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# Acute Effects of Hemodiafiltration Versus Conventional Hemodialysis on Endothelial Function and Inflammation

## A Randomized Crossover Study

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**Abstract:** Endothelial dysfunction and chronic inflammatory process are prevalent in patients with end-stage renal disease (ESRD) on maintenance hemodialysis (HD). The aim of this study was to evaluate the acute and short-term effects of online hemodiafiltration (OL-HDF) versus conventional HD on endothelial function and inflammation.

A prospective, randomized, crossover trial.

Twenty stable ESRD patients undergoing chronic HD treatments were randomly assigned with a 1:1 ratio to conventional HD and to OL-HDF both for 2 weeks (either HD followed by OL-HDF or OL-HDF followed by HD). Markers of endothelial dysfunction such as flow-mediated dilatation (FMD) of the brachial artery, soluble endothelial protein C receptor (sEPCR), and soluble thrombomodulin (sTM) were measured at baseline, after the first dialysis session and after 2 weeks. Meanwhile, serum interleukin 6 (IL-6) and high-sensitivity C-reactive protein (hs-CRP) levels were measured as well.

Both a single OL-HDF session and 2-week OL-HDF significantly improved brachial FMD% ( $18.7 \pm 6.9\%$  at baseline;  $21.5 \pm 5.4\%$  after the first dialysis;  $21.5 \pm 5.7\%$  after 2 weeks;  $P < 0.05$  vs baseline), decreased the levels of sEPCR (from  $394.4 [297.9-457.0]$  ng/ml at baseline to  $234.7 [174.1-345.5]$  ng/ml after the first dialysis, and to  $191.5 [138.2-255.0]$  ng/ml after 2 weeks;  $P < 0.01$  vs baseline) and sTM. In contrast, HD did not change FMD%, even increased the levels of sEPCR and sTM. A reduction in IL-6 level was observed in OL-HDF patients after 2-week dialysis, while IL-6 did not change in HD patients. There was no significant difference in change of hs-CRP level between the OL-HDF and HD treatments.

OL-HDF has both acute and short-term beneficial effects on endothelial dysfunction compared to conventional HD.

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**Abbreviations:** ESRD = end-stage renal disease, FMD = flow-mediated dilatation, HD = hemodialysis, hs-CRP = high-sensitivity C-reactive protein, IL-6 = interleukin 6, NMD = nitroglycerin-mediated dilation, OL-HDF = online hemodiafiltration, sEPCR = soluble endothelial protein C receptor, sTM = soluble thrombomodulin.

## INTRODUCTION

There is an increased incidence and worsening of atherosclerosis in patients on maintenance hemodialysis (HD),<sup>1</sup> which may contribute to the high risk of cardiovascular morbidity and mortality. As an early initiating event in atherosclerosis, endothelial dysfunction is almost universal in patients with end-stage renal disease (ESRD). It is probably caused by uremic toxin retention, inflammation, oxidative stress, and the increased fluid shear stress during dialysis sessions.<sup>2,3</sup> Various endothelial dysfunction markers and methods have been described, such as brachial flow-mediated dilatation (FMD), high-sensitive C-reactive protein (hs-CRP), circulating endothelial cells and progenitor cells.<sup>3-6</sup> Moreover, thrombomodulin (TM), a transmembrane cofactor of the endothelial cells, and endothelial protein C receptor (EPCR), an important regulator of the protein C anticoagulant pathway, which are abundantly produced by endothelial cells during injury, have been defined as new markers of endothelial dysfunction in many disease.<sup>6,7</sup> The soluble forms of these 2 markers significantly increased in patients on chronic HD.<sup>8</sup> In addition, various proinflammatory cytokines are markedly increased in HD patients, and microinflammation induces endothelial cell injury.<sup>3</sup> It appears that HD may induce endothelial dysfunction and chronic HD treatment could predispose patients to vascular disorders.<sup>8</sup>

Conventional HD uses diffusion, the removal of solutes and water across a semipermeable membrane down a concentration gradient, which provides diffusive clearance of low-molecular-weight solutes with limited ability to remove middle-sized solutes.<sup>9</sup> While, hemodiafiltration (HDF) is a combination of convective and diffusive processes for solute removal. Several studies have suggested that HDF are associated with better removal of both small and middle molecules, such as  $\beta_2$ -microglobulin ( $\beta_2$ -MG),<sup>10,11</sup> more advantageous effects on hemodynamic stability,<sup>12</sup> improved survival,<sup>13,14</sup> when compared with conventional HD. In addition, long-term online hemodiafiltration (OL-HDF) with ultrapure dialysate seems to reduce inflammatory activity over time compared to HD.<sup>15</sup> Bellien et al<sup>5</sup> also showed that prolonged HDF significantly improved FMD,

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decreased blood TNF- $\alpha$  mRNA expression in ESRD patients. However, no studies have evaluated the acute effects of OL-HDF on endothelial dysfunction and inflammation in maintenance HD patients. The aim of this study is to assess whether a single or short-term OL-HDF sessions would ameliorate endothelial dysfunction and inflammation compared with conventional HD in these patients.

## METHODS

### Study Design and Patients

This was a randomized, single-blinded, crossover trial designed to compare the acute and short effects of OL-HDF versus conventional HD on endothelial dysfunction and inflammation. All enrolled patients were randomly assigned by concealed allocation with a 1:1 ratio to conventional HD and to OL-HDF both for 2 weeks (thrice a week, either HD followed by OL-HDF or OL-HDF followed by HD). Patients were blinded to treatment order. The study adhered to the Declaration of Helsinki and was approved by the Ethical Committee of Fudan University. All participants provided written informed consent.

Participant flow is shown in Figure 1. Potential participants were screened prospectively using consecutive sampling. Of 185 ESRD patients, who initiated routine HD treatment in Zhongshan Hospital from February 2011 to December 2012, 65 potential patients met the initial inclusion criteria: aged 18 to 80 years, arteriovenous (AV) fistula as vascular access, low-flux HD thrice a week, absence of following ill conditions: diabetes mellitus, liver disease, amyloidosis, vasculitis, severe hyperlipidemia, and cancer. After 6 months of HD treatment, 20 patients were enrolled into the study on the basis of the final inclusion and exclusion criteria. Inclusion criteria included patients being on low-flux HD thrice a week for 6 months, with well-functioning native AV fistulas for blood access, hemoglobin level ranging between 10 and 13 g/dL in the previous 3 months, stable with a minimum dialysis single-pool Kt/V for urea  $\geq 1.2$ . Exclusion criteria included any acute illness such as infection or cardiovascular event (i.e., acute myocardial infarction) or cerebrovascular (i.e., ischemic stroke) event within the past month, severe hypertension (systolic blood

pressure  $> 180$  or diastolic blood pressure  $> 110$  mm Hg), current estrogen/hormone replacement therapy or intravenous iron therapy or oral antiplatelet drugs (such as aspirin, etc.), life expectancy  $< 3$  months because of nonrenal disease. All participants underwent a thorough physical examination with documentation of medical history and medications before study.

### Dialysis Protocol

All participants received thrice-weekly dialysis. They were performed with ultrapure dialysis fluids, defined as  $< 0.1$  colony-forming units/ml and  $< 0.03$  endotoxin units/ml. 1.4 m<sup>2</sup> low-flux polyethersulfone dialyzers (BLS514SD; Sorin Group Italia, Mirandola, Italy) were used for conventional HD. 1.6 m<sup>2</sup> high-flux polyethersulfone hemodiafilters (BLS816SD; Bellco, Italia) were used for OL-HDF. OL-HDF was performed in the postdilution mode, with substitution flow 70 to 80 mL/min, and the total substitution volume 12 to 19 L. Blood flow and dialysate flow rates were set at 250 to 300 mL/min and 600 mL/min respectively, and dialysis time was 240 minutes in all study sessions.

### Blood Sampling and Laboratory Measurements

On the first and the last day of each treatment (OL-HDF, conventional HD), blood samples were obtained from the arterial port of the vascular access immediately before the onset of dialysis and again at 240 minutes during the dialysis sessions. Plasma and serum were collected respectively, for biochemical parameters and other assays. Samples were aliquoted and stored at  $-80^{\circ}\text{C}$  before assay.

As inflammatory markers, hs-CRP was measured by a modification of the laser nephelometric technique (Behring Diagnostics, GmbH, Rarburg, Germany), and interleukin 6 (IL-6) was measured by enzyme-linked immunosorbent assay (ELISA) using commercially available kits (R&D Systems, Minneapolis, MN).

### Determination of Endothelial Function and Markers of Endothelial Injury

With high-resolution ultrasonography, endothelial function was determined noninvasively as the percentage of

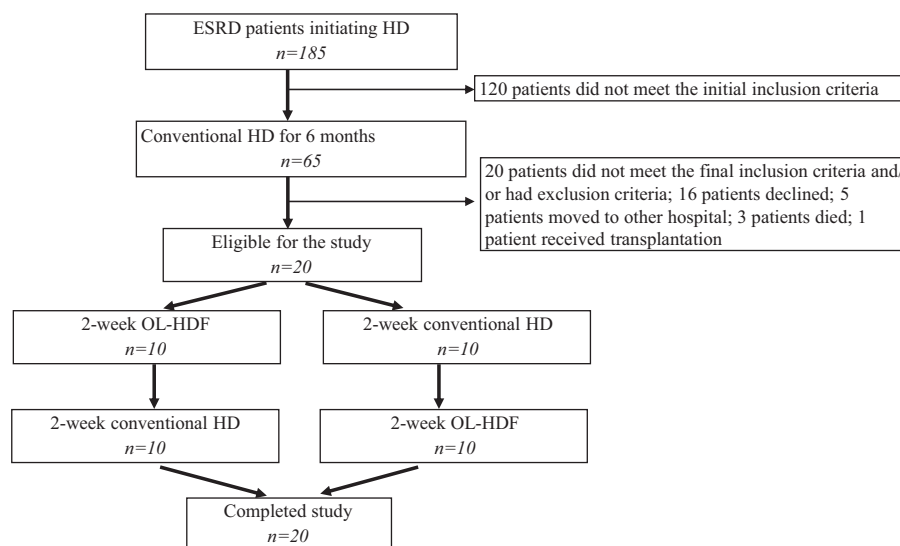


FIGURE 1. Participant flowchart of this study. ESRD = end-stage renal disease, HD = hemodialysis, OL-HDF = online hemodiafiltration.

flow-mediated dilatation (FMD%) of the brachial artery in the nonfistula arm, which was described previously.<sup>4</sup> In brief, brachial artery internal diameter was measured at end-diastole. After basal diameter was recorded, an arterial occlusion cuff was inflated to 200 mm Hg for 5 minutes to create forearm ischemia, and subsequently, deflated to allow reactive hyperemia. Brachial arterial diameter was measured 60 seconds after deflation. FMD was calculated as the percentage change in artery diameter, with the basal diameter before cuff inflation and the peak diameter after cuff deflation. After return to the baseline value, brachial artery nitroglycerin-mediated dilation (NMD), also called as endothelium-independent dilatation, was assessed by measuring changes in artery diameter after sublingual nitroglycerin administration (0.5 mg). FMD% and NMD% were measured before and after the first dialysis session, as well as after the last dialysis session.

As markers of endothelial cell injury, soluble endothelial protein C receptor (sEPCR) and soluble thrombomodulin (sTM) levels in plasma were evaluated using ELISA kits (Quantikine<sup>®</sup> Human EPCR Immunoassay and Quantikine<sup>®</sup> Human Thrombomodulin/BDCA-3 Immunoassay; R&D Systems), according to manufacturer's instructions.

### Statistical Analyses

Data were expressed as mean  $\pm$  standard deviation (SD), median (interquartile range), or percentage, when appropriate. Levels of FMD%, sEPCR, sTM, IL-6, and hs-CRP within OL-HDF and HD patients at each study visit (first dialysis session and 2-week dialysis session) were compared with baseline using paired *t* tests for normally distributed and signed rank test for skewed parameters. Between-group comparisons (patients characteristics at baseline in HD-then-OL-HDF group vs OL-HDF-then-HD group) were analyzed using *t* tests for continuous variables or  $\chi^2$  tests for dichotomous variables.

The differences in change in FMD%, sEPCR, sTM, IL-6, and hs-CRP levels from baseline to first dialysis session and to 2 weeks between the 2 treatment modalities were evaluated using a linear regression model. The natural logarithms (Ln) of sEPCR, sTM, IL-6, and hs-CRP (LnsEPCR, LnsTM, LnIL-6, and Lnhs-CRP) were used for the analyses to improve the fit of the model. A 2-sided  $P < 0.05$  was considered statistically significant. Statistical analyses were performed with SPSS 16.0 (SPSS, Inc., Chicago, IL).

## RESULTS

### Patients

A total of 20 participants completed the study (Figure 1). The baseline characteristics and laboratory data of patients included are listed in Table 1. Thirteen of the 20 patients were men. Mean age was  $49.2 \pm 13.3$  (mean  $\pm$  standard deviation) years. Primary diagnoses of ESRD were: primary glomerulonephritis ( $n = 9$ ), hypertension ( $n = 3$ ), cystic kidney disease ( $n = 2$ ), obstructive nephropathy ( $n = 2$ ), chronic pyelonephritis ( $n = 1$ ), unknown ( $n = 3$ ). Antihypertension drugs were used in 18 patients: calcium channel blockers ( $n = 12$ ), angiotensin converting enzyme (ACE) inhibitors ( $n = 8$ ), and angiotensin receptor blockers ( $n = 5$ ),  $\beta$ -blockers ( $n = 7$ ),  $\alpha$ -blockers ( $n = 7$ ). In addition, erythropoietin was used in 17 patients, and their prescription was unaltered during the whole study. Overall, baseline characteristics and biochemistry were well balanced between the randomizing subgroups (HD-then-OL-HDF and OL-HDF-then-HD).

### Effects of OL-HDF Versus Conventional HD on Endothelial Function

The flow-mediated dilatation (FMD%) of the brachial artery increased after the first dialysis session, as well as after 2-week dialysis, in patients on OL-HDF treatment (baseline,  $18.7 \pm 6.9\%$ ; first dialysis,  $21.5 \pm 5.4\%$ ; 2-week dialysis,  $21.5 \pm 5.7\%$ ;  $P < 0.05$  vs baseline). While, patients on conventional HD did not show an improvement in FMD% during the corresponding dialysis sessions (baseline,  $17.8 \pm 6.3$ ; first dialysis,  $18.6 \pm 7.3\%$ ; 2-week dialysis,  $17.4 \pm 8.9\%$ ). The difference in change in FMD% from baseline to 2 weeks between the 2 treatment modalities was statistically significant ( $P < 0.01$ ; Table 2).

We also evaluated the markers of endothelial cell injury, sEPCR and sTM, during the 2 treatments (Table 3). sEPCR concentration markedly decreased from 394.4 (297.9–457.0) ng/ml at baseline to 234.7 (174.1–345.5) ng/ml after the first dialysis, and to 191.5 (138.2–255.0) ng/ml after 2 weeks in OL-HDF patients ( $P < 0.01$  vs baseline), while slightly increased from 350.0 (285.2–464.0) ng/ml at baseline to 411.8 (320.5–516.3) ng/ml after the first dialysis and to 443.6 (327.9–625.5) ng/ml after 2 weeks in conventional HD patients, with the difference of change between the 2 treatment groups being statistically significant ( $P < 0.01$ ). A similar trend was found for sTM (Table 2).

### Effects on Inflammation

A reduction in plasma IL-6 level was observed in OL-HDF patients after 2-week dialysis sessions (3.41 [0.94–9.54] pg/ml vs 5.60 [2.40–12.95] pg/ml at baseline;  $P < 0.05$ ), while, in HD patients, the IL-6 level did not change significantly (Table 3). From a linear regression analysis, OL-HDF seemed to decrease the IL-6 level during 2-week dialysis sessions when compared to conventional HD treatment ( $P < 0.01$ ). In addition, patients on HD showed a significant increase in hs-CRP level after a single dialysis session, as well as after 2-week dialysis (baseline, 1.35 (0.50–4.23) mg/L; first dialysis, 1.60 (0.63–5.00) mg/L; 2-week dialysis, 3.70 (0.83–8.02) mg/L;  $P < 0.05$  vs baseline). Hs-CRP level in OL-HDF patients remained relatively stable after 2-week dialysis sessions (baseline, 2.05 (0.60–5.55) mg/L; 2-week dialysis, 1.90 (0.90–10.40) mg/L;  $P > 0.05$ ). While, there were no significant differences in change of hs-CRP level between the HD patients and OL-HDF patients ( $P = 0.16$ ).

## DISCUSSION

Endothelial dysfunction is common in ESRD, linked to a variety of disease states, including atherosclerosis, diabetes mellitus, coronary artery disease, and hypertension.<sup>16</sup> As a marker of endothelial function, brachial FMD in HD patients were lower than healthy population.<sup>17</sup> Both sEPCR and sTM, considered as markers of endothelial cell injury, significantly increased during HD procedures.<sup>8</sup> In this randomized crossover study, we demonstrated that a single OL-HDF session or 2-week OL-HDF could significantly improve brachial FMD%, decreased the levels of sEPCR and sTM in maintenance HD patients as compared with conventional HD. It seemed to suggest that short-term OL-HDF might reduce endothelial cell injury and improve endothelial function.

Several studies indicated the advantageous effects of HDF.<sup>5,14,18</sup> Hemodiafiltration may provide hemodynamic and survival benefits above low- and high-flux haemodialysis.<sup>19</sup> Maduell et al<sup>14</sup> evaluated the effects of OL-HDF versus HD on

**TABLE 1.** Baseline Characteristics of Participants

	All Participants (n = 20)	HD-Then-OL-HDF (n = 10)	OL-HDF-Then-HD (n = 10)
Male sex	13 (65)	7 (70)	6 (60)
Age, y	49.2 ± 13.3	50.2 ± 10.9	48.1 ± 15.8
Body mass index, kg/m <sup>2</sup>	21.4 ± 3.4	21.6 ± 4.4	21.3 ± 2.2
Systolic blood pressure, mm Hg	134.1 ± 11.0	134.6 ± 12.7	133.6 ± 9.8
Diastolic blood pressure, mm Hg	83.0 ± 9.9	81.4 ± 10.0	82.4 ± 10.8
Laboratory parameters			
Albumin, g/dl	41.4 ± 2.6	40.3 ± 2.3	42.4 ± 2.7
ALT, U/L	12.3 ± 6.0	10.7 ± 5.1	13.9 ± 6.6
AST, U/L	14.2 ± 5.0	13.2 ± 3.0	15.2 ± 6.5
FBG, mmol/L	4.9 ± 0.5	5.0 ± 0.5	4.9 ± 0.5
Total cholesterol, mmol/L	4.6 ± 0.9	4.4 ± 0.8	4.9 ± 0.9
Triglyceride, mmol/L	1.6 ± 0.9	1.5 ± 1.0	1.8 ± 0.8
HDL-ch, mmol/L	1.1 ± 0.4	1.1 ± 0.4	1.1 ± 0.4
LDL-ch, mmol/L	2.8 ± 0.7	2.6 ± 0.7	2.9 ± 0.7
Serum creatinine, mg/dl	13.3 ± 3.1	12.1 ± 2.6	14.6 ± 3.2
Blood urea nitrogen, mg/dl	89.4 ± 22.4	84.4 ± 24.4	94.4 ± 20.4
Parathyroid hormone, pg/ml	368.1 ± 121.1	385.4 ± 130.0	350.9 ± 115.7
Serum phosphorus, mg/dl	7.7 ± 1.9	7.8 ± 2.2	7.7 ± 1.5
Serum calcium, mg/dl	9.4 ± 0.8	9.3 ± 1.2	9.6 ± 0.8
Hemoglobin, g/dl	11.2 ± 1.2	11.0 ± 0.8	11.4 ± 1.5
Single-pool Kt/V	1.3 ± 0.2	1.3 ± 0.2	1.3 ± 0.2
Antihypertension medications			
Either ACEI or ARB or both	10 (50)	4 (40)	6 (60)
Calcium channel blocker	14 (70)	6 (60)	8 (80)
β-blocker	7 (35)	2 (20)	5 (50)
α-blocker	7 (35)	4 (40)	3 (30)
Other medications			
EPO	17 (85)	8 (80)	9 (90)
EPO dose, U/wk	5500 (3000–10000)	5000 (3000–10000)	6000 (4500–10000)

Data for categorical variables are given as number (percentage); data for continuous variables are given as mean ± standard deviation or median (interquartile range).

ACEI = angiotensin-converting enzyme inhibitor, ALT = alanine aminotransferase, ARB = angiotensin receptor blocker, AST = aspartate aminotransferase, BMI = body mass index, DBP = diastolic blood pressure, EPO = erythropoietin, FBG = fasting blood-glucose, HD = hemodialysis, HDL-ch = high-density lipoprotein cholesterol, LDL-ch = low-density lipoprotein cholesterol, OL-HDF = online hemodiafiltration, SBP = systolic blood pressure.

all-cause mortality and cardiovascular events in chronic HD patients, and found patients who switched to high-efficiency postdilution OL-HDF had a 30% lower risk of all-cause mortality and a 33% lower risk of cardiovascular mortality during 36-month follow-up, as compared with patients who continued on HD. Recently, Bellien et al<sup>5</sup> also demonstrated that high-efficiency OL-HDF decreased uremic toxins and vascular inflammation in chronic HD patients, and was associated with subsequent improvement in endothelial NO-synthase (eNOS) functionality which played a pivotal role in maintaining the integrity of endothelial cells and endothelial function.<sup>20</sup>

Systemic inflammation is commonly observed in patients with ESRD.<sup>21,22</sup> Most circulating acute-phase proteins and proinflammatory cytokines are increased in HD patients when compared with healthy controls,<sup>23</sup> and higher levels of these factors are associated with high risk of cardiovascular events and mortality.<sup>24–26</sup> Previous studies indicated the intradialysis course of CRP levels during HD, yielding different results, with some studies reporting no change,<sup>27,28</sup> whereas some reporting an increase.<sup>29</sup> den Hoedt et al<sup>15</sup> reported CRP and IL-6 concentrations increased in patients treated with HD, and remained stable in patients treated with hemodiafiltration, and proposed

that long-term hemodiafiltration with ultrapure dialysate might reduce inflammatory activity over time. In this study, we observed an increase in hs-CRP level after a single and short-term HD sessions, however, in OL-HDF patients, the level of hs-CRP remained relatively stable after 2-week dialysis. In addition, we also found a decrease in IL-6 level in OL-HDF patients, while no change in HD patients, after 2-week dialysis sessions. These data seemed to suggest a beneficial effect of short-term OL-HDF in amelioration of inflammation.

To our knowledge, this study is the first to evaluate the acute effect of OL-HDF on endothelial function in chronic HD patients. However, there were several limitations. First, our study population was relatively small. We performed a randomized crossover design to increase the power of the study. Second, it was still unclear as to whether acute changes in middle molecules were due to increased clearance or membrane adsorption. Third, a carryover effect of randomization arm (HD-then HDF vs HDF-then-HD) was not considered, which could limit the power to detect significant changes in some parameters assessed. Inrig et al<sup>30</sup> found a carryover effect in a randomized crossover trial testing the effect of low versus high dialysate sodium on patients during HD. Likewise, a carryover effect

**TABLE 2.** Effects of HD Versus OL-HDF on Endothelial Function and Endothelial Cell Injury

	Baseline	First Dialysis	2-Week Dialysis	Change in First dialysis vs Baseline	Change in 2-Week Dialysis vs Baseline	P-value OL-HDF vs HD
FMD (%)						
HD	17.8 ± 6.3	18.6 ± 7.3	17.4 ± 8.9	0.7 ± 5.1	-0.4 ± 8.7	0.02
OL-HDF	18.7 ± 6.9	21.5 ± 5.4	21.5 ± 5.7	2.8 ± 4.7*	2.8 ± 4.9*	
Difference OL-HDF vs HD	0.9 ± 6.0	3.0 ± 6.2	4.1 ± 6.2			
NMD (%)						
HD	16.9 ± 7.0	17.0 ± 7.2	16.1 ± 6.0	0.2 ± 2.5	-0.8 ± 4.3	0.44
OL-HDF	16.5 ± 6.4	17.2 ± 5.6	17.6 ± 7.1	0.7 ± 3.5	1.1 ± 4.6	
Difference OL-HDF vs HD	-0.4 ± 4.1	0.2 ± 4.5	1.5 ± 5.0			
sEPCR (ng/ml)						
HD	350.0 (285.2 to 464.0)	411.8 (320.5 to 516.3)	443.6 (327.9 to 625.5)	11.5 (-39.1 to 88.7)	47.7 (-20.2 to 147.6)*	< 0.001
OL-HDF	394.4 (297.9 to 457.0)	234.7 (174.1 to 345.5)	191.5 (138.2 to 255.0)	-122.9 (-225.1 to 88.0)**	-200.8 (-281.4 to 118.9)**	
Difference OL-HDF vs HD	7.0 (-38.0 to 56.7)	-146.1 (-213.7 to 83.5)	-235.2 (-300.2 to 151.4)			
sTM (ng/ml)						
HD	25.9 (20.7 to 28.7)	29.6 (24.4 to 35.1)	32.0 (24.9 to 37.4)	6.9 (1.5 to 9.7)**	6.8 (-1.0 to 10.0)**	< 0.001
OL-HDF	26.1 (20.7 to 28.4)	24.5 (20.4 to 27.0)	21.5 (17.7 to 28.5)	-1.0 (-2.4 to 2.4)	-1.5 (-5.5 to 11.0)*	
Difference OL-HDF vs HD	-0.6 (-3.8 to 3.0)	-5.3 (-9.2 to 1.7)	-8.9 (-14.0 to 3.3)			

Values expressed as mean ± standard deviation or median (interquartile range).

P-value OL-HDF versus HD from a linear regression model.

FMD = flow-mediated dilatation, HD = hemodialysis, NMD = nitroglycerin-mediated dilatation, OL-HDF = online hemodiafiltration, sEPCR = soluble endothelial protein C receptor, sTM = soluble thrombomodulin.

\*P < 0.05.

\*\*P < 0.01.

TABLE 3. Effects of HD Versus OL-HDF on Inflammation

	Baseline	First Dialysis	2-Week Dialysis	Change in First Dialysis vs Baseline	Change in 2-Week Dialysis vs Baseline	P-value OL-HDF vs HD
IL-6 (pg/ml)						
HD	6.87 (4.56 to 11.34)	4.97 (2.89 to 11.53)	6.14 (2.66 to 15.65)	-0.68 (-2.33 to 1.76)	-1.20 (-3.98 to 1.23)	0.01
OL-HDF	5.60 (2.40 to 12.95)	4.63 (2.11 to 8.33)	3.41 (0.94 to 9.54)	-0.55 (-2.58 to 3.65)	-1.70 (-3.35 to 0.13)*	
Difference OL-HDF vs HD	-2.01 (-5.71 to 0.01)	-1.52 (-4.44 to 0.34)	-2.30 (-3.83 to 0.61)			
hs-CRP (mg/L)						
HD	1.35 (0.50 to 4.23)	1.60 (0.63 to 5.00)	3.70 (0.83 to 8.02)	0.20 (-0.08 to 0.68)*	0.50 (0.00 to 2.40)*	0.16
OL-HDF	2.05 (0.60 to 5.55)	2.20 (0.8 to 6.3)	1.90 (0.90 to 10.40)	0.20 (0.03 to 0.75)*	0.40 (0.00 to 1.55)	
Difference OL-HDF vs HD	0.15 (-0.48 to 0.48)	0.05 (-0.20 to 0.45)	0.10 (-0.45 to 2.43)			

Values expressed as mean ± standard deviation or median (interquartile range).  
P-value OL-HDF versus HD from a linear regression model.  
HD = hemodialysis, hs-CRP = high-sensitivity C-reactive protein, IL-6 = interleukin 6, OL-HDF = online hemodiafiltration.  
\* P < 0.05.  
\*\* P < 0.01.

might exist in this study. Finally, because the aim of the study was to evaluate the acute and short-term effects in OL-HDF versus HD, long-term effect of the intermittent OL-HDF on endothelial dysfunction and inflammation in chronic HD patients could not be assessed.

CONCLUSION

OL-HDF has acute and short-term beneficial effects on endothelial dysfunction in ESRD patients compared to conventional HD, which may be related to the amelioration of inflammation, better removal of middle molecular weight substances. We could speculate a superior effect of OL-HDF in the case of its application to the chronic HD patients, especially with high risk of cardiovascular complications. Further studies are required to assess the possible therapeutic effects of the intermittent OL-HDF on endothelial dysfunction and chronic inflammation that are associated with HD.

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