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Effects of OPC-8212, a new positive inotropic agent, on canine ventricular arrhythmias

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1 OPC-8212, a positive inotropic agent that increases intracellular cyclic AMP and stimulates the Ca current, was compared with amrinone and AR-L 115 in their effects on canine ventricular arrhythmias.

2 OPC-8212, up to 3 mg kg^{-1}, did not worsen 24 h coronary ligation arrhythmias, while it aggravated ventricular arrhythmias seen 48 h after coronary ligation. However, OPC-8212 did not at any time induce ventricular fibrillation in dogs subjected to coronary ligation.

3 OPC-8212, up to 3 mg kg^{-1}, had no arrhythmogenic or antiarrhythmic effect on digitalis-induced arrhythmias, while amrinone and AR-L 115 showed antiarrhythmic effects.

4 With adrenaline-induced arrhythmias, the three drugs aggravated ventricular tachycardia to produce ventricular fibrillation.

5 The absence of a worsening effect of the new positive inotropic agents on digitalis arrhythmia may be helpful in a clinical setting of combined therapy of these new drugs with digitalis.

Introduction

OPC-8212 is a newly synthesized positive inotropic agent the mechanism of action of which has been reported to be different from those of digitalis glycosides or β-adrenoceptor agonists (Yamashita *et al.*, 1984; Taira *et al.*, 1984; Hashimoto *et al.*, 1984c; Takeya *et al.*, 1984). It has been reported that OPC-8212 increases intracellular adenosine 3′:5′-cyclic monophosphate (cyclic AMP) (Taira *et al.*, 1984) and that it increases the slow inward current (called hereafter Ca current) of rabbit sino-atrial node (Satoh & Hashimoto, 1984), guinea-pig ventricular muscle (Takeya *et al.*, 1984) and canine ventricular muscle (Taira *et al.*, 1984). These effects are probably responsible for the drug's positive inotropic action, but may also affect cardiac excitation and induce arrhythmias, because the Ca current is important for the generation of both normal and abnormal cardiac excitation (Noble, 1975; Cranefield & Wit, 1979). Since the occurrence of arrhythmias is deleterious in the treatment of cardiac failure, the present experiment was set up to examine the effects of OPC-8212 on various types of canine ventricular arrhythmias. The arrhythmia models chosen for the present study were produced by coronary ligation, digitalis intoxication and adrenaline infusion and the effects of OPC-8212 were compared with other new positive inotropic agents (Farah *et al.*, 1984), amrinone (Alousi *et al.*, 1979; Endoh *et al.*, 1982) and AR-L 115 (Diederen & Kadatz, 1981; Morita *et al.*, 1984; Endoh *et al.*, 1985).

Methods

Production of two-stage coronary ligation-induced arrhythmias

Seven beagle dogs, weighing 7–9 kg, were anaesthetized initially with thiopentone sodium. As reported earlier (Hashimoto *et al.*, 1982), the chest was opened and a two-stage coronary ligation was performed under halothane anaesthesia.

Experiments were carried out in conscious dogs 24 and 48 h after coronary ligation. The lead II ECG, atrial electrogram from the left atrial appendage, and blood pressure were recorded continuously by use of telemetry systems (Nihon Kohden and Nishimu). OPC-8212 was injected through a cannula in the jugular vein.

Production of digitalis-induced arrhythmias

Twenty one mongrel dogs of either sex, weighing 8–15 kg, were anaesthetized with pentobarbitone sodium, 30 mg kg^{-1}. As reported earlier (Hashimoto *et al.*, 1985), ouabain 40 μg kg^{-1} was injected intravenously and with an additional 10 μg kg^{-1} every 20 min

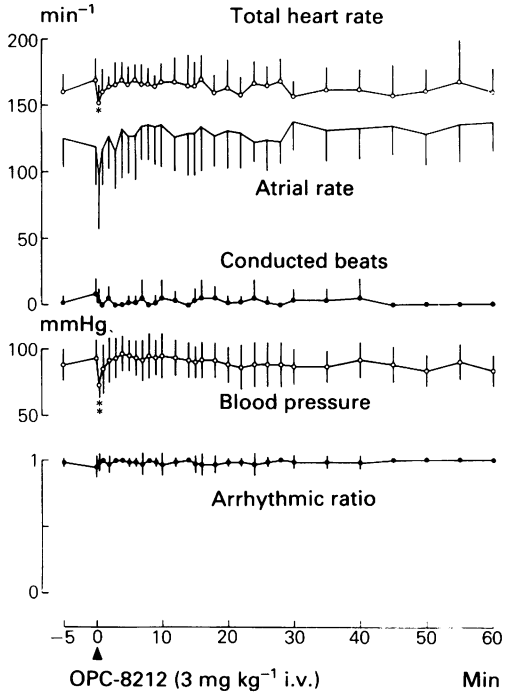

Figure 1 Summary of the effects of OPC-8212, 3 mg kg^{-1} i.v., on 24 h two-stage coronary ligation-induced arrhythmias. $n = 7$. *P < 0.05; **P < 0.01.

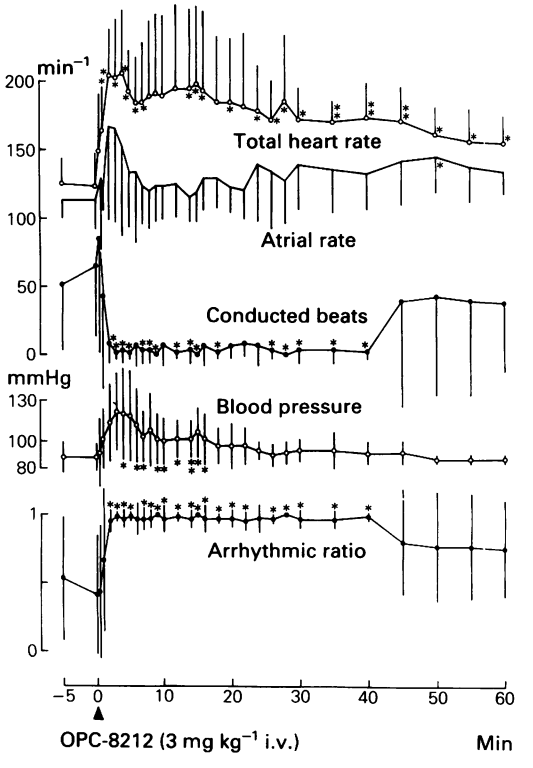

Figure 2 Summary of the effects of OPC-8212, 3 mg kg^{-1} i.v., on 48 h two-stage coronary ligation-induced arrhythmias. Details as in Figure 1. $n = 6$.

until stable ventricular arrhythmias were produced. OPC-8212 and other drugs were injected intravenously through a cannula in the femoral vein.

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

Production of adrenaline-induced arrhythmias

Thirty one mongrel dogs of either sex, weighing 7–15 kg, were anaesthetized initially with thiopentone sodium. As reported earlier (Shibuya *et al.*, 1983), after intubation, 1.0% halothane, vaporized with 95% O_2 and 5% CO_2, was administered with a volume-limited ventilator. Adrenaline was infused through the left femoral vein at a rate of 2.5 μg kg^{-1} min^{-1}. After 3 min of adrenaline infusion, OPC-8212 and other drugs were injected into the right femoral vein.

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

Evaluation of the antiarrhythmic effects

The severity of arrhythmia was expressed by the arrhythmic ratio i.e. number of ventricular ectopic beats divided by the total heart rate. For the three arrhythmias, the arrhythmic ratios before drug injection were almost 1 and there were no spontaneous improvements in these ratios. If the arrhythmic ratio after drug administration was decreased significantly from the zero time value, as determined by Student's *t* test for paired data, the drug was judged as having an antiarrhythmic effect.

Drugs

Drugs used were OPC-8212 (3,4-dihydro-6-[4-(3,4-dimethoxybenzoyl) -1-piperazinyl]-2(1H) -quinolinone, Otsuka Pharmaceutical Co.) dissolved in 60% sulpholane, amrinone hydrochloride dissolved in saline solution and AR-L 115 (2-[(2-methoxy-4-methylsulphinyl)-phenyl]-1-H-imidazo(4,5-β)-pyridine, sulmazole, C.H. Boehringer Sohn, Ingelheime) dissolved in saline solution.

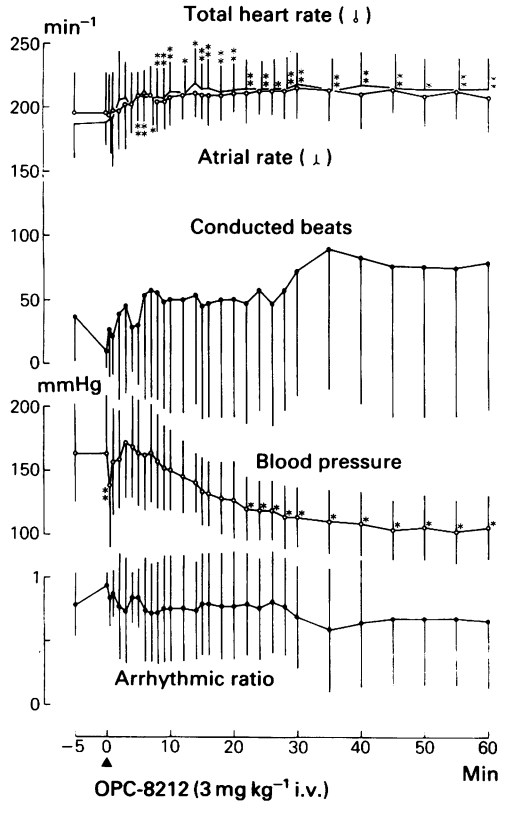

Figure 3 Summary of the effects of OPC-8212, 3 mg kg^{-1} i.v., on digitalis-induced arrhythmias; $n = 6$. Details as in Figure 1.

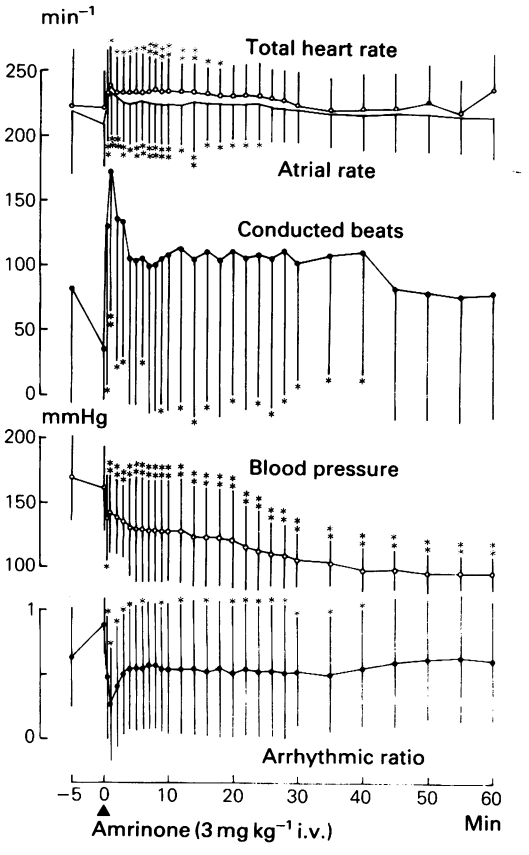

Figure 4 Summary of the effects of amrinone (3 mg kg^{-1} i.v.) on digitalis-induced arrhythmias; $n = 9$. Other details as in Figure 1.

Results

Effects of OPC-8212 on two-stage coronary ligation-induced arrhythmias (Figures 1, 2)

Twenty four hours after coronary ligation, conscious beagle dogs showed ventricular tachycardia, as indicated by the −5 and 0 time values of arrhythmic ratio in Figure 1. The maximal dose of OPC-8212 used in previous canine heart failure model studies (Hashimoto *et al.*, 1984c), 3 mg kg^{-1}, was given, but there was no antiarrhythmic effect, i.e. no statistically significant decrease in the arrhythmic ratio. Heart rate and blood pressure decreased transiently soon after injection, but they returned to their control values quickly. Conscious dogs showed excitement and occasionally vomited within 5 min of OPC-8212 injection. This may be due to the solvent, sulpholane, because the same volume of diluted sulpholane showed similar effects.

The same dogs were used the next day for the study of OPC-8212 effects on the 48 h arrhythmia. As shown in the lower values of the arrhythmic ratio of −5 and 0 time in Figure 2, the 48 h arrhythmia was less severe. At the same dose (3 mg kg^{-1}) OPC-8212 showed an arrhythmogenic effect as shown by the increase in the arrhythmic ratio (Figure 2), and this effect lasted about 40 min. Although the total heart rate and the number of ectopic beats also increased, ventricular fibrillation was never observed. The CNS side effects of excitation, which were sometimes accompanied by convulsions were possibly due to sulpholane and appeared more frequently than on the previous day. Blood pressure was elevated as the dogs showed excitation. The arrhythmogenic effects of OPC-8212 were not induced by the same volume of sulpholane used to inject OPC-8212 (3 mg kg^{-1}).

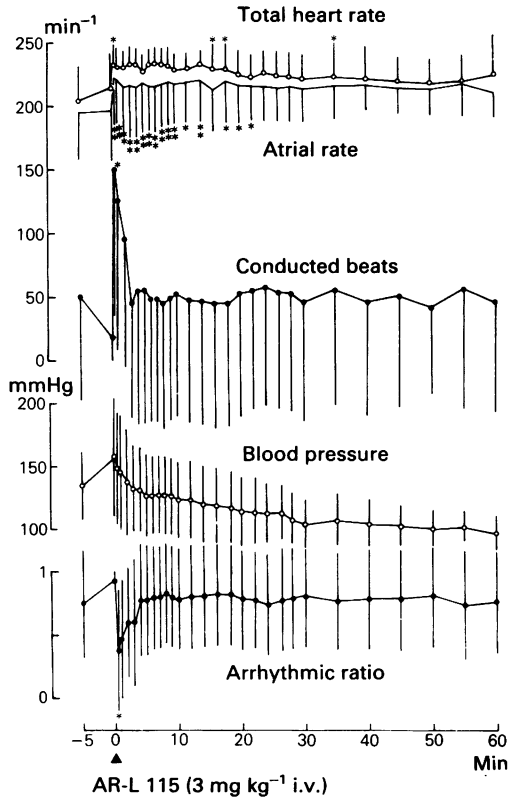

Figure 5 Summary of the effects of AR-L 115 (3 mg kg^{-1} i.v.) on digitalis-induced arrhythmias; $n = 6$. Other details as in Figure 1.

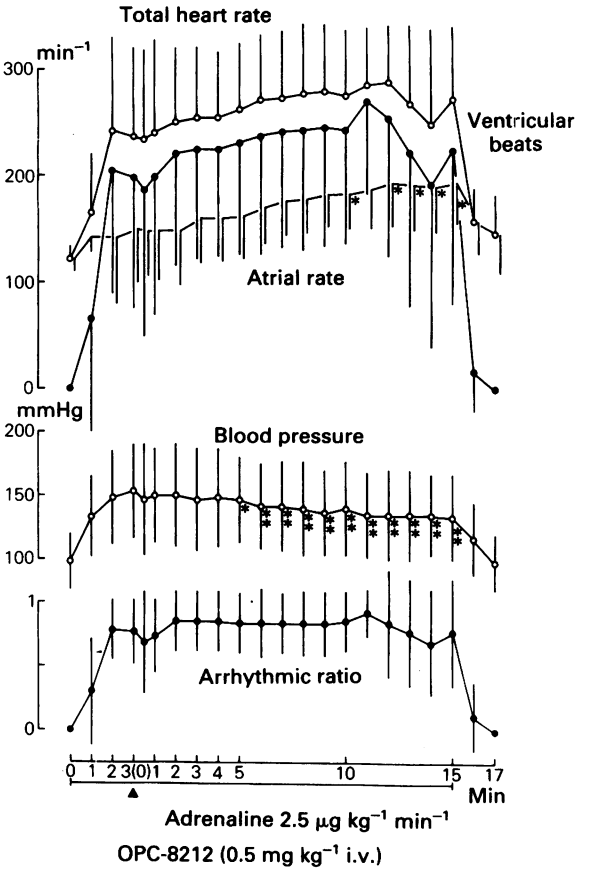

Figure 6 Summary of the effects of a low dose of OPC-8212 (0.5 mg kg^{-1} i.v.) on adrenaline-induced arrhythmias; $n = 6$. Other details as in Figure 1.

Effects of OPC-8212 and other drugs on digitalis-induced arrhythmias

After injection of a total dose of 70–90 μg kg^{-1} ouabain, almost all the beats were of ventricular origin, as shown in the −5 and 0 time values (Figures 3–5). The maximum dose of OPC-8212 (3 mg kg^{-1}) showed no antiarrhythmic effect, but did not induce worsening of arrhythmia, or produce ventricular fibrillation. This dose of OPC-8212 transiently decreased blood pressure, and increased total heart rate and atrial rate. The transient hypotensive effect was also observed with the solvent, sulpholane, given in the same volume as used for OPC-8212. The hypotensive effect observed 20 min after injection might be due to the decrease in the concentration of ouabain, because no supplement of ouabain was given after OPC-8212 injection.

Amrinone (3 mg kg^{-1} i.v.) on the other hand decreased the arrhythmic ratio and this antiarrhythmic effect lasted about 40 min as shown in Figure 4. Amrinone showed a long lasting hypotensive effect. The same dose of AR-L 115 also showed a transient antiarrhythmic effect but of less than 1 min duration (Figure 5), but it had no hypotensive effect. Both drugs increased the total heart rate and atrial rate as with OPC-8212.

Effects of OPC-8212 and other drugs on adrenaline-induced arrhythmias

Adrenaline infusion at a rate of 2.5 μg kg^{-1} min^{-1} induced ventricular tachycardia as shown in the sudden increase in the number of ventricular ectopic beats (Figures 6–8). OPC-8212 (1 and 3 mg kg^{-1}) worsened this adrenaline arrhythmia and induced ventricular fibrillation in 5 out of 6 dogs and 1 out of 2

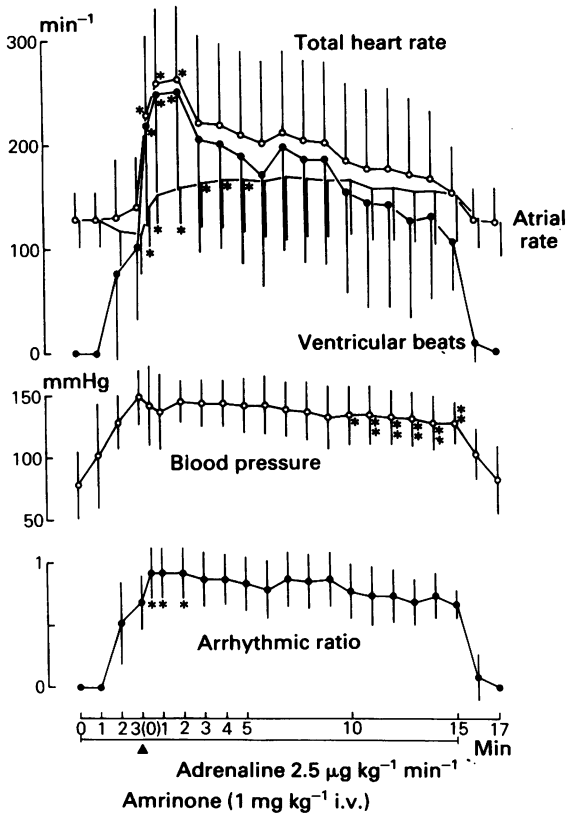

Figure 7 Summary of the effects of amrinone (1 mg kg^{-1} i.v.) on adrenaline-induced arrhythmias; $n = 6$. Other details as in Figure 1.

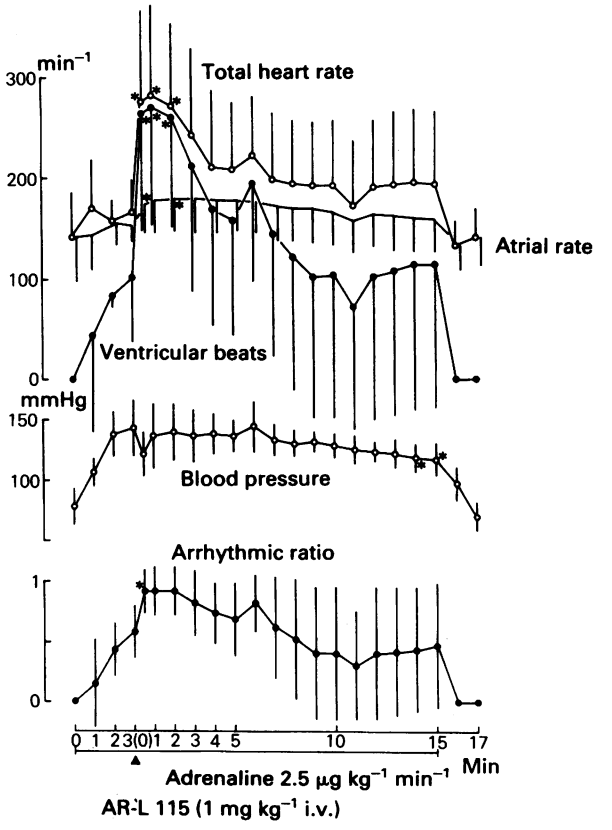

Figure 8 Summary of the effects of AR-L 115 (1 mg kg^{-1} i.v.) on adrenaline-induced arrhythmias: $n = 5$. Other details as in Figure 1.

dogs, respectively. Figure 6 shows the effect of 0.5 mg kg^{-1} OPC-8212 on adrenaline arrhythmia. This low dose of OPC-8212 did not aggravate adrenaline arrhythmia.

Amrinone and AR-L 115 (3 mg kg^{-1}) also aggravated adrenaline arrhythmia and induced ventricular fibrillation in 2 out of 3 dogs and in all of 3 dogs, respectively. The lower (1 mg kg^{-1}) dose transiently increased the arrhythmic ratio both in the cases where amrinone (Figure 7) and AR-L 115 were used (Figure 8). These lower doses of the three drugs did not induce hypotension soon after injection.

Discussion

The present experiments have shown that three new positive inotropic agents, OPC-8212, amrinone and AR-L 115, all aggravate adrenaline arrhythmias but not digitalis arrhythmias. Our previous experiments using three arrhythmia models showed that coronary ligation-induced and digitalis-induced arrhythmias are Na channel-dependent and are suppressed by class 1 antiarrhythmic drugs, while adrenaline arrhythmias are Ca channel-dependent and are suppressed by class 2 β-blockers and class 4 Ca channel blockers (Hashimoto *et al.*, 1982; 1984a,b; 1985; 1986; Shibuya *et al.*, 1983; Komori *et al.*, 1985). Like caffeine (Hashimoto *et al.*, 1986), which is also a phosphodiesterase inhibitor and increases cardiac Ca current, the three drugs we tested have been shown to worsen adrenaline arrhythmias. This worsening of adrenaline arrhythmias may be explained simply by an additive increasing effect of these drugs on the Ca current, which had already been increased by adrenaline and caused the arrhythmia. However, there are controversial reports on the effect of AR-L 115 on Ca current. Diez *et al.* (1984) suggested an increase in Ca influx in rat atrial muscle by AR-L 115, but Achenbach *et al.* (1984) observed no such effect in sheep Purkinje fibres, although they speculated that an increase in in-

tracellular Ca had occurred.

The three drugs had no deleterious effect on the digitalis arrhythmia, and it is consistent with our previous proposal that digitalis arrhythmia is not dependent on the Ca channel (Hashimoto *et al.*, 1985). Though Piwonka *et al.* (1983b) reported no arrhythmogenic and antiarrhythmic effect of amrinone on digitalis arrhythmias, we found that amrinone and AR-L 115 protect against digitalis arrhythmias. These different results may be due in part to the method of production of digitalis arrhythmias used in the two experiments, and also to qualitative differences between the three drugs. The mechanism of amrinone's antiarrhythmic effect in our study cannot be explained by the *in vitro* data of Piwonka *et al.* (1983a) who showed almost no effect of amrinone on the upstroke velocity of normal cardiac action potentials, which is an approximate measure of Na current. Regardless of the different antiarrhythmic effects among the three drugs, OPC-8212, amrinone and AR-L 115, the practical importance of these results is that these new positive inotropic agents can be used concurrently with digitalis without increasing the risk of arrhythmia.

The effect of OPC-8212 on coronary ligation-induced arrhythmias was similar to that reported for amrinone (Piwonka *et al.*, 1983b). Both drugs had no deleterious effects on 24 h coronary ligation arrhythmia, but had arrhythmogenic effects later on, in our case on 48 h coronary ligation arrhythmia. In neither 24 nor 48 h arrhythmias, did OPC-8212 cause ventricular fibrillation. These results indicate that OPC-8212 and the other two positive inotropic agents may be used for the treatment of cardiac failure caused by myocardial damage. However, it should always be borne in mind that these drugs are potentially arrhythmogenic, because of their Ca current or intracellular Ca concentration increasing effects. The present observation on the effects of the positive inotropic agents on digitalis arrhythmia cast doubts on the role of Ca-related phenomenon, such as oscillatory after potential, in the generation of digitalis arrhythmia (Hashimoto *et al.*, 1984b), but further *in vivo* studies may be needed to clarify this problem.

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