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MS-551 and KCB-328, two class III drugs aggravated adrenaline-induced arrhythmias

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1 We investigated the proarrhythmic effects of MS-551 and KCB-328, class III antiarrhythmic drugs using adrenaline-induced arrhythmia models in halothane anaesthetized, closed-chest dogs. In the control period, adrenaline, starting from a low dose of 0.25 to up to 1.0 μg/kg/50 s i.v., was injected to determine the arrhythmia inducing dose and the non-inducing dose. After MS-551 or KCB-328 administration, the adrenaline injection was repeated and the interval between the injection and the occurrence of arrhythmia (latent interval), the changes in arrhythmic ratio (as calculated by dividing the number of ventricular premature contraction by the number of the total heart rate) and the severity of arrhythmia were observed.

2 MS-551 infusion, 1 mg/kg/30 min, decreased the heart rate (HR) by 16% ($P<0.01$) and prolonged the QTc interval by 20% ($P<0.01$). During the 30 min of MS-551 infusion, arrhythmias occurred in three out of seven dogs (torsades de pointes (TdP) type VT in one dog). After these arrhythmias disappeared, MS-551 decreased the latent interval of the adrenaline arrhythmias produced by the inducing dose (30 ± 2 s compared with 43 ± 3 s of the control interval, $P<0.05$), increased the arrhythmic ratio ($P<0.05$) and induced arrhythmias by non-inducing adrenaline doses ($P<0.05$).

3 Effect of a new class III drug KCB-328 infusion, 0.3 mg/kg/30 min, was compared witih MS-551 using the same model. KCB-328 decreased the HR by 21% ($P<0.01$) and prolonged the QTc interval by 25% ($P<0.01$). During the 30 min of infusion, arrhythmias occurred in five out of seven dogs (TdP in two dogs). KCB-328 also decreased the latent interval of the adrenaline arrhythmias produced by the inducing doses (31 ± 3 s compared with 49 ± 7 s of the control period, $P<0.05$), but did not significantly alter the arrhythmic ratio.

4 Adrenaline induced TdP only after MS-551 or KCB-328 was administered, i.e. after MS-551, 1 mg/kg/30 min, 3/7 versus 0/7 in the control; KCB, 0.3 mg/kg/30 min, 3/7 versus 0/7 in the control.

5 To examine the direct arrhythmogenic effect of MS-551 and whether an adrenergic mechanism plays some role on this arrhythmogenesis, a bolus injection of MS-551, 3 mg/kg, was injected either without pre-treatment or after pre-treatment with propranolol 0.3 mg/kg. MS-551 induced arrhythmias in five out of seven dogs (TdP in one dog). Also in the propranolol pre-treated dogs, MS-551 induced arrhythmias in five out of seven dogs (TdP in 1 dog).

6 In conclusion, these observations indicate that MS-551 and KCB-328 induced arrhythmias and intensified proarrhythmic effects of adrenaline, MS-551 being stronger than KCB-328 at the same QTc prolonging doses. The direct arrhythmogenic effect of MS-551 was not influenced by β-blocker treatment.

Keywords: Proarrhythmia; class III antiarrhythmic drugs; MS-551; KCB-328; adrenaline; heart; dog

Introduction

Class III antiarrhythmic agents can slow or terminate narrow excitable gap re-entry arrhythmias by lengthening cardiac repolarization and refractoriness. Conversely, prolongation of repolarization can be associated with proarrhythmic actions mainly inducing ventricular premature contraction (VPC) and, at the worst, TdP tachycardia or VF (Hondeghem & Snyders, 1990; Borggrefe *et al*., 1992), so the use of these drugs is difficult for clinical use. For observation of proarrhythmic actions of drugs, to date, animal experimental models have been used to observe induction of arrhythmias by ambulatory ECG recording in animals with arrhythmogenic substrates, or inducing arrhythmias by exercise stress or programmed electrical stimulation (PES). We have used the adrenaline-induced arrhythmia model to observe effects of antiarrhythmic drugs, and the results were as follows; class II β-blockers and class IV Ca channel blocking drugs suppressed adrenaline-induced arrhythmias, but class I and III drugs have different profiles on these arrhythmias. Among class III drugs, amiodarone suppressed adrenaline-induced arrhythmia; d-solatol and dofetilide had no effect on this arrhythmia; and E-4031 and sematilide even aggravated adrenaline-induced arrhythmias (Awaji *et al*., 1995; Hashimoto *et al*., 1991; Chen *et al*., 1996; Xue *et al*., 1996). Adrenaline administration mimics a clinical situation of sympathicotonia, and human long QT syndrome is often associated with sudden death when sympathetic overactivity is present. Indeed it has been demonstrated that class III drugs, clofilium and d,l-solatol provoked early afterdepolarization and/or delayed after depolarization in canine Purkinje fibers under adrenaline adminstration (Patterson *et al*., 1997). Though it is not clear whether proarrhythmic effects of class III drugs correlate drug-induced worsening of the adrenaline-induced ventricular arrhythmia, we designed the present study to test the proarrhythmic effects of new class III drugs.

A class III drug, MS-551 (1,3-dimethyl-6-{2-[N-(2-hydroxyethyl)-3-(4-nitrophenyl)-propylamino]ethylamino}-2,4 (1H, 3H)-pyrimidinedione hydrochloride) differs from most methylsulfonamide class III drugs (e.g., sotalol, E-4031 and dofetilide) in that MS-551 blocks the delayed rectifier K current (Ik), the transient outward K current (Ito), the inward

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rectifier K current (Ik1), and ATP-sensitive K current (Ikatp) in rabbit and guinea-pig ventricular myocytes (Nakaya *et al*., 1993; Hashimoto *et al*., 1994; Sato *et al*., 1995; Martin *et al*., 1995). MS-551 decreased the reperfusion induced VF in dogs (Hashimoto *et al*., 1995), protected postinfarcted dogs from VF (Friedrichs *et al*., 1995), improved defibrillation efficacy when it was given after the induction of VF in dogs (Murakawa *et al*., 1997), suppressed PES-induced ventricular arrhythmias through prolongation of the effective refractory period (ERP) in dogs with myocardium infarction (Kondoh *et al*., 1994), and was useful for the treatment of tachyarrhythmias by prolonging ERP of the atrium and ventricle in the human (Isomoto *et al*., 1995). Though MS-551 induced ventricular arrhythmias in halothane anesthetized dogs (Hashimoto *et al*., 1995), the precise mode of proarrhythmic effects of MS-551 has not been examined.

KCB-328, 1-(2-amino-4-methanesulfonamidophenoxy)-2-[N-(3,4-dimethoxyphenethyl)-N-methylamino]ethane hydrochloride, is a new synthetic class III antiarrhythmic drug. The electrophysiological studies indicated that KCB-328 increased the action potential duration (APD) and ERP in isolated guinea pig papillary muscles and inhibited Ik without a significant inhibition of Isi, but its APD prolonging effect did not show a reverse frequency-dependency (Lee *et al*., 1996a,b). We investigated the antiarrhythmic effect of KCB-328 in *in vivo* dogs and showed that it suppresses VF induced by coronary artery ligation and reperfusion, and PES-induced ventricular arrhythmias in dogs with old myocardial infarction (Xue *et al*., in press).

The lethal arrhythmias accompanying the long QT syndrome are serious problems for both the patients with congenital long QT syndrome and also for those using class III drugs, and β-blocker therapy is one choice for the treatment of the former. We examined the proarrhythmic effect of the two class III drugs in combination with adrenaline to assess the safety of these drugs. In the present study, we examined (1) whether MS-551 and KCB-328 aggravate adrenaline arrhythmias in dogs and (2) whether a β-blocker, propranolol, influences the proarrhythmic effect of MS-551.

Methods

Experimental preparation

These animal experiments were approved by the Yamanashi Medical University Animal Experimentation Committee and animals were obtained through the Animal Laboratory for Research of Yamanashi Medical University. Adult beagle dogs of either sex weighing 7.5–13.5 kg, was anaesthetized initially with thiopental sodium. After tracheal intubation, 1.0% halothane, vaporized with 100% oxygen, were administered with a volume-limited ventilator (20 ml/kg, 15 strokes/min, Shinano, SN-480-4, Tokyo, Japan). Both vagi was cut at the mid-cervical region. The lead II ECG and atrial electrogram from catheter tip electrodes in the right atrium were continuously monitored. A femoral artery catheter was inserted for blood pressure (BP) monitoring. The ECG, atrial electrogram and BP were recorded with a polygraph system (Nihon Kohden, Tokyo, Japan). A femoral vein was also cannulated for administering drugs and adrenaline.

Production of adrenaline-induced arrhythmia

After surgical preparation, 30–45 min was allowed for stabilization, and then adrenaline diluted in 5 ml saline was intravenously infused for 50 s, according to the method of Hashimoto *et al*. (Hashimoto & Hashimoto, 1972; Matsubara *et al*., 1976). The starting dose of adrenaline was 0.5 μg/kg. If 0.5 μg/kg adrenaline did not produce arrhythima, the dose of adrenaline was increased by increments of 0.25 or 0.5 μg/kg until ventricular arrhythmias were induced. The maximum adrenaline dose in the control period was 2.5 μg/kg, since this dose usually produces severe VT or occasionally fatal VF. If 0.5 μg/kg adrenaline produced arrhythmia, a lower dose of 0.25 μg/kg adrenaline was infused. Between the challenges of the adrenaline infusion, a recovery period of at least 10 min was allowed, during which time the hemodynamic parameter, e.g. HR and BP became stable. We defined the arrhythmia inducing dose of adrenaline as the lowest dose which produced ventricular arrhythmias, including VPC, bigeminy or VT, and the non-inducing dose of adrenaline as the highest dose which did not induce any arrhythmia.

The time in seconds between the onset of adrenaline infusion and the appearance of arrhythmia was defined as the latent interval. After MS-551 or KCB-328 administration and when the QT prolongation became stable, arrhythmia non-inducing and inducing doses were determined.

Protocol

In pilot experiments ($n=3$), the non-inducing and inducing doses of adrenaline before 30 min of saline infusion were 0.5, 0.5 and 0.75 μg/kg (0.58 ± 0.08 μg/kg), and 0.75, 0.75 and 1.0 μg/kg (0.83 ± 0.08) μg/kg, respectively. The arrhythmias of the inducing doses were VPC ($n=2$) and VT ($n=3$). After 30 min of saline infusion, the non-inducing and inducing doses of adrenaline were unchanged, namely 0.5, 0.5 and 0.75 μg/kg (0.58 ± 0.08 μg/kg), and 0.75, 0.75 and 1.0 μg/kg (0.83 ± 0.08) μg/kg, respectively, and the arrhythmias were VPC ($n=2$) and VT ($n=3$), indicating the reproducibility of these values.

As shown in Figure 1, experiments were divided into three groups. In experiment 1, an infusion rate of MS-551, 1 mg/kg/30 min, was chosen as that prolonging the QTc interval approximately 20%. If this infusion of MS-551 alone induced arrhythmias, the inducing and non-inducing adrenaline doses were determined after disappearance of these arrhythmias. In experiment 2, an infusion rate of KCB-328, 0.3 mg/kg/30 min was chosen as that prolonging the QTc interval also approximately 20%. In experiment 3, the direct arrhythmogenic effects of MS-551 were examined using a bolus injection of MS-551, 3 mg/kg, without or after treatment with

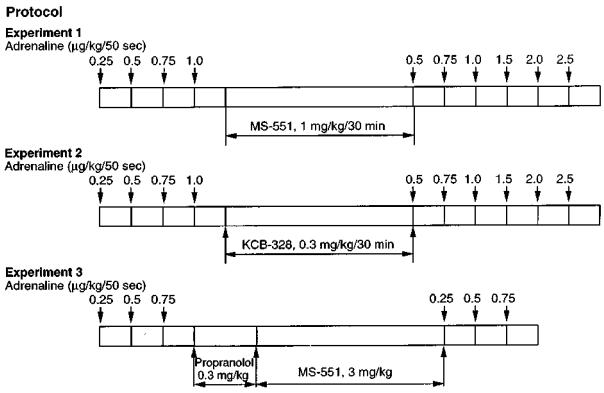

Figure 1 Schematic of experimental protocol of MS-551 and KCB-328 on adrenaline-induced arrhythmias in dogs.

propranolol 0.3 mg/kg. Adrenaline 0.25–0.75 μg/kg/50 s was administered to verify the blocking effect of propranolol.

Drugs

MS-551 and KCB-328 were kindly provided by Mitsui Pharmaceuticals Inc., Tokyo, Japan and C&C Research Laboratories, Kyunggi-do, Korea, respectively. MS-551 and KCB-328 was dissolved in saline. Thiopental sodium (Tanabe Seiyaku, Tokyo), halothane (Takeda Chemical Industries, Osaka) and adrenaline (Daiichi Seiyaku Co., Tokyo) were used.

Evaluation of proarrhythmic effects

The arrhythmic ratio was calculated by dividing the number of ventricular premature contractions by the number of the total heart rate, i.e., the number of VPC plus the number of conducted beats, and the ventricular beats were judged by the different shape of the ventricular complex from the normal QRS complex. VPCs was defined as discrete and identifiable premature QRS complexes (premature in relation to the P wave). VT was defined as a run of four or more consecutive ventricular premature contractions. VF was defined as a signal for which individual QRS deflections can no longer be distinguished from one another (implying morphological instability) and for which a rate can no longer be measured according to the Lambeth Conventions (Walker *et al*., 1988). TdP was defined as the short run of ventricular tachycardia in which the form and axis of the QRS complex undulate in a sinusoidal fashion around the isoelectric line. If the arrhythmic ratio and the severity of adrenaline arrhythmia after MS-551 or KCB-328 administration were increased significantly from that of the control period, or the non-inducing dose of adrenaline induced new arrhythmias, the two drugs were assumed to aggravate the arrhythmia.

Statistics

The hemodynamic data, the QTc interval, the latent interval of arrhythmia observed by the inducing doses of adrenaline and the arrhythmic ratio are expressed as mean ± standard error (s.e.) of means. The hemodynamic data, arrhythmic ratio and the latent intervals of arrhythmia by the inducing doses of adrenaline before and after drug treatments were subjected to paired Student's *t*-test. The severity of arrhythmias was compared by the Wilcoxon signed-ranks test. Differences were regarded as significant if the *P* values were less than 0.05.

Results

Proarrhythmic effects of MS-551 infusion, 1 mg/kg/30 min, on adrenaline-induced arrhythmias (n = 7)

MS-551, 1 mg/kg/30 min, slightly decreased mean blood pressure (mBP) and HR and prolonged QTc interval by 20%, as shown in Table 1. MS-551, 1 mg/kg/30 min, hastened the occurrence of arrhythmias, i.e., decreased the latent interval of arrhythmias produced by inducing doses of adrenaline (0.93 ± 0.13 μg/kg), 30 ± 2 s compared with 43 ± 3 s of the control period ($P < 0.05$), as shown in Figure 2a. MS-551 also aggravated adrenaline arrhythmias, i.e. increased the arrhythmic ratio attained by 0.5 μg/kg/50 s adrenaline (Figure 3a), and altered non-inducing doses of adrenaline (0.50 ± 0.09 μg/kg) to induce ventricular arrhythmias including VF ($P < 0.05$) (Figure 4a), and tended to aggravate arrhythmias produced by inducing doses of adrenaline ($P > 0.05$) (Figure 5a). The incidence of TdP after MS-551, 1 mg/kg/30 min was 2/7 and 1/7 at non-inducing and inducing doses of adrenaline, respectively. During the 30 min of MS-551 infusion alone, ventricular arrhythmias were induced in three out of seven dogs, the severity of arrhythmias were VPC ($n = 1$) and VT ($n = 2$). TdP appeared in one dog, as shown in Figure 6.

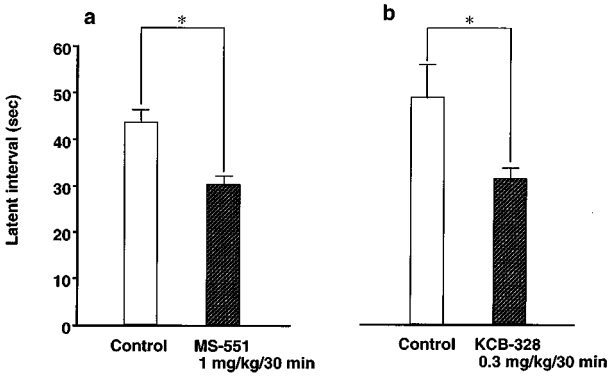

Figure 2 Effects of MS-551 and KCB-328 on the latent interval at the adrenaline inducing dose in dogs. (a) MS-551, 1 mg/kg/30 min group. (b) KCB-328, 0.3 mg/kg/30 min group. Control (open columns) and drug-treatment (hatched columns) were shown in every group. Data shown are the means ± s.e.mean of seven dogs. *$P < 0.05$, by paired Student's *t*-test to compare the latent interval between drug-treatment and control.

Table 1 Effects of MS-551, 1 mg/kg/30 min on hemodynamic and electrophysiologic parameters in dogs

Time	n	*HR (beats/min)*	*BP (mm Hg)*	*QTc interval (ms/s$^{1/2}$)*
Control	7	128 ± 5	126 ± 5	421 ± 16
Pre-MS-551	7	126 ± 5	131 ± 6	427 ± 11
Post-MS-551 (30 min)	7	106 ± 5**	118 ± 7**	512 ± 19**
Before Adr. (0.5 μg/kg)	7	106 ± 5**	120 ± 7*	513 ± 19**
After Adr. (0.5 μg/kg)	5	106 ± 5**	125 ± 6	517 ± 25**
Before Adr. (1.0 μg/kg)	5	105 ± 5**	126 ± 5	520 ± 24**
After Adr. (1.0 μg/kg)	5	109 ± 6**	126 ± 5	514 ± 24**
Before Adr. (1.5 μg/kg)	5	109 ± 6**	126 ± 5	514 ± 24**
After Adr. (1.5 μg/kg)	4	118 ± 8	128 ± 7	503 ± 24*
Before Adr. (2.0 μg/kg)	4	118 ± 8	128 ± 7	503 ± 22*
After Adr. (2.0 μg/kg)	3	120 ± 10	123 ± 11	481 ± 23
Before Adr. (2.5 μg/kg)	3	120 ± 10	124 ± 10	481 ± 23
After Adr. (2.5 μg/kg)	3	120 ± 10	123 ± 11	484 ± 19

HR, heart rate; BP, blood pressure; QTc, corrected QT interval, Adr., adrenaline. Data are mean ± s.e.mean. *$P < 0.05$, **$P < 0.01$ relative to pre-MS-551 value.

Proarrhythmic effects of KCB-328 infusion, 0.3 mg/kg/30 min, on adrenaline-induced arrhythmias (n = 7)

As shown in Table 2, KCB-328, 0.3 mg/kg/30 min, slightly decreased mBP and HR and also prolonged QTc interval by 25%. KCB-328 decreased the latent interval of arrhythmias produced by inducing doses of adrenaline (0.79 ± 0.10 μg/kg), 31 ± 3 s compared with 49 ± 7 s of the control period ($P < 0.05$), as shown in Figure 2b, but did not increase the arrhythmic ratio and the grades of arrhythmia, as shown in Figures 3b, 4b, 5b. The incidence of TdP after KCB-328 was 1/7 and 2/7 by non-inducing and inducing doses of adrenaline, respectively. During the 30 min of KCB-328 infusion alone, arrhythmias occurred in five out of seven dogs, the severity of arrhythmias were VPC ($n = 2$) and VT ($n = 3$). TdP appeared in two dogs, as shown in Figure 6.

Direct arrhythmogenic effects of a bolus injection of MS-551 (n = 7), and effect of propranolol on these effects (n = 7)

After 30 min of injection of MS-551, 3 mg/kg, HR interval decreased from 141 ± 5 to 112 ± 7 beats/min and QTc increased from 369 ± 14 to 490 ± 21 ms/s$^{1/2}$ ($P < 0.01$). Mean BP did not change. Soon after a bolus injection of MS-551, 3 mg/kg alone, arrhythmias were induced in five out of seven dogs and even TdP appeared in one dog. The severity of arrhythmias were VPC ($n = 4$) and VT ($n = 1$). In addition, we investigated whether these arrhythmias are affected by a β-blocker, propranolol treatment. In the control period, HR, mBP and QTc interval were 137 ± 6 beats/min, 122 ± 5 mmHg and 403 ± 12 ms/s$^{1/2}$, respectively ($n = 7$). After 10 min of propranolol injection of 0.3 mg/kg, HR decreased to 115 ± 5 beats/

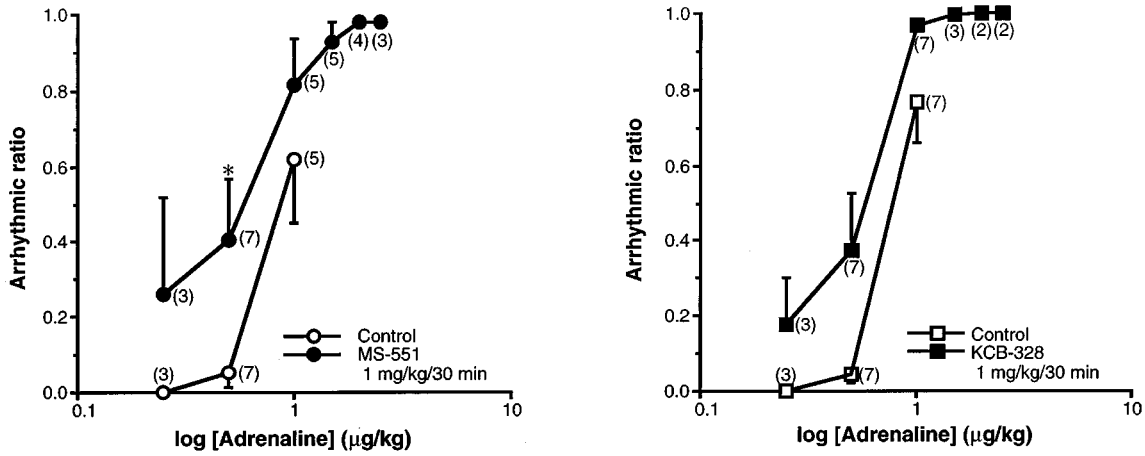

Figure 3 Effects of MS-551 and KCB-328 on the arrhythmic ratio in dogs. (a) MS-551, 1 mg/kg/30 min group. (b) KCB-328, 0.3 mg/kg/30 min group. The arrhythmic ratio in the drug-treated groups were compared with the control group at corresponding adrenaline dose points. Each number in the parenthesis are experimental number. Each data point represents the mean, and vertical lines show s.e.mean of each experiment. *$P < 0.05$, by paired Student's t-test to compare the arrhythmic ratio between drug-treatment and control.

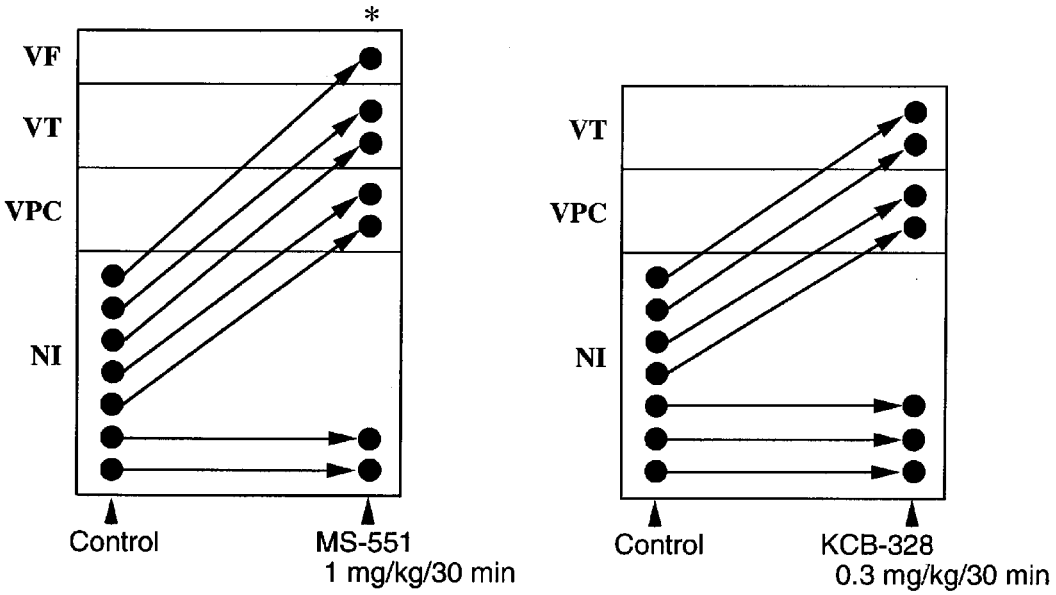

Figure 4 Effects of MS-551 and KCB-328 on the changes of the severity of arrhythmia at the adrenaline non-inducing dose in dogs. (a) MS-551, 1 mg/kg/30 min group ($P < 0.05$). (b) KCB-328, 0.3 mg/kg/30 min group. VF: ventricular fibrillation. VT: ventricular tachycardia. VPC: ventricular premature contraction. NI: non-inducible. *$P < 0.05$, by Wilcoxon signed-ranks test to compare the arrhythmic ratio between drug-treatment and control.

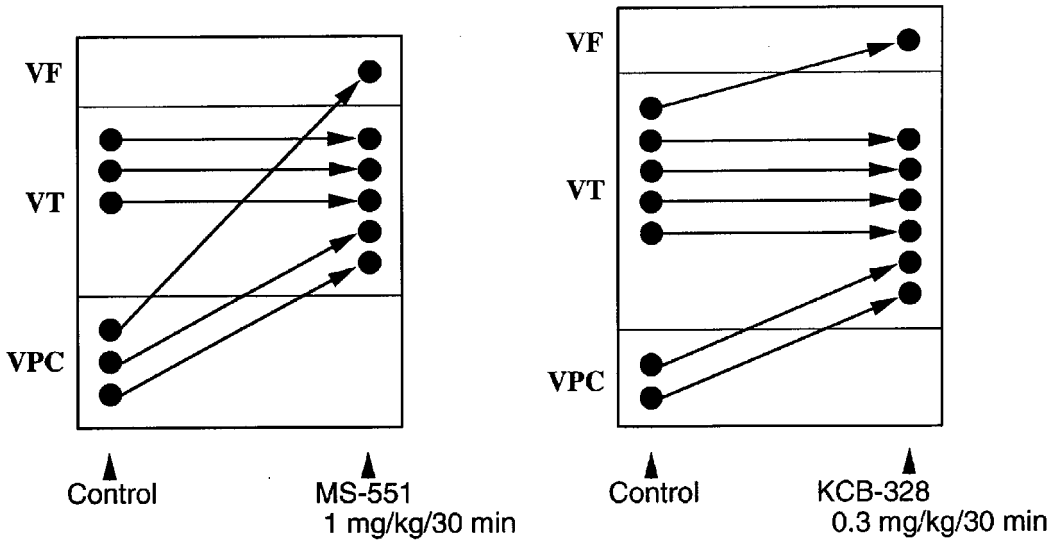

Figure 5 Effects of MS-551 and KCB-328 on the changes of the severity of arrhythmia at the adrenaline inducing dose in dogs. (a) MS-551, 1 mg/kg/30 min group. (b) KCB-328, 0.3 mg/kg/30 min group.

Table 2 Effect of KCB-328, 0.3 mg/kg/30 min on hemodynamic and electrophysiologic parameters in dogs

Time	n	*HR (beats/min)*	*BP (mmHg)*	*QTc interval (ms/s$^{1/2}$)*
Control	7	134±5	128±5	429±4
Pre-KCB-328	7	132±5	129±5	429±5
Post-KCB-328 (30 min)	7	104±4**	117±4**	538±11**
Before Adr. (0.5 μg/kg)	7	104±5**	117±4**	538±11**
After Adr. (0.5 μg/kg)	7	102±5**	121±4	535±13**
Before Adr. (1.0 μg/kg)	7	102±5**	122±5	535±13**
After Adr. (1.0 μg/kg)	3	115±6**	118±6	516±13**
Before Adr. (1.5 μg/kg)	3	115±6**	119±5	515±13**
After Adr. (1.5 μg/kg)	2	123±6	115±5	504±8
Before Adr. (2.0 μg/kg)	2	121±4	115±7	504±8
After Adr. (2.0 μg/kg)	2	121±4	115±5	501±11

HR, heart rate; BP, blood pressure; QTc, corrected QT interval, Adr., adrenaline. Data are mean±s.e.mean. *$P<0.05$, **$P<0.01$ relative to pre-KCB-328 value.

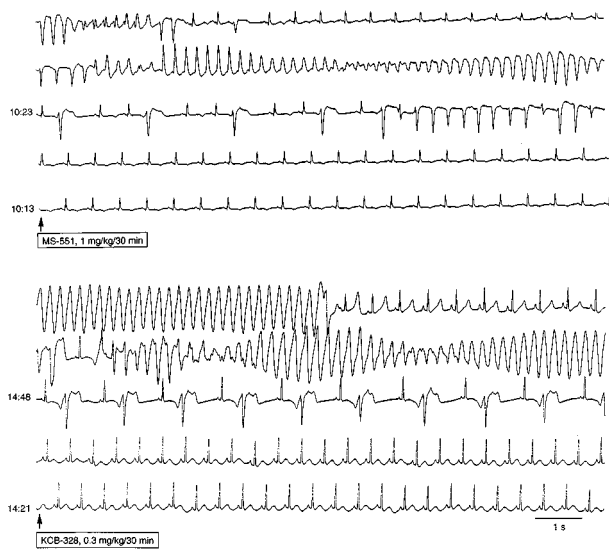

Figure 6 Torsades de pointes in MS-551, 1 mg/kg/30 min (upper) and KCB-328, 0.3 mg/kg/30 min (lower) treated dogs.

min ($P<0.05$), but mBP and QTc were unchanged, i.e. 118±5 mmHg and 409±13 ms/s$^{1/2}$, respectively. After 30 min of 3 mg/kg MS-551 under propranolol treatment, QTc interval was prolonged to 549±22 ms/s$^{1/2}$, and HR decreased to 85±8 beats/min, but mBP was unchanged. Under propranolol administration, arrhythmias were also induced by MS-551 in five out of seven dogs, the severity of arrthymias were VPC ($n=1$), VT ($n=1$) and VF ($n=3$). TdP appeared in one dog. The severity of the ventricular arrhythmias induced by MS-551 were not affected by propranolol ($P>0.05$). At the end of these experiments, HR was not changed by 0.25–0.75 μg/kg/50 s adrenaline administration.

Discussion

In the present study, we tried to demonstrate proarrhythmic effects of class III drugs using halothane anaesthetized dogs. MS-551 not only aggravated halothane-adrenaline arrythmia, but also MS-551 alone induced arrhythmias. The latter arrhythmogenic effect was not blocked by a β-adrenergic receptor antagonist, propranolol. A new Ikr blocking class III

drug, KCB-328, did not aggravate the halothane-adrenaline arrhythmia as much as MS-551, i.e. did not significantly increase the arrhythmic ratio even though the doses were chosen to prolong QT interval at the same extent, but the latent interval of adrenaline arrhythmia was shortened. KCB-328 alone also induced arrhythmias, like MS-551. The incidence of TdP was markedly increased when adrenaline was injected after MS-551 or KCB-328 administration.

Since the early 1980s, it has been recognized that all antiarrhythmic drugs have proarrhythmic potentials to aggravate preexisting tachyarrhythmias or to provoke new ones (Velebit *et al*., 1982; Poser *et al*., 1985; Rae *et al*., 1988; Minardo *et al*., 1988). So far proarrhythmia has been defined on the basis of an increase in the number of spontaneous VPC as documented either by ambulatory ECG recording (The CASCADE Investigators, 1993; Poser *et al*., 1985), exercise stress testing (Rae *et al*., 1988), or PES (Rae *et al*., 1988; Minardo *et al*., 1988; Morganroth *et al*., 1987; Podrid, 1985). Recently, in a number of clinical studies, the proarrhythmia, especially the proarrhythmic effects of class I and III antiarrhythmic agents, has been emphasized due to a significant increase in the number of symptomatic arrhythmias.

To investigate the proarrhythmic effects of class III drugs, we used halothane anaesthetized dogs, which have a low heart rate compared to pentobarbital anaesthetized dogs. It is well known that the arrhythmogenic action of adrenaline is enhanced in the presence of a hydrocarbon inhalation anaesthetic, such as halothane, and the mechanism of these arrhythmias is thought to be abnormal automaticity and triggered activity (Hashimoto & Hashimoto, 1972; Hashimoto *et al*., 1982; Wit & Rosen, 1992), and halothane is known to interfere with the cell to cell coupling (Hashimoto & Hashimoto, 1972). β-Adrenergic catecholamines have various electrophysiological effects. They increased I_{Ca} which must enhance ectopic automaticity, but has other effects such as increasing I_k (Giles *et al*., 1989), I_{to} (Nakayama & Fozzard, 1988), chloride current (I_{Cl}) (Harvey & Hume, 1989). α-Adrenergic agonists increase APD of certain cardiac cells, an effect recently demonstrated to result from an α_1-adrenoceptor-mediated decrease in I_{to} (Fedida *et al*., 1989). MS-551 has been reported not only to inhibit I_k as KCB-328, but also I_{k1}, I_{to} and I_{kATP} in ventricular myocytes (Nakaya *et al*., 1993; Hashimoto *et al*., 1994; Sato *et al*., 1995; Martin *et al*., 1995), and inhibit I_{kAch} in atrial cells (Mori *et al*., 1995). Thus it is difficult to predict the combined effect of catecholamines and class III drugs on the cardiac cell APD. Actually MS-551 aggravated adrenaline-induced arrhythmias in the present study.

The proarrhythmic effects of MS-551 and KCB-328 in the present study may be due to the following causes; they may be class III drug-induced early after depolarization (EAD), EAD induced triggered activity enhanced by exogenous adrenaline, and enhanced depolarizing current of the ectopic heart. In addition, the reverse use dependency of class III drug action may also be arrhythmogenic, because at slower heart rates, the excessive increase in APD is likely to give rise to EADs.

The incidence of adrenaline-induced TdP was increased after the class III drug administration, where MS-551 infusion, 1 mg/kg/30 min, prolonged the QTc interval by 20% and KCB-328 infusion, 0.3 mg/kg/30 min, by 25%. Clinically, isoprenaline or adrenaline infusions induced TdP in a patient with long QT syndrome (Kawade *et al*., 1995; Shimizu *et al*., 1995). A computer simulation of TdP has identified the mechanism and shown that the adrenergic effects of reduced refractory period and increased heart rate may lead to the initiation of TdP (Abildskov & Lux, 1997).

Some studies demonstrated that propranolol and other β-blockers are effective for symptoms and ventricular arrhythmias in patients with long QT syndrome (Garson *et al*., 1993; Leenhardt *et al*., 1995), and the β-blocker may be useful in preventing arrhythmogenic effects of antiarrhythmic drugs under an augmented sympathetic nervous system tone (Sager *et al*., 1994; Sanguinetti *et al*., 1991; Sager & Behboodikhah, 1996). Though the mechanism of the aggravating effect of class III on adrenaline induced arrhythmia was not identified, there may be no β-stimulating effect in these class III drugs. In order to prove this, we tested the proarrhythmic effects of MS-551 alone under propranolol administration. The dose of 0.3 mg/kg propranolol is considered to have no effects on sodium channels. In the present study, MS-551, 3 mg/kg, induced arrhythmias in five out of seven dogs regardless of propranolol administration, showing that propranolol does not prevent MS-551-induced arrhythmias. This suggests that different mechanisms are operating in class III drug induced proarrhythmia and that with long QT syndrome.

In conclusion, MS-551 aggravated adrenaline-induced arrhythmias, and arrhythmias induced by MS-551 were not affected by a β-adrenergic receptor antagonist, propranolol. KCB-328 also aggravated the adrenaline-induced arrhythmia, but the extent of arrhythmogenicity was weaker than MS-551. MS-551 and KCB-328 alone induced arrhythmias, so in the clinical use of class III drugs, an increase of the sympathetic activity may be deleterious in inducing TdP.

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