EXTENDED REPORT

Association between low density lipoprotein and rheumatoid arthritis genetic factors with low density lipoprotein levels in rheumatoid arthritis and non-rheumatoid arthritis controls

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Handling editor Tore K Kvien

Additional material is published online only. To view please visit the journal online (http://dx.doi.org/10.1136/annrheumdis-2012-203202). For numbered affiliations see end of article.

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Accepted 5 May 2013

ABSTRACT

Objectives While genetic determinants of low density lipoprotein (LDL) cholesterol levels are well characterised 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 with non-RA controls.

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

Results Among 567 RA cases and 979 controls, 80% were female and mean age at the first LDL measurement was 55 years. RA cases had significantly lower mean LDL levels than controls (117.2 vs 125.6 mg/dl, respectively, p<0.0001). Each unit increase in LDL GRS was associated with 0.8 mg/dl higher LDL levels in both RA cases and controls (p=3.0x10^{-5}). Each unit increase in RA GRS was associated with 4.3 mg/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. As RA cases carry more RA alleles, these findings suggest a genetic basis for epidemiological observations of lower LDL levels in RA.

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 with 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.1 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 the pathways involved with RA pathogenesis are also associated with lower LDL levels remains unclear.

Genetic determinants of LDL levels are well characterised 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 in LDL levels in the general population.8 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.9 The strongest clinical determinants of LDL levels in subjects not on lipid lowering therapy were age, gender and year of LDL measurement.10 10 Whether the LDL GRS characterised from the general population explains LDL levels in RA is unknown.

Genetic determinants of RA play a major role in the risk of developing RA11 and are associated with dysfunction in immune pathways and inflammation.12 Sepsis, an extreme stage 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 makeup. 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 leucocyte 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 (3.15 mmol/l (121.8 mg/dl) vs 3.48 mmol/l (134.6 mg/dl), p=0.02). No association between the other RA risk alleles and LDL levels was observed.

In this study, we utilised 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 in non-RA subjects. The objectives of this study were: (1) to compare LDL levels in RA cases compared with 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 an RA case cohort and a non-RA control cohort. We hypothesise 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 or direct LDL measurements in the electronic medical records (EMR) of Brigham and Women’s Hospital and Massachusetts General Hospital from 1989 to 2007. Briefly, RA cases were identified using an RA phenotype algorithm which utilises 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 for RA using the algorithm is 94%. For details on the development and validation of this cohort, please refer to Liao et al.\(^ {17}\)

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.\(^ {19}\). Briefly, non-rheumatic disease subjects were selected based on similar age, gender, race and healthcare utilisation to the RA cases. Healthcare utilisation was approximated by using the number of ‘facts’ which are points of contact with the healthcare system—that is, laboratory blood draws, clinic visits and x-rays. For details on the development of the non-RA control cohort and sample collection, please refer to Kurreeman et al.\(^ {19}\). We collected discarded blood samples for each cohort using the Brigham and Women’s Hospital biospecimen repository.

**Variables**

The primary outcome, LDL, was defined by each subject’s first LDL measurement in the EMR, to maximise 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, high density lipoprotein) measured within 1 year of the index LDL. Age at LDL measurement, gender and the year of LDL measurement in the EMR, to maximise the chance of detecting an association with LDL levels.\(^ {24,25}\)

**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.\(^ {20}\) We excluded individuals with call rate <97% and SNPs with missingness >0.02 or departure from Hardy–Weinberg equilibrium (\(p_{\text{HWE}}<0.001\)). We also computed a \(\chi^2\) test to assess the difference in missingness between RA cases and non-RA controls and removed SNPs with \(p_{\text{missing}}<10^{-2}\).

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, We calculated pairwise identity by state statistics using PLINK,\(^ {22}\) and removed one individual from each pair of individuals who were second degree or closer relatives. Principal components analysis 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.

**Genetic risk scores**

We constructed three separate aggregate GRS for LDL, non-HLA RA risk alleles\(^ {24}\) and the RA region\(^ {16}\) to test for association with LDL levels. We refer to these GRS as the LDL GRS, RA GRS and HLA GRS, respectively. Individual SNPs have modest effect sizes and would have limited power to demonstrate an association with LDL levels. GRS allow for testing of SNPs in aggregate (grouped by their association with clearly defined phenotypes, ie, 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 (one test).

The weighted LDL GRS was constructed using 11 SNPs associated with LDL as the lead trait from published studies\(^ {7,23}\) and weighted by published effect sizes for higher levels of LDL (mg/dl) (see online supplementary 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 with non-HLA RA risk loci,\(^ {16}\) 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 OR for risk of RA from the most recent meta-analysis\(^ {20}\) (see online supplementary 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.\(^ {16}\) weighted by the natural log of the published effect sizes for risk of RA.

All three GRSs were constructed using the general formula:

\[
GRS = \sum_{i} w_i X_i
\]

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

**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, total cholesterol and high density lipoprotein levels across prevalent RA and non-RA controls. We applied t tests to compare means and χ² 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,

Our two 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 with 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 levels in RA cases compared with 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 utilised the interaction tests detailed above to determine significant differences in the association between the RA GRS and LDL levels in RA cases compared with non-RA controls.

If the associations of the GRS 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 visualise the trends in LDL levels associated with increasing numbers of LDL and RA genetic alleles, we first determined cut-offs for tertiles of the RA GRS and LDL GRS separately in RA cases and non-RA controls.

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

The study was approved by the Partners Healthcare institutional review board for Brigham and Women’s Hospital and Massachusetts General Hospital. Statistical analyses were conducted using SAS V9.2 (Cary, North Carolina, USA).

**RESULTS**

**LDL levels in RA cases versus non-RA controls**
We identified 567 subjects with prevalent RA and 979 non-RA controls with available LDL measurements spanning the years 1989 to 2007 (table 1). All subjects were of European ancestry and had genotype data available. Mean ages at first LDL measurement were similar in the two groups: 54.5 years 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 that 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, 5 mg/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 with ACPA negative (n=145) subjects, adjusted by age, gender and year of LDL measurement (p=0.94).

**Association of LDL GRS with LDL levels**
We next tested whether an LDL 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 0.72 mg/dl higher LDL level (p=6.2×10⁻⁶). Among controls, each unit increase in the LDL GRS was associated with a 0.85 mg/dl

<table>
<thead>
<tr>
<th>Clinical characteristic</th>
<th>RA cases (n=567)</th>
<th>Non-RA controls (n=979)</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age LDL measured (years)</td>
<td>54.5 (12.3)</td>
<td>55.2 (12.5)</td>
<td>0.29</td>
</tr>
<tr>
<td>Female (n (%))</td>
<td>456 (80.4)</td>
<td>777 (79.4)</td>
<td>0.65</td>
</tr>
<tr>
<td>Mean year LDL measurement (SD)</td>
<td>2000 (4.9)</td>
<td>1998 (5.2)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>ACPA status (n (%))</td>
<td>422 (74.4)</td>
<td>1 (0.6)*</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Lipoprotein levels (mg/dl)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LDL (mean (SD))</td>
<td>117.2 (36.5)</td>
<td>125.6 (41.7)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Tchol, (mean (SD))</td>
<td>199.3 (43.5)</td>
<td>208.3 (49.9)</td>
<td>0.0002</td>
</tr>
<tr>
<td>HDL (mean (SD))</td>
<td>56.1 (18.1)</td>
<td>53.8 (17.8)</td>
<td>0.01</td>
</tr>
</tbody>
</table>

*Out of 156 controls with ACPA data.

ACPAN, anticitrullinated peptide/protein antibodies; HDL, high density lipoprotein; LDL, low density lipoprotein; RA, rheumatoid arthritis; Tchol, total cholesterol.

higher LDL level ($p=9.4\times10^{-8}$) (table 2). The association between the LDL GRS and LDL levels was not significantly different in RA cases and controls (interaction test, $p=0.45$).

As 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 with 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, 4.3 mg/dl lower LDL levels than non-RA controls ($p=0.03$) (table 3).

### Association of an RA GRS and an HLA GRS 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 could 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 five amino acids in three HLA genes.16 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 4.6 mg/dl lower LDL levels ($p=0.007$) among RA cases and a 4.3 mg/dl lower LDL levels in controls ($p=0.02$) (table 2). The association between the RA GRS and LDL levels was not significantly different among RA cases and controls (interaction test, $p=0.17$). 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 ($p<0.001$) (see online supplementary 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.

As 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 with controls, controlling for the RA GRS as well as age, gender and year of LDL measurement. In this model, given a similar RA GRS, RA cases and controls had similar LDL levels ($p=0.30$) (table 3). As we observed no difference between the associations of the LDL GRS or RA GRS and LDL levels in RA cases compared with 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 ($p=9.3\times10^{-7}$) in RA cases and controls (table 4). The lack of interaction between the RA GRS and LDL GRS

### Table 2  Association between the low density lipoprotein genetic risk score and low density lipoprotein levels, and the rheumatoid arthritis genetic risk score with low density lipoprotein levels, among rheumatoid arthritis cases and non-rheumatoid arthritis controls, adjusted by age, gender and year of low density lipoprotein measurement

<table>
<thead>
<tr>
<th>Variables</th>
<th>Association of LDL GRS with LDL levels</th>
<th>Association of RA GRS with LDL levels</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$\beta$ coefficient (SE) $p$ Value</td>
<td>$\beta$ coefficient (SE) $p$ Value</td>
</tr>
<tr>
<td>Age</td>
<td>0.34 (0.08) 1.4x10^{-5}</td>
<td>0.34 (0.08) 1.9x10^{-5}</td>
</tr>
<tr>
<td>Female gender</td>
<td>4.90 (2.41) 0.04</td>
<td>6.23 (2.44) 0.01</td>
</tr>
<tr>
<td>Year of LDL measurement</td>
<td>-2.55 (0.19) 8.6x10^{-19}</td>
<td>-2.60 (0.19) 1.5x10^{-19}</td>
</tr>
<tr>
<td>RA case×GRS</td>
<td>(LDL GRS) 0.72 (0.16) 6.2x10^{-6}</td>
<td>(RA GRS) -4.58 (1.71) 0.007</td>
</tr>
<tr>
<td>Control×GRS</td>
<td>(LDL GRS) 0.85 (0.16) 9.4x10^{-8}</td>
<td>(RA GRS) -4.26 (1.79) 0.02</td>
</tr>
<tr>
<td>Interaction tests</td>
<td>No significant difference in association between LDL GRS and LDL levels in RA cases and non-RA controls, $p=0.45$</td>
<td>No significant difference in association between RA GRS and LDL levels in RA cases and non-RA controls, $p=0.17$</td>
</tr>
<tr>
<td>Note</td>
<td>RA cases with LDL GRS data, n=542</td>
<td>RA cases with RA GRS data, n=541</td>
</tr>
<tr>
<td></td>
<td>Non-RA controls with LDL GRS data, n=963</td>
<td>Non-RA controls with RA GRS data, n=945</td>
</tr>
</tbody>
</table>

GRS, genetic risk score; LDL, low density lipoprotein; RA, rheumatoid arthritis.
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 ($p=1.93 \times 10^{-20}$). As 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 GRAs. RA risk alleles identified mainly though GWAS are common alleles, and were relatively common in the non-RA control group (see online supplementary 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 $^2$ $^29$ 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 healthcare system, the system is not closed and incomplete capture of patient information (ie, 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 was not a random sample of the general population. They comprised individuals who utilised the healthcare system in a manner similar to RA patients, without a rheumatic disease but had 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

### Table 4 Association between rheumatoid arthritis genetic risk score, low density lipoprotein genetic risk score and low density lipoprotein levels in a multivariable model adjusted by age, gender and year of low density lipoprotein measurement in rheumatoid arthritis cases and non- rheumatoid arthritis controls

<table>
<thead>
<tr>
<th>Variable</th>
<th>$\beta$ coefficient (SE)</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>RA case status</td>
<td>$-3.25 (2.14)$</td>
<td>0.13</td>
</tr>
<tr>
<td>RA GRS</td>
<td>$-4.68 (1.76)$</td>
<td>0.008</td>
</tr>
<tr>
<td>LDL GRS</td>
<td>$0.78 (0.16)$</td>
<td>$9.31 \times 10^{-7}$</td>
</tr>
</tbody>
</table>

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

GRS, genetic risk score; LDL, low density lipoprotein; RA, rheumatoid arthritis.

### DISCUSSION

There are three main findings from our study. First, we observed that LDL levels were lower in RA cases compared with non-RA controls, adding to the few published epidemiological studies on this topic. $^1$ $^2$ Second, our findings replicate previous studies that LDL alleles in aggregate are associated with lower LDL levels among non-RA controls, 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 in non-RA controls. By definition, RA

Figure 1 Mean levels of low density lipoprotein (LDL) among rheumatoid arthritis (RA) cases grouped by tertiles of the LDL and RA genetic risk scores (GRS) for (A) RA cases and (B) non-RA controls. (Note: no interaction observed between the RA GRS and LDL GRS.)
and healthcare utilisation 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. 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 analysed several cohorts with data spanning five decades), 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 but the significance and magnitude of these associations are conflicting. 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, as 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.

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Acknowledgements

We would like to acknowledge Gina Peloso, PhD, and Sekar Kathiresan, MD, both at the Center for Human Genetic Research, Massachusetts General Hospital and the Broad Institute, for providing their expertise on the genetics of LDL traits in the general population during the planning and analyses stages of this project.

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Funding

The Informatics for Integrating Biology and the Bedside project is funded by the NIH grant US4-LM008748. KPL is supported by the NIH K08 AR060257 and the Harold and Dorothy Browne Fund; SR by the NIH K08 AR055688, U01HG007033 and the Arthritis Foundation; and EWK by the NIH K24 AR052403, R01-AR049880 and P60-AR047782, RMP NIH R01-AR57108, R01-AR056768, U01-GM092691, R01-AR059648 and the Career Award for Medical Scientists from the Burroughs Welcome Fund.

Competing interests

None.

Ethics approval

The study was approved by the Partners Healthcare institutional review board for Brigham and Women’s Hospital and Massachusetts General Hospital.

Provenance and peer review

Not commissioned; externally peer reviewed.

REFERENCES


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Katherine P Liao, Dorothée Diogo, Jing Cui, et al.

Ann Rheum Dis published online May 28, 2013
doi: 10.1136/annrheumdis-2012-203202

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