Effects of losartan on cerebral and ocular circulation in healthy subjects

Bettina Matulla,¹ Gabriele Streit,¹ Stefan Pieh,² Oliver Findl,² Jesusa Entlicher,¹ Ursula Graselli,¹ Hans-Georg Eichler,¹ Michael Wolzt¹ & Leopold Schmetterer^{1,3}

¹Department of Clinical Pharmacology, ²Department of Ophthalmology B and ³Institute of Medical Physics, Vienna, Austria

Aims The introduction of specific inhibitors of AT₁ receptors, such as losartan, has enabled the investigation of the renin-angiotensin-system (RAS) in humans *in vivo*. We studied the role of the RAS in the cerebral and ocular circulation in healthy subjects. Haemodynamic effects of orally administered losartan were investigated with non-invasive methods.

Methods In a placebo-controlled randomized, double-blind two way crossover design losartan (100 mg orally) or placebo was administered in 10 healthy subjects. The effect of losartan was studied at hourly intervals for 8 h. In addition, the effect of losartan on haemodynamic changes induced by exogenous angiotensin II (Ang II) was assessed. Blood flow velocities in the ophthalmic and the middle cerebral artery (OA, MCA) were measured with Doppler sonography. Pulsatile choroidal blood flow was estimated with laser interferometric measurement of fundus pulsation.

Results Losartan significantly increased fundus pulsation amplitude (+11%, 95% CI: 5 to 16% $P < 0.0001$), tended to increase resistive index in the ophthalmic artery (+12%, 95% CI: 0 to 23%) and tended to decrease mean arterial pressure (−15%, 95% CI: −23 to −1%). Ang II induced effects on cerebral, ocular and systemic haemodynamics were prevented by preceding administration of losartan.

Conclusions The present data suggest that Ang II is not a major determinant of cerebral and ocular blood flow *in vivo*. The observed changes in cerebral and ocular haemodynamic parameters after losartan administration reflect effects on systemic blood pressure.

Keywords: Doppler sonography, fundus pulsation, losartan, angiotensin II, cerebral blood flow, ocular blood flow

Introduction

The renin-angiotensin system (RAS) plays a pivotal role in the regulation of vascular tone. Inhibition of the renin-angiotensin system by angiotensin converting enzyme (ACE) inhibitors has been successfully used in the treatment of hypertension and congestive heart failure [1, 2]. However, ACE is not very specific and has substrates other than Ang I including bradykinin, substance P and neurokinins, which may be responsible for some of the side effects of ACE inhibitors [3]. The development of specific, selective type 1 receptor (AT₁) antagonists has therefore attracted much interest and its blood pressure lowering efficacy in patients with hypertension is investigated in several clinical trials [3, 4].

The role of angiotensin II (Ang II) in the cerebral and ocular circulation is still a matter of debate. Both vasoconstriction and vasodilation have been reported to occur in response to Ang II in the cerebral microcirculation [5, 6]. Local ACE activity has been identified in ocular tissues [7] and there exist specific binding sites for Ang II in the retinal and optic nerve vessels [8]. However, *in vitro* studies suggest

that Ang II does not play an important role in the regulation of ocular blood flow [9, 10].

The involvement of the angiotensin renin system in the regulation of cerebral and ocular blood flow *in vivo* in humans is even less well understood. Results with ACE inhibitors are difficult to interpret, because of the lack of specificity of this class of drugs [11, 12]. We have previously investigated the effect of intravenous infusion of Ang II on cerebral and ocular haemodynamics in healthy subjects [13] and observed dose-dependent changes in haemodynamic parameters in the middle cerebral artery, the ophthalmic artery, and the choroid. However, careful interpretation of these results argues that these changes were mainly caused by systemic haemodynamic effects [13]. The introduction of AT₁ receptor antagonists enables a specific investigation of the role of endogenous Ang II in these vascular beds. In an effort to elucidate the role of Ang II in the regulation of cerebral and ocular perfusion, we investigated the ocular and cerebral haemodynamic effects of losartan using Doppler ultrasound in the middle cerebral [14] and the ophthalmic artery [15] and laser interferometric measurement of fundus pulsation [16, 17].

Methods

Subjects

After approval from the Ethics Committee of Vienna University School of Medicine was obtained, 10 healthy male volunteers were studied (age range: 22–30 years, mean $\pm$ s.d.: 24.8 ± 2.4 years). The nature of the study was explained and all subjects gave written consent to participate. Each subject passed a screening examination that included medical history and physical examination, 12-lead electrocardiogram, complete blood count, activated partial thromboplastin time, thrombin time, fibrinogen, clinical chemistry (sodium, potassium, creatinine, uric acid, glucose, cholesterol, triglycerides, alanine aminotransferase, aspartate aminotransferase, γ -glutamyltransferase, alkaline phosphatase, total bilirubin, total protein), hepatitis A, B, C and HIV-serology, urine analysis, and a urine drug-screen. Subjects were excluded if any abnormality was found as part of the screening unless the investigators considered an abnormality clinically irrelevant. Furthermore an ophthalmic examination, including slit lamp biomicroscopy and indirect funduscopy was performed. Inclusion criteria were normal ophthalmic findings and a refractive error of greater than 3 dioptres in either of both eyes.

Study design

Subjects were studied in a placebo-controlled, randomized, double-blind two way crossover design with washout periods of 7 to 10 days between study days. The study schedule is given in Figure 1. On both trial days subjects arrived after an overnight fast. After steady state conditions were reached, which was ensured by repeated blood pressure measurements during the resting period, haemodynamic baseline measurements were taken. Following an intravenous infusion of

Ang II ($5 \text{ ng kg}^{-1} \text{ min}^{-1}$ over 30 min, Clinalfa AG, Läufelfingen, Switzerland) and a subsequent resting period, a single oral dose of 100 mg losartan (MSD, West Point, PA, USA) or placebo was administered. Placebo tablets were identical in appearance and taste to maintain double-blind conditions. Haemodynamic measurements were repeated at the scheduled time points after drug administration (Figure 1). In addition, three 30 min infusion periods of Ang II ($5 \text{ ng kg}^{-1} \text{ min}^{-1}$) were performed to study the effect of losartan on Ang II-induced haemodynamic changes. These infusion periods were scheduled 180, 300, and 420 min after losartan or placebo, to investigate the time course of losartan effects on Ang II-induced haemodynamic changes. Measurements were performed in a predetermined order (sonography of the middle cerebral artery, fundus pulsation measurement, blood pressure and pulse rate, sonography of the ophthalmic artery). Subjects remained in a sitting position until the end of the study period.

Study methods

Blood pressure and pulse rate: Systolic, diastolic and mean blood pressures (SBP, DBP, MAP) were measured on the upper arm by an automated oscillometric device (HP-CMS patient monitor, Hewlett Packard, Palo Alto, CA, USA). Pulse rate was automatically recorded from a finger pulse-oxymetric device (HP-CMS patient monitor).

Fundus pulsations Pulse synchronous pulsations of the eye fundus were assessed by laser interferometry on the subject's right eye. The method is described in detail by Schmetterer *et al.* [16]. Briefly, the eye is illuminated by the beam of a single mode laser diode with a wavelength (λ) of 783 nm. The light is reflected at both the front side of the cornea and the retina. The two re-emitted waves produce interference fringes from which the distance changes between

Figure 1 Time schedule of the study. Hollow bars indicate time points of haemodynamic measurements. Chequered bars indicate time points of haemodynamic measurements during Ang II-infusion. Numbers in italics indicate the time points of drug administration.

cornea and retina during a cardiac cycle can be calculated. Distance changes between cornea and retina lead to a corresponding variation of the interference order ($\Delta N(t)$). This change in interference order can be evaluated by counting the fringes moving inwards and outwards during the cardiac cycle. Changes in optical distance ($\Delta L(t)$), corresponding to the cornea-retina distance changes, can then be calculated by $\Delta L(t) = \Delta N(t) \cdot \lambda / 2$. The maximum distance change is called the fundus pulsation amplitude (FPA) and estimates the local pulsatile blood flow [17]. FPA was calculated as the mean of at least five cardiac cycles. The short-term and day-to-day variability of the method is small, which allows detection of even small changes in local pulsatile blood flow following pharmacological stimulation [18]. In contrast to systems recording ocular pressure pulse [19, 20], information on the ocular circulation can be obtained with high transverse resolution. To obtain information on the choroidal blood flow, the macula, where the retina lacks vasculature, was chosen for measurements [21, 22].

Doppler sonography

Mean blood flow velocity (MFV), peak systolic flow velocity (PSV), and end diastolic flow velocity (EDV) were determined in the right middle cerebral artery (MCA) with transcranial ultrasound [14] and in the right ophthalmic artery (OA) with colour Doppler ultrasound [15]. MFV was measured manually as time mean of the spectral outline. Measurements in the MCA were performed with a 2 MHz probe, measurements in the OA were performed with a 7.5 MHz probe (CFM 750, Vingmed Sound, Horten, Norway). The OA was measured anteriorly, at the point where it crosses the optic nerve. The sample volume marker was placed approximately 25 mm posterior to the globe. The resistive index in the MCA and the OA was calculated as $RI = (PSV - EDV) / (PSV)$. All parameters were determined as mean values over at least three cardiac cycles.

Data analysis

In analogy to the RI, which is calculated $(PSV - EDV) / PSV$, $(SBP - DBP) / SBP$ was calculated [13]. The pulsatile fraction of blood flow in the ophthalmic artery was calculated as $(MFV - EDV) / MFV$.

All statistical analyses were done using the Statistica® software package (Release 4.5, StatSoft Inc., Tulsa, OK, USA). The effect of losartan on haemodynamic parameters was assessed by repeated measure ANOVA *vs* placebo and *vs* baseline. *Post hoc* comparison was done with *t*-tests at individual time points using Bonferroni's correction. In addition, the effect of losartan on Ang II induced changes in haemodynamic parameters was assessed by repeated measure ANOVA using the data of Ang II infusion periods. The effect of Ang II alone was assessed by paired *t*-tests between values at baseline and during Ang II infusion preceding the administration of losartan or placebo. For data description, values were expressed as percent of baseline and are presented as means $\pm$ s.e.mean. A two-tailed $P < 0.05$ was considered the level of significance.

Table 1 Baseline haemodynamic parameters of the 2 study days. Data are presented as means $\pm$ s.e.mean ($n = 9$)

	Day 1	Day 2
FPA (μm)	3.8 ± 0.4	3.9 ± 0.4
RI (OA)	0.81 ± 0.01	0.79 ± 0.02
MFV (OA) (cm s^{-1})	18.9 ± 1.3	19.4 ± 1.4
RI (MCA)	0.62 ± 0.01	0.64 ± 0.02
MFV (MCA) (cm s^{-1})	49.4 ± 5.2	48.0 ± 4.1
MAP (mm Hg)	80 ± 3	85 ± 3
Pulse rate (beats min^{-1})	70 ± 4	69 ± 4

Results

One subject receiving losartan was withdrawn from the study after onset of headache during Ang II infusion on the first trial day and was not included in the statistical analysis. Nine subjects were therefore eligible for analysis of both trial days. There were no significant differences between baseline values on study day 1 and study day 2 (Table 1).

Haemodynamic effects of losartan

Losartan significantly increased FPA (maximum effect + 11%, 95% CI: 5 to 16%; Figure 2) *vs* baseline ($P = 0.0003$) and *vs* placebo ($P < 0.0001$). As evidenced from *post hoc* comparisons this effect maintained during the whole study period

Figure 2 Effects of losartan (▲) or placebo (▽) on resistive index and mean flow velocity in the ophthalmic artery (RI(OA) and MFV(OA)) and fundus pulsation amplitude (FPA). Data are presented as means $\pm$ s.e.mean ($n = 9$). Asterisks indicate significant changes *vs* baseline.

($P < 0.05$ for all time points). In addition, an increase in RI (OA) was observed (maximum effect +12%, 95% CI: 0 to 23%), which was significantly different *vs* baseline ($P = 0.022$), but not *vs* placebo. This increase in RI also led to an increase in pulsatile fraction of blood flow in the ophthalmic artery ($P = 0.045$ *vs* baseline, NS *vs* placebo, Table 2). These effects were long lasting and maintained during the observation period. MFV in the OA was not affected by losartan. In contrast to the OA, losartan did not exert any significant effects in the MCA.

Losartan reduced mean arterial pressure (maximum effect -15%, 95% CI: -23 to -1%, $P = 0.003$ *vs* baseline, NS *vs* placebo, Figure 3). This was paralleled by an increase in (SBP-DBP)/(SBP) as shown in Table 2 ($P = 0.019$ *vs* baseline, NS *vs* placebo). Pulse rate was not affected by administration of losartan (data not shown).

Haemodynamic effects of Ang II

The haemodynamic changes induced by Ang II infusion before losartan or placebo were comparable between the 2 study days. Ang II significantly reduced FPA by -15% *vs* baseline ($P = 0.006$, Figure 4). In the OA a significant decrease in RI (-6%, $P = 0.019$) and a significant increase in MFV was observed (+13%, $P = 0.01$). In addition, Ang II decreased the pulsatile fraction of ophthalmic artery blood flow ($P = 0.034$, Table 3). As in the OA there was a significant reduction of RI in the MCA (-13%, $P = 0.004$, Figure 4). The effect on MFV in the MCA (+7%), however, was not significant.

As expected, Ang II caused a significant increase in MAP (+13%, $P = 0.005$). Ang II also exerted a significant decrease in (SBP-DBP)/(SBP) as presented in Table 3 ($P = 0.014$). Pulse rate was reduced by -7% during Ang II infusion (NS).

Haemodynamic effects of losartan on Ang II induced changes

Losartan completely prevented the effect of Ang II on FPA ($P < 0.0001$, Figure 4). In addition, the reduction in resistive indices in the ophthalmic artery was significantly blunted by administration of losartan ($P = 0.005$). In the ophthalmic artery losartan also prevented the increase in mean flow velocity as induced by Ang II ($P = 0.011$). The response to

Table 2 The effect of losartan or placebo on (SBP-DBP)/SBP and on the pulsatile fraction of blood flow in the ophthalmic artery (MFV-EDV)/MFV. Data are presented as means $\pm$ s.e.mean ($n = 9$)

	(SBP-DBP)/SBP		(MFV-EDV)/MFV	
	Losartan	Placebo	Losartan	Placebo
Baseline	0.48 $\pm$ 0.03	0.51 $\pm$ 0.01	0.47 $\pm$ 0.03	0.49 $\pm$ 0.04
85 min	0.52 $\pm$ 0.03	0.50 $\pm$ 0.01	0.50 $\pm$ 0.05	0.49 $\pm$ 0.03
145 min	0.54 $\pm$ 0.02	0.49 $\pm$ 0.02	0.52 $\pm$ 0.04	0.45 $\pm$ 0.04
205 min	0.55 $\pm$ 0.02	0.52 $\pm$ 0.02	0.52 $\pm$ 0.03	0.48 $\pm$ 0.03
265 min	0.52 $\pm$ 0.02	0.49 $\pm$ 0.02	0.50 $\pm$ 0.04	0.47 $\pm$ 0.02
325 min	0.53 $\pm$ 0.02	0.48 $\pm$ 0.02	0.51 $\pm$ 0.03	0.48 $\pm$ 0.03
385 min	0.53 $\pm$ 0.02	0.52 $\pm$ 0.02	0.51 $\pm$ 0.05	0.47 $\pm$ 0.03
445 min	0.53 $\pm$ 0.02	0.48 $\pm$ 0.02	0.49 $\pm$ 0.03	0.49 $\pm$ 0.04
505 min	0.52 $\pm$ 0.02	0.51 $\pm$ 0.03	0.53 $\pm$ 0.04	0.48 $\pm$ 0.03

Figure 3 Effects of losartan (▲) or placebo (▽) on resistive index and mean flow velocity in the middle cerebral artery (RI(MCA) and MFV(MCA)) and mean arterial pressure (MAP). Data are presented as means $\pm$ s.e.mean ($n = 9$). Asterisks indicate significant changes *vs* baseline.

Ang II in RI in the middle cerebral artery was also blunted by losartan ($P = 0.005$, Figure 5).

Losartan also blunted the response in MAP ($P = 0.008$, Figure 5) and (SBP-DBP)/(SBP) almost returned to baseline levels ($P = 0.044$). Again, the effects on pulse rate were small.

Discussion

In the present study losartan tended to increase the resistive index in the ophthalmic artery and reversed the Ang II induced decrease in resistive index in the OA and MCA. The RI was introduced as a measure of vascular resistance distal to the investigated artery [23]. Hence one might interpret our data as a paradoxical increase in vascular resistance during administration of losartan. However, changes in proximal resistance can also alter RI [24] and we have previously shown that the Ang II induced changes in RI can be attributed to changes in systemic blood pressure [13]. Our calculations of (SBP-DBP)/(SBP) suggest that the Ang II and losartan induced changes in RIs are paralleled by changes in the pulse pressure amplitude. Our results in the OA and the MCA are more likely to reflect the propagation of the pressure wave along the vascular system, a fact, which has been discussed in more detail in a previous paper [13].

The Ang II induced increase in MFV is also difficult to

Figure 4 Effects of losartan (▨) or placebo (▧) on Ang II induced changes in resistive index and mean flow velocity in the ophthalmic artery (RI(OA) and MFV(OA)) and fundus pulsation amplitude (FPA). Data are presented as means $\pm$ s.e.mean ($n=9$). Asterisks indicate significant changes *vs* placebo.

Figure 5 Effects of losartan (▨) or placebo (▧) on Ang II induced changes in resistive index and mean flow velocity in the middle cerebral artery (RI(MCA) and MFV(MCA)) and mean arterial pressure (MAP). Data are presented as means $\pm$ s.e.mean ($n=9$). Asterisks indicate significant changes *vs* placebo.

Table 3 The effect of losartan or placebo at baseline and with concomitant infusion of Ang II on (SBP-DBP)/SBP and on the pulsatile fraction of blood flow in the ophthalmic artery (MFV-EDV)/MFV. Data are presented as means $\pm$ s.e.mean ($n=9$)

	(SBP-DBP)/SBP		(MFV-EDV)/MFV	
	Losartan	Placebo	Losartan	Placebo
Baseline	0.48 $\pm$ 0.03	0.51 $\pm$ 0.01	0.47 $\pm$ 0.03	0.49 $\pm$ 0.04
15 min (+ Ang II)	0.43 $\pm$ 0.02	0.43 $\pm$ 0.02	0.40 $\pm$ 0.03	0.41 $\pm$ 0.03
235 min (+ Ang II)	0.52 $\pm$ 0.02	0.44 $\pm$ 0.02	0.46 $\pm$ 0.04	0.42 $\pm$ 0.04
355 min (+ Ang II)	0.53 $\pm$ 0.02	0.45 $\pm$ 0.02	0.49 $\pm$ 0.04	0.40 $\pm$ 0.03
475 min (+ Ang II)	0.49 $\pm$ 0.04	0.42 $\pm$ 0.02	0.48 $\pm$ 0.03	0.44 $\pm$ 0.04

interpret. An increase in MFV can either be caused by an increase in blood flow through the vessel or by a decrease in vessel diameter. As we did not measure vessel diameter we cannot decide which of these effects predominates. The interpretation of these data is further hampered since Ang II and losartan caused significant changes in blood pressure and therefore may have provoked autoregulatory mechanisms in the vascular beds under study [25].

In addition, we performed laser interferometric measurement of fundus pulsation, a method which is suitable for

the detection of even small drug-induced effects on pulsatile ocular blood flow [17, 18, 22]. However, it must be kept in mind that the non-pulsatile blood flow component cannot be assessed with this method. There is an ongoing debate concerning the amount of the pulsatile component of blood flow in the eye. The estimates vary between 80% [20] and 50% [26]. Therefore interpretation of the results requires knowledge of drug induced changes in flow pulsatility. In the ophthalmic artery supplying the choroid we observed an increased flow pulsatility after the administration of

losartan. Hence the increase in FPA after losartan administration does not necessarily reflect an increase in choroidal blood flow, but may rather be attributable to a change from non-pulsatile to pulsatile blood flow.

A limitation of our study is that we did not measure intraocular pressure (IOP). It is well known that systemic administration of ACE-inhibitors slightly reduces IOP [27], whereas intravenous Ang II increases IOP [28]. A change in IOP leads to a change in ocular perfusion pressure because the choroidal venous pressure almost equals the IOP [29]. However, as compared with the losartan- and Ang II induced changes in MAP this effect on IOP is certainly small.

The present study provides further evidence that losartan blocks the pressor effects of Ang II *in vivo* [30, 31]. This effect is long-lasting, because losartan is partially converted into the more active metabolite EXP3174 [32], which has a much longer half-life time than losartan itself [33, 34]. This makes losartan an excellent tool to study the RAS *in vivo*. For the investigation of the pharmacodynamic effect of losartan we assumed that the inserted Ang II infusion periods did not influence the succeeding haemodynamic measurements. This assumption is based on the observation that systemic, cerebral, and ocular haemodynamic parameters return to baseline within 5 min after the end of Ang II infusion (unpublished observation). As we have observed a very stable effect of losartan on the parameters under study (Figures 1 and 2) this assumption seems justified.

The results of the present study confirm our previous results that the RAS does not play an important role in the regulation of cerebral and ocular blood flow in healthy subjects [13]. This, however, does not mean that the RAS is not involved in the pathophysiology of cerebrovascular disease. There is evidence from a large number of *in vitro* and *in vivo* studies, that the RAS has a detrimental impact on cardiovascular remodelling and atherogenesis of the cerebrovascular bed [35]. Moreover, there is an association between Ang II and thrombotic stroke and animal experiments suggest that losartan prevents cerebrovascular infarcts [36].

It must be emphasised that Ang II is also assumed to play a role in cerebral myogenic autoregulation [25]. We did not investigate this phenomenon in the present study, although myogenic autoregulation may also be present in the choroidal vasculature [37]. Hence angiotensin receptor blockade may be useful to prevent abnormal cerebral and ocular autoregulation in patients with activated RAS, such as in hypertension. Animal experiments, however, indicate that AT₂ receptors rather than AT₁ receptors are involved in cerebral blood flow autoregulation [38].

In conclusion the data of the present study indicate that the RAS does not play a major role in the regulation of cerebral and ocular blood flow in healthy subjects. Systemic rather than local effects appear responsible for the effects of losartan in the ophthalmic artery and the choroid. Moreover, losartan prevented the systemic and regional effects of Ang II in all vascular beds under study.

Financial support from a grant from the Österreichische Nationalbank (Jubiläumsfonds No. 6346) and a MSD medical school grant is acknowledged.

References

- 1 Brunner HR, Nussberger J, Waeber B. Effects of angiotensin converting enzyme inhibition: a clinical point of view. *J Cardiovasc Pharmacol* 1985; **7**(suppl. 4): 73–81.
- 2 Consensus Trial Study Group. Effects of enalapril on mortality in severe congestive heart failure: results of the Cooperative North Scandinavian Enalapril Survival Study (CONSENSUS). *N Engl J Med* 1987; **316**: 1429–1435.
- 3 Johnston CI. Angiotensin receptor antagonists: focus on losartan. *Lancet* 1995; **346**: 1403–1407.
- 4 Timmermans PB, Wong PC, Chiu AT, Smith RD. The preclinical basis of the therapeutic evaluation of losartan. *J Hypertension* 1995; **13**(suppl. 1): S1–S13.
- 5 Edvinsson L, Hardebo JE, Owan C. Effects of angiotensin II on cerebral blood vessels. *Acta Physiol Scand* 1979; **105**: 381–383.
- 6 Haberl RL. Role of angiotensin receptor subtypes in the response of rabbit brain arterioles to angiotensin. *Stroke* 1994; **25**: 1476–1480.
- 7 Igic R, Kojovic V. Angiotensin I converting enzyme (kinase II) in ocular tissues. *Exp Eye Res* 1980; **30**: 299–303.
- 8 Ferrari-Dileo G, Davis EB, Anderson DR. Angiotensin II binding receptors in retinal and optic nerve head blood vessels. *Invest Ophthalmol Vis Sci* 1991; **32**: 21–26.
- 9 Berg Nyborg NC, Nielsen PJ, Prieto D, Benedito S. Angiotensin II does not contract bovine retinal resistance arteries *in vivo*. *Exp Eye Res* 1990; **50**: 469–474.
- 10 Berg Nyborg NC, Nielsen PJ. Angiotensin II contracts isolated human posterior ciliary arteries. *Invest Ophthalmol Vis Sci* 1990; **31**: 2471–2473.
- 11 Demolis P, Chalon S, Annane D, Duhaze P, Guidicelli JF. Effects of an angiotensin-converting enzyme inhibitor, ramipril, on intracranial circulation in healthy volunteers. *Br J Clin Pharmacol* 1992; **34**: 224–230.
- 12 Demolis P, Carville C, Guidicelli JF. Effects of angiotensin-converting enzyme inhibitor, lisinopril, on cerebral blood flow autoregulation in healthy volunteers. *J Cardiovasc Pharmacol* 1993; **22**: 373–380.
- 13 Krejcy K, Wolzt M, Kreuzer C, Breiteneder H, Schütz W, Eichler HG, Schmetterer L. Characterisation of angiotensin-II effects on cerebral and ocular circulation by noninvasive methods. *Br J Clin Pharmacol* 1997; **43**: 501–508.
- 14 Aaslid R, Markwalder TM, Nornes H. Noninvasive transcranial Doppler ultrasound recording of flow velocity in basal cerebral arteries. *J Neurosurg* 1982; **57**: 769–774.
- 15 Guthoff RE, Berger RW, Winkler P, Helmke K, Chumbley LC. Doppler ultrasonography of the ophthalmic and central retinal vessels. *Arch Ophthalmol* 1991; **109**: 532–536.
- 16 Schmetterer L, Lexer F, Unfried C, Sattmann H, Fercher A. Topical measurement of fundus pulsations. *Opt Eng* 1995; **34**: 711–716.
- 17 Schmetterer L, Wolzt M, Salomon A, et al. Effect of isoproterenol, phenylephrine, and sodium nitroprusside on fundus pulsations in healthy volunteers. *Br J Ophthalmol* 1996; **80**: 217–223.
- 18 Schmetterer L, Strenn K, Findl O, et al. Effects of antiglaucoma drugs on ocular hemodynamics in healthy volunteers. *Clin Pharmacol Ther* 1997; **61**: 583–595.
- 19 Langham ME, Tomey K. A clinical procedure for the measurement of the ocular pulse-pressure relationship and the ophthalmic arterial pressure. *Exp Eye Res* 1978; **27**: 17–25.
- 20 Langham ME, Farrell RA, O'Brien V, Silver DM, Schilder P. Blood flow in the human eye. *Acta Ophthalmol* 1989; **67**(Suppl. 191): 9–13.
- 21 Schmetterer L, Wolzt M, Lexer F, et al. The effect of

- hyperoxia and hypercapnia on fundus pulsations in the macular and optic disc region in healthy young volunteers. *Exp Eye Res* 1995; **61**: 685–690.
- 22 Schmetterer L, Lexer F, Graselli U, Findl O, Eichler HG, Wolzt M. The effect of different mixtures of O₂ and CO₂ on ocular fundus pulsations. *Exp Eye Res* 1996; **63**: 351–355.
 - 23 Pourcelot L. Diagnostic ultrasound for cerebral vascular diseases; In *Present and future of diagnostic ultrasound*. Eds. Donald I, Levi S Rotterdam, The Netherlands: Kooyker 1976: 141–147.
 - 24 Hayreh SS, Beach KW. Optic nerve head sheath decompression, comment. *Ophthalmology* 1993; **100**: 303–305.
 - 25 Strandgaard S, Paulsen OB. Regulation of cerebral blood flow in health and disease. *J Cardiovasc Pharmacol* 1992; **19**(Suppl. 6): S89–93.
 - 26 Krakau CET. A model for pulsatile and steady ocular blood flow. *Graefes Arch Clin Exp Ophthalmol* 1995; **233**: 112–118.
 - 27 Costagliola C, Di Benedetto R, De Caprio L, Verde R, Mastropasqua L. Effect of oral captopril on intraocular pressure in man. *Eur J Ophthalmol* 1995; **5**: 19–25.
 - 28 Karznez TA, Murphy MB, Weber RR, Nelson KS, Tripathi BJ, Tripathi RC. Effects of selective dopamine-1 receptor activation on intraocular pressure in man. *Exp Eye Res* 1988; **47**: 689–697.
 - 29 Maepea O. Pressures in the anterior ciliary arteries, choroidal veins and choriocapillaris. *Exp Eye Res* 1992; **54**: 731–736.
 - 30 Christen Y, Waeber B, Nussberger J, *et al*. Oral administration of DuP 753, a specific angiotensin II receptor antagonist, to normal volunteers. *Circulation* 1991; **83**: 1333–1342.
 - 31 Goldberg MR, de Mey C, Wroblewski JM, Li Q, Schroeter V, Belz GG. Differential effects of losartan and enalapril on local venous and systemic pressor responses to angiotensin I and II in healthy men. *Clin Pharmacol Ther* 1995; **59**: 72–82.
 - 32 Munafo A, Christen Y, Nussberger J, *et al*. Drug concentration–response relationship in normal volunteers after oral administration of losartan, an angiotensin II receptor antagonist. *Clin Pharmacol Ther* 1992; **51**: 513–521.
 - 33 Ohtawa M, Takayama F, Saitoh K, Yoshinaga T, Nakashima M. Pharmacokinetics and biochemical efficacy after single and multiple oral administration of losartan, an orally active nonpeptide angiotensin II receptor antagonist, in humans. *Br J Clin Pharmacol* 1993; **35**: 290–297.
 - 34 Lo MW, Goldberg MR, McCrea JB, Lu H, Furtek CI, Bjornsson TD. Pharmacokinetics of losartan, an angiotensin II receptor antagonist, and its active receptor metabolite EXP3174 in humans. *Clin Pharmacol Ther* 1995; **58**: 641–649.
 - 35 Rossi G, Rossi A, Sacchetto A, Pavan E, Pessina A. Hypertensive cerebrovascular disease and the renin-angiotensin system. *Stroke* 1995; **26**: 1700–1706.
 - 36 Fornes P, Richter C, Vacher E, Bruneval P, Giudicelli JF. Losartan's protective effects in stroke-prone spontaneously hypertensive rats persist durably after treatment withdrawal. *J Cardiovasc Pharmacol* 1993; **22**: 305–313.
 - 37 Kiel JW. Choroidal myogenic autoregulation and intraocular pressure. *Exp Eye Res* 1994; **58**: 529–544.
 - 38 Näveri L, Strömberg C, Saavedra JM. Angiotensin II AT₂ receptor stimulation extends the upper limit of cerebral blood flow autoregulation: agonist effects of CGP 42112 and PD 123319. *J Cereb Blood Flow Metab* 1994; **14**: 38–44.

(Received 21 February 1997,
accepted 10 June 1997)