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*J Clin Invest.* 1983;71(5):1457-1466. <https://doi.org/10.1172/JCI110899>.

### Research Article

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# Acute Effects of Amiodarone upon the Canine Sinus Node and Atrioventricular Junctional Region

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**ABSTRACT** Amiodarone was selectively perfused into the sinus node artery and atrioventricular node artery of 51 dogs. Amiodarone had an immediate negative chronotropic and dromotropic effect. Threshold concentration was 2.5  $\mu\text{g}/\text{ml}$ . 25 and 50  $\mu\text{g}/\text{ml}$  of amiodarone injected into the sinus node artery slowed the heart by  $25.6 \pm 3.1$  and  $33.7 \pm 2.6$  beats/min (mean  $\pm$  1 SEM), respectively. Amiodarone 25 and 50  $\mu\text{g}/\text{ml}$  injected into the AV node artery during AV junctional rhythm slowed the AV junctional pacemaker by  $12.2 \pm 1.8$  and  $17.4 \pm 1.7$  beats/min, respectively. Injections of amiodarone into the AV node artery during sinus rhythm regularly increased AV conduction time sometimes causing 2° AV block at the highest concentration used. Impaired conduction was exclusively measured at the level of the A-H interval in the His electrogram. Neither atropine nor propranolol prevented the negative chronotropic effects of amiodarone. Amiodarone had no significant effect on sinus node response to either stellate stimulation or intranodal administration of norepinephrine. The negative chronotropic action of amiodarone was significantly enhanced when amiodarone was administered in a perfusate containing low (0.6 mM) instead of normal calcium. Taken collectively these observations indicate that amiodarone has immediate depressant electrophysiologic effects on both the sinus node and the AV junction and that these early effects might involve the blockade of the slow channel.

## INTRODUCTION

Amiodarone was originally introduced as an antiarrhythmic drug (1, 2) but is now widely used for the therapy

of both supraventricular and ventricular arrhythmias (3–9). Parenteral administration of the benzofuran derivative as a pseudosolution in dogs (2, 10) and with tween 80 in man (11) causes a prompt decrease in peripheral and coronary vascular resistance (2, 10). The ensuing reflexly mediated response usually includes a brief increase in heart rate (11–13) that often obscures the direct negative chronotropic (14) and inotropic (2) action of amiodarone. Whereas the acute hemodynamic effects abate rapidly after intravenous administration (2, 11, 13), reported maximal antiarrhythmic efficacy after per os administration appears to be achieved after several days or weeks (4, 5, 7, 8, 15, 16). Amiodarone prolongs the duration of the action potential without concomitantly changing the resting membrane potential (10, 14, 17), yet it is still not clear whether amiodarone's antiarrhythmic properties can be ascribed to these electrophysiologic changes (18).

Recently, Goupil and Lenfant (14) demonstrated that amiodarone immediately decreased the slope of diastolic depolarization of rabbit sinus node cells. In the present study we have examined the acute chronotropic action of amiodarone selectively perfused into the sinus node artery of the dog and compared this to both chronotropic and dromotropic actions following selective perfusion into the atrioventricular (AV)<sup>1</sup> node artery. Each effect was investigated in the presence and absence of autonomic blockade.

## METHODS

51 mongrel dogs of either sex were anesthetized with sodium pentobarbital (30 mg/kg i.v.). Ventilation with room air was maintained through a cuffed endotracheal tube. The chest was opened in the right fourth intercostal space and the sinus node artery or the AV node artery or both were cannulated

This work was presented in part at the 31st Annual Scientific Session of the American College of Cardiology in Atlanta, GA, 25–29 April 1982.

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Received for publication 7 September 1982 and in revised form 6 December 1983.

<sup>1</sup> Abbreviations used in this paper: AV, atrioventricular; TEA, tetraethylammonium.

with a small polyethylene catheter. The entire remaining coronary circulation as well as the normal innervation of the heart were preserved intact. Details of these procedures have been published previously (19–21).

Bipolar electrograms were recorded from electrodes sutured on or near the sinus node, Bachmann's bundle, the left atrial appendage, the eustachian ridge, and on the sulcus terminalis at a point equidistant between the sinus node and the eustachian ridge. His bundle electrograms were obtained in each dog using a standard 5F or 6F pacing electrode catheter (10-mm interpolar distance) advanced into the aortic root (22). A surface electrocardiogram, usually lead II, was also recorded in every experiment. Central aortic pressure was routinely recorded from a catheter introduced through the right carotid or left subclavian artery. A tachogram triggered from a bipolar atrial electrogram was routinely obtained. Measurements of conduction time were always made from records obtained at a paper speed of 200 mm/s.

Adequacy of sinus node perfusion is routinely determined with a control injection of Ringer's solution (2 ml) into the sinus node artery (19, 21). The characteristic response of the sinus node to such a Ringer's injection is an immediate but transient decrease in sinus rate with a rapid return to control (Fig. 1) (19, 21). An immediate decrease in sinus rate of at least 20 beats/min is required to be included in these studies (21). Injection of drugs that exhibit a negative chronotropic action not only prevent the rapid return to control sinus rate but often exaggerate the immediate injection bradycardia. For computation in the present study we took the maximum negative chronotropic effect of amiodarone that occurred 30 s after the onset of injection.

Stable AV junctional rhythm was achieved by injecting either 2 ml of eserine (50  $\mu\text{g}/\text{ml}$ ) or 2 ml of racemic verapamil (5–10  $\mu\text{g}/\text{ml}$ ) into the sinus node artery (21, 23). Adequacy of AV junctional perfusion during AV junctional rhythm is also determined with a control injection of Ringer's solution (2 ml) into the AV node artery (21). The characteristic response of the AV junction to such an injection is also an immediate transient AV junctional bradycardia that rapidly returns to control (21). To be included in these studies Ringer's solution injected into the AV node artery must result in an AV junctional bradycardia of at least 12 beats/min (21). After injection of amiodarone computations were also made at the peak of the negative chronotropic action that occurred 30 s after onset of injection. When assessing changes in AV conduction time during sinus rhythm, perfusion was judged to be adequate only if the injection of acetylcholine (0.1  $\mu\text{g}/\text{ml}$ , 2 ml) into the AV node artery caused an immediate complete AV block for at least 2 s. Injection of amiodarone into the AV node artery during sinus rhythm prolongs the A-H interval in the His bundle electrogram. When second degree heart block developed the longest A-H interval preceding the first dropped beat was used for computation.

Amiodarone is a poorly soluble compound (2). As a consequence a variety of adjuvants including ethyl alcohol (24), dimethylformamide (25), and more recently, tween 80 (11) have been used to enhance the drug's solubility. For almost a decade solutions of amiodarone were first prepared from a 5% aqueous stock solution made up in distilled water at a slightly acid pH through gentle warming in a water bath at a temperature of  $\sim 70^\circ\text{C}$  (2). Due to surface tension properties of the amiodarone in water, the solution is in effect a pseudosolution (2). At concentrations  $<1\%$  in either distilled water or saline or plasma the solution slowly precipitates (2). Addition of small amounts of Triton X-100 (pur-

chased from Sigma Chemical Co., St. Louis, MO), however, and gentle warming of the solute up to  $70^\circ\text{C}$  readily permitted true solution in concentrations of 2.5–100  $\mu\text{g}/\text{ml}$  of amiodarone. To achieve 100  $\mu\text{g}/\text{ml}$  of amiodarone we took 0.2 ml of a standard 5% solution of amiodarone in distilled water and added this small volume slowly with a micropipette in 99.8 ml of Ringer's solution to which we had added 60  $\mu\text{l}$  of Triton X-100 previously. Triton X-100 was added slowly (during 10 min) with a micropipette under continuous stirring. The detergent itself used to achieve concentrations of up to 50  $\mu\text{g}/\text{ml}$  of amiodarone had no chronotropic or dromotropic effects, when injected alone into the sinus node or AV node artery. Because intranodal administration of the highest concentration of detergent alone (that used to achieve 100  $\mu\text{g}/\text{ml}$  of amiodarone) caused a minor transient sinus bradycardia we only report in this study the effects of amiodarone at concentrations up to 50  $\mu\text{g}/\text{ml}$ .

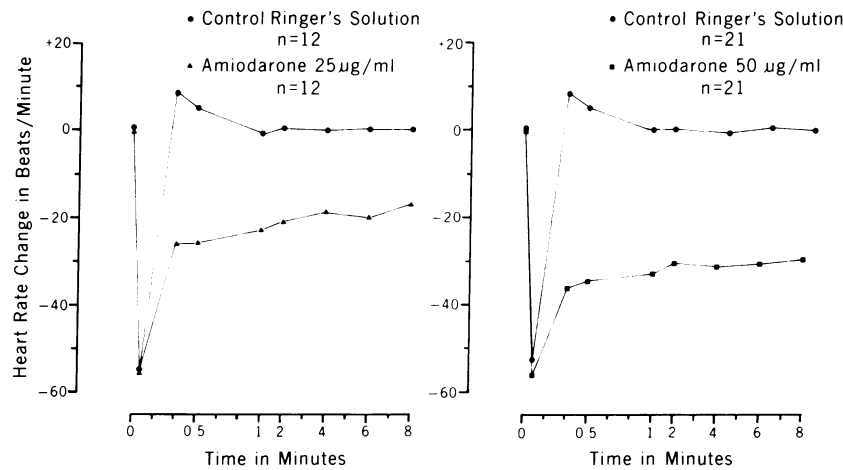
In seven dogs right stellate ganglion stimulations were performed before and after injection of amiodarone into the sinus node artery. Square wave stimulations were performed with a constant current (between 1.5 and 2.5 mA). The trains lasted 10 s, stimulus duration was 2 ms, and the frequencies selected were 0.5, 1, 2, and 4 Hz. In six other dogs the chronotropic responses of the sinus node to norepinephrine 0.006, 0.012, 0.025, and 0.05, and 0.1  $\mu\text{g}/\text{ml}$  were examined before and after intranodal amiodarone. In four other dogs amiodarone was injected into the sinus node artery following systemic autonomic blockade. The latter was achieved with 0.5 mg/kg of atropine sulfate, 1 mg/kg of propranolol hydrochloride, and 1 mg/kg i.v. of phentolamine mesylate. The chronotropic effects of amiodarone diluted in a perfusate containing a low concentration of calcium (0.6 mM) instead of the usual 2.5 mM of the normal Ringer's solution were examined in five dogs. Finally, we also examined the chronotropic action of amiodarone diluted in tween 80 (commercially available injectable form) in five dogs.

The results are expressed as means  $\pm 1$  SE of the mean. In all cases the data were analyzed using an analysis of variance model. The model used was an incomplete block design. Comparisons among dose response means were made using Duncan's multiple range test (26).

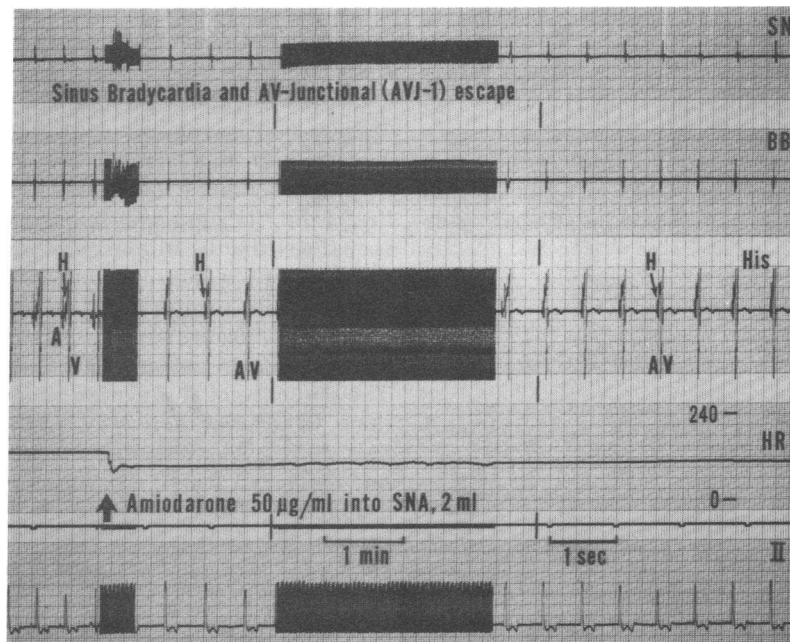
## RESULTS

*Effects of amiodarone on the sinus node.* In the concentrations used (2.5, 25, and 50  $\mu\text{g}/\text{ml}$ , 2 ml) amiodarone always caused a negative and only negative chronotropic effect (Fig. 1). The extent of sinus slowing was dose dependent, averaging  $3.4 \pm 1.2$ ,  $25.6 \pm 3.1$ , and  $33.7 \pm 2.6$  beats/min at the respective concentrations indicated above. In 6 of 21 dogs that received 50  $\mu\text{g}/\text{ml}$  of amiodarone (Fig. 2) the resulting sinus bradycardia was so profound that an AV junctional escape rhythm emerged. In those experiments it was thus not possible to measure the true magnitude of the negative chronotropic action. The duration of the sinus bradycardia is exceptionally prolonged; after 1 h of observation following 50  $\mu\text{g}/\text{ml}$  of intranodal amiodarone, there is essentially little or no recovery. In five other dogs amiodarone (25  $\mu\text{g}/\text{ml}$ ) but dissolved with tween 80 was injected into the sinus node artery. These injections caused an immediate sinus bradycar-

# ACUTE DIRECT NEGATIVE CHRONOTROPIC EFFECT OF AMIODARONE SELECTIVELY PERFUSED INTO SINUS NODE ARTERY



**FIGURE 1** Negative chronotropic effect of amiodarone 25 and 50  $\mu\text{g/ml}$  injected selectively into the sinus node artery. Only four dogs received both 25 and 50  $\mu\text{g/ml}$  of amiodarone, the remaining dogs received either 25- or the 50- $\mu\text{g/ml}$  concentration. Each dot, square, and triangle represents the mean values observed ( $n$  = number of dogs). The dots represent the respective control injections of Ringer's solution. Note the immediate injection bradycardia occurring within 5 s and the postinjection tachycardia occurring within the first 30 s after injection. Both responses are characteristics of the method (21). After amiodarone there is a dose-dependent bradycardia. Changes in heart rate are expressed in beats/minute.



**FIGURE 2** Negative chronotropic effect of amiodarone, 50  $\mu\text{g/ml}$ , 2 ml, injected into the sinus node artery during sinus rhythm. From top to bottom the recordings are bipolar electrograms from the vicinity of the sinus node (SN), Bachmann's bundle (BB), His bundle electrogram (His), heart rate (HR), and electrocardiogram, lead II. Symbols are A (atrium), H (His spike), and V (right ventricle). In this example amiodarone 50  $\mu\text{g/ml}$ , 2 ml, slows the sinus node to such an extent that the next subsidiary pacemaker, located in the AV junctional region escapes.

dia that averaged  $28.8 \pm 4.1$  beats/min 30 s after onset of injection. Neither the extent nor the time course of the bradycardia were any different from the one observed when amiodarone was dissolved with Triton X-100.

The F-test for overall regression = 5.72 is significant,  $P < 0.0010$ . The multiple correlation coefficient,  $r^2 = 0.922$ . The F-test for dose response is  $F = 46.81$  which is significant with  $P < 0.0001$ . A level 0.05 Duncan's test showed that the mean response at each dose level was different from the other two levels.

**Effect of amiodarone upon AV junctional rhythm.** During stable AV junctional rhythm obtained by perfusing either eserine or verapamil into the sinus node artery the injection of amiodarone (2.5, 25, and 50,  $\mu\text{g/ml}$ ) into the AV node artery caused an AV junctional bradycardia that averaged  $1.9 \pm 0.7$ ,  $12.2 \pm 1.8$ , and  $17.4 \pm 1.7$  beats/min, respectively (Fig. 3). AV junctional slowing developed rapidly and peak effect was observed 30 s after onset of injection. Since depression of sinus node automaticity below the level of AV junctional automaticity is difficult to maintain over a period longer than 1 h, exact assessment of the duration of the negative chronotropic effect of amiodarone on the AV junctional pacemaker was not attempted.

The F-test for overall regression,  $F = 6.59$  is significant,  $P < 0.0007$ . The multiple correlation coefficient,  $r^2 = 0.850$ . The F-test for dose response is  $F = 26.19$ , which is significant with  $P < 0.0001$ . A level 0.05 Duncan's test showed that the mean response at each dose level was different from the other two levels.

**Effect of amiodarone upon AV conduction.** In six dogs with a control sinus rate of  $152.4 \pm 4.4$  beats/min amiodarone was selectively perfused into the AV node

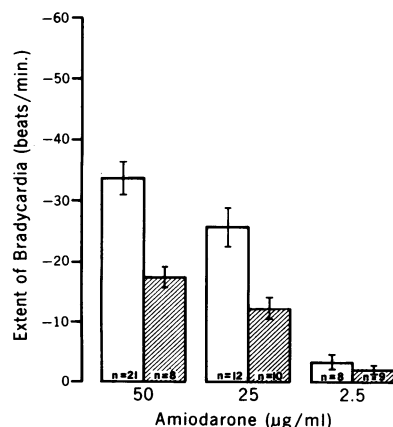


FIGURE 3 Comparison between the negative chronotropic effect of amiodarone 2.5, 25, and 50  $\mu\text{g/ml}$ , 2 ml, injected into the sinus node artery during sinus rhythm (□) and into the AV node artery during AV junctional rhythm (▨). Vertical bars show SEM.

artery during normal sinus rhythm. The control A-H interval in the His bundle electrogram averaged  $37.1 \pm 1.4$  ms. Whereas 2.5  $\mu\text{g/ml}$ , 2 ml of amiodarone had no effect on the duration of the A-H interval, 25  $\mu\text{g/ml}$  caused a  $5.3 \pm 2.4$ -ms prolongation. When the next higher concentration was used (50  $\mu\text{g/ml}$ ) the prolongation averaged  $27.2 \pm 12.2$  ms, respectively (Fig. 4). After 50  $\mu\text{g/ml}$  of amiodarone, one of the six dogs developed 2° heart block (Wenckebach periodicity 3:2) for 3.5 min.

The F-test for overall regression ( $F = 6.51$ ) is significant,  $P < 0.0040$ . The multiple correlation coefficient  $r^2 = 0.838$ . The F-test for significant dose-response is  $F = 21.73$ , which is significant,  $P < 0.0002$ . A level 0.05 Duncan's test showed that the mean response at each dose level was different from the other two levels.

**Effect of amiodarone on sinus node response to intranodal norepinephrine.** In six dogs with a control sinus rate of  $139.2 \pm 9.2$  beats/min the sinus node response to norepinephrine (0.006, 0.012, 0.025, 0.05, and 0.1  $\mu\text{g/ml}$ , 2 ml) was examined before amiodarone. Fig. 5 is a representative example of such an experiment. Injection of amiodarone (50  $\mu\text{g/ml}$ ) into the sinus node artery slowed the sinus rate by 30 beats/min, to a new level of  $109.2 \pm 10.9$  beats/min. After 1 min the sinus rate averaged  $115.3 \pm 10.3$  beats/min and remained stable at that level for the entire duration of the experiment. Then the same concentrations of norepinephrine were again administered into the sinus node artery. Table I summarizes the results obtained, expressed both as absolute sinus rate increase (in beats/minute) and as a percent change in heart rate. Although amiodarone had caused a significant sinus slowing, it did not significantly change the sinus node response to directly administered norepinephrine.

**Effect of amiodarone on sinus node response to right stellate stimulation.** In seven other dogs with a control sinus rate of  $142.9 \pm 6.2$  beats/min right stellate stimulation was conducted at 0.5, 1, 2, and 4 Hz. The sequence of stimulation frequency was randomly done. In order of increasing frequency of stimulus signals, sinus acceleration was  $8.4 \pm 1.6$ ,  $21.1 \pm 3.1$ ,  $43.1 \pm 5.5$ , and  $61.4 \pm 6.5$  beats/min, respectively. Injection of amiodarone (25  $\mu\text{g/ml}$ ) into the sinus node artery decreased the sinus rate to  $118.9 \pm 7.3$  beats/min. Three successive random stimulation frequency response curves were again obtained in the ensuing 15 min after intranodal amiodarone. As illustrated in a characteristic experiment depicted in Fig. 6, the sinus rate increases from stellate stimulations before and after amiodarone are virtually identical.

Two questions were considered in the statistical analysis of this data. Does amiodarone (25  $\mu\text{g/ml}$ ) into the sinus node artery change the control heart rate?

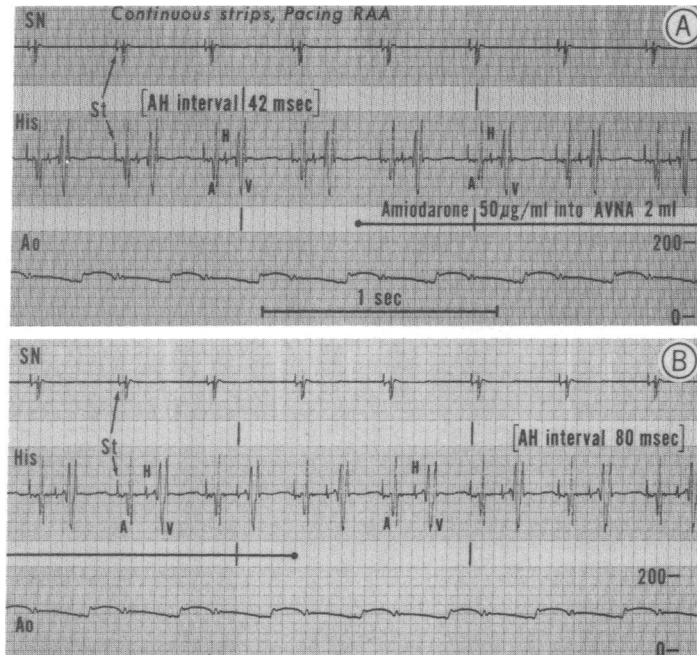


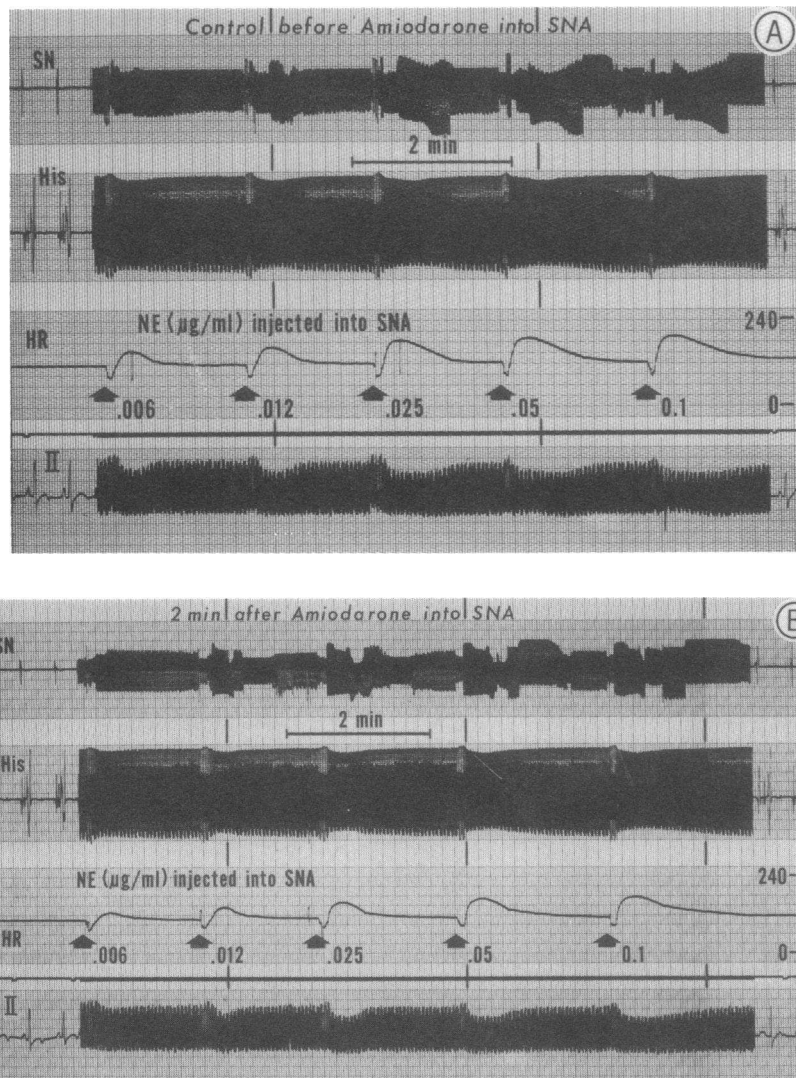
FIGURE 4 Negative dromotropic effect of amiodarone, 50  $\mu\text{g}/\text{ml}$ , 2 ml, injected into the AV node artery. The right atrium is continuously paced at a rate of 158 beats/min. From top to bottom the recordings are bipolar electrograms from the vicinity of the sinus node (SN) and His. St is the stimulus artifact. The bottom trace depicts aortic pressure (Ao) scaled in millimeters of mercury. Note the marked prolongation of the AH interval during the injection of amiodarone.

and does the percent heart rate change resulting from nerve stimulation change as a result of intranodal amiodarone? To answer the first question, a paired  $t$  test was applied to the data. It was found that there was a significant change in the control rate ( $t = 5.76$ , d.f. = 6,  $P < 0.005$ ). To answer the second question the percent change in heart rate was analyzed by analysis of variance. A weighted  $2 \times 4$  factorial model was used for the analysis. Tests for differences in response before and after stimulation were not significant ( $P > 0.47$ ).

**Effect of amiodarone on sinus node automaticity in presence of autonomic blockade.** In four dogs with a control sinus rate of  $148.0 \pm 7.5$  beats/min autonomic blockade was produced by an intravenous administration of atropine (0.5 mg/kg), propranolol (1 mg/kg), and phentolamine (1 mg/kg). Variable amounts of rheomacrodex were continuously administered intravenously in order to maintain mean aortic pressure within 10% of control. Atropine accelerated mean heart rate slightly to  $152.3 \pm 8.2$  beats/min. Two dogs then received first propranolol and then phentolamine, whereas the other two dogs received first phentolamine and then propranolol. After such autonomic blockade the final stable sinus rate was  $112.3 \pm 10.5$  beats/min.

Subsequent administration of amiodarone (25  $\mu\text{g}/\text{ml}$ , 2 ml) into the sinus node artery promptly decreased the sinus rate to  $77.3 \pm 19.4$  beats/min within 1 min, rate then stabilizing at  $81.3 \pm 16.9$  beats/min after  $\sim 3$  min. Every dog remained in sinus rhythm. Autonomic blockade thus has little or no influence on the negative chronotropic action of amiodarone.

**Effect of amiodarone on sinus node automaticity when administered in a perfusate deficient in calcium.** In five other dogs with a control sinus rate of  $143.8 \pm 7.3$  beats/min, amiodarone (25  $\mu\text{g}/\text{ml}$ , 2 ml) was diluted in a perfusate containing 0.6 mM of calcium instead of the 2.5 mM calcium of Ringer's solution, and then was perfused into the sinus node artery. Perfusate containing low calcium (0.6 mM) alone had no other effect than Ringer's solution (Fig. 7). In every dog receiving amiodarone in low calcium containing perfusate the sinus rate decreased immediately and to such an extent that an AV junctional pacemaker escaped (Fig. 8). AV junctional rhythm lasted from 16 s to 16 min, and the rate of this AV junctional rhythm averaged  $92.6 \pm 6.7$  beats/min. Thus, amiodarone diluted in a calcium-deficient perfusate had a greater negative chronotropic action than when diluted in usual Ringer's solution.



**FIGURE 5** Norepinephrine dose-response curves before and after amiodarone. Panel A shows control dose-response curves to norepinephrine 0.006, 0.012, 0.025, 0.05, and 0.1  $\mu\text{g/ml}$  (each bolus 2 ml) injected into the sinus node artery. Panel B demonstrates norepinephrine dose-response curves 2 min after an injection of 50  $\mu\text{g/ml}$  of amiodarone into the sinus node artery, slowing the sinus node by 30 beats. Although in absolute number of beats/minute the positive chronotropic response to norepinephrine is slightly, but not significantly, diminished after amiodarone the relative rate increase in percent is unchanged, compared with the one obtained before amiodarone.

## DISCUSSION

Selective administration of amiodarone into the sinus node artery during sinus rhythm and into the AV node artery during AV junctional rhythm causes an immediate and long lasting negative chronotropic effect. Perfusion of the AV junctional region with amiodarone during sinus rhythm results in a rapidly progressive prolongation of the P-R interval. This impaired AV

conduction was exclusively measured at the level of the A-H interval in the His electrogram. On comparing the negative chronotropic effect on the sinus node with that on the AV junction by the same concentrations of amiodarone, reduction of sinus rate was greater than the slowing of the AV junctional rate. These observations closely resemble those made previously in our laboratory when studying the effects of racemic verapamil (23, 27) and its isomers (28).



TABLE I  
Sinus Node Response to Norepinephrine before and after  
Amiodarone 50  $\mu\text{g/ml}$ , 2 ml Injected Selectively into  
the Sinus Node Artery

	Before		After	
	Rate increase		Rate increase	
	beats/min	%	beats/min	%
Control	139.2 $\pm$ 9.2		115.3 $\pm$ 10.3	
$\mu\text{g/ml}$				
NE 0.006	160.2 $\pm$ 6.1	16.9 $\pm$ 5.7	129.7 $\pm$ 9.9	13.4 $\pm$ 4.2
NE 0.012	178.0 $\pm$ 4.9	30.8 $\pm$ 8.3	146.2 $\pm$ 9.5	29.1 $\pm$ 7.7
NE 0.025	188.8 $\pm$ 4.6	39.3 $\pm$ 9.9	161.2 $\pm$ 9.4	43.2 $\pm$ 9.6
NE 0.05	200.7 $\pm$ 6.2	48.1 $\pm$ 11.2	175.8 $\pm$ 7.4	56.9 $\pm$ 9.9
NE 0.10	208.8 $\pm$ 7.3	54.0 $\pm$ 11.8	188.5 $\pm$ 7.9	68.0 $\pm$ 10.3

NE, norepinephrine.

The recently introduced injectable amiodarone (11) contains tween 80, but the detergent itself can cause hypotension in higher concentrations (29). In the present study we used small amounts of Triton X-100 to enhance amiodarone's solubility. Injection of the diluent alone had no significant depressant effect of its own. Both the negative chronotropic and dromotropic actions observed in this study are thus attributable to an immediate depressing effect of amiodarone. Since complete autonomic blockade did not prevent these acute effects we can further conclude that amiodarone has a direct negative chronotropic and dromotropic action.

Amiodarone does not interfere with the sinus node response to intranodal delivery of norepinephrine. Others (30) however, have described some noncompetitive interaction between amiodarone and catecholamines. Polster and Broekhuysen (31) for example, proposed that the amiodarone interaction (decrease in cyclic AMP) occurred at a point beyond the step of the transmitter binding to the adrenoceptor. Similarly, Lubbe et al. (32) who studied the protective effect of amiodarone against ventricular fibrillation in the rat heart found that in ischemic myocardium the protective electrophysiologic effect was accompanied by a lesser increase in cyclic AMP. In contrast to Bacq et al. (24), who found that amiodarone reduced sympathetic transmitter release in the isolated spleen of the cat, our results in the canine sinus node indicate that concentrations of amiodarone that can be achieved clinically (at least 1–5  $\mu\text{g/ml}$ ) do not cause significant sympathetic neuronal blockade.

In man (3, 4, 7, 11–13, 15) and in experimental animals (2, 33) amiodarone's chronotropic effect has been variously described as positive (11, 13, 25), negative (3, 4, 15, 32–34), or biphasic, with a brief initial increase soon followed by a long-lasting bradycardia (12). The transient positive chronotropic responses that have been observed after intravenous administration are most likely due to the resulting initial transient hypotension that elicits adaptive reflex responses that have both a vagolytic and a sympathomimetic component.

Concerning acute dromotropic effects, several investigators have reported an increase in the P-R interval (35–37) and as in our study the prolongation in

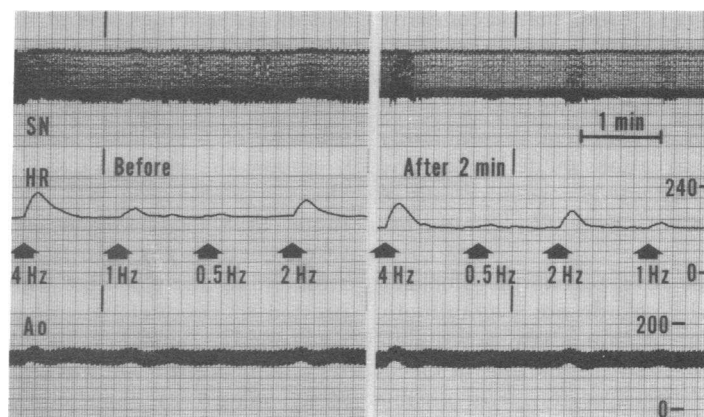


FIGURE 6 Right stellate stimulation frequency response curves before and after 25  $\mu\text{g/ml}$  amiodarone injected into the sinus node artery. The left side panel (control) shows the positive chronotropic response of the sinus node to stellate stimulation with 0.5, 1, 2, and 4 Hz before amiodarone. Intranodal amiodarone slowed the sinus node by 30 beats. 2 min after injection (right side panel) stellate stimulation was repeated. Note that the percent sinus rate change is virtually identical to control.



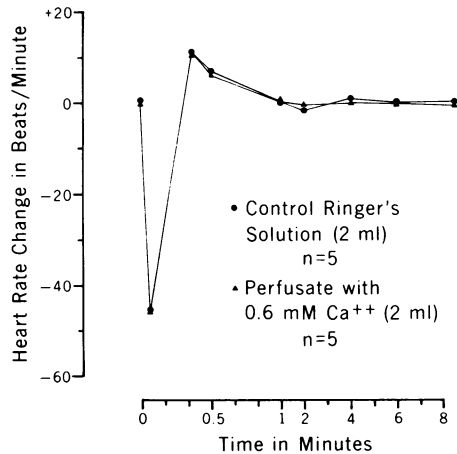


FIGURE 7 The same chronotropic responses are observed when Ringer's perfusate containing low calcium are injected into the sinus node artery. Duration of injection 4 s. Total volume 2 ml.

conduction time was exclusively measured at the level of the A-H interval of the His electrogram (33, 35).

The antiarrhythmic action of amiodarone has often been attributed to two distinct properties of the drug. There is an acute effect that is thought to be related to a noncompetitive antiadrenergic property (30, 31) and a delayed effect that is due to the prolongation of the action potential in both atrial and ventricular cells (10, 14, 17, 34). Long-term oral administration of amiodarone slows the sinus rate (4, 15, 34), increases AV conduction time (5, 8, 38), and may even slow conduction in the specialized tissue distal to the AV node (5). Even with the brief and sometimes intense autonomic adjustments (increase in sympathetic and concomitant decrease in vagal tone) that accompany the transient hypotension caused by intravenous rapid amiodarone, (30 s) (3, 33) or slow (5 min) (36, 37); administration of the drug usually slows sinus rate and prolongs AV conduction (3, 10, 37). The present study is in accord with these observations made in man and further confirms that the effective concentrations of amiodarone are similar in both dogs (25, 33) and humans (11, 13, 38).

Since neither noncompetitive adrenergic blockade nor sympathetic neuronal blockade nor vagomimetic influences can account for the immediate slowing effect by amiodarone on rhythms originating in either the sinus node or AV junctional region, it is possible that these immediate electrophysiologic effects have the same underlying cause as the delayed effects largely attributed to a decrease in potassium conductance (10, 14, 17). For example, Goupil and Lenfant (14) found that within minutes amiodarone increased the action potential duration of rabbit sinus node pace-

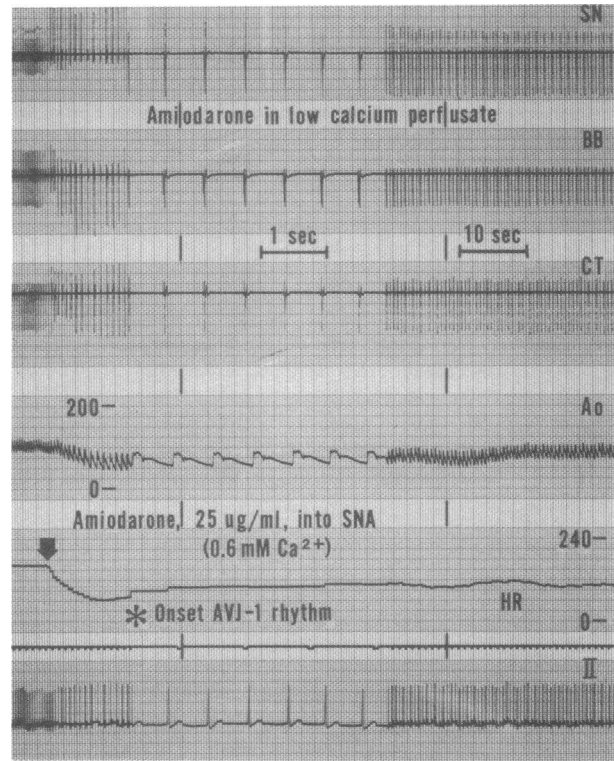


FIGURE 8 This figure shows that amiodarone has a more powerful negative chronotropic action on the sinus node when administered in a perfusate containing low calcium. In low calcium perfusate amiodarone 25  $\mu$ g/ml consistently slows the sinus node to such an extent that the next subsidiary pacemaker, located in the AV junction, escapes.

maker cells. These authors attributed that cellular effect either to a decrease of the delayed outward current, or to an increase of the slow inward current inactivation time constant or both. Tetraethylammonium, which is widely believed to be a relatively selective inhibitor of K<sup>+</sup> conductance (39–41), has been shown to oppose the increase in K<sup>+</sup> conductance that brings about repolarization (41). In related experiments we found that TEA directly increased sinus rate even when contributing effects from release of catecholamines and acetylcholine were controlled (42). If depressed K<sup>+</sup> conductance enhances the rate of firing of sinus node pacemaker cells either directly or indirectly by enhancing the net inward current during the diastolic phase (42), then another explanation must be found for the negative chronotropic action of amiodarone.

One explanation for the reported decrease in the slope of diastolic depolarization produced by amiodarone is that it increases the deactivation time constant of the delayed outward current (14). However,

the circumstantial evidence obtained with TEA in the canine sinus node (showing the exact opposite chronotropic response) (42) argues against this possibility. On the other hand, the negative chronotropic action of amiodarone could, at least in part, be mediated through a reduction of an inward background current similar to the time-independent current previously described in the frog atrial myocytes (43) and sheep Purkinje cells (44).

There is another important observation by Goupil and Lenfant (14), that is that amiodarone significantly decreased the amplitude of the action potential of the sinus node cells. Based on their own previous findings (45) they suggested that this effect was due to a decrease in slow inward current (14). Since the slow inward current has also been shown to contribute to the latter part of diastolic depolarization (46), it is plausible to postulate that amiodarone's immediate negative chronotropic effect could, at least partly, be mediated through a decrease of slow channel conductance. This possibility is supported by our experiments since amiodarone was indeed significantly more powerful in depressing sinus node automaticity when administered in a perfusate containing low calcium. This assumption is particularly attractive because many of amiodarone's immediate electrophysiologic effects occur at strategic sites (sinus node and AV junctional region) where proper function is critically dependent upon the integrity of the slow channel.

## ACKNOWLEDGMENTS

We are grateful to Dr. Charles R. Katholi from the Department of Biomathematics of the University of Alabama in Birmingham for the statistical treatment of the data.

This work was supported by the National Heart, Lung, and Blood Institute (program project grant HL 11,310 and Specialized Center of Research grant on Ischemic Heart Disease HL 17,667) and was conducted while Dr. Gloor was a Fellow of the Swiss National Foundation.

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