The effects of single premature stimuli on automatic and triggered rhythms in isolated canine Purkinje fibers

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ABSTRACT We studied the effects of single premature stimuli on automatic and triggered rhythms occurring in preparations of isolated canine Purkinje fibers. Preparations were made from false tendons, the subendocardial right bundle branch, and infarct zone Purkinje fibers 24 hr after occlusion of the left anterior descending coronary artery, and were studied by standard microelectrode techniques. Single premature impulses almost always produced reset of automatic rhythms, whether the pacemaker had a low (< -60 mV), intermediate (-61 to -70 mV), or high (> -70 mV) maximum diastolic potential. In contrast, single premature impulses imposed on triggered rhythms sometimes were found to alter these rhythms; often, early premature impulses (i.e., during phase 3) resulted in either a shortened first return cycle or a short period of arrest of the rhythm. The results of this study indicate that it may be possible to design simple electrophysiologic tests using single premature stimuli to determine whether an arrhythmia is being caused by an automatic focus or by triggered activity. Circulation 71, No. 4, 813-822, 1985.

IT IS THOUGHT that ventricular ectopic activity can be caused by a variety of mechanisms at the cellular level, including normal automaticity, abnormal automaticity, triggering, and reentrant activation. However, at this time there are no precise tests available to determine which of these mechanisms is causing a particular arrhythmia of the heart in situ. To develop such tests, it will be necessary to better understand the response of automatic and triggered foci in preparations in vitro to stimulus regimens such as those that can be applied to the heart in situ. We have previously reported that short periods of overdrive stimulation can be used to determine the approximate maximum diastolic potential of the pacemaker fibers of a preparation of cardiac Purkinje fibers; preparations with pacemakers with high (> -70 mV) maximum diastolic potentials (and “high potential automaticity”) show postoverdrive suppression, whereas pacemaker fibers with low (-60 to -40 mV) maximum diastolic potentials (which we have referred to as “low potential automaticity” or “abnormal automaticity”) show postoverdrive enhancement or no change in automaticity after a short period of overdrive stimulation. In this study, we report on the effects of single premature impulses on automatic and triggered activity in isolated canine Purkinje fiber bundles and ventricular muscle preparations.

Few studies have been reported on the effects of single premature stimuli on automatic rhythms in ventricular pacemakers. Klein et al. reported on the effects of “extrasystoles” on the return cycle of automaticity in isolated canine Purkinje fibers. They found that premature action potentials elicited during the late stages of repolarization usually produced a shortened “return cycle” (the interval from the upstroke of the premature impulse to the upstroke of the subsequent, or returning, automatic beat) in fibers with normal maximum diastolic potentials “because of the marked abbreviation of the duration of the [premature] action potential.” They also found that “extrasystoles” elicited during phase 4 tended to prolong the return cycle of normal automaticity in isolated Purkinje fibers. However, little information is available on the effects of single premature action potentials on automaticity in partially depolarized pacemaker fibers. This study was designed to elucidate the effects of single premature action potentials on automatic activity in isolated canine Purkinje fibers at both the high (> -70 mV) and low level (-60 to -40 mV) of membrane potential.
During the course of this study, five preparations of subendocardial Purkinje fibers showed "triggered activity." The response of these preparations to single premature stimuli were somewhat different than those of the automatic preparations and may allow the development of electrophysiologic tests to discriminate between automatic and triggered ectopic rhythms. Preliminary reports of this study have appeared previously.  

Methods

These studies were carried out on preparations of canine cardiac tissue with standard microelectrode techniques, as have been described in detail previously. Briefly, healthy adult dogs were anesthetized with sodium pentobarbital, thoracotomies were performed, and the hearts were quickly excised and bathed in cold Tyrode's solution. The composition of the Tyrode's solution was: NaCl 137 mM, NaHCO₃ 12 mM, dextrose 5.5 mM, NaH₂PO₄ 1.8 mM, MgCl₂ 0.5 mM, and CaCl₂ 2.7 mM. For most of the experiments, the KCl concentration in the solution was 4 mM, although in some experiments on automaticity in fibers with normal maximum diastolic potentials the concentration was reduced to 2.7 mM. In the tissue bath, the pH averaged 7.25 to 7.45, and the temperature was maintained at 38±0.5°C (+0.1° in any experiment).

Most experiments were conducted on isolated false tendon preparations, which were devoid of ventricular muscle. Seventeen of these false tendon preparations were obtained from mongrel dogs, and four were obtained from male Siberian huskies. Usually, only one preparation was studied from each heart. In 16 experiments (on 12 fibers from mongrels and on the four fibers from the huskies) we studied the effects of single premature stimuli on normal automaticity in the isolated false tendon preparations. In seven of the preparations from mongrel dogs the fibers were exposed to barium-Tyrode's solution (see below) after the studies on normal automaticity had been completed; in the remaining five preparations only normal automaticity was studied.

In eight experiments we studied the effects of single premature stimuli on automaticity in barium-treated Purkinje fibers. In these studies, false tendons from normal dogs were isolated and superfused in the usual manner, and after a 30 to 45 min control period they were exposed to Tyrode's solution containing barium chloride (125 to 250 μM). After 2 to 5 min of exposure to the barium-Tyrode's, the fibers became automatic and fired from reduced maximum diastolic potentials. In two preparations the pacemaker cells had "intermediate potential automaticity" (which we have previously defined as automaticity occurring from maximum diastolic potentials between -61 and -70 mV), and in the remaining six preparations the pacemaker fibers had "low potential automaticity" with maximum diastolic potentials of -60 mV or less. We have also referred to "low potential automaticity" as "abnormal automaticity,". In 12 experiments the effects of single premature stimuli were tested on sustained rhythmic activity in preparations of infract zone Purkinje fibers obtained 20 to 24 hr after ligation of the left anterior descending coronary artery. At time of study, two to four preparations of subendocardial Purkinje fibers were excised from the endocardial anteroseptal walls in the central zones of the infract, as has been described previously. The preparations were about 2 to 3 cm² in surface area and 1 to 3 mm thick. Of the preparations were placed in the tissue bath and the pacemaker region was identified. Two series of experiments were done; initially, studies were done on eight preparations to attempt to characterize the effects of single premature stimuli on "automaticity" in the infract zone fibers. Four of these preparations had pacemaker activity occurring from maximum diastolic potentials of -40 to -60 mV, one preparation had a pacemaker maximum diastolic potential of -70 mV, and two preparations had pacemaker maximum diastolic potentials that were greater than -80 mV. In seven of these preparations the rhythm did indeed appear to be dependent on automatic mechanisms. However, in one preparation the impulses appeared to be caused by a triggered focus. The results of the first seven experiments indicated that single premature stimuli could produce very different responses in automatic and triggered foci in infract zone Purkinje fibers.

Therefore, after most of this study had been completed, four additional studies were carried out on selected infract zone preparations to better characterize the effects of single premature stimuli on triggered activity. These particular preparations were chosen for study because they had triggered activity as demonstrated by responses to overdrive stimulation. The infract zone preparations also were selected to have maximum diastolic potentials positive to -60 mV in their "triggered foci" because we have previously observed that preparations of infract zone Purkinje fiber with maximum diastolic potentials negative to -60 mV tend to fire in triggered bursts of action potentials, and these were not deemed suitable for study. Thus, if a short period of overdrive stimulation (either burst pacing or a 15 sec train at 300 msec or the shortest cycle length at which the preparation would conduct the impulses 1:1, if that were longer than 300 msec) produced either overdrive enhancement or no change in the rate of the rhythm after cessation of the drive, the rhythm was deemed to be caused by "abnormal automaticity." If the overdrive train was followed by delayed afterdepolarizations (either with or without an intervening salvo of impulses) and a quiescent period, it was deemed to be a "triggered rhythm."

In three preparations we tested the effects of single premature impulses on automaticity occurring in right bundle branch preparations. These preparations were approximately the same size as the infract zone preparations, consisting of a block of tissue from the right septum extending from the base of the main papillary muscle to the atrioventricular ring. The right bundle branch ran diagonally across the preparation, as previously described in detail. In each experiment, a pacemaker fiber was located and a stable impalement was obtained. A Schmitt trigger circuit provided pulsed outputs coincident with phase zero in the "pacemaker" action potential; a delay clock and a counting circuit (which together provided an output pulse after the desired delay every 8 to 40 cycles) triggered an output of an isolated stimulator. The stimulating electrodes were placed close to the pacemaker site (generally within 1 to 2 mm), and 1 to 2 msec bipolar pulses at about 2×threshold were delivered. Single premature stimuli were delivered during the diastolic interval (using the variable delay clock) from the end of diastole to the end of the effective refractory period. Premature impulses were elicited two to four times at a given delay, and then the delay interval was changed. In most experiments the stimuli were moved incrementally back and forth across diastole two to four times (usually in 10 msec increments). If the coupling interval initially had been incrementally increased, it was incrementally decreased in the second phase of the experiment (and vice versa). The results of these different techniques were consistent. A separate series of experiments was begun after it was observed that the fibers obtained from Siberian huskies responded differently to single premature stimuli than did normal fibers obtained from mongrel dogs. In these studies, 4 to 8 msec stimuli (about 2×threshold) were applied to six normal isolated
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FIGURE 1. Effects of single premature stimuli on automatic rhythms in normal preparations of Purkinje fibers. Top, Effects of single premature impulses (indicated by arrows) on rhythm in one preparation from a mongrel dog; early premature impulses shown at left, late impulses at right. Bottom, Left panel shows the one instance in which a single premature impulse led to long pause (“pacemaker annihilation”). Right panel shows unusual effect of midcycle premature stimulus in fiber from Siberian husky. Zero potential calibration marks are to left of each panel. Time calibrations for upper and lower panels are shown below each. Voltage calibration is at center right.

Purkinje fibers after every fortieth spontaneous beat (i.e., 150 to 200 sec apart). These stimuli were used because they consistently produced stable responses (30 to 45 sec periods of acceleration of automaticity). One Purkinje fiber bundle was obtained from each of three different male mongrel dogs and from three different male dogs that appeared to be purebred Siberian huskies. In two studies on the Siberian husky fibers, the responses to single premature stimuli were defined during the first 15 to 60 min; then the fibers were superfused with 0.2 μM propranolol for 30 min and changes in the response were examined.

All action potential data were displayed on chart paper with a Gould 2400 recorder. The data were normalized by dividing the coupling interval of the premature action potential by the previously occurring spontaneous cycle length; this will be referred to as the corrected forced cycle length (CFCL). Likewise, the spontaneous cycle length following the premature (the return cycle length) was also divided by the previously occurring spontaneous cycle length; this will be called the corrected return cycle length (CRCL).

Results

Effects of single premature stimuli on normal automaticity. In general, the corrected recovery cycle lengths that occurred in normal Purkinje fibers obtained from mongrel dogs after single premature impulses were as long as or longer than the control spontaneous cycle length occurring in that fiber (figure 1, top). The results of four experiments on normal automaticity are shown in figure 2 (filled circles). The data are from two false tendon preparations (25 data points), one barium-treated false tendon preparation with maximum diastolic potentials greater than −80 mV (six points), and one 24 hr preparation from the infarct zone with a pacemaker maximum diastolic potential of about −84 mV (28 points). In preparations with normal automatic activity, the CRCL was between 1.0 and 1.5 following all of the premature impulses except one. This atypical response occurred in a normal false tendon preparation; the premature impulse occurred at a CFCL of about 0.25, and the CRCL was prolonged to about 2.75 (figure 1, lower left; figure 2).

In contrast to the results obtained with fibers from mongrel dogs, in two experiments on fibers obtained from male Siberian huskies, transient increases in rate occurred after single stimuli; thus the return cycle lengths were shorter than the spontaneous cycle lengths (figure 2, open circles). After one stimulus at a CFCL of about 0.60 in a fiber from a Siberian husky, a short period of enhanced automaticity occurred as the maximum diastolic potential was transiently decreased.

FIGURE 2. Effects of single premature stimuli on normal automatic rhythms. Filled circles, Data from mongrel dog Purkinje fibers; open circles, data from Siberian huskies.
(figure 1, lower right; figure 2). Transient positive chronotropic responses occurred in the Siberian husky fibers even if the (2 msec) stimulus was delivered during the refractory period and did not elicit a premature action potential.

To further study this phenomenon, experiments were done on fibers from three mongrel dogs and three Siberian huskies, in which 5 to 8 msec stimuli were applied at intervals of about 3 min. The single stimuli did not produce acceleration of automaticity in any of the fibers from the mongrels, but in all three preparations from the huskies (two from the left ventricle and one from the right ventricle) a transient period of enhanced automaticity occurred after single "premature" electrical stimuli. In two of the experiments the preparations were superfused with Tyrode's solution containing 0.2 μM propranolol to block the β-adrenergic receptors, to determine whether this would affect the transient "tachyarrhythmia." Propranolol was added to the superfusate after a control period of 30 to 35 min, during which time the rates and intervals of accelerated automaticity were similar after each of the electrical stimuli. The "tachyarrhythmia" that followed single premature stimulation was obtunded during the first few minutes of exposure to propranolol in each experiment, and within 30 min of exposure, the poststimulation positive chronotropic effect was completely abolished (figure 3). In one experiment the duration of the stimulus was then increased to 10 msec, and the voltage was increased to the maximum allowed by the stimulator (approximately 42 V). Even under these conditions, the positive chronotropic response could not be elicited.

Effects of single premature stimuli on barium-induced automaticity. In eight experiments we studied the effects of single premature stimuli on automaticity in barium-treated Purkinje fibers. Six of the fibers had low potential automaticity and two had intermediate potential automaticity. The responses of these partially depolarized fibers were similar to those of the fibers in normal Tyrode's solution with high potential automaticity. That is, after most of the premature impulses, the preparations showed perfect reset of the pacemaker activity or slight prolongation of the recovery cycle length, relative to control spontaneous cycle length (figure 4). In a few instances after stimuli at short coupling intervals, the preparations showed a relative increase in the recovery cycle length (figure 5). Only two occurrences of recovery cycle lengths deviating by more than 10% from control values were observed in the barium-treated fibers; both of these unusual responses occurred within a 15 sec period in one preparation (figures 5 and 6). After the first stimulus at a forced cycle length of 400 msec (CFCL = 0.55), the spontaneous cycle length shortened. The first spontaneous impulse after the stimulated beat occurred after an interval of 590 msec (or about 20% shorter than the control spontaneous cycle length). The rate of the preparation then slowed gradually, until after 8 beats the cycle length was 650 msec. At that point, another premature stimulus was delivered and the "tachycardia" was terminated. Repeated stimuli (10 times) at this same coupling interval, and thereafter more than 40 additional stimuli at other coupling intervals, failed to elicit additional bouts of accelerated spontaneous impulse initiation in this preparation.

Effects of single premature stimuli on automaticity in preparations of infarct zone Purkinje fibers. In six experiments we studied the effects of single premature stimuli on automaticity in preparations of Purkinje fibers from 24-hr-old infarct zones. With techniques described previously,2 four of the pacemakers showed low potential automaticity, one showed intermediate potential automaticity, and one showed high potential automaticity.
automaticity. The results of the five experiments on preparations with low and intermediate potential automaticity are summarized in figure 7. In general, the results obtained from the infarct zone preparations were similar to those obtained from the other automatic preparations: after a premature impulse, the pacemaker showed perfect reset or a slight (relative) prolongation of the return cycle length.

In five preparations overdrive stimulation protocols indicated that delayed afterdepolarization–induced triggering rather than automaticity caused the sustained rhythmic activity. In two of these preparations, single premature stimuli produced only reset, whereas exceptions to this pattern of reset/slight prolongation of the pacemaker cycle length occurred in the remaining three preparations. In the first of these experiments the preparation was essentially quiescent (firing once every 10 to 20 sec) when impalements were initially obtained approximately 2 min after it was placed in the tissue bath. As the preparation was superfused, the rate of impulse initiation increased, and within 5 min it fired action potentials in pairs (figure 8A, top). Shortly thereafter, the preparation began to trigger regularly (figure 8A, middle). When the preparation was then studied with single premature stimuli, it was found that premature impulses elicited during phase 1 only reset the rhythm (figure 8B, top row) but that impulses during phase 3 could produce “secondary premature responses” (figure 8B, second row, first arrow) or could lead to a relatively long delay until the next spontaneous beat (figure 8B, second row, second and third arrows). On several occasions, single stimuli terminated the triggered activity (figure 8A, bottom; figure 8B, third row). It was also found that this triggered activity could be arrested by a short period of overdrive stimulation (figure 8B, bottom).

In a second experiment on triggered activity in an infarct zone preparation (surface area 10 × 11 mm), it was found that during a 30 sec period of conduction block provoked by burst pacing (figure 9A, bracket), the cycle length of the rhythm was decreased during the period of conduction block. Then this sustained rhythm was transiently suppressed after the conduction block was relieved for 3 beats (figure 9A, asterisks).
However, rhythmic activity persisted elsewhere in the preparation, as indicated by the (presumably) electrotonic deflections present at one site (upper trace): these electrotonic pulses were completely out of phase with the preceding regenerative impulses. Later, a single premature stimulus early in the cycle (figure 9, B, first asterisk) precipitated a similar period of shortened cycle length at one site (upper trace) and conduction block to the second site (lower trace). Then the triggered activity (and the conduction block between the two sites) was terminated after delivery of a single premature stimulus (figure 9, B, second asterisk). This single stimulus produced a regenerative response at the second site (lower trace) and a slightly shortened cycle at the first site (upper trace). Activity at the first site terminated with a small delayed afterdepolarization (figure 9, B, arrow). Again, electrotonic deflections occurred during the quiescent period. The rhythm was restarted, after a warm-up period, by a single stimulated impulse.

A third experiment on the effects of single premature stimuli on triggered activity in infarct zone Purkinje fibers was done simultaneously with the experiment illustrated above (figure 9) on a second preparation from this infarct. This preparation had a surface area of about 9 × 19 mm, and was shown to have triggered activity by burst pacing (figure 10, A). Shortly after this, single premature stimuli were applied and were generally found to produce only reset of the rhythm (figure 10, B). However, starting approximately 30 min later, a different sort of response was produced by single stimuli. Initially single premature stimuli could produce an increase in maximum diastolic potential (10 to 14 mV) before the next cycle (figure 10, C, first panel). Later, the maximum diastolic potential could shift approximately 20 mV negative, and the "triggered rhythm" would restart after 2 to 3 sec (figure 10, C, second and third panels). Later, these abrupt hyperpolarizations of maximum diastolic potential occurred spontaneously. The triggered activity could then be restarted at the low level of membrane potential occurred spontaneously.

![Graph](image-url)

**FIGURE 6.** Effects of single premature stimuli on abnormal automaticity; unusual "triggered" response in a barium-treated Purkinje fiber. Format as in figure 3; for further explanation, see text.

**FIGURE 7.** Effects of single premature stimuli on abnormal automaticity in preparations of infarct zone Purkinje fibers. Format as in figure 2. Each symbol represents data from a different infarct. Data from intermediate-potential pacemakers shown by circles; all other data from low-potential pacemakers.
FIGURE 8A. Triggered activity in preparations of infarct zone Purkinje fibers. Top and middle panels. Onset of sustained rhythmic activity. Bottom panel. Termination of rhythm with single early diastolic extrastimulus (arrow). Voltage calibrations are at the left of each panel; time calibration below each panel at right.

potential by a single premature stimulus (figure 10, C, fourth panel).

Effects of single premature stimuli on automaticity in right bundle branch preparations. Finally, three experiments were done to study the effects of single premature extrasystoles on automatic activity in right bundle branch preparations. These preparations had been stored for 6 to 8 hr in oxygenated Tyrode's solution at room temperature before study (the false tendons from the heart were studied during the storage period). The

FIGURE 8B. Triggered activity, same preparations as in figure 8A. Top row. Effects of single premature action potentials (at arrows) elicited during late, middle, and early diastole (left to right, respectively). Second row. Effects of premature stimuli during phase 3. Third row. Termination of triggered rhythm by single premature stimulus during phase 3. Bottom. Record before (left) and after (right) 15 sec period of overdrive stimulation at cycle length of 300 msec; rhythm terminated by single triggered beat, then delayed afterdepolarization. Format as in figure 3.
fibers in the bundle branch were partially depolarized in all three preparations, but the underlying ventricular muscle cells had normal resting potentials. Since right bundle branch fibers in freshly dissected and superfused preparations have normal maximum diastolic potentials (i.e., > -85 mV), it is reasonable to assume that these preparations depolarized during the period of storage and did not recover during superfusion. In each of the preparations, unidirectional conduction block occurred between the partially depolarized bundle branch fibers and the underlying ventricular muscle cells; impulses could be conducted from the working myocardium to the Purkinje fibers, but not vice versa. In all three preparations the single premature extrasystoles were delivered to the bundle branch via the ventricular muscle. The extrasystoles elicited during phase 4 only reset the rhythm (figure 11). However, extrasystoles elicited during phase 3 produced less predictable responses. In some instances, only subthreshold electrotonic responses occurred (figure 12, A); in other instances, one or two secondary premature responses occurred during the electrotonic pulse (figure 12, B to E).

**Discussion**

These data indicate that automatic rhythms in (1) false tendon Purkinje fibers with normal automaticity occurring from the high level of membrane potential, (2) in partially depolarized Purkinje fibers in barium-treated false tendon preparations, or (3) in surviving subendocardial Purkinje fibers from the central regions of 24-hr-old infarct zones all respond to single premature stimuli in a similar way. That is, each type of preparation examined usually showed reset of the rhythm so that the return cycle following the premature impulse was ±40% of the control spontaneous cycle length.

In most instances, normal Purkinje fibers showed reset of automatic activity or prolongation of the first return cycle by up to 30%; the experiments in which normal pacemakers usually showed shortened return cycles after electrical stimulation were carried out on the fibers from Siberian huskies. These periods of enhanced automaticity were presumably caused by re-

**FIGURE 9.** Effects of burst pacing (A) and single premature stimuli (B) on triggered activity in infarct zone Purkinje fiber. Two impalements maintained in the preparation. Temperature 37.5°C. Time and voltage calibrations are shown at lower left. For further explanation, see text.

**FIGURE 10.** Effects of burst pacing (A) and single premature stimuli (B and C). Temperature 37.5°C. Time and voltage calibrations are shown at lower left. For further explanation, see text.
lease of endogenous catecholamines, since they could be blocked by treatment with 0.2 \( \mu \text{M} \) propranolol. It has been demonstrated that drugs (such as tyramine or bretylium) can liberate enough endogenous catecholamine from isolated canine Purkinje fibers to produce an increase in the rate of automaticity.\(^{11,12}\) The results of the present experiments suggest that it may be necessary to control for the breed of dog used to provide isolated tissues if liberation of endogenous catecholamines can influence the results.

In preparations with abnormal (or low potential) automaticity, where the pacemaker fibers had maximum diastolic potentials of \(-60\) to \(-40\) mV, the effects of single premature stimuli were largely similar to the effects of single premature impulses on preparations with normal (or high potential) automaticity. In both barium- and infarct-induced abnormal automaticity, most extrasystoles were followed by a return cycle that was the same as the control cycle length or slightly prolonged relative to the control. In some of these experiments, however, extrasystoles were followed by return cycles that were up to 20% shorter than the control.

Therefore it appears from these data that it would not be possible to develop a test based on the response to single premature stimuli to discriminate in vivo between normal and abnormal automatic rhythms. However, it may be possible to make this discrimination based on the response to overdrive stimulation for 15 sec or more.\(^2\)

The obvious question, then, is would it be possible to use the responses to single premature stimuli to design a test to discriminate between automatic and triggered rhythms? The results of these experiments suggest that it may be, since the rhythm of triggered activity in infarct zone preparations apparently can be interrupted more easily than can rhythms from automatic foci. In two of the present experiments, rhythms apparently triggered from delayed afterdepolarizations were more easily arrested by single impulses very early in the cycle than by impulses late in the cycle (figures 8 and 9). Similar observations have been made previously by El Sherif et al.\(^{13}\) These results would suggest that triggered activity in infarct zone Purkinje fibers is somewhat different than triggering in simian mitral valve fibers, which is at least as easily interrupted by premature impulses late in the cycle as by early impulses.\(^{14}\) Our results may be related to the phenomenon of “pacemaker annihilation” as described by Jalife and Antzelevitch\(^{15,16}\); however, they do suggest that “annihilation” of a rhythm is more easily elicited in triggered than in automatic preparations. “Annihilation” was repeatedly produced in the triggered preparations, but in all of the automatic preparations was elicited on

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**FIGURE 11.** Effects of single extrastimuli during phase 4 on automaticity in right bundle branch preparation. Upper record in each panel shows transmembrane potential from ventricular muscle cell one cell layer below bundle branch; lower record shows bundle branch implanement. Premature impulses (arrows) delivered via ventricular muscle shown in each panel. Format as in figure 4. For further explanation, see text.

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**FIGURE 12.** Effects of single extrastimuli during phase 3 on automaticity in right bundle branch preparation. Format as in figure 11. For further explanation, see text.
only one occasion (figure 1, top, left panel), and even in this one preparation the event was not reproducible.

The results of the experiment in which single premature stimuli produced "delayed hyperpolarizations" indicate that in some infarct zone Purkinje fibers, it is possible to shift the diastolic membrane potential abruptly up and down between the normal and low level of membrane potential, presumably as a consequence of abrupt increases or decreases of potassium conductance. This would suggest that during the recovery period, it may be possible for triggered activity to occur in infarct zone Purkinje fibers as a consequence of early afterdepolarizations. Indeed, in one previous study, during washout of ethmozin, early afterdepolarizations occurred in an infarct zone Purkinje fiber preparation (results not presented).

Nevertheless, whether these results really could be used as the basis for a test to discriminate between automatic and triggered ventricular rhythms in vivo is open to some question, since the effect of most premature impulses on a (delayed afterdepolarization) triggered rhythm would be to produce only reset. Furthermore, it appears that only a minority of triggered rhythms in infarct zone fibers or ouabain-treated Purkinje fibers can be terminated by premature impulses. Therefore, with respect to most diastolic extrasystoles, a triggered ventricular focus would respond to single premature impulses in a manner similar to an automatic focus. And very closely coupled premature impulses, as would apparently be needed to arrest the triggered activity, might not be conducted well into a partially depolarized focus.

As in our previous study on overdrive stimulation of automatic rhythms,

One factor that must be taken into consideration in the interpretation of the results of [stimulation] protocols in the intact heart is the problem of entrance block into the ectopic focus. In the present in vitro studies, we could be reasonably certain in each preparation that every stimulated impulse was being conducted into the automatic [or triggered] focus. In the intact heart, one could not be certain of this unless the focus were identified and electrograms recorded from it before, during, and after the [stimulated] beat. If pacemaker electrograms could not be recorded, the data would have to be interpreted with more caution.

Obviously, if complete or variable entrance blocks, or variable exit blocks did occur, the interpretation of the results would be even more complicated. Presumably, if the focus (of either an automatic or triggered ventricular rhythm) were "protected" by an entrance block, only "interpolated" extra beats could occur. Or, if the extra beats could alter or affect a zone of exit block, a variety of responses might occur. Finally, sustained reentrant arrhythmias can also be terminated by appropriately timed single premature stimuli and thus might appear to be "triggered" by simplistic application of such a test.

In summary, the results of these experiments suggest that although it may be possible to develop tests to discriminate between automatic and triggered rhythms using single premature stimuli, several technical problems remain to be eliminated, and more information on triggered rhythms will be needed before such a test can be used routinely.

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References
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