Investigation of individual analysis of QT interval in beagle dogs

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Introduction

In the cardiac assessment with the telemetry system, technological advancement has enhanced accuracy of electrocardiogram (ECG) analysis, and enabled easy processing of a large amount of data. With such advanced technologies, detailed analysis such as 24-hour continuous ECG waveform analysis or individual analysis with QT correction formulas are being discussed. In this research, we investigated applicable data-transaction methods by performing analysis of all ECG waveform and individual analysis on data of dog telemetry along with blood collection. Also, we tested 2 compounds, nifedipine, an expected heart rate accelerator, and sotalol hydrochloride, a known QT-prolonging agent.

Materials and Methods

Experiment 1 (Ex1) Investigation of individual analysis

【Data-collection】
Animal: Telemetry transmitters were implanted into 4 male dogs
ECG lead placement:
ECG electrodes were subcutaneously and anteroposteriorly fixed on the thorax.
Acquisition/analysis software: Open ART HEM 4.3 (Notocord system)
Data acquisition: -2 hr to 24 hr after dosing
Feeding: 8.25 hr postpose
Lighting: 6:00 $_{AM}$ to approx. 15 min after feeding
Dosing time: Approx. 11:00$_{AM}$ (Base-point time for non-treatment control was 11:00)
Study design: Shown in Table1 (EX1)
Blood-sampling method: With vacuum blood-collection tubes, 2 mL of venous blood was taken from the cephalic vein; 1 mL of arterial blood, from the access port
Blood sampling: Predose (−0.5 hr), 0.5, 1, 2, 4, 6, 8, and 24 hr postdose. (except for non-treatment control)
【 Data treatment 】
Measurement time point: Predose (−0.5 hr), 0.5, 1, 2, 4, 6, 8, and 24 hr postdose

Data processing:
For each dog, means and medians per minute were obtained, and 3 patterns of data sets, means of 1-minute means, medians of 1-minutes means, and medians of 1-minute medians, were computed for following duration: for 3, 5, 10, 15, 20, and 30 minutes before each time point.
Measurement parameters: SBP, DBP, MBP, HR, RR, PR, QRS, QT, and QTc
QTc formulas:
◆ Matsunaga's formula (QTcM) : QTc=Log600 × QT/LogRR
◆ Fridericia's formula (QTcF) : QTc= QT/RR$^{1/3}$)
◆ Individual formula (QTca, Spence et al., 1998)[1] : QTc=QT/RR$^{\beta}$, LogQT=α+β × LogRR
QTca: QTc corrected by β value of non-treatment control
QTca2: QTc corrected by β value of each dosing
Calculation of β value:
For each individual and each dosing, LogQT and LogRR were obtained by log-transforming QT and RR intervals, respectively. A correction coefficient for the individual formula, which is the slope of the regression line in plotting the data below, was regarded as the β value.
(1) −2 hr predose – 24 hr postdose (all periods)
(2) −2 hr predose – 8 hr postdose (light period)
(3) 9 hr postdose – 19 hr postdose (dark period)

1: Spence et al., Toxicol Sci 1998 Oct;45(2):247-58.

Experiment 2 (Ex2) Evaluation of positive-control drugs

【Data-collection】
The animals, software, feeding, lighting, measurement parameters, and blood-sampling time points were same as Experiment 1.
Positive drug: Nifedipine and sotalol hydrochloride
Dosing time: Approx. 12:00$_{AM}$ (Base-point time for non-treatment control was 12:00)
Study design: Shown in Table 1 (Ex2)

【Data treatment】
Measurement time points: Predose (−0.5 hr), 0.5, 1, 2, 4, 6, 8, and 24 hr postdose
Data processing:
Data are median values of 1-minute means for 10 minutes
Calculation of β value:
LogQT and LogRR were obtained by log-transforming QT and RR intervals, respectively of the non-treatment control (Period 5). A correction coefficient for the individual formula, which is the slope of the regression line in plotting the data between −2 hr predose and 24 hr postdose, was regarded as the β value.

Table 1 Study design of the experiment

(Ex1)

Animal No.	Period 1	Period 2	Period 3	Period 4
1 - 4	Non-treatment control	Water for injection (1st WFI)	Water for injection (2nd WFI)	Water for injection (3rd WFI)

Dosing interval: After measurement of non-treatment, water for injection was dosed 3 times at an interval of 7 days. Exceptionally, the interval between Periods 3 and 4 was 14 days.

(Ex2)

Animal No.	Period 5	Period 6	Period 7	Period 8
1 - 4	Non-treatment control	0.5% methylcellulose solution (0.5% MC)	Nifedipine 10 mg/kg	Sotalol hydrochloride 10 mg/kg

Dosing interval: After measurement of non-treatment, dosing was performed 3 times at an interval of 7 days.

Results and Discussion

Ex1. Investigation of individual analysis

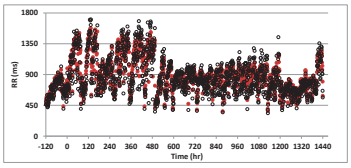

Fig1. Typical RR intervals calculated from means or medians per minute in male dog
Data of Non-treatment animal (No. 3) at 26 hours are shown.
●: 1-minute mean, ○ : 1-minute median

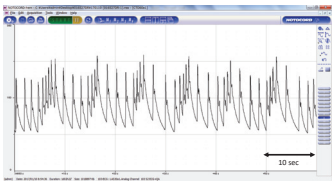

Fig2. Typical respiratory arrhythmia of 1-minute blood pressure waveform in male dog
Data of Non-treatment animal (No. 3) are shown.

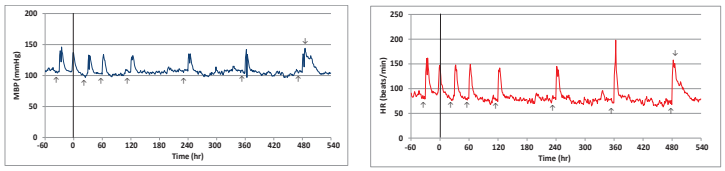

Fig3. Fluctuation of mean blood pressure and heart rates caused by dosing and blood-collection treatment
—: 1-minute mean value of MBP, —: 1-minute means of HR, |: Administration, ↑: Blood sampling ,↓: Feeding
N=12 (3-time dose of water for injection × 4 animals)

✓ For dog-telemetry data, difference in calculation methods of 1-minute values, ie, means or medians, affects the RR interval. It became clear that RR interval values tend to be shortened in use of median values per minute due to respiratory arrhythmia, and so median calculation in primary data treatment appeared to be inappropriate for QT correction.
✓ It became clear that it takes at least 15 minutes for blood-pressure and heart-rate values to return to the normal range in a dog telemetry assessment incorporating blood collection. Analysis by compressing 1-minute means into 10-minute medians minimizes effects of blood-collection treatment and artifacts.

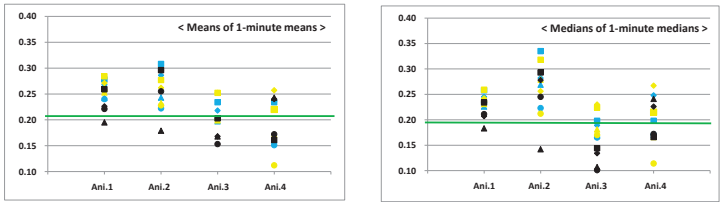

Fig4. Diurnal variation of rate-correction β values derived from QT-RR plots in male dogs
Ani.: Animal, —: Non-treatment control mean (N=4), ●: Light &Dark (Non-treatment control), ▲: Light &Dark (1st WFI), ■: Light &Dark (2nd WFI), ♦: Light &Dark (3rd WFI), ●: Light (Non-treatment control), ▲: Light (1st WFI), ■: Light (2nd WFI), ♦: Light(3rd WFI), ●: Dark (Non-treatment control), ▲: Dark (1st WFI), ■: Dark (2nd WFI), ♦: Dark (3rd WFI)

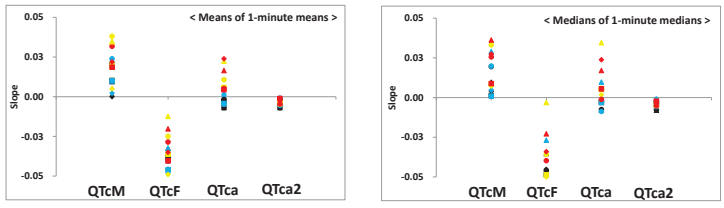

Fig5. Comparison of regression line slope derived from QTc-RR plots in male dogs
●: Ani.1 (Non-treatment control), ●: Ani.1 (1st WFI) , ●: Ani.1 (2nd WFI), ●: Ani.1 (3rd WFI), ▲: Ani.2 (Non-treatment control), ▲: Ani.2 (1st WFI), ▲: Ani.2 (2nd WFI), ▲: Ani.2 (3rd WFI) ■: Ani.3 (Non-treatment control), ■: Ani.3 (1st WFI), ■: Ani.3 (2nd WFI), ■: Ani.3 (3rd WFI), ♦: Ani.4 (Non-treatment control), ♦: Ani.4 (1st WFI), ♦: Ani.4 (2nd WFI) , ♦: Ani.4 (3rd WFI)

✓ Among the tested QT-correction formulas, QTca2, in which correction reflects a β value for each dosing, is the highest in correction capability. The capability of the other formulas is high in this order: QTca, QTcM, and QTcF. Evaluation of unknown compounds is possible with high degree of accuracy by QTca whose capability was close to that of QTCa2.

Ex2. Evaluation of positive-control drugs

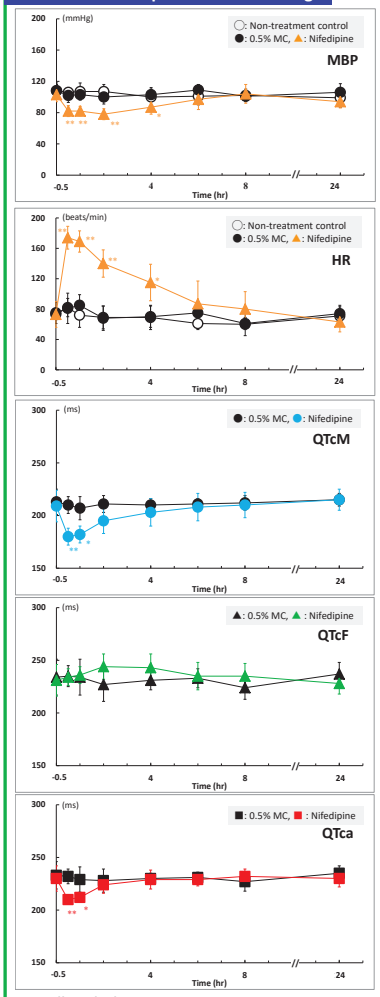

Fig6. Effect of nifedipine on mean blood pressure, heart rate, and electrocardiogram in male dogs
Each value represents the mean ± S.D. of 4 dogs
*: P<0.05, **: P<0.01; Significant difference from 0.5% MC (Student t test)

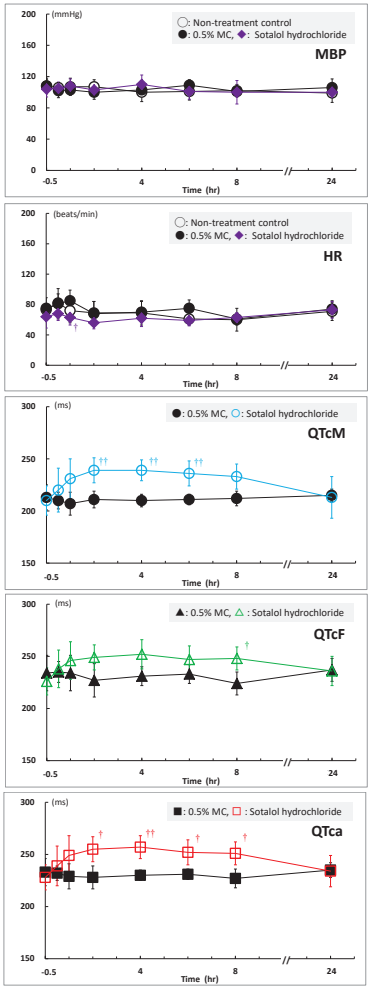

Fig7. Effect of sotalol hydrochloride on mean blood pressure, heart rate, electrocardiogram and in male dogs
Each value represents the mean ± S.D. of 4 dogs
†: P<0.05, ††: P<0.01; Significant difference from 0.5% MC (Student t test)

✓ As for QT interval for nifedipine, the prolongation tendency was detected by QTcF. QTca showed insufficient correction in a high heart-rate range, which did not develop in non-treatment, despite good correction capability was confirmed in cases of moderate heart-rate increase. However, QTca still seems most appropriate for evaluation of heart-rate accelerators since QTca was least affected by heart-rate increase among the tested QTc formulas.
✓ As for QT interval for sotalol hydrochloride, prolongation was detected by all the correction formulas, without any apparent difference by correction method.

Conclusion

In this research, we found out that use of means per minute suites better than medians per minute for individual analysis of QT interval in dogs. Blood collection during the data collection did not interfere with the analysis. The individual analysis was applicable for all the correction formulas studied this time, and the prolongation effect of a known QT-prolonging agent was detected without any apparent difference among formulas. On the other hand, for heart-rate accelerators, it was indicated that the individual correction formula QTca would contribute to high-accuracy correction by minimizing effects of heart-rate increase.

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