PROTOCOL: Project
Project Title: "A short term evaluation of a structured weight loss plan in overweight and obese adults"
Version 1 Date: March 18, 2016

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Purpose

The present study is designed to examine the effect of changes in body weight and related parameters associated with a commercially-available, low-carbohydrate diet plan. A parameter proposed to be studied here is to measure the impact of fructose restriction and weight loss on serum uric acid concentrations and arterial stiffness.

Background

It is well know that consumption of a calorically-restricted diet helps people lose weight. A calorie-restricted diet with low carbohydrate lowers the serum uric acid concentration which is a factor associated with obesity and also represents a particular metabolic risk in women compared to men. The present weight loss study will determine whether women benefit more than men to short-term weight loss occurring during restriction of simple sugars.

Recent epidemiologic data in humans have identified hyperuricemia as a risk factor for cardiovascular (CV)-related disease in women, raising the risk of coronary heart disease by approximately 70% compared to its effect in men (1). Two studies focusing on sex-differences have demonstrated that the relationship between uric acid and CV stiffness observed in the general population is largely driven by the effect in women. These relationships are shown in **figure 1**, where quartiles of serum uric acid (x-axis), are presented for men and women and reveal that the level of arterial stiffness increases with increasing uric acid concentrations only in women (**fig. 1A and 1B**), as does the prevalence of diastolic dysfunction (a composite measure of cardiac stiffness) shown in **figure 1D**.

Negative metabolic effects of dietary fructose - The PI of the present project was an organizer of an NIH consensus conference on the potential negative metabolic effects of dietary fructose (2). The consensus panel called for research to test hypotheses related to whether a reduced-sugar intervention that causes weight loss would improve disease biomarkers for CV risk. With respect to known fructose effects on the metabolic syndrome, supplementation of dietary fructose increases body weight, plasma triacylglycerols (TG), lowers HDLc (3), and raises ambulatory blood pressure (4) and the latter effect can be blocked by the xanthine oxidase inhibitor, allopurinol (5). In rodents, fructose feeding to induce the metabolic syndrome is mediated partly by hyperuricemia but doses needed for this effect can be high, due to rats having the enzyme uricase, which hydrolyzes uric acid. By contrast, humans do not

produce this enzyme and graded doses of fructose in the diet incrementally increase uric acid concentrations (3). Stanhope et al found a linear relationship between fructose supplementation (from 0%, to 10%, 17.5%, and 25% of energy) and increasing serum uric acid, which was higher by 11% at the top dose (*P*<0.0001 for effect of dose). In line with the expert consensus, the design of human studies should allow for better translation of the large amount of preclinical (rodent) data on the detrimental CV effects of fructose feeding in rodents and do so, in humans, by practical dietary means. Lastly, the consensus highlighted one scientific area in critical need of further research – investigation of the effects of dietary sugar intake in at-risk populations, i.e., those already consuming diets high in added sugars.

Figure 1. Relationships between serum uric acid and CV stiffness between the sexes

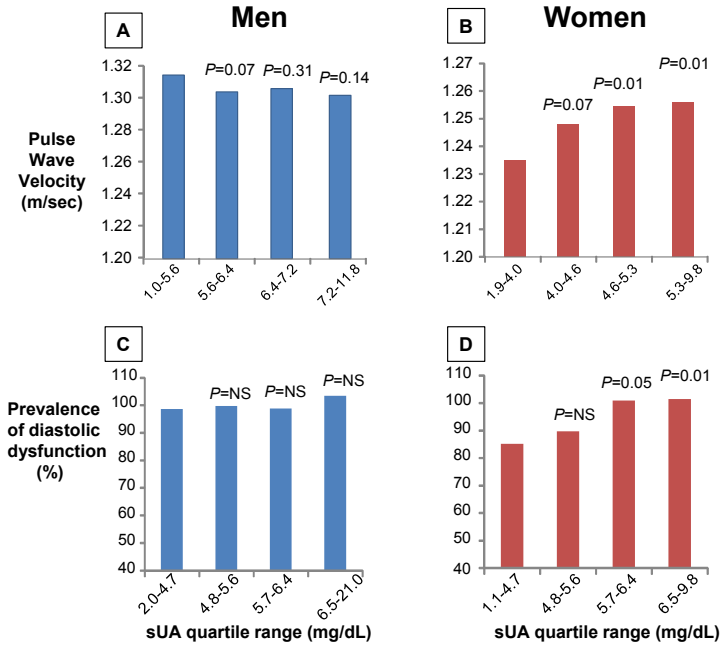


Fig. 1A and 1B modified from Fang et al. The comparison of brachial-ankle pulse wave velocity level among subjects with different quartiles of serum uric acid (sUA) concentrations by gender, based on ANCOVA (adj for age, obesity, SBP, fasting glucose, hypercholesterolemia, hypertrigly-ceridemia, low HDL-C, smoking habit, alcohol consumption and vigorous exercise). *P*-values compare the pulse wave velocity values at quartiles 2, 3 and 4 with quartile 1.

Fig. 1C and 1D modified from Nogi et al. The association between serum UA concentration quartiles and diastolic dysfunction by logistic regression (adj for age, Log(eGFR), SBP, pulse rate, diuretics, both loop or thiazide). *P*-values compare the prevalence of diastolic dysfunction at quartiles 2, 3 and 4 with quartile 1.

As described Stanhope (3), an increased intake of added sugars in the diet is associated with dyslipidemia, CVD, and metabolic syndrome (6, 7) and, as shown in **table 1**, data from the NHANES suggest that the higher the intake of added sugars, the greater the CVD risk (8). The average dietary amount of added sugars in U.S. adults 20–60 years is 13–14% of daily calories and, importantly, is significantly associated with an 18% increase in CVD mortality risk. Posthoc analysis of data from the PI's most recent weight-loss study in subjects with metabolic syndrome agree with this analysis.

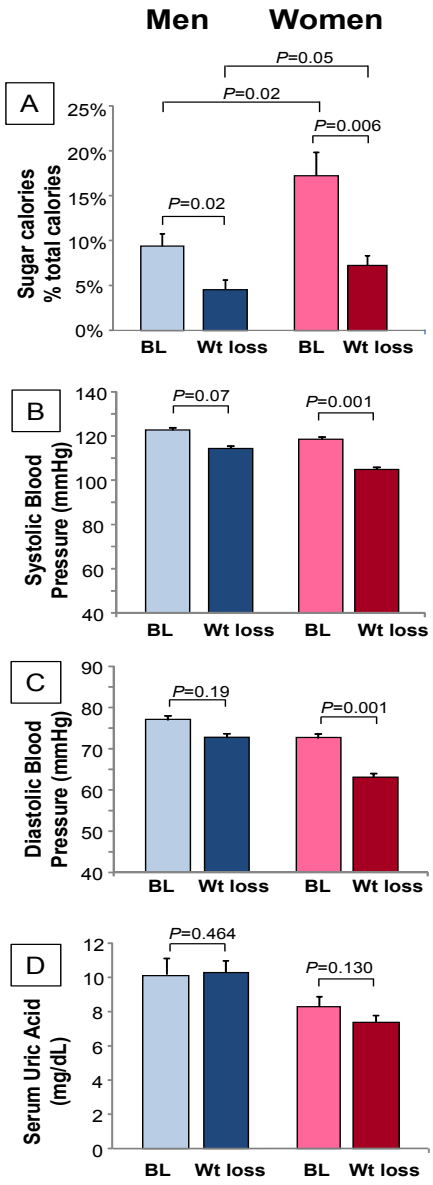
Table 1. NHANES data on intake of added sugars

Quintile of added sugars in the diet		Adjusted hazard ratio of CVD mortality
Q1	0. 0 - 9.6%	1.00 referent
Q2	9.6 - 13.1%	1.07 (1.02-1.12)
Q3	13.1 - 16.7%	1.18 (1.06-1.31)
Q4	16.7 - 21.3%	1.38 (1.11-1.70
Q5	≥ 21.3%	2.03 (1.26-3.27)

n = 11,733. Added sugars represented as the energy from sugars relative to total dietary energy (kcal/kcal). HR (95% confident interval), *P* = 0.004 for dose.

As shown in **figure 2**, ad libitum diets in men contained 9.3 ± 3.6% of energy as added sugars, which was significantly lower than the baseline intake in women (16.6 ± 8.1%, *P*<0.02).

Figure 2. Sex specific changes following fructose-restriction



Data are mean ± SE. Men (n=7) and women (n=9) participated in a program of energy restriction targeted at reducing consumption of added sugars.

These subjects participated in a program designed to reduce dietary sugar intake and body weight. Weight loss was similar in men (-10 ± 3%) and women (-10 ± 2%, *P*=0.99, data not shown) but women reduced their sugar intake more than men (**fig. 2A**). Further, in women, limiting dietary sugars and body weight loss significantly reduced both systolic (**fig. 2B**) and diastolic blood pressure (**fig. 2C**). Even in this relatively small sample size, serum uric acid in women tended to be reduced with weight loss while it was unchanged in men (**fig. 2D**) exhibiting a significant time by sex interaction (*P*=0.006 by ANOVA). Overall, these data demonstrate that the higher added sugar intake of women is a modifiable characteristic that can improve blood pressure and serum uric acid. Our proposed study expands the use of these established procedures by adding a state-of-the-art measure of arterial stiffness - pulse wave velocity (PWV).

The present study is designed to cause weight loss acutely through consumption of a diet that is restricted in calorie and simple sugars. Both men and women will be studied and their results compared to determine whether women benefit more with respect to arterial stiffness. Upon finishing the four-week diet intervention, body weight and blood lipids will be assessed to compare the longer term effect on blood pressure.

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Summary of Study Objectives

We will accomplish the following specific aims:

Aim 1 (weight loss): To induce weight loss, overweight/obese subjects will be fed with a 2-week calorie-restricted diet. This specially formulated diet (see Appendix) will be low in carbohydrates. After this two weeks, the subjects will prepare their own food based on the Atkins online diet program for subsequent 2 weeks. **Hypothesis 1***: After four weeks of a calorie-restricted diet feeding, the subjects will lose 3-6% of their body weight.*

Aim 2 (arterial stiffness improvement): To quantify arterial stiffness improvement by measuring aortic pulse-wave velocity (PWV). **Hypothesis 2:** *After four weeks of feeding, the subjects will have lower serum uric acid level and lower aortic PWV. Further, the female participants will have even more improvement of PWV compared to men.*

AIM 3 (lifestyle impact on body weight): To assess the impact of individual lifestyle factor on the change of body weight, blood pressure and heart rate. **Hypothesis 2:** *During the four weeks of follow-up, the changes of subjects' body weight, blood pressure and heart rate will reflect their lifestyle.*

Impact: The present study to evaluate the effect of a calorically-restricted diet on body weight and arterial stiffness in overweight and obese adults. The data generated by this study will determine whether arterial stiffness can be improved by short-term low carbohydrate diet, and the potential mechanism by which uric acid level impacts CV risk in overweight and obese adults, especially in women. This study will also provide the important preliminary data for an NIH R01 grant submission.

Study Design

The sources of research material will be subject and family medical history, physical exam, screening laboratory tests, Dual-energy X-ray absorptiometry (DEXA, total and regional fat mass and fat-free mass), blood tests, aortic PWV, indirect calorimetry data and behavior questionnaires. Demographic data (plus other related data: emergency contact person, pregnancy status), blood pressure and morphometrics will be measured (height, weight). Lifestyle information including food intake and physical activity will be obtained. Each subject will donate 280 ml of whole blood for all combined blood draws performed for the screening and baseline test and the rest four interim and follow-up tests. **Table 2** shows how the 280 ml of blood given by the subject is allotted over the course of the study. The participant will consume two weeks of either a "Jump Start" or a "Grab-and-go" diet after the baseline visit 1 (please refer to the Appendix for meal composition and calorie density).

Table 2. Accounting of Blood Usage

Procedure	Volume of whole blood (mls)
Screening blood chemistries	40
Baseline visit 1	60
Interim visit 2	30
Follow-up visit 3	60
Follow-up visit 4	30
Final visit 5	60
Total for each subject (mls)	280

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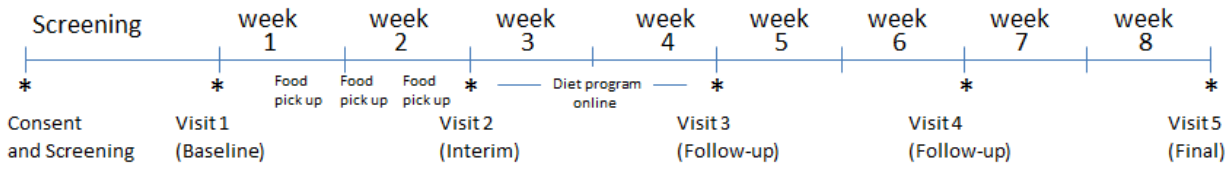
Overall activities

The study tests will be completed through a total of 6 visits (and 3 food pick ups). Please see the study timeline below. First, screening laboratory tests are used to assess general medical status. Second, during the baseline visit 1, the subject's aortic PWV is measured to assess the arterial stiffness. Fasting blood is collected to measure the baseline serum uric acid, creatinine, insulin, glucose and lipid level. A fasting urine sample is collected to measure urinary ketones. Third, the subject's body weight, waist, blood pressure, heart rate and fasting urinary ketone will be determined during the interim visit 2. Fourth, the tests performed at the baseline visit will be repeated during the follow-up visit 3. Blood collected during the baseline and final visits is processed to obtain plasma or serum, and the entire sample is used in the isolation and measurement of blood metabolites and hormones including glucose, TG, FFA, liver function tests, insulin, and plasma hormones. Physical activity before participation in the study and during the 4-week of diet consumption will be monitored by wearing an accelerometer device.

Study Timeline: times of blood draws denoted with an *****



Upon finishing the dietary intervention, the subjects come back for follow-up visit 4 at week 6 and final visit 5 at the end of week 8 to measure blood pressure, heart rate and body weight. The subjects will be asked to fill out questionnaires to assess the behavior and lifestyle change. Once the data acquired, the processed samples will be kept until at least 10 years after publication. The sample tubes are labeled with the subject's study number. No identifying information is present on the tubes. No tools (e.g., cell lines, probes, etc) will be generated from any of this material. All samples are coded to conceal the subjects' identities.

Subject Compensation

For screening visit, subjects receive no compensation, $25 for completing the baseline test, $75 for consuming a 2-week specially formulated diet, and $100 for finishing the follow-up test 3 and $25 each for returning for visits 4 and 5 ($250 total).

Study design and data analysis

Our statistical analysis plan, designed in consult with Greg Petroski, is based both on our preliminary data (**fig. 2**) and data from the increased body of literature that has been generated over the past few years (9-12). The present study is designed to generate data from a future grant and thus, we will use the data generated here for future sample size estimation. For analysis, study group characteristics will be summarized with descriptive statistics and the data will be analyzed as % and absolute change. Paired t-tests will be used to test if the within-group changes are significantly different from zero, and a one-way analysis of variance applied to the "change scores" will be used to test for group (sex)

differences with respect to change. If the overall ANOVA test is significant, pairwise comparisons will be carried out to determine which groups differ. A technique such as the Bonferroni-Holms procedure will be used to correct for multiple testing. Standard ANOVA methods rest on the twin assumptions of normally distributed errors and equal variances across groups. Depending on the distribution of the individual variables, Pearson or Spearman correlation coefficients will be used to examine pairwise associations between continuous variables. Results will reported as sex group means or medians, as are most appropriate for the data, along with 95% confidence intervals for the summary statistics. Statistical analysis will be performed with SAS software (SAS Institute, Cary, NC).

Table 3 below shows the major outcomes measured in this study and the anticipated results at the end of 4-week dietary intervention.

Table 3. Study outcomes and anticipated results at the end of 4-week dietary intervention

Primary outcome variable	Method of detection	Baseline to Visit 3 change
Body weight	Weight scale	↓
Waist circumference	Tape measure	↓
Body fat percentage	DEXA	↓
Blood pressure	Blood pressure cuff	↓
Energy expenditure	Indirect calorimetry	↓
Lipid level	Enzymatic assay	↓
Glucose and insulin level	Enzymatic assay and ELISA	↓
Fructose level	Enzymatic assay	↓
Uric acid level	ELISA	↓
Creatinine level	ELISA	↓
Urinary ketone level	Enzymatic assay	↓
Carotid-femoral PWV	SphygmoCor XCEL device	??

Abbreviations: DEXA, dual-energy X-ray absorptiometry; Indir. cal., indirect calorimetry; ELISA, enzyme-linked immunosorbent assay; PWV, pulse wave velocity

Schedule of Events

Screening

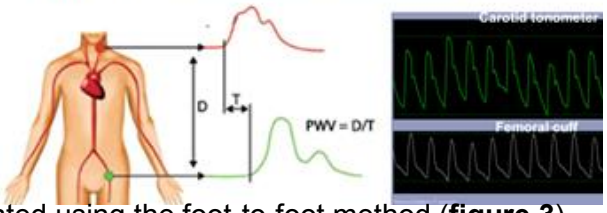
Twenty overweight/obese subjects (10 men and 10 women, 30-55 yrs old) will each participate in the 4-week dietary intervention and 4-week follow-up phase. Eligibility is determined through interactions between study staff and the subjects (initial phone conversation and a screening visit). During the screening visit at the clinical research center (CRC), after signing the consent form, the subject undergoes a fasting blood draw (40 ml) to rule out the presence of diabetes and blood is drawn for serum HbA1c, complete blood count, thyroid, liver (liver enzymes and hepatitis), and kidney function, and a lipid panel are measured. Medical information is obtained including DOB, gender, ethnic/racial category, height, body weight (history of body weight) and medical history (medication use, smoking history). The subject is fed breakfast after blood draw and fills out surveys to assess physical activity, typical food intake, food cravings and sleep quality.

Baseline visit 1

When results from the screening visit are available, eligible/interested subjects are scheduled for their baseline visit 1 after which, they would start the 2-week feeding regimen. The subject is instructed to

wear a study-provided Fitbit Flex® device (Fitbit, San Francisco, CA) four days before the baseline test and during the 4-week dietary intervention to assess the physical activity level. The baseline visit will take place in the CRC. Before the participant comes to the CRC on the study day, he/she is instructed to collect a fasting urine sample. Upon arrival, the participant first voids and then the study staff take the vital signs and measure the height and weight. A catheter is inserted into the vein of one arm to collect 60 ml whole blood. Next, DEXA scan is performed to assess body composition. The resting energy expenditure is subsequently determined using an indirect calorimetry (metabolic cart). The subject lies comfortably in a bed on his/her back and the metabolic cart collects and analyzes the breath to calculate the total energy expenditure. This procedure will take about 30 minutes before continuing for the aortic PWV testing. The subject lies supine again for the aortic PWV testing (**figure 3**).

Figure 3. Aortic Pulse Wave Velocity (PWV)



Carotid-femoral PWV measurement is considered the gold standard, non-invasive index of arterial stiffness. The SphygmoCor XCEL device is used for this assessment, which allows for the simultaneous acquisition of carotid (via tonometer) and femoral (via cuff) pulse waves following the established guidelines (13, 14). Transit time between carotid and femoral pressure waves is calculated using the foot-to-foot method (**figure 3**). Wave 'foots' are identified using intersecting tangent algorithms. PWV is calculated as distance traveled by the pulse wave (i.e., femoral location-sternal notch minus sternal notch-carotid location) divided by pulse transit time. The intra-subject reproducibility for the measurement (2 wks apart, n=15), is as follows: measurement #1, 6.13 m/s; measurement #2, 6.19 m/s, resulting in an average % difference of the two measurements of 0.98% with a SD of that difference among the subjects of 8%. All measurements of PWV are performed by the same investigator.

Consumption of two-week specially formulated diet

The study is designed to first measure how well the commercial meals of low carbohydrate and reduced calorie result in the weight loss over two weeks and then assess the effect of subsequent consumption of a self-prepared, energy-restricted, low-carbohydrate meals on body weight and CV function. All foods given to the subject are commercially available and sold in supermarkets. The subject is assigned randomly to one of two dietary patterns in which all the food is provided for two weeks. Diet 1: The "Grab-and-Go" plan provides Atkins shakes and bars for breakfast, lunch, and snacks. For dinner the subject is given a freshly-prepared meal. Diet 2: The "Jump-Start" plan provides Atkins frozen meals at breakfast, lunch and dinner and these meals are supplemented with fresh salads and vegetables. The reason for the two diets is to understand how subjects like the diets. The diets have the same calories, are otherwise similar in content, and not expected to differ in their ability to cause weight loss.

Beverages: The subject will be asked to consume 8, 8-oz glasses of liquids per day. The subject may also consume coffee or tea without nondairy creamer added, but non-caloric sweeteners, and/or 2 oz of cream per day to the coffee or tea are allowed. Diet soda or other non-caloric beverages are allowed.

We anticipate no medical risks from consuming these diets for 2 weeks. The subjects will be asked to consume only the prescribed foods given to them and no other foods. This may get boring or the subject may experience frustration due to limited food and beverage choices.

Interim visit 2
Before the subject comes for this visit, he/she will be instructed to collect a urine sample at home as he/she did on the Baseline visit 1 and bring the urine sample to the CRC. The study staff will assess the body weight, waist circumference, heart rate and blood pressure. After these measurements, the subject fills out a questionnaire to assess the food craving, appetite and satisfaction on the meal plan. Blood will be collected during this visit (30 ml). Upon completion of the two-week feeding period, the subjects are instructed on how to use the Atkins online diet program (https://www.atkins.com/how-it-works/free-tools) to prepare their own meals which should continue to be restricted in energy and reduced in carbohydrate at home for another two weeks.

Follow-up visit 3
Upon finishing the 4-week dietary intervention, the subject reports for visit 3. All the procedures performed will be the same as the ones performed during the baseline test (collection of 60 ml whole blood) and only questionnaires on food craving, appetite and diet consumption will be filled out.

Follow-up visit 4 and final visit 5
After the follow-up visit 4, the subject returns to the CRC two and four weeks later to assess the original four-week dietary intervention's impact on the longer-term blood pressure and heart rate. Before coming to the visit 4 and 5, the subject is instructed to collect a urine sample first thing in the morning and come to the CRC fasted. We will measure the body weight, blood pressure, heart rate and collect 30 ml whole blood at visit 4 and 60 ml blood at visit 5 to assess plasma glucose, insulin, TG, fructose and uric acid levels. Questionnaires will be administered to assess food intake pattern and food cravings at visit 5.

Human Subjects Involvement and Characteristics

Recruitment and informed consent
The study population includes male and female subjects with BMI (27-40 kg/m^2) and age 30-55y (pre-menopausal women only). The recruitment process will include the typical flyers and notices put in public spaces. These recruitment materials will be posted around the University of Missouri campus, as well as other public places such as swimming pools, churches, and grocery stores. The flyers will state the purpose of the study, eligibility criteria and will tell subjects to call into our clinical trial office for more information. On these calls, the study will be described briefly. A checklist will be gone through to perform initial screening for age, gender, and health status. Subjects who are eligible by the phone conversation will be scheduled for a screening visit and they will be given the consent form (by post,

email or fax) before they come to the screening visit. They may also bring a friend or family member to the visit. The consent form states the purpose of the study, risks, inconveniences, discomforts, and other important information about the study and has been prepared in lay terms so that it is easy to understand. During the screening visit the form is reviewed comprehensively. The subject is instructed to ask questions about any words or information that they do not clearly understand and questions will be answered by the PI and her staff. This will enable us to determine the subject's comprehension of the study. The consent form will be signed before any study procedures are initiated.

Criteria for inclusion

1. Men and premenopausal women
2. 30-55 years of age
3. Sedentary lifestyle, defined as less than three sessions of activity per week, or sessions less than 20 minutes per occasion, or less than 5000 steps per day
4. Must be able to speak, read, and write English (due to the small sample size and pilot nature of this study)
5. Overweight/obese subjects with BMI 27.0 - 40.0 kg/m^2 and any one of the following characteristics of the metabolic syndrome:
   A. Serum HDL cholesterol <40 mg/dL (1 mmol/L) in men and <50 mg/dL (1.3 mmol/L) in women
   B. Blood pressure ≥130/85 mmHg
   C. Fasting plasma glucose ≥100 mg/dL (5.6 mmol/L)
   D. Waist circumference in men of >102 cm (40 in) and in women of >88 cm (35 in)
   E. Serum triglycerides (TG) ≥150 mg/dL (1.7 mmol/L).

Criteria for Exclusion

1. Abnormal thyroid function or known liver disease
2. Diabetes or fasting glucose ≥ 125 mg/dL
3. Use of medications that interfere with protein, carbohydrate or lipid metabolism (e.g., fish oil capsules)
4. Occasional or regular tobacco use
5. History of gout
6. Uncontrolled hypertension
7. Pregnant or peri-menopausal
8. Vegetarian food restrictions (the diets consumed contain some meat, eggs and dairy)
9. Alcohol intake: females > 70 g/wk, males >140 g/wk
10. Moderate or vigorous-intensity physical activity, defined as more than 150 minutes per week of moderate-intensity, or 75 minutes a week of vigorous-intensity aerobic physical activity, or an equivalent combination of both.

Potential risks

Physical risks may include:

1. Blood drawing: During the screening, baseline and follow-up visits, blood will be drawn. The risk of blood drawing may include temporary discomfort from the needle stick, bruising, and rarely, infection. A black and blue mark may result from placement of a catheter; this can be painful, but it carries no significant risks. The total blood loss for the entire study is 300 ml.

2. Indirect calorimetry: The baseline and follow-up tests include calorimetry tests which are painless. However, persons who are uncomfortable in confined spaces may find this test slightly stressful.

3. DEXA: The radiation during DEXA is equivalent to about 2% of the average radiation dose from all sources (natural background radiation, consumer appliances, radon gas, medical tests, etc.) that a person in the United States received each year.

4. Psychological stress: The psychological stress from participation in this study is minimal. However, some of the questions about food intake and physical activity may make the subjects feel uncomfortable.

5. Arterial stiffness and blood pressure: When assessing carotid-femoral PWV, the blood pressure cuff will squeeze the arm and leg tightly; however, any discomfort will be alleviated as soon as the pressure in the cuff is released.

6. Loss of confidentiality: Any time information is collected; there is a potential risk for loss of confidentiality. Every effort will be made to keep subject's information confidential; however, this cannot be guaranteed. The information collected includes: age, ethnicity, gender, height, body weight, blood pressure, and blood test results (e.g., glucose, blood fats) and hormones (insulin). Finally, the investigator will report information to authorities in order to prevent serious harm to the subject or to others. If the Investigators suspect child, elder or disabled person abuse, they will report such concerns to proper authorities as required by law.

7. Risks to Embryo, fetus or breast-fed infant and sperm: The effects of the DEXA on the female or male reproductive systems or on a developing fetus are unknown but could cause harm. For this reason, the subject is required to avoid getting pregnant during this study. Pregnancy status will be checked using a blood test before the DEXA.

8. Other risks: There may possibly be other side effects that are unknown at this time.

Protection against risk

1. Infections from blood drawing site rarely occur. However, if any signs of infection appear, the area will be treated. Subjects will be counseled to not donate blood for 8 weeks after the study.
2. Subjects who have donated blood for any other purpose in a short time (6 weeks) will be excluded from participation. The red blood cell count (hematocrit) will be measured during screening to rule out anemia.
3. A doctor, nurse, or licensed technician will be the one to perform the blood draws.
4. The foods to be consumed will be made of commercially-available ingredients and pose no foreseeable risk. The two-week diet contains small amount carbohydrate which may cause constipation. Although this is rare, the subject will be instructed to drink eight cups of water to reduce this risk.
5. Subjects who participated in any other research study or medical procedure involving ionizing radiation exposure greater than a chest X-ray in the past 12 months will be excluded.

6. When performing carotid-femoral PWV, subjects will be informed about the cuff inflation and durance of the pressure.
7. Subjects will be told that they may refuse to answer any questions, take a break, or stop their participation at any time.

The samples may be shared with other investigators within the University of Missouri and also outside the University (The Sponsor). No identifying information will be shared.

The research plan tests the impact of weight loss by consuming a calorie-restricted, carbohydrate reduced diet and its effect on uric acid concentration and cardiovascular function. The data generated from this study will serve as important preliminary data for an NIH R01 submission. The risks to the subjects are very minimal.

Reducing Risk in Participants

The PI will monitor all procedures and the study results. The PI will report adverse events to the IRB. A data monitoring team with members not associated to this protocol will not be assembled as this study does not have a therapeutic intervention (i.e., is not a clinical trial). The special precautions are as follows:

1. Standard aseptic technique will be used for blood sampling.

2. The tests in this study have been designed for research, not for medical purposes. The subjects will be informed in the event that the PI discovers a possible abnormality; the subject will be given a copy of the screening results and advised to consult with their primary care physician to discuss further tests and/or follow-up.

3. The procedures to maintain confidentiality have been discussed above in the section "privacy."

4. The results of any test or procedure done in this study will follow internal laboratory controls.

5. This study does not involve vulnerable populations.

6. To minimize any risks, BLS certified personnel will be present during all experiments. In case of emergency, during study visits, the research nurse will be present: the University Hospital has in place a system to allow for a code Blue (i.e., cardiac arrest) by dialing 882-7979 in which trained hospital personnel will respond.

Procedures to maintain confidentiality

The health history form and all subject screening and experimental data and pertinent medical paper records will be placed in individual files and coded for de-identification. This information will only be accessible to the principal investigator, co-investigator, or approved research personnel. All records will be kept in a locked filing cabinet, which only the research personnel have access to. Computerized records of experimental data will be similarly coded and will be maintained on a password secure system. The only confidential information to be disclosed would relate to the subject's medical history. The purpose of obtaining a careful medical history is to verify that the patient meets the inclusion/exclusion criteria. All medical and biographical information will be held in strict confidentiality

and no disclosures of personal identity will be made unless specifically requested by the subject. Copies of signed consent forms, as well as the experimental log book, are kept in a locked file cabinet in the laboratory. Only subject ID numbers are used to also insure confidentiality. Participants will not be individually identified in any publications. The participants' right to privacy will be protected to the highest extent possible.

Potential benefits of the proposed research to the subjects and others

Consumption of the research diet may induce up to 3-6% body weight and potentially improve CV function in the overweight/obese subjects. All study procedures are voluntary. The data generated from this study may provide useful information to the public on the effect of weight loss and diet fructose restriction on reducing CV risk and arterial stiffness.

Adverse events and emergencies

Any serious adverse events will be reported directly to the IRB. A detailed report of all adverse events will be submitted to the IRB and a determination of related or unrelated event will be made. In case of emergency, the University Hospital has in place a system to allow for a code Blue (i.e., cardiac arrest) by dialing 882-7979 in which trained hospital personnel will respond. Our Research Nurse is certified in BLS and ACLS. She will activate the Code Response and initiate Basic Life Support until the Code Blue Team arrives.

References:

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