

ENDOTHELIAL FUNCTION AND OXIDATIVE STRESS IN RENOVASCULAR HYPERTENSION

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ABSTRACT

Background It has been reported that renovascular hypertension activates the renin–angiotensin system, leading to an increase in oxidative stress. We sought to determine whether renal-artery angioplasty improves endothelial dysfunction in patients with renovascular hypertension through a reduction in oxidative stress.

Methods We evaluated the response of forearm blood flow to acetylcholine, an endothelium-dependent vasodilator, and isosorbide dinitrate, an endothelium-independent vasodilator, before and after renal-artery angioplasty in 15 subjects with renovascular hypertension and 15 controls without hypertension who were matched for age and sex. Forearm blood flow was measured with the use of a mercury-filled Silastic strain-gauge plethysmograph.

Results The forearm blood flow in response to acetylcholine was less in subjects with renovascular hypertension than in controls, although the forearm blood flow in response to isosorbide dinitrate was similar in the two groups. Angioplasty decreased systolic and diastolic blood pressures, forearm vascular resistance, and urinary excretion of 8-hydroxy-2'-deoxyguanosine and serum malondialdehyde-modified low-density lipoprotein (LDL), indexes of oxidative stress. After angioplasty, the mean (\pm SD) forearm blood flow in response to acetylcholine was increased in the patients with renovascular hypertension (19.3 ± 6.8 vs. 29.6 ± 7.1 ml per minute per 100 ml, $P=0.002$). The increase in the maximal forearm blood flow in response to acetylcholine correlated significantly with the decrease in urinary excretion of 8-hydroxy-2'-deoxyguanosine ($r=-0.51$, $P=0.004$) and serum malondialdehyde-modified LDL ($r=-0.39$, $P=0.02$). Coinfusion of ascorbic acid (vitamin C) augmented the response of forearm blood flow to acetylcholine before angioplasty ($P<0.001$) but not after angioplasty.

Conclusions These findings suggest that excessive oxidative stress is involved, at least in part, in impaired endothelium-dependent vasodilatation in patients with renovascular hypertension. (N Engl J Med 2002;346:1954–62.)

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RENOVASCULAR hypertension caused by renal-artery stenosis leads to stimulation of the renin–angiotensin system and increased production of its main active peptide, angiotensin II. In experimental models of renovascular hypertension, increased vascular oxidative stress plays an important part in the pathogenesis of renovascular hypertension and the enhance-

ment of the oxidation-sensitive mechanism.¹ It has been reported that angiotensin II stimulates the production of reactive oxygen species such as superoxide through the activation of membrane-bound NADH or NADPH oxidase.^{2–4} In addition, both experimental renovascular hypertension and human renovascular hypertension are associated with changes in endothelium-dependent vasodilatation.^{5–7} An imbalance characterized by reduced production of nitric oxide or increased production of reactive oxygen species, mainly superoxide, may promote endothelial dysfunction.^{8–11} One mechanism by which endothelium-dependent vasodilatation is impaired is an increase in the oxidative stress that inactivates nitric oxide. Patients with renovascular hypertension are ideal subjects in whom to determine how endothelium-dependent vasodilatation is affected by excess angiotensin II and an angiotensin II–related increase in oxidative stress. We hypothesized that renal angioplasty would improve impaired endothelial function in subjects with renovascular hypertension by decreasing oxidative stress.

To determine the role of oxidative stress in endothelial function in patients with renovascular hypertension, we evaluated the endothelium-dependent vasodilatation induced by acetylcholine and the endothelium-independent vasodilatation induced by isosorbide dinitrate before and after renal angioplasty and with and without the administration of the antioxidant ascorbic acid (vitamin C).

METHODS

Protocol 1: Endothelial Function in Controls and in Subjects with Renovascular Hypertension

Fifteen subjects with renovascular hypertension (9 men and 6 women; mean [\pm SD] age, 41 ± 15 years) and 15 age- and sex-matched control subjects without hypertension (9 men and 6 women; mean age, 40 ± 13 years) were enrolled in the study. The study protocol was approved by the Ethics Committee of the Hiroshima University Faculty of Medicine. Written informed consent for participation was obtained from all subjects.

Hypertension was defined as a systolic blood pressure of at least 140 mm Hg, a diastolic blood pressure of at least 90 mm Hg, or both, measured with the subject in a sitting position on at least three different occasions in the outpatient clinic of the Hiroshima

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University Faculty of Medicine. The diagnosis of renovascular hypertension was confirmed by renal arteriography. The comparison of plasma renin activity from each renal vein was used to categorize subjects as having isolated unilateral renovascular hypertension or bilateral asymmetric renovascular hypertension. Only subjects with unilateral renal-artery stenosis (six with right-sided stenosis and nine with left-sided stenosis) were included. Six subjects had fibromuscular hyperplasia (four men and two women; mean age, 39 ± 17 years), and nine subjects had atherosclerotic disease (five men and four women; mean age, 46 ± 10 years). Subjects with causes of secondary hypertension other than renovascular disease were excluded on the basis of a complete history; physical, radiologic, and ultrasonographic examinations; and urinalysis. The plasma renin activity and concentrations of aldosterone, angiotensin II, and catecholamine and the serum concentrations of creatinine, potassium, calcium, and free thyroxine were determined; the 24-hour urinary excretion of catecholamines, 17-hydroxycorticosteroids, 17-ketogenic steroids, and vanillylmandelic acid were measured as well. No antihypertensive agents were administered for at least two weeks before the study.

Normal blood pressure was defined as a systolic blood pressure of less than 130 mm Hg and a diastolic blood pressure of less than 80 mm Hg. (Systolic pressures of 130 to 139 mm Hg and diastolic pressures of 80 to 89 mm Hg were considered to represent borderline hypertension, and subjects with such values were excluded from the study.) None of the control subjects had a history of serious medical problems. No subject in either group currently smoked or had a history of smoking.

Forearm vascular responses to acetylcholine (Daiichi Pharmaceutical) and isosorbide dinitrate (Eisai Pharmaceutical) were evaluated in all subjects. The study began at 8:30 a.m. Subjects had fasted the previous night for at least 12 hours. They were kept in the supine position in a quiet, dark, air-conditioned room (temperature, 22°C to 25°C) throughout the study. A 23-gauge polyethylene catheter (Hakkow) was inserted into the left brachial artery for the infusion of acetylcholine and isosorbide dinitrate and for recording the arterial pressure with the use of a pressure transducer (Nihon Kohden) under local anesthesia (1 percent lidocaine). We inserted a second catheter into the left deep antecubital vein to obtain blood samples. After the subjects had spent 30 minutes in the supine position, we measured forearm blood flow and arterial blood pressure. Then, the effects of the infusions of acetylcholine and isosorbide dinitrate on forearm hemodynamics were measured. Acetylcholine (7.5, 15.0, and $30.0 \mu\text{g}$ per minute) and isosorbide dinitrate (0.75, 1.5, and $3.0 \mu\text{g}$ per minute) were infused intraarterially for five minutes at each dose with the use of a constant-rate infusion pump (Terfusion STG-523, Termo). The forearm blood flow was measured during the last two minutes of the infusion. The infusions of acetylcholine and isosorbide dinitrate were carried out in random order. Each study proceeded after the forearm blood flow had returned to base line. During fasting, base-line serum concentrations of total cholesterol, high-density lipoprotein (HDL) cholesterol, low-density lipoprotein (LDL) cholesterol, malondialdehyde-modified LDL, triglycerides, creatinine, insulin, glucose, and electrolytes and plasma concentrations of catecholamines, plasma renin activity, and concentrations of angiotensin II were obtained after the 30-minute rest period. The 24-hour urinary excretion of nitrite and nitrate, as well as 8-hydroxy-2'-deoxyguanosine, was determined.

Protocol 2: Effect of Renal Angioplasty on Endothelial Function in Subjects with Renovascular Hypertension

Vasodilative responses to acetylcholine and isosorbide dinitrate were evaluated in a manner identical to that of Protocol 1 before percutaneous transluminal renal angioplasty and within 4 weeks (mean, 21 ± 3 days; range, 15 to 28) after angioplasty in the 15 subjects with renovascular hypertension. We confirmed that plasma renin activity and blood pressure were in the normal range two weeks after angioplasty in all subjects. No antihypertensive agents were administered after renal angioplasty.

To assess the effect of oxidative stress on endothelium-dependent vasodilatation in subjects with renovascular hypertension, we infused acetylcholine in the presence of an antioxidant, ascorbic acid (Fuso Pharmaceutical), before and after angioplasty in 11 of 15 subjects with renovascular hypertension (8 men and 3 women; mean age, 38 ± 13 years). The forearm vascular responses to acetylcholine alone and in combination with ascorbic acid (24 mg per minute) were evaluated with the use of a protocol identical to Protocol 1. The 24-hour urinary excretion of nitrite and nitrate, and 8-hydroxy-2'-deoxyguanosine were measured before and after angioplasty.

Measurement of Forearm Blood Flow

The forearm blood flow was measured with the use of a mercury-filled Silastic strain-gauge plethysmograph (model EC5R, D.E. Hokanson), as previously described.^{12,13} Forearm blood flow was expressed in milliliters per minute per 100 ml of forearm tissue volume. Four plethysmographic measurements were averaged to determine the forearm blood flow at base line and during the administration of drugs. Forearm vascular resistance was calculated as the mean arterial pressure divided by the forearm blood flow.

Analytical Methods

Routine chemical methods were used to determine serum concentrations of total cholesterol, HDL cholesterol, LDL cholesterol, triglycerides, creatinine, glucose, and electrolytes. Plasma renin activity (Gamma Coat PRA, SRL) and angiotensin II (antiangiotensin II antibody, SRL) were assayed by radioimmunoassay. The plasma and urinary concentrations of catecholamines were measured by high-performance liquid chromatography. Urinary concentrations of nitrite and nitrate were assayed by colorimetric methods with the use of commercially available nitrite and nitrate assay kits (Cayman Chemical). The plasma and urinary concentrations of 8-hydroxy-2'-deoxyguanosine were assayed by enzyme-linked immunosorbent assay (ELISA) with the use of 8-hydroxy-2'-deoxyguanosine kits (Nihon Yushi). The serum concentrations of malondialdehyde-modified LDL were also assayed by ELISA (antimalondialdehyde-modified LDL antibody, SRL). Blood samples obtained before and after renal angioplasty in the same subject were assayed in the same batch to minimize day-to-day variation. The intraassay and interassay coefficients of variation were 6.2 percent and 7.6 percent, respectively, for plasma renin activity; 8.9 percent and 9.4 percent for angiotensin II; 7.9 percent and 8.4 percent for norepinephrine; 6.8 percent and 7.7 percent for malondialdehyde-modified LDL; 4.1 percent and 3.8 percent for nitric oxide; and 5.9 percent and 6.5 percent for 8-hydroxy-2'-deoxyguanosine.

Statistical Analysis

Results are presented as means \pm SD. All reported P values are two-tailed. P values of less than 0.05 were considered to indicate statistical significance. Multigroup comparisons of variables were carried out by the one-way analysis of variance followed by the Bonferroni correction. Comparisons of variables before and after angioplasty were performed with adjusted means by analysis of covariance with the use of base-line data as covariates. Comparisons of time-course curves of variables during the infusions of acetylcholine and isosorbide dinitrate were analyzed by two-way analysis of variance for repeated measures on one factor followed by the Bonferroni correction for multiple paired comparisons. Relations between variables were determined by linear regression analysis. The data were processed with the use of the software package StatView IV (SAS Institute) or Super Analysis of Variance (Abacus Concepts).

RESULTS

Protocol 1

The base-line clinical characteristics of the 15 controls and the 15 subjects with renovascular hyperten-

sion are summarized in Table 1. The systolic and diastolic blood pressures as well as the forearm vascular resistance were higher in subjects with renovascular hypertension than in controls. The plasma renin activity and plasma angiotensin II concentration, serum malondialdehyde-modified LDL concentration, and urinary excretion of 8-hydroxy-2'-deoxyguanosine were higher and the urinary excretion of nitrite and nitrate was lower in subjects with renovascular hypertension than in controls. The other values were similar in the two groups.

The intraarterial infusion of acetylcholine increased forearm blood flow in a dose-dependent manner in both groups. The response of forearm blood flow to acetylcholine was greater in controls than in subjects with renovascular hypertension ($P < 0.001$) (Fig. 1A).

The intraarterial infusion of isosorbide dinitrate also increased forearm blood flow in a dose-dependent manner in both groups, but the response of forearm blood flow was similar in the two groups (Fig. 1B). No significant change was observed in the arterial blood pressure or heart rate with the intraarterial infusion of either acetylcholine or isosorbide dinitrate in either group.

Protocol 2

The base-line clinical characteristics, before and after renal angioplasty, of the six subjects with renovascular hypertension due to fibromuscular hyperplasia and the nine subjects with renovascular hypertension due to atherosclerotic disease are summarized in Table 2. Renal angioplasty decreased plasma renin activity and the

TABLE 1. CLINICAL CHARACTERISTICS OF CONTROLS AND SUBJECTS WITH RENOVASCULAR HYPERTENSION BEFORE AND AFTER ANGIOPLASTY.

CHARACTERISTIC*	CONTROLS (N=15)	SUBJECTS WITH RENOVASCULAR HYPERTENSION (N=15)	
		BEFORE ANGIOPLASTY	AFTER ANGIOPLASTY
		mean ± SD	
Body-mass index	23.2±1.9	23.9±1.8	23.7±1.8
Systolic blood pressure (mm Hg)	115.3±8.4	161.4±17.5†	124.2±7.3‡
Diastolic blood pressure (mm Hg)	67.9±6.2	98.7±8.9†	77.1±6.2‡
Heart rate (beats/min)	66.5±6.3	79.4±9.3	65.2±6.0
Total cholesterol (mmol/liter)	5.11±0.69	5.78±0.98	5.32±0.78
Triglycerides (mmol/liter)	1.12±0.57	1.76±0.79	1.38±0.59
HDL cholesterol (mmol/liter)	1.43±0.41	1.16±0.64	1.21±0.41
LDL cholesterol (mmol/liter)	3.49±0.57	3.78±0.77	3.51±0.57
Serum malondialdehyde-modified LDL (U/liter)	69.1±31.8	126.3±47.1†	78.5±21.0‡
Serum glucose (mmol/dl)	4.7±0.4	5.8±1.2	4.9±0.4
Serum insulin (pmol/liter)	51.2±10.3	42.1±15.4	52.3±9.8
Plasma nitrite and nitrate (μmol/liter)	33.1±19.1	29.2±18.3	35.7±20.2
Plasma norepinephrine (ng/ml)	0.28±0.17	0.37±0.21	0.33±0.19
Plasma renin activity (ng/ml/hr)	1.34±0.55	2.01±0.73†	1.41±0.68‡
Plasma angiotensin II (pg/ml)	17.9±8.4	27.2±10.2†	22.3±11.3‡
Urinary nitrite and nitrate (μmol/mmol of creatinine)	138.9±60.2	87.8±34.7†	120.3±56.4‡
Urinary norepinephrine (μg/mg of creatinine)	73.9±24.1	81.8±29.5	76.3±22.9
Urinary 8-hydroxy-2'-deoxyguanosine (ng/mg of creatinine)	9.7±4.6	19.8±8.7†	10.8±4.1‡
Forearm blood flow (ml/min/100 ml of tissue)	4.7±1.3	4.1±1.6	4.5±1.4
Forearm vascular resistance (mm Hg/ml/min/100 ml of tissue)	18.3±3.9	29.1±5.7†	18.8±4.1‡

*The body-mass index was calculated as the weight in kilograms divided by the square of the height in meters. To convert values for cholesterol (total, HDL, and LDL) to milligrams per deciliter, divide by 0.02586; to convert values for triglycerides to milligrams per deciliter, divide by 0.01129; to convert values for glucose to milligrams per deciliter, divide by 0.05551; to convert values for insulin to microunits per milliliter, divide by 6.0; to convert values for norepinephrine to nanomoles per liter, multiply by 0.05911; and to convert values for plasma renin activity to nanograms per liter-second, multiply by 0.2778.

† $P < 0.05$ for the comparison with controls.

‡ $P < 0.05$ for the comparison with subjects with renovascular hypertension before angioplasty.

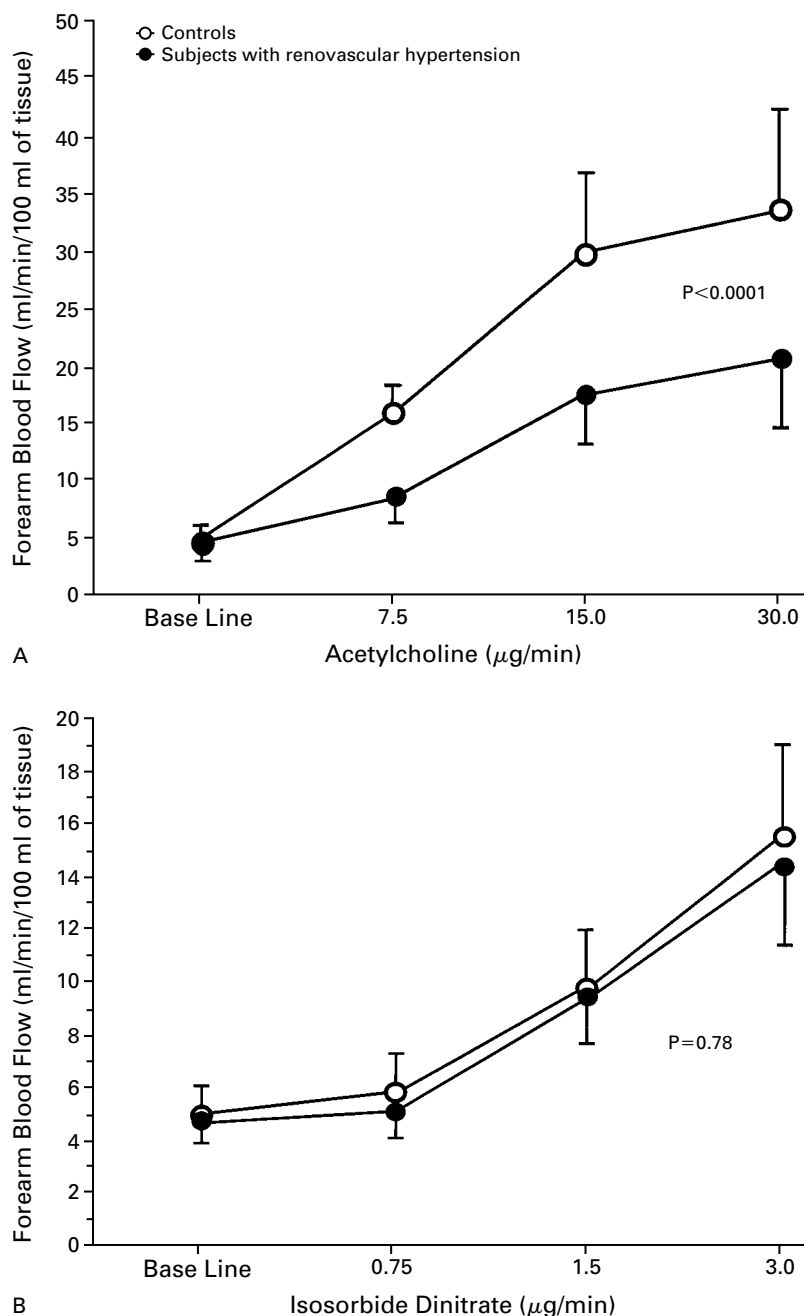


Figure 1. Comparison of the Mean (\pm SD) Response of Forearm Blood Flow to the Administration of Acetylcholine (Panel A) and Isosorbide Dinitrate (Panel B) in Controls and Subjects with Renovascular Hypertension.

plasma angiotensin II concentration, serum malondialdehyde-modified LDL concentration, urinary excretion of 8-hydroxy-2'-deoxyguanosine, systolic and diastolic blood pressures, and forearm vascular resistance and increased urinary excretion of nitrite and nitrate in both groups. Changes in these values were similar in the two groups.

After renal angioplasty, the responses of forearm blood flow to acetylcholine were enhanced in both subjects with renovascular hypertension due to fibromuscular hyperplasia (maximal forearm blood flow, 21.1 ± 8.5 vs. 32.2 ± 8.9 ml per minute per 100 ml; $P = 0.002$) (Fig. 2A) and subjects with renovascular hypertension due to atherosclerosis (maximal forearm

TABLE 2. CLINICAL CHARACTERISTICS OF RENOVASCULAR HYPERTENSION WITH FIBROMUSCULAR HYPERPLASIA AND ATHEROSCLEROSIS BEFORE AND AFTER ANGIOPLASTY.

CHARACTERISTIC*	SUBJECTS WITH FIBROMUSCULAR HYPERPLASIA (N=6)		SUBJECTS WITH ATHEROSCLEROSIS (N=9)	
	BEFORE ANGIOPLASTY	AFTER ANGIOPLASTY	BEFORE ANGIOPLASTY	AFTER ANGIOPLASTY
	mean ± SD			
Body-mass index	23.9±2.1	23.3±1.9	23.8±1.7	23.8±1.8
Systolic blood pressure (mm Hg)	168.5±24.5	119.7±10.3†	159.9±17.1	126.4±9.7†
Diastolic blood pressure (mm Hg)	102.2±9.5	75.8±7.4†	96.8±8.8	79.7±6.8†
Heart rate (beats/min)	80.3±9.5	66.4±6.7	77.9±9.1	65.1±6.0
Total cholesterol (mmol/liter)	5.66±1.03	5.23±0.81	5.79±0.89	5.41±0.77
Triglycerides (mmol/liter)	1.68±0.67	1.40±0.61	1.78±0.71	1.36±0.57
HDL cholesterol (mmol/liter)	1.18±0.69	1.23±0.50	1.15±0.63	1.19±0.39
LDL cholesterol (mmol/liter)	3.71±0.81	3.46±0.60	3.80±0.72	3.52±0.54
Serum malondialdehyde-modified LDL (U/liter)	119.5±51.2	78.3±14.1†	130.7±46.7	77.9±24.2†
Serum glucose (mmol/dl)	5.6±1.2	4.9±0.6	5.8±1.3	4.9±0.4
Serum insulin (pmol/liter)	45.6±20.1	53.5±10.4	41.3±14.6	51.1±9.5
Plasma nitrite and nitrate (μmol/liter)	30.4±21.1	36.3±24.5	28.7±17.9	34.6±19.6
Plasma norepinephrine (ng/ml)	0.41±0.29	0.36±0.22	0.35±0.20	0.32±0.18
Plasma renin activity (ng/ml/hr)	2.24±0.85	1.44±0.73†	1.99±0.71	1.39±0.67†
Plasma angiotensin II (pg/ml)	30.4±13.6	23.7±12.5†	26.9±9.8	21.8±10.9†
Urinary nitrite and nitrate (μmol/mmol of creatinine)	84.5±40.1	122.5±51.8†	88.4±33.9	119.4±60.1†
Urinary norepinephrine (μg/mg of creatinine)	86.7±31.9	79.2±28.8	80.7±28.5	75.4±21.7
Urinary 8-hydroxy-2'-deoxyguanosine (ng/mg of creatinine)	20.2±8.5	9.5±3.1†	19.6±9.3	11.4±3.9†
Forearm blood flow (ml/min/100 ml of tissue)	4.2±1.7	4.6±1.4	4.1±1.6	4.4±1.5
Forearm vascular resistance (mm Hg/ml/min/100 ml of tissue)	29.5±6.1	18.6±4.3†	28.8±5.5	19.1±4.0†

*To convert values for cholesterol (total, HDL, and LDL) to milligrams per deciliter, divide by 0.02586; to convert values for triglycerides to milligrams per deciliter, divide by 0.01129; to convert values for glucose to milligrams per deciliter, divide by 0.05551; to convert values for insulin to microunits per milliliter, divide by 6.0; to convert values for norepinephrine to nanomoles per liter, multiply by 0.05911; and to convert values for plasma renin activity to nanograms per liter·second, multiply by 0.2778.

†P<0.05 for the comparison with subjects before angioplasty.

blood flow, 19.1±6.5 vs. 29.5±7.0 ml per minute per 100 ml; P=0.004) (Fig. 2B). Changes in the responses of forearm blood flow to acetylcholine were similar before and after renal angioplasty in the two groups. The response of forearm blood flow to isosorbide dinitrate was unaffected by angioplasty in both groups. No significant change was observed in the arterial blood pressure or heart rate in response to intraarterial infusion of either acetylcholine or isosorbide dinitrate before or after renal angioplasty. The increase in the maximal response of forearm blood flow to acetylcholine correlated with the decrease in the urinary excretion of 8-hydroxy-2'-deoxyguanosine ($r = -0.51$, $P = 0.004$) and the decrease in the serum concentration of malondialdehyde-modified LDL ($r = -0.39$, $P = 0.02$) (Fig. 3). There was no correlation between the increase

in the maximal response of forearm blood flow to acetylcholine and changes in blood pressure, heart rate, plasma norepinephrine concentration, or other variables such as plasma renin activity or plasma angiotensin II concentration or between these variables and the increase in the maximal response of forearm blood flow to isosorbide dinitrate.

Coinfusion of ascorbic acid augmented the response of forearm blood flow to acetylcholine before angioplasty (maximal forearm blood flow, 20.2±6.7 vs. 28.1±4.8 ml per minute per 100 ml; $P = 0.006$) but not after angioplasty in subjects with renovascular hypertension (Fig. 4). No significant change was observed in the arterial blood pressure or heart rate with the intraarterial infusion of acetylcholine in combination with ascorbic acid.

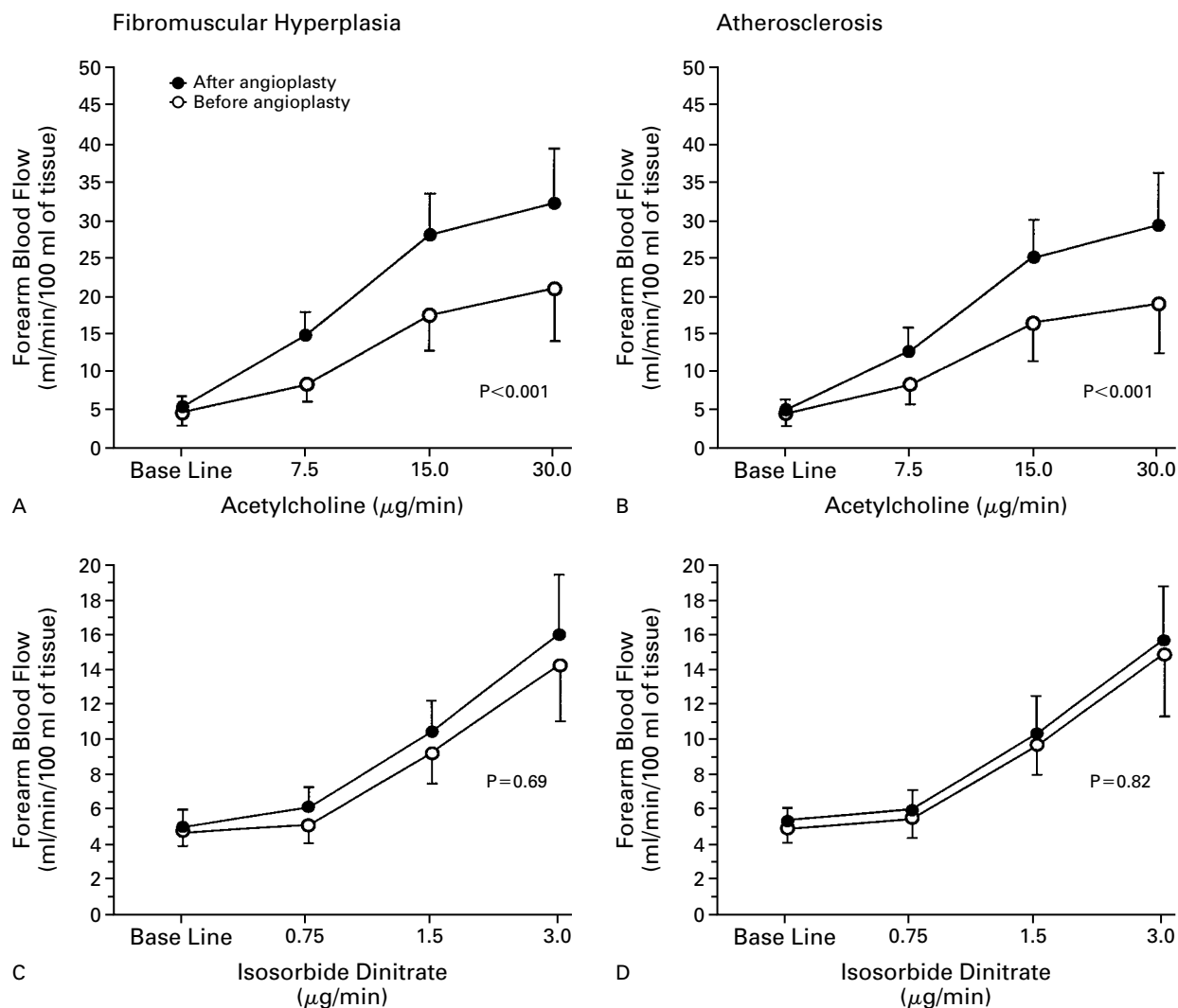


Figure 2. Comparison of the Mean (\pm SD) Response of Forearm Blood Flow to the Administration of Acetylcholine (Panels A and B) and Isosorbide Dinitrate (Panels C and D) before and after Angioplasty in Subjects with Renovascular Hypertension Caused by Fibromuscular Hyperplasia and Atherosclerosis.

DISCUSSION

Patients with elevations of the angiotensin II concentration induced by renovascular hypertension are ideal subjects in whom to study how endothelium-dependent vasodilatation is altered under conditions of increased oxidative stress. Endothelium-dependent vasodilatation induced with acetylcholine was blunted in subjects with renovascular hypertension, most likely through a decrease in the release of nitric oxide.

A balance between ambient levels of superoxide and the release of nitric oxide has a critical role in the maintenance of normal endothelial function.^{14,15} Both 8-hydroxy-2'-deoxyguanosine and malondialdehyde-modified LDL have been used as indexes of oxidative

stress.¹⁴⁻¹⁹ The compound 8-hydroxy-2'-deoxyguanosine is one of the most common markers for evaluating oxidative DNA damage and is a product formed by the specific attack of a hydroxyl radical on DNA.¹⁴ Several studies have suggested that oxidative DNA damage is increased in non-insulin-dependent diabetes mellitus and aging.^{15,16} The concentration of malondialdehyde-modified LDL has been proposed as the biologic signature of clinical in vivo LDL oxidation.^{17,18} Maggi et al.¹⁹ reported that the serum malondialdehyde-modified LDL concentration is higher in subjects with essential hypertension than in normal controls. In the present study, subjects with renovascular hypertension had a higher urinary excretion of

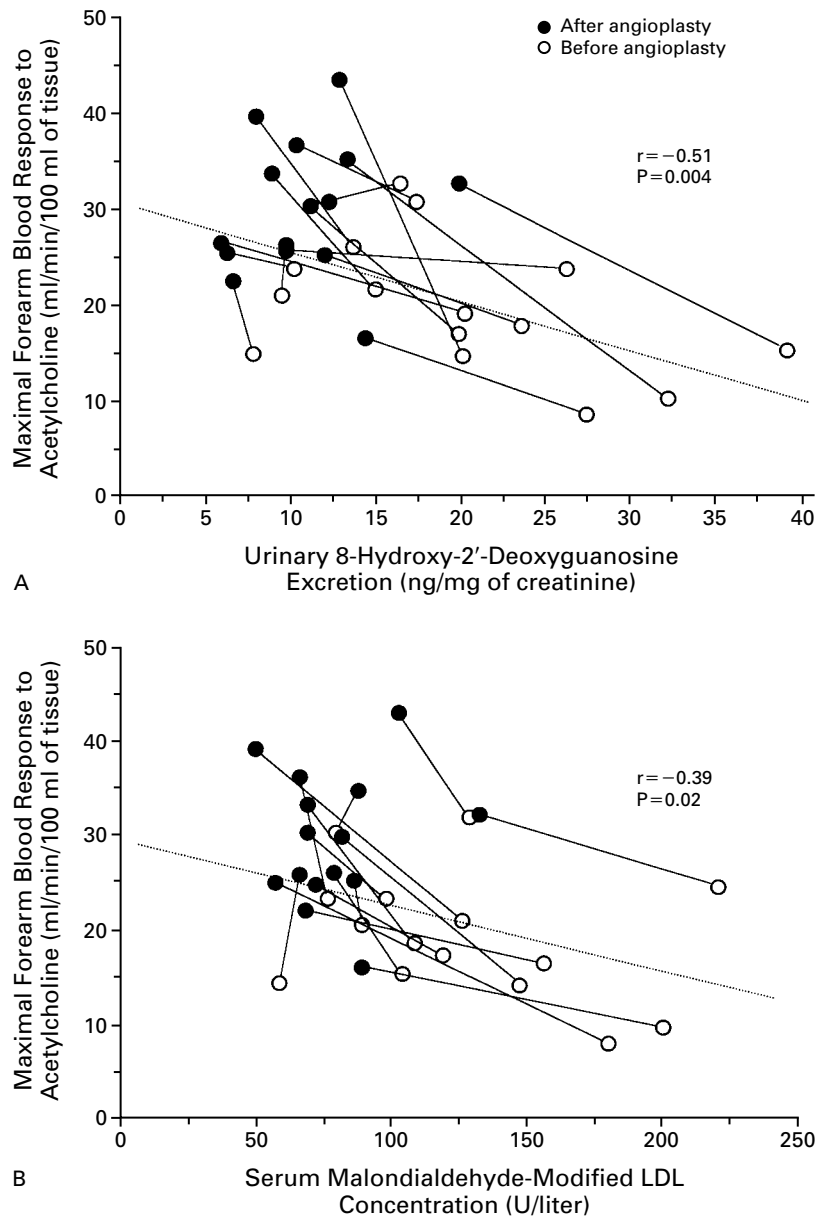


Figure 3. Correlation between the Maximal Response of Forearm Blood Flow to the Administration of Acetylcholine and Urinary Excretion of 8-Hydroxy-2'-Deoxyguanosine (Panel A) and the Serum Malondialdehyde-Modified LDL Concentration (Panel B) before and after Angioplasty in Subjects with Renovascular Hypertension.

8-hydroxy-2'-deoxyguanosine and serum malondialdehyde-modified LDL concentration than controls, suggesting that oxidative stress is increased in clinical renovascular hypertension as well. The improvement of endothelium-dependent vasodilatation correlated with the decrease in urinary excretion of 8-hydroxy-2'-deoxyguanosine and the serum concentration of

malondialdehyde-modified LDL. In addition, ascorbic acid, an antioxidant, augmented the response of forearm blood flow to acetylcholine before, but not after, angioplasty. One possible mechanism by which renal angioplasty improves endothelium-dependent vasodilatation is by decreasing oxidative stress, which may cause endothelial dysfunction directly.

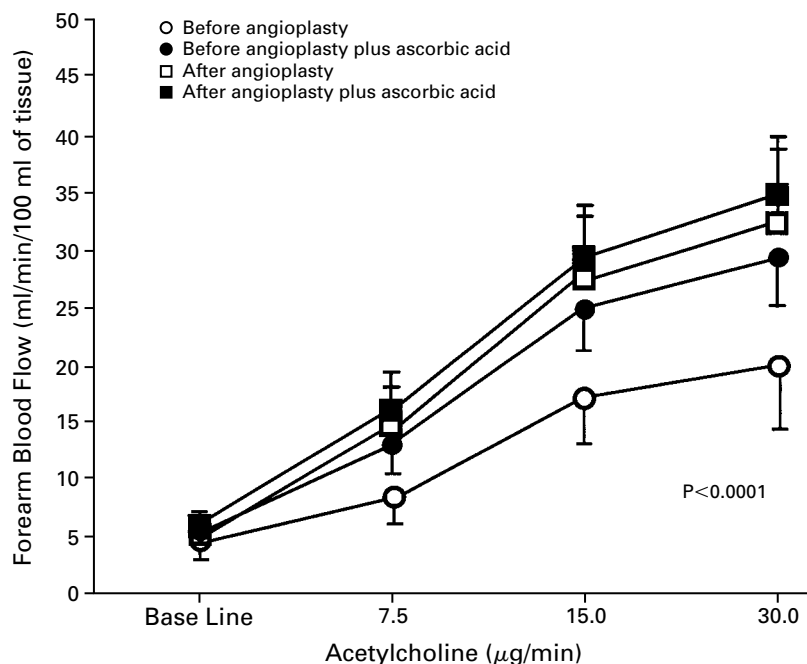


Figure 4. Mean (\pm SD) Effect of Concomitant Administration of the Antioxidant Ascorbic Acid on the Response of Forearm Blood Flow to the Administration of Acetylcholine before and after Angioplasty in Subjects with Renovascular Hypertension.

The principal source of superoxide in renovascular hypertension is an activation of NADH or NADPH oxidase that is induced by angiotensin II.²⁻⁴ Successful renal angioplasty and consequent down-regulation of the renin-angiotensin system may decrease oxidative stress, resulting in improved endothelium-dependent vasodilatation. Therefore, angioplasty may increase the bioavailability of nitric oxide by inhibiting production of angiotensin II. These findings suggest that the role of the renin-angiotensin system in the pathogenesis of atherosclerosis may be due, at least in part, to angiotensin II-induced production of superoxide by vascular cells.

Endothelial function becomes progressively impaired as blood pressure increases, and the degree of dysfunction is related to the severity of the hypertension.^{20,21} It is thought that endothelial dysfunction is improved by antihypertensive therapy. However, several experimental and clinical studies have generated conflicting results concerning this relation.²²⁻²⁴ Although renal angioplasty acutely decreased blood pressure in subjects with renovascular hypertension in the present study, the changes in blood pressure did not correlate with the improvement in the response of forearm blood flow to acetylcholine. In previous stud-

ies, we and other investigators have shown that calcium antagonists, beta-blockers, or diuretics do not improve endothelial dysfunction in subjects with essential hypertension, although all of these drugs have hypotensive effects.²³⁻²⁶ In addition, although clinically effective antihypertensive therapies, such as angiotensin-converting-enzyme inhibitors and aerobic exercise, have restored endothelial function in the forearm circulation in patients with essential hypertension, there is no correlation between the degree of reduction in blood pressure and the augmentation of endothelium-dependent vasodilatation.^{13,23-26} Therefore, a reduction in blood pressure may not itself be involved in the restoration of endothelial function in the forearm circulation.

Norepinephrine, a potent vasoconstrictor, attenuates endothelium-dependent vasodilatation.^{27,28} Stimulation of the renin-angiotensin system modulates autonomic nervous function. However, both plasma and urinary concentrations of norepinephrine were similar before and after angioplasty in the present study. Therefore, the differences in the response of forearm blood flow to acetylcholine after angioplasty cannot be explained by differences in sympathetic activity.

Although subjects with renovascular hypertension due to atherosclerosis might be expected to have lower responses of oxidative stress to renal angioplasty and lower responses of forearm blood flow to acetylcholine than subjects with renovascular hypertension due to fibromuscular hyperplasia, we found no significant difference in the changes in oxidative stress or improvement in endothelium-dependent vasodilatation between the two groups. The increased levels of angiotensin II due to renal-artery stenosis may have caused markedly excessive oxidative stress and may have strongly affected endothelial function. Since five of the nine subjects who had renovascular hypertension due to atherosclerosis were less than 50 years of age, there may be relatively short periods in which atherosclerosis has an influence. In addition, the number of subjects in each group was relatively small. We cannot exclude the possibility that there was selection bias in the results. However, drastic changes in oxidative stress and endothelial function were observed with renal angioplasty. A larger number of subjects are needed to determine conclusively that there is no difference between subjects with fibromuscular hyperplasia and those with atherosclerosis.

Increased production of superoxide impairs endothelium-dependent vasodilatation in the forearm circulation in humans. The dilatation of a stenotic artery by renal angioplasty improved endothelium-dependent vasodilatation in patients with renovascular hypertension through a decrease in oxidative stress.

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