Aalborg Universitet

AALBORG UNIVERSITY
DENMARK

Drug induced T-wave abnormalities

beyond QTc

Graff, Claus

Publication date:
2010

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

[Link to publication from Aalborg University](#)

Citation for published version (APA):
Graff, C. (2010). *Drug induced T-wave abnormalities: beyond QTc*. Medical Informatics Group. Department of
Health Science and Technology. Aalborg University.

Drug Induced
T-wave Abnormalities: Beyond QTc

Dissertation for the Degree of
Doctor of Philosophy
by
Claus Graff

Aalborg University

Faculties of Engineering, Science and Medicine

Dept. of Health Science and Technology

MI - Medical Informatics Group

Acknowledgements

English Summary

A considerable number of drugs from different therapeutic classes have the ability to delay cardiac repolarization, an effect that is reflected on the electrocardiogram as prolongation of the QT interval and the appearance of abnormal T-waves. Drug-induced inhibition of specific cardiac potassium channels has been shown to be the underlying cellular mechanism for QT interval prolongation, changes in the morphology of the T-wave, and the development of life threatening cardiac arrhythmias. To date, drug-induced arrhythmia has consistently involved inhibition of the cardiac I_{Kr} potassium channel, which is encoded by the *KCNH2* (hERG) gene. hERG mutations are also known to provide the pathogenic substrate for inhibition of the I_{Kr} channel in the congenital type 2 form of the long QT syndrome (LQT2). Patients with LQT2 are at risk for arrhythmia and sudden death, with about half of them sustaining major arrhythmic events.

Proarrhythmic liability of drugs is one of the most common reasons for labeling restrictions and withdrawals of drugs from the market today. As a consequence, assessing the potential for a new drug to cause life-threatening arrhythmias is now an integral component of premarketing safety assessment. The corner stone of this assessment is a clinical phase I trial, the "Thorough QT/QTc study", intended to identify drugs that have a pharmacologic effect on cardiac repolarization. This study calls for careful evaluation of drug effects on the QT interval to exclude proarrhythmic risk associated with a drug.

But there is a problem...QT interval prolongation is an inadequate marker of abnormal repolarization and a poor predictor of drug-induced arrhythmias.

The purpose of this thesis has, therefore, been to advance the presently inadequate characterization of drug-induced abnormal repolarization under the conjecture of a central role of hERG encoded I_{Kr} channel inhibition as the repolarization altering mechanism that can lead to proarrhythmia. Phenotypical ECG characteristics of the congenital LQT2 syndrome were used as computerized measures of T-wave morphology to indicate drug-induced repolarization abnormalities.

Four different I_{Kr} inhibiting drugs with different proarrhythmic potential were studied and their effects on T-wave morphology were compared to the effects on the QT interval. It was shown that there is a close relationship between drug influence and the development of specific LQT2-like T-wave morphology, which is consistent with the underlying cellular electrophysiology of I_{Kr} inhibition caused by the various drugs investigated in this work. Drug effects on T-wave morphology could be several times greater than the effects on the QT interval and this difference was positively related to the supposed proarrhythmic potential of the investigated drugs. It was further demonstrated that computerized analysis of LQT2-like T-wave morphology, may provide an opportunity to identify and explore complex drug effects on cardiac repolarization that cannot be described by the QT interval alone. Finally, it was shown that the relationship between QT interval prolongation and a change in T-wave morphology is very different for drugs with different risk profiles. Collectively, this work offers new insights into the characterization of drug-induced repolarization abnormalities that may lead to improved prediction of a drug's proarrhythmic potential.

Dansk Resumé

Et betragteligt antal lægemidler fra forskellige terapeutiske klasser har den egenskab at de forsinker hjertets repolarisering, en effekt der reflekteres på elektrokardiogrammet som et forlænget QT interval og forekomsten af abnormale T-takker. Det er påvist at lægemiddel-inhibering af specifikke kaliumkanaler i hjertet er den cellulære mekanisme, der ligger til grund for QT interval forlængelse, ændringer i morfologien af T-bølgen og udviklingen af livstruende hjertearytmier. Lægemiddel-induceret hjertearytmi har til dato konsekvent involveret inhibering af hjertets I_{Kr} kanal, som *KCNH2* (hERG) genet koder for. hERG mutationer er også kendt for at være det patogene grundlag for inhibering af I_{Kr} kanalen i den medfødte type 2 form af langt QT syndrom (LQT2). Patienter med LQT2 er i fare for hjerteartymi og pludselig død og omkring halvdelen af dem oplever betydelige hjertearytmier.

Proarytmisk risiko for lægemidler er en af de mest almindelige årsager til restriktioner på labels og tilbagetrækninger af lægemidler fra markedet i dag. Følgelig er vurdering af et nyt lægemiddels evne til at forårsage livstruende hjertearytmier blevet en integreret del af premarketings sikkerhedsvurdering. Hjørnestenen i denne vurdering, er et klinisk fase I studie, "Thorough QT/QTc studiet" som har til hensigt at identificere lægemidler der har en farmakologisk effekt på hjertets repolarisering. Dette studie opfordrer til grundig evaluering af medicinpåvirkninger af QT intervallet, for at kunne ekskludere proarytmisk riskio forbundet med et lægemiddel.

Men der er et problem…QT interval forlængelse er en utilstrækkelig markør for abnorm repolarisering og er dårlig til at forudsige om et lægemiddel vil inducere arytmier.

Formålet med denne afhandling har derfor været, at avancere den nuværende utilstrækkelige karakterisering af lægemiddel-induceret abnorm repolarisering, under formodning om en central rolle for hERG kodet I_{Kr} kanal inhibering som den repolariserings-forandrende mekanisme, der kan føre til hjertearytmi. Fænotypiske EKG kendetegn for medfødt LQT2 syndrom blev anvendt som computeriserede mål for T-bølge morfologi til at indikere lægemiddel-inducerede repolariserings abnormaliteter.

Fire forskellige I_{Kr} inhiberende lægemidler med forskellig proarytmisk potentiale blev studeret og deres effekter på T-bølge morfologi blev sammenlignet med deres effekter på QT intervallet. Det blev vist, at der var en tæt sammenhæng mellem lægemiddelpåvirkning og udviklingen af specifik LQT2-agtig T-bølge morpfologi som er forenelig med den cellulære elektrofysiologi der ligger til grund for I_{Kr} inhibering forårsaget af de forskellige lægemidler der blev undersøgt i dette arbejde. Medicinpåvirkning af T-bølge morfologi kunne være adskellige gange større end påvirkninger på QT intervallet og denne forskel var positivt relateret til det formodede proarytmiske potentiale for de undersøgte lægemidler. Det blev desuden demonstreret at computeriseret analyse af LQT2-agtig T-bølge morfologi, kan gøre det muligt at identificere og opdage komplekse medicinpåvirkninger af hjertets repolarisering, som ikke kan beskrives med QT intervallet alene. Slutteligt blev det vist, at sammenhængen mellem QT interval forlængelse og en ændring af T-bølge morfologi, er meget forskellig for lægemidler med forskellige risikoprofiler. Samlet set giver dette arbejde nye indblik i karakteriseringen af lægemiddel-inducerede abnormaliteter der kan føre til bedre forudsigelse af et lægemiddels proarytmiske potentiale

List of Papers

This thesis is based on the following papers, which are referred to in the text by their Roman numerals:

I   **Graff C**, Andersen MP, Xue JQ, Hardahl TB, Kanters JK, Toft E, Christiansen M, Jensen HK, Struijk JJ. Identifying Drug-Induced Repolarization Abnormalities from Distinct ECG patterns in Congenital Long QT Syndrome: A study of d,l-sotalol effects on T-wave morphology.
Drug Saf. 2009; 32: 599-611.

II   Nielsen J, **Graff C**, Hardahl TB, Andersen MP, Kristoffersen J, Struijk JJ, Toft E, Meyer JM. Sertindole causes distinct electrocardiographic T-wave morphology changes.
European Neuropsychopharmacology 2009; 19: 702-707.

III   **Graff C**, Matz J, Christensen EB, Andersen MP, Kanters JK, Toft E, Pehrson S, Hardahl TB, Nielsen J, Struijk JJ. Quantitative Analysis of T-wave Morphology Increases Confidence in Drug-induced Cardiac Repolarization Abnormalities: Evidence from the investigational I_{Kr} inhibitor – Lu 35-138.
J. Clin. Pharmacol. 2009; 49: 1331-1342.

IV   **Graff C**, Struijk JJ, Matz J, Kanters JK, Andersen MP, Nielsen J, Toft E. Covariate Analysis of QTc and T-wave Morphology: New Possibilities in the Evaluation of Drugs Affecting Cardiac Repolarization.
Clin. Pharmacol Ther. 2010; 88: 88-94.

Paper I investigates electrocardiographic manifestations of repolarization abnormalities induced by the torsadogenic drug d,l-sotalol to determine whether a composite ECG measure based on typical LQT2 patterns of T-wave morphology can provide an effective description of sotalol induced repolarization changes that are commonly seen in patients taking this drug.

Paper II examines the relative merits of assessing quantitative T-wave morphology in addition to changes in the QT interval duration in schizophrenia patients exposed to sertindole, a drug which has a possible but uncertain association with the development of Torsade de Pointes arrhythmia.

Paper III analyzes repolarization changes induced by a new antipsychotic that was terminated during development to assess whether a composite electrocardiographic measure of T-wave morphology could be used as a reliable marker to support the evidence of abnormal repolarization, which was indicated by QT interval prolongation.

Paper IV investigates the association between drug-induced T-wave morphology and QTc prolongation for moxifloxacin which has a safe cardiovascular profile and for the torsadogenic drug d,l-sotalol. This study investigates whether covariate analysis of these repolarization measures might add information to drug safety analysis by QT interval assessment alone.

Contents

INTRODUCTION

INTRODUCTION

THE CLINICAL PROBLEM

More than one hundred years after Einthoven recorded his first electrocardiogram (ECG), cardiac repolarization is not completely understood. Clinicians and researchers keep debating the cellular basis of repolarization and are still learning how to extract information about the repolarization process from the ECG.

One of the parameters in the surface ECG that describes repolarization is the QT interval. The QT interval and changes in the QT interval may seem to be related to cellular processes in a simple way: the action potential duration (APD) defines the QT interval and QT prolongation, therefore, is an indicator of increased APD. However, the truth is more complex. Myocardial cells from different parts of the heart, even from different layers of the muscular wall have different APDs and may respond differently to external factors, such as drugs, making the repolarization process more heterogeneous. Adding to the complexity is the coupling of the cells in situ, which also has a great, but poorly quantified, influence on their relative APDs.

QT prolongation has attracted a great deal of attention because of its apparent association with life-threatening cardiac arrhythmias such as Torsades de Pointes (TdP).[1] This arrhythmia refers to a rapid and irregular polymorphic ventricular tachycardia that was first described by Dessertenne in 1966.[2] It is characterized by an electrocardiographic pattern of continuously changing morphology of the QRS complexes that seem to twist around an imaginary baseline. Torsade is commonly non-sustained but can lead to syncope and may degenerate into ventricular fibrillation and sudden death. The likelihood of TdP is increased in a milieu of heterogeneous ventricular repolarization[1,3] and the arrhythmia usually occurs in the setting of T-wave abnormalities, increased U-wave amplitudes and an excessively prolonged heart rate corrected QT interval (QTc).[4,5]

A prolonged QT interval and the concomitant risk for TdP is the hallmark of the various congenital long QT syndromes but may also be induced by several drugs. To date, drug-induced TdP has consistently involved inhibition of the cardiac I_{Kr} channel.[6] The APD prolongation in this case is caused by a reduced rate of potassium ions through the I_{Kr} channels, which are encoded by the *KCNH2* (hERG) gene.[7] It was found[8] that I_{Kr} inhibition is involved in one of the most common forms of congenital long QT syndrome (LQTS) – type 2 and affected by a large number of cardiac and non-cardiac drugs as well.[6]

QT interval prolongation and the proarrhythmic liability of drugs has become one of the most common reasons for labeling restrictions and withdrawals of drugs from the market.[9,10] The first drug to be withdrawn from the market due to cases of arrhythmia was prenylamine in 1988. Many more withdrawals, restrictions, and denied market authorizations have since followed for a range of drugs that belong to different pharmacological classes including the drugs terodiline, astemizole, cisapride, lidoflazine, droperidol, levomethadyl, grepafloxacin, mesoridazine, sparfloxacin, terfenadine, thioridazine, and sertindole. In recent decades, the US Food and Drug Administration (FDA) has expanded its requirements for drug testing. Today, a new drug requires an average of 15 years in research and development to reach the market and the cost was estimated to be as high as 1.7 billion US dollars.[11] Still, only 1 in 10 drugs that enter clinical testing are approved by the FDA and the failure rate for drugs in phase III has increased.[12] This poor state of drug development was addressed in a white paper known as the Critical Path Initiative.[13] The report concluded that the major contributor to the inefficiency in development of drugs was the absence of innovative new methods for preclinical and clinical testing of drugs:

"Not enough applied scientific work has been done to create new tools to get fundamentally better answers about how the safety and effectiveness of new products can be demonstrated, in faster time frames, with more certainty, and at lower costs. In many cases, developers have no choice but to use the tools and concepts of the last century to assess this century's candidates".

Such dilemmas pose obvious challenges for drug developers who are searching for ways to expedite the drug discovery and evaluation processes while at the same time developing safe drugs. As a consequence, it has now been emphasized by the FDA that efforts

must be directed towards the development of new biomarkers that can improve the prediction of cardiac toxicity.[14]

In addition to the Critical Path document, the FDA together with Health Canada released another paper in 2002 that provided the drug development industry with a preliminary guidance on the importance of ECG analysis for drug approval.[15] This concept paper was subsequently subjected to the International Committee on Harmonization (ICH), which culminated in 2005 with the acceptance of the E14 "Guidance for clinical evaluation of QT interval prolongation and proarrhythmic potential of non-antiarrhythmic drugs".[16] The corner stone of the guidance was the establishment of a clinical phase I trial, the "Thorough QT/QTc study", intended to confidently identify drugs that have a threshold pharmacologic effect on cardiac repolarization. This study is presently a required integral part of the development of practically every pharmaceutical compound.

THE THOROUGH QT STUDY -
REGULATORS' ANSWER TO THE PROBLEM

The goal of the Thorough QT study (TQTS) is often misunderstood. This study is not aimed at quantifying the risk of drug-induced TdP. Rather, the purpose of this study is to exclude proarrhythmic risk associated with a drug by establishing whether the investigated drug has any effect on cardiac repolarization, which is the potential basis of drug-induced TdP.

The primary end point in the TQTS is the drug-induced QTc change from baseline corrected for placebo effects. Regulatory guidance on how much QTc prolongation might be related to risk of TdP mainly reflects the regulators' experience with approving QTc prolonging drugs and observing their effect on the market over the past 20 years. The consensus of the relationship between QTc prolongation and risk is the following: 0-5 ms imparts no risk; if the QTc prolongation is greater than 20 ms the risk for TdP is considered elevated or quite high. It is further suggested that a 5-10 ms QTc prolongation is of minimal concern and that the risk associated with a 10-20 ms effect is uncertain.[16]

The TQTS also involves the reporting of electrocardiographic abnormalities of the morphology of the ECG. Specifically, such assessments should be described and presented in terms of the number and percentage of subjects in each treatment group who manifest the appearance or worsening of a morphological abnormality. Particular attention should be directed to the appearance of abnormal U-waves and changes in T-wave morphology that might be indicative of delayed repolarization such as double humps or notched T-waves, indistinct terminal TU complex, delayed inscription or prolonged ST segment, widening, flattening and inversion.[16]

The TQTS is carried out in healthy volunteers to provide indications of QTc prolongation in the target population of patients to be studied during later phases of drug development. The design can be parallel or cross-over depending on the half-lives of the parent compound or metabolites and should be randomized and placebo-controlled. In addition, an active control should be used to demonstrate that the study can discern the level of QT prolongation which is of regulatory concern. In the overwhelming majority of studies, a single 400 mg dose of moxifloxacin has been used because this drug has a relatively consistent and mild QTc prolonging effect and is considered to have a safe cardiovascular profile.

The study is typically performed after significant information is available on the pharmacokinetics of the compound. This is because the number and timing of ECG recordings taken after each dose of the study drug is driven mainly by the pharmacokinetic and pharmacodynamics properties of the drug. Ideally, the time points for ECG recordings should be chosen based on the expected time course of drug related QTc change from baseline so that recordings can be obtained at times around the expected peak plasma concentrations.

The sample size of the TQTS is defined by the requirement of having enough power to detect a 5 ms change from baseline because this level is considered the magnitude of QTc prolongation where risk begins. The study is expected to be able to discern this small degree of QTc prolongation with 80% power and a 0.05 level of significance. For the results of the TQT study to be most predictive all sources of variability must be controlled because a key determinant of sample size is the variance of QTc changes from baseline which in large part is determined by the number of subjects included in the study and to some

degree by the number of ECGs recorded per subject. The typical TQTS usually enrolls 40 to 60 subjects and include 10.000 to 15.000 ECG recordings. An outlier or categorical analysis in the TQTS is explorative because the study is not powered to accurately detect the number of subjects who respond in an exaggerated manner.

It is generally assumed that if a drug truly prolongs the QT interval it will be dose related. This is why the TQTS typically looks at the effect on QTc prolongation of both therapeutic and supratherapeutic doses of a drug. The study should reflect situations in which plasma levels may be markedly elevated in the target population and the supratherapeutic dose level is intended to achieve the highest exposure levels anticipated in clinical practice, e.g. exposure levels caused by drug-drug interactions, overdose, genetic polymorphisms affecting drug metabolism, or in patients with heart disease.

The outcome of a TQTS is classified as negative if the upper limit of the two-sided 90% confidence interval for the largest time-matched difference between the test drug and placebo is below 10 ms.[16] This level of QTc prolongation is chosen to provide reasonable assurance that the mean effect of the study drug on QTc prolongation is not greater than around 5 ms. If the upper limit of the two-sided 90% confidence interval for QTc prolongation exceeds 10 ms, then the test drug is declared positive. In other words, the drug is classified as having affected cardiac repolarization.

The outcome of the TQTS has severe implications for a drug development program. A negative study will almost always allow drug development to proceed without any dedicated focus on QT assessments. A positive study on the other hand would imply more complex additional and expensive monitoring of QTc effects, dose concentration responses and drug interactions in a substantial number of patients in the target population. Thus, a positive study is expected to result in a need to collect a significant number of ECGs during the later phases II and III of clinical testing. This extensive pre-market ECG testing and possibly additional post-market monitoring is extremely costly and may render the drug economically non-viable.

The thorough QT study has attained such a pivotal role in drug testing that it can mean the difference between continuation and termination of a drug development program. In light of the implications for drug developers of the outcome of the TQTS it is important that the study provides the correct guidance. As a whole, the pharmaceutical industry acknowledges the importance of the TQTS but drug developers are also rightfully skeptical of conclusions based on the measurement of the QT interval because this measurement by is increasingly regarded as a poor surrogate marker of the risk of proarrhythmic events.

Important design and measurement issues of the TQTS are continuously debated and it can be expected that changes will be made to the way these studies are conducted. At present it is likely however that the implementation of the TQTS will result in an increasing number of drugs with label statements with regard to QT prolongation based on TQTS outcomes that exceed the regulatory threshold. Such labeling is already in effect. The drug alfuzosin, which does not inhibit the I_{Kr} channel at anywhere near relevant concentrations, and which has not been associated with proarrhythmia after extensive post-marketing experience, but which produces a small QTc prolongation somewhat exceeding 10 ms in a TQT study[17], now bears a label with warnings of cardiac repolarization effects.

THE QT INTERVAL -
A BIOMARKER PROBLEM

The first collection of challenging problems faced by clinicians, regulators and drug developers is related to the task of measuring the QT interval with precision. The measurement problem has existed for more than five decades, since the QT interval was first measured using paper ECG recordings and remains largely unsolved today with the increasing use of digital recordings. Challenges relate to a number of factors, but the most frequently cited difficulties consist of delineating the end of the T-wave when this wave is flat, bifid, biphasic, broad, notched or overlapping with a U-wave. Indeed, measurement of the QT interval may lead to quite significant differences depending on the experience of the reader.[18]

The utilization of QT interval measurements may also be complicated in the presence of heart rate changes. When the heart rate decreases or increases, the QT interval prolongs or shortens, respectively, even

without any drug intervention. Because of this inverse relationship with heart rate, it is customary to obtain a heart rate corrected QT interval. However, despite numerous attempts to derive a universally acceptable formula for heart rate correction of the QT interval, the problem has not been resolved. The QT interval has been corrected for heart rate using a number of methods including fixed population formulas, data driven formulas derived from the population under investigation or individual correction methods.[19] Problems such as over or under correction are known to exist for fixed population methods and there is increasing evidence that the relationship between the QT interval and heart rate is subject-specific.[19] Unfortunately, the effective use of subject-specific correction methods can present important problems in clinical trials because subjects are at rest during the ECG recordings, usually with little variation in their heart rates. In such situations, an individually derived correction method, based on a narrow range of heart rates, may be incorrect. The data driven population based methods on the other hand, rely on the assumption that the relationship between the QT interval and heart rate is the same for all subjects and that a single model properly describes this relationship, which is incorrect.

Correction of the QT interval for heart rate has been the subject of intense debate for almost a century Needless to say, issues surrounding the choice of the most appropriate heart rate correction method are rather complex. Today, there are over 30 different formulas for heart rate correction of the QT interval[20] and all of them yield different QTc values. The search for refined heart rate correction methods is likely to continue although it has been argued that this technical discussion may have limited physiological meaning[21] and that further studies aiming at finding the "correct" heart rate formula are likely to be counterproductive.[22]

The second set of challenges is that, in terms of cardiovascular safety, the ability of QT interval measurements to predict TdP is unclear. Early work suggested a simple relationship between the degree of QT prolongation and proarrhythmia with a 5% increase in baseline risk of proarrhythmic events for every 10 ms increase in the QT interval.[23] Although attractive, this correlation is now known to be an oversimplification of a much more complex relationship between the QT interval and cardiovascular risk. There are numerous experimental and clinical studies showing compelling evidence that the QT interval is a poor surrogate marker of proarrhythmic susceptibility, and there are many shortcomings to the assumption of a simple QT-proarrhythmia relationship.[2,24-26] Different drugs causing comparable absolute or relative increases in the QT interval do not result in similar TdP incidences (e.g. sotalol versus dofetilide or amiodarone versus almokalant). In serial experiments with different doses of the same drug, proarrhythmia has been shown not to depend on the degree of QT interval prolongation.[27,28] Altering drug infusion rates can change the incidence of TdP without affecting the QT interval.[29,30] Sotalol can induce arrhythmia before any prolongation of action potentials is observed in cardiac cells.[31] It is generally estimated that about 10-20% of otherwise healthy individuals have borderline QTc values beyond the so called normal range[32] and among the congenital syndromes, associated with abnormal repolarization, the short QT syndrome poses the greatest risk.

In light of such obvious limitations in characterizing abnormal repolarization, it is difficult to understand why this measurement would be regarded an indispensible and essential electrocardiographic biomarker for drug safety assessment. Nevertheless, presently there are no other established methods. There is little doubt however, that further understanding of the cellular and molecular mechanisms underlying repolarization will enable us to develop better indices of repolarization abnormalities and that such biomarkers will improve prediction of drug-induced proarrhythmic potential.

T-WAVE MORPHOLOGY - ADRESSING THE QT BIOMARKER PROBLEM

Much of our current knowledge about drug-induced ion channel defects has been influenced by our knowledge of the congenital long QT syndrome. There are several types of the congenital long QT syndrome, including type 2 (LQT2) which is caused by loss-of-function mutations of hERG[8] and is the second most prevalent form of this syndrome. The human ether related a go-go gene (hERG) encodes the I_{Kr} channel responsible for the repolarizing current in

the cardiac action potential and patients with LQT2 are at risk for TdP and sudden death[33], with about half of them sustaining major arrhythmic events.

Observations from ECG analysis in patients with congenital long QT syndrome have revealed that specific genetic forms are associated with distinctive T-wave morphologies.[34-37] Of particular interest here, are the patterns described for LQT2, which include flat, bifid, notched and low amplitude T-waves.

Interestingly, the hERG channel appears to be the most pharmacologically promiscuous among all known voltage-gated potassium channels, with drugs from a wide range of therapeutic categories capable of inhibiting the channel.[38,39] Hereby, the unique position of hERG and the I_{Kr} channel is highlighted as the common denominator[40] in congenital and drug-induced long QT syndrome. It is therefore not surprising that electrocardiographic abnormalities similar to those seen in patients carrying mutations in the hERG gene can be produced via direct blockade of hERG/I_{Kr} channels by a large group of diverse therapeutic compounds including many antiarrhythmics, antihistamines, antipsychotics and antibiotics. In fact, profound T-wave morphology changes have been seen with drugs that inhibit the I_{Kr} channel and the severity of these changes has been shown to be more pronounced in patients who develop TdP.[41]

The electrophysiological effect of I_{Kr} inhibition has been studied both in experimental systems, where all cell types are represented[42,43], and using computer models in which the effects of individual action potential currents can be simulated.[44] These studies have demonstrated that there are significant differences in repolarization in the various layers of the myocardium, with the epicardial cells having the shortest action potential duration, endocardial cells having an intermediate duration, and M cells having the longest action potential duration. This physiological transmural dispersion of repolarization usually does not lead to TdP. However, in both the in vitro and in silico work, I_{Kr} inhibition prolongs the action potential of M cells substantially, while endocardial and epicardial action potentials show much smaller changes. Such I_{Kr} related differential effects can give rise to exaggerated transmural dispersion which in turn increases the susceptibility to

reentry, a mechanism presumed to underlie TdP. Physiologically, there is a substantial degree of spatial dispersion in repolarization, not only across the myocardial wall but also between the apex and base of ventricles, between the septum and free walls, as well as between the left and right ventricles. It is not known whether heterogeneity between apex and base or between left and right ventricles are similar to those observed for transmural dispersion, although it seems likely due to the different distribution of repolarization channels in cells of these regions.

Prolongation of the QT interval can result from both homogeneous and heterogeneous prolongation of the predominant myocardial cell types. It is therefore possible that electrocardiographic assessment of drugs using QT interval as the only marker of repolarization, could overlook the effect of dispersion which appears to be a necessary substrate for inducing TdP. In fact, this may be one explanation for the lack of a clear correlation between QT interval prolongation and proarrhythmia. In contrast, T-wave patterns such as those seen in LQT2 are thought to result from a heterogeneous repolarization of the predominant myocardial cell types, which is claimed to be associated with more proarrhythmic potential than homogeneous repolarization.

Drug-induced I_{Kr} inhibition can also cause triangulation of the action potential with loss of notch-and-dome morphology replaced by a linear phase repolarization. Triangulation is reported to be a strong predictor of proarrhythmia and to appear on the ECG as a widened, flattened or notched T-wave.[45] Interestingly, triangulation may be accompanied by either shortening or lengthening of the cardiac action potential duration which indicates that drug-induced T-wave morphology changes may be independent of QT interval measurements.

In conclusion, drug-induced myocardial heterogeneity may manifest, not only as differences in action potential durations, but also as changes of action potential shapes. Importantly, both cellular phenomena translate directly into characteristic changes of T-wave morphology that are distinctive of congenital LQT2. It appears that there are many components to cardiac repolarization beyond just its duration and that there is information to be gained about cardiac repolarization from the analysis of T-

wave morphology changes in drug studies, which goes beyond the information that can be obtained from measurements of the QT interval. For this reason it would be important to quantify T-wave manifestation of the cellular mechanisms in drug studies instead of merely estimating the end of the T-wave to measure the QT interval.

REFERENCES

[1]   Fenichel RR, Malik M, Antzelevich C, et al. Drug-induced torsades de pointes and implications for drug development. *J. Cardiovasc. Electrophysiol.* 2004;15:475-495.

[2]   Dessertenne F. La tachycardie ventriculaire a deux foyers opposes variables. *Arch. Mal. Coeur.* 1966;59:263-272.

[3]   Belardinelli L. Antzelevith C, Vos MA, et al. Assessing predictors of drug-induced torsade de pointes. *Trends Pharmacol. Sci.* 2003;24:619-625.

[4]   Kirchhof P, Franz MR, Abdennasser B, Wilde AM. Giant T_U waves preced torsades de pointes in long QT syndrome. *J. Am. Coll. Cardiol.* 2009;54:143-149.

[5]   Yan GX, Lankipalli RS, Burke JF et al. Ventricular repolarization components on the electrocardiogram: cellular basis and clinical significance. J. Am. Coll. Cardiol. 2003;42:401-409.

[6]   Haverkamp W, Breithart G, Camm AJ, et al. The potential for QT prolongation and proarrhythmia by nonantiarrhythmic drugs: clinical and regulatory implications. *Eur. Heart J.* 2000;21:1216-1230.

[7]   Sanguinetti MC, Curran ME, Spector PS, et al. Spectrum of HERG K+-channel dysfunction in an inherited cardiac arrhythmia. *Proc. Natl. Acad. Sci. USA* 1996;93:2208-2212.

[8]   Keating MT, Sanguinetti MC. Molecular and cellular mechanisms of cardiac arrhythmias. *Cell* 2001;104:569-580.

[9]   Lasser KE, Allen PD, Woolhandler SJ, et al. Timing of new black box warnings and withdrawals for prescription medications. *JAMA* 2002;287:2215-2220.

[10]   Roden DM. Drug induced prolongation of the QT interval. *N. Engl. J. Med.* 2004;350:1013-1022.

[11]   DiMasi JA, Hansen RW, Grabowski HG. The price of innovation: new estimates of drug development costs. *J. Health. Econ.* 2003;22:151–185.

[12]   Mervis J. Productivity counts – but the definition is key. *Science* 2005;309:726.

[13]   FDA, Challenge and Opportunity on the Critical Path to New Medicinal Products March 2004 http://www.fda.gov/ScienceResearch/SpecialTopics/CriticalPathInitiative/default.htm [Accessed January 2010].

[14]   FDA, Critlical Path opportunities list March 2006http://www.fda.gov/ScienceResearch/SpecialTopics/CriticalPathInitiative/default.htm [Accessed January 2010].

[15]   Food and Drug Administration and Health Canada. The clinical evaluation of QT/QTc interval prolongation and proarrhythmic potential for non-antiarrhythmic drugs. Preliminary Concept paper. November 2002.

[16]   Guidance for industry: E14 clinical evaluation of QT/QTc interval prolongation and proarrhythmic potential for non-antiarrhythmic drugs2005http://www.fda.gov/RegulatoryInformation/Guidances/ucm129335.htm [Accessed January 2010].

[17]   FDA review for Uroxatral (alfuzocin HCL tablets).http://www.fda.gov/ohrms/dockets/ac/03/briefing/3956B1_01_FDA-alfuzosin.pdf. [Accessed January 2010].

[18]   Viskin S, Rosovski U, Sands AJ. Inaccurate electrocardiographic interpretation of long QT: The majority of physicians cannot recognize a long QT when they see one. *Heart Rhythm* 2005;2:569-574.

[19]   Malik M. The imprecision in heart rate correction may lead to artificial observations of drug induced QT interval changes. *Pacing. Clin. Electrophysiol.* 2002;25:209–216.

[20]   Malik M. Problems of heart rate correction in assessment of drug-induced QT interval prolongation. *J. Cardiovasc. Electrophysiol.* 2001;12:411-420.

[21]   Boyle NG, Weiss JN. Making QT correction simple is complicated. *J. Cardiovasc. Electrophysiol.* 2001;12:421–423.

[22] Malik M, Färbom P, Batchvarov V, et al. Relation between QT and RR intervals is highly individual among healthy subjects: implications for heart rate correction of the QT interval *Heart* 2002;87:220-228.

[23] Moss, AJ, Schwatz PJ, Crampton RS, et al. The long QT syndrome. Prospective longitudinal study of 328 families. *Circulation*. 1991;84:1136-1144.

[24] Hondeghem LM, Carlsson L, Duker G. Instability and triangulation of the action potential predict serious proarrhythmia, but action potential duration prolongation is antiarrhythmic. *Circulation* 2001;103:2004–2013.

[25] Antzelevitch C. Arrhythmogenic mechanisms of QT prolonging drugs: is QT prolongation really the problem? *J. Electrocardiol.* 2004;37(Suppl):15–24.

[26] Malik M. Detection of drug-induced proarrhythmia: balancing preclinical and clinical studies. *Heart Rhythm* 2005;2:773–776.

[27] Thomsen MB, Verduyn SC, Stengl M, et al. Increased short-term variability of repolarization predicts d-sotalol-induced torsades de pointes in dogs. *Circulation* 2004;110:2453-2459.

[28] Thomsen MB, Volders PG, Stengl M, et al. Electrophysiological safety of sertindole in dogs with normal and remodeled hearts. *J. Pharmacol. Exp. Ther.* 2003;307:776-784.

[29] Carlsson L, Abrahamsson C, Andersson B, et al. Proarrhythmic effects of the class III agent almokalant: importance of infusion rate, QT dispersion, and early afterdepolarisations. *Cardiovasc. Res.* 1993;27:2186-2193.

[30] Detre E, Thomsen MB, Beekman JD, et al. Decreasing the infusion rate reduces the proarrhythmic risk of NS-7: confirming the relevance of short-term variability of repolarisation in predicting drug-induced torsades de pointes. *Br. J. Pharmacol.* 2005;145:397-404.

[31] Hondeghem LM. Frequency dependence of class I and class III antiarrhythmic agents explaining their antiarrhythmic and proarrhythmic properties. In: Franz MR, editor. Monophasic action potentials: bridging cell and bedside. 1st ed. Armonk: Futura Publishing, 2000: 381-394.

[32] Johnson JN, Ackerman MJ. QTc: how long is too long? *Br. J. Sports Med.* 2009;43:657-662.

[33] Priori SG, Schwartz PJ, Napolitano C. Risk Stratification in the Long-QT Syndrome. *N. Engl. J. Med.* 2003;348:1866-1874.

[34] Zhang L, Timothy KW, Vincent GM, et al. Spectrum of ST-T wave patterns and repolarization parameters in congenital long QT syndrome: ECG findings identify genotypes. *Circulation* 2000;102:2849-2855.

[35] Lehmann MH, Suzuki F, Fromm BS, et al. T wave 'humps' as a potential electrocardiographic marker of the long-QT syndrome. *J. Am. Coll. Cardiol.* 1994;24:746-754.

[36] Malfatto G, Beria G, Sala S, *et al.* Quantitative analysis of T wave abnormalities and their prognostic implications in the idiopathic long-QT syndrome. *J. Am. Coll. Cardiol.* 1994;23:296-301.

[37] Lupoglazoff JM, Denjoy I, Berthet M, et al. Notched T waves on Holter recordings enhance detection of patients with LQT2 (HERG) mutations. *Circulation* 2001;103: 1095-1101.

[38] De Ponti F, Poluzzi E, Montanaro N. QT-interval prolongation by noncardiac drugs: lessons to be learned from recent experience. *Eur. J. Clin. Pharmacol.* 2000;56**:**1-18.

[39] De Ponti F, Poluzzi E, and Montanaro N. Organising evidence on QT prolongation and occurrence of torsades de pointes with non-antiarrhythmic drugs: a call for consensus. *Eur. J. Clin. Pharmacol.* 2001;57**:**185-209.

[40] Sanguinetti MC, Jiang C, Curran ME, et al. A mechanistic link between an inherited and an acquired cardiac arrhythmia: HERG encodes the IKr potassium channel. *Cell* 1995;81:299-307.

[41] Houltz B, Darpo B, Edvardsson N, et al. Electrocardiographic and clinical predictors of torsades de pointes induced by almokalant infusion in patients with chronic atrial fibrillation or flutter: a prospective study. *Pacing. Clin. Electrophysiol.* 1998;21:1044-1057.

[42] Antzelevitch C. Heterogeneity of cellular repolarization in LQTS: the role of M cells. *Eur. Heart J. Suppl.* 2001;3(Suppl. K):K2-K16.

[43] Yan GX, Antzelevitch C. Cellular basis for the normal T wave and the electrocardiographic manifestations of the long-QT Syndrome. *Circulation* 1998;98:1928-1936.

[44] Gima K, Rudy Y. Ionic current basis of electrocardiographic waveforms: a model study. *Circ. Res.* 2002;90:889-896.

[45] Shah, RR., Hodeghem, LM. Refining detection of drug-induced proarrhythmia: QT interval and TRIaD. *Heart Rhythm*. 2005;2: 758-772.

PURPOSE OF THIS THESIS

The purpose of this thesis has been to advance the presently inadequate electrocardiographic characterization of drug-induced abnormal repolarization, under the conjecture of a central role of *KCNH2* (hERG) encoded I_{Kr} channel inhibition as the repolarization altering mechanism that can lead to proarrhythmia.

The principal ion channel affected by most proarrhythmic drugs is $I_{Kr,}$ which is the same channel affected in LQT2 patients with loss-of-function hERG mutations. Therefore, this work has attempted to determine whether phenotypical ECG observations in the congenital LQT2 syndrome could be used as computerized measures of T-wave morphology to indicate drug-induced repolarization abnormalities.

The degree of QT interval prolongation is currently considered the gold standard for assessment of abnormal repolarization. This work has therefore further aimed to discover whether a change of T-wave morphology represents a more pronounced manifestation of drug-induced repolarization abnormality than prolongation of the QT interval, because this property would be constructive to expanding electrocardiographic safety evaluation of drugs in clinical trials.

PUBLICATIONS

CONCLUSION

CONCLUSION

This study has investigated the possibility of advancing the presently inadequate electrocardiographic characterization of drug-induced repolarization abnormalities, using phenotypic ECG observations in the congenital LQT2 syndrome as computerized measures of T-wave morphology. This work has further aimed to discover whether a change of T-wave morphology represents a more pronounced manifestation of drug-induced repolarization abnormality than prolongation of the QT interval. Such a property would be constructive to expanding electrocardiographic safety evaluation of drugs in clinical trials.

Considerable evidence has been provided that a composite marker of asymmetry, flatness and notches, which represent typical T-wave patterns indicative of abnormal repolarization in the congenital LQT2 syndrome, could be used as a sensitive indicator of abnormal repolarization induced by drugs. It was shown that such T-wave morphology characteristics could be used to identify repolarization abnormalities due to a variety of drugs belonging to different therapeutic classes, and also across a broad spectrum of drug-induced shape changes, from subtle to exceedingly pronounced. This study offers new insights into the effects of drugs on repolarization and suggests added benefits of using a composite T-wave morphology score for cardiovascular risk stratification in combination with the assessment of the QT interval.

ECG EXPRESSION OF DRUG EFFECTS

After administration of the drugs used in the studies, there was typically a clear increase in the flatness and the asymmetry of the T-wave and often notches in the T-wave occurred. The drug-induced changes were similar to the differences seen between healthy subjects and patients with the LQT2 syndrome. These T-wave features are the hallmarks of severely abnormal repolarization that has often been linked to TdP arrhythmia. Notched T- waves are also included as a morphology characteristic in the diagnostic criteria for the congenital Long QT syndrome. These T-wave characteristics were not seen in subjects prior to drug administration and remained absent with placebo treatment. The development of notches on the T-wave was more frequently associated with administration of drugs that have substantial or apparently elevated torsadogenic potential such as d,l-sotalol and sertindole, respectively.

Each of the individual LQT2 markers that were investigated, including their combined overall expression of T-wave morphology, developed concurrent with blood plasma concentrations of the drug. It was further demonstrated that such morphology patterns were dose dependent and that the T-wave characteristics showed an accumulation over days when dosing to a steady state.

Combined, the results imply that there is a close relationship between drug influence and the development of specific LQT2-like T-wave morphology patterns, which is consistent with the underlying cellular electrophysiology of I_{Kr} inhibition caused by the various drugs investigated in this work. These observations also support the conjecture that the T-wave patterns that are indicative of abnormal repolarization in the congenital LQT2 syndrome, can be used as, electrophysiologically motivated, indices to quantify the degree of drug-induced repolarization abnormalities. This finding advocates further research into the possibility of using quantitative T-wave morphology to categorize drugs, not only by the degree of drug-induced abnormal repolarization, but, more importantly, by their proarrhythmic potential. In addition, LQT2-like patterns of T-wave morphology may be used in pharmacological stress testing to uncover latent repolarization disorders and as guidance for genotyping of people with suspected long QT syndrome.

CONSISTENCY OF DRUG EFFETCS

The administration of drugs often led to prominent changes of T-wave morphology, in some cases without concomitant prolongation of the QTcF interval. The average subject had drug-induced repolarization abnormalities that were easier to discriminate from baseline by the degree of T-wave morphology changes than by QTcF prolongation. Following drug treatment, subjects with a shorter QTcF interval on baseline tended to respond with a larger prolongation of the repolarization interval than patients with longer baseline QTcF values. This tendency was not seen for T-wave morphology.

In general, the drugs, and in particular those associated with high risk of inducing arrhythmia, appear to have a more consistent effect on T-wave morphology than on QTcF prolongation. From a clinical perspective, this favorable property of T-wave morphology analysis could be useful during electrocardiographic monitoring of patients taking certain QT prolonging drugs. Monitoring of the QT interval is already mandatory for some drugs. However, the QT interval may fail to identify all subjects with an increased risk of developing arrhythmia and it would, therefore, be relevant to extend electrocardiographic monitoring to also include evaluation of drug-induced T-wave morphology changes.

The implications for thorough QT studies, of a consistent identification of drug-induced repolarization changes are obvious. The thorough QT study relies on identification of drug-induced QTc change from baseline as the primary end point, and given the low threshold for regulatory concern, the possibility exists that incorrect conclusions will be drawn about the cardiovascular safety of a drug. The thorough QT study also investigates categorical responses, which in the case of inconsistent recognition of drug effects would render this type of analysis less useful. There are also potential problems associated with the QT prolongation being a skewed function of baseline QT interval, which was found to exist for drug-induced prolongation of QTcF but not for changes in T-wave morphology. This concern relates to violation of the assumption of a homogeneous drug response underlying the statistical analyses typically performed in the thorough QT studies. Differential drugs responses could jeopardize the sensitivity and validity of these statistics and possibly lead to wrong conclusions about a drugs' propensity to alter cardiac repolarization.

MAGNITUDE OF DRUG EFFECTS

The changes in T-wave morphology were shown to be more pronounced manifestations of drug-induced repolarization abnormalities than QT interval prolongation. Drug effects on T-wave morphology could be larger for individual subjects, for mean estimates around the time of peak plasma concentrations, and for the maximum time-matched change from baseline, which is the primary end point in drug trials. Drug effects could be more than 10 times greater for T-wave morphology compared to the effects on QT interval prolongation and this factor was positively related to the supposed torsadogenic potential of the investigated drugs. A low drug dose of a torsadogenic drug could have the same effect on T-wave morphology as a higher dose of the drug would have on QT interval prolongation. No changes in T-wave morphology were induced by placebo treatment.

Collectively, these findings highlight the importance extending electrocardiographic analysis of drug effects to the entire repolarization segment instead of merely using the end of the T-wave to measure the QT interval. The literature clearly indicates that an increase in local and/or global heterogeneity, i.e. transmural, apex-base, left-right, of the repolarization process in the heart is associated with an increased risk of arrhythmia. The QT interval does not reflect any of this heterogeneity, whereas the shape of the T wave is directly affected by it. This aspect of repolarization is thus better captured by changes in T-wave morphology than by prolongation of the QT interval. Surely, this property of T-wave morphology extends favorably to investigation of repolarization effects and assessment of cardiac safety in clinical studies of new drugs. Indeed, it was shown that the use of T-wave morphology in drug trials would increase statistical power or, alternatively, would reduce the cost of conducting such trials by enrolling fewer subjects. At the same time, T-wave morphology may also be used in drug studies for categorical analysis to indicate exaggerated response in subjects with a reduced repolarization reserve.

COVARIATE DRUG EXPRESSIONS

This work has demonstrated that computerized analysis of LQT2-like T-wave patterns, may be used to identify and explore complex drug effects on cardiac repolarization that cannot be described by the QT interval alone. It has been shown that drugs with different torsadogenic potential can induce significantly different changes in T-wave morphology for a given QT interval prolongation. Moreover, it was shown that the relationship between QT interval prolongation and changes in T-wave morphology is very different for drugs with different risk profiles.

More pronounced T-wave morphology changes were seen at similar QT interval prolongations for drugs with a supposedly higher proarrhythmic risk.

These findings may add to our understanding of why drugs, which prolong the QT interval to the same extent, can have different incidences of arrhythmia and, vice versa, why drugs, associated with a similar incidence of arrhythmia do not necessarily prolong the QT interval to an equivalent degree.

Certainly, the assumption that a given threshold for QT interval prolongation is indicative of a certain risk is not correct. A covariate description of drug-induced changes, which includes both T-wave morphology and QT interval prolongation, may be important for clinical trials for cardiac safety assessment of new drugs. Using this covariate approach it may be possible to address the shortcomings of the assumption of a simple QT-proarrhythmia relationship. Most notably, it may be possible to characterize drugs with similar QT interval prolonging effects as having different torsadogenic potential.

PUBLICATION LIST

PUBLICATIONS

ORIGINAL ARTICLES

1. **Graff C**, Struijk JJ, Matz J, Kanters JK, Andersen MP, Nielsen J, Toft E. Covariate analysis of QTc and T-wave morphology: New possibilities in the evaluation of drugs affecting cardiac repolarization. *Clin. Pharmacol Ther.* 2010; 88: 88-94

2. Johannesen L, Grove USL, Sørensen JS, Schmidt ML, Couderc JP, **Graff C**. A wavelet-based algorithm for delineation and classification of wave patterns in continuous Holter ECG recordings. *Comput Cardiol* 2010; 37: 979-982

3. Sørensen JS, Johannesen L, Grove USL, Lundhus K, Couderc JP, **Graff C**. A comparison of IIR and wavelet filtering for noise reduction of the ECG. *Comput Cardiol* 2010; 37: 489-492

4. Johannesen L, Grove USL, Sørensen JS, Schmidt M, **Graff C**, Couderc JP. Analysis of T-wave amplitude adaptation to heart rate using RR-binning of long-term ECG recordings. *Comput Cardiol* 2010; 37: 369-372

5. Schmidt S, Holst-Hansen C, **Graff C**, Toft E, Struijk J. Segmentation of heart sound recordings by a duration-dependent hidden markov model. *Physiol. Meas.* 2010; 31: 513-529

6. Haarmark C, **Graff C**, Andersen MP, Hardahl TB, Struijk JJ, Toft E, Xue JQ, Rowlandson GI, Hansen PR, Kanterrs JK. Reference Values of Electrocardiogram Repolarization Variables in a Healthy Population. *J. Electrocardiol.* 2010; 1: 31-39

7. Nielsen J, Andersen MP, **Graff C**, Hardahl TB, Dybro J, Struijk JJ, Meyer JM, Toft E. The Effects of Sertindole on QTd and TpTe *Acta Psychiatr. Scand.* 2010; 12: 385-388

8. **Graff C**, Andersen MP, Xue JQ, Hardahl TB, Kanters JK, Toft E, Christiansen M, Jensen HK, Struijk JJ. Identifying Drug-Induced Repolarization Abnormalities from Distinct ECG patterns in Congenital Long QT Syndrome: A study of d,l-sotalol effects on T-wave morphology. *Drug Saf.* 2009; 32: 599-611

9. **Graff C**, Matz J, Christensen EB, Andersen MP, Kanters JK, Toft E, Pehrson S, Hardahl TB, Nielsen J, Struijk JJ. Quantitative Analysis of T-wave Morphology Increases Confidence in Drug-induced Cardiac Repolarization Abnormalities: Evidence from the investigational I_{Kr} inhibitor – Lu 35-138. *J. Clin. Pharmacol.* 2009; 49: 1331-1342

10. **Graff C**, Matz J, Andersen MP, Kanters JK, Nielsen J, Xue JQ, Toft E, Struijk JJ. T-wave Morphology as a Covariate in Drug-induced QTc Prolongation. *Comput. Cardiol.* 2009 (in Press)

11. Nielsen J, **Graff C**, Hardahl TB, Andersen MP, Kristoffersen J, Struijk JJ, Toft E, Meyer JM. Sertindole causes distinct electrocardiographic T-wave morphology changes. *European Neuropsychopharmacology* 2009; 19: 702-707

12. Haarmark C, Hansen PR, Vedel-Larsen E, Pedersen SH, **Graff C**, Andersen MP, Toft E, Wang F, Struijk JJ, Kanters JK. The prognostic value of the Tpeak-Tend interval in patients undergoing primary percutaneous coronary intervention for ST-segment elevation myocardial infarction. *J. Electrocardiol.* 2009; 42: 555-560

13. **Graff C**, Matz J, Andersen MP, Kanterrs JK, Toft E, Pehrson S, Struijk JJ. Sensitivity of T-wave Morphology and the QT interval to Small Drug-induced Electrocardiographic Changes. *Comput. Cardiol.* 2008; 35: 319-322

14. Andersen MP, Xue JQ, **Graff C**, Kanters JK, Toft E, Struijk JJ. New descriptors of T wave morphology are independent of heart rate. *J. Electrocardiol.* 2008; 41: 557-561

15. Schmidt S, Toft E, Holst-Hansen C, **Graff C**, Struijk JJ. Segmentation of Heart Sound Recordings from an Electronic Stethoscope by a Duration Dependent Hidden Markov Model. *Comput. Cardiol.* 2008; 35: 345-349

16. Kanters JK, Haarmark C, Vedel-Larsen E, Andersen MP, **Graff C**, Struijk JJ, Thomsen PEB, Christiansen M, Jensen HK, Toft E. T(peak)T(end) Interval in Long QT Syndrome. *J. Electrocardiol.* 2008; 41: 603-608

17. Andersen MP, Xue JQ, **Graff C**, Hardahl TB, Toft E, Kanters JK, Christiansen M, Jensen HK, Struij JJ. A Robust Method for Quantification of Ikr-related T-wave Morphology Abnormalities. *Comput. Cardiol.* 2007; 34: 341-344

18. Schmidt S, Holst-Hansen C, **Graff C**, Toft E, Struijk JJ. Detection of Coronary Artery Disease with an Electronic Stethoscope. *Comput. Cardiol.* 2007; 34: 757-760

19. Kanters JK, **Graff C**, Andersen MP, Hardahl TB, Toft E, Christiansen M, Thomsen PEB, Struijk JJ. LQTS Genotyping by ECG: Fact, Fiction, or

Something in Between? *J. Electrocardiol.* 2006; 39: 119-122

20. Struijk JJ, Kanters JK, Andersen MP, Hardahl T, **Graff C**, Christiansen M, Toft E. Classification of the Long QT syndrome Based on Discriminant Analysis of T-wave Morphology. *Med. Biolog. Eng. Comput.* 2006; 44:543-549

21. Struijk JJ, Kanters JK, Andersen MP, Hardahl TB, **Graff C**, Christiansen M, Toft E. Classification of the Long QT syndrome Based on Discriminant Analysis of T-wave Morphology. *Comput. Cardiol.* 2005; 32: 511-514

PATENTS

22. **Graff C**, Andersen MP, Kanters JK, Struijk JJ, Toft E, Hardahl TB. A System and a Method for Analyzing ECG Curvature for Long QT Syndrome and Drug Influence. WO2005058156

23. **Graff C,** Andersen MP, Hardahl TB, Struijk JJ, Xue JQ. A System and a Method for Dimensionality Reduction of ECG Signals for ECG Analysis and Presentation WO2008064682 (Application)

24. Hardahl TB, **Graff C**, Andersen MP, Struijk JJ, Xue JQ, Toft E. System and Method for Analyzing Complex Curvature of ECG Curves. WO2008064679 (Application)

25. Xue JQ, Struijk JJ, Andersen MP, **Graff C**, Hardahl TB. Method and System for Electrocardiogram Evaluation . EP1935336 (Application)

26. Schmidt SE, Struijk JJ, **Graff C**. MultiParametric Classification of Cardiovascular Sound. WO2008000254 (Application)

27. Schmidt SE, **Graff C**, Struijk JJ, Toft E. Method for Segmenting a Cardiovascular Signal. WO2008000255 (Application)

28. Hardahl TB, **Graff C**, Andersen MP, Toft E, Struijk JJ, Kanters J. System and a Method for Analyzing ECG Curvature. US2005177049 (Application)

ABSTRACTS

1. **Graff C**, Toft E, Matz J, Kanters JK, Andersen MP, Nielsen J, Struijk JJ. Covariate analysis of T-wave morphology and QTc prolongation: New insights into drug-induced ECG changes. *WorldPharma*, Copenhagen, Denmark (2010)

2. **Graff C**, Toft E, Matz J, Kanters JK, Andersen MP, Nielsen J, Struijk JJ. Covariate assessment of T-wave morphology and QTc prolongation: New opportunities in the evaluation of drug-induced ECG changes. *Danish Cardiovascular Research Academy*, Sønderborg, Denmark (2010)

3. **Graff C**, Matz, J, Andersen MP, Kanters JK, Nielsen J, Xue JQ, Toft E, Struijk JJ. Drug-induced ECG Changes: Beyond the QT Interval. *Danish Society for Biomedical Engineering*, Brædstrup, Denmark (2009)

4. Schmidt S, Toft E, Holst-Hansen C, Graff C, Struijk J. Segmentation of heart sound recordings by a duration dependent hidden Markov model. *Danish Cardiovascular Research Academy*, Sønderborg, Denmark (2009)

5. Haarmark C, Hansen PR, Vedel-Larsen E, Haahr-Pederrsen S, **Graff C**, Andersen MP, Toft E, Struijk JJ, Kanterrs JK. Alterations in Tpeak-Tend Interval, ST Segment Deviation and QTc in Patients with ST Segment Elevation Myocardial Infarction Immediately After Primary PCI. *European Cardiac Arrhythmia Society*, Paris, France (2009)

6. **Graff C**, Matz J, Andersen MP, Kanters JK, Toft E, Pehrson S, Struijk JJ. Drug-induced Effects on T-wave Morphology and QTc. *Danish Society of Pharmacology and Toxicology*, Odense, Denmark (2008)

7. Nielsen J, **Graff C**, Hardahl TB, Andersen MP, Kristoffersen J, Struijk JJ, Toft E. ECG Changes During Sertindole Treatment. *European College of Neuropsychopharmacology*, Barcelona, Spain (2008)

8. Struijk JJ, Andersen MP, **Graff C**, Kanters JK, Xue JQ, Toft E. Inferior and Lateral Leads Show Similar Tpeak-Tend Intervals in Sotalol-induced QT Prolongation. *International Society for Computerized Electrocardiology*, Riverside, CA, USA (2008)

9. Fanø S, Haarmark C, Jensen GB, Andersen MP, **Graff C**, Struijk JJ, Toft E, Christiansen M, Kanters JK. Methadone Induced hERG-blockage Compared to Congenital hERG Mutations (LQT2) Illustrated by T-wave Morphology Changes. *European Cardiac Arrhythmia Society*, Marseille, France (2008)

10. **Graff C**, Matz J, Andersen MP, Kanters JK, Toft E, Pehrson S, Struijk JJ. Sensitivity of QTc and T-wave Morphology to Small Drug-induced Electrocardiographic Changes. *International*

Society for Computerized Electrocardiology, Riverside, CA, USA (2008)

11. Matz J, **Graff C**, Andersen MP, Kanters JK, Toft E, Pehrson S, Struijk JJ. Sensitivity of T-wave Morphology and the QT Interval to Small Drug-induced ECG Changes. *DIA - Cardiac Safety Conference*, Barcelona, Spain (2008)

12. Haarmark C, Andersen MP, **Graff C**, Hardahl TB, Struijk JJ, Toft E, Hansen PR, Kanterrs JK. T-wave Amplitude in Lead V1 is Bimodally Distributed. *European Cardiac Arrhythmia Society*, Marseille, France (2008)

13. **Graff C**, Matz J, Andersen MP, Kanters JK, Toft E, Pehrson S, Struijk JJ. T-wave Morphology is More Sensitive to Drug-induced Changes than the QT Interval. *Danish Cardiovascular Research Academy*, Sønderborg, Denmark (2008)

14. Haarmark C, Andersen MP, **Graff C**, Hardahl TB, Struijk JJ, Toft E, Christiansen M, Jensen HK, Kanters JK. T-wave Morphology Differences Depending on Genotype in Long QT Syndrome. *European Cardiac Arrhythmia Society*, Marseille, France (2008)

15. **Graff C**, Andersen MP, Xue JQ, Hardahl TB, Kanters JK, Toft E, Christiansen M, Jensen HK, Struijk JJ. T-wave Morphology Reveals Greater Effect of d,l-sotalol than QTc. *International Society for Computerized Electrocardiology*, Riverside, CA, USA (2008)

16. Haarmark C, **Graff C**, Andersen MP, Hardahl TB, Struijk JJ, Toft E, Xue JQ, Rowlandson GI, Kanters JK. Independent Novel T-wave Descriptors of Repolarization. *International Society for Computerized Electrocardiology*, Cancun, Mexico (2007)

17. Haarmark C, **Graff C**, Andersen MP, Hardahl TB, Struijk JJ, Toft E, Xue JQ, RowlandsonGI, Kanterrs JK. Quantifying the T-wave as a Repolarization Marker. *European Cardiac Arrhythmia Society*, Marseille, France (2007)

18. Andersen MP, Xue JQ, **Graff C**, Hardahl TB, Christiansen M, Toft E, Kanters JK, Struijk JJ. Repeatability of T-wave Morphology Measurements: Superiority of a Principal Component Analysis-based Lead. *International Society for Computerized Electrocardiology*, Cancun, Mexico (2007)

19. **Graff C**, Xue JQ, Andersen MP, Kanters JK, Hardahl TB, Toft E, Christiansen M, Struijk JJ. T-wave Convexity Measure Outperforms Notch Criterion as Diagnostic Marker for the hERG Genotype. *International Society for Computerized Electrocardiology*, Cancun, Mexico (2007)

20. Andersen MP, Xue JQ, **Graff C**, Hardahl TB, Christiansen M, Toft E, Kanters JK, Struijk JJ. T-wave Morphology Analysis Identifies Congenital and Drug-induced Long QT Syndrome. *Danish Cardiovascular Research Academy*, Sønderborg, Denmark (2007)

21. Haarmark C, **Graff C**, Andersen MP, Hardahl TB, Struijk JJ, Toft E, Christiansen M, Kanterrs JK. T-wave Morphology in the Long QT Syndrome. *American Heart Association*, Orlando, FL, USA (2007)