Tedisamil and dofetilide-induced torsades de pointes, rate and potassium dependence

¹Terrance D. Barrett, ¹James K. Hennen, ²Peter S. Fischbach, ¹Brian P. O'Neill, ¹Edward M. Driscoll Jr. & ^{*,1}Benedict R. Lucchesi

¹Department of Pharmacology, University of Michigan Medical School, Ann Arbor, Michigan, MI 48019-0632, U.S.A. and

²Department of Pediatrics and Communicable Diseases, Division of Pediatric Cardiology, University of Michigan Medical School, Ann Arbor, Michigan, MI 48019-0632, U.S.A.

1 Tedisamil is a bradycardiac agent that prolongs the QT interval of the ECG and prevents cardiac arrhythmias. Given this profile, tedisamil might be expected to have proarrhythmic actions similar to Class III antiarrhythmic drugs. To address this question, the actions of dofetilide and tedisamil were examined in rabbit isolated hearts in which bradycardia was induced by AV ablation.

2 The QT interval was prolonged in a reverse rate-dependent fashion by dofetilide (3 and 30 nM) and tedisamil (0.3 and 3 μ M).

3 Torsades de pointes was observed in 1/7 hearts treated with 3 nM dofetilide and 0/7 hearts treated with 0.3 μ M tedisamil. The incidence of torsades de pointes was increased to 5/7 in hearts treated with 30 nM dofetilide and to 7/7 in hearts treated with 3 μ M tedisamil (both $P < 0.05$ vs control).

4 The actions of 30 nM dofetilide and 3 μ M tedisamil were also examined in hearts paced at 50, 100, 200 and 50 beats min^{-1} successively. Both drugs caused torsades de pointes in 5/5 hearts paced at 50 beats min^{-1} ; however, the incidence was reduced to 0/5 during pacing at 200 beats min^{-1} . Thus, drug-induced proarrhythmia was bradycardia-dependent.

5 Drug-induced prolongation of the interval between the peak and end of the T-wave (QTa-e) was reverse rate-dependent and was associated with the occurrence of torsades de pointes ($r = 0.91$, $P < 0.01$).

6 The results suggest that tedisamil, like dofetilide, presents a risk for development of torsades de pointes.

British Journal of Pharmacology (2001) **132**, 1493–1500

Keywords: Torsades de pointes; proarrhythmia; QT prolongation; Class III; tedisamil; dofetilide

Abbreviations: I_{Kr} , rapidly activating component of the delayed rectifier K^+ current; QTa-e, interval between the peak and end of the T-wave

Introduction

There is a continuing need for antiarrhythmic drugs that are both safe and effective. Drugs classed by the Vaughan-William's scheme as Class III prolong cardiac action potentials and, therefore, the QT interval of the ECG. It is clear that this class of drugs can have useful antiarrhythmic actions; however, the proarrhythmic actions of such drugs severely limit their utility. For example, ibutilide (Stambler *et al.*, 1997; Oral *et al.*, 1999), dofetilide (Frost *et al.*, 1997; Torp-Pedersen *et al.*, 1999) and sotalol (Kehoe *et al.*, 1990) are associated with a significant risk of torsades de pointes. In contrast, the risk of torsades de pointes with amiodarone is regarded as being low despite its Class III profile (Connolly, 1999). Unfortunately, pulmonary toxicity limits the use of this drug to high-risk patients. Consequently, there is a continuing need for antiarrhythmic drugs.

Tedisamil is an experimental bradycardic agent being developed for the treatment of angina pectoris (Fox *et al.*, 2000). The drug prolongs cardiac action potentials and the QT interval of the ECG in experimental animals (Beatch *et al.*,

1991; Tsuchihashi & Curtis, 1991; Rees *et al.*, 1993; Wallace *et al.*, 1995; Chi *et al.*, 1996) and in man (Bargheer *et al.*, 1994; Fox *et al.*, 2000). Tedisamil brings about these effects *via* blockade of several cardiac ion currents. These include the delayed rectifier K^+ current (Dukes & Morad, 1989; Dukes *et al.*, 1990), transient outward current (Dukes & Morad, 1989; Dukes *et al.*, 1990), the ATP-dependent K^+ current (Bray & Quast, 1992) as well as the fast inward sodium current (Beatch *et al.*, 1991; Nemeth *et al.*, 1996) and the cyclic AMP-activated Cl^- current (Faivre *et al.*, 1998). Experimental studies have demonstrated that tedisamil has antiarrhythmic actions *in vitro* (Tsuchihashi & Curtis, 1991; Chi *et al.*, 1996) and *in vivo* against arrhythmias arising from the atrium (Fischbach *et al.*, 1999) and ventricle (Beatch *et al.*, 1991; Friedrichs *et al.*, 1996; 1998).

Despite its demonstrated antiarrhythmic effectiveness, it is not clear if tedisamil carries with it a similar risk of torsades de pointes to that observed with Class III antiarrhythmic drugs. The aim of the present study was to examine the actions of tedisamil under conditions where torsades de pointes might be expected to occur (i.e., bradycardia and low $[K^+]$) in a rabbit isolated heart model. The QT prolonging

*Author for correspondence; E-mail: benluc@umich.edu

and proarrhythmic actions of tedisamil were compared to dofetilide. Dofetilide is known to prolong the QT interval of the ECG and precipitate torsades de pointes in experimental animals (Buchanan *et al.*, 1993; Bril *et al.*, 1996) and man (Frost *et al.*, 1997; Torp-Pedersen *et al.*, 1999).

Methods

Guidelines for animal research

The procedures used in this study were in accordance with the guidelines of the University of Michigan Committee on the Use and Care of Animals. Veterinary care was provided by the University of Michigan Unit for Laboratory Animal Medicine. The University of Michigan is accredited by the American Association of Accreditation of Laboratory Animals Health Care, and the animal care use program conforms to the standards in The Guide for the Care and Use of Laboratory Animals, Department of Health, Education, and Welfare Publication Number (National Institute of Health) 86-23.

General preparation

Male, New Zealand White rabbits (2–3 kg), were stunned by cervical dislocation. The heart was rapidly removed and mounted on a Langendorff apparatus. Hearts were perfused with a modified Krebs-Henseleit buffer at a constant flow of 20–30 ml min⁻¹ such that the coronary perfusion pressure was 50–70 mmHg. The composition of the buffer (in mM) was: CaCl₂ 1.8, KCl 2.8, KH₂PO₄ 1.2, MgSO₄ 0.83, NaCl 118, NaHCO₃ 24.88, Na pyruvate 2 and glucose 5.55. The final [K⁺] was 4 mM. The buffer was bubbled continuously with 95% O₂/5% CO₂. The heart was maintained at a constant temperature of 37°C using a circulating water bath. An incision was made in the right atrium and the AV node was ablated using forceps. This procedure produced rapid and irreversible AV block and resulted in a low ventricular rate. Pacing leads were inserted into the intraventricular septum and, in the event that heart rate fell below 50 beats min⁻¹, pacing was initiated at this rate. The average resulting ventricular rate was 50 ± 2 beats min⁻¹. An incision was made in the left atrium to allow insertion of a temperature probe and a balloon into the left ventricle. The intraventricular balloon was filled with saline to give an end diastolic pressure of 5 mmHg. A volume conducted ECG was recorded as described by Zabel *et al.* (1997). This permitted simulated leads V1, V3, V5 and lead II to be recorded simultaneously. The ECG leads, coronary perfusion pressure and left ventricular pressure were monitored using Grass Instruments data acquisition hardware (Models 15AX54 and 15AX12, Quincy, MA, U.S.A.) and Polyview software (version 2.1, Grass Instruments, Quincy, MA, U.S.A.). Hearts were allowed 20 min to equilibrate before starting the experiment.

Experimental protocol for concentration-response studies

The protocol was adapted from that described by Johna *et al.* (1998). Hearts were monitored for 5 min before starting an infusion of vehicle control (0.2% dimethylsulphoxide

dissolved in buffer) into a sidearm on the perfusion apparatus. The infusion rate was 20–30 ml h⁻¹ (i.e., 1/60th of the coronary flow rate). After 20 min, the perfusate was switched to a low potassium buffer ([K⁺] 3 mM) for 5 min. Thereafter, the heart was perfused with normal buffer ([K⁺] 4 mM) for 5 min. To assess rate-dependent changes in the QT interval, hearts were paced at 80, 120 and 240 beats min⁻¹ for 1 min, a time sufficient to account for the effects of restitution on the QT interval. In the event that the pacing protocol was interrupted by the occurrence of tachyarrhythmias, the pacing duration was extended such that each heart was paced at a constant rate for at least 1 min. QT intervals were also measured during perfusion with 3 and 4 mM [K⁺] buffer at a pacing rate of 50 beats min⁻¹ or during spontaneous junctional rhythm (depending on which rate was greater).

Hearts then were assigned to receive either continued infusion with vehicle control, dofetilide at successively increasing concentrations of 3 and 30 nM, or tedisamil at concentrations of 0.3 µM, followed by 3 µM. Drug solutions were made up at 60 times the desired concentration and infused at 1/60th of the coronary flow rate into a sidearm on the perfusion apparatus. The protocol described above was repeated at each concentration of the drug. Thus, each heart was treated with two test concentrations of vehicle control, dofetilide or tedisamil.

Experimental protocol for heart rate response studies

In a separate group of hearts, the rate-dependent and pause-dependent proarrhythmic actions of dofetilide and tedisamil were explored. Rabbit isolated hearts were set up as described above. Perfusate [K⁺] was maintained at 4 mM throughout this protocol. Hearts were paced for 20 min at each of 50, 100, 200 and 50 beats min⁻¹, successively. Pauses were inserted in the basic pacing train at various times to assess the occurrence of pause-dependent torsades de pointes (see below for definition). Every 10 min, three pauses were inserted in the basic train. In addition, three pauses were inserted within the first minute at each new pacing rate. The duration of the pause varied between 2–6 s, depending on the rate of the spontaneously junctional rhythm of the heart. In this way, the spontaneous occurrence of torsades de pointes could be assessed during the basic pacing protocol and the occurrence of pause-dependent torsades de pointes could be assessed by introducing a pause in cardiac activity. Hearts were assigned to treatment with vehicle control, 30 nM dofetilide, or 3 µM tedisamil. Infusions were carried out as described above. QT intervals were measured 19 min after commencing pacing at each rate (i.e., just before introducing the final pause in cardiac activity at each pacing rate).

Assessment of serum [K⁺] in conscious rabbits

A blood sample was obtained from the marginal ear vein of 35 rabbits before starting the protocol. Blood samples (1 ml) were heparinised and spun at 4000 × g for 10 min. The [K⁺] of the plasma was measured using a potassium selective electrode (Model 16.40, Instrumentation Laboratory, Lexington, MA, U.S.A.).

Drugs

Tedisamil was obtained from Kali-Chemi (Hanover, Germany) and dofetilide was obtained from Pfizer (Sandwich, U.K.). The test concentrations of tedisamil were selected on the basis of previous studies in our laboratory (Chi *et al.*, 1996; Friedrichs *et al.*, 1998), as well as others (Tsuchihashi & Curtis, 1991), which demonstrated that tedisamil had antiarrhythmic actions over this concentration range (0.3–3 μM) *in vitro* and *in vivo*. The lowest concentration of dofetilide (3 nM) was selected on the basis of a literature report that demonstrated the antiarrhythmic activity of this drug in man (Echt *et al.*, 1995). The highest concentration of dofetilide was intended to be supra-maximal, such that a high incidence of torsades de pointes could be observed (i.e., 30 nM dofetilide served as a positive control).

Definition of end points

The QT interval was measured from the Q wave, or the onset of the R wave if a Q wave was not present, to the point where a tangent drawn to the steepest portion of the T wave crossed the isoelectric line. Other less conventional measures of repolarization time were obtained from the ECG. The QTa interval was measured from the Q wave, or the onset of the R wave if a Q wave was not present, to the peak of the T wave. The interval between the peak and the end of the T wave (QTa-e) was measured as an indicator of the homogeneity of the repolarization process (Antzelevitch *et al.*, 1999). All of the above noted measures were made from lead II of the ECG from three successive beats.

For the purposes of this study, torsades de pointes was defined as a 'polymorphic ventricular tachycardia characterized by an onset with abnormal QT-prolongation and/or abnormal TU complexes, the electrocardiographic configuration of a progressively changing ventricular axis, and spontaneous termination with the exception of rare degeneration into ventricular fibrillation', as recently described by Eckardt *et al.* (1998). A minimum of four beats was required to diagnose torsades de pointes. As the ECG morphology of torsades de pointes is known to depend on the lead considered, monitoring multiple ECG leads was necessary to ensure accurate diagnosis. In the heart rate response studies, pause-dependent torsades de pointes was said to occur if torsades de pointes occurred within 1 min after a pause in cardiac activity.

Statistics

All data are summarized as the mean $\pm$ s.e.mean, $n=4-7$ in each group. At the higher test concentration of each drug and at low pacing rates, QT intervals could not be measured due to the occurrence of premature ventricular beats and torsades de pointes. Repeated measures analysis of variance (ANOVA) was used to test for statistical significance and a Tukey test was used to detect differences between groups. All statistical tests were two tailed and a value of $P<0.05$ was considered statistically significant. The incidence of torsades de pointes was analysed using a Chi Square analysis.

Results

QT prolonging and proarrhythmic actions of dofetilide and tedisamil

The QT interval was longer at lower pacing rates ($P<0.05$) before drug treatment in all groups (Figure 1). Both dofetilide and tedisamil exacerbated this effect and QT intervals were prolonged in a concentration- and reverse rate-dependent fashion relative to vehicle control (Figure 1). The QTa-e interval did not vary with heart rate in vehicle treated hearts but was prolonged in a concentration- and reverse rate-dependent fashion by both dofetilide and tedisamil (Figure 2). Drug-induced QT prolongation was not influenced by perfusion with low $[\text{K}^+]$ buffer (3 mM $[\text{K}^+]$, Figure 3).

The incidence of dofetilide and tedisamil-induced torsades de pointes was concentration-dependent (Figure 4, Table 1). Torsades de pointes was observed in 1/7 hearts treated with 3 nM dofetilide and 0/7 hearts treated with 0.3 μM tedisamil. In hearts treated with either 30 nM dofetilide or 3 μM tedisamil, torsades de pointes was observed in 7/7 hearts ($P<0.05$ vs vehicle control). In drug-treated hearts, torsades de pointes occurred at various times during the 20 min observation period at 4 mM $[\text{K}^+]$. During the 5 min observation period in hearts perfused with 3 mM $[\text{K}^+]$, torsades de pointes occurred in 3/7 hearts treated with either 30 nM dofetilide or 3 μM tedisamil. Perfusion of hearts with low $[\text{K}^+]$ buffer (3 mM) precipitated torsades de pointes in two hearts treated with 30 nM dofetilide in which it did not occur during perfusion with normal $[\text{K}^+]$ buffer (Table 1). Torsades de pointes was not observed in vehicle-treated hearts and ventricular fibrillation was not observed in any group.

QT prolonging and proarrhythmic actions of dofetilide and tedisamil; effect of heart rate

The QT interval exhibited reverse-rate dependent properties such that QT intervals were longer at low pacing rates ($P<0.05$, Figure 5). Both dofetilide and tedisamil exacerbated this phenomenon and QT intervals were prolonged in a reverse rate-dependent fashion (Figure 5). The QTa-e interval was prolonged only at a pacing rate of 50 beats min^{-1} , whereas the QT interval was prolonged at all pacing rates relative to vehicle control (Figure 5). In this way the QTa-e interval was more specifically associated with the occurrence of torsades de pointes ($r=0.91$, $P<0.01$).

In parallel to the reverse rate-dependent actions of both drugs on the QT interval, the incidence of torsades de pointes was greater at lower pacing rates (Table 2). At a pacing rate of 50 beats min^{-1} all hearts treated with either 3 μM tedisamil or 30 nM dofetilide exhibited torsades de pointes. The first time hearts were paced at 50 beats min^{-1} torsades de pointes was observed in 4/5 hearts in each drug-treated group. During pacing at 50 beats min^{-1} the second time, torsades de pointes was observed in the one heart in each drug-treated group in which it was not observed during the first pacing episode at this rate. Thus, the overall incidence of torsades de pointes in drug-treated hearts was 5/5 ($P<0.05$ vs vehicle control).

Figure 1 The effect of vehicle control (top), dofetilide (middle) and tedisamil (bottom) on the QT interval in rabbit isolated hearts paced at 80, 120 and 240 beats min⁻¹. Each point represents the mean $\pm$ s.e. mean, $n=5-7$. The QT interval was measured from lead II. The * denotes statistical significance vs vehicle control at $P<0.05$ (repeated measures ANOVA followed by Tukey test). The # denotes statistical significance from the lower concentration of the same agent.

Once torsades de pointes occurred spontaneously it could commonly be provoked by a pause in cardiac activity and, in addition, torsades de pointes could be provoked after a pause in cardiac activity at pacing rates where it did not occur spontaneously. Torsades de pointes was observed in 1/5

Figure 2 The effect of vehicle control (top), dofetilide (middle) and tedisamil (bottom) on the QTa-e interval in rabbit isolated hearts paced at 80, 120 and 240 beats min⁻¹. Each point represents the mean $\pm$ s.e. mean, $n=5-7$. The QTa-e interval was measured from lead II. The * denotes statistical significance vs vehicle control at $P<0.05$ (repeated measures ANOVA followed by a Tukey test for differences).

hearts treated with 30 nM dofetilide and 2/5 hearts treated with 3 μM tedisamil when hearts were paced at 200 beats min⁻¹ (Table 2, Figure 6). These pause-dependent events occurred following a stepwise increase in the pacing rate from 100 to 200 beats min⁻¹. Spontaneous torsades de pointes was

Figure 3 The effect of vehicle control, dofetilide and tedisamil on the QT interval in rabbit isolated hearts during perfusion with 3 or 4 mM $[K^+]$. Mean QT intervals $\pm$ s.e.mean, $n=4-7$ during perfusion with vehicle control, dofetilide (C1=3 nM and C2=30 nM) or tedisamil (C1=0.3 μ M and C2=3 μ M). PD denotes pre-drug values. QT intervals were measured from lead II. The group mean ventricular rate was 50 ± 2 beats min^{-1} . The * denotes statistical significance vs vehicle control at $P < 0.05$ (repeated measures ANOVA followed by a Tukey test for differences).

Figure 4 Representative ECG record showing torsades de pointes induced in the rabbit isolated heart by 3 μ M tedisamil. Leads V1, V3, V5 and II are shown from top to bottom, respectively. The vertical divisions are 1 s apart and the horizontal divisions represent 0.5 mV. The perfusate $[K^+]$ was 4 mM.

Table 1 Incidence of dofetilide and tedisamil-induced torsades de pointes in rabbit isolated hearts

Treatment	Vehicle control	Dofetilide (3 nM)	Dofetilide (30 nM)	Tedisamil (0.3 μ M)	Tedisamil (3 μ M)
Overall incidence	0/7	1/7	7/7*	0/7	7/7*
$[K^+]$ 4 mM, 20 min	0/7	1/7	5/7*	0/7	7/7*
$[K^+]$ 3 mM, 5 min	0/7	1/7	3/7	0/7	3/7

Results are presented as the group incidence of torsades de pointes. The overall incidence summarizes the occurrence of torsades de pointes during both observation periods. For vehicle control, data are summarized for the entire duration of the experiment in a single column. Statistical significance vs vehicle control was determined using a Chi Square test at a significance level of $P < 0.05$.

not observed at a pacing rate of 200 beats min^{-1} nor could it be precipitated by a pause in cardiac activity after pacing for 10 min at this rate. The proarrhythmic actions of dofetilide and tedisamil were re-established upon reducing the pacing rate to 50 beats min^{-1} . Torsades de pointes was not observed in vehicle controls and ventricular fibrillation was not observed in any group.

Serum potassium concentration in conscious rabbits

Serum $[K^+]$ was measured in conscious rabbits to ensure that the $[K^+]$ studied *in vitro* was relevant to that observed *in vivo*. The serum $[K^+]$ in rabbits averaged 4.2 ± 0.1 mM ($n=35$, range 2.7–6.6 mM).

Discussion

The QT prolonging actions of dofetilide and tedisamil were associated with a similar risk of torsades de pointes. Both the QT prolongation and the proarrhythmic actions of dofetilide and tedisamil were concentration- and reverse rate-dependent. Thus, there was a high incidence of torsades de pointes in hearts treated with either 30 nM dofetilide or 3 μ M

tedisamil under conditions of bradycardia. Tedisamil's profile might be particularly insidious as this drug has bradycardic actions and could therefore precipitate the very conditions under which its proarrhythmic actions are manifested. The data also suggest that the proarrhythmic actions of dofetilide were exacerbated by perfusion with a low $[K^+]$ buffer.

The concentrations tested in the present study have been demonstrated to prolong cardiac action potentials, prolong the QT interval of the ECG and have antiarrhythmic actions *in vitro* and *in vivo* (Beatch *et al.*, 1991; Tsuchihashi & Curtis, 1991; Chi *et al.*, 1996; Wallace *et al.*, 1995; Friedrichs *et al.*, 1996; 1998). Our previous studies in the rabbit isolated heart preparation demonstrated the antifibrillatory effects of tedisamil at a concentration of 3 μ M (Chi *et al.*, 1996). Similarly, a concentration of ~ 0.6 μ M has been documented to be effective in our canine sudden death model (Friedrichs *et al.*, 1998). The concentrations tested in the present study are therefore directly relevant to the observed antiarrhythmic actions of tedisamil and the use of this agent to prevent or revert cardiac arrhythmias is likely to be associated with a risk of torsades de pointes in man. The doses used to treat angina pectoris in man are somewhat lower than those demonstrated to have antiarrhythmic actions in animal models. Despite the lower dose of tedisamil, the QTc interval

was prolonged in clinical trials (Bargheer *et al.*, 1994; Fox *et al.*, 2000). If prolongation of the QTc interval is taken as a valid measure of the risk of torsades de pointes, then the data

Figure 5 The effects of vehicle control, dofetilide and tedisamil on the QT interval (top) and QTa-e interval (bottom) in rabbit isolated hearts paced at 50, 100, 200 and 50 beats min⁻¹. Each point represents the mean $\pm$ s.e. mean, $n=4-5$. Both intervals were measured from lead II. The * denotes statistical significance vs vehicle control at $P<0.05$ (repeated measures ANOVA followed by a Tukey test for differences).

suggest that there is a risk of torsades de pointes with tedisamil at doses used to treat angina pectoris.

It is noteworthy that the QTc interval was not prolonged at effective doses (3 mg kg⁻¹ BID p.o.) in our canine sudden death model (Friedrichs *et al.*, 1998). Torsades de pointes also was not observed in this study. Torsades de pointes also was not observed in our previous *in vitro* study with tedisamil, despite perfusion of rabbit isolated hearts with a low [K⁺] buffer (2.5 mM, Chi *et al.*, 1996). In the latter study, hearts were paced faster than their intrinsic sinus rate. One possible explanation for the apparently incongruent observations is that the proarrhythmic actions of tedisamil are bradycardia dependent (as demonstrated in the present study).

The QT prolonging actions of dofetilide and tedisamil were reverse rate-dependent in keeping with previous observations (Sanguinetti & Jurkiewicz, 1990). Dofetilide is a potent and selective blocker of the rapidly activating component of the delayed rectifier K⁺ current (I_{Kr}). Reverse rate-dependent

Figure 6 Representative ECG record showing pause-dependent torsades de pointes induced in the rabbit isolated heart by 3 μM tedisamil. Leads V1, V3, V5 and II are shown from top to bottom, respectively. The vertical divisions are 1 s apart and the horizontal division represent 0.5 mV. The perfusate [K⁺] was 4 mM. Note the presence of AV block, which is characterized by the dissociation between P waves and the QRS complex.

Table 2 The effect of heart rate on the incidence of dofetilide and tedisamil-induced torsades de pointes in the rabbit isolated heart

Treatment	50 beats min ⁻¹	100 beats min ⁻¹	200 beats min ⁻¹	50 beats min ⁻¹
<i>Spontaneous torsades de pointes</i>				
Vehicle control	0/5	0/5	0/5	0/5
30 nM Dofetilide	4/5*	1/5	0/5	4/5*
3 μM Tedisamil	4/5*	1/5	0/5	4/5*
<i>Pause-dependent torsades de pointes</i>				
Vehicle control	0/5	0/5	0/5	0/5
30 nM Dofetilide	3/5	2/5	1/5	3/5
3 μM Tedisamil	3/5	2/5	2/5	4/5*

Results are presented as the group incidence of torsades de pointes at the pacing rates indicated. Pause-dependent torsades de pointes refers to the occurrence of torsades de pointes following a pause in cardiac activity (see text for definitions). Statistical significance vs vehicle control was tested using a Chi Square test with a significance level of $P<0.05$.

action potential and QT interval prolongation are the hallmarks of drugs with this profile (Hondeghe & Snyders, 1990). The similarity in the actions of dofetilide and tedisamil suggest that tedisamil's QT prolonging and proarrhythmic actions are mediated by blockade of I_{K_r} . While this might explain the observed effects, it is by no means certain. As noted earlier, tedisamil is known to block a number of cardiac ion currents, any of which might contribute to the observed proarrhythmic effects.

The proarrhythmic actions of dofetilide tended to be exacerbated by perfusion with low $[K^+]$ buffer (3 vs 4 mM). Low $[K^+]$ concentrations are known to potentiate the blockade of I_{K_r} produced by dofetilide (Yang & Roden, 1996; Duff *et al.*, 1997). In keeping with this, torsades de pointes occurred in two hearts treated with 30 nM dofetilide during perfusion with 3 mM $[K^+]$ in which torsades de pointes did not occur during perfusion with 4 mM $[K^+]$.

The QTa-e interval was measured in an attempt to assess drug-induced changes in T wave morphology and to relate these changes to the occurrence of torsades de pointes. It has been suggested that changes in T wave morphology and/or the appearance of a U wave may be more predictive of torsades de pointes than QT (or QTc) prolongation *per se* (Jackman *et al.*, 1988; Antzelevitch *et al.*, 1999). The QTa-e interval was prolonged in a concentration- and reverse rate-dependent fashion by both drugs, but was independent of pacing rate in vehicle controls. Moreover, prolongation of the QTa-e interval was associated with a high incidence of torsades de pointes. Thus, this measure might prove to be a

useful, quantitative measure of changes in T wave morphology. Further studies are required to assess the sensitivity, specificity and utility of this measure.

Torsades de pointes occurred commonly after a pause in cardiac activity at low pacing rates and could be precipitated after a pause in cardiac activity during a rate transition at higher pacing rates. Such pause-dependent torsades de pointes might be clinically important (Jackman *et al.*, 1988). A pause in cardiac activity could occur after a premature beat or may also occur in patients with sinus node dysfunction. Patients converted from atrial fibrillation are known to have sinus node dysfunction and antiarrhythmic drugs can exacerbate this dysfunction. In addition, conversion of atrial fibrillation is associated with sudden changes in ventricular rate. It is not surprising, therefore, that torsades de pointes can occur quickly after conversion of atrial fibrillation to sinus rhythm with Class III drugs (e.g., Kowey *et al.*, 1996; Frost *et al.*, 1997). The model described in the present study offers the advantage of allowing such pause-dependence to be examined.

This study was supported by the Cardiovascular Research Fund of the University of Michigan. T.D. Barrett and J.K. Hennen are recipients of a Research Fellowship from the Heart and Stroke Foundation of British Columbia and the Yukon (T.D. Barrett), and of Canada (J.K. Hennen). The authors would like to thank Dr Andy Alt for his contribution to the manuscript.

References

- ANTZELEVITCH, C., SHIMIZU, W., YAN, G.X., SICOURI, S., WEISSENBERGER, J., NESTERENKO, V.V., BURASHNIKOV, A., DI DIEGO, J., SAFFITA, J. & THOMAS, G.P. (1999). The M cell: its contribution to the ECG and to normal and abnormal electrical function of the heart. *J. Cardiovasc. Electrophysiol.*, **10**, 1124–1152.
- BARGHEER, K., DOBDE, F., KLEIN, H.U., TRAPPE, H.J., FRANZ, M.R. & LICHTLEN, P.R. (1994). Prolongation of monophasic action potential duration and the refractory period in the human heart by tedisamil, a new potassium channel-blocking agent. *Eur. Heart J.*, **15**, 1409–1414.
- BEATCH, G.N., ABRAHAM, S., MACLEOD, B.A. & WALKER, M.J.A. (1991). Antiarrhythmic properties of tedisamil (KC8857), a putative, transient outward K^+ channel blocker. *Br. J. Pharmacol.*, **102**, 13–18.
- BRAY, K. & QUAST, U. (1992). Differential inhibition by tedisamil (KC 8857) and glibenclamide of the responses to cromakalim and minoxidil sulfate in rat isolated aorta. *Naunyn-Schmiedeberg's Arch. Pharmacol.*, **345**, 244–250.
- BRIL, A., GOUT, B., BONHOMME, M., LANDAIS, L., FAIVRE, J.F., LINEE, P., POYSER, R.H. & RUFFOLO JR., R.R. (1996). Combined potassium and calcium channel blocking activities as a basis for antiarrhythmic efficacy with low proarrhythmic risk: experimental profile of BRL-32872. *J. Pharmacol. Exp. Ther.*, **276**, 637–646.
- BUCHANAN, L.V., KABELL, G., BRUNDER, M.N. & GIBSON, J.K. (1993). Comparative assessment of ibutilide, D-sotalol, clofilium, E-4031, and UK-68,798 in a rabbit model of proarrhythmia. *J. Cardiovasc. Pharmacol.*, **22**, 540–549.
- CHI, L., PARK, J.L., FRIEDRICH, G.S., BANGLAWALA, Y.A., PEREZ, M.A., TANHEHCO, E.J. & LUCCHESI, B.R. (1996). Effects of tedisamil (KC-8857) on cardiac electrophysiology and ventricular fibrillation in the rabbit isolated heart model. *Br. J. Pharmacol.*, **117**, 1261–1269.
- CONNOLLY, S.J. (1999). Evidence-based analysis of amiodarone efficacy and safety. *Circulation*, **100**, 2025–2034.
- DUFF, H.J., FENG, Z.P., FISET, C., WANG, L., LEES-MILLER, J. & SHELDON, R.S. (1997). $[^3H]$ Dofetilide binding to cardiac myocytes: modulation by extracellular potassium. *J. Mol. Cell. Cardiol.*, **29**, 183–191.
- DUKES, I.D., CLEEMANN, L. & MORAD, M. (1990). Tedisamil blocks the transient outward and delayed rectifier K^+ currents in mammalian cardiac and glial cells. *J. Pharmacol. Exp. Ther.*, **254**, 560–568.
- DUKES, I.D. & MORAD, M. (1989). Tedisamil inactivates transient outward K^+ current in rat ventricular myocytes. *Am. J. Physiol.*, **257**, H1746–H1749.
- ECHT, E.D., LEE, J.T., MURRAY, K.T., VORPERIAN, V., BORGANELLIT, S.M., CRAWFORD, D.M., FRIEDRICH, T. & RODEN, D.M. (1995). A randomized, double-blind, placebo-controlled, dose-ranging study of dofetilide in patients with inducible sustained ventricular tachyarrhythmias. *J. Cardiovasc. Electrophysiol.*, **6**, 687–699.
- ECKARDT, L., HAVERKAMP, W., BORGGREFE, M. & BREITHARDT, G. (1998). Experimental models of torsades de pointes. *Cardiovasc. Res.*, **39**, 178–193.
- FAIVRE, J.F., ROUANET, S. & BRIL, A. (1998). Comparative effects of glibenclamide, tedisamil, dofetilide, E-4031, and BRL-32872 on protein kinase A-activated chloride current in guinea pig ventricular myocytes. *J. Cardiovasc. Pharmacol.*, **3**, 551–557.
- FISCHBACH, P.S., JOHNSTON, P.V., FRIEDRICH, G.S. & LUCCHESI, B.R. (1999). Tedisamil in chronic canine atrial flutter. *J. Cardiovasc. Pharmacol.*, **34**, 212–218.
- FOX, K.M., HENDERSON, J.R., KASKI, J.C., SACHSE, A., KUESTER, L. & WONNACOTT, S. (2000). Antianginal and anti-ischaemic efficacy of tedisamil, a potassium channel blocker. *Heart*, **83**, 167–171.
- FRIEDRICH, G.S., ABREU, J.N., DRISCOLL JR., E.M., BORLAK, J. & LUCCHESI, B.R. (1998). Antifibrillatory efficacy of long-term tedisamil administered in a postinfarcted canine model of ischemic ventricular fibrillation. *J. Cardiovasc. Pharmacol.*, **31**, 55–66.

- FRIEDRICHS, G.S., ABREU, J.N., COUSINS, G.R., CHI, L., BORLAK, J. & LUCCHESI, B.R. (1996). Tedisamil attenuates ventricular fibrillation in a conscious canine model of sudden cardiac death. *J. Cardiovasc. Pharmacol. Therapeut.*, **1**, 313–324.
- FROST, L., MORTENSEN, P.E., TINGLEFF, J., PLATOU, E.S., CHRISTIANSEN, E.H. & CHRISTIANSEN, N. (1997). Efficacy and safety of dofetilide, a new class III antiarrhythmic agent, in acute termination of atrial fibrillation or flutter after coronary artery bypass surgery. Dofetilide Post-CABG Study Group. *Int. J. Cardiol.*, **58**, 135–140.
- HONDEGHEM, L.M. & SNYDERS, D.J. (1990). Class III antiarrhythmic agents have a lot of potential but a long way to go. Reduced effectiveness and dangers of reverse use dependence. *Circulation*, **81**, 686–690.
- JACKMAN, W.M., FRIDAY, K.J., ANDERSON, J.L., ALIOT, E.M., CLARK, M. & LAZZARA, R. (1988). The long QT syndromes: A critical review, new clinical observations and a unifying hypothesis. *Prog. Cardiovasc. Dis.*, **31**, 115–172.
- JOHNA, R., MERTENS, H., HAVEKAMP, W., ECKARDT, L., NIEDERBROKER, T., BORGGREFE, M. & BREITHARDT, G. (1998). Clofilium in the isolated perfused rabbit heart: a new model to study proarrhythmia induced by class III antiarrhythmic drugs. *Basic Res. Cardiol.*, **93**, 127–135.
- KEHOE, R.S., ZHEUTLIN, T.A., DUNNINGTON, C.S., MATTIONI, T.A., YU, G. & SPANGENBERGER, R.B. (1990). Safety and efficacy of sotalol in patients with drug refractory sustained ventricular tachyarrhythmias. *Am. J. Cardiol.*, **65**, 58A–64A.
- KOWEY, P.K., VANDERLUGT, J.T. & LUDERER, J.R. (1996). Safety and risk/benefit analysis of ibutilide for acute conversion of atrial fibrillation/flutter. *Am. J. Cardiol.*, **78**, 46–52.
- NEMETH, M., VIRAG, L., HALA, O., VARRO, A., KOVACS, G., THORMAHLEN, D. & PAPP, J.G. (1996). The cellular electrophysiological effects of tedisamil in human atrial and ventricular fibres. *Cardiovasc. Res.*, **31**, 246–248.
- ORAL, H., SOUZA, J.J., MICHAUD, G.F., KNIGHT, B.P., GOYAL, R., STRICKBERGER, S.A. & MORADY, F. (1999). Facilitating transthoracic cardioversion of atrial fibrillation with ibutilide pretreatment. *N. Engl. J. Med.*, **17**, 1910–1912.
- REES, S.A., TSUCHIHASHI, K., HEARSE, D.J. & CURTIS, M.J. (1993). Combined administration of an $I_{K(ATP)}$ activator and I_{to} blocker increases coronary flow independent of effects on heart rate, QT interval and ischaemia-induced ventricular fibrillation in rat. *J. Cardiovasc. Pharmacol.*, **22**, 343–349.
- SANGUINETTI, M.C. & JURKIEWICZ, N.K. (1990). Two components of cardiac delayed rectifier K^+ current. Differential sensitivity to block by class III antiarrhythmic agents. *J. Gen. Physiol.*, **96**, 195–215.
- STAMBLER, B.S., BECKMAN, K.J., KADISH, A.H., CAMM, J.A., ELLENBOGEN, K.A., PERRY, K.T. & VANDERLUGT, J.T. (1997). Acute hemodynamic effects of intravenous ibutilide in patients with without reduced left ventricular function. *Am. J. Cardiol.*, **80**, 458–463.
- TORP-PEDERSEN, C., MOLLER, M., BLOCH-THOMSEN, P.E., SANDOE, E., EGSTRUP, K., AGNER, E., CARLSEN, J., VIDEBAEK, J., MARCHANT, B. & CAMM, A.J. (1999). Dofetilide in patients with congestive heart failure and left ventricular dysfunction. Danish Investigations of Arrhythmia and Mortality on Dofetilide Study Group. *New Engl. J. Med.*, **341**, 857–865.
- TSUCHIHASHI, K. & CURTIS, M.J. (1991). Influence of tedisamil on the initiation and maintenance of ventricular fibrillation: chemical defibrillation by I_{to} blockade? *J. Cardiovasc. Pharmacol.*, **18**, 445–456.
- WALLACE, A.A., STUPIENSKI III, R.F., BASKIN, E.P., APPLEBY, S.D., KOTHSTEIM, T., GEHRET, J.R., KING, S.W., REMY, D.C. & LYNCH, J.J. (1995). Cardiac electrophysiological and antiarrhythmic actions of tedisamil. *J. Pharmacol. Exp. Ther.*, **273**, 168–175.
- YANG, T. & RODEN, D.M. (1996). Extracellular potassium modulation of drug block of I_{Kr} . Implications for torsade de pointes and reverse use-dependence. *Circulation*, **93**, 407–411.
- ZABEL, M., HOHNLOSER, S.H., BEHRENS, S., LI, Y.G., WOOSLEY, R.L. & FRANZ, M.R. (1997). Electrophysiological features of torsades de pointes: insight from a new isolated rabbit heart model. *J. Cardiovasc. Electrophysiol.*, **8**, 1159–1162.

(Received October 31, 2000

Revised January 18, 2001

Accepted January 18, 2001)