Epicardial Phenol Interrupts Refractory Period Responses to Sympathetic but Not Vagal Stimulation in Canine Left Ventricular Epicardium and Endocardium

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SUMMARY We studied the effects of regional epicardial application of phenol on autonomic neural control of electrophysiological characteristics of the anterior left ventricle. In eight open-chest dogs, we applied a thin line of 88% phenol on the perimeter encircling a multipolar electrode and measured the effective refractory period (ERP) and recorded electrograms in phenol-encircled and untreated areas. Before phenol was applied, sympathetic nerve stimulation shortened ERP in all areas: percent change ($\Delta$) = 12. Phenol application to an area with a radius of 1-2 cm prevented ERP shortening in encircled epicardium during sympathetic stimulation, $\Delta A = 0.9 \pm 0.8$ (SEM), and attenuated ERP shortening in underlying endocardium, $\Delta A = 2.5 \pm 1.6$, compared to ERP shortening in untreated areas, $\Delta A = 10 \pm 1$. A subsequent phenol application to an area with a radius of 2-3 cm prevented ERP shortening during sympathetic stimulation in both encircled epicardium and underlying endocardium. Phenol did not alter electrograms, activation times, or ERP shortening produced by norepinephrine infusion; these data suggest that electrophysiological characteristics of ventricular muscle encircled by phenol were unchanged apart from effects of withdrawal of sympathetic neural influence. To support these functional data, norepinephrine content measured in phenol-treated epicardium and endocardium of three dogs was 7 and 21%, respectively, of the norepinephrine content of untreated areas. In eight additional dogs, vagus nerve stimulation during norepinephrine infusion prolonged ERP by 3-5 msec both before and after phenol encircling an area with a radius of 2-3 cm. In these dogs, phenol did prevent ERP shortening during sympathetic nerve stimulation. We conclude that epicardial phenol interrupts sympathetic neural influences to both epicardial and endocardial sites without impairing responses either to intravenous norepinephrine or to vagus nerve stimulation.


HISTOLOGICAL study of mammalian hearts has suggested that sympathetic efferent nerves approach the heart over the anterior great vessels, travel in the ventricular epicardium, and penetrate into the myocardium to innervate the endocardium (Hirsch, 1971). Vagal efferent nerves approach the heart over the posterior atria and may reach the ventricular myocardium via the ventricular septum as well as the epicardium (Hirsch, 1971). To provide functional confirmation of epicardial routes for sympathetic nerves, Szentivanyi et al. (1967), Randall et al. (1968), and Geis and Kaye (1968) interrupted nerves coursing immediately below the epicardium of the right and left ventricles and eliminated the augmentation of epicardial contractile response to sympathetic nerve stimulation. This observation supported the conclusion that sympathetic nerves traveling in the epicardium innervate the epicardium. These investigators did not test whether the epicardial interventions affected the contractile response of the endocardium to sympathetic stimulation.

The purpose of the present study was to determine whether epicardial nerve interruption produced by application of phenol to the epicardium could prevent effects of sympathetic nerve stimulation on the effective refractory period (ERP) of the epicardium and endocardium of the left ventricle. In addition, we studied the effect of epicardial phenol application on ERP responses to vagal nerve stimulation, because vagal nerves may reach the ventricular myocardium by routes separate from sympathetic nerves.

Methods

Surgical Preparation

Healthy mongrel dogs weighing 11-26 kg were pretreated with morphine (2.3 mg/kg) intramuscularly and anesthetized with a-chloralose (80 mg/kg)
intravenously. Additional amounts of chloralose were given as necessary to maintain anesthesia during the study. No measurements were obtained for at least 10 minutes after each additional dose of chloralose, and intervening dosages were avoided when serial measurements of refractory periods were obtained. The dogs were ventilated by means of a cuffed endotracheal tube and volume-cycled respirator. Arterial PCO₂, pH, and PO₂ were monitored throughout the experiment and were adjusted by varying the tidal volume. Positive expiratory pressure was used to maintain PO₂ in the normal range. Sodium bicarbonate was given intravenously to some dogs to maintain pH above 7.30. Mean values during periods of collection of electrophysiological data were PCO₂ = 32 ± 1 (SEM) torr, pH = 7.32 ± 0.02 and PO₂ = 76 ± 2 torr.

The sternum was split and the open pericardium was sutured to the wound edges to support the heart. The left femoral vein was cannulated to infuse drugs and normal saline, 200 ml/hr, during the surgical procedure. A fluid-filled cannula placed in the left femoral artery was connected to a Statham P-23 transducer to monitor pressure. Mean arterial pressure was determined by electrical filtering and recorded on an oscillographic recorder. To monitor epicardial temperatures continuously, a temperature probe was sutured to an anterior epicardial site located 1-2 cm from the multipolar electrode. The sternotomy was covered by a plastic sheet and, by adjusting the distance of the operating table lamp from the cardiac surface, epicardial temperature was maintained at 37.5°C.

Effective Refractory Period Testing

The region of the sinus node was crushed in all experiments. The atria and ventricles were paced simultaneously at basic cycle lengths of 300-400 msec to maintain control of ventricular rate during all interventions and to eliminate hemodynamic changes from varying ventricular filling owing to different timing of atrial and ventricular contractions. We paced the right atrial appendage with a bipolar hook electrode, and the left ventricle with unipolar cathodal stimuli delivered through one pole of a multipolar plunge needle (22-gauge) electrode. The anode was an electrode 3.5 cm in diameter and located in the subcutaneous tissue of the abdomen.

The ERP was measured by the extrastimulus technique (Krayer et al., 1951) employing a programmable stimulator with separate constant current outputs. The right atrial appendage and each ventricular test site were paced at one and one-half to two times late diastolic threshold. Stimulus duration was 2 msec. Ventricular late diastolic thresholds, measured during each intervention, averaged 29 ± 2 μA and if they varied by more than 10 μA, the data were discarded. Current intensity was kept constant.

The train of eight basic stimuli (S₁) was followed by a late premature stimulus (S₂) that initially produced a propagated ventricular response (capture). The ventricular response to S₂ was recorded in lead II and from an adjacent pair of electrodes on the multipolar needle and displayed on a storage oscilloscope at a rapid sweep speed. The S₁–S₂ interval was shortened by 2-msec intervals (e.g., 160, 158, 156-failed) until failure to capture occurred. Following this, the S₁–S₂ was increased by 9 msec and the shortening of the S₁–S₂ was repeated (e.g., 165, 163, 161, 159, 157-failed) 30 seconds later (Han and Moe, 1969; Janse et al., 1969). Each test site was stimulated at least twice before moving to another test site. A repeat ERP determination resulted in a value within 1 msec of the first, or the data were discarded. The ERP was defined as the longest S₁–S₂ which just failed to capture the ventricle. The longer ERP value was used for analysis. Serial testing after a single anesthetic dose without intervention in this animal model demonstrated ERP shortening of 0 to 1 msec in endocardial and epicardial sites over a 15-minute interval. Control values and the effects of interventions reported in this study generally were obtained in <10 minutes.
lying endocardium of these sites were stimulated and recorded from using multipolar plunge needle electrodes (interelectrode distance = 1 mm) inserted into the myocardium perpendicular to the epicardial surface.

The sites of all electrodes were confirmed at necropsy when the experiment was terminated. In addition, the location of endocardial and epicardial sites to be paced by each multipolar needle were confirmed by recording bipolar electrograms from adjacent poles of the multipolar needle during spontaneous supraventricular rhythm. After recording all electrogram combinations between the left ventricular cavity and epicardium, the endocardial pole chosen for ventricular stimulation was one from which the earliest transmural ventricular activation was recorded and, if possible, one from which a Purkinje spike also was recorded (Durrer and van der Tweel, 1956). The epicardial pole chosen for ventricular stimulation was one from which the latest transmural ventricular activation was recorded. The distance between endocardial and epicardial test sites ranged from 5 to 9 mm.

Electrogram Recording

Bipolar electrograms were recorded during atrial pacing from the two epicardial or endocardial electrodes immediately adjacent to the electrode chosen for stimulation. The signals were amplified, filtered between 1.2 and 500 Hz, displayed on a storage oscilloscope, and simultaneously recorded on a strip chart recorder at 400 mm/sec. Measurements of activation time from a reference bipolar electrogram in the right ventricular outflow tract to the peak of the recorded left ventricular electrogram, total electrogram amplitude, and electrogram duration were performed.

Autonomic Nerve Stimulation

Sympathetic

The proximal communications of both stellate ganglia were cut in all dogs. Shielded stainless steel bipolar electrodes were placed around the anterior and posterior ansa subclavia which still were connected to each ganglion. The electrode nearest the heart always was the cathode. Stimulation of right and left sympathetic nerves was carried out with separate constant current isolators driven at 6–8 Hz with pulses 4 msec in duration. Each nerve was tested separately to determine the current intensity at which the maximal change in heart rate or blood pressure was obtained. That current, ranging from 1 to 7 mA was used for the remainder of the experiment.

Vagus

The cervical vagi were isolated, doubly ligated, and cut in all dogs. Each vagus nerve was stimulated via two Teflon-coated hook electrodes imbedded in the peripheral cut nerve (Lazzara et al., 1973). Pulses were rectangular, 4 msec in duration, and delivered at a frequency of 20 Hz. The current strength was 0.05 mA greater than that required to produce asystole, or complete atrioventricular block (during spontaneous rhythm), and ranged from 0.2 to 1.5 mA. During each experiment, between interventions, bilateral vagal stimulation was repeated without varying the current strength, to be certain that the response to stimulation remained constant.

Because the vagus nerves antagonize effects of sympathetic activity in the ventricles (Kolman et al., 1976; Watanabe and Besch, 1975; Martins and Zipes, 1980) and because the sympathetic nerves were cut in all experiments, determination of the effects of vagal stimulation on ERP was performed during constant infusion of norepinephrine 0.4–1.2 μg/kg per min. The infusion rate was chosen to produce ERP shortening of at least 20 msec.

Protocol

In the first series of experiments in eight animals, the ERP responses to sympathetic nerve stimulation were determined by measuring ERP at all sites prior to and 1 minute after the same intensity and duration of bilateral sympathetic nerve stimulation. Phenol (carbolic acid) 88% then was applied to the epicardial surface of the left ventricle with a wooden stick. A 3- to 5-mm thick line was inscribed on the perimeter of a circle which surrounded the multipolar electrode located in the apical half of the ventricle (Fig. 1). Twenty minutes after the phenol application, ERP measurements were made before and during sympathetic nerve stimulation. If the first encircling application (radius = 1–2 cm) did not prevent ERP shortening in both epicardium and endocardium during sympathetic nerve stimulation, then a second phenol line (radius = 2–3 cm) was applied and ERP testing was repeated. Norepinephrine was infused at 0.4 mg/kg per min for 5 minutes and ERP testing was repeated.

In the second series of experiments in another eight animals, ERP was measured during constant norepinephrine infusion before, during, and after vagus nerve stimulation. The infusion then was stopped and phenol was applied to the epicardium to surround an area with a radius of 2–3 cm. Twenty minutes later, the norepinephrine infusion was resumed and ERP was measured before, during, and after vagal stimulation. The norepinephrine infusion was stopped, and 20 minutes later the ERP response to sympathetic stimulation was measured.

Norepinephrine Measurements

To determine further the effects of epicardial phenol application on sympathetic innervation, we measured norepinephrine content in endocardium and epicardium of three dogs. They were anesthetized with secobarbital (0.6 mg/kg) and the hearts were exposed through a left thoracotomy. A phenol circle with a 2- to 3-cm radius was applied to the anterior left ventricle. The chest was closed and the
dog was allowed to recuperate for 3 days to allow time for norepinephrine depletion from nerves. The dogs then were reanesthetized and the hearts were removed rapidly. Myocardial samples from the outer and inner thirds (epicardium and endocardium, respectively) were taken from the center of the phenol circle and from the untreated posterior left ventricle. Myocardial norepinephrine concentrations were measured using a fluorometric method (Dykstra et al., 1978). The same tissues were examined microscopically for catecholamine histofluorescence by the technique of Falck and Hillarp (Bjorlund et al., 1972).

Statistical Methods

All data were expressed as mean ± SE. Whenever two statistical comparisons were to be made on one control or intervention data group, two-way analysis of variance was performed, employing Duncan’s multiple range test for comparison when F values were significant (Steele and Torrie, 1960). When only one statistical comparison was employed between a control and an intervention, Student’s t-test for paired data was employed.

Results

Sympathetic Nerve Stimulation

Sympathetic nerve stimulation increased mean arterial pressure from 67 ± 6 to 87 ± 9 mm Hg (P < 0.05). Sympathetic stimulation shortened ERP in epicardium of the area to be encircled by phenol from 168 ± 7 to 147 ± 6 msec (P < 0.01), in the underlying endocardium from 170 ± 7 to 148 ± 6 msec (P < 0.01), and in untreated (control) epicardial and endocardial areas from 166 ± 7 to 143 ± 8 msec (P < 0.01).

After phenol encircling (radius = 2-3 cm), mean arterial pressure rose from 58 ± 8 to 84 ± 10 mm Hg (P < 0.01) during repeat sympathetic nerve stimulation. Phenol prevented ERP shortening during sympathetic nerve stimulation in encircled epicardium (165 ± 8 to 163 ± 8 msec P > 0.2) and substantially attenuated ERP shortening in underlying endocardium (164 ± 9 to 158 ± 7 msec, P < 0.01, Fig. 2). The percent ERP shortening in epicardium, percent change (Δ) = 0.9 ± 0.8, and endocardium, Δ = 2.5 ± 1.6, was significantly less (P < 0.01) compared with data for the same areas prior to phenol application, Δ = 12.0 ± 1.5 and 12.5 ± 2.0, respectively, and compared with untreated areas after phenol treatment, Δ = 10 ± 1.

The application of phenol to create a larger circle (radius = 2-3 cm) prevented ERP shortening by the sympathetic stimulation in both encircled epicardium (Δ = -0.32) and underlying endocardium (Δ = -0.8). ERP in untreated areas shortened by 8.7% during sympathetic stimulation.

To determine whether epicardial phenol application caused electrophysiological effects other than those resulting from sympathetic nerve inter-

Discussion

There are three major findings of this study. First, application of phenol to the epicardium prevented ERP shortening produced by electrical stimulation of the sympathetic nerves in the encircled epicardium. This suggests that sympathetic nerves to the area were interrupted. Electrogram amplitude and duration and time of activation, as well as ERP response to norepinephrine, were not altered, suggesting that electrophysiological characteristics of ventricular muscle were unchanged apart from withdrawal of sympathetic neural influence. Second, application of phenol to the epicardium also prevented ERP shortening of the underlying endocardium during sympathetic stimulation, suggesting that sympathetic nerves in the epicardium control refractoriness in both epicardium and endocardium. This conclusion is supported by marked decreases in norepinephrine content and absence of fluoro-
Autonomic Nerve Control of Ventricular Refractoriness/Martins and Zipes

Figure 2 The ERP during sympathetic stimulation is expressed as a percent of the immediately preceding control value. Shortening of ERP is (+)%Δ and prolongation of ERP is (−)%Δ. Data are shown for four interventions: the first sympathetic stimulation, the second stimulation after phenol was applied in a circle with a radius of 1-2 cm, a third stimulation (in some animals) after phenol was applied with a radius of 2-3 cm and cence in phenol-encircled epicardium and endocardium compared to untreated areas. Third, phenol application which prevented ERP shortening in response to sympathetic nerve stimulation did not change the ERP response to electrical stimulation of the vagus.

Sympathetic Nerve Stimulation

Previous histological studies have demonstrated that efferent sympathetic nerves from the right and left stellate ganglia reach the anterior heart via passage over the pulmonary artery and coronary vessels (Hirsch, 1971). Pathways distal to the pulmonary arteries have not been described histologically. However, functional studies have demonstrated that the anterior left ventricle receives its major sympathetic efferent nerve supply from nerves which course in the epicardium from the anterior descending coronary artery to the lateral wall; this course is much the same as that of a diagonal branch of the same artery. The latter information was determined by Szentivanyi et al., 1967; Randall et al., 1968; and Geis and Kaye, 1968, in studies in which they destroyed epicardial nerves by either removing strips of epicardium 1-mm thick or by applying phenol to the epicardium, which produced necrosis to a depth of 0.25 mm (Kaye et al., 1968). When either procedure was used adjacent to the anterior descending coronary artery, a strain gauge sutured to the anterior left ventricular epicardium lateral and distal to that area did not show augmented contractile force during electrical stimulation of right and left sympathetic nerves.

Many previous studies have documented sympathetic influences on cardiac electrophysiological properties (Burgess and Levine, 1976; Han et al., 1964; Kralios et al., 1975; Vassalle et al., 1969; Yawnowitz et al., 1966; Kent et al., 1974; Kolman et al., 1976; Spear and Moore, 1973). The present study demonstrates electrophysiological effects of sympathetic denervation limited to a region of the ventricle encircled by phenol. Thus, phenol application to the epicardium surrounding a test electrode prevents ERP shortening during sympathetic nerve stimulation. Yet, 5 minutes after a thin line of phenol enclosing an electrode was applied, electrogram amplitude, duration, and activation time are unchanged. Interruption of sympathetic nerves by any means would not be expected to affect these finally, during norepinephrine infusion. A: Epicardial test sites encircled by phenol; B: endocardial test sites underlying phenol-encircled epicardium; C: untreated epicardial and endocardial test sites. A phenol circle (radius = 1-2 cm) prevented a significant ERP shortening in treated epicardium and attenuated ERP shortening in treated endocardium compared to the slight decrease in ERP shortening in untreated areas. A 2- to 3-cm phenol circle prevented significant ERP shortening in both epicardium and endocardium. Catecholamine infusion produced ERP shortening in all sites.
TABLE 1  Effects of Phenol Treatment on ERP Response to VNS

<table>
<thead>
<tr>
<th></th>
<th>Before phenol</th>
<th>After phenol</th>
<th>Control</th>
<th>SNS</th>
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<tr>
<td></td>
<td>NE</td>
<td>VNS + NE</td>
<td>NE</td>
<td></td>
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<tr>
<td>Epicardium</td>
<td></td>
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<tr>
<td>msec</td>
<td>162 ± 6</td>
<td>167 ± 6</td>
<td>159 ± 6</td>
<td>149 ± 7</td>
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<tr>
<td>%Δ</td>
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<td>+2.3 ± 0.7</td>
<td>+0.5 ± 0.7</td>
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<td>Endocardium</td>
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<tr>
<td>msec</td>
<td>159 ± 5</td>
<td>162 ± 5</td>
<td>155 ± 5</td>
<td>151 ± 4</td>
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<tr>
<td>%Δ</td>
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<td>+1.4 ± 0.6</td>
<td>+0.2 ± 1.2</td>
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<td>Untreated areas</td>
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<tr>
<td>msec</td>
<td>158 ± 4</td>
<td>162 ± 5</td>
<td>153 ± 5</td>
<td>147 ± 4</td>
</tr>
<tr>
<td>%Δ</td>
<td>−2.8 ± 1.1</td>
<td>−2.2 ± 0.7</td>
<td>+7.4 ± 0.8</td>
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NE = norepinephrine infusion; SNS = sympathetic nerve stimulation; VNS = vagus nerve stimulation. %Δ = percent change between first and second values.

* P < 0.05; † P < 0.01; ‡ P < 0.01 vs. untreated areas.

Electrogram characteristics in normal ventricular muscle (Singer et al., 1967; Wallace and Sarnoff, 1964). Therefore, these data suggest that phenol does not exert direct electrophysiological effects on the myocardium in the area encircled. In addition, phenol encircling did not prevent the shortening of ERP produced by norepinephrine infusion, indicating that adrenergic receptors and the tissue response to receptor stimulation were normal.

Our results also supply the first functional support for the hypothesis that sympathetic innervation to the endocardium arrives via sympathetic nerves which course in the epicardium and penetrate the myocardium as do coronary vessels. Previous investigators (Randall et al., 1968) have suggested this hypothesis but have not produced evidence to support it, since endocardial contractile function was not measured. The present data clearly show that endocardial ERP shortening produced by sympathetic nerve stimulation is prevented by a distant epicardial application of phenol.

These data also suggest a pathway that sympathetic nerves could take through the anterior left ventricular wall. When phenol encircled an area around the electrode with a 1- to 2-cm radius, measured on the epicardium, the endocardial ERP response to sympathetic nerve stimulation was attenuated but not prevented as it was in the epicardium. Presumably some nerves reached the endocardial test site by penetrating the myocardium outside the phenol circle which was between the arrow and electrode in Figure 3. Since a phenol circle with a radius of 2-3 cm prevented ERP shortening in both epicardium and endocardium, the phenol line must have been beyond the point of penetration of most epicardial nerves into the myocardium; i.e., the phenol line was to the right of the arrow in Figure 3. Alternatively, the smaller phenol application may have interrupted sympathetic neural input from one direction (i.e., arising from the anterior descending coronary artery) whereas the larger phenol circle may have interrupted a second sympathetic neural input from another direction (i.e., arising from the circumflex coronary artery; Geis and Kaye, 1968). In either case, the data suggest that the anterior left ventricular endocardium of the dog is innervated by sympathetic nerves which course predominantly in the epicardium and penetrate the myocardium perpendicularly or on a diagonal path, possibly following coronary vessels (see below) to reach the endocardium.

Norepinephrine Measurements

The measurements of norepinephrine content in the phenol-treated epicardium and endocardium support the functional data. A phenol circle with a 2- to 3-cm radius reduced the norepinephrine content...
tent in the endocardium to 21% and in the epicardium to 7% of the norepinephrine content of untreated myocardium. Previous investigators also have measured norepinephrine content of the canine left ventricle 2 weeks after epicardial phenol application (Pace et al., 1969). Norepinephrine content of phenol-treated tissue was 10% of norepinephrine content of untreated tissue. However, these investigators did not separate the endocardium from the epicardium. Thus, our study of norepinephrine content in the phenol-treated anterior left ventricle adds a second line of evidence to support the hypothesis that epicardial sympathetic nerves penetrate the myocardium to innervate the endocardium.

**Vagus Nerve Stimulation**

Recent studies have established that electrical stimulation of the vagus nerves lengthens ERP in canine ventricular myocardium (Kolman et al., 1976; Martins and Zipes, 1980) by antagonism of sympathetic activity. It also is known that electrical stimulation of vagus nerves after stellate ganglionectomy produces inconsistent effects in anesthetized dogs (Martins and Zipes, 1980) so vagal stimulation was performed during norepinephrine infusion in the present study. These data suggest that phenol application to the ventricular epicardium (radius = 2–3 cm) interrupted sympathetic neural influences but did not alter ERP prolongation due to vagal stimulation. There are two possible explanations for these findings. Efferent parasympathetic nerves may be resistant to the necrotizing effects of phenol. This explanation is unlikely since, in several experiments not included in Table 1, phenol application to left atrial tissues along the lateral aspect of the circumflex coronary artery prevented vagus-induced ERP shortening in all ventricular test areas. The more likely explanation is that parasympathetic nerves reach the ventricular myocardium by some route other than the superficial epicardium within 3 cm surrounding the innervated test site (Hirsch, 1971).

The latter explanation is consistent with recent histochemical and functional data. Kent et al. (1974) demonstrated acetylcholinesterase staining of nerves in the endocardium and left septum of human and canine ventricles and comparatively few acetylcholinesterase-staining nerves elsewhere in the ventricular wall. Yet, electrical stimulation of the vagus nerves demonstrated no greater ERP prolongation in endocardium than epicardium (Martins and Zipes, 1980). These data are compatible if one considers that the apparent clustering of acetylcholinesterase stained nerves in the endocardium and left septum is produced by axons which merely travel through these areas to influence endocardium and epicardium remote from the sampled sites. Therefore, the nerves that leave this endocardial “trunk” to innervate the midwall and epicardium of a single histological section appear sparse, but the actual number of acetylcholine-releasing terminals in such a case could be equal in endocardium and epicardium and yield similar functional responses across the ventricular wall.

Employing this reasoning, we believe that the present data are consistent with the hypothesis that the major vagal efferent nerves to the anterior left ventricle of the dog arrive there via pathways in the endocardium. However, we cannot exclude the additional possibility that vagal epicardial pathways traversing the circumflex coronary artery, for example, penetrate the myocardium >3 cm from a test electrode and therefore influence at least endocardial refractoriness after 2–3 cm phenol application. In either case a phenol circle of 2–3 cm selectively removes sympathetic but not vagal influences limited to the treated area.

**Consideration of the Model**

The design of this study demonstrating regional sympathetic denervation required a baseline of bilateral stellate ganglion interruption with superimposed sympathetic nerve stimulation. Comparison of ERP responses in a phenol-treated area to the total response of an untreated area gave an indication of the percent reduction from the major sympathetic neural input to the heart (Randall, 1977). However, this model does not yield important physiological information regarding effects of epicardial phenol treatment on afferent nerves which travel with either sympathetic or vagal efferent nerves.

The application of phenol to epicardial fat produces necrosis of adipose cells to approximately the same depth as produced in myocardium (Kaye et al., 1968). Therefore, sympathetic nerves traveling in or under fat surrounding coronary arteries might not be interrupted by phenol. The contributions of such nerves was not of critical importance in five of eight animals in the present studies since, in these dogs, phenol applications in circles with radii of 1–2 cm did not involve any major coronary vessels yet still prevented ERP shortening during sympathetic stimulation in both endocardium and epicardium. However, in each of the remaining three dogs, phenol applications with 2- to 3-cm radii involved a large coronary vessel around which epicardial fat had been dissected. Had this dissection not been done, further inhibition of ERP shortening during sympathetic stimulation may not have been achieved.

The present study provides electrophysiological evidence for the hypothesis that the major sympathetic neural input to the anterior left ventricle courses over the epicardium and penetrates to the underlying endocardium. It also suggests that parasympathetic nerves reach the anterior left ventricular myocardium by some pathway other than one traveling through the immediate epicardium 3 cm encircling a test site.
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