

Recipe for Heart Health: a Randomized Crossover Trial on Cardiometabolic Effects of Extra Virgin Olive Oil within a Whole-Food Plant-Based Vegan Diet

Andrea Krenek, RDN^{1,2}; Anne Mathews, PhD, RDN¹; Juen Guo, PhD²; Amber Courville, PhD, RDN²; Carl J. Pepine, MD³; Stephanie T. Chung, MBBS²; Monica Aggarwal, MD³

Author Affiliations

¹Food Science and Human Nutrition Department, University of Florida, Gainesville, Florida

²National Institute of Diabetes and Digestive and Kidney Diseases, National Institutes of Health, Bethesda, Maryland

³Division of Cardiology, University of Florida, Gainesville, Florida

Short Title: Recipe for Heart Health

Corresponding Author:

Monica Aggarwal, MD

Monica.aggarwal@medicine.ufl.edu

University of Florida

1329 SW 16th Street

Box 100288

Gainesville, Florida 32610-0288

Manuscript total word count: 5698 words

Clinical Perspective

What is New?

- Both inclusion and exclusion of extra virgin olive oil (EVOO) within a whole food, plant-based (WFPB) vegan diet supports cardiovascular disease risk reduction compared to a standard omnivorous pattern.
- Reduced intake of EVOO in favor of whole food fats yields increased lipid lowering than relatively greater EVOO consumption.
- Addition of EVOO after consuming low amounts within a WFPB diet may impede risk reduction.

What are the Clinical Implications?

- Consideration of optimal sources and quantity of dietary fats, including EVOO, within a risk-lowering vegan dietary pattern may support medical nutrition therapy advice among those at risk for cardiometabolic diseases.

ABSTRACT

BACKGROUND: Whole-food, plant-based (WFPB) vegan diets, low in oils, and Mediterranean diets, rich in extra virgin olive oil (EVOO) both reduce cardiovascular disease (CVD) risk factors. The optimal quantity of dietary fat, particularly EVOO, within a plant-based diet, is unclear.

METHODS: Effects of high (4 tablespoons/day) vs low (<1 teaspoon/day) EVOO intake within a WFPB diet on cardiometabolic markers were compared in adults with $\geq 5\%$ atherosclerotic CVD risk. In a randomized crossover trial with weekly cooking classes, participants followed a high to low (H2L) or low to high (L2H) EVOO WFPB diet for 4 weeks each, separated by a 1-week washout. The primary outcome was difference in low-density lipoprotein cholesterol (LDL-C) from baseline. Secondary measures were changes in additional lipid/lipoprotein, glycemic, and inflammatory markers. Linear mixed models assessed changes from baseline between each phase, with age, sex, and body weight change as covariates.

RESULTS: In 40 participants (75% female; mean[SD] body mass index, 32[7] kg/m²; age, 64[9] years), fat intake comprised 48% and 32% of total energy in the high and low EVOO phases, respectively. Both diets resulted in comparable reductions in LDL-C, total cholesterol, apolipoprotein B, HDL-C, glucose, and high-sensitivity C-reactive protein (all $P < 0.05$). With diet-sequence interactions for LDL-C ($P = 0.003$), differences were detected between diets in the context of diet order (mean[SEM] high-to-low: $\Delta -12.7[5.9]$ mg/dl, $P = 0.04$ vs low-to-high: $\Delta +15.8[6.8]$ mg/dl, $P = 0.02$). Similarly, the L2H order led to increased glucose, total cholesterol, and HDL-C (all $P < 0.05$). Over the first period, LDL-C reductions were $-25.5(5.1)$ mg/dl after low vs $-16.7(4.2)$ mg/dl after high EVOO,

$P=0.162$, which were diminished over the second period (-4.0[4.3] vs -9.7[5.1] mg/dl, $P=0.382$).

CONCLUSIONS: Both plant-based diet patterns improved cardiometabolic risk profiles compared to baseline diets, with more pronounced decreases in LDL-C after the low EVOO diet. Addition of EVOO after following a low intake pattern may impede further lipid reductions.

REGISTRATION: ClinicalTrials.gov Identifier: [NCT04828447](https://clinicaltrials.gov/ct2/show/study/NCT04828447)

INTRODUCTION

Poor diet quality is a leading risk factor for cardiometabolic deaths worldwide.^{1,2} Plant-based diets (PBDs), including Mediterranean Diets (MedDiets) and vegetarian/vegan diets, are recommended for cardiovascular disease (CVD) risk reduction. However, there is continuing debate on the optimal composition and quality of PBDs that confer the greatest cardioprotective benefits.^{3–8} Specifically, the amount of dietary fat in the form of vegetable oils versus whole food sources from which oils are derived (such as olives, nuts, seeds, and avocados) remains controversial.

Vegan diets are comprised of exclusively plant foods without animal products; however, the food composition and quality may vary widely. Low-fat, whole-food plant-based diets (WFPBD) emphasize foods in their whole, unrefined form and minimize heavily processed foods, refined grains, added sugars, and oils while excluding animal foods. In these patterns, dietary fat is often advised as <10-15% of energy intake.⁹ MedDiets also emphasize a foundation of plant-based foods but additionally recommend unrestricted extra virgin olive oil (EVOO) – frequently advised as at least 4 tablespoons (~52 grams) daily – as a central source of dietary fat, with moderate consumption of seafood, poultry, and dairy products and low intake of red meat and animal-derived fats. Dietary fat composition in these patterns can be up to 35-40% of energy intake.^{10–12}

Importantly, both low-fat vegan diets and MedDiets have shown improvements in cardiometabolic risk factors^{7,13–16} and lipid-lowering effects. However, in the large Primary Prevention of Cardiovascular Disease with a Mediterranean Diet Supplemented with Extra-Virgin Olive Oil or Nuts (PREDIMED) study, no overall effects were reported on low-density lipoprotein cholesterol (LDL-C) or total cholesterol for those supplemented with

EVOO compared to the control diet.¹⁷ Further, in a post-hoc analysis of PREDIMED¹⁸, higher conformity to a pro-vegetarian food pattern was associated with a 41% lower mortality. Studies have been conflicting on the benefits of individual components of MedDiets¹⁹⁻²¹, and it is unclear whether EVOO confers LDL-C lowering when consuming a WFPBD at varying CVD risk.

The aim of the Recipe for Heart Health (RFHH) trial was to compare the effects of consuming a high versus low amount of EVOO within a WFPB vegan diet pattern on LDL-C and other cardiometabolic markers. We hypothesized that the WFPBD would lower LDL-C and that there would be a greater change from baseline in LDL-C while consuming the low EVOO WFPBD.

METHODS

The RFHH study was conducted in accordance with ethical standards from the Helsinki Declaration, and it was approved by the University of Florida Institutional Review Board (IRB202002194). All study participants provided written informed consent prior to any study procedures.

Study Design

RFHH was a prospective, open-label, controlled crossover trial (NCT04828447) in adults assigned to follow one of two diet sequences in random order: 4 weeks of a WFPBD high (4 tablespoons (TB) per day) or low (<1 teaspoon (tsp) per day) in uncooked EVOO, followed by 1-week washout before switching to the alternate diet for an additional 4 weeks (eFigure 1a in Supplement 1). Randomization of diet order (High → Low EVOO [H2L] or Low → High EVOO [L2H]) was performed by an independent statistician by

computer generated sequencing. Participants were masked to randomization order until after baseline measures were completed. Six cohorts participated over an 11-month period (May 2021-April 2022). Each cohort attended weekly virtual group culinary medicine cooking classes led by a dietitian/chef during both diet intervention periods. Metabolic, clinical, behavioral, and dietary data were collected before and after each intervention period for a total of four clinical evaluations after study enrollment.

Participants

Adults 18-79 years at borderline to high risk for atherosclerotic cardiovascular disease (ASCVD) according to the ACC/AHA ASCVD Risk Calculator (10-year risk score [>40 years] or lifestyle ASCVD risk [if younger than 40 years] of $\geq 5\%$) were recruited. Participants were excluded if they were secondary prevention patients (defined as history of coronary artery disease with $>50\%$ stenosis, myocardial infarction, or coronary artery calcium score >400 AU), currently or planning to become pregnant, reported they were already following a Mediterranean or vegan food profile that excluded meat, poultry, or fish, or had known warfarin use, end-stage heart failure, active malignancy, end-stage renal disease, HIV, or LDL-C levels ≥ 190 mg/dL.

Dietary Intervention

In addition to weekly gift cards to support grocery purchases, all EVOO was provided, and participants were instructed to consume it only in its raw state with prescribed amounts of 4 TB daily (high EVOO) or as close as possible to no added EVOO (<1 tsp during the low EVOO period). The WFPBD was characterized by abstinence from consumption of animal products, including meat, poultry, fish, seafood, dairy, and eggs, along with heavily processed or refined food items containing refined grains, added sugars, or oils. Foods within categories of whole fruits, vegetables, legumes, whole grains, and nuts/seeds were emphasized. Vitamin B12 was supplemented (≥ 100 mcg/d) as a nutrient not adequately consumed within vegan diets. While nutritionally balanced meals were encouraged, neither specific portion sizes nor controlled caloric or macronutrient composition was implemented. Participants were advised to eat according to individual hunger and fullness cues.

All participants met with the research dietitian/chef in a one-hour virtual session to describe the study diet as well as discuss individualized meal planning and cooking classes before the first diet intervention. Diet fidelity was promoted through ready access to research dietitians and using a comprehensive teaching kitchen/culinary medicine intervention program. Description of the study cookbook and protocol is reported in the Supplemental materials.

Outcomes

The primary outcome was change in LDL-C levels from baseline and between diet periods. Exploratory outcomes were change from baseline post intervention periods in other cardiometabolic biomarkers (blood lipids, glycemic measures, and inflammatory markers), blood pressure, and anthropometrics.

Diet Intake

Diet intake was assessed using the Automated Self-Administered 24-hour (ASA-24) (National Cancer Institute, Bethesda, MD, USA) Dietary Assessment Tool.²² Participants completed 4-7 recalls over a 1–2-week period prior to the study and during each intervention period. Compliance and diet intake were affirmed through verbal weekly check-ins during group cooking classes. Quantitative assessments of dietary intake were collected before and after each diet via trimethylamine-N-oxide (TMAO, reflecting foods from animal sources), and skin carotenoid status²³ (fruit and vegetable intake), reported elsewhere. Participants were asked not to alter exercise habits during the study or make any changes to medications other than as directed by their physicians.

Metabolic and anthropometric measurements

After a 10-12 hour overnight fast, blood and urine were collected at baseline and at the end of each diet period. Fasting lipid profile, lipoprotein (a), apolipoprotein B, glucose, fructosamine, interleukin-6 (IL-6), and high sensitivity C-reactive protein (hsCRP) were analyzed by standard methodologies at the UF Heart Health and Vascular Hospital (Gainesville, Florida). LDL-C was calculated using the Friedwald equation at the University of Florida's core lab. TMAO was measured by liquid chromatography with tandem mass spectrometry (LC-MS/MS) through Cleveland HeartLab (Cleveland, OH).²⁴

All clinic evaluations included measurements of body weight, blood pressure, and heart rate. Waist and hip circumference were measured according to World Health Organization protocols as the midpoint between the lower rib margin and top of the iliac crest (waist) and the maximum circumference (hip).²⁵

Statistical analysis

Power calculations and statistical analyses were performed using SAS 9.4 (Cary, NC) and IBM SPSS Statistics 29.0. Study power calculations were informed by previous crossover trials.²⁶⁻²⁹ To detect a mean difference of LDL-C of 0.30 mmol/L (12 mg/dL) with a $SD \pm 20$ mg/dL, and the assumption that within-subject LDL-C level correlations were 0.3, a total of 40 patients would yield a power of 0.8 and a type 1 error probability of 0.05.

Linear mixed models were used to assess changes from baseline between each phase, including age, sex, and body weight change as covariates. This model allowed for testing differences in diets, order, and their interaction for LDL-C. Individuals were treated as random, while intervention, period, and sequence were treated as fixed effects.

Separate mixed-effects models were used for secondary cardiometabolic outcomes including remaining fasting lipids, TMAO, glycemic measures, inflammatory biomarkers, and blood pressure.

RESULTS

Screening and enrollment began in April 2021, and the last participant visit was completed in May 2022. Of 395 participants assessed for eligibility, 60 attended baseline visits and were randomized to follow one of the two diet sequence interventions for which 40 individuals completed (eFigure 1b in Supplement 1).

Demographics

Thirty female and ten male adults, mean (SD) age 64.4 (8.6) years, 73% non-Hispanic white, BMI 32 (7) kg/m², completed the study. Two participants were <40 years old and thus qualified based on lifetime risk. No significant differences were observed in baseline anthropometric or metabolic characteristics of participants (Table 1 and eTable 1 in Supplement 1). Most participants had obtained at least a college degree (65%) and were primarily responsible for cooking meals (80%).

Circulating Metabolites and Clinical Data

For the primary outcome of change in LDL-C, both diets resulted in decreased levels compared to baseline with no significant differences by group when considering both diet periods (Figure 1a, $P=0.722$). eFigure 8 visualizes mean LDL-C levels by timepoint. With diet-sequence interactions ($P=0.003$), sensitivity analyses revealed that clinically larger LDL-C reductions were observed after the low EVOO diet (mean [SEM] -25.5 [5.1] mg/dl) vs the high EVOO diet (-16.7 [4.2] mg/dl), $p=0.162$ (eTable 2 in

Supplement 1), which were diminished over the second 4 weeks. Similarly, the low EVOO diet yielded greater decreases in total cholesterol (-33.8 [5.6] vs -19.0 [4.6] mg/dl, $p=0.035$), HDL-C (10.5 [2.0] vs -5.0 [1.6] mg/dl, $p=0.025$), apolipoprotein B (-14.8 [3.8] vs -5.5 [3.2] mg/dl, $p=0.053$), and glucose (-17.8 [4.2] vs -8.8 [3.4] mg/dl, $p=0.082$) over period 1 (eFigure 2 in Supplement 1). Differences in LDL-C between diets were detected in the context of which diet was followed first (diet order H2L: Δ -12.7 [5.9] mg/dl, $P=0.04$ vs L2H: Δ +15.8 [6.8] mg/dl, $P=0.02$), Figure 1b. The L2H order also led to increased glucose (Δ +15.3 [5.1] mg/dl, $P=0.004$), total cholesterol (Δ +29.4 [7.4] mg/dl, $P=0.0002$), and HDL-C (Δ +9.8 [2.3] mg/dl, $P<0.0001$), Figure 2. In addition to LDL-C, both diets combined resulted in significant reductions in total cholesterol, apolipoprotein B, HDL-C, glucose, and hs-CRP compared to baseline (eTable 3 in Supplement 1). Table 2 summarizes the overall comparison of the high and low EVOO diets as well as changes in fasting blood measurements by diet sequence order (H2L and L2H).

Anthropometrics

Body weight decreased after both the high and low EVOO diets from 89.0 [3.5] kg to 83.8 [2.8] and 83.1 [2.7], ($P<0.001$), respectively, as shown in eTable 4 and by timepoint in eFigure 9 in Supplement 1. The low EVOO period resulted in greater weight loss compared to the high EVOO diet (mean difference 0.7 [0.3] kg, $p=0.011$).

Dietary Intake

Fat intake comprised 48% and 32% of total energy in the high and low EVOO phases, respectively. eFigures 3-7 in Supplement 1 presents selected nutrient intakes

during each time point for total energy, fat, fiber, sodium, and added sugars. Additional results are available in eTable 6 in Supplement 1.

DISCUSSION

This study was designed to compare effects of consuming a high versus low amount of EVOO within a vegan WFPBD on cardiometabolic risk factors in adults at borderline to high ASCVD risk. Both high and low EVOO WFPBDs led to reduced LDL-C compared to baseline levels, with no differences by group, though diet-sequence interactions were detected. Reductions in LDL-C were observed despite total fat intake comprising 48% and 32% of total energy during the high and low EVOO phases, respectively. While general dietary guidelines recommend approximately 20-35% of total energy from fat, MedDiet patterns tend to be comprised of 35-40% of dietary fat.^{30,31} Our study results corroborate the positive health impact of minimizing most saturated fatty acids, several of which have been shown to downregulate hepatic LDL receptor activity, leading to decreased clearance and increased levels of circulating LDL particles.³² Conversely, the effects of mono- and poly-unsaturated fatty acids are mediated by enhancing LDL receptor activity and thus increasing clearance from circulation.³² While EVOO comprised most of dietary fat during the high EVOO phase, sources during the low EVOO were primarily in the form of unrefined whole plant-based fats (e.g. avocados, nuts, seeds, olives), which retain inherent dietary fiber and intact phytochemicals. Slightly greater intake of saturated fat during the high EVOO period may have contributed towards LDL differences between diet periods. Well-known cholesterol-lowering effects of dietary fiber and potential benefits of phytochemicals in unprocessed forms on cardiometabolic markers may also have supported superior LDL-C decreases during the low EVOO

period. Related to additional nutrient considerations, 42% (high EVOO) and 56% (low EVOO) of energy was derived from carbohydrates, and there were no energy differences between baseline and the high EVOO period. In our study, reduction of low-quality refined carbohydrate sources with concomitant increases in dietary fiber may have yielded benefits. In both groups, intake of dietary fiber increased by ~8-9 g/d compared to baseline estimates. Every gram of soluble fiber could result in up to 1 mg/dL reduction in LDL-C concentrations³³, and every 1 mmol/L (38.67 mg/dl) reduction in LDL cholesterol is associated with reducing risk of major vascular events by at least 25%, exerting effects by upregulation of LDL receptor expression.³⁴

While greater lipid lowering effects occurred with low EVOO intake, results indicated benefits with both diets from baseline intake. Existing support for EVOO intake is based largely on observational analyses in MedDiets that consider its use to replace primarily animal-derived dietary fats that may increase risks for dyslipidemia, including dairy, butter, mayonnaise, lard, and animal meats.³⁵⁻³⁷ Lower risk of CVD mortality with EVOO consumption are not observed when replacing other vegetable oils³⁸, suggesting most benefit when substituting EVOO for other aforementioned animal-based dietary fats, with mixed effects on LDL-C.³⁹ Additional investigation of whole olive consumption in analysis of the PREDIMED study further found a 25% decrease in risk of major cardiovascular events in the highest tertile of baseline consumption, perhaps highlighting benefits of other healthful fat sources within a diet pattern as opposed to solely EVOO intake. The Recipe for Heart Health study may help clarify dose response effects of EVOO on LDL-C within a WFPBD.

Due to carryover effects, we also determined treatment effects by diet order and period, which may be viewed as potentially more valid. Evaluating differences between groups in the context of diet order (H2L vs L2H) lends notable insight around optimal quantity of EVOO within a WFPBD. Our analysis indicated LDL-C lowering was more robust with low EVOO, while addition of EVOO after consuming a diet with lower intake hindered benefit. Nearly all metabolic biomarkers demonstrated similar responses to diet order when comparing the mean difference of change from baseline between groups: addition of EVOO after following a WFPBD with low intake (L2H) led to increases in lipids, glycemic measures, and hs-CRP. In contrast, removal of EVOO after following a diet with high intake (H2L) led to decreases in these measures, suggesting that low EVOO intake may be more optimal for lowering CVD risk than high EVOO intake within this pattern. Analysis by diet order for secondary metabolic outcomes indicated a similar pattern for HDL-C, Lp(a), and glucose as for LDL-C and remaining markers.

Though not statistically significant, the greater decrease observed in LDL-C following the low EVOO diet compared to high intake over the first 4-week diet period may be clinically meaningful. Greater body weight reduction, included as a covariate, coincided with lower EVOO consumption. The low EVOO diet also led to superior improvements in cardiometabolic profile as indicated by lower total cholesterol, apolipoprotein B, glucose, and hs-CRP. Although both diets improved the metabolic phenotype, low EVOO intervention may provide superior LDL-C lowering in individuals at highest risk. Future studies are needed to determine if these short-term effects are sustainable and translate to improvements in cardiac outcomes. Sensitivity analyses that accounted for carryover effects suggested that low EVOO additionally resulted in greatest

HDL-C reductions, though the clinical translation of this finding is beyond the scope of this study as emerging data suggest HDL-C functionality may be of greater importance, with potential U-shaped associations between HDL-C and CVD risks.^{40–42}

Strengths and Limitations

Notably, this rigorous diet intervention study had low attrition, high adherence, and comprehensive metabolic profiling with great interest in the educational programming and community building. In a novel randomized crossover design utilizing a culinary approach, we evaluated an underexplored, debated research question.

Study limitations include diet-sequence interactions (carry-over effects) despite wash-out periods. A relatively short study period may also have precluded subsequent metabolic changes that may become apparent with high and low EVOO intake over time. As uncontrolled macronutrient and energy distribution lends only speculative insight on the influence of EVOO or dietary fat on appetite and intake of other foods, future investigations may aim to match total calorie and nutrient composition to delineate potential differential impacts of oil compared to whole food fat consumption. As our study sample consisted of primarily well-educated, white women, whether results would differ in a more diverse sample is unknown, limiting generalizability. While inclusion of body weight change aimed to account for differences between groups, it is possible that statistical analyses were unable to fully adjust for this difference between diets. This study was additionally conducted during the COVID-19 pandemic, which likely affected study recruitment and participation.

Although diet recalls indicated high levels of adherence and acceptability, and were consistent with TMAO indicators of adherence, self-reported dietary assessments

are prone to recall bias. In this study, substantial burden using the ASA-24 site and lack of food options to fully capture the dietary pattern (e.g., tempeh, seitan, varieties of legumes, non-dairy products, nutritional yeast, nutrient dense “cookies” without added sugars, meatless sloppy joes) could have contributed to reporting bias.

Conclusions

Recipe for Heart Health affirmed that a WFPBD with high and low EVOO levels improved LDL cholesterol and atherogenic lipid profile in participants at risk for ASCVD. Dietary changes resulted in lower cardiovascular risk factors compared to baseline levels, with a greater difference in optimal directions following the transition to a low EVOO diet, suggesting that EVOO may not be the beneficial additive of a Mediterranean diet. Addition of EVOO after following a low EVOO pattern could impede further LDL reductions.

Acknowledgements

Corresponding Author: Monica Aggarwal, MD, University of Florida, 1329 SW 16th Street, Box 100288, Gainesville, Florida 32610-0288
(Monica.aggarwal@medicine.ufl.edu)

Author Contributions:

Concept and design: Krenek, Mathews, Aggarwal.

Acquisition, analysis, or interpretation of data: All authors.

Drafting of the manuscript: Krenek.

Critical review of the manuscript for important intellectual content: Krenek, Mathews, Courville, Pepine, Chung, Aggarwal.

Statistical analysis: Guo, Krenek, Courville, Chung.

Obtained funding: Aggarwal.

Administrative, technical, or material support: Krenek, Mathews.

Supervision: Chung, Courville, Mathews, Aggarwal.

Conflict of Interest Disclosures: Dr. Aggarwal reported support by Flourish Research, Inc., speaking engagement honoraria, and book royalties, outside the submitted work. Dr. Chung is supported by the Intramural Research Program of the NIH, The National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), outside the submitted work. No other disclosures were reported for work presented in this manuscript.

Funding/Support: Funding support for this study was provided by the Purjes Foundation, Dr. Monica Aggarwal Prevention Foundation, and University of Florida Food Science and Human Nutrition Department.

Role of Funder/Sponsor: The funders had no role in the design and conduct of the study; collection, management, analysis, and interpretation of data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

Additional Contributions: We are immensely grateful to our study participants for their devoted time and commitment to this trial. Additionally, we thank the medical staff, research coordinators, and clinical research team nurses at University of Florida Division of Cardiovascular Medicine for their support in recruitment and data collection.

REFERENCES

1. Afshin A, Sur PJ, Fay KA, et al. Health effects of dietary risks in 195 countries, 1990–2017: a systematic analysis for the Global Burden of Disease Study 2017. *The Lancet*. 2019;393(10184):1958-1972. doi:10.1016/S0140-6736(19)30041-8
2. Micha R, Peñalvo JL, Cudhea F, Imamura F, Rehm CD, Mozaffarian D. Association between dietary factors and mortality from heart disease, stroke, and type 2 diabetes in the United States. *JAMA - Journal of the American Medical Association*. 2017;317(9):912-924. doi:10.1001/jama.2017.0947
3. Salvador AM, García-Maldonado E, Gallego-Narbón A, Zapatera B, Vaquero MP. Fatty acid profile and cardiometabolic markers in relation with diet type and omega-3 supplementation in Spanish vegetarians. *Nutrients*. 2019;11(7). doi:10.3390/nu11071659
4. Yokoyama Y, Nishimura K, Barnard ND, et al. Vegetarian diets and blood pressure ameta-analysis. *JAMA Intern Med*. 2014;174(4):577-587. doi:10.1001/jamainternmed.2013.14547
5. Vigiouliouk E, Kendall CW, Kahleová H, et al. Effect of vegetarian dietary patterns on cardiometabolic risk factors in diabetes: A systematic review and meta-analysis of randomized controlled trials. *Clinical Nutrition*. 2019;38(3):1133-1145. doi:10.1016/j.clnu.2018.05.032
6. Satija A, Hu FB. Plant-based diets and cardiovascular health. *Trends Cardiovasc Med*. 2018;28(7):437-441. doi:10.1016/j.tcm.2018.02.004
7. Kahleova H, Levin S, Barnard N. Cardio-metabolic benefits of plant-based diets. *Nutrients*. 2017;9(8):1-13. doi:10.3390/nu9080848
8. Dinu M, Abbate R, Gensini GF, Casini A, Sofi F. Vegetarian, vegan diets and multiple health outcomes: A systematic review with meta-analysis of observational studies. *Crit Rev Food Sci Nutr*. 2017;57(17):3640-3649. doi:10.1080/10408398.2016.1138447
9. Hever J, Cronise RJ. Plant-based nutrition for healthcare professionals: Implementing diet as a primary modality in the prevention and treatment of chronic disease. *Journal of Geriatric Cardiology*. 2017;14(5):355-368. doi:10.11909/j.issn.1671-5411.2017.05.012
10. Estruch R, Lamuela-Raventós RM, Ros E. The Bitter Taste of Extra Virgin Olive Oil for a Sweet Long Life. *J Am Coll Cardiol*. 2020;75(15):1740-1742. doi:10.1016/j.jacc.2020.02.043

11. Marcelino G, Hiane PA, Freitas K de C, et al. Effects of olive oil and its minor components on cardiovascular diseases, inflammation, and gut microbiota. *Nutrients*. 2019;11(8). doi:10.3390/nu11081826
12. Freeman AM, Morris PB, Barnard N, et al. Trending Cardiovascular Nutrition Controversies. *J Am Coll Cardiol*. 2017;69(9):1172-1187. doi:10.1016/j.jacc.2016.10.086
13. Barnard ND, Cohen J, Jenkins DJA, et al. A low-fat vegan diet improves glycemic control and cardiovascular risk factors in a randomized clinical trial in individuals with type 2 diabetes. *Diabetes Care*. 2006;29(8):1777-1783. doi:10.2337/dc06-0606
14. Ornish D, Scherwitz LW, Billings JH, et al. Intensive Lifestyle Changes for Reversal of Coronary Heart Disease. *JAMA*. 1998;280(23):2001-2007. doi:10.1001/JAMA.280.23.2001
15. Ornish D, Brown SE, Billings JH, et al. Can lifestyle changes reverse coronary heart disease?: The Lifestyle Heart Trial. *The Lancet*. 1990;336(8708):129-133. doi:10.1016/0140-6736(90)91656-U
16. Friedman SM, Barnett CH, Franki R, Pollock B, Garver B, Barnett TD. Jumpstarting Health With a 15-Day Whole-Food Plant-Based Program. *Am J Lifestyle Med*. 2022;16(3):374-381. doi:10.1177/15598276211006349
17. Estruch R, Ngel Martínez-González MA, Corella D, et al. Effects of a Mediterranean-Style Diet on Cardiovascular Risk Factors A Randomized Trial. Published online 2006.
18. Martínez-González MA, Sánchez-Tainta A, Corella D, et al. A provegetarian food pattern and reduction in total mortality in the Prevención con Dieta Mediterránea (PREDIMED) study. *Am J Clin Nutr*. 2014;100(suppl_1):320S-328S. doi:10.3945/ajcn.113.071431
19. Tome-Carneiro J, Crespo MC, de las Hazas MCL, Visioli F, Davalos A. Olive oil consumption and its repercussions on lipid metabolism. *Nutr Rev*. 2020;78(11):952-968. doi:10.1093/nutrit/nuaa014
20. Fernandes J, Fialho M, Santos R, et al. Is olive oil good for you? A systematic review and meta-analysis on anti-inflammatory benefits from regular dietary intake. *Nutrition*. 2020;69:110559. doi:10.1016/j.nut.2019.110559
21. Guasch-Ferré M, Liu G, Li Y, et al. Olive Oil Consumption and Cardiovascular Risk in U.S. Adults. *J Am Coll Cardiol*. 2020;75(15):1729-1739. doi:10.1016/j.jacc.2020.02.036
22. ASA24® Dietary Assessment Tool | EGRP/DCCPS/NCI/NIH. Accessed June 23, 2021. <https://epi.grants.cancer.gov/asa24/>

23. Pitts SBJ, Jahns L, Wu Q, et al. A non-invasive assessment of skin carotenoid status through reflection spectroscopy is a feasible, reliable and potentially valid measure of fruit and vegetable consumption in a diverse community sample. *Public Health Nutr.* 2018;21(9):1664-1670. doi:10.1017/S136898001700430X
24. Tang WHW, Wang Z, Levison BS, et al. Intestinal Microbial Metabolism of Phosphatidylcholine and Cardiovascular Risk. <http://dx.doi.org/10.1056/NEJMoa1109400>. 2013;368(17):1575-1584. doi:10.1056/NEJMoa1109400
25. Waist circumference and waist-hip ratio: report of a WHO expert consultation. Accessed June 25, 2021. <https://www.who.int/publications/i/item/9789241501491>
26. Damasceno NRT, Pérez-Heras A, Serra M, et al. Crossover study of diets enriched with virgin olive oil, walnuts or almonds. Effects on lipids and other cardiovascular risk markers *. *NUMECD.* 2010;21:S14-S20. doi:10.1016/j.numecd.2010.12.006
27. Djekic D, Shi L, Brolin H, et al. Effects of a Vegetarian Diet on Cardiometabolic Risk Factors, Gut Microbiota, and Plasma Metabolome in Subjects With Ischemic Heart Disease: A Randomized, Crossover Study. *J Am Heart Assoc.* 2020;9(18):e016518. doi:10.1161/JAHA.120.016518
28. Hall KD, Guo J, Courville AB, et al. Effect of a plant-based, low-fat diet versus an animal-based, ketogenic diet on ad libitum energy intake. *Nat Med.* 2021;27(2):344-353. doi:10.1038/s41591-020-01209-1
29. Crimarco A, Springfield S, Petlura C, et al. A randomized crossover trial on the effect of plant-based compared with animal-based meat on trimethylamine-N-oxide and cardiovascular disease risk factors in generally healthy adults: Study With Appetizing Plantfood—Meat Eating Alternative Trial (SWAP-ME. *Am J Clin Nutr.* 2020;(7):1-12. doi:10.1093/ajcn/nqaa203
30. Álvarez-Pérez J, Sánchez-Villegas A, Díaz-Benítez EM, et al. Influence of a Mediterranean Dietary Pattern on Body Fat Distribution: Results of the PREDIMED–Canarias Intervention Randomized Trial. *J Am Coll Nutr.* 2016;35(6). doi:10.1080/07315724.2015.1102102
31. Davis C, Bryan J, Hodgson J, Murphy K. Definition of the mediterranean diet: A literature review. *Nutrients.* 2015;7(11). doi:10.3390/nu7115459
32. Feingold KR. *The Effect of Diet on Cardiovascular Disease and Lipid and Lipoprotein Levels.*; 2000.
33. Brown L, Rosner B, Willett WW, Sacks FM. Cholesterol-lowering effects of dietary fiber: A meta-analysis. *American Journal of Clinical Nutrition.* 1999;69(1). doi:10.1093/ajcn/69.1.30

34. Silverman MG, Ference BA, Im K, et al. Association Between Lowering LDL-C and Cardiovascular Risk Reduction Among Different Therapeutic Interventions: A Systematic Review and Meta-analysis. *JAMA*. 2016;316(12):1289-1297. doi:10.1001/JAMA.2016.13985
35. Sikand G, Severson T. Top 10 dietary strategies for atherosclerotic cardiovascular risk reduction. *Am J Prev Cardiol*. 2020;4:100106. doi:10.1016/j.ajpc.2020.100106
36. Belardo D, Michos ED, Blankstein R, et al. Practical, Evidence-Based Approaches to Nutritional Modifications to Reduce Atherosclerotic Cardiovascular Disease: An American Society For Preventive Cardiology Clinical Practice Statement. *Am J Prev Cardiol*. 2022;10:100323. doi:10.1016/J.AJPC.2022.100323
37. Arnett DK, Blumenthal RS, Albert MA, et al. 2019 ACC/AHA Guideline on the Primary Prevention of Cardiovascular Disease: A Report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. *Circulation*. 2019;140(11):e596-e646. doi:10.1161/CIR.0000000000000678
38. Guasch-Ferré M, Li Y, Willett WC, et al. Consumption of Olive Oil and Risk of Total and Cause-Specific Mortality Among U.S. Adults. *J Am Coll Cardiol*. 2022;79(2):101-112. doi:10.1016/J.JACC.2021.10.041
39. Schwingshackl L, Krause M, Schmucker C, Hoffmann G, Rucker G, Meerpohl JJ. Impact of different types of olive oil on cardiovascular risk factors: A systematic review and network meta-analysis. *Nutrition, Metabolism and Cardiovascular Diseases*. 2019;29(10):1030-1039. doi:10.1016/j.numecd.2019.07.001
40. Kosmas CE, Martinez I, Surlas A, et al. High-density lipoprotein (HDL) functionality and its relevance to atherosclerotic cardiovascular disease. *Drugs Context*. 2018;7. doi:10.7573/DIC.212525
41. Barter PJ, Rye KA. HDL cholesterol concentration or HDL function: which matters? Published online 2017. doi:10.1093/eurheartj/ehx163
42. Li H, Qian F, Zuo Y, et al. U-Shaped Relationship of High-Density Lipoprotein Cholesterol and Incidence of Total, Ischemic and Hemorrhagic Stroke: A Prospective Cohort Study. *Stroke*. 2022;53(5):1624-1632. doi:10.1161/STROKEAHA.121.034393

Figure Legends

Figure 1: Change in LDL-C level from baseline after the high and low EVOO diets by each 4-week period (a) and by sequence of diet randomization (b). Values are mean+SEM. *P<0.05, **P<0.01, ***P<0.001. Linear mixed models adjusted for age, sex, and body weight change were used for analyses. P values indicate diet effects of the respective intervention in (a) and represent the difference between the high and low EVOO groups in the change in LDL-C from baseline in (b). Abbreviations: H2L, high to low EVOO diet order; L2H, low to high EVOO diet order; LDL-C, low-density lipoprotein cholesterol; EVOO, extra virgin olive oil; mg/dl, milligrams per deciliter.

Figure 2. Change in secondary cardiometabolic outcomes from baseline comparing the high and low EVOO vegan diets by sequence of diet randomization. Values are mean+SEM. *P<0.05, **P<0.01, ***P<0.001. Linear mixed models adjusted for age, sex, and body weight change was used for analyses. P values represent the difference between the high and low EVOO groups in the change in LDL-C from baseline. Abbreviations: HDL-C, high-density lipoprotein cholesterol; apoB, apolipoprotein B; Lp(a), lipoprotein(a); hs-CRP, high-sensitivity C-reactive protein; IL-6, interleukin6; TMAO, trimethylamine N-oxide; H2L, high to low EVOO diet order; L2H, low to high EVOO diet order.

Table 1. Baseline Characteristics of Recipe for Heart Health Study Population by Randomization.

Characteristic	All (n=40)	High EVOO (n=22)	Low EVOO (n=18)
Age, years	64.4 (8.6)	65.5 (6.3)	63.0 (10.9)
Sex, female, n (%)	30 (75%)	14 (64%)	16 (89%)
BMI, kg/m ²	30.7 (7.0)	30.7 (7.0)	30.2 (5.1)
Diabetes diagnosis, n (%)	12 (30%)	4 (18.2%)	8 (44.4%)
Race/ethnicity, n (%)			
African American or Black	9 (22.5%)	5 (22.7%)	4 (22.2%)
Asian or Pacific Islander	1 (2.5%)	0 (0%)	1 (5.5%)
Hispanic/LatinX	1 (2.5%)	1 (4.5%)	0 (0%)
Non-Hispanic White	29 (72.5%)	15 (68.1%)	14 (77.7%)
Other	2 (5.0%)	2 (9.0%)	0 (0%)
Highest level of education achieved, n (%)			
High school degree	1 (2.5%)	0 (0%)	1 (5.6%)
Some college	13 (32.5%)	7 (31.8%)	6 (33.3%)
College degree	11 (27.5%)	6 (27.3%)	5 (27.8%)
Some post-graduate degree	2 (5.0%)	1 (4.5%)	1 (5.6%)
Post-graduate degree	13 (32.5%)	8 (36.4%)	5 (27.8%)
Income, n (%)			
<\$10,000	0 (0%)	0 (0%)	0 (0%)
\$10,000-25,000	3 (7.5%)	0 (0%)	3 (16.7%)
\$25,000-50,000	6 (15%)	5 (22.7%)	1 (5.6%)
\$50,000-75,000	5 (12.5%)	2 (9.1%)	3 (16.7%)
\$75,000-100,000	7 (17.5%)	3 (13.6%)	4 (22.2%)
\$100,000-150,000	6 (15%)	5 (22.7%)	1 (5.6%)
\$>150,000	10 (25%)	7 (31.8%)	3 (16.7%)
Prefer not to answer	3 (7.5%)	0 (0%)	3 (16.7%)
Medication, n (%)			
Lipid-lowering drug	21 (52.5%)	13 (59.1%)	8 (44.4%)
Antihypertensive	24 (60.0%)	12 (54.5%)	12 (66.7%)
Aspirin/other antithrombotic drug	13 (32.5%)	8 (36.4%)	5 (27.8%)
Antiarrhythmic drug	1 (2.5%)	1 (4.5%)	0 (0%)
Insulin	1 (2.5%)	0 (0%)	1 (5.6%)
Oral hypoglycemic drug	9 (22.5%)	2 (9.1%)	7 (38.9%)
Smoking status, n (%)			
Currently smoke	0 (0%)	0 (0%)	0 (0%)
Previously used tobacco	12 (30%)	7 (31.8%)	5 (27.8%)

Data are presented as mean (SD) or n(%).

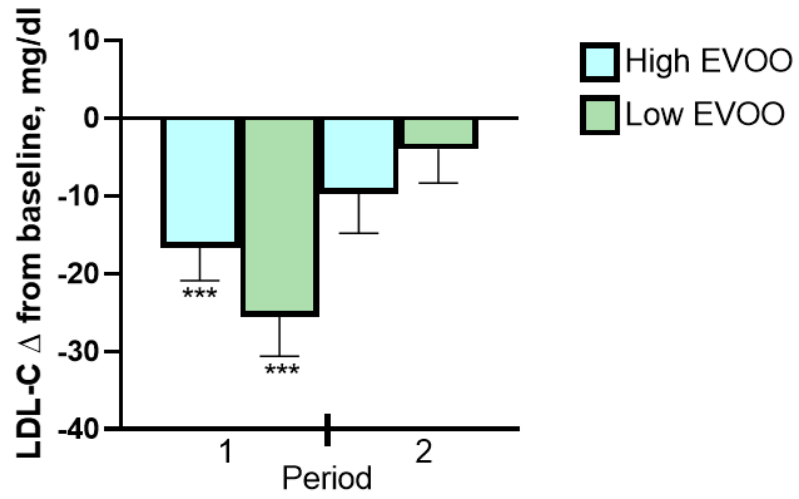
Table 2. Changes in Metabolic Outcomes Comparing the High and Low EVOO Vegan Diets by Sequence of Diet Randomization.

	ΔH2L order	P-value	ΔL2H order	P-value	Δ High vs Low EVOO overall	P-value High vs Low EVOO overall
Total cholesterol, mg/dl	-14.0 (6.5)	0.034	+29.4 (7.4)	0.0002	+7.7 (4.8)	0.115
LDL cholesterol, mg/dl	-12.7 (5.9)	0.036	+15.8 (6.8)	0.022	+1.6 (4.4)	0.722
HDL cholesterol, mg/dl	-1.4 (1.9)	0.488	+9.8 (2.3)	<0.0001	+4.2 (1.4)	0.006
Triglycerides, mg/dl	-0.3 (11.4)	0.982	+20.4 (13.0)	0.121	+10.1 (8.4)	0.236
Apolipoprotein B, mg/dl	-4.1 (4.8)	0.396	+7.4 (5.1)	0.150	+1.7 (3.4)	0.622
Lipoprotein(a), mg/dl	-17.1 (5.5)	0.005	+4.1 (6.6)	0.541	-6.5 (4.2)	0.137
Fructosamine, umol/l	-3.5 (5.6)	0.529	+0.8 (6.4)	0.904	-1.4 (4.1)	0.739
Glucose, mg/dl	-1.9 (4.4)	0.664	+15.3 (5.1)	0.004	+6.7 (3.2)	0.045
Interleukin-6, ng/ml	+0.2 (0.6)	0.768	-1.2 (0.7)	0.098	-0.5 (0.5)	0.272
hs-CRP, mg/l	-0.8 (0.7)	0.261	+1.5 (0.9)	0.078	+0.3 (0.6)	0.534
TMAO, uM	-3.6 (1.6)	0.045	+0.1 (2.0)	0.961	-1.7 (1.2)	0.178

Data represent mean (SEM), showing difference between diets for the change from baseline comparing the high EVOO diet and low EVOO diet within the H2L and L2H diet order. Values are obtained from linear mixed models adjusted for age, sex, and body weight change. To convert mg/dl to millimoles per liter, multiply by 0.0259. Abbreviations: LDL, low-density lipoprotein; HDL, high-density lipoprotein; hs-CRP, high-sensitivity C-reactive protein; TMAO, trimethylamine N-oxide; H2L, high EVOO to low EVOO randomization diet order; L2H: low EVOO to high EVOO randomization diet order.

Figure 1

(a)



(b)

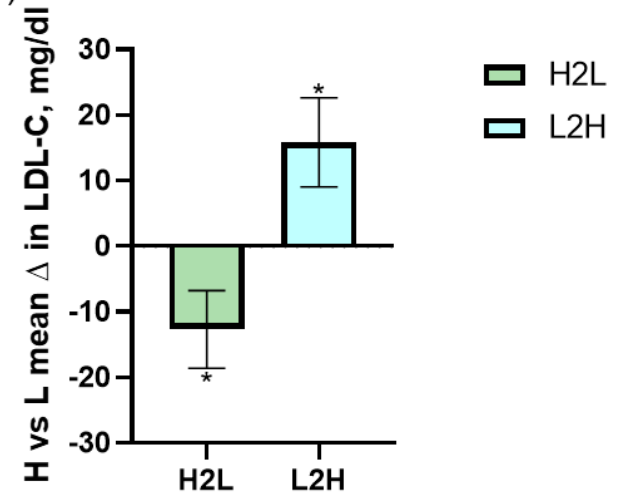


Figure 2

