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GOLD MICRO-PARTICLES FOR KNEE OSTEOARTHRITIS

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Background and aims:

Patients with knee OA (KOA) may display signs of low-grade inflammation.

Animal studies indicate gold ions have a long-acting effect on OA pain. The immuno-modulatory effect of gold ions has for more than 50 years a known anti-inflammatory effect in the treatment of rheumatic arthritis. Gold ions alter the function of macrophages by inhibiting lysosomal enzymes and lowering production of pro-inflammatory cytokines (1-2).

Dissolucytotic metallic gold (DMG) ions have an immune-suppressive effect in laboratory testing (3-6). Gold may decrease inflammation because of various mechanisms such as regulation of the NF-kB (nuclear factor kappa-light-chain-enhancer of activated B cells) pathway (3-6). (Figure 1 - 4). Animal studies prove the effect of gold implantation in arthritic joints (7-10). Injection of DMG in animal models stimulate the immune system (11-12). The carrier for injecting the DMG micro particles is hyaluronic acid (13-14).

No human studies have investigated the effect of intraarticular gold micro particle implants for the treatment of pain and inflammation in KOA. The present open, exploratory study investigated whether gold ions can act as a KOA treatment option through modulation of inflammatory mediators, pain sensitivity, and central pain mechanisms (15).

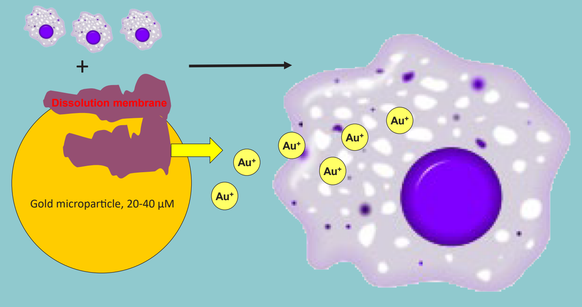


Figure 1. *Macrophages controls the dissolution membrane which liberate the gold ions by oxidation of the surface. Once the ions are liberated, most likely as Au(CN) , they are free to diffuse through the immediate microenvironment. The gold-loaded molecules are taken up into the cells, primarily macrophages, mastcells and histocytes.*

Figure 2. *Close to the gold implant gold-loaded molecular clusters are located outside cells. The two loaded cells are believed to be macrophages loaded with gold ions. The gold ions accumulate primarily in the lysosymes (3).*

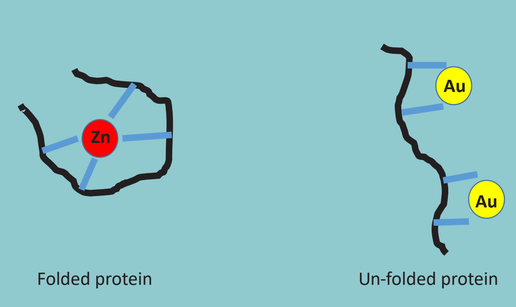


Figure 3. *Once in the intercellular fluid and the intracellular compartments, the gold ions act in the same ways that have been demonstrated for systemically administered gold ions. The effect is related to the ability of the gold ions to unfold the protein structures.*

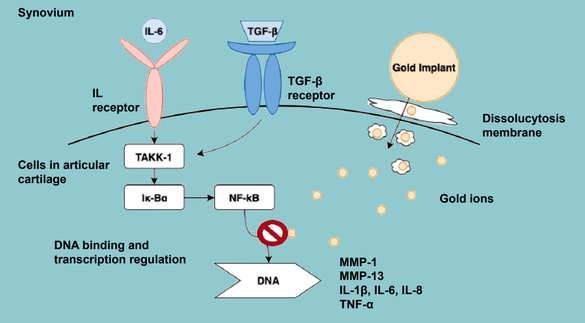


Figure 4. *Gold ions suppress inflammation locally by affecting certain signalling molecules and binding enzymes essential for the inflammatory process. The DNA binding activity and transcription regulation of NF-kB is abolished when AU- ions replace Zn2+ ions. (4)*

Genes	Protein Descriptions	Function	% Change	Ratio	p-value
GOLM1	Golgi membrane protein 1	Immune and Infl..	1907.5	20.07	6.99E-06
TYMP	Thymidine phosphorylase	Regenerative	186.1	2.86	0.012243
POC1A	POC1 centriolar protein homolog A	Regenerative	116.5	2.16	7.01E-06
APCS	Serum amyloid P-component	Immune response	102.2	2.02	5.68E-08
ARHGDIA	Rho GDP-dissociation inhibitor 1	Regenerative	91.5	1.91	0.011445
ANXA5	Annexin A5	Unknown	72.1	1.72	0.000575
DEFA1;DEFA3	Neutrophil defensin 1; Neutrophil defensin 3	Immune response	69.2	1.69	0.003064
CHAD	Chondroadherin	Regenerative	60.0	1.59	0.000362
THBS1	Thrombospondin-1	Immune response	57.0	1.57	0.002468
RGSL1	Regulator of G-protein signaling protein-like	Unknown	53.4	1.53	0.003183
IGHV3-7	Immunoglobulin heavy variable 3-7	Immune response	51.0	1.50	5.66E-06
FCN3	Ficolin-3	Immune response	-33.3	0.66	1.44E-05
LTA4H	Leukotriene A-4 hydrolase	Immune and infl.	-33.7	0.66	0.015647
SBSN	Suprabasin	Unknown	-34.7	0.65	7.16E-07
IGSF22	Immunoglobulin superfamily member 22	Unknown	-35.2	0.64	0.013466
CDC42	Cell division control protein 42 homolog	Immune response	-35.8	0.64	0.002768
NUTF2	Nuclear transport factor 2	Unknown	-39.2	0.60	0.010354
NaN	Immunoglobulin epsilon heavy chain	Sensory percept.	-40.5	0.59	2.16E-05
KRT10	Keratin, type I cytoskeletal 10	Immune and infl.	-43.8	0.56	1.35E-05
CSPG4	Chondroitin sulfate proteoglycan 4	Unknown	-46.9	0.53	0.015744
MPO	Myeloperoxidase	Immune and infl.	-47.3	0.52	0.014761
KRT2	Keratin, type II cytoskeletal 2 epidermal	Immune and infl.	-48.0	0.52	2.14E-05
PGLYRP1	Peptidoglycan recognition protein 1	Immune and infl.	-49.5	0.50	0.004872
FABP5	Fatty acid-binding protein 5	Immune and infl.	-49.5	0.50	0.002271
PNP	Purine nucleoside phosphorylase	Immune and infl.	-49.7	0.50	0.001185
MPP1	55 kDa erythrocyte membrane protein	Inflammatory	-50.0	0.50	0.008299
GPX1	Glutathione peroxidase 1	Immune response	-50.5	0.49	0.014773
GOT1	Aspartate aminotransferase, cytoplasmic	Unknown	-52.6	0.47	0.000849
LCN1	Lipocalin-1	Unknown	-55.6	0.44	0.00013
PIP	Prolactin-inducible protein	Unknown	-56.7	0.43	2.27E-06
KRT14	Keratin, type I cytoskeletal 14	Immune and infl.	-58.0	0.42	1.32E-05
KRT9	Keratin, type I cytoskeletal 9	Immune and infl.	-58.4	0.41	0.000196
KRT1	Keratin, type II cytoskeletal 1	Immune and infl.	-58.5	0.41	3.71E-10
RAC2; RAC1	Ras-related C3 botulinum toxin substrate 2;1	Inflammatory	-60.0	0.40	0.000154
PSMF1	Proteasome inhibitor PI31 subunit	Immune and infl.	-62.2	0.38	0.004097
SERPINB3	Serpin B3	Immune and infl.	-62.5	0.37	4.56E-06
DCD	Dermcidin	Immune response	-62.7	0.37	1.13E-08
KRT6B	Keratin, type II cytoskeletal 6B	Immune and infl.	-63.7	0.36	6.08E-09
SPTA1	Spectrin alpha chain, erythrocytic 1	Unknown	-64.0	0.36	0.001331
CALML5	Calmodulin-like protein 5	Immune and infl.	-67.9	0.32	0.000535
SPTB	Spectrin beta chain, erythrocytic	Unknown	-68.3	0.31	0.013697
PSMC3	26S proteasome regulatory subunit 6A	Immune and infl.	-75.2	0.24	0.01387
CASP14	Caspase-14	Unknown	-76.8	0.23	2.54E-06
EPB42	Erythrocyte membrane protein band 4.2	Unknown	-81.2	0.18	0.014775
ELANE	Neutrophil elastase	Inflammatory	-81.7	0.18	0.003667
MMRN1	Multimerin-1	Unknown	-86.1	0.13	4.79E-08
CTSG	Cathepsin G	Immune and infl.	-87.7	0.12	0.003394
PSMA5	Proteasome subunit alpha type 5	Immune and infl.	-88.1	0.11	0.003873
IGM	Immunoglobulin mu heavy chain	Immune response	-88.1	0.11	1.66E-08

Table 1. *The 49 significantly regulated proteins found in synovial fluid of knee OA patients, 38 downregulated and 11 upregulated, 8 weeks after intra-articular injection of 20 mg gold in 30 knee OA patients. Proteomic analysis using DIA-PASEF analysis of 500ng SF by label free quantification. Functional association of significantly regulated proteins in SF were assessed by STRING analysis.*

Genes	Protein Descriptions	Function	% Change	Ratio	p-value
IGHV1-58	Immunoglobulin heavy variable 1-58	Unknown	94.8	1.94	0.007386875
IGLV3-1	Immunoglobulin lambda variable 3-1	Unknown	-33.8	0.66	0.000235537
IGF2	Insulin-like growth factor II	Unknown	-34.8	0.65	0.001006413
HLA-H	Putative HLA class I histocompatibility antigen, alpha chain H	Unknown	-34.9	0.65	2.27E-07
IL1RAP	Interleukin-1 receptor accessory protein	Immune and infl.	-35.4	0.64	3.22E-06
P4HB	Protein disulfide-isomerase	Inflammatory	-36.3	0.63	0.001831153
IGKJ1	Immunoglobulin kappa joining 1	Immune response	-36.6	0.63	3.28E-06
NaN	Immunoglobulin delta heavy chain	Sensory percept.	-36.9	0.63	0.00185865
YWHAQ	14-3-3 protein theta	Unknown	-38.1	0.61	0.000489174
TUBA1B	Tubulin alpha-1B	Immune and infl.	-38.1	0.61	0.014099739
IGHV1-8	Immunoglobulin heavy variable 1-8	Unknown	-38.2	0.61	0.01411
UBB	Polyubiquitin-B	Immune and infl.	-38.8	0.61	0.019002538
IGKV2-40	Immunoglobulin kappa variable 2-40	Unknown	-40.1	0.59	3.06E-05
IGLV1-36	Immunoglobulin lambda variable 1-36	Unknown	-40.4	0.59	8.19E-07
LCN2	Neutrophil gelatinase-associated lipocalin	Immune and infl.	-40.8	0.59	1.11E-05
GDI2	Rab GDP dissociation inhibitor beta	Immune response	-41.0	0.59	1.69E-07
HSP90AA1	Heat shock protein HSP 90-alpha	Immune and infl.	-41.5	0.58	3.07E-07
KRT17	Keratin, type I cytoskeletal 17	Immune response	-42.5	0.57	0.003159425
ENO1	Alpha-enolase	Immune and infl.	-44.4	0.55	0.010242481
KRT6A	Keratin, type II cytoskeletal 6A	Immune response	-44.6	0.55	1.29E-07
ACTB	Actin, cytoplasmic 1	Immune response	-47.3	0.52	7.51E-06
IGKV2-29	Immunoglobulin kappa variable 2-29	Unknown	-48.2	0.51	7.23E-09
IGHV3-23	Immunoglobulin heavy variable 3-23	Unknown	-49.4	0.50	1.41E-07
HNRNPA1	Heterogeneous nuclear ribonucleoprotein A1	Immune and infl.	-50.4	0.49	0.005957151
HNRNPC	Heterogeneous nuclear ribonucleoproteins C	Immune and infl.	-55.5	0.44	0.009134014
HIST1H2AB	Histone H2A	Immune and infl.	-56.1	0.43	0.001243906
H4C1	Histone H4	Unknown	-61.9	0.38	0.007168713
IGLV2-8	Immunoglobulin lambda variable 2-8	Unknown	-66.2	0.33	3.21E-10
HIST1H2BK	Histone H2B	Immune and infl.	-67.1	0.32	3.96E-07
PTMA	Prothymosin alpha	Immune reponse	-67.3	0.32	0.010697326
S100A11	Protein S100-A11	Immune and infl.	-67.4	0.32	0.00122428
IGKV6D-21	Immunoglobulin kappa variable 6D-21	Immune response	-73.5	0.26	2.15E-06

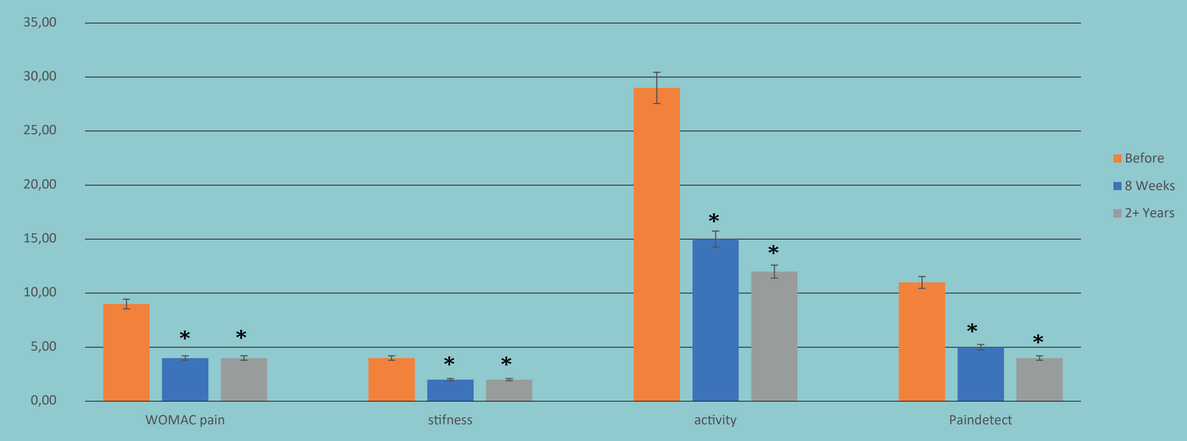
Table 2. *The 32 significantly regulated proteins found in serum of knee OA patients, 31 downregulated and 1 upregulated, 8 weeks after intra-articular injection of 20 mg gold in 30 knee OA patients. Proteomic analysis using DIA-PASEF analysis of 500 ng serum by label free quantification. Functional association of significantly regulated proteins in serum were assessed by STRING analysis.*

Methods:

Thirty patients with moderate KOA were included. Intraarticular injections with 20 mg gold microparticles (72.000 particles, 20-40 µm in diameter) using the patient's synovial fluid (SF) as carrier were performed. Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) subscores for pain, stiffness, and function were assessed at inclusion, 8 weeks and 2 years. The PainDetect questionnaire, pain pressure threshold (PPT), temporal summation (TS), and conditioned pain modulation (CPM), and pain diary were assessed at inclusion and 8 weeks. Proteome analysis was performed on SF and blood samples before and after 8 weeks of treatment.

Results:

At 8 weeks and 2 years follow-up compared to baseline there was a decrease in WOMAC scores and PainDetect ($P < 0.05$) (figure 5). In SF, 38 different proteins were downregulated and 11 upregulated ($P < 0.05$) mainly associated immune response (Table 1). Similarly, 31 proteins were downregulated and 1 upregulated in serum ($P < 0.05$) reflecting key immune response and anatomical structure development processes (Table 2). No adverse effects related to the treatment were recorded.



Figur 5. *WOMAC pain, stiffness and activity, and PainDetect, before treatment, and 8 weeks and 3 years after intra-articular injection of 20 mg gold in 30 knee OA patients (Median and quartiles). * represents significance compared to before treatment.*

Conclusions:

Gold microparticles injected intra-articular in KOA joints may provide pain relief and an inflammatory modulatory effect based on proteome changes found in SF and serum.

References

1. Persellin RH. The effect of gold salt on lysosomal enzymes of the peritoneal macrophage. Arthritis Rheum 1966;9:57-65.
2. Yanni G. Intramuscular gold decreases cytokine expression and macrophage numbers in the rheumatoid synovial membrane. Ann Rheum Dis 1994;53:315-22.
3. Danscher G. In vivo liberation of gold ions from gold implants.. Histochem Cell Biol 2002; 117:447-52.
4. Yang JP, Merin JP, Nakano T, Kato T, Kitade Y, Okamoto T. Inhibition of the DNA-binding activity of NF-kappa B by gold compounds in vitro. FEBS Lett. 1995 Mar 13;361(1):89-96.
5. Larsen A. Gold ions bio-released from metallic gold particles reduce inflammation and apoptosis and increase the regenerative responses in focal brain injury. Histochem Cell Biol 2008;130:681-92.
6. Seifert O. Gene expression profiling of macrophages: implications for an immunosuppressive effect of dissolucytotic gold ions. J Inflam 2012;9:43.
7. Jaeger GT. Double-blind, placebo-controlled trial of the pain-relieving effects of the implantation of gold beads into dogs with hip dysplasia. Vet Rec 2006;158:722-6.
8. Jaeger GT. Two years follow-up study of the pain-relieving effect of gold bead implantation in dogs with hip-joint arthritis. Acta Vet Scand 2007;49:9.
9. Jaeger GT. Gold Bead Implantation in Acupoints for Coxofemoral Arthrosis in Dogs. Animals 2012;2:426-36..
10. Lie KI. Inflammatory response to therapeutic goldbead implantation in canine hip joint osteoarthritis. Vet Pathol 2011;48:1118-24.
11. Havarinasab S. Gold causes geneticallydetermined autoimmune and immunostimulatory responses in mice. Clin Exp Immunol 2007;150:179-88.
12. Hazer DB. Soft tissue response to the presence of polypropylene-G-poly(ethylene glycol) comb-type graft copolymers containing gold nanoparticles. J Biomed Biotechnol 2011;2011:956169.
13. Danscher G. In: Patent US, editor. Medicament and method of treatment of patients with heavy metals 2010. United States Patent Number US 7,655,261 B2.
14. Märki N. Safety of Intra-Articular Gold Microimplants in Horses. J Equine Vet Res 2018;60:59-66.
15. Rasmussen S. https://clinicaltrials.gov/ct2/show/NCT03389906

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