# Antiarrhythmic and Proarrhythmic Effects of Sematilide in Canine Ventricular Arrhythmia Models

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ABSTRACT— Using canine ventricular arrhythmia models induced by two-stage coronary ligation, digitalis, adrenaline, coronary ligation and reperfusion, and programmed electrical stimulation (PES), we examined the antiarrhythmic or proarrhythmic effects of sematilide, *N*-[2(diethylamino)ethyl]-4-[(methylsulfonyl)amino]-benzamide hydrochloride. Sematilide in an intravenous (i.v.) dose range of 0.3 to 6.0 mg/kg prolonged the QTc interval, but had an antiarrhythmic effect only on the arrhythmias induced by PES (7 out of 10 dogs with old myocardial infarction). Sematilide was not effective on coronary ligation and reperfusion arrhythmia or the spontaneously occurring automaticity arrhythmias induced by two-stage coronary ligation, digitalis and adrenaline, respectively, and even aggravated digitalis- and adrenaline-induced arrhythmias. These results indicate that the class III agent sematilide is similar to other new class III agents, such as *d*-sotalol, E-4031 and MS-551, in that it was not effective on the automaticity arrhythmias, but different from these new class III agents, in that sematilide prevented only the induction of ventricular arrhythmias induced by PES and did not suppress the coronary ligation-reperfusion arrhythmias. The PES induced arrhythmias are thought to be induced exclusively by a reentrant mechanism, but the reperfusion arrhythmias may involve not only re-entry, but also automaticity, and we reported the effectiveness of MS-551, E-4031 and *d*-sotalol on the latter arrhythmia. Sematilide is different in that it even aggravated some automaticity arrhythmias.

*Keywords*: Sematilide, Ventricular arrhythmia, Ischemia, Reperfusion arrhythmia, Programmed electrical stimulation

Ventricular arrhythmia is one of the serious problems for patients with coronary artery diseases or myocardial infarction (1) and has been pharmacologically controlled either by slowing conduction and suppressing ventricular automaticity (e.g., class I effects) or by prolonging refractoriness (class III effects) with or without adrenergic modulation (2-4). Given the unfavorable outcome of the Cardiac Arrhythmia Suppression Trial using class I antiarrhythmic agents (5), i.e., the heightened concern regarding proarrhythmic effects of class I agents (6), and also the relatively high level of efficacy associated with amiodarone therapy (7, 8), there has recently been a marked interest in controlling ventricular arrhythmias with agents that exert "pure" class III effects.

Sematilide (*N*-[2(diethylamino)ethyl]-4-[(methylsulfonyl)amino]-benzamide hydrochloride) was synthesized based on the structure-activity relationships of procainamide to get a specific class III agent. Electrophysiological studies indicated that sematilide specifically inhibits the rapid component of delayed rectifier $K^+$ current ($I_{Kr}$), resulting in prolongation of repolarization and refractoriness (9, 10). It has already been reported that sematilide can effectively suppress sustained ventricular tachycardia induced by programmed electrical stimulation (PES) in dogs with previous myocardial infarction (11, 12).

We have studied and classified antiarrhythmic drugs based on their pharmacological effects on canine ventricular arrhythmia models (13) produced by coronary ligation-reperfusion, two-stage coronary ligation, digitalis and adrenaline. Using the same models, we have reported the effects of class III drugs, intravenous amiodarone, *d*-sotalol, E-4031 and MS-551. Except for amiodarone, the class III drugs were not effective on automaticity arrhythmias produced by two-stage coronary ligation, digitalis and adrenaline, but suppressed the ventricular fibrillation (VF) immediately after reperfusion of the coronary artery. To characterize the antiarrhythmic

profile of sematilide, including demonstration of efficacy and at the same time of aggravating potentials, we examined the effects of sematilide and compared them with those of other class III drugs, using the PES-induced arrhythmia model in addition to the above-mentioned models.

## MATERIALS AND METHODS

*Production of two-stage coronary ligation-induced arrhythmia*

Six beagle dogs of either sex, weighing 8.0–11.0 kg, were anesthetized initially with i.v. thiopental sodium, 30 mg/kg, and intubated. As reported earlier (14), the chest was opened and a two-stage coronary ligation of the left anterior descending artery (LAD) was performed under halothane anesthesia.

Experiments were done without anesthesia 48 hr after coronary ligation. The lead II ECG, atrial electrogram from implanted electrodes sutured on the left atrial appendage, and the instantaneous and mean blood pressure were recorded continuously using a telemetry system (Nihon Kohden, Tokyo). In the preliminary experiments, doses of sematilide ranging from 0.3 to 10 mg/kg/10 min were examined. The dose of 3 mg/kg/10 min was chosen, because it prolonged the QT interval maximally without producing proarrhythmic effects. A constant rate infusion of sematilide, 3 mg/kg, was performed for 10 min using a syringe pump (Terumo, Tokyo); and arterial blood samples were drawn from one lumen of the arterial double lumen cannula at 0, 5, 10, 30 and 60 min after the start of the infusion.

*Production of digitalis-induced arrhythmia*

Twelve beagle dogs of either sex, weighing 9.5–11.0 kg, were anesthetized with intravenous pentobarbital sodium, 30 mg/kg, and divided into two groups; one group received 1 mg/kg sematilide and the other 3 mg/kg. As reported earlier (15), 40 μg/kg ouabain was injected intravenously and then followed by an additional 10 μg/kg every 20 min until stable ventricular tachycardia was produced. A constant rate infusion of sematilide, 1 or 3 mg/kg, was performed for 10 min using a syringe pump; and arterial blood samples were drawn from one lumen of the arterial double lumen cannula at 0, 5, 10, 30 and 60 min after the start of infusion.

The lead II ECG, atrial electrogram from catheter tip electrodes in the right atrium and the instantaneous and mean blood pressure were continuously recorded.

*Production of adrenaline-induced arrhythmia*

Six beagle dogs of either sex, weighing 10.0–11.0 kg, were anesthetized initially with intravenous thiopental sodium. As reported earlier (16), after intubation, 1.0% halothane, vaporized with 100% oxygen, was administered with a volume-limited ventilator (20 ml/kg, 15 strokes/min). Adrenaline was infused through the left femoral vein at a rate of 2.5 μg/kg/min using a syringe pump. If multifocal ventricular tachycardia was not induced, a higher infusion rate was employed. At 3 min after the start of adrenaline infusion, a constant rate infusion of sematilide, 0.3 mg/kg, was performed for 10 min using a syringe pump; and arterial blood samples were drawn from one lumen of the arterial double lumen cannula at 0, 5, 10, 15 min after the start of infusion.

The lead II ECG, atrial electrogram from cathether tip electrodes in the right atrium and the instantaneous and mean blood pressure were continuously recorded.

*Production of coronary ligation and reperfusion arrhythmia*

Thirty-nine beagle dogs of either sex, weighing 6.0–13.0 kg, were used. Experimental groups were divided into three groups. Dogs of group 1 were anesthetized with halothane, while dogs of groups 2 and 3 were anesthetized with pentobarbital. Dogs of groups 1, 2 and 3 were treated with 3, 3 and 6 mg/kg/10 min of sematilide, respectively. Dogs of group 1 were anesthetized initially with intravenous thiopental sodium, 30 mg/kg, and then intubated. Anesthesia was maintained by 1.0% halothane, vaporized with 100% oxygen using a volume-limited ventilator (20 ml/kg, 15 strokes/min). Dogs of groups 2 and 3 were anesthetized with intravenous pentobarbital sodium at 30 mg/kg, followed by an infusion of 5 mg/kg/hr.

In all three groups, the chest was opened and the LAD was isolated just proximal to the first diagonal branch. Since the incidence of occurrence of coronary ligation-reperfusion arrhythmia is known to be quite variable, experiments were randomized using a pair of beagles [by coin-flip]; one received drug infusion and the other received 0.9% NaCl infusion. The speed of infusion was 1 ml/min. After 30 min from the start of either sematilide or saline infusion when the change in QTc became stable, the LAD ligation was performed using a silk thread and 30 min later released to examine reperfusion responses. The drug solution was infused for 10 min.

A pair of epicardial electrodes was sutured on the border zone of the ischemic area of the left ventricle for continuous recording of the ventricular electrograms. The QT interval was assessed from the lead II ECG and the ventricular surface electrogram. The QTc interval was calculated by Bazett's formula, $QTc = QT/\sqrt{RR}$. The heart rates were measured from the lead II ECG, and the blood pressure was continuously monitored through a double lumen arterial cannula in the femoral artery. Arterial blood samples were obtained through another lu-

men of the cannula just before 1) the start of sematilide infusion, 2) LAD occlusion and 3) LAD reperfusion.

*Production of PES-induced arrhythmias*

Ten beagles of either sex weighing 8.0−11.5 kg were anesthetized initially with intravenous thiopental sodium, 30 mg/kg, and intubated. The chest was opened and a two-stage coronary ligation of the LAD was performed under halothane anesthesia, and then the chest was closed. Seven to twelve days after surgery, when myocardial infarction was established, the dogs were anesthetized initially with intravenous pentobarbital sodium, 30 mg/kg, and intubated. The chest was again opened under continuous administration of pentobarbital sodium (5 mg/kg/hr) through a cannula in the femoral vein. PES was performed through the stainless-steel bipolar plunge electrodes that were sutured on the non-infarcted myocardium of the left ventricular wall close to the myocardial infarct zone. A silver-coated copper wire was connected to a programmable stimulator (SS-201J, Nihon Kohden) through an isolator (SEN-7203, Nihon Kohden). The lead II ECG, instantaneous and mean blood pressure from a cannula in the femoral artery were continuously recorded.

The basic pacing interval (S1) was set at 300 msec, which was shorter than the cycle length of the spontaneous heart rate. The excitation threshold measurement was preceded by each PES series. After a train of 15 pacing stimuli, a single extrastimulus (S2) was delivered by shortening the S1−S1 interval in a 5 msec-step until arrhythmia or refractoriness occurred. If S2 failed to induce arrhythmias, double extrastimuli (S2 and S3) were delivered with the S1−S2 interval just exceeding the effective refractory period (ERP). S3 was introduced initially at the same interval as S2, which was then decreased again in 5-msec steps. The stimulation protocol was fixed once VF or sustained ventricular tachycardia (SVT) or non-sustained VT (NSVT) or ventricular premature contraction (VPC) was induced or the refractary period was reached.

Arrhythmias that occurred at least twice during the control stimulation protocol were used further for pharmacological analysis. The dogs with SVT and VF were recovered by using a DC defibrillator. Sematilide at 3 mg/kg/10 min was given and after 30 min of the start of infusion, the same stimulation protocol was examined as before. Consecutive VPC of more than 3 is defined as ventricular tachycardia (VT). NSVT is defined as VT lasting less than 30 sec; SVT is a VT lasting more than 30 sec.

The present experiment was approved by the committee on animal experimentation and the animal use and care committee of Yamanashi Medical University.

*Drugs*

The following drugs were used: thiopental sodium (Tanabe Seiyaku, Tokyo), halothane (Takeda Chemical Industries, Osaka), pentobarbital sodium (Tokyo Kasei Kogyo, Tokyo), (−)-ouabain octahydrate (Aldrich Chemical, Milwaukee, WI, USA), adrenaline (Daiichi Seiyaku Co., Tokyo) and sematilide (kindly supplied by Nippon Roussel Co., Ltd., Tokyo).

*Determination of sematilide plasma levels*

The arterial blood samples were collected into heparinized syringes at predetermined times and centrifuged at $3000 \times g$ for 5 min. The plasma was stored at about −80℃ until its assay. The concentrations of sematilide were determined using a high performance liquid chromatographic method in the research laboratory of Nippon Roussel Co., Ltd. (17).

*Evaluation of antiarrhythmic effects*

The severity of two-stage coronary ligation-, digitalis- and adrenaline-induced ventricular arrhythmias was expressed by the arrhythmic ratio: the number of ventricular ectopic beats divided by the total heart rate, which is the number of all beats counted from the 5 sec strip of ECG (i.e., the number of ventricular ectopic beats 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. The arrhythmic ratio before drug injection was almost 1 as shown in the control values of the figures, and there were no spontaneous improvements in these ratios. If the values after drug administration decreased significantly from the 0 time value, as determined by the analysis of variance (ANOVA) followed by the paired $t$-test ($P<0.05$), the drug was judged as having significant effects.

On the coronary ligation and reperfusion arrhythmia model, ANOVA was used to compare the QTc, heart rate and blood pressure values of the drug treated with those of saline treated experiments. The incidence of arrhythmias was compared using the chi squared test. A P value less than 0.05 was considered significant at each time point.

On the electrical stimulation-induced arrhythmia model, ANOVA was used to compare the QTc, heart rate and blood pressure values before and after the drug treatment. The incidence of arrhythmias was compared using the Wilcoxon signed-ranks test, and when $P<0.05$, the drug was judged as having significant effects.

## RESULTS

*Two-stage coronary ligation-induced arrhythmia*

Forty-eight hours after coronary ligation, all dogs

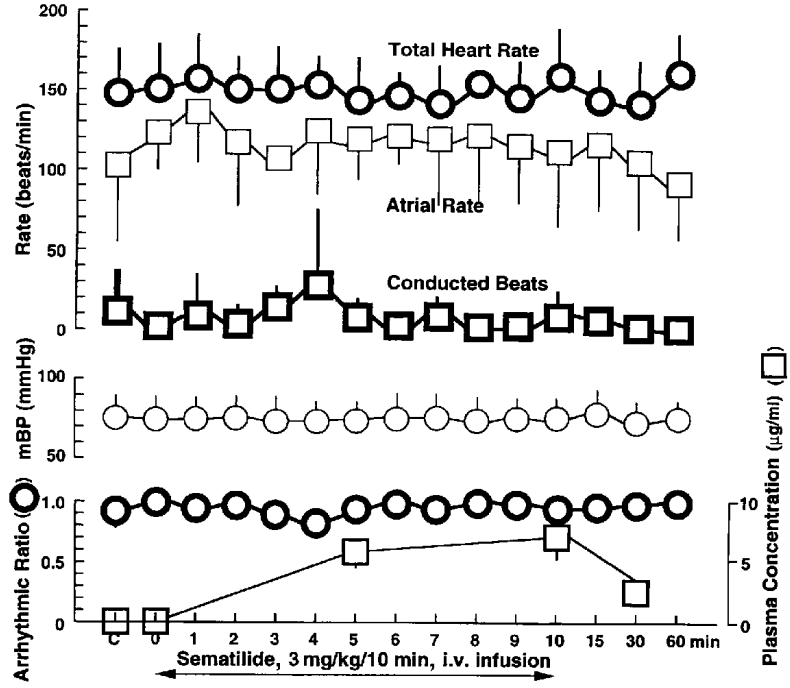


**Fig. 1.** Effects of intravenous infusion of sematilide, 3 mg/kg/10 min, on 48-hr coronary ligation arrhythmia models (n=6). S.D.s are shown at each time point. mBP: mean blood pressure.

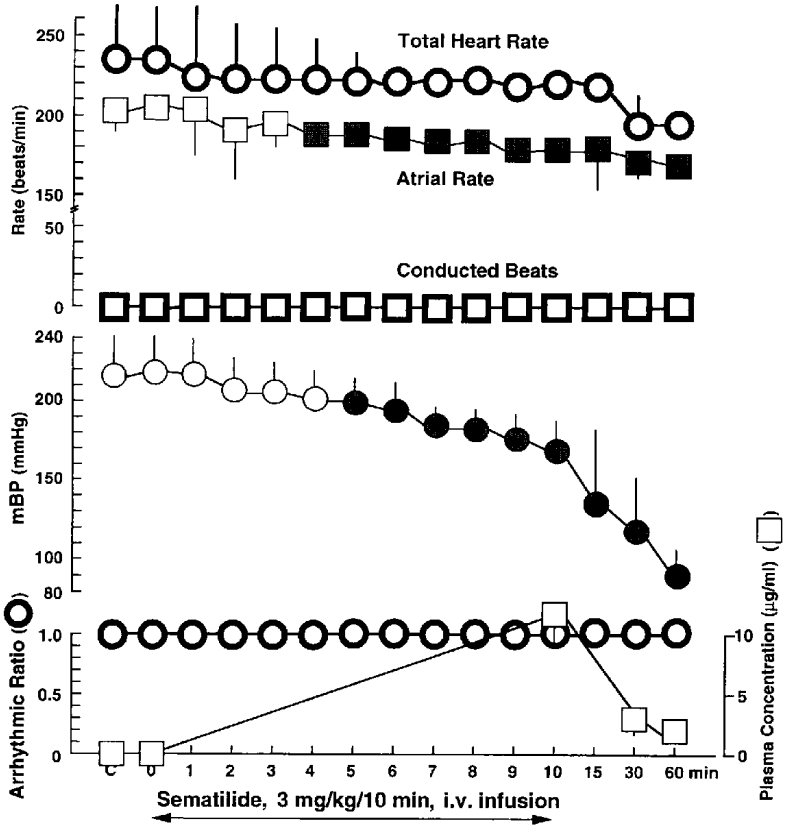


**Fig. 2.** Effects of intravenous infusion of sematilide, 3 mg/kg/10 min, on the digitalis arrhythmia models (n=4). Shaded points represent $P<0.05$, compared with the value at 0 time. S.D.s are shown at each time point. mBP: mean blood pressure.

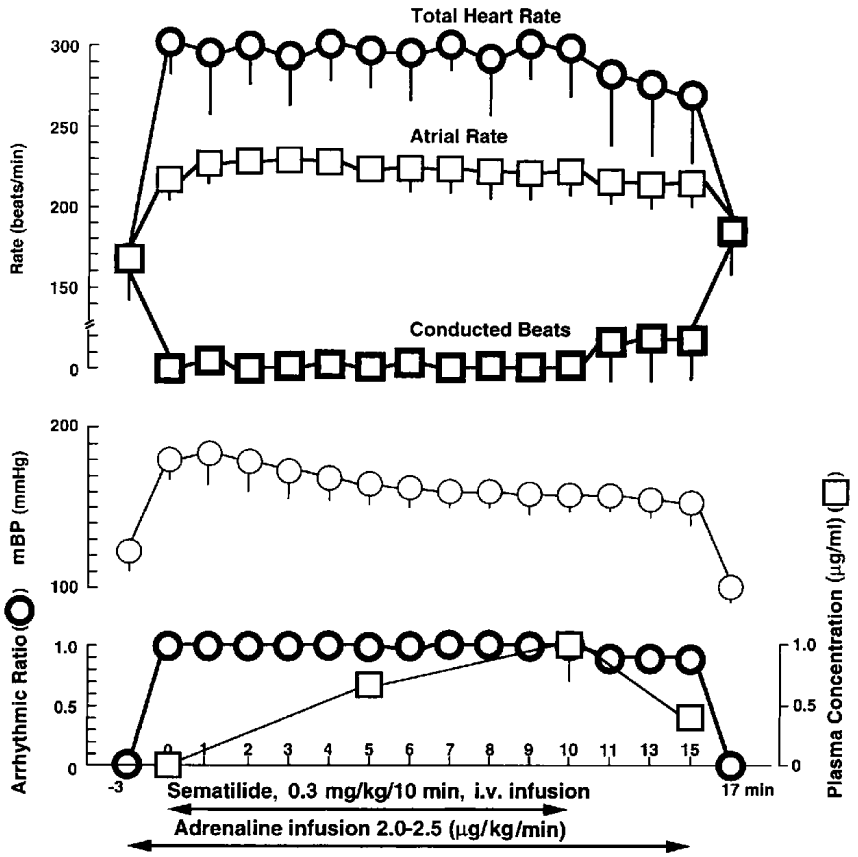


**Fig. 3.** Effects of intravenous infusion of sematilide, 0.3 mg/kg/10 min, on the adrenaline arrhythmia models (n=6). S.D.s are shown at each time point. mBP: mean blood pressure.

showed continuously occurring multifocal ventricular ectopic beats. The arrhythmic ratio of the unanesthetized dogs at the control 0 time was 0.99±0.03 (mean±S.D., n=6) (Fig. 1). Sematilide, 3 mg/kg, transiently increased the number of conducted beats and decreased the arrhythmic ratio, but the changes were not significant. The total heart rate, atrial rate and mean blood pressure were not significantly changed. The mean sematilide plasma concentrations at 0, 5, 10, 30 min were 0.00, 5.84, 7.07 and 2.47 μg/ml (n=6), respectively.

*Digitalis-induced arrhythmia*

After a total i.v. injection of about 70–90 μg/kg ouabain, almost all the beats became of ventricular origin. With a dose of 3 mg/kg/10 min, 2 out of 6 dogs fibrillated within 10 min, but no antiarrhythmic effect was observed in the other 4 dogs (Fig. 2). The mean sematilide plasma concentrations at 0, 10, 30, 60 min were 0.00, 11.65, 2.75 and 2.15 μg/ml (n=4), respectively. With a smaller dose of 1 mg/kg/10 min, no effect was observed on the arrhythmic ratio.

*Adrenaline-induced arrhythmia*

Sematilide in a small dose of 0.3 mg/kg/10 min, which is one tenth of the dose used in other experiments, had no effect (Fig. 3), but a dose of 1 mg/kg/10 min, which is still one third of the dose used in other experiments, induced VF in all 3 dogs soon after the infusion. The total heart rate, atrial rate, conducted beats and mean blood pressure were not significantly changed after 0.3 mg/kg/10 min. The mean sematilide (0.3 mg/kg/10 min) plasma concentrations at 0, 5, 10, 15 min were 0.00, 0.66, 1.04 and 0.43 μg/ml (n=6), respectively.

*Coronary ligation and reperfusion arrhythmia*

*Group 1. Sematilide, 3 mg/kg/10 min, in halothane anesthetized beagles*: The heart rate and mean blood pressure of all beagles anesthetized with halothane were 118±16 beats/min and 110±15 mmHg (n=12). As shown in Fig. 4, sematilide prolongated the QTc interval significantly from 0.39 to 0.56 $sec^{1/2}$ (just before LAD ligation), as compared with that of the control group from 0.36 to 0.38 $sec^{1/2}$. Sematilide significantly decreased the heart rate by 23% (from 117 to 90 beats/min) just before ligation and by 20% (to 94 beats/min) before reperfu-

sion. During the 30 min of coronary ligation, all dogs showed VPC and VT, but no dogs showed torsades de pointes (TdP). There was no change in the heart rate of the control group (118 to 121 beats/min just before occlusion). There were also no significant differences in the number of total VPCs between the drug- and saline-treated groups (125 and 149 beats/30 min during the ligation period, respectively). Immediately after reperfusion, 3 out of 6 dogs given sematilide and only 1 out of 6 dogs given saline infusion fibrillated. These fatal VF occurred soon after reperfusion. In addition, during the 30 min of infusion of sematilide and before applying coronary occlusion, 2 out of 6 dogs showed VPCs. The sematilide plasma concentrations at 0, 10, 29 min (1 min before ligation) and 59 min (1 min before reperfusion) were 0.00, $10.95 \pm 0.47$, $2.15 \pm 0.08$ and $1.18 \pm 0.07$ $\mu$g/ml ($n=4$), respectively.

*Group 2. Sematilide, 3 mg/kg/10 min, in pentobarbital anesthetized beagles*: Changing the anesthesia to i.v. pentobarbital, we repeated the experiment of sematilide with the same dose as that used in the group 1. The heart rate and mean blood pressure under pentobarbital anesthesia were $168 \pm 29$ beats/min and $122 \pm 17$ mmHg, respectively ($n=15$). As shown in Table 1, the QTc interval of the drug treated group also increased significantly from 0.36 to 0.44 $sec^{1/2}$ before ligation and 0.45 $sec^{1/2}$ before reperfusion; in the saline treated group, there were no changes (from 0.36 to 0.36 and 0.39 $sec^{1/2}$). Sematilide decreased the heart rate by 12% just before coronary ligation and stayed at this level until reperfusion. Also in this pentobarbital anesthetized condition, sematilide had no antiarrhythmic effects. The numbers of total VPCs in the drug- and saline-treated groups were 140 ($n=8$) and 412 ($n=7$) beats, respectively, during the 30 min of coronary occlusion. Immediately after reperfusion, 4 out of 8 dogs fibrillated in the drug-treated group (1 case of VF following TdP, while 3 out of 7 in the saline-treated group fibrillated). The sematilide plasma concentrations were 0.00, $8.91 \pm 0.68$, $2.18 \pm 0.20$ and $1.07 \pm 0.05$ $\mu$g/ml ($n=8$) at the same time points obtained in the group 1 experiment.

*Group 3. Sematilide, 6 mg/kg/10 min, in pentobarbital anesthetized beagles*: Applying the same protocol as that of group 2, but even using the twice higher dose of sematilide, we did not observe any antiarrhythmic effects of the drug. As shown in Table 1, the QTc interval further increased from 0.32 to 0.41 $sec^{1/2}$ before ligation and 0.42 $sec^{1/2}$ before reperfusion, and the heart rate decreased 14% and 18% before ligation and reperfusion, respectively (with a non-linear dose-effect when compared with the effects of 3 mg/kg/10 min). There were no significant differences in the number of total VPCs in the drug- and saline-treated groups 132 ($n=6$) and 70 ($n=6$) beats during the 30 min of coronary occlusion, respec-

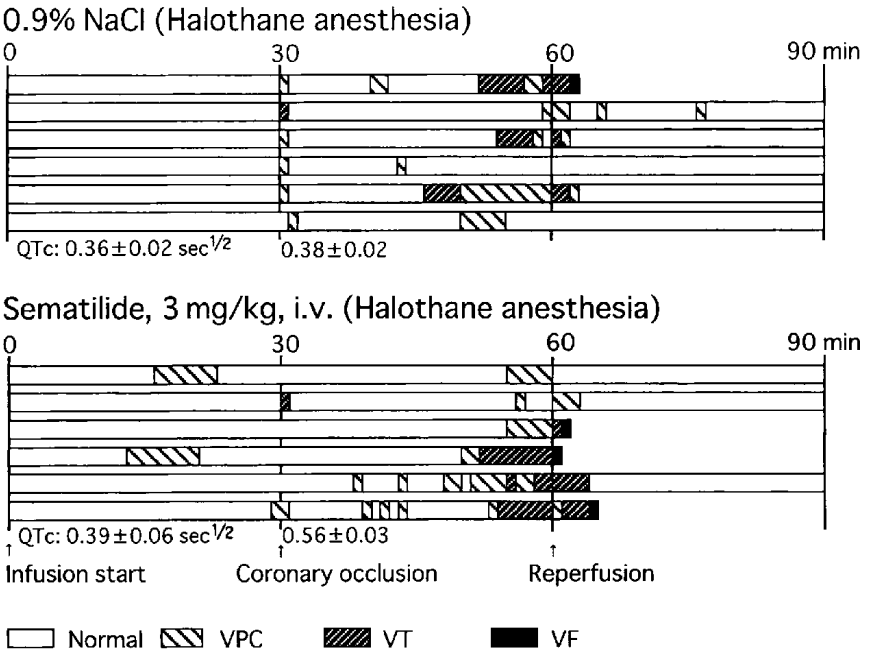


**Fig. 4.** Summary of the effect of sematilide on the coronary ligation-reperfusion experiments in halothane-anesthetized beagles. Each column indicates the responses of each dog. Only one dog died after reperfusion in the saline-treated group, and three dogs died in the sematilide-treated group. VPC: ventricular premature contraction, VT: ventricular tachycardia, VF: ventricular fibrillation.

**Table 1.** Effects of sematilide on the coronary occlusion-reperfusion arrhythmia

| | Group 1 | | Group 2 | | Group 3 | |
|---|---|---|---|---|---|---|
| | Control (n=6) | Drug (n=6) | Control (n=7) | Drug (n=8) | Control (n=6) | Drug (n=6) |
| Anesthetics | Halothane | | Pentobarbital | | Pentobarbital | |
| Dose | Saline | 3 mg/kg | Saline | 3 mg/kg | Saline | 6 mg/kg |
| Control | | | | | | |
| HR | 118±23 | 117±6 | 175±32 | 161±26 | 149±18 | 159±40 |
| QTc | 0.36±0.03 | 0.39±0.06 | 0.36±0.03 | 0.36±0.03 | 0.37±0.04 | 0.33±0.03 |
| 30 min after sematilide | | | | | | |
| ⊿%HR | 2±11 | −23±6** | 1±3 | −12±3** | 1±4 | −14±3** |
| ⊿%QTc | −0.1±6 | 48±26** | 0.1±3 | 26±10** | −1±6 | 27±18** |
| VPC (during 30 min occlusion) | 149±124 | 125±123 | 412±391 | 140±195 | 70±71 | 132±279 |
| VF (after reperfusion) | 1/6 | 3/6 | 3/7 | 4/8 | 3/6 | 3/6 |

HR: heart rate, beats/min. QTc: corrected QT interval, $sec^{1/2}$. VPC: ventricular premature contraction, beats/30 min. VF: ventricular fibrillation. Results are means±S.D. of six to eight dogs. **P<0.01, compared with before administration.

tively. In both groups, 3 out of 6 dogs fibrillated. The sematilide plasma concentrations were 0.00, 16.83±0.89, 3.44±0.15 and 1.90±0.09 μg/ml (n=6) at the same time points obtained in the group 2 experiment.

In these three coronary ligation and reperfusion experiments, the QTc prolonging effect of sematilide persisted until the end of the experiments, showing a non-linear relationship, instead of a hysteresis, between the QTc prolongating effects and plasma concentrations. In addition, reverse use dependent QTc prolongation was observed, i.e., more prominent prolongation in the low heart rate halothane-anesthetized group 1 compared to the prolongation in the high heart rate pentobarbital-anesthetized group 2.

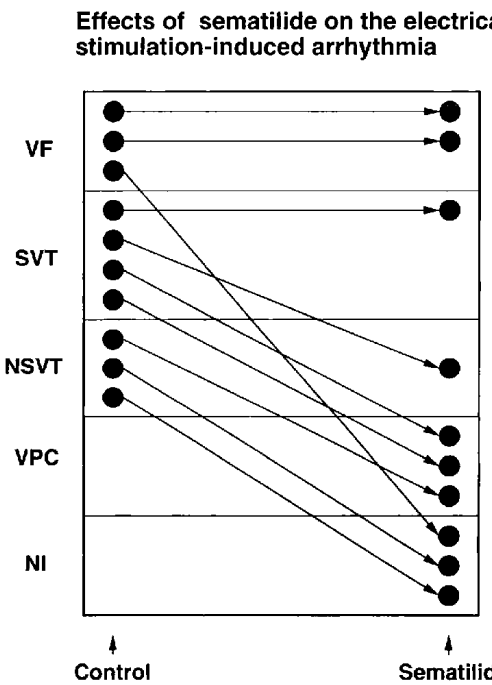


**Fig. 5.** Effects of intravenous infusion of sematilide, 3 mg/kg/10 min, on the programmed electrical stimulation-induced arrhythmia models (n=10). VF: ventricular fibrillation, SVT: sustained ventricular tachycardia, NSVT: nonsustained ventricular tachycardia, VPC: ventricular premature contraction, NI: non-inducible.

*PES-induced arrhythmias in dogs with old myocardial infarction*

By applying a series of PES, 10 dogs showed different types of ventricular arrhythmias: VF, SVT and NSVT, occurred in 3, 4 and 3 dogs, respectively. As shown in Fig. 5, sematilide significantly reduced the severity of these arrhythmias. After sematilide, 2 out of 3 dogs that responded with VF before drug administration, again fibrillated, but in one dog, no arrhythmia became inducible (NI); among the 4 dogs that showed SVT in the control period, 1 dog still showed SVT, but one developed NSVT and the other 2 dogs showed only VPCs; among the 3 dogs that showed NSVT in the control period, one developed VPC and the other 2 dogs became NI. In this experiment, the QTc interval increased by 23% (0.35 to 0.43 $sec^{1/2}$).

## DISCUSSION

*Effects of drug on two-stage coronary ligation-, digitalis- and adrenaline-induced arrhythmias*

In 48-hr two-stage coronary ligation VT, which is less severe than 24-hr VT and has been shown to be easily suppressed by class I $Na^+$ channel blockers (15), sematilide in a dose of 3 mg/kg/10 min, i.v., which is large enough

to increase the QT interval as based on a previous report (12) or from our own coronary ligation-reperfusion experiments, did not suppress this arrhythmia, nor did it aggravate it. There were no cardiovascular side effects such as hypotension and sinus arrest. Sematilide was also ineffective on digitalis-induced arrhythmia at 1 mg/kg/10 min, but aggravated it at 3 mg/kg/10 min (2 out of 6 dogs fibrillated). Consistent with our results, Argentieri reported that sematilide had no protective activity against ouabain-induced arrhythmias in dogs or guinea pigs (18). Similarly, sematilide was not effective on adrenaline-induced arrhythmia with a small dose of 0.3 mg/kg/10 min, but it aggravated it at 1 mg/kg/10 min (all 3 dogs fibrillated soon after the infusion). These results are similar to our previous reports, showing that class III agents, such as *d*-sotalol, E-4031 and MS-551, were ineffective on these arrhythmic models, but different in that sematilide actually aggravated digitalis and adrenaline-induced arrhythmias. E-4031 aggravated adrenaline-induced arrhythmia, but there is no report examining MS-551 on adrenaline arrhythmia (19, 20). These results may demonstrate potential proarrhythmic effects of sematilide.

*Effects of drug on coronary ligation- and reperfusion-induced arrhythmia*

The results of the present investigation on this arrhythmia model indicate that both in halothane- and pentobarbital-anesthetized beagles, sematilide was not effective in suppressing the number of VPC during the 30 min of LAD occlusion, nor was it effective in suppressing the occurrence of coronary reperfusion VF. This is partly due to the very low incidence of reperfusion VF in the control group (1 of 6, 3 of 7 and 3 of 6 dogs in groups 1, 2 and 3, respectively), which is lower than our average occurrence of VF of more than 60% (12). As has been already known, the variable occurrence of reperfusion VF in control dogs made only the prominent antiarrhythmic effects of the drug visible; therefore, it may be difficult to conclude that sematilide is really different from other new class III drugs such as E-4031, MS-551 and *d*-sotalol that suppressed the reperfusion VF (19–21). In these experiments, E-4031 as well as MS-551 was effective in suppressing the occurrence of coronary reperfusion VF in slow heart rate halothane-anesthetized beagles, but *d*-sotalol did not prolong QT as the other two drugs and was not effective. However, when pentobarbital sodium was used as an anesthetic, *d*-sotalol became effective in suppressing the VF. In the present study, the average heart rate in the halothane-anesthetized dogs was 118 beats/min and is significantly lower than that of the pentobarbital anesthetized dogs, 168 beats/min. Sematilide prolongated the QTc interval by 44% in the low heart rate halothane-anesthetized dogs, but only 22% in the high heart rate pentobarbital-anesthetized dogs. Such a reverse use-dependent effect was also observed in the MS-551 experiment; However, *d*-sotalol did not show such a reverse use-dependent QT prolongation (19). The lack of antiarrhythmic effect of sematilide, and also *d*-sotalol's effectiveness observed only when the dogs were anesthetized with pentobarbital, indicate that the mechanism of sup-

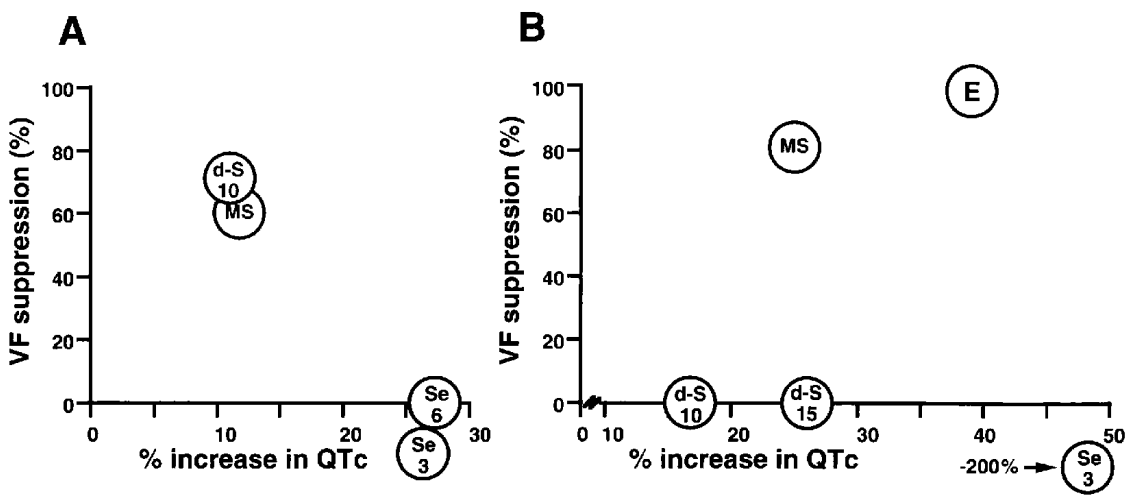


**Fig. 6.** The summary of our results on class III drugs on the reperfusion ventricular fibrillation (VF) versus their QT prolonging effects. A: The anesthetic was pentobarbital. B: The anesthetic was halothane. *d*-S: *d*-Sotalol, MS: MS-551, E: E-4031, Se: Sematilide. Each number under the drug indicates the dose of the drug in mg/kg. The doses of MS-551 and E-4031 were 3.6 mg/kg/hr and 30 $\mu$g/kg+3 $\mu$g/kg/min, respectively. Percent suppression of VF was plotted against QTc prolongation. Minus means an increase in the VF occurrence; for example, in panel B, VF suppression percentage of sematilide (3 mg/kg/10 min) is minus 200%, which means increase the VF occurrence to 200%.

pression is not directly related to the class III QT prolonging actions. Figure 6 shows the summary of our results on class III drugs on the reperfusion VF versus their QT prolonging effects. Percent suppression of VF, calculated as (1 − VF occurrence in the treated group / VF occurrence in the saline group) × 100, was plotted against QTc prolongation. Minus means increase in the VF occurrence. *d*-Sotalol and sematilide did not suppress VF occurrence even though they prolonged the QTc. The lack of a linear relationship also indicates that this arrhythmia model is not suitable for predicting the efficacy of class III drugs. The mechanism of generation of the coronary ligation and reperfusion arrhythmia is thought to be re-entry at and around the acutely infarcted myocardium (22, 23), but it is also reported that the reentrant and non-reentrant mechanisms contribute to arrhythmogenesis during early myocardial ischemia (24, 25). These reports of the existence of reentrant and non-reentrant mechanisms in this model also indicate that this model may not be an appropriate one to evaluate the efficacy of class III drugs. In addition, it is worthy to mention that even when sematilide administration was stopped at 10 min, it prolonged the QTc interval until the end of the experiments. There might exist prominent hysteresis between the prolongating effect of sematilide on QTc and the plasma concentration of sematilide.

*Proarrhythmic effect in coronary ligation- and reperfusion-induced arrhythmia*

The proarrhythmic effect of sematilide was observed during the 30-min control period of the coronary ligation-reperfusion experiments; VPC occurred before applying the coronary ligation in halothane-anesthetized beagles (2 out of 6 beagles), but did not occur in pentobarbital-anesthetized beagles. This may be due to the use of halothane, because arrhythmogenic effects were also observed in our halothane-anesthetized dogs given E-4031 and MS-551, where severe TdP type VT occurred (19, 20). Halothane is known to decrease the sinoatrial rate and the use of halothane in our experiment kept the heart rate relatively low and intensified the QT prolonging effect of class III drugs. Halothane is also known to sensitize the cardiac cell to the arrhythmogenic effect of catecholamines, probably because halothane interferes with the cell-to-cell coupling and thus decreases the conduction velocity (26). So with the concomitant use of halothane, new class III drugs showed arrhythmogenic effects in dogs and such side effects may occur in clinical situations when the QT interval is dramatically increased during the use of class III drugs. It seems that sematilide is a safer drug than MS-551 and E-4031 in that sematilide did not induce Tdp, but due to the small number of dogs used, the conclusion may not be a definite one.

*Effect of drug on PES-induced arrhythmias*

Although sematilide had no antiarrhythmic effects on the four spontaneously occurring arrhythmia models, an antiarrhythmic effect was observed using an arrhythmia model that was induced solely by the re-entry mechanism in dogs with old myocardial infarction by PES. In this experiment, sematilide reduced the severity of arrhythmias that were induced in the control period; for example, VF or SVT to a less severe arrhythmia like NSVT, VPC or even made arrhythmia not inducible. These results are consistent with other reports using similar models of a reentrant mechanism. Sullivan et al. reported antiarrhythmic effects of sematilide in a canine model of 3−8 days after anterior wall myocardial infarction that was created by a LAD occlusion followed by reperfusion. Under a nonsedated condition, dogs were given a PES to induce re-entry arrhythmias (11). Argentieri also demonstrated that sematilide was effective in preventing PES-induced arrhythmias in conscious dogs (18). These studies showed that the QTc prolongation by sematilide must have contributed to the suppression of the re-entry type arrhythmias.

*Concluding remarks*

Although QT prolongation of sematilide was a common property of class III drugs, effects of sematilide on arrhythmias were slightly different from those of other class III antiarrhythmic drugs in their effectiveness on canine ventricular arrhythmia models. In the present paper, sematilide was not effective on the coronary ligation-reperfusion arrhythmia model. The reason for the ineffectiveness was probably due to the variability of the occurrence of this arrhythmia in the control animals as mentioned earlier, but also may be due to the limitation of this arrhythmia model for predicting the effects of class III drugs or due to the difference between sematilide and other class III drugs. Further studies may be necessary to solve this problem. Another important difference was its proarrhythmic effect. As other class III drugs we have examined, sematilide induced ventricular arrhythmias during halothane anesthesia before applying coronary occlusion. In addition, sematilide aggravated digitalis induced VT to VF in 2 dogs and a lower one third dose as compared to that used for other arrhythmia models aggravated adrenaline-induced arrhythmia. Since digitalis and adrenaline arrhythmias are thought to be produced by the automaticity mechanism, class III $I_K$ blockers should not influence the automaticity arrhythmia generation. We have reported that some of the antiarrhythmic drugs having a QT prolonging effect, such as disopyramide, procainamide, pilsicainide, E-4031, aggravated adrenaline arrhythmia (16, 20). This proarrhythmic potency of sematilide may occur in its clinical use, and the induced

arrhythmias may be of a severe or fatal nature. Careful protocol for clinical development must be planned. Also the present experiments show that new, more reliable methods for detecting proarrhythmic drug effects should be developed in the near future.

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