

Low HDL Cholesterol and the Risk of Diabetic Nephropathy and Retinopathy

Results of the ADVANCE study

JAMIE MORTON, MBBS^{1,2}
 SOPHIA ZOUNGAS, PHD^{3,4}
 QIANG LI, MBIOSTAT³
 ANUSHKA A. PATEL, PHD³
 JOHN CHALMERS, PHD³
 MARK WOODWARD, PHD³
 DAVID S. CELERMAJER, PHD^{1,2,5}

JOLINE W.J. BEULENS, PHD⁶
 RONALD P. STOLK, PHD^{6,7}
 PAUL GLASZIOU, PHD⁸
 MARTIN K.C. NG, PHD^{1,2,5}
 ON BEHALF OF THE ADVANCE
 COLLABORATIVE GROUP

OBJECTIVE—Although low HDL cholesterol (HDL-C) is an established risk factor for atherosclerosis, data on HDL-C and the risk of microvascular disease are limited. We tested the association between HDL-C and microvascular disease in a cohort of patients with type 2 diabetes.

RESEARCH DESIGN AND METHODS—A total of 11,140 patients with type 2 diabetes and at least one additional vascular risk factor were followed a median of 5 years. Cox proportional hazards models were used to assess the association between baseline HDL-C and the development of new or worsening microvascular disease, defined prospectively as a composite of renal and retinal events.

RESULTS—The mean baseline HDL-C level was 1.3 mmol/L (SD 0.45 mmol/L [range 0.1–4.0]). During follow-up, 32% of patients developed new or worsening microvascular disease, with 28% experiencing a renal event and 6% a retinal event. Compared with patients in the highest third, those in the lowest third had a 17% higher risk of microvascular disease (adjusted hazard ratio 1.17 [95% CI 1.06–1.28], $P = 0.001$) after adjustment for potential confounders and regression dilution. This was driven by a 19% higher risk of renal events (1.19 [1.08–1.32], $P = 0.0005$). There was no association between thirds of HDL-C and retinal events (1.01 [0.82–1.25], $P = 0.9$).

CONCLUSIONS—In patients with type 2 diabetes, HDL-C level is an independent risk factor for the development of microvascular disease affecting the kidney but not the retina.

Diabetes Care 35:2201–2206, 2012

Diabetes is the primary cause of end-stage kidney disease (1) and loss of vision (2) in developed nations. Microvascular disease is a common complication of type 2 diabetes and develops insidiously with few symptoms until irreversible damage has occurred. The two principal and reversible risk factors for

the development and progression of nephropathy and retinopathy are blood glucose and blood pressure levels (1,2). However, despite the benefits seen with control of these two risk factors, considerable residual risk remains. Identifying additional risk factors for these common complications could aid the tailoring of

risk assessment and development of novel therapeutic strategies.

Reduced HDL cholesterol (HDL-C), characteristic of type 2 diabetic dyslipidaemia (3), is a well-recognized risk factor for macrovascular complications (4). We hypothesized that lower HDL-C levels also may predispose to the development and progression of diabetic microvascular disease. In subjects without diabetes, low HDL-C has been previously reported to be an independent risk factor for the development of chronic kidney disease (CKD) (5–7), but there are limited prospective data on the relationship between HDL-C and the risk of diabetic nephropathy (8–12). There are even fewer data on the relationship between HDL-C levels and retinopathy, with conflicting results in nondiabetic patients (13–15), and no significant association found in those with type 2 diabetes (16–20). Despite the paucity of epidemiological evidence, two large randomized trials have recently reported that fenofibrate, an HDL-C-modifying agent, reduces diabetes-related microvascular disease (21–23).

The Action in Diabetes and Vascular Disease: preterAx and diamicroN-MR Controlled Evaluation (ADVANCE) Study is the largest trial to date of glycemic control and blood pressure lowering in patients with type 2 diabetes at high risk for vascular events (24). The ADVANCE study enrolled >11,000 patients with type 2 diabetes and followed them systematically for the development of microvascular complications. In these analyses, we evaluate baseline HDL-C level as a risk factor for the development of new or worsening microvascular disease, defined as a composite of new or worsening retinopathy and nephropathy.

RESEARCH DESIGN AND METHODS

The study design of the ADVANCE study is reported in detail elsewhere (24). In brief, 11,140 patients with type 2 diabetes, aged at least 55 years at study entry and with at least one other cardiovascular risk factor, underwent factorial randomization to 1) the fixed combination of perindopril and indapamide (4 mg/1.25 mg) or matching placebo and 2) intensive

From ¹The Heart Research Institute, Sydney, New South Wales, Australia; the ²Sydney Medical School, University of Sydney, Sydney, New South Wales, Australia; ³The George Institute for Global Health, University of Sydney, Sydney, New South Wales, Australia; the ⁴School of Public Health, Monash University, Melbourne, Victoria, Australia; the ⁵Department of Cardiology, Royal Prince Alfred Hospital, Sydney, New South Wales, Australia; the ⁶Julius Centre for Health Sciences and Primary Care, University Medical Centre Utrecht, Utrecht, the Netherlands; the ⁷Department of Epidemiology, University Medical Centre Groningen, Groningen, the Netherlands; the ⁸Department of Clinical Epidemiology and Biostatistics, Bond University, Gold Coast, Queensland, Australia.

Corresponding authors: Sophia Zoungas, szoungas@georgeinstitute.org.au, and Martin K.C. Ng, mkcng@med.usyd.edu.au.

Received 12 February 2012 and accepted 2 May 2012.

DOI: 10.2337/dc12-0306. Clinical trial reg. no. NCT00145925, clinicaltrials.gov.

This article contains Supplementary Data online at <http://care.diabetesjournals.org/lookup/suppl/doi:10.2337/dc12-0306/-/DC1>.

© 2012 by the American Diabetes Association. Readers may use this article as long as the work is properly cited, the use is educational and not for profit, and the work is not altered. See <http://creativecommons.org/licenses/by-nc-nd/3.0/> for details.

(gliclazide MR-based, to an HbA_{1c} target of $\leq 6.5\%$) or standard (usual care) glucose control. Patients were followed-up for a median of 5.0 years. The ADVANCE trial enrolled subjects from 20 countries in Asia, Australasia, Europe, and North America. A total of 11,126 patients with HDL-C measurements at baseline were included in these analyses. From this population, 1,602 patients were included in the ADVANCE Retinal Measurement (AdRem) study, which involved serial retinal photography (1,241 had assessable images). Study design and the methods used in AdRem are published in detail elsewhere (25). Approval for the study was obtained from each center's institutional ethics committee, and all participants provided written informed consent.

Baseline and follow-up analysis

At baseline, venous blood was taken for a fasting lipid profile, HbA_{1c}, and creatinine. Blood pressure and BMI also were recorded. Creatinine was remeasured at 4 months and then annually. In addition, urine samples were collected at baseline, 24 months, 48 months, and at the end of the trial for determination of the urinary albumin-to-creatinine ratio (ACR). Fundoscopy also was performed at these times. HDL-C level was repeated at 24 and 48 months or at the end of the study. All samples were analyzed by local laboratories, and all assessments could be repeated at physician discretion. All nonstudy medication was at the discretion of the local physician. Participants in AdRem had retinal stereoscopic photographs taken after the ADVANCE trial randomization visit and at the final visit of the blood pressure arm of the trial. All images were graded centrally according to the Early Treatment of Diabetic Retinopathy Study classification as modified in the UK Prospective Diabetes Study (26).

Outcomes

The main outcomes for this analysis were microvascular events, defined as a composite of total renal and retinal events. Total renal events were defined as the development of new microalbuminuria (urinary ACR 30–300 $\mu\text{g}/\text{mg}$), new macroalbuminuria (urinary ACR of $>300 \mu\text{g}/\text{mg}$), a doubling of creatinine to at least 200 $\mu\text{mol}/\text{L}$, the need for renal replacement therapy, or death as a result of renal disease. Total retinal events were defined as the development of proliferative retinopathy (new blood vessels or fibrous proliferations on the disc or elsewhere, vitreous hemorrhage),

or preretinal hemorrhage), macular edema, diabetes-related blindness, or the use of retinal photocoagulation therapy. Participants could record more than one event relating to renal and retinal disease, but only the first event was analyzed for total microvascular events.

Statistics

Differences in variables at baseline between subgroups of the study population were tested using the Student *t* test, the Mann-Whitney test, or the χ^2 test, as appropriate. Participants were censored at their date of death or, for those still alive at the end of follow-up, the date of their last visit. The regression dilution bias in HDL-C was assessed using a linear mixed model, with HDL-C during follow-up as the outcome and baseline HDL-C as the predictor. To calculate the correction factor, HDL-C measurements after microvascular events were excluded (27). The risks of events associated with baseline HDL-C level were estimated using Cox proportional hazards models, with adjustment for potential confounding baseline covariates including age (continuous), sex (male/female), ethnicity (white/Asian/other), treatment groups (standard vs. intensive glucose control and placebo vs. fixed-dose blood pressure-lowering treatment), history of microvascular disease (yes/no), smoking status (current/previous/never), current alcohol consumption (yes/no), HbA_{1c} (continuous), BMI (continuous), systolic blood pressure (continuous), diabetes duration (continuous), total cholesterol (continuous), creatinine (continuous), and statin use (yes/no). The selection of variables was based on identifying all measured clinical variables of known or suspected prognostic importance for the outcomes of interest. The assumption of the proportional hazards was checked graphically using the log cumulative hazard plot for all variables included in the Cox model. The primary analyses compared patients in the highest with those in the lowest third (or tertile group) of HDL-C. Additional sensitivity analyses were performed comparing the risks in those in the lowest with those in the highest fourths (or quartile groups), excluding patients with microvascular disease, renal disease, or retinal disease at baseline and examining subgroups defined by age, sex, ethnicity, systolic blood pressure, BMI, and age. All *P* values were calculated from two-tailed tests of statistical significance with a type I error rate of 5%. All analyses were performed using SAS version 9.1 (SAS Institute, Cary, NC).

RESULTS—The ADVANCE study enrolled patients with type 2 diabetes at high risk for macrovascular events. At baseline, 10% ($n = 1,155$) had evidence of microvascular disease. During a median follow-up period of 5 years, 32% ($n = 3,585$) of participants developed new or worsening microvascular disease. Almost one-third ($n = 3,161$ [28%]) developed a renal event: microalbuminuria was the most frequent event, followed by macroalbuminuria, doubling of creatinine to at least 200 $\mu\text{mol}/\text{L}$, renal-related death, and the need for renal replacement therapy (25, 3.4, 1.2, 0.3, and 0.2%, respectively). A retinal event occurred in 6% ($n = 680$): the need for laser therapy was the most frequent event, followed by macular edema, proliferative retinopathy, and diabetes-related blindness (3.5, 3.2, 2.6, and 0.4%, respectively).

The mean baseline HDL-C level was 1.3 mmol/L (SD 0.45 mmol/L [range 0.1–4.0]). Table 1 shows the distribution of variables by thirds of baseline HDL-C level. Compared with the lowest third of HDL-C, those subjects in the highest third were less likely to be male (44.8 vs. 70.8%) or taking a statin (24.9 vs. 31.1%) and more likely to have lower BMI (27.7 vs. 29.1 kg/m^2), lower serum creatinine (82.5 vs. 91.0 $\mu\text{mol}/\text{L}$), and to have never smoked (65.3 vs. 49.6%). Low HDL-C also was associated with higher triglycerides and lower total cholesterol and LDL cholesterol. Age, diabetes duration, systolic blood pressure, HbA_{1c}, ethnicity, and nonstatin lipid-lowering medication use all were statistically significantly different between thirds; however, the absolute differences were very small. The relative differences between HDL groups and the baseline variables of statin use, nonstatin lipid-lowering medication use, systolic blood pressure, and HbA_{1c} remained stable over the course of study follow-up (Supplementary Table 1).

Table 2 shows the association, both adjusted and unadjusted, between baseline thirds of HDL-C and the risk of developing new or worsening microvascular disease. Compared with the highest third of HDL-C, those in the lowest third had an 11% higher risk of a microvascular event (unadjusted hazard ratio [HR] 1.11 [95% CI 1.02–1.20], $P = 0.01$). Following multivariable adjustment and taking into account regression dilution, the risk was 17% higher (1.17 [1.06–1.28], $P = 0.001$). This finding was driven by an adjusted 19% higher risk of a renal event (1.19 [1.08–1.32], $P = 0.0005$). There was a similar significantly higher

Table 1—Baseline variables of study subjects by thirds of serum HDL-C

	HDL thirds			P for trend
	Third 1 (<1.1 mmol/L)	Third 2 (1.1–1.34 mmol/L)	Third 3 (>1.34 mmol/L)	
n	3,497	3,924	3,705	
Age at baseline (years)	65.2 (6.4)	65.9 (6.5)	66.1 (6.3)	<0.0001
Male (%)	70.8 (2,475)	57.7 (2,264)	44.8 (1,661)	<0.0001
Diabetes duration (years)	7.7 (6.3)	8.0 (6.4)	8.2 (6.4)	0.001
BMI (kg/m ²)	29.1 (5.3)	28.3 (5.0)	27.7 (5.2)	<0.0001
Systolic blood pressure (mmHg)	144.3 (21.0)	144.9 (21.6)	145.8 (21.9)	0.003
HbA _{1c} (%)	7.6 (1.5)	7.5 (1.6)	7.5 (1.6)	0.01
Serum creatinine (μmol/L)	91.0 (26.9)	86.4 (23.5)	82.5 (25.1)	<0.0001
ACR (μg/mg)	54.9 (119.5)	51.5 (111.8)	51.4 (114.4)	0.2
Total cholesterol (mmol/L)	4.8 (1.2)	5.2 (1.1)	5.6 (1.2)	<0.0001
LDL (mmol/L)	2.9 (0.9)	3.1 (1.0)	3.2 (1.0)	<0.0001
HDL (mmol/L)	0.91 (0.12)	1.20 (0.08)	1.64 (0.29)	
Triglycerides (mmol/L)	2.3 (1.5)	1.9 (1.2)	1.7 (1.2)	<0.0001
Smoking status (%)				
Never	49.6 (1,734)	56.9 (2,232)	65.3 (2,418)	<0.0001
Former	32.9 (1,151)	27.5 (1,081)	22.4 (830)	
Current	17.5 (612)	15.6 (611)	12.3 (457)	
Current alcohol intake (%)				
No	68.4 (2,391)	69.9 (2,744)	70.2 (2,600)	0.2
Yes	31.6 (1,106)	30.1 (1,180)	29.8 (1,105)	
Ethnicity (%)				
White/European	64.7 (2,261)	59.8 (2,348)	55.8 (2,066)	<0.0001
Asian	33.1 (1,159)	38.5 (1,510)	42.3 (1,569)	
Other	2.2 (77)	1.7 (66)	1.9 (70)	
Statin use (%)				
No	68.9 (2,410)	71.1 (2,789)	75.1 (2,784)	<0.0001
Yes	31.1 (1,087)	28.9 (1,135)	24.9 (921)	
Nonstatin lipid-lowering medication (%)				
No	90.2 (3,156)	92.3 (3,620)	92.2 (3,416)	0.002
Yes	9.8 (341)	7.7 (304)	7.8 (289)	
History of microvascular disease (%)				
No	89.8 (3,140)	90.1 (3,537)	88.9 (3,294)	0.2
Yes	10.2 (357)	9.9 (387)	11.1 (411)	
Randomized glucose treatment (%)				
Standard	49.6 (1,736)	50.3 (1,972)	50.0 (1,853)	0.9
Intensive	50.4 (1,761)	49.7 (1,952)	50.0 (1,852)	
Randomized blood pressure treatment (%)				
Placebo	49.4 (1,728)	49.6 (1,947)	51.0 (1,891)	0.3
Perindopril-indapamide	50.6 (1,769)	50.4 (1,977)	49.0 (1,814)	

Data are means (SD) or % (n). Bold values are statistically significant values ($P \leq 0.05$). P values refer to the Student *t* test, the Mann-Whitney test, or the χ^2 test, as appropriate.

risk of developing new microalbuminuria and macroalbuminuria (1.14 [1.03–1.27], $P = 0.01$, and 1.42 [1.07–1.87], $P = 0.01$, respectively). Furthermore, we observed that those patients in the lowest third of HDL-C were more likely to maintain or progress to a worse category of urinary ACR over time, compared with those in the middle and highest third of HDL-C

(66.8 vs. 62.0 and 58.8%, respectively) (Supplementary Table 2).

The direction of the effect was similar, however not statistically significant following adjustment, to the other renal outcomes of doubling of creatinine to at least 200 μmol/L and renal-related death. In contrast to the other renal outcomes, the association with the need for renal

replacement therapy (the end point with the smallest number of events) was non-linear.

There was no association between baseline thirds of HDL-C and the development of retinopathy or any specific type of retinal event. Furthermore, there was no association between baseline HDL-C and a wide range of predefined retinal outcomes

Table 2—Association between baseline thirds of HDL-C (tertile [T] 1, T2, and T3) and subsequent development of microvascular disease

	Unadjusted			Multivariable adjusted*		
	T1 vs. T3 [‡]	P	T2 vs. T3 [‡]	P	T2 vs. T3 [‡]	P
Total microvascular events	1.11 (1.02–1.20)	0.01	1.09 (1.00–1.18)	0.04	1.17 (1.06–1.28)	0.001
Total kidney events	1.15 (1.06–1.26)	0.001	1.13 (1.04–1.23)	0.006	1.19 (1.08–1.32)	0.0005
New microalbuminuria	1.08 (0.98–1.18)	0.1	1.09 (1.00–1.20)	0.05	1.14 (1.03–1.27)	0.01
New macroalbuminuria	1.53 (1.19–1.97)	0.001	1.25 (0.97–1.62)	0.07	1.42 (1.07–1.87)	0.01
Doubling of creatinine to ≥ 200 $\mu\text{mol/L}$	1.71 (1.10–2.66)	0.02	1.33 (0.84–2.09)	0.2	1.44 (0.89–2.35)	0.1
Need for renal replacement therapy	0.80 (0.28–2.31)	0.7	1.54 (0.64–3.70)	0.3	0.74 (0.24–2.31)	0.6
Renal-related death	1.24 (0.56–2.78)	0.6	1.12 (0.50–2.50)	0.8	1.16 (0.47–2.89)	0.7
Total retinal events	0.90 (0.74–1.08)	0.3	0.93 (0.78–1.11)	0.4	1.01 (0.82–1.25)	0.9
New proliferative retinopathy	0.76 (0.57–1.02)	0.07	0.85 (0.64–1.11)	0.2	0.92 (0.67–1.26)	0.6
Macular edema	1.02 (0.79–1.33)	0.9	1.04 (0.81–1.34)	0.8	1.18 (0.88–1.58)	0.3
Diabetes-related blindness	0.71 (0.32–1.59)	0.4	1.19 (0.61–2.35)	0.6	1.03 (0.44–2.41)	0.9
Need for laser therapy	1.07 (0.84–1.38)	0.6	1.09 (0.85–1.39)	0.5	1.18 (0.89–1.55)	0.2

†Bold values are statistically significant values ($P \leq 0.05$). *Adjusted for regression dilution and baseline age, sex, ethnicity, treatment groups, history of microvascular disease, smoking status, current drinking, HbA_{1c}, BMI, systolic blood pressure, diabetes duration, statin use, and baseline creatinine, total cholesterol, and triglyceride levels. ‡Cox proportional HR (95% CI), highest third as reference.

from the AdRem substudy involving 1,241 participants who underwent assessable serial retinal photography, which included progression by the Early Treatment of Diabetic Retinopathy Study classification and the development of the individual signs of retinopathy (data not shown).

Sensitivity analyses by fourths (or quartile groups) of HDL-C revealed similar findings, namely an inverse relationship between HDL-C level and total microvascular events that was driven primarily by renal events, with no relationship found between HDL-C and retinal events. Sensitivity analyses excluding patients with baseline microvascular disease also were similar to the main analysis. When stratified by subgroups defined by age, sex, ethnicity, systolic blood pressure, BMI, or HbA_{1c}, there was no evidence of heterogeneity in the association.

CONCLUSIONS—This study is the largest prospective analysis specifically addressing HDL-C level and risk of microvascular disease in patients with type 2 diabetes. The main finding is that lower baseline HDL-C level is a significant and independent predictor of the development and progression of diabetic nephropathy. Compared with patients in the highest third, those in the lowest third of baseline HDL-C had a 19% higher risk of nephropathy, after adjustment for a wide range of potential confounders and accounting for regression dilution. In contrast, there was no association between baseline HDL-C and the risk of diabetic retinopathy. These findings suggest that differences exist in the pathophysiology between the two types of microvascular disease.

Our findings provide the strongest evidence to date for a role of HDL-C in the development and progression of diabetic nephropathy in patients with type 2 diabetes. Several small prospective studies have shown that low HDL-C predicts progression of microalbuminuria (8–10); however, others have not identified such an association (28,29). The largest previous study evaluating this question included 2,193 patients with type 2 diabetes and normal renal function at baseline. In this study, each 0.26 mmol/L higher level of HDL was associated with a 24% lower risk of developing stage 3 chronic kidney disease (11). This was not a prespecified end point of the ADVANCE study; however, we are the first to show an inverse relationship between baseline HDL-C and total renal microvascular events.

We observed no significant association between HDL-C and a whole spectrum of retinal complications. Although some non-HDL lipid fractions previously have been reported to be associated with the severity and progression of retinopathy in patients with type 2 diabetes, no study has shown an association with HDL-C (16,20), and our study is concordant with this literature. Our findings may be explained by the nature of the retinal events examined as the most frequent retinal end point was laser photocoagulation. Because this procedural event is likely to be driven by health service availability and local usual practice in addition to biological risk in a multicenter, international study such as this, its inclusion may have diluted any causal link, if any such association existed. However, we also found no evidence of an association with any of the specific end points examined in the small proportion of ADVANCE trial participants who underwent serial retinal photography, a more sensitive and objective measure of retinal disease.

The Fenofibrate Intervention in End Point Lowering in Diabetes Trial showed that fenofibrate reduced the development of renal dysfunction and the need for retinal laser therapy (22,30). In support of our findings of an association between HDL-C and renal events but not retinal events, fenofibrate seemed to be more protective against the estimated glomerular filtration rate loss in those with baseline dyslipidemia (defined as low HDL-C and high triglycerides) (30), whereas the retinal benefit was independent of any lipid fraction (22). In much the same way, the more recent Action to Control Cardiovascular Risk in Diabetes eye study showed a benefit of fenofibrate added to statin therapy on the progression of diabetic retinopathy, without an appreciable change in HDL-C level (HDL-C at 1 year 1.03 mmol/L in the fenofibrate group compared with 1.01 mmol/L in the placebo group) (23). The salient conclusion from these two studies of fenofibrate therapy is that treatment reduced both microvascular complications in type 2 diabetes. This effect, at least in part, may be related to HDL-C level for renal but not retinal complications. On the horizon, there is the promising cholesterylester transfer protein inhibitor, anacetrapib, which increases HDL-C by 138% (31). Our findings suggest that investigating the effect of HDL-C–modulating agents on the development of diabetic nephropathy is warranted.

The pathogenesis of diabetic microvascular disease is complex and involves the

interplay of endothelial dysfunction, advanced end-glycation products, oxidative stress, and the abnormal production of cytokines and growth factors (32,33). The pathobiology of diabetic nephropathy and retinopathy is heterogeneous, with no single characteristic lesion (33,34). Although retinopathy and nephropathy can coexist in the same patient, the association is less clear in type 2 rather than type 1 diabetes (35), perhaps an indication of differing pathophysiology. HDL-C itself is a complex of several lipoproteins, cholesterol, and triglycerides and has several postulated mechanisms of vascular action, notably reverse cholesterol transport as well as anti-inflammatory and antioxidant properties (36). Furthermore, there is emerging evidence that HDL-C improves glycemic control by modulating glucose uptake into skeletal muscle (37) and protects against islet β -cell dysfunction (38). Although the exact putative mechanism for protection is unknown, it is clear that HDL-C exerts beneficial effects on many of the pathways known to be detrimental to vascular and kidney biology, but how this could be different in retinopathy is unclear.

Following the development of microalbuminuria, 20–40% of patients with type 2 diabetes will progress to overt nephropathy (1). Many randomized trials have shown a clear benefit for the treatment of this asymptomatic condition; thus, early detection is critical (39). International guidelines recommend screening at the time of diagnosis of diabetes and thereafter annually for patients with normoalbuminuria (1). The optimum timing of creatinine clearance measurement is unknown. The use of HDL-C level, along with other established risk factors, may help identify patients at high risk of development and progression of nephropathy and therefore warrant more frequent testing.

The strengths of this study are its large sample size, with an ethnically diverse population and rigorous collection of data. This allowed precise estimation of the independent effect of HDL-C on the development and progression of predefined diabetic microvascular disease outcomes. Our study also has a number of limitations. Because it is a post hoc observational analysis of data from a randomized controlled trial, the results should be interpreted as hypothesis generating. Despite the size of this study, there were very few events for some of the end points such as the need for renal

replacement therapy and renal death, which limited our ability to provide accurate estimates of the associations for these outcomes. Although the difference was small (<2.7%), adherence to the study ACE inhibitor (perindopril) was slightly lower in the lowest HDL-C third compared with the middle and highest third (77.1, 79.2, and 79.8%, respectively). We cannot exclude that this may have had an effect on renal outcomes. In addition, it is possible that not all subtypes of HDL-C have the same relationship with microvascular outcomes. However, we were unable to measure subtype and function of HDL-C in order to explore these relationships in our study.

In conclusion, in a large population of patients with type 2 diabetes and after adjustment for a wide variety of confounders, low HDL-C level was shown to be an independent risk factor for the development and progression of diabetic nephropathy. Measurement of this lipid fraction may be useful in tailoring screening and therapeutic strategies. Additional research is needed to explore the possible benefits of therapies that increase HDL-C in patients with type 2 diabetes.

Acknowledgments—The ADVANCE study was funded by grants from Servier (the major financial sponsor) and from the National Health and Medical Research Council of Australia (211086 and 358395). J.C. has received research grants from Servier, administered through the University of Sydney. J.C., A.P., M.W., and S.Z. have received honoraria from Servier for speaking at scientific meetings. No other potential conflicts of interest relevant to this article were reported.

J.M. holds a postgraduate scholarship from the National Health and Medical Research Council (NHMRC) (598402) and Heart Foundation (PC 09S 4757). A.P. holds a senior research fellowship from the NHMRC (632938). S.Z. holds a career development award from the Heart Foundation (CR 10S 5330).

J.M. researched data and wrote the manuscript. S.Z., A.A.P., J.C., M.W., D.S.C., P.G., and M.K.C.N. contributed to the discussion and reviewed and edited the manuscript. Q.L., J.W.J.B., and R.P.S. researched data and contributed to the discussion. S.Z. is the guarantor of this work and, as such, had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

This research was presented in abstract form at the European Conference of Cardiology (ESC) Congress, Paris, France, 27 August 2011. All members of the ADVANCE Collaborating Group have been listed in full previously (40).

References

- Molitch ME, DeFronzo RA, Franz MJ, Keane WF, Mogensen CE, Parving HH; American Diabetes Association. Diabetic nephropathy. *Diabetes Care* 2003;26 (Suppl. 1):S94–S98
- Fong DS, Aiello L, Gardner TW, et al.; American Diabetes Association. Retinopathy in diabetes. *Diabetes Care* 2004;27 (Suppl. 1):S84–S87
- Beckman JA, Creager MA, Libby P. Diabetes and atherosclerosis: epidemiology, pathophysiology, and management. *JAMA* 2002;287:2570–2581
- Turner RC, Millns H, Neil HA, et al. Risk factors for coronary artery disease in non-insulin dependent diabetes mellitus: United Kingdom Prospective Diabetes Study (UKPDS: 23). *BMJ* 1998;316:823–828
- Obermayr RP, Temml C, Knechtelsdorfer M, et al. Predictors of new-onset decline in kidney function in a general middle-European population. *Nephrol Dial Transplant* 2008;23:1265–1273
- Schaeffner ES, Kurth T, Curhan GC, et al. Cholesterol and the risk of renal dysfunction in apparently healthy men. *J Am Soc Nephrol* 2003;14:2084–2091
- Muntner P, Coresh J, Smith JC, Eckfeldt J, Klag MJ. Plasma lipids and risk of developing renal dysfunction: the atherosclerosis risk in communities study. *Kidney Int* 2000;58:293–301
- Smulders YM, Rakić M, Stehouwer CD, Weijers RN, Slaats EH, Silberbusch J. Determinants of progression of microalbuminuria in patients with NIDDM. A prospective study. *Diabetes Care* 1997;20:999–1005
- Ravid M, Brosh D, Ravid-Safran D, Levy Z, Rachmani R. Main risk factors for nephropathy in type 2 diabetes mellitus are plasma cholesterol levels, mean blood pressure, and hyperglycemia. *Arch Intern Med* 1998;158:998–1004
- Bruno G, Merletti F, Biggeri A, et al.; Casale Monferrato Study. Progression to overt nephropathy in type 2 diabetes: the Casale Monferrato Study. *Diabetes Care* 2003;26:2150–2155
- Zoppini G, Targher G, Chonchol M, Perrone F, Lippi G, Muggeo M. Higher HDL cholesterol levels are associated with a lower incidence of chronic kidney disease in patients with type 2 diabetes. *Nutr Metab Cardiovasc Dis* 2009;19:580–586
- Molitch ME, Rupp D, Carnethon M. Higher levels of HDL cholesterol are associated with a decreased likelihood of albuminuria in patients with long-standing type 1 diabetes. *Diabetes Care* 2006;29:78–82
- Dodson PM, Galton DJ, Winder AF. Retinal vascular abnormalities in the hyperlipidaemias. *Trans Ophthalmol Soc U K* 1981;101:17–21
- Klein R, Sharrett AR, Klein BE, et al. Are retinal arteriolar abnormalities related to atherosclerosis? The Atherosclerosis Risk in Communities Study. *Arterioscler Thromb Vasc Biol* 2000;20:1644–1650
- Leung H, Wang JJ, Rochtchina E, Wong TY, Klein R, Mitchell P. Dyslipidaemia and microvascular disease in the retina. *Eye (Lond)* 2005;19:861–868
- Klein R, Sharrett AR, Klein BE, et al.; ARIC Group. The association of atherosclerosis, vascular risk factors, and retinopathy in adults with diabetes: the atherosclerosis risk in communities study. *Ophthalmology* 2002;109:1225–1234
- Klein R, Marino EK, Kuller LH, et al. The relation of atherosclerotic cardiovascular disease to retinopathy in people with diabetes in the Cardiovascular Health Study. *Br J Ophthalmol* 2002;86:84–90
- Klein BE, Moss SE, Klein R, Surawicz TS. The Wisconsin Epidemiologic Study of Diabetic Retinopathy: XIII: relationship of serum cholesterol to retinopathy and hard exudate. *Ophthalmology* 1991;98:1261–1265
- van Leiden HA, Dekker JM, Moll AC, et al. Blood pressure, lipids, and obesity are associated with retinopathy: the Hoorn Study. *Diabetes Care* 2002;25:1320–1325
- Uçgun NI, Yildirim Z, Kiliç N, Gürsel E. The importance of serum lipids in exudative diabetic macular edema in type 2 diabetic patients. *Ann N Y Acad Sci* 2007;1100:213–217
- Keetch A, Simes RJ, Barter P, et al.; FIELD study investigators. Effects of long-term fenofibrate therapy on cardiovascular events in 9795 people with type 2 diabetes mellitus (the FIELD study): randomised controlled trial. *Lancet* 2005;366:1849–1861
- Keetch AC, Mitchell P, Summanen PA, et al.; FIELD Study Investigators. Effect of fenofibrate on the need for laser treatment for diabetic retinopathy (FIELD study): a randomised controlled trial. *Lancet* 2007;370:1687–1697
- Chew EY, Ambrosius WT, Davis MD, et al.; ACCORD Study Group; ACCORD Eye Study Group. Effects of medical therapies on retinopathy progression in type 2 diabetes. *N Engl J Med* 2010;363:233–244
- Rationale and design of the ADVANCE study: a randomised trial of blood pressure lowering and intensive glucose control in high-risk individuals with type 2 diabetes mellitus: Action in Diabetes and Vascular Disease: PreterAx and DiamicroN Modified-Release Controlled Evaluation. *J Hypertens Suppl* 2001;19:S21–S28
- Stolk RP, Vingerling JR, Cruickshank JK, et al.; AdRem Project Team and ADVANCE Management Committee. Rationale and design of the AdRem study: evaluating the effects of blood pressure lowering and intensive glucose control on vascular retinal disorders in patients with type 2 diabetes mellitus. *Contemp Clin Trials* 2007;28:6–17
- Stratton IM, Kohner EM, Aldington SJ, et al. UKPDS 50: risk factors for incidence and progression of retinopathy in type II diabetes over 6 years from diagnosis. *Diabetologia* 2001;44:156–163
- Woodward M. *Epidemiology: Study Design and Data Analysis*. 2nd ed. Boca Raton, FL, Chapman & Hall/CRC, 2005, p. 849
- Wirta OR, Pasternack AI, Mustonen JT, Koivula TA, Harmoinen A. Urinary albumin excretion rate and its determinants after 6 years in non-insulin-dependent diabetic patients. *Nephrol Dial Transplant* 1996;11:449–456
- Ravid M, Neumann L, Lishner M. Plasma lipids and the progression of nephropathy in diabetes mellitus type II: effect of ACE inhibitors. *Kidney Int* 1995;47:907–910
- Davis TM, Ting R, Best JD, et al.; Fenofibrate Intervention and Event Lowering in Diabetes Study Investigators. Effects of fenofibrate on renal function in patients with type 2 diabetes mellitus: the Fenofibrate Intervention and Event Lowering in Diabetes (FIELD) Study. *Diabetologia* 2011;54:280–290
- Cannon CP, Shah S, Dansky HM, et al.; Determining the Efficacy and Tolerability Investigators. Safety of anacetrapib in patients with or at high risk for coronary heart disease. *N Engl J Med* 2010;363:2406–2415
- van Dijk C, Berl T. Pathogenesis of diabetic nephropathy. *Rev Endocr Metab Disord* 2004;5:237–248
- Cai J, Boulton M. The pathogenesis of diabetic retinopathy: old concepts and new questions. *Eye (Lond)* 2002;16:242–260
- Jefferson JA, Shankland SJ, Pichler RH. Proteinuria in diabetic kidney disease: a mechanistic viewpoint. *Kidney Int* 2008;74:22–36
- El-Asrar AM, Al-Rubeaan KA, Al-Amro SA, Moharram OA, Kangave D. Retinopathy as a predictor of other diabetic complications. *Int Ophthalmol* 2001;24:1–11
- Tabet F, Rye KA. High-density lipoproteins, inflammation and oxidative stress. *Clin Sci (Lond)* 2009;116:87–98
- Drew BG, Duffy SJ, Formosa MF, et al. High-density lipoprotein modulates glucose metabolism in patients with type 2 diabetes mellitus. *Circulation* 2009;119:2103–2111
- Brunham LR, Kruit JK, Hayden MR, Verchere CB. Cholesterol in beta-cell dysfunction: the emerging connection between HDL cholesterol and type 2 diabetes. *Curr Diab Rep* 2010;10:55–60
- Ruilope L, Izzo J, Haller H, et al. Prevention of microalbuminuria in patients with type 2 diabetes: what do we know? *J Clin Hypertens (Greenwich)* 2010;12:422–430
- Patel A, MacMahon S, Chalmers J, et al.; ADVANCE Collaborative Group. Effects of a fixed combination of perindopril and indapamide on macrovascular and microvascular outcomes in patients with type 2 diabetes mellitus (the ADVANCE trial): a randomised controlled trial. *Lancet* 2007;370:829–840