SUMMARY. Marked QT prolongation with induction of polymorphous ventricular tachycardia ("Torsades de Pointes") is a well-described phenomenon during quinidine therapy, frequently occurring at low plasma quinidine concentrations, low serum potassium, and slow heart rates. We have therefore assessed the dose-electrophysiological effects of quinidine as a function of extracellular potassium and cycle length in canine Purkinje fibers, using standard microelectrode techniques. Quinidine (1 μM) prolonged action potential at 90% repolarization, while leaving phase zero upstroke slope (V_{max}) unchanged at a cycle length 300–8000 msec; at 10 μM, V_{max} depression became evident. Increases in the action potential at 90% repolarization were most marked at long cycle lengths and low extracellular potassium (in contrast to V_{max} depression) and were partially reversed by tetrodotoxin (1 μM). The relationship between log of cycle length and action potential at 90% repolarization was linear (for cycle length 300–8000 msec) in the absence of quinidine. Quinidine increased the slope of this relationship in a concentration-related fashion, whereas increasing extracellular potassium shifted the curve rightward (without changing slope), regardless of the presence or absence of quinidine. Action potentials were also measured following pauses of 5–60 seconds. In the absence of quinidine, the action potential depolarization returned to its baseline value in a monoexponential fashion (time constant 36.0 ± 4.9 sec, mean ± SE, n = 10). In the presence of 1 μM quinidine, this return was better fit as a biexponential process (time constants 4.2 ± 1.2 and 40.7 ± 6.2 seconds, n = 14). At slow stimulation rates (cycle length greater than 4000 msec) in low extracellular potassium (2.7 mM), quinidine produced early afterdepolarizations in 7/14 (50%) of fibers at 1 μM and 14/18 (78%) at 10 μM. Early afterdepolarizations were eliminated by increasing stimulation rates, raising the extracellular potassium concentration to 5 mM, or adding tetrodotoxin. These data suggest that prolongation by quinidine of action potentials at 90% repolarization is multifactorial, with both a 'tonic' prolonging effect and a prominent frequency-dependent action potential shortening effect. At long cycle lengths and low extracellular potassium, low quinidine concentrations consistently produced early afterdepolarizations. The parallels between this form of triggered activity and clinical arrhythmias induced by quinidine suggest that early afterdepolarizations may play a role in quinidine-induced Torsades de Pointes. (Circ Res 56: 857–867, 1985)
window current described by Attwell et al. (1979). The selective sodium channel blocker tetrodotoxin produces similar effects, with shortening of action potential duration becoming apparent at concentrations lower than those producing decreases in $V_{\text{max}}$ (Coraboeuf et al., 1979).

Action potential prolongation occurs when canine Purkinje fibers are exposed to quinidine and ventricular muscle repolarization (as assessed by QT duration) is also prolonged in patients receiving quinidine. Increased [K$^+$], blunts this response in canine Purkinje fibers, whereas lowering [K$^+$] exag- gerates it (Wang and Parker, 1980). Colatsky (1982) attributed action potential prolongation by quinidine in rabbit Purkinje fibers to blockade of the delayed potassium rectifier (iK). However the frequency dependence of the relationship among quinidine, potassium, and action potential duration has not been explored.

Studies of the effects of quinidine in vitro have generally used concentrations $\geq 10 \mu M$ (3 $\mu g/ml$) which produce obvious changes in $V_{\text{max}}$ In fact, concentrations over 30 $\mu M$ frequently produce irreversible depolarization and inexcitability (Weld et al., 1980). However, with few exceptions (Wang and Parker, 1980; Mirro et al., 1981), the effects of much lower concentrations have not been well studied. We therefore undertook an evaluation of the effects of changes in [K$^+$], and cycle length on the electrophysiological actions of a range of quinidine concentrations in canine Purkinje fibers. We found that action potential prolongation was evident at quinidine concentrations lower than those depressing $V_{\text{max}}$. This action potential prolongation, unlike $V_{\text{max}}$, depression, was greatest at long cycle lengths and low [K$^+$]. Furthermore, at long cycle lengths and low [K$^+$], abnormal automaticity resulting from early afterdepolarizations (EAD) was consistently triggered in the presence of such low quinidine concentrations.

Methods

Mongrel dogs (15-20 kg) were anesthetized with intravenous sodium pentobarbital (30 mg/kg), and the heart was excised and immersed in cold (6-8°C) Tyrode's solution (gassed with O$_2$-CO$_2$, 95%-5%) containing (in mm) NaCl (137), NaHCO$_3$ (12), dextrose (5.5), MgCl$_2$ (0.5), Na$_2$HPO$_4$ (1.8), KCl (4), and CaCl$_2$ (2.7). False tendons were removed from either ventricle, and mounted in a tissue bath. They were superfused at 15 ml/min with warmed Tyrode's (containing 2.5 mm KCl) which has been gassed with 95% O$_2$-5% CO$_2$. The pH of the perfusate in the bath was 7.35 and the temperature was 37 ± 0.5°C. Temperature was monitored throughout each experiment with a YSI model 43 thermistor and a model 511 probe (1 mm in diameter). A valve interposed between Tyrode's reservoirs and heat exchanger allowed switching perfusates without disturbing the tissue bath. The dead space was approximately 7 ml (25 seconds).

A pair of Teflon-coated silver-silver chloride wires scraped bare at the ends was connected to a stimulator with isolated output for field stimulation of the tissue. Stimuli were 1-2 msec in duration and 1.5-2 times threshold. Purkinje fiber cells were impaled with glass microelectrodes selected for tip resistances of 10-30 MΩ and filled with 3 M KCl. The microelectrodes were connected through a silver-silver chloride junction to an operational amplifier with high input impedance and variable input capacitance neutralization. The tissue bath was grounded through a similar 3 M KCl-silver-silver chloride junction.

The signal from the operational amplifier was displayed on one channel of a Tektronix R556A oscilloscope and on a Gould paper recorder (model 2200). The signal from the operational amplifier and a sawtooth signal (200 V/sec) triggered from the oscilloscope were differentiated, and the differentiated signal was displayed on a second channel (with a different time base) of the oscilloscope. The differentiator had a linear response in the 100-1000 V/sec range. The output of the differentiator also provided input to a peak hold circuit, which recorded the maximum signal in a preset "window" (selected to encompass $V_{\text{max}}$). The peak hold output was recorded on the second channel of the paper recorder.

To record action potential characteristics, the oscilloscope was triggered by the stimulus timer (plus delay) and its screen photographed. The following parameters were derived from the photographs: takeoff potential, action potential amplitude (APA), overshoot, action potential duration at 50% of APA (APD$_{50}$) and at 90% of APA (APD$_{90}$), and, from the differentiated trace, $V_{\text{max}}$. A 100-mV signal was used to calibrate the amplifier-oscilloscope-recorder before each experiment. A sawtooth signal generator was used to confirm the linear output of the differentiator and to calibrate the peak hold output to the paper record prior to each experiment. The paper record was used to quantify changes in action potential duration (as described below), to detect spontaneous automatidty (whether normal or abnormal), and, using the peak hold circuit output, to follow changes in $V_{\text{max}}$ on a beat-to-beat basis.

After having been positioned in the bath, fibers were stimulated at a cycle length of 500-1000 msec. Following impalement, no measurements were made for at least 30 minutes. To study the cycle length and potassium dependence of quinidine's effects, the following approach was used. The tissue was stimulated at each of a range of cycle lengths, and action potentials were photographed after a stable configuration had been reached. With the protocol we used (serial doubling of cycle length), stable changes usually were achieved within 1 minute, although, at longer cycles, longer periods occasionally were required; this is consistent with reports of others (Colatsky and Hogan, 1980) who used somewhat different protocols. The cycle lengths used were (in order) 300, 500, 750, 1000, 2000, 4000, and 8000 msec. The perfusate then was switched to Tyrode's solution containing 2.7 mm KCl (without correction for osmolar changes), the tissue was stimulated at a cycle length of 500-1000 msec, and approximately 10 minutes was allowed to elapse (until the paper record showed the takeoff potential had stabilized at its more negative value). Action potentials were again photographed. The perfusate then was switched to Tyrode's with 5 mm KCl containing quinidine 0.1 μM. Action potentials were photographed after 30 minutes, the solution switched to Tyrode's with 2.7 mm KCl and quinidine 0.1 μM, and action potentials were rephotographed after 10 minutes (as above). The perfusate then was switched to Tyrode's with 5 mm KCl and 1.0 μM quinidine; the above sequence then was repeated for quinidine concentrations.
of 1.0 \mu M and 10 \mu M. Previous reports indicate that action potential characteristics could be expected to have reached near steady state values in 30 minutes (Mirro et al., 1981). As described below, dose-response information was analyzed only for impalements maintained in a single cell throughout the experiment. In some experiments, only the actions of a single concentration were examined (usually 1.0 or 10 \mu M), particularly to study in greater detail interventions affecting early afterdepolarizations (EAD). These included changes in cycle length and [K+]o, and the addition of tetrodotoxin or lidocaine.

The effects of pauses in stimulation on action potential duration were assessed in the following manner. Tissue was stimulated for at least 3 minutes at a cycle length of 1000 msec. The action potential was then recorded at fast paper speed (100 mm/sec), and the stimulator was then turned off for pauses of 5, 10, 30, or 60 seconds. Action potentials were recorded at fast paper speed for 4–8 seconds after the pause ended and then intermittently thereafter for the next 60–120 seconds. Since true action potential amplitude could not be measured from the paper recorder because of its poor frequency response characteristics, action potential durations were measured at takeoff plus 10 mV. Action potentials were excluded if, because of phase 4 depolarization, the takeoff potential had shifted more than 10 mV positive to the takeoff potential during stimulation at a cycle length of 1000 msec. Reinitiation of stimulation after a pause also precipitated EAD (particularly at 10 \mu M quinidine) in some fibers; these action potentials were also excluded from analysis. We recognize that this approach is not likely to detect changes with time constants under 1–2 seconds (e.g., in i_{la}).

Quinidine sulfate was purchased from Aldrich. Refrigerated stock solutions of 2 mg (base)/ml and 80 \mu g/ml were added to Tyrode’s solution to make up the final concentrations required (1 \mu M quinidine = 324 \mu g/liter). Similarly, tetrodotoxin (Sigma) was made up to a stock of 40 \mu M/ml (1 \mu M = 317 \mu g/liter).

Data Analysis

Two-way analysis of variance was used to analyze quinidine concentration-response data. If significant differences (P < 0.05) were detected, Duncan’s multiple range test was used to analyze paired data. Standard linear regression techniques were used to analyze relationships between cycle length and APD90 and between pause duration and post-pause APD. Student’s paired t-test was used to analyze differences between [K+]o = 2.7 mM and [K+]o = 5 mM at a given quinidine level or pause duration.

Post-pause action potentials were normalized to pre-pause baseline values. For pauses of 30–60 seconds, changes in action potential duration were sufficiently large to assess the time course of their return to baseline. Inspection of these time courses suggested that some were monoexponential and others appeared biexponential. Hence, the following approach was used to separate these possibilities. Each action potential duration vs. time plot was fitted to both mono- (2-parameter) and biexponential (4-parameter) declines using unweighted nonlinear regression (Sheiner, 1981). For each fit, the degrees of freedom (df = number of data points—number of parameters) and the residual sum of squares [RSS = \sum \text{(predicted—observed values)}^2] were calculated. The residual sum of squares of a more complex function (in this case, the biexponential fit) will always be smaller than the RSS of the simpler one. Recognizing this fact, the general linear test (Neter and Wasserman, 1974) allows selection between two models. The test statistic F with 2,df\text{bi} (degrees of freedom) was calculated:

\[ F = \frac{(\text{RSS}_{\text{mono}} - \text{RSS}_{\text{bi}})}{\frac{\text{df}_{\text{mono}} - \text{df}_{\text{bi}}}{\text{RSS}_{\text{bi}}}} \]

[note that \((\text{df}_{\text{mono}} - \text{df}_{\text{bi}}) = 2\). If the biexponential model was "much" better, then \((\text{RSS}_{\text{mono}} - \text{RSS}_{\text{bi}})\) was large, making F larger. If this F was associated with a P < 0.05, the biexponential model was deemed more appropriate.

Results

Action potentials recorded from the same cell at two quinidine concentrations and two cycle lengths (300 and 2000 msec) are presented in Figure 1. At 1 \mu M quinidine, APD90 was prolonged, particularly at the slower stimulation rate. In contrast, Vmax was unaffected at 1 \mu M quinidine and, at 10 \mu M quinidine, Vmax was depressed to a greater degree at the faster stimulation rate. Changes observed in experiments in which impalements were maintained in the same cell throughout (n = 8) are shown in Figure 2 at these cycle lengths. Statistically significant increases in APD90 were noted at quinidine concentrations as low as 1 \mu M, whereas no changes occurred in Vmax overshoot, APA, takeoff potential, or APD50 until

![Figure 1](http://circres.ahajournals.org/)

**Figure 1.** Action potentials recorded from a single Purkinje cell in the absence of drug (top row) in 1 \mu M quinidine (middle row), and in 10 \mu M quinidine (bottom row). Cycle lengths were 300 msec (left) and 2000 msec (right), and [K+]o was 2.7 mM. Calibrations for action potential measurement are shown. At the bottom of each photograph is the differentiated phase 0 trace, photographed at a faster sweep speed, offset on the time axis, and preceded by a 200 V/sec calibration.
10 μM quinidine was used. APD₉₀, unlike APD₅₀, in fact shortened with 10 μM quinidine. Furthermore, the changes in $V_{\text{max}}$, overshoot, APA, takeoff potential, and APD₅₀ were greatest at the more rapid stimulation rate, whereas APD₅₀ changes were greatest at the longer cycle length. With $[K^+]_o = 5$ mM, takeoff potential at a cycle length of 300 msec rose insignificantly from $-84.8 \pm 2.0$ mV at baseline to $-83.8 \pm 1.2$ mV in 10 μM quinidine, while $V_{\text{max}}$ fell from 570 ± 59 V/sec to 330 ± 32 V/sec, a 42% decrease. At a cycle length of 2000 msec, on the other hand, takeoff potential was similarly unchanged ($-84 \pm 1.9$ mV vs. $-84 \pm 1.1$ mV), but $V_{\text{max}}$ fell from 605 ± 72 V/sec to 482 ± 48 V/sec, a 20% drop. With $[K^+]_o$ lowered to 2.7 mM, statistically significant changes in takeoff potential were observed in 10 μM quinidine. The accompanying decreases in $V_{\text{max}}$, shown in Figure 2 may therefore represent both voltage- and drug-dependent effects. Our findings also suggest that the depolarization induced by raising $[K^+]_o$ was not sufficient to move into the steep portion of the takeoff potential-$V_{\text{max}}$ plot, and that therefore the marked $V_{\text{max}}$ decreases in this voltage range were due primarily to drug.

The relationship between cycle length and APD₉₀ in these eight experiments is further analyzed in Figure 3. For the sake of clarity only the means at each cycle length are presented. In low $[K^+]_o$ (2.7 mM), data are presented only for cycle length ≤2000 msec, since, at slower rates in the presence of quinidine, these fibers developed either normal automaticity or EAD. It can be seen that the log (cycle length) vs. APD plot provided good linearization of the relationship between the two parameters with correlation coefficients at baseline of 0.77 and 0.84. [The data could also be plotted as the hyperbolic function $(1/APD) = \text{slope} (1/\text{cycle length}) + \text{intercept}$ (Elharrar and Surawicz, 1983) with similar correlation coefficients (0.75 - 0.85). The asymptotic nature of the relationship is better appreciated in this way, but slope changes are more easily assessed by the log (cycle length) vs. APD linearization]. With increasing quinidine concentrations, linearity was maintained (except perhaps an upward deviation at very long cycle lengths in quinidine 10 μM), but the slopes increased. Regardless of $[K^+]_o$, the
slope at the baseline was 110–112 and rose to 242–246 at quinidine 10 μM. Both 1 and 10 μM quinidine produced statistically significant increases in slope. In contrast, the effect of raising [K+]o was to shift the curves rightward, without changing the slope, suggesting that the mechanism of the APD prolongation induced by lowering [K+]o, was not the same as that produced by adding quinidine.

Figure 4 shows the relationship between the duration of a pause in stimulation and the duration of the first action potential following the pause (normalized to baseline). There was no significant difference in normalized APD change between high [K+]o (5 mM) and low [K+]o (2.7 mM) at any pause duration, either in the drug-free state or in the presence of 1 μM quinidine. As is shown, there was an excellent correlation between pause duration and post-pause increase in normalized APD, in both the presence and absence of drug. However, a low concentration of quinidine markedly increased the slope of this relationship. Neither intercept differed significantly from zero. At longer pauses (≥ 30 sec), in the presence of 1 μM quinidine and low [K+]o (2.7 mM), resumption of stimulation initiated EAD in 3/5 experiments.

Figure 5 illustrates the method of data analysis used to assign the return of APD to baseline and to a mono- or biexponential fit after a 30- or 60-second pause. In 10/11 experiments in the absence of drug, a monoexponential fit was sufficient, whereas, in one instance, the biexponential curve afforded a better fit. In contrast, a biexponential fit was required in 14/19 experiments in the presence of 1 μM quinidine (P < 0.05, x²). The time constants for these fits are shown in Table 1.

As shown in Figure 2, Vmax was unchanged by 1 μM quinidine. However, as shown in Figure 6, the absence of quinidine markedly depressed Vmax at rapid rates (42% at cycle length 300 msec), whereas, at cycle lengths of 4000–8000 msec, Vmax depression was less (13–14%).
was decreased from 8000–1000 msec in the presence of 10 μM quinidine, \( V_{\text{max}} \) declined to its new value 
(Fig. 6, inset) with a mean time constant of 4.6 ± 0.4 sec (n = 12), approximately the same as the "rapid component" of decline in normalized APD in the presence of quinidine in Table 1.

With low \([K^+]_o\) (2.7 mM), stimulation at long cycle lengths (>4000 msec) or normal automaticity at similar low rates frequently caused EAD (Fig. 7), the first of which arose during late phase 2/early phase 3 from a takeoff potential of −40 to −55 mV. The incidence of EAD under these conditions is shown in Figure 8. At 1 μM quinidine, multiple EAD were present in half of the experiments and, in 10 μM quinidine, in 14/18. An example of an EAD arising during phase 3 at a cycle length of 8000 msec is shown in Figure 9.

EAD were readily abolished by a number of interventions. Resumption of more rapid rates of stimulation was effective in each experiment. Raising \([K^+]_o\) to 5 mM was tried in 15 experiments and was always effective (upper panel, Fig. 10). Tetrodotoxin (TTX, middle panel, Fig. 10) was used in eight experiments: 0.1 μM abolished EAD in 1/6 attempts, and 1.0 μM did so in 7/7 attempts. Lidocaine (10 μM) also abolished EAD in 2/2 attempts (lower panel, Fig. 10). The changes in \( V_{\text{max}} \) and APD in the presence of quinidine in Table 1.

FIGURE 6. Relations between maximal phase 0 upstroke slope (\( V_{\text{max}} \)) and cycle length (n = 8 at each point). 
Inset: changes in \( V_{\text{max}} \) (top tracing) when interstimulus cycle length was changed from 8000 to 1000 msec. \( V_{\text{max}} \) declined exponentially to its new value with a time constant of 2.3 seconds in this experiment.

FIGURE 7. Early afterdepolarizations in four different preparations in low \([K^+]_o\) and 10 μM quinidine at slow rates of depolarization (either spontaneous, panels A and B, or stimulated at a cycle length of 8000 msec, panels C and D).
TABLE 1

<table>
<thead>
<tr>
<th></th>
<th>Monoexponential</th>
<th>Biexponential</th>
</tr>
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<tbody>
<tr>
<td>No drug</td>
<td>36.0 ± 4.9</td>
<td>0.91</td>
</tr>
<tr>
<td></td>
<td>( n = 10 )</td>
<td>( n = 1 )</td>
</tr>
<tr>
<td>Quinidine (100 μM)</td>
<td>22.3 ± 2.1</td>
<td>4.2 ± 1.2</td>
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<tr>
<td></td>
<td>( n = 5 )</td>
<td>( n = 14 )</td>
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*Data in Table 1 represent time constants for the return of APD to baseline.*

In contrast to changes shown in Figure 10, an interval of several minutes at the new conditions was required before the more negative resting potential was abruptly restored.

**Discussion**

We have shown that a low concentration of quinidine prolonged action potentials of canine Purkinje fibers and consistently triggered abnormal depolarizations in low \([K^+]_o\) at slow stimulation rates. A number of lines of evidence suggest that this action potential prolongation reflects a non-sodium channel-mediated effect, although, as outlined below, sodium channel blockade may well modulate these action potential changes. First, selective sodium channel blockade (by TTX) shortens APD (Coraboeuf et al., 1979). Second, the effects on APD that we observed were most prominent at slow stimulation rates whereas \(V_{max}\) changes were most evident with rapid stimulation. Third, Colatsky (1982) has shown that quinidine abolished \(i_x\), current tails in rabbit Purkinje fibers while it prolonged the action potential. Fourth, Nattel and Bailey (1983) have examined the time courses of quinidine-induced changes in phase 0, 2, and 3 slopes; they concluded that phase 3 slope changes were due to a mechanism different from that producing phase 0 and 2 changes. Our action potential findings complement those of other workers. Mirro et al. (1981) examined quinidine concentration-response relations in canine Purkinje fibers driven at a cycle length of 1000 msec in \([K^+]_o = 4 \text{ mM}\). They found 1 μM to be without effect, but 3 μM prolonged \(APD_{90}\) and depressed \(V_{max}\). On the other hand, Wang and Parker (1980) found that \(V_{max}\) depression and \(APD_{90}\) shortening were the most prominent effects of quinidine up to concentrations of 20 μM without significant \(APD_{95}\) changes when fibers were driven at a cycle length of 800 msec in \([K^+]_o = 4 \text{ mM}\); lowering \([K^+]_o\) to 2 mM prolonged \(APD_{95}\) in 20 μM quinidine. \(APD_{90}\) changes were not significant in the study of Mirro et al. (1981), but the trend toward shortening at higher concentrations reported by Wang and Parker (1980) was found in our data. As discussed below, this change may reflect blockade of the window current.

The observation that the action potential-prolonging effect of quinidine was evident at a lower concentration than that depressing \(V_{max}\) has several potential implications. First, depression of \(V_{max}\) and resultant conduction slowing has been proposed as the major mechanism of quinidine's antiarrhythmic effect (Vaughan Williams, 1958; Wallace et al., 1966). However, action potential prolongation itself may be antiarrhythmic by decreasing dispersion of repolarization, i.e., by producing disproportionately greater increases in short action potentials. Whereas data in this study were collected from mid-false tendon, others have reported that quinidine exerts disproportionately greater effects on action potential in "post-gate" fibers, thereby reducing dispersion of repolarization (Wang and Parker, 1980). Hence, low concentrations of quinidine might be antiarrhythmic by such an effect on action potential duration without depressing \(V_{max}\). Second, this action (action potential prolongation without changes in \(V_{max}\)) has been held to be a "class III" (amiodarone-like) effect (Vaughan Williams, 1975). The demonstration of APD and \(V_{max}\) effects with different dose-dependencies in this study underlines the difficulties with such classification schemes. Extrapolation from changes in parameters such as action potential duration or \(V_{max}\) to antiarrhythmic effects in vivo is not...
A. INCREASE $K_0$ TO 5 mM

B. TETRODOTOXIN 1 $\mu$M

C. LIDOCAINE 10 $\mu$M (2.1 $\mu$g/ml)

FIGURE 10. Interventions altering early afterdepolarizations. In each experiment, the intervention was made at the beginning of the record (the time from intervention change to tissue bath being 25 seconds). As can be seen, raising $[K^+]_o$ or adding tetrodotoxin or lidocaine abolished EAD, and they recurred when $[K^+]_o$ was changed back to 2.7 mM (top).

justified, as no data are available to link quinidine's antiarrhythmic effects in man to such a "class III" action.

FIGURE 11. Effects of adding tetrodotoxin (TTX) in the presence of quinidine on maximal phase 0 upstroke slope ($V_{\text{max}}$, top panel) and action potential duration at 90% repolarization ($\text{APD}_{90}$, bottom) as a function of a cycle length ($n = 8$). The slope of the cycle length vs. $\text{APD}_{90}$ plots decreased ($P < 0.05$) from 283 ± 35 ($r = 0.81$, $P < 0.001$) to 184 ± 26 ($r = 0.77$, $P < 0.001$) with the addition of TTX, while $V_{\text{max}}$ was slightly depressed but in a cycle length-independent fashion. See text for concentrations.

We did not measure membrane ionic currents in this study. However, our findings do suggest an explanation for the APD changes induced by quinidine which is consistent with other reports. The observation that APD in Purkinje fibers lengthens after a pause and shortens exponentially thereafter has been made previously (Greenspan et al., 1967; Vick, 1971; Miller et al., 1971). The factors responsible are not completely understood. One possibility is activation of the Na$^+$-K$^+$ pump; pump current has been measured in small canine Purkinje fibers following pauses in stimulation, and its magnitude after resumption of stimulation followed an exponential decline with a fairly long (1.3 minute) time constant (Gadsby and Cranefield, 1979). Hence, with a long pause, the pump current would decline to a low value and the first action potential after such a pause would be correspondingly prolonged. The rate of return of APD to baseline would similarly be a function of pause duration. Other phenomena which might contribute to such APD changes include slow inactivation of fast channels (Gintant et al., 1984) or changes in $i_{\text{to}}$, which have been implicated as the major determinant of the duration of premature action potentials (Hauswirth et al., 1972). Changes in the slow inward current might also contribute, although Colatsky and Hogan (1980) suggested that the long time course of return to baseline was not mediated by changes in $i_{\text{to}}$.

Quinidine (1 $\mu$M) produced a characteristic alteration in the time course of APD changes following a pause: the long time constant was unaltered, but a more rapid component was superimposed. The time constant for this rapid process was similar to that for the development of $V_{\text{max}}$ depression in 10
μM quinidine. This finding would be consistent with cycle length-dependent blockade of sodium channels by quinidine, not only during phase 0, but also during plateau when it would accelerate repolarization. Although data obtained in ovine Purkinje fibers (Carmeliet and Saikawa, 1982) suggest that quinidine does exert this action, we cannot be certain that the rapid phase of action potential shortening we observed actually represents this fast channel effect. For example, it is likely that the time course of onset of use-dependent fast channel blockade is concentration-dependent (Gintant et al., 1983) and that, at least under some conditions, $V_{\text{max}}$ may not be linearly related to sodium conductance (Bean et al., 1982). Hence the correlation we found between time constants may be fortuitous; without more detailed measurements, we cannot offer alternative explanations for the change induced by quinidine in the time course of APD changes after a pause.

EAD were seen when action potential was markedly prolonged, i.e., when, in the presence of quinidine, stimulation rate was slowed and [K⁺], lowered. The recordings shown in Figure 12 suggest that, under these circumstances, changes in repolarizing currents were sufficient to enable the development of a stable, depolarized level of resting potential (Gadsby and Cranefield, 1977). These changes would include, as described above, a decrease in $i_x$, induced by quinidine, a decrease in $i_{K_1}$ induced by lowering [K⁺], and possibly, a decrease in the pump current due to a reduction in stimulation rate. The use-dependent nature of quinidine's fast inward current-blocking properties would permit relative preservation of this depolarizing current at slow stimulation rates. Increasing stimulation rate would, by increasing pump current and decreasing fast inward current, reverse the tendency to develop such a stable depolarized level of resting potential. In keeping with this concept, interventions which increased outward current (raising [K⁺]) or decreased inward current (adding TTX) restored a normal level of resting potential. A low level of resting potential can itself result in oscillatory activity (Hauswirth et al., 1969). Hence, quinidine-induced EAD might simply be a reflection of the marked APD increase seen in these experiments, although certain interventions (e.g., tetraethylammonium, Ito and Surawicz, 1981) can markedly prolong APD without being associated with EAD. This oscillatory activity at a low level of resting potential has been attributed to time-dependent changes in $i_x$, accompanied by small increases in background inward current (Hauswirth et al., 1969; Imanishi and Surawicz, 1976) or to increased sodium conductivity (Brown, 1981). Coraboeuf et al. (1980) reported abnormalities in the terminal phase of repolarization in false tendon Purkinje fibers exposed to low [K⁺], and acidosis. Such abnormalities, which, if sufficiently prominent, triggered EAD, were attributed to a decrease in background outward K⁺ currents. In an accompanying computer simulation, the same workers (Coulombe et al., 1980) suggested that such repolarization abnormalities were most sensitive to small changes in time-independent outward K⁺ currents, moderately sensitive to changes in $i_{K_1}$, and insensitive to changes in $i_x$. The computer simulation also suggested that the window current exerted an important facilitatory influence upon the development of EAD. This group of workers did not report APD itself but the data presented suggests that EAD developed when APD (measured at 90% repolarization) prolonged due to terminal phase 3 abnormalities. They also noted that shortening cycle length and raising [K⁺], also abolished EAD, and raised the possibility that these abnormalities could account for arrhythmic activity in the prolonged QT syndrome. We cannot conclude that quinidine-induced EAD and acidosis-induced EAD are due to the same underlying membrane derangement. However, our findings are consistent with the suggestion that the
presence of an inward sodium current at long cycle lengths and reduction of outward potassium current(s) are important contributing factors to the generation of this rhythm abnormality.

Brachman et al. (1983) have reported that cesium administration to dogs triggered a bradycardia-dependent polymorphous ventricular tachycardia very closely resembling Torsades de Pointes. Since they and others (Damiano and Rosen, 1983) showed that cesium precipitated EAD at slow stimulation rates in canine Purkinje fibers, they suggested that EAD triggered Torsades de Pointes. They also showed that both the EAD and the in vivo arrhythmia were abolished by low doses of tetrodotoxin, suggesting that an "unblocked" window current facilitated the development of this rhythm disturbance. In contrast to our findings, cesium precipitated EAD at "usual" [K+]o (4 mM); this difference may reflect the fact that cesium is thought to block iK1, as well as pacemaker and pump currents (Isenberg, 1976), while leaving i, unaffected. Cycle-length dependent EAD have also been reported after N-acetylprocainamide superfusion of canine Purkinje fibers (Dangman and Hoffman, 1981). It is of interest that this agent, the major metabolite of procainamide, did not depress V_{max} in vitro (Dangman and Hoffman, 1981) or conduction velocity in vivo (in dogs or patients) (Jaillon and Winkle, 1979; Jaillon et al., 1981). Furthermore, it can cause bradycardia-dependent polymorphic ventricular tachyarrhythmias in dogs (Dangman and Hoffman, 1981) and has been implicated in the development of Torsades de Pointes (Olshansky et al., 1982). Quinidine-induced EAD are triggered and reversed by the same alterations that can trigger and reverse Torsades de Pointes in patients (changes in [K+]o, cycle length). Hence, it is tempting to speculate that "idosyncratic" arrhythmia induction by low plasma concentrations of quinidine is in fact a reflection of predictable changes modulated in an individual by cleft K+, heart rate, and other factors such as autonomic tone (Malliani et al., 1980). The role of EAD in triggering these distinctive clinical arrhythmias requires further studies in intact hearts.

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INDEX TERMS: Quinidine • Action potential duration • Early afterdepolarizations • Arrhythmogenesis
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