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The Association of Serum Lipids and Inflammatory Biomarkers with Renal Function in Men with Type 2 Diabetes Mellitus

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Abstract

Dyslipidemia and inflammation may promote renal disease via mechanisms of vascular endothelial cell dysfunction in type 2 diabetes mellitus (DM). Sparse data, however, are available on the relation of lipids and inflammatory biomarkers and glomerular filtration rate (GFR) in type 2 DM. We performed a cross-sectional study of 732 men with type 2 DM enrolled in the Health Professionals' Follow-Up Study. Plasma creatinine was used to estimate GFR by the simplified MDRD equation.

In men with a GFR < 60 ml/min/1.73 m^2, triglycerides, non-HDL, apoprotein B, fibrinogen, sTNFR-2, and VCAM were significantly higher when compared to the referent group (GFR ≥ 90 ml/min/1.73 m^2). In multivariable logistic regression, those in the highest quartiles of the following biomarkers had an increased odds for having a GFR < 60 ml/min/1.73 m^2 when compared to those in the lowest quartiles: triglycerides (OR 3.11; 95% CI, 1.52 to 6.36), fibrinogen (OR 5.40; 95% CI 2.14 to 13.65), sTNFR-2 (OR 8.34; 95% CI 3.50 to 19.88), and VCAM (OR 4.50; 95% CI 1.98 to 10.23). An inverse association was observed for HDL (OR 0.48; 95% CI 0.24 to 0.98). We found no association between C-reactive protein and GFR. The results were similar when creatinine clearance by Cockcroft-Gault was used to estimate kidney function.

We conclude that several potentially modifiable lipid and inflammatory biomarkers are elevated in the setting of moderately decreased GFR in men with type 2 DM and may be the link between renal insufficiency and increased risk for cardiovascular events in this population.

BACKGROUND

Studies have generally suggested a positive association between dyslipidemia and inflammation and end-stage renal disease (ESRD) [1–3] or advanced chronic kidney failure [4,5], but the relation between these biomarkers and mild or moderate renal dysfunction has not been well characterized, especially in type 2 diabetes mellitus (DM). Type 2 DM is now recognized as an inflammatory condition associated with insulin resistance [6] and abnormal endothelial vascular reactivity [7]. As the leading cause of kidney disease [8] and an important cause of cardiovascular disease (CVD) in the Western world, type 2 DM is an increasingly prevalent etiology of microvascular and macrovascular disease. A chronic, systemic inflammatory state has been proposed to underline this increased risk for atherosclerotic disease, including renal dysfunction and CVD; however, investigations have not focused on

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the relation between glomerular filtration rate and dyslipidemia or inflammatory biomarkers in individuals with type 2 diabetes. Identification of such serologic markers may reveal new approaches to the prevention of progressive renal insufficiency and CVD in this population.

We undertook a cross-sectional study to examine the relation between renal function and 1) serum lipid levels (total cholesterol, low density lipoprotein, triglycerides, high density lipoprotein, lipoprotein-a, and apoprotein B), and 2) markers of inflammation including C-reactive protein (CRP), fibrinogen, soluble tumor necrosis factor receptor (TNFR-2), intracellular adhesion molecule-1 (ICAM), and vascular cell adhesion molecule-1 (VCAM) in 732 male subjects with type 2 diabetes.

METHODS

Source population

The Health Professionals' Follow-Up Study (HPFS) was established in 1986 when 51,529 U.S. male health professionals, aged 40 to 75 at study initiation**,** returned a mailed questionnaire providing information about diet, lifestyle factors, and medical history [9]. Participants were mailed follow-up questionnaires every two years to update information. In 1993–1994, blood samples were collected and frozen (−130° C) from a subset of these participants (n=18,159) as previously described [10]. Internal review has shown no demographic or clinical differences at baseline between men who provide blood samples and those who did not. Diabetes mellitus (DM) was first identified by self-report on a biennial questionnaire and confirmed by a Diabetes Supplemental Questionnaire (DSQ) in 2000; the validity of the DSQ in confirming DM has been demonstrated in the HPFS cohort [11]. The HPFS DM blood cohort consists of 1000 men (a number occurring by chance) with confirmed diabetes at baseline or during follow-up through 1998 who provided a blood sample in 1993–1994. Exclusion criteria for the current analyses were as follows: a) age of onset of DM ≤ 25 years of age (to attempt to restrict the study to type 2 DM) (n=28), b) subjects with a reported date of DM diagnosis after the date of blood draw (n=228), c) participants who reported on the DSQ that they were on dialysis (n=9) or had a kidney transplant (n=3). After these exclusions, 732 subjects were available for analysis.

Assessment of serum lipids, inflammatory biomarkers, hemoglobin A1c, and creatinine

Serum lipids, inflammatory biomarkers, hemoglobin A1c, and creatinine were measured at the Boston Children's Hospital Laboratory (Nader Rifai, director). Non-HDL cholesterol was defined as the difference between total cholesterol and HDL cholesterol. Hypercholesterolemia was defined as total cholesterol ≥ 200 mg/dl. The specific methodology for these assays have been previously reported [12] The coefficients of variation for all of the assays were ≤ 8% except for: a) C-reactive protein (14.8%) and b) creatinine (22.2%).

Assessment of renal function

Renal function was estimated by the simplified MDRD equation where GFR (ml/min/1.73 m^2) =186 × [PCr (mg/dl)]$^{-1.154}$ × [Age]$^{-0.203}$ × [1.21 if subject is black] [13] and the Cockcroft-Gault equation where CrCl=([140-age (years) × weight (kg)]/Pcr × 72) [14]. Because the direction and magnitude of associations between biomarkers and either MDRD-GFR or CrCl were consistently very similar, only the data using the MDRD-GFR are reported here. No data on proteinuria were available since no urine has been collected from this cohort.

Assessment of covariates

Race and height were initially reported on the 1986 questionnaire. Weight, smoking status, hypertension, and medication use from the 1994 questionnaire were used. Body mass index in

1994 was calculated by [weight (kg)/(height (m))^2]. A weekly metabolic-equivalent (MET) score was calculated from the physical activity section on the 1994 biennial questionnaire. Cardiovascular disease (myocardial infarction, coronary artery bypass grafting, or angina) (n=194) [15] and cancer diagnoses (n=31) [16,17] have also been previously validated in this cohort.

Statistical analyses

Most biomarkers were not normally distributed, therefore lipid and inflammatory biomarker levels stratified by level of GFR are reported as medians (5^{th} to 95^{th} percentile range). The Wilcoxon Signed-Rank test was used for inter-group comparisons with GFR ≥ 90 ml/min/1.73 m^2 as the referent group. Spearman correlation coefficients were calculated for each pair of biomarkers.

The dependent variable of estimated GFR was normally distributed. Linear regression was used to assess the difference in renal function across biomarker quartiles. Logistic regression was used to calculate odds ratios for GFR < 60 ml/min/1.73 m^2. Multivariable models were adjusted for age (continuous, years), hypertension (yes/no), BMI (continuous), cigarette smoking status (never, past, current), physical activity (quartiles, METS/week), duration of type 2 DM (quartiles, years), measured HgbA1c (quartiles), cardiovascular disease (yes/no), and cancer (yes/no). All analyses were performed with SAS software, version 8.2 (SAS Institute, Inc., Cary, North Carolina).

RESULTS

Demographic and clinical information for 732 men with type 2 diabetes in 1994 are shown in Table 1. The mean age of diabetes diagnosis was 54.1 years and the mean duration from time of initial diabetes diagnosis was 11.3 years. In this cohort, 54.2% had hypertension, 8.2% were taking ACE-inhibitor medication, and 6.6% were taking a statin medication. Mean estimated GFR was 79 ml/min/1.73 m^2. When the variables in Table 1 were stratified by estimated GFR levels, the men with GFR < 60 ml/min/1.73 m^2 were older (68 vs. 62.6 years, p<0.001) and had lower activity levels (21 vs. 32.5 mets/week, p=0.02) and a slightly lower BMI (27.9 vs. 28.3, p=0.02), lower HgbA1c (7.0% vs. 7.7%, p=0.008), were older at time of DM diagnosis (56 vs. 52 years, p<0.001), and had higher serum creatinine (1.5 vs. 0.8 mg/dl, p<0.0001) when compared to those in the referent group (GFR > 90 ml/min/1.73 m^2).

The median levels of measured lipid and inflammatory biomarkers according to GFR category are shown in Table 2. Levels of triglycerides, fibrinogen, sTNFR-2, and VCAM were consistently inversely related to GFR. Whereas levels of total cholesterol, LDL, non-HDL, and apoprotein B at lower GFR levels were higher than in the referent group of GFR ≥ 90 ml/min/1.73 m^2, these biomarker levels did not consistently increase across the categories of decreasing GFR.

By Spearman correlation, GFR was inversely and significantly correlated with sTNFR-2 (r=−0.39, p<0.0001) and VCAM (r=−0.23, p<0001). Statistically significant but weaker associations were also observed between GFR and total cholesterol (r=−0.09, p=0.004), triglycerides (r=−0.10, p=0.01), non-HDL (r=−0.10, p=0.008), lipoprotein a (r=−0.09, p=0.02), apoprotein B (r=−0.13, p=0.0006), and CRP (r=−0.09, p=0.02). The remaining biomarkers were not statistically associated with GFR by Spearman analysis.

We compared the differences in GFR between the highest and lowest quartiles of each of these biomarkers. Multivariable adjusted linear regression revealed the GFR was significantly lower by −4.2 to −7.8 ml/min/1.73 m^2 among those men in the highest quartiles of total cholesterol, non-HDL, triglycerides, and apoprotein B. Greater differences in GFR of −8.6 to −15.4 ml/

min/1.73 m^2 were noted in comparing the extreme quartiles of fibrinogen, sTNFR-2, and VCAM (Table 3). Because adding ACE-inhibitor and statin use did not significantly change the coefficients, they were removed from the final model. We also examined data for associations between aspirin use (reported as ≥2 times per week use on the 1994 HPFS questionnaire), biomarkers, and estimated GFR and found no association between those who reported use (n=354 or 48% of the cohort) and GFR<60 ml/min/1.73 m^2 (OR 1.08 [0.69, 1.68]). Odds ratios for individual biomarkers and presence of GFR<60 ml/min/1.73 m^2 did not change significantly when aspirin use was included in the model.

Multivariable logistic regression analysis was used to calculate the risk for having GFR of < 60 ml/min/1.73 m^2 when comparing the highest vs. lowest quartile of lipid and inflammatory biomarkers (Table 4). Those in the highest quartiles of triglycerides, fibrinogen, sTNFR-2, ICAM, or VCAM levels had a 2.07-fold to 8.34-fold increased odds of having a GFR < 60 ml/min/1.73 m^2 when compared to those in the lowest quartiles of these biomarkers. Moreover, the test for linear trend for these biomarkers was significant ($p \leq 0.01$), with the exception of ICAM (p=0.15). In addition, logistic regression was performed to estimate odds of GFR < 60 ml/min/1.73 m^2 with one standard deviation (SD) unit change of each biomarker. The odds were increased for triglycerides (OR 1.46 [1.18, 1.83]), fibrinogen (OR 1.50 [1.20, 1.87]), sTNFR-2 (OR 2.84 [2.18, 3.71]), and VCAM (OR 1.48 [1.20, 1.83]). Again, inclusion of ACE-inhibitor and statin use did not significantly change the results. Using the waist-to-hip ratio in place of BMI as a measure of adiposity in these multivariable models resulted in a change of <10% for the odds ratio for each of the biomarkers.

When compared to those in the lowest quartile, those in the highest quartile of HDL had a decreased odds (OR 0.48 [0.24, 0.98]) of having GFR < 60 ml/min/1.73 m^2. The association was marginally significant when HDL was examined per SD (OR 0.79, [0.60, 1.04]). LDL was inversely associated with GFR in the quartile age-adjusted analysis (OR 0.49 [0.25, 0.98]), and marginally significant in the multivariable model (OR 0.49 [0.24, 1.01]). On closer examination of the data, only 14 individuals who were in the highest quartile of LDL had a GFR of < 60 ml/min/1.73 m^2 as compared to 27 individuals in the lowest quartile of LDL who had a GFR of < 60 ml/min/1.73 m^2. In the multivariable logistic regression model with one SD unit change as the exposure, LDL was not significantly associated with GFR of < 60 ml/min/1.73 m^2 (OR 0.88 [0.70, 1.12]).

We noted that VCAM was positively correlated with ICAM (r=0.53, p<0.0001) and sTNFR-2 (r=0.57, p<0.0001). To examine the independent associations, we also included two correlated biomarkers (in quartiles) simultaneously in the multivariable logistic regression model with the outcome of a GFR < 60 ml/min/1.73 m^2. For example, when compared to the lowest quartile, the highest quartile of VCAM was no longer independently associated with GFR < 60 ml/min/1.73 m^2 (p=0.29) after adjusting for sTNFR-2 (OR 7.39 [2.81, 19.43]). When ICAM and VCAM were included in the same model, the highest quartile of ICAM was no longer associated with moderate renal dysfunction (p=0.88) while the highest quartile of VCAM remained significant when compared to the lowest quartile (OR 4.62 [1.87, 11.43]).

DISCUSSION

We observed important associations between lipid and inflammatory biomarkers and renal function in a cross-sectional study of 732 men with type 2 diabetes. Limited data are available regarding these biomarkers and renal function in type 2 diabetes; this paper reports that several lipid and inflammatory markers were independently associated with reduced renal function in adult onset diabetes. Recent investigations have reported that higher baseline levels of sTNFR-2, IL-6, CRP [18], E-selectin, and ICAM [19] are independent predictors of incident type 2 diabetes in prospective nested case-control analyses of the Nurses' Health Study. The

accumulating evidence that a chronic inflammation is a harbinger of adult-onset diabetes raises provocative questions of which, if any, of these same mechanisms lead to renal dysfunction in diabetes and whether any of these mechanisms also underlie non-diabetic nephropathy.

Overall, triglycerides, fibrinogen, sTNFR-2, and VCAM levels were consistently and strongly inversely associated with GFR in our study of men with type 2 diabetes, whereas HDL was positively correlated with renal function and CRP. These findings suggest that elevated risk for macrovascular and microvascular disease in type 2 diabetes may be progressive endothelial cell dysfunction and atherosclerosis mediated by elevated levels of these biomarkers.

In secondary analyses of data from 1785 participants of the MDRD study, investigators reported lower cholesterol levels at GFR<25 ml/min/1.73 m^2 [20], but direct comparison to our findings of no association between total cholesterol and GFR are difficult because the MDRD study excluded all but diet-controlled diabetics and encompassed a much lower range of GFR than in our current study. Most previous studies have found no relation between total cholesterol and kidney dysfunction or dialysis-dependency [1,21,22]. All of these studies, however, had less than 100 subjects and excluded diabetics. In contrast, several of these same studies reported higher triglyceride levels in subjects with kidney dysfunction or who were dialysis-dependent compared to controls [1,21,23]. Furthermore, a cross-sectional analysis of 5808 elderly participants in the Cardiovascular Heart Study (CHS) also revealed higher triglycerides levels in those with renal insufficiency (defined as sCr ≥1.5 mg/dl in men and ≥1.3 mg/dl in women) compared to those without renal insufficiency; participants with diabetes comprised only 12–14% of this cohort [4].

We also noted that higher HDL levels were more common in those without moderate renal insufficiency, defined as GFR < 60 ml/min/1.73 m^2. This observation is consistent with previous investigations demonstrating that those with kidney dysfunction have 11% to 32% lower HDL levels [1,4,21,23]. Our findings are also consistent with a recent prospective analysis of 4,517 members of Physicians' Health Study demonstrating that those with an HDL < 40 mg/dl at baseline had a two-fold higher risk of renal insufficiency (defined a serum creatinine ≥ 1.5 mg/dl) at 14 years of follow-up [5]. Since all of these previously published studies excluded diabetics or had <15% diabetics, our data confirm the inverse relation between HDL levels and renal dysfunction in type 2 diabetes.

Previously published studies on the association between LDL and presence of kidney disease have been mixed, with a couple of investigations reporting a positive association [21,23], and others reporting no association [1,4,22]. The finding that LDL was inversely associated with GFR < 60 ml/min/1.73 m^2 was opposite to our expectations especially because in our cohort, the highest quartile of LDL was associated with a −2.8 ml/min/1.73m^2 difference in estimated GFR when compared to the lowest quartile. On closer scrutiny, these OR estimates were based on only a few patients who fell within the highest quartile of LDL and had estimated GFR < 60 ml/min/1.73 m^2. LDL was not statistically associated with estimated GFR in any of our other analyses, and therefore, we felt this represented a chance finding.

We did not see significant or consistent associations between renal function and lipoprotein (a) (Lp (a)) or apoprotein B (Apo B). Whereas previous publications have reported higher Lp (a) in subjects with kidney disease, the studies either excluded individuals with diabetes or included diabetics who had much lower levels of renal function (CrCl 2 to 19 ml/min) [24] than those in our current study. We found no previously published literature on the relation between Apo B and kidney function. In contrast, we found fibrinogen levels were consistently inversely correlated with GFR. Fibrinogen has been noted to be positively associated with albuminuria in individuals with diabetes [25,26] and to be elevated in subjects with renal insufficiency [1,2,4].

We found no association between GFR and C-reactive protein (CRP) levels. Although some published investigations have found higher CRP levels in those with chronic renal failure or on dialysis compared to healthy controls, it should be noted that diabetic subjects were excluded [1–4] or comprised a minority (11 to 13%) of the study subjects [27,28]. Our results are consistent with those recently published from the MDRD study (comprised of only 5% non-insulin dependent diabetic subjects), which reported that age-stratified CRP levels in subjects with mean GFR 32.7 ml/min/1.73 m^2 (range 12 to 55 ml/min/1.73 m^2) approximated those in the general population as measured in the NHANES III study [29]. Of note, two studies looking at CRP specifically in people with diabetes have reported an association with albuminuria and higher CRP levels, but did not address the relation between CRP and renal function [25,26].

Tumor necrosis factor alpha (TNF-α) is an inflammatory cytokine with a short plasma half-life; therefore, the soluble form of the TNF receptor type 2 (sTNFR-2) is frequently used as a more stable surrogate marker. Since TNF-α is reported to be elevated in obese subjects [30], our multivariable models included BMI, and we observed a strong and consistent inverse association between sTNFR-2 and renal function. One small study of 64 pre-dialysis patients (mean sCr 6.0 mg/dl) also reported a two-fold elevation in TNF-α levels when compared with compared with 40 healthy controls [2]. The inverse association between GFR and sTNFR-2 may result from decreased clearance of this inflammatory cytokine in renal dysfunction as previously reported in a bilateral nephrectomized mouse model [31].

We found no consistent relation between ICAM and GFR. Higher ICAM levels in dialysis-dependent subjects have been reported [1,3,32], as well as in small studies of subjects with advanced renal dysfunction (mean sCr 6.0 mg/dl, range 1.3–6.7 mg/dl) [2,32]. The cross-sectional CHS investigation reported 6% higher ICAM levels in those with renal dysfunction compared with those without [4]. These CHS results may not be generalizable to subjects with better preserved kidney function since the mean CrCl in the renal dysfunction group was 39 ml/min, which is lower than that in our current cohort where only two subjects had a GFR < 30 ml/min/1.73 m^2. No previously published reports on ICAM and higher GFR levels were found.

We observed a strong and consistent inverse association between VCAM levels and GFR. Like ICAM, elevated VCAM levels have been observed in obese women [30] and in dialysis-dependent subjects [1,3,32]. Although one prospective observational study of 363 type 2 diabetics reported 1.5 to 2.0 increased relative risk for mortality in those with higher VCAM levels, it did not examine its relation to GFR [26]. Interestingly, although both VCAM and ICAM expression are upregulated in inflammatory states, ICAM is constitutively expressed at low levels on normal endothelial cells [33], and VCAM appears to be a more important factor in the initiation of atherosclerotic lesions [34]. Our study found an inverse relation between VCAM and renal function, which may suggest an intensifying atherosclerotic process in those with decreased GFR, in a large cohort of people with type 2 diabetes.

By entering combinations of two significant biomarkers simultaneously into regression models, we observed that VCAM was not an independent predictor of GFR < 60 ml/min/m^2 after controlling for sTNFR-2, suggesting that these molecules may be involved in the same inflammatory pathway. TNF-α infusion stimulates VCAM expression in the endothelial cells from a rabbit aorta [35], but we found no clinical human studies examining this relation.

Limitations to our study include the cross-sectional design, which limits conclusions about mechanism or temporal relation. A prospective study would provide information about how these biomarkers may change with declining kidney function over time; however, the lack of previous data on how these biomarkers relate to GFR in diabetes makes these findings informative. The rare use of ACE inhibitors and statin medications in 1994 in this cohort may

limit the generalizability of our results under current medical practices since these therapies are much more prevalent today in diabetics. Importantly, however, our study allows for a purer physiological examination of the association between GFR and biomarkers, since ACE-inhibitors can elevate serum creatinine and statins usually lower blood lipids. Unfortunately, no information is available on measured albuminuria, which is considered a marker for generalized endothelial vascular injury in the kidney as well as nephrotic range proteinuria, which can increase total cholesterol and LDL. Lastly, renal function was estimated from creatinine –based prediction equations since radio-isotope clearance GFR measurements would have been difficult and impractical in this cohort of >700 men. Estimated CrCl using the Cockcroft-Gault formula has been shown to be as accurate as creatinine clearance measured by 24 hour urine collections in diabetics [36]. Whereas we chose to present the kidney function as GFR estimated by the simplified MDRD equation, the results were very similar when CrCl by Cockcroft-Gault was invoked. There is a growing literature on underestimation of true measured GFR by the MDRD equations in people with serum creatinine values in the normal range [37–39]. We used categories of estimated GFR rather than GFR as a continuous variable, and therefore the relative association between these biomarkers and estimated GFR < 60 ml/min/1.73 m^2 should not be influenced by a systematic bias in the estimation equation. Moreover, the associations see were consistent whether GFR was a continuous (Table 3) or a categorical (Tables 2 and 4) variable. The relatively high CV of plasma creatinine in this study would presumably result in random misclassification and bias the results towards the null.

In conclusion, triglycerides, fibrinogen, sTNFR-2, and VCAM levels are inversely associated with GFR in a large cohort of men with type 2 diabetes, while HDL is positively correlated with renal function. The potential impact of drug therapies on altering these biomarkers and thereby slowing the progression of renal dysfunction is of great clinical interest and importance. Further investigation is also needed to determine whether the elevated levels of triglycerides and inflammatory biomarkers are due to increased production, decreased degradation, decreased excretion, or some combination in the setting of declining renal function. Nevertheless, an exciting new and more focused picture of elevated lipids and chronic endothelial inflammation is emerging as the potential mechanism behind the manifold risk of renal dysfunction and CVD in individuals with type 2 diabetes.

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Table 1

Demographic and Clinical Characteristics in 1994 of 732 Type 2 Diabetics in the Health Professionals Follow-Up Study (HPFS)

Age (years)	65.5±7.9 (47–80)
Race	
Caucasian	675 (92.2)
African-American	11 (1.5)
Other	13 (1.8)
Missing	28 (3.8)
Hypertension	397 (54.2)
Weight (kg)	88.1±16.2 (56.8–210.9)
Body Mass Index (BMI) (kg/m^2)	27.8±4.4 (18.3–56.5)
BMI categories (kg/m^2)	
<22	33 (4.5)
22–24.9	160 (21.9)
25–27.9	228 (31.1)
28–29.9	132 (18.0)
>30	179 (24.5)
Activity (mets/week)	29.9±33.2 (0–228.8)
Cigarette smoking	
Current	42 (5.7)
Past	392 (53.6)
Never	261 (35.7)
Missing	37 (5.0)
Age at diabetes diagnosis (years)	54.1+10.4 (26–78)
Duration of Type 2 diabetes (years)	11.3±9.2 (0.1–43.8)
Measured HgbA1c (%)	7.5±1.6 (4.8–15.6)
ACE-inhibitor medication use	60 (8.2)
Statin medication use	48 (6.6)
Measured serum creatinine (mg/dl)	1.1±0.2 (0.4–2.9)
GFR (ml/min/1.73 m^2)	79±18 (23–183)

Results expressed as mean ± SD (range 0 to 100%) or no. (%)

Hypertension was self-reported as “yes” or “no” on the 1994 questionnaire

ACE = Angiotensin-converting enzyme

GFR= Glomerular filtration rate

Table 2

Median levels of lipid and inflammatory biomarkers stratified by renal function

BIOMARKER	Calculated GFR (ml/min/1.73 m^2)			
	≥90 (n=183)	75–89 (n=235)	60–74 (n=224)	<60 (n=90)
Total Cholesterol (mg/dL)	200 (137–275)	***210 (147–279)*** **	***209 (157–287)*** **	209 (148–282)
LDL (mg/dL)	123 (64–177)	***131 (76–185)*** **	***126(69–193)*** *	122(62–192)
HDL (mg/dL)	39 (25–61)	39 (28–63)	39 (26–65)	37 (25–57)
Non-HDL (mg/dL)	159 (103–235)	***170 (111–233)*** **	***168 (111–242)*** **	***168 (115–239)*** *
TG (mg/dL)	157 (63–396)	165 (65–346)	168 (65–385)	***182 (81–408)*** *
Lipoprotein-a (mg/dL)	8.7 (0.7–64.4)	8.5 (0.8–78.8)	9.3 (0.7–75.8)	8.5 (0.7–61.1)
Apoprotein B (mg/dL)	99 (68–140)	***106 (69–145)*** **	***104 (69–150)*** **	***107 (74–143)*** **
C-reactive protein (mg/dL)	0.17 (0.04–1.41)	0.17 (0.03–1.16)	0.18 (0.03–1.41)	0.21 (0.04–1.17)
Fibrinogen (mg/dL)	415 (301–676)	***451 (301–686)*** *	***484 (338–713)*** ***	***513 (364–835)*** ***
sTNFR-2 (pg/mL)	2,525 (1,631–4,103)	***2,755 (1,838–4,433)*** **	***3,086 (2,080–4,598)*** ***	***3,918 (2,173–5,843)*** ***
ICAM (ng/mL)	338 (232–541)	343 (240–522)	344 (230–499)	359 (256–482)
VCAM (ng/mL)	1,254 (827–1,977)	1,340 (871–2,014)	***1,427 (893–2,141)*** ***	***1,540 (1,073–2,263)*** ***

Results expressed as median (5th–95th percentile values)

* p between 0.05 and 0.01,

** p between 0.01 and 0.001, and

*** p<0.001 for value compared to referent group (GFR ≥ 90 ml/min/1.73 m^2) by Wilcoxon Rank Sum test

LDL= low density lipoprotein, TG=triglycerides, HDL=high density lipoprotein, sTNFR-2= soluble tumor necrosis factor receptor-2, ICAM=intracellular adhesion molecule, VCAM=vascular cellular adhesion molecule

Table 3

Difference in GFR (ml/min/1.73 m^2) for those in the highest vs. the lowest quartile of lipid and inflammatory biomarkers

BIOMARKER	AGE-ADJUSTED [95% CI]	MULTIVARIABLE* [95% CI]
Total Cholesterol (mg/dL)	***−4.2 [−7.8, −0.5]***	***−5.2 [−8.9, −1.5]***
LDL (mg/dL)	−2.0 [−5.8, 1.7]	−2.8 [−6.5, 1.0]
HDL (mg/dL)	2.4 [−1.3, 6.1]	1.9 [−2.0, 5.8]
Non-HDL (mg/dL)	***−5.9 [−9.5, −2.2]***	***−7.1 [−10.8, −3.4]***
TG (mg/dL)	***−7.8 [−11.5, −4.2]***	***−8.4 [−12.2, −4.5]***
Lipoprotein-a (mg/dL)	1.1 [−2.6, 4.7]	1.0 [−2.7, 4.6]
Apoprotein B (mg/dL)	***−7.6 [−11.3, −4.0]***	***−8.0 [−11.7, −4.3]***
C-reactive protein (mg/dL)	−3.1 [−6.7, 0.6]	−3.1 [−7.2, 0.9]
Fibrinogen (mg/dL)	***−11.8 [−15.3, −8.3]***	***−11.2 [−14.9, −7.5]***
sTNFR-2 (pg/mL)	***−15.3 [−18.9, −11.8]***	***−15.4 [−19.1, −11.7]***
ICAM (ng/mL)	−3.5 [−71.7, 0.2]	−3.3 [−7.1, 0.5]
VCAM (ng/mL)	***−9.0 [−12.6, −5.4]***	***−8.6 [−12.2, −4.9]***

LDL= low density lipoprotein, TG=triglycerides, HDL=high density lipoprotein, sTNFR-2=soluble tumor necrosis factor receptor-2, ICAM=intracellular adhesion molecule, VCAM=vascular cellular adhesion molecule

Multivariable models are adjusted for age (continuous, years), hypertension (yes/no), BMI (continuous), cigarette smoking status (never, past, current), physical activity (quartiles, mets/week), duration of Type 2 DM (quartiles, years), measured HgbA1c (quartiles), cardiovascular disease (yes/no), and cancer (yes/no)

Table 4

Logistic regression odds ratios for highest vs. lowest quartile of lipid and inflammatory biomarkers for outcome of calculated GFR < 60 ml/min/1.73 m^2

BIOMARKER	AGE-AJUSTED OR [95% CI]	MULTIVARIABLE OR [95% CI]	p-for-trend
Total Cholesterol (mg/dL)	0.83 [0.43, 1.61]	0.86 [0.43, 1.73]	0.69
LDL (mg/dL)	***0.49 [0.25, 0.98]***	0.49 [0.24, 1.01]	***0.05***
HDL (mg/dL)	***0.43 [0.23, 0.82]***	***0.48 [0.24, 0.98]***	0.07
Non-HDL (mg/dL)	1.14 [0.59, 2.22]	1.13 [0.56, 2.29]	0.98
TG (mg/dL)	***2.68 [1.41, 5.09]***	***3.11 [1.52, 6.36]***	***0.01***
Lipoprotein-a (mg/dL)	1.02 [0.55, 1.87]	0.97 [0.51, 1.83]	0.90
Apoprotein B (mg/dL)	1.54 [0.79, 2.97]	1.48 [0.73, 2.98]	0.49
C-reactive protein (mg/dL)	1.24 [0.65, 2.35]	0.89 [0.44, 1.85]	0.99
Fibrinogen (mg/dL)	***6.25 [2.56, 15.28]***	***5.40 [2.14, 13.65]***	***0.003***
sTNFR-2 (pg/mL)	***9.36 [4.07, 21.51]***	***8.34 [3.50, 19.88]***	***<0.001***
ICAM (ng/mL)	***2.14 [1.09, 4.18]***	***2.07 [1.01, 4.26]***	0.15
VCAM (ng/mL)	***4.91 [2.21, 10.88]***	***4.50 [1.98, 10.23]***	***0.009***

LDL= low density lipoprotein, TG=triglycerides, HDL=high density lipoprotein, sTNFR-2= soluble tumor necrosis factor receptor-2, ICAM=intracellular adhesion molecule, VCAM=vascular cellular adhesion molecule

Multivariable models are adjusted for age (continuous, years), hypertension (yes/no), BMI (continuous), cigarette smoking status (never, past, current), physical activity (quartiles, mets/week), duration of Type 2 DM (quartiles, years), measured HgbA1c (quartiles), cardiovascular disease (yes/no), and cancer (yes/no)

p-for-trend is testing for linear trend across all 4 quartiles of biomarker