Near-Thorough QT Study as Part of a First-In-Man Study

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J. Clin. Pharmacol. 2008; 48; 1146 originally published online Aug 29, 2008;
DOI: 10.1177/0091270008323261

Near-Thorough QT Study as Part of a First-In-Man Study

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Detailed electrocardiographic (ECG) support was provided to a first-in-man, single-ascending-dose study that included 6 cohorts of 8 male volunteers each. In each cohort, 6 and 2 subjects received active compound and placebo, respectively. Long-term 12-lead ECGs were obtained on baseline day –1, dosing day 1, and day 2. Automatic QT-interval measurements were made at 63 time points (28 at baseline and 35 on treatment). Based on QT/RR distribution, 20% of measurements were visually verified. Baseline-corrected time-matched ΔQTc values were obtained at 35 postdose time points. Placebo subjects of all cohorts were pooled. When 2 cohorts of the lowest, middle, and highest doses were pooled (12 subjects per active treatment group), the spreads of placebo-corrected ΔΔQTc values were within the regulatory requirements (single-sided 95% confidence interval <10 milliseconds) at all time points. Thus, this ECG support of the first-in-man study provided data of regulatory acceptable accuracy at a small fraction of the cost of a full thorough QT study.

Keywords: *Electrocardiography; QT interval; first-in-man study*

Journal of Clinical Pharmacology, 2008;48:1146-1157

The so-called thorough QT/QTc study (TQT) is required during the development of practically all new drugs.[1] The ICH E14 regulatory guidance and standard requirements by regulatory bodies prescribe several mandatory components for TQT studies,[1] including the positive control investigation (ie, a pharmacological intervention known to prolong the heart-rate-corrected QTc interval to an expected level) and electrocardiographic (ECG) monitoring covering the pharmacokinetic profile of both the parent compound and known metabolites. Although not strictly required by regulatory guidance, verification of ECG measurements is not infrequently performed by independent laboratories.[2] In addition, the US Food and Drug Administration (but presently not other regulatory bodies) requires submission of electronic ECG waveforms[3] for regulatory review. Perhaps most important, the regulatory guidance sets strict limits for the interpretation of drug-induced QTc changes. Because the interpretation is driven by the upper confidence interval rather than by the mean QTc change,[1] the implementation of the TQT study frequently involves a large number of subjects. All these obligations make the conduct of TQT studies both demanding and costly.

A positive TQT study, that is, a study documenting drug-induced QT-interval prolongation above the threshold of regulatory interest,[4] needs to be interpreted together with risk–benefit considerations of the new drug but almost always has to be followed by intensive ECG monitoring during the advanced phases of clinical development. It is therefore not surprising that compounds with positive TQT study results are often discontinued. Scheduling of TQT studies thus presents a dilemma to drug developers. Conducting such a study before proof of concept investigations is potentially wasteful because the expense of a TQT study is not recouped if proof of concept is eventually not established. Conducting a TQT study after establishing proof of concept may mean a substantial loss if the clinical development needs to be stopped when considerable expense has already accumulated.

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DOI:10.1177/0091270008323261

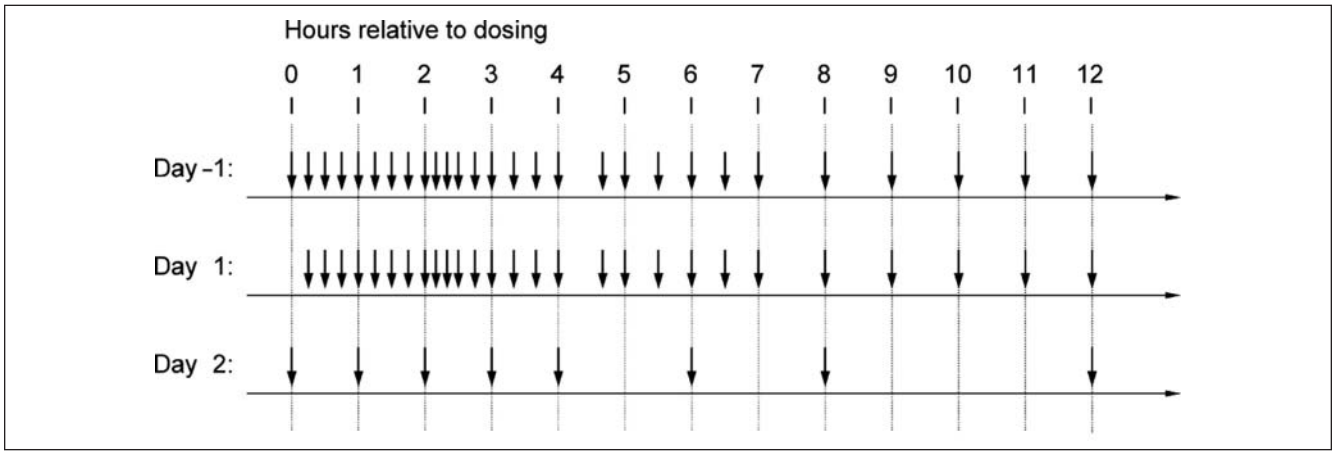

Figure 1. Distribution of the 5-minute periods (study data points) during which the subjects adopted strictly undisturbed supine positions (bold down arrows). The actual times were synchronized according to the dosing time on day 1. The gap between 4:00 and 4:40 hours:minutes after dosing allowed midday meal intake; the data point at 0 hours on day 1 was omitted because of the conflict with dosing procedures.

Many drug developers therefore collect ECG measurement during very early phases of the clinical program in hopes of predicting the outcome of a TQT study. However, such a collection of ECG data in early clinical studies is frequently polluted by substantial errors that limit the predictive value considerably. At the same time, presently available ECG technologies allow fairly accurate ECG measurements to be made during early clinical studies. Therefore, the aim of this article is to describe an example of a detailed ECG support of an early clinical study that may serve as a model for future investigations of this kind.

METHODS

WXYZ is a small molecule, the character of which is not important for the purposes of this text. Following preclinical development, WXYZ was investigated in a standard first-in-man, single-ascending-dose study that was performed in a dedicated clinical unit of Hammersmith Medicines Research in London. The study obtained approval by the regulatory authority and by the institutional ethics committee. All participants gave written informed consent.

First-in-Man Study

The first-in-man study included 6 cohorts, each of 8 young healthy male volunteers (mean age 26.1 ± 5.2 years). In each cohort, 6 volunteers received a single WXYZ dose and 2 received placebo, all in double-blind fashion. The 6 doses followed the standard regimen of a first-in-man, single-ascending-dose study, with each subsequent dose approximately doubling the preceding dose. In this text, the doses are denoted as A (lowest), B, C, D, E, and F (highest).

Electrocardiographic Recordings

In addition to undergoing standard screening and safety ECGs, each subject of the study underwent 3 continuous 12-lead ECG recordings, each of approximately 13 hours duration. These were obtained on the baseline day –1 before dosing, on the dosing day 1, and on the subsequent day 2. The recordings were made using 12-lead ECG monitors (SEER MC recorder, GE Healthcare, Milwaukee, Wis) programmed to obtain a 10-second ECG sample every 10 seconds.[5] Following careful skin preparation, new electrodes were used in Mason-Likar position to obtain each continuous ECG recording. In addition, standard safety ECGs were obtained during the study without electrode interference with the continuous recordings.

The study subjects were handled equally on days –1 and day 1. In addition to being supine in bed for several hours around and after the dosing time, they were strictly motionless and always, within each subject, in the same position during repeated 5-minute intervals between the dosing time and 12 hours afterward. The layout of these intervals (ie, the data points of the study) is shown in Figure 1. The data points were synchronized between day –1 and days 1 and 2 to allow calculation of time-matched differences from day –1 baseline. The data points

were most frequent between 2 and 3 hours post dose when, based on preclinical studies, maximum plasma concentrations of WXYZ were expected.

Electrocardiographic Measurements

For the purposes of analyses reported here, only the continuous ECG recordings were used. Each long-term record was separated into individual 10-second ECG segments, and in each of these the underlying heart rate and noise contents[6] were measured. Subsequently, in each of the 5-minute periods when the subjects were in the stable supine position, five 10-second ECGs were selected, each preceded by at least 90 seconds of stable heart rate with fluctuations <±2 beats per minute. The selected ECGs were not adjacent (ie, every 2 were separated by at least one 10-second ECG sample in between) and had the lowest noise contents among all 10-second ECG, satisfying the stability of preceding heart rate. Where <5 ECGs were found within the target period that satisfied the heart rate stability condition, 4 or 3 ECGs were also accepted to obtain representative data. Subsequently, the QTc measurement results (see further) of the individual 10-second ECGs selected in the 5-minute periods were averaged.

In addition to this ECG selection, further ECGs were selected from the baseline (day –1) recordings to characterize the drug-free QT/RR pattern for the purposes of individualized heart rate correction. Individual QT/RR pattern characterization has to be based on a sufficient heart rate range with comprehensive sampling between the slowest and fastest rates. This is not achievable using only recordings obtained in standardized stable positions because their heart rates do not differ sufficiently. Therefore, all 10-second ECGs were selected in baseline recordings that were preceded by >150 seconds of stable heart rate (variability <±2 beats per minute). These were sorted according to the preceding stable heart rate and divided, in each of the baseline long-term records, into 150 bins that covered the range between minimum and maximum stable heart rates as uniformly as possible. In each bin, the ECG with the lowest noise contents[6] was selected. In addition, 40 further ECGs were selected during hours 8 to 12 when the subjects were not in supine positions and, thus, their heart rates were increased. Thus, altogether, an additional 190 ECGs of 10 seconds each were selected from day –1 recordings.

In each selected ECG 10-second sample, the baseline trend and recording noise were removed using in-house-developed filtering techniques (modification of previously published filtering and baseline wander removal methods[7,8]). Subsequently, representative beats were constructed by superimposing individual beat images within the recording and by calculating the sample-by-sample median value of superimposed voltage values. Synchronized representative complexes (cubic spline interpolated to 1 kHz resolution) were obtained for each ECG lead separately. Positions of Q-onset and T-offset triggers in the representative waveform of each measured ECG sample were placed using in-house-developed pattern classification algorithm. The measurement involved the representative waveforms of all 12 leads.

In each study participant, all the QT and preceding heart rate measurements of selected ECGs (pooling all 3 long-term recordings) were considered, and the following subject-specific linear regression was calculated:

$$\log(QT_i) = \beta + \alpha \times \log(RR_i) + \varepsilon_i$$

where the RR_i interval characterized the stable heart rate preceding the QT_i interval measurement (QT and RR intervals expressed in seconds). From this regression equation, a provisional subject-specific heart rate correction formula was derived, $QTc = QT/RR^{\alpha}$, and 10% of the QT-interval measurements that resulted in the lowest provisional QTc values and 10% that resulted in the highest 10% values were selected for visual verification of the measurements. In these ECGs, the QT-interval measurement was visually verified and, where necessary, manually corrected on a computer screen showing the median beat (superimposition of all 12 leads on the same isoelectric axis) in a resolution of 1 millisecond per pixel. The same cardiologist performed the review of all the ECG patterns selected for manual verification.

After this manual verification of selected ECGs, all the measured ECGs of the same subject were pattern classified with an adjustment algorithm to make sure that similar morphological patterns of QRS onset and T-wave offset were measured systematically. Thereafter, the measurement was again visually checked and where necessary manually corrected (still by the same cardiologist) if the adjustment algorithm suggested that a QT-interval measurement change by >10 milliseconds. The result of this phase was taken as the final QT-interval measurement of the selected ECGs.

Heart Rate Correction

Following the visual verification and correction of automatic QT measurement, the drug-free QT/RR

patterns were obtained in each subject from all the ECGs that were measured in the baseline recordings only. Representative RR-interval durations were derived from stable heart rates preceding the measured ECGs. In each subject, the QT/RR pattern was converted into an individualized heart rate (HR) correction formula including QT/RR curvature optimization.[9,10] These individualized formulas were subsequently applied to all the data of each subject to obtain the final QTc values.

Pharmacokinetic Samples

Following drug administration, plasma levels of WXYZ were measured at 14 time points up to 48 hours after dosing.

Electrocardiographic Study Evaluation

To investigate the accuracy of the ECG measurement, standard deviations of QTc values measured in baseline recordings were evaluated in each subject separately.

Time-matched ΔQTc and ΔHR differences from baseline were calculated in each subject for all QTc readings on days 1 and 2.

The subjects who received placebo in individual cohorts were pooled (n = 12), and their time-matched ΔQTc readings on days 1 and 2 were averaged to obtain a ΔQTc placebo profile of the study. The time-matched ΔQTc readings on days 1 and 2 obtained in patients receiving active WXYZ treatment were averaged to obtain ΔQTc profiles corresponding to drug doses A to F. Because there were only 6 subjects on active treatment per dose, which were too few for meaningful statistics, the dose groups A+B, C+D, and E+F were also pooled to obtain 3 treatment groups of 12 subjects in each group. ΔQTc profiles were also obtained for these pooled groups. The on-treatment ΔQTc profiles were corrected for placebo to obtain ΔΔQTc profiles for each treatment group.

Individual baseline-corrected ΔQTc values were related to corresponding plasma levels of WXYZ. To correct for placebo, ΔQTc values obtained in subjects on placebo were added at 0 plasma level of WXYZ. Subsequently, a linear regression model was calculated between the baseline-corrected ΔQTc values and WXYZ plasma levels.[11] During calculation of the linear regression model, the total weight of placebo readings was adjusted to equal the total weight of the on-treatment readings, thus achieving a properly placebo-corrected relationship.

Unless otherwise stated, values are presented as means with 95% double-sided confidence intervals (CIs) that were calculated assuming normal distribution.

Tests of Methodology

The ECG processing methodology used in the study evaluation relied on, among other things, (*a*) accuracy of QT-interval measurement in carefully selected ECG samples, (*b*) accurate individualized heart rate correction, and (*c*) heart rate correction for the stable preceding heart rate rather than for the RR interval obtained at the moment of QT-interval measurement.

To test whether some of these factors can be safely omitted or simplified, we also reanalyzed the study using exactly the same analytical procedures as just described but using Fridericia-corrected QTc intervals. The correction with Fridericia formula[12] used the original QT-interval measurements and corrected them for the RR interval representative of the preceding heart rate, that is, the same RR interval values as used in the individualized corrections ($QTcF_1$ values were obtained in this way), as well as for the RR interval representative of the heart rate of the 10-second ECG sample in which the QT interval measurement was made ($QTcF_2$ values).

Unless explicitly specified, the term *QTc* used in the text refers to the individualized heart rate correction as described above. (The terms $QTcF_1$ and $QTcF_2$ are used for these methodology test data.)

RESULTS

Of the 48 subjects in cohorts on doses A to F, the baseline data of 1 subject (of cohort D) were not available because of a technical recording error made by the clinical unit, and the subject was thus excluded from the analysis presented here. Full ECG data were available from all other subjects.

Figure 2 shows the distribution of standard deviations of baseline QTc values in individual subjects. The mean value was 4.49 ± 1.03 milliseconds (95% CI, 4.19-4.79 milliseconds).

The top panel of Figure 3 shows averaged ΔHR profiles on placebo and on different WXYZ doses. Although there was a heart rate decrease on treatment until approximately 4 hours post dose on day 1, there were no any systematic differences between active doses and placebo, although noticeable variation in ΔΔHR was present (bottom panel of Figure 3).

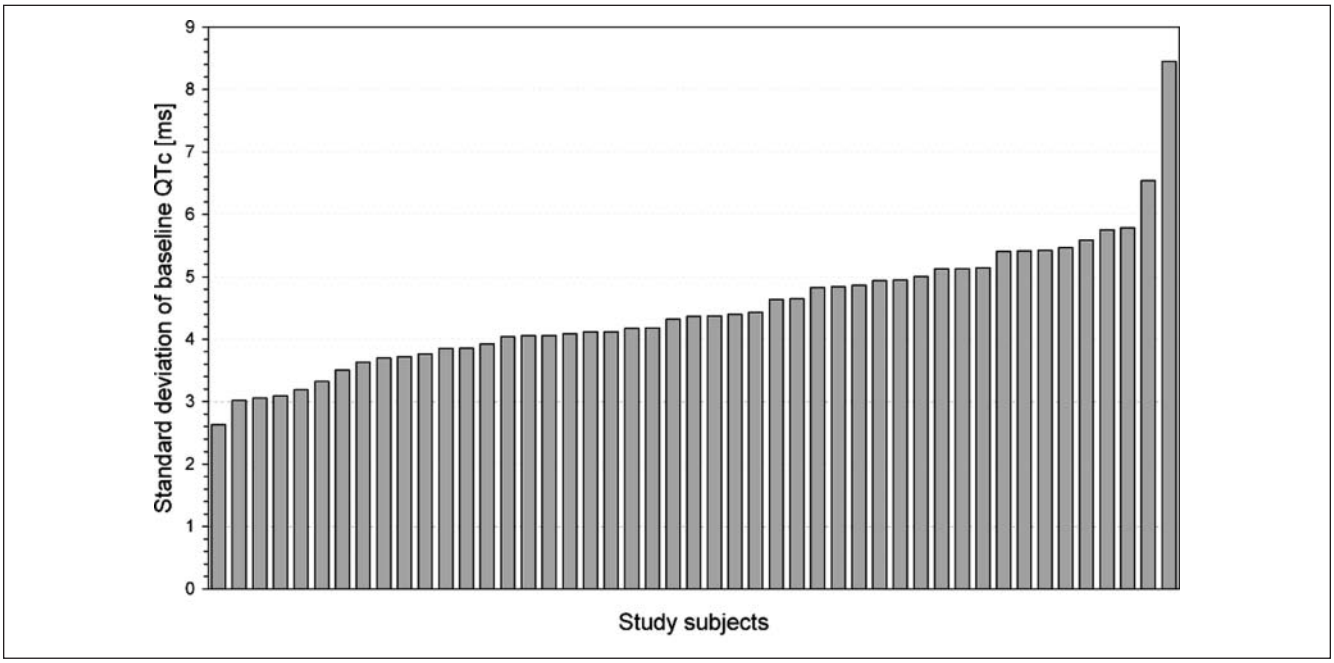

Figure 2 Distribution of standard deviations of QTc values measured in baseline (day –1) recordings of individual study subjects. The standard deviations were calculated from all the measurements made on day –1, that is, both the measurements corresponding to the individual data points and the measurements used to derive the individual QT/RR relationship. (On average, 326 ± 6 QT measurements were used during day –1 in individual subjects.)

Figure 4 shows averaged ΔQTc profile on placebo. There was clear, gradual prolongation of ΔQTc values on placebo of approximately 10 milliseconds between 15 and 135 minutes after dosing.

Figure 5 shows average ΔQTc and ΔΔQTc profiles on individual doses A to F. Note that the ΔQTc profiles replicate the gradual ΔQTc prolongation after dosing that was observed on placebo (most likely attributable to autonomic and psychosomatic conditioning after dosing). The ΔΔQTc profiles show no systematic profile changes and no relationship between QTc changes and drug doses.

Figure 6 shows average ΔQTc and ΔΔQTc profiles on pooled doses A+B, C+D, and E+F. Note in the bottom panel that at all investigated time points, the upper limits of 95% CI are below the 10-millisecond threshold specified in ICH E14 guidance.[1] Thus, when pooling the lowest, middle, and highest 2 doses together, the study passes the so-called intersection union test for the absence drug-induced QTc prolongation according to regulatory guidance.

Figure 7 shows the relationship between WXYZ plasma levels and individual ΔQTc values. The linear regression model predicts a mean 0.6-millisecond QTc interval prolongation per every 1000-ng/mL increase of WXYZ plasma levels. Peaks of group averaged plasma concentrations on doses A to F were found at 2:50, 1:50, 1:20, 1:50, 1:50, and 2:50 hours:minutes after dosing, respectively.

Finally, Figure 8 shows the images corresponding to the bottom panel of Figure 6 obtained with $\Delta\Delta QTcF_1$ and $\Delta\Delta QTcF_2$ values. Figure 8 shows that with Fridericia correction, the variability of the heart rate–corrected QT values is much greater and the stability of the results much lower than with individually corrected QTc. Also, the correction for immediately measured RR interval ($QTcF_2$ values) makes the results even more variable than the correction for the stable preceding heart rate ($QTcF_1$ values).

DISCUSSION

The analysis of ΔΔQTc values shows clearly that single doses of WXYZ did not have any effects on QT-interval duration in the subjects of this study.

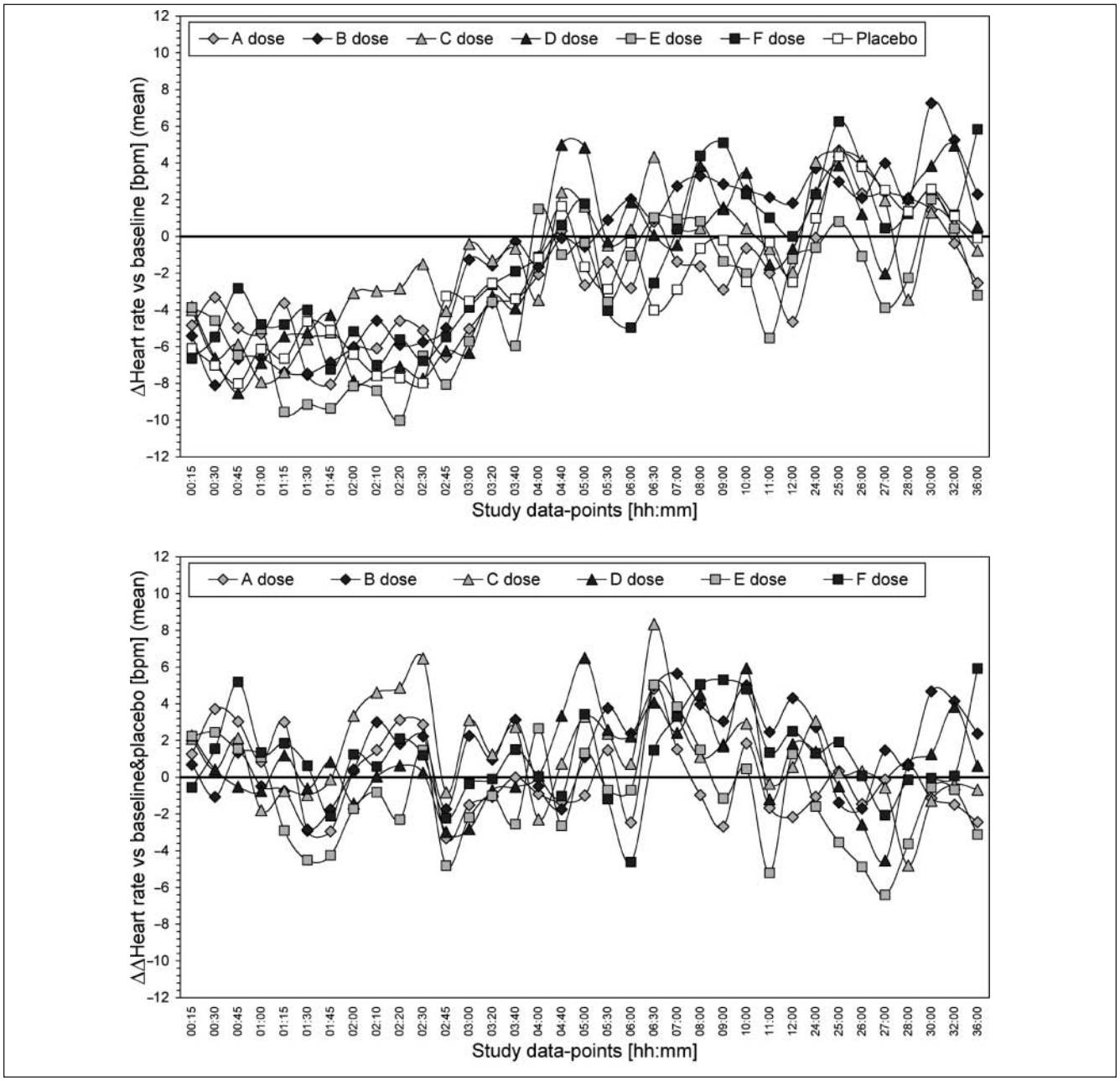

Figure 3. Heart rate changes from baseline (top panel, differences between postdose heart rates and time-matching heart rate values at baseline) and from baseline and placebo (bottom panel, differences between ΔHR on active treatment and on time-matching placebo). Averages for the pooled placebo group and for individual dose groups are shown. bpm, beats per minute.

When the long-term ECGs were obtained accurately and subjected to careful analysis, the very tight threshold of confidence intervals required by the regulatory guidance was satisfied with as few as 12 subjects. The likelihood that single doses of WXYZ have any involvement in cardiac repolarization is therefore very small indeed. Because this ECG analysis was only an add-on to a standard first-in-man, single-ascending-dose study and because the ECG measurement relied, to a substantial degree, on fully

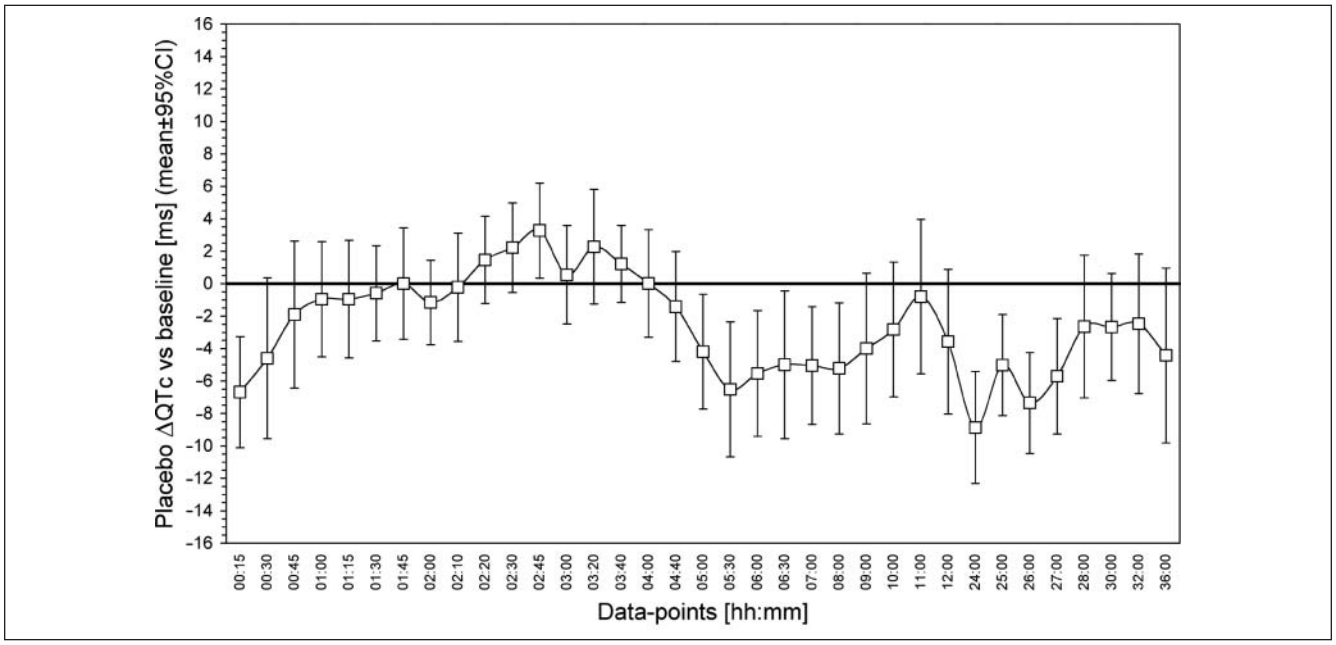

Figure 4. ΔQTc values (differences between postdose QTc values and time-matching QTc values at baseline) in 12 placebo subjects pooled from all 6 cohorts of the study. Double-sided 95% confidence intervals are shown.

automatic ECG processing, these results were obtained for a small fraction of the cost of a standard TQT study.

The success of the ECG support of this first-in-man study, as seen in the very tight confidence intervals of the mean changes as well as in the very small intrasubject variability of baseline QTc intervals, depends on a combination of several factors. These include careful ECG acquisition, systematic checks of heart rate stability before QT-interval measurement, an advanced pattern-matching measurement algorithm, adjustment ensuring systematic measurements of similar ECG morphologies by pattern classification, accurate individual-specific heart rate correction including the estimation of individual QT/RR curvature, and heart rate correction for the preceding stable heart rate rather than for the instantaneous RR-interval measurement.

The reanalysis of the study with Fridericia formula (compare the bottom panel of Figure 6 with the panels in Figure 8) suggests that the technology cannot be substantially simplified without compromising if not entirely spoiling the results. Although using Fridericia formula we obtained $\Delta\Delta QTcF_1$ and $\Delta\Delta QTcF_2$ values still fluctuating around zero, their spread was substantial and the assurance obtained from the accurate analysis with individualized corrections was lost as was the agreement with regulatory guidance. Thus, meaningful and strong data on drug-induced QTc changes can be obtained from very small studies involving as few subjects as was the case here, but the ECG processing technology must be very careful and accurate. Although the Fridericia formula might (in the absence of drug-induced heart rate changes[13]) be acceptable in large studies when averaging over many subjects overwhelms the correction inaccuracies, it is clearly unacceptable for accurate evaluation of small studies such as first-in-man investigations.

The demonstrated ECG support of first-in-man studies in the present implementation naturally does not replace thorough studies. Nevertheless, if similar ECG support were incorporated into a multiple-ascending-dose, first-in-man study and if ECG assay sensitivity were sufficiently established, it is possible that the need for a subsequent, separate TQT study might even be eliminated. A standard TQT study would likely be required subsequently only if the doses used in the first-in-man studies were not sufficiently higher than the expected therapeutic dose or if the duration of the multiple-ascending-dose, first-in-man study was not sufficient to cover the metabolism of the drug.

However, the doses investigated in the first-in-man studies of new drugs are frequently the highest ever administered in the clinical program and, likewise, the

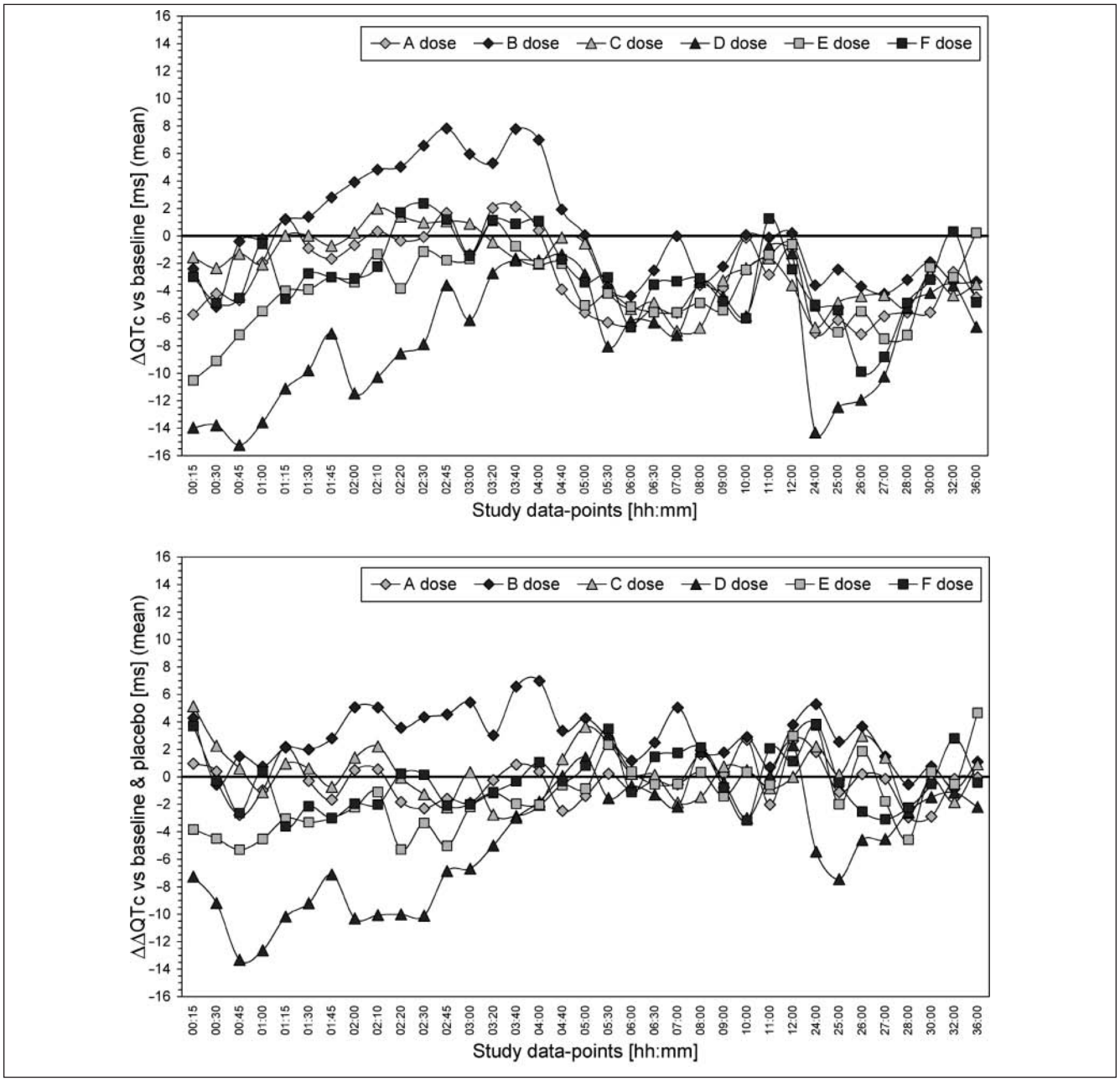

Figure 5. ΔQTc values (top panel, differences between postdose QTc values and time-matching QTc values at baseline) and ΔΔQTc values (bottom panel, differences between ΔQTc values on active drug and time-matched ΔQTc values on placebo) in all 6 active drug cohorts of the study.

duration of the first-in-man, multiple-ascending-dose study frequently covers the pharmacokinetic profile of all known as well as subsequently identified metabolites. Thus, for the majority of new drugs, only the objective proof of assay sensitivity might differentiate the detailed ECG support of the first-in-man studies from a dedicated TQT study. Nevertheless, it should not be difficult to incorporate objective determination of ECG assay sensitivity into the early studies. Prospectively defined ECG influences of provocative maneuvers possibly might be used in single-ascending-dose studies (once sufficient experience with such provocations has accumulated),

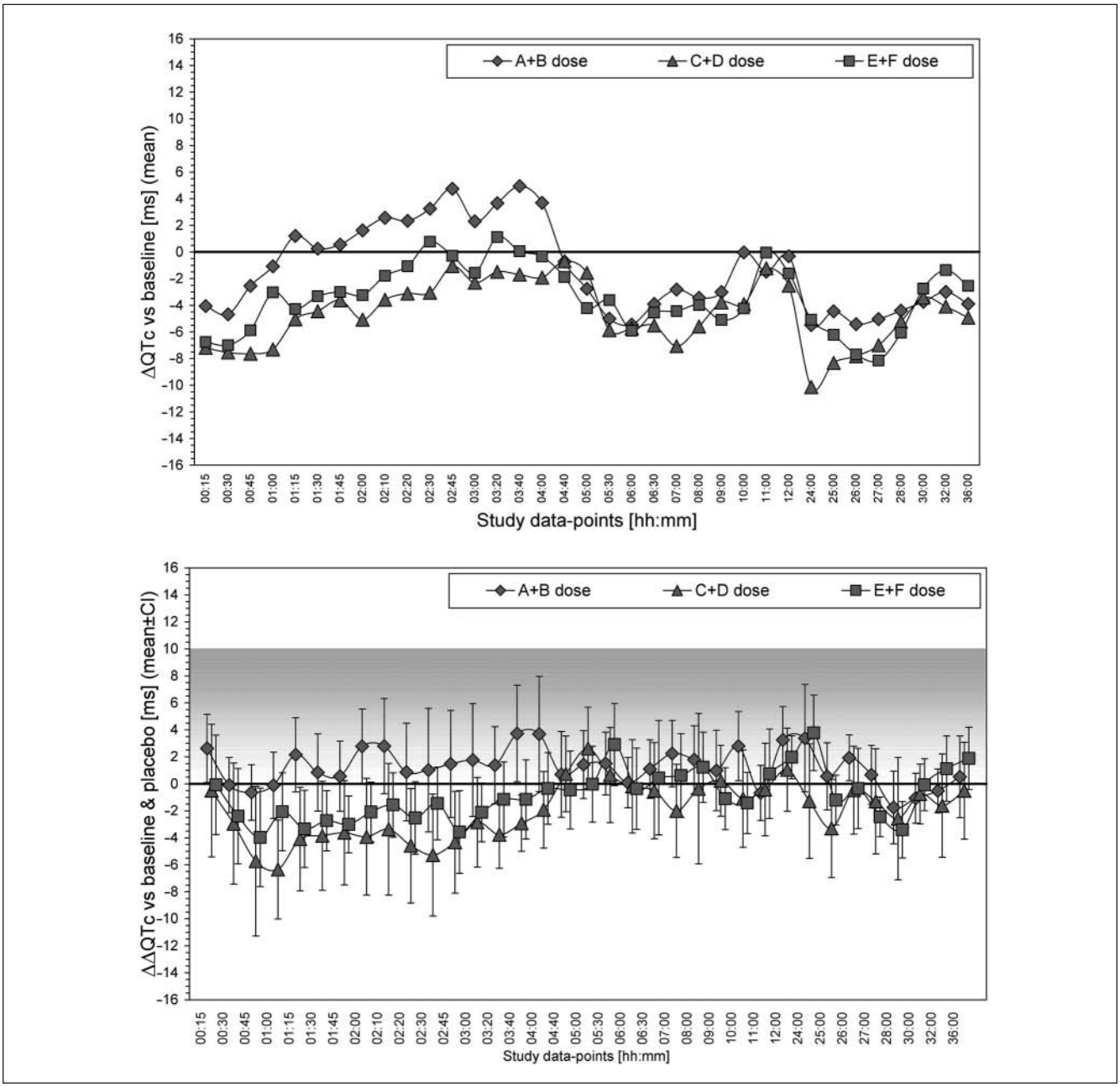

Figure 6. ΔQTc values (top panel) and ΔΔQTc values (bottom panel) for pools of dose cohorts A+B (lowest), C+D (middle), and E+F (highest). Compare with Figure 5. The bottom panel of ΔΔQTc values also shows single-sided 95% confidence intervals (double-sided 90% confidence intervals) that are relevant for the regulatory evaluation of the study. The shaded area shows the regulatory 10-millisecond limit for the upper single-sided 95% confidence intervals.

whereas proper pharmacologic positive control might be incorporated into multiple-ascending-dose studies, preferably not in a separate arm, which would complicate the study design, but in dedicated days of placebo control without interference with the study layout.[14]

Because the precision of this small "near-thorough" QT study (ie, the width of the confidence intervals of QTc changes) fully satisfied the regulatory requirements[1] of the TQT study with comparisons of only 12 subjects on placebo and 12 on active drug, the study

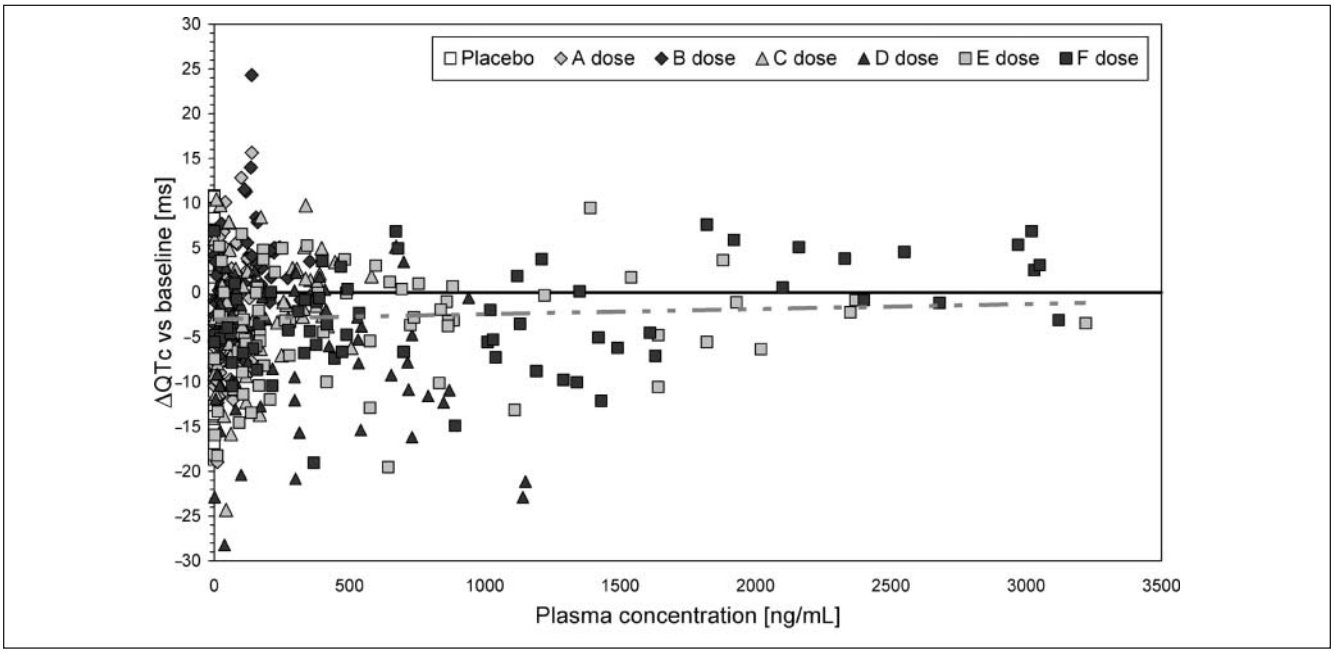

Figure 7. Relationship between drug plasma levels and ΔQTc values measured on placebo (pooled from all 6 cohorts) and on individual active drug doses. The dashed line shows a linear regression model between the plasma levels and ΔQTc values.

also shows that by involving accurate ECG handling, the size of standard TQT studies might be much smaller than is usually the case.[15-18] This is in good agreement with statistical predictions.[19,20]

The analysis of this study also demonstrated the need for a careful placebo control of QT/QTc investigations. As seen in Figures 4 and 5, gradual QTc prolongation after dosing occurred with both placebo and active WXYZ doses. This prolongation was most likely caused by autonomic conditioning with sympathetic overdrive at the time of dosing and a slow return to the baseline sympathovagal balance. Although such autonomic changes might be expected in first-in-man studies, an incorrect conclusion might be reached if the gradual QTc prolongation is not corrected for placebo and if it coincides with the postdose increase of drug plasma levels.

Intentionally, we restricted this text to the study of drug-induced QT-interval changes and did not describe other facets of this first-in-man study of WXYZ. Naturally, many other parameters can be obtained from accurate ECG recordings and processed in a very similar way as the analysis of QTc intervals shown in this text. These parameters include more than the standard ECG measurements, such as PR interval and QRS width. The repeated 5-minute episodes of stable positioning also allow for evaluation of spectral components of heart rate variability[21,22] to investigate drug effects on cardiac autonomic status.

Limitations of the ECG support provided in this study as well as of this text should also be acknowledged. We present the analysis of only 6 cohorts of the original study, which, as is usual with investigations of this kind, included other cohorts. Although the analysis of these other cohorts was fully consistent with the results presented here, the relevant details are not important for the description of the methodology. The selection of automatically measured ECGs for visual verification and correction used a very simple intrasubject classification of provisional QTc values. Such a simple approach might even create some bias. A simple ECG template review might have similar disadvantages. Nevertheless, more advanced morphological classifications that are under development offer much a more precise approach while reducing the number of visual verifications even further. A pattern classification algorithm was used for automatic QT-interval measurements, but comparisons with commercially available software were not made. The experience suggests that if standard commercial algorithms for QT-interval measurement were used, the results

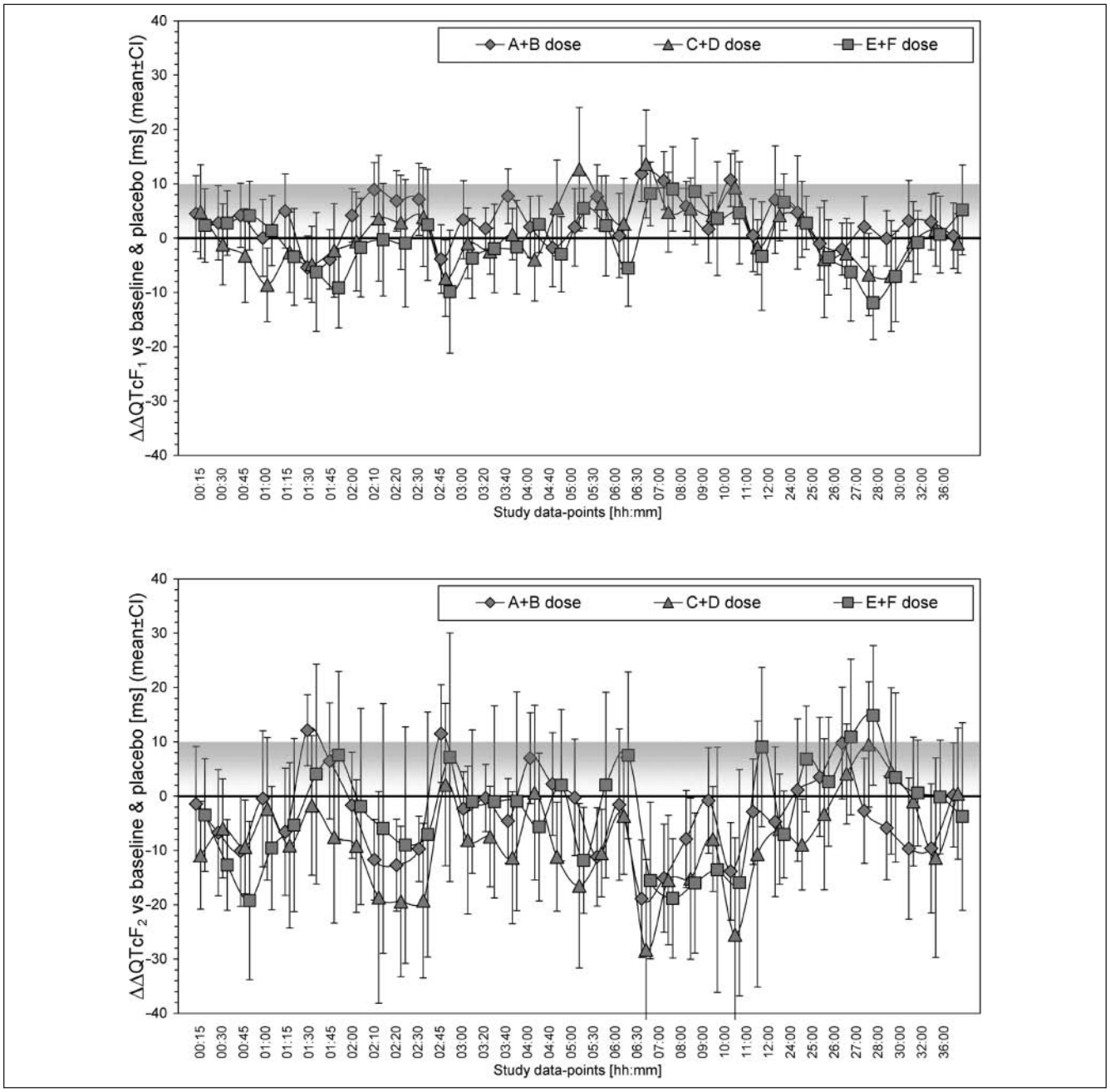

Figure 8. Post hoc reanalysis of the study with Fridericia correction. The top panel ($\Delta\Delta QTcF_1$ values) shows the results with Fridericia correction for preceding stable rate; the bottom panel ($\Delta\Delta QTcF_2$ values) corresponds to Fridericia correction for immediate RR-interval measurement. In both panels, single-sided 95% confidence intervals (double-sided 90% confidence intervals) are also shown. The shaded areas show the regulatory 10-millisecond limit for the upper single-sided 95% confidence intervals. Compare with the bottom panel in Figure 6 and note that whereas in Figure 6, the vertical axis ranges from –16 to +16 milliseconds, the range between –40 to +40 milliseconds had to be used here.

would not have been as stable as presented here.[23,24] Finally, correction for QT/RR hysteresis was not incorporated into the QTc calculations. As recently reported, this correction should be included in further similar projects because it substantially improves the stability of QTc values.[25]

Despite these limitations, the study shows that substantial and accurate ECG data can be obtained in

first-in-man studies (and other small clinical investigations), including assessment of QTc interval changes, the accuracy of which satisfies the confidence interval requirements of regulatory guidance.[1] The evaluation of this very early near-thorough QT study allows investigators to delay the decision of when and how to conduct a true TQT study while providing results at a fraction of the cost of the full thorough investigation.

Financial disclosure: During the clinical study described here, Dr Malik acted as a consultant to Xention, Ltd.

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