Reduced medication and normalization of vascular structure, but continued hypertension in renovascular patients after revascularization

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Abstract

Objective: The effect of invasive treatment of renal artery stenosis on the use of antihypertensive medication, blood pressure, and morphology and function of resistance arteries was investigated in 14 renovascular hypertensive patients before and after treatment compared to normotensive controls. Methods: Use of antihypertensive medication was calculated as defined daily doses (DDD). Resistance arteries were taken from gluteal subcutaneous biopsies and analyzed in a myograph. Results: Prior to invasive treatment, blood pressure of the patients was elevated compared to normotensive controls. Six months after technically successful invasive treatment, patients were still hypertensive compared to time-matched controls. The use of antihypertensive medication was reduced from $4.4 \pm 0.7$ DDD before invasive treatment to $3.0 \pm 0.6$ DDD 6 months after treatment. Vascular structure of resistance arteries, expressed as media/lumen ratio (media thickness to diameter), was greater in patients before invasive treatment ($10.7 \pm 1.0\%$) compared to normotensive controls ($7.9 \pm 0.8\%$). Media/lumen ratio of resistance arteries was reduced to that of the controls 6 months after invasive treatment despite the remaining hypertension. The functional studies showed no difference in response to acetylcholine, adenosine, noradrenaline or angiotensin II between patients and controls before or after treatment. Conclusions: This study shows that hypertension and increased media/lumen ratio of resistance arteries prevail in renovascular hypertensive patients despite antihypertensive medication and that invasive treatment is of benefit as regards use of antihypertensive medication. The study provides the novel information that the remaining hypertension is not due to uncorrected media/lumen ratio of the resistance arteries. © 2001 Elsevier Science B.V. All rights reserved.

Keywords: Antihypertensive/diuretic agents; Arteries; Blood pressure; Hypertension; Remodeling

1. Introduction

In renovascular hypertension, it is often difficult to keep patients normotensive despite massive antihypertensive medication making the patients prone to the consequences and complications of hypertension [1,2]. This fact, together with the hypertension having an apparently known cause which in principal can be removed, makes it tempting to perform invasive treatment of the disease as an alternative to continuation of medical treatment. However, there is a great variation in the results of invasive treatment which can be explained by differences in types of indication for treatment, invasive procedures, parameters investigated and time of follow-up [3–10]. The immediate results of invasive treatment are often good as regards patency of the artery, lowering of blood pressure and reduction of medicine intake, but later follow-up often shows that these beneficial effects are not maintained [11].

One of the well-known consequences of hypertension is change of function and morphology of the resistance vessels [12–14]. There is much knowledge about these abnormalities in essential hypertension and about the influence of medical treatment on the vessels [13,15], but information about the same parameters in human re-

Time for primary review 27 days.
novascular hypertension is sparse [16]. Our aim was therefore to determine if invasive treatment was able to change these conditions in renovascular hypertensive patients as regards blood pressure and use of antihypertensive medication, and for the first time to assess the effect of invasive treatment on morphology and function of peripheral resistance arteries.

2. Methods

2.1. Subjects

Fourteen patients with renovascular hypertension and 14 normotensive controls were included in the study. Their basic characteristics are shown in Table 1. The following parameters were assessed at entry to the study and 6 months after invasive treatment: blood pressure, serum-creatinine, urine-albumin, antihypertensive medication, and function and morphology of resistance vessels. Blood pressure was measured with a mercury sphygmomanometer after at least 10 min rest with the subject in an upright position. Mean value of three measurements was used. The measurements were performed by a nurse unaware of the diagnosis or former blood pressure of the patients and controls. Urine-albumin was measured by Stix-method, Stix being positive when proteinuria exceeded 0.1 g/l. If Stix was positive, proteinuria was quantitated in g/l.

Serum-creatinine was determined and considered abnormal if above 125 μM.

2.2. Patients

Renovascular hypertension was suspected in hypertensive patients on the basis of routine renography showing differences between kidneys in clearance and mean transit time. Final diagnosis was established by intraarterial arteriography. To increase the likelihood of dealing with renovascular hypertension, and not just renovascular disease, renal vein renin ratio was measured in 11 patients. Renal vein renin ratio was calculated as the concentration of renin in the renal vein of the affected side/concentration of renin on the contralateral side. Renal vein renin ratio exceeded 1.5 in eight patients, in two patients the ratio was 1.46 and 1.48, respectively, and in one patient the ratio was unity, but captopril-renography showed marked differences in clearance and mean transit time compared to conventional renography. The plasma renin activity was not measured after removal of the stenoses, because the response of this parameter to removal of a renal artery stenosis is unpredictable [17]. All patients had unilateral stenoses. The mean degree of the stenosis was 75±3% (range 50–95%). Ten stenoses were localized on the left side and four on the right side. There were six smokers among the patients and five patients had symptoms of general atherosclerosis (ischaemia of the legs, angina, retinal thrombosis, atherosclerosis of aorta visualized by arteriography).

All patients received antihypertensive medication when they entered the study. All but one patient were being given calcium-antagonists, and all but one were receiving more than one type of antihypertensive medication. No patients were treated with ACE-inhibitors or angiotensin II-antagonists at entry to the study. The antihypertensive treatment was withdrawn on the day of invasive treatment and resumed afterwards when needed. The choice of antihypertensive treatment was at the discretion of the clinician, which for one patient included an angiotensin II-antagonist. The spectrum of drugs used before and after is shown in Section 3. The duration of antihypertensive treatment prior to entry was 45±15 months (range 6–168 months). Six patients had been treated with acetyl salicylic acid for various periods of time. The amount of antihypertensive medication was measured in defined daily doses (DDD) [18] at entry and 6 months after invasive treatment.

Percutaneous transluminal renal angioplasty (PTRA) was the preferred treatment and was the final treatment for six patients. Percutaneous transluminal renal angioplasty with stent (PTRS) was performed in one patient. In three patients, the renography showed that the kidney beyond the stenosis had less than 10% function left and nephrectomy was performed. Four patients were subjected to renal artery bypass grafting (RABG), two patients because of complications to PTRA and two patients because of simultaneous need for reconstructive surgery of abdominal aorta or femoral artery. The immediate effect of these treatments was judged successful in all patients. No patients were subjected to new invasive treatment procedures during the follow-up. Subcutaneous biopsies were taken from all patients just before invasive treatment and 6 months after.

In the two patients who were subjected to RABG because of complications to PTRA, the second biopsy was taken 6 months after RABG.

2.3. Controls

Controls were healthy volunteers, gender- and age-matched to the patients.

Table 1

<table>
<thead>
<tr>
<th>Patients</th>
<th>Controls</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender (female/male)</td>
<td>4/10</td>
</tr>
<tr>
<td>Blood pressure (mmHg)</td>
<td>175/99±6/3</td>
</tr>
<tr>
<td>Serum-creatinine (μM)</td>
<td>113±9</td>
</tr>
<tr>
<td>Age (years)</td>
<td>53±3</td>
</tr>
<tr>
<td>Proteinuria (+/−)³</td>
<td>6/8</td>
</tr>
</tbody>
</table>

Basal characteristics of the hypertensive patients and normotensive controls. The patients and controls were gender- and age-matched. * P<0.05. *** P<0.001. ³ Stix method.
Controls were normotensive and without any medication or known disease. Blood pressure and urine protein were measured as in patients, and subcutaneous biopsies were taken at the time when patients entered the study. In nine controls, a second biopsy was taken 6 months later at the same time as the second biopsy was taken from the patients.

No controls became hypertensive or received any medication during the study.

2.4. Preparation of subcutaneous resistance vessels

After application of local anaesthesia, a biopsy (1×2.5×0.5 cm) of subcutaneous fat was cut from a buttock with a sterile technique. The biopsy was placed in ice-cold physiological salt solution (see below for composition) and the following day two resistance arteries, 2 mm long, were dissected from the biopsy and mounted in a myograph [19]. In the myograph, vessels were kept at 37°C and bubbled with 5% CO₂ in O₂. After equilibration, morphology was measured in a microscope using an ocular micrometer to measure the different layers at six positions in each vessel. Mean media thickness and internal diameter were calculated and expressed as media/lumen ratio for the vessels being stretched to normalized diameter 1ₐ; 1ₐ was 0.9 times the diameter the vessels would have had when fully relaxed and exposed to a pressure of 100 mmHg [19].

Only vessels with an internal diameter, 1ₐ, of less than 350 μm and effective pressure (pressure against which vessels could contract=active wall tension/(1⁻½) [18]) to K-PSS (see below for composition) of minimum 8 kPa were accepted. Functional studies were performed with the vessels set to diameter 1ₐ. Functional studies were performed as concentration–response curves with noradrenaline (0.01–10 μmol/l), acetylcholine (0.01–100 μmol/l) and adenosine (0.001–1 mmol/l) added to the bath in half logarithmic steps. Acetylcholine and adenosine were added on top of a precontraction with 3 μmol/l noradrenaline for 10 min. Drugs were added every second minute. To check the capability of the vessels to maintain a stable precontraction during acetylcholine and adenosine relaxing curves, a time control curve was performed with 3 μmol/l noradrenaline. Relaxation was assessed with reference to this control curve. Concentration–response curves with angiotensin II (3–100 nmol/l) were performed in six patients and matched controls, while angiotensin II was applied as one concentration (100 nmol/l) in the remaining eight patients and controls.

Dissection of the vessels and experiments in the myograph were performed blindly to ensure that the laboratory technician was not aware whether the biopsy was from a patient before or after treatment or from a control.

2.5. Solutions

Biopsies were kept in physiological salt solution (PSS), which consisted of (mmol/l): NaCl 119, KCl 4.7, CaCl₂ 2.5, MgSO₄·7H₂O 1.17, NaHCO₃ 25, KH₂PO₄ 1.18, EDTA 0.026, glucose 5.5. In K-PSS, NaCl was replaced with 119 mmol/l KCl. The following drugs were used: (−)noradrenaline hydrochloride, adenosine and angiotensin II (Sigma Chemical Co., St. Louis, MO, USA) and acetylcholine (Fluka AG, Buchs SG, Switzerland).

2.6. Ethics

The investigation conforms with the principles outlined in the Declaration of Helsinki (Cardiovascular Research 1997;35:2–3). The protocol was also approved by the Local Scientific Ethical Committee, and all patients and controls gave their informed consent before entering the study.

2.7. Statistical analysis

All values are given as mean±S.E.M. Data were analyzed by Student’s two-tailed paired t-test and linear regression. P<0.05 was considered statistically significant. When more than one vessel was taken per subject, the mean value of each vascular parameter was used for the statistical analysis.

3. Results

3.1. Blood pressure

The patients were hypertensive despite antihypertensive medication when they entered the study (Table 1), and there was a positive correlation between systolic blood pressure and media/lumen ratio of the resistance arteries (r²=0.31, P<0.05). Six months after invasive treatment, blood pressure of the patients was 157/90±6/3 mmHg and blood pressure was still significantly higher than blood pressure of the normotensive controls, 129/81±4/3 mmHg (P<0.05, Fig. 1).

3.2. Use of antihypertensive medication

Before invasive treatment, patients were treated with antihypertensive medication with a DDD of 4.4±0.7 (range 1–8.3 DDD). Six months after invasive treatment DDD was reduced to 3.0±0.6 (P<0.05, range 0–6.2 DDD) including two patients who were normotensive without medication (Fig. 2). The spectrum of drugs used was similar before and after treatment (in DDD) beta-blocker: before 0.6±0.1, after 0.4±0.1; calcium antagonist: before 2.3±0.4, after 1.3±0.4; diuretic: before 1.1±0.3, after 0.8±0.3; other: before 0.5±0.3, after 0.6±0.4).
3.3. Morphological investigations

The media/lumen ratio of resistance arteries was 10.7±1.0% in hypertensive patients before invasive treatment compared to 7.9±0.8% in the normotensive controls (P<0.05). Six months after invasive treatment, media/lumen ratio of resistance arteries was reduced to the same level as the controls, 8.5±0.9 and 8.3±0.7%, respectively (Fig. 3). The reduction in blood pressure during treatment did not correlate with the reduction in media/lumen ratio (r²=0.09, P=0.28). There were no significant differences in internal diameter or media cross-sectional area of the vessels between patients and controls before or after invasive treatment (Table 2).

3.4. Functional investigations

The functional results are shown in Table 3. We found no differences in the maximal relaxation to acetylcholine and adenosine between patients and controls before or after invasive treatment. We found no differences in contractile response to noradrenaline, K-PSS or angiotensin II between patients and controls before or after invasive treatment expressed as wall tension, or (data not shown) as effective pressure. We found no differences in ED₅₀ of acetylcholine, adenosine or noradrenaline between patients and controls before or after invasive treatment.

3.5. Kidney function

Kidney function was evaluated on the basis of serum-creatinine and proteinuria. Mean serum-creatinine at the entry to the study is given in Table 1. Although these

<table>
<thead>
<tr>
<th>Table 2</th>
<th>Morphological measurements in human gluteal resistance arteries</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Before treatment</td>
</tr>
<tr>
<td>Patients</td>
<td>Controls</td>
</tr>
<tr>
<td>Media area (μm²)</td>
<td>16.0±2.2</td>
</tr>
<tr>
<td>Internal diameter (μm)</td>
<td>203±14</td>
</tr>
</tbody>
</table>

There were no differences in media area or internal diameter between patients and controls before or after invasive treatment. Values are mean±S.E.M.
Table 3
Responses of human gluteal subcutaneous resistance arteries to acetylcholine, adenosine, angiotensin II and noradrenaline

<table>
<thead>
<tr>
<th></th>
<th>Before treatment</th>
<th>After treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Patients</td>
<td>Controls</td>
</tr>
<tr>
<td><strong>ED&lt;sub&gt;50&lt;/sub&gt;</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>acetylcholine (−log, μM)</td>
<td>7.4±0.1</td>
<td>7.5±0.1</td>
</tr>
<tr>
<td>Max. relaxation,</td>
<td>0.53±0.09</td>
<td>0.59±0.09</td>
</tr>
<tr>
<td>acetylcholine ( % of max. response)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ED&lt;sub&gt;50&lt;/sub&gt; adenosine (−log, μM)</td>
<td>4.3±0.2</td>
<td>4.2±0.2</td>
</tr>
<tr>
<td>ED&lt;sub&gt;50&lt;/sub&gt; noradrenaline (−log, μM)</td>
<td>6.6±0.1</td>
<td>6.9±0.1</td>
</tr>
<tr>
<td>Max. response, noradrenaline (mN/mm)</td>
<td>2.52±0.26</td>
<td>2.32±0.27</td>
</tr>
<tr>
<td>Max. response, angiotensin II (mN/mm)</td>
<td>1.5±0.3</td>
<td>1.3±0.2</td>
</tr>
<tr>
<td>Max. response, K-PSS (mN/mm)</td>
<td>2.2±0.3</td>
<td>2.1±0.2</td>
</tr>
</tbody>
</table>

There were no differences in responses to acetylcholine, adenosine, angiotensin II or noradrenaline between patients and controls before or after invasive treatment. Values are mean±S.E.M.

values were considered normal and the mean clearance of the patients was 98±11 ml/min (estimated by Gault–Cockcroft formula), serum-creatinine was higher in the patients (P<0.05 before and after treatment compared to controls). Proteinuria investigated by Stix method was negative in eight patients. In six patients, Stix was positive, but quantitation showed that proteinuria was less than 0.2 g/l.

4. Discussion

The main findings of this study are that, prior to invasive treatment, the patients were hypertensive despite substantial antihypertensive medication and that media/lumen ratio of resistance arteries was elevated. Invasive treatment resulted in reduction of use of antihypertensive medication, but patients still had increased blood pressure despite normalization of media/lumen ratio.

Our findings that the patients had considerable hypertension despite massive antihypertensive medication is in agreement with many earlier studies and with the well-known difficulty of achieving normotension with antihypertensive medication as one of the key-characteristics of renovascular hypertensive patients [1,20,21]. This refractory hypertension encourages the clinician to focus on alternatives/supplements to medical treatment, and as the complications to PTRA are few and often of minor importance, renovascular hypertensive patients are frequently offered this type of treatment [22–24]. In this study, only two patients became normotensive without medication, but as blood pressure was also reduced in the remaining group and as a reduction of blood pressure of only 10/5–6 mmHg reduces risk of stroke and coronary disease the invasive treatment was of benefit in general in the whole group [25,26].

We found that invasive treatment reduced antihypertensive medication, but only reduced blood pressure approximately 18 mmHg, 6 months after invasive treatment. Xue et al. [3] also found that antihypertensive medication was reduced 6 months after invasive treatment, but returned to pre-treatment level after 12 months. They also measured a fall in blood pressure after 6 months, even though there was a tendency to rise in blood pressure during the follow-up period. Van de Ven et al. [8] reported a reduction of blood pressure and no change in amount of antihypertensive medication after 6 months, but this population was characterized with a lower level of antihypertensive medication before and after treatment than we observed. Webster et al. [4] found that both blood pressure and amount of antihypertensive medications were reduced after 6 months. Bonelli et al. [27] followed patients for mean 33 months and von Knorring et al. [9] followed patients for mean 48 months, and both found reduced blood pressure and use of medicine at the last visit. Weibull et al. [10] investigated patients with ostial stenoses for 24 months, and at that time there was a fall in blood pressure and a fall in number of patients receiving more than three types of antihypertensive medication. Although the above mentioned studies differ in types of treatment, times of follow-up and methods of assessing positive outcome, the overall view is in agreement with our results that invasive treatment is of benefit as regards blood pressure and/or use of antihypertensive medication. However, as the number of patients still taking antihypertensive medication after treatment in the studies mentioned above ranges from 75 to
95%, it is clear that invasive treatment seldom cures the patients.

Our studies of the resistance vessels showed that they were affected by the hypertensive state, demonstrated by an increased media/lumen ratio of resistance arteries in patients compared to controls, which was correlated to the height of the blood pressure. As the measured 21% increase in media cross-sectional area was not significant, the observed greater media/lumen ratio in the vessels from the patients could not be ascribed to hypertrophic remodeling as reported by Rizzoni et al. [16] (who reported a 53% increase in media cross-sectional area). The reason for the discrepancy is not clear, but is not due to a greater blood pressure of the renovascular patients (172/106 mmHg) or lower variance in measurements in the latter study. Increased media/lumen ratio is a well-known phenomenon in hypertensive vessels [16,28–31], but it may be noted that the media/lumen ratio of resistance arteries from the renovascular patients in our study was higher than in vessels with the same size from patients with untreated essential hypertension observed by Thybo et al. [30], despite similar untreated blood pressure in that study. This suggests that blood pressure is not the only factor influencing the resistance vessel structure in the renovascular patients [32,33].

Our findings that media/lumen ratio of resistance arteries was normalized after 6 months has also been found in vessels from essential hypertensive patients after treatment [13,15,34]. Heagerty et al. [35] and Aalkjaer et al. [14] found that treatment with different antihypertensives for mean 13–14 months reduced media/lumen ratio of resistance arteries. Schiffrin et al. [13] and Thybo et al. [15] demonstrated normalization of media/lumen ratio of resistance arteries when patients were treated with an angiotensin converting enzyme-inhibitor for 1 year, while treatment with a beta-blocker did not affect structure. In another study, Schiffrin et al. [31] showed that treatment with a calcium-antagonist also normalized structure after treatment for 1 year. In our investigations concerning renovascular patients treated with different medical regimens, the media/lumen ratio of resistance arteries was already normalized 6 months after invasive treatment, although blood pressure was not reduced to normotensive level. This suggests that removal of the stenosis either resulted in removal of a factor responsible for the hypertensive changes in the resistance arteries, or in production of an antitrophic factor [36]. In addition, the results also show that the persistence of hypertension was not due to increased media/lumen ratio of resistance arteries.

Concerning the response to acetylcholine, we found no changes in relaxation between normotensive and hypertensive persons in agreement with the results of Thybo et al. [30] and Angus et al. [37]. In contrast, Rizzoni et al. [16] found a reduced relaxation in the renovascular hypertensive patients, and both Rizzoni et al. [16] and Schiffrin et al. [31] found a reduced relaxation in the untreated essential hypertensive patients compared to normotensive controls. It is not obvious what gives the difference, but Rizzoni et al. [16] and Schiffrin et al. [31] used higher concentrations of noradrenaline for precontraction than we, Angus et al. [37] and Thybo et al. [30] did, while the concentration of acetylcholine and the size of the vessels were almost the same in all the studies.

Basal function of the kidneys of the patients in our population, evaluated by serum-creatinine and estimates of clearance, was in the normal range, but quantitatively serum-creatinine was higher than in the normotensive controls. Invasive treatment did not change serum-creatinine, which is in contrast to the findings of Jensen et al. [38], who showed that clearance increased in nearly all patient subjected to PTRA, although they did not become normotensive. We can only speculate whether normalization of blood pressure would cause reduction of serum-creatinine in our population. Persistence of higher serum-creatinine than in controls might be due to nephrosclerosis in the non-stenotic kidney because of longstanding hypertension prior to treatment [39–41].

In conclusion, this study showed that media/lumen ratio of resistance arteries and blood pressure were increased in the renovascular hypertensive patients, despite use of antihypertensive medication and that invasive treatment induced a fall in DDD and a normalization of media/lumen ratio of resistance arteries, although patients were still hypertensive.

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