Effects of Propranolol on the Impulse Activity of Cardiovascular Sympathetic Afferent Fibers

Federico Lombardi, Carlo Casalone, Gabriella Malfatto, Tomaso Gnecci Ruscone, Rodolfo Casati, and Alberto Malliani

SUMMARY  The influence of β-adrenergic receptor blockade on the impulse activity of 21 cardiovascular sympathetic afferent nerve fibers (11 from the thoracic aorta, 10 from the pulmonary veins), isolated from the left sympathetic rami communicantes T-3 and T-4 was studied in anesthetized, vagotomized cats. Aortic pressure, heart rate, and neural discharge were recorded during control conditions and during brief aortic occlusions of comparable amplitude and duration. Administration of dl-propranolol (0.2–0.4 mg/kg) did not modify aortic pressure or neural discharge of the fibers during control conditions, although, as expected, heart rate was diminished. dl-Propranolol administration did change the response of cardiovascular sympathetic afferents to similar aortic pressure increases. Before drug administration, aortic occlusion caused a significant increase in neural discharge of both aortic and pulmonary vein sympathetic afferent fibers, from 0.52 ± 0.12 to 1.64 ± 0.31 and from 0.67 ± 0.10 to 2.08 ± 0.25 impulses/sec, respectively (p < 0.05). After dl-propranolol administration, comparable increases in aortic pressure resulted in slight but not significant increases in neural discharge of aortic and pulmonary vein fibers. Administration of d-propranolol (0.4–0.6 mg/kg), which possesses only membrane-stabilizing properties, did not modify the firing rate of four pulmonary sympathetic afferents, which subsequently decreased their response to pressure rises after administration of dl-propranolol. These results indicate that β-adrenergic receptor blockade reduces the responsiveness to hemodynamic stimuli of sympathetic cardiovascular afferent fibers that are capable of mediating excitatory pressor reflexes. (Hypertension 8: 50–55, 1986).

KEY WORDS  • β-blockers • arterial hypertension • cardiovascular sympathetic afferent fibers • positive feedback reflexes

It has been more than 20 years since Prichard et al.1 noticed that pronethalol, a β-adrenergic receptor blocking agent, was capable of lowering the arterial pressure in hypertensive patients. The serendipity of this observation can still be appreciated since, despite the proliferation and increasing popularity of β-blockers, the mechanism of their antihypertensive action remains uncertain.

Various hypotheses advanced to shed light on the mechanism of action of β-blockers have focused on the role of renin,1,2 baroreceptors,4,5 and blockade of prejunctional β-adrenergic receptors6,7 or have suggested a central nervous system site of action.5,8,9

FURTHER

From the Istituto Ricerche Cardiovascolari, CNR, Patologia Medica, Ospedale di L. Sacco, Università di Milano, Milan (F. Lombardi, C. Casalone, G. Malfatto, R. Canuti, and A. Malliani), and the Servizio di Cardiologia, Ospedale di Merate, Como (T. Gnecci Ruscone), Italy.

Address for reprints: Prof. Alberto Malliani. Istituto Ricerche Cardiovascolari, via Bonfadini 214, 20138 Milano, Italia.

Received November 30, 1984; accepted June 12, 1985.

more, the progressive decrease in total peripheral resistance that occurs despite reduced cardiac output as a result of chronic administration of β-blockers has not been explained adequately.10

Lewis11 and Vaughan Williams12 emphasized that a determinant component of the antihypertensive effects of β-blockers might be related to a diminished response of the heart to various excitatory stimuli. They hypothesized that an attenuation of the sensory input from the cardiovascular system to the central nervous structures eventually could result in a reduction of the sympathetic outflow distributed back to the cardiovascular system. The recent description of excitatory cardiovascular reflexes, mediated by sympathetic afferent fibers13,14 and exhibiting positive feedback characteristics,15-16 seems to offer the functional substratum to this appealing hypothesis.

The present study attempted to determine the effects of dl-propranolol on the impulse activity of aortic and pulmonary vein sympathetic afferent fibers (i.e., a
population of fibers possessing mechanical sensitivity and capable of mediating excitatory sympathetic reflexes).

Materials and Methods

Twenty-six adult cats (weight, 2.5—4 kg) were anesthetized by an intramuscular injection of ketamine (30 mg/kg) followed by administration of chloralose (70 mg/kg, i.v.). The trachea was cannulated, and both vagi were isolated and cut in the neck. Polyethylene catheters were inserted into 1) the thoracic aorta through a femoral artery, 2) the inferior vena cava through a femoral vein, and 3) in five cats, the left atrium through the atrial appendage. The animals were paralyzed with gallamine triethiodide (Flaxedil), 2 mg/kg, and artificially ventilated. The respirator was adjusted to maintain blood gases within physiological limits. All cats were placed on their right side, and the chest was opened by removing the fourth to seventh ribs. The heads of the second and the third ribs were removed retropleurally on the left side to expose the stellate ganglion and its branches. Threads were passed around the distal portion of the descending thoracic aorta and the inferior vena cava. All of these ligatures were then passed through short, rigid polyethylene tubings so that vascular occlusions could be induced by pulling the appropriate ligature.

Variables Recorded

Afferent neural impulse activity was recorded in 21 cats from filaments isolated under a dissecting microscope from the cut peripheral end of the third and fourth left thoracic sympathetic rami communicantes. Filaments were split until discharge from a single, or clearly detectable, active unit was present. The details for recording neural activity of sympathetic fibers with aortic and pulmonary sensory endings have been published previously.17,18 A digital neuronal spike analyzer (custom-made by Dr. D. Malagodi, Milan, Italy) was used to obtain an electrical signal proportional to the number of neural impulses over time and to compute the mean frequency of discharge.19

Arterial pressure and left atrial pressure were measured with a Statham P23De strain gauge; the cathetermanometer system had a flat (±5%) frequency response of 30 Hz, as calculated by its response to a stepwise input of pressure.20 At variance with previous studies,17 left atrial pressure was only measured in cats in which neural recordings were not obtained, in order to prevent any possible damage to the fibers in passage or to the sensory fields located in the pulmonary veins, thus affecting the impulse activity.

Location of the Receptor Endings

A preliminary, approximate localization of the endings was performed at the beginning of the recording phase by extremely light mechanical probing of the aortic wall or the pulmonary veins with a cotton wool ball. An accurate and definite localization was obtained at the end of the experiment by repeating the mechanical probing after the animal had been killed. The destruction of the aortic or pulmonary vein endings caused the disappearance of impulses in the fibers studied.17,18 As myelinated and unmyelinated afferent sympathetic aortic nerve fibers display a similar responsiveness to hemodynamic stimuli,19 the conduction velocity was not determined in these experiments.

Protocol

Afferent sympathetic impulse activity was assessed during control conditions after the preliminary localization of the receptive field. The mechanical properties of these afferent fibers were assessed by their response to increases in aortic pressure produced by distal thoracic aortic occlusion for 47 ± 7 seconds. The effects observed during the initial 8 ± 2 seconds of occlusion (i.e., the rising phase) will be referred to as early effects; those occurring during the subsequent 39 ± 5 seconds (i.e., the plateau of the raised pressure) will be referred to as late effects.

After two or three control aortic occlusions, dl-propranolol (Inderal), 0.2 to 0.4 mg/kg, was injected intravenously over 5 minutes. Impulse activity was then studied during control conditions and in response to occlusion of the thoracic aorta, which produced changes in arterial blood pressure of the same magnitude as those induced before dl-propranolol administration.

The adequacy of β-blockade was assessed in eight animals by comparing the heart rate changes induced by the intravenous administration of isoproterenol (2 μg/kg) before and after dl-propranolol administration.

The relative contribution of the membrane-stabilizing properties of dl-propranolol was assessed in four cats by administering d-propranolol (0.4—0.6 mg/kg), which possess only membrane-stabilizing properties, after control occlusions. Therefore, four pulmonary vein sympathetic afferents were studied under three different experimental conditions: control, after d-propranolol, and after dl-propranolol.

Statistical Analysis

The firing rate of aortic and pulmonary sympathetic fibers is expressed in impulses per second and in impulses per beat, which accounts for the effect of the dl-propranolol-induced bradycardia on the absolute firing rate of the units.

Results are reported as means ± SEM. Multiple comparisons were performed by analysis of variance using the Scheffe method.21 A value of p less than 0.05 was considered significant.

Results

We studied the impulse activity of 21 afferent sympathetic fibers, the endings of which were excited by direct mechanical probing of the thoracic aorta (11 fibers) or one of the pulmonary veins (10 fibers). Of 11 aortic fibers, six had their sensory endings located in the aortic arch and five in the proximal descending tract. Of the 10 fibers from the pulmonary veins, six
had their receptive field located in the vein of the upper lobe and four in that of the central lobe. Aortic sympathetic fibers had a spontaneous impulse activity of 0.52 ± 0.12 impulses/sec at a systolic arterial blood pressure of 130 ± 8 mm Hg and a heart rate of 164 ± 10 beats/min. Pulmonary vein fibers had a spontaneous impulse activity of 0.67 ± 0.10 impulses/sec at a systolic arterial pressure of 135 ± 7 mm Hg and a heart rate of 162 ± 7 beats/min.

Responses to Aortic Occlusion of Aortic Sympathetic Afferents

Occlusion of the thoracic aorta resulted in a significant increase in aortic pressure that was accompanied by a marked augmentation of neural discharge (Table 1). In particular, the impulse activity increased up to 1.97 ± 0.36 and to 1.64 ± 0.31 impulses/sec during, respectively, the early and late phase of the occlusion (Figure 1A; see Table 1). A significant excitation during aortic occlusion also was evident when the firing rate of the fibers was expressed in impulses per beat (see Table 1).

*d/-Propranolol did not significantly modify the control impulse activity of afferent sympathetic aortic fibers (0.52 ± 0.12 vs 0.37 ± 0.09 impulses/sec); however, it produced the expected, marked reduction of heart rate (from 164 ± 10 to 128 ± 8 beats/min). Occlusion of the thoracic aorta after *β*-blockade, which produced a similar arterial pressure rise, resulted in a small but not significant increase in sympathetic afferent discharge during the early and late phases of occlusion (Figure 1B; see Table 1). The attenuation of the responsiveness of afferent sympathetic fibers to similar aortic pressure increases also was significant when the firing rate was expressed in impulses per beat (see Table 1).

Responses to Aortic Occlusion of Pulmonary Vein Sympathetic Afferents

Aortic occlusion resulted in a significant increase in the firing rate of the fibers from the pulmonary veins during both the early and late phase of occlusion. In particular, neural impulse activity increased to 2.54 ± 0.38 and 2.08 ± 0.25 impulses/sec, respectively, together with a significant increase in aortic blood pressure (Figure 2A; Table 2). *d/-Propranolol did not modify significantly the resting impulse activity of pulmonary vein sympathetic afferents (0.67 ± 0.10 vs 0.43 ± 0.09 impulses/sec); however, it resulted in a significant reduction in heart rate. After *β*-blockade, aortic occlusion induced a small but not significant

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control</th>
<th>AO early phase</th>
<th>AO late phase</th>
</tr>
</thead>
<tbody>
<tr>
<td>SAP (mm Hg)</td>
<td>130 ± 8</td>
<td>185 ± 9*</td>
<td>191 ± 10*</td>
</tr>
<tr>
<td>DAP (mm Hg)</td>
<td>99 ± 11</td>
<td>138 ± 7*</td>
<td>133 ± 14*</td>
</tr>
<tr>
<td>Heart rate</td>
<td>164 ± 10</td>
<td>164 ± 9</td>
<td>154 ± 10</td>
</tr>
<tr>
<td>Neural discharge</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Impulses/sec</td>
<td>0.52 ± 0.12</td>
<td>1.97 ± 0.36*</td>
<td>1.64 ± 0.31*</td>
</tr>
<tr>
<td>Impulses/beat</td>
<td>0.20 ± 0.05</td>
<td>0.74 ± 0.14*</td>
<td>0.78 ± 0.18*</td>
</tr>
<tr>
<td>Duration (sec)</td>
<td>31 ± 3</td>
<td>8 ± 2</td>
<td>39 ± 5</td>
</tr>
</tbody>
</table>

Values are means ± SEM. AO = aortic occlusion, SAP = systolic arterial pressure, DAP = diastolic arterial pressure, *p < 0.05, compared with control, †p < 0.05, compared with response obtained before *β*-blockade
increase in neural discharge during comparable aortic pressure rises (see Table 2). As observed for aortic sympathetic afferents, these responses were significantly different from those observed before drug administration (see Figure 2 and Table 2).

d-Propranolol was administered in four cats after control occlusions and did not modify systolic arterial pressure, heart rate, or neural discharge of four pulmonary vein sympathetic fibers, which subsequently reduced their responsiveness to pressure rises of similar amplitude with dl-propranolol administration (Table 3).

Effects of β-Blockade on Arterial–Left Atrial Pressure Relationship

The effects of dl-propranolol on the arterial–left atrial pressure relationship was also determined in five additional cats. β-Blockade did not modify resting mean left atrial pressure (4.9 ± 0.6 vs 4.8 ± 0.7 cm H₂O respectively) or systolic arterial pressure (128 ± 6 vs 123 ± 7 mm Hg). After dl-propranolol administration, however, there was a greater increase in left atrial pressure (7.1 ± 0.7 vs 9.2 ± 1.4 cm H₂O; p < 0.05) in response to arterial pressure rises of similar amplitude.

Discussion

Our results indicate that dl-propranolol can modify the responsiveness of cardiovascular sympathetic afferent fibers to similar increases in arterial blood pressure and therefore seems to be capable of affecting the sympathetic input to the spinal cord.

The general property of cardiovascular sympathetic afferent fibers to yield only a few impulses per cardiac cycle, as compared to the bursts of impulses characterizing, for example, vagal afferent fibers, limits the possibility of investigating the “static” and “dynamic” components of the receptors’ transducing properties. However, the analysis performed in the present study adequately emphasized that after dl-propranolol administration the aortic fibers exhibited markedly reduced responses to similar increases in systolic aortic pressure.

In this context it is important to remember that in anesthetized cats a graded distention of a tract of the descending thoracic aorta, a stimulus that mimics an increase in aortic pressure, produced a reflex increase in arterial pressure, heart rate, and the first derivative of left ventricular pressure. These animals had the baroreceptor afferents and the vagi cut, and the reflex was mediated by aortic sympathetic afferent fibers. In conscious dogs, moreover, it was additionally

---

**Table 2. Effects of dl-Propranolol on Arterial Pressure, Heart Rate, and Neural Discharge of 10 Pulmonary Vein Sympathetic Afferents During Control, Early, and Late Phase of Aortic Occlusion**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Before β-blockade</th>
<th>After β-blockade</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>AO early phase</td>
</tr>
<tr>
<td>SAP (mm Hg)</td>
<td>135 ± 7</td>
<td>179 ± 15*</td>
</tr>
<tr>
<td>DAP (mm Hg)</td>
<td>102 ± 8</td>
<td>140 ± 7*</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>162 ± 7</td>
<td>162 ± 6</td>
</tr>
<tr>
<td>Neural discharge</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Impulses/sec</td>
<td>0.67 ± 0.1</td>
<td>2.54 ± 0.38*</td>
</tr>
<tr>
<td>Impulses/beat</td>
<td>0.26 ± 0.08</td>
<td>0.94 ± 0.16*</td>
</tr>
<tr>
<td>Duration (sec)</td>
<td>30 ± 4</td>
<td>11 ± 2</td>
</tr>
</tbody>
</table>

Values are means ± SEM. See Table 1 for key to abbreviations.

*p < 0.05, compared with control; †p < 0.05, compared with the response obtained before β-blockade.
proved that the same reflex could occur in the presence of an intact cardiovascular innervation. Thus, a similar sympathetic pressure acting on this reflexogenic area would generate a reduced input to the spinal cord and thereby a reduced excitatory reflex after β-blockade.

The results concerning the afferent sympathetic fibers innervating the pulmonary veins can be similarly interpreted. Previous studies reported that these mechanoreceptors, located in a low pressure area, were extremely sensitive to changes in atrial pressure.24,25 They were similarly excited during increases in aortic pressure, due to a concomitant rise in left atrial pressure. The reduction in firing rate of these afferent fibers appears to be unrelated to a change in the absolute level of left atrial pressure induced by aortic occlusion, as β-blockade caused even greater increases in mean left atrial pressure during similar arterial pressure rises. These findings suggest that the reduction of impulse activity of sympathetic afferents from pulmonary veins reflects an alteration of the fine characteristics of the mechanical stimulus induced by dl-propranolol.

Uchida and Murao27 reported that β^-propranolol reduces the activation of myelinated and unmyelinated left ventricular sympathetic afferent fibers during experimental coronary artery occlusion. Our observations indicate that β-blockade also may affect the mechanosensitivity of sympathetic afferent fibers with vascular endings.

The main purpose of our experiments was to prove that a β-blocking agent (and in this regard we selected the most widely used drug) could decrease the afferent impulse traffic from the cardiovascular system to the spinal cord through the thoracic sympathetic afferents. Although this phenomenon was not apparent from the background impulse activity of the fibers, it was easily detectable when the mechanical responsiveness of the fibers was tested with rises in pressure.

The observed reduction in the responsiveness of sympathetic afferents to mechanical stimuli could be either the result of an alteration of the hemodynamic stimulus or of a direct effect of β-adrenergic blockade on the elastic properties of the vessels. It is well known that arterial elastic stiffness can be influenced by an adrenergic activation in both anesthetized28 and conscious29 animals and that changes in the mechanical properties of the arterial wall can affect the transducer properties of the nerve endings.30,31 The present experiments, however, cannot give a definitive answer to this question, as we did not measure pressure-diameter relationship while recording neural activity. Reduction in the sensitivity of ventricular unmyelated vagal afferents has been reported to be secondary to the reduction in contractility resulting from β-adrenergic blockade and was dependent on the level of end-diastolic pressure.32 In addition, the alteration of the arterial–left atrial pressure relationship reported in these experiments and the reduction of the amplitude of atrial contraction during aortic constriction reported by Thames32 as a result of β-blockade indicate that propranolol may well affect the fiber responsiveness by changing the hemodynamic stimulus.

In the present experiments, we used dl-propranolol, a racemic mixture that has both β-adrenergic receptor blocking properties and nonspecific membrane-stabilizing effects. However, it is unlikely that our findings were the result of the anesthetic properties of the drug. In fact, administration of d-propranolol, which has little β-adrenergic receptor blocking activity but retains membrane-stabilizing properties, did not modify the resting impulse activity or the neural response of the pulmonary vein sympathetic afferents studied. These findings are consistent with observations by Thames32 that d-propranolol does not modify the mechanosensitivity of ventricular vagal afferent fibers (although it was affected by dl-propranolol).

The relevance of the present study consists in the demonstration that a population of afferent fibers, which can mediate cardiovascular excitatory reflexes with positive feedback characteristics,13,33 displayed a reduced sensitivity during similar rises in systolic arterial pressure. The activation of sympathetic efferent activity reported by Sundlöf et al.34 after short-term administration of metoprolol in hypertensive patients is difficult to explain in the context of our findings.
However, in our experimental model dl-propranolol was only effective in decreasing fiber responsiveness to short-term arterial pressure rises. This apparent discrepancy is probably the result of different experimental models.

The attenuation of the reflex forearm vasoconstriction response to graded lower body negative pressures reported by Ferguson et al. in normal subjects and the propranolol-induced reduction in the pressor response to exercise seen in normotensive subjects are consistent with our experimental findings.

Human arterial hypertension as a disease of regulation may be caused in part by increased sympathetic tone. According to the positive feedback hypothesis, this increase may depend not only on an augmented afferent component of these excitatory reflexes. Our data provide evidence that the administration of dl-propranolol alters the afferent component of these excitatory reflexes.

References

Effects of propranolol on the impulse activity of cardiovascular sympathetic afferent fibers.
F Lombardi, C Casalone, G Malfatto, T Gnecci Ruscone, R Casati and A Malliani

Hypertension. 1986;8:50-55
doi: 10.1161/01.HYP.8.1.50

Hypertension is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1986 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/8/1/50

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Hypertension 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 Hypertension is online at:
http://hyper.ahajournals.org/subscriptions/