Ventricular Function in Cardiac-Denervated and Cardiac-Sympathectomized Conscious Dogs

By Hubert L. Stone, Ph.D., Vernon S. Bishop, Ph.D., and E. Dong, Jr., M.D.

ABSTRACT

The role of the autonomic nervous system in ventricular function was studied in conscious animals when either the cardiac nerves or the sympathetic nerves to the heart had been eliminated. Regional neural ablation was used to denervate the heart, and blockade of the cardiac sympathetic nerves was performed with propranolol. A reduction in the plateau of the ventricular output was seen in the cardiac-denervated animals (283 ml/min per kg) and in animals with beta-blockade (240 ml/min per kg) as compared with 318 ml/min per kg for the control curves. The difference in stroke volume in the cardiac-denervated animals and the beta-blocked animals at the plateau demonstrates that the chronically cardiac-denervated animal changes its ventricular output solely by a change in stroke volume, whereas the animal with beta-blockade changes heart rate and stroke volume in response to stress. Thus the beta-blocked animals increased their heart rate by a decrease in parasympathetic activity.

ADDITIONAL KEY WORDS ventricular output curves cardiac function stroke volume curves cardiac dynamics cardiac control

Previous experiments on ventricular function curves in the normal conscious animal have indicated that the level of ventricular output attained by a normal animal is probably related to the existing balance of autonomic nervous system activity. In a group of animals that had right ventricular damage, the plateau level of ventricular output was found to remain within normal limits for 1 to 2 weeks following the production of the myocardial lesion. During this time period, the myocardium was visibly damaged from the procedure used to create the lesion. The only explanation of the ability of the animal to maintain his plateau level of ventricular output under the experimental condition was an increase in the level of the sympathetic nervous system activity. However, the influence of the parasympathetic nervous innervation of the heart cannot be ignored, especially since conflicting data on the effect of the vagus nerves on cardiac dynamics are present in the literature.

To begin to investigate the effect of the autonomic nervous system on ventricular function in conscious animals, two methods were used to eliminate either all of the cardiac nerves or the sympathetic nerves to the heart. Regional neural ablation was used to denervate the heart and we chose to chemically block the cardiac sympathetic nerves by using the beta-adrenergic blocking agent, propranolol. This drug has been shown to effectively block the cardiac response to isoproterenol, except when large amounts of isoproterenol are used. Elimination of all cardiac nerves should allow one to examine the intrinsic cardiac regulation by comparing the denervated heart with the normal heart, while removing one side of the auto-
nomic nervous system gives answers concerning the contribution of this part of the system to ventricular function.

Methods

Nine adult mongrel dogs ranging in weight from 7 to 19 kg were used. Three animals were used for cardiac denervation and 6 animals were used for the beta-adrenergic blockade study.

Cardiac denervation was accomplished by the regional neural ablation technique described by Cooper (8). The animals were anesthetized with sodium pentobarbital (30 mg/kg) and a median sternotomy was made under sterile surgical conditions. The phrenic nerves were dissected free from the pericardium and all vessels entering or leaving the heart were stripped. The pericardium was freed and removed as was most of the tissue within this general area. Following this stripping procedure, a sine-wave electromagnetic flowprobe was positioned around the main pulmonary artery and a small polyvinyl catheter was positioned in the left atrium. The catheter was filled with heparinized saline and closed. The flowprobe leads and the catheter were brought under the skin to the anterior surface of the neck, where they were exteriorized. The incision was closed and the animals were allowed to recover. Following a suitable recovery period, the completeness of cardiac denervation was ascertained by measuring the heart rate response to injections of 4 mg atropine, 60 μg/kg tyramine, and 4 μg norepinephrine. There was no heart rate response to the atropine injection and essentially no response to the tyramine injection. However, the injection of norepinephrine produced a tachycardia.

The 6 animals used in the beta-adrenergic blockade experiments were prepared with electromagnetic flowmeter probe and left atrial catheter in similar manner.

During the 2- to 3-week recovery period, all animals were again anesthetized; a small polyvinyl catheter was placed in the right atrium through the right jugular vein and a larger polyvinyl catheter was placed in the left jugular vein. These two catheters were again brought through the skin on the anterior neck surface. All catheters were maintained patent by periodic flushing with heparinized saline solution.

Ventricular function curves were determined as previously described (1). This method uses the rapid infusion of Tyrode's solution into the left jugular vein catheter in the conscious animal while measuring mean femoral arterial pressure, mean right and left atrial pressures, heart rate, mean ventricular output, and pulsatile pulmonary artery flow. All pressures were registered with Statham P23 series transducers zeroed to the midsternal line with the animal reclining on the right side; the recordings were made with an eight-channel Offner recorder. All flowmeter probes were calibrated prior to implantation and were checked upon recovery from the animal (1). The level of flow in late diastole was used as a zero measurement on the electromagnetic flowmeter (Medicon Model K 2000). At least three ventricular function curves were determined in each of the 9 animals with no more than three curves determined in any 7-day period. A total of 11 ventricular function curves were obtained in the 3 cardiac-denervated animals. In the 6 normal animals, a total of 18 control ventricular function curves were determined.

After a minimum of three control ventricular function curves were obtained in the 6 normal animals, they were treated with the beta-adrenergic blocking drug, propranolol, in a dose of 1 mg/kg body weight. The drug was dissolved in normal saline and given slowly intravenously. A ventricular function curve was obtained 20 min after the completion of drug administration. Following determination of several ventricular output curves using propranolol, another control curve was determined and compared with the previous control curves obtained in the same animals. Twenty-one ventricular function curves were determined after beta-adrenergic blockade.

Results

In the 21 determinations of ventricular function following beta-adrenergic blockade, the average values recorded for mean arterial pressure, mean right and left atrial pressures, heart rate, mean cardiac output, and stroke volume prior to giving propranolol were compared with the same values recorded 20 min after beta-blockade. These values can be seen in Table 1. There was no particular change in any of the resting values following beta-adrenergic blockade when compared with the pretreatment values except a slight rise in mean left atrial pressure. The mean left atrial pressure rose from an average of 2.4 mm Hg to an average of 4.9 mm Hg 20 min after beta-adrenergic blockade. The initial mean arterial pressure recorded prior to the determination of ventricular function in the normal dogs averaged 114 mm Hg (±4 mm Hg SEM) and rose an average of 16% at the plateau level of cardiac output. Following beta-adrenergic blockade, the mean arterial pressure at
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Average Resting Values before and 20 Min after Propranolol Treatment

<table>
<thead>
<tr>
<th></th>
<th>MAP (mm Hg)</th>
<th>RAP (mm Hg)</th>
<th>LAP (mm Hg)</th>
<th>H.R. (beat/min)</th>
<th>C.O. (ml/min per kg)</th>
<th>S.V. (ml/beat per kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before</td>
<td>114 ± 4</td>
<td>1.2 ± 0.4</td>
<td>2.4 ± 0.5</td>
<td>101 ± 4</td>
<td>130 ± 6</td>
<td>1.3 ± 0.06</td>
</tr>
<tr>
<td>After</td>
<td>119 ± 4</td>
<td>1.3 ± 0.4</td>
<td>4.9 ± 0.7*</td>
<td>95 ± 4</td>
<td>129 ± 6</td>
<td>1.3 ± 0.06</td>
</tr>
</tbody>
</table>

Mean arterial pressure (MAP), mean right atrial pressure (RAP), mean left atrial pressure (LAP), heart rate (H.R.), mean cardiac output (C.O.), and stroke volume (S.V.). Propranolol was given in a dose of 1 mg/kg body weight. The values are mean and standard error of the mean.

*Significantly different with a probability value of less than .01.

the beginning of the determination of ventricular function averaged 119 mm Hg (±4 mm Hg SEM) and rose an average of 15% at the plateau level of cardiac output.

VENTRICULAR OUTPUT CURVES

Ventricular output curves were determined by the rapid infusion of Tyrode's solution into the right side of the heart. The infusion was continued until a point was reached where the atrial pressures continued to rise and the cardiac output had attained a constant level. The changes in ventricular output, heart rate, and stroke volume were then plotted against the mean right and left atrial pressures at the various points along the curve.

Figure 1 shows the average ventricular output curves for the control and beta-adrenergic blocked animals plotted against mean right and left atrial pressures. In the control experiments, the ventricular output rises rather rapidly to an average plateau value of 318 ml/min per kg (±13 ml/min per kg SEM). In previous work (1) it was demonstrated that in a single animal the plateau level of ventricular output remained fairly constant from one determination to another and was reproducible several weeks apart. Following beta-adrenergic blockade, the average curve for all of the animals approached the plateau value of ventricular output more slowly than during the control period. The average plateau value of ventricular output was also found to be reduced to a value of 240 ml/min per kg (±11 ml/min per kg SEM). The reduction in plateau level of ventricular output was present in all animals studied following blockade. Statistical comparison of the normal versus the beta-blocked values for plateau ventricular output shows this to be a highly significant reduction in ventricular function (P values less than .001).

The curve in Figure 1 labeled with a D represents the average ventricular output curve obtained from the 3 cardiac-denervated animals. It should be noticed that this average curve again rises rather slowly to the plateau of ventricular output. The plateau of ventricular output in this group of animals averaged 263 ml/min per kg (±15 ml/min per kg SEM). A statistical comparison between the average plateau of ventricular output of the cardiac-denervated animal and the group of control animals showed a significant difference (P values between .02 and .01), but there was no significant difference between the cardiac-denervated and the beta-blocked groups.

HEART RATE AND STROKE VOLUME

Figure 2 shows the average heart rate plotted against mean right and left atrial pressures during the determination of ventricular output curves. The average heart rate in the control group rose to a plateau level of 158 beat/min (±5 SEM) during experiments, but following beta-blockade it rose to a value of only 130 beat/min (±4 SEM). The reduction in peak heart rate following blockade was found to be significantly different from the control peak heart rate value (P value less than .001).

The remaining curve in Figure 2 represents the average heart rate response of the cardiac-denervated group of animals. The average initial heart rate in this group was 118 beat/min (±11 SEM) and rose to an average
FIGURE 1
Average mean ventricular output of 6 animals plotted against mean right and left atrial pressures. The bars through the points represent 1 standard error of the mean. The symbols C, D, and BB stand for control, denervated, and beta blockade, respectively.

FIGURE 2
Average heart rate of 6 animals plotted against mean right and left atrial pressures. The bars through the points represent 1 standard error of the mean. Symbols are the same as those in Figure 1.

FIGURE 3
Average stroke volume of 6 animals plotted against mean right and left atrial pressures. The bars through the points represent 1 standard error of the mean. Symbols are the same as those in Figure 1.
value of only 126 beat/min (±11 SEM) at the plateau of the ventricular output curve. There was a significant difference between this peak heart rate and that of the group of control animals (P value between .01 and .001). When the average peak heart rate of cardiac-denervated animals was compared with that of beta-blocked animals, no difference was found. However, one major difference should be noticed in Figure 2 concerning the manner in which these two groups attained the peak heart rate. The average heart rate of the cardiac-denervated group hardly changed at all during the determination of the ventricular function curve; there was a change in the beta-blocked group similar to the normal, but with a reduced peak heart rate.

The average values for the stroke volume response can be seen in Figure 3. In this figure the changes in stroke volume are plotted against both mean right and left atrial pressures for the control experiments and for the beta-adrenergic blockade experiments. During the control experiments, the average stroke volume rose to a value of 1.9 ml/beat per kg (±0.09 SEM) at the plateau level of ventricular output. Following beta blockade, the maximum average stroke volume was 1.7 ml/beat per kg (±0.09 SEM). The change in the stroke volume observed at the plateau level of ventricular output following blockade was found not to be statistically different from the control experiments.

In Figure 3 the stroke volume response of the cardiac-denervated animals is labeled with the letter D. It should be noticed that the maximum stroke volume for this group of animals was higher than both the control and beta-blocked groups. The average stroke volume at the plateau of ventricular output was 2.2 ml/beat per kg (±0.18 SEM). A statistical difference was found in the comparison of the cardiac-denervated and beta-blocked groups (P value between .02 and .01), but not between the control and cardiac-denervated groups. The major difference between the cardiac-denervated and the beta-blocked groups is that the cardiac-denervated animals had a higher plateau value for ventricular output at a lower heart rate.

**Discussion**

In the present series of experiments, in a comparison of the cardiovascular parameters measured before and 20 min following propranolol injection, there was no change in mean arterial pressure, right atrial pressure, cardiac output, heart rate or stroke volume. These results are in agreement with those of Schroder and Werkö (3) but do not agree with the data of others (10), who found changes in resting heart rate and cardiac output following beta blockade. A small rise in mean left atrial pressure was noted, as has been observed by Murray and associates (11), but the increase was only 2.5 mm Hg, which represents a minor change.

The major finding in this study was the reduction in the plateau level of ventricular output following beta blockade. In the normal animal, the plateau of the ventricular output curve represents some balance between the autonomic nervous system activity (namely, the heart rate response) and the stroke volume output. The activity of both parts of the autonomic nervous system can certainly be modulated by many reflexes such as those originating from the cardiac and the carotid sinus regions. As the venous pressure was increased, the heart rate also increased, but the level of increase was different for the control and beta-blocked animals, as can be seen in Figure 2. The only means by which the beta-blocked animal can increase the heart rate is by reducing the activity of the parasympathetic nervous system. Therefore, the reduction in heart rate seems to determine the level of the ventricular output curve under the conditions of these experiments. This is emphasized by referring to Figure 3, in which it will be noticed that the peak stroke volume was hardly changed after beta blockade as compared with the normal response.

Ventricular work was not shown in these experiments. However, it can be stated, as has been done previously (1), that the work will depend largely on the level of ventricular output and heart rate attained during the
course of the experiment. Since the ventricular output curve was reduced, one would expect the ventricular minute work curve to be depressed. This is the case, since no difference in arterial pressure response was found. However, the stroke work curve was almost identical since there was no difference in the stroke volume during the control and beta-blocked periods. The small shift in the initial mean left atrial pressure did not produce any significant shift along the pressure axis in the average curves as has been reported (12).

Therefore, the major effect of beta blockade is the same as stated previously, namely, reduction in the plateau levels of ventricular output and heart rate.

The comparison of the present series of experiments with the results obtained under identical conditions in cardiac-denervated animals shows three important features. The plateau value of ventricular output attained in the denervated animal and beta-blocked animal is approximately the same, as is the heart rate response. However, one major difference exists in the heart rate response, as should be noticed in Figure 2. In the denervated animal, the heart rate essentially does not change during the determination of ventricular function, but in the animals with beta blockade the heart rate does increase from its initial value and reaches approximately the same level as that found in the denervated animals. The increase in heart rate observed in the beta-blocked animals must result from a decrease in the parasympathetic nervous system activity or the heart rate would not have changed at all. The difference in stroke volume in the cardiac-denervated animals and in the beta-blocked animals demonstrates that the chronically cardiac-denervated animal changes its ventricular output solely by a change in stroke volume, whereas the animal with partial nervous innervation changes the heart rate in response to cardiac stress and therefore changes the stroke volume to a lesser degree than the chronically denervated animal. The difference in the heart rate and stroke volume responses of these two groups of animals to identical conditions emphasizes the contribution of the parasympathetic nervous system to normal ventricular function, namely, by maintaining some regulation of stroke volume and heart rate during cardiac stress. Therefore, in the normal animal, the heart rate is changed by the interaction of the two systems by having a decrease in parasympathetic activity as well as an increase in the sympathetic activity.

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References
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