Cardiovascular Effects of Dofetilide in Conscious, Telemetered Beagle Dogs

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Figure 1. Effects of Vehicle and Dofetilide on QTcI in Conscious, Telemetered Beagle Dogs

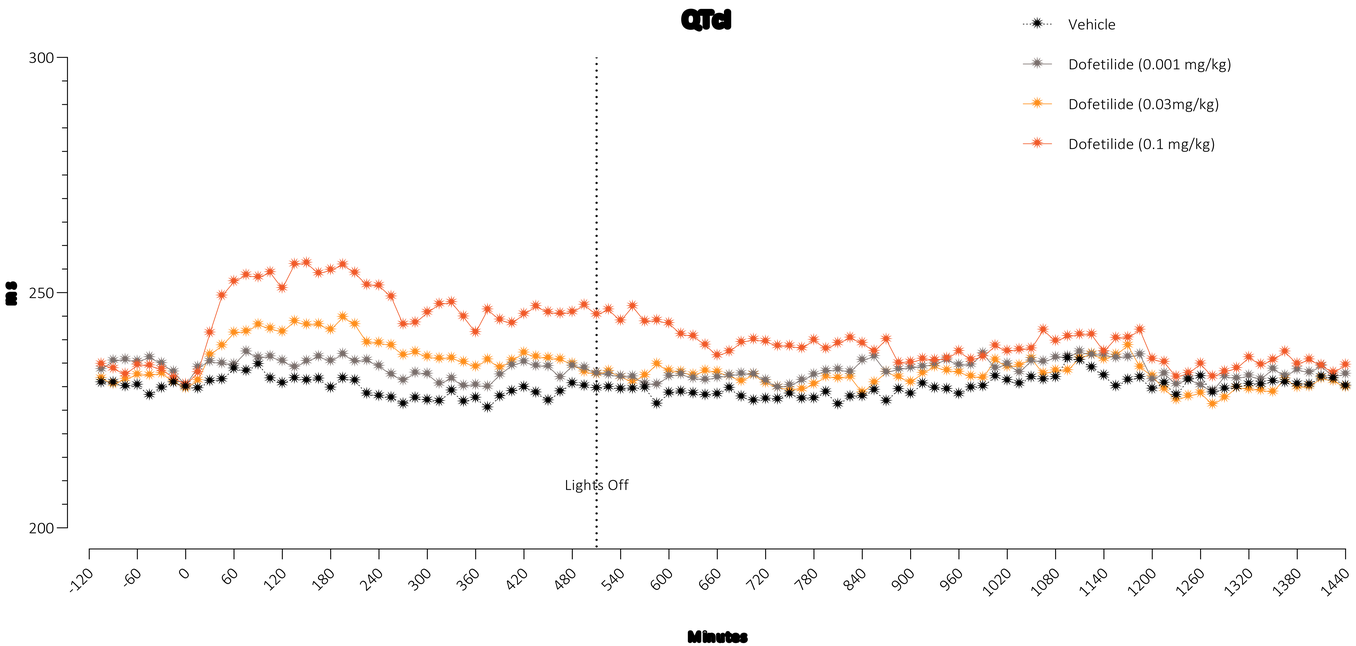


Figure 2. QT Correction Factors vs. HR After Dofetilide Administration in Conscious, Telemetered Beagle Dogs

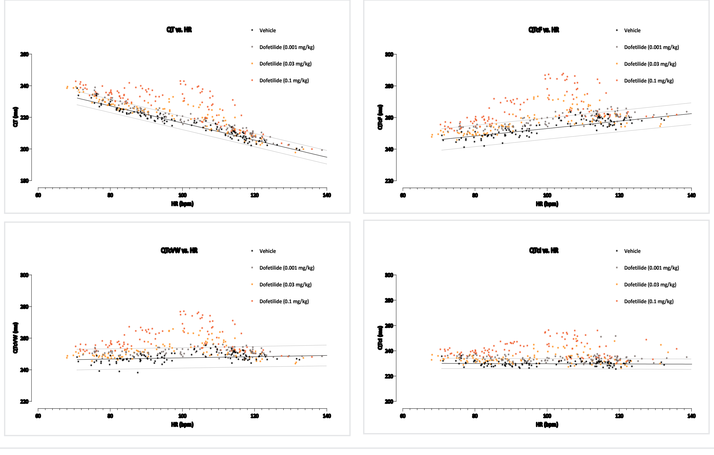
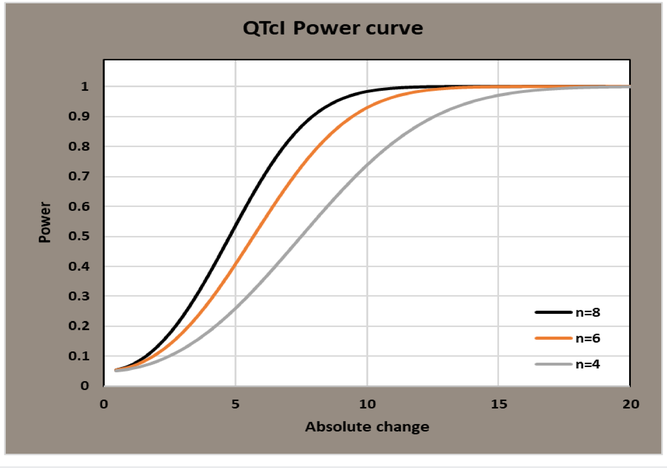


Figure 3. QTcI Power Curve After Administration of Dofetilide (n=8 vs. n=6 vs. n=4)



Introduction

Laboratory specific environmental controls and activities can result in a great deal of variability in data collected at individual test sites. Therefore, it is of great importance for each site to perform verification studies to supplement other validation activities. Well executed verification studies provide researchers, sponsors, and regulatory agencies with a level of confidence in the ability of a study model to pick up changes in important parameters. An integral step when designing a verification study for cardiovascular safety pharmacology is the selection of the dose levels that will be administered. A clear dose response should be considered as a main goal. Additionally, sensitivity of the model should be established using statistical power analysis. For this reason, doses levels that produce subtle changes should be included to provide insight to the range of sensitivity one could expect from a particular study design.

The FDA developed specific guidelines (ICH S7B) to provide strategies for assessing the potential of a test substance to prolong the QT interval. Prolongation of the QT in known to be associated with an increased risk of ventricular tachyarrhythmia and, in some cases, torsade's de pointes (TdP) in humans. TdP is a potentially fatal form of polymorphic ventricular tachycardia associated with a prolonged QT interval.

In accordance with ICH guidelines, we designed this study with the purpose of evaluating the sensitivity and validity of the beagle dog telemetry model at Inotiv to be an effective preclinical predictor of QT prolongation in humans. The canine was selected as it is the most frequently used non-rodent species for studies of pharmaceutical products.

Dofetilide was selected as a positive reference compound as it is a Class III anti-arrhythmic known to cause QT prolongation.

Methods

Eight male beagle dogs that had been previously implanted with digital telemetry devices were used in this study. Digital L11 and L21 devices (Data Sciences International) were implanted onsite at Inotiv. The animals were administered 0.5% methylcellulose in water and dofetilide at doses of 0.001, 0.03 and 0.1 mg/kg using a Latin square dose design. The route of administration for all doses was oral gavage. There was a washout period of 7 days between doses.

Prior to dosing, all animals were acclimated to the room environment for minimum of 2 days. Inherent variability of the model was assessed with the collection of baseline data on 3 separate occasions. All instances of data collection were uninterrupted at ~24 h in duration.

Clinical observations were made using video monitoring equipment in an effort to mitigate room disturbance. Rooms were cleaned and animals were fed each morning prior to data acquisition start.

A Data Sciences Telemetry Recording System with Ponemah (P3) software was used for recording and analysis of the physiological data. Systolic blood pressure (SBP), diastolic blood pressure (DBP), mean arterial pressure (MAP), heart rate (HR), core body temperature (BT), and Lead II ECG were recorded. An ECG waveform morphology assessment was completed for each dog at the 0.1 mg/kg dose level using P3 data insights. Data was collected continuously from at least 2 h prior to dosing through 24 h post dose on each study day. Sampling rates for both ECG and ABP recordings was set to 500 Hz. Derived parameters were automatically calculated by the P3 system.

All telemetry data was organized into phases (8 intervals of 15 minutes) for each 24-hour collection period. Each parameter was analyzed, by phase, with a repeated measure analysis of covariance (RANCOVA). Covariates included baseline data, dosing day, treatment, time after dosing and the interaction of each treatment group and time after dose. SAS ® software was used for the analysis procedures. A Dunnett's test was used to compare the treated groups with vehicle control. For comparative purposes, this statistical analysis was performed using both a 24-hour baseline period collected prior to dose administration and the 2 hours baseline immediately preceding each dosing day.

Retrospective statistical power analysis (RSPA) was conducted on the following parameters: HR, SBP, DBP, MAP, BT, PR Interval, RR Interval, QRS Interval, QT Interval, QTcI, QTcVW and QTcF using three different methods. The first method used all 8 animals from the study design. The second method involved using only 6 animals from the study design (first 3 animals with L21 implant and first 3 animals with implant L11R). The third method involved using only 4 animals from the study design (all animals with L11R implant).

RSPA was conducted, and minimum detectable effect size (changes from control group) were calculated based on observed variance from the analysis model at a power of 0.8 and a significance level of 0.05. A noncentral t distribution was used for the power calculation. For each study parameter, the standard deviation was calculated using the error degrees of freedom for the test of Treatment effect in the statistical model and the median of standard errors for the difference in least square means between treated groups and the control group in all blocks of study.

Results

A dose dependent increase in QTcI intervals was observed as expected (Figure 1). Control values observed during the course of the study matched those of previous historical data collected at the test site. ECG morphology chances after 0.1 mg/kg dofetilide induced several different conduction disturbances with the most common being ectopic ventricular contractions and T-wave morphology changes. A dose response was observed at all 3 dose levels by plotting the relationship between uncorrected QT vs. HR. Of the 4 correction factors analyzed, QTcVW and QTcI most appropriately corrected for HR (Figure 2). Minimum detectable differences were calculated and reported for all parameters as the absolute change that would be detectable with a given statistical power (Table 1). Power curves were generated to show the relationship between sample size (n=8 vs. n=6 vs. n=4). As expected, statistical power increase with sample size. In this study, using n=4 animals, we were able to statistically identify an ~10 ms change in QTcI produced with administration of dofetilide (Figure 3).

Table 1. Retrospective power analysis at 80% power and 0.05 level of significance (Calculated using data derived from n=4 animals)

Parameter	Absolute Change (Minimum Detectable Difference) n=4
SBP (mmHg)	14.4
DBP (mmHg)	9.0
MAP (mmHg)	10.5
HR (bpm)	12.6
QT (ms)	13.7
QTcI (ms)	10.8
QTcV (ms)	9.8
QTcF (ms)	9.8
PR (ms)	4.0
QRS (ms)	2.1

Conclusions

- Data from the 0.5% methylcellulose group matched previous Inotiv historical control data.
- Dofetilide at all 3 dose levels was associated with a significant increase in the QT.
- Conduction disturbances noted after administration of dofetilide were similar to results from similar studies found in scientific literature.
- QTcVW and QTcI provided the most appropriate QT corrections across variable heart rates.
- The n=4 Inotiv telemetry beagle dog model exhibits low inherent intra-animal variability and high sensitivity to detect small but significant increases in all parameters including QT/QT(c) intervals (~10 ms minimum detectable change).