

Original Article

Effect of losartan and amlodipine on proteinuria and transforming growth factor- β 1 in patients with IgA nephropathy

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Abstract

Background. Transforming growth factor- β 1 (TGF- β 1) is the major profibrotic cytokine involved in many renal diseases, and urinary TGF- β 1 reflects intrarenal TGF- β 1 production. Urinary TGF- β 1 excretion is reported to be significantly increased in patients with immunoglobulin A (IgA) nephropathy. The aim of the present study was to compare the effects of losartan and amlodipine on proteinuria, as well as on serum and urine TGF- β 1 levels in IgA nephropathy patients with hypertension and proteinuria.

Methods. The initial 4 week washout period was followed by 12 weeks of active treatment, in which patients were randomized to once-daily treatment with losartan 50 mg (group 1, $n=20$) or amlodipine 5 mg (group 2, $n=16$). Urinary protein and TGF- β 1 excretion, serum TGF- β 1 and other clinical parameters were determined at baseline and during 12 weeks of active treatment.

Results. Both treatments controlled blood pressure (BP) to a similar degree, and renal function and other biochemical parameters did not change during the study period. Urinary protein and TGF- β 1 excretions were significantly elevated in IgA nephropathy patients. Losartan significantly reduced urinary protein (from 2.3 ± 1.5 g/day at baseline to 1.2 ± 1.5 g/day at 12 weeks, $P < 0.05$) and urinary TGF- β 1 excretion (from 31.2 ± 14.0 pg/mg creatinine at baseline to 22.1 ± 13.5 pg/mg creatinine at 12 weeks, $P < 0.05$). In contrast, amlodipine had no effect on urinary protein and TGF- β 1 excretion. Both losartan and amlodipine failed to reduce serum TGF- β 1 levels.

Conclusion. Losartan and amlodipine, with similar control of BP, showed different effects on urine protein or TGF- β 1 excretion. Whereas losartan improved both urinary parameters, amlodipine did not. These

differences might be important for the management of IgA nephropathy.

Keywords: amlodipine; IgA nephropathy; losartan; proteinuria; transforming growth factor- β 1

Introduction

Transforming growth factor- β 1 (TGF- β 1) is a profibrogenic cytokine involved in the synthesis of extracellular matrix, decreasing its degradation and stimulating the synthesis of integrin matrix receptors [1]. Progressive nephropathies are characterized by the enhanced accumulation of extracellular matrix in the kidney, and overproduction of TGF- β 1 can result in pathological tissue fibrosis through the accumulation of extracellular matrix proteins [2]. Many studies have shown that TGF- β 1 expression is increased in various types of human glomerulonephritis, including diabetic nephropathy, lupus nephritis, human immunodeficiency virus nephropathy and immunoglobulin A (IgA) nephropathy [3–5].

Treatment with blockers of angiotensin II actions, such as angiotensin-converting enzyme (ACE) inhibitors or angiotensin II receptor antagonists (ARAs), retarded renal disease progression in humans and ameliorated proteinuria in several models of kidney damage [6]. Experimental animal and *in vitro* studies recently have demonstrated that ACE inhibitors and ARAs may also decrease the synthesis and secretion of renal TGF- β 1 [7,8]. In a prospective study, Campistol *et al.* [9] demonstrated that losartan reduces TGF- β 1 plasma levels in kidney transplant patients with chronic allograft nephropathy, while amlodipine did not significantly change them. Similarly, captopril reduced TGF- β 1 serum levels in patients with type 1 diabetes and overt nephropathy participating in the Collaborative Study Captopril Trial [10]. It has been speculated that inhibition of angiotensin II action induced by losartan may decrease TGF- β 1 production

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by the kidneys in chronic renal disease as well as reduce the urinary protein excretion independently of its blood pressure (BP) lowering effects.

Urinary TGF- β 1 excretion reflects TGF- β 1 production in the kidney, and clinical and experimental studies have suggested that measurement of urinary TGF- β 1 content may be a useful non-invasive procedure for the evaluation of the renal production of this polypeptide [11]. In humans, urinary TGF- β 1 excretion has been shown to be significantly increased in patients with membranous nephropathy [12], diabetic nephropathy [13], crescentic glomerulonephritis and IgA nephropathy [12].

The aim of this study was to compare the effects of the ARA, losartan, with those of the calcium channel blocker, amlodipine, on proteinuria, urinary TGF- β 1 excretion and serum TGF- β 1 levels in IgA nephropathy patients with hypertension.

Patients and methods

Patient selection

Thirty-eight hypertensive IgA nephropathy patients with serum creatinine <3.0 mg/dl, and urine protein excretion >1 g/day were screened for enrollment.

Exclusion criteria included evidence or suspicion of diabetes, history of malignant hypertension, systolic BP >210 mmHg, cerebrovascular accident, transient ischaemic attacks or myocardial infarction within the previous 12 months, clinically significant arteriovenous conduction disturbances and/or arrhythmias, unstable angina, history of heart failure, serum potassium ≥ 5.5 mmol/l or ≤ 3.5 mmol/l, treatment with oral corticosteroids, and concomitant use of agents that may affect BP and proteinuria except α - or β -blockers, diuretics and nitrates. Urine and serum samples from healthy volunteers ($n=22$) served as controls.

Study design

The study consisted of 1 week of screening, a 4-week washout period and a 12-week active treatment period in which patients were randomized to once-daily treatment with losartan 50 mg or amlodipine 5 mg. After screening assessment, current antihypertensive medications, other than α - or β -blockers and/or diuretics, were discontinued during the washout period. Additional antihypertensive agents other than ACE inhibitors, ARAs and calcium channel blockers were prescribed to achieve the goal of 125/75 mmHg during the active treatment period. All trial medications were administered once daily in the morning. The subject's diets were unchanged, with no additional sodium or protein restriction.

The antihypertensive effects of the study medication were evaluated by the results of the clinic BP measurements at each out-patient visit (baseline, 4 and 12 weeks). Clinic BP was measured with a standard mercury sphygmomanometer and an appropriately sized cuff, and with Korotokoff phases I and V for the systolic and diastolic values, respectively. The means of three intermediate readings were recorded. Mean arterial BP was calculated as the sum of one-third of the systolic and two-thirds of the diastolic BP. Tolerability of study treatment was assessed by monitoring spontaneous

reports of adverse experiences at each visit. All patients provided written informed consent to participate, and the protocol was approved by the ethics committee of our hospital.

Measurement methods

A fasting blood sample was taken and a fresh morning random urine collection was obtained at baseline, 4 weeks and 12 weeks of active treatment for routine haematology, blood chemistry, serum TGF- β 1 and urinalysis evaluations. Twenty-four hour urine collections were performed at baseline, 4 weeks and 12 weeks of active treatment for measurement of protein, sodium, potassium, urea excretion and urinary TGF- β 1 levels.

Urine preparation

Aliquots of validated 24 h urine collections were centrifuged for 5 min to remove cells and particulate matter, and the supernates were treated with 1 mM phenylmethylsulfonyl fluoride (PMSF) and stored at -20°C . Samples were thawed rapidly and centrifuged for 5 min at 2000 r.p.m. to remove any urates or phosphates before use for assays.

TGF- β 1 assay of urine and serum samples

Urine and serum samples were analysed for TGF- β 1 using a commercially available solid-phase enzyme-linked immunosorbent assay (ELISA; Quantikine; R&D Systems, Minneapolis, MN). To activate latent TGF- β 1 into immunoreactive TGF- β 1 detectable by the immunoassay test, we performed an acidification and neutralization procedure (pH 7.2–7.6). The TGF- β 1 in the urine sample was activated by incubation with 1.0 N HCl for 10 min followed by neutralization with 1.2 N NaOH/0.5 mol/l HEPES buffer. The TGF- β 1 in the serum sample was activated by 2.5 N acetic acid/10 M urea and neutralized by 2.7 N NaOH/1 M HEPES. Urinary TGF- β 1 excretion is reported per mg of urinary creatinine to correct the variation in urine concentration. TGF- β 1 standard curves for urine and serum samples were prepared using RD5I and RD6M calibrator diluents, respectively. The minimum detectable level of TGF- β 1 with the test was 7 pg/ml.

Statistical analysis

All results are expressed as the mean \pm SD. Analysis of variance for repeated measures and paired *t*-test were used for within-group comparisons. Due to the non-normality of the serum levels of TGF- β 1 and 24 h urine data, changes from baseline were analysed using the Wilcoxon signed-rank test. Independent samples *t*-test and Mann–Whitney U-test were used for between-treatment group comparisons. Statistical significance was defined as a value of $P < 0.05$ (two sided).

Controlling for a type 1 error of 5% and taking a power of 80% (type II error of 20%), 16 patients would be needed to be recruited in each group in order to detect between-group changes from baseline of 50%. It was decided to recruit ~ 38 patients to allow for patients who may drop out of the study.

Table 1. Baseline patient characteristics

	Control (n=22)	Losartan (n=20)	Amlodipine (n=16)
Male/female	16/6	9/11	9/7
Age (years)	28.2 \pm 8.9	39.3 \pm 8.7	44.3 \pm 13.4
Weight (kg)	65.1 \pm 9.8	62.1 \pm 9.2	62.3 \pm 8.1
SBP (mmHg)	120 \pm 11	131 \pm 16	131 \pm 12
DBP (mmHg)	76 \pm 9	89 \pm 9	86 \pm 11
MAP (mmHg)	91 \pm 10	103 \pm 10	102 \pm 9
Antihypertensive medications			
Washout period	–	β -Blockers (2) Thiazide diuretics (2)	β -Blockers (2) Loop diuretics (1) Thiazide diuretics (2)
Active study period	–	α -Blockers (1) β -Blockers (2) Thiazide diuretics (8)**	α -Blockers (1) β -Blockers (2) Loop diuretics (1) Thiazide diuretics (3)
Urine protein (g/day)	0.05 \pm 0.02	2.3 \pm 1.5*	2.1 \pm 0.7*
Ccr (ml/min/1.73 m ²)	111 \pm 27	63 \pm 22*	63 \pm 24*
Urine TGF- β 1 (pg/mg creatinine)	2.3 \pm 1.1	31.2 \pm 14.0*	31.3 \pm 20.6*
Serum TGF- β 1 (ng/ml)	30.2 \pm 7.1	31.7 \pm 11.4	33.1 \pm 6.5

SBP, systolic blood pressure; DBP, diastolic blood pressure; MAP, mean arterial blood pressure; Ccr, creatinine clearance.

* P <0.05 vs control; ** P <0.05 vs amlodipine.

Table 2. Changes in biochemical profiles following treatment

	Losartan		Amlodipine	
	Baseline	12 weeks	Baseline	12 weeks
Hct (%)	39.6 \pm 4.6	39.0 \pm 3.8	39.8 \pm 5.3	38.4 \pm 4.4
Sodium (mEq/l)	140 \pm 2	140 \pm 3	139 \pm 2	138 \pm 2
Potassium (mEq/l)	4.3 \pm 0.3	4.2 \pm 0.5	4.2 \pm 0.5	3.9 \pm 0.2
Total protein (g/dl)	6.8 \pm 0.5	7.0 \pm 0.3	6.8 \pm 0.3	6.8 \pm 0.3
Albumin (g/dl)	4.0 \pm 0.3	4.1 \pm 0.4	4.0 \pm 0.3	3.9 \pm 0.3
Creatinine (mg/dl)	1.5 \pm 0.6	1.5 \pm 0.6	1.5 \pm 0.6	1.5 \pm 0.7
Uric acid (mg/dl)	6.4 \pm 1.6	6.4 \pm 1.6	7.1 \pm 1.9	6.8 \pm 2.1

Results

Patient characteristics and general parameters

Table 1 presents the baseline clinical characteristics of the losartan and amlodipine groups. Baseline gender distribution, age, weight and BP were similar in the two treatment groups. In both groups, antihypertensive drugs taken during the washout period were maintained during the active treatment period. There were no significant differences in the use of combination therapy ($P=0.95$) or other antihypertensive agents, such as β -blockers ($P=0.159$), for the two groups of patients during the whole study period (Table 1). However, more patients were using diuretics in the losartan group during the active study period.

Baseline urine TGF- β 1 levels of the losartan- and amlodipine-treated IgA nephropathy patients were significantly higher than those of the normal healthy controls (31.2 \pm 14.0 and 31.3 \pm 20.6 pg/mg creatinine vs 2.3 \pm 1.1 pg/mg creatinine, respectively, P <0.05). Baseline serum TGF- β 1 levels did not differ among the three groups.

Haematocrit, serum sodium, potassium, total protein, albumin, creatinine and uric acid did not change significantly from baseline to the end of either treatment period (Table 2). The creatinine clearance and changes in creatinine clearance after 12 weeks of treatment did not differ significantly between the two treatment groups (Table 3).

Blood pressure responses to treatment

The BP measurements are summarized in Figure 1. Twelve weeks of active treatment with losartan or amlodipine alone or in combination with other antihypertensive agents induced significant reductions in BP; from baseline mean arterial BP (MAP) 103 \pm 10 mmHg to 89 \pm 9 mmHg with losartan (P <0.01) and from 102 \pm 9 mmHg to 89 \pm 8 mmHg with amlodipine (P <0.01) after 12 weeks of treatment. Diastolic BP was reduced from 89 \pm 9 mmHg to 77 \pm 10 mmHg on losartan (P <0.01) and from 86 \pm 11 mmHg to 77 \pm 7 mmHg on amlodipine (P <0.01). Systolic BP was reduced from 131 \pm 16 mmHg to 113 \pm 9 mmHg

Table 3. Changes in urine parameters following treatment

	Losartan			Amlodipine		
	Baseline	4 weeks	12 weeks	Baseline	4 weeks	12 weeks
U _{Na} (mEq/day)	198 ± 50	199 ± 55	203 ± 55	204 ± 58	196 ± 58	199 ± 54
U _K (mEq/day)	64 ± 23	58 ± 13	57 ± 15	76 ± 24	72 ± 23	65 ± 22
U _{Pro} (g/day)	2.3 ± 1.5	1.4 ± 1.4*	1.2 ± 1.5*	2.1 ± 0.7	2.1 ± 0.8	2.2 ± 1.6
Random urine PCR	1.9 ± 0.9	1.3 ± 1.0	1.1 ± 1.2*	1.7 ± 1.1	1.9 ± 1.2	2.0 ± 1.4
Ccr (ml/min/1.73 m ²)	63 ± 22	64 ± 25	62 ± 22	63 ± 24	64 ± 22	64 ± 25
nPNA (g/kg/day)	1.3 ± 0.3	1.2 ± 0.2	1.2 ± 0.3	1.2 ± 0.3	1.2 ± 0.3	1.2 ± 0.2

U_{Na}, urinary sodium excretion; U_K, urinary potassium excretion; U_{Pro}, urinary protein excretion; PCR, protein creatinine ratio; Ccr, creatinine clearance; nPNA, normalized protein nitrogen appearance; * $P < 0.05$ vs baseline.

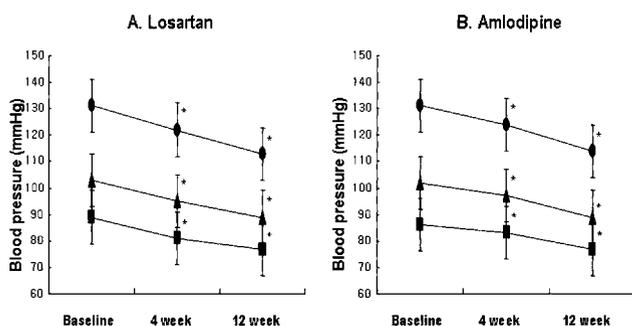


Fig. 1. Blood pressure responses to losartan (A) and amlodipine (B) treatment. * $P < 0.05$ vs baseline; circles, systolic blood pressure; squares, diastolic blood pressure; triangles, mean arterial blood pressure.

on losartan ($P < 0.01$) and from 131 ± 12 mmHg to 114 ± 9 mmHg on amlodipine ($P < 0.01$). No significant differences with regard to the control of BP were observed between the two treatment groups.

Effects on urine protein excretion

After 4 weeks of active treatment with losartan, urinary protein excretion significantly decreased from a baseline value of 2.3 ± 1.5 g/24 h to 1.4 ± 1.4 g/24 h (42.7% reduction from baseline). Urine protein excretion decreased further to 1.2 ± 1.5 g/24 h at 12 weeks (54.4% reduction from baseline) in patients on losartan (Table 3, Figure 2). Twelve weeks of amlodipine therapy increased urine protein excretion from a baseline value of 2.1 ± 0.7 g/24 h to 2.2 ± 1.6 g/24 h (not significant). The mean protein/creatinine ratio of the random urine specimen also significantly decreased from 1.9 ± 0.9 at baseline to 1.1 ± 1.2 after 12 weeks of losartan treatment ($P < 0.01$), whereas amlodipine treatment caused a slight increase in protein/creatinine ratio (from 1.7 ± 1.1 to 2.0 ± 1.4 after 12 weeks of active treatment). There were no significant changes in daily urinary sodium and potassium excretion, and normalized protein nitrogen appearance (nPNA) remained unchanged during the entire study period in both treatment groups (Table 3).

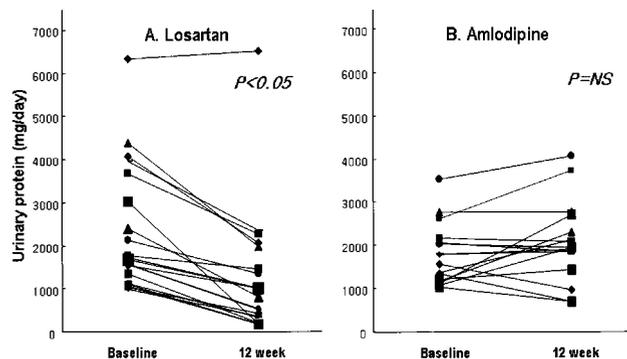


Fig. 2. Individual patient data for urinary protein excretion at baseline and after treatment with losartan (A) and amlodipine (B). NS, not significant.

Effects of treatment on urine and serum TGF- β 1

Baseline urinary TGF- β 1 levels did not differ between losartan- and amlodipine-treated patients; however, baseline urinary levels of total TGF- β 1 in IgA nephropathy patients were significantly greater than those of healthy controls (Table 1). In the losartan-treated group, urinary excretion of TGF- β 1 decreased significantly from baseline 31.2 ± 14.0 pg/mg creatinine to 23.0 ± 12.4 pg/mg creatinine after 4 weeks of treatment ($P < 0.05$) and was decreased further to 22.1 ± 13.5 pg/mg creatinine at 12 weeks of active treatment ($P < 0.05$). Conversely, in patients treated with amlodipine, urinary excretion of TGF- β 1 showed no significant change after the 12 weeks of treatment (Figure 3). After 12 weeks of treatment, urinary TGF- β 1 levels were significantly lower in the losartan treatment group compared with the amlodipine treatment group (22.1 ± 13.5 vs 29.6 ± 19.5 pg/mg creatinine, $P < 0.05$).

Serum levels of TGF- β 1 in control subjects did not show any significant difference when compared with those of IgA patients receiving losartan or amlodipine treatment (Table 1). Furthermore, serum levels of TGF- β 1 did not show any significant changes during treatment with either drug. There was no correlation between urine and serum TGF- β 1 levels.

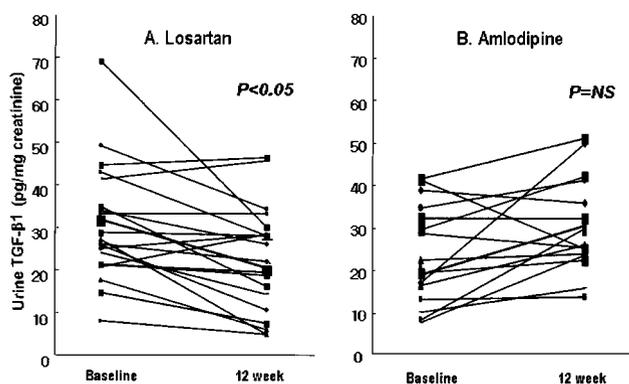


Fig. 3. Individual patient data for urinary excretion of TGF- β 1 at baseline and after treatment with losartan (A) and amlodipine (B). In losartan-treated patients, there was a 28% reduction (95% CI, 10–45, $P = 0.004$) in urinary TGF- β 1 excretion from 31.2 (95% CI, 23.7–38.6) to 22.1 (95% CI, 14.7–27.4) pg/mg creatinine. NS, not significant.

Correlation between urinary TGF- β 1 excretion and laboratory parameters

The relationship between urinary TGF- β 1 excretion and laboratory indices such as parameters of renal function and amount of urinary protein excretion were examined. Baseline urinary TGF- β 1 excretion did not correlate with creatinine clearance ($P = 0.86$, $r^2 = 0.021$) or baseline urinary protein level ($P = 0.96$, $r^2 = 0.121$). No significant correlation could be demonstrated between changes in urinary TGF- β 1 excretion and changes in proteinuria ($P = 0.86$, $r^2 = 0.256$) or BP in losartan-treated patients.

Discussion

In the present study, we demonstrated that despite similar control of BP with both losartan and amlodipine, the effects on proteinuria and urine TGF- β 1 excretion were completely different. Whereas losartan caused a significant decrease in both proteinuria and urine TGF- β 1 excretion, amlodipine caused no significant changes in either parameter. This different effect on proteinuria and urinary TGF- β 1 excretion could have potential repercussions for treatment of IgA nephropathy.

Treatment with blockers of angiotensin II actions, such as ACE inhibitors or ARAs, retarded renal disease progression and ameliorated proteinuria in several models of kidney damage [6]. In patients with renal disease, both with and without diabetes, it has been demonstrated that losartan reduces proteinuria to a similar extent to ACE inhibitors [3]. In patients with proteinuric IgA nephropathy and mild renal impairment, ACE inhibition and angiotensin II receptor blockade with enalapril or irbesartan for 28 days improved glomerular size selectivity and reduced proteinuria [14]. Longer treatment with enalapril or losartan also showed beneficial effects in patients with

IgA nephropathy with renal impairment and non-selective proteinuria. Such patients responded to therapy with improvement in protein selectivity, decrease in proteinuria and improvement in renal function [15]. The antiproteinuric effect of losartan has also been demonstrated in a double-blind, cross-over study comparing losartan with amlodipine in hypertensive patients with non-diabetic nephropathy. In this study, both losartan and amlodipine significantly lowered BP, but only losartan significantly reduced proteinuria after 4 weeks of treatment [16]. Our study also demonstrated that losartan alone or with the addition of low doses of other antihypertensive medications reduced proteinuria without significantly altering creatinine clearance in IgA nephropathy patients. The magnitude of reduction in urinary protein excretion after losartan treatment was $54.4 \pm 24.2\%$ at 12 weeks. This improvement in proteinuria is similar to the results of other clinical studies evaluating antiproteinuric effects of ACE inhibitors or ARAs, which vary from 30 to 60% according to various series [14,15].

Furthermore, the results of the present trial demonstrated that losartan was significantly more effective than amlodipine in reducing the urinary excretion of protein. These findings are in agreement with those reported by Holdaas and colleagues, who showed that urinary albumin excretion was significantly decreased in patients with non-diabetic renal disease following 4 weeks of treatment with losartan but was increased with a comparable course of amlodipine treatment [16]. The calcium channel blocker amlodipine documented no antiproteinuric effect despite a comparable antihypertensive effect in this study. The renal effects of calcium channel blockers remain controversial and may vary among different agents depending on the balance between direct renal actions and indirect effects due to a reduction in systemic BP. Dihydropyridine calcium channel blockers such as amlodipine dilate both afferent and efferent arterioles, which may increase proteinuria despite the reductions in systemic BP [17]. Results of clinical studies on the ability of calcium channel blockers to retard progression to renal failure have been variable and generally less striking than those with ACE inhibitors [6].

Renal TGF- β 1 expression is enhanced in patients with IgA nephropathy, and it has been reported that TGF- β 1 plays a pivotal role in the progression of IgA nephropathy [3]. Urine TGF- β 1 excretion reflects intrarenal production of TGF- β 1 [11], and several studies have found that urinary TGF- β 1 excretion is increased in patients with IgA nephropathy [18,19]. However, these clinical studies have not evaluated the effects of ARAs or calcium channel blockers on urinary TGF- β 1 excretion in IgA nephropathy patients. In the present study, patients with IgA nephropathy had up to 20 times higher urinary TGF- β 1 excretion compared with the healthy control group. At the end of the study, the reductions in urinary TGF- β 1 excretion after ARA treatment were fairly consistent with the data from previous experimental animal

studies, in which ACE inhibitors and ARAs decreased the synthesis and secretion of renal TGF- β 1 [7]. The main decrease in urinary TGF- β 1 excretion occurred within the first 4 weeks of losartan treatment, with a small further decrease at 12 weeks. A similar improvement in urinary TGF- β 1 level with losartan therapy has also been seen by Agarwal *et al.* [20] in patients with diabetic and non-diabetic glomerulonephritides. In their study, 4 weeks of add-on ARA treatment of patients already treated with the maximal dose of lisinopril resulted in early improvement in urinary TGF- β 1 excretion without changes in proteinuria or BP. However, treatment with losartan failed to normalize the urinary TGF- β 1 excretion to the control group level. Mean values of urinary TGF- β 1, although lower than at baseline, were significantly higher than those in the control group after 12 weeks of losartan therapy. This might have been due to production of renal TGF- β 1 via mechanisms other than angiotensin II stimulation, such as persistent proteinuria, pre-existing glomerular inflammation or non-strict dietary salt restriction in our study. Other reasons for sub-maximal response to ARA treatment might be the low dose of losartan or short treatment duration. The optimal antiproteinuric dose of losartan in non-diabetic patients with nephrotic range proteinuria has been reported to be 100 mg per day [21]. Further studies with increased dose and longer duration are needed to answer these questions.

Proteinuria and urine TGF- β 1 excretion may also decrease as a consequence of declining renal function or improved glomerular permselectivity. In this study, however, creatinine clearance did not change during the 12 weeks of treatment with losartan. The pattern of early urinary TGF- β 1 reduction after losartan treatment coincided with that of proteinuria reduction. This finding suggests that reduction in urinary TGF- β 1 excretion may reflect secondary effects of protein reduction by improved glomerular permselectivity caused by losartan. However, baseline urinary TGF- β 1 levels did not correlate with levels of proteinuria in our patients. In addition, the degree of reduction in urine TGF- β 1 levels after losartan treatment also did not correlate with the degree of reduction in proteinuria. Previous studies in diabetic nephropathy and membranous glomerulonephritis reported a strong correlation between increased urinary levels of TGF- β 1 and the extent of proteinuria [12,13]. However, those studies found no relationship between change in proteinuria and reduction in urinary TGF- β 1 excretion rate. In addition, other studies measuring urinary levels of TGF- β 1 in IgA nephropathy patients also found no correlations between baseline proteinuria and urine TGF- β 1 excretion [18,19]. Furthermore, in the present study, 20% of patients treated with losartan showed no significant decrease in urinary TGF- β 1 excretion, while only one patient did not respond to the antiproteinuric effect of losartan. These findings support the presence of diverse mechanisms involved in the stimulation of intrarenal TGF- β 1 production, such as angiotensin II [22],

proteinuria in the tubulointerstitium [23] and increased mechanical stress [24]. Therefore, it is difficult to discern clearly whether reductions in urinary TGF- β 1 excretion reflect an impairment in renal TGF- β 1 synthesis or a passive effect of proteinuria reduction by improved permselectivity.

Sharma *et al.* [10] showed that in patients with diabetic nephropathy enrolled in the Captopril Collaborative Study Group Trial and studied at baseline and 6 months after initiation of treatment with captopril, ACE inhibition lowered serum TGF- β 1 levels. However, proteinuric patients with IgA nephropathy in our study showed similar serum TGF- β 1 levels to those of the healthy control group, and both losartan and amlodipine failed to decrease serum TGF- β 1. Because serum TGF- β 1 levels reflect both platelet-derived and circulating TGF- β 1, an effect of losartan on platelet activation or production of TGF- β 1 in other cells may have contributed [10].

Our finding that losartan is associated with a significant reduction in urinary levels of TGF- β 1 suggests that ARAs may provide renal protection by blocking the production of renal TGF- β 1. However, due to the short observation period and lack of correlation with proteinuria reduction, which is a well known predictor of renoprotection, there is uncertainty regarding the long-term renoprotective effects of urinary TGF- β 1 reduction *per se*. Whether ARA-induced reduction in urinary TGF- β 1 excretion is associated with long-term renal protection requires larger and longer prospective, randomized controlled trials. Because we did not observe complete reductions in proteinuria and urine TGF- β 1 excretions, it remains unclear whether an even higher dose of an ARA would produce further reductions in urinary TGF- β 1 excretion.

In summary, the present study demonstrated that losartan and amlodipine were highly effective in the control of hypertension in proteinuric IgA nephropathy patients with hypertension. Whereas losartan treatment significantly reduced both proteinuria and urine TGF- β 1 excretion, amlodipine increased proteinuria and was without effect on urine TGF- β 1 despite similar BP reductions. These differences might be important for the management of proteinuric IgA nephropathy patients with mild to moderate hypertension.

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Conflict of interest statement. None declared.

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