Original Article

Effects of Soy on Metabolic Biomarkers of Cardiovascular Disease in Elderly Women with Metabolic Syndrome

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Abstract

Background: This study ascertained the effects of soy, in the forms of textured soy protein (TSP) and soy nuts, on lipid profiles, apolipoproteins, inflammatory and prothrombotic markers, and blood pressure in elderly women diagnosed with metabolic syndrome (MetS).

Methods: This was a 12-week parallel, randomized, controlled trial conducted in rural health centers of Babol, Iran. Participants were 75 women, ages 60–70 years, who were diagnosed with MetS. Subjects were randomized to one of the following 3 groups: i) soy nut (35g/d), ii) TSP (35g/d), and iii) control. Blood biochemical markers measured at baseline and at the end of the study included: triglycerides (TG), cholesterol, HDL-C, LDL-C, VLDL-C, ApoB100, ApoAI, C-reactive protein (CRP), and fibrinogen.

Results: Soy nuts significantly improved LDL-C, VLDL-C, and ApoB100levels ($P < 0.05$), while fewer, significant improvements were observed in these variables in the TSP group compared to mean changes from baseline ($P < 0.001$). Similar results were found for ApoAI in the treatment groups ($P < 0.01$). Serum total cholesterol (TC) decreased significantly in the treatment groups compared with the control group ($P < 0.005$). Differences from the control group in terms of TG, HDL-C, fibrinogen, CRP, and blood pressure were not significant.

Conclusion: Both forms of soy improved lipid profiles. The group that consumed soy nuts had greater improvement than the TSP group. Therefore, moderate daily intake of soy may be a safe, inexpensive, and practical method to improve the risk of cardiovascular disease (CVD) and reduce the need for medical treatment.

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Keywords: Aging, dyslipidemia, inflammatory factor, metabolic syndrome, soy foods

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Introduction

Metabolic syndrome (MetS) has emerged as a cluster of risk factors for atherosclerotic diseases. Dyslipidemia with proinflammatory and procoagulant status associated with MetS ultimately leads to cardiovascular complications in these patients.[1,2] It is suggested that proinflammatory status plays an essential role in the pathophysiology of MetS through enhanced adipose tissue-derived cytokine expression and therefore, insulin resistance.[3] Fibrinogen and C-reactive protein (CRP) as acute-phase proteins increase in response to a high-cytokine state. Thus, they may be metabolically interconnected and can be used as screening tools for individuals at high risk for cardiovascular disease (CVD).[4] Thus, reducing the levels of these protein scan be a practical target for new therapies for individuals with MetS. Dyslipidemia and inflammatory markers increase with age, particularly in women due to multiple age-related physiological mechanisms and endocrine changes from menopause.[1,5] Hence older women with MetS are at high risk of heart disease.

Nutrition has a major role in MetS.[6–8] Several studies have been conducted on the effect of diet on lipid profile and inflammatory markers, especially the effect of soy intake; however, most have been performed on healthy people, or hypercholesterolemic and diabetic patients.[9–13] The results of these studies are contradictory.[14] Soy is a plant-derived estrogen that through its anti-inflammatory and antilipemic properties may protect against CVD.[10]

There are a variety of soy products. However, the manufacturing process for soy products may have destroyed some of soy's active factors. Soy nuts are roasted whole soy beans that are a non-processed, non-fermented product. In contrast, textured soy protein (TSP) obtained by manufacturing; in this process, 95% of its fat is extracted, with a reduction in many of its nutrients. Therefore, we have designed this study to determine the effects of soy nut, as the soybean natural state, compared with TSP, a processed soy product, on metabolic biomarkers of MetS in women ≥ 60 years. This research evaluates the effects of 2 types of soy on lipid profiles, apolipoproteins, CRP, fibrinogen, and blood pressure in older women diagnosed with MetS.

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Table 1. Nutrient composition of textured soy protein (TSP) and soy-nut.

Nutrients per 35 g	TSP	Soy nut
Protein (g)	18.2	13.8
Fat (g)	0.45	8.7
Total carbohydrates (g)	11.4	11.5
Fibre (g)	11.9	10.5
Total Isoflavones (mg)	96.2	117.2
Diadzein (mg)	38.5	47.6
Genistein (mg)	48.8	60.2
Glycitein (mg)	8.9	9.45

TSP = textured soy protein

Table 2. Baseline participant characteristics.

Variables	Soy nut (n = 25)	TSP (n = 25)	Control (n = 25)
Age [a]	63.8 ± 2.82	64.6 ± 2.91	64.1 ± 2.81
Menopausal age	48.2 ± 3.91	47.7 ± 4.72	48.6 ± 3.62
Currently in menopause [a]	15.5 ± 3.64	16.8 ± 61	15.5 ± 3.75
Weight (kg)[a]	72.1 ± 11.30	69.2 ± 7.61	71.0 ± 15.43
BMI (kg/m^2)[a]	28.8 ± 3.88	27.5 ± 2.59	28.5 ± 5.47
Education status [b]			
Illiterate	22(88)	23 (92)	22(88)
Literate	3(12)	2 (8)	3 (12)
Economic status [b]			
Dependent	2 (8)	1 (4)	2(8)
Independent	23 (92)	24 (96)	23 (92)
Living arrangements [b]			
Alone	4 (16)	2(8)	2 (8)
Husband only	19(76)	19 (76)	20 (80)
Children	2 (8)	4 (16)	3 (12)

[a] Values are mean ± SD; [b] Values are listed as number (%); TSP = textured soy protein

Materials and Methods

Participants

The study was approved by the Ethics Committee of the University of Putra, Malaysia and Babol University of Medical Sciences, Iran. A total of 200 women, 60–70 years old, were screened for study participation. The study was conducted in rural health centers of Babol, Iran in 2009. MetS was determined according to Adult Treatment Panel (ATP) III guidelines: 1) waist circumference > 80 cm; 2) serum HDL-C < 50 mg/dL; 3) triglycerides (TG) ≥ 150 mg/dL; 4) fasting blood glucose (FBG) ≥ 100 mg/dL; and 5) systolic blood pressure ≥ 130 mmHg, diastolic ≥ 85 mmHg. To be enrolled in the study, subjects had to have ≥ 3 of the above-mentioned criteria without the need for medical treatment of diabetes, hypertension, and hyperlipidemia. Exclusion criteria were: currently or previously using estrogen therapy; soy products or supplements; treatment with aspirin; taking antibiotics; history of CVD, hyper- and hypothyroidism; kidney, liver; breast cancer or any cancer; vegetarian diet; smoking; and allergic reaction to soy consumption. Finally, a total of 75 women, 60–70 years old, who met the inclusion criteria, were included in the study. All participants provided informed written consents.

Study procedures

This study was a 12-week open-label randomized, controlled trial. Data were collected between July and December 2009. Participants were randomized into 3 groups by the proportional randomization method using a table of random numbers generated by Microsoft Excel. Group A (n = 25) received 35 g soy nuts and group B (n = 25) took 35 g TSP daily for 3 months. Group C (n = 25), as the control group received nothing. Soy nuts and TSP were packed and given to the participants in small, 490 g bags that were to be consumed over 2 weeks, and provided a daily soy dose of 35 g. The people in group B were trained on how to prepare their meals with TSP. The soy nuts and TSP used in this study were produced and packed by Max Soy Company, Tehran, Iran. The nutrient composition of soy consumed in the study, based on Max Soy Company analysis, is shown in Table 1.

Participants were asked not to change their habitual diet and physical activity levels for the duration of the study. To assure there were no changes in diet and activity, dietary intake and physical activity levels were all measured at baseline and during each month of intervention. Each participant completed her food record (3-day food record) and physical activity questionnaire [recorded as metabolic equivalent (MET)-minute/week using the International Physical Activity Questionnaire (IPAQ)] each month and gave them to the researchers. These questionnaires were analyzed in controlling the participant's compliance to ensure that there were no changes in diet and physical activity levels throughout the treatment period.

The participants were visited every 2 weeks and were phoned weekly for monitoring compliance, and noting complaints about the soy or any changes in their health status. In every 2-week meeting, empty packages were returned and new packages distributed to be used for the next 2weeks.Monthly visits were performed to gather the completed questionnaires for physical activity and food records of the participants in all 3 groups, and to measure their blood pressures. Metabolic biomarkers were measured on all 3 groups before and after 12 weeks of intervention.

Measurements

After 10–12 hours of overnight fasting, 10 mL of venous blood was drawn, of which 1.8 mL of the blood samples were collected in citrate tubes to measure fibrinogen. The remainder of the blood samples were collected into test tubes. Serum was separated by centrifugation within 15 minutes of collection. The aliquots were frozen and stored at -80°C for subsequent analyses.

Table 3. Clinical characteristics of participants according to metabolic syndrome (MetS) criteria.

Variables	N = 75 (%)[a]	Soy nut [b]	TSP [b]	Control [b]	*P*-value
WC	63 (84.00)	93.3 ± 9.69	91.6 ± 10.92	92.4 ± 14.7	0.87
FBG	49(65.33)	104.8 ± 9.85	104.3 ± 11.12	102.5 ± 11.44	0.75
TG	69(92.00)	212.1 ± 40.31	211.9 ± 42.53	212.6 ± 48.84	0.99
HDL-C	69(92.00)	44.2 ± 6.76	43.1 ± 4.71	44.2 ± 7.25	0.78
SBP	35 (46.66)	127.3 ± 4.41	127.6 ± 4.48	127.4 ± 4.64	0.97
DBP	21 (28.00)	79.4 ± 6.47	80.6 ± 4.34	81.4 ± 6.15	0.48

TSP-Textured soy protein; WC-Waist circumference; FBG-Fasting blood glucose; TG-Triglycerides; BP-Systolic blood pressure; DBP-Diastolic blood pressure. [a]Values are number (%) of MetS components. [b]Values are mean ± SD of MetS components. TSP = textured soy protein.

Table 4. Mean ± SE of macronutrient and micronutrient intake by participants.

	Treatment Groups			
Nutrients	**Soy nut (*n* = 25)**	**TSP (*n* = 25)**	**Control (*n* = 25)**	***P*-value[1]**
Total calorie (kcal)	1943.0 ± 50.45	1939.0 ± 53.39	1959.0 ± 54.65	0.52
CHO (g) percentage	267.2 ± 30.12 55.0 ± 3.19	275.3 ± 31.14 56.8 ± 4.21	277.2 ± 30.11 56.6 ± 3.18	0.35
Protein (g) percentage	74.9 ± 8.24 15.4 ± 3.01	81.4 ± 9.24 16.8 ± 3.34	79.8 ± 8.52 16.3 ± 3.15	0.24
Total fat (g) percentage	63.9 ± 9.11[a] 29.6 ± 5.65	56.87 ± 8.52 26.4 ± 5.35	59 ± 9.73 27.1 ± 5.95	<0.05
SFA (g) percentage	11.9 ± 3.81[a] 5.5 ± 1.78	11.6 ± 2.92[a] 5.4 ± 1.31	16.5 ± 4.73 7.6 ± 2.21	<0.001
MUFA (g) percentage	20.0 ± 5.21 9.25 ± 1.56	20.5 ± 5.45 9.5 ± 1.68	19.6 ± 5.18 9.0 ± 1.54	0.51
PUFA (g) percentage	31.9 ± 7.42[ab] 14.8 ± 2.86	24.8 ± 6.58 11.5 ± 2.44	22.8 ± 6.19 10.5 ± 2.24	<0.001
Fibre (g)	39.3 ± 8.24[a]	40.8 ± 9.21[a]	25.3 ± 5.13	<0.001
Phosphorus (mg)	803.0 ± 20.41[ab]	510.0 ± 21.11	400.2 ± 19.04	<0.001
Potassium (mg)	3684.1 ± 176.44[ab]	2420.2 ± 179.56	2297.0 ± 185.32	<0.001
Calcium (mg)	1110.2 ± 92.34[a]	1080.2 ± 88.45[a]	770.1 ± 76.41	<0.001
Folic acid (mcg)	170.1 ± 30.15	168.8 ± 27.11	170.8 ± 29.14	0.82
Magnesium (mg)	450.3 ± 5.22[ab]	320.1 ± 5.31	300.0 ± 5.06	<0.001
Zinc (mg)	11.8 ± 5.62	10.2 ± 3.91	10.9 ± 4.73	0.14
Iron (mg)	20.7 ± 6.61	19.7 ± 6.91	20.3 ± 7.8	0.27
Vit. A(RE)	8282.1 ± 65.12	8380.3 ± 60.21	8250.5 ± 66.35	0.38
Vit. E (mg)	8.4 ± 1.51	8.2 ± 1.52	8.6 ± 1.41	0.22
Vit. C (mg)	70.9 ± 12.14	69.1 ± 12.15	72.1 ± 12.11	0.87
Vit. B1 (mg)	1.1 ± 0.49	1.2 ± 0.67	1.0 ± 0.59	0.52
Vit. B2 (mg)	1.7 ± 0.47	1.4 ± 0.51	1.9 ± 0.42	0.29
Vit. B6 (mg)	0.76 ± 0.11	0.71 ± 0.09	0.92 ± 0.12	0.27
Vit. B12 (mcg)	2.1 ± 0.62	1.9 ± 0.71	2.1 ± 0.60	0.26

TSP= textured soy protein; SE= standard error; CHO= carbohydrate; SFA= saturated fatty acid; MUFA= monounsaturated fatty acid; PUFA= polyunsaturated fatty acid. Food intake analyzed by Nutritionist IV, version 2.0 (software version 4.0 for windows, 1994, First Databank, San Bruno, CA). [1] *P*-value is the difference in the three groups under study by GLM Repeated Measures. [a] Significant difference with the control group; [b] Significant difference with the TSP group.

The total cholesterol (TC) and TG levels were measured using an Elitech kit from France; LDL-C, HDL-C, VLDL-C and FBG were analyzed by a Pars Azmoon kit from Iran. All lipid profiles and FBG were assayed on a Mindray-BS300 chemistry autoanalyzer (Mindray-BS300, Nanshan, Shenzhen, China). ApoAI and ApoB100 were measured by ELISA (Diagnostic Mabteck AB, Sweden). Fibrinogen was analyzed 1 hour (at the latest) after sampling by a MAHSA-YARAN kit (Iran) through quantitative determination of plasma fibrinogen by the Clauss method (clotting method) according to the manufacturer's guidelines. The quantity of serum CRP was determined by human microplate immunoenzymetric assay (AccuBind ELISA Kit, Monobind Inc., Costa Mesa, CA, USA). Serum isoflavone daidzein was randomly measured by HPLC to determine the participants' adherence.[15] Blood pressure of the participants was assessed twice on the right arm after patients were instructed to remain seated for 10 minutes; a calibrated mercury sphygmomanometer was used. The average of two seated systolic and diastolic blood pressure measurements were used for data analysis.

Statistical analysis

Statistical analysis was performed using SPSS Windows version 17. Changes from the baseline within each group were evaluated using the paired *t*-test; changes from baseline between groups were compared by ANOVA. Analysis of Covariance (ANCOVA) was also run to determine the effect of the intervention on the treatment groups after removing the variances for pre-treatment levels of the variables. The Generalized Linear Model (GLM) measure analysis was applied to detect the changes in mean of physical activity level and dietary intake of the participants during the 12-week intervention. Blood pressure was analyzed by the Kruskal-Wallis test. The two-tailed $P < 0.05$ was considered significant.

Table 5. Mean ± SD of cardiovascular disease (CVD) biomarkers before and after intervention.

	Baseline			12 week (Absolute change)			
Variables	**Soy nut**	**TSP**	**Control**	**Soy nut**	**TSP**	**Control**	***P*-value***
SBP (mm Hg)	127.3 ± 4.41	127.6 ± 4.48	127.4 ± 4.64	123.1 ± 6.56 (-4.2 ± 1.30)	124.3 ± 7.43 (-3.3 ± 1.52)	126.4 ± 4.78 (-0.95 ± 1.22)	0.20
DBP (mm Hg)	79.4 ± 6.47	80.6 ± 4.34	81.4 ± 6.15	75.8 ± 8.68 (-3.5 ± 1.81)	78.6 ± 5.26 (-1.9 ± 1.36)	80.0 ± 2.73 (-1.4 ± 1.15)	0.13
TC(mg/dL)	229.9 ± 25.17	229.4 ± 28.03	233.2 ± 23.86	200.7 ± 23.76 (-29.2 ± 3.56)[ab]	205.2 ± 26.60 (-24.3 ± 3.54)[ab]	224.5 ± 23.80 (-8.7 ± 5.47)	<0.001
TG(mg/dL)	212.1 ± 40.30	211.9 ± 42.50	212.6 ± 48.80	199.8 ± 42.4 (-12.2 ± 1.86)[a]	200.3 ± 43.5 (-11.6 ± 1.95)[a]	208.5 ± 49.4 (-4.1 ± 1.12)	0.14
HDL-C (mg/dL)	44.2 ± 6.76	43.0 ± 4.71	44.2 ± 7.25	46.3 ± 5.7 (2.1 ± 0.49)[a]	44.8 ± 4.4 (1.7 ± 0.64)[a]	43.7 ± 7.8 (-0.57 ± 1.46)	0.08
LDL-C (mg/dL)	154.0 ± 28.40	154.7 ± 28.8	152.3 ± 25.00	131.0 ± 25.27 (-23.1 ± 2.03)[ab]	134.5 ± 27.1 (-20.1 ± 3.00)[ab]	151.5 ± 29.8 (-0.80 ± 3.84)	<0.001
VLDL-C (mg/dL)	41.7 ± 7.69	42.4 ± 8.50	42.4 ± 10	34.5 ± 8.9 (-7.1 ± 1.11)[ab]	37.4 ± 9.7 (-5 ± 0.67)[ab]	42.0 ± 9.9 (-0.37 ± 1.23)	<0.001
ApoAI (g/L)	1.8 ± 1.02	1.8 ± 1.10	1.6 ± 1	2.0 ± 1 (0.19 ± 0.03)[ab]	1.9 ± 1 (0.18 ± 0.03)[ab]	1.6 ± 0.9 (-0.02± 0.01)	<0.001
ApoB100 (g/L)	1.5 ± 0.48	1.5 ± 0.50	1. 5 ± 0.8	1.2 ± 0.4† (0.31 ± 0.04)[ab]	1.2 ± 0.5 (-0.28 ± 0.01)[ab]	1.6 ± 0.7 (0.06 ± 0.05)	<0.001
CRP (μg/mL)	3.2 ± 1.80	3.1 ± 2.16	3.0 ± 2.28	2.9 ± 1.9 (-0.25 ± 0.16)	2.9 ± 1.9 (-0.16 ± 0.17)	3.0 ± 1.54 (-0.04 ± 0.32)	0.82
Fibrinogen (mg/dL)	316.7 ± 33.79	316.5 ± 42.61	314.2 ± 42.95	297.8 ± 29.75 (18.8 ± 8.98)	302.0 ± 30.57 (-14.5 ± 9.53)	306.4 ± 30.2 (-7.8 ± 6.24)	0.55

TSP= textured soy protein; SD= standard deviation; SBP= systolic blood pressure; DBP= diastolic blood pressure; HDL-C= high density lipoprotein; LDL-C= low density lipoprotein; VLDL-C= very low density lipoprotein; Apo AI= apolipoprotein AI, ApoB100= apolipoprotein B100; CRP= C-reactive protein. Absolute change refers to the change from the baseline (week 12, baseline values).*Significant difference between treatment groups compared to the control group (ANCOVA; $P < 0.05$). [a] Significant difference between after and before (paired *t*-test). [b] Significant difference with the control group (ANOVA).

Results

All participants completed the study. Both the soy nut and TSP were well tolerated. There were no serious complaints pertaining to consumption of soy, except for flatulence in a small number of individuals. Demographic and clinical characteristics of the women at baseline are presented in Tables 2 and 3. There were no significant differences among the groups with regard to baseline variables. Results of the food record analysis are presented in Table 4. There were no significant differences in energy, protein, carbohydrate, monosaturated fat and the most micronutrients between the treatment groups and the control group. However, total fat, saturated fat, polyunsaturated fat, fiber, as well as some of the micronutrients were significantly different in the treatment groups compared to the control group. The activity level (MET-minutes/week) of the participants remained the same during the study (Figure 1). These results suggested good compliance of the participants.

Table 5 shows the mean values of biomarkers of CVD in all groups at baseline and after 12 weeks of intervention with their absolute changes. No significant difference in the baseline values of these variables was observed. Comparison of lipid profile values before and after the intervention within the groups showed significant changes within the treatment groups for all variables of lipid profiles ($P < 0.001$). The most change was observed in the soy nut group. Changes from baseline showed significant differences between the groups for lipid profiles. A post-*hoc* comparison of the groups showed a significant difference between the control group, the soy nut and TSP groups with regards to TC, LDL-C, VLDL-C, ApoB100 and ApoAI. The greatest change was seen in the soy nut group.

ANCOVA confirmed the outcomes obtained from the previous analysis. Following soy nut and TSP intervention, there were significant differences in the mean TC ($P < 0.001$), LDL-C ($P < 0.001$), VLDL-C ($P < 0.001$), ApoAI ($P < 0.001$), and ApoB100 ($P < 0.001$) between the treatment and control groups. There were no significant differences in mean changes of CRP, fibrinogen, and blood pressure in the groups. The mean change for TG and HDL-C was not significant, although they improved more in the treatment groups (Table 5). A significant difference between the 2 treatment groups for lipid profile was not seen. Compared with the control group, the serum isoflavonedaidzein increased significantly in the soy nut (percentage change: 58%; $P < 0.001$) and TSP (percentage change: 39.9%; $P < 0.001$) groups.

Discussion

The results of the present study showed that daily consumption of 35 g of soy, in the form of soy nuts and TSP for 12 weeks had beneficial effects on lipid profiles and apolipoproteins, but no effect on CRP, fibrinogen, and blood pressure in elderly women with MetS. The effects of soy nuts were of more benefit in comparison with TSP.

In this study, consumption of both soy nuts and TSP resulted in decreased serum TC, LDL-C, VLDL-C, and ApoB100 levels and

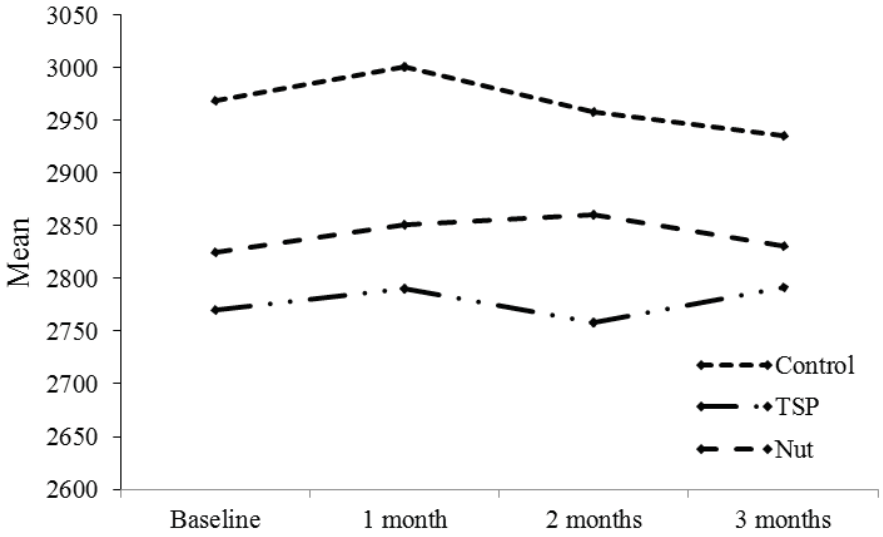

Figure 1. Mean changes of total physical activity in the groups during the study.

increased ApoAI. TG decreased by 5.8% and HDL-C increased by 4.7% in the soy nut group, whereas there was a 5.5% decrease in TG and a 4.0% increase in HDL-C in the TSP group, which was not statistically significant. The results of this intervention were consistent with the results of 3 comprehensive reviews on soy consumption and measures of cardiovascular health.[16–18] All 3 reports concluded that there was a significant effect of soynut[16] and TSP[17,18] isoflavones when consumed at levels similar to this research on LDL-C (11%–15.3%), TC (7.5%–17.2%), and ApoB100 (8%–14.6%) but not on HDL-Cor TG concentrations.[17–19] The clinical significance of soy intake on HDL-C concentration has remained controversial. However, among the previous reports, even those with a high isoflavone intake, only a few studies have noted significant changes in HDL-C levels.[11] The varying results in different studies can be addressed by Zhan and Ho's study. They, in a meta-analysis on 23 clinical trials, reported that higher initial TC concentrations and intakes > 80 mg isoflavone are more effective on lipid profiles. The strongest effects of soy protein on decreasing TC and LDL-C levels occurred within the short initial period of intervention; whereas improvements in HDL-C levels were only observed in lengthy studies of more than12 weeks.[20] Anotherexplanation may be the difference in 7S globulin concentration among soy varieties in the different studies.[21–22]

The results of this study, which was consistent with 2 recent studies, showed that serum levels of ApoAI increased significantly in the treatment groups.[17,19] The significant increase in ApoAI did not correspond with the results of HDL-C because no significant increase in the level of HDL-C was observed. The difference in change in ApoAI (11%) versus the change in HDL-C (4.7%) is unclear because there is one ApoA molecule in the HDL particle. One explanation could be that the particle size of HDL in hyperlipidemic subjects tends to be smaller, which indicates that HDL-C maturation might be abnormal in hyperlipidemic subjects.[23] Another argument is that the HDL-C levels in contrast to ApoAI levels are calculated, but not measured, which may have led to an underestimation in HDL-C change. Hermansen et al. have also found the apparent differences in changes of ApoB (30%) versus changes in LDL-C (10%).[24] However, though improvements in HDL-C and TG measurements were not significant, their importance was similar to reductions in TG and HDL-C levels observed in other studies.[20,25] This has possible clinical relevance, particularly since TG and HDL-C levels are stronger predictors of cardiovascular risk in women than in men.

The proposed mechanisms for a hypocholesterolemic effect of soy are: 1) the higher arginine to lysine and methionine amino acid profile of soy protein[17]; 2) up-regulation of LDL receptors by the 7S globulin protein exists in soy[21–22]; 3) soy fibre content[26]; 4) isoflavone content[27]; and 5) trace components such as plant sterols, phytates, folates and oxalates that could be altered by the processing method of the soy products.[17] Regarding the use of soy nuts and TSP in this study, Matthan et al. have stated that soy protein rather than soy isoflavones are useful in reducing serum lipid. Interestingly, they have shown that processing of the soy products appears to have a slight effect on their influence on lipids.[17] This finding confirms the results of our study where TSP and soy nuts both positively improved lipid profiles, though this effect is less than soy nuts. By comparing the composition of food in the two groups (Table 4), this difference might be attributed to higher values of PUFA, isoflavone, and micronutrients such as magnesium, calcium, potassium and phosphorus in the soy nut compared to TSP.

The results of this study showed that consumption of soy nuts and TSP led to 8.2% and 5.43% reduction in CRP levels, though this difference was not significant. Researchers showed that genistein has an anti-inflammatory effect both in vitro and *in vivo*.[13] Although a recent meta-analysis that assessed 14 randomized clinical trials reported that neither soy foods nor soy isoflavone impacted biomarkers of inflammation.[28] Results of a recent study were consistent with many published studies that evaluated the effects of short-term consumption of soy on CRP concentration[17,29–33] while others have shown the positive effects of soy on CRP in long-term experiments.[34–35] Considering all the factors, a possible explanation for the differences in results is that the dose that has been used in this study (35 g/d) was insufficient; higher amounts might have produced better results for CRP. However, high doses may be connected to other health risks.[36] It is possible that the effects mediated

by the isoflavone are too modest to be detected over the 3-month study period, while most effective trials about the use of soy and CRP levels have been undertaken for approximately 1 year.[34,35] Participants' overall baseline CRP level in this study was 3.1 µg/mL. Possibly soy protein has a positive effect only in persons with more elevated CRP levels.

The findings suggested that hyperfibrinogenemia (> 350 mg/dL) as a component of MetS could possibly explain the increased cardiovascular risk.[37] According to reports, the formation of thrombin and platelet activity is inhibited by genistein *in vitro*.[38] In this study, although procoagulant fibrinogen levels did not reach statistical significance, it decreased 5.9% by the consumption of 117 mg isoflavone vs. a 4.6% decrease with consumption of 96 mg isoflavone. Dent et al. showed no change in fibrinogen with consumption of 40 g/d soy protein in 6 months.[39] In a study by Hermansen et al., the consumption of 50 g/d soy protein with 165 mg/day isoflavone in diabetic patients resulted in a non-significant decrease in fibrinogen levels.[24] These results were consistent with those obtained in the current study.

In the present study, TSP and soy nut did not create a significant reduction in blood pressure. Some studies have reported beneficial effects of soybeans on blood pressure.[16,40] It is not quite clear whether this beneficial effect is related to the isoflavone content, soy protein or higher concentrations of proteins rich in specific amino acids.[39] However, several studies did not report significant effects of soy isoflavone intake on blood pressure.[11,17,24,29] This difference can partly be related to the study population since studies about the effects of soy on blood pressure have mostly been undertaken in healthy or hypertensive people in different age and sex groups; the current study has been conducted exclusively on elderly women with MetS. Physiologic changes during the aging process, along with the MetS can complicate the response to treatment.

We did not take into consideration equol production capacity of individuals in response to soy intake, thus more research is needed to investigate the relationship between equol production and response to soy intake**.** There was no placebo in the study since participants in the treatment groups were consuming natural soy products rather than medications, thus it was not feasible to use a placebo for the control group. We aimed to determine the net effect of intervention on people who were on a normal daily diet. Although this study was an open-label randomized, controlled trial, laboratory and research personnel who performed blood sampling, biochemical and statistical analyses were blinded to the treatment interventions, therefore reducing study bias. This study attempted to additionally reduce bias by having an increased sample size, appropriate randomization strategy, and good compliance by the participants.

The results of this study showed that including soy as part of a daily diet has a beneficial effect on atherogenic lipids in older women with MetS. The effect of soy nut was more significant than TSP. This study has not shown significant improvements in blood pressure, CRP and fibrinogen levels. Although the soy effect on lipid profiles is better understood, the anti-inflammatory and anticoagulant mechanisms of soy in humans are not clear. More research is needed to further study these effects.

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