Onset of glomerular hypertension with aging precedes injury in the spontaneously hypertensive rat

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Tolbert, Evelyn M., Joseph Weisstuch, Helen D. Feiner, and Lance D. Dworkin. Onset of glomerular hypertension with aging precedes injury in the spontaneously hypertensive rat. Am J Physiol Renal Physiol 278: F839–F846, 2000.—The changes in renal hemodynamics that develop with aging in spontaneously hypertensive rats (SHR) were examined. Micropuncture studies revealed that glomerular capillary pressure was elevated in SHR at 9 mo of age compared with 3-mo-old SHR and 9-mo-old normotensive Wistar-Kyoto rats. Glomerular hypertension developed because of a small increase in systemic blood pressure and a decline in preglomerular vascular resistance, allowing transmission of elevated systemic pressure to the glomerular capillaries. The hemodynamic alterations were not a compensatory response to injury, inasmuch as vascular and glomerular morphology were normal in 9-mo-old SHR. To determine the mechanism of these changes, the activity of several vasoactive systems was examined. Similar changes in renal hemodynamics were observed in young and old SHR after blockade of nitric oxide production and after intravenous administration of endothelin. However, ANG II produced a proportionally greater reduction in glomerular filtration rate than renal blood flow in older SHR. These data suggest that reduced endogenous activity of the renin-angiotensin system leads to glomerular hypertension in aging SHR. Late development of glomerular hypertension may contribute to the subsequent appearance of glomerular sclerosis and progressive renal failure in these rats.

glomerular hemodynamics; angiotensin II; endothelin; nitric oxide

SYSTEMIC HYPERTENSION is the second most common cause of end-stage renal disease in the United States and accounts for approximately one-fourth of all patients receiving dialysis. A number of hypotheses have been proposed to explain the association between elevation in systemic blood pressure and progressive renal disease, including atherosclerotic vascular disease leading to ischemic nephropathy (28, 35) as well as progressive glomerular sclerosis (21). In many experimental models of chronic renal disease, glomerular injury has been associated with glomerular hypertension (29); however, elevation in glomerular pressure is not characteristic of all models of hypertension. For example, although spontaneously hypertensive rats (SHR) develop progressive glomerular sclerosis with aging, previous reports indicate that, before the onset of injury, glomerular capillary pressure (Pgc) is normal in SHR (4, 5, 17). Interestingly, proteinuria and morphological evidence of glomerular injury develop slowly in SHR and are not apparent in rats until ~1 yr of age (20). In contrast, most studies of glomerular hemodynamics in SHR have been performed in much younger animals, up to ~4 mo of age. Therefore, the purpose of this study was to determine whether glomerular pressure increased with aging in SHR and, therefore, might account for the eventual appearance of glomerular sclerosis and renal failure in older rats. Finding that glomerular hypertension developed in aging SHR, we also sought to determine whether it was an adaptive response to nephron loss or was functional in nature and humorally mediated.

METHODS

Summary of Groups

The institution’s Animal Use Committee approved all studies. Male SHR or their normotensive controls, Wistar-Kyoto (WKY) rats, were studied at 3 or 9 mo of age. Table 1 provides a summary of the number of animals of each strain at each time point that were examined in the specific protocols. For all studies, rats were housed in the standard fashion in an approved facility and had free access to food and water.

Micropuncture Studies

Micropuncture studies of glomerular hemodynamics were performed in 3- and 9-mo-old SHR and 9-mo-old WKY rats. One group of 9-mo-old SHR were given enalapril (13.3 mg·kg⁻¹·day⁻¹) in the drinking water for 8 wk. Micropuncture was not performed in young WKY rats, inasmuch as these studies have been previously performed by us (17) and others (4, 5) and are reported elsewhere.

Rats were anesthetized with thiobutabartal (Inactin, Andrew Lockwood, Sturtevant, WI; 100 mg/kg ip) and prepared in the standard fashion for micropuncture (9). A polyethylene (PE-50) catheter was inserted into the femoral artery, and mean arterial pressure (MAP) was measured by a Statham P23B pressure transducer connected to a recorder (model 7A, Grass Instruments, Quincy, MA). A tracheostomy was performed, and polyethylene catheters were inserted into the left and right jugular veins and the left ureter for infusion of test substances and collection of urine. The left kidney was exposed via a subcostal incision, placed on a Lucite holder, and illuminated with a fiber-optic light. All rats received an initial infusion of rat plasma equal to 10 ml/kg body wt...
Table 1. Description of groups

<table>
<thead>
<tr>
<th>Strain of Rat</th>
<th>Age, mo</th>
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<th>Type of Study</th>
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<tr>
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Study I: Glomerular hemodynamics

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<td>Clearance</td>
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<td>Clearance</td>
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Study II: L-NAME infusion

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<td>7</td>
<td>Clearance + flow probe</td>
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Study III: Endothelin-1 infusion

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<tr>
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<tr>
<td>SHR</td>
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<td>6</td>
<td>Clearance + flow probe</td>
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Study IV: ANG II infusion

followed by a sustained infusion of plasma at ~0.5 ml/h, adjusted to maintain a stable hematocrit. Rats also received a 0.5-ml iv bolus of [3H]methoxyinulin (Dupont) followed by a sustained infusion at 0.5 ml/h. After a 45-min equilibration period, timed collections of proximal tubule fluid were obtained by micropuncture for determination of inulin concentration by liquid scintillation counting. Urine and plasma content of inulin was measured by the same method, and single-nephron glomerular filtration rate (SNGFR) and glomerular filtration rate (GFR) were calculated by the standard formulas (8). Mean Pgc was estimated using the stop-flow pressure technique, as described by Allison et al. (1). The determinants of ultrafiltration and segmental vascular resistance were calculated using the standard formulas as set forth by Baylis et al. (8).

Whole Kidney Studies

Studies involving NO. SHR and WKY rats aged 3 and 9 mo were prepared for clearance studies in the manner described above for the micropuncture experiments, except the left kidney was not manipulated and immobilized. The rats received a 0.5-ml bolus of a solution of inulin (10%) and p-aminoinhippurate (PAH, 0.4–0.5%) in normal saline followed by a sustained infusion of the inulin-PAH mixture at a rate of 0.5 ml/h. After a 45-min equilibration period, two timed urine collections of 15 min with midpoint blood collections were taken as described above. After the two baseline periods, the NO synthase inhibitor N-nitro-l-arginine methyl ester (L-NAME) was infused intravenously in three escalating doses: 0.12 µg·kg⁻¹·min⁻¹ (low), 0.4 µg·kg⁻¹·min⁻¹ (intermediate), and 1.2 µg·kg⁻¹·min⁻¹ (high). Two urine and blood collections were made at each dose for determination of GFR and renal plasma flow (RPF). The concentrations of L-NAME were selected on the basis of preliminary studies in which the low dose was determined to represent the maximal amount of the agent not associated with an increase in systemic blood pressure.

The anthrone method (17) was used to determine inulin concentration in urine and plasma samples. PAH concentration in urine and plasma samples was determined using established methods (40). GFR and RPF were calculated as the clearances of inulin and PAH, respectively, according to the standard formulas.

Studies involving ANG II. Studies were performed on groups of 3- and 9-mo-old SHR. Rats were prepared for determination of inulin clearance as described above with the following modifications. For convenience, rather than the clearance of PAH, an ultrasonic flow probe was used to measure RPF. For this technique, a 6-mm section of the left renal artery was dissected free from the renal vein, with care taken to preserve the renal nerves. A 1-mm flow probe, connected to an ultrasonic blood flowmeter (model T106XM, Transonic Systems, Ithaca, NY), was placed adjacent to the renal artery for renal blood flow (RBF) determination. MAP was measured via a femoral artery catheter connected to an Abbott Transpac pressure transducer connected to a second channel of the ultrasonic flowmeter. The flowmeter was connected to a Compac computer running WINDAS software. All rats received infusions of isoncotic rat plasma and labeled methoxyinulin in normal saline, as described above. After two timed urine collections under basal conditions, rats received an infusion of ANG II (0.2 µg·kg⁻¹·min⁻¹). GFR and RPF measurements were repeated 30–45 min after initiation of the ANG II infusion, as previously described (37). GFR (inulin clearance), filtration fraction (FF), RPF, and renal vascular resistance (RVR) were calculated using standard formulas.

Studies of the role of endothelin. Male 3- and 9-mo-old SHR were anesthetized and prepared for clearance study exactly as described for the ANG II study. After baseline collections were made, an intravenous infusion of endothelin (ET)-1 (50 ng·kg⁻¹·min⁻¹) was started. After a 30- to 45-min stabilization period, timed urine and blood collections and MAP and RBF measurements were recorded as described above. The ET-1 dose and protocol were adapted from the study of Tank et al. (37). GFR, FF, and RVR were calculated.

Morphological Studies

Morphological studies were performed on the kidneys of 9-mo-old SHR and WKY rats that were fixed by perfusion with Formalin at the measured MAP for 5 min after micropuncture study. Two coronal slices of each kidney were embedded in Parafilm and processed for light microscopy. Three-micrometer-thick slices were stained with hematoxylin-eosin and periodic acid-Schiff. The extent of glomerular, vascular, and tubulointerstitial abnormalities was assessed in a blinded fashion by one observer. All glomerular and vascular profiles appearing in a single slice were examined. The total number of glomeruli showing evidence of focal or global sclerosis was counted, and the percentage of abnormal glomeruli was calculated for each animal. Tubulointerstitial disease was assessed in an semiquantitative fashion and scored on a scale of 0 to 3 for each animal.

Statistics

Statistical analysis was performed on a personal computer with use of SigmaStat software (Jandel Scientific Software, San Rafael, CA). Micropuncture data were compared by one-way ANOVA followed by multiple pairwise comparisons with Tukey's test. For studies involving two groups, the t-test was used. Statistical significance was defined as P < 0.05. Values are means ± SE.

RESULTS

Micropuncture and Morphological Studies

Results from the micropuncture studies of systemic and renal hemodynamics and 3 (young)- and 9 (old)-
mo-old SHR and 9-mo-old WKY rats are shown in Table 2. Body weight increased significantly in SHR between 3 and 9 mo of age; however, 9-mo-old WKY rats were significantly larger than similarly aged SHR. As has been repeatedly observed, MAP was already significantly elevated in SHR at 3 mo of age but increased further with aging on average by 14 mmHg by the time rats were 9 mo old. These values in SHR were markedly elevated compared with values obtained in WKY rats that were normotensive at 9 mo of age. The mean value for \( P_{gc} \) was 54 mmHg in SHR at 3 mo of age. This value was not significantly greater than that observed in normotensive WKY rats, in which the average value for \( P_{gc} \) was 53 mmHg. These values for \( P_{gc} \) in young SHR and old WKY rats are similar to those previously reported for SHR and WKY rats at 3 mo of age (17). Therefore, the data are consistent with previous reports indicating that glomerular pressure is not elevated in young, adult SHR (4, 5, 17). Again, consistent with previous data (17), the mean value for afferent resistance (\( R_a \)) in young SHR was significantly greater than that observed in WKY rats, indicating that \( P_{gc} \) is normal in young SHR, because increased preglomerular vascular resistance prevents transmission of elevated systemic blood pressure to the glomerular capillaries. Our data demonstrate that \( P_{gc} \) also does not increase with aging in WKY rats up to 9 mo of age.

In contrast, the mean value for \( P_{gc} \) was increased in older SHR to 61 mmHg, a value significantly greater than that observed in young SHR and old WKY rats. This value for \( P_{gc} \) is similar to that observed in other models of hypertension and renal disease in which elevations in glomerular pressure have been associated with glomerular injury (2, 18). This increase in \( P_{gc} \) resulted from the small rise in renal perfusion pressure combined with a 35% decline in preglomerular vascular resistance.

Because a decline in preglomerular resistance was responsible in part for glomerular hypertension in older rats, we next sought to determine the reason for this change. A decrease in \( R_a \) could result from vasodilation developing as a compensatory response to an ischemic loss of nephrons secondary to hypertension-induced atherosclerosis. However, evidence suggests that this was not the case in SHR at 9 mo of age. First, the GFR averaged 0.9 ± 0.3, 1.0 ± 0.4, and 1.3 ± 0.9 ml/min in young and old SHR and WKY rats, respectively (Table 2), and these values were not significantly different from each other. Therefore, there was no evidence for a loss in kidney function leading to renal vasodilation. More importantly, morphological analysis of kidney tissue harvested from SHR at 9 mo of age revealed no evidence of renal vascular or glomerular injury in these rats. These data suggest that the changes in renal hemodynamics in older SHR are functional in nature and not the result of compensatory adaptation to a partial loss of kidney tissue.

Logically, if not in response to renal injury, then a decline in \( R_a \) might result from increased production of an endogenous vasodilator or decreased production of a vasoconstrictor. To examine this hypothesis, we performed a series of studies to compare the activity of several vasoactive systems in young and old SHR.

Role of NO in Age-Associated Glomerular Hypertension

To examine the role of NO, RBF and GFR were measured during increasing blockade of NO synthase with the competitive inhibitor L-NAME. We reasoned that if increased endogenous production of NO accounted for a decrease in preglomerular resistance in older rats, then these rats should display an exaggerated vasoconstrictor response when NO production was blocked. The results of these studies are shown in Fig. 1. Young and old rats experienced parallel declines in RPF and GFR in response to increasing doses of L-NAME. That renal NO production was not enhanced in older SHR is further illustrated by Fig. 2, which shows the average percent reduction in GFR, RPF, and FF in young and old rats at baseline and at the highest dose of L-NAME. Again, rats displayed proportionally similar vasoconstrictor responses to blockade of NO production. These data provide no evidence for a significant increase in NO production contributing to renal vasodilation in older SHR.

We also compared responses to L-NAME in WKY rats and SHR. The percent changes in GFR, RPF, and FF in these rats at baseline and at the highest dose of L-NAME are shown in Fig. 3. Because similar responses were observed in young and old SHR (Fig. 2) and WKY rats (data not shown), data from 3- and 9-mo-old animals are combined in Fig. 3. Administration of L-NAME produced a significantly greater decline in GFR in SHR than in WKY rats, and this was associated with a trend toward lower RPF as well. These data suggest that RPF and GFR are more dependent on endogenous production of NO in SHR than in WKY rats in young and old animals.

### Table 2. Micropuncture data

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>Body Wt, g</th>
<th>Hct, %</th>
<th>MAP, mmHg</th>
<th>( P_{gc} ), mmHg</th>
<th>SNGFR, ml/min</th>
<th>( Q_A ), ml/min</th>
<th>( K_t ) nl·min⁻¹·mmHg⁻¹</th>
<th>( R_a ), dyn·s·cm⁻²</th>
<th>( R_{ef} ), dyn·s·cm⁻²</th>
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<tbody>
<tr>
<td>Young SHR</td>
<td>12</td>
<td>307 ± 8</td>
<td>50 ± 1</td>
<td>170 ± 4</td>
<td>55 ± 2</td>
<td>62 ± 8</td>
<td>184 ± 29</td>
<td>0.061 ± 0.011</td>
<td>3.36 ± 0.59</td>
<td>1.40 ± 0.22</td>
</tr>
<tr>
<td>Old WKY</td>
<td>6</td>
<td>550 ± 19*</td>
<td>44 ± 1</td>
<td>110 ± 4*</td>
<td>53 ± 2</td>
<td>71 ± 8</td>
<td>242 ± 45</td>
<td>0.098 ± 0.014</td>
<td>0.90 ± 0.09*</td>
<td>0.67 ± 0.08</td>
</tr>
<tr>
<td>Old SHR</td>
<td>12</td>
<td>375 ± 5†</td>
<td>45 ± 1</td>
<td>184 ± 5†</td>
<td>61 ± 1†</td>
<td>72 ± 3</td>
<td>227 ± 26</td>
<td>0.111 ± 0.052</td>
<td>2.18 ± 0.26</td>
<td>0.98 ± 0.09</td>
</tr>
<tr>
<td>Old SHR + enalapril</td>
<td>5</td>
<td>379 ± 10</td>
<td>48 ± 1</td>
<td>137 ± 14</td>
<td>58 ± 1</td>
<td>55 ± 1</td>
<td>222 ± 43</td>
<td>0.049 ± 0.021</td>
<td>1.56 ± 0.31</td>
<td>1.01 ± 0.18</td>
</tr>
</tbody>
</table>

Values are means ± SE; n, number of animals; Hct, hematocrit; MAP, mean arterial pressure; \( P_{gc} \), glomerular capillary pressure; SNGFR, single-nephron glomerular filtration rate; \( Q_A \), afferent blood flow; \( K_t \), ultrafiltration coefficient; \( R_a \) and \( R_{ef} \), afferent and efferent resistance; SHR, spontaneously hypertensive rats; WKY, Wistar-Kyoto rats. *P < 0.05 vs. young SHR; †P < 0.05 vs. old WKY; ‡P < 0.05 vs. old SHR.
Alternatively, a reduction in preglomerular resistance might result from a decline in the production or action of an endogenous vasoconstrictor substance. To assess the contribution of ET to the control of RVR in older rats, we examined the renal vascular response to intravenous administration of a moderately pressor dose of exogenous ET-1 in young and old SHR. As in the previous experiment, an exaggerated vasoconstrictor response to exogenous ET would suggest that the endogenous ET effect was decreased under basal conditions. The results of this experiment are shown in Fig. 4, which compares the percent change from baseline in RPF, GFR, and FF in young and old rats given ET-1. Administration of ET-1 produced quantitatively similar changes in renal hemodynamics in young and old rats. Therefore, the study suggests that the effects of endogenous ET on renal hemodynamics are similar in 3- and 9-mo-old SHR.

Role of ANG II

Somewhat different results were obtained when the effects of ANG II on preglomerular vascular resistance in young and old SHR were examined. The results of these studies are summarized in Fig. 5. Although there were statistically similar percent declines in RPF and GFR in young and old rats infused with ANG II, there was a proportionally greater reduction in RPF than GFR in young than in old rats. Thus FF increased by ~40% in young rats compared with a decline of 11% in older animals. This pattern of response to exogenous ANG II (see below) might occur if the endogenous vasoconstrictor action of ANG II on the afferent arteriole declined with aging in SHR.

To examine the role of endogenous ANG II in regulating renal hemodynamics in older SHR in another way, micropuncture studies were performed in a group of 9-mo-old SHR after administration of the angiotensin-converting enzyme (ACE) inhibitor enalapril. The results of this experiment are shown in Table 3. Administration of enalapril was associated with a significant decline in MAP of ~50 mmHg in SHR. Despite this marked fall in perfusion pressure, P gc was not significantly reduced by enalapril. An analysis of the segmental vascular resistances revealed only a modest decline in R A with no change in efferent resistance (R E) in enalapril-treated rats. This pattern of change, a selective decrease in R A associated with constancy of P gc, is...
the expected autoregulatory response. Autoregulatory changes in vascular resistance of this type are observed in isolated renal arterioles and probably result from intrinsic myogenic responses within these vessels. Therefore, these data are consistent with our hypothesis that preglomerular vascular tone in older SHR is not dependent on endogenous ANG II production under basal conditions.

DISCUSSION

Systemic hypertension is identified as causal in ~30% of patients developing end-stage renal disease in the United States (38). Evidence suggests that a significant subset of these patients have hypertensive nephrosclerosis, in which elevation in systemic blood pressure leads directly or indirectly to progressive glomerular sclerosis. For example, in the pilot study for the African-American Study of Kidney Disease, the majority of black hypertensive patients with mild-to-moderate renal insufficiency who underwent renal biopsy had lesions consistent with hypertensive nephrosclerosis (21). Notable in this study was the marked prevalence of global glomerular sclerosis in these individuals, which affected ~50% of all the glomeruli examined and correlated significantly with the degree of elevation in systolic blood pressure.

The genesis of glomerular sclerosis in patients with essential hypertension is still uncertain. A number of factors have been implicated, including increased frequency of genes that convey susceptibility to end-organ damage (10), congenital reduction in nephron number with associated nephron hypertrophy and hyperfunction (11, 21), and glomerular capillary hypertension. Regarding hemodynamic factors, elevation in glomerular pressure has been closely associated with the development of glomerular sclerosis in several experimental models of hypertension (42) but is not observed in all settings (4, 5). Whether glomerular hypertension precedes glomerular injury in hypertensive humans is unknown.

On the basis of studies of renal hemodynamics in SHR, Bauer et al. (6) proposed a sequence of changes in renal structure and function that might lead to glomerular hypertension and progressive glomerular injury in patients with essential hypertension. In young SHR, increased preglomerular vascular resistance prevents increased systemic pressure from reaching the glomerular capillaries (4, 5). Consistently, clearance studies early in the course of human essential hypertension reveal a hemodynamic pattern similar to that observed in young SHR, including an increase in RVR, a decrease in RBF, and preservation of GFR (31, 32). Bauer et al. hypothesized that whereas vasoconstriction might protect the glomerular capillaries in young hypertensive animals, eventually, atherosclerotic vascular disease developed, leading to glomerular ischemia and a partial loss of renal mass. Once a critical fraction of renal mass is lost, compensatory reductions in RA occur, allowing transmission of elevated pressure to the glomerular capillaries, thereby promoting further nephron destruction.

Late onset of glomerular hypertension has also been proposed to explain glomerular sclerosis and decline in renal function with normal aging; however, the existing data are inconsistent on this point. Maneuvers such as protein restriction and ACE inhibition that reduce glomerular pressure lessen age-associated glomerular sclerosis in rats (3); however, conflicting results have been obtained when Pgc has been directly measured in normotensive geriatric rats. Anderson et al. (3) reported that Pgc was increased in normotensive Munich-Wistar rats at 2 yr of age, primarily as a result of a decline in RA. Similarly, Zhang and co-workers (43) and Fujihara et al. (22) observed an increase in Pgc of 6–7 mmHg in geriatric compared with adolescent normotensive Sprague-Dawley rats. In both of these studies, morphological examination revealed significant glomerular injury in the kidneys of older rats at the time of micropuncture study. On the other hand, Reckelhoff et al. (34) reported that Pgc was not increased in 20- to 22-mo-old Sprague-Dawley rats, despite the presence of glomerular sclerosis. These authors concluded that glomerular hypertension did not account for age-associated glomerular injury. Komatsu et al. (27) assessed glomerular hemodynamics in 73-wk-old SHR, a time when significant glomerular injury was already present. They observed an average increase in glomerular pressure of only 4 mmHg compared with similarly aged WKY rats. Once again, whether glomerular hypertension was the cause or a consequence of injury in this study cannot be determined.

To our knowledge, the present study is the first to document the onset of glomerular hypertension before the appearance of glomerular injury in SHR. As recently reviewed (42), Pgc is determined by the MAP and the relative resistances of the afferent and efferent arterioles. Increases in MAP or RE or declines in RA tend to cause Pgc to rise. As shown in Table 2, RE did not increase with aging in SHR and, therefore, could not account for the development of glomerular hypertension in older SHR. Instead, the increase in Pgc resulted from the rise in renal perfusion pressure combined with a 35% decline in preglomerular vascular resistance.
The relative contributions of the increase in MAP and the decline in RA to the change in Pgc can be assessed by an analysis of the equation for RA (Eq. 1), in which RA is calculated as the pressure drop from renal artery to glomerular capillary divided by the initial glomerular blood flow (GBF)

$$RA = (MAP - Pgc)/GBF$$  \hspace{1cm} (1)

Solving for Pgc yields Eq. 2, from which one may predict that increases in MAP or declines in RA * GBF will be associated with an increase in Pgc. In the present study, MAP increased by ~10% in older rats, and this would tend to increase Pgc. However, at constant resistance, a rise in pressure will be associated with an increase in blood flow rate. In fact, although not statistically significant, the mean value for afferent blood flow (Q˙A) was ~25% greater in old than in young SHR. The effect of an increase in GBF if Ra remains constant would be an increase in RA * GBF as well, and this would have offset the effect of MAP to increase Pgc. Therefore, although the observed increase in MAP in older SHR tended to cause Pgc to rise, there would have been little net increase in Pgc if RA had not also declined.

The calculation of RA is based on determination of five separate parameters during the micropuncture study (MAP, Pgc, SNGFR, and the afferent and efferent arteriolar protein concentrations). Because the inherent variability in each of these measurements is combined in the resistance calculation, the standard deviation of this parameter is typically large, and it may be difficult to demonstrate statistically significant differences on the basis of a reasonable number of experiments. Therefore, despite the fact that the magnitude of the difference in RA between young and old rats was not great enough to be statistically significant, it nevertheless appears to have been biologically relevant. RA also tended to be lower in older SHR, and this tended to offset the effects of the reduction in RA on Pgc. However, RA/RE also tended to increase, by ~10%, with aging in SHR, consistent with a proportionally greater decrease in RA than in RE.

Although onset of glomerular hypertension with aging has been previously reported, an increase in Pgc was evident at a much earlier age in SHR than in the normal rats studied by Anderson (3) or SHR studied by Komatsu et al. (27). Glomerular sclerosis is also an early event in SHR compared with normotensive rats and is observed in SHR kidneys at 1 yr of age (20). It is attractive to speculate that the premature increase in glomerular pressure and injury in SHR results, at least in part, from the marked elevation in systemic blood pressure in these rats. Our data are not entirely consistent with the hypothesis of Bauer et al. (6). According to their theory, afferent vasodilation and glomerular hypertension develop in response to vascular disease and ischemic loss of nephrons. However, neither vascular nor glomerular lesions were observed in SHR at 9 mo of age at the time when Pgc was already elevated. Instead, the decline in RA appeared to be functional in nature. To determine the mechanism of this effect, we compared the relative sensitivity of the renal vasculature to NO, ET, and ANG II in young and old rats. Quantitatively similar constrictor responses to NO blockade and ET-1 administration were observed in young and old rats (Figs. 2 and 3), suggesting that alterations in the activity of these vasoactive mediators did not account for glomerular hypertension in older SHR. These findings are somewhat different from those observed in aging normotensive rats, in which enhanced vasoconstrictor response to blockade of NO production (24, 33) and infusion of ET (37) is reported. The explanation for these divergent findings is uncertain but may result from the differences in the age, gender, and strain examined as well as the absence of renal injury in rats in the present study.

A somewhat different response was observed when ANG II was infused. A proportionally greater decline in GFR than RPF was observed in old rats, leading to a significant reduction in FF. This pattern of change is consistent with the hypothesis that infused ANG II preferentially constricted the afferent arteriole in older SHR. If this is so, then decreased endogenous production of ANG II may have contributed to the age-associated decline in RA in SHR. Alternatively, a proportionally greater reduction in GFR than in RPF might result if exogenous ANG II caused the ultrafiltration coefficient (Kf) to decline significantly. ANG II has been demonstrated to reduce Kf in rats, presumably by inducing constriction of mesangial cells, thereby reducing the glomerular capillary surface area available for ultrafiltration. However, no significant differences in Kf were observed between young and old SHR (Table 2) or between old SHR treated or untreated with enalapril (data not shown). Therefore, there is no direct evidence in the present study that supports the notion that ANG II produced a greater decline in GFR than in old SHR.

Some controversy exists regarding the precise localization of the vasoconstrictor effects of ANG II on the renal microcirculation. ANG II exerts its effects on glomerular hemodynamics by binding to specific receptors that are widely distributed along the renal microvasculature (19). Myers et al. (30) were the first to examine the influence of ANG II on the preglomerular, glomerular, and postglomerular pressures and flows in a mammalian species. An intravenous infusion of a mildly pressor dose of ANG II resulted in relative stability of SNGFR, despite a significant decline in Qa in normal, hydropenic rats. This fall in Qa was associated with increases in RA and RE, presumably the result of constriction of afferent and efferent arterioles. Of interest to the present study is the extent to which the rise in RA with ANG II was a direct effect of the hormone or was dependent on the marked rise in renal perfusion pressure that occurred in response to the relatively high dose of ANG II employed in these studies. To examine this question, Myers et al. carried out experiments with an identical dose of ANG II in a separate group of Munich-Wistar rats in which constriction of the aorta prevented the rise in renal perfusion.
pressure. When the aorta was constricted during infusion of ANG II, the increase in $R_A$ with ANG II was strikingly attenuated. These studies suggested that ANG II preferentially constricts the efferent arteriole.

Somewhat different results have been obtained using techniques in which responses of the preglomerular and postglomerular vessels to angiotensin have been directly visualized. For example, in studies of the effects of ANG II on isolated perfused afferent and efferent arterioles, although the efferent arteriole was somewhat more sensitive, constriction of both vessels was noted in response to ANG II (15, 25). Consistent with this view, studies in the hydronephrotic rat kidney revealed that blockade of angiotensin receptors with saralasin was associated with dilatation of the arcuate, interlobular, and afferent arteriole as well as of the efferent arteriole, even under controlled arterial pressure (36). Similar observations have also been made in the blood-perfused juxtamedullary nephron preparation (12–14). Taken together, the data clearly indicate that ANG II can directly raise preglomerular vascular resistance in some settings.

Considerable evidence exists for a decline in activity of the renin-angiotensin system with aging. Plasma renin levels decrease with aging in humans (41), in normal rats (32), and in SHR (35). Single-nephron levels also decline in normal rats (26, 34). Baylis (7) compared renal hemodynamic responses to infused ANG II and ANG II blockade in conscious normotensive rats at 3–5 or 19–22 mo of age. In contrast to the present study, similar hemodynamic responses were observed in young and old rats in response to infused ANG II. When the renin-angiotensin system was blocked, renal vasodilation was seen in old, but not in young, rats, consistent with an increased effect of endogenous ANG II on renal hemodynamics in older rats. In contrast, Corman and Michel (16) failed to observe a significant effect of converting enzyme inhibition on RPF or GFR in female Wistar rats at 30 mo of age.

In summary, a rise in blood pressure and functional decline in preglomerular vascular resistance develop with aging in SHR, leading to glomerular hypertension that antedates the appearance of significant vascular or glomerular injury in these rats. Responses to manipulations of several vasoactive systems in SHR suggest that the changes in renal hemodynamics in older rats result from a decline in endogenous activity of the renin-angiotensin system predominantly at the level of the afferent arteriole. These data are consistent with the hypothesis that glomerular injury and progressive loss of kidney function in SHR result, at least in part, from the late onset of glomerular hypertension in this strain. It is attractive to speculate that a similar sequence of events accounts for hypertensive nephrosclerosis and progression to end-stage renal disease in a subset of patients with essential hypertension.

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