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Soy provides modest benefits on endothelial function without affecting inflammatory biomarkers in adults at cardiometabolic risk

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Abstract

Scope—Systemic inflammation, endothelial dysfunction, and oxidative stress are involved in the pathogenesis of the metabolic syndrome (MetS). Epidemiological evidence supports an association between whole soy food consumption and reduced risk of cardiovascular disease (CVD). The objective of this randomized, controlled, crossover study was to evaluate the effects of soy nut consumption on inflammatory biomarkers and endothelial function and to assess whether isoflavone metabolism to secondary products, equol and/or *O*-desmethylangolensin (ODMA), modifies these responses.

Methods and Results—*n*=17 adults at cardiometabolic risk were randomly assigned to the order of two snack interventions, soy nuts and macronutrient-matched control snack, for four weeks each, separated by a two week washout period. Outcome measures included biomarkers of inflammation, oxidative stress, and glycemic control (ELISA and clinical analyzers), endothelial function and arterial stiffness (peripheral arterial tonometry (PAT)), and isoflavone metabolites (LC-MS/MS). Results revealed that consuming soy nuts improved arterial stiffness as assessed by the augmentation index using PAT (*P*=0.03), despite lack of improvement in inflammatory biomarkers. Addition of equol and/ODMA production status as covariates did not significantly change these results.

Conclusions—Soy nuts when added to a usual diet for one month provide some benefit on arterial stiffness in adults at cardiometabolic risk.

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Keywords

cardiometabolic risk; endothelial function; inflammation; isoflavone; soy

1) Introduction

Background

Cardiovascular disease (CVD) is the leading cause of morbidity and mortality in the US [1]. Metabolic syndrome (MetS) is a clustering of risk factors for CVD and diabetes mellitus [2], affecting approximately 34% of adults in the US and its prevalence is increasing [1]. Because of the heterogeneity of MetS pathologies and more than one definition of MetS, a more broad term, cardiometabolic risk, has been coined that refers to CVD risk factors which contribute to MetS [3]. Traditional risk factors fail to predict up to 50% of the development of CVD, emphasizing the importance of other cardiometabolic risk factors [4]. Among these are multiple biological pathways that underlie the majority of vascular pathologies and involve inflammation, endothelial dysfunction [5], and oxidative stress [6].

Few investigations have studied the effects of whole foods on inflammatory biomarkers [7] and foods rich in polyphenols seem to provide significant benefit to endothelial function [8]. Epidemiological evidence demonstrates that whole soy food consumption is associated with reduced cardiometabolic risk in Asian populations [9, 10]. Further, there are limited data from randomized clinical trials on biological effects of whole soy foods consumed by Western populations. Soy nuts are soybeans that are minimally processed, non-fermented, and roasted. Soy nuts provide a concentrated source of the unique soy nutrient profile: high quality protein, PUFAs, minerals, dietary fiber, phytosterols, and isoflavones, among other bioactive compounds [11].

Of all whole soy foods, soy nuts contain the highest level of isoflavones [11–13]. In Asian countries, the mean isoflavone consumption is 25–50 mg/day [14, 15], whereas in Western countries 1–2 mg/day is typical [16, 17]. One of the primary isoflavones, daidzein, is metabolized by gut microflora and can undergo a ring cleavage to form *O*-desmethylangolensin (ODMA) and/or deoxygenation to convert to equol [18]. Not all individuals are equol producers [19] or ODMA producers [20]. Up to 60% of Asian populations and 35% Western populations are equol producers [21] and 80–95% of individuals are ODMA producers [20].

Further, the equol hypothesis has been proposed [19], which suggests that many of the biological effects of soy relate to metabolic actions due to equol, a metabolite produced by gut microflora in only some individuals [18, 19]. Equol is a biologically active isoflavone metabolite with estrogenic effects, affinity for estrogen receptors, and antioxidant activity [19]. The positive effects of soy on cardiometabolic risk factors may not be mediated primarily through estrogen receptors [22] and the food matrix may impact isoflavone metabolism [23] with additive or synergistic effects on health outcomes [11]. Few studies have examined effects of soy consumption on cardiometabolic risk factors by examining the relationship to equol and/or ODMA producers [24].

Objectives

Therefore, the purpose of this study was to determine whether soy consumption would influence cardiometabolic risk factors. The hypothesis was that the consumption of a whole soy food snack (soy nuts), compared to a macronutrient-matched control snack, would significantly attenuate inflammatory biomarkers and endothelial dysfunction in adults at cardiometabolic risk. A secondary hypothesis of this study was that participants who produce equol and/or ODMA would have significantly better inflammatory, vascular endothelial, and glycemic responses to soy consumption.

2) Materials and Methods

Study Participants

This study was approved by the University of California, Davis Institutional Review Board and informed consent of all participants was obtained. Participants were recruited from the surrounding county via flyers, newspapers, websites, and advertisements during community events. Interested participants were screened for inclusionary and exclusionary criteria. Participants who qualified after this initial screening were then invited to provide a fasting blood draw and anthropometric measurements to confirm cardiometabolic risk.

All participants were postmenopausal women and men over the age of 45 years, ensuring that participants had a low estrogen metabolic milieu. Participants were also required to meet the MetS blood pressure criteria of $\geq 130/85$ mmHg and at least one of the remaining four criteria for MetS: elevated waist circumference by population definition, elevated triglycerides ≥ 150 mg/dL (1.7 mmol/L), reduced HDL-cholesterol < 50 mg/dL (1.3 mmol/L) for women and < 40 mg/dL (1.0 mmol/L) for men, or elevated fasting glucose ≥ 100 mg/dL (5.6 mmol/L). A risk factor was also met by use of medication for these parameters [2].

Exclusionary criteria included smoking, soy allergy, unwilling to avoid soy products during the entire study, other chronic diseases (diabetes, liver disease, etc.), > 2 medications per cardiometabolic risk factor, taking other medications or supplements that interfere with study outcomes (selective estrogen receptor modulators, hormone replacement therapy, etc.), reported difficulty with phlebotomy, regular blood donor, menses within the past year, acutely ill, elite athlete, vegetarian or vegan, and on a weight loss diet.

Study Design

The study was conducted at the University of California, Davis Ragle Human Nutrition Research Center. The study design was a randomized, controlled, crossover trial, so that each participant served as his or her own control and received both snacks in a random order. The snacks consisted of soy nuts or a macronutrient-matched control snack that were provided for four weeks, separated by a two week washout period to remove any potential carryover effect (Figure 1). There were four study days: before and after each snack intervention as baseline and follow-up visits, respectively. For each visit, participants arrived after an overnight fast to the research center. Reactive hyperemia peripheral arterial tonometry (PAT) measurements were obtained first, followed by anthropometrics and a

blood draw. Participants returned unused snack containers and provided a spot urine sample at the two follow-up visits. Food records were obtained three days prior to each snack intervention and three days during each snack intervention for a total of 12 days of food records.

Snack Interventions

The soy nuts were donated in bulk (Genisoy® Food Company, Inc., Fairfield, CA) and were salted and oil roasted for palatability. The soy nuts were analyzed for isoflavone content using HPLC according to AOAC Method 2001.10. 101 mg was the target aglycone equivalent level due to its beneficial effects from other studies [25, 26]. Based on this analysis, 70 g of soy nuts provided 101 mg of aglycone equivalents that was distributed as 55 mg of genistein, 42 mg of daidzein, and 4 mg of glycitein. The soy nuts were weighed to the nearest gram and amounted to roughly 0.5 of a cup.

The control snack consisted of cookies developed and baked in the metabolic kitchen at our research center. The control snack was matched to soy nuts for kcals, macronutrients, fiber, and sodium and consisted of three small cookies comprised of vanilla whey protein powder (Body Fortress), sugar cookie mix (Betty Crocker), and generic unflavored fiber, prepackaged egg whites, and canola oil (Table 1).

The snack interventions were portioned into 30 containers so that there were enough for four weeks plus extra to account for potential spillage. Participants were instructed not to consume other soy products (soy milk, tofu, etc.) during the study to avoid potential background diet confounding effects. Participants were also instructed to maintain their usual dietary patterns and physical activity levels throughout the study.

Biochemical Assays

Approximately 65 mL of blood were drawn into serum separator tubes, heparin plasma separator tubes, and EDTA tubes. Plasma blood samples were centrifuged at 1,800 g for 15 minutes at 4°C and serum at 1,000 g for 15 minutes at room temperature. Serum and plasma samples were then aliquoted and frozen at -80°C until lab analyses could be performed in batches. Biochemical assays were analyzed at the University of California, Davis, with select assays performed at the Food and Drug Administration laboratories at the Institute for Food Safety and Health (soy nut analysis) and University of Hawai'i, Mānoa (urinary isoflavone and creatinine analysis). All were performed according to manufacturers' instructions and included appropriate quality controls.

Tumor necrosis factor (TNF)- α , IL-1 β , IL-6, and IL-10 were analyzed by ELISA (R&D Systems, Minneapolis, MN). IL-18 and oxidized LDL (oxLDL) were also analyzed by ELISAs (MBL International, Woburn, MA and Mercodia, Winston Salem, NC, respectively). High sensitivity CRP and insulin were analyzed by an Immulite 1000 Immunoassay Analyzer (Fisher Scientific, Pittsburgh, PA). Glucose, lipid panel, and clinical chemistries were analyzed by a Cobas c111 Analyzer (Fisher Scientific, Pittsburgh, PA). Baseline hemoglobin A1c was analyzed by a direct enzymatic assay (Diazyme, Poway, CA). Glucose and insulin levels were used to assess insulin resistance using the homeostasis model assessment (HOMA)-insulin resistance and HOMA- β cell function equations by

Matthews et al. [27], quantitative insulin sensitivity check index (QUICKI) equation by Katz et al. [28], and the glucose: insulin ratio [29].

Urine samples were collected in individual urine collection containers, then aliquoted and frozen at -80°C until lab analyses could be performed in batches. Urine samples were analyzed for isoflavone metabolites using LC-MS/MS according to the protocol developed by Franke et al. [30, 31]; creatinine was determined with a test kit based on the Jaffé reaction (Randox Laboratories, Crumlink, UK) on a Cobas MiraPlus CC Clinical Analyzer (Roche Diagnostics, Indianapolis, IN). From the isoflavone metabolites, equol producers and ODMA producers were calculated using equol/daidzein = 0.018 with a daidzein threshold of $2\text{ nmol/mg creatinine}$ for equol producers [21] and a detectable level of 339 nmol/L for ODMA producers [32].

Vascular Function

PAT measurements were performed using the EndoPAT 2000 system and software (Itamar Medical Inc., Caesarea, Israel). In brief, participants rested in a quiet, dimly lit, and temperature controlled room in a bed for at least 15 minutes. A PAT probe was placed on one finger on each hand to record pulse wave amplitude during baseline, occlusion, and reactive hyperemia. After a baseline period, the stimulus of occlusion was applied as a blood pressure cuff inflated to a suprasystolic blood pressure (50 mmHg above usual) for five minutes. Reactive hyperemia was induced with the blood pressure cuff deflation [33].

The PAT measurements included the reactive hyperemia index and augmentation index [34]: a higher reactive hyperemia index reflects flow mediated vasodilation [33] and a lower augmentation index reveals less arterial stiffness [35]. The reactive hyperemia index is defined as the ratio of the mean pulse wave amplitude during reactive hyperemia relative to the baseline period [36], normalized to the control arm [37]. A reactive hyperemia score of 1.67 represents endothelial dysfunction, 1.68–2.0 intermediate endothelial function, and > 2.1 protected endothelium (<http://www.itamar-medical.com/>). Augmentation index is a measure of arterial stiffness [35], which is defined as the percentage of the peak pressure attributed to the pressure of the inflection point corresponding to the reflected pulse wave [36]. A relatively lower augmentation index is considered beneficial, as a higher augmentation index has been significantly associated with more cardiometabolic risk factors [35].

Food Records

Food records were obtained to estimate current dietary intake and assess potential changes in dietary intake. The study dietitians instructed participants on how to fill out three day food records and the food records were analyzed using nutrition software (Food Processor SQL Version 10.2, ESHA Research, Salem, OR).

Statistical Analysis

A retrospective power analysis revealed adequate power for endothelial function using a sample size of $n=18$ per snack intervention, type I error rate of 0.05, observed differences, and observed variability. Primary outcome measures were endothelial function,

inflammatory biomarkers, and glycemic control indices. Curve fitting and interpolation of ELISA values were obtained by Graphpad 4.03 (Graphpad Software, La Jolla, CA). Variables not meeting normality and/or homogeneity of variance assumptions were transformed, usually by log transformation. Data analysis was performed using a linear mixed model using Proc Mixed over the snack intervention relative differences, calculated as (follow-up visit – baseline visit) / baseline visit (SAS Version 9.3, Cary, NC). Main effects were snack, sequence, and snack*sequence interaction and random effects included participant. Covariates were tested using baseline outcome measurements as continuous variables and equol producers and ODMA producers as dichotomous variables. Statistical significance was considered with $P < 0.05$ and data are presented as means \pm SD or least squares means (LSM) \pm SE.

3) Results

Participant Characteristics

Out of the 18 participants who were enrolled, one participant withdrew from the study due to a job change that required a move out of the county (Figure 2). Of the 17 participants who completed the study, 15 met the criteria for MetS [2]. Despite the addition of ~340 calories a day for eight weeks, participants had no significant change in weight (Table 2).

Biochemical Assays and Vascular Function

The inflammatory and oxidative stress biomarkers did not reveal any significant differences between soy nuts and control snack for CRP, TNF- α , IL-6, IL-18, IL-10, and oxLDL. Baseline levels of the inflammatory biomarkers, CRP and IL-6, suggest systemic inflammation [38, 39], yet the other inflammatory biomarkers fall within normal limits [39–42] (Table 4) except for IL-1 β , which was non-detectable.

Of the PAT measurements, baseline reactive hyperemia index demonstrated intermediate endothelial dysfunction and was not significantly different between soy nuts and control snack (Table 5). However, the change in augmentation index (%) was significantly different between snack intervention ($P=0.03$), with a lower LSM for soy nuts 19.7 ± 4.1 , compared to the control snack 23.5 ± 4.1 (Figure 3).

There were no significant differences between the two snack interventions at baseline for fasting glucose. Fasting glucose (mg/dL) declined after both snack interventions, but the LSM remained significantly higher after soy nuts 107.7 ± 2.2 , compared to the control snack 105.8 ± 2.2 ($P=0.02$) (Figure 4). Hemoglobin A1c revealed good long-term glycemic control and none of the participants reported a diagnosis of diabetes mellitus. Insulin was not significantly different between soy nuts and control snack, yet there was evidence of insulin resistance at baseline according to the HOMA equations [43], QUICKI equation [28], and glucose: insulin ratio [29], which did not change after the snack interventions (data not shown).

Food Records

Averaging the baseline food records revealed that the participants consumed a diet consistent with the typical American diet: approximately 48% carbohydrate, 34% fat, and 17% protein; excessive intake of saturated fat, cholesterol, and sodium; and inadequate intake of dietary fiber, calcium, and potassium, as compared to the Dietary Reference Intakes. The food records during snack interventions revealed some significant differences in dietary intake between soy nuts and control snack, reflecting expected compositional differences between the two snack interventions (Table 6).

Equol and/or ODMA Production

Six isoflavonoids were quantified: the native genistein and daidzein and their metabolites, dihydrogenistein, dihydrodaidzein, equol, and ODMA. As expected, levels of these isoflavonoids were significantly higher in response to soy nuts, compared to the control snack ($P<0.05$) (Table 3). 47% of participants ($n=8$) were equol producers and 71% ($n=12$), ODMA producers. Adding equol producers or ODMA producers as covariates in the linear mixed model did not result in any significant findings, suggesting that equol and/or ODMA production did not influence inflammatory or oxidative stress biomarkers, vascular function, or glycemic control in this population.

Compliance

Compliance to soy nuts and control snack was estimated at 91% based on counting returned snack containers. Compliance was also assessed by the food records and mean levels of isoflavonoids. As expected, a robust response in urinary isoflavones and metabolites occurred during the soy nut snack intervention. Additionally, small amounts of urinary isoflavones and metabolites were detected in samples during the control snack intervention, likely reflecting intakes of other legumes and dairy products [44]. Collectively, compliance to the snack interventions was good.

4) Discussion

Summary

This study demonstrated that soy nut consumption, compared to a control snack, significantly improved, i.e. reduced, arterial stiffness, independent of any significant effects on biomarkers of inflammation and oxidative stress. Two similarly designed studies have also investigated soy nuts and cardiometabolic risk factors [25, 26, 45–48]. Consistent with the current study, these studies found improvements in endothelial function, which were assessed by biochemical markers predictive of vascular reactivity, such as nitric oxide [45–47] and soluble vascular cell adhesion molecule-1 [25, 26]. These studies also found significant differences beyond endothelial markers, including reduced inflammatory biomarkers CRP [26, 45], TNF- α , and IL-18 [45]. Inflammation may have been modulated to a greater extent in that study because the participants were more at-risk for CVD by meeting all five of the MetS criteria and not receiving medications for cardiometabolic risk factors, so beneficial responses were more likely to occur [45–47].

Biochemical Assays and Vascular Function

The finding of improved augmentation index based on PAT measurement is consistent with the literature on soy and vascular function. A meta-analysis of 17 trials investigating isoflavone containing soy products revealed a modest improvement in endothelial dysfunction as assessed by flow mediated dilation [49], an older endothelial function measurement [8]. A separate meta-analysis examined endothelial function in postmenopausal women and found that positive responses to soy isoflavone supplementation were dependent on baseline vascular function levels [50]. Of particular relevance to the current study, a review on arterial stiffness found that four out of five studies investigating soy or isoflavones had beneficial effects using other validated measures of arterial stiffness [51]. The current study finding of improved arterial stiffness with consumption of soy nuts as measured by PAT adds to the growing literature on soy consumption and arterial compliance.

The dietary data supports the hypothesis that the soy was responsible for the beneficial changes in augmentation index in this study. Nutrient analysis of the food records confirmed that there were no significant changes in macronutrient intakes or total energy between the snack interventions that might influence vascular function. Although the PUFA content with the soy nuts was greater than the control snack, recent studies have not shown differential effects of chronic PUFA and MUFA intake on arterial stiffness [52–54]. A beneficial effect of a high potassium diet on endothelial function has been reported, but the relevance to the current snack interventions is unclear, as the levels of potassium were 1.5 fold higher than the dietary potassium during the soy nut intervention used in this study [55].

Recent studies analyzing biochemical inflammatory biomarkers and vascular adhesion molecules after soy consumption have shown mostly non-significant findings [56–61]. Two recent studies with modest favorable results in select biomarkers reflect interventions of 6–12 months and, in one, the response was apparent only in mildly hypertensive women [59, 61]. A meta-analysis of 14 trials studying postmenopausal women and soy foods containing isoflavones found that CRP was non-significantly different from controls, although a subgroup analysis revealed that soy isoflavones significantly reduced CRP in postmenopausal women with elevated CRP at baseline [56]. Another review found that only two of seven randomized clinical studies demonstrated that soy foods influenced pro-inflammatory cytokines [57] and one of these studies found that a high isoflavone soy diet actually increased IL-6 in women [62]. A daidzein rich supplement was reported to downregulate genes associated with inflammatory pathways, including Toll-like receptors, but were not mediated through the estrogen-responsive genes [22]. Despite this molecular finding, the preponderance of the evidence from multiple clinical trials and meta-analyses suggests that soy foods and components do not modulate inflammation as detected by these select circulating biochemical assays.

Some studies have found beneficial effects of soy on biomarkers of oxidative stress when investigating other soy products and different oxidative stress biomarkers: reduced plasma malondialdehyde levels [46, 60], increased total plasma antioxidant capacity [46, 63, 64], reduced LDL oxidative stress capacity lag time [23], and decreased advanced oxidation protein products [63]. Consistent with the current study, other studies have not demonstrated

a protective effect against oxidative stress [63–65]. One of these studies found that neither a high or low soy diet revealed antioxidant effects, as assessed by urinary isoprostanes in women [65]. This finding is particularly important, as isoprostanes are stable metabolic products of *in vivo* lipid oxidation and are considered the gold standard as a marker for oxidative stress [66].

In the current study, blood glucose decreased after both snack interventions but to a greater extent following the control snack. Two recent studies on soy supplementation reported improved glycemic control [67, 68]: equol supplements significantly decreased hemoglobin A1c levels in adults with MetS [67] and genistein tablets decreased glucose, insulin, and HOMA-insulin resistance in postmenopausal women with MetS [68]. However, two recent meta-analyses on glycemic control reported no overall effect of soy intake on glycemic control [69, 70]. Notably, when only studies utilizing whole soy foods or a soy diet were analyzed for glycemic control outcomes, a significant decrease in glucose was found [69]. Although this subgroup finding contrasts with the current study, soy nuts did decrease glucose, but did not reach significance within four weeks. It is possible that with increased length of exposure, the results may have achieved statistical and clinical significance.

Limitations and Strengths

This study had several limitations. First, it is possible that the effects of soy on inflammation and endothelial dysfunction occur locally at the lesion site, rather than at the systemic level, and therefore are not detected by changes in circulating inflammatory biomarkers [71]. Furthermore, there is great variability in inflammatory biomarkers and the best one to use has not yet been identified. The current study had a small sample size and short duration as a pilot trial. A larger and longer-term study may detect true differences in outcomes.

The current study is now the third study to investigate soy nut consumption [25, 26, 45–48] and the first soy nut study to use a usual diet as the background diet. It has a strong study design as a randomized, controlled, crossover study. Additionally, it is the only soy nut study to assess vascular function utilizing PAT.

Conclusion

In conclusion, consumption of soy nuts attenuates arterial stiffness, as assessed by PAT. Soy nuts did not result in clinically significant effects on inflammatory biomarkers and glycemic control. Interestingly, the ability of the individual to synthesize the isoflavone metabolites, equol and/or ODMA, did not alter the study findings. Overall, this study suggests that soy nuts have modest effects on attenuating endothelial dysfunction over four weeks in adults with features of MetS, which may contribute to the beneficial cardiovascular effects of soy consumption.

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manuscript, and FMS had primary responsibility for the final manuscript. All authors read and approved the final manuscript.

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Abbreviations

CVD	Cardiovascular Disease
CRP	C-reactive protein
DASH	Dietary Approaches to Stop Hypertension
HOMA	homeostasis model assessment
LSM	least squares means
ODMA	<i>O</i> -desmethylangolensin
oxLDL	oxidized LDL
MetS	metabolic syndrome
PAT	peripheral arterial tonometry
QUICKI	quantitative insulin sensitivity check index
TNF-α	tumor necrosis factor- α

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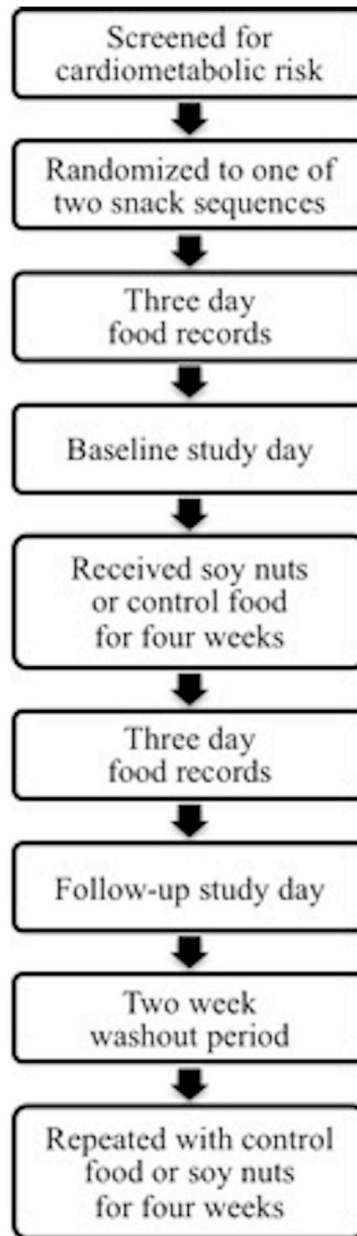


Figure 1.
Study Flow

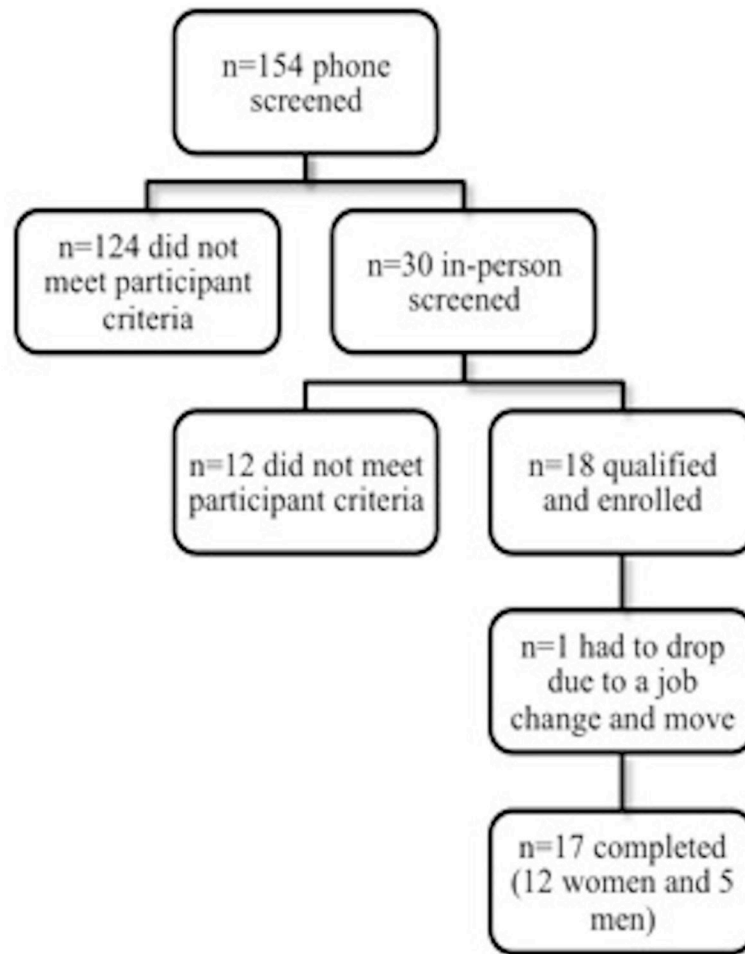


Figure 2.
Participant Flow

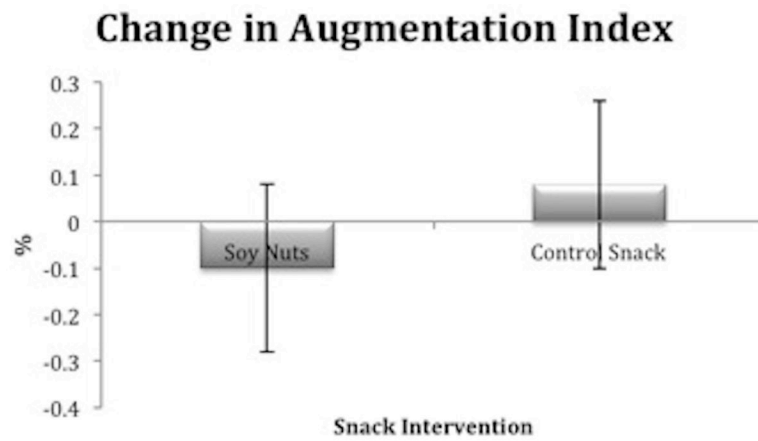


Figure 3. Relative change in augmentation index (%) was significantly different after soy nuts, compared to the control snack ($P=0.03$). The least squares means (LSM) \pm SE after soy nuts was 19.7 ± 4.1 and after control snack, 23.5 ± 4.1 .

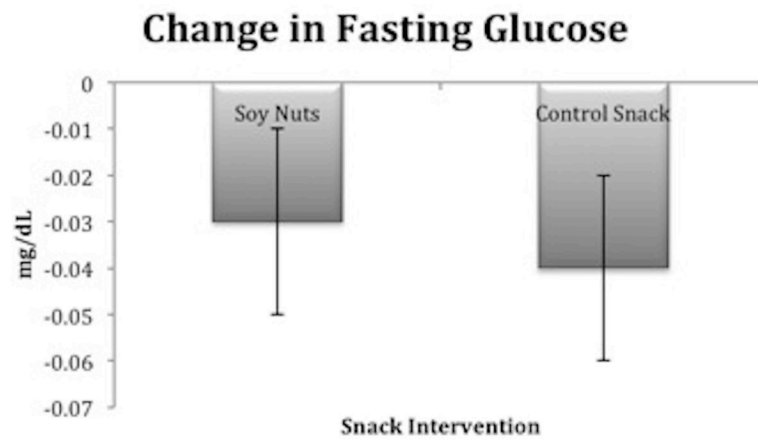


Figure 4. Relative change in glucose (mg/dL) was significantly different after soy nuts, compared to the control snack ($P=0.02$). The least squares means (LSM) \pm SE after soy nuts was 107.7 ± 2.2 and after control snack, 105.8 ± 2.2 .

Table 1

Snack Composition

Nutrient	Units	Soy Nuts^a	Control Snack^b
Calories	(kj)	1,382	1,457
	(kcal)	330	348
Fat	(g)	18	17
Sodium	(mg)	114	113
Potassium	(mg)	1,030	170
Carbohydrate	(g)	23	27
Sugars	(g)	5	5
Fiber	(g)	12	12
Protein	(g)	25	25
Isoflavones ^c	(mg)	101	Non-detectable

^a70 g of soy nuts

^b65 g of cookies

^caglycone equivalents

Table 2Participant Characteristics (n=17)^a

Characteristic	Units	Mean ± SD
Age	(years)	56 ± 5
Gender	(% women)	71
Weight	(kg)	86.7 ± 12.6
BMI	(kg/m ²)	31.2 ± 4.0
Ethnicity	(% Caucasian)	59
Metabolic syndrome ^b	(number of criteria out of 5)	3.5 ± 1.1
Waist circumference	(cm)	106.5 ± 10.6
Triglycerides	(mg/dL)	168.4 ± 78.6
HDL-cholesterol	(mg/dL)	46.4 ± 12.7
Systolic blood pressure	(mmHg)	142 ± 12
Diastolic blood pressure	(mmHg)	87 ± 10
Glucose	(mg/dL)	96.4 ± 13.3
Total cholesterol	(mg/dL)	213.4 ± 30.7
LDL-cholesterol	(mg/dL)	129.0 ± 31.1

^aParticipant characteristics were obtained from the screening visit.

^bMetabolic syndrome was defined according to the joint interim statement [1].

Table 3

Isoflavone Metabolite Levels in Urine (n=17)

Isoflavone	Units	Soy Nuts Mean \pm SD	Control Snack Mean \pm SD
Genistein	(nmol/mg creatinine)	24.4 \pm 22.5	0.5 \pm 0.9
Dihydrogenistein	(nmol/mg creatinine)	10.1 \pm 18.1	0.8 \pm 2.6
Daidzein	(nmol/mg creatinine)	54.9 \pm 78.0	1.1 \pm 2.0
Dihydrodaidzein	(nmol/mg creatinine)	32.3 \pm 42.0	1.8 \pm 3.9
Equol	(nmol/mg creatinine)	13.7 \pm 19.9	1.1 \pm 2.0
ODMA	(nmol/mg creatinine)	22.5 \pm 36.1	0.2 \pm 0.3

ODMA, *O*-desmethylangolensin.

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Table 4

Inflammatory and Oxidative Stress Biomarkers (n=17)

Biomarkers	Units	Baseline ^a or Change ^b	Soy Nuts	Control Snack	P value ^c
CRP	(mg/L)	Baseline	3.4 ± 4.7	3.7 ± 5.5	NS
		Change	1.0 ± 0.4	0.1 ± 0.4	
TNF-α	(pg/mL)	Baseline	1.3 ± 0.8	1.3 ± 0.7	NS
		Change	0.01 ± 0.05	-0.03 ± 0.05	
IL-6	(pg/mL)	Baseline	2.0 ± 1.0	1.9 ± 1.1	NS
		Change	0.08 ± 0.1	0.2 ± 0.1	
IL-18	(pg/mL)	Baseline	49.5 ± 15.2	55.5 ± 18.9	NS
		Change	0.08 ± 0.04	-0.01 ± 0.04	
IL-10	(pg/mL)	Baseline	0.2 ± 0.2	0.3 ± 0.1	NS
		Change	1.5 ± 0.8	0.3 ± 0.8	
OxLDL ^d	(U/L)	Baseline	53.0 ± 12.8	48.2 ± 8.4	NS
		Change	-0.1 ± 0.3	0.1 ± 0.3	

CRP, C-reactive protein; OxLDL, oxidized LDL; TNF-α, tumor necrosis factor-α.

^a Baseline values are depicted as mean ± SD.

^b Relative change is calculated as (follow-up visit – baseline visit) / baseline visit. These values are depicted as least squares mean (L-SM) ± SE.

^c P value corresponds to the transformed variable.

^d n=8

Table 5

Vascular and Glycemic Outcomes (n=17)

Outcomes	Units	Baseline ^d or Change ^b	Soy Nuts	Control Snack	P value ^c
Reactive hyperemia index	(-)	Baseline	2.0 ± 0.6	1.8 ± 0.6	NS
		Change	0.06 ± 0.07	0.08 ± 0.07	
Augmentation index	(%)	Baseline	25.2 ± 20.3	23.8 ± 16.5	0.03
		Change	-0.1 ± 0.2	0.08 ± 0.2	
Glucose	(mg/dL)	Baseline	109.0 ± 8.0	107.8 ± 11.1	0.02
		Change	-0.03 ± 0.02	-0.04 ± 0.02	
Insulin	(μU/mL)	Baseline	12.9 ± 6.5	12.7 ± 7.5	NS
		Change	0.08 ± 0.08	0.1 ± 0.08	

^a Values are depicted as mean ± SD.

^b Relative Change is calculated as (follow-up visit – baseline visit) / baseline visit. These values are depicted as least squares mean (LSM) ± SE.

^c P value corresponds to the transformed variable.

Table 6

Food Records (n=17)

Nutrient	Units	Baseline	Soy	Control	P value ^b
Calories	(kj)	8,884 ± 2,441	9,596 ± 3,848	8,667 ± 1,968	NS
	(kcal)	2,122 ± 583	2,292 ± 919	2,070 ± 470	NS
Fat	(g)	80.7 ± 31.4	87.4 ± 38.9	86.1 ± 28.5	NS
Saturated fat	(g)	24.6 ± 11.9	25.5 ± 13.8	20.6 ± 10.3	NS
Trans fat	(g)	0.9 ± 0.8	0.8 ± 1.1	0.7 ± 1.0	NS
MUFA	(g)	20.6 ± 9.4	20.7 ± 14.8	15.6 ± 6.5	NS
PUFA	(g)	11.4 ± 6.0	17.1 ± 8.0	8.5 ± 3.7	0.04
Cholesterol	(mg)	324 ± 162	286 ± 168	232 ± 151	NS
Sugars	(g)	86.4 ± 41.3	93.4 ± 53.5	83.1 ± 39.0	NS
Sodium	(mg)	3,008 ± 1,044	2,710 ± 1,057	2,646 ± 873	NS
Carbohydrate	(g)	254.5 ± 81.7	267.6 ± 129.0	232.2 ± 65.0	NS
Fiber	(g)	21.1 ± 7.7	34.4 ± 15.6	31.8 ± 10.5	NS
Protein	(g)	90.7 ± 26.3	108.7 ± 35.0	98.0 ± 20.2	NS
Calcium	(mg)	733 ± 264	849 ± 407	698 ± 333	NS
Potassium	(mg)	2,131 ± 782	3,220 ± 1,819	2,067 ± 935	0.04

^a Values are depicted as mean ± SD.^b P value corresponds to the difference between the snack interventions.