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Angiotensin II-induced renal responses in anesthetized rabbits: effects of N^{ω}-nitro-L-arginine methyl ester and losartan [1]

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Abstract

Intrarenal arterial infusion of angiotensin II (4 ng/kg/min) reduced renal blood flow, glomerular filtration rate and urinary Na^+ excretion (UNaV) without affecting fractional Na^+ excretion (FENa) in anesthetized rabbits. Losartan (10 μg/kg/min) abolished these angiotensin II-induced renal responses. The renal blood flow, glomerular filtration rate and UNaV responses were potentiated during intrarenal arterial infusion of N^{ω}-nitro-L-arginine methyl ester (L-NAME, 10 μg/kg/min). A high dose of L-NAME (50 μg/kg/min) also potentiated the renal blood flow and UNaV responses but not the glomerular filtration rate response. Angiotensin II reduced FENa during L-NAME infusion at either dose. In L-NAME-pretreated rabbits, losartan abolished the angiotensin II-induced renal blood flow and glomerular filtration rate responses, but the reduction in FENa still remained. The present study suggests that in the rabbit kidney (1) nitric oxide attenuates the angiotensin II-induced (angiotensin AT1 receptor-mediated) vasoconstriction and (2) angiotensin II can evoke losartan-resistant tubular Na^+ reabsorption, but the tubular action is concealed by nitric oxide.

Keywords: Angiotensin II; Nitric oxide (NO); Losartan; L-NAME (N^{ω}-nitro-L-arginine methyl ester); Kidney; (Rabbit)

1. Introduction

Nitric oxide (NO) has been suggested to regulate peripheral vascular tone as a vasodilator and thereby participates in cardiovascular homeostasis (Manning et al., 1993). NO is also considered to modulate renal function. Since NO synthesis inhibitors such as N^{ω}-monomethyl-L-arginine (L-NA) and N^{ω}-nitro-L-arginine methyl ester (L-NAME) reduce renal blood flow, urine flow rate (UV), and urinary Na^+ excretion (UNaV) (Rees et al., 1990; Tolins et al., 1990; Naess et al., 1992), endogenous NO may contribute to the control of renal circulation and urine formation as a vasodilator and diuretic substance.

Angiotensin II is well known to regulate renal hemodynamics by its potent vasoconstrictor action. Angiotensin II is also suggested to act directly on the renal tubular system to enhance Na^+ reabsorption (Fagard et al., 1976). Angiotensin II receptors are classified into two main subtypes: angiotensin AT_1 and AT_2 receptors. AT_1 is the major subtype of angiotensin II receptor in the rabbit kidney (Herblin et al., 1991). An angiotensin AT_1 receptor antagonist losartan suppresses angiotensin II-induced vasoconstriction in the pump-perfused rat kidney (Fontoura et al., 1991) and angiotensin II-enhanced Na^+ reabsorption in the isolated rabbit convoluted proximal tubules (Eiam-Ong et al., 1993). Thus angiotensin II may control renal function through stimulation of vascular and tubular angiotensin AT_1 receptors.

Many studies have shown that endogenous NO exerts restrained influences upon angiotensin II-induced renal responses in the kidney (Ohishi et al., 1992; Ito et al., 1993; Takenaka et al., 1993; Alberola et al., 1994; Baylis et al., 1994; Evans et al., 1994; Hajj-Ali et al., 1994; Sigmon et al., 1994; Matsumura et al., 1995). However, the site and mechanism of their interaction are still controversial. Systemic inhibition of angiotensin-converting enzyme with captopril counteracts a L-NA-induced decrease in renal blood flow and increases the proximal tubular Na^+ reabsorption rate reduced by L-NA in conscious rabbits (Evans et al., 1994). In the dog kidney, however,

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L-NA is reported to enhance decreases in renal blood flow, glomerular filtration rate and UNaV but not a decrease in fractional excretion of Na^+ (FENa) induced by intrarenal arterial infusion of angiotensin II (Matsumura et al., 1995).

In the present study, we examined whether NO interacts with vasoconstriction and tubular Na^+ reabsorption induced by exogenous angiotensin II in the rabbit kidney in vivo. We also confirmed whether angiotensin AT_1 receptors mediate the renal effects of angiotensin II. Changes in renal hemodynamics and urine formation in response to intrarenal arterial infusion of angiotensin II were compared before and during infusion of losartan or L-NAME in anesthetized rabbits.

2. Materials and methods

2.1. Preparation

Male Japanese white rabbits (2.5–3.5 kg) were anesthetized with sodium pentobarbital (40 mg/kg) injected through a marginal ear vein. The trachea was cannulated and the rabbit was artificially ventilated with room air (stroke volume 50 ml, 25 rpm). A double rumen catheter was inserted into the right femoral vein for drug administration. Anesthesia was maintained by continuous infusion of pentobarbital (2–4 mg/kg/h, i.v.) throughout the experiments. Inulin, dissolved in plasma extender solution (Hespander, Kyorin, Tokyo, Japan), was given i.v. at a priming dose of 50 mg/kg and at a maintenance dose of 1 mg/kg/min (0.1 ml/kg/min). The right femoral artery was cannulated for collection of arterial blood samples and measurement of arterial blood pressure with a pressure transducer (model TP-200T, Nihon Kohden, Tokyo, Japan). The left kidney was exposed by a retroperitoneal flank incision, and the animal was suspended by clamping one of its lumbar spinous processes to facilitate manipulation. A catheter for urine collection was inserted into the ureter. The renal nerves were dissected away from the renal vessels and cut after ligation to exclude possible effects of angiotensin II and L-NAME on the neural control of renal function. An electromagnetic flow probe (1.5 mm in diameter, Nihon Kohden) was attached to the renal artery to measure renal blood flow with a square-wave flowmeter (model MF-27, Nihon Kohden). A curved 30-gauge needle connected to PE 10 tubing with two side branches (for single or combined intrarenal arterial infusion of drugs) was placed in the renal artery for drug infusion. The plasma extender solution was continuously infused via this catheter (0.1 ml/min) throughout the experiments. Blood pressure, heart rate and renal blood flow were recorded with a polygraph system (model PM-6000, Nihon Kohden). After completion of surgery, more than 90 min were allowed for stabilization with continuous monitoring of urine flow and hemodynamics.

2.2. Experimental protocols

Urine was collected over a 10-min period and 1 ml of arterial blood was withdrawn at the midpoint of urine collection to obtain basal values. Angiotensin II was then infused into the renal artery at 4 ng/kg/min for 15 min. Five minutes after the start of angiotensin II infusion, 10-min urine sampling and blood sampling were performed and the infusion was stopped. Ten minutes after the end of angiotensin II infusion, urine and blood samples for recovery values were collected. Then intrarenal arterial infusion of vehicle (0.9% saline, Group I, $n = 8$), L-NAME at 10 μg/kg/min (Group II, $n = 7$), L-NAME at 50 μg/kg/min (Group III, $n = 7$) or losartan at 10 μg/kg/min (group IV, $n = 6$) was started. Twenty minutes after the start of drug infusion, urine and blood sampling and angiotensin II infusion were performed again.

In Group V ($n = 6$), the urine and blood sampling and angiotensin II infusion (4 ng/kg/min for 15 min) were performed during simultaneous infusion of L-NAME (50 μg/kg/min) and losartan (10 μg/kg/min).

In Group VI ($n = 7$), urine and blood samples were obtained by the same manner as in Group III expect that acetylcholine (0.3 μg/kg/min), instead of angiotensin II, was infused into the renal artery before and during L-NAME infusion.

The experimental groups, the order of drug administration and the sampling periods were summarized in Fig. 1.

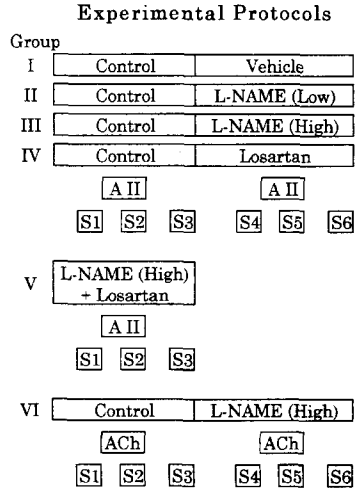

Fig. 1. Summary of the experimental protocols. A II, angiotensin II (4 ng/kg/min); ACh, acetylcholine (0.3 μg/kg/min); S1-S6, sampling periods (S1 and S4, basal; S2 and S5, ANG II or ACh; S3 and S6, recovery). In Groups I–IV, angiotensin II was infused before and during infusion of vehicle (0.9% saline), L-NAME (low, 10 μg/kg/min; high, 50 μg/kg/min) or losartan (10 μg/kg/min). In group V, angiotensin II was infused during simultaneous infusion of L-NAME and losartan. In Group VI, acetylcholine was infused before and during infusion of L-NAME. All drugs were infused into the renal artery. The experimental period before infusion of vehicle, L-NAME or losartan is expressed as 'Control'.

2.3. Measurements

Blood samples were transferred to ice-chilled tubes containing ammonium EDTA (6 mg/ml blood) and then centrifuged to obtain plasma samples. Glomerular filtration rate was determined as inulin clearance. Inulin concentration in plasma and urine was measured by the anthrone method. Na^+ and K^+ were measured by flame photometry (model 775A, Hitachi).

2.4. Statistics

All values were expressed as means ± S.E. Data for UV, UNaV and FENa were transformed to logarithms before application of statistical procedures. Effects of drugs (L-NAME and losartan) on the values before and during angiotensin II infusion and during the recovery period were analyzed by analysis of variance (ANOVA) for multifactor repeated measures. When ANOVA showed a statistical difference, the values in the control period and in the drug infusion period were compared in terms of simple main effects. Student's paired *t*-test was used to compare values before and during angiotensin II infusion and the percentage changes induced by angiotensin II in the control period and the drug infusion period. Differences at $P < 0.05$ were considered to be statistically significant.

3. Results

The values obtained before angiotensin II infusion, during angiotensin II infusion and after stopping angiotensin II infusion are expressed as 'Basal' , 'ANG II' and 'Recovery', respectively, in the table and figures.

In the control period (before infusion of vehicle, L-NAME or losartan), intrarenal arterial infusion of angiotensin II reduced renal blood flow and glomerular filtration rate and increased renal vascular resistance (Groups I–IV, Table 1 and Figs. 2 and 3). Filtration fraction tended to be elevated by angiotensin II infusion, but the change was not statistically significant (Table 1 and Figs. 2 and 3). Angiotensin II infusion reduced UV and UNaV whereas FENa remained unchanged. After the angiotensin II infusion was stopped, each value returned nearly to its basal level. Angiotensin II did not affect systemic blood pressure or heart rate in all experimental periods (Table 1).

Vehicle infusion did not affect renal blood flow, renal vascular resistance, glomerular filtration rate, filtration fraction, UV, UNaV or FENa (group I, Table 1). During the vehicle infusion period, angiotensin II caused similar renal responses as observed in the control period.

Intrarenal arterial infusion of L-NAME at the low dose (10 μg/kg/min, Group II) reduced renal blood flow,

Table 1
Systemic and renal hemodynamics and urinary parameters in Groups I, IV, V and VI

Group	Experimental period		Parameter								
			MAP (mm Hg)	HR (bpm)	RBF (ml/min)	RVR (U)	GFR (ml/min)	FF (%)	UV (ml/min)	UNaV (μEq/min)	FENa (%)
I	Control	Basal	95 ± 4	236 ± 16	25 ± 3	4.5 ± 0.5	4.6 ± 0.3	38 ± 4	0.43 ± 0.05	52 ± 5	8.5 ± 1.0
		ANG II	97 ± 4	234 ± 16	19 ± 2 [a]	5.8 ± 0.6 [a]	3.8 ± 0.4 [b]	40 ± 4	0.34 ± 0.05 [b]	41 ± 4 [b]	8.6 ± 1.3
		Recovery	98 ± 4	232 ± 16	23 ± 2	4.6 ± 0.4	4.4 ± 0.3	38 ± 5	0.45 ± 0.08	48 ± 6	8.7 ± 1.4
	Vehicle	Basal	94 ± 5	227 ± 16	23 ± 3	4.6 ± 0.4	4.5 ± 0.3	39 ± 2	0.45 ± 0.07	48 ± 6	8.0 ± 1.2
		ANG II	93 ± 4	224 ± 16	17 ± 2 [a]	6.6 ± 0.8 [a]	3.6 ± 0.4 [a]	42 ± 4	0.36 ± 0.06 [b]	40 ± 6 [b]	8.4 ± 1.3
		Recovery	93 ± 4	220 ± 16	20 ± 2	5.1 ± 0.4	4.3 ± 0.4	41 ± 4	0.44 ± 0.08	47 ± 8	8.1 ± 1.5
IV	Control	Basal	93 ± 4	229 ± 16	29 ± 3	3.3 ± 0.3	4.5 ± 0.5	28 ± 2	0.43 ± 0.06	39 ± 5	6.7 ± 0.9
		ANG II	92 ± 4	230 ± 16	24 ± 2 [a]	4.0 ± 0.3 [a]	4.1 ± 0.5 [b]	31 ± 3	0.35 ± 0.06 [a]	35 ± 5 [a]	6.5 ± 0.8
		Recovery	90 ± 3	236 ± 16	28 ± 3	3.4 ± 0.3	4.4 ± 0.3	29 ± 2	0.40 ± 0.06	40 ± 5	6.8 ± 0.9
	Losartan	Basal	81 ± 4 [c]	231 ± 18	34 ± 4 [c]	2.6 ± 0.2 [c]	4.2 ± 0.6	23 ± 3 [c]	0.24 ± 0.03 [c]	27 ± 5 [d]	4.8 ± 0.7 [d]
		ANG II	80 ± 4 [c]	229 ± 18	33 ± 4 [c]	2.6 ± 0.2 [c]	4.2 ± 0.5	23 ± 2 [c]	0.25 ± 0.04	27 ± 5	4.8 ± 0.7 [d]
		Recovery	78 ± 4 [c]	228 ± 18	35 ± 4 [c]	2.5 ± 0.2 [c]	4.0 ± 0.4	21 ± 1 [c]	0.25 ± 0.04 [c]	26 ± 6 [c]	4.8 ± 0.9 [d]
V	L-NAME	Basal	94 ± 4	195 ± 12	22 ± 0	4.4 ± 0.4	2.9 ± 0.2	23 ± 2	0.41 ± 0.07	33 ± 5	8.3 ± 1.3
	+	ANG II	93 ± 3	193 ± 14	21 ± 0	4.6 ± 0.4	3.1 ± 0.2	25 ± 2	0.36 ± 0.06	29 ± 5	6.8 ± 1.1 [b]
	losartan	Recovery	92 ± 4	190 ± 14	23 ± 0	4.2 ± 0.4	3.0 ± 0.3	23 ± 2	0.41 ± 0.07	34 ± 5	8.1 ± 1.3
VI	Control	Basal	100 ± 3	215 ± 6	24 ± 2	4.4 ± 0.4	2.5 ± 0.5	19 ± 3			
		ACh	99 ± 3	211 ± 8	30 ± 2 [a]	3.4 ± 0.3 [a]	2.1 ± 0.4	13 ± 2 [a]			
		Recovery	97 ± 4	206 ± 6	23 ± 2	4.3 ± 0.4	2.5 ± 0.6	19 ± 3			
	L-NAME	Basal	101 ± 3	186 ± 12 [c]	15 ± 1 [c]	7.0 ± 0.9 [c]	2.0 ± 0.4	24 ± 5 [c]			
		ACh	103 ± 2	185 ± 13 [c]	17 ± 1 [c]	6.4 ± 0.8 [c]	2.1 ± 0.4	22 ± 3 [c]			
		Recovery	103 ± 2 [d]	177 ± 14 [c]	15 ± 1 [c]	7.3 ± 0.8 [c]	2.0 ± .03	26 ± 4 [c]			

Values are means ± S.E.M. MAP, mean arterial pressure; HR, heart rate; RBF, renal blood flow; RVR, renal vascular resistance (U: mm Hg/ml/min); GFR, glomerular filtration rate; FF, filtration fraction; UV, urine flow rate; UNaV, urinary Na^+ excretion; FENa, fractional excretion of Na^+. Angiotensin II (ANG II; 4 ng/kg/min), acetylcholine (ACh 0.3 μg/kg/min), losartan (10 μg/kg/min) and L-NAME (50 μg/kg/min) were infused into the renal artery. [a] $P < 0.01$, [b] $P < 0.05$, compared with the corresponding basal value. [c] $P < 0.01$, [d] $P < 0.05$ compared with the corresponding control value.

glomerular filtration rate, UV, UNaV and FENa (Fig. 2). Similar changes were observed during infusion of L-NAME at the high dose (50 μg/kg/min, Group III, Fig. 3). In the L-NAME infusion period, angiotensin II caused similar changes in renal blood flow, glomerular filtration rate, renal vascular resistance, UV and UNaV as observed in the control period, but angiotensin II significantly reduced FENa (from 4.7 ± 0.9 to 3.9 ± 0.9% in Group II and from 6.6 ± 1.1 to 5.1 ± 1.4% in Group III, Figs. 2 and 3, respectively) and elevated filtration fraction (Group III, Fig. 3). Mean arterial pressure slightly rose during L-NAME infusion, but angiotensin II did not affect the arterial pressure in either experimental group. The values of mean arterial pressure (mm Hg) at basal, angiotensin II and recovery points were 100 ± 2, 100 ± 2 and 101 ± 2, respectively, in the control period and 104 ± 3 ($P < 0.05$ vs. recovery in the control period), 106 ± 2 and 108 ± 3, respectively, in the L-NAME (50 μg/kg/min) infusion period.

Fig. 4 shows the angiotensin II-induced responses (percent changes in the renal hemodynamic and urinary parameters from the basal levels) in the control and vehicle or L-NAME infusion period. Vehicle infusion (Group I) did not affect the angiotensin II-induced renal responses. L-NAME infusion at the low dose (Group II) potentiated the angiotensin II-induced decreases in renal blood flow, glomerular filtration rate, UV and UNaV and the increase in renal vascular resistance. L-NAME infusion at the high dose (Group III) also potentiated the renal blood flow, renal vascular resistance, UV and UNaV responses but not the glomerular filtration rate response; accordingly the elevation of filtration fraction was enhanced during L-NAME infusion.

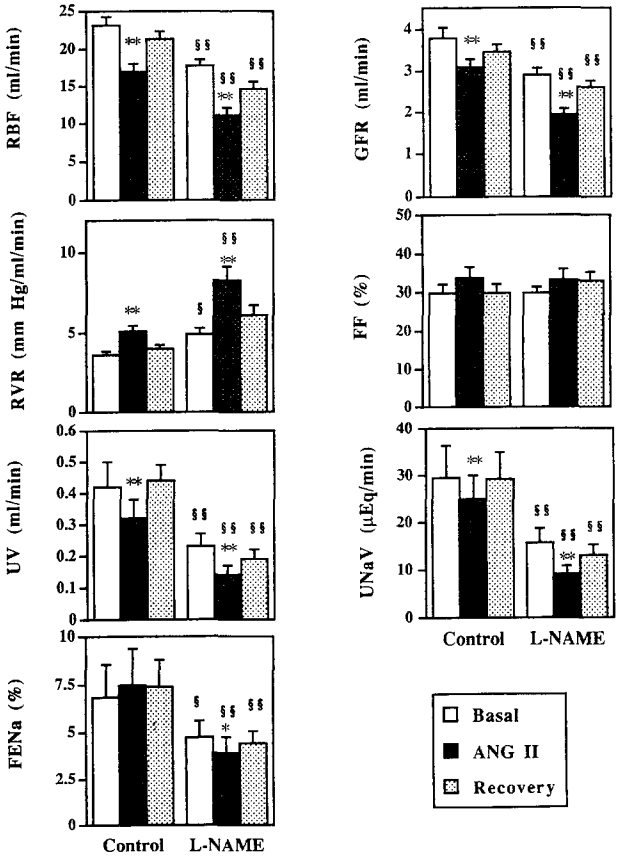

Fig. 2. Effects of angiotensin II on renal functions before and during infusion of L-NAME at 10 μg/kg/min (Group II). ANG II, angiotensin II (4 ng/kg/min); RBF, renal blood flow; GFR, glomerular filtration rate; RVR, renal vascular resistance; FF, filtration fraction; UV, urine flow rate; UNaV, urinary Na^+ excretion; FENa, fractional excretion of Na^+. Values are means ± S.E. $n = 7$. ** $P < 0.01$, * $P < 0.05$ compared with the corresponding basal value. §§ $P < 0.01$, § $P < 0.05$ compared with the corresponding value in the control period.

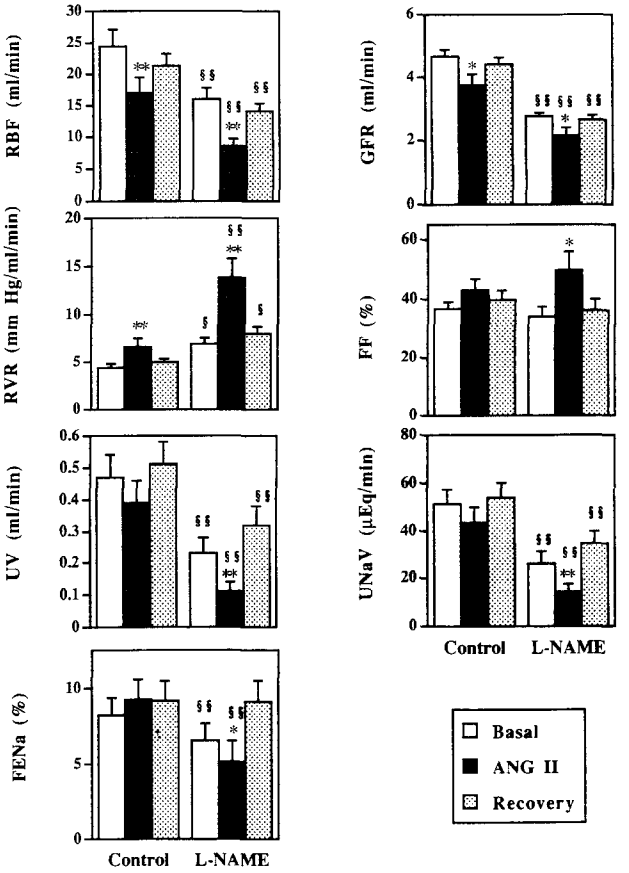

Fig. 3. Effects of angiotensin II on renal functions before and during infusion of L-NAME at 50 μg/kg/min (Group III). Abbreviations are as in Fig. 2. $n = 7$. ** $P < 0.01$, * $P < 0.05$ compared with the corresponding basal value. §§ $P < 0.01$, § $P < 0.05$ compared with the corresponding value in the control period.

Intrarenal arterial infusion of losartan increased renal blood flow and reduced mean arterial pressure, renal vascular resistance, filtration fraction, UV, UNaV and FENa without affecting glomerular filtration rate (Group IV, Table 1). In the losartan infusion period, angiotensin II did not affect the renal parameters (Table 1).

In the animal pretreated with the combination of L-NAME and losartan (Group V), angiotensin II infusion reduced FENa (the change from basal level was −21 ± 14%) without affecting renal blood flow, renal vascular resistance, glomerular filtration rate or filtration fraction

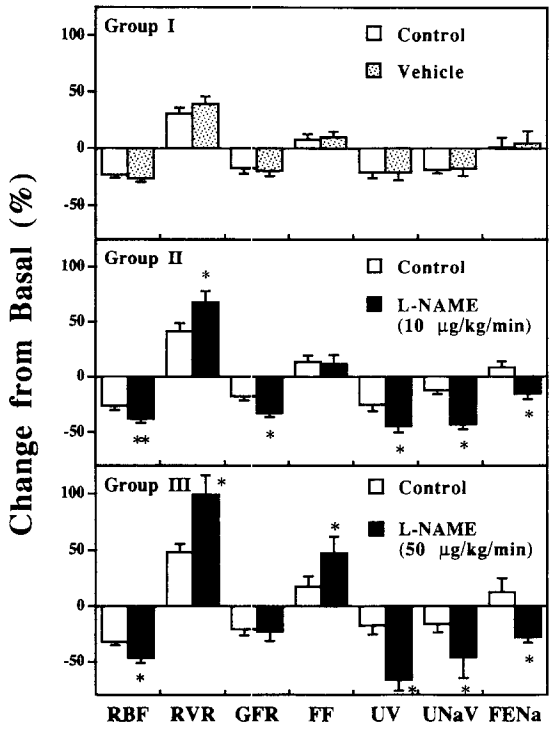

Fig. 4. Effects of vehicle and L-NAME on angiotensin II-induced changes in renal functions. Abbreviations are as in Fig. 2. Values are percent changes from basal values in response to angiotensin II infusion. ** $P < 0.01$, * $P < 0.05$ compared with the corresponding control value.

(Table 1). Angiotensin II infusion also reduced UV ($-16 \pm 12\%$) and UNaV ($-12 \pm 13\%$) although the changes were not statistically significant.

Intrarenal arterial infusion of acetylcholine increased renal blood flow and reduced filtration fraction (the changes from basal levels were $25 \pm 3\%$ and $-28 \pm 11\%$, respectively; Group VI, Table 1). L-NAME infusion (50 μg/kg/min) reduced renal blood flow and glomerular filtration rate, but the reduction in glomerular filtration rate was smaller than the change in renal blood flow and was not statistically significant. Thus L-NAME significantly elevated the filtration fraction in this experimental group. In the L-NAME infusion period, acetylcholine failed to cause statistically significant changes in renal blood flow ($13 \pm 6\%$) and filtration fraction ($1 \pm 14\%$). We also confirmed that intrarenal infusion of L-arginine (0.5 mg/kg/min) completely counteracted the changes in renal hemodynamics and urinary Na^+ excretion induced by the high dose of L-NAME (data not shown).

4. Discussion

In the present study, the effects of L-NAME on changes in renal hemodynamics and urine formation induced by angiotensin II were examined in anesthetized rabbits. Because both L-NAME and angiotensin II may enhance neurotransmitter release from renal sympathetic nerve endings (Matsuoka et al., 1991; Egi et al., 1994), all experiments were performed in surgically denervated kidneys to eliminate the influence of sympathetic tone on renal functions.

Intrarenal arterial infusion of angiotensin II elevated renal vascular resistance and reduced renal blood flow, glomerular filtration rate, UV and UNaV. Angiotensin II has been suggested to enhance Na^+ reabsorption at the proximal tubular site in micropuncture experiments with the rat kidney (Harris and Navar, 1985, Ichikawa and Brenner, 1980) and in Li^+ clearance experiments with the dog kidney in vivo (Olsen et al., 1985). In the present study, however, FENa remained unchanged during angiotensin II infusion. Angiotensin II therefore does not seem to evoke renal tubular Na^+ reabsorption in our experimental conditions. Angiotensin II may cause antinatriuresis through reduction of glomerular filtration rate in the rabbit kidney in vivo.

The angiotensin II-induced renal responses were completely suppressed during intrarenal arterial infusion of losartan. During vehicle infusion, angiotensin II induced the renal responses by the same degree as observed in the control period. Angiotensin II may cause vasoconstriction through stimulation of angiotensin AT_1 receptors, as commonly accepted (Fontoura et al., 1991; Loutzenhiser et al., 1991), in the rabbit kidney. Losartan reduced basal UV, UNaV and FENa, which may result from the reduction of systemic blood pressure. In our preliminary experiments, lower doses of losartan elevated UV, UNaV and FENa without affecting systemic hemodynamics but did not completely inhibit angiotensin II-induced renal responses.

Intrarenal arterial infusion of L-NAME (10 and 50 μg/kg/min) reduced renal blood flow and glomerular filtration rate. Thus endogenous NO may contribute to maintaining the renal circulation and filtration function. We confirmed that the acetylcholine-induced increase in renal blood flow was almost abolished during infusion of L-NAME, implying that L-NAME can effectively suppress NO production at the renal vessels of the rabbit. L-NAME also reduced UV, UNaV and FENa, which indicates that L-NAME promotes renal tubular Na^+ reabsorption. This is consistent with results obtained in other in vivo studies (Lahera et al., 1991; Alberola et al., 1992). Tracey et al. (1994) have identified NO synthase in LLC-PK_1 cells, implying that renal tubular cells can produce NO. Thus endogenous NO produced at the renal tubular site seems to act as a natriuretic factor.

During infusion of L-NAME, the angiotensin II-induced increase in renal vascular resistance and decreases in renal blood flow, UV and UNaV (evaluated by percent changes from the basal values) were greater than the responses observed in the control period. Matsumura et al. (1995) has shown that L-NA augments the renal hemodynamics and urinary responses induced by angiotensin II in dogs. Consistent with their results, our present study also suggests that renal NO plays a protective role in the kidney in

vivo by counteracting vasoconstriction and antinatriuresis in response to increased angiotensin II level.

Infusion of L-NAME at the low dose (10 μg/kg/min) enhanced the angiotensin II-induced decrease in glomerular filtration rate, which may contribute to the enhanced decreases in UV and UNaV. However, the high dose of L-NAME (50 μg/kg/min) also potentiated the angiotensin II-induced UV and UNaV responses without affecting the glomerular filtration rate response. Since angiotensin II reduced FENa during L-NAME infusion at each dose, angiotensin II may be able to evoke renal tubular Na^+ reabsorption in the NO-depleted state. Although in the dog kidney the enhanced antinatriuresis has been exclusively related to the augmented hypofiltration (Alberola et al., 1994; Matsumura et al., 1995), the renal tubular action of angiotensin II revealed by inhibition of NO production may participate in the enhanced UV and UNaV responses in the rabbit kidney. Evans et al. (1994) reported that captopril reduced and L-NA increased the rate of renal tubular reabsorption of Na^+ in conscious rabbits. Although combined administration of these two drugs nullified their responses, the authors concluded that the renal tubular actions of angiotensin II and NO may be independent. Our present study, however, demonstrates that there may be some kind of interaction between angiotensin II and NO at the renal tubular site, since angiotensin II itself did not reduce FENa in the absence of L-NAME.

We also examined whether the renal tubular action of angiotensin II is mediated by angiotensin AT_1 receptors. Because the direct tubular action of angiotensin II is concealed by NO, effects of losartan on the angiotensin II-induced responses were examined in animals pretreated with L-NAME. Losartan prevented the angiotensin II-induced renal blood flow, renal vascular resistance and glomerular filtration rate responses. However, angiotensin II reduced FENa and slight decreases in UV and UNaV still remained. Takenaka et al. (1993) reported that L-NA increased the endogenous angiotensin II level in renal tissue. Thus, this dose of losartan might not be sufficient to abolish the angiotensin II-induced renal tubular responses in the NO-depleted state. Kageyama et al. (1990) reported that nisordipine but not saralasin abolished the angiotensin II-induced decrease in FENa in anesthetized dogs. Keiser et al. (1992) reported that a selective angiotensin AT_2 receptor antagonist elicited natriuresis with no change in renal blood flow or systemic blood pressure. It is therefore also possible that the angiotensin II-induced renal tubular response is mediated by mechanisms other than losartan-sensitive receptors.

In summary, the present study suggests that in the rabbit kidney in vivo (1) angiotensin II contracts renal vasculature through stimulation of angiotensin AT_1 receptors, (2) angiotensin II can also enhance renal tubular Na^+ reabsorption, and (3) endogenous NO attenuates the vascular action and masks the tubular action of angiotensin II.

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