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The effect of angiotensin II receptor antagonists on kidney function in two-kidney, two-clip Goldblatt hypertensive rats

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Abstract

The effect of blockade of the renin–angiotensin system on kidney function using non-peptide angiotensin AT_1 receptor antagonists was investigated in renovascular hypertensive rats. An angiotensin converting enzyme inhibitor, captopril and two angiotensin AT_1 receptor antagonists, losartan and GR138950 (1-{[3-bromo-2[2-[[(trifluoro-methyl)sulphonyl]amino]phenyl]-5 benzofuranyl]methyl}-4-cyclopropyl-2-ethyl-1H-imidazole-5-carboxamide) were administered in Na^+-deplete two-kidney, two-clip Goldblatt hypertensive rats over a 3-day period. Captopril, losartan (30 mg/kg body weight) and GR138950 (5 mg/kg body weight) significantly ($P < 0.001$) lowered the systolic blood pressure in the hypertensive rats from 290 ± 5, 252 ± 9 and 238 ± 13 mmHg to 152 ± 17, 148 ± 9 and 123 ± 6 mmHg, respectively. The magnitude of reduction in blood pressure in these three groups of rats was similar and occurred with comparable marked increases in plasma levels of urea and creatinine indicative of acute renal failure. These findings demonstrate an important role for angiotensin II in the maintenance of renal function during blood pressure reduction in renovascular hypertensive states during restriction of dietary Na^+ intake. © 1997 Elsevier Science B.V.

Keywords: Renovascular hypertension; Renal failure, acute; Angiotensin converting enzyme inhibition; Na^+ depletion; Angiotensin AT_1 receptor antagonists, non-peptide

1. Introduction

It is recognised that the renin–angiotensin system plays an important role in the maintenance of the cardiovascular system and renal function in both normotensive and hypertensive animals and humans. A blockade of the renin–angiotensin system has been proven to be beneficial in the treatment of essential and renal hypertension, the prevention and reversal of cardiac hypertrophy together with a reduction in proteinuria.

It is evident that the angiotensin converting enzyme inhibitors reduce proteinuria and improve the morbidity and mortality particularly in patients with renal failure due to diabetic nephropathy (Johnston, 1995); yet, it is apparent that they may cause a deterioration in renal function in patients with bilateral renal artery stenosis, renal artery stenosis in a transplanted kidney, stenosis of the artery supplying a solitary kidney, in severe congestive cardiac failure and volume depletion (Johnston, 1995). The original peptide angiotensin II receptor antagonist, saralasin, was used initially to obtain a better understanding of the role of angiotensin II because of the relative non-specificity of the angiotensin converting enzyme inhibitors (Helmchen et al., 1982) but because of its known agonist actions of raising blood pressure in some patients (Case et al., 1976) it has not been developed as a clinical tool.

More recently, non-peptide angiotensin II receptor antagonists have been discovered and developed of which losartan is the first of this new class of agents to reach the clinics. These compounds have the potential of acting as effective antihypertensive agents but appear to be without the side effects of the angiotensin converting enzyme inhibitors. However, whether this class of compounds might have adverse effects on renal function in renovascular hypertensive patients is now an important question. A number of experimental studies have found that losartan lowered blood pressure to the same extent as captopril in a

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high renin hypertensive rat model, the renal artery ligated rat whereas in this situation saralasin had a restricted hypotensive action (Wong et al., 1990). In one-kidney, one-clip (1K1C) Goldblatt hypertensive Na^+-restricted rats, losartan lowered systemic blood pressure and renal vascular resistance to a similar degree to enalapril over a 6 day study period, but enalapril reduced glomerular filtration rate to a greater extent than losartan (Demeilliers et al., 1995). Imamura et al. (1995) also reported that both losartan and enalapril reduced proteinuria in 2K1C hypertensive rats as well as lowering systemic blood pressure, increasing glomerular filtration rate and renal plasma flow in the non-clipped kidney without altering renal haemodynamics in the clipped kidney despite a reduced renal perfusion pressure. Furthermore, Inada et al. (1994) reported that a single dose of losartan caused a dose-dependent antihypertensive effect in normo-reninaemic 1K1C hypertensive rats for more than 24 h, while in normo-reninaemic two-kidney, two-clip (2K2C) hypertensive rats Basso et al. (1995) found that EXP 3174, the active metabolite of losartan, acutely lowered blood pressure suggesting that a vascular renin–angiotensin system might contribute to the high blood pressure.

Although it has been established that the non-peptide angiotensin II receptor antagonists can effectively lower blood pressure in experimental rat models of renal hypertension, their impact on renal function has yet to be assessed in detail. Recently we developed a rat model of renovascular hypertension based on the report of Helmchen et al. (1982), in which kidney function was dependent on angiotensin II in so far as angiotensin converting enzyme inhibitors induced renal failure (Abdi and Johns, 1996). The objective of this study was to determine whether non-peptide angiotensin II receptor antagonists would also cause a deterioration of renal function under these conditions.

2. Materials and methods

2.1. Animal preparation

Male Wistar rats were purchased from Charles River and fed a normal Na^+ diet (Lillico, Surrey, UK), with a Na^+ content of 0.32 wt% and offered tap water ad libitum. They were kept in cages, 6 per cage, in an automatically lighted room (8.00 a.m. to 8.00 p.m.) at a constant temperature (22 ± 1°C). When animals reached an appropriate weight of 130–170 g, they were anaesthetised with a mixture of O_2/N_2O/fluothane and silver clips, 0.25 mm internal diameter, were slipped around each renal artery as close as possible to its exit from the aorta. The wound was sutured and the animals were allowed to recover. The animals were closely observed during the immediate post-operative recovery period and kept in individual cages for 3 to 4 days after which they were housed in groups of 3 to 4 animals in a continuously ventilated cage.

The animals were maintained for a further six weeks before they entered the metabolic study. Four weeks following surgery, they were fed a 'no' Na^+ diet (virtually Na^+ free ICN 902 diet, ICN Pharmaceuticals, Irvine, CA, USA) plus distilled water. The animals remained on this diet for two weeks before entering the metabolic study.

2.2. Metabolic study

The animals were placed in the metabolic cages, initially for 24 h, 2–3 days before the main study, in an attempt to allow them to acclimatise and during this time urine flow and water intake were determined gravimetrically and systolic blood pressure was measured using a tail cuff sphygmomanometer. To study the effect of drugs, the animals were placed in the metabolic cages at 8.00 p.m. and urine collections and water intake were measured every 24 h. The experimental drugs were given by gavage in a volume of 3 ml/kg body weight (b.wt.) with either tap water or distilled water as vehicle once a day between 9.00 and 10.00 p.m. Systolic blood pressure was measured each morning of days 1, 2 and 3 between 10.00 and 12.00 a.m. On the afternoon of day 4, the animals were briefly anaesthetised, using O_2/N_2O/fluothane, a carotid artery was cannulated and a blood sample was taken into EDTA (to achieve a concentration of 1 mg/ml); it was immediately centrifuged and the plasma placed into a -20°C deep freeze. Urine samples were stored deep frozen until assay. Urinary and plasma Na^+ was estimated using flame photometry (Corning, 410C, Halsted, Essex, UK), plasma creatinine and urea levels were measured using routine methodologies in the Clinical Chemistry Department of the Queen Elizabeth Hospital (Birmingham, UK). Plasma renin activity levels were estimated using a commercial radioimmunoassay kit (C.I.S., UK).

2.3. Blood pressure measurement

Systolic blood pressure was measured by means of the tail cuff technique using the Harvard apparatus system. Over the 6-week period of hypertension development, the animals were trained to accept the tail cuff by having the systolic blood pressure measured once per week.

2.4. Animal groups and drugs

The doses of the drugs (captopril, losartan and GR 138950, 1-{[3-bromo-2[2-[[(trifluoro-methyl)-sulphonyl]amino]phenyl]-5 benzofuranyl]methyl}-4-cyclopropyl-2-ethyl-1H-imidazole-5-carboxamide) were chosen such that approximately equivalent reductions in the blood pressure were achieved and they corresponded approximately to those used by others.

The animals were divided into 4 groups:

Group I, vehicle-treated rats: distilled water (3 ml/kg b.wt. per 24 h) was given by gavage once a day between 9.00 and 10.00 p.m. Six animals were studied with a body weight of 314 ± 7 g and kidney weight of 528 ± 27 mg/100 g b.wt.

Group II, captopril-treated rats: Captopril was dissolved in distilled water and given by gavage (3 ml/kg b.wt. per 24 h) once a day between 9.00 and 10.00 p.m. such that the animals received 30 mg/kg b.wt. Eight animals were studied with a body weight of 308 ± 11 g and kidney weight of 655 ± 33 mg/100 g b.wt.

Group III, losartan-treated rats: Group III rats were divided into two groups. In group III_a, animals received losartan 15 mg/kg b.wt. (11 animals, body weight 308 ± 11 g, kidney weight 669 ± 35 mg/100 g b.wt) once a day between 9.00 and 10.00 p.m. and group III_b animals were given 30 mg/kg b.wt. losartan (eleven animals, body weight 334 ± 13 g, kidney weight 636 ± 12 mg/100 g b.wt.) once a day between 9.00 and 10.00 p.m. The drug was dissolved in distilled water and given by gavage (3 ml/kg b.wt. per 24 h).

Group IV, GR138950-treated rats: The drug (potassium salt) was dissolved in distilled water and given by gavage (3 ml/kg b.wt. per 24 h). In group IV (eight animals with body weight of 350 ± 16 g and kidney weight of 616 ± 27 mg/100 g b.wt.), 5 mg/kg b.wt. of GR138950 was given by gavage once per day for three days between 9.00 and 10.00 p.m.

2.5. Statistics

The means $\pm$ S.E.M are given and the percentage and absolute changes quoted in the text are the averages generated for each group of rats. All data were compared using unpaired Student's *t*-test as appropriate and significance was taken when $P < 0.05$.

3. Results

Table 1 indicates that all groups of bilaterally clipped rats fed with a 'no' Na^+ diet had developed hypertension before entering the metabolic study, having systolic blood pressures markedly elevated compared with the normotensive rats studied previously (Abdi and Johns, 1996).

3.1. Group I, vehicle administration

The blood pressure of the vehicle-treated rats showed little variation over the three days of vehicle administration (Table 1). The plasma renin activity of the group was 32.3 ± 1.3 ng Ang I/ml per h which indicated the activation of renin–angiotensin system in Na^+-depleted bilaterally clipped rats. Plasma concentrations of urea and creatinine were 7.2 ± 0.6 mmol/l and 44.8 ± 1.5 μmol/l, respectively, which were comparable with those of normotensive rats fed a 'no' Na^+ diet (Abdi and Johns, 1996). Urine flow, water intake and Na^+ excretion of this group remained at a stable level during the three days of vehicle administration (Table 2).

3.2. Group II, captopril treatment

Table 1 shows that captopril caused a significant ($P < 0.001$) fall in blood pressure of the group II rats of some 134 mmHg in the first 24 h which was sustained for the subsequent 48 h and the average fall in blood pressure during the three days of captopril administration was 138 ± 16 mmHg. The plasma concentrations of urea and creatinine were significantly ($P < 0.01$) higher than vehicle-treated group (45.1 ± 9.0 mmol/l and 189.9 ± 29.2 μmol/l, respectively).

The final plasma renin activity of captopril-treated group was 11.0 ± 2.5 ng Ang I/ml per h which was significantly lower ($P < 0.01$) compared to those of the vehicle-treated

Table 1
Effect of captopril, losartan and GR138950 on systolic blood pressure and plasma urea and creatinine levels in 2K2C-GHT rats

Group	Systolic blood pressure (mmHg)					Plasma level	
	Control	24 h	48 h	72 h	Average change	Urea (mmol/l)	Creatinine (μmol/l)
Vehicle ($n = 6$) 3 ml/kg b.wt. per 24 h	277 ± 15	271 ± 12	275 ± 11	262 ± 19	-6 ± 6	7.2 ± 0.6	44.8 ± 1.5
Captopril ($n = 8$) (30 mg/kg b.wt. per 24 h)	290 ± 5	156 ± 20 [c]	146 ± 17 [c]	152 ± 15 [c]	-138 ± 16 [f]	45.1 ± 9.0 [e]	189.9 ± 29.2 [e]
Losartan ($n = 11$) (15 mg/kg b.wt. per 24 h)	247 ± 5	183 ± 9 [c]	185 ± 9 [c]	172 ± 8 [c]	-67 ± 6 [f]	35.9 ± 5.0 [f]	127.0 ± 23.2 [d]
Losartan ($n = 11$) (30 mg/kg b.wt. per 24 h)	252 ± 9	167 ± 14 [c]	135 ± 11 [c]	134 ± 8 [c]	-108 ± 10 [f]	43.3 ± 5.8 [f]	155.9 ± 25.0 [e]
GR138950 ($n = 8$) (5 mg/kg b.wt. per 24 h)	239 ± 13	121 ± 11 [c]	130 ± 8 [c]	118 ± 8 [c]	-116 ± 14 [f]	43.4 ± 7.8 [e]	196.0 ± 47.9 [d]

Control represents the values obtained during the first 24 h in the cage before vehicle or drug administration while '24 h', '48 h' and '72 h' represent the values obtained during the first, second and third 24 h of drug treatment, respectively.

[a] $P < 0.05$.

[b] $P < 0.01$.

[c] $P < 0.001$, compared to the control values of the same group.

[d] $P < 0.05$.

[e] $P < 0.01$.

[f] $P < 0.001$, compared to vehicle-treated group.

Table 2
Effect of captopril, losartan and GR 138950 on fluid balance in hypertensive rats

Group	Water intake (ml/kg per h)				Urine flow (ml/kg per h)				Na^+ excretion (μmol/kg per h)			
	Control	24 h	48 h	72 h	Control	24 h	48 h	72 h	Control	24 h	48 h	72 h
Vehicle ($n=6$) 3 ml/kg b.wt. per 24 h	5.4±0.6	5.7±0.6	4.9±0.7	5.4±0.7	3.6±0.5	3.8±0.6	3.3±0.5	3.1±0.5	3.5±0.6	3.9±0.7	4.7±0.8	6.4±2.4
Captopril ($n=8$) (30 mg/kg b.wt. per 24 h)	4.8±0.5	3.2±0.6 [a]	3.5±1.1	5.7±1.1	3.0±0.5	1.5±0.7	2.2±0.6	4.0±0.9	8.7±2.7	14.2±6.4	21.6±7.2	23.2±4.3 [a]
Losartan ($n=11$) (15 mg/kg b.wt. per 24 h)	5.2±0.6	3.3±0.6 [a]	2.0±0.3 [c]	3.0±0.4 [b]	3.5±0.4	2.0±0.6	1.4±0.3 [c]	1.5±0.3 [c]	3.6±0.4	6.9±2.7	10.2±2.2 [b]	9.5±1.9 [b]
Losartan ($n=11$) (30 mg/kg b.wt. per 24 h)	5.2±0.8	2.4±0.3 [b]	2.4±0.3 [b]	2.7±0.4 [b]	3.7±0.6	1.3±0.2 [b]	1.6±0.2 [b]	1.9±0.2 [a]	10.3±2.2	13.6±4.9	12.7±2.8	13.7±2.3
GR138950 ($n=8$) (5 mg/kg b.wt. per 24 h)	5.6±0.6	2.0±0.4 [c]	1.9±0.1 [c]	2.8±0.3 [c]	4.4±0.6	1.2±0.3 [c]	1.2±0.2 [c]	2.0±0.2 [c]	9.2±1.8	10.5±2.0	6.6±2.5	14.3±2.5

Control represents the values obtained during the first '24 h' in the cage before vehicle or drug administration while '24 h', '48 h' and '72 h' represent the values obtained during the first, second and third '24 h' of drug treatment, respectively.

[a] $P < 0.05$.

[b] $P < 0.01$.

[c] $P < 0.001$, compared to the control values of the same group.

rats. Although there was a small but significant ($P < 0.05$) reduction in water intake on the first day of captoptil treatment, on the second and third days it was not significantly different from the control levels (Table 2). Urine flow fell slightly, but not consistently, on the first day of captopril administration, but thereafter was at levels close to the control values. Concomitantly, Na^+ excretion tended to rise over the initial two days of dosing and by day three was significantly ($P < 0.05$) higher by some 2–3 fold (Table 2).

3.3. Group III, losartan treatment

Administration of 15 mg/kg b.wt. losartan (group III_a) once daily caused an immediate fall in blood pressure ($P < 0.001$) on day one of some 64 mmHg and remained at this level for the remainder of the study which averaged some 67 ± 6 mmHg (Table 1). The higher dose of losartan, 30 mg/kg b.wt. (group III_b), reduced blood pressure by approximately 85 mmHg ($P < 0.001$) on the first day of treatment but then fell even further on days two and three such that the average reduction was 108 ± 10 mmHg (Table 1). In both losartan-treated groups of rats, urea concentration rose to levels much higher ($P < 0.001$) than those observed in the vehicle-treated group (Table 1). Creatinine concentrations also rose in both losartan-treated groups and were significantly higher ($P < 0.05$ in those received losartan at low doses, and $P < 0.01$ in rats receiving high dose losartan) than the vehicle-treated group (Table 1). Urine flow and water intake in both losartan groups were significantly ($P < 0.01$) reduced by day one of treatment and remained at this level over the whole of the treatment period (Table 2). Na^+ excretion was significantly ($P < 0.01$) elevated in the animals given the low dose of losartan on the second and third days of treatment whereas in the animals receiving the high dose of losartan Na^+ excretion did not change in a consistent way (Table 2).

3.4. Group IV, GR138950 treatment

In group IV, administration of 5 mg/kg b.wt. GR138950 daily significantly ($P < 0.001$) lowered blood pressure on the first treatment day by 118 mmHg and over the three day period gave an average fall of some 116 ± 14 mmHg (Table 1). The plasma concentration of urea was significantly higher in group IV (Table 1) compared to that observed in vehicle-treated rats and averaged 43.4 ± 7.8 mmol/l ($P < 0.01$). Creatinine levels were also elevated in group IV rats and averaged 196.0 ± 47.9 μmol/l which was significantly higher ($P < 0.05$) than the vehicle-treated rats (Table 1). Both water intake and urine flow were significantly ($P < 0.001$) reduced by GR138950 treatment after the first treatment day and remained depressed for the remainder of the study (Table 2). Na^+ excretion remained at a constant unchanged level for the whole of the treatment period (Table 2).

4. Discussion

This study set out to compare the effects of blockade of angiotensin AT_1 receptors, using non-peptide antagonists, with that of angiotensin converting enzyme inhibition on blood pressure, renal function, water intake and renal excretion of water and Na^+ in Na^+-deplete 2K2C hypertensive rats (Abdi and Johns, 1996). It is recognised that the renin–angiotensin system is highly activated in patients suffering from severe renal artery stenosis, those with malignant hypertension and patients with severe congestive heart failure (Antonios and MacGregor, 1995) and it is these individuals who are likely to be treated with the blockers of the renin–angiotensin system. Therefore, the Na^+-deplete 2K2C hypertensive rat is an appropriate model for investigating the action of drugs which are designated for renin–angiotensin system blockade in high-renin patients.

Surprisingly, in the captopril-treated group, the final plasma renin activity levels were somewhat lower compared to vehicle-treated rats. This finding was consistent with our earlier observation in which it was noted that there was a significantly lower plasma renin activity in captopril-treated 2K2C hypertensive rats fed a 'no' Na^+ diet but not in those which were fed with a low or normal Na^+ diet (Abdi and Johns, 1996). The degree of reduction in the plasma renin activity levels of captopril-treated rats appeared to be related to the magnitude of decrease in blood pressure and elevation of urea and creatinine plasma levels and it is possible to suggest that the renal renin was not able to enter the circulation as a consequence of the reduced perfusion through the kidney. In contrast to our findings, other investigators have reported increased plasma renin activity levels after renin–angiotensin system blockade with either angiotensin converting enzyme inhibition or angiotensin II receptor antagonism in Na^+-deplete renovascular hypertensive rats fed a low Na^+ diet (Helmchen et al., 1982; Garcia et al., 1996). Helmchen et al. (1982) reported 7- and 3-fold increases in the final plasma renin activity levels in Na^+ deplete 2K2C rats treated for 4 days with captopril and saralasin, respectively (Helmchen et al., 1982). Garcia et al. (1996) also reported a 3-fold increase in the final PRA of Na^+-deplete 1K1C hypertensive rats treated with losartan for 7 days (Garcia et al., 1996) but they did not report on kidney function. In our study, the blood pressure of the rats treated with the high dose of captopril was reduced to a level comparable to that obtained in sham-operated rats reported by ourselves (Abdi and Johns, 1996) and other investigators (Helmchen et al., 1982; Garcia et al., 1996). There was one difference in the experimental protocol of the present study compared with the two other reports in that a 'no' Na^+ rather than a low

Na^+ diet was used. However, in our previous study, the plasma renin activity of the hypertensive rats fed a low Na^+ diet was increased but not to the same extent as that achieved following the feeding of the 'no' Na^+ diet (Abdi and Johns, 1996). It is also possible that the different time intervals between drug administration and withdrawal of blood samples might also contribute to these different observations.

Administration of captopril caused a significant fall in blood pressure in the hypertensive rats suggesting that angiotensin converting enzyme inhibition could reduce blood pressure by removing the action of angiotensin II (Helmchen et al., 1982) or potentiating bradykinin-mediated effects (Gavras et al., 1992) on the cardiovascular system. Blockade of angiotensin II formation by angiotensin converting enzyme inhibition has been demonstrated to lower blood pressure in renovascular hypertensive subjects by acute vasodilation (Conway et al., 1979) and chronically to cause a reversal of cardiac (Dahlof, 1993) and vascular (Wang and Prewitt, 1990) hypertrophy. Administration of angiotensin converting enzyme inhibitors has been found to increase circulating bradykinin (Wong et al., 1990; Campbell et al., 1994) which has been suggested as being, at least partially, responsible for vasodilation (Gavras et al., 1992) and reduced cardiac hypertrophy (Dahlof, 1993) during angiotensin converting enzyme inhibition.

Losartan and GR138950 are two different angiotesin AT_1 receptor antagonists both of which caused a dose-dependent reduction in blood pressure in these hypertensive rats. The angiotensin AT_1 receptor antagonists have been shown to reduce blood pressure in normotensive rats (Inada et al., 1994) as well as various experimental models of hypertension such as the spontaneously hypertensive rat (Inada et al., 1994; Kett et al., 1996), 2K1C rats (Inada et al., 1994; Sigmon and Beierwaltes, 1993) and DOCA/salt hypertensive rats (Wang and Prewitt, 1990) at doses similar to those used herein. In normo-reninaemic Na^+-replete 2K2C rats, 10 mg/kg bwt losartan acutely lowered blood pressure from 156 ± 9 to 132 ± 5 mmHg (Basso et al., 1995) while very high doses of losartan some 77 mg/kg b.wt. was needed to lower blood pressure of Na^+-replete 1K1C hypertensive rats by 25 mmHg for 24 h (Inada et al., 1994) and one week administration of 20 mg/kg b.wt. per 24 h losartan failed to lower the blood pressure of Na^+-replete 1K1C-GHT rats (Garcia et al., 1996). However, chronic administration of losartan (20 mg/kg b.wt. per 24 h) normalised the blood pressure of Na^+-deplete 1K1C rats indicating that angiotensin II had become important in maintaining hypertension in 1K1C-Goldblatt rats when the animals were placed in a Na^+ retaining state. Lowering blood pressure to the same extent with either angiotensin converting enzyme inhibition or angiotensin II receptor antagonism suggests that removal of the actions of angiotensin II via angiotensin AT_1 receptors on the cardiovascular system is a major component in the antihypertensive actions of angiotensin converting enzyme inhibition (Inada et al., 1994), the contribution of the kinin–bradykinin system being slight.

A rise in the urea and creatinine in the plasma of patients is considered to be closely related to their renal clearance and reflects filtration and secretion of these nitrogenous products by the kidney (Schuster and Seldin, 1992; Walser et al., 1993). The retention of urea and creatinine in the plasma of Na^+-depleted 2K2C-hypertensive rats during either angiotensin converting enzyme inhibition or angiotensin AT_1 receptor antagonism was the most important finding of this study. Indeed severe reversible azotemia has been reported in captopril-treated patients with bilateral renal artery stenosis (Hirick et al., 1983; Chrysant et al., 1983; Textor et al., 1985) as well as Na^+ depleted 2K2C- and 1K1C-hypertensive rats (Helmchen et al., 1982; Demeilliers et al., 1995). Moreover, in earlier reports, it was shown that a reduction in the renal clearance of urea and creatinine was not caused by a simple fall in renal perfusion pressure because lowering blood pressure of 2K2C animals to the same extent by non-specific vasodilators such as hydralazine (Helmchen et al., 1982; Abdi and Johns, 1996) and minoxidil (Helmchen et al., 1982) did not cause renal failure. Using a bradykinin receptor antagonist, Kon et al. (1993) were able to increase glomerular filtration rate during angiotensin converting enzyme inhibition suggesting that deterioration of renal function was partly because of a selective efferent arteriolar dilation caused by bradykinin during angiotensin converting enzyme inhibition. Demeilliers et al. (1995) demonstrated that the plasma creatinine level at the end of a 6-day period of treatment with either enalapril (an angiotensin converting enzyme inhibitor) or losartan were higher than vehicle-treated 1K1C Na^+-deplete rats (Demeilliers et al., 1995). Interestingly, Demeilliers et al. (1995) noticed that glomerular filtration rate and renal vascular resistance were reduced to the same extent in both enalapril- and losartan-treated groups. The findings of the present study supports earlier reports of retention of creatinine (Helmchen et al., 1982; Demeilliers et al., 1995) and urea (Helmchen et al., 1982) during angiotensin converting enzyme inhibition and angiotensin II receptor blockade. However, Demeilliers et al. (1995) supported the bradykinin dependent hypothesis suggested by Kon et al. (1993) because renal plasma flow and filtration fraction in the losartan-treated group were significantly higher than enalapril-treated group (Demeilliers et al., 1995). Neither renal plasma flow nor filtration fraction were measured in this study and it is difficult to determine whether there was a dissociation between these variables during renin-angiotensin system blockade as noted by Demeilliers et al. (1995). Nonetheless, a combined increase in plasma level creatinine and urea supports our earlier suggestion that the withdrawal of angiotensin II vasoconstrictor action at both afferent and efferent arterioles would be the major cause of renal function deterioration (Abdi and Johns, 1996).

The very low basal levels of Na^+ excretion in 2K2C hypertensive rats fed the 'no' Na^+ diet demonstrates that homeostatic mechanisms for Na^+ preservation were probably highly activated. Angiotensin II is known to have a Na^+-retaining effect via its action at the proximal tubules (Cogan, 1990) and therefore blockade of its action, either by angiotensin converting enzyme inhibition, or peptide angiotensin II receptor antagonists, or non-peptide angiotensin AT_1 receptor antagonists, would be expected to cause a natriuresis. Interestingly, although there was an overall increase in Na^+ excretion during captopril administration, which was similar to that reported previously (Abdi and Johns, 1996), this did not occur with either of the angiotensin AT_1 receptor antagonists. The reason for this difference is unclear but may reside in the different mechanism of action of the two classes of compound and may perhaps reflect some renal effects of the potentiated actions of bradykinins. It is important to note that as blood pressure falls, Na^+ excretion is reduced, thus in the present study, the lower systemic blood pressure would have opposed the natriuretic effects of renin–angiotensin system blockade in the 2K2C hypertensive rats (Demeilliers et al., 1995). In support of this, Garcia et al. (1996) found no difference in the pattern of Na^+ excretion during administration of losartan into either Na^+-deplete and Na^+-replete groups (Garcia et al., 1996), a finding similar to those reported herein and previously (Abdi and Johns, 1996) during captopril administration in Na^+-replete and Na^+-deplete 2K2C hypertensive rats.

It was apparent that captopril did not alter the fluid balance of animals supporting our earlier findings that captopril had no significant effect on the fluid balance of Na^+-deplete hypertensive animals. In contrast, in the groups treated with losartan and GR138950, water intake and urine flow were reduced significantly and remained low until the end of study. Again, this may be related to the fact that animals had gone into renal failure, as evidenced by the elevated plasma urea and creatinine levels such that perfusion and filtration did not occur. In conclusion, these findings show that in a high renin model of hypertension, the 2K2C Goldblatt hypertensive rat, both losartan and GR138950 are effective antihypertensive agents. Equivalent hypotensive doses of the angiotensin converting enzyme inhibitor captopril and the angiotensin AT_1 receptor antagonists, losartan and GR 138950, are associated with marked elevations in plasma urea and creatinine levels and are indicative that renal failure had taken place. These findings highlight, at an experimental level, the importance of angiotensin II in the intrarenal regulation of glomerular haemodynamics, but at the clinical level, emphasise the fact that in man with renovascular hypertension and who may be volume depleted with a high plasma renin, angiotensin AT_1 receptor antagonists may have to be used with caution as is the case with angiotensin converting enzyme inhibitors.

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