Pressure Natriuresis in Nitric Oxide-Deficient Hypertensive Rats: Effect of Antihypertensive Treatments

LOURDES A. FORTEPIANI,* ELENA RODRIGO,† M. CLARA ORTÍZ* VICTORIA CACHOFEIRO,† NOEMÍ M. ATUCHA* LUIS M. RUILOPE,† VICENTE LAHERA,† and JOAQUÍN GARCÍA-ESTAÑ*
*Departamentos de Fisiología, Facultades de Medicina, *Murcia and †Madrid, Spain.*

Abstract. Chronic inhibition of nitric oxide (NO) synthesis has been shown to result in arterial hypertension and an important blunting of the pressure diuresis and natriuresis response (PDN). The mechanisms mediating these abnormalities are not completely understood. In the present study, the role of several antihypertensive drugs to ameliorate these alterations was evaluated. The PDN relationships have been evaluated in rats chronically (8 wk) treated with the NO synthesis inhibitor N^G-nitro-L-arginine methyl ester (L-NAME; 40 mg/kg per d in the drinking water). Appropriate groups of rats were simultaneously treated with the angiotensin II receptor blocker candesartan at a low (1.5 mg/kg per d) and high (2.5 mg/kg per d) dose, with the converting enzyme inhibitor captopril (60 mg/kg per d) and with the calcium channel blocker verapamil (100 mg/kg per d). Chronic treatment with L-NAME significantly elevated mean BP (163.6 ± 6.5 mmHg *versus* 105.1 ± 3.6 in controls), reduced GFR and renal blood flow (RBF), and shifted to the right the PDN responses. Chronic administration of low-dose candesartan, captopril, or verapamil prevented the arterial hypertension and improved renal hemodynamics, but these levels were not completely normalized. High-dose administration also improved renal hemodynamics but induced reduced BP below the levels of control animals. Despite the normalization of the elevated BP, the PDN responses of these hypertensive treated groups were not normalized, and the slopes of the respective diuretic or natriuretic responses were very similar to those of the hypertensive untreated rats. The results indicate that interruption or blockade of the renin-angiotensin system and calcium channel blockade are effective treatments for the NO-deficient arterial hypertension and renal vasoconstriction. However, the PDN responses are not normalized, and this finding suggests that the antihypertensive treatment is not enough to overcome the renal alterations associated with the chronic deficiency of NO.

Nitric oxide (NO) is an important vasodilator and natriuretic substance that plays an important role in the control of renal hemodynamics and sodium excretion (1). Several investigators have shown clearly that acute and chronic inhibition of NO production induced by oral administration of NO synthase inhibitors such as N^G-monomethyl-L-arginine acetate or N^G-nitro-L-arginine methyl ester (L-NAME) produces arterial hypertension in animals (2–5). The mechanisms underlying this effect are not completely established, but there is considerable evidence suggesting that the renin-angiotensin system is a major participant in the renal and systemic alterations induced by the chronically reduced NO availability (6–11). This participation is supported by experiments showing that chronic AT_1 receptor blockade or converting enzyme inhibition prevents the development of L-NAME-induced hypertension (6).

Recently, NO has been shown to be an important modulator of the renal excretory response to changes in arterial pressure, the so-called pressure diuresis and natriuresis (PDN) mechanism. Separate studies have shown that either acute (12,13), short-term (14), or long-term (15) inhibition of NO synthesis profoundly reduces the PDN response, and this process may well be one of the mechanisms responsible for the arterial hypertension that is produced after NO synthesis inhibition. However, the mechanisms involved in the resetting of the PDN mechanism of the L-NAME hypertensive model are not known. Thus, in the present study we have evaluated the role of the renin-angiotensin system as mediator of the reduced renal excretory response to changes in arterial pressure in a model of chronic deficiency of NO. For this purpose, we have administered concomitantly with the NO synthesis inhibitor the AT_1 receptor blocker candesartan, at two different doses, the converting enzyme inhibitor captopril, and the calcium channel blocker verapamil.

Materials and Methods

Male Sprague Dawley rats (Charles River, Barcelona, Spain) were used for the study. All experiments were performed according to the guidelines for the ethical treatment of the animals of the European Union. Animals initially weighing approximately 150 g were maintained on standard rat chow and tap water throughout the study and were randomly divided into six groups that received the following treatments for 8 wk in the drinking water: (*1*) Control group ($n = 8$), which received no treatment; (*2*) L-NAME (40 mg/kg per d, $n = 7$); (*3*) L-NAME + low-dose candesartan (same dose of the NO synthesis inhibitor plus 1.5 mg/kg per d of the AT_1 blocker, $n = 9$); (*4*)

Received April 7, 1998. Accepted July 28, 1998.
Correspondence to Dr. Joaquín García-Estañ, Departamento de Fisiología, Facultad de Medicina, 30100 Murcia, Spain. Phone: 34-968-364880; Fax: 34-968-364150; E-mail: jgel@fcu.um.es
1046-6673/1001-0021$03.00/0
Journal of the American Society of Nephrology

L-NAME + high-dose candesartan (same dose of the NO synthesis inhibitor plus 2.5 mg/kg per d of the AT_1 blocker, $n = 9$); (*5*) L-NAME + captopril (same dose of the NO synthesis inhibitor plus 60 mg/kg per d of the converting enzyme inhibitor, $n = 7$); (*6*) L-NAME + verapamil (same dose of the NO synthesis inhibitor plus 100 mg/kg per d of the calcium channel blocker, $n = 6$). All of the treatments were adjusted throughout the study by measuring water intake and body weight. Both L-NAME and verapamil were purchased from Sigma (Madrid, Spain), and the other drugs were gifts as stated in the Acknowledgments.

Surgical Preparation

All experiments were performed as described previously (15–17), after a method originally described by Roman and Cowley (18), in rats fasted for 16 h before the experiment. The animals were anesthetized with inactin (100 mg/kg, intraperitoneally; Research Biomedical International, Natick, MA) and placed on a heated surgical table to maintain rectal temperature at approximately 37°C. The right femoral artery was cannulated, and basal mean arterial pressure (MAP) was measured before any other intervention was done (Hewlett-Packard 1280 transducer, Hewlett-Packard 8805D amplifier). Catheters were also inserted into the right femoral vein for infusions and into the right carotid artery for blood sampling and BP monitoring. A tracheostomy tube was placed to facilitate respiration. The left kidney was exposed by a midline abdominal incision and denervated by stripping the adventitia from both the renal artery and vein, and by applying 95% ethanol containing 10% phenol to each vessel to destroy any remaining nerve fibers. The left ureter was catheterized to collect urine. Silk ligatures were placed around the superior mesenteric and celiac arteries, and two adjustable clamps were placed on the aorta above and below the renal arteries to allow for increasing or decreasing renal perfusion pressure (RPP). A flow probe (0.8 mm in diameter) was placed around the left renal artery to measure renal blood flow (RBF) with a electromagnetic flowmeter (Skalar 1421, Skalar, The Netherlands). Finally, the abdominal opening was covered with a piece of Parafilm (American National Can, Greenwich, CT) to minimize evaporation. All animals received an intravenous infusion of 0.9% NaCl solution containing 1% bovine serum albumin at a rate of 2 ml/100 g per h. Plasma levels of sodium and water-retaining hormones were maintained at fixed high levels by adding aldosterone (20 ng/kg per min), corticosterone (10 ng/kg per min), vasopressin (0.05 ng/kg per min) and norepinephrine (100 ng/kg per min) to the infusion solution. [^{3}H]Inulin (1 μCi/ml, New England Nuclear, Itisa, Madrid, Spain) was included in the infusion solution to measure GFR. At least 45 min elapsed before the experiment was started.

Experimental Procedure

RPP measured either at the femoral or the carotid catheter, was continuously recorded throughout the experiment on a Hewlett-Packard model 7754A polygraph. After the stabilization period, RPP was lowered to approximately 100 mmHg by tightening the clamp above the renal arteries; after 15 min of stabilization, urine and plasma samples were collected. The aortic clamp was then released so that the kidney was perfused to approximately 125 mmHg, and after a period of stabilization, one more clearance period was recorded. Finally, the clamp below the renal arteries was occluded to elevate further RPP (to approximately 150 mmHg), and after a period of stabilization, a final clearance period was obtained. In some animals, it was necessary to momentarily occlude the celiac and mesenteric arteries to elevate RPP. In the hypertensive animals, the RPP levels to which the excretory function was measured were higher (120, 140, and 160) to not study the renal function below the different autoregulation range of these animals. Urine samples were collected in all periods into preweighed plastic vials, during 10 min. Blood samples (150 μl) for the determination of hematocrit and plasma inulin were obtained from the femoral or carotid catheter into heparinized microhematocrit tubes, in the middle of each clearance period. At the end of the experiment, the pressor and renal vasoconstrictor effect of angiotensin II (AngII) was tested by infusing a bolus dose of 50 ng. Then, the animal was euthanized by thoracotomy, and the left kidney was removed and weighed.

Analytical Techniques

[^{3}H]Inulin in plasma and urine was measured by counting aliquots of the samples dissolved in scintillation fluid (Ecoscint H, National Diagnostics, Atlanta, GA) in a β-counter (Wallac 1409, EG&G Instruments, Madrid, Spain). GFR was calculated as the clearance of radioactive inulin (urine to plasma concentration ratio $\times$ urine flow), and was normalized per gram kidney weight. Urine flow was determined gravimetrically. Sodium concentration was measured by flame photometry (Corning 435, Izasa, Barcelona, Spain).

Statistical Analyses

Data are presented as means $\pm$ SEM. A repeated-measures ANOVA was used to obtain the statistical significance between and within groups. If the global analysis was significant, a post hoc Duncan's test was carried out. The slopes of the relationships between RPP and the excretory parameters were calculated by linear regression analysis, and the differences between groups were obtained by unpaired *t* test. Differences were considered statistically significant at $P < 0.05$.

Results

The values of MAP in the experimental groups are shown in Figure 1. MAP was significantly elevated in the group treated chronically with L-NAME (163.6 $\pm$ 6.5 mmHg) compared with the values recorded in the control animals (105.1 $\pm$ 3.6). All of the treatments significantly reduced MAP in the L-

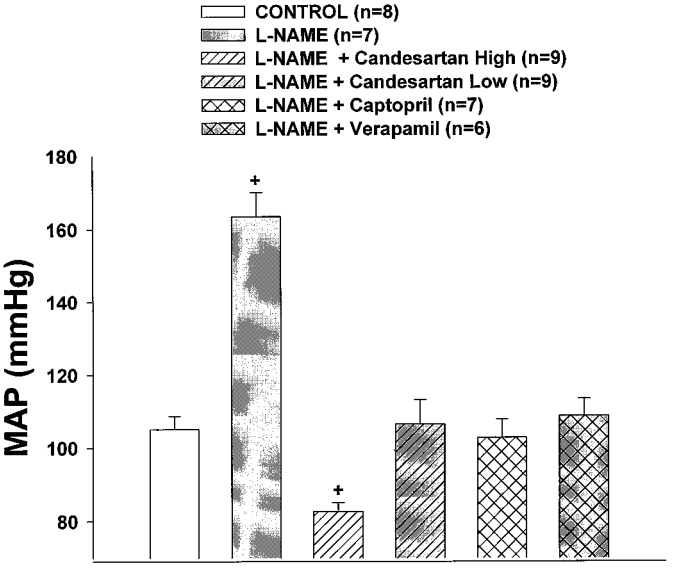

Figure 1. Mean arterial pressure in the experimental groups. *$P <$ 0.05 *versus* control.

NAME-treated groups. Thus, MAP in the animals simultaneously and chronically treated with L-NAME and the high dose of candesartan was 82.7 ± 2.5 mmHg, a value significantly lower than that of the controls. However, the group that received a lower dose of candesartan showed a MAP similar to that of the controls (108.8 ± 4.7). Chronic treatment with captopril or verapamil also completely prevented the elevation in MAP (106.6 ± 6.7 and 102.8 ± 5.1, respectively).

Figure 2 shows the PDN responses of the control animals and the animals that received L-NAME alone or in combination with two different doses of the AT_1 receptor blocker candesartan. As observed, the responses to changes in RPP were blunted in the L-NAME-treated groups in comparison with those of the control animals, so that the slopes of these relationships were very reduced (Table 1). The treatment with the high dose of candesartan did not significantly improve the PDN responses, and the slope of the PDN responses was even lower than in the L-NAME-treated group (Table 1). In contrast, treatment with a lower dose of the AT_1 blocker slightly improved the PDN responses, but without reaching the values observed in the control group (Table 1).

The renal hemodynamics of these groups are shown in Figure 3. Both GFR and RBF were significantly reduced in the L-NAME-treated animals, and treatment with both doses of candesartan normalized GFR and significantly elevated RBF, although it was not completely normalized. Hematocrit was 0.43 ± 0.003 in the lower RPP period of the control animals, 0.44 ± 0.01 in the L-NAME-treated group, 0.44 ± 0.007 in the group treated with the low dose of candesartan, and 0.44 ± 0.008 in the group treated with the high dose of candesartan. There were no significant changes in hematocrit in any group during the course of the experiments.

Figure 4 shows the PDN responses of the animals that received L-NAME and captopril or verapamil simultaneously, and for ease of comparison, also those of the control animals and the L-NAME-treated animals, already shown in Figure 2. As observed, captopril or verapamil slightly improved the responses, in a similar way to the group receiving the low dose of candesartan, but the slopes of the PDN responses were far from reaching the control values (Table 1). The renal hemodynamics of these groups are shown in Figure 5. Treatment with captopril or verapamil normalized GFR and significantly elevated RBF, although without reaching the values of the control group. Hematocrit was 0.45 ± 0.006 in the lower RPP period of the captopril-treated group and 0.46 ± 0.005 in the group treated with verapamil, and there were no significant changes during the course of the experiment.

Table 2 shows the determination coefficients of the linear regression analysis. As observed, control rats had the highest coefficients, and all of the L-NAME-treated rats had lower values. However, all of these coefficients were significant, thus, suggesting that the excretory responses were correlated with RPP.

The efficacy of the AngII blockade was assessed by the administration of a 50-ng bolus of AngII. In control animals, AngII elevated BP by 23.6 ± 3.0% and decreased RBF by 28.6 ± 3.2%. In the L-NAME-treated animals, AngII increased BP by 20.3 ± 0.8% and decreased RBF by 31.7 ± 5.9%. The simultaneous administration of candesartan almost abolished the pressor effect of AngII, increasing by 2.6 ± 0.4% in the

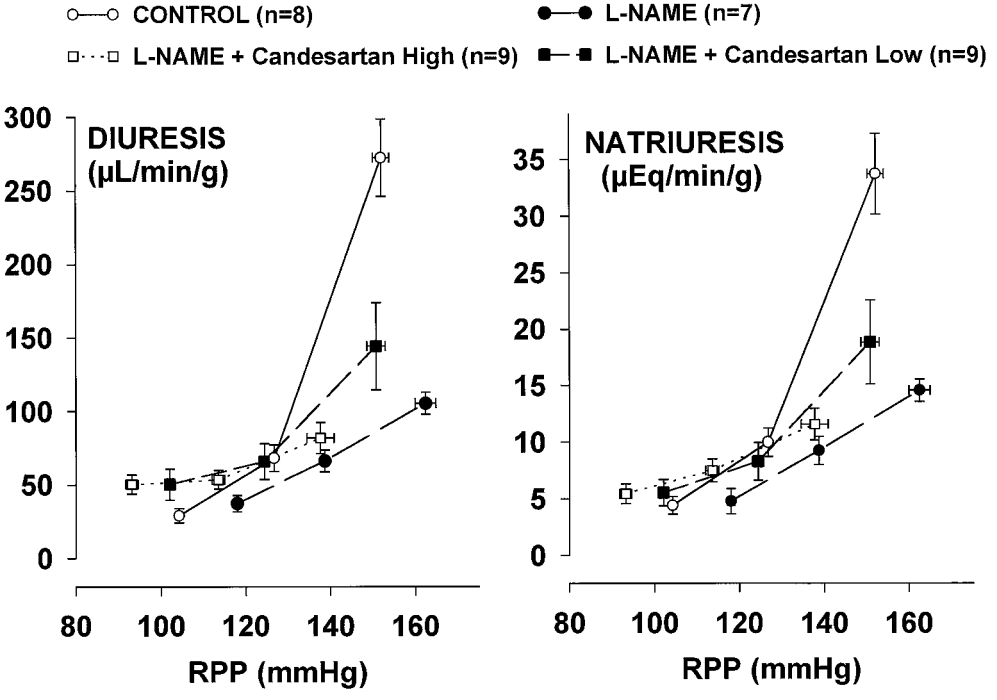

Figure 2. Changes in diuresis and natriuresis in response to changes in renal perfusion pressure (RPP) in control animals and in the groups chronically treated with the nitric oxide (NO) synthesis inhibitor N^G-nitro-L-arginine methyl ester (L-NAME) alone or in combination with two doses of the AT_1 receptor blocker candesartan.

Table 1. Slopes of the linear regression curves between renal perfusion pressure and the excretory parameters[a]

Group	Diuresis	Natriuresis	FE Water	FE Sodium
Control	5.14 ± 0.60	0.62 ± 0.07	0.28 ± 0.03	0.25 ± 0.03
L-NAME	1.43 ± 0.25[b]	0.19 ± 0.04[b]	0.15 ± 0.03[b]	0.13 ± 0.03[b]
L-NAME + low-dose candesartan	2.27 ± 0.64[b]	0.32 ± 0.08[b]	0.16 ± 0.04[b]	0.15 ± 0.04[b]
L-NAME + high-dose candesartan	0.84 ± 0.24[b]	0.15 ± 0.03[b]	0.05 ± 0.02[b,c]	0.06 ± 0.02[b,c]
L-NAME + captopril	2.23 ± 0.35[b]	0.28 ± 0.06[b]	0.18 ± 0.03[b]	0.16 ± 0.03[b]
L-NAME + verapamil	1.87 ± 0.71[b]	0.26 ± 0.09[b]	0.13 ± 0.04[b]	0.13 ± 0.03[b]

[a] Data are the mean ± SEM; FE, fractional excretion.
[b] $P < 0.05$ *versus* the control group.
[c] $P < 0.05$ *versus* the L-NAME-treated group.

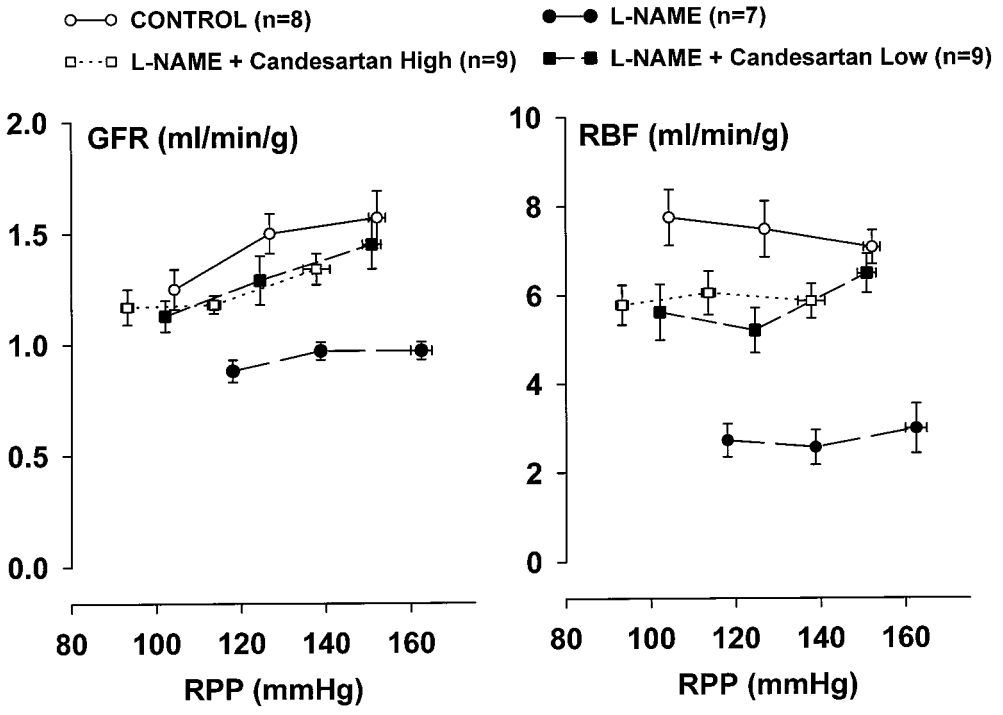

Figure 3. Changes in GFR and renal blood flow (RBF) in response to changes in RPP in control animals and in the groups chronically treated with the NO synthesis inhibitor L-NAME alone or in combination with two doses of the AT_1 receptor blocker candesartan.

high-dose group and 6.5 ± 1.2% in the low-dose group. Similarly, the renal vasoconstrictor effect of AngII was prevented by the AT_1 receptor blocker, decreasing by 5.1 ± 3.3% in the high-dose group and 5.2 ± 2.9% in the low-dose group. In the L-NAME + captopril group, AngII administration increased MAP by 25.2 ± 2.6% and decreased RBF by 30.5 ± 4.5%. In the L-NAME + verapamil group, AngII administration increased MAP by 26.5 ± 3.6% and decreased RBF by 28.7 ± 3.7%.

Discussion

The results of the present study confirm previous observations indicating that the renin-angiotensin system plays an important role contributing to the maintenance of the elevated BP levels observed when NO formation is chronically inhibited (6–11). This participation is clearly due to the action of AngII acting through AT_1 receptors, because both the converting enzyme inhibitor captopril and the AT_1 receptor blocker candesartan completely prevented the elevation in BP elicited by the administration of the NO synthesis inhibitor. Moreover, we have also found a dose-related effect on MAP, because the high dose of candesartan (2.5 mg/kg) not only prevented but induced a clear hypotension, thus stressing the importance of the AT_1 AngII receptors in this type of hypertension. Similarly, calcium channel blockade with verapamil also effectively prevented L-NAME hypertension, which agrees with other studies (19,20), showing the importance of calcium entrance as an underlying mechanism responsible for the increase in vascular tone.

Arterial hypertension is not the only sign resulting of the chronic inhibition of NO synthesis. Important renal alterations have also been described, among them renal vasoconstriction

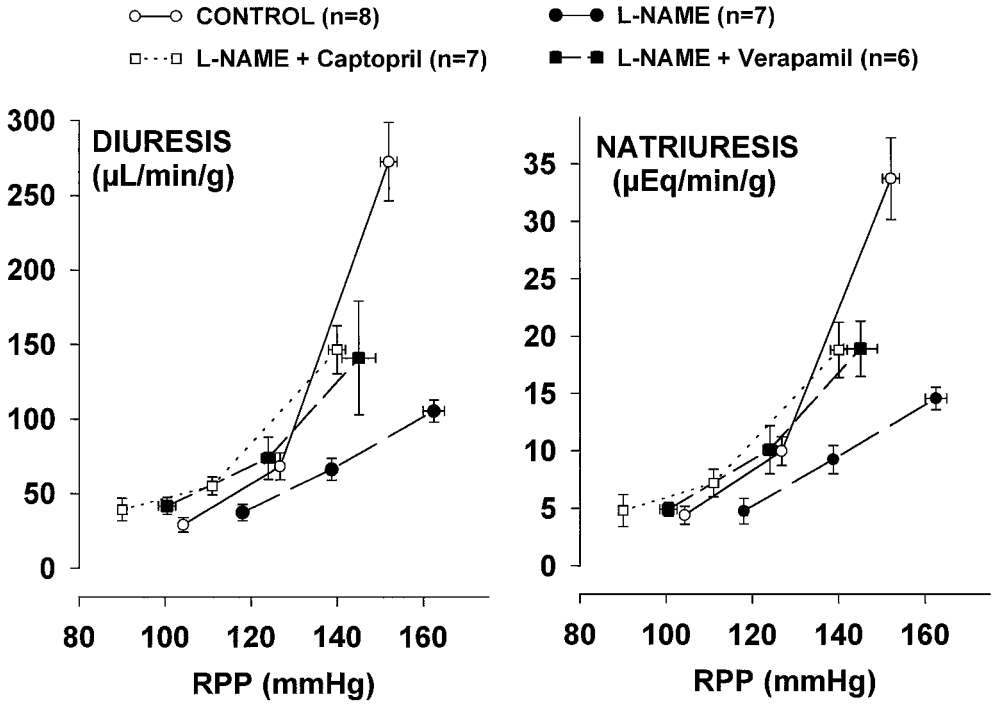

Figure 4. Changes in diuresis and natriuresis in response to changes in RPP in control animals and in the groups chronically treated with the NO synthesis inhibitor L-NAME alone or in combination with captopril or verapamil.

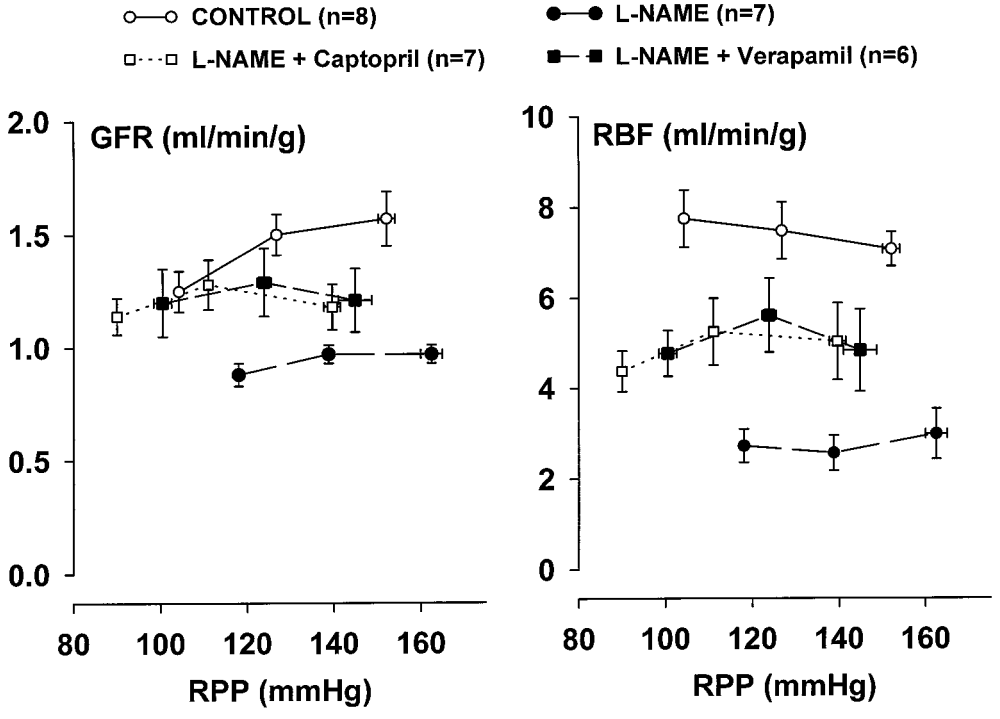

Figure 5. Changes in GFR and RBF in response to changes in RPP in control animals and in the groups chronically treated with the NO synthesis inhibitor L-NAME alone or in combination with captopril or verapamil.

and reduced GFR and a blunted excretory response to changes in RPP, the so-called PDN response (14,15). The PDN mechanism is thought to play a central role in the ability of the kidney to manage the long-term control of BP (21–23). Thus, the reduced PDN response shown by the animals chronically devoid of NO (15) may well be an important mechanism contributing to the elevation of MAP and the maintenance of the arterial hypertension of this model. Thus, the present results

Table 2. Determination coeficients (r^2) of the linear regression curves between renal perfusion pressure and the excretory parameters[a]

r^2	Diuresis	Natriuresis	FE Water	FE Sodium
Control (22)	0.77	0.77	0.80	0.78
L-NAME (19)	0.64	0.56	0.63	0.52
L-NAME + low-dose candesartan (25)	0.35	0.47	0.24	0.37
L-NAME + high-dose candesartan (25)	0.37	0.44	0.39	0.42
L-NAME + captopril (19)	0.68	0.57	0.68	0.55
L-NAME + verapamil (16)	0.30	0.31	0.43	0.48

[a] Numbers in parentheses are the degrees of freedom. All of the coeficients were significant at $P < 0.05$.

confirm previous data from our laboratory, which showed that chronic treatment with L-NAME blunts the PDN response (15). As observed previously (15), these results also indicate that this reduced response is due to both a decrease in GFR and to an increase in tubular reabsorption. Other investigators (14) have reported that chronic NO inhibition (15 d) merely displaces, in a parallel manner, the PDN relationships toward higher pressures. However, the results of this article and those previously published also in 8-wk treated rats (15) indicate that the PDN relationships are displaced to the right, but the slope of the relationship is clearly lower than that of the control animals.

In the present work, we have used several antihypertensive drugs to analyze their ability to ameliorate the renal abnormalities of L-NAME hypertension. In fact, all of the drugs tested completely prevented the elevation in MAP associated with the chronic deficiency of NO. However, none of these drugs were effective in normalizing the reduced PDN response of the L-NAME-treated animals. Thus, treatment with the lower dose of candesartan, captopril, or verapamil slightly but not significantly improved the slopes of the PDN relationships in comparison with those of the L-NAME-treated rats. This interesting result indicates a dissociation of the ability of these drugs to prevent the elevation in MAP and to normalize the PDN response. Thus, the blockade of AngII or the inhibition of its formation and the blockade of calcium entry can decrease the elevated vascular tone and thus prevent the arterial hypertension due to the absence of NO. However, it seems that the inhibition or blockade of AngII is not enough to improve the PDN response, and this finding may be due to several reasons. First, all of the AngII that cannot interact with the AT_1 receptor blocked by candesartan, could affect the tubular function through the activation of the AT_2 receptors. Although the exact role of renal AT_2 receptors is not completely clear yet (24,25), recent work indicates that the activation of AT_2 receptors blunts the PDN response (26,27). However, this reason is not valid to explain the response of the L-NAME + captopril-treated rats or that of the L-NAME + verapamil-treated group. Second, the lack of normalization of the excretory responses may also be related to the intrarenal deficit of NO, which is not reduced by the antihypertensive therapies. This is an unresolved issue, because there are conflicting reports about this topic. Thus, Tojo *et al.* (28) have published that angiotensin-converting enzyme inhibition with imidapril increased nitrite production and neuronal and endothelial type NOS immunoreactivities in the kidneys of L-NAME hypertensive rats. However, Takase *et al.* (19) found that verapamil or trandolapril did not increase constitutive NOS activity in the kidneys of L-NAME-hypertensive rats, despite a complete prevention of the arterial hypertension. Third, it is known that changes in arterial pressure elevates sodium and water excretion through the inhibition of tubular reabsorption in several nephron segments (23). These tubular effects may be induced by changes in renal interstitial pressure (16,23), intrarenal medullary hemodynamics (23), or both. Recent results from our laboratory (11) indicate that the chronic administration of the AT_1 receptor blocker losartan only moderately improves the reduced papillary blood flow shown by the L-NAME hypertensive rats, but blunts the arterial hypertension and reduced renal hemodynamics. Thus, the failure of these treatments, at least that of candesartan, to normalize the PDN response of the L-NAME hypertensive animals may be related to the fact that papillary blood flow is not normalized with the blockade of the AT_1 receptors. Clearly, more experiments will be necessary to determine the mechanisms behind the blunted PDN responses of the L-NAME hypertensive rats.

The inability of the different antihypertensive treatments to normalize the PDN response is also striking in view of the important beneficial impact that these drugs have on the severely reduced GFR and RBF of the NO-deficient rats. Thus, all of these drugs almost normalized renal hemodynamics, and therefore, they are able to restore a "normal" renal function. However, this "normalized" kidney seems to be still devoid of NO and some of its NO-dependent functions, such as pressure natriuresis are, therefore, not functional.

In summary, inhibition of AngII formation, blockade of AT_1 AngII receptors, and calcium channel blockade are effective treatments to completely prevent the arterial hypertension produced by the chronic inhibition of NO synthesis. These treatments are also very effective in ameliorating the reduced GFR and RBF of these NO-deficient animals. Despite all of these beneficial effects, however, the pressure natriuresis mechanism is not normalized, and this may be due to the chronic absence

of NO. Overall, these results suggest that NO is a very important long-term controller of sodium and water excretion.

Acknowledgments

This study was supported by grants SAF95-1549-C02-01 (to Dr. Lahera) and SAF95-1549-C02-02 (to Dr. García-Estañ) from the Comisión Interministerial de Ciencia y Tecnología de España. Candesartan was kindly provided by Takeda. Captopril was kindly provided by Squibb. Dr. Fortepiani (PN95-42849062) and Dr. Ortíz (FP94-29000129) are recipients of predoctoral fellowships from the Ministerio de Educación y Ciencia.

References

1. Romero JC, Lahera V, Salom MG, Biondi ML: Role of endothelium-dependent relaxing factor nitric oxide on renal function. *J Am Soc Nephrol* 2: 1371–1387, 1992
2. Arnal JF, Warin L, Michel JB: Determinants of aortic cyclic guanosine monophosphate in hypertension induced by chronic inhibition of nitric oxide synthase. *J Clin Invest* 90: 647–652, 1992
3. Ribeiro MO, Antunes E, de Nucci G, Lovisolo SM, Zatz R: Chronic inhibition of nitric oxide synthesis, a new model of arterial hypertension. *Hypertension* 20: 298–303, 1992
4. Navarro J, Sánchez A, Sáiz J, Ruilope LM, García-Estañ J, Romero JC, Moncada S, Lahera V: Hormonal, renal and metabolic alterations during the hypertension induced by chronic inhibition of nitric oxide in rats. *Am J Physiol* 267: R1516–R1521, 1994
5. Fernández-Rivas A, García-Estañ J, Vargas F: Effects of chronic increased salt intake on nitric oxide synthesis inhibition-induced hypertension. *J Hypertens* 13: 123–128, 1995
6. Pollock DM, Polakowski JS, Divish BJ, Opgenorth TJ: Angiotensin blockade reverses hypertension during long-term nitric oxide synthase inhibition. *Hypertension* 21: 660–666, 1993
7. Morton JJ, Beattie EC, Speirs A, Gulliver F: Persistent hypertension following inhibition of nitric oxide formation in the young Wistar rat: Role of renin and vascular hypertrophy. *J Hypertens* 11: 1083–1088, 1993
8. Qiu C, Engels K, Baylis C: Angiotensin II and α1-adrenergic tone in chronic nitric oxide blockade-induced hypertension. *Am J Physiol* 266: R1470–R1476, 1994
9. Bank N, Aynedjian HS, Khan GA: Mechanism of vasoconstriction induced by chronic inhibition of nitric oxide in rats. *Hypertension* 24: 322–328, 1994
10. Zanchi A, Schaad NC, Osterheld M-C, Grouzmann E, Nussberger J, Brunner HR, Waeber B: Effects of chronic NO synthase inhibition in rats on renin-angiotensin system and sympathetic nervous system. *Am J Physiol* 268: H2267–H2273, 1995
11. Ortíz MC, Fortepiani LA, Ruiz-Marcos FM, Atucha NM, García-Estañ J: Role of AT_1 receptors in the renal papillary effects of acute and chronic nitric oxide inhibition. *Am J Physiol* 274: R760–R766, 1998
12. Salom MG, Lahera V, Miranda-Guardiola F, Romero JC: Blockade of pressure natriuresis induced by inhibition of renal synthesis of nitric oxide in dogs. *Am J Physiol* 262: F718–F722, 1992
13. Majid DS, Williams A, Navar LG: Inhibition of nitric oxide synthesis attenuates pressure-induced natriuretic responses in anesthetized dogs. *Am J Physiol* 264: F79–F87, 1993
14. Guarasci GR, Kline RL: Pressure natriuresis following acute and chronic inhibition of nitric oxide synthase in rats. *Am J Physiol* 270: R469–R478, 1996
15. García-Estañ J, Navarro J, Atucha NM, Quesada T, Romero JC, Cachofeiro V, Lahera V: Chronic effect of nitric oxide and prostaglandin inhibition on pressure diuresis and natriuresis in rats. *Kidney Int* 49: S141–S143, 1996
16. García-Estañ J, Roman RJ: Role of renal interstitial hydrostatic pressure in the pressure diuresis response. *Am J Physiol* 256: F63–F70, 1989
17. García-Estañ J, Atucha NM, Quesada T, Vargas F: Involvement of renin-angiotensin system in the reduced pressure natriuresis response of hyperthyroid rats. *Am J Physiol* 268: E897–E901, 1995
18. Roman RJ, Cowley AW: Characterization of a new model for the study of pressure natriuresis in the rat. *Am J Physiol* 248: F190–F198, 1985
19. Takase H, Moreau P, Kung CF, Nava E, Luscher TF: Antihypertensive therapy prevents endothelial dysfunction in chronic nitric oxide deficiency: Effect of verapamil and trandolapril. *Hypertension* 27: 25–31, 1996
20. Navarro-Cid J, Maeso R, Rodrigo E, Muñoz-García R, Ruilope LM, Lahera V, Cachofeiro V: Renal and vascular consequences of the chronic nitric oxide synthase inhibition. Effects of antihypertensive drugs. *Am J Hypertens* 9: 1077–1083, 1996
21. Guyton AC, Coleman TG, Cowley AW: Arterial pressure regulation: Overriding dominance of the kidney in the long-term regulation and in hypertension. *Am J Med* 52: 584–594, 1972
22. Hall JE, Mizelle HL, Hildebrandt DA, Brands MW: Abnormal pressure natriuresis: A cause or a consequence of hypertension? *Hypertension* 15: 547–559, 1990
23. Cowley AW: Long-term control of arterial blood pressure. *Physiol Rev* 72: 231–300, 1992
24. Muñoz-Garcia R, Maeso R, Rodrigo E, Navarro J, Ruilope LM, Casal MC, Cachofeiro V, Lahera V: Acute renal excretory actions of losartan in spontaneously hypertensive rats: Role of AT2 receptors, prostaglandins, kinins and nitric oxide. *J Hypertens* 13: 1779–1784, 1995
25. Siragy HM, Carey RM: The subtype 2 (AT_2) angiotensin receptor mediates renal production of nitric oxide in conscious rats. *J Clin Invest* 100: 264–269, 1997
26. Lo M, Liu K-L, Lantelme P, Sassard J: Subtype 2 of angiotensin II receptors controls pressure-natriuresis in rats. *J Clin Invest* 95: 1394–1397, 1995
27. Madrid MI, Garcia-Salom M, Tornel J, De Gasparo M, Fenoy FJ: Effect of interactions between nitric oxide and angiotensin II on pressure diuresis and natriuresis. *Am J Physiol* 273: R1676–R1682, 1997
28. Tojo A, Kobayashi N, Kimura K, Hirata Y, Matsuoka H, Yagi S, Omata M: Effects of antihypertensive drugs on nitric oxide synthase activity in rat kidney. *Kidney Int* 55: S138–S140, 1996