

Diuretic and Enhanced Sodium Restriction Results in Improved Antiproteinuric Response to RAS Blocking Agents

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Angiotensin-converting enzyme inhibitors, angiotensin receptor blockers, and diuretics may exert synergistic antiproteinuric effects. Eighteen patients with a proteinuria >1 g/24 h after 6 mo of treatment with ramipril at 5 mg/d were assigned to receive in random order ramipril at 10 mg/d, valsartan at 160 mg/d, or combined ramipril at 5 mg/d and valsartan at 80 mg/d in addition to their antihypertensive treatment. The treatment periods lasted 4 wk and were separated by a 4-wk washout with ramipril at 5 mg/d. At the end of this crossover sequence, patients received combined ramipril at 5 mg/d, valsartan at 80 mg/d, and an increased furosemide dosage for an additional 4-wk period. The primary end point was the urinary protein/creatinine ratio for two 24-h urine collections at the end of each treatment period. No significant differences were noted between the study end points of the ramipril 10, valsartan 160, and combined ramipril 5 and valsartan 80 treatment groups. However, the urinary protein/creatinine ratio was lower with combined ramipril 5 and valsartan 80–increased furosemide dosage than with valsartan 160 and combined ramipril 5 and valsartan 80, with a similar tendency compared with ramipril 10. Combined ramipril 5 and valsartan 80–increased furosemide dosage decreased systolic home BP and increased serum creatinine but did not significantly increase the number of symptomatic hypotension cases compared with the other three treatments. Thus, in patients with severe proteinuria and hypertension, a cautious increase in diuretic dosage in addition to combined angiotensin-converting enzyme inhibitors and angiotensin receptor blockers decreases proteinuria and BP but may expose the patient to prerenal failure.

J Am Soc Nephrol 16: 474–481, 2005. doi: 10.1681/ASN.2004060505

Proteinuria is a major risk factor for progression to ESRD in both diabetic and nondiabetic nephropathies (1,2). Angiotensin II is a key player in the development of renal failure, either directly by promoting tissue fibrosis or indirectly through its action on glomerular hemodynamic and proteinuria (1,3–5). Therefore, inhibition of the renin-angiotensin system (RAS), through either angiotensin-converting enzyme inhibitors (ACEI) or angiotensin II receptor blockers (ARB), may have a positive impact on proteinuria and renal failure progression (2). ACEI significantly slow down renal failure progression in type 1 diabetes (6), as well as in nondiabetic nephropathies (7–12). ARB demonstrated a similar nephroprotective effect in type 2 diabetes (13,14). However, despite treatment with ACEI or ARB, many patients present residual proteinuria and progress to ESRD.

ACEI and ARB antagonize the RAS at different levels, suggesting that their combination may be beneficial (15). Several studies have shown that dual blockade of RAS by ACEI and ARB can decrease proteinuria more than ACEI and ARB alone in IgA nephropathy (16,17), type 1 (18) and 2 (19) diabetes, and

mixed primary nephropathies (20–24). This dual blockade of the RAS is also more effective at preventing renal failure progression than each form of monotherapy (25). However, only one study has shown that combined half doses of ACEI and ARB decrease proteinuria better than optimal doses of ACEI or ARB, demonstrating a true synergistic antiproteinuric activity (26). This latter study included only nondiabetic patients who had well-equilibrated BP and were taking fewer than two antihypertensive drugs and no RAS blocking agent before their inclusion in the study. However, most patients with severe proteinuria and renal failure do not reach the targeted BP in the absence of multiple drug regimens including a RAS blocking agent (7,8), particularly in the case of diabetes (13,14). Furthermore, the antiproteinuric effect of ACEI may be blunted by high salt intake and can subsequently be restored by diuretics (27). The aim of this study was to determine (1) whether the synergistic effect of ACEI and ARB on proteinuria is general to patients with renal failure of various causes, including diabetes, irrespective of their baseline BP, and (2) whether the antiproteinuric effect of combined ACEI and ARB can be increased by raising diuretic dosage.

Materials and Methods

Patients

The study population was selected from the outpatient clinic of the Nephrology-Clinical Immunology Department of Nantes University Hospital. The inclusion criteria were as follows: age between 18 and 80 yr, glomerulopathies not requiring or resistant to immunosuppressive

Received June 29, 2004. Accepted November 12, 2004.

Published online ahead of print. Publication date available at www.jasn.org.

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treatments, proteinuria >1 g/24 h after 6 mo of treatment with ramipril at 5 mg/d in addition to a conventional antihypertensive treatment, and changes in daily proteinuria $<50\%$ at three consecutive tests over a 2-mo period. Conventional antihypertensive treatments included calcium channel blockers, β -blockers, α -blockers, central acting drugs, and diuretics (furosemide 20 to 80 mg/d) whenever required for BP control. Diuretics were also prescribed to prevent pitting edema. The only patients excluded were those with serum creatinine levels >250 $\mu\text{mol/L}$ (2.82 mg/dl), a serum creatinine level increase $>20\%$ after introduction of ramipril at 5 mg/d, a contraindication or intolerance to ACEI or ARB, or an office systolic BP <110 mmHg. We sought to include both diabetics and nondiabetics, with either controlled or uncontrolled BP. Written informed consent was obtained from each patient before inclusion. The protocol was approved by the local ethics committee and conducted according to good clinical practice.

Study Design

This was a single-center, prospective, randomized, open-label crossover study. Patients were assigned to switch from a daily 5-mg dose of ramipril in random order to (1) 10 mg of ramipril, (2) 160 mg of valsartan, or (3) a combined half-dose of each treatment (*i.e.*, 5 mg of ramipril and 80 mg of valsartan). A Youden square design ensured that every treatment was represented in every period with the same frequency, to control for period effect (*i.e.*, a time-dependent trend that can affect the experiment as a whole, regardless of the treatment under evaluation). Three treatment sequences were defined (1, 2, 3; 2, 3, 1; and 3, 1, 2), with a treatment factor at three levels and a period factor at three levels. Treatment periods lasted 4 wk and were separated by a 4-wk washout with ramipril at 5 mg/d, to control for carryover effect (*i.e.*, the persistence of the effect of a treatment applied in one period on a subsequent period of treatment), because the acute hemodynamic effects of ACEI and ARB on proteinuria are fully reversible within 4 wk (28). It was believed to be unethical to perform a washout with no ramipril and no diuretics in these patients with severe hypertension and proteinuria.

Although patients were given diuretics (furosemide 20 to 80 mg/d) for uncontrolled hypertension or to prevent pitting edema, we put forward the hypothesis that nonclinically apparent overhydration might interfere with the antiproteinuric effect of the RAS-blocking agents. Therefore, immediately after this crossover sequence, patients entered a fourth treatment period with (4) combined ramipril at 5 mg/d, valsartan at 80 mg/d, and an increased furosemide dosage for an additional 4-wk period. Furosemide treatment at 40 mg/d was initiated in patients who did not receive diuretic therapy at the time of their inclusion in the study, and furosemide dosage was increased by 40 mg/d in the remaining patients. The furosemide dose could be increased by only 20 mg/d in patients with low systolic BP and high blood urea nitrogen over serum creatinine ratio, to avoid prerenal failure. Patients were advised not to change their usual protein and sodium intake throughout the crossover study but to avoid excessive salt intake during the last treatment period of diuretic dose reinforcement.

At the end of each treatment period (1-wk run-in, 4-wk test-treatment and washout periods), two 24-h urine samples were obtained for protein, creatinine, and sodium measurements, and blood was drawn to measure serum creatinine, sodium, potassium, and albumin. Home BP was measured and printed using a validated apparatus (OMERON 705CP) twice with a 2-min interval, in the morning as well as in the evening, 3 d/wk, during run-in and test-treatment periods. At the end of each treatment period, a physical examination that included measurement of body weight, heart rate, and sitting BP was performed.

Questions concerning symptomatic hypotension and side effects were also posed at this time.

Study Measures

The primary end point was the mean urinary protein/creatinine ratio in two consecutive 24-h collections of urine at the end of each treatment period. Secondary end points were mean 24-h proteinuria, home systolic and diastolic BP, and serum creatinine levels. Tolerance was evaluated by the number of home systolic BP <100 mmHg and the number of cases of symptomatic hypotension.

Sample Size

We hypothesized that patients would present a mean urinary protein/creatinine ratio of 3.5 ± 2 g/g with ramipril at 5 mg/d. Assuming a reduction of 0.7 ± 0.65 with ramipril at 10 mg/d and valsartan at 160 mg/d (from 3.5 to 2.8 g/g) and a reduction of 1.4 ± 0.65 with combined ramipril at 5 mg/d and valsartan at 80 mg/d (from 3.5 to 2.1 g/g), it was estimated that to give the study an 80% power to detect a statistically significant difference ($\alpha = 0.05$), 18 patients had to complete the crossover design sequence.

Statistical Analyses

An intention-to-treat statistical analysis was performed using the maximum bias method. During the crossover study, missing proteinuria, serum creatinine, or systolic or diastolic BP values were imputed using the worst value of their subgroup for the hypothesis (*i.e.*, highest proteinuria and serum creatinine and lowest BP). Missing 24-h creatininuria values were replaced by the mean value of all of the other creatininuria dosages of the corresponding patient. The absence of period and carryover effects on efficacy measures (*i.e.*, urinary protein/creatinine ratio and 24-h proteinuria) was confirmed. Linear models (mixed and fixed) were performed. Serum creatinine, systolic BP, and treatments were considered as fixed effects, and patients were considered as a random effect. Tukey tests were used to localize the difference. The primary comparison was made between combined ramipril 5-valsartan 80, ramipril 10, and valsartan 160. A secondary comparison was made between combined ramipril 5, valsartan 80, and increased furosemide dose and the three other treatments and between each of these treatments and the baseline treatment with ramipril 5.

A subgroup analysis was made in individuals with and without diabetes, because patients with type 2 diabetes and overt nephropathy may develop abnormalities in size-selective function of the glomerular barrier that are not improved by low dose of ACEI (29). Other variables, including home BP and serum creatinine levels while on combined ramipril 5, valsartan 80, and increased furosemide dose, were compared with each of the other three treatments.

Tolerance was evaluated using a multiple Kruskal-Wallis test. Qualitative criteria were compared using Fisher exact test. The level of statistical significance was set to 5%, in bilateral situation. The SPLUS 6.2 Professional software was used.

Results

Patients

The clinical characteristics of the 18 enrolled patients are listed in Table 1. The patients included 12 men and six women, all of whom were white and had a mean age of 49.3 ± 20.4 yr, a mean 24-h proteinuria with ramipril at 5 mg/d of 3.71 ± 2.10 g/d, and a mean serum creatinine level of 151.2 ± 63.9 $\mu\text{mol/L}$ (1.71 mg/dl). Seven patients had diabetes, four had IgA nephropathies, four had focal segmental glomerulosclerosis, one

Table 1. Patient characteristics at baseline

Gender ratio M:F	12:6
Age (mean \pm SD)	49.3 \pm 20.4 yr
24-h proteinuria (mean \pm SD)	3.71 \pm 2.1 g/d
Serum creatinine (mean \pm SD)	151.22 \pm 63.9 μ mol/L
Systolic home BP (mean \pm SD)	149.06 \pm 29.1 mmHg
Diuretic	9/18 patients
mean furosemide dose	21.1 mg/d (range 0–80)
no. of antihypertensive drugs	2.6 (range 1–6)
Diagnosis	
diabetic nephropathy	7
IgA nephropathy	4
focal segmental glomerulosclerosis	4
minimal-change disease	1
amyloidosis	1
mesangioproliferative glomerulonephritis	1

had minimal-change disease, one had mesangioproliferative glomerulonephritis, and one had amyloidosis. The mean number of antihypertensive drugs was 2.6, including diuretics in nine of 18 patients. Home systolic BP was 149.1 \pm 29.1 mmHg and was >135 mmHg in 13 of 18 patients. The furosemide dose was increased from 21.1 mg/d (range, 0 to 80) to 47.8 mg/d (range, 20 to 120) during the last treatment period.

Efficacy Measurements

No period ($P = 0.51$) or carryover effects ($P = 0.10$) were observed during the crossover study. Changes in serum creatinine ($P = 0.0002$) and systolic ($P = 0.042$) but not diastolic ($P = 0.63$) home BP had a significant effect on the urinary protein/creatinine ratio. Therefore, serum creatinine and systolic home BP were used as fixed effects in the mixed model that analyzed the impact of study treatments.

In the mixed model, the urinary protein/creatinine ratio was not significantly different between combined ramipril 5 and valsartan 80 (3.01 \pm 2.68 g/g), ramipril 10 (2.98 \pm 2.02 g/g), and valsartan 160 (3.20 \pm 2.32 g/g) ($P = 0.39$ with serum creatinine and systolic BP as fixed effects and $P = 0.48$ without). The urinary protein/creatinine ratio did not significantly differ between any of these three treatments and the baseline treatment with ramipril 5 (3.71 \pm 3.00 g/g) ($P = 0.17$ for ramipril 10, $P = 0.70$ for valsartan 160, and $P = 0.66$ for combined ramipril 5 and valsartan 80; Figure 1). However, the mixed and fixed model showed that the urinary protein/creatinine ratio significantly differed between combined ramipril 5, valsartan 80, and increased furosemide dose (2.12 \pm 1.76 g/g) and the other three treatments ($P = 0.029$), with borderline significance without serum creatinine and systolic BP as fixed effects ($P = 0.066$). The urinary protein/creatinine ratio was indeed significantly lower (Tukey test) with combined ramipril 5, valsartan 80, and increased furosemide dose than with ramipril 5, valsartan 160, and combined ramipril 5 and valsartan 80. Although the decrease in urinary protein/creatinine ratio with combined ramipril 5, valsartan 80, and increased furosemide dosage

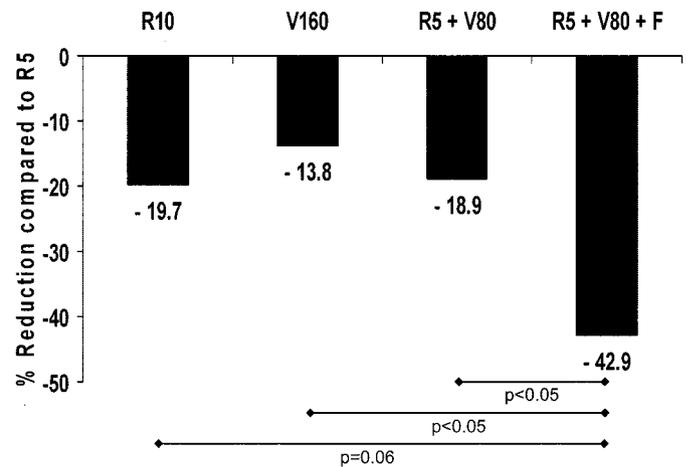


Figure 1. Urinary protein/creatinine ratio: Changes (in percentage) compared with ramipril at 5 mg/d (R5) after ramipril at 10 mg/d (R10); valsartan at 160 mg/d (V160); combined ramipril 5 and valsartan 80 (R5 + V80); and combined ramipril 5, valsartan 80, and increased furosemide dosage (R5 + V80 + F).

did not reach statistical significance compared with ramipril 10, a similar tendency was observed ($P = 0.060$; Figure 1).

In the mixed model, the 24-h proteinuria level did not significantly differ between combined ramipril 5 and valsartan 80 (3.01 \pm 2.07 g/d), ramipril 10 (3.60 \pm 2.90 g/d), and valsartan 160 mg/d (3.02 \pm 1.51 g/d) ($P = 0.63$ with serum creatinine and systolic BP as fixed effects and $P = 0.70$ without). The proteinuria level did not significantly differ between any of these three treatments and the baseline treatment with ramipril 5 (3.71 \pm 2.10 g/d) ($P = 0.80$ for ramipril 10, $P = 0.47$ for valsartan 160, and $P = 0.78$ for combined ramipril 5 and valsartan 80; Figure 2). The mixed and fixed model with Tukey tests showed that the proteinuria level was lower with combined ramipril 5, valsartan 80, and the increased furosemide dose (2.06 \pm 1.53 g/d) compared with ramipril 5, ramipril 10, valsartan 160, and

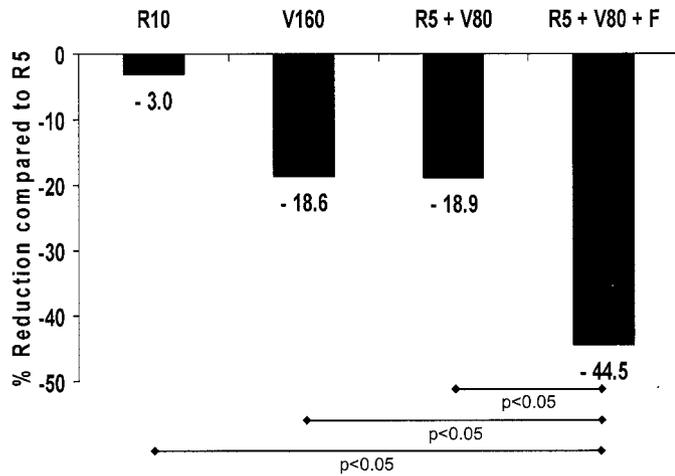


Figure 2. Proteinuria levels: Changes (in percentage) compared with R5 after R10, V160, R5 + V80, and R5 + V80 + F.

combined ramipril 5 and valsartan 80 ($P = 0.010$), with borderline significance without serum creatinine and systolic BP as fixed effects ($P = 0.080$; Figure 2).

There was no significant difference between ramipril 10, valsartan 160, and combined ramipril 5 and valsartan 80 in serum creatinine levels ($165.4, 163.1, \text{ and } 162.7 \mu\text{mol/L}$; $9.4, 7.9, \text{ and } 7.6\%$ versus baseline ramipril 5, respectively), systolic home BP ($142.4, 148.9, \text{ and } 144.1 \text{ mmHg}$; $-4.5, -0.1, \text{ and } -3.3\%$ versus baseline ramipril 5), and diastolic home BP ($78.9, 81.2, \text{ and } 77.7 \text{ mmHg}$; $-4.9, -2.1, \text{ and } -6.4\%$ versus baseline ramipril 5). However, systolic home BP was significantly decreased by combined ramipril 5, valsartan 80, and the increased furosemide dose ($135.2 \pm 23.5 \text{ mmHg}$) compared with valsartan 160 ($P = 0.003$) and combined ramipril 5 and valsartan 80 ($P = 0.025$) but not compared with ramipril 10 ($P = 0.15$; Figure 3). Similarly, diastolic home BP was decreased by combined ramipril 5, valsartan 80, and the increased furosemide dose ($73.3 \pm 10.5 \text{ mmHg}$) compared with valsartan 160 ($P = 0.003$)

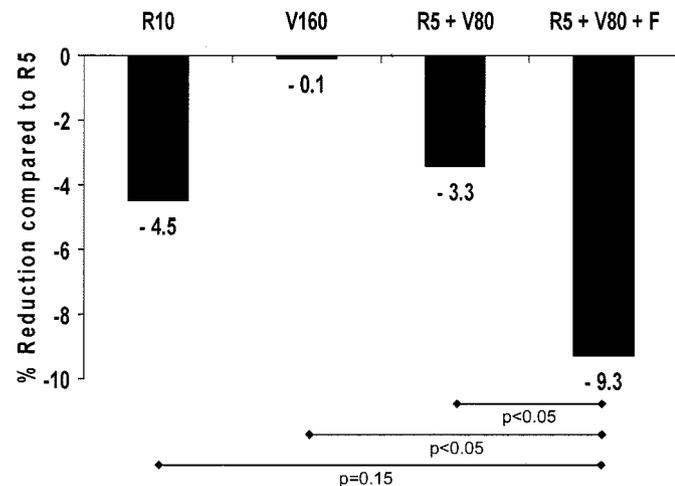


Figure 3. Systolic home BP: Changes (in percentage) compared with R5 after R10, V160, R5 + V80, and R5 + V80 + F.

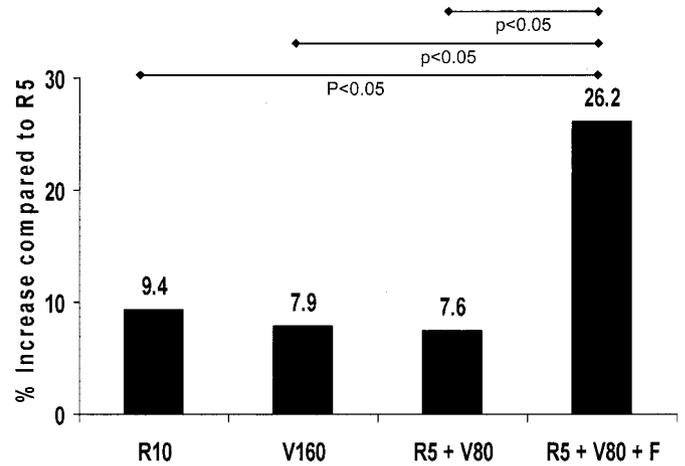


Figure 4. Serum creatinine levels: Changes (in percentage) compared with R5 after R10, V160, R5 + V80, and R5 + V80 + F.

and combined ramipril 5 and valsartan 80 ($P = 0.019$) but not compared with ramipril 10 ($P = 0.08$). Furthermore, serum creatinine levels were increased by combined ramipril 5, valsartan 80, and the increased furosemide dose ($190.8 \pm 71.7 \mu\text{mol/L}$) compared with ramipril 10 (15% ; $P = 0.025$), valsartan 160 (17% ; $P = 0.001$), and combined ramipril 5 and valsartan 80 (17% ; $P = 0.030$; Figure 4).

Subgroup Analysis in Individuals with and without Diabetes

The urinary protein/creatinine ratio was greater in individuals with diabetes than in those without diabetes at baseline with ramipril 5 ($P = 0.033$). In both individuals with and without diabetes, the decrease in the urinary protein/creatinine ratio from baseline was not significantly different between ramipril 10, valsartan 160, and combined ramipril 5 and valsartan 80. However, there was a trend for high doses of ACEI or ARB to decrease the urinary protein/creatinine ratio more in individuals with diabetes than in those without diabetes ($P = 0.08$; Figure 5).

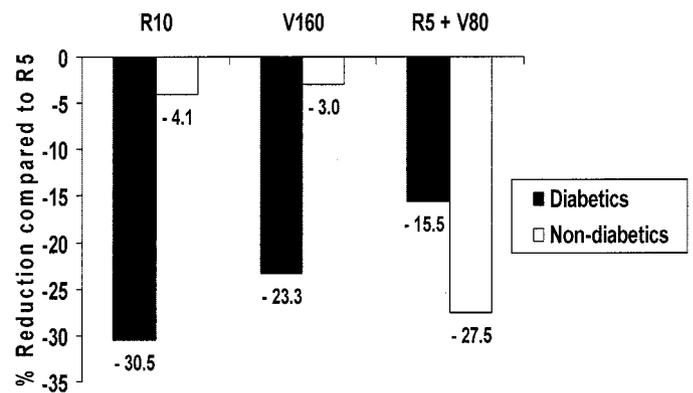


Figure 5. Urinary protein/creatinine ratio in individuals with diabetes (■) and individuals without diabetes (□): Changes (in percentage) compared with R5 after R10, V160, and R5 + V80 (Tukey test).

Table 2. Clinical characteristics at the end of each treatment period^a

	R5	R10	V160	R5 + V80	R5 + V80 + F
Home systolic BP	149.06 ± 29.1 ^b	142.44 ± 30.0 (<i>P</i> = 0.15)	148.88 ± 26.5 ^b	144.12 ± 29.7 ^b	135.23 ± 23.5
Home diastolic BP	83.00 ± 13.0 ^b	78.94 ± 14.4 (<i>P</i> = 0.08)	81.25 ± 13.4 ^b	77.65 ± 13.5 ^b	73.31 ± 10.5
24-h proteinuria (g/24 h)	3.71 ± 2.1 ^b	3.60 ± 2.9 ^b	3.02 ± 1.5 ^b	3.01 ± 2.1 ^b	2.06 ± 1.5
Urinary protein/creatinine (g/g)	3.71 ± 3.0 ^b	2.98 ± 2.0 (<i>P</i> = 0.06)	3.20 ± 2.3 ^b	3.01 ± 2.7 ^b	2.12 ± 1.8
Serum creatinine	151.22 ± 63.9 ^b	165.44 ± 73.0 ^b	163.13 ± 65.0 ^b	162.65 ± 79.7 ^b	190.79 ± 71.7
Serum potassium	4.07 ± 0.48	4.21 ± 0.56	4.32 ± 0.59	4.36 ± 0.73	4.53 ± 0.82
Serum albumin	35.5 ± 5.5	35.8 ± 4.6	34.9 ± 6.7	37.7 ± 5.6	35.9 ± 5.3
No. of home systolic BP <100	0 (1 wk)	1.63 ± 3.9 (4 wk)	0.81 ± 2.6 (4 wk)	1.82 ± 4.0 (4 wk)	3.77 ± 6.7 (4 wk)
Symptomatic hypotension	0 (1 wk)	0.35 ± 1.5 (4 wk)	0.38 ± 1.5 (4 wk)	0.59 ± 1.7 (4 wk)	0.29 ± 1.1 (4 wk)
24-h urinary sodium	165.7 ± 62.3	167.7 ± 90.7	147.4 ± 62.2	155.5 ± 57.4	128.9 ± 58.5

^aR5, ramipril at 5 mg/d; R10, ramipril at 10 mg/d; V160, valsartan at 160 mg/d; R5 + V80, combined ramipril at 5 mg and valsartan at 80 mg/d; R5 + V80 + F, combined ramipril at 5 mg, valsartan at 80 mg/d, and increased furosemide dosage. Data are mean ± SD.

^bComparisons *versus* R5 + V80 + F: *P* < 0.05. Statistical trends are illustrated by *P* values.

Adverse Events

No significant difference in the number of symptomatic hypotension cases was noted between treatment groups (Table 2). There was also no notable clinically significant difference in serum potassium levels between treatments with an increase of <0.5 mmol/L between the ramipril 5 group and the combined ramipril 5, valsartan 80, and increased furosemide dosage group (Table 2). Although the number of home SBP values <100 mmHg was numerically higher with combined ACEI and ARB, there was no difference in the number of individuals with symptomatic hypotension (Table 2).

The only missing data concerned the third treatment period for patient 9 (combined ramipril 5 and valsartan 80) and patient 12 (valsartan 160) and the last treatment period with combined ramipril 5, valsartan 80, and the increased furosemide dosage for both patient 9 and 12, because these two patients dropped out before the last treatment period of the crossover study. One experienced laryngeal edema in the context of increased ACEI dosage and viral infection, and the other started a pregnancy. These patients were included in the intention-to-treat analysis using the maximum bias method. This imputation method altered the statistical difference in urinary protein/creatinine ratio between the combined ramipril 5, valsartan 80, and increased furosemide dosage group and the ramipril 10 group, with statistical significance becoming only borderline (*P* = 0.06). Other unrelated adverse events included a traumatic radial fracture and hospitalizations for depression and cataract surgery.

Discussion

Our study shows that the synergistic effect of combined half doses of ACEI and ARB was not confirmed in patients with persisting severe proteinuria under ACEI treatment, because combined half doses of ACEI and ARB did not decrease proteinuria more than either given as a full-dose monotherapy. Several studies have shown that adding ARB without reducing the dose of ACEI decreases proteinuria, demonstrating an additive effect (16–23). Even after ACEI and ARB were increased

to search for the optimal antiproteinuric dose, the combination of the two optimal doses had a stronger effect on proteinuria than either ACEI or ARB alone (24). Only one study compared the antiproteinuric effect of combined half doses of ACEI and ARB with full doses of each given as a monotherapy and demonstrated not only additive but also synergistic antiproteinuric effects of ACEI and ARB, particularly in patients with severe proteinuria (26). In contrast, combined ACEI and ARB treatment was not superior to either given as a monotherapy in patients with low levels of proteinuria (30,31). Combined ACEI and ARB may be superior to each treatment alone for BP but not for microalbuminuria control (31).

The benefit of combined half doses of ACEI and ARB seemed limited compared with the full dose of each monotherapy in our study. There was not even a trend in favor of combined half doses of ACEI and ARB compared with full doses of each monotherapy, which would have suggested that increasing the size of our population would reveal a significant difference between the groups. However, our study differed from the Campbell study in three major ways (26). First, highly selected nondiabetic patients were included in the Campbell study, because BP was controlled with fewer than two antihypertensive drugs and no RAS-blocking agent. In contrast, we included patients with a more severe hypertensive condition, with a mean home systolic BP >149 mmHg despite ramipril at 5 mg/d and a mean of 2.6 antihypertensive drugs. Second, our patients had a lower salt intake throughout the study: 10 g/d, which is below the 12 g/d of the Campbell study (26). Moreover, nine of our 18 patients also received diuretics at baseline, and serum creatinine levels increased significantly after the furosemide dose was raised during the last treatment period, suggesting that our patients were not overhydrated. Excessive salt intake can inhibit the antiproteinuric effect of ACEI (27) and could make patients more dependent on combined ARB. Finally, two different ACEI were used. Although the mechanism of the putative synergistic effect of combined ACEI and ARB on proteinuria, fibrosis (32), and renal failure progression (25) remains

to be determined, differential effect of ACEI and ARB on tissue angiotensin II might play a role (33).

Some discrepancy between urinary protein/creatinine ratio and 24-h proteinuria changes in the ramipril 10 group was noted, which was only partly due to imputation of missing data. We anticipated that good quality 24-h urine samples might be difficult to obtain; therefore the primary outcome measurement, urinary protein/creatinine ratio, was chosen to circumvent this problem. This discrepancy had a marginal statistical impact, because the only consequence was that combined ramipril 5, valsartan 80, and increased furosemide dosage treatment significantly decreased 24-h proteinuria with only a similar trend for the urinary protein/creatinine ratio ($P = 0.06$) compared with ramipril 10.

A secondary comparison in this study suggests that a cautious increase in diuretics in addition to combined ACEI and ARB enabled a better control of both proteinuria and BP but significantly increased serum creatinine levels. Although a better BP control and a rise in serum creatinine both may contribute to decreasing proteinuria, the beneficial effect of increased diuretic dosage on proteinuria was confirmed by a statistical adjustment for changes in serum creatinine and systolic home BP. When serum creatinine and systolic home BP were not included as fixed effects in the linear mixed model, differences between treatment groups were slightly decreased, showing that these variables contributed mainly to a loss of data homogeneity and only to part of the observed differences. It is unlikely that the proteinuria decrease observed with combined ramipril, valsartan, and increased furosemide dosage could be due to the absence of a washout period before this last treatment period, because the urinary protein/creatinine ratio was not significantly higher during the washout periods with ramipril 5 compared with ramipril 10, valsartan 160, and combined ramipril 5 valsartan 80. However, we did not test for the effect of increased diuretic dosage in association with ramipril 10 or valsartan 160, and these latter strategies might be just as effective. Indeed, a sodium load can inhibit the antiproteinuric effect of ACEI, and this effect could be restored by a diuretic (27,34,35). A secondary comparison of our study suggests that this may hold true for combined ACEI and ARB. In fact, the additive effect of combined full-dose ACEI and ARB in the Laverman study was achieved during sodium restriction (24). However, further studies are required to determine whether diuretics increase the antiproteinuric effect of maximum RAS blockade with combined ACEI and ARB at their maximum recommended dose.

In an unplanned subgroup analysis, the decrease in the urinary protein/creatinine ratio from baseline with low doses of ACEI tended to be greater in individuals with diabetes than in those without diabetes with high dose of ACEI or ARB, without reaching statistical significance ($P = 0.08$). Caution should be exercised in interpreting this small subgroup analysis, which, nevertheless, is in line with previous observations. Indeed, individuals with type 2 diabetes develop abnormalities in size-selective function of the glomerular barrier that may not be improved by low doses of ACEI (29) and may contribute to the

questionable impact of ACEI on renal failure progression in these patients (36).

The method used in our study, with 4-wk washout periods with ramipril at 5 mg/d between treatments, enables a comparison of tolerance, because no carryover effect is observed after a 4-wk withdrawal of a previous ACEI or ARB treatment. However, it was believed to be unethical to obtain a true baseline with a washout of all RAS-blocking agents and diuretics in these patients with severe hypertension and proteinuria. There was no difference in the frequency of symptomatic hypotension between treatments, and these were always mild. Although the tolerance of combined ACEI and ARB is reported to be excellent in short-term studies (37), the safety of long-term treatments remains to be determined. The rise in serum creatinine during diuretic treatment reinforcement was <18%, in the range of accepted changes for subsequent nephroprotection. However, caution should be exercised during long-term treatments, because acute extrarenal salt loss might potentially lead to more severe hypotensive episodes and prerenal failure. Therefore, long-term studies are required to evaluate the impact of aggressive proteinuria lowering with maximum tolerated doses of diuretics and RAS-blocking agents on renal failure progression. Inadequate monitoring of prerenal failure might indeed potentially lead to a J-curve, with a poor long-term renal outcome despite a low level of proteinuria.

Future antiproteinuric treatments may include other drugs in addition to ACEI, ARB, and diuretics. A low-protein diet and angiotensin II blockade produce additive therapeutic effects in experimental glomerulonephritis (38), but such effects have not been demonstrated in humans with optimal RAS blockade. Nonsteroidal anti-inflammatory drugs may be difficult to use in this context of potential prerenal failure (39). Statins will probably prove to be the safest step toward optimal treatment of patients with refractory proteinuria (40,41).

In conclusion, our study does not confirm that combined half doses of ACEI and ARB are superior to full doses of each given as monotherapy in patients with severe proteinuria and hypertension. Nevertheless, a secondary comparison of our study suggests that a cautious increase in diuretics in addition to RAS-blocking agents decreases both proteinuria and BP. However, signs of prerenal failure should be monitored on long-term treatment.

Reference

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