Electrophysiological effects of E-3753, a new antiarrhythmic drug, in guinea-pig ventricular muscle

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- 1 The electrophysiological effects of E-3753, a new antiarrhythmic drug, were studied in guinea-pig papillary muscles.
- 2 E-3753 (10^{-7} – 10^{-4} M) produced a concentration-dependent decrease in the action potential amplitude and V_{max} of the upstroke, shortened the action potential duration (APD) and shifted the resting membrane potential to less negative values. E-3753 also shortened the effective refractory period (ERP), lengthening the ERP relative to APD.
- 3 E-3753 (10^{-5} M) shifted the membrane responsiveness curve along the voltage axis to more negative potentials.
- 4 In the presence of E-3753 (10^{-5} M) trains of stimuli at rates between 0.5 and 3 Hz led to an exponential decline in V_{max} (onset rate at 3 Hz, 0.05 ± 0.009 per action potential), to a new steady-state level. This use-dependent V_{max} block was augmented at higher rates of stimulation. The time constant for the recovery of V_{max} from the use-dependent block was 41.1 ± 4.8 s.
- 5 E-3753 (10^{-5} – 10^{-4} M) had no effect on the characteristics of the slow action potentials elicited by isoprenaline in ventricular fibres depolarized by 27 mM KCl.
- 6 The slow onset of use-dependent block during repetitive activity and the slow offset kinetics of use-dependent V_{max} block suggest that E-3753 exhibits class Ic antiarrhythmic actions in ventricular muscle fibres but does not exhibit class IV (Ca antagonist) antiarrhythmic actions.

Introduction

E-3753, 1H-imidazol-2-methanol, α -(3-trifluoromethylphenyl)- α -methyl-1-methyl-0-(3-terbutylaminopropane)-2ol (Figure 1), is a new antiarrhythmic drug which exhibits local anaesthetic properties (Colombo *et al.*, 1987). E-3753 has been shown to be efficacious in suppressing cardiac arrhythmias in a variety of experimental models. Thus, E-3753 was as potent as lignocaine in reversing ventricular arrhythmias induced by ouabain in the anaesthetized guinea-pig and 2–3 times more potent than quinidine, lignocaine and

lorcainide in suppressing the experimental arrhythmias induced by mechanical lesions in rat isolated right ventricles (Farré *et al.*, 1988). In contrast, E-3753 had no effect on the positive inotropic and chronotropic responses induced by isoprenaline in guinea-pig isolated atria or on the maximum upstroke velocity of the slow action potentials induced in K-depolarized guinea-pig papillary muscles (Delpón *et al.*, 1988). These results suggested that E-3753 had no class II (antisympathetic) or class IV (Ca antagonist) antiarrhythmic actions. However, preliminary microelectrode studies in guinea-pig atrial and ventricular muscle fibres indicated that E-3753 decreased action potential upstroke velocity (V_{max}), suggesting a class I mechanism of action, i.e., fast sodium channel block (Delpón *et al.*, 1988).

Thus the present experiments were undertaken to: (1) characterize the cellular electrophysiological effects of E-3753 in guinea-pig ventricular muscle fibres and (2) determine the effects of this drug on the onset and recovery kinetics of the sodium channel as reflected by its effects on V_{max} . Moreover, since the

Figure 1 Chemical structure of E-3753.

prevailing classifications of antiarrhythmic drugs (Harrison, 1983; Vaughan Williams, 1984) are based on changes in action potential duration, refractoriness and use-dependent block of fast sodium channels, the present experiments were also carried out in order to (3) place E-3753 in the appropriate class I subgroup (i.e., class Ia, Ib, Ic).

Methods

General procedure

Guinea-pigs of either sex weighing 250–350 g were killed by a blow on the head. The hearts were rapidly excised and papillary muscles 2 to 3 mm in length and less than 1 mm in diameter were isolated from both ventricles. The muscles were pinned to the bottom of a Lucite chamber and superfused continuously at a constant rate of 7 ml min^{-1} with Tyrode solution of the following composition (mM): NaCl 137, KCl 5.4, CaCl_2 1.8, MgCl_2 1.05, NaHCO_3 11, NaH_2PO_4 0.42, glucose 5.5. The solutions were bubbled with 95% O_2 and 5% CO_2 and maintained at $34 \pm 0.5^\circ\text{C}$. The preparations were initially driven at 1 Hz and a period of 1 h was allowed for equilibration during which a stable impalement was obtained. Driving stimuli were rectangular pulses (2 ms duration, twice threshold strength) delivered to the preparation from a multipurpose programmable stimulator (Cibertec CS-220). Electrical stimulation was applied to the surface of the preparation through Teflon-coated bipolar electrodes of silver wire.

Transmembrane action potentials (AP) were conventionally recorded through glass microelectrodes filled with 3 M KCl (tip resistance of 10–20 M Ω). The microelectrode was connected via Ag-AgCl wire to high impedance, capacity neutralizing amplifiers (WPI model 701). The maximal rate of depolarization (V_{max}) of the action potential was obtained by electronic differentiation (Tamargo & Delgado, 1985). The differentiator used had an upper limit of linearity of 1000 V s^{-1} and possessed variable input filters (3 Hz–260 kHz). The suitable frequency filter for minimizing noise without reducing the V_{max} was elected for each individual experiment. Another distortion of V_{max} can arise from the formation of the foot of the rising phase of the electrical stimulus. This phenomenon is accompanied by a reduced interval between stimulus artifact and the upstroke of the action potential, particularly when strong electrical stimuli are applied. In order to diminish this artefact, stimulus intensity and duration were adjusted throughout each experiment to maintain a constant latency (1–2 ms) between stimulus and upstroke of the action potential to minimize latency-

induced alterations in V_{max} (Valenzuela *et al.*, 1988). Both action potential and V_{max} were displayed on a storage oscilloscope (Tektronix 5104N) and the oscilloscope traces were photographed with a kymographic camera (Grass C4). The effective refractory period (ERP) was measured by introducing premature test-stimuli of twice threshold strength at different intervals from the preceding basic action potential. Interpolation and shift along the voltage axis were carried after every eighth basic drive stimulus. All experimental results were obtained from a single continuous impalement through the whole experiment.

To study the rate-dependent effects of E-3753 on V_{max} , preparations were driven at a basal rate of 0.05 Hz. Following the equilibration period, the preparations were driven by trains of stimuli at varying rates ranging from 0.5 to 3 Hz for 40 s. Rest periods of 5 min, which were sufficient to ensure full recovery from use-dependent decrease in V_{max} , were interpolated between the trains of stimuli (Valenzuela *et al.*, 1988). A similar experimental protocol was followed after exposure to 10^{-5} M E-3753. This experimental protocol indicated that E-3753 at any given rate produced two types of V_{max} inhibition, a tonic and a phasic blockade. Tonic blockade is the decrease of V_{max} of the first action potential preceded by the rest periods, whereas phasic blockade is the decrease of V_{max} during a train from the value of the first action potential to a new steady-state.

Recovery of V_{max} from rate-dependent block was studied by applying a single test stimulus at various coupling intervals after a stimulation train for 5 s at 4 Hz. The intensity and duration of the test and conditioning stimuli were adjusted to obtain a constant latency from the stimulus artefact to the initiation of the upstroke of the action potential. The effect of E-3753 on the relationship between membrane resting potential and V_{max} , i.e. membrane responsiveness, was studied in papillary muscles driven at 1 Hz and the membrane potential was lowered by adding to the perfusate KCl in steps from 2.7 mM to a final concentration of 15 mM (Valenzuela *et al.*, 1987).

Drugs

E-3753 (Esteve, S.A.) as a powder was prepared by dissolving 10 mg in 0.4 ml of 0.1 N HCl and adding 9.6 ml of H_2O to yield a stock solution of 1 mg ml^{-1} . Further dilutions were carried out in Tyrode solution to obtain final concentrations between 10^{-7} M and 10^{-4} M (0.039 – $39.9 \mu\text{g ml}^{-1}$). After 1 h equilibration, control measurements were performed. The preparations were then superfused with Tyrode solutions containing the drug at various concentrations for 30 min.

Throughout the paper, data are given as

mean $\pm$ s.e.mean and Student's *t* test was used to estimate the significance of differences from control values. For statistical comparison of more than two groups, a one-way analysis of variance and Scheffé's test for critical difference was used (Wallenstein *et al.*, 1980). A *P* value of less than 0.05 was considered to be significant.

Results

Effects of E-3753 on transmembrane action potentials

The effects of E-3753 in a range of concentrations between 10^{-7} and 10^{-4} M were studied in ventricular muscle fibres. The effects of E-3753 were usually evident within 10 min of beginning the perfusion and reached steady-state values within 30 min. Results obtained under control conditions and 30 min after each increment in drug concentration are shown in Table 1. In ventricular muscle fibres E-3753 at concentrations higher than 10^{-7} M produced a significant decrease ($P < 0.05$) in V_{max} , which at concentrations $\geq 10^{-5}$ M was accompanied by a significant decrease in action potential amplitude ($P < 0.05$) and a progressive shift of the resting membrane potential to less negative values. At concentrations between 10^{-7} and 10^{-5} M E-3753 had no effect on ventricular repolarization, whereas at higher concentrations it increased the slope of phases 2 and 3 which led to a significant shortening of both APD₅₀ and APD₉₀ values ($P < 0.05$).

In 11 papillary muscles the ventricular effective refractory period (ERP) averaged 179.8 ± 9.1 ms. At 5×10^{-5} M and 10^{-4} M, E-3753 shortened the ERP to 157.5 ± 14.3 ms ($P < 0.05$) and 143.5 ± 10.8 ms ($P < 0.05$), respectively. This shortening was accompanied by a greater shortening in the APD₉₀ values, and therefore, at both concentrations E-3753 also produced a significant decrease in the ERP/APD

Figure 2 Effect of E3753 on the relationship between V_{max} and the resting membrane potential at which the action potential is generated, i.e. membrane responsiveness. Ordinate scale: normalized V_{max} values. Abscissa scale: resting membrane potential (mV). (●) Controls. (○) E-3753 10^{-5} M.

ratio from 1.01 ± 0.03 to 1.11 ± 0.03 ($P < 0.05$) and 1.18 ± 0.06 ($P < 0.05$).

Effects of membrane responsiveness

The relationship between V_{max} and the membrane resting potential from which the action potential originates, i.e. membrane responsiveness, was examined in four papillary muscles driven at basal rate of 1 Hz. The membrane resting potential was depolarized in steps from its original resting level to approximately -60 mV by increasing the K^+ concentration in the Tyrode solution from 2.7 to 15 mM. The results of a typical experiment are shown in Figure 2. In the absence of the drug, the membrane potential at which V_{max} was reduced to half of its maximum value (V_h) was 61.0 ± 1.2 mV ($n = 6$). Addition of E-3753 shifted the membrane responsiveness curve along the voltage axis to more negative potentials. The average shift in V_h determined in 6 preparations exposed to 10^{-5} M E-3753 was -6.2 ± 1.2 mV. These results

Table 1 Electrophysiological effects of E-3753 on transmembrane potentials in guinea-pig papillary muscles

Concentration (M)	Resting potential (mV)	V_{max} ($V s^{-1}$)	APA (mV)	APD ₅₀ (ms)	APD ₉₀ (ms)
0	-86.5 ± 0.6	194.9 ± 7.3	118.8 ± 0.7	151.6 ± 11.5	176.8 ± 11.0
10^{-7}	-85.7 ± 0.6	185.0 ± 8.4	117.7 ± 0.9	143.4 ± 15.7	167.9 ± 16.3
10^{-6}	-85.0 ± 0.6	$176.0 \pm 7.8^*$	117.4 ± 0.8	148.2 ± 15.1	174.1 ± 15.5
10^{-5}	$-82.7 \pm 0.6^{***}$	$133.2 \pm 7.7^{***}$	$116.4 \pm 0.6^*$	143.2 ± 14.8	169.1 ± 15.4
5×10^{-5}	$-80.7 \pm 0.8^{***}$	$80.0 \pm 4.1^{***}$	$111.2 \pm 0.6^{***}$	$116.7 \pm 12.7^*$	$142.2 \pm 14.6^*$
10^{-4}	$-78.8 \pm 0.8^{***}$	$50.6 \pm 3.7^{***}$	$101.7 \pm 2.2^{***}$	$97.0 \pm 9.0^{**}$	$126.6 \pm 10.4^{**}$

Mean values $\pm$ s.e.mean, $n = 11$.

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

APA = action potential amplitude. APD = action potential duration measured at 50% (APD₅₀) and 90% (APD₉₀) or repolarization.

indicated that the effect of E-3753 was more pronounced at low (less negative) than at normal resting membrane potentials.

Use-dependent effects of E-3753

The effectiveness of many class I antiarrhythmic drugs at inhibiting I_{Na} is strongly dependent on the frequency of stimulation. Thus, the influence of stimulation frequency on the depressant effect of E-3753 on V_{max} was studied in papillary muscles by applying trains of pulses of 40 s in duration at different rates (0.5–3 Hz) which were separated by a 5 min period of rest. In untreated muscles the value of V_{max} remained almost unchanged during the stimulation trains at rates ranging from 0.5 to 3 Hz (Figure 3). Following the perfusion with 10^{-5} M E-3753 the V_{max} of the first action potential in each train was reduced from 224 V s^{-1} to 207 V s^{-1} , i.e. there was a 6.4% tonic (resting) V_{max} block. During the trains of pulses there was a gradual decrease of V_{max} from beat to beat until a new steady-state occurred (use-dependent or phasic block), which depended on the stimulation frequency. In the absence of the drug, the percentage decrease of V_{max} from the first action potential of the train to a new steady-state level increased progressively when the frequency of stimu-

Figure 3 Onset of use-dependent depression of V_{max} induced by 10^{-5} M E-3753 in guinea-pig papillary muscles driven by trains of stimuli at various rates (0.5–3 Hz). Stimulation caused a use-dependent block which disappears, whereas the tonic V_{max} block persisted. The tonic block (%) results from $[1 - V_{max}(\text{first beat, drug})/V_{max}(\text{rested, drug free})] \times 100\%$ and the use-dependent block (%) from $[1 - V_{max}(\text{ss, drug})/V_{max}(\text{first beat, drug})]$. $V_{max}(\text{ss})$ is the steady-state value attained during continuous stimulation and $V_{max}(\text{first beat})$ is the value of the first beat of each train of stimuli (●) 0.5 Hz, (○) 1 Hz, (△) 2 Hz, (▲) 3 Hz.

Table 2 Effect of 10^{-5} M E-3753 on the onset kinetics of use-dependent V_{max} block

Frequency of stimulation (Hz)	τ_{on} (s)	Onset rate per action potential (AP^{-1})
0.5	10.7 ± 0.7	0.18 ± 0.01
1.0	10.3 ± 0.6	$0.08 \pm 0.007^{***}$
2.0	9.5 ± 0.3	$0.05 \pm 0.004^{***}$
3.0	$5.6 \pm 0.5^{***}$	$0.05 \pm 0.009^{***}$

Mean values $\pm$ s.e.mean, $n = 6$.

*** $P < 0.001$.

lation increased from 0.5 ($2.8 \pm 0.7\%$) to 3 Hz ($10.8 \pm 1.0\%$). In the presence of 10^{-5} M E-3753 the use-dependent V_{max} block significantly increased at all stimulation rates tested ($P < 0.001$), but this increase was more marked at 2 ($51.1 \pm 2.6\%$) and 3 Hz ($54.2 \pm 4.6\%$) than at 0.5 ($27.8 \pm 2.0\%$) and 1 Hz ($37.7 \pm 2.5\%$), i.e. was more marked at fast than at slow frequencies of stimulation.

Onset kinetics of phasic V_{max} blockade can be defined either in terms of a time- or an event-dependent process. In both cases, the decline of V_{max} during stimulation trains represents a first order reaction and can be well fitted by a single exponential. Thus, the time-constant (τ_{on}) and the onset rate per action potential (AP^{-1}) at which V_{max} fell to the new steady-state level could be calculated from the regression lines in semilogarithmic plots. Table 2 shows that between 0.5 and 2 Hz τ_{on} values were almost similar (10.7 ± 0.7 s, 10.3 ± 0.6 s and 9.5 ± 0.3 s, respectively. $P > 0.05$), whereas at 3 Hz the τ_{on} significantly shortened to a value of 5.6 ± 0.7 s ($P < 0.05$). On the other hand, at frequencies of stimulation between 1 and 3 Hz the rate-constants (K) showed similar values, but K was significantly increased ($P < 0.05$) at the lower stimulation frequency.

Recovery kinetics of phasic V_{max} block

The effect of E-3753, 10^{-5} M, on the kinetics of the offset (recovery) of phasic V_{max} block was studied in 6 papillary muscles driven at a basal frequency of 0.05 Hz. Muscles were driven by a train of stimuli at a frequency of 4 Hz for 5 s and a test-stimulus was applied at variable coupling intervals from 2 to 36 s. Recovery from use-dependent V_{max} block occurs as a monoexponential process for which a single time constant (τ_{re}) can be estimated by regression analysis of the data. Under control conditions, the τ_{re} was 26.5 ± 1.3 ms whereas in the presence of E-3753 the τ_{re} was prolonged to 41.1 ± 4.8 ms ($P < 0.001$). This value indicates that E-3753 must be considered as belonging to the kinetically slow group of class I antiarrhythmic drugs (Campbell, 1983a).

Effect on the slow action potentials

The effects of E-3753 were also studied on slow action potentials in muscles driven at a basal rate of 0.12 Hz. When the K concentration in the Tyrode solution was increased from 5.4 (control) to 27 mM, the membrane potential was depolarized to -43.0 ± 1.0 mV ($n = 6$) and the preparations became inexcitable due to voltage-dependent inactivation of the Na channels. Excitability, i.e. slow action potentials, were induced by adding 10^{-6} M isoprenaline to the perfusate. E-3753 at any of the concentrations tested (10^{-5} , 5×10^{-5} and 10^{-4} M) did not modify the resting membrane potential, amplitude, V_{max} and duration of the slow action potentials. These results suggest that it did not exhibit, in addition, class IV antiarrhythmic actions.

Discussion

In this paper we studied the effects of E-3753, a new antiarrhythmic class I drug, on the electrophysiological properties of guinea-pig ventricular muscles fibres. Also, the onset and recovery kinetics of use-dependent V_{max} block were analysed in order to determine the subclassification of E-3753.

The present results indicated that in ventricular muscle fibres E-3753 produced a concentration-dependent decrease of V_{max} values; this effect occurred even at concentrations which had no effect on other AP characteristics or resting membrane potential. These findings suggested that the depressive effect on V_{max} is probably due to an inhibition of the fast inward sodium current, and the drug can be classified as a class I, i.e. membrane stabilizing antiarrhythmic agent (Vaughan Williams, 1984). In this regard, E-3753 has about the same degree of local anaesthetic activity as lignocaine (unpublished data). At higher concentrations, E-3753 decreased the action potential amplitude and shifted the resting membrane potential to less negative values, thus producing a further decrease in phase 0 characteristics. Moreover, E-3753 produced a progressive shortening of the APD and ERP, although the shortening of the APD₉₀ was greater than that of the ERP and therefore the ERP/APD₉₀ ratio increased.

There is evidence that during the fast upstroke of the cardiac action potential, the majority of ionic current crossing the cell membrane at the time of V_{max} is a sodium current (Hondeghe, 1978; Walton & Fozzard, 1979). Thus, although the use of changes in V_{max} to measure peak sodium current and sodium conductance has been questioned (Cohen *et al.*, 1984), there is little doubt that V_{max} is mainly generated by the fast inward sodium current. Whether or not possible non-linearities between V_{max} and sodium

conductance (G_{Na}) affect the present results remains to be determined in reliable voltage-clamp experiments, under the same conditions of temperature and external sodium concentration as used in this study.

The modulated-receptor hypothesis (Hondeghe & Katzung, 1977; 1984) is a useful model for investigating the use-dependent effects of class I antiarrhythmic drugs. According to this hypothesis, reduction of sodium inward current is due to an accumulation of drug-associated non-conducting channels, the affinity of the drug being modulated by the state (resting, activated, inactivated) of the channel. At therapeutic concentrations most class I antiarrhythmic drugs exhibit little or no tonic V_{max} block (Campbell 1983a,b; Hondeghe & Katzung, 1984; Tamargo *et al.*, 1988) and E-3753 is no exception. The fact that little or no resting block was found at normally polarized ventricular fibres suggests that E-3753 has little affinity for the resting state of the sodium channel. As compared to periods of quiescence, during the periods of stimulation channels spend proportionately more time in the open state during the upstroke and in the inactivated state during the plateau. Thus, if like other antiarrhythmic drugs E-3753 has higher affinity for the activated and inactivated states of the channel (Hondeghe & Katzung, 1984), an accumulation of blocked channels during the stimulation period leading to a use-dependent V_{max} block would be expected.

The decrease of V_{max} increased progressively when the preparations were stimulated at progressively increasing driving rates and the onset rate of use-dependent block became slower as the stimulation rate was increased. The slope of the curves relating the intensity of the use-dependent block to the frequency of stimulation was not as steep as those described with lignocaine (Chen & Gettes, 1976; Hondeghe & Katzung, 1980), amiodarone (Varro *et al.*, 1985) and mexiletine (Campbell, 1983a), but flat like those of flecainide, lorainide (Campbell & Vaughan Williams, 1983), propafenone (Kohlhardt & Seifert, 1983) and 5-hydroxypropafenone (Valenzuela *et al.*, 1988). Moreover, at driving rates (2 and 3 Hz) that reduced V_{max} by about 50%, the onset rate of the use-dependent V_{max} block per action potential was 0.05 ± 0.009 AP⁻¹. These values are similar to those for procainamide which has been proposed as an intermediate kinetics class I drug by Campbell (1983a,b).

The recovery from use-dependent V_{max} block (reactivation) by E-3753 was far slower than those of most antiarrhythmic drugs (Campbell, 1983a,b; Courtney, 1980a,b; 1987), probably reflecting the unbinding of the drug from inactivated sodium channels. In fact, the values found in this study are

intermediate between those described with CCI 22277 (Campbell, 1982) and encainide (Campbell, 1983a). E-3753 also shifted the membrane responsiveness curve along the voltage axis to more negative membrane potentials, indicating a greater inhibition of V_{max} at less negative potentials. This effect suggests that this drug possesses a somewhat enhanced affinity for the inactivated state of the sodium channel. However, voltage-clamp studies are required to determine whether E-3753 has a higher affinity for the open and/or inactivated state of the channel. Thus, it can be predicted that E-3753 might cause greater inhibition of excitability and conduction velocity in depolarized cardiac tissues due to ischaemia or other pathological conditions than in normally polarized cardiac tissues. Therefore, since E-3753 exhibits slow onset and offset kinetics of use-dependent block, it can be classified as a kinetically slow class Ic agent according to the classification of Vaughan Williams (1984). The kinetics of the onset and particularly of the offset seem to correlate reasonably well with the physicochemical properties of the drug (Courtney, 1980a,b; Sada & Ban, 1981; Campbell, 1983a). It has been demonstrated that binding and unbinding of class I antiarrhythmic drugs to the receptor site are favoured by low molecular weight and high lipophilicity and inhibited by ionization (Courtney, 1980a,b; 1987). The present data seem consistent with this hypothesis, since E-3753 exists mostly in an ionized form at physiological pH and has a larger molecular weight (399) compared to class I antiarrhythmic drugs with intermediate and fast kinetics (Campbell, 1983a; Courtney, 1987).

Class I antiarrhythmic drugs characterized by a slow onset of and recovery from V_{max} block are often very effective in suppressing both early and late premature beats since they will find a significant fraction of non-conducting sodium channels (Hondeghe & Katzung, 1980; 1984). Moreover, in the presence of tachyarrhythmias sodium channels

are in activated and/or inactivated states for a longer period and there is less time for recovery from block between beats. Thus, it would be possible to select a dose of an antiarrhythmic drug with long τ_{re} (i.e., E-3753) which suppressed conduction in the presence of tachycardia but produced minimal changes at normal sinus rates (Hondeghe & Katzung, 1984; Tamargo *et al.*, 1988).

The present data also reveal that E-3753 shortens the APD. This shortening might result from: (1) a decrease in the slow inward Ca current, (2) an inhibition of the slow inward Na 'window current' flowing during the plateau (Carmeliet & Saikawa, 1982) and/or (3) an increase in the time-dependent outward K current (Carmeliet & Vereecke, 1979). The first possibility was investigated using slow action potentials. E-3753 was without effect on the slow action potentials elicited by isoprenaline in K-depolarized fibres, thus suggesting that it did not exhibit class IV activity in guinea-pig papillary muscles. A more likely explanation for the shortening in APD would be a reduction in the Na current flowing during the plateau. This current is thought to utilize the rapid Na channel forming the basis for a relationship between V_{max} inhibition and APD shortening (Carmeliet & Saikawa, 1982), which can thus be considered to be a manifestation of the class I action exhibited by E-3753. However, an increase of outward K current may also be responsible for the shortening of the APD, but this possibility was not investigated in this study.

In conclusion, on the basis of the present results obtained in guinea-pig atrial and ventricular muscle fibres, E-3753 can be classified as class I antiarrhythmic agent with effects similar to those previously described for class Ic drugs.

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