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Angiotensin receptor blocker vs ACE inhibitor effects on HDL functionality in patients on maintenance hemodialysis

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Abstract

Background and Aims—Angiotensin receptor blockers (ARB) and angiotensin converting enzyme inhibitors (ACEI) reduce cardiovascular events in the general population. Maintenance hemodialysis (MHD) patients are at high cardiovascular risk but few studies have directly addressed the comparative efficacy of these drugs. MHD disrupts the normally atheroprotective actions of high density lipoprotein (HDL), therefore, we compared ACEI or ARB treatment on HDL functions in MHD.

Methods and Results—HDL was isolated at the starting point (pre) and 3–6 months later (post) in 30 MHD randomly assigned to placebo, ramipril or valsartan. Outcomes included cholesterol efflux, inflammatory cytokine response, effects on Toll-like receptors (TLR), superoxide production, methylarginine and serum amyloid A (SAA) levels. HDL from ARB- or ACEI-treated subjects was more effective in maintaining efflux than HDL of placebo. HDL from ARB- or ACEI-treated subjects but not placebo lessened cellular superoxide production. In contrast, neither ARB nor ACEI improved HDL anti-inflammatory effect. Indeed, HDL of ACEI-treated subjects potentiated the cytokine responses in association with activation of TLR but did not alter the HDL content of methylarginines or SAA.

Conclusion—Both ACEI and ARB stabilized HDL cholesterol acceptor function and sustained cellular anti-oxidative effects but not anti-inflammatory effects, and ACEI-treatment instead amplified the HDL inflammatory response. The findings reveal possible utility of antagonizing

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angiotensin actions in MDH and suggest a possible mechanism for superiority of ARB vs ACEI in the setting of advanced kidney disease.

Keywords

HDL; angiotensin-converting enzyme inhibitors; angiotensin receptor blocker; cholesterol; oxidant stress

Introduction

Although decades of epidemiological studies have firmly established the inverse relationship between HDL levels and cardiovascular events[1], recent studies emphasize HDL's functionality as a better predictor of cardiovascular disease (CVD)[2, 3]. Chronic kidney disease (CKD) impairs HDL functions including the capacity to accept cholesterol from lipid-loaded cells as well as the ability to lessen inflammation and oxidant stress, especially in patients with advanced CKD on maintenance hemodialysis (MHD)[4–7]. The MHD population is also unique in its lack of benefit from standard lipid-lowering interventions that highlights an urgent need to develop additional therapeutic strategies.

Numerous epidemiological studies document fewer cardiovascular events and increased survival of non-CKD individuals treated with angiotensin-converting enzyme inhibitors/angiotensin receptor blockers (ACEI/ARBs)[8, 9]. By contrast, there are few studies of cardiovascular benefits of interruption of the renin-angiotensin-aldosterone system in MHD patients and the results suggest variable effects in this population. Fosinopril, an ACEI, provided no protection against CV events in MHD patients while the ARB telmisartan reduced all-cause and CV mortality in MHD patients with heart failure[10, 11]. We reported that ARB use, in combination with other antihypertensives, but not with ACEI, is associated with less CV death in MHD patients[12]. We also showed that ARB treatment exerts greater anti-inflammatory effects during hemodialysis compared with ACEI[13]. In a separate study, we found that short-term treatment with ACEI, but not ARB, increased circulating levels of asymmetric dimethylarginine (ADMA), which has been linked to vascular dysfunction, CVD and mortality in CKD[14]. Interestingly, HDL of patients with moderate CKD was reported to be enriched in symmetrical dimethylarginine (SDMA) linked to enhanced nicotinamide adenine dinucleotide phosphate-oxidase-dependent reactive oxygen species (ROS) production and reduced endothelial NO bioavailability[7]. HDL enrichment in serum amyloid A (SAA) has been shown to promote systemic inflammation in MHD patients[15]. Impaired anti-inflammatory function of HDL was recently linked to reduced cholesterol efflux and abnormal plasma membrane lipid homeostasis that correlated with SAA in patients with systemic lupus erythematosus[16]. Elevated plasma SAA associated with acute phase reaction in subjects with infections, cancer or autoimmune diseases have been linked to reduced cholesterol efflux by remodeled HDL[17]. Other compositional changes such as low apolipoprotein levels in hemodialysis patients have been postulated as contributing to low cholesterol efflux capacity[18]. Together, these findings underscore the potential for pleiotropic effects of ACEI/ARB inhibition on vascular inflammation and oxidative stress in across CKD.

To date no study has compared the effects of ARB versus ACEI on HDL function in patients on MHD. To evaluate the effects of treatment with either ACEI or ARB on HDL functionality in MHD patients, we performed a substudy as a part of a randomized double blind placebo-controlled study (NCT00878969). The outcome measures were HDL cholesterol efflux capacity, cellular anti-inflammation and anti-oxidant response before and 3–6 months after treatment.

Methods

Study population

The study was approved by Vanderbilt University Institutional Review Board and was performed according to the Declaration of Helsinki. All subjects underwent adequate hemodialysis ($Kt/V > 1.2$) thrice weekly for at least six consecutive months. Patients with a history of active connective tissue disease, acute infection or use of immunosuppressive drugs, advanced liver disease, gastrointestinal dysfunction requiring parental nutrition or active malignancy, history of myocardial infarction or cerebrovascular event within three months prior to study or with an ejection fraction lower than 30% were excluded. Not all subjects were available for every assay. However, changes between pre and post were given individual.

Study protocol

This was a substudy within a randomized, double-blind, placebo control design (NCT00878969). Subjects were randomized to treatment with ramipril (King Pharmaceuticals, Bristol, TN), valsartan (Novartis Pharmaceuticals, East Hanover, NJ) or placebo. Patients receiving either ACEI or ARB prior to study discontinued that medication for three weeks (washout period). If, at any time, systolic blood pressure exceeded 160 mmHg or diastolic exceeded 100 mmHg, other antihypertensive medications were maximized. Following the initial washout period, thirty subjects who were randomly assigned to one of the three treatment arms: ramipril (5 mg/d, n=10), valsartan (160 mg/d, n=8) and matching placebo (n=12). The participant characteristics are shown at Table 1.

HDL isolation and macrophage cholesterol efflux capacity (CEC) to HDL

Blood samples were obtained at the starting point of the study (pre) and 3–6 months later (post). The blood was collected at the beginning of a dialysis treatment and HDL fraction isolated by density gradient ultracentrifugation. THP-1 cells were plated, differentiated into macrophages then enriched with acetylated-low density lipoprotein (LDL) (100µg/ml, Intracel, Frederick, MA) and exposed to HDL (50mg/ml) and LPS (50 ng/ml) for 24h. Cholesterol content was measured by the demanding and rigorous gas chromatography method because flux of cholesterol between cells and HDL is bi-directional and analysis of cholesterol efflux by analysis of cholesterol mass is superior to other methods since other methods disregard the influx of cholesterol whereas gas chromatography determines the net flux of cholesterol[19]. Cholesterol efflux determined as the percent at baseline versus after incubation with HDL. Cell protein content was measured by bicinchoninic acid assay[5, 20].

Macrophage inflammatory cytokine and oxidative stress response and HDL composition

HDL modulation of inflammatory response was measured using the established cytokine response in LPS-activated macrophages[5]. Quantification of interleukin (IL)-1 β , monocyte chemoattractant protein-1 (MCP-1), tumor necrosis factor- α (TNF- α), Toll like receptor (TLR)2, TLR4, and 18S rRNA levels was performed by real-time reverse transcriptase polymerase chain reaction (PCR) using CFX96™ Real-Time System (BIO-RAD, Hercules, CA). Probes for IL-1 β (Hs99999029_m1), MCP-1 (Hs00234140_m1), TNF- α (Hs99999043_m1), TLR2(Hs01872448_s1), TLR4(Hs00152939_m1) and 18S rRNA (Applied Biosystems, Foster City, CA). To evaluate intracellular production of superoxide, we measured the formation of superoxide specific product of dihydroethidium, 2-hydroxyethidium, using high-performance liquid chromatography (HPLC) analysis. THP-1 macrophages were washed with Kreps-Hepes buffer and incubated in Kreps-Hepes buffer with 10 μ M (micromole/Liter) DHE for 30 min, then scraped and collected in 300 μ l of methanol. Samples were homogenized and filtrated through 0.22 μ l filters and run on HPLC machine. 2-Hydroxyethidium was expressed per milligram protein[21]. Plasma and HDL levels of ADMA and SDMA were measured by HPLC with fluorescence detection[22]. SAA concentrations from plasma and purified HDL were measured by a double sandwich ELISA kit (Immunology Consultants Laboratory Inc).

Statistical analysis

Results are expressed as a mean of triplicate assays in efflux and duplicate assays in mRNA of inflammation cytokine response and ROS measurements. This is an exploratory analysis. No multiple comparison adjustment was done. Descriptive statistics are presented as counts for categorical variables and median (interquartile range [IQR]) for continuous variables. The differences between pre- and post-values were tested with the Wilcoxon signed rank test. Kruskal Wallis test was used to examine the change among three treatment groups, comparisons between treatment groups (ACEI, ARB) and the placebo arm of pre- and post-value differences were performed with the Wilcoxon rank sum test. Statistical analyses were performed using R version 3.3.3 (<http://www.r-project.org/>). A 2-sided significance level of 5% was required for consideration as statistically significant.

Results

Patient demographics

Table 1 summarizes demographic, clinical characteristics, and HDL composition. Patients were mostly African American (60%), female (60%), with a median age of 53 [IQR: 45, 63] years. Causes of ESRD were diabetes mellitus (29%), hypertension (39%), polycystic kidney disease (6%), non-steroidal anti-inflammatory drug (NSAID)-induced chronic kidney disease (3.3%), and unknown (23%). The groups were similar with the exception of higher percentage of smokers in placebo and ACEI groups.

Effects of ARB and ACEI treatment on HDL cholesterol efflux capacity (CEC)

Compared to baseline level at randomization, CEC of HDL from the placebo-treated group was significantly lower at the end of the study ($P < 0.001$) whereas no differences were

observed for CEC of HDL from ARB- and ACEI-treated groups (Figure 1A). Comparison of changes between pre and post CEC among the groups showed that decrease in CEC observed in placebo-treated subjects was significantly lower than treatment with ACEI ($P=0.02$) whereas there was a trend for ARB treatment to achieve the same effect without reaching statistical significance ($p=0.06$) (Figure 1B).

Effects of ARB and ACEI treatment on HDL cellular response to oxidative stress

The oxidative stress level of LPS-exposed macrophages in the presence of HDL isolated at baseline versus after placebo treatment was not different. By contrast, HDL from ARB-treated subjects significantly reduced macrophage oxidant stress (Figure 2A). Similarly, HDL from ACEI-treated subjects significantly reduced macrophage oxidant stress (Figure 2A). There was no difference in the anti-oxidant effect between the two treatment groups (Figure 2B).

Effects of ARB and ACEI treatment on HDL cellular inflammatory response

Macrophage inflammatory response (IL-1 β , MCP-1 and TNF- α) to HDL of placebo-treated subjects isolated at baseline randomization and second visit was not different (Figure 3A, 3C, 3E). Similarly, inflammatory cytokine responses to HDL of ARB-treated subjects before and after treatment were not different. On the other hand, compared to baseline randomization, HDL isolated from MHD treated with ACEI caused a greater inflammatory response. Thus, comparison of the pre and post inflammatory cytokine response in each of the three treatment groups showed significant potentiation in IL-1 β and TNF- α with a directionally similar response in MCP-1 to HDL isolated from ACEI-treated subjects (Figure 3B, 3D, 3F). ARB/ACEI treatment also affected the TLR response (Figure 4). TLR2 response was numerically highest in placebo and ACEI-treated groups although neither reached statistical significance (Figure 4A). For TLR4, compared to HDL of ARB or placebo, HDL of ACEI-treated patients potentiated the response in THP-1 cells (Figure 4C), although intergroup comparison did not reach statistical difference (Figure 4D).

Effects of ARB and ACEI treatment on ADMA, SDMA, SAA

Although plasma levels of ADMA at the end of the study were numerically lower, this reduction was not statistically significant in any group (Table 2). Plasma SDMA levels also did not change with any intervention. ADMA/SDMA levels were below detection before the interventions and therefore could not be compared to levels at the end of the study. At the end of the study, neither ADMA nor SDMA levels were different among the groups. Plasma SAA level was not affected by either treatment. SAA content in HDL was also not different before/after treatment or among the treatment groups at the 2nd visit.

Discussion

Angiotensin receptor antagonists and angiotensin converting enzyme inhibitors are widely and interchangeably used in many clinical settings, although recent findings suggest that in MHD patients, they may have different effects. The current study examined ACEI and ARB treatment on HDL function in MHD. Compared to placebo, HDL of MHD patients treated with either ARB or ACEI supported the capacity to elicit cholesterol efflux from lipid-

loaded macrophages (Figure 1). Thus, while CEC of placebo-treated subjects fell by 36%, the CEC of HDL from subjects treated with ARB or ACEI was not reduced over the same time frame. The degree of preservation of efflux capacity was similar in ARB and ACEI groups. These data support the intriguing possibility that uremia-related HDL dysfunction can be modified by therapeutic interventions. These observations complement our previous studies showing that in *vitro* activation of cellular transporters by LXR agonists can increase efflux to dysfunctional HDL of MHD patients[5]. Therapeutic interventions can improve HDL's cholesterol efflux capacity in other chronic illnesses. For example, cholesterol efflux was increased by treatment with immunomodulatory agents in patients with rheumatoid arthritis[23, 24]. Several groups, including ours, have shown that although the magnitude of efflux increase is small, the benefit may be especially relevant in individuals with the most depressed cholesterol levels and the highest disease activity scores[23, 24].

To date, few studies have directly addressed possible cardiovascular benefits of ARB and ACEI in MHD. Compared to placebo-treated group, ARB/ACEI had a similar protective on the efflux capacity of HDL. As these are prevalent MHD patients, the CEC decrease does not reflect potentially adverse effects of dialysis initiation. On the other hand, it is possible that lower CEC reflects withdrawal of ARB/ACEI therapy in the placebo-treatment group. Indeed, 40% of placebo-treated subjects were maintained on ARB/ACEI prior to randomization. In any case, compared to placebo, antagonism of angiotensin II activity preserved CEC. In view of studies showing that CEC is a marker of atherosclerosis as detected by radiographic assessments and can predict cardiovascular events[2, 3], our findings suggest that ARB/ACEI intervention may prevent a decline in cholesterol efflux capacity in the MHD population. In non-diabetic nephrotics, lisinopril did not affect plasma cholesterol efflux capacity although the ACEI treatment did not modify elevated pre-beta HDL that may be the driver for cholesterol efflux in this setting[25]. Kopecky et al. reported that cholesterol efflux does not predict cardiovascular risk in diabetic patients on maintenance hemodialysis[26]. However, the authors acknowledge that the adverse consequences of years of diabetes and the long-term evolution in CVD may obscure differences in efflux capacity observed at baseline versus during follow up. Although the study did not investigate concomitant use of medications, in patients with the lowest CEC, atorvastatin reduced the risk for all cardiac events combined, results that remained significant after adjustment for multiple relevant risk factors. Since the half-life of HDL is 3–5 days, our data underscore the possibility that therapeutic interventions with ARB and ACEI can modulate CEC of the continuously renewed HDL over a period of weeks-to-months. Whether such interventions can be used to alter CV risk in the MHD population remains to be determined.

Complementing the effects of ARB and ACEI on HDL capacity to elicit cholesterol efflux, HDL of ARB- and ACEI-treated groups significantly lessened macrophage production of superoxide (Figure 2). As with efflux, there was no difference in this effect between the ARB and ACEI groups. These data are interesting in view of the extensive evidence that CKD potentiates cellular production of superoxide radicals and that the HDL of CKD patients have impaired antioxidant activity[4, 7, 26–29]. Indeed, a prospective study observed that high levels of oxidized HDL are associated with increased CIMT, while the combination of high ox-HDL and high interleukin-6 predicts not only a greater increase in

carotid intima-media thickness but also an increased risk for CVD events and CVD-related mortality in MHD patients[28]. In multivariable models adjusting for established clinical and biochemical risk factors, diminished antioxidant properties of PON-1 predicted higher risk of incident long-term adverse cardiovascular events (heart attack, stroke, or death) in patients with CKD[6]. Given the clinical evidence that inhibition of angiotensin II with ARB or ACEI can lessen oxidative stress and reduce CVD events in the general population, and experimental studies showing that ARB can ameliorate CKD-accelerated atherosclerosis, the current findings suggest that ARB/ACEI modulation of HDL-driven mechanisms (cholesterol efflux and anti-oxidant protection) may have beneficial effects in the MHD population[30].

The potentially advantageous effect of both ARB and ACEI on HDL-mediated cholesterol efflux and oxidant stress appears confounded by consequences specific to each treatment. HDL of MHD patients is known to have defective anti-inflammatory function[5]. Neither ARB nor ACEI improved the macrophage inflammatory cytokine response to uremic HDL, and HDL of ACEI-treated group rather potentiated the cytokine response (Figure 3). These findings are interesting in view of the observations that while antagonism of the renin-angiotensin-aldosterone system in the general population has become a cornerstone of evidence-based therapy for protection against cardiovascular morbidity and mortality, this approach in MHD has yielded mixed results[8–15]. Our group reported that ARB in combination with other antihypertensives, but not with ACEI, has a beneficial effect on cardiovascular death in MHD patients[12]. In another study, we found that although 7-day treatment with either ARB or ACEI reduced circulating levels of IL-6 and IL-8 in MHD patients, ACEI-treatment increased pro-inflammatory IL-1 β and reduced levels of the anti-inflammatory IL-10[13]. The current study extends those findings with the novel observation that in MHD patients, ACEI but not ARB treatment leads to HDL that amplifies the macrophage proinflammatory cytokine response. In this connection, the recent landmark study in patients with myocardial infarction treated with monoclonal antibody targeting interleukin-1 β with canakinumab showed significantly reduced recurrent cardiovascular events compared placebo[31]. Interestingly, we previously reported that interventions with interleukin1 receptor antagonist or IL-1 trap reduced markers of systemic inflammation and oxidative stress and improved endothelial function in CKD patients[32]. In recent studies we show that the anti-inflammatory and anti-oxidative properties of HDL in these CKD patients is improved by treatment with IL-1blockers[33].

Several mechanisms may contribute to the heightened inflammatory response to HDL from ACEI-treated individuals[34, 35]. We reported that MHD patients treated for one week with ACEI, but not ARB, have increased circulating levels of methylarginine, specifically ADMA[17]. This is notable because loss of anti-inflammatory and anti-oxidant functions of HDL from CKD patients was linked to SDMA through pathways that include Toll-like receptors[7]. In our study, plasma ADMA or SDMA were not significantly affected by treatment with ARB or ACEI. Further, neither ARB nor ACEI affected the ADMA or SDMA content in HDL. Although the TLR2 and TLR4 response to HDL from placebo-treated group tended to increase and reached significance for TLR4 in ACEI-treated group (Figure 4C), there was no statistical difference among the groups (Figure 4B and 4D). These findings do not support cytokine potentiation occurring through mechanisms involving HDL

methyarginine content regulation of TLRs in our MHD population. HDL also carries SAA, which can directly affect the anti-oxidative and anti-inflammatory capacity of the lipoprotein[16, 36]. In the current study, there were no differences in these levels among the two treatment arms and placebo. These findings suggest that SAA is not responsible for the proinflammatory effects of HDL of ACEI-treated patients, at least based on our small study.

Limitations of our study include the small number of patients. This was a hypothesis generating analysis and we cannot make confirmatory conclusions regarding differences among the ARB-ACEI- and placebo-treated groups. Nonetheless, it is a first look at the relationship between HDL functionality and effects of treatment with ACEI, ARB or placebo. Future experiments with larger sample size will be needed to further assess this potential interaction. ACEI and placebo groups had more individuals with a history of smoking. Since smoking can affect HDL efflux capacity, we cannot exclude a possible positive effect of previous non-smoking status in ARB versus ACEI and placebo groups. The study also lacked the ability to examine correlation to the hard endpoint such as CV events or mortality. Nonetheless, these limitations were offset by a number of strengths including the randomized, double-blind, placebo control design that limits the inter-subject variation as well as assessment of the study parameters over time in the same subjects. The study also presented a comprehensive analysis of HDL efflux capacity, cellular anti-inflammation and anti-oxidant responses, and for the first time, examined the effects of ARB and ACEI treatment on HDL functionalities that may have clinical relevance in MHD patients.

In summary, our study shows treatment with ARB and ACEI stabilize HDL cholesterol acceptor function of MHD patients and improves HDL-mediated antioxidant function. On the other hand, ACEI rather potentiates the cellular inflammatory response. The findings suggest potential utility of antagonizing angiotensin actions in MDH patients and propose a possible mechanism for difference of ARB vs ACEI in this population.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

None

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Abbreviations

ARB	angiotensin receptor blockers
ACEI	angiotensin converting enzyme inhibitors
HDL	high density lipoprotein
MHD	maintenance hemodialysis
TLR	Toll-like receptors

SAA	serum amyloid A
IL-1β	interleukin-1 β
MCP-1	monocyte chemoattractant protein-1
TNF-α	tumor necrosis factor- α
ADMA	asymmetric dimethylarginine
SDMA	symmetric dimethylarginine

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Highlights

- HDL cholesterol acceptor function in MHD patients was stabilized by treatment with ACEI and ARB, which also sustain cellular anti-oxidative effects.
- By contrast, anti-inflammatory effects of HDL were not improved by either ARB or ACEI, and ACEI-treatment instead amplifies the HDL inflammatory response.
- Enhanced inflammatory response to HDL of ACEI-treated subjects were associated with activation of TLR-4 but not HDL content of potential modulators of HDL function such as methylarginines or serum amyloid A.
- The results suggest a possible mechanism for different effects of ARB vs ACEI in MHD patients.

Figure 1.

Cholesterol acceptor capacity of HDL from MHD patients before (pre) and after 3–6 month treatment with ARB, ACEI or placebo (post). (A) Cholesterol content in THP-1 macrophages was measured by gas chromatography and efflux determined as percent at baseline versus after incubation with HDL. $N=11, 7, 10$ for placebo, ARB and ACEI. Bars show median values compared by paired t-test. (B) Intergroup comparison of pre/post values: $N=11, 7, 10$ for placebo, ARB and ACEI.

Figure 2.

Cellular production of reactive oxygen species (ROS) in response to HDL from MHD patients before (pre) or after 3–6 month treatment with ARB, ACEI or placebo (post). (A) ROS measured cellular formation of superoxide-specific product of dihydroethidium, 2-hydroxyethidium, by high-performance liquid chromatography. N=12, 7, 9 for placebo, ARB and ACEI. Bars show median values compared by Wilcoxon Signed rank test. (B) Intercomparison of pre/post values. N=12, 7, 9 for placebo, ARB and ACEI.

Figure 3.

Cytokine response to HDL from MHD patients before (pre) or after 3–6 month treatment with ARB, ACEI or placebo (post). mRNA expression for interleukin-1 β (IL-1 β), N=9, 7, 9 for placebo, ARB and ACEI; monocyte chemoattractant protein (MCP)-1, N=9, 7, 9 for placebo, ARB and ACEI; and tumor necrosis factor (TNF)- α measured by real-time PCR in each group (A, C, E), N=9, 7, 9 for placebo, ARB and ACEI. Bars show median values compared by Wilcoxon Signed rank test. Intergroup comparisons of pre/post (B, D, F). N=9, 7, 9 for placebo, ARB and ACEI.

Figure 4.

Toll like receptor (TLR)-2 (A) and TLR-4 (C) mRNA expression of THP-1 cells exposed to HDL from MHD patients before (pre) or after 3–6 month treatment with ARB, ACEI or placebo (post) measured by real-time PCR. N=6, 5, 6 for placebo, ARB and ACEI, respectively. Intergroup comparisons of pre/post (B, D). N=6, 5, 6 for placebo, ARB and ACEI.

Table 1

Baseline characteristics by study groups

Parameter	ARB	ACEi	Placebo	P Value
N	8	10	12	
Age (yr)	55.0 [50.0, 64.0]	54.5 [52.0, 63.8]	51.7	0.51
Gender (male: female)	3:5	5:5	4:8	0.72
Race (African-	5:3:0	6:3:1	8:4:0	0.98
Smoker (yes: no)	0:8	3:7	6:6	0.06
Diabetes (yes: no)	3:5	7:3	8:4	0.31
Statin use (yes: no)	2:6	7:3	7:5	0.15
BMI (kg/m ²)	29.9 [26.2, 33.6]	27.6 [26.4, 34.9]	31.7	0.98
Glucose (mg/dL)	105.0 [90.2, 110.8]	91.0 [83.0, 141.8]	90.0	0.51
HDL cholesterol (mg/dL)	33.0 [25.0, 41.0]	41.0 [35.0, 51.5]	37.0	0.71
Triglycerides (mg/dL)	123.0 [89.2, 110.8]	101.0 [65.2, 154.0]	152.5	0.57
LDL cholesterol (mg/dl)	74.0 [70.0, 79.0]	63.0 [50.0, 73.0]	85.0	0.35
Total cholesterol (mg/dL)	146 [126, 157]	126 [112, 138]	161	0.16

Data are presented as median with interquartile range [IQR] for continuous variables and as counts for categorical variables.

P value was calculated using the Pearson chi-square for categorical variables and the Wilcoxon rank sum test for continuous variables.

Effects of ARB and ACEI treatment on asymmetric dimethylarginine (ADMA), symmetric dimethylarginine (SDMA), serum amyloid A (SAA) in plasma and HDL

Table 2

		Plasma (µg/ml)		HDL-total protein (µg/ml)			
		Pre	Post	P value	Pre	Post	P value
ADMA							
ARB	0.71 [0.69–0.75]	0.46 [0.40–0.60]	1.00	Below detection	0.013 [0.009–0.029]	–	–
ACEI	0.67 [0.60–0.68]	0.44 [0.37–0.49]	0.06	Below detection	0.025 [0.022–0.048]	–	–
Placebo	0.66 [0.60–0.75]	0.52 [0.49–0.57]	0.13	Below detection	0.025 [0.019–0.035]	–	–
SDMA							
ARB	3.0 [2.9–4.6]	4.5 [3.0–4.9]	0.75	Below detection	0.120 [0.086–0.252]	–	–
ACEI	3.7 [3.4–4.7]	3.1 [2.3–3.2]	0.19	Below detection	0.164 [0.123–0.256]	–	–
Placebo	3.7 [3.0–4.1]	4.0 [3.3–4.4]	0.38	Below detection	0.136 [0.111–0.150]	–	–
SAA							
ARB	0.51 [0.50–0.85]	0.86 [0.47–1.57]	0.19	0.17 [0.15–0.23]	0.44 [0.21–0.69]	0.38	0.38
ACEI	0.85 [0.41–1.03]	0.62 [0.37–0.67]	0.58	0.12 [0.08–0.38]	0.18 [0.09–0.19]	0.38	0.38
Placebo	0.59 [0.44–0.83]	0.73 [0.46–1.38]	0.47	0.18 [0.15–0.29]	0.14 [0.08–0.25]	0.73	0.73

Data are presented as median with interquartile range. P value compares Pre vs Post using Wilcoxon signed rank test