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No effect of angiotensin II AT₂-receptor antagonist PD 123319 on cerebral blood flow autoregulation

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Abstract

Blockade of the renin-angiotensin system with angiotensin-converting enzyme inhibitors (ACE-I) or angiotensin AT₁-receptor antagonists shift the limits of autoregulation of cerebral blood flow (CBF) towards lower blood pressure (BP). The role of AT₂-receptors in the regulation of the cerebral circulation is uncertain. Hence, the present study investigated the effect on CBF autoregulation of blocking of angiotensin AT₂-receptors with PD 123319 in spontaneously hypertensive rats (SHR). Anaesthetised and ventilated SHR were given PD 123319, 0.36 mg/kg/min, intravenously, and compared with a control group. CBF was measured by the intracarotid ¹³³xenon injection method and BP was raised by noradrenaline infusion and lowered by controlled haemorrhage in separate groups of rats. The limits of autoregulation were determined by computed least-sum-of-squares analysis. PD 123319 did not influence baseline CBF, but resulted in a minor BP decrease (10 control and 10 treated rats). The lower limit of CBF autoregulation (eight treated and eight control) as well as the upper limit of CBF autoregulation (eight treated and eight control) were not significantly different in PD 123319 and control animals (lower limit treated 102±4 mmHg and control 94±4; NS, and upper limit treated 171±10 mmHg and control 162±7; NS). These findings indicate that acute AT₂-receptor blockade does not influence CBF autoregulation.

Introduction

Cerebral blood flow (CBF) is autoregulated, i.e. is kept constant within a wide range of perfusion pressure. Autoregulation of CBF is mediated mainly by calibre changes in the smaller cerebral resistance vessels, which constrict when perfusion pressure increases, and dilate when perfusion pressure decreases. Beyond the upper limit of autoregulation, the arteries and arterioles are unable to constrict further, and the CBF rises, eventually resulting in forceful vasodilation, damage of the blood-brain barrier, cerebral oedema formation and risk of cerebral haemorrhage. Below the lower limit of CBF autoregulation, the arteries and arterioles will be submaximally dilated and as the perfusion pressure falls, CBF will fall, resulting in cerebral ischaemia, with risk of irreversible damage.¹

The renin-angiotensin system (RAS) modulates CBF autoregulation. Angiotensin-converting enzyme inhibitors (ACE-I) and AT₁-receptor blockers (ARBs) shift both the upper and lower limits of

autoregulation towards lower blood pressure (BP) and shorten the autoregulatory plateau.^{2,3} It has been suggested that selective blockade of the AT₁-receptor in large cerebral arteries causes dilation of these large cerebral arteries with a compensatory autoregulatory constriction further downstream in small arteries and arterioles.³

The function of the AT₂-receptor is poorly understood. It is less widely distributed in the body than the AT₁-receptor. The pressor effect of angiotensin II (Ang II) is mediated by the AT₁-receptor, and the role of the AT₂-receptor in BP regulation is still unclear.⁴ All of the well-known effects of Ang II, such as aldosterone, vasopressin and oxytocin release, negative feed-back on renin release, and renal salt and water retention are mediated by the AT₁-receptor.⁵ Previous studies have failed to clarify the function of the AT₂-receptor in the cerebral circulation.^{6,7}

The aim of the present work was to investigate whether blockade of this receptor influences autoregulation of CBF. The study was undertaken in spontaneously hypertensive rats (SHR) using the selective AT₂ antagonist, PD 123319.⁸

Methods

The study was carried out in 52 male SHR, aged around three months and weighing between 260 and 350 g. The rats were supplied by Møllegaarden, Lille Skensved, Denmark. They had unlimited access to food and water prior to the study.

Surgery

Anaesthesia was induced with 4% halothane in a mixture of 30% O₂ and 70% N₂O. After tracheotomy, anaesthesia was maintained with 0.8% halothane in 30% O₂ and 70% N₂O, by controlled ventilation at normocapnia. Rats were paralysed with suxamethonium i.p. (10 mg/kg bolus and 3.5 mg/kg/hour), in order to prevent interference from spontaneous respiration with regard to the stability of the arterial carbon dioxide tension (PaCO₂). A rectal thermostat-controlled heating table maintained body temperature at around 37°C. Both femoral veins were cannulated (pp50 catheter, Holm & Halby, Denmark) for drug and donor blood administration. Both femoral arteries were cannulated (pp50 catheter, Holm & Halby, Denmark), one for continuous BP measurement with a transducer (Simonsen & Weel, Denmark), and one for blood gas sampling. PaCO₂, PaO₂ and pH were measured, using an arterial blood gas

analyser (ABL 500 and 605, Radiometer, Denmark). PaCO₂ was kept constant (between 37 and 41 mmHg) during the experiment by adjusting the ventilation volume. For CBF measurement, the intra-arterial ¹³³Xe technique was used.⁹ The scalp and the temporal muscle over the right hemisphere were removed. The right external carotid artery was exposed on the right side near the carotid bifurcation. Extracerebral branches, including the pterygopalatine artery, were ligated, in order to minimise extracerebral distribution of ¹³³Xe.¹⁰ Rats were then heparinised (3300 IU/kg), and a fine catheter (pp25, Holm & Halby, Denmark) was introduced retrogradely into the external carotid artery, with the tip at the bifurcation, for ¹³³Xe administration. After surgery, anaesthesia was maintained with 0.6% halothane in 30% O₂ and 70% N₂O, and the rat was left to stabilise for 30 minutes, before any CBF measurement was made.⁹

Cerebral blood flow measurement

For each CBF measurement a bolus of 30–70 µl ¹³³Xe,¹⁰ 3.5 mCi/ml (370 MBq/ml) was injected into the right internal carotid artery. A collimated sodium iodide crystal placed over the skull, ipsilateral to the injection site, measured the clearance of ¹³³Xe from the brain during the first 20 seconds after the injection. CBF was then determined from the initial slope of the washout curve.^{9,11} The correction for background activity and any activity remaining from previous ¹³³Xe injections was slightly different, as we recorded the data directly on a computer allowing for a mathematical correction. The correction was the background recorded before any ¹³³Xe plus the remaining ¹³³Xe recorded immediately before the new injection and assumed to decay with a constant time constant during the short period of recording. At the end of each measurement MABP (mmHg), CBF (ml/100 g/min), PaCO₂ (mmHg), PaO₂ (mmHg), arterial pH and rectal temperature (°C) were recorded. Three baseline measurements were made before drug administration and manipulation of the BP, to confirm baseline stability. The values were averaged for the determination of baseline CBF level in each rat.

PD 123319 administration

PD 123319 (Parke-Davis, Michigan, USA) dissolved in saline, 0.36 mg/kg/min, was administered by intravenous infusion (0.56 ml/hour). PD 123319, at doses of 0.36 and 1 mg/kg/min, has been shown not to affect baseline CBF.^{6,7} The lowest dose was chosen in order not to interfere with AT₁-receptors.

Time course of the effect of PD 123319

PD 123319 was administered to 10 SHR. Ten rats received vehicle (saline) as control. CBF was measured at baseline and at 2, 5 and 10 minutes during drug administration. Thereafter, CBF was measured at 15 minute intervals for 120 minutes.

Autoregulation study

The lower limit of CBF autoregulation was studied in 16 SHR. Eight animals received PD 123319, while

eight served as controls. PD 123319 or saline was administered intravenously, and the BP was allowed to stabilise for 10 minutes after the injection and prior to the commencement of the autoregulation study. Haemorrhagic hypotension was subsequently induced by withdrawing blood into a syringe. By this means, BP was reduced stepwise to the lowest obtainable level. Throughout the study, CBF was measured at 10 to 15 mmHg BP intervals.

The upper limit of CBF autoregulation was studied in 16 SHR. Eight received PD 123319 and eight served as controls. BP was increased gradually to the highest obtainable level by the intravenous infusion of norepinephrine (NE), 0.025–0.5 µg/min, and CBF was measured at 10 to 15 mmHg BP intervals.

Investigation of the lower and upper limits of CBF autoregulation were made in separate groups of rats, since rats are no longer in a normal physiological condition when their BP has been brought to the extremes outside the limits of autoregulation.

Calculation of the autoregulation curve

The lower and upper limits of CBF autoregulation in each individual rat were defined as the mean arterial blood pressure (MABP) value of the intersection of two lines: one slope regression line for all points from 1 to *i*, and one horizontal line for the point *i* + 1 to *N* (all measurements ranked in order of increasing BP). The horizontal line had to cross the slope line between the slope MABP values at *i* and *i* + 1, but was otherwise chosen as, or as close as possible to, the mean of the points *i* + 1 to *N*. Going through all data points (*i* = 2 to *N* – 2), the best fit for the autoregulation curve was then defined by the least-sum-of-squares method of the combined line.¹² Autoregulation was defined as preserved when the total sum of squares for the best fit was smaller than that of one straight regression line through all data points. The upper limit of autoregulation was calculated using the horizontal line corresponding to the data point with lower BP values and the slope corresponding to those with higher BPs. Otherwise, calculations were performed as for the lower limit.

Data analysis

The Wilcoxon rank sum test was used for intergroup comparisons of the baseline values from the time-course study and the autoregulation study. Analysis of variance (ANOVA) was used for statistical comparison of the time-course groups (control versus PD 123319). The Wilcoxon rank sum test was used for statistical comparison of the effect of time in the time-course group. Values are expressed as mean ± SD. The limits of autoregulation were evaluated by pooling all control and PD 123319 animals separately and applying a SE estimate of the curves, according to a *t*-statistics test, testing the intersection of two different lines.¹² Values are expressed as mean ± SEM. *p* < 0.05 was considered statistically significant.

Results

Baseline values in the different groups are shown in

Table 1 Baseline values.

Group	MABP (mmHg)	CBF (ml/100 g/min)	PaCO ₂ (mmHg)	PaO ₂ (mmHg)	pH	Temperature (°C)
Time course, control	143±12	104±14	39.0±0.5	117±4	7.43±0.01	37.6±0.3
Time course, PD 123319	141±8	98±14	39.1±0.4	111±2	7.46±0.0**	37.0±0.2*
Lower limit, control	129±10	108±10	39.0±0.6	124±2	7.44±0.01	37.4±0.4
Lower limit, PD 123319	144±10	99±8	39.2±0.8	111±4	7.45±0.01*	37.2±0.2
Upper limit, control	140±10	112±12	38.9±0.6	115±2	7.45±0.0	37.1±0.3
Upper limit, PD 123319	152±10	104±10	38.9±0.4	112±3	7.45±0.0	37.2±0.2

Values are expressed as means ± SD. (n=8 per group, except time course groups, for which n=10).

* p<0.05; ** p<0.01 when compared with control. MABP = mean arterial blood pressure. CBF = cerebral blood flow.

Figure 1 (A) The time course study. Mean arterial blood pressure measured at regular intervals, in control (▲) and PD 123319 (●) treated SHR rats. (B) The time course study. Cerebral blood flow measured at regular intervals, in control (▲) and PD 123319 (●) treated SHR rats. Cerebral blood flow values are given as percentages of baseline values.

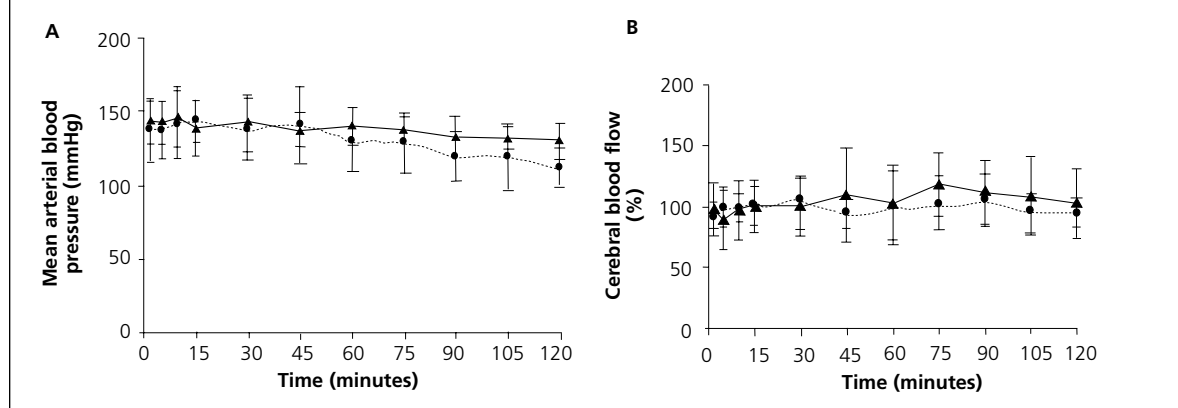


Table 1. There were no significant differences in any of the baseline parameters between the groups ($p>0.05$), except for pH ($p<0.01$) and temperature ($p<0.05$) in the time-course groups and pH ($p<0.05$) in the lower limit groups. These alterations were very small, and not likely to influence the results of the study. PaCO₂, PaO₂, pH and temperature were constant and within the normal range during the study, except for slight alterations caused by extreme hypotension and hypertension induced at the end of the experiment.

Time-course study

Effect of PD 123319 on mean arterial blood pressure

Administration of PD 123319, 0.36 mg/kg/min, caused a significant decrease in MABP in the time-course study. During the first 10 minutes, there was no reduction in MABP, (138±21 to 141±23 mmHg, $p>0.05$). During the rest of the study, there was a significant reduction in MABP, (143±14 to 112±13 mmHg, $p<0.001$). The overall reduction was from 138±21 to 112±13 mmHg ($p<0.01$). The BP-lowering effect commenced approximately one hour after the start of the drug infusion

(Figure 1A). The difference in BP between the control and PD 123319-treated groups was significant only for the last measurement (time = 120 minutes, Figure 1A).

Effect of PD 123319 on cerebral blood flow

There was no effect on CBF during the time-course study, (92±11 to 94±11 ml/100 g/min, $p>0.05$). (Figure 1B)

Lower limit of cerebral blood flow autoregulation

CBF autoregulation was preserved in both PD 123319-treated and control groups. The lower limit of CBF autoregulation was 94±3.8 in the control group and 102±3.9 mmHg in the PD 123319-treated group (NS).

Upper limit of cerebral blood flow autoregulation

CBF autoregulation was preserved in both PD 123319-treated and control groups. The upper limit of CBF autoregulation was 162±7.1 in the control group and 171±10.2 mmHg in the PD 123319-treated group (NS).

Figure 2 The pooled autoregulation curve showing the lower and upper limit in (A) control group and (B) PD 123319-treated rats. Cerebral blood flow values are given as percentages of baseline values.

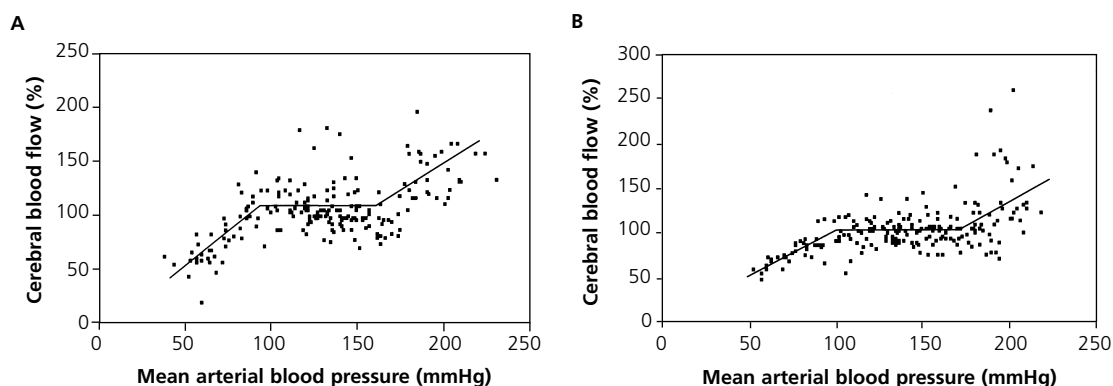


Table 2 Blockade of the renin-angiotensin system and autoregulation of the cerebral blood flow. Comparison of studies from our laboratory performed using the same method.

Study	Drug	Action	Shift lower limit	Shift upper limit
Barry <i>et al.</i> 1984	Captopril, 10 mg/kg	ACE inhibition	WKY – 20 mmHg, no p-value SHR – 20 mmHg, no p-value	WKY – 60 mmHg, no p-value SHR – 60 mmHg, no p-value
Torup <i>et al.</i> 1993	Ceranapril, 1 mg/kg	ACE inhibition	WKY – 10 mmHg, $p < 0.05$ SHR – 15 mmHg, $p < 0.01$	WKY – 23 mmHg, $p < 0.05$ SHR – 15 mmHg, $p < 0.01$
Vraamark <i>et al.</i> 1995	Candesartan 0.1 mg/kg	AT ₁ inhibition	WKY – 12 mmHg, $p < 0.01$ SHR – 7 mmHg, $p < 0.05$	WKY – 18 mmHg, $p < 0.05$ SHR – 30 mmHg, $p < 0.01$
Present study	PD 123319 0.36 mg/kg/min	AT ₂ inhibition	SHR + 8 mmHg, NS	SHR + 9 mmHg, NS

In Figure 2, normalised CBF values from the studies of the lower and upper limits have been combined to show the whole autoregulation curve in control (2A) and PD 123319-treated (2B) animals.

Comparison with earlier studies from our group

In Table 2, results from studies made by our group with the ACE-I, captopril and ceranapril, and the ARB, candesartan, are shown.^{3,13,14} ACE inhibition and AT₁-receptor blockade, in contrast to AT₂-receptor blockade, shifts the limits of CBF autoregulation to lower pressure limits.

Discussion

The main observation of the present study was that PD 123319 had no effect on the limits of autoregulation. Furthermore, PD 123319 significantly lowered MABP in the time-course study, without influencing CBF.

It is unlikely that the methodology of the study would have obscured any effect of PD 123319 on CBF autoregulation. The intra-arterial ¹³³Xe injection method for CBF measurement in the rat, as used in this study, allows repetitive measurements at short intervals, and is thus well suited to the study of cerebrovascular reactivity.⁹ The method

requires universal anaesthesia, which might influence CBF. In the present study, the rats were anaesthetised with halothane and paralysed with suxamethonium. Both of these agents have ganglion-blocking properties, but any effect on CBF would be present in both control and in AT₁-receptor antagonist studies, and thus are unlikely to influence the comparisons between the groups. Anaesthesia with halothane causes a decrease in MABP, without an increase in plasma renin activity, but again any effect on CBF would be present in both groups.¹⁵ Norepinephrine has no effect on CBF.¹⁶ Haemorrhagic hypotension leads to α -adrenergic vasoconstriction, which shifts the autoregulatory curve towards higher BP.¹⁷ This effect, blunted by halothane and suxamethonium, would be present in both control and PD 123319-treated groups.

Ang II receptors in large cerebral arteries in rats have been suggested to be of the AT₂-receptor subtype.¹⁸ However, the AT₁-receptor subtype is also expressed, at least in the middle cerebral artery.¹⁹ Ang II-mediated vasodilation has been found in rat brain arterioles^{20,21} and in dog middle cerebral arteries.²² Ang II-mediated vasoconstriction has been reported in cat middle cerebral artery strips.²³ This dual effect of Ang II suggests stimulation of two types of Ang II receptors in the

cerebral circulation, with AT₁-receptors mediating vasoconstriction and AT₂-receptors mediating vasodilation.

ACE-I shift the limits of CBF autoregulation towards lower BP.² In a previous study from our group, the ARB CV-11974 (candesartan) was shown to have a similar effect,³ whereas the present study shows that AT₂-receptor blockade has no effect on CBF autoregulation (Table 2). Studies by other groups have given somewhat contradictory results. Thus, Strömberg and co-workers showed that PD 123319 and losartan, an ARB, both shifted the upper limit of CBF autoregulation towards higher BP.^{6,7} They suggested that stimulation of the AT₂-receptor causes vasoconstriction, and that vasodilation is mediated through the AT₁-receptor. According to these studies, PD 123319 functions as an agonist at the AT₂-receptor, causing vasoconstriction, and losartan acts as an antagonist at the AT₁-receptor, also causing vasoconstriction, resulting in a shift of the upper limit of autoregulation towards higher BP.^{6,7} In a previous report, Strömberg and co-workers found that losartan shifted the upper limit of CBF autoregulation towards lower BP, which is consistent with a vasoconstrictory role of the AT₁-receptor.²⁴ These earlier results of Strömberg and co-workers are in agreement with Vraamark and co-workers.³ Another study in rats revealed that blockade of AT₁-receptors, in contrast to blockade of AT₂-receptors, abolished the blood flow reduction resulting from intracarotid Ang II infusion.²⁵ The BP-lowering effect of PD 123319, as demonstrated in the time-course study, is difficult to explain. Although the PD 123319 is not absolutely specific for AT₂-receptors, it is unlikely that it had any major effect on AT₁-receptors.

The absence of a significant cerebrovascular response to PD 123319, as observed in the present study, could be due to the nature of the AT₂-receptor in the SHR strain. Endo and co-workers recently showed that Ang II caused more marked vasoconstriction in SHR kidney afferent arterioles than in WKY afferent arterioles, possibly due to an impaired modulator function of the AT₂-receptor in SHR afferent arterioles.²⁶ SHR was chosen in the present study because it is a model of human essential hypertension. Carried out in WKY rats, a more pronounced effect on CBF autoregulation might have been seen with a shift of the limits of autoregulation towards even higher BP.

In conclusion, the present study showed no significant effect of the AT₂-receptor on CBF autoregulation in SHR.

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References

1. Paulson OB, Strandgaard S, Edvinsson L. Cerebral autoregulation. *Cerebrovasc Brain Metab Rev* 1990;2:161-92.
2. Paulson OB, Waldemar G. ACE inhibitors and cerebral blood flow. *J Hum Hypertens* 1990;4(suppl 4):69-73.
3. Vraamark T, Waldemar G, Strandgaard S, Paulson OB. Angiotensin II receptor antagonist CV-11974 and cerebral blood flow autoregulation. *J Hypertens* 1995;13:755-61.
4. Stroth U, Unger T. The renin-angiotensin system and its receptors. *J Cardiovasc Pharmacol* 1999;33(suppl 1):S21-S28.
5. Timmermans PBMW, Smith RD. The diversified role of angiotensin II-receptor blockade. *Blood Pressure* 1996;5(suppl 2):53-61.
6. Strömberg C, Näveri L, Saavedra JM. Nonpeptide angiotensin AT₁ and AT₂ receptor ligands modulate the upper limit of cerebral blood flow autoregulation in rats. *J Cereb Blood Flow Metab* 1993;13:298-303.
7. Näveri L, Strömberg C, Saavedra JM. Angiotensin II AT₂ receptor stimulation extends the upper limit of cerebral blood flow autoregulation: Agonist effects of GCP42112 and PD123319. *J Cereb Blood Flow Metab* 1994;14:38-44.
8. Brechler V, Jones PW, Levens NR, de Gasparo M, Bottari SP. Agonistic and antagonistic properties of angiotensin analogs at the AT₂ receptor in PC12W cells. *Regulatory Peptides* 1993;44:207-13.
9. Hertz MM, Hemmingsen R, Bolwig TG. Rapid and repetitive measurements of blood flow and oxygen consumption in the rat brain using intraarterial Xenon injection. *Acta Physiol Scand* 1977;101:501-03.
10. Hertz MM, Bolwig TG. Blood-brain barrier studies in the rat: an indicator dilution technique with tracer sodium as an internal standard for estimation of extracerebral contamination. *Brain Research* 1976;107:333-43.
11. Olesen J, Paulson OB, Lassen NA. Regional cerebral blood flow in man determined by the initial slope of the clearance of intra-arterially injected ¹³³Xe. *Stroke* 1971;2:519-40.
12. Schmidt JF, Waldemar G, Vorstrup S, Andersen AR, Gjerris F, Paulson OB. Computerized analysis of cerebral blood flow autoregulation in humans: validation of a method for pharmacologic studies. *J Cardiovasc Pharmacol* 1990;15:983-8.
13. Barry DI, Jarden JO, Paulson OB, Graham DI, Strandgaard S. Cerebrovascular aspects of converting-enzyme inhibition I: effects of intravenous Captopril in spontaneously hypertensive and normotensive rats. *J Hypertension* 1984;2:589-97.
14. Torup M, Waldemar G, Paulson OB. Ceranapril and cerebral blood flow autoregulation. *J Hypertension* 1993;11:399-405.
15. Miller ED. Renin with anesthesia and surgery. In: *The Renin-Angiotensin System*. Edited by Robertson I, Nicholls MG. London: Gower; 1993:81.1-81.7.
16. Olesen J. The effect of intracarotid epinephrine, norepinephrine and angiotensin on the regional blood flow in man. *Neurology* 1972;22:978-87.
17. Fitch W, MacKenzie ET, Harper AM. Effects of decreasing arterial blood pressure on cerebral blood flow in the baboon. *Circ Res* 1975;37:550-7.
18. Tsutsumi K, Saavedra JM. Characterization of the AT₂ angiotensin II receptors in rat anterior cerebral arteries. *Am J Physiol* 1991;261:H667-70.
19. Nishimura Y, Takeshi I and Saavedra JM. Angiotensin II AT₁ blockade normalizes cerebrovascular autoregulation and reduces cerebral ischemia in spontaneously hypertensive rats. *Stroke* 2000;31:2478.
20. Haberl RL, Anneser F, Villringer A, Einhäupl KM. Angiotensin II induces endothelium-dependent vasodilation of rat cerebral arterioles. *Am J Physiol* 1990;258:H1840-H1846.
21. Brix J, Haberl RL. The AT₂ receptor mediates endothelium-dependent dilation of rat brain arterioles [abstract]. *FASEB J* 1992;6:A1264.
22. Toda N. Hemolysate inhibits cerebral artery relaxation. *J Cereb Blood Flow and Metabol* 1988;8:46-53.
23. Edvinsson L, Hardebo JE, Owman C. Effect of angiotensin II on cerebral blood vessels. *Acta Physiol Scand* 1979;105:381-3.
24. Strömberg C, Näveri L, Saavedra JM. Regulation of rat cerebral blood flow by angiotensin II [Abstract]. *FASEB J* 1992;6:A1011.
25. Kramar EA, Harding JW, Wright JW. Angiotensin II- and IV-induced changes in cerebral blood flow. Roles of AT₁, AT₂, and AT₄ receptor subtypes. *Regulatory Peptides* 1997;68:131-8.
26. Endo Y, Arima S, Yaoita H, Tsunoda K, Omata K, Ito S. Vasodilation mediated by angiotensin II type 2 receptor is impaired in afferent arterioles of young spontaneously hypertensive rats. *J Vascular Research* 1998;35:421-7.