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CLINICAL RESEARCH
Lipids

U-shaped relationship of HDL and risk of infectious disease: two prospective population-based cohort studies

Christian M. Madsen[1,2,3], Anette Varbo[1,2,3], Anne Tybjærg-Hansen[2,3,4,5], Ruth Frikke-Schmidt[2,3,5], and Børge G. Nordestgaard[1,2,3,4]*

[1]Department of Clinical Biochemistry, Herlev and Gentofte Hospital, Copenhagen University Hospital, Herlev Ringvej 75, 2730 Herlev, Denmark; [2]The Copenhagen General Population Study, Herlev and Gentofte Hospital, Copenhagen University Hospital, Herlev Ringvej 75, 2730 Herlev, Denmark; [3]Faculty of Health and Medical Sciences, University of Copenhagen, Blegdamsvej 3B, 2200 Copenhagen, Denmark; [4]The Copenhagen City Heart Study, Frederiksberg Hospital, Copenhagen University Hospital, Nordre Fasanvej 57, 2000 Frederiksberg, Denmark; and [5]Department of Clinical Biochemistry, Rigshospitalet, Copenhagen University Hospital, Blegdamsvej 9, 2100 Copenhagen, Denmark

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Aims Preclinical evidence has indicated that HDL may play an important role in the immune system; however, very little is known about the role of HDL in the immune system in humans. We tested the hypothesis that low and high concentrations of HDL cholesterol are associated with risk of infectious disease in the general population.

Methods and results We included 97 166 individuals from the Copenhagen General Population Study and 9387 from the Copenhagen City Heart Study with measurements of HDL cholesterol at baseline. The primary endpoint was any infectious disease requiring hospital admission, ascertained in the Danish health registries from baseline in 2003–13 or 1991–94 through 2014; 9% and 31% of individuals in the two studies experienced one or more infectious disease events. Using restricted cubic splines, there was a U-shaped association between concentrations of HDL cholesterol and risk of any infection. Following multifactorial adjustment, individuals with HDL cholesterol below 0.8 mmol/L (31 mg/dL) and above 2.6 mmol/L (100 mg/dL) had hazard ratios for any infection of 1.75 (95% confidence interval 1.31–2.34) and 1.43 (1.16–1.76), compared to those with HDL cholesterol of 2.2–2.3 mmol/L (85–95 mg/dL). In the Copenhagen City Heart Study, corresponding hazard ratios for any infection were 2.00 (1.16–3.43) and 1.13 (0.80–1.60).

Conclusion Low and high HDL cholesterol concentrations found in 21% and 8% of individuals were associated with higher risk of infectious disease in the general population. These findings do not necessarily indicate causality.

Keywords Lipids • Lipoproteins • HDL • Infectious disease • General population • Epidemiology

Introduction

In recent years, a protective role of HDL in cardiovascular disease has been challenged. First, genetically low concentrations of HDL cholesterol were not causally associated with high risk of cardiovascular disease.[1,2] Second, randomized clinical trials with HDL cholesterol-elevating drugs were unable to reduce vascular disease risk.[3] However, HDL is conserved and present in many species, indicating an important yet unknown biological role from an evolutionary standpoint, although this has not been clearly determined.

HDL may have a role in immunity, as HDL cholesterol concentrations decrease rapidly at the onset of sepsis and as low HDL is associated with poor outcome in these patients.[4,5] A possible explanation for such phenomena could be the ability of HDL to neutralize and clear pro-inflammatory endotoxins.[6,7] HDL may also be involved in protection against certain parasitic infections, as apolipoprotein L-I found in HDL particles confers human resistance towards infection with *Trypanosoma brucei brucei* by lysing the parasite.[8] Mechanistically, preclinical studies suggest that HDL can regulate innate and adaptive immunity,[9] as HDL takes part in the regulation of the proliferation of

* Corresponding author. Tel: +45 38683297, Fax: +45 38683311, Email: boerge.nordestgaard@regionh.dk

haematopoietic stem cells from the bone marrow and is also able to modulate the maturation and function of immune cells through an effect on cell surface receptors.[10–12]

Despite the evidence indicating that HDL is an important modulator of the immune response, little is known about the relationship of HDL with the immune system in humans, and specifically whether HDL cholesterol concentrations associate with infectious disease risk. Indeed, the association between HDL concentrations and risk of infectious disease has only been investigated in five studies with 204 to 1719 hospitalized patients showing that low HDL cholesterol at admission was associated with high risk of developing infectious complications in relation to hospitalization[13–17]. However, we recently found that both low and high HDL cholesterol is associated with high all-cause mortality.[18]

We tested the hypothesis that low and high concentrations of HDL cholesterol are associated with risk of infectious disease in the general population.

Methods

A detailed description of the cohorts, endpoints, laboratory analyses, covariates, and statistical analyses is given in the Supplementary material online, *Methods*.

The Copenhagen General Population Study

The primary study cohort was comprised of 97 166 individuals from the prospective Copenhagen General Population Study with baseline measurements of HDL cholesterol. It was initiated in 2003 and recruitment is ongoing.

The Copenhagen City Heart Study

A total of 9387 individuals from the 1991 to 1994 examination of the Copenhagen City Heart Study with baseline measurements of HDL cholesterol were included as an independent cohort for confirmation of the results obtained in the Copenhagen General Population Study.

Endpoints

Infectious disease endpoints were based on data from the national Danish Patient Registry, which records all hospital contacts in Denmark. We only included infectious events listed as the primary cause of admission until the end of follow-up on 8 November 2014. Infectious diseases were grouped based on the International Classification of Diseases codes as shown in Supplementary material online, *Table S1*. For the endpoint infectious disease-related death, data were retrieved from the national Danish Register of Causes of Death.

Laboratory analyses, genotyping, and covariates

HDL cholesterol, LDL cholesterol, triglycerides, and apolipoprotein A1 were all measured non fasting using standard hospital assays.[19] Genotyping was done using TaqMan-based assays. Covariates for statistical adjustment were chosen a priori based on known association with HDL cholesterol and risk of infectious disease. Multifactorial adjustment was for age, gender, body mass index, smoking, alcohol intake, physical activity in leisure time, diabetes mellitus, ischaemic heart disease, lipid-lowering therapy, estimated glomerular filtration rate (eGFR), LDL cholesterol, and triglycerides.

Statistical analyses

Statistical analyses were performed using Stata 13.1, and *P*-values were two sided. Only for adjustment in the few individuals with missing information on certain covariates (see Supplementary material online, *Tables S2 and S3*), we used multiple imputations with chained equations to fill out the missing values.

Associations between risk of infectious disease and HDL cholesterol, apolipoprotein A1, triglycerides, and LDL cholesterol on a continuous scale were examined using restricted cubic splines, with knots at equally spaced percentiles, which were used in multiple-event Cox proportional hazards regression. These analyses were used to help define meaningful reference groups using categories of HDL cholesterol, with the category having the lowest risk as the reference group. Individuals were also stratified into four clinically relevant HDL cholesterol groups with 0.5 mmol/L (19 mg/dL) intervals and into 11 groups with 0.2 mmol/L (8 mg/dL) intervals.

To allow for repeated infectious disease events for each individual, multiple-event Cox proportional hazards regression using the approach described by Andersen–Gill with robust standard errors was used as the primary analysis. Age was used as timescale, and analyses were conducted with delayed entry (left truncation) and censoring at death or emigration (0.4%). Conventional single-event Cox proportional hazards regressions were carried out as sensitivity analyses and were used for analyses of infectious disease-related death. Infectious disease events were also analysed prospectively using negative binomial regression, Fine and Gray competing risk regression, and logistic regression.

For genetic analyses, genotypes were combined into an unweighted allele score of HDL cholesterol increasing alleles. As genotypes are present from birth, we included infectious disease events from birth or 1977 (registry start), whichever came last. Multiple-event Cox proportional hazards regression adjusted for age and gender was used for the genetic analyses.

Risk estimates and confidence intervals (CIs) were corrected for regression dilution bias using a non-parametric method. *P*-values are for linear trend, and we tested for interactions by incorporating two-factor interaction terms between HDL cholesterol and included covariates in categories, or as continuous variables, as indicated. *P*-values for interaction were obtained using the Wald test.

Results

Individuals included from the Copenhagen General Population Study were followed for a median of 6 years (range 0–11 years), and 8282 (9%) individuals experienced one or more infectious disease events during follow-up. Correspondingly, individuals from the Copenhagen City Heart Study were followed up for 20 years (0–23 years), and 2904 (31%) developed an infectious disease event. Baseline characteristics for both cohorts are shown in Supplementary material online, *Tables S4 to S12*.

HDL cholesterol at baseline was associated with parameters of the immune system as plasma C-reactive protein (CRP) decreased 29% (95% CI 27–30%; $R^2 = 0.02$) and blood leucocytes decreased 0.51×10^9/L ($0.49–0.53 \times 10^9$/L; $R^2 = 0.02$) per 1 mmol/L (39 mg/dL) higher HDL cholesterol (see Supplementary material online, *Figure S1*).

HDL cholesterol, apolipoprotein A1, triglycerides, and LDL cholesterol on a continuous scale and risk of infectious disease

Using restricted cubic splines, both low and high concentrations (U-shaped) of HDL cholesterol were associated with high risk of any

infection (*Figure 1*). The same was observed for concentrations of apolipoprotein A1, whereas no clear association was seen for concentrations of triglycerides and risk of any infection. Low concentrations of LDL cholesterol were also associated with high risk of any infection.

HDL cholesterol and risk of infectious disease

When HDL cholesterol concentrations were divided into 0.2 mmol/L (8 mg/dL) intervals, the risk of any infectious disease showed a U-shaped pattern with both low concentrations and very high concentrations being associated with high risk, compared with the reference group with HDL cholesterol concentrations of 2.2–2.3 mmol/L (85–93 mg/dL) (*Figure 2*, lower part). Individuals with HDL cholesterol below 0.8 mmol/L (31 mg/dL) had a multifactorially adjusted hazard ratio for any infection of 1.75 (95% confidence interval 1.31–2.34) as compared to those with HDL cholesterol of 2.2–2.3 mmol/L (85–93 mg/dL). Individuals with HDL cholesterol ≥2.6 mmol/L (100 mg/dL) also showed high risk of infection with a corresponding hazard ratio of 1.43 (1.16–1.76).

In age- and gender-adjusted models, there was an inverse association between concentrations of HDL cholesterol and risk of any infectious disease when HDL cholesterol concentrations were divided into 0.5 mmol/L (19 mg/dL) intervals (P for trend < 0.0001; *Figure 2*, upper part). This association was attenuated, but persisted, following multifactorial adjustment with a hazard ratio for any infection of 1.24 (1.05–1.46) for individuals with HDL cholesterol below 1.0 mmol/L (39 mg/dL) compared to individuals with HDL cholesterol above 2.0 mmol/L (77 mg/dL).

The population-attributable risks of infectious disease in individuals with low (<1.2 mmol/L (<46 mg/dL)) and high (≥2.4 mmol/L (≥93 mg/dL)) HDL cholesterol were 3.2% and 1.9%, respectively.

HDL cholesterol and risk of infectious disease subgroups

For infectious disease subgroups, except the group other infections, there were inverse associations between HDL cholesterol concentrations and risk of infectious disease in age- and gender-adjusted models (*Figure 3*). This association persisted after multifactorial adjustment for gastroenteritis and bacterial pneumonia with hazard ratios of 1.68 (95% confidence interval 1.10–2.56) and 1.34 (1.02–1.76) for individuals with HDL cholesterol below 1.0 mmol/L (39 mg/dL) compared to individuals with HDL cholesterol above 2.0 mmol/L (77 mg/dL). In the remaining subgroups of infectious disease, the association with HDL cholesterol did not persist after multifactorial adjustment. The relationship between HDL cholesterol and infectious disease subgroups was also examined using 0.2 mmol/L (8 mg/dL) categories of HDL cholesterol, although with less statistical power (see Supplementary material online, *Figure S2*). The high risk of any infectious disease observed with high concentrations of HDL cholesterol seemed to be driven by bacterial pneumonia, skin infection, and urinary tract infection.

Grouping the infectious disease subgroups by likely aetiology into bacterial and viral showed that both low and high concentrations of HDL cholesterol were associated with high risk of bacterial infectious disease, whereas only low concentrations of HDL cholesterol were

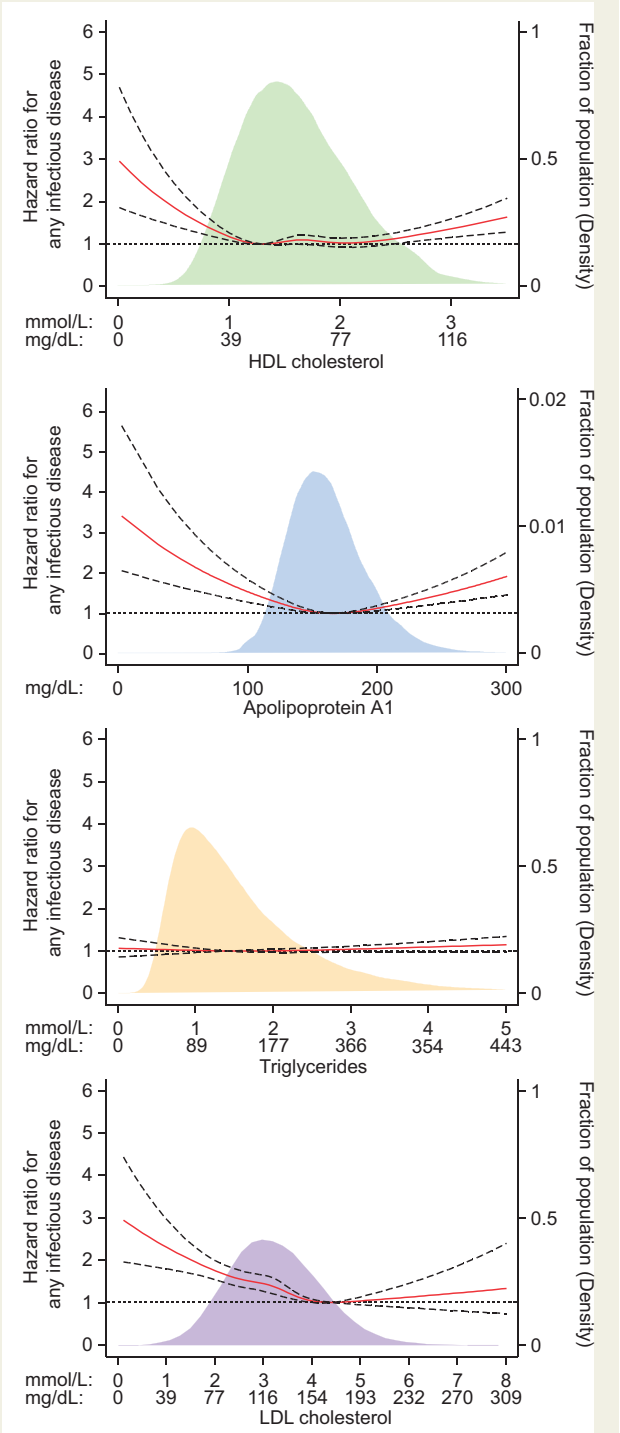


Figure 1 HDL cholesterol, apolipoprotein A1, triglycerides, and LDL cholesterol on a continuous scale and risk of any infectious disease in 97 166 individuals from the Copenhagen General Population Study. Analyses were conducted using restricted cubic splines, with hazard ratios and 95% confidence intervals from multiple-event Cox proportional hazards regression. The values of HDL cholesterol, apolipoprotein A1, triglycerides, and LDL cholesterol with the lowest hazard ratio were chosen as reference. The light green, blue, yellow, and purple areas indicate the distribution of concentrations of HDL cholesterol, apolipoprotein A1, triglycerides, and LDL cholesterol, respectively.

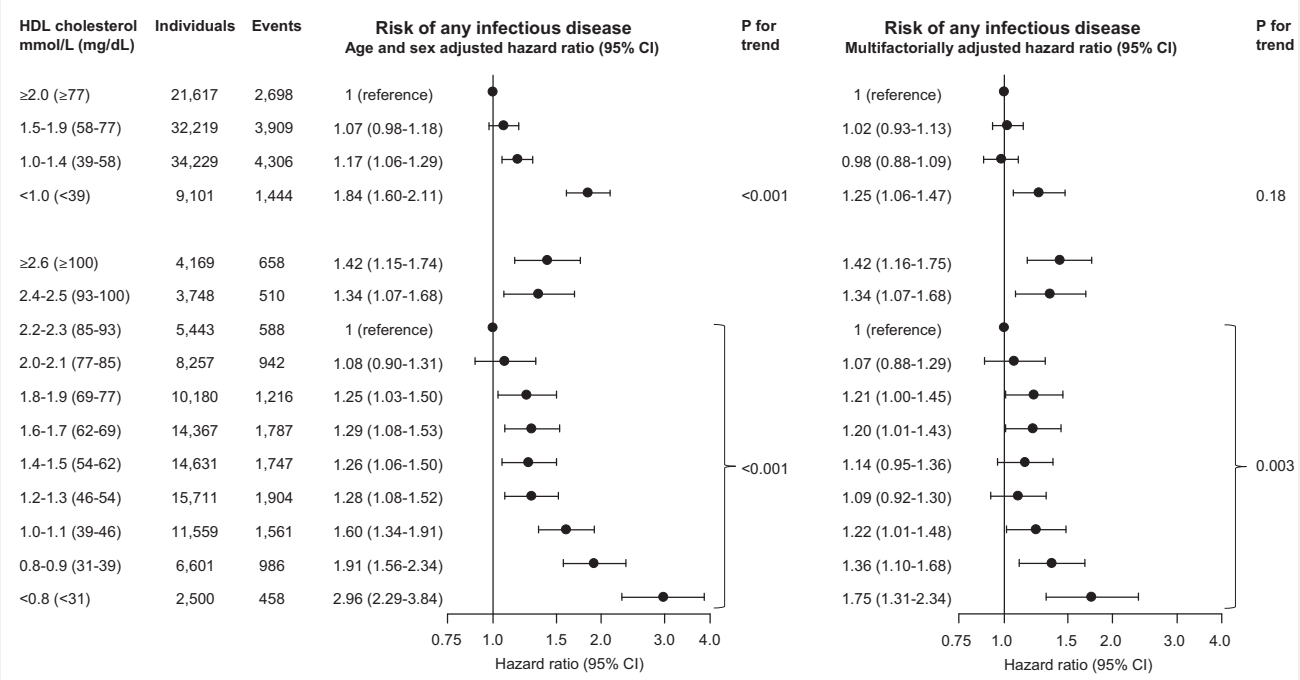


Figure 2 HDL cholesterol and risk of any infectious disease in 97 166 individuals from the Copenhagen General Population Study. Individuals are in the top part divided into 0.5 mmol/L (19mg/dL) intervals, and in the bottom part into 0.2 mmol/L (8 mg/dL) intervals of HDL cholesterol. Hazard ratios were from multiple-event Cox proportional hazards regression.

associated with high risk of viral infectious disease, albeit the number of events of viral disease were limited (see Supplementary material online, *Figure S3*).

Sensitivity analyses

The association between low HDL cholesterol and high risk of any infectious disease was robust after stratification for age, gender, body mass index, smoking, alcohol intake, physical activity, statin use, diabetes, ischaemic heart disease, triglycerides, LDL cholesterol, eGFR, and birth year (*Figure 4*). Specifically, results were similar when stratifying based on alcohol intake, plasma CRP, and blood leucocytes (*Figure 5*). When further adjusting for plasma CRP or blood leucocytes, the association was slightly attenuated for plasma CRP adjustment but persisted for the groups with extreme high and low concentrations of HDL cholesterol (see Supplementary material online, *Figure S4*). As women on average have higher HDL cholesterol than men, we also examined restricted cubic spline models in men and women separately (see Supplementary material online, *Figure S5*), with a similar overall pattern in both gender.

Results from individuals without missing information on covariates, i.e. a complete case analysis, were similar to those with imputed missing covariates (compare Supplementary material online, *Figure S6* with *Figure 2*). Furthermore, when examining the association between HDL cholesterol and risk of infectious disease using (i) single-event Cox proportional hazard regression (see Supplementary material online, *Figure S7*), (ii) competing risk regression with death as competing event (see Supplementary material online, *Figure S8*), (iii) negative binomial regression (see Supplementary material online, *Figure S9*), and (iv) cross-sectional logistic regression (see Supplementary material online, *Figure S10*), results were similar to those obtained from multiple-event Cox proportional hazard regression shown in *Figure 2*, with similar risk estimates.

Finally, examining at infectious disease-related death, the association with HDL cholesterol was similar to what was observed for any infectious disease, as both low and high concentrations of HDL cholesterol were associated with high risk of infectious disease-related death (*Figure 6*).

HDL cholesterol and risk of infectious disease: independent confirmation

Associations between concentration of HDL cholesterol and risk of infectious disease were also investigated in an independent cohort, the Copenhagen City Heart Study. Using 0.5 mmol/L (19 mg/dL) cut-points of HDL cholesterol, a similar association was observed as in the Copenhagen General Population Study for low concentrations of HDL cholesterol (compare *Figure 7* with *Figure 2*). Individuals with HDL cholesterol below 1.0 mmol/L (39 mg/dL) had a multifactorially adjusted hazard ratio for any infection of 1.54 (95% CI 1.16–2.04) compared to individuals with HDL cholesterol above 2.0 mmol/L (77 mg/dL). Also, a similar trend as in the Copenhagen General Population Study was observed in the Copenhagen City Heart Study when examining 0.2 mmol/L (8 mg/dL) intervals of HDL cholesterol, as individuals with HDL cholesterol below 0.8 mmol/L (31 mg/dL) had a multifactorially adjusted hazard ratio of 2.00 (95% CI 1.16–3.43) compared to individuals with HDL cholesterol of 2.2–2.3 mmol/L (95% CI 85–93 mg/dL); individuals with very high

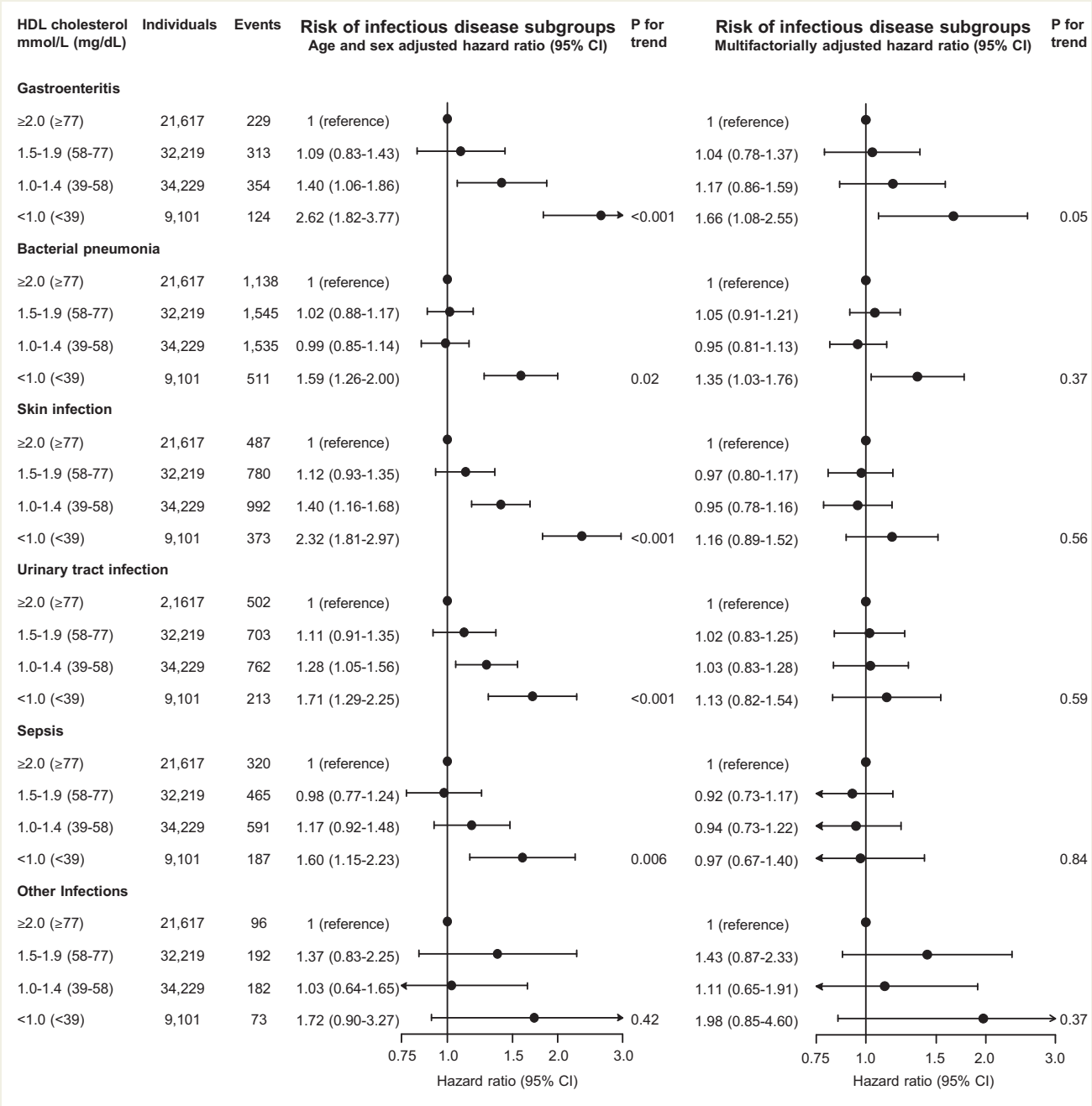


Figure 3 HDL cholesterol and risk of infectious disease in subgroups of diagnoses in 97 166 individuals from the Copenhagen General Population Study. Hazard ratios were from multiple-event Cox proportional hazards regression.

concentrations of HDL cholesterol, i.e. above 2.6 mmol/L (100 mg/dL), had a hazard ratio of 1.13 (95% CI 0.80–1.60) for risk of infectious disease.

Genetically determined HDL cholesterol and risk of infectious disease

The concentration of HDL cholesterol increased with increasing number of HDL cholesterol increasing alleles in an unweighted allele score of two common variants in the genes encoding hepatic lipase (LIPC) and cholesteryl ester transfer protein (CETP) (*P* for trend <0.001; left lower panel *Figure 8*). The risk of any infectious disease was lower with higher numbers of HDL cholesterol increasing alleles and hence higher genetically determined HDL cholesterol (left upper panel *Figure 8*). This seemed to be most pronounced in those with HDL cholesterol above the median (centre and right panel *Figure 8*). The risk of infectious disease related to the two variants in *LIPC* and *CETP* separately are shown in Supplementary material online, *Figure S11*. All covariates (not including apolipoprotein A1) were equally distributed across the different allele scores, except for LDL cholesterol, which

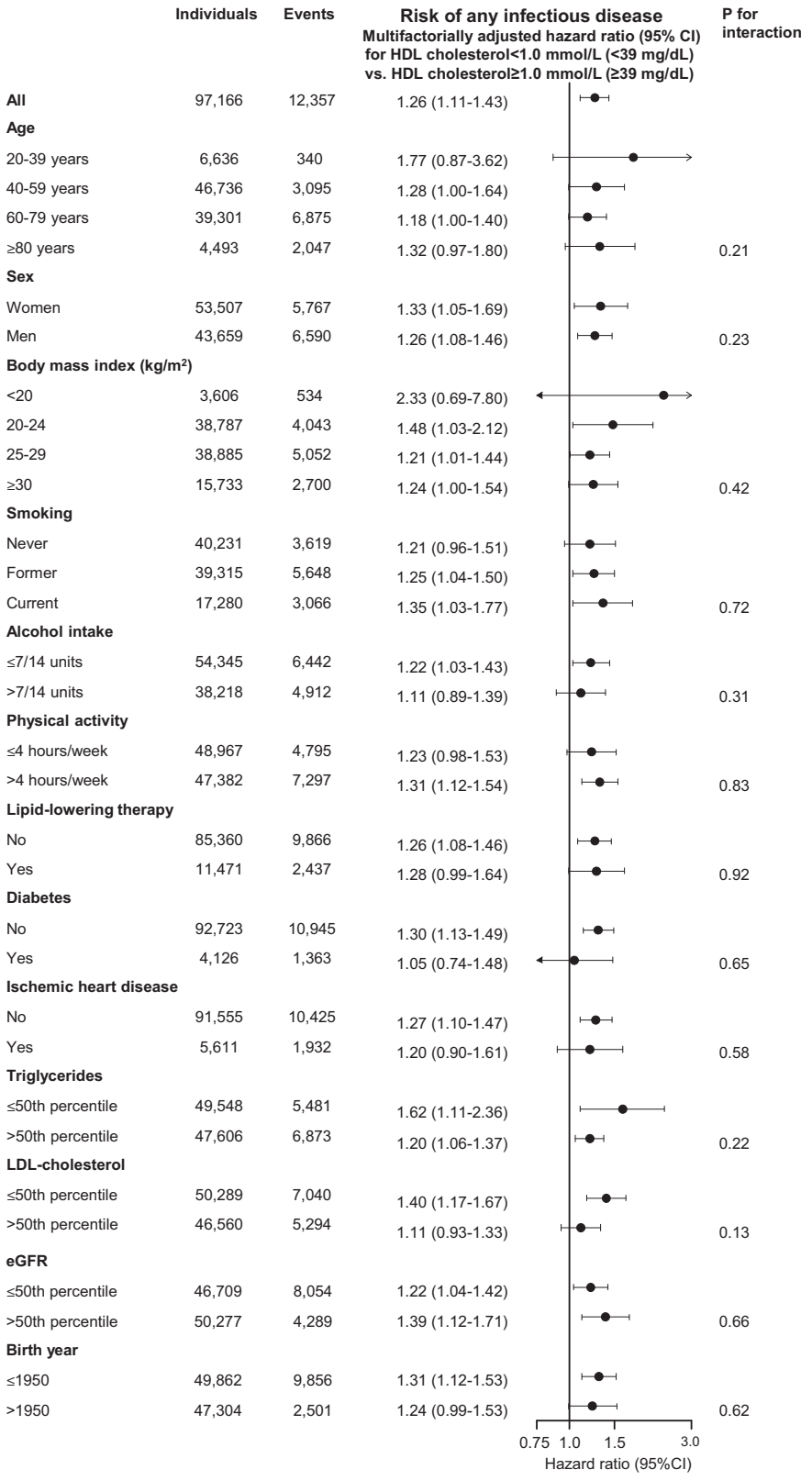


Figure 4 Risk of any infectious disease in different strata, for HDL cholesterol above vs. below 1 mmol/L in 97 166 individuals from the Copenhagen General Population Study. Hazard ratios were from multiple-event Cox proportional hazards regression. *P* for interaction was from Wald test. The total number of individuals varies slightly with the availability of the variables used for stratification.

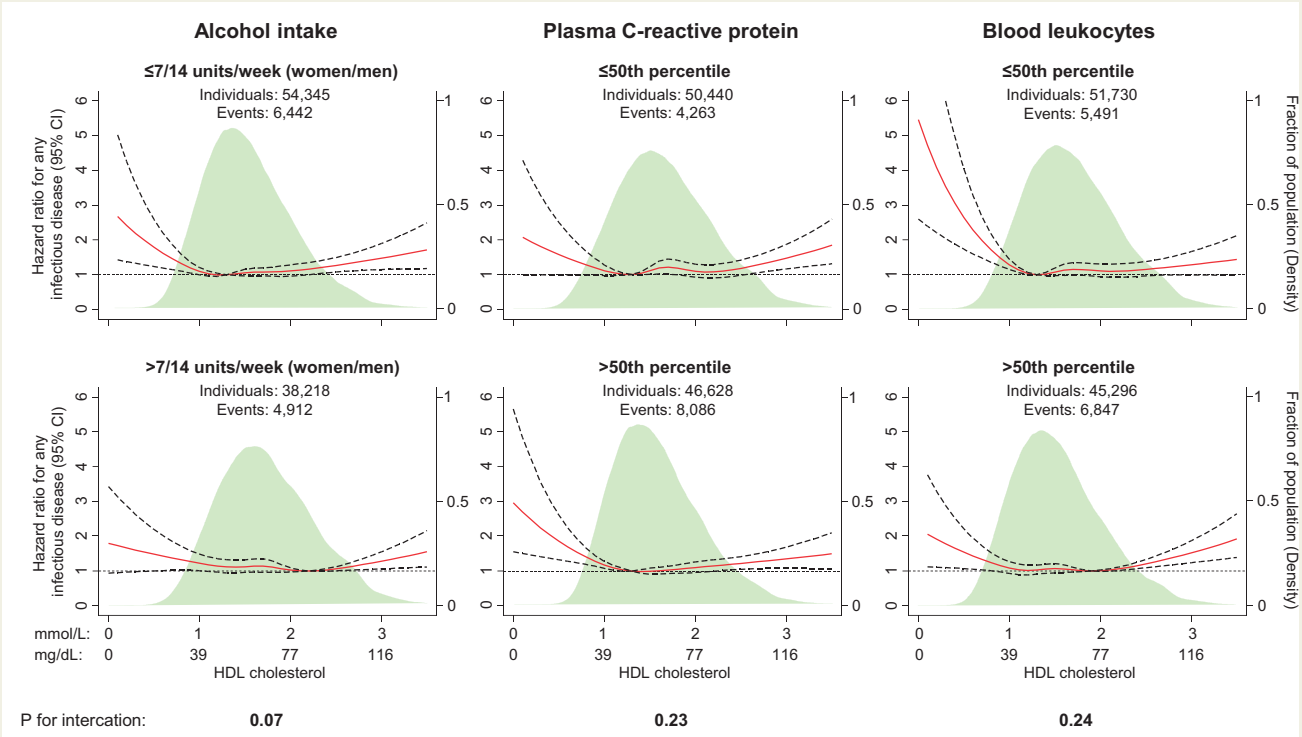


Figure 5 Risk of any infectious disease stratified by alcohol intake, plasma C-reactive protein, and blood leucocytes for HDL cholesterol on a continuous scale in 97 166 individuals from the Copenhagen General Population Study. Analyses were conducted using restricted cubic splines, with hazard ratios and 95% confidence intervals from multiple-event Cox proportional hazards regression. The value of HDL cholesterol with the lowest hazard ratio was chosen as reference. *P* for interaction was from Wald test. The light green areas indicate the distribution of concentrations of HDL cholesterol. The total number of individuals varies slightly with the availability of the variables used for stratification.

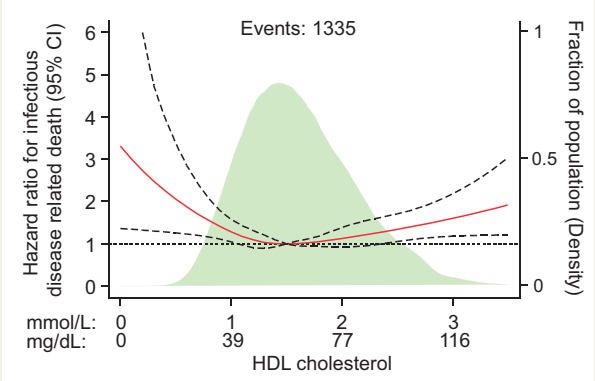


Figure 6 HDL cholesterol on a continuous scale and risk of infectious disease-related death in 97 166 individuals from the Copenhagen General Population Study. Analyses were conducted using restricted cubic splines, with hazard ratios and 95% confidence intervals from multiple-event Cox proportional hazards regression. The value of HDL cholesterol with the lowest hazard ratio was chosen as reference. The light green area indicates the distribution of concentrations of HDL cholesterol.

decreased with increasing number of HDL cholesterol increasing alleles (see Supplementary material online, *Table S12*). However, further adjusting the association between the allele score and risk of infectious disease for LDL cholesterol did not attenuate the association rather it was strengthened (data not shown).

Discussion

This study showed that low and high concentrations of HDL cholesterol found in 21% and 8% of the individuals in the general population were associated with high risk of infectious disease (*Take-home figure*). The highest risk estimates were seen for gastroenteritis and bacterial pneumonia. These findings are novel and were confirmed for low HDL cholesterol in an independent cohort. Importantly, these observational findings do not necessarily indicate causality.

Possible mechanisms and comparison with existing evidence

It is likely that the explanation for high infectious disease risk differ in individuals with low and high HDL cholesterol. Relevant for low HDL cholesterol, preclinical studies have provided several possible mechanisms by which HDL might contribute to normal functioning of the immune system.[9] Studies in mice have shown that HDL helps to control the proliferation of bone marrow progenitor cells.[10,11] Specifically, HDL inhibits the proliferation of haematopoietic stem cells in the bone marrow, thereby controlling the development of immune cells and avoiding inappropriate leucopoiesis. In later phases of the immune response, HDL acts as a modulator of immune cell

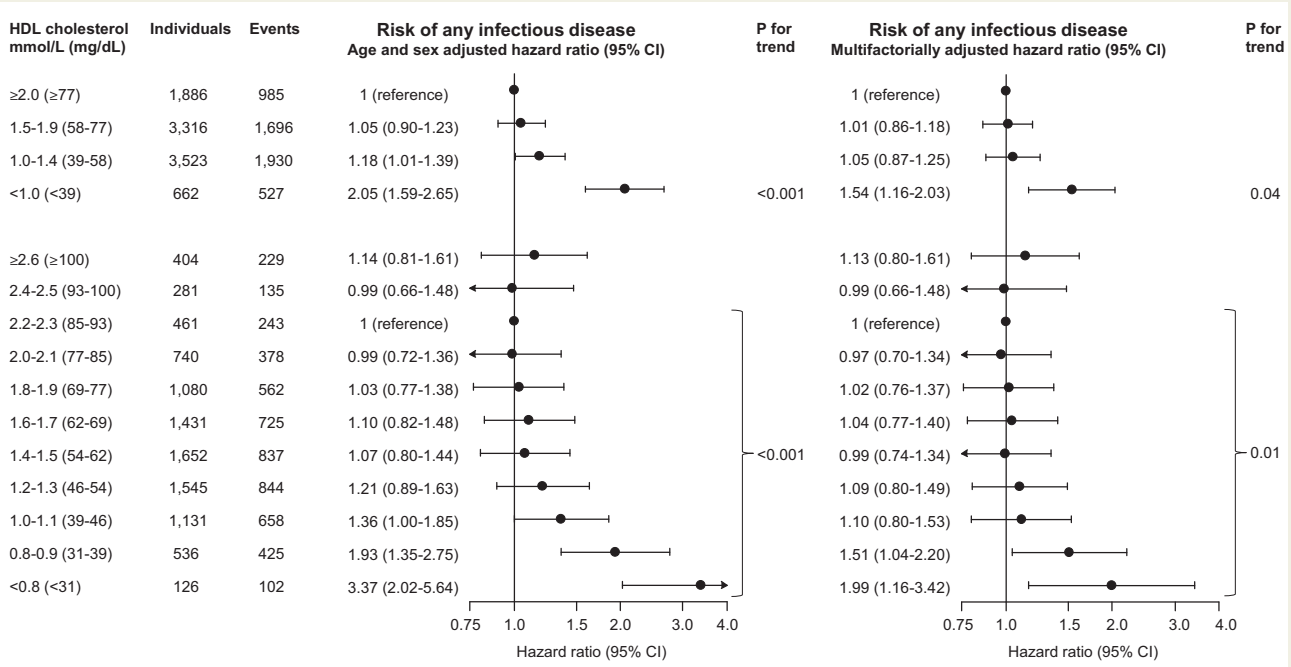


Figure 7 HDL cholesterol and risk of any infectious disease in 9387 individuals from the Copenhagen City Heart Study. Individuals are in the top part divided into 0.5 mmol/L (19 mg/dL) intervals and in the bottom part into 0.2 mmol/L (8 mg/dL) intervals of HDL cholesterol. Hazard ratios were from multiple-event Cox proportional hazards regression.

function and activity, which in part is believed to be based on the ability of HDL to change the cholesterol content in lipid rafts in the cell membrane, thereby affecting the activity of different cell surface receptors and the function of immune cells.[9,12] Additionally, HDL carries lipopolysaccharide-binding protein (LBP), which enables neutralization and clearance of endotoxins.[20] In future studies, it would be interesting to determine the association between HDL concentrations and concentrations of LBP in the HDL particles and how this affects infectious disease risk. Of note, potential defensive activities of HDL might also happen at mucosal barriers where HDL is secreted rather than in the blood stream.

Relevant for high HDL cholesterol, the very high concentrations of HDL cholesterol could be due to genetic variants that might have detrimental effects affecting disease susceptibility such is the case for certain mutations in, e.g. *LIPC* and *SCARB1*, which are associated with high risk of coronary heart disease.[21,22] Alternatively, the functional property of the HDL particles may be compromised in individuals with very high HDL cholesterol leading HDL to no longer function properly. HDL is an extremely complex particle that varies greatly in size and composition, and it has been attributed to several different functions, including being anti-inflammatory.[23] Hence, it is possible that some people with extreme high concentrations of HDL cholesterol may have HDL particles that are dysfunctional.

Reports on the association between HDL concentrations and infectious disease risk in humans are limited to five studies with 204 to 1719 hospitalized patients.[13–17] In accordance with our results, these studies have shown an association between low concentrations of HDL cholesterol at admission and high risk of infectious disease during hospital admission or shortly following. To the best of our knowledge, this study is the first one to examine the relationship between concentrations of HDL cholesterol and risk of infectious disease in two large prospective studies of the general population.

In addition to the high risk of infection seen at low concentrations of HDL cholesterol, we also observed high risk of infectious disease among those with high concentrations of HDL cholesterol in the Copenhagen General Population Study but not in the Copenhagen City Heart Study. An explanation could be that high concentrations of HDL cholesterol can be due to genetic variations that could have detrimental effects, as shown for cardiovascular disease.[22,24] Interestingly, in the ILLUMINATE trial with the CETP inhibitor torcetrapib, the torcetrapib group, with elevated HDL cholesterol, had more infectious disease-related deaths than the placebo group with lower HDL cholesterol (9 vs. 0 events).[25] This might reflect a qualitative difference in HDL raised in response to CETP inhibition different from that of ambient HDL measured in the general population.

Concentrations of HDL cholesterol show an inverse correlation with concentrations of triglycerides[2]; however, adjusting for triglycerides in the multifactorially adjusted analysis gave similar risk estimates and plasma concentrations of triglycerides were not associated with the risk of infectious disease. Hence, it is unlikely that the correlation between HDL cholesterol and triglycerides explained the findings in this study. Furthermore, low concentrations of apolipoprotein A1 were also associated with high risk of infectious disease similar to HDL cholesterol. In light of the association with HDL cholesterol, the absence of any relationship between risk of infection disease and plasma triglyceride concentrations is surprising. This could be explained by the fact that HDL cholesterol or triglycerides are not perfectly correlated and that it really is HDL *per se* that is important

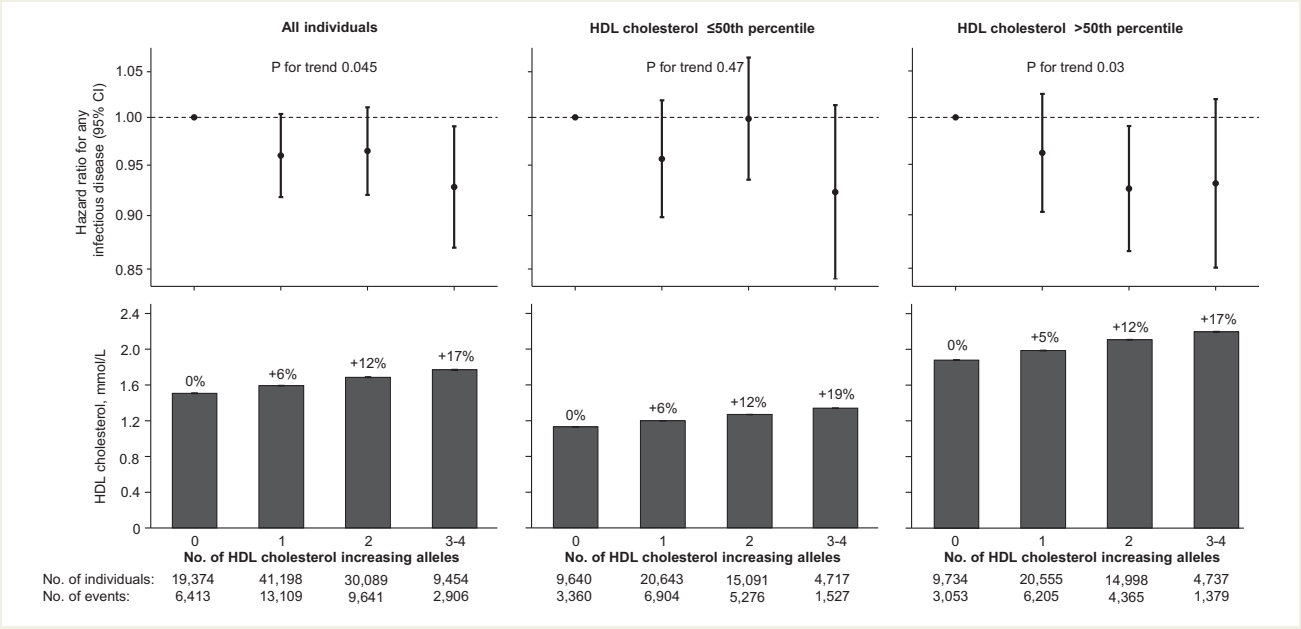


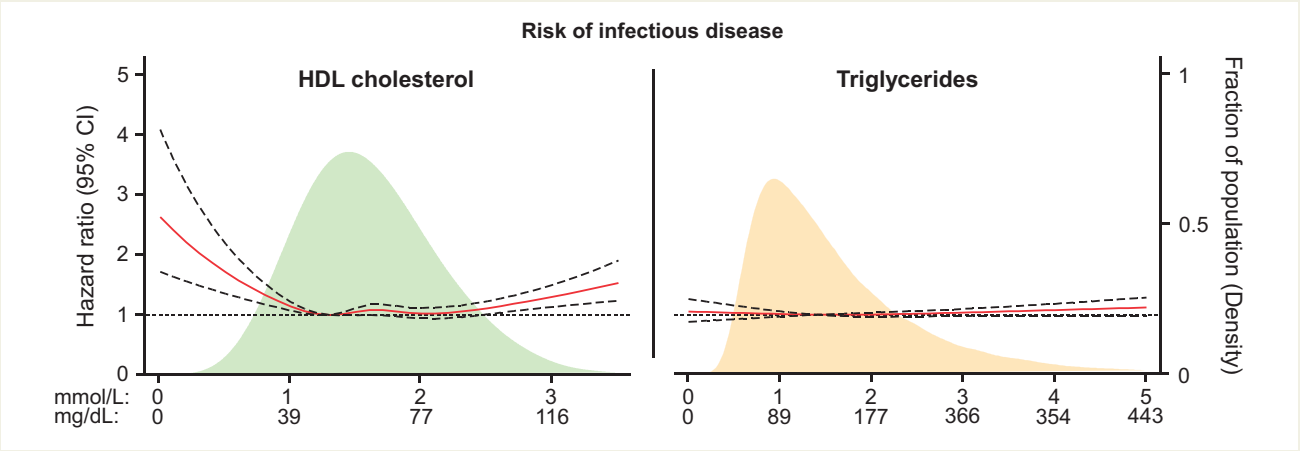
Figure 8 Genetically determined HDL cholesterol and risk of any infectious disease in 100 115 individuals from the Copenhagen General Population Study and Copenhagen City Heart Study combined. Lower panel shows the mean (±SEM) concentration of HDL cholesterol for individuals with the different allele scores with the percent change in HDL cholesterol compared to individuals in the group with zero HDL cholesterol increasing alleles. Upper panel shows the risk of any infectious disease with hazard ratios from multiple-event Cox proportional hazards regression adjusted for age and gender. For this analysis, infectious disease events were included from 1977 (start of the registry) or from birth, whichever came last. The left column includes all individuals, whereas the centre and right columns include individuals with HDL cholesterol below or above the median. Individuals were divided by the median of HDL cholesterol after the effect of the genotypes on the concentration of HDL cholesterol was removed.

for the risk of infectious disease. In addition, there are situations where a very high level of HDL cholesterol is associated with a high rather than a low concentration of plasma triglycerides, as is seen in people who consume excessive amounts of alcohol. However, stratifying for alcohol intake provided similar results in those with low and high alcohol intake.

Although recent results from genetic studies and randomized clinical trials have failed to show a beneficial effect of raising HDL cholesterol for the prevention of cardiovascular disease, there is still a strong focus on HDL cholesterol in this context in the current European Society of Cardiology (ESC)/European Atherosclerosis Society (EAS) Guidelines for the Management of Dyslipidaemias.[26] This focus should in the future perhaps be directed more at triglyceride-rich remnant lipoproteins when it comes to the prevention of cardiovascular disease,[27] while other possible roles of HDL outside of cardiovascular disease should receive additional attention, such as increased all-cause mortality[18] or the presently documented U-formed relationship with infectious disease.

Strengths and limitations

Due to the observational nature of our study, we are unable to conclude whether low HDL cholesterol is a causal risk factor for infectious disease. Thus, we cannot exclude that the association could be due to residual confounding or reverse causation; especially, this could be the case at the extremes of the HDL cholesterol distribution. Further to this, we also observed a strong attenuation of the association between HDL cholesterol and the risk of infectious disease after multifactorial adjustment, indicating that part of the association could be explained by possible confounders. HDL cholesterol is reduced in many chronic conditions which themselves are associated with high risk of infectious disease such as diabetes.[28] However, we did not observe an association between high concentrations of triglycerides, which are often present in individuals with poor health status, and risk of infectious disease, and also we adjusted and stratified our analyses for diabetes with similar results. Furthermore, there seemed to be reduced risk of infection in those with genetically higher HDL cholesterol, which could indicate a causal relationship, as genetic variants are not subject to reverse causation and are less likely to be confounded. This is because genetic variants are present at birth and because the random assortment of genetic variants during gamete formation leads to equal distribution of confounders in individuals with the different genotypes. However, the results from the genetic analyses has to be interpreted with caution and needs to be confirmed in additional studies, as the association between HDL cholesterol and risk of infectious disease does not appear to be linear and as the effect of the allele score on HDL cholesterol concentrations was only modest. Another limitation relates to the use of the national registries for diagnoses of infectious diseases. These only capture infections severe enough to lead to hospital contact and not all are confirmed by microbiological examinations. In addition, there might be a referral bias for individuals who are generally less well who are more often referred to hospital and treated for infections.



Take-home figure HDL cholesterol and triglycerides on a continuous scale and risk of any infectious disease in 97 166 individuals from the Copenhagen General Population Study. Analyses were conducted using restricted cubic splines, with hazard ratios and 95% confidence intervals from multiple-event Cox proportional hazards regression. The values of HDL cholesterol and triglycerides with the lowest hazard ratio were chosen as the reference. The light green and yellow areas indicate the distribution of concentrations of HDL cholesterol and triglycerides respectively.

That said, such a referral bias will not operate for the endpoint infectious disease-related death, and results using this endpoint were similar to the overall results. Also, the use of data from the national Danish registries on infectious disease has previously been suggested to provide valid results.[29] Furthermore, generalizability of the results may be reduced by the fact that we only investigated White individuals of Danish descent, but we are not aware of data to indicate that the results of this study may be different in other ethnic groups.

Strengths include the large number of prospectively recruited individuals from the general population, no losses to follow-up, and detailed information on covariates. In addition, we confirmed our findings in an independent cohort including individuals examined in another time period.

Conclusions

In conclusion, we found that low and high concentrations of HDL cholesterol found in 21% and 8% of all individuals in the general population were associated with high risk of infectious.This indicates that HDL may play a role in normal function of the human immune system.

Supplementary material

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

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