STATISTICAL ANALYSIS PLAN

Cardiometabolic effect of low-fat and full-fat dairy in healthy and cardio-metabolically at-risk adults

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Table of Contents

1. Administrative information 3
Clinicaltrials.gov Identifier: NCT07605442 (Registered May 18, 2026) 3
Key Personnel 3
2. Introduction 3
Background and Rationale 3
Specific Aims 6
Hypothesis 6
3. Study Methods 6
Trial design 7
Randomization method, allocation concealment, blinding 7
Sample size estimate 7
Hypothesis testing framework 8
Interim analyses 10
Timing of outcome assessment 11
4. Trial Population 11
Recruitment 11
Screening and eligibility criteria 11
Early withdrawal of participants 12
Presentation of baseline characteristics 13
5. Analysis Population 13
6. Hypothesis Testing 13
Primary outcome: 13
Secondary outcomes: 13
7. Statistical Analyses 13
References 14

1. Administrative information

Clinicaltrials.gov Identifier: NCT07605442 (Registered May 18, 2026)

Key Personnel

Principal Investigator: Dr. Kristina Petersen PhD, APD, FAHA is an Associate Professor in the Department of Nutritional Sciences at Penn State University. Dr. Petersen is the Principal Investigator of the clinical trial. Dr. Petersen will be responsible for general study oversight and administration, protocol development and implementation, institutional review board (IRB) submission, data analysis and management, and training study personnel required for protocol execution.

Statistician: Dr. Prabhani Kuruppumullage Don PhD is an Associate Research Professor in the Department of Statistics at Penn State University. Dr. Kuruppumullage Don is a Co-investigator and will oversee all aspects of statistical design and analysis for the study. Specifically, she assisted with the power calculations and the development of the statistical analysis plan. She will also provide consultation on execution of the statistical analysis plan and critically review all of the manuscripts for statistical considerations.

Clinical Research Center: The Clinical Research Center (CRC) at Penn State University is staffed with experienced clinical research employees consisting of physicians, a nurse practitioner, and registered nurses who will work closely with the PI and study personnel throughout the clinical trial to facilitate the research protocol.

Metabolic Kitchen Manager: The Metabolic Kitchen manager will be responsible for overseeing food preparation, procurement, and provision to study participants and will conduct adherence monitoring during the study.

Study Coordinators: Study personnel involving the research laboratory coordinator and research staff will be responsible for recruitment activities, data collection, and study procedures and will facilitate clinical trial operations.

2. Introduction

Background and Rationale

Dietary guidance issued by federal agencies and authoritative organizations for general health and cardiovascular disease (CVD) risk reduction recommends intake of low-fat or fat-free dairy rather than full-fat dairy.[1–4] This recommendation is made because of cardiovascular concerns about the higher saturated fat content of full-fat dairy as well as the higher energy content. However, observational and clinical trial evidence suggest that full-fat dairy intake is not associated with increased risk of CVD[5] or worsening of LDL-cholesterol (LDL-C),[6] a key risk factor for CVD. Although there has been limited empirical investigation of the cardiovascular implications of meeting dairy recommendations with full-fat dairy so concerns about population guidance to consume full-fat dairy instead of low-fat dairy remain. To address this evidence gap and inform population-level guidance for dairy intake, the goal of the proposed

study is to determine if full-fat dairy has equivalent effects on LDL-C and other cardiometabolic disease (CMD) risk factors to low-fat dairy in both healthy adults and those at risk for CMD.

Prospective cohort studies generally show that higher total dairy intake is either not associated with CVD risk or is associated with lower risk of CVD.[7] A recent systematic review and meta-analysis of 27 prospective cohort studies showed that higher total dairy intake was associated with an approximate 4% relative risk reduction for CVD, which equated to a 2% relative risk reduction per 1 daily serving increase in dairy.[5] A systematic review and meta-analysis of 24 prospective cohort studies published a few years earlier showed a similar relative risk reduction estimate, although it was not statistically significant (RR 0.96; 95% CI 0.92, 1.00).[8] In these analyses, dairy intake was estimated by diet assessment methods that are limited by measurement error. Recently, circulating and tissue levels of pentadecanoic acid (15:0) and heptadecanoic acid (17:0) have emerged as objective biomarkers of dairy intake. Notably, in a systematic review and meta-analysis of prospective cohort studies, high vs. low levels of 15:0 (RR 0.88; 95% CI 0.78, 0.99; 17 studies) and 17:0 (RR 0.86; 95% CI 0.79, 0.93; 12 studies) were associated with lower risk of total CVD.[9] Collectively, evidence from prospective cohort studies suggests that total dairy intake (or objective biomarkers of intake) is associated with either neutral or beneficial CVD outcomes.

Epidemiological evidence also suggests that higher vs. lower intake of full-fat dairy is either not associated with CVD risk or associated with lower risk.[7] There has been less investigation of the association between full-fat dairy relative to lower-fat dairy and CVD risk. The 2025 Dietary Guidelines Advisory Committee (DGAC) conducted a systematic review that included 9 prospective cohort studies (15 comparisons) and concluded that substituting higher-fat dairy with lower-fat dairy is not associated with a difference in risk of CVD in adults.[10] However, the evidence was graded as limited because of concerns with consistency, precision, risk of bias, directness, and generalizability. Thus, evidence from prospective cohort studies collectively suggests cardiovascular benefits of dairy intake regardless of the fat content.

Clinical trial evidence suggests that dairy intake, irrespective of the fat content, does not have adverse effects on anthropometric measures, lipids/lipoproteins, or blood pressure.[6] In a systematic review and network meta-analysis including 8 randomized controlled trials (RCT) with a duration ≥ 12 weeks, high intake of full-fat dairy (MD -7.60 mmHg; -17.18, 1.97) and low-fat dairy (MD -5.22 mmHg; 95% CI -10.01, -0.43) improved systolic blood pressure compared to a control/low dairy.[6] In these analyses, there was no effect of high dairy vs. low dairy (or dairy subtypes) on body weight, LDL-C, or triglycerides. Only a few clinical trials have examined the effect of full-fat dairy vs. low-fat/fat-free dairy on CVD risk factors.

Most clinical trials show no difference in blood pressure with full-fat dairy vs. low-fat dairy. [11–13] In a randomized crossover study of young adults who were normotensive (n=43), no difference in blood pressure (primary outcome blood pressure) was observed following intake of 3.5 servings/day of full-fat milk and yogurt, compared to low-fat milk and yogurt, after 8 weeks; however, body weight increased with full-fat vs. low-fat dairy.[11] In a 3-period randomized crossover study of generally healthy adults, a Dietary Approaches to Stop Hypertension (DASH) diet with full-fat dairy and lower in sugars reduced blood pressure compared to the control

Average American diet, and to a similar extent as a DASH diet with low-fat dairy.[14] Collectively, clinical trial evidence suggests full-fat dairy has similar blood pressure effects to low-fat dairy.

Clinical trials have generally shown no difference in the effect of full-fat dairy vs. low-fat/fat-free dairy on lipids/lipoproteins.[12–15] In a 12-week RCT, there was no effect of intake of 80 g/d/2400 kcal of regular fat cheese vs. the same amount of low-fat cheese on LDL-C (primary outcome), total cholesterol, HDL-C, or triglycerides in participants at risk for CVD.[12] Similarly, another 12-week RCT conducted in adults with metabolic syndrome showed no effect of 3.3 servings/day of full-fat milk, yogurt, and cheese compared to the same amount of low-fat dairy or a limited dairy diet ≤3 servings/week on lipids/lipoproteins.[13] Therefore, despite the saturated fat content of full-fat dairy, the effect on lipids/lipoproteins does not appear to differ from low-fat dairy.

The lack of effect of dairy fat on lipids/lipoproteins may be related to the structure of milk fat globules, which include a tri-layered membrane rich in bioactive phospholipids and proteins that enclose the milk fat.[7] In an 8-week RCT, intake of 40 g/day of whipping cream [contains milk fat globule membrane (MFGM)] resulted in lower LDL-C (-12 mg/dL; 95% CI -22, -1.5) than 40 g/day of butter oil in men and women with overweight or obesity.[16] However, this study was not able to identify the mechanism(s) for the attenuated lipid impairment associated with MFGM. In a 4-week randomized controlled trial, intake of 5 g/day of milk polar lipids reduced LDL-C, apoB, and triglycerides compared to the control (0 g/day) in postmenopausal women.[17] Furthermore, it was shown that the milk polar lipids reduced intestinal cholesterol absorption, resulting in a reduction in intestinal-derived chylomicrons and delivery of unabsorbed lipids to the colon. While further research is needed in this area, the available evidence suggests that the milk polar lipid content of full-fat dairy may explain the lack of adverse lipid/lipoprotein effects, supporting the biological plausibility of the non-different findings observed between full-fat and low-fat dairy in RCTs.

RCT evidence suggests that there is no difference in the effect of full-fat dairy vs. low-fat dairy on markers of glycemic control.[12,15,18,19] Schmidt et al. observed no difference in the effect of 3.3 servings/day of full-fat vs. low-fat milk, yogurt, and cheese on glucose tolerance (primary outcome), systemic insulin sensitivity, pancreatic β-cell function, liver fat content, fasting glucose, HbA1c, and HOMA-IR after 12 weeks in adults with metabolic syndrome.[18] Similarly, in a randomized crossover trial (n=13), no difference in 120-minute glucose (following oral glucose tolerance test; primary outcome), glucose tolerance, fasting glucose or insulin, HOMA-IR, or insulin sensitivity was observed with intake of full-fat or non-fat yogurt in adults with prediabetes after 3 weeks.[19] In this study, fructosamine was lower after intake of full-fat yogurt compared to non-fat yogurt. Thus, available RCT evidence suggests that full-fat and low-fat/non-fat dairy have similar effects on glycemic control.

Well-designed, adequately powered studies directly comparing the effect of low-fat and full-fat dairy on risk factors for CVD are needed to inform dietary guidance. The 2025 DGAC concluded that insufficient evidence exists to determine the effect of higher-fat dairy intake, compared to lower-fat versions, on blood lipids and blood pressure in adults and older adults.[20] Only 3 clinical trials[11,12,21] met the inclusion criteria for the DGAC systematic review.[10] Although, as described above, additional clinical trials exist, they were excluded because of small sample

sizes (<30 participants/group). Under the current process for Dietary Guideline development, empirical evidence from adequately powered clinical trials demonstrating that full-fat dairy does not adversely affect cardiometabolic health, particularly lipids/lipoproteins, compared to low-fat dairy, will be needed to prompt any shift in recommendations.

All of the prior trials were designed to test for superiority rather than equivalence, i.e., full-fat dairy has a different effect than low-fat dairy on the outcomes of interest. Under the superiority testing framework, a non-significant difference (failure to reject the null hypothesis) is viewed as inconclusive and not as the same effect. Alternatively, a trial designed using the equivalence testing framework enables direct testing of the hypothesis that full-fat and low-fat dairy have the same effect. Given prior research suggests that full-fat dairy has similar effects on LDL-C and other CMD risk factors to low-fat dairy, directly testing whether these two types of dairy are equivalent would strengthen the evidence base and enable inferences to be drawn about the similarity of low-fat and fat-free dairy.

Dietary guidance is issued at the population level and therefore, evidence generalizability is important. Critically, most of the prior research on the effect of full-fat dairy vs. low-fat dairy was conducted in adults at risk for CVD or CMD, and research in healthy adults is limited. This is relevant because it is well established that excess adiposity impairs LDL-C responsiveness to dietary intervention.[22] Thus, individuals with overweight/obesity may respond differently to full-fat dairy vs. individuals with a healthy body weight. A direct comparison of full-fat vs. lower-fat dairy in both healthy and cardio-metabolically at-risk adults will inform population-level dietary guidance as well as precision public health recommendations.

Specific Aims

The overarching goal is to characterize and compare the effect of full-fat dairy to low-fat dairy on LDL-C and other CMD risk factors in healthy adults and those at risk for CMD to inform population-level dietary guidelines for dairy. This goal will be achieved with the following objectives:

1. Characterize the effect of full-fat dairy compared to low-fat dairy on LDL-C and other CMD risk factors in healthy adults and adults at risk for CMD after 6 weeks.
2. Compare the effect of full-fat dairy to low-fat dairy in healthy adults vs. adults at risk for CMD after 6 weeks.

Hypothesis

This clinical trial will test two hypotheses: 1) intake of 3 cup-equivalents/day of full-fat dairy (milk, yogurt & cheese) has ***equivalent effects*** on LDL-C and other CMD risk factors to 3 cup-equivalents/day of low-fat dairy (milk, yogurt & cheese) in adults with normal BMI and optimal/near optimal LDL-C, and in adults with overweight/obesity and elevated LDL-C after 6 weeks; 2) the effect of full-fat dairy on LDL-C and other CMD risk factors, compared to low-fat dairy, is ***equivalent*** in adults with normal BMI and optimal/near optimal LDL-C compared to adults with overweight/obesity and elevated LDL-C after 6 weeks. This trial is expected to demonstrate equivalent effects of full-fat and low-fat dairy on LDL-C and other CMD risk factors in both healthy and at-risk adults.

3. Study Methods

Trial design

This is a randomized cross-over clinical trial with 2 parallel arms (see Figure 1). Arm 1 will include adults with normal BMI (18.5-24.9 kg/m^2) and optimal/near optimal LDL-C (≤ 129 mg/dL) and Arm 2 will include adults with overweight/obesity (25-39.9 kg/m^2) and elevated LDL-C (130-190 mg/dL). Within each arm, participants will undergo two 6-week dietary conditions in random sequence order, separated by a 4-week washout: 1) 3 cup-equivalents/day of full-fat dairy; 2) 3 cup-equivalents/day of low-fat dairy.

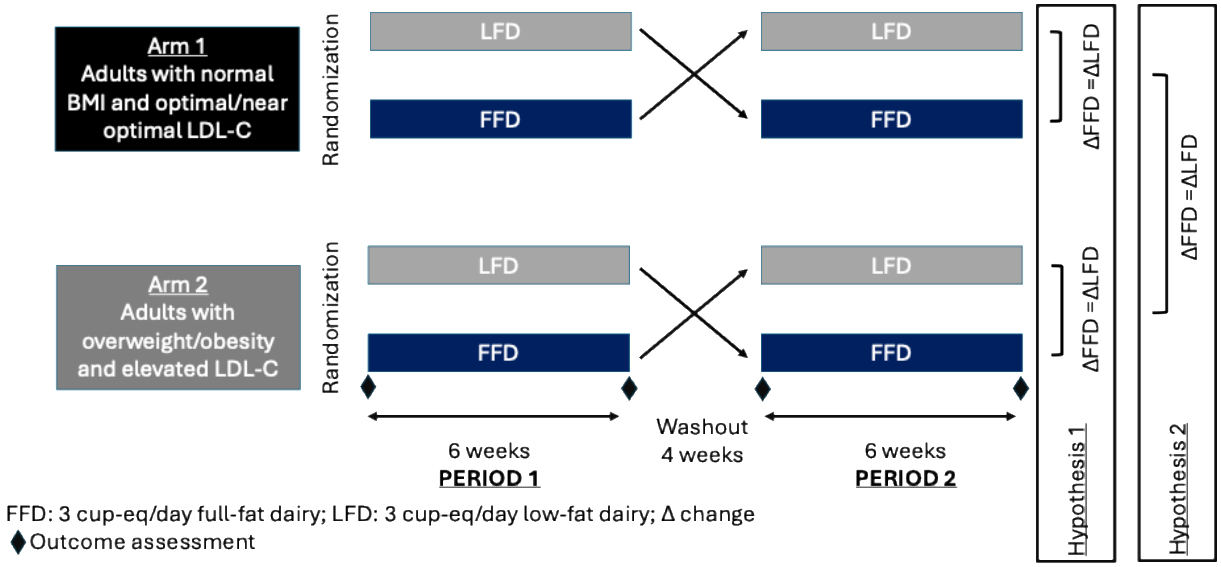


Figure 1: Study Design

Randomization method, allocation concealment, blinding

Condition sequence will be randomized at the individual level. The randomization schedule will be stratified. Strata 1 will be for participants with a normal BMI (18.5-24.9 kg/m^2) and optimal/near optimal LDL-C (≤ 129 mg/dL). Strata 2 will be for participants with overweight/obesity (25-39.9 kg/m^2) and elevated LDL-C (130-190 mg/dL). Block sizes of 2, 4 and 8 will be used. For each stratum, the randomization sequence will be uniform and balanced with regard to first-order carryover effects (i.e., AB and BA). Randomization sequences will be computer-generated by a person who is not involved in recruitment or data collection and will be uploaded into REDCap. REDCap will be used to ensure allocation concealment. Prior to baseline testing for period 1, the metabolic kitchen manager will use the randomization module in REDCap to reveal the participant's randomization. The person generating the randomization sequence and the metabolic kitchen manager will be the only study team members with user access to randomization in REDCap. The nurses drawing blood and laboratory staff conducting the assays will be blinded to the randomization schedule. The randomization code will be unsealed only once all participants have completed the study and the dataset has been reviewed and cleaned. Participants will be told which type of dairy they are consuming and will not be blinded.

Sample size estimate

For the first hypothesis, 75 subjects per arm are needed to test within-arm equivalence in the 6-week change in LDL-C (primary outcome), assuming a between-condition mean difference of

0 mg/dL (SD 14.6 based on a prior study from our lab looking at a single food intervention in a similar population[23]), an equivalency margin of ±5 mg/dL, 80% power, and $p<0.05$. An equivalency margin of 5 mg/dL is used because a change in LDL-C of <5 mg/dL is not associated with a clinically meaningful change in ASCVD risk.[24] Furthermore, prior studies comparing full-fat dairy to low-fat dairy show effect estimates of -0.8 to 3.1 mg/dL.[12–15] 75 subjects per arm will also provide adequate power to test the second hypothesis; the same equivalency margin will be used for the second hypothesis. Overall, 150 subjects are needed; 173 subjects will be recruited to account for an ~15% attrition rate.

Hypothesis testing framework

Primary Endpoint (6-week change from baseline):

- **LDL-C:** LDL-C will be assessed on two consecutive days at the beginning (baseline) and the end of each period (endpoint) using the Modified Sampson–NIH Equation, which has a higher degree of accuracy than other LDL-C estimation equations and direct LDL-C.[25] The change in LDL-C will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). For hypothesis 1, the equivalency hypothesis will be tested using paired samples t-tests with two one-sided tests (TOST) for equivalence with bounds set at -5, 5 mg/dL. Equivalence will be established if the 90% confidence interval for the between-condition mean difference is entirely within the predefined equivalence bounds (i.e., -5 to 5). For hypothesis 2, between-arm (Arm 1 vs. Arm 2) equivalency will be tested using independent samples t-tests with TOST equivalence analysis. The same equivalency margin will be used as for hypothesis 1.

Secondary Endpoints (6-week change from baseline)

The statistical procedures outline for the primary outcome will be used for all of the secondary outcomes. For each secondary outcome the equivalence bounds are as follows:

- **ApoB:** ApoB will be assessed on one day at the beginning and the end of each period. The change will be calculated as the endpoint value minus the baseline value. The equivalence bounds will be set at -4, 4 mg/dL. The minimally important difference for apoB is 4 mg/dL.[26]
- **Triglycerides:** Triglycerides will be assessed on two consecutive days at the beginning and the end of each period. The change in triglycerides will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). The equivalence bounds will be set at -8.9, 8.9. The minimally important difference for triglycerides is 8.9 mg/dL.[26]
- **Total cholesterol:** Total cholesterol will be assessed on two consecutive days at the beginning and the end of each period. The change in total cholesterol will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). The equivalence bounds will be set at -10, 10 mg/dL. This is expected to represent an approximate 5% difference.
- **HDL-cholesterol:** HDL-cholesterol will be assessed on two consecutive days at the beginning and the end of each period. The change in HDL-cholesterol will be calculated

as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). The equivalence bounds will be set at -4.6, 4.6 mg/dL. A 4.6 mg/dL increase in HDL-cholesterol is associated with a 10% reduction in risk for coronary heart disease.[27]

- **Non-HDL cholesterol:** non-HDL-cholesterol will be assessed on two consecutive days at the beginning and the end of each period by subtracting HDL-cholesterol from total cholesterol. The change in non-HDL cholesterol will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). The equivalence bounds will be set at -4, 4. The minimally important difference for non-HDL-cholesterol is 4 mg/dL.[26]
- **Fasting plasma glucose**: Glucose will be assessed on two consecutive days at the beginning and the end of each period. The change in glucose will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). The equivalence bounds will be set at -9, 9 mg/dL. The minimally important difference for fasting glucose is 9 mg/dL.[26]
- **Fasting insulin**: Insulin will be assessed on two consecutive days at the beginning and the end of each period. The change in insulin will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). The equivalence bounds will be set at -0.83, 0.83 µIU/mL. The minimally important difference for fasting insulin is 0.83 µIU/mL.[26]
- **Fructosamine:** Fructosamine will be assessed on one day at the beginning and the end of each period. The change will be calculated as the endpoint value minus the baseline value. The equivalence bounds will be set at -5, 5 µmol/L. A minimally important difference has not been defined for fructosamine. This margin has been selected based on a prior study from our lab, which did not show an effect of the intervention on fasting glucose or fructosamine.[28]
- **Homeostatic model of insulin resistance (HOMA-IR):** Fasting glucose and insulin will be assessed on two consecutive days at the beginning and the end of each period. The mean of the two measurements taken at each timepoint will be used to calculate HOMA-IR. HOMA-IR will be calculated as (insulin µIU/mL × glucose mg/dL) / 405. The change in HOMA-IR will be calculated as the endpoint value minus the baseline value. The equivalence bounds will be set at -0.5, 0.5. Limited data exist to define the minimally important difference; this margin is estimated to reflect a 25% change. Baseline HOMA-IR is expected to be ~2.0.
- **C-reactive protein (CRP):** high sensitivity CRP will be assessed on two consecutive days at the beginning and the end of each period. The change in CRP will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). Values above 10 mg/L will be removed from analyses because this reflects acute inflammation. The equivalence bounds will be set at -0.5, 0.5 mg/L. The minimally important difference for CRP is 0.5 mg/L.[26]

- **Body weight:** Weight will be assessed on two consecutive days at the beginning and the end of each period. The change in weight will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values). The equivalence bounds will be set at -0.5, 0.5 kg. The minimally important difference for weight is 0.5 kg, which represents weight maintenance.[26]
- **Central blood pressure:** Systolic and diastolic blood pressure will be measured at the beginning and the end of each period. The change in systolic and diastolic will be calculated as the mean endpoint value (i.e., mean of the last two measurements of three taken or if brachial systolic blood pressure is inconsistent (i.e., >10 mm Hg difference), a fourth measurement will be taken, and the 2 closest measurements from measurements 2-4 will be used for analysis) minus the mean baseline value. The equivalence margins will be set at the same bounds are for peripheral blood pressure because evidence suggests similar risk prediction for central and peripheral blood pressure.[29]
- **Peripheral blood pressure:** Systolic and diastolic blood pressure will be measured at the beginning and the end of each period. The change in systolic and diastolic will be calculated as the mean endpoint value (i.e., mean of the last two measurements of three taken or if brachial systolic blood pressure is inconsistent (i.e., >10 mm Hg difference), a fourth measurement will be taken, and the 2 closest measurements from measurements 2-4 will be used for analysis) minus the mean baseline value. For systolic blood pressure the equivalence bounds will be set at -4, 4 mmHg. This is based on the FDA requirement for approval of new antihypertensive medications.[30] For diastolic blood pressure, the equivalence bounds will be set at -2, 2 mmHg based on the minimally important difference of 2 mmHg. [26]
- **Pulse wave velocity (PWV):** carotid-femoral PWV will be measured at the beginning and the end of each period. The change will be calculated as the mean endpoint value (i.e., mean of three readings within 0.5 m/s) minus the mean baseline value. The equivalence bounds will be set at -0.7, 0.7 m/s. A 0.7 m/s higher PWV is associated with a 10% increase in cardiovascular events.[31]
- **Healthy Eating Index-2020:** The Healthy Eating Index-2020 (HEI-2020)[32] will be calculated from 24-hour recalls conducted at the beginning and the end of each period. Prior to the beginning of each period and during the last week of each period, participants will be asked to complete two 24-hour recalls (total four per period). At each timepoint, the HEI-2020 will be calculated by averaging the results of the two recalls. The change in the HEI-2020 will be calculated as the mean endpoint value minus the mean baseline value. The equivalence bounds will be set at -2.2, 2.2 points. A clinically significant difference in the Healthy Eating Index is 5-6 points based on an effect size of 0.5.[33] Therefore a small effect size (0.2) corresponds to a 2.2 point difference.

Interim analyses

No interim analyses will be performed.

Timing of outcome assessment

4. Trial Population

Recruitment

Participants will be recruited from University Park and State College, PA and surrounding areas using public advertisements and recruitment flyers posted on campus and in the local community (State College/University Park area).

Screening and eligibility criteria

Individuals responding to advertising will be emailed information about the study and complete a pre-screening survey via REDCap. Potentially eligible individuals will be telephone screened. Based on the answers to the questions, participants will be deemed eligible or ineligible by the staff member assessing eligibility in consultation with the PI. Eligible individuals will be scheduled for a clinic screening visit. At the clinic screening appointment, anthropometrics and blood pressure will be measured. Fasting blood samples will be assessed for a complete blood count, comprehensive metabolic panel, iron, phosphate, lipid panel, and uric acid. If a potential participant takes thyroid medicine, they must provide a current (within 6 months) lab test. If they do not have one, thyroid-stimulating hormone will be measured. Inclusion/exclusion criteria will be assessed.

At the screening visit, participants must meet all the following inclusion criteria and none of the exclusion criteria to participate in this study.

Inclusion criteria

- Age 25-70 years
- Arm 1 Healthy Adults: BMI 18.5-24.9 kg/m^2 and LDL-C ≤ 129 mg/dL assessed by the Modified Sampson–NIH Equation
- Arm 2 Adults at risk for cardiometabolic diseases: BMI 25-39.9 kg/m^2 and elevated LDL-C 130-190 mg/dL assessed by the Modified Sampson–NIH Equation
- Currently consume milk, yogurt, and/or cheese daily
- Willing to eat 3 servings of dairy (milk, yogurt, cheese) for two 6-week periods during the study

Exclusion criteria

- Hemoglobin <13.2 g/dL for men or < 11.7 g/dL for women at screening
- Fasting triglycerides >350 mg/dL at screening
- ≥10% change in body weight within the 6 months prior to enrollment
- Blood pressure >140/90 mmHg at screening
- Fasting glucose ≥126 mg/dL
- Type 1 or type 2 diabetes
- Takes any (prescription or over-the-counter) anti-hypertensive, lipid-lowering, glucose-lowering or anti-inflammatory drugs, or drugs that alter body weight
- Intake of supplements that affect the outcomes of interest (i.e., lipids, blood pressure, glucose, body weight, inflammation and microbiome) and are unwilling to cease during the study period

- Unwilling to refrain from starting to take any supplements, vitamins, nutritional products, or health foods that are not prescribed by a doctor for the duration of the study
- History of liver, kidney, or autoimmune disease
- Prior cardiovascular event (e.g., stroke, heart attack)
- Current pregnancy or intention of pregnancy within the next 12 months
- Lactation within the prior 6 months
- Dairy allergy/intolerance/sensitivity/dislike
- Antibiotic use within the prior four weeks
- Oral steroid use within the prior four weeks
- Use of tobacco or nicotine-containing products within the past 6 months
- History of cancer at any site within the past 10 years (eligible if ≥10 years without recurrence) or non-melanoma skin cancer within the past 5 years (eligible if ≥5 years without recurrence)
- Participation in another clinical trial within 60 days of baseline
- Currently following a restricted or weight-loss diet
- Prior bariatric surgery
- Intake of >14 alcoholic drinks/week and/or not willing to avoid alcohol consumption for 48 hours prior to test visits
- Does not speak and/or understand English
- Unwilling to refrain from donating blood and/or plasma during the study
- Weight <110 lb
- Unwilling/unable to eat 3 servings of dairy/day during the study
- Lactose intolerance or sensitivity
- Unwilling to contact study staff before enrolling in other health-related research and avoid participating in any research that may interfere with this study
- For individuals taking thyroid medication: abnormal thyroid stimulated hormone (TSH) concentration, or change in dose of thyroid medication within the last 6 months
- Principal Investigator discretion related to the potential participant's ability to adhere to the study requirements, including being able to come to attend visits

Early withdrawal of participants

Participants will be withdrawn from the study for the following reasons:

- Risks to the other participants/research team members, disruptive behavior during the study visit, or food pick-ups
- Diagnosis of a disease listed as an exclusion criterion or a serious medical condition requiring active intervention (assessed by review of medical history form or participant report)
- Prescription of anti-hypertensive, lipid-lowering, or glucose-lowering drugs or drugs that affect body weight (assessed by review of medical history form or participant report)
- Prescription of steroids, antibiotics or anti-inflammatory drugs for longer than 1 week
- Pregnancy

Presentation of baseline characteristics
Baseline demographic and clinical characteristics will be reported for the total analysis population and by arm and randomization according to CONSORT guidelines.

5. Analysis Population

Analyses will be conducted consistent with intent-to-treat principles. All available data from randomly assigned participants will be included in data analyses. A secondary per-protocol analysis will be conducted that will only include participants with a high level of adherence defined as consuming ≥ 90% of the provided dairy (≥2.7 cup-equivalents/day) on ≥90% of the study days (≥ 38 days) based on self-reported intake from the daily monitoring survey. This will be treated as a sensitivity analysis to determine the robustness of the findings.

6. Hypothesis Testing

Primary outcome:
The primary outcome is 6-week change in LDL-C. LDL-C will be assessed on two consecutive days at the beginning and the end of each period using the Modified Sampson–NIH Equation.[25] The change in LDL-C will be calculated as the mean of the endpoint measures (i.e., mean of day 1 and day 2 values) minus the mean of the baseline measures (i.e., mean of day 1 and day 2 values).

Secondary outcomes:
The secondary outcome variables will be 6-week change (end of period minus period baseline) in:

- ApoB
- Triglycerides
- Total cholesterol
- HDL-cholesterol
- Non-HDL-cholesterol
- Fasting glucose
- Fasting insulin
- Fructosamine
- Insulin resistance assessed by the homeostatic model of insulin resistance (HOMA-IR)
- C-reactive protein (CRP)
- Body weight
- Central and peripheral blood pressure
- Carotid femoral pulse wave velocity (PWV)
- Diet quality assessed by the Healthy Eating Index-2020

7. Statistical Analyses

The primary analysis will include all data collected from randomly assigned participants consistent with intent-to-treat principles. A secondary per-protocol analysis will be conducted that will only include participants with a high level of adherence defined as consuming ≥ 90% of the provided dairy (≥2.7 cup-equivalents/day) on ≥90% of the study days (≥ 38 days). This will be treated as a sensitivity analysis to determine the robustness of the findings. Assumptions for

all statistical tests will be checked and confirmed prior to conducting analyses for hypothesis testing. The change from baseline will be calculated by subtracting the endpoint value from the baseline value for each condition. To test hypothesis 1, within-arm (Arm 1 and Arm 2) equivalency for all outcomes will be tested using paired samples t-tests with two one-sided tests (TOST) for equivalence. Equivalence will be established if the 90% confidence interval for the mean difference falls entirely within the predefined equivalence bounds. Equivalency margins for all outcomes are defined based on clinical importance. For hypothesis 2, between-arm (Arm 1 vs. Arm 2) equivalency will be tested using independent samples t-tests with TOST equivalence analysis. The same equivalency margins will be used for hypotheses 1 and 2. Statistical significance $p<0.05$.

References

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