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Myeloperoxidase Levels Predict Accelerated Progression of Coronary Atherosclerosis in Diabetic Patients: Insights from Intravascular Ultrasound

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Abstract

Objective—While inflammation has been proposed to contribute to the adverse cardiovascular outcome in diabetic patients, the specific pathways involved have not been elucidated. The leukocyte derived product, myeloperoxidase (MPO), has been implicated in all stages of atherosclerosis. The relationship between MPO and accelerated disease progression observed in diabetic patients has not been studied.

Methods—We investigated the relationship between MPO and disease progression in diabetic patients. 881 patients with angiographic coronary artery disease underwent serial evaluation of atherosclerotic burden with intravascular ultrasound. Disease progression in diabetic (n=199) and non-diabetic (n=682) patients, stratified by baseline MPO levels was investigated.

Results—MPO levels were similar in patients with and without diabetes (1362 vs. 1255 pmol/L, p=0.43). No relationship was observed between increasing quartiles of MPO and either baseline (p=0.81) or serial changes (p=0.43) in levels of percent atheroma volume (PAV) in non-diabetic patients. In contrast, increasing MPO quartiles were associated with accelerated PAV progression in diabetic patients (p=0.03). While optimal control of lipid and the use of high-dose statin were associated with less disease progression, a greater benefit was observed in diabetic patients with lower compared with higher MPO levels at baseline.

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Conflict of Interest

All other authors have reported that they have no relationships to disclose.

Conclusions—Increasing MPO levels are associated with greater progression of atherosclerosis in diabetic patients. This finding indicates the potential importance of MPO pathways in diabetic cardiovascular disease.

Keywords

diabetes mellitus; myeloperoxidase; atheroma progression; intravascular ultrasound; coronary atherosclerosis

Diabetic patients have a markedly increased incidence of adverse cardiovascular events, in association with an extensive atheroma burden and an accelerated plaque progression [1–4]. In these patients, the combination of impaired glycemic profile with hypertension, dyslipidemia and obesity are considered to have a detrimental impact on the artery wall and lead to the development of extensive atherosclerotic plaque.

In addition to these metabolic abnormalities, increasing evidence supports a central role for inflammation in the pathogenesis of both atherosclerosis and diabetes [5–8]. Previously, a greater inflammatory cell infiltration was observed on pathological examination of atherosclerotic plaques of diabetic patients [10, 11]. Moreover, a positive correlation has existed between inflammation and progression of carotid intima-media thickness in diabetic subjects [12, 13]. However, it remains to be fully elucidated which specific inflammatory pathways are upregulated in diabetic atherosclerosis.

Myeloperoxidase (MPO) is a pro-oxidant enzyme released from granules of activated neutrophils, monocytes, and certain tissue macrophages [14, 15]. While a major biological function of MPO is the defense of the organism against infections by generating antimicrobial oxidants, free radicals and other reactive oxidant species [16, 17], this activity can also lead to oxidative damage of endothelium and vessel wall [18, 19]. MPO promotes oxidation of low-density lipoprotein (LDL) [20], and apolipoprotein A-I in high-density lipoproteins (HDL), which impairs its ability to promote reverse cholesterol transport [21–23]. MPO also directly scavenges nitric oxide, diminishing nitric oxide bioavailability leading to endothelial dysfunction [24–26]. Recent study has reported that HDL-associated MPO activity and content were increased in diabetic patients [27]. Furthermore, MPO-derived oxidants impaired the endothelial-protective effect of HDL, leading to endothelial dysfunction [27]. Given that endothelial dysfunction has been considered to associate with the development of atherosclerosis, we hypothesized that MPO may contribute to the initiation and propagation of atheromatous plaque in diabetic patients.

Intravascular ultrasound (IVUS) permits high resolution imaging of the arterial wall [28] and has been employed in clinical trials to assess the effect of various anti-atherosclerotic drugs on coronary atherosclerosis [29–31]. The purpose of the current study was to investigate the relationship between systemic level of MPO, atheroma burden and progression in non-diabetic and diabetic patients with coronary artery disease (CAD).

Methods

Study Population

From 1184 patients in 3 clinical trials that evaluated the effect of medical therapies on the progression of coronary atherosclerosis by using IVUS, we identified and analyzed 881 patients with available serum MPO level at baseline. Of these patients, there are 682 non-diabetic and 199 diabetic patients with follow-up IVUS data. Three clinical trials analyzed in the current study were the REVERSAL (Reversal of Atherosclerosis With Aggressive Lipid Lowering) study [29] the CAMELOT (Comparison of Amlodipine Versus Enalapril to Limit Occurrences of Thrombosis) study [30] and the ACTIVATE (Acyl:Cholesterol Acyltransferase Intavascular Atherosclerosis Treatment Evaluation) study [31]. These clinical trials employed serial IVUS examination to assess the impact of intensive lipid lowering, antihypertensive therapy and acyl:cholesterol acyltransferase inhibition on progression of coronary atherosclerosis. Eligibility criteria in these clinical trials included the presence of stable CAD, defined as a lumen stenosis >20% in at least one major epicardial coronary artery on visual estimation of the coronary angiogram performed for a clinical indication. Each study was approved by the institutional review boards of the participating clinical trial sites, and all participants in the trials provided informed written consent before enrollment.

Acquisition and analysis of IVUS images

The methods for acquisition and analysis of IVUS images have been described previously [29–31]. In brief, a target vessel without luminal stenosis >50% within a segment of at least 30 mm in length was selected for imaging. This vessel was required to not have undergone previous revascularization or represent the culprit vessel for a prior myocardial infarction. After anticoagulation therapy and administration of intracoronary nitroglycerin, an imaging catheter containing a high-frequency ultrasound transducer (30 to 40MHz) was inserted distally within the coronary artery. Ultrasonic images were continuously recorded on videotape during withdrawal of the catheter at a constant rate of 0.5mm/s. Imaging was performed within the same coronary artery at baseline and at the end of the study, which ranged from 18 to 24 months.

The recorded images were digitized for subsequent analysis. An anatomically matched segment was defined at the 2 time points on the basis of proximal and distal side branches (fiduciary points). Cross-sectional images spaced precisely 1 mm apart were selected for measurement. The leading edges of the lumen and external elastic membrane (EEM) were traced by manual planimetry. Plaque area was defined as the area occupied between these leading edges.

The percent atheroma volume (PAV) was calculated as the proportion of the entire vessel wall occupied by atherosclerotic plaque and was selected by virtue of its strong association with clinical outcomes.

$$\text{PAV formula} \quad \text{PAV (\%)} = \left[\frac{\sum (\text{EEMarea} - \text{LUMENarea})}{\sum \text{EEMarea}} \right] \times 100$$

Biochemical Analyses of MPO Levels

Systemic concentrations of MPO were measured at baseline using a commercially available FDA-cleared ELISA (CardioMPO Test, Cleveland Heart Lab, Cleveland, Ohio). The inter-assay and intra-assay coefficients of variance were 2% and 6%. The lower detection limit was 13 pmol/l, and the upper detection limit was 5,223 pmol/l. Samples with MPO values in excess of this were remeasured upon dilution. Spike and recovery studies with isolated human MPO into plasma matrix showed >95% accuracy [35]. Samples were analyzed in random order to avoid systemic bias by laboratory personnel, who were blinded to all clinical and laboratory phenotypic data.

Statistical Analysis

Continuous variables were expressed as mean $\pm$ SD if normally distributed (or median and interquartile range if not normally distributed) and categorical variables as percentage. In non-diabetic and diabetic patients respectively, clinical characteristics, medication use and atherosclerotic plaque burden at baseline were compared across quartiles of baseline MPO levels using polyserial correlation analysis for normally distributed continuous variables and Spearman rank correlation analysis for non-normally distributed continuous variables, and Cochran-Armitage trend test for categorical variables.

Serial changes in plaque burden (expressed as least-squares mean $\pm$ SEM) across the MPO quartiles were assessed with a random effect mixed model adjusting for baseline plaque burden, in non-diabetic and diabetic patients respectively, with trial set as a random factor to control for heterogeneity across the 3 studies. A test of trend on the serial changes across the increasing MPO quartiles was also conducted within each of the patient groups. Since there was no trend in the non-diabetic patients, subsequent analyses focused on diabetic patients. A multivariable mixed model adjusting for clinical characteristics was then constructed for diabetic patients through model selection. First, covariates that were univariately contributive to the relationship of atheroma progression and MPO quartiles (with a p-value <0.10) were chosen. These covariates were further selected by bootstrap resampling (1000 iterations) and the final covariates were baseline and change in LDL-C, diastolic blood pressure and concomitant insulin use. The relationship of atheroma progression against baseline MPO was further examined in diabetic patients by checking the effect of the interactions between high-level MPO and intensive risk factor control of LDL cholesterol and high-dose statin use. A 2-sided probability value of <0.05 was considered statistically significant. All statistical analyses were performed using SAS software, version 9.2 (SAS Institute, Cary North Carolina).

Results

Clinical Characteristics

In non-diabetic patients, patients with the highest MPO level were more likely to be a current smoker ($p<0.001$) and less likely to be female ($p=0.02$) (Table 1). Also, higher level of baseline high sensitivity C-reactive protein (hsCRP) ($p=0.01$) and lower levels of baseline LDL cholesterol ($p<0.001$), HDL cholesterol ($p=0.001$) and fasting glucose ($p=0.01$) were observed in this group. The prevalence of other coronary risk factors and the use of

established anti-atherosclerotic therapies were similar across the groups. Risk factor controls with treatments were summarized in Supplementary Data, Table 1. On therapy, non-diabetic patients with the highest MPO level had lower level of HDL cholesterol ($p=0.001$) and higher levels of triglyceride ($p=0.003$) and hsCRP ($p=0.01$) at follow-up. Non-diabetic patients with the highest MPO level were more likely to be treated with beta-blocker at follow-up ($p=0.03$). There were no significant differences in the use of other anti-atherosclerotic therapies at follow-up.

In diabetic patients, patients with the highest MPO level were less likely to have a history of hypertension ($p=0.04$) and dyslipidemia ($p=0.04$) (Table 2). Diabetic patients with the highest MPO level tended to be less likely to receive an angiotensin-converting enzyme inhibitor although this comparison failed to meet statistical significance ($p=0.05$). A lower level of LDL cholesterol was observed in this group ($p=0.02$). Diabetic patients with the highest MPO level also had higher level of hsCRP, but this comparison was borderline significance ($p=0.05$). At follow-up, diabetic patients with the highest MPO level had higher level of hsCRP ($p=0.01$) and were less likely to be treated with high dose statins ($p=0.02$) (Supplementary Data, Table 2). There was no significant difference in serum MPO level at baseline between non-diabetic and diabetic patients (1362 (IQR [775, 2263]) vs. 1255 (IQR [584, 2294]) pmol/L, $p=0.43$).

Atheroma Burden and Progression

In non-diabetic patients, baseline PAV was similar across the groups ($p=0.81$) (Table 1). On serial evaluation, no significant relationship was observed between increasing quartiles of MPO and serial changes in PAV ($p=0.43$ for trend) (Figure 1-a, left panel). In contrast to non-diabetic patients, diabetic patients with 4th quartile of MPO exhibited greater baseline PAV although this comparison showed borderline significance (Table 2, $p=0.05$). In addition, serial IVUS imaging showed greater progression of PAV in association with the increased level of MPO, especially, 3rd and 4th quartile of MPO ($p=0.02$ for trend, Figure 1-a, right panel). Multivariable analysis, adjusting for baseline and change in LDL-C, diastolic blood pressure and concomitant insulin use, revealed that higher MPO levels remained independently associated with greater progression of PAV ($p=0.03$ for trend, Figure 1-b).

In diabetic patients, plaque regression defined as change in PAV $< 0\%$ was negatively associated with MPO quartiles ($p=0.04$), whereas this relationship in non-diabetic patients was not significant ($p=0.19$).

Intensive Risk Control and Atheroma Progression in Diabetic Patients

The impact of intensive control of LDL cholesterol and the effect of high-dose statin use were summarized in Figure 2. While intensive lowering of LDL cholesterol to levels less than 80mg/dL was associated with less progression of PAV, a greater benefit was observed in diabetic patients with low MPO (change in PAV [95% confidence interval]: -0.63% [-1.89 – 0.64] vs. 1.33% [0.37 – 2.28], $p=0.02$) compared to those with high MPO levels (change in PAV [95% confidence interval]: 1.91% [0.58 – 3.25] vs. 2.16% [1.26 – 3.05], $p=0.76$) (Figure 2, left panel, $p=0.14$ for interaction test). Even in multivariable analysis which adjusted for baseline PAV, diastolic blood pressure and concomitant insulin use,

these results were still consistent (diabetic patients with lower MPO: $p=0.01$, diabetic patients with higher MPO: $p=0.82$; $p=0.10$ for interaction test). The impact of high dose statins was also investigated in diabetic patients in whom data about dose of statins were available in the current analysis ($n=150$). While high dose statins were beneficial in diabetic patients with lower MPO levels (change in PAV [95% confidence interval]: -0.96% [$-2.39-0.46$] vs. 1.37% [$0.34-2.39$], $p=0.01$), this impact was diminished in diabetic patients with higher MPO level [95% confidence interval]: 1.24% [$-0.43-2.92$] vs. 2.29 [$1.34-3.24$], $p=0.28$) (Figure 2, right panel, $p=0.33$ for interaction test). These results persisted in multivariable analysis with the adjustment for baseline PAV, diastolic blood pressure and concomitant insulin use (diabetic patients with lower MPO: $p=0.01$, diabetic patients with higher MPO: $p=0.33$; $p=0.23$ for interaction test).

Discussion

The current analysis demonstrates that elevated systemic MPO levels are associated with a particularly aggressive form of atherosclerotic disease progression in diabetic patients. In these patients with high MPO level, the benefits from achieving a favorable control of LDL-C and the use of high-dose statin were significantly diminished. The current study thus indicates the importance of MPO pathways in diabetic cardiovascular disease.

Even on aggressive high potency statin intervention, a majority of diabetic patients still possess substantial residual risk of clinical events and rapid progressive atherosclerotic plaque [1–4]. In these patients, inflammation is considered an essential contributor to all stages of atherosclerosis [5–12], and elucidation of the factors involved in this complex cascade should reveal more accurate predictors of cardiovascular risk and targets for therapeutic intervention. However, the role of specific inflammatory mediators associated with diabetic atherosclerosis remains to be defined. In recent studies, higher systemic concentrations of MPO have been observed to be associated with the presence of angiographic CAD, and both increased cardiovascular risks and long-term cardiovascular mortality independent of classic cardiovascular risk factors [32–37]. Thus, MPO may serve as both a potential marker of cardiovascular disease and a potential target for treatment. In the current IVUS study, serial IVUS imaging demonstrated a significant association between elevated systemic MPO levels and atheroma progression in diabetic patients but not in non-diabetic patients despite similar level of MPO in these two groups. The reasons for the enhanced associations in the diabetic state are unknown. Since the MPO measure performed is one of mass, and not activity, and MPO requires hydrogen peroxide as a substrate to be catalytically active, it is tempting to speculate that the increased association of systemic MPO to atheroma burden progression in diabetics is observed because of enhanced overall systemic reactive oxidant species tone in diabetics, with increased potential for MPO catalytic activity.

Recent observations show that MPO modifies apolipoprotein A-I in HDL, impairing its capacity to promote cholesterol efflux from macrophage [21, 22]. Also, Sorrentino et al. demonstrated that diabetic patients exhibited more enhanced HDL associated MPO activity with a substantially increased lipid peroxidation of HDL [27]. In addition, exposure of HDL to MPO-generated nitrating and chlorinating oxidants rapidly impaired its capacity to

stimulate endothelial nitric oxide production. Another study using diabetic rats reported that MPO activity in vessels is markedly increased [38]. Furthermore, MPO uses hydrogen peroxide stimulated by hyperglycemia and thereby, amplifying endothelial dysfunction, which is a major contributing factor to the diabetic vascular complications [38]. These MPO-mediated effects might result in the development of diabetic atherosclerosis.

In the current era of high potency statin therapy, coronary atherosclerosis still continued to progress even with intensive risk factor control in diabetic patients. The present study reveals that increased systemic MPO levels help to distinguish amongst those diabetics at higher risk for atherosclerosis progression. In addition, MPO level also enables to predict the response to anti-atherosclerotic therapy such as intensive LDL cholesterol control or high-dose statin in diabetic patients with CAD. This finding may identify MPO as an attractive target for pharmacological intervention, particularly within diabetic subjects. The present studies suggest that modifying systemic MPO activity within diabetic patients may be a viable pharmacologic strategy, and a targeted reduction of MPO activity, particularly within diabetics, may become feasible in the future.

A number of caveats should be noted with regard to the current analysis. 303 patients (26%) were excluded from 1184 patients in 3 previous IVUS trials due to a lack of available MPO data. These missing data might affect the current results. Our findings should be considered as hypothesis-generating and exploratory in nature because three IVUS trials included in the current analysis were not designed specifically to study the association between MPO levels and atheroma progression in diabetic patients. The current study focused on the contribution of only MPO level to atheroma progression in diabetic patients with CAD. As a variety of biomarkers have been considered to implicate in diabetic atherosclerosis, further investigation of other biomarkers associated with disease progression would be required. Despite this analysis being one of the largest available for serial IVUS imaging, the overall diabetic study population still remains relatively small. The present findings should be clarified in larger diabetic cohorts. Further, there are differences in each IVUS trial with regard to different pharmacological interventions and study population. While these heterogeneities across studies exist, all trials were conducted by the same group, using identical imaging protocols and with analysis by 1 core laboratory. The use of random effect in a mixed model statistical approach was applied to account for potential differences between studies. Nevertheless, we cannot exclude the possibility that there is residual heterogeneity in the analysis despite adopting these measures. As all patients had a diagnosis of CAD at the time of a clinically indicated angiography, it remains unknown if the current findings can be translated to the setting of primary prevention, or whether MPO levels are associated with increased atherosclerosis progression risk within subjects at risk for developing diabetes (i.e., with prediabetes). While a relationship between MPO level and atheroma progression was significant in diabetic patients, the difference of change in PAV was around 1% between diabetic patients with lower and higher MPO level, and thereby the ultimate translation of this finding to differences in cardiovascular events remains to be determined. Recently, we have demonstrated a direct relationship between the burden of coronary atherosclerosis, its progression, and adverse cardiovascular events [39]. In this study, given that even a 0.5% difference in progression rate identified a group with adverse clinical outcomes, our finding in IVUS measures of diabetic patients seems to have great

impact on clinical outcome. Thus, relatively small changes on ultrasound appear to associate with major differences in cardiovascular event rates.

In summary, the current study shows that systemic MPO levels are associated with accelerated coronary atheroma progression in diabetic patients with CAD. Intensive risk modification therapies are less effective for the prevention of atheroma progression in diabetic patients with an increased systemic MPO level. Collectively, the present findings suggest the MPO pathway may contribute to the development of accelerated atherosclerotic vascular disease observed in diabetes.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Stanley L. Hazen reports being listed as co-inventor on pending and issued patents held by the Cleveland Clinic relating to cardiovascular diagnostics; having been paid as a consultant or speaker for Abbott Laboratories, Inc, Bristol Myers Squibb, Cleveland Heart Lab, Inc, Esperion, Liposcience, Inc., Merck & Co, Inc, and Pfizer Inc; having received research funds from Abbott Laboratories, Inc, Cleveland Heart Lab, Inc., Esperion, and Liposcience Inc.; and having the right to receive royalty payments for inventions or discoveries related to cardiovascular diagnostics from Abbott Laboratories, Inc, Cleveland Heart Lab, Inc, Frantz Biomarkers, LLC, Liposcience, Inc, and Siemens.

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Abbreviation

CAD	coronary artery disease
DM	diabetes mellitus
EEM	external elastic membrane
HDL	high-density lipoprotein
hsCRP	high sensitivity C-reactive protein
IVUS	intravascular ultrasound
LDL	low-density lipoprotein
MPO	myeloperoxidase

PAV percent atheroma volume

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Figure 1-a **Figure 1-b** **Figure 1. Atheroma Progression in Non-diabetic and Diabetic Patients**

(a) Unadjusted changes in PAV in non-diabetic and diabetic patients stratified according to the quartiles of baseline MPO levels

Left panel: before adjustments for baseline and change in LDL-C, diastolic blood pressure and concomitant insulin use in non-diabetic patients

Right panel: before adjustments for baseline and change in LDL-C, diastolic blood pressure and concomitant insulin use in diabetic patients

(b) Adjusted changes in PAV in diabetic patients stratified according to the quartiles of baseline MPO levels

Change in PAV is shown in least-square mean.

LDL-C = low-density lipoprotein cholesterol, MPO = myeloperoxidase, PAV = percent atheroma volume

Figure 2. LDL-C Control, High-dose Statin Therapy and Atheroma Progression in Diabetic Patients

Changes in PAV in diabetic patients with baseline low or high MPO level

Left panel: stratified according to on-treatment levels of low-density lipoprotein cholesterol (LDL-C) < or ≥ 80mg/dl

Right panel: stratified according to low or high dose statin use at follow-up.

Change in PAV is shown in least-square mean.

LDL-C = low-density lipoprotein cholesterol, MPO = myeloperoxidase, PAV = percent atheroma volume

Table 1

Baseline Clinical Characteristics in Non-Diabetic Patients

	1st quartile of MPO (2-773.4 pmol/L)	2nd quartile of MPO (774.9-1359.8pmol/L)	3rd quartile of MPO (1365-2263.4 pmol/L)	4th quartile of MPO (2277.2-23018.5pmol/L)	p-value for trend
age (years)	57.9 ± 9.0	59.4 ± 9.9	56.3 ± 9.7	57.2 ± 10.2	0.19
female (%)	30.0	33.3	23.4	22.4	0.02
BMI	29.7 ± 5.0	29.6 ± 5.2	30.2 ± 5.6	30.6 ± 6.3	0.09
Hypertension (%)	60.6	69.0	66.1	64.1	0.32
Hyperlipidemia (%)	90.6	90.1	91.8	84.1	0.05
current tobacco use (%)	16.5	20.5	23.7	34.9	<0.001
Medication					
statin (%)	77.6	68.4	73.7	69.4	0.10
high-dose statin (%)	23.0	19.2	25.4	24.2	0.35
beta-blocker (%)	71.2	73.7	78.4	77.1	0.07
ACE inhibitor (%)	40.0	29.8	32.7	35.9	0.28
Biochemistry Data					
fasting glucose (mg/dl)	98.6 ± 17.2	96.9 ± 13.4	97.5 ± 17.5	93.2 ± 14.6	0.01
LDL cholesterol (mg/dl)	127.0 ± 39.0	120.0 ± 39.9	109.8 ± 36.9	105.9 ± 33.3	<0.001
HDL cholesterol (mg/dl)	44.8 ± 12.8	44.3 ± 11.8	40.9 ± 11.4	41.4 ± 10.7	0.001
Triglyceride (mg/dl) *	158.0 (103.2, 204.0)	143.0 (106.2, 205.0)	156.0 (106.2, 221.3)	141.8 (107.0, 201.0)	0.75
hsCRP (mg/dL) *	2.4 (1.2, 5.4)	2.4 (1.0, 5.7)	2.3 (1.1, 5.1)	3.7 (1.8, 7.4)	0.01
baseline PAV (%)	39.2 ± 9.5	38.2 ± 10.4	38.6 ± 9.2	39.3 ± 10.4	0.81

ACE = angiotensin-converting enzyme, BMI = body mass index, hsCRP = high sensitivity C-reactive protein, HDL = high-density lipoprotein, LDL = low-density lipoprotein,

* Median (Interquartile range)

Table 2

Baseline Clinical Characteristics in Diabetic Patients

	1st quartile of MPO (85576.5 pmol/L)	2nd quartile of MPO (583.5-1243.6 pmol/L)	3rd quartile of MPO (1255.3-2282.8 pmol/L)	4th quartile of MPO (2293.7-16739.5 pmol/L)	p-value for trend
age (years)	58.6 ± 9.1	57.8 ± 8.2	59.1 ± 10.5	57.9 ± 9.6	0.87
female (%)	30.6	30.0	40.0	20.0	0.23
BMI	32.6 ± 5.4	33.5 ± 6.7	33.3 ± 7.1	31.7 ± 4.5	0.46
Hypertension (%)	87.8	90.0	84.0	76.0	0.04
Hyperlipidemia (%)	100.0	94.0	94.0	92.0	0.04
current tobacco use (%)	18.4	16.0	18.0	22.0	0.30
Medication					
statin (%)	79.6	68.0	74.0	70.0	0.21
high-dose statin (%)	33.3	19.0	16.0	26.3	0.32
beta-blocker (%)	65.3	74.0	68.0	66.0	0.44
ACE inhibitor (%)	61.2	60.0	66.0	42.0	0.05
pioglitazone (%)	20.4	10.0	26.0	8.0	0.18
insulin (%)	36.7	30.0	32.0	30.0	0.27
Biochemistry					
fasting glucose (mg/dl)	152.8 ± 48.1	144.4 ± 43.3	160.2 ± 75.8	150.5 ± 72.1	0.66
LDL cholesterol (mg/dl)	122.5 ± 41.8	107.6 ± 37.8	107.0 ± 34.5	102.7 ± 44.6	0.02
HDL cholesterol (mg/dl)	40.1 ± 8.7	41.4 ± 13.2	40.5 ± 10.5	38.8 ± 9.5	0.50
triglyceride (mg/dl) *	161.2 (118.5, 251.5)	197.5 (141.6, 276.0)	201.5 (139.0, 265.0)	180.0 (123.9, 256.7)	0.81
hsCRP (mg/dL) *	2.9 (1.1, 5.6)	2.7 (1.1, 6.1)	3.8 (1.4, 7.2)	3.8 (1.6, 8.9)	0.05
baseline PAV (%)	40.8 ± 9.6	41.2 ± 8.6	41.8 ± 10.0	44.5 ± 8.9	0.05

ACE = angiotensin-converting enzyme, BMI = body mass index, hsCRP = high sensitivity C-reactive protein, HDL = high-density lipoprotein, LDL = low-density lipoprotein, PAV = percent atheroma volume,

* Median (Interquartile range)