Temporary Treatment of Prepubescent Rats with Angiotensin Inhibitors Suppresses the Development of Hypertensive Nephrosclerosis

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Abstract. Hypertensive nephrosclerosis is a leading cause of end-stage renal disease; therefore, strategies to prevent the development of renal disease require close study. Here it is demonstrated that transient treatment of prepubescent rats with angiotensin inhibitors attenuated their susceptibility to the development of hypertensive nephrosclerosis after maturation.

Stroke-prone spontaneously hypertensive Izumo strain rats were divided into four groups, treated with vehicle, the angiotensin-converting enzyme inhibitor (ACEI) delapril (40 mg/kg per d), the angiotensin receptor antagonist (AT1R-Ant) candesartan cilexetil (1 mg/kg per d), or the vasodilator hydralazine (25 mg/kg per d) from weaning to puberty (3 to 10 wk of age), and then monitored without treatment for 6 mo. BP in the ACEI- and AT1R-Ant-treated groups remained significantly decreased, compared with the untreated and hydralazine-treated groups. Moreover, marked proteinuria and nephrosclerosis developed in the untreated and hydralazine-treated groups at 30 wk but were suppressed in the ACEI- and AT1R-Ant-treated groups. Of interest, plasma renin activity, plasma angiotensin II concentrations, and renal renin mRNA levels were reduced by 

Recent studies using knockout mice demonstrated that the R-A system also plays an important role in kidney development. Targeted deletion of components of the R-A system (angiotensinogen, ACE, or AT1 receptors) resulted in the development of kidneys with multiple abnormalities, including vascular hypertrophy, mesangial expansion, and tubular atrophy (for review, see reference 6). Interestingly, the same changes could be produced by treating newborn rats from 0 to 3 wk with ACEI or AT1R-Ant, suggesting that these 3 wk represent a critical time window for the developmental effect of the R-A system on kidney maturation (6).

In this study, we examined the effects of treatment of these rats with angiotensin inhibitors at a later stage of infancy, i.e., from weaning (3 wk) to puberty (10 wk), on the development of renal lesions at 6 mo, well after discontinuation of the treatments. Our results suggest that, unlike treatment from 0 to 3 wk, treatment from 3 to 10 wk suppresses the development of renal lesions in this model. Moreover, we found that permanent changes in the R-A system may be involved in the striking beneficial effects of these treatments.

Materials and Methods
Animal Treatments

Studies were conducted using 3-wk-old, male, SHRSP Izumo strain (SHRSP/Izm) rats maintained by the Disease Model Cooperative Research Association (Kyoto, Japan). All experiments were performed in accordance with the animal experimentation guidelines of
Keio University School of Medicine. Rats were allowed free access to standard rat chow (CE-2; Nippon, Clea, Japan) containing sodium (0.26 g/100 g) and potassium (1.06 g/100 g). Drinking water was monitored daily, using drinking bottles. Rats were housed five/cage except for the period of urine collection for protein measurements, when they were housed individually in metabolism cages. Rats were randomly divided into four groups. Rats in group 1 (n = 6) were untreated SHRSP/Izm rats. Rats in groups 2 (n = 8) and 3 (n = 8) were SHRSP/Izm rats treated with the ACEI delapril (0.4 g/L in drinking water; the calculated dose for a 200-g rat drinking 20 ml/d is 40 mg/kg per d) or the AT1R-Ant candesartan cilexetil [dissolved in drinking water at 0.01 g/L, as described by Mackenzie et al. (7); the calculated dose for a 200-g rat drinking 20 ml/d is 1 mg/kg per d], respectively, for 7 wk (from 3 to 10 wk of age). Rats in group 4 (n = 5) were SHRSP/Izm rats treated with the vasodilator hydralazine (0.26 g/100 g) and potassium (1.06 g/100 g). Drinking water was provided at 50 mM Tris-HCl (pH 8.3), 50 mM KCl, 2 mM MgCl₂, 0.2 mM dNTP, 15 pmol of each primer, 5 µCi of [³²P]dCTP, and 2.5 µL of Taq polymerase, using a Perkin-Elmer-Cetus thermal cycler for 25 cycles. Renin primers were 5'-GGCACTTGTGTGTGAGG-3' and 5’-ACCCGATGCTGATTGTATGCCC-3’, corresponding to the sense and antisense sequences of bases 851 to 870 and 1203 to 1224, respectively, in the rat renin sequence. AT1 receptor primers were 5'-GGAAACAGCTTGTTGTGTGAGG-3' and 5'-ACCCGATGCCTGATTGTATGCCC-3', corresponding to the sense and antisense sequence of bases 113 to 130 and the antisense sequence of bases 719 to 739 in the rat AT1a and AT1b receptor sequences, respectively. AT2 receptor primers were 5’-ATGAAGGACATCACTTACATACATG-3', corresponding to the sense sequence of bases 1 to 23 and the antisense sequence of bases 478 to 499, respectively. GAPDH primers were 5’-TCCCTCAAGATTGTCGACAAGC-3' and 5’-AGATCCACAACCCTATACATTCC-3', corresponding to the sense sequence of bases 451 to 470 and the antisense sequence of bases 739 to 758, respectively. Plasma renin activity (PRA) was determined by RIA of AngI formed by incubation of plasma for 1 h at 37°C. Plasma AngII levels were analyzed by RIA as described previously (8).

Histologic Studies
Kidneys and thoracic aortae were fixed in 10% phosphate-buffered formalin and then embedded in paraffin blocks. Histologic sections from the rat kidneys were stained with periodic acid-Schiff stain, and sections from aortae were stained with Azan (Mitsubishi Kagaku, Tokyo, Japan). Slides were examined by light microscopy, and renal histopathologic changes were scored as described previously (2). For assessment of glomerular damage, the number of glomeruli exhibiting focal or global ischemic or proliferative damage was enumerated and expressed as a percentage of the total number of glomeruli examined. Blood vessels were graded 0 to 4 for arteriolar sclerosis, on the basis of the severity of hyalinosis and thickening of the vascular wall. Tubulointerstitial changes, including interstitial inflammation and tubular atrophy, were assessed and graded 0 to 3, as follows: grade 1, involvement of <20% of the cortical interstitium; grade 2, involvement of 20 to 40% of the interstitium; grade 3, involvement of >40% of the interstitium.

Reverse Transcription-PCR Analysis of Renal Gene Expression
Total RNA was purified from the kidneys of five animals in each group, using the acid guanidinium-phenol-chloroform method, and was quantified by spectrophotometric measurement of absorbance at 260 nm. Renin and AT1 and AT2 receptor subtype mRNA were analyzed by reverse transcription-PCR (RT-PCR), as reported by us previously (2,9). In brief, 1 µg of total RNA was reverse-transcribed in a reaction mixture containing 10 mM Tris-HCl (pH 8.3), 50 mM KCl, 5 mM MgCl₂, 1 mM dNTP, 1 U of RNase inhibitor, 2.5 µM (50 pmol) random hexamers, and 2.5 U of Moloney murine leukemia virus reverse transcriptase, in a volume of 20 µl. The reverse-transcribed product was amplified with renin, AT1 receptor, AT2 receptor, or glyceraldehyde-3-phosphate dehydrogenase (GAPDH) sense and antisense primers, in a reaction mixture containing 10 mM Tris-HCl (pH 8.3), 50 mM KCl, 2 mM MgCl₂, 0.2 mM dNTP, 15 pmol of each primer, 5 µCi of [³²P]dCTP, and 2.5 U of Taq polymerase, using a Perkin-Elmer-Cetus thermal cycler for 25 cycles. Renin primers were 5’TGGCCACCTGTGGGTGAGG-3’ and 5’-ACCCGATGCGATTGTATGCCC-3’, corresponding to the sense and antisense sequences of bases 851 to 870 and 1203 to 1224, respectively, in the rat renin sequence. AT1 receptor primers were 5’-GGAAACAGCTTGTTGTGTGAGG-3’ and 5’-ACCCGATGCCTGATTGTATGCCC-3’, corresponding to the sense sequence of bases 113 to 130 and the antisense sequence of bases 719 to 739 in the rat AT1a and AT1b receptor sequences, respectively. AT2 receptor primers were 5’-ATGAAGGACATCACTTACATACATG-3’, corresponding to the sense sequence of bases 1 to 23 and the antisense sequence of bases 478 to 499, respectively. GAPDH primers were 5’-TCCCTCAAGATTGTCGACAAGC-3’ and 5’-AGATCCACAACCCTATACATTCC-3’, corresponding to the sense sequence of bases 451 to 470 and the antisense sequence of bases 739 to 758, respectively. To assess the relative levels of AT1a and AT1b receptor- amplified PCR products, the PCR products were incubated for 90 min at 37°C in the presence of EcoRI (10 U). Because the AT1a receptor (but not the AT1b receptor) contains an internal EcoRI site, EcoRI digestion under these conditions results in two fragments (428 and 178 bp in length) in the case of AT1a receptor DNA and one fragment (606 bp in length) in the case of AT1b receptor DNA (2). Levels of transforming growth factor-β (TGF-β) subtype mRNA were assessed using primers specific for TGF-β1, TGF-β2, and TGF-β3 isoforms, as described in detail by Gao et al. (10). Preliminary experiments confirmed that these PCR were performed within the linear phase of the PCR amplification. Reaction products were resolved by electrophoresis through 8% polyacrylamide gels. Gels were dried using a gel dryer, and incorporated radioactivity in each band was quantified using a laser image analyzer (model BAS 2000; Fuji Film Co., Tokyo, Japan).

Materials
Delapril and candesartan cilexetil were generously provided by Takeda Chemical Industries (Osaka, Japan). RT-PCR and electrophoresis reagents were obtained from Perkin Elmer (Branchburg, NJ) and Bio-Rad (Hercules, CA). Other chemicals were from Sigma Chemical Co. (St. Louis, MO).

Statistical Analyses
Results are expressed as mean ± SEM. Statistical comparisons were made by ANOVA, followed by Scheffe’s F test for comparisons between groups. Values of P < 0.05 were considered statistically significant.

Results
Effects of Prepubescent Treatment with Angiotensin Inhibitors on BP and Cardiovascular Hypertrophy in SHRSP/Izm Rats
As shown in Figure 1, the time period from 3 to 10 wk corresponded to the time of development of hypertension in...
untreated rats. The rats in groups 2 to 4, which were treated with ACEI, AT1R-Ant, or hydralazine, respectively, exhibited decreased BP, compared with the untreated rats, during the time of treatment. No significant differences in BP were observed between the different treatment groups at that stage. After cessation of treatment at 10 wk, BP continued to increase in the treated groups, whereas the BP was already at a plateau level in the untreated control rats. Interestingly, the BP in the hydralazine-treated rats attained a level similar to that in the control rats at 16 wk of age, whereas the BP in the ACEI- or AT1R-Ant-treated rats remained below 200 mmHg.

After euthanasia of the rats at 30 wk, the hearts and aortae were assayed and yielded consistent results, as demonstrated in these findings, the changes in plasma AngII levels were also observed in the ACEI- and AT1R-Ant-treated groups, compared with either control or hydralazine-treated rats. Representative histologic sections are shown in Figure 3. In the control SHRSP and hydralazine-treated rats, prominent thickening of small to medium-sized arteries were observed. These changes were dramatically reduced in the ACEI- and AT1R-Ant-treated groups.

Effects of Prepubescent Treatment with Angiotensin Inhibitors on Proteinuria and Renal Histologic Changes at 30 Wk in SHRSP/Izm Rats

Urinary protein levels were examined at 14 and 30 wk, as shown in Figure 2a. At 14 wk, proteinuria had not yet developed in SHRSP/Izm rats (comparative values for normotensive WKY/Izm rats were 5.4 ± 1.3 mg/100 g per d). In contrast, marked proteinuria was evident at age 30 wk for the control rats. This development of proteinuria was completely suppressed in the ACEI- and AT1R-Ant-treated groups (to 6.5 ± 1.2 and 7.9 ± 2.9 mg/100 g per d, respectively), whereas no such suppression was observed in the hydralazine-treated group.

The histologic findings for the different groups are presented in Figure 2b. As evident from the histologic scoring, suppression of the glomerular, vascular, and interstitial changes were observed in the ACEI- and AT1R-Ant-treated groups, compared with either control or hydralazine-treated rats. Representative histologic sections are shown in Figure 3. In the control SHRSP and hydralazine-treated rats, prominent thickening of small to medium-sized arteries were observed. These changes were dramatically reduced in the ACEI- and AT1R-Ant-treated groups.

Effects of Prepubescent Treatment with Angiotensin Inhibitors on PRA and Plasma AngII Levels at 30 Wk in SHRSP/Izm Rats

PRA values for the different groups were as follows: control, 13.1 ± 2.4 ng/ml per h; ACEI, 6.0 ± 1.3 ng/ml per h; AT1R-Ant, 6.3 ± 0.6 ng/ml per h; hydralazine, 15.7 ± 6.7 ng/ml per h. These data revealed that PRA was decreased to <50% in the angiotensin inhibitor-treated groups. To confirm these findings, the changes in plasma AngII levels were also assayed and yielded consistent results, as demonstrated in Figure 4.

Effects of Prepubescent Treatment with Angiotensin Inhibitors on Renal Gene Expression at 30 Wk in SHRSP/Izm Rats

Renal expression of renin mRNA was examined using our previously reported RT-PCR method. As presented in Figures

Table 1. Parameters of cardiovascular hypertrophy in treated and untreated SHRSP/Izm rats

<table>
<thead>
<tr>
<th></th>
<th>SHRSP Control (n = 6)</th>
<th>Delapril (n = 8)</th>
<th>Candesartan (n = 8)</th>
<th>Hydralazine (n = 5)</th>
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<tr>
<td>Blood pressure (mmHg)</td>
<td>228 ± 2</td>
<td>185 ± 5&lt;sup&gt;c,e&lt;/sup&gt;</td>
<td>184 ± 2&lt;sup&gt;c,e&lt;/sup&gt;</td>
<td>223 ± 3</td>
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<tr>
<td>Kidney weight (g)</td>
<td>1.41 ± 0.05</td>
<td>1.34 ± 0.04</td>
<td>1.35 ± 0.03</td>
<td>1.26 ± 0.07</td>
</tr>
<tr>
<td>Kidney weight/body weight (g/100 g)</td>
<td>0.45 ± 0.02</td>
<td>0.45 ± 0.03</td>
<td>0.44 ± 0.02</td>
<td>0.49 ± 0.07</td>
</tr>
<tr>
<td>Aortic wall thickness (media/lumen ratio × 100)</td>
<td>8.6 ± 0.1</td>
<td>9.1 ± 0.3</td>
<td>8.4 ± 0.2</td>
<td>8.8 ± 0.2</td>
</tr>
<tr>
<td>Heart weight (g)</td>
<td>1.68 ± 0.05</td>
<td>1.46 ± 0.05&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.49 ± 0.06</td>
<td>1.52 ± 0.18</td>
</tr>
<tr>
<td>Heart weight/body weight (g/100 g)</td>
<td>0.55 ± 0.02</td>
<td>0.48 ± 0.02&lt;sup&gt;b,d&lt;/sup&gt;</td>
<td>0.48 ± 0.03&lt;sup&gt;b,d&lt;/sup&gt;</td>
<td>0.55 ± 0.04</td>
</tr>
<tr>
<td>Body weight (g)</td>
<td>315 ± 7</td>
<td>300 ± 7</td>
<td>307 ± 7</td>
<td>283 ± 10</td>
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</table>

<sup>a</sup> Results are mean ± SEM. SHRSP/Izm, stroke-prone spontaneously hypertensive Izumo strain.
<sup>b</sup> P < 0.05, <sup>c</sup> P < 0.01 versus SHRSP control; <sup>d</sup> P < 0.05, <sup>e</sup> P < 0.01 versus hydralazine.
5 and 6, renin/GAPDH mRNA ratios were reduced to <50% in the angiotensin inhibitor-treated groups. In contrast, no such changes in the mRNA for angiotensin receptor subtypes (AT1a, AT1b, and AT2) were detected. TGF-β1 mRNA levels were also significantly (P < 0.05) reduced in the angiotensin inhibitor-treated groups, whereas no significant changes in TGF-β2 or TGF-β3 mRNA levels were observed (Table 2).

Discussion

The R-A system plays an important role in the control of BP and renal function. Recent evidence, particularly from targeted deletion of components of the R-A system, has demonstrated that the R-A system may also be involved in the development of normal kidneys. Experiments using ACEI and AT1R-Ant have demonstrated that the period from 0 to 3 wk after birth is
the critical time window for the actions of the R-A system in kidney development (6,11).

Other studies have shown that the use of ACEI or AT1R-Ant during a later period (up to 2 mo after birth) can attenuate the full development of hypertension in spontaneously hypertensive (SHR) rats (12–15). Wu and Berecek (13) reported that treatment of newborn rats with the ACEI captopril for 2 mo also attenuated central responses to AngI and AngII, suggesting alterations in the central R-A system. In other studies, the same group also noted decreases in plasma arginine vasopressin (AVP) levels (16) and alterations in endothelial function in these rats (17). However, the exact mechanisms for the changes in BP are still unclear.

In this study, we focused on the effects of angiotensin inhibitor treatment of prepubescent rats on the nephrosclerosis observed in SHRSP rats. These rats were derived from the parent SHR strain by selection of rats with a propensity to develop stroke and malignant nephrosclerosis while receiving a high-salt diet (18). With a normal-salt diet, these rats do not develop fulminant hypertension but develop nephrosclerosis with histologic changes very similar to the changes observed in human benign hypertensive nephrosclerosis, including characteristic sclerosis of small to medium-sized arteries in the kidney. We previously showed that the renal changes are not evident at 14 wk but are well developed after 22 wk (2).

In our study, treatment of prepubescent rats in the developmental stage of hypertension (from 3 to 10 wk) resulted in a decrease in the BP plateau reached after 14 wk, consistent with the results of studies using the parent SHR strain. The fact that this phenomenon was observed with both ACEI and AT1R-Ant demonstrated that the effect was attributable to inhibition of angiotensin actions at the AT1 receptor. Although ACEI also decrease the degradation of bradykinin and diminish stimulation of AT2 receptors (19), the mirroring of ACEI effects by AT1R-Ant makes it unlikely that these actions were involved in the observed effects. Moreover, this effect was not observed with the vasodilator hydralazine, demonstrating that the decrease in BP per se during the treatment period was not the main cause of this phenomenon. Of interest, we found that the heart weight/body weight ratios were lower in the angiotensin inhibitor-treated groups but aortic wall hypertrophy was not attenuated by treatment with angiotensin inhibitors. These results are similar to those observed using SHR rats (20).

We next examined proteinuria and renal histologic lesions in the different groups. In the angiotensin inhibitor-treated groups, the levels of proteinuria were similar to the levels observed for normotensive WKY/Izm rats of the same age. In other words, the development of proteinuria was completely suppressed by the interventions. Histologic examinations also revealed dramatic improvements in the renal lesions. In particular, marked renal arteriolar hypertrophy was observed in both the SHRSP control and hydralazine-treated groups but was virtually absent in the ACEI- and AT1R-Ant-treated groups. The development of nephrosclerosis in SHRSP rats is known to be accompanied by increases in TGF-β expression (3). In concert with the improvements in the histologic changes, reductions in TGF-β1 mRNA levels were observed in the kidneys of the angiotensin inhibitor-treated rats in this study.

Next, we examined potential mechanisms for the observed changes. As noted in the introduction, the inability to suppress renin activity has been implicated in the pathogenesis of the changes observed in SHRSP rats (4). In these rats, PRA increases progressively and is always significantly higher than that in normotensive WKY rats (5). We therefore examined the
R-A system in these rats and found that both PRA and plasma AngII levels were significantly reduced, compared with control SHRSP rats. Similar results were found at the mRNA level, suggesting that the excess renin production characteristic of this strain was attenuated in the treated rats.

It has been demonstrated that high levels of renin and hence of AngII are associated with cerebrovascular, renal, and cardiovascular lesions in SHRSP rats and that there is a correlation between PRA and the severity of cerebrovascular (4) and renal (4,21) damage. Therefore, it is possible that the decreases in PRA and AngII levels observed in our angiotensin inhibitor-treated rats played an important role in the observed beneficial effects of treatments with these agents.

It is presently unclear why treatment from 3 to 10 wk of age caused a sustained decrease in R-A activity in these rats. As mentioned above, the time window of 0 to 3 wk after birth has been demonstrated to correspond to the time window for the actions of the R-A system on the development of normal kidney morphologic features. We speculate that the time period from 3 to 10 wk may include the time window for the maturation of the adult R-A system. One possibility is that the R-A system has a positive feedback effect on its own development at this stage; therefore, blockade of the R-A system during this time period may result in permanent attenuation of the R-A system. Interestingly, a recent study by St. Lezin et al. (22), using congenic SHR strains, demonstrated that the susceptibility to hypertension-induced renal damage in SHR rats is genetically determined by a region on chromosome 1q. It would be interesting to determine whether genes affecting the activity and maturation of the R-A system are located in this region.

An important issue concerns the cause-and-effect relationship between nephrosclerosis and BP. The fact that hypertension precedes the development of nephrosclerosis in this model (2) suggests that hypertension is the cause of the renal damage, not vice versa. The question of whether the reduction in BP per se in the ACEI- and AT1R-Ant-treated rats contributed to the amelioration of the renal changes thus arises. Interestingly, Nakamura et al. (3) administered hydralazine to SHRSP rats from 12 to 24 wk of age and reduced the BP to approximately 160 mmHg, which is similar to the values for our ACEI- and AT1R-Ant-treated groups. Those authors found that BP reduction was effective in reducing both proteinuria and histologic changes in their rats, which suggests that the lower BP in our treated groups played a significant role in the suppression of

**Figure 6.** Quantitation of signals from RT-PCR analyses of renin and angiotensin receptor mRNA in the kidneys of 30-wk-old SHRSP/Izm rats. Rats were treated from 3 to 10 wk of age with vehicle (SHRSP), delapril (dela) (40 mg/kg per d), candesartan cilexetil (cand) (1 mg/kg per d), or hydralazine (hydra) (25 mg/kg per d). *, P < 0.05 versus vehicle-treated rats; +, P < 0.05 versus hydralazine-treated rats (n = 5/group).

**Table 2.** RT-PCR analysis of TGF-β isoform mRNA in the kidneys of treated and untreated SHRSP/Izm rats

<table>
<thead>
<tr>
<th>Isoform</th>
<th>SHRSP Control (n = 5)</th>
<th>Delapril (n = 5)</th>
<th>Candesartan (n = 5)</th>
<th>Hydralazine (n = 5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>TGF-β1/GAPDH</td>
<td>1.00 ± 0.23</td>
<td>0.35 ± 0.10b</td>
<td>0.43 ± 0.08b</td>
<td>0.72 ± 0.29</td>
</tr>
<tr>
<td>TGF-β2/GAPDH</td>
<td>1.00 ± 0.18</td>
<td>1.28 ± 0.14</td>
<td>0.65 ± 0.15</td>
<td>0.60 ± 0.38</td>
</tr>
<tr>
<td>TGF-β3/GAPDH</td>
<td>1.00 ± 0.17</td>
<td>0.96 ± 0.13</td>
<td>0.82 ± 0.11</td>
<td>1.20 ± 0.55</td>
</tr>
</tbody>
</table>

a Results are mean ± SEM. SHRSP control mean values were arbitrarily assigned the value 1. RT, reverse transcription; TGF-β, transforming growth factor-β; GAPDH, glyceraldehyde-3-phosphate dehydrogenase.

b P < 0.05 versus SHRSP control.
nephrosclerosis. However, the same group found that treatment with AT1R-Ant from 12 to 24 wk had a greater effect in abolishing the nephrosclerotic changes, suggesting that the combination of BP reduction and R-A system inhibition had the greatest nephroprotective effect. These findings are compatible with the assumption that BP reduction played a significant role in the attenuation of renal changes in our rats, with further protection being afforded by suppression of the R-A system.

Another interesting question is why the BP increased to reach values comparable to those for control untreated animals after the cessation of hydralazine therapy. Both Christensen et al. (23) and King et al. (24) reported a washout period for hydralazine of approximately 2 to 3 wk in rats, after which the hypotensive effect disappears. This probably explains why the effects of hydralazine were only temporary in this study. A related question is why the ACEI and AT1R-Ant had longer effects on BP reduction. One possibility is related to the “vascular amplifier” hypothesis. This hypothesis relates the establishment of hypertension to a positive feedback loop involving vascular hypertrophy and increases in BP, and it is the subject of ongoing debate (25). Interestingly, Adams et al. (15) reported that treatment of SHR rats either from 4 to 9 wk or from 4 to 14 wk resulted in long-term attenuation of SHR rat hindquarter resistance properties to values similar to those for normotensive WKY rats. Those authors proposed that the prevention of peripheral vascular changes in the young rats by the angiotensin inhibitors during this susceptible period could inhibit the development of the vascular amplifier mechanism, resulting in the observed long-term reductions in BP. However, Morton et al. (14) reported that treatment of SHR rats with either captopril or losartan from 3 to 7 wk of age resulted in decreased BP in the absence of changes in the mesenteric resistance artery media/lumen ratio. Those authors concluded that the persistent hypotensive effect was not related to the vascular structural changes. Therefore, the precise role of the vascular amplifier mechanism in the observed changes remains unclear. As mentioned above, Berecek and co-workers (13,16,17) have postulated that alterations in the central R-A system, decreases in plasma AVP levels, and alterations in endothelial function may be involved in the persistent hypotensive effect. These possibilities are not mutually exclusive, and we speculate that changes in the central and peripheral R-A systems, changes in the levels of other hormones such as AVP, and vascular functional and structural changes could all contribute to the sustained antihypertensive and nephroprotective effects of angiotensin inhibitors administered during the critical period in rat maturity.

One important clinical implication of this study is the possibility that patients with genetic susceptibility to renal disease might be treated for their innate susceptibility by intervention at an appropriate stage, before the signs of the disease become apparent. The timing of this intervention may be quite early, i.e., during the preadolescent stage. Of interest, studies by Raizada and co-workers (26,27) showed that a single injection of a retroviral vector containing antisense sequences for ACE or the AT1 receptor could cause permanent blockade of the R-A system. Moreover, the antisense sequence is integrated into the genome and is then transmitted to offspring, resulting in blockade of the R-A system in the F1 and F2 generations (26). Although these methods are effective in providing permanent cardiovascular protection in both the parents and the offspring (27) and offer possibilities for a total cure for hypertension and its sequelae, the effects may be too drastic and long-lasting for use in human subjects. The administration of orally active compounds for a limited time may be a more acceptable option. Obviously, it is not presently possible to extrapolate our results to other types of renal disease. However, the results do provide us with reasons to think that, in the future, genetic susceptibility to renal disease may be treated with appropriate interventions at an earlier stage of development. Because several renal diseases, including the most common autosomal dominant disease (polycystic kidney disease), have strong genetic components, we speculate that earlier intervention, perhaps with angiotensin inhibitors, may shift the therapeutic paradigm for renal disease away from the management of established disease and toward more effective disease prevention.

References


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