ORIGINAL RESEARCH

CURRENT DEVELOPMENTS IN NUTRITION

Nutritional Biochemistry and Physiologic Mechanisms

Elevated LDL Cholesterol with a Carbohydrate-Restricted Diet: Evidence for a "Lean Mass Hyper-Responder" Phenotype

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ABSTRACT

Background: People commencing a carbohydrate-restricted diet (CRD) experience markedly heterogenous responses in LDL cholesterol, ranging from extreme elevations to reductions.

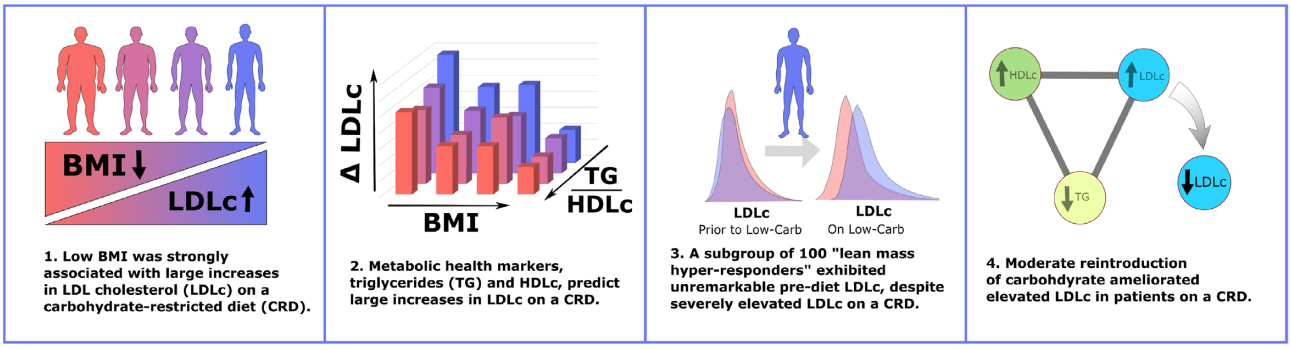
Objectives: The aim was to elucidate possible sources of heterogeneity in LDL cholesterol response to a CRD and thereby identify individuals who may be at risk for LDL cholesterol elevation.

Methods: Hypothesis-naive analyses were conducted on web survey data from 548 adults consuming a CRD. Univariate and multivariate regression models and regression trees were built to evaluate the interaction between body mass index (BMI) and baseline lipid markers. Data were also collected from a case series of five clinical patients with extremely high LDL cholesterol consuming a CRD.

Results: BMI was inversely associated with LDL cholesterol change. Low triglyceride (TG) to HDL cholesterol ratio, a marker of good metabolic health, predicted larger LDL cholesterol increases. A subgroup of respondents with LDL cholesterol ≥200 mg/dL, HDL cholesterol ≥80 mg/dL, and TG ≤70 mg/dL were characterized as "lean mass hyper-responders." Respondents with this phenotype ($n = 100$) had a lower BMI and, remarkably, similar prior LDL cholesterol versus other respondents. In the case series, moderate reintroduction of carbohydrate produced a marked decrease in LDL cholesterol.

Conclusions: These data suggest that, in contrast to the typical pattern of dyslipidemia, greater LDL cholesterol elevation on a CRD tends to occur in the context of otherwise low cardiometabolic risk. *Curr Dev Nutr* 2022;6:nzab144.

GRAPHICAL ABSTRACT



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Supplemental Survey, Supplemental Tables 1–3, Supplemental Figures 1–5, and Supplemental Case Series are available from the "Supplementary data" link in the online posting of the article and from the same link in the online table of contents at https://academic.oup.com/cdn/.

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Abbreviations used: AIC, Akaike Information Criterion; ASCVD, atherosclerotic cardiovascular disease; CRD, carbohydrate-restricted diet; LCD, low-carbohydrate diet; LMHR, lean mass hyper-responder; TG, triglyceride; VLCD, very-low-carbohydrate diet.

Introduction

Carbohydrate-restricted diets (CRDs) hold promise for weight loss, type 2 diabetes, and other chronic health conditions, but this dietary strategy may cause elevated LDL cholesterol, an important risk factor for atherosclerotic cardiovascular disease (ASCVD). Some studies report marked increases in LDL cholesterol with consumption of a CRD (1–4); however, others show no clinically meaningful increases (5–12). The sources and mechanistic basis of heterogeneity in response to carbohydrate restriction among studies and among individuals are poorly characterized, limiting translation of this dietary strategy to public health and patient care.

Plausible sources of heterogeneity include ratio of saturated to unsaturated fatty acid content, genetic susceptibility (13), and degree of carbohydrate restriction [differentiating between a moderately restrictive low-carbohydrate diet (LCD; 50 to 130 grams/day available carbohydrate) and a more intensively restrictive very-low-carbohydrate diet (VLCD; <50 grams/day)]. A novel source of interindividual heterogeneity may relate to cardiometabolic health measures.

Many studies of CRD involve participants with obesity (6–8, 10–12), metabolic syndrome (8), or type 2 diabetes (14), conditions associated with adverse metabolic health markers related to insulin resistance, notably including low HDL cholesterol and high triglycerides (TG) (15). Among these participants, relatively minor LDL cholesterol elevations have been observed. For example, in a nonrandomized study including 262 patients with type 2 diabetes consuming a VLCD, LDL cholesterol increased by a mean of only 11 mg/dL after 2 years (14). Similarly, in a randomized crossover trial including participants with obesity and metabolic syndrome, isocaloric substitution of fat for carbohydrate did not raise LDL cholesterol despite a 2-fold increase in saturated fat intake (8).

By contrast, among lean and metabolically healthy participants, marked elevations in LDL cholesterol have been reported. In an observational study of 20 ultra-endurance runners, those habitually consuming a 10% compared with a 57% carbohydrate diet had substantially higher LDL cholesterol (161 vs. 88 mg/dL, respectively; $P < 0.001$), with striking consistency within groups (1). In a 4-week crossover feeding study of 17 lean healthy young women, a VLCD increased LDL cholesterol by 70 mg/dL compared with a standard diet. Notably, all participants exhibited an increased LDL cholesterol on the VLCD (2).

Based on anecdotal reports in social media communities of CRD consumers, one coauthor (DF) proposed in 2017 the existence of a "lean mass hyper-responder" (LMHR) phenotype defined as high LDL cholesterol (≥ 200 mg/dL) with consumption of a CRD in association with high HDL cholesterol (≥ 80 mg/dL) and low TGs (≤ 70 mg/dL) (16). This lipid pattern differs from that commonly seen in obesity, in which mild elevations and preponderance of small dense LDL cholesterol is associated with other adverse lipid concentrations, including atherogenic dyslipidemia (low HDL cholesterol and high TGs) (15, 17). To test the hypotheses that LDL cholesterol elevation is associated with both leanness (as measured by BMI) and metabolic health (as evidenced by a low ratio of TG to HDL cholesterol) we analyzed a survey of adults consuming a CRD. In addition, we explored in a case series whether a moderate increase in carbohydrate intake, within the context of a CRD, might ameliorate LDL cholesterol elevation.

Methods

Ethics statement

Respondents to the web survey were informed that their anonymized data may be used in scientific reports. Patients in the case series provided written informed consent for the publication of the data included herein. Review of existing anonymized aggregate data, previously collected by survey, and case reports, for the purpose of this study, was determined to not constitute human subject research by the Institutional Review Board at Boston Children's Hospital.

Web survey

The "Cholesterol Super Survey" is a publicly available ongoing questionnaire created by a coauthor (DF) in January 2020 with the aim of describing changes in LDL cholesterol among consumers of a CRD. The survey, advertised through social media, includes questions about height, weight, dietary intake, medications, and current and past lipid test results. A copy of this survey is available in the **Supplemental Survey** and responses used in this manuscript were collected between 16 January and 30 November 2020.

A priori inclusion and exclusion criteria

Inclusion criteria were as follows:

- Current consumption of a CRD, with ≤130 grams/day available carbohydrate (i.e., excluding dietary fiber)
- Not taking lipid-lowering medication (e.g., statins, ezetimibe, PCSK9i, fibrates)
- Most recent lipid data ("current") on CRD provided, included LDL cholesterol, HDL cholesterol, and TG, as well as lipid data from prior to current CRD
- Lipid test on CRD obtained in 2018 or later; current and prior lipid tests obtained within 5 years (1825 days) of each other
- Current and prior lipid tests obtained after a fast of 12–16 hours

Exclusion criteria were as follows:

- Respondents with reported ages <18 or >100 years
- Respondents reporting potentially unreliable data: BMI (in kg/m^2) <10 or >50, LDL cholesterol <30 or >1000 mg/dL, HDL cholesterol <20 or >200 mg/dL, TG <20 or >1500 mg/dL

Descriptive statistics

Data management and statistical analyses were performed using R version 4.0.3 and the packages: *tidyverse, readxl, performance, gtools, MASS, bootStepAIC, lmtest*, and *car*. We filtered data using the inclusion/exclusion criteria described above, to obtain our final sample of $n = 548$ (after excluding 355). Descriptive statistics were obtained using *tableone::CreateContTable | CreateCatTable* and quantile values were obtained using *stats::quantile* for the 5th, 25th, 50th, 75th and 95th percentiles. Data were tested for normality using *stats::shapiro.test*.

Exploratory analyses and regression models

To identify factors associated with LDL cholesterol changes, we first ran a linear model with all lipid and anthropometric factors in our dataset other than current LDL cholesterol. Subsequently, we evaluated the relevance of potential non–LDL cholesterol factors associated with LDL

cholesterol change by analyzing their contribution to the explanatory capacity of the model [Akaike Information Criterion (AIC)], the consistency of their coefficient signs, and the consistency of their statistical relevance. This procedure was performed via a bootstrap AIC consistency diagnosis in which 100 independent samples were drawn at random from the dataset using *bootStepAIC::boot.stepAIC*.

We next built univariate and multivariate linear regression models to further probe for relations among lipid markers, BMI, and increases in LDL cholesterol with a CRD. Models were constructed using *stats::lm* and *stats::glm* and "binomial" as family identity if the independent variable was dichotomous. To compare the all-around performance of different models for predicting LDL cholesterol change on a CRD, we used *performance::compare_performance*. To evaluate linear assumptions of the models we used *performance::check_model*. This can be graphically evaluated in the publicly available code.

To illustrate the relation among TG/HDL cholesterol, BMI, and LDL cholesterol change as a 3-dimensional bar graph, TG/HDL cholesterol and BMI data were partitioned into quartiles using *gtools::quantcut*.

In a complementary approach for finding potential predictors of LDL cholesterol change on a CRD without using any preconceived definitions, we performed a regression tree using *rpart::rpart* with method = "anova". This method uses as splitting criteria $SS_T - (SS_L + SS_R)$, where $SS_T = \Sigma\ (y_i - y)^2$ is the sum of squares for the node and SS_R, SS_L are the sums of squares for the right and left, respectively. In other words, it chooses the split that maximizes the between-groups sum-of-squares in a simple ANOVA and does not consider any pre-established cutoff points (18). BMI, sex, age, and all prior non–LDL cholesterol lipid markers were included, and the algorithm selected the relevant variables.

Characterizing the LMHR phenotype

To characterize the LMHR phenotype, we used previously proposed criteria (16): LDL cholesterol ≥200 mg/dL, HDL cholesterol ≥80 mg/dL, and TG ≤70 mg/dL. For comparison of LMHR to non-LMHR respondents' prior lipid data, the statistical significance threshold was established at 0.05 and evaluated by Mann-Whitney *U* test. Comparison BMI plots, including distribution curves and violin-box plots, were created using *ggplot2* and *ggstatsplot* with default settings for producing the density plots.

To compare data collected in our survey against a population reference, the NHANES IV dataset, we used *RNHANES::nhanes_load_data* for datasets *"TRIGLY_G,"* *"HDL_G,"* and *BMX_G,"* and DEMO_G" for *"2011–2012"*. *BMX_G"* and DEMO_G" were subsequently merged by SEQN (individual identifier) and filtered by age ≥18 years to exclude children.

Sample size and statistical power assessment

Due to the descriptive nature of our study, no outcome-based sample size was established a priori. However, we estimated that our sample size is large enough to detect f^2 differences as small as 0.02 with an α of 0.05 and a statistical power of 0.8 in linear multiple regressions with as a many as 3 predictors; and to detect R^2 as low as 0.1 with ORs >2.0 assuming the same statistical error parameters. Nonetheless, interaction terms were avoided and no more than 3 predictors were evaluated in each linear model. Statistical power and sample size assessments were performed using G*Power version 3.1.9.4 (open access software by the University of Dusseldorf).

Raw data and analysis code availability

The anonymized raw data from the web survey and the step-by-step commented code for reproducing both quantitative and graphical analyses and are publicly available at: https://github.com/AdrianSotoM/LMHR.

Case series

Patients presenting to the clinic of a coauthor (TK) with a history of elevated LDL cholesterol following initiation of a VLCD containing <25 grams/day carbohydrate and no personal history of myocardial infarction or stroke were initially counselled on standard-of-care pharmacologic options to lower LDL cholesterol, including statins. Patients included in this series refused pharmacotherapy and instead opted to pursue an empiric clinician-supervised dietary therapy, with reintroduction of 50–100 grams carbohydrate/day in the form of fruits or starchy vegetables. Body fat percentage was measured by a Valhalla Scientific BCS Elite 4-point bioimpedance scale.

Results

Survey respondents reported markedly elevated LDL cholesterol and optimal TG/HDL cholesterol ratio

Among 903 respondents, 23 reported available carbohydrate intake >130 grams/day, indicating that the sample was primarily composed of those consuming an LCD or VLCD. A further 332 were excluded as described in the Methods, yielding 548 for analysis (**Figure 1**).

Table 1 presents quantile distributions for the included sample. The mean age was 51 ± 12 years, with 90% of respondents between 31 and 69 years, and 58% male. Mean BMI was 24.1 ± 4 and mean available carbohydrate intake was 27 grams/day. Mean current LDL cholesterol on a CRD was 236 ± 107 mg/dL, whereas prior LDL cholesterol was 145 ± 59 mg/dL. Mean current TG/HDL cholesterol was 1.1, an optimal level (19, 20), consistent with the expected effects of a CRD (9). Mean time between lipid tests was 724 days.

Leanness and metabolic health are associated with large LDL cholesterol increase on a CRD

In a hypothesis-naive multiple linear regression model using a bootstrap AIC consistency diagnosis (**Supplemental Table 1**), low BMI was consistently (100% of random samples) associated with LDL cholesterol increase on a CRD ($\beta = -4.5$, $P = 1.0 \times 10^{-4}$).

Table 2 shows the results of linear models of baseline metabolic health markers and LDL cholesterol change on the diet. Low TG ($\beta = -0.17$, $P = 0.015$) and high HDL cholesterol ($\beta = 0.6$, $P = 0.007$) predicted greater increases in LDL cholesterol (model 1). TG to HDL cholesterol ratio was also a strong and highly significant predictor ($\beta = -9.9$, $P = 1.1 \times 10^{-4}$) (model 2), although this relation was attenuated with inclusion of current BMI (model 3).

To explore this finding, we examined the ability of prior lipids to predict current BMI. Whereas both prior TG ($\beta = 0.01$, $P = 2.3 \times 10^{-5}$) and HDL cholesterol ($\beta = -0.06$, $P = 6.9 \times 10^{-12}$) were strongly associated in the expected directions, the association with LDL

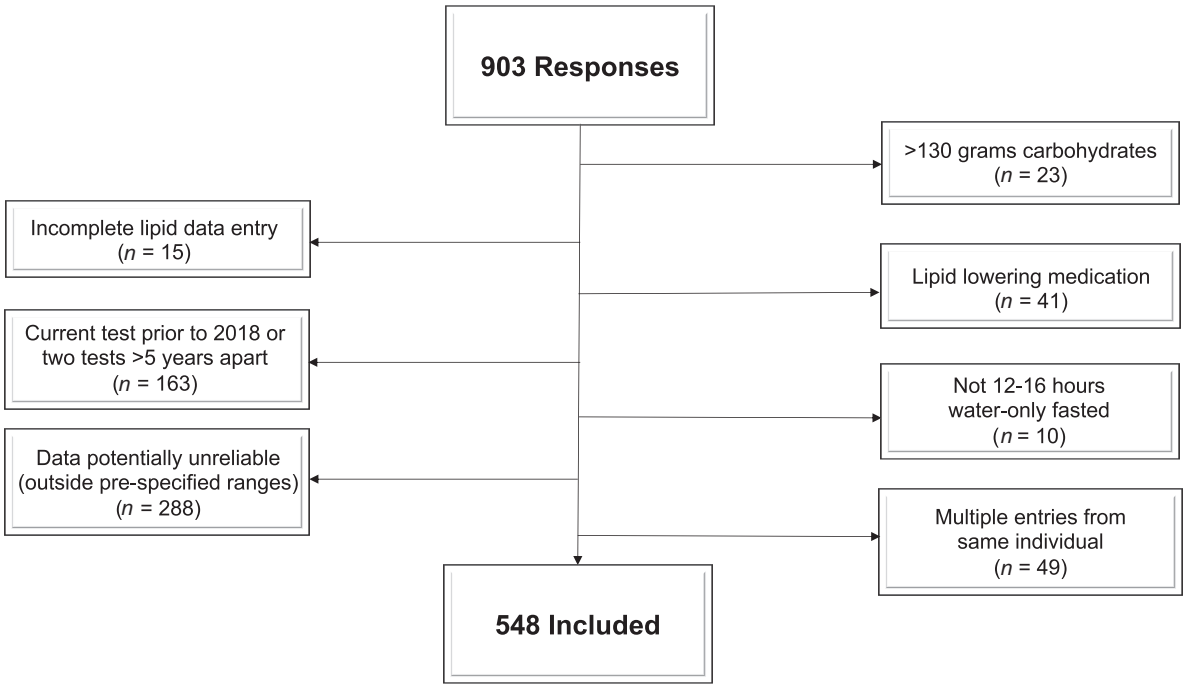

FIGURE 1 Study flow chart. All filters were applied in parallel according to a priori criteria described in Methods. Because filters were applied in parallel some participants were excluded for >1 reason. Thus, numbers excluded for individual reasons exceed the total excluded.

cholesterol was null ($\beta = -0.001$, $P = 0.6$) (model 4). Categorized by quartiles (**Figure 2**), these relations are nearly monotonic along both axes. Comparing respondents in the lowest BMI and lowest TG/HDL cholesterol quartiles with those in the highest BMI and highest TG/HDL cholesterol quartiles, LDL cholesterol increased by median of 135 vs. 35 mg/dL ($P = 1.08 \times 10^{-9}$), a 3.9-fold relative difference. Analyzing all cases with a BMI above 25, LDL cholesterol median change was 59 mg/dL, with 26 showing decreased LDL cholesterol. Similar relations were obtained with use of current TG/HDL cholesterol ratio during consumption of a CRD (**Supplemental Figure 1**).

TABLE 1 Web survey descriptive data[1]

			Percentile				
	Mean	SD	5th	25th	50th	75th	95th
Age, y	51	12	31	43	51	59	69
BMI, kg/m^2	24.1	4.0	19.1	21.6	23.5	26.2	31.7
Carbohydrate, grams	27	24	0	10	20	35	80
Current							
LDL cholesterol	236	107	112	166	209	281	460
HDL cholesterol	76	22	45	59	73	90	115
TG	72	37	27	47	66	86	140
TG/HDL cholesterol ratio	1.1	0.8	0.3	0.6	0.9	1.3	2.7
Prior							
LDL cholesterol	145	59	75	108	134	166	249
HDL cholesterol	63	21	35	48	60	75	101
TG	98	67	31	54	79	120	233
TG/HDL cholesterol ratio	1.8	1.7	0.4	0.8	1.2	2.4	5.3
Change							
ΔLDL cholesterol	91	103	-29	25	72	130	302
ΔHDL cholesterol	13	17	-14	2	13	23	41
ΔTG	–26	59	–134	–42	–12	5	39
ΔTG/HDL cholesterol ratio	–0.8	1.4	–3.7	–1.1	–0.3	0	0.6

[1]Quantile distributions shown for data from 548 eligible responses: males, $n = 319$ (58.2%); females, $n = 228$ (41.6%) (1 individual did not identify sex). TG, triglyceride.

TABLE 2 Prior metabolic health markers predict LDL-cholesterol increases and low BMI[1]

Model and term	β	SE	95% CI	P
Δ LDL cholesterol				
Model 1				
Intercept	68.5	18.1	33, 104	1.7×10^{-4}
Prior HDL cholesterol	0.60	0.22	0.17, 1.04	0.007
Prior TG	–0.17	0.07	–0.3, –0.03	0.015
Model 2				
Intercept	108.9	6.4	96.3, 121.5	2×10^{-16}
Prior TG/HDL cholesterol ratio	–9.9	2.5	–14.9, –4.9	1.1×10^{-4}
Model 3				
Intercept	242.5	26.4	190, 294	2×10^{-16}
Prior TG/HDL cholesterol ratio	–4.5	2.7	–9.81, 0.7	0.09
BMI	–5.9	1.1	–8.2, –3.7	2.7×10^{-7}
Current BMI				
Model 4				
Intercept	27.0	0.7	25.5, 28.4	2×10^{-16}
Prior LDL cholesterol	–0.001	0.003	–0.007, 0.004	0.6
Prior HDL cholesterol	–0.06	0.008	–0.07, –0.04	6.9×10^{-12}
Prior TG	0.01	0.002	0.006, 0.016	2.3×10^{-5}

[1] $n = 548$. Linear regression models reveal that prior high HDL cholesterol and low TG (model 1) and low TG/HDL cholesterol ratio (model 2) predict larger LDL cholesterol changes on a CRD. When BMI is added as a covariate in model 3, prior TG/HDL cholesterol lost significance due to collinearity. Model 4 shows that prior high HDL cholesterol and low TG, but not LDL cholesterol, predict low current BMI. β Values reflect the magnitude of the increase, such that each unit change in the input variable is associated with a proportional unit change in the output (e.g., in model 3, a 1 kg/m^2 decrease in BMI is associated with a 5.9 mg/dL larger increase in LDL cholesterol). CRD, carbohydrate-restricted diet; TG, triglyceride.

Model comparisons suggest prior TG/HDL cholesterol plus BMI best predicts LDL cholesterol change

We compared the all-around performances of 4 models to explain LDL cholesterol change on a CRD, including the following: *1*) prior HDL cholesterol and TG, *2*) prior TG/HDL cholesterol ratio, *3*) BMI, and *4*) prior TG/HDL cholesterol + BMI. As shown in **Supplemental Figure 2** and **Supplemental Table 2**, the model including only BMI performed better than TG/HDL cholesterol ratio alone; however, the TG/HDL cholesterol plus BMI model performed best.

As a parallel approach for identifying the most important predictors of large LDL cholesterol changes on a CRD, without use of any preconceived definition, a machine learning regression tree was

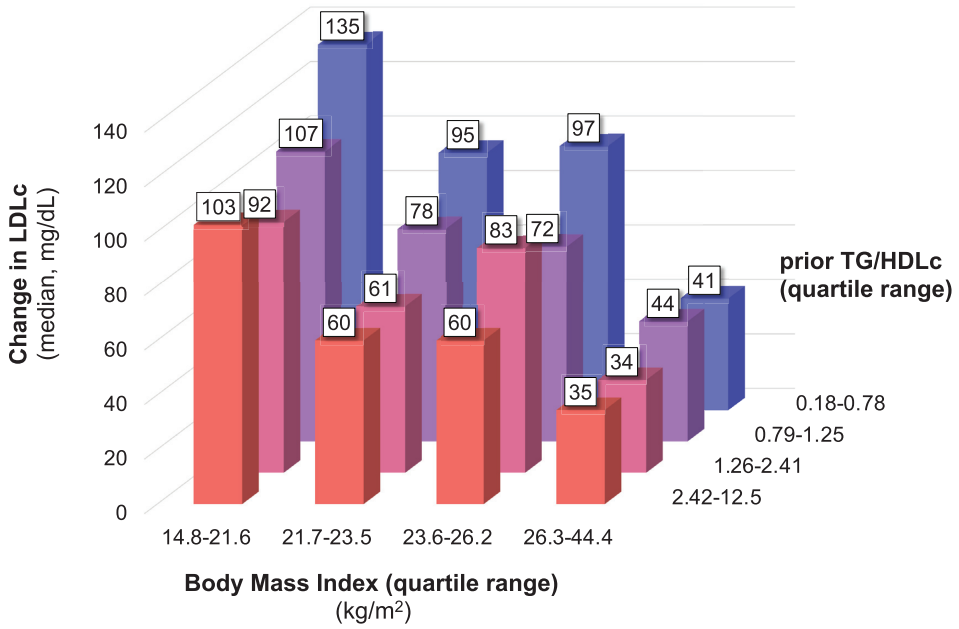

FIGURE 2 BMI and TG/HDL cholesterol ratio predict LDL cholesterol increases on a CRD. Median LDL cholesterol change according to quartiles of TG/HDL cholesterol ratio prior to CRD and of BMI ($n = 34$ per cell) is shown. CRD, carbohydrate-restricted diet; HDLc, HDL cholesterol; LDLc, LDL cholesterol; TG, triglyceride.

TABLE 3 Characterization of LMHR to non-LMHR phenotypes[1]

	Non-LMHRs ($n = 448$)			LMHRs ($n = 100$)			
	Mean	SD	Median	mean	SD	median	*P*
Age, years	51	12	51	51	12	52	0.68
BMI, kg/m^2	24.6	4.1	23.9	22.0	2.7	21.8	1.2×10^{-10}
Carbohydrate, grams	29	24	20	23	19	20	0.07
Sex, % male	61%			45%			0.003
Current							
LDL cholesterol	217	96	191	320	115	286	
HDL cholesterol	71	20	70	99	16	95	
TG	77	38	72	47	15	46	
TG/HDL cholesterol ratio	1.2	0.9	1	0.5	0.2	0.5	
Prior							
LDL cholesterol	145	58	135	148	65	133	0.85
HDL cholesterol	61	19	58	76	22	72	3.0×10^{-11}
TG	104	68	85	66	57	57	3.7×10^{-12}
TG/HDL cholesterol ratio	2.1	1.8	1.4	1.0	0.9	0.7	3.1×10^{-15}
Change							
ΔLDL cholesterol	72	89	61	172	118	146	
ΔHDL cholesterol	11	16	11	23	20	23	
ΔTG	–27	60	–14	–20	55	–10	
ΔTG/HDL cholesterol ratio	–0.8	1.5	–0.4	–0.5	0.9	–0.3	

[1]The LMHR subgroup has lower BMI. The 2 groups do not differ in prior LDL cholesterol, even though the LMHR subgroup has exceptionally high LDL cholesterol on a CRD. *P* values were omitted for variables that define the phenotype or are closely related to those variables. CRD, carbohydrate-restricted diet; LMHR, lean mass hyper-responder; TG, triglyceride.

developed (**Supplemental Figure 3**). BMI emerged as the first branch point, at 26 kg/m^2. Prior HDL cholesterol and TG were also identified as meaningful branch points.

LMHRs have similar pre-diet LDL cholesterol versus nonresponders

As the patterns emerging from our hypothesis-naive analysis were consistent with the LMHR phenotype, we used the a priori cutoffs, as described in Methods, to select respondents who satisfied all 3 criteria for the phenotype ($n = 100$, 18%). Mean LDL cholesterol, HDL cholesterol, and TGs for LMHRs on a CRD were 320 ± 115, 99 ± 16, and 47 ± 15 mg/dL, respectively (**Table 3**). As compared with non-LMHR respondents ($n = 448$), LMHRs were similar in age and were more likely to be female. Notably, the LMHR subgroup had markedly lower mean BMI (22.0 vs. 24.6, $P = 1.2 \times 10^{-10}$) and larger median LDL cholesterol increases (146 mg/dL vs. 61 mg/dL, $P = 1.9 \times 10^{-18}$) (**Supplemental Figure 4**A, B). With regard to baseline lipid data, LMHRs had higher HDL cholesterol ($P = 3.0 \times 10^{-11}$) and lower TG ($P = 3.7 \times 10^{-12}$), but similar LDL cholesterol. Although LMHRs had much higher LDL cholesterol on a CRD, median prior LDL cholesterol values were 133 mg/dL for LMHRs and 135 mg/dL for non-LMHRs ($P = 0.85$).

Recognizing the highly selected nature of our sample, we compared BMI and lipid values, both prior and current, of our respondents with NHANES data (**Figure 3**, **Supplemental Table 3**). Overall, respondents were leaner (BMI: LMHRs, 22.0 ± 2.7; non-LMHRs, 24.6 ± 4.1; NHANES, 28.5 ± 6.8) and had higher LDL cholesterol and HDL cholesterol, and lower TG, correlated with differences in BMI. Differences in BMI, LDL cholesterol, HDL cholesterol, and TG were most pronounced in LMHRs.

Moderate reintroduction of carbohydrate lowers LDL cholesterol among patients consuming a VLCD

Five patients consuming a VLCD with <25 grams/day available carbohydrate presented to a primary care practice with LDL cholesterol of 239 to 665 mg/dL, representing extreme elevations from pre-diet levels (113 to 141 mg/dL). Genetic testing for familial hypercholesterolemia was negative in each case. After refusing statin therapy, the patients chose to pursue an empiric clinician-supervised protocol with moderate reintroduction of carbohydrate (50 to 100 grams/day). Case histories of each patient are presented in the **Supplemental Case Series**. As summarized in **Table 4**, this dietary intervention was associated with a large decrease in LDL cholesterol in all patients, ranging from –100 mg/dL to −480 mg/dL. The 2 patients who met criteria for LMHR (MI and IA) showed the largest increases in LDL cholesterol upon initiation of a VLCD and the largest reductions in LDL cholesterol with moderate reintroduction of carbohydrate.

Discussion

The results of this study suggest that a large elevation in LDL cholesterol on a CRD is more likely to occur in people who are lean and metabolically healthy. Low TG/HDL cholesterol ratio prior to consumption of a current CRD and low BMI were strongly associated with a larger LDL cholesterol increase. Moreover, we found evidence to support the existence of an LMHR phenotype, using an a priori definition, characterizing individuals with unremarkable baseline LDL cholesterol and marked LDL cholesterol elevation on a CRD. This phenotype contrasts with the usual pattern of atherogenic dyslipidemia observed in populations at high risk for ASCVD, in which normal to high LDL cholesterol occurs with high TG and low HDL cholesterol (15, 17).

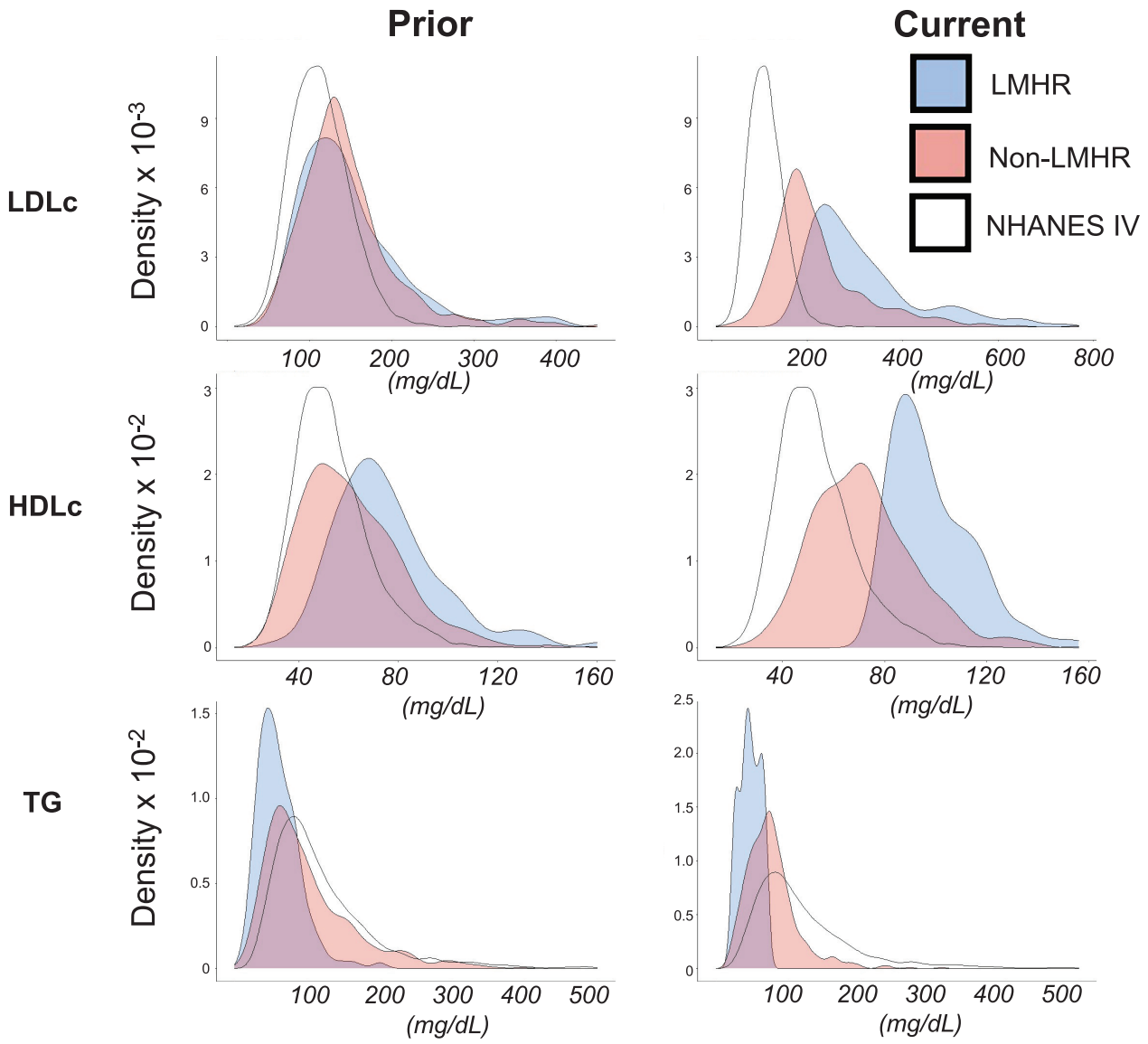

FIGURE 3 Comparison of LMHR and non-LMHR subgroups among respondents with US nationally representative data (NHANES IV). LMHRs and non-LMHRs possess higher LDL cholesterol on a CRD, as well as high HDL cholesterol and lower TG both prior and on a CRD, as compared with adults in the NHANES IV (2011–2012) dataset. Differences from NHANES were more pronounced for LMHRs (see Supplemental Table 3 for details). CRD, carbohydrate-restricted diet; HDLc, HDL cholesterol; LDLc, LDL cholesterol; LMHR, lean mass hyper-responder(s); TG, triglyceride.

The significance of elevated LDL cholesterol in the context of good metabolic health markers warrants consideration. As the prevalence of obesity, type 2 diabetes, and metabolic syndrome increase, alongside a global downward trend in LDL cholesterol (due to, in part, to more powerful LDL cholesterol–lowering pharmacotherapy), a high TG/HDL cholesterol ratio and an LDL cholesterol profile characterized by small particles now comprise the dominant dyslipidemia among those with ASCVD (21). Our respondents showed the opposite pattern. Recent data from the Women's Heart Study, a large prospective trial assessing risk factors for ASCVD, found that atherogenic dyslipidemia (including high TG and low HDL cholesterol) may contribute more to ASCVD risk than high LDL cholesterol (22). In the Scandinavian Simvastatin Survival Study (23), individuals with isolated elevated LDL cholesterol, compared with those who also had high TG and low HDL cholesterol, were at lower risk for coronary events and benefited less from statins, findings consistent with other studies (24). Furthermore, a CRD tends to increase LDL particle size (25), resulting in a lipoprotein profile associated with lower risk at a given LDL cholesterol concentration (22, 26) (although we do not have particle-size data for our respondents). In some circumstances, elevated LDL cholesterol on a CRD may be associated with reduced small LDL particle number (1, 27). However, these data cannot exclude the important possibility of increased ASCVD risk

TABLE 4 Case series summary[1]

Patient initials	Pre-VLCD	VLCD	LCD	LDL cholesterol decrease
IA[2]				
Total cholesterol	214	797	294	–480
LDL cholesterol	116	665	185	
HDL cholesterol	81	122	95	
TG	84	50	72	
TG/HDL cholesterol ratio	1.0	0.4	0.8	
MI[2]				
Total cholesterol	209	698	497	–223
LDL cholesterol	122	583	360	
HDL cholesterol	72	97	122	
TG	54	70	67	
TG/HDL cholesterol ratio	0.8	0.7	0.5	
RO				
Total cholesterol	197	311	180	–124
LDL cholesterol	137	239	115	
HDL cholesterol	45	65	54	
TG	62	56	36	
TG/HDL cholesterol ratio	1.4	0.9	0.7	
NM				
Total cholesterol	179	387	272	–122
LDL cholesterol	113	317	195	
HDL cholesterol	49	59	61	
TG	86	54	56	
TG/HDL cholesterol ratio	1.8	0.9	0.9	
AN				
Total cholesterol	218	423	318	–100
LDL cholesterol	141	336	236	
HDL cholesterol	57	69	66	
TG	98	74	64	
TG/HDL cholesterol ratio	1.7	1.1	1.0	

[1]Lipid values for five patients refusing LDL cholesterol–lowering pharmacotherapy before a VLCD, during consumption of a VLCD, and on an LCD (i.e., after moderate reintroduction of carbohydrate). Results are in mg/dL except for TG/HDL cholesterol ratio. LCD, low-carbohydrate diet; LMHR, lean mass hyper-responder; TG, triglyceride; VLCD, very-low-carbohydrate diet.
[2]Indicates those patients satisfying criteria for LMHR during consumption of VLCD. Patients are ordered according to TG/HDL cholesterol ratio on a VLCD.

associated with elevated LDL cholesterol on a CRD, especially among individuals with severe elevations, a possibility that requires prospective study.

One major interpretive issue of our observational study involves selection bias. Our sample had substantially lower BMI and better TG/HDL cholesterol ratio compared with US nationally representative data, and participants were previously aware of their LDL cholesterol change by virtue of the study design. Thus, our findings are not directly generalizable. Nevertheless, even with a sample biased toward leanness and good metabolic health markers, we had sufficient heterogeneity to observe strong associations over a large range in LDL cholesterol response to a CRD. If these associations were extrapolatable to a broader population, we would expect an even smaller increase in LDL cholesterol among individuals with high BMI as compared with the median LDL cholesterol increase observed among respondents in the highest BMI and TG/HDL cholesterol quantiles (see Figure 2). Such individuals are more characteristic of patients with diet-related chronic disease, such as type 2 diabetes, for whom a CRD holds particular interest. This possibility is consistent with several clinical trials of a CRD involving participants with obesity or type 2 diabetes, in which little or no LDL cholesterol increase occurred (6–8, 10, 11, 14).

Changes in saturated fat intake were not measured in this study and may have contributed to any individual's increases in LDL cholesterol. However, this possibility implies an inverse association between BMI and saturated fat intake. As depicted in **Supplemental Figure 5**, this alternative explanation presupposes that leaner participants with a low TG/HDL cholesterol ratio preferentially consumed substantially more saturated fat than participants with a higher BMI and TG/HDL cholesterol ratio.

We believe a more likely explanation involves physiological mechanisms relating directly to energy metabolism. By analogy, sodium-glucose cotransporter 2 (SGLT2) inhibitors, which promote fat oxidation and ketosis, increase LDL cholesterol (and, interestingly, reduce ASCVD risk), raising the possibility that a shift in substrate oxidation from carbohydrate to fat intrinsically elevates LDL cholesterol (28). Thus, reduced intake of carbohydrate may increase systemic trafficking of lipid energy through VLDL lineage particles coincident with high lipoprotein-lipase–mediated remodeling of VLDL into LDL and HDL (29) and resulting in a profile of elevated LDL cholesterol and HDL cholesterol, and reduced TG. We speculate that this effect may be greatest in lean, insulin sensitive individuals with high energy demands, a possibility consistent with other research (30–32) and warranting future investigation.

With regard to other limitations, we did not conduct a detailed dietary assessment and thus cannot assess other contributory influences, beyond saturated fat, on lipids. In addition, data were collected by self-report, and bias due to misreporting cannot be excluded. In contrast to laboratory data, to which respondents may have direct access, recall of prior BMI may be especially susceptible to bias. In our survey, we did not interrogate prior BMI and, therefore, further study is needed in the use of BMI for prognostic purposes. This issue would not apply to the predictive associations involving prior lipids, nor to characterization of the LMHR phenotype.

A strength of the study included its large sample size with excellent power to test a priori hypotheses, including in hypothesis-naive exploratory models. Despite issues of generalizability, the survey findings receive some clinical support from a patient case series. Furthermore, we have made all data and analysis codes publicly available, to facilitate reanalysis and further investigation.

In conclusion, the results of this study identify major potential sources of heterogeneity in LDL cholesterol response to a CRD. This finding suggests that patients with obesity and related disease, for whom a CRD may hold special promise, may be at low risk of experiencing a clinically significant increase in LDL cholesterol with this dietary intervention, while potentially experiencing improvements in other ASCVD risk markers, including atherogenic dyslipidemia, lipoprotein insulin resistance, and lipoprotein(a) [Lp(a)] (12). In contrast, lean, physically active individuals may be unique susceptibility to LDL cholesterol increases on a CRD. Prospective observational research and interventional studies will be needed to explore these findings, assess the associated ASCVD risk, and examine causal mechanisms. This study should not be interpreted as implying cardiovascular safety of the LMHR phenotype.

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Data Availability

Data described in the manuscript, code book, and analytic code are publicly and freely available at https://github.com/AdrianSotoM/LMHR.

References

1. Creighton BC, Hyde PN, Maresh CM, Kraemer WJ, Phinney SD, Volek JS. Paradox of hypercholesterolaemia in highly trained, keto-adapted athletes. BMJ Open Sport Exerc Med 2018;4(1):e000429.
2. Buren J, Ericsson M, Damasceno NRT, Sjodin A. A ketogenic low-carbohydrate high-fat diet increases LDL cholesterol in healthy, young, normal-weight women: a randomized controlled feeding trial. Nutrients 2021;13(3):814.
3. Retterstol K, Svendsen M, Narverud I, Holven KB. Effect of low carbohydrate high fat diet on LDL cholesterol and gene expression in normal-weight, young adults: a randomized controlled study. Atherosclerosis 2018;279: 52–61.
4. Valsdottir TD, Henriksen C, Odden N, Nellemann B, Jeppesen PB, Hisdal J, Westerberg AC, Jensen J. Effect of a low-carbohydrate high-fat diet and a single bout of exercise on glucose tolerance, lipid profile and endothelial function in normal weight young healthy females. Front Physiol 2019;10:1499.
5. Sharman MJ, Gomez AL, Kraemer WJ, Volek JS. Very low-carbohydrate and low-fat diets affect fasting lipids and postprandial lipemia differently in overweight men. J Nutr 2004;134(4):880–5.
6. Seshadri P, Iqbal N, Stern L, Williams M, Chicano KL, Daily DA, McGrory J, Gracely EJ, Rader DJ, Samaha FF. A randomized study comparing the effects of a low-carbohydrate diet and a conventional diet on lipoprotein subfractions and C-reactive protein levels in patients with severe obesity. Am J Med 2004;117(6):398–405.
7. Hays JH, DiSabatino A, Gorman RT, Vincent S, Stillabower ME. Effect of a high saturated fat and no-starch diet on serum lipid subfractions in patients with documented atherosclerotic cardiovascular disease. Mayo Clin Proc 2003;78(11):1331–6.
8. Hyde PN, Sapper TN, Crabtree CD, LaFountain RA, Bowling ML, Buga A, Fell B, McSwiney FT, Dickerson RM, Miller VJ, et al. Dietary carbohydrate restriction improves metabolic syndrome independent of weight loss. JCI Insight 2019;4(12):e128308.
9. Dong T, Guo M, Zhang P, Sun G, Chen B. The effects of low-carbohydrate diets on cardiovascular risk factors: a meta-analysis. PLoS One 2020;15(1):e0225348.
10. Dashti HM, Al-Zaid NS, Mathew TC, Al-Mousawi M, Talib H, Asfar SK, Behbahani AI. Long term effects of ketogenic diet in obese subjects with high cholesterol level. Mol Cell Biochem 2006;286(1-2):1–9.
11. Samaha FF, Iqbal N, Seshadri P, Chicano KL, Daily DA, McGrory J, Williams T, Williams M, Gracely EJ, Stern L. A low-carbohydrate as compared with a low-fat diet in severe obesity. N Engl J Med 2003;348(21): 2074–81.
12. Ebbeling CB, Knapp A, Johnson A, Wong JMW, Greco KF, Ma C, Mora S, Ludwig DS. Effects of a low-carbohydrate diet on insulin-resistant dyslipoproteinemia—a randomized controlled feeding trial. Am J Clin Nutr 2021. doi: https://doi.org/10.1093/ajcn/nqab287.
13. Griffin BA, Mensink RP, Lovegrove JA. Does variation in serum LDL-cholesterol response to dietary fatty acids help explain the controversy over fat quality and cardiovascular disease risk? Atherosclerosis 2021;328: 108–13.
14. Athinarayanan SJ, Adams RN, Hallberg SJ, McKenzie AL, Bhanpuri NH, Campbell WW, Volek JS, Phinney SD, McCarter JP. Long-term effects of a novel continuous remote care intervention including nutritional ketosis for the management of type 2 diabetes: a 2-year non-randomized clinical trial. Front Endocrinol 2019;10:348.
15. Bagheri P, Khalili D, Seif M, Rezaianzadeh A. Dynamic behavior of metabolic syndrome progression: a comprehensive systematic review on recent discoveries. BMC Endocr Disord 2021;21(1):54.
16. Feldman D. Cholesterol code. Are you a lean mass hyper-responder? CholesterolCode.com2017 [Internet]. Available from: https://cholesterolcode.com/are-you-a-lean-mass-hyper-responder/.
17. Rizzo M, Barylski M, Rizvi AA, Montalto G, Mikhailidis DP, Banach M. Combined dyslipidemia: should the focus be LDL cholesterol or atherogenic dyslipidemia? Curr Pharm Des 2013;19(21):3858–68.
18. Therneau TM, Atkinson EJ. An introduction to recursive partitioning using the RPART routines [Internet]. Foundation M. 2019. Available from: https://cran.r-project.org/web/packages/rpart/vignettes/longintro.pdf.
19. Boizel R, Benhamou PY, Lardy B, Laporte F, Foulon T, Halimi S. Ratio of triglycerides to HDL cholesterol is an indicator of LDL particle size in

patients with type 2 diabetes and normal HDL cholesterol levels. Diabetes Care 2000;23(11):1679–85.
20. Moriyama K. The association between the triglyceride to high-density lipoprotein cholesterol ratio and low-density lipoprotein subclasses. Intern Med 2020;59(21):2661–9.
21. Libby P. The changing landscape of atherosclerosis. Nature 2021;592(7855):524–33.
22. Dugani SB, Moorthy MV, Li C, Demler OV, Alsheikh-Ali AA, Ridker PM, Glynn RJ, Mora S. Association of lipid, inflammatory, and metabolic biomarkers with age at onset for incident coronary heart disease in women. JAMA Cardiol 2021;6(4):437–47.
23. Ballantyne CM, Olsson AG, Cook TJ, Mercuri MF, Pedersen TR, Kjekshus J. Influence of low high-density lipoprotein cholesterol and elevated triglyceride on coronary heart disease events and response to simvastatin therapy in 4S. Circulation 2001;104(25):3046–51.
24. Jeppesen J, Hein HO, Suadicani P, Gyntelberg F. Low triglycerides-high high-density lipoprotein cholesterol and risk of ischemic heart disease. Arch Intern Med 2001;161(3):361–6.
25. Falkenhain K, Roach LA, McCreary S, McArthur E, Weiss EJ, Francois ME, Little JP. Effect of carbohydrate-restricted dietary interventions on LDL particle size and number in adults in the context of weight loss or weight maintenance: a systematic review and meta-analysis. Am J Clin Nutr 2021;114(4):1455–66.
26. St-Pierre AC, Cantin B, Dagenais GR, Mauriege P, Bernard PM, Despres JP, Lamarche B. Low-density lipoprotein subfractions and the long-term risk of ischemic heart disease in men: 13-year follow-up data from the Quebec Cardiovascular Study. Arterioscler Thromb Vasc Biol 2005;25(3):553–9.
27. Norwitz NG, Loh V. A standard lipid panel is insufficient for the care of a patient on a high-fat, low-carbohydrate ketogenic diet. Front Med 2020;7:97.
28. Basu D, Huggins LA, Scerbo D, Obunike J, Mullick AE, Rothenberg PL, Di Prospero NA, Eckel RH, Goldberg IJ. Mechanism of increased LDL (low-density lipoprotein) and decreased triglycerides with SGLT2 (sodium-glucose cotransporter 2) inhibition. Arterioscler Thromb Vasc Biol 2018;38(9):2207–16.
29. Kontush A. HDL and reverse remnant-cholesterol transport (RRT): relevance to cardiovascular disease. Trends Mol Med 2020;26(12):1086–100.
30. Bak AM, Vendelbo MH, Christensen B, Viggers R, Bibby BM, Rungby J, Jorgensen J, Moller N, Jessen N. Prolonged fasting-induced metabolic signatures in human skeletal muscle of lean and obese men. PLoS One 2018;13(9):e0200817.
31. Bartelt A, John C, Schaltenberg N, Berbee JFP, Worthmann A, Cherradi ML, Schlein C, Piepenburg J, Boon MR, Rinninger F, et al. Thermogenic adipocytes promote HDL turnover and reverse cholesterol transport. Nat Commun 2017;8(1):15010.
32. Cinkajzlova A, Mraz M, Lacinova Z, Klouckova J, Kavalkova P, Kratochvilova H, Trachta P, Krizova J, Haluzikova D, Skrha J, et al. Angiopoietin-like protein 3 and 4 in obesity, type 2 diabetes mellitus, and malnutrition: the effect of weight reduction and realimentation. Nutr Diabetes 2018;8(1):21.