Electrophysiological Effects of Disopyramide and Quinidine on Guinea Pig Atria and Canine Cardiac Purkinje Fibers

Dependence on Underlying Cholinergic Tone

MICHAEL J. MIRRO, AUGUST M. WATANABE, AND JOHN C. BAILEY

SUMMARY We studied the role of the anticholinergic properties of disopyramide and quinidine in mediating the electrophysiological effects of these agents on isolated cardiac tissue. In right atria, disopyramide, quinidine, and procainamide administered alone elicited negative chronotropic responses. However, after cholinergic stimulation with physostigmine (1 × 10^{-6} M), disopyramide and quinidine produced a positive chronotropic response. Procainamide, when administered under the same conditions of increased cholinergic stimulation, elicited negative chronotropic responses similar to those observed when it was given alone. In Purkinje fibers, disopyramide, quinidine, and procainamide superfused alone for 5 minutes significantly prolonged action potential duration (APD). When administered to Purkinje fibers pretreated with isoproterenol (1 × 10^{-7} M) plus acetylcholine (3 × 10^{-7} M), disopyramide and quinidine shortened APD. Atropine also shortened APD of fibers exposed to both isoproterenol and acetylcholine. In these experiments, isoproterenol consistently shortened APD, and acetylcholine consistently blunted these effects of isoproterenol. Thus, disopyramide and quinidine, alone, prolong APD, but when given in the setting of increased adrenergic-cholinergic stimulation, they shorten APD. Procainamide, when administered to Purkinje fibers pretreated with isoproterenol and acetylcholine, prolongs APD as when it is administered alone. These data demonstrate that disopyramide and quinidine exert important anticholinergic electrophysiological effects on the atria and the ventricular conducting systems. These anticholinergic effects may contribute to the antiarrhythmic and arrhythmogenic properties of these compounds. Circ Res 46:660-668, 1980

THE cholinergic limb of the autonomic nervous system regulates the electrophysiological properties of both atria (Hoffman and Suckling, 1953) and ventricles (Bailey et al., 1972; Kent et al., 1973; Corr and Gillis, 1974; Myers et al., 1974; Bailey et al., 1979). In atria, muscarinic receptor stimulation decreases spontaneous sinoatrial rate and modifies action potential configuration of atrial muscle (Hoffman and Suckling, 1953). In canine cardiac Purkinje fibers, muscarinic agonists produce electrophysiological effects by modulating the actions of β-adrenergic receptor stimulation on action potential configuration (Bailey et al., 1979). When muscarinic agonists act on nonautomatic Purkinje fibers, they produce minimal (Gadsby et al., 1978; Danilo et al., 1979) or no effects (Bailey et al., 1979) on action potential configuration, but when they act on Purkinje fibers that are under the influence of β-adrenergic receptor stimulation, they exert potent electrophysiological effects (Bailey et al., 1979).

Anticholinergic agents exert well-known electrophysiological effects on atria (Tanz et al., 1978), and recently, muscarinic antagonists have been shown to modify ventricular fibrillation threshold as well as Purkinje fiber action potential configuration (Corr and Gillis, 1974; Bailey et al., 1979). Drugs other than atropine and its analogues, which possess anticholinergic properties, also would be expected to produce electrophysiological effects on atria and ventricles because of their ability to antagonize the effects of muscarinic receptor activation.

The antiarrhythmic agents, disopyramide, quinidine, and procainamide, exert similar electrophysiological effects on automaticity, action potential duration, and refractoriness of atrial and ventricular tissues studied in vitro in the absence of cholinergic stimulation (Rosen and Hoffman, 1973). However, when studied in intact animals, disopyramide (Mokler and VanArman, 1962; Birkhead and Vaughan Williams, 1977) and quinidine (James and Nadeau, 1964; Wallace et al., 1966), but not procainamide (Josephson et al., 1973), have been shown to antagonize cholinergic effects on sinoatrial automaticity and AV nodal refractoriness. We have hypothesized, therefore, that disopyramide and quinidine...
might exert electrophysiological effects on cardiac tissues studied in vitro in the presence of muscarinic receptor activation that are different from their effects in the absence of muscarinic receptor activation. The purpose of the present study was to investigate in vitro the role of the anticholinergic properties of disopyramide and quinidine in mediating the electrophysiological effects of these agents on isolated cardiac tissues. The anticholinergic electrophysiological properties of these agents were compared to the effects of the muscarinic receptor blocking drug, atropine. Additionally, the electrophysiological anticholinergic effects of procainamide were compared with those of disopyramide and quinidine.

Methods

Experiments on Guinea Pig Right Atrium

Guinea pigs of either sex weighing 400–600 g were injected intraperitoneally with heparin sulfate (500 U) 30 minutes prior to use. Each guinea pig was stunned with a blow to the head, after which the heart was removed rapidly and placed in cool oxygenated Tyrode’s solution. The right atrium, including the area of the sinus node, was excised and pinned to the floor of a wax-bottomed Lucite muscle chamber (7-ml volume) and was superfused with Tyrode’s solution that was gassed with 95% O₂-5% CO₂. Temperature was maintained at 37 ± 0.5°C. The composition of the Tyrode’s solution was (mm): Na⁺, 138; K⁺, 4.0; Ca²⁺, 1.25; HCO⁻₃, 20.0; H₂PO₄⁻, 0.9; Mg²⁺, 0.5; and glucose, 5.5. The osmolarity of this solution was 285 mOsmol, and the pH was 7.40 ± 0.05.

Conventional microelectrode techniques were used (Draper and Weidmann, 1951). Data were displayed on a Tektronix 5100 series oscilloscope, photographed with a Tektronix C-95 Polaroid oscilloscope camera, and stored on an eight-channel Honeywell 7600 tape recorder. To record the rate of spontaneous electrical discharge of the right atria, an action potential-triggered tachometer was used. The tachometer produced a linear ramp that was triggered by the upstroke of each successive action potential. The tachometer was calibrated by stimulating the preparation at known constant basic cycle lengths and measuring the height of the ramp. Control spontaneous cycle length was recorded for at least 15 minutes prior to any experimental intervention.

Two series of experiments were conducted to determine the effects of each antiarrhythmic agent on spontaneous rate in the presence and absence of cholinergic stimulation. Each experiment was performed on a single preparation, and five to eight atria were studied at each drug concentration.

1. Experiments were performed in which disopyramide, quinidine, procainamide, or atropine was superfused for 5 minutes to quantify the direct atrial rate slowing effect of each drug.

2. Experiments also were conducted in which disopyramide, quinidine, procainamide, or atropine was superfused for 5 minutes over atria pretreated with physostigmine to analyze the anticholinergic effects. In these experiments, physostigmine 1 × 10⁻⁶ M was administered alone for 15 minutes, and then antiarrhythmic drug or atropine was added to the superfuse for an additional 5 minutes with continued physostigmine administration. Thus, changes in atrial rate produced by each antiarrhythmic drug were assessed after 5 minutes of drug exposure, in both the presence and absence of physostigmine. The concentrations of disopyramide, quinidine, and procainamide studied are comparable to concentrations that are considered to be therapeutic (Winkle et al., 1975).

Experiments on Canine Cardiac Purkinje Fibers

Adult mongrel dogs of either sex, weighing 12–15 kg, were anesthetized with sodium pentobarbital (30 mg/kg, intravenously). Their hearts were removed rapidly through a right thoracotomy and immersed in cool oxygenated Tyrode’s solution. Purkinje fibers were excised and fixed to the floor of a wax-bottomed Lucite muscle chamber (7-ml volume) and superfused with Tyrode’s solution.

The preparations were stimulated at a constant basic cycle length of 800 msec using a bipolar extracellular stainless steel electrode. Conventional microelectrode techniques were used (Draper and Weidmann, 1951). Transmembrane resting potential, action potential amplitude, action potential duration at 100% repolarization, and dV/dt phase 0 were monitored. Continuous single cell impalements were maintained throughout all experiments. A 15-minute control period, during which no detectable alterations of the monitored action potential parameters were noted, preceded the initiation of each experiment.

Two separate series of experiments on Purkinje fibers were performed to determine the effect of each drug on action potential duration in the presence or absence of muscarinic receptor activation.

1. Disopyramide, quinidine, procainamide, or atropine was superfused alone for 5 minutes over Purkinje fibers to determine their direct effects on action potential duration. These effects were reversed after a 10-minute washout period.

2. Experiments also were conducted in which each of the drugs was superfused for 5 minutes over Purkinje fibers exposed to isoproterenol and acetylecholine to determine the effects of the antiarrhythmic drugs on action potential duration in the presence of simultaneous β-adrenergic and muscarinic receptor activation. Sodium metabisulfide (1 × 10⁻⁶ M) was added to all isoproterenol-containing solutions to prevent oxidation.

In these experiments, after a control period, isoproterenol (1 × 10⁻⁷ M) was added first and superfused for 5 minutes. This concentration of isopro-
terenol consistently produced significant shortening of action potential duration (Bailey et al., 1979). The effect reached a steady state within 3 minutes of drug exposure and could be reversed by a brief washout. After 5 minutes of exposure to isoproterenol, action potential durations again were measured. Acetylcholine (3 x 10^{-7} M) then was added for 5 minutes to the Tyrode's solution already containing isoproterenol, and the measurements of action potential duration then were repeated. This concentration of acetylcholine consistently produced significant prolongation of Purkinje fiber action potential duration in the presence of isoproterenol. We previously have reported that acetylcholine antagonizes the isoproterenol-induced shortening of action potential duration, and this effect of acetylcholine is blocked by atropine (Bailey et al., 1979). The prolongation of action potential duration caused by acetylcholine reached a steady state within 3 minutes and could be reversed rapidly (<3 minutes) by washout. Finally, after 5 minutes of exposure to the isoproterenol-acetylcholine combination, disopyramide, quinidine, procainamide, or atropine was added to the bath. The effects of these drugs in the presence of simultaneous β-adrenergic and muscarinic receptor stimulation were maximal within 3 minutes and could be reversed by a 5-minute washout period. After the preparation had been exposed to the isoproterenol-acetylcholine-drug combination for another 5 minutes, action potential duration again was measured.

Acetylcholine chloride, atropine sulfate, β-isoproterenol HCl, physostigmine sulfate, and quinidine sulfate were purchased from Sigma Chemical Company. Procainamide HCl was purchased from E.R. Squibb and Sons, Inc., and d,l-disopyramide phosphate was a gift from the G.D. Searle Company. Disopyramide was studied at concentrations of 1 x 10^{-6}, 7 x 10^{-6}, and 1.4 x 10^{-5} M; quinidine at concentrations of 1 x 10^{-6}, 1 x 10^{-5}, and 2 x 10^{-5} M; and procainamide at 2 x 10^{-5} M.

The electrophysiological data were analyzed by the paired Student's t-test for single drug interventions. Experiments performed with multiple drug interventions were analyzed by an analysis of variance to determine whether significant differences existed among groups of data, and the multiple t method then was applied to analyze differences within each group. All results are expressed as mean ± SD. The differences between the means were considered significant when P < 0.05 (Dixon and Massey, 1969).

**Results**

**Effects of Disopyramide, Quinidine, and Procainamide on Spontaneous Rate of Guinea Pig Atria**

The spontaneous rate of the isolated guinea pig right atrial preparation did not vary by more than 4% throughout the 15-minute control period which preceded the initiation of each experiment.

Spontaneous atrial rate was not altered by superfusion with atropine (1 x 10^{-6} M) (Fig. 1, panel A). In contrast, disopyramide (7 x 10^{-6} M), quinidine (1 x 10^{-5} M), and procainamide (2 x 10^{-5} M) superfused alone reduced spontaneous rate. In the experiments illustrated in Figure 1, these agents reduced rate by 11%, 13%, and 11%, respectively (Fig. 1, panels B-D) after 10 minutes of exposure to drug. In a series of such experiments (Table 1), changes in atrial rate were quantified after 5 minutes of exposure to drug. With prolonged exposure to antiarrhythmic drug (data not shown), slowing of atrial rate continued throughout a 30-minute period. This direct atrial rate slowing effect ob-

**Figure 1** Effects of atropine, disopyramide, quinidine, and procainamide alone on spontaneous cycle length of guinea pig right atria. Cycle length (msec) is plotted on the vertical axis, and time (minutes) is plotted on the horizontal axis. The initial portion of each tachometer recording represents the last 2 minutes of the 15-minute control period. The interruption of the tachometer recording after control is the time when drug infusion was initiated. A: Atropine 1 x 10^{-6} M; B: disopyramide 7 x 10^{-6} M; C: quinidine 1 x 10^{-5} M; D: procainamide 2 x 10^{-5} M. See text for discussion.
TABLE 1 The Effect of Antiarrhythmic Agents on Rate of Spontaneously Beating Guinea Pig Right Atria

<table>
<thead>
<tr>
<th>Control (n)</th>
<th>PS</th>
<th>PS/drug</th>
<th>Drug alone</th>
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<tbody>
<tr>
<td>223 ± 49 (12)</td>
<td></td>
<td></td>
<td>221 ± 57 (atropine 1 × 10⁻⁶ M)</td>
</tr>
<tr>
<td>264 ± 39 (9)</td>
<td></td>
<td></td>
<td>232 ± 32 (disopyramide 7 × 10⁻⁵ M)</td>
</tr>
<tr>
<td>209 ± 39 (9)</td>
<td></td>
<td></td>
<td>170 ± 47 (quinidine 1.4 × 10⁻⁵ M)</td>
</tr>
<tr>
<td>198 ± 34 (7)</td>
<td></td>
<td></td>
<td>168 ± 30 (quinidine 1 × 10⁻⁵ M)</td>
</tr>
<tr>
<td>233 ± 75 (12)</td>
<td></td>
<td></td>
<td>194 ± 57 (quinidine 2 × 10⁻⁵ M)</td>
</tr>
<tr>
<td>208 ± 69 (5)</td>
<td></td>
<td></td>
<td>184 ± 59 (procainamide 2 × 10⁻⁵ M)</td>
</tr>
</tbody>
</table>

Values = mean ± SD depolarizations per minute; n = number of guinea pig right atria studied; PS = physostigmine (1 × 10⁻⁶ M).
* Slowed compared to control after 5 minutes of drug exposure, P < 0.01.
† Slowed compared to control after 15 minutes of drug exposure, P < 0.005.
‡ Accelerated compared to rate observed at 15 minutes of PS, P < 0.05.

Served with disopyramide, quinidine, and procainamide following prolonged drug exposure was reversible only after a prolonged washout period (30 minutes).

Physostigmine (1 × 10⁻⁶ M) alone produced a gradual and continuous slowing of spontaneous rate of guinea pig right atria, as expected. In Figure 2, panel A, it increased spontaneous cycle length by 107% after 15 minutes and 126% after 20 minutes. Atropine (1 × 10⁻⁶ M) added to the superfusate already containing physostigmine antagonized the reduction in rate induced by physostigmine as expected (Fig. 2, panel B). Disopyramide (7 × 10⁻⁶ M) or quinidine (1 × 10⁻⁵ M) added to the superfusate already containing physostigmine also antagonized.

FIGURE 2 Effects of atropine, disopyramide, quinidine, and procainamide on spontaneous cycle length of guinea pig right atria pretreated with physostigmine. Cycle length (msec) is plotted on the vertical axis, and time (minutes) is plotted on the horizontal axis. The initial portion of each tachometer recording represents the last 1 minute of the 15-minute control period. The first interruption of the tachometer recording is the point at which physostigmine infusion was initiated, and the second interruption is the time point at which antiarrhythmic drug infusion was initiated. A: Physostigmine (1 × 10⁻⁶ M) superfused alone slows spontaneous rate; B: atropine (1 × 10⁻⁶ M) administered to an atrial preparation treated with physostigmine accelerates spontaneous rate; C: disopyramide (7 × 10⁻⁶ M) administered to a physostigmine-pretreated preparation accelerates spontaneous rate; D: quinidine (1 × 10⁻⁵ M) administered to a right atrial preparation which is treated with physostigmine elicits a positive chronotropic response; E: procainamide (2 × 10⁻⁵ M) administered to a right atrial preparation treated with physostigmine produces a negative chronotropic response, similar to the response observed with procainamide alone. See text for discussion.
the reduction in rate induced by physostigmine (Fig. 2, panels C and D). Thus, in the setting of muscarinic receptor activation, these antiarrhythmic agents produced an effect similar to that of atropine and opposite to that produced when the drugs were given alone. Procainamide did not modify the rate-slowing effect of physostigmine (Fig. 2, panel E). The anticholinergic effects observed with atropine, disopyramide, and quinidine reached maximum at 4 minutes and were rapidly reversible (data not shown). Summary data describing the effects of these drugs on spontaneous rate of guinea pig right atria are given in Table 1.

Effects of Disopyramide, Quinidine, and Procainamide on Action Potential Duration of Purkinje Fibers

Disopyramide, quinidine, and procainamide, when administered alone to isolated canine cardiac Purkinje fibers for 5 minutes, prolonged action potential duration by 5%, 12%, and 10%, respectively (Fig. 3). A 5-minute period was chosen because the atropine-like effects of these drugs (see below) reached steady state within 3–4 minutes, and our purpose was to compare the direct effects (action potential duration prolongation) to the atropine-like effects (action potential duration shortening) following identical periods of exposure. Prolonged exposure of fibers to each antiarrhythmic drug alone revealed that action potential duration prolongation continued throughout a 60-minute period (data not shown).

In a second series of experiments, each antiarrhythmic drug was administered to a Purkinje fiber under conditions of simultaneous β-adrenergic and muscarinic receptor stimulation to elucidate the atropine-like properties of these compounds. Isoproterenol shortened action potential duration of isolated cardiac Purkinje fibers as expected (Fig. 4B). Acetylcholine reversed this β-adrenergic receptor-mediated shortening of action potential duration (Fig. 4C). This effect of acetylcholine could be abolished by atropine (see below). Like atropine, disopyramide antagonized the acetylcholine-induced reversal of action potential duration shortening produced by isoproterenol (Fig. 4D). Quinidine had similar anticholinergic effects (Fig. 5, A–D). Thus, when administered together with isoproterenol and acetylcholine, both disopyramide and
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**Figure 6** Effects of procainamide on action potential duration of a Purkinje fiber preparation treated with isoproterenol and acetylcholine. A: Control; B: isoproterenol (1 x 10^-7 M) administered for 5 minutes; C: acetylcholine (3 x 10^-7 M) administered for 5 minutes in combination with isoproterenol; D: procainamide (2 x 10^-6 M) superfused for 5 minutes with continued treatment with isoproterenol and acetylcholine. Zero potentials and calibrations as in Figure 3. See text for discussion.

Quinidine antagonized the effects of acetylcholine. The effects of procainamide, however, were different. When this agent was added to acetylcholine and isoproterenol, action potential duration continued to lengthen (Fig. 6).

Summary data from a series of such experiments on Purkinje fibers are provided in Table 2. Disopyramide, quinidine, and procainamide, when given alone, all prolonged action potential duration. Disopyramide superfused for 5 minutes at concentrations of 1 x 10^-6 M, 7 x 10^-6 M, and 1.4 x 10^-5 M produced 1%, 7%, and 5% prolongation of total action potential duration, respectively. Linear regression analysis of control and post-drug exposure action potential durations demonstrated that the effects of disopyramide were not dependent on the control Purkinje fiber action potential duration (r = 0.21). Additional experiments (data not shown) performed with disopyramide at concentrations of 3 x 10^-6 M and 3 x 10^-5 M demonstrated 2% and 9% prolongation of action potential duration, respectively, thereby demonstrating that the direct effects of this compound were concentration-dependent over a narrow range. The concentrations 7 x 10^-6 M and 1.4 x 10^-5 M were chosen since they are comparable to concentrations attainable clinically (Winkle et al., 1975; Niarchos, 1976).

Quinidine administered alone at concentrations of 1 x 10^-6 M, 1 x 10^-5 M, and 2 x 10^-5 M produced 1%, 8%, and 13% prolongation of action potential duration, respectively. As with disopyramide, the duration of action potential in the control state was not a determinant of the response to quinidine (r = 0.39). Procainamide (2 x 10^-5 M) also prolonged action potential duration. Thus, when administered alone, quinidine, disopyramide, and procainamide prolonged action potential duration. However, when administered after pretreatment with isoproterenol and acetylcholine, disopyramide (7 x 10^-6 M) and quinidine (2 x 10^-5 M), but not procainamide (2 x 10^-5 M), induced shortening of action potential duration. In these latter experiments, isoproterenol was first given to shorten action potential duration, and then acetylcholine was administered. This muscarinic agonist reversed the action potential duration shortening induced by isoproterenol. When disopyramide and quinidine were added after acetylcholine, action potential duration was again reduced further, or action potential duration prolongation was at least, prevented, presumably because

**Table 2** The Effect of Antiarrhythmic Agents on Action Potential Duration of Canine Cardiac Purkinje Fibers

<table>
<thead>
<tr>
<th>Control (n)</th>
<th>ISO</th>
<th>ISO/ACh</th>
<th>ISO/ACh/drug</th>
<th>Drug alone</th>
</tr>
</thead>
<tbody>
<tr>
<td>367 ± 49 (5)</td>
<td>343 ± 16 (5)</td>
<td>313 ± 40 (5)</td>
<td>351 ± 27 (9)</td>
<td>373 ± 27 (6)</td>
</tr>
<tr>
<td>269 ± 30†</td>
<td>232 ± 33†</td>
<td>256 ± 32†</td>
<td>253 ± 27†</td>
<td>254 ± 30†</td>
</tr>
<tr>
<td>280 ± 34§ (atropine 1 x 10^-6 M)</td>
<td>343 ± 25§ (disopyramide 1 x 10^-6 M)</td>
<td>288 ± 25 (disopyramide 7 x 10^-6 M)</td>
<td>320 ± 17 (disopyramide 1.4 x 10^-5 M)</td>
<td>313 ± 18 (quinidine 1 x 10^-6 M)</td>
</tr>
</tbody>
</table>

Values = mean ± sd of total action potential duration in msec; n = number of Purkinje fibers studied; ISO = isoproterenol (1 x 10^-7 M); ACh = acetylcholine (3 x 10^-7 M).

* Prolonged from control after 5 minutes of drug alone, P < 0.01.
† Shortened from control after 5 minutes of ISO, P < 0.001.
‡ Prolonged from ISO after 5 minutes of ISO plus ACh, P < 0.01.
§ Shortened from ISO/ACh after 5 minutes of ISO/ACh plus drug, P < 0.05.
∥ Prolonged from ISO/ACh after 5 minutes of ISO/ACh plus drug, P < 0.05.
the action potential duration prolonging effects of acetylcholine were blocked. These effects of disopyramide and quinidine were similar to those observed with atropine.

Disopyramide exerted its greatest atropine-like effect at the lowest concentration studied (1 × 10⁻⁶ M). In contrast, quinidine produced its greatest atropine-like effect at the highest concentration studied (2 × 10⁻⁵ M). This atropine-like effect of quinidine was not observed at concentrations of 1 × 10⁻⁶ M and 1 × 10⁻⁵ M. Presumably, disopyramide and quinidine exerted direct effects as well as anticholinergic effects under these experimental conditions. At the higher concentrations (1.4 × 10⁻⁵ M), the direct effects of disopyramide predominated over the anticholinergic effects, whereas quinidine, at the higher concentrations (2 × 10⁻⁵ M), demonstrated greater anticholinergic effects than direct effects. In this experimental model, procainamide did not possess atropine-like effects and prolonged action potential duration in the presence or absence of cholinergic stimulation.

Discussion

The physiological properties of the heart are modulated by the two limbs of the autonomic nervous system. These two limbs generally exert opposite effects on cardiac function, the sympathetic limb producing excitatory effects and the cholinergic limb producing inhibitory effects. Drugs that modify the function of one or both limbs of the autonomic nervous system would be expected to exert effects that are dependent on the level of the autonomic tone regulating the heart when the drug is administered. The present study demonstrates that, under specific conditions in vitro, disopyramide and quinidine, two widely used antiarhythmic agents, produce qualitatively different electrophysiological effects on atria and Purkinje fibers, depending on whether or not the tissues are under the influence of muscarinic receptor activation.

When administered alone to isolated guinea pig right atria, disopyramide, quinidine, and procainamide all slowed spontaneous rate. This inhibitory effect on sinoatrial automaticity of these three agents has been observed previously (Vaughan Williams, 1958; West and Amory, 1960; Sekiya and Vaughan Williams, 1963; Nye and Roberts, 1966; Rosen and Hoffman, 1973). When these three agents were given to atrial tissues under the influence of increased muscarinic receptor stimulation, however, the electrophysiological effects of the drugs were no longer similar. Under this latter condition and at the specific duration of exposure studied, disopyramide and quinidine exerted positive chronotropic effects similar to those of atropine, whereas procainamide continued to produce a negative chronotropic effect as it did when administered alone.

In an early investigation by Mokler and Van-Arman (1962), disopyramide, when administered alone to conscious dogs at high doses (60–100 mg/kg, orally, or 5 mg/kg, intravenously), increased spontaneous heart rate. In the same study, these investigators observed that disopyramide (5 mg/kg, intravenously) abolished the negative chronotropic effects of vagal stimulation in an anesthetized dog. Furthermore, in a single isolated perfused rabbit heart experiment, they demonstrated that disopyramide (1 × 10⁻⁵ M) abolished the negative chronotropic effects of acetylcholine on atrial automaticity. This early study, therefore, suggested that high concentrations of disopyramide can exert anticholinergic effects in intact animals. The data from the present study extend these early observations concerning the anticholinergic properties of this agent. Disopyramide, administered in concentrations which are considered therapeutic (Winkle et al., 1975), antagonized the negative chronotropic effects of muscarinic receptor activation in isolated guinea pig atria. Furthermore, after the specific interval of drug exposure studied, this anticholinergic effect, which increased automaticity, predominated over the direct effect, which was to slow spontaneous atrial rate.

Quinidine also has been shown to produce anticholinergic effects in intact animals. James and Nadeau (1964) perfused quinidine (1 × 10⁻⁵ to 1 × 10⁻³ M) directly into the sinus node artery of intact anesthetized dogs and observed that it produced negative chronotropic effects when administered alone. These investigators also demonstrated that quinidine (1 × 10⁻⁴ to 1 × 10⁻³ M) prevented the negative chronotropic effects of vagal stimulation and acetylcholine administration. Thus, the data from the study by James and Nadeau suggested that quinidine possessed anticholinergic properties, although the concentrations of quinidine used (1 × 10⁻⁴ to 1 × 10⁻³ M) were well within the range considered to be toxic in intact animals. The present results in isolated guinea pig right atria confirm those earlier observations and also demonstrate that therapeutic concentrations (Winkle et al., 1975) of quinidine (1 × 10⁻⁴ M) can exert atropine-like effects on atrial automaticity. As with disopyramide, the anticholinergic effects of quinidine, which increased spontaneous rate, predominated over the direct rate slowing effects of this drug for the time that quinidine was administered simultaneously with physostigmine.

The anticholinergic effects of quinidine and disopyramide in atria were readily demonstrated because of the well-known direct negative chronotropic effects of muscarinic receptor stimulation in atria. By contrast, it is well known that activation of muscarinic receptors in quiescent ventricular tissues produces minimal or no direct inotropic (Levy et al., 1966; Dempsey and Cooper, 1969; Watanabe and Besch, 1976) and electrophysiological effects (Bailey et al., 1972; Gadabry et al., 1978; Danilo et
al., 1979). However, if muscarinic receptors are activated during simultaneous β-adrenergic receptor stimulation, a situation which is likely to occur physiologically, the inotropic effects of muscarinic receptor activation are pronounced (Watanabe and Besch, 1975). This phenomenon, which has been termed “accentuated antagonism” by Levy (1971), recently has been shown to be true also for electrophysiological effects of muscarinic activation (Bailey et al., 1979). In the setting of simultaneous activation of both β-adrenergic and muscarinic receptors in ventricular tissues, the effects of anticholinergic agents such as atropine are easily demonstrable, because these agents abolish the muscarinic-induced antagonism of β-adrenergic activation. In the present study, therefore, to elicit anticholinergic effects of antiarrhythmic agents on cardiac Purkinje fibers, the agents were given to fibers that were simultaneously treated with isoproterenol and acetylcholine. As we have demonstrated previously, acetylcholine antagonized the shortening of action potential duration induced by isoproterenol. This effect of acetylcholine was reversed by atropine. In fibers treated with isoproterenol and acetylcholine, disopyramide and quinidine also reversed the acetylcholine antagonism of the action potential duration shortening induced by isoproterenol. In other words, in the setting of simultaneous β-adrenergic and muscarinic receptor stimulation, these antiarrhythmic agents produced anticholinergic effects. When these same two antiarrhythmic agents were given alone to isolated cardiac Purkinje fibers in the absence of β-adrenergic and muscarinic receptor stimulation, they prolonged action potential duration. Procainamide prolonged action potential duration of isolated cardiac Purkinje fibers whether given alone or in the presence of β-adrenergic and muscarinic receptor stimulation. Thus, the electrophysiological effects of disopyramide and quinidine on cardiac Purkinje fibers depended on the degree and the nature of autonomic receptor stimulation present during drug administration.

Interaction between the sympathetic and cholinergic nervous systems in regulating cardiac function occurs at multiple levels in the intact animal, including the vasomotor centers of the central nervous system (e.g., via modulation of efferent sympathetic activity by inhibitory baroreceptor afferents), sympathetic nerve terminals wherein activation of presynaptic muscarinic receptors inhibit norepinephrine release, and postsynaptically at the level of β-adrenergic and muscarinic receptors (Levy, 1971; Higgins et al., 1973; Watanabe and Besch, 1975; Watanabe et al., 1978). This latter level of interaction can only be studied in an isolated tissue system free from influences by potentially interacting presynaptic sympathetic and vagal nerve fibers. The present results must, however, be extrapolated cautiously to intact animals and humans. Activation of autonomic receptors by exogenously administered neurotransmitters may not necessarily correspond physiologically to activation by the receptors in vivo by catecholamines and acetylcholine released from nerve endings. Moreover, the time course of the anticholinergic and direct effects of the agents studied was different. Whereas the anticholinergic effects of disopyramide and quinidine in atria and Purkinje fibers reach steady state within 3–5 minutes, the direct effects (slowing of spontaneous rate in atria and prolongation of action potential duration in Purkinje fibers) continued to increase progressively during 30 minutes of drug exposure. To dissect out the two components of effect of disopyramide and quinidine and for experimental reasons (detailed in methods), 5 minutes was chosen as the time to analyze and compare the data. The relevance of these electrophysiological findings, obtained after relatively brief exposure to the antiarrhythmic agents, to effects in intact animals and humans to whom the drug was administered chronically remains to be established. It is possible that, during chronic administration of the drugs to human subjects, the direct electrophysiological effects predominate. Nevertheless, the chronic extracardiac anticholinergic effects of these drugs are well recognized.

Current classifications of antiarrhythmic agents are based on the electrophysiological effects of these drugs on normal cardiac fibers. Disopyramide and quinidine are thought to exert their antiarrhythmic effects because of their common ability to reduce the maximum rate of depolarization and to prolong the refractory period of Purkinje fibers and ventricular muscle. However, previous studies have demonstrated that the electrophysiological effects of antiarrhythmic agents can be markedly altered by manipulation of numerous in vitro conditions. Some of these conditions include the extracellular potassium ion concentration (Armitage, 1957; Holland, 1957; Singh and Vaughan Williams, 1971; Chen et al., 1975; Danilo et al., 1977) and the presence of myocardial ischemia or infarction (Bassett et al., 1970; Allen et al., 1978; Cardinal and Sasyunik, 1978). Moreover, our data indicate that disopyramide and quinidine, prototypic group one antiarrhythmic drugs (Rosen and Hoffman, 1973), exert diametrically opposed electrophysiological effects on automaticity of guinea pig right atria and action potential duration of canine cardiac Purkinje fibers, depending on the degree of β-adrenergic and/or muscarinic cholinergic receptor stimulation. The present findings, together with the previous observations, indicate that the pharmacological properties of antiarrhythmic agents must be characterized in vitro under conditions mimicking physiological and pathological conditions in the intact animal.

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M J Mirro, A M Watanabe and J C Bailey

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