Enhancement of Triggered Activity in Ischemic Purkinje Fibers by Ouabain: A Mechanism of Increased Susceptibility to Digitalis Toxicity in Myocardial Infarction

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Enhanced susceptibility to toxic arrhythmias by digitalis administration has been reported in clinical and experimental myocardial infarction. To investigate the mechanism responsible for this phenomenon, the effects of superfusion with normal Tyrode's solution and superfusion with Tyrode's solution containing \(4 \times 10^{-8} M\) of ouabain in ischemic Purkinje fibers were compared. Ischemic Purkinje fibers of small endocardial preparations from 1 day old myocardial infarcts in 18 dogs were used for the study. During control conditions, these endocardial preparations demonstrated delayed afterdepolarizations and triggered activity. Superfusion with normal Tyrode's solution resulted in a gradual increase in maximal diastolic potential and action potential amplitude, a decrease in delayed afterdepolarization amplitude and slowing and termination of triggered activity.

Superfusion for 90 minutes with Tyrode's solution containing ouabain resulted in: 1) an increase in the magnitude of delayed afterdepolarizations in preparations demonstrating subthreshold delayed afterdepolarizations, 2) sustention of triggered activity in preparations showing nonsustained triggered activity, and 3) shortening of cycle lengths of the triggered activity in preparations demonstrating sustained triggered activity before superfusion with ouabain. These effects occurred despite the gradual increase in maximal diastolic potential and action potential amplitude. Superfusion of normal Purkinje fibers with Tyrode's solution containing \(4 \times 10^{-8} M\) of ouabain for 90 minutes did not result in delayed afterdepolarizations or triggered activity. Thus, ouabain at a concentration that has no toxic effect on normal Purkinje fibers may enhance arrhythmias in ischemic Purkinje fibers by increasing the magnitude of delayed afterdepolarizations and enhancing triggered activity.

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Increased susceptibility to the toxic effects of digitalis in recent or acute myocardial infarction remains controversial (1). In a study (2) of patients with acute myocardial infarction, only 6% of the patients receiving a full dose of acetylstrophanthinid developed ventricular ectopic beats. A double-blind study (3) in patients with acute myocardial infarction also showed no difference in the incidence of rhythm disturbances between digoxin-treated and control patients. However, previous experimental studies (4–8) found that animals with myocardial infarction tolerated digitalis poorly. A number of clinical studies (9–11) also demonstrated the increased incidence of ventricular arrhythmias and mortality in patients with myocardial infarction who received digitalis.

The mechanism responsible for the increased susceptibility to digitalis remains unclear. After stabilization in normal Tyrode's solution for more than 2 hours, ischemic Purkinje fibers showed oscillatory potentials as a result of ouabain superfusion at subtoxic concentrations (12). Recently, it was found (13) that triggered activity arising from delayed afterdepolarizations in the ischemic Purkinje fibers was an important mechanism of arrhythmias in the early phase of experimental myocardial infarction. It was also noted that the delayed afterdepolarization and triggered activity occurred within minutes after placement of the endocardial preparations in the tissue bath and could not be elicited after the ischemic fibers had been superfused with normal Tyrode's solution for more than 2 hours.

The purpose of this study was to evaluate the effects of ouabain at a concentration that did not induce arrhythmia...
in normal Purkinje fibers on such delayed afterdepolarizations and triggered activity seen in ischemic Purkinje fibers. The results may elucidate one possible mechanism responsible for the increased susceptibility to digitalis toxicity in patients with previous myocardial infarction.

Methods

Experimental preparations. Small endocardial preparations (up to 20 × 20 × 5 mm in dimension) from 1 day old myocardial infarcts in 18 dogs were studied using standard microelectrode techniques. Twenty to 26 hours after left anterior descending coronary artery ligation performed under pentobarbital (30 mg/kg) anesthesia, dogs were reanesthetized using the same drug and the heart was removed. Small preparations of endocardial tissue overlying the infarcted area were sliced and mounted in the superfusion chamber. Border zones were excluded from the experimental analysis. The ischemic preparations from those hearts were randomized into two groups, one group studied during 90 minutes of superfusion with normal Tyrode’s solution and another group during 90 minutes of superfusion with normal Tyrode’s solution containing ouabain at 4 × 10⁻⁸M concentration. The Tyrode’s solution contained 4.0 mM of potassium chloride and 2.7 mM of calcium chloride. The concentrations of other substances are similar to that of a previous study (13). The pH was 7.2 to 7.4 as measured by Radiometer ABL 1 Acid-Base Laboratory. The solutions and preparations were gassed with a mixture of 95% oxygen and 5% carbon dioxide.

We also studied the effect of superfusion with Tyrode’s solution containing ouabain at a concentration of 4 × 10⁻⁸M on normal Purkinje fibers taken from the normal endocardial surface of both the left and right ventricles of the same hearts to determine whether delayed afterdepolarizations and triggered activity could be induced during and after 90 minutes of superfusion.

Protocol. All preparations were studied at 36°C temperature and driven by rectangular pulses of 2 ms duration and at twice diastolic threshold delivered by a Bloom DTU programmable stimulator through bipolar platinum electrodes (Rhodes). Six to 10 stimuli at decreasing cycle lengths from 1,500 to 600 ms were used to induce delayed afterdepolarizations and triggered activity (13). Initially, the preparation was stimulated at a cycle length of 1,500 ms for six beats. If no triggered activity was induced, the number of beats was gradually increased until 10 beats were delivered. If no triggered activity was induced, the stimulation cycle length was decreased by 100 ms and the series from 6 to 10 beats was repeated. This stimulation protocol was continued until triggered activity was induced or a cycle length of 600 ms and a series of 10 beats were achieved. Between trains of stimuli, the endocardial preparations that did not show spontaneous electrical activity were driven at a cycle length of 1,500 ms. Transmembrane potentials were measured using glass capillary microelectrodes filled with 3 M of potassium chloride and having a resistance of 10 to 30 MΩ. Each microelectrode was coupled to a W.P. Instrument model M-707 microprobe system and the potentials were displayed on a Tektronix 5111 storage oscilloscope and recorded using a Gould-Brush 220 chart recorder. Details of the recording technique were reported previously (13).

Definition of terms. The terms used in this study conform with the terminology suggested by Cranefield (14). If after repolarization, the membrane potential reaches a level negative to the level before the upstroke, an early hyperpolarization occurs. An early hyperpolarization may terminate in a return to the level of membrane potential prevailing before the action potential or may lead into a delayed afterdepolarization. A delayed afterdepolarization occurs when the membrane potential, after complete repolarization, returns to a level positive to the level of membrane potential before the action potential.

Data analysis. Data are presented as means ± standard deviation. Analysis of variance with repeated measures were used to analyze the data at various times of superfusion with normal Tyrode’s solution and ouabain. When significant differences were detected among the groups, Scheffé’s multiple comparison test was applied to compare individual mean values (15). Student’s paired t test was used to compare the action potential duration and amplitude and slope of delayed afterdepolarization before and after ouabain superfusion. All differences were considered significant when the probability level was less than 0.05.

Results

Normal Purkinje Fibers

Superfusion of 10 preparations of normal Purkinje fibers with Tyrode’s solution containing 4 × 10⁻⁸M of ouabain for 90 minutes resulted in no delayed afterdepolarizations or triggered activity. Figure 1 shows action potentials recorded from a normal Purkinje fiber stimulated at a cycle length of 1,000 ms for 10 consecutive beats. No delayed afterdepolarization or triggered activity was seen after the stimulated beats before and after superfusion with ouabain for 90 minutes. It can also be seen that ouabain superfusion shortened action potential duration at 50% repolarization from 270 to 250 ms and at 90% repolarization from 390 to 360 ms. However, when the action potential durations at 50% repolarization and at 90% repolarization before and after ouabain superfusion were compared, no statistical difference was found (action potential duration at 50% repolarization before ouabain 277.0 ± 18.9 ms, after ouabain 273.0 ± 21.1 ms; action potential duration at 90% repolarization before ouabain 389.0 ± 38.7 ms, after ouabain
Figure 1. Effect of 90 minutes of superfusion of Tyrode’s solution containing $4 \times 10^{-8} \text{M}$ of ouabain on normal Purkinje fibers. Panel A is control, panel B is after 90 minutes’ ouabain superfusion. Ouabain shortened action potential duration (inserts) during stimulation at a cycle length of 1,000 ms when compared with control. No delayed afterdepolarizations or triggered activity can be seen after ouabain superfusion. S = stimulus artifact; TL = time lines.

379.0 $\pm$ 42.5 ms). Similarly, ouabain superfusion did not significantly change membrane potentials at rest (before ouabain $-86.3 \pm 3.9$ mV, after ouabain $-85.7 \pm 3.2$ mV).

Ischemic Purkinje Fibers

Superfusion with normal Tyrode’s solution. The depressed cells in the endocardial preparations from the infarcted area showed gradual recovery during superfusion (16). The process of recovery was characterized by an increase in the resting potential, action potential amplitude and maximal rate of rise of phase 0.

Delayed afterdepolarization and triggered activity. Preparations that showed delayed afterdepolarizations or triggered activity immediately after impalement maintained these characteristics for a period of at least 45 minutes. During this period, either sustained activity (activity lasting more than 1 minute), separate runs (each lasting less than 1 minute) or single triggered beats were observed. The rate of triggered activity gradually decreased as the amplitude of the maximal diastolic potential increased. At membrane potentials more negative than $-80$ mV, triggered activity could not usually be induced. Twelve of 29 preparations obtained from the endocardial side of the infarcted area for this part of the study showed a delayed afterdepolarization that achieved threshold potential, resulting in sustained triggered activity when stimulated at cycle lengths of 600 to 1,500 ms and for 6 to 10 beats. Those preparations showing sustained triggered activity were subjected to 90 minutes of superfusion with normal Tyrode’s solution. The remaining preparations showed only nonsustained triggered activity lasting less than 1 minute, subthreshold afterdepolarization or no delayed afterdepolarization during superfusion with normal Tyrode’s solution for 15 to 30 minutes. Six of the preparations demonstrating nonsustained triggered activity and subthreshold depolarization were then subjected to ouabain superfusion, the results of which will be discussed later. Eleven other endocardial preparations that did not show delayed afterdepolarizations or triggered activity became quiescent within 45 minutes of superfusion with normal Tyrode’s solution.

Figure 2 shows an example of the time course of sustained triggered activity during continuous superfusion with Tyrode’s solution. Twenty-five minutes after placement of the preparation in the tissue bath, automatic action potentials were followed by a prominent early afterhyperpolarization and a small but distinct delayed afterdepolarization (Fig. 2A). The fourth automatic action potential triggered a fast rhythmic activity at a cycle length of 600 ms. A single action potential recorded at an expanded time scale illustrates a maximal diastolic potential of $-48$ mV and a steep slope of the delayed afterdepolarization (Fig. 2A, right). Thirty minutes after initiation of triggered activity, the amplitude of the maximal diastolic potential and action potential amplitude increased (Fig. 2B). This was associated with an increase in the cycle length of the rhythmic activity and a decrease in the slope of the delayed afterdepolarization (Fig. 2B, right). Sixty minutes later spontaneous termination of the rhythmic activity occurred, after the cycle length of

Figure 2. Time course of sustained triggered activity in ischemic Purkinje fibers superfused with normal Tyrode’s solution. Action potentials recorded to the right of each panel show gradual decrease in the slope of delayed afterdepolarizations. See text for details. T = time line.
the rhythmic activity had increased to 2,500 ms and the slope of the diastolic potential had markedly decreased or completely disappeared (action potential in Fig. 2C (right) and during cessation of triggered activity). The absence of delayed afterdepolarization recorded when the rhythmic activity ceased can be explained by either a marked diminution of delayed afterdepolarization or a shift in the location of the cell with dominant triggered activity. The termination of the triggered rhythm was associated with marked improvement of maximal diastolic potential (−86 mV) and action potential amplitude (−98 mV).

The effects of 45 and 90 minute superfusion with normal Tyrode's solution on 12 ischemic subendocardial preparations showing sustained triggered activity are tabulated in Table 1. Maximal diastolic potentials, action potential amplitudes and cycle lengths of the triggered activity increased significantly during superfusion with normal Tyrode's solution. In addition, after 45 and 90 minutes of superfusion, one and five preparations, respectively, lost their sustained triggered activity despite stimulation at various cycle lengths and for up to 10 beats.

**Superfusion with Tyrode's solution containing 4 × 10⁻⁸ M of ouabain.** We evaluated the effects of superfusion with Tyrode's solution containing ouabain on two groups of ischemic preparations.

1) **Preparations that showed delayed afterdepolarizations but no sustained triggered activity before ouabain superfusion.** Six such preparations showing these characteristics during superfusion with normal Tyrode's solution for 15 to 30 minutes were superfused with ouabain. Ouabain superfusion gradually increased the amplitude of the delayed afterdepolarizations. This gradual increase in delayed afterdepolarizations was followed by the appearance of sustained triggered activity. In two preparations that showed only nonsustained triggered activity (triggered activity lasting less than 1 minute), ouabain superfusion also resulted in the development of sustained triggered activity. Figure 3 shows an example of the effect of ouabain superfusion on subthreshold delayed afterdepolarizations. During superfusion with normal Tyrode's solution (Fig. 3A), membrane potential at rest was −44 mV, and stimulated beats at a cycle length of 1,500 ms were followed by a small delayed afterdepolarization of about 6 mV in magnitude. After superfusion with ouabain for 25 minutes (Fig. 3B), the magnitude of the delayed afterdepolarization increased to about 10 mV. At the same time, the membrane potential at rest increased from −44 mV during control to −50 mV; this was not caused by ouabain superfusion, but by the natural time course of improvement in superfused ischemic fibers. An increase in maximal diastolic potential and action potential amplitude was also observed at this time. Further superfusion (Fig. 3C) resulted in larger delayed afterde-

Table 1. Effects of 45 and 90 Minutes of Superfusion With Normal Tyrode’s Solution on Sustained Triggered Activity in Ischemic Purkinje Fibers

<table>
<thead>
<tr>
<th></th>
<th>Control (n = 12)</th>
<th>Duration of Superfusion From Initiation of Triggered Activity</th>
<th>45 Minutes</th>
<th>90 Minutes</th>
<th>90 Minutes</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>n</td>
<td>p₁</td>
<td>n</td>
<td>p₁</td>
</tr>
<tr>
<td>Maximal diastolic potential (mV)</td>
<td>54.3 ± 5.2</td>
<td>67.9 ± 6.9</td>
<td>12</td>
<td>&lt;0.01</td>
<td>77.7 ± 4.1</td>
</tr>
<tr>
<td>Action potential amplitude (mV)</td>
<td>55.5 ± 6.7</td>
<td>74.3 ± 8.0</td>
<td>12</td>
<td>&lt;0.01</td>
<td>89.3 ± 8.0</td>
</tr>
<tr>
<td>Cycle length (ms)</td>
<td>756.7 ± 109.0</td>
<td>962.7 ± 138.1</td>
<td>11*</td>
<td>&lt;0.01</td>
<td>1,225.0 ± 310.6</td>
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</tbody>
</table>

*One preparation lost triggered activity. †Five preparations lost triggered activity. n = number of preparations; NS = not significant; p₁ = compared with control; p₂ = compared with 45 minutes of superfusion.
polarization and the development of sustained triggered activity, since the magnitude of the delayed afterdepolarization after seven consecutive stimulated beats was sufficient to induce regenerative action potentials. As a result of these regenerative potentials, the last two stimuli occurred during the refractory period of the first two triggered beats.

In the six preparations studied, ouabain superfusion for 26.8 ± 4.5 minutes resulted in an increase in the delayed afterdepolarization amplitudes from 5.7 ± 2.0 to 11.3 ± 2.1 mV (p < 0.001) before sustained triggered activity occurred. The increase in the amplitude of delayed afterdepolarizations occurred in the presence of minimally changed membrane potentials at rest (−56.7 ± 7.3 mV before and −60.2 ± 6.7 mV after ouabain superfusion).

2) Preparations that showed sustained triggered activity before ouabain superfusion. In 8 of 10 preparations that showed triggered activity before ouabain superfusion, triggered activity persisted for more than 90 minutes after ouabain superfusion began and the rate of the activity increased. This increase in the rate of triggered activity was due to an increase in the slope of the delayed afterdepolarization (Fig. 4). During control conditions, five stimulated action potentials (Fig. 4A) resulted in sustained triggered activity with a cycle length of 720 ms (Fig. 4A and B). Ouabain superfusion for 90 minutes resulted in shorter cycle lengths of triggered activity (Fig. 4C and D). The shorter cycle length occurred most probably as a result of an increase in the slope of delayed afterdepolarization after the ouabain superfusion (dashed lines in Fig. 4B, C and D). A significant increase in the slope of delayed afterdepolarization was seen in five preparations showing acceleration of the triggered activity during superfusion with ouabain (slope before ouabain superfusion 29.2 ± 5.9 mV/s and after 90 minutes of ouabain superfusion 57.2 ± 11.8 mV/s; p < 0.01). In the other three preparations, impalement in the cell showing the dominant rhythm could not be maintained; therefore, analysis of the slope of the delayed afterdepolarization could not be performed. The increase in the rate of triggered activity and the slope of delayed afterdepolarizations occurred despite an increase in maximal diastolic potential and action potential amplitude. In contrast, such an increase in maximal diastolic potential and action potential amplitude in preparations superfused with normal Tyrode’s solution was accompanied by a decrease in the rate of triggered activity and slope of delayed afterdepolarizations (Fig. 2). Table 2 lists the effects of 90 minutes of ouabain superfusion on 10 preparations showing sustained triggered activity before ouabain superfusion. In contrast to preparations superfused with normal Tyrode’s solution (Table 1), preparations superfused with Tyrode’s solution containing ouabain showed shorter cycle lengths of triggered activity after 90 minutes of superfusion (cycle lengths before superfusion with Tyrode’s solution containing ouabain 836.6 ± 66.1 ms, after superfusion 602.5 ± 117.7 ms; p < 0.01) (Table 2). The shortened cycle length of triggered activity occurred despite the increase in maximal diastolic potential and action potential amplitude due to the time course of the preparations. When the cycle lengths of the triggered activity observed in preparations superfused with Tyrode’s solution containing ouabain for 90 minutes were compared with those observed in preparations superfused with normal Tyrode’s solution for the same length of time, a significant difference was observed (602.5 ± 117.7 ms versus 1,225.0 ± 310.6 ms, p < 0.01). Furthermore, in only 2 (20%) of 10 preparations did triggered activity cease after 90 minutes of superfusion when ouabain was present in the superfusate, whereas 5 (42%) of 12 preparations lost triggered activity after 90 minutes of superfusion when no ouabain was added.

**Discussion**

Enhancement of triggered activity in ischemic Purkinje fibers by ouabain. Recent in vivo studies (8) suggest that dogs with previous infarction are more susceptible to digitalis-induced ventricular tachycardia. The tachycardia was found to originate from the infarcted or periinfarcted areas and have focal characteristics. In vivo and in vitro canine studies (13,17) also suggested that the ventricular arrhythmias occurring in the early phase of myocardial infarction could be due to a focal mechanism in the Purkinje network underlying the infarction, possibly caused by delayed afterdepolarizations and triggered activity. Furthermore, previous studies (18–23) suggested that digitalis-

**Figure 4.** Acceleration of triggered activity by superfusion with Tyrode’s solution containing ouabain. A and B indicate control and C and D indicate activity after 45 and 90 minutes, respectively, of superfusion with ouabain. *Dashed lines* indicate the slope of delayed afterdepolarizations. *Arrowheads* indicate the stimuli (S) that initiated the triggered activity. Acceleration of triggered activity occurred despite an increase in maximal diastolic potential and action potential amplitude. CL = cycle length (in ms); other abbreviations as in Figure 1.
induced ventricular arrhythmias also originated from the Purkinje fibers and are caused by delayed afterdepolarizations and triggered activity. The similar basic mechanisms responsible for ventricular arrhythmias caused by myocardial infarction and ouabain through their synergistic actions conceivably may explain the postinfarction increased susceptibility to digitalis-induced ventricular arrhythmias.

A nonsteady state usually characterized isolated preparations of ischemic subendocardial Purkinje fibers in normal Tyrode’s solution. The trend in any one experiment was for resting and maximal diastolic potentials, action potential amplitude and maximal rate of rise of phase 0 to increase. The gradual increase of the resting and maximal diastolic potentials was characteristically associated with gradual attenuation of delayed afterdepolarizations and gradual decline of triggered activity associated with slower rhythmic activity. Finally, triggered activity could no longer be initiated. This type of recovery of Purkinje fibers overlying a 1 day old myocardial infarct has been previously emphasized by Lazzara et al. (16), who speculated on its possible mechanism. In the group of preparations superfused with ouabain, there was still an increase in maximal diastolic potential and action potential amplitude secondary to the time course of superfusion.

Because of the unstable characteristics of the action potentials recorded from Purkinje fibers surviving myocardial infarction and the relatively slow action of ouabain, we decided to compare the action potential characteristics of ischemic Purkinje fibers superfused with normal Tyrode’s solution and Tyrode’s solution added with ouabain. This was our attempt to determine how ouabain changes the time course of ischemic Purkinje fibers superfused with normal Tyrode’s solution. Our approach can be contrasted to the approach taken by Brennan and Bonn (12), who made their control observations at least 2 hours after isolation of the preparation. As a result of this approach, they failed to observe delayed afterdepolarization and triggered activity in their control preparations. This was not surprising because the average maximal diastolic potentials for their “ischemic cells” was $-78.6 \pm 2.7$ mV. From the study showing delayed afterdepolarizations and triggered activity in ischemic cells (13), it was clear that the phenomenon could be seen in depolarized preparations and the gradual improvement of maximal diastolic potentials during prolonged superfusion would result in disappearance of delayed afterdepolarizations and triggered activity. Therefore, what Brennan and Bonn observed was the “tail” of a disappearing phenomenon. This explains the small percent of their preparations that showed delayed afterdepolarizations despite ouabain superfusion. Furthermore, since they did not observe sustained triggered activity in their control preparations, they could not describe the effect of subtoxic ouabain superfusion on the rate of triggered activity.

**Methodologic considerations.** The concentration of ouabain in this study was chosen on the basis of previous studies (24) showing that this range of “therapeutic concentration” of ouabain resulted in positive inotropic effect and not in intracellular potassium loss of rabbit atrial tissue. This concentration is in the same range as that used by Brennan and Bonn (12), and lower than concentrations used by Rosen et al. (22) to induce delayed afterdepolarizations in normal Purkinje fibers. In normal Purkinje fibers, superfusion with $2 \times 10^{-7} M$ of ouabain resulted in delayed afterdepolarizations and triggered activity in 25 to 35 minutes (22), whereas superfusion with $4 \times 10^{-7} M$ of ouabain for 90 minutes did not induce delayed afterdepolarizations or triggered activity.

In the experiments in which ouabain resulted in an increase in the amplitude of the delayed afterdepolarizations, one may ask whether the change in maximal diastolic potential alone could have increased the amplitude of the delayed afterdepolarizations and resulted in triggered activity, as has been described (25) in strophanthidin-induced delayed afterdepolarizations in normal Purkinje fibers. Although this cannot be completely ruled out, it seems unlikely in the ischemic preparations. In experiments in which the amplitudes of the delayed afterdepolarizations that resulted from a constant pattern of stimulated action potentials were followed during superfusion with normal Tyrode’s solution, there was a gradual decline in amplitudes as the maximal diastolic potential became more negative (13).

Superfusion with Tyrode’s solution containing ouabain

<table>
<thead>
<tr>
<th>Table 2. Effects of 90 Minutes of Superfusion With Tyrode’s Solution Containing Ouabain on Sustained Triggered Activity in Ischemic Purkinje Fibers</th>
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</thead>
<tbody>
<tr>
<td>Control (n = 10)</td>
</tr>
<tr>
<td>Maximal diastolic potential (mV)</td>
</tr>
<tr>
<td>Action potential amplitude (mV)</td>
</tr>
<tr>
<td>Cycle length (ms)</td>
</tr>
<tr>
<td>52.3 ± 4.6</td>
</tr>
<tr>
<td>59.7 ± 6.8</td>
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<tr>
<td>836.6 ± 66.1</td>
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*Two preparations lost triggered activity. n = number of preparations; NS = not significant. Probability (p) values are compared with control.
produced sustained triggered activity in ischemic fibers that showed only nonsustained triggered activity. Ouabain also shortened the cycle length of triggered activity and decreased the percent of preparations that lost triggered activity after 90 minutes of superfusion (to be contrasted to the longer cycle lengths and higher percent of loss of triggered activity in preparations superfused with normal Tyrode's solution). Those effects of ouabain occurred despite minimally changed time course of maximal diastolic potential and action potential amplitude of the ischemic preparations. The observation that 2 of the 10 preparations lost triggered activity despite ouabain superfusion may also be explained on the basis of the two competing effects, that is, enhancement of triggered activity by ouabain and suppression of triggered activity when maximal diastolic potential increased. It is possible to speculate, therefore, that in some preparations improvement in action potential characteristics and decline in triggered activity during superfusion might overshadow a potentially slower effect of ouabain in enhancing delayed afterdepolarizations and triggered activity.

Mechanism of the action of ouabain. How does ouabain enhance delayed afterdepolarization and triggered activity in ischemic Purkinje fibers? The triggered activity associated with digitalis toxicity is believed to be associated with an increase in intracellular ionic calcium concentration (26). This critical increase in intracellular ionic calcium concentration, in turn, causes oscillatory changes in this concentration and membrane conductance, which permits a transient inward current (27,28). The mechanism for diastolic afterdepolarizations and triggered activity in ischemic Purkinje fibers has not been studied. It may be postulated that the reduction of sodium-potassium adenosine triphosphatase pump activity as a result of ischemia results in an increase in intracellular sodium concentration (29,30). This increase reduces sodium gradient which, in turn, causes a secondary decrease in calcium ion extrusion by calcium-sodium exchange and an increase in intracellular calcium concentration (31). Thus, it may also be postulated that the common action of ischemia and digitalis in causing an increase in intracellular ionic calcium concentration may work synergistically to cause delayed afterdepolarizations and triggered activity in Purkinje fibers.

Limitations of the study. The concentration of the calcium ions in the superfusate used in this study as well as other studies (13,22,25) is about twice the physiologic concentration in the extracellular space in vivo. Since delayed afterdepolarizations and triggered activity are very sensitive to the changes in extracellular calcium concentration (13), some consideration should be given to the relevance of these in vitro findings as they are related to the in vivo conditions. In favor of the relevance of these in vitro results is the fact that catecholamine level is low in vitro when compared with that in vivo. High catecholamine levels in vivo may potentiate this phenomenon, allowing for the possibility that they could occur in vivo in the presence of a much lower ionic calcium concentration.

This study did not investigate changes in refractory periods and conductions secondary to ouabain superfusion, which might possibly increase the propensity for ventricular reentrant arrhythmias. However, such significant changes in refractory periods and conductions were not detected in a previous study (12).

Conclusions. Ouabain at a concentration that had no toxic effect on normal Purkinje fibers was found to enhance arrhythmias in ischemic Purkinje fibers from a 1 day old myocardial infarct by increasing the magnitude of delayed afterdepolarizations and enhancing triggered activity. These findings may represent the mechanism responsible for the increased susceptibility to digitalis toxicity due to previous myocardial infarction.

References


