Selective In Situ Parasympathetic Control of the Canine Sinoatrial and Atrioventricular Nodes

By Ralph Lazzara, Benjamin J. Scherlag, Morton J. Robinson, and Philip Samet

ABSTRACT

Methods were devised for the selective alteration of parasympathetic control over the sinoatrial (SA) or the atrioventricular (AV) node in anesthetized, thoracotomized mongrel dogs. Two epicardial sites were located at which parasympathetic nerve fibers enroute to the SA or the AV node could be stimulated or blocked. Selective nerve stimulation was accomplished with brief pulses (0.05 msec), and blockade was accomplished with topically applied lidocaine. At the intercaval site, only the SA node was affected. At the site near the coronary sinus ostium, only the AV node was affected in terms of parasympathetic control, but sometimes there were modest sympathetic effects on the SA node. The effects of stimulation at these sites, except for the SA speeding produced by stimulation at the site near the coronary sinus, were blocked by atropine. The effects were also blocked by ganglionic blockade. Probably, preganglionic parasympathetic fibers to the nodes are concentrated at these sites.

KEY WORDS intracardiac nerves vagal stimulation parasympathetic effects ganglionic blockade His bundle recording atropine lidocaine

Many investigators have observed autonomic effects attributed to the direct stimulation of nerve elements in preparations of excised cardiac tissues (1-10). Vincenzi and West (8) developed the technique of subthreshold stimulation, which permits the selective stimulation of autonomic fibers within the heart with brief pulses without the excitation of cardiac cells. This technique allows comparatively sharp localization of the resultant autonomic effects.

This paper describes the application of a variant of the technique of subthreshold stimulation that is applicable to the intact heart. There are epicardial sites at which the selective stimulation of intracardiac nerves produces highly specific, localized vagal effects on the sinoatrial and the atrioventricular nodes. Also, specific, localized blockade of vagal efferent impulses to either node can be accomplished.

Methods

Eleven mongrel dogs were anesthetized with sodium pentobarbital (30 mg/kg, iv), and their hearts were exposed through a right thoracotomy and a pericardiotomy. The vagosympathetic trunks were isolated in the neck. Bipolar plunge wires (stainless steel, 0.005 inches in diameter) were placed along the sinoatrial (SA) node, on the body of the atrium, near the mouth of the coronary sinus, and in the His bundle area (11). In several experiments, the His bundle electrogram was recorded with an electrode catheter at the atrioventricular (AV) junction (12). A bipolar electrode probe (interelectrode distance, 2-3 mm) was used for epicardial stimulation. A thorough exploration of the epicardial surface of the right atrium and some of the accessible surface of the left atrium was made with the stimulating electrodes. In some instances several contact electrodes were sutured onto the epicardial surface for the selective stimulation of intracardiac nerves. For stimulation of the cervical vagosympathetic trunk, the bare ends of two silver wires were inserted into the sheath of the cervical vagosympathetic trunks (Fig. 1). This method of stimulation was based on a comparison with a standard external contact electrode commonly used for nerve stimulation. Each length of Teflon-coated silver wire (0.012 inches in diameter) was passed through a 20-gauge hypodermic needle; the tip of each wire was stripped free of Teflon and bent over the bevel of the needle to form a small hook about ¾ inch long (Fig. 1). In six dogs, the right and the left common carotid sheaths were dissected in the neck, and the respective vagosympathetic trunks were tied or crushed centrally. The hypodermic needles containing the wire electrodes were inserted longitudinally into the nerve trunk. The right and the left trunks were used alternately. Each needle was carefully withdrawn so that the hook remained within the nerve. The wire was then inserted further to ensure that the portion of exposed silver wire was completely buried within the nerve. A commercially produced bipolar electrode was applied to the outside of the contralateral vagosympathetic nerve trunk. The nerve was separated from the carotid sheath only for the length necessary for the installation of the external electrode. The wire electrodes,
Changes in intra-atrial conduction were assessed by comparing the times between the stimulus artifact and a distant atrial electrogram or between two electrograms from different atrial sites. The effective refractory period of atrial muscle was determined with premature 2-msec stimuli of twice the late diastolic threshold voltage delivered through bipolar electrodes of stainless steel wire (0.005 inches in diameter). The refractory periods were compared at the same paced rates.

Lidocaine (1–3 ml of a 1% solution) was applied topically to the epicardial sites at which electrical stimulation produced either SA slowing or AV block. Atropine (2 mg, iv) was administered to produce cholinergic blockade. Trimethaphan camsylate, a ganglionic blocking agent, was administered by intravenous infusion of 500 mg in 500 ml of normal saline at a rate sufficient to lower mean blood pressure at least 30 mm Hg.

Results

Comparison of the effects of the internal wire electrodes and the external commercially produced electrodes is shown in Figure 2. The voltage used in each case was just the intensity needed to produce sinus asystole for at least 5 seconds; the average threshold voltage was 3.7 v for the external electrodes and 0.5 v for the internal wire electrodes. Figure 2 depicts the relationship between sinus slowing and the frequency of stimulation with both types of electrodes. The curves labeled 1 were taken immediately after insertion of the electrodes, and those labeled 2 were taken after 3 hours. Although the effects of the two types of electrodes were similar initially, there was a marked decrease at all
local nervous control of SA, AV nodes

frequencies in the effectiveness of the external unit at the end of 3 hours (compare curve 2° with 1°). The internal wire electrodes, however, maintained their effectiveness (compare curve 2° with 1°). The internal wire electrodes showed several advantages over the externally applied electrodes. Nerve stimulation was selectively achieved with the internal electrodes without electrical leakage to contiguous muscles. Stimulation through the internal wire electrodes produced effects equal to those of the external electrodes at lower voltages. Because of this lower threshold and the insulating effects of the nerve sheath, the stimulus artifact produced by the internal wire electrodes was much less conspicuous than that produced by the external electrodes. Finally, the internal electrodes did not lose their effectiveness for at least 3 hours.

Exploration of the epicardial surface of the atria uncovered two epicardial loci (SA site and AV site) where it was possible to specifically alter vagal tone on the SA or the AV node. The SA site was located in the intercaval region of the right atrium just posterior to the sulcus terminalis and near the pericardial reflection. It was at the anterior, superior border of a triangular accumulation of fat in the area. At this site, selective stimulation of intracardiac nerves produced slowing of the sinus rate which could be graded by altering the

---

**FIGURE 3**

Effects of selective stimulation of intracardiac nerves at SA site. The tracings from the top down are leads II (L-2) and aVR of the electrocardiogram, electrograms from the sinus node area (SA), the AV junction (Hb), and the mouth of the coronary sinus (CS), and a recording of the stimulus artifact from the nerve stimulator. A: With selective stimulation of intracardiac nerves there was a prompt slowing of the SA rate with the emergence of a functional pacemaker. The atrial rate (AR) and the functional rate (NR) are shown above the L-2 trace. B: Control record during atrial pacing. The SA electrogram shows the pacer artifact (PI). C: Selective stimulation of intracardiac nerves applied during atrial pacing produced no change in the times between the atrial and the His bundle deflections or between the pacer artifact and the atrial deflections. D: Atrial pacing was discontinued to show the sinus slowing from selective stimulation of intracardiac nerves.
stimulus voltage (Figs. 3 and 4). It was always possible to slow the sinus rate sufficiently to allow the emergence of escape junctional rhythms at rates of 70-120/min. There was no change in intratrial conduction, AV conduction (Fig. 3B–D), or the refractory period of the atrial muscle (Table 1) during cardiac stimulation at the SA site.

The AV site was located in a pocket formed between the terminations of the inferior vena cava and the coronary sinus on the posterior inferior right atrium. Although this site was slightly more difficult to localize than was the SA site, it was found in every dog, and selective stimulation of intracardiac nerves produced AV block without any change in the intra-atrial conduction or the refractory period of the atrial muscle (Fig. 5, Table 1). The degree of AV block could be controlled by changing the voltage of the pulses utilized in selective stimulation of the intracardiac nerves. In some dogs there was no change in the sinus rate, but in others a slight, gradual increase in sinus rate occurred (Fig. 4). This sinus speeding was not dependent on a high degree of AV block and slowing of the ventricular rate, because it occurred

<table>
<thead>
<tr>
<th>TABLE 1</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
<tr>
<td>Refractory Periods (msec) of Atrial Muscle</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Control</td>
</tr>
<tr>
<td>SSIN-SA</td>
</tr>
<tr>
<td>SSIN-AV</td>
</tr>
</tbody>
</table>

All values are means ± sd. CS = mouth of the coronary sinus, SA = near the SA node, Mid-RA = a central and anterior point in the body of the right atrium, SSIN-SA = selective stimulation of intracardiac nerves in the SA site, and SSIN-AV = selective stimulation of intracardiac nerves in the AV site.

FIGURE 4
Chronotropic effects of selective stimulation of intracardiac nerves at the SA site (A–C) and the AV site (D). The tachometer recording of the SA rate is shown on the third trace (AR). The other traces are labeled as they are in Figure 3. Note the graded sinus slowing with increasing stimulus voltage in A–C. In C a junctional pacemaker supervened and gradually speeded. The recovery was interrupted by ectopic atrial contractions. The sinus speeding from selective stimulation of the intracardiac nerves at the AV site was comparatively slow in development and decline. Note that there were no dropped QRS complexes (leads II, aVR) in this run.
in first-degree block (Fig. 4). The speeding effect was sometimes obtained at a site slightly different from the optimal site for producing AV block.

Topical epicardial application of lidocaine to the SA and the AV sites resulted in abolition of the effects of selective stimulation of intracardiac nerves. Also, the effects of stimulation of the cervical vagosympathetic trunk on the SA or the AV node were greatly attenuated or, in some experiments, abolished by application of lidocaine. Records from representative experiments before and after application of lidocaine to the SA and the AV sites are shown in Figures 6 and 7, respectively.

After atropinization, selective stimulation of intracardiac nerves at the SA and the AV sites no longer produced sinus slowing or AV block (Fig. 8). The positive chronotropic effect on the SA node from selective stimulation of the intracardiac nerves at the AV site was not affected by atropinization. Ganglionic blockade (Fig. 9) also abolished or attenuated the vagal effects of selective stimulation of intracardiac nerves.

Figures 10 and 11 are photomicrographs of sections from the SA and the AV sites, respectively. There are numerous nerve elements within the epicardium including abundant ganglion cells and myelinated nerves.

Discussion

In 1921 Lewis and co-workers (13) suggested that intracardiac nerves might be excited by stimuli applied directly to the intact heart. This suggestion aroused little interest; no purposeful application of intracardiac nerve stimulation in the intact heart ensued. However, the phenomenon has been encountered in studies on excised cardiac tissue, most often as an unanticipated experimental anomaly induced by the stimuli used for driving the cardiac cells. However, Ursillo (3) and later other investigators (7-9) showed that the autonomic nerve elements could be selectively stimulated, especially if brief pulses were used, and that these effects could be experimentally useful. Vincenzi and West (8) suggested that postganglionic nerve endings were stimulated, resulting in the release of neurohormones. Lewartowski (9) also concluded that in atrial preparations postganglionic elements were stimulated, because ganglionic blockade did not diminish the effects. However, in our studies the autonomic effects were profoundly suppressed by ganglionic blockade. This observation indicates that in the intact heart at the SA and the AV sites, preganglionic fibers enroute to the nodes are stimulated during the application of selective stimulation of intracardiac nerves.

![Figure 5](image-url)

**Figure 5**

Effects of selective stimulation of intracardiac nerves at the AV site. Recordings are labeled as they are in Figure 3. Complete heart block occurred within a second after stimulation was begun, and a ventricular escape pacemaker emerged. There was no change in sinus rate or in the interval from the SA electrogram to the CS electrogram.
The effects of selective stimulation of intracardiac nerves at the SA and the AV sites differ in still another respect from the effects reported in excised preparations. In excised tissues, intracardiac nerve stimulation has almost invariably produced mixed sympathetic and parasympathetic effects in the same area. However, with selective stimulation of intracardiac nerves at the SA site, there were purely parasympathetic effects on the SA node. No sympathetic effects were unmasked with atropine. With selective stimulation of intracardiac nerves at the AV site, there were sometimes mixed effects, but the effect on either of the nodes was pure. The effects on the AV node were always purely parasympathetic; the effects on the SA node were always purely sympathetic. The latter effects were not prominent and not constant. Within the limits of the methods used, no effects on ordinary atrial muscle were detected. However, the measurement of conduction time is an insensitive index of vagal effect on ordinary atrial muscle. Measurement of the refractory period is a more sensitive index of vagal effect, but it is uncertain whether the atrial muscle closely adjacent to the nodes was sampled.

Application of lidocaine to the SA and the AV sites produced great attenuation of the nodal effects of stimulation of both cervical vagal sympathetic trunks; in some experiments there was complete blockade. This observation, in conjunction with the...
Local application of lidocaine to the AV site. Recordings are labeled as they are in Figure 6 before (A) and after (B) topical applications of lidocaine to the AV site. In A there was 2:1 heart block during selective stimulation of intracardiac nerves and complete heart block during cervical vagal stimulation. In B the effects of both maneuvers on AV conduction were almost completely blocked despite the use of higher stimulus voltage. There were still negative chronotropic effects from cervical vagal stimulation.

**FIGURE 7**

Photomicrograph of a section from the SA site. A collection of nerve elements including ganglion cells and myelinated fibers is enclosed within the broken line. The needle tract is visible in the upper right corner. Note the inflammatory cell infiltrate around the needle tract. Hematoxylin and eosin stain.

**FIGURE 10**

Photomicrograph of a section from the AV site. A collection of nerve elements including ganglion cells and myelinated fibers is enclosed within the broken line. The needle tract is visible in the upper left corner and is surrounded by inflammatory cell infiltrate. Hematoxylin and eosin stain.
Abolition of effects of selective stimulation of intracardiac nerves by atropine. Labels are as they are in Figure 3. A: Control. B: Effects before atropine of selective stimulation of intracardiac nerves at the AV site. C: Effects before atropine at the SA site. D: Stimulation at the SA site produced no effects after atropine. E: Stimulation at the AV site produced no effect after atropine.

Abolition of effects of selective stimulation of intracardiac nerves by ganglionic blockade. Labels are as they are in Figure 3. The aortic blood pressure is shown on the lowest trace. The two lines denote 0 and 100 mm Hg. The stimulus artifact of the nerve stimulation appears on the Hb trace. A and B: Effects of selective stimulation of intracardiac nerves at SA and AV sites, respectively, before trimethaphan infusion. C and D: Lack of effects of stimulation of the SA and AV sites, respectively, during the infusion.
results of ganglionic blockade, indicates that most if not all of the preganglionic parasympathetic fibers influencing the nodes converge at these sites. The slight residual effects after lidocaine application could have been mediated by preganglionic fibers bypassing these sites. However, some nerve fibers at the SA and the AV sites may have escaped blockade.

Numerous anatomic studies (14-21) have demonstrated that the atrial wall in the vicinity of the vena caval terminations is replete with nerve fibers and ganglia. Preganglionic and postganglionic effenter fibers, ganglion cells, and afferent fibers have been identified. However, there has been no specific anatomic localization of the preganglionic fibers destined for the SA and the AV nodes. This study suggests that the preganglionic parasympathetic fibers from both vagi which influence SA nodal function converge at the SA site and that the preganglionic parasympathetic fibers influencing AV nodal function converge at the AV site. Also, there may be sympathetic fibers enroute to the SA node at the AV site. This convergence of fibers at the SA and the AV sites provides an opportunity to specifically increase (selective stimulation of intracardiac nerves) or decrease (local nerve block) vagal influence over the SA or the AV node.

Acknowledgment

We would like to acknowledge Sheldon Cantor, Marie Ellis, Klaus Jurachek, Duane Postlethwaite, Israel Dingle, and David Young for their efforts in this study.

References

Selective In Situ Parasympathetic Control of the Canine Sinoatrial and Atrioventricular Nodes
RALPH LAZZARA, BENJAMIN J. SCHERLAG, MORTON J. ROBINSON and PHILIP SAMET

Circ Res. 1973;32:393-401
doi: 10.1161/01.RES.32.3.393

Circulation Research is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1973 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circres.ahajournals.org/content/32/3/393

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation Research can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation Research is online at:
http://circres.ahajournals.org/subscriptions/