Asymmetric dimethyl arginine levels in chronic hemodialysis patients

Aydın ÇİFCİ1, Salih İNAL2*, Coşkun KAYA3

1Department of Internal Medicine, Kırıkkale Yüksek İhtisas Hospital, Kırıkkale, Turkey
2Department of Nephrology, Faculty of Medicine, Süleyman Demirel University, Isparta, Turkey
3Department of Nephrology, Faculty of Medicine, Ondokuz Mayıs University, Samsun, Turkey

* Correspondence: salihinal@yahoo.com

1. Introduction
Patients with chronic kidney disease (CKD) of any stage have significantly increased risk of accelerated atherosclerosis and increased cardiovascular mortality rate (1). In end-stage renal disease (ESRD) patients, more than half of all deaths are related to cardiac events and this rate is extremely higher than the rate among the general population (1). The high prevalence of cardiovascular mortality in this group of patients could not be easily explained by traditional risk factors such as diabetes, hypertension, and dyslipidemia. Recently, there have been considerable data supporting the idea that chronic inflammation is a nontraditional risk factor contributing to the accelerated atherosclerosis in CKD patients (2,3). Predicting the cardiovascular risk earlier in CKD patients is crucial in order to decrease the prevalence of cardiovascular events (4).

Endothelium-derived nitric oxide (NO) is a major regulator of vascular dilatation and is derived from arginine by nitric oxide synthase (5). Via the vasodilator effects, NO reduces both afterload and preload, resulting in prevention of left ventricular hypertrophy (6). Besides being an endothelial-based strong vasodilator, it also inhibits leukocyte adhesion to vascular walls, thrombocyte aggregation, and vascular smooth muscle cell proliferation (5). Asymmetric dimethyl arginine (ADMA) is an endogenous competitive inhibitor of endothelial-based NO synthase and by reducing NO production it may promote endothelial dysfunction, hypertension, and atherosclerosis (7).

ADMA is partially eliminated by the kidneys; therefore, renal failure increases its level (7). In previous studies, ADMA levels were found to be higher in ESRD patients both on peritoneal dialysis and hemodialysis (HD) compared with healthy controls (8,9). Increased levels of ADMA are thought to be associated with the increased frequency of cardiovascular events seen in ESRD patients (10,11), and it is suggested that ADMA might be a novel
cardiovascular risk factor for both ESRD patients and
the general population (7,12). Additionally, its relation
with nutritional status in maintenance HD patients was
assessed in a recent trial and a negative correlation was
found between ADMA levels and nutritional parameters
(13).

The objective of our study was to investigate serum
ADMA levels in patients with ESRD who were on chronic
HD treatment and to assess their potential correlations
with C-reactive protein (CRP), albumin, and cholesterol
levels as the established cardiovascular and nutritional
parameters.

2. Materials and methods
This cross-sectional, single-center study enrolled
maintenance HD patients and healthy control subjects.
Seventy-six prevalent HD patients treated at our institute
were screened. Exclusion criteria were history or existence
of malignancy, chronic liver diseases, autoimmune
diseases, symptoms of active infection or inflammation,
hepatitis B and C virus infection, and steroid treatment.
Forty-nine adult HD patients (18–80 years of age) who
fulfilled the inclusion criteria were recruited into the study.
The study protocol was approved by the Medical Ethics
Committee of Ankara Education and Research Hospital
and was performed in accordance with the principles of
the Helsinki Declaration. Informed consent was obtained
from all participants.

Forty-nine patients were on maintenance HD treatment
3 times weekly for 4 h with standard bicarbonate dialysate
and with synthetic membranes. Twenty-two healthy
volunteers, with similar age and sex characteristics and
without any clinical signs of chronic disease or laboratory
abnormality were recruited as controls from subjects who
were admitted to our hospital for routine examination.
Demographic and clinical data were recorded for all
subjects and body mass index (BMI) was calculated as
the ratio of weight (kg) to the square of height (m). Blood
samples were drawn after 12 h of fasting, once after the
weekend. Serum albumin, CRP, creatinine, calcium,
phosphate, total cholesterol, low-density lipoprotein (LDL)
cholesterol, high-density lipoprotein (HDL) cholesterol,
hemoglobin, and white blood cell counts were measured
by using standard methods in a routine biochemistry
laboratory. Serum samples that were separated for ADMA
measurement were stored at –70 °C for a short period of
time, and the tests were performed by the enzyme-linked
immunosorbent assay (ELISA) method (ADMA ELISA
Kit, Immundiagnostik, Bensheim, Germany).

2.1. Statistical analysis
Data were presented as means ± standard deviations.
Comparisons of numeric variables between groups were
performed by Student’s t-test or the Mann–Whitney U
test, as appropriate. Categorical variables were compared
using the chi-squared test and were shown as frequency
and percentages. Univariate correlation was established by
using either Pearson's or Spearman's correlation coefficient.
All statistical calculations were performed using SPSS 13.
All P-values were calculated as 2-sided, and P < 0.05 was
considered significant.

3. Results
The present study involved 49 prevalent HD patients and 22
age- and sex-matched controls. Mean age was 55.3 ± 14.2
years for HD patients and 49.3 ± 15.0 years for the control
group (P = 0.11). The patients were on HD treatment for
64.1 ± 4.5 months on average. Primary causes of ESRD
were diabetic nephropathy (n = 21, 42.8%), hypertension
(n = 14, 28.6%), chronic glomerulonephritis (n = 8, 16.3%),
and unknown etiology (n = 6, 12.2%). The anthropometric,
demographic, and biochemical parameters as compared
between the control group and the HD patients are given
in Table 1. HD patients had significantly higher ADMA
levels compared with healthy controls (0.51 ± 0.25 vs.
0.35 ± 0.15, P = 0.002). While white blood cell counts and
BMI values were similar between the 2 groups, CRP and
LDL cholesterol were found to be significantly higher and
albumin and HDL cholesterol significantly lower in HD
patients compared with healthy controls.

ADMA concentrations were positively correlated with
mean age (P = 0.02, r = 0.360), LDL cholesterol levels (P =
0.006, r = 0.325), and serum CRP levels (P = 0.02, r = 0.268).
Additionally, there was a significant negative correlation
between ADMA concentrations and serum albumin levels
(P = 0.005, r = –0.331). Table 2 shows the correlations of
ADMA with clinical and laboratory parameters and the
Figure illustrates these correlations.

4. Discussion
In our study, serum ADMA levels of chronic HD patients
were found to be significantly higher than those of healthy
controls. This result is consistent with the previous similar
studies in the literature regarding ADMA levels in ESRD
patients (9,14,15). Circulating levels of ADMA had a
positive correlation with levels of CRP. This finding might
suggest that a connection exists between endothelial
dysfunction and inflammation in ESRD patients. Recently
some studies pointed out the association of ADMA
levels with biomarkers of inflammation in some chronic
conditions like familial Mediterranean fever, inflammatory
bowel disease, and chronic kidney disease (16–18).

Additionally, serum ADMA levels were inversely
correlated with serum albumin levels in the present study.
As serum albumin level is a marker of nutritional status,
we think that this result suggests an association between
nutrition and inflammation in ESRD patients. This result
is consistent with the findings of a recent trial studying nutritional status in HD patients (13). Unlike the results of similar studies, we could not find any significant relation between ADMA and BMI (13,19). This may be due to the small number of patients in our study.

Findings on the relation between circulating ADMA concentrations and serum cholesterol levels are conflicting. In one study, ADMA levels were found to be higher in patients with metabolic syndrome compared with normal subjects, but no correlation was found between ADMA and cholesterol levels (20). In another study of ESRD patients, ADMA showed significant correlation with serum cholesterol levels (19). Consistent with the latter report, in our study ADMA showed a positive correlation with serum LDL cholesterol levels. LDL cholesterol may have a role in the pathway of the atherosclerotic process of ADMA.

It is reported that there is a positive and significant correlation between ADMA levels and carotid intima media thickness in chronic HD patients (21). In another study, inflammation and endothelial dysfunction as assessed by circulating levels of ADMA were found to be an independent factor for explaining all-cause mortality and cardiovascular events in ESRD patients (18). Similar

<table>
<thead>
<tr>
<th>Variable</th>
<th>HD patients (n = 49)</th>
<th>Healthy controls (n = 22)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean age (years)</td>
<td>55.3 ± 14.2</td>
<td>49.3 ± 15.0</td>
<td>0.11</td>
</tr>
<tr>
<td>Sex (male/female)</td>
<td>26/23</td>
<td>7/15</td>
<td>0.09</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>24.7 ± 3.8</td>
<td>25.6 ± 3.6</td>
<td>0.32</td>
</tr>
<tr>
<td>Time on dialysis (months)</td>
<td>64.1 ± 4.5</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Creatinine (mg/dL)</td>
<td>7.50 ± 1.78</td>
<td>0.79 ± 0.25</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Albumin (g/dL)</td>
<td>3.6 ± 0.3</td>
<td>4.2 ± 0.3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hemoglobin (g/dL)</td>
<td>10.8 ± 1.5</td>
<td>13.4 ± 1.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>WBC (U/L)</td>
<td>6810 ± 2019</td>
<td>6787 ± 1384</td>
<td>0.96</td>
</tr>
<tr>
<td>LDL cholesterol (mg/dL)</td>
<td>147.7 ± 24.3</td>
<td>132.8 ± 23.7</td>
<td>0.02</td>
</tr>
<tr>
<td>HDL cholesterol (mg/dL)</td>
<td>31.1 ± 6.2</td>
<td>46.1 ± 13.4</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>ADMA (µmol/L)</td>
<td>0.51 ± 0.25</td>
<td>0.35 ± 0.15</td>
<td>0.002</td>
</tr>
<tr>
<td>CRP (mg/dL)</td>
<td>15.2 ± 17.2</td>
<td>2.7 ± 1.2</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

HD: Hemodialysis, WBC: white blood cell count, LDL: low-density lipoprotein, HDL: high-density lipoprotein, ADMA: asymmetric dimethyl arginine, CRP: C-reactive protein.

<table>
<thead>
<tr>
<th>Variable</th>
<th>P-value</th>
<th>r</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean age</td>
<td>0.02</td>
<td>0.360</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>0.64</td>
<td>0.056</td>
</tr>
<tr>
<td>Time on dialysis (months)</td>
<td>0.67</td>
<td>−0.062</td>
</tr>
<tr>
<td>Creatinine (mg/dL)</td>
<td>0.07</td>
<td>0.210</td>
</tr>
<tr>
<td>Albumin (g/dL)</td>
<td>0.005</td>
<td>−0.331</td>
</tr>
<tr>
<td>Hemoglobin (g/dL)</td>
<td>0.06</td>
<td>−0.220</td>
</tr>
<tr>
<td>WBC (U/L)</td>
<td>0.17</td>
<td>−0.162</td>
</tr>
<tr>
<td>LDL cholesterol (mg/dL)</td>
<td>0.006</td>
<td>0.325</td>
</tr>
<tr>
<td>HDL cholesterol (mg/dL)</td>
<td>0.10</td>
<td>−0.197</td>
</tr>
<tr>
<td>CRP (mg/dL)</td>
<td>0.02</td>
<td>0.268</td>
</tr>
</tbody>
</table>

ADMA: Asymmetric dimethyl arginine, WBC: white blood cell count, LDL: low-density lipoprotein, HDL: high-density lipoprotein, CRP: C-reactive protein.
to this finding, a recent study also reported that ADMA level is a death predictor in ESRD patients (22). These results strengthen the hypothesis that interventions aimed at preventing endothelial dysfunction and inflammation may decrease the high cardiovascular risk for these patients. However, it is still unknown whether lowering the ADMA levels would be of any clinical benefit. The limitations of our study are the cross-sectional design and small sample size. Additionally, some anthropometric measurements and clinical parameters representing nutritional status could be added to the study design. A prospective study with a larger sample size would be more helpful for this issue.

In conclusion, in the present study we have found significantly higher serum ADMA levels in chronic HD patients compared with healthy individuals. Our results suggest that ADMA might be a potential novel biomarker of inflammation and atherosclerosis in patients on chronic HD. Measuring ADMA levels may promote awareness of early cardiovascular risk increase in HD patients.

References


