Experimental Studies

Antiarrhythmic Effects of the Class 1c Antiarrhythmic Drug, Flecainide, on Canine Ventricular Arrhythmia Models

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SUMMARY

The properties of a class 1c antiarrhythmic drug, flecainide, were examined using 3 canine ventricular arrhythmia models: (1) digitalis-, (2) adrenaline- and (3) two-stage coronary ligation-induced arrhythmias. The minimum effective plasma concentration of flecainide was determined for each model. Flecainide suppressed the arrhythmias. The minimum effective plasma concentrations for arrhythmias induced by digitalis, adrenaline and 24 hr coronary ligation, were 0.9 ± 0.2 , 1.4 ± 0.6 , 1.5 ± 0.4 mcg/ml, respectively (mean $\pm$ SD, n=6). These minimum effective plasma concentrations of flecainide were almost equal to the reported *in vitro* concentration that suppress the Na channels of isolated canine ventricular tissues. Thus, flecainide may suppress these arrhythmias by inhibiting the Na channels of cardiac cells.

Additional Indexing Words:

Ventricular arrhythmia Coronary ligation Digitalis Adrenaline Flecainide Class 1c antiarrhythmic drug

FLECAINIDE, N-(2-piperidinylmethyl)-2, 5-bis (2, 2, 2-trifluoroethoxy)-benzamide monoacetate, is a new class 1c antiarrhythmic drug, according to the Vaughan Williams classification system.¹⁾ It was reported to have an antiarrhythmic effect in both animal models²⁾ and human arrhythmias. Oral doses have been effective for the treatment of both paroxysmal supraventricular tachycardia (including Wolff-Parkinson-White syndrome) and ventricular arrhythmias.³⁾⁻⁵⁾ It is also reported to have few clinical side effects.^{6),7)}

We have previously examined the effects of several antiarrhythmic drugs on 3 canine ventricular arrhythmias (digitalis-, adrenaline- and two-stage coronary ligation-induced arrhythmias).⁸⁾⁻¹³⁾ These studies have shown that the efficacy of the drugs on these arrhythmia models are correlated with the drug's electrophysiological action on cardiac cells. For example, all class

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I Na⁺ channel blockers except lidocaine were effective on both digitalis-induced and coronary ligation-induced arrhythmias; they showed variable effects on adrenaline arrhythmia.¹³⁾ In the present study, we examined the typical class 1c drug flecainide on our arrhythmia models to determine whether its slow kinetics reflect a different pharmacological mode of action as compared to other class I antiarrhythmic drugs.

METHODS

Production of digitalis-induced arrhythmia:

Six mongrel dogs weighing 7–13 kg were anesthetized with pentobarbital sodium (30 mg/kg). As reported earlier,¹¹⁾ 40 mcg/kg ouabain was injected intravenously into the femoral vein and then followed by an additional dose of 10 mcg/kg every 20 min until stable ventricular arrhythmia was produced. Flecainide, 3 mg/kg, was injected rapidly as an intravenous bolus through a cannula in the femoral vein. The lead II electrocardiogram (ECG), atrial electrogram from catheter tip electrodes in the right atrium, and blood pressure were recorded continuously. Venous blood samples were taken from the jugular vein 5 min before and 1, 3, 5, 10, 15, 30 and 60 min after flecainide injection.

Production of adrenaline-induced arrhythmia:

Six mongrel dogs weighing 7–13 kg were anesthetized initially with thiopental sodium. After intubation, 1.0% halothane, vaporized with 100% oxygen, was administered with a volume-limited ventilator.¹²⁾ Adrenaline was infused through the left femoral vein at rates of 2.5–3.5 mcg/kg/min for 18 min. When stable and severe ventricular arrhythmia was produced by 3 min of adrenaline infusion, 3 mg/kg flecainide was injected into the right femoral vein.

The lead II ECG and blood pressure were continuously recorded. Venous blood samples were taken from the jugular vein 1 min before and 1, 3, 5, 10 and 15 min after flecainide injection.

Production of two-stage coronary ligation-induced arrhythmia:

Six beagle dogs, weighing 7–9 kg, were anesthetized initially with 30 mg/kg of thiopental sodium. As reported earlier,⁸⁾ the chest was opened and the two-stage coronary ligation was performed under halothane anesthesia.

Experiments were done without anesthesia 24 hr after coronary ligation. The lead II ECG, atrial electrogram from the left atrial appendage,

and blood pressure were recorded continuously using telemetry systems (Nihon Kohden and Nishimu). Three mg/kg flecainide was injected through a cannula inserted into the jugular vein. Venous blood samples were taken from the cannula 5 min before and 1, 3, 5, 10, 15, 30 and 60 min after injection of flecainide.

Determination of plasma concentration of flecainide:

Venous blood samples were centrifuged and the plasma was stored in a freezer at about -80°C before plasma flecainide was determined using a high-performance liquid chromatograph (HPLC), according to the method described by Chang et al.¹⁴⁾ There was an excellent linearity ($r=0.998$) in the calibration curve. The limit of detection was 50 ng/ml and the recovery was $99.4 \pm 1.2\%$ (mean $\pm$ SE, $n=3$) when 1 ml of spiked plasma (500 ng) was determined.

Determination of the minimum effective plasma concentration:

The severity of arrhythmia was expressed by the number of conducted beats or ventricular ectopic beats (the sum of the two is the total heart rate), and the arrhythmic ratio (the number of ventricular ectopic beats divided by the total heart rate). For 3 types of arrhythmias, there were no conducted beats and the arrhythmic ratios were 1 before drug injection, and there were no spontaneous improvements. The minimum effective plasma concentration of flecainide was determined by obtaining the last minute of a statistically significant decrease ($p < 0.05$) in the conducted beats compared with that at 0 time for the digitalis- and two-stage coronary ligation-induced arrhythmia models. For the adrenaline-induced arrhythmia model, the last minute of a statistically significant increase in the ventricular beats compared with that at 0 time was used. Student t-test for paired data was used to determine the significance of difference. The corresponding plasma concentration was then calculated from the experimentally derived plasma concentration-time equations, to estimate the minimum effective plasma concentration. Wilcoxon's signed rank test was used for the statistical analysis of the arrhythmic ratio.

RESULTS

Effects of flecainide on digitalis-induced arrhythmia:

After injection of a total dose of 70–80 mcg/kg ouabain, almost all the beats were of ventricular origin. Preliminary experiments using 1 and 3 mg/kg i.v. flecainide showed that a 1 mg/kg dose had almost no antiarrhyth-

Fig. 1. Summary of the effects of flecainide, 3 mg/kg i.v., on digitalis-induced arrhythmia. Flecainide suppressed the arrhythmia soon after injection and the antiarrhythmic effect lasted 16 min. * $p < 0.05$, ** $p < 0.01$.

mic effect. Therefore, a 3 mg/kg dose was used in this study. As shown in Fig. 1, the number of conducted beats increased significantly ($p < 0.05$) from 2 to 16 min after flecainide injection. This change was accompanied by a reduction in the total heart rate, the blood pressure and the arrhythmic ratio. The plasma concentration-time curves corresponded well with predictions from a two-compartment open model. The parameters of the curve were analyzed using the non-linear regression program "MULTI"¹⁵⁾ and an NEC PC-9801Vm computer (Tokyo, Japan). The parameters of the flecainide concentration-time equation, expressed as $\text{concentration} = Ae^{-\alpha t} +$

Fig. 2. Summary of the effects of flecainide, 3 mg/kg i.v., on halothane-adrenaline-induced arrhythmia. Flecainide decreased the total heart rate, number of ventricular beats, and arrhythmic ratio. The antiarrhythmic effect lasted 3 min.

$Be^{-\beta t}$: $A=2.9\pm2.0$ mcg/ml, $\alpha=0.44\pm0.18/\text{min}$, $B=1.1\pm0.2$ mcg/ml and $\beta=0.015\pm0.008/\text{min}$ (mean $\pm$ SD, $n=6$). The minimum effective plasma concentration was calculated as the value at 16 min, 0.9 ± 0.2 mcg/ml.

Effects of flecainide on adrenaline-induced arrhythmia:

As reported previously,¹²⁾ adrenaline infusion for 3 min at rates of 2.5–3.5 mcg/kg/min induced a tachycardia characterized by almost exclusively ventricular ectopic beats. Two doses of flecainide, 3 and 5 mg/kg, were examined in a preliminary study. Since the 5 mg/kg dose either induced ventricular fibrillation or exacerbated the arrhythmia, a 3 mg/kg dose was used.

Fig. 3. Effects of flecainide on 24 hr two-stage coronary ligation-induced arrhythmia. Flecainide, 3 mg/kg i.v., decreased the total heart rate and arrhythmic ratio, but the blood pressure increased for 60 min.

As shown in Fig. 2, flecainide rapidly decreased the total heart rate, the atrial rate, and the arrhythmic ratio. A significant decrease in the number of ventricular beats was observed at 1 and 2 min after injection. The blood pressure was reduced for 17 min. The plasma concentration-time curves fitted reasonably well with that predicted by the one compartment open model theory for the 5 data points during the short observation period of 15 min. The parameters of the flecainide concentration-time equation, expressed as $\text{concentration} = Ae^{-\alpha t}$: $A = 1.8 \pm 0.9$ mcg/ml and $\alpha = 0.07 \pm 0.04/\text{min}$ ($n=6$). We estimated the minimum antiarrhythmic flecainide plasma concentration for canine halothane-adrenaline-induced arrhythmia as 1.5 ± 0.5 mcg/ml.

Effects of flecainide on two-stage coronary ligation-induced arrhythmia:

Twenty-four hours after coronary ligation, the dogs showed ventricular tachycardia (-5 and 0 time values in Fig. 3). Preliminary experiments using $3-6$ mg/kg flecainide, i.v., showed that 6 mg/kg induced vomiting, but that the antiarrhythmic effect of this dose was not stronger than that of 3 mg/kg. Therefore, a 3 mg/kg dose was used. Flecainide decreased the total heart rate, increased the number of conducted beats, and decreased the arrhythmic ratio for up to 10 min. The atrial rate was unaffected, but the blood pressure increased for 60 min. The plasma concentration time curve corresponded with predictions of a two compartment model. The parameters of the curve of the 24 hr experiments: $A=17.9\pm 28.2$ mcg/ml, $\alpha=1.48\pm 1.12$ /min, $B=1.87\pm 0.54$ mcg/ml and $\beta=0.02\pm 0.006$ /min ($n=6$). The minimum antiarrhythmic plasma concentration for the canine 24 hr coronary ligation-induced arrhythmias was calculated as 1.5 ± 0.4 mcg/ml (at 10 min).

DISCUSSION

The present study confirmed previous reports²⁾ that flecainide was effective in 3 canine ventricular arrhythmia models. The minimum antiarrhythmic plasma concentration of flecainide for digitalis-induced arrhythmia, 0.9 mcg/ml, was significantly lower than the concentrations for adrenaline and coronary ligation-induced arrhythmias. These data indicate that flecainide is most effective on digitalis-induced arrhythmia. Flecainide is a potent antiarrhythmic drug, as judged by the low effective plasma concentrations. It resembles cibenzoline and aprindine,¹³⁾ in its pattern of efficacy in these 3 canine models. From these results, flecainide can be expected to be effective on the some clinical ventricular arrhythmias as other class 1 drugs.⁸⁾⁻¹³⁾ These results also confirmed our previous conclusion that canine and human antiarrhythmic concentrations of drugs are similar,^{11),13)} since $0.2-1.5$ mcg/ml is reported to be the clinical effective plasma concentration of flecainide.¹⁶⁾

Electrophysiological studies of flecainide with isolated rabbit atria,^{1),17)} have indicated that flecainide decreases the max dV/dt of cardiac action potential at concentrations around $1-5$ mM, which corresponds to $0.4-2$ mcg/ml. If the plasma protein binding data of about 40% in humans¹⁶⁾ are applicable to dogs, a concentration of $0.7-3.3$ mcg/ml flecainide in plasma can suppress cardiac Na^+ channels. This concentration corresponds to our minimum effective plasma concentration data of 0.9 to 1.5 mcg/ml. Therefore, Na^+ channel blockade may be the mechanism of suppression of digitalis

and coronary ligation arrhythmias.¹³⁾ We also assumed that suppression of Ca^{2+} channels might be the mechanism of suppression of adrenaline induced arrhythmia. *In vitro* cardiac electrophysiological studies indicated that flecainide in high concentrations (5 to 10 mcg/ml) slightly suppressed Ca^{2+} -mediated cardiac action potentials.¹⁸⁾ This effect might be responsible for the suppression of adrenaline arrhythmias by flecainide.

The association and dissociation kinetics of flecainide binding to Na^{+} channels have suggested that it belongs to a slow drug.¹⁷⁾ This class Ic property of flecainide seems to be reflected in the relatively slow onset of antiarrhythmic action, even after intravenous injection (Figs. 1–3). Other class I antiarrhythmic drugs have shown a maximum antiarrhythmic effect within 1 min of injection,^{8)–12)} but for flecainide it took about 3 to 5 min to reach its maximum effect. Another Ic drug, E-0747, showed the same characteristics as flecainide.¹⁹⁾ Therefore, a slow action may be the only characteristic property of class Ic drugs, because other characteristics are common to flecainide and class Ia or Ib drugs, such as phenytoin, mexiletine, tocainide, cibenzoline, aprindine and propafenone. It is not surprising that classification of class I drugs by the kinetic properties does not correlate well with their effectiveness on *in vivo* canine arrhythmia models. Other approaches such as making arrhythmia models of rapid or relatively slow ventricular tachycardia and examining different class I drugs may give more appropriate classification comparable to that based on the kinetic properties of the drugs.

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