Asymmetric Dimethylarginine Plasma Concentrations Differ in Patients with End-Stage Renal Disease: Relationship to Treatment Method and Atherosclerotic Disease

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Abstract. Asymmetric dimethylarginine (ADMA) is an endogenous inhibitor of endothelial nitric oxide (NO) synthase. Its concentration is elevated in patients with end-stage renal disease (ESRD), in part because it is excreted via the kidneys. In this study, the plasma concentrations of ADMA, symmetric dimethylarginine, and L-arginine were determined in relation to plasma nitrate levels (as an index of NO formation) for a group of 80 patients with ESRD. The effects of two treatment methods, i.e., hemodialysis (HD) and peritoneal dialysis (PD), and the role of the presence of atherosclerotic disease were evaluated. Forty-three patients receiving HD and 37 patients receiving PD were compared with healthy control subjects. Plasma L-arginine and dimethylarginine levels were determined by HPLC, using precolumn derivatization with o-phthalaldehyde. Plasma nitrate levels were determined by gas chromatography-mass spectrometry. Predialysis ADMA concentrations in HD-treated patients were approximately sixfold higher than those in the control group (6.0 ± 0.5 versus 1.0 ± 0.1 µmol/L; P < 0.05). Plasma nitrate concentrations were significantly lower in HD-treated patients, which suggests that ADMA may inhibit NO synthase. In contrast, plasma ADMA levels and nitrate concentrations in PD-treated patients were similar to those in control subjects. Plasma L-arginine concentrations were not significantly decreased in patients with ESRD. ADMA concentrations were significantly decreased 5 h after HD, compared with baseline values. ADMA levels were significantly higher in HD-treated patients with manifest atherosclerotic disease than in HD-treated patients without atherosclerotic disease (7.31 ± 0.70 versus 3.95 ± 0.52 µmol/L; P < 0.05). This study confirms that ADMA is accumulated in ESRD. PD-treated patients exhibit significantly lower ADMA levels than HD-treated patients. Accumulation of ADMA may be a risk factor for the development of endothelial dysfunction and cardiovascular disease in patients with ESRD.

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Endothelium-derived nitric oxide (NO) plays an important role in the regulation of BP and platelet aggregation. It is synthesized by stereospecific oxidation of the terminal guanidino nitrogen of the amino acid L-arginine (1). NO is produced by the action of a family of NO synthases, with endothelial, neuronal, and macrophage isoforms (2).

The synthesis of NO can be selectively inhibited by guanidino-substituted analogs of L-arginine, such as N-monomethyl-L-arginine, which act as competitive antagonists at the active site of the enzyme (3). Asymmetric dimethylarginine (ADMA) has been recently characterized as an endogenous inhibitor of NO synthase (4). It is synthesized and metabolized by human endothelial cells (5). ADMA and its biologically inactive stereoisomer symmetric dimethylarginine (SDMA) are at least in part eliminated via urinary excretion (6). Vallance et al. (7) were the first to report elevated plasma levels of ADMA and SDMA in a small group of patients with end-stage renal disease (ESRD). In their study, dimethylarginine (DMA) levels were elevated approximately sixfold, compared with those for healthy control subjects. Those authors suggested that the high incidence of conditions such as hypertension, atherosclerosis, and immune dysfunction among patients with ESRD might be caused at least in part by dysfunction of the L-arginine/NO pathway secondary to accumulation of ADMA (7).

Data from several experimental studies suggest that ADMA concentrations in a pathophysiologically high range (3 to 15 µmol/L) significantly inhibit vascular NO formation (8,9). Moreover, it was recently found by our group that plasma ADMA levels are also elevated in atherosclerotic patients and in hypercholesteremic subjects with normal renal function, suggesting that mechanisms other than decreased renal elimination may contribute to the elevation of plasma ADMA levels in hypercholesterolemia and atherosclerosis (10). It is therefore unclear whether elevated ADMA concentrations in patients...
with ESRD are a cause or a consequence of accompanying cardiovascular disease.

The aim of this study was to determine the plasma concentrations of ADMA, SDMA, and \( \text{L-arginine} \) and their relationship to NO formation (measured as plasma nitrate concentrations) in patients with ESRD, compared with age-matched control subjects with normal renal excretory function. The effects of two treatment methods, i.e., hemodialysis (HD) and peritoneal dialysis (PD), on these parameters were also evaluated.

**Materials and Methods**

**Patients and Study Design**

Four groups of individuals were included in this study, after they had given informed consent for participation. The first group consisted of 43 patients receiving HD (20 female and 23 male patients; mean age, 64.9 ± 1.6 yr) who had been treated with HD for a median of 38 mo (range, 1 to 200 mo), with residual diuresis of 582 ± 95 ml of urine/24 h. Sixteen of the 43 (37%) HD-treated patients were anuric. The average diuresis among the remaining 27 patients was 776 ± 116 ml/24 h.

For these patients, a venous blood sample was drawn at midweek before dialysis. Patients were treated with HD three times each week and were in clinically stable conditions. The mean serum creatinine level was 707 ± 108 \( \mu \text{mol/L} \), and the mean arterial BP was 141.0 ± 3.4/78 ± 1.7 mmHg. Average serum cholesterol and triglyceride levels were 5.29 ± 0.17 and 2.09 ± 0.16 mmol/L, respectively. The body mass index averaged 23.6 ± 0.6, and the mean serum albumin level was 37.6 ± 1.1 g/L. Patients underwent standard bicarbonate HD with biocompatible membranes (Hemophan® [Gambro Medizintechnik, Munich, Germany] and polyamide membranes, sterilized with steam). The average dialysis time was 270 ± 6 min, blood flow was 290 ± 8 ml/min, and dialysate flow was 500 ml/min. The delivered dialysis dose measured by urea reduction rate was 67.2 ± 1.1%, well above the suggested dialysis dose (by urea reduction rate) of 65% (11).

Subgroup analysis was performed for patients with ESRD with atherosclerotic vascular disease (11), which was defined as the presence of clinically and angiographically confirmed peripheral arterial occlusive disease (Fontaine stage IIB to IV) alone (11) or in combination with a history of prior myocardial infarction (11), compared with patients with ESRD without vascular disease (11). For a subgroup of eight HD-treated patients, additional heparinized blood samples for the analysis of ADMA, SDMA, \( \text{L-arginine} \), and nitrate were drawn 1, 5, and 18 h after the end of a dialysis session (dialysis duration, 4.5 h). These patients (five female and three male patients; mean age, 63.1 ± 4.5 yr) had been receiving HD for a median of 18 mo (range, 1 to 82 mo). The mean serum creatinine concentration was 760 ± 49 \( \mu \text{mol/L} \), and the mean arterial BP was 133.8 ± 9.7/81.3 ± 3.7 mmHg. Average serum cholesterol and triglyceride levels were 5.33 ± 0.50 and 2.44 ± 0.42 mmol/L, respectively.

The second group consisted of 37 patients (16 female and 21 male patients) receiving PD (24 patients undergoing nightly intermittent PD, nine continuous ambulatory PD, three continuous cyclic PD, and one intermittent PD; mean age, 46.4 ± 2.5 yr), who had received PD for a median of 55 mo (range, 7 to 168 mo). These patients exhibited residual diuresis of 658 ± 129 ml of urine/24 h. Thirteen of the 37 (35%) patients receiving PD were anuric. The average diuresis among the remaining 24 patients was 1015 ± 151 ml/24 h (\( P \) not significant versus HD). The weekly \( \text{Kt/V}_{\text{urea}} \) value for the patients receiving PD was 2.13 ± 0.09, well above the recommended weekly \( \text{Kt/V}_{\text{urea}} \) value of 2.0 (12).

The mean serum creatinine concentration was 922 ± 47 \( \mu \text{mol/L} \), and the mean BP was 137.2 ± 2.8/83.8 ± 1.6 mmHg. Average serum cholesterol and triglyceride levels were 6.16 ± 0.24 and 2.80 ± 0.26 mmol/L, respectively. The body mass index averaged 23.2 ± 0.6, and the mean serum albumin concentration was 36.7 ± 0.8 g/L. Heparinized blood samples from fasting subjects were obtained during routine morning visits in the PD clinic.

The control group consisted of 37 elderly, healthy, normotensive subjects with no apparent disease (13 female and 24 male patients; mean age, 68.3 ± 1.1 yr), who exhibited mean serum creatinine concentrations of 79.3 ± 3.5 \( \mu \text{mol/L} \). Average cholesterol and triglyceride levels were 4.86 ± 0.11 and 1.61 ± 0.12 mmol/L, respectively. The presence of cardiovascular or metabolic disease was excluded by medical histories, physical examinations, and routine laboratory tests.

An additional control group consisted of 33 patients with manifest arteriosclerotic disease and normal renal function. These patients (13 female and 20 male patients; mean age, 61.4 ± 4.0 yr) had peripheral arterial occlusive disease, as verified by angiography or duplex-sonography. The serum creatinine concentration in this group was 96.0 ± 4.4 \( \mu \text{mol/L} \); average cholesterol and triglyceride levels were 6.24 ± 0.22 and 1.81 ± 0.14 mmol/L, respectively, and BP was 152.1 ± 3.2/79.4 ± 2.0 mmHg.

**Biochemical Analyses**

The plasma concentrations of \( \text{L-arginine} \), \( \text{N}^{\text{G}, \text{N}^{\text{G}}} - \text{ADMA} \) (ADMA), and \( \text{N}^{\text{G}, \text{N}^{\text{G}}} - \text{SDMA} \) (SDMA) were measured by HPLC with precolumn derivatization with \( \text{o-phthalaldehyde (OPA)} \), using a modification of a previously published method (13). \( \text{L-Homoarginine (10 \( \mu \text{mol/L} \) ) was added to 0.5 ml of plasma as an internal standard. Plasma samples and standards were extracted on solid-phase extraction cartridges (CBA Bond Elut; Varian, Harbor City, CA). The recovery rates were 82.9 ± 3.8%. Eluates were dried under nitrogen and resuspended in double-distilled water for HPLC analysis. Standards and samples were incubated with OPA reagent (5.4 mg/ml OPA in borate buffer, pH 8.4, containing 0.4% 2-mercaptoethanol) for exactly 30 s before automatic injection into the HPLC system. The OPA derivatives of \( \text{L-arginine} \), ADMA, and SDMA were separated on a 250-× 4.5-mm (inner diameter), 7-\( \mu \text{m} \), Nucleosil phenyl column (Macherey and Nagel, Düren, Germany), with the fluorescence detector set for an excitation wavelength of 340 nm and an emission wavelength of 450 nm. Samples were eluted from the column with 99.6% citric acid/methanol (2:1, pH 6.8), at a flow rate of 1 ml/min. Standard curves generated for ADMA and SDMA in water and in pooled human plasma showed linearity over a concentration range from 0.1 to 16 \( \mu \text{mol/L} \). The coefficients of variation of this method were 5.2% within assays and 5.5% between assays; the detection limit was 0.1 \( \mu \text{mol/L} \).

The ADMA/creatinine ratio was calculated, as ADMA (micromolar)/creatinine (micromolar) \( \times 10^{-3} \). For patients for whom multiple samples were drawn before and after HD sessions, to assess the different effects of HD treatment on methylargnine levels, as opposed to serum creatinine levels. Plasma nitrate was assayed as its pentafluorobenzyl derivative by gas chromatography-mass spectrometry, as described previously (14). The detection limit of the method was 20 fmol of nitrate or nitrate. Intra- and interassay variabilities were <3.8%. Serum creatinine concentrations were determined spectrophotometrically with the alkaline picric acid method, in an automatic analyzer (Beckman, Galway, Ireland). Protein concentrations were determined spectrophotometrically using the biuret method. All other
laboratory data were obtained from routine laboratory tests, using certified assay methods.

**Statistical Analyses**

Data are presented as mean ± SEM unless otherwise stated. Statistical significance was tested using ANOVA, followed by the Fisher protected least significant difference test for comparisons between patient groups. Repeated measurements were tested for statistical significance using ANOVA and the Scheffé F test. Statistical significance was accepted for P < 0.05.

**Results**

**Plasma l-Arginine, ADMA, SDMA, and Nitrate Concentrations in ESRD**

Compared with control subjects, HD-treated patients exhibited significantly higher plasma ADMA concentrations (P < 0.05) (Figure 1). Plasma SDMA levels were also significantly higher, but l-arginine concentrations were not significantly different between the two groups (Table 1). Plasma nitrate concentrations were significantly lower in HD-treated patients than in control subjects (Table 1).

ADMA levels in PD-treated patients were not different from those in control subjects (P > 0.05) (Figure 1). However, SDMA concentrations in the PD-treated group were significantly higher than those in control subjects (P < 0.05) (Table 1). l-Arginine and nitrate concentrations and the l-arginine/ADMA ratio were not significantly different, compared with those for the control group (P > 0.05) (Table 1).

HD-treated patients exhibited significantly higher mean ADMA concentrations, compared with PD-treated patients. SDMA, l-arginine, and nitrate concentrations did not differ significantly between these groups. The l-arginine/ADMA ratio in the HD-treated patients was significantly lower than that in the PD-treated patients (Table 1).

**Effects of HD on the Time Course of Plasma l-Arginine, DMA, and Nitrate Levels**

For eight HD-treated patients, the changes in the plasma levels of l-arginine, DMA, and nitrate were assessed before and after one HD session. One hour after the end of dialysis, no change in plasma ADMA and SDMA concentrations was observed. When the concentrations of these l-arginine analogs were expressed with respect to serum creatinine concentrations, there was a significant elevation 1 h after dialysis, compared with predialysis concentrations (ADMA, 0.86 ± 0.20 versus 2.02 ± 0.33; P < 0.05). Five hours after the end of the HD session, ADMA concentrations were significantly decreased, below values measured 1 h after dialysis. ADMA levels then continuously increased until 18 h after dialysis (Figure 2). A similar time course was observed for SDMA levels (Table 2). However, it seemed that the reduction in plasma SDMA levels at 5 h after dialysis was not as marked as that for ADMA. Plasma l-arginine concentrations remained unchanged after HD. Plasma nitrate levels and total protein concentrations did not change significantly.

**ADMA Concentrations with Respect to Renal Function and Atherosclerosis**

HD-treated patients with atherosclerosis exhibited significantly higher ADMA levels than did HD-treated patients without atherosclerosis (7.31 ± 0.33 μmol/L [n = 11]; P < 0.05) (Figure 3). Similarly, patients with peripheral arterial occlusive disease with normal renal function exhibited significantly higher ADMA levels than did healthy control subjects (3.23 ± 0.33 μmol/L [n = 33] versus 1.01 ± 0.05 μmol/L [n = 37]; P < 0.05) (Figure 3). There was no significant difference in ADMA levels between HD-treated patients without atherosclerosis and patients with peripheral arterial occlusive disease with normal renal function. A similar pattern of SDMA concentrations was observed (Table 3). Plasma l-arginine concentrations did not differ significantly between control subjects, patients with peripheral arterial occlusive disease, and patients with ESRD with atherosclerosis. However, patients with ESRD without atherosclerosis exhibited lower plasma l-arginine concentrations than did patients with ESRD with atherosclerotic disease.

**Table 1.** ADMA, SDMA, l-arginine, l-arginine/ADMA ratio, and nitrate concentration as determinants of nitric oxide synthesis in control subjects (control) compared with PD and HD patients

<table>
<thead>
<tr>
<th>Agent</th>
<th>Control (n = 37)</th>
<th>PD (n = 37)</th>
<th>HD (n = 43)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SDMA</td>
<td>0.8 ± 0.1</td>
<td>5.1 ± 0.6</td>
<td>5.2 ± 0.4</td>
</tr>
<tr>
<td>ADMA/SDMA</td>
<td>1.31 ± 0.21</td>
<td>0.72 ± 0.11</td>
<td>1.21 ± 0.09</td>
</tr>
<tr>
<td>l-arginine</td>
<td>75.5 ± 3.9</td>
<td>84.0 ± 9.3</td>
<td>75.9 ± 7.2</td>
</tr>
<tr>
<td>l-arg/ADMA</td>
<td>79.2 ± 5.0</td>
<td>68.9 ± 11.3</td>
<td>13.7 ± 1.4</td>
</tr>
<tr>
<td>Nitrate</td>
<td>39.1 ± 1.9</td>
<td>36.0 ± 1.5</td>
<td>23.9 ± 1.7</td>
</tr>
</tbody>
</table>

*Data are mean ± SEM. Results are given as μmol/L. ADMA, asymmetric dimethylarginine; DMA, symmetric dimethylarginine; PD, peritoneal dialysis; HD, hemodialysis.

b P < 0.05 versus control.

c P < 0.05 versus PD.
The major findings of this study are that: (1) ADMA and SDMA are accumulated in patients with ESRD; (2) ADMA levels are significantly lower in patients with ESRD treated with PD than in those treated with HD; (3) elevated ADMA levels are accompanied by low plasma levels of nitrate, the oxidative metabolite of NO; and (4) regardless of renal function, patients with atherosclerosis have higher plasma levels of ADMA than those without atherosclerosis.

**Accumulation of ADMA in ESRD**

Our finding that DMA levels are significantly higher in patients with ESRD than in healthy control subjects is consistent with the first description by Vallance et al. (7), who studied nine HD-treated patients. They reported a mean total DMA level of 8.7 ± 0.7 μmol/L. This concentration was six times higher than that measured for control subjects (1.2 ± 0.1 μmol/L) (7). In other studies, lower concentrations of DMA were reported. MacAllister et al. (15) found elevated ADMA and SDMA levels (0.9 ± 0.1 and 3.8 ± 0.4 μmol/L, respectively) in six HD-treated patients. Anderstam et al. (16) reported ADMA levels of 0.6 ± 0.2 μmol/L in 19 patients, and no difference between PD and HD. However, in these studies ADMA and SDMA levels in control subjects were also very low. The relative increase in DMA levels was 2.5- to sixfold in all of these studies. Comparing two treatment methods for patients with ESRD, i.e., HD and PD, we found that PD-treated patients exhibited lower plasma ADMA concentrations than did HD-treated patients. This difference may be caused by differences in dialytic clearance of ADMA with the two treatment methods or the metabolism of ADMA.

**Metabolism of ADMA**

The origin of ADMA in human plasma is currently unclear. Animal studies suggest that the relatively low levels of ADMA and SDMA present in plasma from healthy rabbits are derived mainly from the degradation of methylated proteins (6). Different methyltransferases seem to be responsible for L-arginine methylation in various tissues; the enzyme present in the vasculature mainly yields ADMA, as judged by the observation that cultured human endothelial cells release more ADMA than SDMA (17,18).

Urinary excretion is the main elimination route for SDMA in rabbits, whereas Nω-monomethyl-L-arginine and ADMA are partly eliminated by other metabolic pathways (6). In rats, Ogawa and coworkers (19) demonstrated that 14C-labeled ADMA is metabolized to citrulline by the enzyme DMA dimethylaminohydrolase (DDAH), which is present in various tissues of rats and human subjects (5,20,21). These tissues include the kidneys, the vasculature, and other tissues in which this enzyme is colocalized with isoforms of NO synthase and may affect NO-mediated cell function (20,22). DDAH presence and activity have also been demonstrated in human en-

**Table 2.** Time course of plasma L-arginine, SDMA, SDMA/creatinine ratio as determinants of nitric oxide synthesis in 10 HD patients before, 1, 5 and 18 h after a 4.5-h dialysis session

<table>
<thead>
<tr>
<th>Agent</th>
<th>Pre-HD (n = 8)</th>
<th>1 h Post-HD (n = 8)</th>
<th>5 h Post-HD (n = 8)</th>
<th>18 h Post-HD (n = 8)</th>
</tr>
</thead>
<tbody>
<tr>
<td>L-arginine (μmol/L)</td>
<td>69.44 ± 9.55</td>
<td>67.32 ± 12.78</td>
<td>51.50 ± 7.11</td>
<td>62.55 ± 7.60</td>
</tr>
<tr>
<td>SDMA (μmol/L)</td>
<td>3.97 ± 0.85</td>
<td>6.25 ± 0.63</td>
<td>4.55 ± 0.56</td>
<td>5.72 ± 0.62</td>
</tr>
<tr>
<td>SDMA/creatinine (×10⁻³)</td>
<td>0.60 ± 0.14</td>
<td>1.70 ± 0.26</td>
<td>0.99 ± 0.11</td>
<td>1.14 ± 0.13</td>
</tr>
<tr>
<td>Nitrate (μmol/L)</td>
<td>32.41 ± 2.68</td>
<td>40.51 ± 5.48</td>
<td>39.89 ± 4.26</td>
<td>35.61 ± 2.56</td>
</tr>
<tr>
<td>Protein (mg/ml)</td>
<td>64.08 ± 1.74</td>
<td>69.2 ± 1.91</td>
<td>69.96 ± 3.29</td>
<td>64.99 ± 2.59</td>
</tr>
</tbody>
</table>

* Data are mean ± SEM. Abbreviations as in Table 1.
inhibition of DDAH activity causes vasoconstriction, which is

Effects of ADMA on NO Elaboration

Several studies performed in vitro suggest that ADMA, at concentrations between 1 and 10 μmol/L, inhibits NO elaboration by NO synthase in the presence of L-arginine in isolated blood vessels, in cultured macrophages, and in cultured endothelial cells (8,9,17,18). Because intracellular ADMA levels have been shown to be approximately one order of magnitude greater than the levels in conditioned cell culture media, the accumulation of ADMA observed in patients with ESRD may be sufficient to cause clinically relevant inhibition of endothelial NO elaboration (17).

In 1992, Vallance et al. (7) hypothesized that inhibition of vascular NO formation caused by accumulated ADMA might be responsible for cardiovascular disorders such as hypertension and atherosclerosis, which are frequently observed in ESRD. Since then, the presence of dysfunctional endothelium-dependent vasodilation in this disease has been confirmed in several clinical investigations: Joannides et al. (24) showed that flow-induced, NO-dependent forearm vasodilation was impaired in adult patients receiving HD. Kari et al. (25) found impaired flow-induced, NO-dependent vasodilation in children with chronic renal failure. This was associated with elevated ADMA levels and reduced levels of nitrosothiols in plasma. Moreover, Hand et al. (26) demonstrated that the defect of endothelium-dependent vasodilation that was present in patients with ESRD before HD was reversed after HD sessions. Furthermore, L-arginine, but not D-arginine, restored endothelial function independently of HD. This study strongly supports the hypothesis that levels of ADMA present in the plasma of patients with ESRD induce clinically relevant inhibition of endothelial NO synthase activity, because competitive inhibition of NO synthase by ADMA can be overcome by excess L-arginine (17,27).

In this study we measured the plasma concentration of nitrate, a final oxidative metabolite of NO. Nitrate levels were significantly lower at baseline in HD-treated patients than in matched healthy control subjects. After HD, nitrate levels transiently increased and finally returned to baseline. These data are in accordance with studies performed by other investigators who also found evidence for reduced basal NO production in patients with ESRD (28) and increased NO forma-

Table 3. L-arginine and SDMA plasma concentrations in healthy controls, PAOD patients, and ESRD patients maintained on hemodialysis without or with concomitant atherosclerosis

<table>
<thead>
<tr>
<th>Agent</th>
<th>Control (n = 37)</th>
<th>PAOD (n = 33)</th>
<th>ESRD (n = 11)</th>
<th>ESRD + Atherosclerosis (n = 22)</th>
</tr>
</thead>
<tbody>
<tr>
<td>L-arginine</td>
<td>75.5 ± 3.9</td>
<td>78.6 ± 6.4</td>
<td>51.9 ± 8.6</td>
<td>98.9 ± 11.3</td>
</tr>
<tr>
<td>SDMA</td>
<td>0.8 ± 0.1</td>
<td>2.4 ± 0.5</td>
<td>3.8 ± 0.4</td>
<td>6.1 ± 0.5</td>
</tr>
</tbody>
</table>

* Data are mean ± SEM. Results are given as μmol/L. PAOD, peripheral arterial occlusive disease; ESRD, end-stage renal disease.
tion during HD (29,30). Using stable-isotope techniques, Rhodes et al. (31) recently found that in human subjects the major portion of circulating nitrite/nitrate is derived directly from the \( L \)-arginine/NO pathway. Although we cannot exclude the possibility that differences in dietary nitrate intake accounted at least in part for differences in plasma nitrate levels between patient groups in this study, accumulated evidence from these studies suggests that NO production is altered in ESRD. This is further supported by the recent finding of increased ADMA impaired endothelium-dependent, NO-mediated vasodilation ESRD. This is further supported by the recent finding of ADMA from these studies suggests that NO production is altered in ESRD. This is further supported by the recent finding of ADMA.

Consequences of Reduced NO Elaboration in ESRD

Aside from volume changes during HD, acute alterations in NO formation during HD may contribute to peridialytic BP instability in HD-treated patients (32–34). Changes in ADMA-induced inhibition of endothelial NO production may contribute to this phenomenon. In the long term, chronic reduction of NO synthase activity secondary to accumulation of ADMA may contribute to the pathogenesis of hypertension and atherosclerosis. Chronic inhibition of NO synthesis has been shown to produce hypertension (35) and to accelerate atherosclerosis in animal models (13,36,37). Chronically elevated ADMA concentrations may induce similar proatherogenic effects, like those observed in these experimental models (17).

Our group previously reported that plasma ADMA concentrations were elevated two- to threefold in patients with generalized atherosclerosis and peripheral arterial occlusive disease (10). This increase, which was associated with reduced urinary excretion of nitrate and cGMP, was independent of the presence of impaired renal excretory function. Moreover, we found a twofold elevation of ADMA levels in the plasma of asymptomatic young hypercholesterolemic subjects with normal renal function (38). We therefore investigated, in this study, whether the presence of atherosclerosis was also associated with increased ADMA levels in patients with ESRD. We found a similar relative elevation of ADMA levels in patients with ESRD and atherosclerosis, compared with patients with ESRD without atherosclerosis, and in patients with peripheral arterial occlusive disease with normal renal function, compared with control subjects. From these data it remains unresolved whether patients with ESRD and atherosclerosis exhibited higher ADMA levels because of the presence of atherosclerosis or whether they suffered from atherosclerosis as a consequence of higher ADMA levels. However, the elevation of ADMA levels in patients with peripheral arterial occlusive disease with normal renal function, compared with control subjects, suggests that mechanisms other than reduced renal excretion may also contribute to the greater accumulation of ADMA in patients with ESRD with peripheral arterial occlusive disease, compared with patients with ESRD without atherosclerosis. The concept of accelerated atherosclerosis in patients chronically undergoing dialysis has been widely accepted since it was first published (39). However, in a recent review, London and Druke (40) argued that the high mortality rates resulting from cardiovascular disease are not proof of accelerated atherosclerosis in patients undergoing maintenance dialysis. Many patients undergoing dialysis have more or less marked vascular lesions at the start of dialysis treatment, and the risk factors present in the predialysis phase may be of primary importance for the manifestation of cardiovascular disease. Multiple known cardiovascular risk factors have been shown to be present in patients with ESRD (41–44). Elevated ADMA levels may be a newly identified, preexistent risk factor for atherosclerosis.

In conclusion, we have demonstrated that ADMA is accumulated during chronic renal failure. Different dialysis treatment strategies affect ADMA levels differently. The presence of atherosclerosis is associated with higher ADMA levels in patients with normal renal function, as well as in patients undergoing dialysis, but this phenomenon may be unrelated to renal handling of ADMA. Reduced NO elaboration secondary to accumulation of ADMA may be an important pathogenic factor for atherosclerosis in chronic renal failure.

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