ALINEA

NUTRITION

MARCH 2022

www.alineanutrition.com

TABLE OF
CONTENTS

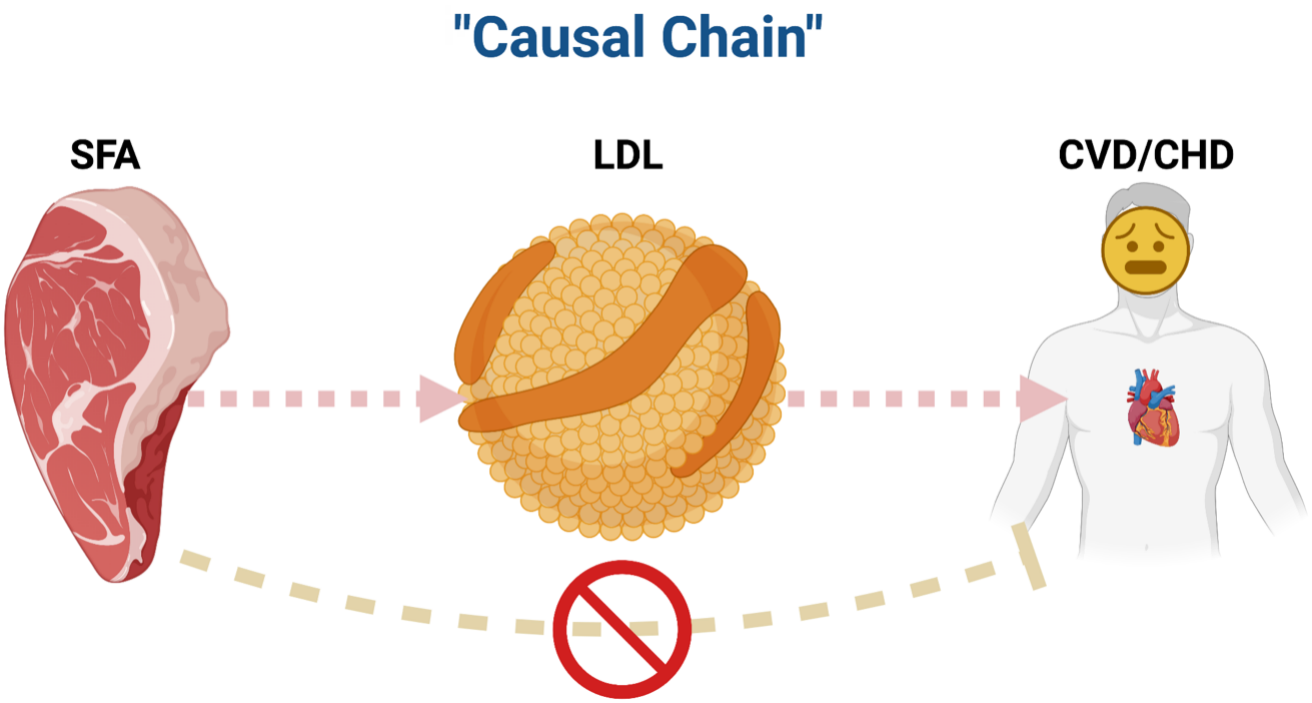
Kelly RK, Watling CZ, Tong TYN, Piernas C, Carter JL, Papier K, Key TJ, Perez-Cornago A. Associations Between Macronutrients From Different Dietary Sources and Serum Lipids in 24 639 UK Biobank Study Participants. Arterioscler Thromb Vasc Biol. 2021 Jul;41(7):2190-2200.

What We Know, Think We Know, or Are Starting to Know

The epidemiology of diet and cardiovascular disease could hardly get more controversial. And thankfully the study we Deepdive into today has gone some way to helping resolve some of the conflict. But we' ll get to that…

Most of the controversy is generated by studies claiming that there is no association between saturated fat and 'hard endpoints' , i.e., cardiovascular [CVD] or coronary heart disease [CHD] events and/or mortality. The whys and wherefores of the controversy, we have covered in some detail in this article on the saturated fat 'no association' meta-analyses, this article on the substitution effects of replacing saturated fat with other macronutrients, together with their respective video lectures.

To sum it up in a sentence, the association is there, but it depends on levels of intake, what nutrient replaces saturated fat in the diet, and whether blood cholesterol levels have been adjusted for. The reason why this latter factor is important is illustrated below:



Take a look at the **Figure**, above; we know that LDL-cholesterol is the primary causal risk factor for CVD/CHD [1,2]. And we know that saturated fat raises blood cholesterol and LDL-C to a greater magnitude than any macronutrient [3–5]. So, the causal chain would link saturated fat to CVD/CHD *via* the effects of SFA on LDL-C. However, many analyses make the assumption that saturated fat has direct effects. These analyses usually adjust for LDL-C, in effect giving us the relationship represented by the bottom inhibitor/stop-sign line; a lack of, or weak, association.

And that is only just saturated fat; what of carbohydrate? Siri-Tarino *et al*. [6] suggested that replacing SFA with carbohydrate did not lower CVD risk, but did not distinguish between carbohydrate subtypes, e.g., complex vs. refined, added sugars vs. intrinsic sugars [i.e., sugars naturally occurring in foods]. Thus, the question over associations between specific effects of types of carbohydrates in the diet.

All of which leads us to the present study. Generally, most nutritional epidemiological research does not focus on clinical risk factors, i.e., measuring blood cholesterol or blood pressure, because it is expensive and requires participants to attend a clinical visit in person. But some of the more recent large-scale cohorts have the capacity to do so. The study we look at today investigated the associations between macronutrients and blood lipid levels in the UK Biobank cohort.

The Study

The UK Biobank is a large observational cohort of participants aged between 40-69yrs, registered with the National Health Service [NHS] in the United Kingdom. Participants completed a validated baseline 24hr dietary recall, and were invited to complete up to four subsequent 24hr recalls over the course of a year.

Dietary macronutrient intakes were based on the averages of a minimum of two 24 h recalls per participant and expressed as a percentage of energy, and adjusted for total energy intake*. At least two completed 24hr recalls were required to be included in the study. The dietary assessment also separated macronutrients by food source, e.g., animal fat and plant fat, dairy fat and non-dairy fat, etc.

Participants also had blood samples taken at baseline, and a subgroup of participants had a repeated blood sample taken 4yrs later.

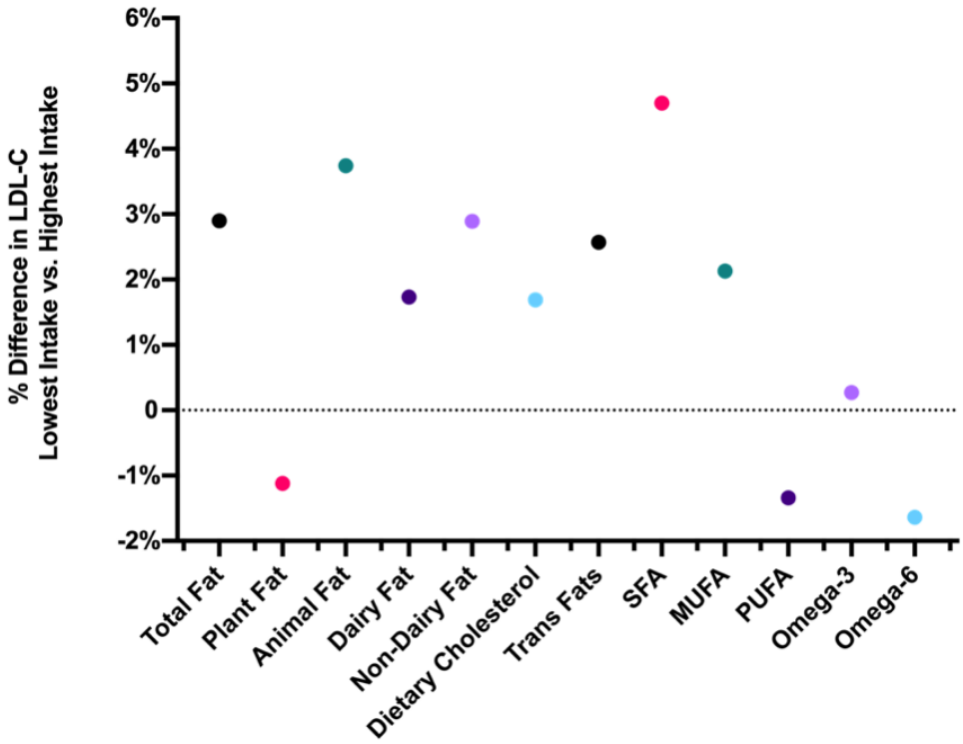
The primary exposure of interest were macronutrients and their food sources, while the outcomes of interest included a full blood lipid profile of total cholesterol [TC], LDL-C, HDL-C, triglycerides [TG], and ApoB [a marker of all atherogenic lipoproteins beyond LDL] and ApoA1 [a marker for all HDL particles].

*Geek Box: Adjusting for Total Energy Intake in Epidemiology

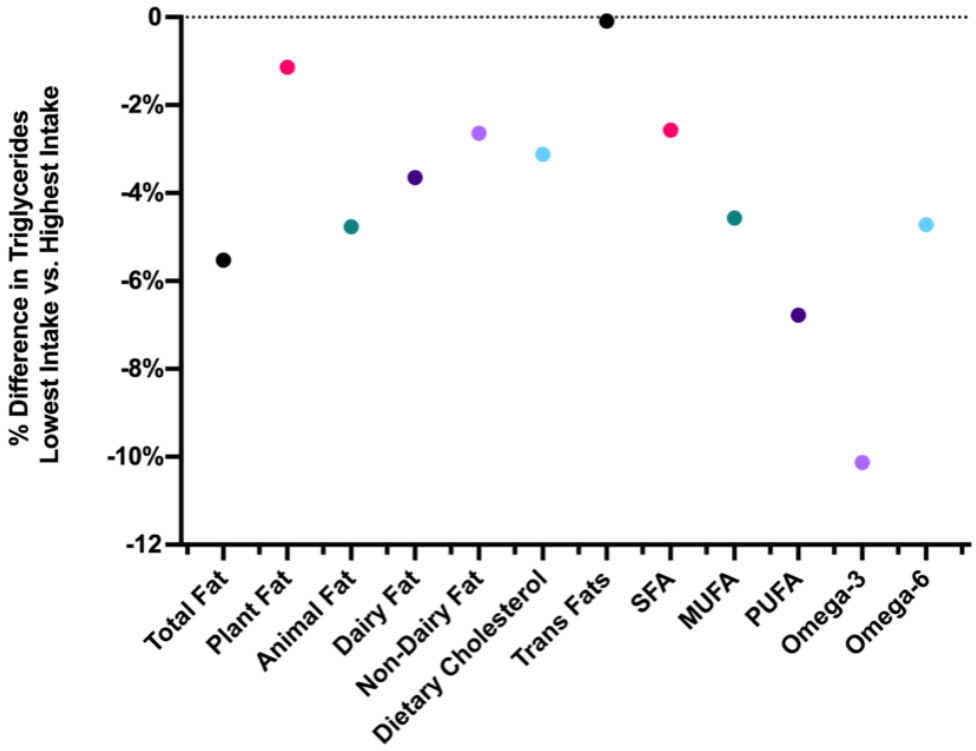
Total energy intake is a critical factor to account for in any epidemiological analysis of diet and disease for three main reasons. First, the level of energy intake itself may be the primary factor influencing disease risk. Secondly, individuals vary in their body size and physical activity levels, and consequently in their total energy intake; this means absolute levels of nutrient intake will vary from person to person, which could introduce random error into the analysis. Finally, if a nutrient is a more direct cause of disease than total energy intake, then the effect of a nutrient of interest would be distorted by total energy intake [because as energy increases or decreases, the intake of the nutrient would increase or decrease]. The best analogy for adjusting for total energy in epidemiology is that it is seeking to achieve the same effect as having isocaloric diets in an intervention. Let's say you want to compare two diets, one high in fat and lower in carbohydrate vs. the opposite: if one diet had less energy, we would say the effects were likely due to this difference in total energy intake. This is the same for epidemiology: the exposure of interest is the composition of diet, independent of total energy intake. For epidemiology, other methods have been proposed to more assess total energy intake, in particular adjusting for body weight and adjusting for physical activity. However, these methods do not cancel out measurement errors because of body weight and physical activity are independently related to energy intake. However, because both total energy and individual nutrients are calculated from the same foods, the errors for both are strongly correlated. Therefore, by carefully adjusting the intake of a nutrient for total energy intake, these correlated errors cancel each other out and the validity of the measure of a nutrient is improved. There are a number of methods of adjusting for total energy, including the nutrient-density method, the energy-adjusted residual method, the energy partition method, and multivariate methods. Each has certain advantages and limitations, and it is important to consider what the variable of interest is, how that variable relates to other factors, and the implications for biological plausibility of the chosen method.

Results: 24,639 participants were included in the final analysis. Of the included participants, 8,113 had completed 2 of the 24hr recalls, 6,921 had completed 3, 5,953 had completed 4, and 3,652 had completed 5. For this results section, we will focus on the effects on LDL-C and triglycerides.

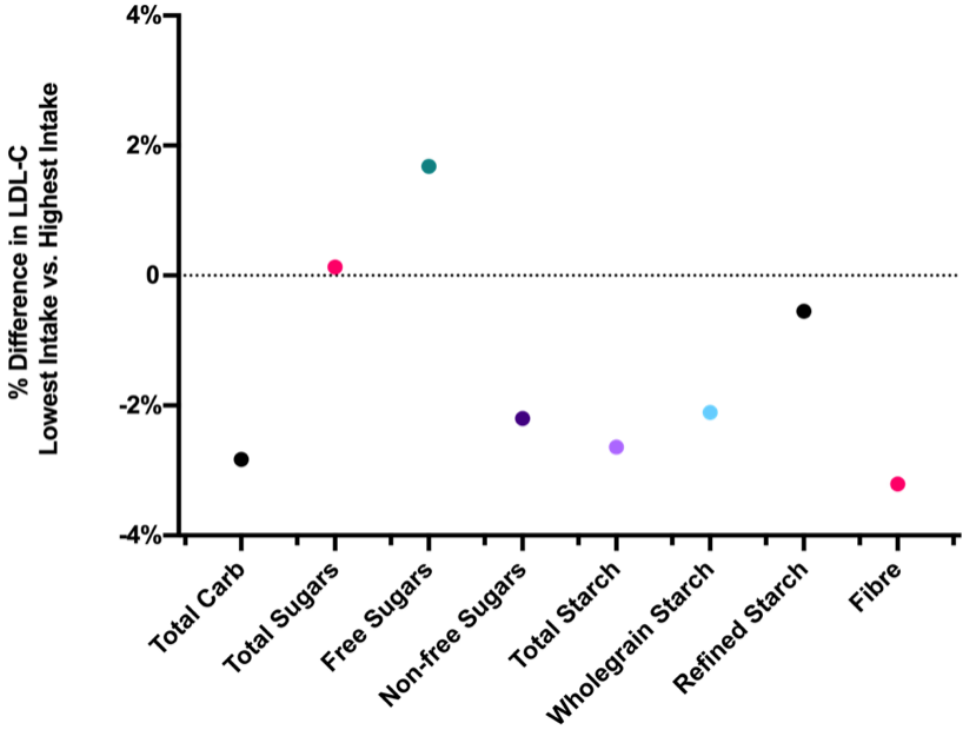
- ***Dietary Fat & LDL*:** The effects of different fat sources on LDL-C are summarised in the Figure, below. The left Y-axis is the percentage difference in LDL-C comparing the lowest level of each fat source to the highest level; the X-axis labels each fat source analysed. Of note, the highest saturated fat level was associated with a 4.7% higher LDL-C level, while the highest polyunsaturated fat level was associated with 1.34% lower LDL-C.



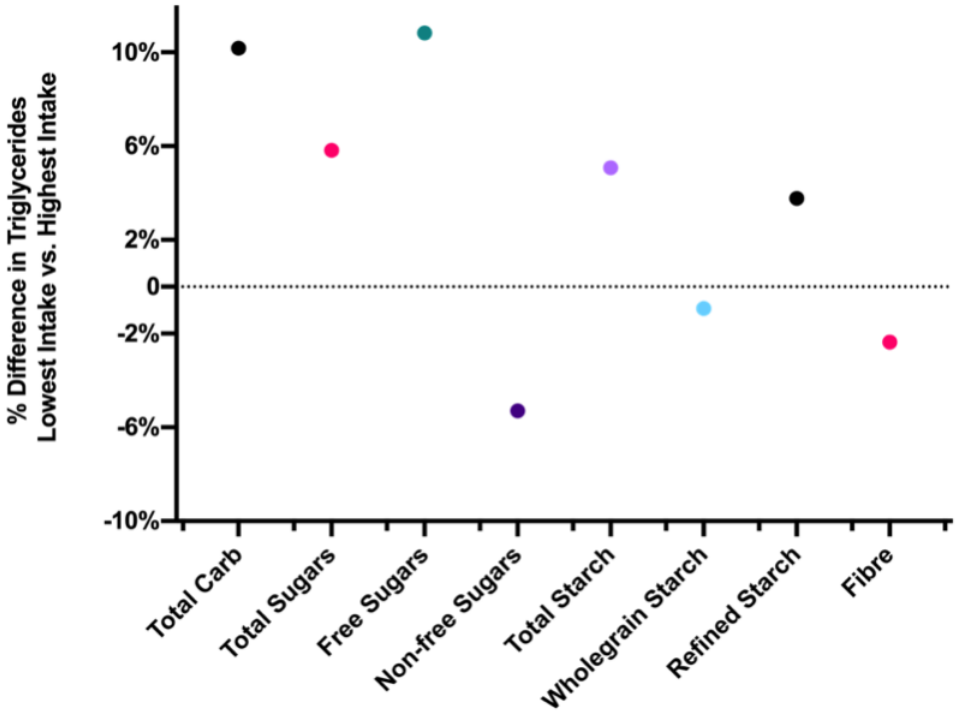
- ***Dietary Fat & TG*:** The effects of different fat sources on TG are summarised in the Figure, below. The left Y-axis is the percentage difference in TG comparing the lowest level of each fat source to the highest level; the X-axis labels each fat source analysed. Of particular note, the highest polyunsaturated fat level was associated with 6.78% lower TG, while the highest dietary intake of omega 3 fatty acids was associated with a 10.13% lower TG.



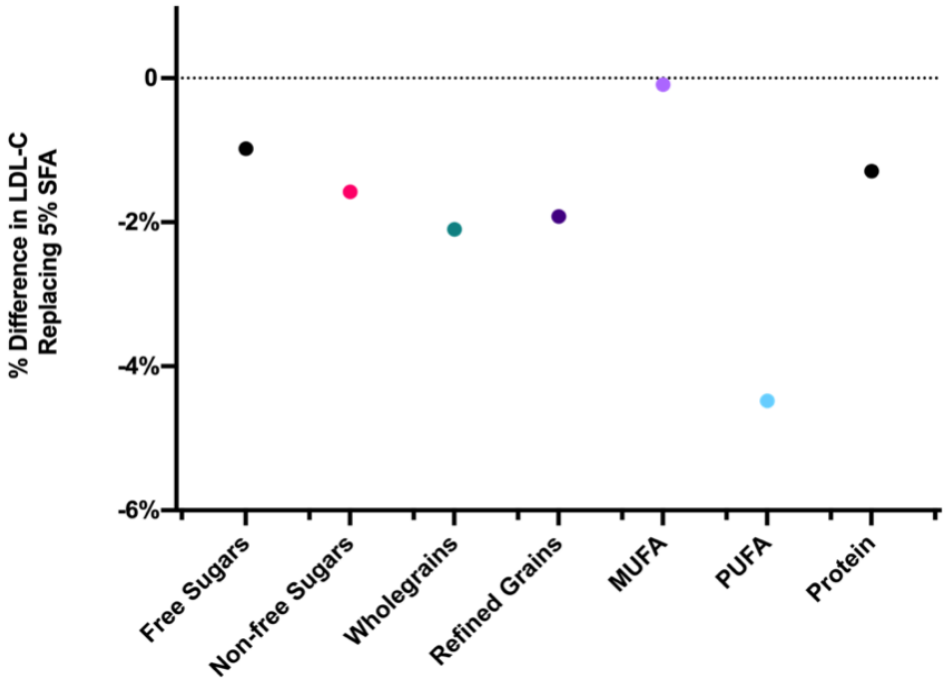
- ***Carbohydrate & LDL*:** The effects of different carbohydrate sources on LDL-C are summarised in the Figure, below. The left Y-axis is the percentage difference LDL-C comparing the lowest level of each carbohydrate source to the highest level; the X-axis labels each carbohydrate source analysed. Of note, the highest free sugar level was associated with a 1.68% higher LDL-C level, while the highest fibre level was associated with 3.21% lower LDL-C.



- ***Carbohydrate & TG*:** The effects of different carbohydrate sources on TG are summarised in the Figure, below. The left Y-axis is the percentage difference TG comparing the lowest level of each carbohydrate source to the highest level; the X-axis labels each carbohydrate source analysed. Of note, the highest free sugar level was associated with a 10.83% higher TG level, while the highest non-free sugar intake was associated with 5.3% lower TG.



- ***Substitution Analysis on LDL-C*:** The effects of replacing 5% of energy from saturated fat with an isocaloric exchange of 5% of energy from another nutrient was also analysed, as is summarised in the Figure, below. The percentage on the left Y-axis is the difference in LDL-C after replacing saturated fat; the X-axis is the nutrient replacing saturated fat. Of note, replacing 5% energy from saturated fat with 5% energy from polyunsaturated fat was associated with a 4.48% lower LDL-C level.



The Critical Breakdown

Pros: The dietary assessment used repeated measures of diet, and 2/3 of all participants had completed at least three 24hr recalls [*n* = 9,605 completed 4-5]. Although 24hr recalls have the potential for measurement error when a single recall is administered, having repeated assessments would be expected to improve accuracy. The study adjusted for total energy intake, which is important because the results may then be taken to represent the effect of a macronutrient independent of the effects of energy intake. The study used clinical measures of blood lipids and had repeated blood lipid measurements in a subgroup of participants. The analysis included a substitution analysis which modelled the effects of replacing 5% of energy from saturated fat with 5% energy from other macronutrient [i.e., isocaloric substitution]. Several sensitivity analyses were also conducted to determine whether the associations differed according to number of completed 24hr recalls, repeated blood lipid measurements, or extremely high or low blood lipid levels. The study had a good sample size.

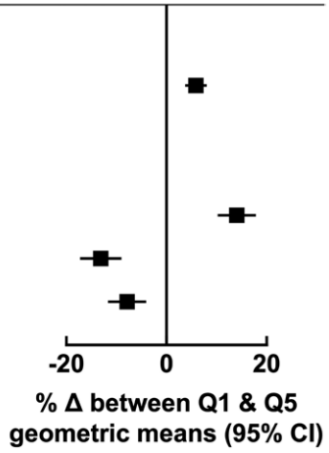
Cons: The main limitation of the present study is the potential selection bias and generalisability. As the study requires compliance and participants willing to commit to repeated measurements over time, this may lead to some selection/healthy user bias, i.e., people with a better health profile than the general population may be more inclined to participate in research. Further, the majority of the participants are White ethnicity, which may lead to generalisability issues for the wider British population. That said, both of these limitations are *maybes*; the reality is that this is a very well designed and executed work of nutritional epidemiology.

Key Characteristic

Accurately capturing representative diet is one of the most well-known challenges for nutritional epidemiology. One of the major criticisms in this regard is the lack of repeated measures of diet and other important variables over time. This study thus has a major strength in conducting repeated dietary assessments and having a subgroup of participants with repeated blood lipid measurements 4yrs later.

The authors conducted a sensitivity analysis to see whether the associations in the main analysis remained in the participants with repeated blood lipid measurements, which you can see in the Figure, below; the findings not only mirrored the main analysis but were strengthened. Participants with repeated blood lipid measurements still showed a near 5.86% higher LDL-C level from saturated fat intake and 14% higher TG from free sugars. These findings strengthen the primary analysis, showing stability over time of the effects of different nutrients on these lipid outcomes.

Nutrients	Geometric mean in Q1 (95% CI)	Geometric mean in Q5 (95% CI)	% Δ between Q1 & Q5 geometric means
LDL-C			
SFA	3.48 (3.43 - 3.52)	3.69 (3.64 - 3.73)	5.86%
Triglycerides			
Free sugars	1.37 (1.34 - 1.40)	1.58 (1.54 - 1.62)	14.02%
Non-free sugars	1.56 (1.52 - 1.60)	1.37 (1.33 - 1.40)	-13.14%
n-3 fatty acids	1.52 (1.48 - 1.56)	1.40 (1.37 - 1.44)	-7.88%



Interesting Finding

The finding in relation to types of dietary sugars is novel and adds more nuance to our understanding of the relationship between diet and CVD/CHD risk. In this study, "free sugars" were classified as added sugars, i.e., sugars added to drinks or those found in honey and fruit juices. "Non-free sugars" were classified as remaining dietary sugars minus free sugars, which could include sugars from granola and other such foods.

In some ways, this finding is corroborating wider research, including from controlled feeding studies. For example, it is known that high intakes of free sugars lead to increased TG synthesis and circulating levels of both fasting and postprandial TGs [7]. However, it has become somewhat of a dogma to state that carbohydrates generally raise TGs, and while this is evident in the present study, it is important to know that these findings relate to *total carbohydrate*, without distinguishing between *type/source of carbohydrate*. In fact, more fibre and wholegrain carbohydrates prevent the elevation in TG often associated with high total carbohydrate intake [8].

Similar to the point made in the Research Lecture on fructose, it may not be sugars *per se*, but the source and related metabolic activity of dietary sugars that determines health effects.

Relevance

Isn't it interesting when that apparently 'unreliable' arena of nutritional epidemiology produces findings that are exactly what would be predicted from 70yrs worth of tightly controlled human metabolic ward studies and intervention studies?

Take the findings in relation to SFA and PUFA; in the main analysis of the effects of each macronutrient on LDL-C, SFA exhibited twice the cholesterol-raising effects as the cholesterol-lowering effect of PUFA [3,4]. In an analysis of over 390 metabolic ward studies by Clarke *et al*. [5], the effect of replacing 5% energy from SFA with PUFA was an average 0.11mmol/L lower LDL-C level, while another analysis of RCTs showed an average reduction of 0.25mmol/L from replacing 5% SFA with PUFA [9].

This is practically what the present study found, i.e., a 5% lower LDL-C from replacing SFA with PUFA when the LDL-C level in the highest SFA group was 3.69mmol/L would be ~0.18mmol/L. Not too shabby for epidemiology, huh.

And just in case you find yourself stuck with someone who insists on hard endpoints, bear in mind intervention studies also show that this replacement of SFA with PUFA lowers risk of CVD/CHD events and mortality [9,10].

What about omega-3? The pronounced triglyceride-lowering effect of omega-3 fatty acids is precisely why they are of interest for CVD/CHD, and high-dose EPA used in intervention studies [11,12]. And bear in mind that the difference in effects on LDL-C between plant and animal sources of fat was one of the first findings in the 1950's metabolic ward research [13]. And those studies also used omega-6 rich sources of vegetable fat to lower LDL-C [13,14], another finding consistent with the present study.

You may have noticed MUFA was associated with increased LDL-C; however, there may be distinct effects of MUFA from animal sources and MUFA from plant sources [15], and the present study did not distinguish between food sources of MUFA, so I would be inclined not to read too much into that finding.

And as discussed under ***Interesting Finding***, above, the results regarding types of carbohydrates and sources of sugar are perhaps the most relevant finding in the present study, given the results for dietary fat are largely congruent with over a half-century of data.

Application to Practice

Bear in mind that the lowest and highest SFA intakes in the present study were 7% and 15%, respectively; there are many who would argue there is no additional benefit to lowering SFA given current population averages, which are ~12-13% in the UK. I respectfully differ; both the reduction in LDL-C *and* the evidence from interventions supporting lower risk of CVD/CHD would support lowering SFA in individuals who are at the higher end of population intakes. And the evidence certainly supports replacing those SFA food sources with PUFA-rich foods; vegetable oil-based spreads [e.g., Flora], vegetable cooking oils [e.g., rapeseed oil], nuts and seeds.

And of carbohydrates and sugars? In the present study, the lowest and highest intakes of free sugars were 26g [5% total energy] and 83g [18% total energy], respectively. The highest intake was thus in a range that is associated with significant increases in CVD/CHD risk [16]. But the highest non-free sugar intake was 97g [21% total energy]. The message would be to think less about "sugar" and more about food sources, total intake, and total diet, i.e., the big picture of carbohydrate intake. Recall that overall, the findings in relation to fibre and wholegrains are what we would expect also; an overall benefit to blood lipids, although of a much smaller magnitude than the effects of modifying dietary fat intake.

References

1. Ference BA, Ginsberg HN, Graham I, Ray KK, Packard CJ, Bruckert E, et al. Low-density lipoproteins cause atherosclerotic cardiovascular disease. 1. Evidence from genetic, epidemiologic, and clinical studies. A consensus statement fromthe European Atherosclerosis Society Consensus Panel. European Heart Journal. 2017;38(32):2459–72.

2. Borén J, Chapman MJ, Krauss RM, Packard CJ, Bentzon JF, Binder CJ, et al. Low-density lipoproteins cause atherosclerotic cardiovascular disease: pathophysiological, genetic, and therapeutic insights: a consensus statement from the European Atherosclerosis Society Consensus Panel. European Heart Journal. 2020;41(24):2313-2330.

3. Keys A, Anderson JT, Grande F. Prediction of Serum-Cholesterol Responses of Man to Changes in Fats in the Diet. The Lancet. 1957;Nov 16(73(7003)):959=966.

4. Hegsted MD, Ausman LM, Johnson JA, Dallal GE. Fat and Serum Lipids: The American Journal of Clinical Nutrition. 1993;57(6):875–83.

5. Clarke R, Frost C, Collins R, Appleby PN, Peto R. Dietary lipids and blood cholesterol: quantitative meta-analysis of metabolic ward studies. BMJ. 1997;314:112–7.

6. Siri-Tarino PW, Sun Q, Hu FB, Krauss RM. Saturated fatty acids and risk of coronary heart disease: Modulation by replacement nutrients. Current Atherosclerosis Reports. 2010;12(6):384–90.

7. Parks EJ. Changes in fat synthesis influenced by dietary macronutrient content. Proceedings of the Nutrition Society. 2002;61(2):281–6.

8. Anderson JW. Dietary fiber prevents carbohydrate-induced hypertriglyceridemia. Current atherosclerosis reports. 2000;2(6):536–41.

9. Mozaffarian D, Micha R, Wallace S. Effects on coronary heart disease of increasing polyunsaturated fat in place of saturated fat: A systematic review and meta-analysis of randomized controlled trials. PLoS Medicine. 2010;7(3):e1000252.

10. Hooper L, Martin N, Abdelhamid A, G DS. Reduction in saturated fat intake for cardiovascular disease. Cochrane Database of Systematic Reviews. 2015;(6):1–150.

11. Bhatt DL, Steg PG, Miller M, Brinton EA, Jacobson TA, Ketchum SB, et al. Cardiovascular risk reduction with icosapent ethyl for hypertriglyceridemia: REDUCE-IT. New England Journal of Medicine. 2019;380(1):11–22.

12. Green CJ, Pramfalk C, Charlton CA, Gunn PJ, Cornfield T, Pavlides M, et al. Hepatic de novo lipogenesis is suppressed and fat oxidation is increased by omega-3 fatty acids at the expense of glucose metabolism. BMJ Open Diabetes Research & Care. 2020;8(1):e000871.

13. Ahrens EH, Blankenhorn DH, Tsaltas TT. Effect on Human Serum Lipids of Substituting Plant for Animal Fat in Diet. Proc Soc Exp Biol Med. 1954;Aug-Sep(86(4)):872–8.

14. Beveridge JMR, Connell WF, Mayer GA, Firstbrook JB, DeWolfe MS. The Effects of Certain Vegetable and Animal Fats on the Plasma Lipids of Humans. The Journal of Nutrition. 1955;56(2):311–20.

15. Zong G, Li Y, Sampson L, Dougherty LW, Willett WC, Wanders AJ, et al. Monounsaturated fats from plant and animal sources in relation to risk of coronary heart disease among US men and women. American Journal of Clinical Nutrition. 2018;107(3):445–53.

16. Yang Q, Zhang Z, Gregg EW, Flanders W, Merritt R, Hu FB. Added sugar intake and cardiovascular diseases mortality among us adults. JAMA Internal Medicine. 2014;39:376