Altered Control of Hindlimb Vascular Resistance by Vagal Afferents in Spontaneously Hypertensive Rats
Difference in The Early and Late Stage of Hypertension

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SUMMARY. The aim of this study was to examine whether control of vascular resistance by vagal afferents is altered in the early as well as late stage of hypertension. We examined the effects of vagotomy on hindlimb vascular resistance as well as on arterial baroreflex control of hindlimb vascular resistance in spontaneously hypertensive rats and Wistar-Kyoto rats, 12 and 35 weeks old. Vagotomy in rats with the intact arterial baroreceptors increased hindlimb vascular resistance in all four groups. Hindlimb vascular responses to graded sympathetic nerve stimulation were closely linear up to 6 Hz in all groups, and the slope of the response was approximately 1.4 times steeper in spontaneously hypertensive than in Wistar-Kyoto rats, in both the young and old groups. The increase in hindlimb vascular resistance after vagotomy normalized by the slope of the response to sympathetic nerve stimulation was greater in young spontaneously hypertensive rats than in young Wistar-Kyoto rats, but was less in old spontaneously hypertensive rats than in old Wistar-Kyoto rats. Vagotomy increased the gain of arterial baroreflex control of hindlimb vascular resistance in young spontaneously hypertensive rats, and in two groups of Wistar-Kyoto rats, but not in old spontaneously hypertensive rats. The percent increase in the gain of arterial baroreflex control of hindlimb vascular resistance after vagotomy tended to be greater in young spontaneously hypertensive rats than in young Wistar-Kyoto rats, but was less in old spontaneously hypertensive than in old Wistar-Kyoto rats. Central venous pressure and left ventricular end-diastolic pressure were higher in spontaneously hypertensive than in Wistar-Kyoto rats in both young and old groups. Left atrial distensibility assessed by obtaining the atrial pressure-volume relationship was comparable between young spontaneously hypertensive and young Wistar-Kyoto rats, but was less in old spontaneously hypertensive than in old Wistar-Kyoto rats. These results indicate that vagal afferents exert tonic inhibition on control of hindlimb vascular resistance in spontaneously hypertensive as well as in Wistar-Kyoto rats, and that tonic inhibitory influence of vagal afferents on control of hindlimb vascular resistance is altered in spontaneously hypertensive rats, augmented in young, but attenuated in old, spontaneously hypertensive rats, compared with that in age-matched Wistar-Kyoto rats. It is considered that altered control of vascular resistance by vagal afferents in spontaneously hypertensive rats may result from changes in vagal afferent activity. (Circ Res 55: 763-772, 1984)

IT is suggested that reflex originating from the cardiopulmonary receptors contributes to altered control of vascular resistance in acute myocardial ischemia (Toubes and Brody, 1970; Felder and Thames, 1979; Abboud et al., 1981), left ventricular outflow obstruction (Mark et al., 1973a, 1973b), or during salt depletion (Takishita and Ferrario, 1982). However, the role of vagal afferents from the cardiopulmonary receptors in control of vascular resistance in hypertension is not well understood.

In hypertension, there are several known changes that might alter the activity of vagal afferents. First, there is an elevation of atrial pressure in hypertension (Noresson et al., 1979) which stimulates the cardiopulmonary receptors that mostly localize in the atrium in rats (Thoren et al., 1979a). On the other hand, recent studies (Ricksten et al., 1979; Thoren et al., 1979b) indicate that the cardiopulmonary receptors as well as the arterial baroreceptors are reset in the established stage of hypertension in spontaneously hypertensive rats, which may offset atrial hypertension. However, it is not known when, in the time course of arterial hypertension, resetting of the cardiopulmonary receptors or atrial hypertension may develop. Atrial hypertension may be caused, in part, by decreased venous distensibility (Haraldsson et al., 1981; Nilsson et al., 1981) which may be present in the early stage of hypertension (Takeshita and Mark, 1979). Thus, it may be possible that atrial hypertension is present before resetting of the cardiopulmonary receptors develops. Second, there may be impaired arterial baroreflex in hyper-
tension (Alexander and DeCuir, 1966; Brown et al., 1976), which may augment the effects of vagal afferents on control of sympathetic outflow. It is reported that the inhibitory influence of the cardiopulmonary receptors on sympathetic outflow is increased when the inhibitory input from the arterial baroreceptors is reduced (Koike et al., 1975; Mancia et al., 1976). Third, changes in the central nervous system in hypertension (Bunag et al., 1975, 1976; Gonzalez et al., 1983) might alter reflex control of vascular resistance by vagal afferents, as well as by the arterial baroreceptors. It may be considered that the overall consequence of these various factors would determine the effects of vagal afferents on control of vascular resistance in hypertension.

Although it is reported that the cardiopulmonary receptors are reset in the established stage of hypertension in rats (Ricksten et al., 1979; Thoren et al., 1979a, 1979b), a recent study by Mark and Kerber (1982) suggests that, in young men with borderline hypertension, cardiopulmonary baroreflex control of forearm vascular resistance is augmented. It is possible that the effects of vagal afferents on control of vascular resistance are different in the stage of hypertension.

The purpose of this study was to examine the role of vagal afferents in control of hindlimb vascular resistance in the early as well as late stage of hypertension. We examined the effects of vagotomy on hindlimb vascular resistance as well as on arterial baroreflex control of hindlimb vascular resistance in spontaneously hypertensive rats (SHR) and Wistar-Kyoto rats (WKY). 12 and 35 weeks old. We also examined atrial distensibility, central venous pressure, and left ventricular end-diastolic pressure in these rats, since these variables are important determinants of cardiac receptor activation and, thus, the activity of vagal afferents from the cardiopulmonary receptors.

Methods

General Methods

Rats used in this study were bred in our laboratory. Four groups of male rats were studied: young (12 ± 1 weeks old, weight 250 ± 10 g) and old (35 ± 1 weeks old, weight 345 ± 5) SHR and age-matched young (12 ± 1 weeks old, weight 291 ± 7 g) and old (35 ± 1 weeks old, weight 421 ± 10 g) WKY. After weaning, rats were raised on regular laboratory chow (CRC-4, Oriental Yeast Co., Ltd.).

Rats were anesthetized with urethane, 1 g/kg, given intraperitoneally. A tracheal cannula was inserted to facilitate spontaneous respiration with a mixture of oxygen and room air. A cannula was positioned in the left axillary artery for continuous recording of systemic arterial pressure. Another cannula was inserted in the left iliac artery via the left femoral artery for the measurement of hindlimb perfusion pressure. A third cannula was positioned in the left femoral vein which was used for drug injection. Systemic arterial and hindlimb perfusion pressures were measured with Statham pressure transducers (P23ID) and recorded on a Nihonkoden Medical Recorder. \( P_{ao2}, P_{aco2}, \) and \( pH \) were measured periodically, and it was confirmed that \( P_{ao2} \) was above 200 mm Hg and \( pH \) was between 7.30 and 7.45. The experiments were done with rats placed on a heating pad to maintain body temperature around 37°C.

Hindlimb Perfusion

The effects of vagal afferents on control of vascular resistance were examined in the vascularly isolated right hindlimb perfused at a constant flow. The abdominal aorta was exposed through a midline incision. After sodium heparin (500 U/kg) was given, the aorta was ligated distal to the renal arteries and two cannulas were inserted. Blood from the proximal aorta was pumped at a constant flow into the distal aorta perfusing the right hindlimb, using a perfusion pump (Cole-Parmer Masterflex, model 7013). To examine arterial baroreflex control of hindlimb vascular resistance, we included a delay system (20-30 seconds) in the extracorporeal perfusion circuit, which prevented the direct vascular effects of drugs modifying vascular resistance. Changes in vascular resistance (this method was adopted from that of Guo, Thames, and Abboud) (Guo et al., 1982). The delay system consisted of a polyethylene coil which was immersed in a thermoregulated bath. The temperature of the bath was maintained at 37°C. While the delay circuit was filled with approximately 3 ml of blood withdrawn from the aorta, the equivalent volume of blood obtained from a donor rat was intravenously infused. Blood flow to the hindlimb was adjusted initially so that the perfusion pressure closely approximated the existing systemic arterial pressure. With this delay system, the maximal reflex change in hindlimb perfusion pressure occurred before an intravenously injected drug reached the perfused hindlimb (Fig. 1). At constant flow, changes in perfusion pressure reflected changes in vascular resistance.

Effects of Vagotomy on Control of Hindlimb Vascular Resistance

Through a midline incision in the neck, bilateral vagal nerves were exposed with a dissecting microscope and looped with fine threads for easy access for subsequent section. Great care was taken not to injure the vagal as well as adjacent nerves. Atropine (0.2 mg/kg, iv) was administered before the protocols were begun, to eliminate the influence of vagal efferents on the heart.

After completion of surgery, a period of 20-30 minutes was allowed for stabilization before beginning the protocols. To evaluate the role of vagal afferents in control of hindlimb vascular resistance, we measured hindlimb perfusion pressure, and the gain of arterial baroreflex control of hindlimb vascular resistance before and after vagotomy in atropinized rats. The latter measurements were done 10 minutes after vagotomy.

The following additional studies were done. First, we examined hindlimb perfusion pressure and the gain of arterial baroreflex control of hindlimb vascular resistance before and 15 minutes after sham vagotomy in young SHR. Second, to determine whether changes in hindlimb perfusion pressure after vagotomy were neurally mediated, we examined the effects of vagotomy on hindlimb perfusion pressure in young SHR in which lumbar sympathetic nerves were cut. Third, we examined whether intravenous phenylephrine produces reflex hindlimb vasoconstriction after bilateral vasodilation occurred in response to intravenous phenylephrine in these rats (n = 5), which
indicates that aortic depressor nerves remained intact after vagotomy. Before carotid sinus denervation, carotid artery occlusion produced reflex hindlimb vasoconstriction which was absent after carotid sinus denervation.

**Arterial Baroreflex Control of Hindlimb Vascular Resistance**

To examine arterial baroreflex control of hindlimb vascular resistance, we injected two doses of phenylephrine (0.5–2 µg) and nitroprusside (0.3–1.5 µg), intravenously, by bolus. The low and high doses of the drugs were adjusted to change arterial pressure by approximately 20 and 40 mm Hg, respectively. The volume of injection was 10–30 µL and the bolus injection was followed by a saline flush of 50 µL. With an increase or decrease of arterial pressure, hindlimb perfusion pressure decreased or increased by reflex, respectively (Fig. 1). The gain of arterial baroreflex control of hindlimb vascular resistance was assessed by obtaining the slope of the regression line relating the maximal changes in arterial pressure to the maximal reflex changes in hindlimb perfusion pressure. Injections of the drugs were separated by at least 5 minutes, by which time, arterial pressure, as well as hindlimb perfusion pressure, had returned to a baseline level.

**Responses to Lumbar Sympathetic Nerve Stimulation**

We should consider the difference in vascular responses to changes in sympathetic nervous activity between SHR and WKY when changes in hindlimb vascular resistance after vagotomy as well as the gains of arterial baroreflex control of vascular resistance are to be compared between SHR and WKY. To assess the difference in vascular responses to changes in sympathetic nervous activity between SHR and WKY, we examined hindlimb vascular responses to graded lumbar sympathetic nerve stimulation in four groups of rats. The lumbar sympathetic chain was cut and stimulated electrically (10 V, 3 msec duration) at a baseline frequency of 1.5 Hz to restore perfusion pressure to the level that was present before vagotomy. The frequency of stimulation was then increased or decreased transiently by 1, 2, 3, or 4 Hz for 20 seconds, and the corresponding changes in perfusion pressure were recorded. Then we increased baseline frequency of electrical stimulation to 4.0 Hz in order to increase baseline perfusion pressure to the level similar to that seen after vagotomy. The frequency of stimulation was again increased or decreased transiently by 1, 2, 3, or 4 Hz for 20 seconds from this new baseline level.

**Measurements of Central Venous Pressure (CVP) and Left Ventricular End-Diastolic Pressure (LVEDP)**

Atrial pressure is an important determinant of cardiac receptor activation in rats. Mean left atrial pressure approximates LVEDP. Thus, we measured CVP and LVEDP in four groups of rats. The venous catheter was positioned in the superior vena cava through a jugular vein. The arterial catheter was inserted into a carotid artery and was advanced into the left ventricle.

We also measured changes in CVP and LVEDP during intravenous injections of phenylephrine or nitroprusside. If CVP and LVEDP were changed by phenylephrine or nitroprusside, the assessment of arterial baroreflex control of hindlimb vascular resistance using these drugs might not be valid, since then the drugs stimulated not only the arterial baroreceptors but also the cardiopulmonary baroreceptors.

**Estimation of Left Atrial Distensibility**

A previous study indicated that the cardiopulmonary stretch receptors are localized mostly in the atrium, but not in the ventricle, in rats (Thoren et al., 1979a). Thus we determined left atrial distensibility in four groups of rats: young SHR (weight 257 ± 3 g), old SHR (384 ± 4 g), young WKY (278 ± 4 g), and old WKY (399 ± 4 g).

Under urethane anesthesia, a catheter was inserted into an external jugular vein, and heparin (500 U/kg) was given intravenously to prevent coagulation. After diastolic arrest of the heart by intravenous potassium (1 mEq/kg), the heart and lungs were excised and kept moist in the ice-chilled Krebs solution. A double lumen catheter (Argyle 18 G) was inserted into the left atrium by way of the
left pulmonary vein, while other pulmonary veins were ligated. A tight ligature was put around the atroventricular groove to obtain the isolated left atrium (Rickstein et al., 1980).

The isolated left atrium was rinsed to remove the blood. A great care was taken to exclude air bubbles. Left atrial pressure was recorded via the catheter with the recording transducer at the level of the left atrium. The left atrium was first allowed to empty passively against atmospheric pressure. No collapse of the atrium was seen during this procedure. From this basal level, the left atrial volume was increased by infusing saline into the left atrium at a rate of 100 µl/sec, and left atrial pressure was recorded continuously. The infusion was stopped when left atrial pressure reached 15 mm Hg. Thereafter, the left atrium was allowed to empty passively to reach the basal level again. The initial left atrial volume at the basal state was measured, using a microsyringe (Hamilton Co. Inc.), by removing the contained solution until the left atrial cavity completely collapsed. The procedures were repeated five times in each experiment, and the average value was used for later analysis. The left atrial pressure-volume curve was obtained by relating left atrial pressure and the infused volume relative to the initial volume (% of the initial volume). Left atrial pressure fluctuated greatly until it reached 2–2.5 mm Hg, but, thereafter, the rise of left atrial pressure was smooth as infusion was continued at a constant rate. Therefore, the curves were constructed from left atrial pressure of 2.5 to 12.5 mm Hg.

Data Analysis

To obtain the gain of arterial baroreflex control of hindlimb vascular resistance, we first calculated the ratio of the reflex change in hindlimb perfusion pressure to the change in arterial pressure with the low or high dose of the drug, separately. The average difference in the calculated ratios between the low and high doses of phenylephrine or nitroprusside was small (10 ± 1% for both phenylephrine and nitroprusside), which indicated that the regression lines with phenylephrine or nitroprusside were closely linear. Thus, we averaged the ratios of the reflex changes in hindlimb perfusion pressure to the changes in arterial pressure with the low and high dose of the drugs, and used the averaged value as the gain of arterial baroreflex for later analysis. Student's t-test was used for comparisons between SHR and WKY, and paired t-test for comparisons before and after interventions in the same group. P < 0.05 was considered as a statistically significant difference. All values are expressed in mean ± SE.

### Results

#### Arterial and Hindlimb Perfusion Pressure before and after Vagotomy

Mean arterial pressure before vagotomy was higher in SHR than in WKY in both young and old groups (P < 0.01 for both groups) (Table 1). Mean arterial pressure was not different between the two groups (young vs. old) of either SHR or WKY (Table 1). Hindlimb perfusion pressure before vagotomy was adjusted to the level of systemic arterial pressure so that hindlimb perfusion pressure before vagotomy was higher in SHR than in WKY, both in young and old groups (P < 0.01 for both groups) (Table 1).

Vagotomy increased systemic arterial pressure only transiently. After transient elevation, systemic arterial pressure declined gradually over a period of 10–15 minutes (Fig. 2). At 10 minutes after vagotomy when the measurements of changes in systemic as well as hindlimb perfusion pressure after vagotomy were done, systemic arterial pressure was not significantly different from that at control in any group of rats (Table 1). The magnitudes of transient increase and the rate of subsequent descent of arterial pressure were quite variable between rats, and were not different as a group between SHR and WKY in either the young or old group (data are not shown).

In contrast, the elevation of hindlimb perfusion pressure after vagotomy was sustained during an observation of 10–15 minutes (Fig. 2). At 10 minutes after vagotomy, hindlimb perfusion pressure was significantly elevated in all four groups of rats (P < 0.05 for old SHR, P < 0.01 for the other three groups) (Table 1). The elevation of hindlimb perfusion pressure after vagotomy (Δ perfusion pressure in Table 1) was greater (P < 0.05) in young SHR than in young WKY, but there was no difference between old SHR and old WKY.

The increase in hindlimb perfusion pressure after vagotomy must be influenced by the difference in vascular responses to changes in sympathetic activity. In all four groups, the increases in hindlimb perfusion pressure in response to graded sympathetic nerve stimulation were linear over the ranges

### Table 1

<table>
<thead>
<tr>
<th></th>
<th>Baseline MAP (mm Hg)</th>
<th>Baseline Hindlimb PP (mm Hg)</th>
<th>After vagotomy MAP (mm Hg)</th>
<th>After vagotomy Hindlimb PP (mm Hg)</th>
<th>% increase of hindlimb PP (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SHR-Y</td>
<td>156 ± 5*</td>
<td>148 ± 6*</td>
<td>6 ± 3</td>
<td>54 ± 5*</td>
<td>37 ± 4*</td>
</tr>
<tr>
<td>SHR-O</td>
<td>168 ± 4*</td>
<td>164 ± 8</td>
<td>-6 ± 5</td>
<td>20 ± 4†</td>
<td>8 ± 2*</td>
</tr>
<tr>
<td>WKY-Y</td>
<td>95 ± 2</td>
<td>95 ± 3</td>
<td>5 ± 2</td>
<td>21 ± 2†</td>
<td>23 ± 3</td>
</tr>
<tr>
<td>WKY-O</td>
<td>100 ± 3</td>
<td>103 ± 3</td>
<td>-1 ± 2</td>
<td>24 ± 4†</td>
<td>23 ± 4</td>
</tr>
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</table>

SHR-Y = young SHR, SHR-O = old SHR, WKY-Y = young WKY, WKY-O = old WKY in this and subsequent tables.

* P < 0.01, SHR vs. WKY (between age-matched groups).
† P < 0.05, SHR vs. WKY (between age-matched groups).
‡ P < 0.05, before vs. after vagotomy.
of hindlimb perfusion pressure which were observed before and after vagotomy (Fig. 3). The slope of this linear portion of the response curve was 1.4 times steeper in young SHR than in young WKY, and in old SHR compared with old WKY (Fig. 3).

In rats with sham vagotomy, systemic arterial pressure did not change, but hindlimb perfusion pressure increased by 6 ± 2 mm Hg (P < 0.05) during an observation of 15 minutes. In rats with lumbar sympathectomy, the increase in hindlimb perfusion pressure after vagotomy was less than 3 mm Hg (n = 3), which suggests that the increase in hindlimb perfusion pressure after vagotomy was mediated by the sympathetic nervous system.

**Arterial Baroreflex Control of Hindlimb Vascular Resistance before and after Vagotomy**

Before vagotomy, the gain of arterial baroreflex control of hindlimb vascular resistance in response to phenylephrine was not different between young SHR and young WKY, or between old SHR and old WKY (Table 2). The gain of arterial baroreflex in response to nitroprusside also was not different between young SHR and young WKY, but was greater in old SHR than in old WKY (P < 0.05) (Table 2).

Vagotomy increased the gain of phenylephrine-induced reflex vasodilation in young SHR (P < 0.01), young WKY (P < 0.01), and old WKY (P < 0.05), but not in old SHR (Table 2; Fig. 4). Similarly, the gain of nitroprusside-induced reflex vasoconstriction was increased by vagotomy in young SHR (P < 0.01), young WKY (P < 0.05), and old WKY (P < 0.05), but not in old SHR (Table 2; Fig. 4). The percent increase in the gain of phenylephrine-induced reflex vasodilation after vagotomy tended to
Figure 4. The arterial baroreflex slope in response to arterial hypertension and hypotension induced by intravenous phenylephrine and nitroprusside, respectively, before and after vagotomy in four groups of rats.

Figure 5. The arterial baroreflex slope in response to arterial hypertension and hypotension induced by intravenous phenylephrine and nitroprusside, respectively, before and after vagotomy in four groups of rats.

Figure 6. Changes in hindlimb perfusion pressure in response to graded lumbar sympathetic nerve stimulation in young SHR. The stimulation of sympathetic nerves was varied from a low baseline frequency at 1.5 Hz (open circles) and a high baseline frequency at 4 Hz (closed circles). The slopes of the changes in perfusion pressure were linear and not altered by the difference in a baseline frequency. Bars indicate SE.
TABLE 3

<table>
<thead>
<tr>
<th></th>
<th>CVP (mm Hg)</th>
<th>LVEDP (mm Hg)</th>
</tr>
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<tbody>
<tr>
<td>SHR-Y</td>
<td>2.6 ± 0.1*</td>
<td>7.1 ± 0.4*</td>
</tr>
<tr>
<td>SHR-O</td>
<td>2.0 ± 0.3*</td>
<td>5.6 ± 0.7*</td>
</tr>
<tr>
<td>WKY-Y</td>
<td>1.2 ± 0.2</td>
<td>3.1 ± 0.4</td>
</tr>
<tr>
<td>WKY-O</td>
<td>0.8 ± 0.3</td>
<td>2.8 ± 0.4</td>
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* P < 0.01, SHR vs. WKY (between age-matched groups).

Left Atrial Distensibility

The initial volume of the isolated left atrium at atmospheric pressure was not significantly different between young SHR (63 ± 3 μl) and young WKY (69 ± 3 μl) or between old SHR (80 ± 6 μl) and old WKY (87 ± 4 μl). The relationship between the relative volume change of the left atrium and left atrial pressure was comparable between young SHR and young WKY (Fig. 7A), but the curve in old SHR was shifted toward the pressure axis, compared with that in old WKY (Fig. 7B).

Discussion

The results of this study allow three conclusions. First, vagal afferents exert tonic inhibition on hindlimb vascular resistance as well as on arterial baroreflex control of hindlimb vascular resistance in WKY and SHR with the intact arterial baroreceptors. Second, tonic inhibitory influence of vagal afferents on control of hindlimb vascular resistance is altered in SHR, compared with that in age-matched WKY. Third, the alteration in inhibitory influence of vagal afferents in SHR is different in the stage of duration of hypertension. Inhibitory influence of vagal afferents is augmented in young SHR but attenuated in old SHR, as compared with the results in age-matched WKY.

Tonic Inhibitory Influence of Vagal Afferents on Control of Hindlimb Vascular Resistance

Bilateral vagotomy produced an immediate elevation of hindlimb perfusion pressure which was sustained during 10–15 minutes of observation in all four groups of rats (Table 1; Fig. 1). Hindlimb perfusion pressure increased gradually in rats with sham vagotomy, but the increase in hindlimb perfusion in time control rats was considerably smaller than that seen after vagotomy. The increase in hindlimb vascular resistance after vagotomy was neurally mediated, since it was not observed in rats with lumbar sympathectomy. Since systemic arterial pressure was not lowered during this period, the increase in hindlimb vascular resistance was not caused by reflex from the arterial baroreceptors. We considered the possibility that aortic depressor nerves were cut with vagi, and denervation of the aortic baroreceptors might have contributed to the increase in hindlimb vascular resistance after vagotomy. However, this possibility appears unlikely, since aortic depressor nerves infrequently run with vagi in rats (Krieger and Marseillan, 1963), but the elevation of hindlimb perfusion pressure after vagotomy was a persistent finding in these rats. We observed reflex hindlimb vasodilation in response to intravenous phenylephrine in several rats after bilateral carotid sinus denervation and vagotomy, which indicates that aortic depressor nerves were not cut with vagotomy. Thus, it appears that vagal afferents exerted tonic inhibition on hindlimb vascular resistance in these rats.

The results also suggest that vagal afferents tonically inhibited arterial baroreflex control of hindlimb vascular resistance in three groups of rats other than old SHR. Bilateral vagotomy increased the gains of arterial baroreflex control of hindlimb vascular resistance in response to arterial hypertension, as well as hypotension, in these rats (Table 2; Fig. 4).
We considered the possibility that the increases in the gains of arterial baroreflex after vagotomy in three groups of rats were caused by the mechanisms other than tonic inhibition of arterial baroreflex by vagal afferents. First, we considered the possibility that vascular responses to a given increase or decrease in sympathetic activity might be augmented when baseline sympathetic tone to the hindlimb was increased. However, this possibility is unlikely, since changes in sympathetic activity gave comparable and linear changes in hindlimb perfusion pressure when they were instituted at a higher or lower baseline level of sympathetic activity (Fig. 6). Second, it might be possible that the increase in the gain of arterial baroreflex was due to the difference in the set point of arterial pressure in the stimulus-response curve of the arterial baroreceptors. However, this possibility is also unlikely, since systemic arterial pressure changed little after vagotomy at the time of measurements.

These results in rats are different from the results in rabbits. Guo et al. (1982) recently reported that bilateral vagotomy in rabbits with the intact arterial baroreceptors did not alter hindlimb vascular resistance or the gain of arterial baroreflex control of hindlimb vascular resistance. The reason for the difference in the results between this study and that by Guo et al. is not clear, but it is possible that the role of vagal afferents in control of hindlimb vascular resistance is different in species.

Vagotomy produced a sustained elevation of hindlimb perfusion pressure, but systemic arterial pressure was only transiently elevated, and declined gradually over the following 10–15 minutes to a level similar to that before vagotomy (Table 1; Fig. 3). A gradual recovery of arterial pressure toward control after transient elevation following vagotomy has been described (Hosomi and Yokoyama, 1982). The mechanisms of the recovery of arterial pressure after vagotomy is unknown, but is not due to the reflex adjustment by the arterial baroreceptors, since it occurs in animals with the arterial baroreceptors denervated (Hosomi and Yokoyama, 1982).

**Altered Control of Hindlimb Vascular Resistance by Vagal Afferents in SHR**

The magnitude of elevation of hindlimb perfusion pressure after vagotomy was greater in young SHR than in young WKY, but was not different between old SHR and old WKY (Table 1). However, the difference in vascular responses to changes in sympathetic activity between SHR and WKY should be taken into consideration in comparing the elevation of hindlimb perfusion pressure after vagotomy between SHR and WKY. The slope of hindlimb vascular responses to graded sympathetic nerve stimulation was 1.4 times steeper in young and old SHR as compared with that in corresponding age group of WKY (Fig. 3). We attempted to normalize the difference in vascular responses to changes in sympathetic nervous activity by taking the ratio of the increase in hindlimb perfusion pressure after vagotomy to the slope of vascular response to graded sympathetic nerve stimulation. The ratio was greater in young SHR than in young WKY, but was less in old SHR than in WKY.

Thus, we consider that tonic inhibitory influence of vagal afferents on arterial baroreflex control of hindlimb vascular resistance was greater in young SHR than in young WKY, but was less in old SHR than in old WKY.

To assess the effect of vagotomy on the gain of arterial baroreflex control of hindlimb vascular resistance, we obtained the percent increase in the gain after vagotomy. The percent increase in the gain after vagotomy should be affected little by the difference in vascular responses to changes in sympathetic activity, since the influence of altered vascular responses must be similar before and after vagotomy. The percent increase in the gain of phentolamine-induced reflex vasodilation after vagotomy tended to be greater (0.5 < P < 0.1), and that of nitroprusside-induced reflex vasoconstriction was greater in young SHR than in young WKY. In contrast, the increase in the gain of phentolamine-induced reflex vasodilation, as well as nitroprusside-induced reflex vasoconstriction, was less in old SHR than in old WKY.

These effects of vagotomy on the gains of arterial baroreflex cannot be explained by the difference in the magnitudes of the increase in sympathetic activity produced by vagotomy between SHR and WKY, since hindlimb vascular responses to a given increase or decrease in sympathetic activity were not altered by the difference in baseline sympathetic activity (Fig. 6).

It is reported that intravenous phentolamine produced reflex vasodilation in the hindlimb in rabbits with sinoaortic denervation (Guo et al., 1982). Phenylephrine-induced reflex vasodilation in rabbits with sinoaortic denervation was abolished by bilateral vagotomy, which suggested that reflex vasodilation was mediated by vagal afferents. However, we did not observe reflex hindlimb vasodilation in response to phentolamine in rats with sinoaortic denervation with intact vagi (unpublished observation). Furthermore, the results by Guo et al. suggest that vagal afferents do not contribute to phentolamine-induced reflex vasodilation in animals with intact arterial baroreceptors (Guo et al., 1982). In this study, we examined whether CVP and LVEDP were altered by intravenous phentolamine or nitroprusside. If there were changes in CVP and LVEDP, they might have affected the gains of arterial baroreflex by modifying vagal afferents activity. However, CVP and LVEDP were not significantly altered by phentolamine or nitroprusside in either SHR or WKY at a time when reflex changes in hindlimb perfusion pressure occurred.

Thus, we consider that the tonic inhibitory influence of vagal afferents on arterial baroreflex control of hindlimb vascular resistance at least tended to be
greater in young SHR than in young WKY, but was less in old SHR than in old WKY.

Possible Mechanisms for Altered Control of Vascular Resistance by Vagal Afferents in SHR

The mechanisms for altered control of vascular resistance by vagal afferents in SHR might involve changes in the activity of vagal afferents from the cardiopulmonary receptors. To explore this possibility, we examined variables that are important to determine the activity of vagal afferents. We measured CVP and LVEDP and determined left atrial distensibility, since it is reported that the cardiac receptors localize mostly in the atrium in rats (Thoren et al., 1979a).

Left atrial distensibility was determined by obtaining the pressure-volume relationship during rapid infusion of saline into the left atrium at a rate of 100 μl/sec. A few points should be noted for proper interpretation of the results of this method. First, we determined only the dynamic distensibility of the left atrial wall, but not the static distensibility. However, in the in vivo situation, the cardiac receptors are activated during the atrial v-wave, which lasts for 80 to 100 msec in rats (Ricksten et al., 1980). Thus, the atrial wall is distended very rapidly in vivo. It was therefore of interest to study the dynamic but not the static distensibility of the left atrium. Second, the rate of saline infusion was fixed and was not adjusted to the initial volume of the left atrium. If the rate of infusion relative to the initial volume was different between groups, it is not valid to compare the curves, since the curve must be influenced by the rate of infusion. However, the initial volume of the left atrium was not different between young SHR and young WKY, or between old SHR and old WKY. Thus, the rate of infusion relative to the initial volume was different for groups between young SHR and young WKY or between old SHR and old WKY.

CVP and LVEDP were elevated in young SHR, compared with the values in young WKY. Left atrial distensibility was comparable between the two groups. If the distensibility measured in this method truly reflected the distensibility at the sites of main receptor location, we may reasonably assume that activation of the atrial receptors was greater and, thus, that vagal afferent activity was greater in young SHR than in young WKY.

On the other hand, left atrial distensibility was decreased in old SHR, compared with that in old WKY. These results are compatible with the results reported by Ricksten and co-workers in SHR that were older than our young SHR (Ricksten et al., 1980). Resetting of the cardiac receptors found in older SHR (Thoren et al., 1979b) was attributed to the decreased distensibility of the atrial wall, as well as to other factors, presumably changes in the cardiac receptors themselves (Ricksten et al., 1980). CVP and LVEDP were higher in old SHR than in old WKY. However, because of the decreased distensibility of the atrial wall and possible changes in the receptors, vagal afferent activity might have been less in old SHR than in old WKY, despite elevated atrial pressure.

It is reported that there is a central interaction between the input from the arterial baroreceptors and that from the cardiac receptors with vagal afferents (Koike et al., 1975; Mancia et al., 1976; Chen et al., 1978). This interaction is such that the inhibitory influence of the cardiac receptors on sympathetic outflow is increased when the inhibitory input from the arterial baroreceptors is reduced (Koike et al., 1975; Mancia et al., 1976). In other words, the excitatory response to interruption of inhibitory vagal afferents by vagotomy is greater when the inhibitory influence from the arterial baroreceptors is reduced. We considered the possibility that this mechanism might have contributed to the difference in inhibitory influence of vagal afferents between SHR and WKY. However, when the steepest portion of the stimulus-response relationship of arterial baroreflex before vagotomy was compared (Konner, 1971), there was no difference between SHR and WKY in either young or old rats. We assumed that the greater gain represented the steepest portion of the stimulus-response relationship of phenylephrine-induced reflex vasodilation or nitroprusside-induced reflex vasoconstriction (Mancia et al., 1979). The gain of arterial baroreflex was assessed by relating changes in arterial pressure to reflex changes in hindlimb perfusion pressure. When the gain of arterial baroreflex is calculated in this way, it is influenced by the difference in vascular reactivity or release of neurotransmitters from the nerve endings. Thus, it may not represent the true gain of arterial baroreflex in terms of reflex control of sympathetic outflow to the hindlimb. However, when we normalize reflex changes in hindlimb perfusion pressure by changes in hindlimb perfusion pressure in response to graded sympathetic stimulation, the steepest portion of the normalized gains was not different between SHR and WKY in either young or old rats (data are not shown). Thus, it appears unlikely that the central interaction between arterial and cardiac receptor reflex would account for the difference in inhibitory influence of vagal afferents on control of hindlimb vascular resistance between SHR and WKY.

We did not examine the possible contribution of central nervous system to the findings. It is conceivable that the modulation of the input from vagal afferents by the central nervous system at the brainstem might be altered in SHR.

In conclusion, the results of this study indicate that vagal afferents contribute importantly to control of hindlimb vascular resistance in SHR and WKY. However, control of hindlimb vascular resistance by vagal afferents is significantly altered in SHR, compared to that in WKY. The results suggest that the alteration in control of vascular resistance by vagal afferents in SHR is different in the stage or duration
of hypertension: augmented in the early but attenuated in the late stage of hypertension as compared with the results in age-matched WKY. The difference in the results between young and old SHR was not due simply to the age-related changes, since it was not found in WKY. Although we have no direct evidence, we favor the view that the alteration in control of vascular resistance by vagal afferents in SHR, either in the early or late stage of hypertension, may be due to the alteration in the activity of vagal afferents from the cardiac receptors. These results may suggest that the influence of vagal afferents should be taken into consideration in evaluating reflex or neural control of vascular resistance in SHR.

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Altered control of hindlimb vascular resistance by vagal afferents in spontaneously hypertensive rats. Difference in the early and late stage of hypertension.

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