Chronic alpha-1-adrenergic blockade increases sympathoneural but not adrenomedullary activity in patients with essential hypertension

Marie-Cécile Jacobs, Jacques W.M. Lenders, Jacques J. Willemsen* and Theo Thien

Objective: Doxazosin, a selective \(\alpha_1\)-adrenoceptor antagonist, lowers blood pressure by reducing peripheral vascular resistance without causing reflex tachycardia. To discover whether antihypertensive treatment with an \(\alpha_1\)-adrenoceptor blocker is accompanied by an increase in sympathoadrenomedullary activity, we studied plasma catecholamine kinetics before and during treatment with doxazosin.

Patients and methods: Eleven patients with essential hypertension were studied before and after 3 months' treatment with doxazosin (4–8 mg a day). \(^3\)H-noradrenaline and \(^3\)H-adrenaline were infused simultaneously and blood samples were collected to calculate plasma catecholamine kinetics before and during sympatho-adrenomedullary stimulation (lower-body negative pressure).

Results: Doxazosin decreased systolic and diastolic blood pressure and forearm vascular resistance, whereas the heart rate did not change significantly. During doxazosin, baseline arterial plasma noradrenaline increased from 0.97±0.07 to 1.21±0.07 nmol/l, and this appeared to be due to an increase in total body noradrenaline spillover from 1.54±0.15 to 1.84±0.16 nmol/min; noradrenaline clearance did not change significantly. Forearm noradrenaline spillover also increased, from 0.89±0.18 to 1.48±0.23 pmol/100ml per min. In contrast, arterial plasma adrenaline, total body adrenaline spillover and adrenaline clearance were not significantly affected by doxazosin treatment. The response of plasma noradrenaline and total body and forearm spillover of noradrenaline to lower-body negative pressure (−40 mmHg) was significantly increased during doxazosin administration, whereas the response of the adrenaline kinetic parameters were not altered.

Conclusions: The blood pressure reduction induced by a chronic administration of the \(\alpha_1\)-adrenoceptor blocker doxazosin elicits a baroreceptor-mediated reflexive increase in sympathoneural but not in adrenomedullary activity. The latter finding might partly explain why the heart rate is not increased during chronic treatment with this \(\alpha_1\)-adrenoceptor blocking drug.


Keywords: Doxazosin, catecholamines, \(\alpha_1\)-adrenoceptor, hypertension

Introduction

Doxazosin is a selective \(\alpha_1\)-adrenoceptor blocking drug with a slow onset of action; it lowers blood pressure by decreasing systemic vascular resistance [1,2]. In contrast to the acute administration of \(\alpha_1\)-adrenoceptor blockers, chronic administration in hypertensive patients does not elicit a reflexive increase in the heart rate. This suggests that baroreflex-mediated sympathetic activity is not increased during chronic treatment with doxazosin. The lack of reflex tachycardia has also been attributed to the absence of an interruption of the presynaptic inhibitory effect by noradrenaline upon neuronal noradrenaline release [3]. Central \(\alpha_1\)-adrenoceptor antagonism has

From the Department of Internal Medicine, Division of General Internal Medicine, and the *Laboratory of Experimental and Chemical Endocrinology, St Radboud University Hospital, Nijmegen, the Netherlands.

Sponsorship: This study was supported by the Dutch Heart Association (grant no 89.212)

Requests for reprints to Dr J.W. M. Lenders, Department of Medicine, Division of General Internal Medicine, St Radboud University Hospital, Geert Groteplein Zuid 8, 6525 GA Nijmegen, the Netherlands.

© Rapid Science Publishers ISSN 0263-6352
been suggested as an alternative mechanism for the absence of reflex tachycardia [4]. However, treatment with prazosin, a similar α1-adrenoceptor blocker to doxazosin but with a rapid onset of action, induces an increase in plasma levels of noradrenaline [5–7]. Although it has been assumed that the increased plasma noradrenaline levels are caused by an increase in baroreflex-mediated sympathetic activity, no definitive explanation is available for the increase in plasma noradrenaline levels. Elevated plasma noradrenaline levels might be due to a reflexive increase in sympatheuoneal release of noradrenaline or to a reduced clearance of noradrenaline.

The main objective of the present study was to assess the effects of a chronic blood pressure reduction induced by the α1-adrenoceptor blocker doxazosin on the baroreflex-mediated activity of the sympatho-adrenomedullary system. For this purpose, we used the isotope dilution method with a steady-state infusion of tritiated catecholamines [8]. Before and after chronic doxazosin treatment (3 months), spillover and clearance of noradrenaline and adrenaline were assessed in essential hypertensives and this was examined before and during stimulation of the sympatho-adrenomedullary system by two different intensities of lower-body negative pressure.

Subjects and methods

Subjects
Eleven subjects with essential hypertension (mean ± SD age 35.8 ± 2.3 years) participated in the study. Before entry all participants had a normal physical examination and none were found to suffer from cardiovascular or other diseases. Secondary hypertension was excluded according to standard clinical criteria, and all subjects had a normal renal function. The mean ± SD Quetelet index was 23.6 ± 0.7 kg/m². Antihypertensive treatment was according to standard clinical criteria, and all subjects had a normal renal function. The mean ± SD Quetelet index was 23.6 ± 0.7 kg/m². Antihypertensive treatment was withdrawn at least 4 weeks before the study; after it was withdrawn, blood pressure was measured three times at 2-week intervals. The mean ± SD basal systolic/diastolic blood pressure before treatment with doxazosin was 147 ± 3.95/2.2 mmHg and the heart rate was 77 ± 3 beats/min. All subjects gave written informed consent and the study protocol was approved by the Hospital Ethics Committee.

Study protocol
All patients were studied twice: before and after treatment with doxazosin for 3 months. Each patient started with doxazosin at 2 mg once a day and blood pressure was monitored every 3 weeks. The dose was increased every 3 weeks by 2 mg to a maximum of 8 mg a day unless diastolic blood pressure had fallen by at least 25% on the current dose. Patients with a sufficient blood pressure fall remained on the titrated dose until the end of the 3-month period. After 3 months, two patients were taking 4 mg a day, one was taking 6 mg a day and nine were taking 8 mg doxazosin a day.

On each study day, the subjects were allowed a light breakfast. All were required to abstain from alcohol, nicotine and caffeinated foods and beverages for at least 24 h before each study day. All studies were carried out in the morning in a room with constant temperature. During the study the subjects remained supine in a lower-body negative pressure box that was used to stimulate sympatho-adrenomedullary activity. After instrumentation, radiotracer infusions (see below) were started and the subjects rested for 30 min. During the last 3 min, baseline recordings of blood pressure, heart rate and nine forearm blood flow curves were obtained. Then arterial and venous blood samples were drawn simultaneously to determine plasma concentrations of endogenous and tritiated catecholamines.

Thereafter, lower-body negative pressure was applied at 15 mmHg for 15 min. Blood pressure, heart rate and forearm blood flow recordings and blood samples were collected in sequence beginning after 12 min of lower-body negative pressure. A rest period of 30 min ensued and then another 15 min of lower-body negative pressure at 40 mmHg was applied, and blood pressure, heart rate and forearm blood flow recordings and blood samples were obtained as before.

Procedures
A brachial artery was cannulated to monitor blood pressure and the heart rate (Hewlett-Packard GmbH, Böblingen, Germany) and to draw arterial blood samples. An intravenous catheter was inserted into a deep brachial vein in the ipsilateral arm for collecting venous blood samples. A forearm venous catheter in the contralateral arm was used for simultaneous infusion of 3H-noradrenaline and 3H-adrenaline. Forearm blood flow was recorded by venous occlusion strain-gauge plethysmography with air-filled cuffs [9]; during this measurement and while blood samples were drawn, the hand circulation was excluded by inflating a wrist cuff to 100 mmHg above systolic blood pressure [10].

To assess catecholamine kinetics, 3H-noradrenaline (L-[O-2,5,6-3H]-noradrenaline) and 3H-adrenaline (L-[N-methyl-3H]-adrenaline), with high specific activity, were infused intravenously. Tritiated catecholamines were obtained from DuPont New England Nuclear (Hertogenbosch, the Netherlands), sterilized using a micropore filter (0.22 μm) and diluted in 0.9% NaCl containing acetic (0.2 mol/l) and ascorbic (1 mg/ml) acid. The aliquots were stored until used at –80°C for a maximum of 3 months. Sterilization, dilution and aliquoting were carried out under nitrogen. Just before use, an aliquot of each radiotracer was diluted in 0.9% NaCl.

After a bolus injection of each tracer at 15 μCi/m²2, both tracers were infused continuously for 90 min at a rate of 0.35 μCi/m²2 per min. The weights of the two syringes containing the radiotracers were measured before and
after the infusion to verify the infusion rate. Samples of the infusate were taken at the end of the infusion and stored at -80°C until assayed.

**Analytical methods**

Blood samples were collected in prechilled tubes containing ethyleneglycol-bis-(β-aminoethylether)-N,N',N'-tetraacetic acid (0.25 mol/l) and glutathione (0.2 mol/l). The blood samples were placed on melting ice. Plasma was separated by refrigerated centrifugation and frozen until assayed within 2 months of collection. Plasma samples were analysed for concentrations of noradrenaline, adrenaline, $^3$H-noradrenaline and $^3$H-adrenaline, using high-performance liquid chromatography with fluorimetric detection after selective precolumn derivatization of the catecholamines with the fluorescent agent 1,2-diphenylethylene diamine [11]. Using a Gilson fraction collector, model 201-202, (Gilson Medical Electronics, Villiers le Bel, France) connected to an automatic sample injector, Wisp 710B, (Waters Associates, Milford, Massachusetts, USA), we collected $^3$H-noradrenaline and $^3$H-adrenaline into scintillation vials for 1 min, starting at the beginning of the peaks of noradrenaline and adrenaline in the standard mixture.

**Data analysis**

Forearm vascular resistance was calculated by dividing mean arterial blood pressure by forearm blood flow and was expressed in arbitrary units (AU). The average of the haemodynamic data during 3 min of recording was calculated.

The clearance of noradrenaline from arterial plasma was calculated by dividing the rate of $^3$H-noradrenaline infusion by the steady-state arterial plasma concentration of $^3$H-noradrenaline. Total body noradrenaline spillover, the estimated rate of appearance of endogenous noradrenaline in arterial plasma, was calculated by multiplying the steady-state arterial plasma noradrenaline concentration by the clearance. Analogously, noradrenaline spillover in the forearm ($S$, pmol/min per 100 ml), was estimated as:

$$S = PN_A f + [P(N_V - N_A)]$$

where $P$ is forearm plasma flow, $N_A$ is arterial plasma noradrenaline, $N_V$ is venous plasma noradrenaline and:

$$f = (3H-N_A - 3H-N_V)/3H-N_A$$

The function $f$ represents the fractional extraction of the tracer in the forearm. The forearm plasma flow, in units of ml/min/100 ml, was calculated from the forearm blood flow and haematocrit. The clearance of noradrenaline in the forearm (ml/min per 100 ml) was calculated by multiplying the forearm plasma flow by the function $f$.

The clearance of adrenaline from arterial plasma and the estimated rate of appearance of endogenous adrenaline into arterial plasma were calculated according to similar formulas.

Results are expressed as means±SEM unless indicated otherwise. To test the effects of doxazosin on baseline plasma kinetic variables, the Wilcoxon signed rank test was used. This test was also used to test for the effects of lower-body negative pressure on each variable. $P<0.05$ (two-sided) was considered significant.

**Results**

**Haemodynamic data**

Doxazosin decreased systolic blood pressure from 147±3 to 135±4 mmHg and diastolic pressure from 95±2 to 85±3 mmHg ($P<0.05$). The heart rate decreased slightly, from 77±3 to 70±2 beats/min, but this was not significant. Forearm vascular resistance decreased significantly ($P<0.05$), from 89±7 to 64±6 AU.

Doxazosin had no effect on the blood pressure and heart rate response to a lower-body negative pressure of −15 mmHg but the increase in forearm vascular resistance during lower-body negative pressure at −15 mmHg was smaller after doxazosin treatment (+7±2 AU) than before (+31±8 AU, $P<0.05$). The blood pressure and heart rate responses to lower-body negative pressure at −40 mmHg were also unaffected by doxazosin, but the increase in forearm vascular resistance was again significantly smaller after doxazosin treatment (+5±2 AU) than before (+32±9 AU, $P<0.05$).

**Plasma noradrenaline kinetics**

After treatment with doxazosin, all subjects had an approximately 20% higher plasma noradrenaline level than before (Table 1). This increase in arterial plasma noradrenaline appeared to be due to an increase in total body noradrenaline spillover of about 20% (Fig. 1), since total body clearance of noradrenaline was not significantly altered (Table 1). Forearm noradrenaline spillover was also significantly increased after doxazosin treatment (Fig. 1) while the local clearance of noradrenaline by the forearm was not significantly affected (Table 1).

The changes in plasma noradrenaline, total body and forearm noradrenaline spillover and total body and forearm noradrenaline clearance in response to lower-body negative pressure (−15 mmHg) were not affected by doxazosin. However, with −40 mmHg lower-body negative pressure, the increase in plasma noradrenaline was significantly larger after doxazosin (+1.33±0.11 nmol/l) than before (+0.88±0.10 nmol/l; $P<0.05$). Total body and forearm noradrenaline spillover increases were also significantly higher during −40 mmHg lower-body negative pressure after doxazosin treatment than before (Fig. 2). The decreases in total body noradrenaline clearance during lower-body negative pressure at −40 mmHg were not significantly affected by doxazosin and the same applied to the reductions in forearm noradrenaline clearance (Fig. 2).
Table 1. Plasma levels and kinetics of noradrenaline and adrenaline before and after treatment with doxazosin.

<table>
<thead>
<tr>
<th></th>
<th>Before doxazosin</th>
<th>After doxazosin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Noradrenaline</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Arterial plasma level (nmol/l)</td>
<td>0.97 ±0.07</td>
<td>1.21 ±0.07*</td>
</tr>
<tr>
<td>Total body spillover (nmol/l)</td>
<td>1.54 ±0.15</td>
<td>1.84 ±0.16*</td>
</tr>
<tr>
<td>Total clearance (l/min)</td>
<td>1.57 ±0.10</td>
<td>1.52 ±0.09</td>
</tr>
<tr>
<td>Forearm spillover (pmol/100 ml per min)</td>
<td>0.89 ±0.18</td>
<td>1.48 ±0.23*</td>
</tr>
<tr>
<td>Forearm clearance (ml/100 ml per min)</td>
<td>0.58 ±0.06</td>
<td>0.82 ±0.10</td>
</tr>
<tr>
<td>Adrenaline</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Arterial plasma level (nmol/l)</td>
<td>0.21 ±0.03</td>
<td>0.15 ±0.02</td>
</tr>
<tr>
<td>Total body spillover (nmol/l)</td>
<td>0.42 ±0.07</td>
<td>0.27 ±0.04</td>
</tr>
<tr>
<td>Total clearance (l/min)</td>
<td>1.94 ±0.10</td>
<td>1.81 ±0.11</td>
</tr>
<tr>
<td>Forearm clearance (ml/100 ml per min)</td>
<td>0.70 ±0.07</td>
<td>0.89 ±0.12</td>
</tr>
</tbody>
</table>

*P<0.05 versus before doxazosin.

Plasma adrenaline kinetics
After the administration of doxazosin, basal arterial plasma adrenaline was lower than before doxazosin treatment but the difference did not reach significance (Table 1). Total body adrenaline spillover was also lower after doxazosin but, again, the decrease was not significant. Total body clearance of adrenaline was not affected by doxazosin (Table 1).

The changes in adrenaline kinetics in response to lower-body negative pressure of -15 and -40 mmHg were not significantly altered by doxazosin.

Discussion
The main new finding of the present study is that long-term antihypertensive treatment with the α₁-adrenoceptor blocker doxazosin elicits a selective increase in sympathetic but not adrenomedullary activity. This was demonstrated by the increase in plasma noradrenaline and total body and forearm noradrenaline spillover with no concurrent increase in plasma levels and total body spillover of adrenaline.

Chronic antihypertensive treatment with the α₁-adrenoceptor blocker doxazosin resulted in a significant decrease in blood pressure and forearm vascular resistance. The heart rate did not increase, as shown previously for prazosin. The following three explanations for the unchanged heart rate have been put forward: vasodilation through α₁-adrenoceptor blockade does not induce a reflexive increase in sympathetic activity because of resetting of the arterial baroreceptor reflex; blockade of central α₁-adrenoceptors [4]; selective α₁-adrenoceptor blocking drugs preserve the normal presynaptic α₂-adrenoceptor-mediated auto-inhibition of neuronal noradrenaline release by noradrenaline [3]. However, in the present study plasma noradrenaline levels were increased, as were total body and forearm noradrenaline spillover. This is in accord with previous studies [5–7] on prazosin. An increase in the spillover of noradrenaline into the blood compartment can be caused by an increase in sympathetic nerve traffic, by a decrease in neuronal uptake of noradrenaline or by decreased inhibition of noradrenaline release through presynaptic α₂-adrenoceptors. The last mechanism is unlikely to have occurred in the present study, since the affinity of doxazosin for α₂-adrenoceptors is about 400 times lower than that for α₁-adrenoceptors [12]. Alternatively, presynaptic α₁-adrenoceptors might be considered a possibility; however, although present in some species [13,14], no presynaptic α₁-adrenoceptors are functionally present in human blood vessels. Thus, an effect of doxazosin on presynaptic α₂-adrenoceptors or on eventual presynaptic α₁-adrenoceptors has to be rejected as a possible explanation for the increase in noradrenaline spillover. The most likely explanation for the increase in noradrenaline spillover, both systemically...
and in the forearm vascular bed, is that there was a reflexive increase in sympathetic nerve traffic due to the blood pressure reduction. This argues strongly against a resetting of the arterial baroreceptor reflex during chronic treatment with doxazosin.

If there is, indeed, an increase in baroreflex-mediated sympathoneural activity during chronic $\alpha_1$-adrenoceptor blockade, then why is there no increase in the heart rate? Since cardiac $\beta_1$-adrenoceptors are equally sensitive to noradrenaline as to adrenaline in vitro, the increased noradrenaline spillover may be expected to elicit an increase in the heart rate. However, in vivo, noradrenaline usually induces a decrease in the heart rate because of the baroreflex-mediated increase in vagal nerve activity. An alternative explanation for the absence of an increase in the heart rate could be provided by the absence of an increase in plasma adrenaline and in total body noradrenaline spillover. Total body noradrenaline spillover reflects adrenomedullary secretion of adrenaline. Apparently, the blood pressure reduction induced by doxazosin does not stimulate adrenomedullary secretion of adrenaline but only the sympathoneuronal release of noradrenaline. This differentiated response to a reduction in blood pressure has also been described for other vasodilating antihypertensive drugs and is therefore not a specific effect of $\alpha_1$-adrenoceptor blockers [15].

During lower-body negative pressure at $-40\ mmHg$, both arterial and cardiopulmonary baroreceptors were de-activated and a marked increase in sympatho-adrenomedullary activity ensued, as illustrated by the increase in forearm vascular resistance, in plasma noradrenaline and forearm noradrenaline spillover and in total body noradrenaline and adrenaline spillover. During doxazosin treatment, the blood pressure and heart rate responses to lower-body negative pressure were not significantly altered but forearm vascular resistance response was lower than before doxazosin. This is in variance with a previous study with prazosin, demonstrating unaltered responses by peripheral vascular resistance to isometric exercise and exposure to cold [16].

Taken together, these data indicate that doxazosin is a very powerful $\alpha_1$-adrenoceptor antagonist that blocks postsynaptic $\alpha_1$-adrenoceptors adequately, even during strong sympathetic stimulation. During chronic treatment with doxazosin there is a clear increase in baroreflex-mediated sympathoneural activity whereas adrenomedullary activity is not increased. This latter finding might provide a partial explanation for the absence of an increase in the heart rate during chronic treatment with doxazosin.

References