Supernormal Excitability and Conduction in the His-Purkinje System of the Dog

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ABSTRACT

The electrophysiological characteristics of the period of supernormal excitability and supernormal conduction were investigated in the isolated canine His-Purkinje system. Strength-interval curves were determined as the minimum transmembrane current required to bring the impaled fiber to threshold potential following a conducted action potential. During the period of supernormal excitability, 17.0 ± 4.6% (SD) less current than that required during diastole was needed to reexcite fibers throughout the left and right bundle branch-Purkinje system. A period of supernormal excitability was not found in the His bundle proximal to its pseudobifurcation or in ventricular muscle. The period of supernormal excitability was voltage dependent in the bundle branch-Purkinje system; it began during phase 3 at full repolarization (88.8 ± 5.6 [SD] mV) and reached minimum current requirements at about 74.3 ± 5.8 mV. Action potentials evoked during this period were conducted faster than they were during diastole. The maximum rates of depolarization of these supernormally conducted action potentials were not greatly depressed compared with control rates. A period of supernormal conduction was not observed in the His bundle. When the external potassium concentration was increased from 2.7 mM to 5.0 mM or 7.5 mM, both the supernormal period of excitability and the period of supernormal conduction were eliminated in Purkinje fibers.

KEY WORDS

cardiac excitability
supernormal period
threshold potential
intracellular stimulation
premature beats

table: supernormal period: cardiac refractory period

A supernormal period of excitability was described in ventricular muscle by Hoff and Nahum in 1938 (1). This supernormal period occurred during ventricular recovery and was manifest as a decrease in the current required to reexcite the myocardium through surface stimulating electrodes. More recently, cardiac excitability has been evaluated using both unipolar cathodal and anodal surface stimulation (2-4). In general, supernormal periods or "dips" in the excitability curves seem to be associated with anodal stimulation but not with cathodal stimulation. The complexity of the response of the myocardium to surface stimulation has been further demonstrated by Hoshi and Matsuda (5), who have shown that cells in the region of a surface anode can be depolarized as well as hyperpolarized. In our attempt to relate changes in excitability to changes in impulse conduction, we felt that it was necessary to utilize a method of stimulation that more closely corresponds to the physiological stimulus offered by a conducted impulse: intracellular stimulation eliminates many of the complexities associated with surface stimulation, since depolarizing currents are injected into single impaled cells. By intracellularly injecting depolarizing current, Weidmann (6) has demonstrated a period of supernormal excitability in isolated Purkinje fibers from sheep and calves. This supernormal period results partly because, during the later phase of repolarization, the threshold potential has recovered more completely than has the membrane potential. The membrane potential, therefore, has to undergo a smaller degree of additional depolarization to reach threshold potential, and this depolarization can be brought about by a weaker depolarizing current. Since the experiments of Weidmann (6), relatively few studies on supernormal excitability have used intracellular stimulation. Childers et al. (7) have reported a period of supernormal excitability in the specialized atrial fibers of Bachman's bundle and an associated period of supernormal conduction between the right and left atria. In a study of
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Conduction of premature impulses in isolated canine Purkinje fibers, van Dam et al. (8) found that no change in conduction velocity occurred during late repolarization; however, the earliest impulses that could be conducted were conducted in a way which indicated that the activity probably originated at a distance from the stimulating electrode. Therefore, the distal recording site appeared to be activated earlier than sites proximal to the stimulating electrode. This phenomenon makes the evaluation of supernormal conduction difficult, since an early premature beat which is apparently conducted faster than it would be during diastole might actually be conducted as a dissociated wave front.

The purpose of the present study was to characterize conduction and excitability of premature beats in the isolated canine His-Purkinje system and to relate conduction characteristics to the electrophysiological properties of the tissue. To distinguish true supernormal conduction from conduction with activity originating at a distance from the stimulating site, transmembrane potentials were recorded at the site of intracellular stimulation.

Methods

Experiments were performed on isolated canine Purkinje fiber-papillary muscle preparations and isolated canine His bundle-brunch branch preparations. Hearts were excised from dogs anesthetized with sodium pentobarbital (30 mg/kg, iv), and right and left Purkinje fiber-papillary muscle tissues were rapidly removed and placed in Tyrode's solution. The His bundle and the proximal bundle branches were exposed by dissection according to the method of Elizari et al. (9) and maintained in Tyrode's solution. The Tyrode's solution was equilibrated with 95% O₂-5% CO₂ and maintained at 37°C. Transmembrane potentials were recorded using standard glass microelectrodes filled with 3M KCl. The preparations were paced at a constant basic cycle length of 800 msec with silver bipolar surface electrodes. Our technique for stimulating and recording from a single microelectrode has been described previously (10). Briefly, it involves rapid relay circuits that electronically switch the microelectrode between the record mode and the stimulate mode. In the stimulate mode, depolarizing current pulses 4-10 msec in duration were delivered through the microelectrode. The current intensity was measured as the voltage drop across a series 100-kohm resistor. Following delivery of the current pulse, the system was switched within 4 msec to the record mode, and the resulting changes in transmembrane potential were recorded. The recordings were displayed on an oscilloscope and photographed on 35-mm film. The analogue data were then projected on a film reader (Microinsurance Inc.), and the currents, potentials, and time intervals were measured. Current was measured to the nearest 0.002 x 10⁻⁴ amps, potential was measured to the nearest 0.5 mv, and time was measured to 0.03 msec.

Figure 1A demonstrates the method used to evaluate membrane excitability, voltage changes in the region of the stimulating electrode, and conduction time of the evoked responses. The preparations were impaled with three intracellular microelectrodes. PF₁ is a recording from a Purkinje fiber with a microelectrode capable of both stimulation and recording. PFᵢ is a differential recording from an impalement within a space constant of the PFᵢ fiber, and PFᵢ is a recording several millimeters downstream from PFᵢ and PFᵢ. An action potential (arrow) was evoked by applying an intracellular depolarizing current through the PFᵢ electrode at just threshold intensity. The potential at which the cells near the stimulation electrode produced an all-or-none response was obtained by displaying the record from the PFᵢ fiber on an expanded time scale. One test of close electrical coupling between the site of current injection (PFᵢ) and the site of voltage recording (PFᵢ) is that, during diastole with currents slightly greater than threshold intensity, the latency between the beginning of the current pulse and the evoked action potential recorded at PFᵢ can be made extremely brief. During threshold voltage determinations, the intensity of current was adjusted so that the action potential was evoked with a consistent latency of about 4 msec. The conduction time of the evoked response was obtained by observing on an expanded time scale the difference in activation times between PFᵢ and PFᵢ; the excitability at the stimulation site was measured as the threshold current intensity. In some preparations, the input resistance of the membrane was also measured. As shown in Figure 1B, a hyperpolarizing current was injected (arrow) through the PFᵢ electrode. The input resistance was calculated as the ratio of the voltage displacement of the membrane potential at PFᵢ to the current intensity of the injected pulse. By scanning the conducted beat with a hyperpolarizing pulse, the input resistance could be obtained throughout the time course of an action potential.

Results

OCCURRENCE OF SUPERNORMAL EXCITABILITY IN THE HIS-PURKINJE SYSTEM

Using intracellular stimulation, excitability curves were determined in the His-Purkinje system as the minimum current required to reexcite a fiber at various times following a conducted action potential. Measurements of complete excitability curves following conducted activity in 58 cells sampled throughout the His-Purkinje system and ventricular muscle demonstrated a consistent pattern. Figure 2 compares typical excitability curves from the proximal portion of the His bundle (A), a right-sided false tendon (B), a transitional Purkinje type fiber (C), and a ventricular muscle cell (D). A period when the current required to reexcite the fibers was reduced (supernormal period of excitability) was not observed in either His bundle fibers or ventricular muscle fibers; there was sim-

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FIGURE 1
Analogue data demonstrating the technique used to measure excitability, threshold potential, conduction time, and input resistance. In both A and B, all traces were recorded simultaneously. The 100-msec time pulses apply to traces PF, PF, and PF. The 1-msec time pulses apply to traces (PF) and (PF) and to the current record. PF indicates transmembrane action potentials recorded with a microelectrode capable of both passing current and recording potentials. The PF transmembrane action potentials were recorded within 0.02 mm (within a space constant) of the PF recording site using a differential amplifier. The transmembrane action potentials in PF were recorded about 4 mm downstream from the PF electrode. (PF) and (PF) are the PF and PF recordings on an expanded time scale; the current record (on the expanded time scale) is the voltage drop across a 100-kohm precision resistor. The vertical bar to the left of the current record indicates 0.3 μA. A: A depolarizing current pulse was delivered through the PF electrode at just threshold intensity (arrow). The action potentials evoked as a result of this depolarizing current are shown on the expanded time scale. B: A hyperpolarizing current pulse was delivered through the PF electrode (arrow). The resulting potential change is displayed on the expanded time scale.

VOLTAGE DEPENDENCE OF THE SUPERNORMAL PERIOD

In all 29 cases in which a supernormal period of excitability was measured, the reduced current requirements for excitation occurred during late repolarization (phase 3) before the membrane potential had returned to its resting value. The Purkinje cells exhibited their minimum current requirements at an average potential of 74.3 ± 5.8 mv (Table 1). At more depolarized levels, the current requirements increased until the tissue became absolutely refractory. The regions of the His bundle that did not exhibit periods of supernormal excitability also exhibited low normal resting potentials. In 14 impalements in the proximal and middle portions of the His bundle in six different preparations, the resting potentials averaged only 69.6 ± 6.1 mv (Table 1). The low resting potentials in the proximal portions of the His bundle could have resulted from the extensive dissection required to isolate this preparation. However, we feel that this possibility is unlikely because the other electrophysiological characteristics of the proximal His bundle were not typical of damaged tissue (Table 1). That is, the action potential amplitudes were comparable to those in...
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Electrophysiological Basis of Supernormal Excitability

To investigate the mechanisms of supernormal excitability, transmembrane potentials were recorded near the current-passing electrode during determinations of the excitability curves (Fig. 1A). Figure 3 shows representative results from similar experiments in three preparations in which excitability and local voltage changes were measured in the His bundle and false tendon cells. The threshold potentials measured in this experiment (crosses along solid line) were at a more depolarized level than the true membrane threshold potential determined by a voltage clamp, because cable properties complicated the system. The cells near the site of the injected current were depolarized to a greater degree than the true membrane threshold potential before a conducted response was obtained. This phenomenon has been analyzed by Noble and Hall (11) and Fozzard and Schoenberg (12) and will be discussed later in the present paper. The potential records and the corresponding intracellular currents necessary to elicit conducted action potentials are plotted on the same time axis in Figure 3. It is obvious that the His bundle did not exhibit a supernormal period of excitability in association with the later period of repolarization (Fig. 3A). The threshold potential of the His bundle during repolarization was always in a less recovered state than the action potential. In Figure 3B, however, the Purkinje fiber exhibited a reduced current requirement for excitation during the later part of repolarization. During this supernormal period of excitability, the threshold potential recovered at a greater rate than did the action potential. Therefore, during the later part of repolarization, the threshold potential in the Purkinje fiber was closer to the membrane potential. During this time, a smaller degree of depolarization was required to bring the tissue to a firing level. This difference between His bundle and Purkinje fibers with regard to recovery of full excitability is one factor that explains the absence of the supernormal period of excitability in His bundle fibers and its presence in Purkinje fibers.

The membrane input resistance was also determined for both His bundle and Purkinje fibers (Fig. 4). For both the His bundle and the Purkinje fibers, an increased input resistance was associated with the plateau of the action potential. This increased resistance gradually returned during repolarization to a steady level at diastole, corresponding to the

Other tissues, and the diastolic threshold current requirements were low relative to those seen in damaged tissue. In addition, the action potential depolarization rates and the conduction velocities were characteristic of normal tissue (see Fig. 6A). The proximal portions of the bundle branches near the pseudobifurcation underwent the same dissection, and these cells exhibited greater resting potentials and possessed periods of supernormal excitability.

The absence of supernormal excitability in the His bundle may be related to the fact that the resting potential of His bundle cells levels off during diastole at approximately the potential at which Purkinje fibers exhibit minimum current requirements, suggesting that supernormal excitability may actually be a reflection of a "loss of excitability" during diastole as Purkinje fibers repolarize beyond the potential at which minimum current requirements occur.

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**TABLE 1**

Electrophysiologic Characteristics of Ventricular Tissues

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Rest potential (mv)</th>
<th>Action potential amplitude (mv)</th>
<th>Absolute refractory potential (mv)</th>
<th>Diastolic threshold current (% change from diastolic current)</th>
<th>Supernormal threshold potential (mv)</th>
<th>Supernormal period duration (msec)</th>
</tr>
</thead>
<tbody>
<tr>
<td>His bundle (14)</td>
<td>69.6 ± 6.1</td>
<td>94.6 ± 8.7</td>
<td>62.0 ± 5.7</td>
<td>0.666 ± 0.419</td>
<td>74.3 ± 5.8</td>
<td>88.2 ± 23.6</td>
</tr>
<tr>
<td>Bundle branches (24)</td>
<td>88.8 ± 5.6</td>
<td>105.5 ± 6.6</td>
<td>55.1 ± 6.6</td>
<td>0.318 ± 0.119</td>
<td>74.3 ± 5.8</td>
<td>88.2 ± 23.6</td>
</tr>
<tr>
<td>Ventricular muscle (18)</td>
<td>82.4 ± 4.3</td>
<td>93.6 ± 6.4</td>
<td>77.8 ± 4.9</td>
<td>1.54 ± 0.57</td>
<td>74.3 ± 5.8</td>
<td>88.2 ± 23.6</td>
</tr>
</tbody>
</table>

All values are means ± sd. Number of excitability curves measured is given in parentheses. No supernormal period was found in His bundle or ventricular muscle preparations.

Behavior of membrane impedance first described by Weidmann in 1951 (13). In the four preparations in which input resistance was measured, the ratio of peak input resistance during the plateau to input resistance during diastole was consistently about 40% greater for the Purkinje fibers than it was for the His bundle fibers, i.e., during the plateau phase a given current produced a greater displacement of membrane potential relative to diastole in the Purkinje fibers than it did in the His bundle fibers.

This difference in input resistance between His and Purkinje fibers also contributes to the presence of a supernormal period of excitability in Purkinje fibers and its absence in His bundle fibers.

In five preparations, high external potassium concentrations of 5.0 mM and 7.5 mM eliminated the period of supernormal excitability in Purkinje fibers. Figure 5 shows superimposed data in the same Purkinje fiber recorded at external potassium concentrations of 2.7 mM and 5.0 mM. The broken

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Representative examples of simultaneous determinations of threshold potential and excitability for the His bundle (A) and for a Purkinje fiber (B). In the top graphs, the broken lines indicate the voltage time courses of the transmembrane action potentials. The crosses along the broken lines represent the potentials during the recovery of the action potential at which each of the current pulses was delivered (the take-off potentials). The crosses above each of the take-off potentials (connected by a solid line) are the thresholds potentials at which an all-or-none response was evoked. The corresponding threshold currents are plotted in the bottom graphs on the same time axis.

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A characteristic 12-mv depolarization and a reduction in the duration of the action potential were associated with the increase in potassium concentration from 2.7 mM to 5.0 mM; the membrane also became more excitable during diastole. There was a 41% decrease in the current necessary to excite the fiber during diastole in the 5.0 mM potassium solution (0.355 vs. 0.210 μamp). This increase in excitability at moderately high levels of external potassium was first reported by Siebens et al. (14) using surface stimulation. Even though the fiber was more excitable during diastole, 5.0 mM potassium eliminated the period of supernormal excitability. Plotted together with the action potential time courses in Figure 5 are the threshold potentials for excitation in 2.7 mM and 5.0 mM potassium. Even though the membrane potential was depolarized 12 mv during diastole in 5.0 mM potassium, the threshold potential was 7 mv closer to the resting membrane potential. Dominguez and Fozzard (15) have established that this phenomenon is the cause of the increased excitability seen during diastole in moderately high potassium solutions. The excitability of the fiber in the 2.7 mM potassium solution recovered to a greater degree than did the membrane potential during the recovery phase. This decrease in the distance between membrane potential and threshold potential during the late part of phase 3 was associated with the supernormal period of excitability, as described previously in Figure 3B. In contrast, in the higher concentration of potassium, the excitability did not recover to as great a degree as did the membrane potential during the repolarization phase. Therefore, in 5.0 mM potassium the distance between the membrane potential and the threshold potential was always greater during the recovery phase than it was during diastole. This difference in high potassium solutions partially accounts for the disappearance of the period of supernormal excitability. In this preparation the input resistance of the same cell was determined in both normal and high potassium (Fig. 5). During diastole, the input resistance was decreased in 5.0 mM potassium, i.e., for a given current, the membrane potential was displaced to a smaller degree in 5.0 mM potassium than it was in 2.7 mM potassium. This effect of moderate increases in external potassium on the diastolic input resistance has also been described by Dominguez and Fozzard (15). The decrease in input resistance associated with high potassium solutions should antagonize the effect on excitability of the decrease in the voltage difference between threshold and resting potential in high potassium solutions. Figure 5 also shows that, although there was a decrease in resistance with 5.0 mM potassium, the peak input resistance in both cases was approximately the same. However, in the 5.0 mM potassium solution, the curve was shifted to the

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**Figure 4**

Input resistance determinations for the His bundle (A) and a Purkinje fiber (B). The time courses of the action potentials are displayed in the top graphs. The abscissas are the times after the upstroke of the action potential. The ordinates of the bottom graphs are the input resistances measured as the ratio of the membrane voltage displacement to the injected current intensity.
Effect of elevating the potassium concentration in the Tyrode's solution from 2.7 mM to 5.0 mM on membrane excitability, threshold potential, and input resistance. The data with both 2.7 mM and 5.0 mM potassium are superimposed and plotted on a common time axis. The broken lines represent the voltage time course of the action potential of the same cell (one impalement) in 2.7 mM and 5.0 mM potassium. The threshold currents necessary to excite the fiber in the two potassium solutions are displayed above the voltage time course curve; dots represent 2.7 mM potassium and crosses indicate 5.0 mM potassium. Input resistances at various times during the action potentials in the two concentrations of potassium are plotted at the bottom of the figure. The respective threshold potentials in 2.7 mM and 5.0 mM potassium are plotted above the action potential time courses.

left. This shift was greater than the corresponding decrease in action potential duration in the 5.0 mM potassium solution; therefore, at a given membrane potential during repolarization, the input resistance was less in the 5.0 mM potassium solution than it was in the 2.7 mM potassium solution. This effect also acts to reduce the supernormal period of excitability in the 5.0 mM potassium solution. Additional experiments were carried out in 7.5 mM potassium. At this higher potassium level, diastolic excitability was decreased in contrast to the finding in 5.0 mM potassium solution. However, although diastolic excitability was decreased, the supernormal period was eliminated as it was in the experiments described in Figure 5.

The change in the excitability of the Purkinje fiber in 2.7 mM and 5.0 mM potassium (Fig. 5) again suggests that supernormal excitability is a reflection of a loss of excitability associated with repolarization of Purkinje fibers beyond the potential exhibiting minimum current requirements. In the 5.0 mM potassium solution the Purkinje fiber repolarized and maintained a diastolic potential at about the potential of minimum current requirements. In this sense, one might say that in 5.0 mM potassium the fiber did not lose excitability with repolarization as it did in 2.7 mM potassium but was maximally excitable throughout diastole.

RELATIONSHIP BETWEEN SUPERNORMAL EXCITABILITY AND SUPERNORMAL CONDUCTION

Experiments were also performed to investigate the relationship between excitability and conduction using a third microelectrode to impale a fiber several millimeters from the current-passing and voltage-recording electrodes (PF, in Fig. 1). Figure 6 presents representative experiments on His bundle (A) and Purkinje fibers (B). Two superimposed action potentials from proximal (P) and distal (D) cells are shown at the top of Figure 6A and B. In this figure, P corresponds to the PF, cell in Figure 1A, and D corresponds to the PF cell. By passing current through a recording electrode, excitability during repolarization was determined as previously described. In addition, the conduction times of the evoked responses were determined as the time difference between activation of the proximal and distal cells (Fig. 6, middle graphs). The rates of maximum depolarization for the evoked responses were also electronically determined and are plotted in the graphs at the bottom of Figure 6. All data are on the same time axis; zero time is the upstroke of the action potentials.

The excitability curve for the His bundle fiber in Figure 6A exhibits no supernormal period of excitability. The plot of the conduction times for these prematurely evoked His bundle responses demonstrates a corresponding increase in conduction times as the beats became more premature. There was no indication of a decreased conduction time associated with the repolarization phase in the His bundle. The maximum rate of depolarization (dV/dt) for both proximal and distal cells decreased precipitously as the premature beats were evoked progressively earlier during the relative refractory period.

Corresponding data for the Purkinje fiber are presented in Figure 6B. The excitability curve for the Purkinje fiber shows the typically observed decrease in current requirement associated with repolarization. A decrease in conduction time corresponds to the period of supernormal excitability.
i.e., premature beats evoked during the time when the proximal cell was supernormally excitable were conducted to the distal cell with a shorter conduction time than those beats evoked during diastole. The plot of maximum dV/dt for the proximal and distal Purkinje cells indicates that the rate of rise of the action potentials of the premature beats did not decrease as precipitously as the relative refractory phase was infringed upon. In fact, because of the distance between proximal and distal cells, the distal cell (crosses) showed very little decrease in maximum rate of rise before conduction block occurred between proximal and distal recording sites. The period of supernormal conduction described in Figure 6B was consistently associated with the period of supernormal excitability in all five Purkinje fiber preparations studied. However, with the very earliest premature beats, there was an inconsistency in conduction behavior in the various preparations. The very early premature beats evoked before the period of supernormal excitability exhibited conduction times between the proximal and distal cells which could be either increased or decreased, depending on the particular experiments. This very early phase of the relative refractory period was associated with depressed rates of rise of action potentials as shown in the bottom graph of Figure 6B. Also the current required to excite the Purkinje fibers was increased during this time. It is probable that the slow rate of rise of the action potentials coupled with the increased current requirements caused the slowing of conduction. In some cases, the conduction time for very early premature beats appeared to approach zero, i.e., proximal and distal electrode activation was very nearly simultaneous. This phenomenon was described by van Dam et al. (8), who suggested that it was due to activation at a distance from the stimulation site. During these very premature beats, our voltage records at the site of intracellular stimulation indicated that a propagated subthreshold response (16) did occur, and the real site of all-or-none activation was at some distance from the stimulation site with retrograde reflection to the proximal site and antegrade conduction to the distal recording site.
Figure 7 demonstrates that high potassium solutions eliminate the period of supernormal conduction as well as the supernormal period of excitability in Purkinje fibers. The figure shows superimposed data from a single cell impalement in 2.7 mM and 5.0 mM potassium; this preparation is the same as that described in Figure 5. The 5.0 mM potassium solution caused a 12-mv depolarization and a reduction in action potential duration. Also associated with the change in membrane potential was a 41% decrease in the current required to excite the fiber during diastole. Along with increased excitability during diastole in the 5.0 mM potassium solution, a decrease in conduction time between proximal and distal recording electrodes occurred.

In the 5.0 mM potassium solution, the supernormal period of excitability was eliminated as indicated in the excitability curves at the right of the action potentials. The supernormal period of conduction was also eliminated. The slight decrease in dV/dt in association with the 5.0 mM potassium solution was probably the result of the slight depolarization.

Discussion

In our experiments the supernormal period of excitability was voltage dependent; it always terminated a full repolarization, and the minimum current threshold during the supernormal period occurred at 74.3 ± 5.8 mv. The supernormal period of excitability was restricted to the bundle branch-Purkinje system. The absence of the supernormal period in the His bundle was associated with a relatively low resting potential in this tissue. Ventricular muscle, which also did not exhibit a supernormal period, became refractory to our stimulation at a potential that was greater than the potential of the supernormal period in the Purkinje fibers. The ventricular muscle cells exhibited the highest diastolic current thresholds of the tissues that we studied (Table 1). Since the maximum current output through our experimental apparatus was about 5 µamp, the true absolute refractory period was probably not measured in these ventricular fibers. However, in all cases the point at which we could no longer stimulate the impaled muscle fiber was on the steep ascending limb of the excitability curve; therefore, we can definitely rule out the possibility that we missed a period of supernormal excitability. The ventricular muscle cells located near the transitional type Purkinje fibers tended to have lower diastolic thresholds than those in the high ventricular septum, possibly indicating that adjacent Purkinje type fibers have some influence on the excitability of ventricular muscle cells. However, a supernormal period of excitability was never recorded in any ventricular muscle fiber. Part of the complexity of the excitability curves generated by surface stimulation (1-4) may result because of the relatively large area of influence of surface electrodes. In fact, the supernormal excitability of ventricular myocardium observed when surface stimulation is used may be due to stimulation of adjacent Purkinje fibers, and its appearance may therefore partly depend on electrode configuration and location relative to the Purkinje system.

The data in Figures 3-5 indicate that the appearance of a period of supernormal excitability in the
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Purkinje system and its absence in the His bundle are primarily due to the differences in behavior of the threshold potential in these two tissues. In Purkinje fibers during repolarization, the excitability recovers to its resting state faster than does the membrane potential. Therefore, there is a period of time during repolarization when the membrane potential is closer to threshold potential than it is during rest. This phenomenon does not occur in the His bundle. Threshold potential in these experiments is not membrane threshold potential; rather it is a threshold potential that partly depends on the cable properties of the fiber. This concept of "liminal length" was first described by Rushton (17) and later analyzed by Noble and Hall (11) and Fozzard and Schoenberg (12). Briefly, the concept is that, at the point of current injection, the potential of the fiber must be raised to a level considerably more depolarized than the membrane threshold potential to generate a conducted response. A certain minimum amount of membrane (liminal length) must be raised above the membrane threshold potential to counter the repolarizing effect of local currents from the adjacent inactive membrane. A change in the measured threshold potential at the point of current injection could be due either to a change in the true membrane threshold potential, to a change in the cable properties, or to both. Using voltage-clamped Purkinje fibers, Weidmann (6) found that the period of supernormal excitability was due to the rapid recovery of the membrane excitability. The rapid recovery of excitability measured by intracellular stimulation in our experiments on Purkinje fibers was probably due to a combination of the recovery of the membrane threshold potential and the changes in cable properties associated with repolarization. Since a conducted wave of activity relies on local depolarizing currents to excite fibers downstream, the factors that modify excitability as measured by intracellular stimulation in our experiments would be expected to similarly modify excitability in the case of a conducted beat.

The changes in input resistance that occur during repolarization do not appear to contribute as much to supernormal excitability as does the behavior of the threshold potential. Figures 4 and 5 indicate that there is a greater input resistance during the time of repolarization than during diastole for both His bundle and Purkinje fibers. Therefore, in both cases the current required to hyperpolarize the membrane a given amount during repolarization is less than that required during diastole. However, the reduced threshold current requirement during repolarization occurs only in the Purkinje fibers. The relative increase in input resistance associated with the plateau of the action potentials is less for the His bundle than it is for Purkinje fibers. In the His bundle, the increase in input resistance is apparently too small to oppose the factors that contribute to the absence of supernormal excitability. In addition, the relative unimportance of the input resistance in determining excitability is seen in Figure 5 where the potassium-depolarized Purkinje fiber is considered. A reduced threshold current requirement during diastole was associated with the increase in potassium even though an associated substantial decrease in the diastolic input resistance still existed.

The basis for the lower resting potential in the His bundle is unknown. The slower rate of recovery of the membrane excitability compared with the membrane potential in the His bundle may be analogous to the behavior of Purkinje fibers in high potassium solutions. The possibility that repolarization in the His bundle proceeds by an ionic mechanism different from that in Purkinje fibers has been suggested for ventricular muscle by Giebisch and Weidmann (18).

According to the local circuit theory of conduction, a reduced current requirement for excitation during late repolarization would mean that cells could be brought to threshold potential sooner downstream and, therefore, an increment in conduction velocity would be realized. Our experiments demonstrate a direct correlation between a period of supernormal excitability and a period of increased conduction velocity. Our contention that supernormal excitability is a cause for supernormal conduction is supported by two findings. First, the absence of a supernormal period of excitability in the His bundle is associated with the absence of supernormal conduction, and the presence of supernormal excitability in Purkinje fibers is associated with supernormal conduction. Second, in Purkinje fibers when supernormal excitability is eliminated by depolarization with moderately high potassium, supernormal conduction is also eliminated.

Unexpected acceleration in conduction in the intact animal can be due to a wide variety of mechanisms (19); one such mechanism is supernormal excitability. Our experiments indicate that supernormal excitability associated with late repolarization may cause supernormal conduction in the bundle branch-Purkinje system. However, this mechanism probably does not accelerate conduc-
tion in the His bundle and ventricular muscle. In addition, supernormal excitability in the bundle branch–Purkinje system is eliminated by elevated extracellular potassium concentrations and cannot be a mechanism for supernormal conduction under these conditions.

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