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High-density lipoprotein cholesterol, coronary artery disease, and cardiovascular mortality

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Aims High-density lipoprotein (HDL) cholesterol is a strong predictor of cardiovascular mortality. This work aimed to investigate whether the presence of coronary artery disease (CAD) impacts on its predictive value.

Methods and results We studied 3141 participants (2191 males, 950 females) of the LUdwigshafen RIsk and Cardiovascular health (LURIC) study. They had a mean ± standard deviation age of 62.6 ± 10.6 years, body mass index of 27.5 ± 4.1 kg/m^2, and HDL cholesterol of 38.9 ± 10.8 mg/dL. The cohort consisted of 699 people without CAD, 1515 patients with stable CAD, and 927 patients with unstable CAD. The participants were prospectively followed for cardiovascular mortality over a median (inter-quartile range) period of 9.9 (8.7–10.7) years. A total of 590 participants died from cardiovascular diseases. High-density lipoprotein cholesterol by tertiles was inversely related to cardiovascular mortality in the entire cohort ($P = 0.009$). There was significant interaction between HDL cholesterol and CAD in predicting the outcome ($P = 0.007$). In stratified analyses, HDL cholesterol was strongly associated with cardiovascular mortality in people without CAD [3rd vs. 1st tertile: HR (95% CI) = 0.37 (0.18–0.74), $P = 0.005$], but not in patients with stable [3rd vs. 1st tertile: HR (95% CI) = 0.81 (0.61–1.09), $P = 0.159$] and unstable [3rd vs. 1st tertile: HR (95% CI) = 0.91 (0.59–1.41), $P = 0.675$] CAD. These results were replicated by analyses in 3413 participants of the Athero*Gene* cohort and 5738 participants of the ESTHER cohort, and by a meta-analysis comprising all three cohorts.

Conclusion The inverse relationship of HDL cholesterol with cardiovascular mortality is weakened in patients with CAD. The usefulness of considering HDL cholesterol for cardiovascular risk stratification seems limited in such patients.

Keywords High-density lipoprotein cholesterol • Atherosclerosis • Cardiovascular mortality

Introduction

Epidemiological data have provided broad evidence that low concentrations of high-density lipoprotein (HDL) cholesterol indicate increased cardiovascular risk.[1–4] Although less consistently,[5] this relationship is even apparent in patients treated with statins.[6] Therefore, raising HDL cholesterol has become a therapeutic target in coronary artery disease (CAD).[7]

Inhibition of cholesterol-ester transfer protein (CETP) is associated with a substantial increase of HDL cholesterol.[8–12]

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Nevertheless, the use of torcetrapib did not improve but rather worsened prognosis in the ILLUMINATE trial.[11] Treatment with dalcetrapib did not reduce the risk of cardiovascular events in the recently published dal-OUTCOMES trial either.[12] These disappointing results may in part be attributed to off-target effects of CETP inhibitors, most importantly an increase in systolic blood pressure and pro-inflammatory activity.[11,12] An alternative explanation would be that raising HDL cholesterol is less beneficial in certain subgroups, for example, in patients with CAD. Notably, ILLUMINATE included patients at high cardiovascular risk and all participants of the dal-OUTCOMES trial had suffered a recent acute coronary syndrome.[11,12]

Therefore, the aim of the present study was to investigate whether CAD at baseline affects the prognostic value of HDL cholesterol for cardiovascular mortality. The exploratory analyses were performed in the 'LUdwigshafen RIsk and Cardiovascular health' (LURIC) cohort.[13,14] Replication of the findings obtained from LURIC was sought in the Athero*Gene*[15,16] and the ESTHER[17,18] cohorts. Considering that their clinical presentation, treatment, and prognosis differ markedly, stable and unstable CAD were analysed separately.

Methods

Study design, participants, and clinical characterization

LURIC

A total of 3316 patients, who were referred for coronary angiography to the Ludwigshafen Heart Center in Southwest Germany, were recruited between July 1997 and January 2000.[13] Inclusion criteria were German ancestry, clinical stability except for acute coronary syndromes, and the availability of a coronary angiogram. The indications for angiography in individuals in clinically stable condition were chest pain and/or non-invasive test results consistent with myocardial ischaemia. Individuals suffering from any acute illness other than acute coronary syndromes, chronic non-cardiac diseases, or malignancy within the 5 past years, and those unable to understand the purpose of the study were excluded. Subjects with missing information on the clinical presentation of CAD, missing laboratory measurements, or missing information on the cause of death were additionally ruled out, resulting in a subgroup of 3141 participants for the present analyses. Coronary artery disease was diagnosed if coronary angiography revealed stenosis of one or more vessels $\geq$20%. Unstable angina was diagnosed according to Braunwald. Acute myocardial infarction was defined as a myocardial infarction that had occurred within the 4 weeks prior to enrolment into LURIC. A definite ST-elevation myocardial infarction was diagnosed if typical electrocardiogram changes were present along with prolonged chest pain, refractory to sublingual nitrates, and/or enzyme or troponin T elevations ($>$0.1 g/L). Non-ST-elevation myocardial infarction was diagnosed if symptoms and troponin T criteria but not the ECG criteria for ST-elevation myocardial infarction were met.[13] The functional capacity of patients with cardiac disease, especially heart failure, was estimated according to a classification developed by the New York Heart Association (NYHA).[13] Left ventricular function was estimated using echocardiography.[13]

Athero*Gene*

A total of 3800 patients, who underwent coronary angiography at the Department of Medicine II of the Johannes Gutenberg-University Mainz or the Bundeswehr-Zentralkrankenhaus Koblenz, were recruited between June 1999 and March 2000.[15,16] The exclusion criteria were evidence of haemodynamically significant valvular heart disease, surgery or trauma within the previous month, known cardiomyopathy, known cancer, febrile conditions, or use of oral anticoagulant therapy within the previous 4 weeks.[15,16] Subjects with missing information on the clinical presentation of CAD, missing laboratory measurements, or missing information on the cause of death were additionally ruled out, resulting in a subgroup of 3413 participants for the present analyses. Coronary artery disease was diagnosed if the coronary angiogram showed at least one stenosis $>$30% in a major coronary artery. Unstable angina was diagnosed according to Braunwald. Acute myocardial infarction was either ST-segment elevation with significant elevation in at least two contiguous leads or non-ST-elevation myocardial infarction based on clinic and positive in-house troponin concentrations.[16]

ESTHER

A total of 9949 subjects were recruited for the 'Epidemiologische Studie zu Chancen der Verhütung, Früherkennung und optimierten Therapie chronischer Erkrankungen in der älteren Bevölkerung (German)' by their general practitioners during a routine health check-up between 2000 and 2002 in the German federal state Saarland.[17,18] The distribution of socio-demographic baseline characteristics and common prevalent chronic diseases were similar to the distribution in the respective age categories in the German National Health Survey, which is a representative sample of the German population,[17] a fact supporting the population-based character of the study. We excluded study participants due to unmeasured HDL cholesterol, due to an unknown cause of death, and due to self-reported history of CAD that was not physician-confirmed, leaving a subgroup of 5738 participants for the present analysis. Coronary artery disease was defined by physician-reported CAD.[17,18]

Ethical approval and written informed consent

All three studies were approved by the local ethics committees and performed in accordance with the Declaration of Helsinki. All participants gave written informed consent.[13–18]

Follow-up

In the LURIC cohort, there was a follow-up for cardiovascular mortality with a mean $\pm$ standard deviation duration of 8.9 $\pm$ 3.0 years (median and inter-quartile range: 9.9, 8.7–10.7). Information on the vital status was obtained from local person registries. Using death certificates, two experienced clinicians independently classified the causes of death. They were masked to any other data of the study participants. In a few cases of a disagreement or uncertainty concerning the coding of a specific cause of death, classification was made by a principal investigator of the LURIC study (W.M.).[13] In the Athero*Gene* cohort, there was a follow-up for cardiovascular mortality with a mean $\pm$ standard deviation duration of 4.5 $\pm$ 2.0 years (median and inter-quartile range: 4.4, 2.9–6.3). Information about the causes of death and clinical events was obtained from hospital and general practitioner charts. In the ESTHER cohort, there was a follow-up for cardiovascular mortality with a mean $\pm$ standard deviation duration of 9.1 $\pm$ 1.6 years (median and inter-quartile range: 9.4, 8.9–9.9). Deaths were identified by inquiry at the residents' registration offices. All deaths coded with ICD-10 codes I00-I99 were considered to be cardiovascular deaths.

Laboratory analyses

All analyses were performed in fasting blood samples. In the LURIC and Athero*Gene* cohorts, the blood samples were collected before

angiography.[13–16] In the ESTHER cohort, the blood samples were taken during the health check-up.[17,18]

In the LURIC cohort, the lipoproteins were separated using a combined ultracentrifugation–precipitation method (β-quantification).[13] Cholesterol was measured with enzymatic reagents from WAKO (Neuss, Germany) on a WAKO 30 R or Olympus AU640 (Tokyo, Japan) analyser.[13] Triglycerides were quantified with an enzymatic colour assay on a Hitachi 717 analyser (Roche, Mannheim, Germany).[13] Apolipoproteins A1, A2, and B were measured by turbidimetry (Rolf-Greiner Biochemica, Flacht, Germany).[13] In the Athero*Gene* cohort, total cholesterol and triglycerides were measured enzymatically (Roche Diagnostics, Mannheim, Germany). High-density lipoprotein cholesterol was measured after masking apolipoprotein B immunologically (Rolf Greiner Biochemica, Flacht, Germany). LDL cholesterol was calculated using the Friedewald formula.[15,16] In the ESTHER cohort, total cholesterol, LDL cholesterol, HDL cholesterol, and triglycerides were measured with standard methods.[17,18]

Statistical analysis

Tertiles of HDL cholesterol were formed in each cohort. The baseline characteristics are presented as counts and percentages of subjects in cases of categorical data and as means and standard deviations or medians with inter-quartile ranges in cases of continuous data for the tertiles of HDL cholesterol. The χ^2 test and ANalysis Of VAriance were used to compare the distributions of the variables across the tertiles of HDL cholesterol. Triglycerides (Shapiro–Wilk *W* test) were transformed logarithmically before being used in parametric statistical procedures. The Cox proportional hazards model was used to examine the association between HDL cholesterol and time to cardiovascular death. For exploratory purposes, the associations of the HDL cholesterol tertiles with cardiovascular mortality were first studied in the entire LURIC cohort. Two pre-defined models of adjustment were used [model 1: adjusted for sex, age, and CAD; model 2: adjusted for sex, age, body mass index, systolic and diastolic blood pressure, diabetes mellitus, smoking, glomerular filtration rate, triglycerides, low-density lipoprotein cholesterol, medication use (insulin, oral antidiabetic, β-blocker, ACE-inhibitor, calcium antagonist, diuretic, acetyl salicylic acid, and statin), and CAD]. Moreover, the interaction between the HDL cholesterol tertiles and CAD with regard to cardiovascular mortality was studied by including the interaction term as a covariate. In the next step, stratified analyses in people without CAD, in patients with stable CAD, and in patients with unstable CAD were conducted using the aforementioned models of adjustment. For replication, the associations of the HDL cholesterol tertiles with cardiovascular mortality were studied in the Athero*Gene* and ESTHER cohorts, which were stratified accordingly. All statistical tests were two-sided and *P*-values <0.05 were considered significant. The SPSS 19.0 statistical package (SPSS, Inc., Chicago, IL, USA) was used in the LURIC and ESTHER cohorts. The R statistical software package (http://www.r-project.org) was used in the Athero*Gene* cohort. For meta-analyses of the results obtained from the LURIC, Athero*Gene*, and ESTHER cohorts, HRs were pooled by the inverse of the variance method in a fixed effects model with the Comprehensive Meta-Analysis® software.

Role of the funding source

The funding source was not involved in study design, in the collection, analysis, and interpretation of data, in the writing of the report, and in the decision to submit the paper for publication.

Results

LURIC

Baseline characteristics

The study participants (2191 males, 950 females) had a mean ± standard deviation age of 62.6 ± 10.6 years, body mass index of 27.5 ± 4.1 kg/m^2, and HDL cholesterol of 38.9 ± 10.8 mg/dL. High HDL cholesterol was associated with female gender, lower body mass index and waist circumference, and lower proportion of subjects with diabetes mellitus (*Table 1*). High-density lipoprotein cholesterol was positively related to total, low-density lipoprotein, and very low density lipoprotein cholesterol and inversely related to triglycerides (*Table 1*). Furthermore, HDL cholesterol was positively related to apolipoprotein A1, apolipoprotein A2, and inversely to apolipoprotein B (*Table 1*). Prevalent stable and unstable CAD, cerebral vascular disease, and peripheral vascular disease were more frequent in people with low HDL cholesterol (*Table 1*). Moreover, low HDL cholesterol went in parallel with impaired left ventricular function (*Table 1*). Use of insulin, oral antidiabetics, β-blockers, ACE-inhibitors, diuretics, statins, and acetyl salicylic acid were more frequent in subjects with low HDL cholesterol (*Table 1*). The baseline characteristics stratified for patients without CAD, patients with stable CAD, and those with unstable CAD are shown in Supplementary material online, *Table S1*. There were no major differences in the mean HDL cholesterol concentrations among the corresponding HDL cholesterol tertiles of the subgroups without CAD, with stable CAD, and with unstable CAD (Supplementary material online, *Table S1*).

Prospective analyses

A total of 925 deaths occurred during the follow-up. Among these, 590 were due to cardiovascular diseases. In the entire cohort, the HDL cholesterol tertiles were inversely related to cardiovascular mortality ($P = 0.009$) (*Table 2*). Further, investigations disclosed significant interaction between the HDL cholesterol tertiles and CAD with regard to cardiovascular mortality in model 1 ($P = 0.146$ and $P = 0.004$ for 2nd and 3rd tertile vs. 1st tertile, respectively) and in model 2 ($P = 0.178$ and $P = 0.007$ for 2nd and 3rd tertile vs. 1st tertile, respectively). Subsequent analyses were, therefore, stratified by the presence of CAD. By doing so, the tertiles of HDL cholesterol were strongly associated with cardiovascular mortality in participants without CAD ($P = 0.005$), but not in participants with stable ($P = 0.162$) or unstable ($P = 0.675$) CAD (*Table 3*). When we independently formed the HDL cholesterol tertiles within the subgroups resulting in similar numbers of participants for the three tertiles, the associations of the HDL cholesterol tertiles with cardiovascular mortality still remained significant in participants without CAD, but not in participants with stable and unstable CAD (data not shown).

Athero*Gene*

Baseline characteristics

The study participants (2568 males, 845 females) had a mean ± standard deviation age of 61.8 ± 10.0 years, body mass index of 27.5 ± 3.9 kg/m^2, and HDL cholesterol of 49.2 ± 14.6 mg/dL.

Table 1 Baseline characteristics according to tertiles of high-density lipoprotein cholesterol in the LURIC cohort

Number	1st tertile 1048	2nd tertile 1083	3rd tertile 1010	*P*-value[e]
Male sex	868 (82.8)	776 (71.7)	547 (54.2)	<0.001
Age, years	61.8 ± 11.1	62.6 ± 10.2	63.6 ± 10.4	0.001
Body mass index, kg/m^2	28.1 ± 4.1	27.8 ± 4.1	26.6 ± 3.9	<0.001
Waist circumference[a], cm	101 ± 11	100 ± 11	96 ± 12	<0.001
Hypertension	767 (73.2)	798 (73.7)	732 (72.5)	0.822
Systolic blood pressure, mmHg	139 ± 24	142 ± 23	143 ± 23	0.001
Diastolic blood pressure, mmHg	80 ± 12	82 ± 11	82 ± 11	<0.001
Diabetes mellitus	510 (48.7)	438 (40.4)	293 (29.0)	<0.001
Smoking				<0.001
Never	285 (27.2)	383 (35.4)	468 (46.3)	
Former smoker	504 (48.1)	492 (45.4)	410 (40.6)	
Current smoker	259 (24.7)	208 (19.2)	132 (13.1)	
Lipids[b], mg/dL				
Total cholesterol	182 ± 41	194 ± 37	203 ± 36	<0.001
LDL cholesterol	106 ± 35	120 ± 33	124 ± 33	<0.001
HDL cholesterol	28 ± 4	38 ± 3	51 ± 8	<0.001
VLDL cholesterol	46 ± 33	37 ± 24	28 ± 18	<0.001
Triglycerides	173 (128–241)	149 (114–201)	122 (93–161)	<0.001[c]
Apolipoprotein A1	108 ± 15	128 ± 13	155 ± 19	<0.001
Apolipoprotein A2	36 ± 8	42 ± 8	48 ± 9	<0.001
Apolipotrotein B	106 ± 26	106 ± 24	101 ± 23	<0.001
GFR, mL/min/1.73 m^2	80 ± 20	83 ± 19	81 ± 18	0.022
CAD				<0.001
No CAD	142 (13.5)	233 (21.5)	324 (32.1)	
Stable CAD	509 (48.6)	535 (49.4)	471 (46.6)	
Unstable CAD	397 (37.9)	315 (29.1)	215 (21.3)	
Presentation of unstable CAD				<0.001
Unstable angina	223 (21.3)	217 (20.0)	181 (17.9)	
NSTEMI	61 (5.8)	34 (3.1)	19 (1.9)	
STEMI	113 (10.8)	64 (5.9)	15 (1.5)	
NYHA functional class				0.339
1	527 (50.3)	569 (52.5)	509 (50.4)	
2	302 (28.8)	319 (29.5)	314 (31.1)	
3	189 (18.0)	157 (14.5)	157 (15.5)	
4	30 (2.9)	38 (3.5)	30 (3.0)	
Left ventricular function (echo)[d]				<0.001
Normal	542 (53.1)	665 (62.9)	723 (72.7)	
Mildly impaired	163 (16.0)	155 (14.7)	116 (11.7)	
Moderately impaired	136 (13.3)	123 (11.6)	76 (7.6)	
Severely impaired	77 (7.5)	36 (3.4)	28 (2.8)	
Peripheral vascular disease	147 (14.0)	86 (7.9)	66 (6.5)	<0.001
Cerebrovascular disease	123 (11.7)	87 (8.0)	76 (7.5)	0.001
Medication use				
Insulin	70 (6.7)	59 (5.4)	38 (3.8)	0.013
Oral antidiabetic	125 (11.9)	84 (7.8)	46 (4.6)	<0.001
β-Blocker	730 (69.7)	697 (64.4)	552 (54.7)	<0.001

Continued

Table 1 *Continued*

Number	1st tertile 1048	2nd tertile 1083	3rd tertile 1010	*P*-value[e]
ACE-inhibitor	637 (60.8)	565 (52.2)	455 (45.0)	<0.001
Calcium antagonist	156 (14.9)	169 (15.6)	174 (17.2)	0.331
Diuretic	366 (34.9)	286 (26.4)	244 (24.2)	<0.001
Statin	576 (54.9)	530 (48.9)	408 (40.4)	<0.001
Acetyl salicylic acid	790 (75.4)	773 (71.4)	672 (66.5)	<0.001

Values are means ± standard deviations or medians (25th–75th percentiles) in cases of continuous variables and numbers (percentages) in case of categorical data.
[a]Numbers: 1035/1073/994.
[b]To convert values for total, LDL, HDL, and VLDL cholesterol to millimoles per litre, multiply by 0.02586; to convert values for triglycerides to millimoles per litre, multiply by 0.01129.
[c]ANalysis Of VAriance of logarithmically transformed values.
[d]Numbers: 918/979/943.
[e]For differences across the three groups calculated with χ^2 test and ANalysis Of VAriance for categorical and continuous data, respectively.

Table 2 Cardiovascular mortality according to tertile of high-density lipoprotein cholesterol in the LURIC cohort

	n	CD	HR	*P*-value
Model 1[a]				
1st tertile	1048	248 (23.7)	1.0 reference	
2nd tertile	1083	190 (17.5)	0.71 (0.59–0.86)	<0.001
3rd tertile	1010	152 (15.0)	0.59(0.36–0.80)	<0.001
Model 2[b]				
1st tertile	1048	248 (23.7)	1.0 reference	
2nd tertile	1083	190 (17.5)	0.82 (0.67–1.00)	0.052
3rd tertile	1010	152 (15.0)	0.74 (0.59–0.93)	0.009

n, number; CD, number of cardiovascular deaths (percentage of tertile); HR, hazard ratio (calculated with Cox proportional hazards model).
[a]Adjusted for sex, age, and CAD.
[b]Adjusted for sex, age, body mass index, systolic and diastolic blood pressure, diabetes, smoking, glomerular filtration rate, triglycerides, LDL cholesterol, medication use (insulin, oral antidiabetic, β-blocker, ACE-inhibitor, calcium antagonist, diuretic, acetyl salicylic acid, and statin), and CAD.

The baseline characteristics of the Athero*Gene* cohort stratified for patients without CAD, patients with stable CAD, and those with unstable CAD are shown in Supplementary material online, *Table S2*.

Prospective analyses

A total of 381 deaths occurred during the follow-up. Among these, 264 were due to cardiovascular diseases. Low HDL cholesterol was strongly associated with increased cardiovascular mortality in people without CAD (*Table 4*). The association between HDL cholesterol and cardiovascular mortality was less pronounced in patients with stable CAD and turned non-significant after multivariate adjustment (*Table 4*). In patients with unstable CAD, there was no significant association between the tertiles of HDL cholesterol and cardiovascular mortality (*Table 4*).

ESTHER

Baseline characteristics

The study participants (2606 males, 3132 females) had a mean ± standard deviation age of 62.0 ± 6.6 years, body mass index of 27.7 ± 4.3 kg/m^2, and HDL cholesterol of 53.5 ± 15.3 mg/dL. The baseline characteristics of the ESTHER cohort stratified for people without CAD and patients with stable CAD are shown in Supplementary material online, *Table S3*.

Prospective analyses

A total of 586 deaths occurred during the follow-up. Among these, 196 deaths were due to cardiovascular diseases. High-density lipoprotein cholesterol was inversely related to cardiovascular mortality in people without CAD (*Table 4*). This association reached statistical significance comparing the 2nd with the 1st tertile of HDL cholesterol (*Table 4*). In patients with stable CAD, there was no significant association between the tertiles of HDL cholesterol and cardiovascular mortality (*Table 4*).

Meta-analysis

The meta-analysis comprised 12 292 participants of the LURIC, Athero*Gene*, and ESTHER cohorts. In the subgroup of 5791 people without CAD, the HDL cholesterol tertiles were significantly, inversely related to cardiovascular mortality (*Table 5*). In contrast, the HDL cholesterol tertiles were not significantly related to cardiovascular mortality in the 4304 patients with stable CAD and the 2197 patients with unstable CAD (*Table 5*).

Discussion

The present data from a total of 12 292 participants of the LURIC, the Athero*Gene*, and the ESTHER cohorts show that CAD modulates the association of HDL cholesterol with cardiovascular mortality.

High-density lipoprotein cholesterol tertiles were significantly inversely related to cardiovascular mortality in the entire LURIC cohort. Inclusion of the interaction term between CAD and HDL cholesterol indicated variation in the predictive value of HDL cholesterol for cardiovascular mortality according to the presence of CAD. In stratified analyses, the association between the HDL cholesterol

Table 3 Cardiovascular mortality according to tertile of high-density lipoprotein cholesterol stratified for coronary artery disease in the LURIC cohort

	No CAD				Stable CAD				Unstable CAD			
	n	CD	HR	*P*-value	*n*	CD	HR	*P*-value	*n*	CD	HR	*P*-value
Model 1[a]												
1st tertile	142	22 (15.5)	1.0 reference		509	138 (27.1)	1.0 reference		397	88 (22.2)	1.0 reference	
2nd tertile	233	20 (8.6)	0.44 (0.24–0.81)	0.008	535	114 (21.3)	0.74 (0.58–0.95)	0.017	315	56 (17.8)	0.75 (0.53–1.04)	0.085
3rd tertile	324	19 (5.9)	0.27 (0.14–0.50)	<0.001	471	94 (20.0)	0.67 (0.51–0.87)	0.003	215	39 (18.1)	0.65 (0.44–0.96)	0.030
Model 2[b]												
1st tertile	142	22 (15.5)	1.0 reference		505	138 (27.1)	1.0 reference		397	88 (22.2)	1.0 reference	
2nd tertile	233	20 (8.6)	0.54 (0.28–1.06)	0.074	535	114 (21.3)	0.83 (0.64–1.08)	0.162	315	56 (17.8)	0.88 (0.62–1.26)	0.485
3rd tertile	324	19 (5.9)	0.37 (0.18–0.74)	0.005	471	94 (20.0)	0.81 (0.61–1.09)	0.159	215	39 (18.1)	0.91 (0.59–1.41)	0.675

n, number; CD, number of cardiovascular deaths (percentage of tertile); HR, hazard ratio (calculated with Cox proportional hazards model).
[a]Adjusted for sex and age.
[b]Adjusted for sex, age, body mass index, systolic and diastolic blood pressure, diabetes, smoking, glomerular filtration rate, triglycerides, LDL cholesterol, and medication use (insulin, oral antidiabetic, β-blocker, ACE-inhibitor, calcium antagonist, diuretic, acetyl salicylic acid, and statin).

tertiles and cardiovascular mortality was strong in people without CAD, whereas it was weak and non-significant after multivariate adjustment in patients with stable and unstable CAD. Very similar observations were made in the *AtheroGene* cohort. The aforementioned differences were less pronounced in the ESTHER cohort, possibly due to the lack of coronary angiograms and consequently a large proportion of undiagnosed, silent CAD. Finally, a meta-analysis comprising all three cohorts was performed and the results were in support of the LURIC findings.

Previous studies have not specifically addressed an interaction of HDL with CAD, probably because very few studies have collected precise information on both, CAD at baseline and cardiovascular death. However, our results are in agreement with evidence from the dal-OUTCOMES trial.[12] In this cohort of 15 871 patients, who had suffered acute coronary syndromes, HDL cholesterol was not predictive of the primary endpoint.[12] Moreover, the present observations are confirmed by recent data on the cholesterol efflux capacity from macrophages to serum, which is positively related to HDL cholesterol.[19,20] In cross-sectional analyses, low cholesterol efflux capacity was repeatedly associated with higher prevalence of CAD.[19,20] At the same time, the cholesterol efflux capacity was positively associated with the risk of future vascular complications in patients with CAD, whereas no such paradox association was seen in an outpatient cohort.[20]

Two potential reasons that may explain the interaction between the HDL cholesterol tertiles and CAD with regard to cardiovascular mortality shall be highlighted: first, dysfunctional HDL may account for weaker associations of HDL cholesterol with cardiovascular mortality in CAD. High-density lipoprotein is considered to represent the major vehicle of reverse cholesterol transport.[21–23] In addition, HDL may exert anti-inflammatory effects, prevent low-density lipoprotein oxidation, and play an important role in nitric oxide synthesis.[21–23] Even anti-thrombotic potency has been suggested.[21–23] Nevertheless, there is growing knowledge that the vascular protective properties of HDL are impaired in certain diseases.[24,25] Most importantly, the anti-inflammatory and anti-oxidative effects of HDL were demonstrated to be reduced in CAD.[26,27] In the LURIC cohort, the apolipoprotein A1 and A2 composition of HDL did not explain the key finding (Supplementary material online). Future studies within the LURIC cohort will address other aspects of HDL functionality. Second, multimodal treatment of cardiovascular risk factors and co-morbidity may have blunted the relationships of HDL cholesterol with cardiovascular mortality in patients with CAD. However, exploratory analyses within the LURIC cohort did not support this possibility (Supplementary material online).

It is a limitation of our study that HDL cholesterol was measured once at baseline only. Therefore, we were not able to adjust for possible moderate fluctuations of HDL cholesterol during the follow-up, for example due to the start of statin treatment or an increase of the statin dose.[28] The major strength of this work is the detailed clinical and metabolic investigation of the LURIC participants, including coronary angiography. Moreover, we want to emphasize the long duration of the follow-up with a large number of fatal cardiovascular events. In addition, our conclusions rely on replication in 3413 participants of the Athero*Gene* cohort and 5738 participants of the ESTHER cohort, and on a meta-analysis comprising all three cohorts.

Table 4 Cardiovascular mortality according to tertiles of high-density lipoprotein cholesterol stratified for coronary artery disease in the ESTHER and Athero*Gene* cohorts

	No CAD				Stable CAD				Unstable CAD			
	n	CD	HR	*P*-value	*n*	CD	HR	*P*-value	*n*	CD	HR	*P*-value
Athero*Gene*												
Model 1[a]												
1st tertile	35	6 (17.1)	1.0 reference		674	65 (9.6)	1.0 reference		509	46 (9.0)	1.0 reference	
2nd tertile	45	2 (4.4)	0.29 (0.06–1.44)	0.128	643	39 (6.1)	0.65 (0.43–0.96)	0.031	421	36 (8.6)	0.95 (0.62–1.48)	0.828
3rd tertile	73	2 (2.7)	0.16 (0.03–0.82)	0.027	673	42 (6.2)	0.57 (0.38–0.86)	0.007	340	26 (7.6)	0.68 (0.42–1.12)	0.127
Model 2[b]												
1st tertile	n.a.[c]	n.a.[c]	n.a.[c]	n.a.[c]	663	65 (9.8)	1.0 reference		503	45 (8.9)	1.0 reference	
2nd tertile					633	38 (6.0)	0.88 (0.58–1.33)	0.539	416	36 (8.7)	1.06 (0.67–1.69)	0.792
3rd tertile					659	41 (6.2)	0.73 (0.47–1.13)	0.159	337	26 (7.7)	0.90 (0.53–1.52)	0.685
ESTHER												
Model 1[a]												
1st tertile	1546	47 (3.0)	1.0 reference		362	37 (10.2)	1.0 reference		n.a.[d]	n.a.[d]	n.a.[d]	n.a.[d]
2nd tertile	1665	29 (1.7)	0.58 (0.36–0.92)	0.020	255	29 (11.4)	1.18 (0.72–1.93)	0.502				
3rd tertile	1728	35 (2.0)	0.74 (0.46–1.17)	0.199	182	19 (10.4)	1.14 (0.64–2.02)	0.659				
Model 2[b]												
1st tertile	1184	37 (3.1)	1.0 reference		282	26 (9.2)	1.0 reference					
2nd tertile	1324	23 (1.7)	0.58 (0.34–1.00)	0.050	211	24 (11.4)	1.80 (0.98–3.33)	0.059				
3rd tertile	1410	29 (2.1)	0.80 (0.50–1.27)	0.226	151	14 (9.3)	1.70 (0.80–3.61)	0.168				

n, number; CD, number of cardiovascular deaths (percentage of tertile); HR, hazard ratio.
[a]Adjusted for sex and age.
[b]Adjusted for sex, age, body mass index, systolic and diastolic blood pressure (hypertension in the Athero*Gene* cohort), diabetes, smoking, glomerular filtration rate, triglycerides, LDL cholesterol, and medication use (insulin, oral antidiabetic, β-blocker, ACE-inhibitor, calcium antagonist, diuretic, acetyl salicylic acid, and statin).
[c]Sample size too low.
[d]No patients with unstable CAD in this cohort.

Table 5 Results of meta-analyses for cardiovascular mortality according to tertiles of high-density lipoprotein cholesterol stratified for coronary artery disease in the LURIC, ESTHER, and AtheroGene cohorts

	No CAD			Stable CAD			Unstable CAD		
	HR	*P*-value	Heterogeneity (Q; P; I^2)	HR	*P*-value	Heterogeneity (Q; P; I^2)	HR	*P*-value	Heterogeneity (Q; P; I^2)
Model 1[a]									
1st tertile	1.0 reference			1.0 reference			1.0 reference		
2nd tertile	0.51 (0.35–0.73)	<0.001	1.0; 0.61; 0%	0.77 (0.64–0.94)	0.009	3.7; 0.16; 45%	0.82 (0.63–1.07)	0.143	0.7; 0.40; 0%
3rd tertile	0.49 (0.34–0.71)	<0.001	8.1; 0.02; 75%	0.67 (0.56–0.85)	<0.001	3.8; 0.14; 48%	0.66 (0.49–0.90)	0.008	0.2; 0.88; 0%
Model 2[b,c]									
1st tertile	1.0 reference			1.0 reference			1.0 reference		
2nd tertile	0.56 (0.37–0.86)	0.007	0.0; 0.87; 0%	0.92 (0.75–1.13)	0.440	5.3; 0.07; 62%	0.94 (0.71–1.25)	0.681	0.4; 0.53; 0%
3rd tertile	0.63 (0.43–0.94)	0.021	3.2; 0.07; 68%	0.84 (0.67–1.06)	0.148	3.8; 0.15; 48%	0.91 (0.65–1.27)	0.564	0.0; 0.98; 0%

HR, hazard ratio.
[a]Adjusted for sex and age.
[b]Adjusted for sex, age, body mass index, systolic and diastolic blood pressure, diabetes, smoking, glomerular filtration rate, triglycerides, LDL cholesterol, and medication use (insulin, oral antidiabetic, β-blocker, ACE-inhibitor, calcium antagonist, diuretic, acetyl salicylic acid, and statin.
[c]For participants without CAD including data from LURIC and AtheroGene cohorts only.

To sum up, the association of HDL cholesterol with cardiovascular mortality is weakened in the presence of CAD. The usefulness of considering HDL cholesterol for cardiovascular risk stratification may be limited in secondary prevention.

Supplementary material

Supplementary material is available at *European Heart Journal* online.

Authors' contributions

B.O.B. and W.M. designed the LURIC study. G.S., W.M., and M.E.K. performed the statistical analysis in the LURIC cohort. H.S. performed lipid analysis in the LURIC cohort. G.S. wrote the manuscript. S.B. designed the Athero*Gene* study. S.A. performed the statistical analysis in the Athero*Gene* study. H.B. designed the ESTHER study. B.H. was responsible for mortality data collection in the ESTHER study. B.S. and U.M. performed the statistical analysis in the ESTHER study. B.S. performed the meta-analysis. T.B.G., A.R., H.S., U.L., H.B., B.S., R.B.S., S.B., G.G., and A.N. contributed to the interpretation of the results and reviewed/edited the manuscript. All authors have read, approved, and take full responsibility for the manuscript as submitted.

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