ORIGINAL ARTICLE

Fibrinolysis and insulin sensitivity in imidapril and candesartan (FISIC study) recipients with hypertension

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The aim of this study was to evaluate the effects of imidapril and candesartan on fibrinolysis and insulin sensitivity in normoweight hypertensive patients. After a 2-week wash-out period, 61 patients with mild-to-moderate hypertension were randomized to imidapril or candesartan for 12 weeks. Blood pressure (BP), plasma tissue plasminogen activator (t-PA) and plasminogen activator inhibitor-1 (PAI-1) antigen activities were evaluated at baseline and during treatment. The patients underwent a euglycemic–hyperinsulinemic clamp (insulin sensitivity was evaluated as glucose infusion rate during the last 30 min) and a desmopressin test (with desmopressin infusion in the brachial artery) to evaluate endothelial ability to release t-PA. Imidapril and candesartan induced similar systolic/diastolic BP reductions ($-16/12.6$ and $-16.1/12.2$ mm Hg, respectively, $P < 0.001$ vs. baseline). Imidapril increased glucose infusion rate ($+1.1$ mg min $^{-1}$ per kg, $P < 0.02$), whereas candesartan did not change it. Both drugs decreased PAI-1 antigen activity after 4 weeks of treatment; subsequently, only the decreasing effect of imidapril was sustained throughout the 12 weeks, whereas candesartan increased PAI-1 activity at week 12 ($P < 0.05$ vs. baseline, $P < 0.01$ vs. imidapril). Activity of t-PA decreased with candesartan (from 0.48 ± 0.16 to 0.43 ± 0.14 IU ml $^{-1}$, $P < 0.05$) but not with imidapril. Activity of t-PA in response to desmopressin was increased more by imidapril ($+4.45$ IU ml $^{-1}$) than by candesartan ($+2.73$ IU ml $^{-1}$, $P < 0.01$ vs. imidapril). These results indicate that in normoweight hypertensive patients, despite similar BP reduction, imidapril but not candesartan improved the fibrinolytic balance, suggesting that mechanisms other than Ang II inhibition, possibly including bradykinin-mediated effects on insulin sensitivity and endothelial function, may be responsible for these different effects.

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INTRODUCTION

A growing body of evidence indicates that the rennin–angiotensin–aldosterone system, which has an important pathophysiological role in hypertension, can influence the balance of the fibrinolytic system by stimulating production of plasminogen activator inhibitor-1 (PAI-1) and by reducing tissue plasminogen activator (t-PA) production.^{1–3} Angiotensin II (Ang II) induces synthesis of PAI-1 in cultured endothelial cells,^{4,5} murine brain astrocytes⁶ and vascular smooth muscle cells.^{4,7} In agreement with these *in vitro* results, infusion of Ang II into healthy volunteers and hypertensive subjects produces a rapid increase in circulating PAI-1.⁸ Aldosterone is another factor through which the rennin–angiotensin–aldosterone system may influence PAI-1 level, as demonstrated *in vitro* and *in vivo*.^{1,9} Because bradykinin increases the release of t-PA from endothelial cells,^{10,11} angiotensin converting enzyme (ACE), which degrades bradykinin and catalyzes the conversion of Ang I to Ang II, might contribute to the reduction in t-PA production.

Clinical use of ACE inhibitors (ACE-Is) has been associated with a reduction in PAI-1 in most of the trials.^{12–17} The effects of ACE-Is on t-PA are more controversial, as some studies have shown increased t-PA levels,^{18,19} other studies have reported decreased t-PA levels^{12,20} and still others have observed unchanged t-PA levels during ACE inhibition.^{13,21,22} One possible explanation for these contrasting findings might be that t-PA exists in several forms, including free active t-PA and t-PA bound to PAI-1 and other inhibitors.²³ Moreover, the systemic levels of t-PA may not reflect the local profibrinolytic capacity in terms of availability of active t-PA at the organ level, which in turn depends on the magnitude of local t-PA release from the endothelium.^{24,25} Data on the effects of Ang II AT1 receptor blockers (ARBs) on fibrinolysis are even more controversial, with some studies showing a reduction in PAI-1 plasma level^{26–28} and others showing no effect.^{19,29–32} Reasons for such different findings might be because of differences in experimental models, study population characteristics, duration of treatment and possible methodological bias. Some direct

comparisons on the effects of ACE-Is and ARBs on plasma PAI-1 and t-PA have been performed, and again the results were controversial:^{33–38} Some studies have shown that both ACE inhibition and AT1 antagonism were associated with a significant improvement in plasma fibrinolysis,³⁴ whereas others have observed a decrease in PAI-1 and/or an increase in t-PA with ACE-Is but not with ARBs.^{33,35–37} Only a few studies have reported an improvement of fibrinolytic balance with ARBs.³⁸

With this background, the present study was undertaken to compare the effects of 12-week treatment with the ACE-I imidapril and the ARB candesartan on plasma PAI-1 antigen level and activity and on plasma t-PA activity level in a homogeneous population of normoweight (body mass index (BMI) $\leq 25 \text{ kg m}^{-2}$) hypertensive patients. The capacity for acute t-PA release from vascular endothelium before and after treatment with the two drugs has also been assessed through arterial infusion of desmopressin.^{39,40} *In vitro* studies show that proinsulin and insulin stimulate the synthesis of PAI-1 in human hepatocytes and endothelial cells,^{41,42} which has also been confirmed *in vivo*.⁴³ Additionally, insulin resistance with resulting hyperinsulinemia is frequently accompanied by increased PAI-1 level.^{44–46} Owing to this close relationship between insulin sensitivity and the fibrinolytic system, the effects of imidapril and candesartan on insulin sensitivity, as assessed by euglycemic–hyperinsulinemic clamp,⁴⁷ were evaluated.

METHODS

Study design

This was a 12-week, prospective, randomized, open-label, blinded-endpoint (PROBE),⁴⁸ parallel group study with two treatment arms.

Study population

Male and female normoweight outpatients, aged 18–65 years, with mild-to-moderate essential hypertension (defined as sitting systolic BP ≥ 140 and < 180 mm Hg and sitting diastolic BP ≥ 90 and < 105 mm Hg after a 2 week run-in wash-out period) associated with at least one other cardiovascular risk factor (smoking, echocardiographic left ventricular hypertrophy, male with age > 55 years, family history of cardiovascular disease or carotid intima–media thickness ratio < 0.9) were considered for enrollment.

Patients with secondary hypertension, BMI $> 25 \text{ kg m}^{-2}$, creatinine clearance $< 80 \text{ ml min}^{-1}$, total cholesterol $\geq 250 \text{ mg dl}^{-1}$, diabetes mellitus, history of myocardial infarction or stroke within 6 months before the study, congestive heart failure or any severe diseases likely to interfere with the conduction of the study were excluded, as were those with known contraindications or intolerance to ARB or ACE-Is.

Ethics

The study was performed in accordance with the Declaration of Helsinki and its amendments, and all patients gave their written informed consent to participate in the study at the time of enrollment.

Study protocol

After a 2-week run-in wash-out period, patients fulfilling the inclusion criteria were randomized to 5 mg imidapril or 8 mg candesartan, both given once a day in the morning (at approximately 0800 h) for 12 weeks. After 2, 4 and 8 weeks, a dose titration consisting of a progressively increasing amount of imidapril to 20 mg or candesartan to 32 mg was permitted in non-responder patients (systolic BP ≥ 140 mm Hg and/or diastolic BP ≥ 90 mm Hg). The doses of the two drugs were chosen to be equipotent with respect to blood pressure (BP) lowering. From the time of enrollment until the completion of the study, all participants maintained their usual diet and level of physical activity and avoided changes in body weight. No concomitant medication was allowed. Medication compliance was assessed by a residual capsule count at each visit.

Blood pressure, PAI-1 antigen level and activity and t-PA activity were evaluated at the end of the wash-out period and after 1, 2, 4, 8 and 12 weeks of treatment, whereas fasting plasma glucose, fasting plasma insulin, insulin sensitivity and the t-PA activity response to desmopressin were evaluated only at the end of the wash-out period and of each treatment period.

The BP measurements were obtained from each patient (right arm) in the seated position by using a standard mercury sphygmomanometer (Korotkoff I and V) with a cuff of appropriate size. Measurements were taken in the morning before daily drug intake (that is, 24 h after dosing, at trough) and after the subject had rested 10 min in a quiet room. Three successive BP readings were obtained at 1-min intervals and averaged.

Evaluation of fibrinolytic parameters

Venous blood was drawn from an antecubital vein for blood sampling at the same hour in the morning, approximately between 0800 and 0900 h, because it was confirmed that PAI-1 concentration peaked during this period in the preliminary experiments. Blood samples were collected on ice and centrifuged immediately at 0°C for 20 min. All plasma or serum samples were separated and stored at -70°C until assaying. Blood for measuring PAI-1 antigen was collected in Vacutainer tubes containing $0.105 \text{ mmol l}^{-1}$ acidified sodium citrate. PAI-1 antigen level was determined using a two-site enzyme-linked immunosorbent assay (Biopool AB, Umea, Sweden). PAI-1 activity was measured with an assay based on the method of Veheijen *et al.*,⁴⁹ with standardized commercial kits (Biopool AB, Umea, Sweden). Because t-PA is bound by PAI-1, which inactivates it, the antigen level differs from the active level, which is determined only by t-PA unbound to PAI-1. Therefore, we measured only plasma t-PA activity, which was determined using an immunofunctional assay (Chromalize, Biopool AB, Umea, Sweden).

Desmopressin test

The capacity for acute stimulated t-PA release from vascular endothelium was assessed by the desmopressin test,^{39,40} which was performed after an overnight fast on the second-to-last day of the wash-out period and of each treatment period. An arterial polyethylene catheter was introduced percutaneously into the brachial artery of the non-dominant arm, and an indwelling cannula was introduced retrogradely into a deep antecubital vein of the same arm for venous blood sampling from the muscle vascular bed; the cannula was flushed with heparinized (5 IU ml^{-1}) saline. After catheterization, patients rested for 30 min in the supine position. Then, a venous blood sample was obtained to evaluate baseline t-PA activity. Soon afterwards, the arterial desmopressin infusion began. Desmopressin was diluted in saline and was administered at a dose of 70 ng min^{-1} for 15 min. The infusion was carried out at a constant rate of 1 ml min^{-1} by means of a syringe infusion pump. After 15 minutes, another venous blood sample was obtained to evaluate the t-PA activity response to desmopressin. The t-PA release experiments were not corrected for flow because previous data have demonstrated that the t-PA increase in response to desmopressin is independent of flow increase.^{39,40}

Evaluation of insulin sensitivity and glucose metabolism parameters

Insulin sensitivity was assessed by euglycemic–hyperinsulinemic clamp, according to the technique of De Fronzo *et al.*⁴⁷ At 0900 h, after the subjects had fasted for 12 h overnight, an intravenous catheter (18 g polyethylene cannula, Venflon, Viggo, Sweden) was placed in an antecubital vein for infusion of insulin and 20% glucose. A second catheter was inserted retrogradely into a wrist vein. The hand was heated (about 70°C) in a thermoregulated box to arteriaize venous blood within 20–40 min. Plasma glucose was assessed at 5–10 min intervals during the clamp. A 10-min priming infusion of insulin (Humulin R, Lilly, Indianapolis, IN, USA) was administered at rate of 1 mU min^{-1} per kg for 2 h, during which the plasma glucose concentration was held constant at the basal state (95 mg dl^{-1}) by a variable infusion of exogenous glucose. The amount of glucose required for maintaining isoglycemia equals the whole-body disposal of glucose, provided that endogenous glucose production is essentially absent. During insulin infusion, normal fasting blood glucose level was maintained by adjusting the infusion rate of a 20% glucose solution. The amount of glucose taken up (mg min^{-1} per kg) was calculated for each 10 min interval after the

first 20 min of the clamp. Insulin sensitivity was calculated from the mean glucose uptake rate for the last 30 min of the clamp and expressed as the amount of glucose infused during that time (glucose infusion rate) in mg min^{-1} per kg.

Blood glucose in the fasting state and during the glucose clamp was measured by the glucose oxidase method (Beckman Auto-Analyzer, Fullerton, CA, USA). Plasma insulin concentration was determined by radioimmunoassay.

Statistical analysis

The results were expressed as means $\pm$ s.d. All data were analyzed by a split-plot method for analysis of variance; the data for PAI-1 were also analyzed for multiple comparisons by the Dunn-Sidak method for split-plot analysis of variance. Differences were considered significant when the *P*-value was less than 0.05.

RESULTS

A total of 68 patients were screened between February and September 2008. At the end of the 2-week run-in period, 61 patients were randomized to receive imidapril 5 mg ($n=30$) or candesartan 8 mg ($n=31$). Their main demographic and clinical characteristics are shown in Table 1. The two treatment groups were comparable in terms of age, sex, BMI, baseline sitting BP, total cholesterol, creatinine clearance, smoking habits, prevalence of left ventricular hypertrophy and of carotid intima-media thickness >0.9 and duration of hypertension. Five patients withdrew after randomization (two withdrew the informed consent and three complained of side-effects). A total of 56 patients, 28 in the imidapril group and 28 in the candesartan group, completed the study. At 8 weeks, 96.4% of patients in both treatment groups required dose titration.

The BP effects of the two agents are shown in Table 2. Both imidapril and candesartan significantly reduced sitting systolic BP (-16 and -16.1 mm Hg, respectively; both $P<0.001$ vs. baseline) and diastolic BP (-12.6 and -12.2 mm Hg, respectively; both $P<0.001$ vs. baseline). The effects of the two drugs on BP were statistically similar and sustained for the duration of the study.

Treatment with either imidapril or candesartan decreased plasma PAI-1 antigen during the first 4 weeks; subsequently, only the effect of imidapril was sustained throughout the 12-week period, whereas in the candesartan group PAI-1 antigen returned to the baseline value at week 8 and significantly increased at 12 weeks (both were statistically

different from baseline, $P<0.05$, and from imidapril, $P<0.01$) (Table 3) (Figure 1). Similar results were obtained for PAI-1 activity, which paralleled PAI-1 antigen behavior in both treatment groups (Table 3).

At the end of 12 weeks, t-PA activity had decreased from candesartan treatment (from 0.48 ± 0.16 to 0.43 ± 0.14 IU ml^{-1} at week 12, $P<0.05$), but not from imidapril (from 0.47 ± 0.15 to

Table 2 Effect of treatment on blood pressure

Time	Imidapril (n=28)	Candesartan (n=28)
<i>Seated SBP mm Hg</i>		
Baseline	148.4 $\pm$ 4.8	149.0 $\pm$ 4.9
Week 1	144.1 $\pm$ 5.1*	145.6 $\pm$ 4.9*
Week 2	142.8 $\pm$ 4.8†	143.5 $\pm$ 4.9†
Week 4	136.8 $\pm$ 4.9‡	136.9 $\pm$ 5.1‡
Week 8	133.6 $\pm$ 4.2‡	134.3 $\pm$ 4.5‡
Week 12	132.4 $\pm$ 4.1‡	132.9 $\pm$ 4.4‡
<i>Seated DBP mm Hg</i>		
Baseline	98.7 $\pm$ 4.4	98.5 $\pm$ 4.0
Week 1	95.4 $\pm$ 4.5*	95.8 $\pm$ 3.9*
Week 2	94.8 $\pm$ 4.3†	94.1 $\pm$ 3.8†
Week 4	90.1 $\pm$ 3.9‡	88.9 $\pm$ 4.1‡
Week 8	86.9 $\pm$ 2.6‡	87.1 $\pm$ 3.2‡
Week 12	86.1 $\pm$ 2.6‡	86.3 $\pm$ 3.2‡

Abbreviations: DBP, diastolic blood pressure; SBP, systolic blood pressure.

* $P<0.05$; † $P<0.01$; ‡ $P<0.001$ vs. baseline.

Table 3 Effect of treatment on plasma PAI-1 antigen, PAI-1 activity and on t-PA activity

Time	Imidapril (n=28)	Candesartan (n=28)
<i>PAI-1 antigen (ng ml^{-1})</i>		
Baseline	21.9 $\pm$ 10.9	21.0 $\pm$ 11.2
Week 1	15.2 $\pm$ 7.1*	15.3 $\pm$ 7.4*
Week 2	14.0 $\pm$ 6.7*	17.1 $\pm$ 8.1*
Week 4	15.5 $\pm$ 6.5*	18.9 $\pm$ 8.8
Week 8	16.0 $\pm$ 6.5*	21.3 $\pm$ 9.4†
Week 12	15.4 $\pm$ 6.8*	24.5 $\pm$ 10.6‡§
<i>PAI-1 activity (IU ml^{-1})</i>		
Baseline	9.2 $\pm$ 3.6	9.7 $\pm$ 3.8
Week 1	6.8 $\pm$ 2.4*	7.1 $\pm$ 2.5*
Week 2	6.5 $\pm$ 2.3*	8.1 $\pm$ 3.1*
Week 4	6.7 $\pm$ 2.5*	8.8 $\pm$ 3.7
Week 8	7.1 $\pm$ 2.8*	9.8 $\pm$ 4.1†
Week 12	6.4 $\pm$ 2.2*	11.5 $\pm$ 4.6‡§
<i>t-PA activity (IU ml^{-1})</i>		
Baseline	0.47 $\pm$ 0.15	0.48 $\pm$ 0.16
Week 1	0.50 $\pm$ 0.13*	0.47 $\pm$ 0.15
Week 2	0.49 $\pm$ 0.13*	0.46 $\pm$ 0.15
Week 4	0.47 $\pm$ 0.12	0.44 $\pm$ 0.14*
Week 8	0.47 $\pm$ 0.13	0.43 $\pm$ 0.14*‡
Week 12	0.49 $\pm$ 0.14	0.43 $\pm$ 0.14‡§

Abbreviations: PAI-1 antigen, plasminogen activator inhibitor-1 antigen; t-PA, tissue plasminogen activator. * $P<0.05$ vs. baseline; † $P<0.02$ vs. imidapril; ‡ $P<0.05$ vs. imidapril; § $P<0.01$ vs. imidapril.

Table 1 Subjects' characteristics

Parameters	Imidapril (n=30)	Candesartan (n=31)
Age (years)	53.0 $\pm$ 10.8	53.2 $\pm$ 9.7
Gender (Male/female)	16/14	16/15
Male >55 years	10	8
BMI (kg m^{-2})	23.8 $\pm$ 1.08	23.9 $\pm$ 1.07
SBP (mm Hg)	148.3 $\pm$ 5.2	148.8 $\pm$ 4.8
DBP (mm Hg)	97.6 $\pm$ 4.7	98.8 $\pm$ 4.2
Total cholesterol (mg dl^{-1})	207.6 $\pm$ 30.5	207.4 $\pm$ 29.1
Creatinine clearance	103.7 $\pm$ 11.1	103.6 $\pm$ 11
Family history for CVD (n)	10	10
Smokers (n)	7	8
Patients with LVH (n)	4	3
Patients with CIMT >0.9 mm (n)	3	4
FPG (mg dl^{-1})	88.9 $\pm$ 8.8	89.1 $\pm$ 8.9
FPI (μIU^{-1} per ml)	9.4 $\pm$ 2.7	9.5 $\pm$ 2.8
Duration of hypertension (years)	8.4 $\pm$ 2.5	8.6 $\pm$ 2.4

Abbreviations: BMI, body mass index; CIMT, carotid intima-media thickness; CVD, cardiovascular disease; DBP, diastolic blood pressure; FPG, fasting plasma glucose; FPI, fasting plasma insulin; LVH, left ventricular hypertrophy; SBP, systolic blood pressure.

$0.49 \pm 0.14 \text{ IU ml}^{-1}$, $P < 0.01$ vs. candesartan). The changes in t-PA activity over time are shown in Table 3 and Figure 1.

Desmopressin infusion markedly increased plasma t-PA activity to a similar extent in both treatment groups at the end of wash-out period ($+1.4 \text{ IU ml}^{-1}$ in the imidapril group and $+1.4 \text{ IU ml}^{-1}$ in the

candesartan group). At the end of the treatment period, the t-PA activity response was further increased, but the increase was significantly greater in the imidapril group in which a threefold increase was observed ($+4.45 \text{ IU ml}^{-1}$), than in the candesartan group in which a twofold increase was observed ($+2.73 \text{ IU ml}^{-1}$, $P < 0.01$ vs. imidapril) (Figure 2).

The effects on glucose metabolism are shown in Table 4, which provides data on the effects of therapy on fasting plasma glucose, fasting plasma insulin and insulin sensitivity. Neither drug significantly affected fasting plasma glucose or fasting plasma insulin, whereas glucose infusion rate, which is considered an index of insulin sensitivity, was significantly increased by imidapril (by a mean of 1.1 mg min^{-1} per kg, $P < 0.01$) but not by candesartan ($+0.1 \text{ mg min}^{-1}$ per kg, $P = \text{non significant}$).

No significant change in BMI was observed in either of the treatment groups.

DISCUSSION

The results of this study show that in normoweight patients with mild-to-moderate hypertension, despite a similar BP-lowering effect, imidapril and candesartan displayed different effects on the

Figure 1 Changes in mean plasma PAI-1 antigen concentration and t-PA activity over time in response to treatment with imidapril or candesartan. * $P < 0.05$ vs. baseline; $^{\dagger}P < 0.02$ vs. imidapril; $^{\circ}P < 0.01$ vs. imidapril $P < 0.05$ vs. baseline; $^{\$}P < 0.05$ vs. imidapril; $^{\circ}P < 0.01$ vs. imidapril.

Table 4 Effect of treatment on metabolic parameters

Parameters	Imidapril (n=28)	Candesartan (n=28)
FPG (mg dl ⁻¹)		
Baseline	88.9 ± 8.8	89.1 ± 8.9
Week 12	88.4 ± 8.7	89.2 ± 8.8
FPI (μ IU ⁻¹ per ml)		
Baseline	9.4 ± 2.7	9.5 ± 2.8
Week 12	9.1 ± 2.6	9.4 ± 2.7
GIR (mg min ⁻¹ per kg)		
Baseline	5.2 ± 2.0	5.2 ± 1.8
Week 12	6.3 ± 1.8* [†]	5.3 ± 1.7

Abbreviations: FPG, fasting plasma glucose; FPI, fasting plasma insulin; GIR, glucose infusion rate. * $P < 0.01$ vs. baseline; $^{\dagger}P < 0.01$ vs. candesartan.

Figure 2 Plasma t-PA activity in response to desmopressin infusion at the end of the wash-out period and at the end of each treatment period. * $P < 0.01$ vs. Candesartan.

fibrinolytic balance. In fact, after 12 weeks of treatment, imidapril significantly reduced PAI-1 antigen level and activity without affecting plasma active t-PA level, whereas candesartan did not change PAI-1 but did decrease plasma active t-PA. In agreement with some previous observations,⁵⁰ when considering the effect of ACE inhibition and Ang II AT1 receptor blockade on plasma PAI-1 antigen over time, we observed a different time courses of the effects: short-term treatment with either imidapril or candesartan significantly reduced plasma PAI-1 antigen, whereas in the long term only imidapril decreased PAI-1 antigen. Similar results were obtained for plasma PAI-1 activity. Different time courses of effects of ACE inhibition and Ang II AT1 receptor blockade on t-PA concentrations were also observed, because t-PA level decreased significantly over time in candesartan but not in imidapril treatment.

The different effects of imidapril and candesartan on the fibrinolytic balance observed in the present study were not caused by a difference in BP-lowering effects because the extent of BP decrease was similar in both treatment groups. As obesity is one of the factors that activate renin angiotensin system (RAS), only patients with normal body weight were included in the study to avoid the influence of body weight on the fibrinolytic process.⁵¹ No significant change in BMI was observed throughout treatment with either imidapril or candesartan, so the observed modifications in the fibrinolytic parameters were not related to changes in body weight.

One possible explanation for the dissimilar effects of imidapril and candesartan on the fibrinolytic balance might be their different effects on kinin metabolism. Unlike ARBs that do not affect the metabolism of bradykinin, ACE-Is may exert a beneficial effect on fibrinolysis by blocking the degradation of bradykinin, which stimulates t-PA synthesis and release from endothelial cells through a bradykinin type-2 receptor-dependent pathway.^{10,11}

The dissimilar effects of imidapril and candesartan on PAI-1 could have been due in part to increased PAI-1 expression by the hexapeptide Ang II metabolite Ang IV and to stimulation of AT4 receptor, as reported *in vitro* in human endothelial cells.⁵² These effects could have increased PAI-1 antigen concentration after short-term AT1 receptor blockade and after 2 week AT1 blockade, when AT4 receptors may be upregulated.⁵³ This mechanism is difficult to determine in a clinical setting and remains undetermined in the present study. Another explanation is that Ang II-induced PAI-1 expression is mediated by AT1 receptors, and the brevity of the effect of candesartan might then depend on AT1 receptor upregulation.⁵⁰ More specifically, after PAI-1 was decreased in weeks 1 and 2 because of AT1 blockade, AT1 was upregulated and PAI-1 was increased. It has also been hypothesized that the different time courses of the effects of ACE-I and ARB on PAI-1 level result from differences in the duration of suppression of tissue Ang II, which is suppressed by ACE-I and not suppressed by ARB.⁵⁰

Furthermore, the dissimilar effects on fibrinolysis of imidapril and candesartan might depend on their different effects on insulin sensitivity, whose relationship with PAI-1 level, observed in cross-sectional studies, has been confirmed by interventional studies aimed at reducing insulin resistance, which have demonstrated a parallel decrease in plasma insulin and PAI-1 levels.^{45,46,54} In the present study, imidapril significantly increased insulin sensitivity, as assessed by euglycemic-hyperinsulinemic clamp, whereas candesartan did not. These findings, which are in agreement with previous observations by ourselves and other authors,^{35,55–57} besides providing another possible explanation for the different effects of the two drugs on the fibrinolytic balance, also suggest that mechanisms other than Ang II inhibition, such as the increase in endogenous

kinins because of ACE inhibition, might be responsible for the observed different influences of imidapril and candesartan on insulin sensitivity.

Systemic administration of desmopressin increases t-PA activity.^{39,40} Although the mechanisms behind this response are not fully defined, it has commonly been assumed that desmopressin induces endothelial release of t-PA.^{39,40} Therefore, in the present study the capacity for acute t-PA release from vascular endothelium before and after treatment with imidapril or candesartan was evaluated through desmopressin infusion as an index of endothelial function. We observed: (1) a greater increase in t-PA activity in response to desmopressin after 12-weeks treatment with both imidapril and candesartan than compared with baseline; and (2) this increase was significantly more pronounced with imidapril than compared with candesartan. These findings suggest that antihypertensive therapy with both the drugs improved the capacity for acute t-PA release, probably through an improvement of endothelial function. The reduction in the vascular shear stress resulting from the hypotensive action of both imidapril and candesartan might partly explain such an improvement compared with baseline. However, the different influences of ACE inhibition and Ang II AT1 receptor blockade on bradykinin metabolism and kinin-mediated t-PA release from human endothelium might be one possible explanation for the observed greater improvement with imidapril.

Although it is accepted that imidapril is a selective ACE-I and candesartan is an ARB, the selectivity of the compounds is generally relative, and the potential actions associated with the compound themselves cannot be completely excluded, which may constitute a limitation of this study. Further studies with other agents are needed to generalize the current investigation to the characteristics of these classes of agents.

The results of this study show that in normoweight hypertensive patients, 12-weeks treatment with the ACE-I imidapril or the ARB candesartan, despite similar BP-lowering effects, displayed different effects on the fibrinolytic balance and insulin sensitivity: imidapril but not candesartan significantly improved them. Different time courses of the effects of the two drugs on the fibrinolytic parameters and a significantly greater t-PA activity increase in response to desmopressin with imidapril were also observed. These findings imply that mechanisms other than direct inhibition of Ang II-mediated signaling, such as the kinin-mediated effects on insulin sensitivity and bradykinin-stimulated t-PA release from human endothelium, may have crucial roles in the differential effects of imidapril and candesartan on the fibrinolytic balance, whereas Ang II signaling could also contribute to the difference.

Many other studies have demonstrated the beneficial effects of ACE-I on prothrombotic state,^{12–17} as well as insulin sensitivity^{58–60} in addition to their efficacy in normalizing elevated BP, which might result in better cardiovascular protection by these drugs. In this regard, a meta-analysis of clinical studies has shown that ACE-I but not ARB may reduce the risk of major coronary disease events.⁶¹ Although various underlying mechanisms might give rise to this significant difference, the positive effects of ACE-Is on the fibrinolytic balance might contribute to the greater reduction in myocardial infarction rate achieved in clinical trials with these drugs.

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