Differentiating electrophysiological effects and cardiac safety of drugs based on the electrocardiogram: A blinded validation

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BACKGROUND The ventricular components (QRS and QT) on the electrocardiogram (ECG) depend on the properties of ventricular action potentials that can be modulated by drugs via specific ion channels. However, the correlation of ECG ventricular waveforms with underlying ion actions is not well established and has been extensively debated.

OBJECTIVE To conduct a blinded in vitro assessment of the ionic mechanisms for drug-induced ECG changes.

METHODS AND RESULTS Fourteen cardiac and noncardiac drugs with known effects on cardiac ion channels were selected by the study sponsor, and were tested in the rabbit left ventricular wedge preparation with recording of the ECG and contractility. The investigators who performed the experiments and analyzed the data were blinded to names, concentrations, and molecular weights of the drugs. The compounds were prepared by the sponsor and sent to the investigators as 56 stock solutions. The effects of I_{Kr}, I_{Ks}, $I_{Ca,L}$, I_{Na} blocker, and I_{KATP} opener on QRS, QT, and $T_{p\text{-}e}$, were evaluated. Disclosure of the names and concentrations after completion of the study revealed that there were highly correlated ECG changes with underlying ionic mechanisms and proarrhythmic potential of drugs that, respectively, target I_{Kr}, I_{Ks}, $I_{Ca,L}$, I_{Na}, and I_{KATP}. Among ECG parameters, $T_{p\text{-}e}$ was more useful in differentiating drugs' actions.

CONCLUSIONS Specific electrophysiological action and the consequent proarrhythmic potential of a drug can be accurately determined by analysis of drug-induced changes in ECG in the rabbit left ventricular wedge preparation. Change in $T_{p\text{-}e}$ provides the most relevant information.

KEYWORDS QT; $T_{p\text{-}e}$; $T_{p\text{-}e}$/QT ratio; proarrhythmias; ionic channels; I_{Kr}; I_{Ks}; $I_{Ca,L}$; I_{Na}; I_{KATP}

ABBREVIATIONS ECG = electrocardiogram; $\mathbf{I_{Ca,L}}$ = inward L-type calcium current; $\mathbf{I_{KATP}}$ = ATP-sensitive potassium current; $\mathbf{I_{Kr}}$ = delayed rectifier potassium current with its rapid; $\mathbf{I_{Ks}}$ = slow component potassium current; $\mathbf{I_{Na}}$ = inward sodium current; **LV** = left ventricular; **TdP** = torsades de pointes; **TDR** = transmural dispersion of repolarization

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Introduction

The transmembrane action potential of ventricular myocytes functions as an electromotive generator for the ventricular waveforms on the body surface electrocardiogram (ECG) that extend from the QRS complex (depolarization) to the T wave (repolarization).[1] The cardiac action potential is the consequence of a dynamic balance between inward and outward currents across the cell membrane, which adapt to physiological demands and pathophysiological stresses, and can be altered by a variety of cardiac and noncardiac drugs. In the ventricles, major ion currents participating in depolarization and repolarization processes include the inward sodium current (I_{Na}), inward L-type calcium current ($I_{Ca,L}$), and delayed rectifier potassium current with its rapid (I_{Kr}) and slow component (I_{Ks}). However, correlation of individual ventricular repolarization waveforms on ECG with underlying ion and cellular mechanisms is not well established and has been a matter of intense debate.[2,3]

In 1996, Yan and Antzelevitch[4] first developed an arterially perfused ventricular wedge preparation, in which a transmural ECG can be recorded simultaneously with transmembrane action potential across the ventricular wall, in an attempt to establish the cellular and ionic basis for the repolarization waveforms on the ECG.[1,4,5] Data obtained in this model during the past one and half decades have greatly enhanced our understanding of ventricular waveforms on the ECG, specifically their physiological significance and role in arrhythmogenesis associated with a wide variety of sudden cardiac death syndromes.[1,2,4,6] One of the key findings is the presence of 3 cell types (epicardium, M cells, and

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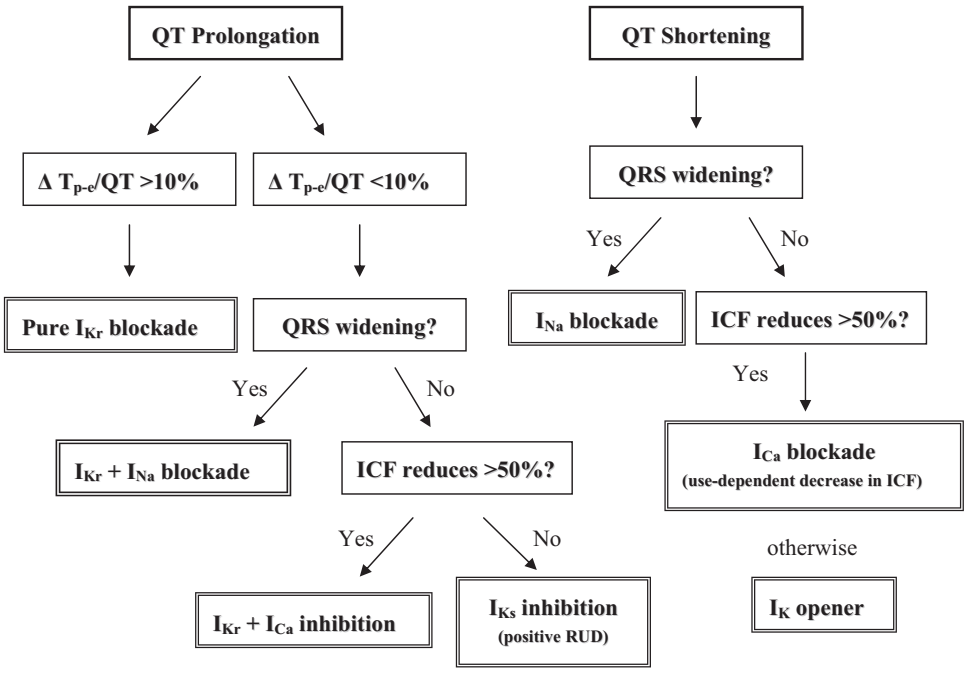

Figure 1 This schematic depicts our interpretation of the rabbit left ventricular wedge data to determine the ionic basis of drug-induced changes on the electrocardiogram and contractility. ICF = isometric contractile force; RUD = reverse use-dependence.

endocardium) with different repolarization characteristics across the ventricular wall of the wedge preparation in which all the myocytes are electrically coupled.[2,5] Heterogeneous dispersion of repolarization across the ventricular wall is responsible for registration of the T wave on ECG and can be quantitatively defined by the interval from the peak to the end of the T wave ($T_{p\text{-}e}$).[2,5] The rabbit left ventricular (LV) wedge data, of which the transmural dispersion of repolarization (TDR) represented by $T_{p\text{-}e}$ and $T_{p\text{-}e}$/QT ratio is a key component, have been found to be useful in predicting drug-induced QT prolongation and torsades de pointes (TdP) liabilities.[7,8]

However, debate over whether the wedge data particularly related to TDR can be directly applied to the intact heart and clinical ECG has been rekindled.[2,3] Therefore, we believe it is important to further validate the concepts derived from the ventricular wedge preparation in a different approach from that of the previous studies. In previous studies, drugs with well-established ionic actions and clinical profiles were used to delineate the ionic mechanisms for a variety of clinically important ECG phenomena.[1,5,9] In contrast, this study was designed specifically to differentiate the ionic mechanisms of a number of "unknown" cardiac and noncardiac drugs based on ECG changes induced by each individual drug. This validation study used the rabbit LV wedge preparation and was conducted by investigators who were blinded to the tested drugs. Our objectives were to (1) test clinical prediction of the electrophysiological concepts established from the wedge preparation in drug research and development; (2) correlate changes in ECG waveforms with underlying ionic mechanisms, which may be potentially translated into a better understanding of the ionic basis for human ECG waveforms; and (3) establish changes in the ECG associated with proarrhythmic potential of commonly used drugs.

Methods

Arterially perfused rabbit LV wedge preparations

Surgical preparation of the rabbit LV wedge and its electrophysiological recordings have been described in detail in previous publications.[7,9] The preparation was placed in a small tissue bath and arterially perfused with Tyrode's solution containing 4 mM K^+ buffered with 95% O_2 and 5% CO_2 (temperature: 35.7°C ± 0.1°C; mean perfusion pressure: 35–45 mm Hg).

A transmural ECG signal was recorded. The QT interval was defined as the time from the onset of the QRS to the point at which the final downslope of the T wave crossed the isoelectric line. The $T_{p\text{-}e}$ interval was defined as the time from the peak to the end of the T wave. A force transducer was connected to the wedge preparation for recording contractility.

Study protocols

Fourteen compounds were prepared by the sponsor as 56 (4 × 14) dimethyl sulfoxide (DMSO) stock solutions in which each stock solution was used for a specific concentration and was sufficient for only 4 wedge experiments. Each compound was tested at 4 different concentrations in 4 wedge preparations. The compound's identity, concentration range, molecular weight, and order of testing were unknown to the investigators performing the experiments and analyzing the results.

After a 1-hour equilibration period during which the preparation achieved electrical stability,[7] each preparation was exposed to each of 4 drug concentrations for a period of 20 minutes in a concentration escalation manner prior to the beginning of data collection. Three basic cycle lengths of 500, 1000, and 2000 ms were used to pace the preparation from the endocardial surface.