GENETIC INFLUENCES ON THE RELATIONSHIP

BETWEEN MEAT CONSUMPTION & LDL-C LEVELS

By

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ABSTRACT

Meat consumption is known to be related to increased concentrations of low density lipoprotein cholesterol (LDL-C), which is directly related to several cardiometabolic diseases. Though some clinical trials have elucidated how meat consumption disproportionately impacts blood biomarker levels in some groups more than others, little is known regarding how genetic factors might modify this association. In this study, a genetic risk score (GRS) comprised of SNPs known to be related to an increased genetic risk of high LDL-C levels was calculated for 402,434 participants in the UK Biobank and tested for interactions with both red meat and processed meat consumption, as defined by responses to a food frequency questionnaire. Red meat consumption was found to have a significant interaction with the GRS (β=0.01, p=0.011). In particular, those with a higher LDL-C GRS were more likely to consume red meat, though all groups displayed a positive association with red meat consumption. As such, across all levels of genetic risk, lower red meat consumption was found to be consistently associated with a reduced risk of high LDL-C levels, consistent with the recommendations of nutritional guidelines.

INTRODUCTION

Meat consumption has long been regarded as a risk factor for a wide range of metabolic disorders, including several cardiovascular diseases, which account for the top 5 leading causes of death globally.[1] This association, however, has recently been contested as varying studies present different perspectives on whether meat consumption is in fact detrimental to cardiovascular health.[2,3] Recent meta-analyses have concluded that meat consumption, especially of processed and red meats, is a significant risk factor for coronary heart disease (CHD) and type 2 diabetes.[4] Meanwhile, a recent systematic review concluded that red meat consumption has no particularly detrimental effects for human health and subsequently recommended meat consumption not be decreased in the general population, claiming that this dietary practice is associated with minimal risk of the aforementioned cardiovascular detriments.[5]

The observational nature of the studies analyzed in these reviews has often been blamed for the disparities in guidelines and recommendations surrounding meat consumption. Many of these studies have focused on cancer incidence associated with red and processed meat consumption over time, but few have sought to examine the relationship between meat consumption and related biomarkers predictive of cardiometabolic diseases.

Meat's inherently high level of saturated fatty acid content has been shown to impact low density lipoprotein cholesterol (LDL-C) levels.[6] Based on the results of a recent randomized controlled trial, those consuming red meat heavy diets (as opposed to those consuming nonmeat and white meat diets) have been shown to present higher levels of LDL-C cholesterol due to high saturated fatty acid levels in this diet compared to plant-based protein diets.[12] High levels of LDL-C and high intake of saturated fatty acids are known to be important biomarker risk factors influencing the risk of cardiovascular diseases.[7,8] Increased levels of LDL-C, in particular, has been consistently correlated with an elevated risk for CHD.[9,10] LDL-C has also been studied in a genetic context through Mendelian Randomization, establishing causal relationships between genetic predisposition towards higher LDL-C levels and subsequent CHD.[11]

The strength of the association between LDL-C levels and poor cardiovascular outcomes such as CHD thus confirms the potential for use of the LDL-C biomarker as a proxy for cardiometabolic disease risk. The question remains, however: does meat consumption impact cardiometabolic disease risk? A *genetic* analysis of this question seeks to determine whether the association of meat consumption with LDL-C levels differs in varying subgroups based on genetic predispositions for high LDL-C levels.

Using serum LDL-C levels and genetic risk for higher lipoprotein levels as an indicator of cardiometabolic diseases is a useful tool in establishing the relationship between meat consumption and cardiometabolic disease outcomes. To this end, this thesis aims to illuminate the associations and interactions of genetic risk for increased LDL-C levels and meat

consumption in the UK Biobank, a large prospective cohort study comprising a vast amount of genetic and phenotypic data.

METHODS

UK Biobank Data

The UK Biobank is a large prospective cohort study of 502,549 participants with a large array of genetic and phenotypic data. The data stored in the UK Biobank includes, but is not limited to, demographic, blood/urine/saliva sample, questionnaire, and genomic data collected between 2006 and 2010 through several assessment centers in 22 locations across the UK. This study analyzed the data of 488,377 UK Biobank participants between the ages of 37 and 73 with predominantly European ancestry. The UK Biobank's collection strategies and procedures have been described in greater detail previously.[13] Basic quality control procedures were performed to exclude individuals (i.e. those reporting non-European ancestry) and some SNPs, as is cited in genotyping methods described previously.[14] Participants taking cholesterol-lowering medications were removed from this sample to yield a total of 402,434 individuals whose genetic risk score (described in detail later) was derived.

Meat Consumption Variables

These variables were defined from dietary data collected in a self-reported food frequency questionnaire (FFQ). Frequency of weekly beef, pork and lamb intake (excluding processed meat) and frequency of weekly processed meat intake were asked on an online questionnaire completed by participants at the assessment centers. These two variables were coded as total red meat (TRM) consumption and processed meat (PM) consumption. Beef, pork, and lamb were all considered red meats for this analysis.[15] Each of the three meats were coded as a continuous variable into 6 groups ranging from consumption of "less than once per week" to "once or more daily". The TRM variable was then created as a summation of the individual beef, pork, and lamb variables. The PM variable was coded categorically as a dichotomous variable where those consuming processed meat less than once a week were coded as "0" and all others as "1."[16] Participants with missing data for these variables were excluded from analyses.

LDL-C Variable

The UK Biobank measured LDL-C levels directly in mmol/mL through a serum test of all participants from blood collected in a non-fasting state at the baseline measurement. This variable was inverse-normalized throughout all analyses to conform to assumptions of linear regression.

Development of a LDL-C Genetic Risk Score

To derive the LDL-C genetic risk score (GRS), 37 SNPs known to be associated with LDL-C levels were used.[17] For each subject, a GRS was derived by summing the total number of risk alleles, each weighted by their beta coefficient from the association of each SNP with LDL-C levels. This resulted in a GRS ranging from 0.903 to 3.32, that was assigned to all participants.

Statistical Analysis

A genetic risk score was created and analyzed for interactions with both TRM and PM consumption using multiple linear regression, with covariates of BMI, sex, and age. This interaction was tested by including the product of the GRS with meat consumption as a covariate in the linear regression. Following this, individuals were divided into tertiles of LDL-C genetic risk. Within these tertiles, the association of TRM and PM consumption with LDL-C was tested with the inclusion of BMI, sex, and age as covariates. All analyses were conducted using R version 3.6.1.

RESULTS

The descriptive characteristics of the sample used in this analysis are described in Table 1. Division of the sample into tertiles did not result in any apparent significant differences between participants in any of the 3 groups.

We found that, phenotypically, both red and processed meat consumption are positively associated with LDL-C levels in the entire sample (β= 0.02 (p=1.5E-101); β= 0.005 (p=0.05)). The primary analysis also demonstrated a consistent and positive association between LDL-C and the LDL-C GRS, confirming that the GRS, though created in a different population sample, consistently predicted LDL-C levels in UK Biobank participants.

Table 1. Descriptive Characteristics of the Sample, Grouped by GRS Tertile

	Low LDL-C GRS *(n= 132595)*	**Medium LDL-C GRS** *(n= 132669)*	**High LDL-C GRS** *(n= 132519)*
Mean Age	55.7 (8.1)	55.6 (8.07)	55.3 (8.06)
Sex	75,141 female 57,454 male	76,338 female 56,331 male	77,226 female 55,293 male
Townsend Deprivation Index[a]	-1.320 (3.1)	-1.403 (3.0)	-1.464 (3.0)
BMI	27.17 (4.7)	27.0 (4.6)	26.86 (4.6)
Serum LDL-C	3.473 (0.8)	3.713 (0.8)	3.950 (0.8)
TRM[b]	2.117 (1.5)	2.087 (1.4)	2.068 (1.4)
PM	91,777=0 40,509=1	92,394=0 40,026=1	92,385=0 39,900=1

[a]The Townsend Deprivation Index is a measure of material deprivation. Higher values indicate increased material deprivation.[18]

[b]Average reported level of red meat intake per week.

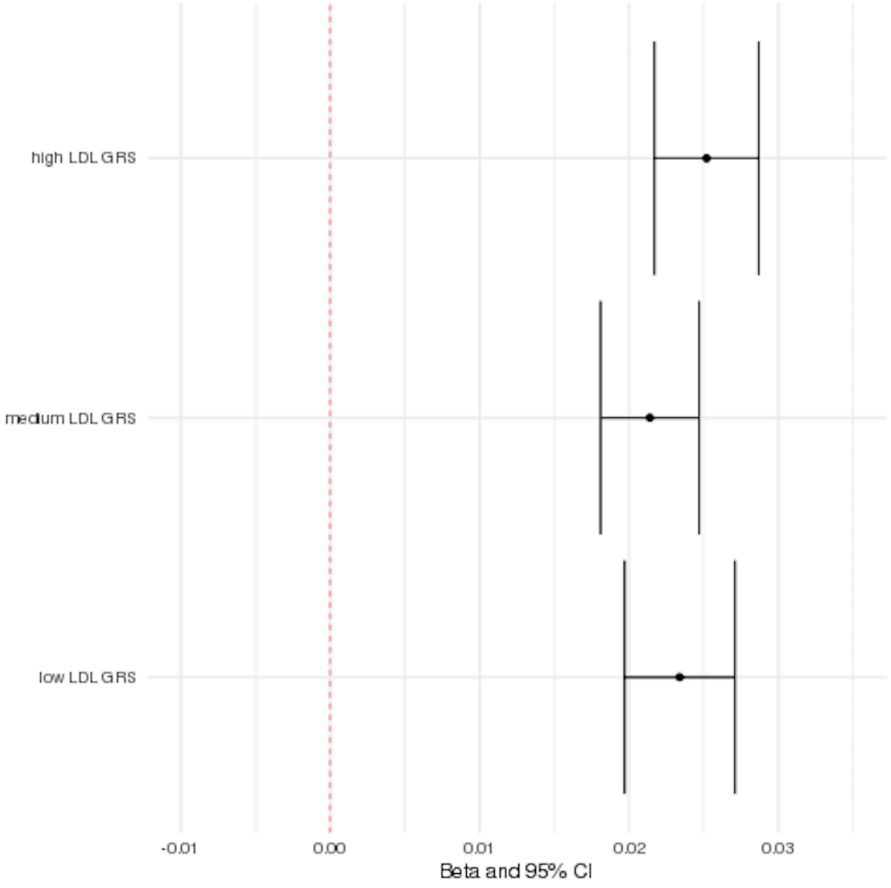
Interaction of LDL-C GRS with TRM & PM

We found a significant interaction between LDL-C GRS and TRM consumption (β=0.01, p=0.011), but not with PM consumption (p=0.879). The sample was divided into tertiles of LDL-C GRS to understand the differential association of meat consumption with LDL-C on individuals of varying levels of genetic risk. Participants were distributed among those with GRSs between 0.89 and 2.00, 2.01 and 2.21, and 2.22 and 3.15. **Table 2** describes the results of the linear regression within each GRS tertile. We found a weaker association of meat with LDL-C among people with lower LDL-C GRS. Across all levels of genetic risk, red meat consumption showed a significant association with lowered risk for high LDL-C levels. **Figure 1** depicts these results in a forest plot.

Table 2. Association of TRM with LDL-C, Grouped by GRS Tertile

GRS Tertile	Estimate	Std. Error	t-value	p-value	95% CI
Low	2.14E-02	1.68E-03	12.748	3.35E-37 *	(0.018, 0.025)
Medium	2.52E-02	1.79E-03	14.08	5.45E-45 *	(0.022, 0.029)
High	2.34E-02	1.87E-03	12.517	6.37E-36 *	(0.020, 0.027)

Figure 1. Forest Plot of Associations of Total Red Meat Consumption with LDL-C, Grouped by GRS Tertile



DISCUSSION

This study sought to understand the effects, if any, of meat consumption on different groups of people separated by genetic risk for high LDL-C levels. Self-reported consumption of both red and processed meats were found to be positively associated with LDL-C levels. The results of the GRS analysis indicate the positive and significant association of TRM consumption with LDL-C levels across all categories of genetic predisposition to higher LDL-C levels. Lower meat consumption is consistently associated with a reduced risk of high LDL-C levels in all groups of genetic risk.

These findings correspond with the positive relationship between LDL-C and red meat intake. In two meta-analyses of different diets and their associations with several blood biomarkers, individuals consuming meat-based diets were found to have higher levels of LDL than diets that substituted red meat for nuts and legumes.[19,20] The previously mentioned RCT found LDL-C levels to be higher in those consuming red meat and white meat diets than nonmeat sources, even regardless of the saturated fatty acid content of the meats.[12] A systematic review seeking to understand diets most likely to prevent CHD purported that substitution of red meat for other types of protein sources reduced CHD risk.[21] A meta-analysis of the effects of soy protein intakes on serum lipid concentrations found that substitution of animal protein with soy protein significantly reduces LDL-C levels, as well.[22]

The study also found that meat consumption had a greater association for those in the sample who had a higher LDL-C GRS. Given this result, it is likely that those with a higher genetic predisposition to high LDL-C levels be more cautious than others in controlling their meat intake. Since such an analysis has not, to our knowledge, been studied in in-person trials or interventions, future experimental studies would be needed to understand this relationship further.

The strengths of this study include the large sample size of the use of directly measured serum LDL-C levels. The use of a LDL-C GRS that was developed in a different dataset and then applied to a UKB sample confers the fidelity of this instrument in accurately predicting the genetic risk of the sample. While the above confer strengths to this study, a cross-sectional study design and lack of representative meat phenotype serve as limitations of the analysis. In particular, reliance on participants reporting of "typical" meat intake in a week may incur memory bias and provides room for inaccurate self-reporting of diet, a known limitation of the food frequency questionnaire.

In conclusion, this analysis suggests the maintenance of current reccomendations for moderate meat consumption for improved cardiovascular health in the general population, and perhaps modification to suggest those at high risk for LDL-C levels limit their red meat consumption more than others. Future directions of this study may include the addition of more LDL-C SNPs into the development of the genetic risk score, analysis of the GRS concurrently with a dietary risk score, and use of a refined meat consumption phenotype that more accurately reflects quantities of meat consumed by participants.

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