Characterization of Intrarenal Arterial Adrenergic Receptors in Renovascular Hypertension

Neil D. McElroy and Ben G. Zimmerman

α-Adrenergic receptor subtypes were investigated using [3H]prazosin, an α1-selective antagonist, and the α2-selective antagonist [3H]rauwolscine in a smooth muscle plasma membrane enriched microsomal fraction prepared from rabbit intrarenal arterial vasculature. Both radioligands displayed single components on Scatchard analysis. The specific binding of [3H]prazosin was of high affinity (0.54±0.04 nM) with a maximum binding capacity (B max) of 212±15 fmol/mg protein. The maximum number of [3H]rauwolscine binding sites was 64±4 fmol/mg of protein with a dissociation constant (K d) of 5.60±0.27 nM. Binding of both radioligands was rapid, saturable, and specific. α1- and α2-adrenergic receptors in the intrarenal arterial membrane preparation were also characterized at 2-, 4-, 6-, and 10–12-week intervals during the course of development and maintenance of chronic two-kidney, one clip (2K1C) Goldblatt hypertension and in age-matched sham-operated normotensive control rabbits. The α1-adrenergic receptor affinity for [3H]prazosin binding in hypertensive rabbits was significantly increased in the stenotic, but not contralateral, kidney at 2 weeks; however, at 6 weeks the receptor affinity of both kidneys was significantly increased compared with those of the normotensive control group. No difference in α1-adrenergic receptor affinity was seen at 12 weeks, and there were no changes in B max at any of the weekly intervals. Neither the K d, nor B max, for [3H]rauwolscine in either kidney showed a significant difference between hypertensive rabbits and normotensive control rabbits. These studies demonstrate the existence in the rabbit intrarenal arterial vasculature of binding sites with α1- and α2-adrenergic receptor specificity. Further, intrarenal arterial α1-adrenergic receptor affinity is significantly increased early (to at least 6 weeks) but not later (at most 12 weeks) during 2K1C Goldblatt hypertension. (Hypertension 1989;13:851–858)

Enhanced reactivity of regional vascular beds to various vasoconstrictor stimuli is a common feature in several models of experimental and human hypertension.1,2 Such increased vascular reactivity may involve, in part, adrenergic receptor changes,3,4 which have been shown to precede the elevation of blood pressure.5 Adrenergic receptors located on the postjunctional membrane mediate catecholamine-induced vasoconstriction and therefore directly influence vascular resistance. Increased vascular reactivity may result from differences in a number of factors, including cell membrane properties, level of membrane potential, and postreceptor phenomena. The sensitivity of blood vessels to catecholamines has also been shown to be influenced by α-receptor number and affinity6 and may be modulated by a variety of physiological and pathological conditions.7,8 Several investigations have concerned changes in vascular reactivity of the kidney in experimental hypertension. Because of the pivotal role of the kidney in maintaining elevated arterial pressure, alterations in renal vascular reactivity could lead to changes in renal hemodynamics that importantly influence the development and maintenance of hypertension. Enhanced renal vascular tone may be due to vasoconstrictor hyperresponsiveness9 or to increased activity of the renal sympathetic innervation.10 However, observations of increased sympathetic nerve activity in experimental hypertension differ,10,11 and to our knowledge there is no firm evidence of increased renal nerve activity in renovascular hypertension. Information on adrenergic functional alterations in renovascular hypertension have come largely from in vivo experiments, studies on isolated blood vessels and perfused kidney, or from whole kidney
homogenates. Thus, previous investigations have not provided a means of determining specifically alterations in renal vascular adrenergic receptors in hypertension, nor have the subtypes of these receptors in renal vessels been defined. The present investigation was conducted on a novel intrarenal arterial vascular preparation to characterize these receptors during various stages in the development and maintenance of the hypertensive process.

Materials and Methods

Animal Protocol

Experiments were initiated with male New Zealand White rabbits having body weights of less than 1 kg. Rabbits were anesthetized with 10–15 mg/kg pentobarbital via the marginal ear vein. Hypertension was induced by the Goldblatt method, with placement of a silver clip (0.22-mm slit) on the left renal artery in close proximity to the aorta. Young rabbits were used to facilitate hypertension development by allowing the renal artery to grow into the clip. Rabbits in the normotensive group underwent a sham operation with an identical silver clip placed near the renal artery. All rabbits were maintained on standard laboratory chow and tap water ad libitum.

Arterial pressures were determined in the conscious state by means of a permanent catheter placed in the carotid, auricular, or lower abdominal aorta (via the femoral artery). The arterial line was also used to obtain blood samples for determination of biweekly plasma renin activity (PRA). Animals were studied after 2, 6, and 12 weeks and 2, 4, and 10 weeks of sustained hypertension for \( \alpha_r \) and \( \alpha_2 \)-adrenergic receptors analysis, respectively.

Membrane Preparation

Rabbits were killed by rapid pentobarbital overdose, and the kidneys, abdominal aorta, and thoracic aorta were immediately excised. Kidneys were bisected and immersed in ice-cold modified Tris-HCl buffer (50 mM Tris-HCl, 10 mM MgCl₂, pH 7.4). Plasma membrane vesicles were prepared by modifications of methods from Kwan et al. 

Binding Assays

\([\text{H}]\text{Prazosin and [H]}\text{rauwolscine were used to assess } \alpha_r \text{- and } \alpha_2 \text{-adrenergic receptors in the plasma membrane fraction prepared from intrarenal arterial and aortic smooth muscle over the course of the induction and maintenance of two-kidney, one clip (2K1C) Goldblatt hypertension. Experiments were performed on fresh or frozen (in modified Tris-HCl buffer, pH 7.4, at -60° C) plasma membrane-enriched F₂ fractions. Previous experiments of radioligand-receptor binding have shown that membranes stored frozen up to 1 month give results comparable with those from freshly prepared membranes.}

In all radioligand binding assays, a modified Tris buffer (50 mM Tris-HCl and 10 mM MgCl₂, pH 7.2) was used in the incubation medium. Subdued filament room lighting was used to prevent photolysis of \([\text{H}]\text{rauwolscine radioligand.}

No degradation of either radioligand was observed in the presence of membrane during incubation under these conditions. The reactions were terminated by adding 3.0 ml ice-cold buffer (4°C) to the incubation mixtures. Membranes were collected on 0.1% polyethylene-amine pretreated Whatman GF/C microfibre glass filters by rapid vacuum filtration followed by three washes with 5.0 ml ice-cold buffer. The filters were air dried and placed in 5.0 ml scintillation solution containing pseudocumene-1,2,4 trimethylbenzene, Biosolve-emulsifier, PPO primary fluor, and bis-o-methyl styrylbenzene secondary fluor. Filters were left to equilibrate in scintillation cocktail at room temperature for 12 hours and counted on a Beckman Model LS5801 liquid scintillation counter. Quench correction in the counting of tritium was made using the external standard ratio method. Counting efficiency was 43%. Blanks were run for all the concentrations of radioligands as for binding with membranes except that in blank tubes there was no membrane, but they contained the same volume of modified Tris buffer.
Specific binding for \[^3H\]prazosin and \[^3H\]rauwolscine was defined as the radioactivity displaceable by 10 \(\mu\)M phentolamine and comprised 80–95% of total bound counts at a concentration of these radioligands near their dissociation constant \((K_d)\) values. Justification for the use of 10 \(\mu\)M phentolamine to define specific binding was based on displacement curves between various concentrations of phentolamine and the radioligands in which maximum displacement was reached at a concentration not greater than 5×10^{-6} M phentolamine.

Protein content of the membrane was determined by the method of Bradford, using crystalline serum bovine albumin as the standard. Data in all figures, text, and tables refer to specific binding and have been normalized in terms of the protein concentration.

\[^3H\]Prazosin (0.02–5.0 nM) and \[^3H\]rauwolscine (0.1–40.0 nM) were used for construction of Scatchard plots in duplicate for each assay. The incubation media contained 250 \(\mu\)l buffer with or without unlabeled drug and 50 \(\mu\)l diluted radioligand. The binding interactions were started by adding 100 \(\mu\)l membrane suspension (1 mg/ml) to make a final volume of 400 \(\mu\)l. Incubations were carried out in a covered rotary shaking water bath at 25\(^\circ\)C for 45 minutes, at which time equilibrium had been reached for all concentrations of each radioligand used in the study. Inhibition experiments were similar to saturation studies except that in these assays aliquots of the tissue homogenates were incubated with \[^3H\]prazosin or \[^3H\]rauwolscine for 45 minutes at 25\(^\circ\)C in the presence of at least 12 increasing concentrations of the various antagonists and \((-)\)epinephrine.

The reaction kinetics of the radioligands \[^3H\]Prazosin and \[^3H\]rauwolscine were studied using one concentration of the radioligand under standard assay conditions but at different incubation times. For dissociation studies, binding was allowed to proceed for 45 minutes before 100 \(\mu\)M phentolamine was added at specified intervals to the assay mixture. Kinetic rate constants were determined as detailed in Figure 3 and by the method described by Williams and Lefkowitz.

Data Analysis

Saturation radioligand binding curves were analyzed by the method of Scatchard\(\text{ }\) using the computerized weighted least-squares curve-fitting algorithm for determination of \(K_d\) and maximum binding capacity \((B_{\text{max}})\) radioligand binding parameters according to Munson and Rodbard. The apparent dissociation constant \((K_d)\) values for the competitive unlabeled antagonist were calculated by the method of Cheng and Prusoff. The \(K_d\) for each agonist and antagonist was calculated from the equation: \(K_d = IC_{50}/(1 + \frac{L}{K_d})\), where \(IC_{50}\) is the concentration of an unlabeled antagonist giving 50% inhibition of radioligand binding, \(L\) and \(K_d\) are the concentration of free tritiated ligand in the assay system and the equilibrium dissociation constant of the radioligand as determined from saturation experiments, respectively. To check whether ligand-receptor interaction studies have followed the law of mass action in saturation studies or displacement by the unlabeled compounds, the data have been transformed into the Hill equation: \(\log B/(B_{\text{max}} - B) = \log [L] - \log K_d\). Log B/(B_{\text{max}} - B) was plotted against log [L] to obtain the value of the Hill slope \((n_h)\) and the Hill binding dissociation constant \((K_d)\). Differences between mean values of \(K_d\) and \(B_{\text{max}}\) were tested for significance using a grouped t test with a Bonferroni transformation. The difference was taken to be significant when \(p<0.05\). Slopes of linear regression lines were fitted by the least-squares method.

Drugs

Radioligands \[^3H\]prazosin (specific activity, 26.0 Ci/mmol) and \[^3H\]rauwolscine (specific activity, 73.7 Ci/mmol) were purchased from New England Nuclear, Boston, Massachusetts. Other drugs were obtained as follows: rauwolscine (Regitine) and phentolamine (CIBA-GEIGY Corp., Summit, New Jersey), prazosin HCI and \((-)\)epinephrine (Sigma Chemical Co., St. Louis, Missouri), idazoxan (gift from Reckitt & Colman, Kingston-Upon-Hull, UK).

Results

[^3H]Prazosin Binding

Specific saturation binding data disclosed a single class of sites in the intrarenal arterial preparation over the concentration range of \[^3H\]prazosin studied (Figure 1A). \(K_d\) and \(B_{\text{max}}\) were 0.54±0.04 nM and \(212\pm15\) fmol/mg protein, respectively.

Specific binding of \[^3H\]prazosin to this fraction was rapid, with a half-time of approximately 3 minutes. In the presence of 100 \(\mu\)M phentolamine, \[^3H\]prazosin dissociated from the membranes with a half-time of approximately 2 minutes. According to the pseudo first-order rate equations, the apparent rate constant for the pseudo first-order reaction of association \((K_{\text{app}})\) of \[^3H\]prazosin binding at 0.5 nM concentration was 0.253/min (Figure 2A, insert), and the second-order rate constant \((K_i)\) was \(2.04\times10^8\) M⁻¹/min. The second-order dissociation rate constant \((K_{-1})\) was \(0.151/min\) (Figure 2B). The ratio of the \(K_{\text{app}}/K_i\) yielded a value of 0.74 nM, which compares well with the \(K_d\) values obtained by the equilibrium binding experiments (Figure 1A).

[^3H]Rauwolscine Binding

The specific binding of increasing concentrations of \[^3H\]rauwolscine in the F₂ fraction of the intrarenal arterial preparation was saturable, and the linearity of the Scatchard analysis indicated a single class of binding sites over the concentration range studied (Figure 1B). The \(K_d\) and \(B_{\text{max}}\) were 5.6±0.27 nM and \(64\pm4\) fmol/mg protein, respectively. Hill plots of the data were linear with an average Hill coefficient of 0.99±0.02.
The specific binding of $[^{3}H]$rauwolscine (7.2 nM) to the membranes of the intrarenal arteries was also time dependent with a half-time of about 3 minutes (Figure 2C). Assuming pseudo first-order kinetics, the association reaction reached apparent equilibrium with the $K_{eq}$ value of 0.209/min. In the dissociation experiments, $K_{d}$ was 0.091/min (Figure 2D). From the $K_{d}$ and the $K_{eq}$, a $K_{ip}$ value of $1.53 \times 10^{7}$ M/min was calculated. The dissociation constant calculated from the ratio of $K_{d}/K_{ip}$ was 5.9 nM, which is in close agreement with the $K_{d}$ calculated from the Scatchard analysis.

**Specificity of $[^{3}H]$Prazosin and $[^{3}H]$Rauwolscine Binding**

Data for these competition studies are shown in Table 1. Investigation of competition between unlabeled antagonists versus $[^{3}H]$prazosin and $[^{3}H]$rauwolscine disclosed that $[^{3}H]$prazosin and $[^{3}H]$rauwolscine were labeling distinct sites. The unlabeled prazosin was about 7,300-fold more potent in competing at the $[^{3}H]$prazosin recognition sites than at the $[^{3}H]$rauwolscine sites. In contrast, unlabeled rauwolscine displayed the reverse relation; rauwolscine was about 200-fold more potent at the $[^{3}H]$rauwolscine sites than at the $[^{3}H]$prazosin sites. Unlabeled $\alpha$-adrenergic antagonists competed for $[^{3}H]$prazosin binding sites with an order of potency of: prazosin > phentolamine > rauwolscine indicative of an $\alpha_{1}$-adrenergic receptor subtype. Competition curves with unlabeled antagonists against $[^{3}H]$rauwolscine revealed the reverse order of potency expected of an $\alpha_{2}$-adrenergic receptor subtype. The $\alpha$ agonist (-)-epinephrine competed equally for the $[^{3}H]$prazosin or $[^{3}H]$rauwolscine binding sites.

**Effects of Two-Kidney, One Clip Goldblatt Hypertension**

The time course of changes of mean arterial blood pressure and PRA in sham-operated and hypertensive rabbits is summarized in Table 2. 2K1C Goldblatt hypertensive rabbits in both the $\alpha_{1}$ and $\alpha_{2}$-adrenergic receptor studies maintained an increase in mean arterial blood pressure of at least 20 mm Hg over the sham-operated controls. Plasma renin activity in 2K1C hypertensive rabbits was increased approximately fourfold or greater to at least 4-6 weeks in both groups.

The binding affinity of $[^{3}H]$prazosin to $\alpha_{1}$-adrenergic receptors was significantly higher in the intrarenal arterial preparation derived from the stenotic, but not the contralateral, kidney of hypertensive rabbits at 2 weeks; at 6 weeks the receptor affinity of both kidneys was significantly increased compared with that in sham-operated normotensive control rabbits (Figure 3). However, at 12 weeks no difference was observed in the affinity of $\alpha_{1}$-adrenergic receptor binding. $B_{max}$ did not differ between sham-operated and hypertensive rabbits over the intervals studied. Both the $K_{d}$ and $B_{max}$ in microsomes derived from aortic smooth muscle of these rabbits did not differ over these time intervals (data not shown).

The results of $[^{3}H]$rauwolscine binding in the intrarenal arterial preparation are shown in Figure 4 at 2-, 4-, and 10-week intervals. Neither the $\alpha_{2}$-adrenergic receptor affinity nor the total number of binding sites in either kidney showed a significant difference between the hypertensive rabbits and their normotensive sham-operated controls.

**Discussion**

This study identified and characterized pharmacologically for the first time $\alpha_{1}$- and $\alpha_{2}$-adrenergic receptors in the intrarenal arterial vasculature. The
Figure 2. Kinetic analysis of [3H]prazosin and [3H]rauwolscine binding to the F2 fraction of the rabbit intrarenal arterial preparation. Results shown are means of two determinations performed in duplicate. Panel A, time course of association for specific [3H]prazosin binding at the radioligand concentration of 0.5 nM. Inset, determination of the second-order rate constant for [3H]prazosin binding. $B_\infty$ is the amount of [3H]prazosin bound at equilibrium, and $B_t$ is the amount bound at each time, $t$. The slope of the line relating $\ln(B_\infty - B_t)$ and time, determined by linear regression analysis is equal to the apparent rate constant for the pseudo first-order reaction of association ($K_{ap}$). Second-order rate constant ($K_t$) is calculated according to the equation $K_t = K_{ap} - K_d/[^*L]$, where $K_d$ is the dissociation rate constant and $[^*L]$ is the concentration of [3H]prazosin used in the assay. These results indicated $K_{ap} = 0.253/min$ and $K_t = 2.04 \times 10^7 M^{-1} min$ with a linear regression correlation coefficient ($r$) = 0.98. Panel B, time course for dissociation of [3H]prazosin determined by the addition of phentolamine to a final concentration of 10 μM. Inset, first-order rate plot of [3H]prazosin dissociation. Dissociation rate constant ($K_{d1}$) is equal to the slope of the line relating $\ln(B_t/B_0)$ and time, where $B_t$ refers to the specific binding at each time, $t$, and $B_0$ represents the binding at equilibrium before addition of phentolamine. $K_{d1} = 0.151/min$ with $r = 0.96$. Panel C, time course of association for specific [3H]rauwolscine binding at the radioligand concentration of 7.2 nM. Inset, determination of the second-order rate constant for [3H]rauwolscine binding. These results indicated $K_{ap} = 0.201/min$ and $K_t = 1.58 \times 10^7 M^{-1} min$ with $r = 0.99$. Panel D, time course of dissociation of [3H]rauwolscine on addition of phentolamine to a final concentration of 10 μM. Inset, first-order rate plot of [3H]rauwolscine dissociation. $K_{d1} = 0.091/min$ with $r = 0.99$.

$K_d$ and $B_{\text{max}}$ for the $\alpha_1$-adrenergic receptors in the intrarenal arterial vessels and aorta obtained in this study are within the range of previously reported values for vascular smooth muscle. $\alpha_1$-Adrenergic receptor number in the intrarenal vessels was approximately fourfold greater than that of the $\alpha_2$. In 2K1C Goldblatt hypertension, the $\alpha_1$-adrenergic receptors exhibited a significant affinity increase early in the course of development of the hypertension. This increase was, however, not sustained during the
TABLE 1. Inhibition Constants and Hill Coefficients for Competition of \([\text{H}]\text{Prazosin}\) and \([\text{H}]\text{Rauwolscine}\) Binding Sites in Rabbit Intrarenal Arterial Smooth Muscle Microsomes

<table>
<thead>
<tr>
<th>Drug Competitor</th>
<th>([\text{H}]\text{Rauwolscine})</th>
<th>([\text{H}]\text{Prazosin})</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(K_i(nM))</td>
<td>(n_H)</td>
</tr>
<tr>
<td>Rauwolscine</td>
<td>1.45±0.21</td>
<td>1.06±0.05</td>
</tr>
<tr>
<td>Idazoxan</td>
<td>4.02±0.38</td>
<td>0.93±0.03</td>
</tr>
<tr>
<td>Phentolamine</td>
<td>28.6±4.1</td>
<td>1.05±0.04</td>
</tr>
<tr>
<td>Prazosin</td>
<td>528±68</td>
<td>0.94±0.03</td>
</tr>
<tr>
<td>(-)Epinephrine</td>
<td>1,480±120</td>
<td>0.99±0.06</td>
</tr>
</tbody>
</table>

Inhibition constant (\(K_i\)) values calculated according to Cheng and Prusoff\(^{20}\); values are mean±SEM of duplicate determinations from two separate experiments in membrane preparations of six rabbits per group. Concentrations of \([\text{H}]\text{prazosin}\) and \([\text{H}]\text{rauwolscine}\) used in these experiments were 0.2–0.6 nM and 4.0–6.0 nM, respectively. \(n_H\), Hill coefficient; N.D., not determined.

12-week interval when PRA was normal. An increase in \(\alpha_1\)-adrenergic affinity may relate to altered vascular reactivity in the hypertensive rabbit. Because no changes were seen in the vascular \(\alpha_2\)-adrenergic receptors, they do not appear to be involved in the hypertension.

A relation between affinity of the \(\alpha_1\)-adrenergic receptor in vascular smooth muscle and the sensitivity of the arterial contractile response has been demonstrated in a series of 12 rabbit arteries.\(^{23}\) Accentuated adrenergic receptor–mediated responses have also been demonstrated in renovascular hypertension both in vitro and in vivo. This was manifested by increased \(\alpha_1\)-adrenergic receptor affinity in isolated aortic smooth muscle from Grollman hypertensive rats\(^{24}\) and by a decreased \(E_D_{50}\) of norepinephrine and phenylephrine for renal vasoconstrictor responses in the 2K1C Goldblatt hypertensive dog.\(^{23}\) Vascular wall hypertrophy did not appear responsible for the altered reactivity in the latter study since there was a temporal difference in the reactivity change between norepinephrine and a thromboxane analogue. Studies in the artificially perfused renal vascular preparation\(^{26}\) or in isolated arterial strips\(^{27}\) have shown that alterations in reactivity persist in preparations devoid of humoral and neurogenic control. Therefore, it may be concluded that an intrinsic change occurs in the vascular smooth muscle resulting in enhanced sensitivity to external stimuli.

Regional differences in affinity of the \(\alpha_1\)-adrenergic receptors of arterial vascular smooth muscle cells have been demonstrated.\(^{23}\) This may be an important determinant of differential reactivity of rabbit vascular smooth muscle. In the present study, an increase in affinity was seen in one region, the intrarenal vasculature, but not in the aorta of 2K1C Goldblatt hypertensive rabbits. This regional difference would seem to support the potential for regional variation in receptor plasticity. The regional difference, and the temporal variation in affinity, may explain, in part, failure of previous investigations of \(\alpha\)-adrenergic receptors in hypertension to demonstrate an affinity alteration. The present study is consistent with the previous findings of Niels and Bevan\(^{28}\) where arteries from spontaneously hypertensive rats were shown to be more sensitive to norepinephrine than arteries from Wistar-Kyoto rats. This increased sensitivity was associated with an increased norepinephrine \(K_i\) for the \(\alpha\)-adrenergic receptor but not with differences in receptor reserve or relative receptor occupation.

It is possible that the increased intrarenal arterial \(\alpha_1\)-adrenergic receptor affinity seen in the present study was related to the increase in plasma angiotensin II due to the early rise in PRA. At the 2-week

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**Table 2.** Mean Arterial Blood Pressure and Plasma Renin Activity in Goldblatt Hypertensive and Sham-Operated Normotensive Control Rabbits

<table>
<thead>
<tr>
<th>Weeks after procedure</th>
<th>MAP (mm Hg)</th>
<th>PRA (ng/ml/hr)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sham</td>
<td>Hypertensive</td>
</tr>
<tr>
<td><strong>Group 1</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>79±4</td>
<td>108±7*</td>
</tr>
<tr>
<td>6</td>
<td>93±6</td>
<td>114±4*</td>
</tr>
<tr>
<td>12</td>
<td>91±6</td>
<td>109±6*</td>
</tr>
<tr>
<td><strong>Group 2</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>65±3</td>
<td>98±4*</td>
</tr>
<tr>
<td>4</td>
<td>76±3</td>
<td>114±6*</td>
</tr>
<tr>
<td>10</td>
<td>85±2</td>
<td>107±6*</td>
</tr>
</tbody>
</table>

Group 1, \(n=5\); Group 2, \(n=6\) for hypertensive and sham-operated rabbits in each time period. Group 1 and Group 2 correspond to \(\alpha_1\)- and \(\alpha_2\)-adrenergic receptor experiments, respectively. MAP, mean arterial pressure; PRA, plasma renin activity. \(*p<0.05\) by grouped \(t\) test when compared with sham-operated rabbits.
postclipping interval, PRA had maximally increased at which time α₁-adrenergic receptor affinity of the stenotic kidney was significantly enhanced. PRA remained elevated at the 6-week interval when α₁ affinity of both the stenotic and contralateral kidney was significantly increased. At 12 weeks both PRA and affinity had returned to the control level. During this stage in the hypertension when receptor affinity normalized, other factors (e.g., structural) may play a more important role in increased vascular reactivity.

The present study revealed an early increase in affinity of the intrarenal arterial but not in aortic α₁-adrenergic receptors in 2K1C Goldblatt hypertension. This receptor affinity change is unlikely to represent the sole basis of the hypertension, but it may be important in regard to several factors (e.g., structural remodeling, altered sodium balance, or increased sympathetic tone) that eventually sustain the elevated blood pressure.

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**References**

7. Colucci WS, Gimbrone MA, Alexander WR: Regulation of myocardial and vascular α-adrenergic receptor affinity:
Effects of guanine nucleotides, cations, estrogens, and catecholamine depletion. **Circ Res** 1984;55:78-88
20. Cheng YC, Prusoff WH: Relationship between the inhibition constant (K_i) and the concentration of inhibitor which causes 50 percent inhibition (I_50) of an enzyme reaction. *Biochem Pharmacol* 1973;22:3099-3108

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