identification of these proteins allows us to explore *in silico* their putative function using bioinformatics and to identify novel proteins and regulatory candidates involved in egg formation or molecules of interest as therapeutic molecules.

Eggshell formation

The egg possesses an efficient physical protection with it's calcified eggshell. As mentioned in the abstract, research in nutrition has led to some innovation for improving hen performance and egg quality by testing the effect on egg quality of novel rearing strategies or nutritional additives with a black box approach.

The acquisition of knowledge on the how the eggshell is elaborate, and which are the mechanisms and the molecular keys actors involved in it formation, appear to be thus powerful indicators to unlock this black box and contribute to enhanced quality of eggshell.

Secretion of ionic precursors

The eggshell results from a massive secretion of calcium and bicarbonate, which contributes to a linear deposition of CaCO3 at a rate of 0.33 g/h between 10 to 22 hours after ovulation of the yolk in the oviduct (Nys and Guyot, 2011). The main characteristics of Ca2+ secretion in the uterus are (i) the daily discontinuity of the ability of the uterus to transfer Ca2+ and HCO3-, (ii) its synchronization with the ovulation process during a particular period of the daily ovulatory cycle, and (iii) its additional stimulation by the presence of the egg in the dilated uterus (Nys et al., 1999; Bar, 2009). The transfer of calcium occurs mainly via the uterine glandular cells, as confirmed by the presence of calbindin 28 kDa (Wasserman et al., 1991)] and carbonic anhydrase (CA) in these cells. It also involves many transcellular transporters of other ionic species (Na+, K+, Cl-, H+), which participate in the process of calcium secretion and in the maintenance of cellular ionic homeostasis (Eastin et Spaziani, 1978; Jonchère et al., 2012;Brionne et al., 2014). Hen uterine glandular cells must transfer large amounts of calcium into the uterine lumen against the concentration gradient, while preserving a low level of intracellular calcium (<0.0002 mM). Ca2+ is not stored in the uterus before eggshell calcification, but comes from blood plasma. Bicarbonate is mainly derived from CO2 passive diffusion through cellular membranes. Trans-epithelial transfer of Ca2+ occurs in three steps, as observed in all transporting epithelia: (i) Ca2+ influx through a downhill gradient, (ii) intracellular Ca2+ transport involving calbindin 28kD protein (Wasserman et al, 1991) and storage of Ca^{++} in endoplasmic reticulum and (iii) active output into the lumen through Ca2+ pump extrusion. Transcriptomic studies comparing the uterus and other parts of the oviduct, or comparing gene expression in the uterus at a period with or without eggshell formation, revealed numerous candidate transporters (Jonchère et al., 2012; Brionne et al., 2014). Calcium secretion through epithelial glandular cells involves TRP Ca2+ channels (cell uptake / entry), CALB1 (intracellular transfer), endoplasmic Ca2+ pumps type 2 and 3 (ATP2A2 and 3, uptake by endoplasmic reticulum) and inositol trisphosphate receptors type 1, 2, and 3 (ITPR1, 2 and 3, output from the reticulum). Calcium is then extruded from the glandular cells by plasma membrane Ca2+ pumps (ATP2B1 and 2) and Ca2+/Na+ exchangers (SLC8A1 and 3). The low intracellular Ca2+ concentration, which is crucial for cell survival, is ensured by CALB1, endoplasmic Ca2+ pumps and ITPRs.

The bicarbonate precursor of eggshell calcite is mainly derived from blood carbon dioxide (CO2). It is also supplied at a low level from plasma by the Na+/HCO3- co-transporters (SLC4A4, A5 and A10). Carbonic anhydrase 2 (CA2) catalyzes the reversible hydration of intracellular CO2 to HCO3-, which is then secreted into the uterine fluid through the HCO3-/Cl- exchanger SLC26A9. Large currents of Na+, K+, and Cl- contribute to maintenance of physiological ionic concentrations in the cell. Na+ is absorbed from the uterine fluid by Na+/Ca2+ exchangers and by amiloride-sensitive Na+ channels at the apical membrane, and is extruded from the cell at the basolateral membrane against the electrochemical gradient by the Na+/K+-ATPase, which transports Na+ out and K+ into the cells (Jonchère et al., 2012; Brionne et al., 2014). In conclusion, identification of the ionic transporters in the uterus underlines some numerous similarities with those involved at the intestinal level, even if the uterus is secreting Ca^{++} in contrast to the intestine, which is absorbing this ion. However, regulation differs largely between these tissues, intestinal calcium absorption being directly under the control of vitamin D, in contrast to the uterus.

All calcium transporters are up-regulated during eggshell formation, with the exception of the TRPVs (Jonchère et al., 2012; Brionne et al., 2014). However, the intensity of differential expression varies from 16-fold for calbindin, to 2-3 fold for Ca2+/Na+

exchanger, endoplasmic reticulum pumps, PMCA, CA2, Na+/HCO3- exchangers, Na+/K+ exchanger and K+ voltage-gated channel. The presence of vitamin D response element (VDRE) and of the estrogen response elements (EREF) in the promoter region of some of these genes underlines the possibility of regulation by vitamin D and estrogen but deeper studies are obviously needed to confirm this possibility. Mechanical stretch of the uterine wall induced by the presence of an egg can also up-regulate ion transfer. As shown for the Na+/K+-ATPase (Lavelin et al., 2001). It is noteworthy that in some molluscan or echinoderm species, there is evidence that the formation of the initial $CaCO_3$ crystal occurs intracellularly as an amourphous calcium carbonate (ACC phase) within a membrane envelope (Wilt et al., 2008). In hens, there is no direct experimental evidence for the presence of $CaCO_3$ containing vesicles in uterine cells, but this hypothesis cannot be currently discarded.

Biomineralisation of the eggshell

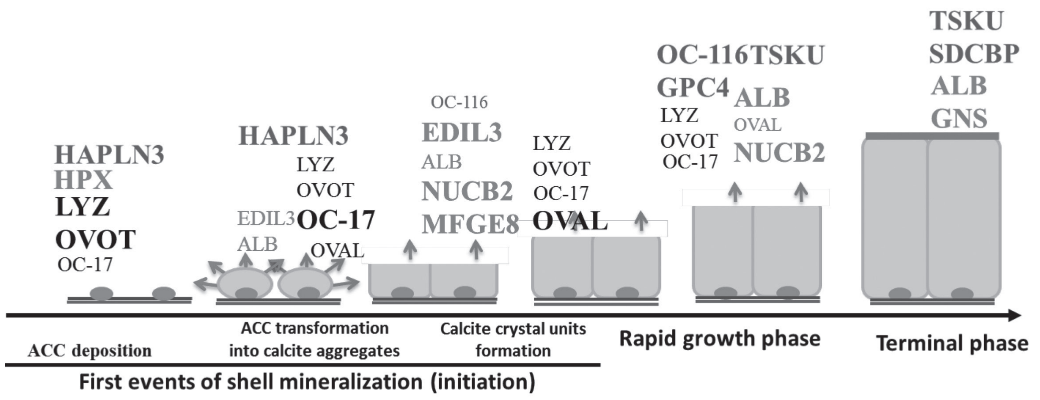
The avian eggshell is a porous bio-ceramic that is formed at body temperature in a cell-free environment. The avian eggshell forms in a confined space, the distal segment of the hen oviduct, in the uterine fluid that is supersaturated with respect to calcium and bicarbonate (about 70-fold relative to the solubility product of calcite) and contains the organic precursors of the shell matrix. Its distinctive features, as compared to bone or teeth, are the nature of the mineral deposit – CaCO3 in the form of calcite, as well as the absence of cell- directed assembly during its fabrication, which is initiated at organic cores present on the outer surface of the eggshell membranes (Nys et al. 1999, 2004; Hincke et al. 2012). The mineralized shell is anchored to nucleation sites, which are located on the surface of the outer shell membrane. The multi-directional growth of calcite crystals towards the outer surface leads to the formation of inverted cones that join to form a compact layer. Eggshell mineral initially originates from the accumulation of amorphous calcium carbonate (ACC) particles, which then dissolve and become concentrated at specific organic sites on the eggshell membrane fibers that are rich in proteins and sulfated proteoglycans. These components are thought to promote the nucleation and stabilization of ACC with calcite short-range order, which predetermines the calcite composition of the mature eggshell (Rodríguez-Navarro et al. 2015).After this initial event, radial growth of calcite crystal is progressively replaced by competitive growth between crystals arising from adjacent sites of nucleation (Garcia-Ruiz and Rodriguez-Navarro 1994). This high degree of control of size, shape and orientation of the calcite crystals in avian eggshells is responsible for its unique ultrastructure and exceptional mechanical properties. This high degree of textural organization is due on the one hand to competition for space between crystals growing from adjacent nucleation sites (Garcia-Ruiz and Rodriguez-Navarro 1994), and on the other hand, to the interaction of calcium carbonate with the organic matrix components during the nucleation and growth phases. Proteins and proteoglycans from the organic matrix interact with the initial amorphous calcium carbonate to form the calcite crystal, and then inhibit the growth of the faces parallel to the C axis of the crystal to elongate the calcite crystallites; together, these processes favor the growth of crystals that are roughly perpendicular to the egg surface (Garcia-Ruiz and Rodriguez-Navarro 1994; Nys et al. 1999).

The eggshell matrix composition is currently well characterized. It is firstly composed of ubiquitous components (clusterin, antimicrobial proteins, antiproteases), proteoglycans (keratan and dermatan sulfate glycoaminoglycans), proteins previously identified in the egg white (ovalbumin, ovotransferrin, lysozyme) and also of proteins identified in bone tissue (SPP1, OC 116). In addition, the shell matrix is constituted of proteins which are extremely specific to the distal oviduct of birds (isthmus and uterus) and to eggshell formation: in chicken, these have been characterized as ovocleidin-17, ovocalyxin -21, -25, -32, -36 (Nys et al. 1999; Nys et al. 2004; Hincke et al. 2012). Ovocleidin-17 is amongst the most abundant matrix protein and has been shown to modify the morphology of calcite crystal (Reyes-Grajeda, 2004). Modelling of ovocleidin-17 suggests the involvement of this protein in the crystallization of calcite from ACC nanoparticles (Freeman et al. 2010).
A recent quantitative proteomic analysis of the organic matrix proteins of the uterine fluid, where shell mineralization takes place, and of eggshell extracts, throughout the different steps of eggshell formation (ACC transformation in calcite, nucleation phase, rapid growth phase and arrest of shell formation), reveals the presence of more abundant proteins at particular phases of the process (OC 17, OC 116, EDIL3, LOXL2, ovalbumin, ovotransferrin, lysozyme, etc.…) and underlines the likely involvement of these proteins in the control of eggshell formation (Marie et al. 2015a; Marie et al. 2015b). Significant associations between gene polymorphism and eggshell quality indicate the influence of organic matrix proteins on eggshell fabric: ovalbumin gene polymorphism is associated with breaking strength, shell deformation, eggshell thickness and crystal size, OC116 with shell thickness, elastic modulus and crystal orientation, SPP1 with fracture toughness, ovotransferrin with crystal size and OCX-32 with shell thickness and crystal orientation (Dunn 2011; Takahashi et al. 2010; Sun et al. 2016).

Egg Antimicrobial proteins

Identification of novel molecules

More than 150 different proteins have been identified in the egg white (Mann, 2007); however, the ten most abundant proteins represent about 95% of the total proteins and the remaining 5% is composed of a large number of minor proteins. The main proteins involved in antimicrobial properties of egg are lysozyme, ovotransferrin, some protease inhibitors (ovomucoid, ovoinhibitor, ovostatin and cystatin) and some vitamin-binding protein (avidin, riboflavin-binding protein) (Guyot et al., 2017). Recently, additional proteins have been identified in the egg white and indeed are generally present in other compartments of the egg (eggshell, vitelline membrane). The avian beta-defensins are small (2-6 kDa) cysteine-rich cationic proteins that are components of the host innate



Major mineralogical and proteomics events involved in the eggshell calcification process.

defense system, as previously observed in invertebrates, plants and other vertebrates. These molecules possess in general a broad-spectrum activity directed against Gram-positive and Gram-negative bacteria, but also against fungi and viruses. AvBD11 is a longer beta-defensin (9.2 kDa) containing 6 disulfide bonds that is composed of two beta-defensin motifs. It exhibits antimicrobial activities against *Salmonella* Enteritidis, *Salmonella* Typhimurium, *Escherichia coli*, *Listeria monocytogenes* and *Staphylococcus aureus* (Herve-Grepinet et al. 2010). AvBD11 activity is inhibited by heparin suggesting that the heparin-binding site is involved in its mechanism of action (Guyot et al. 2016). Gallin is a cationic peptide containing 3 disulfide bonds. Its antibacterial spectrum is more restricted as the synthetic gallin is active only against *E. coli* but not against *Salmonella enterica, Staphylococcus aureus* and *Listeria monocytogenes* (Herve et al. 2014). Amongst other egg proteins, are Tenp and OVAX. Tenp (Transiently Expressed in Neural Precursor) is a protein belonging to the BPI/ LBP (Bactericidal Permeability Increasing protein / Lipopolysaccharide binding protein)/PLUNC (Palate, lung, and nasal epithelium clone protein) family and is able to bind bacterial lipopolysaccharide and mediate damages of the inner membrane of bacteria. Tenp isolated from Emu egg white exhibits antibacterial activities against some Gram-positive bacteria but not against the Gram-negative bacteria *Escherichia coli* and *Salmonella* Typhimurium (Maehashi et al. 2014). Ovalbumin related protein X belongs to the ov-serpin family (serine protease inhibitors), which includes ovalbumin. OVAX is 100-times less concentrated than ovalbumin in egg white and shares high sequence identity with ovalbumin. It is noteworthy that OVAX, in contrast to ovalbumin, exhibits antimicrobial activity against *Listeria monocytogenes* and *Salmonella* Enteritidis (Rehault-Godbert et al. 2013). Indeed, heparin inhibits its activity probably by blocking the heparin-binding site of OVAX and competing with the bacteria at the antibacterial site of OVAX. The use of heparin-affinity chromatography, to concentrate the antimicrobial activity of egg white have revealed antimicrobial proteins in addition to OVAX and AvBD11 in this fraction: VMO-1 (Vitelline Membrane Outer layer protein 1), pleiotrophin and beta-microseminoprotein-like (Guyot et al. 2017).

Regulation of antimicrobial activities by degree of hen microbial contamination

Numerous antibacterial compounds are present in the egg white and might interact synergically. In addition, physicochemical/environmental conditions (temperature, and egg white alkaline pH, viscosity, CO_2, ionic composition) are known to affect bacterial growth and egg white antimicrobial activity (Guyot et al. 2017). Genetics, age and immunological status of the hen, and egg storage conditions might affect the antimicrobial potential of the egg because of their well-known effect on egg white technological properties. Heritabilities for the antibacterial effects against Staphylococcus aureus (0.16) and Salmonella Enteritidis (0.11) are higher than those of antimicrobial proteins (Sellier et al. 2007), but remain too low, for practical applications in animal selection. We have explored whether stimulation of hen immunity can affect the innate protection of the egg as reflected in the yolk IgY antibodies. The systemic administration of lipopolysaccharide (bacterial endotoxin) to hens stimulates the antibacterial properties of egg white proteins against *Staphylococcus aureus* (Bedrani et al. 2013a) but the underlying mechanisms of this stimulation were not elucidated. Three extreme rearing conditions of hens (germ-free, specific pathogen free and conventional hens) with large difference in bacterial load were utilized to reveal any influence on egg white antibacterial properties (Bedrani et al. 2013b).

Antibacterial activities of egg white proteins against *Staphylococcus aureus* and *Streptococcus uberis* were increased in specific pathogen free and in conventional groups by comparison to the germ-free group, confirming that egg white antibacterial activities can be reinforced by microbial environment of hens but the magnitude of the effect remains limited. Lysozyme and antiprotease activities were not significantly altered by these rearing conditions.

Change in egg a**ntimicrobial activity during egg storage**

The effect of egg storage on egg white physico-chemical properties is well established but the consequences on egg white antimicrobial activity have not been thoroughly studied . During egg storage, antimicrobial proteins in egg white are challenged by the alkaline pH and storage conditions (time, temperature). A proteomic study conducted on eggs stored at three different storage temperatures (4°C, 20°C, 37°C) revealed the formation of protein complexes in egg white with increasing storage temperature (Qiu et al. 2012b). A decreased lysozyme activity by 4% to 17% has been observed in egg white following storage during 28 days at 15°C (Trziszka et al. 2004). Egg white antichymotrypsin and antipapain activities were not altered by storage time or temperature, but antitrypsin activity deteriorated after 2 weeks of storage at 37°C (Rehault-Godbert et al. 2010). No data are available for minor egg white antibacterial proteins. The growth of *Salmonella* is favoured at 20°C for eggs not older than one week, as compared to eggs stored at this temperature for 2-3 weeks (Duboccage et al. 2001). The comparisons of eggs stored for different times at variable temperature (Rehault-Godbert et al. 2010) reveals that egg white antibacterial activity is significantly increased within few days following oviposition. The activation was more rapid at higher temperature (between 0 and 5 days following oviposition for egg stored at 37°C) but the antimicrobial activity is then progressively altered after two weeks of storage. Therefore, some conditions of egg storage reinforce the antibacterial potential of the egg white against *Salmonella*. The molecular mechanisms for this effect remain unknown, although egg white thinning or an increase in pH might be involved.

Innovative therapeutic molecules

Some novel antimicrobial proteins might be used as new molecules for human heath either after being purified or concentrated in egg white fractions using affinity chromatography, however the low protein concentration and the animal origin of these proteins of interest might limit further developments. Alternatively, a synthetic peptide route can be adopted. Other approaches have been proposed using partial proteolysis of egg material.

Egg-derived peptides

Egg peptides (from egg white, egg yolk and more recently vitelline membranes) have long been recognized as potent bioactive molecules exhibiting antihypertensive, antioxidant, immunomodulatory, anti-inflammatory and antimicrobial activities (Mine et al, 2007; Liu L et al., 2017 ; Liu Y et al., 2017; Lee et al. 2017 ; Meram et al. 2017). Most of these active egg-derived peptides have been obtained *in vitro* after limited proteolysis of egg components or of purified proteins, by digestive enzymes including pepsin, trypsin and chymotrypsin proteases. These *in vitro* data suggest that such peptides could naturally occur in the gastrointestinal tract after egg consumption, thereby contributing to digestive health. To date, published studies refer to hydrolysates from raw egg white, egg yolk and vitelline membrane or are related to hydrolytic peptides from major egg proteins such as ovotransferrin (Liu L et al., 2017), lysozyme (Rao S et al. 2012) or phosvitin (Xu et al., 2007). However, there is currently a lack of information on less abundant proteins, which may also have a high potential for development and that likely exhibit a diversity of activities. Indeed, more recently, some have shown activities in endotoxin neutralization (Ma et al., 2012) or bone organogenesis (Liu et al., 2013, Shang N., 2018). The fact that the egg constituents support embryonic development suggests that we might also discover interesting activities related to the defense of the embryo and regulation of its growth, by investigating the dynamics and the generation of natural egg-derived peptides during incubation. Actually, although many efforts have been focused on proteomic approaches of egg components, there is currently a lack of information regarding egg peptidomes in fertilized eggs during embryonic development. Such data combined with the development of assays for screening many distinct activities and by using bioinformatic tools in parallel to predict biological activities (Basilicata et al., 2018) could drastically increase the development of egg-derived hydrolysates (either purified from egg, generated by *in vitro* proteolysis or obtained by chemical synthesis) of increased values for health industries.

Conclusions

The identification of novel egg and oviduct proteins has allowed the mechanisms of egg formation to be to deciphered, and novel molecules to be to developed, which can be used for improving human health. High throughput technology was initially mainly qualitative and descriptive but proteomics is now more quantitative allowing exploration at a large scale of the proteins providing the precursors of the egg involved in its formation (eggshell) and controlling their properties (all compartments). This quantification can be used to explore the regulation of the synthesis of the active proteins in the process and possibly to reinforce some properties of the egg compartment by exploring how genetic or physiology of hens can explain the change in egg properties. That might be used to explore how some nutritional additives might reinforce egg quality. This information on proteins (polymorphisms) is currently in use to improve the selection of hens and should improve its efficiency. There is clearly a high potential for this technology to permit better control egg quality but also improvements in other sectors such as pharmaceutical industry.

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Preincubation and incubation conditions, hatching time and broiler growth

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Summary: It is well known that chick viability and broiler growth are important factors determining the profitability of broiler industry. In order to meet the expectation of broiler sector, the production of high-quality day-old chicks is fundamental. Pre-incubation factors such as breeder age, egg weight, duration of egg storage may determine hatching egg characteristics. Several studies have shown that breeder age affects egg weight that may affect broiler performance. Longer egg storage duration (even after eight days of storage) can reduce hatchability, increase embryonic mortality, and depress broiler growth. The quality of day-old chicks and broiler performance are highly affected by incubation temperature, humidity, turning, lighting and spread of hatch (hatching time). Optimal incubation temperature for broilers is 37-38 °C. Relatively small deviations in incubation temperature result in embryo growth retardation and decrease hatchability. On the other hand, short time cyclic or constant high or low incubation temperature may alter the thermotolerance of broilers. High and low incubation temperatures have also been shown to alter bone and muscle development. Broiler breeder eggs are incubated in complete darkness. Studies showed that lighting during incubation could increase embryonic growth and hatchability and reduce stress susceptibility and fear response. Different wavelengths of light can affect embryo growth differently. The first hours after hatching are considered as another critical factor for broiler growth and performance. Chicks do not hatch exactly at the same time; hatching occurs over a 24 to 48 h window. The chicks that hatch early remain for several hours without food and water, which impairs intestine and muscle development and weight gain. All these factors during pre-incubation and incubation stages influence each other and determine the overall broiler performance. Today, there is an increasing evidence that incubation environment does not only determine hatching success but also affects the potential performance of broiler. The purpose of the presentation is to discuss the most relevant pre-incubation and incubation factors that affect embryo development and broiler performance.

Keywords: *Breeder age, egg storage, incubation, embryo, chick, growth*

Introduction

The quality of day-old chick is an important starting point for broiler industry. Day-old chick quality is related to preincubation and incubation conditions Management practices from the broiler breeder farm to the hatchery have an impact on hatching performance, day old chick quality and broiler performance. Among many other factors, breeder age and feeding determine hatching egg characteristics and broiler growth. Breeder flock age and egg size (weight) always have been linked together. Preincubation egg storage is a common practice in commercial hatcheries and provides flexibility to meet meat type chick's demand of the industry. However, it is known that the length of the storage period can influence hatching success. Incubation conditions have an unquestionable effect on broiler growth however; these conditions have interacted with preincubation conditions. Environmental factors during incubation such as temperature, lighting, turning of eggs, the gaseous environment of the embryo are of critical importance for embryonic and posthatch growth. Incubation conditions experienced by birds can have long-lasting effects on growth and well-being. During critical stages of incubation, environmental factors have the potential to affect birds' ability to adapt to the environment. Hatching time (late vs. early hatched) can influence the homogeneity of day-old chicks, thus affects the growth rate of broilers. This presentation aims to discuss the most relevant preincubation and incubation conditions that affect embryo development and broiler growth.

Broiler breeder age and feeding

Egg provides essential nutrients for the growing embryo. Egg yolk, which contains 99 % of the lipids and 47 % of the protein present in the egg, is the main energy supply for the growing embryo. It is known that egg weight and yolk percentage increases with the breeder age increase, whereas heavier eggs at any given age have relatively lower egg yolk than lighter eggs (Vieira and Moran 1998). It must be noted that egg size has a greater effect on chick weight than yolk size (Wolanski *et al.*, 2007). Embryonic growth is related to uptake and utilization of egg nutrients. Peebles *et al.* (2001) noted that residual yolk sac uptake was faster in embryos from hens at 36 wk compared with those at 27 wk. O'Sullivan *et al.* (1991) and Nangsuay *et al.* (2011) reported that the eggs from old breeders had the higher availability of resources, thus the chicks from old-breeders had higher nutrient assimilation to yolk-free body weight than chicks from young breeders. A better nutrient assimilation may results in more available energy for growth (Nangsuay *et al.*, 2011), in turn, potentially affect broiler performance. Therefore, breeder age effect was more pronounced during the first weeks of laying period. The breeder age has been reported to influence the development of intestine during embryonic growth. Chicks hatched from old breeders had significantly longer and wider villus, larger villus area, and higher goblet cell number in jejunum at d of hatch (Yalcin *et al.*, 2013). This result indicates that intestine of chicks from older breeders are morphologically more developed than the chicks from younger breeders, therefore these chicks have an ability to better adapt to posthatch feeding. Indeed, broilers from old breeders were heavier than those from young breeders at d of hatch and this trend continued throughout the posthatch period. Breeder age had a significant effect on the feed consumption from 0 to 21 d but did not have an effect from 22 to slaughter age (Ulmer-Franco *et al.*, 2010; El Sabry *et al.*, 2013). Researchers have increasingly focused on the maternal nutrition that can potentially affect embryo and posthatch development. Among different nutrients in the maternal diet, vitamins and natural antioxidants have been suggested to play a significant role. A clear effect of the maternal dietary vitamin D3 on the progeny performance and particularly on bone characteristic was highlighted by Atencio *et al.* (2005). The antioxidant composition of breeder diet can serve as a major adaptive mechanism for protection of tissue during the oxidative stress experienced at hatching and early posthatch period (Surai, 2000). The antioxidant (vitamin E, selenium, canthaxanthin, caretonoids) enriched maternal diets influenced antioxidant enzyme activities and decreased lipid peroxidant reaction in chicks (Karadas *et al.*, 2005; Zhang *et al.*, 2011). The results suggest that the effect of maternal nutrition on progeny is more pronounced under conditions of poor flock uniformity and stress.

Preincubation egg storage

Ideally, eggs should be incubated 1-2 day(s) after they are laid. This allows CO_2 and H_2O to be released from the egg and albumen pH increases from 7.6 to 9.0. This optimizes early embryo development because optimum pH for embryonic development is between 7.9 and 8.4 (Reijnick, 2008). However, this is rarely practical and due to market demand for the one-day-old chick, usually, the egg storage duration varies from a few days to 14 days. The negative effects of prolonged egg storage on hatchability and chick quality are well documented in the literature (Reijnick *et al.*, 2008, Tona *et al.*, 2004). The prolonged of egg storage prior to incubation influence embryo development, length of the incubation period (Lapao *et al.*, 1999; Christensen et al., 2002) and hatchability with the greatest effects for eggs stored longer than 8 d (Yalcin and Siegel, 2003). Prolonged egg storage duration reduces embryo viability and chick quality and these effects are more pronounced in eggs from old breeders (Lapoa *et al.*, 1999; Tona *et al.*, 2004). However, storage effect on posthatch growth was more obvious in younger than in older breeders (Tona et al., 2004). A trend for reduced lung (Yalcin and Siegel, 2003), heart weights (Christensen *et al.*, 2002), villus width and surface area and goblet cell number (Yalcin et al., 2016) in embryos from eggs stored for 14 d were reported. Moreover, expression of H+-dependent peptide transporter (PepT1), which plays an important role in the uptake of amino acids is, lower in chicks from stored 14 days before incubation than those from eggs stored 3d. Therefore lowered villus surface area along with downregulated expression of PepT1 in chicks from eggs stored 14 d could lead to depressed growth (Yalcin *et al.*, 2016). Alleviating the negative effects of long storage, Fasenko *et al.* (2001) reported that prestorage incubation of 6 h at 37.5°C could be used to improve hatchability of eggs stored for 14 d. Dymond *et al.* (2013) reported that 4-h of preincubation at 37.5°C at 4 – to 5-d intervals during egg storage could be used. They showed that short-term preincubation is an effective method to increase embryo survival, improve hatchability and chick performance.

Incubation temperature

In poikilotherm embryos, the incubation temperature is one of the major environmental factors that affect development. It is important to incubate eggs at optimum temperatures, which range from 37 to 37.8°C (Decuypere and Michels, 1992). The organs and physiological systems of embryo begin to develop during the first week of embryonic stage and continue posthatch. The deviations from optimum temperature may affect embryo size, organ, bone and muscle development, intestinal maturation (Yalcin and Siegel, 2003; Molenaar *et al.*, 2011; Wineland *et al.*, 2006). Previous studies showed that the first week of embryo development is utmost importance for hatchability, chick weight, and broiler performance. Low incubation temperature (eggshell temperature) from day 0 to 10 of incubation reduced hatchability and body weight gain of broilers from 0 to 21 and 21 to 42 d (Joseph *et al.*, 2006). Incubation temperatures during the first week of embryo development also affect bone development. Low (36.9°C) or high (39°C) incubation temperatures from 0 to 8 d of incubation increase the subsequent incidence of tibial dyschondroplasia, which is probably the result of delayed Hsp-90 driven chondrocyte differentiation (Yalçın *et al.*, 2007). High incubation temperatures (39.7°C) from d 16 to hatch resulted heavier chicks at hatch compared with control incubation, however, these broilers had significantly lower body weight during the posthatch (Hulet *et al.*, 2007). The incubation temperature has a particular importance in thermoregulation of poultry. Evidence from numerous studies demonstrated that the development of thermoregulation system extends from embryonic period through early prenatal. The environment experienced by the embryo during critical periods, which are defined as developmental periods when an organism is particularly susceptible to internal or external stressors, can determine the "set point" of thermoregulation system for the posthatch. The efficient timing to alter the "set point" of thermoregulation could be linked to development or maturation of the hypothalamus-hypophysis-thyroid axis. Several studies have shown that a 1–2°C cyclic increase in the optimum incubation temperature from day 10 to 18 or day 16 to 18 of incubation can induce thermotolerance without impairing the development of embryos or subsequent growth (Yahav *et al.*, 2004; Collin *et al.*, 2005; Yalcin *et al.*, 2008a,b; Piestun *et al.*, 2008). Acquisition of thermotolerance is reflected by better body weight gain, reduction in plasma thyroid hormone concentration and lower body temperature than untreated group when exposed to heat stress conditions (Yalcin *et al.*, 2008b). Similarly, cyclic 1°C reductions of the incubation temperature improve posthatch cold tolerance and contribute to the decline in ascites syndrome in broilers under cold environmental stress (Shinder *et al.*, 2009; Yalcin *et al.*, 2012; Akşit *et al.*, 2012). Loyau *et al.* (2014) also showed that cold incubation induced long-term effects on some antioxidant pathways and genes involved in energy metabolism in broilers. This could enhance the welfare and thermotolerance of broilers when exposed to cold conditions.Manipulation in the incubation temperature during critical periods of muscle development may also have a stimulatory effect on muscle growth. Higher incubation temperatures (39.5°C for 3 or 6 h/d) between 16 and 18 d of incubation has been shown to have a positive effect on breast muscle weight until slaughter age (Piestun *et al.*, 2011). Recently, it was reported that 39.5°C for 13 h/day from 14 to 18 d of incubation reduced the number of broilers with moderate to severe myopathic attributes compared to control (Clark *et al.*, 2017).

Oxygen and carbon dioxide concentration during incubation

The oxygen (O_2) and carbon dioxide (CO_2) concentration is controlled through ventilation during incubation. Eggs are normally incubated in a standard gaseous environment of 21% O_2 and 0.1-0.5% CO_2. However, studies showed that higher CO_2 concentration enhanced embryo growth and stimulated hatching process, depending on the level of CO_2 and timing of high CO_2 exposure. De Smit et al. (2008) showed that increasing incubator CO_2 concentration up to 0.7 % by nonventilation during the first 10 d of incubation had no effect on hatchability, chick weight, but decreased incubation duration and shortened the interval between internal piping and hatch. Everaert et al. (2007) reported that chicken embryos can tolerate 4% concentrations of CO_2 between 10 to 18 d of incubation with no effect on embryonic growth, hatchability, and relative growth up to 7 d. Hypoxic conditions during the first week of incubation stimulate blood vessel development. The study by Druyan *et al.* (2012) investigated the effect of 17 % O_2 concentration for 12 h/d from 5 to 12 d of incubation on chorioallantoic membrane (CAM) development and cardiovascular parameters. The 12 h of hypoxic condition resulted in a significant increase in the vascular area of the CAM and subsequently (on 13 and 14 d of incubation) increase in their blood oxygen-carrying capacity and oxygen consumption and T_4 concentration. Broilers grew better than control until slaughter age and coped better with severe environmental conditions (Druyan et al., 2018). Similarly, between 16 and 18 d of incubation, 17 % O_2 for 12 h/d led to moderate changes to give the embryo the ability to cope with stress, while continuous 17 % O_2 significantly decreased in hatch weight (Haron *et al.*, 2016).

Lighting during incubation

Numerous studies have shown that exposing eggs to light during incubation may improve embryonic growth and chick quality. The color, source, and level of the light are important factors affecting the results. Huth and Archer (2015) reported that 12 h of light/d at 250 lux at egg level during incubation could improve chick quality and later reduce the stress susceptibility of broilers compared with continuous dark incubation conditions. Lighted incubation either 16 h of light and 8 h of dark (Özkan *et al.,* 2012a,b) or 12 h of light and 12 h of dark (Archer and Mench, 2014) establishes circadian rhythms of melatonin, which is synthesized mainly in pineal gland and eyes during the dark period of day (Zeman et al., 1992). The decrease in stress susceptibility in chicks from lighted incubation may be due to melatonin rhythms, which could alter the HPA axis (Özkan *et al.*, 2012a,b) or asymmetric development of brain structures, which may affect responses of birds to environmental stressor due to changes in visual perception of fearful stimuli (Rogers, 1995). The melatonin rhythm induced during incubation by lighting may have long-lasting effects on the rhythmicity of some behaviors via epigenetic effects (Archer and Mench, 2014). Lighting during incubation also affects muscle development and incidence of leg problems. Cool white fluorescent lighting from d14 to 21 of incubation (Özkan *et al.*, 2012a) and intermittent (15 min light:15 min dark) green LED lighting (0.1 W/m^2 at eggshell level) from d 5 to 21 (Rozenboim *et al.*, 2004) increased breast muscle weight of chicks at hatching.

The spread of hatch (hatching time) and early nutrition

Hatching duration is influenced by breeder age, egg storage duration, and incubation conditions. Even if all these factors are standardized, there is still a spread of the hatch between 24 and 48 h (Careghi *et al.*, 2005; van de Ven *et al.*, 2011). Because chicks are held in the incubator until most of the chicks have already hatched, the time from the first hatch to feed access is longer for early hatched chicks compared to late hatched. Therefore, early hatched chicks can lose about 4-10% of their body weight due to the hatch window duration (Careghi *et al.,* 2005; Yalcin *et al.,* 2013). The loss in body weight was found as 0.092 g/h during the first 6 h after hatch, then chicks lost 0.113 g/h and 0.132 g/h between 9 and 15 h and 15 and 21 h, whereas yolk-free body weight was not affected. This indicates that a longer hatch window would increase body weight losses and decreases the hatch weight at the pullout, due to reduced yolk sac weight (Yalcin *et al.,* 2013). The decrease in yolk sac weight is explained by nutrient transfer from the yolk sac into the intestine, which helps the early growth of small intestine (Uni *et al.*, 1998; Noy and Sklan, 2001; Yadgari *et al.,* 2011). Indeed, the increase in the weights of the duodenum, jejunum and, ileum was found as 64.7, 46.5, 47.8 %, respectively, during the hatch window (Yalcin *et al.*, 2016). Moreover, hatch time has an impact on muscle development in broilers, with accelerated muscle growth in early hatched and midterm hatched compared to late hatched chicks (Powel et al., 2016). van de Ven *et al.* (2013) and Lamot *et al.* (2014) suggested that early hatched chicks had a different growth and metabolic pattern compared with midterm and late hatched chicks within a hatch window. Higher plasma glucose level was found in late hatched versus in the early hatched chicks, probably due to lower metabolism, which was associated with lower T_3 levels and decreased growth rate (Careghi *et al.*, 2005; Tona *et al.*, 2005; van de Ven *et al.,* 2011). Hatching time also affects adipose tissue deposition. Recent studies have shown a trend for more adipose tissue deposition in late hatched compared to early hatched broilers at 40 d (Powell *et al.*, 2016). Breeder age is one of the factors affecting the spread of hatch. Hatching distribution of chicks from old breeders is concentrated in the late periods of incubation (495-500 h of incubation) while during the early hours of hatching process (480-485 h of incubation), more chicks are hatched from young breeders (El-Sabry *et al.*, 2013). Tona *et al.* (2003) reported no significant effect of egg storage duration on the spread of hatch but about 80% of the chicks from eggs stored for 3 d hatched before 490 h of incubation, whereas such percentage was obtained after 500 h of incubation for eggs stored for 18 d. Early access to feed after hatch positively affects growth and development during the early postnatal stage (Lamot *et al.*, 2014). The relative organ growth is delayed in chicks withheld from feed and water for 32 h after hatch compared with those with the direct feed and water access from hatch onward (van de Ven *et al.*, 2013). Longer duration of feed deprivation after the hatch lowers intestinal weight and enzymatic activity, and villus development (Geyra *et al.,* 2001; Maiorka *et al.*, 2003).

Conclusion

A wide range of factors influences the structural and functional development of the chicken embryo.Embryo development highly depends on maternal effects and storage duration of the egg before incubation.Changing in the environmental conditions of a developing organism might change the developmental trajectories during prenatal and postnatal growth.Cyclic changes in incubation temperature, ligh:dark rhythm, and gaseous environment have a potential to reduce stress response of broilers under suboptimal environmental conditions.

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Modern Myopathies in Young Broilers

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Summary: Incidence of myopathies in broiler breast meat have been on the rise over the past 5-10 years. These include the so-called Wooden Breast Syndrome (WB), White Striation (WS), and "spaghetti meat". These myopathies cause loss of myofibers and fibrosis, leading to poor meat processibility, economic losses at the processing plant, and negative customer perception. The prevalence of these myopathies has increased in many countries, with WB prevalence reaching 30% in heavy birds in the US, leading to an estimated $200M loss per year. Currently, identification of WB and WS mainly relies on physical and visual inspection performed by plant workers. Given the speed of meat processing at the plant, inspections are necessarily time consuming and might result in decrease sensitivity in detecting of these myopathies. This presentation will review our current understanding of the WB and WS problems and industry efforts to reduce the incidences at the farm, detect them at the plant, as well as find ways to utilize some of the fillets affected by such problems.

Keywords: *broilers, myopathy, wooden breast, white striation*

Literature Review

Over the past decade, a number of reports have indicated a significant rise in the incidence of myopathies in young broilers. Two of the most prevalent and economically relevant myopathies are the so-called Wooden Breast Syndrome (WB) and White Striation (WS). The WB is a relatively new occurrence that has not been as well characterized as the WS which has been reported since 2010. In general, the WB consists of multifocal loss of myofibres associated with markedly increased amount of fibrous tissue, mainly affecting the breast fillets of young broiler chickens (35-49 days old). This accumulation of fibrous tissue affects the tactile characteristics of the raw breast fillet, making it firmer upon palpation compared to normal (Mudalal *et al.*, 2015). In some cases, a clear viscous fluid and/or petechial multifocal lesions on the fillet surface are also observed. On the other hand, WS is characterized by white stripes on the surface of chicken breast fillets, although it can be seen also on some thigh muscles. These striations typically run parallel to the muscle fibre direction, and are microscopically characterized by acute myodegeneration and necrosis. Although described separately, these myopathies have overlapping histological lesions, where necrosis, myodegeneration, inflammation, fibrosis, accumulation of adipose tissue, and regenerative changes are variably described (Kuttappan *et al.*, 2016). Although WB shows the highest level of fibrosis and chronic lipidosis compared to WS, it is not clear whether the two myopathies share a common etiology and if they might represent the two extremes of a single disease spectrum (de Brot *et al.*, 2016; Velleman, 2015). The exact etiology and pathogenetic mechanisms behind the occurrence of WB and WS remain unknown, however a few hypotheses have been proposed. A recent study with RNA-seq analysis suggested that localized hypoxia, oxidative stress, higher levels of intracellular calcium, and muscle fiber type switching associated with modern fast-growing broilers could be associated with occurrence of these myopathies (Mutryn *et al.*, 2015). Various studies reported that fillets with higher degrees of WS were heavier and thicker than normal fillets (Kuttappan *et al.*, 2016). Moreover, feeding birds with high energy and protein diets, age, gender, feed restriction, and genetics had all been associated with increased severity of WS (Barbut, 2015). However, many of these variables could have slaughter body weight as an underlying factor (Kuttappan *et al.*, 2016). Overall, it remains unclear how much genetics, management, tissue microanatomy (microvascular density), and nutrition might influence the development of both WB and WS (Kuttappan *et al.*, 2016; Soglia *et al.*, 2018). Similarly, the timeframe for the development of WB and WS remains uncertain: although initially regarded as a disease of broilers close to processing age, recent data suggest that WB lesions might be observed as early as one week of age (Anonymous, 2017). Industry concerns are related to poor processibility of the meat affected by the WB and WS conditions, economic losses at the plant, and negative customer perception (e.g., necrotic tissue in a chicken breast fillet is unappealing and consumers associate these myopathies with injured/old bird). Both syndromes also negatively affect protein functionality (e.g., reduced water binding) in processed products (Barbut, 2015). The prevalence of these myopathies has been increasing in many countries, and WB prevalence has been recorder to be as high as 27-32% (Russo *et al.*, 2015; Tijare *et al.*, 2016; Owens, 2015). Owens (2015) suggested that the incidence of severe WS has increased from 3-19% in 2009 to 15-27% in 2015. An article published in The Wall Street Journal reported that the estimated prevalence of WB in breast meat produced in the broiler industry could range from 5 to 10% (Gee, 2016). This would translate, in a conservative estimate, to $200 million losses annually for the US poultry industry alone (Gee, 2016). Other countries are also struggling with the problem and various numbers have been published. In any case, systematic studies regarding the prevalence of WB and WS around the world show that these are global problems.

In order to control and limit the possible economic impact of WB and WS, both basic and applied research are needed. Basic research is necessary for an in-depth understanding of the development of these diseases, in order to eliminate possible genetic and environmental causes. Applied research is needed to understand disease prevalence, develop diagnostic tools to accurately identify the problem before slaughter, and to implement a systematic scoring system for both syndromes. To date, some equipment/sensor manufacturers are working on finding effective methods for detection of breast myopathies (in particular WB and WS) and implement them in fast moving processing lines. Currently, identification of WB and WS mostly relies on visual and physical (palpation) inspection performed by plant workers. Given the speed of a modern meat processing broiler line (10,000 – 15,000 birds per hour), visual and physical inspections are time consuming, impractical, and might result in decrease sensitivity in detecting WB or WS lesions. A system to detect these myopathies using infrared light spectroscopy has been suggested (Wold *et al.*, 2017) and tried in a few plants, but is still considered expensive to install and not as effective as visual inspection and palpation. Therefore, gaining better understanding of these two myopathies including ways to reduce/eliminate their occurrence (e.g., via genetic improvements, modifying feeding/husbandry practices) is very important to the industry. In addition, developing and validating new in-line automated detection methods is of great importance to the meat processing industry.

This presentation will review our current understanding of the WB and WS problems and industry efforts to reduce the incidences at the farm, detect them at the plant, as well as find ways to utilize some of the fillets affected by these problems.

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Alternative protein sources for poultry nutrition

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Summary: The supply of protein and essential amino acids will undoubtedly be a great challenge for European poultry nutritionists when the transition to 100% organic feed ingredients for organic poultry production becomes reality. To avoid costly and energy consuming oversea transports connected to the import of soya beans, much research effort has focussed on the exploitation of alternative locally grown protein sources to increase the sustainability of organic production systems. Besides the cultivation of protein crops like legumes (peas, lupin, and faba been) and seeds (sunflower, rapeseed), new approaches including the feeding of protein concentrates from green biomass obtained by bio-refinement are discussed. At present, many of the alternative protein sources require a high degree of energy demanding processing and refining, i.e. drying and extraction in order to increase protein quality and quantity. These processes have to be optimised with respect to their efficiency in order to make local protein sources a competitive alternative to soya beans and fishmeal.

Keywords: *protein sources, local, amino acids, organic, technology, poultry*

Introduction

To strengthen the sustainability of organic poultry production in Europe, increased use of locally grown resources is required to avoid the import of organic protein feed from non-European countries (EU, 2007; EU 2012) involving energy demanding transports. Currently, at least 20% of the raw material used in organic feed production must be produced primarily on-farm or in the same region in cooperation with other organic crop producers or feed manufacturers (Magdelaine et al., 2010; EU, 2012). Because of a lack of organic protein sources, the transition to 100% organic feed ingredients for organic livestock has been postponed repeatedly in the EU, but is expected to take effect from January 1. 2019 (EU, 2017).

In organic animal production, the use of industrially produced protein feed based on oil seeds, i.e. soya bean meal is not permitted due to the use of chemical solvents for oil extraction. The same holds true for the use of synthetic amino acids, which makes the formulation of well-balanced poultry feed difficult. In layers, any imbalance in the amino acid supply does not only impair production results, but may cause stress to the hens thus provoking unwanted behaviour like feather pecking (Ambrosen and Petersen, 1997). Further, a lack of essential amino acids, especially methionine, reduces egg production and egg quality (Hammershøj and Steenfeldt, 2005). The requirement of these essential amino acids is currently covered by a surplus of dietary protein, which involves high nitrogen excretion, poor production results and a high risk of intestinal disorders.

In Europe, the feed for organic poultry production is currently based on grains such as wheat, barley, maize and oat as well as protein crops such as soya beans, soya bean cake, sunflower cake, rapeseed, rapeseed cake, lupin, pea and faba bean. Further, organic poultry diets often contain fishmeal contributing with valuable sulphur containing amino acids. However, seen in light of global overfishing, it may be questioned whether fishmeal may be considered as sustainable protein source. The composition of the feed depends to a high degree on the availability and price of raw materials (Sundrum et al., 2005). Although the European production of soya beans per year has more than doubled during recent years (1.2 to 2.5 million tonnes from 2013-2017, https://www.indexmundi.com/), organic livestock production still depends to a high degree on the import of soya beans from oversea countries, i.e. USA, Brazil, Argentina and China.

Currently, in many European countries, significant research efforts focus on the use of alternative sources of available protein that comply with the expected transition to 100% organic feed ingredients for organic animal production. As consumer demand for organic products is rapidly increasing, there is an additional pressure for alternative protein sources. The aim of this paper is to provide an overview of accessible protein sources that could be considered in this relation. The content of protein and selected amino acids of the protein sources discussed in the present paper is summarised in Table 1.

Vegetable sources

Legumes and seeds

Due to their ability of nitrogen fixation providing nitrogen for the following crop, legumes have a great advantage particularly in organic farming. Soya beans fixate nitrogen and it is indisputable that soya beans have a very good amino acid profile for poultry. Therefore, a large local production of more climate robust soya beans in Europe can be an important supplement to other home-grown legumes with a lower content of sulphur containing amino acids. In Denmark, cultivation experiments with soya beans (variety: Merlin) have been carried out over several years. The protein content (> 45% DM) and the amino acid profile of this variety showed high quality (Afrose, 2015; Steenfeldt & Hammershøj, 2015) and was even superior to that of soya beans grown in the largest producing countries like the USA, Brazil and China (40-43% DM), (Grieshop & Fahey, 2001). However, the harvest yield was relatively low and varied substantially from year to year. New varieties and the development of an optimal cultivation management are needed to achieve a higher and more stable yield, before locally grown soya beans can contribute to the self-sufficiency of protein. However, soya beans may have a potential as European protein source in the longer term (Van der Poel et al., 2013).

Faba beans, peas and lupins grow well in temperate climate. The average protein content is approx. 23% dry matter (DM) in peas and 30% DM in faba beans, but compared to soya beans, the content of methionine and cysteine is much lower in both legumes (Sauvant et al. 2004; Masey O'Neill et al., 2012). In experiments with broilers, it has been shown that the extrusion of peas improves DM, crude protein and amino acid digestibility and reduces their contents of phytic P, trypsin inhibitor activity and amount of resistant starch resulting in improved nutrient digestibility and AME_N values (Hejdysz et al., 2016).

In general, lupin has a higher protein content than peas and faba beans (30-45% DM), but the content of the essential amino acids methionine and cysteine is very low (Petterson, 2000). The starch content is likewise low in lupins, and the NSP content is almost twice as high as in other protein rich plants, which may limit the amount of lupin that can be used in feed poultry. In line with this, the results of Kubiś et al. (2018) showed that the graded inclusion of white lupin meal into layer diets (up to 300 g/kg) resulted in depressed performance, AMEN and eggshell quality. Likewise, dietary inclusion of 25% yellow lupin seeds significantly decreased the mean laying rate and egg weight (Rutkowski et al., 2017).

Rapeseed and rapeseed expellers are of interest due to high levels of sulphur containing amino acids (Sauvant et al., 2004). Both ingredients can be used in organic feed formulations to a certain extent taking the high oil content in whole seeds into consideration. However, the cultivation of rape requires a considerable nitrogen supply to the soil compared to legumes.

Sunflower has a high fat content (~50% DM), while the protein content is approximately 17-18 % DM (WPSA, 1992; Sauvant, 2004; CVB, 2009). After mechanical removal of the oil, the protein content in sunflower expellers increases up to 25-38% DM, depending on the extent of de-hulling (WPSA, 1992; CVB; 2009). Expelled sunflower has a reasonable high content of the sulphur-containing amino acids and is considered a good protein source for poultry (Van Krimpen et al., 2016).

Other potential raw materials of interest in organic farming systems are quinoa, hemp and sainfoin. The protein content of quinoa seed is 14-16% DM, and the methionine content is 2-3g/kg DM, which is higher than in most other grain products (Ruales & Nair, 1992; Steenfeldt and Hammershøj, 2015; Nowak et al. 2016). Sainfoin is mainly used as roughage for ruminants and horses, but these protein rich seeds can be seen as a potential raw material for poultry. Baldinger et al. (2014) found a concentration of 30% protein in whole sainfoin seeds. This percentage increased to 42% after de-hulling. The methionine and cysteine content was close to 10 g/kg (whole seed). The protein and amino acid content of hemp seed (Cannabis genus) is also high, and the methionine content can be compared to that of soya beans (Callaway, 2004). Currently limitations for the use of these raw materials are quite ineffective harvest methods, which have to be optimised through development of better technical equipment.

Dried distillers grains with solubles (DDGS)

DDGS is a cereal by-product of distillation for bio-energy, ethanol production with an average protein content of approximately 31% depending on the grain source (Pottgüter, 2015). Generally, maize DDGS has a significantly lower protein content (27-30%) than wheat DDGS ~36%). However, the protein content varies from facility to facility and even from batch to batch and requires a nutrient analysis before diet formulation. In their review on the use of maize DDGS as ingredient in poultry diets, Salim et al., (2010) suggest that high quality maize DDGS can be fed to broilers and layers without adverse effects on growth and performance. Further, DDGS appears to have some positive environmental effects including a reduction of ammonia emission from manure and an improved utilization of organic phosphorus. However, experiments with layers showed that the substitution of 0, 25%, 50% or 75% soybean meal with DDGS decreased egg production as dietary inclusion of DDGS increased (Ghazalah et al., 2011). For organic poultry production, the use of DDGS from bio-ethanol seems not feasible, but the inclusion of DDGS from local organic distilleries and breweries could be considered. However, the production of DDGS involves a costly and energy demanding drying process, which is a significant disadvantage of this protein source.

Protein from green biomass

In Europe, there is an increasing interest for the production of protein from green biomass via bio-refinement. Important research in that field has been carried out in Austria, Denmark, Germany, Ireland and the Netherland (Mandl, 2010), which is expected to result in the establishment of several bio-refinery plants in the coming years. Concentrating the protein fraction from green biomass such as e.g. red clover, clover grass, alfalfa, oilseed radish, using a bio-refining method, makes it possible to obtain a vegetable protein and amino acid source that can optimize feed formulation of organic poultry diets. Introduction of a new protein source with higher content of essential amino acids for poultry will increase the sustainability of organic poultry production using local grown plant material, while decreasing the strong dependency for soya bean import in Europe. Recent research strongly indicates that the content of protein and the important sulphur containing amino acids in green crop protein have a quality comparable with soya bean (Santamaria-Fernadez et al., 2017). The principles of green bio-refining is that fresh harvest of green biomass first is mechanically treated in a screw press resulting in two fractions, a green juice and a press cake. The press cake can be used for cattle feed or as material for biogas production, whereas the protein in the green juice can be precipitated by different methods (fermentation, heat or acid treatment). After centrifugation, the green juice is separated in a protein concentrate and a brown juice, where the dried protein concentrate can be used in feed for monogastric. The protein content in the concentrate can vary to a high extent depending on the extraction procedure, plant species and development stage of the plants. In a recent study by Santamaría-Fernández et al. (2017), the protein content in red clover, clover grass, alfalfa and oilseed radish was in the range between 38 and 48% DM, and the methionine content was in the range of 7.8-9.1g/kg DM, where a novel refining technique using lactic acid fermentation was used to extract the protein.

Very few studies have been conducted to evaluate the effect of green protein concentrate on egg production and growth performance of poultry. However, Ameenuddin et al. (1983) studied the effect of diets containing red clover protein concentrate for broilers, and Fasuyi et al. (2007) leaves of Amaranthus in layer diets. They found no negative effect on bird performance. However, the inclusion levels of Amaranthus leaves should not exceed 10%. In a recent study carried out in Denmark (manuscript in prep.), inclusion levels of 0, 4, 8 and 12% clover grass protein concentrate in diets were tested in layers during a period of 12 weeks (30-42 weeks of age), where the protein concentrate was added at the expense of soya beans. The protein content was 35% DM and the methionine 6.3g/kg DM. The inclusion of green protein concentrate had no effect on egg production compared to the control group (av. 93%), egg weight (av. 62g), feed intake (av. 102g/h/d) or FCR (av. 2.25g/g). There was a significant effect on yolk color ($P<0.0001$), where lightness (L*) was lower and yellowness (b*) and redness (a*) higher with increasing inclusion of clover grass protein concentrate.

The improvement of the biorefining methods to increase the protein content of leaf protein concentrates is currently addressed in several research projects.

Microalgae and macroalgae

Spirulina (*Arthrospira platensis*) belongs to the microalgae, which are single-celled fresh water plants, which grow wildly in lakes and can be cultivated in ponds. Gerrard et al. (2015) reported that organically produced Spirulina algae might replace the use of soya beans in diets for broilers. No significant differences were observed between the control containing soya bean and the Spirulina diet regarding welfare parameters (plumage and food pad quality). The ash content is usually high including high concentrations of mainly sulphur, potassium, sodium and chloride, which has to be taken into account when formulating diets for poultry (van der Poel et al., 2013). In a recent study by Gaillard et al. (2018), the content of protein and amino acids was analysed in different seaweed species in order to study the degradability in dairy cows. The protein content varied to a high degree being generally low in brown algae (7-16% DM), whereas green and red algae species provide a higher protein content (12-39% DM). The harvest season (spring or autumn) had a considerable influence on the quality of the seaweed, which was highest in spring. However, knowledge is lacking on the amino acid digestibility of seaweed in poultry and its value as protein source for monogastric animals need to be studied. The present limitations for the use of micro- and macroalgae are related to accessibility, economy and questions of certification. However, there is a growing interest in using algae for bioenergy, thus there is a potential for a high degree of development in this sector.

Improving the nutritive value of available feed ingredients

Fermentation of feed or feed ingredients

The improvement of the value of available feed resources by fermentation may be an attractive option for organic producers, thus increasing sustainability of their production system. The feeding of fermented compound feed with porridge like consistency (DM content approx. 400 g/kg) to poultry is possible and offers some advantages as shown in an experiment with layers (Engberg et al., 2009). Related to bacterial growth, the dry matter protein content of the feed increases by approx. 7% units. Further, intrinsic plant phytases activated during the steeping process release phytate bound phosphorus thus being available for the bird. This is of special importance for organic production, as dietary supplementation of exogenous enzymes, e.g. microbial derived phytase, is not permitted. Another advantage is the low pH of fermented feed (pH 4.5), acting as a barrier for acid sensitive bacteria, i.e. *E. coli, Salmonella* and Campylobacter colonising the gastrointestinal tract. However, feed with a high water content may present a hygienic risk factor. In order to avoid that, the fermentation of single feed ingredients has shown to beneficially influence their nutritive value.

Xu et al. (2012) showed that solid-state fermentation of rapeseed meal with *Lactobacillus fermentum* and *Bacillus subtilis* over 30 days decreased the content of isothiocyanates dramatically. The addition of up to 10% fermented rapeseed meal at the expense of soybean meal could be applied safely in broiler diets. Ensiling of whole grains (maize and wheat) may likewise increase the nutritional value of the raw material and support gastrointestinal health.

Nutritional value of roughage

In organic poultry production, it is mandatory to provide roughage like fresh hay, silage (of grass/herbs, maize, pea/barley, alfalfa, hemp, sunflower), jerusalem artichokes and other vegetables (beet roots, carrots and kale), which is often fed in relative small amounts several times a day. Roughage plays an important role in crop rotation and therefore supports the sustainability of the organic framing system. Roughage of high quality (i.e. high protein content and low fibre content) can supplement the basic feed with a certain amount of nutritional components (Afrose et al., 2015). Some studies have focussed on the possibilities to increase the use of locally available raw materials. Better knowledge of the nutritional value of various types of roughage may aid the transition to 100% organic feed and may offer the opportunity to take the nutrient content of roughage into account when formulating the diets. It has been shown that depending on the roughage type, the consumption of roughage may account for up to 30% of the total feed intake. Silages based on maize, maize cob, alfalfa, grass and hemp as well as carrots and kale are attractive roughage items for layers. A high roughage intake may reduce the consumption of basic compound feed, thus saving feeding costs. However, it is important that the roughage is of high quality, i.e. the fibre content must not be too high (Steenfeldt & Hammershøj, 2015; Afrose, 2015).

Animal protein sources

Mussels and starfish

Protein of animal origin can supply poultry with a more balanced amino acid composition than vegetable protein sources, and fishmeal is an excellent source for essential amino acids (Sauvant et al. 2004). However, fishmeal is an expensive product and may be a limited resource in future. Marine invertebrates such as mussels have a high concentration of protein and essential amino acids, which quality wise can be compared to fishmeal (Jönsson and Elwinger, 2009). Mussels are filter feeders consuming algae and phytoplankton. Long-line mussel production in fiords and near-shore areas can produce a quality product in a sustainable way and at the same time improve the water quality by removing nitrogen and phosphorus (Lindahl et al., 2005). Results from our laboratory showed that mussel meal contained (% of DM) 15 % raw fat and 60 % protein with an amino acid composition similar to that of fishmeal. Our experiments with layers receiving 4-12 % mussel meal at the expense of fishmeal showed a ood egg production, which was not different from that of layers receiving fishmeal (av. 93%). Feed intake was on av. 114g/h/d and FCR 2.33g/g. No differences with regard to shell quality was observed, but mussel meal significantly improved yolk colour ($P<0.0001$). Further, digestibility of total amino acids increased significantly ($P<0.0001$) with increasing inclusion of mussel meal (Afrose et al., 2016). This indicates that mussel meal has a high value as protein and methionine source for laying hens. A significant drawback of mussels are their shells, which have to be separated from the meat before further processing. The inclusion of starfish meal as protein source for laying hens (4-8% of the feed) showed similar positive results as mussel meal. The protein and methionine content of starfish meal can even be higher than in mussel meal, corresponding to 70% DM and 17 g methionine /kg DM, respectively (Afrose et al., 2016). The use of starfish in organic feed seems immediately tempting as starfish are considered a pest in mussel production consuming mussels in great quantities. The first starfish factory is currently being built in Denmark, where starfish from a nearby fjord is harvested and processed.

Insects

The European Commission is expected to authorize insect proteins in feed for poultry very soon (IPIFF, 2017). Currently, the authorization is restricted to aquaculture and to seven insect species including house fly, black soldier fly, yellow meal worm, lesser mealworm, house cricket, banded cricket and field cricket. With respect to the use of insects in poultry nutrition, the reader is referred to the reviews of Makkar et al. (2014) and Józefiak et al. (2016). Insects contain significant amounts of protein in the range of 42% up to 63% on DM basis depending on the species. Most research regarding the nutritive value of insects in poultry nutrition has been carried out with larvae of the yellow mealworm and the black soldier fly (Makkar et al., 2014). The complete replacement of soya bean meal by mealworm meal (CP content 51.9, ether extracts 21.6%) in broiler diets did not impair growth performance and carcass traits. Dietary inclusion of larvae meals (Yellow mealworm or Black soldier fly) substituting 250 g/kg of a basic diet (based on maize and soya bean) revealed, that both larvae meals are valuable protein sources for broilers (De Marco et al., 2015). A limiting factor for the use of insect meals in poultry diets is their content of chitin, which varies depending on the species and the age of the larvae reducing nutrient digestibility (Bovera et al., 2016). The dietary addition of chitinase may be a tool to overcome this problem. On the other hand, as discussed by Gasco et al. (2018), insect meals rich in chitin may have even health promoting effects. However, insects have a clear potential as protein source for poultry. In order to be competitive in price, it is necessary to intensify insect production, which involves an improvement of equipment and automatization as well as availability of abundant and cheap growth substrates. Due to their temperature requirement for optimal growth, and development, insect meal still has a significant carbon footprint compared to other plant derived protein sources (Tallentire et al., 2018). For the use in organic poultry nutrition, the question remains whether or not insects may comply with the rules of organic production. Further, considering insects as livestock, it can be discussed, that insects similar to other organic animal production should be produced on substrates grown under organic production conditions. However, this is a matter demanding clarification and a political decision.

Conclusions and Perspectives

Currently a number of protein sources as alternative to soya bean meal and fishmeal are available for organic poultry nutrition in Europe. An increasing amount of the protein crops can be cultivated locally and included in crop rotation systems, thus increasing the sustainability of organic production. Different technologies have the potential to improve the nutritive value of readily available feed ingredients and to increase the content of protein with an optimal amino acid profile for poultry. This will offer the opportunity to balance diets for poultry while taking actual amino acid requirements into consideration and thus avoiding the surplus protein supply resulting in increased environmental nitrogen burden.

However, the development of technology in the different sectors still requires a lot of effort in order to increase the competitiveness of alternative protein sources compared to soya beans imported from countries outside Europe. In particular, the energy demanding drying of wet feed ingredients, presents a serious challenge with respect to energy costs, thus increasing carbon footprints considerably (De Bohr et al., 2014).

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Table 1. Concentration of protein (% DM) and amino acid (kg/DM) in different vegetable and animal sources

Source	Protein % of DM	Amino acids, g/kg DM			Reference
		Methionine	Cystine	Lysine	
Vegetable protein					
Legumes					
Faba bean	25.0-31.1	2.1-2.2	3.7-3.9	16.4-20.0	Sauvant et al., 2004; Masey O'Neil et al., 2012
Pea	23.1-24	1.6-2.4	3.24-3.5	15.7-17.4	Sauvant et al., 2004; Hejdysz et al., 2016
Lupin. blue	32.4-35.1	2.1-2.2	4.6-5.0	15.1-16.1	Petterson, 2000; Hammershøj and Steenfeldt, 2005
Lupin. yellow	39.0-41.8	3.0-3.2	9.6-10.0	18.7-22.6	Petterson, 2000; Rutkowski et al. 2017
Lupin. white	39.0	2.6	5.5	17.3	Petterson, 2000;
Soya beans	39.7-48.8	4.1-6.3	6.1-8.8	24.6-29.1	Grieshop and Fahey, 2001; Sauvant et al., 2004; Steenfeldt and Hammershøj, 2015; Afrose 2015
Other Seeds					
Sunflower	17.2-18	4.1	3.1-3.3	6.3-6.8	WPSA, 1992; Sauvant et al., 2004
Sunflower expeller	26.0-37.5	5.7-8.2	4.6-6.7	9.0-13.0	WPSA, 1992; CVB, 2009;
Rapeseed	20.7-21.5	4.5	5.1-5.6	12.6-12.9	WPSA 1992; Sauvant et al., 2004
Rapeseed expeller	36.2-38	7.7	9.6	20.9	WPSA, 1992; CVB, 2009
Hemp	26.5-34.2	6.2-7.5	4.4-6.0	10.0-11.0	Callaway, 2004; House et a., 2010
Quinoa	14.2-15.6	2.6-2.9	2.0-2.5	7.2-8.1	Ruales and Nair, 1992; Steenfeldt and Hammershøj, 2015; Nowak et al. 2016
Sainfoin seed	30.6	5.2	4.5	16.9	Baldinger et al., 2014
Sainfoin seed dehulled	42.4	6.9	6.0	22.7	Baldinger et al., 2014
DDGS					
DDGS wheat	36.3	5.5	6.8	7.6	Pottgüter, 2015
DDGS maize	29.7	5.7	5.5	8.6	Pottgüter, 2015

Table 1. Continued

Source	Protein % of DM	Amino acids, g/kg DM			Reference
		Methionine	Cystine	Lysine	
Green biomass concentrate					
Red clover	38.0	8.5	2.5	26.8	Santamaria-Fernandez et al., 2017
Clover grass	40.0	8.2	2.4	23.9	Santamaria-Fernandez et al., 2017
Alfalfa	42.0	7.8	3.4	21.5	Santamaria-Fernandez et al., 2017
Oilseed radish	48.0	9.1	4.7	25.4	Santamaria-Fernandez et al., 2017
Algae					
Brown algae	6.8-16.4	1.4-3.1	1.0-2.4	2.8-7.5	Gaillard et al., 2018
Green algae	12.2-34.1	2.6-4.7	2.2-11.3	5.3-15.5	Gaillard et al., 2018
Red seaweed	14.9-39.7	2.3-7.3	5.1-6.3	7.1-18.2	Gaillard et al., 2018
Roughage					
Maize silage	8.5-9.2	1.4-1.6	1.2	1.8-3.1	Steenfeldt and Hammershøj, 2015; Afrose, 2015
Alfalfa silage	22.3-23.5	2.5-3.5	1.1-1.7	3.4-6.2	Steenfeldt and Hammershøj, 2015; Afrose, 2015
Hemp silage	17.3	2.6	1.8	5.6	Afrose, 2015
Kale	22.7-27.6	3.58-3.8	2.4-3.4	12.1-13.8	Hammershøj and Steenfeldt, 2012; Afrose, 2015
Animal protein					
Marine sources					
Musselmeal, de-shelled	60.5-71.1	14.1-17.7	8.9-11.1	43.5-53.5	Jönsson and Ewinger, 2009; Afrose et al., 2016
Starfishmeal	70.0	17.5	5.3	43.4	Afrose et al., 2016
Insect meals					
House fly	50.4	11.1	3.5	30.7	Makkar et al., 2014
Black soldier fly	42.1	8.8	1,4	27.8	Makkar et al., 2014
Yellow mealworm	52.8	7.9	4.2	28.5	Makkar et al., 2014
House cricket	63.3	8.9	5.1	34.2	Makkar et al., 2014

Nonessential amino acids – the forgotten nutrients?

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Running headline: Nonessential amino acids

Summary: Nonessential amino acids (AA), particularly glycine and serine, recently have received increasing attention in the context of crude protein (CP) reduction in poultry feed. Reduced CP concentrations in the feed enable to decrease nitrogen emissions related to farm animal husbandry by a more efficient use of protein in feedstuffs. The possibility to reduce CP concentrations without impaired growth performance by only optimizing the concentrations of essential AA in poultry feed is limited. CP concentrations can be further reduced considerably when glycine and serine are additionally considered in feed formation. The response to glycine and serine, preferentially expressed as glycine equivalents (Gly_{equi}), was determined to be highly variable between studies. Known factors influencing the Gly_{equi} requirement are the concentrations of cysteine, threonine, choline, and arginine in the feed. Uric acid synthesis also seems to contribute considerably to the requirement for Gly_{equi} because glycine is needed for uric acid synthesis. The variable requirement for dietary Gly_{equi} means that static recommendations for Gly_{equi} concentrations in poultry feed lead to high safety margins in Gly_{equi} supply or holds the risk of Gly_{equi} deficiency. Variable recommendations for Gly_{equi} depending on concentrations of other nutrients in the feed would overcome this issue. Considering the concentrations of Gly_{equi} and essential AA in feed formulation enables to formulate feed for broiler chickens from day 1 to day 21 post-hatch with about 15 to 16% CP without affecting growth performance. Glycine and serine are the first nonessential AA of which requirement values were quantified. Requirements for other nonessential AA are unknown to date.

Keywords: *nonessential amino acids, poultry, glycine, serine*

Introduction

The 20 proteinogenic AA have been categorized into several schemes since they have been discovered, which was in the 19th century for most AA. One of these schemes is a classification in essential and nonessential AA. A perception has developed that considers nonessential AA less important in feed formulation. This perception may have been supported by the term dispensable AA, which is used synonymously to nonessential.

The requirement for essential AA has been investigated extensively since the middle of the last century. The increasingly advancing knowledge of the requirement for essential AA has enabled the reduction of the CP content in the feed, thereby avoiding essential AA deficiency. An early recommendation of the NRC (1960) was 28% CP in feed for 0 to 8 week-old broiler chickens, while the latest recommendation for CP of the NRC (1994) is 23% for 0 to 3 week-old broiler chickens. Current practical recommendations for this age period usually range between 21% and 22% CP (e.g. DLG, 2017).

A further reduction of CP concentration in feed is advantageous because of economic drawbacks of feeding excess CP. Beyond that, the worldwide increasing demand for meat and other animal products and concomitant limitation of arable land results in a foreseeable shortage of protein-rich feedstuffs. Consequences of increased crop prices have been shown to especially affect affordability of food in developing countries (Fazeni and Steinmüller, 2011). Feeding protein above the requirement of the animals results in an excretion of nitrogenous compounds which can have negative effects on the environment.

The possibility to reduce the CP content in poultry diets without undesirable effects on performance was repeatedly shown to be limited even when the requirement of essential AA was considered. It recently was recognized that the nonessential AA glycine and serine need to be considered when CP contents in poultry diets are aimed to be decreased further. However, assessing requirements and recommendations for glycine and serine currently is far from being completed. Amongst other reasons, this is because the relevance of glycine and serine in low CP feed became clear only recently and because assessing requirements for nonessential AA needs further development of methods.

This contribution aims to summarize the current knowledge on the requirement for nonessential AA and consequences thereof for CP reduction in poultry. Considerations on how recommendations might evolve are also included.

Approaches of lowering crude protein concentrations

Current research mainly aims to find ways to reduce the CP concentration without compromising growth and feed efficiency. Dean *et al.* (2006) summarized that in literature growth and feed efficiency was reduced in broiler chickens fed with diets containing less than 19% to 20% CP even when the requirement for essential AA was met. Several potential reasons for these undesirable effects were discussed by Aftab *et al.* (2006) and should not all repeated herein.

Numerous studies have investigated whether the requirement for essential AA differs between low and standard CP feeds. Results consistently did not overcome the reduced growth performance induced by low CP feed. Other studies have investigated the effect of nonspecific nonessential AA-nitrogen, which was shown to have no effect on the growth performance in some studies. In other studies, a growth-increasing effect compared to the low CP feed without addition of nonspecific nonessential AA-nitrogen was reported. However, the level of growth that was achieved with the standard CP level could not be attained. Consideration of nonspecific nonessential AA-nitrogen alone seems to be not sufficient because it assumes unlimited interconversion of nonessential AA. Instead, several studies showed that adequate supply of specific nonessential AA is required in low CP diets (Siegert and Rodehutscord, 2015).

Growth-increasing effects were determined when glycine or serine were specifically supplemented to low CP feed. After two studies had shown that glycine-supplemented feed that contained about 17% CP caused no difference in growth performance compared to feed with 22% CP (Corzo *et al.*, 2005; Dean *et al.*, 2006), this subject received even more attention in broiler nutrition research.

Relationship between glycine and serine

Glycine can be metabolized from serine by splitting the hydroxymethyl group of serine. This reaction can be reversed by adding CH3 from tetrahydrofolic acid (Velíšek and Cejpek, 2006). For poultry, it is generally assumed that the interconversion of glycine and serine is not limited in metabolism (Akrabawi and Kratzer, 1968; Sugahara and Kandatsu, 1976). Therefore, glycine and serine are usually assessed together in poultry nutrition studies. Most authors and groups, including the NRC (1994), considered the analogue effect of dietary glycine and serine by using the sum of both AA (Gly+Ser), neglecting that serine has the same effect as glycine only on an equimolar basis. Dean *et al.* (2006) proposed to describe the physiological value in a better way by calculating the glycine equivalent (Gly_{equi}) as the sum of the concentration of glycine and the molar equivalent of serine, which is 0.7143:

Gly_{equi} (g/kg) = glycine (g/kg) + [0.7143 × serine (g/kg)]

Physiological functions of glycine and serine

Like any other proteinogenic AA, glycine and serine are incorporated in proteins. Whole body accretion of glycine and serine in broiler chickens from day 8 to day 21 was determined between 7.8 and 11.4 g glycine accretion and between 4.2 and 5.5 g serine/16 g N accretion (Fatufe *et al.*, 2004; Fatufe AND RODEHUTSCORD, 2005).

Proteins incorporating glycine and serine

Collagen and elastin: The proteins richest in glycine are collagen and elastin, where glycine is incorporated at every third position in the primary structure. In broiler slaughter processing, low skin strength due low collagen content in consequence of low dietary supply with glycine can have economic implications (Christensen et al., 1994).

Keratin: Keratin is rich in both glycine and serine. In avian species, keratin is mainly present in feathers and claws. Feather development of birds fed with glycine deficient diets is impaired (Robel, 1977).

Mucin: Mucin proteins are rich in serine because serine provides attachment sites for the oligosaccharide chains which have a high proportion in mucins. The physiological functions of mucin are described as lubrication of the gut epithelium, protection of the epithelium against acidic conditions and proteases, and selective diffusion barrier for nutrients. Further microbiota-associated functions are fixation of commensal bacteria, protection of the epithelium from pathogens, and substrate for bacterial fermentation. Ospina-Rojas et al. (2013) found that intestinal mucin secretion of broilers linearly increased with the proportion of glycine and serine in the diet.

Processes using metabolization products of glycine or serine

Uric acid: In birds, ammonia is detoxified and excreted as uric acid being the main excretion product of nitrogen metabolism. The formation of each molecule of uric acid requires one molecule of glycine to build the purine ring. In addition, protein synthesis and cell proliferation depends on DNA synthesis, which requires glycine to form purines (Wang et al., 2013).

Creatine: Glycine is an integral part of creatine along with arginine. Creatine can either be directly supplied by feed derived from animal products or produced by endogenous synthesis. Arginine reacts with glycine in a first step to form guanidino acetic acid. In a second step, guanidino acetic acid is methylated to form creatinine.

Cysteine synthesis: Most species metabolize cysteine from methionine. Serine is required when cystathionine is formed from homocysteine. Cystathionine further reacts to cysteine and ammonia (Velíšek and Cejpek, 2006).

Bile salts: Primary bile salts are synthesized from cholesterol in the liver and then conjugated with either glycine or taurine. The proportion of bile salts conjugated with glycine or taurine is different between animal species. In broiler chickens, bile salts were found to be almost exclusively conjugated with taurine (Hofmann *et al.*, 2010). Glycine deficiency was found to affect fat digestibility in laying hens and broiler chickens, and therefore, the MEN concentration in feed (Han and Thacker, 2011; Ospina-Rojas *et al.*, 2013). These authors discussed this to result from different levels of bile production as a consequence of glycine availability. However, this theory in not fully supported by results of Hofmann *et al.* (2010) who found that formation of glycine-conjugated bile salts was particularly low in broiler chickens.

Responses of broiler chickens to glycine and supplements

Responses to a nonessential AA supply were mostly investigated using broiler chickens in their first 21 days post-hatch. Therefore, this chapter will focus on this age period. However, research on older broiler chickens is also considered.

Since the importance of glycine and serine in poultry nutrition has been recognized, the number of published studies in this area has largely increased. A compilation of some dose-response studies showed that the response to dietary Gly_{equi} was inconsistent (Figure 1. Glyequi concentrations needed to achieve 95% of the maximum gain:feed response (based on the 10 studies used by Siegert et al., 2015b)). Therefore, factors influencing the response to Gly_{equi} are addressed in the following chapters and get back to some of the physiological functions mentioned above.

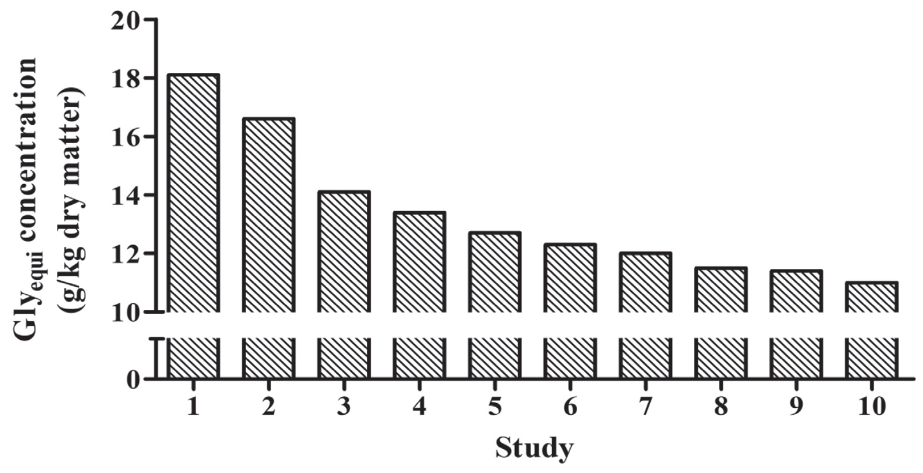

Figure 1. Gly_{equi} concentrations needed to achieve 95% of the maximum gain:feed response (based on the 10 studies used by Siegert *et al.*, 2015b)

Effect of cysteine

Powell *et al.* (2011) found that the impact of glycine and serine on the gain:feed response is explained by the conversion of methionine to cysteine, for which serine is required.

They found an increased gain:feed response upon addition of glycine to feed adequate in total sulfur amino acids (TSAA) but deficient in cysteine. Further inclusion of methionine showed no increase in growth, but the addition of cysteine above the requirement reduced the growth-increasing effects of glycine supplementation. Low CP concentrations are often associated with the inclusion of pure ingredients, such as dl-methionine or analogues. Because cysteine is usually not added, the ratio between methionine and TSAA (Met/TSAA ratio) in the feed is increased upon supplementing dl -methionine, disregarding a specific requirement of cysteine.

The concentrations of both methionine and cysteine in the feed have a substantial impact on the response of broiler chickens to dietary Gly_{equi} (Siegert *et al.*, 2015b) (Figure 2). Fulfilling the requirement for both methionine and cysteine reduces the necessity of the conversion of methionine to cysteine.

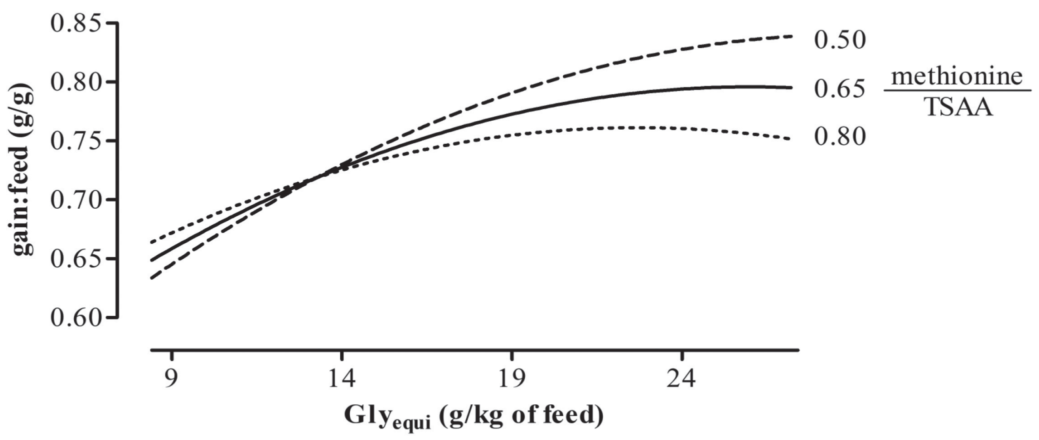

Figure 2. Effect of the Met/TSAA ratio on the gain:feed response of broiler chickens to dietary Gly_{equi} in a meta-analysis compiled from 10 peer-reviewed studies investigating the growth response of up to 21 day old broiler chickens (modified from Siegert *et al.* [2015b]).

Effect of endogenous precursors of glycine and serine

Threonine can be metabolized to glycine (Meléndez-Hevia *et al.,* 2009). Another precursor is choline, which, with betaine as an intermediate step, is metabolized to glycine if homocysteine is available. Other endogenous precursors of glycine are glyoxylate in combination with alanine, and trimethyllysine when glycine is produced as a by-product of carnitine synthesis. However, glyoxylate has no relevance in broiler feeding, and the potential quantity of glycine metabolized from trimethyllysine is low (Meléndez-Hevia *et al.,* 2009).

Threonine: Interactions between the Glyequi and threonine concentrations in the feed have been reported (Corzo *et al.*, 2009; Ospina-Rojas *et al.,* 2013) and later quantified (Siegert *et al.*, 2015a [Figure 3]; Lambert et al., 2015). An increase in threonine concentration reduced the Gly_{equi} concentration required to achieve certain response levels. One molecule of threonine can be converted to one molecule of glycine. Taken the molar weights of both molecules, the replacement value due to the endogenous conversion of one mass unit of threonine cannot exceed 0.63 mass units of glycine. However, a much higher replacement can be derived from broiler responses (Siegert *et al.*, 2015a; Lambert *et al.*, 2015; Ospina-Rojas *et al.*, 2013). This apparent discrepancy most likely results from a relative excess of other AA in threonine-limited diets. Increasing the threonine concentration probably reduced catabolism of AA (other than threonine), thereby reducing the need for Glyequi to be used for uric acid formation.

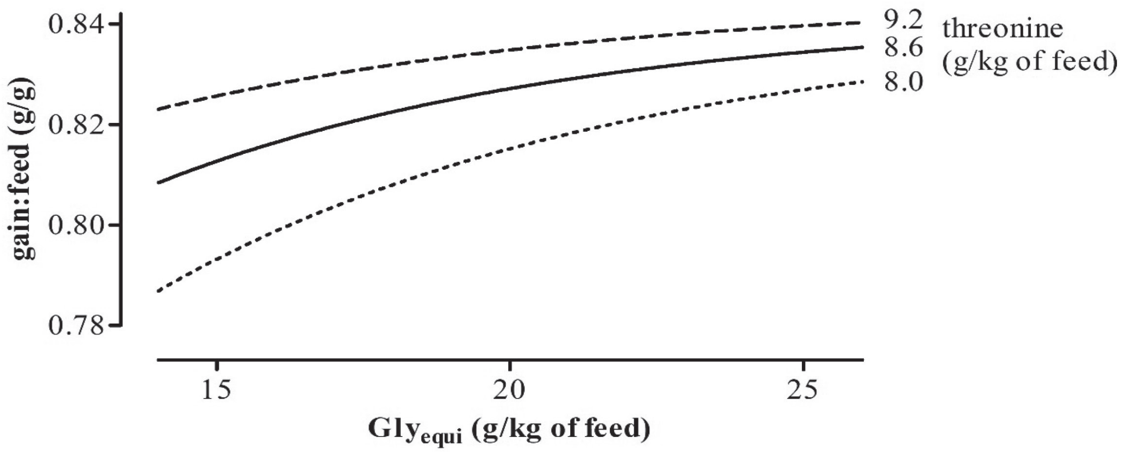

Figure 3. Effect of the threonine concentration on the gain:feed response of broiler chickens to dietary Gly_{equi} from 7 to 21 days post-hatch (nutrient concentrations calculated for feed with 88% dry matter; modified from Siegert *et al.* [2015a]).

Choline: Choline can affect the growth response of broiler chickens to Glyequi supplements (Siegert et al., 2015a). However, the effect of choline on the response to Glyequi was lower compared to the effect of threonine.

Arginine

Arginine and glycine are precursors of guanidino acetic acid, which further reacts to form creatine (Akinde, 2014). Significant interaction effects for combinations of components including creatine, guanidino acetic acid, arginine, and glycine on the growth response were reported for broiler chickens in several studies (e.g. Dilger *et al.*, 2013; Ringel *et al.*, 2008). To our knowledge, however, no literature is available investigating different combinations of Gly_{equi}, arginine, and guanidino acetic acid in the feed simultaneously.

Current status and perspectives of crude protein reduction in feed for broiler chickens

Recently published studies have shown that reduced growth performance is not to be expected when free glycine is supplemented to industry-type feed with CP concentrations below 19% (van Harn *et al.*, 2017; Hilliar *et al.,* 2017). Such feed could be used by the industry.

However, it is worth further investigating to which level the CP concentration in feed can be reduced when Gly_{equi} concentrations are adequate. The growth response of broiler chickens from day 7 to day 21 post-hatch was similar with 16.7% and more than 20% CP in Gly_{equi} adequate feed (Siegert *et al.*, 2015c). In another study, a similar level of growth performance was observed with feed based on corn and soybean meal that contained 16.3% CP and adequate concentrations of essential AA and Gly_{equi} (Hofmann *et al.*, 2018). However, the latter study showed that growth performance was reduced when feed with 14.7% CP was fed although concentrations of essential AA and Gly_{equi} were adequate. The next-limiting nitrogenous nutrient in feed for broiler chickens, presumably another nonessential AA, currently is unknown. Glycine and serine are the first nonessential AA of which requirement values were quantified. The possibility to further reduce the CP concentration in feed without adverse effects on performance can be expected when the role of other nonessential AA is better understood and their requirement values are assessable.

For broiler chickens from 21 to 35 days post-hatch, Gly_{equi} was found to become growth-limiting at CP concentrations below 16% to 17% in the feed (Belloir *et al.*, 2017). As in younger birds, the response to dietary Gly_{equi} at this age was also different between studies (Ospina-Rojas *et al.*, 2014; Vasconcellos *et al.,* 2011). To our knowledge, no literature is available investigating the effect of influencing factors on the response to Gly_{equi} in the feed in different age periods. Interrelations most likely are similar to younger broiler chickens, but the extent of interrelations might differ.

Results of other growing poultry species and laying hens

To our knowledge, no dose-response studies using Gly_{equi} with other growing poultry species than broiler chickens are published. However, the effect of low CP feed supplemented with free glycine has been investigated in White Pekin ducks (Xie *et al.*, 2017) und quails (Wen *et al.*, 2017). These studies showed that supplementing feed with free glycine enables to reduce the CP concentration in feed without reducing growth performance below the limit of CP reduction when only concentrations of essential AA are considered in feed formulation.

Literature on responses of laying hens to nonessential AA is very scarce. Han and Thacker (2011) described a linear increase in feed intake, egg production, and egg weight of laying hens with increasing Gly_{equi} in feed containing 16.9% CP and Gly_{equi} concentrations ranging from 14.6 to 16.2 g/kg. This study indicates that the Glyequi supply can become relevant in laying hens, although the CP concentration was not markedly reduced below what is currently common practice.

The metabolism is not substantially different between poultry species. The interactions between Gly_{equi} and other nutrients that were reported for broiler chickens probably are also relevant in other growing and laying poultry species, though their quantitative dimension might be different.

Gly_{equi} requirements of broiler chickens and consequences for recommendations

Several studies indicated that considering Gly_{equi} in feed formulation is more relevant than considering the concentrations of some essential AA. Waguespack *et al.* (2009) considered Gly_{equi} as the fourth-limiting of all proteinogenic AA after methionine+cysteine, lysine, and threonine in a feed based on corn and soybean meal for broiler chickens from 1 to 18 days post-hatch. Similar results were published by Ospina-Rojas et al. (2014), who characterized valine and Gly_{equi} as equally limiting after methionine+cysteine, lysine, and threonine in a corn-soybean meal-based feed for broiler chickens from 1 to 21 days post-hatch.

The requirement for Gly_{equi} has been repeatedly shown not to be constant. Instead, the requirement for Gly_{equi} varies considerably depending on the dietary concentrations of other nutrients, such as threonine, arginine, cysteine, and choline. As indicated by Siegert *et al.* (2015a) and Hofmann *et al.* (2018), the amount of nitrogen that has to be excreted via uric acid is another very relevant determinant of the requirement for dietary Gly_{equi}. The variable requirement for dietary Gly_{equi} means that a static recommendation for Gly_{equ}i in poultry feed would imply high safety margins in Gly_{equi} supply or hold the risk of Gly_{equi} deficiency.

Based on existing literature, the requirement for Gly_{equi} in feed for up to 21 day old broiler chickens is estimated to vary between 11 and 20 g/kg. The lower end of this range is approached when uric acid formation and the need to metabolize cysteine from methionine is low, and when concentrations of endogenous precursors like threonine and choline in the feed are high. Further quantification of the effect of these influencing factors on the requirement for Gly_{equi} in feed is needed.

Current possibilities for crude protein reduction

Compared to nowadays common practice, the scientific knowledge enables to reduce the CP concentration in the feed by 5 to 6 percentage points. Several aspects are not in support of utilizing this potential currently. Such aspects include the mostly imprecise knowledge of concentrations and digestibility of AA of raw materials. Another aspect with specific regard to the European Union is list of raw materials and feed additives currently approved. This is because the concentration of Gly_{equi} in plant proteins is relatively constant. Therefore, using only plant and mineral feedstuffs in feed formulation causes the dietary concentration of Gly_{equi} to depend mainly on the CP concentration of the feed (Siegert and Rodehutscord, 2015). In other regions of the world, animal byproducts can be used to elevate the Gly_{equi} concentration in diets. Feed additives potentially suitable for elevating the Gly_{equi} concentration in feedstuff are free glycine and l-serine, which are currently also not approved in the European Union (European Commission, 2014).

Conclusions

Increasingly restrictive legislation in regard to emissions caused by animal husbandry in the European Union and expected price increases for protein sources makes it necessary to further reduce of CP concentrations in animal feeds. This needs an optimization of the AA composition according to the requirement of the animals. Considering Gly_{equi} in feed formulation is necessary to reduce the CP concentration in poultry feeds below the current standard. The Gly_{equi} requirement of broiler chickens is variable depending on other constituents of the diet. Therefore, variable recommendations for Gly_{equi} would contribute to supply the animals according to their requirement and to reduce nitrogen emissions from poultry farming. This potential can be best used when plant feedstuffs are combined with animal byproducts or free AA or both.

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Technology for Precision Livestock Farming in the Poultry Industry

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1. Introduction

The European Union is a major producer of poultry meat with about 6 billion chickens being raised annually. In conventional production systems birds are slaughtered after 42 days at a weight of about 2.5kg. In the current economic climate poultry farmers are required to grow more and more birds in order to make their business profitable. It is not uncommon to see broiler farmers raise close to 2 million birds per year (6 buildings x 6 flocks per year x 50,000 birds). It has become impossible for a farmer to observe all of his animals often enough by audio-visual (direct, human) observations. Farmers are rarely faced with an easy life, as technical issues arise daily, e.g. climate control problems, blocked feeder lines, electricity problems, drinking lines, failing lightning systems and so on. To carry out daily assessments of the welfare alongside the daily management routine takes vital hours from the farmer's day, affecting the profitability of their business. It has been known for many years that, whatever production system is used, the farmer remains the most important factor in obtaining good results at the level of animal health and productivity (Geers et al., 1985).

Access to feed, water and a suitable thermal environment are primary components of good health and welfare, so the farmers must quickly be made aware of any restriction or deviation from acceptable conditions. Moreover, whether or not feeders and drinkers are working still need to be checked manually. For this the clever application of technology has remarkable potential to assist the farmer. Twenty years ago carrying personal computer in a satchel was a novel experience, yet nowadays we have powerful devices that fit in the pocket. This evolution in the ICT world has had a significant impact on the agricultural industry too. Agricultural production have, for the last number of years, witnessed an explosion of new software systems, monitoring devices and machines that all exploit the latest capabilities in sensing, communication, processing and power management (van Evert et al., 2017).

The field of Precision Livestock Farming (PLF) offers a technology-based approach to commercial livestock farming. PLF builds on the basis that fully automated continuous monitoring of animals will enable farmers to detect and control, in real time, the health and welfare status of their animals, and that this information can be used for management. Modern technology makes it possible to use cameras, microphones and sensors sufficiently close to the animal so that they can replace the farmers' eyes and ears in monitoring individual animals, and this can take place during 7 days a week, 24 hours a day, 3600 seconds per hour. The development of precision livestock farming tools requires the connection of sensor data (derived from cameras, microphones or other sensors) with the biological responses of animals through the implementation of real-time algorithms.

The objective of this paper is to demonstrate how PLF can impact the poultry sector, going from laboratory to systems that are operational in commercial farms as well as other technologies that will be available in future commercial PLF systems. Moreover, the examples show how real time automated data analyses can generate added value for the farmer. PLF systems can replace the ears and the eyes of the farmer and work 24 h a day and 7 days a week to support him.

2. What is Precision Livestock Farming?

Precision Livestock Farming (PLF) focussed not on the development of ICT-based gadgets but on implementation of process knowledge into solutions that can really improve livestock monitoring and efficiency of processes to grow meat and animal products like milk and eggs. This way PLF can create systems that are based on continuous automated real-time monitoring and control of animal health and welfare, production/reproduction and environmental impact of livestock production. The real time aspect and being part of the management system is quite different from other solutions like the use of so called Iceberg indicators (FAWC, 1979) or Welfare Quality approach (Welfare Quality).

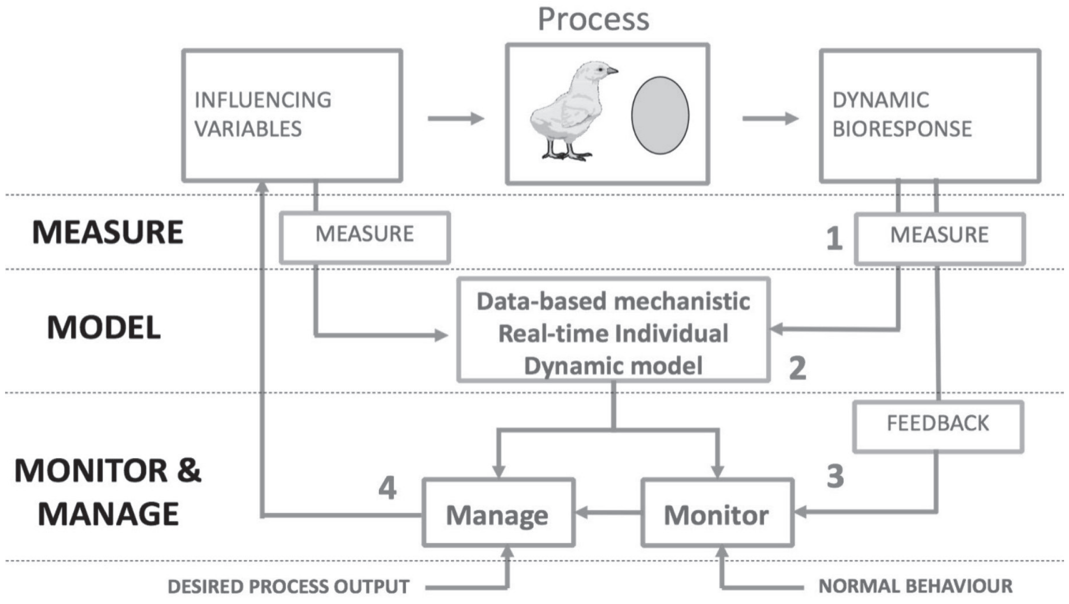

Figure 1 Schematic of the key principles of precision livestock farming (Aerts et al., 2003)

Over the last years, there is an increasing interest in the automated monitoring of animal welfare indicators. Vocalisations of chickens have been researched in relation with welfare, social separation, thermal comfort, feather pecking, stress, diseases and growth. Additional to sound analysis, also image analysis is used to monitor animals in livestock houses. Cameras have, same as microphones, the advantage that they are not mounted to the animal itself and therefore no extra stress for the animals is induced. Image analysis has been used in the past to measure some important bioresponses regarding health, welfare and growth parameters of animals. Examples are: estimating animal weight, assessing the gate score of chicken broilers, measuring the water intake in pigs and assessing the lameness of broiler chickens.

The PLF approach comprises the simultaneous measurement of variables on the animals (including developing embryos) and in their micro-environment, and the implementation of real-time dynamic models for monitoring and control purposes (e.g., Berckmans, 2006, 2017 and Norton and Berckmans, 2017). PLF research has to date shown potential in monitoring and control of various processes ranging from bees to poultry, pigs and cows. This research has led to the development of tools (some now on the market) which aim to combine audio and/or video information in an online automated system, in order to monitor, model & manage the biological responses of the animals. Figure 1 shows the primary scheme of PLF-based monitoring and management (Aerts et al., 2003).

As seen in the figure, PLF systems rely on the following basic principles:

1. the ability to continuously **measure** relevant field data (i.e. sound, image, sensor) at a suitable frequency level,
2. a compact mathematical **model** that is able to predict online and in real time the time-variant, complex, dynamic response of these field data,
3. one or more target variables (e.g. heat stress level) that can be unambiguously linked to a (field data related) feature variable (e.g. egg shell temperature, embryo heart rate and motion) which is **monitored** in real time,
4. if possible: a predictive controller, enabling to **manage** in real time the target value responses using the dynamics of the feature variable in the control.

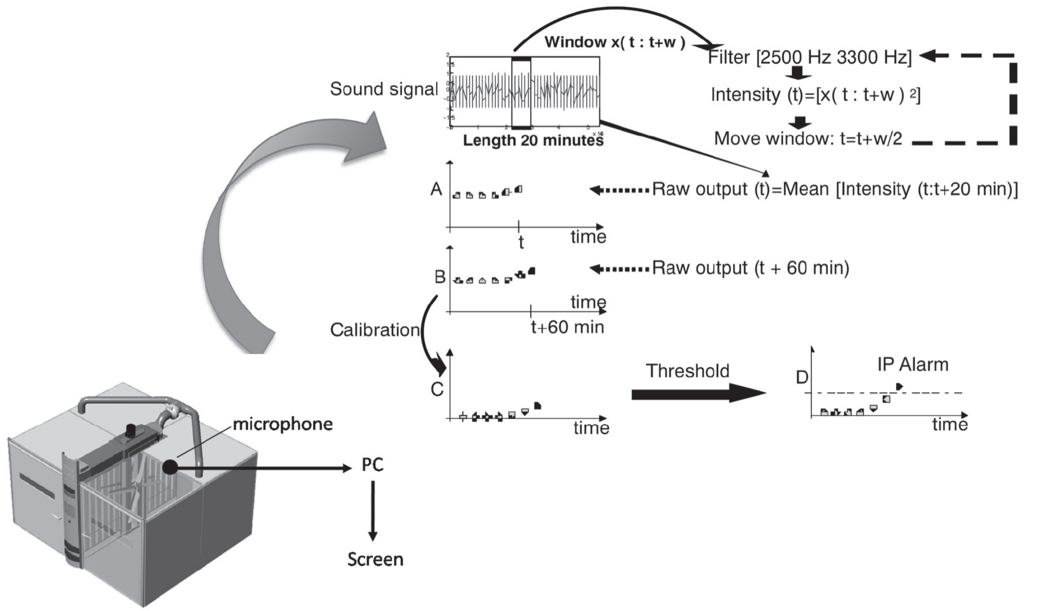

Figure 2 Scematic of methodology developed for automatic external pipping detection in industrial incubators (Silva et al., 2010)

To ensure the bio-response is correctly measured the following global analysis framework for developing such a methodology are distinguished here:

- bio-signal registration, by use of a sensor,
- real-time signal data pre-processing,
- identifying a number of events out of the monitored bio-signals (an event can be defined as a timeframe in the signal with a certain meaning),
- classifying the events, based on their signal characteristics,
- correlating these events with output data characteristics.

This approach is fundamental to the development of new monitoring and management methodologies for the incubator and chicken growing environment. Figure 2 demonstrates the translation of the general methodology to the incubator setting (Silva et al., 2010).

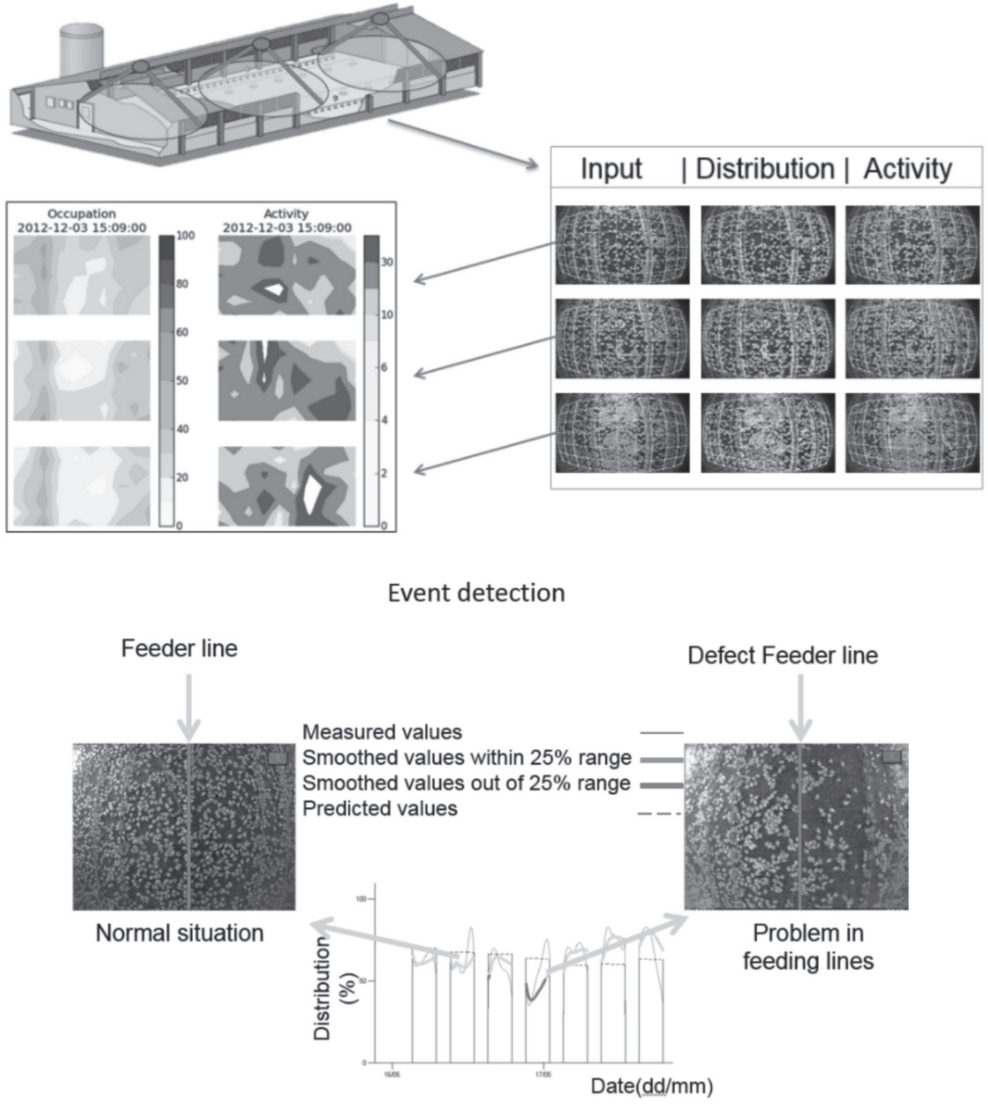

Figure 3. Behaviour of broilers as analysed in real time by the eYeNamic system

3. Examples of PLF Technology in poultry production

3.1. Camera based broiler behaviour monitoring

Whilst environmental sensors provide information on the average house environment, they cannot give an animal-based measure of the animal and micro-climate interaction. To address such challenges a Precision Livestock Farming tool called eYeNamic (Fancom BV, the Netherlands) has been developed. This system is camera-based and continuously monitors the broiler flock, enabling the detection of daily problems in broiler houses. For a typical broiler house, the system employs up to 4 cameras mounted in the ceiling, that give pictures of the distribution of the birds (de Montis et al., 2015). The system uses an algorithm that compares the actually measured distribution of animals with a predicted value at that time of the day. When the real measured value is more than 25 % different from the predicted value, an alarm can be given to the farmer (Figure 3). This alarm indicates a problem in the barn, such as a blocked water line (Kashiha et al., 2013).

Recent research with the eYeNamic system (Penz-Fernandez et al., 2017) has revealed correlations between activity and distributions indices and welfare problems, specifically hock burn and footpad lesions (Figure 4). The relation between the deviations in occupation patterns and the foot pad lesion scores is positive, indicating that birds which tend to cluster together for long periods present an increased chance for food pad lesions. On the other hand the relation found between the deviations in the activity pattern and the hock burn scores is negative, indicating that a higher activity of the flock would improve the hock burn scoring. This can be related with the fact that having less active broiler chickens staying still for longer periods on badly conditioned litter can worsen this kind of lesions (Haslam et al. 2007). This latter result is in agreement with the work of Dawkins et al. (2013), in which they found that the percentage of birds walking was negative correlated with the hock burns.

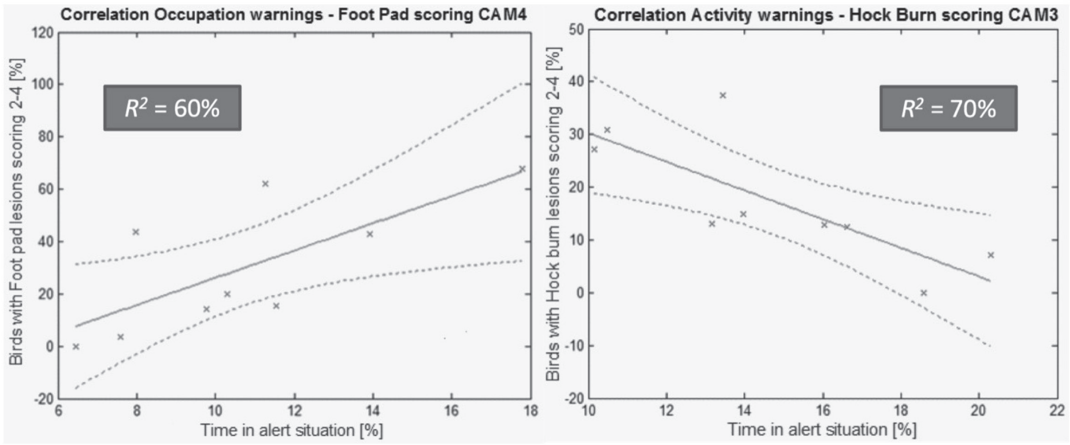

Figure 4. Correlation between time spent in alarm situation and welfare scores

Another camera-based monitoring approach similar to the eYeNamic has been recently developed by a research group in Oxford University (Dawkins et al., 2013). The approach is based on an analysis approach called optical flow. This approach derives the general flow of birds from the images, as well as the variance, skew and kurtosis of the flow. The approach has already been correlated with health and welfare indicators, proving that a general indication of problems is possible while the specific disease diagnosis is less convincing.

3.2 Continuous sound analysis for an activity of broilers

Microphones and cameras have the advantage that they are not in contact with the animals thus do not induce additional stress. Also, they can monitor multiple animals / group of animals at the same time and the equipment can be used to monitor different production cycles. Therefore, the price of the equipment can be spread over multiple animals and multiple production cycles. When comparing microphones and cameras, microphones have the advantage that there is no need for light, they are not affected by blind spots, and they don't need to be cleaned in contrast to the cameras' lenses. Looking at housing conditions in commercial companies microphones could be used to monitor animals living on different levels in the house (e.g. aviary system for laying hens) whereas cameras attached to the roof are restricted to only a 2D top view image of this house. This example shows the potential to use sound to monitor activity in a broiler house.

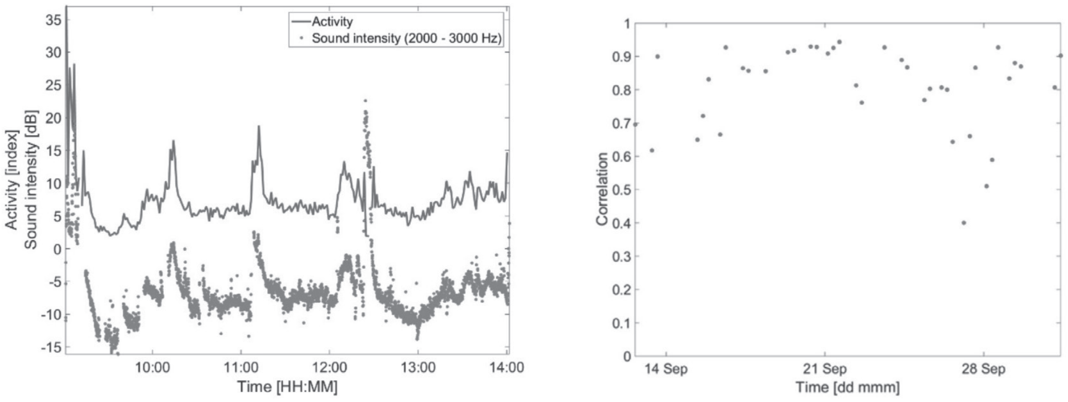

Figure 5. Relation between activity derived from sound and image sensors

Sound has the advantage over image technology that it can be used in the dark, also there are no blind spots. In addition, the pixel activity tend to decrease to the end of the round because all space is filled with broilers and when one moves their spot is taken by another bird and no activity is recorded. To demonstrate a application of sound for activity analysis three cameras and microphones recorded continuously during a production cycle in a commercial broiler farm. The activity was calculated from the images and the sound power produced by the chickens was calculated from the sound. To calculate the energy a band pass filter (2000 – 3000 Hz) was used. Additionally, all sounds with very noisy background (e.g. heater) were removed as they affect the accuracy of the sound power estimation. The Pearson correlation between the activity and the sound power was calculated for the first 21 days of the flocks' life during light periods. Light periods in which less than 95% of the camera information and less than 50% of the sound information was available were removed from this study. The correlation (mean (standard deviation)) between camera and microphone that monitored the same region was 80.5% (12.8%), 75.5% (10.3%), and 58.6% (16.6%). The results from the first and second camera and microphone indicate that sound could be used to assess the activity in a broiler house and hence, sound power could be used as feature variable to detect welfare and health issues (Figure 5). In the last dataset, a decrease of correlation could be observed in proportion with the age of the birds.

3.3 Sound-based growth monitoring

A further aspect of high importance in a broiler farm is the growth trend of the flock, since it is an important part of modern broiler production representing the efficiency and profitability of the processing plant. The average weight of the flock is generally evaluated either manually or automatically using samples of birds chosen at random within a poultry house. The manual measurement of the weight of a representative number of animals in a building is time and labour intensive, since buildings may hold up to 50k birds. Today, many farms use "step-on scales" placed on the floor of the poultry house to automatically collect the average weight of the birds in the flock. Even if the weighing system gives an accurate weight value each time a bird steps onto it, the weight is only representative for the birds that access the automated weighing system and certainly not representative for all the birds in the flock.

The accuracy of automated weighing is limited due to (a) the reluctance of heavy birds to visit the weighing scale (which requires the bird to climb up onto the scale) at the end of the production period and (b) the walking ability of fast-growing broilers that decrease with age, reducing their mobility and willingness to move. Moreover, sick, lame and very heavy birds reduce their locomotor activity, and extend the time periods spent in resting and lying behaviour. Therefore, while current automatic weighing systems reduce time wasted by the farmer for manual weighing of birds, they may fail to continuously follow the growth trend of the whole flock, whilst simultaneously not estimating the weight of sick, lame and very heavy birds that are reluctant to move and to jump onto the automated scale. During the EU-PLF project (EU-PLF, 2016), the use of sound recordings collected with the commercially available SoundTalks® monitoring system, allowed to find a clear and inversely proportional relation between Peak Frequency of the sounds emitted by broilers and their weight as shown in Figure 6.

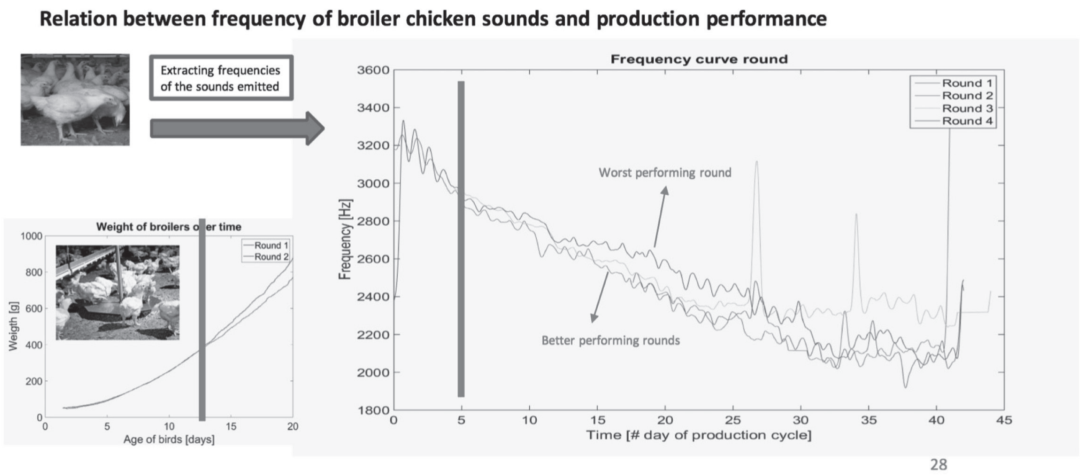

Figure 6. Link with the peak frequency extracted from the sound and the weight evolution of broilers (EU-PLF, 2016)

3.4. Out-of-comfort monitoring

Another challenge for the broiler farmers is to manage the house to stay in the optimal thermal comfort zone for the broilers. Broiler performance is heavily conditioned by environmental parameters such as temperature, relative humidity, air and litter quality and ventilation speed. Broilers are reared under different temperature and humidity ranges according to their age and a tightly controlled environment improves animal health, well-being, and production efficiency. To this end, temperature, relative humidity and ventilation rate are constantly controlled and managed by automated systems. Several investigators have reported that broilers can tolerate a wide range of relative humidity and still perform efficiently, but fast changes in relative humidity can rapidly and negatively influence litter conditions, that have been associated with lowered carcass quality and increased leg and foot abnormalities.

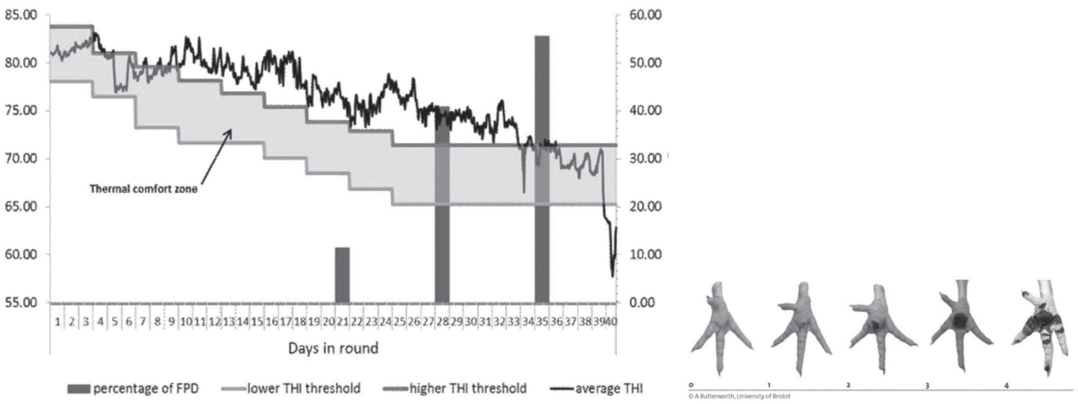

Figure 7. Relation between environmental variables and leg problems

Among leg disorders, footpad dermatitis is a significant welfare concern to the broiler chicken farming industry. Footpad dermatitis is characterised by skin lesions that can start from a discoloration of the skin finally turning into epidermal necrosis. These lesions may become a gateway for bacterial infections thereby affecting the bird's health and the walking ability of the birds, with the reduction of the animals welfare. Indeed, lame birds may also find difficult to reach food and water. Since the animal health strongly depends on good welfare, during the last years many progresses has been made in developing new indices and procedures to assess animal's health and welfare status. But, so far, the most commonly used outcome measure of living birds on a large scale is to observe individual birds walking and score them using a ranked scale. The Welfare Quality® protocol for boilers requires a lot of trained manpower/labour and it is timeconsuming and could potentially create biosecurity risks moving assessors between farms. In order to develop an automated prediction system to detect footpad dermatitis and lameness, data from the climate control system (FarmManager, Fancom) and data from manual welfare scoring where merged to find a correlation between climate and leg problems (Figure 12). The analysis shows that footpad dermatitis and lameness were strictly linked to the environmental conditions and that can be controlled with the automated system (Tullo et al., 2015).

4. Conclusions

PLF systems are becoming available in products and are getting operational in commercial farms. PLF devices can do much more monitoring than what any farmer or human observer can do and therefore add significant value to their livelihood. From examples presented we can see that value creation can be done in several ways: saving labour time, saving time in detecting problems, giving less stress to the farmer, solving problems on the spot immediately, and giving objective information about what happens to the animals and others.

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Managing On-Farm Health and Welfare Risks to Promote Sustainability in Poultry Industry

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Abbreviated title: Risks Management in Poultry Production

Summary: The poultry industry is a complex but well-organized system with a long history of pragmatic measures for the control of animal health and welfare risks. However, the sector is facing the impact of recent agricultural policies on the sustainability and multifunctional nature of production systems. Therefore, there is a need to critically analyze the sustainability of the poultry production systems, covering the economic, social and environmental aspects, including their adaptive capacity to socioeconomic and climate changes. The aim of this paper is to discuss how risk management as a methodology and mindset influence on priorities and decisions concerning sustainable development in poultry industry. Two models are presented to demonstrate how health and welfare risks can be managed to contribute to sustainable development in poultry production: a) management of *Campylobacter spp*. risks in broiler production (FAO/WHO, 2009; EFSA, 2011) and b) management of heat stress risks in laying hen production. In both cases, different scenarios are discussed to weight alternatives and select the most appropriate regulatory action, integrating the results of risk assessment with engineering data and with social, economic and environmental concerns to reach a decision. Finally, contradictions between risk management and sustainable development are questioned as they have a different point of departure, different perspectives on scope and time frames, different understandings about causes and consequences and different values defining their contents.

***Keywords**: poultry industry, sustainability, health and welfare risks, management*

Introduction

Poultry production systems are complex but well organized systems with a long history of realistic measures for the control of animal health and welfare risks. The sector, on the other hand, is facing the impact of recent agricultural policies on the sustainability and multifunctional nature of production systems and needs to comply with the new the EU's common agricultural policy (CAP) to structure and safeguard the stability of market prices in the sector, to facilitate the marketing of products (animal and environment friendly, disease free) and to establish the rules in the trade with third countries, providing stability for the producers and processors. In this direction, poultry industry should use a holistic approach of the risk management process, which will allow risk analysis and evaluation at strategic, operational, financial, environmental and social level. This integrating approach, the continuous and systematic analysis and evaluation of all risks shall represent a real fundament for the sustainable development of the management of the sector, thus generating an efficient management of all possible risks as well as the opportunities that may result. Furthermore, the poultry industry needs to fully integrate the concept of sustainable development and risk management into their strategy, not only to minimize potential losses, but also to fructify the new business opportunities which derive from the principle of sustainable development. These can include new products and services that can favor sustainable development, new technologies that can support and improve poultry farm sustainability and financial performance, or new business models in order to develop emerging markets and support the creation of sustainable communities. A recent comparative study in Germany and Thailand showed that social, animal welfare and economic issues dominate the current discussion of sustainable poultry production (Soisontes, 2017). In this context, there is a need to critically re-analyze the sustainability of the poultry production systems, covering the economic, social and environmental aspects, including their adaptive capacity to socioeconomic and climate changes.

The aim of this paper is to discuss how risk management as a methodology and mindset influence on priorities and decisions concerning sustainable development in poultry industry. The motivation for this review originates from the necessity of having a holistic approach for the risk management process in the sector, which will allow risk analysis and evaluation at a strategic, operational, financial, environmental and social level.

The Risk Management Process

In a rapidly changing world, businesses must regularly assess threats and opportunities as they strive to maintain a competitive edge. This analysis is referred to as risk management and it grows more and more important in poultry farming as the decision factors realize its multiple applications with supporting the industry's sustainable development in an increasingly pressing competitive environment (Mirela, 2012). Risk management is also potentially a tool for informing and implementing sustainability. Both risk and sustainability are multidimensional constructs which can be indicated in varied ways.

Traditional risk analysis has emerged as a structured model for improving food control systems, with the objectives of producing safer food, reducing the numbers of foodborne illnesses and facilitating domestic and international trade in food. Nowadays, this model is moving towards a sustainable approach to food safety, where the entire food chain together with environment and society needs to be considered in efforts to produce safer food (FAO/WHO, 2009).

In general, risk in poultry production is dependent on the existence of three factors: (1) a source from which risk agents are generated or released into the environment, (2) a route of human exposure to the risk agents, e.g., distribution and consumption of poultry products, and (3) a mechanism by which the exposures can generate adverse health effects, e.g., through microbial and chemical factors, which determine the health consequences resulting from human consumption or other contact with poultry products. A risk model may be developed by identifying and linking all significant influences on these three factors.

The risk analysis process includes hazard identification, risk assessment and risk management (Regulation (EC) 178/2002). The fourth component, risk communication, is used to inform farmers and workers of the need to implement appropriate biosecurity measures. Choice between qualitative, semi-qualitative or quantitative approaches should be made according to the purpose or the type of questions to be answered, and the data and resources available for a specific risk assessment. Quantitative data should be used whenever possible without diminishing the utility of available qualitative information and expert knowledge (EFSA Journal, 2012). In any case, good communication between risk assessors, risk managers, and all interested parties, is essential to the risk analysis process.

Risk Management of *Campylobacter spp.* in broiler production

Campylobacter spp. is well recognized as the leading cause of bacterial foodborne diarrheal disease worldwide and consists a serious economic and social concern with a direct impact on poultry flock productivity and welfare. The European Food Safety Authority (EFSA) has concluded that approximately 180,000 cases occur annually in the EU (EFSA, 2009). The reported incidence of Campylobacter infections has markedly increased in many developed countries within the last 20-year period.

Several risk factors can result in the introduction of *Campylobacter* into the flocks making it difficult to keep chicken flocks free of *Campylobacter* throughout the rearing period (Natsos *et al.*, 2016). Lack of biosecurity measures (Gibbens, *et al.*, 2001; Herman *et al.*, 2003, Cardinale *et al.*, 2004) , season (Bouwknegt *et al.*, 2004; Barrios *et al.*, 2006; Huneau-Salaün *et al.*, 2007; Zweifel *et al.*, 2008; McDowell *et al.*, 2008; Ellis-Iversen *et al.*, 2009; Jore *et al.*, 2010; EFSA, 2010; Lawes *et al.*, 2012; Chowdhury *et al.*, 2012) , age (Berndtson *et al.*, 1996; Evans and Sayers, 2000; Bouwknegt *et al.*, 2004; Barrios *et al.*, 2006; McDowell *et al.*, 2008; EFSA, 2010; Ansari- Lari *et al.*, 2011; Chowdhury *et al.*, 2012; Lawes *et al.*, 2012; Sommer *et al.*, 2013), partial depopulation practices (Hald *et al.*, 2000; Hald *et al.*, 2001; Slader *et al.*, 2002; Ellis-Iversen *et al.*, 2009; Hansson *et al.*, 2010; EFSA, 2010; Lawes et *al.*, 2012), flock size (Berndtson *et al.*, 1996; Barrios *et al.*, 2006; Guerin *et al.*, 2007; Nather *et al.*, 2009), type of production system (Van Der Zee *et al.*, 2005, Näther *et al.*, 2009), presence of other animals on farm (van de Giessen *et al.*, 1996; Bouwknegt *et al.*, 2004; Cardinale *et al.*, 2004; Lyngstad *et al.*, 2008; Ellis-Iversen *et al.*, 2009; Hansson *et al.*, 2010; Sommer *et al.*, 2013), water quality (Pearson *et al.*, 1993; Zimmer *et al.*, 2003), presence of rodents (Gregory *et al.*, 1997; Huneau-Salaün *et al.*, 2007; McDowell *et al.*, 2008; Sommer *et al.*, 2013), type of bedding material (Natsos *et al.*, 2017), contaminated surface water (Messens *et al.*, 2009), personnel and farm equipment (Ramabu *et al.*, 2004) and mechanical transmission via insects (Berndtson *et al.*, 1996; Refregier-Petton *et al.*, 2001) are considered to be some of the risk factors associated with horizontal transmission.

There is a need for the poultry industry to employ risk management strategies that predictably reduce Campylobacter levels in pre-harvest poultry and poultry carcasses. Ghareeb *et al.* (2013) reviewed current control measures to address risks of Campylobacter colonisation in poultry production. These include water treatment, hygienic and biosecurity measures, supplementing plant-derived additives to the feed, vaccination, passive immunisation, application of prebiotic, and probiotics (competitive exclusion microflora).

The Food and Agriculture Organization (FAO) and the World Health Organization (WHO) assembled an expert drafting group to develop a risk assessment framework for Campylobacter spp. in broiler chickens (FAO/WHO, 2009). The model developed includes the risk characterization component to complete the risk assessment.

Comparisons of risk for a variety of scenarios and mitigation measures were also conducted. The scenario changing 'between-flock prevalence', for instance, refers to the prevalence of contaminated flocks at the farm. Altering the between-flock prevalence could be achieved by the use of colonization-resistant breeds, if this were a technically feasible option, or through improved biosecurity where that is considered a key determinant of flock contamination.

In 2010, before deciding on risk management measures and setting performance objectives and/or targets, the Commission has requested EFSA to help with decision making process with the development of quantitative model to assess the public health benefit resulting from potential control options (EFSA, 2011). The model developed was able to evaluate quantitatively the effect of interventions scenarios on the risk of campylobacteriosis from broiler meat in EU Member States (MS). This uses probability mathematics to determine the distribution of microbial load on a bird or carcass through the various stages from shed to chilled carcass, together with the prevalence of *Campylobacter*-contaminated flocks and the within-flock prevalence for those contaminated flocks.

One of the success stories of *Campylobacter* risk management strategy, being in existence since 2006, can be found in New Zealand's Food Safety System (New Zealand Government, 2017). More than a 50% reduction in foodborne campylobacteriosis cases from 2007-2012 was the key achievement by the implementation of the above strategy. The future goal is to implement a *Campylabacter* risk management strategy as a systematic and inclusive manner that results in continuous reduction in the incidence of foodborne campylobacteriosis. Risk communication and stakeholder representation at all steps are also incorporated in the strategy to ensure understanding of the comprehensive risk management strategy and to share and obtain feedback on results from the work programme on an ongoing basis.

Risk Management of heat stress in laying hens

Understanding and controlling environmental conditions is crucial to successful poultry production and welfare. Heat stress is one of the most important environmental stressors challenging poultry production worldwide (Nienaber and Hahn, 2007; Daguir, 2008; Kadim, Al-Qamshui, 2008; Nardone *et al.*, 2010; Renaudeau *et al.*, 2012). Understanding and buffering environmental conditions is thus imperative to successful poultry production.

Typical conditions leading to heat stress occur where environmental temperatures are 26 ^{o}C and relative humidity is greater than 40%. As with other livestock, birds maintain a relatively constant internal temperature through physiological and behavioural thermoregulation. Physiological thermoregulation entails changes in metabolism to control body temperature. In contrast, behavioural thermoregulation involves changes in posture, orientation and/or environment in order to retain a constant body temperature. Several biotic factors such as a species' genetic potential, life stage and nutritional status, may determine the level of vulnerability to heat stress (Thornton *et al.*, 2009). According to Huey *et al.* (2012), species with high physiological capacities to buffer environmental variations and high potential for rapid evolution are better equipped to survive rapid warming. The level of tolerance to high environmental temperatures is also determined by the species' place of origin. Animals that have not been exposed to high temperatures are generally more vulnerable to heat stress than those that have adapted (acclimatized) over time (Sirohi & Michaelowa, 2007).

The detrimental effects of heat stress on laying hens range from reduced growth and egg production to decreased egg quality and safety (Mashaly *et al.*, 2004). Moreover, the negative impact of heat stress on poultry welfare has recently attracted increasing public awareness and concern (Lara and Rostagno, 2013). Many studies have been published about the effects of heat stress on the efficiency of broiler production. As previously seen, exposure of birds to high environmental temperature generates behavioral, physiological and immunological responses, which impose detrimental consequences to their productivity. Heat stress results in estimated total annual economic loss to the U.S. livestock production industry of $1.69 to $2.36 billion; from this total, $128 to $165 million occurs in the poultry industry (St-Pierre *et al.*, 2003). Therefore, managing risks for heat stress is strongly linked with the sustainability of the sector.

On the other hand, risk analysis methods have been recently sought to be applied to animal welfare as well (Müllller-Graf *et al.*, 2007). The use of a risk analysis approach to evaluate issues related to animal welfare can be useful to better identify and rank heat stress risk factors, and to prioritise possible management measures. The European Food Safety Authority (EFSA) has launched risk analysis studies in which animal welfare was considered as endpoint of interest (EFSA, 2012).

The main identified risks for hens' heat stress results from a negative balance between the net amount of energy flowing from the animal's body to its surrounding environment and the amount of heat energy produced by the animal. This imbalance may be caused by variations of a combination of environmental factors such as sunlight, thermal irradiation, and air temperature, humidity and movement but also, characteristics of the animal such as metabolism rate, and thermoregulatory mechanisms.

The intervention strategies to deal with heat stress conditions have been the focus of many published studies, which apply different approaches, including environmental management (such as facilities design, ventilation, sprinkling, shading, *etc.*), nutritional manipulation (*i.e.*, diet formulation according to the metabolic condition of the birds), as well as inclusion of feed additives in the diet (e.g., antioxidants, vitamins, minerals, probiotics, prebiotics, essential oils, *etc.*) and water supplementation with electrolytes (Sossidou, 2012). Nevertheless, effectiveness of most of the interventions has been variable or inconsistent. More recently, two innovative approaches have been explored, including early-life conditioning (*i.e.*, perinatal heat acclimation) and genetic selection of breeds with increased capacity of coping with heat stress conditions (*i.e.*, increased heat tolerance). However, these potential opportunities, although promising (particularly, for poultry production in hot climatic regions), still require further research and development.

Risk Management and Sustainable Development in poultry industry

Modern management needs to be risk-sensitive, to follow the implementation and utilization of reliable and efficient systems, to elaborate action plans and security schemes which include ranking the objectives on operational levels, adaptable to the permanent changes. An efficient management system does not limit itself to a 'short time horizon', also but considers further perspectives. In these situations, proactive management turns into prospective management, its purpose being to identify the risks that might arise as a consequence of strategy or environment modifications.

Today's poultry industry needs to fully absorb the concept of sustainable development and risk management into their strategy, not only to minimize potential losses but also to exploit new business opportunities which result from the principles of sustainable economic development.

Despite the fact that risk analysis has emerged as a structured model for improving animal health and welfare, new threats have to be addressed for the sustainability of poultry business as related to a rapidly changing environment and changes in society and economy (OECD, 1995). Increased complexity and rapid changes in modes of production produce ambiguity and uncertainty. Hence, business and technology management becomes more risky and vulnerable. Management of risks and hazards is based on past experience and often rely on partial analysis with a limited time frame and this may lead to a paradoxical situation where risk management and extended use of risk analysis could hamper long term sustainable development (Olsen *et al*., 2006). Taking the development and scientific foundation of risk analysis and risk management into account, it is likely to claim that there have been weak linkages between trends within risk management on one side, and sustainable development on the other side. Societal safety, defined as 'the ability of society as such to sustain important societal functions and to secure the life, health and basic needs of its citizens under different types of stress', could have been the linking pin between risk analysis and sustainable development.

Conclusions

Risk management and sustainability are closely linked. Today's poultry industry needs to completely absorb the concept of sustainable development and risk management into their strategy, not only to minimize potential losses but also to exploit new business opportunities which result from the principles of a sustainable economic, social and environmental development.

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Campylobacter infection compromises broiler health, welfare and performance

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Introduction

Campylobacter is the major, principally chicken-associated, zoonotic pathogen in the EU, infecting ~9 million people annually at an approximate cost to the economy, through impact on public health systems and lost productivity, of €2.4 billion (EFSA 2011).

Most chickens grown in the EU are reared in broiler systems where birds are housed usually at a final stocking density of 38Kg/M$^{2.}$ Modern, rapidly growing broiler breeds can reach slaughter weight (2.2Kg) ~36 days of age, although slower-growing breeds reared in 'high welfare' systems and stocked at a lower final density, usually 30Kg/M2, reach slaughter weight at around 50 days of age.

The modern broiler chicken is a very different animal from earlier breeds and has been bred selectively to increase weight gain, muscle mass and food conversion efficiency. A seminal paper on the changes in chicken because of selective breeding is by Zuidhof et al (2014). In this paper, the authors compared birds from 1957, 1978 and 2005. Using live weight as an indicator of change it was shown that at 28 days of age: the 1957 birds weighed 316g, the 1978 animals were 632g and the 2005 ones had a weight of 1396g. The 2005 birds were 440% heavier than their 1957 counterparts at this time point. Birds grown in 2018 are likely to be quite different from those reared in 2005.

It is important to better understand the animal and public health consequences of such changes. Chicken production should be sustainable, with a safe product and good bird welfare. The international poultry industry can find this difficult to always achieve and birds can experience poor welfare, often in the form of pododermatitis and hock marks, contact dermatitis of the feet and lower legs respectively. These problems relate mainly to poor gut health, manifesting as diarrhoea, causing wet litter, which ferments, becomes corrosive, resulting in the skin lesions. A variety of factors has been shown to affect the incidence of contact dermatitis, including the house environment, use of antibiotics and season and bird breed (Pagazaurtundua and Warriss, 2006, Kjaer *et al.*, 2006). Work in the UK in 2003-6 (Bull *et al.*, 2008, Rushton *et al.*, 2009) found marked flock-to-flock differences in incidences of health markers in UK commercial production systems but there was a clear link between bird health and welfare and *Campylobacter* infection.

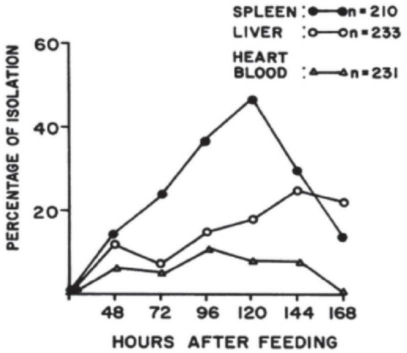
In vivo behaviour of *Campylobacter* in chickens and the public health threat

Despite much research, carriage rate in chickens and numbers of human cases remain high across much of the EU. The importance of chicken in human infection has resulted in many studies being undertaken over the last 30 years. Most work on chickens and *Campylobacter* has been from a public health perspective, largely with a focus on lowering levels in the gut to reduce numbers on carcass surfaces. Therefore, few studies on artificial infection have looked outside colonisation of caeca. Much of work on interventions to lower the perceived public health risk of surface-contaminated carcasses has been driven by a paper published in 2003, (Rosenquist *et al.*, 2003), which estimated that a two-log reduction in levels of *Campylobacter* on chicken carcasses would have a large public health benefit. However, this work may have underestimated the heat resistance of *Campylobacter* because relevant data to properly assess this were not available at that time and recent work has shown that some strains of *C. jejuni* can exhibit high heat resistance when attached to chicken muscle (De Jong et al 2012). For the past few years the UK Food Standards Agency (FSA) have undertaken a joint programme to reduce the % of chicken carcasses heavily contaminated with *Campylobacter*. Thus far this initiative has not resulted in any significant change in the number of human cases in the UK.

Tissue contamination is more important than that on the surface: There is long-standing and mounting evidence that the major health threat from *Campylobacter*-positive chickens is not from surface contamination (see above); edible tissues are also positive, almost exclusively with *C jejuni*, (Berndtson et al 1992; Whyte,Hudson and Graham, 2006; Scherer et al 2006; Luber and Bartelt, 2007, Hansson *et al.*, 2015) and contaminated chicken liver is a major vehicle for human infection (Edwards *et al.*, 2014), as is undercooked flesh. Contamination of chicken liver and muscle with *C. jejuni* is a public health threat that seems to be increasing but is not fully understood. Levels in livers can be high (Whyte, Hudson and Graham, 2006) and this can be associated with disease (Luber and Bartelt, 2007).

Routes of contamination of chicken tissues: The routes of contamination of chicken liver and muscle are yet to be fully explored and/or explained. The former may well be disease-associated, as *C. jejuni* can cause the disease Vibrionic Hepatitis in chickens and past work has found an association with the appearance of disease in liver and high numbers of *Campylobacter* in the tissues of this organ (Luber and Bartelt, 2007).

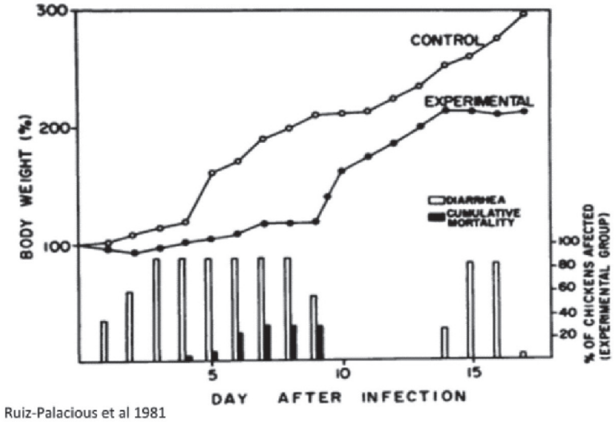
Fig 1: Damage to gut leads to spread to tissues



Sanyal et al 1984

Contamination of muscle tissues may be associated with septicaemia or bacteraemia in chickens. Figure 1 from a paper published over 30 years ago (Sanyal *et al.* 1984) shows that strains of *C. jejuni* can spread readily from the gut and in the study quoted, ~15% of infected birds had *Campylobacter* in their blood. More recent US work found that 12% of blood samples taken aseptically from naturally infected broilers at slaughter were *Campylobacter*-positive (Richardson *et al.*, 2011). It is possible that when the animals are bled at slaughter *Campylobacter* lodges in the small blood vessels in muscle tissues. More attention needs to be paid to preventing the contamination of edible tissues, which cannot be controlled in the slaughterhouse.

Fig 2: Weight gain and health in broilers given *C. jejuni*

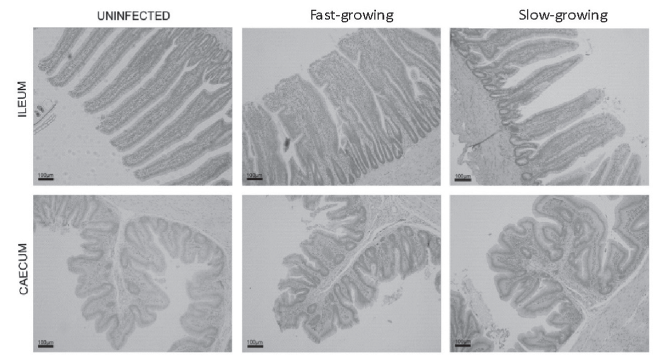


Ruiz-Palacious et al 1981

One of the problems common to most types of research is that when published work is quoted people focus on the results and pay rather less attention to the techniques used. This is particularly important when examining the infection dynamics of *Campylobacter* and chicken. In published work, studies used a variety of *Campylobacter* strains and different types of chicken, including both broilers and hens. It is important to recognise that the outcomes of infection and the *in vivo* behaviour of the bacteria, can be influenced strongly by bacterial strain, bird type and the environment they share. Thus, one *Campylobacter* strain may not represent the population of the bacterium and commonly used commercial broiler types show quite different innate immune responses to infection (Humphrey *et al.*, 2014).

***C. jejuni* damages broiler gut health:** There is strong evidence from studies over 30 years (Ruizpalacios,Escamilla and Torres, 1981, Sanyal *et al.*, 1984, Humphrey *et al.*, 2014, Awad *et al.*, 2014) showing that *Campylobacter* can have a negative impact on broiler gut health (Fig. 2; Ruizpalacios,Escamilla and Torres, 1981). The data in Figure 2 show that approximately 90% of the birds had diarrhoea, there was cumulative mortality of around 40% and the final body weight of the infected birds was around 30% lower than the controls.

Fig 3: In broilers *C. jejuni* M1 damages gut epithelia



Recent UK work (Humphrey *et al.*, 2014) supports the earlier studies on gut damage (Figure 3) and also shows that *Campylobacter* strains differ in impact on chickens (Chaloner *et al.*, 2014). This creates difficulties when studies are compared, given that broiler strains show differences in innate immune responses to *C jejuni* (Humphrey *et al.*, 2014). Recent work (Han *et al.*, 2016) has also shown that the innate immune responses of laying hens to *Campylobacter* differ from those of broilers.

It is important for academic engagement with the poultry industry, that work using artificial infections is supported by studies in commercial systems. Field work in the UK found significant relationships between broiler health/welfare and *Campylobacter* infection of commercial flocks. Neill *et al*. (1984) in Northern Ireland showed that infection of commercial flocks at any age was associated with wet litter, and in birds infected at <2 weeks old, there was raised mortality. Later UK work (Bull *et al.*, 2008, Rushton *et al.*, 2009) produced similar data with ~800 pre-thinned commercial flocks from 214 farms and three poultry companies. The companies made all health and production data for each flock available to the research team. Data were analysed using random effects logistic regression and/or generalised linear mixed models. The outcomes of this work were essentially in agreement with the earlier observations of Neill et al (1984) that flocks with high levels of contact dermatitis and/or condemnation for infection at slaughter (mainly avian pathogenic *E. coli*, APEC) were more likely to have *Campylobacter*. US work around the same time also showed a link with APEC (Russell et al 2003).

The field studies quoted above were not able to determine whether *Campylobacter* benefited from poor broiler gut health or if they caused it. The work from the 1980s, quoted above (Figures 1 and 2), suggest that the latter may be important. UK work compared effects of *C. jejuni* infection in 'fast' (2.2kg in ~35 days) and 'slow' broilers (2.2kg in ~50 days (Williams et al 2013). Birds were given the same diet and reared at the same density. Post-infection, 'fast' birds had wet litter and high levels of pododermatitis. 'Slow' birds had dry litter and little pododermatitis. It was not recorded if birds had diarrhoea but the available data suggest that they did. This was the first study to show that *C. jejuni* affects broiler types differently (Williams et al 2013).

Subsequent work (Humphrey *et al.*, 2014) using the same *C. jejuni* strain and bird types as the above study, found that 'fast-growing' chickens had severe diarrhoea whereas 'slow-growing' birds did not. There was much damage to gut mucosa with 'fast' birds, particularly in the ileum (Fig. 3). In 'slow' birds, with normal faeces, the villi were less affected (Fig. 3). The bacterial mechanisms for such effects were not determined and such work needs to be undertaken. Preliminary analyses of innate immune responses (Humphrey *et al.*, 2014) showed that gut damage in 'fast' birds was associated with dysregulation of pro-inflammatory cytokines. Work by another group, using 'fast' birds and a different *C. jejuni* strain, found similar damage to ileal mucosa but did not report diarrhoea (Awad *et al.*, 2014). Damage to gut mucosa has also been reported in some other studies (Naseri,Rahimi and Khaki, 2012, Ruizpalacios,Escamilla and Torres, 1981, Sanyal *et al.*, 1984), but not all (Naseri,Rahimi and Khaki, 2012). Awad *et al.*, (2015) found that infection compromised gut barrier function and performance and host nutrient uptake (Awad et al 2014), which is consistent with the damage to the ileum shown in Figure 3. Other studies report a range of impacts of infection with *C. jejuni*. As with Awad et al (2014) and Humphrey et al. (2014), other work has reported differences in whether birds suffered diarrhoea or not. Sanyal et al.(1984), Ruizpalacios et al. (1981) and Sang (1989) reported this, particularly in young chickens, but others did not. Other work found that infection of young birds (<3 days of age) with *C. jejuni* was lethal (Welkos, 1984) and this has also been seen in turkey and ostrich chicks (Lam *et al.*, 1992). The data indicate that bird type impacts on the effects of *Campylobacter* but bacterial strain is also important. These, and earlier data, show that *Campylobacter* can be chicken pathogens although strains behave differently. A working hypothesis could be that: "*Campylobacter* in chickens contain genotypes that are pathogenic (harmful) and others that are less so (i.e. benign to chickens)". It is possible that the 'harmful' strains are more likely to be 'invasive', leaving the chicken gut and infecting edible tissues in birds. *Campylobacter* colonisation starts with low-level invasion of gut mucosa (Young, Davis and DiRita, 2007). With some strains this may be more aggressive. There are now powerful economic and bird welfare reasons for better *Campylobacter* control on farm.

***Campylobacter* and commercial broiler performance**: Many factors affect broiler performance, including breeder flock age and stocking density (Sasaki *et al.*, 2014), season (Campe *et al.*, 2013), and light intensity (Olanrewaju *et al.*, 2014).. Good gut health in broilers is key to better growth rate and feed efficiency (Montagne,Pluske and Hampson, 2003). Given how *Campylobacter* can affect the gut (Fig. 3) it is no surprise that infected birds may have poorer performance. An Indian study (Singh *et al.*, 2012) compared broiler flocks with good and poor feed conversion ratios (FCRs). Using metagenomics to characterise gut microbiota, it was found that in birds with good performance (low FCR) the % of sequences assigned to *Campylobacter* was 0.05, whereas in poorly performing flocks (high FCR) it was 12. This means a highly significant 240-fold difference in the presence/levels of *Campylobacter* between the flock types. In 2014, with UK 'fast-growing' commercial flocks (>150 in total), the impact of *Campylobacter* on FCRs was examined. The average economic impact due only to *Campylobacter*-associated poorer FCR was high at ~£25 per 1,000 birds in infected flocks (Sparks; personal communication), even when all confounding factors had been removed and the differences between *Campylobacter*-positive and -negative flocks was highly significant ($p<0.01$). Recent work in Ireland found that high-performance birds (low FCRs) had both a lower *Campylobacter* prevalence and a higher gross income per bird. An important risk factor for high FCR was poor biosecurity and its impact, which could be *Campylobacter*-related, was ~€100 per thousand birds (Smith *et al.*, 2016b). Given the size of the EU industry, potential losses due to *Campylobacter*-related poorer FCRs alone could be €millions per year. There are also other costs like wastage from *Campylobacter*-related mortality and culling. Preliminary analysis of published experimental data on the financial impact of reported reduced bird performance, but not mortality, due to *Campylobacter* was, on average, ~£25 per 1,000 birds, although in Fig. 2, from a 1981 paper, it was greater (Ruizpalacios, Escamilla and Torres, 1981)

Campylobacter in commercial chickens comprise a diverse population and data show that some strains may pose a real risk to bird health and performance. Identifying the 'more chicken-pathogenic' strains and mechanisms by which they affect the birds is an important prerequisite for effective control and improving broiler welfare. Little is currently known about this, but clearly damage to gut mucosa and diarrhoea will affect nutrition and the former will also contribute to the spread of *Campylobacter* to edible tissues. Recent work found that nutrient transporter expression in the broiler gut was compromised by *C. jejuni* (Awad *et al.*, 2014). This may have a negative effect on performance and down-regulation of mRNA expression of glucose and amino acid transporters may result in nutrient accumulation in the intestinal lumen, favouring *C. jejuni* replication (Awad *et al.*, 2014). Increased gut damage and physiological disruption have clear implications for bird performance in a system with inherent low economic returns.

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Viral infections of poultry – the globally challenging situation

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Abbreviated title: Challenges of poultry viral diseases

Summary: Many challenges are facing the poultry industry, and viral infections are of a particular concern. Viruses can spread rapidly, they are not sensitive to antibiotics and cause devastating economic losses in poultry industries worldwide. Common markets and presence of international poultry companies provide for easier and faster spread of poultry viral infections to different countries, regions and even continents. In addition, recent spillovers of certain poultry-derived viruses into migratory bird populations have resulted in unpredictable outbreaks in remote poultry operations. The most important viral diseases representing major challenges to the poultry industry today are discussed here. None of such viral infections have been controlled to a satisfying extent at the global level so far, on the contrary more pathogenic or antigenic variant strains are increasingly emerging. Vaccination is possible for many viral infections of poultry, but it does not always warrant required protection. Undoubtedly, proper use of poultry vaccines through suitable vaccination programs has a precious impact on controlling and preventing viral infections and therefore in reducing economic losses. Nevertheless, poorly planned and/or improperly performed vaccinations can promote the emergence and enhance the transmission of virulent pathogens. Another issue is that vaccination cannot be a substitute for biosecurity and hygiene management, although there are such misconceptions. High competitiveness and demands for lower poultry production costs have altered the management system resulting in lowered emphasis in biosecurity and other basic disease control practices. Therefore, it can be concluded that management systems in poultry production that neglect fundamental disease control and eradication principles are in a great proportion responsible for emergence and spread of viral infections nowadays. All this makes viral infections of poultry a globally challenging situation and requires inclusion of specialized poultry veterinarians in disease consideration and decision making bodies already at the stage of planning new poultry operations.

Keywords: *avian influenza, HPAI, Newcastle disease, infectious bronchitis, infectious bursal disease, vaccination, biosecurity*

Introduction

There are many challenges facing poultry industry related either directly to the efficiency of poultry meat and egg production or indirectly through the poultry market issues, food safety and public health concerns, consumer perception, animal welfare, increasing legislative requirements and so on. Emergence and transmission of infectious diseases have an enormous impact on the poultry industry and pose a serious threat to the health of humans and wild birds (Wang *et al.*, 2013). Viral infections are of a particular concern. Viruses can spread rapidly, they are not sensitive to antibiotics and cause devastating economic losses in poultry industries worldwide. Vaccination is possible for many viral infections of poultry, but it does not always warrant required protection. In many cases, even when protected from a disease, vaccinated and apparently healthy poultry may carry the field virus and therefore play a role of a Trojan horse shedding it silently.

The chicken and turkey industries have been leaders in all agricultural industries toward larger units and corporate enterprise. Together with establishing of common international companies and markets, this results in easier and faster spread of poultry viral infections to more poultry farms, regions, countries and even continents. In addition, recent spillovers of certain poultry-derived viruses into migratory bird populations result in unpredictable outbreaks in remote poultry operations. Here we discuss most important viral diseases representing the major challenges to the poultry industry today.

Avian influenza

Avian influenza (AI) viruses are highly contagious, extremely variable viruses that are widespread in birds. Influenza viruses infecting poultry can be divided into 2 groups. The extremely virulent viruses cause highly pathogenic avian influenza (HPAI), with flock mortality as great as 100%. These viruses have been restricted to subtypes H5 and H7, although not all H5 and H7 viruses cause HPAI. All other viruses cause a milder, primarily respiratory, disease (LPAI) unless exacerbated (Capua and Alexander, 2009). Influenza viruses may infect all types of domestic or captive birds in all areas of the world. The frequency with which primary infections occur in any type of bird usually depends on the degree of contact there is with feral birds. Secondary spread is typically associated with human involvement, either by birds or bird product movement or by transferring infective faeces from infected to susceptible birds, but potentially wild birds can be involved (Alexander and Capua, 2008). Until recently, HPAI viruses were rarely isolated from wild birds, but the situation with HPAI of the H5 subtype dramatically changed in the last decade. The widespread presence of HPAI H5N1 viruses in poultry in the Far East inevitably resulted in spill-over into wild bird populations in May 2005 (Alexander and Capua, 2008) and there was strong evidence that wild birds could spread the virus long distances (Normile, 2006). This virus spread during 2005 and 2006 to a large number of European and African countries infecting poultry and wild bird populations (Salzberg *et al.*, 2007; Hesterberg *et al.*, 2009). A similar situation was repeated in 2014 when HPAI H5N8 virus spread by migratory wild birds from the Far East to Europe as well as North America (Lee *et al.*, 2015). The Far East origin HPAI H5N8 virus and its local reassortants caused the worst avian influenza outbreak in USA history, resulting in the destruction of more than 47 million chickens, turkeys and other commercially-raised birds with economic loss estimated to exceed $3 billion in direct and indirect costs in the US alone.

After the 2005-2006 H5N1 and the 2014-2015 H5N8 epidemics, another HPAI H5N8 virus introduced by migratory wild birds affected Europe. Up to August 2017, 1,112 outbreaks in domestic and 955 in wild birds in 30 European countries have been reported. This was the largest ever recorded epidemic by a HPAI virus in the continent in terms of number of poultry outbreaks, geographical extent and number of dead wild birds. (Brown *et al.*, 2017; Napp *et al.*, 2018). The virus remained circulating in poultry in Europe at few locations even in 2018. New introductions of HPAI viruses by migratory wild birds from remote areas and consequent infections of poultry are therefore to be expected in the future.

Although LPAI viruses cause a milder disease, infection of poultry with H9N2 virus resulted in vast losses during the last few decades. Since mid-1990 this infection was recorded in poultry in several European countries, South Africa, USA, South Korea and China, and the infection more recently became widespread and endemic in poultry throughout Asia, Middle East and North Africa (Capua and Alexander, 2009). Considering that there is no mandatory poultry surveillance of AI viruses other than H5 and H7 subtypes, there is currently a serious risk of introduction, spread and establishment of H9N2 virus into poultry in countries currently free of this infetion.

Vaccination against AI is possible and is nowadays practiced mostly against H9N2 virus, but vaccination against H5 and H7 subtypes is also not an exception, particularly in order to combat against HPAI viruses. Nevertheless, vaccination against AI is a complex issue due to high variability of influenza subtypes and even significant antigenic differences among AI viruses of a single subtype. This may result in poor efficacy of even properly used vaccines of the same subtype as the circulating virus. Optimal vaccination with AI vaccines when selected properly and administered correctly will protect against clinical signs and mortality, reduce the levels and duration of virus excretion and increase the resistance of the host to infection by raising the minimum infectious virus dose needed to infect the bird (Capua *et al.*, 2004). However, it is possible that suboptimal vaccination may result in the reduction of disease without affecting transmission, resulting in an endemic situation (Webster *et al.*, 2006), particularly if there is scarce or no practice of other measures for controlling AI. For this reason, vaccination should only be part of a wider control strategy, which must include improved biosecurity and the detection of field-exposed flocks within the vaccinated population (Capua and Marangon, 2007).

Certain AI viruses posses significant zoonotic capacity, although none of them has adapted to humans and there is no efficient human-to-human transmission so far. For most of people who contracted AI virus infection there was evidence of direct contact with infected poultry, mainly in regions with endemically infected poultry, while transmission to humans through poultry meat or eggs has never been confirmed. Nevertheless, consumer perception can be often the opposite. For example, detection of zoonotic H5N1 HPAI virus in 2005/2006 in Croatia exclusively in wild birds had a significant impact on the local poultry industry due to the fear of infection and consequently decreased poultry meat consumption up to 20% at the peak of the crisis (Savić *et al.*, 2007).

Newcastle disease

Newcastle disease (ND) is another devastating disease of poultry causing mortality up to 100% in fully susceptible chicken flocks. In contrast to HPAI hardly a single commercial flock of poultry is reared that, if not infected, is not influenced in some way by measures aimed at controlling ND and spread of the virus. In many areas where poultry are reared commercially reliance is placed on vaccination to keep ND in check, but ND nevertheless represents a major limiting factor for increasing poultry production in many countries (Alexander, 2001). Like for HPAI, economic importance of ND is not only due to direct losses in infected flocks and cost of vaccination, but also the impact that may ensue due to trading restrictions and embargoes placed on areas and countries where outbreaks have occurred (Aldous and Alexander, 2001). Although these embargoes are generally justifiable, in many instances they are not driven by epidemiological reasons, but present a political decision of importing country. We can bear witness to

situations where a country with endemic ND places ban on import of poultry and poultry products from a country with a limited ND outbreak which was promptly contained by stamping out and compartmentalisation.

The disease is continuously present in many countries around the world, in some of them causing sporadic outbreaks or sudden epidemics while in other countries being endemic. ND is, among several other viral diseases of poultry, notifiable to the World Organization of Animal Health (Office International des Epizooties - OIE). Only two poultry viral diseases, HPAI and ND, were listed on the former OIE list A which represented transmissible diseases of major importance in the international trade of animals and animal products. Nevertheless, ND seems to be highly underreported to OIE, since some countries are formally free from this infection while research articles in respectable journals describing serious and continuous ND outbreaks in those countries can be found. Lack of diagnostic capacity and/or monitoring of this infection can be also an issue in underreporting to the international organisations. As it was mentioned for AI, virulent ND viruses may infect and replicate in vaccinated birds without causing clinical disease (Parede *et al.*, 1990; Capua *et al.*, 1993; Guittet *et al.*, 1993) and this may lead to endemicity and also masking of the infection and significantly hindering passive monitoring that relies on clinical symptoms and mortality of infected poultry.

Unlike AI viruses, ND viruses are antigenically unique representing a single serotype termed avian avulavirus 1 (AAvV-1, formerly avian paramyxovirus 1). This makes vaccination much more simple and straightforward since there is practically no antigenic mismatch between field virus and ND vaccines. Live and inactivated vaccines against ND have been widely used since the 1950's. When administered correctly to healthy birds, live or viral-vectored vaccines are able to prevent clinical disease and mortality in chickens upon infection with virulent NDV. In contrast to vaccines against AI, availability of live vaccines against ND allows mass administration in poultry premises which reduces labour and cost. It should be mentioned that live ND vaccines vary in their tropism and capacity to replicate in naïve chickens and therefore the vaccine choice should fit local circumstances. ND vaccines and vaccination have been recently reviewed by Dimitrov *et al.* (2017).

Infectious bronchitis

Infectious bronchitis (IB), also known as avian IB, is an acute and highly contagious viral disease that affects chickens of all ages. It is ubiquitous in most parts of the world where poultry are reared and is considered the most contagious of poultry diseases. The morbidity usually reaches 100%, but mortality is low reaching as high as 25% or more in chickens less than six weeks of age and usually is insignificant in older chickens (Cavanagh and Gelb, 2008). Despite low mortality, the economic importance of IB most likely surpasses that of ND. Survey of 176 countries included in the OIE Animal Health Yearbooks from 2006 to 2009 determined IB as a poultry disease causing second greatest losses, behind HPAI and followed by LPAI, ND and infectious bursal disease (IBD) (Anonymous, 2011).

Despite its suggestive name, IB assumes a variety of clinical forms along with respiratory disease. Infection of the oviduct at a very young age can result in permanent damage while in hens usually causes drop or cessation of egg-laying as well as production of thin-walled and misshapen shells with loss of shell pigmentation. Certain IB strains can severely damage kidneys and cause acute nephritis, urolithiasis and mortality, especially in young chickens (Jackwood and de Wit, 2013).

Another important issue is that IB virus exists in the form of many different antigenic or genotypic types i.e. serotypes or genotypes, commonly referred to as variants. The virus has an enormous capacity to change both by spontaneous mutation and by genetic recombination (Cavanagh and Gelb, 2008). Such events may result in the emergence of new variants of which most eventually disappear, some become endemic or have only limited importance while only few become dominant i.e. widespread in larger regions, even continents. These new variants were usually arbitrarily named by researches using full or abbreviated name of geographical location or year of isolation, sometimes combined with ordinal numbers, and so on. As a result there are several serotypes/genotypes that were variously termed by different researchers, making a lot of confusion. For example designations LX4, D388, A2, Korean-II (K-II), Japanese-III (JP-III) and QX-like refer to a single IB virus variant probably most commonly known just as QX. Another good example is where designation 4/91, 793B or CR88 is used for the same variant. A recent attempt to harmonize IB virus variant classification based on genotyping included six genotypes (or G's) designated using Roman numerals (GI to GVI) which all together comprise 32 distinct lineages designated by Arabic numerals (Valastro *et al.*, 2016). According to this classification, variant QX for example should be termed GI-19, while designation GI-13 refers to variant 4/91. In spite of this systematic classification and numerical designation, the older arbitrary designations for IB virus variants are still preferred (de Witt *et al.*, 2018).

Strict biosecurity and one-age farm system are essential control measures, but vaccination is normally an inevitable tool to increase the resistance of the chickens against challenge with field IB virus. Live attenuated and inactivated vaccines are available, but since there are numerous antigenic variants of IB virus, the vaccination is not nearly as straightforward as it is in the case of ND. Vaccines of a certain IB virus serotype or genotype are normally able to protect a well-vaccinated chicken against a homologous challenge. Often there is a partial protection against strains of other serotypes or genotypes that can vary from high to low (de Wit *et al.*, 2011). Today, vaccines of various serotypes/genotypes are commercially available, but not of all currently circulating serotypes/genotypes. This is actually not necessary because certain IB vaccine strains provide high cross-protection against certain heterologous serotypes or genotypes (Bijlenga *et al.*, 2004). Therefore, beside classification into serotypes and genotypes, strains that induce protection against each other in chickens are placed into the protectotype (de Wit *et al.*, 2011). It has been also demonstrated in several studies that vaccination with two antigenically distinct live attenuated vaccines can result in a broad cross-protection against many different IB virus variants. This means that tools for successful vaccination against most of IB virus variants are available, such as planning vaccination by use of protectotypes as well as combination of antigenically distinct vaccines. Nevertheless, it is crucial how to use these tools i.e. how to design efficient IB vaccination program for a given epidemiological situation, otherwise even abundant vaccination programs may fail. Knowing exact epidemiological situation requires systematic and continuous serological and/or virological monitoring of chicken flocks as well as accurate diagnosis in order to discriminate used vaccinal strains from circulating field IB viruses (Savić, 2017).

Infectious bursal disease (Gumboro disease)

Infection with infectious bursal disease (IBD) virus causes clinical disease only in chickens younger than 10 weeks, while older birds usually show no clinical signs.

Turkeys, ducks, guinea fowl, pheasants and ostriches may be infected, but the clinical disease is absent in these species (Eterradossi and Saif, 2013). Although the problems caused by IBD are restricted to a single poultry species and a short age span, this disease continues to be among leading causes of economic losses in the poultry industry mainly due to the large number of broiler chickens which fall into this host category during their entire life span. Historically, IBD was causing mainly subclinical disease in broilers with insignificant mortality but with immunosupression leading to mainly indirect losses due to secondary infections, growth retardation and condemnation of carcasses at the abattoir. The situation drastically changed in the mid 1980s when highly virulent strains were detected in Western Europe causing mortality of 5 to 15% with occasional extremes of 25% in broilers and up to 60% mortality in pullets (van den Berg *et al.*, 1991). These highly virulent strains, also referred to as very virulent IBD (vvIBD) strains, have rapidly spread all over the world, causing dramatic losses (van den Berg, 2000). The infection with vvIBD viruses could be easily recognised in the field due to the acute form of the disease, typical gross lesions and notably increased mortality, leaving impression that vvIBD viruses entirely replaced classical strains which therefore became neglected. Nevertheless, recent studies have shown presence of non-vvIBDV field strains in Europe which can easily be undetected due to the focus on sampling of clinically ill birds (de Wit *et al.*, 2018). Therefore, beside strict biosecurity and hygiene management, vaccination is a key method for preventing not only clinical disease and mortality, but also for reducing economic losses due to possibly undetected subclinical IBD infections (McIlroy *et al.*, 1992). Similarly to ND, IBD viruses that infect chickens belong to a single serotype, yet vaccination against IBD is not that straightforward. The main reason is very strong interference of maternally derived antibodies (MDA) with both live IBD vaccines and field IBD strains which means that vaccination with live vaccines in presence of IBD specific MDA may be partially or completely inefficient. Chickens passively acquire MDA from their parents and these antibodies start to decay soon after hatching and last for just a few weeks, depending primarily on their initial level and type of the bird. As chickens are most susceptible to IBD virus in their first weeks of life, active immunity to the virus has to be induced early after hatching (Müller *et al.*, 2012). Live IBD vaccines show a different degree of attenuation. Generally, more attenuated vaccines are neutralised by lower antibody levels while more virulent strains can break through higher antibody levels. As a consequence, chickens are sooner susceptible to infection with field virus than they are susceptible to attenuated IBD vaccines. The aim of vaccination with live IBD vaccines should be to vaccinate chickens as early as possible but late enough to avoid neutralisation of vaccine virus by MDA. Therefore, in flocks with significantly uneven MDA levels, two vaccinations may be required, one for birds with initially lower MDA levels and the other for those with initially higher MDA levels. Nevertheless, there is a timeframe when chickens will become susceptible to field infection, particularly with vvIBD viruses, but not yet ready for vaccination with live vaccines. This sensitive period is even longer since it takes several days for developing active immunity to vaccination. It can be shortened by use of less attenuated live vaccines, like "intermediate" and "intermediate plus (hot)" vaccines which are capable to break through higher MDA levels and therefore are suitable for application in earlier age then "mild" vaccines. "Intermediate" and "intermediate plus" vaccines also induce higher immune response after vaccination, but increased efficacy of live IBD vaccines is chiefly in correlation with decreased safety. This means that "intermediate plus" vaccines in particular can induce immunosuppression comparable to that caused by classical field IBD strains. Recently, live viral vector IBD vaccines comprising remarkable combination of safety and efficacy are available. They are suitable for *in ovo* vaccination at day 18 of incubation or at one-day old chickens in the hatchery regardless of the MDA presence. Immune complex vaccines can be applied in the same manner, although these two types of vaccines are quite different in their fundamental biological properties. Such advanced vaccines are generally more expensive and have to be applied by injection individually to each chicken in contrast to conventional live IBD vaccines which are suitable for mass vaccination in the farm.

Discussion and conclusions

In addition to the above mentioned viral infections of poultry, several others constantly pose challenge to the poultry industry, depending on the poultry species affected, type of birds, age and somewhat on the geographical region. These are, for example, infectious laryngotracheitis, turkey rhinotracheitis, duck virus hepatitis, fowl pox, Marek's disease and others. Although only few most important poultry viral infections were discussed here individually, they all share some common characteristics and issues. None of these diseases have been controlled in satisfying extent at the global level so far, on the contrary more pathogenic or antigenic variant strains are increasingly emerging. Development of new vaccines effective against emerging strains is an ongoing activity while the use of available vaccines has been widely practiced in the poultry industry for decades. Undoubtedly, proper use of poultry vaccines through suitable vaccination programs has a precious impact on controlling and preventing viral infections and therefore in reducing economic losses. However, imperfect vaccination can enhance the transmission of virulent pathogens while vaccines that do not prevent transmission can create conditions that promote the emergence of pathogen strains that cause more severe disease in unvaccinated hosts (Read *et al.*, 2015). Even some live attenuated vaccines themselves may revert to higher levels of virulence following bird-to-bird passage and then can cause severe disease in unvaccinated contacts (Coppo *et al.*, 2013). Therefore, poorly planned and/or improperly performed vaccinations present a double-edged sword with unwanted consequences that will not necessarily occur in short terms but definitively will occur in long terms. Another issue is that vaccination cannot be a substitute for biosecurity and hygiene management, although there are such misconceptions. High competitiveness and demands for lower costs of poultry production have altered the management system resulting in lowered emphasis in biosecurity and other basic disease control practices. Many new and modern farms are built in a manner of multi-flock complexes with no possibility of all-in, all-out and neither of one-age farm practices. Instead, continuous production in such multi-age farms enables continuous circulation of viral pathogens from older to younger flocks. Abundant bird-to-bird passage of field virus in such complexes favours emergence of new antigenic variants or more pathogenic strains. It is also very difficult, if not impossible, to design efficient and safe vaccination program in such farms, particularly if the infection is already present at the farm. Thinning of broiler flocks in order to increase profit is another practice that largely contributes to spread of viral infections among flocks and farms.

It can be concluded that management systems in poultry production that neglect fundamental disease control and eradication principles are in a great proportion responsible for emergence and spread of viral infections nowadays. Even in cases of massive HPAI outbreaks in poultry in USA and Europe that occurred after virus introductions by migratory wild birds we should bear in mind that the virus was poultry-derived, although it emerged on the other side of the globe. Also, presence of international companies and common markets provide for easier and faster spread of viral infections between widespread locations. All this makes viral infections of poultry a globally challenging situation jeopardizing poultry locally and requires inclusion of specialized poultry veterinarians in disease consideration and decision making bodies already at the stage of planning new or reconstructing existing poultry operations.

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Metagenomic insights into the dynamics of microbial communities in poultry and poultry products: current challenges and future opportunities

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Abbreviated title: Metagenomics applications in poultry science

Summary: The poultry gut microbiome represents gut microorganisms and genomes as well as genes belonging to those microorganisms. Since the gut microbiome is involved in the regulation of multiple host metabolic pathways, a deep understanding of the relationships between gut microbiome and host should provide new strategies to improve poultry health and productivity as well as poultry meat safety. Using metagenomic sequencing it is possible to investigate such relationships, testing whole populations of microorganisms in their natural habitats and in the context of their interrelationships with the host. In this review, metagenomics techniques currently available are summarised, as well as possible approaches to data analysis. Furthermore, selected metagenomic studies showing how to explore chicken gut taxonomic and gene compositions, using targeted and shotgun sequencing, are presented. Finally, in field metagenomics applications to drive improvement of poultry feed efficiency, map variations in antibiotic resistance genes in poultry productions and characterise the microbiota colonising poultry carcasses are shown, along with key technical issues to address in order to support metagenomics researches in poultry science.

Keywords: *Metagenomics, next generation sequencing, microbiota, functional genes, feed efficiency, antibiotic resistant genes, meat safety.*

Introduction to metagenomics

Metagenomics refers to a non-culture based approach providing insight into the overall functional repertoire of a microbial community, including information on its metabolic capabilities and potential functional interactions among its members (Chistoserdova, 2009). Metagenomics has become an emerging field of microbial ecology since the first use of the concept in 1998 in relation to soil microorganisms (Handelsman *et al.*, 1998). In the poultry framework, metagenomics is the study of the metagenome, corresponding to the total DNA that can be extracted from a specific site within a bird, frequently represented by the gastrointestinal tract. The collective microbial community inhabiting the investigated site is named microbiota, whereas the term microbiome refers to the collective genomic content of the microbiota (Tremaroli and Bäckhed, 2012).

Originally, metagenomics studies depended on the cultivation of clonal cultures, followed by functional expression screening (Handelsman *et al.*, 1998). However, such cultures are not able to represent the total community profile and may overlook the vast majority of the microbial biodiversity. Thus, natural microbial communities typically contain a wide diversity of organisms, viruses and other chromosomal and extra-chromosomal genetic elements.

The advent of next-generation sequencing (NGS) technologies has made it possible to perform sequencing-based metagenomic analyses. Next-generation sequencing, also defined as massively parallel sequencing, is a sequencing technology, which overcome the limited scalability of traditional Sanger sequencing, allowing for millions of sequencing reactions to happen in parallel. NGS platforms allow researchers to retrieve high-throughput data at a reasonable cost, fuelling the development of sequence-based metagenomic analyses aimed at decoding entire microbial communities (Thomas *et al.*, 2012).

NGS technologies can be divided into several types, based on the sequencing process used. They are typically represented by Ion PGM, Ion Proton and Ion SS or SSXL by ThermoFisher; MiSeq, NextSeq, HiSeq (2500) and HiSeq X Ten by Illumina (Vincent *et al.*, 2017); HeliScope by Helicos BioSciences; PacBio RS by Pacific BioSciences (Mohinudeen *et al.*, 2017). The combination of read length and number of reads defines the throughput of each instrument in number of bases per run. Liu *et al.*, (2012) reviewed these systems, summarising the first-hand data analysis to discuss the advantages and specifics associated with each sequencing system.

Besides technical differences, the goal for any metagenomic project is to understand the following issues: 1) community composition/structure, including taxonomic breakdown and relative abundance of the various species; 2) genic contribution of each member of the community, including number and functional capacity; 3) intra-species and/or intra-population gene heterogeneity (Scholz *et al.*, 2012).

Targeted and shotgun metagenomics

Two distinct metagenomics approaches are commonly used: the first is referred to as marker-gene metagenomics or targeted metagenomics, the second as shotgun metagenomics. To assess the taxonomic composition of a sample, targeted metagenomics is straightforward (Chistoserdova, 2016). One of the most general target for sequencing is the hypervariable region in genes encoding 16S rRNA. Bacterial 16S rRNA gene possesses nine hypervariable regions, flanked by highly conserved regions, which are generally designated as polymerase chain reaction (PCR) primer sites. Sequence variations in hypervariable regions allow accurate bacterial taxonomic estimation by comparing against 16S rRNA gene sequences deposited in public databases, such as GreenGenes, the Ribosomal Database Project, SILVA, etc (Choi *et al.*, 2015). The overall sequencing output of the 16S rRNA is a cluster of nearly identical sequences, referred to operational taxonomic units (OTUs) (Cole *et al.*, 2014). The representative sequences, which are either the most abundant sequences or sequence with the least distance summation within all sequences in the same OTUs, are then matched to those in a public database to obtain taxonomic classification. In addition to providing taxonomic information, the OTUs provide information on population diversity, indicating richness and evenness of individual species in a sample as alpha diversity (Colwell, 2009). This information is also used to account for the degree of divergence between different communities or sample types.

Besides taxonomic composition, the more difficult question asked via metagenomics concerns the genic contribution of each member of the investigated community, including number and functional capacity, as well as intra-species or intra-population heterogeneity of the genes (Scholz *et al.*, 2012). To address these issues shotgun metagenomic sequencing is the most suitable approach. Shotgun sequencing is the process of randomly breaking (often by shearing) a long DNA molecule (for example, a complete chromosome) and then sequencing the resultant DNA fragments, which each come from a different location in the original long DNA molecule (Weinstock, 2012). Shotgun sequence data provide information on the organisms that make up communities but also information on functional genes in the sample. The challenge for metagenomic sequencing is now complete the framework associating each specific microorganism with its own gene set.

Identification and assembly of genomes and genetic elements in complex metagenomic samples

At present, thousands of marker gene assisted and shotgun metagenomics projects have been undertaken, comprising millions of samples available in the public domain. After sequencing of a metagenomics sample, the primary task is to analyse the vast amount of data. The simplest way to analyse the metagenome is by using short reads directly after the quality sequence-filtering step (Thomas *et al.*, 2012). These short reads can be used as taxonomic markers and matched against a marker gene database (e.g. Phylosift, EggNOG) and functional markers, quantifying abundance of genetic pathways (Darling *et al.*, 2014; Powell *et al.*, 2014). These analyses provide taxonomic (e.g. MEGAN) and functional (e.g. KEGG pathway) contents of single data sets to rapidly search for specific targets and test hypothesis generated from an experimental design. Alternatively, assembly algorithms can be used to reconstruct short reads into a sequence contig, which is a set of overlapping sequences representing the contiguous DNA fragment (Mende *et al.*, 2012; Thomas *et al.*, 2012). Although obtaining a complete individual genome from metagenomic sequences is still challenging, data collected are sufficient to characterize the major functions of the microbial communities as well as to identify their taxon, by assigning to public genome reference databases (Howe *et al.*, 2014; Nielsen *et al.*, 2014).

Even though current computational analysis strategies for metagenomic data rely largely on comparisons to reference genomes, they represent only a fraction of the species and viruses present. Moreover, bacterial genomes from different isolates of the same species usually show considerable genetic heterogeneity, when compared (Nielsen *et al.*, 2014). This variation may be the result of clonal differences, environmental adaptation or possibly artefacts from the cultivation process (Nielsen *et al.*, 2014). Therefore, reference genomes represent only a small proportion of the biological diversity of microbial systems, and thus methods relying on them place limitations on structuring and analysing metagenomic data (Nielsen *et al.*, 2014). In particular, they limit our ability to segregate metagenomic data into coherent biological entities and fail to describe previously unknown species, phages and modules of genetic variation within microbial species (Nielsen *et al.*, 2014). *De novo* assembly of genomes from complex metagenomic data is inherently difficult due to the many sequence ambiguities that confuse the assembly process. Hence, a typical metagenomic assembly will result in a large set of independent contigs that are not easily aggregated into biological entities (Wooley et al., 2010). Proper structuring of the complete metagenomic composition is important not only for understanding the microbial communities (Raes et al., 2008), but also for making statistical associations between the metagenomic data and descriptors of the system (Nielsen *et al.*, 2014).

Metagenomic sequencing to investigate the chicken gut taxonomic composition

The poultry gut microbiota constitutes a complex ecosystem composed by a large variety of microorganisms. It plays an important role in maintaining the host normal gut functions and health, and its imbalance, or dysbiosis, can produce negative effects on gut physiology (De Cesare *et al.*, 2017). Signs of dysbiosis in broilers are thinning of the small intestine, increased water content and presence of indigested residues in the faces (De Cesare *et al.*, 2017). The chicken GIT harbours a very diverse microbiota that aids in the breakdown and digestion of food and comprised over 900 species of bacteria (Apajalahti *et al.*, 2004; Wei *et al.*, 2013). It is estimated that only 10–60 % of caeca bacteria can be cultured (Stanley *et al.*, 2014) and that only around 45 % of chicken intestinal bacteria can be confidently assigned to a known genus (Apajalahti *et al.*, 2004). Therefore, much of the GIT microbiota remains largely unexplored making it a major source of untapped biological potential in terms of newly identified bacteria, encoded enzyme activities and potentially probiotic bacterial strains (Stanley *et al.*, 2014).

The bacterial communities originating from different sections of the chicken GIT are so different that it has been suggested that they should be considered as separate ecosystems (Wielen *et al.*, 2002). They are, however, highly connected and seed and influence microbiota both up and downstream in the GIT (Stanley *et al.*, 2014). A detailed overview of the chicken indigenous bacteria for each gut section is given in the review by Rehman *et al.*, (2007). In mammals, the caecum has a negligible role in digestion; however, in birds, the caeca are an important site of fermentation (Mead 1989; Clench and Mathias, 1995), influencing animal health and performance; therefore, caeca microbiota profiles are widely investigated. Caeca microbiota has the ability to digest foods rich in cellulose, starch and resistant polysaccharides (Clench and Mathias, 1995). Moreover, caeca are a major site of water absorption and nutrient transport and absorption (Stanley *et al.*, 2014).

De Cesare *et al.*, 2017 investigated the caeca microbiota composition of one-day old chicks and more than 95% of bacterial population was represented by Firmicutes (85.5%) and Proteobacteria (9.61%). Both these phyla were largely represented also in the caeca of birds reared for 41 days. However, the relative frequency of abundance of Firmicutes in one-day old chicks was significantly lower than that observed at 41 days ($P=0.01$), whereas that of Proteobacteria was significantly higher ($P=0.0067$). Within Firmicutes, in one-day old chicks Bacilli was the most abundant class, followed by Clostridia. On the contrary, at 41 days, Clostridia represented the most abundant class (i.e., 70.5%) followed by Bacilli (i.e., 20.7%). The mean relative abundances of Clostridia and Bacilli in the birds at the end of the rearing period were significantly higher ($P=0.0086$) and lower ($P=0.0094$) in comparison to those of one-day old chicks, respectively. At both sampling times, Gammaproteobacteria was the most representative class of Proteobacteria and it was significantly higher in one-day old chicks in comparison to 41 days of age ($P=0.015$). Moreover, in one-day old chicks Enterobacteriaceae was the most represented family (7.63%) in comparison to the birds at 41 days, where the relative abundance of the same family was as low as 0.77%. Within Bacilli class, the most represented family in one-day old chicks was Lactobacillaceae (33.5%), followed by Enterococcaceae (3.72%), Streptococcaceae (1.79%) and Bacillaceae (1.17%). This distribution was similar at 41 days, except for Bacillaceae representing the second most abundant family 41 days. In comparison to one-day old chicks, Lactobacillaceae, Enterococcaceae and Streptococcaceae decreased significantly (i.e., P=0.023, P=0.042 and P=0.01, respectively) at 41 days. Lachanospiraceae was the most represented family identified within the Clostridia class in one-day old chicks (13.25%). On the contrary, at 41 days Ruminococcaceae was the most represented family (29.53%) and showed a relative frequency of abundance significantly higher than in one-day old chicks (P=0.00044). Overall, among the first 30 bacterial species identified in one-day old chicks the most represented were *Lactobacillus johnsonii, Lactobacillus crispatus, Escherichia coli, Ruminococcus torques, Lactobacillus helveticus, Lactobacillus gasseri, Ruminococcus obeum, Ruminococcaceae bacterium* D16, *Clostrifium hylemonae* and *Eubacterium limosum.* At 41 days, the most represented species were *Faecalibacterium prausnitzii, Lactobacillus crispatus*, *Ruminococcus torques, Subdoligranulum variabile*, *Ruminococcaceae bacterium* D16, *Lactobacillus johnsonii, Pseudoflavonifractor capillosus, Ruminococcus obeum, Clostridium difficile* and *Blautia hydrogenotrophica. Ruminococcus lactaris* was significantly higher in one-day old chicks in comparison to 41 days, whereas *Faecalibacterium prausnitzii* and *Subdoligranulum variabile* showed a significantly higher relative frequency of abundance at 41 days in comparison to one-day old chicks.

Metagenomic sequencing to assess the chicken gut gene composition

Metagenomic investigations allow clarifying the interconnections between metabolic pathways resulting from the interactions within the microbiota and between the microbiota and the host, analysing such interactions *in vivo*. Using metagenomic analysis it is possible to investigate which bacteria have specific genes coding for enzymes associated to metabolic pathways. Sergeant *et al*., (2014) predicted 108103 complete coding sequences (CDS) in chickens. Among these sequences, the authors found sequences from over 200 different non-starch polysaccharide-degradating enzymes, with glucanases acting on oligosaccharides predominate over endoglucanases acting on full-length polymers of cellulose and xylan. In Bacteroidetes the authors found evidence of over 500 polysaccharide utilization systems including representatives of a novel class of such systems that incorporates β 1-4 endoglucanases and β 1-4 endoxylanases. In Megamonas, the same authors found genetic evidence of co-ordination of polysaccharide degradation with sugar transport and utilization. Finally, in Clostridales they found two endoglucanase genes associated with sporulation genes, leading to speculated that such genes may be implicated in breakdown of the cell wall or formation of the spore cortex.

De Cesare et al., 2017 investigated the gene composition in the caeca of chickens tested at 1 and 41 days of age. The data showed the mean relative abundance of the KEGG pathways related to metabolism and genetic information processing in one-day old chicks corresponded to 20.9 and 16.2%, respectively. These values were significantly lower than those detected at 41 days (i.e., 53.8 vs 55.8% respectively). On the contrary, the environmental information processing and cellular processes pathways were significantly higher in one-day old chicks. In relation to specific metabolism pathways, one-day old chickens showed relative frequencies of abundance of aminoacid and carbohydrate metabolisms significantly lower (4.54 and 3.55%, $P<0.001$) than those detected at 41 days (i.e., 16.2 and 16.1%, respectively).

These examples show the potential application of metagenomic analysis to check gene settings in the chicken gut in order to verify their distribution and how the metabolic pathways are correlated one to another in different groups of bacteria. However, to exploit such correlation metagenomics results should be implemented with results from other omics like transcriptomic and proteomic.

Metagenomic study to identify targets to drive improvement of feed efficiency

Feed efficiency is a comprehensive trait to evaluate the efficacy of nutrient and energy metabolism (Yan *et al*., 2017). Improving feed efficiency can decrease the cost to producers, preserve additional edible resources for humans and reduce the excrement effluent as well as emission of greenhouse gases (Yan *et al*., 2017). Feed conversion ratio (FCR) and residual feed intake (RFI) are the major indices for assessing the feed efficiency of animals (Yan *et al*., 2017). FCR has been used in breeding for a long time because of its convenience and effect on improving growth (Yan *et al*., 2017). On the other hand, RFI has been proposed for measuring feed efficiency in breeding because of its phenotypic independence from maintaining body weight and body weight gain (Yan *et al*., 2017). Feed efficiency is a complex trait because it is influenced not only by the host genetics and physiological state but also by the intestinal microbiota, which would affect the nutrient digestion and energy absorption of the host (Yan *et al*., 2017). Singh *et al*., 2014 investigated the difference in microbial communities between good and poor feed efficiency broilers testing the microbiota in faecal samples and assessing FCR; *Acinetobacter*, *Anaerosporobacter* and *Arcobacter* were dominant in the poor efficiency group, whereas *Escherichia/Shigella*, *Faecalibacterium* and *Helicobacter* were dominant in the better efficiency group. However, the abundances of *Lactobacillus* and *Bacteroides* were similar in both groups. Mignon-Grasteau *et al*., 2015 quantified the microbial 16S rDNA in the cecum by quantitative polymerase chain reaction (qPCR) and observed higher ratios of *Clostridium leptum*, *Clostridium coccoides* and *Lactobacillus salivarius* to *E. coli* in the better efficiency group. Yan *et al*., 2017 investigated the role of duodenum, caecal and faeces microbiota in hen feed efficiency evaluated as RFI. Microbiota analysis suggested that the dominant microbes in duodenum and faeces had greater similarity than those in the caecum. Moreover, the microbial community diversity had higher diversity in the caecum than the other two sites. Significantly different taxa between better feed efficiency (BFE) and poor feed efficiency (PFE) hens were present in cecum. Notably, there were significantly higher proportions of Lactobacillus and Akkermansia in the BFE group. At species level, *Bacteroides coprophilus*, *Lactobacillus delbrueckii*, *Veillonella dispar*, *Lactobacillus reuteri* and *Prochlorococcus marinus* were significantly higher in the BFE group, whereas *Faecalibacterium prausnitzii*, *Parabacteroides distasonis* and *Thermobispora bispora* were found to be significantly higher in the PFE group. These results indicated the prominent role of cecal microbiota in the feed efficiency of chickens and suggested plausible uses of *Lactobacillus* to improve the feed efficiency of host.

Metagenomic mapping of variations in antibiotic resistance genes

A further application of metagenomics concerns the investigation antibiotic resistance genes (ARGs) in poultry productions, meaning the chicken guts, the poultry faeces and the farm environments. Animal faecal microflora harbours a vast reservoir of ARGs that could be acquired by human commensals and pathogens (Xiong *et al*., 2018). Antibiotic resistant bacteria and ARGs in animal excretion may be transported into the environment via manure application, leakage, runoff and airborne particulate matter, globally contributing to the aggregation of resistance in the environment (Xiong *et al*., 2018). The quantification of the relative contribution of poultry productions in the dissemination of antibiotic resistant bacteria and ARGs might support the work of international organizations who are monitoring their spreading in the environment. The World Health Organization (2014) initiated a global surveillance of antibiotic resistance in 114 countries, with their first surveillance report released in April 2014; the Public Health Agency of Canada also provided a 4-year surveillance report on antibiotic resistant organisms (2014); the U.K. Parliament released a note titled 'Antibiotic resistance in the environment' (2013) and the U.S. White House issued an executive order on national strategy (2014) for combating antibiotic resistance (Yang *et al.*, 2016).

Xiong *et al.*, 2018 applied metagenomic sequencing to investigate variations in ARG and bacterial host abundance in the faeces of broilers treated for 5 days with a low dose (0.2 g/L) and a therapeutic dose (2 g/L) of chlorotetracycline in the drinking water. The ARG abundance in the DNA extracted from the samples tested was determined using ARGs-OAP (accessible through http://smile.hku.hk/SARGs), which is an online pipeline for fast annotation and classification of ARG-like sequences from metagenomic data (Yang *et al.*, 2016).

The results obtained showed that therapeutic dose of chlortetracycline inhibited multidrug resistance genes (mdtA, mdtC, mdtK, ompR, and TolC) and promoted the abundance of tetracycline resistance genes (tetA and tetW) (Xiong *et al.*, 2018). The reduction of multidrug resistance genes was linked to the loss of Proteobacteria and, in particular, to the decrease of Escherichia/Shigella from 72 to 58% (Xiong *et al.*, 2018). The enrichment of tetracycline resistance genes was due to the emergence of Bifidobacterium harbouring tetW (Xiong *et al.*, 2018). Overall, these data indicated that the changes in the structure of antibiotic-induced faeces microbial communities accompany changes in the abundance of bacterial hosts carrying specific ARGs in the faeces microbiota. These findings should contribute to optimize therapeutic schemes for the effective treatment of antibiotic resistant pathogens in poultry farms.

Metagenomic investigation of poultry carcasses

Metagenomic investigations of food microbiota have until recently been less reported in the literature, perhaps because microbial communities of food are generally considered to have a low richness in terms of diversity (Kergourlay *et al.*, 2015). However, the development of NGS technologies and their application in the field of food ecosystems revealed that these communities are perhaps more rich than expected and that some of them might play a yet unsuspected role (Kergourlay *et al.*, 2015). Ercolini (2013) recently reviewed high throughput workflow for food analysis by NGS.

De Cesare *et al.*, 2018 applied metagenomics sequencing to investigate the microbiological profile of chicken carcasses collected from animals fed different diets by using shotgun metagenomic sequencing. The results showed that Proteobacteria and Firmicutes represented more than 98% of the whole bacterial population associated to carcass skins in all groups but their abundances were different between carcasses belonging to animals fed different diets. Significant differences were identified between carcasses even at species level, with particular reference to *Clostridium perfringens* and *Salmonella enterica* having a direct impact on the carcass food safety profile.

Future challenges

Although scientific publications concerning shotgun metagenomic applications in poultry science are rapidly increasing, there is an urgent need to solve two key issues. The first one is how many birds to sample within each experimental group in order to collect robust results. The second issue is how much sequencing is necessary to enable to see less abundant species, or even strains and less abundant genes in the communities under investigation. Without the definition of sequencing threshold, comparative taxonomic and functional gene analyses within and between experimental groups, over time as well as space, will remain of little value, because a small part of the sampled system will be compared to one another.

Nielsen *et al.*, 2014 suggested a method based on binning co-abundant genes across a series of human metagenomic samples that enables comprehensive discovery of new microbial organisms, viruses and co-inherited genetic entities without using reference genomes. In their study, the authors collected data from 396 human gut metagenome samples and used *Bifidobacterium animalis* subsp. *lactis* as a benchmark species because 19 samples originated from individuals who had consumed a defined fermented milk product containing that strain. The results showed that using co-abundance profiles that strain can be segregated accurately using as few as 18 samples (Nielsen *et al.*, 2014). In order to evaluate the sequencing depth Ni *et al.*, 2013 suggested considering that prokaryotic genomes have 1 to 15 small subunit (SSU) ribosomal (r) RNA gene copies, which range from 139 kb to 13,034 kb. The diversity of both SSU rDNA copies and prokaryotic genome sizes could significantly disturb the accurate estimation of the depth for metagenomic sequencing. Retriving the 2339 SSU rDNA sequence of three human faecal specimen from the study by Eckburg *et al.*, 2005, Ni *et al.*, 2013 suggested a computational approach demonstrating that the estimated amounts for sequencing specimens were 7.00 Gb at the species level, 6.93 Gb at the genus level, 7.10 Gb at the family level and 6.54 Gb at the order level. These results imply that at least 7 Gb is required for sequencing to enumerate the gene contents of prokaryotes with relative abundance of more than 1% in the human faecal microbiota (Ni *et al.*, 2013).

Overall, these results suggest to set metagenomic projects to investigate chicken gut samples and faeces considering to collect at least 18 samples from each experimental group and to sequence each sample at 7Gb. Even though the biological variability between birds belonging to the same experimental group might be lower in comparison to that associated to human specimen, this sampling parameters should guarantee results robustness in the poultry science context. The selection of the sequencing depth has the biggest impact on sequencing costs. As order of magnitude, the shotgun metagenomic sequencing of a sample at 7 Gb double cost sequencing of the same sample at 3.5 Gb. However, in the first case the sequencing results have much more value. Therefore, when a shotgun metagenomic sequencing project is set, then 7 Gb should be selected as sequencing coverage. Otherwise, a targeted amplicon sequencing is a much more cost effective alternative.

To standardize a set of sequencing parameters, including the sequencing threshold, the European Commission funded a Horizon 2020 project name COMPARE (www.compareurope.eu) with the specific aim to speed up the detection of and response to disease outbreaks among humans and animals worldwide by new genome technology.

Conclusion

The role of the gut microbiome in chicken productivity and health as well as meat product safety is subject to intensive study. Even if our knowledge of the gut microbiome composition, metabolic functions and influence on animal health, welfare, and performance is far from complete, metagenomic sequencing seems to be one of the most effective research strategy to fill this gap of knowledge.

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Microbiota, chicken’s gut health and antibiotic reduction/resistance

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Abbreviated title: Chicken gut microbiota

Summary: The development of gut microbiota in chickens in commercial production is characterized by gradual succession and replacement of *Proteobacteria*, *Firmicutes* and *Bacteroidetes*. However, this development can be manipulated by oral administration of cecal or fecal microbiota from adult hens or by antibiotic therapy. *Firmicutes* are major butyrate producers while *Bacteroidetes* are responsible for propionate production. *Bacteroidetes* also exhibit a broader potential for metabolism of complex polysaccharides. Chickens respond to the colonization with gut microbiota and the most characteristic feature is antibody production and secretion in intestinal tract. Understanding of function of individual microbiota members should allow to produce defined competitive exclusion products and thus compensate for the absence of any contact between newly hatched chicks and adult hen in commercial production.

Keywords: *chicken; microbiota;* Salmonella*; tetracycline; streptomycin; gut response*

Development of chicken gut microbiota

Colonisation of the intestinal tract of warm blooded animals begins immediately after birth or hatching with parents being the first and most important source of beneficial microbiota. Initial gut colonizers are recruited from facultative anaerobes of the family *Enterobacteriaceae* (phylum *Proteobacteria*). Soon after, representatives of *Firmicutes* begin to appear in chickens within a week after hatching, and lastly, representatives of *Bacteroidetes* appear as part of the intestinal tract microbiota (Lu et al., 2003; Mariat et al., 2009; O’Toole, and Claesson, 2010).

The development of gut microbiota in chickens in commercial production is characterized by an absence of contact between newly hatched chicks and adult hens. The initial colonization of the digestive tract in newly hatched chicks is therefore dependent on the microbiota present in the hatchery or the housing environment, and if a pathogen appears in the environment, the poorly populated gut of young chickens may represent an ideal ecological niche for its multiplication. This is why competitive exclusion products containing complex microbiota from healthy adult hens are used for early chicken colonisation and the prevention of infection with pathogens (Hoszowski and Truszczynski, 1997; Endt et al., 2010).

Four developmental stages of microbiota composition were recorded during the chicken’s whole life (Videnska et al. 2014b). The first stage was associated with the first week of the chicken's life and was characterised by a high presence of the representatives from the phylum *Proteobacteria* which formed nearly 50 % of microbiota in the first days of life. At the family level, *Proteobacteria* were represented mainly by family *Enterobacteriaceae* and genus *Escherichia*. The remaining part of caecal microbiota of this age category was formed by representatives of family *Lachnospiraceae* (phylum *Firmicutes*).

The second stage of caecal microbiota development was recorded in chickens 2-4 weeks of age reared at the conventional farm. This stage was characterised by a drop in *Proteobacteria* to less than 10 % by week 2 of life and by nearly absolute dominance of the representatives of families *Lachnospiraceae* and *Ruminococcaceae* (phylum *Firmicutes*) which formed around 90 % of the total microbial population in two-week-old chickens (Endt et al., 2010; Zhu et al., 2002; Yin et al., 2010). Representatives of *Lachnospiraceae* (mainly of genera *Blautia* and *Roseburia*) dominated in the caecum in two-week-old chickens and were outcompeted by representatives of *Ruminococcaceae* (mainly of genus *Faealibacterium*) at week 3 of life. Minority families of caecal microbiota belonging to phylum *Firmicutes* included *Lactobacillaceae*.

The third stage of caecal microbiota development was characteristic for chickens 2-6 months of age. During this stage a gradual succession of the representatives of *Firmicutes* and their replacement with the representatives of *Bacteroidetes* was observed. Within phylum *Bacteroidetes*, representatives of *Rikenellaceae* (genus *Alistipes*) were the first to appear at week 4 followed by the representatives of *Porphyromonadaceae* and *Bacteroidaceae* which were recorded for the first time in 12-week-old chickens. By week 26, i.e. at approx. 6 months of age, representatives of phylum *Bacteroidetes* comprised 55 % of the total caecal microbiome of hens.

The fourth stage was characteristic of hens older than 7 months. Caecal microbiota of this age category was formed mainly by representatives from *Firmicutes* and *Bacteroidetes*, each of them forming approx. half of the total microbiome. The representatives of *Bacteroidaceae*, though appearing for the first time at week 12, remained at a low level of representation (not exceeding 2 % of the total microbiota) until week 34 when they increased to approx. 10% and then fluctuated between 10 and 30 % of the total microbiome. Within stage 4, *Proteobacteria* re-appeared in the caecal microbiota of hens aged 34 weeks, from which point in time they formed around 5 % of the total microbiota. However, genera composition of representatives of *Proteobacteria* in the caecum of hens aged 34 weeks or older and one-week old chickens were different since the representatives of *Proteobacteria* in old hens belonged to genera *Desulfovibrio*. The microbial community in the caecum of chickens older than the age of broiler fattening time, i.e. older than 6 weeks, has been characterised much less frequently. Zhao et al. observed around 37 % of *Firmicutes* and 10 % of *Bacteroidetes* in 8-week-old chickens (Zhao et al., 2013) and Nordentoft et al. (2011) characterised microbiota of 18-week-old hens in which the majority of microbiota was formed by the representatives of *Firmicutes* and *Bacteroidetes*, followed by minority populations belonging to phyla *Proteobacteria*, *Actinobacteria* and *Fusobacteria*. In our previous study on the disturbances induced by antibiotic therapy, the microbiota of 15-week-old control chickens differed from that of the 46-week-old chickens with representatives of *Clostridiales* dominating in the faeces of the younger birds (Videnska et al., 2013b), and Callaway et al. (2009) described the microbiota of 75-week-old hens, two thirds of which were formed by the representatives of *Bacteroidetes*. Finally, we recently characterised faecal microbiota of broilers and hens originating from different European countries and found out that microbiota of broilers, *i.e.* chickens at 3-4 weeks of age, were enriched for *Firmicutes* whilst representatives of *Bacteroidetes* were present mainly in adult hens (Videnska et al., 2014a). Collectively this indicates that conclusions from our longitudinal study are correct though it is clear that the precise timing and/or percentages of representatives of individual phyla will differ from study to study and there might be extensive bird-to-bird variation.

Can be the microbiota development experimentally modified?

Since *Enterobacteriaceae* are replaced with different *Firmicutes* representatives in the second week of life followed by the appearance of *Bacteroidetes* later in life (Videnska et al., 2014b), we tested whether this development can be experimentally modified. Newly hatched chickens were orally inoculated with cecal microbiota from 1-, 3-, 16-, 28- and 42-week-old and composition of caecal microbiota was determined in inoculated chickens one week later. The inoculum itself (and therefore the microbiota present in the donor chickens) was the most decisive factor for the composition of cecal microbiota in the recipient chickens since the chickens inoculated with the cecal extract from different donors formed separate clusters (Varmuzova et al., 2016). Newly hatched chickens can be therefore colonized by microbiota of any composition.

Is the microbiota of different composition protective against Salmonella Enteritidis infection?

Next we determined the protective effect of cecal microbiota from 1-, 3-, 16-, 28- and 42-week-old hens against *S.* Enteritidis (Varmuzova et al., 2016). Following *Salmonella* Enteritidis infection, *Salmonella* counts in the ceca were significantly higher in the non-colonized chickens and the chickens inoculated with microbiota from 1-week-old donors than in the chickens inoculated with microbiota from 3-week-old or older chickens. Similar results were recorded also for liver colonization. Fecal transplantation of microbiota from 3-week-old or older chickens considerably increases chicken resistance to *Salmonella* infection.

Can the microbiota be used therapeutically to control Salmonella Enteritidis infection?

This question we addressed in an experiment in which newly hatched chickens were inoculated with microbiota on the day of hatching and challenged with *Salmonella* Enteritidis 24 hours later, or microbiota and *S.* Enteritidis were administered simultaneously on day 1 of life, or the chickens were first infected with *Salmonella* Enteritidis on the day of hatching and microbiota were provided to the chickens therapeutically 24 hours later. Parallel or secondary administration of cecal microbiota did not protect chickens against *Salmonella* Enteritidis infection. Only chickens that were provided with microbiota prior *Salmonella* Enteritidis challenge were effectively protected against this challenge (Varmuzova et al., 2016).

How extensive is the effect of antibiotic therapy at microbiota composition?

To obtain an insight into the changes occurring to the fecal chicken microbiota in response to tetracycline or streptomycin therapy, sequencing of the V3/V4 regions of the 16S rRNA genes was undertaken (Videnska et al., 2013a). Therapy with both antibiotics reduced the complexity of gut microbiota 2 days after antibiotic therapy. Chao1 index estimated the total number of OTUs present in faeces to 4,592 before the treatment which decreased to 709 and 263 after 2 days of streptomycin and tetracycline therapy, respectively. The interruption of antibiotic therapy for 12 days allowed for a rapid recovery of microbiota since Chao1 index increased to 1,675 and 1,860 after streptomycin and tetracycline therapy, respectively. However, the repeated antibiotic administration decreased the microbiota complexity again as Chao1 index decreased to 490 and 364 after repeated streptomycin or tetracycline therapies, respectively.

Analysis at the lower taxonomical levels showed that the therapy with both antibiotics reduced the prevalence of *Bifidobacteriales*, *Bacteroidales*, *Clostridiales*, *Desulfovibrionales*, *Burkholderiales* and *Campylobacterales*. On the other hand, the orders *Enterobacteriales* and *Lactobacillales* increased after the administration of both tetracycline and streptomycin. When we analysed the composition of the orders in which the increase in prevalence was recorded, since around 98 % of all *Enterobacteriales* were formed by *Escherichia*, the increase observed for the whole order after both streptomycin and tetracycline administration was caused by the increase of genus *Escherichia*. The order *Lactobacillales* comprised of 10 different genera, out of which the genera *Lactobacillus*, *Enterococcus*, *Paralactobacillus* and *Streptococcus* formed more than 99 % of all *Lactobacillales*. However, only representatives of the genus *Enterococcus* increased in after the therapy with both antibiotics.

What are the major metabolic pathways expressed by major gut colonisers?

Firmicutes colonising chicken cecum were characterised by the acquisition of Fe^{2+} by the FeoAB transport system and lipid metabolism using acyl-CoA intermediates. *Firmicutes* used the ATP binding cassette (ABC) and phosphotransferase system (PTS) transporters for the uptake of carbohydrates, glycerol and phosphate. Unique proteins expressed by certain representatives of *Firmicutes* included SudA and SudB subunits of sulfide dehydrogenase in *Roseburia* or CO dehydrogenase/acetyl-CoA synthase in *Blautia*. Major butyrate producers can be found among *Anaerotruncus*, *Faecalibacterium*, *Megasphaera*, *Oscillibacter*, *Subdoligranulum* and *Butyrivibrio* which expressed acetyl-CoA acetyltransferase, and at least one of two additional enzymes necessary for butyrate production, *i.e.* 3-hydroxyacyl-CoA dehydrogenase or enoyl-CoA hydratase (Polansky et al., 2016).

Bacteroidetes were characterised by the expression of lipid metabolism dependent on acyl carrier proteins, expression of the pentose cycle and respiration of fumarate. Of some rather unusual proteins, *Parabacteroides* expressed 3'-phosphoadenosine 5'-phosphosulfate sulfotransferase and *Bacteroides* expressed sulfate adenylyltransferase, which are both involved in sulphate assimilation. *Alistipes* expressed glutamate decarboxylase metabolising glutamate into γ-aminobutyric acid (GABA). Representatives of phylum *Bacteroidetes* were mainly propionate producers. For propionate production, *Alistipes*, *Bacteroides*, *Parabacteroides*, *Paraprevotella*, *Prevotella* and *Tannerella* expressed cobalamin-binding methylmalonyl-CoA mutase and/or methylmalonyl-CoA epimerase. Representatives of phylum *Bacteroidetes* exhibited also extended enzymatic activities in polysaccharide degradation since α-amylase, α-1,2-mannosidase, endo-1,4-β-mannosidase or glycogen debranching α-1,6-glucosidase were specifically expressed in *Bacteroidetes* and not *Firmicutes*. *Bacteroides*, *Alistipes*, *Paraprevotella* and *Tannerella* were characterised by high expression of xylose isomerase. In addition, *Bacteroides* expressed sucrose-6-phosphate hydrolase, α-L-fucosidase, α-glucosidase, glycosyl hydrolase and D-arabinose 5-phosphate isomerase (Polansky et al., 2016).

How does the chicken respond to gut microbiota colonisation?

Chicken response was determined by protein mass spectrometry using proteins extracted from cecal wall of 7 days old chickens (Volf et al., 2016). Chickens in control group remained untreated throughout the whole experiment while chickens in the experimental groups were inoculated with cecal extract obtained either from 4- or 40-week old donor hens on day 1 of life. Expression of ISG12-2, ES1 protein homolog, 2'-5'-oligoadenylate synthetase-like, constant region of IgM immunoglobulin, immunoglobulin λ light chain, cytidine deaminase, MRP-126 protein, protein deleted in malignant brain tumor 1-like, angiopoetin related protein 6, hemopexin, NK-lysin, ribonuclease homolog RSFR, lysozyme G-like 2, avidin, CUB and zona pellucida containing protein 1 and aldose reductase were induced by microbiota. On the other hand, fructose-bisphosphate aldolase B, transthyretin, calbindin D28, retinal dehydrogenase 1, alcohol dehydrogenase 1C, aldo-keto reductase family 1 member B1-like and estradiol 17-β-dehydrogenase 2-like were down-regulated in recipient chicken due to the administration of gut microbiota.

Conclusions

Although microbiota development in commercially hatched and raised chickens follow particular pattern, this pattern can be easily modified from the very first day of chicken life. Chickens do respond to the colonization. At the animal level, colonization of chickens with gut microbiota increases their resistance to *Salmonella* infection. At tissue and cellular level, chickens respond to the colonization by modified expression of numerous proteins. Major metabolic pathways and biological processes of individual microbiota members can be determined what allows for the predictions of their role in the whole microbial communities. Next logical step is extensive culture of gut anaerobes and construction of mixtures of defined composition which would compensate for the absence of contact between the hen and chickens.

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99 **Visceral fat of broiler and layer chickens and its apparent endocrine role in female reproduction /** Abstract ID: 294
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102 **EDIL3 and MFGE8: key proteins in the biomineralization of the hen eggshell /** Abstract ID: 403
L. Stapane[3], N. Le Roy[3], J. Ezagal[3], J. Poirier[3], V. Labas[1,2], L. Combes-Soia[1,2], J. Gautron[3]
[1]Université François Rabelais de Tours, Tours, France, [2]PAIB, PRC, INRA Centre Val de Loire, Nouzilly, France, [3]BOA, INRA, Université de Tours, Nouzilly, France

103 **Effectiveness of yeast cell wall in layers on intestinal and ovarian colonization of Salmonella enteritidis /** Abstract ID: 469
M. A. Bonato[2], C. Hofacre[1], L. L. Borges[2], G. Mathis[3], R. Berghaus[1]
[1]The University of Georgia - Poultry Diagnostic and Research Center, Athens, United States, [2]ICC Industrial Comércio Exportação e Importação Ltda., São Paulo, Brazil, [3]Southern Poultry Research Group, Inc., Athens, United States

104 **Effect of dietary microalgae oil supplementation on fatty acid composition and sensory profile of table eggs in laying hens /** Abstract ID: 247
J. Wang[1], G. Qi[1], S. Wu[1], H. Zhang[1]
[1]Feed Research Institute, Chinese Academy of Agricultural Sciences, Beijing, China

105 **Effect of yolk on functional properties of chicken eggs /** Abstract ID: 30
M. A. Grashorn[1], F. Hüeber[1], N. Kretzschmar[1]
[1]University of Hohenheim, Stuttgart, Germany

106 **Evaluation of antioxidative capacity of egg by using the ORAC and SOAC assay /** Abstract ID: 307
Y. Wang[1], Y. Tanaka[1], H. Hatta[1]
[1]Kyoto Women's University, Kyoto, Japan

107 **Influence of eggshell quality and egg white protein composition on Salmonella spp. contamination /** Abstract ID: 343
N. Domínguez Gasca[1], M. Hincke[2], A. Muñoz[1], A. Rodríguez-Navarro[1]
[1]Department of Mineralogy and Petrology, Granada, Spain, [2]Department of Cellular and Molecular Medicine, Ottawa, Canada

108 **Production and characteristics of functional egg based products with high biological and nutritive value /** Abstract ID: 117
V. Mazo[1], I. Stefanova[1], A. Kavtarashvili[1], I. Mokshantseva[1]
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109 **Proteomics and ultrastructural study of the Guinea fowl eggshell at key stages of the biomineralization /** Abstract ID: 405
N. Le Roy[3], A. Brionne[3], L. Combes-Soia[2], V. Labas[2], A. Rodriguez-Navarro[1], C. Rivard[4], Y. Nys[3], J. Gautron[3]
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112 **Characterization of productive traits and incidence of breast myopathies in a broiler chicken crossbred proposed for alternative production systems /** Abstract ID: 607
M. Zampiga[1], M. Petracci[1], A. Meluzzi[1], F. Sirri[1]
[1]Alma Mater Studiorum - University of Bologna, Ozzano dell'Emilia - Bologna, Italy

113 **Comparison of protein profile by SDS-PAGE of broiler breast meat affected by white striping, wooden breast and spaghetti meat myopathies /** Abstract ID: 408
F. Soglia[1], G. Baldi[1], E. Babini[1], C. Cavani[1], M. Petracci[1]
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114 **Effect of myopathy occurrence on quality of broiler chicken breasts during storage /** Abstract ID: 277
F. Gratta[2], L. Fasolato[2], M. Birolo[2], E. Novelli[2], M. Petracci[1], C. Zomeño[2], A. Piccirillo[2], A. Pascual Guzmán[2], G. Xiccato[2], A. Trocino[2]
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115 **European Union Approaches Toward Safe Poultry Meat /** Abstract ID: 507
H. Mohamed Hafez[1]
[1]Institute of Poultry Diseases, Free University Berlin, Berlin, Germany

116 **Evolution of macroscopic and microscopic broiler breast alteration related to White Striping /** Abstract ID: 220
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117 **Is meat of turkey breeders really different from that of standard turkeys? /** Abstract ID: 337
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118 **Occurrence and characterization of biofilms in drinking water systems of broiler houses /** Abstract ID: 238
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119 **Poultry meat quality and functionally food /** Abstract ID: 642
F. Rutz[1], E. Xavier[1], D. Lopes[1], V. Roll[1], J. Nunes[1], A. Roll[1]
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120 **Raw and cooked meat texture as affected by white striping, wooden breast and spaghetti meat myopathies /** Abstract ID: 352
G. Baldi[1], F. Soglia[1], C. Cavani[1], M. Petracci[1]
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121 **Rearing factors affecting myopathy rate and meat quality in broiler chickens /** Abstract ID: 264
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122 **Volatile fingerprints of raw and cooked meat of different commercial chicken genotypes /** Abstract ID: 529
A. Cartoni Mancinelli[3], E. Siletti[1], S. Mattioli[3], A. Dal Bosco[3], B. Sebastiani[2], L. Menchetti[4], A. Koot[1], S. Van Ruth[1], C. Castellini[3]

123 **White striping prevalence and its effect on meat quality of broiler chicken breast fillets under commercial conditions /** Abstract ID: 628
S. Golzar Adabi[2] , E. Demirok Soncu[1], N. Ceylan[3], N. Kolsarıcı[1]
[1]*Ankara University, Ankara, Turkey,* [2]*Huvepharma Turkey,* İstanbul, *Turkey,* [3]*Ankara University, Faculty of Agriculture, Ankara, Turkey*

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126 **Applying cold incubation profiles during the last week of incubation in a commercial incubator: effects on broiler embryonic mortality, hatchability, and chick quality**
Abstract ID: 324
I. van Roovert[2], M. van Eijk-Priester[1,2], J. Wijnen[1,2], C. van der Pol[2]
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127 **Assessment of GnRH analogues efficacy in enhancing the reproductive functionality in broiler breeders /** Abstract ID: 591
G. Kolluri[1], J. Mohan[1], H. Ali[1], J. Singh Tyagi[1], G. Marappan[1]
[1]*ICAR-Central Avian Research Institute, Bareilly, India*

128 **Both the rooster and incubation temperature affect embryonic metabolism and day-old chicken quality in laying hens /** Abstract ID: 100
H. van den Brand[1], S. van de Kraats[1], A. Sözcü[2], R. Jöerissen[3], M. Heetkamp[1], I. van den Anker[1], M. Ooms[1], B. Kemp[1]
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129 **Changes in Gizzard Development Caused by High Incubation Temperatures in the Chicken (*Gallus gallus*) Embryo /** Abstract ID: 94
R. Noiva[1], A. Menezes[1], H. L. Shivaprasad[2], M. C Peleteiro[1]
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130 **Chick yolk mineral levels during their sojourn in the hatcher /** Abstract ID: 245
R. Hopcroft[1], W. Muir[1], P. Groves[1]
[1]*University of Sydney, Camden, Australia*

131 **Effects of in ovo injection of N-Carbamylglutamate (NCG) on hatchability, quality of chickling, growth performance and carcass composition in broiler chickens /** Abstract ID: 255
G. Qi[1], H. Zhang[1], J. Wang[1], S. Wu[1], Y. Ma[1], F. Zhang[1]
[1]*Feed Research Institute, Chinese Academy of Agricultural Sciences, Beijing, China*

132 **Embryonic thermal manipulation of the Japanese quail: impacts on physiology and hypothalamic methylome and transcriptome /** Abstract ID: 233
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133 **Influence of Incubation Temperature on the Development of the Bursa of Fabricius in the Chicken (*Gallus gallus*) Embryo /** Abstract ID: 93
R. Noiva[1], A. Menezes[1], H. L. Shivaprasad[2], M. C Peleteiro[1]
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134 **Temperature variations during incubation and postnatal period affect performance, metabolism, health and gene expression in the blood of fast-growing chickens /** Abstract ID: 420
A. Collin[6], V. Coustham[6], N. Couroussé[6], S. Crochet[6], C. Praud[6], T. Bordeau[6], E. Godet[6], E. Cailleau-Audouin[6], P. Chartrin[6], I. Gabriel[6], C. Schouler[7], T. Larcher[4], C. Le Bourhis[2], K. Germain[5], O. Zemb[3], A. Travel[1], C. Berri[6], L. A. Guilloteau[6]
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135 **The Effect of Combined Monochromatic Photostimulation on Reproductive Activities of Broiler Breeders /** Abstract ID: 384
J. Bartman[1], S. Zaguri[1], L. Dishon[1], N. Avital Cohen[1], I. Rozenboim[1]
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136 **The in ovo critical period for somatotrophic axis elevation by green light photostimulation /** Abstract ID: 187
L. Dishon[1], N. Avital-Cohen[1], J. Bartman[1], S. Zaguri[1], I. Rozenboim[1]
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137 **Ultrastructural development of the Yolk Sac Tissue during incubation /** Abstract ID: 371
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140 **Adenoviral gizzard erosion in pullet and layer flocks and potential means of prophylaxis due to live vaccination with an apathogenic FAdV-A (CELO) /** Abstract ID: 260
B. Grafl[2], D. Liebhart[2], A. Schachner[1], M. Hess[1,2]
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141 **Amino acid substitutions in the Receptor Binding Domain (RBD) of Avian Influenza Viruses subtype H9N2 in Israeli isolates, with human health implications /** Abstract ID: 353
A. Lublin[1], I. Shkoda[1], A. Biton[1]
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142 **An innovative approach for analysing and evaluating poultry farms odour related to animal health and welfare /** Abstract ID: 357
V. Ferrante[1], G. Grilli[2], F. Borgonovo[1], L. Susanna[1], E. Tullo[1], M. Guarino[1]
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143 **A probiotic containing viable spores of Bacillus licheniformis improves gut health after coccidiosis vaccination /** Abstract ID: 212
V. Hautekiet[1], M. Vereecken[1]
[1]Huvepharma nv, Berchem, Belgium

144 **A probiotic containing viable spores of Bacillus licheniformis prevents outbreak of necrotic enteritis /** Abstract ID: 210
V. Hautekiet[1], A. Kanora[1]
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145 **A probiotic containing viable spores of Bacillus licheniformis reduces caecal colonization of Campylobacter /** Abstract ID: 211
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146 **A retrospective analysis to identify the contribution of the breeder on health and performance of broiler chickens /** Abstract ID: 78
I. de Jong[1], J. van Riel[1]
[1]Wageningen Livestock Research, Wageningen, Netherlands

147 **Assessment of innate immune response by applying mono or polyvalent live Newcastle disease and Infectious bronchitis vaccinations /** Abstract ID: 85
A. Ghalyanchilangeroudi[4], P. Hesari[3], R. KH Farahani[1], S. Ali Ghafouri[1], H. Hosseini[5], N. Sadri[4], A. Homayounmehr[3], R. Ehsan[2], H. Abdollahi[1], V. Karimi[4], M. Hossein Fallah[6], M. Jabbarifakhr[4]
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148 **Assessment of the efficacy of fecal sampling in representing gastrointestinal microbiota in chickens /** Abstract ID: 624
W. Yan[2], J. Zheng[2], C. Wen[2], C. Ji[1], D. Zhang[1], Y. Chen[1], C. Sun[2], N. Yang[2]
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149 **Autogenous vaccine against Escherichia coli and Gallibacterium anatis reduces losses and improves production on layer farms /** Abstract ID: 627
Ž. Gottstein[1], L. Lozica[1], D. Horvatek Tomić[1], G. Nedeljković[1], M. Lukač[1], E. Prukner-Radovčić[1]
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150 **Avian botulism: recent investigations from diagnosis to management (in France) /** Abstract ID: 328
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151 **Bacillus subtilis 29784 prevents a pro-inflammatory response in an induced inflammation condition using the Caco-2 cells model /** Abstract ID: 419
D. Prévéraud[1], L. Rhayat[1], E. Devillard[1], E. Eckhardt[1], M. Maresca[2]
[1]Adisseo France SAS, Commentry, France, [2]Aix Marseille University, CNRS, Centrale Marseille, Marseille, France

152 **Bio-monitoring of mycotoxins in blood serum and feed to assess exposure of broiler chickens in Belgium /** Abstract ID: 480
G. Antonissen[3], M. Lauwers[1], N. Caekebeke[4], M. Ringenier[4], F. De Meyer[2], J. Dewulf[4], S. Croubels[1]
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153 **Beyond effects on the microbiota, Bacillus Subtilis 29784 shows direct effects on the host /** Abstract ID: 429
D. Prévéraud[1], V. Jacquier[1], A. Nelson[2], L. Rhayat[1], E. Devillard[1]
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154 **Biomarkers as a tool to better understand the effect of mycotoxins on gut barrier functioning and immunosuppression /** Abstract ID: 34
A. Koppenol[1], L. Caron[2]
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155 **Coccidiosis Control With Farm Management to Prepare for "No Antibiotics Ever" Production /** Abstract ID: 298
L. Newman[1], A. Montoya[1]
[1]MSD Animal Health, Madison, NJ, United States

156 **Comparative study on primers for molecular detection of different infectious bronchitis virus genotypes** / Abstract ID: 84
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157 **Comparision of Commercial Broilers flocks vaccinated with a novel DNA construct vaccine applied in hatchery and reared in different field conditions** / Abstract ID: 49
D. Furmanek[1] , M. Mamczur[2]
[1]MSD, Warsaw, Poland, [2]Avi-vet Poultry Practice, Tarnowo Podgórne, Poland

158 **Comparison of different anticoccidial programs on coccidiosis control of broilers under floorpen conditions: shuttle versus full** / Abstract ID: 246
M. Marien[1], B. Dehaeck[1], M. Vereecken[1], M. Geerinckx[1], K. De Gussem[1]
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159 **Comparison of preventive ionophore supplementation on gut health and performance in broilers: evaluation in a necrotic enteritis model** / Abstract ID: 252
M. Marien[1], B. Dehaeck[1], M. Vereecken[1], M. Geerinckx[1], K. De Gussem[1]
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160 **Comparison of the gut microflora composition between different commercial layer farms: feasibility and limitations** / Abstract ID: 329
S. Rautenschlein[3], H. Kathy Scholtes[3], J. Kamphues[2], I. Rychlik[4], F. J. Hoerr[1]
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161 **Comparison of Two European Gel-Type Diluents vs. Water** / Abstract ID: 276
L. Newman[1]
[1]MSD Animal Health, Madison, NJ, United States

162 **Comparison two type of inactived avian infectious bronchitis vaccines (M41 and M41/Dutch variants) in protection against Variant-2** / Abstract ID: 83
A. Ghalyanchilangeroudi[3], V. Karimi[3], R. KH Farahani[2], H. Hosseini[4], T. Zabihi[3], M. Hossein Fallah[1]
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163 **Comprehensive epidemiological investigations support the role of Histomonas meleagridis for systemic translocation of *Escherichia coli* in chickens** / Abstract ID: 269
S. Paudel[2], B. Stessl[1], C. Fürst[2], D. Jandreski-Cvetkovic[2], C. Hess[2], M. Hess[2]
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164 **Delivery of oligodeoxynucleotides containing CpG motifs (CpG-ODN) by the intrapulmonary route against bacterial septicemia in neonatal broiler chickens** / Abstract ID: 443
S. Gomis[1], K. B. Goonewardene[1], S. Popowich[1]
[1]University of Saskatchewan, Saskatoon, Canada

165 **Dermanyssus gallinae as a vector of selected bacterial and viral diseases in flocks of hens in Poland** / Abstract ID: 603
L. Gawel[1], J. Urbanowicz[1], P. Falkowski[1], K. Bobrek[1], A. Gaweł[1]
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166 **Development and application of a quantitative real-time PCR for the detection of Enterococcus cecorum** / Abstract ID: 151
A. Jung[1], H. Petersen[1], L. Mohr[1], S. Rautenschlein[1]
[1]Clinic for Poultry, University of Veterinary Medicine Hannover Foundation, Hannover, Germany

167 **Different strains of Clostridium perfringens cause different levels of severity of necrotic enteritis in broiler chickens** / Abstract ID: 304
K. Gharib Naseri[1], R. Swick[1], M. Choct[1], N. Morgan[1], C. Keerqin[1], S. Wu[1]
[1]University of New England, ARMIDALE, Australia

168 **DNA construct vaccine applied in hatchery as a tool to combat ILT virus circulating in the field** / Abstract ID: 52
D. Furmanek[1], P. Stachów[1]
[1]MSD, Warsaw, Poland

169 **Effect of age of vaccination on antibody titers, clinical signs occurrence and performances after challenge with H9N2 on broilers** / Abstract ID: 355
Y. Bensassi[1]
[1]MSD Animal Health, Casablanca, Morocco

170 **Effect of fluralaner treatment on the production results at commercial layers farms in Poland** / Abstract ID: 303
S. Doner[1], D. Furmanek[1]
[1]MSD Animal Health, Warsaw, Poland

171 **Effect of selected yeast fraction on the growth of Clostridium perfringens: quantitative determination of growth inhibition and adsorption capacity** / Abstract ID: 244
G. Avantaggiato[2], E. Santovito[2], D. Greco[2], V. Marquis[1], R. Raspoet[1], V. D'Ascanio[2]
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172 **Effects of a dry hydrogen peroxide (DHP) air sanitation system used in an egg cooler on hatchability and chick quality** / Abstract ID: 301
B. Jordan[1], E. Melo[3], J. McElreath[1], R. Stephens[2], L. Lara[3], N. Cox[4], J. Wilson[1]
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173 **Effects of diet dilution on bone development and mineralization of coccidiosis-infected broilers** / Abstract ID: 510
I. Oikeh[2], P. Sakkas[2], J. Taylor[2], D. Blake[1], I. Kyriazakis[2]
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174 **Effects of feeding Diamond V fermentation metabolites on Avian Pathogenic E. coli prevalence and antibiotic resistance of E. coli in ceca samples taken from commercial broilers and turkeys** / Abstract ID: 284
W. Abdelrahman[1], J. A. Byrd[1], D. R. McIntyre[1], H. O. Pavlidis[1], J. P. Carroll[2], S. Carlson[2]
[1]Diamond V, Cedar Rapids, United States, [2]Iowa State University, Ames, United States

175 **Efficacy of a monensin/nicarbazin combination in the control of coccidiosis in turkeys under floor pen conditions using a seeder model** / Abstract ID: 251
M. Marien[1], B. Dehaeck[1], M. Vereecken[1], M. Geerinckx[1], K. De Gussem[1]
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251 **Use of perches and grids during light and dark period by growing chickens differing in growth intensity** / Abstract ID: 359
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254 **Welfare assessment of laying hens in four housing systems in Slovenia** / Abstract ID: 217
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258 **Amniotic feeding of flavanonein last week of incubationon, tibia growth plate and long bone mineralisation of hatched chick** / Abstract ID: 622
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ORAL PRESENTATIONS

Nutrition

A study of coccidiostat and phytase supplementation effects on precaecal phytate breakdown and P digestibility in broiler chickens

Abstract ID: 129

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A recent study has shown that P digestibility and phytate (InsP6) breakdown varied widely between institutions even though identical experimental diets were used. It was speculated that the coccidiostats used in the pre-experimental phase at some institutions contributed to this variation. Our objective was to study whether coccidiostats affect InsP6 breakdown and P digestibility, and whether interactions with P, Ca, and phytase supplements exist. A total of 560 male Ross 308 broilers were randomly assigned to 8 treatments with 7 replicated pens each. Birds were fed maize-soybean meal-based diets in 2 phases (day 1-10 and 10-25). Diets were based on the recommendations used in Germany with the exception of P and Ca in phase 2. Diets varied in P and Ca (4.2 g P and 6.5 g Ca/kg DM or 7.0 g P and 10.4 g Ca/kg DM), phytase (0 or 1500 FTU/kg of a modified, E. coli-derived 6-phytase, QuantumTM Blue, AB Vista), and coccidiostat (0 or 50 mg/kg of Narasin and Nicarbazin each) supplementation. Digesta from the terminal ileum was collected at the end of the trial and pooled on a pen basis. Feed and digesta were analyzed for titanium (indigestible marker), P, InsP6, myo-inositol (MI), and crude protein (CP). Data were analyzed using a three-way ANOVA and significance declared at $P \leq 0.05$. Coccidiostats neither influenced precaecal P digestibility and InsP6 disappearance nor ileal MI concentration significantly. Phytase supplementation increased precaecal P digestibility significantly by 18-29 percentage points. PCa supplementation had no effect on P digestibility in the absence of phytase but decreased P digestibility with phytase added significantly by 12 percentage points. Phytase increased precaecal InsP6 disappearance significantly by 39-65 percentage points and ileal MI content by 1.2-1.9 g/kg DM. PCa supplementation decreased InsP6 disappearance significantly by 11-36 percentage points and MI by 1.3-2.0 g/kg DM. Precaecal CP digestibility was significantly increased by the combined supplementation of phytase and coccidiostat without (82 vs. 78 %) and with PCa supplementation (82 vs. 80 %). We conclude that the coccidiostats used in this trial had no effect on endogenous phytases up to the terminal ileum. Results should be verified using other coccidiostats. Further work is warranted to determine if the effect of coccidiostats on precaecal CP digestibility is correlated with microbial protein reduction.

Keywords: *Broiler, Coccidiostat, Phytase, Phytate, Protein*

Barley in broiler diets: Optimum inclusion and response to carbohydrase enzyme

Abstract ID: 178

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Two trials were conducted to investigate the feed value of barley in broiler diets. In Trial 1, nitrogen-corrected apparent metabolisable energy (AMEn) and standardised ileal digestibility (SID) of amino acids (AA) of two barley grains and a sample of wheat were determined. A 3 × 2 factorial arrangement of treatments was used in both assays with three grain types (normal starch hulled barley [NSH], waxy starch hull-less barley [WSHL] and wheat) and two levels of carbohydrase supplementation (Ronozyme multigrain, DSM Nutritional Products, Singapore; 0 and 200 g/tonne feed). A significant interaction was observed between grain type and enzyme supplementation for AMEn ($P < 0.01$), with the greatest response being observed in WSHL. Grain type had a significant ($P < 0.001$) effect on the SID of nitrogen (N) and average AA digestibility, whereas enzyme effect was significant ($P < 0.05$) only for N digestibility. Birds fed wheat and WSHL based diets had the highest and lowest SID of N and AA, respectively, with NSH diets being intermediate. In Trial 2, the interaction between the inclusion level of barley and supplementation of a carbohydrase enzyme on growth performance of broilers was investigated in a 5 × 2 factorial arrangement of treatments with five levels of NSH (0, 141, 283, 424, 565 g/kg, as fed basis) substituted for wheat and two levels of enzyme (0 and 150 g/tonne of feed). The diets were formulated using the AMEn and SID of AA for wheat and NSH, obtained from Trial 1. The inclusion level of NSH had a significant ($P < 0.01$ to 0.001) effect on weight gain (WG), feed intake (FI) and feed per gain (F:G). Regardless of enzyme supplementation, WG showed a gradual increase up to 283 g/kg inclusion of NSH. Inclusion of NSH at 424 and 565 g/kg significantly ($P < 0.05$) suppressed FI. Increasing levels of NSH resulted in lower ($P < 0.05$) F:G. Enzyme addition improved WG ($P < 0.05$) and F:G ($P < 0.001$) at each level of barley inclusion. These data suggest that optimum inclusion level of NSH, with respect to broiler growth, is 283 g/kg of diet and feed efficiency can benefit from the supplementation of carbohydrases.

Keywords: *Barley, Broilers, Digestible amino acids, Metabolisable energy, Performance*

Calcium level in pre-starter diets of broilers: too high?

Abstract ID: 555

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During incubation, the embryo has limited access to minerals, except Calcium (Ca). The yolk is the main source of the phosphorus (P) needed for skeletal development but at hatch yolk P storage is minimal. Such physiological limitation might delay bone mineralization after hatch especially when chicks are fed starter diet at recommended Ca levels of 9-10.5 g/kg. Therefore, a trial to investigate early performance and bone quality when chicks received pre-starter diet formulated with reduced Ca level was carried out. Four pre-start diets (provided from placement up to 4th day), based on corn and soybean meal, were formulated to contain 4.0, 6.0, 8.0 and 10 g/kg Ca. All diets were equivalent in respect to digestible P (4.6 g/kg), AME (2800 kcal/kg) and dLys (11.5 g/kg). A total of 4960 male day-old Ross 308 chicks were allocated to 80 experimental units which were randomly assigned to experimental diets (20 replicates/treatment). At the end of 4th day all chicks received a starter (4-10 d), grower (10-28 d) and finisher diets (28-37 d) formulated with standard Ca level. Significance was set at $P<0.05$. Increasing dietary Ca concentrations from 4 to 10 g/kg Ca decreased body weight gain in the 2nd, 3rd and 4th days. Chicks fed with 4g/kg Ca increased feed intake during the first day and had improved feed conversion ratio from 0 to day 4. Tibia ash weight decreased at days 3 and 4 in chicks fed with 4 g/kg Ca. Paradoxical effect of decreasing dietary Ca improving performance but reducing mineralization at day 4 did not affect tibia ash weight and breaking strength during grow-out period. In fact, only 3 days after eating a standard Ca diet, the birds fed the low Ca had similar tibia ash compared to 10 g/kg Ca and were still gaining more weight at 7 and 14 days of age. Growth benefits observed in earlier ages were lost at 37 days although a one point improvement on feed conversion was observed in the global period. Increasing P availability early in life might support energy production, which in turn promotes growth. Results indicate that in the first days after hatch chicks seem to require less than the recommended calcium level in a starter diet. Decreasing calcium to 4 g/kg improved early growth and feed intake with positive carry over effects observed in the grow-out phase without compromising bone mineralization at market age.

Keywords*: Bone quality, Calcium, Performance, Pre-starter diet*

Chemical Analysis and Tannin Content of Some Raw and Germinated Sudanese Local Varieties of Sorghum and Their Potential as Poultry Energy Feed Source for Food Security in Rural Areas

Abstract ID: 594

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To reduce poultry feed cost and to alleviate the competition between man and poultry in energy source, the current study has been carried out to evaluate the chemical composition of some Sudanese sorghum varieties and their nutrient potential as feedstuffs. Three Sudanese sorghum varieties *Feterita*, *Wad Ahmed* and *Mogod* were brought from cereal grains markets of Khartoum and Janub Kurdufan states and subjected to proximate analysis and tannin content determination. Mineral content of *Feterita* and *Mogod* was evaluated and *Wad Ahmed* tannin content was assessed after five days of germination. The proximate analysis of *Feterita*, *Wad Ahmed* and *Mogod* showed the following composition: crude protein% (CP) 16.65, 11.03 and 16.24, crude fibre% (CF) 1.97, 2.44 and 3.06, fat% 3.86, 2.81 and 3.88 and metabolizable energy MJ/kg (ME) 14.25, 14.48 and 13.49, respectively. Calcium and phosphorus content values for wad *Ahmed* and *Feterita* were 0.03 and 0.41 and 0.02 and 0.32%, respectively. *Feterita* lysine and methionine concentrations were 0.3501 and 0.2925%, respectively. The percentage of tannin of *Wad Ahmed* was 0.1379%. Germination of *Wad Ahmed* seeds increased tannin content by six times. The same pattern of increasing was observed for fibre content of *Wad Ahmed* germinated seeds. The results indicated that sorghum variety *Mogod* could be used as alternative energy source with *Feterita* and *Wad Ahmed*. Because *Wad Ahmed* and *Feterita* are preferred to be consumed by man in rural areas and their production and prices are fluctuated according to rainy season. Moreover, *Mogod* is characterized by high disease tolerance and capability to be produced in very harsh climatic conditions.

Keywords*: Composition, Poultry, Sorghum, Tannin, Varieties*

Comparison between inorganic and hydroxychloride zinc on performance and carcass characteristics in broiler chickens: a meta-analysis

Abstract ID: 333

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Zinc hydroxychloride (ZHC) is a less reactive alternative Zn source than the inorganic Zn sulphate ($ZnSO_4$). Due to its unique characteristics and better bioavailability, ZHC will be less reactive in the feed and may cause less negative effects in the animals, resulting in better growth performance and carcass quality. The aim of this study was to compare the efficacy of ZHC to that of $ZnSO_4$ in broiler chickens raised under different circumstances. Nine studies were performed at different research facilities throughout Europe with similar experimental set-ups. In all studies, 80 ppm of Zn was added to the basal feed via the premix, resulting in a total Zn of about 120 ppm in the diet. This is representative of commercial practices in Europe. In the treatment where the Zn source was changed from $ZnSO_4$ to ZHC, all Cu in the premix (15 ppm) was changed from $CuSO_4$ to copper hydroxychloride as well. The growth performance and carcass measurement results of all nine studies with the two mineral sources were compared in a meta-analysis. Only the overall results of the studies, from one-day old chicks until slaughter, were used. In the meta-analysis a distinction was made between "Challenged" and "Non-challenged". In this study Challenged was defined as dietary, in which a wheat based diet was used; or as environmental, in which re-used litter or lower hygienic conditions were applied to mimic commercial practice. Results were obtained using random effects meta-analysis. The results show a clear distinction between Challenged and Non-challenged studies, in which greater responses to changes in mineral source were observed under challenged conditions. In the challenged studies there was a trend for increased average daily gain (P=0.09) with ZHC. In all studies (challenged and non-challenged) the breast meat yield was increased by 0.4% for the hydroxychloride trace minerals supplemented birds compared to birds fed sulphates (P=0.02). In conclusion, the meta-analysis of the nine studies indicated a beneficial effect of ZHC on growth performance and carcass qualities, which may have positive impacts on profitability and economics of broiler production.

Keywords*: Broiler chickens, Meta-analysis, Zinc hydroxychloride, Zinc sulphate*

Dietary arabinoxylan-oligosaccharides (AXOS) kick start arabinoxylan degradation in the ageing broiler

Abstract ID: 299

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Endoxylanases are frequently used in cereal-based broiler feeds to improve the nutritional quality of the feed. One of the major proposed mechanism of action of these enzymes is the generation of arabinoxylan-oligosaccharides (AXOS) that exert prebiotic effects in the gastrointestinal (GI) tract of broilers. It is however hypothesized that the age of broilers and the age-related development of their intestinal microbiota influences the efficacy of these endoxylanases. The objective of this study was to measure and understand possible age-related changes in and impact of AXOS supplementation on arabinoxylan (AX) degradation in different sections of the GI tract of broilers. Therefore, a feeding trial was performed on 480 one-day old chicks (Ross 308) receiving a wheat-based diet supplemented with or without AXOS (0.5%), containing no endoxylanases. Digesta samples from the jejunum, ileum and caeca and fecal samples were collected at six different ages: d 5, d 10, d 15, d 21, d 28 and d 35 and examined for AX concentrations and digestibility, intestinal viscosity and microbial endoxylanase activities. In the first two weeks, microbiota were able to solubilize the wheat water-unextractable arabinoxylan (WU-AX), thereby increasing intestinal viscosity and water-extractable arabinoxylan (WE-AX) concentrations in the GI tract. Establishment of an adult and stable microbial community at three weeks resulted in a further augmentation in the solubilization of WU-AX and an increase in WE-AX fermentation at the main microbial sites; ileum and caeca. Remarkably, in birds receiving AXOS supplemented feed, AXOS seem to boost the AX solubilizing capacity of the intestinal microbiota, observed by significantly higher total tract wheat WE-AX concentrations of AXOS birds compared to control birds. Furthermore, AXOS supplemented birds younger than 10 d of age showed a higher solubilization of WU-AX and an increased fermentation of WE-AX at the fecal level in comparison to the control birds. This study shows that the age of broilers and their age-related microbial development are important factors in determining the capacity of broilers to degrade wheat arabinoxylan. Moreover, prebiotic AX derived oligosaccharides will kick start the wheat AX degrading capacity of the developing microbial community at young broiler ages.

Keywords: Arabinoxylan degradation, Arabinoxylan-oligosaccharides, Broiler age, Gastrointestinal tract

Dietary Schizochytrium oil supplementation affect n-3 LC-PUFA enrichment in egg yolk, plasma lipid metabolism and egg yolk quality in laying hens

Abstract ID: 249

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Previously, we found that Schizochytrium oil (SO) could be used as an additive in layers to enrich eggs with docosahexaenoic acid (DHA). However, the mechanisms of the retention rate for DHA in yolk reducing with dietary DHA concentration are ambiguous. Hence, we investigated the possible mechanism by examine changes in plasma lipid metabolism. Four hundred and fifty 28-wk-old Hy-Line Brown laying hens were randomly allocated into five dietary treatments supplemented with 0, 0.25, 0.50, 1.00, or 1.50% SO feed. Each treatment contained six replicates with 15 hens each. The feeding trial lasted for 4 wks. The results showed that plasma lipid metabolites including total triglyceride, total cholesterol, high-density lipoprotein cholesterol and low-density lipoprotein cholesterol were reduced with dietary SO supplementation, while the non-esterified fatty acid, oxidized low-density lipoproteins and thiobarbituric acid-reactive substances were increased. An eicosapentaenoic acid (EPA) + DHA content of 182.32 mg per egg was observed for supplementation with 1.00% SO. Supplementing 1.00% and 1.50% SO in the diet led to approximately the same EPA + DHA enrichment (≈17%) in the yolk. More than 1.00% SO diet showed significantly greater yolk color than other groups. The results indicated dietary supplementation with 1.0% SO increased the EPA + DHA content in eggs, improved egg yolk color, and modulated lipid metabolism in hens.

Keywords: Egg yolk, N-3 LC-PUFA, Plasma parameter, Schizochytrium oil

Digestibility coefficients of pea, faba bean and lupin seeds following application of protease in broiler chickens

Abstract ID: 86

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Pea, faba bean or lupin seeds could be used as a valuable source of amino acids for birds, but nutrient utilization from above raw materials is lower than from soybean meal. Cibenza EP150 contains protease produced by fermentation of a single strain of Bacillus licheniformis. The aim of the study was to determine nutrient digestibility after protease addition. In the trial, 252 18-day old birds (ROSS 308) were used. Chickens were divided into 10 dietary treatments with 7 replications and two birds per replica. Birds from day 18 to 23 of age were fed experimental diets. Diets in 8 of the treatments were fed a basal diet containing yellow lupin, blue lupin, pea or faba bean meals in 30:70 proportion. Birds in 2 other treatments were fed only the basal diet. All diets were supplemented or not with Cibenza EP150. From day 20 to 21, excreta were collected and on day 22 all birds were sacrificed by electric stunning and ileal digesta samples were collected (2 birds per sample, 7 replications per treatment). Results of dietary protease activity was as follows: basal diet and other unsupplemented diets – not detected; supplemented diets – from 258 to 414U/g. The highest ($P<0.05$) crude protein pre-caecal digestibility was determined for yellow and blue lupin (0.77). Faba bean and pea seeds were characterized by 7pp lower crude protein pre-caecal digestibility ($P<0.05$). Apparent crude protein pre-caecal digestibility was found to be significantly affected by the applied protease. For faba bean, the difference in crude protein digestibility was 1pp and 2pp for pea and both lupin seeds. Apparent Asp, Gly, Lys and Arg ileal availability was improved after protease inclusion ($P<0.05$). The AMEN was the highest ($P<0.05$) for pea seeds and the lowest ($P<0.05$) for blue lupin seeds. Protease supplementation had a significant effect on the AMEN of investigated proteinaceous raw materials. The overall difference between supplemented and unsupplemented treatments was 57 kcal/kg. The inclusion of B. lichenformis protease evaluated in the present study shows promising results. The exogenous protease significantly enhanced crude protein pre-caecal digestibility coefficients. It appears that the relatively poor inherent digestibility of crude protein in pea, faba bean and lupins can be addressed by dietary inclusion of this exogenous protease.

Keywords: AME, Amino acids, Protein raw materials

Digestibility of Conventional and Organic Feedstuffs in Laying Hens

Abstract ID: 471

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Aim of the study was to determine the *in vivo* apparent ileal and faecal digestibility of phosphorus (P) and nitrogen (N) and derived apparent metabolisable energy (AME) values in laying hens of six feedstuffs with both a conventional and organic origin: maize, wheat, soybean expeller, sunflowerseed expeller, wheat middlings and peas. The experiment was performed with fourteen treatments: two basal diets with either a high or low protein level and twelve test diets consisting of a mixture of the test feedstuff and a basal diet in a ratio ranging from 25:75 to 40:60. The two basal diets were tested with eight replicates each. Each of the twelve test feedstuffs was tested with six replicates. A replicate consisted of eight laying hens. The experiment was performed with high productive Bovans Brown laying hens from 25 to 27 weeks of age. Birds were housed in digestibility cages with a wire floor. Excreta was collected during three days at the end of the experimental period. Ileal chyme was collected at the final day of the experimental period. TiO_2 was used as inert marker in the experimental diets to calculate nutrient digestibility. Faecal AMEn values as well as ileal digestibility of P and N were determined.
Differences in AMEn value and ileal P digestibility between conventional and organic batches of each feedstuff were small, and ranking of the feedstuffs per origin showed a similar pattern. Highest AMEn value was observed for maize being 3,683 and 3,640 kcal/kg for the conventional and organic batch, respectively. Ileal P digestibility was highest for soybean expeller with 31.1% and 28.8% for the conventional and organic batch, respectively. Origin had limited effect on ileal N digestibility, except for wheat middlings, in which ileal N digestibility ranged from 84.3% in the organic batch to 65.5% in the conventional batch.
In conclusion, there were small differences in AMEn value and faecal gross energy digestibility, and ileal P and N digestibility between conventional and organic test feedstuffs. Other factors, such as crop variety, regional differences, and processing, but also nutrient analysis, might have caused small differences in nutrient composition and digestibility between organic and conventional feedstuffs. In general, this study showed that origin only had marginal effects on nutrient digestibility coefficients, and therefore it is advised to use the same digestibility coefficients for calculating the digestible nutrient contents and AMEn value of both conventional and organic feedstuffs.

Keywords: Digestibility, Laying hens, Nitrogen, Organic feedstuffs, Phosphorus

Dose-effect relationship and safe use of a matrix-encapsulated phytogenic product based on Carvacrol, Cinnamaldehyde, Capsaicin and Cineol in diets for broiler and laying hens

Abstract ID: 325

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The first study evaluated the tolerance and dose depending efficacy of the phytogenic product in diets for broiler chickens from day 1 to 35 of age (400 animals, 4 treatment groups). The basal starter and grower diets were supplemented with 0, 100 mg/kg, 1.000 mg/kg and 10.000 mg/kg of the product. Body weight gain, feed intake, feed conversion ratio, health status, and blood parameters were used for demonstrating the tolerance and efficacy of the product. Compared to the control group, supplementation with 100 mg/kg showed significant improvements in body weight gain in the starter period (+4%) and significant improvements in feed conversion ratio (FCR) in the grower period (+4%) resulting in an overall improvement in FCR of 3%. At 1.000 mg/kg supplementation, a significant improvement in FCR of 6% was observed over the entire feeding period. Results recorded for hematological parameters were within the reference range of healthy broiler chickens. Based on blood parameters and optimal health conditions no negative effects were found when feeding up to 10.000 mg/kg of the product in feed. The second study evaluated the dose depending efficacy and tolerance of the phytogenic additive in laying hens from week 20 to 43 of age (200 animals, 5 treatments). A basal diet for laying hens was supplemented with dose levels of 0, 100, 250, 500, and 5.000 mg/kg of the product, respectively. Responses were demonstrated on body weight, body weight gain, feed intake, egg production, feed conversion ratio and health status throughout the 168 day feeding period. Blood profile measurements were included at the end of the study. Inclusion levels from 100 mg/kg onwards improved laying performance, egg mass and egg weight in comparison to the control group and reduced conversion ratio compared to the control group. Blood parameters obtained in the layers at the end of the feeding trial showed that the product at an inclusion of 5.000 mg/kg did not affect the measured parameters, when compared to the control group.
In conclusion, both studies revealed that graded inclusion levels of a defined matrix encapsulated phytogenic blend can significantly increase production parameters of both broiler and laying hens without negatively affecting animal health up to 100 fold the dose recommended for the inclusion into diets.

Keywords: Dose-effect, Efficacy, Phytogenic additives, Product safety, Tolerance

Dried yeast and its enzymatically hydrolyzed derivatives can improve performance and organ development of broiler chickens challenged with Salmonella lipopolysaccharide

Abstract ID: 81

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The efficacy of dried whole yeast and its enzymatically hydrolyzed derivatives on performance and organ development of broiler chickens challenged with Salmonella lipopolysaccharide was considered. Eight diets based on maize and soybean were offered to Ross 308 broiler chickens from 0 to 35 d. The eight treatments used in this experiment consisted of a Negative control [NC] (without supplementation); Positive control [PC] (without supplementation but challenged); Whole yeast [WY]; Yeast cell wall [YCW];Yeast glucan [YG];Yeast manno-protein [YMP]; Zinc bacitracin [ZNB] (antibiotics); Salinomycin (antibiotics). All yeast treatments were included at 2 g/kg diet while Zinc bacitracin and Salinomycin antibiotics were included at 0.0267 and 0.05 g/kg diet, respectively. Except for the birds in the NC group, birds in the other groups were inoculated with Salmonella lipopolysaccharide (LPS) intraperitoneally on days 13, 15 and 16. Each of these 8 treatments were replicated 6 times with 9 birds per replicate. There was no significant ($P > 0.05$) difference in feed intake (FI) across the dietary treatments at 10 d pre-LPS administration. Compared to birds on NC diet, birds on PC diet had reduced FI while other treatment groups had comparable FI at 24 and 35 d post-LPS administration. Broiler chickens on ZNB, WY and YCW diet had better BWG and FCR compared to other dietary groups at 10 d. A decline ($P < 0.05$) in BWG and FCR was observed for birds in the PC group compared to the NC group while the negative effect of the LPS on BWG and FCR was ameliorated across all the yeast and antibiotics groups at 24 and 35 d. There was no significant ($P > 0.05$) difference observed in the weight of all visceral organs measured at 10 d. Except for the spleen and bursa, the weight of other visceral organs were not significantly different at 24 d. Birds on the PC diet had heavier spleen but lighter ($P < 0.05$) bursa than birds on the NC and other groups. These results show that supplementation of dried whole yeast and it derivatives at 2 g/kg can improve broiler performance under mild stress conditions and could serve as an alternative to antibiotics in broiler chicken diets.

Keywords: Antibiotics, Broiler chickens, Lipopolysaccharide, Stress, Yeast

Effect of a multi-strains yeast fractions product plus anticoccidial on performance and gut health of broiler chickens under Eimeria challenge

Abstract ID: 562

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The aim of this study was to investigate the combined effect of dietary supplementation of a multi-strains yeast fractions product (MSYF) and an anticoccidial on performance and health of broiler chickens orally challenged with coccidian oocysts. A total of 192 male one-day-old Ross-308 chicks were randomly allocated into 4 groups with 6 replicates of 8 chicks each, housed in separate cages. All groups were offered *ad libitum* water and the same basal diets of meal feed. The first group (negative control, NC) was unchallenged, whereas the second group (positive control, PC) was orally challenged with *Eimeria* oocysts. The other two challenged groups, had their diets further supplemented with either an anticoccidial (salinomycin 60 mg/kg feed) or a combination of an anticoccidial plus the examined MSYF (0.4 g/kg feed). The three challenged groups were orally inoculated with sporulated oocysts (3.5×10^4 *Eimeria acervulina*, 7.0×10^3 *Eimeria maxima*, and 5.0×10^3 *Eimeria tenella* per bird) at 14 days of age. Performance parameters were weekly recorded, along with fecal oocyst numbers and mortality. On 7 days post inoculation, coccidiosis lesions were scored and samples were taken for histopathology evaluation. Counts of total aerobes, total anaerobes, coliforms and lactobacilli were also enumerated in digesta samples from jejunum and cecum by conventional microbiological techniques. All data were subjected to one-way analysis of variance for parametric ones and Kruskal-Wallis test for nonparametric ones. The combination of MSYF plus salinomycin when compared to PC had improved ($P<0.05$) body weights of chicken in the measurements up to day 28 of age, as well as higher ($P<0.05$) jejunum and cecum lactobacilli counts and lower ($P<0.05$) oocysts counts during the acute phase of the challenge. Moreover, PC had lower ($P<0.05$) duodenum villus height compared to the other three groups and lower ($P<0.05$) jejunum villus height compared to NC and the MSYF plus anticoccidial group. The intestinal lesion scores were higher ($P<0.05$) in PC group, whereas the anticoccidial and the MSYF plus the anticoccidial groups tended ($P<0.10$) to have less severe macroscopic lesions. Performances on several of the measured parameters of the combination of MSYF and anticoccidial were situated between those of the anticoccidial alone and NC. In conclusion, a combination of MSYF and salinomycin exerted a substantial improvement in growth performance and intestinal health in *Eimeria* challenged birds.

Keywords: Chicken, Coccidiosis, Gut microflora, Histopathology, Multi-strain yeast fractions product

Effect of licorice essential oils on meat quality and cecal microbial population of broiler chickens

Abstract ID: 48

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This experiment was conducted to evaluate the effects of licorice essential oils on meat oxidative stability and cecal microbial populations of broiler chickens. Six hundred and twenty five day-old Arian broiler chicks were used in a completely randomized design with 5 treatments, 5 replicates and 25 birds per each. Dietary treatments were included: control (basal diet), antibiotic (150 mg Avilamycin /kg), probiotic (100 mg Protexin / kg), licorice essential oil (200 mg/kg) and licorice essential oil (400 mg/kg). Dietary treatments were iso-caloric and iso-nitrogenous and fed from 1-42 days of age. At the end of the experiment (day 42) two birds from each replicate were slaughtered, thigh meat samples were collected and immediately transferred into freezer (-20°C) for subsequent malondialdehyde concentration analysis as a meat oxidative stability index (0, 50, 100 and 150 hours after slaughter). Cecal contents of each slaughtered birds were also collected and transferred to the laboratory for counting microbial population (E. coli and lactobacilli). The results of meat oxidative stability index at different times after slaughter showed that concentration of malondialdehyde in meat samples of birds fed diets containing 200 or 400 mg/kg licorice essential oil was lower than other treatments ($P<0.05$). The population of E. coli and lactobacilli in the intestine of birds received antibiotic was less than other treatments ($P<0.01$). Nevertheless, lactobacilli population in the intestine of birds fed with licorice extract tended to increase in comparison with others. This study showed that licorice essential oil may have beneficial effects on meat quality of broiler chickens.

Keywords: Licorice essential oils, Meat quality, Probiotic

Effect of low protein diets on nitrogen utilization and litter quality of broilers

Abstract ID: 630

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Reducing dietary crude protein (CP) is the most efficient strategy to reduce nitrogen (N) excretion by broilers that has also been reported to improve litter quality through reduction in water consumption and litter moisture (LM). Given the high amount of data available in literature on dietary CP reduction in broilers, a meta-analysis was performed to quantify the impact of reducing CP on N balance (N intake, excretion, retention, and retention efficiency), daily water consumption (DWC) and LM. The effect of the trial has been tested as a random effect and those of supplying the indispensable amino acids (IAA) at their requirement or not (IAAreq and IAAnot-req) and bird age (0-21 or 21-42 days) were also investigated as fixed effects in this meta-analysis. Based on a dataset of 116 trials for N balance criteria, the analysis revealed that reducing the dietary CP linearly decreases N intake ($P<0.001$; $R^2 = 99\%$) and N excretion ($P<0.001$; $R^2 = 99\%$) with a higher effect for 21-42 d than 0-21 d broilers for excretion (CP x Age, $P<0.001$). Besides, reducing dietary CP increases linearly the efficiency of N utilization ($P<0.001$; $R^2 = 98\%$) without impact of IAA level or age, meaning that age effect observed on N excretion is due to a higher consumption. Nitrogen efficiency of broilers can thus be increased by 2.3 % per % of dietary CP reduction. Based on a dataset of 22 trials for DWC, the analysis revealed that DWC of broilers (expressed in proportion to the highest CP level within trial) decreases linearly with the reduction of dietary CP level ($P<0.001$; $R^2 = 72\%$); increasing CP by 1 point % increases DWC by 2%. Regarding LM, based on 12 trials, the response to CP ($P<0.001$; $R^2 = 67\%$) showed that the LM of broiler might be reduced by 2.4% per point % of dietary CP reduction. This meta-analysis helps to quantify the effects of reducing dietary CP levels on N balance and on the litter quality of broilers. The combined reduction of N excretion and LM when reducing the dietary CP level may decrease the occurrence of footpad dermatitis by improving the litter quality. These results are useful to formulate low CP diets for a more sustainable broiler production.

Keywords: Broiler, Litter quality, Low protein diets, Nitrogen

Effect of microencapsulated dry chestnut wood extract and salts of butyric acid on production parameters of broiler chickens and laying hens

Abstract ID: 567

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Three studies were conducted to evaluate the effects of microencapsulated dry chestnut wood extract and salts of butyric acid on production performance of broiler chickens and laying hens. The experiment on broilers was conducted on 120 male Ross 308 chickens in the experimental facilities of the Faculty of Veterinary medicine in Zagreb. Animals were divided into groups of 20 animals and were assigned to randomized treatments (0, 250 and 500 g/t) with two replications per treatment. The group that received 500 g/t of feed achieved the best results of average body weight (2.943 kg) and feed efficiency (1.55kg of feed per kilogram of gain) at the end of the investigated period (day 42.). After slaughter, there was no significant difference in dressing percentage and weight of the liver. During the study were no significant differences in mortality between groups. The trials on laying hens were conducted on two commercial layer farms in Croatia. The diet of layers was supplemented with 500 g/t of additive to evaluate its effect on performance and health status. The trials lasted for 32 weeks (18^{th} week until 50^{th} week) and 13 weeks (16^{th} week until 28^{th} week) of layers age and were conducted on 31,770 and 92,887 birds, respectively. The birds were divided into two groups (experimental and control) and monitored for production results and health status. The experimental group of layers on Farm 1 achieved higher egg production (209.2 vs. 200.1), had better feed conversion (2.13 vs. 2.35) and had significantly lower mortality (2.9% vs. 6.9%) in the observed period. The control birds had significantly higher ADFI (122.7g vs. 115g) and there was no difference in the mass of produced eggs. The supplemented animals on Farm 2 had higher egg production (786.7 vs. 765.4), a better feed to egg ratio (142g vs. 150g) and a significantly lower mortality (0.6% vs. 1.4%) during the trial. Overall, we can conclude that the tested feed additive had a positive influence on the majority of production parameters that were monitored and can be recommended to the industry as a safe and effective way to increase production efficiency.

Keywords: Broilers, Butyric acid, Chestnut extract, Laying hens, Performance

Effect of novel soya bean meal processing technologies on broiler performance and digestibility

Abstract ID: 546

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Locally produced soybean meal (SBM) may be an important constituent of European broiler diets in the future. In the present trial, extrusion or cooking processes in combination with dehulling and pressing were used to produced 4 partly defatted SBMs from European soybeans (var. Ecudor) harvested in 2015 in the South of France (30 T/ha; 44 CP % DM, 25 Trypsin Inhibitor (TI) units/mg). Broiler starter (d0-14) and grower (d15-28) diets were offered to 288 Ross 308 male broilers as coarse mash in a 2x2 design: 2 processing methods ((Extrusion-pressing (E) vs Flaking-pressing-cooking (F)) x 2 hulling methods ((de-hulling (D) vs no dehulling (ND)). TI values were 2.6, 3.5, 3.6 and 7.6 TIU/mg for E/ND, E/D, F/ND and FD meals, respectively. The KOH protein solubility was increased by dehulling for E (70 vs. 76%) and F processes (82 vs. 89%, for non- and dehulled- meals, respectively). Variables measured on broilers consisted of ADG, ADFI, FCR, DM and CP digestibility and jejunal histomorphometry at d14 and d28 of age. In addition, carcass yield and carcass part yield, organ weight, and empty gastrointestinal tract (GIT) weight and length per small intestinal GIT segment were assessed at d28. Processing method did not affect any of the variables tested. On the other hand, hull presence increased ($P < 0.05$) ADFI over the starter period, but not over the grower period. In addition, hulled treatments increased proventriculus, gizzard and jejunum weight, and reduced carcass yield at d28 of age likely due to their higher fibre content. Method of processing and hulling significantly interacted ($P < 0.05$) for ADFI and ADG at the end of the starter period being the highest for the E/ND treatment, but overall broiler performance was similar between dietary treatments. Similarly, small intestinal architecture and DM and CP digestibility were not affected by dietary treatments at either d14 or d28 post hatch. In conclusion, all 4 methods of production resulted in comparable results in relation to performance variables. Hull removal did not confer a significant advantage, possibly due to the adaptive growth of the gizzard and proventriculus, aside from increased carcass yield.

Keywords: Broiler, Digestibility, European grown soybean meal, Extrusion, Flaking

Effect of pea extrusion and probiotic supplementation on performance, microbiota activity and biofilm formation in the gastrointestinal tract of broilers

Abstract ID: 131

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Peas (*Pisum sativum* L.) contain some antinutritional factors but are a valuable source of dietary protein and energy for poultry, and may be used to partially replace soybean meal (SBM) and wheat in broiler diets. The potential for the nutritional improvement of broiler diets by pea extrusion and *Bacillus subtilis* spore-based probiotic supplementation and their influence on microbiota activity in broilers was investigated. The fluorescence in situ hybridization (FISH) with rRNA-targeted probe was applied to investigate the spatial organization of Bacillus subtilis in the gastrointestinal tract (GIT) of broilers. Raw or extruded pea meals were included in wheat-SBM-based diets at 250 g/kg, a wheat-SBM diet served as a control. Pea-based diets were unsupplemented or supplemented with probiotic (*Bacillus subtilis*). The diets were fed to Ross 308 broilers kept in the individual cages from d 9 to d 28, weight gain and feed intake were monitored. During following 2 days chicks were sacrificed, GIT sections were excised, immediately fixed and analyzed for the presence of *Bacillus subtilis* spp., ileal and caecal digesta were sampled and analyzed for short-chain fatty acids concentration (SCFA). Inclusion of pea meal, raw or extruded did not affect body weight gain compared to control. Feeding pea diets with probiotic compromised feed utilization due to higher feed intake. The concentrations of SCFA in ileum were higher whereas in caecum did not differ in groups fed diets with pea compared to control birds and did not depend on experimental treatments. *Bacillus subtilis* were detected in all GIT sections, but they were more abundant in birds fed diets with probiotic. In the crop and duodenum the bacteria were present mostly in the feed, in the jejunum and ileum formed biofilm-like structures, while in caecum structured biofilms were observed on villi surfaces. It may be concluded that partial replacement of SBM and wheat by raw peas is possible, but the application of extrusion process seems not economically justified. Supplementation of pea diet with probiotic did not cause an excessive fermentation in the distal part of the gut. The *Bacillus subtilis* spore-based probiotic administration did likely enhance biofilm formation in the distal GIT sections probably with positive effect of the gut health but without clear effects on broiler productivity

Keywords: Bacillus subtilis, Broiler chicken, Extrusion, Histo-FISH, Pea

Effect of pH-manipulation of a wheat-based diet on phytase-induced phytate degradation in an in vitro simulation of crop retention

Abstract ID: 459

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Phytases can exert their effect in the crop, but the efficacy may be improved by reducing pH trough adding acids to the diet. An in vitro experiment designed to simulate crop environment was performed. Organic acids were used to manipulate pH in the feed. A pilot trial with propionic acid, formic acid, acidic acid and citric acid was conducted in order to determine the dose required to achieve the chosen pH levels, and assess stability in regards to pH. Each acid was added to the feed in quantities sufficient to reach pH 5.5, 4.5 and 3.5, respectively. The samples were then incubated in a water bath at 40 °C for 1 hour, with repeated pH measurements. Formic acid was chosen for the main trial due to its properties. A wheat-based broiler feed was ground and mixed with water at a feed: water ratio of 1:3. Formic acid was added in order to reach the following pH levels: 6.2 (without acid), 5.5, 4.5, and 3.5. Thereafter phytase was added at a rate of 5000 FTU per kg dry feed. All samples were mixed well, and incubated in a 40 °C thermal bath. Samples were taken after 10, 30, 60, 90 and 120 minutes of incubation. 0.5 g of sample and 0.5 ml 4 % trichloroacetic acid solution where immediately mixed to stop the phytase activity. The sample was then centrifuged, and the supernatant was stored at -21 °C until analysis. A colorimetric method was used to analyze free phosphate. A significant difference ($P \leq 0.05$) in amounts of P released was observed between all the tested pH levels, with higher P release at lower pH levels. Thus amount of released phosphate after 10 minutes, was twice as high at pH 3.5 compared to pH 6.2. Generally, the effect of incubation time was not significant, except for at pH 3.5, where more phosphate was released at 30 and 60 minutes incubation than at 10 minutes.

Keywords: Crop, Organic acids, Phytase, Phytate degradation

Effect of sexing and dietary incorporation of sugar cane molasses on broiler performance and carcass characteristics

Abstract ID: 592

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Sorghum is used as a source of energy in poultry rations in Sudan, in spite of its high price and competition between humans and animals. There are several alternatives for sorghum grains such as sugar cane molasses which is available and cheap source of energy as agro-industrial by-product. The unsexed one-day old broiler chicks are commercially used in Sudan. The data concerning the performance of male broiler chicks in comparison with unsexed one are lacking and need to be investigated. The objectives of the study were to evaluate the effect of partial replacement of sorghum grains by cane molasses on broiler performance and to compare the performance of male broiler chickens with unsexed one. A (2 × 3) factorial arrangement was used in a completely randomized design to study the effect of two sexing pattern (male and unsexed chicks) and three dietary levels of molasses (0, 10 and 24%) on broiler performance. A total of six treatments were employed and each treatment was replicated three times with six birds each. The experiment lasted for 42 days. Feed consumption, weight gain, and feed conversion ratio (FCR) were recorded. At the end of the experiment, two birds from each experimental unit were selected according closed to average weight and slaughtered. Carcass weight, weights of some internal organs and cuts were measured. Blood samples were collected for determination of cholesterol, triglycerides, protein and potassium. The results revealed that significant improvement ($p \leq 0.05$) in feed consumption, body weight gain and FCR as the level of molasses increased. The greater feed consumed and the highest weight gain were observed with birds fed on diets incorporate with 24% molasses. Males were better in weight gain and feed consumption than unsexed birds, while no differences in FCR. The greater blood protein and potassium levels were recorded with birds fed 24% molasses. Sexing had no significant effect ($p \geq 0.05$) on blood constituents. The weights of carcass, thigh, drumstick and breast were not affected by experimental treatments. The current study concluded that, the sugar cane molasses could be used as alternative energy source up to 24% in broiler diets without negative impact in performance and male were advisable to be adopted for commercial broiler production than unsexed one. Economically, the cost of diets containing 10 and 24% molasses were lower than control one.

Keywords: Broiler, Carcass, Molasses, Performance, Sexing

Effect of split feeding on performance and eggshell quality in laying hens

Abstract ID: 414

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The aim of this study was to maintain shell quality in Dekalb white hens by using an alternative system, split feeding. Two split feeding concepts were compared to two conventional diets. Within each feeding system, 2 ratios of fine (FL) and coarse limestone (CL) (50FL:50CL and 30FL:70CL) were used. In the conventional systems hens received the same diet during the day, containing either 50FL:50CL or 30FL:70CL. In the split system however, hens were fed either 50FL in the morning and 50CL in the afternoon or 30FL in the morning and 70CL in the afternoon. In total 4 treatments were compared, each had 4 replicates of 384 birds housed in enriched cages. The experiment was divided into 5 periods, treatments were compared within a period (34-46, 47-55, 56-65, 66-75 and 76-90 wk of age).
Laying % tended to be higher in the conventional feeding system compared to the split ($P = 0.095$), but only in the first period. Egg weight was similar in the feeding systems during the experiment. Feed intake was reduced in the split system in the second period ($P = 0.011$) and in the third and last period it tended to be lower compared to the conventional system ($P = 0.096$, $P = 0.076$). Split feeding also improved FCR in the second and last period ($P = 0.026$, $P = 0.044$). Not only feeding system, but also limestone ratio affected performance: in the first 4 periods feed intake and FCR were both significantly lower when hens were fed 50FL:50CL compared to those fed 30FL:70CL. Limestone ratio also affected cracked eggs %: feeding 50FL:50CL resulted in more cracked eggs compared to 30FL:70CL ($P = 0.035$), but only in the first period. In the fourth period cracked eggs % tended to be influenced by the interaction ($P = 0.083$): in the split system feeding 50FL:50CL could reduce cracked eggs % whereas in the conventional system feeding 30FL:70CL resulted in lower cracked eggs %. Shell thickness and breaking strength both decreased as hens aged ($P \leq 0.001$) but neither split feeding nor different limestone ratios could improve these traits.
The split feeding approaches used in this study resulted in better feed efficiency compared to the conventional systems. Therefore, it is worthwhile to fine-tune the split feeding concept in further studies so that it could be used as an alternative system to maintain shell quality in longer laying cycles.

Keywords: Alternative feeding, Laying hen, Shell quality, Split feeding

Effects of an organic acid combination with polyphenols via drinking water supplementation on broiler growth performance and ascites incidence under heat stress conditions

Abstract ID: 309

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This study was conducted to investigate the effects of an organic acid combination with polyphenols via drinking water supplementation on broiler growth performance and ascites incidence under heat stress conditions. A total of 240 one-day old Cobb 500 broiler chickens were randomly assigned to three treatment groups: control (no supplementation), S-1 (supplementation during 1-42 days) and S-2 (supplementation during 21-42 days). The amount of organic acid combination with polyphenols was applied as 0.2% in the drinking water (2 g/liter drinking water). The combination of organic acid included formic acid, lactic acid, propionic acid and acetic acid. During the experiment, room temperature was 28-30°C to induce heat stress in broilers. Each experimental group consisted of four replicates, each containing 20 chickens (10 females and 10 males). Growth performance, body weight gain and feed conversion rate were determined between d 1-21 and d 22-42. To determine the ascites incidence, chickens that died after 14 days of age were submitted to gross examination necropsy for hydropericardium, right ventricular hypertrophy and abdominal fluid accumulation. The hearts were removed and dissected to obtain heart weight, right ventricle (RV) and total ventricle (TV) weights to calculate RV:TV ratio as an index of ascites.The RV:TV values greater than 0.3 were considered as ascites. Data was analyzed using GLM Procedure of SAS. Body weight on d 21 was found to be lower in control and S-2 compared to S-1 (935.6 and 932.4 vs. 989.4 g, $P<0.01$). Final body weight was 10.5% and 7.8% higher in S-1 and S-2, respectively, when compared to control (2382.2 g, $P=0.002$). Accordingly, body weight gain between day 1 and 42 was found to be higher in S-1 and S-2 ($P<0.01$). . Feed conversion ratio was affected ($P<0.01$) with the highest FCR for control (1.96) and lower for S-1 (1.74) and S-2 (1.79). The RV: TV ratio was found to be the highest in control group with a value of 0.31, compared to S-1 (0.27) and S-2 groups (0.27, $P<0.001$). These findings demonstrated that organic acid combination with polyphenols could be used in broilers drinking water to improve body weight gain and feed conversion rate and decrease the susceptibility against ascites under heat stress conditions.

Keywords: Ascites, Broiler, Heat stress, Organic acid, Polyphenols

Effects of dietary supplementation of different sources of selenium and different levels of selenium yeast on laying hens

Abstract ID: 265

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Two feeding trials were conducted to investigate the selenium (Se) sources on laying hens. In trial 1, 648 Hy-Line laying hens at 18 week of age were randomly allotted into 6 groups with 6 replicates of 18 hens each, with the aim to compare the effect of different Se sources on performance,egg quality, plasma antioxidant capacity and egg Se concentration. Chicks were fed a basal diet without exogenous Se addition or a basal diet supplemented with sodium selenite (SS), selenium yeast (SY), selenomethionine (SM), nano-selenium (NS) or selenomethionine hydroxy-analogue (HMSeBA) containing 0.30 mg/kg Se. In trial 2, 576 Hy-Line laying hens at 21 week of age were randomly assigned to 6 groups with 6 replicates of 16 hens each, to investigate the effects of different levels of SY on performance, plasma biochemical parameters and egg Se deposition. Layers were fed a basal diet without SY addition or a basal diet containing 0.30, 0.60, 0.90, 3.0 or 15.0 mg/kg Se from SY, respectively. Both trials lasted for eight weeks followed an adaptation period of one week. In trial 1, Se sources did not affect laying performance and egg quality. The five sources selenium in the feed could increase the superoxide dismustase activity and decrease the malondialdehyde content in plasma. The glutathione peroxidase (GSH-Px) activities in SY group and NS group were much higher, and GSH-Px activity in the SY group was slightly higher than that in the NS group. Egg Se content could be deposited more efficiently by HMSeBA or SY addition. In trial 2, compared with hens fed the basal diet, dietary SY addition at 0.30~3.0 mg/kg Se may improve the production performance. Addition of SY at 15.0 mg/kg Se caused decrease in feed intake and body weight gain. Eggshell thickness, yolk color and eggshell rate in the group of 15.0mg/kg Se were significantly decreased. The activities of aspartate and alanine aminotransferase were markedly elevated in layers fed SY at 15.0 mg/kg Se, indicating the adverse effect on liver and the body health by the higher addition of SY. Taken together, Se sources did not affect laying performance and egg quality, and egg Se content could be deposited more efficiently by HMSeBA or SY addition among Se sources. Supplementation of SY in the feed of laying hens should be less than 15.0 mg/kg Se.

Keywords: Antioxidant activity, Egg Se content, Laying hens, Production performance, Se sources

Effects of supplementing microalgae in laying hen diets on productive performance, color and content of carotenoids and fatty-acid profile of yolks

Abstract ID: 105

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Hens egg is an excellent source of nutrients and use of various additives in feed mixtures can affect nutrient content of eggs, including proportion of omega-3 fatty acids in total fatty acids. Omega-3 fatty acids are known for their positive influence on the prevention and treatment of various diseases. Various authors state that algae are a good source of omega-3 fatty acids due to the similar fatty acid composition of fish. The aim of this study was to determine the effect of 0.5 and 1% algae (Schyzochytrium limacinum) supplementation in hens diet on the production indicators, color, carotenoid content and the fatty acid profile of the yolk. In total, 36 TETRA - SL brown hens 35 to 43 weeks old were randomly allocated to one of three treatments. Each treatment was assigned to 12 hens, individually caged on wire-mesh floor. All hens were fed with the same feed mixtures 0.5% (treatment B) and 1% (treatment C) of algae was added to basic diet (control, A). During eight weeks of trial period, inclusion of algae in hens diet resulted in an increase ($P < 0.05$) of average egg weight (B: 67.83 and C: 65.75 g) compared to the control (A: 64.46 g). Furthermore, hens fed experimental treatments obtained greater egg production over the trial period (B: 94.50 and C: 95.09%) compared to the control (A: 93.46%). Addition of 0.5% and 1% of the algae increased ($P < 0.05$) ω-3 fatty acids content in the eggs (B: 1.23 and C: 1.56% vs. A: 0.82%) and contents of EPA (A: 0.07 and C: 0.25%) and DHA (A: 0.45; B: 0.92; C: 1.19%) in total fatty acids. The omega 6/omega 3 ratio was lower ($P < 0.05$) in eggs from experimental treatments (B: 14.89 and C: 11.45) compared to the control (A: 22.29). Increasing proportion of algae in hens diet increase carotenoid content in egg yolk and consequently its color. The largest difference was observed between control without algae supplementation and the treatment containing 1% algae. The obtained results indicate the justification of 1% algae supplementation in hens diet due to the improved hens production indicators and the quality of eggs.

Keywords: Eggs quality, Laying hens, Laying performance, Omega-3 fatty acids

Effects of two limestone sources differing in particle size in broiler diets with or without phytase

Abstract ID: 351

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This study determined the effects of a coarse limestone source (75% of particles between 0.3-0.6 mm; Ca=374 g/kg) versus a fine source (<0.09 mm; Ca=383 g/kg), in diets with and without phytase on broiler performance during 0-21d of age. The fine source showed in vitro Ca solubility of 100% vs 26% for coarse limestone after 30 minutes at pH 3. Male Ross 308 broilers were used in a 3x2 factorial design with three phytase dose levels in corn-SBM based diets and the two limestone sources, respectively. Without phytase a starter (0-10d) and grower (10-21d) were formulated (Aviagen specifications, 2014) as positive control (PC) for each limestone source. Next, these diets were made with reduced total P (1.87 g/kg), retainable P (1.59 g/kg), Ca (1.99 g/kg) and Na (0.4 g/kg) contents only by exchange of MCP-P, limestone, salt and diamol, but supplemented with 500 or 1000 FTU/kg of a *Buttiauxella* phytase as NC500 and NC1000, respectively. Each dietary treatment (6) was tested with 8 replicate floor pens (30 birds/pen). Pelleted diets and water were provided *ad libitum*. At 21d, ileal digesta samples were collected from 12 birds per pen, pooled for phytate P (IP6) analysis. BWG, FI and FCR were determined and analysed via ANOVA ($P \leq 0.05$ considered as significant). Coarse limestone showed a better BWG (263 vs. 256 g; $P<0.001$), FI (283 vs. 278 g; $P=0.006$) and FCR (1.076 vs. 1.084; $P=0.014$) than fine limestone during the starter phase. NC500 had a worse performance than PC and NC1000 for all parameters. From 0-21 d no longer significant effects were shown on FCR, while an interaction effect was shown on FI and BWG. With fine limestone, NC500 had a lower BWG and FI compared to PC and NC1000, while there were no differences with coarse limestone. Ileal IP6 content was lower with coarse vs fine limestone (1.60 vs 1.77% digesta DM, $P< 0.05$). NC1000 had lower ($P < 0.001$) IP6 content than NC500, and both phytase doses had lower IP6 content vs PC. When compared to PC, NC500 and NC1000 reduced IP6 concentration by 70 and 88%, respectively. In conclusion, a less fine limestone source in diets with or without phytase can have a beneficial effect at performance of young broilers. Effects of factors like birds Ca requirement and age, digestible Ca in diet, limestone solubility in gizzard, particle distribution and origin need further study.

Keywords: Broiler performance, IP6 content, Limestone source, Phytase

Efficacy of a novel silica supplement fed to broilers

Abstract ID: 392

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A broiler bird trial was designed to assess the effect of inclusion of a novel silica, with and without phytase and with two different calcium to phosphorus ratios in a 2*2*2 factorial. 336 male Cobb 500 chicks were allocated one of 8 dietary treatments from day of hatch to 21 days of age. Birds were distributed into 48 pens with 7 birds per pen on arrival. Wheat-soya bean meal based basal diets were provided designed to meet the requirements of the age and strain of birds. The experimental protocol was approved by the Nottingham Trent University College of Science Ethics committee. The silicon supplement fed at 0ppm or 1000ppm, phytase at either 0FTU/kg or 1500FTU/kg (Quantum Blue, ABVista, UK) and a Ca:P ratio of either 1.9:1 (Low) or 2.2:1 (High). Bird bodyweight gain and feed intake were calculated weekly. On d21, two birds per pen were euthanized by cervical dislocation. Feet were removed at the tibial-tarsal joint and foot ash content measured. Tibias were removed at the femoral-tibial joint and bone strength assessed by breaking on a texture analyser with a three-point jig, and tibia ash percentage measured after ether extraction. Univariate analysis was performed using SPSS v.24 with Ca:P ratio, Phytase level and Silicon level as factors. Phytase did not improve bird performance, possibly due to the relatively poor bodyweight gain associated with feeding mash diets, or because phosphorus was present at more than sufficient levels in all diets. Addition of the silica supplement to the diet did significantly improve both bodyweight gain ($p=0.045$) and feed intake ($p=0.037$) at d21 in both high and low Ca:P diets irrespective of phytase addition. As expected, both tibia and foot ash were increased by the addition of phytase ($p=0.008$ and $p=0.002$ respectively) but, interestingly, the Si supplement did not show any benefit. Bone strength did not show any improvement with phytase addition, but there was a significant increase in bone strength when Si was added, perhaps due to an improvement in collagen strength or cross linking, but this requires further investigation. The data from this trial suggests that silica supplementation may improve bird performance and bone tensile strength in broiler chicks in the starter period but increased bone mineralization does not appear to be the mechanism for the observed response.

Keywords: Calcium, Lameness, Phosphorus, Phytase, Silica

Emulsifier and multi-carbohydrase in a maize, SBM, rapeseed meal and palm oil diet for broiler chickens

Abstract ID: 77

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Under normal conditions, the gastrointestinal tract of a chicken is an aqueous environment. Fatty acids, as hydrophobic components, have to aggregate to form micelles to get absorbed. Emulsifiers naturally mediate this process. Our results showed that use of exogenous emulsifiers had a positive effect also on neutral detergent fibre total tract digestibility. We assumed that improved fat digestibility reduced its content in digesta and, consequently, enhanced carbohydrate availability for microbe enzymes. It is common practice is to use carbohydrases to prevent negative effects of carbohydrates. On the basis of the above, it could be assumed that the use of exogenous emulsifiers and carbohydrases in poultry diets may have an additive effect on carbohydrate utilization by poultry. The experiment was conducted with 480 one day old ROSS 308 male chickens. Birds were randomly located in floor pens and assigned to 4 dietary treatments (15 replication in each, 8 birds per replication). The first group was fed a basal diet (BD) (maize-SBM, rapeseed meal (RM) and palm oil) without any supplementation. The second treatment consisted of a BD and an emulsifier additive (E), whereas BD in the third group was supplemented by an enzyme (S), (Superzyme OM - Canadian Bio-Systems Inc., Calgary, Canada). In the fourth group, both supplements were added to the diet (E+S). The content of palm oil in the diet changed during the experiment from 2% (starter diet) to almost 4% (in finisher diet). The highest BWG was noted for E+S treatment in starter and grower and finisher period ($P<0.05$). There were no differences between treatments in feed intake, across all feeding periods. During starter and finisher period birds fed E+S were characterized by the lowest FCR ($P<0.05$). The results of our study indicate positive effects of feeding of the emulsifier when the compound was used either alone or in combination with the multi-carbohydrase enzyme.
Research supported by the National Science Centre, Poland, grant 2015/19/D/NZ9/03580

Keywords: Broiler chickens, Emulsifiers, Enzyme

Feeding Broiler Breeders with Guanidinoacetic Acid (GAA) supplementation affect Creatine egg resources, elevates laying percent and improves progeny's performance

Abstract ID: 385

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Creatine plays an important role in energy metabolism and was shown to be a crucial component in broiler nutrition. We hypothesized that supplementary creatine precursor Guanidinoacetic Acid (GAA) will contribute to nutrient deposition in the egg and will consequently affect breeding performance and hatchlings quality. In this study, eighty 26-week-old Cobb500 broiler breeder hens and twenty males strain were distributed into individual cages and kept under standard conditions for 32 weeks. The breeders were randomly divided into 5 groups: a control group (no GAA in the diet) and four treatment groups, which were fed with 0.05%, 0.10%, 0.15% or 0.20% GAA in their diet. Eggs were examined for Creatine, Creatinine and GAA levels in both yolk and albumen. Broiler breeder performance (percentage of lay, egg deformations, hatchability, egg and chick weights), progeny body weight gain and breast meat yield were also examined at day 20. Results showed that Creatine concentrations in egg yolks were directly correlated with increasing administration of GAA. Creatine levels in treatment groups fed with 0.05%, 0.10%, 0.15% and 0.20% CreAMINO (GAA supplementation), compared to the control group were elevated by +33%, +73%, +46% and +106% after 11 weeks, respectively. In all 5 groups, no significant differences were found in the creatinine and GAA concentrations in the yolks, and in the concentrations of creatine, creatinine, and GAA in the albumen. Laying percentages, along 35 weeks of eggs production, showed that the 0.10% CreAMINO group had a significant increase, compared to the control: +3.6% at ages 30-35w, +2.6% at ages 36-41w, +3.1% at ages 47-58w and +18.3% at ages 53-58w. There were no significant differences in egg deformations, hatchability percentage, or egg and chick weights across treatment groups. Treatment groups of 0.15% and 0.20% CreAMINO also demonstrated significant increase in laying percentage, but only at older age (52-58 weeks); +16.7% and +14.8% respectively. Studying the effect of maternal-plus-broiler CreAMINO feeding showed an advantage on body weight and breast muscle percentages compared to the control group. An experiment conducted on 100 progenies of the broiler breeder hens fed with 0.00% or 0.15% CreAMINO, showed a significant increase in BW at 20d (of +15%) and in their breast muscle percentage (of 1.5%). These results show that GAA supplementation in broiler breeder diet has beneficial effects on laying and progeny growth performance probably due to the elevated levels of Creatine in the hatching egg.

Keywords: Broiler breeder, Creatine, Egg, Hatching

Formulation of Bacillus-based probiotics is key to product performance

Abstract ID: 257

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Probiotic-based products have received more attention in poultry productions since the focus on reduction of antibiotics has increased in recent years. Several Bacillus-based products are available but often users find a lack of consistent effects. Much focus has been on selection of the right strain but this factor is only one of several which needs to be considered to develop a consistent probiotic product. A probiotic strain screened and selected for performance in broilers was assessed in a range of assays related to formulation. Simple and optimized formulations were compared and analyzed with respect to demixing, flowability and particle size distribution (PSD) to assess the impact on homogeneity in feed. The optimized formulation had several beneficial characteristics when compared to simple formulations: The PSD was 233 um (Dv50) where other products were at either <100 or >500. In assessments of simple and optimized formulations of products, significant differences were found in a demixing study: In comparisons of bacterial counts (CFU) between bottom, middle and upper layer of a feed formulation the difference between upper and lower layer was a significant 142% in the simple formulation, while the difference in the optimized formulation was insignificant at 7%. In recovery trials from nine in vivo studies the average in-feed recovery was above 80% and the CV below 20% proving good in-feed homogeneity. A careful development of the correct formulation is an often overlooked key feature to ensure product performance. Our studies have shown that an optimized formulation with a correct PSD will result in less demixing, better flowability and thus higher in-feed homogeneity - as well as more hassle-free usage. Especially in the starter feed it's crucial that the in-feed counts of Bacillus spores are equal in each feed pellet as the chicks only ingest tiny amounts. Therefore, product performance is closely linked not only to strain selection but also to formulation features of the product.

Keywords: Bacillus, DFM, Probiotics, Product formulation

Glycine plus serine requirements of broilers fed low-protein diets

Abstract ID: 203

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Low-protein diets, supplemented with free amino acids might reduce the amount of imported soybean meal. In low-protein broiler diets glycine (Gly) and serine (Ser) may become deficient for maximal growth. The optimal level of digestible Gly+Ser (dGly+Ser) in low-protein broiler diets, however, is not clear yet. Therefore, a Gly+Ser dose-response study with 910 male broilers housed in 70 floor pens (0.75 m^2) from 0 – 35 d was performed. Broilers received the same starter diet from 0 – 10 d, and subsequently seven different dietary treatments were applied: a control treatment (normal protein diet), five low-protein dietary treatments with increasing concentrations of dGly+Ser (from 12.4 to 15.7 g/kg and from 11.4 to 14.9 g/kg, in grower (10- 28 d) and finisher phase (28 – 35 d) respectively), and a treatment in which 0.7 g/kg extra digestible threonine (dThr) was supplemented to the diet with the lowest dGly+Ser level. Crude protein content of the low-protein diets was in both phases 30 g/kg lower than the control. Within each feeding phase diets had similar essential digestible amino acids contents (lysine, cysteine, methionine, threonine, tryptophan, isoleucine, arginine and valine). Each treatment was replicated 10 times. A replicate was a pen with 13 broilers. Feed and water were provided for ad libitum intake. Growth performance, slaughter yields, litter quality, litter composition and footpad score were measured as response parameters.The dGly+Ser level in low-protein diets had no effect on the growth performance, litter quality, litter composition and footpad health. Broilers fed with the highest dGly+Ser dose had a lower breast meat yield compared with broilers fed the treatments with the lowest two dGly+Ser doses. Supplementing extra dThr to low-protein diets low in dGly+Ser had no beneficial effects on the growth performance, litter score and footpad score, but it negatively affected breast meat yield. Feeding low-protein diets had compared to the control no effect on the growth performance, but it resulted in a better litter quality, better footpad health and a more efficient protein conversion. As a consequence, less N was excreted. From this study, it can be concluded that dGly+Ser doses in low-protein diets of 12.4 g/kg and 11.4 g/kg in grower and finisher phase, respectively, is sufficient. It is also possible to reduce the crude protein content of grower and finisher diet with 30 g/kg, provided that the amino acid balance and level in the diets meet the bird's requirements by supplementation of free amino acids.

Keywords: Broilers, Glycine+serine requirement, Low protein

Hydroxy-selenomethionine improves feed conversion of heat stressed finisher broilers associated with enhanced Se bioavailability and antioxidant response

Abstract ID: 297

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Selenium (Se), under the form of selenocysteine, is an essential component of selenoproteins, amongst them major antioxidant enzymes such as glutathione peroxidases. It was shown that dietary hydroxy-selenomethionine could enrich tissues to a greater extent in selenomethionine and total Se and differently affected the expression of the selenogenome as compared to sodium selenite and seleno-yeast. Heat stress in broilers is known to induce oxidative stress. It was hypothesized that dietary supplementation with hydroxy-selenomethionine will be beneficial to heat stressed finisher broilers due to specific regulation of the selenogenome. A total of 720 one-day-old male Ross 308 broilers were allocated to 3 treatments with 12 replicates (20 birds each). Treatments were: no supplemental Se, Na_2O_3Se at 0.3 mg/kg Se, and hydroxy-selenomethionine at 0.3 mg/kg Se, added to corn-soybean meal diets, fed for 39d. A chronic cyclic heat stress model (temperature increase to 34°C with 50-60% RH, 6h daily) was applied in finisher phase (d25-39). One bird per pen was sampled on d26 (acute heat stress) and d39 (chronic heat stress) to determine selenium concentration in serum, glutathione peroxidase and glutathione redox status in erythrocytes or serum, liver and breast muscle. In the finisher period, when the heat stress protocol was implemented, an improvement in feed efficiency was observed (-5 and 6 points as compared to non-supplemented and Na_2O_3Se supplemented birds, respectively) (both $P<0.05$). Mortality in the finisher period, which increased substantially with cyclic heat stress, was numerically lowest in treatment with hydroxy-selenomethionine; -3.5 and 4.2% as compared to non-supplemented and Na_2O_3Se supplemented birds, respectively. Se supplementation dramatically increased Se levels in serum, and notably levels were higher for the hydroxy-selenomethionine fed birds as compared to supplemented birds. Corroborating with this, GPx activity in erythrocytes, breast muscle and liver increased multi-fold when diets were supplemented with Se. In conclusion, hydroxy-selenomethionine improved performance of heat stressed finisher broilers, but it remains to be established whether differential effects on the expression of other selenoproteins might be involved.

Keywords: Feed conversion, Glutathione peroxidase, Heat stress, Selenium

Identification of candidate genes for calcium absorption along the small intestine of the laying hen

Abstract ID: 582

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About 10% of total body Calcium (Ca) is transferred daily for eggshell biomineralization in laying hens, thus requiring an optimal balance between Ca absorption and mobilization from bone stores. Data from mice indicate that intestinal Ca absorption occurs through transcellular and paracellular pathways. In hens, the transcellular pathway implies a well-described intracellular Ca carrier Calbindin-D 28K (CALB1), whose expression increases together with Ca uptake during sexual maturity, while the paracellular pathway remains uncharacterized. The objective of the present study was thus to further characterize candidate genes of the transcellular pathway, to detect new ones for the paracellular pathway, in the hen's intestine and to assess their variation with sexual maturity.
Candidate genes were selected by combining a literature review and a bioinformatics approach using RNAseq data from chicken gastrointestinal tract. Specific primer pairs for 29 candidate genes were designed and tested on intestinal RNA from laying hens. Based on PCR product sequencing, melting curves analysis and RT-qPCR efficiency, 17 were validated. Samples of intestinal mucosa (duodenum, jejunum and ileum) were collected from 12, 15 and 17 week-old pullets or 23 week-old laying hens (n=6 per group). All were submitted to RNA extraction, reverse transcription and processed for gene expression measures by high-throughput microfluidic RT-qPCR (BioMark HD system).
The preliminary study validated specific primer pairs for candidate genes from the transcellular (7) and the paracellular pathways (10) in intestinal samples from laying hens. For the transcellular pathway, CALB1 was the highest expressed gene, followed by ATP2B1> CALM1>CACNA1C> SLC8A1>PKD2> CACNA1D. Surprisingly, unlike in mice, TRPV6 was not detected in the intestine of hens, despite using primers validated for other chicken tissues. For the paracellular pathway, Claudins (CLDN) Occludins (OCLN), Junctional Adhesion Molecules (JAM) and Tight Junction Proteins (TJP) were detected (CLDN1>TJP2>TJP1>CLDN10>CLDN2>OCLN>TJP3> CLDN12>JAM2>CLDN5).
The present study validated conditions for quantitative measurement of mRNA expression of numerous candidate genes for intestinal Ca absorption in the domestic hen. We observed a relatively high expression of CACNA1C and CACNA1D, which could compensate the absence of TRPV6 expression, and account for the apical entry of Ca in the transcellular pathway. We detected the expression of 10 candidate genes belonging to the paracellular pathway including CLDN2 and CLDN12, associated with vitamin D dependent Ca entry in other model species. Transcriptional profiling of those candidate genes during sexual maturity will contribute to an integrated view of intestinal Ca absorption pathways in the hen.

Keywords: Calcium, Intestine, Laying hens, Nutrition

Impact of fumonisins and deoxynivalenol on laying hens and the effect of a counteracting strategy

Abstract ID: 400

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In 2017 more than 18,700 samples of different feed and feed raw materials from 72 countries were analyzed as part of the BIOMIN Mycotoxin Survey Program. In Europe 51% of the 2,649 investigated samples were contaminated with fumonisins (FUM) and 65% of the 4,795 investigated samples were contaminated with deoxynivalenol (DON). In light of the well-documented negative effects of FUM and DON on the health and performance of poultry, the high prevalence of these mycotoxins is a cause for concern and necessitates the development of counteracting strategies.
In autumn 2016 EFSA issued a positive opinion for the use of a fumonisin esterase (FUM*zyme*®) and a life bacterial strain (Biomin BBSH®) in diets for all avian species. FUM*zyme*® biotransforms the toxic fumonisin B1 (FB1), cleaving off the two tricarballylic acid side chains to form the much less toxic hydrolyzed fumonisin B1 (HFB1). The levels of FB1 and HFB1 in feces are recognized biomarkers for evaluating the efficacy of FUM degrading feed additives. Biomin BBSH® is a feed additive that contains viable cells of bacterial strain DSM 11798 (genus nov., sp. nov., formerly *Eubacterium*), which degrades DON by deepoxydation.
In this study we evaluated the combined efficacy of FUM*zyme*® and Biomin BBSH® to counteract the negative effect of DON and FUM on layer performance. To this end a total of 150 laying hens (Lohmann Brown, 20 weeks old) were randomly assigned to 3 groups with 10 pens each (5 layers per pen). Group 1 was the negative control receiving basal feed, group 2 the positive control receiving feed contaminated with 10 ppm FUM and 3 ppm DON and group 3 the additive group receiving feed contaminated with 10 ppm FUM and 3 ppm DON supplemented with Biomin BBSH® and FUM*zyme*®. The trial lasted for 12 weeks. Statistical analysis was done with ANOVA (IBM SPSS 22.0).
Feed intake, body weight and weight gain were not significantly different between the groups. However, a negative impact on egg mass, egg shell parameters, laying rate and feed to egg mass was observed in the positive control group compared to control and additive groups, indicating that the feed additives counteracted the negative effect of FUM and DON on these parameters. Furthermore, the additive group showed a significantly ($p<0.05$) reduced concentration of FB1 and a significantly increased concentration of HFB1 in excreta compared to the positive control group, indicating gastrointestinal FB1 degradation by FUM*zyme*®. In conclusion, an additive containing a specific enzyme (FUM*zyme*®) to detoxify FUM and a bacterial strain to biotransform DON (Biomin BBSH®), allows the egg producer to reduce negative effects of frequently occurring mycotoxins on the animals.

Keywords: Biotransformation, Deoxynivalenol, Enzyme, Fumonisins, Mycotoxin

Influence of calcium solubility on egg production and egg quality in 68-week-old laying hens

Abstract ID: 444

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Calcium carbonate is the main component of a bird's eggshell, with limestone being the major calcium source in laying hen diets. Limestone characteristics, such as solubility and particle size, are known to vary between sources and can affect calcium digestibility and phytase efficacy as recently shown in broilers. These characteristics may also affect the efficiency of laying hens to use calcium for eggshell formation. The aim of the study was to evaluate calcium availability of different limestone sources for laying hens, measured by egg production and egg (shell) quality. Calcium availability was evaluated using five limestone sources with varying acid solubility values. Particle size was standardized across all sources to eliminate confounding effects of limestone source and acid solubility. Each limestone source was ground to contain a particle size ratio of 50 % fine (250µ; acid solubility ranging from 51.5% to 68.0%) and 50% coarse (1455µ; acid solubility ranging from 27.1% to 45.0%). In total 720 laying hens (Hy-line W36, 68 to 76 weeks of age) were used, with 144 hens divided over 6 replicates per treatment. For each diet, limestone from the various sources were exchanged on a one on one basis while keeping calcium content equal. Hens were restricted fed to assure equal intake of each limestone source. BW gain, feed intake, egg production, egg mass, feed efficiency and eggshell quality were measured. Data were subjected to mixed model analyses using PROC GLIMMIX (SAS). Additionally, correlation regressions were applied with acid solubility of either fine or coarse limestone as independent variables and egg(shell) parameters as dependent variables. Laying hens fed a limestone source with lower acid solubility produced more second class eggs ($P<0.05$), mainly because of an increased number of produced broken eggs. Correlation regressions showed higher egg mass production (lay percentage x egg weight) throughout the study by laying hens fed diets containing a higher solubility of the fine limestone ($R=0.324$; $P<0.05$) and higher solubility of the coarse limestone ($R=0.282$; $P<0.05$). Additionally, the percentage of egg shell (relative to total egg weight) was higher in eggs produced by laying hens fed limestone having a higher solubility ($R=0.318$; $P<0.05$). In conclusion, higher solubility of the coarse limestone fraction resulted in increased egg shell quality, with a heavier relative egg shell weight and a higher solubility of both fine and coarse limestone resulted in increased egg mass output.

Keywords: Acid solubility, Egg production, Egg shell quality, Laying hens, Limestone

Influence of insect (Hermetia illucens) or algae meal (Spirulina platensis) in broiler diets on growth performance, precaecal digestibility and intestinal microbiota of meat type chickens

Abstract ID: 590

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Alternative protein sources, such as processed insect meals or algae meals are in special focus of animal nutrition in order to examine potential replacement of soybean meal (SBM). As part of the multidisciplinary project "sustainability transitions" the aim of the study expected to evaluate the potential of *Hermetia illucens* larvae meal (HM) and meal of the microalgae *Spirulina platensis* (SM) in meat type chicken diets with complete substitution of SBM during the complete growth period.

In total, 336 one-day-old male chickens (Ross 308) from a commercial hatchery were randomly allotted to five diets and feed supply on free choice level. The control diet (main ingredients: wheat, corn, SBM) contained 39% SBM (starter period, d1-21) and 33% SBM (grower period, d22-34). The experimental diets replaced 100% of SBM by the alternative proteins under study. Diets with both of the alternative proteins and the control diet were amino acid (AA) supplemented according to current ideal AA ratio (IAAR) recommendations. In a further step, the supply of calculated first limiting AA (LAA) was reduced to 80% of its requirement recommendation for further evaluation of the individual dietary AA efficiency according to the 'Goettingen approach'. Experimental diets were compared to the control based both on zoo-technical response which was under weekly control (growth, feed intake, feed and protein conversion ratio, mortality) and several physiological parameters at the end of the study (Nutrient deposition, dietary protein quality, precaecal digestibility and microbial parameters in the gut).

First results indicate that the dry matter intake is significant lower with 100% substitution rate of SBM. However, feed and protein conversion ratio for the AA completed SM and HM based diets were similar or even superior as compared to the control diet. Except for Clostridium spp. the isolated bacteria from digesta and total bacterial counts were not significantly influenced by the dietary treatments. Parasites were not detected. Based on further results, the potential of alternative proteins of *Hermetia* and *Spirulina* in chicken diets will be discussed.

Keywords: Alternative proteins, Growing chickens, Growth performance, Microbiology, Precaecal digestibility

Influence of use of organic minerals in laying hen diets on productive performance, egg quality and bone strength

Abstract ID: 638

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One of the most important problems in poultry industry is eggshell quality and it has been estimated that eggs with damaged shells account for 6–10% of all produced eggs, what leads to great economic losses. The other problem observed in alternative egg production is poor bone quality related mainly with keel bone fractures which is one of the greatest welfare problems facing commercial egg production. Compared to inorganic sources, the organic mineral sources are reported to have several advantages, including protection from undesired chemical reactions in the gastrointestinal tract, easy passage through the intact intestine wall, and possibly different absorption, metabolic pathway, and mechanisms. The aim of the present experiment was to study the effect of two sources (inorganic and organic) of trace elements zinc, manganese, copper, iron and Se in the diet for hens on laying performance, eggshell quality and chosen parameters of bones. Experiment was carried out on 30 TETRA – SL Brown laying hens from 35 to 50 weeks, randomly assigned to two experimental treatments. Each treatment contained 15 laying hens, individually caged on wire-mesh floor. All layers were fed the same basal diet contained in 1 kg 52 mg Zn and 30 mg Mn. In experimental treatments basal diet was supplemented with 50 ppm Zn, 75 ppm Mn, 36 ppm Fe, 4 ppm Cu, 0,15 ppm Se in inorganic forms as ZnO, MnO, $FeSO_4$, $CuSO_4$ and Na_2SeO_3 resp. (group K), and with 30 ppm Zn, 30 ppm Mn, 5 ppm Cu, 5 ppm Fe, 0,2 ppm Se in organic forms as proteinate complexes. At the end of the experiment ten hens from each group were sacrificed and the tibias from both legs were prepared – left tibias for determination of ash and the right tibias for mechanical properties in three points bending test were determined. Egg production and egg weight were not affected ($P>0.05$) by dietary treatments. Dietary treatments in organic forms of microelements improved ($P < 0.05$) eggshell breaking strength and led to high content of Ca in tibias ash 184.48 in group P related to 179.77 g/kg in group K. Also in group P there was higher ($P < 0.05$) bone breaking strength relative to group K; 155.90 vs. 108.66 (N). Obtained results indicate that use of organic complexes could alleviate the negative effect of hen age on eggshell and bone breaking strength.

Keywords: Laying hens; organic complexes; laying performance; eggshell quality; tibia bones

In-ovo fed nutrients accelerate intestinal brush border maturation towards hatch

Abstract ID: 193

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In-ovo feeding (IOF) is a feeding strategy aimed to influence the developing small intestine at the final stage of incubation. During this period, the small intestine undergoes rapid changes in preparation for post-hatch digestion and absorption. These include the formation of intestinal villi, lined with microvilli expressing digestive enzymes and nutrient transporters, creating the functional intestinal brush border. Additionally, a glycocalyx layer coats the apical surface of microvilli, functioning as a digestive surface and protective barrier. Enrichment of the amniotic fluid with nutrients through IOF during this time point is expected to improve small intestinal functionality at hatch. The current study aimed to identify structural and functional maturation processes within the intestinal enterocyte brush border region of the chick embryo during the last days of incubation and evaluate the effect of IOF of L-glutamine and NaCl on these processes. Observations of an embryonic jejunum at day 17 of incubation (E17) under a scanning electron microscope revealed the presence of microvilli on the apical surface of enterocytes even prior to villi formation, with no glycocalyx layer. At day of hatch (DOH), villi structures were evident and microvilli were significantly longer, denser and covered by a glycocalyx layer. Correspondingly, a qPCR analysis of Villin and Ezrin, genes encoding microvillar components, showed significant increases in expression within jejunal segments from E17 to DOH. PepT1, a peptide transporter and a common marker for mature enterocyte absorptive potential, had a similar significant increase in expression from E17 to DOH. IOF of L-glutamine and NaCl at E17, induced a significant increase in DOH jejunal microvilli lengths, compared to non-IOF hatchlings. qPCR analysis of these DOH jejunal segments revealed that IOF of L-glutamine and NaCl created a significant increase in expression of Villin, Ezrin and PepT1 genes, compared to non-IOF hatchlings. These results indicate that IOF of L-glutamine and NaCl at E17 accelerate maturation processes that occur from E17 to DOH in the small intestine. This includes upregulation of genes involved in microvilli formation and structure, longer microvilli and an increased expression of nutrient transporters.

Keywords: Gene expression, In-ovo feeding, Microvilli, Scanning electron microscopy

Isocaloric and isonitrogenous replacement of soybean meal with other legumes influenced growth performance and ileal amino acid digestibility in young broiler chickens

Abstract ID: 135

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A total of 240 Ross 308 male broiler chicks at zero-day old were allocated to 4 treatments in a randomized complete block design. Each treatment had 6 replicates with 10 birds each. The treatments consisted of a wheat-soybean meal control diet and three additional diets in which lupin, faba bean or mix of faba bean plus barley (harvested from faba bean and barley mixed cropping) partially replaced soybean meal. The diets were iso-caloric and iso-nitrogenous and were fed to broiler chickens for 21 days. Birds and feed were weighed on days 0 and 21, and ileal digesta were collected on day 21. There were no significant treatment effects on weight gain or feed intake but birds receiving diets with lupin or faba bean plus barley mix had higher FCR ($P < 0.01$) compared with the control treatment. Birds receiving faba bean-containing diet had similar FCR with the control. There were no treatment effects on ileal digestible dry matter ($P = 0.14$) and energy ($P = 0.13$). However there were significant ($P < 0.01$) treatment effects on ileal digestibility of nitrogen and all amino acids. Generally ileal nitrogen and amino acid digestibility was lowest ($P < 0.01$) in diet containing faba bean plus barley mix but greatest ($P < 0.01$) in diet with lupin, the amino acid digestibility in birds receiving control and faba bean-only diets were intermediate and similar. It was concluded that partial substitution of soybean meal with faba bean supported growth performance and amino acid utilization comparable to wheat-soybean meal diet and that the poorer FCR in lupin-containing diet was not due to impact on intake or amino acid digestibility. Therefore faba bean can partly replace soybean meal in broilers but the effects of lupin on growth performance needs to be further understood before it can be included at appreciable levels.

Keywords: Broilers, Growth performance, Ileal digestibility, Legumes

Micro-Mineral composition and deposition efficiency in the broiler hatching egg.

Abstract ID: 312

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The hatching egg's nutrients contain proteins, fat, vitamins, macro and micro-minerals and is the nutrients source for the embryo, thus a sufficient nutrient content of the egg is at high importance. Micro-minerals have a fundamental role as catalysts in enzymes reactions and also take part in hormone secretion and immune-activity. Therefore, it is important to include micro-minerals sources in poultry diet. Nowadays, poultry nutritionists can choose between different sources of micro-minerals: Organic (OM), Inorganic (IM) or a mixture of both Organic and Inorganic minerals (OM+IM). In this study, 30 Cobb500 broiler breeder hens were fed for 12 weeks (31-43 weeks) conventional diets which included the same level of Zn, Mn and Fe from 3 different sources: IM; OM; OM+IM. 15 hatching eggs (5 per each treatment) were sampled at day lay, for their egg parameter (EggW, YolkW and AlbumenW). In addition, yolk and albumen from each egg was analyzed for the concentration of Zn, Mn and Fe, using ICP-AES. Results shows that OM+IM treatment had heavier egg (73.89 g versus 71.94 g and 71.88 g in the IM and OM, respectively). It was found that the deposition of Zn, Mn and Fe are premier in the yolk, compare to the albumen; Zn, Mn and Fe concentration in the yolk was 82, 1.26 and 129 ppm, respectively, while their concentration in the albumen was 0.87, 0 and 1.20 ppm, respectively. In order to determine the deposition efficiency of these micro-minerals in the egg, the ratio between the levels of Zn, Mn and Fe in the feed and between their levels in the yolk was calculated. Results show that Zinc had the highest deposition efficiency as 14.67% of the Zinc levels in the feed was invested in the yolk. Manganese had the lowest deposition efficiency (0.28%) from the feed, while Iron had intermediate levels (3.31%). Comparing the deposition efficiency of these micro-minerals between IM;OM; OM+IM diets shows that OM+IM had higher Zinc and Iron deposition efficiency in the egg compared to IM and OM (by +18% and +24%, respectively), while Manganese's deposition efficiency was similar between the three diets. It can be concluded that Zinc has a significant role for the production of the hatching egg since almost 15% of Zn from the feed is deposited in the yolk. Moreover, diets containing a mixture of IM and OM have higher deposition efficiency of Zn and Fe in the yolk.

Keywords: Deposition efficiency, Hatching egg, Micro-minerals, Yolk

Nanoparticles as feed additives in poultry nutrition

Abstract ID: 520

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Poultry is one of the fastest growing industries in the world especially in developing country like India. Increase in demand for meat and eggs leads to need of innovation and growth in research for attaining higher productivity. Nanotechnology is one such application which could be exploited by poultry feed industry where in, nano-particles (NP) can be used as a supplemental source of trace minerals in diets. Applications of Nano-particles includes administration of nutrients, supplements, probiotics and drugs, diagnosis and treatment of diseases, identity registry for individual animals and in use of hormonal immune-sensors in the management of reproduction. Nano-particles that tend to increase the surface area for better interaction with biological support; they prolonged the compound residence time in gut and reduce the influence of intestinal clearance mechanisms. Nano-particles have several novel properties like higher bioavailability, because of greater surface area, higher surface activity, high catalytic efficiency and stronger adsorbing ability. Nano-particles help in direct transportation of compounds to targeted organs or systems while avoiding fast degradability. Previously nanoparticles were synthesized by chemical method, which leads to excretion of chemicals into the environment but now it involves plant extracts which consists of sugars, polyphenols, terpenoids, proteins, etc. which are highly biodegradable and hence no adverse effect to the environment. Nano technology promises tremendous potential for poultry production provided more research and innovation is done in this aspect.

Keywords: Bioavailability, Nanoparticles, Poultry feed

OH-Methionine is as efficacious as DL-Methionine to sustain growth performance in ducks and improves their antioxidant capacity

Abstract ID: 263

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The objective of the present study was to compare the bio-efficacy of OH-Methionine (OH-Met or 2-hydroxy-4-methylthiobutanoic acid) to that of DL-methionine (DLM) as sources of methionine in terms of growth performance, carcass traits, feather growth and redox status of Cherry Valley ducks between 1 and 42 days of age. Six hundred and thirty male ducks were randomly allotted to 9 dietary treatments with 7 replicates of 10 birds each. The first group received a basal diet deficient in total sulfur amino acids. In groups 2 to 5 and groups 6 to 9, the basal diet was supplemented with four increasing doses of methionine as either DLM or OH-Met. The dietary supplementation with DLM and OH-Met improved body weight gain and feed intake as well as carcass, breast meat and feather weights compared with the basal diet. No significant difference was observed between the two methionine sources on growth performance, carcass traits and feather growth. The concentrations of some redox parameters in the pectoralis major muscle were improved by addition of methionine to the basal diet.
However, OH-Met significantly increased the total antioxidant capacity, the activities of glutathione peroxidase, and the concentration of reduced glutathione in the pectoralis major muscle, compared to DLM. No significant difference between methionine sources was found on the concentrations of oxidized glutathione and malondialdehyde in the pectoralis major muscle. In conclusion, DLM and OH-Met have equal biological value for the performance, carcass traits and feather growth of Cherry Valley ducks.
OH-Met demonstrated a better antioxidant capacity than DLM which would slow down the oxidative process in the meat during post-mortem storage and thereby would contribute to a better duck meat quality.

Keywords: DLM, Duck, Efficacy, HMTBA, Methionine

Performance and intestinal microflora of broilers fed the probiotic Bacillus amyloliquefaciens H57

Abstract ID: 492

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Probiotics have been shown to improve animal production and prevent enteric infections. In this study, the effects of the novel, spore forming strain, *Bacillus amyloliquefaciens* H57 (H57) on productivity and the intestinal microbiota of broiler chickens was evaluated.
A sorghum and soybean based mash diet with or without H57 spores (~10^7cfu/g), was fed to day old male, broiler chicks for 21 days. Chick growth and feed consumption was measured weekly and the ileal and caecal contents of 24 birds (2 from each replicate cohort of 15 chicks) were collected on day 21. Culture independent microbial profiling of the digesta samples was undertaken by sequencing V6 to V8 variable regions of the 16S rRNA gene with Illumina sequencing.
Permutational multivariate analysis of variance of operational taxonomic units from control and H57 treated birds indicated that H57 significantly modified the microbial community structure both in the ileum ($P < 0.005$) and caecum ($P < 0.005$). Microbiota diversity (Shannon index) was significantly reduced ($P < 0.05$) in the ileum by dietary H57 addition (control 4.35 vs H57 group 3.92) while diversity was not affected in the caecum (rarefaction with sequencing depth of 25,000 reads per sample in both sites) with a shift in the relative abundance of multiple bacterial genera in both sites. The most prominent change was an increase in the relative abundance of Bacteroides in the caecum from 0.0002% in control birds to 17.4% in the H57 treated birds; becoming the most dominant taxon. Quantification of H57 in digesta samples by real time qPCR indicated that H57 did not multiply in the intestine.
Together with the change in microbial community structure, H57 significantly improved ($P < 0.005$) growth rate. The average daily weight gain of H57 fed chicks was improved by some 6.9% (34.8 g/day/control bird vs 37.2g/day/H57 bird) resulting in a higher ($P < 0.01$) body weight at day 21(845g vs 896g). Similarly, H57 improved ($P < 0.05$) the feed conversion ratio by about 6% during the study without effect on feed intake.
In conclusion, dietary supplementation of the probiotic *Bacillus amyloliquefaciens* H57 to chicken modified the intestinal microbiota and improved growth and feed efficiency.

Keywords: Bacillus amyloliquefaciens, Broilers, Probiotic

Performance enhancing effect of a natural resin acid composition in broiler chickens under a variety of challenge conditions

Abstract ID: 481

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Coniferous trees secrete viscous rosin at the site of injury to inhibit invasion of pathogenic microbes. Rosin contains a mixture of diterpene carboxylic acids collectively referred to as resin acids, according to species and geographical origin. The antimicrobial and anti-inflammatory potential of resin acids is well documented and utilized *e.g.* in rosin salves that accelerate wound healing. The natural resin acid mixture of Norway spruce (*Picea abies*) and Scots pine (*Pinus sylvestris*) consists mainly of abietic, dehydroabietic and pimaric acids, and is found in tall oil fatty acids (TOFA).
Supporting the gastrointestinal health and performance of broiler chickens is important especially in antibiotic-free feeding. In our previous research, a patented and standardized resin acid composition (RAC; Progres®, Hankkija Ltd), sustainably produced from tall oil, inhibited the growth of several pathogenic Gram-positive bacterial cultures, including *Clostridium perfringens* and *Staphylococcus aureus*. Here we studied the potential of RAC as a performance enhancer for broiler chickens in different environmental conditions and feeding regimens, and also with and without intentional challenge.
Eight feeding trials without intentional challenge were run in four European research institutes, using local feeding and management systems. RAC at 0.500-0.750 g/kg feed significantly ($P < 0.05$) improved feed conversion ratio in four trials and the daily growth also in four trials, compared with non-supplemented control treatments.
In total, statistical improvement in performance was measured in five out of eight experiments (62.5%). Intentional challenge procedures, including *Eimeria maxima*, *Clostridium perfringens*, high stocking density, low temperature and/or coccidiosis vaccine, were implemented in additional five broiler experiments in European and US research institutes. Significant ($P < 0.05$) improvement of feed conversion, bird growth and/or mortality was reported in four out of five (80%) challenge trials.
Improved foot pad health was observed in both trials in which it was measured. In trials with *Clostridium perfringens* -challenge, RAC did not prevent the onset of necrotic enteritis nor did it reduce the formation of lesions or decrease the pathogen load. However, RAC improved the performance of challenged broiler chickens more often than unchallenged birds. Antimicrobial effects of resin acids unlikely fully explain the improved bird performance in our trials. Future research has been directed to reveal whether modulation of intestinal microbiota, reduced inflammatory processes in the gut epithelium, or other physiological functions are involved in the mode-of-action of dietary resin acid supplementation.

Keywords: Broiler chicken, Necrotic enteritis, Performance, Resin acid, Rosin

Phytate disappearance and myo-inositol release in gnotobiotic broiler chickens

Abstract ID: 138

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It has been shown that the degradation of phytate ($InsP_6$) in the digestive tract of broilers was reduced by dietary P and Ca supplements, but increased by phytase addition. When phytase is not supplemented to low-P diets, prececal phytate degradation in broiler chickens is substantial. This indicates endogenous enzymes are involved in phytate breakdown. However, the contribution of epithelial or microbial phosphatases have not been quantified. This study investigated the effects of dietary P/Ca and phytase levels on prececal $InsP_6$ disappearance and degradation products in gnotobiotic broilers. In two runs, 16 pens in 8 germfree isolators were allocated to one of 4 dietary treatments. Sterilized eggs were hatched in isolators and up to 10 male Ross 308 chickens were placed in each pen. Treatments fed from day 10-15 included irradiated diets without (PCa-; 4.1 g P and 6.2 g Ca/kg dry matter (DM)) or with (PCa+; 6.9 g P and 10.4 g Ca/kg DM) monosodium phosphate and limestone supplementation and without (Phy-) or with (Phy+) 1500 FTU/kg of a modified E. coli 6-phytase (Quantum™ Blue). TiO_2 was used as indigestible marker. On day 15, digesta from the terminal ileum was collected. Isolators did not remain germfree, but results indicated no or minor effects of identified contaminants on analyzed traits. A two-way ANOVA was performed and significance was declared at $P < 0.05$. Prececal $InsP_6$ disappearance was 42% in the PCa-Phy- treatment and 17% in PCa+Phy-. No $InsP_{3-4}$ isomers were found in ileal digesta. The concentration of myo-inositol (MI) in treatment PCa-Phy- (6.1 µmol/g DM) was significantly higher compared to PCa+Phy- (1.7 µmol/g DM), which suggested a rapid degradation of the lower InsPs by mucosal phosphatases and their inhibition by PCa. Supplementation of phytase significantly increased $InsP_6$ disappearance and removed effects of PCa supplements (72% in PCa-Phy+ and 67% in PCa+Phy+; $P > 0.05$). However, PCa supplement reduced lower InsPs degradation in the presence of phytase, resulting in significant lower MI concentrations in ileal digesta of PCa+Phy+ (3.4 µmol/g DM) compared with PCa-Phy+ (12.2 µmol/g DM). It is concluded that epithelial-derived phosphatases contribute significantly towards $InsP_6$ degradation in broilers. The potential of endogenous phosphatases to degrade $InsP_6$ and lower InsPs, however, is markedly reduced by dietary P and Ca supplements.

Keywords: Broiler, Gnotobiotic, Phosphatase, Phytase, Phytate

Phytogenic premix effects on gene expression of intestinal antioxidant enzymes and broiler meat antioxidant capacity

Abstract ID: 401

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The aim of this study was to investigate the effects of administration level of a dietary phytogenic premix (Anco®Fit - Poultry) on the gene expression profile of antioxidant enzymes (i.e. CAT, SOD, GPX2, GPX7) and transcription factor Nrf2 at intestinal level. In addition, broiler liver and meat lipid oxidation and total antioxidant capacity (TAC) were determined.

Depending on phytogenic premix (PP) inclusion level (0, 750, 1000 and 2000 mg/kg diet) in a three stage feeding programme formulated to meet Cobb 500 nutritional requirements, treatments were: PP-0, P-750, PP-1000 and PP-2000. Feed and water were available *ad libitum*. Each one of the 4 treatments had 125 broilers arranged in 5 replicates of 25 chickens each. At 42d, 2 birds per treatment replicate were analyzed for gene expression and 4 birds per treatment replicate were pooled for biochemical analyses.

Data were analyzed by ANOVA, taking the treatment as fixed effect. Statistical significant effects ($P \leq 0.05$) were further analyzed and means were compared using Tukey HSD test. In addition, polynomial contrasts tested the linear and quadratic effect of PP inclusion levels.

Gene expression of SOD was up-regulated in the duodenum (P=0.027), jejunum (P=0.026) and ceca (P=0.023) in PP-1000 and PP-750 compared to PP-0. Expression of GPX2 was up-regulated in the duodenum (P=0.032) and jejunum (P=0.013) in PP-1000 and in ceca (P=0.006) in PP-2000 compared to PP-0, respectively. In addition, Nrf2 was up-regulated in ceca (P=0.024) in PP-1000 compared to PP-0. Intestinal mucosa TAC was higher in duodenum (P=0.011) and ceca (P=0.050) in PP-1000 compared to PP-0. Lipid oxidation was delayed in a linear pattern with increasing PP inclusion level in breast (P_L=0.020) and liver (P_L=0.046). Moreover, the PP inclusion level resulted in higher breast (P=0.005), thigh (P=0.002) and liver (P=0.040) TAC. In particular, breast and thigh TAC increased in a quadratic pattern reaching plateau at PP-1000, whereas liver TAC continued to increase linearly.

Overall, a consistent PP inclusion effect on meat, liver and intestinal antioxidant capacity has been shown with PP-1000 being the most effective.

Keywords: Antioxidant capacity, Antioxidant enzymes, Chicken, Phytogenic

Preference tests in laying hens; the choice between insect type and treatment.

Abstract ID: 461

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Several literature studies have been conducted on the feasibility of the application of insects as a protein source in animal feed. It was found that insects contain high levels of protein and fat, minerals and vitamins and their amino acid compositions as well as digestibility make insects an interesting protein source for animal feed meeting the protein requirements of poultry for maintenance, plumage development, growth and egg production. The use of insects in poultry feed is in a start-up phase: several subsequent studies have been carried out, initially focusing on examining the possibilities and the feasibility of using insects as a protein source. The aim of this experiment was to test the uptake of insects by laying hens in addition to their standard diet and to see whether laying hens have a preference for a certain insect species, and whether heat treatment (blanching) has an effect on their uptake. Two experiments were performed to investigate whether laying hens show a preference for a certain type of insect and treatment. In each experiment 30 laying hens were offered black soldier flies (BSF), lesser mealworms and mealworms, all blanched and non-blanched.
When all insect types of larvae were considered, the consumption of blanched insect was significantly higher than that of non-blanched insect ($P = 0.002$). The consumption of blanched insect larvae was significantly higher than that of non-blanched insect larvae for lesser mealworm ($P = 0.028$) and mealworm ($P = 0.028$) (Figure 3). When comparing the total insect consumption to the layer feed uptake of the hens, it was found that overall there was less insect consumed than layer feed ($P = 0.022$). The consumption of BSF was also lower than the layer feed consumption during the trial ($P = 0.028$), while the consumption of lesser mealworm and mealworm were not significantly lower than the layer feed consumption.

Keywords: Black soldier fly, Laying hens, Lesser mealworm, Mealworms, Novel proteins

Relationship between variations in particle size distribution of mash diets and the occurrence of performance and health issues in German hen flocks

Abstract ID: 237

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The objective of this study was to examine the relationship between variations in particle size distribution (PSD) of mash diets and the occurrence of performance and health issues in German hen flocks. For this purpose, 98 samples of mash diets were collected from farms suffering from no obvious problem (group 1, $n = 31$), decline in performance (group 2, $n = 42$) or feather pecking and cannibalism (group 3, $n = 25$) since feeding new batches. Carrying out proximate analysis concentrations of apparent metabolisable energy (AME_N), crude nutrients, sodium, calcium and phosphorous were determined in samples. In accordance with German industrial norms DIN 66165-1 and 66165-2, the PSD of each sample was examined via dry sieving analysis using automatic sieve shaker equipped with 3150, 2000, 1400, 1000, 800, 500, 400 and 200 µm sieves (DIN ISO 3310-1). Samples' average particle size and uniformity were described by geometric mean diameter (GMD) and geometric standard deviation (GSD), respectively. For statistical evaluation, one-factorial ANOVA with "group" as fixed effect and Pearson correlation analysis between GMD and nutrient concentrations were performed. Group 3 showed significantly higher proportions of coarse particles (> 1000 µm, mainly insufficiently ground grains) and lower proportions of fine (≤ 400 µm) and medium-sized particles (> 400 but ≤ 1000 µm) leading to higher GMD than in group 1 and 2 ($p \leq 0.05$). Indicating higher uniformity in particle size the GSD of group 3 was slightly lower than in group 2 ($p \leq 0.05$). Interestingly, Pearson correlations revealed that the higher samples' GMD the higher their starch and AME_N concentrations ($r=0.591$, $P < 0.001$) and the lower their mineral concentrations (ash: $r=-0.679$, calcium: $r=-0.674$, sodium: $r=-0.596$, $P < 0.001$). Accordingly, the coarser group 3 showed significantly higher starch and AME_N concentrations than the finer group 2 ($P < 0.01$). In conclusion, this study indicated that increasing coarseness and uniformity of particles in mash diets might serve as risk factors for feather pecking and cannibalism in hen flocks probably due to separation processes between mineral (fine) and starch containing (coarse) components in mash diets. Regarding differences in GMD, GSD and associated nutrient concentrations, sieving analysis for PSD determination in mash diets can be considered as valuable tool for diagnosing nutrition-related performance and health issues in hen flocks.

Keywords: Dry Sieving Analysis, Feed Coarseness, Hen Flocks, Mash Diets, Particle Size Distribution

Response of broiler chicken fed diets supplemented with dry powdered gluco- and manno-protein yeast cell wall extracts

Abstract ID: 21

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This study was designed to measure the response of broiler chickens fed diets containing inactive gluco- and manno-protein yeast cell wall powdered extract on their performance and physiological function. Nine diets based on maize and soybean were offered to 486 Ross 308 broiler chickens from 0 to 35d. The test diets contained enzymatically hydrolyzed inactive gluco- (HGT Auxoferm®) and manno-protein (MP Auxoferm®) yeast cell wall components/powder at four levels: (0.5, 1.0, 1.5 and 2.0 g/kg diet), respectively. These yeast extracts were compared with a control diet (without yeast supplementation) in a 2 x 4 + 1 factorial design. Each of the 9 treatments was replicated 6 times, with 9 birds per replicate. Birds were kept in a climate-controlled environment and fed on starter (1-10 d), grower (11-24 d) and finisher (25-35 d) diets. Feed and water were offered ad libitum. Feed intake (FI) and body weight gain (BWG) were measured on 10, 24 and 35 d, while FCR (corrected for mortality) was calculated from FI and BWG. On 10 and 24 d, one bird per replicate was electrically stunned, killed by cervical dislocation and visceral organs were weighed. At 35 d, two birds per replicate were euthanised and dissected to obtain the relative weight of breast, thighs and drumsticks. The dietary treatments did not have any effect on FI, BWG and FCR at 10 d. However at 24 d, there was an improvement ($P < 0.05$) in BWG and FCR for broiler chickens fed gluco- and manno-protein yeast cell wall extract at 2.0 g/kg diet compared to birds on the control diet. Except for the weight of the small intestine that was significantly increased ($P < 0.05$) for birds on higher levels of manno-protein compared to birds on the control diet at 24 d, the weight of all other visceral organs was not significantly affected ($P > 0.05$) by the dietary treatments at 10 and 24 d of age. At 35 d, there was significant improvement ($P < 0.05$) in BWG, FCR, dressing percentage of carcass, relative breast and drumstick weight for broiler chickens fed higher levels of both gluco- and manno-protein yeast cell wall extracts compared to birds on the control diet. In conclusion, supplementation of diets with inactive dried gluco- and manno-protein yeast cell wall extract/powder at 2.0 g/kg diet can improve broiler chicken performance and carcass yield.

Keywords: Broiler chickens, Glucan, Manno-protein, Yeast

The effect of feeding Diamond V fermentation metabolites on reducing Salmonella prevalence, numbers and NARMS panel antibiotic resistance in samples taken from commercial broiler breeder hens

Abstract ID: 285

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A field study was conducted in commercial broiler breeder hens to determine the effects of feeding a fermentation metabolites product (FM) on reducing the prevalence, numbers, and antibiotic resistance of *Salmonella*. Three breeder flocks within a single company were evaluated in this field study. Baseline samples taken prior to the application of the FM served as the control (CON). Samples included boot swabs (N=4/house) and cloacal swabs (100/house) taken when the flocks were 29, 33, and 37 weeks of age. The FM was then added to the commercial diet and fed at 1.25 kg/MT. Ten weeks later, when the breeder flocks were 39, 43, and 37 weeks of age, samples were taken again (boot swabs: N=8/house and cloacal swabs: N=100/house). Boot swabs and cloacal samples were shipped overnight to Iowa State University where they were analyzed for prevalence and numbers of *Salmonella*, with positive samples from cloacal swabs tested for antibiotic resistance using a panel of 19 different antibiotics as outlined in the National Antimicrobial Resistance Monitoring System (NARMS) program. Data were analyzed in SAS using the GLM procedure with feeding treatment as the main effect, house as a random effect, and significance considered at $P \le 0.05$. As measured with boot swabs, feeding the FM resulted in a not significant reduction ($P = 0.0780$) in *Salmonella* prevalence compared to CON (20.8% vs. 50.0% respectively), with a significant reduction ($P = 0.0437$) observed in *Salmonella* numbers (12.2 vs. 115.1 CFU/g). Cloacal swab *Salmonella* prevalence was significantly reduced ($P < 0.0001$) compared to CON (7.0% vs. 27.6%, respectively) after 10 weeks of feeding the FM. A significant reduction ($P < 0.0001$) was also observed for *Salmonella* numbers in the FM fed birds vs. CON (114.9 vs. 21,888.7 CFU/g of cloacal contents, respectively). A total of 19 different antibiotics, representing eight different classes of antibiotics are contained within the *Salmonella*-specific NARMS panel. A total of 10,248 individual *Salmonella* isolates obtained from cloacal swabs were tested, and the inclusion of the FM resulted in a significant reduction ($P < 0.05$) in the resistance of *Salmonella* isolates to 17 of the 19 antibiotics tested in the NARMS panel. These data suggest that the addition of the FM to the diet of commercial broiler breeder hens is an effective in-feed intervention for the reduction of *Salmonella* prevalence and numbers as measured either by boot or cloacal swabs. These data also suggest that the addition of the FM is an effective in-feed intervention for reducing the antibiotic resistance in *Salmonella*.

Keywords: Antibiotic, Breeders, Fermentation metabolites, Resistance, Salmonella

The effect of feed restriction on performance, organ development and blood picture of broiler chickens

Abstract ID: 229

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Feed restriction in broiler chickens has been applied to improve health and affects performance, body development and physiological characteristics. The aim of the study was to evaluate the effect of feed restriction and its intensity on performance, selected internal organs development and health status. In the experiment broiler cockerels Ross 308 were split into three groups of feeding regime, group one was fed *ad libitum* during the whole experiment, group two was restricted from 8th to 14th day of age and received 80% of *ad libitum* (R80) and group three was restricted at the same age, and was fed 65% *ad libitum* (R65). Restricted chickens before and following feed restriction period were fed *ad libitum*. Chickens were fattened until 35 days of age and organ percentage and blood picture were evaluated at 14 and 35 days of age. Final live weight was significantly ($P \leq 0.05$) lower in the group R65 (1965 g) compared to the *ad libitum* and R80 groups (2098 g and 2026 g). Feed restriction decreased ($P \leq 0.001$) feed intake in both restricted groups (94 % and 92 % of *ad libitum*), on the other hand, feed conversion ratio was not affected by feeding regime. Mortality of chickens was reduced in the restricted groups for 26 % in the group R80 and 42 % in the group R65. Liver percentage was significantly lower immediately after feed restriction as well as at the end of the experiment in both restricted groups, however, heart and stomach percentage was not affected by feeding regime and age. Blood picture measurements were significantly affected by age. Feed restriction increased percentage of lymphocytes immediately after restriction, whereas at 35 days the measurement was numerically higher in both restricted groups. Results of the study show that mainly restriction 65 % of *ad libitum* in the second week of fattening deteriorated performance of cockerels but had a positive effect on mortality. However, restriction on 80 % *ad libitum* is assumed to be suitable because cockerels reached a higher European production efficiency factor than the *ad libitum* fed cockerels.

Keywords: Blood picture, Chicken, Liver, Quantitative feed restriction

The effect of pelleting on starch digestion rate of soft and hard wheat in broiler chickens.

Abstract ID: 116

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Wheat can comprise up to 65% of a poultry diet as a source of energy and contributes 60-65% of the apparent metabolizable energy. Previous research has suggested soft wheat has lower AME than hard and durum wheat and this variation in terms of AME and digestibility may vary within wheat cultivars. A 2 × 4 factorial experiment (two wheat types and four different feed processing) was carried out to investigate the effect of feed processing of soft and hard wheat on starch damage level caused by processing and its effect on starch digestibility in broiler chickens. A total of 280-day-old male Ross 708 broiler chicks were fed up to 28 days of age. Each of the 8 treatments were assigned to 7 replications of five broiler chickens per cage. Starch damage (as a % of total starch) was higher in hard wheat than soft wheat ($P = 0.0001$). Body weight gain and feed intake were affected by feed processing ($P < 0.05$), but feed efficiency wasn't affected. There are no differences in the digestive tracts length, however, full and empty weight of organs and digestive tracts were affected by wheat type and feed processing ($P < 0.05$). Starch disappearance in anterior jejunum was higher in pelleted diets and particularly in pelleted and fine grinding diets ($P=0.006$). In pelleted diets both types of wheat (3038 vs 3015 kcal/kg soft vs hard) had similar AME values. In mash diets the AME values were lower and there was a difference between the wheat types (2885.15 vs 2787.29). SEM analysis revealed that starch granules in the ileum were almost completely digested in pelleted diet, however, in mash diets some starch granules remained intact. In conclusion, pelleting causes starch damage in both hard and soft wheat, increasing apparent metabolizable energy and grow rate in broiler chickens, it also reduces the difference between hard and soft wheat in regards to energy utilization and growth rate in broiler chickens.

Keywords: Broiler, Pelleting, SEM, Starch digestion, Wheat type

Valine requirement under low crude protein diets in Ross PM3 broiler chickens

Abstract ID: 464

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Monitoring dietary crude protein (CP) content participates to reducing feed cost, soybean meal usage, nitrogen and ammonia emissions, foot pad lesions and intestinal challenges. In order to successfully reduce dietary CP in broiler diets, attention must be paid to thet limiting amino acids (AA). In classical corn soybean meal diet, Valine is the fourth limiting AA after Met, Lys and Thr and its dietary content must therefore be adequately supplied. A total of 1920 Ross PM3 broilers were fed a common starter and grower diet from 0 to 9 and 10 to 21, days of age, respectively. From 22 to 36 days, broilers were attributed to one of the 8 experimental treatments, with 6 replicates (floor pens) of 40 broilers per treatment. The experimental design consisted in a high CP diet (T1; CP = 19%, dLys = 0.9%) and seven low CP diets (T2 to T8; CP = 16%, dLys = 0.9%). Those seven low CP diets differed in digestible Val to Lys (from 67 to 87%) by addition in L-Val. Growth performance (BW, ADG, ADFI, FCR,EPI) were evaluated . Abdominal fat and breast meat yields were evaluated by slaughtering 30 birds per treatment (5/pen). By contrast analysis, it was determined that the high CP diet was significantly lower in body weight at 36d, ADG and ADFI and higher in CV than all other treatments except the 67% dVal:Lys treatment. For FCR and EPI, the high CP diet was significantly higher and lower, respectively, than the T6 treatment (CP = 16%, dVal:Lys = 83%). The high CP diet was also significantly lower in abdominal fat and breast meat yield than all other treatments. ADG, FCR, EPI responded linearly and quadratically to dVal/Lys ratio. Homogeneity (coefficient of variation) only responded linearly while abdominal fat and breast meat yields did not respond significantly to Val addition. Using a quadratic and a curvilinear-plateau model, it was possible to estimate the dVal:Lys requirement for ADG (84 and 77%), FCR (96 and 103%) and EPI (86 and 85%). From the present trial, it can be concluded that reducing dietary CP improved growth performance and breast meat yield but only when Val was adequately supplied. Val requirement of modern finishing Ross PM3 broilers was estimated to be higher than usual recommendations and optimal requirement although depending on model and performance criteria is established at 80% dVal:Lys.

Keywords: Low Protein Diets, Requirement, Valine

Whole wheat supplemented with butyric acid influence growth performance, gut development and apparent ileal digestibility of protein and amino acids in broilers fed different protein sources

Abstract ID: 421

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896 one-d-old mixed-sex (Ross 308) broilers were used to evaluate the effects of protein source (PS), diet structure (DS) and butyric acid (BA) supplementation on growth performance, gut development and its morphology, carcass characteristics, apparent ileal digestibility (AID) of protein and amino acids (AA). A 2 × 2 × 2 factorial arrangement of 8 treatments with soybean meal (SBM) and rapeseed meal (RSM) diet was employed for 35 d. Whole wheat (WW) feeding significantly ($P < 0.05$) improved feed intake (FI), body weight gain (BWG) and, feed conversion ratio (FCR). Protein sources significantly influenced ($P < 0.05$) the growth performance of the birds. Broilers fed SBM based diets had significantly ($P < 0.05$) greater FI, BWG and FCR. Supplementation of BA also significantly ($P < 0.05$) improved BWG and FCR. Whole wheat fed birds had significantly ($P < 0.05$) lower weights of crop, duodenum, jejunum and ileum, whereas significantly ($P < 0.05$) greater weights of proventriculus, gizzard, and liver and more acidic gizzard pH. Butyric acid supplementation significantly ($P < 0.05$) increased the weights of liver and gizzard, whereas the weights of duodenum and ileum were significantly ($P < 0.05$) decreased. Whole wheat, SBM and BA significantly ($P < 0.05$) improved villus height (VH) and villus height to crypt depth ratio (VCR), whereas crypt depth (CD) was significantly ($P < 0.05$) reduced by these dietary factors. Apparent ileal digestibility of protein and most of the amino acids was significantly ($P < 0.05$) improved by WW, SBM and BA supplementation. The WW feeding resulted in significantly greater ($P < 0.05$) carcass weight (CW) with and without giblets, leg quarter yield (LQY), breast meat yield (BMY) and lower abdominal fat (AF). Soybean meal resulted in significantly greater ($P < 0.05$) CW with and without giblets, LQY, BMY and AF. Butyric acid supplementation significantly ($P < 0.05$) improved the BMY only. A WW based diet supplemented with BA may improve the gut health and ileal digestibility of protein and most of the amino acids resulting in improved growth performance and carcass yield in broilers.

Keywords: Amino acids, Broilers, Butyric acid, Digestibility, Whole wheat

ORAL PRESENTATIONS

Breeding and Genetics

Broiler body composition is transgenerationally affected by reduced dietary protein levels in breeder hens

Abstract ID: 437

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In mammalian species it has already been demonstrated that maternal undernutrition can program their offspring. However, far less is known about these possible programming effects in chickens. Therefore, an experiment was designed in which three generations of breeder hens (pure line A) were raised on different levels of dietary protein. The F0 generation was divided in a control group (C) on a standard diet and in a reduced protein group (RP) with a 25% balanced reduction in crude protein and amino acids during their entire lifespan. The female offspring of F0 was then subdivided in a C and RP group, resulting in 4 F1 breeder groups. Female progeny of these F1 breeders was then again raised as breeders of the F2 generation, which were all fed a C diet: C/C/C, C/RP/C, RP/C/C and RP/RP/C, with letters defining the feed treatments in F0/F1/F2 generations. All breeder hens were raised according to standard management guidelines and fed to reach the target body weight (BW). From sexual maturity onwards, all hens were artificially inseminated with semen from roosters fed a standard diet. Male progeny of F2 breeders was raised as broilers for 6 weeks on C or low protein diets (10% reduction in protein and amino acids), resulting in 8 treatments. Feed was provided *ad libitum*, light and temperature schedules followed standard management guidelines. Body composition of these broilers was examined at day 22, 29 and 37 by dual energy X-ray absorptiometry scanning (DEXA). These procedures were performed on living chickens, allowing the same broilers to be scanned at all three time points. On control diets, broilers descending from breeders receiving RP feed in the F0 generation (groups RP/C/C and RP/RP/C) had an increased fat percentage and decreased lean tissue percentage compared to broilers descending from breeders receiving C diets in the F0 generation (C/C/C and C/RP/C breeders). The same observations were made for broilers on the low protein diet, namely an increase in fat percentage and decrease in lean percentage when F0 grandparent breeders were fed an RP diet. Furthermore fat percentage increased with age, with the strongest increase between day 22 and 29, for broilers on both control and low protein diets. Simultaneously, lean percentage decreased with age, especially between day 22 and 29, again regardless of the current broiler diet. Together, it can be concluded that F0 RP breeder diets influenced F2 offspring body composition in a transgenerational way.

Keywords: Body composition, Broilers, DEXA, Transgenerational

Different evolutionary dynamics revealed by functional SNP classes in global chicken groups

Abstract ID: 207

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Evolutionary forces such as selection and genetic drift have played a huge role in the chicken diversification. Such forces led to genomic alterations (c.f. fixation of favorable alleles) accountable for the wide phenotypic variation present in chickens, and the differentiation of contemporary chicken breeds from their ancestral state. Consequently, the various functional classes of single nucleotide polymorphisms (SNP) might have been subjected to different evolutionary forces in various breeds which resulted in the variation of phenotypes. This study used data from the SYNBREED chicken diversity panel (SCDP), which encompassed DNA samples of a wide range of chicken breeds provided by partners across the world. The data consisted of 173 populations (3231 individuals) which were genotyped at 580K SNP loci with the Affymetrix Axiom® Genome-Wide Chicken Genotyping Array. Eighteen groups were established from these populations based on the chicken types. One group represented the wild type. The SNPs were classified into six functional classes. We estimated and compared derived allele frequency (AF) spectra and observed heterozygosity (H_o) across the various SNP classes. The differences in AF between synonymous and missense SNPs can be used to study whether amino acid changes were neutral or reflected selection. Due to the functional implication of missense SNPs, a higher rate of fixation or loss can indicate possible directional or purifying selection. We found that some breed types exhibited deviating patterns of AF spectra from the wild type. There were three patterns for the differences in AF distribution between missense and synonymous SNPs, i) for the wild type and many of the breed types, there was an over-representation of missense SNPs compared to synonymous ones towards the rare and common AF bins, indicating an erosion of some (presumably undesired) variants and fixation of favorable ones. ii) A more rapid fixation of missense SNPs than synonymous SNPs which was observed in some breeds, predominately the crested types, showing subjection to directional selection. iii) A lack of difference between the two SNP classes which was only observed in the egg laying types. Such similarities in the two classes may be explained by linkage drag. There was an excessively higher rate of fixation and loss of missense SNPs in the white layer type than in the rest of studied chicken types, indicating an intensive selection pressure with possibly small effective population size. The difference in the level of H_o in the six SNP classes were systematic across the studied breed types whereby the missense sites exhibited significantly lower levels of H_o than the rest of the SNP classes, except for the white layer type which had no difference in H_o between missense and synonymous sites. The results demonstrate that chicken breed types have experienced different evolutionary forces at the various SNP classes.

Keywords: Chicken diversity, Functional annotation, SNPs

Effect of long-term heat stress on production, egg quality and physiological traits in four experimental lines of layers differing in heat tolerance and feed efficiency

Abstract ID: 494

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This study compares the effects of heat exposure on production, egg quality and physiological traits in 4 experimental lines differing for traits important for heat tolerance and feed efficiency: the Fayoumi, a dwarf naked-neck leghorn line (LS) and two lines of Rhode Island Red divergently selected for high (R+) and low (R-) residual feed intake. A hundred and sixty-nine hens (50 Fayoumi, 48 LS, 34 R+ and 37 R- hens, representing the "control group", were reared under thermo-neutral (22°C) ambient temperature (AT) throughout the experiment. A hundred and eighty-six hens (50 Fayoumi, 46 LS, 43 R+ and 47 R- hens, representing the "stressed group", were reared at 22°C until 28 wk and then under high (32°C) AT until 32 wk of age. Body weights were measured at 28 and 32 weeks. Feed intake, residual feed intake, and egg mass were determined from 28 to 32 wk of age. Egg quality traits were recorded on three eggs per hen. Blood was taken from 12 randomly selected birds per genotype and AT group at 32 wk of age and blood pH, carbon dioxidepartial pressure (pCO_2); bicarbonate content (HCO_3) and triiodothyronine (T3) concentration were measured. Significant genotype x AT group interactions were observed for most traits. The Fayoumi exposed at high AT showed, for all traits, no significant difference from the control group. On the contrary, under heat-stress the R+ and R- showed significant reduction of feed intake (-26% and -24%, respectively), egg mass (-8%, -11%, respectively-), body weight (-5%, R+ and R-), albumen height (-13%, R+), yolk weight (-5%, R-), and fracture force (-15% for the R-). Compared to the controls the LS hens exposed to heat stress did not have reduced egg mass and altered egg quality traits but did exhibit a significant reduction of feed intake (-10%) and body weight (-5%). A significant reduction was observed for pCO_2 in the R+ and R- (-16% in both lines) and for HCO_3 in R+, R- and LS (12%, 10% and 11%, respectively), consistent with a respiratory alkalosis condition. A significant genotype x AT interaction was observed for the T3 concentration, mostly due to the significant T3 reduction in the heat stressed R+ hens but not in the other genotypes. In conclusion, the present study showed that Fayoumi hens exhibited greater thermo-tolerance compared with the other genotypes. Heat-stress affected negatively the R+ and R-, although the R+ seemed to cope better to heat decreasing T3.

Keywords: Feed efficiency, Heat stress, Heat tolerance, Layers

Evaluation of growth and egg production performance of Iranian improved indigenous hens in rural areas

Abstract ID: 609

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This study was carried out to investigate on growth rate, egg production performance and livability of Iranian improved indigenous chickens in rural area of five different Iranian provinces (Esfahan, Azarbayejangarbi, Fars, Khorasan Razavi, and Mazandaran). Accordingly, 1500 indigenous chickens were used in a Block Completely Randomised design. Initially, day-old chicks were provided from indigenous breeding center of each province and reared for 45 days. Then, for evaluation of their growth and egg production performance, two dominant climatic regions were determined for each province. In each climate, two towns and three villages in each town were chosen. Furthermore, six families were determined as experimental units in each village. Twenty one 45 day-old chicks with 7 to 1 hen to rooster ratio were delivered to each family. Chickens' body weight measured on 8, 12, 24, 48 and 72 weeks of age. Additionally, number of eggs produced by the hens in each family were recorded daily basis. Produced eggs also were collectively weighed and recorded biweekly. Total average body weight of the indigenous hens were 687.7, 974.8, 1491.6, 1795.2 and 1920.6 at 8, 12, 24, 48 and 72 weeks of age, respectively. Differences between average body weight for different provinces at these ages were significant ($P<0.05$), while climate had no effect on the average body weight. Average egg production performance was affected by province ($P<0.05$) while no effect observed for climates. Least average egg production belonged to Azarbayegangarbi (22.2%) that was significantly ($P<0.05$) lower than Esfahan (32.4%), Khorasan Razavi (39.1%), Fars (37.8%) and Mazandaran (37.8%) provinces. Province had significant effect on average egg weight in most stages and total production period ($P<0.05$), but climate had no effect on this parameter. Totally, according to the results, average egg production percentage, annually number of eggs and egg weight of improved Iranian indigenous hens in rural areas were 33.7%, 123 and 50.7 g, respectively. Average livability of the indigenous chickens also during rearing and laying periods in rural conditions were 81.4 and 63.5%, respectively.

Keywords: Egg production performance, Egg weight, Indigenous hen, Livability

Exploiting extreme phenotypes to investigate haplotype structure and detect signatures of selection for body weight in broilers

Abstract ID: 372

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In the present study, we used a total number (n=700) of male broilers with records on body weight at 35 days (BW35) by taking the 5% lower (n=350, average BW35=1876.5 g, L) and upper (n=350, average BW35=2417.4 g, H) tails of a broiler population (n=3,500). We performed haplotype blocks (HB) analysis in the two tail populations and detected signatures of selection using the Wright's fixation index F_{ST} and pooled (both tails) data. Number of HBs was lower in the H than in the L tail (34,005 vs. 38,442) while the average HB length was higher in the H when contrasted to the L tail (9878.5 bp vs. 7597.3 bp). The genome coverage by HBs was higher in the H when compared to the L tail (0.37 vs. 0.32). A total number of 53 signatures of selection dispersed in 18 autosomes were suggested by markers exhibiting F_{ST} values higher than 0.20. A number of 192 QTLs related to growth traits (e.g. carcass weight) were found to lie within genomic regions 50 kb downstream and upstream the selection signatures with 96 QTLs having been reported to affect body weight. The search for putative candidate genes within the specified regions revealed 67 positional candidate genes, some of them related to growth traits (e.g. *PSMB4, MYOM2, ATP8B2, CAMK2D)*. Three genes (*GNAO1, MYOM2* and *TPM3*) were found to participate in muscle contraction via functional enrichment analysis.

Keywords: Broilers, Extreme phenotypes, Haplotype blocks, Signatures of selection

Exploiting linkage disequilibrium in GWAS analysis

Abstract ID: 644

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A genome-wide association study (GWAS) is the first step to identify the genetic cause of a disease or the genetic base of a trait. Though being straightforward this approach has some daunting aspects that make it complicated and error prone, e.g. population stratification and correction for multiple testing. Many solutions have been suggested to overcome such problems over the years.
An additional problem that arises while analyzing polygenic traits is that the effects of single markers are so weak that they often do not exceed the significance threshold after correction. To overcome the drawbacks of the existing methods we propose a novel approach which exploits the linkage disequilibrium among genetic markers. For this aim, a GWAS was performed using a linear mixed model for chicken egg-related traits. To obtain Wald-statistics genotypic data was permuted to create 1000 permuted genotypic files and the same GWAS analysis was performed for these permuted data sets. Sliding windows comprising marker under investigation and a certain number of flanking markers were considered to calculate average Wald-statistic values. A heuristic approach based upon the observed variance in the average Wald-values from the permuted data sets was applied to assess the significance of the signal in the Wald-value from the real data set. The parameter settings were chosen such that no false positives were seen in the permuted data. These average values were then plotted against the physical positions of the markers on the chromosome to identify the associated loci. The results show that by applying our novel approach we could obtain more associated loci than with classical GWAS.

Keywords: GWAS, Permutation, Wald-statistic value

Gene-based mapping and pathway analysis of feather pecking in laying hens

Abstract ID: 551

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Feather pecking in laying hens is a frequent and serious economic and welfare issue and extensive research efforts have been put into this trait during the last decades. The motivation for this behavior is, however, still not completely clear. In a previous study, we identified genomic regions associated with feather pecking in a meta-analysis jointly exploiting selection signatures from divergently selected lines of White Leghorn as well as GWA analysis in a large F_2-design established from these lines. The animals were recorded for feather pecking at 27 weeks of age applying established ethograms. To gain insight into the biological meaning of these single marker results, we conducted post-GWAS enrichment and pathway analyses based on significantly associated genes. Therefore, the SNP content of the Illumina 60k SNP chip was mapped to genes annotated on chicken chromosomes GGA1-28 based on genome assembly galGal4. SNPs were allocated to genes according to the transcription-start and -end positions including exonic and intronic as well as variants 5kbp up- and downstream. In total, 23,010 out of the 29,376 SNPs analyzed in the initial mapping study were assigned to 10,758 genes. Based on the minimum p-value for each of these genes and a relaxed nominal significance level of 5×10^{-4}, a list of 284 significantly associated genes was defined that was subjected to enrichment and pathway analyses using the tools PANTHER and DAVID. Most notably, the gene list was found to be most significantly enriched for the GO terms neuron-neuron synaptic transmission and glycinergic synaptic transmission (biological process), extracellularly glycine-gated chloride channel activity and glycin binding (molecular function) as well as post-synaptic membrane (cellular component). The most enriched pathway was the p53 feedback loop. Detailed results will be presented at the conference. Furthermore, gene- and pathway-based association mapping results will be presented.

Keywords: Chicken, Feather pecking, Pathway analysis

Gene pool preservation in poultry- actuality and solutions

Abstract ID: 417

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Narrowing of the genetic diversity in farm poultry is getting more and more actual problem. Economical reasons lead to disappearing of numerous local and small scale breeds and populations. FAO supervises gene pool preservations programs worldwide, but this activity needs to be better coordinated and scientifically supported. The narrowing of genetic diversity results in vanishing of valuable genes and alleles and will negatively impact on the efficiency of poultry breeding in the future. To optimize preservation programs and use gene pool populations most efficiently, there are needed wide investigations of genetic processes, which occur in such populations, find the ways of preservation (as live birds, frozen sperm, conserved tissues or cells etc.), find the ways of use of such populations in experimental or commercial breeding programs, find the methods of restoration of vanished poultry breeds. All-Russian Institute of Farm Animal Genetics and Breeding carries out extended investigations in this field. It has a collection farm, where 40 chicken breeds, originated from Russia and other countries are being preserved and investigated. On the base of this material we carry out investigations of: genetic diversity in breeds; search for SNP-markers related to egg productivity (shell quality, egg mass and egg performance) and meat productivity (body weight, content of abdominal fat); develop methods of population reproduction by natural mating and artificial insemination (optimization of sex ratio, frequency and dosage of insemination); improvement of technique of cryopreservation of individual and mixed ejaculates (methods of sperm dilution, freezing, thawing), improvement of fertilization and early embryo livability after use of individual cryoprotected ejaculates. There is being established a bank of frozen sperm- currently it contains 18 breeds, totally about 600 dozes . There are developed methods of restoration of vanished unique breeds (unique disappeared Russian chicken breed Pavlovskaya). The gene pool breeds are used for creation of new colored breeds (Pushkinskaya), meat-type crosses for farmers and small producers, specialized chicken populations for vaccine production. The Institute investigates various theoretical aspects of gene resources preservation not only local and rare breeds, but also commercial strains, which lost their competitiveness, but still have some valuable traits . Research activity, related to this problem, should be intensified and better cooperated on the international level.

Keywords: Genetic resources, Poultry, Preservation, Rare breeds

Genetic relationship between the keel bone palpation score and performance traits in white layers

Abstract ID: 526

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The presence of keel bone damage in layers is a known problem since many years. Interactions between keel bone damage with traits as the housing system, nutrition, management and genetics have been proven. The aim of this study is to analyse the relationship between the subjective keel bone palpation score and relevant breeding characteristics in a white Leghorn breeding program. For two generations, the data of two pure lines (A, B) from the LSL breeding program were collected. The palpation was conducted in generation one at the age of 46 and 70 weeks of life and in the second generation at 82 weeks of life. In total, 17,926 hens were investigated by palpation with a four graded scoring system. At the age of 46 weeks of life 76.5 % in line A and 14.2 % in line B had a deformation or fracture in the keel bone. At 70 weeks of age the percentage of hens with keel bone indications was 73.4 % in line A and 15.8 % in line B. Layers from the second generation showed similar results. With 82 weeks of age the two lines varied in their incidence of keel bone indications between 79.4 % and 13.7 %, respectively. Based on a substantial intra-observer reliability test, the Kappa values for the palpation score range between 0.65- 0.7 for both lines. At 46 weeks of life the estimated heritability was h^2 = 0.30 for line A and h^2 = 0.15 for line B. In both lines the palpation score at 46 weeks of life was negatively correlated with the early egg number r_g= -0.24 to -0.54. Nearly no relationship was investigated for the traits body weight (r_g= -0.01 to -0.06), egg number at peak production (r_g= -0.02 to +0.12) and egg weight (r_g= -0.01 to +0.10). A slightly negative correlation was found with the early egg breaking strength (r_g= -0.13 to +0.04), however, the late egg breaking strength (r_g= -0.02 to +0.06) appears not to be affected by palpated keel bone indications.
The study showed the influence of selective breeding to reduce keel bone damage at the expense of a later onset of lay. Egg quality traits appeared not to be affected by the palpation score.

Keywords: Breeding, Keel bone damage, Layers, Palpation

Genome-wide association study for eggshell crystal structures in chicken

Abstract ID: 625

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Eggshell provides a relatively independent and stable internal environment. Improving eggshell quality could reduce the economic losses from the damaged egg. Eggshell crystal structure is the basis of eggshell quality. In this study, we constructed an F_2 population derived from the cross of White Leghorn and Dongxiang chickens and measured parameters of eggshell crystal structure. In total, 927 hens at 66 weeks from this F_2 population were selected for SNP genotyping. Eggshells were collected one per hen. The crystal parameters were determined using a Rigaku R-AXIS SPIDERX X-ray diffraction. The integral intensity of the first nine major diffraction peaks, total integral intensity (TA) and crystal orientation degree (OD) were obtained. Genome-wide association studies were performed using a mixed linear model. The results showed that TA had a positive correlation with the ultrastructure and common quality traits of eggshells. There was a negative correlation between TA and OD, indicating that the larger the crystals, the lower the degree of orientation, the more prone to random distribution. The coefficients of variances of TA and OD were 9.4% and 23.73%, and their SNP-based heritabilities were 0.23 and 0.06. And 621 SNPs located in 55.7 ~ 69.3 Mb in GGA1 were significantly associated with TA. According to their mRNA and protein expression in uterine tissue cells and their function, we suggested that DERA gene may play an important role in the regulation of eggshell crystal growth. There might be three possible pathways: a) DERA provides energy for the synthesis of matrix protein and ions transport in the uterine tissue; b) DERA reduces the stress of high Ca^{2+} concentration to cells to maintain cell activity, and provides Ca^{2+} stably for the growth of eggshell crystals; c) DERA affects the growth of eggshell crystals through its regulation on uterine fluid ATP concentration. The current study could provide now insight for the genetic mechanism of eggshell crystal structure and eggshell quality.

Keywords: Chicken, Crystal structure, Eggshell, GWAS, X-ray

Male germ line transplantation as an efficient technique of transgenesis in chicken

Abstract ID: 364

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The recent progress in primordial germ cells (PGC) derivation and long-term cultivation opened new avenues in reproductive biotechnology, particularly in chicken where embryo stem cells are not available. We recently developed a new technique of transgenesis in chickens using the transplantation of gene-modified PGCs directly into the testes of sterilized adult roosters. In this environment, the PGCs are able to mature into the functional spermatozoa and restore spermiogenesis. We will present this technique and document it by introduction of blue fluorescence protein (BFP) reporter into the genome of White Leghorn chicken. PGCs were isolated from the early embryo circulation, cultured in vitro and transduced by nucleofection of BFP-transposon and phi31C Integrase. BFP-positive clones were further subcultured and transplanted after 110 days of in vitro culture. Three months after transplantation, the first spermatozoa were observed in the seminal fluid. BFP was positively tested in DNA of the spermatozoa by PCR. The sperms were also used for artifitial insemination. Compared with the existing approaches, our novel technique is fast, efficient and reduces the number of experimental animals.

Keywords: Chicken germ cells, Spermiogenesis, Transgenic chicken

The Epigenetic Effects of Feeding Increased Methyl Donors to Japanese Quail

Abstract ID: 548

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Changes in DNA methylation leads to changes in gene expression and ultimately phenotypes, which can affect the health and growth of an organism. Environmental signals, including nutrition, are known to impact epigenetic modifications of DNA and a diet that is high in methyl donors (HiMet) may alter DNA methylation in progeny compared to a control (CON). The methyl donors included 7030 mg/kg choline chloride, 5 mg/kg betaine, 1.5 mg/kg Vitamin B12, 7.5 mg/kg folic acid, 12 mg/kg pyridoxine, and 99 mg/kg zinc sulfate in addition to nutrients endogenous to the dietary feedstuffs. In the following study, quail parents were fed either the HiMet or CON diet in the starter and layer diets, yielding the following treatments: HiMet-HiMet, HiMet-CON, CON-HiMet, and CON-CON. For the parent generation of this multi-generational trial, 300 Japanese Quail were placed in a brooder at day of hatch. 150 chicks received the CON diet while the remaining 150 received the Hi-MET diet. At 6 wk, half of each group continued on the same diet while the other half switched diets. Two generations of progeny from both treatments were evaluated, all receiving a control diet. To determine methylation, all samples were subject to bisulfite conversion and directly sequenced to interpret methylated and unmethylated regions of the genome. By evaluating the differentially methylated regions using Ingenuity Pathway Analysis, loci involved in immune function were affected by diet, specifically those associated with inflammation. Evaluation of egg wt from the parent flock indicated significantly larger eggs from the HiMet-HiMet and CON-HiMET treatments compared to eggs from the CON-CON and HiMet-CON treatments. When evaluating the first generation of progeny, the hatch weight of the chicks from parents who received HiMet-HiMet and CON-HiMET layer was significantly larger than those whose parents received the other diets ($p>0.0001$). However, in weeks 1 through 4 of grow out no significant difference in BW was detected. While there were no significant differences detected in BW of the second and third generations of progeny, significant differences in egg weight persisted. All comparisons were analyzed using JMP ANOVA. Feeding a high methyl donor diet may have an epigenetic effect by altering factors that are involved in immune function that could result in changes in performance. Further research is warranted on the manipulation of breeder diets to impart positive epigenetic effects on progeny because it may be a useful alternative to feed additives in market birds and be of significant economic impact to the poultry industry.

Keywords: DNA Methylation, Epigenetics, Methyl Donors, Performance

The Use of Axial-Transmission Ultrasound to determine Bone Quality in the Laying Hen

Abstract ID: 509

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Keel and long bone fracture are of concern for laying hen welfare. However, there is genetic potential within commercial layers to lay a high number of eggs and have good bone quality. Unfortunately, the methods used in genetic trials have relied on killing birds to make the phenotypic measurements followed by retrospective selection. A reliable measure of bone quality in the living hen is required if genetic selection is to offer a practical solution to improve bone quality.
Ultrasound can be used to assess bone quality in humans. Axial-transmission ultrasound uses emitters and receivers positioned on the same axis and the ultrasound wave is transmitted along the cortex. Axially transmitted speed of sound (SOS) indicates overall bone strength by reflecting properties such as mineral density, elasticity, cortical thickness and microstructure.
Bone is composite of apatite nanocrystals reinforcing an organic matrix. Using analytical techniques such as Fourier-transformed Infrared Spectroscopy (FTIR) and Two-dimensional X-ray Diffraction (2D-XRD), parameters related to chemical composition, degree of mineralization, crystallinity and structural organization of bone tissue, which influence the mechanical properties of bone, were measured. To produce a range of bone quality different levels of calcium (1.8, 3.6, 5.4%, n=20) were fed in a replicated design to laying hens for 6 weeks from 19 weeks of age. The proximal humerus was used to perform SOS measurements with a bone sonometer (Omnisense 7000, Sunlight Medical Inc., Israel). Correlations were made between the SOS data and biomechanical and physico-chemical measurements made post mortem.
SOS did not correlate with any of the biomechanical measurements.There was weak but significant correlation between SOS and bone mineral crystallinity as determined by both 2D-XRD (r=0.340; p=0.016; R^2=11.6%) and FTIR (r=0.307; p=0.024; R^2=9.4%).
The humeri in these birds were pneumatised and contained no medullary bone. However, in a separate experiment using commercial pedigree hens whose humeri contained varying amounts of medullary bone, there was a clear correlation between SOS and medullary bone content (n=190; r = 0.338; p<0.001; R^2=11.4%).
These results suggest that SOS may reflect some aspects of bone quality in the living hen, but that medullary bone may obscure the relationship between SOS and bone quality.
This work was funded by ERANET Animal Health and Welfare grant BB/M029829/1.

Keywords: Bone quality, Breeding, Selection, Ultrasound

Visceral fat of broiler and layer chickens and its apparent endocrine role in female reproduction

Abstract ID: 294

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The mammalian adipose tissue plays a central role in energy-balance control, whereas the avian visceral fat hardly expresses leptin, the key adipokine in mammals. Therefore, to assess the endocrine role of adipose tissue in birds, we compared the transcriptome and proteome between two metabolically different types of chickens, represented by the main commercial lines, broilers and layers, which were selected by breeding towards efficient meat and egg production, respectively. Broilers and layer hens grown up to sexual maturation under free-feeding conditions, differed 4.0-fold in weight and 1.6-fold in ovarian-follicle counts, yet the relative accumulation of visceral fat was comparable. RNA-seq and mass-spectrometry (MS) analyses of visceral fat revealed differentially expressed genes between broilers and layers, 1,106 at the mRNA level (FDR≤0.05), and 203 at the protein level (P≤0.05). In broilers, Ingenuity Pathway Analysis revealed activation of the PTEN-pathway, and in layers increased response to external signals. The expression pattern of genes encoding fat-secreted proteins in broilers and layers was characterized in the RNA-seq and MS data, as well as by qPCR on visceral fat under free feeding and 24 hr-feed deprivation. This characterization was expanded using available RNA-seq data of tissues from red junglefowl, and of visceral fat from broilers of different types. These comparisons revealed expression of new adipokines and secreted proteins (LCAT, LECT2, SERPINE2, SFTP1, ZP1, ZP3, APOV1, VTG1 and VTG2) at the mRNA and/or protein levels, with dynamic gene expression patterns in the selected chicken lines (except for *ZP1*; FDR/P≤0.05) and following feed deprivation (*NAMPT*, *SFTPA1* and *ZP3*) (P≤0.05). Interestingly, the majority of these newly identified gene products in visceral fat are implicated in female reproduction. Strikingly, some of the most prominent adipokines in mammals, leptin, *TNF*, *IFNG*, and *IL6* were expressed at a low (FPKM/RPKM<1) and non-differential mRNA level, in the divergently selected broiler and layer lines and in fed *vs* feed-deprived chickens ($P \leq 0.05$). In summary, our study revealed similarity in the relative accumulation of visceral fat in broiler and layer hens grown at *ad libitum* access to food, reflecting selective breeding effort to reduce fattening in broilers. Nevertheless, our comparative gene expression analysis, demonstrated a significant difference between broiler and layer hens in the endocrine activity of the visceral fat. This difference seems to relate to the strong difference in the growth and reproduction efficiencies between the two strains.

Keywords: Adipokines, Adipose tissue, Broilers, Layers, Reproduction

ORAL PRESENTATIONS

Eggs Safety and Quality

EDIL3 and MFGE8: key proteins in the biomineralization of the hen eggshell

Abstract ID: 403

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The eggshell is an important physical barrier against mechanical stress and microbial injuries allowing an harmonious development of the chick embryo. The egg is a basic ingredient for human consumption and the shell integrity is crucial to maintain its hygiene. The shell is made of 95% calcium carbonate (calcite), and 3.5% organic matrix (OM, proteins, proteoglycans...). Its formation results from an acellular biomineralization process in the uterus, involving a transitional amorphous calcium carbonate phase (ACC). The OM controls the ACC stabilization, promotes crystal nucleation, the calcite growth and drives the morphology and size of crystals. These OM-mineral interactions provide to the eggshell its ultrastructure and its mechanical properties. My PhD project focuses on two proteins (EDIL3, MFGE8) with predicted functions associated to calcification, and overabundant in the shell during the key events of the biomineralization process. We have first performed *in silico* analysis to characterize these two proteins and their corresponding genes. High similar organization of EDIL3 and MFGE8 proteins were determined with 55% identical residues and 2 similar domains in both proteins, including one EGF-like calcium-binding domain. Two distinct genes encode EDIL3 and MFGE8 on chromosomes Z and 10, respectively. The phylogenetic reconstruction suggests a duplication event of an ancestral gene for these two proteins at about 430 Ma. In a second approach, we have performed real-time quantitative RT-PCR in order to assess the expression of these genes in several tissues collected in the hen at different stages of the biomineralization. We observed a specific expression of *edil3* in the oviduct regions involved in shell calcification (isthmus and uterus). This transcript is overexpressed during the primary events of the process when the protein is overabundant. We quantified the level of abundance of these two proteins in the eggshell, the uterus and the uterine fluid using specific antibodies. We will also use *in situ* hybridization and immunohistochemistry to localize EDIL3 and MFGE8, in the uterine cell populations that synthesize these two proteins. In a third approach, we are purifying these two proteins using chromatography in order to analyze their interactions with the mineral phase. These data will characterize the role of this protein in the shell calcification process. Some polymorphisms present in the coding sequence of these proteins are already investigated on grandparents layer selected lines and will be associated to eggshell quality parameters for the selection of laying hens with reinforced eggshell quality.

Keywords: Biomineralization, EDIL3, Eggshell, Laying hen, MFGE8

Effectiveness of yeast cell wall in layers on intestinal and ovarian colonization of Salmonella enteritidis

Abstract ID: 469

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Salmonella enteritidis (SE) is currently the most common serotype associated with the human disease. In laying hens is especially important because colonize the ceca and internal organs, resulting in SE translocation to the ovary and as a consequence, can be found in eggs, by either ovarian or intestinal tract infection. The yeast cell wall (YCW) from *Saccharomyces cerevisiae*, highly concentrated in β-glucans and MOS has been demonstrated to be promisor. This study was designed to evaluate the use of YCW to mitigate intestinal and ovarian colonization by SE. For this, 200 Hyline W36 pullets at 10 weeks of age were distributed in 2 treatments: Control and YCW supplemented group (from *Saccharomyces cerevisiae* at 0.5 kg/MT, ImmunoWall® product from ICC Brazil Company). At 12 weeks of age all pullets were light stimulated, then at 16 weeks of age each bird was orally gavaged with a Nalidixic acid-resistant strain of SE ($3x10^9$ CFU/bird). On 7 and 14 days post-challenge hens were euthanized by cervical dislocation, cecas and ovaries aseptically removed, weighed and placed into sterile plastic bags. Prevalence of SE was determined by culture in tetrathionate (42°C) and after XLT-4 (25 mg/ml Nalidixic acid). Enumeration of SE in ceca was by the MPN method of Berghaus and Thayer (log 10). Statistical analysis was by Kruskal-Wallis test and Dunn's procedure ($P<0.05$). The ovarian prevalence was 41.7 and 4.2% from Control group *vs.* 33.3 and 2.1% in YCW supplemented group, at 7 and 14 days, respectively. The ceca prevalence was 93.8 and 47.9% by YCW *vs.* 97.9 and 53.2% by Control, at 7 and 14 days, respectively. The colonization (enumeration) was significantly reduced by YCW at 7 days (2.23 YCW *vs.* 2.70 Control, and 0.07 YCW *vs.* 0.07 Control, at 14 days). In summary, the SE challenge was effective in colonizing ceca and ovaries. The treatments with YCW did not reduce the prevalence of SE in the ceca, at this high challenge, but significantly reduce the level in those ceca. Also was effective in numerically lowering SE prevalence in ovaries at both 7 and 14 days post-challenge.

Keywords: Food safety, Nalidixic acid, Saccharomyces cerevisiae

Effect of dietary microalgae oil supplementation on fatty acid composition and sensory profile of table eggs in laying hens

Abstract ID: 247

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Microalgae is the original source of long-chain n-3 polyunsaturated fatty acids (PUFA) in the marine food chain and particularly rich in docosahexaenoic acid (DHA, C22:6n3). The effects of dietary supplementation with graded levels of microalgae oil on egg fatty acid composition, sensory properties were explored, and the relationship between yolk fatty acid and sensory attributes were further investigated. Five hundred and forty Hy-Line Brown hens (30 wk of age) were randomly assigned to 1 of the 6 dietary treatments with 6 replicates of 15 birds each. The diets were isonitrogenous and isoenergetic, containing 0, 0.25, 0.5, 1, 2.5, or 5% microalgae oil. The trial lasted 10 weeks. Compared to the control group, dietary 5% microalgae oil supplementation yielded a more than 9-fold increase in yolk total n-3 PUFA contents (36.46 mg/g yolk, $P<0.001$), and the significant variation of DHA contents contributed to ~84% of this increase. But yolks from hens fed 5% microalgae oil had lower scores of aroma, flavor and overall acceptability than those of other groups ($P<0.001$). Interestingly, dietary supplementation with 0.25% microalgae oil increased the scores of yolk overall acceptability compared with the control ($P<0.001$). Results from partial least squares analysis indicated a strong association between fishy aroma, fishy flavor, sour flavor and specific yolk fatty acids. Egg aroma and flavor were more related to OA (oleic acid, C18:1n9), SA (Stearic acid, C18:0) and AA (Arachidonic acid, C20:4n6). Fishy and seaweed aroma, and fishy and sour flavor appeared to be more associated with n-3 PUFAs, including DHA, ALA (α-Linolenic acid, C18:3n3) and ETE (Eicosatrienoic acid, C20:3n3). And sweet flavor revealed higher correlation with AA. In fact, acceptance scores of egg yolks decreased as the DHA ($R^2=0.807$) and total n-3 PUFAs concentration increased ($R^2=0.823$). The eggs were within acceptance when yolk DHA concentration was less than 16.44 mg/g. These findings would contribute to beneficially altering the fatty acid profile and sensory profile for the production of DHA enriched eggs by using microalgae oil.

Keywords: DHA, Eggs, Laying hens, Microalgae oil, Sensory profile

Effect of yolk on functional properties of chicken eggs

Abstract ID: 30

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Chicken eggs are widely used in the manufacturing of foods. The relation of egg contents (yolk to albumen) may affect the quality of pastries and yolk impurities in the albumen may affect the foam quality of albumen. Therefore, two experiments have been conducted aiming both at quantifying the effect of different yolk proportions on the quality of whole egg biscuits (Exp1) and at determining the effect of different yolk impurities in the albumen on foam quality (Exp2). In total, 240 and 256 fresh eggs, weight class L, have been used in Exp1 and for Epx2, purchased from a local layer farm, respectively. In Exp1 yolk proportions have been adjusted to 25, 28, 31, and 34 %. For preparation of the biscuit 180 g whole egg mass were mixed with 55 g wheat flour, 55 g corn starch and 106 g sugar. Exactly 60 g dough was baked in a convection oven for 20 min. at 185°C. The volume and the stability of the cake were measured (N=4x32=128 cakes). In Exp2, in total 100 ml albumen were prepared containing 0, 0.5, 1.0, 2.0 or 4.0% yolk. The albumen was mixed with the Hobart Mixer N50 for 180 sec. After mixing, the foam volume was measured and the durability of the foam was determined by recording the development of drainage within 30 and 60 min., respectively (N=5x10=50 foams). With increasing yolk proportions the cake volume increased from 248 to 276 ml ($P<0.05$) and the maximum force to cut the cake decreased from 8.5 to 6.7 N ($P<0.05$), respectively. Yolk impurities in the albumen decreased the foam volume significantly from 1175 to 470 ml. The biggest difference was observed between 0% yolk and 0.5% yolk (1175 and 900 ml, respectively), whereas, the volume decreased linearly from 0.5 to 4.0% impurity. In contrast, the drainage amount was highest for 4.0% impurity (56 ml), but did not differ much between the other treatments (38-44 ml). In conclusion, yolk proportion plays an important role in cake quality. As the yolk proportion decreased over years due to the breeding on high egg mass production the baking quality of today's chicken eggs deteriorated. Even small yolk impurities in the albumen significantly reduce the foam volume, whereas, the foam durability is less impaired.

Keywords: Albumen foam, Baking quality, Egg, Functional properties, Yolk

Evaluation of antioxidative capacity of egg by using the ORAC and SOAC assay

Abstract ID: 307

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Reactive oxygen species (ROS) was proved to be related to many multifactorial degenerative diseases such as diabetes, cardiovascular disease, cancer, neurodegenerative disorders and aging. Oxygen radical absorbance capacity (ORAC) and a singlet oxygen absorption capacity (SOAC) assay are two methods for evaluation of antioxidative capacity of food, while the ORAC assay focuses mainly on the hydrophilic antioxidants, the SOAC assay is performed in ethanol-chloroform-D_2O (50:50:1, v/v/v) solution focusing mainly on the lipophilic antioxidants. In the current study, after being adapted to a high-throughput 96-well microplate assay format, SOAC assay and ORAC assay were applied to evaluate the antioxidative capacity of 9 kinds of hen egg powder sold in Japan. Results showed that no significant difference among ORAC values of whole egg powder (WEP), egg white powder (EWP) and egg yolk powder (EYP). The highest SOAC value was observed in green tea-fed hen WEP (32.7 μmol α-toc E/g WEP), rice-fed hen WEP showed the lowest SOAC value (almost zero). Furthermore, compared with basic-fed hen EYP, green tea-fed hen EYP showed almost 3 times higher SOAC value (61.7 μmol α-toc E/g EYP) than that of basic-fed hen (23.5 μmol α-toc E/g EYP), the SOAC value of rice-fed hen EYP was only 3.6 μmol α-toc E/g EYP. At last, the SOAC values of both Japanese tea (green tea) and Chinese tea (brown tea) were measured, although SOAC value of Chinese tea (311.4 μmol α-toc E/g tea powder) was only half of that of Japanese tea (646.4 μmol α-toc E/g tea powder), the SOAC value of WEP from hen fed by 1% of Chinese tea was 18.1 μmol α-toc E/g WEP, while that of Japanese tea-fed hen was 31.5 μmol α-toc E/g WEP. This result showed that about 80% of antioxidant compounds in Japanese tea or Chinese tea could be transferred to hen eggs, especially in yolk part. Fed by tea should by an effective way to improve the antioxidative capacity of eggs.

Keywords: Antioxidative capacity, Egg, ORAC value, SOAC value

Influence of eggshell quality and egg white protein composition on Salmonella spp. contamination

Abstract ID: 343

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The hen egg contain all nutrients and vitamins necessary for the developing chick embryo and is also an inexpensive source of protein for the human diet. However, eggs are also associated with food borne diseases such as Salmonellosis. The egg contents are protected by the cuticle, eggshell mineral and shell membranes which act as an effective physical barrier to prevent bacterial ingress. Additionally, the egg white contains proteins (i.e. lysozyme, ovotransferrin, avidin) which exhibit potent antibacterial and bacteriostatic activities. Thus, food safety of the table egg is highly dependent on both eggshell quality and egg white composition. However, these characteristics are highly variable as they are dependent on many factors including hen age, genetics, diet and hen housing. We have analyzed changes in eggshell quality and egg white protein composition with hen age and evaluated how the associated changes affect the susceptibility of eggs to be contaminated by Salmonella. Freshly laid eggs from hens of different age groups (25, 35 and 52 weeks old) were exposed to a *Salmonella* suspension, and after 21 days the egg white contents were analyzed for *Salmonella* growth. The cuticle quality, eggshell thickness and porosity of *Salmonella* positive and negative eggs were evaluated using scanning electron microscopy (SEM) and attenuated total reflection-Fourier transform infrared spectroscopy (ATR-FTIR). Additionally, the concentrations of the main egg white proteins (ovalbumin, ovotransferrin, lysozyme,) were determined, in contamination -positive and -negative eggs, by ultra performance liquid chromatography (UPLC). The results showed that there were no significant differences regarding eggshell quality between Salmonella positive and negative eggs (e.g. eggshell thickness, P=0.186; Polysaccharides, P=0.605; CO3, P=0.804; Amides, P=0.641). The main difference between Salmonella positive and negative eggs was that the first ones have a significantly lower amount of antimicrobial egg white proteins (ovotransferrin = 12.458 % egg white proteins, lysozyme = 0.067 %) than the last ones (ovotransferrin = 16.478 %, lysozyme = 0.269 %). Another interesting finding was that the concentration of antimicrobial proteins increased with the age of hens. Thus, the increase in the concentration of proteins in eggs laid by older hens could in part compensate for the increasing risk of bacterial contamination of these eggs due to their poorer eggshell quality and internal quality. In conclusion, the cuticle and eggshell quality and integrity determine if bacteria can penetrate the shell or not. However *Salmonella* growth and survival in the egg contents during storage depends on egg white protein composition.

Keywords: Eggshell quality, Lysozyme, Ovotransferrin, Salmonella, UPLC

Production and characteristics of functional egg based products with high biological and nutritive value

Abstract ID: 117

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Functionality of a foodstuff depends on the presence of sufficient amounts of functional food ingredients (FFIs) in it; FFIs are the products decreasing the risks of alimentary diseases, maintaining and improving the health of consumers according to the data of evidentiary medical studies. The aim of the study presented was development and characterization of FFIs for subsequent inclusion into the functional egg based products with high biological and nutritive values. Initial stage of the research involved biofortification of chicken eggs (via hen diets) with organic forms of selenium (Se), vitamin E, and ω-3 polyunsaturated fatty acids (PUFAs). The diets were optimized on the basis of the analysis of livability, egg productivity, and feed efficiency in layers and morphological and chemical composition of the eggs. Supplementation of diets with aforementioned nutrients increased their contents in eggs: more than 2 times for Se, almost 3 times for vitamin E, more than 20 times for ω-3 PUFAs. These biofortified eggs were than processed to obtain macro-FFI for functional egg products, coagulated egg albumen (CEA) additionally enriched with calcium and/or iodine. The investigation of properties of this macro-FFI included determination of concentration of trypsin inhibitor (ovomucoid) in CEA, its antigenicity in vitro, nutritive and biological value in vivo. After heat treatment and soft acid-salt hydrolysis concentration of ovomucoid in the resulted CEA decreased two-fold; concentration of antigenic ovalbumin units in CEA was 2.2%. Biological and nutritive values of CEA were evaluated in trials on 50 growing Wistar rats (males) randomly allotted in three treatments. Control treatment (CT) were fed semi-synthetic diet (20% of casein on the caloric base). Experimental treatments (ET2 and ET3) were fed isocaloric and isonitrous diets with the substitution of EA and CEA, respectively, for 50% of casein. True protein digestibility was similar in all treatments. Protein efficiency ratio (PER) in CEA-fed ET3 (1.96±0.04) was significantly higher in compare to CT (1.49±0.05,$P<0.01$), and EA-fed ET2 (1.60±0.02, $P<0.05$). Fermentolyzate of EA (containing 58% of peptides with molecular weights 6.9-1.1 KDa and 21% with molecular weights <1.1 KDa) obtained by 5-hour hydrolysis of CEA by enzymatic preparation Protozyme-B was chosen for the production of micro-FFI – complex of peptides with zinc (Zn).The resulting concentration of organic Zn in this micro-FFI was 19 mg/g. FFIs developed were included into the receipts of two functional egg-based foodstuffs with high biological and nutritive value.

Keywords: Antigenicity, Biological value, Coagulated albumen of chicken egg, Function food ingredient

Proteomics and ultrastructural study of the Guinea fowl eggshell at key stages of the biomineralization

Abstract ID: 405

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The bird eggshell is a protective calcium carbonate ($CaCO_3$) layer, essential to the prevention against microbes. This complex biomineral is composed of 95% $CaCO_3$ (calcite) and 3.5% organic matrix (OM) that contains proteins and proteoglycans. The OM controls the nucleation, orientation and growth of calcite crystals and is made of more than 700 proteins. The precipitation of $CaCO_3$ is initiated by amorphous calcium carbonate (ACC), which is transformed into calcite during the eggshell biomineralization process. In the hen eggshell, calcite crystals follow the same orientation, perpendicular to the surface of the egg, which confers to the shell its resistance properties to mechanical injuries. The Guinea fowl eggshell ultrastructure presents a dramatic shift of the calcite crystal orientation at the middle of the eggshell deposition providing superior mechanical properties. Specific organic matrix proteins and/or differential of abundance of proteins are suspected to be involved in this process. In this context, our objective is to better characterize the calcification process of Guinea fowl eggshell in order to learn from this model. To reach this objective, we have investigated the Guinea fowl eggshell proteome at different stages of the biomineralization process. We sampled eggshells at different stages of mineralization: nucleation of crystals, shift of crystal orientation and at end of the calcification. OM was extracted from shells and proteins were separated using 15% SDS-PAGE gel, and 20 bands were excised at each stage. After trypsin digestion, resulting peptides were analyzed using nano-LC-MS/MS approach allowing the identification of 168 proteins. Amongst them 27 are shared between all stages and 62 are only found at stages when crystal shift occurs. In order to determine key proteins involved in the shift of crystals, we have compared these 168 proteins with eggshell proteomes from other avian species. We have identified several specific proteins not reported in other bird species. We will also explore the organo-mineral composition in the Guinea fowl and chicken eggshells, using high-resolution infra-red analyses Synchrotron SOLEIL, to get insight on the different calcification mechanisms in a comparative approach. This study is the first investigation on Guinea fowl eggshell matrix proteins, and highlight species-specific proteins, which could be involved in the shift of crystal orientation, and potentially responsible for the particular high mechanical properties observed for this species.

Keywords: Biomineralization, Chicken, Guinea fowl Eggshell, High-resolution Infra-red, Proteomics

ORAL PRESENTATIONS

Poultry Meat Quality and Safety

Characterization of productive traits and incidence of breast myopathies in a broiler chicken crossbred proposed for alternative production systems

Abstract ID: 607

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This study aimed to characterize growth performance, incidence and severity of foot pad dermatitis (FPD) and breast myopathies (white striping, WS; wooden breast, WB; spaghetti meat, SM), proximate composition of breast meat of a broiler chicken crossbred (CB; Hubbard RedBro ♂ x Ross 308 ♀), and to compare these results to those obtained from a fast-growing genotype (FG-F; Ross 308 ♀) widely used in Italy for organic production. 864 1-d-old chicks, equally divided in males (CB-M) and females (CB-F), were housed in 12 replicates/group. Similarly, 432 1-d-old FG-F were housed in 12 replicates. Body weight (BW) and feed consumption were recorded at 28, 45, 56, and 81d to calculate daily weight gain (DWG), daily feed intake (DFI) and feed conversion rate (FCR). At processing (81d), slaughter yields, incidence and severity of FPD and breast meat abnormalities (NORM, MOD, SEV) were evaluated. Twelve breasts/group were used to assess crude protein, fat, dry matter and ashes. At 81 d, CB-M reported higher BW (5,023 vs. 4,002 vs. 4,443 g, respectively for CB-M, CB-F, FG-F, $P<0.001$), DWG (63 vs. 51 vs. 56 g/bird/d; $P<0.001$) and DFI (180 vs. 156 vs. 150 g/bird/d; $P<0.001$). FG-F showed the lowest FCR (2.686 vs. 2.860 vs. 3.095, for FG-F, CB-M, CB-F; $P<0.001$). Slaughter yields exhibited similar values. CB-F showed higher percentage of birds without FPD (92 vs. 89 vs. 55%, respectively for CB-F, CB-M, FG-F; $P<0.001$), whereas FG-F reported the highest of those with SEV lesions (10 vs. 2 vs. 0%, respectively; $P<0.001$). FG-F exhibited the highest percentage of breasts with SEV myopathies (WS: 48 vs. 11 vs. 6%, respectively for FG-F, CB-M, CB-F; WB: 72 vs. 6 vs. 12%; SM: 30 vs. 4 vs. 7%; $P<0.001$). CB-F breasts had higher protein content than FG-F (24.8 vs. 23.6%; $P<0.05$). These results highlight the potential effects of using a crossbreed in alternative farming systems, where minimum slaughter age are imposed by law, rather than a fast-growing one to limit the incidence and severity of breast myopathies while maintaining high growth performance.

Keywords: Breast myopathy, Broiler chicken, Crossbreeding, Genotype, Meat quality

Comparison of protein profile by SDS-PAGE of broiler breast meat affected by white striping, wooden breast and spaghetti meat myopathies

Abstract ID: 408

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In the past decade, the broiler industry faced the occurrence of emerging muscular abnormalities (White Striping – WS, Wooden Breast – WB and Spaghetti Meat – SM) mainly affecting those high growth-rate hybrids exhibiting an increased breast muscle development. Although several studies have been carried out to evaluate the impact of these abnormalities on the main quality traits and technological properties of the meat, only few have been focused on the protein profile. In addition, no have simultaneously explored the effect of these abnormalities (WS, WB and SM) on the sarcoplasmic and myofibrillar protein profiles. For this purpose, 20 *Pectoralis major* muscles were selected from the same flock of male broilers (Ross 308, 45 day-old, weighing about 3.0 kg) and classified by visual appearance as: Normal (NORM), WS, WB and SM. After extracting the myofibrillar and the sarcoplasmic protein fractions, SDS-PAGE electrophoresis was run (in duplicate) on 5 mg of proteins by using 7.5% polyacrylamide hand-cast gels and loading a molecular-weight marker into each gel. After running, the gels were stained with Coomassie Brilliant Blue R-250 and images acquired by using a densitometer. With regard to the myofibrillar protein fraction, it was evidenced a remarkably higher desmin (53 kDa) content in WB in comparison with NORM muscles (5.1 *vs.* 4.5%; $P<0.05$), whereas WS and SM exhibited intermediate values (4.7 and 4.9%). This finding, together with the presence of vimentin (54 kDa), might be considered as an indicator of the regeneration processes taking place within the muscle tissue. On the contrary, if compared to NORM, no significant differences ($P>0.05$) were found in the relative abundance of the sarcoplasmic protein bands in WS, WB and SM affected muscles. In spite of these few differences, an increased number of high-molecular weight bands was observed in both the myofibrillar and the sarcoplasmic protein fraction of the abnormal muscles and hypothesized to arise from the degenerative processes associated with the occurrence of these abnormalities. Overall, it was confirmed that the occurrence of breast myopathies is associated with profound degenerative and regenerative processes involving both cytoskeletal (desmin and vimentin) and myofibrillar proteins and, if compared to WS and SM, the WB condition resulted in more severely impaired muscles.

Keywords: Broiler myopathies, Protein profile, Spaghetti meat, White striping, Wooden breast

Effect of myopathy occurrence on quality of broiler chicken breasts during storage

Abstract ID: 277

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Literature well describes quality traits of broiler breast with myopathies, but little is known about meat shelf life. Thus, 48 normal, 48 white striped (WS), and 48 wooden (WB) breasts were selected at slaughtering from male chickens (49 d) and kept at 4°C to measure technological traits, microbial total viable count (TVC), proximate composition, and fatty acid profile at 24, 72, 120, 168, 216, and 264 h. Data were analysed with meat type (normal, WS, WB), storage time, and interaction as main effects. The log10 CFU TVC/g were modelled by DMFit for microbial growth parameters. Shelf life end was defined at 7 log10 CFU/g. Normal breasts had lower weight than WS and WB (237 vs. 280 and 312 g); the former also exhibited lower cooking losses than WS and WB (22.0% vs. 23.8% vs. 26.9% ($P<0.001$). Normal meat showed higher redness than WS and WB (a*: 0.88 vs. 0.41 and 0.43; $P<0.001$). Regarding proximate composition, WB exhibited a higher water content compared to normal and WS samples (75.0% vs. 73.4% and 73.6%, lower protein (21.4% vs. 23.9%) and higher lipid content (1.88% vs. 1.09%) than normal breasts ($P<0.001$). Normal breasts had higher saturated fatty acids (FA) and lower polyunsaturated FA rates than WS and WB (30.5% vs. 35.4%; $P<0.001$). During storage, lightness decreased; yellowness showed the highest value after 24 h and the lowest value after 168 h (b*: 8.28 vs. 6.98; $P<0.05$). Cooking losses averaged 24.2% after 72 h, decreased until 22.0% between 120 and 168 h, and reached 26.6% between 216 and 264 h ($P<0.001$). The shear force was the highest at 24 h (5.08 kg/g), then decreased and reached the lowest value by 120 h (2.83 kg/g) ($P<0.001$). In normal meat, Lag phase of TVC (46.3 vs. 85.2 and 77.8 h) and shelf life (130 vs. 149 and 192 h) were shorter compared to WS and WB. In conclusion, proximate composition and technological traits were impaired by myopathies; however, overall quality of normal and abnormal meat had similar onset during storage, while microbial shelf life increased.

Keywords: Microbial spoilage, Shelf life, White striping, Wooden breast

European Union Approaches Toward Safe Poultry Meat

Abstract ID: 507

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Incidents of foodborne disease in humans have increased considerably world-wide in the last few years. Although the sources of infection are mostly unknown, poultry products have repeatedly been implicated as the main source.
Many reports during recent years have shown that *Campylobacter* spp. and *Salmonella* are the most common causes of human foodborne bacterial diseases linked to poultry. In some areas also verotoxin producing *Escherichia coli*, *Listeria, Yersinia*, *Staphylococcus aureus*, *Clostridium perfringens*, *Clostridium botulinum* and *Bacillus cereus* have surfaced as an additional foodborne pathogen. In addition, the development of antibiotic resistance in bacteria, which are common in both animals and humans, such as Methicillin Resistant *Staphylococcus aureus* (MRSA) and Extended-spectrum beta-lactamase (ESBL) bacteria, are also an emerging public health hazard.
Beside the current legislations, the main strategy to control microbial food borne hazards should include Good Animal Husbandry Practices (GAHPs) at the farm level, which should be applied to poultry houses and environment and the feed. In addition, reducing colonization by using feed additives, competitive exclusion treatment or vaccines is a possibility during transport and slaughtering. In all cases agent surveillance and monitoring programmes must be adapted and followed strictly in aim to allow early intervention.
The main strategy to control food borne infections in poultry should include: monitoring, cleaning the production pyramid from the top especially of vertically transmitted microorganism such as Salmonella by culling infected flocks, hatching egg sanitation and limiting introduction and spread of infections at the farm level through effective hygiene measures which should be applied in poultry houses, their environment and the feed. An intensive and sustained rodent control is essential and needs to be well planned and routinely performed and its effectiveness should be monitored. Household pets also constitute a serious hazard. Buildings therefore should be pet proof. In addition, reducing bacterial colonization by using feed additives, competitive exclusion or use of vaccines are further possibilities. In addition, effective education programs must be implemented to increase public awareness of the necessary measures to be taken for protection against foodborne contamination of food products.

Keywords: Antibiotic resistance, Campylobacter, EU legislations, Food safety, Salmonella

Evolution of macroscopic and microscopic broiler breast alteration related to White Striping

Abstract ID: 220

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White striping (WS) is a pathological modification typical of heavy broiler breast, macroscopically characterized by white striation running parallel to muscle fibers. This lesion changes breast characteristics and can cause a meat downgrade with related economic losses.The aim of this study was to develop a histological score for WS lesions in broilers and the evaluation of the evolution of macroscopic and microscopic lesions during a production cycle. For this study, superficial pectoral muscle of 138 broilers, euthanized at different ages (12, 25 and 51 days), were scored macroscopically for WS (0=absence, 1=mild, 2=severe), and sampled for histopathology analysis. Sections were cut at a thickness of 4 µm and stained with hematoxylin-eosin, then firstly observed to set up a grading of peculiar lesions. The following lesions were selected and scored: myocyte degeneration (score 1-4), presence of adipocytes (score 0-3), fibrosis (score 0-3), diffused inflammation (score 0-3) or vasculitis (score 0-3). The degree of association between WS macroscopic scores and the presence of each histological lesion selected was evaluated using the rank correlation by means of the Spearman's Rho coefficient. A strong correlation was found between WS and myocyte degeneration (Rho= 0,543 P=0,000), WS and presence of adipocytes (Rho= 0,683 P=0,000), WS and fibrosis (Rho= 0,406 P=0,000). Because of these results only these parameters were used to calculate an histological overall score. The overall score resulted to increase with time, as a result of the evolution of single lesions. At 12 and 25 days, myocytes resulted swollen and degenerated (average score 1.8 and 2.2), other lesions are rare (average score under 1). The absence of inflammation in this period led to suppose that the lesion originates from an apoptotic phenomena instead of an autoimmune mechanism. At 51 days, after degeneration, myocytes seems to be substituted by adipocytes and connective tissue, and the pathology becomes chronic. Diffused inflammation and vasculitis worsen progressively, but they are not strongly correlated with WS. In conclusion WS is confirmed to be a chronic alteration of muscle fibers beginning before the second week of age and immune system seems not to be involved in the onset of the pathology, but further studies will be necessary to better understand the pathogenesis of this disease.

Keywords: Broiler, Histopathology, Lesion score, White striping

Is meat of turkey breeders really different from that of standard turkeys?

Abstract ID: 337

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The technological, nutritional and sensorial quality of breasts and thighs with shanks of turkey male and female breeders was characterized by comparison with breasts and thighs with shanks of growing male and female turkeys from Grademaker line (Hybrid Turkeys, n = 20 birds per sex and per physiological stage). The turkey breeders were slaughtered at 397 and 410 days of age and 10.42 and 32.67 kg of body weight for the females and males, respectively. The growing turkeys (standard birds) were slaughtered at 75 and 103 days of age and 5.89 and 13.48 kg of body weight for the females and males, respectively. Water loss after cooking was more important for the male fillets compared to females (15.3 % vs 11% for breeders and 9.8% vs 8,6% for standard respectively) and more important for breeders compared to growing line. In the other side, male fillets showed lower technological yield than the one of females (72.4 % vs 86.4% for breeders and 81.3% vs 84.3% for standard respectively). Furthermore, breeders fillets had more lipids and minerals than growing turkeys (3% and 2.5 % for femelle and male breeders vs 1% for both female and male standard respectively). The differences observed between males and females on one hand and between standard and breeder turkeys on the other hand were mainly induced by differences in slaughter ages and sexual dimorphism on body weight. The meat of female breeders has characteristics close to those of female and male standard turkeys whereas the meat of male breeders is clearly distinguishable.

Keywords: Breeders, Carcass, Meat Quality, Turkey

Occurrence and characterization of biofilms in drinking water systems of broiler houses

Abstract ID: 238

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Water quality in the drinking water system (DWS) plays an important role in the general health and performance of broiler chickens. Conditions in the DWS of broilers are ideal for microbial biofilm formation. Since pathogens might reside within these biofilms, they are a potential source of waterborne transmission of pathogens to livestock and humans, thereby causing a health risk. Yet, knowledge about the presence, importance and composition of biofilms in the DWS of broilers is largely missing. In this study, we therefore aim to monitor the occurrence, and chemically and microbiologically characterize biofilms in DWS. The bacterial load in DWS was determined by enumerations of total aerobic flora (TAC) and *Pseudomonas* spp.. The dominant flora was identified based on the 16S rRNA gene sequence and their biofilm-forming capacity was evaluated. Also, proteins, carbohydrates and uronic acids were quantified to analyse the presence of extracellular polymeric substances of biofilms. Despite disinfection of the water and the DWS, average TAC was 6.03±1.53 log CFU/20cm^2 ranging from 1.87 to 9.00 log CFU/20cm^2. Enumerations for *Pseudomonas* spp. were on average 0.88 log CFU/20cm^2 lower. Among the dominant flora collected from TAC, three species, *Stenotrophomonas maltophilia*, *Pseudomonas geniculata* and *Pseudomonas aeruginosa*, were the most identified, i.e. respectively 17, 14 and 11% of the isolates. Yet at species level, most of the identified microorganisms were farm specific. Almost all the isolates belonging to the three most abundant species were strong biofilm producers. Overall, 92% of all tested microorganisms were able to form biofilm under lab conditions. Furthermore, 63% of the DWS surfaces appeared to be contaminated with microorganisms combined with at least one of the analyzed chemical components, which is indicative for the presence of biofilm. The three earlier mentioned dominant species are also considered as opportunistic pathogens and could consequently be a potential risk for animal health. Additionally, the biofilm-forming capacity of these organisms could promote attachment of other pathogenic bacteria such as *Campylobacter* spp. and *Salmonella* spp.

Keywords: Biofilm, Broiler, Drinking water system, Pseudomonas spp., Stenotrophomonas maltophilia

Poultry meat quality and functionally food

Abstract ID: 642

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New concepts indicate that there is a tendency of production animals to not only fulfill their requirements caused by the progress in genetics, but also to provide an enriched food that fulfills the human nutritional needs together with an additional function, such as health promotion or disease prevention. Animal body composition undergoes continuous turnover. In monogastric animals, there is a close relationship between the diet and body lipid composition. There is a health interest in omega 3 fatty acids such as linolenic (ALA) eicosapentaenoic (EPA) and docosahexaenoic (DHA) omega 3 fatty acids. They differ in their function. For example, ALA, found mainly in vegetable oils, is more a source of energy and metabolic support, alternatively, EPA, found in algae or in fish that ate algae, fights off cardiovascular diseases and has an anti-inflammatory role. DHA, found in algae or in fish that ate algae, is important for brain and eye development, immunity, fertility and heart health. Metabolically, there are some degrees of conversion and retroconversion among ALA, EPA and DHA. Conjugated linoleic acids (CLA) can be formed by heating, refining, or partial hydrogenation of oils. Increasing dietary CLA results in a reduction of chicken abdominal fat content, an increase in saturated fatty acids, but not in unsaturated fatty acids. For humans, CLA fights off cancer and obesity. Chicken meat is also a good source of minerals, mainly selenium, phosphorus, potassium, magnesium, iron and zinc. Meat can be enriched with selenium (Se) when the chicken is fed a diet containing an organic selenium source. There are many different organic selenium forms available on the market and they are not all the same, having different responses in the bird. In addition to a decreased meat drip loss and increased in shelf life, Se-enriched chicken meat fed to humans can help to prevent cancer, arthritis, cardiovascular diseases, abortion, susceptibility to disease, thyroid hormones problems and much more. Chicken meat is also a good source of B-vitamin complex. The main interest in enriching meat with vitamins is with the liposoluble tocopherol, as vitamin E is a powerful natural antioxidant. It protects cell membranes from peroxidation, providing a longer shelf life. Therefore, human health status can be enhanced by feeding chicken with appropriate ingredients to enrich the chicken meat.

Keywords: DHA enriched meat, Meat quality, Selenium enriched meat

Raw and cooked meat texture as affected by white striping, wooden breast and spaghetti meat myopathies

Abstract ID: 352

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In recent past, several studies have evaluated the impact of emerging poultry meat abnormalities (White Striping, WS; Wooden Breast, WB; Spaghetti Meat, SM) on the main meat quality traits. Although their detrimental effects on meat quality have been widely established, there have been no studies carried out by simultaneously evaluating the effect of muscular abnormalities on raw and cooked meat texture. For this purpose, forty-eight *Pectoralis major* muscles were selected from the same flock (Ross 308, 45 day-old, weighing about 3.0 kg) and classified as follows according to the presence of breast abnormalities: 12 Normal (N), 12 WS, 12 WB and 12 SM. Peak force (kg) measurements were carried out through a 40% compression test using a TA-Hdi Texture Analyser at a speed test of 3mm/sec on raw and cooked meat samples (1×1.5 cm), obtained after cutting the fillets in order to separate the superficial section of muscle from the deep one. Raw meat texture was found to be significantly modified by myopathies both in the superficial and deep layer of breast muscle. Regardless the sampling position, if compared to normal samples, WB fillets exhibited a significantly higher hardness (superficial: 4.45 *vs.* 2.95 kg; deep: 3.41 *vs.* 2.01 kg; $P<0.001$), while WS and SM showed intermediate values (superficial: 3.53 and 3.55 kg; deep: 2.88 and 2.92 kg). This finding can be likely due to the massive accumulation of connective tissue detected in WB fillets by previous histopathological studies. Otherwise, following cooking, superficial portion of SM exhibited softer texture if compared with normal, while WS and WB did not differ from each other (2.65 *vs.* 3.36, 3.41 and 2.95 kg; $P<0.05$). Thus, overall these findings showed that: i) WS did not affect both raw and cooked meat texture; ii) WB was superficially harder only before cooking; iii) SM became superficially softer following cooking.

Keywords: Broiler myopathies, Spaghetti meat, Texture, White striping, Wooden breast

Rearing factors affecting myopathy rate and meat quality in broiler chickens

Abstract ID: 264

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The present work aimed at evaluating the effect of some rearing factors on performance and myopathy rates of broiler chickens slaughtered at 45 d of age. A total of 768 chicks were housed in 16 pens (48/pen) according to a factorial arrangement: 2 genders x 2 pens shapes (short, 2.4 x 2.5 m vs. long, 1.2 x 5 m) x 2 feeding treatments (standard diet vs. the same diet supplemented with 150 mg sodium butyrate/kg). In the long pens, feeders and drinkers were placed at the opposite sides to increase animal activity. Performance and meat quality data were analysed by a mixed model with gender, pen shape, dietary treatment, and interactions as the main effects, and pen as random effect. The same effects on myopathy rates were analysed by PROC CATMOD. Final live weight was higher in males than in females (3586 vs. 2965 g; $P<0.001$) and in broilers from short pens compared to those from long ones (3327 vs. 3234 g; $P<0.001$). The Na-supplementation had no effect on performance. At gross examination, 35.9%, 14.6%, and 14.1% of breasts showed white striping (WS), wooden breast (WB), or spaghetti meat (SM), respectively. Breasts with SM and WB also showed WS. Gender did not affect WS, whereas WB rates were lower (7.28% vs. 21.9%; $P<0.01$) and SM rates higher (25.2% vs. 3.13%; $P<0.001$) in females than males. Pen shape and dietary treatment did not affect myopathy rate. However, females fed Na-butyrate tended to show a lower SM rate (16.7% vs. 33.3%; $P=0.06$). Myopathies affected meat quality: WB had higher lightness index (50.7 vs. 48.2, on average), cooking losses (29.9% vs. 24.2%), and shear force (3.85 vs. 2.85 kg/g) than other meat types ($P<0.01$). In conclusion, WB occurred more in males than females, whereas SM showed an opposite trend. Differences in SM rate according to Na-supplementation deserve further investigations for confirmation and clarification of the action mechanisms.

Keywords: Spaghetti meat, White Striping, Wooden Breast

Volatile fingerprints of raw and cooked meat of different commercial chicken genotypes

Abstract ID: 529

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Volatile organic compounds (VOCs), e.g. odour and aroma, are important for the consumer acceptability of food. Is well known that the cooking process induce lipid oxidation and chicken meat, compared to red meat (e.g. calf), is more susceptible to this type of deterioration mainly due to the high level of unsaturated fatty acids. Therefore, the analysis of VOCs appears a powerful tool to evaluate the quality of meat products. The aim of this study was to evaluate the aroma in raw and cooked meat samples of 4 chicken genotypes with different growth rates. Ross was chosen as fast-growing genotype, Campese and Naked Neck as medium-growing and Leghorn as slow-growing. Two crossbreeds (Leghorn x Sassó and Leghorn x Ross) were studied as well. Furthermore, we investigated the influence of the rearing system (i.e. organic, conventional, free-range) and sex (for both Ross and Campese). The volatile fingerprints of non-cooked and cooked meat were measured by proton-transfer mass spectrometry (PTR-MS) to establish the quantity of VOCs, whereas the molecule identification was performed with PTR-TOF-MS and GC-MS. All obtained data were subjected to multivariate data analysis. Eleven main peaks related to fatty acid profile have been identified (Methanol, Ethanal, Ethanol, Butanal, Pentanal, Hexanal, Xylenes, Heptanal, Octanale, Penthyl furan, Nonanale). The results showed that the cooking process increased the release of VOCs in comparison with the raw meat (5 and 250-fold higher for the Methanol and Ethanal, respectively). As expected, also the genotype strongly influenced the VOCs. In fact, PCA demonstrates 6 spatially separated clusters one for each genotype in both cooked and non-cooked samples. The effect of sex was evident but appeared to be genotype dependent. In fact, this discrimination between genetic strains was clearly visible in a medium-growing genotypes (Campese) whereas, in fast-growing genotype (Ross) this distinction was not apparent.

The breeding system is another variable which affected VOCs. Ross genotype, reared under organic conditions, showed different VOCs profile than their conventional counterparts.

In conclusion, all the analysed variables affected the aroma's release mainly in cooked meat samples and the effect of genotypes was also relevant.

Accordingly, PTR-MS could be a suitable technique for VOCs fingerprints profiling between different genotypes, rearing systems and sex in poultry meat.

Keywords: Aroma, Genotypes, Rearing Systems, Volatile Organic Compounds

White striping prevalence and its effect on meat quality of broiler chicken breast fillets under commercial conditions

Abstract ID: 628

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White striping (WS) characterized by white striations on *Pectoralis major* muscle is an emergent problem for broiler industry, thus the impact of WS on meat quality has become a current research topic nowadays. From this viewpoint, a two-year follow-up study to reveal the prevalence of WS in broiler integrations was carried out in Turkey. As a second part of the study, a laboratory experiment was conducted to determine the differences in proximate composition, color, fatty acid and amino acid profile of breast fillets scored visually as normal (no striations), moderate (striations<1 mm thick), and severe (striations>1 mm thick). In addition, the effect of WS on oxidative quality of those breast fillets was also examined with storage study at 4°C and -18°C. The two-year follow-up study indicated that the WS incidence increased with increasing age and more than 50% of broiler breast fillets obtained from 32-35 and 36-39 day of age had white stripes with different scores. Lower protein and higher fat content were measured in moderate and severe scored fillets ($P<0.01$). Stripes with severe score induced the formation of less redness color on the ventral surface ($P<0.05$) and darker color on the dorsal surface ($P<0.01$). The individual amino acid amount significantly decreased with increasing level of WS due to possible effect of muscle degeneration and high collagen content ($P<0.05$). Moderate and severe scored fillets were rich in polyunsaturated fatty acids, especially oleic, linoleic and linolenic acid, which referred that they were sensitive to lipid oxidation. That judgement was also proved with higher TBARS values in moderate and severe striped fillets during both storage conditions. Moreover, higher carbonyl and lower sulphydryl content were measured in all breast fillets stored at 4°C. The freeze-thawing induced the formation of carbonyl compounds but it did not differ –*SH* groups. In brief, different scores of WS may affects the physicochemical and oxidative quality of broiler breast fillets, however more future studies are needed to assert an accurate and clear judgement.

Keywords: Amino acid profile, Oxidation, Physicochemical parameters, White striping

ORAL PRESENTATIONS

Reproduction and Incubation

Applying cold incubation profiles during the last week of incubation in a commercial incubator: effects on broiler embryonic mortality, hatchability, and chick quality

Abstract ID: 324

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During incubation, an eggshell temperature (EST, as a reflection of embryo temperature) of 37.8°C was long considered to be optimal for broiler embryonic development. However, an EST of 36.7°C (Cold) from embryonic day (E)15 onward may result in a more developed heart at hatching than 37.8°C EST throughout (Control; Maatjens *et al.*, 2016). Maatjens *et al.*'s study was performed in large incubation chambers with low air velocity, unlike commercial practice. To study Cold EST in a commercial situation with high air velocity and egg density, three trials were conducted. In all trials, EST for Cold was maintained at 37.8°C, decreased to 36.7°C with varying profiles in the last week, and then maintained at 36.7°C till hatching. Cold treatments were always compared to Control (37.8°C EST throughout incubation). 3,000-10,800 broiler eggs from a 30-39 week old parent flock were used. Firstly, EST was decreased within 30 minutes, on E15. Compared to Control, Cold resulted in 2.5x higher embryonic mortality around the time of the EST decrease (P = 0.015), 2.2% more second grade chicks (P = 0.049), and chicks were 0.4cm shorter (indicating lower development) at hatch (P = 0.001). Possibly, the EST decrease happened too early or abruptly. Secondly, EST was decreased gradually in 1 day, from E16-E17. No differences were found in hatchability (P = 0.68) or chick length (P = 0.93), but embryonic mortality around the time of the EST decrease tended to be 1.7x higher for Cold than for Control (P = 0.070). It was thought that an even slower EST decrease may optimize Cold further. Thirdly, EST was decreased using three different profiles. EST was decreased gradually from E16-E17, or quickly (to 36.9°C on E17) and then slowly (to 36.7°C on E18), or slowly (to 37.5°C on E17) and then quickly (to 36.7°C on E18). Embryonic mortality, hatchability, and navel quality did not differ between the Cold profiles and Control (P > 0.29). To conclude, results differed from the low air velocity trial situation. When EST was decreased from 37.8°C to 36.7°C abruptly or too early in development, hatchability and chick quality decreased. A slow transition from E16-E18 can result in chick quality and hatchability similar to Control. Knowledge on these optimal EST decrease profiles can be used to further investigate the effect of Cold incubation during the last days of incubation on post hatch performance and possibly apply it to commercial practice.

Keywords: Chick quality, Eggshell temperature, Hatchability, Incubation

Assessment of GnRH analogues efficacy in enhancing the reproductive functionality in broiler breeders

Abstract ID: 591

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Meat type hens are found to be more prone to Erratic Oviposition and Defective Egg Syndrome than egg type which includes follicular atresia, internal ovulation and laying, soft shelled and multiple yolk eggs and ovipositions that are not occurring in sequence. Even though feed restriction is practiced to be a viable option, it has raised welfare oriented concerns. Therefore, considering this research problem and with a goal of addressing reproductive anomalies, we have ascertained the influence of Gonadotropin Releasing Hormone (GnRH) analogue in broiler breeders. Seventeen weeks old broiler breeders were randomly divided into three groups (n=10) and injected with GnRH analogue-Buserelin acetate (Receptal™) @ 200 µg-0.2 ml (T_2) and 400 µg-0.4 ml (T_3) intramuscularly on weekly basis for a period of 10 weeks. Control (T_1) birds were injected with distilled water. One way ANOVA generated records on egg production indicated a significant ($P<0.05$) difference, with T_2 having higher egg number ($34.88^a \pm 2.82$) followed by T_1 ($34.16^b \pm 3.3$) and T_2 ($32.77^b \pm 3.15$). Further, Hen House Egg Production (HHEP) also indicated that low dose of GnRH analogue resulted in higher (314) than control (284) and T_3 (273) groups. Similarly, this group (T_2) was also found to have a reduced (4.17%) incidence of double yolk eggs than T_1 (6.81%) and T_3 (5.37) groups. Higher (5.1%) recurrence of blood spots in T_2 group is indicative of rapid ovulation and oviposition. Yolk colour was found to be mildly influenced in low dose treated group having 7.68±0.2. Other estimates like shell colour, shape index and egg weight was not influenced significantly under GnRH analogue treatment. As far as ovarian morphology is concerned, higher number of pre ovulatory follicles was observed with regards to low dose treated groups than others. Ovary with ovarian follicles weighed $44.65^b \pm 5.85$g, $72.36^a \pm 4.82$g and $41.15^b \pm 3.56$ g for T_3, T_2 and T_1 groups respectively. Low dose (T_2) treated birds also showed significantly ($P<0.05$) higher liver weights of $57.13^a \pm 6.45$ g than higher dose ($37.06^c \pm 4.58$g) and control groups ($41.4^b \pm 7.53$g) respectively. Further, 200 µg of GnRH analogue improved the oviduct weights ($68.44^a \pm$ 10.36g) than others. Fertility studies also indicated a higher fertility in low dose treated group (71%) followed by high dose (56.63%) and control (68.33) respectively. T_3 had higher (0.76±0.14 IU/L) of LH followed by T_2 (0.64±0.11). While low GnRH dose resulted in lowered concentrations (23.39 ±1.22 µg/L) significantly ($P<0.05$). In conclusion, lower dose (200 µg) of GnRH analogue significantly ($P<0.05$) improves HHEP along with ovarian and oviduct health and reduced reproductive anomalies in broiler breeders.

Keywords: Broiler breeder, Double yolk eggs, GnRH analogue, Ovarian health

Both the rooster and incubation temperature affect embryonic metabolism and day-old chicken quality in laying hens

Abstract ID: 100

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During incubation, the main factors driving embryonic metabolism and development are nutrient availability, oxygen availability and embryo temperature. Both nutrient and oxygen availability are expected to be particularly affected by the hen and thus the hen is thought to majorly determining embryonic metabolism and day-old chicken quality. However, in wild birds it has been suggested that the rooster is of influence on offspring quality, directly or via affecting egg size and egg composition. In poultry, the role of the rooster in embryonic development and metabolism is hardly investigated. In case the rooster affects egg composition, this can mean that the incubation temperature needs to be adjusted to obtain optimal embryo temperature. The role of incubation temperature on embryonic metabolism and development in the broiler chicken is extensively investigated, but much less information is available regarding laying hen chickens. The aim of the experiment was to investigate the role of the rooster and the incubation temperature on laying hen embryonic development and chicken quality. Eggs of two genetic crossbreds (AB and BB; 51 to 59 weeks of age) were used. Hens originated from the same breeder flock, were housed at the same farm, obtained the same management and diet but were mated with a different rooster. In six consecutive batches, eggs of both crossbreds (59.0 to 61.0 gram) were incubated at an eggshell temperature (EST) of 37.8^{o}C during the first 14.5 days and at an EST of 36.7, 37.8 or 38.9^{o}C from day 14.5 of incubation onward. In all batches, eggs of both crossbreds were used, but EST differed among batches. Egg composition was determined in fresh eggs, heat production was determined between day 14.5 and 18.5 of incubation and day-old chicken quality was determined at 6 hours after hatching or at pulling. Yolk weight tended to be higher (Δ=0.28 gram; P=0.08) in AB than in BB crossbreds, whereas other egg components did not differ. Heat production between day 14.5 and 18.5 of incubation was higher in the AB than in the BB crossbred (Δ=2.61%; P<0.001). At pulling, AB chickens were lighter, had less red hocks, more red beaks and worse navel scores than BB chickens. An EST of 36.7^{o}C resulted in later hatching time, higher heart weight and higher intestine weight than an EST of 38.9^{o}C. It can be concluded that both the rooster and incubation temperature appears to affect embryonic metabolism and day-old chicken quality.

Keywords: Chicken quality, Embryonic heat production, Incubation temperature, Laying hens, Rooster

Changes in Gizzard Development Caused by High Incubation Temperatures in the Chicken (*Gallus gallus*) Embryo

Abstract ID: 94

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Large deviations from the ideal incubation temperature of 37.8ºC have been known to interfere with chick development, hatchability and post-hatch survival and performance. Among other changes, lesions of the koilin layer and gizzard mucosa have been found in newly-hatched chicks and embryos, indicating a possible congenital background to the multifactorial syndrome of gizzard erosion and ulceration in poultry. Seven hundred forty specific pathogen-free eggs were divided into two groups. One was incubated as control (incubation temperature - 37.8ºC; relative humidity - 50-55% during the first 18 days of incubation and 60-65% during the last 3 days of incubation), and the other was challenged with a constant high incubation temperature of 38.9ºC (50-55% relative humidity). Eggs were removed from the incubator and sequential break-outs were performed to evaluate the effects of different temperature manipulations on embryonic development by histopathology. Cellular hypertrophy with mildly basophilic, PAS-positive amorphous content and cell rupture were found in nearly all challenged embryos (>85%) analyzed between days 12 and 16, in most cases affecting between 15% and 50% of the extension of the mucosal layer, which appeared vacuolated at low magnification. By day 18, areas of formation of irregularly-shaped spaces in the mucosa were identified, with apparent detachment of more superficial layers of the mucosa with formation of vesicles. The glandular mucosa in these embryos appeared disorganized, irregular and less cellular when compared to the well-formed, column-like architecture found in control embryos. Among the suspected causes of gizzard erosion/ulceration in newly hatched chicks are breeder diet, breeder age and capillary fragility combined with a transient increase in blood pressure during hatching. The lesions found in this study indicate that, although temperature may not be the only factor involved, it may also play a fundamental role in this syndrome by acting as a facilitating or exacerbating stimulus for the development of gizzard lesions, with fragility and irregularity of the mucosal surface and subsequent changes in the secretion and deposition of the koilin layer. Because chicks start ingesting feed in the first twenty-four hours after hatching, the combination of a fragile koilin layer with the input of the low-pH digesta from the proventriculus, as well as the presence of feed particles, could be enough to increase susceptibility to gizzard erosion/ulceration and lead to a lower subsequent post-hatch performance.

Keywords: Chicken, Gizzard, Incubation, Temperature, Ulceration

Chick yolk mineral levels during their sojourn in the hatcher

Abstract ID: 245

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The length of time a chick spends between hatching and accessing feed, known as chick sojourn, impacts chick quality. To quantify physiological changes in chicks during their sojourn, data were collected and combined from four trials. In each trial Cobb 500 eggs were incubated and chick hatch time recorded. In total five hundred and thirty chicks were removed from the incubator and sampled at various time intervals post-hatch, resulting in 9 different possible sojourn lengths (0-5 hours sojourn (HS), 6-11HS, 12-17HS, 18-23HS, 24-29HS, 30-35HS, 36-41HS, 42-48+HS). Measurements collected included chick body weight, chick length, residual yolk weight, serum Ca and P, and femoral bone ash percentage (BA). The residual yolk was assayed for ten minerals. As the length of the sojourn increased chick weight, residual yolk weight and BA decreased, yolk free body weight remained constant and chick length, bone mineral weight and serum Ca increased. Serum P decreased until 24-29HS, and then increased to 36-41HS. Yolk concentrations of Ca, Fe, K, Mn, Na and Sr increased over time, whereas Cu, Mg, P and Zn tended to decrease. Generally, yolk mineral quantities decreased as HS increased, however Fe increased at 36HS, Na increased until 24HS, Mn increased from 12-41HS, and P increased from 36HS. Chicks grow in length prior to feeding, utilizing their residual yolk. During this growth, the non-mineral component of the femur grows faster than the mineral component. This could be detrimental to skeletal integrity. Homeostatic mechanisms such as parathyroid hormone secretion may be responsible for the increase in serum P after it reached its lowest value at 24HS, which is likely to have initiated resorption of minerals from bones. Yolk mineral data is open to interpretation. One possibility is that selective uptake of minerals, such as P, through the yolk sac membrane is occurring by exchange with Na, until 24HS. After 24HS yolk transport could be nonselective and through the yolk stalk only. Minerals may passively move from blood circulation to the yolk if their concentration is higher in the serum. The complex physiology of chicks during their sojourn warrants further investigation. Mineral interactions may reveal vital information about chick nutrient requirements during this critical period.

Keywords: Hatch window, Minerals, Residual yolk

Effects of in ovo injection of N-Carbamylglutamate (NCG) on hatchability, quality of chickling, growth performance and carcass composition in broiler chickens

Abstract ID: 255

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N-carbamylglutamate (NCG) has been shown to increase litter size in sows and enhance performance in neonatal piglets. The modulation effects of NCG on embryonic and neonatal animal may convey benefit to developmental broiler embryos. Two incubation trials were conducted to investigate the effects of *in ovo* injection of NCG on hatchablility, chick quality, and posthatch growth and carcass composition in broilers. In the first trial, a total of 504 Ross 708 broiler hatching eggs were hand injected at 17 day of incubation (DOI) with 100 µL sterile saline (0.85%) alone or containing 1 or 2 mg NCG. In the second trial, a total of 1800 Ross 308 broiler hatching eggs were injected at 17 DOI with 100 µL sterile saline (0.85%) alone or containing 1, 2, 4 or 6 mg NCG. In trial 1, NCG injection did not affect hatchability, embryo (chick) BW as percentage of set egg weight or yolk sac weight as percentage of embryo (chick) weight on 19 and 21 DOI. After 48 h holding time without access to feed and water, BW loss or BW loss as percentage of set egg weight markedly decreased in chicks hatched from NCG injection treatments in comparison with those in saline or non-injected treatments. In trial 2, *in ovo* injection of NCG did not affect hatchability, BW as percentage of set egg weight. Chicks' BW at 21 day of age, and ADG during 0~21 day were significantly higher in NCG injected groups compared with saline or non-injected groups. Feed efficiency was improved in 6 mg NCG/egg groups than other groups during starter phase. Breast muscle percentage in 1 and 2 mg NCG/egg groups at 42 d of age significantly improved in comparison with those in saline or non-injected groups. Overall, in ovo supplementation of NCG may serve as an effective approach to improve chickling quality, early growth and breast muscle development in broiler chicks.

Keywords: Carcass composition, Growth performance, In ovo injection, N-carbamylglutamate (NCG)

Embryonic thermal manipulation of the Japanese quail: impacts on physiology and hypothalamic methylome and transcriptome

Abstract ID: 233

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Early environmental challenges during the embryogenesis are known to affect the phenotype, health and disease risk of animals. Numerous analyses have shown that this modulation of gene activity is associated to epigenetic alterations. The epigenome is therefore an important contributor to phenotypic plasticity, and learning how environmental exposures translate into persisting epigenetic changes may open new doors to improve the robustness and resilience of developing farm animals. In chicken, numerous studies reported the utilization of the thermal-manipulation (TM) of eggs during the incubation period to improve the thermotolerance of chickens at slaughter age, but the molecular mechanisms underlying this phenomenon are still unclear.
We took advantage of a highly inbred line of Japanese quails (*Coturnix japonica*) to perform a comprehensive study of the impact of TM on bird phenotype, methylome and transcriptome. Quail eggs were either incubated in control conditions (at 37.8°C and 56% relative humidity RH) or in TM conditions (from embryonic day 0 – E0 – to E13, 39.5°C during 12h/day and 65% RH and then 37.8°C and 56% RH). Short and long-term effects of the treatment on zootechnical, physiological and metabolic parameters were analyzed and heat tolerance was further tested by a heat challenge (HC) at 35 days of age (D35). Concomitantly, the short-term influence of TM was also investigated at hatch by methylome and transcriptome approaches. A significant influence of TM was observed on quail growth during the first week of life but no longer during later growth period. A modification of blood flow through the regulation of surface temperature was observed in female quails at D35. The analysis of blood at D35 revealed no huge difference between control and TM quails. In hypothalamus, several differentially expressed genes and regions of differential methylation were identified and are currently under study. The functionnal analyses revealed that the embryonic treatment affected several regulation pathways at both methylation and transcript levels.
This study was supported by the ANR JCJC "QuailHeatE" Research Program [ANR-15-CE02-0009-01].

Keywords: Embryogenesis, Physiology, Quail, Thermo-manipulation, Transcriptome

Influence of Incubation Temperature on the Development of the Bursa of Fabricius in the Chicken (*Gallus gallus*) Embryo

Abstract ID: 93

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The ideal incubation temperature for chicken eggs is defined as 37.5 to 37.8°C. Large deviations from this, in either direction, have been known to interfere with embryo development, hatchability and post-hatch survival and performance. A thousand ten specific pathogen-free eggs were divided into three different groups. One group was incubated as control (incubation temperature - 37.8ºC; relative humidity - 50-55% during the first 18 days of incubation and 60-65% during the last 3 days of incubation), and the remaining two groups were challenged with: A) constant high incubation temperature (38.9ºC) and B) constant low incubation temperature (36,7ºC). Eggs were removed from the incubator and sequential break-outs were performed in order to evaluate the effects of the manipulations on embryonic development by histopathology. Significant differences were visible between Treatment A embryos and controls from day 12 onward, and between Treatment B embryos and controls from day 14 onward. A greater number of lymphoid follicles was observed in the bursa of Fabricius of Treatment A embryos between days 12 and 14 of incubation when compared to control embryos of the same age. These follicles were also larger in size than controls. At day 16the bursal follicles of Treatment A embryos appeared slightly less cellular when compared to controls. This became more evident by day 18 when the cellular scaffolding for bursal follicles was found outlining these structures in similar numbers and dimensions to control-embryo bursas but containing very low numbers of (or even no evident) lymphoid cells. In embryos challenged with Treatment B, the bursa of Fabricius consistently had a lower number of follicles (which were also smaller in size) and a decreased number of lymphoid cells when compared to the controls. These findings suggest that incubation temperatures higher or lower than optimal have a detrimental effect on the development of the bursa of Fabricius that can lead to a lower subsequent post-hatch performance. Post-hatch immune status is vital to chick response to vaccines administered to day-old chicks. With the progressively more generalized use of in ovo vaccines at day 18 of incubation, the matter of embryonic immunity and delayed bursal development proves all the more critical. To the best of the authors' knowledge, no reports have been published on the effects of sub-optimal temperatures on the embryonic development of avian lymphoid organs.

Keywords: Bursa of Fabricius, Chicken, Immunity, Incubation temperature

Temperature variations during incubation and postnatal period affect performance, metabolism, health and gene expression in the blood of fast-growing chickens

Abstract ID: 420

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For adapting the developing chick embryo to thermal variations of later breeding environment, programs of temperature fluctuations during incubation are studied.
A multidisciplinary approach was applied to evaluate the effects on performance, pathophysiological markers and blood gene expressions of standard conditions (I0) or temperature variations (I1) during incubation combined with control rearing conditions (C) or cooler start followed by late heat exposure (V). Condition I1 including elevated temperatures during mid-incubation and cold stimulations during late embryogenesis did not affect hatchability. Chicken body weight was lower in I1 group than in I0, and in V than in C condition from d11 to slaughter age. Feed conversion ratio was lower for I1 chickens in C condition compared to both incubation groups in V condition. Chickens exposed to V condition were less affected by diarrhea than C chickens at 11d of age. Mortality was greater in I1 chicks than in I0 before 11d of age, whereas at 41d, overall mortality and morbidity rates were higher in C than in V condition. Meat quality was affected by both incubation and postnatal conditions: incubation I1 induced lower values of breast meat ultimate pH (pHu) measured 24h after slaughter, increased drip loss and shear force compared to I0. Postnatal V condition resulted in lower breast yield, greater abdominal fat and thigh percentages. It also reduced the occurrence of white striping and drip loss and increased pHu of breast meat compared to C, with a reduction in wooden breast only in I1 group. Both factors interacted to affect the intramuscular lipid content of breast meat that was lower in I1 than in I0 chickens only under C conditions. Postnatal but not incubation condition changed blood redox equilibrium compared to controls. Incubation I1 increased the blood expression of Acot11 involved in the thermogenic response to cold in mammals, while the expressions of HUS1 involved in mitotic proliferation and of SOST involved in bone quality were lower in V than in C condition. Thus the adaptive capacities and metabolism of broilers were strongly challenged by postnatal temperature fluctuations and this was not mitigated by thermal stimulations during incubation. However, varying postnatal environment limited growth, total mortality and meat quality defects compared to standard conditions at 42d.
Funding: Integrated Management of Animal Health metaprogram of INRA for the "GISA-ROBUSTCHICK" project (www.gisa.inra.fr/en)

Keywords: Blood transcriptome, Health, Incubation, Muscle integrity, Temperature variations

The Effect of Combined Monochromatic Photostimulation on Reproductive Activities of Broiler Breeders

Abstract ID: 384

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Artificial targeted lighting has a key role in stimulating poultry reproduction. Light perception mechanism in birds composed of two main components: retina and extra-retinal sites, which are located in the brain. Previous works have shown that photostimulation of the eye inhibits reproduction activities, whereas photostimulations of the brain will increase reproduction activity. Differential photostimulation of the retina and of the extra retinal sites was based on the retina's sensitivity to the green and yellow wavelengths on the one hand, and on the penetration abilities of the red wavelengths on the other hand. The purpose in this study was to examine the use of innovative lighting combinations on litter rearing broiler breeders. 150 broiler breeder females and 15 males (Ross) at 20 weeks. of age were divided into 5 light treatment rooms (n=30, n=3 respectively). Light treatments: 14 hr white (Control), long day (14 hours) red light (630 nm) and short day (6 hours) green light (560 nm) (red-green), long day green and short day red (green-red), long day red light and short day blue light (480 nm) (red-blue), long day blue light and short day red light (blue-red). The lighting treatment began from 20 weeks of age until 60 weeks. All rearing protocols conducted according to Ross recommendations. Daily egg production and egg weight recorded. Eggs were placed in incubator and fertility and hatchability were measure. Monthly blood samples collected for plasma steroids level. In the end of the experiment, tissues collected for mRNA gene expression. Egg production was significantly the highest in the red-blue group with an average of 84.7%, compare to Ross recommendation of 71.9%. and all other groups. Similarly, red-blue group laid 161 eggs/hen compare to Ross primary breeder recommendation of 140.9 egg/hen. The white group produced 136.3 egg/hen, blue-red 140.3 egg/hen, green-red 133 egg/hen and red-green 143.8 eggs/hen. No significant differences were detected in fertility and hatchability results of treated groups compare to control and Ross primary breeder recommendation. We conclude for the first time that targeted illumination can improve breeder's production in experimental model of broiler breeder flock.

Keywords: Broiler-breeder, Fertility, Hatchability, Photostimulation

The in ovo critical period for somatotrophic axis elevation by green light photostimulation

Abstract ID: 187

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In ovo green light (GL) photostimulation of meat type birds, elevated body weight and muscle growth at market age. The mechanism of this phenomenon was due to elevation of the somatotrophic axis activity. The objective of this study was to find the in ovo critical period for stimulating the somatotrophic axis by GL photostimulation. Two hundred equal weight fertile broiler eggs (Cobb 500), were divided to 4 in ovo treated groups: 1. Incubated at dark condition (Negative Control n=60), 2. GL photostimulated from embryonic day 0 (ED0) until ED20 (hatching day) (positive control n=60), 3. GL photostimulated between ED10 and ED20, (n=50) and 4. GL photostimulated between ED15 and ED20 (n=30). At ED10 and every other day until ED20, 10 eggs from each treatment groups were sampled. Eggs were opened, blood samples were drown for GH ELISA assay, hypothalamus, liver and breast muscle samples were collected for mRNA gene expression of GHRH, GHR (Growth Hormone Receptor) and IGF-1 (Insulin Like Growth Factor 1), by Real Time PCR. After finding no significant interaction between treatment and ED, all statistical analyses were conducted with the JMP software using one-way ANOVA. In-ovo GL photostimulation from ED0, caused a significant elevation ($P < 0.05$) in plasma GH levels (between ED14-ED20) of between 35% and 100%, compared to negative control. Hypothalamic GHRH mRNA gene expression significantly increased in 30% (on ED16 and ED20), and both liver GHR (on ED12 and ED16-18) and IGF-1 (on ED16-18) were significantly elevated by 20%-100%, compared to the negative control. In-ovo GL photostimulation from ED10 showed positive effect (compared to the negative control) on GH plasma levels, with no effect on mRNA gene expression. Green light photostimulation from ED15 showed elevation in somatotrophic axis activity similarly to the positive control group ($P < 0.05$). In ovo, green light photostimulation of broiler embryos, from ED15-ED20 significantly elevated somatotrophic axis activity similar to the positive control group. We suggest that the critical period for GL photostimulation acceleration of somatotrophic axis is between ED15-ED20.

Keywords: Broilers, Development, In ovo photostimulation, Somatotrophic axis

Ultrastructural development of the Yolk Sac Tissue during incubation

Abstract ID: 371

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The yolk sac tissue (YST) is an extra-embryonic tissue that envelopes the yolk during incubation. The YST serves as the primary site of digestion, absorption and nutrient transfer from the yolk content to the developing embryo. Contrary to mammals, in which maternal nutrients are transferred continuously to the embryo via the placenta, chicken maternal nutrients are deposited into the oocyte during vitellogenesis and constitute the primary nutritional source in the fertile egg. The YST develops during the first week of incubation and reaches maximum functionality by day 15 (E15). Towards hatch, the YST dimensions decrease while final intestinal development occurs, in preparation for the shift from embryonic to post-hatch nutrition. The aim of the current study was to characterize morphological changes in the YST during different stages of incubation. Observations of YST samples by Scanning Electron microscope (SEM) revealed villi structures, comprised of endodermal epithelial cells (EEC's) surrounding a central blood vessel. Along the incubation period, changes are evident in EEC morphology. At E15, EEC's are bloated, while at DOH, they appear shriveled. Measerments of EEC sizes showed a significant decrease from E15 to DOH, with average cell perimeters of 116.82 μm and 95.43 μm respectively. It was found that YST EEC's contain microvilli on their apical membrane. At E15, the majority of microvilli were located at EEC borders. An increase in their quantity was evident towards hatch (from E15 to DOH), with higher uniformity of microvillar distribution, covering the entire cell surface. Results of the current study suggest that the increasing numbers of microvilli in the YST towards hatch is related to an increase in nutrient demand by the embryo. It is expected that YST development and functionality will be affected by incubation conditions through changes of YST cell dimensions, including microvilli localization and quantity during the last days of incubation.

Keywords: Embryo, Scanning electron microscope, Yolk

ORAL PRESENTATIONS

Poultry Health

Adenoviral gizzard erosion in pullet and layer flocks and potential means of prophylaxis due to live vaccination with an apathogenic FAdV-A (CELO)

Abstract ID: 260

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The gizzard is an important organ for processing feed and various nutritive factors are known to influence the function. Adenoviral gizzard erosion (AGE), caused by infection with pathogenic Fowl Adenovirus species-A (FAdV-A), has been well documented from natural outbreaks in broiler flocks in concurrence with extensive economic losses due to growth retardation and subsequently reduced end weights. Sporadically, gross lesions in the gizzard together with the isolation/detection of FAdV have been described also from pullet or layer flocks. Recently, 15 cases of AGE have been confirmed from natural outbreaks as well as from retrospective analysis of gizzard samples from pullets and layers kept in free range and enriched cage systems. Available production data from three outbreaks showed an increased mortality of up to 1.07% over a period of two to three weeks. Necropsy of affected birds revealed the typical pathomorphological lesions characterized by areas of erosion in the koilin layer, non-suppurative inflammation and degeneration of glandular epithelial cells in the gizzard mucosa as well as the presence of basophilic intranuclear inclusion bodies. In all outbreaks FAdV-A could be isolated by cell culture and/or detected by PCR from affected gizzards. Sporadically, FAdV-C and -E could be also detected from gizzards of two pullet flocks. However, pathological changes, in particular intranuclear inclusion bodies, were directly attributed to FAdV-A by a species specific *in-situ* hybridization. The pathogenicity of FAdV-A as well as the aetiological role of FAdV-E, both isolated from AGE affected pullets, was investigated in 3- and in 20-week-old specific-pathogen-free (SPF) layers. Furthermore, the protective effect of a single (17^{th} week of life) and double (14 and 17^{th} week of life) live vaccination with the apathogenic FAdV-A CELO strain against challenge with the pathogenic FAdV-A was investigated in 20 week-old SPF layers. Post-mortem examinations were performed at 7, 10 and 14 days post infection (DPI) with a special focus on pathological changes in the gizzard. Characteristic pathomorphological changes in gizzards of pullets and layers from 7 DPI onwards confirmed pathogenicity and aetiological role of the isolated FAdV-A in the development of AGE. The investigations confirmed that FAdV-E was just isolated in co-incidence as no pathological changes were observed in 3- and 20-week-old SPF birds. Furthermore, for the first time it could also be demonstrated that live vaccination with the apathogenic CELO strain prevents the development of AGE in layers due to a later experimental challenge with a pathogenic FAdV-A strain.

Keywords: Adenoviral gizzard erosion, Fowl adenovirus, Layers, Pullets, Vaccination

Amino acid substitutions in the Receptor Binding Domain (RBD) of Avian Influenza Viruses subtype H9N2 in Israeli isolates, with human health implications

Abstract ID: 353

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Influenza A viruses of the H9N2 subtype are considered one of the most likely candidates to cause a new influenza pandemia in humans. Currently, the subtype is endemic in many countries including Israel, and causes considerable economic losses in the poultry industry. Human cases of H9N2 virus infection have been reported since the late 1990s. The Receptor Binding Domain (RBD) that is part of the Haemagglutinin (HA) gene is responsible for binding the virus to residues of sialic acid in the oligosaccharide receptors on the host cell surface. Avian viruses and human viruses have different specificities in their ability to infect avian or human host cells. The type of the amino acids in the RBD is considered responsible to the degree of affinity of the virus to host cells. The H9N2 virus is documented in Israel since yr. 2000. In this study we examined the amino acid substitutions, those that may have human health implications, in the RBD of 201 Israeli isolates during yrs. 2000-2014. One of the most important findings reveals that 94.9% of the isolates during yrs. 2006-2013 and 100% of the 2014 isolates have amino acid leucine at position 226 of the HA gene. Leucine at this residue affects cell tropism and has implications on the capability of the virus to affect humans, due to better specificity to human's cells. The results of this survey indicate high genetic variability among the H9N2 viruses associated with specific amino acid substitutions. This and other variations highlight the need of continuous monitoring alterations in H9N2 viruses, to get a better understanding of the risk to human health posed by these viruses.

Keywords: Avian Influenza H9N2, Haemagglutinin gene, Receptor Binding Domain, Sialic acid

An innovative approach for analysing and evaluating poultry farms odour related to animal health and welfare

Abstract ID: 357

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Volatile organic compounds (VOCs) produced by pathogens, host-pathogen interactions and biochemical pathways were explored in several study as biomarkers for their capacity of diagnosing pathologies in livestock and in humans. VOCs are present everywhere such as in blood, breath, faeces, sweat, skin, urine and vaginal fluids and their qualitative and quantitative composition is influenced by pathophysiological responses to infections, toxins or endogenous metabolic pathway perturbations. In poultry, VOCs analysis has been explored to evaluate air quality in sheds but they have never been monitored to determine if birds were affected by enteric pathologies. These enteric disorders represent one of the most important groups of diseases they affect poultry and cause illness, mortality and economic losses. For this reason, monitoring the health status of broilers and an early detection of any health problem is of great importance in intensive farming, especially nowadays that antibiotics are banned. Nowadays, the preventive use of antibiotics in intensive farming system is common and this management practice involves spreading of drugs in the environment, contributing to the phenomenon of antibiotic resistance. The prompt reaction to any change in health, welfare and productive status is the key for the reduction in drugs usage and for the improvement of animal wellbeing. Due to the high priority of this issue, it is of great importance the early detection of any health problem in intensive farming. Precision Livestock Farming, through the combination of cheap technologies and specific algorithms, can provide valuable information for farmers starting from the huge amount of data collected in real time at farm level.This study was aimed to the application of a PLF diagnostic tool, sensible to the variation of volatile organic compounds, to promptly recognise enteric problems in intensive farming, supporting veterinarians and enabling specific treatments in case of disease.

Keywords: Early warning system, Health, PLF, Poultry welfare, Volatile organic compounds

A probiotic containing viable spores of Bacillus licheniformis improves gut health after coccidiosis vaccination

Abstract ID: 212

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The probiotic (B-Act®) is a feed additive, consisting of viable spores of a unique *Bacillus licheniformis* strain (BL). The current approved coccidiosis vaccines contain live parasites which will multiply in the gut to evoke immunity. Although attenuated oocysts are being used minimal damage will be present in the gut. Objective of the 40 day study was to determine, if the probiotic can reduce gut lesions after coccidiosis vaccination in broilers. A randomized block design with 20 replications of 5 birds per pen was used. On day 1 all birds were vaccinated with an attenuated coccidiosis vaccine (Huveguard MMAT) containing oocysts of *Eimeria maxima*, *Eimeria mitis*, *Eimeria acervulina* and *Eimeria tenella*. The treatment groups were: 1. no additives (control group); 2. BL 0.5 kg ($1.6x10^9$ cfu *Bacillus licheniformis*/kg of feed), fed continuously. Bird weights and feed consumption were measured on day 40. Coccidiosis lesion scores on 10 birds were evaluated on day 15, 22 and 29 of age together with dysbacteriosis score and evaluation of foot pad lesions. Final weight between treatment groups was similar however FCR was markedly improved in the probiotic versus the control group. In addition vaccinating the birds with a coccidiosis vaccine combined with feeding BL improved lesions, dysbacteriosis and foot pad lesion score. BL is not impairing recycling of the vaccine as seen in lesions on day 15, but reduces the negative effect of the field strains starting around day 21. Feeding of this unique strain of *Bacillus licheniformis* in coccidiosis vaccinated birds improves feed conversion and reduces gut and foot pad lesions.

Keywords: Bacillus licheniformis, Coccidiosis vaccination, Probiotic

A probiotic containing viable spores of Bacillus licheniformis prevents outbreak of necrotic enteritis

Abstract ID: 210

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The probiotic is a feed additive, consisting of viable spores of a unique *Bacillus licheniformis* strain (BL). Necrotic enteritis continues to be an important disease of broiler chickens. The depression of growth rate and feed efficiency of birds is unavoidable due to intestinal damage and the subsequent reduction in digestion and absorption of feed. Objective of the 28 day study was to determine, if the probiotic can reduce the effects of *Clostridium perfringens* induced necrotic enteritis in broilers. A randomized block design with 6 replications of 8 birds per pen was used. The treatment groups were: 1. non medicated, non-infected (NMNI); 2. non-medicated, infected (NMI); 3. BL ($1.6x10^9$ cfu *Bacillus licheniformis*/kg of feed), fed continuously. On day 13 all birds were challenged orally with coccidiosis (*Eimeria maxima*, approximately 5000 oocysts) and on day 18, 19 and 20 all birds, except NMNI were challenged with *Clostridium perfringens* (2 $ccx10^8$ cfu/bird). Bird weights and feed consumption were measured on day 28. Significance was set at $P<0.05$. This study reproduced clinical necrotic enteritis (NE; 18.8% NE mortality for NMI). The feed conversion was significantly improved for BL compared to NMI (1.680 vs 2.079). Also the total weight gain was significantly improved for BL compared to NMI (874g vs 643g). The NMNI had a better feed conversion (1.635) and total weight gain (905) compared to all other groups. Feeding this unique *Bacillus licheniformis* strain demonstrated significant improvements in performance as well as reducing mortality in *Clostridium perfringens* induced necrotic enteritis in broilers.

Keywords: Bacillus licheniformis, Necrotic enteritis, Probiotic

A probiotic containing viable spores of Bacillus licheniformis reduces caecal colonization of Campylobacter

Abstract ID: 211

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The probiotic feed additive consists of viable spores of *Bacillus licheniformis*. *Campylobacter* is one of the most common causes of food poisoning. The incidence and prevalence of campylobacteriosis have increased over the last 10 years. Poultry is a major reservoir and source of transmission of *Campylobacter* to humans. Objective of the 37 day study was to determine, if the probiotic (BL) can reduce *Campylobacter* colonization in broilers. At the start of the study, twenty one-day-old broilers were randomly assigned to one of the four treatments: 1. no additives (control group); 2. BL 0.5 kg ($1.6x10^9$ cfu *Bacillus licheniformis*/kg of feed), fed continuously; 3. BL 1 kg ($3.2x10^9$ cfu *Bacillus licheniformis*/kg of feed), fed continuously; 4. BL 2 kg ($6.4x10^9$ cfu *Bacillus licheniformis*/kg of feed), fed continuously. At day 21, twenty *Campylobacter* spp. positive birds were added to each group. The concentration of *Campylobacter* spp. (log10 cfu/g caecal content) was measured at day 37 by inoculating decimal dilutions on *Campylobacter* spp. specific plates. Significance was set at $P<0.05$. This study demonstrates that the probiotic fed at 2 kg/mton of feed significantly reduced *Campylobacter* concentration in the caecum of broilers compared to the control group (4.75 log10 cfu/g versus 6.55 log10 cfu/g). BL at a level of 0.5 kg and 1 kg/mton of feed showed a tendency ($P=0.085$ and $P=0.0562$) to decrease the amount of *Campylobacter* in the caeca of the birds (6.01 log10 cfu/g versus 5.59 log10 cfu/g respectively). Under the present study conditions feeding of BL significantly reduced *Campylobacter* spp. colonisation of the caeca. A significant dose-response effect was observed.

Keywords: Bacillus licheniformis, Campylobacter, Probiotic

A retrospective analysis to identify the contribution of the breeder on health and performance of broiler chickens

Abstract ID: 78

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Data on performance and health are routinely collected in the various stages of the broiler production chain. In The Netherlands, routinely collected data are usually stored and solely used by the owner(s), and the various databases are not connected. Connecting databases may however provide insight to improve the quality of the chain. We were interested in the contribution of the parent stock to the performance of the broiler flock, as transgenerational effects have been reported and variation in performance and health between breeder farms and flocks exist. Broiler flock data at depopulation, collected by the slaughter plant (growth, uniformity, rejections, first week and total mortality) and from a national database (antibiotic treatments) were linked to breeder flocks and farms by unique identifiers for the period between 2011-2016. This resulted in 2174 broiler flock records (at house level). Within these records, 74 broiler farms, 88 breeder farms, and 209 breeder flocks were identified. A mixed model analysis was used to simultaneously estimate effects of season, parent flock age, time, and the variance components that determine the contribution of the chain phase to the broiler performance parameters. Results showed no systematic effects of the breeder farm on the various parameters at broiler level. Systematic effects of breeder flock were relatively small; the largest effect was found on rejections in the broiler flock (estimated contribution to the variance component: 7%). The largest contributions to the variance component were found for broiler farm: 14% (antibiotic treatment) to 59% (growth index). Also the phase between egg laying at the breeder farm and chick placement at the broiler farm (here called: chick delivery) had a large contribution to the variance component: rejections: 27%, first week mortality: 52%). Negligible effects were found for house at broiler farm and specific breeder-broiler farm combinations. A moderate effect of broiler house within a chick delivery was found. It can be concluded that systematic effects of breeder flock and farm on broiler performance and health could not be found, and that broiler farm and chick delivery had a large contribution on the variation in broiler performance and health. This does not exclude that transgenerational effects exist, but these may be relatively short lasting, and thus could not be found in the present analysis, or might be overruled by more influential factors after egg laying at the breeder farm.

Keywords: Broiler, Data analysis, Health, Performance

Assessment of innate immune response by applying mono or polyvalent live Newcastle disease and Infectious bronchitis vaccinations

Abstract ID: 85

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Newcastle disease (ND) and Avian infectious bronchitis (IB) remains a significant concern to the poultry industry; however, it can be managed with effective vaccination programs. Vaccinating day-old chicks with ND and IB vaccines plays an important role in flock immunizations, and the immune response considers as one of interesting discussion in avian immunology. This could be done whether with separate or mixed two types of vaccines. Several studies were evaluated the humoral immunity responses with different vaccination programs, but few do so for innate immune responses, especially mediators of inflammation. 20 day-old SPF chicks were divided into four groups and vaccinated with IB, ND, ND/IB, and ND + IB vaccines via the eye drop route. Tracheal samples were taken 48 hours post vaccination, and RNA was extracted. The innate immune response was assessed by quantifying the IFN-γ, IL6, IL8, IL10, IL12, IL15, and IL18 mRNA by Quantitative Real-time RT-PCR. Surprising differences were found between the groups. The highest expression of genes (Fold-Change) that involved in mucosal and cell-mediated immune response for the preliminary immunity of chicks was recorded with group vaccinated with ND/IB. Our findings highlight the mucosal and cellular immune response changes against two common respiratory pathogens in different vaccination strategy. We concluded that with commercial combined vaccines ND/IB vaccine, which is a factory-mixed vaccine of two viruses, could induce stronger local and antiviral response than vaccinating with ND and IB separately causing higher immunity level.

Keywords: Avian Infectious Bronchitis, Immunity, Newcastle disease, Vaccination

Assessment of the efficacy of fecal sampling in representing gastrointestinal microbiota in chickens

Abstract ID: 624

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Because of the convenience and noninvasiveness of fecal sampling, most studies use fecal samples as a proxy to study the gut microbiota, despite the increasing recognition that fecal microbial populations may not be fully representative of those in the gastrointestinal (GI) tract contents or mucosa. Therefore, a comprehensive understanding of the efficacy of using fecal samples as a proxy to study the GI microbiota would help improve longitudinal analyses of microbiota. Using chickens as a model, we collected 1 026 samples from 206 animals, including duodenum, jejunum, ileum, cecum and feces samples. Most taxa in the small intestine (94.10 – 94.82%) and ceca (99.57%) could be identified in feces. Microbial community membership was reflected with a gut anatomic feature, but community structure was not. Excluding shared microbes, the small intestine and ceca contributed 26.69 and 2.36% of the total fecal members, respectively. The composition of Firmicutes members in the small intestine and that of Actinobacteria, Bacteroidetes and Proteobacteria members in ceca could mirrored that observed in fecal samples well (ρ = 0.68 – 0.79 and 0.66 – 0.79, respectively, $P < 0.05$). All sites clustered into 2 or 3 enterotype-like clusters, which is the first reports of enterotype-like clusters in the duodena, jejuna, ilea and ceca in chickens. Feces from different clusters were observed to reflect the GI microbiota with different efficacy. Our results provide evidences that the good potential of feces to identify most taxa in chicken guts, but microbial structure analyses using feces as a proxy for gut should be interpreted with caution, which will help extend the understanding in chicken gut microbiota and provide suggestions in usage of fecal samples for gut microbial study.

Keywords: Enterotype-like clustering, Feces, Gastrointestinal tract, Spatial relationships

Autogenous vaccine against Escherichia coli and Gallibacterium anatis reduces losses and improves production on layer farms

Abstract ID: 627

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Gallibacterium anatis and avian pathogenic *E.coli* (APEC) is one of the most frequent pathogen combination found in layer flocks today, causing serious problems with increased mortality and drop in egg production combined with high multidrug resistance. If other pathogens are involved, like MS, MG, IB, ART etc, loses could be significantly higher. The purpose of this study was to determine influence of bivalent autogenous vaccines on flock performance, made on farmers request from local *G.anatis* and APEC strains. Farms were visited or carcass/swab samples were sent for isolation of APEC and *G.anatis* strains, which were confirmed by MALDI-TOF spectrometry and selected for vaccine production. Antigen solution is dispersed in oil adjuvant and given in 0,4 ml with 10^8 CFU per dose twice per pullet at 9 and 18 weeks of age. Flocks were also monitored for other diseases by serology, together with production parameters and weekly mortality and compared to previous flocks. Farms included in this study showed high incidence of colibacillosis with high percentage of fibrinous poliserositis and high percentage of APEC strains isolated from bone marrow and liver, but also *G. anatis* strains were isolated from liver, oviduct or egg follicles. Both species were also multidrug resistant. Nonvaccinated flocks reached peak, but production was later irregular with elevated mortality, on average around 0,7% per week, sometimes reaching around 2,6% per week. Those flocks were MS, MG and ART negative, and IB vaccination program was adjusted to cover broad spectrum of strains. Vaccinated flocks reached peak without later fluctuations, but mortality was 0,078% per week on average, with max 0,10% per week. Results showed significant influence of autogenous vaccine on flock performance, confirming that combination of APEC and *G. anatis strains* were the main causative agents of problems in previous flocks. Also, due to high multidrug resistance of isolated strains and fact that egg production is not compatible with drug application, autogenous vaccines seems to be method of choice for successful egg production.

Keywords: APEC, Autogenous vaccines, Egg production, Gallibacterium anatis, Layers

Avian botulism: recent investigations from diagnosis to management (in France)

Abstract ID: 328

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Avian botulism is a re-emerging problem in Europe for the last decade. A raise in poultry botulism outbreaks was recorded in 2007 and 2008 which triggered the creation of a French National Reference Laboratory for avian botulism in 2011. Various projects have been carried out since then to address and improve the knowledge about avian botulism and to help the national authorities to set up a management system. First, a diagnosis method has been developed through the analysis of 63 suspicions and showed that analysis of 4 livers using Real-Time PCR allowed the confirmation of 97 % of avian botulism outbreaks while analysis of serum using mouse lethality assay (considered as Gold standard up to now) only allowed the confirmation of 77 % of the outbreaks. In a second step, the limit of detection was defined and tended to be 5 spores per gram of type C and D *Clostridium botulinum* spores and 250 spores per gram of type E in avian liver. Specificity of the method was shown to be 100 % and sensitivity 95.35%. The methodological optimizations regarding the detection of *C. botulinum* in various matrices have resulted in the improvement of outbreak confirmation but have also allowed the identification of critical contaminated areas in poultry farm after an outbreak and the demonstration of spore persistence within and around the poultry house, even several months after the outbreak. This has also resulted in the delivery of a performant tool for the validation of cleaning and disinfection operations. Analysis of poultry manure samples after a botulism outbreak has shown to be highly contaminated even after several weeks of storage and needs relevant and careful management to avoid recurrence between flocks and cross contamination between farms and livestock productions. Although major achievements have been realized since the creation of the French NRL for avian botulism, many perspectives are still to be implemented in order to prevent new botulism outbreaks.

Keywords: Avian botulism, Detection, Diagnosis, Manure

Bacillus subtilis 29784 prevents a pro-inflammatory response in an induced inflammation condition using the Caco-2 cells model

Abstract ID: 419

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The protection of the animals against exogenous challenges, including infectious diseases and digestive disorders, is mainly provided by the intestine. Maintaining intestinal barrier integrity and functions is therefore critical to avoid deterioration of animal health status. Among the different ways to achieve this, probiotic supplementation could turn out to be a good solution.
In a previous study, we have shown in a Caco-2 cells model that *Bacillus subtilis* 29784 is able to prevent the disruption of the intestinal barrier by improving TransEpithelial Electrical Resistance (TEER) and inflammatory status. The objective of the present study was to explore the mechanisms involved into the anti-inflammatory properties of *B. subtilis* 29784.
Caco-2 cells model was used to evaluate the impact of the *B. subtilis* strain on the NFkB signaling pathway, known to be a major player in inflammation. Vegetative cells of *B. subtilis* 29784 were applied for 16 hours to a 14 day-differentiated Caco-2 cells monolayer established in a Transwell system. Caco-2 cells exposed or not to bacterial cells were then stimulated basolaterally with IL-1β, and nuclear and cytosolic fractions were extracted. Western blotting was finally carried out to measure IkB protein level in the cytosol and nuclear translocation of NFkB.
IL-1β treatment on Caco-2 cells induced a fivefold increase in NFkB amount in the nuclear fraction, demonstrating activation of the signal pathway by the pro-inflammatory stimulus. When cells were pre-incubated with *B. subtilis* 29784, there was no increase in nuclear level of NFkB. This indicates that the bacterial strain is able to prevent the translocation of NFkB from the cytoplasm to the nucleus. In order to activate the NFkB signaling pathway, IkB must be first degraded. As expected, cytosolic level of IkB decreased in the IL-1β treated cells. When the Caco2 cells were first exposed to *B. subtilis* 29784, IkB degradation was very much reduced, explaining the decrease in NFkB translocation.
The results obtained from this study suggest that *B. subtilis* 29784 exerts its immunomodulatory properties by inhibiting IkB degradation, thus preventing NFkB translocation and, by doing this, the expression of pro-inflammatory cytokines, such as IL8.

Keywords: Bacillus, Caco-2, Inflammation, NFkB

Bio-monitoring of mycotoxins in blood serum and feed to assess exposure of broiler chickens in Belgium

Abstract ID: 480

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Mycotoxin exposure in animals is mainly monitored by feed analysis, since maximum guidance levels are only available for feed. However, uneven distribution and non-proportional spread of mycotoxins in feed hampering representative sample collection and evaluation of mycotoxin exposure in animals. Therefore the aim of this study was to comparatively evaluate mycotoxin exposure in broiler chickens by mycotoxin feed analysis and by analysis of mycotoxins and their *in vivo* phase I and II metabolites in serum, as candidate biomarkers for exposure. A cross-sectional study was performed on 40 different randomly selected broiler rearing farms in Belgium. During a farm visit at the animal's age of 28 days, a pooled feed sample was collected. Feed samples were analysed by a validated multi-mycotoxin LC-MS/MS method. Furthermore, also serum samples were collected from 10 randomly selected chickens per farm. Serum concentrations of mycotoxins and major *in vivo* phase I metabolites were analysed quantitatively, while the presence of phase II metabolites was determined in a qualitative approach by an UPLC-HRMS method. Deoxynivalenol (DON) was the most prevalent, contaminating 74% of the feed samples, with an average concentration in positive samples of 270±171 µg/kg and a maximum of 750 µg/kg. Besides, also the acetylated forms 3- and 15-acetylDON (3- and 15-ADON) were present in half of the samples, however, in lower concentrations (8±3 µg 3-ADON/kg and 10±7 µg 15-ADON/kg). Remarkably, only in 18% of the farms DON was detected in serum samples (11±19 ng/mL). A maximum serum concentration of 49 ng DON/mL was detected in chickens fed a diet contaminated with 191 µg DON/kg. Besides, 15-ADON was not detected in serum and 3-ADON was only detected in 10% of the farms. Similarly, deepoxy-DON (DOM-1) was only detected in 10% of the farms. Although, sulfate conjugates are the most important phase II metabolites, these metabolites were only detected in a few serum samples. A similar tendency was observed for other mycotoxins. In conclusion, bio-monitoring of mycotoxins and related phase I and II biomarkers in blood serum gives an underestimation of the actual mycotoxin exposure in broiler chickens, most probably due to the limit oral absorption and fast and efficient elimination of many mycotoxins in this animal species.

Keywords: Biomarker, Broiler chicken, Deoxynivalenol, Mycotoxins, Serum

Beyond effects on the microbiota, Bacillus Subtilis 29784 shows direct effects on the host

Abstract ID: 429

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Administered in adequate amounts, probiotics could improve gut health in poultry. The present work investigates the effect of *Bacillus subtilis* 29784 on gut health, at host and microbiota level. An *in vitro* study was conducted to assess the direct effect of *Bacillus subtilis* 29784 on intestinal barrier function and inflammation using a Caco-2 cell model. In an in vivo study, 1.600 day-old Cobb 500 male broilers were fed during 42 days either a Control diet or a Control diet supplemented with *B. subtilis* 29784 (1 x 10^8 CFU/kg). At 42 d, intestinal tissues and contents were collected for histomorphological analysis and 16S rDNA gene sequencing. *In vitro* data showed that *B. subtilis* 29784 can increase Trans Epithelial Resistance and IL8 production, suggesting that this probiotic can improve intestinal functions. In the growth performance study, *B. subtilis* 29784 increased final BWG of broilers (+5.4%; $p < 0.0001$) and improved feed efficiency (-5.8%; $p < 0.0001$). Intestinal morphology was also modified with an increase of intestinal microvilli height (+18% in ileum and +17% in cecum; $p < 0.001$). In addition, *Ruminoccocus* and *Lachnoclostridium* genera were found in higher relative abundance in *Bacillus*-treated birds. Both bacteria are known to be involved in the degradation of polysaccharides and production of butyrate. In conclusion, *B. subtilis* 29784 can increase broiler performance, presumably through effects on intestinal microbiota and host physiology, as shown *in vivo* by an improvement of intestinal absorption capacity and *in vitro* by intestinal barrier integrity and inflammation status.

Keywords: Bacillus, Gut integrity, Inflammation, Microbiota

Biomarkers as a tool to better understand the effect of mycotoxins on gut barrier functioning and immunosuppression

Abstract ID: 34

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Intestinal gut health is key for overall performance and health and can be impaired by different factors. One of these factors is the presence of mycotoxins in the feed. Mycotoxins are immunosuppressive leading to higher susceptibility to secondary infections and production losses. The indirect effect of mycotoxicosis on performance losses finds its origin into impaired gut barrier functioning and decreased gut health (bacteria dislocation) with an altered immune response as a consequence. To gain information on the effect of mycotoxins on the animals immune reaction, biomarkers were used as a tool in this in vivo experiment. Biomarkers give the opportunity to detect changes in biological parameters in a very early stage after contamination, including immune responses. Ninety six one-day-old broiler chickens were used to evaluate the effect of feeding naturally contaminated rations with low levels (17 ppm) of fumonisins and the protective effect of a commercial mycotoxin eliminator on circulating and intestinal immune cells, blood biochemistry and haematological variables. Contamination with fumonisins lead to a significant increased sphinganine to sphingosine ratio, decreased hematocrit value, decreased total leukocyte count and an increased ratio of albumin to globulin in the blood biochemistry. All these signs are correlated with a decreased general health status. The immune response to fumonisins was clearly demonstrated by a significant effect on the amount of circulating helper T-lymphocytes, regulatory T-lymphocytes and terminally activated cytotoxic T-lymphocytes. The number of circulating monocytes and macrophages was also significantly decreased by contaminating the broilers, whereas the use of an effective mycotoxin eliminator resulted in higher levels of available immune cells again and hence give immune support to the animal to overcome not only mycotoxicosis but also secondary infections. In conclusion, there is still much to reveal on the effect of mycotoxins in the gastro intestinal tract. However, in this research, it is clearly demonstrated that mycotoxins, fumonisins in this particular case, affects immune responses orchestrated by impaired gut barrier functioning and metabolic changes. Biomarkers, such as circulating lymphocytes, are a great tool to better understand how mycotoxins cause production losses by immunosuppression.

Keywords: Blood biochemistry, Broilers, Circulating lymphocytes, Mycotoxins

Coccidiosis Control With Farm Management to Prepare for "No Antibiotics Ever" Production

Abstract ID: 298

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Pressure from retailers to produce poultry completely without antibiotics, including ionophores ("No Antibiotics Ever") or without antibiotics used in human medicine (allowing ionophores) has forced many U.S. broiler integrators to adapt part or all of their weekly production to the requirements of these labels. Subclinical coccidiosis, particularly *E. maxima*, remains an important underlying factor in dysbacteriosis, necrotic enteritis and poor performance, so improving coccidiosis control (even without ionophores) has been essential to achieving good performance in the face of these market demands. Performance and coccidiosis lesion scores were monitored at two large integrator complexes after changes in one key management factor (down time between flocks) was altered. Coccidiosis control programs during the monitoring period included ionophores, "hybrid" vaccine-ionophore or vaccine only programs. Coccidiosis lesion scores were monitored by regular post-mortem sessions involving 60+ birds representing 12 flocks of incremental age from <17 days to slaughter. Despite the small sample size, the resulting average overall *E. maxima* lesion score (and to a lesser degree, *E. acervulina* lesion score) correlated to feed conversion ratio. In each case, the overall lesion score was less than a microscopic *E. maxima* lesion score of +1 or a gross E. acervulina score of + 0.25. Each complex produces 1.8 kg broilers with a low energy feed model. Complex A started with *E. acervulina* at an average + 0.24 and *E. maxima* +0.62 with FCR approximately 1.80. After management adjustments, the average *E. acervulina* was + 0.14 and *E. maxima* was +0.39 with FCR approximately 1.65. Complex B started with similar coccidiosis lesion scores: *E. acervulina* + 0.20 and *E. maxima* +0.66 and FCR approximately 1.80. After management adjustments, the average *E. acervulina* was +0.07 and the *E. maxima* was +0.36 and FCR was again approximately 1.65. The key change was an increase in down time from an average of 10 days to 16 days. Complex A calculated the cost of 7 days' increased down time for the 1.5 million bird/week complex at US$ 2.8 million annually, while the return for 10 points (100 gm) FCR was US$ 4.3 million annually. The extra down time paid for itself in performance, while reducing the overall coccidiosis challenge, making it easier to implement the reduction or elimination of antibiotics. A simple management change can aid in the elimination of antibiotics without severe performance consequences.

Keywords: Coccidiosis, Down time, FCR

Comparative study on primers for molecular detection of different infectious bronchitis virus genotypes

Abstract ID: 84

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Infectious bronchitis (IB) is always involved in avian respiratory complex and is included in almost all cases. To detect this virus, several diagnostics such as serology and molecular assays are used. Molecular RT-PCR based tests are the best approaches to trace and detect the genome of the virus and are extensively used around the world. Different primers are available based on loci to detect IB virus. The candidate primers were aimed at 5'UTR, nucleoprotein, matrix, polymerase and 3'UTR regions. Four IBV genotypes (793/B, variant 2, QX, and Massachusetts) were evaluated in this study. Different dilutions of each genotype, from 1 to 100 ng cDNA, were prepared and evaluated with candidate primers in similar conditions and three replicates. PCR products were run in similar conditions and imaged with Gel Doc to be analyzed using Image J Software. The pixels were recorded, and finally, the differences between groups were analyzed with ANOVA. The results indicated nucleoprotein and 3'UTR primers as the most sensitive ones being able to detect all genotypes in all dilutions Especially in low concentrations and were so sensitive. This study showed that the primers were not suitable to detect all genotypes in low concentrations, though designed for. It is recommended to use two gene targets for multiplex PCR, e.g. 3'UTR, N gene for the detection of IBV. Since avian infectious bronchitis is a gamma coronavirus, this protocol could also be used for the detection of gamma coronaviruses in other birds as the suitable primers.

Keywords: IBV, Molecular detetcion, RT-PCR

Comparision of Commercial Broilers flocks vaccinated with a novel DNA construct vaccine applied in hatchery and reared in different field conditions

Abstract ID: 49

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Herpesvirus of Turkey (HVT) is a naturally-occurring virus from turkeys, also known as Marek's disease virus serotype 3. This virus has been used for decades as a vaccine to aid in the protection against Marek's disease in chickens. Because it is a naturally-occurring non-pathogenic virus (not treated or attenuated), it cannot revert to virulence when used as a vaccine. Like other herpesviruses, HVT does produce a persistent infection, inducing life-long immunity. HVT has a 159, 160 base-pair genome encoding for an estimated 99 different proteins. The genome includes unique long (UL) and unique short (US) genomic regions and short repeat (RS) and long repeat (RL) segments. HVT was selected as the appropriate backbone for genetic construct vaccines because of the characteristics of non-pathogenicity, persistence of infection, and stability when genetic material is inserted into its genome to encode for proteins that are not naturally expressed by the virus. Unique and proprietary genes enable researchers to construct an HVT virus that encodes not only for proteins that contribute to Marek's disease protection, but also to protection against Newcastle disease (ND) or Infectious bursal disease (IBD). A novel DNA construct vaccine (Innovax ND-IBD, MSD Animal Health) was administered on two different farms with differing management levels, different building types (single and multi-age) and different vaccination program histories. The flocks were compared with respect to serology (day old chicks, 21 days and slaughter at 42 days), histopathology of bursas of Fabricius at 21 and 42 days, infectious bursal disease (IBD) PCR on bursal tissue at 42 days, infectious bronchitis PCR from cloacal swabs at 42 days and final production performance results. Both flocks were placed and reared in Poland, in distant regions with medium-to-low Newcastle disease challenge in the field, but a history of high IBD challenge pressure.The novel DNA construct vaccine performance parameters of both flocks were compared to analyze the influence of management and vaccination history of the site on final production outcome.

Keywords: Broiler management, IBD, ND, Novel DNA construct vaccine

Comparison of different anticoccidial programs on coccidiosis control of broilers under floorpen conditions: shuttle versus full

Abstract ID: 246

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The efficacy of different anticoccidial programs for the control of coccidiosis in broilers was assessed in a floorpen study by comparing parasitological and zootechnical results between 4 groups: 1 untreated control group versus 3 treated groups. Treatment groups consisted of 7 replicate pens, each containing 20 broilers (Ross 308); i.e. 28 pens and 560 animals in total. All treated groups received nicarbazin/narasin (Combi) at 50/50 mg/kg in the starter feed (D0-D22) followed by (D22-D40) either nicarbazin/narasin (50/50 mg/kg) or by salinomycin at a concentration of 70 mg/kg or by narasin at a concentration of 70 mg/kg. Broilers were exposed to the natural coccidiosis pressure present in the farm; no artificial inoculation was performed. In order to evaluate coccidiosis pressure, lesion scoring) was performed on day (D)28 and D35. Feed intake and body weight (BW) was measured throughout the trial period. Daily weight gain (DWG) and feed conversion rate (FCR) were calculated. A low natural coccidiosis challenge was present, resulting in a total mean lesion score (TMLS) of 0.8 in the untreated control group on D28 and D35. TMLS were low in general in the other groups as well, with scores ranging from 0.2 (Combi-salinomycin) to 0.8 (Combi full) at D28 and from 0.7 (Combi-salinomycin) to 1.3 (Combi-narasin) at D35. The zootechnical results are clearly in the benefit of Combi-salinomycin. This is the only group with a significant ($P < 0.05$) improvement of the end body weight compared with the untreated control group. In comparison with the untreated control group (BW 3.29kg; DWG 79g/day), BW at D40 and DWG (D0-D40) were significantly higher in the Combi-salinomycin group (BW 3.44kg; 83 g/day). No significant differences could be seen in the other groups. No significant differences were seen on FCR. Under the conditions of the current trial it was shown that the shuttle program nicarbazin/narasin until D22 followed by salinomycin until slaughter is showing the best zootechnical results, indicating an advantage of using this programme over using a shuttle with narasin and using a full programme nicarbazin/narasin.

Keywords: Anticoccidials, Broilers, Coccidiosis, Floor pen

Comparison of preventive ionophore supplementation on gut health and performance in broilers: evaluation in a necrotic enteritis model

Abstract ID: 252

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Coccidiosis is one of the most important predisposing factors for the development of necrotic enteritis (NE). In this study the effect of in-feed prevention with coccidiostats on gut health and performance was evaluated in a NE-model in broilers. Trial groups consisted of 2 control groups (infected untreated control; IUC and uninfected untreated control; UUC) and 3 treatment groups receiving different coccidiostats in the feed: monensin sodium at 100 ppm, narasin at 70 ppm and salinomycin at 60 ppm from day (D)1 till the end of the study at D20. Each group consisted of 6 replicates of 16 birds. The induction of NE was performed by means of individual oral challenge with *Eimeria maxima* (10.000 oocysts) on D9 followed by an oral *Clostridium perfringens* (2.0 x10^8 cfu) challenge on D14. Intestinal lesions (coccidiosis and NE), mortality and performance (body weight gain (BWG), feed intake (FI) and feed conversion ratio (FCR)) were determined at D20. Challenge was successful and resulted for the IUC group in significant higher lesion for coccidiosis and NE (D15 and D16) and deteriorated BWG, FI and FCR compared to UUC in the acute infection phase (D9-D20). No significant difference was seen in NE related mortality. At the peak of infection, coccidiosis lesions were significantly reduced by monensin and salinomycin and NE lesions by all coccidiostats. The coccidiostats were all able to diminish the negative effects on performance of NE in comparison to the IUC: significant higher BWG and FI and significant lower FCR. More specifically, BWG, FI and FCR in the monensin-group and FCR in the salinomycin-group were even not significantly different from the UUC, indicating complete prevention from the negative effects of the challenge. In the current trial it was shown that the use of coccidiostats, in this case ionophores, can have a preventive effect on the occurrence of coccidiosis and the related NE-problems: by taking away one of the most important predisposing factors, coccidiostats can directly influence the risk for development of NE. Under the conditions from this trial the best results on coccidiosis lesion scores and performance were seen with monensin followed by salinomycin and then by narasin, probably linked with the effect on coccidiosis control.

Keywords: Broilers, Coccidiosis, Coccidiostats, Ionophores, Necrotic enteritis

Comparison of the gut microflora composition between different commercial layer farms: feasibility and limitations

Abstract ID: 329

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Many experimental but also field studies indicate that animal health is significantly influenced by the composition of its gut microflora. The composition is influenced by a variety of factors including feed, animal's age and breed, and the host immune system. It is suggested that management factors and environment may also significantly contribute to the bacterial colonization of the gut. But still it is difficult to identify influencing factors. Individual variations between birds kept under comparable environmental conditions may be high and study results have been controversial. The aim of this project was to compare the gut microflora composition of laying hens during one production cycle kept on four different farms within two differently fed groups on each farm (n = 1000-4200 birds/group). Gut contents of duodenum, jejunoileum and cecum were collected at the age of 24 – 26 and 55 - 56 weeks from a total of 10 clinically healthy birds/group, and investigated by Illumina sequencing. In addition health parameters including the macroscopical as well as microscopical evaluations of gut sections as well as body weight were determined. The body weight development was at most time points, which were investigated during the production cycle, comparable between feeding groups within one farm but significantly different between farms ($P < 0.05$). Neither macroscopical nor histological lesions were detected for the investigated birds, and variations in villi height as well as mucosal thickness and crypt depth as determined for the ileum were only minor between groups and farms. Our results indicate that age had a significant influence on the microflora composition throughout all farms ($P < 0.05$). Interestingly, despite similar genotype and age at the time of sampling on three farms, gut microflora composition varied significantly between farms ($P < 0.05$). The influence of the diets differing in their crude fiber content was less clear and the differences between feeding groups kept on one farm under comparable housing and management conditions were for most farms and time points not significant ($P > 0.05$). This study clearly shows that the farm environment is of major importance for the established gut microflora composition. Diet composition may modify the gut microflora but effects may be more difficult to detect under field conditions as compared to experimental studies conducted in a controlled environment.

Keywords: Commercial farms, Diet, Gut health, Layers, Microflora

Comparison of Two European Gel-Type Diluents vs. Water

Abstract ID: 276

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Control of coccidiosis by vaccination is gaining interest around the world; a result of increased demands for "no antibiotic ever" production. Coccidiosis immunity begins with uniform vaccine application, but must continue under field conditions for multiple complete life cycles of each *Eimeria spp*. parasite to induce immunity. The best opportunity for uniform initial application is the hatchery, where conditions are controlled and chicks are confined to a chick box for spray via a spray cabinet. Although coccidiosis vaccines are often mixed with water for application via spray cabinet, gel-type diluents have been introduced with the goal of increasing initial oocyst uptake via day-old spray. Water and two commercial gel-type diluents available in Europe were evaluated with respect to oocyst uptake, challenge response, the ability to maintain oocysts in solution, the onset of immunity, and the influence of an oral adjuvant contained in one of the gels on the onset of coccidiosis immunity. Results show significantly better uptake of oocysts from a precocious strain coccidiosis vaccine using gel spray application instead of traditional water spray. Birds challenged at 21 days post-spray vaccination had significantly better group weights after challenge with *E. tenella*, *E. necatrix* or *E. brunetti* when vaccinated using a gel diluent vs. water. Comparison of two commercial gel diluents demonstrated better oocyst suspension in the more viscous formulation vs. the more aqueous gel. The oral adjuvant in one gel did not induce an earlier onset of immunity when the same precocious vaccine strains were administered in each diluent via gavage, indicating that the "work" of immunity building largely takes place in the field, after the initial vaccination. Getting as many oocysts into the birds as uniformly as possible to initiate the immunity building process is more important than having an oral adjuvant to initiate the process. The study demonstrates that uniform and optimized uptake of oocysts of precocious vaccines can have measurable effects on the onset of immunity and the response to challenge at 21 days of age.

Keywords: Coccidiosis, Gel diluent, Immunity

Comparison two type of inactived avian infectious bronchitis vaccines (M41 and M41/Dutch variants) in protection against Variant-2

Abstract ID: 83

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Avian infectious bronchitis (IB) is a contagious poultry disease with huge losses around the world, and the incidence of its virulent genotypes is very common. Variant 2 is one of IBV genotype with respiratory and kidney tropism considered as a very opportunistic genotype recently. It is spread from the middle east to Europe with no effective control parameters available. Strategies to control this virus has not been completely successful and finding an appropriate one is so helpful. Different killed IB vaccines are available throughout the world, from M41 (Massachusetts) only to mixed M41 and Dutch variant's (D274 and D1466) vaccines. In this study, 3 out of 5 groups were vaccinated with H120 vaccine (Priming), and two received no vaccine as control groups. After three weeks after the first vaccination, two of the three vaccinated groups, one was vaccinated with M41 and another with M41+Dutch vaccines. Three weeks post vaccination (Inactive), all four groups except the control one was challenges with variant 2 virus and the average ciliostasis score, cross-protection, and virus shedding (q-RT-PCR) rates were evaluated. The relative protection rates were 60% and 63% for M41, and M41+Dutch vaccinated groups, respectively. Also, in the M41+Dutch vaccinated group, the viral shedding was lower than M41 group. Although the difference was not significant, it could be concluded that adding Dutch variants in killed vaccines helps to protect the flock against variant 2. It is necessary to mention that adding variant 2 to the killed vaccine could also improve the efficacy of vaccines.

Keywords: Cross Protection, IBV, Vaccination

Comprehensive epidemiological investigations support the role of Histomonas meleagridis for systemic translocation of *Escherichia coli* in chickens

Abstract ID: 269

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Histomonosis caused by *Histomonas meleagridis* infection in poultry is of growing attention, especially due to the lack of promising therapy and prophylaxis. In addition to turkeys, the significance of *H. meleagridis* in layers has now been acknowledged and it can cause pathomorphological lesions mainly localized in caeca. Recent field reports suggested that histomonosis in laying and breeding chickens often coincide with colibacillosis, a systemic bacterial infectious disease caused by *Escherichia coli*. Considering the fact that the gut harbours a pool of *E. coli* isolates and *H. meleagridis* destroys intestinal integrity, it can be hypothesized that the parasite might act as a predisposing factor for systemic translocation of the bacteria which leads to colibacillosis. In this study, we performed pheno- and genotyping of *E. coli* isolates from intestine and systemic organs of birds affected by histomonosis and colibacillosis in order to investigate the systemic dissemination of the bacteria. In total, 29 birds were necropsied from 11 affected flocks that showed elevated mortality and drop in egg production. From each of the birds, up to three *E. coli* isolates were collected from intestine, heart and liver, thus a total of 251 isolates were characterized by serotyping, phylogenetic grouping, *in vitro* virulotyping and pulsed-field gel electrophoresis (PFGE). Macroscopically, egg peritonitis was recorded in all birds while fibrinous typhlitis, a typical lesion of histomonosis was additionally seen in 18 birds. Serotyping revealed that most of the isolates belonged to serotype O2:K1 (n=94) followed by O78:K80 (n=59) or O1:K1 (n=20). Isolates from the intestine and extra-intestinal organs were mainly assigned to phylogroup B2 (n=136) that also contained a relatively higher number of virulence-associated genes. *E. coli* isolates collected from 19/29 birds had at least one common phylogenetic group among all three organs within a bird. The PFGE cluster analysis resulted in 46 different profiles among which eight PFGE types that included 60.6% of total isolates (n=152/251) were shared among intestine, heart and liver indicating for the presence of identical *E. coli* types in the gut and extraintestinal organs. Collectively, the epidemiological findings provided a strong support for the assumption that *E. coli* present in the intestine are translocated into systemic organs during the course of *H. meleagridis* infection in chickens. Thus the parasite can not only act as a primary pathogen but also support others to exacerbate colibacillosis.

Keywords: Chickens, Colibacillosis, Escherichia coli, Histomonas meleagridis, Histomonosis

Delivery of oligodeoxynucleotides containing CpG motifs (CpG-ODN) by the intrapulmonary route against bacterial septicemia in neonatal broiler chickens

Abstract ID: 443

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Oligodeoxynucleotides (ODN) containing CpG motifs (CpG-ODN) are effective immunostimulatory agents against a variety of viral, bacterial, and protozoan diseases in different animal species including poultry. We have recently demonstrated that *in ovo* delivery of CpG-ODN confers protection against bacterial septicemias in neonatal chickens. The objective of this study was to investigate the effectiveness of intrapulmonary (IPL) delivery of CpG-ODN against *Escherichia coli* infection in neonatal chicks. Chicks were IPL delivered CpG-ODN or saline at hatch. Three-days later chicks were challenged with $1x10^4$ or $1x10^5$ cfu of *E. coli*. Chicks treated with CpG-ODN by the IPL route had significantly lower clinical signs and bacterial load compared to the group treated with saline ($P<0.05$). CpG-ODN treated groups were significantly protected against *E. coli* septicemia. We have seen a dose and exposure time-dependent immunoprotective effects of IPL of CpG-ODN in chicks. We found that IPL delivery of CpG-ODN can induce protective immunity as early as 6 h post-administration and remain effective until day 5 post-treatment. This study has demonstrated that CpG-ODN delivery by IPL route can be a promising alternative to antibiotics for inducing protective immunity in chicks during the neonatal life.

Keywords: Alternatives to antibiotics, Immunostimulation, Intrapulmonary, Neonatal broiler chickens, Septicemia

Dermanyssus gallinae as a vector of selected bacterial and viral diseases in flocks of hens in Poland

Abstract ID: 603

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The poultry red mite (*Dermanyssus gallinae*) is the most important parasite of laying hens kept in cages. The infestation of *Dermanyssus gallinae* in layers results in stress, decreasing egg production, anaemia and it also creates public health concerns.
Apart from these direct effects of hematophagus parasitism poultry red mites (PRM) has also been considered as a vector for a number of avian viral and bacterial pathogens of poultry.
The purpose was to determine the role of PRM as a vector of selected pathogens in flocks of laying hens in Poland i.e. *Mycoplasma gallisepticum*, *Mycoplasma synoviae*, *Salmonella* spp., *Campylobacter* spp., adenoviruses and the viruses of the Marek's disease. The research was carried out in in *Dermanyssus gallinae* obtained from 35 flocks of laying hens kept in cages. From the samples each 10 specimen of *Dermanyssus gallinae* representing the collective sample from which then the DNA was isolated were selected at random for the research. The DNA genetic material isolated with use of the commercial set for isolation of DNA genome served as a matrix for PCR reaction thus facilitating determination of presence of the specified pathogens of laying hens. Conditions of the reaction and starters were determined based on the literature data. The obtained products will be sequenced in order to determine their genetic affiliation and to differentiate them from the vaccination strains. From the performed research it results that the presence of the genetic material of *Mycoplasma gallisepticum*, *Mycoplasma synoviae*, adenoviruses was not stated in any isolate of Dermanyssus gallinae.
However the presence of *Salmonella* spp. was observed in 10/35 and of *Campylobacter* sp. in 18/35 of the researched samples.

Keywords: Dermanyssus gallinae, PCR detecion, Poultry pathogens

Development and application of a quantitative real-time PCR for the detection of Enterococcus cecorum

Abstract ID: 151

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Enterococcus cecorum (EC) infection is an important bacterial disease in broiler chickens today. However, many aspects of epidemiology and pathogenesis are still unknown. Other detection methods than classical cultivation are needed to answer these questions. In the present work, the validation and application of a newly developed quantitative TaqMan real-time PCR (qPCR) assay based on the 16S-rRNA-gene for the detection of EC is described. Fifty EC strains isolated from different animal species were successfully detected with the qPCR, while none of the other 26 examined bacterial species were tested positive during validation procedure. The detection limit of the PCR was 6.25 CFU/mL PBS. Additionally, the qPCR assay was also more sensitive than classical cultivation method, using intestine and organ samples from broilers. Application of the PCR setup was tested in two different broiler production cycles of one farm. In cycle 1, broilers showed signs of EC infection from day 24 post hatch onwards while broilers in cycle 2 developed no clinical signs. Two different colonization patterns were found in the two cycles with the qPCR using cloacal swabs. Broilers in cycle 1 showed significantly ($P \leq 0.05$) higher detection rates of EC at the day of placement and throughout the cycle than broilers of cycle 2. Additionally, varying detection rates were found in different parts of the intestine within one cycle. The presented qPCR for EC is highly specific, considerable more sensitive than classical isolation and was able to show differences in colonization patterns in a broiler flock with later EC disease outbreak compared to a healthy cycle. These findings may be explained by infection with different strains, pathogenic EC isolates are probably more effective in colonization than commensal isolates. A high correlation was found between qPCR results from cecum and cloacal swabs in this study, indicating that cloacal swabs can be used to examine intestinal colonization of broilers with EC. The new qPCR setup may be used to answer open questions concerning epidemiology and pathogenesis and significantly improves the diagnostic of EC infections.

Keywords: Broiler chickens, Colonization, Enterococcus cecorum, QPCR

Different strains of Clostridium perfringens cause different levels of severity of necrotic enteritis in broiler chickens

Abstract ID: 304

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Necrotic enteritis (NE), mainly caused by the gram-positive anaerobic bacterium *Clostridium perfringens* (*Cp*), is an enteric disease with great economic impact for the broiler industry. The prohibition of in-feed antibiotics has resulted in an increase in NE incidences globally. The severity of the disease in broilers depends on many factors. However, the type of *Cp* strains used plays a critical role. This study examined the impact of two *Cp* strains, i.e. NE18 and NE36, on performance, lesion score and gut permeability of broilers. Ross 308 broilers (n = 468) were assigned to a 2 × 3 factorial design (Feed: With/Without antibiotic) × (Challenge: No / NE18/ NE36). Oral administrations of *Eimeria* species (d9) and two stains of *Cp* (d14) were used to induce NE in the challenged groups. On d16 two birds from each pen were inoculated with FITC-d and serum samples were subsequently obtained for leaky gut evaluation. FITC-d amount was measured using a microplate reader (Synergy HT, Multi-mode microplate reader, BioTek Instruments, Inc., VT, USA). Broiler performance (Feed Intake, FI; weight gain, WG; and feed conversion ratio, FCR) was analysed for d0-24 period. Intestinal lesion scoring and gut permeability were measured at d16. Weight gain in the challenged group was significantly lower ($P < 0.001$) compared to the unchallenged birds at d24. Challenge also reduced the FI significantly ($P < 0.001$). FCR was significantly higher in the groups challenged with NE18 ($P < 0.001$) and NE36 ($P < 0.001$) compared to non-challenged birds. Antibiotic diets significantly decreased FCR ($P < 0.05$) and increased WG ($P < 0.05$) in all groups. Birds challenged with NE36 showed higher FCR ($P < 0.001$) compared to the birds challenged with NE18 indicating a more severe impact of the NE36 than NE18. Ileal lesion scores and gut permeability were not significantly different between the two strains, but numerically higher number of lesions and gut permeability were observed in birds challenged with NE36. Supplementation with antibiotics reduced ($P < 0.05$) gut permeability in the birds of all groups. The findings of this study indicate that the severity of necrotic enteritis disease in challenge model can be modulated by the use of different *Cp* strains.

Keywords: C. perfringens, Lesion score, Necrotic enteritis, Performance, Strain

DNA construct vaccine applied in hatchery as a tool to combat ILT virus circulating in the field

Abstract ID: 52

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Herpesvirus of Turkey (HVT) is a naturally-occurring virus from turkeys, also known as Marek's disease virus serotype 3. This virus has been used for decades as a vaccine to aid in the protection against Marek's disease in chickens. Because it is a naturally-occurring non-pathogenic virus (not treated or attenuated), it cannot revert to virulence when used as a vaccine. Like other herpesviruses, HVT does produce a persistent infection, inducing life-long immunity. HVT has a 159, 160 base-pair genome encoding for an estimated 99 different proteins. The genome includes unique long (UL) and unique short (US) genomic regions and short repeat (RS) and long repeat (RL) segments. HVT was selected as the appropriate backbone for genetic construct vaccines because of the characteristics of non-pathogenicity, persistence of infection, and stability when genetic material is inserted into its genome to encode for proteins that are not naturally expressed by the virus. But the insertion of genetic material does not automatically result in a stable HVT virus that can express the desired antigenic proteins encoded by the added genes. Special sequences of genes must be added to the beginning and end of the desired insertion called "promoters" and "terminators." These unique and proprietary genes enable researchers to construct an HVT virus that encodes not only for proteins that contribute to Marek's disease protection, but also to protection of ILT , to minimize losses due to ILTV infection. Infection Laryngotracheitis is a common problem of poultry industry worldwide and used to be called "man-made disease", since majority of ILT outbreaks are caused by CEO ILT vaccine virus reversed to different levels of pathogenicity. Innovax ILT is a genetic construct vaccine, which creates an opportunity to eliminate from immunoprophylaxis programs use of conventional ILT vaccines based on CEO or TCO ILT virus.The paper shows how serology performed in conventional ELISA test (from BioChek) versus IDvet gI specific test (from IDvet) can help to learn about ILT status of birds in commercial poultry units and furthermore – to decide about final vaccination program, accordingly to this knowledge. The paper also analyzes potential reasons for vaccine failures.

Keywords: DNA construct vaccine, Innovax ILT, Serological investigation

Effect of age of vaccination on antibody titers, clinical signs occurrence and performances after challenge with H9N2 on broilers

Abstract ID: 355

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In order to evaluate the impact of vaccination age on protection against H9N2 field challenge MDA (maternally derived antibodies) positive broiler chicks were vaccinated at different ages (D1, D7 and D14) and challenged at 28 days in controlled conditions. In this trial, one group was non-vaccinated and challenged in the same controlled conditions, to compare the seroconversion, clinical signs, lesions, virus shedding and body weight. Vaccination seroconversion in non-challenged groups showed high ELISA titers for the groups vaccinated at D7 and D14. Those vaccinated at D1 showed similar titers as the non-vaccinated chicks. As for the non-vaccinated groups seroconversion after challenge showed the same evolution for the D1. The birds vaccinated at D7 and D14 reacted strangely differently after D35. High titers were observed in the D7 group and the lowest titers in the D14 group. Clinical signs scoring and necropsy showed no difference between all groups and only two mortalities were noted in the group vaccinated at D1. Virus shedding measured by PCR was continuous in non-vaccinated flocks while less shedding was noted in birds vaccinated at D7 and D14. Body weight evolution and FCR showed an advantage for groups vaccinated at D7 and D14. The experiment shows that late vaccination for H9N2 in MDA positive broiler birds between day 7 and D14 can provide higher titers and better performances.

Keywords: Antibodies, Challenge, H9N2

Effect of fluralaner treatment on the production results at commercial layers farms in Poland

Abstract ID: 303

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Fluralaner is a novel izoxasoline, which is used for reducing infestation of *Dermanyssus gallinae* at commercial layers and broilers breeders flocks. The relevant publications and research show, that *Dermanyssus gallinae* infestation is still very important problem in poultry production, because of avian diseases spreading like salmonellosis, mycoplasmosis, Newcastle disease or avian influenza. Red mites infestation can induce also stress in the birds, cannibalistic behavior, increased feed and water intake and decrease animal health. At commercial layers farms, high level of red mites infestation leads to a drop in eggs production, higher mortality and increasing morbidity of diseases. The aim of the study was to monitoring of red mites infestation and production results after fluralaner treatment at commercial layers farms in Poland. The level of red mites infestation before fluralaner application was very high or medium. The parasites were visible on the eggs and on the layers cages as an colonies. Usually the mortality rate was increasing at intervals of few weeks as a result of virus or bacterial infections like *Escherichia coli*. During the study, production rates like egg yield, mortality and all information about the treatment and pathogens infections were collected during the minimum 6 months after fluralaner treatment. The collected data was compared with production rates collected 3 months before fluralaner application. Additional at all farms, which were included to this studies, were provided red mites infestation level monitoring every 1 month using red mites traps. The preliminary results show a decreasing mortality and health improvement in commercial layers flocks and red mites infestation reduction after fluralaner treatment.

Keywords: Fluralaner, Layers, Production results, Red mites

Effect of selected yeast fraction on the growth of Clostridium perfringens: quantitative determination of growth inhibition and adsorption capacity

Abstract ID: 244

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Yeast fractions are used worldwide as promising nutritional solutions to enable the reduction of antibiotics use with respect to the promotion of health and performance in livestock, based on their capacity to bind enteropathogenic bacteria, and for their immunomodulatory activity. The *in vitro* and *in vivo* mode of action of YCW yeast fractions, with respect to improving animal performance, is still not entirely understood. The inhibition mechanism exerted by yeast fractions on pathogens seems to be limited to some specific Gram-negative enteropathogens (*Salmonella* and *E. coli*), although several *in vivo* studies report their effect also on Gram positive pathogens like clostridia. The literature lacks *in vitro* evidences for antimicrobial effect of yeast fractions on *C. perfringens* strains. The effectiveness of yeast fractions in inhibiting the growth of several *C. perfringens* strains was quantitatively determined, and one product out of four materials was selected as the best candidate for *C. perfringens* inhibition. The selected product, at an optimal dosage of 1.25 mg/mL, increased the lag phase duration, and reduced the maximum growth rate and the final cell count in a significant manner with respect to the control. Interestingly, the study proved that the product adsorbed *C. perfringens* cells in a dose and time dependent manner. Equilibrium isotherms showed that the cell adsorption onto the product was fast, stable over the time, and occurred with high affinity and capacity. The selected product sequestered up to ca. 10^4 cells of *C. perfringens* strains per mg, including the NetB toxin producing strain. To the best of our knowledge, this is the first report showing the *in vitro* efficacy of yeast fraction products to inhibit the growth of *C. perfringens*, and to reduce the culturable cells by an adsorption process. The *in vitro* approach proposed herein is as a powerful approach to study the adsorption of aerobic or anaerobic pathogens by eubiotics, thanks to the possibility of modulating culture conditions.

Keywords: Adsorption, Antimicrobial, Clostridium, Eubiotics, Yeast fraction

Effects of a dry hydrogen peroxide (DHP) air sanitation system used in an egg cooler on hatchability and chick quality

Abstract ID: 301

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In commercial poultry production, hatcheries are a source of continual contamination. A sanitation method that could continually clean and disinfect a hatchery in support of daily cleaning procedures would be valuable and ,for this reason, a novel, commercially available, gaseous dry hydrogen peroxide (DHP) system has been introduced to continually combat microbes in the air and on surfaces in hatcheries. Preliminary studies have shown that this sytem can indeed reduce the microbial population in hatcheries and on the surface of eggs, however the effects of this new system on hatchability and chick quality need to be evaluated. A total of 3,960 fertile eggs were collected from an approximately 40 week old Ross 308 broiler breeder flock maintained at the Poultry Science research farm and distributed in 2 treatments: control (no disinfection) and treated. For the treated group, one DHP air sanitizer was placed inside an egg cooler at the Poultry Science research farm and two other machines were placed in the common area outside. Both areas were treated for 7 days prior to placement of eggs, and then eggs were collected and placed inside the cooler over a four day period. Eggs were stored for 3 days after the last collection prior to placement in the incubator. During pre-treatment and egg storage, DHP levels were measured inside the cooler and air samples were taken to evaluate environmental microbial load. After storage, eggs were placed into a single stage Natureform incubator. For the control group, all DHP machines were removed from the cooler and external room five days before placing the eggs and the egg cooler was cleaned. During the treated phase of the trial, a daily increasing level of DHP was measured in the egg cooler, with an average level of 12 ppb, and a concomitant reduction in microbial load from air samples was seen, confirming that the room was treated and the system was destroying microbes as expected. A 1.1% increase in hatch and a 2.5% increase in hatch of fertile were seen for the treated group, as well as a 1.7% decrease in early dead and a 1% decrease in late dead embryos. In total, this data demonstrates the positive effect that reducing microbial loads on hatching eggs using the DHP system can have on hatchability.

Keywords: Dry hydrogen peroxide, Hatchability, Hatchery sanitation, Microbial reduction

Effects of diet dilution on bone development and mineralization of coccidiosis-infected broilers

Abstract ID: 510

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Growth rate (GR) of modern fast-growing broilers may adversely affect their long bone development. Furthermore, broiler coccidiosis impairs aspects of bone mineralization. We tested the hypothesis that delaying GR of fast-growing broilers via diet dilution will allow for better bone mineralization with more pronounced effects during coccidiosis. 384 male Ross 308 day old broilers were distributed into 48 pens and offered a common starter diet for 6 days. Thereafter, diet was gradually diluted at 4 levels of cellulose which was expected to limit FI and consequently ADG. Following a period of adaptation birds were allocated to one of dilution treatments of a high quality diet at d10: 0, 5, 10, 15%. Hence, 4 groups (R0-3, respectively) were formed and half the pens of each group were orally infected with 7000 sporulated *Eimeria maxima* oocysts at d13 (d0 post infection (pi)). Six birds per treatment combination were sampled for tibia and femur bones at d12 pi for assessing breaking strength (BS; N), ash weight (AW; g), as a proportion of BW at dissection, bone density (robusticity index; RI) and percentage ash (PA). Diet, infection status and their interaction were treated as factors and data were analysed with GLM. Following infection, R1-3 birds showed significantly reduced BW by approximately 8, 14 and 23% respectively in comparison to R0 birds. Infection with *E. maxima* significantly reduced ($P < 0.05$) femur and tibia BS, and tended to reduce tibia AW ($P < 0.1$). There was not interaction between infection and diet dilution. Diet dilution affected ($P < 0.05$) femur RI, BS, and tended to increase ($P < 0.1$) femur PA and tibia AW; R3 birds had significantly higher femur BS in comparison to R0 birds. In conclusion, aspects of bone mineralization were improved at reduced GR irrespective of infection status.

Keywords: Bone mineralization, Broiler chicken, Coccidiosis, Diet dilution, Growth rate

Effects of feeding Diamond V fermentation metabolites on Avian Pathogenic E. coli prevalence and antibiotic resistance of E. coli in ceca samples taken from commercial broilers and turkeys

Abstract ID: 284

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Colibacillosis is an economically important disease that is caused by an extraintestinal pathogenic *E. coli* known as Avian Pathogenic *E.* coli (APEC). Large-scale field trials were conducted to determine the effects of feeding a fermentation metabolites product (FM) on reducing the prevalence of APEC and the resistance of *E. coli* to the antibiotics ceftiofur, florfenicol and enrofloxacin in ceca samples taken from commercial broilers and turkeys. Ceca samples were taken in commercial processing plants. A total of six broiler companies (98 houses) and four turkey companies (34 houses) were monitored. Birds in houses were fed a company standard diet (CON) or a diet that contained 1.25 kg/MT of the FM. One cecum/bird was collected from 50-100 birds per house (3,100 broiler and 1,770 turkey ceca) during evisceration at the processing plant. Ceca were shipped overnight to Iowa State University for analysis. APEC prevalence was determined using a pentaplex PCR assay while antibiotic resistance was evaluated using micro-broth dilution. Data were analyzed in SAS using the GLM procedure with feeding treatment as the main effect, company as a random effect, and significance considered at $P \leq 0.05$. Feeding the FM significantly reduced ($P < 0.0001$) APEC prevalence compared to CON for broilers (32.1% vs. 74.7%, respectively) and turkeys (43.6% vs. 92.2%, respectively). For broilers, feeding the FM significantly lowered antibiotic resistance ($P < 0.0001$) compared to CON (florfenicol 25.0% vs. 90.7%; ceftiofur 26.9% vs. 68.0%; enrofloxacin 15.4% vs. 56.3%, respectively). In turkeys, feeding the FM significantly lowered antibiotic resistance ($P < 0.0001$) compared to CON (florfenicol 26.6% vs. 87.2%; ceftiofur 39.6% vs. 75.3%; enrofloxacin 22.3% vs. 42.4%, respectively). These data suggest that the addition of the FM to the diet in commercial broilers and turkeys can reduce the prevalence of APEC as well as the antibiotic resistance of *E. coli*.

Keywords: APEC, Antibiotics, Broilers, Fermentation metabolites, Resistance

Efficacy of a monensin/nicarbazin combination in the control of coccidiosis in turkeys under floor pen conditions using a seeder model

Abstract ID: 251

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The efficacy of monensin/nicarbazin at a concentration of 40mg/kg (40ppm) monensin sodium and 40mg/kg (40ppm) nicarbazin, administered in feed for 85 days was assessed in turkeys after an experimental coccidiosis challenge. Body weight (BW), daily weight gain (DWG) and feed conversion ratio (FCR), intestinal lesion scoring of seeder birds (D22) and contact birds (D28, D31 and D34) (relevant pathogenic species, *E. meleagrimitis* and *E. adenoeides*; scoring system developed at ANSES - Laboratory of Ploufragan - France) and oocyst shedding of monensin/nicarbazin-treated birds were compared to an infected untreated control (IUC) and an uninfected untreated control (UUC) group. Each group consisted of 8 replicate pens (30 Hybrid Converter males/ pen). On D17, in IUC and monensin/nicarbazin group, 8 birds (=seeders) per pen were inoculated with a mixture of *Eimeria meleagrimitis*, *E. adenoeides* and *E. dispersa* (European origin). Birds from UUC were sham-inoculated. The challenge was successful as proven by the significantly higher oocyst production and intestinal lesion scores in the IUC. To prevent cross-contamination of the UUC, this group was located at the front of the house and throughout the trial a negative house effect was seen on the performance of the UUC. Nonetheless, the impact of the infection on performance was seen as BW, DWG and FCR in the UUC group was better (especially in acute phase of infection) compared to the inoculated birds from the IUC. Administration of monensin/ nicarbazin significantly suppressed oocyst shedding and resulted in significantly lower *E. meleagrimitis* and *E. adenoides* lesion scores as compared to IUC. On D22, inoculated birds from the monensin/nicarbazin group had significantly lower total coccidiosis lesion scores compared to birds from IUC. In the contact birds, total coccidiosis lesion scores tended to be lower on D28 and D34 and were significantly lower on D31. Performance of monensin/nicarbazin supplemented birds was positively affected: BW, ADG and FCR were significantly improved in comparison to IUC. The results from this trial demonstrate the efficacy of a monensin/nicarbazin combination for prevention of coccidiosis in turkeys. Under present study conditions, we can conclude, based on lower oocyst shedding, lower coccidiosis lesion and better performance, that the administration of monensin/nicarbazin proved efficacious against coccidiosis infection in turkeys.

Keywords: Coccidiosis, Coccidiostats, Turkeys

Efficacy of different vaccination programs against colibacillosis after intratracheal challenge at the beginning of laying period in commercial layers

Abstract ID: 356

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Vaccination, either commercial live and inactivated and/or autogenous vaccines, has been used for the protection against colibacillosis. In the present study, the efficacy of different vaccination programs in commercial layers was assessed after experimental intratracheal challenge with different serotypes of *E.coli* strains at the beginning of lay. Commercial pullets from a rearing farm were divided in 4 groups according to the colibacillosis vaccination schedule: A: control unvaccinated; B: vaccinated 3 times with a live-O78 strain commercial vaccine; C: vaccinated twice with an inactivated oil adjuvanted autogenous vaccine containing 3 *E.coli* strains (serotypes O18, O78 and O111); D: 2 administrations of a live-O78 strain commercial vaccine followed by 1 injection of the inactivated oil adjuvanted autogenous vaccine. Twenty birds from each group were placed in 3 different experimental rooms and challenged intratracheally with a different *E.coli* strain (serotypes O18, O78 and O111) used for the production of the autogenous vaccine. The evaluation of the efficacy of each vaccination program, was made by daily monitoring of the birds and recording of mortality. Dead birds were necropsied for identifying characteristic lesions in target organs (heart, liver, air sacs, lungs, peritoneum). One week after challenge, all birds were euthanized and necropsied. Samples from bone marrow and organs were collected, in an attempt to recover *E.coli*. Groups A and B showed 45% and 35% mortality respectively when challenged with O78 strain and between 0% and 10% when challenged with O18 and O111 strains. Finally, groups C and D had 0% mortality after each challenge with O78 and O111 strain, whereas challenge with O18 strain lead to 5% and 0% mortality for groups C and D respectively. Concerning the evaluation of lesion scoring after O78 challenge, the lowest lesion score for all target organs was reported to Group C birds. In the O18 strain challenge, birds from Group D showed the lowest lesion score for all target organs, whereas in the third challenge – strain O111 – the lesion score was similarly low for both groups C and D. In conclusion, our study showed that the use of at least one autogenous vaccine including 3 different serotypes of *E.coli* strains had a positive effect in bird protection after challenge with each one of these 3 strains. This protective effect is particularly pronounced when mortality is high in the control group - which is dependent on the virulence level of the strain.

Keywords: Autogenous vaccine, Commercial layers, Escherichia coli, Intratracheal challenge, Serotype

Efficacy of fluralaner administered through drinking water with or without brilliant blue in layer hens suffering from poultry red mite infestation and effect of treatments on performance

Abstract ID: 145

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Fluralaner is an acaricide appropriate for drinking water treatment of chickens infested with poultry red mites (PRM). In some farms, accurate administration of medicines through the drinking water can be complex because of the numbers of birds involved and infrastructure of the housing and watering systems. To visualize the product flow and to avoid mistakes it can be useful to add a dye such as brilliant blue to medicated water. It was the aim of the present study to evaluate the effect of combined administration of brilliant blue and fluralaner on the efficacy of treatment against PRM. A trial was set up in 2 similar infested houses of the same farm with 71,000 and 56,000 layer hens of 67 weeks old, belonging to the same breed and kept in enriched cages. All hens were treated with fluralaner at a dose of 0.5 mg/kg body weight through the drinking water, twice with a 7-days interval. Fluralaner was added from a stock solution to the drinking water lines over a period of approximately 10 hours. In the first house, the fluralaner stock solution was used as such while in the second house, 1.5 grams of brilliant blue were added per liter of fluralaner stock solution. In order to evaluate effects of the treatments, a mite trap assay was used at 2 days before and at 19 and 91 days after the first administration of product. At each occasion, 20 mite traps were evenly distributed over each house for 48 hours. Then, the number of mobile mite stages in the traps was counted under a reversed microscope. The percentage of mite reduction after treatment was over 99.9% in both flocks. At 91 days after treatment, no mites were found at all. The results demonstrate that fluralaner is very efficient in eliminating mites from poultry houses and that efficacy is not reduced by concurrent use of brilliant blue. Performance of the flocks was monitored by gathering production data on a weekly basis from 5 weeks before until 5 weeks after the first treatment administrations. Although egg production in both flocks was already around 7% above the production standards prior to treatment, it rose by 0.71% and 0.83% afterwards. Egg masses increased accordingly and feed conversion ratios went down. These data demonstrate that even in flocks with a superior production level, performance can be increased through effective control of PRM.

Keywords: Brilliant blue, Compatibility, Fluralaner, Performance, Red mite

Efficacy of Gallifen® 40 mg/ g premix against natural infections of Ascaridia galli and Heterakis gallinarum in layer chickens

Abstract ID: 28

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Worm infections are common in poultry and have a significant impact on the health status and zootechnical performance. Next to nutrient absorption, helminth infections reduce bodyweight gain, egg production and hatchability. Irritation of the intestinal mucosa can lead to diarrhoea. Heterakis gallinarum is also well known to play an important role in the transmission of Histomonas meleagridis. Two flocks were selected and housed in similar housing conditions and managed in a similar manner. One flock was treated with Gallifen 40 mg/ g premix at a target dose of 1.0 mg fenbendazole/ kg bodyweight for 5 consecutive days, corresponding to 17 ppm fenbendazole in the feed. The second flock was untreated and considered as the untreated control group. At the day on which the treatment started and 5 days after the end of the treatment, 30 and 25 layers respectively, were necropsied and A. galli and H. gallinarum worm counts (adult stadia, immature L4 and L5) were performed. For both the treated and the control group, the geometric means (GM) at the end of the study period of 10 days were calculated and the percentage reduction determined. At study completion, adult A. galli worms were found in 64 % of the birds (GM = 2.61) randomly selected from the untreated control flock and in none of the 25 birds from the treated flock. Immature L5 A. galli worms were observed in 36 % of the birds (GM = 0.61) of the untreated control birds but none in the treated group. No L4 A. galli stadia were counted in the control group. Thus, both adult and immature L5 A. galli worm counts were reduced by 100 %. Adult H. gallinarum worms were present in 92 % of the untreated layers (GM = 44.1) and in 36 % (GM = 1.1) of the treated flock, resulting in an adult worm count reduction of 97.5 %. A reduction by 86.7 and 94.5 % in the immature L4 and L5 worm counts respectively was noted. This study confirmed the efficacy of Gallifen 40 mg/ g premix at a target dose of 1.0 mg fenbendazole per kg bodyweight per day for 5 consecutive days against both adult and immature stages of A. galli and H. gallinarum.

Keywords: A. galli, Gallifen fenbendazole efficacy, H. gallinarum, Heterakis Ascaris

Efficacy of specific compositions of 1-Monoglycerides of Short- and Medium Chain Fatty Acids in controlling Salmonella typhimurium and other serotypes of Salmonella spp. in broiler chickens and in vitro conditions

Abstract ID: 165

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Specific compositions of 1-Monoglycerides of Short- and Medium Chain fatty acids showed antibacterial efficacy *in vitro* against *S. typhimurium*, *S. enteritidis, S. jawa*, *S. heidelberg* and *S. dublin*, *S. infantis*, *S. alachua* and other serotypes at pH 6-7, corresponding to the pH values of the gut of chickens. The MIC of the 1-Monoglycerides compositions against above *Salmonella* serotypes resulted to be comprised between 0.01% and 0.1%, while *Lactobacillus plantarum* and *Lactobacillus acidophilus* were not inhibited. A trial with broiler chickens was carried out to assess the efficacy of a specific 1-Monoglycerides composition in reducing *Salmonella typhimurium* counts in the caeca after experimental infection. Sixty female one-day old Ross 308 broiler chicks were randomly housed in isolators and allotted to two treatments. The control group received a standard commercial feed, while the 1-Monoglycerides group received the same feed supplemented with 0.3% of the composition during the whole experimental period. At 7 days of age all the birds were challenged *via* endoesophageal inoculation with 1mL of saline solution containing $1x10^7$ CFU/bird of *S. typhimurium*. At day 14, 24 and 34 ten chickens from each group were sacrificed; samples from caeca were analyzed for *Salmonella* spp. counting. Results showed that in the 1-Monoglycerides group the Salmonella CFU counts was reduced by 6 $logs_{10}$ compared to the control group (p = 0.02. Two-way ANOVA). In the control group a mortality of 30% was recorded, while no mortality was observed in the 1-Monoglycerides group. The 1-Monoglycerides composition prevented *Salmonella typhimurium* colonization in the caeca and the pathogenic effects of the bacterium which caused high mortality in the control group. The *in vitro* trial showed a selective antibacterial effect of the 1-Monoglycerides compositions against *Salmonella* spp. without inhibiting beneficial *Lactobacillus*. Specific compositions of Short- and Medium-Chain 1-Monoglycerides may represent an alternative to enteric antibiotics in broiler feed.

Keywords: Antibiotic-free, Monoglycerides, Salmonella Typhimurium, Salmonella spp.

Establishment of the cecal microbiota of broiler chickens supplemented with protected sodium butyrate alone or in combination with essential oils and challenged with coccidia and Clostridium perfringens

Abstract ID: 143

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The objective of this study was to determine the effects of protected sodium butyrate (SB), and protected sodium butyrate plus essential oils (carvacrol and ginger; SBEO) on the establishment of the cecal microbiota of broilers induced to necrotic enteritis (NE). Birds were assigned to 4 treatments (8 replicates of 58 birds): (1) unchallenged and unsupplemented control; (2) challenged and unsupplemented control; (3) SB supplementation and challenged; (4) SBEO supplementation and challenged. On d 13, challenged birds were orally inoculated with ~5,000 Eimeria maxima oocysts. On d 18-19, the same birds were exposed to C. perfringens via drinking water. Cecal excreta was collected at d 12, 18, 21, and 28 for microbiota analysis through 16s rRNA sequencing using Illumina platform. All sequence data processing was performed using QIIME v. 1.9.1 software. NE challenge impaired FCR in the overall experimental period (d 1-41) by 5%, but SB and SBEO supplementation completely reverted this effect (P = 0.01). The effect of time point within each experimental group was evaluated. The inclusion of SB alone or in combination with EO contributed significantly to the establishment of the cecal microbiota, as shown by the diversity indexes. The community structure and abundance of the cecal microbiota were significantly different across the experimental groups and ages. The cecal microbiota in the four ages evaluated was dominated by bacteria belonging to the Firmicutes, followed by Bacteroidetes and Proteobacteria. The most abundant genera observed in the cecal microbiota were: Ruminococcus, Lactobacillus, and Bacteroides, followed by unclassified Ruminococcaceae and Clostridiales. In the unchallenged control birds, it was observed that Ruminococcus decreased (P = 0.006), whereas Bacteroides and Clostridiales increased (P ≤ 0.02) as the birds aged. In the challenged control group, however, it was also observed that the frequency of Coprococcus and Blautia decreased as birds aged (P ≤ 0.01) and, Clostridiales did not increase as birds ages in this group. Supplementation of SB, but not SBEO, increased the frequency of Lactobacillus (P = 0.01) on d 12 compared to d 18 and 28, and prevented the reduction in the frequency of Blautia as the birds aged. SB and SBEO improved growth performance of NE challenged broilers, partially due to their modulating effects on the cecal microbiota.

Keywords: Broiler chickens, Intestinal microbiota, Necrotic enteritis

Estimates on the significance of chronic E.coli infections of the reproductive tract in laying hens

Abstract ID: 535

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Good health in layer flocks is crucial to animal welfare, egg yield and production economy. Oviduct infections are the most frequent cause of non-outbreak related mortality in egg laying hens, often manifested in a chronic state. The chronicity and manifestations suggest that these infections are subclinical and thereby present for a substantial period of time and thus non-apparent to the farmer. This period is of high risk regarding both decreased welfare of the affected hens and of transmitting pathogenic bacteria to the other hens in the flock or with the eggs. *Escherichia coli* has previously been isolated from the oviduct of healthy hens in a flock with *E. coli* associated oviduct infections. However, currently it is not possible to estimate the level of transmission from chronically infected birds. Consequently, the impact of such infections on production parameters cannot be estimated either.
The aim of the present study was to estimate the losses (e.g. mortality, reduction in egg production) of chronic *E. coli* infections using an experimental model of salpingitis. Eighty-four 26 week old Bovans Brown layers were divided into three groups; two groups were inoculated into the oviduct with two different *E. coli* strains isolated from clinical cases of chronic salpingitis. The third group served as control and received sterile media. After infection the infection groups showed a pronounced egg-drop (50 to 60% below the control group) and egg yield stayed significantly lower four weeks post infection. The egg loss during the six week long observation period was 30% plus a mortality rate around 20% for the infection groups, depending on the *E. coli* strain. Furthermore, the *E. coli* inoculum strains were detected in the egg contents up to 7 days post infection (17-50%), whereas contents from eggs from the control group were sterile. After the observation period the hens were necropsied. The severity of the observed pathology was strain dependent with chronic salpingitis and/or peritonitis present in 29 or 64% of the hens. In conclusion, experimental chronic infections in the reproductive tract were successfully established for the first time. The significance of chronic salpingitis on the egg yield was demonstrated and losses estimated. Furthermore, the detection of the inoculum in the egg content underlines the risk of transmission of pathogenic *E. coli* via eggs.

Keywords: Chronic salpingitis, E. coli, Egg loss, Layers

Effects of various housing conditions and genotypes on health and performance of tom turkeys

Abstract ID: 369

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The primary objective of genetic selection in turkeys has focused on traits that improve profitability such as body weight, feed conversion or breast meat yield resulting in heavy turkeys with more health problems, for example decreased locomotor activity. The provision of a roofed veranda or access to a free range might enhance walking ability and can be a way to promote the natural behaviour. This study investigated the effects of three different turkey genotypes (G1 to G3) differing in their final body weights and two housing conditions (barn as control, barn plus veranda and free range) on health and performance. In two trials, 1,352 one day-old male turkeys were randomly allocated to 12 littered floor pens (each 18 m^2). Six groups each were kept without or with veranda (12 m^2) and free range (240 m^2). Eight pens each contained 54 males (G1; G2, heavy turkeys for further processing) and four pens 61 males (G3, lighter genotype). Turkeys were fitted individually with two transponders and kept for 20 weeks. The use of veranda and free range (OA) as well as health and performance data (mortality, leg posture, walking ability, and body weight) were recorded and statistically analysed per individual tom using the GLM procedure of SAS®. Significant means were separated using Tukey-Test. Mean time per day in OA were significantly lower for strain G1 (13.4 hrs/day) compared to G2 (16.5 hrs/day) and G3 (15.9 hrs/day, $P<0.05$). G3 stayed 0.96 hrs in the barn and 1.07 hrs in OA per visit (in comparison: G2: 1.12 hrs vs. 1.19 hrs, G1: 1.8 hrs vs. 1.15 hrs, respectively). Walking ability and leg posture were significantly influenced by trial and genotype ($P<0.001$). The lightest genotype G3 showed the best leg posture and walking ability, following by G2 and G1. Mortality varied between trials and was lowest in G3 compared to G1 and G2. The OA reduced the mortality in heavier genotypes G1 and G2. The results show that the lightest genotype G3 seems best suited among the tested genotypes for alternative housing systems such as a veranda or free range. Furthermore, the results indicate that the access to an outside area can also improve the health and welfare of heavier common turkey genotypes in this study.

Keywords: Different genotypes, Free range, Roofed veranda, Turkeys

Expressional analysis of few immune relavant genes and bio-molecule at nasal mucosal surface against bacterial pathogen Avibacterium paragallinarum

Abstract ID: 305

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Avibacterium paragallinarum is the causative agent of infectious coryza (IC), mainly affects nasal mucosal surface and its associated structure of chicken. The disease involves poultry populations globally (directly or insidiously). The disease is paltry known to inflict other avian species; however some reports reflects involvement of pet and ornamental birds. International scenario reveals large numbers of vaccine failure owing to emergence of new pathotype(s), however our academic surmise additionally suggest failure in understanding of host mediated disease interference/susceptibility mechanism at gene level. In the present study, we attempted investigation on immuno-protective mechanism of chicken and Japanese quail at an early age against bacterial pathogen *Avibacterium paragallinarum*following intranasal inoculation (1.8×10^8 CFU/ml) in both species. Requisite sample i.e. nasal turbinates were taken at 3, 5, 12 day post exposure for qPCR and *in-situ* hybridization (ISH) technique to elucidate the temporal immune response regulations as well as tissue/site specific expressional labelling of biomolecule (COX-2 expression). Gene expression studies revealed biphasic up-regulation of IL1-β, explaining second peremptory protective response against infection in either species towards later stages (i.e. 12 DPI) with conjoint and contiguous effort by IL-4 up-regulation, an immunological directive that point towards Th-2 like response at nasal mucosal surface with prominent expression in chicken. Through ISH techniques, COX-2, a central inflammatory molecule which normally coincides with IL1-β expression corresponds to a protective response were mainly seen labelling respiratory epithelial cells of nasal septum and infraorbital sinuses in case of chicken and sensory epithelial cells of posterior turbinates in case of Japanese quail at both nuclear and cytoplasmic level, unlikely haddepressing expression towards later time point i.e. 12 day post infection as contrary to IL-1-β resurgence. Therefore, this study displayed tangible variations and putatively indicates towards complex intricate pathways getting involved in handling infections by either of the two avian species.

Keywords: Chicken, Gene expression, In-situ Hybridization, Infectious coryza, Japanese quail

Field trials and experiences with fluralaner against poultry red mite in the UK

Abstract ID: 115

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Poultry red mites (*Dermanyssus gallinae*) are the primary mite threat in Europe. These pests can cause anemia, increase mortality and disease susceptibility while eroding production. Parameters like feed efficiency, egg production, egg quality and weight gain. Current control methods lack sufficient efficacy to keep mite infestations under satisfactory control at farm level. Exzolt is a new and unique product designed to combat poultry red mite, Dermanyssus gallinae. The active ingredient is fluralaner, a new isoxazoline compound – a substituted benzamine derivative. It can be easily administered through the drinking water to layer and breeding chickens. Laboratory and pre-registration trials have demonstrated over 99 % killing rate and very fast efficacy. It can be used during not only during the rear but also during the laying period as well due to its zero days egg withdrawal period claim. The product offered a new opportunity to achieve control of PRM. The trials were designed to determine its efficacy under field conditions, and the impact on key production parameters such as age of production onset, egg production, egg quality and egg weight. We have chosen three farms for the trials with history of high PRM infestation level and failure of other products to reduce the challenge level satisfactorily. We monitored the red mite infestation level before and after the treatment using traps and software form AviVet. The product was administered via water at the recommended dose level of 0.5 mg per kg in two doses 7 days apart. The treatment had no adverse effect on water and food consumption and production parameters. Red mite infestation level dropped within 2- 3 days to a very low level after the first dose of treatment, and remained very low after the second treatment for 3 – 6 months. The treated flocks have become more active, their combs brighter, started laying eggs earlier and their egg production improved by 3-6 percent. Producers also reported fewer second class eggs, improvement in egg shell colour and egg weight. Considering the increased production rate and improved FCR the estimated ROI is 2.5:1. No regrowth of mite populations was observed in any of the treated farms to date and no adverse effects associated with the treatment have been reported.

Keywords: Dermanyssus gallinae, Fluralaner, Isoxazoline, Mite infestation, Poultry red mite

Fungal fermented feed ingredient for improved gut health in broiler chickens

Abstract ID: 584

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Fermentation is an old technique to preserve and enhance food functionality. Several bioactive compounds in feed and bacterial material are produced during fermentation. Those compounds may be beneficial for gut health, such as bioactive peptides, prebiotics, peptidoglycans, glucans, enzymes and vitamins. Mushrooms are well-reported and specific species show immunomodulatory properties and are used in curing of all types of diseases. Those mushrooms can also be used in solid-state fermentations to ferment feed materials.
In our research, we study the *in vitro* and *in vivo* efficacy of a solid-state fermented rye by the fungus *Agaricus subrufescens* (almond mushroom). Exposure of a chicken macrophage cell line (HD-11) to the fermented rye showed an increase in nitric oxide production in a dose dependent way after 24 hours *in vitro*. The production of NO is one of the effector pathways used by macrophages to defend the host against invading pathogens. Furthermore, the fermented rye demonstrated *in vitro* a high adhesion capacity to several Gram-negative pathogens, which may contribute to reduce the risk of colonization in the intestine. Moreover, the signs of dysbacteriosis were significantly reduced in a broiler study using an objective scoring method on the proximal and distal intestinal tract. Further *in vivo* evaluations in broilers are ongoing studying effects on performance, immunity and microbiota.
The results indicate that rye fermented with *Agaricus subrufescens* may be beneficial for gut health and control of bacteria in broiler chickens.

Keywords: Feed fermentation, Gut health, Immunity, Mushroom

Gel vs spray administration for infectious bronchitis virus: which is better?

Abstract ID: 300

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Infectious Bronchitis Virus (IBV) is an economically important respiratory disease seen throughout the poultry industry. Multiple methods of control have been employed with live vaccination being primary. Often live vaccination occurs in the hatchery on day of hatch in an attempt to prime the chicks immune system in a setting where application variables can be controlled. Novel administration methods are also being evaluated to optimize vaccine coverage and minimize stress. In this study, two types of gel products were compared to conventional aqueous spray. Massachusetts-type vaccine was mixed with water, a spray-type gel, and a drop-type gel and applied to groups of 100 chicks. In all groups the water used to dilute the vaccine, mix the gel powder, or dilute the gel diluent to the appropriate concentration was chilled to 4C prior to use. Chick activity was recorded for 5 minutes, and external and rectal body temperatures were recorded every 5 minutes for 1 hour after vaccination. The 3 groups of vaccinates and an unvaccinated control group were then reared in separate colony houses. Choanal cleft swabs of the vaccinates were taken on days 3, 5, 7, 10, and 14 to assess vaccine coverage based on real time-PCR. A random sample of birds from each group were taken 28 days post vaccination, challenged with a pathogenic Mass-type IBV, and placed in isolator units. Five days post challenge, the birds were observed for respiratory signs, swabbed for real-time PCR, and tracheas were taken for ciliostasis scoring. Chicks vaccinated with a spray-type gel experienced more severe temperature fluctuations than chicks vaccinated via drop-type gel or aqueous spray, though all chicks experienced a drop in rectal temperature compared to pre-vaccination. Aqueous spray vaccination produced higher uptake and replication after vaccination and better post challenge protection than either gel types. This data suggests that application of live IB vaccine via gel has no benefit over the traditional method of aqueous spray, and that using chilled water to prepare gel diluents can negatively impact chick temperature post vaccination.

Keywords: Gel application, Infectious bronchitis virus, Spray application

Identification of faecal protein biomarkers for intestinal health in broilers

Abstract ID: 413

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Gut health is crucial for performance, health and welfare of broilers. Next to classical pathogen-induced (e.g. coccidia and bacteria such as *Clostridium perfringens*) intestinal pathologies, more subtle digestive disorders are an increasing problem. These are often characterized by intestinal inflammation and villus shortening, which affect performance. Dietary stressors and/or subclinical enteric infections may be the underlying causes. There is a great need for reliable, rapid and preferably non-invasive biomarkers to diagnose these types of subclinical gut health disturbances.
Day-old broilers were randomly assigned to two groups, ie. control and challenged. From D12, all animals were fed a wheat (57.5%) based diet supplemented with 5% rye. From D12 to D18, animals from the challenge group received a cocktail of antibiotics. Hereafter, a bacterial cocktail that included *E. coli* and *C. perfringens* was given daily by oral gavage from D18 till D20. On D19, the animals were administered a coccidial challenge consisting of *Eimeria acervulina* and *E. maxima.* On D26 and D28, birds were euthanized and macroscopically scored for gut appearance and coccidial lesions. A sample of duodenum was fixed in formalin for measurement of histological parameters, i.e. villus length, crypt depth and infiltration of T-lymphocytes. Proteomics analysis was performed on colonic content via high performance liquid chromatography-mass spectrometry.
A significant difference between treatments was observed for both macroscopic gut appearance and total coccidiosis score at D26 (p<0.001). Significantly shorter villi, deeper crypts and a lower villus-to-crypt ratio were observed in duodenal sections of the challenged birds at D26 and D28 (p<0.001). Levels of CD_3^+ T-cells showed a significant increase at both D26 and D28 (p<0.001) in challenged birds. Proteomics resulted in identification of several proteins that correlated significantly with (decreased) villus length, mucosal T-lymphocyte density and/or the macroscopic gut appearance score. According to their function, the proteins could be categorized as markers for inflammation, plasma leakage, tissue destruction, epithelial cell death and tight-junction damage.

Keywords: Broiler, Faecal biomarker, In vivo model, Intestinal health

Impact of the Poultry Red Mite, Dermanyssus gallinae on the European poultry production systems

Abstract ID: 396

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Dermanyssus gallinae, also known as the Poultry Red Mite (PRM) is the most debilitating blood-feeding arthropod attacking birds and mammals. It costs circa 230 million euros per year to the European poultry industry and has a prevalence around 80% in many European countries. Since the first paper published 90 years ago on Dermanyssus we have seen over 85 countries publishing on such pest with many papers targeting control methods. As in other pest arthropods, resistance against some pesticides (esp. some of the authorized acaricides) were recorded in some PRM populations. Besides, due to the specific biology of this mite (which spends almost its whole life in the environment at a distance from the host), the efficiency of most sprayed substances is insufficient due to the product not reaching the largest part of the PRM population. Many countries have worked on approaches linked to their national legislation and what products were available and legalised or depending of the research priorities their national funding bodies were interested to support. Such national approach created fragmented data and a lack of potential replication and comparability between countries. Fortunately, since 2014 a new EU initiative from the COST scheme under the COREMI FA1404 Cost Action (www.coremi.eu) has developed a strong network (28 countries and over 300 members) to work together. In recent years the research focus has been towards new control methods including organic/biological approaches, antigen candidates for potential vaccines or understanding the vector capacity, reproduction or detoxification of *Dermanyssus gallinae* and many more. Such collaborative work has created new knowledge on such pest, increasing co-authorship of scientific publications between the COREMI members.
The authors would like to thank the European Cooperation in Science and Technology (COST Action (FA1404 – COREMI)*"Improving current understanding and research for sustainable control of the poultry red mite* Dermanyssus gallinae").

Keywords: Control methods, Dermanyssus gallinae, One health, Poultry Red mite, Poultry health

Influence of alternative husbandry systems on post-mortem findings and prevalence of important bacteria and parasites in layers - determined from end of rearing until slaughter

Abstract ID: 318

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In the present study 66 layer flocks housed in different alternative husbandry systems, barn system/conventional free range system/organic system, were monitored from placement of birds on the farm until slaughter to evaluate a possible influence on the occurrence of selected non-infectious as well as infectious diseases. Post-mortems were performed and the occurrence of extraintestinal *E. coli* and *G. anatis* was investigated. No specific post-mortem findings were noticed in pullets. Non-infectious syndromes were mainly found in layers independent from the husbandry system at the end of production, predominantly plumage damage, keel bone deformations, foot pad lesions and cloacal prolapse. Most prominent post-mortem findings in layers were reproductive tract lesions and the presence of intestinal helminths. From each and approximately 65% of the flocks *E. coli* and *G. anatis* were isolated, respectively. Therefore, the majority of reproductive tract lesions could be associated with *E. coli* alone or *G. anatis* co-infections. No significant differences were seen in regard to the housing system, but the prevalence of *G. anatis* increased with the age of birds. The prevalence of *Ascaridia galli* and *Heterakis gallinarum* was not influenced by the housing system, but significantly increased with age. Cestodes were present in 6 flocks. Histomonosis was detected twice. *Dermanyssus gallinae* was found in 5 pullet and 20 layer flocks. Additional investigations were performed on demand. Again, reproductive tract lesions were the most prominent post-mortem findings. In one flock each histomonosis and erysipelas was diagnosed, respectively. Severe affection by *D. gallinae* was found once. Necrotic enteritis was seen in two layer flocks. With the present study important information was revealed with consequences on health management in layer flocks kept in alternative housing systems. Main pathological lesions were a) bacterial infections of the reproductive tract, and b) infestation of helminths. This information is of importance regarding the limited options of antibiotic and anthelminthic treatments in layers flocks due to the limited availability of approved substances and technical constraints with regard to an efficient treatment.

Keywords: Alternative husbandry systems, Bacteria, Layers, Longitudinal study, Parasites

Insights on Avian Metapneumovirus Subtype B circulation in Europe

Abstract ID: 370

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Avian Metapneumovirus (aMPV) causes an upper respiratory tract infection in turkeys, leading to turkey rhinotracheitis, and in some other avian species is involved in the etiology of multi-factorial diseases such as swollen head syndrome of chickens. AMPV is classified as the type species of the genus *Metapneumovirus*, family Pneumoviridae, of which 4 subtypes (A, B, C and D) have been recognized based on the nucleotide sequence differences. Sequences of partial G gene (about 400 nucleotides) of European subtype B aMPV strains collected from 2014 to 2018, during routine molecular diagnostic activity, were analyzed. Nucleotide and amino acid sequences were edited and assembled using BioEdit software, then were aligned using Clustal W and compared with other sequences of subtype B aMPVs collected prior to that period or retrieved from GenBank. Sequences of most commonly used commercial vaccine strains were also included. Phylogenetic analysis, performed using PhyML method, demonstrated that aMPV subtype B has evolved in Europe from its first appearance without displaying a clear relation with the Country of detection. In recent times different field virus populations co-exist in Europe (maximum genetic distance 5,5%) suggesting that the protection achieved using current commercial vaccines should be evaluated. Along with field strains, a few vaccine-derived viruses are circulating. These strains were found mostly during outbreaks of respiratory disease, confirming that reversion to virulence of live vaccines is likely to occur. Insights offered by molecular characterization of aMPV will allow to better address the strategies to control aMPV infection in poultry.

Keywords: Avian metapneumovirus, Philogenetic analysis, Virus evolution

Interaction of Campylobacter jejuni with the gut barrier of broiler chickens: Camplyobacter jejuni has diametral effects on broiler gut health

Abstract ID: 323

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Campylobacter is an important food borne pathogen and despite its high prevalence in chickens, the strategy used by *Campylobacter* to colonize the chicken gut is hardly understood. In our recent studies we were able to demonstrate that an experimental *Campylobacter jejuni* infection has severe consequences on gut health. Although, caeca are the part of the gut with *Campylobacter* accumulation the infection does also influence certain functions within the small intestine. In addition, *Campylobacter* does not only reside in the gut, it can also disseminate to various internal organs, like liver and spleen. Colonization of internal organs might pose an additional risk to consumers via poultry consumption. Changes in the intestinal barrier function revealed an increased intestinal permeability, the down regulation of certain nutrient transporters and an impact on mucous production, altogether influencing nutrient uptake. In consequence, it was found that glucose uptake and amino acid availability were reduced following *C. jejuni* infection. We were able to demonstrate that *C. jejuni* can cross the intestinal epithelial barrier and facilitate the translocation of *Campylobacter* itself as well as of other enteric microorganisms such as *E. coli* to extra-intestinal organs of infected birds. Moreover, *C. jejuni* colonization could be associated with an alteration of the gut microbiota with infected birds having a significantly lower abundance of *E. coli* at different gut sites. On the contrary, the level of *Clostridium* spp. was higher in infected birds compared with birds from the negative controls, demonstrating that the infection of chickens with *C. jejuni* was associated with significant changes in the composition of the intestinal ecosystem. Altogether, these findings indicate that *Campylobacter* is not entirely a commensal and can be recognized as an important factor contributing to an impaired chicken gut health.

Keywords: Campylobacter jejuni, Chicken, Gut

Investigation of the effect of live attenuated Escherichia coli vaccination on experimentally induced salpingitis in layers

Abstract ID: 534

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Escherichia coli infection is a common production disease in the poultry production. In young birds the infection mainly causes airsacculitis, sepsis, and polyserositis, whereas in egg-laying hens oviduct infections are the primary manifestation. Oviduct infections contribute significantly to mortality, with regards to both the normal mortality and during outbreaks of colibacillosis in adult layer flocks. Vaccination is one strategy to minimize the losses due to *E. coli* infections. In young birds (broilers and pullets) vaccination with live vaccine (Poulvac® E. coli) can decrease the losses due to colibacillosis. Despite, not marketed nor documented and validated for effect on oviduct infections the vaccine is widely used in Danish layers. The aim of the present study was to investigate the effect of vaccination against *E. coli* salpingitis (Poulvac® E. coli, 2 and 14 weeks of age) in a recently established experimental model. Two groups of Bovans Brown layers (vaccinated and unvaccinated) were each divided into three groups. Each of the six groups was surgically inoculated with either a highly virulent *E. coli* strain (outbreak simulation), a moderately virulent *E. coli* strain (simulation of non-outbreak related salpingitis), or sterile media. Two days post infection (dpi) the vaccinated group receiving the outbreak strain contracted severe clinical symptoms and were for animal welfare reasons euthanized. The clinical symptoms in the unvaccinated group infected with the outbreak strain were slightly less severe and 60% of the hens were euthanized. The mortality rate in this group was significantly lower than in the vaccinated group. Ten dpi all remaining hens were killed and necropsy and bacteriological investigation were performed. No significant difference in the lesion scores was observed between vaccinated and unvaccinated birds. However, significant strain dependent differences were observed in lesion scores resulting in severe lesions in the vaccinated birds infected with the outbreak strain. Similar differences were observed regarding bacteriological re-isolation of the inoculum strain.
In conclusion, vaccination with live attenuated *E. coli* at two and 14 weeks of age does not protect against salpingitis in egg-laying hens under experimental conditions with both regards to outbreak associated *E. coli* salpingitis and non-outbreak related salpingitis. However long-term unspecific vaccination effects on causes of normal mortality and production parameters remain to be addressed.

Keywords: E. coli, Layers, Salpingitis, Vaccination

Monitoring after coccidiosis vaccination- sample collection and dynamics in different housing systems

Abstract ID: 272

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Coccidiosis vaccination in rearing pullets is gaining more interest as birds are reared on the ground or in aviary systems. Cycling of the live coccidiosis vaccines, an important and necessary part for immunity build up, is challenging as bird are reared in low density, alternative housing systems and dry environmental conditions. The aim of this study was to follow oocyst excretion in rearing pullets, housed in different environmental conditions after hatchery spray vaccination, using a standardized sampling method. Two Austrian pullet flocks were closely followed after coccidiosis vaccination in the same hatchery with two European coccidiosis vaccines containing the species *Eimeria (E.) maxima, E. mitis, E. acervulina, E. tenella* and *E. necatrix* and *E. brunetti.* Birds were moved to commercial farms where they were reared in aviary systems or on the ground. In the aviary system birds were housed on the wired floor cages for a period of 35 days, after that, pullets had access to the different levels and the floor. The ground reared pullets were reared in 2 different rearing farms: with a movement at the age of 35 days. In both rearing systems oocyst excretion was followed. Weekly fresh excreted droppings were collected during three consecutive days. Individual samples were investigated by the McMaster oocyst counting technique, followed by species identification. After individual investigation, the three samples of the same week, were pooled in the lab to one sample. On this sample the same techniques of oocyst counting and species identification were applied, using the same standardized method. Results of the individual samples showed for both farms a peak of oocyst excretion at the age of 14 days. After opening of the aviary system a second peak was seen at the age of 58 days. No clinical signs nor performance impact was associated with the OPG increases. As expected excretion was lower in the aviary housing system in comparison to the ground reared pullets. Oocyst excretion was variable over the three sampled days within one week, this resulted in differences of OPGs between these samples and the pooled samples. Pooled samples of three consecutive days resulted in higher OPG counts than the averages of the three individual samples. Less species were seen in the pooled samples. Sample collection on three consecutive days was worthwhile as it resulted in a clearer picture on oocyst excretion as well as on the different *Eimeria* species present.

Keywords: Coccidiosis, Coccidiosis vaccination, Oocyst excretion

New insights on application of IBV vaccines

Abstract ID: 502

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Infectious bronchitis virus (IBV) is, in spite of vaccination(s), one of the major causes of respiratory problems, poor egg production and mortality in chickens in many parts of the world. The vaccines have shown under experimental conditions that they are able to provide a high level of protection against a homologous challenge. This raises the question: why do we have outbreaks in vaccinated flocks?
One of the possible reasons of (partial) failure of the protection induced by vaccination(s) could be the timing and way of vaccine application. Under field conditions, IBV vaccines are usually not applied individually, but by mass application methods. IBV strains are non-enveloped virus which are much easier to inactivate than to keep them infectious. This is the same for the vaccines of IBV. This aspect of IBV is relevant as the mass-application methods have been developed or implemented for practical reasons (human comfort) and are not necessarily favorable for the take of the vaccine virus. This paper presents work showing that mass application of vaccines against IBV needs a high level of attention and care. This includes the latest new insights on the application of IBV vaccines and how to monitor the take of the vaccines.

Keywords: Application, IBV, Vaccination

Overview on the efficacy of paromomycine as a treatment for commercial turkeys experiencing outbreaks with Histomonas meleagridis

Abstract ID: 379

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Paromomycin was used for treatment of turkeys that experienced outbreaks with *Histomonas meleagridis* in commercial turkey farms in Austria. An overview will be given on 9 outbreaks which occurred in 5 different turkey farms (3 commercial and 2 organic farms). Outbreaks appeared in the period of 2015-2017 in turkey flocks aged between 2 weeks and 13 weeks. In 3 farms different outbreaks were experienced over the years. In the farms treatment with paromomycin was initiated in the drinking water at a dosage of 12.5 mg/kg bodyweight (BW) for 13-14 days. Mortality was variable and ranged between 10-50%. It was observed that early diagnostic, and consecutively early treatment, were the main points for a successful treatment. Initiating treatment early after detection of the first dead bird in the farm, had the highest success rate. This was possible when farmers were motivated to perform necropsy on every dead turkey on the farm, even before any clinical signs were observed. In farms where the disease was only diagnosed when turkeys were already diseased during several days, success rate of the product was poorer. This can be explained by the low bioavailability of the product in combination with the fast spreading of the parasite to the liver. In farms where both males and females were affected during an outbreak, treatment of females appeared more successful than treatment of the turkey toms. It can be concluded that early diagnostic and early start of paromomycin at a dose rate of 12.5 mg/kg BW appeared successful in controlling *Histomonas meleagridis* in field conditions. For this reason, a sound monitoring system using available diagnosis tools such as PCR is crucial, in particular on problem farms.

Keywords: Blackhead, Histomonas meleagridis, Paromomycin, Treatment, Turkeys

Phytogenics improve resilience against coccidiosis and secondary intestinal infections in broilers

Abstract ID: 508

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Resilience of birds against intestinal infections is key to maintain production performance under challenge conditions. Phytogenics (bioactive secondary plant compounds) comprise a large group of natural compounds with a broad spectrum of activities: anti-oxidant and anti-inflammatory effects, enhanced immune response and improved nutrient digestibility. Also, direct effects on pathogens are described like antimicrobial and anticoccidial effects. Use of phytogenics are a base to reduce pathogenicity of the intestinal microbiota and maintain or optimize the intestinal barrier as first line of defense. By blending and standardization of such actives, their different modes of action can be combined into effective additives that improve the resilience of birds against intestinal infections.

The effect of a blend of quillaja saponins, and selected essential oils from the myrtaceae plant family was tested in male Cobb 500 broilers, using a combined *Eimeria* and *Clostridium perfringens* (Cpf) infection in a 28-day cage study (8 birds per cage). This challenge model was chosen as secondary bacterial infections strongly aggravate the impact of a cocccidiosis infection on production performance. All birds were inoculated on day 14 with 5,000 *E. maxima* oocysts and part of the birds at day 19, 20 and 21 with Cpf. Production performance was measured during the 28-day trial period. Necrotic enteritis lesions were scored in three birds per cage on day 21 and oocysts excretion was determined per cage on the same day. Treatments were 'not Cpf infected and not treated' (NINT), 'Cpf infected not treated' (INT), 'Cpf infected and treated with 50 g/t BMD' (IT) and 'Cpf infected and treated with the phytogenic blend' (IPB). Measurements were replicated 8 times.

BWG was reduced by 160 g and FCR increased by 0.305 due to the Cpf infection. INT showed high mortality. IPB resulted in a numerical reduction in OPG, presumably due to less favourable intestinal conditions for Eimeria reproduction. Addition of BMD or the blend of phytogenic actives resulted in a similar production performance as in NINT. These results were confirmed in a 35-day floor pen study with the same layout. It was concluded that resilience of birds against intestinal pathogens was improved by feeding the specific phytogenic blend.

Keywords: Broiler, Coccidiosis, OPG, Performance, Saponins

Prevalence of Gallibacterium anatis isolated from layer poultry farms in Croatia

Abstract ID: 623

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Objectives: *Gallibacterium anatis* is a part of normal microflora of lower reproductive tract and upper respiratory tract in layers. Recently, it has been considered one of the most prevalent causative agents of different reproductive and respiratory disorders in laying hens beside avian pathogenic *E. coli* (APEC). Both bacteria cause similar lesions, such as peritonitis, folliculitis and salpingitis, which makes it difficult to differentiate the cause. Additionally, they also often appear as coinfections, especially in immunocompromised poultry. Alongside those lesions, production decrease and high mortaEditlity rates are often reported which makes it a serious problem on poultry farms. The purpose of this study was to determine prevalence of *G. anatis* on layer poultry farms in order to assess the current situation in Croatia and investigate the possible solutions to *G. anatis* infection.

Materials and Methods: During this epizootiological survey, poultry carcasses as well as pharyngeal and cloacal swabs from live animals were brought from seven layer poultry farms to the Department where necropsy was done. During examination, swab samples were taken from pharynx, trachea, lungs, liver, oviduct, follicles and bone marrow. Samples were then plated on 5% sheep blood agar and incubated aerobically at 37°C for 24 hours. Identification was done by gram staining, biochemical tests and MALDI-TOF spectrometry method. Afterwards, they were stored in BHI broth at -80°C for further analysis.

Results and Conclusion: All of the farms included in epizootiological survey were positive for *G. anatis.* Most of the isolates originated from pharynx and cloaca. In some cases coinfection with *E. coli*, *Hafnia alvei, Mycoplasma sp.* or IB virus was present, so we assume *G. anatis* is an opportunistic pathogen which caused the infection in immunocompromised chickens. Due to often reported quickly acquired antimicrobial resistance after antibiotic therapy, use of autogenous vaccine combined with adjustment of the immunoprophylaxis program is recommended.

Keywords: Gallibacterium anatis, Layers, Peritonitis, Poultry

Prevention of MG-MS infection using live vaccines

Abstract ID: 24

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With the rapid growth of poultry production world-wide, there has been concentration of large numbers of birds into small areas, leading to increased risk of exposure to pathogenic mycoplasmas. In some areas, poultry production is so concentrated that from an epidemiological point of view, it is almost like a very large multi-age farm. Mycoplasmosis is still one of the costliest disease for the modern poultry industry. The infected birds/flocks are carriers for the life. *M. gallisepticum* (MG) is causing respiratory syndromes, egg-drops and increased mortality during production, embryonic death during hatch. The MG is causing severe post vaccination reactions, serious respiratory syndromes, increased mortality, medication costs and increased condemnation at slaughter with poor weight gain and FCR in the vertically infected broilers. *M. synoviae* (MS) is traditionally considered the second most important avian mycoplasma species. It has been associated with respiratory disease and subsequent condemnations due to airsacculitis in broilers and peritonitis and mortality in commercial layer hens, although subclinical infections of the respiratory tract seem predominant. MS is also known to cause synovitis in chickens. Since the year 2000 a novel eggshell apex abnormality (EAA) has been increasingly found. Recent studies have shown that MS, could be a significant factor in initiating E. coli peritonitis, a major cause of mortality in layers. The control of the disease caused by MG and MS can be achieved by three methods: by eradication, by antibiotic treatment and by application of vaccines. Antibiotic treatments reduce mycoplasmas population, clinical signs, losses and decrease contamination of eggs and embryos, but increase resistance to antibiotics and residues in poultry products. The first commercially available Mycoplasma vaccines were oil-emulsion bacterins. Bacterins protect against egg production losses generating plenty of humoral antibody, but provide little protection against colonization by field strains of mycoplasma in the respiratory tract. Major disadvantages of bacterins are the need for 2 doses for optimal protection and the cost of administration. Significant advances in the control of mycoplasmosis have been realised since the introduction of third generation attenuated live *M. gallisepticum* and *M. synoviae* vaccines. These third generation mycoplasma vaccines were specifically attenuated (selected for temperature sensitivity (ts) and selected to combat the effects of wild mycoplasma challenge. Field experiences utilizing live vaccines have been very successful in commercial layers and broiler breeders. These experiences suggest that live vaccines may be viable tools for the eradication of mycoplasma infection.

Keywords: Live mycoplasma vaccines, MG-MS infection, Mycoplasma infection

Released-active drugs enhance poultry production performance

Abstract ID: 132

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Modern healthcare in poultry production suffers from common problems that can not be dissolved with the help of existing therapeutic approaches. Similarly to the human medicine, veterinary pharmacology has an acute necessity in safe and efficient drug products. Recent fundamental development of the released-active drugs (the active pharmaceutical ingredient produced by gradual decrease of drug substance's concentration) has established its efficacy in the human medicine and is now being transferred into the veterinary field. Applying the released-activity technology to the antibodies raised to physiologically active molecules provides us with the specificity of action together with the high profile of safety (both for the animals and the poultry products consumers). One of the major challenges in poultry production, viral infections, could be met with antiviral immunity boosting. The complex drug Polyferon, containing the released-active affinity purified antibodies to gamma-interferon, CD4 and beta-subunit of the insulin receptor, has shown the ability not only to increase the broilers production performance parameters but also to stimulate the vaccination efficiency. Previously published laboratory studies have shown that the complex drug and its separate components improve the resistance of the organism against several viral strains (including avian influenza) and partake in the energy homeostasis regulation. Field trials of the complex drug on poultry factory in Russia (n=27500 in placebo group and n=27150 in Polyferon group) also provided the evidence of the broilers livability (95.4% vs 97.1%) and body weight (2011.16±13.94 g vs 2119.64±24.30 g on the day 35) statistically significant increase. Feed conversion was shown to be much higher in the Polyferon-treated group – 1.68kg per 1kg of body weight gain vs 1.81 in the placebo group. Together these data illustrate the significant increase of economic effectiveness of poultry production with the use of the drug. Polyferon is the first marketed released-active drug for poultry production, but the forthcoming research results suggest that new components could be efficiently used as a separate drug or be added to the Polyferon composition in order to stimulate antibacterial immunity of the livestock and to further improve the production performance. Immunity stimulation is not the only direction of the released-active drugs application. One of the latest experimental studies shows the promising results of the released-active treatment for the hatching eggs, another study compound is aimed at the laying hens performance improvement.

Keywords: Immunostimulation, Polyferon, Poultry performance, Released-activity

Similar longitudinal development of cecal microbiota diversity within four broiler houses at two different farms

Abstract ID: 581

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Gut microbiota influences health and production performance of broiler chickens. However, there is a lack of fundamental knowledge of microbiota dynamics within and between broiler flocks and farms. Therefore, for four broiler houses at two different farms the longitudinal development of cecal microbiota diversity was analysed using alpha and beta diversity metrics and multivariate analyses.
The two farms had a different feed supplier and management, but the broiler flocks in the two houses within the same farm were from the same breed (Ross 308), hatchery and parent flock and received the same feed and management. Coccidiostatic drugs were standardly applied in the feed in all flocks. During one production round cecal content was collected at the chicks' day of arrival and on days 2, 7, 14, 21, 28, and 35. For each time point, cecal samples from nine birds from each of the two poultry houses at each farm were analysed, resulting in a total of 324 analysed samples . All four flocks showed excellent production performance, and no antibiotic treatments were applied. The diversity of the cecal microbiota was assessed by 16S ribosomal RNA gene amplicon Illumina HiSeq sequencing.
From around the second week of age onwards, the alfa diversity such as the number of observed OTUs, the richness, and evenness of the cecal microbiota seemed to stabilize at both farms. According to the beta diversity based on unweighted UniFrac distances, the age of the broilers most strongly affected cecal microbiota diversity. Furthermore, unweighted UniFrac distances were slightly higher than the corresponding weighted UniFrac distances between the two farms. This suggested that the abundant taxa of the two farms are phylogenetically related. The two broiler houses within the same farm showed no difference in cecal microbiota diversity.
In conclusion, based on the alpha diversity the cecal microbiota development of the two different farms was comparable, despite the different feed supplier and management. In all broiler houses, the age of the broilers within the production round had a similarly large effect on gut microbiota dynamics.

Keywords: 16S rRNA, Broilers, Field study, Microbiota

Supplementation of Diamond V fermentation metabolites in feed or drinking water to reduce stress in broilers

Abstract ID: 289

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Reducing stress is an important goal in animal production and previous research has demonstrated the ability of fermentation metabolites products (FM) to reduce the stress response of broilers during a heat stress. Three studies were conducted to evaluate the effects of adding the fermentation metabolites in the feed (FM1) or in the water (FM2) to broilers exposed to acute stress in addition to normal rearing stressors. Treatments included: control non-stressed (CNS), control stressed (CS), the FM1 added to the feed at 1.25 kg/MT, and the FM2 added to water at 160mL/100L. All treatments except CNS received the following stressors: Live cocci-vaccination on Day 1, reared on reused litter, heat stress (35^{o}C) and feed/water withdrawal for 12h on Day 18. On Day 19, blood was collected and analyzed for plasma corticosterone (CORT) and heterophil/lymphocyte ratios (HL) from 60 birds/ treatment. On Day 41, CORT and HL were also determined in 24 birds/ treatment and bilateral bone traits were measured in 60 birds/treatment to assess physical asymmetry (ASYM). In all three experiments, CNS birds had lower CORT (average 50% lower) and HL (average 42% lower, $P < 0.05$) than CS birds (average per trial 1130 pg/mL and 0.17) on Day 19. Results for all three experiments showed that birds provided FM1 or FM2 had significantly lower CORT (average 45% lower) and HL (average 27% lower) than CS ($P < 0.05$) on Day 19. Also in all 3 experiments, birds supplemented with FM1 or FM2 had significantly lower CORT (average 61% lower), HL (average 31% lower), and ASYM (average 18% lower) than both CNS (average per trial 2456 pg/mL, 0.24, and 2.49mm) and CS (average per trial 2421 pg/mL, 0.23, and 2.48mm) on Day 41 ($P < 0.05$). Supplementing broiler chickens with FM1 or FM2 improved animal welfare measured by reduced stress indicators after acute stress or the effects of normal rearing stress in both experiments.

Keywords: Broilers, Fermentation metabolites, Heat, Stress

The effects of treatment with fluralaner on poultry red mite infestation and on production of laying hens housed in enriched cages and aviaries

Abstract ID: 196

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Dermanyssus gallinae, the poultry red mite (PRM), is an important cause of welfare and health problems in laying hens, yet the control remains a challenge. Recently, a new systemic acaricide was approved in the European Union: fluralaner. A field study was performed to evaluate the effects of treatment with fluralaner on PRM infestation level and on egg production of laying hens kept in different housing systems. 31 000 hens of 2 different breeds were housed under controlled conditions in 12 climate and light independent compartments equipped with enriched cages and 2 different types of aviaries. At the age of 50 weeks, fluralaner was administered twice 7 days apart at a dose rate of 0.5 mg/kg BW through the drinking water. PRM were monitored with the Mite Monitoring System (MMS), which is a visual scoring system and by counting of PRM in 2 types of traps. The laying percentage was recorded daily for each compartment from 4 weeks before until 6 weeks after the first treatment. For evaluation, obtained data were compared to the production standard of the breeds.Before treatment, all compartments except one were infested with PRM. The infestation levels varied from light to very high. After the treatment, no mites were detected in 11 of the 12 compartments. In the remaining compartment, the percentage of mite reduction was more than 99.99%. In the 4-week period post-treatment, the laying percentage rose in 10 of the 12 compartments, on average with 2.79%. In conclusion, fluralaner administered through drinking water was found effective in reducing the number of PRM in infested laying hen houses with at least 99.99%, irrespective of breed and housing system. This was accompanied with a sharp increase in egg production. Under conditions of PRM infestation, treatment with fluralaner can therefore mean an important aid to welfare of laying hens and profitability of laying farms.

Keywords: Fluralaner, Poultry red mite, Production

Transcriptomics approaches in enteric inflammation models for the assessment of dietary supplements

Abstract ID: 311

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It is well known that intestinal inflammation and leaky gut are hallmarks of poor gut health, resulting ultimately in reduced performance. Recent studies demonstrated that the better performance of animals fed antibiotic growth promoters (AGPs) is also driven by reduction of the innate inflammatory response in the intestinal tract. Now that the use of AGPs is discouraged in animal production, it is important to identify dietary compounds that are as effective as AGPs. Until recently, few models of enteric inflammation were developed in poultry to properly evaluate alternatives to AGPs. Therefore, a first feeding trial, 2x3 factorial design, was conducted in broiler chickens in which the interaction of heat stress with three experimental diets was evaluated (16 birds/diet). Broilers were fed from hatch to d28, i) corn-soy-based feed (control group), ii) deoxynivalenol-contaminated feed (DON group, 15 mg DON/kg), and iii) rye-wheat-barley-based feed (RWB group, 31-22-10%). At day 28, half of the birds in each group were kept at 36°C for 10 hours to induce heat stress, and then intestinal samples were collected from all the birds. The expression of 25 genes associated with inflammation was evaluated via qPCR in the duodenum, jejunum and ileum. Multivariate analysis and hierarchical clustering showed that the nutritional challenges (DON and RWB) modulated the overall gene expression, with stronger changes when combined with heat stress. The top genes consistently affected all along the intestinal tract were HSP70, IL-6, IL-8, TLR2, FABP2 (-16 to +7 fold change). A second feeding trial was conducted using an overdose of a coccidial vaccine (25x) to induce alterations in the ceca of 35 day-old broiler chickens. Six days post infection, the ceca of 10 birds either challenged or unchallenged were sampled for running RNA sequencing analysis (30+30 Mio reads, 2*75 bp). Reads were mapped with reference genome Galgal5, quantified with HTSeq count and normalized in edgeR. 563 genes were found differentially expressed (-13 to +58 fold change), and enrichment pathways analysis showed that up-regulated genes were associated with cytokine signaling and innate immune system whereas down-regulated genes were related to lipid metabolism. In conclusion, enteric inflammation models are useful *in vivo* tools for assessing anti-inflammatory compounds, and measurement of the gene expression via transcriptomics approaches is a robust method to characterize this inflammation.

Keywords: Gut barrier, Inflammation model, QPCR, RNA sequencing, Transcriptomics

Treatments of gastrointestinal problems in broiler breeders and pullets after coccidiosis vaccination – a retrospective study

Abstract ID: 282

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Coccidiosis control in breeders on a worldwide level is mainly achieved by vaccination. Also in rearing pullets housed in alternative systems the interest of vaccination is increasing. Inadequate immunity build up against all the *Eimeria* species included in the applied coccidiosis vaccines might lead to coccidiosis outbreaks during rearing or even during production. Coccidiosis infections may lead also to secondary gastrointestinal problems that require antibiotic treatment. In a retrospective study treatment interventions for coccidiosis and/or secondary gastrointestinal problems of a European breeder and rearing pullet integration were compared. In both cases treatment interventions after vaccination with a live attenuated vaccine containing the species *Eimeria (E.) maxima (2x), E. mitis, E. acervulina, E. tenella, E. praecox, E. necatrix and E. brunetti (Vaccine 1)* were compared with treatments after application of 2 live attenuated vaccines containing the species *E. maxima, E. mitis, E. acervulina, E. tenella (Vaccine 2)* and *E. necatrix* and *E. brunetti (Vaccine 3).* Vaccination of the breeders was applied on farm (for Vaccines 2 and 3 at the same day or split application); while for rearing pullets vaccines were applied in the hatchery. 379 broiler breeder flocks and 182 pullet flocks vaccinated with vaccine 1 and 107 flocks (as well for broiler breeders as pullets) vaccinated with vaccines 2+3 were included in the study. For both integrations the number of treatment interventions were counted, starting 5 weeks after the last vaccination when immunity should be established. For broiler breeders the number of treatment interventions decreased significantly (P=0.016) from 7% (vaccine 1) to 0.9% (vaccine 2+3). In rearing pullets a decrease from 17% (vaccine 1) to 3% (vaccine 2+3) was noted (P < 0,005). Beside improvement of management, different application methods (for breeders split application), a higher reproduction potential of the vaccines 2 and 3 might explain the results. Indeed, beside correct application, cycling of the different live *Eimeria* species included in the vaccines is an important and necessary part for immunity build up. The number of attenuations during development of live coccidiosis vaccines may lead to different reproduction potential properties of the available vaccines. High oocyst excretion after vaccination might contribute to a more solid immunity, leading to lower treatment interventions during rearing and production.

Keywords: Broiler breeders, Coccidiosis vaccination, Gastrointestinal problems, Rearing pullets, Treatment intervention

ORAL PRESENTATIONS

Poultry Housing and Management

Effect of time period between hatching and the onset of feeding on the physiology and growth of broiler chicks

Abstract ID: 434

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In commercial conditions, day-old chickens are subjected after hatching to a delay in their access to food and water until they arrive at the broiler house. The objectives of this study were to evaluate the effects of three transport times commonly used in Spain (2, 14, and 26 h) on the chick metabolism, development of digestive organs and performances during their first week of life. Thus, three experimental groups of 1,200 day-old chickens corresponding to every transport time were housed in our experimental farm in pens containing 100 birds each and provided with feed and water. Each treatment was replied in 12 pens.
Before housing, 12 birds per treatment were randomly chosen to analyze several metabolic indicators, and other 12 chicks per treatment were used to determine initial weight and proportions of yolk sac and digestive organs. Blood parameters were analyzed again 24 h and 48 h after housing. Liver glycogen and weight of yolk sac and digestive organs were measured 3 and 7 days after housing in 12 chicks per treatment. Mortality, weight gain, and feed consumption and conversion were determined at the same periods on a pen basis. All measurements were made at equal times after housing. Birds subjected to 26 h of fasting showed greater values for hematocrit, serum total proteins and β-hydroxybutyrate, but differences disappeared at 24 h after housing. Plasma glucose levels increased quickly in all groups after the onset of feeding and decreased thereafter. Initial yolk sac weight and percentage were lower in chicks fasted for 26 h, and in all groups were negligible after 3 days. The relative weight of intestine greatly increased from 0 to 3 days, being lower at 0 days in the 2 h group, without further differences among groups. Initial body weight was lower in birds fasted for 26 h, but they were able to eat more feed in the first week and to perform compensatory growth. At 7 days, no differences were found for feed conversion. Mortality rates were normal without significant differences among groups in any period.
In conclusion, a delay in access to food and water up to 26 h after hatching did not show a permanent harmful effect on bird's welfare and performances. Day-old chicks subjected to the longest fasting times showed slight negative effects on metabolic indicators and initial performances, but they were able to compensate them in their first 7 days of life.

Keywords: Broiler chick, Fasting, Performances, Physiology

Improving external egg quality in enriched cages

Abstract ID: 195

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A big challenge in managing hens in enriched cages is maintaining a high egg quality. European legislation, establishing welfare of laying hens, foresees a nest area in cages with adequate nest material. Nest design should focus on attracting the birds and can have a direct impact on outer eggshell quality. In enriched cages, hens in the nests are often only separated from the other hens by a plastic curtain at the back of the nest. Birds also experience direct light in the nest. The Experimental Poultry Centre (EPC) investigated the effect of providing a more darkened nest area on percentage of nest eggs and external egg quality. 6144 White and 6144 brown hens were kept in 4 climate independent compartments, each containing 2 rows with 4 levels each of enriched cages. Half of the groups were equipped with standard, half open nests and half of the groups with darkened nests. External egg quality was automatically recorded daily and the percentage of nest eggs was observed at 31, 36, 41, 46, 51 weeks of age. In both white and brown hens, the percentage nest eggs differed strongly between the groups with and without a darkened nest. In white hens, 78.7% of the eggs were laid in the darkened nest vs 52.6% of the eggs in the open nest at 31 weeks of age. In brown hens, the difference was even more explicit (90.6% in darkened nest vs 48.9% in open nest). Similar results were found at different ages. In brown hens, the percentage nest eggs did not have an impact on the dirtiness of the eggs resulting in an equal percentage of dirty eggs. In white hens, however, enclosing the nest resulted in 10% less dirty eggs. We can conclude that a small adjustment of the enriched cage design can have a large impact on the percentage of nest eggs in brown and white hens and influence the percentage of dirty eggs in white hens.

Keywords: Egg quality, Enriched cages, Nest area

Influence of the housing system on mating behavior in two broiler breeder hybrids

Abstract ID: 228

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Unlike layers, broiler breeders are commonly kept in floor housing systems without perches or aviary tiers. Several European countries now require perches for broiler breeders. Mating normally occurs on the litter so we hypothesized that hens might use perches or aviary tiers to avoid males and that males would show more courtship behavior to attract females. Relatively fast (Ross 308) and slow growing (Sasso) hybrids were kept separately in semi-detached barns with control pens (C) consisting of litter, raised slats, feeding systems for males and females, drinker line, and nestboxes.
Aviary pens (A) additionally included a four-tier aviary on the slats, and perch pens (P) included six wooden perches in an A-frame on the slats. Each treatment was replicated three times within the barn for each hybrid. The different elements of mating behavior were coded from video recordings when the birds were 35 and 40 weeks old. Hatching rates per pen were determined at the ages of 39 and 45 weeks. The number of mating acts was analyzed with generalized linear mixed models using the log-link function. The experimental unit was the pen nested in treatment and hybrid (confounded with the factor 'barn'). The treatment did not affect the frequency of matings ($F_{2,14} = 0.00$, $P = 0.99$) but Ross 308 mated less frequently at 40 weeks of age compared with 35 weeks of age (interaction hybrid x age: $F_{1,14} = 6.93$, $P = 0.02$, contrast Ross age 35 vs. Ross age 40: $t_{14} = 2.75$, $P = 0.02$). However, matings were more frequent on the slats in C and P than in A ($F_{2,13} = 6.37$, $P = 0.01$) and more frequent on the litter in Ross 308 ($F_{1,13} = 12.45$, $P = 0.004$). Thus males followed the location of the hens. Hens solicited a higher percentage of matings in P and A than in C ($P = 0.05$) without a change in male courtship behavior. The hatching rate per pen was correlated with the corresponding mating success rate but not with the treatment. We therefore conclude that perches and aviaries do not interfere with hatching rates.

Keywords: Aviary, Broiler breeder, Mating behavior, Perch

Innovative agroforestry models: chickens in olive orchards and geese in vineyards

Abstract ID: 530

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In modern agriculture, one of the most important problems, is the soil compaction, caused by overuse of machinery, intensive cropping and short crop rotations. The agroforestry practice, where animals are raising in plants integrated systems partly solve this problem.
The Italian hills have a high prevalence of orchards, vineyards and olive groves and the agroforestry application to these territories added a further value to the systems by enhancing local productions. The aim of this study was to evaluate the applicability of agroforestry systems in poultry production, with special focus on meat quality, environmental impact and profitability.
Two case studies were analyzed:
1) an olive grove intercropped with wild asparagus and grazed by chickens;
2) a vineyard grazed by geese.
In the first case study Naked neck (NN) chickens were reared in an olive groves until 81 days of age. Whereas, in the second one, *Romagnola* geese were reared in the 1 ha of vineyard for 180 days of age. At the end of both experimental trials 10 animal/group were slaughtered and the *Pectoralis major* muscle was dissected for the qualitative analyses.
In both cases the tree/vineyard productions (olive and grape) was not negatively affected by the introduction of the animals, however, many factors affect the success of the systems:
i) the choose of suitable animal genotypes characterized by a high rusticity and foraging behaviour;
ii) an appropriate animal density, in relation to the animal species: geese needed more space than chickens (50 vs 10 m^2/animal);
iii) the pasture rotation to allow grass recovery and the optimal use of the forage species, which, in turn, positively affected the meat quality, and finally
iiii) the space availability, which promoted the kinetic activity and the development of the drumstick muscle (higher meat/bone ratio) simultaneously reducing fat deposition and lipid content of meat.
One of the negative side effect of the extensive system is the need of more land for grazing. However, there is no reason why animals should occupy land that is not already productive. These results, taken together, demonstrate that the integration of poultry (chickens and geese) with orchards allows a more rational use of renewable and not renewable resources. Integrated production systems determine closed links between animal breeding and plant production, resulting in a more circular agriculture and higher profits, due to a double income (animals and plant culture).

Keywords: Agroforestry, Chickens, Geese, Pasture, Sustainability

Microbial-mineral manure additive reducing odours

Abstract ID: 69

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Poland has become the biggest poultry meat producer in the EU with close to 20 percent of all EU-28 broiler production. Unfortunately poultry production is strongly linked with livestock odor emission. Thus, there is a strong need to find a method which will allow to control concentration of odorous compounds. The aim of the study was to investigate the effectiveness of the innovative microbial-mineral manure additive (MA), which was developed at Lodz University of Technology, in ammonia emission mitigation. The experiment was carried out in a poultry house equipped with a battery cage system, where laying hens were kept. At the beginning of each week the manure was removed from conveyor belts below the cages. On the last day of each cycle, air samples were taken from the poultry house's space to determine the concentration of NH_3. During the control cycles (5 cycles) MA was not applied on the manure. During similar experimental cycles (also 5 cycles), 70 g of MA were applied per cage on the 2nd day of each cycle, topically onto the manure. Throughout the experiment, hens were fed the same feed in order to eliminate the variability of feed ingredients, which could affect the amount of excreted nutrients, and thus the concentration of odorous compounds. The concentration of ammonia was increasing with every week of the investigation. During the control cycles it was between 4,67 ppm on the beginning and 12,56 ppm at the end of the investigation. In comparable periods of the experimental cycles it was between 3,93 and 12,85 ppm, respectively at the beginning and the end of experiment. No statistically significant differences were noted between the control and experimental cycles. The MA did not affect the ammonia content in the poultry house's air most probably, because of the very low moisture content in the manure, which was being systematically dried up while lying on the belt under the cage. It can be assumed that the effectiveness of the MA decreases with the increase of dry matter content in the manure, thus the use of investigated litter additive in the cage system is ineffective.
The research was financially supported by the National Centre for Research and Development, grant no. PBS2/B8/14/2014: "Innovative bioadditive for poultry production premises".

Keywords: Ammonia, Laying hens, Manure, Microbial-mineral

On farm hatching in broilers – Effect on production, welfare and profitability

Abstract ID: 201

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On farm hatching is gaining more and more attention in broiler practice. Instead of day-old chicks, eggs are being transported to the broiler house after 18 days of incubation and chicks hatch out in a production environment with immediate access to feed and water. Early intake of feed particles can stimulate gut development, supporting an optimal start of the production cycle. Several commercial concepts are already available, differing in the extent of automation, hatching conditions and investment cost. The Experimental Poultry Centre (EPC) investigated the effect of on-farm hatching on production, welfare and profitability. The trial was conducted during 3 production cycles in 4 climate independent rooms, each divided in 4 pens (± 1550 chickens/pen). Each room consisted of 2 pens where chicks hatched on-farm (HOF) and two control (C) pens with day-old chicks from the hatchery (same parent stock). The commercial system "Home hatching@" of the company Pe-Da was used. Production data were monitored, foot pads were scored at day 39 (2 cycles) and litter dry matter % was measured weekly. The contribution margin (CM) was simulated (total income of meat production minus the costs for feed and chicks/eggs, corrected with an extra heating cost for on farm hatching). Differences were seen in mortality, feed intake and slaughter weight, although production efficiency numbers did not differ significantly. Litter dry matter % was higher in HOF groups. Foot pad dermatitis was seen less in the HOF groups, with 48.5% of the chicks scoring 0 vs 22% in the control group. On-farm hatching takes three days longer than a conventional production cycle (e.g. 43 vs 40 days). If a farmer can manage to fit in the concept in a 7 week cycle length, CM can be 0.22 euro per chick per year higher. When a farmer needs to extend the cycle length to 7.5 weeks, the yearly number of production cycles will reduce and we calculated a loss of 0.075 euro per chick per year. We conclude that on-farm hatching can be beneficial for animal welfare and profitability if cycle length can be maintained.

Keywords: Broilers, On farm hatching, Profitability

Perch use by broiler breeders in commercial flocks

Abstract ID: 482

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In Europe, access to perches is not only recommended for laying hens but also for poultry breeders. In Germany, broiler breeders are kept in single tier floor barn systems with a litter and a raised slatted area. Except for a part of northern Germany (Lower Saxony), perches are not commonly provided. However, there are no legal requirements on perch design, i.e. for the material and the floor or grid below. The on-farm study focused on the detailed description of the present perches in commercial broiler breeder flocks with special regard to perching behaviour.
A total of nine broiler breeder flocks housed in commercial farms in northern Germany were included in the study. Perch use was recorded at two times during production (in the first and in the last third). Therefore, scouting cameras were installed, and the number of hens perching was counted during 48 h in 15 min intervals.
The nine commercial farms offered perches in different areas of the barn (in the slatted area as well as in the litter area), made of diverse materials (plastic, wood, metal), and at various heights (38.0 – 48.0 cm above the littered area, and 0.0 – 15.5 cm above the raised slatted area). In general, perch use was higher at night (Ø 1.93 birds/m) than during day-time (0.65 birds/m), and more hens resided on the perches installed in the litter area (Ø 3.42 birds/m) than on the slats (1.93 birds/m). Furthermore, the material and the height had an effect on perching behaviour.
The status quo survey in broiler breeder farms with perches in Germany showed that a diversity of perch variations is present, which differ in use intensity by the birds. Nevertheless, they all seem appropriate to enable resting behavior of broiler breeders. The results of our on-farm study indicate that perch use can be increased by improving perch location in the barn and perch height.
The study was supported financially by the Lower Saxony Ministry of Food, Agriculture and Consumer Protection.

Keywords: Broiler breeder, On-farm, Perching

Providing ramps during rearing improves bone strength in laying hen pullets

Abstract ID: 313

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The rearing phase is one of the most crucial phases in the life of a laying hen due to development of locomotor/cognitive skills and skeletal integrity needed for the future laying environment. As part of a larger project investigating the design of rearing aviaries on behaviour during the rearing and laying phases, initial analyses focused on pullet bone properties. The aim of this study was to investigate the provision of ramps during rearing on bone biomechanical properties and production parameters. Laying hen pullets were reared in a rearing barn with eight pens (n= 600 birds per pen) from day of hatch until 17 weeks of age (WoA). Four of the pens contained an aviary system with an offset structure (i.e., vertical tiers stacked in an off-centered manner) whereas the other pens contained an aviary with a direct structure (i.e. tiers stacked on top of each other). Within each aviary structure type, two pens contained ramps (R) that connected the different tiers providing a continuous pathway between the highest and lowest tiers compared to pens without ramps (NR). At 16 WoA, 10 birds per pen (n= 80) were randomly selected, euthanized, weighed and the left tibia and humerus collected. Bones underwent a 3-point bending test with outputs used to calculate Young's modulus, maximum load and work to maximum load for each bone. Feed consumption per hen and mortality were recorded at the pen-level on a daily basis and summarized every four weeks (WoA 4, 8, 12 and 16). Statistical analyses were conducted using linear mixed effects models in R. Birds reared in aviaries with ramps had a higher Young's modulus for the humerus compared to birds reared without ramps (R: 167.8 ± 25.2 N/mm vs. NR: 152.3 ± 23.5 N/mm, P= 0.0035). Compared to tibiae, humeri had a higher maximum load (humerus: 3.2 ± 0.38 N vs. tibia: 3 ± 0.28 N, P < 0.001) as well as a higher value for work to maximum load (humerus: 0.51 ± 0.07 [J] vs. tibia: 0.38 ± 0.08 [J], P < 0.001). Mortality increased with age (WoA 4: Ø 0.54 %, WoA 8: Ø 0.89 %, WoA 12: Ø 0.97 %, WoA 16: Ø 1.02 %, P< 0.001). Feed consumption per bird did not differ between treatments. These results suggest that access to ramps and consequent changes in moving behaviour (e.g. WAIR) during rearing improve wing bone strength in pullets.

Keywords: Bone strength, Laying hen pullet, Ramps, Rearing aviary

Qualification and continuing education for people working in the poultry sector

Abstract ID: 208

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The framework of the poultry production is constantly changing. New production systems and techniques, an increasing importance of animal welfare as well as management tools and the current level of scientific knowledge are challenging the competences of people who work with poultry at different stages of production. Furthermore qualification and continuing education in order to approve, expand or renew knowledge becomes more and more important in quality assurance schemes (e.g. supplier contracts) and is even required e.g. in the German Animal Welfare Act. Therefore, on the one hand people are 'required' to participate in further training and on the other hand people participate voluntarily in order to improve their competences. Thus, continuing education – and the certificates to validate that it has happened – is therefore mandatory for those involved in the poultry industry. This emerging demand opens the discussion for a closer integration of vocational training, continuing education and advanced further education. Presently, the "in" catchwords are "Dual Course Studies" and "Open Universities". Within a project, funded by the German Federal Ministry for Education and Research (BMBF), the University of Applied Sciences Osnabrück founded the "Osnabrücker Poultry Academy" (OPA) which worked out a concept that may meet all demands and requirements and may be best explained using the example of the certificate course "Poultry Professional" (PP). The PP is an on-the-job certificate course targeting people working in the poultry sector who want or need to approve, renew or expand their knowledge and skills. The course, which comprises 300 hours workload and covers level 5-6 of European Qualification Framework (EQF), is designed as a blended learning education program combining online digital media with traditional classroom methods. The gained certification "Poultry Professional" and acquired knowledge must be maintained by collecting 20 continuing education credits (CEPS) within 2 years. Therefore the "Poultry Professionals" have state-of-the-art knowledge. However, the same program can also be completed on EQF level 6-7 which includes further examinations and workload (600 hours). Along with the certificate, successful participants then receive 20 ECTS which will be approved in the master program "Applied Poultry Sciences", a dual master program currently being created by the OPA. This would be the first step to a higher qualification. Continuing education offers quality assurance in the whole production chain and addresses inter alia the sensitive issues of food safety and animal welfare and thus, may promote the public image of the poultry industry.

Keywords: Academy, Lifelong learning, Poultry, Qualification

The role of farms of Galliformes birds during 2016-2017 Avian Influenza Epizootic in Europe

Abstract ID: 375

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Avian influenza viruses pose significant threats to avian and human health and causes tremendous damage to the poultry industry, in terms of mortality, welfare, economic losses and trading restrictions. The objective of the study was to estimate the role of farms of Galliformes birds during 2016-2017 Avian Influenza Epizootic in Europe. During 2016-2017 (until 11/12/2017) there were 2,853 cases of high pathogenic avian influenza in Europe, which led to the death of 11,183,710 birds. The majority of the cases were in wild and backyard birds, with exception of France and Hungary, where the cases were mostly in duck-goose farms. In addition, among 8 European countries whose reports came from farms of Galliformes birds, there were 130 cases of HPAI in fattening turkey, 36 in layer, while in broiler farms there were only 11. Particularly, in Germany (85) there were 76 cases in turkey, 5 in layer, 2 in breeder and another 2 in turkey breeder farms. In Italy (59) there were 39 cases in turkey, 16 in layer and 4 in broiler farms. In Hungary (18) there were 10 cases in turkey, 6 in layer and 2 in broiler farms. In France (10) there were 5 cases in broiler, 3 in layer, 1 in breeder and 1 in turkey farms. In Holland (5) there were 3 cases in layer and 2 in breeder farms. In UK (5) there were 4 cases in turkey and 1 in breeder farms. In Sweden (2) there were 2 cases in layer farms. Finally, in Greece (1), there was one reported case in a layer farm. Overall, 74% of cases were reported in turkey farms, 17% in layer farms, 6% in breeder farms and 3% in broiler farms. Although wild birds are natural hosts and reservoir for all types of avian influenza viruses, farms of Galliformes birds play a crucial role to the spread of the disease into an area. Since the introduction of avian influenza in farms might lead to severe damages and trade restrictions, it is essential to eliminate the exposure of poultry to wild birds through a number of restrictive measures. In conclusion, the type of poultry farms should be taken into account for the implementation of biosecurity measures, since fattening turkey and layer chicken farms pose a major risk factor for an avian influenza outbreak.

Keywords: Avian influenza epizootic, Europe, Farms, Galliformes birds

ORAL PRESENTATIONS

Poultry Welfare

An integrated farming concept with the use of dual-purpose chickens: investigations of aspects related to animal health and immunity

Abstract ID: 374

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There is an increasing concern in the public perception of commercial poultry production in many European countries. These include ethical concerns over killing day-old layer males as well as beak trimming and losses associated with cannibalism especially in layer-type chickens. Also health problems which are associated with fast-growing genotypes are under discussion. A newly bred line of dual-purpose chickens, which allows the use of the females for egg and of the males for meat production, may provide a possible approach to overcome some of these concerns. The aim of this project was to compare both genders of dual-purpose chickens with a commercial layer-genotype regarding performance, health and immunity parameters. By using an integrated farming concept, pullets, layers and males were all kept on one farm. Overall, both genders of the dual-purpose chickens were calmer than the layer-type birds, which correlated with lower cannibalism and reduced mortality rates. Furthermore the immune responses varied between these genotypes despite the same vaccination program and management in the field. Significantly higher serum antibody levels were detected in dual-purpose chickens after vaccination against Newcastle disease and infectious bronchitis compared to the layer-type birds ($P < 0.05$). With an average of 270 eggs/hen of the dual-purpose line the egg production was lower compared to an averay of 311 eggs/hen of the commercial layer line. On the other hand, male birds of the dual-purpose line had significantly higher body weight at the time points of slaughter at 75, 63, and 64 days in placements 1, 2, and 3, respectively, as compared to the males of the layer line. Overall, the integrated farming concept provided an environment for raising healthy birds, which was comparable or better to commercial settings in the field. Therefore, this new farming concept is from the point of animal welfare an interesting alternative to other commercial settings. But certainly, aspects of sustainability, economy and consumer perception of this concept as well as its products have to be evaluated further before this concept may find application in the field.

Keywords: Both genders, Dual-purpose chicken, Integrated farming concept, Poultry health

A probiotic containing viable spores of Bacillus licheniformis reduces lameness in broilers

Abstract ID: 209

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The probiotic is a feed additive, consisting of viable spores of a unique *Bacillus licheniformis* strain (BL). Objective of the 53 day study was to determine, if the probiotic can reduce and delay incidences of lameness in bacterial chondonecrosis and osteomyelitis (BCO) induced broilers. Broilers were randomly assigned to 3 treatment groups with 40 birds per treatment and housed on wire to induce BCO without any bacterial challenge. Treatment groups were: 1. no additives (control group; CG); 2. enrofloxacin group (EG; enrofloxacin 10mg/kg body weight from day 38-47) or 3. BL (1.6×10^9 cfu *Bacillus licheniformis*/kg of feed), fed continuously. Lameness percentage at the end of the trial was determined. Significance was set at $P<0.05$. Day of first lameness incidences – defined as 3% of broilers being lame - was identified.The study shows, that the addition of enrofloxacin and BL can significantly reduce lameness in broilers compared to broilers receiving no additives. The lameness percentage was reduced from 36% in CG to 10% in EG and 16% in the probiotic group. Differences between the EG and the BL group were not significant. The first incidences of lameness were reported at day 40 in CG, while in the probiotic and EG groups the onset of lameness took place on day 45 and 46, respectively. Feeding of this unique *Bacillus licheniformis* strain reduced the occurrence of lameness in BCO induced broilers significantly. No significant differences in the occurrence and the day of onset of lameness between enrofloxacin and BL treated broilers making the probiotic a viable alternative to the usage of antibiotics.

Keywords: Bacillus licheniformis, Lameness, Probiotic

Assessing the Welfare of end of lay hens during the catching and packing process of depopulation

Abstract ID: 422

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The process of depopulation has been reported to cause severe injuries, yet studies investigating depopulations in modern aviary systems or more subtle effects on welfare are lacking. The current work assessed physical, physiological, and behavioural measures of welfare on 15 commercial farms. We hypothesized that measures of stress and fear (plasma corticosterone, breathing rate, tonic immobility, and cloacal / comb temperature) and injury (fractures, luxations and plasma creatine kinase concentration) would be affected by depopulation relative to baseline measures collected within 2h prior to depopulation. We also hypothesized that the distance hens are carried and duration into the depopulation would influence these measures. Separate groups of hens were assessed for baseline measures and during depopulation, with each further split into subgroups (6-17 hens per farm; grand total N=603). Blood samples and temperature (comb and cloaca) were collected in two subgroups once for baseline and depopulation (N=290), breathing rate and tonic immobility (TI; # inductions required (max=3), latency to first head movement (LAT1) and standing upright (LATSU)) assessed in the others (N=313). In addition, "catchers" were equipped with head cameras in order to identify collisions with the aviary furniture. Linear mixed models with a nested design (individuals within farms) were used to test for differences between baseline and depopulated hens. The number of TI-inductions was analyzed using a multinomial logistic regression. In total 27 out of 319 hens (~8%) exhibited at least one fresh fracture or luxation after depopulation. Average creatine kinase concentration was elevated by 5.8% ($p<0.05$) after depopulation compared to baseline. Furthermore cloacal temperature was elevated by 0.9 ^{o}C ($p<0.05$), and comb temperature reduced by 1.9 ^{o}C ($p<0.05$). Breathing rate was increased by five chest movements per minute higher ($p<0.05$) and corticosterone levels by 16% ($p<0.05$). There was no difference between baseline and depopulation for LAT1 ($p=0.46$, overall mean ± SE=74.5s ± 5.4s) and LATSU ($p=0.5$, overall mean ± SE=117.1 ± 8.4s). However, TI-induction was more likely to require three attempts ($z=2.5$; $p<0.05$) or fail ($z=2.5$; $p<0.05$) in baseline hens. With respect to duration into depopulation, fractures and luxations ($p=0.06$), collisions with pen furniture ($p=0.07$), LAT1 ($p=0.06$), LATSU ($p=0.03$) and corticosterone levels ($p=0.09$), tended to be higher towards the end. Carry distance was not related to any of the presented measures. These results indicate that depopulation can be physically and psychologically challenging for hens. Future work will focus on alternative methodologies to reduce injuries, stress and fear of hens.

Keywords: Depopulation, Laying hens, Welfare

Being fearful or calm individual predisposes different development of keel bone in laying hens

Abstract ID: 240

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We evaluated hypothesis suggesting that an individual hen that is predisposed to be more fearful and less explorative has problems to deal with challenging commercial situations such as flying off perches which may have a devastating effect on keel bone development. The laying hens (n=93) of Slovenian Styrian breed were individually tested four times (17, 18, 29 and 33 weeks) for 3 min in an open-field test. We primarily used this breed in order to increase the understanding of ethological needs of primitive chickens to enhance their welfare if they are to be reared in more intensive production systems and secondly as their variation in behavioural responses is expected to be larger. A commercial flock of hens with 15 cockerels was housed on sawdust with wooden perches (190 cm x 4 cm x 6 cm). A hen was placed in the pre-starting area for 30 s. Then, using one-zero sampling every 10 s, fear responses were scored, ranging from 20 (calm animal) to 80 (fearful animal), as well as explorative responses (i.e. latency to leave the area, to reach the central zone, frequencies of crossing central zone, pecking and preening). Keel bone fractures and deformations were recognized by palpation at 33 weeks of age, which is at the pick of a laying period. Based on fear responses, hens were categorised into three fear groups (High, n=19; Intermediate, n=67 and Low, n=7 hens). Fearful hens (from High fear group) were least explorative, indicated by the lowest number of hens that left the area ($Hi^2=8.07$, $P<0.02$), the lowest frequency of pecking (pre-starting area: 34.65%; $Hi^2=74.96$, $P<0.001$; main area: 9.9%; $Hi^2=43.54$, $P<0.001$) and preening (pre-starting area: 11.88%; $Hi^2=33.57$, $P<0.001$; main area: 3.96%; $Hi^2=73.78$, $P<0.001$). Interesting, not only exploration, but also keel bone development was associated with hens' fearfulness. Fearful hens were individuals with most keel bone deformations (42.1%; $Hi^2=13.54$, $P<0.01$) while the least with keel bone fractures (10.5%; $Hi^2=10.77$ $P<0.02$) and with both types of damages (21%; $Hi^2=8.24$, $P<0.02$). The reverse picture to that seen in fear hens was observed in calm hens. It is therefore suggested that emotional states such as fear and exploration have a significant impact on keel bone development which provides a new finding in the field of poultry welfare.

Keywords: Exploration, Fear, Keel bone, Open-field test, Primitive chicken breed

Comparing stress levels and behavior in two lines of layer hens transported with and without break

Abstract ID: 466

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The present study examines the effects of transport on the welfare and stress level of 18-week old pullets in journeys from the rearing farm to the farm of laying hens. For this purpose two practice-relevant transport variants were compared: "night" (transportation with a break) and "direct" (transportation without a travel break). Animal subjects were free-ranged, organic mixed held pullets H&N Super Nick (HNS) and H&N Brown Nick (HNB) of the breeder and distributor H&N International, Cuxhaven, Germany. The study was conducted of 15 different flocks with a total of 30'210 pullets of Switzerland's biggest organic egg supplier, hosberg AG. Altogether 5751 birds were sampled to measure corticosterone metabolites (CM) out of feces with an enzyme immunoassay (EIA). Individual droppings were collected before transportation to determine baseline concentrations of CM and 0 h, 3 h, 6 h, 10 h, 24 h, 48 h, and 72 hours after the end of transportation to measure stress response. Taking account of the circadian rhythm flocks transported "direct" were sampled additionally 34 h and 58 h. Further 750 pullets were weighted and 600 were scored for their health (i.e. plumage condition) and behavior generally before and after transportation. The statistical analysis of the data was performed with the programming language and environment for data analysis and graphics R (version 3.4.0). Overall, mean baseline concentrations of excreted CM ranged from 43 to 66 ng/g for both variants. Mean CM at 0 h (first samples after end of transportation) were highest with 323 ng/g and 173 ng/g for "night" and "day", respectively and 298 ng/g and 191 ng/g for HNS and HNB, respectively. Weight loss during transportation was significantly correlated (negative) with the transport variant. Birds of variant "night" lost 2.1% weight (95% KI: [-2,6; -1,5]). HNB had 0.5% (95% KI: [0,3; 0,7]) higher loss in weight than HNS. Scoring results for plumage condition did not highly differ before and after transportation. The results show that differences in stress levels and behavior appear most between the two evaluated layer lines and not between transport variants. Therefore it should be taken into consideration that not all transportation variants may be optimal for all layer lines. Further investigations on this topic are necessary.

Keywords: Corticosterone metabolites, Pullet, Stress parameter, Transport, Welfare

Development of a user-friendly protocol and webtool for monitoring and benchmarking broiler chicken welfare during the pre-slaughter phase

Abstract ID: 577

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The pre-slaughter phase (i.e. the phase at the end of the production period from the catching & loading of the animals until slaughter) is a critical period for broiler welfare and farmer income. We aimed to develop a pre-slaughter broiler chicken welfare monitoring protocol which meets several criteria: user-friendly, reliable, valid, discriminative, allows benchmarking of individual measures as well as an overall integrated welfare score (IWS), and based on expert opinion. We identified, from literature, 16 potential animal-based welfare measures that can be assessed at the slaughter plant. We collected data on these 16 measures for 81 transportations of commercial Belgian flocks. Three measures were removed because they were not affected by the pre-slaughter phase in our sample of transportations. In order to downscale the protocol further, another 4 measures were removed by considering the amount of time that could be saved and the relative weights that were allocated to the various measures by a panel of 9 poultry experts. The final protocol includes measures of fractures, bruising, panting/huddling, splay-legged birds, crowding, supine birds, stuck body parts, mortality and carcass rejections. Inter-observer reliability of the measures that could be tested was high (>73%). It takes circa 50-60 min for an experienced assessor per flock. In order to calculate the IWS, data were aggregated in three steps based on expert opinion (n = 17 or 19). First, observed prevalences were converted to measure scores (from 0 to 100) reflecting expert acceptability for broiler welfare. Then, measure scores were multiplied by weights reflecting the experts' ranking of the importance of these measures for broiler welfare, and by a compensation reduction factor based on the measure score rank, and then summed. The IWS gives more weight to low measure scores, which corresponds with how experts scored overall welfare of hypothetical flocks. Application on 53 flocks showed a good spread of IWS and good sensitivity to extremely low scores. The protocol describing the precise methodology and the accompanying webtool for calculating the IWS and for benchmarking are freely available (http://survey.ilvo.vlaanderen.be:3838/WQ/). It can be used in quality assurance schemes and for identifying best practices, potential preventative actions and risk factors for broiler welfare during the pre-slaughter phase.

Keywords: Animal welfare, Broiler chicken, Expert opinion, Integrated welfare score, Transportation

Economic Supporting Model to Determine Optimal Age of Layers Flock Replacement

Abstract ID: 198

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This Model is an economic supporting tool in determining the optimal length of laying period. The model uses the financial Excel PMT Function, to calculate the Average Monthly Balance (AMB) based on the Present Value of cash flows related to various laying periods examined. It should be noted that the approach taken in this analysis is an additional option to an already known method described in the literature for determining the optimal age of replacement in layer flock, namely the Marginal Net Revenue approach (MNR). The model is implemented to support consideration in the following cases: 1. Examining force molting versus no molting flock profitability, 2.Examining optimal flock age of replacement based upon monthly intervals, 3. Comparing the option of replacing to a new flock versus the existing one in cases in which the current flock gains poorer performance than initially expected upon regular professional norms (relate to monthly layer performance tables in Israel). The monthly production costs comprise the follows components: flock purchase, chicken-house cleaning and disinfection, feed, labor, energy and miscellaneous. The monthly revenues: table eggs & broken eggs. The cash flow of the laying period is the series of figures resulting from subtraction of production costs from the revenue in each and every month. The maximal AMB represents the optimal age to replace the flock. Based on Israeli economic conditions: 1. Induced molting has an economic advantage yet Israel bans induced molting in laying hens for animal welfare reasons, 2. 23 months of age was found to be the optimal time for a normative flock replacement without induced molting, 3. The optimal age for replacement of poorer flock performance may differ compared to the normative one. It will be determined by the intersection of the Nominal Monthly Balance (NMB) of poorer performance versus AMB of a normative flock. The model efficiently supports economic based time replacement decisions for standard conditions layer flocks as well as for poorer flock performance.

***Keywords**: AMB approach, Economic Supporting Model, Layers flock replacement decision*

Effect of a chemical Litter Amendment on Animal-based welfare Indicators and Litter Quality in a European commercial Chicken Husbandry

Abstract ID: 179

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Foot pad dermatitis is considered as one the main welfare concerns in broiler husbandry. Several studies have shown that litter moisture is one of the main reason for foot pad lesions and also promotes microbial growth of nitrifying bacteria. The aim of the current study was to evaluate possible effects of sodiumbisulfate (SBS) as a litter additive on foot pad health (FPD), hock burn (HB) and litter parameters. Two different application rates of SBS were examined on a commercial broiler farm. 30,000 chicks per group were placed on 700g/m² spread speltgranulate and kept for 36 days. In the first experiment (TRT1) 250g/m² SBS were spread on top of the litter 20 hours before the chicks were housed; in the second experiment (TRT2) the amount of SBS was reduced to 150g/m². Each experiment consisted of one treatment group (SBS) and a control group without treatment (CON). Both experiments were repeated twice. Litter parameters (pH and percentage of dry matter) and condition of foot pads, hocks, both in accordance to the 5-point scale of Welfare Quality®, and body weight development of the same randomly sampled birds (n = 60 per group) were recorded weekly. Mortality rate was higher in SBS groups when compared to the CON groups (TRT1 2.8 vs 2.0%; TRT2 2.9 vs. 2.3%), whereas SBS had no effect on body weight development. The incidence of FPD was significantly reduced in both SBS treated groups compared to CON groups ($p<0.05$) with TRT1 group showing the best results. The incidence of HB was not affected by SBS ($p>0.05$) but by dry matter content of litter ($p<0.05$). At time of housing, SBS reduced the pH of litter to 1.7 and 2.0 in TRT1 and TRT2 when compared to 6.5 and 6.7 in control groups. Litter pH in TRT groups increased over time and approached the pH of control groups by day 15. The results of the current study indicate that SBS treatment may be beneficial regarding foot pad health in broilers. However, further studies are needed to investigate alternative SBS application rates and dispersing intervals and to verify the results, especially regarding mortality rate and the interaction between pH, litter moisture and climate conditions (ammonia).

Keywords: Broiler, Footpad dermatitis, Litter treatment, Sodiumbisulfate, Welfare

Effect of Feed Color on Growth Performance and Tonic Immobility in Broiler Chicks

Abstract ID: 528

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Vision is important in poultry performance and welfare. Color of feed is one of the factors that can affect welfare and performance of poultry. This experiment was conducted to investigate effect of different feed colors on growth performance and tonic immobility in broiler chicks. A total of 240-day-old broiler chickens (Ross 308) were randomly allotted to 4 treatments with 5 replicate pens in a completely randomized design. The treatments concluded: control and three colored feed (blue, green and red). Broilers received dietary treatments from 1 to 42 d in three phases: starter (1-14 d), grower (14-28 d) and finisher (28-42 d). Growth performance parameters (feed consumption, weight gain and feed conversion ratio) were measured in different growth periods. Tonic immobility as fear reaction were determined at 35 d. Feed consumption were increased in green and red feed ($P< 0.01$). Broilers in colored feed groups were heavier than control ($P< 0.05$). Lower feed conversion ratio was observed in blue feed and control ($P< 0.05$). Broilers in blue feed group showed short duration of tonic immobility (less fearful) and in red feed long duration ($P< 0.05$). The results demonstrated, blue feed had favorable effects on performance and welfare of broiler chicks.

Keywords: Broiler chick, Feed color, Performance, Tonic immobility

Effect of keel bone fractures on laying hen behaviour in a non-cage housing system

Abstract ID: 342

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Non-cage housing systems such as aviaries allow laying hens to perform a variety of natural species-specific behaviour. However, they are suspected to increase the risk of keel bone damage due to their complexity and height that favour crashes and collisions. Keel bone fractures in laying hens pose a problem for animal welfare since they are likely to be associated with pain and might hinder or restrict hens' ability to perform natural species-specific behaviour. The aim of this study was to determine whether keel bone fractures affect laying hen behaviour in a non-cage system and whether this alteration is caused by pain. 80 Brown Nick and 80 Nick Chick hens were housed in 8 identical pens (20 hens/pen), whereas 10 hens/pen were individually marked, and video recorded at 37 and 39 weeks of age. All hens received an analgesic in their water supply (Paracetamol 20 mg/kg) prior to one of the two video collections for 7days with video recordings taking place on day six and seven. To assess keel bone fractures, hens were x-rayed after both data collections and keel bones were scored using three classifications: no fracture, healed fractures and a visible fracture gap. Statistical analysis was done by applying (generalised-) linear mixed effects models. Frequency of rapid comfort behaviour (e.g. tail shaking, feather ruffling, wing flapping and stretching) was affected by the interaction of the keel bone state and analgesic ($P=0.01$) where the frequency was relatively greater with a visible fracture gap and no analgesic and lower with the same keel bone score but with administration of the analgesic. Duration of feather maintenance increased with the administration of the analgesic ($P=0.01$) while the probability of perch or nest access by flying and jumping decreased when a fracture gap was visible ($P=0.004$). In conclusion, certain comfort and mobility behaviour were affected by keel bone fractures, especially when a fracture gap was visible. Since the analgesic reversed the fracture effect in one of the behaviour types, pain cannot be dismissed as a factor that influences behaviour in birds with keel bone fracture.

Keywords: Analgesic, Behaviour, Keel bone fractures, Laying hen

Effects of a treatment against Dermanyssus gallinae with fluralaner on welfare parameters in laying hens

Abstract ID: 283

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Poultry Red Mites infestation (PRM, *Dermanyssus gallinae*) is widely recognized as a major animal welfare threat in layers. This study investigated the effect of a new drinking water PRM treatment containing fluralaner on the improvement of welfare parameters in layers, including specific blood biomarkers of stress and anemia, mortality and performances.The study involved a commercial layer house with enriched cages, containing 12,700 29 week-old hens infested with PRM. The infestation level was determined weekly by quantitative and qualitative mite traps, over a study period of 13 weeks. Seven weeks after the study start, the hens were administered a fluralaner solution in drinking water, twice 7 days apart, at the recommended dose. One week before treatment (baseline, Week-1), and again at Week 1 (i.e. immediately after the 2nd administration) and at Week 6, blood stress biomarkers and haematological parameters were measured from a sample of 50 hens. Egg production and mortality were recorded daily, before and after treatment. Mite counting in the traps revealed a high infestation till treatment (ca. 1500-2200 mites per trap in average) which almost disappeared after treatment (< 2 mites per trap from Week 3 till Week 6). Blood corticosterone concentration decreased from 4.0 ng/mL (baseline) to 2.7 ng/mL at Week 1 (P=0.02) and to 1.7 ng/mL at Week 6 (P=0.003). The heterophil-lymphocyte ratio decreased from 0.6 to 0.3 and to 0.1 (P<0.0001 at both time points). Treatment had no effect on ovotransferrin and adrenaline levels.Haemoglobin concentration increased from 7.0 g/dL (baseline) to 8.0 g/dL at Week 1 and to 7.8 g/dL at Week 6 (P<0.0001 at both time points). Similarly, mean corpuscular haemoglobin increased from 32.9 pg (baseline) to 33.9 pg (P=0.06) and 36.9 pg (P<0.0001) at the same post-treatment time points.The daily mortality rate decreased from 0.012% before to 0.007% after treatment (P=0.04) whereas the daily egg production increased from 85.2% to 91.6% after treatment (P<0.0001). This study demonstrated that effective PRM control with fluralaner induced a significant improvement of the layers welfare parameters.

Keywords: Fluralaner, Hen welfare, Poultry red mites, Stress biomarkers

Effects of hybrid and ramps status on health parameters and behaviour of laying hens: An on-farm study

Abstract ID: 593

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Multi-tier systems are increasing in popularity, due to the freedom of movement and expression of natural behaviour they provide for hens in comparison to cage systems. Complex environments can lead to high levels of keel bone damage, ramps between tiers can help alleviate this problem by increasing safe transitions. A previous experimental study showed that ramps benefited foot pad health and brown hens had better foot pad health in comparison to white hens. In the current study, commercial flocks (ranging 42-85 weeks of age) of white hens without ramps (n=4), brown hens with ramps (n=7) or without ramps (n=7) were scored for keel bone damage and foot health. Jumping/flying and uncontrolled movements (falling from a tier/perch and crashes with structures of conspecifics) were monitored. Data were analysed using generalised linear mixed models (keel and foot scores) or linear mixed models (behaviour), with ramp status, brown vs white birds and age as fixed factors, and flock as random factor. Due to sample size, interactions were only tested for behavioural data. Behavioural data were square root transformed where necessary and time of day was included as a fixed factor. Behavioural data are presented as the number of movements per bird in an area during a 15-minute observation period. Keel bone fracture prevalence tended to be lower in flocks with ramps (64% ± 5%) compared to those without (73% ± 3%, P=0.0993). Foot pad lesions where more prevalent in white flocks (36% ± 8%) than in brown flocks (20% ± 3%, P=0.031). Bumble foot was less prevalent in flocks with ramp access (5% ± 2%) than flocks without (13% ± 3%, P=0.016). White hens jumped/flew downward more than brown hens (P=0.010) (1.5 ± 0.3; 0.6 ± 0.1, respectively) and tended to jump/fly more irrespective of the direction (3.8 ± 0.7; 2.1 ± 0.3, P=0.052). Jumps/flight occurred more during the dimming period (3.7 ± 0.4) than during daylight (2.5 ± 0.4, P=0.019). There was an interaction between time of day and age for uncontrolled movements (P=0.027), these movements decreased with age during dimming but were steady over daylight.
In summary, ramps between tiers can improve health by decreasing keel fracture and bumble foot prevalence, although fracture prevalence was still high. The mechanism behind the reduced fracture incidence requires further study, as ramps did not affect the occurrence of behaviours thought to cause keel fractures (falling/colliding) and white hens moved more and may be affected by ramps differently.

Keywords: Behaviour, Hybrid, Keel bone damage, Laying hens, Ramps

Electrical stunning of poultry: impact factors of stunning efficiency and meat quality

Abstract ID: 334

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The objective of stunning animals before bleeding is to induce unconsciousness, which lasts until the animal dies. The exact rules are described by the European Union in the regulation n°1099/2009 from the 24th of September 2009 on the protection of animals at the time of killing. The stunning method should ensure immediate loss of consciousness from bleeding to death of animals, to spare them unnecessary physical hardship, stress and fear. In France, electrical stunning using an electric water bath is the method generally used to stun broilers. The regulation requires a minimum current value distributed to each animal depending on the frequency applied. The objective was to establish electric parameters allowing to stun broilers in a reversible way and respecting animal protection, without degrading products quality. Three statutory electrical parameters (100 mA-50 Hz, 150 mA-400 Hz, 200 mA-1000 Hz) and three below the regulations (60 mA-400 Hz, 100 mA-400 HZ and 100 mA-600 Hz) have been evaluated. Our results showed that stunning efficiency was better for the statutory parameters with frequencies under 400 Hz, and on the contrary, the product quality was better with the low intensities (lower than 100 mA) and high frequencies (superior or equal to 400 Hz) with carcasses presenting less red wings. Regarding only animal protection, awakening tests allowed us to conclude that the statutory parameters (100 mA / 50 Hz, 150 mA / 400 Hz and 200 mA / 1000 Hz) as well as the parameters below the regulations 100 my / 400 Hz stunned in an effective way the chickens. Indeed, when there was a return of consciousness, it was always 45 s after stunning as recommended by EFSA, and for statutory parameters 100 mA / 50 Hz and 150 mA / 400 Hz, stunning was irreversible for all birds. The only parameters allowing a reversible stunning, were those below the regulation values, worth namely 60 mA / 400 Hz and 100 mA/600 Hz. However, with these two parameters, more than 40 % and up to 77 % of chicken presented signs of returning to consciousness within 45 seconds, which is problematic for animal protection. Finally, the solution to ally stunning efficiency and products quality will doubtless pass by a sum of compromise and a good control of the slaughter process.

Keywords: Animal protection, Electrical stunning, Meat quality

Environmental enrichment increases positive activity in broiler chickens

Abstract ID: 130

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Provision of biologically relevant enrichments may enable animals to perform highly motivated behavior which may reduce frustration and boredom, and also increase their physical activity. Most Norwegian broiler companies have now made it mandatory to use a variety of enrichments in their broiler production, including provision of peat, roughage and elevated platforms. The combined effects of these enrichments may encourage exploration, locomotion, comfort activities and possibly contribute to improved leg health. The aim of this pilot study was therefore to investigate effects of commercially applied environmental enrichments on behavior and lameness in broilers. During 2 consecutive flocks, a total of 18,200 broilers (mixed sex Ross 308) were randomly allotted to either an enriched or control treatment in a commercial broiler house. The house was divided along the middle by a plastic wall, ensuring that no birds or enrichment materials could move between treatments. The flocks were observed at 16 and 30 days of age to investigate differences between enriched (peat, bales of lucerne hay, and elevated platforms) and control birds with regards to behavioral activities and lameness. At 30 days of age, 50 random birds from each treatment per flock were gait scored to assess lameness. More running ($P<0.001$), worm-running ($P=0.006$), play fighting ($P=0.015$), dust bathing ($P=0.009$) and ground pecking while standing ($P<0.001$) was observed at 16 than 30 days. Across both ages, enriched birds showed more wing flapping ($P=0.016$), wing stretching ($P=0.002$), body shaking ($P=0.002$), ground scratching ($P<0.001$) and ground pecking while both standing ($P<0.001$) and lying ($P=0.003$) compared to control birds. Even when no enrichments were close, enriched birds showed more body shaking ($P=0.008$), and ground pecking while standing ($P<0.001$) and lying ($P=0.010$) than birds in comparable locations in control pens. There was a tendency for a lower gait score (i.e. reduced lameness) in the enriched treatment ($P=0.077$). In general, the combination of peat, bales of lucerne hay and elevated platforms increased specific exploratory, locomotory and comfort behaviors. Interestingly, even when no enrichments were close to the observation patch, enriched birds still showed more activity in the form of ground pecking and body shakes. In conclusion, enriched birds showed higher levels of several activities than control birds, also in areas where no enrichments were present.

Keywords: Behavioral activity, Broiler, Enrichment, Lameness

‘European Broiler Ask’ – The new era for poultry welfare

Abstract ID: 340

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Animal welfare is a subject that is growing in the poultry industry’s agenda year on year, mainly due to an increasing demand for transparency from consumers. Animal welfare groups have a great part in increasing consumer awareness, but also in raising the animal welfare agenda with food companies. One strategy animal protection groups have adopted to increase awareness is to form coalitions where animal welfare organizations engage to make a stronger case for animal welfare. First in the USA and more recently in Europe, animal welfare groups have formed a coalition to produce a statement for higher welfare broiler production. In Europe, the coalition already has 25 animal protection organisations participating. The statement covers five key issues, identified by the coalition as priorities for the improvement of broiler welfare: 1) a stocking density of a maximum of 30 Kg per square meter; 2) a switch to breeds with measurable, higher welfare outcomes; 3) environmental improvements including 50 lux of natural light, provision of enrichment materials and no cages nor multi-tier systems; 4) adoption of multi-step controlled atmosphere stunning systems or electrical stunning without live inversion and 5) the use of third party auditing to demonstrate compliance. The statement was built based on science and coalition members’ agreement on what the most pressing issues for broiler welfare are. One of the main outcomes of the coalition was to unify the call to improve broiler welfare ‘from farm to fork’, avoiding a multitude of slightly different requests coming from the different animal welfare groups to food companies and consumers. Over 80 companies in the US and in Europe already have commitments in line with the coalitions’ statement. This work aims to explore the science behind the criteria of the statement, the impact it had on the US and the first results in Europe. In addition, the paper will discuss the strategies companies are adopting to respond to the coalition proposal and the challenges at supply chain level.

Keywords: Industry, NGO coalition, Poultry welfare

Evaluating environmental enrichment for broiler chickens using the transect method

Abstract ID: 33

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Environmental enrichment (EE) consists in additions and modifications to production environments that facilitate animals’ biological adaptation improving animal welfare. We hypothesized that peat moss, wood shaving bales and perching structures (boxes and perches) would have EE effects on broiler welfare indicators assessed during on-farm transect walks and at slaughter. Twenty-four Ross 308 broiler flocks received one, two or three EE types, while six were raised with no EE. At 28 (±2) days of age, the transect method was used to assess frequencies (%) of immobile, lame, sick, small, dirty, and dead chickens and frequencies of chickens with featherless areas, tail wounds and other wounds (head and back wounds). Data on first-week mortality and total mortality were also collected for each flock. The effects of underfloor heating (UFH), windows and lighting programme (18 h continuous vs 16 h intermittent) were included in the analysis along with the initial stocking density and body weight at 28 days. An equivalent model was used for slaughter outcomes, that included foot pad dermatitis (FDP) score, frequencies (%) of total condemnations (TC) and condemnations due to heart problems (CHP), liver problems (CLP), perosis (CP), wounds (CW), emaciation (CE), and dead on arrival (DOA). Preliminary results showed that the frequency of dead was lower in flocks with wood shaving bales ($P<0.05$), UFH ($P<0.05$), and an intermittent lighting programme ($P<0.05$). The frequency of immobile chickens was lower in houses with windows ($P<0.05$) but higher in flocks with perching structures ($P<0.05$). Regarding slaughter outcomes, the incidence of CHP was lower with the use of wood shaving bales and in houses with windows, whereas a higher incidence of DOA was observed in houses with windows. Lower stocking densities were associated with a lower incidence of CW. In addition, positive correlations were detected between the incidence of small birds and first-week mortality ($P<0.001$), total mortality ($P<0.001$), and CE ($P<0.001$). The incidence of immobile birds was positively correlated with first-week mortality ($P<0.01$), total mortality ($P<0.01$) and CW ($P<0.01$), while tail wounds were correlated with CP ($P<0.01$) and CW ($P<0.01$). In general, wood shaving bales, UFH, intermittent lighting, and lower stocking densities positively affected on-farm welfare and/or slaughterhouse outcomes. Windows had both positive and negative effects, peat had no detectable effects on the measured indicators, and perching structures had a negative effect. Moderate to high correlations between on-farm and slaughterhouse variables indicated that the transect method was useful for predicting slaughterhouse outcomes.

Keywords: Broiler, Environmental Enrichment, Transect, Welfare

Evaluation of development of foot pad dermatitis in Pekin ducks under commercial conditions

Abstract ID: 39

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Foot pad dermatitis (FPD) is an important animal welfare concern in poultry husbandry systems. Its occurrence, causes, scoring methods and its suitability as an welfare and management assessment tool are very well described in broilers and turkeys, however, very little is known in Pekin ducks as a water fowl. The aim of the current study was to determine the onset and development of foot pad alterations in Pekin ducks. Therefore, occurrence, areas of occurrence (metatarsal foot pads, toe pads, webs) and development of severity of foot pad alterations were investigated during rearing and fattening period on two commercial farms. Data collection was performed consecutively over four production cycles at each farm. In total, 6,700 ducks were sampled randomly, whereas data was collected on six sampling days per farm and cycle. Over all farms and cycles, first symptoms of FPD in terms of hyperkeratosis occurred at the toe pads (2nd, 3rd and 4th toe) 4 days post-hatch. Additional alterations occurred 9 days post-hatch at the metatarsal foot pads. At the end of the fattening period 89 and 82 % of sampled ducks showed alterations at the metatarsal pads and pads of the 3rd toe, respectively. However, only 0.3 % of ducks showed severe alterations at the end of fattening and no duck showed alterations at the webs. Conditions of foot pads were affected by season and amount of dispersed bedding material (straw). Occurrence of FPD did not differ between seasons, however severity of FPD alterations was higher ($P \leq 0.05$) during winter season (December – March) when compared to summer season (June – October). Regardless of season, foot pad health was worse in ducks reared with < 2 kg straw per duck and production cycle than in ducks reared with > 2 kg straw ($P \leq 0.05$). The results of the current study indicate, that slight lesions can occur at an early stage of rearing and localization and severity of alterations change over time. In order to address animal welfare aspects, continuous evaluation of the status quo of foot pad health in the flock is highly recommended even at an early stage of production as certain management measures may need to be adopted to promote and maintain food pad health. However, further investigations are needed to identify and understand risk factors and pathophysiological mechanisms that influences the occurrence of FPD in Pekin ducks.

Keywords: Duck, Health, Monitoring, Welfare

Feeding Black Soldier Fly larvae to laying hens: effects on production performance

Abstract ID: 474

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An experiment was conducted using 400 Bovans Brown layers to determine effects of feeding Black Soldier Fly (BSF; *Hermetia illucens*) on production performance. BSF larvae were fed alive to hens from 20-36 wks of age, additionally to unlimited mash feeding. Four feed treatments were used, with 20 units having 20 hens each. Adult larvae were either provided daily in the feed bin (24 g; T1), spread daily in the litter (24 g; T2), not being fed (T3), or crawled out through the slatted floor after development in the manure (T4). For the latter, 4-5 days old larvae (about 70 g) were spread in manure, twice a wk. Eggs were collected daily to determine laying rate (hen-day egg production (HDEP)) and egg weight. Feed intake was measured weekly. At 36 weeks of age, hens were individually weighed and uniformity (% of hens that was within 10% of average weight) was determined. Finally, cumulative mortality was determined. For analyses of feed intake, egg weight, and feed conversion, mixed models were used, with treatment and period (4 periods between 20-36 weeks), and their interaction, as predictors. For single measures, such as analysis of HDEP, a one-way ANOVA was used. Data was analysed using SPSS 23.
Uniformity, feed intake, hen weights, egg weights, and cumulative mortality were not affected by treatments. However, T4 hens had a higher feed conversion ratio of 2.45±0.11 and 2.23±0.10 kg feed/kg egg, respectively, in the first 3 and last 4 weeks of the experimental period (treatment x period interaction: $P=0.016$). Averages for the other treatments in these periods were relatively high and ranged between 2.79-2.99 and 2.39-2.63 kg feed/kg egg, respectively. T4 hens also had the highest HDEP (94.59±1.74%), together with T1 (93.54±1.74%) and T2 hens (93.56±1.74%), compared to T3 hens without provision of larvae (86.39±1.56) (treatment effect: $P=0.011$). This could partly be explained by eating of eggs in T3: additional observations indicated a higher risk for occurrence in T3 hens (80%), whereas this was 20% in T1, 50% in T2, and 0% in T4 hens.
To conclude, BSF larvae supplementation had a positive effect on production performance. Likely this was due to an increase in welfare, especially by a reduction in undesirable pecking, rather than being a nutritional effect. This seems especially true in T4 hens, having the best feed conversion, combined with best foraging conditions. The research was funded by the Centre of Expertise Agrodier.

Keywords: Black soldier fly, Larvae, Laying hens, Production performance, Welfare

Genetic diversity – Potential for diversity in adaption and productive quality among chicken breeds

Abstract ID: 344

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The advantages of heritage breeds in modern food production might not be obvious but could offer alternatives in terms of animal welfare, food quality and regional identity. On the other hand, the use of day-old male chicks from layer lines for meat production gains ethical and welfare-associated interest in the public. We tested three genetics - Lohmann Dual as dual-purpose hybrid, Lohmann Brown as layer hybrid and Rhinelander as traditional breed - for their potential in terms of animal welfare, behaviour and meat production. Therefore, we raised 844 Lohmann Dual, 714 Lohmann Brown and 458 Rhinelander conventional and recorded their feed consumption, weight gain, mortality and evaluated their scores according to the Welfare Quality® assessment protocol for poultry. Sexes were separated with 10 weeks of life, concurrent with the first date of slaughtering. Remaining cocks were fattened until 20 weeks of life. We found no evidence for any welfare-related morphological changes, beside the Rhinelander whose feet were conspicuous beginning with 15 weeks of life. Fattening performances revealed that Lohmann Dual show a daily weight gain between 28g (day 43) and 36g (day 71; $P \leq 0.001$), Lohmann Brown show values between 16g (day 43), 21g (day 71 and 134; $P \leq 0.001$), and Rhinelander show 13g (day 43) and 18g (day 71 and 134; $P \leq 0.001$). For the latter, a prolonged fattening period up to 20 weeks would accommodate their slower growing rates whereas the peak performance of Lohmann Dual is reached around day 71. European Production Index was highest for Lohmann Dual at day 71 (165 points) and lowest for Rhinelander (28 points at day 134). Rhinelander tended to be more insecure ($P \leq 0.01$) and fearful ($P \leq 0.000$) during the qualitative behaviour assessment than Lohmann Brown or Lohmann Dual. Adaptive behaviours might have been favoured during intensive breeding which is not applicable to heritage breeds in general. We conclude that the fattening of male layers as well as of heritage breeds is possible with surprisingly good carcasses but far away from any productive efficiency. Comparable slow growing broiler lines strike with 305 points e.g., so dual purpose reach approximately 50% of performance points in twice the time. These breeds might meet modern requirements of ethics as well as the idea of animal production in the public. Based on their performances, these non-broiler breeds will have to find real fans in order to find their production niche.

Keywords: Animal welfare, Dual-purpose, Fattening performance, Heritage breed, Layer

Hens in mixed meat turkey flocks separated by wire fence from toms showing high mortality due to histomonosis are infected based upon laboratory diagnostics

Abstract ID: 261

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Histomonosis, caused by the protozoan parasite *Histomonas meleagridis*, is a severe disease which may lead to high mortalities in turkeys. However, outbreaks are described in which hens, separated by wire fence, survive the disease despite high mortality in toms. The present study investigated environmental dust, serum samples and cloacal swabs from turkey hens and toms raised in mixed houses. Investigations were performed on three different meat turkey farms in which outbreaks started in the 4th week of life. Initially, pathognomonic caecal and hepatic lesions were observed in dead turkey toms and/or hens. Histomonosis was further diagnosed by histopathology and/or 18S rRNA real-time PCR using tissue samples from dead birds. In turkey toms mortality went up to nearly 100% within two weeks and the rest of the toms were euthanized. In contrast, turkey hens showed no significant increase in mortality and were kept until slaughter at 10 to 12 weeks after the start of the outbreak. Dust samples collected within compartments of hens and toms confirmed the presence of *H. meleagridis* DNA within the whole houses up to 6 weeks after the first signs of histomonosis. Serum samples were investigated with an indirect *H. meleagridis* sandwich ELISA. Within two weeks post initial diagnosis and prior to euthanasia up to 73% of the investigated toms developed antibodies. Within the same period of time also up to 70% of the investigated hens were positive but much lower OD-values were noticed. Until the end of the investigation the number of positive hens per farm increased up to 100% with mean OD-values approaching the levels noticed in toms prior to euthanasia. Only a very limited number of cloacal swabs were found positive which might be due to the intermittent excretion of the parasite. For the first time it could be demonstrated that turkey hens kept in the same house as turkey toms got infected during fatal outbreaks in toms. This is somewhat different to experimental studies in which usually no differences between sexes are reported. However, the underlying mechanism of the phenomenon needs to be addressed in future studies.

Keywords: ELISA, Histomonas meleagridis, Histomonosis, PCR, Turkeys

Identification of risk factors and prevalence of injuries at different stages of the broiler slaughter process

Abstract ID: 102

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Transport and handling of broilers during the (pre)slaughter process are risk factors for welfare. The impact of preslaughter treatments on injuries and thus the effect on welfare is poorly known. Moreover, it is unclear which proportion of carcass damage can be attributed to the conscious phase, and affects welfare, and which proportion of carcass damage can be attributed to handling after stunning and killing of birds and is related to product quality. We first analysed routinely collected data of a Dutch slaughter plant to identify risk factors for carcass damage. It was included whether or not prevalence of carcass damage was related to flock welfare status. Data collected in 2014-2016 from five farms with low foot pad dermatitis (FPD) score (<40 points) and five farms with high FPD score (>80 points) were analysed (N=771 flocks), assuming that FPD score was indicative of flock welfare status. A regression model was applied. The model showed positive associations between live body weight (P=0.000; B=0.001), number of dead-on-arrival (P=0.001; B=0.004), and wing damage. A negative association was found between wing damage and number of broilers per tray (P=0.037; B=-0.015). No relation between FPD score and carcass damage was found. Second, prevalence of injuries or damage during the slaughter process was determined in 20 flocks. Wing, leg and breast bruises, wing dislocations, and wing and leg fractures were scored between lairage and post-plucking. An increase in wing fractures from lairage (0,99%), post-shackling (1.67%), post-stunning (2.73%) and post-plucking (5.02%) was found (P=0.000 between all stages). Medium and large breast bruises increased between lairage and post-plucking (P=0.000). Small wing bruises decreased between lairage and post-plucking (P=0.047). This study showed that flock welfare status was not related to injuries, and that weight and crate density could be identified as risk factors for wing damage. Injuries and damage do mainly occur during the slaughter process. However, it was difficult to determine whether or not carcass damage originated from handling live animals, and thus is an animal welfare problem, or after stunning and therefore is a product quality issue. It is advised to develop an accurate bruise and damage scoring system that can be used to determine whether or not carcass damage was caused in live animals.

Keywords: Broiler, Handling, Injuries, Slaughter, Welfare

Implementation of project results for the rearing of pullets under practical conditions

Abstract ID: 156

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The evaluation of a scientific project on the welfare of laying hens revealed, that the factor "feather pecking at rearing" had a significant influence on "feather damage at layer farm" and the factor "injuries at rearing" had a significant influence on the "injuries of adult laying hens at the layer farm". Even though, the rearing of pullets is one of the major aspects for feather pecking and injuries in adult layers, no legal regulations on the rearing of pullets (e.g. stocking density) exist in Germany. However, guidelines with recommendations for the prevention of feather pecking and cannibalism in layers and pullets have been presented by the German state Lower-Saxony and the German Federal Ministry of Food and Agriculture. These also consider the rearing of pullets. Core requirements are a stocking density of 18 pullets/m^2 usable area, 36 pullets/m^2 usable ground area and 54 pullets/m^2 litter area from the day 35 of rearing onwards. Own research and experience on farm showed, that the implementation of these recommendations, especially regarding the stocking density, is in some cases not possible without substantial rebuilding or remodeling of the housing systems. The recommended stocking density of 18 pullets/m^2 usable area was considerably exceeded on several farms (up to 34 pullets/m^2 usable area from the 35th day of rearing onward). However, it was also observed, that even on farms with 18 pullets or less per m^2 usable area, the stocking density regarding the usable ground area was exceeded. This is due to the fact that construction and setup of aviaries in barns with narrow corridors lead to an increase of stocking density regarding the ground and litter area. Rebuilding or remodeling of aviaries can be associated with limitations or deterioration of animal welfare. Consequently, pullets might have to be housed in the cages longer without access to litter or the presence of an increased risk of limited accessibility to resources, if cages are opened too early. It should be discussed which systems used under practical conditions fulfil both, the ethological needs of the pullets (litter from the first day of life) and legal requirements (pullets getting used to the housing system).

Keywords: Practical conditions, Pullets, Rearing, Stocking density

Influence of different environmental enrichment programmes on behaviour and utilisation of floor space of pullets and laying hens kept on commercial farms

Abstract ID: 484

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Feather pecking and cannibalism are the most common behaviour disorders in laying hens. Deficient husbandry- and management conditions, both in terms of rearing as well as in the later laying period, particularly the lacking satisfaction of the execution of the characteristic foraging- and food intake behaviour are considered to be the cause. Environmental enrichment (EE) seemed to have positive effects on behaviour and welfare in different projects and on several farms. Although the suitability of EE to prevent behavioural disorders, like feather pecking and cannibalism on commercial farms is largely unresearched.
Therefore, this project was supposed to provide video-based evidence for an increase in the utilisation of the floor area by deployment of various EE during the rearing- and the laying period. The 92.000 animals (Lohmann brown) of the study were housed on a commercial rearing and production (laying) farm with an aviary system, monitored throughout an entire breeding- and laying period.
To examine the impact of a supply of diverse EE on the animals's behaviour, there were five groups supplied with different EE programmes, with a repetition of eight (rearing, 2545/1820 birds per flock), as well as four flocks (laying period, 4611 birds per flock) per group. One group had access to lucerne bales, another was provided with pecking blocks, a third group was fed with wheat grains daily, the next group was given both pecking blocks and wheat grains, while the last group served as a control group, without any EE. Animals in a defined area were counted on video stills and average values of animals per square meter were calculated. Video analysis of the usage behaviour in the rearing and the laying period (mean value of all five dates) shows, animals with EE programme used the floor space more intensively (group with lucerne as EE 7.31 birds/m^2, with pecking blocks and wheat grains 6.85 birds/m^2, with only wheat grains 6.14 birds/m^2, with only pecking blocks 5.80 birds/m^2) than the hens of the flocks without EE (control group 4.58 birds/m^2).
Deployment of various EE could increase the utilisation of the floor space during rearing- and the laying period. Therefore, different EE materials could help enable the expression of natural behaviour with hens kept on commercial farms.

Keywords: Cannibalism, Environmental enrichment, Feather pecking, Laying hen, Pullet

Organic fattening of turkeys: investigations on animal welfare

Abstract ID: 42

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As organic fattening of turkeys differs from fattening turkeys under conditions of traditional farming, (e.g. restriction of work with feed additives containing amino acids, prohibition of routine beak trimming, lower stocking density and access to free range area), these altered conditions may have an influence on animal health and welfare. Therefore, in the presented study animal welfare indicators were investigated during rearing and fattening of turkeys reared under organic poultry farming system. The design of the study was similar to previous examinations in conventionally reared turkeys presented at former conferences. The project was financially supported by the German Federal Ministry of Food and Agriculture through the Federal Scheme for Organic Farming and Other Forms of Sustainable Agriculture (BÖLN). Nine rearing farms and 15 fattening farms were visited at five specific time frames (3rd-5th; 35th-42th; 70th-77th and 106th-112th fattening day, and one visit between 10th-17th day after the turkeys were placed in the fattening barn [5th-6th week of life]), this scheme was repeated in a second period. The data of 1,860 turkeys (1,320 hens, 540 toms) were collected in the study. Flocks with Kelly Bronze BBB (both sexes) and B.U.T. turkeys (hens) were examined. At each visit on the farms 60 randomly picked animals of each sex were examined concerning different health parameters with focus on pecking injuries, foot pad condition, and breast skin alterations. Information about flock health and management was evaluated by standardized questionnaires. In addition litter samples from defined areas and feed samples were collected to be analyzed in lab. This was done with the purpose to investigate the influence of litter (especially humidity) and feed (especially NSPs, crude protein and amino acid composition) on animal health and animal welfare. The results obtained will be discussed in comparison with those gained in other studies from conventional reared fattening turkeys.

Keywords: Animal welfare, Fattening turkeys, Foot pad dermatitis, Indicator, Organic poultry farming

Production performance of two dual-purpose chicken breeds in a mobile stable system

Abstract ID: 404

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Debates on culling day-old male egg-type chicks are constantly growing. Besides the in-ovo sex determination and the fattening of male chicks of layer lines, the use of Dual Purpose Breeds is considered as a strategy to avoid killing of day-old male egg-type chicks. The current study investigated the performance of two Dual Purpose Breeds in a mobile production system under organic conditions. 509 Lohmann Dual (LD) and 505 Lohmann Dual*experimental* (LDex) day old chicks were reared in two mobile stable barns as hatched. The cockerels were slaughtered on day 85, the hens remained in their respective mobile barn for a prolonged laying period (74 weeks, including molting). Body weight (BW) development, group feed consumption and mortality was recorded at regular intervals. At time of slaughter of the males, randomly selected birds (n = 30 / group and genotype) were dissected to determine slaughter weight (SW) and carcass composition. Additionally, laying performance (quantitative and qualitative) of the hens was recorded and evaluated in frequent intervals during the laying period. Average BW and SW of cockerels at d 85 was 2,650 (SD: 262.1) and 1,786 g (SD: 185.3) in LD and 2,176 (SD: 263.9) and 1,449 g (SD: 179.3) in LDex, respectively. Over the fattening period, average daily weight gain was 34.6 g in LD and 29.7 g in LDex. Percentage of breast muscle (16.3 %) and legs (32 %) did not differ between genotypes. In 68 weeks of production, average laying performance (excluding molting) was 62.2 % in LD and 66.9 % in LDex; an average LD hen produced 289 eggs whereas it was 307 eggs in LDex. Average egg weight during 68 weeks of production was around 60.5 g in both genotypes, whereas LD hens had a higher feed consumption per kg egg mass (X:1) when compared with LDex (2.9 kg vs. 2.4 kg). The performance of both Dual Purpose Breeds is not comparable with those of specialized hybrids. The use of such breeds may still be sufficient if the products realize higher prices at the market as their production is more expensive and less efficient when compared to specialized hybrids. However, when taking animal welfare and ethical aspects into account, the use of Dual Purpose Breeds may still be considered as one potential strategy to avoid culling of day olds.

Keywords: Culling, Free range, Organic, Welfare

Specification for Light Sources in Poultry

Abstract ID: 499

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Birds` eyes generally differ from human eyes, especially regarding spectral sensitivity and its ability to resolve temporally varying stimuli. Contrary to the bird's perception, the entire visual spectrum of a human being excludes the UVA range, whereas ultraviolet wavelengths contribute to brightness perception in birds. Furthermore human eyes are able to detect flicker between 20 and 60 Hz, whereas it is assumed that chickens eyes are able to detect flicker at higher frequencies (100 Hz). Public interest about welfare in domestic poultry draws attention to housing conditions, especially to lighting which plays a decisive role for the development of conduct disorders such as feather pecking and cannibalism. This is probably due to discrepancies between lighting systems and perception of poultry. It can be assumed that natural daylight corresponds most closely to the requirements of a bird. Therefore a number of new product developments of lighting systems are placed on the market which promise an equivalent spectrum to natural light and an absence of flicker. Estimation of natural light-like illuminants depends on knowledge about natural light relating to the spectral composition. Critical flicker fusion frequency has rarely been assessed in birds. Furthermore flicker fusion frequencies are varying because of its heavily dependent on light intensity. The ability to resolve temporally varying stimuli increases with rising light intensity. As a precaution it is advisable to strive for flicker rates in lighting systems including a buffer-range (160 Hz). Currently the fitting of these new natural light-like lighting systems generally doesn`t solve problems with conduct disorders. It seems that new lighting systems for poultry barns often do not correspond to expectations. Own results of spectral analysis and lamp flicker rates also revealed that the development of natural light-like illuminants with reliable spectral composition and absence of flicker seems to be difficult and needs further technical development work.

Keywords: Flicker, Lighting system, Poultry barn, Spectral composition

Spontaneous intake of essential oils after a negative postnatal experience in chicks and long-term effects on blood transcriptome

Abstract ID: 427

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The perinatal period is a critical period for broiler chicks as they are exposed to various environmental changes, possibly stressful, in the hatchery and during the transport to the breeding houses. We tested the ability of broiler chicks to drink spontaneously essential oils (EO) to mitigate the effects of negative experiences during this perinatal period. Chicks were either immediately placed in the rearing facility (Control group C), or submitted to a 24h-delayed period before their placement (D group) mimicking the possible transport delay in commercial conditions. In experiment 1, half of each group had only access to water and the other half to water and one EO (cardamom, marjoram or verbena) from day 1 to 13 of age. The verbena EO intake was higher in D group than in C group from day 1 to 6 of age. In experiment 2, half of C and D groups had only access to water and the other half was offered water and the 3 EO simultaneously. The marjoram EO intake was the highest compared to the other EO from day 1 to 6 of age whatever the treatment. In this experiment, D group chicks drank less verbena EO from day 1 to 2 of age and the intake of each EO increased with age, except for verbena intake in C group.
EO intake did not allow overcoming the reduced growth in D group but tended to overcome the reduction of the relative percentage of *Pectoralis major* muscle.
Transcriptome analysis of blood cells showed long-term effects of the delay to placement on gene expression at slaughter age. Differential expression of transcription factors in D group was related to oxidative stress in males. EO intake limited the overexpression of several genes involved in oxidative stress and inflammation in D groups.
In conclusion, chicks are able to make spontaneous choices for EO according to their postnatal experience. These choices had a few beneficial effects on their performance, and long-term regulatory effects on blood transcriptome, which could participate to maintain chicken welfare and health.
Work funded by the Integrated Management of Animal Health metaprogram of INRA for the 'GISA – WHELP' project (www.gisa.inra.fr/en).

Keywords: Blood transcriptome, Essential oils, Health, Negative Postnatal experience, Welfare

Temperature training in the hatcher – effects on robustness, performance and production efficiency in dual- and laying-type cockerels

Abstract ID: 64

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Routine culling of one day old male laying-type chicks is an ethical problem. The use of dual purpose chickens for egg and meat production and breeding of laying-type cockerels may provide an alternative. To improve breeding economy alongside with robustness pre-hatching temperature training (PTT) with short-term warm stimulation (plus 1°C above standard for 2 hours daily) was used. In broiler chickens PTT has long lasting effects on thermal adaptability and various body functions (e.g. reduction in energy metabolism). Hence, PTT experienced chicks have more energy available for the primary performance as well as for adaptation, immune and stress responses, which meets important animal health and welfare but also economical aspects. 2880 eggs (Lohmann Brown-LB/Lohmann Dual-LD) were incubated from day 1 to 17 under normal incubation temperature (37.2-37.4°C) and until hatching in two groups with different temperature programs (control: 37.2-37.4°C / PTT). For a subsequent 70 days growing period 320 one-day-old cockerels (160 LB/160 LD) from each incubation group were randomly distributed in 8 treatments (8 pens/treatment; 10 birds/pen) resulting from two origin of cockerels, two incubation programs, and two dietary protein/energy levels (200 g crude protein/11 MJ AMEN/kg – low level, LL; 215 g/12 MJ AMEN/kg – high level, HL). PTT had no negative effect on hatching results in LB and LD cockerels. In LD, but not so pronounced in LB cockerels, performance and production efficiency were significantly improved by PTT. In LD cockerels, for instance, the final body weight on day 70 achieved in the LL and HL feed group 2558 and 2528 g, respectively (control: LL 2432 g/HL 2482). From day 1 until 49, the lowest feed to gain ratio ($p<0.05$) of 1.81 kg/kg was calculated for the PTT group of LD cockerels fed with HL feed. Slaughter after 70 days showed significant differences in breast meat and legs, and percentage of abdominal fat between the bird's origin and of the different incubated groups. While the thyroid weight was influenced by the male's origin only, the bursa weight was depended on origin of birds and experienced PTT at the age of 70 days. Possibly, robustness may benefit from PTT in both, LB and LD cockerels, which were indicated by increased mean relative bursa weight of 18 and 21%, respectively. In conclusion, PTT can be a practicable tool to improve robustness alongside with production efficiency in laying-type and dual cockerels.

Keywords: Dual chickens, Incubation, Laying-type cockerels, Performance, Robustness

The effect of different eggshell temperature patterns during incubation on broiler chicken behavior determined by an automatic tracking system

Abstract ID: 194

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In the last decades, it has been generally assumed that a continuous eggshell temperature (EST) of 37.8ºC throughout incubation optimizes hatchability and hatchling quality. However, recent findings using broiler eggs suggest that a higher EST (38.9ºC) during the second week of incubation or a lower EST (36.7ºC) in the third week of incubation may improve hatchling quality even further. The combination of a high and a low EST during incubation has not been investigated before. Furthermore, consequences of EST patterns during incubation on behaviour post hatching such as activity and anxiety has also never been investigated to our knowledge. The aim of this study was therefore to evaluate effects of EST patterns during incubation on behaviour post hatching of broiler chickens using an automatic tracking system. Eggs of a Ross flock were incubated in a 2x2 experiment design with a high EST (38.9ºC) or normal EST (37.8ºC) in the second week of incubation and a low EST (36.7ºC) or normal EST (37.8ºC) in the third week of incubation. Post hatching, five males and five females were housed in a 2 m^2 pen with 8 replicates per EST treatment and reared until day 42 of age (n=320). At day 7 and 34 of age, an open field test was performed to evaluate anxiety by social isolation in a novel environment (n=128). At day 29 and 40 of age, activity of the broilers was evaluated using TrackLab (Noldus, Wageningen, The Netherlands; n=64). Chickens of four pens were transported to a PhenoLab test room and placed in 2 m^2 pens. Measurements started after 15 minutes of acclimatization. The ultra-wideband tracking system TrackLab consisted of active tags that were placed in backpacks on 2 males and 2 females per pen. The location of each bird was measured twice every second by triangulation of the received signal between four beacons that were placed in every corner of the test room and was based on the time and the angle of arrival of the signal. Data of the x,y location of the chickens was obtained by TrackLab and distance moved (cm) and speed (m/s) from individual chickens of all four EST treatments were calculated. Results of this study are currently processed and will be presented at the conference.

Keywords: Behaviour, Broiler chicken, Incubation, Tracking

The point and purpose of rearing male chicks

Abstract ID: 22

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The issue of killing male chicks is surrounded by much emotion, which is why the economy has been looking for solutions for a long time. Embryonic early detection is an important approach, but it involves considerable effort and expenses. The results have not been effective so far. On the one hand, it is criticized by animal welfare organizations, and on the other, commercial selection requires extensive effort and costs. However, there are alternatives in the field of organic production. The market is structured differently. Consumers have different expectations in terms of animal welfare requirements, the origin of organic products, and price. Ideologically, the group of buyers is quite willing to pay more for organically produced products. So-called dual-purpose chickens are one way of fulfilling NGOs' and consumers' animal welfare expectations. This form of chicken keeping is also expensive because of lower laying performance and significantly poorer feed conversion compared to conventional broiler fattening. Both products, eggs and meat, must be marketed, which often leads to limitations in terms of food retail trade (LEH) specifications. Many organic producers are therefore increasingly concentrating on rearing male chicks. There are no legal criteria for rearing yet. The interest group Bio-Initiative (IG Bio) has therefore set binding standards based on environmental principles. This means, for example: requirements for barn equipment, stocking density, perches, usable areas, water, food, and range. Although the production costs for this are higher, the appeal to emotion offers better sales opportunities in the food trade and especially in convenience. Bio-Initiative has obliged participating companies to stop killing male chicks and make 100% of their output available for fattening until the end of the year. This utilization also makes an important contribution to animal welfare. The aim of this presentation is to present the point and purpose of raising male chicks and to evaluate the requirements of standards from a business and market economy perspective. Furthermore, the marketing concept plays a decisive role. The significantly higher rearing costs must be compensated and achieved through marketing strategies. The so-called broiler rooster concept is intended to serve as an example. Specifications for neutral control along the process chain, traceability, and producer identification must also be assured. This particularly applies to the origin of the chicks in order to prevent manipulation.

Keywords: Bio-Initiative standards, Broiler rooster concept, Embryonic early detection, Environmental principles, Marketing

The use of pheromones in laying hens in aviary housing system

Abstract ID: 200

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Feather pecking is an important welfare and economic problem in laying hens. The development of this behavior is multifactorial and the large group housing (enriched cages and aviaries) makes it difficult to control. The first weeks in the layer house are known to be a stressful period for the hens. Tools to reduce the stress level can prevent or delay the start of feather pecking within a group of hens. Pheromones already showed promising results to reduce stress in different species. In this study, the effect of the systemic analogue of MHUS (Mother Hen Uropygial Secretion) was examined in aviary housed hens. A double blind study with MHUS analogue was performed with 10300 hens (Isa Brown) in 4 identical aviaries. Two groups received the active product while the 2 control groups received a placebo. Treatment started on the day before arrival of the hens and continued until 41 weeks of age. Production data were recorded, a flock behavior checklist was performed and feather cover was assessed every 4 weeks together with keelbone evaluation. In general, until 41 weeks of age, hens performed well. However, already small differences were detected between the 2 groups at this age. Hens that were exposed to the MHUS analogue showed lower mortality rates compared to hens without MHUS (resp. 0,6% 1,2%) and slightly higher laying percentage (resp. 93,8% and 92,9%), egg weight (resp. 63,1g and 62,7g) and higher feed intake (resp. 118,5 and 116,7g/hh/day) compared to the control groups. A trend towards better feather cover and less keelbone abnormalities was detected in the treated hens compared to the control groups. The use of the MHUS analogue in laying hens appears to give promising results. Further work is needed to evaluate the effect on both rearing hens and on end of lay hens in order to investigate if the MHUS analogue could help the sector in prolonging the laying cycle without feather pecking.

Keywords: Feather pecking, Pheromones

To catch or not to catch? That is not the only question in an educational program for catchers

Abstract ID: 45

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Catching is an inevitable part of the preslaughter chain for broiler chickens and may evoke stress and fear. Therefore, the fitness of the animals prior to catching is extremely important. In addition, physical injuries and pain may be inflicted, and previous studies found traumatic wing fracture in approximately 1 % of manually caught broilers. Catching related injuries may also lead to mortality. A Norwegian study showed that 25 % of dead-on-arrivals had a fatal trauma; e.g liver ruptures with bleeding to the abdomen and fractures to the skull and vertebra. In Norway, ethical audits are used as internal animal welfare revision at all major poultry abattoirs. The thorough revisions investigate the entire pre-slaughter chain, from farm prior to catching, during transport and through the stunning and killing process at the slaughter houses. The audits, along with the above-mentioned studies, have revealed that there is a need for more and better education for catching personnel. Education must focus on the fitness for transport e.g. is the animal healthy and without injuries? The catchers must be able to distinguish between fit animals that can be caught, and unfit animals that should not be caught. In addition, catching technique, handling of live birds and the correct way to stun and kill broilers on farm are essential. Currently, the development of an extended education program is in progress in Norway to meet these challenges. The education will consist of an e-learning course for catching-team leaders, and a one-day classroom education for catchers, team leaders and drivers. The education will incorporate both catchers and drivers, since both actors may contribute to enhance welfare during the catching process. In addition, animations as a teaching tool are developed. These will focus on the most basic "does" and "don'ts" in training of new catchers. The animations are short, easily available and meant to be used prior to entering the farm, for catchers with no previous catching experience. Due to language challenges, the animations speechless. The effect of this educational program will be evaluated later through ethical audits and registration of wing fractures on the slaughter line. The program will be adjusted if the effect is lower than expected.

Keywords: Broiler, Catching, Education, Pre-slaughter chain, Welfare

Use of perches and grids during light and dark period by growing chickens differing in growth intensity

Abstract ID: 359

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In commercial housings meat chickens most often are kept in deep litter systems equipped with drinkers and feeders but without any other structures. By offering most often either perches or grids at a certain height several studies indicated that also meat chickens are motivated to rest on elevated structures. In the present study we systematically tested the usage of perches and grids in relation to their height, the growth intensity and age of chickens, and time of day. In two trials 400 Ross 308 (Ross: fast growing), 400 Lohmann Dual (Dual: medium growing), and 400 Lohmann Brown Plus (LB: slow growing) chickens were kept in groups of 50 birds of the same strain in eight compartments (2x3m), respectively. Each compartment was equipped with both plastic perches and grids at three different heights (10, 30, 50 cm) and a ramp in between. Single-phase pelletized feed and water were available *ad libitum*. After the first three days the stable was artificially lightened from 04:00 am to 08:00 pm. Number of birds on elevated structures were recorded by an infrared video camera and their usage was analyzed for two days each week of live by time-sampling in 30 minutes intervals. Effect of the type of elevated structure was tested separately for daytime and nighttime with pooled data from all strains; effect of weeks of age, height of structures and their interaction was tested separately for each strain. All analyses were done with Generalized Linear Models (GLM) and compartment number was included as random factor in each model. All three strains showed a preference for grids during both light periods and dark periods except for LB at nighttime throughout the entire observation period (all $P<0.01$). At the end of the fattening period chickens used the structures at 50 cm more frequently than those at 10 or 30 cm during dark periods ($P<0.05$). At light periods all three strains showed an increasingly usage with increasing age except Ross which showed a decrease of usage from the fourth week. During dark periods, however, none of the strains showed a decrease of usage. Results indicated that meat chickens differing in growth intensity are motivated to use elevated structures. Especially for chickens with fast and medium growth grids are more suitable than perches and should be offered to meet their behavioral demand for roosting.

Keywords: Elevated structures, Growing chickens, Growth intensity

Value creation by a novel ceiling-based mobile broiler monitoring system (“chickenboy”) for improving animal health, welfare and productivity

Abstract ID: 409

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Citizens and consumers are increasingly worried about the living conditions of intensely farmed chickens and the health and welfare problems in crowded broiler houses such as diarrhoea, leg and foot diseases, pododermatitis, poor air quality, respiratory diseases and other health and behavioural issues. Most of these problems are recognized rather late because of the low surveillance frequency in large broiler herds and the impossibility to monitor chickens permanently and individually under current production conditions. This paper reports about a new mobile livestock robot (called “ChickenBoy”) which runs permanently on ceiling-based rails and monitors continuously and in real time the birds, faeces and litter conditions, food and water supply by visual and infrared cameras, temperature, relative humidity and CO2 (NH3 optional) close above the heads of the animals. The system can identify dripping nipple drinkers, anomalous faeces structures, wet litter areas and immobile and dead birds. All data are stored in a protected, cloud-based system, mapped across the barn floor and presented to farmers via PC or smart phone. This enables them to survey animals continuously and at any time. They can take early action in case of animal disorders or indisposition and the data mapping leads him directly to the points of technical failure or animal health or welfare problem in the barn. First experiences in broiler farms in Spain show the great potential of the ChickenBoy to improve the quality of life of broilers. The value of this new intelligent PLF (Precision Livestock Farming) technology is that it enhances the confidence of farmers in their production, increases understanding of animals, improves health and welfare of the animals, stabilizes the productivity and may create more confidence of consumers by the high transparency in the so surveyed broiler production. Estimates show that the system can become profitable after about one year already. The savings result from lower labour and health costs, lower mortality and higher productivity. A short demonstration video is shown. (www.faromatics.com).

Keywords: Broiler, ChickenBoy, Health, Precision Livestock Farming, Productivity, Welfare

Walking on tiptoes: Description of alterations of the digital pads as one indicator of foot pad dermatitis in turkeys

Abstract ID: 441

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Foot pad dermatitis (FPD) is an accepted indicator for animal welfare. Most scoring systems evaluating the severity of FPD, describe lesions of the metatarsal foot pad (FP). As there is little information in literature, the aim of this study was to describe lesions on the digital pads (DP) in turkey feet.
At the slaughterline, a total of 250 feet were scored by an automated camera system, providing a five-scale scoring level with rising severity (0-4), based on the size of the lesion on the FP. Digital pictures of these feet were used for further examination. Each DP was scored using a five-scale score to evaluate the size of the affected necrotic area (0-4, with rising severity) and a three-scale score was used providing information on the grade of swelling (0-2). Furthermore, the amount of affected digitals was evaluated. Descriptive statistics is provided for first data analysis. Feet which were expected to be intact according their scoring of the FP (n=50), were observed to show lesions on up to every digital, providing scoring levels for each state of severity in both, necrotic areas and level of swellings. No linear relationship between FP lesion and DP lesion was detected (i.e. rising severity in FP lesion score did not results in rising severity of DP lesion). Each DP parameter was found to decrease in severity from FP lesion score 0 to score 1, whereas severity increased from FP score 1 to score 2. The severity in DP lesions did not increase with rising severity in FP lesions (2, 3 and 4). Furthermore, using the Friedmann-Test, a significant effect of the individual digital in both, affected necrotic area and grade of swelling (both $p<0.0001$) could be shown. The severity of necrotic areas was highest in the exterior digit, whereas the interior digit was most affected by swellings.
As a conclusion lesions of the digitals do not develop equivalent to the lesions of the metatarsal foot-pads. Even between digitals seems to be a difference in pathogenesis. Therefore, scoring systems, rating the severity of FPD should include alterations of the digitals as one further relevant indicator of animal welfare.

Keywords: Digital-pad, Foot-pad-dermatitis, Turkey

Welfare assessment of laying hens in four housing systems in Slovenia

Abstract ID: 217

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Factors such as diseases, skeletal health, behaviour stress, nutrition and genetics can influence the welfare of laying hens. In Slovenia, no assessment of commercial layers has been conducted focusing on welfare indicators. The present study is the first to provide results of the indicators of Lohmann brown hens in enrichment battery cages (n=37.860), aviaries (n= 13.800), litter system with (n=3.820) or without outdoor access (n=4.420). Four flocks, each per housing system, were placed in laying farms at the age of 16 weeks and had the same feed supplier. Flocks were visited at two time points; at the beginning of the laying period at 22-24 weeks and at 50-55 weeks of age. At each visit, environmental conditions (i.e., temperature, NH_3, CO_2, relative humidity and air speed) were measured at three different locations within the barn. Feather condition, keel bone deformities and foot pad lesions were scored in 100 randomly selected hens per flock. Hens' behaviour was scored in the novel object and the avoidance distance test following the procedure described in Welfare Quality® protocol. Additionally, pathological examinations were performed and production results were compared.
At the first visit, selected environmental parameters revealed good and comparable conditions in barns with aviaries and enrichment battery cages systems. In both houses with deep litter system, the level of NH_3 exceeded 20 ppm, but no respiratory disorders, eye pathology or foot pad lesions were confirmed. Keel bone deformities were detected in enrichment cage system (8 %) and in aviaries (1 %), but not in hens kept on litter. At the second visit, the highest score of keel bone deviations was confirmed in aviaries (67 %), followed by battery cage system (48 %), in litter system with or without outdoor access much lower percentage of hens with keel bone damage was identified (6 %) by palpation, although at necropsy multiple keel bone fractures and deviations as well as osteoporosis were confirmed in dead or culled hens from all systems. Higher feather scores were observed in hens kept in enrichment battery cages (1.50) and aviaries (1.08) compared to those on litter with (0.6) or without outdoor access (0.18). Furthermore, at both visits, hens' behaviour tests showed that hens kept in aviaries were least fearful animals. Production results did not differentiate between systems, although in the litter system with outdoor access significantly better egg shell strength was confirmed

Keywords: Different housing systems, Laying hens, Welfare assessment

ORAL PRESENTATIONS

Physiology

Amniotic feeding of flavanonein last week of incubationon, tibia growth plate and long bone mineralisation of hatched chick

Abstract ID: 622

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The effect of ovo injection of phytoestrogen, flavanone (Naringin & Hesperidin), a source of citrus flavonoids, was investigated on day-chick epiphysial growth plate histology of tibia zones (reserve, proliferation, pre-hypertrophic, hypertrophic, early calcification or mineralisation), long bone Ca and P content and blood serum alkaline phosphate (ALP) status. Fertile Ross eggs (n=700), obtained from a commercial broiler breeder company Karaj/Iran, were weighted and distributed into 7 groups of 100 eggs. On 14th and 17.5th days of incubation, 4 groups were injected via amnion sac of low and high levels (15 or 30 mg) of Flavanone/ 0.5 ml saline /egg, respectively in 4 treatments. The other three groups were used as the sham controls (normal saline, 0.5 ml/egg) in 14th and 17.5th days of incubation and un-injected control. The right proximal part of tibia and the left femur and tibia were used for measuring the bone characteristics. The experiment was approved by the Animal Ethics Committee of the Tarbiat Modares University. The data were analyzed using the General Linear Model procedure of the SAS® and the means were compared by Tukey test at 5% probability. In ovo feeding of flavanone increased the growth plate zones thickness of the reserve and pre-hypertrophic zones on the 14th day of embryonic injections. Proliferative zone thickness in the high level of flavanone in day 17.5th of incubation was more than other treatments and control group. Hypertrophic and early mineralization zone showed no meaningful differences. ALP in 14th day of the substance injection increased rather than other groups. Content calcium in femur ofday-hatched treated chicks was more in low doses in 14 and 17.5 days of incubation compared to control but changes in phosphorous content was more in high level of flavanone in day 17.5 compare to high level in 14th day of embryonic injection. Tibia bone ash, calcium was lower in chicks receiving lower flavonoids and phosphorous in 17.5 day of incubation was lower by receiving15 mg of flavanone. In ovo injection of flavanone (as a phytoestrogen) at 15 or 30 mg/ egg or in different day of embryonic had positive effects on bone properties or regulation of changes in chondrocytes phase in the growth plate of newly hatched chicks. It needs to have more investigations studying the effects of flavonoids on prenatal long bone that would affect postnatal growth and changes to form stronger bone in fast growth phase of broiler.

Keywords: Amniotic feeding, Flavanone, Growth plate, Hatched chick

Body weight is affected by early life feeding strategy and hatch moment in broiler chickens

Abstract ID: 230

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In conventional hatcheries, chickens have no access to nutrition (including water) in the hatcher after hatching. As not all eggs hatch simultaneously, but in a so-called hatch window of approximately 20 – 40 h, hatchlings are deprived from nutrition before the majority of eggs hatch. Subsequently, hatchlings undergo several procedures before being transported to the broiler farm. We estimate this delay can last for 72h, depending on hatching conditions, internal hatchery procedures and transport duration. The goal of our study was to test the hypothesis that the provision of nutrition immediately after hatch until placement (early nutrition; EN) enhances weight gain, intestinal development, and reduces intestinal permeability (IP), compared with delayed nutrition (DN). Furthermore, we evaluated the interaction with hatch moment as we expected different body weight gain in early hatchers (first half of hatchers) compared with late hatchers (second half of hatchers). Incubated eggs (n = 432, embryonic age: 17 d) were obtained and hatched. Every 3 h, hatchlings were pulled, sexed, and distributed among either EN or DN (withheld from water and feed for 72 h). To ensure all DN chicks had a 72 h delay to nutrition, ages were registered as biological age. Late hatchers had higher ($P < 0.01$) BW at hatch compared with early hatchers. At 72 h p.h., early hatchers receiving EN had higher ($P < 0.001$) BW compared with later hatchers, whereas no effect of hatch moment was found in DN chickens. This may indicate that early hatchers benefit more from EN compared with late hatchers. No effects of sex on BW development were found. EN chickens had increased villi width at 4 d ($P < 0.001$), but not at 10 and 14 d, indicating that effects of EN might be short-term. Effects of hatch moment or sex were not found. Oral administration of fluorescein isothiocyanate dextran (FITC-d), and subsequent determination of FITC-d levels in plasma after 2h, was used as an indicator for IP. FITC-d concentration at day 4 was higher ($P = 0.10$) in DN chickens compared with EN. Our results indicate that effects of EN are rather short-term with respect to intestinal development under optimal conditions.

Keywords: Delayed Nutrition, Early Nutrition, Intestinal Permeability

Comparative assessment of renal plasma flow and tubular secretion in five different poultry species by determining the para-aminohippuric acid clearance

Abstract ID: 483

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The kidneys play a major role in the elimination of metabolic waste products, drugs and toxic substances. Renal excretion is thrived by the effective renal plasma flow (eRPF), glomerular filtration (GFR), tubular secretion (TS) and reabsorption. Recently, our research group validated intravenous (IV) iohexol administration as sensitive marker to assess the GFR in six different bird species. The eRPF is the volume of plasma that reaches the kidney per time unit. Tubular secretion can be defined as active transport from the peritubular capillaries to the renal tubules. Para-aminohippuric acid (PAH) undergoes both glomerular filtration and tubular secretion. Administering doses of PAH as high as 100 mg/kg bodyweight (BW) results in saturated active transport, and hence, maximum transport capacity can be measured as a marker for TS. Lower dosages of 10 mg PAH/kg BW are considered as marker for eRPF due to the absence of transporter saturation. The aim of the current study was to validate plasma PAH clearance as a potential marker to assess eRPF and tubular secretion in five poultry species, and to propose a kinetic approach for the simultaneous measurement of GFR+eRPF, and GFR+TS in these species. PAH was administered IV to broiler chickens, laying hens, turkeys, Muscovy ducks, and racing pigeons with eight birds (4♂/4♀) per species. Each animal received successively a single bolus of 10 mg PAH/kg BW and 100 mg/kg BW in a crossover design, with a three days wash out period between both administrations. Simultaneously with both PAH administrations, a single IV bolus of 64.7 mg/kg BW of iohexol was also administered, followed by blood sampling until 10h post administration. Plasma endo- and exo-iohexol, and PAH concentrations were simultaneously determined by an in-house validated UPLC-MS/MS method. The area under the plasma concentration-time curve of both iohexol stereoisomers and PAH ($AUC_{0\text{->}\infty}$) was calculated by using the logarithmic trapezoidal method with extrapolation to infinity. The total body clearance of endo- and exo-iohexol and PAH was determined by dividing the administered dose by the $AUC_{0\text{->}\infty}$. Allometric scaling demonstrated a clear association between the bird's BW and the clearance of the different markers. More detailed results will be presented at the conference.

Keywords: Glomerular filtration, Kidneys, Renal plasma flow, Tubular secretion

Effects of organic minerals, fish oil and hydrolysed collagen in broiler diets on growth performance and tibia characteristics

Abstract ID: 113

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Nutrition is one of the most important factors for growth and bone development in broiler chickens. Alterations in ingredients and adjustment of nutrients in broiler diets might positively affect bone development; it might contribute to the reduction of locomotion related problems. This study was designed to evaluate effects of dietary organic minerals (OTM), fish oil (FISH) and hydrolysed collagen (COL) on growth performance and tibia characteristics of broiler chickens. A total of 384 one-day-old Ross 308 male broilers were used in a completely randomized pen design with 4 dietary treatments and 8 replicates per treatment. In the OTM diet, the inorganic calcium, phosphorus and trace elements were replaced by their organic varieties. In the FISH diet, palm oil and soybean oil were substantially (91%) replaced by fish oil. In the COL diet, soybean meal was partly (14%) replaced by hydrolysed collagen. Results showed that the OTM and COL groups reached higher body weight at 42 day of age with lower feed conversion ratios. On day 28, 35 and 42, gait score (GS), tibial rotation (TR), varus valgus deformity (VV), metatarsal breaking strength (MBS), tibia proximal length (TL), tibia lateral cortex thickness (TT), femoral and metatarsal sides tibia proximal head thickness (THT), tibia mineral content (TMC), tibia mineral density (TMD), tibia breaking strength (TBS), tibia stiffness (TSF) and tibia energy to fracture (TEF) were measured (n=3/replicate). Tibia parameters were corrected for differences in BW. Chickens of the OTM treatment had longer TL at day 42; longer THT at day 28; higher TMC at day 42; higher TMD at day 28, 35 and 42; higher TBS at day 42; higher TSF at day 35 and 42; and higher TEF at day 42 compared to the other treatments. All tibia parameters of the FISH treatment showed a general tendency to lower values. All tibia parameters of the COL and control treatments placed between OTM and FISH treatments throughout the experiment. It can be concluded that replacing inorganic Ca, P and trace elements in broiler diets by their organic varieties seems to stimulate growth performance and tibia development.

Keywords: Broiler, Collagen, Fish oil, Organic minerals, Tibia characteristics

Impact of thermal manipulation during embryogenesis on hepatic metabolism in ducks

Abstract ID: 162

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Thermal manipulation during embryogenesis has an immediate and long-term influence on physiology in birds. In this study, we aimed to test whether rising incubation temperature during embryonic days (ED) 12–27, has an impact on the development, growth and adult phenotype in mule duck, the most popular bird used for fatty liver prodution. Four groups of 500 eggs were incubated as follow : the control group at 37,6°C, 50% of relative humidity (RH) 24h/day ; the first treated group at 38,6°C, 65% RH 24h/day ; the second treated group at 38,6°C, 65% RH 16h/day and the third treated group at 39,1°C, 65% RH 16h/day. To determine the effect of thermal manipulation we look at the zootechnical performance during the embryonic development, the breeding, and before/after 21 meals of overfeeding. At 15 days of age, and before/after the overfeeding we collected blood, muscles and liver samples for RNA and protein extraction. The first observation was a decrease in incubation time in all treated groups compared to the control. Sexe ratio and hatching rates were not changed while body weight and temperature were significantly lower at hatching in all treated groups. At 15 day of age, the difference of body weight was totaly catched up, while lower temperature was maintained all along the breeding. Two weeks post hatching no differences were noticed on weights of liver, leg muscle, or adipose tissue, while breast muscle presented a small but significant increase in two treated groups compared to the control. From day 45 to 59 post hatching, total body weight of the 3 treated groups has significantly exceeded the control group, leading to a strong negative correlation between body weight and temperature. Finally, zootechnical results after overfeeding demonstrated a significant increase of liver weight in all treated groups. These preliminary results suggest that embryonic thermal manipulation in mule ducks could accelerate growth and adult performance without alteration of hatchability or sexe ratio. This innovative project could become an interesting and inexpensive strategy for fatty liver production.

Keywords: Fatty liver and meet production, Incubation temperature, Physiology

Monitoring of turkey breeders growth and composition through rearing to photostimulation as influenced by body weight category at 16 weeks of age

Abstract ID: 336

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In order to improve turkey breeders' feeding management, and especially growth pattern and fat deposition prior to the onset of lay, we have evaluated the feasibility to measure these two parameters, associated with turkey breeders body weight category during the second half of the rearing phase by using non-invasive tools such as medical imaging. More specifically, we have identified 3 groups of hens at 16 weeks according to their respective body weights: light, medium and heavy, and kept them together. We have monitored every 2 weeks their individual body weights up to 28 week-old. To monitor body composition, 4 turkeys (1 light, 2 mediums and 1 heavy) were randomly selected every 4 weeks and analyses were performed with a CT-Scanner of bio-medical grade (Siemens Somatom Definition AS ; 100K Volta and 120 mA/s). 500 images were performed every 0.6 mm, 0.45 Pitch and reconstruction filter Safire I26 was used to better characterize fat tissues. Pearson correlation between the body weight of hens from all groups and the volume of adipose tissue determined in vivo was 0.97. For total muscle and offal volume we also found r = 0.97, and for total bone volume r = 0.76. Photostimulation was associated with elevated plasma triglycerides concentration. A correlation of 0.6 was found between ovary weight and fat volume. Interestingly both age and group had an effect on body weight. Average body weight remained different for all three groups throughout trial period and maintaining the order of selection: light remained smaller than medium, which in turn remained lighter compared with the heavy group ($P < 0.001$). In addition, at all ages, animals in heavy group had more adipose tissue then medium and light, but the difference between medium and light on this volume decreased with hen age. In conclusion the present study highlights the importance of CT-Scan in order to follow the changes of body composition through a non-invasive method and to associate these data with growth rate. Similar experiments are in progress on turkey at sexual maturity.

Keywords: Body weight, Computed Tomography, Hens, Rearing, Turkey

The postprandial activity of digestive enzymes in pancreatic juice and blood serum in chicken

Abstract ID: 98

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Presently it is well documented that digestive enzymes enter not only the intestine but also the bloodstream; normally the enzymes are circulating constantly in definite ranges of concentration and activity. Physiological roles of the circulating enzymes, however, still remain unclear. The aim of our study was to determine the interrelationships between the postprandial enzymatic activities in pancreatic juice and blood serum in chicken. The trials were performed on pullets and cockerels (White Leghorn) aged 90-120 days. Pancreatic juice was sampled in chronic experiments on 3 pullets with fistulae in main pancreatic duct; the samples were collected for 10 days during 3 postprandial hours. Blood samples were taken from the axillary vein from 5 birds. Pancreatic amylase activity was determined using Roy-Smith method; trypsin activity using casein hydrolysis; lipase activity using semi-automatic analyzer Sinnowa BS3000P (China). The enzymatic activities in blood serum were determined on semi-automatic analyzer Chem Well 2900 (USA) using specific reagent kits Human (Germany) for amylase and lipase and BAPNA for trypsin. The increases in enzymatic activities in pancreatic juice in 1 hour after the feeding were 76.7% for amylase, 95.5% for protease, and 7.9% for lipase in compare to the respective basal levels; tryptic activity in serum at this moment significantly increased by 66.0% ($P<0.05$) while activities of amylase and lipase remained unchanged. These data are in contrast to the data of Laporte and Tremolieres (1971) who had found the reversed correlation between postprandial tryptic activities in pancreatic juice and blood serum in rats; these authors had suggested that active circulatory trypsin is a negative feedback regulator of pancreatic trypsin synthesis. The authors also had stressed the crucial role of cholecystokinin in this regulation: it inhibits the circulatory trypsin activity and restore the level of trypsin synthesis in the pancreas. However, we found positive correlation between postprandial levels of pancreatic and circulatory trypsin activities ($r=0.36$). The circulatory tryptic activity was found to be the most responsive to feed stimulus.
The study was financed by the sub-program "The investigation of adaptive mechanisms in the digestive system in mammals and poultry to the diets with different ingredients" implemented by the Russian Academy of Sciences (The Presidium's Resolution, July 5, 2017).

Keywords: Chicken, Digestive enzymes in blood, Pancreatic juice, Trypsin

ORAL PRESENTATIONS

Antibiotic resistance

Dietary nucleosides improved performance and gut health lately in broilers

Abstract ID: 519

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Globally, the gut health studies revolves around manipulating their environment but strengthening the intestinal barrier could be an ideal approach which could have more direct effect. The nucleotides will be a limiting factor for the gut mucosa during their early rapid proliferation phase. A biological experiment with 240 chicks was carried out to assess the nucleosides supplementation in an antibiotic free rearing system. Divided into six groups with each containing 40 as follows; Control: without antibiotics; T1: Adenosine 0.1%; T2: Guanosine 0.1%; T3: cytosine 0.1%; T4: Uridine 0.1% and T5: 0.1% mixture of all each in equal proportion. The supplementation was carried out for initial 14 out of 42 days experiment. The production performance, serum biochemistry, slaughter studies and intestinal histo-morphology supported by scanning electron microscopy were carried out at 7, 14 and 42 d of experimental periods. The data were subjected to ANOVA single factor and means were compared to Tukey's Multiple Range Test. The body weight was comparable ($P>0.05$) during the pre-starter phase but starter and finisher stage had higher ($P<0.05$) body weight in T5 (2035g) followed by T1 (1923g). Higher ($P>0.05$) feed intake in supplemented groups with comparable feed efficiency with that of control. The digestive enzyme amylase activity is higher ($P<0.05$) in T3 (648.67 units) and T5 (653.32 units) and lipase activity at T3 (190.72 units) than other groups, respectively. The plasma biochemical constituents, mainly total protein, calcium, phosphorus and consistently higher uric acid content (evidence of intestinal transport) in supplemented groups than control during 7, 14 and 42d of age. The plasma glucose content was unexpectedly comparable among the treatment groups at all assessed age periods. At 7d, the intestinal weight on %body weight was higher ($P<0.001$) in T1 than other groups but the intestinal length expressed to body weight was longest ($P<0.001$) in T5 birds. The histo-morphological studies indicated better intestine development as villi length higher and lower crypt depth in T5 than the other groups at 7, 14 and 42d. The scanning electron microscopy also confirmed lengthier villi in supplemented groups as compared to control. The present study revealed the role of dietary nucleosides supplementation in strengthening the gut cellular structure in an antibiotic free rearing system in broilers. However, as long as feeding nucleosides concern, Adenosine alone or combination of all needs further studies to conclude.

Keywords: Adenosine, Broilers, Gut health, Nucleosides, Performance

Microfloral rehabilitation: Normalisation of Gut Function

Abstract ID: 41

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Gut health and its management is an intricate and complex area governed by numerous factors including nutrition, microbiology, immunology and physiology. When gastrointestinal health is compromised, nutrient digestion and absorption are affected, feed conversion becomes reduced, susceptibility to disease is heightened, all of which can have negative economic impacts. The overall population of microorganisms in the gut is referred to as the microbiome and is recognised as a very diverse community of bacteria, fungi, protozoa, and viruses. Its' diversity varies along the different regions of the GI tract, with regions having less tolerable conditions containing a reduced microbial diversity in comparison to regions more favourable to microbial growth. Within the GI tract, there are multiple interactions between the host, intestinal environment and microbial cells in addition to feed components. These interactions underline the critical role of the microbiota in the health and well-being of the host, although the exact way in which this is achieved is not yet fully understood. The diversity within the microbiome plays a critical role in gut health with beneficial microbes forming a protective barrier lining the gut, preventing the growth of pathogenic bacteria such as Salmonella, Campylobacter, Clostridia and Escherichia amongst others. Poor intestinal health is often associated with increased pathogen colonisation, susceptibility to infectious disease and increased mortality. While there is a need for therapeutic use of antimicrobials to treat disease, globally it is recognised that there is an urgent need to further limit and restrict their use in production settings. The main drawback of the therapeutic use of antimicrobials to combat poor intestinal health is their non-specific effects on the gut microbiome and the reduction in overall gut microfloral diversity noted with their administration. Without intervention, the use of antimicrobials can lead to a vicious cycle whereby their use reduces overall microfloral diversity and selects for the expansion of resistant species to the detriment of non-resistant commensal strains. This reduced diversity allows for the continued proliferation of resistant species and can have negative impacts on health and performance. One strategy to combat the negative consequences of therapeutic antibiotic use is to repair and rehabilitate the gut microfloral diversity. Profiling and understanding the role of intestinal microbial communities is important for the development and understanding of new and existing feed additives thus allowing the manipulation of diets to improve performance, health and welfare. Dietary supplements, which focus on rehabilitating or repairing the gut microflora to aid intestinal health and decrease the animal's susceptibility to disease will prove key in developing non-antibiotic pathogen control strategies.

Keywords: Alternatives, Antimicrobial, Diversity, Microbiome, Nutritional Strategy

"Ecology from farm to fork of microbial drug resistance and transmission" Interventions to reduce antibiotic use

Abstract ID: 148

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"Ecology from farm to fork of microbial drug resistance and transmission" (EFFORT, www.effort-against-amr.eu) is an EU FP7 project that started on December 2013 and will last five years. The Effort project is based on field studies in 10 European countries that aim to link the antimicrobial usage (AMU), antimicrobial resistance (AMR) in different food-producing animal, the (farm) environment, and food of animal origin as well as companion animals and wildlife to quantify the exposure of humans to AMR through different exposure pathways. One part of the project is dedicated to the study of on-farm (Pig, Poultry) interventions tailored by veterinarians to assess their effect on AMU, animal welfare and performance. In Belgium, these farm specific interventions affect over 1.5 million broiler places. Each farm situation is analyzed individually by the supervising veterinary and an action plan is proposed. In the final stage of the project we have identified some strategies that reduce the antibiotic usage in broiler farms in Belgium. Focus is on improving intestinal health with better diagnostics to control coccidiosis and bacterial enteritis, implementing vaccination for coccidiosis and using feed additives. For coccidiosis vaccination, the average days of treatment with antibiotics decreased from 8.8 days before vaccination to 4.5 days in the cycles after vaccination. When we focused on the treatment days for gut-health associated issues, we could see a decrease of 54%. Although less antibiotics where used, the performances where significantly improved during and after these vaccination rounds. The positive effect on the EPEF could be seen up to 3 rounds after last vaccination. Other interventions that have a positive effect to reduce antibiotic use are improved diagnostics, improved management and biosecurity, using alternative feed and water additives and education of the farmer.

Keywords: Animal health, Animal welfare, Antimicrobial resistance, Coccidiosis, Eco-epidemiology

Effective replacement of in-feed antibiotics with a blend of free and buffered organic acids on performance and gut health in broilers

Abstract ID: 583

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Raising broilers with reduced or without antibiotics becomes currently more important. There is a need in finding new alternatives, replacing antimicrobial growth promoters (AGP). This study explores the effect of a drinking water additive (DWA), a blend of free and buffered organic acids, on growth performance, health status and gut microbiota of broiler chickens.
990 day-old male broiler chickens were randomly assigned to 3 treatments (n = 15, 22 birds per pen), including: 1) negative control (NC), 2) positive control (PC, 40 ppm Bacitracin zinc in feed) and 3) a drinking water additive (DWA), at 0.15%. The birds were raised on floor pens with litter and received a commercial two-phase diet. During the 42-day trial period, the body weight and feed intake were recorded per pen. At day 21 and day 42 footpad dermatitis was evaluated. Intestinal and crop samples were taken from 1 bird per pen to evaluate intestinal health parameters, tight junction protein gene expression, histology, volatile fatty acids and bacterial counts.
The results indicated that DWA significantly increased body weight (3.164 kg) on day 42, compared to NC (2.972 kg) and PC (2.981 kg) ($P<0.001$). Feed intake during day 1-21, day 22-28 and day 1-42 ($P < 0.05$) shows significant improvement for DWA compared to NC and PC ($p<0.05$). Feed conversion ratio during day 1-21 and 21-42 was significant improved for DWA compared to NC, and significantly lower during day 1-42 for DWA (1.560) compared to NC (1.597) and PC (1.582) ($p<0.05$). DWA decreased the footpad dermatitis scores on day 21 and 42 ($P<0.05$), whereas the PC did not. Gene expression of the tight junction protein Claudin-1 in the jejunum showed a significant increase occurred in DWA group compared to NC ($p< 0.05$). Also, the number of goblet cells was increased in the jejunum of the DWA group compared to both controls. ($P<0.05$). The amount of volatile fatty acids, acetate, propionate and butyrate was significantly higher in the DWA group on day 42. Enterobacteriaceae counts in the crop tended to be lower in the DWA group ($p = 0.054$) and were significantly lower in the cecum ($p<0.05$) compared to NC and PC at day 42.
The results indicate that using a DWA containing free and buffered organic acids could replace the use of in-feed antibiotics in broilers, while maintaining performance. Moreover, the DWA showed improvement of different gut health parameters.

Keywords: Antibiotic replacement, Gut health, Organic acids, Performance, Tight junctions

Effect of disinfectant use on antibiotic susceptibility of E. Coli isolates from broiler houses

Abstract ID: 512

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Farm disinfectants are widely used in primary production, but there is an increasing concern that the use of disinfectants is selecting for antibiotic resistance. In order to determine the effect of farm disinfectants use on antimicrobial susceptibility, *Escherichia coli* was isolated from environmental samples taken after cleaning and disinfection at 25 broiler houses. In addition to the sampling, information about the composition of the applied farm disinfectants was collected by questioning.
To monitor antimicrobial susceptibility, 200 *E. coli* isolates were subjected to a panel of 14 antibiotics and for each isolate the MIC (Minimum Inhibitory Concentration) was determined using broth dilution. For a selection of 57 *E. coli* isolates, biocide susceptibility testing was performed by determining the MBC (Minimal Bactericidal Concentration) of commonly used disinfectant components: a quaternary ammonium compound (QAC), glutaraldehyde (GA), formaldehyde (F) and a peracetic acid (PA) - hydrogen peroxide (H_2O_2) formulation.
Results showed high levels of antibiotic resistance to ampicillin (77%), sulfamethoxazole (62%), ciprofloxacin (60%), trimethoprim (56%), tetracycline (53%) and nalidixic acid (46%). About 58% of the isolates were resistant to four or more antimicrobial agents, and only 13% were susceptible to all 14 antibiotics tested. Resistance to the tested disinfectant components benzalkoniumchloride (QAC), formaldehyde, glutaraldehyde and a peracetic acid-hydrogen peroxide formulation could not be found. However, significant associations were found between the use of QAC-GA and tetracycline resistance and between the use of PA-H_2O_2 and ampicillin resistance. In conclusion, these results indicate the possibility that the practical use of disinfectants can select for antibiotic resistance, but further research focusing on this association is needed.

Keywords: Antibiotic resistance, Broilers, Desinfectants, E. coli

Good management and poultry welfare - perfect combination for a responsible use of antimicrobials

Abstract ID: 345

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Responsible use of antibiotic is a growing need that can no longer be ignored. In the European Union, several countries have been setting up targets for antibiotic use in the industry. With a lower use of antibiotics the industry needs to undergo significant changes to keep on meeting consumer and market demand. Good management and good animal welfare should always underpin a plan for antibiotic reduction. These two pivotal pillars will provide sustainability to any antibiotic stewardship programme adopted by industry. ValVerde, a poultry producers located in Northern Italy, developed a speciality brand called Gran Selezione- Pollo dal Piedmont. The brand produces 26,000 broilers per year and it is located in the Piedmont region. The brand has adopted slower growing breeds: in most cases New Hampshire Red and some local traditional breeds the birds are kept at a maximum stocking density of 30 kg/m^2. Natural light, 1 straw bale for 1000 birds, perches and outdoor access are provided as environmental enrichment for the broilers. The new vaccination portfolio includes 7 different vaccines in the hatchery. Feed additives have also been introduced: linseeds are used in a concentration of 4% and polyphenols are bought as a premix which is then mixed with the feed with a ratio of 200 to 250 g per 100 kg. Only natural herbs are used with medicinal purposes. The farm has been running for 2 years without antibiotic or coccidiostats use. Currently the slaughter weight is between 2.5 and 3 kg, which is equivalent to a slaughter age between 90 and 100 days. The feed conversation ratio is approximately 3:1. The average mortality it is situated between 3 and 4%, depending on the season. Last year, due to the success of the concept, the brand has had an increase of 71% in their sales. Antibiotic reduction is becoming a mandatory practice in animal production. Examples such us the Pollo Dal Piemonte Gran Selezione illustrate how a food company can tackle the issue and how good management and higher welfare are pivotal in reducing the need for antibiotics and other antimicrobial drugs such as coccidiostats.

Keywords: Antibiotics, Coccidiostats, Italy, Poultry welfare

Practical evidence-based approaches towards reduction of antimicrobial use and antimicrobial resistance

Abstract ID: 493

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In the post-AGP era, the focus is shifting towards reduction of antimicrobial use (AMU) and thereby preventing antimicrobial resistance (AMR) in poultry production, which requires implementation of the right interventions.
The complexity involved in assessing the effectiveness of interventions aimed at reducing AMU/AMR hinders effective decision making. A systematic, evidence-based approach is proposed in order to choose and evaluate candidate interventions in an evidence-based and practical way.
Objective setting: Clarity of the objective is needed, as interventions for reduction of AMU may differ from those for prevention of AMR. Moreover, a clear definition facilitates the evaluation of effectiveness
Selection of interventions: Disease is generally multifactorial. Therefore interventions might focus on different aspects of poultry production, e.g. nutrition, incubation, genetics, disease monitoring and vaccination, housing, and climate. The following evidence based approaches can be used as methods for selecting interventions to reduce AMU/AMR: Based on BEST PRACTICE, which can be defined as a program, activity or strategy that is known to work effectively with successful outcomes and is supported by reliable data sources;
Analysis of the quantitative variation in current practices to identify key SUCCESS FACTORS, and their influence on disease occurrence.
NOVEL INTERVENTIONS, not used in current practice, can be selected by reviewing of experimental studies done using reproducible challenge models of the target disease.
Measuring intervention effectiveness
Each approach constitutes an evidence based method for selecting interventions aimed at reducing AMU/AMR. For the evaluation of effectiveness of interventions following activities can be considered:
Evaluation within the target population, under representative challenge conditions;
Implementation of health surveillance program, monitoring disease occurrence, AMU and AMR;
Identification of farm factors and practices that interact with the intervention or directly influence disease occurrence;
Studies addressing if there is synergy, antagonism or additivity among interventions.
Dependent on existing surveillance systems, production database, legislative environment, urgency, available resources and expertise, different approaches will be preferred. A clear pre-assessment, and the use of illustrative examples is helpful in the decision making on the approach of choice.

Keywords: AMR, AMU, Epidemiology, Evidence Based, Intervention

Retrospective study on antimicrobial resistance profiles from E. coli isolated from diagnostic samples

Abstract ID: 316

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Antimicrobial testing is an important part of bacteriological investigations in a diagnostics laboratory. Despite certain disadvantages the agar gel diffusion test is the method of choice due to its easy handling and options for modification. Between April 2014 and December 2017 a total of 1290 *E. coli* isolates from layer, broilers and turkeys were investigated for their resistance profile using a standardized setting with ampicillin, amoxicillin, colistin, doxycycline, enrofloxacin, neomycin, oxalic acid, tretracycline, tilmicosin, trimethoprim/sulfamethoxazole and tylosin. Overall, high resistance was noticed against the macrolides tylosin and tilmicosin with nearly none of the isolates being sensitive. A difference in the sensitivity could be seen within the group of aminoglycosides, with more isolates being resistant to neomycin compared to spectinomycin. This tendency was consistent throughout the testing period. No significant differences were found in the sensitivity against colistin, oxalic acid and trimethoprim/sulfamethozaxol over the years. For most of the antibiotics significant differences were noticed between layers and turkeys / broilers with a much higher number of isolates being sensitive from layers. Finally, over the 4 year-period an increase of sensitivity was noticed, especially for tetracyclines (doxycycline and tetracycline), but also penicillines (ampicillin, amoxicillin) and quinolones (enrofloxacin) which might reflect the more prudent use of in recent years. Furthermore, within a comprehensive clinical study the microdilution method (Micronaut system) was applied to investigate the antibiotic resistance profiles of 219 *E. coli* isolates. These isolates were obtained from the bone marrow of 66 broiler birds, and up to 3 colonies from the primary culture of each bird were investigated. Interestingly, antimicrobial resistance profiles varied substantially within a bird and identical profiles were only obtained for isolates from 14 birds. This highlights the difficulty to select the right clone for antimicrobial resistance testing, an issue so far very much neglected in routine diagnostic investigations.

Keywords: Agar gel diffusion test, Antibiotics, E. coli, Microdilution, Resistance profiles

Should enzymes be part of your antibiotic reduction strategy?

Abstract ID: 140

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In the post-AGP era, enzymes play a crucial role by reducing feed costs, compensating for increases in production costs and performance challenges. Enzymes are conventionally looked at as a nutritional feed additive with a nutrient matrix attached to them. However, as science of application of enzymes in feed and their effects on digestion kinetics in the gut evolve, it is becoming increasingly clear how enzymes influence the conditions in the gastrointestinal tract (GIT): enzyme application does not only facilitate digestion but can also be used as a tool for influencing microbiota aiming to improve intestinal health. Dietary challenges posed by fiber content in the diet (especially soluble arabinoxylans), have consequences such as higher digesta viscosity and increased digesta transit time, leading to proliferation of microflora in the small intestine, competition for available nutrients, overgrowth of pathogenic bacteria, as well as decreased nutrient digestibility; especially of fat. These events can very well be reversed by the addition of xylanase which reduces viscosity by degrading soluble arabinoxylans and reduces/limits proliferation of microflora in the small intestine. Furthermore, the degradation of arabinoxylan -by xylanase- in the upper gastrointestinal tract produces xylo-oligosaccharides which have a prebiotic effect on the beneficial microbial population in the lower gastrointestinal tract. A reduction in pathogen levels in the digesta by the application of xylanase in the diet has been shown in several studies. The beneficial shift in microbial activity in the upper and lower GIT positively impacts gut health, reducing the risk of disease, and consequently the need for antimicrobial treatment. Furthermore, how enzymes help to improve performance of other gut health feed additives is also being studied in more depth. The holistic effects of xylanase on the digestion process in the small intestine makes it a fundamental part of the diet, which should also be considered within an overall antibiotic reduction strategy.

Keywords: Enzyme, Gut health, Prebiotic, Xylanase

The role of the poultry veterinarian in reduction of antibiotic use, intervention study in broiler farms in different EU countries

Abstract ID: 558

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“Ecology from farm to fork of microbial drug resistance and transmission” (EFFORT) is an EU FP7 project, that started on December 2013 and will last five years, that studies how to reduce antimicrobial resistance in farm animals. During the project intervention studies were performed in broiler farms in 3 different EU countries, involving more than 1300 broiler flocks in 115 farms, to determine actions that can contribute to reduce antimicrobial use and to abolish the use of specific antibiotics which are critically important for use in humans. A risk analysis was made for each individual farm based on information collected by the veterinarian in an audit questionnaire about the farmer, farm infrastructure and management, animal health status and disease management. Data on performance and antimicrobial use were collected for each farm retrospectively and during the study. Every six months an action plan was designed by the veterinarian for each farm based on the risk analysis with the aim to develop strategies to improve health status and reduce AMU. Multi-level interventions have been implemented in the broiler farms and were evaluated by the veterinarian during regular follow up visits to the farms, assessing success and reasons of failures of different actions. Most interventions focus on improving health status by improving diagnostics and prevention, other actions include training of the farmer to detect problems in early stages and to improve biosecurity and hygiene on the farm, and implementing innovative strategies in feed and water additives. Reducing AMR by reducing AMU in farm animals is a challenge for the veterinarian and the farmer, it is also an incentive for the poultry veterinarian to assume his role as gatekeeper of food safety, animal health and welfare.

Keywords: Antimicrobial resistance, Disease management, Food safety, Veterinary expertise

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348 **Effect of different exogenous enzymes on the early life performance of broilers /** Abstract ID: 432
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349 **Effect of different levels of Macleaya cordata alkaloid extract in low protein diets on performance, Ileal protein digestibility and plasma amino acid concentration in broiler chickens /** Abstract ID: 610
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350 **Effect of digestible Trp to Lys ratio and low dietary crude protein diets on laying performance and egg quality in 36 to 50 weeks old laying hens /** Abstract ID: 467
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351 **Effect of dried tomato pomace on the performance, egg traits, blood parameters and immune response of laying hens /** Abstract ID: 16
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352 **Effect of feeding high quality crude glycerol as energy source in broiler diets /** Abstract ID: 423
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353 **Effect of feeding yeast fraction alone or in combination with organic selenium on oxidative status and meat quality of broilers under heat stress /** Abstract ID: 360
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354 **Effect of fiber source and content on volume and physical properties of the final diet /** Abstract ID: 31
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355 **Effect of high dietary levels of α-tocopherol acetate on immune response of light and heavy weight male broiler breeders /** Abstract ID: 58
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356 **Effect of high fibre layer diets on layer performance and egg quality /** Abstract ID: 349
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357 **Effect of nonanoic acid supplementation on broiler performance /** Abstract ID: 341
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358 **Effect of partial substitution of D3 by 25-OH-D3 on production parameters and gait scoring in commercial broiler farms /** Abstract ID: 547
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359 **Effect of prebiotic on intestinal microflora, carcass and meat chemical composition of broiler chickens /** Abstract ID: 46
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360 **Effect of probiotic supplementation on performance and egg quality of laying hen during the late egg cycle /** Abstract ID: 410
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361 **Effect of soapnut shell powder supplementation on reproductive performance of broiler breeders /** Abstract ID: 500
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362 **Effect of source of Selenium on Se deposition and egg quality /** Abstract ID: 416
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363 Effect of two phytases at two doses on performance of broilers during 0-21d of age / Abstract ID: 268

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364 Effect of xylanase in corn-soybean meal diets on growth performance of broiler chicken / Abstract ID: 476

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365 Effects of a fat coated betaine based product on breast meat yields in turkeys and on performances and blood cells membrane integrity in heat-stressed laying hens / Abstract ID: 273

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366 Effects of a global enzyme solution on growth performance and bone quality of broilers from 1 to 28 days of age / Abstract ID: 190

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367 Effects of conditioning time and sodium bentonite on pellet quality and performance in growing male broilers / Abstract ID: 368

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368 Effects of dietary inclusion of high- and low-tannin faba bean (Vicia faba L.) seeds on microbiota, histology and fermentation processes in the gastrointestinal tract of finisher turkeys / Abstract ID: 286

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369 Effects of dietary inulin supplementation on growth performance, gut health and bone quality of broilers / Abstract ID: 183

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370 Effects of dietary tuna oil levels and feeding periods on growth performance and meat quality of Thai crossbred chickens / Abstract ID: 455

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371 Effects of different heat damaged protein ingredients on protein digestibility and caecal proteolytic fermentation in broilers / Abstract ID: 236

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372 Effects of different levels of natural glauconite and zeolite on performance, tibia bone characteristics and blood parameters of broiler chicken / Abstract ID: 572

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373 Effects of in ovo and dietary supplementation of flavanone onantioxidant defence system and performance of postnatal broiler / Abstract ID: 527

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374 Effects of multispecies synbiotic on performance parameters of commercial turkey hens / Abstract ID: 347

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375 Effects of probiotic (Saccharomyces cerevisiae) and antibiotic Neomycin on performance and some carcass characteristics of broiler chickens / Abstract ID: 559

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376 Effects of supplementing D or L- Methionine on productive performance and egg quality in laying hens subjected to chronic cyclic heat stress / Abstract ID: 144

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377 Efficacy of an algo-clay complex on decreasing mycotoxins liver toxicity on broiler / Abstract ID: 450

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378 Emulsifier and carbohydrase in a maize-wheat-SBM-tallow diet for broiler chickens / Abstract ID: 55

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379 **Encapsulated sodium butyrate to improve gut development and control enteric bacteria in broilers /** Abstract ID: 418
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380 **Ensiling of mixed forages of beans and grains as feed for organically raised laying hens /** Abstract ID: 446
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381 **Evaluating probiotic application alone or in combination with antibiotic growth promoters on broiler performance and health status /** Abstract ID: 29
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382 **Evaluating probiotic application alone or in combination with antibiotic growth promoters on broiler performance and health status /** Abstract ID: 362
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383 **Evaluation of a global enzyme solution on growth performance and carcass characteristics of broilers fed corn-wheat-soybean based diets reduced in metabolizable energy and nutrients /** Abstract ID: 188
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384 **Evaluation of egg quality and vitamin D transfer following vitamin D2-rich yeast supplementation in laying hens /** Abstract ID: 54
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385 **Growth performance of broilers chicken fed different concentration of trona (sodium sesquicarbonate) treated feather meal /** Abstract ID: 13
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386 **Growth response of Ross 308 broiler chickens fed different dietary energy levels /** Abstract ID: 166
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387 **Guanidinoacetic acid supplementation in broiler chickens fed corn-soybean diets affects performance in the finisher period and energy metabolites in breast muscle independent of diet nutrient density /** Abstract ID: 453
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388 **Hydroxy trace minerals allow dosage reduction without growth impairment of broilers /** Abstract ID: 553
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389 **Ileal phosphorus digestibility of soybean meal in broilers of different ages /** Abstract ID: 381
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390 **Implication of dietary willow bark extract (Salix alba) on performance and caecal microflora of broilers (14-28 days) reared at 32°C /** Abstract ID: 361
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391 **Increasing dietary amino acid content in grower and finisher phases tends to increase the incidence rate of wooden breast in broiler chickens /** Abstract ID: 407
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392 **Influence of cereal type and fat source on performance, pellet quality and gastrointestinal traits in broiler starters fed pelleted diet /** Abstract ID: 616
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393 **Influence of different crude fiber contents in the diet on the performance of laying hens /** Abstract ID: 19
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394 **Influence of different sources and length of fibers in the diet on the growth of broiler chickens /** Abstract ID: 20
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395 **Influence of graded dietary levels of seeds of three species of lupin on growth performance, nutrient digestibility, and excretion of total and free sialic acids of broiler chickens /** Abstract ID: 107
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396 **Influence of particle size and fiber source on performance and nutrient utilization in broiler starters fed pelleted diets /** Abstract ID: 621
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397 **Influence of Wheat Particle Size, Insoluble Fibre Sources and Whole Wheat Inclusion on Gizzard Musculature Development and Nutrient Utilisation in Broilers** / Abstract ID: 221
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398 **Investigation on growth performance and ileal digestibility of probiotics (Bacillus subtilis and Bacillus licheniformis) supplementation in broilers** / Abstract ID: 18
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399 **Is pathogen induced anorexia in broilers infected with Eimeria maxima influenced by food quality?** / Abstract ID: 557
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400 **Larvae of synanthropic flies in diets for broilers** / Abstract ID: 596
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401 **Lupine in Diets for Laying Hens** / Abstract ID: 118
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402 **Lysine and protein requirements of laying Japanese quail** / Abstract ID: 402
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403 **Manipulating energy density and dietary electrolyte balance in ISA Brown laying hen diets diluted with Kikuyu grass (Pennisetum clandestinum)** / Abstract ID: 486
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404 **Manipulation of Meat Quality of The Late Period of Laying Hens through Supplementation Probiotic, Metabolizable Energy and Digestible Essential Amino Acids** / Abstract ID: 70
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405 **Meat quality of slow-growing chickens housed on pasture** / Abstract ID: 110
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406 **Mineral composition of eggs and vitelin sac of day-old chicks from broiler breeders in different ages** / Abstract ID: 213
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407 **Molecular modulation by GOS prebiotic injected in ovo in broiler chickens under heat-stress** / Abstract ID: 451
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408 **Mycotoxin Survey 2017 - The threat in Europe** / Abstract ID: 399
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409 **Ontogeny of feather pecking behaviour with focus on early nutrition** / Abstract ID: 232
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410 **Optimum digestible valine to digestible lysine ratio for growth rate and feed efficiency of broiler chickens** / Abstract ID: 327
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411 **Performance, Immune Related Organs Weight, and Blood Metabolites of Broiler Chicks Fed Diets Included Immune Responses Enhancers** / Abstract ID: 182
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412 **Performance of Bacillus probiotics is strain specific** / Abstract ID: 514
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413 **Performance of layers and egg quality traits as affected by the dietary supplementation of layers' diet with Spirulina sp.** / Abstract ID: 487
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414 **Phytase storage stability under different trace mineral premixes** / Abstract ID: 470
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415 **Processed animal proteins can replace soybean meal in broiler diets /** Abstract ID: 114
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416 **Profile of fatty acids of yolk sacs of embryos from broiler breeders supplemented with CLA /** Abstract ID: 222
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417 **Reduction of contamination of broiler carcasses with Salmonella spp. through the supplementation with Saccharomyces cerevisiae boulardii CNCM I-1079: a multi-analysis /** Abstract ID: 271
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418 **Research of Physiological and Microbiological Peculiarities of Meat Chicken Breeds Digestive System in Fetal and Post-Fetal Periods for the Purpose of Creating New Feeding Techniques Ensuring the Fullest Possible Implementation of Genetic Potential of Poultry /** Abstract ID: 108
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419 **Response of Japanese quails fed diets containing graded levels of Honey-flavoured sun-dried cassava peel meal as a replacement for maize /** Abstract ID: 321
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420 **Scientific review of selenium enrichment data in different organs in broilers fed organic selenium source (Alkosel) /** Abstract ID: 56
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421 **The adverse effect of surplus dietary L-arginine supply on parameters of the nitrogen metabolism in moderate restrictively fed growing cockerels /** Abstract ID: 241
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422 **The comparison of farm formulated and commercial diets on broiler production performance and carcass yield /** Abstract ID: 536
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423 **The effectiveness of Taraxacum officinale under conditions of the intestinal microbial imbalance in broiler chickens /** Abstract ID: 608
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424 **The effect of a combination of prebiotics and herbs rich in polyphenols on the growth performance and uniformity in broilers /** Abstract ID: 415
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425 **The effect of alfalfa and ascorbic acid on performance and egg quality /** Abstract ID: 109
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426 **The effect of bee bread as a feed additive on selected egg quality and hens' performance /** Abstract ID: 266
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427 **The effect of feed additives on cecal short chain fatty acids production /** Abstract ID: 606
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428 **The effect of incremental dietary levels of canola meal on growth performance of broiler chickens /** Abstract ID: 275
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429 **The effect of polyphenols and vitamin E on the growth performance, meat quality** and **antioxidant status of broiler chickens under heat stress conditions /** Abstract ID: 234
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430 **The effect of synbiotic and probiotic preparations on the growth performance, gastrointestinal tract development and health status of turkeys /** Abstract ID: 239
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431 **The effects of microbial muramidase inclusion in broiler diets on growth performance, apparent ileal digestibility and intestinal histology /** Abstract ID: 153
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432 **The effects of moderate feed restriction and surplus dietary arginine supply on relative digestive organ weights in growing cockerels of a dual-purpose breed /** Abstract ID: 243
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[1]Chair for Nutrition Physiology and Animal Nutrition, University of Rostock, Rostock, Germany, [2]Institute of Animal Nutrition, Friedrich-Loeffler-Institute, Braunschweig, Germany

433 **The effect of particle size and insoluble fiber sources on growth performance and gastrointestinal tract development in broiler starters /** Abstract ID: 613
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[1]Razi University, Kermanshah, Iran, Islamic Republic Of, [2]Massey University, Palmerston North, New Zealand

434 **The efficiency of xylanase in broiler chickens fed with increasing dietary level of rye /** Abstract ID: 248
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435 **The intestinal inositol phosphate pattern in broilers as influenced by phytase source /** Abstract ID: 314
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436 **The length of collection period affects the estimate of dietary metabolisable energy in broilers /** Abstract ID: 159
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[1]College of Agriculture, Križevci, Croatia, [2]Harper Adams University, NEWPORT, United Kingdom

437 **The use of a prestarter contributes to a better start and optimal results of broilers /** Abstract ID: 564
S. De Vos[1], S. Cardinaels[2], K. De Baere[2]
[1]INVE België, Baasrode , Belgium, [2]Experimental Poultry Centre – Province of Antwerp, Geel, Belgium

438 **Trace mineral sources and zinc levels influenced growth performance, tissue mineral content and carcass yield of 35-day old male broilers /** Abstract ID: 189
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439 **Transfer of different forms of vitamin K from hens to broilers /** Abstract ID: 491
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440 **Triticale grain in diets for laying hens /** Abstract ID: 99
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[1]Federal Scientific Center "All-Russian Research and Technological Poultry Institute" of Russian Academy of Sciences , Sergiev Posad, Russian Federation

441 **Use of a calcium particulate and feeding time in combination with 25-hydroxycholacifereol to reduce fracture susceptibility in laying hens /** Abstract ID: 635
L. Lazarov[2], M. Toscano[1]
[1]University of Bern, Division of Animal Welfare , Zollikofen, Switzerland, [2]Trakia University, Stara Zagora, Bulgaria

442 **Xylanase and xylo-oligosaccharide have a positive effect on body weight and indicators of immune function in broilers at 28 days of age /** Abstract ID: 192
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443 **Association of Heterosis, Dominance effect on Body Weight and the Expression of Growth Hormone Gene and Insulin Like Growth Factor – I Gene in Slow Growing Chicken /** Abstract ID: 320
A. Molee[1], P. Kaewnakian[1], R. Bunnom[1], S. Boonanuntanasarn[1], W. Molee[1]
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444 **Bone stability and performance level of phylogenetically divergent chicken lines after dietary calcium restriction /** Abstract ID: 280
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445 **Chicken sperm cryopreservation as a tool of maintenance genetic diversity in small scale populations /** Abstract ID: 382
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446 **Direct and indirect effects of selected morphometric traits on body weight of fulani ecotype chickens of Nigeria /** Abstract ID: 11
A. Egena[1], Y. Sulayman[1], A. Ayotunde[1], O. Falowo[1]
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447 **Discovering genes and pathways in the shell gland which may be involved in egg cuticle formation by gland shell RNA-seq data analyses /** Abstract ID: 533
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448 **Does Chronic Heat Stress Affect Hypothalamic AMPK and (An)Orexigenic Neuropeptide Gene Expression in Broilers? /** Abstract ID: 439
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449 **Egg production and egg shell quality of phylogenetically divergent chicken lines during dietary calcium depletion /** Abstract ID: 358
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450 **Establishing of in vivo Transfection of Primordial Germ Cells in Chicken Eggs using the Sleeping Beauty Transposon System /** Abstract ID: 515
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451 **Genetic characterization and population structure of local Polish and Italian poultry breeds /** Abstract ID: 614
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452 **Genome-wide association analysis of egg production in chickens across the whole laying period /** Abstract ID: 626
Z. Liu[2], C. Sun[2], Y. Yan[1,2], A. Liu[1], N. Yang[2], G. Wu[1]
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453 **Germline chimera production from cryopreserved primordial germ cell lines of a Hungarian indigenous chicken breed /** Abstract ID: 123
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454 **Identification and genetic analysis of extreme feather pecking behaviour in laying hens /** Abstract ID: 258
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455 **In ovo genotyping of chicken using DNA isolated from allantoic fluid /** Abstract ID: 389
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456 **Marker genes in poultry of gene pool flock /** Abstract ID: 88
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457 **Morphological blood indices in turkeys of preserved breeds /** Abstract ID: 133
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458 **New possibilities in the gene conservation of Hungarian indigenous chicken breeds /** Abstract ID: 80
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459 **Optimization of live bodyweight and mortality during the selection of preparental lines of Hisex Brown chicken /** Abstract ID: 124
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460 **The effect of plumage colour and serotonin transporter gene polymorphisms on feather pecking behaviour in laying hens /** Abstract ID: 157
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461 **The transgenic technologies improving the efficiency of poultry production /** Abstract ID: 87
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462 **The use of infertile interspecific hybrids for a novel model of PGC reintroduction applicable in gene preservation for poultry /** Abstract ID: 65
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463 **Verification of effect on the growth rate in Hinai-jidori chickens (Japanese meat-type chicken) by selection using the CCKAR g.420 C < A polymorphism /** Abstract ID: 224
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464 **Effect of citric acid addition on functional and phisical properties of pasteurisated liquid whole eggs during 4 weeks of storage /** Abstract ID: 641
H. Medić[1], N. Marušić Radovčić[1], S. Karlović[1], A. Režek Jambrak[1]
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465 **Effect of maize hybrid antioxidant potential on egg yolk oxidative stability – correlation analysis /** Abstract ID: 542
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466 **Egg parameters of two Hungarian indigenous chicken breeds /** Abstract ID: 219
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467 **Evaluation of quail eggs quality as affected by storage period, temperature and egg size** Abstract ID: 146
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468 **Egg production in China /** Abstract ID: 214
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469 **Influence of lycopene on malondialdehides, fatty acid profile and sensory and texture properties of fresh and stored eggs, using rapeseed oil in laying hens nutrition /** Abstract ID: 477
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[1]Kaunas University of Technology, Kaunas, Lithuania, [2]Lithuanian University of Health Sciences, Kaunas, Lithuania

470 **Influence of organic acid mixture on laying hens productivity, egg quality parameters and sensory properties /** Abstract ID: 566
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471 **Methods of surface disinfection of eggs prior to incubation /** Abstract ID: 160
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472 **New functional egg products based on melange and enriched with macro- and micronutrients /** Abstract ID: 120
I. Stefanova[1], V. Mazo[1], A. Kavtarashvili[1], L. Shahnazarova[1], A. Klimenkova[1]
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473 **Our experiences in enriching table eggs with n-3 PUFA /** Abstract ID: 63
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474 **Production of outdoor and equol enriched eggs for the benefit of human health /** Abstract ID: 563
V. Tosar[1], G. Rousseau[1], E. Froidmont[1]
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475 **Quality of omega-3 eggs enriched with lutein /** Abstract ID: 59
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476 **Quality of table eggs from different housing systems /** Abstract ID: 170
L. Perić[1], M. Đukić Stojčić[1], I. Jajić[1]
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477 **The comparative efficiency of organic vs inorganic forms of selenium and vitamin E in hen diets for biofortification of table eggs /** Abstract ID: 125
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478 **The effects of feeding Diamond V Original XPC on reducing Salmonella prevalence, numbers and NARMS panel antibiotic resistance in cloacal swabs taken from commercial layers /** Abstract ID: 288
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479 **The technology of new functional foodstuffs based on egg albumen enriched with calcium and iodine** / Abstract ID: 119
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480 Poultry Meat Quality and Safety

480 **Carcass composition and edibility yield of two heavy broiler strains** / Abstract ID: 496
G. Hahn[1], M. Judas[1], M. Spindler[1]
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481 **Differences in performance and carcass yield of Ross 308, JA757 and ISA Dual chickens** / Abstract ID: 227
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482 **Effect of dietary ferric tyrosine (TYPLEX™ chelate) supplementation on Campylobacter jejuni counts, on quality and sensory characteristics of breast and thigh meat of broiler chicken exposed to natural Campylobacter jejuni challenge** / Abstract ID: 92
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483 **Effect of dietary marine microalgae (Schizochytrium. SHG104) on growth performance, meat quality and fatty acid composition in broiler** / Abstract ID: 319
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484 **Effect of different concentrations of the Powdered Hawthorn (Crataegus elbursensis) extract on the physical quality of chicken's breast meat** / Abstract ID: 540
Z. Ansari Pirsaraei[1], R. Taherian[1], A. Jafari Sayadi[1], P. Biparva[1]
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485 **Effect of incorporating an emulsifier complex (DIGESTFAsT) on broiler meat quality during the storage** / Abstract ID: 218
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486 **Effect of laying-type cockerels DOMINANT CZ hybrid combination on carcass parameters and meat quality** / Abstract ID: 488
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487 **Effect of organic acid mixture on broiler chicken meat and liver quality** / Abstract ID: 565
R. Gruzauskas[1], V. Buckiuniene[1], V. Slausgalvis[2], V. Sasyte[1], A. Raceviciute-Stupeliene[1], A. Dauksiene[1], J. Al Saifi[2], R. Stankevicius[1]
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488 **Effect of Rosemary powder (Rosmarinus Officinalis) feed incorporation on organic broiler chickens performance and carcass quality** / Abstract ID: 180
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489 **Effects of feeding Diamond V Original XPC on Salmonella prevalence, numbers, and antibiotic resistance in ceca samples taken from commercial broilers** / Abstract ID: 287
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490 **Inter-muscular variation in ultimate pH and colour of broiler muscles** / Abstract ID: 326
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491 **Physical and chemical meat characteristics of chicken ross 308, JA757 and isa dual hybrid** / Abstract ID: 226
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492 **Phytochemicals for controlling Campylobacter jejuni in poultry: An update on novel delivery methods for increased efficacy and understanding of antimicrobial action** / Abstract ID: 292
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493 **Productivity and meat quality in floor vs. cage housed broilers** / Abstract ID: 122
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494 **Salt reduced chicken wings** / Abstract ID: 424
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495 **The effect of polyphenols and vitamin E in the finishing feed for turkeys on meat quality** / Abstract ID: 278
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496 **The effects of preparation Polyferon on the productive performance and meat quality in broilers** / Abstract ID: 121
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499 **The effect of combined monochromatic lighting on sperm quality of broiler breeder males** / Abstract ID: 377
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500 **Air Sample based Flock Diagnostics** / Abstract ID: 597
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501 **Anticoccidial efficacy of a multi-strains yeast fractions product in commercial broiler chickens exposed to a mixed challenge of Eimeria acervulina, E. maxima, and E. tenella** / Abstract ID: 647
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502 **AviPro® IBD Xtreme vaccination in broiler farms in high risk area for very virulent infectious bursal disease virus** / Abstract ID: 32
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503 **Bacillus subtilis 29784 maintain performances of broilers in stressed conditions** / Abstract ID: 436
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505 **Compared effect of copper-exchanged zeolite and brown macroalgae Ascophyllum nodosum on fecal excretion of Avian Pathogenic Escherichia coli in a chicken intestinal carriage model** / Abstract ID: 186
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506 **Comparison in ossification zones between broiler and dual-purpose breeds** Abstract ID: 147
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507 **Comparison of gut microbiota in hens** / Abstract ID: 600
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508 **Detection of Mycoplasma synoviae infections in vaccinated broiler breeder flocks** / Abstract ID: 35
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509 **Dietary ferric tyrosine affects broiler chicken performance, intestinal health and Campylobacter counts exposed to natural Campylobacter jejuni challenge** / Abstract ID: 89
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510 **Dried algae rich in beta-(1,3)-glucan as a replacement for in-feed antibiotics /** Abstract ID: 435
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511 **Drinking water vaccination with Gumboro 228E: monitoring by PCR and serology following extension of the period during which the vaccine solution is dispensed /** Abstract ID: 517
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512 **Effect of incorporating an emulsifier complex (DIGESTFAsT) on meat quality during the storage /** Abstract ID: 181
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513 **Effect of the Saccharomyces sp. and Bacillus subtilis based probiotics on broiler chickens performance and caecum microbiome community /** Abstract ID: 544
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516 **Efficacy study of a live attenuated Newcastle Disease vaccine in SPF and commercial pullets /** Abstract ID: 216
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518 **Efficiency of natural plant extract feed additives on slow growing broilers compare to coccidiosis vaccine /** Abstract ID: 205
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519 **Elimination of poultry red mites (dermanyssus gallinae) by essential oils /** Abstract ID: 302
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520 **Evaluation of sphinganine sphingosine ratio and liver weights of broilers exposed to Fumonisins /** Abstract ID: 141
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521 **Evaluation of three serological methods to monitor humoral response of a live bivalent Newcastle Disease vaccine /** Abstract ID: 215
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522 **Field study on production performances improvement in commercial meat turkeys by using a vector vaccine rHVT-ND compared with live vaccines program in France /** Abstract ID: 62
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534 **Husbandry factors affecting digestive microbiota diversity in organic broiler chickens /** Abstract ID: 267
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526 **In vitro evaluation of the anticoccidial potential of five ethanolic Tunisian plant extracts /** Abstract ID: 177
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528 **New generation of inert substances in D. gallinae control** / Abstract ID: 580
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529 **Phylogenetic Analysis of Newcastle Disease Viruses during 2012 to 2017 in Taiwan** / Abstract ID: 306
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530 **Prevalence of Mycoplasma synoviae in layer poultry flocks in Croatia** / Abstract ID: 639
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531 **Probiotic Properties of isolated lactobacilli from southwest and north west of Iran free range chicken intestine** / Abstract ID: 174
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533 **Retrospective evaluation of the economic cost of avian colibacillosis in broilers based on four years of records in a French production flow** / Abstract ID: 448
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534 **Screening of antiprotozoal action of plants extracts: in vitro and in vivo results** / Abstract ID: 71
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535 **The effect of Lactobacillus acidophilus,Thymus vulgaris and acetic acid in broiler chickens challenged with Salmonella enteritidis** / Abstract ID: 617
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538 **Vaccination against adenoviral gizzard erosion in chicken using a novel antigen delivery system** / Abstract ID: 430
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539 **Yeast cell wall effects on immunity, gut integrity and performance of broilers** / Abstract ID: 465
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541 **Behavioural Parameters and Final Body Weight of Slow-Growing Broilers With and Without Access to Pasture Area** / Abstract ID: 310
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542 **Feed additive sulfur to control ammonia** / Abstract ID: 615
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543 **Performance and eggs quality in layers fed with different percentage of Camelina sativa meal** / Abstract ID: 197
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544 **Practical experience with manipulable environmental enrichment material in modern poultry housing systems in Germany** / Abstract ID: 479
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545 **The use of optic fiber lightguides in cage batteries for laying hens** / Abstract ID: 91
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546 **The use of waste heat exchanger in commercial broiler production** / Abstract ID: 126
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549 **Animal health in different slow growing premium broiler genotypes** / Abstract ID: 391
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550 **An investigation into the bone mineralisation of meat chickens from hatch until three days of age** / Abstract ID: 223
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551 **Behavioral suitability of cocks with three different genetic backgrounds – dual-purpose hybrid, layer and heritage breed – for meat production** / Abstract ID: 428
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552 **Blood levels of cortisol and differential leukocytes as measure of stress among egg-type chickens on alternative housing systems** / Abstract ID: 611
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553 **Differential expression of magnum proteins in laying hens treated with corticosterone** / Abstract ID: 619
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554 **Effect of broiler hatching system and diet composition on indicators of welfare and performance** / Abstract ID: 127
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555 **Effect of intermittent lighting schedule on behaviour, food conversion ratio and performance in welfare tests in early life of chicks** / Abstract ID: 386
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556 **Effects of particle size and dietary levels of perlite on performance and tibia bone characteristics of broiler chickens** / Abstract ID: 568
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557 **Elimination of poultry red mites (dermanyssus gallinae) by using essential oils and their active ingredients** / Abstract ID: 601
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558 **Genotype Effect on Gentle, Severe and Aggressive Pecking Behaviours and Feather Condition in Laying Hens** / Abstract ID: 136
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559 **Improving Poultry Welfare in China** / Abstract ID: 339
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560 **Influence of dietary vitamin A levels on tibial dyschondroplasia in broiler chickens /** Abstract ID: 574
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561 **Keel bone fractures in laying hens affect laying performance but not egg quality /** Abstract ID: 103
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562 **Killing individual poultry on farm: a matter of animal welfare and feasibility /** Abstract ID: 134
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Types of Commercial Shell Eggs in Retail Markets in the Southeastern United States

Abstract ID: 605

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Based primarily on consumer concern regarding the well-being of commercial laying hens, the U.S. egg market is undergoing rapid change in which the diversity of types of eggs available for the retail marketplace is increasing. With this change, there is a lack of information regarding the range of egg types now available to the retail consumer. To determine the range of shell egg products available to consumers, a survey of retail stores was conducted. In a southeastern U.S. metropolitan area with a population of approximately 160,000, the types of eggs for sale in seven retail grocery stores was determined. Carton labels of shell eggs were observed, and egg type was determined by production system (e.g., traditional, cage-free, pasture-raised, organic, vegetarian), nutritional claims (e.g., omega-3 fortified, pasteurized), color (brown or white) and brand name. Excluding size and grade, a total of 26 different types of eggs were identified across the seven stores. The number of types of eggs varied among the stores and ranged from 6 – 20. Price also varied by egg type and ranged from US$1.80 - US$6.59 per dozen. Results of this study demonstrate that consumer choice of shell eggs has greatly increased, and suggest the need to provide information on the various egg types to consumers. Because retail sales ultimately will affect the evolution of commercial production systems, data from this study also provide insight into the technical and scientific gaps that must be addressed to maintain needed production levels.

Keywords: Eggs, Markets, Production, Retail

Addition of lysolecithin to broiler diets improves growth performance across fat levels and sources

Abstract ID: 350

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Fat inclusion in broiler diets provides a concentrated energy source, capable of increasing growth rates and efficiency while improving feed milling processes. At the same time, fat digestion can be impaired, due to insufficient lipase activity or bile salt secretion. Therefore, the use of lysophospholipid-based nutritional emulsifiers to increase the absorption of fats and other nutrients has been shown to be of practical and commercial interest. This study aimed to quantify the effect of increasing doses of lysophospholipid-based products (LPL) on nutrient availability and growth performance of broiler chickens. A total of 33 separate experimental reports were collated according to predetermined selection criteria to provide: 16 performance trials with LPL applied on top, and 17 performance trials with LPL applied in diets reformulated with reduced energy. Accounting for differences in product composition, product dose in feed was converted into the equivalent dose of lysolecithin (g LPL/tonne feed). Data on ADG and FCRc were analysed using the restricted maximum likelihood (REML) method with trial as a random effect. Overall, neither bird growth performance nor response to LPL supplementation were significantly affected by the ratio of unsaturated over saturated fatty acids of the dietary fats. In on top performance trials, FCRc was significantly reduced by LPL at 250 g/t compared to the control, with 125 g/t returning an intermediate value (1.599 control, 1.581 for 125 g/t LPL, 1.576 for 250 g/t LPL, $P=0.0516$). In reformulated trials FCRc was not significantly affected ($P=0.7099$), confirming that LPL supplementation at 125 and 250 g/t could recover the average dietary energy reductions of 57.88 and 73.11 kcal, respectively. In conclusion, this study shows that the addition of LPL at 125 g/t and above to broiler diets can consistently improve growth performance across a range of husbandry conditions, dietary main grains and fat sources.

Keywords: Broiler, Lysolecithin, Lysophospholipid, Meta-analysis, Performance

A multi-analysis evaluating the effect of Pedioccus acidilactici MA18/5M on performances of broiler chickens and laying hens

Abstract ID: 322

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The usage of *Pediococcus acidilactici* MA18/5M (PA) in poultry has been documented in literatures over the last twenty years. The objective of this multi- analysis is to evaluate the effect of PA in broiler and layers by assembling the results of the publications evaluating the effect of this probiotic. A total of 25 studies have been identified comparing the performances of poultry with or without the supplementation of PA. Each publication reports at least the two main performance criteria 1) average daily gain (ADG) for broilers or laying rate for laying hens and 2) feed conversion ratio (FCR). In total 14 studies in broilers chickens and 10 studies in laying hens are selected. Data are analyzed using a Mixed Model (SPSS 22.0) with treatment as fixed effect. Study was added as a random effect to the model. The multi-analysis shows a beneficial effect of the probiotic of the performances both of broiler chickens as well as laying hens. For broilers a significant higher production (+5.7% ADG, $P<0.05$) and improved FCR (6.3% better FCR, $P<0.05$) are recorded. For layers an improvement of the laying rate (+2.8%, $P<0.05$) as well as the feed efficiency (feed efficiency improved with 2.8%, $P<0.05$) is demonstrated. In addition supplementation of the feed with PA lowers the cholesterol content in blood and eggs of the supplemented birds. Results from this multi-analysis show that supplementation with PA can improve the performances of poultry, both in the case of broilers as well as laying hens.

Keywords: Broilers, Laying hens, Pedioccus acidilactici, Performances

A multi-enzyme complex with ferulic acid esterase improves apparent metabolizable energy and growth performance in broilers

Abstract ID: 576

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Non-starch polysaccharides (NSPs) present in the plant cell wall limit nutrient digestibility and thus the performance of animals. Addition of exogeneous hydrolases, (especially xylanase, cellulase and beta-glucanase) to the animal's diet help in degrading these NSPs in the feed. Esterified (di)ferulic acids also play an important role in digestibility of NSPs by influencing the chemical structure of this component. Feruloyl esterase (FAE) catalyzes the cleavage of these ester bonds, helping the main chain hydrolases in degrading the plant cell wall. The present study investigates the efficacy of adding a multi-enzyme product containing FAE, xylanase, cellulase, beta-glucanase and alpha-amylase in improving the performance of broilers, given a maize based diet. Two hundred and forty, 1-day old male Ross 308 broiler chickens were divided into three groups with 8 replicates per treatment and 10 birds per replicate. The trial lasted 35 days. A corn-soybean meal based control diet was provided as the positive control (PC) diet. A negative control (NC) diet was formulated to reduce apparent metabolizable energy, digestible aminoacids and crude protein by 65 kcal/kg and 2,5% respectively. The experimental group received the NC diet supplemented with the multi-enzyme product at 250 ppm. There was no significant difference in feed intake (FI), body weight gain (BWG) and feed conversion ratio (FCR) between the PC group and the treated group. Birds of the NC group performed significantly worse than those of the PC and treated group for FI (3319 g and 3413 g *vs.* 3618 g, respectively; $P<0.05$), BWG (2177 g and 2215 g *vs.* 2089 g, respectively, $P<0.05$) and FCR (1.53 and 1.54 *vs.* 1.73; $P<0.05$) In conclusion, the study shows that adding multiple enzyme preparations containing FAE improves performance of broilers fed cheaper low density diets making possible to match performance of broilers fed more expensive high density diets.

Keywords: Broilers, Ferulic acid esterase, Multi-enzyme complex, Performance

Analytical Hierarchy Process (AHP) used to hierarchize high fibre layer diet formulations

Abstract ID: 367

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The comparative evaluation of the efficiency of layer formulations enriched in fibre by the inclusion of alfalfa or alfalfa and sunflower meal was done using the findings of a 4-week feeding trial on 160, TETRA SL layers (56 weeks) assigned to 4 groups, using the analytical hierarchy process (AHP). The analysed diet formulations had 6% fibre, which resulted from: 11.95% alfalfa (E1); 11.95% alfalfa +0.15% enzyme (E2); 6% alfalfa + 15% sunflower meal (E3); 6% alfalfa + 15% sunflower meal +0.15% enzyme (E4). Layer performance (feed intake, feed conversion ratio, laying percentage and egg weight), and the microbiological load of the feeds and droppings were monitored throughout the experimental period. The analytical hierarchy process (AHP) is a structured method used to organise and analyse complex decisions, using mathematics and psychology. Rather than prescribing a "correct" decision, AHP helps the decision-makers to find one that fits best their purpose and the context of the problem. According to this methodology, a group of evaluators make pair comparisons which express the relative importance of the criteria set on the basis of the results produced by the analysed formulations. AHP users first decompose the decision problem into a hierarchy of sub-problems, which are easier to understand and which can be analysed independently. A coherent analysis presumes a panel of three evaluators/experts in the evaluated field and in related fields, which express their opinions on the basis of evidences (feeding trial results), on own experience and knowledge, with the purpose to evaluate the advantages and disadvantages of each of the four diet formulations. Three types of hierarchizing criteria were set: (c1) technical (laying percentage and average egg weight); (c2) economic (laying percentage and feed conversion ratio); (c3) ecologic (Escherichia coli and *Salmonella spp* counts in the compound feeds and droppings). Under the conditions of this experiment, formulations E3 and E4 performed the best.

Keywords: Alfalfa, Fibre, Hierarchize, Layer diets, Methodology

Antioxidant activity of grape seeds meal in broiler diets enriched in polyunsaturated fatty acids

Abstract ID: 338

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Winery by-products have high concentrations of polyphenols, which improve the oxidative stability of the meat, hence its quality. A feeding trial was conducted on 80, Ross 308 broilers, reared on the floor (14-49 days), using 3% grape seeds meal as a natural antioxidant in diets enriched in polyunsaturated fatty acids (PUFA). The broilers had free access to the feed and water and a 23 h light regimen. The basal ingredients of the conventional diet were: corn, wheat, gluten, soybean meal and 2% flaxseeds meal. Compared to the control formulation, the experimental formulation included 3% grape seeds meal, both in the growing and finishing stages. The grower (14-28 days), formulations had 17.19 MJ/kg gross energy, 20.60% crude protein and 5.73% ether extractives, while the finisher formulations (29-49 days) had 18.41 MJ/kg gross energy, 20.50% crude protein and 6.50% ether extractives for both groups. Flaxseeds meal had 69.04% PUFA, of which 53.77% Ω3, 15.27% Ω6 and 0.28 Ω6/Ω3 ratio. Grape seeds meal had 13.24% crude protein, 7.48% ether extractives, 29.45 mg gallic acid equivalent/g polyphenols and an antioxidant capacity of 143.31 mM Trolox equivalents/g sample. PUFA concentration in the feeds was 61.71% total fatty acids (C) and 69.54% total fatty acids (E) for the growing phase, respectively 59.75% total fatty acids (C) and 61.18% total fatty acids (E) for finisher stage. In the end of the feeding trial, samples of poultry blood were collected for serum biochemical determinations; six broilers per group were slaughtered and samples of meat broiler were collected and assayed for lipid degradation after 7 days of storage at +4^{0}C. In the experimental group, the peroxide value decreased by 3.37% in the breast meat, and by 9% in the thigh meat; fat acidity decreased by 2.46% in the breast meat, and by 0.03% in thigh meat, compared to control group. Blood glucose levels decreased from 241.00 mg/dL (C) to 214.85 mg/dL (E), cholesterol from 139.30 mg/dL (C) to 98.80 mg/dL (E) and triglycerides from 64.20 mg/dL (C) to 47.15 mg/dL (E). These results prove the antioxidant activity of the 3% grape seed meal which inhibits lipid degradation reactions, improving the oxidative status of broiler meat from the experimental group.

Keywords: Antioxidant, Broiler, Grape seeds meal, PUFA

A symbiotic supplement results in a propionic acid decline and limited microbiota shift in duck cecal content

Abstract ID: 390

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Pre- and probiotic supplements can be promising candidates for substituting antibiotic growth promoters in poultry production. Researches in this area have focused mainly on broiler chickens, whereas gut health of ducks has received less attention. A symbiotic mixture (containing inulin as a prebiotic and a *Bifidobacterium*, a *Lactobacillus* and an *Enterococcus* strain; each one of chicken origin) were tested on the composition of cecal microbiota and on cecal short-chain fatty acid (SCFA) production of Cherry Valley ducks. In total, 120 day-old ducks were purchased from a commercial hatchery and raised in deep litter pens. Ducks were allocated into two treatment groups (a control and a symbiotic) with six replicates of 10 ducks per pens. Two ducks were sacrificed on day 42 in each pen and samples were pooled for each pen after DNA extraction. For both treatment group, 6 samples were analysed with 16S rRNA gene targeted Illumina MiSeq sequencing. Overall, main bacterial phyla were *Bacteroidetes* (59% relative abundance, RA), *Firmicutes* (33% RA), *Proteobacteria* (4% RA) and *Actinobacteria* (1% RA). *Bacteroidia* and *Clostridia* represented 75% of the bacterial classes (56 and 19%, respectively). At the genus level, *Bacteroides, Prevotella, Faecalibacterium, Alistipes* and *Selenomonas* were most abundant (31, 16, 4, 3 and 3% RA, respectively). No differences were observed between the control and symbiotic group at phylum and class level ($P > 0.1$). However, less *Lactobacillaceae* ($P \leq 0.05$) was found in ducks received the symbiotic supplement. Propionic acid concentration was found lower ($P \leq 0.05$), whereas a trend was observed for lower total SCFA concentration ($P \leq 0.1$) in the symbiotic group. As a conclusion, symbiotic supplement altered duck cecal microbiome and microbial fermentation profile, however resulted in minor microbial shifts.
This work was supported by the CEPI (INTERREG V-A Austria-Hungary 2014-2020 Cooperation Programme) and EFOP-3.6.3-VEKOP-16-2017-00008 projects, which are financed by the European Union and the European Social Fund.

Keywords: Duck, Microbiome, Poultry, Short-chain fatty acids, Symbiotic

Bacillus subtilis in broiler diets with different levels of protein, amino acids and energy

Abstract ID: 152

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The current study investigated the impacts of *Bacillus subtilis* (BAS) inclusion in broiler diets with optimized nutrients content (standard) or with protein/energy deficiency (PED), on nutrients digestibility and growth performance. Six experimental diets, a diet with optimized nutrient content and two diets with different levels of protein-, AA- and -energy deficiency, without or with BAS (1.6 × 109 CFU/kg for starter and 0.8 × 109 CFU/kg for grower diets), were fed to broilers for 42 days. Apparent ileal digestibility (AID) of crude protein (CP), ash, starch and gross energy (GE) was determined at the end of the experiment. The impacts of BAS inclusion in standard broiler diets on gut histology, bacterial activity and composition were also investigated. Performance and AID data were assessed by ANOVA using GLM procedure, applying a 2 (BAS presence/ absences) × 3 (nutrients levels) factorial arrangement of treatments. Histology, bacterial metabolic activity and composition data obtained from chickens received standard diets without (S) and with BAS (SB), were subjected to an independent Student's t-test.
The PED in the diets was enough to cause nutritional stress and negatively influence growth performance. Supplementation of BAS in the diets with PED, ameliorated growth performance, which seemed to be caused by improvements in AID of CP, starch and GE as results of adding BAS to PED diets ($P \leq 0.05$). Comparing only S and SB experimental groups, displayed no effect on bacterial metabolism and composition in the ileum and caecum, except a decline in number of ileal lactobacilli and a decline in caecal L/D-lactate ratio for SB group. Furthermore, inclusion of BAS in S diet shallowed duodenal crypt depth (CD) and increased duodenal villus heigth to CD ratio while, it did not affect other histological variables in the duodenum, jejunum and ileum. In conclusion, adding BAS to the diets of broilers under nutritional stress may be able to retrieve growth performance by improving nutrients digestion and availability. However, the biological mechanisms for these beneficial impacts deserve to be studied further.

Keywords: Bacillus subtilis, Energy deficiency, Nutritional stress, Protein deficiency

Beneficial effects of over recommended dietary alpha-tocopherol acetate on reproductive performance of overweight roosters

Abstract ID: 57

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This study aimed to investigate the effect of dietary supplementation of over recommended alpha-tocopherol acetate on reproductive performance of Ross 308 male broiler breeders. In all, 60 roosters (5 months old) were randomly allotted to a 2×5 completely randomized factorial design with 6 birds in each. Included factors were 5 levels of vitamin E (VE; 0, 100, 200, 300 or 400 mg/kg feed) and 2 weight groups of roosters (standard and heavy, SW and HW respectively). To have heavy weigh roosters, half of experimental birds fed 30% more than standard requirement for 5 weeks naming adaptation period. Semen quality parameters, including sperm total and forward motility, plasma membrane integrity and functionality, semen concentration, sperm abnormality and ejaculate volume were recoded weekly for 10 weeks while semen MDA and blood testosterone concentrations were assessed in the 5th and 10th weeks of trail. Fertility rate was estimated using collected eggs following AI during the 11th and 12th weeks. According to the results, feeding different levels of VE dose responsively improved semen parameters, blood testosterone concentration and body weight with VE400 level achieving higher value. Except for a positive relationship observed in body weight groups with semen concentration and ejaculate volume, other traits were negatively correlated with body weight. Considering the interactive effects of BW, VE and time, sperm total and forward motility as well as ejaculated volume were not affected, but other traits were significantly changed by varying degrees. Dietary supplementation of 400 mg VE could significantly improve semen concentration, plasma membrane functionality, sperm abnormality, semen malondialdehyde (MDA) and blood concentration in both SW and HW groups compared to respective control groups. Regarding the interactive effect of BW, VE and time, blood testosterone and semen concentrations had a significant upward trend, however, both MDA levels and sperm abnormality experienced a significant downward trend in VE treated groups during the experiment. Body weight increment had no significant effect on fertility rate, however, it was dose responsively increased by VE supplementation in both SW and HW groups. To conclude, reproductive performance of roosters in both standard and heavy weight groups were beneficially affected by different levels of dietary VE in the current study.

Keywords: Fertility, Heavy weight, Rooster, Standard weight, Vitamin E

Biological characterization of a plant extract containing 1,25-dihydroxyvitamin D3-glycosides in poultry

Abstract ID: 167

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In three experiments in different species of poultry, the biological activity of a standardized extract of the plant *Solanum glaucophyllum* (SG) was explored. SG is known to contain high levels of 1,25-dihydroxyvitamin D_3-glycosides (1,25DHVD). Dried leaves of SG were extracted with water/ethanol and standardized with maltodextrin to an analytical content of 50 µg 1,25DHVD /g. This mixture was formulated into a fully water-soluble powder that can easily be applied via the drinking water.The biological activity was estimated in a bioassay with Japanese quails. Laying quails received a vitamin D free diet until laying rate dropped below 10 %. At this time-point the animals were repleted by adding vitamin D_3 (VD3) or the test extract in ascending doses. Regain of laying is earlier with the extract than with regular VD3. This is because VD3 needs two metabolic conversions to become the active form 1,25DHVD, while the extract already contains the metabolic active form. A single dose of the extract was applied to male Ross broilers of 1 kg body weight and blood serum concentration of 1,25DHVD was measured. Plasma kinetics showed a peak value between 6 and 12 hours post-application with an increase of 180% as compared to the pre-application level. In contrast, a single dose of synthetic 1,25DHVD resulted in a blood maximum within 1-2 hours. This difference can be explained by the necessary cleavage of the glycosides before absorption. 66 weeks old LSL laying hens fed a commercial layer diet containing 75 µg/kg VD3 were allocated to 4 treatment groups. control; 47 mg extract/animal and day for 5 consecutive days via drinking water (T1); 5x T1; 10x T1 and 5x T1 but for 10 days, respectively. No adverse effects were observed. Only animals in group 5x T1 for 10 consecutive days showed a slight reduction in laying rate. Plasma calcium remained unchanged, whereas plasma phosphorus showed a small increase. Plasma 1,25DHVD was increased in a dose-dependent manner. A tendency to a slight elevation of 25-hydroxyvitamin D (25HVD) was observed in the high doses.The extract from SG provided a faster onset of action compared to VD3 but uptake was delayed in comparison to synthetic 1,25DHVD, reducing the risk of hypercalcemia. In layers the observed increase of 1,25DHVD did not result in significantly elevated serum Ca-levels and did not influenced 25HVD level.

Keywords: 1, 25-dihydroxyvitamin D3-glycosides, Plant extract, Plasma kinetics, Vitamin D activity

Broiler chicken pigmentation is influenced by maize hybrid as the only source of carotenoids

Abstract ID: 250

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Increased concern about use of synthetic additives in food chain led to search for suitable natural sources in broiler pigmentation. These natural sources could considerably increase production costs while carotenoids from maize, a major component in diet, remain neglected. The aim of this research was to explore commercial maize hybrids suitable for increased broiler pigmentation. In total, 325 1-day-old male Ross 308 broiler chicks were allocated in 25 pens, and pens were in complete randomized design assigned to one of five dietary treatments (5 treatments×5 cages) differed only in maize hybrid. Four experimental (Bc 572, Kekec, Mejaš and Riđan) and one control (unknown hybrid) diets were not supplemented with pigments. On day 32 of the trial, broilers were humanely euthanized, and colour according to the CIE L*a*b* was determined in chilled breast meat and skin, shank skin and abdominal fat using a Minolta CR-410 Chroma meter. Maize hybrids differed in carotenoid profile which resulted in variable daily intake ($P<0.05$) of lutein [329 (Bc 572) – 639 μg (Riđan)], zeaxanthin [397 (Riđan) – 885 μg (Bc 572)], β-cryptoxanthin [70 (control) – 148 (Bc 572)] and β-carotene [100 (Kekec) – 154 (Bc 572)]. Lightness scores were similar among treatments (59.03, 65.98, 69.57 and 73.21 for meat, skin, fat and shank, respectively). Redness was significantly affected by maize hybrid only in shank skin; control had the lowest (4.70) while Bc 572 had the highest value (7.99) in accordance with increasing zeaxanthin and β-cryptoxanthin intake ($r=0.54$ and 0.65, respectively, $P<0.01$). Treatments showed the highest impact on broiler pigmentation in yellowness score ($P<0.05$) with obtained ranges: 14.50-17.14 for meat, 18.28-22.69 for skin, 19.19-21.51 for fat, and 51.77-58.57 for shank. Control had lowest values in all ranges while Riđan and Bc 572 had the highest values in meat and fat. Among commercial hybrids, there are ones with high carotenoid content providing cost-effective pigmentation in broiler chickens.

Keywords: Broiler pigmentation, Carotenoids, Maize hybrid

Brown macroalgae fermented in vitro by broiler cecal microbes

Abstract ID: 202

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The brown macroalgae *Saccharina latissima* contains bioactive polysaccharides not found in terrestrial plants, such as the β-glucan laminarin, that are known to promote gut health. Whole *S. latissima* harvested in April (5% of dry matter laminarin), whole *S. latissima* harvested in June (20% of dry matter laminarin) and a laminarin extract (90% purity) from *S. latissima*, were tested for their *in vitro* fermentation characteristics to evaluate their prebiotic potential compared to inulin. Six replicates of 1 gram organic matter of each substrate were fermented in 100 ml Duran bottles containing a buffer and broiler (Ross 308, 38 days of age) cecal content that had been filtered through double layers of cheese cloth under CO_2-flow to maintain anaerobic conditions. Fermentation was allowed to continue for 30 hours while gas volume was recorded, and samples for short chain fatty acid (SCFA) ratios were taken after 4, 6 and 30 hours. There was no difference in the fermentation characteristics between the two whole algae of different harvest times, both fermented fast (no gas produced after 6 hours), while laminarin and inulin produced slowly progressing gas profiles up to 30 h. Yet both the fast and slowly fermenting substrates produced similar final gas volumes, around 70 ml. At the end of fermentation, the SCFA profile of the whole algae had resulted in mainly (55%) propionate while laminarin and inulin resulted in mainly acetate (53%). The proportion of butyrate, an important enterocyte energy substrate, was higher from laminarin fermentation (24%) than from inulin (20%, $P<0.001$). Laminarin fermented the slowest and also produced a higher final SCFA concentration of 107 mmol/L than inulin (92 mmol/L, $P=0.012$). Thus laminarin extracted from *S. latissima* shows potential similar or superior to that of inulin as a future prebiotic. Whole algae might be of interest for this purpose too but their potential was not evident from this study, they may hold other valuable physiological and nutraceutical characteristics.

Keywords: Brown algae, In vitro fermentation, Prebiotic

Ca and P Requirements of Broiler Chickens?

Abstract ID: 629

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A large scale broiler trial was conducted to evaluate Calcium (Ca) and Available Phosphorus (Pa) requirements of broiler chickens in research facilities of Beypiliç, one of the biggest broiler integrator in Turkey. Total 11200 Ross 308 broiler chicks were randomly distributed into 7 dietary treatments, each has 8 replicates with 200 broiler chicks. Current 2014 Ca and Pa recommendation of Ross 308 broilers was considered as positive control (treatment 1) for starter, grower and finisher phase. The rest 6 dietary treatments (2 to 7) were formulated to meet 93.8% of Ca and P level of positive control for starter period, and 93.4, 90.8, 89,8, 87.2, 87.2 and 85.6 % respectively for grower and finished phase.

Body weight (BW), body weigh gain (BWG), feed intake (FI), feed conversion (FCR), mortality, and tibia ash and P content were determined to evaluate and examine the effects of reduced levels of Ca and Pa in broiler chicks. Growth performance and also tibia parameter results of the experiment showed that all of the birds fed the reduced Ca and Pa diet did not show any growth depression and impairment. Besides it was interestingly obtained that all reduced Ca and Pa diets had better BWG and FCR than positive control ($P<0.05$). No significant differences was observed among the treatments for FI and mortality ($P>0.05$). There was also no significant differences among the groups in ash and phosphorus content of the tibia ($P>0.05$). The tibia ash content was between 35.07 to 36.59% and tibia P level was between 17.31 to 17.36% among the treatments.

The results of the present study showed that the current Ca and P recommendation of Ross 308 broilers could be higher than real requirement in practical conditions, and needs to be updated. So it was concluded that at least 10 % reduction can be possible in dietary Ca and Pa level of broiler diets after starter period, and this would not cause any impairment in broiler growth and bone development.

Keywords: Available phosphorus, Broilers, Calcium, Performance, Tibia ash

Charcoal in the feed or litter and its effects on nitrogen retention and performance of intensive broilers

Abstract ID: 47

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The influence of adding activated charcoal to feed and litter was tested in a broiler barn regarding the production performance of 5400 Ross 308 broilers as well as nitrogen retention in the litter. The as hatched one day old chicks were evenly distributed to 20 compartments and randomly allocated to five different treatments, allowing 4 compartment replications with 270 animals each. They were slaughtered at the age of 37 days. Treatments consisted in a) a control group receiving standard feed (C), b) adding every third day about 0.3 kg/m2 charcoal to the litter (CL), c) adding 1% charcoal to the feed (CF), d) combining a) and b) (CLCF) and e) adding 2.6% of a carbon feed containing charcoal and effective microorganisms (EMC). All the groups received the same feed, based on an organic type recipe without any synthetic amino acids, but with conventional components (starter: 12.5 MJ ME, 21.5% CP; grower: 13.0 MJ ME, 22.1% CP, finisher: 13.1 MJ ME, 19.7% CP). After 37 days, CF and EMC were significantly heavier than the others (C: 1800 g BW, CL: 1788 g BW; CF: 1903 g BW; CLCF: 1848 g BW; EMC: 1895 g BW). Their FCR was significantly higher as well, due to spillage of feed into the litter, which amounted to an estimated 10-20% (C: 1.72, CL: 1.69; CF: 1.99, CLCF: 1.84; EMC: 2.06). Mortality was not influenced. The litter was dryer and less encrusted with the addition of charcoal to feed or litter. CF and EMC lead to signif-icantly less foot pad lesions than the control and CL (C: 56.3%, CL: 65%; CF:13.8%; CLCF: 26.3%; EMC: 3.8%). Significantly less ammonia nitrogen was found in CL, CF and CLCF than in the control. EMC lowered it slightly (C: 7.8 g/kg DM, CL: 5.9 g/kg DM; CF: 6.1 g/kg DM, CLCF: 6.0 g/kg DM; EMC: 7.0 g/kg DM). In conclusion, adding charcoal or EM carbon feed to a standard feed improved broiler performance, litter quality and foot health. Charcoal as feed or litter additive can lower the ammonia nitrogen contents in the litter and thus lower ammonia emissions. Further investigations, especially with layers, are necessary to confirm reduction of long term ammonia emissions. Additionally, the uniformity of charcoal structure and quality as well as the process of fabrication should be looked into in order to improve the documentation of the effect of charcoal as a feed and litter additive.

Keywords: Broiler, Charcoal, Feed, Nitrogen retention

Clinoptilolite clay relieves broilers from footpad lesions

Abstract ID: 552

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Clinoptilolite is known for the good cation exchange capacity and can be used to reduce the amount of harmful ammonia produced in the intestinal tract from fermentation of undigested protein. Clinoptilolite binds ammonia on gut level, preventing the need to remove ammonia via the liver and kidneys into urine. Clinoptilolite also binds water and together with the increased gut health by capturing ammonia it results in less wet faeces and therefore lower amounts and less severe footpad lesions. The effect of clinoptilolite on footpad lesions and animal performance was tested in two trials. Trial 1 entailed a practical trial of 37 days with two broiler houses (each containing 14.000 broilers, Cobb500), located in Germany executed by Agroproduct Qualitätssicherungs-GmbH. Trial 2, was performed in Belgium by Zootechnical centre KU Leven, with 900 Ross 308 broilers and lasted 42 days. Both trials compared a commercial broiler diet with a similar diet diluted with 1% clinoptilolite*. Trial 1 measured performance, footpad scores from 0 (no lesions) to 2 (severe lesions) and litter quality from 1 (dry, crumbly) to 5 (wet). Trial 2 measured performance and footpad lesions from 0 to 4 (with increments of 1): 0 being no lesions, 4 meaning severe lesions. Results of trial 1 showed a higher daily weight gain of 0.5 g/bird/day in the clinoptilolite vs. control group (average daily gain 65.1 g/d in clinoptilolite group vs. 64.6 g/d in control group). Feed intake and feed conversion ratio were similar for both groups. Smaller wet areas in the clinoptilolite farm house vs. control resulted in better litter scores in clinoptilolite group vs. control (2.3 vs. 2.9). Performance data in trial 2 showed no significant differences between control and clinoptilolite group. There were better footpad results for the clinoptilolite group vs. control group: no severe lesions (score 2) and more score 0 and 1 vs. control. Results from day 28 showed significantly higher incidence of low scores for footpad lesions (score 0) and significantly less score 3. It was concluded from these two trials that reducing the energy content and price of the feed by dilution with 1% clinoptilolite did not cause impaired growth of the birds and the addition of clinoptilolite improved litter quality and footpad lesion scores.
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Keywords: Ammonia, Broiler performance, Clinoptilolite, Footpad health

Combining acidic and neutral proteases improved growth performance in broilers

Abstract ID: 575

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A trial was conducted on straight-run broilers to evaluate the combined effect of acidic and neutral proteases on growth performance of broilers. Average 3% reduction in amino acids was made in negative control diet (NCD) as compared to positive control diet (PCD) using a commercial calculator, and this diet was then supplemented with combined proteases. This trial was lasted for 35 days under normal conditions of temperature and humidity. Growth performance was evaluated as BW, FCR and European Performance Index (EPI). Though statistically non-significant changes ($P>0.05$) in growth parameters, however protease never exceed positive control diet in d-21 and d-35. A little improvement ($P>0.05$) in BW at d-35 was observed by supplementing combined protease in NCD (2243 vs. 2205) with similar pattern in FCR (1.53 vs. 1.55). Conclusively such improvement in performance can be evaluated economically at commercial level for further use of proteases.

Keywords: Broiler, Growth, Protease

Comparative Effects of Sodium Salts (Sulphate vs Bicarbonate) in Diets for Broiler Chickens

Abstract ID: 438

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A study was conducted to evaluate the use of sodium sulphate (Sodifeed®) in broilers feeding compared to sodium bicarbonate.
Four hundred and eighty 1-day-old male Ross 308 broilers were distributed into 16 floor pens, at 30 per pen. Broilers were fed *ad libitum* a starter diet from 1 to 21 days (22.2% CP, 3050 kcal/kg AME) and a grower diet from 21 to 34 days (20.5%. CP, 3150 kcal/kg AME), both in 3 mm pellet form.
Two treatments were tested according to sodium supply, T-1: 0.30% sodium bicarbonate plus 0.31-0.32% sodium chloride, and T-2: 0.30% sodium sulphate plus 0.27-0.28% sodium chloride. The two diets were formulated to supply the same sodium content and Dietary Electrolytic Balance (DEB = 250 mEq/kg).
The broiler performance (growth, feed intake, feed to gain ratio) was evaluated at 21 and 34 days of the experiment. The characteristics of litter were also evaluated at 21 and 34 days. At the end of the trial, the carcass and breast yields and the tibia bone ash percentage of two birds per replicate were determined. The drip loss of breast meat were measured after 7 days of storage at 4ºC.
Data were statistical evaluated by an analysis of variance and treatment means were compared using t Student test ($P<0.05$). No statistical differences between treatments were found on productive parameters along the experimental periods evaluated. Litter characteristics (dry matter, ammonia concentration and pH) were similar ($P>0.05$) irrespective to dietary treatments tested. The results of carcass (80.8%) and breast meat yield and ash percentage of tibia bone of broilers 39 days old showed no differences between both diets. After 7 days of cold storage the drip loss were also similar (12.5% on breast meat of broilers fed bicarbonate and 12.4% on breast meat of broilers fed sulphate).
Results of the present experiment suggest that the sodium sulphate salt could be comparable to sodium bicarbonate as sodium supply to high DEB diets at lower cost.

Keywords: Broiler performance, Carcass yield, Litter, Sodium bicarbonate, Sodium sulphate

Computed tomography (CT) in evaluation of different inorganic phosphorus sources in broilers diet

Abstract ID: 378

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The aim of this trial was to determine the impact of monobasic calcium phosphate (MCP), as source of inorganic phosphorus in broilers diet, on production performances and bone mineralization, by using computed tomography (CT). The trial included 300 broilers (Cobb 500), both sexes, from the same hatchery. Broilers were divided in three groups, by one hundred birds each. During the trial, which lasted 42 days and was divided in three phases (1-21, 21-35 and 35-42 days), groups were fed with different experimental diets. The low-P basal diet was formulated without addition of monobasic calcium phosphate, and the other 2 diets was formulated with addition of 2 different MCP (one was provided by "Elixir Group" D.O.O., Šabac, Serbia, and the other one was originating from Russia). Production results (average body weight, average daily gain, feed intake and feed to gain ratio) were monitored during the trial. At the end of experiment, on the forty-second day, right legs were collected from 6 birds in each group, in order to obtain the samples of tibia. Measurements were carried out by the CT, and the tissue density (radiodensity) was expressed in Hounsfield units (HU). Additionally, linear bone parameters were measured, such as bone length and inner and outer radii of the cross-section at half of the dyaphisis length, in order to calculate cross sectional area of medullar cavity and cortex thickness. Both experimental groups that received MCP, as a P source achieved significantly higher production results compared to the group of broilers fed with basal diet ($P<0.001$). Furthermore, the differences were observed between the two experimental groups - production results, as well as bone tissue density of the first experimental group (fed with addition of MCP – Elixir), was significantly higher, compared to the group fed with MCP from Russia ($P<0.05$). Tibia length and cross sectional area in the same group was higher, but without statistical significance ($P>0.05$). The results of the experiment indicated that MCP, as inorganic P source has significant impact on growth, production performances and level of mineralization of bones, and CT can be used as one of the methods in evaluation of inorganic phosphorus sources.

Keywords: Broilers, Computed tomography, Monobasic calcium phosphate, Nutrition, Phosphorus

Content and deposition of carotenoids in egg yolk from hens fed diets differentiated in maize hybrid

Abstract ID: 254

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Maize is the main energy feed material in hen's diet, and the only major component with relevant carotenoid content. Carotenoids give yellow to orange colour of egg yolk, however, contribution of maize carotenoids to yolk pigmentation is neglected. There are a variety of maize hybrids available for planting that contain different carotenoid profile and this study aimed to explore the effect of six maize hybrids in 56%-proportion in diets without added pigments on the deposition of individual and total carotenoids (TC) into egg yolks. In total, 90 Tetra-SL hens, by three in each cage, were allocated to six dietary treatments in complete randomized design (6 treatments×5 cages). After depletion period, hens were fed five experimental (Bc 572, Kekec, Mejaš, Riđan and Pajdaš) and one control (unknown hybrid) diets for 12 weeks. Eggs were collected every three days until stabilisation, determined using yolk colour assessed with Minolta CR-410 Chroma meter, and then once weekly until the end of 12th week. Carotenoid profile of diets and egg yolks was determined using HPLC method, and values from day 35 were used for calculations of deposition deficiencies. Redness was the colour parameter the most sensitive to changes in yolk carotenoid profile, and it stabilised after day 13 of the trial [$P<0.001$, 8.11 (control) – 13.24 (Bc 572)]. Carotenoid profile in yolks reflected one in maize hybrids with highest contents of lutein (13.29 µg/g) and β-carotene (0.78 µg/g) in eggs from hens fed with Riđan while eggs from hens fed with Bc 572 had highest contents of zeaxanthin (15.85 µg/g) and β-cryptoxanthin (0.93 µg/g). Riđan also obtained highest deposition efficiencies of zeaxanthin and β-carotene (28.51% and 8.85%) while Kekec and Bc 572 were the most efficient in deposition of β-cryptoxanthin (11.65%) and lutein (28.65%), respectively. Mejaš was the most efficient in deposition of TC due to the high values obtained for lutein and zeaxanthin while control was the least efficient in deposition of all carotenoids. Experimental maize hybrids had sufficient carotenoid content to achieve high redness values of egg yolks with high deposition efficiencies providing a suitable replacement for pigment supplements in hen's diet.

Keywords: Carotenoids, Deposition efficiency, Egg yolk, Laying hens, Maize hybrid

CuO nanoparticles influence on quail physiological parameters

Abstract ID: 106

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The research was carried out to investigate CuO nanoparticles influence on japanese quails (Coturnix coturnix japonica) physiological parameters. For experiments was used japanese quails (Coturnix coturnix japonica). Quails was grown from first day to 12 weeks age. For the trial quails were divided in to three groups. First group was served as control and quails were fed with standart feed. Feed for quails in second group was enriched with inorganic $CuSO_4 \times 5H_2O$ additive. Feed for quails in third group was enriched with CuO nanoparticle additive. Following parameters was investigated: total protein level in blood serum, glucose level in blood serum, alanine aminotransferase (ALT) level (activity) in blood serum, aspartate aminotransferase (AST) level (activity) in blood serum, ceruloplasmin level in blood serum, influence of Cu additives for quails growing. The experiment and analysis of the study showed: total protein level in blood serum increased. Highest level of total protein was observed in 7 week age quails blood serum in 3-rd trial group increased 16,83% in comparison with control group ($P<0,05$). Highest level of glucose in blood serum was observed in 12 week age 3-rd trial group increased 4,63% in comparison with control group. Highest increasing of alanine aminotransferase (ALT) level in blood serum was observed in 12 week age 3-rd trial group: increased 3,69% in comparison with control group. Highest increasing of aspartate aminotransferase (AST) level (activity) in blood serum was observed in 12 week age 3-rd trial group: increased 6,33% in comparison with control group ($P<0,05$). Highest increasing of ceruloplasmin level in blood serum was observed in 7 week age 3-rd trial group: increased 9,9% in comparison with control group ($P<0,05$). Highest growing was observed in 3-rd trial group: 7 week age weight increased from 206,73 to 218,12 g, i.e. increased 6,70% in comparison with control group ($P<0,05$).

Keywords: Blood parameters, Cooper, Nanoparticles, Quails, Trace element

Daily egg mass production and associated utilization of dietary calcium in laying hens of different genetic origin

Abstract ID: 242

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The objective of this study was to examine the utilization of dietary calcium (Ca) by hens from high and low performing lines and characterize potential line-dependent differences in hens' Ca metabolism. Thirty-six hens from four genetically diverse lines (WLA/R11: high/low performing white layers; BLA/L68: high/low performing brown layers; n=9 hens per line) were housed in metabolic cages and ad libitum fed with a commercial diet (43.8 g Ca/kg DM). In the 42^{nd} week of age a Ca balance trial was performed for 5 days. Birds were weighed at the beginning and end of the trial, while residual feed as well as weight and composition of laid eggs were recorded daily. Excrements of each bird were collected twice a day. Balance parameters were related to birds' metabolic body weight ($kg^{0.75}$). Total Ca retention was calculated by subtracting Ca excretion from intake, whereas Ca retention in eggs was estimated by multiplying daily shell, yolk and albumen production with their average Ca concentration (370, 1.4, 0.11 mg/g shell, yolk, albumen). Subtracting Ca retention in eggs from total Ca retention remaining Ca retention in body was calculated. The quotient of daily Ca retention in egg and daily Ca intake described Ca utilization for egg synthesis. Statistical analysis was performed as one-factorial ANOVA with "line" as fixed effect. Daily Ca intake and excretion did not differ between high and low performing hens with the exception for significantly lower Ca excretion by WLA hens ($P<0.05$). According to higher daily shell production ($P<0.01$), high performing hens absolutely retained more Ca in eggs than low performing ones ($P<0.05$). But, both high and low performing hens deposited approximately 72% of retained Ca in eggs and did not differ in remaining Ca retention in body. However, due to lower daily Ca excretion, high performing WLA apparently utilized dietary Ca more efficient for egg synthesis than low performing lines ($P<0.01$). In conclusion, this study indicates that slight increases in dietary Ca consumption and especially a more efficient utilization of dietary Ca for egg synthesis ensure higher Ca retention in eggs of intensively selected, high performing hens. However, to understand the mechanisms of obviously selection-induced adaptations in hens' Ca metabolism further research is necessary.

Keywords: Balance Study, Calcium Metabolism, Daily Egg Mass, Genetic Background

Determination of microbiocoenoses in the intestine of the Hisex Brown hens in ontogenesis

Abstract ID: 599

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Microbiocoenoses in the gastrointestinal tract, especially in the ceca, play an important part in life processes of poultry. Identification of the structure and taxonomic composition of microorganisms in the cecum using molecular genetic methods serves as a crucial approach in understanding how a cecal microbiota interplays with the chicken organism during ontogenesis. For this purpose, we studied an intestinal bacterial community composition in the ceca of the Hisex Brown laying hens at age of 40, 155 and 315 days using T-RFLP and RT-PCR. In the chickens studied, development of the cecal microbial communities, changes in their content, and appearance of new microorganisms occurred in the ontogeny. A broader spectrum of bacteria was found in 40- and 155-day-old birds (221±11 and 258±9 phylotypes, respectively) as compared with 315-day-old laying hens (178±8 phylotypes). Also, 315-day-old birds showed the least content of unidentified phylotypes. In the ceca of adult hens, there was a change in the dominant microbial taxonomic groups including a higher proportion of acid-utilising bacteria of the class Negativicutes and cellulolytic bacteria of the class Clostridia, with a lower content of the classes Bifidobacteriales and Bacillales. Lactobacteria (order Lactobacillales) showed a greater content in 315-day-old laying hens (33.15±1.05%) as compared with 40- and 155-day-old birds (5.13±0.23% and 24.58 ± 0.86%, respectively). The variety and number of bacteria in the ceca conventionally attributed to various pathogens of poultry diseases, including the genera ***Enterobacter, Pantoea, Listeria, Acinetobacter*** and ***Mycoplasma***, families ***Campylobacteraceae*** and ***Pasteurellaceae***, and phylum Fusobacteria, increased with the age of birds. Thus, in the course of molecular genetic studies, the species composition and dynamics of the microbiocoenoses in the cecum of the Hisex Brown laying hens was determined as related to their ontogeny.
This research is supported by a grant of the Ministry of Education and Science of the Russian Federation, Contract No. 14.W03.31.0013.

Keywords: Hisex Brown hens, Microbiocoenoses, T-RFLP

Dietary black soldier fly as suitable ingredient for Muscovy duck: preliminary results on carcass and breast meat traits

Abstract ID: 281

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Many studies have been conducted in poultry to substitute the conventional protein sources (fishmeal and soybean meal) with insect meals, due to their high protein content and the good and balance amino acid profile. The aim of this study was to evaluate the effect of dietary administration of a partially defatted black soldier fly larva (BSF) meal on carcass characteristics and breast meat quality in Muscovy duck (*Cairina moschata domestica*). A total of 256 females broiler Muscovy ducklings (average live weight, LW: 71.32±2.70g) were randomly allotted in 32 pens (8 replicates/treatment) and reared from day 3 to day 48. Four isonitrogenous and isoenergetic diets were formulated with increasing inclusion level of BSF (0, 3, 6 and 9%; BSF0, BSF3, BSF6 and BSF9, respectively) in substitution of corn gluten meal and divided in 3 feeding phases: starter (1-14d), grower (14-35d) and finisher (35-48). At day 48, 2 animals/replicate were slaughtered and dissected to determine their carcass yields. The weights of spleen, bursa of Fabricius, liver, heart and abdominal fat were recorded. Breast and thigh muscles were then excised from 16 ducks/treatment and weighted. Ultimate pH (pHu) and L*, a*, b* colour values were then measured on breast muscle. Data were analysed by one-way ANOVA evaluating the effect of dietary BSF inclusion level by polynomial contrasts. Significance level was set at $P<0.05$. No significant difference was observed for final LW (2515.68±92.42g on average) among groups. Hot and cold carcass weights showed a quadratic response ($P<0.05$) to increasing BSF larva meal, with a minimum corresponding to BSF6; however, refrigeration losses were not affected by treatments (2.01±0.08% on average). No significant effects, related to BSF meal utilization, were observed for the weights of spleen, bursa of Fabricius, liver and heart. The weight of abdominal fat showed a quadratic response to increasing BSF larva meal with a minimum corresponding to BSF6 ($P<0.05$). Breast and thigh yields did not differ among groups (20.37±0.21% and 29.53±0.53% on carcass weight, respectively). As well, pHu and L*, a*, b* colour values were not different among groups. The obtained results showed that the inclusion of BSF meal, with the exception of BSF6, did not affect carcass characteristics and meat traits, confirming the potentiality of BSF meal in Muscovy duck.

Keywords: Cairina moschata domestica, Hermetia illiucens, Insect meal, Meat quality

Dietary camelina cake beneficially affect the fatty acids profile of immune tissue in broiler chicks

Abstract ID: 383

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Nowdays, there is an increased interest to manipulate the n-3 polyunsaturated fatty acids (PUFA) profile of poultry meat in order to increase the consumption of these fatty acids by humans. Moreover, there is some concern that the n-3 PUFA enriched diets may detrimental affect immunity function in chickens. Little is known about the effect of camelina cake (CC), as rich source of n-3 PUFA, on fatty acids profile of immune tissue in broiler chicks. The study was conducted to evaluate the effects of CC in broiler finisher phase diets on immune organs (spleen, thymus and bursa of Fabricius). Three-wk-old Cobb 500 broilers (N=600) were randomly assigned for 20 d experimental period into 2 groups. Broilers were fed with isocaloric and isonitrogenous diets: control (C; corn-soybean meal-canola meal based diet) and experimental (CC; that contained 80 g camelina cake/kg diet and replace canola meal from based diet). At slaughter, spleen, thymus and bursa of Fabricius were removed, weight and collected from bird (n=12 per each treatment). The fatty acids profile of immune tissue was determined by gas chromatography. Results shown that the fatty acid profile of immune organs reflected the composition of the diet. Feeding diets with CC increased the total content of n-3 PUFA in all immune tissue (> 4.03 fold in spleen, 3.24 fold in thymus and 3.29 fold in bursa; $P= 0.0001$) compared with C diet. Conversely, the total level of n-6 PUFA decreased in bursa (< 1.51 fold; $P= 0.0001$), thymus (< 1.45 fold; $P= 0.0001$), spleen (< 1.28 fold; $P= 0.002$) as effect of CC diet. From the total of n-6 PUFA, the higher concentration of arahidonic acid was found in spleen (2.11%; $P= 0.013$), thymus (2.05%; $P= 0.041$) and bursa (0.59%; $P= 0.007$) of chicks fed CC diet. The dietary CC inclusion decreased n-6:n-3 ratio at recommended level for health (4.87% in thymus, 3.72% in bursa and 3.64% in spleen; $P= 0.0001$), compared with C diet. In conclusion, our study suggested that feeding camelina cake in broiler chicks diets beneficially affect the fatty acids profile of immune tissue and may be an effective tool to improve the immune status.

Keywords: Broilers, Camelina cake, Immune organs, N-3 fatty acids

Dynamic modulation of intestinal microbiota induced by tannins and antibiotics feed additives in poultry

Abstract ID: 454

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Antibiotic growth promoters (AGPs) have been used for decades in animal production as a means to maintain animal health and improve feed efficiency. Global concern about emergence and transference of antimicrobial resistance is rising and therefore the development and testing of alternatives to AGPs is urgently needed. Information about the effects of phytogenic compounds on chicken intestinal microbiota is required to understand the potential of these bioactive natural products as well as their mechanisms of action. The aim of this study was to evaluate and compare the effects of a blend of tannins and antibiotics treatments on chicken microbiota using high-throughput sequencing of 16S rRNA gene amplicons.
Cecal contents were obtained at 21 and 40 days of age from chickens raised under experimental and commercial conditions. DNA was extracted from each pooled cecal sample and the V3-V4 region of the 16S rRNA gene was amplified. High-throughput sequencing was performed in the Illumina MiSeq platform. Bioinformatics and statistical analyses were done with QIIME2 and STAMP software.
A significantly different microbial profile was found between experimental and commercial samples and also between the two sampling ages analyzed. Significant differences in the diversity metrics across dietary treatments were also detected in both environments. Tannins increased *Firmicutes* to *Bacteroidetes* ratio, a parameter linked to feed conversion efficiency, in both experimental and productive conditions. This was mainly due to the growth of members of order *Clostridiales* and genus *Lactobacillus* and the decrease of genus *Bacteroides*. Tannins also increased the relative abundance of other potentially probiotic genera as *Bifidobacterium* and *Enterococcus*. The relationship between the intestinal microbiota profile obtained and chicken productivity remains unclear, although certain members of order *Clostridiales* and phylum *Proteobacteria* showed different degrees of correlation with productive parameters. Tannins and antibiotics have a different impact on cecal microbiota, affecting specific bacterial groups. This study glances at the model to go in deep using metagenomics analysis to evaluate the impact of feed additives on the intestinal microbiome of poultry.

Keywords: Feed additives, Growth promoters, High-throughput sequencing, Intestinal microbiota, Tannins

Effectiveness of a phytogenic feed additive on growth performance of broiler chickens raised under optimal health conditions

Abstract ID: 331

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A healthy digestive system is a key factor for high growth performance and feed efficiency in poultry production. Phytogenic feed additives with antimicrobial and health promoting effects seem to have a beneficial influence especially under challenging health conditions. However, in modern and highly sophisticated livestock farming, the animal health is usually at a higher level. Thus, the question arises whether a phytogenic additive also improves the performance under optimal conditions. This was tested within an official project at a trial facility in Iran. The objective of this study was to determine the effectiveness of the feed additive Anta®Phyt (dosage: 200 g/t, prior to pelleting) on growth parameters and mortality. This phytogenic product is composed of plant extracts including selected components of the hop plant, on a natural carrier. In total 25,000 broiler chicks (Cobb 500) were randomly assigned to two dietary treatments (with and without Anta®Phyt) with two replications. Birds were fed experimental diets based on corn and soybean meal in a three-phase feeding program from day 1 to 42 of age. Feed and water were provided ad libitum throughout the trial. To evaluate the growth performance, body weight was measured weekly. At day 42, the treatment group showed improved (+45 g) growth performance in terms of live weight gain (1820 ± 200 vs. 1745 ± 150 g, P=0,08) and feed conversion ratio (1,61 ± 0,20 vs. 1,66 ± 0,10 g/g, P=0,13) in comparison to the negative control. In both groups, the broilers showed a good health status and cumulative mortality was low (2%). However, the treatment with the phytogenic additive led to an even reduced mortality in the last two weeks (0,5% vs. 0,7%), so that a lower total live weight loss was achieved.These results indicate that supplementation of the phytogenic additive Anta®Phyt to the broiler diet has beneficial effects on growth and efficiency, even in herds with good health status.

Keywords: Broiler, Feed additives, Phytogenic

Effect of a mycotoxin mitigating agent on the oral absorption of deoxynivalenol and ochratoxin a in broilers

Abstract ID: 137

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Limited information is available in literature concerning the effect of mycotoxin binders on the in vivo absorption of mycotoxins. The European Food Safety Authority (EFSA) stipulates that, in addition to preliminary in vitro trials, in vivo testing of mycotoxin binders is necessary to evaluate the efficacy of the binder. Though, in vivo studies where non-specific parameters (e.g. growth, feed conversion, et al.) are measured, do not suffice to demonstrate the efficacy of these products. In vivo toxicokinetic studies, based on the absorption, distribution, metabolisation and excretion of the mycotoxin, are mandatory to evaluate the possible effects of mycotoxin binders on the oral absorption of mycotoxins in animals. Consequently, a study was performed to determine the efficacy of a SAN-protect® on the oral absorption of the mycotoxins deoxynivalenol (DON) and ochratoxin A (OTA) in broiler chickens. Sixteen healthy 4-weeks-old broilers were randomly allocated in 2 groups of 8 animals. After 1 week of acclimatization, the broilers were administered OTA (0.25 mg/kg) and DON (0.5 mg/kg) without mycotoxin binder (group 1) or in combination with SAN-protect® (group 2). Blood samples were taken at different time points pre- and post-administration and OTA and DON were quantified in plasma with a validated LC-MS/MS method. The effect of the mycotoxin binder on the oral absorption of DON and OTA was evaluated by statistical comparison of toxicokinetic parameters between the mycotoxin and mycotoxin + binder treated broilers, with special emphasis on $AUC_{0-\infty}$, Cmax and Tmax and relative oral bioavailability (F). Administration of SAN-protect® resulted in a lower systemic exposure ($AUC_{0-\infty}$) to both DON (P=0.130) and OTA (P= 0.003) in broilers. After SAN-protect® supplementation, a relative oral bioavailability of 47.5% and 67.4% was observed for DON and OTA respectively, compared to the control group (group 1).

Keywords: Deoxynivalenol, Ochratoxin A, SAN-protect®

Effect of betaine hydrochloride on intestinal health of broilers

Abstract ID: 554

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Betaine as osmoregulator increases water retention capacity of the gut and therefore is known to support intestinal health. When intestinal health of broilers is impaired often wet litter can cause footpad lesions. These footpad health problems cause impaired performance and have a negative impact on economics of broiler production and on animal welfare. Whereas European diets have been free of antibiotics for several years, the implementation of antibiotic free diets is currently of high interest in other parts of the world. Litter quality and footpad health are of high concern when in-feed antibiotics are being withdrawn from the diet. To evaluate the effect of betaine on footpad health and broiler performance, a performance trial was conducted comparing a control diet with a similar diet with 0.1% betaine hydrochloride* addition. These diets were fed to 870 broilers (Ross 308), housed in pens with 30 birds each. The trial lasted from day 1 to 42 of age and performance parameters, litter quality and footpad scores were measured. Litter quality was measured by scores from 1 (dry compact litter) to 3.5 (wet sticky litter; with 0.5 increments). Footpad lesions were scored from 0 (no lesions) to 4 (severe lesions; with 1 increment). There were no significant differences in end body weight and feed intake between the control group and the betaine group. Feed conversion ratio of the betaine group was numerically improved compared to control (1.586 vs. 1.607). Litter quality was significantly improved in the betaine group over the course of the trial with on average a score of 2 in the betaine group vs. 2.2 in the control group. This improved litter score resulted in improved footpad health. At day 28 there were significantly more birds with lower footpad lesion scores in the betaine group vs. the control group (score 0 and 1) and significantly more higher scores in the control group vs. the betaine group (score 2 and 3). On day 42 this same trend was shown, with numerical differences. The positive effect of betaine on litter quality and footpad health shown in this trial suggest that betaine hydrochloride can improve intestinal health in broilers.
* Excential Beta-Key (Orffa)

Keywords: Betaine hydrochloride, Broiler performance, Footpad health, Intestinal health

Effect of crude protein levels with supplement of limiting amino acids on broiler performance

Abstract ID: 598

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Content of crude protein in the diets has a significant effect on broilers growth and carcass quality. On the other hand it is the most expensive ingredient in complete feed diets. The main source of crude protein in diets is soybean meal but EU including Czech Republic is depending on import of this feed and it belongs to the most expensive feed component. That is why feeding of diets with reduced crude protein levels with supplement of limiting amino acids could improve economical and environmental aspects and welfare of broilers. The aim of the study was to evaluate the effect of four crude protein (CP) levels supplemented with limiting amino acids in the broiler diets on broilers growth, feed conversion ratio (FCR) and carcass quality. Hybrid ROSS 308, 2640 chickens, were divided into four groups and they fed grower diets with 22; 20.8; 19.5 and 18% of crude protein. The experiment period lasted from 10 to 30 days of age. Starter and finisher diets were the same for all groups. Grower diets were optimized in digestible limiting amino acids (lysine, methionine, leucine, isoleucine, threonine, tryptophan a valine). The lowest growth intensity was observed in the broilers fed 18% CP ($P<0.05$) on the other hand the highest weight was found in the group fed 19.5% CP ($P<0.05$). The best FCR was found at feeding 19.5% CP and the worst at feeding 18% CP ($P<0.05$). The carcass yield and breast yield were the highest in group fed 19.5% CP ($P<0.05$). The highest proportion of abdominal fat was found in group fed 18% CP ($P<0.05$). On the basis of the results we can recommend the level 19.5% crude protein in grower diets supplemented by limiting amino acids without negative effect on FCR and broiler growth.

Keywords: Broiler, Carcass quality, Crude protein level, FCR, Growth

Effect of dietary inclusion of hemp (Cannabis Sativa L.) And dill seed (Anethum Graveolens) vis-a-vis antibiotic growth promoter on microbial quality of intestinal contents in broiler chickens

Abstract ID: 523

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An experiment was conducted with dietary inclusion of hemp *(Cannabis sativa* L.*)* and dill *(Anethum graveolens)* seeds in varying combinations against antibiotic growth promoter in broiler diet. The objective was to study the possible synergistic effect of hemp seed (HS) and dill seed (DS) on microbial quality of intestinal contents in broiler chickens as against antibiotic growth promoter and control diet. In this experiment 352 CARIBRO-VISHAL broiler chickens were grouped into 11 dietary treatments for 42 days comprising hemp and dill seed in various combinations in addition to basal diet. Each treatment had 4 replicates with 8 birds per replicate, *viz.*T1 (control-basal diet) T2 (0.1%HS), T3 (0.1%HS + 0.1%DS) T4 (0.1%HS+0.3%DS), T5 (0.2%HS), T6 (0.1%HS+0.1%DS), T7 (0.1%HS+0.3%DS), T8 (0.3%HS), T9 (0.1%HS+0.1%DS), T10 (0.1%HS+0.3%DS) and T11 (basal diet+0.025% BMD). At 42 days post-hatch, 6 healthy birds from each treatment group were sacrificed to collect cecal and jejunal contents. Total coliform and lactobacillus count was enumerated by the method described by APHA (2001) by using MacConkey and MRS agar as nutrient media respectively. The colony count was expressed in terms of Log_{10} cfu/g of intestinal contents. Significant reduction ($P<0.001$) of total coliform count both in cecum and jejunum was observed in treatment birds with concomitant increase in seed level. Also, almost all treatment birds showed significantly higher ($P<0.001$) lactobacillus count in both cecum and jejunum than control (T1) birds, hence showing direct relationship with inclusion level of both seeds. It is concluded here that, the seed combination at any level could help in reducing the coliform and increasing the lactobacillus count in broiler chickens.

Keywords: Broiler, Coliform, Dill, Hemp, Lactobacillus

Effect of dietary supplementation of peppermint on performance, meat physicochemical properties and carcass characteristics of broiler chicks under hot climatic conditions

Abstract ID: 335

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Improved animal performance and reduced heat stress can be accomplished by herbal plants as feed supplements. The present study was conducted to investigate the effect of dietary peppermint leaves supplementation on performance, meat physicochemical properties, carcass characteristics and internal organs in broiler chicks under hot climatic conditions of Upper Egypt. A total of 192 one-day-old Ross 308 broiler chicks were assigned to 4 dietary treatments that included peppermint leaves at concentrations of 0, 5, 10, or 15 g/kg for 35 days. Each treatment had 6 replicate pens with 8 birds. In addition to performance variables, 12 randomly-selected birds from each treatment were slaughtered at the last trial day to evaluate carcass traits and meat quality. The results showed that body weight and body weight gain were increased (linear, $P<0.01$) with the increase in dietary peppermint leaves concentration during starter, grower and entire trial periods. In addition, the feed intake linearly increased ($P<0.01$) with increasing peppermint levels in the grower and the entire trial periods and, in turn, caused linear improvements ($P<0.01$) in feed conversion values during the grower and entire trial periods. Although pH values and drop loss percentage of breast and leg muscles were not affected by dietary peppermint levels, increasing peppermint levels decreased ($P<0.001$) cook loss percentage of breast and leg muscles. On the other hand, dietary supplementation of peppermint leaves had no effect ($P\geq0.05$) on the relative weights of dressing, breast, leg, liver, heart, gizzard, spleen and pancreas. Interestingly, abdominal fat percentage was decreased by supplemental peppermint levels. The present findings indicate that peppermint leaves can be used as an effective feed additive up to 15 g/kg to improve performance of broiler chicks in hot climatic regions.

Keywords: Broilers, Carcass criteria, Growth performance, Meat quality, Peppermint

Effect of different dietary levels of alfalfa (Medicago sativa) meal on the serum and yolk cholesterol concentration of laying hens

Abstract ID: 295

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Even though the egg is considered the "perfect food of nature," especially because of the high protein content of high biological value and the high digestibility of the nutrients, fear of cholesterol is a barrier to the evolution of the average individual consumption. Among the nutritional ways used to lower the cholesterol in eggs, is feeding hens with high fiber diets. A 4-week feeding trial was conducted on 90, Tetra SL layers (age 42 weeks) assigned to 3 groups (C, E1 and E2) to investigate the effect of dietary alfalfa meal *(Medicago sativa)* on the serum and yolk cholesterol concentration. Birds were housed in cages (4 hens/cage) on three tiers in an experimental hall with controlled microclimate (16h light, temperature 20.01 ± 1.44^{0}C, humidity $65\pm6.38\%$). The control group (C) received a conventional corn-soybean meal diet with 2800 kcal/kg of metabolizable energy and 18.38% crude protein. Unlike diet C, the diets of the experimental groups also included 11.95% (E1) and 16.34% (E2) alfalfa meal, which increased the fibre to 6.86% (E1) and 8.23% (E2), compared to 3.71% in group C diet. On the last experimental day, blood samples were collected from 6 hens/group in order to determine the serum energy profile (blood glucose, cholesterol and triglyceride). In the last week of the experiment, 18 eggs/group were randomly selected, and used to form 6 average yolk samples (3 eggs/sample) assayed for cholesterol concentration. The results show that the serum cholesterol in the hens from experimental groups significantly decreased ($P\leq0.01$), (95.85 mg/dL in E2 respectively 108.58 mg/dLin E1), compared to group C (144.16 mg / dL). The level of cholesterol in eggs decreased significantly with the increased level of dietary fibre. The cholesterol concentration in the eggs collected from the E2 group (8.23% fibre) was 236.27 mg cholesterol/egg, significantly lower ($P\leq0.01$) than in group C (289.28 mg cholesterol/egg) and E1 group (263.98 mg cholesterol/egg). It can be concluded that the use of 16.34% alfalfa meal *(Medicago sativa)* in layer diets, had beneficial effects both on the serum cholesterol concentration and on cholesterol concentration in the egg.

Keywords: Alfalfa meal, Blood serum, Egg cholesterol, Fiber, Laying hens

Effect of different exogenous enzymes on the early life performance of broilers

Abstract ID: 432

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Feed enzymes are commonly added to wheat based diets to counter the possible anti-nutritional effects caused by non-starch polysaccharides (NSP). The enzyme most frequently added to wheat-based diets is xylanase, because arabinoxylan is the main NSP present in wheat. In addition to arabinoylan, however, wheat also contains significant amounts of β-glucan and cellulose. Hence, other enzymes like β-glucanases and cellulases may also play an important role in wheat-based feeds. In the current broiler trial, a comparison was made between four commercially available xylanases and an enzyme mixture (xylanase, cellulase, β-glucanase, peptidase and α-amylase).
In total 1440 male broiler chickens (Ross 308) were fed a diet with a high wheat content (56% wheat) with or without the addition of an enzyme preparation. Four commercially available xylanase preparations and an enzyme mixture were used in this trial. All enzyme preparations were added based on an equal xylanase activity (Xylazyme-AX kit, Megazyme) and the dosages were within the commercial application dosages given by the suppliers. One-day-old broilers were assigned randomly to 48 pens (6 dietary treatments x 8 replicates) and broiler performance during the early rearing period of the broilers (d1-13) was recorded.
The use of the enzyme mixture resulted in the highest improvement of feed conversion ratio compared to the control diet (1.293 compared to 1.360 in the control diet; $P=0.006$). Moreover, it significantly improved the body weight of the broilers compared to the control and to the other treatments ($P<0.001$). In the early life of the chicken, its digestive capacity is not yet fully developed. Hence, in this period, exogenous enzymes such as amylase and peptidase could increase the digestive capacity of the chicken. *In vitro* research showed that adding a mixture of other polysaccharidases and peptidase to xylanase increased the degradation of the water-unextractable-part of NSP (Smeets *et al.*, 2014; 14th EPC, Norway), putatively by reducing the cage-effect caused by the plant cell walls. Different component enzymes can attack different portions of the cell wall at once and thereby increase the possibility of xylanase to degrade the NSP.

Keywords: Broiler performance, Exogenous enzymes, Wheat, Xylanase

Effect of different levels of Macleaya cordata alkaloid extract in low protein diets on performance, Ileal protein digestibility and plasma amino acid concentration in broiler chickens

Abstract ID: 610

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This experiment was conducted to evaluate the effect of different levels of *Macleaya cordata* alkaloid extract (Sangrovit X10®) (0, 180, 360, 540 mg/kg) in low protein diets (100 and 95% of CP requirement for Ross 308) on performance, Ileal protein digestibility and plasma amino acid concentrations of broiler chickens. Accordingly, 560 male broiler chicks (Ross 308) were assigned to 8 dietary treatments based on a factorial arrangement (2×4) in a completely randomized design. Five replicates of 14 birds allocated to each treatment. Birds fed low protein diet supplemented with 540 mg/kg Sangrovit X10® had higher body weight with same FCR at 35 days of age compared to control groups fed low or normal protein diets ($p<0.05$). In addition, supplementation of 540 mg/kg Sangrovit X10® improved ileal protein and organic matter digestibility and reduced plasma uric acid concentration than control group at 35 d of age. Plasma lysine concentration significantly was lower in birds fed low protein diets ($P<0.05$) and Sangrovit X10® supplementation could not ameliorate this trend. Totally, it seems that dietary supplementation of Macleaya cordata alkaloid extract would be able to improve performance via protein digestibility and amino acid availability in broiler chicks fed low or normal protein diets.

Keywords: Amino acid availability, Broiler chicks, Performance, Protein digestibility, Sangrovit

Effect of digestible Trp to Lys ratio and low dietary crude protein diets on laying performance and egg quality in 36 to 50 weeks old laying hens

Abstract ID: 467

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Lowering dietary crude protein (CP) in poultry diets allows to reduce feed costs and environmental impact and to improve the health and welfare of birds. However, in order to implement successful low CP diets, dietary amino acid (AA) levels need to be adequately provided. In a typical corn-soybean meal based diet, Trp is the third limiting AA after Met and Lys. In the present experiment, two levels of digestible Trp to Lys ratio in a low CP diet were compared to a regular CP diet. A total of 960 commercial layers were placed into 3 treatments with 16 replicates of 20 hens from 36 to 50 weeks of age. All diets were iso ME (2720kcal/kg) and iso digestible Lys (0.69%).The regular CP diet (T1) was formulated to have 17.0% CP, while the low CP diet was 15.6% CP. The digestible Trp to Lys ratio was 24% in the regular CP diet, and either 19% (T2) or 24% (T3) in the low CP diet, obtained by L-Trp supplementation. Laying rate, weekly egg number, egg weight, weekly egg mass, daily feed consumption, feed conversion ratio and individual body weight were evaluated. Egg weight, static stiffness, shell proportion, shell index, shell thickness and fracture force were measured in around 200 eggs per treatments. Significant effects were observed for egg weight, weekly egg mass, shell proportion, shell index and shell thickness. Reducing dietary CP resulted in lower egg weight and egg mass but egg mass of laying hens fed the low CP diet with L-Trp was not significantly different from the high CP diet, indicating that adequately supplying dTrp:Lys allows to maintain performance in low CP diets. For egg quality traits, hens fed the low CP diet with 24% dTrp:Lys showed significantly higher shell thickness and shell index than low CP diet with 19% dTrp:Lys. In addition, shell proportion was significantly higher when hens were fed the low CP diet with 24% dTrp:Lys compared to the two other treatments. In conclusion, maintaining dietary dTrp:Lys at 24% allows to maintain laying performance of hens fed low CP diets while enhancing egg quality.

Keywords: Eggshell quality, Low Crude Protein Diets, Tryptophan

Effect of dried tomato pomace on the performance, egg traits, blood parameters and immune response of laying hens

Abstract ID: 16

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This study was conducted to evaluate the effect of different levels of dried tomato pomace on the performance, egg quality traits, blood parameters and immune response of laying hens. One hundred fifty, Hy-line W36 hens at 73-81 weeks old, were randomly assigned to 6 treatments of 25 each. Different levels of dried tomato pomace (0, 2, 4, 6, 8, and 10%) were added to the diets of these six groups. Percent of egg production, feed conversion ratio (FCR), feed intake, percent of breaking eggs, egg shell thickness, haugh unit, shell strength, egg mass, egg weight and yolk color were recorded. The FCR, egg production, breaking egg %, shell thickness, haugh unit, egg shell weight, egg mass, egg weight were not significantly affected by the addition of dried tomato pomace. However, it did have an effect on feed intake, shell strength and yolk color. Blood parameters such as cholesterol, HDL, and LDL were also not affected. The addition of 6% pomace had an effect on the immune response; lymphocyte/ heterophile ratio was affected. It was concluded that using up to 10% dried tomato pomace in the diets of laying hens had no adverse effect on the performance and egg traits of birds but did improve their immune response.

Keywords: Blood parameters, Immune response, Laying hen, Performance, Tomato pomace

Effect of feeding high quality crude glycerol as energy source in broiler diets

Abstract ID: 423

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Thailand has promoted the alternative energy, biodiesel, to replace using fossil fuel. The raw materials for biodiesel production are mainly palm oil and palm wax. By using proprietary process, approximately 10% of high quality crude glycerol (low methanol) was produced as by-product of biodiesel production. The aim of the study was to investigate the use of high glycerol as alternative energy source for broiler. High quality crude glycerol contains 84.6% glycerol, < 0.01% methanol, 5.7 %wt NaCl and 6,340 kcal GE/kg. Our digestibility study indicated that high quality crude glycerol contained 3,697 kcal ME/kg. Feed study was conducted to determine the effect of using high quality crude glycerol in broiler wdiets. A total of 2000, 1-day old, Ross 308 were divided into 4 treatments including, corn-soy basal diet (CS), CS with 2.5, 5 and 7,5% crude glycerol, respectively. Each treatment consisted of 10 replications with 50 birds each (25 male and 25 female). All diets were isocaloric and isonitrogenous. Birds were raised in an evaporative cooling system house with feed and water provided *Ab libitum* for 35 days period. There was no significant difference in feed intake, weight gain, FCR and mortality. The results revealed that high quality crude glycerol can be used in broiler diet at 7.5% without a negative effect on growth performance.

Keywords: Alternative energy, Broiler, High quality crude glycerol, Metabolizable energy

Effect of feeding yeast fraction alone or in combination with organic selenium on oxidative status and meat quality of broilers under heat stress

Abstract ID: 360

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The study was conducted to evaluate the effect of feeding yeast fraction alone or in combination with organic Se (Safmannan® and Selsaf®, Phileo Lesaffre Animal Care, France) on growth performance, oxidative status and meat quality of broilers under heat stress. A total of 2000 day-old Ross 308 broilers were randomly assigned into 4 treatments using Completely Randomized Design. Each treatment consisted of 10 replicates with 50 birds each (25 males and 25 females). All chicks were housed in an environmentally controlled house. The dietary treatments were no feed additive (T1), adding 250 g/ton yeast fraction throughout the experiment (T2), adding 250, 500 and 250 g/ton yeast fraction in starter, grower and finisher diets (T3) and T2 with 100 g/ton of organic Se in grower and finisher diets (T4). All diets were formulated according to Ross 308 recommendation. The trial was run for 35 days during heat stress (June-July/hot summer and rainy season of Thailand). Feed in pellet form and water were provided *Ad-libitum* throughout the experiment. There was no significant difference in feed intake, body weight gain, feed conversion ratio and mortality among dietary treatments. Nevertheless, feeding yeast fraction tended to lower mortality during 22-35 day of age (DOA). Feeding yeast fraction and organic Se had no effect on carcass trait and pH of breast meat ($P>0.05$). However, drip loss at 24 hours tended to lower when feeding broiler with diet containing yeast fraction 500 g/ton during grower (T3) and yeast fraction in combination with organic Se during grower and finisher period (T4; $P=0.0954$). Malondialdehyde concentration in the serum was decreased by supplementation of yeast fraction and yeast fraction with organic Se in diets (T4) during 21 DOA ($P=0.1771$) and 35 DOA ($P=0.1217$). The results from the current study indicated beneficial effect of feeding yeast fraction and organic Se in improving oxidative status and meat quality of broilers during heat stress.

Keywords: Broiler, Heat stress, Organic Se, Oxidative status, Yeast fraction

Effect of fiber source and content on volume and physical properties of the final diet

Abstract ID: 31

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The crude fiber fraction in poultry diets contains mainly insoluble polysaccharides like cellulose and lignin, but also non-starch polysaccharides (NSP). Insoluble fibers are believed to have beneficial effects on the digestion system of the birds, by this improving wellbeing, health level and performance. In laying hens, it was observed that increased dietary fiber contents can reduce the risk of feather pecking and cannibalism. Therefore, it is recommended to use feed with increased fiber contents in feeding layer pullets and laying hens. As there is limited information on the effects of dietary fiber sources and contents on volume and physical properties of the final compound feed a laboratory study was conducted. Alfalfa meal, cellulose (Arbocel®), spelt dust and wheat straw were used as fiber sources and were included in a layer diet, based on soybean meal, wheat, corn and corn starch, in way that a crude fiber content of 4, 5 and 6 % was reached, respectively (N=4x3=12+3 control=39 diets). A diet without the addition of any fiber source was included as a control. With the mixer M20GP of the manufacturer Lödige (Germany) 5 kg of each experimental diet was produced. In a sample of 200 g of the final mixture the volume, the distribution of particle sizes was determined (N=78). Water binding capacity was measured in the native fiber source. Furthermore, the crude fiber content of the diets were determined according to Henneberg and Stohmann (1860). The highest feed volume was determined for cellulose (403 mL), followed by wheat straw (345 mL), spelt dust (320 mL) and alfalfa meal (280 ml). Without supplementation of a crude fiber source the volume was 275 mL. The volume amounted to 400, 370 and 337 ml for a fiber content of 4, 5 or 6 %, respectively. Wheat straw and spelt dust contained mainly particles > 3mm, whereas, in alfalfa meal particles < 2mm were dominating. Water binding capacity of fiber sources were 2.96, 4.36, 5.90 and 9.04 g water/g fiber source for spelt dust, alfalfa meal, wheat straw and cellulose, respectively. The analyzed fiber contents of test diets have been in the expected range. The results underline that fiber sources and contents in diets affect the volume and the water binding capacity of the feed. This may affect the nutrient intake by the bird.

Keywords: Crude fiber, Feed, Feed volume, Fiber content, Fiber source

Effect of high dietary levels of α-tocopherol acetate on immune response of light and heavy weight male broiler breeders

Abstract ID: 58

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Effect of overdosing dietary α-tocopherol acetate (α-TOH) on immune response of heavy weight roosters investigated in a precision study. A total of 60 roosters (Ross 308) in a light (LW, n=30) and heavy weight (HW, n=30) were kept in the individual cages. Roosters were randomly divided into 10 treatments in a 2×5 factorial experiment. Each group received a basal diet supplemented with graded levels of α-TOH (0, 100, 200, 300, 400 mg/kg diet) for 10 weeks (from 25 to 35 week of age). Blood samples were collected at fifth and tenth week of the experiment. Afterward, humoral immune system evaluated by sheep red blood cell (SRBC) haemagglutination assay (HA) and haemagglutination inhibition (HI) tests including antibody response to Newcastle disease viruses (NDV) and avian influenza viruses (AIV). Cell-mediated immune response evaluated by cutaneous basophil hypersensitivity (CBH) test. Positive linear relationship was observed between incremental levels of α-TOH and both humoral and cell mediated immunity ($P \le 0.05$). So that, all immune parameters amplified by diet supplemented with 400 mg/kg α-TOH ($P \le 0.05$). Results showed that CBH in LW roosters was significantly higher than HW roosters ($P \le 0.05$). However, the interaction of BW and α-TOH doses didn't have any significant effect on immune responses ($P \ge 0.05$). In conclusion overdosing diet with α-TOH four times more than strain nutrients recommendation had beneficial effects on immune response of both heavy and light body weight male broiler breeders.

Keywords: Avian influenza, Broiler breeder males, Immune response, Newcastle disease, α-tocopherol

Effect of high fibre layer diets on layer performance and egg quality

Abstract ID: 349

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The effect of high fibre layer diets was tested in an 8-week feeding trial on 120, Tetra SL layers (28 weeks), assigned to 5 groups (C, E1, E2, E2 and E4), using alfalfa (*Medicago sativa*) (15.93% crude protein and 30.58% crude fibre) as fibre source. The layers (4 birds/cage) were housed in a three tier ZUCAMMI battery located in an experimental hall with controlled environment (16 light regimen, 20.01±1,44^{0}C temperature; 65±6.38% humidity). Compared to the conventional diet of the control group (2760 kcal/kg metabolizable energy; 17.7% crude protein and 3.71% crude fibre), the diets for the experimental groups included pelleted alfalfa: 11.95% (E1, E2) and 16.34% (E3, E4). The dietary alfalfa increased the dietary fibre level up to 6% (E1, E2) and to 7% (E3, E4). In order to improve fibre digestibility, the formulations for groups E2 (6% crude fibre) and E4 (7% crude fibre) were supplemented with an enzymatic product (0.15%). Layer performance was monitored throughout the experimental period. The average daily feed intake increased significantly (P≤0.05) in group E4 (120.96 g/layer/day) compared to the control group C (111.26 g/ layer/day). The laying percentage was better in group C (99.08%) compared to the experimental groups E1 (93.7%); E2 (95.92%); E3 (95.03%) and E4 (94.64%). In the end of the feeding trial (week 8), we collected randomly 18 eggs/group. After the external and internal quality parameters of the eggs were measured, we formed 6 yolk samples/group (3 eggs/sample) and assayed them for polyunsaturated fatty acids (PUFA) concentrations. The use of dietary alfalfa in the diets for the experimental groups increased significantly (P≤0.05) yolk colour intensity compared to the control. A significant (P≤0.05) increase of the Haugh units was noticed in group E3 (84.47) compared to group C (78.59); however, the differences were not statistically significant for the other groups. Significant (P≤0.05) increases of PUFA concentration in the yolk were noticed in the experimental groups: 31.305 g/100g total fatty acids (E1); 31.45 g/100g total fatty acids (E2); 33.75 g/100g total fatty acids (E3) and 34.872 g/100g total fatty acids (E4), compared to the control group C, 23.97g/100g total fatty acids. The pelleted alfalfa had a positive influence on the quality parameters of the eggs, without affecting the production parameters.

Keywords: Alfalfa, Egg quality, Eggs, Fibre, Polyunsaturated fatty acids

Effect of nonanoic acid supplementation on broiler performance

Abstract ID: 341

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Medium-chain fatty acids have been shown *in vitro* to have antimicrobial activity against *Escherichia coli*, *Salmonella typhymurium*, *Campylobacter jejuni*, and *Clostridium perfringens*. Among them, caproic, caprylic, capric and lauric acids are the most studied. Nonanoic acid (C9), a medium-chain fatty acid used as a flavouring agent in animal feed and naturally present in numerous plants, has also demonstrated antimicrobial and antifungal effects. This experiment was conducted to evaluate the effect of nonanoic acid supplementation at different doses in broilers fed corn/soybean meal based diets from 1-42d. A total of 1,100 Ross 308 one-day-old broilers, half male and half female, were placed in 50 floor-pens (22 broilers/pen). Five treatments were applied based on the inclusion of nonanoic acid in the diets at 0, 100, 300, 500 and 1,000 mg/kg. Observations of feed intake, growth, BW, and feed efficiency were recorded at 7, 21 and 42d. Blood haematology and biochemistry, necropsy and histopathology were analysed at 42d in one bird/replicate. Data were analysed as a randomized complete design with nonanoic acid supplementation as main effect. Additionally, the dose-response was analysed, testing for L&Q effects. All blood parameters were within the normal range for the weight and age of the birds studied and no significant differences between treatments were observed in any of the haematology parameters analysed. No significant differences between treatments were observed in any of the histopathology parameters evaluated. Nonanoic acid supplementation increased feed intake up to 500 mg/kg (L:P=0.0698; Q:P=0.0092). Broilers receiving 500 mg/kg nonanoic acid at 7d or 1,000 mg/kg at 21d tended to be heavier than controls. At 42d, treatment effect was not significant, but nonanoic supplementation increased BW linearly (P=0.0263). From 7-21d and for 0-21d, growth was increased and feed conversion was reduced by nonanoic acid supplementation (L&Q, P<0.05). For the global study (0-42d), nonanoic acid supplementation at 1,000 mg/kg tended to improve EPEF over the control (+7.0%, 316 vs 338; P≤0.10). There was also L&Q relationship between the nonanoic acid supplementation and improved growth, FCR and EPEF for the whole study. It is concluded that, at dosages up to 1,000 mg/kg, nonanoic acid was well tolerated by broilers. A trend for improved growth, FCR, and EPEF was observed with nonanoic supplementation. Further work is needed to determine if these improvements in performance were due to nonanoic acid improving palatability of the feed and thus increasing feed intake or because of an antimicrobial effect in the feed.

Keywords: Broiler, Medium-chain fatty acids, Nonanoic acid, Performance

Effect of partial substitution of D3 by 25-OH-D3 on production parameters and gait scoring in commercial broiler farms

Abstract ID: 547

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Partial substitution of D3 by 25-OH-D3 (25D3) may improve aspects of performance and animal welfare in broiler farms. Two identical broiler houses within the same farm, each housing approximately 18000 male Ross 308 birds vaccinated against coccidiosis, were offered diets with vitamin D in the form of D3 (5000 in the starter and 4000 over the grower period IU/kg) or the equivalent IU/kg following partial substitution (1:1) with 25D3. Diets were alternated between houses over a total of 4 production cycles. Diets were offered according to the commercial practices, maintaining identical nutrient specifications between cycles. Performance variables such as final BW, FCR, percentage mortality and percentage of culled birds were assessed at the end of each cycle. In addition, approximately 150 birds were gait scored (GS) within the last 3 days prior to the end of each production cycle, which lasted between 42 and 48 days of age. Furthermore, 150 birds from each house were assessed in the slaughter line for the incidence of foot pad dermatitis (FPD) and percentage of carcass downgrades. Overall, dietary supplementation with D3 or 25D3 did not bear an effect on performance, incidence of FPD, percentage of carcass downgrades, percentage mortality or percentage of culled birds. Although average GS did not differ significantly ($P > 0.05$) between dietary treatments, partial substitution with 25D3 significantly increased ($P < 0.05$) the percentage of birds rated with a GS of 1, tended to increase ($P < 0.1$) the percentage of birds with a GS of 0 + 1, tended to decrease ($P < 0.1$) the percentage of birds with a GS of 2+3, whilst the proportion of birds with GS of 4 + 5 was not affected. In conclusion, although offering 25D3 did not improve aspects of performance it may mediate improvements in the walking capacity of intensively raised broilers.

Keywords: 25-OH-D3, Commercial broiler production, Gait score, Performance, Vitamin D

Effect of prebiotic on intestinal microflora, carcass and meat chemical composition of broiler chickens

Abstract ID: 46

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Effect of the prebiotic (MOS) on the broiler chicken's intestinal microflora, carcass and meat chemical composition was investigated. The broilers in both groups were fed at libitum with the dry not granulated standard wheat – corn meal compound feed of the same composition and the same nutritional value, except the addition of the prebiotic (MOS) for group 2. This prebiotic was included in the feed premixes from the first day of life to 38 days of age. The results showed that the components of the prebiotic helped to maintain the microflora balance of the intestinal tract of poultry, had a positive impact on the carcass and meat chemical composition. The prebiotic (MOS) help to maintain the microflora balance of the intestinal tract of broiler chicken's, had a positive impact on the carcass and meat chemical composition. The prebiotic had a positive effect on the population of "beneficial" bacteria in the intestinal tract of the broilers from the trial group. For male and female broilers the biggest increase was determined on carcass yield and muscle weight ($P < 0.05$). The results of chemical analysis of the broiler chickens' meat at the age of 38 days showed that this prebiotic had positive influence on some meat chemical composition parameters. Under the impact of the prebiotic (MOS) there were more dry matter, protein and fat in the meat of male and female broilers meat.

Keywords: Broiler chickens, Carcass composition, Intestinal microflora, Meat chemical composition, Prebiotic

Effect of probiotic supplementation on performance and egg quality of laying hen during the late egg cycle

Abstract ID: 410

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This study was conducted to evaluate the effect of probiotic (Saltose) on performance and egg quality of laying hen during the late egg cycle. A total of 360 laying hens (Lohmann Brown) at 64 weeks of age were assigned to 3 dietary treatments, with 8 replicate cages and 15 birds per cage. The dietary treatments consisted of a control diet (corn-soybean meal basal diet), and a control diet supplemented with 250 and 500 ppm probiotic, respectively. The diets in mash form were offered for 8 weeks during late of egg cycle production (64 to 72 weeks of age). The different levels of probiotic supplementation had no significant (P>0.05) effect on body weight change, feed intake, feed conversion ratio, hen day egg production, hen-housed egg production, egg weight, egg mass, and percentage of livability. There were no significant (P>0.05) differences in egg shell breaking strength, albumen height, haugh unit, yolk color, and egg component among birds fed the diets containing the different probiotic levels. However, egg shell thickness was significantly (P<0.01) increased with an increasing in probiotic levels in layer diet. The highest shell thickness was observed for layer fed a diet containing 500 ppm probiotic. Under the condition of present study, there were concluded that feeding probiotic (Saltose) had no effect on laying hen performance. The egg shell thickness of laying hen could be improved with probiotic supplementation.

Keywords: Egg production, Egg quality, Laying hen, Performance, Probiotic

Effect of soapnut shell powder supplementation on reproductive performance of broiler breeders

Abstract ID: 500

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Present experiment was conducted to investigate the effect of soapnut shell powder (SSP) on serum hormone profile, fertility, hatchability and total embryonic mortality of broiler breeders. 120 broiler breeders (96 females and 24 males) of about 38 weeks age were randomly distributed equally to four different treatments consisting of three replicates of 8 female and 2 male birds. All the birds were fed on a basal diet (ICAR, 2013) for 42 days. T1 (control), T2, T3 and T4 were supplemented with 0, 0.0176%, 0.026% and 0.0528% SSP (dose equivalent: 0, 50, 75 and 150ppm saponin), respectively (saponin yield from soapnut shells were 28.4% on DMB). The serum testosterone (ng/ml) was significantly higher (P<0.001) for T4 (5.64) and T3 (4.56) treatment groups compared to T1 (3.63) and T2 (4.38) treatment groups. The seminal plasma testosterone (ng/ml) level among the treatments was significantly higher (P<0.05) for T4 (1.65) treatment group compared to T1 (1.41), T2 (1.47) and T3 (1.55) treatment groups. The serum estrogen level in females were insignificant (P>0.05) among the treatments. The fertility (%) was significantly higher (P<0.01) in SSP supplemented groups (T3-90.23%) compared to control (T1-84.23%). Higher (P<0.001) hatchability on total egg set (TES) and fertile egg set (FES) was observed in T3 87.87 and 95.38%, whereas, lowest was in T1 75.32 and 84.76%, respectively. Total embryonic death was found to be significantly (P<0.001) reduced in dietary treatment groups and was lowest in T3 (4.73%) whereas, highest in T1 (9.06%). Thus, it can be concluded that dietary supplementation of 0.026% (dose equivalent: 75ppm) soapnut shell powder improved reproductive performance of broiler breeders.

Keywords: Broiler breeders, Fertility, Hatchability, Soapnut, Testosterone

Effect of source of Selenium on Se deposition and egg quality

Abstract ID: 416

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Despite the extensive research that has been conducted comparing inorganic with organic selenium (Se) sources there has not been sufficient research conducted to directly compare levels of commercially-available sources of selenium which are approved for use in poultry feeds. Dusting potential of the preparation of organic selenium L-Selenomethionine need to be less than 0.2mg Selenium /m³ air according to EU regulation. Therefore different coating procedures are used which can affect Se bio-availability. The objective of the trial was to compare Se deposition in the whole egg when hens were fed diets supplemented with various sources of Se.
Laying hens (Lohmann brown), 40 weeks of age at the start of the experiment, were used in this trial. Duration of the trial was 3 weeks: 2 weeks adaptation and 1 week for egg collection. Pelleted feed and drinking water were provided *ad libitum.* The trial consisted of 4 treatments, each with 2 replicates of 16 animals. A corn based, Se free diet was used as a control diet (T1), for the other dietary treatments 0.20 mg/kg inorganic selenite (T2) or organic Se (T3 and T4) was added on top of the control diet. A different source of organic Se was used for T3 and T4.
There was no dietary effect on laying rate, egg weight, daily egg mass, daily feed intake and feed conversion. Selenium content of the eggs significantly increased if Se was supplemented to the diets. Bio-availability of organic Se was higher than selenite as higher Se contents were observed in T3 and T4 compared with T2. By comparing both organic Se sources, Se_1 showed significantly higher bioavailability than Se_2. There were no soft-shelled eggs during this trial and the incidence of cracked eggs and dirty eggs was not significantly influenced by dietary treatments. Mortality percentage was not different between treatments as no hens died during the trial.
It can be concluded that a significant effect of the preparation of supplemented Se on Se content of the eggs was noticed. Significantly higher Se levels were obtained for the organic versus inorganic Se source. Furthermore, a significant effect of the source of organic Se was obtained with Se_1 being more available compared with Se_2.

Keywords: Deposition, Laying hens, Selenium

Effect of two phytases at two doses on performance of broilers during 0-21d of age

Abstract ID: 268

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This study evaluated the effect of two phytases on performance of broilers fed diets containing fine limestone. The limestone had a particle size of <0.09 mm, with *in vitro* Ca solubility of 100% after 30 minutes at pH 3. Five dietary treatments were a positive control (PC, Aviagen specifications NL, 2014) without phytase and 4 test diets that supplemented with *Buttiauxella* phytase at 500 or 1000 FTU/kg (PhyB500 and PhyB1000), or with *Citrobacter* phytase at 1000 or 2000 standard FTU/kg (PhyC1000 and PhyC2000). Test diets were formulated with reduced P (0.187%), retainable P (0.159%), Ca (0.199%) and Na (0.04%) content by varying only MCP, limestone, salt and diamol levels in NC vs PC. Each treatment was tested with 8 replicates (Ross-308 male day-old chickens, 30 birds/pen). Pelleted diets (based on corn, SBM, rapeseed meal and sunflower meal) and water were provided *ad lib*. in two phases (0-10d and 11-21d). At 21 day, the left tibia bone was collected from four birds per pen and pooled per pen for defatted tibia ash measurement. Ileal digesta samples were collected from 12 birds per pen, pooled for phytate P (IP6) analysis. BWG, FI, FCR were determined per phase and for overall 0-21d. Treatment means were compared using Tukey's HSD using JMP 11. In comparison to PC, for overall 0-21d, PhyC1000 showed lower ($P<0.05$) FI (1278 vs 1323g), BWG (1052 vs 1081g) and tibia ash (499 vs 510). PhyB500 showed lower ($P<0.05$) FI (1289 vs 1323) and tibia ash (502 vs 510). PhyB1000 and PhyC2000 maintained all performance parameters to the level of PC. The Ileal IP6 content was reduced by 68, 87, 44 and 60% vs PC, respectively, for PhyB500, PhyB1000, PhyC1000 and PhyC2000 ($P<0.001$). Among the phytase treatments, for overall 0-21d, PhyB1000 had greatest BWG and tibia ash, significantly higher than PhyC1000 (1091 vs 1052g BWG; 508 vs 499g/kg DM for tibia ash), and higher BWG than PhyB500 (1091 vs 1054g), but not significantly differ from PhyC2000 (1091 vs 1070g for BWG). FCR was lower ($P< 0.05$) with PhyC1000 vs PhyB500 during 0-10d but no significant differences were found during 11-21 and 0-21d. PhyB1000 had the lowest IP6 content in ileal digesta, significantly lower than PhyB500, PhyC1000 and PhyC2000 (0.46 vs 1.17, 2.04 and 1.46% DM, respectively), indicating a higher IP6 degradation rate. In conclusion, PhyB showed higher efficacy than PhyC, most likely due to a higher IP6 degradation rate.

Keywords: Broiler, IP6, Performance, Phytase

Effect of xylanase in corn-soybean meal diets on growth performance of broiler chicken

Abstract ID: 476

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The positive effect of xylanases on non-starch polysaccharides rich cereals is well established; however, their effect on corn-based diets is often questioned. The objective of this study was to evaluate the effects of a bacterial origin xylanase on growth performance of broilers fed corn-soybean meal based diets. Cobb male chicks (n=1,440; 1-42 d) were divided into 3 treatments (T), which differed only in feed formulation: T1, standard diet (positive control; PC); T2, diet with 150 kcal/kg reduction in metabolizable energy (negative control, NC); and T3: as T2 with the addition of a *Bacillus subtilis* xylanase (100 g/t; Puratos & Jefo Nutrition). The recommended uplift for the xylanase tested for corn-based diets is 80 kcal/kg in broilers. The purpose of using a greater energy difference between the PC and NC diets was to generate a substantial drop on performance. The hypothesis was that the tested xylanase would be able to partially prevent the performance loss caused by the 150 kcal/kg reduction on dietary metabolizable energy. Growth performance was evaluated by feeding phase and the feed cost/kg of live weight was calculated at 42 days. The difference on body weight gain and feed conversion ratio was observed starting at 14 days throughout the trial. For the overall period, the reduction of 150 kcal/kg on the NC diet caused a drop ($P<0.05$) of 4.6% on average daily gain (ADG) and feed conversion ratio (FCR), which was partially reverted by the addition of the xylanase to the NC diet ($P<0.05$; 49% for ADG and 45% for FCR). The NC diet supplemented with the xylanase had the lowest cost/kg of live weight (USD 0.08 in comparison to the NC diet and USD 0.21 to the PC diet). The recovery in performance confirms the recommended uplift of 80 kcal/kg in broiler diets and the efficacy of this bacterial origin xylanase in reduced energy corn-soybean meal based diets to improve growth performance while reducing the feed cost of broiler chicken diets.

Keywords: Broiler, Corn-soybean meal diet, Growth performance, Xylanase

Effects of a fat coated betaine based product on breast meat yields in turkeys and on performances and blood cells membrane integrity in heat-stressed laying hens

Abstract ID: 273

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Betaine, the trimethyl derivative of the amino acid glycine, has osmoprotective properties and can act as a methyl donor. Hence, dietary betaine supplementation may improve heat stress resistance and breast meat yield (BMY) in poultry. However, betaine may be difficult to manipulate in feed and premix plants due to its high hygroscopicity. Fat coating allows reducing such caking issues and, in addition, improves betaine bioefficiency in broilers. Our aim was to evaluate the effect of a specific fat-coated betaine product (BeTane; vectorization technology) on heat stress resistance and BMY in other poultry species. In a first study, 416 day old male B.U.T. Premium turkeys allocated to a randomized complete block design in 32 pens, received one of the following feeds: NC (negative control, no betaine), B1000 (NC + 1000 mg/kg non-coated betaine), CB500 (NC + 500 mg/kg coated betaine) and CB1000 (NC + 1000 mg/kg coated betaine), each replicated 8 times. Birds were sacrificed at 105 d and BMY was measured. There was no significant effect of betaine addition on overall growth performances. Turkeys fed BeTane through CB500 and CB1000 diets tended to have a higher BMY than their NC and B1000 fed counterparts (+0.35 and +0.49 points, respectively). In a second study, 100 ISA brown laying hens allocated to a randomized complete block design in 50 cages, with 2 birds per cage and 25 replicates per group, received either NC or CB600 (NC + 600 mg/kg coated betaine). The trial was conducted between 71 and 81 weeks of age and high temperature was applied every day (30°C from 8am to 6pm). Blood hemolysis score was measured as cellular damage indicator at 81 weeks of age. Heat stress induced performances variability, but CB600 fed laying hens tended to have a higher exported egg mass (+1.4 g/day) and a lower feed intake per egg (-8.8 g) than NC birds. Birds fed CB600 had, in addition, significantly lower hemolysis scores than NC fed birds (-11%, $P<0.01$). Overall, our results suggest that the positive effect of BeTane observed in broilers can be extrapolated to other poultry species. BeTane can improve BMY in meat poultry and alleviate the detrimental effects of heat stress on laying performance, with a protective effect against cellular damages induced by heat stress.

Keywords: Fat-coated betaine, Heat stress, Laying hens, Meat yields, Turkeys

Effects of a global enzyme solution on growth performance and bone quality of broilers from 1 to 28 days of age

Abstract ID: 190

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The ability of dietary enzyme(s) to increase feed efficiency have been well established, however, the drive to further increase nutrient efficiency necessitates higher enzyme efficiency. The efficacy of a global enzyme solution containing multi-carbohydrase and phytase activities (Enz) supplemented in diets reduced in ME, digestible AA (dAA), Ca and available P (avP) was assessed on performance and bone quality of broilers between 0 and 28 days of age. Five corn-wheat-soybean based dietsdiets were assigned to 60 floor pens with 20 Ross PM3 male broilers each (n=1,200) in a randomized complete block design. The diets included a nutrient adequate-PC; two negative control diets (NC1 and NC2) reduced in avP by 0.18% unit, Ca by 0.16% unit, dAA by 4%, with reduction in ME by 5 (NC1) and 7% (NC2). NC1 and NC2 were supplemented with the Enz. Data were analyzed using Proc Mixed of SAS 9.4. Compared to the PC diet, d0-28 BW gain and FCR were depressed with NC1 (4.9 and 6.4%, respectively) and NC2 (5.1 and 6.4%, respectively). Whereas the d0-28 feed intake was maintained. Inclusion of Enz increased BW gain by 6.8±0.7% and decreased FCR by 2.4±0.1% ($P < 0.001$) in both NC1 and NC2. Also, the NC1 and NC2 decreased bone ash by 7.6±0.5% versus PC which was fully recovered by Enz supplementation (+6.8±0.8%). Similarly, decreases in bone Ca and P in the NC1 and NC2 birds were alleviated with the Enz ($P < 0.001$), whereas no effect was observed on blood Ca and P levels. Apparently, the deficiency of energy, dAA, avP, and Ca in the NC1 and NC2 maintained feed intake but worsened feed to gain efficiency, BW and bone mineralization. The Enz supplementation of the NC diets alleviated the adverse effects. Overall, these results evidenced that use of the global enzyme solution (Enz) enabled to further reduce feed value through lower dietary energy which would significantly reduce feed cost while maintaining performance and skeleton development.

Keywords: Bone, Broiler, Enzyme, Nutrient-deficiency, Performance

Effects of conditioning time and sodium bentonite on pellet quality and performance in growing male broilers

Abstract ID: 368

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The present study was conducted to evaluate the effects of conditioning times and processed sodium bentonite (PSB)-based pellet binder (G. BindÔ) on pellet quality and performance in growing broiler chickens (d 11-24). A total of 540 day-old male broiler chicks were fed a commercial starter diet up to d 10. On day 11, birds were assigned to a 2×3 factorial arrangement including two conditioning times (2 and 4 minutes) and three levels of PSB (0, 7.5 and 15 g/kg) with 6 replicates of 15 chicks each. All diets were tested for pellet durability (PDI) and hardness, feed intake and weight gain were recorded to calculate growth performance. Results indicated that the diet with a 2 minute conditioning time and containing 15 g/kg PSB had the highest ($P< 0.05$) pellet durability index and hardness. Chickens fed the diet conditioned for 2 minutes showed better weight gain and FCR It is concluded that a corn-soybean meal diet conditioned for 2 minutes and containing 15 g/kg PSB may improve pellet quality and performance.

Keywords: Broiler, Conditioning time, Pellet quality, Performance

Effects of dietary inclusion of high- and low-tannin faba bean (Vicia faba L.) seeds on microbiota, histology and fermentation processes in the gastrointestinal tract of finisher turkeys

Abstract ID: 286

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This study evaluated the effects of dietary replacement of soybean meal (SBM) with graded levels of faba bean (FB) seeds with high or low tannin content (HT or LT) on the gastrointestinal function and growth performance of turkeys at 13 – 18 weeks of age. Hybrid Converter turkeys were distributed into 7 dietary treatments: a control *wheat-soybean meal-based* (FB_0) diet and experimental diets where SBM was partially replaced with HT or LT seeds at 10, 20 and 30%. Seven replicate pens of 30 birds each per treatment were used. The LT diet decreased jejunal crypt depth (vs. FB_0) with no significant effect on the caecal histological parameters. In comparison with the FB_0 diet, diets containing HT and LT FB increased the total bacterial counts and *Bacteria* domain and decreased the counts of *Bacteroides*. Diets containing LT FB reduced the abundance of *Salmonella*, relative to the FB_0 diet and diets containing HT FB. The LT diet decreased the counts of total bacteria and *Bacteria* domain, in comparison with the HT treatment. The highest ileal short-chain fatty acid (SCFA) concentrations were observed for the LT_{20} diet. The LT diets stimulated SCFA production in the caeca, relative to the FB_0 diet, and the opposite effect was noted when HT and SBM dietary treatments were compared. In comparison with the HT diets, LT diets led to a desirable increase in the concentrations of all major fatty acids (acetic, propionic and butyric) in the caecal contents. It can be concluded that FB seeds enhanced fermentation processes in the gastrointestinal tract of turkeys. In comparison with HT FB, LT seeds improved selected parameters of intestinal function, including a decrease in the counts of *Salmonella*, increased SCFA production (including butyrate), and a decrease in the pH of intestinal digesta. In conclusion, both LT and HT FB seeds, the latter containing up to 7.1 g/kg tannins, can be included in finisher turkey diets at up to 30% as a safe and effective substitute for SBM.

Keywords: Faba bean, Microbiota, SCFA, Tannin, Turky

Effects of dietary inulin supplementation on growth performance, gut health and bone quality of broilers

Abstract ID: 183

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The present experiment was conducted to investigate the efficacy of 1% dietary inulin supplement (Orafti® SIPX 70%) with or without dietary phytase (Quantum Blue 5G) and endo-xylanase enzymes (Econase XT 25P) on growth performance, gut health, calcium and ash content of tibia of broiler chickens. One-day old male broiler chickens (Ross 308) were placed in 24 deep litter pens (32 chickens per pen) and were randomly assigned to four experimental dietary treatments for 39 days (6 pens per treatment). Dietary treatments were as follows: 1) endo-xylanase and phytase supplementation without inulin, 2) endo-xylanase and phytase supplementation + 1% inulin, 3) diet without enzyme and inulin supplementations, 4) diet without enzyme supplementations + 1 % inulin. Experimental animals were fed starter (d0-10), grower I (d11-21), grower II (d22-32) and finisher (d33-39) diets. Both enzymes were added at the inclusion level of 100 mg/kg. According to our results, inulin supplementation at 1% did not influence the growth performance (body weight, body weight gain), feed intake and feed conversion ratio of broilers significantly ($P>0.05$). However, dietary inulin significantly reduced the concentration of *E. coli* and total number of coliform bacteria in the caecum. Furthermore, higher concentration of butyrate was measured in the ceacal content of broilers fed inulin supplemented diets ($P<0.05$). Also, dietary inulin treatment led to higher ash and calcium content of tibia bones ($P<0.05$). These important and significant effects of dietary inulin were not influenced by the absence or presence of phytase and endo-xylanase enzymes. This study indicates that 1% dietary inulin can be used to improve gut health and bone quality of broiler chickens.
This research was supported by the Hungarian Government and the European Union, with the co-funding of the European Regional Development Fund in the frame of Széchenyi 2020 Programme GINOP-2.3.2-15-2016-00054 project.

Keywords: Bone quality, Broiler, Gut health, Inulin

Effects of dietary tuna oil levels and feeding periods on growth performance and meat quality of Thai crossbred chickens

Abstract ID: 455

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The aim of the present study was to determine the effects of dietary tuna oil levels and feeding periods on growth performance, carcass composition and meat quality of the 'Korat meat chicken', a type of Thai indigenous crossbred chicken. Seven hundred 3-wk-old mixed-sex chicks were randomly allotted to an augmented factorial in completely randomized design model with 7 treatments of 4 replicate pens each. The basal diet based on corn-soybean and 4.5% rice bran oil was used as control. In the experimental diets, part of rice bran oil content was substituted with 1.5, 3.0 or 4.5% tuna oil and fed to chicks 3 or 6 wk before slaughtering at 9 wk of age. The results showed that body weight, feed intake, feed conversion ratio (FCR), meat yields and abdominal fat were not different among treatments ($P>0.05$). There were no differences in pH, cooking loss and shear force of breast and thigh meat among treatments ($P>0.05$). However, the dietary tuna oil and feeding period influenced drip loss in thigh meat and meat color in breast and thigh meat ($P<0.05$). The drip loss in thigh meat of chicks fed dietary tuna oil with different feeding period was lower than control group. The b* value of breast and thigh meat of chicks fed dietary tuna oil with different feeding period was lower than control group. It is concluded that tuna oil supplementation in diet with different feeding period was not showed the detrimental effects on growth performance and meat yields but influenced drip loss and meat color of Thai crossbred chicken.

Keywords: Crossbred chicken, Feeding period, Growth performance, Meat quality, Tuna oil

Effects of different heat damaged protein ingredients on protein digestibility and caecal proteolytic fermentation in broilers

Abstract ID: 236

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Poor digestible proteins or overfeeding of protein may lead to increased fermentation of protein and peptides in the caeca of broilers, which results in the production of toxic metabolites. This proteolytic fermentation may be an important nutritional cause of gut health issues in broiler practice. In order to determine the effects of different quality proteins on caecal proteolytic fermentation, performance and gut health, a total of 480 one day old ROSS 308 male broilers are divided over 6 dietary treatments, with 8 replicate pens with 10 broilers each. Diets contained 20% of a regular soybean meal (SBM), dehulled sunflower meal (SFM) or a mildly processed dehulled rapeseed meal (RSM) as is, or heat damaged by secondary toasting at 136°C for 20 minutes (hSBM, hSFM or hRSM, respectively). From 27 days of age 5 broilers from each pen are housed on slats to allow for faecal collection. On day 29, 30 and 31 broilers are culled for the collection of ileal and cecal content as well as to sample and weigh various organs. Both ileal and faecal apparent digestibility of amino acids (AA) is determined using two markers in the feed; TiO_2 as marker for the solid fraction and Co-ETDA as marker for the soluble fraction. Two markers are used because solid and soluble fractions of digesta behave differently in the gastrointestinal tract. The difference in AA profile between the ileal and faecal samples indicates the level and type of hindgut fermentation. Caecal content is stored for later determination of the microbiota profiles and proteolytic metabolites such as ammonia, biogenic amines and branch chain fatty acids. Results will be presented at the conference in due time.

Keywords: Broilers, Performance, Protein digestibility, Proteolytic fermentation

Effects of different levels of natural glauconite and zeolite on performance, tibia bone characteristics and blood parameters of broiler chicken

Abstract ID: 572

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An experiment with 300 one-day old Ross male broiler chicks was conducted to determine the effects of glauconite and zeolite on the broiler's performance, tibia bone and blood characteristics. Five experimental treatments [control, glauconite (2 and 4 %), and zeolite (2 and 4 %)] were used in a completely randomized design with 4 replicates. During the experiment weight gain, feed consumption and feed conversion ratio (FCR) were measured periodically. At 42 days of age, one chick per replicate was slaughtered to determine calcium and phosphorus of blood serum and tibia bone parameters. Analysis of variance and separation of means by Duncan's multiple range tests were conducted by SAS software. The results indicated that by adding 4% zeolite to diet, weight gain was increased in starter and total rearing period ($P<0.05$). In comparison with other treatments, feed consumption at 0-42 days of age was increased significantly ($P<0.05$) in 4% zeolite treatment except 4% glauconite group. There were no significant differences in FCR among treatments ($P>0.05$). Also, adding 4% zeolite led to significantly ($P<0.05$) increase in tibia bone volume compared to 2% zeolite group but experimental diets had not significant effect on tibia bone relative weight, length and density ($P>0.05$). Supplementation of diets with glauconite and zeolite did not have effect on serum Ca and P content at the end of experiment ($P>0.05$).

Keywords: Broiler; Glauconite; Zeolite; Performance; Tibia

Effects of in ovo and dietary supplementation of flavanone onantioxidant defence system and performance of postnatal broiler

Abstract ID: 527

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Oxidative metabolism increases during the late embryonic development, substantially over the incubation period few days before the hatching. Flavanones from citrus fruits such as hesperidin and Naringin plays role as natural plant antioxidants and immune system promoter stimulator. This experiment was conducted to investigate the effects of amniotic injection and dietary feeding of flavanoid (Naringin and Hesperidin) on hatchability, performance, antioxidant status and stability in serum and meat by chicks divided randomly in four replication with 10 Ross birds after hatching for growing period until 28 day. Treatments based on acompletely randomized design were (Low or high additives) 1, 2 and 3) 50, 100, 175 mg/kg flavonoids in diet, respectively, 4 and 5) 15, 30 mg/egg injection of flavonoid respectively and 6) injection of 15 mg/ egg and feeding 50 mg/kg flavonoid and 7) injection of 30 mg/ egg and feeding175 mg/kg and 8) control group (no injection nor additives of flavonoids). Data were analyzed using the General Linear Model procedure of SAS. Treatment means were compared using the Duncan's Multiple Range Test, and values were considered statistically different at $P<0.05$. Intra amniotic injection at 17.5th day of incubation showed more hatchability with 15 mg/ egg (low) flavanone and sham group than the un-injected and 30 mg/ egg (high) substance injection (80 vs 85 %). Treatments 4 and 6 did have lower daily feed intake ($P<0.05$) but daily gain weight or feed efficiency until 28 day old of fast-growing broilers showed no difference. In ovo injection with 30 mg/egg flavanone significantly increased serum superoxide dismutase (SOD) in the hatch-day chick than other treatments but glutathione peroxidase (GSH-PX) didn't show changes. At 4 weeks after hatching these antioxidant capacities were improved in treatments that received dietary flavonoid compare to 15 mg in ovo feeding (treatment 4) and control. Reduction of Malondialdehyde in tight and breast meat was observed in treatments (except 4) at 28 days than in the control group. Ovo feeding of flavonoid decreased feed intake. Flavanones had positive effects on hatchability, primary defence mechanism of newly hatched chicks and growing broilers. Meat quality improved by dietary flavonoid with or without in ovo feeding of flavonoid.

Keywords: Broiler, Glutathione peroxidase, Late embryonic growth, Malondialdehyde

Effects of multispecies synbiotic on performance parameters of commercial turkey hens

Abstract ID: 347

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Probiotics and prebiotics influence the intestinal tract in a positive way thus improving the health, well-being and performance of animals. A 14-week study was conducted with Hybrid Converter female birds to evaluate the effect of a multispecies synbiotic feed additive on growth performance. 480 one-day old chicks were randomly assigned to two dietary treatments with 20 replicates per group and 12 birds per replicate. Birds were kept in floor pens on wood shavings, environmental conditions were according to the recommendations of the management guide. Animals of both groups were fed with a standard commercial corn-soybean based mashed diet in a four phase feeding program (starter (0-28 days), grower I (28-56 days), grower II (56-84 days), grower III (84-98 days)). The diets were formulated to have similar calculated nutrients and did not contain antibiotic growth promoters or coccidistats. Feed and water were provided *ad libitum.* The experimental group received a synbiotic multispecies product (PoultryStar® me^{EU}, BIOMIN GmbH) at 0.5 kg/ton while the control group was given diets without the synbiotic. The birds were kept under observation for 98 days and production performance parameters were recorded. By the addition of the synbiotic product the feed conversion ratio was significantly improved over the period 0-28 days (1.52 vs. 1.57, P=0.046), 0-84 days (2.02 vs. 2.10, P=0.002) and over the total trial period of 98 days (2.32 vs. 2.38, P=0.036). Furthermore, a significantly higher live weight could be observed after 28 days (1000 g vs. 963 g, P=<0.001) and after 84 days (6520 g vs. 6407 g, P=0.003) and a numerically higher live weight after 98 days in comparison to the control group. Therefore, it was concluded that the multispecies synbiotic was effective in enhancing turkey performance.

Keywords: Feed additive, Performance, Synbiotic, Turkey

Effects of probiotic (Saccharomyces cerevisiae) and antibiotic Neomycin on performance and some carcass characteristics of broiler chickens

Abstract ID: 559

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This study was carried out to investigate the effects of probiotics (*Saccharomyces cerevisiae*) and antibiotic Neomycin on performance and some carcass characteristics of broiler chickens for 42 days using a completely randomized design. A total of 108 day-old male broiler chicks (Ross 308) were randomly assigned to 3 treatments, 3 replicates and 12 chicks per replicate. The experimental treatments consisted of: control treatment (basal diet), Saccharomyces cerevisiae live yeast (0.1 % of diet) and Neomycin (0.02 % of diet). Results showed that experiment treatments had not significant effect on body weight gain, feed intake, and feed conversion ratio in experimental period but conversion ratio in grower period had significantly decreased in Neomycin treatment compared to Saccharomyces cerevisiae treatment (P<0.05). In Saccharomyces cerevisiae treatment significantly increased Ileum height, gizzard, caeca, proventriculus weight at 42d of age. Spleen weight in Saccharomyces cerevisiae and Neomycin treatment significantly increased (P<0.05). No significant differences were observed in among groups other performance parameters. This study showed that inclusion of probiotics in broiler diets had not remarkable effects on performance and carcass characteristics, but adding probiotics significantly increased Ileum height, gizzard, caeca, proventriculus and spleen weight of broiler chickens.

Keywords: Broiler chickens, Carcass characteristics, Neomycin, Performance, Saccharomyces cerevisiae

Effects of supplementing D or L- Methionine on productive performance and egg quality in laying hens subjected to chronic cyclic heat stress

Abstract ID: 144

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The objective was to evaluate performance and egg quality of laying hens given D and L-Methionine (Met) under chronic cyclic heat stress (HS). A total of 240 18-week-old HyLine hens, raised during pullet phase under HS (35°C/7h/d), were distributed in a completely randomized design, with a 2 by 5 factorial arrangement (4 replicates/6 birds each). Temperature and diets were the main factors. Two rooms were used, differing only in temperature: control room (CR) - 21°C/24h/d, and heat room (HR) – 32°C/8h/d. The diets were given as a percentage of HyLine TSAA requirement and were: T1=70% (no Met supplementation), T2=85%+D-Met, T3=100%+D-Met, T4=85%+L-Met, and T5=100%+L-Met. Body weight gain (BWG), feed intake (FI), feed conversion (FC), egg production and quality were evaluated at 34 wks. Means were analyzed using ANOVA and compared by Tukey`s test ($P<0.05$). No interactions were found for BWG, FC, egg production, egg, yolk, and shell weights, specific gravity nor shell thickness. BWG and egg production were affected by room and diet. T1 and HR had the lowest values for both variables. FC was affected by diets, T1 had the worse FC compared to other treatments. Egg weight was higher for T3 and T5 than for T1, whereas T2 and T4 showed intermediate results. Yolk weights were higher for T5 versus T1. Specific gravity and shell thickness were higher for T1 compared to T2, T3 and T4, whereas T5 was similar to all treatments. Shell weight was influenced by temperature, with CR showing higher values than HR. There was a significant interaction between the factors for FI and albumen weight. In CR T2, T4 and T5 had higher FI than T1 and the treatments in HR, and in HR T1 had the lowest value. For albumen weight, in CR T3, T4 and T5 showed higher results compared to T1; T2 showed intermediate value. In HR, T5 had higher albumen weight compared to T4 and no other comparisons were significantly different. 100% of TSAA with L-Met improved egg and yolk weight when compared to 70% of TSAA, which had the worse results for productive performance compared to the other treatments at 34 weeks of age. In conclusion, Met supplementation is essential to maintain good performance and improve egg quality during HS.

Keywords: Egg quality, Heat stress, L- Methionine, Laying hens

Efficacy of an algo-clay complex on decreasing mycotoxins liver toxicity on broiler

Abstract ID: 450

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The aim of this study was to measure the efficacy of an algo-clay complex on T2-HT2 toxins, fumonisins and aflatoxins individual liver toxicity. Three trials were conducted by the Samitec Institute of Analytical (Brazil). 1080 broilers chickens (Cobb 500) were used in total for the 3 trials. 360 animals were allocated to 5 treatments with 6 replicates within each of the 5 tests treatments or 12 replicates within the control treatment. Each group contained 10 animals. The study was run from day 1 to day 21. 3 trials were set up allowing to test each mycotoxin individually at a contamination level of 2.8ppm for aflatoxins, 100ppm for fumonisins and 2ppm for T-2/HT-2 toxins. Treatments differed by the presence each individual mycotoxin object of study, alone or with an inclusion of the algo-clay complex at 2.5kg/ton or 5kg/ton. Performance and liver parameters were measured: feed intake (FI), body weight (BW), individual relative liver weight (RWL), Sphinganine-to-Sphingosine ratio (Sa/So) for fumonisins, Lamic/Samitec Index (LSI) for aflatoxins and total plasma proteins (TPP). The inclusion of 0.50% of algo-clay complex in the diets containing mycotoxins significantly improved FI and BW compared to diets containing mycotoxins only ($P\leq0.05$). In each study, the RWL of the birds receiving 0.50% of the algo-clay complex, was significantly improved when compared to those receiving mycotoxins only. The inclusion of 0.50% of algo-clay complex in the diets containing 2.8 ppm of aflatoxins improved significantly the LSI compared with those diets containing aflatoxins only ($P\leq0.05$). The inclusion of 0.25% and 0.50% of algo-clay complex in the diets containing 100 ppm of fumonisins diminished significantly the Sa/So compared with those from the birds fed with fumonisins only ($P\leq0.05$). The inclusion of 0.25% and 0.50% of algo-clay complex improved TPP ($P\leq0.05$). The algo-clay complex decreased significantly ($P\leq0.05$) the deleterious hepatic effects and performance losses caused by very high level of 3 types of mycotoxins on broiler.

Keywords: Algae, Clay, Mycotoxins, Poultry

Emulsifier and carbohydrase in a maize-wheat-SBM-tallow diet for broiler chickens

Abstract ID: 55

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Under normal conditions, the gastrointestinal tract of a chicken is an aqueous environment. Fatty acids, as hydrophobic components, have to aggregate to form micelles to get absorbed. Emulsifiers naturally mediate this process. Our results showed that use of exogenous emulsifiers had a positive effect also on neutral detergent fibre total tract digestibility. We assumed that improved fat digestibility reduced its content in digesta and, consequently, enhanced carbohydrate availability for microbe enzymes. It is common practice is to use carbohydrases to prevent negative effects (high digesta viscosity or/and digestibility deterioration) of carbohydrates. On the basis of the above, it could be assumed that the use of exogenous emulsifiers and carbohydrases in poultry diets may have an additive effect on carbohydrate utilization by poultry. The experiment was conducted with 480 one day old ROSS 308 male chickens. Birds were randomly located in floor pens and assigned to 4 dietary treatments (15 replication in each, 8 birds per replication). The first group was fed a basal diet (BD) (maize-wheat-SBM-tallow) without any supplementation. The second treatment consisted of a BD and an emulsifier additive (E), whereas BD in the third group was supplemented by xylanase (X). In the fourth group, both supplements were added to the diet (E+X). The content of tallow in the diet changed during the experiment from 0 (starter diet) to over 6 % (in finisher diet). There was a tendency ($P=0.051$) to improved FI after E and E+X addition. Body weight gain at day 42 of experiment increased when the diet was supplemented with X or E+X, ($P<0.001$). The overall FCR was improved after X, E or E+X addition ($P<0.01$). The lowest FCR (0-42d) was noted for E+X treatment ($P<0.01$). Fat digestibility was improved ($P<0.05$), after X, E and E+X addition. Differences were more pronounced at 28 than 35d of the experiment. The results of our study indicate positive effects of feeding of the experimental emulsifier throughout the 42 d broiler growth period when the compound was used either alone or in combination with xylanase.

*Research supported by the National Science Centre, Poland, grant 2015/19/D/NZ9/03580

Keywords: Broiler chicken, Carbohydrase, Emulsifier, Tallow, Wheat

Encapsulated sodium butyrate to improve gut development and control enteric bacteria in broilers

Abstract ID: 418

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Two studies were conducted to evaluate the encapsulated sodium butyrate (Na-B) with different releasing time on broiler performance and gut development following challenge with the *Salmonella* Typhimurium and *Clostridium perfringens*. Both studies included 6 treatments: non-challenged control, challenged control, 2 h releasing time product (500, 1000 ppm) and 3-4 releasing time product (500, 1000 ppm). In Exp. 1, a total of 396 male broilers were placed 11 birds per cage into 6 replicates. Birds were orally gavaged with 0.1 mL of 10^7 cfu/mL *Salmonella* Typhimurium on d 4. Ceca were collected from 2 birds per cage on d 11 and 21. Intestinal samples were collected on d 21. In Exp. 2, a total of 336 male broilers were placed 8 birds per cage into 7 replicates. Challenge treatment birds were orally gavaged with ~5,000 oocysts of *Eimeria maxima* on d 14, following a 10^8 cfu/mL *Clostridium perfringens* challenge on d 19, 20 and 21. On d 21, 3 birds were randomly selected per cage and scored for intestinal lesions. Results were analyzed using a one-way ANOVA via JMP. There was no significant difference on *Salmonella* colonization between the challenged control and Na-B treatments in Exp. 1. For the intestinal results, we only found adding Na-B product showed a numerically higher ($P = 0.08$) on the jejunum villus height than the challenge control treatment in Exp. 2, but no difference was observed on villus height to crypt depth ratio. Both products at 500 or 1000 ppm showed the significantly ($P < 0.05$) lower lesion scores compared to the challenged control in Exp. 2. In conclusion, adding encapsulated Na-B showed the potential to mitigate the impact on necrotic enteritis in broilers. However, no significant influence on *Salmonella* control was evident in current study.

Keywords: Broiler, Growth, Gut health, Sodium butyrate

Ensiling of mixed forages of beans and grains as feed for organically raised laying hens

Abstract ID: 446

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In an attempt to maximize the use of regional protein sources and more specifically for organically produced laying hens, pulses have received a lot of attention lately, in particular field beans and peas. However, the presence of anti-nutritional factors (ANF) like tannins, glycosides (such as vicine and convicine in faba beans), trypsine and protease inhibitors together with a low ileal digestibility of methionine and cysteine of beans limit the inclusion of faba beans in poultry feeds. The ensiling of these faba beans and the combination of these with cereals is a possible solution to reduce these ANF and to compensate the shortage in certain amino acids.
In order to assess to which extent the ANF of faba beans are reduced when these are ensiled as moist beans in combination with cereals, one needs to develop a method to identify vicine and convicine in dried faba beans as well as in ensiled beans. For this, an LC-PDA-HRMS metohd is currently being been implemented. In this method finely grounded beans are extracted with water and the extract is separated in HILLIC mode on an Acquity UPLC ® BEH Amide column (1.7 µm; 2.1 x 150 mm, Waters). Vicine and convicine are detected at 273 nm with the PDA detector. High resolution mass spectrometry (HRMS) is used to identify breakdown products of these compounds. Due to the lack of commercial available convicine standard, convicine is quantified using the response of the vicine calibration curve. Different varieties of dried and ensiled faba beans are currently being analysed for their content in vicine and convicine. Data from these analyses is important to assess the reduction or not in vicine and convicine levels of ensiled faba beans. The method and information acquired with these analyses will be used to demonstrate that ensiling of mixed forages of beans and grains is a successful technique to reduce the levels of vicine and convicine. In this way, faba beans can be valorised and used as protein source in organically produced laying hens, without detrimental effects for the performance, production and quality of the eggs.

Keywords: Convicine, Faba beans, Laying hens, Silages, Vicine

Evaluating probiotic application alone or in combination with antibiotic growth promoters on broiler performance and health status

Abstract ID: 29

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A study was conducted to compare broiler performance and health status under the influence of probiotic application, alone or in combination with antibiotic growth promoters (AGP's). A total of 1260 Ross 308 male day old broiler chicks were randomly assigned to seven treatments, each consisting of 6 replicates with 30 birds per replicate. The trial was conducted for a period of 42 days Starter 0-9 days, Grower 10-24 days, Finisher 1 25-35 days, Finisher II 36-42 days). The dietary treatments included a corn-soybean based control diet without growth promoter (T1) and the treatment diets containing either Bacitracin, (100 ppm, T2), Colistin (10 ppm, T3), Probiotic (PoultryStar® me, 0.5 kg/t, T4) or a combination of probiotic and Bacitracin (T5), Probiotic and Colistin (T6) and Probiotic, Bacitracin and Colistin (T7). Results revealed that probiotic application (alone) in broiler feed resulted in significantly higher body weight gain (BWG) than its combination with Bacitracin ($P<0.05$) during the critical phase of rearing from 0-9 days. None of the other treatments showed any significant improvement in BWG, feed intake (FI) or feed conversion ratio (FCR) compared to the only probiotic application (T4) during the overall rearing period from 0-42 days ($P<0.05$). FCR was lower in the probiotic group (T4, 1.87) than control (T1, 1.93) during overall rearing period. Likewise, bird mortality was also lower in the probiotic group (T4, 1.11%) compared to control (T1, 2.78%). Hence, it can be concluded that PoultryStar® me (probiotic employed) could serve as a replacement and an effective alternative for the use of AGP's like Bacitracin and Colistin in broiler feeds. The probiotic can serve this purpose alone without any combination with AGP's like Bacitracin and/or Colistin and thus could be cost effective for broiler production.

Keywords: Antibiotic growth promoters, Broilers, Performance, Probiotic

Evaluating probiotic application alone or in combination with antibiotic growth promoters on broiler performance and health status

Abstract ID: 362

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A study was conducted to compare broiler performance and health status under the influence of probiotic application, alone or in combination with antibiotic growth promoters (AGP's). A total of 1260 Ross 308 male day old broiler chicks were randomly assigned to seven treatments, each consisting of 6 replicates with 30 birds per replicate. The trial was conducted for a period of 42 days Starter 0-9 days, Grower 10-24 days, Finisher 1 25-35 days, Finisher II 36-42 days). The dietary treatments included a corn-soybean based control diet without growth promoter (T1) and the treatment diets containing either Bacitracin, (100 ppm, T2), Colistin (10 ppm, T3), Probiotic (PoultryStar® me, 0.5 kg/t, T4) or a combination of probiotic and Bacitracin (T5), Probiotic and Colistin (T6) and Probiotic, Bacitracin and Colistin (T7). Results revealed that probiotic application (alone) in broiler feed resulted in significantly higher body weight gain (BWG) than its combination with Bacitracin ($P<0.05$) during the critical phase of rearing from 0-9 days. None of the other treatments showed any significant improvement in BWG, feed intake (FI) or feed conversion ratio (FCR) compared to the only probiotic application (T4) during the overall rearing period from 0-42 days ($P<0.05$). Although statistically insignificant, FCR was lower in the probiotic group (T4, 1.87) than control (T1, 1.93) during overall rearing period. Likewise, bird mortality was also lower in the probiotic group (T4, 1.11%) compared to control (T1, 2.78%). Hence, it can be concluded that PoultryStar® me (probiotic employed) could serve as a replacement and an effective alternative for the use of AGP's like Bacitracin and Colistin in broiler feeds. The probiotic can serve this purpose alone without any combination with AGP's like Bacitracin and/or Colistin and thus could be cost effective for broiler production.

Keywords: Antibiotic growth promoters, Broilers, Performance, Probiotic

Evaluation of a global enzyme solution on growth performance and carcass characteristics of broilers fed corn-wheat-soybean based diets reduced in metabolizable energy and nutrients

Abstract ID: 188

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The use of a global enzyme solution, combining multi-carbohydrolase and phytase, in poultry diet allows to reduce to a greater extent the nutrient levels compared to individual enzyme approach. This experiment was conducted to investigate if supplementation with a global enzyme solution, containing multi-carbohydrolase and phytase (Enz) could alleviate the negative effects of a large deficiency in energy (ME), digestible amino acid (dAA) and minerals on performance and carcass characteristics. A total of 1,020 Cobb 500 male chicks were fed a common diet for the first ten days then randomly assigned to 5 dietary treatments with 12 pens of 17 birds. Bird were fed an adequate nutrient diet (positive control; PC) or one of 2 negative control diets (NC1 and NC2). Both NC diets were reduced in avP and Ca by 0.18% and 0.16% units, respectively, and differently reduced in ME and dAA by 5% (NC1) or 7% (NC2) compared to the PC or these NC diets supplemented with or without Enz (1,250 xylanase U and 1,000 FTU/kg diet). Body weight (BW) and feed intake (FI) were measured at 28 and 42 days of age. Body weight gain (BWG) and feed conversion ratio (FCR) were calculated. Additionally, on d42, eight randomly selected birds per pen were used for carcass yield determination. Results showed that nutrient reductions in the NC1 and NC2 relative to the PC significantly decreased ($P < 0.0001$) the BWG 10-42 d by 21.1 and 15.8%, respectively, FI by 13.2 and 8.1%, respectively, and increased FCR by 10 and 9.2%, respectively. Moreover, the 42 d carcass, breast and leg yields were also degraded for birds fed NC diets by 2.14, 1.40, and 0.53% unit, respectively ($P < 0.0001$). The supplementation of those nutrient deficient diets (NC1 or NC2) with Enz fully restore the performance, carcass, breast and leg yields to the same levels as the broilers fed PC diet. The results indicate that the global enzyme supplementation (Enz) has the capacity to restore the performance and carcass characteristics suggesting the possibility to lower feed cost while maintaining performance.

Keywords: Broilers, Carcass characteristics, Enzyme combination, Performance

Evaluation of egg quality and vitamin D transfer following vitamin D2-rich yeast supplementation in laying hens

Abstract ID: 54

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Vitamin D (VD) is a fat-soluble vitamin generated in human skin by UV light. It plays a central role in bone mineralization and calcium homeostasis and is involved in numerous human disorders. VD deficiency contributes to higher risk of metabolic bone disease and other non-skeletal chronic diseases. These research results trigger a new demand for VD supplements and fortified food. In this context, VD enriched eggs would present interesting and natural sources of VD for humans. This study investigates the effects of a VD2-rich yeast supplementation in laying hens on the transfer of different VD forms in the egg and on different parameters of egg quality. 96 laying hens were divided in 2 groups: control group (C: 2200 IU VD3/kg in the basal diet) and group D supplemented with VD2-rich yeast (D: 2200 IU VD3/kg in the basal diet + 10000 IU VD2/kg, Vitacell, Lallemand). The trial lasted 91 days and the level of different forms of VD was analyzed by LC-MS-MS in 2 eggs pooled per pen at the end of the supplementation period. The parameters of egg quality (Haugh units, yolk and eggshell weight and eggshell resistance) were also measured at different time-points during the trial (38, 50, 63 and 74d). Data comparison between both groups was statistically analyzed by T-test and significant results were reported for $P < 0.05$. The transfer of VD was significantly improved in group D (VD2: 51.9 IU; VD3: 32.3 IU; 25(OH)D2: 17.2 IU) compared to group C (VD2: 1.2 IU; VD3: 20.5 IU; 25(OH)D2: 1.5 IU) ($P < 0.001$), while there was no significant difference for 25(OH)D3 (C: 27.1 IU; D: 24.7 IU). The total VD content therefore reached 126.2 IU of VD equivalent/2 eggs in group D (vs 50.3 IU for group C, $P < 0.001$). Besides egg VD transfer, parameters of egg quality were increased by 2% in group D for Haugh units ($P < 0.05$), relative yolk weight ($P < 0.05$) and relative eggshell weight ($P < 0.1$). Eggshell resistance was also improved by 5% ($P < 0.001$) and eggshell appeared stiffer in group D (3% less deformation). These results underlined the interest of supplementing laying hens with this VD2-rich yeast to significantly enrich VD in eggs for human health purposes.

Keywords: Egg quality, Laying hens, Vitamin D, Yeast

Growth performance of broilers chicken fed different concentration of trona (sodium sesquicarbonate) treated feather meal

Abstract ID: 13

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In the study, the growth performance of broilers chicken fed feather meal treated with different concentrations of Trona (sodium sesquicarbonate) was evaluated using one hundred and twenty birds. The birds were randomly allotted to four dietary treatments. The treatments represented feather meal which was treated with 0, 5, and 10% Trona being included into the diets at 3%. The control diet had no feather meal included. Completely randomized design was used for the two phase study (starter and finisher). The parameters evaluated were feed intake, body weight, body weight gain, feed conversion ratio and apparent nutrient digestibility. Results revealed that total feed intake, final body weight per bird and mean body weight gain per bird per week were not significantly ($P>0.05$) affected by the feeding of the experimental diets to the birds at both the starter and finisher phases of the experiment. Crude protein, ash, ether extract and nitrogen free extract digestibility were affected significantly ($P<0.05$) by the feeding of the diets at the starter phase, while only crude protein and nitrogen free extract digestibility were affected significantly (p$P<0.05$) at the finisher phase of the experiment. It was concluded that feather meal treated with up to 10% Trona when fed at 3% inclusion in the diets led to no negative effect on growth performance of broilers chicken.

Keywords: Broilers, Feather meal, Growth performance, Trona

Growth response of Ross 308 broiler chickens fed different dietary energy levels

Abstract ID: 166

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In broiler chicken production, the energy intake is considered a primary factor, able to affect not only growth and production parameters but also involved in several metabolic functions. The broiler energy requirements are already been deeply studied, however, with the constant genetic evolution of commercial hybrids, an update on this point could be useful to follow this evolution and to adapt the formulation approaches. The aim of this trial was to evaluate how broilers Ross308 react to different levels of dietary energy during the finisher period (d21-d43). A total of 2700 one day-old male broilers (Ross 308) were randomly assigned to 6 treatments, each consisting in 6 floor pens as replicates (75 chicks/pen). Diets were split in 2 phases, a starter common diet (1-21 d) and finisher (21- 43d) where broilers received one out of the 6 experimental diets varying in their energy content (+100 Kcal/kg), from 2700 kcal/kg to 3200 kcal/kg, at 20% of crude protein. Feed consumption (FC), body weight (BW), average daily gain (ADG) and feed conversion ratio (FCR) were determined on the pen basis. Differences were studied by one-way ANOVA, followed by Tukey's post hoc test ($P<0.05$). At day 43, there were different steps in the effect of energy concentration on feed intake. From 2700 to 2900 kcal/kg, consumption increases very slightly, indicating perhaps a reaction of birds to meet energy requirements. BWG follows this increase. Conversely, from 2900 to 2700 kcal/ kg, chickens should logically increase their feed intake in order to adjust their energy intake, but the lower feed intake recorded could be explained by a limited capacity of ingestion. From 3000 to 3200 kcal/kg, the feed intake decreases linearly but the BWG remains constant, indicating that 20%CP is sufficient. There is a significant 0.04 point FCR improvement for each increment of 100 kcal/kg. In conclusion, 2900 kcal/kg is enough at 20% CP to maximize the BWG, but there is still a room of improvement, potentially more than 0.1 point FCR, to optimize feed efficiency from 2900 kcal/kg to 3200 kcal/kg.

Keywords: Broiler chickens, Dietary energy, Ross 308

Guanidinoacetic acid supplementation in broiler chickens fed corn-soybean diets affects performance in the finisher period and energy metabolites in breast muscle independent of diet nutrient density

Abstract ID: 453

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Guanidinoacetic acid (GAA) is the single immediate endogenous precursor of creatine. It was hypothesized that dietary GAA would have different effects on performance and energy metabolites in breast muscle depending on the nutrient density (ND) of corn-soybean based diets. A total of 540 day-old male Ross 308 broilers were allocated to 9 dietary treatments with 6 replicates (10 birds each) in a 3 × 3 factorial arrangement with 3 levels of ND (low, 2800; medium, 2950 and high, 3100 kcal ME/kg; and with the other nutrients being constant relative to ME) and with 3 levels of GAA (0, 0.6 and 1.2 g/kg) for 42 days. In the starter and grower period, increasing levels of ND improved BW, ADG, ADFI and FCR, with the exception of ADFI in the starter period. GAA supplementation did not change these performance characteristics. All performance indicators responded markedly to increasing ND in the finisher period, whereas the highest GAA level reduced ADFI compared to control (156 *vs*. 162 g/d) and concomitantly FCR (1.81 *vs*. 1.93). No interactive effects were noted for any performance trait. The high ND level resulted in a higher breast yield at d42, associated with higher fat content and darker color compared to the other ND levels, whereas GAA supplementation did not affect carcass and breast traits. At the end of the experiment, creatine was elevated when feeding GAA at 1.2 g/kg (5455 *vs*. 4338 mg/kg fresh muscle). To conclude, ND had a substantial effect on performance and carcass traits, whereas the effect of GAA was limited to FCR in the finisher period and independent of diet ND level.

Keywords: ATP, Breast, Broiler, Creatine, Energy, Guanidinoacetic acid

Hydroxy trace minerals allow dosage reduction without growth impairment of broilers

Abstract ID: 553

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Long term use of inorganic trace minerals (TM) have resulted in heavy metal deposition in soil. Continuous use of high dosages of TM in animal feed has been linked to cause antimicrobial resistance of several bacterial strains causing human health hazards. Future changes in EU regulation will demand to reduce the amount of TM in animal feed. Hydroxy TM have a higher bioavailability than inorganic sulphate and oxide TM. Next to higher amounts of minerals available to the animal's metabolism, they show a positive effect on intestinal health, supporting animal performance. To investigate the effect of reduced dosing of hydroxy minerals* on mineral deposition, animal health and performance, a diet with practical dosages of inorganic copper (15 ppm) and zinc sulphates (120 ppm) was compared with a diet containing lower dosages of hydroxy copper (10 ppm) and zinc (80 ppm). These diets were fed to 870 broilers (Ross 308), housed in pens with 30 birds each. The trial lasted from day 1 to 42 of age and copper deposition in liver, performance parameters and footpad scores were measured. Footpad lesions were scored from 0 (no lesions) to 4 (severe lesions). Results showed 16% higher copper deposition in liver for hydroxy treatment vs. control (15.38 vs. 13.29 ppm/dry matter). Performance results showed no growth impairment due to reduction of mineral dosage. Feed conversion ratio of the hydroxy treatment was significantly better than the control treatment in the first 10 days of trial (1.072 vs. 1.327) and numerically improved in the grower phase (d11-28: 1.485 vs. 1.506) and in the whole period (1.569 vs. 1.607). Footpad lesion results from day 28 showed numerically higher incidence of lower scores (0 and 1) in the hydroxy treatment and higher scores (2 and 3) in the control treatment. Results of this trial show that reducing the dosing of high bioavailable TM in broiler feeds is possible without a negative effect on growth performance. Hydroxy TM at lower dosing than inorganic sulphate mineral sources also show higher copper deposition and improved footpad health. These findings provide a good indication for future research about the effects of higher bioavailable TM sources and the reduction of mineral contents in poultry feeds.
* Excential Smart C and Z (Orffa)

Keywords: Broiler performance, Footpad health, Intestinal health, Mineral reduction, Trace minerals

Ileal phosphorus digestibility of soybean meal in broilers of different ages

Abstract ID: 381

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Poultry industry is facing an ever increased challenge due to the increase in the prices for feed phosphates and public concerns of environmental pollution associated with excessive excretion of phosphorus. Accurate determination of phosphorus (P) digestibility, as an estimation of availability, in poultry feed ingredients is crucial for precision feed formulation. There are many factors, such as different ingredients, batches and bird's age may contribute to variations in P digestibility. The aim of current study was to examine the age effect on ileal P digestibility and retention of soybean meal (SBM) in broilers. Forty-eight male Ross 308 birds were fed the experimental diet containing the test ingredient of SBM which provided the only source of P in the experimental diet. The dietary concentration of Ca and P were 6.5 and 2.9 g/kg diet, respectively. Celite (a source of acid insoluble ash) acting as indigestible marker was added to allow calculation of P digestibility. The same diet was offered in mash form to 4 replicate cages with 6 birds per cage from day 22 to 27 and from day 37 to 42, respectively. Commercial broiler feeds were fed prior to each five days of experimental period. At day 27 and 42, excreta and digesta from the lower half of the ileum were collected, pooled per cage, freeze dried and ground to pass through 0.5 mm sieve. The diets and digesta samples were analysed for P and acid insoluble ash contents. Ileal P digestibility and retention were calculated. The results indicate that both ileal P digestibility (62.5 vs 59.7) and P retention (55.5 vs 37.6) of SBM were higher in younger (day 27) than older (day 42) broilers. The differences, however, were only significant ($P<0.05$) for P retention. Perhaps this reflects changes in broiler requirements for P as they age.

Keywords: Age, Broiler, Ileal phosphorus digestibility

Implication of dietary willow bark extract (Salix alba) on performance and caecal microflora of broilers (14-28 days) reared at 32°C

Abstract ID: 361

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The 14-d feeding trial was performed on 60, Cobb 500 broiler chicks (14-28 days) assigned to 2 groups (C, E) housed in an experimental hall with 32 °C air temperature, 36% humidity and 23 h light regimen. The conventional diet (C), with corn and soybean meal as basic ingredients, had 3082.48 kcal/kg metabolisable energy and 19.99 % crude protein. Compared to the diet of C group, the diet of experimental group (E) included 0.5 % willow bark extract (*Salix alba*). The following performances were monitored throughout the experimental period: bodyweight; average daily feed intake; average daily weight gain; feed conversion ratio. According to the experimental protocol approved by the Ethics Commission of IBNA at the age of 28 days, 6 broilers/ group were slaughtered and samples of caecal content were collected for bacteriological examination. Feed conversion ratio (14-28 days) was higher at broiler chicks of E group (1.52 kg feed/kg gain) compared to broilers chicks of group C (1.46 kg feed/kg gain), but it was not statistically assured ($P \geq 0.05$). As regards to the other performances (body weight, average daily gain, average daily intake) there were no significant differences ($P \geq 0.05$) between the two groups although in absolute value they were higher for the control group. Throughout the experimental period, under heat stress, no mortalities were recorded. The pathogenic bacteria, *Enterobacteriaceae and E.coli,* (expressed as colony forming units) were significantly ($P \leq 0.05$) lower at group E compared to group C. There were no significant differences ($P \geq 0.05$) between groups regarding the staphylococci number. The colony forming units number of lactobacilli was significantly ($P \leq 0.05$) higher in the caecal content of broiler chicks fed with E diet (10.274 lg10 CFU/g wet caecal digesta) compared to that of broiler chicks fed with C diet (10.264 lg10 CFU / g wet caecal digest). In the broiler's caecal content of the two groups, *Salmonella spp*. was absent. The addition of 0.5 % willow bark extract in diets of broiler (14-28 days), reared under heat stress exerted a positive effect on the caecal microflora.

Keywords: Broiler, Heat stress, Microflora, Performance, Willow bark extract

Increasing dietary amino acid content in grower and finisher phases tends to increase the incidence rate of wooden breast in broiler chickens

Abstract ID: 407

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Wooden breast (WB) is an identified muscle disease, characterized by a hardening of breast muscle. The cause of WB is likely due to the high growth rate and breast meat yield of modern broiler chickens. The objective of the current study was to investigate the effect of dietary amino acid (AA) content on the occurrence of WB in broiler chickens. A total of 702 broiler chickens were used. The study was divided into four phases: starter (day 0-10), grower I (day 10-20), grower II (day 20-31) and finisher (day 31-42). A 2 x 3 x 2 factorial design was applied, with two breeds (Ross 308 and Cobb 500), three AA levels in the starter diet (low, medium or high), and two AA levels in the grower and finisher diets (medium or high). Digestible lysine content of the medium AA diet of each phase was 1.25, 1.15, 1.05 and 0.98 %, respectively. For the low and high AA diets, digestible lysine content was reduced and increased by 10 %. Average daily feed intake, average body weight gain and feed conversion ratio were determined for each period and over the experiment. At 42 days of age, all broiler chickens were dissected to determine the breast meat yield (%) and the score of WB. No two-way or three-way interactions were found for breast meat yield nor the occurrence of WB. Cobb 500 showed a higher breast meat yield than Ross 308 (30.1 vs. 29.0 %; $P < 0.001$). In addition, in the current study Cobb 500 showed a higher probability for a higher WB score than Ross 308 (0.58 vs. 0.38; $P < 0.001$). Decreasing or increasing AA content by 10 % in the starter diet did not increase either the breast meat yield or the incidence rate of WB. Increasing AA content by 10 % in the grower and finisher diets increased the breast meat yield (29.9 vs. 29.2 %; $P < 0.001$). Moreover, broiler chickens fed the high AA diet in the grower and finisher phases tended to show a higher probability for a higher WB score (0.51 vs. 0.44; $P = 0.07$). In conclusion, increasing dietary AA content in the grower and finisher phases tends to increase the incidence rate of WB in broiler chickens.

Keywords: Amino acid, Broiler chickens, Wooden breast

Influence of cereal type and fat source on performance, pellet quality and gastrointestinal traits in broiler starters fed pelleted diet

Abstract ID: 616

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This study was conducted to evaluate the effect of cereal type and fat source on performance, pellet quality, digesta transit time, viscosity of ileum and weight of gizzard and small intestine in broiler starters. Two cereal types (maize and wheat) and four fat sources: soy oil, fish oil, tallow and palm oil were evaluated in a 2 × 4 factorial arrangement of treatments. In maize-based diets, weight gain of birds fed soy oil was higher ($P<0.05$) than those fed fish and palm oil and similar ($P>0.05$) to those fed tallow, while, in wheat-based diets, inclusion of tallow increased ($P<0.05$) weight gain compared to other fat sources. Feeding maize-based diet increased ($P<0.001$) feed intake, also, feed intake ($P<0.01$) of broilers received tallow was higher than those fed other sources of fat. In maize-based diets, birds fed soy oil had better ($P<0.001$) feed per gain than those fed tallow, but was similar ($P<0.01$) to those fed fish and palm oil. However, in wheat-based diets, feeding tallow and soy oil improved feed to gain ($P<0.01$) compared to palm and fish oil diets in wheat diets. Pellet durability index (PDI) of soy and palm oil containing diets was better ($P<0.001$) than fish oil and tallow in maize-based diets, while lower ($P<0.001$) PDI was observed at fish oil diets ($P<0.001$) that based on wheat. Viscosity of ileum digesta was not influenced by fat sources in maize-based diets, but the lowest and highest ($P<0.001$) viscosity of ileum was observed at fish and soy oil, respectively, with those fed tallow and palm oil diets being intermediate in wheat based diets. In maize-based diets, relative weights of gizzard in birds fed soy oil was greater ($P<0.05$) than those fed tallow and palm oil diets. Although, in wheat-based diets, different fat source had no effect on relative weights of gizzard. While in maize-based diets, birds fed different fat source had similar ($P<0.001$) small intestine weights, in wheat-based diets, palm oil fed birds had heaviest small intestine that was significantly higher than tallow and fish oil fed birds ($P<0.01$). Overall, the present data suggest that fat source and main cereal of the diet affected weight gain, PDI and feed per unit gain of broiler starters fed pelleted diet and the observed effect differed depending on the main cereal used.

Keywords: Cereal type, Fat source, Pellet

Influence of different crude fiber contents in the diet on the performance of laying hens

Abstract ID: 19

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It is recommended to use in laying hen feed 40-50 g crude fiber per kg. The following study was carried out to verify the influence of increasing proportions of fiber 3/6/9 g/kg in diets on the performance of laying hens. 252 Lohmann Brown-Plus hens were randomly divided into 3 groups. The hens were kept in pens (21 hens per pen) with 4 pens per group. The study commenced when the hens were 21 weeks old and continued until the 6th laying month (168 days). In the treatment CF (crude fiber)/NDF (Neutral Detergent Fiber) concentration was increased per kg feed. Data were analyzed via ANOVA (SAS). Daily feed intake and feed conversion of hens from the 3% CF group were significantly higher compared with other groups. However, no effect of substituting CF through straw/oat in the diet was seen on laying intensity, egg weight and daily egg mass production. In the 3rd laying month the egg composition showed a significantly higher percentage of egg albumen and as a result a reduced part of egg yolk in highest CF group. In this trial a content of 66 and 84 g CF/kg feed significantly improved feed conversion. The highest content of CF/NDF per kg feed led to a heavier egg weight, though as a result of a greater albumen and a reduced yolk content.

Keywords: Crude fiber, Egg composition, Hens, Performance

Influence of different sources and length of fibers in the diet on the growth of broiler chickens

Abstract ID: 20

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It is recommended to use 35 g crude fibers per kg in broiler starter feed and with older broilers 40-45 g/kg. Information on recommendations for an optimal length of the fiber in broiler feed cannot be found. Therefore, the following study was carried out to investigate the influence of different sources and lengths of fibers in feed on growth performance of broiler chickens. 630 one-day old male chickens (ROSS) were randomly distributed into six treatment groups (two fiber origins – wheat/oat, 3 fiber lengths (SANACEL® – 30/200/400 or 300 µm) and one control group (9 pens/group) over a study period of 42 days. Data were analyzed via a two-way ANOVA. The results of this study indicate that an inclusion rate of different fiber lengths not significantly influenced feed intake, final body weight, carcass yield and proportional content of gizzard. However, the fiber length of 300/400 µm decreased the feed to gain ratio significantly. In this trial a fiber length of 300/400 µm with a fiber concentration of approximately 40 g/kg feed proved to be especially favorable for the development of the birds.

Keywords: Broiler, Crude fiber, Fiber length, Growing performance

Influence of graded dietary levels of seeds of three species of lupin on growth performance, nutrient digestibility, and excretion of total and free sialic acids of broiler chickens

Abstract ID: 107

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Study was conducted to investigate the effect of different dietary levels of seeds of three lupin species (narrow-leaved, yellow, and white) on the broilers growth performance, nutrient digestibility, excretion of total and free sialic acids and AMEN value of the diets. A total of 1280 one-day-old male broiler chicks of the Ross 308 strain were used in the experiment. A corn-soybean meal control diet and diets containing 50, 100, 150, 200, and 250 g/kg of three lupin species were fed to 10 replicate cages of 8 broiler chickens per treatment throughout the 35-day study. Fresh water and feed were provided ad libitum throughout the experiment. Titanium dioxide added to grower diets at 3.0 g/kg was served as an internal marker used to calculate the DM and nitrogen retention, apparent ileal digestibility (AID) and AMEN values, and to determine the content of total and free sialic acids. Growth performance of birds fed yellow and white lupin diets was similar to the control group. The use of narrow-leaved lupin decreased body weight gain and increased feed conversion ratio. Narrow-leaved lupin inclusion linearly and quadratically decreased the AID of dry matter (DM) and starch. Retention of DM and nitrogen, and AMEN values decreased (linear $P < 0.05$; quadratic $P < 0.05$), with increasing narrow-leaved lupin inclusion in the diet. There was a quadratic effect of narrow-leaved lupin level on the increase of sialic acid excretion. Excretion of sialic acid was negatively correlated with AMEN value and retention of DM and nitrogen. To conclude, yellow and white lupins could be alternative ingredients for soybean meal in broiler chicken diets without any adverse effect on growth performance. The narrow-leaved counterpart did not provide the satisfactory outcome.

Keywords: Broiler, Lubin, Performance, Sialic acid

Influence of particle size and fiber source on performance and nutrient utilization in broiler starters fed pelleted diets

Abstract ID: 621

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The study was conducted to evaluate the influence of particle size and insoluble fiber sources on growth performance and coefficient of apparent ileal digestibility (CAID) of nutrients in broiler starters fed pelleted diets. Two particle sizes (fine, 2 mm and coarse, 8 mm) and five sources of fiber (none, 1% cellulose, 3% rice hull, 3% sunflower hull and 3% oat hull) were evaluated in a 2 × 5 factorial arrangement of treatments. Each treatment was replicated 6 times. In fine grinding, inclusion of RH, OH and cellulose resulted in higher ($P < 0.05$) weight gain compared to control diet (with no additional fiber), whereas, in coarse grinding, all fiber sources improved weight gain from d 1 to 21 ($P > 0.01$). Fiber inclusion ($P < 0.01$) and fine grinding ($P < 0.01$) improved feed to gain ratio. Coarse grinding ($P < 0.01$) compared to fine grinding, also inclusion of RH, SFH and OH compared to control reduced pH of gizzard and proventriculus. Fat digestibility was not influenced ($P > 0.05$) by fiber sources in coarse ground diets, whereas, fiber inclusion increased ($P < 0.01$) CAID of fat compared to control diet in fine ground diets. Fiber inclusion resulted in higher calcium digestibility compared to control diet ($P<0.05$). In fine grinding, inclusion of RH and OH improved ileal protein digestibility, while, in coarse grinding, all fiber sources increased CAID of protein ($P<0.05$). Feeding RH, SFH and OH improved ($P < 0.05$) the CAID of phosphorus, also coarse grinding resulted in higher phosphorus digestibility than fine grinding ($P < 0.05$). In conclusion, inclusion of rice hulls, oat hulls and sunflower hulls, through improving feed to gain, reducing gizzard and proventriculus pH and improving nutrient digestibility affected growth performance of broilers fed pelleted diets.

Keywords: Broiler, Fiber source, Particle size, Pellet

Influence of Wheat Particle Size, Insoluble Fibre Sources and Whole Wheat Inclusion on Gizzard Musculature Development and Nutrient Utilisation in Broilers

Abstract ID: 221

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The influence of wheat particle size, insoluble dietary fibre and whole wheat (WW) inclusion on gizzard musculature, apparent metabolisable energy (AME) and ileal nutrient digestibility in broilers was examined in this study. The seven experimental treatments were fine (2 mm, control), medium (5 mm) and coarse (8 mm) wheat, control diet diluted with either 10 g/kg lignocellulose (LC), 30 g/kg oat hulls (OH) or 30 g/kg wood shavings (WS) and WW replacing the finely ground wheat during second and third weeks. A total of 350, one-d-old male broilers (Ross 308) were individually weighed and allocated to 35 cages (10 broilers/cage). Feeding OH and WS diets increased ($P < 0.05$) diameter of caudodorsal thick muscle and caudoventral thin muscle of the gizzard by an average of 84 and 50%, respectively. Diets containing fine wheat particles and LC resulted in the lowest ($P < 0.05$) and, OH and WS diets resulted in the greatest ($P < 0.05$) diameter of gizzard cranioventral thick muscle. Feeding medium wheat, coarse wheat, LC and WW diets resulted in similar AME to the fine wheat diet, but the AME was lower ($P < 0.05$) in OH and WS diets compared to the fine wheat diet. Feeding OH, WS and WW diets improved ($P < 0.05$) apparent ileal digestibility of starch, while medium wheat, coarse wheat and LC diets had no effect on starch digestibility. Overall, the present data indicate that gizzard musculature responses to structural components are likely influenced by the presence of coarse particles and, more importantly, the fibre composition. Incorporation of structural components to poultry diets can benefit digestive function, as a result of increased mechanical stimulation of proventriculus and gizzard.

Keywords: Broilers, Fibre Source, Particle size, Whole wheat

Investigation on growth performance and ileal digestibility of probiotics (Bacillus subtilis and Bacillus licheniformis) supplementation in broilers

Abstract ID: 18

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Alternatives to antibiotic growth promoters for broiler production are important in Thailand. This study was conducted to determine the effect of probiotics (*Bacillus subtilis* and *Bacillus licheniformis*) supplementation on growth performance and ileal digestibility in broilers. A total of 288, 1-d-old female Ross-308 broilers were randomly allotted to 4 treatments, each with 6 replicates. T1 was the control group receiving basal corn-soybean meal diet; T2 was supplemented with 200 ppm amoxicillin while T3 and T4 groups were supplemented with 2 levels of probiotic at 2.5 x 107 and 5 x 107 cfu/kg feed respectively. Growth performance parameters were calculated on d 21 and 42. Diets were added with 2% acid insoluble ash (AIA) as an indigestible marker on d 18-21 and d 39-42 for the determination of coefficient of apparent ileal digestibility (CAID). Broilers supplemented with either probiotic at both levels or antibiotic had a better ADG and FCR ($P < 0.05$) than the control group (d1-42). Mortality rates in the probiotic groups were lower than amoxicillin and control groups ($P = 0.053$). Broilers with the high level of probiotic inclusion (T4) improved ($P < 0.05$) CAID of dry matter (DM) and crude protein (CP) compared with the control group (T1) but did not differ from the amoxicillin group (T2) on d 42. In conclusion, the supplementation of both probiotic levels improved growth performances during the period of trial. Probiotic inclusion at the level of 5x107 cfu/kg feed could increase the ileal digestibility of DM and CP on d 42.

Keywords: Digestibility, Growth performance, Probiotics

Is pathogen induced anorexia in broilers infected with Eimeria maxima influenced by food quality?

Abstract ID: 557

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Pathogen induced anorexia (a voluntary reduction in feed intake; FI) is a common consequence of parasitic infections. We addressed the hypothesis that pathogen induced anorexia is independent of food quality. This was tested by reducing food quality through diet dilution with an inert, bulky ingredient (Arbocel cellulose) in both infected and non-infected birds. We hypothesised that diet dilution would reduce FI of non-infected birds. On the other hand, birds subjected to a coccidial infection and offered diluted diets would show a lower degree of pathogen induced anorexia (compared to infected birds offered non-diluted diets), as a further reduction in nutrient intake would not serve a benefit to the host. A total of 384 male Ross 308 day old broilers were distributed into 48 pens and offered a common starter diet for 6 days. A gradual diet dilution was implemented from D7 until D9, and birds were offered the full dietary treatment on D10 (0, 5, 10, 15%; No Fibre, Low Fibre, Medium Fibre, High Fibre; NF, LF, MF and HF, respectively). At D13 (D0pi; post infection) half of the pens were infected with 7,000 sporulated *E. maxima* oocysts.
FI was measured daily until D15 pi and faeces were collected from D4pi until D10pi to measure oocysts per gram (OPG). We observed a significant interaction between diet and infection on D6pi ($P = 0.006$) and D7pi ($P <0.001$), when the anorexia was at its maximum. In the absence of infection, diet dilution resulted in the expected dose dependent reduction in FI. However, the opposite trend was seen in the presence of infection. On D6pi and D7pi HF infected birds ate significantly more than the NF infected birds ($P <0.001$). However, there were no significant differences in OPG excretion between the different dietary treatments from D5-D9pi ($P = 0.997$). In conclusion, our results suggest that infected birds fed a diluted diet did not reduce their FI to the same extent as birds fed a non-diluted diet, as if birds were trying to regulate their energy or nutrient intake. Therefore, pathogen induced anorexia is dependent on food quality and should be taken into account when treating infected birds.

Keywords: Anorexia, Coccidiosis, Diet Dilution, Feed intake

Larvae of synanthropic flies in diets for broilers

Abstract ID: 596

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The aim of the study was the evaluation of effects of native larvae of synanthropic flies as a substitute for fishmeal in diets for broilers on growth and feed efficiency. Nutrient contents (%) in 100 g of air-dried larvae: crude protein 53.7, crude fat 24.8, crude fiber 5.4, calcium 0.3, phosphorus 1.5, sodium 0.6, lysine 3.4, methionine 1.5, cystine 0.5; ME 10.48 MJ/kg.
The study was performed on 4 treatments of Cobb-500 broilers (35 birds per treatment) reared from 1 to 38 days of age. Control treatment 1 was fed standard broiler diets containing 6 and 4% of fishmeal according to age periods 1-4 and 5-6 weeks of age. In diets for treatments 2 and 3 blanched larvae substituted for 50 and 100% of fishmeal, respectively; in treatment 4 minced larvae substituted for 100% of fishmeal. All diets were isonitrous and isocaloric and complied with breeder's recommendations.
Mortality in treatment 2 was 0%, in other treatments 2.9%. Live bodyweight in all experimental treatments was significantly higher in compare to control; in treatment 2 by 5.85% ($P<0.05$), in treatment 3 by 12.7% ($P<0.001$), in treatment 4 by 6.5% ($P<0.05$). Feed conversion ratio in experimental treatments 2-4 was lower in compare to control by 4.6; 5.2 and 8.1%, respectively, due to improvements in digestibility of dietary nutrients. Digestibility of protein in treatments 2-4 was higher in compare to control by 1.6; 0.7 and 2.1%; fat by 5.0; 2.2 and 6.5%; fiber by 5.4; 3.2 and 6.3%, respectively. Nitrogen assimilation was higher by 3.7; 1.4 and 5.4%, amino acid assimilation higher by 4.2; 2.4 and 4.7%. Concentration of total free amino acids in blood serum was higher in treatments 2-4 by 2.3; 4.7 and 2.5%, respectively, in compare to control treatment 1. Eviscerated carcass yield was higher in all experimental treatments (average 68.6% vs. 68.1% in control). Taste panel trial evidenced excellent sensory characteristics of bouillon and meat of larvae-fed broilers.
It was concluded that blanched or minced larvae of synanthropic flies can be effectively included to broiler diets as a protein source and a substitute for expensive fishmeal.

Keywords: Broilers, Fishmeal, Growth efficiency., Larvae of synanthropic flies, Protein

Lupine in Diets for Laying Hens

Abstract ID: 118

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The modern cultivars of lupine were studied as a possible substitution of animal-derived protein sources, soybean meal and sunflower cake in diets for hens. It was found that the dehulling of lupine allows the production of plant protein source with protein content no less than 40% and fiber content 3.5%. The trial was performed on 5 treatments of cage-housed SP-789 layers (30 birds per treatment; KBN cage batteries) fed diets with 0, 5, 7, 10 and 15% of dehulled and toasted lipine (Dega cultivar, alkaloid content 0.025%) during 6 months of the productive season. At the age of peak egg production and at the end of the trial (35 and 58 weeks of age) the layers were inseminated and fertile eggs were obtained with subsequent incubation (Danki incubator with standard regime; 100 eggs per treatment). Artificial insemination was performed according to the standard protocol with the use of new patented medium for sperm dilution (1:3); single dose of diluted sperm was 0.1 ml per hen. During 1-18 days of incubation the temperature was set at 37.70C and controlled with 0.10C accuracy, since day 19 the temperature was set at 37.20C; relative humidity levels were 52-53 and 52-75%, respectively. Fertility of eggs laid at 35 weeks of age in lupine treatments was 96-100% vs. 96% in control; hatchability was 87.88-94.00 vs. 84.38%. The intensity of lay at this age in four lupine treatments was 79.33; 83.44; 83.67 and 82.00% vs. 80.11% in control. At 58 weeks of age egg fertility in lupine treatments was 100, 99, 95 and 96% vs. 94% in control; hatchability 89.00; 88.89; 91.58 and 92.71% s. 86.17% in control. The intensity of lay at this age in four lupine treatments was 76.67; 79.68; 83.66 and 79.46% vs. 75.86% in control. The results of the trial evidenced that low-alkaloid white lupine can be effectively used in diets for parental flocks of chicken at inclusion levels up to 15%.

Keywords: Egg hatchability, Feed efficiency, Laying hens, Lupine, Mortality

Lysine and protein requirements of laying Japanese quail

Abstract ID: 402

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For laying hens, crude protein (CP) and digestible lysine (DL) requirements affect production parameters. These requirements have already been deeply studied and are very well known. For laying quails, they need to be updated and better evaluated. Our first approach of this study was to compare amino acid profiles of eggs coming either from laying hens or laying quail in order to deduce the different targets in amino acid profiles between the two species. Then, the aim of this trial was to test different CP and DL levels during quails laying period (from 9 week old to 20 week old). A total of 2143 Japanese quails were randomly assigned to 5 treatments, each consisting in 20 cages as replicates (24 quails per cage). Quails received one out of the 5 experimental diets varying in their CP content (-1.5%, -3%), from 20% to 17%, and in their DL content (-0.1%, -0.2%), from 0.95% to 0.75%. The experimental diets were iso-nutrients for all the other parameters. Feed intake, egg production (EP), egg weight, egg yield (EY) and feed conversion ratio (FCR) were recorded weekly. Differences were studied by one-way ANOVA with repeated measures, followed by a Bonferroni's post hoc test ($P<0.05$). At 20% of CP, EP was not affected by the different DL levels. FCR significantly increased by lowering dietary DL level from 0.95% to 0.75% (-0.04 point; $P<0.001$). At 0.75% of DL, by reducing the CP content of 1.5 points from 20% to 18.5% did not shown any detrimental effect on performance while at 17% of CP, it significantly ($P<0.001$) decrease EY (-0.4g/day), and increase FCR (+0.08 point) compared to 20% of CP. Decreasing DL from 0.95% to 0.75% showed no effect on EP. Dietary DL at 0.75% seems to be enough to reach and even exceed the laying quail requirements for this nutrient. Based on these results, dietary CP could be reduced from 20% to 18.5% without any detrimental impact on production performance. The DL requirements of laying quail could be fixed between 20.6 and 20.8 mg/g of egg.

Keywords: Crude protein requirement, Egg production, Japanese quail, Lysine requirement

Manipulating energy density and dietary electrolyte balance in ISA Brown laying hen diets diluted with Kikuyu grass (Pennisetum clandestinum)

Abstract ID: 486

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Pasture intake as a proportion of the diet of free-range layers is a major factor influencing the difference in performance between conventional cage and free range systems. The objective of this study was to investigate in a 2 x 2 factorial design the effect of feeding two levels of dietary electrolyte balance (150 mEq/kg and 250 mEq/kg) and two dietary energy density (standard at 11.73 MJ/kg AME and high density at 12.15 MJ/kg AME) when diluted with 7.5 % of fresh Kikuyu (*P.clandestinum*) grass, on performance, egg quality traits, blood parameters and nutrient digestibility in laying hens over 42 days. A total of 144 ISA Brown laying hens at 32 weeks of age were used in the experiment with twelve replications of three birds each for each treatment diet. Although high DEB diet depressed egg production rate by 4% ($P \le 0.01$) and showed a trend towards lowered feed intake ($P=0.090$), significant positive effects were observed on egg quality with enhanced egg shell thickness ($P \le 0.01$). High density diets showed a significant improvement in percentage yolk ($P \le 0.05$) and albumen weight ($P \le 0.05$). Higher density diet also showed a significant increase in higher apparent metabolisable energy (AME) ($P \le 0.01$). Higher DEB diets significantly improved dry matter (DM) digestibility ($P \le 0.05$), and AME ($P \le 0.05$) . The results of this study show that both a high energy density diet and high DEB (250 mEq/kg) diet had a significant effect on the performance of laying hens fed diets diluted with 7.5% Kikuyu grass.

Keywords: Dietary electrolyte balance, Dietary energy, Free range, Grass, Laying hens

Manipulation of Meat Quality of The Late Period of Laying Hens through Supplementation Probiotic, Metabolizable Energy and Digestible Essential Amino Acids

Abstract ID: 70

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This study was conducted to evaluate the effect of probiotic, dietary metabolizable energy (ME) and digestible essential amino acids on meat quality of the late period of laying hens. A 2x2x3 factorial arrangement of treatment comprising of 2 dietary ME levels (2,800 and 2,900 kcal of ME/kg feed) and 2 levels of digestible lysine-methionine ratio (DLM); (0.81:0.44 and 0.97:0.53%) with supplementation of commercial probiotic at the level of 0, 0.1 and 0.2% diet. Isa brown laying hens (n=540, 60 wks old) were randomly divided into 12 treatments (5 replicates of 45 hens per treatment). The trial lasted 16 wks. There was no interaction on meat quality between probiotic, ME and DLM. However, it was also found that there was a significant ($P<0.05$) improved on meat quality in term of abdominal fat (AF), water holding capacity (WHC), cooking loss (CL) and shear force (SE) when increased the levels of ME and DLM while supplementation of probiotic had no effect ($P>0.05$) on meat quality. In conclusion, diet which composed of 2,900 kcal ME/kg feed and 0.97:0.53 % DLM is the optimum level to improve meat quality of the late period of laying hens.

Keywords: Digestible essential amino acids, Late period of laying hens, Meat quality, Metabolizable energy, Probiotic

Meat quality of slow-growing chickens housed on pasture

Abstract ID: 110

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The aim of this study was to evaluate the effect of pasture intake in 300 broilers of three chicken genotypes (Dominant Sussex D104, Dominant Brown D102 and Dominant Tinted D723) on the concentration of carotenoids and vitamins, oxidative stability and sensory evaluation of breast muscle. The chickens were kept until 49 days of age in indoor pens and then they were moved to the mobile boxes on pasture from 50th to 77th day of age (stocking density 9.2 chickens per m^2). The mobile boxes were moved twice a day to restrict grassland damage. The dominant species in the pasture were Lolium perenne, Festuca pratensis and Trifolium pratense. Higher pasture intake was in 70 days of age ascertained in genotype Dominant Brown D102 (33.8 g/pcs/day) and Dominant Tinted D723 (34.4 g/pcs/day). It corresponds with higher concentration of lutein ($P \leq 0.003$; 0.234 mg/kg) and zeaxanthin ($P \leq 0.002$; 0.223 mg/kg) in breast muscle of Dominant Tinted D723. The highest concentration of α-tocopherol in breast muscle ($P<0.001$) was ascertained in Dominant Brown D102 (4.572 mg/kg DM) and Dominant Tinted (4.642 mg/kg DM) in comparison to Dominant Sussex D104 (3.435 mg/kg DM). The significant differences in concentration of retinol were not found. Higher oxidative stability ($P \leq 0.012$) of fresh breast muscle was recorded in Dominant Brown D102 and Dominant Tinted D723. However after 5 days of storage, significant differences were not detected. In addition, the meat of cockerels Dominant Tinted D723 showed the highest tenderness based on the sensory evaluation ($P \leq 0.022$). Results of the study suggest that Dominant Tinted D723 had the highest pasture intake which positive influenced some quality characteristics of meat.

Keywords: Broiler, Carotenoids, Genotype, Meat quality, Pasture

Mineral composition of eggs and vitelin sac of day-old chicks from broiler breeders in different ages

Abstract ID: 213

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Maternal nutrition is an important key to maintain the development of embryos and neonate chicks. Diet composition affects egg content which is transferred to vitelline sac to guarantee good starter and growing development in broiler chickens. Minerals are enrolled in important physiological and immune functions that are essential for breeders and chicks and the egg is the unique way to maintain normal embryo and neonate metabolic functions. Then, is important to understand the transference of nutrients from breeders to embryos and consequently to neonate chicks. Based in this findings, the objective of this study was to evaluate the egg yolk and vitelline sac mineral composition from broiler breeders in three different stages of production cycle. The birds were evaluated during the initial egg production (32 weeks), peak egg production (42 weeks) and after peak production (52 weeks), checking if the Hubbard broiler breeders age can interfere on the mineral egg content with the advancing age. Three experiments were conducted in commercial company and at the Poultry Sector from Department of Animal Production at the Veterinary Medicine and Animal Science Faculty, Federal University of Goiás in Goiania, Brazil. A group of 60 samples of diets, yolks and vitelline sac were selected to chemical analysis. The yolks and vitelline sac samples were separated and frozen at - 18°C and then lyophilized. Analysis were performed for Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg), Copper (Cu), Iron (Fe), Manganese (Mn) and Zinc (Zn) using atomic absorption spectrometry. It was found that the age of breeders interfered on the yolk and vitelline sac composition. In yolk, P, K, Mg, Cu, Mn and Zn were reduced in eggs from 52-week age hens, and increased for Mg, with no effect observed for Ca and Fe. In vitellin sac, P levels were increased for 52-week age hens and reduced for Ca in 42 and 52-week of age hens, with no effect on K, Mg, Cu, Fe, Mn and Zn. This results indicated that the nutritional content of minerals for embryos and neonate chicks were affected, which indicates that as old the breeders are, minerals availability should reduce for starter chick development.

Keywords: Diets, Mineral content, Neonate chick, Vitellin sac, Yolk composition

Molecular modulation by GOS prebiotic injected in ovo in broiler chickens under heat-stress

Abstract ID: 451

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Galactooligosaccharides (GOS) prebiotics delivered *in ovo* stimulate development of indigenous microflora in the chicken embryo. Such stimulation creates eubiotic environment in the guts which positively influences intestinal function and health. This study aimed to evaluate the impact of a GOS delivered *in ovo* on the modulation of the transcriptomic responses in intestinal tissues of broiler chickens challenged with heat. On d 12 of egg incubation, a single dose of physiological saline (CON) or CON+3.5 mg GOS/egg (GOS) was injected into the air chamber. A total of 300 male chicks/group were split into 2 groups and reared in floor pens (6 replicates/group/environmental condition) either in thermoneutral (TN, 25°C) or chronic heat-stress (HS, 30°C for 24h/d from 32 to 42 d) conditions. At slaughter (day 42) jejunum and cecum mucosae were collected from 2 birds/replicate. RNA was extracted for microarray analysis on transcriptome by GeneChip Chicken Gene 1.0ST Array. Data were analyzed by Transcriptomic Analysis Console software (Affymetrix). Transcripts were considered as differentially expressed genes (DEG) when showing
a ≥ 2-Fold Change (log2 ratio) and a False Discovery Rate<0.05 between treatments. Pathway analysis was carried out using Gene Set Enrichment Analysis (GSEA) and a KEGG list of gene sets (from MSigDB, Broadinstitute).
No interaction between HS and in GOS treatments was detected in jejunum and cecum for the DEG. No DEG were detected for GOS vs. CON treatment in either tissue. Conversely, HS vs. TN up- and down-regulated 12 and 13 genes in jejunum and 2 and 9 genes in cecum, respectively. Significantly enriched gene sets were observed in jejunum for lipid metabolism and oxidation (i.e. peroxisome; sphingolipid metabolism; cytochrome p450 metabolism) in GOS, probably due to the prebiotic effect, and for cell cycle and mitosis in CON. No enriched gene set was found in cecum. GSEA confirmed a negative effect of HS on the gut health showing enrichment in gene sets such as for cytochrome p450 metabolism, linked to oxidative metabolism, and for retinol metabolism (possibly linked to retinol involvement in oxidative stress preventing) in jejunum, while gene sets for genes involved in extracellular matrix (ECM) organization, collagen formation and for ECM glycoproteins encoding were found in cecum, likely due to oxidative stress again. The ongoing analyses on gut metagenomic sequencing and on immune response will further help in elucidating GOS impact in heat-stressed and unstressed chicks.

Keywords: Heat-stress, In ovo injection, Poultry, Prebiotic, Transcriptome

Mycotoxin Survey 2017 - The threat in Europe

Abstract ID: 399

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Poultry are particularly sensitive to mycotoxins. Therefore, it is imperative to monitor the prevalence of mycotoxins in agricultural commodities and the severity of mycotoxin contamination worldwide. To elucidate the threat of these toxic metabolites, BIOMIN has been conducting an annual mycotoxin survey for almost fifteen years, monitoring the incidences of different mycotoxins in finished feed and agriculture commodities used in animal feed.
In 2017, 18,757 samples sourced worldwide were analyzed for major mycotoxins, including aflatoxins (Afla), zearalenone (ZEN), deoxynivalenol (DON), T-2 toxin (T-2), fumonisins (FUM) and ochratoxin A (OTA). Of these samples, 4,812 originated from Europe. Samples were analyzed using liquid chromatography coupled to tandem mass spectrometry, high performance liquid chromatography or enzyme-linked immunosorbent assay.
In total, 90% of samples from Europe contained at least one of the six main mycotoxins (based on samples for which at least three mycotoxins were analyzed). The *Fusarium* mycotoxin DON was the most common mycotoxin and was detected in 65% of all samples at a mean concentration of 555 ppb. FUM was present in 51% (mean of 777 ppb), ZEN in 44% (mean of 72 ppb) and T-2 in 33% (mean of 36 ppb) of samples. Aflatoxin (produced by *Aspergillus* species) and ochratoxin A (produced by *Penicillium* and *Aspergillus* species) occurrence are often related to storage conditions but also to grain damage in the field. Afla was present in 16% (mean of 6 ppb) and OTA in 24% (mean of 8 ppb) of samples.
Trends of mycotoxin occurrence in single commodities (corn, finished feed, etc.) were similar to the trend observed for the whole dataset. In corn samples, for example, DON and FUM were the most common mycotoxins, detected in 72% (mean of 882 ppb) and 73% (mean of 1506 ppb) of samples, respectively Furthermore, prevalence of ZEN (40% positive tested samples), T-2 (22% positives) and Afla (22% positives) was rather high in European corn.
The survey results indicate that mycotoxins remain a serious concern in agricultural production. An effective mycotoxin risk management program should be applied to protect poultry species from negative effects of mycotoxins.

Keywords: Europe, Mycotoxins, Survey

Ontogeny of feather pecking behaviour with focus on early nutrition

Abstract ID: 232

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Feather pecking (FP) by laying hens is a significant welfare issue in the poultry industry. Pecking at and pulling out the feathers of conspecifics can seriously reduce the well-being of hens, and cause economic losses for the farmer. It is important to better understand the origin of this unwanted behaviour, which eventually might lead to preventive measurements. The ontogeny of FP behaviour remains to be unravelled: it has been suggested to be different forms of re-directed behaviour (e.g. foraging, ground pecking and dustbathing), and is also associated with social exploration, frustration and feather eating. Combined with nutrition, several options are possible. Hens might re-direct their behaviour because there is no proper foraging material or they are not satiated. If hens are unable to fulfil their drive for feeding related behaviour, they may get frustrated and consequently peck their conspecifics. Powerful behaviour-related hormones, such as serotonin and dopamine are related to FP, but the precise mechanisms are still unknown. Unpublished work of our group revealed some effects of a 10% decreased energy level in the diet on serotonin and dopamine turnover (P=0.058 and P=0.014, resp.) in the brain of laying hens. These results support the hypothesis that endocrine signals of stress and behaviour in blood and the brain could be related to nutritional factors. Previous findings also showed that nutritional interventions, such as diet structure and fibre content, reduced FP and cannibalism. Other studies indicate a relationship between early life nutrition and later life pecking behaviour. Nutrients can act as precursors to behaviour related hormones and satiety related gut hormones (e.g. PYY, ghrelin, CCK), that may influence eating time and pecking related behaviour. It is hypothesized that nutritional strategies during early stages of rearing could influence pullets in such a way that FP during the laying period can be reduced or even prevented. A series of experiments will be conducted aiming to pinpoint this precise period of sensitive during the rearing of pullets and reveal the relationship between nutrition and FP behaviour. Additionally, the effects of nutritional intervention during these sensitive periods on FP in later life will be studied.

Keywords: Behaviour, Feather pecking, Laying hens, Nutrition, Rearing

Optimum digestible valine to digestible lysine ratio for growth rate and feed efficiency of broiler chickens

Abstract ID: 327

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Steady concerns to protect the environment has led the nutritionists to reduce the crude protein level in farm animal diets, as 1% reduction in CP will lead to 10% reduction in the N excretion in broilers (Gomide et al. 2011). However, the reduction in CP urges to the supplementation of the synthetic amino acids in order to meet the animal maintenance and production requirements. Since the trend of supplementation of L-Valine increased significantly in the last years, the present study was designed to determine the valine to lysine ratio of the modern broiler chickens. A total of 960 male Cobb 500 male broilers were housed in 48 floor pens (20 birds per pen). The basal diet consisted of corn, wheat, SBM and peas. The experiment comprised in total 8 dietary treatments based on supplementation of L-Valine at 6 graded levels to a basal diet (= negative control: NC) with a low d.Val content and a low d.Lys content (-7.5%) from 0-35 days. The NC diet contained a d.Val: d.Lys ratio of 0.63 and the other diets contained a ratio of 0.68, 0.73, 0.78, 0.83, 0.88 and 0.93. A positive control (PC) diet was also formulated to contain a d.Val: d.Lys ratio of 0.80 with a normal d.Lys level. Body weight gain (BWG) and feed conversion ratio (FCR) were measured from d0-35. The raw data were analyzed through ANOVA, whereas, different regression methods were applied to identify the optimal d.Val: d.Lys ratio, determined by establishing the first point at which the quadratic polynomial response curve intersected the plateau value established from the linear response plateau model. Birds responded significantly ($P<0.05$) to the supplementation of L-Valine to the basal diet. Based on the ANOVA the maximum response for BWG (2,404 g) and FCR (1.445 g/g) was achieved at a d.Val: d.Lys ratio of 0.78. When the data was evaluated through the regression analysis, the optimum dose-response relationship of d.Val: d.Lys ratio to ensure the best BWG and FCR was 0.76 and 0.78 respectively, whereas based on laboratory analysis the optimum total Val: Lys ratio was 0.79 (BWG) and 0.81 (FCR) in male Cobb 500 broilers from 0-35 days of age.

Keywords: Broilers, Cobb, Ideal ratio, Valine

Performance, Immune Related Organs Weight, and Blood Metabolites of Broiler Chicks Fed Diets Included Immune Responses Enhancers

Abstract ID: 182

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A study was conducted to compare the effects of a commercial immune responses stimulator (Bio-Herbal) with two products (A and B) on growth performance, immune related organs weight and blood metabolites of broiler chicks. Product A (IASRI1) is an immune responses stimulator based on vitamin C, vitamin E, Selenium, Garli (*Allium Sativum*) powder, Paper (*Piper nigrum*) powder, Lemon (*Citrus Limon*), and Mentha (*Mentha Piperita*) essences. Product B (IASRI2) is too an immune responses stimulator based on vitamin C, vitamin E, Selenium, Onion (*Allium Cepa*) powder, Lemon (*Citrus Limon*), Rosemary (*Rosmarinus Officinalis*) and *Eucalyptus* essences.This experiment was conducted in a completely randomized design with 4 treatment, 8 replication (except control group that had 4 replication) and 15 day old chicks (Ross 308) per each replicate. Experimental treatments were: (1) The basal diet (control); (2) the basal diet + 200g/ton Bio-Herbal; (3) The basal diet + 1 kg/ton IASRI1; (4) The basal diet+ 1kg/ton IASRI2. Daily feed intake, daily weight gain, feed conversion ratio and mortality percentage were evaluated in starter (1-14 days), grower (15-28days), finisher (29-42 days) and total (1-42 days) periods. Immune responses of broiler chicks (SRBC, IgG, IgM, PHA AND DNCB) were studied in 35 days of age. At the end of experiment carcass characteristics and related organ weight and blood biochemical metabolites were studied. Experimental treatments had significant effect ($p<0.05$) only on mortality percentage in starter period. Mortality percentage in treatments 2 and 3 were less than control group (1.66 and 1.72 vs. 10.23 %). Experimental treatments had no significant effect on related organs weight (Liver, Boursa, Spleen, and Gizzard). Experimental treatments had no significant effect on immune responses (SRBC,IgG, IgM, PHA, DNCB). There were significant difference on HDL, LDL/HDL and CHO/HDL between experimental treatments. HDL content in treatments 2 and 3 were less than control group. LDL/HDL ratio in treatment 3 were higher than other treatments. According to the results of this experiment immune responses stimulator A (ASRI1) were better than Bio-Herbal and immune responses stimulator B (ASRI2).

Keywords: Body organ weight, Broiler, Immune response, Performance

Performance of Bacillus probiotics is strain specific

Abstract ID: 514

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Gut health is a major factor to be taken into consideration for the optimum performance of birds. It is becoming increasingly clear that the intestinal microbiota mediates key physiological processes thereby influencing the host. Probiotics can positively impact these processes, however their effects are strain dependent. In the present study, we describe the screening for a novel strain of Bacillus subtilis which was phylogenetically different from commonly applied Bacillus strains. It was found to have specific impact on animal performance.
A novel strain of Bacillus subtilis was identified by in vitro screening of safety, performance and robustness and by comparisons of whole genome sequences.To study the phylogeny of the selected strain *(B. subtilis* DSM29784), a comparative analysis of the *gyrB* gene sequences was performed.
Animal trials were conducted in three trial facilities to assess the impact of strain DSM29784 on broiler performance. Broilers were fed 1E8CFU/kg feed and feed conversion was measured at day 35.
Screening a range of Bacillus strains revealed large differences in strain characteristics even between closely related strains. The *gyrB* gene analysis suggested that DSM29784 is a novel subspecies of *B. subtilis*.
In vivo results showed a significant improvement in animal performance in all three animal studies when compared to negative controls with an average FCR improvement of 3.8% at day 35 ($p<0.05$). In comparison other Bacillus strains showed less consistency in results.
This novel strain of Bacillus was found in an extensive screening and tested in animal studies. The results of these in vivo tests showed significant improvement in animal performance which was found to be linked to the strain's impact on the host microbiota and host epithelial wall. These data support that the effects of probiotic bacteria are strain specific and that a probiotic strain with certain specific properties can benefit the health and growth of commercial broilers.

Keywords: Bacillus, Nutrition, Probiotics

Performance of layers and egg quality traits as affected by the dietary supplementation of layers' diet with Spirulina sp.

Abstract ID: 487

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The aim of this study was to evaluate the effect of dietary supplementation of layers' diet with *Spirulina* sp. on the performance of laying hens and egg quality traits, as well as on fatty acid profile and oxidative stability of egg yolk. One hundred and ninety-two, 60 weeks old, ISA Brown laying hens, were randomly allocated into 2 dietary treatments (C, S). Each treatment was comprised of 8 replicates (cages) and each replicate was comprised of 12 layers. During the experimental period (45 days) the layers of treatment C (control diet) fed on a typical diet for laying hens based on corn and soybean meal whereas those of treatment S fed on the same diet supplemented with *Spirulina* sp. so that it corresponds to consumption of 1g of spirulina/hen/day. Egg production was daily recorded. Eggs were taken every 15 days from the beginning of the experiment and for three consecutive days. A total of 144 eggs were examined to determine the fatty acid profile and oxidative stability of egg yolk while another 64 eggs were examined for their quality characteristics. Results showed no significant differences ($P\geq0.05$) between treatments in egg, albumen and yolk weight and height, in egg and yolk diameter, as well as in shell hardness, except for the yolk color ($C=7.84\pm0.88$ *vs* $S=8.59\pm0.71$, $P\leq0.05$). The fatty acid profile showed that total saturated, unsaturated, monounsaturated and polyunsaturated fatty acids, n-3 and n-6 fatty acids, as well as, n-3:n-6 ratio were not significantly ($P\geq0.05$) affected by the treatments. Egg production rate was calculated at 82.96% for the control group and 79.92% for the spirulina group, but difference between groups was not significant ($P\geq0.05$). The oxidative stability data are currently under statistical processing and results will be published soon.

Keywords: Egg quality, Layers, Performance, Spirulina diet

Phytase storage stability under different trace mineral premixes

Abstract ID: 470

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Phytase inclusion to monogastric diets is commonplace in the industry today. Its ability to release phytate-bound phosphorus (P) from cereal based feed ingredients aims to satisfy the animal's P requirements, while simultaneously reducing inorganic P inputs at formulation. Given the benefits of exogenous phytase, and in appreciation of the complexity of modern day feed formulations, it has become increasingly necessary to identify and avoid any potential negative ingredient interactions which may impede enzyme function.
The present study assessed the antagonistic effects of dietary trace minerals on phytase activity under simulated storage conditions with a variety of mineral premix packs. Inorganic trace minerals (ITMs) are often supplemented to monogastric feeds at levels exceeding those recommended by the National Research Council (NRC). Comparatively, organic trace minerals (OTMs), such as Bioplex® chelates, can be used at lower inclusion rates due to their greater bioavailability, with partial inclusion rates typically ranging between 25-50% of NRC values.
Results indicated that, during the storage of a commercial phytase in different premix packs over a 7-week period, mineral source and composition had a significant effect on phytase activity ($p \leq 0.05$). The most notable loss of enzyme function occurred with an inorganic mineral premix formulated to NRC specifications, retaining only 27.9% phytase activity after 7 weeks. Comparatively, a commercial inorganic-based mineral premix demonstrated approximately 70-75% activity over the course of the study. Two commercial organic mineral premixes were also assessed, one based on OTMs as Bioplex® chelates and the other containing amino acid complexes. The amino acid complex based premix had a final activity similar to the inorganic premix, with 71.7% phytase activity recovered. Conversely, the chelate-based premix demonstrated the greatest level of enzyme function, retaining 87.4% phytase activity after 7 weeks of storage.
This study emphasises the potential antagonisms that may occur between feed components over prolonged storage times, as well as the importance of mineral source in retaining enzyme function and overall feed ration value.

Keywords: Inorganic, Mineral, Organic, Phytase, Stability

Processed animal proteins can replace soybean meal in broiler diets

Abstract ID: 114

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Abstract Processed animal proteins (PAPs) have long been banned from farm animal diets in the EU, and nutritional values in present feeding tables may not be representative for present PAPs. Two experiments were conducted to determine i) nutrient digestibility and AMEn of two different porcine protein meals in broilers, and ii) the effect of replacing soybean meal (SBM) by these PAPs on growth performance, gut health, bone quality and blood characteristics related to Ca and P metabolism. PAP1 (62% crude protein, 15% fat, 18 g/kg Ca, 15 g/kg P) was derived after sterilisation under pressure (EU-method 1) and PAP2 (62% crude protein, 11% fat, 84 g/kg Ca, 46 g/kg P) after applying EU-method 7, which is less specifically defined. In the digestibility study, three treatments (basal diet, basal+18% PAP1, basal+7% PAP2) were replicated in six pens with 12 Ross_308 male broilers from 14 – 23d per pen. The performance study comprised three treatments with starter, grower, and finisher diets without PAPs (control), with 8.0, 8.5, and 9.0% PAP1, and with 8.0, 7.0, and6.0% PAP2, respectively. Each treatment was replicated in eight pens with 10 Ross 308 male broilers from 0 – 42d per pen. PAP1 and PAP2 replaced 4.5 to 8.5% and 5.5 to 7.0% dietary SBM, respectively. PAP2 had a higher precaecal digestibility of dry matter (76.6 vs. 60.9%) and crude protein (68.8 vs. 60.9%), but lower digestibility of calcium (40.3 vs. 48.8%) and phosphorus (49.9 vs. 56.4%) compared to PAP1. PAP2 had a higher total tract digestibility of dry matter (65.6 vs. 54.5%), organic matter (67.1 vs. 55.0%) and ash (52.2 vs. 42.3%), but a lower digestibility of crude fat (47.2 vs. 79.6%). AMEn content of PAP1 and PAP2 was 11.6 and 9.3 MJ/kg, respectively. Feed intake and BWG of PAP1 fed birds in the performance study was higher than in control or PAP2 fed birds. FCR was higher in both PAP treatments, but corrected to a similar BW of 3850 g, FCR was not affected by dietary treatments. Apart from a higher villus/crypt ratio in the PAP1 fed birds, none of the gut morphology parameters were affected by treatments. Blood serum parameters, footpad and litter scores, and tibia quality parameters were also not affected by dietary treatments. The current study provided up to date nutrient characteristics of the tested PAPs, while performance data showed that these PAPs can partly replace SBM without compromising growth performance, gut health, or bone quality.

Keywords: Broilers, Digestibility, Growth performance, Processed animal proteins, Soybean meal

Profile of fatty acids of yolk sacs of embryos from broiler breeders supplemented with CLA

Abstract ID: 222

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CLA has been associated with improvement of immunity and performance of broilers; however, it may change the fatty acid profile of yolks when offered to broiler breeders. We aimed to evaluate the fatty acid profile of yolk sacs (YS) of chick embryos from broiler breeders fed CLA. Two breeder flocks fed control or CLA-supplemented (0.025%) diets were raised in a completely randomized design. At the end of supplementation, 270 eggs per treatment were distributed in two hatches. The fatty acid profile of the yolk sacs of 18 days old embryos were evaluated, applying the randomized block design, and each block consisted of each age which corresponds to different hatches. Data were submitted to ANOVA and F test to compare the means at 5% probability. Yolk sacs of embryos from supplemented breeders showed higher percentages of myristic ($P=0.008$), palmitic ($P<0.001$), palmitoleic ($P<0.001$) and stearic ($P<0.001$) acids, higher values for saturated fats ($P<0.001$) and total fat ($P=0.014$), and higher saturated:unsaturated fatty acids ratio ($P<0.001$). The yolk sac of control embryos showed higher omega-3 ($P<0.001$) and omega-9 ($P=0.003$) composition, and higher percentages of monounsaturated ($P=0.009$), polyunsaturated ($P=0.016$), oleic ($P=0.003$), cis-11-eicosenoic ($P=0.013$), cis-11,14-eicosadienoic ($P=0.013$), arachidonic acid ($P=0.002$) and docosahexaenoic ($P<0.001$) fatty acids. Embryos from supplemented breeders absorbed higher amounts of polyunsaturated fatty acids, indicated by the lower percentage of these in the yolk sac, suggesting a greater incorporation of polyunsaturated fatty acids into the organism. On the other hand, the use of CLA for breeders resulted in higher percentages of saturated fats in yolk sacs, suggesting lower demand for energy sources and, probably, higher efficiency of their use during embryonic life. It is important to notice that docosahexaenoic acid, essential for brain function, decreased in yolk sacs of embryos from supplemented breeders, indicating greater uptake by the embryo. In conclusion, CLA modifies the fatty acid profile and increases the saturated: unsaturated fatty acids ratio in the content of yolk sacs of 18 days old embryos.

Keywords: Chick embryo development, Omega-6, Saturated:unsaturated fatty acids ratio

Reduction of contamination of broiler carcasses with Salmonella spp. through the supplementation with Saccharomyces cerevisiae boulardii CNCM I-1079: a multi-analysis

Abstract ID: 271

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Salmonellosis remains one of the most frequent foodborne zoonosis, constituting a worldwide major public health concern. Currently, at a global level, the main sources of infection for humans are meat products, including the consumption of contaminated poultry meat. Alternatives to antibiotics to reduce the carriage of foodborne pathogens get more attention in recent years. There is evidence that some probiotics such as live yeast could be effective on this subject. A multi-analysis is performed with the objective of assembling the results available on the use of one specific live yeast (*Saccharomyces cerevisiae boulardii* CNCM I-1079 (LY)) on the reduction of *Salmonella spp.* in broiler chickens. Five studies from literature reporting the effect of this probiotic on contamination of chicken carcasses with *Salmonella spp.* are included in the analysis. Each selected publication contains information on *Salmonella spp.* carriage in the cecum as well as contamination of the carcass. In all five studies, LY was added to the broiler feed at the same level ($1x10^9$ CFU/kg of feed). Data are analyzed using a binary logistics model (Generalized Linear Mixed Model – SPSS 22.0). Results show that supplementation of broiler feed with LY significantly reduced the number of animals positive for *Salmonella* carriage (Control: 37% positives versus LY: 12% positives; $P<0.05$). The number of carcasses contaminated with the foodborne pathogen were also significantly reduced in the LY-groups (Control: 41% positives versus LY: 20% positives; $P<0.05$). Results from this multi-analysis suggest that supplementing broiler feeds with *Saccharomyces cerevisiae boulardii* CNCM I-1079 can help reduce *Salmonella spp.* carriage of and thereby reducing the contamination of carcasses.

Keywords: Broiler, Saccharomyces cerevisiae boulardii, Salmonella

Research of Physiological and Microbiological Peculiarities of Meat Chicken Breeds Digestive System in Fetal and Post-Fetal Periods for the Purpose of Creating New Feeding Techniques Ensuring the Fullest Possible Implementation of Genetic Potential of Poultry

Abstract ID: 108

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At the project course the embryo chicken, meat poultry and hybrids (broilers) digestive processes through embryonation and early ontogenesis observations were carried out. It has been found that during chicken embryonic development period the hybrids have better developed digestion as compared to the parental lines, which is characterized by the lipolytic and amylolytic activity presence in the embryonic tissue homogenate (7 days) and intestinal and pancreatic tissue (14 days). The day-old chicken has high levels of pancreatic tissue pancreatic enzymes, besides hybrids and parental lines performance differences are not significant at the prime ontogenesis period. The chicken body weight gain depends on the pancreas development and physiology level, which to 35 age days increases in the hybrids up to 62.7 times, in Plymouth Rock chickens breed to 49.9 times, in Cornish to 40.7 times. The highest chicken growth rate is noted in the first two weeks of bringing up, then it's gradually decreasing to 28-35 days of age, but the hybrids feature is that their growth rate is ahead of the parental line by 4.1-6.9%, and of body weight by 19.4-30.2%. The pancreatic enzymes (amylase and protease) activity dynamics in plasma tends to decrease with the chicken age, excepting of lipase, in which the dynamics has inverse trend - the wave-like enzyme activity increase to 35 age days. Protease activity reduce, apparently, due to age-related blood pressure changes in which regulation proteolytic blood enzymes are involved through the kinin system. The hybrids pancreas digestive function formation period is faster, which is confirmed by a more intensive pancreas mass gain and high enzymes activity at 7 days age when different chick groups management carried out with the same nutritionally and energy diet. This dynamic is maintained up to 14 days. During this period the chickens rations differ: hybrids get more nutritious feed. At 14 days age in the pancreatic tissue amylase and protease dynamics there is a high level of enzymes activity, lipase reaches its maximum level to 21 day age, which indicates the digestive system functional formation completion. During this period the growth rate and relative pancreas mass rate are stabilizing. From 14 to 35 days age hybrids outperform parental lines in body weight and the pancreas weight, as well as the pancreas tissue proteolytic enzymes activity. The study was financed by Russian Science Foundation, grant RSF №16-16-04089

Keywords: Amylase, Broilers, Pancreatic enzymes, Protease

Response of Japanese quails fed diets containing graded levels of Honey-flavoured sun-dried cassava peel meal as a replacement for maize

Abstract ID: 321

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A study was carried out to investigate the response of Japanese quails fed diets containing graded levels of sun-dried cassava peel meal flavoured with 6 % honey. The experiment lasted for four weeks, one hundred and eighty (180) unsexed two weeks old Japanese quails with initial body weight of 30.83 g were used. The birds were randomly allotted to four dietry treatments replicated thrice with 15 birds per replicate making total of 45 birds per treatment and designated T1, T2, T3 and T4 containing 0 %, 50 %, 75 % and 100 % sun-dried cassava peel meal flavoured with honey as replacement for dietary maize respectively. The parameters measured were initial weight, average feed intake, average daily weight gain, feed conversion ratio, final body weight gain and nutrients digestibility. All data collected were subjected to analysis of variance (ANOVA) using SAS. The results shows that there were significant ($P<0.05$) differenes in final body weight. However, there were no significant ($P>0.05$) differences in initial body weight, average feed intake, average weight gain and feed conversion ratio. The nutrients digestibiliy results showed that all parameters measured were significantly ($P<0.05$) different. It was observed that all treatments had digestibility values above 50 % which means they all had high apparent nutrients digestibility. It was concluded that honey flavoured sun-dried cassava peel meal can replace maize completely in the diets of growing Japanese quails without any deleterious effect on growth performance and nutrients digestiility of the birds.

Keywords: Cassava peel, Honey, Japanese quails, Sun-dried

Scientific review of selenium enrichment data in different organs in broilers fed organic selenium source (Alkosel)

Abstract ID: 56

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Selenium (Se) is an essential element for animal and human health, playing a role in the immune system, antioxidant defences, reproduction and ageing. The results of various studies showed that Se-enriched animal products (egg, meat, milk) may be used as a new potential source of this scarce element in human nutrition. Different Se sources (inorganic/organic) are available on the market and Se bioavailability in different organs represents an accurate measurement of Se product efficiency and quality. The objective of this study was to review existing information of 13 studies (performed on broilers, ducks and pheasants; 85% of these studies were performed by external research institutes) on Se enrichment in blood and different organs (liver, breast and thigh muscle, Fabricius bursa and feathers). Depending on the studies, animals were supplemented with inorganic Se (sodium selenite), organic Se (ALKOSEL) or a mix of organic and inorganic Se. For each study, the percentage of Se improvement in the different organs following organic Se supplementation was therefore calculated after comparison to control (no added Se) or inorganic Se. This source of organic Se induced Se enrichment ranging from +15% to 316% in blood, from +11% to 526% in liver, from +13% to +38% in feathers, from +4% to +1364% in thigh muscle and from +7% to +1549% in breast muscle. Se level was also improved by 31% in Fabricius Bursa, the major immune organ in the bird and by 24% in bone following organic Se supplementation. Moreover, one study displayed a reduction of 12% of Se excreted in feces with this source of organic Se, underlining better Se bioavailability for the animals. Besides the improvement of Se bioavailability in these organs, organic Se supplementation was also able to improve zootechnical performance (body weight, ADG, feed intake, FCR) in several studies. This scientific review underlines the robustness of Se deposition in blood and different organs (blood, liver, muscles, Fabricius bursa, feathers) following organic Se (ALKOSEL) supplementation, compared to control diets or inorganic Se (SS) and confirms the results of another meta-analysis performed on laying poultry showing better Se deposition in egg compartments (albumen, yolk, eggshell, whole egg) following supplementation with this Se-enriched yeast.

Keywords: Bioavailability, Broilers, Organic selenium

The adverse effect of surplus dietary L-arginine supply on parameters of the nitrogen metabolism in moderate restrictively fed growing cockerels

Abstract ID: 241

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The objective of this study was to examine effects of surplus dietary arginine (Arg) on the nitrogen (N) metabolism of growing cockerels nutritionally stressed by moderate feed restriction. From day 28 to 49 thirty-two Lohmann Dual cockerels were kept in metabolic cages and supplied with two diets differing in Arg concentration only (CON: 1.37% Arg; ARG: 2.04% Arg). Both diets were fed ad libitum (AL) and restrictive (RES) (n=8 birds per diet*regime). During the study daily feed intake (DFI) of RES birds complied with 75% of AL birds' DFI. From day 35 to 49 a N balance trial with two periods of 7 days each was carried out. Birds were weighed weekly and residual feed was recorded daily. Excrements of each bird were collected twice a day. Beside the determination of daily weight gain (DWG), N intake (DNI), N excretion (DNE) and N balance (DNB), the N efficiency ratio (NER; quotient of DWG and DNB) and N utilisation (NU; quotient of DNB and DNI) were calculated. Balance parameters were related to birds' metabolic body weight ($kg^{0.67}$). At day 35, 42 and 49 serum samples were taken from each bird to analyse N metabolism associated parameters including total protein, albumin, uric acid and urea. Statistical analysis was performed as three-factorial ANOVA with "diet", "regime", "week" as fixed effects using a mixed model. Throughout the study AL birds showed 20% higher mean DWG, DNI, DNE and DNB than RES birds ($P<0.001$). As both diets were isocaloric and isonitrogenous, mean DWG and DNI did not differ between CON and ARG birds. However, latter ones showed 9% higher mean DNE ($P=0.01$) and slightly lower NU than CON birds ($P<0.001$). Additionally, a minor diet-dependent difference in mean DNB was detected in AL cockerels only ($P<0.05$). While the feeding regime did not influence serum levels of examined parameters, mean serum urea level was 20% higher in ARG than in CON birds and slightly correlated with mean DNE ($r=0.248$; $P<0.05$). In conclusion, sole surplus Arg supply did not improve N balance and subsequent body growth in moderate restrictively fed cockerels. On the contrary, present results even indicated that additional dietary Arg has mainly been metabolised to urea and renally excreted impairing metabolic utilisation of dietary N.

Keywords: Arginine, Balance Study, Cockerels, Feed Restriction, Nitrogen Metabolism

The comparison of farm formulated and commercial diets on broiler production performance and carcass yield.

Abstract ID: 536

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The high cost of commercial feeds is a major contributor to the increased cost of broiler production in Lesotho. A completely randomized study was implemented at the National University of Lesotho to evaluate the influence of farm formulated diets on broiler performance and carcass yield using locally available feed resources and to carry out economic analysis to establish which feed was cheaper between commercial and farm formulated feeds. The two treatments were made up of control (commercial feeds) and treatment (farm formulated) and each treatment was replicated four times with 30 birds per replicate. The experimental diet was based on maize, dried brewery grain and soyabean, and it was formulated to satisfy the National Research Council requirements for broilers. A total of 240 day-old Ross 308 chicks were reared under deep litter system in a well-ventilated house with a lighting programme of 20 hours per day. Feed and water were provided ad libitum during the trial. Data collection on performance was done on weekly basis while carcass parameters data was done on day 42 by slaughtering ten chickens per replicate. Economic analysis was compiled at the end of trial as the cost per kilogram feeds and feed cost per kilogram weight gain. According to the findings there were no significant ($p>0.05$) difference between broilers fed commercial and farm formulated feeds on, feed intake, body weight, feed conversion ratio, carcass weight, dressing percentage, heart and the liver. On the other hand there were significant ($p<0.05$) difference between broilers fed commercial and farm formulated feeds on, gizzard weight and intestinal weight whereby birds fed farm formulated had higher weights than those fed commercial diets. Economic analysis results revealed that the use of farm formulated diets reduced the feed cost by 7%. The implications of these findings suggest that farm formulated diet can be used to obtain same production performance and carcass yields as commercial diet. Farm formulated diets were relatively cheaper to produce than the commercial, resulting in a more economic production of broiler. It is recommended that poultry farmers could consider formulating their own feeds provided that they have necessary skills or can source out professionals to assist them.

Keywords: Broiler performance, Carcass yield, Farm formulated, Feed costs

The effectiveness of Taraxacum officinale under conditions of the intestinal microbial imbalance in broiler chickens

Abstract ID: 608

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The aim of the study was to determine the effectiveness of *Taraxacum officinale* extract, along with (or without) various periods of application of coccidiostat in feed, salinomycin, under conditions of impaired intestinal balance. Shortening the period of salinomycin application by its withdrawal from the 22nd day of age was aimed to minimize the amount of its consumption during rearing. A total of 240 1-d-old, Ross 308 chickens were randomly assigned to 1 of 6 treatments, each comprising 5 replicate pens, with 8 male birds per replicate. A 3x 2 factorial arrangement was employed with the following main experimental factors: the period of use of salinomycin (70 ppm) – 0, or shortened to the 21st day of age, or standard; feeding strategy - no feed additive or dry *Taraxacum officinale* extract (2 g/kg feed). The intestinal imbalance was experimentally induced by challenge model including feeding birds for 4 days with an inoculum from three *Clostridium perfringens* strains producing a netB toxin, and a single infection of *Eimeria spp.* as a 10-fold dose of live anticoccidial vaccine Paracox-5. The basic feed mixtures compositions were provocative and contained 10% rye, 10% wheat, and 2% fishmeal. The lack of salinomycin in feed resulted in the lower BWG and increased FCR compared to both other periods of coccidiostat use in the period 1-35 d of age. The growth performance indices obtained in the groups of chickens receiving coccidiostat in feed up to 21d were comparable to the standard period of salinomycin use. The use of herbal extract improved FCR, both in the absence of coccidiostat in compound feed and in case of its standard application period. Moreover, salinomycin residues in feces collected on the 42nd day indicate the circulation of this substance, despite its withdrawal from compound feed from the 22nd day of life.This study was supported by the National Centre for Research and Development, Poland (grant “GUTFEED” number: BIOSTRATEG1/267659/7/NCBR/2015).

Keywords: Broiler chickens, Growth performance, Herbal extract, Intestinal imbalance

The effect of a combination of prebiotics and herbs rich in polyphenols on the growth performance and uniformity in broilers

Abstract ID: 415

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Prebiotics, e.g. gluconic acid and mannan-oligosaccharides (MOS), added to feed can influence the composition and activity of the microbiota and improve growth performance. Polyphenols derived from herbs are known for their anti-oxidant and anti-inflammatory properties. They may reduce the risk of oxidative stress and inflammation in the digestive tract which could potentially jeopardize growth performance. The aim of this study was to investigate the effect of dietary supplements consisting of gluconic acid, MOS and herbs rich in polyphenols (GMH) on broiler performance and uniformity. 640-day-old male chicks (Ross 308) were randomly allocated to two groups (Control group (C) no supplementation, Treatment group (T) supplemented with 1kg GMH per tonne of feed). Every group consisted of 8 replicate pens with 40 birds per replicate. The duration of the trial was 35 days, and birds were fed with mash corn-soybean diet *ad libitum*. Birds were weighed individually on day 1, day 14 and day 28 and in groups on day 7, 21 and 35. Feed intake (FI) was determined weekly per replicate. Body weight gain (BWG), feed conversion ratio (FCR) and European broiler index (EBI) were calculated. The uniformity of the group was assessed using the Shapiro-Wilk test on days 14 and 28 (IBM SPSS Statistics, version 24). The test evaluates whether the body weight (BW) distribution in the groups are statistically significantly different ($P<0.05$) from a normal distribution Gauss Curve. In a 35-day period BW, BWG, FI, FCR, and EBI were improved by 5.2, 5.3, 3.6, 1.3 and 5.3% respectively in the supplemented group compared to the C group. The uniformity test showed that C group BWs exhibited a distribution significantly different ($P<0.001$) from a normal Gauss Curve whereas birds in the T group showed a normal distribution. In conclusion, the dietary GMH supplementation improved the performance and uniformity among the broilers. Further studies are necessary to evaluate the relationship between the improvement in growth performance to the gastrointestinal tract microbiota and morphology, as well as the possible reduction in oxidative stress in broilers.

Keywords: Gluconic acid, Herbs, Mannan-oligosaccharides (MOS), Polyphenols, Prebiotic

The effect of alfalfa and ascorbic acid on performance and egg quality

Abstract ID: 109

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The purpose of the experiment was to find out how the addition of dehydrated alfalfa (DA) and ascorbic acid (AA) into hen diet influences the performance characteristics and egg quality characterized by physical parameters, carotenoids content in yolk and oxidative stability of yolks. The hens Hisex Brown were divided into 6 treatments according to level of DA (0 and 40 g/kg) and concentration of AA (0, 100 and 200 mg/kg) in diet. Hen-day egg production and egg mass production was reduced by the supplement of DA ($P<0.001$) and AA ($P=0.017$). The addition of DA itself increased feed intake per egg ($P<0.001$) and feed conversion ratio ($P<0.001$). The heaviest eggs ($P<0.001$) were ascertained in hens fed the diet with 100 mg/kg of AA (67.6 g) and the lightest were laid by the hens with 100 mg/kg of AA and 40 g/kg of DA (65.3 g) in diet. The ascorbic acid supplement improved egg content quality, i.e. Haugh units ($P<0.001$), albumen index ($P<0.001$) and yolk index ($P<0.001$). Whereas, DA addition increased values of yolk colour ($P<0.001$) and had a negative effect on shell quality. The interaction of DA and AA was detected in zeaxanthin concentration in yolks ($P=0.002$). The highest concentration was found in eggs from hens fed the diet with DA supplement and 100 or 200 mg/kg of AA (17.33 or 17.25 mg/kg dry matter of yolk). In addition, 40 g/kg of DA significantly increased the concentration of another carotenoids, lutein ($P<0.001$) and ß-carotene ($P<0.001$) in yolks, and oxidative stability of fresh eggs ($P<0.001$). The results suggest that DA in amount of 40 g/kg in diet is a good source of carotenoids and in combination with AA the better deposition of carotenoids in yolks occurs. On the other hand, DA and AA deteriorate performance and DA shell quality characteristics.

Keywords: Alfalfa, Ascorbic acid, Carotenoids, Physical parameters of egg quality, Vitamins

The effect of bee bread as a feed additive on selected egg quality and hens' performance

Abstract ID: 266

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The aim of this study was to evaluate the influence of bee bread used as a natural feed supplement (dose 5% wt. of feed mixture) for laying hens. The effect of bee bread on egg quality and hens' performance were determined. The experiment with a total of 36 ISA Brown laying hens lasted for 7 weeks. Egg production and feed consumption were monitored and calculated. Egg traits such as eggshell strength and thickness, egg mass, yolk colour and albumen height were determined. The positive effect of bee bread feed supplementation on eggshell strength and thickness was observed ($P<0,05$). In experimental group the tendency to intensify of egg yolk colour was observed. Probably it was a result of high content of plant dyes in bee bread. However, effect of bee bread on other analyzed egg quality traits, egg production and feed consumption did not reveal any differences among groups. Feed conversion ratio (FCR) per 1 egg was 12,5% better in the experimental group than in the control one. Supplemetation did not negatively influence birds' health conditions. Result of the study suggested that using bee bread as a supplement for hens feed could positively affect selected egg quality traits and feed conversion ratio.

Keywords: Bee product, Poultry feeding

The effect of feed additives on cecal short chain fatty acids production

Abstract ID: 606

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The aim of the study was to find the effect of different feed additives on cecal short chain fatty acids production in broiler chickens at 15 days of age.
In total 288 Ross 308 chickens, both sexes, were used in the experiment. They were housed in 24 cages, 12 chickens per cage. Chickens were divided into four treatment, each treatment had six replication with twelve chickens. Till seven days of age all chicken fed the same starter diet. From seven to fifteen days of age chicken fed the same diets supplemented with different additive at level 0.7%. Control diet was without any supplement, A diet was supplemented with cellulose arbocel, B diet was supplemented with fiber on the basis of sugar beet, BioFaser and C diet was supplemented with Bactria Control SF1 product on the basis of organic acids.
At day 15 all birds were euthanized according to legislation on the protection of animals used for scientific purposes, by a blow to the head and bleeding. The caeca of each bird was immediately dissected and digesta of 12 chickens was gently squeeze out to the tube and immediately frozen. Later the digesta was homogenized and used for short chain fatty acid measurement.
Diet with arbocel had significant positive effect on acetic acid production ($P<0.05$). There was no significant difference in production of butyric, propionic and lactic acids among the treatments. Research was supported by the Ministry of Agriculture of the Czech Republic (Project No. QJ1610219).

Keywords: Acetic acid, Butyric acid, Lactic acid, Propionic acid

The effect of incremental dietary levels of canola meal on growth performance of broiler chickens

Abstract ID: 275

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Solvent-extracted canola meal (CM) is commonly used source of protein for poultry. Dietary inclusion level of CM, however, has been historically limited to 5-10% due to concerns related to high fiber content and the presence of glucosinolates. A four-week feeding trial was conducted to determine the effect of varying inclusion levels of CM on the growth performance of broiler chickens. The experiment had four phases of six treatment groups: pre-starter (0, 3, 6, 9, 12, 15 % of CM), starter (0, 4, 8, 12, 14, 18 % of CM), grower 1 (0, 5, 10, 15, 20, 25 % of CM) and grower 2 (0, 6, 12, 18, 24, 30 % of CM), each lasting for one week. Diets were balanced for SID AA contents by replacing soybean meal (SBM) in a corn-SBM basal diet with graded levels of CM. One-day-old broiler chickens (housed 5 birds/cage) were allotted into the six treatment groups with ten replicates/cages per treatment. Body weight gain (BWG) and feed intake (FI) of birds were monitored weekly and feed conversion ratio (FCR) was calculated. Completely randomized design using the GLM procedure of SAS was used. Overall, the result showed that although NDF content of diets differed substantially with increased levels of CM bird performance was not significantly (P>0.05) affected by CM inclusion levels. Irrespective of the phase and CM inclusion levels, FI, BWG, and FCR averaged 2,019 g/bird/28 d, 1,387 g/bird/28 d, and 1.46 g/feed/g gain and were similar to 1,921 g/bird/28 d, 1,325 g/bird/28 d, and 1.45 g feed/g gain for the control treatment, respectively. Incremental levels of CM inclusion didn't have a significant (P>0.05) effect on NDF digestibility. It could be concluded that CM can effectively replace SBM when used up to 30% in broiler chicken diets assuming the diets are formulated on a digestible AA basis. Canola fiber has minimal effect on nutrient utilization as seen from the excellent performance of broiler chickens fed diets of different fiber content.

Keywords: Broiler chicken, Canola meal, Growth performance, Inclusion level

The effect of polyphenols and vitamin E on the growth performance, meat quality and antioxidant status of broiler chickens under heat stress conditions

Abstract ID: 234

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A 35-day experiment was conducted on 120 Ross 308 male broiler chickens (6 treatments, 20 birds in each group) to determine the effect of vitamin E and natural polyphenols (onion and grape seed extracts, Proviox) on the growth performance, meat quality and antioxidant status of birds exposed to heat stress. Dietary supplementation with vitamin E and polyphenols was applied in the following experimental design: group I (negative control) – without supplementation; group II (positive control) – without supplementation; group III – supplementation with 100 mg/kg of vitamin E; group IV – supplementation with 200 mg/kg of vitamin E; group V – supplementation with 100 mg/kg of vitamin E and 100 mg/kg of polyphenols; group VI – supplementation with 200 mg/kg of polyphenols. Broiler chickens from groups II to VI were exposed to elevated ambient temperature (34^{o}C/8 h from 21 day). The following parameters were monitored: growth performance, carcass quality, blood indicators (GPx, SOD, TAS, α-tocopherol) and meat quality (pH, color, drip loss, water holding capacity). It was found that dietary supplementation with vitamin E, vitamin E and/or polyphenols in broiler chickens exposed to heat stress improved their body weights (P≤0.01), body weight gains and feed intake until day 28 (P≤0.05). The analyzed feed additives had no impact on carcass dressing percentage in broiler chickens exposed to heat stress. The highest percentage of breast muscles in the carcass (P≤0.05) was observed in chickens fed diets with an increased content of vitamin E (IV) and vitamin E and polyphenols (V). Increasing dietary inclusion levels of vitamin E and/or polyphenols enhanced the activity of blood antioxidant enzymes (SOD, GPx) and increased serum α-tocopherol concentrations in heat stress-exposed birds (P≤0.01). The total antioxidant status (TAS) was highest in the negative control group (I) and group IV (200 mg/kg). The experimental diets had no effect on the pH value of meat. The breast meat of heat stress-exposed chickens fed the control diet (group II) and the vitamin E-supplemented diet (group III) was darker than the breast meat of birds fed diets with vitamin E and/or polyphenols and birds from the thermoneutral group. The *b** value (yellowness) of meat significantly decreased (P≤0.05) in groups with polyphenol supplementation (V and VI). The dietary inclusion of vitamin E and/or polyphenols reduced drip loss of breast meat in chickens exposed to heat stress (P≤0.01). The results of the study suggest that addition of polyphenols (VI) improve the growth performance, and increased content of vitamin E (IV) improve the antioxidant status of broiler chickens under heat stress condition.

Keywords: Antioxidant status, Broiler chicken, Heat stress, Polyphenols, Vitamin E

The effect of synbiotic and probiotic preparations on the growth performance, gastrointestinal tract development and health status of turkeys

Abstract ID: 239

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An experiment involving 600 BIG-6 female turkeys was conducted for 105 days to evaluate the effect of synbiotic and probiotic preparations on the growth performance, gastrointestinal tract development and health status of birds (6 treatments, 5 replications per treatment). Turkeys received a basal diet without additives (control group I) and diets with the addition of probiotic preparations BioPlus 2B (group II) or Cylactin (group III) and synbiotic preparations S1 (*L. reuteri*, *L. plantarum*, *L. pentosus*, *S. cerevisiae*), S2 (*L. reuteri*, *L. plantarum*, *L. pentosus*, *S. cerevisiae*, *L. rhamnosus*) and S3 (*L. reuteri*, *L. plantarum*, *L. pentosus*, *S. cerevisiae*, *L. rhamnosus*, *L. paracasei*) - groups IV, V and VI, respectively. The following parameters were monitored: growth performance, carcass quality, the chemical composition of meat, the structure (length, weight) and functional parameters (pH, viscosity) of particular segments of the gastrointestinal tract and health status of birds (lysozyme, γ-globulin, ceruloplasmin and total protein). Dietary supplementation with probiotic and synbiotic preparations increased the average body weight (BW) of birds ($P \le 0.01$), improved the feed conversion ratio (FCR) and the European Efficiency Index (EEI) ($P \le 0.05$). The analyzed feed additives had no impact on slaughter yield or the chemical composition of turkey meat. The weight of proventriculus increased in the Cylactin- and synbiotic-supplemented groups. The pH of small intestinal digesta was higher in turkeys fed diets supplemented with probiotics and synbiotic preparation S2 (tendency), compared with the control group. The viscosity of small intestinal digesta was significantly reduced in birds that received synbiotic preparation S3 ($P \le 0.05$) and probiotic preparation BioPlus 2B (tendency), compared with the Cylactin- and synbiotic S1-supplemented groups. Synbiotics S2 (tendency) and S3 ($P \le 0.05$) decreased the pH of cecal digesta. The addition of probiotic and synbiotic preparations to turkey diets increased the serum levels of lysozyme (tendency) and γ-globulin ($P \le 0.05$) and decreased ceruloplasmin activity ($P \le 0.01$). The results of the study suggest that synbiotic and probiotic preparations improve the growth performance and immune status of turkeys.
This study was carried out as part of a research project funded by the National Centre for Research and Development, PBS3/A8/32/2015.

Keywords: Gastrointestinal tract, Health status, Probiotics, Synbiotics, Turkey

The effects of microbial muramidase inclusion in broiler diets on growth performance, apparent ileal digestibility and intestinal histology

Abstract ID: 153

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The current study evaluated the effects of different inclusion levels of microbial muramidase (Muramidase 007, DSM Nutritional Products) on gastrointestinal functionality by determination of apparent ileal digestibility (AID) of nutrients, investigation of intestinal histomorphology and quantification of resulting growth performance. Four maize-wheat-soybean experimental diets were produced without (C) and with different dosages of muramidase: low (L, 25,000 LSU(F)/kg), medium (M, 35,000 LSU(F)/kg), and high (H, 45,000 LSU(F)/kg); diets were fed to broilers for 35 days. At the end of the experiment, AID of ash, crude fat (CF), CP, Ca and P were determined and samples of the mid jejunum and ileum were collected for histomorphological observations. Data were subjected to ANOVA analysis using GLM procedure. Orthogonal polynomial contrasts were used to assess linear and quadratic effects of different levels of the muramidase. At the end of the trial, Muramidase 007 supplementation linearly increased body weight gain and decreased feed conversion ratio (FCR) and broilers received diets with the enzyme had lower FCR compared with those in C group ($P \le 0.05$). Adding the muramidase to broiler diets, also linearly increased the European poultry efficiency factor ($P \le 0.05$). Inclusion of the muramidase in broiler diets linearly increased AID of CP, CF and P ($P \le 0.05$) and H group had higher AID of CF and CP compared to C group ($P \le 0.05$). Microbial muramidase supplementation linearly increased ileal villus height to crypt depth ratio and decreased the number of ileal CD45 cells ($P \le 0.05$). Broilers fed M and H diets had fewer number of CD45 cells in the ileum compared to those in C group ($P \le 0.05$). In conclusion, the results of the present study demonstrated that inclusion of the microbial muramidase in broiler diets could increased AID of key nutrients and improved growth performance in broilers. Broiler feed supplementation of the muramidase can therefore be considered an interesting prospect to improve gastrointestinal functionality in broiler chickens. Biological mechanisms causing these improvements need to be studied further.

Keywords: Digestibility, Feed efficiency, Gastrointestinal functionality, Muramidase, Nutrient availability

The effects of moderate feed restriction and surplus dietary arginine supply on relative digestive organ weights in growing cockerels of a dual-purpose breed

Abstract ID: 243

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The objective of this study was to examine effects of moderate feed restriction and surplus dietary arginine (Arg) on relative digestive organ weights in growing cockerels. From day 28 to 49 thirty-two Lohmann Dual cockerels were kept in metabolic cages and supplied with two diets differing in Arg concentration (CON: 1.37% Arg; ARG: 2.04% Arg). Both diets were fed ad libitum (AL) and restrictive (RES; n=8 birds/diet*regime). During the study daily feed intake (DFI) of RES birds complied with 75% of AL birds' DFI. Birds were weighed weekly and residual feed was recorded daily. At day 49 birds were slaughtered and gastrointestinal content was removed and weighted in order to determine ingesta free body weight (IFBW). Absolute weights of liver, pancreas and sections of the gastrointestinal tract from crop to ceca were recorded. The length of intestinal sections from duodenum to ceca was measured additionally. For statistical evaluation organ weights were related to birds' IFBW (g per 100 g IFBW) and the length of each intestinal section was related to its absolute weight (mm per g organ) as well as to the length of the entire small intestine (%). Statistical analysis was performed as two-factorial ANOVA with "diet" and "regime" as fixed effects using general linear model. Due to 25% lower DFI, RES birds showed lower IFBW ($P<0.001$) and relative gizzard weights ($p<0.001$), but higher relative weights of pancreas ($P<0.05$) and crop ($P<0.001$) than AL birds. The additional Arg supply did not affect relative organ weights. Regarding the length of intestinal sections, the ileum, entire small intestine and ceca of AL birds were longer per gram than that of RES birds ($P<0.05$). Furthermore, as proportion of entire small intestine, the jejunum was shorter and the ileum was longer in AL than in RES birds ($P<0.05$). Likewise, ARG diet caused a longer jejunal and shorter ileal proportion of the small intestine than CON diet ($P<0.01$). In conclusion, this study indicated that moderate feed restriction leads to marked changes in the gastrointestinal development of growing birds. In this way moderate feed restriction and additional dietary Arg might affect intestinal digestion and subsequent nutrient absorption by changing relative lengths and weights of certain digestive organs.

Keywords: Arginine, Cockerels, Feed Restriction, Gut Length, Organ Weights

The effect of particle size and insoluble fiber sources on growth performance and gastrointestinal tract development in broiler starters

Abstract ID: 613

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The effects of particle size and inclusion of insoluble fiber sources on growth performance and gastrointestinal tract development was studied in broiler starters from d 1 to 21. A total of 660 Ross 308 male broilers was randomly allocated to 6 treatments arranged factorially, each treatment replicated 5 times. Two corn particle sizes (2 mm as fine and 5 mm as coarse) and three insoluble fiber sources (cellulose, rice hull, RH and sunflower hull, SFH) were evaluated in this study. The main effect of fiber source on weight gain and feed to gain was significant, broilers fed sunflower hull had higher weight gain ($P<0.01$) and better feed to gain ratio ($P<0.01$) than those fed RH and cellulose. Feed intake was not influenced by particle size and fiber sources. Inclusion of cellulose resulted to higher carcass weight ($P<0.05$) and small intestine weight ($P<0.01$), also, coarse grinding decreased carcass weight than fine grinding ($P<0.01$). Particle size and fiber source did not affect length of small intestine segments and relative weight of pancreas, proventriculus and ceca ($P>0.05$). Gizzard pH was reduced in broilers fed coarse ground corn ($P<0.05$) and sunflower hulls containing diet ($P<0.05$) compared to those fed fine ground corn and cellulose, respectively. Also, pH of ceca ($P<0.05$) was reduced in birds fed SFH than those fed cellulose and RH. Broilers received cellulose and SFH had heavier gizzard ($P<0.01$) compared to RH fed birds. In conclusion, the present data suggested that inclusion of SFH in diet of broiler chickens, through reduced gizzard and ceca pH and enhanced gizzard development and functionality, is beneficial to growth performance, while corn particle size had little effect on gastrointestinal tract characteristics in broilers fed mash diets.

Keywords: Broilers, Insoluble fiber, Particle size

The efficiency of xylanase in broiler chickens fed with increasing dietary level of rye

Abstract ID: 248

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The aim of this experiment was to evaluate the effect of xylanase addition to the diet with increasing content of modern hybrid rye (Brasetto variety) on performance indices and viscosity of small intestine content in broiler chickens. A total of 560 1-d-old, Ross 308 chickens were randomly assigned to 1 of 10 treatments, each comprising 7 replicate cages, with 8 male birds per cage. A 5x 2 factorial arrangement was employed, with five dietary levels of ground rye (0, 5, 10, 15, and 20%). All the diets were not supplemented or supplemented with xylanase (200 mg/kg of feed; Ronozyme WX, (CT) with minimum xylanase activity of 1000 FXU/g). In the starter rearing period (1-21 days of age) the inclusion of rye to the diet, even at the lowest dietary level (5%) negatively affected body weight gain (P<0.05) without effect on feed intake and feed conversion ratio. In older chickens (the grower-finisher rearing period; 22-42 days of age) none of the dietary levels of rye (5-20%) affected growth performance indices. Similarly, no significant effect of increasing dietary level of rye was found for entire rearing period (1-42 days of age). Diet supplementation with xylanase improved body weight gain and feed conversion ratio in chickens from 1 to 21 days of age. No positive effect of the enzyme was found in older birds. No significant effects of the used experimental factors on the results of slaughter analysis, i.e. the carcass yield, breast meat yield, abdominal fat and relative weight of the liver, were noticed. The high dietary concentration of rye (20%) increased the viscosity of small intestine content (P<0.05), however diet supplementation with xylanase significantly alleviated this effect. The findings of this experiment indicated that modern hybrid rye grain may be used at a 20% dietary level in broiler chickens nutrition during second feeding phase, i.e. from 22 to 42 days of age, without any detrimental influence on growth performance indices, while enzyme (xylanase) positively affected body weight gain and feed conversion ratio in younger chicks (1-21 days of age).
This study was supported by the National Centre for Research and Development, Poland (grant “ENERGYFEED” number: BIOSTRATEG2/297910/12/NCBR/2016).

Keywords: Broiler chickens, Growth performance, Intestinal viscosity, Rye, Xylanase

The intestinal inositol phosphate pattern in broilers as influenced by phytase source

Abstract ID: 314

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Recent data demonstrates that the inositol-phosphate (InsP) isomers generated in broiler digesta are phytase specific. This study investigated the effects of different phytase sources and doses on broiler s gastrointestinal InsP levels. Male Ross 308 chickens, (8 treatments, 9 pens with 10 broilers each), were fed pelleted corn-soybean based diets from d1-35. Starter and finisher diets (PC) contained 9.5 and 8 g Ca and 7.5 and 6 g P respectively or were reduced by 1.5 g P and 1.6 g Ca per kg feed (NC) without or with phytase (modified *E. coli* 6-phytase, PhyA or chimeric 6-phytase, PhyB; 400, 1250 or 3500 FTU/kg). Birds were slaughtered at d35, ileal contents analysed for $InsP_{2-6}$, specific isomers as well as myo-inositol (MI) and blood plasma for MI. Data were analysed by ANOVA and significance declared at $P < 0.05$. In control birds $InsP_{5+6}$ disappearance at the ileum was low (-22 and 0.08% in PC and NC respectively, P>0.05) which did not correspond to ileal P digestibility of 55 and 46% respectively (P>0.05). In broilers fed phytase supplemented diets $InsP_{5+6}$ disappearance increased to 28-35%, 66% and 93-94% (P<0.05) respectively and ileal P digestibility to 54-58%, 64% and 69-70% respectively, with no differences between phytase sources. Feeding 1250 FTU/kg feed resulted in higher ileal InsP4 levels for both phytases compared to NC and PC birds (P<0.05). But the identity of the ileal $InsP_4$ isomers differed between phytases with D- or L-Ins(2345)P_4 predominantly recovered in Phy A fed birds, whereas equal amounts of D- or L-Ins(2345)P_4 and D-or L-Ins(1234)P4 were recovered in Phy B-fed birds (P<0.05). While ileal MI levels did not differ, plasma MI levels increased with phytase application rate. It is concluded that intestinal InsP degradation and P digestibility is optimised by increasing the phytase application rate. Effects of phytase sources on the occurrence of phytase specific InsP isomers need further investigation for example in respect to their nutrient binding properties.

Keywords: Broiler, Inositol phosphates, Phytase source

The length of collection period affects the estimate of dietary metabolisable energy in broilers

Abstract ID: 159

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The aim of the study was to evaluate the effect of two collection periods, 48h and 96h on N – corrected apparent metabolisable energy (AMEn) of three dietary treatments with different energy density when fed to broiler chickens. A basal diet containing 499.5 g/kg of wheat, 235.0 g/kg soybean and 100.0 g/kg of maize, as major ingredients, was mixed. The basal diet was then split into 3 batches and one of them was used as a control (Diet 1), a second lot had 100 g/kg of vegetable oil added (Diet 2), and the third lot had 100 g/kg of soy hulls added (Diet 3). The diets were fed as mash and did not contain any coccidiostat, antimicrobial growth promoters, prophylactic, or other similar additives. A flock of birds was reared in a common pen and fed a standard diet until 24 d of age. One hundred forty four male Ross 308 broiler chickens were then randomly allocated to 48 raised floor pens (0.36 m^2), giving three birds per pen, each diet being replicated 16 times. The birds were fed the experimental diets for 4 days adjustment period prior to their allocated collection period (48 hours (8 replicates per dietary treatment) or 96 hours (8 replicates per dietary treatment)). Water and food were supplied ad libitum throughout the study. At the start of the collection period when birds were 28 d age, the solid floor of each pen was replaced with a wire mesh and all excreta were collected for 48, or for four 96 hours, respectively, immediately dried at 60^{o}C and then milled. Feed intakes were also measured for the same period as excreta collection. Data were statistically analysed by ANOVA using a 2 x 3 factorial arrangement of treatments. The main effects were the collection period (48 or 96 h) and the dietary treatment used. The determined AMEn values for 48h collection period were higher compared to 96h collection period ($P<0.05$), 13.63 vs 13.34 MJ/kg DM (SEM=0.086). Diet 2 had the highest AMEn ($P<0.001$), followed by diet 1 and diet 3, i.e. 12.87 and 12.25 MJ/kg DM, respectively (SEM=0.106). Feeding high energy diets produced relatively high AMEn. There were no ($P>0.05$) collection period x dietary treatment interactions. There were no differences ($P>0.05$) in average daily bird feed intake. The results demonstrated that length of collection period has an impact on the estimate of dietary AMEn values.

Keywords: Broilers, Length of collection, Metabolisable energy

The use of a prestarter contributes to a better start and optimal results of broilers

Abstract ID: 564

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Early feeding in broiler nutrition is an important parameter affecting the development of the day-old chick and its later performance. A large-scale trial has been set up in 2 consecutive rounds each consisting of 8 pens of 1150 broilers. The objective of this trial was to investigate the effect of a prestarter feed on the production performances, foot pad dermatitis and litter score of the broilers. Day-old chicks (as hatched) were fed a prestarter feed (P, mash/pellet mixture) or a common starter (S, crumble). In each round, 4 pens received either P or S (30 g/chick) on chick paper, while feeders in all pens were filled with S. Further, all pens were fed according to the same feeding schedule with increasing levels of whole wheat until day 39. At day 32, each pen was thinned by removing 280 animals.
In the first round, mortality was very low and not affected by the treatment. However, in the second round, overall mortality was higher and tended to be reduced in treatment group P compared to group S. In one control pen S, an antibiotic treatment on day 3 was necessary due to yolk sac infection, whereas no antibiotics were used in the P pens.
In both rounds, body weight at day 39 was numerically higher in the P group showing a difference of 29 g on average. As feed intake was not affected by the dietary treatment, FCR was also lower in the P treatment ($p=0.08$). This resulted in a higher European Production Efficiency Factor ($p=0.06$) and a profit of 0.004 € per chick.
In this trial no differences between treatment groups were noticed with regard to litter quality and foot pad lesions.
It can be concluded that the application of a prestarter feed contributes to a better start of day-old chicks resulting in less mortality and a technical and economical benefit.

Keywords: Broiler, Early nutrition, Prestarter

Trace mineral sources and zinc levels influenced growth performance, tissue mineral content and carcass yield of 35-day old male broilers

Abstract ID: 189

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A total of 900 Ross 308 male broiler chicks at zero-day old were allocated to four treatments in a randomized complete block design and 2×2 factorial treatment arrangement. Each treatment had 15 replicates with 15 birds each. The factors were two sources (sulphate or hydroxy) of two trace minerals (Zn and Cu) and two levels (low or high) of Zn. Zinc and Cu were supplied as zinc sulphate monohydrate and copper (II) sulphate pentahydrate or zinc hydroxychloride and copper hydroxychloride. Diets containing sulphate Zn and Cu are denoted sulphate trace minerals (STM) and those containing hydroxychloride Zn and Cu are denoted hydroxy trace minerals (HTM). The supplementary dietary Cu level was 15 ppm. The supplementary low dietary Zn level was 20 ppm or 80 ppm for high Zn level. The wheat-soybean meal based diets was fed throughout the experiment. Birds and feed were weighed on days 0, 21 and 35. On day 35, seven birds per pen were processed for carcass evaluation, and the left tibia bone, liver and blood were collected from additional four randomly selected birds per pen. There was no significant trace mineral source × Zn level interaction on any of the growth performance response. Broiler chickens receiving HTM had lower ($P < 0.05$) FCR whereas broiler chickens receiving lower Zn supplemental level had greater ($P < 0.01$) weight gain than those receiving the higher level of Zn supplementation. Broiler chickens receiving HTM had greater % breast yield ($P < 0.05$) than those receiving STM and broiler chickens receiving the lower Zn level had greater ($P < 0.05$) % carcass yield and breast. Broiler chickens receiving higher level of Zn, irrespective of the trace mineral source, had greater ($P < 0.01$) tibia and plasma Zn levels. On the other hand, liver Cu was greater in broiler chickens receiving HTM. It was concluded that hydroxy Zn and Cu are more efficacious than sulphate Zn and Cu in promoting growth performance and enhancing meat yield in the male broiler chickens but the reasons for lower performance of the chickens at higher supplemental Zn level need to be further investigated because it was not related to Zn availability or deposition in tissues.

Keywords: Broiler, Copper, Growth performance, Meat yield, Zinc

Transfer of different forms of vitamin K from hens to broilers

Abstract ID: 491

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Vitamin K consists of a group of structurally related compounds; phylloquinone (K1), that is synthesized by plants, and the menaquinones (MKs also known as K2) synthesized by bacteria. Menadione (K3) is routinely added to poultry diets. It is designated K3 and does not have a side-chain. There is increasing evidence that vitamin K plays a significant role in bone metabolism, in addition to its recognized role in blood coagulation. The aim of this study was to determine the efficacy of vitamin K transfer from the maternal diet to the chick.
Ross broiler breeder pullets were obtained at 20 weeks of age and placed into four floor pens with each pen containing 12 hens and 2 roosters. The pair of roosters was rotated between pens on a weekly basis. A basal diet was formulated that met nutrient requirements of the hens and to it was a added per kg either No vitamin K; 3.0 mg Vitamin K3; 3.0 mg vitamin K1 (QAQ) or 6.0 mg vitamin K1 (QAQ) to make up the four experimental diets 1, 2, 3, and 4, respectively. Quinaquanone (QAQ) is a patented product of Agricure Pty Ltd. It is a stabilised soluble formulation and contains both K1 and K2 in the ratio of 10:1. Eggs were incubated and after hatching,chick livers were removed and analysis for vitamin K1 and vitamin K2 (MK4, menaquinone).MK4 was measured as it is believed that it is alkylated from of menadione in the liver.
Low hepatic concentrations of Vitamin K1 were found in chicks from hens fed diets without supplementation with this vitamin. Dietary supplementation resulted in significant ($P<0.05$) increases in hepatic concentrations that reflected levels in the diet. In contrast concentrations of MK4 remained relatively constant across treatments although there was a doubling $P<0.05$) of the concentration of liver values from diet 4 compared to those from diets 1 and 2. The ramifications of the hepatic reserves of vitamin K from feeding hens different forms of vitamin K, found in this study, for the skeletal development and health of the chick awaits elucidation.

Keywords: Broiler, Broiler breeder, Liver, Vitamin K

Triticale grain in diets for laying hens

Abstract ID: 99

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At present heavy-yielding hybrid feed-grade cereals gain growing importance worldwide as energy sources in poultry diets. Triticale is a wheat-rye hybrid; its nutritive characteristics are close to wheat, except higher content of total protein and especially sulfur-containing amino acids. The contents of antinutritive factors (arabinoxylans, phytin, resorcinols and other phenolic metabolites, protease inhibitors, tannins) in triticale depend on the cultivar. The aim of our study was the determination of reasonable levels of the inclusion of triticale into the compound feeds for laying hens (cross SP-789, 30 birds per dietary treatment). The trial lasted for 6 months of the productive season. It was found that triticale can be included into the diets for layers as a partial (up to 30% of total diet) substitute for wheat without detrimental effects on egg productivity. Increased substitution level (45%) decreased the intensity of lay (by 1.4%) and increased feed expenses (kg) per 1 kg of eggs laid (by 2.2%). Full substitution of triticale for wheat (60% of total diet) significantly ($P<0.001$) decreased the intensity of lay (by 12.0%), increased feed expenses (by 21.4%), impaired digestibility and assimilation of dietary nutrients. The substitution of triticale for wheat had no effect on egg morphology (with the exception of the highest triticale level, 60%, which led to certain insignificant decreases in eggshell thickness and resilience); concentrations of vitamins A, E, B2 in yolk and liver fell within the physiologically normal ranges. Histological investigation of hepatic and duodenal tissues revealed structural abnormalities, granular and fatty degeneration of the hepatocytes only at maximal dietary triticale level (60%).

Keywords: Compound feeds, Duodenum, Intensity of lay, Laying hens, Liver

Use of a calcium particulate and feeding time in combination with 25-hydroxycholacifereol to reduce fracture susceptibility in laying hens

Abstract ID: 635

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Keel bone fractures (KBF) in laying hens is a serious problem for the laying hen industry with high frequencies found in all genetic lines and housing systems. The aim of our study was to investigate a nutritional solution to reduce KBF by providing calcium particulate in the two hour period immediately before the dark phase (CaT) alongside a second factor (25-hydroxycholecalciferol, HyD) known to increase calcium absorption. For the experiment, a total of 240 birds were used that were exposed to one of four treatment combinations in a 2X2 factorial design (Factor 1: CaT^+ or Control (CaT^-); Factor 2: HyD vs. Control (HYD^-). All birds were housed in a commercial aviary until 33 weeks of age when they were moved to a test barn and randomly assigned to one of the eight pens (30 hens/pen). After one week of habituation to the novel housing, all birds began to receive one of the four treatment combinations (n = 2 replicate pens/treatment combinations).
In order to provide bird-level egg production data, at 37 weeks of age, twenty focal birds/pen were selected to ingest a gelatin capsule containing one of three fat soluble dye colors for three consecutive days (Appleby and McRae, 1983). The method allowed individual eggs to be traced back to a specific hens despite being group housed. At +2, +3, +4 and +5 days after the third day of capsule administration, all eggs were collected and underwent quantification of biomechanical strength, mass and thickness of the egg shell. The ability of the treatment combinations to reduce fracture susceptibility was tested using an *ex vivo* impact testing protocol
At 39 weeks of age after which the keels were removed and scored for damage using an established three-point severity scale. Bird-level egg measures were statistically analyzed using „Statistica v. 6.1"(Stat-Soft Inc., 2002). Fracture susceptibility was statistically analyzed using logistic regression with fracture occurrence as a binomial response and treatment combination, collision energy, and body mass as predictors. The likelihood of an experimental fracture occurring in the CaT^-/HYD^- was 3.6 times more likely compared to the CaT^+ /HYD^+ treatment (OR CI (1.1 to 5.8). At the same time, the strength of the eggs is highest in the CaT^+ /HYD^- group (54.4N), followed by CaT^+ / HYD^+ (53N) and CaT^- /HYD^+ (52.2N). The egg shell thickness is again greatest at CaT^+ / HYD^-(0.367mm), followed by CaT^+ /HYD^+ (0.363mm) and CaT^- /HYD^- (0.358mm).

Keywords: CaT, Egg parameter, Keel bone damage, Laying hens

Xylanase and xylo-oligosaccharide have a positive effect on body weight and indicators of immune function in broilers at 28 days of age

Abstract ID: 192

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The objective of this study was to investigate the effect of xylanase and xylo-oligosaccharides (XOS) on indicators of the gut immune response of broilers fed a nutritionally deficient diet and raised on new or mix of recycled and new (50/50) (dirty) litter. Six hundred and forty male Ross 308 broilers were used in this 28 day study. The treatments were arranged into a 2 × 4 factorial arrangement with 8 replicates per treatment. The factors were 2 litter types (dirty litter or clean litter) and 4 additives (control- without any additive, xylanase alone, xylanase + β- glucanase and 0.025% XOS). The energy and protein contents of the diets were reduced by 1MJ/kg and 30g/kg to 12.0 MJ/kg and 200 g/kg, respectively, and all diets contained 500FTU of phytase. On day 28, birds and feed were weighed and 2 birds per pen were randomly selected and euthanised. The spleen, bursa of fabricius and gizzard were removed and weighed. There was no significant effect of treatment on feed intake or FCR. Birds receiving diets containing xylanase + β- glucanase or XOS gained significantly ($P < 0.05$) less weight than those receiving control diets or diets containing xylanase alone. There was a significant ($P < 0.05$) litter type × additive type interaction for bursa of fabricius weight. Birds raised on dirty litter and receiving xylanase and β-glucanase had significantly lighter bursa than those raised on dirty litter and receiving XOS but there was no such effect for birds raised on clean litter. There was no significant effect of treatment on gizzard or spleen weight. In conclusion, litter type had no effect on growth performance however; the use of xylanase + β-glucanase decreased the weight of an organ associated with immune function compared to XOS. These observations indicate that carbohydrases supplementation possibly influences immune response in broilers, the effect of this on genes related to inflammatory and immune responses are being further investigated.

Keywords: Beta-glucanase, Broiler, Prebiotics, Xylanase, Xylose oligosaccharides

Association of Heterosis, Dominance effect on Body Weight and the Expression of Growth Hormone Gene and Insulin Like Growth Factor – I Gene in Slow Growing Chicken

Abstract ID: 320

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The aims of this study was, to investigate the relationship between genotype effect of *Growth Hormone* gene (*GH*) and *Insulin Like Growth Factor – I (IGF-I)* gene on heterosis, dominance effect, and the level of expression of slow – growing chicken. Korat chicken (KR) was used as a representative of slow – growing chicken. They were produced and 630 KRs were selected by *GH*, and *IGF-I* genotype. Completely Randomized Design was applied, each loci were divided into 3 groups by genotype, A1A1, A1A3, A3A3 and, AA, AC, and CC for *GH* gene, and *IGF-I* gene, respectively, each group composed with 4 replications. Age at 0, 2nd, 4th, 6th, 8th, and 10th week, chickens were weighted. The age at 2nd - 10th week, livers were collected. Total RNAs were isolated, level of mRNA was measured by qPCR. Heterosis (H) of each group were calculate by LSMeans, and dominance effect (D) were estimated by BLUP method, ASREML 4.1 software was applied. Correlation between growth traits, and H, D were analyzed. Significant effects of genotypes were analyzed by ANOVA. TUKEY's range test was used to test the significant difference of the mean. $P< 0.05$ was accepted to significance. Significant correlation between growth traits, and H, D at the most age were detected ($P<0.05$). Significant association between *GH* gene and H, D at all of age were found, while significant differences were detected at some of ages in case of *IGF-I* gene. Non – significant difference of mRNA level in different genotype of *GH* gene in all of ages were detected, while significant different was detected at the age of 2 weeks in case of *IGF-I* gene ($P<0.05$). The results can be concluded that heterosis, particularly dominance effect play a main role for growth traits. *GH* gene is mainly role in growth in most of age of chicken, but it is disaccorded with the expression of *GH* gene that significant different between different genotype cannot be detected.

Keywords: Dominance effect, Gene expression, Heterosis, Slow growing chicken

Bone stability and performance level of phylogenetically divergent chicken lines after dietary calcium restriction

Abstract ID: 280

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It has been observed, that poor bone quality and a high prevalence of fractures are important issues in laying hens. Since calcium is indispensable for eggshell as well as bone formation, long-term selection for increased egg production might be accompanied by a decrease of bone stability. The aim of this study was to get insights into the hens' adaptation response to a nutritional calcium restriction in relation to their laying performance and phylogenetic origin. The animal model consisted of four purebred layer lines, two brown and two white-egg strains, differing in performance level. In total 132 hens (33 per line) were semi-randomly placed in six 8 m^2 floor-housing pens. Two customary wheat-based diets, which only varied in terms of calcium content, were fed ad libitum. With regard to nutritional recommendations of calcium for high performing laying hens, the diets can be classified as adequate (Ca+; 4.26%) or deficient (Ca-; 1.09%). At the beginning of the 31st week of age, the control groups (2 pens) were fed the Ca+ diet over the entire 21-week experiment, while the test groups (4 pens) were fed alternatingly with both diets. In the latter case, a 3-week period of Ca- was followed by a 6-week period of Ca+ two times, ended with a third period of Ca-. At the end of the 52nd week of age, the animals were sacrificed, tibia and humeri dissected and their bone size measures and breaking strength recorded. Analysis of variance revealed a highly significant ($P<0.0001$) influence of line and diet on tibia and humerus breaking strength, while its interaction was only significant ($P<0.02$) for tibia. Both Ca- fed white-egg layer lines showed lower breaking strengths than their brown-egg counterparts. Within each phylogenetic group, there was no consistent tendency towards breaking strengths in relation to their laying performance. The results suggest that the genetic line has a major effect on bone stability. Laying intensity decreased considerably among all test groups during the deficient feeding periods compared to the controls (for details see Bues et al., same conference). Radiologically assessed bone mineral data and analyses of calcium related blood parameters are in progress and will provide deeper insight into the hens' adaptation response.

Keywords: Bone breaking strength, Laying hens, Laying performance

Chicken sperm cryopreservation as a tool of maintenance genetic diversity in small scale populations

Abstract ID: 382

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Gene resources preservation in poultry is an important task, and has obvious peculiarity in small scale populations. To avoid inbreeding in such populations there is needed to provide sex ratio up to 1:4-1:5. But such sex ratio leads to increased expenses due to keeping of additional amount of cocks and due to particularity of sexual behavior does not ensure genetic diversity in progeny. Use of the frozen individual ejaculates enables to solve this problem and makes possible and easy and safe exchange of genetic material between flocks. There is a problem of decreasing of fertilization ability of chicken sperm during the freezing-thawing cycle. After insemination by mixed native sperm the fertility percentage usually reaches 90-98%; after use of individual native ejaculates fertility is on the level of 70-90%; after use of individual frozen ejaculates – 17-60%. The degree of such decreasing in big extent depends on the breed and individual properties of a cock even if the cocks have been previously selected according to the quality of their native semen. Our research group has created a method of stepwise evaluation and choosing of the cocks for the goals of their sperm cryopreservation. It includes estimation of spermatozoa concentration (≥ 4,0 bln/mL), motility (≥80%), sperm cholesterol concentration (≤ M of cocks group) . Freezing was carried out in pellets with DMA. Application of this method enables to increase fertility up to 59-79%. Cryopreservation of chicken sperm makes possible not only to keep small scale populations, but also facilitate to preserve genetic material for their further use in order to restore vanished breeds and populations, to make molecular -genetic investigations and certifications.

Keywords: Chicken sperm, Cryopreservation, Gene pool, Genetic diversity

Direct and indirect effects of selected morphometric traits on body weight of fulani ecotype chickens of Nigeria

Abstract ID: 11

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A structural equation model (path analysis) was used to evaluate the direct and indirect effects of selected morphometric traits on body weight of Fulani ecotype chickens. The morphometric parameters measured were: body length, body girth, wing length, shank length and thigh length. Measurements were taken at 4, 8 and 12 weeks, respectively. Results showed that body weight correlated positively and significantly ($P<0.05$; $P<0.01$) with the morphometric traits at week 4, 8 and 12, respectively. However, thigh length did not have a significant ($P>0.05$) correlation with body weight at week 4. With the exception of thigh length at week 4, the traits all had positive and significant ($P<0.05$) direct effects on the body weight of the chickens. The sums of the indirect effects were observed to be positive and greater than the individual direct effects. The indirect effects were mostly felt via shank length at week 4, body girth at week 8, and thigh length at week 12, respectively. Percentage combined contribution of the morphometric traits indicated that body length via shank length (0.76), body girth via thigh length (30.78) and wing length via thigh length (237.70) made the greatest contributions to the body weight of the chickens at week 4, 8, and 12, respectively. The results revealed the existence of positive association between the morphometric traits and body weight of Fulani ecotype chickens which could be exploited for improvement of the chickens.

Keywords: Body weight, Direct and indirect effects, Fulani ecotype chickens, Morphometric traits

Discovering genes and pathways in the shell gland which may be involved in egg cuticle formation by gland shell RNA-seq data analyses

Abstract ID: 533

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The cuticle of an egg is formed in the shell gland and we know there is considerable genetic variation in cuticle deposition. We can use hormonal treatments to produce eggs without cuticles. In an effort to try to understand more about the factors which may control variation in cuticle deposition, we have analysed global gene expression in the shell gland with a view to elucidating the molecular mechanisms and biological pathways involved in cuticle deposition in the shell gland. In order to do that we have sequenced RNA from 8 biological replicate samples of chicken shell gland in each of two experimental conditions; GnRH and AVT, this resulted in hens laying eggs with and without cuticle respectively, but at the same chronological time. Quality control and data processing steps resulted in rejection of one replicate from the AVT group resulting in 8 GnRH and 7 AVT biological replicates of “clean” data totalling 889M aligned sequence reads. Of a total of 18346 genes present in the chicken genome, our sequencing reads were assembled onto 17108 genes (genes with reads per count per million (RCPM) ≥ 1 in at least 2 samples). After fitting gene-wise generalized linear models and performing likelihood ratio test, a total of 12248 genes are differentially expressed between the shell gland of the two conditions. After correcting for False Discovery Rate (FDR) we obtained a list of 3431 DE genes (representing around 18,7% of the total number of annotated genes in the chicken genome) of which 46 are up-regulated (fold change ≥ 1.5) and 115 are down-regulated (fold change ≤ -1.5). The set of down-regulated genes was particularly enriched with genes annotated with GO terms related to glycosaminoglycan binding. The results of these studies have expanded our knowledge of important genes and pathways involved in the final stages of egg formation including the deposition of the cuticle. We are further examining the expression of genes discovered by these experiments and believe it will lead to greater understanding of the function of the shell gland. Funded by BBSRC LINK grant BB/K0070921/1 ‘Cute-Egg’ with Aviagen and Lohmann Tierzucht.

Keywords: Cuticle, RNAseq, Shell gland

Does Chronic Heat Stress Affect Hypothalamic AMPK and (An) Orexigenic Neuropeptide Gene Expression in Broilers?

Abstract ID: 439

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Poultry production yield and welfare both benefit from a controlled environment. Heat stress is one of the most important environmental challenges to poultry production worldwide, mainly affecting growth rate and egg production. Many of the heat-stress induced effects can be traced back to a reduced feed intake. Feed intake is regulated by complex mechanisms involving brain-gut neuropeptides, but these neuropeptide pathways in broiler chickens remain unclear. It is hypothesized that orexigenic and anorexigenic neuropeptides are affected by AMP-activated protein kinase (AMPK) in the hypothalamus, and interfere with the expression and/or activity of enzymes involved in lipid metabolism and mTOR signaling (mammalian target of rapamycin). This study investigates the effects of chronic heat stress (40°C and 60% relative humidity) on the gene expression of peptides in the hypothalamus of broiler chickens at 16 days of age. At day 7, the chicks were surgically implanted with a stainless steel cannula system into the third ventricle. This allowed for intracerebroventricular injections (ICV) of 150 µg of AICAR, an AMPK activator. Throughout the experiment, performance, feed intake and feed conversion ratios were recorded. AMPK protein levels in hypothalamus and peripheral tissues are determined by Western Blot. The results from changes in AMPK protein levels in hypothalamus and peripheral tissues and from hypothalamic mRNA levels of AMPK, mTOR, neuropeptide Y (NPY), agouti-related peptide (AgRP), pro-opiomelanocortin (POMC), cocaine- and amphetamine-regulated transcript (CART), corticotropin-releasing hormone (CRH), acetyl-CoA carboxylase (ACC), thyrotropin releasing hormone (TRH), fatty acid synthase (FAS), carnitine palmitoyltransferase (CPT1), sterol regulatory element-binding protein 1 (SREBP1) and melanin-concentrating hormone (MCH), will be discussed in relation to the heat stress-induced alterations in feed intake.

Keywords: AMPK, Broilers, Feed intake, Hypothalamus, ICV injection

Egg production and egg shell quality of phylogenetically divergent chicken lines during dietary calcium depletion

Abstract ID: 358

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In high performing laying hens, approximately 2 g Ca are needed for egg shell calcification daily. To meet this high demand, hens rely on a complex interplay of intestinal absorption and mobilization of Ca from the skeleton. To investigate whether performance level has affected the capacity to adapt to changes in Ca supply, this study was performed on four different purebred layer lines that varied in laying performance and phylogenetic origin. In total 132 hens were semi-randomly placed in six 8 m^2 floor-housing pens (22 hens each). The diets applied to the hens ad libitum can be classified as adequate (Ca+; 4.26%) or deficient (Ca-; 1.09%). With beginning of the 31st week of age, the control groups were fed with Ca+ diet over the entire 21-week experiment. The test groups received a Ca- diet three times for a 3-week-period each. The first and the second Ca- period were followed by 6-week recovery periods (Ca+ diet). Throughout the Ca- periods, all eggs were collected and analyzed for egg weight, shell weight, breaking strength and shell thickness. After the third Ca- period (52nd week of age), the animals were sacrificed. Statistical analysis of laying performance and shell quality during Ca- periods revealed different responses. While high-performing white-layers (WLA) showed almost no significant reduction in laying rate, there were significant and distinct deteriorations in every other measured parameter starting 2 to 7 days after change of feed. In contrast, there was a decrease in laying rate in high-performing brown-layers (BLA) during the first Ca- period and even though there was a reduction in shell quality, it was not as pronounced as in WLA hens. In low-performing lines (L68, R11) egg shell quality parameters were only slightly affected. While there was no reduction in laying rate in brown-layer L68 hens, it decreased in white-layer R11 hens. In general, both brown-layer lines (BLA, L68) showed better shell quality and bone stability (see Jansen et al., same conference) than their white-layer counterparts (WLA, R11).

Keywords: Ca-depletion, Egg shell quality, Laying hens

Establishing of in vivo Transfection of Primordial Germ Cells in Chicken Eggs using the Sleeping Beauty Transposon System

Abstract ID: 515

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Here, we established a direct in vivo transfection method of chicken primordial germ cells. Therefore a complex of Lipofectamine 3000 and the Sleeping Beauty (SB) transposon system was injected into fertilized eggs. For the fluorescence detection of transfected cells a ubiquitously expressed Venus reporter transposon plasmid was employed. Microinjection of the transfection mix was performed into incubated fertilized eggs at 2,5 days of development. At this stage, migratory primordial germ cells peak in the blood circulation. In a first set of experiments, we analyzed the treated embryos at day 11 of embryonic development. In a second step, we optimized the molar ratio of the reporter transposon and the SB transposase plasmid, we achieved the highest efficiency of gonadal transfection and a good vitality of the embryos at a 6:1 ratio. Using this ratio, we were able to hatch 10 cocks for further testing of transfected sperm. The female chicks and those embryos which died during the experiment were used to estimate the gonadal transfection efficiency. The gonads were isolated and the fluorescence signal of the Venus reporter was detected in 34% of the hatched chicks. Transfected primordial germ cells were verified in 46,6 % of 56 investigated gonads. The fluorescence microscopy data were confirmed by Venus specific PCR. Primordial germ cell transduction was verified by SSEA1 immunohistochemistry. In conclusion, these results suggest that the Sleeping Beauty transposon system is another efficient method for direct in vivo transfection and the production of transgenic chicken.

Keywords: Chicken, In vivo transfection, PGCs, Sleeping Beauty

Genetic characterization and population structure of local Polish and Italian poultry breeds

Abstract ID: 614

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Aims of this study were to estimate and compare the internal variability of 4 ducks and 11 chickens populations, all under plan of conservation, and evaluate their genetic distances from evolutionary perspective. The duck breeds analyzed were the following: Germanata Veneta (AGV) and Mignon (AMG) from Italy; Pekin Krajowy (33 P) and Pomniejszona (2K) from Poland. The 11 chicken breeds were: Miniature Cochin (MCO), Gold Italian (GI), Green Legged Partridge (GLP), Silver Italian (SI), White Leghorn (WL), Ermellinata di Rovigo (ER), Polverara (PL), Padovana (PD), Pèpoi (PP), Robusta Lionata (RL) and Robusta Maculata (RM); 5 of these were reared in Poland. Genetic characterization of the four duck populations was carried out through a panel of 23 microsatellite markers selected from different linkage groups found in literature. The analysis involved~180 individuals for each breeds. An average of 11.36 and 10.74 alleles per locus were identified in duck and in chickens respectively, with fragments length from 68 to 485 bp. The duck and chicken breeds showed good heterozygosity: 0.69 in Polish duck populations, 0.45 for the Italian one and a range between 0.28 and 0.58 in all chickens. Generally, the inbreeding coefficient (F_{is}) was higher for the Italian breeds, AMG and PP in particular (0.13 and 0.18 respectively) with minimum reached by GLP (0.11). The molecular inbreeding (f_{ij}) was higher for the Italian duck breeds (MeanKin=0.37; MeanKinSubp=0.53), and lower for the Polish breeds (MeanKin=0.22; MeanKinSubp=0.29), while in chickens the breed ranged from 0.12 (GI) and 0.18 (PP) evidences limited coancestry. When each Italian duck population was compared with a Polish breed f_{ij} showed intermediate values. The Nei's minimum distances (D_N) and Reynolds distances (D_R) were low between the Polish populations (0.13 and 0.09 respectively); these were associated to AGV (D_N=0.19 and D_R=0.26 for 33P; D_M=0.17 and D_R=0.23 for 2K). Finally AGV was very distant from AMG (D_M=0.26 and D_R=0.32). Hence, the analysis of the population structure revealed a marked differentiation between the Italian populations which had also a great internal homogeneity. Instead, both Polish breeds showed more similarity. This result was confirmed by a factorial analysis. Tomiuk and Loeschcke's D_{TL} genetic distance values shown that the PD and PL breeds are the most related (D_{TL}=0.1) followed by PD and PP (D_{TL}=0.11) while, when considering the "Polish breed", the two Italian derived GI and SI breeds were the most related (D_{TL}=0.31); these data confirm a clear separation between the six Italian chicken breeds and the five "Polish breed". In conclusion, the information obtained from this study represented a useful tool for the monitoring of the conservation activities, which until now has been performed in the good way, leading each population to show its own identity.

Keywords: Genetic characterization, Italy, Local poultry breeds, Poland, Population structure

Genome-wide association analysis of egg production in chickens across the whole laying period

Abstract ID: 626

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Age at first egg (AFE) and egg number (EN) are economically-important traits related to egg production and have aroused great concern in poultry industry. To better understand the genetic architecture of AFE and the longitudinal ENs from 23 to 80 weeks of age which were divided into four stages (EN1, EN2, EN3, EN4), we performed a multivariate genome-wide association study (GWAS) in a population of 1,078 Rhode Island Red hens employing chicken 600K high density SNP arrays. Results indicated that the heritability estimates were high for AFE (0.51) and moderate for ENs (0.14 ~ 0.24). In addition, a genomic region spanning from 117.87Mb to 118.36Mb (~0.49Mb) in GGA1 was detected to be associated with EN and the six genome-wide significant SNPs could accounted 10.77%, 2.89%, 8.65% and 1.32% of phenotypic variance from EN1 to EN4. Four promising genes including *POLA1, PDK3, PCYT1B* and *PRDX4* around the significant SNPs were considered to be candidate genes for egg number. Findings in our study could provide worthy theoretical basis to improve egg production based on marker assisted breeding selection.

Keywords: Egg production; GWAS; Heritability; Candidate genes

Germline chimera production from cryopreserved primordial germ cell lines of a Hungarian indigenous chicken breed

Abstract ID: 123

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The current way of preserving most of the economically important or indigenous chicken breeds is to maintain them in in situ populations, which poses numerous risks, such as epidemics (e.g. avian influenza), environmental disasters or inbreeding. Therefore, preservation of the genome itself is a high priority, although it is faced with difficulties. Semen biobanking lacks the ability to conserve the W chromosome and the mitochondrial DNA, and embryo cryopreservation is not yet established. Nowadays, primordial germ cell (PGC) based biobanking is the most promising solution. In this study, by using the unique characteristics and accessibility of PGCs (they migrate through the vascular system to reach the genital ridges), biobank for an indigenous Hungarian chicken breed – the Partridge colour Hungarian – was established and then tested. Blood samples were collected from each embryo individually, then the isolated blood, containing the PGCs, was cultured in a PGC selective medium. After that, PGC samples were collected for DNA, RNA isolation and immunohistochemistry to characterize the quality of the cultured lines. As a next step, parallel vials were frozen from each PGC line. The cryopreserved samples have been stored in liquid nitrogen since then. To evaluate the freezing process and to prove the functional integrity and migration ability of PGCs, some of the vials were thawed and the cells were injected into recipient embryos. As a first step, the cells were labelled with an in vivo fluorescent dye, thus the migration of the injected cells was followed toward the developing gonads, and the ratio of the colonization was analysed. As a next step, after injection, the eggs were incubated to hatching. The hatched chicks were then kept until maturation and are going to be cross-tested with animals from the donor genotype to examine the germline transmission. During the study, 21 PGC lines from Partridge colour Hungarian chicken were established with a derivation rate 31.1%. The PGC lines were frozen and then successfully thawed with a cell viability of 50%. The preserved cells were capable of colonizing the gonads of the recipient embryos; furthermore, we have 24 adults (13 roosters and 11 hens) which presumably contain the donor PGCs. This is the first demonstration of a successful PGC-based cryopreservation project on a Hungarian indigenous breed and the first initiative in Hungary to establish a biobank based on PGCs. The research was funded by Horizon 2020 (n°677353 IMAGE) and by VEKOP-2.3.2-16-2016-00012.

Keywords: Biobanking, Cryopreservation, Germplasm, Hungarian indigenous breed, Primordial germ cell

Identification and genetic analysis of extreme feather pecking behaviour in laying hens

Abstract ID: 258

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Feather pecking is a serious economic and welfare problem in the domestic fowl. It is usually recorded as number of severe pecks or bouts of pecks delivered towards group mates. This trait follows a Poisson distribution with a mean close to zero. It has recently been shown that the distribution is not homogenous and comprises a sub-population of extreme feather peckers. In a large F2 cross with about 900 individuals, about 20 percent of the hens were clustered to the extreme feather peckers with a mean about ten times larger than the mean of the remaining birds. These results provide strong empirical evidence for the occurrence of a sub-group of extreme feather peckers, which was previously reported in the literature. In the present study we fitted a two component negative binomial distribution to the same data set and clearly confirmed the existence of a sub-group of extreme feather peckers. We estimated the posterior probability of each hen belonging to the extreme feather pecking sub-group and used this probability as observations. The Spearman correlation between these two traits (posterior probability of extreme feather pecking and common feather pecking) was 0.67. The heritability of the hens' posterior probability of extreme feather pecking was 0.14. A genomewide association analysis is in progress to identify trait associated SNPs for these two traits and thus to investigate if they have to some extend a different genetic basis. The results will provide us important information if it comes to a selection against feather pecking behaviour. Further they will also stimulate research on potential differences in motivation of the extreme feather pecking sub-group and the remaining hens.

Keywords: Breeding, Extreme feather pecking, Laying hens

In ovo genotyping of chicken using DNA isolated from allantoic fluid

Abstract ID: 389

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As part of the EU project IMAGE (Innovative Management of Genetic Resources), we want to show how *ex-situ* collections could efficiently be used to recover a rare characteristic from a chicken gene bank collection to upgrade *in-situ* populations. As a model, the introduction of blue egg shell color as a single monogenic dominant trait from a gene bank population into a high performing, white egg layer chicken line will be demonstrated. Two marker-assisted backcross generations followed by an intercross-generation will be generated aiming at a high performing White Leghorn-like line which is homozygous for blue egg shell color. Since we only need males carrying a blue egg shell allele for further breeding of backcross generations and males and females carrying a blue egg shell allele for the intercross generations, we are interested in *in-ovo* sorting of eggs in order to reduce the number of chicks hatched. The objective of the present study is to prove the possibility of *in-ovo* sampling of allantoic fluid for blue egg shell genotype and sex determination in chicken. To achieve this, we aim at the use of an innovative approach by genotyping DNA isolated from hatching eggs at the time of first candling. The causal mutation of dominant blue egg shell color is a 4.2 kb retroviral insertion on chromosome 1 upstream of *SLCO1B3* at 65.22 Mb and can be determined by PCR. We developed a competitive allele-specific PCR (KASP) assay for genotyping the blue egg shell mutation. Using KASP we are able to genotype up to 379 animals in one reaction. Sex determination was done by PCR according to Fridolfsson and Ellegren (1999). On the basis of sequence differences between CHD1-W and CHD1-Z, we also developed a KASP assay for sex determination. First results suggest that DNA of sufficient amount and quality for genotyping can be obtained from allantoic fluid at day 10, and to a lesser degree also at day 6 of incubation. Our results are in concordance with Li *et al.* (2015) who demonstrated the possibility of *in-ovo* sexing of ducks at embryonic stage 28 corresponding to incubation day 6 in chicken. Current experiments focus on the identification of the earliest possible day for sampling and the most suitable method for DNA isolation.

Keywords: Allantoic fluid, Blue egg shell, Chicken, In-ovo genotyping, KASP

Marker genes in poultry of gene pool flock

Abstract ID: 88

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The study was aimed at the identification of poultry carrying marker genes which can be used in modern selection programs. The study was performed on poultry of conserved gene pool flock (150-200 birds per genotype) of Russian poultry breeds. Spangled Orloff chicken were tested and selected for nuclear and mitochondrial markers associated with the resistance to viral diseases, together with the directional selection for the genes related to the exterior parameters: genes of E (e+, ey) and PR loci, Mb, co+. Differentiation of the breed as carriers of e+ and ey genes was enhanced. In previous generation the occurrence of e+ gene was 67.2%; in the last generation (2017) this parameter was 75.0%. Maximal occurrence of ey gene in the selected generation was increased by 22.6% (from 53.6 to 76.2%). The occurrence of controlled molecular markers carrying valuable allele 357 of the microsatellite locus LEI0258 (associated with the resistance to Marek's disease) was found to persist during the breed's conservation. The occurrence of 357/357 genotype in Spangled Orloff population was 48.5%. Day-old chicks of experimental slow-feathering E5 line (Plymouth Rock) were evaluated according to feather phenotype; five groups were selected differing in growth rates of remex and tectrix and their ratio. Crossing of females from these groups with fast-feathering Cornish males led to the different sexing accuracy rates (68.7-100%) in progeny. The lowest sexing accuracy (68.7%) was found in the progeny of the females with similar lengths of well-developed remex and tectrix; this group is excluded from the further selection. The genes modifying the expression of silver and gold genes were identified in Creamy Guinea fowl. Family selection for gene ig (inhibiting fluff color in males and enhancing it in females) allowed for the sexing accuracy at 1 day of age 95%. The selected groups of poultry carrying valuable marker genes can be successfully used in further selection.

Keywords: Chicken, Genes, Guinea fowl, Markers, Polymorphism

Morphological blood indices in turkeys of preserved breeds

Abstract ID: 133

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The number of commercially used breeds, lines and crosses of turkeys is limited. The main attention is paid to the productivity of turkeys of popular crosses of White Broad-Breasted breed and to a lesser extent certain preserved breeds. In addition, only few data are presently available on the blood indices in turkeys of rare genotypes. The studies were performed at the Center for Selection & Genetics "North-Caucasian Zonal Experimental Station for Poultry" on 4-week turkeys of the breeds of gene pool flock included in the "State Register of breeding achievements": Bronze North Caucasian (BNC), White North Caucasian (WNC), Silver North Caucasian (SNC), Black Tikhoretskaya (BT), Moscow White (MW), and Uzbek Fawn (UF). The poults were grown in closed poultry houses on deep litter in compliance with the recommendations concerning zoohygienic conditions, nutrition, and healthcare. Age of 4 weeks was chosen due to the fact that at this age genotype and sex become primary factors influencing the blood indices, and due to higher risk of different diseases related to the immune deficiencies and paratypical factors. The concentration of red blood cells (RBC) in the blood of matured turkeys of all breeds averaged 2.75×10^{12}/L and fell within the normal range (2.5-3.5×10^{12}/L). The difference between the highest RBC concentration in BNC turkeys and the lowest in MW and UF turkeys was 1.1%. The difference between the minimal and maximal leukocyte concentration averaged 18.31×10^{9}/L was 0.98×10^{9}/L or 5.2%. Concentration of hemoglobin fell within the normal range (70-110 g/L). Concentration of hemoglobin in SNC turkeys was higher than in the other five breeds by 0.02-2.32 g/L. According to the rank assessment of these data BNC turkeys had the highest priority, followed by WNC and SNC; MW was assessed as outsider breed. In comparison, the valuation rates (scores from 0 to 5) given by the selectionists to the youths of these breeds were: 1.0 for WNC; 2.0 for MW and SNC; 3.0 for BNC; 4.7 for UF and BT genotypes. It was concluded that in similar rearing conditions circulatory concentrations of RBC, leukocytes, and hemoglobin differed between studied breeds of turkeys, even between closely related genotypes.

Keywords: Blood morphology, Breeds, Turkey

New possibilities in the gene conservation of Hungarian indigenous chicken breeds

Abstract ID: 80

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According to the FAO report, 32% of chicken breeds disappeared or are in critically endangered status. In vivo and in vitro gene conservation systems ensure the survival of this valuable genetic source. Continuous improvement of the applied methods for maintenance of gene banks is indispensable. Nowadays - regarding to avian species – semen freezing is the only practically used preservation method. Cryopreservation of oocytes and embryos is impossible, because of their biophysical traits, therefore the female genome falls out of gene conservation. For the maintenance of it, orthotopic transplantation of early ovary of day old chicks can be a suitable method. However, earlier studies proved that not all breeds are suitable as a recipient for this purpose. The aim of the study was to optimize the donor/recipient pairing and cryopreservation protocol of the early gonads of Hungarian indigenous chicken breeds. The genetic distance of the successful pairings from intensive lines were examined, then native breeds were compared to them. According to the results, donor/recipient combinations were created which could be efficient for transplantation. White Leghorn or Novogen White formed appropriate pairs with Yellow Hungarian, Black and Speckled Transylvanian Naked Neck and Partridge-colour chicken breeds. The implanted native organs adhered (75-80%), and a histological examination proved, that ovaries and testicles are capable to produce gametes. The gonadal tissues of these breeds were cryopreserved by vitrification using a modified method of Wang et al. (2008). The transplanted frozen / thawed gonads were adhered in 72% of Hungarian Partridge colour, 57% in Black and 43% of Speckled Transylvanian Naked Neck and 20% of Yellow Hungarian chicken. In order to check the donor derived progeny raising of hens is in progress now. As the interventions can be carried out under simple circumstances and the recipient hens can produce high number of donor gametes from donor gonads, the method can be a promising tool in the in vitro gene conservation of Hungarian indigenous chicken breeds. The research leading to these results has been conducted as part of the IMAGE project which received funding from the EU's Horizon 2020 Research and Innovation programme under the grant agreement n°677353, as well as it was supported by GÉNNET_21 (VEKOP-2.3.2-16-2016-00012 / Hungary).

Keywords: Chicken, Gene conservation, Gonadal cryopreservation, Gonadal grafting

Optimization of live bodyweight and mortality during the selection of preparental lines of Hisex Brown chicken

Abstract ID: 124

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The study was aimed at the determination of optimal live bodyweight (LBW) and mortality ranges in growing and adult chicken of purebred lines of Hisex Brown hybrid layers (A, B are lines of hybrid paternal form, C, D – for maternal form) selected by breeding farm "Sverdlovsky" since 2014. For individual evaluation of productivity the birds were kept in individual cages within cage batteries; the productive performance was estimated at 16, 35, 52 and 65 weeks of age. Phenotypic correlations between LBW and productive traits were calculated to define the normal range of LBW. Uniformity in LBW within a line was assessed as the percentage of birds falling within a range of average +/- 10%. Average maturity age in hens of line A was significantly ($P<0.001$) lower by 11.5 days compared to line B; the difference between lines C and B was 7.1 days ($P<0.001$). The best average egg production was found in line A (302.2 eggs per hen). Average egg production in lines A and B was higher by 5.3-8.8% and 3.8-7.4% compared to lines C and D, respectively ($P<0.001$). Egg weight at 52 weeks of age in lines C and D was higher by 1.0-1.8 and 1.9-2.7%, respectively, compared to lines A and B ($P<0.05$). Uniformity coefficient for LBW in males of line A was 73.7%, in females 83.2%. Uniformity coefficients for LBW in females of lines B, C, and D were higher by 3.5; 1.0 and 1.2% compared to line A. The best uniformity in males was found in line D (90.1%). Correlation between maturity age and LBW was modest in all lines (negative in line A). Correlation of LBW and egg production was negative in lines B, C, and D. Significant positive correlation between LBW and egg weight was found in all lines. LBW in females and males at all ages was slightly higher in lines A and B in compare to lines C and D. Selection of all lines for LBW improved uniformity coefficients for LBW; the correlations between LBW and productive traits were neutral or positive. Mortality levels in growing and adult birds selected for normalized LBW fell within the range 2-3%. It was concluded that productive performance in selected purebred lines of Hisex Brown hybrid layers was higher compared to non-selected. Normal ranges of LBW and mortality in growing and adult chicken determined in the study will be considered in further breeding and test trials.

Keywords: Layer chicken, Live bodyweight, Mortality, Productivity, Reproductive performance

The effect of plumage colour and serotonin transporter gene polymorphisms on feather pecking behaviour in laying hens

Abstract ID: 157

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Feather pecking in laying hens is a serious welfare problem and the motivation for this behaviour is still unclear. Only few studies focused on the role of the victims and it seems that some birds attract more feather pecking than others. In the present study, feather pecking was recorded in 216 hens from three experimental lines bred from a commercial medium heavy brown hybrid. These lines differed in genotype of a single nucleotide polymorphism (SNP) in the serotonin transporter (SERT) gene, being wildtype (W/W), mutants (D/D) or heterozygotes (W/D). Through several years of crossings of this commercial hybrid, however, a range of colour phenotypes had emerged and these were categorized in beige (white with some reddish spots, n=31), brown (n=100), grey (white with dark grey stripes, n=19) and white (n=66). The number of feather pecks (severe or gentle) and aggressive pecks given and received were recorded on an individual basis twice a day (1 h/morning and 1 h/afternoon) for 23 days at the age of 32 to 35 weeks. Plumage condition was scored as well. Severe feather pecking was the predominant type of pecking (on average 7.8, 1.3 and 3.6 severe, gentle and aggressive pecks given per hen, respectively) and therefore only severe feather pecking is presented here. Colour morphology did not affect performance of severe feather pecking but genotype did. W/D hens pecked more and lsmeans for pecking per bird were 12.6, 8.1 and 3.5 for W/D, W/W and D/D hens, respectively (P<0.001). There were significant effects of colour morph as well as genotype on number of pecks received. Beige (3.26) and grey hens (2.47) received more pecks as compared with brown (1.66) and white (1.24) hens (P < 0.05). WW hens received more pecks (1.13, 1.63 and 2.77 for D/D, W/D and W/W, respectively) (P < 0.001). Plumage condition was not affected by genotype but colour morph. Brown hens had the worst scores (ChiSquare=81.4, P<0.001). In conclusion, beige and grey plumage colour attract more pecking than white and brown in contrast to earlier reports. This suggests that plumage with a combination of colours (speckled) attracts more pecking than even colours. On the other hand, the darkest hens had the most feather damage. Further, it was a new finding that hens carrying the W/D genotype of the SERT gene polymorphism pecked more and that hens carrying the W/W genotype received more severe feather pecking, and this needs further investigation.

Keywords: Feather pecking, Genetic crossing, Plumage colour, Plumage condition, Welfare

The transgenic technologies improving the efficiency of poultry production

Abstract ID: 87

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Practical applications of the transgenesis in poultry can be related to the improvements in productivity and to the development of lines resistible to viral diseases. However, technically simple and effective technologies for the production of genetically modified poultry lines are still to be developed. Authors developed a method for the induction of the transgenicity in poultry by microinjections of foreign DNA into the oocytes in chicken and quails. The method is based on the surgical operation to gain access to an oocyte for subsequent DNA microinjection and natural formation of tertiary oocyte coats in the reproductive tract of a female. This research has been commenced back in 1990s on White Leghorn chicken and Estonian quails. It was found that the ovulation on experimental birds occurs in 20-25 min after the oviposition. After ca. 5 min an ovulated oocyte enters the infundibulum where the fertilization occurs. Freshly fertilized oocytes in the hens were accessed via laparotomy in 15-20 min after the oviposition with preliminary application of an anesthetic and a muscle relaxant. Microinjection to the blastodisc region was performed via the transparent oviduct wall using 2-4 µm micropipettes; DNA injection volumes in different experiments varied from 200 to 1000 pL. Eggs laid on the next day after the microinjections were incubated, the poults hatched were raised. The transgenicity in the progeny was detected by blot-hybridization and PCR; the expression of the transgenes was determined using ELISA analysis of blood serum. The effectiveness of the induction of transgenesis varied from 10 to 25% in different experiments. A substantial part of transgenes obtained were mosaic. This technology was applied in the production of transgenic chicken with human genes of β-galactosidase (pCMVlacZ), somatotropin (pSMThGH9), and β-interferon (pMT-hIFNb1). Microinjection of gene construct pMTbGH (2×att) into the oocytes of quail eggs resulted in the population of transgenic Estonian quails carrying the bovine somatotropin gene. The productive and reproductive performance in this transgenic line was assessed in 33 generations. Average bodyweight in females and average egg weight in transgenic line were higher in compare to normal Estonian quails by 5-15 and 10-20%, respectively.

Keywords: DNA microinjection, Hens, Quails, Transgenesis, Zygote

The use of infertile interspecific hybrids for a novel model of PGC reintroduction applicable in gene preservation for poultry

Abstract ID: 65

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Related to the gene preservation activities of our Institute, the aim of our research was to produce an infertile interspecific hybrid (recipient) which is able to receive primordial germ cells (PGCs) from native poultry breed (donor) and according to our expectations; this genotype may appear in the offsprings of hybrids. To achieve our goals, the first step was crossing female Hungarian guineafowls with Hungarian yellow roosters by artificial insemination. The following year we reversed the crossing procedure with Hungarian yellow hens and guineafowl males. During the research we were investigating the hatching time, fertility, the phenotype of dead embryos and the incidence of chromosomal abnormalities. Development of the gonads of hybrids was being observed during the incubation and the hatched hybrids were raised until maturity. We described the phenotype of the hybrids and 6 hybrids were sacrificed every two weeks between the 16th and 30th week of growth in order to document the development of their gonads. We sampled the gonads for histological and genetic examination in order to prove that the individuals are hybrids and infertile. In the first year the hybrids hatched between the 21st and 27th day of incubation. 31.7% of the eggs were infertile; the ratio of early embryonic death was 29.58% and 13.3% of the embryos died between the 1st and the 5th day of incubation. The ratio of hatched hybrids was 6.65% and 10% of the embryos was viable, but could not break the eggshell of guineafowl. During the investigation of chromosomes 2.7% of the samples showed abnormalities (mosaicism or aneuploidy), but this ratio is average compared to previous literary data. As the result of the investigation of chromosomes, 56.76% of the embryos was male, 43.24% was female. Three kinds of phenotype were observed on the hatched hybrids. In the second year, the reversed crossing procedure (Hungarian yellow hens with Hungarian guineafowl males) was unsuccessful due to the high rate of infertile eggs (98.4%). From 701 incubated eggs only one hatched (0.14%). Based on our results, the conclusion is that these interspecific hybrids (female Hungarian guineafowls with Hungarian yellow roosters) may be suitable to receive primordial germ cells (PGCs). According to our plans, in the next steps of this research, first we will inject fluorescent labeled cells, then primordial germ cells of a native chicken breed into 3 days old hybrid embryos.

Keywords: Domestic fowl, Gene preservation, Guinea fowl, Hybrid, Primordial germ cells

Verification of effect on the growth rate in Hinai-jidori chickens (Japanese meat-type chicken) by selection using the CCKAR g.420 C < A polymorphism

Abstract ID: 224

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We previously reported the significant association between a single-nucleotide polymorphism (SNP; g.420 C < A) in the cholecystokinin type A receptor gene (CCKAR) and growth traits in the Hinai-dori breed which is a breed of indigenous chicken in Japan, and results showed that the A allele had a superior effect on the growth traits compared to the C allele. Moreover, we demonstrated that the SNP improved the growth rate in the low-growth line of the Hinai-dori breed. Furthermore, we observed that the A allele had a superior effect on the growth traits compared to the C allele in the Hinai-jidori chickens, a cross between Hinai-dori sires and Rhode Island Red dams, has been commercialized as a popular brand meat-type chicken in Japan. In the present study, we verified the effect of the growth rate in Hinai-jidori chickens by selection using the CCKAR g.420 C < A polymorphism. A/A individuals in the Hinai-dori breed and Rhode Island Red were selected by genotyping of the SNP, respectively. A/A Hinai-jidori individuals were produced by mating of Hinai-dori sires and Rhode Island Red dams (improved group). The chicks of improved group and conventional group were hatched on the same day. Thirty chicks of improved group and sixty-three chicks of conventional group were randomly divided into four pens at 4 weeks of age, and raised in the poultry shed until 23 weeks of age. The body weight of each individual was measured at 4, 14, and 23 weeks of age, and the average daily gain was calculated from the body weights. The data showed that the body weight at 23 weeks of age, the average daily gain between 14 and 23 weeks of age, and between 4 and 23 weeks of age in improved group were significantly greater than those of conventional group ($P < 0.05$). Our findings suggest that the A allele of the g.420 SNP of CCKAR improves the growth rate of the Hinai-jidori chickens and the SNP could be a candidate marker to improve the growth rate in selective breeding of meat-type chickens.

Keywords: Body weight, Cholecystokinin type A receptor gene, Growth rate, Hinaji-jidori chicken, Single-nucleotide polymorphism

Effect of citric acid addition on functional and phisical properties of pasteurisated liquid whole eggs during 4 weeks of storage

Abstract ID: 641

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Eggs and egg products have important and irreplaceable part in human nutrition because of high content of protein, valuable nutrients, wide range of applications, relatively low cost and high availability in most countries. Traditionally, eggs are sold as shell eggs, but their consumption under the form of egg products has increased, especially pasteurized liquid whole eggs (LWE). LWE have many properties such as gelling, emulsifying, foaming, coloring and flavoring food. For this reason knowledge of functional properties of LWE is important. The process of pasteurization has influence on the microbiological quality of LWE. Also, citric acid is often added to LWE which is a natural preservative and has effect on pH decrease and helps preserve the color of the egg products. So the aim of this study was to examine influence of citric acid addition (0, 300, 400 and 500 mg/L) on functional and physical properties of LWE during 4 weeks of storage. The pH, color (L*a*b*), protein solubility, foaming properties (foaming capacity and foam stability) and emulsifying properties (emulsifying capacity and the percentage of emulsion) were measured. The addition of citric acid influenced pH value and also the addition of 300 mg/L of citric acid had a positive effect on the preservation of protein solubility throughout the storage period. Addition of citric acid and storage statistical influenced L * and b * values. L * value increases with addition of citric acid and weeks of storage while a * value was affected by weeks of storage. Best foaming capacity and foam stability had samples with addition of 300 mg/L of citric acid while the lowest foam stability had samples with addition of 500 mg/L. The emulsifying capacity was the highest in samples with addition of 400 mg/L while the percentage of emulsion was the highest in samples with 300 mg/L of citric acid. Although addition of 500 mg/L citric acid gave the best bacteria reduction, functional properties were significantly changed. Addition of 300 mg/L gave satisfactorily bacteria reduction without affecting functional properties.

Keywords: Citric acid, Emulsifying, Foaming, Liquid whole eggs, Protein solubility

Effect of maize hybrid antioxidant potential on egg yolk oxidative stability – correlation analysis

Abstract ID: 542

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Lipid oxidation during food production and processing is of main concern today. Due to the egg shell stability, severe oxidation in egg yolk will not occur before processing. These conditions are usually tested in induced oxidation methods. Major preventive factors of lipid oxidation are natural or synthetics antioxidants. Yellow maize grain, paramount cereal in poultry nutrition, is the cereal offering significant concentrations of natural antioxidants: carotenoids, vitamin E, and phenols. The objective of the study was to explore the relationship between distinctive amounts of antioxidants found in maize hybrids and oxidative stability of eggs during induced oxidation.

In total 90 hybrid Tetra-SL lying hens were fed diets based on five different maize hybrids (Bc 572, Mejaš, Kekec, Pajdaš and Riđan) while control diet contained hybrid of unknown origin. The hens were grouped, three in one cage, in a random block design with five repetitions per hybrid. Levels of carotenoids and tocols in hexane extracts were determined using HPLC while bound, free and total phenols and flavonoids were determined spectrophotometrically using the Folin-Ciocalteu reagent (galic acid equivalents) and Al-complexation reaction (catechin equivalents), respectively. The Fe-induced lipid oxidation measured as TBARS was used to determine oxidative stability of egg yolks.

Egg yolk stability was significantly affected by maize hybrid (*P*<0.001), which differed in contents of all evaluated natural antioxidants (*P*<0.001). Their contents varied as follows, all on DM basis: lutein (6.21-12.27 µg/g), zeaxanthin (9.63-16.66 µg/g), β-kryptoxanthin (1.22-2.79 µg/g), β-carotene (1.87-2.90 µg/g), total tocols (69.10-89.76 µg/g), bound phenols (3575-4824 µg/g), free phenols (959-1289 µg/g), total phenols (4620-5784 µg/g), bound flavonoids (639.4-835.4 µg/g), free flavonoids (142.2-280.0 µg/g) and total flavonoids (821.8-978.2 µg/g). The correlation analysis showed negative correlation between concentration of TBARS substances and contents of lutein (*P*<0.05), total tocols (*P*<0.05), free phenols (*P*<0.01), bound phenols (*P*<0.001), total phenols (*P*<0.001), free flavonoids (*P*<0.001) and total flavonoids (*P*<0.001). Results clearly show that antioxidant properties of maize hybrids define egg yolk oxidative stability. In order to minimise expenses and effects of possible lipid oxidation during processing of shelled eggs animal nutritionists should choose maize hybrids not based only on their energy value and should account them for their antioxidant potential.

Keywords: Antioxidants, Egg yolk, Maize hybrids, Oxidative stability

Egg parameters of two Hungarian indigenous chicken breeds

Abstract ID: 219

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Indigenous chicken breeds could provide a response to the rapidly changing consumerpreferences, furthermore theycould be suitable for production in sustainable agriculture.In the aspect of the gene preservation of the breeds, the exact description of their phenotypical traits and production parameters is needed. Our aim was to investigate the egg quality (weight, index, shell thickness, shell breakage strength) of Speckled Hungarian chicken and Yellow Hungarian chicken. Besides, we establishedtwo crossbred genotypes: Speckled Hungarian chicken × Tetra Harco and Yellow Hungarian chicken × Tetra Harco. Every day 15 eggs were randomly selected for weight and index measurement (n=945). In the case of shell thickness and shell breakage, the eggs were measured once a week on the same days (n=270). The egg shell breaking strength was measured by TA®XT PLUS TextureAnalyser (with the flat compression breakage force measurement). The egg shell thickness was detected by digital micrometer. Correlations ofthe egg parameters of Speckled Hungarian chickens and Yellow Hungarian chickenswerecompared.The crossbred genotypes were compared to the purebred genotypes. In case of thickness and shell breakage the Speckled Hungarian chickens' eggs (0.34 mm, 4.7 kg), did not differ from the eggs of Speckled Hungarian chicken × Tetra Harco (0.33 mm, 4.5 kg), neither the Yellow Hungarian chickens' eggs (0.32 mm, 4.0 kg) from the eggs of Yellow Hungarian chicken × Tetra Harco (0.33 mm, 4.2 kg). The egg weight of the crossbred genotypes was more favourable, than the purebred genotypes. The Speckled Hungarian chicken (62.47 g) and Yellow Hungarian chicken (62.41 g) produced M – sized eggs and the crossbred genotypes produced (66.61 g, 67.65 g) L – sized eggs. The Speckled Hungarian chicken had a significantly higher egg index (1.33) than the Speckled Hungarian chicken × Tetra Harco (1.30). Positive correlation (r=0.6) was found between the shell thickness and the shell breakage strength of Yellow Hungarian chicken.Correlations werenot observable in other egg parameters. According to the results,the weight of the eggs of indigenous breeds compared (M) are corresponding to the Hungarian consumer demand but the eggs of the crossbred genotypes (L) have more favourable sales. During correlation-analysis, our results were corresponding to the literature in both breeds.

Keywords: Crossing, Egg, Egg shell, Speckled Hungarian, Yellow Hungarian

Evaluation of quail eggs quality as affected by storage period, temperature and egg size

Abstract ID: 146

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This study was conducted to evaluate the effect of different storage time, temperature, egg weight and their interaction on the evaluation of quality parameters of the quail eggs, obtained from different small-scale farms in Prishtina region. At the beginning of the experiment, the eggs were weighed, marked and divided in the groups by size (≤10, 10.01-1.00, 11.0-12.00 and 12.0-13.00). The eggs were stored for 7, 14, 21, 28 days at two temperatures (4-6 and 20-24 ^{0}C). At the end of each storage period all the eggs were weighed individually to calculate egg weight loss. Loss of egg weight was significantly ($P<0.001$) influenced by storage duration. High differences on the egg weight loss also were observed by storage temperature ($P<0.001$) and by eggs size group ($P<0.001$). Storage duration significantly affected albumen weight (g and %) and index ($P=0.075$, $P<0.001$, and $P<0.0001$), yolk weight (g and %) and index ($P=0.0254$, $P<0.001$, and $P<0.001$), egg yolk color ($P<0.001$), ratio yolk/albumen ($P<0.001$), Haugh Unit ($P<0.001$), albumen pH ($P<0.001$), yolk pH ($P<0.001$) and eggshell thickness ($P<0.001$). Albumen weight, albumen percentage, yolk index and yolk pH significantly decreased with increasing duration of storage ($P<0.001$, $P=0.0291$, $P<0.001$, $P<0.001$ respectively). Albumen and yolk percentage, yolk index and color, ratio yolk/albumen, albumen pH, yolk pH, eggshell percentage and eggshell thickness were not significantly affected by egg weight group size. Based on the findings in this study, in order to keep egg quality parameters mainly Haugh Unit and to avoid losses, quail eggs can be stored up to 28 days without missing the freshness.

Keywords: Egg quality, Quail eggs, Storage duration, Storage temperature

Egg production in China

Abstract ID: 214

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China produces more than 40% of all the eggs in the world. The average annual growth rate of egg production was approximately 0.6 million tonnes per year from 2000 to 2016. In 2016, the total eggs produced in China reached a peak of 31 million tonnes. Eggs contribute approximately 6% of the protein supply in the diets of Chinese citizens. Egg consumption in rural families is still much lower than that in urban families (5.9 kg eggs per capita vs. 10.5 kg eggs per capita in 2012), but with the rapid increase in urbanization it is expected that the future demand for eggs will continue to increase. In the five years since 2012, the annual revenue for the poultry industry has grown at an average rate of 8.7% per annum. In the last decade, egg production costs at farm level have generally changed with the change in layer feed prices. Both the egg production cost and the layer feed price reached a peak in 2013. Average cost of the egg production was 8.7 Yuan/kg ($1.31 /kg) and the average feed price was 2350 Yuan/ tonne ($352 /tonne). Variation in feed costs remain the major, but not the only, influence on production costs. Ten Chinese provinces produces 80% of the national eggs. In order to meet the consumer's demands in the whole nation there is a substantial movement of eggs around the country. Intensification of egg production methods have occurred throughout the country. In 2008, 43% of all eggs produced were from flocks with sizes less than 2000 hens, but in 2014 this size of egg producer only supplied 4% of the total eggs produced in China. Eggs are primarily produced in cage systems regardless of the size of the enterprise. Ninety percent of eggs are produced from cage systems with 9% in free-range systems and 1% in barn systems. Chinese consumers are becoming more focused on the quality and safety of eggs. Therefore the future developments in the egg production market may concentrate on egg quality, safety and traceability of eggs.

Keywords: China, Egg production, Laying hens

Influence of lycopene on malondialdehides, fatty acid profile and sensory and texture properties of fresh and stored eggs, using rapeseed oil in laying hens nutrition

Abstract ID: 477

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The trial was conducted to investigate the effect of rapeseed oil and lycopene on laying hens productivity, MDA (malondialdehydes), fatty acid concentration and texture properties of eggs. A total of 40 laying hens, 30 weeks old, were assigned into two groups. Control group was with a standard compound feed and supplemented with 4.5 % rapeseed oil, experimental group was supplemented with 4.5 % rapeseed oil + 25 g/kg of lycopene. The laying hens received 125 g of compound feed per day. MDA concentration in the egg yolk were determined by high performance liquid chromatography, with a high pressure gradient HPLC system (Varian ProStar). Fatty acids concentration were determine with gas chromatograph Shimadzu GC -2010 Plus. For the egg sample for texture analysis was used Shafer et al. (1998) method. Statistical Analysis. The results of the experiment were analysed using the 1-way ANOVA test, and significant differences between groups were determined by Duncan's multiple range test. Statistica 8.0. for Windows™ software was used. Differences were considered significant at $P<0.05$. Statistically significant results on laying hens productivity were not determined. Statistically significant results were get on these fatty acids concentration in the egg yolk: MUFA-increased 2.55%, n-3 – 1.57%, IP (index of peroxidability) -1.14%, decreased –n-6-7.66 compared to the control group ($P<0.05$). The MDA concentration in fresh and storage at 28 days had tendency to decreased 40 and 35 % respectively ($P<0.05$) compared to the control group. Used feed additives did not had effect on texture properties of egg, just egg yolk color of fresh egg increased by 2.09 times and stored at 28 days – 1.61 times compared to control group ($P<0.05$). The results of this study clearly demonstrate that supplementation of lycopene and rapeseed oil had effect on MUFA, n-3, IP and egg yolk color intensity.

Keywords: Fatty acids, Lycopene, MDA, Rapeseed oil, Texture properties of egg

Influence of organic acid mixture on laying hens productivity, egg quality parameters and sensory properties

Abstract ID: 566

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The aim of this study was to investigate the effects of organic acid mixture on laying hens productivity, egg quality parameters and sensory properties of eggs. A total of 60 laying hens, 30 weeks old, were assigned to three experimental groups and fed experimental diets for 8 weeks. Control group was fed a standard compound feed, experimental group I – was supplemented with 0.25 kg/t of organic acid mixture, experimental group II – 0.5 kg/t of organic acid mixture. The laying hens received 125 g of compound feed per day. Egg weight, albumen high, Haugh unit, intensity of egg yolk color were established by multifunctional automatic egg characteristics analyzer „Egg Multi-Tester EMT-5200“, hardness of eggshell – by „Egg Shell Force Gauge MODEL-II“ device, and thickness of eggshell – by electronic micrometer „Mitutoyo“. A sensory panel for the descriptive analysis consisted of 9 assessors experienced in sensory evaluation of different food products. The assessors were selected and trained according to the standards. The sensory evaluation was performed according to a standardized sensory descriptive method.The results of the experiment were analysed using the 1-way ANOVA test, and significant differences between groups were determined by Duncan's multiple range test. Statistica 8.0. for WindowsTM software was used. Differences were considered significant for $P<0.05$. The results showed that on laying hens productivity parameter organic acid mixture did not have any statistical significant effect. Eggshell thickness decreased by 8 % ($P<0.05$) when compound feed was supplemented with 0.5kg/t of organic acid mixture. Albumen hardness decreased by 15 % ($P<0.05$), in experimental group I when eggs were stored for 30 days. In conclusion laying hens diet may be supplemented with organic acids without any negative effect on eggs quality and sensory parameters.

Keywords: Egg quality, Laying hens, Sensory properties

Methods of surface disinfection of eggs prior to incubation

Abstract ID: 160

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Most modern poultry farms are integrated enterprises with tremendous flocks concentrated on the restricted area. Incubatory is one of the most critical areas within the farms since microorganisms can proliferate during the incubation, penetrate the eggshell, and contaminate the embryos with subsequent decreases in hatchability of eggs and livability of hatched chicks. Disinfection of premises, incubators, and eggs on the farms involves treatments with different disinfectants which can be unsafe for poultry, personnel, and the environment (formaldehyde, ozone, etc.). A mixture of active substances in anolyte ANK (Russia) inhibit bacterial growth and prevent adaptation to its biocidal effect while low total concentration of active oxygen and chlorine guarantees its safety for the personnel and the environment even after long-term application. The efficiency of electrochemically activated solutions as disinfectants was earlier proved by many authors. Prokopenko et al. (2015) recently tested anolyte "ANK Super" as a disinfectant for the eggs. The effects of disinfection of eggs prior to incubation with anolyte "ANK Super" on egg hatchability and chick quality were studied. Eggs from control group 1 were treated with large-drop formaldehyde aerosol; experimental groups were treated with ANK Super: aerosol for group 2 was produced by "APA" device (large-drop aerosol), for group 3 by electrogenerator 2610 (small-drop aerosol, or "cold fog"). All aerosols were pulverized on eggs for 10 min; after 20 min of exposure ventilation was switched on for 30 min to dry the eggshell. Hatchability of eggs in ANK-treated groups was higher by 5.0 and 2.7% in groups 2 and 3 in compare to control; hatch of chicks higher by 3.7 and 2.5%, the percentage of high-quality chicks higher by 2.14 and 7.14%. At 2 weeks of age live bodyweight in chicks from ANK-treated eggs was higher by 5.18 and 4.38% in compare to control; mortality level lower by 0.5 and 1.0%. It was concluded that treatment of eggs with the aerosols of new disinfectant anolyte ANK Super improves egg hatchability, hatch and quality of chicks.

Keywords: Anolyte ANK Super, Disinfection, Hatchability, Incubation eggs, Live bodyweight

New functional egg products based on melange and enriched with macro- and micronutrients

Abstract ID: 120

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An egg can be a concentrate of nutraceuticals maintaining consumers' health and decreasing the risk of certain diseases; the eggs are an important part of balanced human diet. The aim of our study was to develop a range of new functional egg products based on coagulated melange and enriched with calcium, iodine, and zinc. The study on the effects of coagulation regime on pH of the coagulated melange allowed the estimation of coagulation temperature further optimized using the data on the influence of final coagulation temperature on the yield of coagulated product. Coagulated melange is rich in polyunsaturated fatty acids (PUFAs; 20.86% of total fatty acids); the decrease in concentration of phospholipids in coagulated melange in compare to initial melange was acceptable (5.2% in coagulated and 5.5% in intact melange). The technologies of functional foodstuffs (FFSs) based on the melange involve biofortification of eggs with certain essential nutrients (ENs) via hen nutrition with subsequent enrichment with other ENs during the stages of further egg processing. The eggs cannot be effectively biofortified with calcium and zinc in sufficient amounts and hence the eggs were biofortified with ω-3 PUFAs, vitamin E, and selenium; the melange obtained was additionally supplemented with calcium and iodine at the stage of soft acid-salt hydrolysis; at the stage of production of final egg products an organic form of zinc was added. The parameters of the processing were determined ensuring contents of the ENs in amounts corresponding to no less than 20% of daily requirements and in a bioavailable form. The supplementation with ENs was performed at the stage of thermal treatment of acidified melange; mineral eggshell-based additive was used as a calcium source, dried laminaria powder as iodine source. These ENs were evenly distributed within the products; no off-tastes in the final products were found. The increase in final temperature of coagulation was found to increase the yield of coagulated product and the content of calcium in it while the content of iodine and iodine losses were not influenced. This multi-stage approach to the enrichment of egg products with essential macro- and micronutrients favors sufficient conservation of the latter within the final egg products. The final product – enriched coagulated melange – contains moisture 69.2%, fat 11.3%, protein 14.4%, Ca 399.4 mg/100 g, iodine 0.258 mg/100 g, zinc 2.5 mg/100 g; ω-6/ω-3 PUFA ratio is 2.0.
The study was financed by Russian Science Foundation, grant 16-16-04047.

Keywords: Chicken eggs, Coagulation, Enrichment, Functional foodstuffs, Phospholipids

Our experiences in enriching table eggs with n-3 PUFA

Abstract ID: 63

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Eggs are a good source of nutrients, such as protein, vitamins and minerals. By modifying laying hens' feed, it is possible to influence the content and profile of fatty acids in eggs, aiming to increase the n-3 polyunsaturated fatty acids (PUFA) and to enable production of eggs with increased content of desirable functional ingredients. Such eggs represent a functional foodstuff and their consumption has health benefits. Functional ingredients or nutricines, such as n-3 PUFA, are efficient in protection against chronic diseases (arthritis, psoriasis, Crohn's disease), as well as in prevention of cardiovascular diseases. Omega-3 fatty acids are polyunsaturated fatty acids with a double bond on the n-3 position, i.e. on the third C atom. In the human nutrition, the most important n-3 fatty acids are α-linolenic (ALA, C18:3), eicosapentaenoic (EPA, C20:5) and docosahexaenoic (DHA, C22:6). ALA is a precursor of long-chain PUFA. Since conventional table eggs contain high portion of n-6 PUFA and low portion of n-3 PUFA, which affects the unfavorable n-6/n-3 PUFA ratio, laying hens' feed is supplemented with forages or mixtures that have a good source of n-3 PUFA. Thus, different oils, such as linseed, rapeseed, fish and algae oils, as well as their combinations, supplemented to laying hens feed significantly increase the portion of total n-3 PUFA and reduce the portion of n-6 PUFA in egg yolks. The results of our researches show that meal composition affects the FA profile in eggs. ALA in egg yolk may increase from 1.9 to 4.4 times, EPA from 3.3 to 7 times, and DHA from 1.7 to 5.5 times. Total n-3 PUFAs can be increased from 2.2 to 2.8 times. At the same time, the ratio of n-6 / n-3 PUFAs is reduced. Supplementation of oils may have negative effects on organoleptic properties of eggs (odor and taste), which occur because of the increased n-3 PUFA oxidation due to higher number of double bonds. For this reason, there are organic or synthetic antioxidants (Se, carotenoids, vitamin E) supplemented to laying hens' feed to prevent unwanted oxidative processes in eggs, as well as the occurrence of undesirable taste and odor. In this way, produced eggs have improved nutritional value and extended shelf life. Our researches have confirmed the efficient enrichment of eggs with n-3 PUFA by supplementing linseed, rapeseed, soybean and fish oils to laying hens' feed. Individual concentrations of ALA, EPA and DHA depend on the composition of forage (fat) in laying hens' diet. Vegetable oils (linseed, rapeseed oils) affect the increase of ALA, while the fish and algae oils affect the increase of EPA and DHA in egg yolks.

Keywords: DHA, EPA, Egg, Fish oil, Vegetable oils

Production of outdoor and equol enriched eggs for the benefit of human health

Abstract ID: 563

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Equol is a bacterial metabolite belonging to the family of polyphenols, arising from transformation of specific isoflavones (daidzein, formononetin). This compound would present positive effects on human health by improving prevention or treatment of some diseases like hormonodependant cancers, cardiovascular diseases or osteoporosis. However a small part of western population is able to produce it. As daily and high quality product, egg could be a good way to provide equol in our diet. The aim of this study was to evaluate the production of outdoor and equol enriched eggs in a field trial. Laying hens had access to outdoor areas covered by different plant species rich or poor in isoflavones. Four modalities (repeated twice) were tested: ray grass (control), red clover (rich in isoflavones), chicory (palatable) and white clover (very common). Vegetation cover was maintained in a vegetative state thanks to regular cutting but was subject to climatic variations. In addition to plants prairie laying hens received a balanced diet. Eight coops were used with 15 laying hens from June to October 2017. Eggs were counted and weighed daily. Grasses and eggs were sampled each fortnight to evaluate seasonal evolution of isoflavones and equol concentrations. Visual observations of laying hens indicate an exploration of the whole outdoor area and ingestion of vegetation cover. There is not a significant difference with eggs production in function of plants prairie species. First results showed that equol was more concentrated in egg yolk with red clover allowance. Chemical analyses are still in progress and final results will be available in a couple of weeks. In addition to health benefits, this work promotes outdoor production which responds to consumers' expectations for productions with higher animal welfare and environmental standards.

Keywords: Egg, Equol, Outdoor production, Red clover

Quality of omega-3 eggs enriched with lutein

Abstract ID: 59

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The aim of this study was to determine how lutein addition and storage time influence indicators of external and internal quality of omega-3 eggs. The study was conducted on a total of 150 hens which were divided into control (C - without the addition of lutein) and two experimental groups (E1 - added 200 mg/kg of lutein and E2 - added 400 mg/kg of lutein in the mixture). Experiment lasted for 5 weeks. Indicators of egg quality were analyzed in two terms, on fresh eggs (one day after collection) and on eggs stored for 28 days in a refrigerator. In both terms, 20 eggs from each group were analyzed. The lutein content was determined on eggs (n=8 per group) collected in the third and fifth week of the experiment. Statistical analysis of results showed a significant effect ($P<0.001$) of lutein addition on content of lutein in egg yolks. There was 20.04 µg lutein/g yolk in control group while in E1 and E2 groups values were 108.24 and 118.64 µg lutein/g yolk, respectively. Storage time significantly influenced egg weight and eggshell thickness, while lutein addition did not have influence on indicators of external quality of eggs. Highest egg weight, both for fresh and stored eggs, was recorded in C group, while stored eggs in all groups were lighter compared to fresh eggs. All indicators of internal egg quality were significantly affected by storage time ($P<0.001$). Albumen weight, albumen height and Haugh units decreased with time of storage, while the yolk weight and pH of yolk and albumen increased. Addition of lutein caused higher pH albumen values in fresh eggs ($P<0.001$). There was significant influence of both lutein and storage time ($P<0.001$) on the color of egg yolks. Color of fresh yolks increased from 12.90 (C group) to 13.75 with the addition of 200 mg/kg lutein, while further addition of lutein did not affect the increase in egg yolk color. Yolk color of stored egg changed from 13.05 (C) to 14.10 (E1) and 14.40 (E2). There was more intense yolk color in stored eggs in relation to fresh eggs in all three experimental groups, although the difference was significant only in the E2 group. Based on the obtained results, it can be concluded that the addition of lutein significantly affects egg yolk color while other indicators of internal quality of omega-3 eggs were significantly influenced by storage time.

Keywords: Eggs, Lutein, Omega-3, Quality, Storage

Quality of table eggs from different housing systems

Abstract ID: 170

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One of the most important factors that affect egg quality traits is the production system. In recent years consumers become aware of the strong connection between food and their health, and this one of the reasons for the increase of their interest to so-called "healthy food". Many consumers consider the free range and organic eggs healthier compared to the eggs produced in cages. This attitude is usually based on the belief that there is a significant difference in their chemical composition. Therefore, the aim of this work was to determine the chemical composition and the internal and external quality of eggs produced in conventional cages, free range and organic system that can be found on the market. Samples of 30 eggs were taken from three different production systems: cages, free range or organic system. Egg weight was measured on a precision scale. Shell breaking force was determined by Egg Force Reader (Orka Food Technology Ltd, Israel). Yolk color was determined using the Roche yolk color fan. Albumen height was measured with a tripod micrometer. Protein content in yolk and albumen was determined by Kjeldahl method and fat content was determined using the Soxlet method. The results showed a significant infuence of housing systems on protein content of eggs ($P<0.01$). Higher protein content were determined in organic (11.10% in albumen and 17.04% in yolk) and free range eggs (11.11% in albumen and 16.58 % in yolk) compared to eggs from conventional cages (9.66% in albumen and 15.88% in yolk). The differences in fat content were not significant. Regarding the egg quality traits, yolks form organic eggs were much paler (5.69 points) compared to other two systems (14.9 in cages and 14.24 free range). This can be explained by the fact that the use of synthetic pigments is not allowed in organic production. No significant differences were established in shell breaking force or albumen quality. This research confirmed that there are differences in the chemical composition of eggs from different production systems. The difference was the most prominent in protein content which was significantly lower in eggs from conventional cages, indicating better nutritional quality of eggs from organic and free range production.

Keywords: Cages, Eggs, Free range, Organic, Quality

The comparative efficiency of organic vs inorganic forms of selenium and vitamin E in hen diets for biofortification of table eggs

Abstract ID: 125

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The efficiency of organic and inorganic forms of selenium (Se) and vitamin E (VE) as dietary supplements for laying hens for egg biofortification was studied in a trial on commercial "SP-789" layers (White Leghorn) kept in cage batteries from 140 to 200 days of age (7 treatments, 30 birds (6 replicates of 5 birds) per treatment, each replicate caged separately). Control treatment 1 was fed standard commercial wheat-based diet (basic diet BD) containing wheat 57.2%, wheat bran 4.7%, soybean meal 10.36%, sunflower cake 8.56%, corn gluten 3%, sunflower oil 3%. The dietary contents of ω-6 and ω-3 PUFAs were 3.69 and 0.12%, respectively, VE content 10 ppm, Se 0.2 ppm (as sodium selenite). Treatments 2-4 were fed BD where sunflower oil was substituted by flaxseed oil (3%) and flaxseed cake (5%) was added. The contents of ω-6 and ω-3 PUFAs were 2.49 and 2.16%, respectively, VE content 150 ppm, Se 0.5 ppm. Treatments 5-7 were fed the same diet as treatments 2-4 supplemented with "Fatty Acids" preparation (1.5%) based on the processed wastes of oil industry and containing >90% of fat, >11,300 µg/g of natural α-tocopherol, 280 µg/g of natural carotenoids. The contents of ω-6 and ω-3 PUFAs were 2.50 and 2.23%, respectively, VE content 150 ppm, Se 0.5 ppm. In treatments 1-4 VE source was synthetic DL-α-tocoferyl acetate, in treatments 5-7 preparation "Fatty Acids" (an organic form). Se source in treatment 2 and 5 was Sel-Plex®, in treatments 3 and 6 Sel-Plex® + "DAFS-25" (1:1), in treatments 4 and 7 Sel-Plex® + sodium selenite (1:1). All diets were supplemented with enzyme preparation "Fecord" (100 ppm). All experimental treatments had better indices of egg productivity and feed efficiency compared to control. Se content in 100 g of edible part of the eggs (59.3- 61.5 µg) was 2.2-2.5 times higher in compare to control; VE content (6.16-8.82 mg) 2.0-2.8 times higher; total content of ω-3 PUFAs (655-960 mg) 3.4-5.0 times higher (with ω-6/ω-3 ratio 2.3-2.9:1 vs. 14.2:1 in control); α-linoliec acid (400-618 mg) 5.2-8.0 times higher. The best results were found in the treatment 7.
The study was financed by Russian Science Foundation, grant 16-16-04047.

Keywords: Aying hens, Selenium, Table eggs, Vitamin E, Ω-3 polyunsaturated fatty acids

The effects of feeding Diamond V Original XPC on reducing Salmonella prevalence, numbers and NARMS panel antibiotic resistance in cloacal swabs taken from commercial layers

Abstract ID: 288

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A field study was conducted using two sets of independent paired commercial layer houses within a single company to determine the effects of feeding Original XPC™ (XPC) on reducing the prevalence and numbers of *Salmonella*, including an evaluation of antibiotic resistance. Birds were fed a company standard diet (CON) or a diet that contained 0.75 kg/MT of XPC. The genetics used were ISA Brown. At 42 weeks of age, individual cloacal swabs were taken from 400 birds per house (total N= 1,600; N=800/treatment). Cloacal samples were shipped overnight to Iowa State University where they were analyzed for prevalence and numbers of *Salmonella*, with positive samples tested for antibiotic resistance using a panel of 19 different antibiotics as outlined in the National Antimicrobial Resistance Monitoring System (NARMS) program. Data were analyzed in SAS using the GLM procedure with feeding treatment as the main effect, trial as a random effect, and significance considered at $P \leq 0.05$. Feeding XPC significantly reduced ($P < 0.0001$) *Salmonella* prevalence compared to CON (44.5% vs. 69.1%, respectively). A concomitant significant reduction ($P < 0.0001$) was also observed for *Salmonella* numbers in XPC-fed birds vs. CON (2,835.4 vs. 29,487.6 CFU/g, respectively). The NARMS panel contains 19 different antibiotics which covers eight classes of antibiotics. Across both trials, a total of 6,669 individual *Salmonella* isolates were tested, and the inclusion of XPC in the diet resulted in a significant reduction ($P < 0.05$) in the resistance of *Salmonella* isolates to 18 of the 19 antibiotics tested in the NARMS panel. These data suggest that the addition of Original XPC to the diet of commercial layers is an effective in-feed intervention for the reduction of *Salmonella* prevalence, numbers, and antibiotic resistance.

Keywords: Antibiotic, Layers, Resistance, Salmonella, XPC

The technology of new functional foodstuffs based on egg albumen enriched with calcium and iodine

Abstract ID: 119

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Growing interest to functional foodstuffs (FFSs) beneficial for the consumers' health requires the development of modern technologies improving bioavailability and conservation of functional food ingredients (FFIs) within the functional foodstuffs. We used egg albumen as a raw material for the development of new FFSs; native albumen is FFI per se containing 12-14% of well-balanced protein and low amount of fat; hence the albumen can be used in FFSs promoting muscle growth in consumers. At present large segments of consumers are short in dietary calcium and iodine supply. FFSs positioned as the sources of calcium and iodine (as FFIs) should provide these minerals in amounts corresponding to no less than 20% of daily requirements and in a bioavailable and secure form. The sources of these FFIs were chosen; the technologies of enrichment were developed ensuring target concentrations of the FFIs in final FFSs. Mineral additive based on the eggshell (Ca content 72.5%) was used as calcium source (1%); powder of dried laminaria (iodine content 189 mg/100 g) was chosen as a source of bioavailable iodine (0.2%). Both FFIs were added to the egg albumen during its thermal treatment and soft acid-salt hydrolysis. The resulting coagulated egg albumen enriched with calcium and/or iodine have neutral taste without off-tastes and off-odours. The influence of initial supplementation with the sources of calcium and/or iodine and the regimes of thermal treatment and hydrolysis on the efficiency of enrichment of final FFSs was studied; the optimal parameters of these processes were determined. The yield of coagulated albumen was found to be primarily dependent on final coagulation temperature. The following FFSs were developed: 1) calcium-enriched coagulated albumen; 2) calcium-enriched coagulated albumen with cream; 3) calcium- and iodine-enriched albumen; 4) calcium-and iodine-enriched albumen with fruity filler. Protein content in these FFSs is 14.6; 12.4; 15.1; 13.0%, respectively. Calcium-enriched albumen with cream (FFS 2) contains 2.0% of fat (due to 20% of milk cream in this receipt); fat content in other FFSs is insignificant. FFSs 2 and 3 contain 5.23 and 2.32 mg/kg of iron, respectively; 4423.0 and 3622.0 mg/kg of calcium; 1228.0 and 1101.2 mg/kg of sodium; 36.0 mg/kg of phosphorus; 13.84 and 13.85 µg/100 g of selenium. FFS 4 contains 0.25 mg of iodine in 100 g.

The study was financed by Russian Science Foundation, grant 16-16-04047.

Keywords: Egg albumen derived functional foodstuff, Enrichment with calcium and iodine, Soft acid-salt hydrolysis

Carcass composition and edibility yield of two heavy broiler strains

Abstract ID: 496

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In Germany, consumers increasingly prefer convenience-cut chicken parts to entire carcasses. Therefore, an increasing demand for heavier chickens can be observed as they are better suited for dissection. So far, only few data on the composition of heavy chicken carcasses exist, in particular with respect to the proportion of edible tissue. These data are relevant for the evaluation of population-wide poultry meat consumption, or in the context of human dietary plans with respect to food nutrition facts.
A sample of 100 carcasses from two fast-growing strains was selected from a representative German slaughterhouse. Carcasses (without giblets) were separated into parts according to Working Group V of WPSA, where breast, leg (separated into thigh and drumstick) and wing form the valuable parts. For each valuable part, we determined the edible fraction (muscles with tendons, skin and fat) by separating bones and cartilage. For entire carcasses, we calculated the respective percentage of edible and non-edible tissues.
The two strains differed in carcass weight, with mean values of 2285g and 2032g, respectively. In consequence, also the weight of most parts differed significantly between strains. The heavier strain had also higher percentages of thigh and wing, while the lighter strain was balanced by a higher breast percentage. The average carcass had 77.4% of valuable parts, which did not differ between strains. These parts of both strains had comparable percentages of edible tissue, except for the breast. The lighter strain (i.e. with the lighter breast) had slightly more edible breast tissue than the heavier strain (86.8% vs. 86.1%). Thigh, drumstick and wing had overall 86.0%, 72.3% and 75.7% of edible tissue, respectively. Such close percentages of edible tissue had not been expected.
The influence of carcass weight irrespective of strain was controlled by separating two classes above or below 2100g. Apart from the absolute weight differences, only the edible percentage of wings was slightly higher in the heavier group (76.0% vs. 75.3%).
The total percentage of edible tissue in broiler carcasses was not affected by strain or weight class. Based on our results, on average 64% of a commercial broiler carcass is edible tissue suited for human consumption. A proportion of 36% leftovers for broiler carcasses appear to be rather high.

Keywords: Carcass composition, Chicken, Dissection, Edible portions, Valuable cuts

Differences in performance and carcass yield of Ross 308, JA757 and ISA Dual chickens

Abstract ID: 227

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In the present, different meat type of chicken genotypes have been used for meat production depending on intensive or organic production. The effect of genotype on meat quality has been well described; however, information about performance parameters are limited. The aim of the study was conducted to compare fattening performance and carcass composition of cockerels and pullets of fast-, medium- and slow-growing genotypes of chickens fattened in identical conditions. One thousand and two hundred 1-day old chickens were split into six groups according to genotype (Ross 308, JA 757 and ISA Dual) and sex. Chickens were housed in littered pens and fed *ad libitum* the same diets. Chickens Ross and JA 757 were fattened for 35 days and ISA Dual 70 days. Final weight of males (2183 g, 1906 g and 1851 g) significantly differed from females (1929 g, 1699 g and 1459 g). Cockerels and pullets ISA Dual had significantly lower weight than Ross 308 and JA 757. As expected, feed conversion ratio was the significantly highest in ISA Dual females and males, whereas between Ross and JA 757 males and females did not differ. Zero mortality was observed in both sexes ISA Dual and in JA 757 females. Chickens ISA Dual reached only 27 and 20 % of European production efficiency factor of Ross 308 chickens, and in chickens JA 757 was 90 and 84 %. The lowest dressing out percentage ($P \leq 0.001$) and breast meat percentage ($P \leq 0.001$) were in males and females ISA Dual. On the other hand, ISA Dual cockerels had the highest thigh percentage ($P \leq 0.001$), and females abdominal fat percentage ($P \leq 0.001$). In conclusion, the performance of show that medium-growing chickens JA 757 grew faster than needed for organic production, whereas, ISA Dual males are suitable for organic production due to slow growth and low mortality.

Keywords: Carcass composition, Chicken, Fattening performance, Genotype

Effect of dietary ferric tyrosine (TYPLEX™ chelate) supplementation on Campylobacter jejuni counts, on quality and sensory characteristics of breast and thigh meat of broiler chicken exposed to natural Campylobacter jejuni challenge

Abstract ID: 92

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Food-born enteric diseases due to contaminated meat is a major concern for the broiler industry. Considerable global efforts are being made to prevent human campylobacteriosis by non-antibiotic means. Identification and large scale use of feed additives with protective properties against Campylobacter jejuni infection is an urgent priority. The aim of the present study was to evaluate the efficacy of dietary TYPLEX™ chelate against C. jejuni infection in broiler chickens and the effects on intestinal health and meat quality. A total of 384 male one-day-old broiler chicks were used in a 42-day trial. They were randomly allocated into four treatments with six replicates. Control group (T1) was fed basal diets in mash form. The other three groups were fed the same basal diets further supplemented with TYPLEX™ chelate at 0.02 g/kg feed (T2), 0.05 g/kg (T3) or 0.20 g/kg feed (T4). TYPLEX™ chelate (Akeso Biomedical Inc.) contains ferric tyrosine, an iron chelate (III) with L-tyrosine (4-hydroxyphenylalanine). Diets did not contain any other added iron compounds, coccidiostats or antibiotics. Feed and water were provided ad libitum. At day 20, broilers were exposed to natural C. jejuni challenge by means of contaminated litter from a commercial farm. On day 42, all chickens were slaughtered and from each pen 6 birds were randomly selected for post-mortem analyses. Caeca were collected for microbiological analyses; Breast (Pectoralis major) and thigh meat (Pectoralis major) samples were analysed for moisture, crude protein and fat content, by NIRS; Meat quality and organoleptic properties were evaluated by a taste panel. TYPLEX™ chelate had a significant effect against C. jejuni colonization in the gastrointestinal tract, ($P \leq 0.05$) limiting intestinal damage and lowering C. jejuni loads in the chicken intestine ($P \leq 0.05$). In the breast meat samples, the T4 group had a tendency ($P=0.087$) for higher protein content compared to the T3 group. In the thigh meat samples, the T3 group had significantly ($P=0.006$) lower fat compared to groups T1 and T4. The T4 group had significantly ($P=0.002$) lower protein content than groups T1, T2 and T3. Regarding breast meat, the T3 group had significantly better scores in tenderness ($P=0.002$) and juiciness ($P=0.008$) compared to T1 and T4. No significant differences were noted for cooked thigh meat. In conclusion, the protective effect of TYPLEX™ chelate against C. jejuni challenge resulted in a healthier gut with beneficial effects on nutrient absorption and metabolism that affected meat tissue formation. This research was financed by Akeso Biomedical, Inc., USA.

Keywords: Broiler chickens, Campylobacter jejuni, Feed additives, Intestinal health, Meat quality

Effect of dietary marine microalgae (Schizochytrium. SHG104) on growth performance, meat quality and fatty acid composition in broiler

Abstract ID: 319

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This study aimed to investigate the effect of dietary marine microalgae (*Schizochytrium*. SHG104) on growth performance, meat quality and fatty acids composition in broiler. A total of 480 male one-day broiler chickens (BW = 39.6 ± 0.4 g) were randomly assigned to 4 dietary treatments with 4 replicated pens consisting of 30 chicks. The basal diet was formulated to be adequate in energy and nutrients. Three additional diets were prepared by adding 20 g/kg marine microalgae (MA), 20 g/kg salmon oil (SO) or 20 g/kg flaxseed oil (FO) to the basal diet. The experimental diets were fed ad libitum to the birds during 35 d. Results indicated that BW gain and final weight were greater ($P<0.05$) in MA treatment than other treatments. But, there were no significant differences in feed intake and feed conversion ratio during the 35 d feeding trial. There were no significant differences in total cholesterol, total protein, triglyceride, albumin, glucose, AST and ALT during the feeding trial. Significant ($P<0.05$) differences were found in white blood cells and heterophils among broilers fed on diets supplemented with FO. The concentrations of dry matter (DM), crude ash, crude protein, and crude fat in the thigh meat were not different among dietary treatments. The pH value was not significantly affected by dietary treatments except increased FO treatment at 1 and 5d. There was no significant differences in all treatments at 7d but, MA treatment was lower than other dietary treatments. WHC was not significantly affected in all treatments and no correlations between pH and WHC was observed. TBARS values of all the dietary treatments increased during storage but broilers supplemented with MA significantly have the lower TBARS value than SO and FO treatments in thigh meat at 7d. In fatty acid composition of thigh meat, DHA content of broilers fed with MA was significantly ($P<0.05$) highest and omega-6/omega-3 ratio was improved in all treatments compared with the basal treatment. These results suggest that dietary MA may be used as a functional ingredient to improve body weight gain, and omega-3 composition of thigh meat.

Keywords: Broiler, Fatty acids, Flaxseed oil, Marine microalgae, Salmon oil

Effect of different concentrations of the Powdered Hawthorn (Crataegus elbursensis) extract on the physical quality of chicken's breast meat

Abstract ID: 540

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The quality of the meat depends on the different physical, chemical and structural features of the meat. Hawthorn has been used as herbal drug for a long time. Some useful properties of this planet include the improvement of the cardio-vascular system, hypo-clostromia, anti-oxidant characteristics and so on.
The purpose of the current study was investigating the effect of different amount of the Hawthorn powder on the physical quality of the chicken's Brest meat. To do so, 150 one-day male chicks of 308 Ross were used based on the completely randomized design with four treatments, three replicates and ten chicks per replicate.
The treatments included: treatment 1: Control diet; treatment 2: control diet+1.4 g / kg Hawthorn powder; treatment 3: control diet+2.8 g / kg Hawthorn powder; treatment 4: control diet + 5.6 Grams per kilogram of Hawthorn powder. Meat rigors were measured by texture profile analysis (TPA). To measure the meat tenderness, a compressive test with a dental probe (TA 7) was used, and a cylindrical probe (TA 25) was used to test the quality of the material. The hardness and deformation in hardness were measured by the compressive test. Moreover, the characteristics of the meat tissue, including factors such as hardness, cohesiveness, adhesiveness, elasticity, chewiness and gumminess were measured through TPA test. In this regard, the samples of breast meat chicks were gathered at the day 42. After the rigormortis process, the samples were boiled for 30 minutes in the temperature of 90^0c. Then, the samples with the size of 2*2*2cm for the TPA test and with the size of 2*4*2 cm for the compressive test were used. In compressive test, results showed that treatments didn't affect hardness (179.0, 244.3, 115.2, 172.4 g, respectively) and deformation in hardness factors (14.3%) of the breast meat ($P<0.05$). In addition, the hardness (29.84, 25.77, 2075.10, 1837.4 g, respectively), cohesiveness (0.08, 0.10, 0.11, 0.12), adhesiveness (.0.468, 0.483, 0.513, 0.483 mj, respectively), elasticity (0.521, 0.466, 0.518, 0.466 mm, respectively), chewiness (86.34, 82.75, 67.05, 54.56 mj, respectively) and gumminess factors (1420.6, 1313.5, 873.3, 882.8 g, respectively) in TPA test were not under the influence of the different treatments ($P<0.05$). The findings of the current study showed that hawthorn powders did not reduce the physical characteristics of the meat in mentioned test. Hence, regarding the useful properties of this plant, further research suggests to investigate the magnitude effect of this supplementary fruit for the chickens.

Keywords: Chicken, Compressive test, Hawthorn, Meat quality, TPA

Effect of incorporating an emulsifier complex (DIGESTFAsT) on broiler meat quality during the storage

Abstract ID: 218

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The objective of this study was to evaluate the effect of a biosurfactant (DIGESTFAsT) additive combining lysophospholipid emulsification activity with choline , L-carnitine and betaine, extracted from artichoke. A total number of 100 one-day-old JV chicks were allotted two groups of 7 pens with 7 chickens each. Two diets based on maize and soy bean meal, Control (without additive) (C) and Experimental (E) (with the additive, 500 g/Ton), were prepared and each diet was distributed to a group. At the end of the fattening essay (39 days of age), 32 broilers (8 males and 8 females from each diet) were slaughtered. After a six-days-storage period (+4°C), variations of the color, total phenol index, aerobic bacteria and fecal and total *Coliform* were analyzed on breast meat samples. Our findings show that breast meat color measured after 24h was similar in both groups. However, after the storage period, luminance index in E samples lot was higher than in C ($P = 0.038$), while total phenol content in breast meat increased ($P = 0.04$) in E chickens' meat. The analyzed germs after storage, were higher in C group ($P=0.026$). Moreover, the tested feed supplement tended to reduce ($P = 0.08$), meat oxidation during the storage period, in chickens fed E diet. In conclusion, DIGESTFAsT complex preserves breast meat quality by reducing the multiplication of total germs, inhibiting its oxidation and increasing total phenol content.

Keywords: Broiler, Emulsifier, Meat quality, Storage

Effect of laying-type cockerels DOMINANT CZ hybrid combination on carcass parameters and meat quality

Abstract ID: 488

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The aim of the experiment was to evaluate carcass parameters and meat quality with focus on physico-chemical properties in four hybrid combinations of cockerels of the DOMINANT CZ commercial program. The experiment was performed in the experimental facility of Faculty of AgriSciences, Mendel University in Brno. Four different hybrid combinations of DOMINANT CZ laying-type cockerels from the hatchery Lihen Studenec, s.r.o. were observed (D 104 Sussex, D 109 Black, D 459 Red Barred and D 853 Red). A total of 20 cockerels of each hybrid combination were used for observation, they were housed in the same place, fed the same diets. The experiment was finished at the age of 18 weeks and selected parameters of carcass value and meat quality were analyzed. The results revealed significant differences in live weight of cockerels, which reached the highest level in D 853 hybrid ($P<0.05$). Also carcass weight was the highest in D 853 hybrid ($P<0.05$). Carcass value ranged from 65.6 % in D 109 to 67.8 % in D 853 ($P<0.05$). No significant differences were found among the observed hybrids in the proportion of breast muscle in the carcass, while the proportion of thighs in the carcass was the highest in D 109 ($P<0.05$) in comparison with all the other hybrids. Evaluation of dry matter content in breast muscle showed differences among the hybrids. The highest values were found in D 109 and D 853, significantly lower was the content of dry matter in D 104 ($P<0.05$). The fat content in the breast muscle was not statistically different among the cockerels. Evaluation of redness of the breast muscle sample (a*) showed no statistical differences among the hybrids, but the values of lightness (L*) and yellowness (b*) were different. Brighter breast muscle samples were found in D 853 hybrid ($P<0.05$). Evaluation of thigh muscle showed no effect of hybrid combination on dry matter content. The highest fat content ($P<0.05$) was found in D 853 hybrid and the lowest fat content in thigh muscle sample was found in D 109 hybrid. The values of L* were the highest in D 853 hybrids ($P<0.05$), the values of a* in D 459 hybrids ($P<0.05$) and the values of b* in D 853 hybrids ($P<0.05$). In conclusion, there were statistically significant differences found in the evaluated parameters among the cockerels of the observed hybrids. The D 853 hybrid was characterized by the highest values of live weight and carcass weight, but also the highest values of L* and b* and the lowest value of a*.

Keywords: Breast and thigh muscle, Carcass, Colour of meat, Laying-type cockerel, Live weight

Effect of organic acid mixture on broiler chicken meat and liver quality

Abstract ID: 565

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The trial was conducted to investigate the effect of organic acids mixture supplementation on broiler chickens performance, MDA (malondialdehydes) concentration in liver and breast muscle. During a 5-week feeding experiment, 1000 one-day-old Ross 308 broiler chickens were fed by the same diet ad libitum with a crumbled wheat-soybean meal based diet. Control group diet was standard compound feed, experiment group I - standard compound feed supplemented with 0.5 kg/t of organic acid mixture, experimental group II – supplemented with 1 kg/t of organic acid, experimental group III – 1.5 kg/t of organic acid preparation and experimental group IV – 2 kg/t of organic acid preparation. MDA concentration in the meat were determined by high performance liquid chromatography as described by Mendes (2009), with a high pressure gradient HPLC system (Varian ProStar). The results of the experiment were analyzed using the 1-way ANOVA test, and significant differences between groups were determined by Duncan's multiple range test. Statistica 8.0. for WindowsTM software was used. Differences were considered significant at $P<0.05$. Statistically significant results on broiler chickens productivity were not determined. The highest concentration of MDA in fresh liver of broiler chicken were found in control group,whlile in the remaining groups it decreased from 2 to 26% ($P<0.05$). After a storage of 3 months – the highest concentration of MDA was found again in control group, while in experimental groups it decreased from 2.32 to 2.5 times ($P<0.05$) compared to the control group. The same trend was observed when data of MDA of fresh and storage breast muscle were analysed. In fresh breast muscle the highest concentration of MDA was detected in control group, while in the remaining groups it decresed from 5 to 22% ($P<0.05$). In 3 months stored brest, this parameter decreased from 11 to 22 % ($P<0.05$) in all experimental groups compared to the control group. In conclusions, the organic acids preparation had positive effect on lipid oxidation process in liver and broiler chicken brest muscle.

Keywords: Chicken broiler, MDA, Organic acid

Effect of Rosemary powder (Rosmarinus Officinalis) feed incorporation on organic broiler chickens performance and carcass quality

Abstract ID: 180

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The aim of the present study was to evaluate the effect of Rosemary powder (RP) on growth performance and carcass quality of organic broiler chickens. A total of 100 one-day-old chicks (Hubbard JV) were randomly allotted into 12 groups of 8 to 9 chicks each (6 replicates per treatment). Six groups of birds were fed the basal diet while the other six groups were fed the basal diet supplemented with 0.25 % of RP. At the end of the fattening, 12 chickens (6 males and 6 females) from each treatment were slaughtered and carcass quality analysis was made. Aerobic bacteria and fecal and total *coliform*and oxydation in meat samples from breast fillets were analyzed after 72h storage period (72h at +4°C). Final live weight of chicken, weight gain, feed intake, feed conversion ratio and mortality rate during the rearing period did not differ significantly between groups. Same findings were obtained for eviscerated hot and chilled carcass yield (74.02% and 71.34%). Not significant change (P>0.05) was detected on proportion of viscera (intestines, gizzard, gizzard fat, liver and heart). After 24h and at the end of the storage period, total bacteria population was higher in the control diet (1.04 10^2 and 2.27 10^2 vs 9.54 10^2 vs and 19.08 10^2 (P<0.05) while fecal coliforms (24h) were lower (P<0.05) in RP lot (2.72 102 vs 2.72 103). Variation in meat oxydation during storage was also lower (P=0.01) in RP group (8.10^{-4} vs 8. 10^{-3} of MDA mg/kg). Based on this, it can be concluded that the dietary supplementation of 0.25% RP did not have any adverse effects on organic broiler performance and carcass traits and RP supplementation improved the microbiological stability and inhibited oxydation of broilers' breast meat.

Keywords: Broiler, Carcass, Organic, Performance, Rosemary

Effects of feeding Diamond V Original XPC on Salmonella prevalence, numbers, and antibiotic resistance in ceca samples taken from commercial broilers

Abstract ID: 287

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Large-scale field trials were conducted to determine the effects of feeding Original XPC™ on reducing pre-harvest *Salmonella* in broilers at commercial processing plants, including an evaluation of *Salmonella* virulence and antibiotic resistance. In this study, a total of 217 commercial broiler houses from 11 companies were monitored. Birds were fed a company standard diet (CON) or a diet that contained 1.25 kg/MT of Original XPC™ (XPC). Birds were fed a commercial diet with or without XPC an average of 50 days. Depending on the company protocol, at processing, one cecum was collected from between 50 to 100 birds per house (7,591 total cecum samples) during evisceration at the processing plant. Ceca samples were shipped overnight to Iowa State University where they were analyzed for prevalence and numbers of *Salmonella*, with positive samples tested for antibiotic resistance. Data were analyzed in SAS using the GLM procedure with feeding treatment as the main effect company as a random effect and significance considered at $P \leq 0.05$. Feeding XPC significantly reduced ($P < 0.0001$) *Salmonella* prevalence compared to CON (6.9% vs. 19.4%, respectively). A significant reduction ($P < 0.0001$) was also observed for *Salmonella* numbers in XPC fed birds vs. CON (8.0 vs. 147.0 CFU/g, respectively). All positive samples were measured for resistance to the antibiotics florfenicol, ceftiofur and enrofloxacin. A total of 21,029 *Salmonella* isolates were evaluated for antibiotic resistance and feeding XPC significantly lowered ($P < 0.0001$) resistance compared to CON (florfenicol 1.4% vs. 17.7%; ceftiofur 0.5% vs. 13.6%; enrofloxacin 0.0% vs. 6.5%, respectively). In summary, the addition of XPC resulted in a significant reduction in both *Salmonell*a prevalence and numbers. *Salmonella* recovered from fed XPC birds were less to susceptible to three different classes of antibiotics. These data suggest that the addition of Original XPC™ to the diet is an effective in-feed intervention for *Salmonella* in broilers.

Keywords: Antibiotic, Broilers, Resistance, Salmonella, XPC

Inter-muscular variation in ultimate pH and colour of broiler muscles

Abstract ID: 326

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Breast, legs and wings represent the cut-up of main commercial interest in the broiler industry. In particular, breast is principally marketed for fresh consumption, while legs are mainly used for the preparation of minced and pre-cooked products. Previous studies have mainly evaluated the pH_u and colour of broiler breast muscles, while there is a lack of knowledge about wing, drumstick and thigh muscles. Thus, this study aimed at providing reference values of colour and pH_u of principal muscles of the most important commercial cut-up of broiler chickens. For this purpose, 10 muscles (*P. major* and *minor* from breast, *M. scapulotriceps* from wing, *E. iliotibialis anticus, F. cruris medialis, E. iliotibialis lateralis* from thigh and *P. longus, F. perforatus digiti III, G. pars interna, F. cruris lateralis* from drumstick) from 7 chicken carcasses (male broiler Ross 308, medium-size, 44 days-old slaughtered at 2.8 kg) were used to assess pH and colour (L*a*b*) at 24 h *post mortem.* A large inter-muscular variability was found, especially as regards pH_u (range 5.85-6.57) and a* values (range 1.9-12.6), while L* (range 50.1-55.5) and b* (2.1-7.3) showed a slighter variation range. If cut-up were compared (breast-wing, thigh and drumstick), breast-wing part exhibited a significantly lower pH_u if compared with drumstick (5.96 *vs.* 6.46; P<0.05), while thigh showed intermediate values (6.37). In addition, if compared with thigh and drumstick, breast-wing muscles presented lower a* and b* values (3.0 *vs.* 9.0 and 8.3; 3.4 *vs.* 5.5 and 5.6, respectively; P<0.05), while L* did not differ. Thus, leg muscles' pH_u and colour significantly differ from what observed in breast and wing likely due to the different fibre composition and intramuscular fat accumulation. In conclusion, chicken drumstick and thigh muscles have very different peculiarities and their properties should be carefully considered especially when used for further processing.

Keywords: Breast, Chicken meat, Colour, Leg, PH

Physical and chemical meat characteristics of chicken ross 308, JA757 and isa dual hybrid

Abstract ID: 226

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The aim of this study was to evaluate the effect of genotype and sex on physical meat characteristics and chemical composition in fast- (Ross 308), medium- (JA757) and slow-growing (ISA Dual) chickens. In total 1200 chickens were fattened under identical conditions. For meat quality analysis, 40 birds per each genotype (1:1 male to female) were slaughtered at 2 kg live weight and 24 hours post mortem the samples of meat were taken. The chemical characteristics and drip loss were detected in the *Pectoralis major* (PM) muscle and physical meat (pH, colour) characteristics were measured in *Pectoralis major* and *Biceps femoris* (BF). The Ross 308 reached 2 kg live weight at 32 days of age, JA757 at 35 days of age and ISA Dual at 70 days of age. From physical meat characteristics, pH measured in PM was affected by genotype (P=0.005) with lower values in ISA Dual and JA757 compared to the Ross. In BF muscle lower pH was detected in ISA Dual than in Ross and JA757. Interaction of genotype and sex were detected in colour parameter L* (lightness) in PM, with the lightest meat in cockerels of ISA Dual. Other genotypes and sexes did not differ in lightness of breasts. The Ross chickens had significantly more intensive colour parameters a* (redness; P=0.009) and b* (yellowness; P=0.002) than both genotypes JA757 and ISA Dual. Similarly in BF muscle, the highest (P=0.028) lightness had cockerels of genotype ISA Dual, on the other hand, Ross had darker thigh meat. The drip losses of PM was lower in JA757 and ISA Dual of both sexes, however, the highest drip loss was in hens of Ross 308. The chemical characteristics were not affected by interaction of genotype and sex. The Ross 308 had the lowest dry matter (P<0.001), crude protein (P<0.001) and higher ether extract (P<0.001) than JA757 and ISA Dual. We can conclude that even though the meat of slow- and medium-growing chickens had lighter colour, it had better chemical meat quality parameters.

Keywords: Chemical composition, Chicken, Genotype, Pectoralis major, Physical parameters

Phytochemicals for controlling Campylobacter jejuni in poultry: An update on novel delivery methods for increased efficacy and understanding of antimicrobial action

Abstract ID: 292

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The contamination of poultry products with *Campylobacter jejuni* remains one of the leading causes for foodborne illness globally. Chickens act as the host for *C. jejuni*, wherein the pathogen colonizes the ceca thereby leading to contamination of the carcass during slaughter. Reducing *C. jejuni* cecal colonization in poultry gut and survival in poultry products would potentially reduce the risk of human infections. With growing consumer preference for minimally processed, natural food products and increasing concerns over the use of synthetic chemicals in food industry, there is significant impetus on developing safe and effective strategies for improving the microbiological safety of poultry products. One of the missions of our laboratory has been to provide the poultry industry with efficient antibiotic alternatives for the control of *Campylobacter* in conventional and the organic poultry sectors. The use of phytochemicals as antimicrobial feed additives and food bio-preservatives is one such technology that is safe, effective and environmentally friendly. We have tested a plethora of phytochemicals for their anti-*Campylobacter* efficacy, both in preharvest and postharvest poultry. Our results indicate that plant-based, GRAS (Generally Recognized as Safe) status compounds such as trans-cinnamaldehyde (obtained from cinnamon bark), carvacrol (from oil of thyme), eugenol (from clove oil), and beta-resorcylic acid (from Brazilian wood) are very effective in reducing *C. jejuni* colonization in the gut when used as in-feed or in-water (with nanoemulsions for increased solubility) supplements in poultry. For example, in two separate trials, in-water supplementation of 0.25% trans-cinnamaldehyde nanoemulsion and in-feed supplementation of 0.5% beta-resorcylic acid consistently reduced *C. jejuni* colonization by at least 1.5 log CFU/g of cecal material in 14-day old broiler chickens ($P<0.05$; 10 birds/treatment/trial). Moreover, these compounds are also effective in reducing the survival of *C. jejuni* on chicken meat when applied as an antimicrobial wash or coating treatments. Washing with 0.5-2% beta-resorcylic acid for 30 sec and with 0.5% trans-cinnamaldehyde nanoemulsion for 1 min reduced *C. jejuni* by 1-2 log CFU/ml on chicken skin and meat ($P<0.05$). Follow up mechanistic studies (using next generation sequencing and proteomics analysis) have revealed that phytochemicals such as trans-cinnamaldehyde, beta-resorcylic acid and eugenol modulate key *C. jejuni* genes and proteins that facilitate colonization in poultry gut, persistence in poultry processing environment, biofilm formation and survival in meat products. Currently, using microbiome and *in vivo* metaproteomic approaches, we are analyzing the effect of aforementioned compounds on gut health and productivity parameters in poultry.
Funded by USDA-NIFA-OREI-2017-51300-26815.

Keywords: Campylobacter, Gut health, Phytochemicals, Poultry safety

Productivity and meat quality in floor vs. cage housed broilers

Abstract ID: 122

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The effects of management system (floor vs. cage housing) and slaughter age (38, 42, and 49 days) on the productive performance and meat quality in broilers were studied in the 2x3 experiment on 6 treatments (400 broilers per treatment in two replicates of 200 birds) of Cobb-500 broilers since 1 day of age. Live bodyweight in cage-housed broilers was insignificantly higher in compare to floor-housed; mortality level lower by 0.9-1.0%; feed conversion ratio better by 0.6-1.2%. Slaughter yield and carcass quality were also slightly better in cage-housed birds. Eviscerated carcass yields increased with the increase in slaughter age; these parameters were the highest at 49 days of age (73.9% in cage- and 73.3% in floor-housed broilers), higher by 3.1-2.7 and 2.7-2.2% in compare to 38 and 42 days of age, respectively. There were no significant differences in the relative yields of cuts and edible offal between housing systems at all slaughter ages. The highest protein content in breast meat was found in floor-housed broilers (from 20.6% at 38 days to 22.8% at 49 days of age); in cage-housed broilers the corresponding parameters were 20.0 and 20.4%. Fat content in breast meat in cage-housed birds was significantly higher ($P<0.05$) in compare to floor-housed. Average taste scores of bouillon and meat taste determined in a taste panel examination were higher in floor-housed broilers at all studied ages; within the single management system these scores tended to increase with age. It was concluded that growth and feed efficiency were better in cage-housed broilers while meat quality parameters were slightly better in floor-housed birds.
The study was supported by Russian Science Foundation, grant 17-16-01028.

Keywords: Broilers, Feed efficiency, Live bodyweight, Meat quality, Mortality

Salt reduced chicken wings

Abstract ID: 424

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Elevated NaCl consumption has been associated with hypertension and cardiovascular disease. Therefore the EU and the UK have passed legislations and set targets to regulate the salt content of food products. The aims of the present study were to reduce the salt content of spicy marinated and breaded chicken wings (‘hotwings’) by 25% and to investigate and realize the maximal permitted salt content in hotwings according to UK targets without reduction of salt perception by consumers.
The reference hotwings contained 2.43% salt (NaCl), of which 0.21% in the chicken meat, 1.05% in the marinade and 1.17% in the breading layer. Salt reduction in the marinade was selected as starting point due to the spiciness of the marinade, which was suspected to compensate for salt perception partially. In this way, it was achievable to reduce the salt content of the hotwings by 20.34% without causing sensorial differences. The remaining 4.66% salt reduction to meet the target of 25% salt reduction were achieved by reduction of salt in the breading. A sensory panel (n=31) evaluated the parameters saltiness, tenderness, juiciness, and color, and perceived the 25% salt reduced hotwings as comparable to the reference product.

The maximal allowed salt content according to UK targets depends on the type of product and its nutritional value. Hotwings fall into category 3 ‘Battered or breaded chicken portions and pieces’. The portion size for hotwings was established to be six pieces of 1st part hotwings. Including experimentally determined oil absorption during frying (6.2%) and moisture loss (28.1%), this portion size contains 390 kcal and is therewith allowed to contain a maximum of 2 grams of salt. One portion of 25% salt reduced hotwings contains 2.88 gram salt and is therewith not sufficiently salt reduced to comply with the UK targets. It was therefore investigated whether salt replacers were suitable. By using a salt replacer (50% sodium replaced by potassium and natural flavouring) instead of all common salt in the marinade and part of the common salt in the breading, a product was obtained that contained 2 gram NaCl per six 1st part hotwings. Also for the salt replaced hotwings, the sensory panel evaluated the same parameters as for the 25 % salt reduced hotwings described above, and perceived them as comparable to the reference product.

Keywords: Chicken wings, NaCl reduction, Replacing, Salt reduction, Sensory

The effect of polyphenols and vitamin E in the finishing feed for turkeys on meat quality

Abstract ID: 278

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The consumption of turkey meat is increasingly turning to more cut-up and processed products. The meat industry is facing with problem of meat discoloration of red meat linked with oxidation of meat. In this study the effect of a combination of polyphenols and vitamin E (G2) distributed in the feed during the last 28 days of breeding was compared to a negative control without added antioxidant (G1) in the feed of turkeys on oxidation susceptibility and drip losses of turkey meat, essentially from thighs. Three commercial turkey farms with two identical buildings each were selected (only male, same genetic and same age in both buildings). The turkeys from one building received G1 whereas G2 was distributed in the other building. At slaughtering, the 2 groups were monitored until cut-up and put into trays. The thighs meat was stored until 15 days and measurements were done at different storage time (3, 9, 13 and 15d). Drip losses (n=30), lipid oxidation (TBARS) (n=20), color score (n=120) and grey aera (mm^2) (n=50) were measured. The color score visually classified meat in 4 classes from 1 (acceptable) to 4 (unacceptable). Meat technological quality was assessed by drip loss measurement which was lower in G2 (0.64 versus 0.81for G1 at 3d, $P<0.05$). Meat from turkey receiving antioxidants (G2) showed lower TBARS value at 3 and 9 days of conservation ($P < 0.001$). Percentage of meat trays of Score 1 and 2 decreased more rapidly in G1 than G2 until 15 days of storage. This result was confirmed by the decrease of the grey area surface at 9 days of storage in G2 (respectively 29.9% versus 35.6%; $P < 0.05$). The combination of different antioxidant molecules (vitamin E and polyphenols) in finisher feed of turkey improved meat appearance and storage ability (color and oxidation susceptibility) as well as its technological quality (drip loss). Further investigations need to be realised on a larger scale to confirm these results.

Keywords: Antioxidant sources, Color, Meat quality, Turkey

The effects of preparation Polyferon on the productive performance and meat quality in broilers

Abstract ID: 121

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The trial was performed in the Institute's vivarium to evaluate the effects of immunomodulator Polyferon (Materia Medica, Russia) on the productive performance and meat yields and quality in Ross-308 broilers. The birds (35 per treatment) were reared in cage batteries R-15 (Russia) from 1 to 37 days of age. In control treatment 1 drinking water for broilers contained no additives; in experimental treatment 2 the water was supplemented with Polyferon (to achieve daily doses 0.005 g/bird from 1 to 22 days of age and 0.010 g/bird from 22 to 37 days). Live bodyweight in treatment 2 was significantly higher compared to control (by 6.7%, $P<0.05$). Mortality level in this treatment was 0% while in control 2.9%. The yield of eviscerated carcass in treatment 2 was 73.8%, higher by 1.5% compared to control; percentage of high-quality carcasses higher by 5.1%. There were no differences between the treatments in relative weights of liver, heart, gizzard, lungs, and kidneys; all the organs were well developed, no symptoms of pathological changes were found. The yield of edible carcass cuts was higher in Polyferon-treated broilers (82.5 vs. 81.2% in control), mainly due to higher muscle yield (63.7%, higher by 1.2% compared to control). Bone yield in treatment 2 was lower by 0.54%; yield of inedible carcass parts in treatment 2 was also lower compared to control. There were no differences between the treatments in chemical composition of breast and thigh meat. It was concluded that immunomodulator Polyferon in drinking water for broilers (0.005 g/bird from 1 to 22 days of age and 0.010 g/bird from 22 to 37 days) decreases mortality, improves live bodyweight and meat yields.

Keywords: Broilers, Feed conversion ratio, Live bodyweight, Meat yield, Mortality

Development of cryopreservation protocols of Copper turkey' semen

Abstract ID: 259

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Successful cryopreservation of Copper turkey's semen has importance in gene conservation and breed protection. Since the number of our indigenous turkey breeds, along with the other endemic poultry species, has declined drastically, they are included in *in vivo* and *in vitro* gene conservation projects. Since the existed differences in their tolerance for cryopreservation, different species require species – or even breed-specific freezing protocols. The aim of our research was to develop a freezing protocol that allows us the efficient storage of Copper turkey's semen. We tested different freezing protocols which efficiency was tested by *in vitro* methods. We used Lake's diluent and dimethyl-acetamide as cryoprotectant in a final concentration of 6%. Freezing/Thawing protocols:1.*Programmed, dynamic freezing in straw*, from +5°C to -35°C, with the rates of - 7°C/minute, then to -140°C with the rate of -20°C/minute. Thawing of the 0,25 ml straw was done at 5°C and 60°C (1/A, 1/B) 2.*Static freezing in 0,25 ml straw*, holding it at 1 cm above the surface of liquid nitrogen for 10 minutes, thawing was done at 5°C and 60°C. (2/A, 2/B) 3.*Pellet method*: 25 µl of diluted semen was directly dropped into liquid nitrogen, thawing was done at 65°C by a special thawing instrument (3). We got the highest rate of live, intact sperm (22,8%) and survival (29,2%) in the case of static freezing, followed by thawing at 5°C (2/A), although the rate of live, but morphologically abnormal cells was the highest (19,6%) in this case too. Among the morphological abnormalities midpiece disorders dominated both in fresh and thawed samples. After thawing, the rate of morphologically abnormal cells decreased in almost all cases, which suggests that abnormal cells are less tolerant against the process of freezing/thawing. The rate of acrosome-degenerations was higher as well after freezing, which were the most prominent in the case of pellet-method (from 0,08% to 2,6%).To determine the true fertilizing ability of thawed spermatozoa, we are planning artificial insemination tests, so after obtaining those results we can start the efficient storage of Copper turkey's semen in gene bank environment.

Keywords: Cryopreservation, Gene conservation, Sperm, Turkey

Effect of constant or weekly varied eggshell temperature during incubation on broiler performance up until slaughter age

Abstract ID: 560

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During incubation, embryo development is particularly influenced by temperature. The vast majority of commercial hatcheries aim at a constant eggshell temperature (EST) of 37.8°C throughout incubation. However, it has been shown recently that lowering EST (36.7°C) in the last week of incubation might improve embryo development, as higher (yolk-free) body mass and relative organ weights at hatch were found. In addition, raising EST slightly in phases during which oxygen is not limited yet (before the last week of incubation), might improve embryo development as well. It is hypothesized that a lower EST (36.7°) in the last week of incubation and a higher EST (38.9°C) in the second week of incubation improve embryo development and perinatal chick quality compared to a constant intermediate EST (37.6°) throughout incubation. Moreover, EST during incubation might have long term effects on broiler performance as the incubation period covers a substantial part of their whole lifespan and as it is known for many animal species that perinatal experiences have an impact in later life. To test this hypothesis, Ross 308 eggs from a prime parent flock were incubated in a 2x2 experimental design. All eggs were incubated at a normal (37.8°C) EST until embryonic day (E) 7. Thereafter, eggs were either incubated at a normal (37.8°C) or high (38.9°C) EST during the second week (E7 – E14) of incubation and a normal (37.8°C) or low (36.7°C) EST during the last week (E14 – E21) of incubation. Within 6 hours after hatch, chick development was evaluated by chick weight, length, navelscore, and organ weights. Posthatch, 5 males and 5 females were housed in a 2 m^2 pen with 8 replicates per treatment (n=320) and reared until slaughter age (D42). Growth and feed intake were monitored weekly. At D28, D35, and D39 gait was scored from all animals. At slaughter, foot-pad dermatitis, hock burns, and carcass characteristics were determined. Preliminary results indicate that a low EST from E15 onwards results in slower growth and on average 103 g. lower body weight at slaughter. Other parameters were not significantly different or not analysed yet (e.g. carcass characteristics) at the moment of abstract submission.

Keywords: Broiler, Eggshell temperature, Incubation, Performance

The effect of combined monochromatic lighting on sperm quality of broiler breeder males

Abstract ID: 377

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Artificial targeted illumination has pivot role in reproductive processes in poultry. Light absorption mechanism in poultry consists of two main components: the eye (retinal photoreceptors) and the extraretinal photoreceptors locate in the brain. Previous studies showed that photostimulation of extra-retinal photoreceptors elevate reproductive activities, while retinal photostimulation suppressed reproduction. All those studies were conducted on female chicken. In this study, we tested the effect of combined monochromatic photostimulation on reproductive activities of broiler breeder males. Fifty broiler breeder roosters (Ross), at 21 weeks of age, were divided to 5 light treatment rooms with individual cages (n=10). Light treatments: White (Control), long day of blue light (480 nm) with a short day of red light (630nm) (blue-red), long day of red light with blue light (blue-red), long day of green light (560nm) with short day of red light (green-red) and long day of red light and a short day of green light (red-green). Each week until 65 weeks of age semen samples were collected and analyzed for: volume (ml), motility (sample were ranked from 1-8 when 1 shows total lack of motility and 8 indicates excellent movement with turbulence), sperm count by a hemocytometer, and vitality of a sperm by Eosin-Nigrosine staining. Furthermore, once a month blood samples were drown for plasma hormonal assay. At 65 weeks of age roosters were killed and tissue samples (hypothalamus, pituitary gland, retina and testes) were taken for mRNA gene expression of GnRH-I, LH and FSH, and steroid respectively. Semen motility from red-green group was significantly elevated than all the other groups. Semen volume in red-green and blue-red groups were higher than all other groups. Semen concentration per ml was similar in all groups. however, concentration per ejaculation in green-red group was higher during the whole experiment. Testes weight was significantly higher in the red-green treatment group. There were no significant differences in viability. Further endocrine and genomic assay are conducting in order to understand the mechanism of this phenomena.

Keywords: Broiler-breeder-male, Photostimulation, Semen

Air Sample based Flock Diagnostics

Abstract ID: 597

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In the poultry industry diagnostic has typically been based on investigation and testing of individual hen or chickens selected for spot testing. Assuming that the infection in the flock is wide spread this approach is very effective. However, if the aim is to detect an emerging infection at an early stage spot testing is not necessarily an ideal solution due to pure statistics. In a population of say 20.000 animals an infection needs to be rather spread in order for a spot test of 20 animals likely will detect it.
Air sample based flock diagnostics provide a new approach to both monitoration and diagnostic of pathogens in large poultry flocks. Here we will present the latest results with this new approach to flock diagnostics based on the air samples collected with the newly developed handheld air sampling equipment (AeroCollect™) from FORCE Technology.
The AeroCollect™ equipment has been specifically developed to be suited for sample collection in dusty and harsh environment such as poultry houses. In this work we have successfully demonstrated the detection of *Campylobacter*, infectious bronchitis virus, avian reovirus, chicken astro virus and *e. coli* out of which the first two are already validated for screening and diagnostic use.
The samples are collected by walking around covering the whole house for a minimum of five minutes while sampling with the AeroCollect™. After collection the samples are analysed in the laboratory using standard qPCR and RT-qPCR techniques. Each sample can be tested for up to 10 different pathogens. If the test results are positive it is possible to determine the strain of the bacteria or virus directly from the already collected air samples without the need for further test material because in contrast to the antibody based blood diagnostic approaches the pathogens are detected directly in the sir samples. Furthermore, it is possible to test previously vaccinated flocks to investigate if another strain of the virus is spreading in the flock.

Keywords: Air samples, Bacteria, Flock diagnostics, Pathogen surveillance, Virus

Anticoccidial efficacy of a multi-strains yeast fractions product in commercial broiler chickens exposed to a mixed challenge of Eimeria acervulina, E. maxima, and E. tenella

Abstract ID: 647

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Coccidiosis is a major enteric disease that poultry industry tries to control as much as possible, since it is also a risk factor for necrotic enteritis. Anticoccidial drugs remains the most affordable solution but the development of some coccidia resistance encouraged the application of alternate strategies. Besides management programs including vaccination, nutritional solutions have been evaluated. The potential of a proprietary multi-strains yeast fractions product (YANG) is assessed in the present study using a randomized block design with 9 replications of 10 birds per cage. The treatments were nonmedicated non-infected (NMNI), nonmedicated infected (NMI), YANG dose 1 (400 g/t), YANG dose 2 (1000 g/t), YANG dose 3 (3000 g/t). Day of hatch male chicks (Ross 708) were placed into cages and issued treatment feeds (D0). Treatment feeds were available *ad libitum* throughout the experiment (D0-28). On D14, birds were challenged with *E acervulina* (EA), *E. maxima* (EM) and *E. tenella* (ET) at an average total of 150 000 oocysts, while NMNI birds received 1 mL of distilled water. On D5 and D6 post-infection (PI), fecal OPG was counted. Lesion scoring and Bursa of Fabricius (BF) weight were reported for D6 PI. Feed conversion PI (D14-20 and D14-28) was reduced ($P<0.05$) for YANG, doses 2 and 3 being the most consistent over time against NMI. Lesion scores for all 3 doses were reduced versus NMI. Specifically, ET was reduced for YANG dose 2, EA for YANG dose 3 and EM by all doses. A negative correlation was reported for OPG with YANG dose, EM being more sensitive to YANG doses 2 and 3. BF (% body weight) was higher for YANG dose 2 when compared to NMI. Coccidiosis related mortality was numerically but non-significantly reduced by all YANG doses. Specific nutritional solution such as YANG helps birds to mitigate coccidiosis negative effect, but the animal response looks dose sensitive, with 1000g/tonne as the most consistent.

Keywords: Coccidiosis, Eimeria, Yeast fractions

AviPro® IBD Xtreme vaccination in broiler farms in high risk area for very virulent infectious bursal disease virus

Abstract ID: 32

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Infectious bursal disease (IBD) might cause dramatic economic losses to the poultry industry especially in regions with presence of very virulent infectious bursal disease virus (vvIBDV). Vaccination programs that provide early protection against IBD are normally advised when there is a history of vvIBDV outbreaks or in epidemiologically uncertain areas. Therefore, less attenuated live IBD vaccines (intermediate plus or hot) that can be administered at an earlier age are normally used in such field conditions. The objective of this study was to evaluate the efficacy of AviPro® IBD Xtreme to prevent field vvIBDV infections in commercial broilers. This intermediate plus IBD vaccine contains the V217 strain that allows earlier vaccination due to its ability to overcome high maternally derived antibodies (MDA). Twenty broiler farms in Germany located in a very high density broiler production area with confirmed vvIBDV infection history were included in this study. Blood samples of day old broiler chickens were collected and tested for IBD specific antibodies with BioChek® ELISA kits. They presented high MDA level as were coming from parent stock vaccinated with live and inactivated IBD vaccines. Optimal day of vaccination was calculated in each broiler flock using Deventer formula and AviPro® IBD Xtreme breakthrough titer (636 ELISA units). Vaccination with AviPro® IBD Xtreme was via drinking water at 15 or 16 days of age. All broiler flocks were observed for clinical signs during the whole production period and zootechnical results were monitored. At the end of the cycle, from five birds per farm bursas were collected for Retrotranscription Polymerase Chain Reaction (RT-PCR) and sequencing of VP1 and VP2 genes. The V217 vaccine strain was detected in all bursal tissues and no clinical signs of IBD disease were observed in any of the broiler flocks. There was no evidence for the presence or influence of a field vvIBDV infection supported by the facts that no field IBD virus was found on the bursas, no clinical or pathological signs associated to IBD were observed and no negative influence was noted on the performance data. This field study shows that AviPro® IBD Xtreme represents an effective vaccination strategy to prevent field vvIBDV infections in broiler chickens. Additionally, serological results demonstrates that high breakthrough titer of AviPro® IBD Xtreme allows early IBD vaccination which correlates with early production of neutralizing antibodies.

Keywords: Gumboro disease, Infectious bursal disease virus, Live intermediate plus vaccines

Bacillus subtilis 29784 maintain performances of broilers in stressed conditions

Abstract ID: 436

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Clostridium perfringens (Cp) induces subclinical necrotic enteritis (NE) and causes important economic losses in the broiler industry. *Bacillus subtilis* 29784 was selected through an *in vitro* screening process showing potential to inhibit several strains of *Cp*. The present study aimed to assess the effect of supplementing diet with *B. subtilis* 29784 on growth parameters and livability of broilers using a model of microbiota dysbalance in comparison with another *B. subtilis strain (Bs A).* A well-established stress model inspired from the pharmaceutical industry was used. Cobb male broiler chicks were orally given *Eimeria maxima* oocysts on d14 and then with a virulent *Cp* strain once daily on d19, 20, and 21. 320 chicks were randomly distributed into 4 treatments with 8 cage replicates of 8 birds per treatment. The four treatments were: (1) Non-stressed chicks, non-supplemented diet; (2) stressed chicks, non-supplemented diet; (3) stressed chicks, *Bs* A (5.10^8 cfu/kg) supplemented diet; and (4) stressed chicks, *B. subtilis* 29784 (5.10^8 cfu/kg) supplemented diet. Performance (body weight gain, feed intake, and feed conversion ratio) and total mortality were measured from d0 to d28.
The induced disbalance was effective, as shown by a significant reduction of broilers' BWG (-36%), an increase of FCR (+42%) and total mortality (+20.3%). The animals receiving *B. subtilis* 29784 showed significant improvement of performance (+50.3% on BWG and -26.5% on FCR), compared to stressed animals, reaching the same performance than the non-challenged birds. The total mortality was also decreased. *Bs* A was also able to improve the performance and the livability of the stressed birds, but did not allow to fully restore the performance.
This study showed that *B. subtilis* 29784 is able to maintain the performance of broilers subjected to a strong stress. And, depending on the *Bacillus* strain, the level of performance can differ. This implies that strain specificity is an important criteria to be taken into consideration when selecting probiotics. Choosing those with strong robustness will help the animals to cope with a strong environmental stress.

Keywords: Bacillus, Clostridium perfringens challenge, Dysbiosis

Comparative Correlation between Bursa Bodyweight Index with different types of live Infectious Bursal Disease Vaccines in Broiler

Abstract ID: 171

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The weight and size of the primary lymphoid organs (Bursa of Fabricius and Thymus) are an important parameter can estimate the immunosuppressive in poultry. A safety investigation of IBD vaccines in the broiler is generally using a Bursa Bodyweight Index (BBI), the ratio of the Bursal weight to bodyweight. This study showed the investigation and comparison the correlation between the BBI with the different types of the live IBD vaccines (IBD Winterfield Strain and IBD Immune complex) in broilers in Thailand. Age of all healthy broiler samples between 26 to 30 days were randomly sampling from the broiler farms in the South, East, West and Central region of Thailand during January 2017 to January 2018 to measure the bodyweight and the Bursa of Fabricius's weight for calculation of the BBI. Two broiler sample groups were divided depending on IBD vaccine types and their vaccination programs in the hatchery and farms. The first group, 128 birds were only vaccinated with the IBD Immune Complex Vaccine at hatchery by subcutaneous injection and the second one, 122 birds were only vaccinated with the live IBD Winterfield Strain Vaccine on 14 days old via drinking water at farms. The BBI data of the broilers from both groups was analyzed statistically using Wilcoxan method ($P<0.05$). The result showed the BBI information of the broilers from both groups are statistical different significantly. The average BBI of the first (IBD Immune Complex Vaccine) and the second (live IBD Winterfield Strain Vaccine) groups are 1.07 ± 0.63 and 1.93 ± 0.91, respectively. This study concluded that the broilers vaccinated by the live IBD Winterfield Strain Vaccine at farms played the important role to enhance a good BBI which means less immunosuppression than the ones vaccinated by the live IBD Immune Complex Vaccine alone at hatchery.

Keywords: Bursa Bodyweight Index, IBD Immune Complex Vaccine, IBD Winterfield Strain Vaccine, Immunosuppression

Compared effect of copper-exchanged zeolite and brown macroalgae Ascophyllum nodosum on fecal excretion of Avian Pathogenic Escherichia coli in a chicken intestinal carriage model

Abstract ID: 186

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Avian colibacillosis ranks as one of the most commonly encountered bacterial diseases in poultry worldwide. Being the main reservoir of Avian Pathogenic *Escherichia coli* (APEC), the lower gastrointestinal tract and resulting birds' excreta play a critical role in APEC spread in poultry houses. In this context, we evaluated the effect of selected feed supplements on excreta APEC loads using an intestinal carriage model. Thirty two specific pathogen free day-old Leghorn PA12 chicks were orally inoculated with 1.13 x 10^9 CFU of an APEC strain (APEC O2: K1: H5) and randomly assigned to 3 treatments in a blind study. Chicks of the untreated control group (UC) were fed a basal diet, whereas chicks in the treated groups were respectively fed the basal diet supplemented with 0.5% of a mixture containing Copper-exchanged Zeolite (CeZ) or 0.5% of dry *Ascophyllum nodosum* biomass (AS). Excreta samples were collected at different days (d2, d5, d7, d12, d14, d19, d21) and fecal shedding of total *E. coli* and APEC were monitored by bacterial counts. For bacterial counts, statistical differences between the groups were tested by Kruskal-Wallis test with pairwise comparisons. Differences in birds' growth were tested by one-way ANOVA followed by Tukey's post-hoc test. Inoculated APEC represented the majority of excreted *E. coli* until 14 days of age (over 80%). Unexpectedly, beyond the intestinal carriage, APEC oral inoculations led to some cases of colibacillosis infections with a total mortality rate of 19%. Compared to the UC, both CeZ and AS did not significantly alter APEC and *E. coli* excretion over the period d0 - d14. CeZ significantly reduced fecal shedding of APEC at d19 by 1 $\log_{10}$ CFU/g ($P = 0.007$) and the reduction was maintained numerically at d21 by 0.8 $\log_{10}$ CFU/g. AS treatment did not significantly alter fecal APEC counts at d19 and d21, but numerically reduced total *E. coli* counts at d19 by 1 $\log_{10}$ CFU/g ($P = 0.05$) and significantly at d21 by 1 $\log_{10}$ CFU/g ($P = 0.035$). However, the final weight of birds fed AS was reduced by -19 % ($P = 0.004$) compared to the UC, which could indicate a negative impact on growth. Overall, these results suggest that CeZ and AS could be interesting ingredients to contain the fecal shedding of pathogenic *E. coli* and limit their propagation in poultry houses, particularly after the first weeks of life.

Keywords: APEC intestinal carriage, Ascophyllum nodosum, Copper-activated zeolite, Fecal excretion, Poultry

Comparison in ossification zones between broiler and dual-purpose breeds

Abstract ID: 147

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Bone damage in broilers has been a widely studied subject over the years.The main factor stated as the cause of this is the continued genetic selection that broiler lines have been subjected to throughout the years, with the purpose of increasing growth performance and carcass quality and composition. This study aimed at investigating histological differences in the ossification of long bones in broiler strains which might help explain the higher incidence of bone lesions in broiler lines. Fifty Cobb birds of at slaughter age (50-55 days; average 1.5 kg) and 5 Rhode Island Red exemplars of the same age (average 700 g) were used. Full-thickness sections of femoral head cartilage and bone from both legs were collected and fixed in 10 % buffered formalin, subsequently decalcified using a commercial solution and later routinely processed for histopathological analysis. Histology slides were observed and photographed using an optical microscope. The different ossification zones were divided into five layers: joint/reserve cartilage, proliferation zone, hypertrophic zone, superficial ossification zone (<30% ossified matrix) and deep ossification zone (<30% remaining cartilage). These were measured against the total thickness of the femoral head ossification center using appropriate software and the obtained data subsequently statistically treated. As expected, the comparison between the left and right femoral heads of each bird showed no statistically significant (95% confidence interval) difference in ossification which could have proven a confusion factor. Comparison between the thickness of the various layers of ossification in the dual-purpose and the broiler group resulted in statistically significant differences between the hypertrophic zone and the deep ossification zone of the samples tested, both of which were considerably more extensive in the broiler group. This study indicates that broiler strains, which are privileged for their fast growth and weight gain will tend to rest considerably more weight on bones which are made fragile by a thicker layer of hypertrophic cartilage (poor in extracellular matrix) and only partially-ossified bone, in comparison to the slower-growth strain used for comparison.

Keywords: Broilers, Cartilage, Dual-purpose, Histology, Ossification

Comparison of gut microbiota in hens

Abstract ID: 600

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Changes in the composition of the intestinal microbiota in the cecum of poultry could have a direct impact on the quality and safety of poultry products. This study presents the results of comparative molecular genetic analysis of the cecal microbiocoenoses in laying hens of two commercial crosses, Hisex Brown and Lohmann Brown, during ontogeny. According to the analysis of overall taxonomic representation, more than 70% phylotypes determined can be attributed to three phyla, Firmicutes, Bacteroidetes, and Proteobacteria. Less represented were Actinobacteria, Tenericutes and Fusobacteria, and the presence of significant amounts of unidentified bacteria was also revealed. During ontogenesis, birds exhibited marked changes in the ratio of the number of phylotypes and taxonomic groups of the intestinal microbiota. Chickens of both crosses went through several stages in the development of microbial communities, including a stabilization period at the age of 20 to 40 weeks, as evidenced by the biodiversity assessment using ecological indexes. The stabilization period was characterised with a significant increase in representatives of class Clostridia involved in the metabolism of carbohydrates, and in bacteria with high antagonistic properties (genera ***Lactobacillus*** and ***Bacillus***). There was also a significant reduction of number of opportunistic and pathogenic taxa, such as families ***Campylobacteraceae*** and ***Enterobacteriaceae***, order Pseudomonadales, and phylum Tenericutes. Despite the similar conditions of housing and feeding, the Lohmann Brown hens had a maximum level of representatives of the normal flora observed by 40 weeks of age. This probably determines a smaller number of pathogens like ***Staphylococcus***, family ***Campylobacteraceae***, and phyla Tenericutes and Fusobacteria found by 40 to 60 weeks of age and greater stability of intestinal microbiocoenosis in the Lohmann Brown birds as compared with the Hisex Brown chickens.
This research is supported by a grant of the Ministry of Education and Science of the Russian Federation, Contract No. 14.W03.31.0013.

Keywords: Gut microbiota, Hens

Detection of Mycoplasma synoviae infections in vaccinated broiler breeder flocks

Abstract ID: 35

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Mycoplasma Synoviae is a major poultry pathogen which causes significant economic losses to the poultry industry. The aim of study was detection Mycoplasma synoviae infections in vaccinated broiler breeder flocks and their progeny. For this purpose, from 20 broiler breeder farms and their progeny 2 times in 30 and 60 weeks of age 20 tracheal swab and 20 blood samples from each farm were collected. All 20 broiler breeder farms before vaccination against Mycoplasma Synoviae were confirmed free from Mycoplasma synoviae infections. Tracheal swab transferred to cap tubes containing PPLO broth media and kept at 4°C less than 24 hrs send to laboratory for further proceeding. Antibody against Mycoplasma Synoviae was measured by rapid serum agglutination (RSA) and Elias tests. Tracheal swabs were examined by high resolution melting PCR (HRM PCR) for detection Mycoplasma Synoviae field isolate. The results showed that vaccination broiler farm against Mycoplasma Synoviae up to 40 weeks could protect all broiler breeder farms from Mycoplasma Synoviae field infection and produce good antibody titers. With increasing age and at age 60 weeks one farms and its progeny was positive for Mycoplasma Synoviae which was differ from Mycoplasma Synoviae included in live vaccine. This study showed vaccination by present vaccine against Mycoplasma Synoviae in broiler breeder farms could not protect older broiler breeder farms in older ages and probably Mycoplasma Synoviae infection transferred to progeny. Therefore vaccinated farms should be monitor by molecular methods.

Keywords: Broiler breeder, ELISA, HRM PCR, Mycoplasma synoviae, RSA

Dietary ferric tyrosine affects broiler chicken performance, intestinal health and Campylobacter counts exposed to natural Campylobacter jejuni challenge

Abstract ID: 89

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Campylobacter is an important pathogen commonly found in chickens that can cause severe acute gastroenteritis in humans. Despite intensive efforts to prevent food-borne transmission of Campylobacter, no effective strategy exists to reduce Campylobacter loads in farmed broilers. This study examined the capacity of a novel feed additive to lower Campylobacter jejuni populations and to improve growth efficiency of broiler chickens. A total of 384 male one-day-old broiler chicks were used in a 42-day trial. They were randomly allocated into four treatments with six replicates (16 chicks per pen). Control group (T1) was fed basal diets (Starter 1-21 days; Grower 22-42 days) in mash form. The other three groups were fed the same basal diets further supplemented with ferric tyrosine at 0.02 g/kg feed (T2), 0.05 g/kg (T3) or 0.20 g/kg feed (T4). Ferric tyrosine is an organo-iron complex of iron (III) with L-tyrosine (4-hydroxyphenylalanine). Diets did not contain any other added iron compounds, coccidiostats or antibiotics. Feed and water were provided ad libitum. At day 20, broilers were exposed to natural C. jejuni challenge by means of contaminated litter from a commercial farm. Faecal swabs, caecal and litter samples were taken on days 25 and 42 on trial, to confirm the presence of C. jejuni using PCR amplification and conventional techniques. After 42 days on trial all birds were slaughtered and faecal and intestinal samples collected for further analysis. Dietary supplementation with ferric tyrosine did not affect performance parameters at the examined levels of this trial. At day 25, pen litter samples analysed positive for C. jejuni, with significantly ($P \leq 0.001$) lower C. jejuni counts for the T4 group, compared to groups T1, T2 and T3. At the end of the study, C. jejuni litter counts did not differ significantly among groups, but C. jejuni counts in bird caeca were significantly ($P=0.004$) reduced, by 2 log10 in the T4 group, compared to the T1 Control and T3 groups. On day 42, diarrhoea was observed on the litter in only 1 of 6 pens in the T4 group, but in 5 of 6 pens in the T1 control group. In conclusion, these data suggest that ferric tyrosine in the feed at 0.20 g/kg feed exerts a protective effect against C. jejuni. Acknowledgements: This research was financed by Akeso Biomedical, Inc., USA.

Keywords: Broiler chickens, Campylobacter jejuni, Ferric tyrosine, Growth performance, Intestinal health

Dried algae rich in beta-(1,3)-glucan as a replacement for in-feed antibiotics

Abstract ID: 435

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In response to the European ban on antibiotic growth promotors (AGP) in 2006 and the increasing pressure to reduce therapeutic antibiotic usage, alternatives such as probiotics, phytogenic substances and natural immune modulators are gaining traction in the industry. Beta-glucans have been known for their immune modulating effects. They have been demonstrated to stimulate both specific and non-specific immune responses and are added to feed to enhance animal performance and resistance to diseases. To date, the most common source of β-glucans are branched β-glucans incorporated into the cell wall of yeasts. In contrast, the current research evaluates an algal source of β-glucan. The algae *Euglena gracilis* produces a highly bioavailable linear non-cell-wall-bound β-(1,3)-glucan as a storage carbohydrate.
Earlier *in vitro* experiments have shown an increased secretion of cytokines by macrophages (eg. TNFα and IL-10) and an increased recruitment of immune cells to the intestine in response to the algal β-glucan. In the broiler performance trial described in the current research, the possibility of replacing an in-feed antibiotic with a dried algae product in broiler chickens was assessed. In total, 432 one-day-old male broiler chickens (Ross 308) were randomly assigned to 48 pens (18 birds/pen x 8 pens x 3 treatments). The treatments consisted of a control treatment with the AGP Zn-bacitracin (50 mg/kg) and two treatments with dried algae product (Aleta^{TM}, 50g/T and 100g/T). Challenge was induced in the trial by feeding the broilers a diet with a high concentration of low digestible protein (fishmeal), a high concentration of fiber (wheat and barley) and by re-using litter from a commercial farm. Broiler performance during starter (0-10d), grower (11-21d) and finisher periods (22-39d) was recorded.
Overall, no significant differences were observed between the birds fed the AGP and the birds fed the dried algae. The supplementation of the highest dosage (100g/T) of dried algae resulted in a body weight and FCR of 2319 g and 1.658 respectively, whereas the addition of the AGP resulted in a body weight and FCR of 2257 g and 1.680 respectively. It can be concluded that the dried algae were equally effective as the AGP in maintaining animal performance during nutritional and environmental challenge conditions.

Keywords: Algae, Beta-glucan, Broiler

Drinking water vaccination with Gumboro 228E: monitoring by PCR and serology following extension of the period during which the vaccine solution is dispensed

Abstract ID: 517

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Infectious bursal disease (Gumboro disease) is one of the most problematical diseases for poultry producers. In addition to biosecurity measures, vaccination is the best means of prevention. When vaccination using an attenuated strain is carried out via the drinking water, if the chickens are not given enough time to drink it, this can cause a lack of homogeneity in the amount of vaccine ingested, and therefore poorer protection of the flock.
The authors monitored groups of broilers and breeders which had access to the vaccine solution for approximately 6 to 8 hours following a period of at least 2 hours during which water was withheld. Monitoring of the vaccination involved serology when the chicks were introduced, in order to determine the optimum age for vaccination, and checking the decrease in maternally derived antibodies by serology on the day of vaccination. The vaccination protocol was monitored by the veterinary surgeon who was present when vaccination was carried out. Finally, replication of the vaccine virus strain 228E was checked 3 weeks after vaccination by serology and by PCR Nobivet ^{O}Gumbo+ on cloacal swabs.
Extending the vaccination period improves vaccination compliance on the part of the farmer by simplifying the process and allowing savings in time. The increase in the number of vaccinated flocks and the improvement in the quality of vaccination allows protection of all the flocks to be improved within a production area.

Keywords: Infectious Bursal Disease, PCR, Vaccination

Effect of incorporating an emulsifier complex (DIGESTFAsT) on meat quality during the storage

Abstract ID: 181

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The objective of this study is to evaluate the effect of a biosurfactant (DIGESTFAsT) additive combining lysophospholipid emulsification activity with choline, L-carnitine and betaine, extracted from artichoke. A total number of 100 one-day-old JV chicks were allotted two groups of 7 pens with 7 chickens each. Two diets based on maize and soy bean meal, Control without additive) and Experimental (with the additive, 500 g/Ton), were prepared and each diet was distributed to a group. At the end of the fattening essay (39 days of age), 32 broilers (8 males and females from each diet) were slaughtered. After a six-days-storage period (+4°C), variations of the color, total phenol index, aerobic bacteria and fecal and total *coliform* were analyzed on breast meat samples. Our findings show that breast meat color measured after (24h) was similar in both groups. However, after 6 days of storage the luminance index in E lot was higher than that T ($P = 0.038$), while total phenol content in breast meat increased ($P = 0.04$) in E birds. The analyzed bacteria after storage were higher in the control group ($P<0.026$). Moreover, it the feed supplement tends to reduce ($P = 0.08$), meat oxidation during the storage period, in chickens receiving the diet E. In conclusion, the emulsifier complex preserves breast meat quality by reducing the multiplication of total germs, inhibiting its oxidation and increasing total phenol content.

Keywords: Broiler, Emulsifier, Meat quality, Storage

Effect of the Saccharomyces sp. and Bacillus subtilis based probiotics on broiler chickens performance and caecum microbiome community

Abstract ID: 544

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Bacillus sp. and *Saccharomyces sp*. are considered to be prospective as probiotics due to their antibiotic synthesize ability and some other properties. The aim of this research was to evaluate the effect of the probiotics on the broiler chickens. With use of molecular-genetics methods (T-RFLP and Real-time PCR) there were evaluated the amount and composition of caecum microbiome in 37 days old broiler chickens Cobb 500. Chickens of the Group I were fed with balanced compound feed from the first day. Chickens of the Group II were fed with the same compound feed and addition of probiotic on the base of derivates from live cells of *Saccharomyces sp.* In the Group III from the first day as a probiotic product was added Cellobacterin-T (BIOTROF Ltd, Russia, based on *Bacillus sp*.). Taxonomical domination of community belonged to the phylum *Firmcutes* including mostly *Clostridia* and less *Bacillus sp., Lactobacillus sp., Enterococcus sp*. Moreover various opportunistic and pathogenic microflora was found including avian respiratory disease causing *Pasteurellaceae, Mycoplasma sp.* and others. Addition of probiotics into the feed resulted in increasing of total microbiome size and decreasing of its biodiversity. Highest probiotical effect demonstrated usage of the Cellobacterin-T. In the Group III chicken microbiome community showed increase of *Bacillus sp.* amount on 1.38 times, *Lactobacillus sp*. amount on 1.47 times and also decrease of *Campylobacter sp.* amount on 3 times and family *Enterobacteriaceae* amount on 1.44 times compared to the control group. Yeast probiotic usage revealed a positive effect on *Clostridia* amount but also led to the rise of *Campylobacter sp., Pasteurella sp. and Mycoplasma sp*. amount. Usage of probiotics made positive effect on broiler growth and productivity rate. Highest growth rate, weight increase, FCR, vitamin A, E, B2 and carotenoid accumulation in liver showed the chickens of the Group III. Addition of the yeast probiotic to the feed stimulated better feed intake.

Keywords: Bacillus sp., Broiler chickens, Real-time PCR, Saccharomyces sp., T-RFLP

Effects of 2 forms of vitamin D on performance, leg health and coccidial invasion level in broiler flocks vaccinated and non-vaccinated against coccidiosis

Abstract ID: 561

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An experiment was conducted in 2 identical houses within the same farm over two production cycles to compare the effects of dietary supplementation with either D3 or 25-OH-D3 (5000 IU/kg). Each house had approximately 20 000 Ross 308 birds which were maintained for 42 days. In the first cycle birds were vaccinated against coccidiosis and in the second they received in feed coccidiostat. Birds in the first house received vitamin D3 and in the second house 25-OH-D3 over the first cycle and dietary treatments were alternated in the second. We hypothesized that 25-OH-D3 would result in improved performance and leg health, effects being more pronounced at increased prevalence of coccidiosis. Finally, we investigated whether 25-OH-D3 affected aspects of coccidial invasion and oocyst production. Performance indicators EEF and AWG for each house were assessed at the end of each cycle. 10 birds from each house within each cycle were euthanized at 15, 25 and 35 day of the cycle and their intestines were dissected for lesion scoring. Faecal samples were collected from the litter at 15, 25 and 35 days of age for determination of oocysts per gram (OPG). 150 birds were gait scored in each house at day 39 of each production cycle. Effect of vitamin D source was investigated with dependent t-tests. Performance, OPG, lesion scores and coccidial invasion were higher in vaccinated flocks. Although EEF and AWG were higher for 25-OH-D3 birds it did not reach statistical significance ($P > 0.1$). For all sampling days lesion scoring was numerically lower for 25-OH-D3 than D3, but it was significantly lower ($P < 0.001$) only for chickens that received coccidiostat at d35 of age. OPG was similar for both sources of vitamin D ($P > 0.1$). Gait score in vaccinated chicken was better for 25-OH-D3 than D3 ($P < 0.001$) but similar for chickens receiving coccidiostats ($P < 0.05$). In conclusion, offering 25-OH-D3 improved leg health to a higher degree in vaccinated flocks. In addition it reduced the degree of coccidial invasion but not OPG.
This work has received funding from the EU-FP7 PROHEALTH project
(grant agreement n° 613574)

Keywords: Broilers, Coccidiosis, Coccidiostat, Vaccination, Vitamin D

Efficacy of natural zeolite and glauconite dietary supplementation on carcass characteristics, gut pH and performance of broiler chickens

Abstract ID: 573

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An experiment with 300 one-day old Ross male broiler chicks was conducted to determine the effects of glauconite and zeolite on the broiler's carcass characteristics, gut pH and performance. In this experiment, glauconite added for first time in broiler diet as new feed additive. Five experimental treatments [control, glauconite (2 and 4 %), and zeolite (2 and 4 %)] were used in a completely randomized design with 4 replicates. During the experiment energy efficiency ratio (EER), protein efficiency ratio (PER), European efficiency ratio (EEF) and European broiler index (EBI) were measured periodically. At 42 days of age, one chick per replicate was slaughtered to determine carcass characteristics and gut pH. Analysis of variance and separation of means by Duncan's multiple range tests were conducted by SAS software. The results indicated that adding zeolite and glauconite to diet do not affected EER, PER, EEF and EBI at total rearing phase ($P>0.05$). Also, by adding 4% glauconite to diet, carcass percentage and breast relative weight was increased ($P<0.05$) in relation to control and 2% glauconite group. For thigh relative weight, there were no significant differences between treatments ($P>0.05$). Supplementation of diets with glauconite and zeolite did not have effect duodenum, jejunum and ileum pH at the end of experiment ($P>0.05$). In conclusion, we can use glauconite on broiler diet without any side effect whereas some beneficial effects were seen in carcass characteristics.

Keywords: Broiler, Carcass, Glauconite, Gut pH, Zeolite

Efficacy study of a live attenuated Newcastle Disease vaccine in SPF and commercial pullets

Abstract ID: 216

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Newcastle disease (ND) is one of the most important notifiable diseases presenting an enormous global economic impact. Vaccination combined with diagnostics and biosecurity are the key measures to control ND. Worldwide, the most commonly used ND vaccines are live attenuated vaccine viruses formulated with different strains as Hitchner B1 or LaSota, which are nowadays some of the most widely used vaccine strains. The main differences among those attenuated vaccines are the tropism and capacity to replicate in birds, which is highest in LaSota and results in higher levels of neutralizing antibodies compared to other strains. The efficacy of a live attenuated ND vaccine consisting of a ND LaSota strain, was examined in SPF and commercial pullets. In the first part of the study, SPF layers were individually identified and housed in three isolators based on two treatment groups and a control group. At 21 days of age, one treatment group was vaccinated via ocular and another treatment group via drinking water with the live ND LaSota vaccine. The control group remained unvaccinated. Blood samples were collected from the three groups at 21 days post-vaccination and tested for ND specific antibodies using Hemaglutination Inhibition (HI) test. All vaccinated and unvaccinated chickens were challenged at 21 days after vaccination with a pathogenic ND virus strain and observed for 10 days. All the chickens in control group died. No clinical signs of the disease were detected during the 10 days observation period and a sound seroconversion was observed in the two vaccinated groups. In the second part of the study, day-old commercial pullets derived from a layer breeder flock vaccinated with live and inactivated ND vaccines were individually identified and housed in isolators. Birds were vaccinated via ocular with the live ND LaSota vaccine at 35 days of age. Blood samples were collected from all pullets at vaccination day and at 7, 14, 28 and 42 days post ND vaccination. They were tested for ND specific antibodies with IDEXX® and BioChek® ELISA kits and HI test. The three serological methods showed high and homogeneous antibody response with the beginning of seroconversion at 7 days post-vaccination. The results of these studies confirmed the efficacy of the live attenuated ND LaSota vaccine applied via ocular or drinking water to induce strong humoral immune response and protect pullets from morbidity and mortality induced by ND virus infection.

Keywords: Newcastle Disease, Pullets, Vaccination

Efficiency of a specially modified bentonite to decrease the toxic effects of Aflatoxin B1 in ducklings

Abstract ID: 164

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Aflatoxins (AF) are secondary toxic metabolites produced by *Aspergillus flavus* and *Aspergillus Parasiticus*. Aflatoxin B1 (AFB1) is the most hepatic-toxic among AF. The symptoms of aflatoxicoses are loss of appetite, growth reduction, immunosuppression, liver damage, particularly in young poultry, and among them, ducklings are recognized to be the most susceptible specie. Bentonites are well known for their ability to bind and detoxify AFB1. However, bentonite's efficiency as detoxifiers may depend from their chemical structure and their geological deposit and origin. The aim of this study was to assess AFB1-binding ability of a commercial bentonite (CB) in comparison with a specially modified bentonite (SMB) by evaluating growth performance and total plasmatic protein (TPPs) in ducklings. A total of 120 one day-old Pekin ducklings were randomly assigned to 4 treatments, each consisting of 3 pens. A standard basal diet was used as a positive control (PC) while the same diet contaminated with 90 ppm of AFB1 served as a negative control (NC). NC diet served as a base for the two experimental diets: SMB diet = NC+2 kg/t of SMB and CB diet = NC+2 kg/t of CB. Individual body weights (IBW) and individual average daily gain (IADG) were recorded at day 0 and day 15. At day 15, all the ducklings were euthanized and blood samples were collected to perform TPPs analyses. Differences were studied by one-way ANOVA, followed by Tukey's post hoc test ($P<0.05$). At day 15, ducklings fed NC showed the lowest IBW, IADG and TPPs and statistically lower than PC ($P<0.001$). SMB achieved the same IBW, IADG than PC while for the same parameters CB showed a statistical lower performance both than PC and SMB ($P<0.001$). Concerning TPPs results, both SMB and CB were not able to reach the PC result. However, SMB showed higher TPPs level than NC ($P<0.001$) while CB showed the same TPPs concentration than NC. These findings showed the potential of SMB to limit the negative effects of AFB1 contamination in poultry.

Keywords: Aflatoxin, Bentonite, Ducklings

Efficiency of natural plant extract feed additives on slow growing broilers compare to coccidiosis vaccine

Abstract ID: 205

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More growing societal and government concerns lead to use alternatives in order to prevent coccidiosis in poultry. This trial aims to compare efficiency between two coccidiostatic alternatives: vaccine and natural feed additives. 4400 one day female JA757N were reared at 11 chicks/m^2 for 86 days. All animals were vaccinated with Bioral H120 (D14) and GUMBORO (D21 and 28). They received *ad libitum* a starter feed from D1 to D28, a grower feed from D28 to D56 and a finisher feed until slaughtering. Crossed experimental plan with 2 repetitions was designed as control group (group C), 2 cases of 1100 broilers vaccinated with coccidiosis vaccine at the hatchery; the experimental group (group E) of 2*1100 broilers, received supplemented feeds with plants extracts (phenols, phenylpropanoids, diallyldisulfides and polyphenols at 363 ppm in the starter and grower and phenylpropanoids and phenols at 58 ppm in finisher). Mean of weight, weight gain, feed consumption and mortality were recorded. At D29 and 43, 8 broilers in both groups were autopsied and oocysts count was performed according to MacMaster method. Oocyst counts were classified according 3 classes: low excretion (0-100 000 OPG), medium excretion (100 000-1 000 000 OPG) and high excretion (>1 000 000). Weights of group E were 0.28% higher than group C with a FCR 2.25% higher. Growth curves stay very similar throughout animal life without difference at any period. Mortality rates were numerically 0.4% lower for group E (1.57% vs. 1.18%; P=0.18). Both groups had 75% of broilers with a low excretion. At day 43, low excretion represents 31% for the group C instead of 38% for the group E; medium excretions are higher for group E (50% vs. 31%) and high excretion is higher for group C (38% vs. 13%). No significant difference was obtain on oocyst excretion (P=0.19). Moreover intestinal lesion score remain very low in both groups: 0 vs 0.25 at D19 for control and experimental group and 0 for both groups at D43. One hypothesis to explain the relatively high excretion without lesions in control group is the vaccine immunizes chicken for 5 strains of *Eimeria* and haven't activity against other *Eimeria* strains on the contrary to natural feed additives. This study indicates that both prevention strategies are comparable for *Eimeria* management on slow growing broilers and further studies should be launched on higher broiler densities.

Keywords: Broiler, Coccidiosis, Plant extracts

Elimination of poultry red mites (dermanyssus gallinae) by essential oils

Abstract ID: 302

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The objective of the experiments was to monitor mortality of poultry red mites (*Dermanyssus gallinae*), caused by plant essential oils (clove bud, lavender and cinnamon). The active ingredient in clove bud is eugenol ($C_{10}H_{12}O_2$), in lavender is linalool ($C_{10}H_{18}O$), in cinnamon is cinnamaldehyde (C_9H_8O). The acaricidal activity of these oils against poultry house-collected red mites was examined using direct contact method – by glass vial bioassay. Oils were tested at different concentrations (µL/cm^2). Oils were dissolved in water and Tween 85 and the solutions were spread on the filter paper at the bottom of the glass vial. The solvent of solution was then evaporated. Twenty movable poultry red mites, all their stages of development, were placed on the filter paper impregnated by oil at the bottom of the glass vials. The mortality of the poultry red mites in the glass vials was measured after 24h. All used oils caused mortality of poultry red mites. The highest mortalities were observed with clove bud and cinnamon, which caused more than 99% mortality of poultry red mites at concentrations 0.5; 0.25; 0.12 and 0.06 µL/cm^2. Cinnamon caused more than 90% mortality and clove bud caused 81.8% mortality at concentration 0.03 µL/cm^2. Lavender caused more than 99% mortality at concentration 0.5 µL/cm^2. At concentration 0.5 µL/cm^2 there was no significant difference between the efficiency of lavender, cinnamon and clove bud. Lavender caused less than 50% mortality at other concentrations. At concentration 0.015 µL/cm^2 clove bud and lavender caused less than 10% mortality. The results of these performed experiments suggest that clove bud, lavender and cinnamon essential oils are potential poultry red mites control agents.

Keywords: Botanical pesticides, Cinnamaldehyd, Eugenol, Linalool

Evaluation of sphinganine sphingosine ratio and liver weights of broilers exposed to Fumonisins

Abstract ID: 141

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Fumonisins (FUM) are mycotoxins produced by molds (*Fusarium* spp.), present in feed raw materials such as corn, and negatively affecting poultry after ingestion. As FUM are inhibiting the conversion of sphinganine (Sa) into sphingosine (So), part of the sphingolipid synthesis, the Sa/So ratio in serum has been proposed as a biomarker to evaluate exposure to FUM in farm animals. Additionally, liver damage can be noticed in FUM fed birds. The aim of this trial was to determine the effect of feeding a diet contaminated with FUM B1+B2 on the Sa/So ratio and liver weight in broilers. In addition, the effect of supplementing a mycotoxin binder to the diet was assessed. A 21-day trial was performed at the institute Samitec in Brasil. In total, 360 one-day old COBB broilers were divided over 3 treatments: a negative control diet, a 100 ppm FUM B1+B2 contaminated diet, and a FUM B1+B2 contaminated diet supplemented with a mycotoxin binder at 0.25%. At day 21, all birds were necropsied to determine liver weight and blood samples (n=12/treatment) were taken to evaluate the Sa/So ratio by HPLC-MS. FUM contamination significantly increased average Sa/So ratio and liver weight compared to the negative control group. Addition of the mycotoxin binder decreased the Sa/So ratio up to the level of the control group and decreased liver weight numerically. Results show that 100 ppm FUM negatively impacts broilers, as shown by the increased liver weight and the Sa/So ratio. The use of a mycotoxin binder could limit the impact of those FUM induced detrimental effects in broilers.

Keywords: Broiler, Fumonisin, Liver, Mycotoxin binder, Sa/so ratio

Evaluation of three serological methods to monitor humoral response of a live bivalent Newcastle Disease vaccine

Abstract ID: 215

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Newcastle disease (ND) is a highly contagious and lethal avian disease caused by certain strains of avian Paramyxovirus Type 1. The most important factors in the prevention of ND are the conditions under which birds are reared, the degree of biosecurity and the vaccination strategy. The most widely used ND vaccines include live attenuated vaccine viruses. They may be presented as monovalent ND vaccines or bivalent and polyvalent vaccines in which more viruses are included in the same commercial product. AviPro® ND-IB Sohol is a live bivalent vaccine containing the Newcastle disease virus LaSota strain, which is of lentogenic B1 type and the infectious bronchitis virus Massachusetts type, Holland strain. The objective of this study was to compare different serological methods for assessing the seroconversion profile against ND induced by AviPro® ND-IB Sohol under laboratory conditions. Thirty commercial pullets derived from a layer breeder flock vaccinated with live and inactivated ND vaccines were included in this trial. At their arrival, five of them were euthanized to measure the level of maternally derived ND antibodies. The other twenty-five day-old pullets were individually identified and housed in one isolator. At 5 weeks of age they were vaccinated with AviPro® ND-IB Sohol. Blood samples were collected from the pullets at vaccination day and at 7, 14, 28 and 42 days post-vaccination. All the samples were tested for ND specific antibodies with IDEXX® and BioChek® ELISA kits and Hemaglutination Inhibition (HI) test. The three serological tests showed high and homogeneous ND antibody titers and quick onset of seroconversion that was detectable at 7 days post-vaccination. Regarding the evolution of ND geometric mean titers the same trends were observed with BioChek® and IDEXX® ELISA kits: Antibody titers increased sharply from vaccination day until 2 weeks post-vaccination when they started to decline gradually. The HI test showed also increasing MT that started to decrease at 4 weeks post-vaccination. The results of the three serological assays showed very similar seroprofiles demonstrating that all of them are valid tests to assess vaccine take and monitor humoral immune response induced by AviPro® ND-IB Sohol. The early decline of ND antibody titers confirmed that vaccination of laying hens always requires the application of several doses to maintain immunity throughout their lives. In addition, this study demonstrates that AviPro® ND-IB Sohol vaccination elicits a sound seroconversion in pullets.

Keywords: Newcastle Disease, Pullets, Vaccination

Field study on production performances improvement in commercial meat turkeys by using a vector vaccine rHVT-ND compared with live vaccines program in France

Abstract ID: 62

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In turkey meat production, Paramyxovirus infections induce respiratory and digestive troubles, resulting on economical losses and poor production performances. In few cases, secondary infections as colibacillosis or Ornithobacterium rhinotracheale can occur and can affect flock performances as increase of mortality and antibiotic consumption. Vaccination with live vaccines is widely used but boost vaccinations are needed to have a consistent protection until slaughter. The aim of this study was to investigate the effects of a rHVT-ND vaccine (Vectormune ND®) on production performances. 262 flocks have been followed: 152 flocks vaccinated at farms (control group) with 1 to 3 live vaccines and 110 flocks (test group) vaccinated at day 1 in the hatchery with a rHVT-ND vaccine. Production performances of both groups (FCR, ADG, antibiotic consumption) have been compared with a statistical analyzed. As compared to the control group, an improvement on different criteria was observed in the test group as a mean body weight 312 g heavier at slaughter ($P = 7{,}74x10^{-5}$), a better mean ADG (107,31 g vs. 106,44 g) ($P = 2.6x10^{-3}$) and a strong decrease of antibiotic consumption of 40% ($P = 1{,}31x10^{-5}$). rHVT-ND vaccine is off label in turkeys but can contribute to improve prophylaxis against Paramyxovirus infection.

Keywords: Hatchery, Paramyxovirus, Production performances, Turkey, Vaccination

Genetic characterization of Avibacterium paragallinarum isolates from chickens in South Korea during 2011-2016

Abstract ID: 393

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Avibacterium paragallinarum (*A. paragallinarum*), a causative agent of infectious coryza, is an acute respiratory disease of chickens causing economic losses from retarded growth in growing birds and reduction (10-40%) in egg production in layers. Recently we obtained four field isolates of *A. paragallinarum* with different nicotinamide adenine dinucleotide (NAD) requirements including three NAD-independent isolates and an isolate with increased NAD dependency. Studies were needed for the genetic relatedness among different NAD requiring isolates. In this study, we genetically characterized the Korean field isolates of *A. paragallinarum* from chickens in South Korea during 2011-2016. The isolates (n=24) was identified by HPG-2 PCR known to be specific for *A. paragallinarum*. The genotyping by the enterobacterial repetitive intergenic consensus-based PCR (ERIC-PCR) and phylogenetic analysis of bacterial 16S ribosomal RNA (rRNA) and hemagglutinin antigen (hagA) genes of the isolates were performed using primers and conditions as previously described. Korean field isolates showed genetic differences among various NAD requiring field strains. Five ERIC-PCR patterns were recognized from Korean field strains and distinguished from those of eleven reference strains. The phylogenetic analysis showed that Korean isolates were divided into three distinct groups. Most isolates (20/24) showing typical requirement for NAD belonged to the same phylogenetic group as well as the same ERIC-PCR pattern. Our results indicate that the Korean field isolates have genetic diversity among strains and variants have their distinct genetic characteristics. Although the ERIC-PCR fingerprinting and phylogenetic anlaysis of 16S rRNA and hagA genes is not suitable for molecular serotyping, these could be used for molecular tools for epidemiological study for infectious coryza outbreaks.

Keywords: Avibacterium paragallinarum, ERIC-PCR, NAD, Phylogenetic

Husbandry factors affecting digestive microbiota diversity in organic broiler chickens

Abstract ID: 267

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Biodiversity of cecal microflora of organic broilers was assessed in 34 flocks reared under commercial conditions in France. In each flock, 5 healthy chickens were autopsied and 1 cecum per chicken was collected for a molecular fingerprinting analysis using Temporal Temperature gradient Gel Electrophoresis (TTGE). The mean of the similarity coefficients for one broiler with the 4 other chickens analyzed in a flock was then calculated (N=169). It was linked with the husbandry management and the broiler digestive health as described in an epidemiological questionnaire and in the autopsy. A multiple linear regression model was used with a random effect accounting for the flock effect. The similarity coefficient between a broiler and the 4 other ones in a flock was 58% (IC95% [54-61]). It tended to be lower when a broiler was infested by helminths (*Heterakis*) in ceca (55% [51-59] vs. 62% [57-67]) suggesting a relationship between cecal microflora diversity and parasitic infestation. The diversity of cecal microflora within a flock was more important in small organic farms where broilers are often reared in mobile houses with low level of biosecurity and drinking water from a private water supply. On the contrary the diversity was lower in farms with more standardized rearing management and technical support from a broiler production company. No relationship was observed between the cecal microflora diversity and the occurrence of digestive disorders in broilers or the broiler mortality rate.

Keywords: Digestive health, Microbiota diversity, Organic broiler

Influence of the phytobiotic additive on weight parameters, microbiome balance and immune response in broiler chickens

Abstract ID: 543

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A study aiming to investigate the effect of the phytobiotic additive containing essential oils on broiler chickens has been carried out in 2017-2018. In the vivarium was set up an investigation on 4 groups of broilers ("clean"control, infected control,"clean"group treated with the phytobiotic additive and infected group, treated with phytobiotic). Broilers in two groups were infected with the *Salmonella enteritidis* culture. Samples of litter for microbiome analysis by T-RFLP method (42 days chicken) and samples of ceca for immune-response genes expression by qPCR with ΔΔCt method (20 days chicken) were tested in the laboratory.
In the non-infected phytobiotic-treated group the content of "normal" microbiome representatives (cellulolytics, *Bifidobacterium sp., Bacillus sp., Lactobacillus sp., Selenomonas sp.*) increased on 26% compared to the control non-infected group. Among the infected broilers, the content of normal microbiome increased on 15% in the phytobiotic-treated group compared to the control group. The content of opportunistic enterobacteria, including *S. enteritidis*, decreased in the phytobiotic-treated group on 32% compared to the control group. The total share of opportunistic microbiome decreased in the phytobiotic-treated on 42% compared to the control group. Immune-response interleukin-6 and gallinacin-10 genes expression among experimental broilers in next after infection (day 20) was estimated with qPCR method. IL6 - anti-inflammatory cytokine- refers to signaling molecules. Gal10 - beta-defensin - plays a vital role in the innate immune response to bacterial infections. Expression of the IL6 increased on 1.62 times after infection with *S. enteritidis*. Adding phytobiotic into feed resulted in IL-6 expression increase on 3.41 times in non-infected group and increase on 4 times in infected group. Expression of the Gal10 gene increased on 1.84 times after infection. Adding phytobiotic into feed resulted in Gal10 expression increase on 17.51 times in non-infected group and increase on 2.1 times in infected group. Thus, there was revealed the positive effect of the phytobiotic additive on the poultry health (weight, microbiome, immune response) in infected and non-infected broiler chickens.
This work was supported by the state contract № 14.W03.31.0013.

Keywords: Broiler chickens, Genes expression, QPCR, Salmonella enteritidis, T-RFLP

In vitro evaluation of the anticoccidial potential of five ethanolic Tunisian plant extracts

Abstract ID: 177

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Resistance to coccidiostats and the negative effects of coccidiosis on poultry industry throughout the world have stimulated the researcher to look for alternative solutions of control, and the use of plant extracts is one of the alternative plans to treat or to prevent this disease. The aim of the present study was to evaluate 70% aqueous-ethanolic extracts at various doses (100-1000 µg mL^{-1}) of five selected plants collected from the southern region of Tunisia (leaves of *Artimesia herba alba asso*, *Urtica dioica*, *Ceratonia siliqua*, peel of *Punica granatum* and cloves of *Allium sativum*) on in vitro *Eimeria tenella* sporozoite invasion. The efficacy evaluation criteria, was the inhibition of sporulation (%). The oocystes used in this study were obtained after the autopsy of 36 day-old Hubbard broiler chicks. The harvested oocysts were provided 27.5°C temperature with 60-80 % humidity and continuous aeration.The obtained results have shown that all tested extracts have an ability to replace chemical anticoccidials (Amprolium). Sporulation inhibition rates at 1000 µg mL^{-1} concentration of *C.siliqua*, *A.herba alba asso* or *U.dioica* extracts were 66.75%, 61.73% and 62.37 %, respectively, higher than Amprolium (59.15 %). Extracts effects of *A.sativum* and *P.granatum* (1000 µg mL^{-1}) and *A.herba alba asso* and *C.siliqua* (800 µg ml^{-1}) were similar (P> 0.05) to Amprolium on *E.tenella* sporulation inhibition. Mean value of non-sporulated oocysts rates of all extracts ranged between 5.35% and 59.77% for 100 µg mL^{-1} to 1000 µg mL^{-1} doses, respectively. According to our findings, extracts of the five selected plants can be used as alternatives to the chemical anticoccidial (Amprolium) to inhibit *E.tenella* sporulation. However, their incidence is related to their concentrations. Furthermore chemical analysis and *in vivo* essays must be performed to detect the active biomolecules with anticoccidial effects and to evaluate efficiency and also toxicity on animals.

Keywords: Anticoccidial, Eimeria tenella, Ethanolic plant extracts, In vitro

In vitro techniques to evaluate anticoccidial properties of phytobiotics

Abstract ID: 256

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The aim of this *in vitro* assessment was to quantify the ability of oregano and thyme essential oils against *Eimeria* oocysts *in vitro* and to find out the mechanism of action involved in those oocysticidal properties. Several pharmaceutical products are being used in order to protect broiler chicken against coccidian infections. The anticoccidials have been added to the chicken diets for more than seven decades. However, the indiscriminate, overuse and misuse of anticoccidial drugs caused development of resistant strains of coccidian. Among other alternative solutions, phytobiotics, at relatively low concentrations could have significant benefits when used to treat livestock against coccidiosis: good palatability, low cost, less toxicity, and fewer residues. Although, there are several substances with potential antiparasitic activity, it is necessary, the effectiveness of those alternative substances to be screened by different *in vitro* techniques before *in vivo* evaluation with challenged birds in experimental trials. To examine the *in vitro* anticoccidial effects of different phytobiotics it is necessary to assess, i) the ability of phytobiotics to destroy *Eimeria* oocysts and ii) to study the *in vitro* inhibition of *Eimeria spp* sporozoite invasion into host cells by phytobiotics. In the first technique, the followed steps were used: isolation, purification, counting of *Eimeria* oocysts, identification of *Eimeria* species and determination of the LC50 of the tested phytobiotics. The effect of phytobiotics on the oocyst destruction in parallel with the release of substances absorbing at 273 nm after treatment is provided. The treatment of *Eimeria* oocysts with the oregano and thyme oil, led to their lysis in a dose and time dependent manner as shown by the release of substances absorbing at 273 nm. In the second technique, a parasite invasion assay was conducted to inhibit invasion in Madin Darby Bovine Kidney (MDBK) cells in 2 different levels. Each essential oil was added together with sporozoites to MDBK cell cultures and invasion was evaluated after incubation for approximately 20h. Oregano essential oil was found to be a more effective inhibitor of parasite invasion up to 50%, for similar concentrations compared to thyme oil. Furthermore, toxic effect against cell culture is less severe by thyme oil in similar concentrations. *In vitro* techniques are fast and inexpensive tools to screen phytobiotics in different levels or combinations as alternative anticoccidials.

Keywords: Anticoccidials, Broiler chickens, In vitro techniques, Phytobiotics

New generation of inert substances in D. gallinae control

Abstract ID: 580

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The dominant method of physical control of *D. gallinae*, the use of inert substances, has thus far relied on compounds based on SiO2, which have been applied in powder form or as a water suspension. The new generation of inert substances are oils (P 547-17 (patent application); the product contained (purified) paraffinic and silicone oil with an emulsifier), which are applied in the form of water emulsions. The combination of paraffin and silicon oils on non-absorbent surfaces creates a persistent layer, which shows a long and residual effect. The main characteristics established in the course of laboratory and clinical testing are the following: high efficacy to directly exposed specimens; an extremely long residual effect which is conditioned by the quality of the surface; no particular application device needed; suitable for machine-industrial use; greater applicability to surfaces which possess fewer requirements in cage construction; higher penetrative ability with impurities; greater killing capacity; greater requirements for hygiene maintenance; possibility of use in biosecurity (treatment of transport cages). Optimal use requirements: empty, hygienically prepared facility with rest period, ideally in temperature conditions for mite activity with additional treatment of concrete floors with inert substance. In the case of optimised procedure, the ongoing clinical testing has so far shown that 7 months after the procedure, *D. gallinae* findings were negative, with early detection method.

Keywords: Control, Dermanyssus gallinae, Inert substance

Phylogenetic Analysis of Newcastle Disease Viruses during 2012 to 2017 in Taiwan

Abstract ID: 306

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Thirty Newcastle disease virus (NDV) isolates from domestic poultry and wild birds during 2012 to 2017 in Taiwan were genetically characterized. The phylogenetic analysis of the variable region of the F gene indicated that there are at least five genotypes of NDV circulating in domestic poultry and wild birds. Five isolates belonged to class II genotype I, four to genotype II, nine (isolated from pigeons) to genotype VI, five to genotype VII, and the remaining seven isolates belong to class I NDV. The isolates from poultry farms were classified into genotypes I, II, and VII. Based on the phylogenetic analysis and the motif of the fusion glycoprotein cleavage site, the genotypes I and II isolates were lentogenic NDV vaccine strains. All of the genotype VII velogenic isolates belong to the sub-genotype VIIe, and these results suggested that sub-genotype VIIe NDV was a prevalent virulent strain circulating among chicken farms in Taiwan. In this study, half of the NDV isolates were collected from wild birds, and our results indicated that wild bird population carried avirulent NDV with genetic divergence regularly and may act as one of the important reservoirs.

Keywords: Newcastle disease, Paramyxovirus, Phylogenetic analysis

Prevalence of Mycoplasma synoviae in layer poultry flocks in Croatia

Abstract ID: 639

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Mycoplasma synoviae (MS) can cause serious problems on layer farms with drop in egg production and quality of eggs. Statistics confirm that more than 70% of layer farms in Europe are positive for MS. Once positive, flocks have to cope with recurrent problems and continuous losses during all production period together with high investment in treatment. Vaccination showed to be successful in elimination of the MS problem, using different vaccinal strains, but vaccination must be properly done and on time. Despite that, continuous monitoring should be established to determine possible threats for the farm as well as success of vaccination. Since farmers in the last two years continuously reported clinical signs of MS, aim of the study was to determine the prevalence of MS on layer poultry farms in Croatia. Around 300 blood samples from layer flocks were collected during 2017/2018 on 5 large layer farms previously non-vaccinated for MS. Sera were stored on -20°C till analyzed. Level of specific serum antibodies for MS were determined using commercial ELISA kit. Results showed high prevalence of MS on tested farms, especially in the last half a year, with over 80% of positive flocks. Average titres were in range of 2500 to 12 000. On multi-age farms usually younger flocks had lover titres with higher SD, while older flocks had higher average titres with lower SD. There were several cases of acute manifestation with poor egg quality and egg production decrease, what was confirmed by molecular diagnostics, together with simultaneous infection with *M. gallisepticum* (MG) on one farm. This survey confirms above mentioned statistics of continuous threat to layer egg production from MS and MG as well. This also confirms the need to modify the vaccination program and protect the production using available MS vaccines and to continuously monitor their impact on wild strain presence.

Keywords: Layer hens, MG, MS, Mycoplasma

Probiotic Properties of isolated lactobacilli from southwest and north west of Iran free range chicken intestine

Abstract ID: 174

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The purpose of this study was to isolate, identify, and investigate the probiotic properties of lactic acid bacteria isolated from the digestive tract of native domestic chickens from northwest and southwest of Iran, as potential probiotic bacteria with antibacterial activity against *Zoonoses.* In this study, two hundred and sixteen isolates of gram positive and negative catalase isolates of lactic acid were initially isolated. After an initial analysis of the ability to tolerate acid and bile, 13 isolates were selected to be identified using the 16 S rRNA gene sequencing. After that, the ability of thirteen isolated lactobacilli isolates to tolerate acid pH = 3 and the ability to control digestive tract *Zoonoses* was studied. The results of the study of the 16S rRNA gene showed that all isolates were belong to the Lactobacillus species. Sequencing results of the isolates were recorded in the Gene Bank with MG547723 to MG547735 access numbers. Of the 13 isolates, seven isolates were belong to *L. reuteri*, three isolates were *L.crispatus* and three isolates were *L. salivarius*. All of these isolates were able to tolerate pH = 3 for 3 hours with a decrease of less than one logarithmic unit. All isolates had tolerance to 0.3% bile and reached to more than 50% growth rate. Among isolates, two isolates of *L. salivarius* (*L. salivarius* ABRIIN39, *L. salivarius* ABRIIN27) and one isolate *L. reuteri* (*L. reuteri* ABRIIN31) had the ability to tolerate bile and reached to more than 90% growth rate. Three isolates belonging to *L. salivarius* (*L. salivarius* ABRIIN33 *L. salivarius* ABRIIN39; *L. salivarius* ABRIIN27;) were more effective to inhibit common *Zoonoses* bacteria. The result of this study revealed that *lactobacilli* isolated from the native chicken digestive tract have promising probiotics potential and some of them have good potential for inhibition of gastrointestinal *Zoonoses*.

Keywords: 16 S rRNA sequencing, Intestinal pathogens., Iranian Native chicken, Lactobacilli, Probiotic

Proof of concept of the efficacy of a modified live Escherichia coli vaccine in preventing cellulitis in an SPF broiler model

Abstract ID: 447

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Few studies have investigated the role of Avian Pathogenic *E. coli* in the pathogenesis of cellulitis since the seminal publication by Boulianne & Messier (2003). That study linked cellulitis in broilers to the ability of specific *E. coli* strains (including the O78:K80 serotype) to adhere to the skin through the interaction between type 1 fimbriae and fibronectin.
A challenge experiment was performed in day-old SPF broiler chicks with two O78 *E. coli* strains to assess the efficacy of an *E. coli* vaccination with a modified live vaccine in controlling cellulitis lesions. The spray vaccine was applied to 160 chicks according to manufacturer specifications, with 80 unvaccinated chicks as controls. Thirty minutes after vaccination, groups of 20 chicks were transferred to isolators (4 rooms with each 3 isolators for vaccinated chicks and one isolator for unvaccinated control chicks). At 14 days of age, all birds in room #1 were inoculated intratracheally with a high dose (10^9 CFU/bird) of an O78 reference strain known to consistently reproduce cellulitis. In room #2, all birds were inoculated subcutaneously (SC) with a low dose (10^5 CFU/bird) of an O78 reference strain known to consistently reproduce cellulitis. In room #3, all birds were inoculated SC with a low dose (10^5 CFU/bird) of an O78 field strain (29A) isolated from a commercial US broiler flock being with a high condemnation rate due to lesions. The birds in room #4 were not challenged. Birds were observed daily for morbidity and mortality until 21 days of age, when they were sacrificed, necropsied and scored for *E. coli* lesions (grade 0 = absence, to grade 3 = extensive signs). All birds that died before day 21 were also necropsied. The hypothesis was to observe a significant reduction in *E.coli*-associated lesions and/or mortality in the vaccinated groups as compared to controls (p value < 0.05, Fischer's exact test). None of the birds in room #4 showed grade 3 cutaneous lesions or died before being sacrificed. In room #1, a high level of mortality was observed within 24 h after challenge, in relation with the high inoculum titre. In rooms #2 and #3, grade 3 lesions and internal lesions were significantly lower in challenged vaccinated birds than in unvaccinated controls.
Under these experimental conditions, a modified live *Escherichia coli* vaccine was found to be efficacious in reducing the number of birds severely affected by *E. coli*-associated skin lesions.

Keywords: Broiler, Cellulitis, Modified live Escherichia coli vaccine

Retrospective evaluation of the economic cost of avian colibacillosis in broilers based on four years of records in a French production flow

Abstract ID: 448

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This descriptive retrospective epidemiological study assesses the impact of an avian colibacillosis (AC) outbreak on the technical and economic performances of broiler batches of a single production flow in France (159 production sites). Data collected were: the zootechnical performance, health history and meat inspection for all broiler batches slaughtered between February 2008 and December 2012 (n=3,702).
Three batch types were identified: i. standard production (slaughter at 41 days of age), representing 75.1% of batches, ii. slow growing (JA® type) production (39 days), representing 21.1% of batches and iii. production for export (34 days) for the remaining 3.8%.
Performance data included: mortality rate at 10 days of age, final mortality rate, final Feed Conversion Ratio (FCR, condemned birds excluded), slaughter weight, age at slaughter and condemnation rate. Whenever a disease or treatment was recorded in the flock information sheet of the batch, further diagnostic results were collected (sample, necropsy, bacteriological results and/or antibiotic sensitivity results); only batches with bacteriologically confirmed AC were considered affected. All other batches were considered as non-affected (NA). For each of the three production systems, AC-affected batches were compared to NA batches. Mortality and condemnation rate distributions were non-normal, and were analysed using the non-parametric Kruskal-Wallis test. FCR and weights were normally distributed (ANOVA analysis).
For the export production (12.2% AC prevalence), none of the performance/health parameters analysed seemed to be significantly affected by AC.
For the JA production (4.9% AC prevalence), mortality rate at 10 days and final mortality rate were the only parameters that were significantly increased (2.16% in AC group *vs.* 1.16% in NA group, p<0.001 and 3.35% *vs.* 2.29%, p=0.001, respectively).
For the standard production (25.4% AC prevalence), mortality rate at 10 days, final mortality rate and condemnation rate were also significantly impacted (AC: 2.01%, 5.06%, 1.16% and NA: 1.59%, 3.96%, 0.91%, respectively; p<0.001 in each case). The economic impact of AC was assessed using the data provided by the production flow management. These revealed increased final mortality representing an average 400 € loss per AC-affected batch for standard production, and increased condemnation rate representing an average 40 € loss. Also, age at diagnosis was significantly associated with increased 10-day and final mortality rates when AC was confirmed during the first week of age. When AC was diagnosed after 8 days of age, this was associated with significantly increased final mortality and condemnation rates.

Keywords: Broiler, Colibacillosis, Cost, Economic

Screening of antiprotozoal action of plants extracts: in vitro and in vivo results

Abstract ID: 71

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Demedication and natural healthy alternatives are clearly identified as long-term societal trends. One consequence in animal nutrition is to reduce the use of chemical coccidiostats. Phytogenic have already be proven to be efficient to prevent coccidiosis but their mode of action remain unclear. The objective of this work is to investigate and to compare the in-vitro and in-vivo antiprotozoal action of plants extracts used to replace chemical coccidiostats. For the in vitro experience, Trypanosoma brucei gambianse reference strain has been exposed to 6 different plant extracts in triplicate to determine antiprotozoal activity thanks two parameters: MEC (minimal concentration to kill 100 % of trypanosomes) and IC50(concentration inhibiting 50% of parasite growth). The in vivo experience consisted on two consecutive coccidiosis challenged trials where male chicks Ross 308 were reared in cages from 8 to 35 days. Each trial was composed of 2 replicates of 8 birds, 1 negative control and 3 experimental groups. A 1 mL inoculum/bird (E.tenella 200 000 ookysts; E.acervulina 10 000 ookysts; E.maxima 10 000 ookysts) was nebulized at day 14. For 3 plant extract genus(Acanthaceae, Allioideae, Lamiaceae), we observed simultaneously a strong in-vitro trypanostatic activity (IC50 values between 1 and 10 µg/mL), a statistically significant in-vivo reduction of ookysts at D26 (P<0,0001) and improved lesion index (0.33 vs. 2.64) and FCR(-8%,-6%-11% respectively). 2 plants extracts do not show correlation between the in vitro and in vivo experimentations. When Myrtaceae extract showed also a strong in vitro activity (6µg/mL), in vivo results such as lesion index (3.33 vs 3.00) and FCR (-2%) were not different from control. On the opposite Urticaceae extract showed very low in vitro trypanostatic activity (IC50 : 506µg/ml), all in vivo parameters (lesion: 0.67; FCR-8%) were strongly improved compared to control. In this study, we demonstrate selected plant extracts can help to prevent coccidiosis. However, the direct antiprotozoal action of most extracts as Acanthaceae, Allioideae or Lamiaceae seems to explain the improved performances; others secondary effects of plants must be investigate to explain the Myrtaceae and Urticaceae performances on broilers.

Keywords: Antiprotozoal activity, Coccidiosis prevention, Natural plant extracts

The effect of Lactobacillus acidophilus,Thymus vulgaris and acetic acid in broiler chickens challenged with Salmonella enteritidis

Abstract ID: 617

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The present study was conducted to investigate the effect of supplementation of *Lactobacillus acidophilus* (LA) with and without *Thymus vulgaris* aqueous extract (TV_{AE}) and acetic acid (AA) in drinking water on the performance, *Lactobacillus* spp. population, *Salmonella enteritidis* (SE) shedding and colonization of broilers challenged with SE from d 3 to 24. A total of 420 male broilers (Ross 308®) were randomly divided into 7 treatments (5 replicates per treatment) consisted of: 1-Control (CN), 2- Antibiotic (trimethoprim-sulfadiazine TS, 0.02%), 3-LA (0.02%), 4-AA (0.5%), 5-TV_{AE} (0.5%), 6-LA (0.01%)+ AA (0.25%), 7-LA (0.01%)+ TV_{AE} (0.25%). Administration of LA and LA+ TV_{AE} significantly improved weight gain during the study ($P < 0.001$). Broilers received AA had lower weight gain and feed intake and the least feed to gain ratio than CN ($P < 0.001$). Water intake was reduced in birds received AA and LA+ AA on d 3-6 and 13-15 ($P < 0.001$). LA+ TV_{AE} administration decreased SE colonization on d 15 in the cecum ($P = 0.04$). Also, on d 8, AA treatment ($P < 0.05$) and on d 15 ($P < 0.001$), all treatments decreased SE colonization in ileum compared to CN. Furthermore, on d 8, birds obtained TS, AA, TV_{AE} and LA+ TV_{AE} had higher *Lactobacillus* spp. population in the ileum ($P < 0.05$), but on d 15 only TS increased *Lactobacillus* spp. population in cecum compared to CN ($P < 0.01$). LA+ TV_{AE} administration reduced the faecal shedding of SE compared to CN on d 8 ($P < 0.05$). In conclusion, oral administration of LA+ TV_{AE} showed positive effects on the productive performance, reducing SE load and increasing *Lactobacillus* spp. population of challenged birds and probably have potential to replace as a new growth promoter instead of antibiotics in poultry industry.

Keywords: Acetic Acids., Broiler, Lactobacillus acidophillus, Salmonella enteritidis, Thymus vulgaris aqueous extract

Using aloe vera plant extract in ovo injection and drinking water on the immune system and digestive tract bacterial populations in broilers chickens

Abstract ID: 176

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In this study, 540 fertile eggs from broiler chicks Ross 308 used in a completely randomized design with 6 treatments and 5 replications. The treatments were: 1) non injection treatment (negative control: NC), 2) distilled water injection (positive control: PC), 3) injection of Aloe Vera gel extract at 100 ppm; 4) injection of Aloe Vera gel extract Level of 200ppm, 5) Injection of Aloe Vera shell extract at 100 ppm and 6) Injection of Aloe Vera shell extract at a level of 200 ppm. Experimental solutions were injected to the air sac of the eggs with using 13 mL insulin syringes at No. 22, and the eggs were placed in standard incubation conditions. In the next stage, the chicks were fed in floor up to 42 days (10 bird /replications) under the same standard diet conditions (according to the cataloge2007 Ross 308 recommendation). To evaluate antibody titers against SRBC, 0.1 mLof 5% SRBC solution was injected on each day of the experiment on days 18 and 35 of the breast muscle, and then carried out on taken blood samples in 22 and 42days. Blood samples were taken to determine the antibody titre against Newcastle virus, white blood cell count, and serum lipoprotein levels. On the 42 day, the immune organs (such as the Spleen, Thymus and Bursa fabrisius), liver, heart, and abdominal fat were weighed after the slaughter of broiler. Statistical analysis was performed using SAS software and GLM procedure. Comparison of the meanings was done using Duncan's multiple range test. The results showed that the weight of abdominal fat was lower in chickens that received 100 µg of Aloe Vera gel or shell extract than chickens fed with control diet ($P < 0.05$). The use of Aloe Vera gel extract significantly reduced LDL and total cholesterol levels. However, there was no difference in the concentration of triglyceride and plasma HDL in the treatments. The antibody titer response to SRBC and Newcastle disease and influenza vaccines were not affected by experimental treatments ($P > 0.05$). Birds that received in ovo injection of 100 micrograms Aloe Vera gel extract from the second week to the end of breeding period had a higher growth rate than other treatments. The results of this experiment indicate that the use of Aloe Vera gel extract improves the immune system in broiler chicks.

Keywords: Aloe vera plant extract, Blood parameters, Broiler chickens, Immune system, In ovo injection

Using a RT-PCR to identify birds vaccinated with a recombinant turkey herpes virus (HVT) vectored infectious laryngotracheitis (ILT) vaccine.

Abstract ID: 387

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Advances in vaccine technology have added new perspectives to the poultry sector, so much so that the number of recombinant vaccines has steadily increased. Diagnostics needs to adjust to this trend and tools and methods for assuring the quality of vaccines throughout the delivery pathway must be identified. The aim of this trial was to find the best timing and the best matrix to detect a recombinant turkey herpes virus (HVT) vectored infectious laryngotracheitis (ILT) vaccine using a PCR specific to HVT. Thirty-five chicks from commercial laying hens were subcutaneously vaccinated in hatchery with one dose of HVT-ILT vaccine and 2 doses of Rispens Marek's vaccine. Chicks were transported to the lab and housed in a BSL3 isolator. Starting from 10 days up to 40 days of age, 5 chicks were randomly selected and humanely euthanized, every 5 days. Spleens, blood and feathers were collected from each animal and singularly analyzed. Total RNA was extracted from homogenized feather pulps and spleens as well as peripheral blood mononuclear cells using a commercial kit following the producer's protocol. HVT quantitative Real Time RT-PCR (qRRT-PCR) was performed. Ten days post vaccination (p.v.), only 1 bird tested positive; 15 days p.v., the birds were all positive for at least one matrix; at 20 and 25 days p.v., 3 out of 5 birds tested positive; at day 35 p.v., the positive birds were 4 out of 5; at 30 and 40 days p.v., all birds were positives for at least one matrix. Comparing the average loads of virus DNA in the different matrixes, the highest value was recorded in the feather pulps followed by the spleens and blood. Between 30 and 40 days p.v., 14/15 (93%) of the birds resulted positive for at least 1 matrix, in particular 12/15 (80%) birds had both positive feather pulps and spleens. Considering these findings, we assume that the recommended window to detect this HVT-ILT vaccine virus is 30-40 days p.v. and that the most preferable and practical tissue to collect is the feather pulp. Further studies will follow to improve the sensitivity and the specificity of the diagnostic test used and to evaluate its use under field conditions.

Keywords: RT-PCR, Recombinant HVT vaccine, Vaccination

Vaccination against adenoviral gizzard erosion in chicken using a novel antigen delivery system

Abstract ID: 430

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Antigen delivery systems into chicken antigen-presenting cells represent an important novel strategy in vaccine development. In this study, we verified the ability of fowl adenovirus antigens fused with streptavidin to be targeted by specific biotinylated monoclonal antibody (Ly75) into dendritic cells and induce virus-specific protective immunity. Specific pathogen-free chickens were used in the study. Field isolate from local outbreak of adenoviral gizzard erosion was obtained and genetically analyzed and identified. Hexon protein and envelope protein IIIa peptides based on the analyzed virus were produced in *E. coli* and used as antigens in the antigens-streptavidin-antibody immunization complex. The isolated virus was also used as challenge infection for immunized chickens and as a reference control. Nine days post-challenge chickens were sacrificed and koilin and mucosal erosions were macroscopically evaluated and scored. The immunized group showed a significant decrease in infection symptoms with average score of 0.9 in comparison to average score 2.1 in the non-immunized group. The control group that was administered the bare streptavidin-antigens complex showed an average score of 1.1. Based on the results we conclude that our system is able to induce partial protective immunity against fowl adenovirus and may support further investigation in the area.

Keywords: Adenovirus, Antigen delivery system, Dendritic cells, Gizzard erosion

Yeast cell wall effects on immunity, gut integrity and performance of broilers

Abstract ID: 465

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A study was performed to evaluate the effects of the yeast cell wall (YCW) compared to zinc bacitracin (ZBC) on the immunity, gut integrity and performance parameters of broilers. For this, 504 male Hubbard® chicks (1 day old) were distributed in a completely randomized design with 3 treatments: 1–Control; 2–Control with ZBC (50 g/MT); 3-YCW (from *Saccharomyces cerevisiae* at 0.5 kg/MT, ImmunoWall® product from ICC Brazil Company), with 14 replicates of 12 birds each. The diets were divided into: pre-initial (1-7 days); initial (8-21 days); growth (22-33 days) and final (34-42 days). The birds were housed in pens with reutilized litter from a commercial farm (2nd time used). The BWG, FI, FCR and Production factor were measured at 7, 21 and 42 days. At 21 days, 8 birds per treatment were selected and slaughtered to collect the ileum. The samples were prepared for histology and immunohistochemistry analyzes and were evaluated macrophages, CD4+, and CD8+ cells count; lamina propria (LP) and epithelial thickness; enterocytes proliferation; epithelial plasma infiltration; mixed inflammatory infiltration of LP; goblet cells; congestion and necrosis. These parameters were qualified by "I See Inside" (ISI) index methodology (Kraieski, 2017). The data were analyzed by GLM produced from SAS and the means compared by Tukey test at 5% of significance. The effects were also analyzed by orthogonal contrasts by F test at 5% of significance. Significant differences ($P<0.05$) were found for immunohistochemistry, where the treatment with YCW supplementation result in lower macrophages compared to others treatments, and lower CD8+ cells count compared to ZBC treatment. For CD4+ cells count and ISI index (total), no statistical differences were found ($P>0.05$). However, YCW improved ($P<0.05$) the mixed inflammatory infiltration of LP and congestion and decrease ($P<0.05$) the goblet cells count compared to Control and ZBC treatments. The Control group decreased ($P<0.05$) the LP thickness. Regarding the performance parameters, the YCW improved ($P<0.05$) the FCR compared to the Control and ZBC. The supplementation of YCW in the broilers diet improved the FCR at 42 days and resulted in the best response of the immune parameters analyzed compared to others groups.

Keywords: Antibiotic, Immunohistochemistry, Nutrition, Poultry, Saccharomyces cerevisiae

Assessing an improved nestbox design for laying hens within a commercial setting

Abstract ID: 511

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The aim of the present study was to investigate whether nests in the middle of the nest row could be made more attractive to improve welfare and reduce mislaid eggs by installing partitions inside the nestboxes or on the balconies. The study was divided in two parts with different methods to improve the attractiveness of the nestboxes, each conducted on a single farm: 1) installation of wooden partitions inside a nestbox and 2) installation of wooden partitions on the nestbox balconies. In both experiments, aggression in front of the nest and the number of nest visits (number of nest entrances + number of nest leavings) was recorded by video observations on three consecutive days in the first five hours after the lights were turned on. Additionally, the total number of eggs per nest was counted. Using this design, a correlation between nest visits and eggs laid per nest and differences between nest visits in nests with or without partitions could be calculated.

The placement of partitions inside the nestboxes to improve the attractiveness of the nest resulted in ambiguous results (Experiment 1) where no correlation could be found between the number of nest visits and the number of eggs in the nest. Similarly,installation of partitions on the balcony as performed in Experiment 2 had no effect on the number of nest visits or the amount of aggression observed on the balconies. Most eggs were laid on the lower nest row level and in the front nests (end-nests) of the nest row. In conclusion, the installation of a partition inside the nest or on the balcony only had consequences on the number of nest visits but not on the number of aggressions or the number of laid eggs in the respective nests. Therefore, no improvements could be found with the installation of partitions inside the nestboxes or on the nestbox balconies in respect to animal welfare.

Keywords: Aggression, Animal welfare, Laying hens, Nestbox, Partitions

Behavioural Parameters and Final Body Weight of Slow-Growing Broilers With and Without Access to Pasture Area

Abstract ID: 310

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The aim of the present study was to compare the behavioural parameters and final body weight of a slow growing broiler genotype with and without access to a pasture area. A total of 210 one day old male chicks of Hubbard ISA Red JA were used in the experiment. Each experimental group consisted of three replicates, each containing 35 male chicks. The chicks were fed with starter diet (22.0% CP and ME 12.8 MJ/kg) between the 1st and 28th days and a grower diet (20.00% CP and ME 13.2 MJ/kg) between the 29th and 84th days. Pasture area was covered with about 30% Alfalfa (Medicago sativa), 10% White clover (Trifolium repens) and 60% Perennial ryegrass (Lolium perenne) for chicks with access to pasture area. The behaviour of the broilers was compared by observing the behaviour of 3 randomly selected marked birds on video tape at 8th week and were monitored for eating, preening, drinking, spot pecking, feather pecking, walking-standing, resting- lying and other behaviour. Broilers were weighed at the end of the 8th week (84th day) to determine the final body weight. Data was analyzed using GLM Procedure of SAS. A higher percentages of eating and resting-lying behaviours were observed in broilers without access to pasture, whereas a higher percentages of drinking, preening, spot pecking, feather pecking and walking-standing were observed for broilers with access to pasture area. At the 84th day, final body weight was determined to be lower in broilers with access to pasture area than the others without access to pasture area (respectively 2780.4 g and 2986.8 g, $P<0.01$). This study clearly showed that accessing to pasture has a positive impact for behavioural parameters for broilers by increasing of activity and exhibiting of natural behaviours.

Keywords: Behaviour, Broiler, Pasture, Slow growing, Welfare

Feed additive sulfur to control ammonia

Abstract ID: 615

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The use of litter acidifying agents is a common practice in the USA and they are utilized to control ammonia levels, which is an important aspect in poultry management. In a previous study performed on clean bedding material, sulfur at a feed inclusion rate of 2.27kg/ton with or without the addition of sodium bisulfate at 22.67kg/92.9m^2 as a litter amendment reduced ammonia levels without impacting bird live performance. A follow up study was performed using built-up litter in place of fresh bedding to determine if this change impacted either ammonia levels or live bird performance. Eight hundred straight run broilers were randomly assigned to 32 pens (25 birds/pen). Inclusion levels for treatments containing sulfur and/or sodium bisulfate were 2.27kg/ton of feed and 45.36kg/92.9m^2 onto the litter respectively. The treatments were as follows: (Con)control, (S)sulfur, (S+SB) sulfur+sodium bisulfate and (SB)sodium bisulfate. SB was spread onto the surface of built-up litter in the assigned pens 24 hours before placement of the chicks. Live performance data was recorded at D0, D9, D30 and D38. Ammonia readings were taken at D9, D24 and D38. At trial termination (D38) birds were scored for footpad dermatitis, pen litter quality scored and litter was collected for moisture and pH analysis. Data was analyzed using the GLM procedure, if significant ($P \leq 0.05$) means were separated using Tukey HSD. There was no significance between treatments for AFCR on D9 or D38. Day 9 BW were significantly higher ($p<0.05$) for SB and Con as compared to S. However these differences were not maintained by D38. There was no significance between treatments for ammonia on D23 or D30. Day 9 ammonia values were significantly higher ($p<0.05$) for Con when compared to S+SB and SB. Litter pH was lower in S and S+SB compared to Con at D0. On D9, 23 and 38 the treatment with S+SB had significantly lower pH then Con. There was no significance between treatments on D38 for footpad dermatitis, litter scores, or moisture levels. Based upon the results, the addition of sulfur at 2.27kg/ton did not negatively affect BW, pH, footpad score or litter moisture as compared to the other treatments at D38.

Keywords: Ammonia, Broiler, Sulfur

Performance and eggs quality in layers fed with different percentage of Camelina sativa meal

Abstract ID: 197

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The seeds of camelina are a rich source of polyunsaturated fatty acids (33% omega-3) and antioxidants and, therefore, potentially able to fortify foods. The CAMFEED project aimed to enrich eggs with omega-3 and antioxidant compounds. These eggs were laid from hens fed with camelina meal (CM) obtained from *Camelina sativa* lines with a reduced glucosinolate content. CM contains about 15% of residual oil (about 1/3 of omega-3), a high amount of protein (35-40%) and a high content of antioxidants (vitamin E and polyphenols). Hy-Line® brown laying hens of 18 weeks of age were divided into six pens of 40 animals each reared under the same environmental conditions; the layers were fed with three different diets (isoenergetic and isoproteic) containing 0%(C), 10%(CAM10) and 20%(CAM20) of CM respectively. The number of eggs collected and laid on the floor was recorded weekly, as well as mortality and feed consumption. The level of animal welfare was assessed according to the Welfare Quality®. Weight of the egg, egg yolk, albumen, shell and the mechanical properties of the shell were evaluated on samples of about 40 eggs per pen. The eggs were sampled at different intervals and the evaluation of the nutritional quality based on omega-3 fatty acids and cholesterol was performed. Preliminary results showed a good persistency of deposition curve. Eggs production (eggs laid*100/number of hens at time of recording) was similar among the three treatments and according with the Hy-Line®. Welfare assessment didn't show any differences among treatments; injuries were almost absent, plumage and leg conditions were good. Omega-3 fatty acids in egg yolk showed an increment consistent with the increasing in CM in the diet (CAM10 = × 3; CAM20 = × 4). These results underlined the high adaptability of the hens to the housing systems and feed protocols.

The project was funded by Fondazione Cariplo.

Keywords: Camelina sativa, Egg quality, Laying hens, Omega-3, Performance

Practical experience with manipulable environmental enrichment material in modern poultry housing systems in Germany

Abstract ID: 479

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As part of a German-wide survey a representative number of poultry experts (39 experts) and owners (22 farmers) were involved to investigate the experience with manipulable environmental enrichment materials and techniques in modern poultry production. The study focused on the detailed description of the existing environmental enrichments, with special regard to inserting material, advantages and disadvantages, recommendations to the number of birds per material as wells as economic aspects. Data were acquired using a structured questionnaire, interviews and direct on-farm observations.
The evaluation of the housing conditions of pullets and laying hens showed that in addition to access dry litter, especially the use of foraging, organic environmental enrichment has proven itself to minimize the risk of behavioural disorders. Therefore freely suspended bales of alfalfa hay as well as pecking blocks offered elevated are attractive elements for hens. An animal/enrichment material ratio of 500-1,500/1 has been proven for each material. Other materials such as maize silage (up to 15g/animal) as well as grain (3-5 g/animal) offering daily per hand or automatically stimulate feeding and foraging behaviour.
Environmental enrichment is not commonly provided in conventional fattening broiler chicken systems. Therefore, provision of a loose and dry litter over the entire fattening period is important to stimulate pecking, scratching and dust bathing behaviour. Access to environmental enrichment is recommended in organic production systems as well as numerous labelling systems. Bales of straw and pecking blocks for every 1,000 broiler chickens are mandatory.
A special challenge is to find attractive environmental enrichments for turkeys. The behaviour of turkeys requires an individual flock-based offer. In general manipulable organic materials, such as hay, offered in baskets (1 basket for a maximum of 2,500 turkeys), or pressed large straw bales (each for every 1,000 turkeys) are recommended. Temporary use of non-functional materials (for example plastic elements, such as lids of PET bottles and metallic polished materials) has proven in practice to distract turkeys if behavioural disorders occur.
The survey showed that depending on poultry species the German poultry production systems used miscellaneous environmental enrichment strategies to provide animal welfare.
This research had been funded by the Association for Technology and Structures in Agriculture (KTBL) Darmstadt, Germany

Keywords: Animal welfare, Manipulable enrichment material, Practical survey

The use of optic fiber lightguides in cage batteries for laying hens

Abstract ID: 91

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The efficiency of optic fiber lightguides for cage-housed laying hens was studied on White Leghorn layers (cross "SP-789") from 120 to 260 days of age with intermittent lighting regime (2L:5D:3L:2D:3L:9D) and average light intensity 10 lux. Local light sources for control treatment were LED lamps, for experimental treatment optic fiber lightguides; local lamps were positioned above the egg-collecting troughs in all cage tiers. Overhead light sources were LED lamps with warm white specter (color temperature 2800-3200 K). It was found that mortality level in experimental treatment (1.0%) was lower compared to control while average live bodyweight in layers at 240 days of age (1644 g) was significantly higher (by 2.9%, $P<0.05$). Higher bodyweight promoted higher average egg weight in experimental treatment (58.3 g) by 0.7% in compare to control ($P>0.05$). Average egg production and egg mass output during the trial were similar in both treatments (103.2-103.3 eggs and 6.00-6.03 kg). However, lower mortality in experimental treatment improved egg productivity parameters calculated per initial hen: egg production (102.5 eggs) was better by 2.6%, egg mass output (5.89 kg) by 2.9% in compare to control. Average daily feed consumption in experimental treatment (112.1 g/hen/day) was lower by 4.7% compared to control. Lower feed consumption and higher egg output per initial layer improved feed conversion ratio per 10 eggs (1.30 kg) and per 1 kg of egg mass laid (2.23 kg) by 5.1% in compare to control. There were no significant differences in absolute and relative weights of the yolk (14.6-14.7 g and 24.6-25.0%), albumen (37.6-38.3 g and 63.8-64.6%), eggshell (64.-6.6 g and 10.8-11.2%), in eggshell thickness (368-369 µm), and albumen/yolk ratio (2.56-2.62); in the experimental treatments, however, the trends are seen toward the higher relative weights of yolk and eggshell, lower relative albumen weight, and lower albumen/yolk ratio.

Keywords: Laying hens, Light emitting diodes, Local lighting, Optic fiber lightguides, Productivity

The use of waste heat exchanger in commercial broiler production

Abstract ID: 126

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The regeneration of waste heat is a promising direction for the improvement of energy efficiency in the heating of poultry houses. The regeneration means partial recovery of heat transmitted into the poultry house for subsequent reutilization for the same purpose. This approach can provide significant energy savings. Poultry and especially fast-growing broiler crosses have higher metabolic rates in compare to other productive animals and produce more CO_2 and heat per 1 kg of live bodyweight: a chick with the weight 1 kg produces 1.44-2.34 liters of CO_2 and 8.1-10.1 Wt of heat per hour while growing piglet with the same weight produces 0.26-0.65 liters and 1.5-3.6 Wt, respectively. The analysis of the dynamics of heat supply and waste within a cage-equipped house for broilers proved that 75-82% of energy supplied will be transferred outside the house with the polluted air exhausted by the ventilation system. There are waste heat exchangers in the market which can recuperate 60-80% of this waste energy to heat the cold inlet air. These exchangers, however, are designed for the use with relatively undusted air (air dust loads up to 0.5 mg/m^3). Air dust load within a poultry house can reach 4-10 mg/m3; the dust is predisposed to the adhesion compromising the regeneration of used filters. Certain foreign companies produce plastic exchangers though capital investments in this case will be unreasonably high. An exchanger made of galvanized iron sheets was designed with relatively wide gaps between the exchange surfaces allowing easy flushing of the adherent dust with water stream. Production cost for this exchanger is 10-15 lower in compare to known serial models; this device can act with strongly dusted air mediums. The moisture from the outlet air condensing on the sheets flows down to a trough and then directed to the canalization. The testing of this exchanger within the standard systems of ventilation and heating in a poultry house resulted in the improvement by 30-35% of energy efficiency during cold winter season (average inlet air temperature during 5 months -7.2^{o}C). Since the production cost of this device is low the payback period is less than a year.

Keywords: Air dust load, Broiler chicks, Energy regeneration, Heat expenses, Ventilation

A bacillus subtilis probiotic can improve performance and welfare conditions of broilers

Abstract id: 426

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In Europe, the welfare of broilers is receiving increasing attention and footpad dermatitis (FPD) has already been used in several countries as an indicator of the welfare of broilers. FPD also affects farmer income. Wet and sticky litter are major causes of FPD. We hypothesize that the benefits on gut health by probiotics may improve litter quality and may thus partly represent a solution to decrease FPD. Thus, we evaluated the effect of *Bacillus subtilis* 29784 on performance but also on litter quality, occurrence and severity of FPD of broiler.
Three hundred and twenty day-old male chicks were randomly distributed into 2 treatments with 8 pen replicates of 20 birds each (density: 13.3 birds/m^2). All birds were housed in the same environmentally controlled house, with a wood shaving litter, and fed on corn-soybean meal based diets. The two treatments were: Control and Control + *B. subtilis* 29784 (1 x 10^8 CFU/kg of feed). The experiment was conducted until 35 days and broilers were euthanized with inhaled carbon dioxide gas. At 35 days, weight gain and feed intake were measured. Litter quality and FPD were scored (Welfare Quality® method, 2009). Mortality was recorded at every day. Data were analysed using ANOVA, and Tukey's test with a level of significance at 5%.
Mortality rate was not influenced by dietary treatments and was 4% in average. Probiotic supplementation significantly improved WG (+4.8%) and FCR (-2.9%). 60% of pens in the Control group had highly degraded litter (Score 4), whereas this condition was absent in the pens of the probiotic group ($P = 0.031$). The probiotic addition improved FPD scores, with 21% decrease of pens scored as "4, highly necrotic" ($P = 0.008$) and 11% decrease of pens with combined scores "3+4, highly and visibly necrotic" ($P = 0.014$).
As expected, the improvement of litter score resulted in an increase of FPD. Better litter condition could be at least partially explained by the positive effect of *B. subtilis* 29784 on gut health parameters, such as microbiota shifts and inflammation reduction. Therefore, the dietary supplementation of *B. subtilis* 29784 aimed to improve both performance and well-being of broilers.

Keywords: Bacillus, Broiler, Footpad dermatitis, Welfare

A method for improvement of the productivity in thermal-stressed broiler chickens

Abstract ID: 158

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The controversy between high productivity and low thermotolerance in broiler chicken is becoming an increasingly important problem in the light of ongoing global warming, and hence the search the ways of mitigation of negative impact of hyperthermia on the productivity of broilers also gains increasing importance. Four trials were performed on Ross-308 broilers to develop a method improving the thermotolerance of finisher broilers during hot season. At 4 days of age the chicks were subjected to controlled hyperthermal condition (temperature was 4^{o}C above the recommended level) during 12 or 24 hours as a thermal training; control treatment was not trained. During the last week prior to the slaughter (at 35 or 38 days of age) the thermal stress was induced in all treatments (8^{o}C above the recommended level). In trials 3 and 4 drinking water for broilers during the last week of age was supplemented with KCl (0.5%). Mortality levels after finisher thermal stress in thermally trained (for 12 and 24 hours) treatments were lower by 2.8 and 5.7%, respectively, in compare to untrained control; live bodyweight higher by 2.3 and 4.2%, ADWG higher by 2.3 and 4.3%; FCR better by 5.9 and 11.3%, EPEF by 11.7 and 24.5%. Combinations of thermal trainings with KCl led to lower mortality levels compared to control (by 2.0%), higher live bodyweight (by 4.6 and 9.7%) and ADWG (by 6.9 and 12.4%), better FCR (by 6.1 and 9.9%) and EPEF (by 13.4 and 22.9%). Training for 24 hours improved the digestibility of nutrients from the finisher diet compared to control and 12-hour training: crude protein by 3.07 and 0.36%, crude fat by 1.51 and 2.19%, free-nitrogen extract by 1.35 and 2.58%, crude fiber by 3.46 and 1.82%, crude ash by 1.85 and 1.75 times. This effect was also accompanied by the improvements in the jejunal morphology. Training for 24 hours and KCl improved meat yields by 0.12-1.22%. The conclusion was made that the combination of 24 hours of thermal training (additional 4^{o}C) at 4 days of age and supplementation of drinking water with KCl during the last week of age mitigates the negative impact of the thermal stress in finisher broilers.

Keywords: Broiler chickens, Electrolyte, Hyperthermia, Mortality, Productivity

Animal health in different slow growing premium broiler genotypes

Abstract ID: 391

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Conventionally raised meat-producing chicken in Germany are usually fast growing strains, whose breeding characteristics are in particular a fast growth, an efficient feed conversion and optimal slaughtering results, but also robustness and resistance.
As a result of rapid growth, a higher burden of the organism on health disorders such as cardiovascular problems or diseases of the skeletal system can occur. Rapid growth combined with high animal weight is the most common cause of gait problems of fast-growing broiler strains. For this reason, four slower growing broiler strains for production under an animal welfare label have been investigated for possible health problems.
Therefore, 750 unsexed broiler chicks of each of the commercial slow growing breeds (Rowan Ranger, Hubbard JA 957, Hubbard JA 987 and Cobb Sasso 175) were housed and at the end of the fattening period (42 days) 50 broilers each were examined with the focus on animal-based welfare parameters. Furthermore, litter quality, performance of fattening and slaughter yield of the broilers were recorded. The best gait was performed by the Hubbard JA 957 broilers (52% showed a normal gait, 42% a mild inexplicit alteration). Even though, the broilers of the breeds Rowan Ranger and Cobb Sasso 175 were assessed with slightly inferior scores in the gait analysis, 90% of all assessed birds coincided with the list of criteria for a livestock-appropriate housing and treatment of broilers in the scope of the animal welfare label issued by the German Animal Welfare Federation. Only Hubbard JA 987 broilers could not comply with the limit, because 26% of the examined broilers showed considerable deviations in the gait score analysis. There was a significant correlation between the gait score of the broiler and the weight, the soiling of plumage and the prevalence of hock burns.
Only isolated alterations were seen in the foot pads. Lesions were observed in 12% of the broilers of the breed Cobb Sasso 175, where the most humid litter was assessed. Considering the alterations of hock burns, similar observations were made, however no significant differences were detected between the breeds. Thus, all breeds satisfy the list of criteria regarding the foot pad dermatitis and hock burn. In the breeds with lower weights (Rowan Ranger and Hubbard JA 957) fewer broilers with skin scratches were found than in the heavy breeds Cobb Sasso 175 and Hubbard JA 987.

Keywords: Animal welfare, Broiler, Gait score, Slow growth

An investigation into the bone mineralisation of meat chickens from hatch until three days of age

Abstract ID: 223

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We have previously reported that later hatching chicks have higher femoral bone ash (BA) at take-off (TO) from the incubator than their earlier hatching counterparts. This study was designed to further explore BA levels in young chicks. In particular, the effect that chick hatch time and, the time that chicks spend in the incubator before TO have on BA, and changes in BA during the first few days post-hatch (PH) as the chick transitions onto diet, were evaluated. Cobb 500 eggs were incubated at 37.8°C, with transfer to hatching trays at 18 days of incubation (DOI). From 19.5 until 21.5 DOI the hatching trays were observed every 6 hours (i.e. 19.5, 19.75, 20, 20.25, 20.5, 20.75, 21, 21.25 and 21.5 DOI), and hatched chicks were recorded. At each of these observation points a subset of chicks from throughout the hatching window were sampled and their weight, length and femoral BA were measured. At 21.5 DOI all remaining chicks were TO and placed in pens with feed and water. Chicks from all hatch times were then sampled 1, 2 and 3 days PH. The hatch window of this batch of chicks had a normal distribution, with the greatest number of chicks hatching between 20.25 and 20.5 DOI. From hatch, all chicks increased in length. At 2 and 3 days PH mean chick weight was significantly higher than mean chick weight prior to TO. Throughout the hatch window BA was highest for all chicks 6-18 hours after their respective hatch time. In contrast BA was significantly lower in chicks that had only just hatched, or, that had hatched at least 24 or more hours earlier. At TO chicks that had hatched later (>20.5 days) had significantly higher BA when compared to the earlier hatching (<=20.5 days) chicks. Interestingly at day 1 PH the later hatching chicks experienced a decline in BA compared to their BA at TO, but, their BA increased on day 2 and most notably on day 3 PH. For the earlier hatching chicks BA was lowest at TO and then increased continually across the first 3 days PH. At day 3 PH there was no significant difference in BA between early and late hatching chicks. This study has demonstrated a benefit for BA when chicks remain in the incubator for up to 18 hours after hatch before TO.

Keywords: Bone ash, Hatch time, Incubation, Meat chicken

Behavioral suitability of cocks with three different genetic backgrounds – dual-purpose hybrid, layer and heritage breed – for meat production

Abstract ID: 428

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The actual practice of killing 48m day-old male chicks of layer lines in Germany each year generated an ethical debate over the last decade, which forces agriculture and government to find appropriate alternatives to this practice. One idea is the reawakening of the fattening of these male chicks in terms of meat production. For this purpose, also dual-purpose hybrid lines and heritage breeds are in a promising position. The current study investigates on the behavioral suitability for meat production of cocks of three different breeds in terms of animal welfare. The dual-purpose hybrid Lohmann Dual (n = 844), the layer Lohmann Brown (n = 714) and the traditional breed Rhinelander (n = 458) were raised under conventional conditions and slaughtered at an age of 10 and 20 weeks, respectively. Until week 10, hens and cocks were raised together. Several parameters concerning animal health and welfare have been monitored weekly for each breed according to the Welfare Quality® Assessment Protocol for Poultry, including the avoidance distance test (ADT), the novel object test (NOT) and parameters related to the behavioral complex of dominance behavior, pecking at conspecifics and feather pecking. Within all measurements, Rhinelander showed more fearful behavior than the other two breeds. Rhinelander avoided human contact actively ($p \leq 0.000$) during ADT and showed also higher fear levels towards novel objects ($p \leq 0.000$) than Lohmann Brown and Lohmann Dual. Lohmann Brown and Lohmann Dual showed no significant behavioral differences except for their fearfulness towards novel objects, with Lohmann Brown showing to be the least fearful breed ($p \leq 0.03$). Also, breeds did not differ significantly in their social interaction complex. The results indicate that in terms of animal welfare the fattening of laying lines is possible. In the past, commercial breeds have been genetically selected for less aggressive behavior and more adaptive behavior towards humans and their environment, in contrast to traditional breeds like Rhinelander. Thus, Rhinelander showed to be the least suitable breed for using the male chicks for meat production as they are more susceptible to stress during the fattening period.

Keywords: Animal welfare, Dual purpose, Heritage breed, Layer

Blood levels of cortisol and differential leukocytes as measure of stress among egg-type chickens on alternative housing systems

Abstract ID: 611

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Chickens under different housing systems show diverse responses in terms of performance, sentience, emotions and pain. Egg-type chickens being housed in battery cages are under bad welfare condition hence the recent global shift on to the use of alternative systems for chickens to eliminate stress. With this shift, methods of measuring chicken's response to good and bad welfare still remain inconclusive with various techniques being reported. Changes in levels of many blood metabolites and enzymes have been reported as good indicators of stress among chickens but their reliabilities and reproducibility still remain controversial. Therefore, an investigation was carried out to establish changes in blood levels of cortisol, total white blood cell counts, differential leukocytes and heterophil-lymphocyte ratio as reliable ways of measuring stress in egg-type chickens on different housing systems in humid tropics. 225; 8-weeks old each of Nera Black pullets (NBpx) and Super Brown pullets (SBpx) were randomly allotted into three housing systems; Conventional Battery Cage (CBc), Deep Litter System (DLs) and Deep Litter System with outside run (DLr) in a Randomized Complete Block Design (RCB). Each housing system constitutes a treatment group with seventy-five chickens in three replicates for each strain. Routine management practices were strictly observed. Blood collection via wing web veins started at age of 12 weeks and bi-weekly thereafter while the experiment lasted for 24 weeks. Blood sample were analyzed for concentrations of serum cortisol,total white blood cell counts (TWBc), differential leukocytes such as lymphocytes, heterophils. Data generated were subjected to analysis of variance statistically while heterophil-lymphocyte ratios were calculated for each of the experimental groups. The TWBc of the hens were not significantly ($p>0.05$) affected by the housing system and strain. The values for this ranged from 5.05×10^3/µl for NBpx on CBc to 5.10×10^3/µl for LBpx on DLr. However, the heterophils, lymphocytes and cortisol levels were significantly ($p<0.05$) affected by the housing systems only. Hens on CBc had highest levels of heterophils and cortisol but lowest levels of lymphocytes than others. Hens on Cbc also had the highest values for H/L ratio with the lowest from the hens on DLr. The results of this experiment suggest that these blood indices except total white blood cell counts can be used confirmatorily to measure the stressful condition of egg-type chickens on different housing systems alternative housing systems with good reliability.

Keywords: Blood, Housing, Layers, Measurement, Stress

Differential expression of magnum proteins in laying hens treated with corticosterone

Abstract ID: 619

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Stressful environment, including experimental setting, can affect both egg production and quality that are assumed to be caused via gene and protein expression in the ovary and oviduct in laying hens. The objective of this study was to investigate the effects of dietary corticosterone as a stress model on the expression of magnum proteins in laying hens using proteomics analysis. Forty, 47 week-old Single Comb Brown Hy-Line Leghorn laying hens were housed in individual cages in a room with light regimen of 15 h lights (on at 06:00 h) and temperature of 20 ± 2 °C during the entire experimental period. Feed and water were provided ad libitum. After adaptation to the experimental environment for the first 14 days, hens were divided into two groups and provided for the next 14 days with either control- or corticosterone-containing diet at 30 mg/kg. For each hen, feed intake, egg production and egg weight were monitored daily in the morning and body weight measured weekly throughout the experimental period. Protein, extracted from magnum tissues obtained on day 14, was subjected to two-dimensional electrophoresis (2DE). Corticosterone treatment gave rise to 318 protein spots that were up- or down-expressed at least 1.2 fold compared with control. Three 2DE images of each treatment were processed to determine the intensity of protein spots by an image master and were statistically analyzed by a t-test. From 90 protein spots, 45 proteins were identified by MALDI-TOF/TOF-MS/MS analysis and bioinformatics. Corticosterone treatment resulted in significant up- or down-regulation of cytoskeleton-associated proteins, proteases, protease inhibitors, transport proteins, DNA-associated proteins, enzymes, stress-associated proteins, protein folding-associated proteins, and immune-related proteins. The results of the current study suggest that stress can exert detrimental impact on protein expression in hens' magnum.

Keywords: Corticosterone, Laying hens, Magnum proteins, Proteomics, Stress

Effect of broiler hatching system and diet composition on indicators of welfare and performance

Abstract ID: 127

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On-farm hatching systems for broilers are increasingly being used. Eggs are transported to the farm at d18 of incubation and chickens hatch in the house, where they have immediately access to feed and water. We showed that on-farm hatching of broiler flocks resulted in better performance, better litter quality and less footpad dermatitis compared to flocks that hatched in the hatchery. Because of the high performance of on-farm hatched flocks, we hypothesised that feeding on-farm hatched flocks a low-density diet would result in similar performance, but lower feed costs compared to hatchery hatched broiler flocks fed a standard diet. On-farm hatched (OH) and hatchery-hatched (HH) flocks were fed either a standard (S) diet or a low-energy/low protein diet (LE) (2% reduction in energy and lysine) in a 2x2 experimental setup. The experiment comprised three production cycles, with two replicates per hatching system/diet per cycle, resulting in N=6 replicates per treatment combination. Chickens (Ross 308 as hatched, same parent stock per cycle) were housed in pens (1150 chickens/pen) in mechanically ventilated rooms. For on-farm hatching the X-treck system (Vencomatic, The Netherlands) was used. Measurements included performance, welfare indicators and carcass yield. Data were analysed using the GLM procedure. In agreement with earlier studies, OH flocks had better footpad dermatitis scores (FPS) as compared to HH flocks (0.72 vs. 1.35; $P<0.001$), which was probably due to the significant better visual litter score (5.5 vs. 4.6; $P=0.02$). Diet had no effect on FPS and litter quality. LE diet resulted in a better gait score as compared to the S diet ($P=0.03$). No differences were found in body weight at d39 and mortality. Total feed conversion ratio was better for HH as compared to OH flocks ($P=0.03$), in contrast to results of previous experiments. Carcass% was higher for HH than OH flocks ($P=0.05$), but no diet effect was found, and no other differences in carcass composition were found. In conclusion, our hypothesis that providing a low energy/protein diet to on-farm hatched flocks would result in equal performance as compared to control flocks fed a standard diet, could not be confirmed. The present experiment confirmed the reduced risk for footpad dermatitis and better litter quality in on-farm hatched as compared to hatchery hatched flocks.

Keywords: Broiler, Diet, Hatching, Performance, Welfare

Effect of intermittent lighting schedule on behaviour, food conversion ratio and performance in welfare tests in early life of chicks

Abstract ID: 386

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The mother hen shows chicks what to peck and when to rest by protecting them in the dark and warmth underneath her. This way the hen controls chick behaviour, helps with synchronisation of their activity and buffers response of chicks to stress. Fearful chicks with unsynchronised behaviour are more likely to develop behavioural problems (e.g. feather pecking) or hurt themselves (e.g. panic response). Keeping mother hens with their chicks is not commercially viable and chicks are hatched and reared in large groups using artificial incubation, lighting schedule and brooders. In our study eighty 1-day-old Hy-Line Brown chicks were randomly assigned into two groups of forty, each housed five chicks per pen. Forty chicks were kept under the continuous lighting schedule (C) with 14 hours of light and 10 hours of darkness per day (14L : 10D), while the other 40 chicks were kept under the intermittent lighting schedule (IT), based on pre-established natural brooding patterns of broody hens and chicks. Body weight was measured three times a week and feed consumption was measured five times a week during first 16 days of life. Behaviour (locomotion, comfort behaviours, feeding behaviours), activity and synchronisation of chicks within pen were also recorded on day 1, 2, 7 and 14. On day 16 chicks were subjected to tests measuring their welfare status, including human approach, novel object and tonic immobility tests and their performance was evaluated. Data were tested for normality and the groups were compared using independent samples t-test. Behaviour of IT chicks was more synchronised on day 1 and 2 ($P < 0.05$ and $P < 0.001$, respectively), and they were more active on both days ($P < 0.05$ and $P < 0.001$, respectively) with no differences on day 7 and 14. The feed conversion ratio of IT chicks was lower in comparison with C chicks (day 8 $P < 0.001$; day 15 $P < 0.001$). Chicks reared under IT lighting schedule weighed more on day 8 ($P < 0.01$), but not on day 15. Tests measuring welfare status did not prove any differences between chicks reared under different lighting schedules. To conclude, rearing chicks under IT lighting schedule enhanced their early behavioural synchronisation and feed conversion ratio with no detrimental effects on welfare. This synchronisation could potentially have positive effect on behavioural problems later in life.

Keywords: Behaviour, Chicks, Feed conversion ratio, Lighting schedule, Welfare

Effects of particle size and dietary levels of perlite on performance and tibia bone characteristics of broiler chickens

Abstract ID: 568

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This study was to investigate the effects of different levels perlite on performance and tibia bone characteristics of broiler chickens. 336 one day old broiler chicks reared for 42 days in a completely randomized design with 7 treatments, 4 replicates and 12 chicks per each. The experimental treatments were control (without perlite supplementation), perlite levels (2 and 4%), and particle size (fine, medium, coarse). The results showed that inclusion of 4% fine perlite increased feed intake in comparison to other groups significantly in 3-6 and 1-6 weeks of age ($P<0.05$). As body weight gain, supplementation of 4% coarse perlite led to lowest in comparison to other treatments at finisher and total rearing phase ($P<0.05$). But, control group and 2 % medium and coarse perlite supplemented broilers had lowest feed conversion ratio ($P<0.05$). Tibia bone ash content increased with no perlite addition and in groups supplemented with 4% fine and 2% coarse perlite compared to 4% coarse group significantly ($P<0.05$).

Keywords: Broiler, Particle size, Performance, Perlite, Tibia

Elimination of poultry red mites (dermanyssus gallinae) by using essential oils and their active ingredients

Abstract ID: 601

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The objective of the performed experiments was to monitor mortality of poultry red mites (*PRM, Dermanyssus gallinae*), caused by plant essential oils (clove bud, lavender and cinnamon) and their active ingredients. The active ingredient in clove bud is eugenol ($C_{10}H_{12}O_2$), in lavender is linalool ($C_{10}H_{18}O$), in cinnamon is cinnamaldehyde (C_9H_8O). The acaricidal activity of these oils and their active ingredients against poultry house-collected red mites was examined using direct contact method – by glass vial bioassay. Oils and their active ingredients were tested at different concentrations (µL/cm^2). Oils were dissolved in water and Tween 85 and the solutions were spread on the filter paper at the bottom of the glass vial. The solvent of solution was then evaporated. Twenty movable poultry red mites were placed on the filter paper impregnated by oil or its active ingredient at the bottom of the glass vials. The mortality of the poultry red mites in the glass vials was measured after 24h.

All used oils and their active components caused mortality of poultry red mites. The highest mortalities were observed with clove bud and cinnamon, which caused more than 99% mortality of poultry red mites at concentrations 0.5; 0.25; 0.12 and 0.06 µL/cm^2. Cinnamon caused higher than 90% mortality of PRM and clove bud caused 81.8% mortality both at concentration 0.03 µL/cm^2. There was no significant difference in efficiency on PRM mortality between these two oils.

Lavender caused more than 99% mortality at concentration 0.5 µL/cm^2.

At concentration 0.5 µL/cm^2 there was no significant difference between the efficiency of lavender, cinnamon and clove bud. Lavender caused less than 50% mortality at other concentrations. At concentration 0.015 µL/cm^2 there was significant difference between the efficiency of clove bud and lavender, however both caused less than 10% mortality. The results of these performed experiments suggest that clove bud, lavender and cinnamon essential oils in merit further study as potential poultry red mites control agents.

Keywords: Botanical pesticides, Cinnamaldehyd, Eugenol, Linalool

Genotype Effect on Gentle, Severe and Aggressive Pecking Behaviours and Feather Condition in Laying Hens

Abstract ID: 136

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Feather pecking in laying hens is a serious behavioural problem and has negative impact on bird's welfare and productivity. Genetics, nutritional and managemental factors beside developmental stages of birds affecting feather pecking have long been studied. However, there is no clear solution available by now. Therefore, this study aimed to investigate differences in feather pecking behaviour and feather condition of local layer hybrids that developed and commercialyy available in Ankara Poultry Research Institute. A total of 576 hens from three genotypes namely Atabey (white egg), Atak, and Atak-S (brown egg), 192 each, were used. Birds were kept at enriched cages in two adjacent environmentally controlled rooms by 64 wk. Each room contained 18 cages (240×63.5×68 cm) with 16 hens each. Genotypes were represented with 6 cages per room (12 cages/genotype) as experimental unit. Observations for feather pecking behaviour were made using video records obtained from 4 cages/genotype in 2 consequent days on 24 and 30 weeks of ages during photoperiod (14 h). However, time sampling (1 minute in every hour) was used for data collection; and gentle, severe and aggressive pecks given to cage mates were counted. Each bird was scored for feather condition at the age of 64 wk. Each body part (head-upper neck, back, ramp, tail, cloaca-abdomen, breast, and wings) was scored using a scale 0 (best) to 5 (worst). Behaviour data were analysed by generalized estimating equations (GEE) using logistic regression procedure (PROC GENMOD) of SAS statistical software with a model including age, genotype, age by genotype interaction, and room effect. Cage effect was nested within genotype and room effects. ANOVA was performed for feather condition and the model included genotype, room, and interaction effects; and cage within room and genotype. Statistical significance was based on $P \leq 0.05$. Severe and aggressive pecking counts significantly differed with genotype ($P \leq 0.05$). Odd ratios revealed that both brown egg layers, Atak and Atak-S, showed a higher tendency to perform severe pecks as compared with the white egg layer Atabey (OR=3.17 and 3.44 for Atak and Atak-S genotypes using Atabey as reference). Higher aggressive peck counts were observed in Atak-S than Atak (OR=1.75) and Atabey (OR=2.24). Gentle feather peck counts did not differ with genotype. There were significant differences among genotypes for feather condition ($P \leq 0.05$) and Atabey had the best score (17.03) while Atak was worst (28.15) and Atak-S (26.27) was intermediate.
This study has been funded by TAGEM/HAYSUD/A/16/A06/P03/01.

Keywords: Feather condition, Feather pecking, Genotype, Laying hens

Improving Poultry Welfare in China

Abstract ID: 339

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China is the largest producer of pig meat, chicken and eggs in the world. As production continues to increase, interest in and understanding of the concept of animal welfare is gradually growing also. Compassion in World Farming, an established, fast-growing and influential global NGO focused on the promotion of farm animal welfare, and the Chinese government-backed organisation – the International Cooperation Committee of Animal Welfare (ICCAW) – have worked in partnership since 2014 to bring higher welfare practices into farm animal production. Through this partnership, Compassion aims to improve welfare standards and raise public awareness on farm animal welfare issues across China; through awards, media activities and education. Compassion launched the Good Pig Production Award in China in 2014 to bring higher welfare practices into local pig production, recognising Chinese producers for improving pig welfare, food safety and environmental standards. Now in its third year the awards continues to encourage welfare improvements from pig producers and in 2017 the programme extended further with the launch of the Good Egg Production Awards and Good Chicken Production Awards. These awards recognise producers that are committed to cage free production and improving the welfare of broilers and laying hens by incorporating key elements into poultry systems such as increased space allowance, provision of enrichment, use of slow growing, dual purpose breeds, and the ending of routine mutilations such as beak trimming. The awards consist of a one star (basic criteria) to five star (all criteria) rating. To ensure maximum impact with this awards programme applicants must be medium to large scale producers who meet basic requirements agreed and set out by Compassion and ICCAW. Applications are reviewed by an expert judging panel and eligible producers undergo an on-site inspection undertaken by experts from or on behalf of Compassion to evaluate the current welfare conditions and likelihood of meeting any additional commitments to improve. The awards have already attracted interest from a number of very large producers including OSI (Weihai) Poultry Development CO., LTD., a wholly-owned subsidiary of the U.S. OSI Group and Hubei CP Foods, a wholly owned subsidiary of Thailand's Charoen Pokphand Group. To date, through this programme in China over 90 million birds are set to benefit from the Good Production Award winners' policies and practices.

Keywords: Awards programme, Broiler welfare, China

Influence of dietary vitamin A levels on tibial dyschondroplasia in broiler chickens

Abstract ID: 574

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Tibial dyschondroplasia (TD) is a disease that affects birds with accelerated growth. The condition is characterized by the presence of an unmineralized cartilage mass that extends distally from the growth plate of the metaphysic at the proximal end of the tibia and occasionally to the distal end of the tibia, proximal end of the tarsometatarsus and proximal ends of femur and humerus which results in bone deformity and lameness. It is a consistent observation that broilers and turkeys are most suseptible to the development of TD under environmental conditions that maximize the growth rate and treatments that eliminate the occurrence of TD are those restrict growth. The symptoms can range from subclinical to severe bone deformities and lameness. The severity of the disease is usually scored on the basis of the size and location of the lesions. A range of nutritional, environmental and genetic factors may be cause TD. One of investigated nutritional factors is vitamins and their interaction. Although, vitamin A is considered to be essential for normal skeletal tissue development, this fat-soluble vitamin has been reported to cause a decrease in bone ash and an incidence in leg problems when fed at high levels. Apparently, pharmacological levels of vitamin A interfere with metabolism and utilization of the other fat-soluble vitamins. It is possible that interactions with vitamin D cause a decrease in calcium absorption and influence the development of TD in broilers. Also, vitamin A competitively inhibits utilization of the other fat-soluble vitamins such as vitamin D3.

Keywords: Broiler, Tibial dyschondroplasia, Vitamin A

Keel bone fractures in laying hens affect laying performance but not egg quality

Abstract ID: 103

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Although up to 90% of laying hens manifest keel bone fractures, it is not known to what extent individual hens are affected. The aim of this study was to investigate the effect of keel bone fracture severity on individual productivity of laying hens. Focal hens (75 LSL, 75 LB) were housed in 10 identical pens containing a commercial aviary system (15 LSL focal hens + 210 LB or 15 LB focal hens + 210 LSL per pen). Eggs of focal hens were identified by orally administering a dye on three consecutive days, resulting in a hen-specific colour pattern in the yolk. Eggs were collected at seven time points (37-61 weeks of age; WoA) for five days to determine individual laying performance (N=4813). Egg quality was assessed in all eggs laid on the first three days of egg collection (N=2868). Radiographs were performed on the last day of data collection for each time point with resulting images used to score fracture severity on a continuous scale. Additionally, the state of the keel bone was scored as inactive (absent, healed or inactive fractures), healing or fresh. Linear mixed effects models were used for statistical analyses. At WoA 37, hens with the highest observed fracture severity laid more eggs (100% laying performance) than hens without fractures (95%) whereas at WoA 61, performance in hens with the same fracture severity was 12% lower than in hens without fractures (86% vs. 98%; $P=0.005$). Hens with fresh fractures had a lower performance than hens with healing and inactive fractures at WoA 37 (94%, 96% and 98%, respectively) but higher performance at WoA 61 (98%, 96% and 92%, respectively; $P=0.003$). Egg quality parameters were not affected by fractures but were associated with an age*hybrid interaction (egg mass: $P=0.0495$, shell breaking strength: $P=0.016$, shell width: $P=0.001$). The absence of a relationship between fractures and egg quality suggests that hens prioritized maintaining egg quality over egg numbers while coping with a physiological challenge. As young hens with high performance were associated with high severity fractures and older hens with high performance suffered from fresh fractures more often, high productivity might be linked with fracture susceptibility. The dramatic decrease in performance at the end of lay in hens with severe fractures not only raises substantial economic concerns for producers, but also implies reduced fitness and consequent welfare problems in laying hens suffering from fractures.

Keywords: Individual productivity, Keel bone fractures, Laying hen welfare

Killing individual poultry on farm: a matter of animal welfare and feasibility

Abstract ID: 134

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European Council Regulation 1099/2009 stipulates that on-farm killing of sick and injured poultry is done humanely and that the level of suffering is minimized. The regulation allows cervical dislocation for small poultry (<3kg), while larger poultry (>3kg) can be killed by a percussive stroke on the head. Given the huge number of birds involved worldwide, it is important to ascertain whether both methods are truly the most humane for killing individual poultry. The aims of this research was to investigate several killing methods and to evaluate the feasibility of those methods using them on-farm. Three killing methods were evaluated in chickens at age of 2, 4 and 6 weeks, namely manual cervical dislocation (CD), captive bold device (CB) and electrocution (EL). In 8-, 12- and 16-week-old turkeys, five methods were tested, namely mechanical CD (broom stick and neckcrusher), CB, EL and nitrogen gas (N2). The evaluation was based on efficacy (killing was successful or not), on time measurements when reflexes stopped (pain response, pupil reflex, nictitating membrane, wing flapping, leg paddling, rhythmic breathing and heartbeat) and on measuring electroencephalography (EEG) to record electrical activity of the brain. For both chickens and turkeys, head electrocution using the electric stunner of FAF did not appear to be a good killing method. For younger poultry, the down feathers provides poor electrical conduction. Larger animals are too heavy resulting in cutting injuries around the neck region caused by the fixation system. Therefore, this method was canceled after the first age categories. In chickens, CD and CB were both 100% efficient killing methods, EL 91.7%. The time until cessation of the pupil reflex and nictitating membrane reflex was significant longer in CD compared to CB for all three age categories ($P<0.001$). Wing flapping and leg paddling did not occur after EL, resulting in a significantly shorter duration compared to CD and CB ($P<0.001$). The time lag until cessation of rhythmic breathing was slightly longer after CD compared to CB and EL. The time until complete cardiac arrest was similar for the three methods. The EEG results and the results for turkeys are currently being analyzed and will be presented at the conference as well. Based on the reflex measurements, it can be concluded that CD as well as CB are relatively humane methods for killing broiler chickens at various ages. Which method to recommend for commercial on-farm use, however, also depends on the practical feasibility.

Keywords: EEG, Killing, On-farm, Poultry, Welfare

Non-beak trimmed hens versus beak trimmed hens kept in aviary

Abstract ID: 199

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Feather pecking is an important welfare and economic problem in laying hens. Because of national legislation or market demands, more and more hens in Europe are non-beak trimmed. It has been shown that the consequences of feather pecking in non-beak trimmed hens (NBT) can be much larger compared with beak trimmed hens (BT). This can mean a serious decrease of animal welfare and health and can strongly influence flock performance. Field studies show different outcomes of NBT flocks and conclusions are often contradictory. The Experimental Poultry Centre therefore compared 3840 BT hens with 3840 NBT hens in a controlled set-up. All hens (Isa Brown) were from the same breeder flock, were born on the same day, were reared in identical conditions in the same rearing house, were transported in the same way to the layer house and were placed in 4 identical aviaries (2 with NBT hens, 2 with BT hens). All groups received pecking blocks and roughage as distraction. Production data were recorded, a flock behavior checklist was performed and feather cover was assessed every 4 weeks. The aim of this study was to evaluate the onset and evolution of feather pecking in the groups.The hens were moved to the laying house in spring and encountered high ambient temperatures, discouraging a sufficient feed intake during the first weeks. Together with the increasing nutrient needs due to the final maturation and development of the reproductive system, this caused an imbalance and stress. Already at 22 weeks of age the first signs of feather pecking were detected in the NBT hens while all BT hens were still intact. Feather damage increased rapidly in the NBT hens which resulted at 39 weeks of age in bald hens with obvious signs of cannibalism. Measures were taken to stabilize the situation: extra distraction was offered to keep the hens busy, the light was dimmed gradually and eventually shifted towards red light. Especially the latter stabilized the situation and decreased the number of wounds on the back and vent area. In conclusion, even when hens have identical origin, youth, housing and management, differences can be seen between non-beak trimmed and beak trimmed hens in the onset and evolution of feather pecking.

Keywords: Beaktrimming, Cannibalism, Feather pecking

Pecking behavior of pullets in a commercial aviary

Abstract ID: 270

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In aviary housing systems pullets are usually confined within aviary cages during the first 4 weeks of life. From the 5th week of life onwards the aviary is opened and the animals have access to a litter area. The influence of the stocking density during the rearing period on the occurrence of feather pecking is discussed controversy in literature. On the other hand there is a broad agreement, that litter and enrichment reduce feather pecking. The aim of our project was to study the ontogeny of pecking behavior of pullets to optimize the housing conditions accordingly. We observed the animal behavior in 3 different groups: Group 1: 22 pullets/m^2 from day 35 onwards, without enrichment, group 2: 18 pullets/m^2 from day 35 onwards with enrichment, group 3: 22 pullets/m^2 from day 35 onwards, with enrichment. Management was equal for all animals. Our results already showed significant differences between the groups, while the pullets were kept in the aviary cages: In group 1 significantly more gentleand severe feather pecking was observed in comparison to groups 2 and 3. After opening the aviaries with access to the litter area from week 5 onwards these differences could still be observed: both gentle and severe feather pecking occured significantly more often in group 1 then in groups 2 and 3. These findings allow the conclusion that the enrichment in groups 2 and 3 led to a reduced feather pecking rate. The results found in literature could therefore be verified in a large commercial farm. At the same time we found that group 3 showed significantly more severe feather pecking then group 2 after opening the aviary to give access to the litter area from day 35 onwards. Therefore a reduced stocking density seemed to have a positive (reducing) effect on the feather pecking rate while enrichment already is presented to all animals. In this project the sole effect of a reduced stocking density (without any enrichment) could not be investigated, but would be an interesting subject for further studies.

Keywords: Enrichment, Feather pecking, Pullet behavior

Plumage condition, foot and keel bone disorders in laying hens: effects of different production systems

Abstract ID: 397

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Plumage condition, foot and keel bone disorders (KBD) were measured on laying hens reared in enriched cages and organic production. The research was carried out on beak-trimmed Lohmann brown hens at the end of the production cycle. At 72 weeks of age, 100 hens were randomly selected from each housing system and were inspected for plumage condition, and foot and keel bone disorders. The plumage condition ranged from 1 (severe feather damage) to 4 (perfect feather coverage), and bumble foot were scored according to Tauson et al. (2005). The footpad lesions were recorded according to Ekstrand et al. (1998). The length of the claw was recorded, whether claws are too long or normal. The prevalence of KBD was assessed by using the palpation technique of Wilkins et al. (2004).
Feathering of neck, breast, tail, wings, back and cloaca/vent was significantly worse in hens from enriched cages compared to organic hens (3.1 vs 4.0). Rearing system had a significant effect ($P < 0.001$) on claw length. Even 94% of hens from the cage had too long claws at the end of the production cycle, while in organic production only 8%. There was no significant effect of production system on bumble foot.
Foot pad lesions were more frequent in layers from organic production (10%), but deep and severe lesions were only observed in a few cases. Caged hens had significantly less foot pad lesions (1%) compared to organic hens. The overall range of KBD in cages was significantly higher than in the organic system (43% vs 4%).
Based on the results of this study, it could be stated that the plumage condition, foot and keel bone disorders (KBD) were seriously influenced by the rearing systems at the end of production cycle. Generally it can be concluded that rearing laying hens in organic production resulted in better welfare parameters, which is certainly the result of more favorable production conditions.

Keywords: Enriched cage, Laying hens, Organic production, Welfare

The effect of feather pecking phenotypes on affective states and decision making in laying hens

Abstract ID: 293

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Feather pecking in commercial laying hens is a maladaptive behaviour that reduces welfare of laying hens. A number of experiments have shown behavioural and physiological differences between the line selected for high level of feather pecking (HFP) and low level of feather pecking (LFP). In this experiment we investigated whether these two lines differ also in their affective states and cognitive abilities by using a cognitive bias test. Twenty HFP and twenty LFP laying hens were trained to discriminate feeders of two different colours. Half of the animals were trained to approach the white feeder to obtain the reward (mealworm) and not to approach the black feeder to avoid the punishment (water spraying). The other half of animals was trained in a reversed way. After reaching a discrimination criterion the hens were exposed to the feeder with an ambiguous colour (grey). The mean latency to reach the feeder was measured. Thirty six out of forty hens reached the learning criterion and learnt to discriminate the positive and the negative feeder colours. The learning process was slower in HFP line. However, the HFP line approached the ambiguous feeder significantly faster than hens from the LFP line (HFP 13.59 ± 0.59 s, LFP 16.68 ± 0.79 s, $P<0.05$). Possible interpretation based on cognitive bias hypothesis is that their decision making under ambiguity was more optimistic. Nevertheless, another possible explanation is the higher activity and propensity to search stimuli for pecking in HFP line.

Keywords: Affective states, Cognitive bias, Feather pecking, Laying hens

The influence of egg production on keel and long bone quality in laying hens

Abstract ID: 330

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Bone fractures are one of the most serious animal welfare problems in the egg production industry. Particularly, the prevalence of keel bone fractures is very high, reaching up to 97 % of hens near the end of a production cycle with affected hens likely to suffer pain. Since a lot of calcium is required for egg shell formation, high egg production and the associated calcium mobilization from the skeleton might be one factor contributing to this multifactorial problem. The aim of this study was to investigate how egg production affects keel bone health and characteristics of long bones. A total of 76 laying hens of the layer line WLA with an average laying performance of 320 eggs per year were kept in a group floor housing system from hatch until 62 weeks of age. 38 hens were given a Suprelorin® implant with the active component deslorelin acetate that prevented egg production for three months at a time. Subcutaneous implants were renewed every three months to cover the whole production cycle (group S). The other 38 hens were kept as control animals (group C). To detect keel bone fractures, all hens were radiographed repeatedly. In the 62nd week of age, hens were euthanized and dissected. Strength and chemical composition of tibiotarsi were analyzed using the following methods: three-point bending test, thermogravimetric analysis, infrared spectroscopy, X-ray diffraction and scanning electron microscopy. Hens of group S, which were prevented from egg laying, showed significantly fewer keel bone fractures from 32nd week of age onwards (group S: 1.64 % prevalence; group C: 67.46 % prevalence; $P<0.001$). Tibiotarsi of group S had a significantly larger diameter ($P<0.05$) and were mechanically stronger ($P<0.001$) compared to group C. The degree of mineralization of cortical bone was significantly lower in group S hens compared to group C hens ($P<0.05$). Medullary bone was present in tibiotarsi of group C but not of group S hens. These results give strong evidence that egg production plays an important role in the etiology of keel bone fractures. This is consistent with the differences in long bone characteristics which have been found: bone diameter and bone strength seem to be negatively influenced by egg production. The negative contribution to bone breaking strength of a smaller bone diameter cannot be compensated by the higher degree of mineralization of cortical bone and the presence of medullary bone in productive laying hens.

Keywords: Bone quality, Egg production, Keel bone, Laying hen, Radiography

The influence of litter material on broiler production performance and feet health

Abstract ID: 537

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A completely randomized study with four treatments replicated four times was designed with the aim to find alternative litter material that could replace wood shaving and be able to minimize the incidence of feet pad dermatitis (FDP) and hock burns in broilers. The four litter treatments were wood shavings (control), dry pine tree leave (DPL), decomposed kraal manure (DKM) and sand. Bedding material was laid at the depth of 10 cm, and wet patches were removed when necessary. A total of 320 day-old Ross 308 chicks were used with 20 birds per replicate. Broiler feet health data was collected daily in the form of observation for occurrences of foot pad dermatitis (FDP), hock burns and mortality. Broiler body temperature was measured on weekly basis using clinical thermometers. FDP was assessed by an eye and scored according to the protocol of Berg (1998). Feed intake, body weight, growth rate and feed conversion ratio were measured on weekly basis. There were significant ($p<0.05$) differences in FPD incidences between litter materials where the highest (64%) were observed in sand, followed by DPL with 16%. The control had no incidences while fewer incidences (9.85%) were observed in DKM. Contrarily, there were no significant ($p>0.05$) differences in the hock burns, mortality and body temperature between different litters materials. However, hock burns incidences were high in DPL and sand which had equal numbers of mortality at 0.33% while DKM recorded no hock burn and mortality incidences. The lowest and highest body temperature was recorded for DKM and DPL respectively. Broiler production performance did not differ significantly ($p>0.05$) between litter materials. However, birds reared in wood shavings and DKM had better growth rates than the rest of the treatments. The highest feed intake was observed in the sand while the lowest was recorded in the control group. The lowest body weight and growth rate was observed in DPL and sand respectively. The highest feed intake in sand could have been used to keep the animals warm because sand was the coldest of all litters. Based on incidences of FDP, hock burns, mortality, feed intake and growth rate, DKM was found to be an appropriate alternative litter material that can replace wood shavings.

Keywords: Broiler litter material, Decomposed kraal manure, Feet health, Sand, Wood shavings

The informative value of the assessment of injuries of layers at slaughter with respect to cannibalism

Abstract ID: 154

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Detection of cannibalism is essential for the improvement of hens' welfare in subsequent laying periods. The validity of the assessment of skin injuries at slaughter compared to on farm was evaluated in hens, being non-beak trimmed (19 flocks) or beak trimmed (8 flocks). 438 hens each from 27 flocks of 10 farms were examined at the evisceration line after a previous on farm examination by the same observer (30 hens/flock). Time intervals between "on farm" and "at slaughter" assessments varied from 1 to 22 weeks. On average 7.1% of the laying hens had skin injuries, non-beak trimmed hens more (8.1%) than beak trimmed hens (4.5%). With a threshold for the definition of cannibalism set at 10% of injured hens in a flock, 6 flocks were affected by cannibalism. A correlation between the assessment results of skin injuries at slaughter with those on farm was found (0.639; $P<0.001$, Kendall's-Tau). The difference of assessed skin injuries between the two assessment methods was 24.4% (31.4% of hens affected on-farm, 7.1% at slaughter). The coefficient of variation for repeated measurements of 129.3% demonstrated a large variation in the assessed prevalence of injuries. After data transformation into a binary form of either "cannibalism" or "no cannibalism," we conducted a receiver operating characteristics–curve analysis to determine the accuracy of estimating the prevalence of cannibalism on-farm via examinations at slaughter. The results showed that a threshold of 1.4% of hens with injuries at slaughter indicated the presence of cannibalism defined as 10% or more of the hens being injured on-farm with a sensitivity of 100.0% and a specificity of 73.3%. To conclude, the detection of preceding cannibalism at slaughter is possible, and the detection threshold to diagnose cannibalism from injuries is much lower at slaughter than on-farm.

Keywords: Assessment of injuries, Cannibalism, Layer, Slaughter

The role of dopamine in control of feather pecking in laying hens

Abstract ID: 579

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Feather pecking (FP) in laying hens, or more exactly severe FP resulting in feather damage, represents both welfare and economic problem of egg industry. Involvement of both serotoninergic and dopaminergic neurotransmitter systems in control of these adverse behaviours has been implicated. Dopamine is a neurotransmitter involved in a wide range of functions, including reward or various behavioural problems. Using systemic treatment with dopamine receptor agonists and antagonists we assessed the role of dopamine D1 and D2 receptors in control mechanisms underlying FP. Based on preceding behavioural tests 24 LSL hens with the high (HFP) and 24 hens with the low FP frequency (LFP) were selected. Each bird was subjected to 6 consecutive i.p. treatments every other day in a balanced order: 1 mg/kg apomorphine (mixed D_1/D_2 agonist), 4 mg/kg SKF38393 (D_1 agonist), 0.2 mg/kg SCH23390 (D_1 antagonist), 7 mg/kg spiperone (D_2 antagonist), 10 mg/kg bromocriptine (D_2 agonist) and saline (control). One hour after the injection following behaviours were recorded in a test box in a presence of another hen: floor pecking, wall pecking, drinking, gentle and severe FP, aggressive pecking, head-shaking and preening. Three-way ANOVA proved highly significant effects of drug, group (HFP vs. LFP) and time (twelve 5-min periods) ($p<0.001$). Moreover, there were significant effects ($p<0.001$) of drug x group interaction, showing that response to drugs differed between peckers and non-peckers, and drug x time interaction, implying that the time course of response differed between drugs. Apomorphine stimulated mostly floor pecking, head-shaking (in both groups) and preening (in HFP group), but also suppressed gentle FP, severe FP and aggressive pecking in HFP. SCH23390 suppressed and SKF38393 stimulated both gentle and severe FP in HFP hens. Both spiperone and bromocriptine suppressed gentle and severe FP in both groups. Our experiment proved the role of dopaminergic system and mainly D1 receptors in control of FP behaviour of laying hens.

Keywords: Agonist, Antagonist, Dopamine, Feather pecking, Receptors

Preferences of male turkeys for different kinds of manipulable materials

Abstract ID: 442

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Feather pecking and cannibalism display a major problem in turkey housing regarding animal welfare. One approach to reduce the occurrence of feather damages and injuries is the offer of manipulable materials. Generally, these materials are provided manually, however as a perspective in future the development of automatic providing systems is desirable. The aim of this study was to examine, if male turkeys show preferences for several kinds of material, which are also suitable for the usage in automatic systems. Sunflower seeds, maize, oat and wheat were simultaneously provided in two pens with 317 male turkeys (B.U.T.6) respectively. The turkeys of both pens differed in beak condition (beak trimmed vs. intact beak). The materials were offered in special feeders consisting of a tube with two lateral lines of holes. The diameter of the holes was 10 to 14 mm. For determining potential preferences, the number of animals using the feeders was measured by time sampling in the 8th, 12th, 16th and 20th week of age. At all time points, observations were conducted in 10 minutes frequencies over a period of 120 minutes. Data was first tested on normal distribution. Afterwards a Generalized Linear Mixed Model was used for further analysis.
The number of turkeys using the feeder was affected by the kind of material ($P \leq 0.01$), the beak condition ($P \leq 0.001$) and the age ($P \leq 0.001$). The highest numbers of animals were observed at the feeders with sunflower seeds, followed by maize and wheat. Least animals were recorded at the feeders filled with oat. Beak-trimmed turkeys used the feeders less frequently then untrimmed birds. With increasing age, the number of turkeys using the feeders decreased.
Sunflower seeds seem to be more attractive for turkeys than different kinds of grain. Therefore, this material could be appropriate for the use in automatic systems. The special feeders proved to be more suitable for untrimmed birds. Unattached of the kind of material the turkeys showed less interest with age, which is a challenge regarding the prevention of feather pecking and cannibalism.

Keywords: Beak-trimming, Behavioural disorders, Enrichment

Reducing gut lesions caused by bacterial enteritis with a specific blend of esterified fatty acids

Abstract ID: 40

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Modern turkey breeds remain vulnerable to enteric diseases and bacterial challenges such as *E.coli, Salmonella* and *Clostridium*. Therefore it is essential to create a healthy intestine from the very start and to maintain optimal gut functions under high pressure conditions throughout the whole growth period. The efficacy of a specific combination of esterified butyric and lauric acid on gut health and overall performance was tested in turkeys. A total of 1400 healthy one-day-old turkey females were randomly divided into 4 treatment groups with 7 replications each. The experimental groups comprised of a negative control group (NC), fed a basal, commercial diet; a positive control group (PC), fed the same basal diet, but receiving antibiotics via the drinking water throughout the entire experiment and two treatment groups receiving a feed either a high (HD) or low (LD) degressive supplementation of a specific combination of esterified butyric and lauric acid. All birds were individually orally inoculated with a bacterial and Eimeria cocktail during 3 consecutive days from the age of 19 days. They were reared up to 105 days. On day 26, three animals per pen were evaluated for intestinal gut health and coccidiosis. A macroscopic intestinal lesion scoring system, as an assessment of the macroscopic pathology and physical damage caused by bacterial enteritis, was combined with lesion scoring for coccidiosis, as subclinical coccidiosis plays a major role in the development of bacterial enteritis. The LD treatment shows a near-significant trend for a lower coccidiosis scoring compared to the HD, as well as for a lower bacterial enteritis scoring compared to both HD and NC. Higher butyrate concentration reduces the mucin production. A thinner mucosa layer may allow parasites and pathogenic bacteria, like *Clostridium perfringens*, to reach the gut wall easier and inflict more damage. Glycerol monolaurate reduces immune cell proliferation at high concentrations, whereas it increases the proliferation of T-cells at lower concentrations. Moreover, flaccidity in LD and PC is significantly lower than in HD groups and the LD groups show a trend towards lower inflammation compared to HD and PC, which may be due to over-stimulation of the immune system at higher concentration of glycerol monolaurate. Also antibiotics perturb commensal microbial communities and affect susceptibility to infection by intestinal pathogens and development of inflammatory bowel diseases.

Keywords: Bacterial enteritis, Butyric, Coccidiosis, Glyceride, Lauric

Six sigma: process improvement methodology for poultry industry

Abstract ID: 521

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The total poultry population in India is more than 800 million. Its annual growth rate is 5.57 % and while egg and broiler production rate is 11.44 %. Poultry sector provides employment to more than 6.5 million people directly and indirectly in sectors like feed, pharmaceutical, equipment and other support services. Globalization and quick access to information, products and services have changed the way of poultry business. Traditional business models are no longer working effectively.This is why Six Sigma Quality has become an important part of business management system. With the application of six sigma (SS) one can develop and deliver near-perfect products and services of poultry. SS can be defined as a strategy for improvement of quality of output of a process by identification and elimination of causes or defects and minimizing variation in process outputs. "Sigma" is a statistical term which measures the deviation of process from perfection. It also can be defined as anything outside of customer specifications. It is a data driven approach based on measurement of the process variation using statistical process control. It is a tool set, can be used in conjunction with other methods for process improvement. The immediate goal of SS is defect reduction, which leads to yield improvement and higher yields improve customer satisfaction. This is intended to lead to cost reduction. It uses a set of quality management methods for quality improvement of a process. It aims to highlight process improvement opportunities through systematic measurement. It is based on utilizing an extensive set of statistical and advanced mathematical tools, techniques and methodologies that produces significant results quickly. To achieve Six Sigma quality, a process must produce no more than 3.4 defects per million opportunities i.e. chances for nonconformance, or not meeting the required specifications.

Six sigma gives extremely fast rate of improvement in the areas like cost reduction, defect reduction, customer satisfaction, productivity and market growth. It will have great positive impact in poultry feed, post harvest technology, hatchery and day to day poultry management. Poultry industry is yet to be completely exposed to six sigma techniques. So, we should adapt and implement six sigma process rapidly of the process for betterment of poultry industry.

Keywords: Defects, Poultry, Process Improvement, Six Sigma

Support an approach to reduce the antimicrobial use at poultry farm level: integrate human factors comprehension in technical advices toward farmers

Abstract ID: 513

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A part of the variability of antimicrobial uses at farm level is generally explained by structural, sanitary or zootechnical factors. However, a few studies about bovine, pig or rabbit production, underlined that psycho-sociological factors could explain some farmer's choices of livestock management practices and stewardship of animal health measures. Therefore, a survey of 68 broiler farmers was conducted in 2016, in the Brittany and Pays-de-la Loire regions (France). Considering the "job demands-resources model" described by Bakker and Demerouti (2008), the aim of our survey was to study the influence of the difference of farmer's perception about constraints and resources in their job. The question sheet allowed collecting structural and organizational data of the farm and its poultry production, farmer's experience and his poultry livestock management practices. The farmers also had to indicate their perception of working conditions with a scoring scale: constraints, professional and personal resources. Moreover, they had to evaluate the global perception of their job (positive and negative stress perception, tension and stress symptoms), and the level of personal involvement, their motivation, their global satisfaction in their job. Linear regression allowed describing predictive models of levels of stress perception (positive, negative stress, declared symptoms), levels of involvement and global satisfaction of work based on the perception scores. These models did not allow establishing a direct link with livestock management practices, or antimicrobial uses. Nevertheless, significant differences and correlations appeared between perceptions scores and some practices. For example, farmers who declared to be particularly watchful about early postnatal rearing conditions of broilers, seemed to declare less symptoms of stress than the others on average [$F(1,61) = 4,8$; $P = 0,03$]. Moreover, farmers who declared rather an increase of their antimicrobial uses over these ten past years, had lower perception score of constraints in their job than the others on average [$F(2,61) = 4,0$; $P = 0,02$]. The results of this survey were presented to a student group who received a training course to become technical advisers in poultry production. The pedagogic exercise aimed to raise awareness on the importance to integrate human factors comprehension in technical advices to lift barriers, understand expectations and foster transitions towards prudent use of antimicrobials.

Keywords: Antimicrobial use, Pedagogic exercice, Poulry, Psycho-sociological factor

Assessment of the development of parental broiler Cobb 500 by non-invasive imaging techniques

Abstract ID: 185

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In order to follow the composition of parental broilers (Cobb500) during growth from hatching to adulthood (32 weeks of age), we evaluated kinetics of fattening, growth rate, reproduction parameters and body composition of the animals by using non-invasive tools such as medical imaging and blood sample analysing. The use of CT scanner allowed us to monitor the development of body composition on the same animals who are sleeping (fatness, bone, muscle, ovary and testis growth). These analyses were associated with biochemical blood analysis. Parental broilers fed the recommended diet by breeders. The analyses were performed with a CT scanner of bio-medical grade (Siemens As Definition; 100K Volta and 120 mA/s). Five hundred images were performed every 0.6 mm, 0.45 Pitch and reconstruction filter Safire I26 was used to characterize fat tissues, skeletal and muscle. Image processing by thresholding was used to quantify and monitor the tissues from 4 weeks to 32 weeks of age. The acquired data were used to compare males and females. A significant difference in volume of adipose tissue was observed between male and female. From 20 weeks of age, shortly before the onset of lay, the females had 1.6 fold more adipose tissue than males ($P < 0,001$). In the female group, the increase in adipose tissue is associated to elevated plasma triglycerides levels (average 8 fold more in comparison to male at the beginning of the laying period. In addition, females, from 16 weeks of age, present a slow bone development in comparison to male ($P < 0,001$). The ratio bone/bodyweight is 30% lower in female compared to male chicken at 32 weeks of age. A section of the pectoral muscle volume was also analysed but no difference has been noted between both sexes. The correlation between the number of yellow follicles of hens and volume of ovarian adipose tissue determined in vivo was 0.80. In conclusion, the use of CT scanner and ultrasound system has permitted to follow the body composition of an actual parental breed without dissection.

Keywords: CT scanner, Cobb500, Growth, Parental broiler

Determening the haemoglobin concentration in hen's (Gallus gallus) blood using spectophotometry (cyanmethemoglobin method) with and without centrifugation: should the rutine method be reevaluated?

Abstract ID: 618

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OBJECTIVE: To determine whether the routine haemoghlobin determination method with Drabkin's solution should be corrected for chicken blood samples.
MATERIALS AND METHODS: Blood was drawn from a wing vein on lithium heparin from 15 Brown hissex hens. Haemoglobin concentration was measured using standard cyanmethaemoglobin method (Drabkin's solution) on a spectrophotometer at 540 nm against water. The same samples were centrifugated at 1600xg, 10 min at room temperature and haemoglobine concentration was once again measured using the same protocol.
RESULTS: After the centrifugation, at the bootom of the test tube, the red stained sediment was noticed. The sediment was microscopicaly assesed and incomplete erythrocytes lysis was determined.
Comparing haemoglobin concentrations before and after the centrifugation, it was determined that after the centrifugation haemoglobin concentrations were 60 to 130% lower, comparing to those without centrifugation.
CONCLUSION: Cyanmethemoglobin method is well known and world wide used rutine haemoglobin concentration determination. However, during routine chicken blood analyzes, it was noticed that some specimens were more cloudy then the others, having elevated absorbances, and respectively, haemoglobin concentrations. In these research was fund that analyzing mixture was not cloudy just due to red blood cell's nuclei but also unlysed red blood cells.
It was concluded, if reliable haemoglobin concentration values are needed, the correction of the method should be considered.

Keywords: Centrifugation, Chicken, Corrections, Cyanmethemoglobin, Haemoglobin

Effect of emulsifier and carbohydrase in a maize-wheat-SBM-tallow diet on metabolic and hormonal profile in broiler chicken

Abstract ID: 473

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In view of the desire to improve the process of breeding chickens for fattening, feed additives (like emulsifier and carbohydrase) are used to improve the digestibility and use of feed. Emulsifiers in a natural way mediate the absorption of hydrophobic diet components as free fatty acids contained in micelles. Literature data and our previous research demonstrate that the addition of exogenous emulsifiers improves the digestibility of fats and may cause a decrease in their content in digesta. The consequence of reduced fat content in digestion is to improve the availability of carbohydrates for the intestinal microflora which may adversely effect on the digesta viscosity or/and digestibility deterioration. In order to prevent these changes, carbohydrases are also used in practice. Although the effect of these feed additives on breeding parameters is relatively well known there is still little data on their effect on the metabolic and hormonal profile of broiler chickens. The aim of the present experiment was to evaluate the effects emulsifier and xylanase on hormonal profile and lipids and glucose levels in serum blood of broiler chickens. The experiment was conducted on ROSS 308 male chickens. Birds were divided into four groups. The first group was fed a basal diet (BD) (maize-wheat-SBM-tallow) without any supplementation. The second treatment consisted of BD and an emulsifier additive (E), whereas BD in the third group was supplemented by xylanase (X). In the fourth group both supplements were added to the diet (E+X).
Blood parameters levels were determined by enzymatic, colorimetric assays. Hormone concentrations in serum were analyzed using specific ELISA or RIA kits. Statistical analyses were performed using One-Way ANOVA with Duncan's post-hoc test.
We found that xylanase supplementation statistically significant reduced the levels of total ghrelin ($p \le 0.05$) and spexin ($p \le 0.05$) in all experimental groups in comparison to the birds fed a basal diet. The increase of insulin concentration was observed in the second group (E) and the fourth group (E+X) compared to BD birds. However, these differences were not statistically significant. The markers of liver damage and biochemistry parameters concentrations did not change, suggesting no negative effect of xylanase on metabolism.
The research was supported by National Science Center Poland 2015/19/D/NZ9/03580 SONATA

Keywords: Emulsifier, Hormones, Physiology, Xylanase

Expression of spexin and its receptors in chicken tissues

Abstract ID: 468

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Spexin (SPX) is a novel peptide coded by *SPX* gene involved in regulation of energy metabolism, puberty and reproduction in fish and mammals. It was previously showed that SPX inhibits food intake, long chain fatty acid uptake into the isolated mouse adipocytes and it is also involved in regulation of insulin secretion in fish. Spexin exerts a variety of physiological roles in the neuroendocrine and metabolic system through its interactions with two galanin receptor subtypes (GalR2 and GalR3). However, little is known about the characteristics of spexin gene and receptors in birds. Our previous research conducted on mammals (Kolodziejski et al. 2018) showed that SPX could determine the metabolic status. Therefore, we decided to investigate expression of SPX gene and its receptor in various chicken tissues. The identification of SPX and GALR2/R3 genes in chicken tissues was conducted using tissues from ROSS 308 male chickens. For determination of SPX gene total RNA was isolated from tissues using Tripure Isolation Reagent according to the manufacturer's instructions and cDNA was generated from 1 µg total RNA. Real-time polymerase chain reaction (PCR) was performed using gene-specific primers. We found that SPX and GALR2 and GALR3 genes are expressed in various chicken tissues (like: fat, liver, intestine, muscle, brain and spleen). A wide distribution of SPX gene in broiler chicken might suggest a potential role of SPX in regulation of metabolism of these species.
The research was supported by National Science Center Poland 2015/19/D/NZ9/03580 SONATA and 2015/19/N/NZ4/00572 PRELUDIUM grants.

Keywords: Chicken, Galanin receptors, Spexin

Growth rate of broiler chickens is influenced by early life feeding strategy

Abstract ID: 231

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After hatching in conventional systems, broiler chickens have a delay to nutrition that can last for 72h, depending on length of the hatch window, internal hatchery procedures and transport duration. Previous research on early life feeding strategies has shown negative effects on bodyweight (BW) gain after delayed nutrition (DN), compared with early nutrition (EN). However, it is not known whether DN chickens can (partially) compensate for their lower BW between hatch and slaughter. In this study, we tested the hypothesis that DN chickens have an increased growth rate, as a result of compensatory growth. Data from 3 independent experiments were used. In these studies, broilers were subjected to either EN or DN with different durations of DN (38 to 72 h) and days to slaughter (14 to 35 d). In all experiments, DN groups had lower BW compared with EN which was sustained until slaughter. Relative differences in BW, however, decreased from 114 to 176% post placement to 102 – 112 % at slaughter (35 d). Growth curves of DN and EN chickens were analysed to study whether compensatory growth could explain the differences in BW between EN and DN. Absolute average daily gain (aADG) was higher in EN chickens from start until slaughter. To analyse the growth curve independent of BW, relative ADG (rADG) between two ages was calculated as follows:

$$rADG\ /\ (\%) = \frac{(BW_{end} - BW_{start})\ /\ (Age_{end} - Age_{start})}{BW_{start}}$$

Differences in rADG between DN and EN chickens were greater in the first 14 d (DN: 63%, EN: 47%; P < 0.001), but smaller in the remaining grow-out period (14 – 28 d: DN: 18%, EN: 16%; 28 – 35 d: DN: 8%, EN: 7%; both P <0 .001). Based on these results, it seems that DN broilers compensate for their lag in BW during the first 14 d post placement. As differences in absolute BW were still present at 35 d, the increase in rADG seems insufficient to catch up with EN broilers. EN chickens have higher aADG until slaughter, however, rADG is lower, showing that growth rate is influenced by feeding strategy. Previous literature describes interactions between compensatory growth and nutrient composition of diets on nitrogen and fat retention. This may give reason for future work to evaluate effects of early life feeding strategy on carcass traits.

Keywords: Compensatory Growth, Delayed Nutrition, Early Nutrition

Highly sensitive embryonic marker of the prospective meat productivity in poultry

Abstract ID: 101

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Meat productivity in poultry is determined by growth rate, bodyweight, efficiency of feed consumption, and other genetically predetermined parameters. Histological studies proved that the rate of the myogenesis is irrespective of productivity type. The differences in muscle weight are related to number and thickness of muscle fibers. This difference emerging only during the posthatch development. However, certain biochemical factors acting during the embryogenesis determining postnatal muscle growth. Genotype can predetermine the synthesis of these factors, the presence and sensitivity of their targets. According to certain authors, the activity of these factors are related to nitric oxide (NO). Our data confirm the intense NO production in developing bird embryo. The intensity of this process is similar in egg- and meat-type poultry. The intensity of NO oxidation to nitrate, however, is repeatedly higher in meat-type breeds as compared to laying breeds. Oxidation intensity is, therefore, determined by certain structure(s) within the embryo. Experiments with intraembryonic injections of NO donors evidenced that these structures appear (or activated) as early as on days 2-4 of incubation and then acting until the end of embryogenesis. The appearance (or activation) of these structures is apparently genetically predetermined. The differences in individual intensities of embryonic NO oxidation within a single breed do not exceed 10% irrespective of parental age, diet, or egg weight. The structures responsible for embryonic NO oxidation still unknown. We examined over 50 breeds in 5 poultry species and found that the intensity of embryonic NO oxidation in breeds selected for meat productivity was repeatedly higher as compared to the initial breeds. Green light, a well-known exogenous stimulator of muscle growth, was also found to enhance (by 20-30%) the intensity of embryonic NO oxidation. The intensity of embryonic NO oxidation to nitrate is therefore a highly sensitive marker of the presence (or activation) of embryonic structures promoting muscle growth. This embryonic parameter can be used as a criterion in the selection of poultry for improved postnatal growth rate. The study was financed by subprogramme "Investigation of the adaptation of the digestive system in mammals and poultry to diets with different ingredient composition", resolution of the Presidium of Russian Academy of Sciences No 132, 07/05/2017.

Keywords: Embryo, Meat productivity, Nitric oxide (NO)

Investigations on the effects of heat stress on hepatic cell culture models of chicken origin

Abstract ID: 489

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Heat stress is one of the main stress factors in intensive poultry farming, contributing to animal welfare and production issues. Cellular prooxidant (mainly H_2O_2) production is commonly increased by heat stress, while the enhanced heat shock protein (HSP) expression effectively facilitates the metabolic adaptation to the altered environmental conditions. As the liver plays central role in the heat stress response, in which the specific role of different cell types is not fully elucidated, the main goal of the present study was to investigate the hepatic cellular response to heat stress in broilers.
In vitro experiments were carried out on primary hepatic cell culture models of chicken origin. Hepatocytes and Kupffer cells were isolated from the liver of 20-day-old Ross 308 broilers by multi-step perfusion, collagenase-mediated digestion and a subsequent differential centrifugation. To study the specific role of different cell types, hepatocyte mono-cultures and co-cultures with cell ratio of 6:1 (hepatocyte to Kupffer cells) were prepared from freshly isolated cells, the latter model mimicking a mild inflammatory state. After 24 h of culturing, cell cultures were incubated at 44°C for 1 or 2 h, while control cells were cultured at 37°C during the entire trial. Viability of cells was assessed directly following heat exposure and after 20 h recovery time with MTS and CCK-8 assays; the concentration of H_2O_2 in culture media was monitored with the Amplex red method.
Cell viability was significantly ($P<0.05$) decreased by heat stress on both cell culture models directly after heat exposure. A prolonged deterioration in cell viability was observed following 20 h recovery time only if cells were exposed to heat stress for the longer incubation time ($P<0.05$). In this latter case, cellular H_2O_2 production was significantly ($P<0.05$) increased on both cell cultures, but with a remarkably higher extent on co-cultures than on hepatocyte mono-cultures.
According to our results, the applied cell culture models seem to be proper tools for studying hepatic cellular response to heat stress. It can be concluded based on our results that heat stress can highly diminish viability of liver cells in chicken. The heat stress-evoked elevation in H_2O_2 production of cultured cells suggests that the hepatic release of prooxidants may play central role in the oxidative stress related to heat stress response. Further, the key function of Kupffer cells in the stress-induced prooxidant production was also proven in the present study.

Keywords: Animal health, Animal welfare, Heat stress, Kupffer cells, Liver

Qualitative versus quantitative feeding in the broiler breeder: a route to manipulating satiety?

Abstract ID: 578

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The orexigenic AGRP neurones in the hypothalamus of broiler breeder chickens are sensitive to feeding history (Dunn *et al.* 2013). In broiler breeder hens food restriction improves reproduction and health but increases hunger. We are interested in understanding the way that food intake is controlled to try and tackle this 'broiler breeder paradox'. One approach has been diets diluted with fibre. We hypothesised that when diets were diluted with fibre and fed iso-energetic with the usual ration to maintain the target growth curves, the expression of AGRP and the anorectic gene, POMC would not differ from quantitatively restricted hens.
Broiler breeder hens were reared on a standard restriction to 6 weeks of age when they were assigned randomly to 4 replicated treatment groups (n=24); standard restriction on a typical commercial diet (R), the same diet with 20% (OH20) or 40% oat hulls (OH40) added to attain the usual growth curve, finally one treatment was released to *ad-libitum* feeding with the commercial diet (AL). Hens were killed at 12 weeks of age and the hypothalamus containing the orexigenic and anorectic peptide expressing neurones were collected for measurement of gene expression by RT-PCR.
The body mass on the quantitatively (R) and qualitatively restricted diets (OH20&40) were similar whilst the AL hens had a higher mass (R, 1324±20g; OH20, 1312±22g; OH40, 1349±16g; AL, 2851±75g). As anticipated, there was a highly significant ($P<0.001$) effect of diet on AGRP expression with R, OH20 and OH40 being ~10 fold greater than AL hens. There was no significant difference within the restricted hens (R, 0.50±0.05; OH20, 0.55±0.06; OH40, 0.47±0.06; AL, 0.05±0.01). In contrast, but as observed previously, there was no statistical effect of diet on POMC expression (R, 165±49; OH20, 157±29; OH40, 137±24; AL, 204±38).
The results strongly suggest that although the OH40 diet was fed near *ad-libitum* it did not result in an effect on orexigenic gene expression in the brain. The diets show benefits in allowing near *ad-libitum* feeding. However, different methods, including behavioural assessment and measurements of how gut fill inhibits intake, will be required to understand their welfare benefits.
We acknowledge BBSRC grant BB/L000199/1 and strategic programme grant BB/P013759/1.

Keywords: Broiler breeder, Gene expression, Qualitative feeding, Quantitative feeding

The effect of age and diet type on the hepatic and intestinal CYP activity in broiler chicken

Abstract ID: 412

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The CYP450 enzymes play an important role in the metabolism of xenobiotics. They are mainly localized in the liver, however a significant amount can be found also in the small intestines, which determine the amount and form of consumed xenobiotics reaching the portal circulation. The aim of this study was to follow the activity of hepatic and intestinal CYP1A2 enzymes at different ages in broiler chickens, fed on various cereal types of diet.
Sixty male Ross 308 broiler chickens were fed with maize- or wheat-based diet (the latter with xylanase and glucanase supplementation). Tissue samples from liver and mucosal scraping samples of duodenum and ileum were taken at week 1, 3 and 6 (n=10/group at all age). After homogenization of the samples S9 fractions were isolated by differential centrifugation. The CYP1A2 activity was determined by luminescent CYP Glo Assays.
In the liver, the activity of CYP1A2 enzymes was significantly higher ($P<0.001$) in the wheat based diet group compared to maize based diet, regardless of age. However, concerning diet type no significant difference was found either in the duodenum or the ileum. In contrast CYP1A2 activity in the liver did not change with age, but we found a significant increase (week 1 vs. week 6: $P=0.048$) in the duodenum and a marginal significant increasing trend (week 1 vs. week 3: $P=0.072$; week 1 vs. week 6: $P=0.096$) in the ileum.
Our results suggest, that the composition of the diet could influence the activity of the CYP enzymes in the liver, thus the ability of the liver to metabolize xenobiotics. The lack of significant effect of diet type on the intestinal CYP activity could be due to the higher individual variability of these enzymes in the small intestines. The influence of age on the intestinal, but not on the hepatic enzyme activity can be explained by the fact, that the stable function of liver is developed already at age of one week in broiler chicken. However, due to the changing quantity and quality of consumed feed during raising, the relevance of small intestinal detoxification thus the activity of intestinal CYP enzymes could increase with age.

Keywords: Age, Broiler chicken, CYP450, Diet

Antibiotic resistance pattern of Staphylococcus aureus isolates recovered from broiler breeder farms in Iran, 2014-2017

Abstract ID: 112

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Staphylococcus aureus is a highly versatile pathogen of a large number of domestic animals, including avian species. *Staphylococcus aureus* is gram positive bacteria that could infect primarily or secondarily poultry species with major concern in farming. The most common site of staphylococcal infections are bones, joints and tendon sheaths that cause lameness in broiler breeder flocks. lameness in broiler breeder flocks, especially in males, may result in severe economic losses. The aim of this study was to determine antibacterial susceptibility pattern of *Staphylococcus aureus* isolated from birds with symptoms of lameness. During 2014-2017, 64 isolates of *Staphylococcus aureus* were recovered from breeder flocks using standard bacteriologic procedures and the respective drug resistance patterns were determined to a panel of 20 antimicrobial agents by agar disk-diffusion method. Due to bacterial resistance to antibiotics, the diameter of growth prevention varies. Results of antibiogram test were reported as sensitive, moderate sensitive, intermediate and resistant classes. The findings of this study demonstrated that *Staphylococcus aureus* isolates were 62, 59 and 53% resistant to Erythromycin, Clindamycin and Doxycycline, respectively. The multi-drug resistance (MDR) was found among all isolates. The MDR pattern was variable and ranged from 0 to 16 drugs. In total, all 64 isolates generated 56 different patterns of antimicrobial resistance. The relationship between the occurrence of resistance and the consumption of the antimicrobial was studied. The findings of the present study may be useful for choosing an appropriate antibiotic by clinician to control infection.

Keywords: Antimicrobial susceptibility, Breeder, Lameness, Staphylococcus aureus

Does disruption of the disulphide bonding in Ovodefensins affect its antibacterial activity?

Abstract ID: 545

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The EU ban on prophylactic antibiotic usage in farm animals has left a demand for alternatives to ensure the efficiency of animal production whilst maintaining animal health and welfare. Our research found that the in-feed inclusion of naturally-occurring antimicrobial peptides, specifically a family called ovodefensins that are found in eggs, do improve growth and intestinal health in broilers. We wish to identify structural features of the ovodefensins required for activity with the aim of designing variants with improved activity.
Ovodefensins differ from classical defensins in the spacing of the 6 conserved cysteine residue motif – $CX_5CX_3CX_{11}CX_3CC$ or $CX_3CX_3CX_{11}CX_4CC$. Although mammalian and avian defensins have been characterised, the spacing or bonding of the cysteine residues has not been studied. NMR analyses show that disulphide bridges form between C1-C5, C2-C4, C3-C6 resulting in a β-defensin fold. In human β-defensin 1, reduction of disulphide bonds results in potent antibacterial activity. Therefore our aim was to synthesise and test ovodefensins variants where cysteine was substituted for alanine.
The peptide where alanine was used to replace cysteine at C3-C6 was unable to be produced in yields appropriate for characterisation. All others cysteine substitutions could be synthesised. Interestingly the peptide with the most potent antibacterial activity was that in which the C1-C5 cysteines had been substituted. A dose of 10µM gave more than 50% reduction compared with a negative control against *S. aureus*. When all cysteines were replaced significant antibacterial activity versus *S. aureus* remained, similar to the unaltered structure with ~ 50% reduction at 100µM. Disrupting the C2-C4 disulphide bond led to lower antibacterial activity even at higher doses (100µM to 200µM).
These results show the replacement of all cysteines and therefore all disulphide bonding led to no change in antibacterial activity. However, some disulphide bonding could be beneficial for the ovodefensin functioning because the removal of only the C1-C5 bond led to a significant increase in antibacterial activity. Future research in the form of a feed trial will be needed to assess whether this modified ovodefensin provides benefit to growth and intestinal health.

Keywords: Antibacterial activity, Disulphide bonds, Function, Growth, Ovodefensin

Isolation and characterization of ESBL and/or AmpC producing Escherichia coli isolates from broiler meat and caecum

Abstract ID: 395

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Antimicrobial resistance to third generation cephalosporins has increased dramatically in E.coli in human and veterinary medicine. Selective pressure, induced by the use of antimicrobial preparations, was the main force for the emergence and dissemination of drug resistance among pathogenic and commensal bacteria. From 2013 to 2016 in the EU/EEA countries, significant increase of bacterial resistance to third-generation cephalosporins was noted. Third generation cephalosporins are critically important for treatment in human medicine, therefore, their use in veterinary medicine is of great concern. More than 50% of sold antibiotics are used to treat animals. Their bacterial microflora plays a significant role in the emergence and transmission of antimicrobial resistance. In this study, 1885 samples of broiler caecum and 200 samples of broiler meat were examined, by the EURL-AR protocol, for E.coli isolates which produce ESBLs and/or AmpC and/or carbapenemases. Broth microdilution method was used for cefotaxime and/or ceftazidime resistance screening, and resistant isolates were examined with another commercial microtiter plate which contain only cephalosporins and carbapenems. According to phenotypic characteristics we decided if bacterial isolates are presumptive ESBL and/or AmpC and/or carbapenemase producers. A total of 1016 caecum samples (53.9%) and 126 mean samples (63%) were positive for E.coli which produce ESBL and/or AmpC. All samples were negative for E.coli which produce carbapenemases. Broth microdilution method was made for 300 E.coli isolates from broilers caecum and 126 E.coli isolates from broiler meat. According to the EFSA criteria 186 isolates (62%) were characterised as presumptive AmpC producers, 109 (36.3%) were characterised as a presumptive ESBL producers and 5 (1.67%) were found to be AmpC + ESBL producers. From 126 E.coli isolates from broiler meat 96 isolates (76.2%) were characterised as presumptive AmpC, 29 (23%) isolates are presumptive ESBL and 1 isolate (0.8%) was presumptive AmpC + ESBL producer.

Keywords: Broiler, Cephalosporinases, E.coli, Resistance

Prevalence and Antimicrobial Resistance of Salmonella from Poultry Processing Plants

Abstract ID: 612

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Salmonella in one of the top causes of bacterial foodborne infections in the US, which points out the importance of controlling this pathogen for public health. Poultry and poultry products, are commonly associated with *Salmonella*, and interventions during production and processing are necessary to manage the risk of infection due to consumption of poultry products. In recent times, demand for organic and antibiotic-free chicken has increased due to consumer perception of these products as safe and because of concerns over increasing antimicrobial resistance (AMR) in pathogens like *Salmonella*. However, the microbiological effect of these management practices is not clear. This study was conducted to determine the efficacy of antibiotic-free (ABF) management practices by assessing the prevalence of AMR *Salmonella* in a commercial broiler processing plant. Fecal samples, carcass rinses, and environmental samples (water, air, and equipment) were collected over a 1-year period. *Salmonella* was isolated by using the XLT4 agar and the presumptive positive *Salmonella* were further confirmed by PCR. These confirmed isolates were then tested for antimicrobial resistance using commercially prepared broth micro-dilution plates and the National Antimicrobial Resistance Monitoring System (NARMS) protocol. Results showed that organic chickens were associated with higher levels of *Salmonella* during early processing steps, however post-chill, no differences ($p>0.05$) were observed between organic and conventional carcasses. In addition, prevalence of AMR *Salmonella* was lower in this study than reported by NARMS for chickens at slaughter. Out of the 274 isolates (162 from organic and 112 from conventional processing), 20.8% were resistant to atleast one antimicrobial and 13.1% were resistant to three or more classes of antimicrobials making them multidrug resistant (MDR), whereas 79.2% (217/274) of isolates tested were not resistant to any antimicrobials. These findings reinforce the importance of interventions to control overall pathogen prevalence and highlight the continuing need for research on management strategies for AMR pathogens.

Keywords: Organic, Poultry processing, Salmonella

The choice of specific phages can increase the therapeutic efficacy of repeated phage therapy against colibacillosis in broilers

Abstract ID: 463

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Phage therapy presents an alternative to antibiotic treatment in the control of bacterial infections. However, the strong host immune response and rapid clearance from the bloodstream after repeated application of phage is a serious limitation of phage therapy which needs to be overcome. The aim of the study was to investigate if pre-immunization of broilers with one specific phage (TM1) would limit the ability of another phage to treat colibacillosis infection.
To this end, *in vitro* and *in vivo* antibacterial activity and immunogenicity of three phages (TM2, TM3 and TM4) were studied in pre-immunized broiler chickens.
The phages were previously observed to differ from each other with respect to their morphologies and restriction endonuclease digestion profiles. One phage (TM3) showed low immunogenicity and high survivability in broilers and was therefore selected to study its efficacy in the treatment of experimentally induced colibacillosis. The study included 7 treatments with 4 replicate pens of 15 chickens per pen. The treatments were: 1) negative control (non-immunized chickens, not challenged with *E. coli*); 2) pre-immunized chickens not challenged with *E. coli*; 3) pre-immunized chickens challenged with *E. coli* and not treated with phage; 4) non-immunized chickens challenged with *E. coli* and treated with phage TM1; 5) pre-immunized chickens, challenged with *E. coli*, and treated with phage TM1; ,6) non-immunized chickens challenged with *E. coli* and treated with phage TM3; 7) pre-immunized chickens, challenged with *E. coli*, and treated with phage TM3. Colibacillosis was induced by injecting of 0.2 mL *E. coli* O78:K80 (10^8 CFU) into the right thoracic air sac when birds were 12 day of age. The phages were administered by i.m. injection of 0.2 mL (10^{11}PFU) into the thigh muscle. Control chickens were injected with the same volume of PBS.
At 14 d post challenge, chickens pre-immunized with phage TM1 and treated with phage TM3 (treatment 7) showed significantly lower mortality and higher body weights (16.6%, 1135g,) than untreated birds (43.3%; 828g, treatment 3) and those treated with phage TM1 (36,7%; 976g, treatment 5). Non-immunized chickens treated with phage TM3 (treatment 6) showed significantly lower mortality than non-immunized chickens treated with phage TM1 (treatment 4) (10% *vs.* 20%). Likewise, the treatment with phage TM3 resulted in significantly higher body weights compared to the treatment with phage TM1 (1136g *vs.* 1041g).
In conclusion, the present study demonstrated that the inhibitory effect of broiler humoral immunity on phage therapy could be overcome when using a distinct phage with low immunogenicity, which is suggested to increase the therapeutic effectiveness of repeated phage therapy.

Keywords: Broiler chicken, Colibacillosis, Escherichia coli, Immunogenicity, Repeated phage therapy

Differential Impact of Yeast Cell Wall Preparations on Growth Kinetics and Antibiotic Sensitivity of Antimicrobial Resistant Bacteria

Abstract ID: 472

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Antimicrobial resistance (AMR) has the potential to become one of the greatest problems of our generation given the ever-increasing rise in bacterial strains which are becoming less sensitive to existing treatments. Currently, there is political pressure worldwide to restrict the use of antibiotics in animals to therapeutic use only, following on from the 2006 EU ban on the use of antibiotics for all non-therapeutic use as antibiotic growth promoters (AGPs).
Presently, there is a need to find alternatives to antibiotics for pathogen control within the agriculture industry. Of the functional ingredients currently in use for microbial control, yeast based preparations are widely used in animal nutrition and have been shown to improve animal performance while at the same time control the growth and proliferation of pathogens.
In this study mannan rich fractions (MRF) and glucan rich fractions (GRF) isolated from the cell wall of differing strains of *Saccharomyces cerevisiae* were assessed and compared with respect to their capacity to modulate the growth of antibiotic resistant bacterial strains.
The growth kinetics of ESBL producing *Escherichia coli* and multi- drug resistant *Salmonella* and *Escherichia coli* were examined in the presence of MRF and GRF. All strains were grown over a period of 18 hours in Mueller Hinton broth supplemented with MRF or GRF in conjunction with the antibiotics; ticarcillin, cefotaxime and ciprofloxacin at concentrations up to 1 mg/mL.
Our results demonstrate that MRF and GRF have varying abilities to modulate the growth kinetics of antimicrobial resistant bacteria. Moreover, differences were noted between sources of MRF and GRF with respect to the ability of individual preparations to influence resistant bacterial growth. Additionally, the effect of these preparations in modulating the sensitivity of individual resistant strains to antibiotics was assessed. Notable differences were observed in the ability of MRF and GRF preparations to effect changes in sensitivity to antimicrobials. As with the growth kinetic response, different preparations effected differential responses with respect to sensitisation to antibiotics. Overall this study indicates that yeast cell wall fractions have the potential to impact on growth of antibiotic resistant bacteria but that different preparations effect different responses.

Keywords: Antibiotic resistance, Mannan-rich fraction, Saccharomyces cerevisiae

A Ferula based product limits the decrease of performance in old laying hens raised in commercial conditions

Abstract ID: 346

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The impact of the dietary inclusion of a phytonutrient from *Ferula* spp. family on laying hen performance was evaluated in commercial conditions.At 20 weeks of ages, 80'000 Tetra SL Longlife hens were housed in enriched cages in a building containing 4 rows of 6 floors each. Two weeks before the start of the trial, the building was separated in two groups, to collect performance data per group before first product supplementation. From 61 weeks of age until start of culling at the end of 74 weeks, each part of the building was allocated to one of the two following treatments: 1) standard commercial diet (control) or 2) standard diet supplemented with 100 g/t of a *Ferula* based product (NEX, NexTend). Feed intake, mortality, number of eggs produced per class, laying rate, FCR and egg mass sold were collected daily. During the trial period, the weekly average was calculated for each outcome. Data collected during the trial phase were then analyzed by using linear regression, and differences in regression coefficients between the two treatments were assessed using GraphPad Prism. Before the trial, hens in future NEX group had lower performance than those allocated to control treatment: lower number of eggs and laying % (-3.0%, $P < 0.01$), and greater feed intake and FCR (respectively +7.6% and +10.7%, $P \leq 0.01$). During the supplementation period, regression analysis showed that the slopes of NEX group were higher than those of the control, for laying % (-0.68 vs -0.47 %/week, $P < 0.01$), number of eggs of grade A (-1502 versus -2039 eggs/week, $P < 0.01$), and egg mass sold (-92.04 vs -124 kg/week, $P = 0.07$), and lower for FCR (-0.018 vs -0.024 g/g/week, $P = 0.09$). These findings suggest that in commercial conditions NEX supplementation could slow down the performance decrease of hens at the end of the laying period. This could therefore limit the drop of production and benefit at the farm level.

Keywords: Laying hens, Performance, Plant extract

Application of IgY antibody for prevention of atopic dermatitis - Suppression effect of anti-S.aureus IgY on S. aureus growth

Abstract ID: 411

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S. aureus (SA) has been considered one of causative bacteria of atopic dermatitis. Normally, the resident bacteria on healthy skin including *S. epidermidis* (SE) inhibit the growth of SA. However, it has been known that SA grows predominant on skin in atopic constitution. The purpose of this study is to evaluate the growth inhibitory effect of anti-SA IgY against *S. aureus*. This study might lead to a novel application of an anti-SA IgY for prevention of atopic dermatitis using a skin care cream containing an anti-SA IgY antibody.

Control IgY or anti-SA IgY was adjusted to a concentration of 10 mg/ml, and each of the IgY solution and SA solution was mixed at a volume ratio of 1: 1. Bacterial growth was monitored by absorbance at 600 nm. Cell growth was also monitored by ATP chemiluminescence assay at 0, 4, 6, 8, 12 and 24 hours incubation at 37 ° C. HPLC analysis was applied to calculate specific binding IgY ratio in IgY protein sample against SA and SE. In addition, the growth inhibitory effect of anti-SA IgY against SE was also determined.

The specific binding antibody ratio in the anti-SA IgY protein was 18.1% against SA. This value was about two times higher than that against SE. In addition, it was found that the growth inhibitory effect of anti-SA IgY showed delaying the lag phase of SA growth curve for 4 hours compared to that of Control IgY. However, there was no delaying effect of the anti-SA IgY on the growth of SE. Four hours of bacteriostatic on SA growth could be a prevention measure against atopic dermatitis in using a skin care cream containing an anti-SA IgY antibody.

Keywords: Anti- S. aureus IgY, Atopic dermatitis, Bacteriostatic, Egg yolk antibody

Confirmed and suspected cases of poisoning in wild birds in Croatia from 2010 to 2017: a pathomorphological survey

Abstract ID: 587

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During the years 2010 to 2017, a pathomorphological survey of the causes of death of wild birds in Croatia was made. A total of 5457 birds were examined. In 274 wild birds of various species, a probable or a definite cause of death was established. Intoxication was toxicologically confirmed only in 26 (9.48%) cases. *Clostridium botulinum* toxin type C caused death in 19 (73.07%) birds (pheasants; wild ducks). In seven cases, intoxication with rodenticide and carbofuran was confirmed in water birds and birds of prey.
Some experts worldwide believe that the only relevant poisoning in wild birds is acute and easily recognizable lethal poisoning toxicologically confirmed. On the contrary, the majority of literature data emphasize that the most dangerous (especially as a source of secondary intoxication for humans and animals) are chronically intoxicated animals (especially wild birds). In these cases, as unspecific symptoms and gross lesions are usually hidden behind various bacterial and/or viral infections, toxicological analysis is unnecessarily ignored. This may be one of the major causes of failure in the implementation of the concept of "One Health". As it is known that the biomarkers of exposure and toxic effects for some poisons are similar in humans and in sentinel animals (e.g., the effect of organophosphorus insecticides), much more attention should be paid to comprehensive surveillance and/or detailed research and care of ill (poisoned) (wild) mammals and (wild) birds.

Keywords: Wild birds; intoxication; pathomorphology, „One Health"; Croatia

Evaluation of protective immune response against Salmonella in laying hens

Abstract ID: 440

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Salmonellosis remains the second most frequently reported cause of food-borne zoonosis in humans in the EU. Vaccination programmes against most common serovars are part of the control measures implemented to reduce the prevalence at farm level. Understanding the mechanisms of protection that vaccines confer to animals can help to develop effective and safe vaccines.
Our objective was to compare mucosal immune response to *Salmonella* Enteritidis and *Salmonella* Typhimurium field strain infection in two groups of vaccinated (n=25) and non-vaccinated (n=33) laying hens. Vaccine used was a bivalent live attenuated vaccine against the two abovementioned serovars. Immunological parameters were evaluated at 7, 14 and 21 days post infection (samples from euthanized animals): IgY levels in serum; IgA levels in bile; RNA expression in ileal mucosa of γ-IFN, IL-2, IL-6, IL-10, TLR-4, TLR-5 and MCH-II; and quantification of immune cell populations in villus and gut-associated lymphoid tissue (GALT) from small intestine.
We did not find significant differences between vaccinated and non-vaccinated groups regarding IgY levels in serum nor IgA levels in bile. We found significant differences in γ-IFN and IL-10 expression level comparing vaccinated and non-vaccinated animals. Vaccinated animals showed significant higher expression of γ-IFN (correlates with *Salmonella* clearance) and lower expression of IL-10 (controls inflammatory response during infection). Results of quantification of immune cell populations will also be presented.
This study indicates that humoral responses may not be correlated with protection of *Salmonella* vaccines. Understanding of cell-mediated immunity can be a better way to determine effectiveness of new vaccines.

Keywords: Cellular immunity, Live attenuated vaccines, Lymphoid tissue, Mucosal immunity

Evaluation of the impact of a highly concentrated chlorine neutralizer on poultry live vaccine titer simulating a vaccination with a dosing pump for drinking water distribution

Abstract ID: 650

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Drinking water (DW) vaccination is probably one of the easiest methods to deliver a live vaccine to a large number of birds. However, good quality of water as well as a quick and uniform intake by the flocks are critical to ensure effective vaccination. The use of a chlorine neutralizer is highly recommended to help guarantee the live vaccine viruses titer taken by the birds. Three main types of water distribution systems to vaccinate birds by DW: header tank distribution, medication tank distribution, or dosing pump distribution. Dosing pump injectors (also known as Medication pumps) are widely used and they allow a proper dosage of additives into the drinking system (antibiotics, probiotics, etc.), supplements (vitamins, minerals, etc.) and also vaccines. When proportionally pump injectors are used, drinking water distributed live vaccines may be injected directly into the water together with chlorine neutralizer in high concentrated dose.
The objective of the present experimental study was to simulate a vaccination using a pump injector and therefore to evaluate the impact of chlorine neutralizer in high concentrated dose (approx. 50 times) on live vaccine titer. Two different vaccines were tested: one live attenuated vaccine against Newcastle disease (VG/GA AVINEW vaccine strain in effervescent formulation) and a live attenuated vaccine against Infectious Bursal Disease (S-706 strain). In group 1 (G1), no water stabilizer was added to the vaccinal solution (control group). In group 2 (G2), commercial thiosulfate (Thiosulfate Blue®), 50-fold concentrated were added in vaccine solution. In group 3 (G3), commercial stabilizer (NeOStab®), 50-fold concentrated were added in vaccine solution. The vaccine titer for each tested vaccine and for each experimental group was measured after the vaccine reconstitution and at three different time-points after mixing with chlorine neutralizer: 0.5, 1 and 2 hours after reconstitution. A statistical analysis was performed in order to correlate the titres of the vaccines reconstituted in highly concentrated chlorine neutralizers with the control groups (ANOVA test, STATGRAPHICS Centurion XVII®).
The ANOVA didn't highlight any effect and interaction: there weren't found any statistically significant differences between the results obtained with the Chlorine Neutralizers and those obtained without a neutralizer at any time-points after reconstitution (0.5, 1 and 2 hours).
The results of the present study showed that the two chlorine neutralizers tested in the study did not have any impact on the titers of the two commercial live attenuated vaccines tested.

Keywords: Chlorine Neutralizer, Live Attenuated Vaccines, Medication Pump, Vaccination

Exposure of poultry workers to air dust in poultry slaughterhouses in France

Abstract ID: 524

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More than 32,000 people work in poultry slaughterhouses in France. Among them workers handling and shackling live birds may be exposed to aerial dust and microorganisms. ACCROCH'AIR project aimed to assess the potential impacts of those bioaerosols on respiratory health of workers handling poultry. Thirty visits were carried out in 17 slaughterhouses in the Western part of France to quantify workers' exposure to aerial dust. Ambient dust strongly varied among the 27 slaughter lines studied according to the workplace arrangement. Closed shackling cabins showed high levels of aerial dust despite specific ventilation systems; ceiling air ducts improved air circulation and tended to lower dust concentration. Aerial dust concentration was lower when poultry were handled in very large warehouses where dust quickly diffused away from workers. However exposure to inhalable dust (diameter < 100 µm) was often high whatever the workplace arrangement was. Exposure to respiratory dust (< 5 µm) was also high but under the regulation threshold. Workers' exposure closely correlated to ambient dust concentration suggesting that preventive measures implemented in the shackling area are likely to have a significant impact on workers' exposure. An in-depth diagnostic of available technical solutions for collective prevention is needed for every identified risky workplace. Wearing an adequate respiratory mask is strongly recommended for workers handling poultry.

Keywords: Air dust, Exposure, Poultry, Slaughterhouse, Worker

Japanese quail chorioallantoic membrane - experimental in vivo model

Abstract ID: 478

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Avian chorioallantoic membrane (CAM) is an excellent *in vivo* experimental model for studies in physiology, pharmacology, cancer research and other fields. CAM is thin, highly vascularized extraembryonic membrane which functions as a primary respiratory organ of the embryo. It holds structural similarity to e.g. buccal or bladder mucosa, lungs, placenta and is suitable for *in vivo* testing of potential drugs, toxicity testing or transplantation studies. Most often used is chicken CAM, however, here we are presenting various possibilities and advantages of Japanese quail CAM.
Japanese quail embryos were cultivated *ex ovo* in plastic cultivation plates from embryonal day (ED) 3. On ED 7 tested substances were applied topically on CAM surface. We examined pro and antiangiogenic effect of leptin and fractionated and unfractionated heparins. Fractal and histological analysis showed stimulating effect of leptin and inhibitory effect of fractionated heparin. In another series of experiments we explored possible use of quail CAM for photodynamic diagnosis of cancer and yeast infection. TE1 tumour spheroids
were cultivated on CAM surface from ED7. On ED8 photosensitizer hypericin or hypericin with low density lipoproteins (LDL) as a delivery molecule was added. In comparison to white light, presence of hypericin very well visualized the tumour spheroid position in the fluorescent image, with gradual increase of fluorescence intensity in time. Addition of LDL improved and accelerated the detectability of tumour. Similar results we obtained with different yeast strains, simulating microbial infection.
Our results demonstrate broad usability of Japanese quail CAM model for the study of normal and pathological angiogenesis, photodynamic diagnosis and therapy of cancer or microbial infections.
This research was supported by APVV-15-0485 and VEGA 2/0096/18.

Keywords: Angiogenesis, Cancer, Chorioallantoic membrane, Microbial infection, Quail

Presence of Campylobacter spp. in farm-reared Red-legged Partridge (Alectoris rufa)

Abstract ID: 646

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Nowadays, more than 70 million game birds are produced in Europe just among red-legged partridges and pheasants for hunting purposes. In the USA the upland game bird industry represents a $5.000 million impact and growing business. This production is usually made in semi-intensive systems, where the birds can contact with the external environment through the flight pen. In these species, 84% of the antibiotics prescriptions are related to intestinal disorders. Little is known about the role of *Campylobacter spp.* in these birds, but some reports show that farm-reared and restocked partridges can act as carriers of enteropathogens increasing the risk of transmission to natural populations throughout the releases.
During the summer of 2017, two batches of 2080 and 4400 partridges were followed in France, to identify the presence of *Campylobacter spp.* in each intestinal segment. Five animals per batch were weekly collected to evaluate gut lesions and alterations, and then performing a pool of intestinal content. The intestinal content of each segment was collected with a sterile inoculation loop and diluted in saline peptone, homogenized and then seeded in a specific culture media for *Campylobacter spp.* (CASA) within 30 minutes of collection. The study lasted for 10 weeks and revealed that the access to the flight pen determines the presence and charge of *Campylobacter spp.* with high significant correlations in the duodenum (P = 0.95), jejunum/ileum (P = 0.85) and caecum (P = 0.81). These results show that the flight pen can represent a risk of contamination, but it remains unclear if it represents a potential risk for other species, or for partridges themselves like a trigger-point for some enteritis.
Since the production of free-range poultry is similar to that of game birds because they have continuous daytime access to open-air runs, one can speculate that the same problem may exist in Poultry, with a high risk of food contamination by *Campylobacter spp.*

Keywords: Campylobacter spp., External Environment, Flight pen, Game Birds, Red-legged Partridge

Sensory evaluation and consumers' acceptance of raw sausages from chicken and pork

Abstract ID: 184

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The aim of this research was to compare consumers' attitudes toward chicken delicacy salami and royal salami, meat products from Zlatiborac Meat Company, Serbia. Chicken and pork meat for chicken and royal salami, respectively, were prepared by the meat industry (Zlatiborac, Mačkat) using their own production protocols. Sensory evaluation was performed by Serbian consumers (n=1018) over six days during the period from 10 a.m. to 4 p.m. in four retail stores in Belgrade. The origin and market name of the products were unknown to the consumers. Consumers were males (37%) and females (63%) older than 18 years of age. They were asked to express their perception of taste (good, satisfactory, non-satisfactory), salt content (well balanced, not salty enough, too salty) and smoke flavour (balanced, too weak, too strong) of the products. A questionnaire administered was a modified version of the DLG-5-points-scheme. After sensory evaluation, each consumer answered the questionnaire, which concerned general socio-demographic information related to ages and education levels. Female consumers evaluated the taste of royal salami as good (90%), a higher percentage compared to chicken salami (84%). Two groups of consumers in relation to age (30-39 years old and 50-59 years old) evaluated the taste of royal salami as good, higher than the percentage rating chicken salami as good (90% vs. 81%; 90% vs. 78%, respectively). However, only consumers with elementary school education evaluated the taste of royal salami as good and smoke flavour as balanced, which was a higher percentage of positive scores in comparison with chicken salami (92% vs. 77%; 90% vs. 83%, respectively). The salt content of royal salami was evaluated as well balanced, with a higher percentage of consumers reporting this than for chicken salami, by male and female consumers (90% vs. 79%; 92% vs. 77%, respectively), as well as by all age groups of consumers (18-29 years, 30-39 years, 40-49 years, 50-59 years and 60 and older). The same attitudes were seen regardless of consumers' education levels (elementary school, high/secondary school or graduate degree). Sensory evaluation of the chicken and royal salamis showed that consumers were satisfied with their taste, salt content and smoke flavour. More than 80%, 75% and 90% of consumers evaluated the taste, salt content and smoke flavour of the products as good, well balanced and balanced, respectively. Additional confirmation of the quality of the products from Zlatiborac Meat Company is reflected in DLG awards obtained in the period 2009-2017.

Keywords: Chicken delicacy salami, Consumers' attitudes, Royal salami

The physical and chemical evaluation of potential litter material sources for suitability as broiler litter

Abstract ID: 538

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The high mortality rates in broiler rearing houses associated with poor quality litter material was a concern for this study. The study was carried out at the National University of Lesotho farm to evaluate the physical and chemical properties of the potential litter material that could replace the wood shavings. The three potential sources were compared to control and the four treatments were as follows wood shavings (control), dry pine tree leaves (DPL), decomposed kraal manure (DKM) and sand. A total of 320 day-old Ross 308 chicks were reared in a well-ventilated house with 24 hour photoperiod during the first week and 20 hour thereafter. Feeds and water were provided ad libitum. General routine management for broilers was observed throughout the experimental period. Weekly data was collected on production performance while physical and chemical samples of litter materials were collected three times at the beginning of trial, at end of 3rd and 6th weeks. Samples were collected from five locations within each pen for a total of sixteen, thoroughly mixed and subsamples taken for analysis. Bulk density, moisture content, pH, water holding capacity and ammonia emissions and were determined according to Brake et al (1992). Temperature was measured by thermometer. The physical and chemical properties were all differing significantly ($p<0.05$) between the litter treatments with exception of temperature. Compared to the control the DKM was found to be outstanding in terms of bulk density, moisture content and temperature, DPL was the best in water holding capacity and pH while sand was good in ammonia emissions only. Sand was low in temperature, water holding capacity which means it was colder and wetter because of poor water holding capacity, it was also the bulkiest of them all. All production performance parameters were statistically ($p>0.05$) similar between the litter materials. The highest feed intake and body weight were found in sand, followed by DKM while the highest FCR and growth rate were found in DKM and DPL respectively. The lowest growth rate was observed in sand. The mortality rate was zero in both control and DKM. When considering the physicochemical properties and production performance it was clear that DKM was the best alternative litter material that can replace wood shavings.

Keywords: Broiler, Litter material, Physical and chemical properties, Wood shavings

The production of ammonia and greenhouse gas emissions from Slovak poultry farming in 2016

Abstract ID: 525

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Poultry farming plays an important role in livestock farming in the territory of Slovakia. Poultry meat is easily digested and therefore belongs to dietary foods. Meat and eggs are significant sources of quality animal proteins. Broiler fattening is very fast and the most affordable relative to other farm animals. On the other hand, poultry farming brings also pitfalls by producing harmful gases emitted into the air. Due to the severity of the problem, we observed ammonia, methane and nitrous oxide emissions from poultry farming. We used the established methodologies and we also accepted the country-specific conditions for poultry farming. First, we detected emission factors and then emissions of observed gases. We found that ammonia emission factors for laying hens, broilers, turkeys, ducks and geese were 0.45 kg, 0.20 kg, 0.89 kg, 0.65 kg and 0.31 kg per animal and year, respectively. The ammonia emissions for laying hens, broilers, turkeys, ducks and geese amounted 2,840 t, 1,115 t, 112.18 t, 112.76 t and 7.43 t per year, respectively. In case of methane, the emissions factors for laying hens and turkeys were 1.02 kg and 0.09 kg per animal and year, respectively. The methane emission factors for broilers, ducks and geese were the same and they reached 0.02 kg per animal and year. The methane emissions for laying hens, broilers, turkeys, ducks and geese achieved values 6,405.52 t, 111.07 t, 11.38 t, 3.48 t and 0.48 t per year, respectively. In case of nitrous oxide, the emission factors were the same for all poultry and they reached 0.001 kg per animal and year. The nitrous oxide emissions for laying hens, broilers, turkeys, ducks and geese achieved values 7.565 t, 3.142 t, 0.326 t, 0.345 t and 0.021 t per year. The highest ammonia, methane and nitrous oxide emissions were produced by laying hens and the lowest by geese. It was mainly related to the number of animals when the highest numbers were detected in laying hens and the lowest in geese. In 2016, we kept 12,130,501 poultry animals that produced 4,187.4 t of ammonia, 6,532 t of methane and 11.4 t of nitrous oxide emissions.

Keywords: Ammonia, Emission, Methane, Nitrous oxide, Poultry

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