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The association between low density lipoprotein (LDL) and RA genetic factors with LDL levels in rheumatoid arthritis and non-RA controls

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Abstract

Objectives—While genetic determinants of LDL cholesterol levels are well characterized in the general population, they are understudied in rheumatoid arthritis (RA). Our objective was to determine the association of established LDL and RA genetic alleles with LDL levels in RA cases compared to non-RA controls.

Methods—Using electronic medical records (EMR) data, we linked validated RA cases and non-RA controls to discarded blood samples. For each individual, we extracted data on: 1st LDL measurement, age, gender, and year of LDL measurement. We genotyped subjects for 11 LDL and

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44 non-HLA RA alleles, and calculated RA and LDL genetic risk scores (GRS). We tested the association between each GRS and LDL level using multivariate linear regression models adjusted by age, gender, year of LDL measurement, and RA status.

Results—Among 567 RA cases and 979 controls, 80% were female and the mean age at 1st LDL measurement was 55 years. RA cases had significantly lower mean LDL levels than controls (117.2 vs. 125.6mg/dL, respectively, $p < 0.0001$). Each unit increase in LDL GRS was associated with 0.8mg/dL higher LDL levels in both RA cases and controls ($p = 3.0 \times 10^{-7}$). Each unit increase in RA GRS was associated with 4.3mg/dL lower LDL levels in both groups ($p = 0.01$).

Conclusions—LDL alleles were associated with higher LDL levels in RA. RA alleles were associated with lower LDL levels in both RA cases and controls. Since RA cases carry more RA alleles, these findings suggest a genetic basis for epidemiologic observations of lower LDL levels in RA.

Keywords

Rheumatoid arthritis; low density lipoprotein; genetics; human leukocyte antigen

INTRODUCTION

Low density lipoprotein (LDL) cholesterol, a major risk factor for coronary artery disease (CAD), has been observed to be lower in rheumatoid arthritis (RA) patients compared to individuals of similar age and gender from the general population.[1, 2] Despite lower LDL levels, RA patients are at higher risk for CAD than the general population.[3] The increased risk for CAD, as well as lower LDL levels has been attributed to the excess inflammation in patients with RA.[4, 5] However, the extent to which pathways involved with RA pathogenesis are also associated with lower LDL levels remain unclear.

The genetic determinants of LDL levels are well characterized in the general population.[6, 7] Genome-wide association studies (GWAS) and related approaches have identified ~30 alleles that explain approximately 12% of the variation of LDL levels in the general population.[7] A longitudinal study, collecting serial LDL levels over the course of 9 years found that the association between an aggregate LDL genetic risk score (GRS) and LDL levels was robust and remained relatively stable over time.[8] The strongest clinical determinants of LDL levels in subjects not on lipid lowering therapy were age, gender and the year of LDL measurement.[9, 10] Whether the LDL GRS characterized from the general population explains LDL levels in RA is unknown.

The genetic determinants of RA play a major role in the risk of developing RA[11] and are associated with dysfunction in immune pathways and inflammation.[12] Sepsis, an extreme state of inflammation, is associated with lower and in some cases, undetectable LDL levels. [13, 14] While the exuberant inflammation of sepsis is mainly caused by an acute response to environmental pathogens, the immune dysregulation in RA is a chronic inflammatory process and may be determined in large part by an individual's underlying genetic make-up. This suggests that the genetic factors associated with dysfunction in immune pathways that lead to RA may influence LDL levels in these patients.

Thus far, only one study has investigated whether RA susceptibility alleles are associated with LDL levels.[15] This study examined the association of single nucleotide polymorphisms (SNPs) with lipid levels in three RA risk genes [*PTPN22*, *TRAF1/C5*, *STAT4*], and the human leukocyte antigen shared epitope (*HLA-SE*) alleles. They observed that RA patients carrying one or more risk alleles for *TRAF1/C5* had significantly lower LDL levels than those who did not (3.15mmol/L [121.8 mg/dL] vs. 3.48 mmol/L [134.6mg/dL], $p=0.02$). No association between the other RA risk alleles and LDL levels was observed.

In this study we utilized RA genetic risk alleles as markers of RA specific pathways of immune dysregulation. If RA specific pathways are associated with LDL levels, then we would expect an association of RA risk alleles with LDL levels in RA as well as non-RA subjects. The objectives of this study were: (1) to compare LDL levels in RA cases compared to non-RA controls, (2) to test whether alleles associated with LDL levels in the general population were also associated with LDL levels in RA patients; and (3) to test whether RA risk alleles were associated with LDL levels in a RA case cohort and a non-RA control cohort. We hypothesize that genetic variants associated with higher LDL levels in the general population will also be associated with higher LDL levels in RA, but that a higher RA aggregate GRS will be associated with lower LDL levels. In this study, we also applied newly developed methods to study the associations of the *HLA-SE*[16] with LDL levels.

METHODS

Study population

We studied a validated RA cohort[17, 18] and a non-RA control cohort[19] of European ancestry with available calculated LDL (LDL-C) or direct LDL (LDL-D) measurements in the electronic medical records (EMR) of Brigham and Women's Hospital (BWH) and Massachusetts General Hospital (MGH) from 1989–2007. Briefly, the RA cases were identified using an RA phenotype algorithm which utilizes a combination of International Classification of Diseases, 9th Revision (ICD9) and clinical data extracted using bioinformatics methods to mine narrative text (natural language processing). The positive predictive value (PPV) for RA using the algorithm is 94%. For details on the development and validation of this cohort please refer to Liao, et al., *Arthritis Care and Research* 2010. [17]

The non-RA cohort was selected by first excluding all patients with an ICD9 code for a rheumatic disease as reported in Kurreeman, et al., *American Journal of Human Genetics*. [19] Briefly, non-rheumatic disease subjects were selected based on similar age, gender, race and health care utilization to the RA cases. Health care utilization was approximated by using the number of “facts” which are points of contact with the health care system, i.e. laboratory blood draws, clinic visits, x-rays. For details on the development of the non-RA control cohort and sample collection please refer to Kurreeman, et al., 2010.[19] We collected discarded blood samples for each cohort using the BWH Biospecimen Repository.

Variables

The primary outcome, LDL, was defined by each subject's first LDL measurement in the EMR, to maximize the chance of selecting subjects prior to any lipid lowering intervention. We excluded patients who had an electronic prescription for an HMG-CoA reductase inhibitor (statin) prior to their first LDL level. We extracted other lipid levels [total cholesterol (Tchol), high density lipoprotein (HDL)] measured within one year of the index LDL. Age at LDL measurement, gender, and the year of LDL measurement were also extracted from the structured EMR data.

RA cases must have prevalent RA at the time of the first LDL measurement. Prevalent RA was defined as subjects in the RA cohort with 1 RA ICD9 code or narrative text mention of RA prior to the 1st LDL measurement.

Genotyping

We genotyped individuals using the Illumina BeadExpress (n=384 SNPs) and the Illumina ImmunoChip.[20] The ImmunoChip dataset clustering and initial filtering were performed as described previously.[21] We excluded individuals with call rate<97%, and SNPs with missingness>0.02 or departure from Hardy-Weinberg equilibrium ($P_{HWE}<0.001$). We also computed a chi-square test to assess the difference in missingness between RA cases and non-RA controls and removed SNPs with a $P_{missing}<10^{-2}$.

To address population stratification, we selected a set of common SNPs (MAF>5%) in the filtered ImmunoChip data, pruned to remove SNPs in linkage disequilibrium (LD). We calculated pairwise identity-by-state (IBS) statistics using PLINK,[22] and removed one individual from each pair of individuals who were 2nd degree or closer relatives. Principal components analysis (PCA) was subsequently performed using EIGENSTRAT[23] with HapMap phase III samples. We limited our study to individuals of European ancestry because the majority of published genetic data for lipid alleles were conducted in this population, and to mitigate confounding from population stratification.

Classical allele genotypes for *HLA-DRB1*, *HLA-B* and *HLA-DPB1* were imputed using methods from Raychaudhuri, et al, Nature Genetics 2012.[16]

Genetic risk scores

We constructed three separate aggregate genetic risk scores (GRSs) for LDL, non-HLA RA risk alleles [24] and the HLA region[16] to test for association with LDL levels. We refer to these GRSs as the LDL, RA and HLA GRSs, respectively. Individual SNPs have modest effect sizes and would have limited power to demonstrate an association with LDL levels. The GRSs allow for testing of SNPs in aggregate (grouped by their association with clearly defined phenotypes, i.e. LDL, RA), allowing for increased power to detect an association with LDL levels[24, 25]. For example, rather than testing the association of each of the 44 non-HLA RA risk alleles with LDL levels (44 tests), we tested the RA GRS (aggregated score of 44 SNPs) with LDL levels (1 test).

The weighted LDL genetic risk score (GRS) was constructed using 11 single nucleotide polymorphisms (SNPs) associated with LDL as the lead trait from published studies[7, 25] and weighted by published effect sizes for higher levels of LDL (mg/dL) (Appendix 1). Not all alleles associated with LDL were examined due to the timing of genotyping with Illumina BeadExpress for this study relative to the publication of the LDL genetic studies.

Due to the strong genetic contribution of the HLA compared to non-HLA RA risk loci,[11] we studied separately, the association of the HLA RA (with the HLA GRS) and non-HLA RA risk alleles (with the RA GRS) with LDL levels. If an association was observed, this approach would allow us to determine whether it arose from the HLA or the non-HLA RA risk loci. We created an RA GRS using 44 published non-HLA SNPs associated with RA risk,[11, 20] weighted by the natural log of the odds ratios (OR) for risk of RA from the most recent meta-analysis[20](Appendix 1). Individuals with missing genotypes for a given SNP were assigned twice the expected frequency of the risk allele in the samples with the same phenotype (RA cases or non-RA controls). We constructed the HLA GRS as per Raychaudhuri, et al. Nature Genetics 2012,[16] weighted by the natural log of the published effect sizes for risk of RA.

All three GRSs were constructed using the general formula:

$$GRS = \sum_i^n w_i X_i$$

Where i =SNP; w_i =natural log of published ORs (HLA and non-HLA RA risk alleles) or the published effect size (LDL, mg/dL); X_i =number of risk alleles (0, 1, or 2).

Laboratory analyses

Anti-citrullinated peptide/protein antibodies (ACPA) were measured using the INOVA CCP3 IgG ELISA.[19] We determined positivity based on the manufacturer cut-off for ACPA 20 units.

Statistical analyses

We conducted univariate analyses to compare age, gender, LDL, year of LDL measurement, and total cholesterol (Tchol) and high density lipoprotein (HDL) levels across prevalent RA and non-RA controls. We applied t-tests to compare means and chi-square tests to compare differences in proportions. To determine differences in LDL levels between RA cases and non-RA controls, we constructed a linear regression model with RA cases status and LDL, adjusted by age, gender, and year of LDL measurement. To test whether ACPA status was associated with LDL levels among RA cases, we constructed a linear regression model with ACPA status and LDL levels, adjusted by age, gender, and year of LDL measurement. All models were adjusted by factors with known significant associations with LDL levels: age and gender as was done in previous genetic association studies of LDL from the general population[7, 25] as well as year of LDL measurement which was recently identified to have secular trends over time.[9]

Our 2 main analyses were: (1) to determine the association between the LDL GRS and LDL levels in RA cases and non-RA controls, and (2) to determine the association between the RA GRS and LDL levels in RA cases and non-RA controls. We tested the association between the LDL GRS and LDL levels in RA cases and non-RA controls by constructing a multivariate linear regression model containing age, gender, year of LDL measurement, and two interaction terms, RA case*LDL GRS and control*LDL GRS. The interaction terms provide the magnitude of effect for the association between the LDL GRS and LDL levels separately for RA cases (β coefficient for 'RA case*LDL GRS') and non-RA controls (β coefficient for 'control*LDL GRS').

To determine if the associations between the LDL GRS and LDL levels were significantly different in RA cases compared to non-RA controls (differences in the β coefficients for 'RA case*LDL GRS' and 'control*LDL GRS'), we constructed a multivariate linear regression model with age, gender, year of LDL measurement, RA case status (yes/no), RA case status*LDL GRS, and association with LDL levels. A significant interaction term (RA case status*LDL GRS, $p < 0.05$), would suggest a significant difference in the association between the LDL GRS and LDL in RA cases compared to non-RA controls.

We conducted the same steps above to determine the association between the RA GRS and LDL levels among RA cases and non-RA controls as well as the HLA GRS and LDL levels among RA cases and non-RA controls. We also utilized the interaction tests detailed above to determine significant differences in the association between the RA GRS and LDL levels in RA cases compared to non-RA controls.

If the associations of the GRSs and LDL levels were similar in the two groups, we tested whether given the same LDL or RA GRS, if RA cases had similar LDL levels to non-RA controls. To test for this difference, we constructed a linear regression model with RA case status (yes/no) and LDL levels, adjusted by age, gender, year of LDL measurement and the LDL or RA GRS.

To visualize the trends in LDL levels associated with increasing numbers of LDL and RA genetic alleles, we first determined cutoffs for tertiles of the RA GRS and LDL GRS separately in RA cases and non-RA controls. We then calculated the mean LDL levels in tertiles of the RA GRS and LDL GRS.

For our sensitivity analyses, we reanalyzed the data using the highest LDL reported for each patient in the EMR as the outcome, regardless of statin use.

This study was approved by the Partners Healthcare Institutional Review Board (IRB for BWH and MGH). Statistical analyses were conducted using SAS 9.2 (Cary, NC).

RESULTS

LDL levels in RA cases vs. non-RA controls

We identified 567 subjects with prevalent RA and 979 non-RA controls with available LDL measurements spanning from 1989 to 2007 (Table 1). All subjects were of European ancestry and had genotype data available. The mean ages at first LDL measurement was

similar in the two groups: 54.5 for RA cases and 55.2 years in controls. The majority of subjects in both groups were women (80%). In the univariate analyses, RA cases had a mean LDL of 117.2 mg/dL, which was significantly lower than in non-RA controls (mean LDL 125.6 mg/dL, $p<0.0001$).

We compared LDL levels in RA cases and controls, adjusting for age, gender and year of LDL measurement, and observed that LDL was on average 5mg/dL lower in prevalent RA cases than in non-RA controls ($p=0.003$). Among RA cases, there was no significant difference between mean LDL levels in ACPA positive ($n=422$) compared to ACPA negative ($n=145$) subjects, adjusted by age, gender and year of LDL measurement ($p=0.94$).

Association of LDL genetic risk score with LDL levels

We next tested whether an LDL genetic risk score (GRS) was associated with LDL levels in RA cases and non-RA controls. Among RA cases, each unit increase in the LDL GRS was associated with a 0.72mg/dL higher LDL level ($p=6.2\times 10^{-6}$). Among controls, each unit increase in the LDL GRS was associated with a 0.85 mg/dL higher LDL level ($p=9.4\times 10^{-8}$, Table 2a). The association between the LDL GRS and LDL levels was not significantly different in RA cases and controls (interaction test, $p=0.45$).

Since the association between the LDL GRS and LDL level was similar in the two groups, we investigated whether LDL levels were significantly different in RA cases compared to controls, controlling for the LDL GRS as well as age, gender and year of LDL measurement. We observed that RA cases still had on average 4.3mg/dL lower LDL levels than non-RA controls ($p=0.03$) (Table 3a).

Association of RA genetic risk score and an HLA genetic risk score with LDL levels

Since neither clinical factors nor an LDL GRS could explain the difference in LDL levels between RA cases and controls, we tested whether genetic factors enriched in RA patients may explain the observed difference. First, we tested an RA GRS composed of 44 non-HLA RA risk SNPs for association with LDL levels in both RA cases and non-RA controls. Second, we tested an RA risk model composed of SNPs within the HLA region, which have been imputed to tag 5 amino acids in three HLA genes.[16] And third, we tested the non-HLA RA GRS after controlling for the effect of the LDL GRS in a combined analysis of RA cases and non-RA controls.

We observed that each unit increase in the RA GRS was associated with 4.6mg/dL lower LDL levels ($p=0.007$) among RA cases, and a 4.3mg/dL lower LDL levels in controls ($p=0.02$) (Table 2b). The association between the RA GRS and LDL levels was not significantly different among RA cases and controls (interaction test, $p=0.17$). Out of 44 non-HLA RA risk alleles, 28 (64%) were associated with lower LDL levels, whereas only half would be expected by chance alone. No single RA risk allele was significantly associated with LDL levels after adjusting for multiple comparisons ($p<0.001$) (Appendix 2B).

We observed no association between an aggregate HLA GRS and LDL levels in either RA cases or non-RA controls ($p=0.90$). No single amino acid residue was associated with LDL levels in either RA cases or controls.

Since the association between the RA GRS (non-HLA) and LDL level was similar in RA cases and controls, we also tested whether LDL levels were similar in RA cases compared to controls, controlling for the RA GRS as well as age, gender, year of LDL measurement. In this model, given a similar RA GRS, RA cases and controls had similar LDL levels ($p=0.30$) (Table 3b).

Since we observed no difference between the associations of the LDL GRS or RA GRS and LDL levels in RA cases compared to controls (interaction tests all non-significant), we report a combined estimate for the association of the GRS and LDL levels from RA cases and controls for subsequent analyses. To determine whether the RA GRS was independently associated with LDL levels beyond the LDL GRS, we included both the RA GRS and LDL GRS in one model (Table 4). We observed that the RA GRS remained associated with lower LDL levels ($p=0.008$) while the LDL GRS remained associated with higher LDL levels ($p=9.3\times 10^{-7}$) in RA cases and controls (Table 4). The lack of interaction between the RA GRS and LDL GRS ($p=0.42$) suggests that the association of RA risk alleles with LDL levels was independent of the association of LDL alleles with LDL levels. In general, RA cases or controls with the highest LDL GRS and the lowest RA GRS had the highest LDL levels (Figure 1).

Sensitivity analyses

We extracted the highest LDL level recorded in the EMR for each subject and observed similar effect sizes for association between GRS and LDL level, regardless of statin status. Each unit increase in the LDL GRS was associated with 0.64mg/dL higher LDL level, using the highest LDL level as the outcome (compared to 0.81mg/dL using the first LDL measured as the outcome). Each unit increase in the RA GRS was associated with a 3.5mg/dL lower LDL level (compared to 4.6mg/dL).

DISCUSSION

There are three main findings from our study. First, we observed that LDL levels were lower in RA cases compared to non-RA controls, adding to the few published epidemiologic studies on this topic.[1, 2] Second, our findings replicate previous studies that LDL alleles in aggregate are associated with higher LDL levels among non-RA controls,[7] and demonstrate that the same LDL alleles have similar effects in RA subjects. Third, we observed associations that non-HLA RA risk alleles in aggregate are associated with lower LDL levels in both RA cases and non-RA controls.

These findings provide a potential explanation for why LDL levels are lower in RA than non-RA controls. By definition, RA patients carry more RA risk alleles than non-RA controls. In this study, RA cases had a significantly higher RA GRS than non-RA controls ($p=1.93\times 10^{-21}$). Since carriage of a higher number of RA risk alleles was associated with lower LDL levels, RA patients as a group had lower LDL levels than non-

RA controls. Moreover, the association of the RA GRS with lower LDL remained after adjusting for clinical factors associated with LDL levels (age, gender, year of LDL measurement) and the LDL GRS. RA risk alleles identified mainly through GWAS are common alleles, and were relatively common in the non-RA control group (Appendix 3). Thus, we also had adequate power to detect an association between the RA GRS and LDL levels in non-RA controls.

These results suggest that pathways associated with RA immune dysregulation, represented by RA risk alleles, are associated with lower LDL levels in prevalent RA. This is in agreement with prior hypotheses that increasing immune dysregulation prior to the onset of RA could be the cause of decreasing LDL levels up to RA onset [2, 26] and during active disease.

Our study represents a comprehensive analysis of the association of RA risk alleles on LDL cholesterol levels. Rather than focusing on a small number of individual RA risk alleles, we studied the effect of known RA risk alleles in aggregate using the most up-to-date genetic data. [20] We also employed recent methods of determining HLA status to test for association of HLA alleles with LDL levels. Toms, et al., studied four RA risk alleles including the HLA-SE and found that only one, *TRAF1/C5*, was significantly associated with lower LDL levels. In our study, *TRAF1* (rs3761847), was not associated with lower LDL levels. However, in agreement with Toms et al., [15] we found no association between HLA RA risk alleles and LDL level.

There were limitations to this study. First, although 89% of subjects have a primary physician in our health care system, the system is not closed and incomplete capture of patient information, i.e. statin prescriptions, lipid levels, is a possibility. We excluded subjects on statins to remain in concordance with previous lipid GWAS meta-analyses. [7] Second, the non-RA control cohort in this study is not a random sample of the general population. They comprise individuals who utilize the health care system in a manner similar to RA patients, without a rheumatic disease but have other chronic diseases, including those with non-rheumatic inflammatory disease. This approach allowed for the creation of a comparison group of individuals with similar age, gender, and health care utilization and therefore similar opportunities to have lipid studies in the system as the RA cohort. In contrast, the general population or healthy individuals are difficult to define in an EMR. They may have less data, making it difficult to determine if they were regularly followed in our healthcare system. Third, we did not have accurate information on fasting status, although a recent study demonstrated that fasting and non-fasting variations in LDL levels at the population level were small. [10] Finally, our models included all variables known to be significantly associated with LDL levels from published studies: age, gender and year of LDL measurement. Our approach follows those of previous large genetic association studies of LDL conducted in the general population (which analyzed several cohorts with data spanning 5 decades), [6, 7] and allows for comparison of results with these studies. Other RA clinical factors such as disease activity and RA treatments may also be associated with LDL levels, however, the significance and magnitude of these associations are conflicting. [27] Therefore, these factors were not included as covariates in this study.

In summary, we observed that RA cases had lower LDL levels than controls. This difference can be partially explained by the higher burden of RA risk alleles in RA cases. Future studies are needed to replicate our results and to determine the clinical implications of these findings. Specifically, since LDL levels in RA patients reflect the combined effects of distinct gene subsets involved in LDL metabolism and RA pathogenesis (and immune dysregulation), LDL levels may have different prognostic implications for CAD in RA patients. More investigation is needed to determine the target LDL levels for CAD risk prevention in RA patients who have a higher burden of RA risk alleles.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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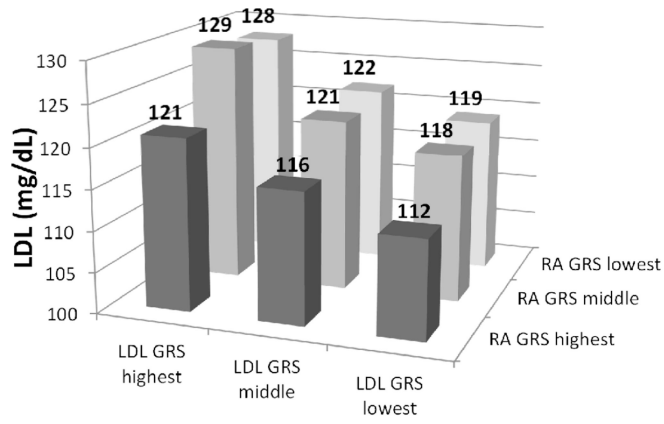
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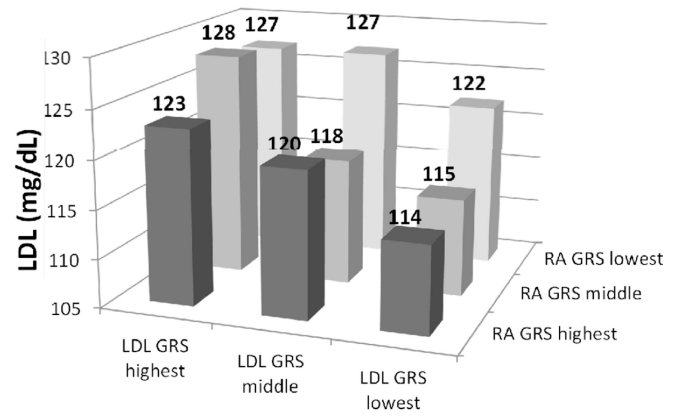
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a. RA cases, n=516



b. Non-RA controls, n=930

Figure 1.

Mean levels of LDL among RA cases grouped by tertiles of the LDL and RA GRS for **(a)** RA cases and **(b)** non-RA controls. (Note: no interaction observed between the RA GRS and LDL GRS)

Table 1

Clinical characteristics of prevalent RA cases compared to non-RA controls.

Clinical characteristics	RA cases, n=567	Non-RA controls, n=979	p-value
Age LDL measured, yrs	54.5 (12.3)	55.2 (12.5)	0.29
Female, n (%)	456 (80.4)	777 (79.4)	0.65
Mean year LDL measurement (SD)	2000 (4.9)	1998 (5.2)	<0.0001
ACPA status, n (%)	422 (74.4)	1 (0.6)*	<0.0001
Lipoprotein levels (mg/dL)			
LDL, mean (SD)	117.2 (36.5)	125.6 (41.7)	<0.0001
Tchol, mean (SD)	199.4 (43.5)	208.3 (49.9)	0.0002
HDL, mean (SD)	56.1 (18.1)	53.8 (17.8)	0.01

* out of 156 controls with ACPA data

Table 2

The association between (a) the LDL GRS and LDL levels, and (b) the RA GRS and LDL levels, among RA cases and non-RA controls, adjusted by age, gender and year of LDL measurement.

Variables	(a)Association of LDL GRS with LDL levels		(b)Association of RA GRS with LDL levels	
	Beta coefficient (SE)	p-value	Beta coefficient (SE)	p-value
Age	0.34 (0.08)	1.4×10^{-5}	0.34 (0.08)	1.9×10^{-5}
Female gender	4.90 (2.41)	0.04	6.23 (2.44)	0.01
Year of LDL measurement	-2.55 (0.19)	8.6×10^{-39}	-2.60 (0.19)	1.50×10^{-39}
RA case*GRS	(LDL GRS) 0.72 (0.16)	6.2×10^{-6}	(RA GRS) -4.58 (1.71)	0.007
Control*GRS	(LDL GRS) 0.85 (0.16)	9.4×10^{-8}	(RA GRS) -4.26 (1.79)	0.02

RA cases with RA GRS data, n=541 Non-RA controls with RA GRS data, n=945

Interaction tests: No significant difference in association between LDL GRS and LDL levels in RA cases and non-RA controls, p=0.45

No significant difference in association between RA GRS and LDL levels in RA cases and non-RA controls, p=0.17

Note: RA cases with LDL GRS data, n=542 Non-RA controls with LDL GRS data, n=963

Table 3

Association between RA case status (yes/no) and LDL levels in a model containing **(a)** the LDL GRS, and **(b)** the RA GRS, adjusted by age, gender and year of LDL measurement.

Variables	(a)LDL GRS		(b)RA GRS	
	Beta coefficient (SE)	p-value	Beta coefficient (SE)	p-value
RA case status	-4.31 (2.04)	0.03	-2.15 (2.11)	0.31
GRS	0.81 (0.15)	3.0×10^{-7}	-4.32 (1.75)	0.01

RA cases with RA GRS data, n=541 Non-RA controls with RA GRS data, n=945

Interaction tests: No significant differences in association of GRSs between RA cases and controls. Therefore, GRSs combined into one average estimate in these models.

Note: RA cases with LDL GRS data, n=542 Non-RA controls with LDL GRS data, n=963

Table 4

The association between RA GRS, LDL GRS and LDL levels in a multivariable model adjusted by age, gender and year of LDL measurement in RA cases and non-RA controls.

Variable	Beta coefficient (SE)	p-value
RA case status	-3.25 (2.14)	0.13
RA GRS	-4.68 (1.76)	0.008
LDL GRS	0.78 (0.16)	9.31×10^{-7}

Note: Subjects with both complete LDL GRS and RA GRS data, RA cases, n=516; Non-RA controls, n=930