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Protocol Title: Safety and Tolerability of Soy Fiber in the Elderly: A Dose Escalation Study

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Abstract

Background

Apathy, defined as the absence or lack of motivation and emotional detachment, is a clinical feature of depression. Depressive symptoms and insulin resistance are conditions that are interdependent. We hypothesize that increasing insulin sensitivity in apathetic insulin resistant individuals will reduce apathy. Our preliminary studies show that a low glycemic index (GI) diet and exercise (D+E) intervention for seven days and 12 weeks increases insulin sensitivity in older adults with obesity. The key feature of a low GI diet is its high dietary fiber (DF) content. In preparation for an efficacy trial, we will conduct a dose escalation trial in the elderly with obesity and determine the maximum tolerated dose of a novel DF from whole young soy pods (soy) delivered in soft foods.

Design and Method

Aim

Determine the maximum tolerated dose of dietary fiber from soy in elderly subjects with obesity and measure fecal short chain fatty acids (SCFA) as a biomarker of compliance.

Hypothesis

Since this is a dose finding study there is no hypothesis for the maximum tolerated dose; however, fecal SCFA will increase with the dose.

Study Design

We propose to conduct a dose escalation trial. We will test the tolerability to 10 g, 20 g, and 30 g of soy containing 4 g, 8 g, and 12 g of DF respectively. At each dose, eight subjects will incorporate the foods into their usual diet for one week. We will evaluate tolerability to each dose and measure fecal SCFA as a biomarker of compliance. Subjects who satisfy the eligibility criteria and express willingness to consume the study foods will be enrolled.

At baseline, weight and vital signs will be measured. Subjects will provide a stool sample collected within the last 48 hours and frozen in containers provided for the purpose. Part of the fecal samples will be archived for future analysis. The study dietitian will interview each subject in order to assess the subject's current intake. Based on this information, subjects will be counseled on incorporating the study foods in their diet with the appropriate fluid intake. Subjects will receive the study foods containing 4g DF/10g soy/day for one week and complete a tolerability questionnaire.

Subjects will consume the soft foods containing soy along with their regular diet for one week, and will be advised to return all used and unused containers at the end of the week. Compliance will be assessed from an interview with the subject and an evaluation of used and unused containers returned by subjects. Pre-intervention tests and measures will be repeated, and adverse events will be recorded. If a dose is tolerated, subjects will receive the next escalated dose for one week (8 g DF/20 g soy/day or 12 g DF/30 g soy/day). At the end of each subject's participation in the study, a fasting blood sample will be drawn for evaluation of CBC, and blood chemistry panel as safety markers.

The results of the present study will guide the design and implementation of a clinical trial to determine the effect of a low GI D+E program on insulin sensitivity and apathy in elderly subjects with obesity and insulin resistance. These results will be significant, in that they will potentially provide a cost-effective intervention that will substantially improve the health and quality of life for the growing number of elderly individuals with obesity who have developed insulin resistance and apathy.

DESCRIPTION AND RATIONALE

The US population is expected to be comprised of approximately 24% older adults (> 65 years) by 2050.[1] More than 35% of US adults over 60 years of age are obese and nearly half of older adults are insulin resistant.[2] Depressive symptoms, which have an incidence of 6.8 per 100 person-years in the elderly (> 70 years),[3] and insulin resistance are conditions that are interdependent;[4,5] yet, the effect of improving insulin sensitivity on depressive symptoms such as apathy has not been investigated.

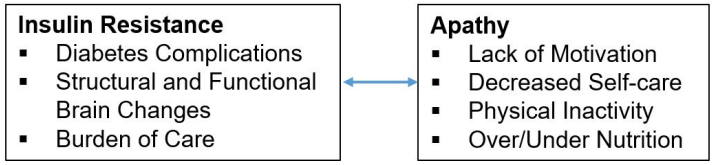

Figure 1. Relationship between insulin resistance and apathy

Apathy. Apathy is a motivation disorder that results in diminished goal directed behavior, lack of concern about one's health, and emotional indifference.[6] Apathy occurs in more than 30% of individuals with major depression and is most prevalent in depressed older adults.[7,8] The prevalence of apathy in the absence of clinical depression in cognitively-intact older adults (Geriatric Depression Scale [GDS] endorsed apathy items ≥ 2) ranges from 14% to 26%.[9,10] Impaired glucose metabolism is associated with depressive symptoms, particularly in the elderly[11-15] (Figure 1). The biological link between depression and insulin resistance appears to be an overactive dysregulated hypothalamic-pituitary-adrenal (HPA) axis. The resulting hypercortisolemia promotes obesity and insulin resistance.[11,16] Since insulin resistance and depressive symptoms are interdependent,[4,5] improving insulin sensitivity has therapeutic potential in the treatment of apathy. The presence of apathy predicts chronicity of depression, poor response of depressive symptoms to treatment, and it is associated with disability that transcends the rest of the depressive syndrome.[7,17-19] Apathy is a source of significant burden on the caregiver and current interventions lack the precision needed to address apathy and its underlying mechanisms.[20] Identifying distinct pathways for intervention will guide the course of treatment and facilitate customized interventions for a pressing, unmet, aging need.

Exercise and Insulin Resistance. The literature supports the benefits of exercise on insulin sensitivity in the overweight and obese elderly.[21-24] Different doses of aerobic exercise ranging from 90 minutes/week to 300 minutes/week have been shown to improve insulin sensitivity. Adherence to the exercise prescription varies across the studies and the dose that would most likely foster compliance with a regimen, and yet be effective, remains unclear. The appropriate exercise regimen for older insulin resistant individuals that will be of relevance in real-world clinical practice settings constitutes a significant gap in the literature. Further, the effects of exercise on apathy are unexplored; but, evidence lends support to the usefulness of exercise in treating depression and preventing its onset.[25-27]

Diet and Insulin Resistance. Some food sources of carbohydrates have a lower potential to raise blood glucose concentrations than other carbohydrate-rich food sources. This potential of individual foods, is measured by the glycemic index (GI), which provides a numerical value to represent the effect of the food on blood glucose concentrations.[28,29] In a tightly controlled study, a low GI diet and exercise (D+E) based lifestyle intervention for 12 weeks improved insulin sensitivity in obese insulin resistant older adults.[30] Supplementing the high GI diet with dietary fiber (DF) produced similar results.[30] The key feature of the low GI diet is its high DF content. Another randomized controlled trial (RCT) showed that a low GI diet that included legumes high in soluble DF reduced glycosylated hemoglobin (Hb_{A1C}) in subjects with type 2 diabetes, but the improvements in glycemia did not differ from a regular diet high in insoluble wheat DF.[31] Thus, the DF content of a low GI diet appears to be an important mediator of its effects on glucose metabolism.

Dietary Fiber in Soybean. The whole pods of young soybean (edamame) provide approximately four times the DF in an equivalent amount of the green soybean.[32] A high-DF diet prevents the stress related rise in cortisol.[33,34,35,36] The majority of the clinical and epidemiologic evidence supports the anti-depressive effects of diets high in soybeans.[37] Observational studies in Asian populations showed an association between soybean consumption and reduced incidence of type 2 diabetes and cardiovascular disease.[38-40] A meta-analysis of nine RCTs determined that consumption of whole soy products, reduced hyperglycemia.[41] Including soybean in a Western diet in the amounts consumed in Asian cultures poses a challenge. However, milled to a flour the whole pods of young soybean (soy) taste like other legume flours, and contain far more DF. Moreover, soy contains DF, protein, isoflavones, and phytochemicals in their original proportions. In rodents, soy increases fecal fat and glucose excretion, and the synthesis of short chain fatty acids (SCFA).[42] Therefore, we hypothesize that soy will potentiate the effects of a low GI diet on insulin sensitivity.

Summary. Depressive symptoms, which are common in the elderly (> 70 years), and insulin resistance are conditions that predispose to each other. Therefore, improving insulin sensitivity may be beneficial in reducing depressive symptoms in the obese elderly. Apathy, being a common depressive symptom in the elderly and among individuals with diabetes, represents a particularly attractive therapeutic target for interventions designed to improve glucose metabolism. A low glycemic index (GI) diet and exercise (D+E) intervention increases insulin sensitivity in obese older adults. To maintain the benefits of a low GI diet in the elderly, foods must be DF-dense, yet soft and tasty. The effect of whole pods of young soybean (soy) on insulin sensitivity has never been explored in humans. In preparation for conducting an efficacy trial of the effect of soy on insulin sensitivity, we will first conduct a dose escalation study in an elderly population to determine the maximum dose of fiber from soy that wil be tolerated.

HYPOTHESIS AND SPECIFIC AIMS

Edamame or green soybean in the pod has approximately four times more dietary fiber than soybean without the pod. The whole soy pod (soy), rich in phytonutrients, increases fecal fat and glucose excretion and the synthesis of short chain fatty acids in mice. The majority of the clinical and epidemiologic evidence supports the anti-depressive effects of diets high in soybeans. In previous controlled feeding and exercise studies, we found that a low GI D+E for one week (n = 32) or 12 weeks (n = 22) increases insulin sensitivity in older adults with obesity. These data form the basis of a proposal to test the hypothesis that insulin sensitivity mediates apathy in depression. No study to date has empirically tested this hypothesis. The success of the efficacy trial depends on a determination of the optimal dose of dietary fiber from soy that will be tolerated by the elderly. Therefore, the present study will determine the maximum tolerated dose of dietary fiber from soy. The results will inform the development of a randomized controlled trial (RCT) to test the efficacy of a low GI D+E program (with and without soy) to increase insulin sensitivity and reduce apathy in the elderly.

Aim. Determine the maximum tolerated dose of dietary fiber from soy in elderly subjects with obesity**.** This aim requires a dose escalation trial in the elderly with obesity. We will test the tolerability to soft foods including 10 g, 20 g, and 30 g of soy containing 4 g, 8 g, and 12 g of dietary fiber, respectively. At each dose, eight subjects will consume the foods for one week. We will evaluate tolerability to each dose and measure fecal short chain fatty acids as a biomarker of compliance with the intervention.
Hypothesis: Since this is a dose finding study there is no hypothesis for the maximum tolerated dose; however, fecal short chain fatty acids will increase with the dose.

STUDY TIMELINE

This is a dose escalation trial. We expect to complete the study within four months. Each eligible subject's participation in the study will be approximately four weeks. Primary analyses are expected to be completed within a month of the study completion.

STUDY ENDPOINTS

Eligible subjects will consume each dose of soy for one week. They will report any adverse events and complete a tolerability questionnaire. Our primary aim is to determine the maximum tolerated dose. The secondary outcome will be the change in fecal SCFA with each escalating dose.

RESEARCH DESIGN AND METHODS

To determine an acceptable dose of DF, we will conduct a dose escalation trial in eight elderly subjects with obesity and test the tolerability to soft foods containing 4 g, 8 g, and 12 g of DF/day from soy for one week at each dose. The soy contains approximately 39% DF.[42] Therefore, subjects will consume soft foods containing 10 g, 20 g, and 30 g of soy/day to provide the required DF doses and the phytonutrients in soy. This clinical trial is patterned according to the National Cancer Institute's (NCI's) recommendation for dose escalation to determine toxicity to a drug.[43] We are testing a food, and we know that relatively large doses of DF are tolerated if daily intake is divided into small portions throughout the day.[44] However, we will evaluate tolerability to DF from soy, and measure fecal SCFA as a biomarker of compliance with the intervention. The maximum recommended starting dose (MRSD) for DF may be estimated at 4 g which added to average DF intake in the US of approximately 17g[45] is not in excess of the recommended adequate intake of 25 - 38 g/day.[46] The dose escalation scheme recommended for first-in-human trials is rapid dose escalation initially (MRSD x 2) until the estimated desirable dose (8 g) is reached and then at a more cautious escalation (e.g. MRSD x 1.5).[47] Subjects will consume soft foods containing 4 g/day of DF from soy for one week, then proceed to 8 g/day for one week, followed by 12 g/day for one week, incorporated into their usual diet.

Adverse events will be recorded as they occur and collated for each week. Fecal SCFA and gastrointestinal (GI) symptoms will be evaluated at the end of each week (Figure 2).

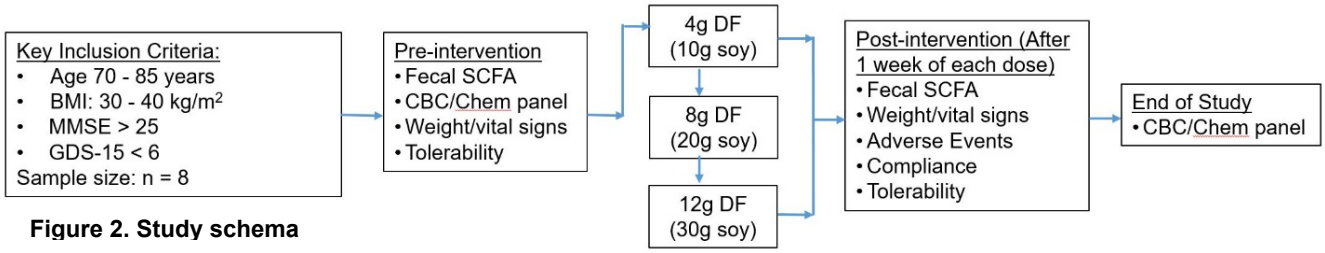

Figure 2. Study schema

Study Subjects

This study will enroll 10 healthy adult male and female subjects. Subjects will be enrolled on the basis of the following eligibility criteria:

Inclusion Criteria

- Adult (70 - 85 years).
- Body mass index between 30 and 40 kg/m^2.
- No evidence of diabetes (fasting blood sugar <126 mg/dL).
- No evidence of dementia (MMSE score > 25).

- No evidence of depression (Geriatric Depression Scale-15 [GDS-15] < 6.
- Are weight stable (< 3 kg weight change in the past three months).

Exclusion Criteria
Subjects will not be eligible for participation if they meet one or more of the following exclusion criteria:

- Have type 1 or type 2 diabetes.
- Had diagnosis of cancer (within the last five years).
- Report advanced disease of any major organs.
- Report clinically significant gastrointestinal malabsorption syndromes such as chronic diarrhea, or celiac disease.
- Have serum triglyceride concentrations > 400 mg/dl.
- Clinically significant abnormal laboratory markers (as determined by the medical investigator).
- Subjects with anticipated surgery during the study period.
- Subjects with a reported history of substance abuse or alcoholism or significant psychiatric disorder that would interfere with the ability to complete the study.
- Subjects who are current smokers or have smoked within the previous three months. Smoking is not permitted during the study.
- Subjects who are unable to provide a baseline blood or fecal sample or if they have any condition that impedes testing of the study hypothesis or makes it unsafe to consume the food being tested in the study (determined by the investigative team).
- Women on hormone replacement therapy unless weight has been stable over the last six months.

Recruitment. Subjects will be recruited through the use of printed material, targeted solicitation through the Pennington Biomedical Research Center (PBRC) email listserv and social media. Participants will complete an online screening and will be contacted by a PBRC recruiter for a brief telephone interview to assess eligibility criteria, prior to being scheduled for a clinic visit. Participation will also be solicited with the help of the LA CATS Center community advisory board. Subject eligibility criteria will be evaluated at a single screening visit. The screening visit will occur in the outpatient clinic, in the morning following confirmation of an overnight (at least ten hours) fast. Subjects who provide informed consent will proceed with the tests and measurements of the screening visit. Subjects who satisfy the eligibility criteria will be enrolled in the study. The schedule of assessments in provided in Table 1.

CLINIC VISITS

Subjects will complete one screening visit. There will be four study visits **unless a participant is intolerant to a dose and cannot be advanced to the next higher dose.**

Screening Visit (2 hours)

Subjects will report to Pennington Biomedical in the morning following an overnight fast (except for water) that began no later than 10 hours prior to the study appointment. The screening visit includes explanation of the study purpose, procedures, and signing of the informed consent. If the participant agrees to participate by signing a consent form, the following tests and measurements will be performed:

- Self-report of personal and family medical history.
- Blood draw for CBC/Chemistry 15 panel.
- Height, weight, vital signs (blood pressure, pulse, and temperature).
- Concomitant medication use.

- Questionnaires (GDS-15 and MMSE).

Eligible subjects will be contacted by a coordinator to schedule the baseline visit. Subjects will collect a stool collection kit from the coordinator before the baseline visit.

Table 1. Schedule of Assessments for Dose Response Study

Procedure	Screening 1	Baseline	Visit 1	Visit 2	Visit 3
PBRC Outpatient Unit					
Infromed Consent	x				
Height	x				
Weight, and Vital Signs	x	x	x	x	x
Chemistry 15 Panel/CBC	x				x*
Medical History Questionnaire	x				
MMSE and GDS-15	x				
Adverse Events**			x	x	x
Medications	x	x	x	x	x
Tolerability Questionnaire**		x	x	x	x
Food Dispensation		x	x	x	
Nutrition Counseling**		x	x	x	
Compliance with Study Foods			x	x	x
Fecal Sample (SCFA)***		x	x	x	x
Fecal Sample (Archives)		x	x	x	x
* Or at a prior visit if the dose is not tolerated					
** By phone call on day prior to the study visit.					
***Measured at Scioto Biosciences					

Baseline (1 hour)

Subjects will report to the clinic for the visit. The following tests and measurements will be performed:

- Collection of fecal sample.
- Measurement of weight and vital signs.
- Recording of medications.
- Dispensation of study foods.
- Nutrition counseling.
- Completion of tolerability questionnaire.
- Dispensation of stool collection kit.

Visit 1 (Week 1, 1 hour)

Assessment of adverse events, completion of the tolerability questionnaire, and nutrition counseling will be completed by phone on the day prior to the visit to avoid unnecessary food preparations in the event that the participant does not tolerate the dose. Subjects will report to the clinic for the visit and the following tests and measurements will be performed:

- Collection of fecal sample.
- Measurement of weight and vital signs.
- Recording of changes in medications.
- Dispensation of study foods.
- Assessment of compliance by metabolic kitchen staff.

- Dispensation of stool collection kit.

A blood draw for CBC/Chemistry panel will be performed if participants are not being advanced to the next dose. Participants will be advised to report after a ten hour fast.

Visit 2 (Week 2, 1 hour)

Assessment of adverse events, completion of the tolerability questionnaire, and nutrition counseling will be completed by phone on the day prior to the visit. Subjects will report to the clinic for the visit and the following tests and measurements will be performed:

- Collection of fecal sample.
- Measurement of weight and vital signs.
- Recording of changes in medications.
- Dispensation of study foods.
- Assessment of compliance by metabolic kitchen staff.
- Dispensation of stool collection kit.

A blood draw for CBC/Chemistry panel will be performed if participants are not being advanced to the next dose. Participants will be advised to report after a ten hour fast.

Visit 3 (Week 3, 1 hour)

Assessment of adverse events and completion of the tolerability questionnaire will be completed by phone on the day prior to the visit. Subjects will report to the clinic in the morning following an overnight fast (except for water) that began no later than 10 hours prior to the study appointment and the following tests and measurements will be performed:

- Collection of fecal sample.
- Measurement of weight and vital signs.
- Recording of changes in medications.
- Blood draw for CBC/Chemistry 15 panel.
- Assessment of compliance by metabolic kitchen staff.

STUDY PROCEDURES

Whole soy pods (soy). The plants will be grown under carefully monitored conditions at the LSU Agricultural Center and harvested at reproductive stage six or when the green pods contain soybeans that fill the pods. The whole pods will be processed at the United States Department of Agriculture (USDA, New Orleans, LA), Food Processing and Sensory Quality Research Unit, where scientists work with foods for research purposes. Slicing the pods into thin cross-sections injures plant cells and activates the synthesis of an anti-microbial compound (glyceollin) synthesized by the plant *de novo* in response to stress. Glyceollins have been shown to improve insulin sensitivity and cognition.[48-52] The sliced pods will be lyophilized and milled. Nutrient analysis, will be performed at Eurofins Food Chemistry Testing (Madison, WI) using methods of analysis of the Association of Official Analytical Chemists.[53]

Food Development. The soy will be incorporated in foods and prepared by the PBRC metabolic kitchen. The PBRC metabolic kitchen is staffed with research dietitians who have a primary responsibility to develop foods for research purposes, plan diets, and execute their delivery. In addition, the applicant is a registered dietitian with experience in food development across cuisines and cultures.

Nutrition Counseling. Subjects will be counseled by the study dietitian who will interview the subject in order to assess the subject's current intake. Based on this information, subjects will be counseled on incorporating the test foods in their diet. We expect that subjects will experience GI symptoms from increased DF intake, but from the applicant's practice as a

registered dietitian and evidence in the literature we know that these symptoms resolve.[54] To enhance compliance with the protocol, subjects will receive counseling on managing GI symptoms.

Food Tolerability. Subjects will complete a version of the GI symptoms questionnaire designed as a validated assessment for dyspepsia,[55] and modified to make it specific by only including the symptoms that may be experienced with high DF intake. The scale is ranked as none, mild, moderate, and severe for each of four GI symptoms (bloating, abdominal rumbling, flatulence, and abdominal pain). Additionally, subjects will report their stool frequency ranging from 0 to 9+, and stool consistency based on the Bristol Stool scale.[56]

Blood and Fecal Assessments. Chemistry 15 panel and CBC, measurements will be performed according to standard PBRC procedures for blood draws and the relevant measurement. The total amount of blood that will be drawn across the study is approximately 20 ml. Homogenized fecal samples will be analyzed for SCFA as previously described.[42,57]

MMSE. A cut-off score of 25 on the MMSE administered at screening will be used to exclude subjects with dementia.[58]

GDS-15. The Geriatric Depression Scale is validated for evaluating symptoms of apathy and depression,[59,60] and will be used as a screening tool. A score > 6 on the GDS-15 has been shown to effectively screen for depression.[61]

SAMPLE SIZE AND ANALYSIS

The primary objective of the study is to determine the maximum tolerated dose of DF. Six subjects at each dose are sufficient to detect tolerability.[43,62,63] However, we will enroll 10 subjects to test each dose of DF from soy. Dose safety will be investigated by compiling by treatment (e.g. 4 g dose, 8 g dose, 12 g dose) a list of adverse events such as frequency of headaches, nausea, vomiting. For GI symptoms the scale is ranked as none (no symptoms), mild, moderate, and severe for each of four GI symptoms and the assessment of intensity is provided in the data safety monitoring plan. Moderate and severe ratings will be considered intolerance to the dose. Stool frequency of one to three/day, or three times/per week is considered regular. Therefore, stool frequency of less than one and more than nine over a 72 hour period will be a consideration for determining intolerance. Stool consistency of Type three to five on the Bristol Stool Scale is considered normal.[64] Any other rating will be considered in the determination of intolerance. To be considered intolerance, subjects must rate stool frequency and consistency as being of moderate to severe intensity.

According to the NCI convention for dose limiting toxicity to a drug, 1) if two out of three (66%) subjects are intolerant to the dose, the next lower dose is the maximum dose tested; and 2) if one out of three (33%) are intolerant to a dose, then that dose is considered the maximum tolerated dose. Therefore for a food, 1) if five out of eight (63%) are intolerant, we will adopt the next lower dose; and 2) if three out of eight (37%) are intolerant to a dose, then that dose will be taken as the maximum tolerated dose.

A mixed effect linear model will be used to estimate changes in the fecal SCFA while accounting for subject correlation across time using all available data. The outcome will be within subject changes over time.

DATA SAFETY MONITORING

The National Institute on Aging-approved data safety monitoring plan and appointment of the safety officer is provided in the Appendix.

DATA COLLECTION AND QUALITY ASSUARANCE

The only people who will know that these patients are research participants are members of the research team. No information about them, or provided by them during the research, will be disclosed to others without their written permission, except if it is necessary to protect their rights or welfare (for example, in case of injury or emergency care), or if it is required by law. When the results of the research are published or discussed in conferences, no information will be included that would reveal the identity of these patients. All data will be kept in locked files, and subjects will be identified by codes when the data gathered in this procedure is presented or published. Authorized representatives of the National Institutes of Health may need to review records of individual participants. As a result, they may see their name; but they are bound by rules of confidentiality not to reveal the patients' identity to others.

Privacy

The subjects will be interviewed in the privacy of an exam room and their records will be protected by a secure medical records area and a password-protected electronic database monitored by the Pennington Research Computing group. Subjects will be asked to sign a written consent after reading it, having it reviewed with them by the study staff and having all their questions answered. The consent conversation will be conducted in the privacy of an exam room and the subject will be allowed to take the consent home to discuss their decision with their family or counselor, if desired. Study procedures will be conducted by trained staff in accordance with PBRC outpatient clinic standards of practice and with subjects' informed consent. Confidential subject information including medical records and test results will be available only to persons authorized by the Pennington. Information collected from subjects will be the minimum amount of data necessary to accomplish the research purposes.

Data and Specimen Management

Study participants will be assigned unique subject identification (ID) numbers. Study subject ID numbers will be used on all data collection instruments, to include questionnaires, data collection forms, and computer records. The forms used for data collection and questionnaires such as the medical history questionnaire will be the standard forms used in PBRC studies. A master list linking the participants' names and ID numbers will be kept in a password-protected computer file with access restricted to the PI and co-PI's. Data collection forms will be kept secure, or password-protected if computerized, and under the control of the PI, co-PIs, and medical investigator. Only personnel assigned to the research study by the PI will have access to the data. Hard-copy data records will be stored for a minimum of 3 years.

The PBRC has a fully integrated, campus-wide, automated data management system. All data are entered into a central database using existing methodology that has been fully validated and undergoes continuous quality assurance by the PBRC Research Computing Core. All data are backed up daily, and the Research Computing Core at the PBRC oversees all data management. The research team has extensive experience using the procedures and methods required to conduct this study. Standard operating procedures in place throughout the units at Pennington Biomedical will be utilized for repeatable, valid data collection and quality.

In accordance the standards of practice followed by the Clinical Chemistry core, blood samples will be stored frozen at PBRC until analysis can be completed. Fecal samples will be shipped to Scioto Biosciences, Indianapolis, IN for analysis. Packaging and shipping of biological samples will be overseen by the PI, shipped by the laboratory and will be completed in accordance with

International Air Transport Association regulations to ensure that viable biological samples reach their intended destination.

WITHDRAWAL OF SUBJECTS

We will attempt to retain program participants once randomized for study completion through the end of study visit. It is our desire to analyze results on all participants who were enrolled in the study. In accordance with the declaration of Helsinki/Tokyo/Venice/Hong Kong, participants have the right to withdraw from the program at any time for any reason. The investigator also has the right to withdraw participants from the program treatments in the event of inter-current illness, adverse experience, treatment failure, protocol violation, or other reasons. Should a participant decide to withdraw from treatment, all efforts will be made to complete and report follow-up observations as thoroughly as possible.

PAYMENT FOR PARTICIPATION

At the completion of all study visits and procedures, participants will be paid $125 for the time spent in the clinic. If subjects' participation in the study ends early, they will receive $25 for completion of each visit.This compensation is in line with all the other studies conducted at the Pennington Biomedical Research Center.

EMERGENCY CARE AND COMPENSATION FOR RESEARCH-RELATED INJURY

No form of compensation for medical treatment is available from the Pennington Biomedical Research Center. In the event of injury or medical illness resulting from the research procedures the research volunteer (from any group) will be referred to their physician/surgeon or a treatment facility. The Pennington Biomedical Research Center is a research facility and provides medical treatment only as part of research protocols. Should a volunteer require medical treatments, community physicians and hospitals must provide them to him/her.

SHARING OF RESULTS WITH SUBJECTS

The screening lab will be shared with the subjects on their first testing visit. At the end of the study, a manuscript will be prepared for submission to a peer-reviewed journal. A summary of the results will be posted on ClinicalTrials.gov which subjects may access. Since subjects are elderly, a written summary will be provided.

RESOURCES AVAILABLE

The outpatient research units are well equipped and staffed to carry out the requirements of this study and appropriate standards of practice are in place to ensure appropriate research procedures.

ECONOMIC BURDEN TO SUBJECTS

There are no costs for which the subjects will be responsible.

CONSENT PROCESS

Written informed consent will be obtained in the outpatient research clinic by the coordinators and one of the physicians will be available to answer questions if needed. A waiting period will be allowed, if desired by the participant. The coordinators and investigators will be available for questions throughout the study.

APPENDIX

DATA & SAFETY MONITORING PLAN (DSMP)

Candida Joan Rebello

Grant #: 5K99AG065419 - 02

Lifestyle intervention to reduce apathy in the elderly

Brief Description of Intervention: To determine an acceptable dose of dietary fiber from soy, a dose escalation trial in eight older subjects with obesity will be conducted. The tolerability to soft foods containing approximately 4 g, 8 g, and 12 g/day of dietary fiber (depending on the nutrition analysis of the soy flour) from 10g, 20g, and 30g of soy flour for one week at each dose will be assessed, and the maximum tolerated dose will be determined.

Brief Description of Project Design

- Eight subjects will consume soft foods containing soy along with their regular diet for one week.
- Compliance will be assessed from an interview with the subject and an evaluation of used and unused containers returned by subjects.
- Subjects will provide a stool sample for measurement of short chain fatty acids as an objective biomarker of compliance.
- Subjects will complete a tolerability questionnaire at the end of each week. If a dose is tolerated, subjects will receive the next escalated dose for one week.

Stage of Behavioral Intervention Development Stage I

NIH Phase III Clinical Trial? No.

Multiple Site Trial? No.

List of Specific Aims

- Determine the maximum tolerated dose of dietary fiber from soy in elderly subjects with obesity and measure fecal short chain fatty acids (SCFA) as a biomarker of compliance.

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1.0 PARTICPANTS SAFETY

1.1 Potential Risks and Benefits for Participants

The study involves the following procedures which may pose a potential risk:

- **Blood Draws.** There is the possibility of discomfort, pain, and bruising at the vein on the arm where the needle is inserted. There may also be a small risk of bleeding and a very small risk of infection at the site of the blood draw. Sterile technique and trained personnel minimize these risks.

- **Safety Assessments**. Complete blood count (CBC), and chemistry 15 panel (glucose, creatinine, potassium, uric acid, albumin, calcium, magnesium, creatine phosphokinase, alanine aminotransferase, alkaline phosphatase, iron, cholesterol [total, high density lipoprotein, low density lipoprotein], and triglycerides) reports will be used to screen for eligibility prior to enrollment and will be used as a safety assessment following completion of each dose of dietary fiber during the study. The medical investigator will review the reports, and clinically significant changes in laboratory values will be investigated, and the subject will be referred for treatment, as appropriate.

- **Whole soy pods**. The soy being tested in the study is a food. The soybean will be grown at the Louisiana State University (LSU) Agricultural Center and milled to a flour at the United States Department of Agriculture (USDA, New Orleans, LA.) Food Processing and Sensory Quality Research Unit, where scientists work with foods for research purposes. This research examines an approach that mimics soybean consumption from parts of the world where its efficacy and health benefits are known.[38-40] We expect that subjects will experience GI symptoms from increased dietary fiber intake, but from the applicant's practice as a registered dietitian and evidence in the literature we know that these symptoms resolve.[54] In the general population, supplementation with dietary fibers such as inulin and guar gum is well-tolerated at 15 g/day.[44] Therefore, we expect that supplementation of the usual diet with 4 g, 8 g, and 12 g dietary fiber from soy will be tolerated by the elderly.

- **Study Foods.** There is ordinarily no risk associated with the foods provided by the Pennington Metabolic kitchen. Additionally, subjects will be asked to inform staff about any food allergies or intolerances. A dietary questionnaire is routinely administered at the screening visit when the study involves a meal service. Research dietitians are responsible for managing the dietary component of specific study protocols. A continuous quality assurance program is followed to check food item weights, recipe procedures, packed meal and tray assembly, and food temperatures. Documentation is maintained for each study. All Metabolic Kitchen staff members receive training in food sanitation and in research diet preparation.

- **Anthropometric Measurements and Vital Signs.** The PBRC outpatient clinic staff are trained to perform these procedures in accordance with PBRC standards of practice.

- **Fecal Samples.** The clinical chemistry core staff will collect the fecal samples in accordance with standard operating practices.

In addition to the potential risks listed above, participants may experience a previously unknown risk or side effect.

Potential Benefits

Constipation is a common problem in the elderly. With the increase in dietary fiber, participants may experience benefits in their bowel movements, and as a result, in their general well-being.

1.2 Adverse Event and Serious Adverse Event Collection and Reporting

Unanticipated Problem

An unanticipated problem is any incident, experience, or outcome that meets all of the following criteria: (1) unexpected, in terms of nature, severity, or frequency, given (a) the research procedures that are described in the protocol-related documents, such as the IRB-approved research protocol and informed consent document; and (b) the characteristics of the study population; (2) related or possibly related to participation in the research (possibly related means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); (3) suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized. Unanticipated problems include other incidents, experiences, and outcomes that are not adverse events.

Adverse Events

An Adverse Event (AE) is defined as any untoward medical occurrence in a patient or clinical investigation subject, temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product. We define AE as any unfavorable and unintended sign (including a clinically significant abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of the study food.

We will define a serious adverse event (SAE) as any untoward medical occurrence that results in death, is life threatening, requires inpatient hospitalization or prolongation of existing hospitalization, results in persistent or significant disability/incapacity or in a congenital anomaly.

Safety will be assessed by recording all adverse events. The study team will inquire regarding adverse events while minimizing the chance for bias when detecting AEs/SAEs. The study team will employ open-ended and non-leading verbal questioning of the subject as the preferred method to inquire about AE occurrence. For examples, appropriate questions include: “How are you feeling?”, “Have you had any (other) medical problems since your last visit/contact?”, or “Have you taken any new medicines since your last visit/contact?” It is the responsibility of the investigator to attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. In such cases, the diagnosis will be documented as the AE/SAE and not the individual signs/symptoms.

Assessment of Intensity. An assessment of intensity for each AE and SAE reported during the study will be provided by the investigator and the investigator will assign it to one of the following categories:

Mild: An event that is easily tolerated by the subject, causing minimal discomfort and not interfering with everyday activities.

Moderate: An event that is sufficiently discomforting to interfere with normal everyday activities.

Severe: An event that prevents normal everyday activities.

An AE that is assessed as severe will not be confused with an SAE and both AEs and SAEs can be assessed as severe. Severe Adverse Event is a category utilized for rating the intensity of an event.

Assessment of Causality. The investigator is obligated to assess the relationship between study treatment and the occurrence of each AE/SAE. A "reasonable possibility" is meant to convey that there are facts/evidence or arguments to suggest a causal relationship, rather than that a relationship cannot be ruled out. The investigator will use clinical judgment to determine the relationship. Alternative causes, such as concomitant therapy, other risk factors, and the temporal relationship of the event to the study treatment will be considered and investigated.

Events meeting the definition of an AE:

- Any abnormal laboratory test results (clinical chemistry) or other safety assessments (e.g., vital signs measurements), including those that worsen from baseline, and felt to be clinically significant in the medical and scientific judgment of the investigator.
- Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition.
- New conditions detected or diagnosed after study treatment administration even though it may have been present prior to the start of the study.
- Signs, symptoms, or the clinical sequelae of a suspected interaction.
- Signs, symptoms, or the clinical sequelae of a suspected overdose of either study treatment or a concomitant medication (overdose per se will not be reported as an AE/SAE).

Events that do not meet the definition of an AE:

- Any clinically significant abnormal laboratory findings or other abnormal safety assessments that are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the subject's condition.
- Medical or surgical procedure (e.g., endoscopy, appendectomy); the condition that leads to the procedure is an AE
- Situations where an untoward medical occurrence did not occur (social and/or convenience admission to a hospital).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

AEs will be reported per IRB policies, and to the NIA on an annual basis. The reporting of AEs will follow the standards of practice as outlined by the Pennington Biomedical Research Center (PBRC) IRB in the 'Unanticipated problems Involving risks to subjects or others' policy. Once the investigator determines that an event meets the definition of an AE that must be reported, the investigator will not wait to receive additional information before notifying the responsible parties of the event and completing the appropriate forms. An assessment of causality at the time of the initial report will be provided. Email transmission of the AE data collection tool will be the preferred method to transmit this information followed by notification by telephone and/or

fax. A copy of the AE report will also be sent to the institution officials. New or updated information will be recorded in the originally completed data collection tool.

We are testing tolerability to a dietary fiber at supplementation levels known to be tolerated. In some instances, increased intake of dietary fiber may result in gastrointestinal symptoms which resolve over time. Nevertheless, should an SAE occur, a summary will be reported to NIA Program Officer and to the Safety Officer quarterly, in accordance with NIA policies. When SAEs occur that are unexpected (i.e., have not been previously reported for the study's intervention) and that are related to the intervention, they will be reported to NIA Program Officer and to the Safety Officer within 48 hours of study's knowledge of the SAE. All deaths will be reported within 24 hours of study's knowledge of death. The report of death will be submitted to NIA Program Officer and to the Safety Officer.

The investigators will abide by PBRC policy for ensuring prompt reporting to the IRB, NIA, and the Safety Officer, of any Unanticipated Problem involving risks to study participants or others (45 CFR 46.103(b)(5)). Accordingly, events meeting the definition of unanticipated problems will be reported within 48 hours, when possible, to avoid potential harm to subjects. PBRC Unanticipated Problems reporting procedures require completion and submission to the IRB of forms which include detailed information on the event and the actions that will be taken to prevent reoccurrence. Reports of Unanticipated Problems, as defined above, will be forwarded to OHRP using ohrp@osophs.dhhs.gov, within two weeks of the event.

Data Safety Monitoring Plan. For each AE, the seriousness, intensity, and relationship to study product will be assessed, documented, and supported by an entry in the subject's medical records. During the study, each subject will be carefully monitored for any adverse events. After the initial AE/SAE report is completed and sent, the investigator is required to follow each subject at subsequent visits/contacts. As currently practiced, it is planned that all AEs and SAEs will be followed in clinic until: 1) resolution; 2) the condition stabilizes; 3) the event is otherwise explained; or 4) the subject is lost to follow-up.

1.3 Protection Against Study Risks

Informed Consent Process.

The subjects will be interviewed in the privacy of an exam room and their records will be protected by a secure medical records area and a password-protected electronic database monitored by the Pennington Research Computing group. Subjects will be asked to sign a written consent after reading it, having it reviewed with them by the study staff and having all their questions answered. The consent conversation will be conducted in the privacy of an exam room and the subject will be allowed to take the consent home to discuss their decision with their family or counselor, if desired. Written informed consent will be obtained in the outpatient research clinic by the coordinators and one of the physicians will be available to answer questions if needed. A waiting period will be allowed, if desired by the participant. The coordinators and investigators will be available for questions throughout the study.

Minimizing risks

Study procedures will be conducted by trained staff in accordance with PBRC outpatient clinic standards of practice and with subjects' informed consent. Continuous monitoring by the PI and/or the medical investigator of the study and institution of a safety officer will minimize all potential risks and discomforts. Research participants will be immediately withdrawn from the

study upon evidence of any significant adverse event if the investigative team deems that the safety of the participant is in jeopardy.

2.0 INTERIM ANALYSIS

The primary objective of the study is to determine the maximum tolerated dose of dietary fiber. Dose safety will be investigated by compiling by treatment (e.g. 4 g dose, 8 g dose, 12 g dose) a list of adverse events such as frequency of headaches, nausea, vomiting. For GI symptoms the scale is ranked as none (no symptoms), mild, moderate, and severe for each of four GI symptoms and the assessment of intensity is provided in the section on adverse events. Moderate and severe ratings will be considered intolerance to the dose. Stool frequency of one to three/day, or three times/per week is considered regular. Therefore, stool frequency of less than one and more than nine over a 72-hour period will be a consideration for determining intolerance. Stool consistency of Type three to five on the Bristol Stool Scale is considered normal.[64] Any other rating will be considered in the determination of intolerance. To be considered intolerance, subjects must rate stool frequency and consistency as being of moderate to severe intensity. If a dose is tolerated, subjects will receive the next escalated dose for one week.

Stopping Rules. The study foods will be stopped, and further dosing will be halted until un-blinded safety information can be reviewed in the event that:

- A death occurs.
- Two or more subjects experience the same SAE following administration of study product.
- Based on AEs, laboratory findings, or clinical findings the Investigator determines that review of pertinent safety information is required.

Dosing may only resume if, after review of safety information, both the Investigator and IRB agree that it is safe to proceed.

3.0 Data and Safety monitoring

The Principal Investigator (PI) will be responsible for ensuring participants' safety on a daily basis. The Safety Officer advises the NIA Program staff and Principal Investigator on participant safety, evaluates the progress of the study, reviews procedures for maintaining the confidentiality of data, the quality of data collection, management, and analyses.

3.1 Frequency of Data and Safety Monitoring

Safety reports are sent to the Safety Officer at least twice a year and will include an analysis of study progress, data and safety issues.

3.2 Data Analysis and Coordination

The proposed safety officer is a physician in the PBRC Outpatient Clinic. The safety officer is independent of the study. The PI will discuss tolerability with the Safety Officer during the dose titration to determine whether the next escalation of dose is indicated. All of the data except raw data identifying individual participants will be made available to the Safety Officer and upon request to the Program Officer.

At the end of the study, a manuscript will be prepared for submission to a peer-reviewed journal. A summary of the results will be posted on ClinicalTrials.gov which subjects may access.

3.3 Content of Data and Safety Monitoring Report

The study team for this trial will prepare a written report for the Safety Officer. This report will summarize the current status of the study, including enrollment and toxicity information. All subjects in the study receive the study product and tolerability to the dose of dietary fiber is being assessed. Hence, there is no masking of data.

3.4 Safety Officer and Affiliation

The following individual has accepted the position of Safety Officer (pending NIA approval)

Raoul Manalac, M.D.

Assistant Professor, Pennington Biomedical Research Center

Dr. Manalac is trained and boarded in internal medicine with additional certifications in metabolic disease (Obesity Medicine, Clinical Lipidology) and acute inpatient medicine (Fellow of Hospital Medicine). Furthermore, he has a Master of Science in Clinical Trial Research and has served as Primary and Medical Investigators on numerous studies. He is an active clinician in both the inpatient and outpatient realms giving him familiarity with the monitoring parameters and actions necessary to address abnormal findings.

3.5 Conflict of Interest for Safety Officer

Dr. Manalac has no direct involvement with the study or conflict of interest with the investigators conducting the study. Dr. Manalac will sign a confidentiality agreement. He will be expected to follow the PBRC guidelines for disclosing conflicts of interest.

3.6 Protection of Confidentiality

Participant identities will not be revealed to the Safety Officer. The only people who will know that these individuals are research participants are members of the research team. No information about them, or provided by them during the research, will be disclosed to others without their written permission, except if it is necessary to protect their rights or welfare (for example, in case of injury or emergency care), or if it is required by law. When the results of the research are published or discussed in conferences, no information will be included that would reveal the identity of these patients.

3.7 Safety Officer Responsibilities

The responsibilities of the Safety Officer are to:

- Review the entire IRB-approved study protocol with regard to participant safety, recruitment, intervention, data management, quality control and analysis and the informed consent document.
- Recommend changes to the protocol and the informed consent form, when applicable.
- Discuss with the PI and communicate the relevant data parameters and the format of the information to be regularly reported.
- Recommend participant recruitment be initiated after receipt of a satisfactory IRB-approved protocol.
- Review study data. These data can be related to safety, recruitment, retention, protocol adherence and trial operations.
- Identify needs for additional data relevant to safety issues and request these data from the study investigators.

- Communicate with the PI and periodically review developing data on safety and endpoints.
- Consider the rationale for continuation of the study, with respect to progress of retention, protocol adherence, data management, safety issues, and outcome data and make a recommendation for or against the trial's continuation.
- Review and make recommendations on proposed protocol changes, and/or new protocols proposed during the trial.
- Provide advice on issues regarding data discrepancies found by the data auditing system or other sources.
- Review manuscripts of trial results if requested by the NIA Program Officer who may seek Safety Officer review of manuscripts reporting major outcomes prior to their submission for publication.

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