In increased vascular reactivity to vasoconstrictor substances has been observed both in clinical hypertension and in animal models of hypertension. However, the mechanisms responsible for this increased reactivity are not completely understood. Folkow et al. have provided evidence suggesting that this hyperresponsiveness to hypertension is due to structural changes in the walls of the resistance vessels which result in an increased vessel reactivity.
wall-lumen ratio; thus, according to this theory, the hyperresponsiveness would be a result of the hypertension and not a cause of it. On the other hand, several findings would argue that the mechanism postulated by Folkow et al. is inadequate. It has been observed that in hypertensive animal models there is a lowered threshold for pressor substances, a finding which cannot be explained on the basis of structural modifications in the vessel walls. Also, increased responsiveness of in vitro aortic strips from hypertensive animals has been seen. Furthermore, rats in which the vascular bed of one leg was protected from the elevated arterial pressure during the development of hypertension still had abnormally high increases in resistance in response to pressor substances. These studies suggest that the exaggerated pressor responses of hypertensives to pressor stimuli may be due to some alteration in the responsiveness of the vascular smooth muscle cells; according to this theory, the hyperresponsiveness could be a cause of the hypertension. Because these two theories are not mutually exclusive, it is possible that both mechanisms could play a role in the vascular hyperresponsiveness in hypertension.

There is considerable evidence pointing to an interaction between the vasoconstrictor effects of angiotensin II and the sympathetic nervous system or norepinephrine. Infusions of angiotensin II have been shown to potentiate the vascular effects of sympathetic stimulation or norepinephrine infusion. Because disturbances of the renal circulation are known to promote renin release, it is possible that the renin-angiotensin system may be involved in the hyperresponsiveness to norepinephrine seen in renal hypertensive animal models.

The present study examined the pressor effects of norepinephrine in rabbits with hypertension due to renal artery stenosis of at least 30 days' duration, and in rabbits with renal artery stenosis of only 3 days' duration. It was reasoned that if vascular hyperresponsiveness is a cause of hypertension, then it should be manifest early in the disease, before the hypertension becomes fully developed. These studies also investigated a possible role for angiotensin II in the pressor responses to norepinephrine in these two animal models by the infusion of angiotensin II and by the use of the compound [1-sarcosine, 8-isoleucine]angiotensin II, a competitive antagonist of angiotensin II.

Methods

Male New Zealand White rabbits weighing 2.5-3.6 kg were used. All rabbits were fed a commercial diet (Purina rabbit chow) containing 0.167 mEq of Na and 0.467 mEq of K per gram. Water was available ad libitum. The rabbits were divided into three groups: 19 rabbits served as normal controls, 15 rabbits had renal artery stenosis of 3 days' duration (3-day clipped rabbits), and 20 rabbits had renal artery stenosis of over 30 days' duration (chronic renal hypertensive rabbits). Renal artery stenosis was produced by the method of Brooks and Muirhead. The rabbits for renal artery stenosis were anesthetized with halothane (5% with nitrous oxide and oxygen) and, with sterile surgical procedures, the abdomen was opened by a ventral midline incision; the left renal artery was constricted by a small silver clip with a gap size of 0.6 mm, and the right kidney was removed. The abdomen was closed and the rabbit was allowed to recover. For the 3-day clipped rabbits, the experiment was performed 3 days later whereas, in the chronic renal hypertensive rabbits, the experiment was performed at least 30 days after renal artery stenosis.

On the day prior to an experiment, each rabbit was anesthetized with halothane, and a polyvinyl catheter (Fr. 5 infant feeding tube) was placed in the lower aorta by way of the femoral artery. In the rabbits in which cardiac output would be measured, a similar Fr. 5 polyvinyl catheter also was placed in the femoral vein; another similar catheter was placed in the jugular vein so the tip of the catheter would lie near the right atrium. After insertion of the catheters, each rabbit was placed in a rectangular box to restrict its movements, and it remained in the box throughout the experiment. The following day, the experiments were performed on conscious rabbits. A few hours prior to an experiment, either one (in experiments 1 and 2) or two (in experiments 3 and 4) catheters of polyethylene tubing (PE 50) were placed in the marginal ear veins; one catheter was used for the infusion of norepinephrine and, in experiments 3 and 4, the other catheter was used for the infusion of angiotensin II or the angiotensin II antagonist, [1-sarcosine, 8-isoleucine]angiotensin II.

Mean arterial pressure was measured through the arterial catheter with a Statham transducer (P23Db). Heart rate was determined by recording pulsatile arterial pressure at a fast paper speed (5 mm/sec). The technique used in this laboratory for measuring cardiac output in rabbits by dye dilution has been described previously. After the rabbit had been heparinized, cardiac output was determined by injecting 0.2 ml of indocyanine green dye (Cardio-green, Hynson, Westcott, and Dunning, Inc.), 2.5 mg/ml, into the jugular vein while arterial blood was pumped through a densitometer cuvette and back into the animal through the venous catheter by a roller pump, at a rate of 10.0 ml/min. The arterial catheter was attached to the pressure transducer by a three-way stopcock so that either the cardiac output or the arterial pressure could be determined by turning the stopcock. The dye concentration in the blood and the arterial blood pressure were recorded on a Hewlett-Packard model 7754A recorder. Each cardiac output determination was made in triplicate, and the average of the three determinations was accepted as the measured...
value. Repeat cardiac output measurements varied by an average of less than 4%.

At the beginning of each experiment and before the rabbit was given heparin, a 2-ml sample of arterial blood was obtained. One milliliter was added to a tube containing ethylenediaminetetraacetate and was immediately placed in an ice bath; this sample was for the determination of plasma renin activity (PRA). The remainder of the sample was placed in a clot tube for serum urea nitrogen (SUN) determination. Blood samples for PRA were spun in a refrigerated centrifuge, and the plasma was stored frozen at −4°C until assayed. PRA was determined by radioimmunoassay of generated angiotensin I, as described by Cohen et al. SUN was determined by the procedures described by Fawcett and Scott.

**Experiment 1: Determination of Pressor Sensitivity to Norepinephrine**

Six normal rabbits, six 3-day clipped rabbits, and six rabbits with chronic renal hypertension received infusions of norepinephrine in doses of 25, 50, 100, and 200 ng/min per kg of body weight. Each dose of norepinephrine was infused for 5 minutes, and the mean arterial pressure was recorded continuously. The mean arterial pressure during the 1-minute period prior to norepinephrine infusion was taken as the control pressure, and the mean arterial pressure during the 5th minute of norepinephrine infusion was taken as the pressor response. An interval of at least 5 minutes was allowed between infusions, and the next dose was not infused until the mean arterial pressure had returned to the original control level. The norepinephrine (Levophed, Winthrop Laboratories) was diluted with 5% dextrose in water to prepare the solutions for infusion; a new ampul of norepinephrine was used for each experiment, and the solutions were prepared just prior to use. The norepinephrine solutions for 100 ng/min per kg were infused at 0.68 ml/min, and the other norepinephrine doses were infused at 0.34 ml/min.

**Experiment 2: Determination of Pressor Responsiveness to Norepinephrine**

Thirteen normal rabbits, 10 3-day clipped rabbits, and 12 rabbits with chronic renal hypertension received infusions of norepinephrine in doses of 400, 800, and 1200 ng/min per kg of body weight. The procedures for the infusions were the same as in experiment 1. The norepinephrine solutions for 400 and 1200 ng/min per kg were infused at 0.34 ml/min, and the 800 ng/min per kg solution was infused at 0.68 ml/min. Measurements of cardiac output and heart rate were made in six normal rabbits, five rabbits with renal artery stenosis of 3 days' duration, and seven rabbits with chronic renal hypertension before and during the 5th minute of norepinephrine infusion at 800 ng/min per kg.

**Experiment 3: Potentiation of Pressor Responsiveness to Norepinephrine by Angiotensin II**

The effect of angiotensin II in subpressor and pressor doses on the elevation in mean arterial pressure produced by a standard dose of norepinephrine was investigated in six normal rabbits, seven 3-day clipped rabbits, and five rabbits with chronic renal hypertension. Norepinephrine at a dose of 400 ng/min per kg was infused into the marginal ear vein for 5 minutes and the pressor response was recorded. During this period, isotonic saline was infused into the opposite marginal ear vein at 0.2 ml/min. After the end of the norepinephrine infusion, we waited for at least 5 minutes for the mean arterial pressure to return to preinfusion levels; the saline infusion then was discontinued and solution of synthetic angiotensin II [Asp7, Ile8]angiotensin II, Schwarz/Mann) in isotonic saline was infused into the marginal ear vein at a subpressor dose at 0.2 ml/min for 5 minutes. Norepinephrine at 400 ng/min per kg again was infused intravenously (iv) for 5 minutes while the angiotensin II infusion was continued. When all infusions were completed and the mean arterial pressure had again reached the preinfusion level, angiotensin II was infused iv in a pressor dose for 5 minutes; then norepinephrine at the same dose of 400 ng/min per kg again was infused concurrently with the angiotensin II infusion for 5 minutes. During these experiments the mean arterial pressure was recorded continuously.

To determine the subpressor dose of angiotensin II to be used in each experiment, an initial infusion of angiotensin II was made at doses of 5, 2.5, and 1.3 ng/min per kg. For the normal control rabbits, an angiotensin II infusion of 5 ng/min per kg was the subpressor dose in all experiments. In four of the 3-day clipped rabbits, angiotensin II infused at 5 ng/min per kg was the subpressor dose, but in one rabbit a lower angiotensin II dose of 2.5 ng/min per kg was required to be subpressor, and in one rabbit a dose of 1.3 ng/min per kg was needed. Rabbits with chronic renal hypertension all required an angiotensin II infusion of 2.5 ng/min per kg for a subpressor dose, except one, which required a dose of 1.3 ng/min per kg. The pressor dose of angiotensin II employed in each experiment was determined by preliminary infusions of angiotensin II at several dose levels to find a dose that would elevate the mean arterial pressure by less than 15 mm Hg. For the normal, control rabbits, the pressor dose of angiotensin II was 25 ng/min per kg; this increased mean arterial pressure by an average of 8 ± 1 (SEM) mm Hg. For the rabbits with renal artery stenosis of 3 days' duration, an angiotensin II dose of 25 ng/min per kg was used in three, whereas one rabbit required 5 ng/min per kg, and one required an angiotensin II dose of only 2.5 ng/min per kg. These
doses of angiotensin II in the 3-day clipped rabbits increased the mean arterial pressure by an average of 7 ± 1 mm Hg. All of the rabbits with chronic renal hypertension except one required 2.5 ng/min per kg as a pressor dose of angiotensin II; the exception required 1.3 ng/min per kg. The average increase in mean arterial pressure in chronic renal hypertensive rabbits with the pressor doses of angiotensin II was 9 ± 2 mm Hg.

**Experiment 4: Angiotensin II Antagonism and Pressor Responsiveness to Norepinephrine**

The effect of a competitive antagonist of angiotensin II, [1-sarcosine, 8-isoleucine]angiotensin II, on the pressor responsiveness to norepinephrine was studied in six normal rabbits, six 3-day clipped rabbits, and six rabbits with chronic renal hypertension. After 20 minutes of saline infusion into the marginal ear vein at 0.14 ml/min, the initial control mean arterial pressure was recorded. Norepinephrine was infused iv at 800 ng/min per kg for 5 minutes and the pressor response was determined. The norepinephrine infusion was stopped, and, when the mean arterial pressure had returned to the baseline level, the pressor response of a bolus of 300 ng of synthetic angiotensin II in isotonic saline, injected iv, was noted. After the pressor response had subsided, a saline solution of the angiotensin II antagonist was infused into the marginal ear vein at a rate of 300 ng/min per kg of body weight (flow rate of 0.14 ml/min). After 30 minutes of infusion of the angiotensin II analogue, a bolus of 300 ng of angiotensin II was injected again to test the completeness of the blockade. While the infusion of the angiotensin II antagonist was continued, the infusion of norepinephrine at 800 ng/min per kg was repeated for 5 minutes and the pressor response was noted again.

**Statistics**

For comparisons of values between groups of rabbits, the test statistic used was Student’s t-test for paired observations.22 For comparisons within a group of rabbits, Student’s t-test for group observations was used. Significance levels were selected as either $P < 0.05$ or $P < 0.01$. In experiment 1, the response threshold for each group of rabbits was calculated by the u-test to determine the smallest norepinephrine dose which would produce a pressor response that was significantly ($P < 0.05$) greater than zero.

**Results**

The initial values for mean arterial pressure, heart rate, PRA, and SUN for all three groups of rabbits in all experiments are summarized in Table 1. In the rabbits with renal artery stenosis of over 30 days’ duration (chronic renal hypertensive rabbits), the mean arterial pressure was significantly ($P < 0.01$) higher than that of the normal group of rabbits; however, the mean arterial pressure for all 3-day clipped rabbits was only slightly higher and not significantly different from that of the normal rabbits. Heart rate was significantly ($P < 0.01$) less in the 3-day clipped rabbits than in the normal rabbits, but in the rabbits with chronic renal hypertension the heart rate was unchanged from normal. In the 3-day clipped rabbits, the PRA was slightly lower but not significantly different from values for the normal rabbits, but the PRA in the rabbits with chronic renal hypertension was significantly ($P < 0.01$) decreased below that of the normal rabbits. The SUN was slightly but significantly ($P < 0.01$) elevated in both the 3-day clipped rabbits and in the rabbits with chronic renal hypertension.

**Experiment 1: Determination of Pressor Sensitivity to Norepinephrine**

Infusions of norepinephrine in doses of 25, 50, 100, and 200 ng/min per kg all produced greater increases in mean arterial pressure in the 3-day clipped rabbits and in the chronic renal hypertensive than in the normal control rabbits (see upper panel, Table 2). Also, the degree of elevation in arterial pressure with these doses of norepinephrine was almost the same in the 3-day and in the 30-day renal artery stenosis rabbits. The threshold dose of norepinephrine for the normal group of rabbits was 50 ng/min per kg, but both the 3-day clipped rabbits and the chronic renal hypertensive rabbits had a lower threshold of 25 ng/min per kg. Thus, both the

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Values for All Rabbits</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Normal rabbits (n = 19)</td>
</tr>
<tr>
<td>Mean arterial pressure (mm Hg)</td>
<td>88 ± 2</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>268 ± 12</td>
</tr>
<tr>
<td>Plasma renin activity (ng A I/ml hr)</td>
<td>3.5 ± 0.6</td>
</tr>
<tr>
<td>Serum urea nitrogen (mg/100 ml)</td>
<td>20.1 ± 1.0</td>
</tr>
</tbody>
</table>

*Values are means ± SEM. A I = angiotensin I.

† = $P < 0.01$ and ‡ = $P < 0.05$ when compared to the normal rabbit group by Student's t-test for group observations.
3-day clipped rabbits and the chronic renal hypertensive rabbits showed an increased sensitivity to norepinephrine.

**Experiment 2: Determination of Pressor Responsiveness to Norepinephrine**

The results of this experiment are summarized in the lower panel of Table 2. The pressor responses in the 3-day clipped rabbits and in the rabbits with chronic renal hypertension were significantly \( P < 0.01 \) greater than in the normal rabbits at all three of these higher dose levels of norepinephrine. The elevations in mean arterial pressure produced by these doses of norepinephrine were almost the same in the 3-day clipped rabbits and in the chronic renal hypertensive rabbits.

Combining the results of this experiment with the pressor responses to the lower doses of norepinephrine obtained in experiment 1 allowed the determination of complete dose-response curves for the three groups of rabbits. The dose-response curve for each group could be described adequately by the equation: \( y = a (\log x)^n \), where \( y \) was the change in mean arterial pressure, \( x \) was the norepinephrine dose, and \( a \) and \( n \) were constants. The values for these constants and the correlation coefficients for each of the three groups of rabbits are given in Table 3. These calculated dose-response curves are shown in Figure 1. The lower curve in this figure represents the dose-response characteristics of the normal group of rabbits. The dose-response curves for the 3-day clipped rabbits and the chronic renal hypertensive rabbits were very similar and showed considerable overlap. Because these curves were difficult to distinguish, the upper curve of this figure represents the approximate dose-response curve for both groups of rabbits.

Table 4 summarizes the data describing cardiac output, total peripheral resistance (TPR), heart rate, and stroke volume before and during the infusion of norepinephrine at 800 ng/min per kg for all members of the three groups of rabbits in which these determinations were performed. Cardiac output values are given in ml/minute per kg of body weight. Stroke volume, in ml/beat per kg, was calculated by dividing the cardiac output by the heart rate. The TPR values are expressed in the arbitrary units that result from dividing the arterial pressure by the cardiac output.

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**Table 2** Increases in Mean Arterial Pressure in Response to Several Dose Levels of Norepinephrine

<table>
<thead>
<tr>
<th>Norepinephrine infusion (ng/min per kg)</th>
<th>Normal rabbits ((n = 6))</th>
<th>3-day clipped rabbits ((n = 6))</th>
<th>Chronic renal hypertensive rabbits ((n = 6))</th>
</tr>
</thead>
<tbody>
<tr>
<td>25</td>
<td>1 ± 0.5</td>
<td>3 ± 0.3* (T)</td>
<td>3 ± 0.7† (T)</td>
</tr>
<tr>
<td>50</td>
<td>2 ± 0.6 (T)</td>
<td>6 ± 1.2†</td>
<td>5 ± 0.4*</td>
</tr>
<tr>
<td>100</td>
<td>3 ± 0.7†</td>
<td>8 ± 2.4*</td>
<td>9 ± 0.9*</td>
</tr>
<tr>
<td>200</td>
<td>5 ± 0.8</td>
<td>13 ± 2.4*</td>
<td>15 ± 2.5*</td>
</tr>
<tr>
<td>400</td>
<td>7 ± 0.7</td>
<td>19 ± 2.1*</td>
<td>18 ± 2.3†</td>
</tr>
<tr>
<td>800</td>
<td>13 ± 1.4</td>
<td>26 ± 2.9*</td>
<td>23 ± 2.0*</td>
</tr>
<tr>
<td>1200</td>
<td>19 ± 2.0</td>
<td>31 ± 6.4†</td>
<td>32 ± 2.1†</td>
</tr>
</tbody>
</table>

\( n = 13 \)  \( n = 10 \)  \( n = 12 \)

* = \( P < 0.01 \) and † = \( P < 0.05 \) when compared to the normal rabbit group by Student's \( t \)-test for group observations. T = threshold response \( (P < 0.05 \) that the response is greater than zero, by \( t \)-test).

**Table 3** Constants for the Equation \( y = a (\log x)^n \) Used to Describe the Norepinephrine Dose-Response Curves, and the Correlation Coefficients \( (r) \)

<table>
<thead>
<tr>
<th></th>
<th>Normal rabbits</th>
<th>3-day clipped rabbits</th>
<th>Chronic renal hypertensive rabbits</th>
</tr>
</thead>
<tbody>
<tr>
<td>a</td>
<td>0.282</td>
<td>1.160</td>
<td>1.121</td>
</tr>
<tr>
<td>n</td>
<td>3.562</td>
<td>2.913</td>
<td>2.944</td>
</tr>
<tr>
<td>r</td>
<td>0.983</td>
<td>0.996</td>
<td>0.990</td>
</tr>
</tbody>
</table>

---

**Figure 1** Changes in mean arterial pressure (mm Hg) in response to the infusions of various doses of norepinephrine in three groups of rabbits. 3-clip = 3-day clipped rabbits; CRH = chronic renal hypertensive rabbits. Variations are SEM. Lower curve represents normal rabbits. Upper curve represents 3-day clipped rabbits and chronic renal hypertensive rabbits; they were very similar. * = \( P < 0.05 \) and ** = \( P < 0.01 \) that the change in mean arterial pressure is greater than in normal rabbits.
in mm Hg by the cardiac output in ml/min per kg. As can be seen from Table 4, the control cardiac output values were not significantly different among the three groups of animals, although the mean for the normal group was slightly higher than for the other two groups. In each group the cardiac output failed to change during norepinephrine infusion. The TPR was somewhat higher in the 3-day clipped rabbits than in the normal control group, but this difference was not statistically significant; however, the rabbits with chronic renal hypertension did show a significant (P < 0.05) elevation in TPR compared to the normal rabbit group by Student’s t-test for paired observations.

Experiment 3: Potentiation of Pressor Responsiveness to Norepinephrine by Angiotensin II

The results of this experiment are summarized in Table 5. The infusion of angiotensin II in pressor doses resulted in enhanced pressor responses to norepinephrine in normal rabbits but did not alter the pressor responses to norepinephrine in the 3-day clipped rabbits or in the chronic renal hypertensive rabbits. Likewise, infusion of angiotensin II in pressor doses resulted in enhanced pressor responses to norepinephrine in normal rabbits but, again, failed to potentiate the effect of norepinephrine on arterial pressure in 3-day clipped rabbits or in rabbits with chronic renal hypertension.

Experiment 4: Angiotensin II Antagonism and Pressor Responsiveness to Norepinephrine

Infusion of norepinephrine at 800 ng/min per kg into normal rabbits resulted in an increase in mean arterial pressure from 87 ± 3 to 101 ± 1 mm Hg, for an average increase of 14 ± 3 mm Hg. Infusion of the angiotensin II antagonist for 30 minutes did not alter the mean arterial pressure, and the infusion of norepinephrine concurrently with the angiotensin II analogue produced a rise in mean arterial pressure from 85 ± 3 to 99 ± 2 mm Hg for an average increase of 14 ± 4 mm Hg. Thus, the angiotensin II antagonist did not alter the pressor responsiveness of normal rabbits to norepinephrine (see Fig. 2).

Infusion of norepinephrine at 800 ng/min per kg into 3-day clipped rabbits increased the mean arterial pressure from an average of 97 ± 4 to 123 ± 6 mm Hg, for an average increase of 26 ± 3 mm Hg. Again, a 30-minute infusion of the angiotensin II antagonist did not produce any changes in mean arterial pressure. However, when norepinephrine was infused during angiotensin II blockade, the

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**Table 4** Effect of Infusion of Norepinephrine, 800 ng/min per kg

<table>
<thead>
<tr>
<th>Normal rabbits (n = 6)</th>
<th>3-day clipped rabbits (n = 5)</th>
<th>Chronic renal hypertensive rabbits (n = 7)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cardiac output (ml/min per kg)</td>
<td>237 ± 36</td>
<td>230 ± 30</td>
</tr>
<tr>
<td>Total peripheral resistance</td>
<td>0.40 ± 0.06</td>
<td>0.45 ± 0.06</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>302 ± 15</td>
<td>230 ± 21</td>
</tr>
<tr>
<td>Stroke volume (ml/beat per kg)</td>
<td>0.80 ± 0.16</td>
<td>1.03 ± 0.24</td>
</tr>
</tbody>
</table>

Values are means ± SEM.

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**Table 5** Effect of Norepinephrine Infusion (400 ng/min per kg) on Mean Arterial Pressure, and the Influence of Concurrent Infusions of Subpressor and Pressor Doses of Angiotensin II

<table>
<thead>
<tr>
<th>Normal rabbits (n = 6)</th>
<th>3-day clipped rabbits (n = 5)</th>
<th>Chronic hypertensive rabbits (n = 5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>88 ± 2.3</td>
<td>90 ± 4.3</td>
</tr>
<tr>
<td>Norepinephrine infusion</td>
<td>95 ± 1.7</td>
<td>104 ± 5.4</td>
</tr>
<tr>
<td>Change</td>
<td>+7 ± 1.4</td>
<td>+15 ± 2.8</td>
</tr>
<tr>
<td>Control</td>
<td>87 ± 2.7</td>
<td>91 ± 4.4</td>
</tr>
<tr>
<td>A II subpressor dose</td>
<td>88 ± 2.6</td>
<td>92 ± 4.5</td>
</tr>
<tr>
<td>Norepinephrine infusion</td>
<td>100 ± 1.5</td>
<td>105 ± 6.2</td>
</tr>
<tr>
<td>Change</td>
<td>+12 ± 2.7</td>
<td>+13 ± 2.6</td>
</tr>
<tr>
<td>Control</td>
<td>85 ± 3.0</td>
<td>86 ± 4.4</td>
</tr>
<tr>
<td>A II pressor dose</td>
<td>92 ± 2.7</td>
<td>92 ± 3.9</td>
</tr>
<tr>
<td>Norepinephrine infusion</td>
<td>108 ± 3.3</td>
<td>103 ± 4.4</td>
</tr>
<tr>
<td>Change</td>
<td>+17 ± 3.7</td>
<td>+11 ± 2.3</td>
</tr>
</tbody>
</table>

Values are means ± SEM for the mean arterial pressure in mm Hg. A II = angiotensin II.

* = P < 0.05, compared to the change in mean arterial pressure in the same group of rabbits without infused angiotensin II; Student’s t-test for paired observations.
mean arterial pressure rose from 92 ± 3 to 109 ± 5 mm Hg for an average increase of only 17 ± 2 mm Hg. This increase in mean arterial pressure with norepinephrine was significantly (P < 0.01) less than that seen with the same dose of norepinephrine prior to infusion of the angiotensin II antagonist (see Fig. 3) and was almost the same as the response of normal rabbits to this dose of norepinephrine. Thus, the angiotensin II antagonist abolished the hyperresponsiveness of the 3-day clipped rabbits to this dose of norepinephrine.

In the rabbits with chronic renal hypertension, the infusion of norepinephrine at 800 ng/min per kg increased the mean arterial pressure by 31 ± 6 mm Hg (from 117 ± 2 to 147 ± 7) before the administration of the angiotensin II antagonist. Mean arterial pressure did not vary during the infusion of the angiotensin II antagonist, and after 30 minutes of angiotensin II analogue infusion, the average pressor response to the test dose of norepinephrine was 21 ± 4 mm Hg (from 114 ± 3 to 135 ± 6); this was significantly (P < 0.05) less than that observed before the angiotensin II analogue infusion (see Fig. 4). Thus, although the angiotensin II antagonist did not totally abolish the hyperresponsiveness of chronic renal hypertensive rabbits to norepinephrine, it did produce a substantial attenuation of the pressor response.

The intravenous injection of 300 ng of synthetic angiotensin II into normal rabbits, 3-day clipped rabbits, and chronic renal hypertensive rabbits resulted in mean arterial pressure elevations of 15 ± 3, 16 ± 2, and 26 ± 8 mm Hg, respectively. After administration of the angiotensin II analogue, the pressor response to this dose of angiotensin II was completely abolished.

**Discussion**

The values for cardiac output during the control period for 3-day clipped rabbits and chronic renal hypertensive rabbits were not different from those of the normal group of rabbits. Previous studies from this laboratory also have demonstrated that...
Responses to norepinephrine in rabbits with renal stenosis may be involved in the genesis of renal hypertension; Ogden et al. reported that rabbits with renal artery stenosis developed increased pressor responses to norepinephrine before they developed hypertension. In the present study, the infusion of norepinephrine at a dose of 800 ng/min per kg resulted in significant increases in TPR with no changes in cardiac output in all three groups of rabbits. Thus, the elevations in mean arterial pressure which occurred in response to this dose of norepinephrine were reflections of increases in circulatory resistances, presumably due to contractions of arteriolar smooth muscle cells. These experiments revealed that the infusion of norepinephrine in rabbits with renal artery stenosis of only 3 days' duration resulted in increases in arterial pressure to the same degree as rabbits with renal artery stenosis of over 30 days' duration. Because 3 days constitute insufficient time for structural changes to develop in the arterioles, and because the 3-day clipped rabbits were not hypertensive, the hypertensive responsiveness to norepinephrine which occurred in this group of rabbits cannot be attributed to morphological changes in the vessel walls. In 1940 Ogden et al. reported that rabbits with renal artery stenosis developed increased pressor responses to vasopressin before they developed hypertension. Also, McQueen observed an increased responsiveness to norepinephrine in the perfused hindquarters of rats with renal artery stenosis of 2 days' duration. The results of the present study confirm the findings that the pressor hyperresponsiveness to norepinephrine precedes the onset of hypertension; these findings are in accord with the hypothesis that increased vascular responses to pressor substances may be involved in the genesis of renal hypertension.

The contention that the increased pressor responses to norepinephrine in rabbits with renal artery stenosis may be due to factors other than structural changes in the vessel walls is supported further by the observation that the threshold dose of norepinephrine required for a pressor response was lower both in the 3-day clipped rabbits and in the rabbits with chronic renal hypertension than it was in the control group of rabbits. However, these experiments examined only the pressor responses to these lower doses of norepinephrine, and cardiac output was not determined. Because norepinephrine may produce increases in cardiac output in situations where the rise in arterial pressure does not reflexly inhibit the myocardial contractions, the increased sensitivity to norepinephrine seen in rabbits with renal artery stenosis could have been due to increased vascular or cardiac sensitivity to norepinephrine.

Although the exaggerated pressor responses to norepinephrine in the rabbits with renal artery stenosis probably were due to increased responsiveness of the vascular smooth muscle cells to norepinephrine, these studies were performed on rabbits with intact autonomic reflexes. Thus, these experiments do not exclude the possibility that altered autonomic control of the cardiovascular system could occur following renal artery stenosis, and that diminished sympathetic tone could have contributed to the heightened pressor responses to norepinephrine observed in the rabbits with renal artery stenosis.

Many studies have revealed that angiotensin II interacts with the sympathetic nervous system and with norepinephrine to potentiate their vasoconstrictor effects. Sakurai and Hashimoto reported that angiotensin II increased the vasoconstrictor effects of norepinephrine in isolated perfused rabbit ears. The response to norepinephrine of the perfused hindlimbs of rats was observed by Sato and Masuyama to be enhanced by angiotensin II. Paini and Bourdois reported that angiotensin II increased the vasoconstrictor responses to norepinephrine and sympathetic nerve stimulation in the mesenteric blood vessels of the cat. A similar potentiating effect of angiotensin II on the constrictor action of norepinephrine and sympathetic nerve stimulation in the perfused caudal artery of the rat was noted by Nicholas. The potentiating effect of angiotensin II on the vascular responses to norepinephrine or sympathetic nerve stimulation has been attributed to an increased amount of norepinephrine being released from terminal nerve endings or to an inhibitory effect of angiotensin II on the reuptake of norepinephrine. Also, Day and Moore provided evidence that angiotensin II may potentiate the constrictor effects of norepinephrine and other pressor agents by inhibiting the active extrusion of sodium from the vascular smooth muscle cells, thereby causing a nonspecific sensitization. Yet another possibility is that angiotensin II may be acting...
on the central nervous system to increase the sympathetic tone. In the present study, the infusion of angiotensin II in subpressor or pressor doses in normal rabbits potentiated the pressor response to norepinephrine at 400 ng/min per kg; this finding is in agreement with the results obtained by others, as cited above. It should be noted that in the present study the potentiated pressor response to norepinephrine in normal rabbits was almost the same as the pressor response to this same dose of norepinephrine in 3-day clipped rabbits and was only slightly less than the norepinephrine pressor response in the rabbits with chronic renal hypertension. On the other hand, in rabbits with renal artery stenosis of 3 days' duration and in rabbits with renal artery stenosis of over 30 days' duration and hypertension, the infusion of angiotensin II in subpressor or pressor doses failed to alter the pressor responses to norepinephrine. These findings suggest that angiotensin II may be involved in the hyperresponsiveness to norepinephrine of hypertensive and prehypertensive animals with renal artery stenosis.

The experiments in which endogenously produced angiotensin II was antagonized by the use of a competitive inhibitor of angiotensin II provided further support for this hypothesis. Zimmerman found that the angiotensin II antagonist, [1-sarcosine, 8-alanine]angiotensin II, blocked the potentiating effect of angiotensin II on the vasoconstrictor response to norepinephrine or sympathetic nerve stimulation in the perfused hindpaw of the dog. In the present study, the infusion of an angiotensin II antagonist had no effect on the pressor responses to norepinephrine in normal rabbits. However, the infusion of this compound into 3-day clipped rabbits totally abolished the hyperresponsiveness to infused noradrenaline and, in rabbits with chronic renal hypertension, this angiotensin II analogue attenuated the increased pressor responsiveness to norepinephrine. These findings provide strong evidence that angiotensin II is involved in the vascular hyperresponsiveness to norepinephrine seen in animals with renal artery stenosis.

Failure of the angiotensin II antagonist to abolish the hyperresponsiveness to norepinephrine in chronic renal hypertensive rabbits suggests that some additional mechanism also may play a role in rabbits with renal artery stenosis of long duration; this additional mechanism may be structural changes in the arterioles.

The angiotensin II antagonist failed to lower the mean arterial pressure in the chronic renal hypertensive rabbits. Similar results have been reported previously from this laboratory. Because the angiotensin II antagonist diminished the pressor hyperresponsiveness of chronic renal hypertensive rabbits to norepinephrine but failed to diminish or alleviate the hypertension, these results suggest that the hyperresponsiveness to norepinephrine which is angiotensin mediated may not be involved in maintaining the elevated arterial pressure in rabbits with chronic renal artery stenosis and hypertension.

From the present data we can only speculate as to the mechanisms whereby angiotensin II increases the pressor responses to norepinephrine in rabbits with renal artery stenosis. Although Romero et al. observed increases in plasma renin activity 3 days after renal artery stenosis and contralateral nephrectomy in rabbits, in the present study, the plasma renin activity was not elevated in rabbits with renal artery stenosis of 3 days' duration and, in fact, was greatly reduced in the rabbits with chronic renal hypertension. Therefore, the potentiating effect of angiotensin II in these rabbits probably was not due to elevated plasma levels of angiotensin II. The most likely explanation is that, in the rabbits with renal artery stenosis, there may be an increased number of angiotensin II receptors, or there may be an increased affinity of angiotensin II for these receptors.

Acknowledgments

We are very grateful for the [1-sarcosine, 8-isoleucine]angiotensin II which was provided in part by Daichi Seiyaku Company, Ltd., Tokyo, Japan. The indocyanine green dye (Cardio-Green) was generously provided by Dr. T.R. Carski, of Hynson, Westcott, and Dunning, Inc. The anti-angiotensin I antiserum used in the radioimmunoassay for renin was provided by Dr. E.L. Cohen, of the University of Michigan School of Medicine.

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The role of angiotensin II.

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doi: 10.1161/01.RES.43.3.437

_Circulation Research_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
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Print ISSN: 0009-7330. Online ISSN: 1524-4571

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