

Two Probabilistic Methods to Characterize and Link Drug Related ECG Changes to Diagnoses from the PTB Database: Results with Moxifloxacin

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Abstract

The QT interval is the lead biomarker for predicting risk of severe ventricular pro-arrhythmia in drug safety assessment. QT is a weak predictor, with no direct link to clinical endpoints.

We propose two methods that might bridge the gap. Method A is based on the study level probabilistic intra-subject clustering of 12-lead based ECG waveform similarities, comparing pre-treatment with placebo and treatment within and between study periods.

Method B: Population related approach where all 12-lead ECG waveforms from a study are compared with the fully diagnosed 30.000 ECGs in the PTB database. ECGs are assigned a probability for normality and a limited number of cardiac main diagnoses contained therein.

In a crossover study with moxifloxacin 200 and 400 mg iv, QT-prolongation and method A revealed significant treatment effects, while method B showed no relevant treatment related change to non-normal diagnosis.

1. Introduction

During the past decades many drug substances have been investigated for their potential to cause serious adverse cardiac effects, especially life-threatening arrhythmias like ventricular tachycardia and Torsade de Pointes (TdP). Guideline ICH E14 reflects the need for assessment of these risks [1].

The QT interval is the only commonly acknowledged biomarker for the assessment of drug-related pro-arrhythmic risk. Unfortunately, QT is known to be a weak biomarker, as some drugs, like Micronesia, prolong the QT interval significantly, but show a rather low risk profile. Other drugs are regarded dangerous but exhibit mild QT prolongation only.

Despite of all efforts to improve the predictability of QT for pro-arrhythmia, (e.g., different heart rate correction methods, waveform morphology parameters, U wave detection, age, sex [2]) there is still no reliable

method for pro-arrhythmia assessment. The search for markers of pro-arrhythmia other than QT is limited to material from clinical phase-I studies, which lacks information on medically meaningful terminal endpoints.

Two basic ideas were pursued to perform non-QT related risk assessment for drugs :

(1) The study level approach (method A) which performs intra-subject correlative clustering of ECG waveform similarities, comparing waveform changes by verum (=moxifloxacin) with those by placebo.

(2) The population level approach (method B) which has the intention of linking ECG waveforms with diagnoses for study ECGs. The method is based on the PTB database that contains waveforms, metadata and diagnoses performed by cardiologists for 30,000 ECGs. Each study ECG is correlatively compared to every ECG in the database. Based on this correlation a probability for association with normal and non-normal diagnoses is estimated. Verum treatment that has a propensity for pro-arrhythmia should increase the probability of non-normal diagnoses that are related with pro-arrhythmia. This method may provide a novel medically meaningful biomarker.

A proof-of-concept evaluation was done using a single-blinded crossover design phase-I study in ten subjects (volunteers), treated with isotonic saline, 200 mg (M200) and 400 mg moxifloxacin (M400) iv. infusion.

Moxifloxacin (Bayer Avelox[®]) is an antibiotic known to induce mild QT-prolongation that is considered safe with a low pro-arrhythmia risk profile. It is widely used as a positive control in thorough QT studies according to ICH E14 [1].

Therefore, the expected study outcome is a significant QT prolongation by 400 mg moxifloxacin, and a borderline result for the 200 mg treatment. Method A was expected to reveal a significant treatment effect, while method B was not expected to show a significant treatment effect, being expressed by an increase in risk probabilities assigned to non-normal diagnose status.

2. Methods

The clinical study M04-704 was conducted in the Abbott Clinical Research Phase-I Unit (ACPRU) at Abbott GmbH & Co. KG, Ludwigshafen/Rhein. The purpose of the study was to investigate the effects of single dose 200 mg and 400 mg iv. moxifloxacin (both together named “verum”) in comparison to placebo (isotonic saline solution). The drug was administered as a 250 ml infusion over one hour. ECGs and drug blood plasma levels were measured within a post-observation period of 24 hours per treatment.

The standard adult treatment dose of 400 mg moxifloxacin is known to induce QT prolongation exceeding 12 ms. The 200 mg dose was included in the study, as the ICH-E14 guideline decrees the inclusion of a positive control with a mean effect of about 5ms on the QT/QTc interval (i.e., an effect that is close to the QT/QTc effect that represents the threshold of regulatory concern, which is 5 ms average and 10 ms upper 95% confidence limit)[1].

The study was conducted as a two-period, single-blinded, placebo-controlled, randomized crossover design with 2 study periods of 3 full days plus a post-observation day. Each full day had 7 visits with recording of triplicate resting ECGs. An additional triplet ECG was recorded 24 hours after last treatment on day 4. Each ECG triplet was followed by a blood draw for the assessment of drug blood plasma concentration. A wash-out period of two weeks was allowed between the periods. Eight adult male and two female volunteers (all Caucasian; aged 27 to 45 years) in good health were selected to participate in the study according to the protocol selection criteria.

All subjects gave informed written consent. The study was conducted in accordance with the protocol, ICH and good clinical practice (GCP) guidelines, applicable regulations and guidelines governing clinical study conduct and ethical principles according to the Declaration of Helsinki. The investigator ensured that the study was conducted in accordance with the provisions as stated by Food and Drug Administration (FDA).

For reasons of study result comparability, we briefly report the results by use of the classical evaluation scheme, a standard “delta” QT analysis (difference of the QTcF difference profile comparing to baseline profile).

Methods A and B both require identical preprocessing steps. Detection of fiducial points P, Q, R, S, T for a single 10 second standard ECG is followed by the selection of one characteristic ECG beat, which is normalized for heart rate according to the Fredericia correction formula.

Method A is based on lead-by-lead cross correlation of the selected characteristic ECG beat with the beats selected for all other ECGs of each subject (in our example, a total of 134 ECGs). We calculated the Euclidean distance (based on median and standard

deviation of the 12 cross correlations). The cross correlation results for each subject were accumulated per study day and visit, resulting in up to 21 single ECG distance vectors. Calculation of Euclidean cluster distances for each subject for the complete study was performed. For each subject treatment effect (verum vs placebo) a t-test for dependent samples was performed.

Method B is based on a PTB developed approach [3] exploiting ECG waveform similarities linked to medical diagnoses. PTB’s database of 12-lead waveforms contains demographic subject descriptions, findings, details on anamnesis, cardiologist-assessed diagnosis and medication details. It contains approximately 30,000 ECGs with manifold cardiac diagnoses from normal to severest grades of cardiac diseases.

The similarity of two ECG patterns is determined by cross-correlation. The normalization of waveforms allows to compare ECGs with different heart rates. Through comparison of the characteristic beat waveform with the waveforms stored in the database a subset of database ECGs is selected that best matches the corresponding leads of the undiagnosed ECG. This procedure allows assigning a pattern of most probable diagnoses to the undiagnosed ECG. The resulting probability vector of best match diagnoses (BMD) is the basis of the subsequent statistical calculations.

The method has been validated [4] by a database of more than 10,000 ECGs with unknown diagnoses, post classified by cardiologists. Depending on group of cardiac diagnoses, sensitivities and specificities between 72% and 95% were achieved in 8,500 individual examinations. Rare diagnoses could not be evaluated for performance due to sparse data (~1500 ECGs).

The processing steps for method B are [5]:

- (1) Lead by lead cross-correlation of the selected characteristic ECG beat with about 30,000 ECGs in the ECG database of PTB .
- (2) Calculation of Euclidean distance (from median and standard deviation of the cross correlations per ECG).
- (3) Selection of the 50 best-matched ECGs from the database for each study ECG.
- (4) Each of these best-matched ECGs contributes its probability weights of BMD resulting in a probability vector of diagnoses. This BMD vector reflects the most probable associations between each study ECG and its medical endpoint.
- (5) On the subject level, a time and treatment related plot of this BMD vector reveals a potential drug related shift of risk profile.
- (6) A treatment summary of the risk profile per subject is compiled.
- (7) On the study level a treatment-related summary of the risk profile is compiled.
- (8) A generalized linear model (GLM) with Tukey’s bi-weight robustification method is used to

analyse the probability for BSD category “normal” as the target variable, with study factors treatment, period, hour and subject, and their interactions as independent variables.

The significance level was set at $\alpha=0.05$ for all statistical tests performed.

3. Results

3.1. Standard QTc evaluation

The standard evaluation method with $\Delta QTcF$ (see Figure 1) resulted in a maximum difference at hour 1 (=end of infusion) of 7.3 ms for M200, and 17.1 ms for M400. The “double delta” method, $\Delta\Delta QTcF$, estimates the differences of the $\Delta QTcF$ profile of treatment versus placebo; this resulted in differences of 6.7 ms for M200 and 13.5 ms for M400.

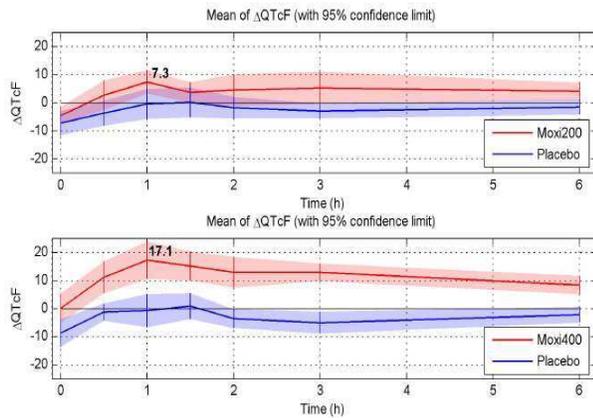


Figure 1: Standard $\Delta QTcF$ is shown for each visit of the 10 s ECG recording scheme. The upper part shows the baseline corrected mean for the ten subjects (and three replicates) with a 95% confidence limit for day 1 of treatment (M200), while the lower part shows the same for day 2 (M400). The figure at C_{max} (hour 1) denotes the mean value.

3.2. Result for method A

As an example of processing steps, intermediate results for subject 108 are shown (see figure 2).

The accumulated results for all 10 subjects by treatment are shown in figure 3. The intra-subject comparison by a paired t-test allowed an intra-individual assessment of waveform similarities (resp. dissimilarities). In 4 out of 10 subjects we observed no significant waveform similarity change by moxifloxacin treatment (M200 and M400 taken together vs. placebo).

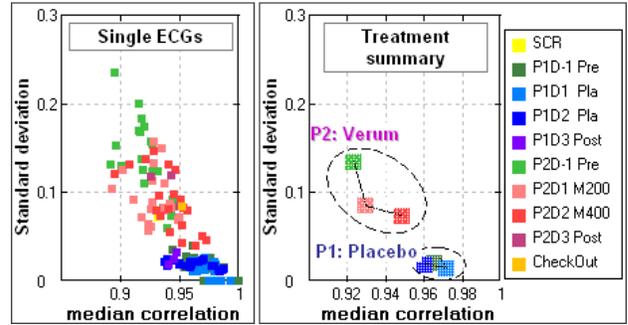


Figure 2: Method A: subject 108. Left: intermediate similarity plot (median correlation vs standard deviation) for the first ECG vs. each other in the study time series. Right: median by day and treatment.

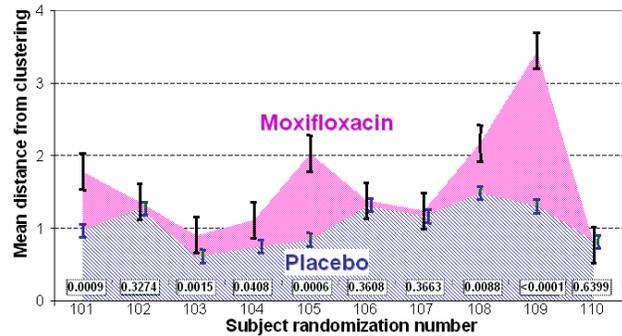


Figure 3: Method A: Mean difference between pre-treatment and treatment by subject. Bars denote one standard deviation around the mean. The boxes at the bottom show the p-value of the t-test for paired data.

3.3. Result for method B

Figure 4 shows an example of the BMD time series results. Visual inspection shows some daytime variation, and a clear distinction between periods. There is clearly no association with verum treatment in this case. Most of the BMD profiles look similar, showing differences between subjects and periods, but no distinguishable treatment related patterns.

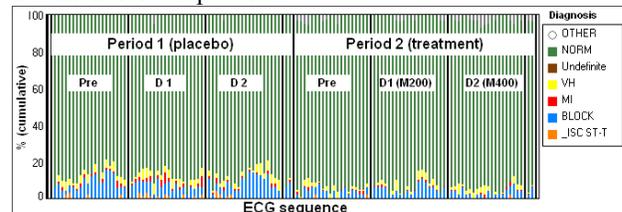


Figure 4: Method B: subject 108. Best match diagnosis (BMD) probabilities for the ECG sequence of all 134 ECGs as cumulative percentage of diagnoses.

For the BMD category “normal” the robustified GLM analysis was performed using the square transformation identified as optimal by the Box-Cox procedure. ANOVA (Table 1) showed a dominant effect by the

random factor subject, and a subject-by-treatment interaction. Hour (time of ECG measurement relative to treatment) and hour-by-treatment interaction were also significant. However, there was neither a significant general treatment effect, nor a general period effect. Period-by-subject interaction could not be tested in this model. However, inspection on the subject level indicated differences, albeit inconsistent.

Source	df	F-Ratio	Significance (p)
SUBJECT (S)	9	42.250	<0.001 *
PERIOD (P)	1	0.340	0.5601 -
HOUR (H)	7	4.672	< 0.001 *
TREATMENT (TRT)	1	0.043	0.8409 -
S*TRT	9	24.670	<0.001 *
H*TRT	7	2.551	0.0132 *
Residual	1281		(alpha=0.05)

Table 1: Model B ANOVA summary. * in last column denotes a significant, - a non-significant test result.

BMD medians (see figure 5) by subject, sequence and treatment show considerable variation that is possibly attributable to external uncontrolled influences.

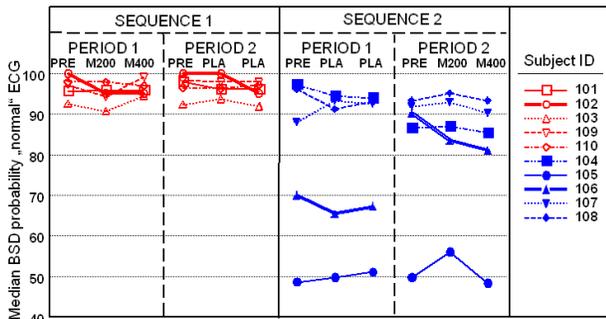


Figure 5: Method B: Probabilities for category “normal” by subject, treatment sequence, period and treatment.

Of note, two of the subjects randomized into sequence 2 had been included in the study despite of their cardiac problems (subject 105: right bundle branch block; subject 106: ST-elevation).

4. Discussion and conclusions

Study results for QT prolongation showed an average „double-delta“ QTcF difference of 6.7 ms for 200 mg and 13.5 ms for 400 mg moxifloxacin treatment.

Results for method A showed larger similarities of ECG patterns (based upon Euclidean distance) between baseline and placebo conditions than under treatment conditions. Six out of ten subjects could be termed as responders, while four did not show any significant treatment related effects.

According to method B, there was no significant general treatment influence. This confirms the expected

outcome, because the arrhythmic propensity of moxifloxacin is generally low.

Nevertheless, some subjects responded to treatment with inconsistent direction of change while others did not. Although, variations in treatment effect were also observed with method A, the direction of change was consistent.

Table 1 shows a significant, albeit minor hour-by-treatment interaction effect, which may indicate that moxifloxacin changes the BMD assignment to category “normal”.

In contrast to the statistical results in table 1, figure 5 does not indicate that moxifloxacin systematically changes BMD probabilities to category “non-normal”.

Both methods yield additional evidence on moxifloxacin treatment-related changes, and allow insights that the usual QT analyses cannot provide. Method A allows judgment on intra-individual waveform changes, and has the potential to identify persons responsive to treatment. Method B showed no relevant treatment related changes in terms of diagnostic BMD probabilities. and bears the potential to detect signs of pro-arrhythmic risk.

To confirm the diagnostic potential of the proposed methods a proof-of-concept study including known pro-arrhythmic drugs like Sotalol will be needed.

Acknowledgements

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References

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