Modest Protective Effects of Isoflavones from a Red Clover-Derived Dietary Supplement on Cardiovascular Disease Risk Factors in Perimenopausal Women, and Evidence of an Interaction with ApoE Genotype in 49–65 Year-Old Women[1]

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ABSTRACT Data suggest that soy protein, a source of isoflavones, may have favorable effects on cardiovascular risk factors. Women ($n = 205$), ages 49–65 y, were randomized into this double blind, placebo-controlled trial of 43.5 mg red clover–derived isoflavones/d. A total of 177 women completed the trial. There were no differences between treatments for changes from baseline to 12 mo in total cholesterol, LDL cholesterol, triglycerides, HDL cholesterol, systolic and diastolic blood pressures, fibrinogen, and plasminogen activator inhibitor type 1 (PAI-1) ($P \geq 0.1$). Interactions between treatment and menopausal status were significant for changes in triglycerides and PAI-1 ($P = 0.02$ and $P = 0.01$), and changes were significant among perimenopausal women. In the isoflavone and placebo groups, changes in triglycerides were -0.2 ± 0.6 and 0.4 ± 0.6 mmol/L, $P = 0.02$, and changes in PAI-1 were -3.06 ± 5.88 and 4.95 ± 6.25 IU/L, $P = 0.004$, respectively. Interactions between apolipoprotein E (apoE) genotype and treatment tended to be significant for changes in total and LDL cholesterol ($P = 0.06$ and $P = 0.05$), and differences between treatments were significant in E2/E3 women. In the isoflavone and placebo groups, changes in total cholesterol were -0.61 ± 0.79 and 0.18 ± 0.79 mmol/L, $P = 0.03$, and changes in LDL cholesterol were -0.84 ± 0.79 and -0.04 ± 0.69 mmol/L, $P = 0.02$, respectively. Although there were potentially beneficial changes in triglycerides and PAI-1 among perimenopausal women consuming isoflavones, this study suggests that isoflavones alone are not responsible for the well-documented effects of soy protein on blood lipids. A larger study is required to confirm the effect modification by apoE genotype. J. Nutr. 134: 1759–1764, 2004.

KEY WORDS: • *isoflavone* • *phytoestrogen* • *cardiovascular disease* • *apoE* • *randomized controlled trial*

Elevated levels of cholesterol in the blood are a major determinant of heart disease, and it has been estimated that for every 1% reduction in blood cholesterol there is a corresponding 2.5% reduction in the incidence of heart disease (1). Thus, strategies to reduce blood cholesterol could result in considerable reductions in the number of deaths from heart disease.

It has been known since the 1940s that substitution of animal protein in the diet for soy protein can have beneficial effects on blood lipids, lipoproteins, and atherosclerosis (2). In 1995, a meta-analysis of 38 controlled clinical trials in humans provided evidence that an average daily intake of 47 g soy protein was associated with significant reductions in total cholesterol, LDL cholesterol, and triglycerides, and that changes in total and LDL cholesterol were directly related to the initial serum cholesterol concentration (3).

Soy protein is an important source of isoflavones (4), and several investigators have attempted to assess the contribution of isoflavones to the lipid-lowering effects of soy protein. However, the data are somewhat conflicting. Studies in animals fed intact soy protein or alcohol-washed soy protein (containing little or no isoflavones) provided some evidence that the isoflavone component of soy may be responsible for the lipid-lowering effects (5–7). However, Greaves et al. (8,9) did not see a protective effect of an isoflavone rich soy extract that was added to casein on the blood lipid profile of ovariectomized monkeys, whereas there was a significant lipid-lowering effect with intact soy protein. Similarly, in humans, some studies have shown significant blood lipid-lowering effects of intact soy protein compared with alcohol-washed soy protein (10), but others have shown little or no difference between soy protein containing isoflavones and soy protein with a low isoflavone content (11). It is possible that some other component of soy, such as saponins, plant sterols, or proteins, might be acting in combination with, or independently of the isoflavones to produce lipid-lowering effects. In examining isoflavone supplements, a study of a red clover–derived isoflavone supplement demonstrated a significant increase in serum HDL

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cholesterol after 6 mo (although there was no control group for comparison) (12), but there was little or no effect in another study using 2 different doses of isoflavones (43.5 and 87 mg/d) (13). Among postmenopausal Japanese women taking capsules of a soy isoflavone extract, there was a significant decrease in total and LDL cholesterol (14), but other studies of soy-derived isoflavone supplements showed no effect on blood lipids (15–18).

Some (19–22), but not all (15), studies showed significant reductions in blood pressure with soy protein interventions, and, similar to the effects of soy protein on blood lipids, the effects may be related to the degree of hypertension at baseline. Fibrinogen and plasminogen activator inhibitor type 1 (PAI-1)[3] are involved in the clotting process, and elevated levels are important risk factors for cardiovascular disease (CVD) (23–27). However, a few recent studies showed no effect of soy protein interventions on fibrinogen or PAI-1 (28–31).

The aim of the present study was to examine the effects of a red clover–derived isoflavone supplement, providing 26 mg biochanin A, 16 mg formononetin, 1 mg genistein, and 0.5 mg daidzein daily for 1 y, on some risk factors for CVD, in a randomized, double-blind, placebo-controlled trial. The apolipoprotein E (apoE) genotype is an important factor affecting blood lipid profiles (32); therefore, women were genotyped for polymorphism in the gene that encodes for apoE to determine potential gene-treatment interactions.

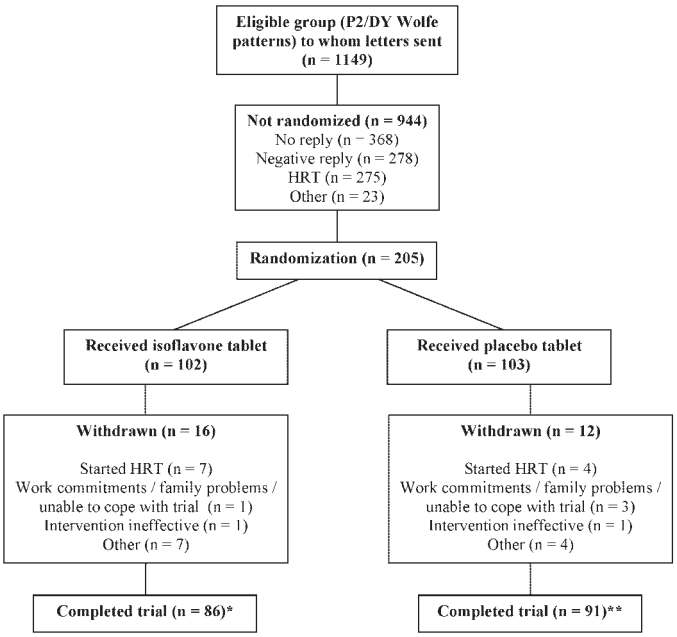

FIGURE 1 Flow chart describing the progress of volunteers through the trial. *Includes 2 women who completed the trial but were excluded from all analyses because they took oral contraceptives (OCs) or they were treated for alcoholism. **Includes 1 woman who completed the trial but was excluded from all analyses because she took OCs.

SUBJECTS AND METHODS

Study participants and intervention. Between November 1997 and May 1999, women were recruited from the Breast Screening Unit, Addenbrooke's Hospital, Cambridge, UK. The primary outcome measure was breast density, and women were selected for the study according to the extent of dense tissue on their most recent mammogram; mammograms from 1908 women were classified according to their Wolfe pattern (33), and women with Wolfe's P2 or DY breast patterns (i.e., dense breast patterns; $n = 1149$, 60% of all mammograms classified) were sent a recruitment letter, which contained a short description of the study and a reply slip. Full details of recruitment procedures were published elsewhere (34). A total of 205 women were randomized to receive a daily isoflavone tablet (Promensil, Novogen) or a placebo of identical appearance (**Fig. 1**). The isoflavone content of the Promensil tablets was not measured in our laboratory, but an independent study showed that they contained the quantity and type of isoflavones stated by the manufacturer (35). Randomization was performed using random number generation in Microsoft Excel, and researchers and study participants were unaware of the tablet allocation throughout the study. The code regarding who had been taking isoflavone or placebo tablets was broken when all volunteers had completed all stages of the study. All study procedures were approved by the Dunn Human Nutrition Unit Ethics Committee and the Cambridge Local Research Ethics Committee.

Urine collections. Women were asked to make 24-h urine collections at baseline and after ~6 and 12 mo on the study; the completeness of all urine collections was assessed using the *p*-aminobenzoic acid (PABA)-check test (36). Samples containing 85–110% of the ingested PABA were designated satisfactory. For samples with PABA recoveries between 70 and 85%, indicating that all tablets had been taken but that the urine collection was incomplete, the urinary excretion of isoflavones was adjusted up to 93% PABA recovery (37). Samples with <70% recovery were designated incomplete. Samples with >110% PABA recovery were designated unsatisfactory, because additional sources of PABA might have been consumed (e.g., a multivitamin), and completeness of the sample could not be accurately determined.

Urinary excretion of genistein, daidzein, formononetin, and biochanin A were measured by HPLC using a modified method of Setchell et al. (38) and Franke et al. (39); for full details of the method see (34). Baseline or final urine samples were unavailable for 2 women in the isoflavone group and 3 women in the placebo group.

Blood samples. Blood samples were taken from fasting subjects by the study nurse at baseline, and after ~12 mo on the study. Participants were asked to refrain from eating or drinking beverages (except water) from midnight until after the sample had been taken the following morning. A total of 35.5 mL blood was drawn at each visit; the first sample after venipuncture was drawn into a 4.5-mL sodium citrate tube for fibrinogen and PAI-1 analysis; tubes were kept cool before centrifugation. Tubes were centrifuged for 10 min at 2000 rpm at 5°C and after centrifugation, plasma was removed and stored at −20°C. Fibrinogen concentrations were determined according to the method of Clauss (40). The CV was 3.57%. PAI-1 activity was determined with an indirect enzymatic method (Spectrolyse, Biopool), and the CV was 7.91%. Baseline and 12 mo blood samples from each participant were analyzed within the same batch.

Blood (9.0 mL) was drawn into a serum tube and left at room temperature for at least 1 h before centrifugation. Tubes were centrifuged for 10 min at 2000 rpm at 5°C, and after centrifugation, serum was removed and stored at −20°C. Total and HDL cholesterol were measured using a Technicon RA1000 analyzer and enzymatic kit T01–1684-01, and total triglycerides were measured using enzymatic kit T01–1868-01 (Miles, Inc.). LDL cholesterol concentrations were determined using the Friedewald formula (41). The CVs for total cholesterol were 2.7% at a level of 2.59 mmol/L and 1.4% at a level of 8.75 mmol/L; triglycerides 1.9% at levels of 2.43 and 3.13 mmol/L; and HDL cholesterol 1.8% at a level of 1.39 mmol/L. All blood samples from each participant were analyzed within the same batch.

Estradiol was measured using previously described methodology (42), and follicle-stimulating hormone (FSH) was measured by enzyme immunoassay on an Abbott Axsym automated analyzer (Abbott Diagnostics). Menopausal status was determined using levels of es-

[3] Abbreviations used: apoE, apolipoprotein E; CVD, cardiovascular disease; FSH, follicle-stimulating hormone; PABA, *p*-aminobenzoic acid; PAI-1, plasminogen activator inhibitor type 1.

tradiol and FSH in the baseline serum sample only; women were classified as premenopausal if FSH < 30 IU/L and estradiol > 100 pmol/L; postmenopausal if FSH > 30 IU/L and estradiol < 100 pmol/L; and perimenopausal if FSH > 30 IU/L and estradiol > 100 pmol/L or if FSH < 30 IU/L and estradiol < 100 pmol/L. In addition, if a woman had noted on her baseline questionnaire that she was currently menstruating, but her baseline hormone profile was that of a postmenopausal woman (i.e., FSH > 30 IU/L and estradiol < 100 pmol/L), she was classified as perimenopausal.

Whole blood (~1 mL) was removed from the baseline lithium heparin tube and was stored at −20°C for subsequent DNA extraction; DNA was extracted from 200 μL whole blood using a QIAGEN kit. The presence of polymorphic restriction sites in the apoE gene was detected via an HhaI digestion according to the method of Loktionov et al. (43). Genotype data were available for 78 and 84 women in the isoflavone and placebo groups, respectively.

Blood pressure. Blood pressure was measured at baseline and after ~12 mo using an Omron M4 blood pressure monitor (Hutchings Healthcare). Before the measurement, all study participants were required to have been sitting down for at least 5 min. Information on time of measurement, past history of high or low blood pressure, and medications taken for maintenance of blood pressure (if any) was collected before each measurement.

Data analysis. All statistical analyses were performed using the SAS statistical package version 6.12 (SAS Institute). A *P*-value of <0.05 was considered statistically significant, and data are presented as means ± SD. χ^2 analysis was used to test for differences between treatment groups for the number of withdrawals, and for the numbers of women who were pre-, peri-, and postmenopausal. One-way ANOVA was used to determine the effect of apoE genotype on baseline serum lipids, blood clotting factors, and blood pressure. Data on the E2/E4 genotypes were not included in analyses by genotype due to the small number of women with this genotype ($n = 3$). Changes in serum lipids, blood clotting factors, and blood pressure were calculated as 12-mo data minus baseline data. There was a significant difference between treatment groups for baseline levels of LDL cholesterol (4.21 ± 0.94 and 3.88 ± 1.00 among women in the isoflavone and placebo groups, respectively, $P = 0.03$); therefore, changes in LDL cholesterol were also assessed as a percentage of baseline, i.e., [(12 mo data − baseline data)/baseline data] × 100. Unpaired *t* tests were used to test for differences between treatment groups for changes from baseline to 12 mo. A general linear model (PROC GLM) was used to determine interactions between apoE genotype and baseline measures, between apoE genotype and changes from baseline to 12 mo, and between menopausal status and changes from baseline to 12 mo. Three women who completed the study were excluded from all analyses because of regular use of oral contraceptives during the study (isoflavone $n = 1$, placebo $n = 1$) or being treated for alcoholism (isoflavone $n = 1$). Data from 3 and 4 women in the isoflavone and placebo groups, respectively, were excluded from analyses on serum lipid levels because they had taken medications such as statins or lipid-lowering drugs, and data from 9 women in each treatment group were excluded from analyses on blood pressure because they had taken antihypertensive medication. One woman in the placebo group was taking an antifibrinolytic agent, and her data were excluded from analyses of blood-clotting factors. LDL cholesterol data were unavailable for 1 woman in the isoflavone group and triglyceride data were unavailable for 1 woman in the isoflavone group.

RESULTS

Withdrawals, participant characteristics, and urinary isoflavone excretion. There were no differences between treatment groups in the number of withdrawals ($\chi^2 = 1.123$, $P = 0.29$) (**Fig. 1**). Women in the isoflavone and placebo groups were 55.1 ± 4.7 and 55.2 ± 4.9 y old ($P = 0.65$), respectively, and their BMI were 25.3 ± 3.9 and 25.3 ± 3.5 kg/m^2 ($P = 0.90$), respectively. BMI increased from baseline to 12 mo among women in both treatment groups (mean change 0.38 ± 1.09 and 0.51 ± 0.97 kg/m^2 among women in the isoflavone and placebo groups, respectively; $P = 0.42$). The number of women who were pre-, peri-, and postmenopausal were 13, 12, and 56, respectively, among women in the isoflavone group (data unavailable for 3 women), and 15, 14, and 61, respectively, among women in the placebo group ($\chi^2 = 0.04$, $P = 0.98$).

All 3 common alleles of apoE (E2, E3, and E4) were detected, and the most frequent genotype was E3/E3 ($n = 91$), followed by E3/E4 ($n = 43$), E2/E3 ($n = 25$), and E2/E4 ($n = 3$). There were no E2/E2 or E4/E4 genotypes within this study population. One-way ANOVA showed a significant effect of apoE genotype on baseline levels of total and LDL cholesterol ($P = 0.004$ and $P = 0.0001$, respectively); total cholesterol was significantly lower among women with the E2/E3 genotype (5.58 ± 0.83 mmol/L) than among women with the E3/E3 (6.28 ± 1.01 mmol/L) or E3/E4 (6.49 ± 1.33 mmol/L) genotypes. LDL cholesterol differed significantly among all 3 genotypes; E2/E3 = 3.29 ± 0.74 mmol/L, E3/E3 = 4.08 ± 0.96 mmol/L, and E3/E4 = 4.50 ± 0.91 mmol/L. There were no interactions between apoE genotype and baseline levels of fibrinogen or PAI-1 ($P = 0.17$, and $P = 0.27$, respectively; data not shown).

According to the PABA-check method, 58, 72, and 77% of the women had complete urine collections at baseline, 6, and 12 mo, respectively. A further 22, 11, and 13% had PABA recoveries between 70 and 85% at baseline, 6, and 12 mo, respectively. Total isoflavone excretion (sum of daidzein, genistein, formononetin, and biochanin A) among women in the isoflavone and placebo groups (excluding women with < 70% or > 110% PABA recovery), was 4344 and 1755 nmol/24 h at baseline ($P = 0.23$), 19464 and 1234 nmol/24 h at 6 mo ($P < 0.001$), and 19462 and 914 nmol/24 h at 12 mo ($P < 0.001$), respectively. Isoflavone excretion changed from baseline to 6 mo ($P < 0.001$) and from baseline to 12 mo ($P < 0.001$) among women in the isoflavone group, but did not change from baseline to 6 mo ($P = 0.80$) or from baseline to 12 mo ($P = 0.15$) among women in the placebo group. Inclusion of women with incomplete or unsatisfactory collections did not alter these findings. The increase in isoflavone excretion among women taking the isoflavone tablets and the lack of change among women taking the placebo indicate good compliance.

Serum lipids. There were no treatment effects on changes in total cholesterol, LDL cholesterol, triglycerides, or LDL cholesterol (**Table 1**). When data were analyzed as a percentage of baseline data, the 2 groups also did not differ. There was a significant interaction between treatment group and menopausal status for the change in triglycerides ($P = 0.02$), and the difference between treatment groups was significant among perimenopausal women (**Table 2**). Interactions between apoE and treatment group for changes in total and LDL cholesterol tended to be significant ($P = 0.06$ and $P = 0.05$, respectively). Differences between treatment groups were significant among women with the E2/E3 genotype (**Table 3**).

Blood clotting factors. There were no differences between treatment groups for changes in fibrinogen or PAI-1 among all women (Table 1). The interaction between treatment group and menopausal status was significant for the change in PAI-1 ($P = 0.01$), and the difference between treatment groups was significant among perimenopausal women (Table 2). There were no interactions between apoE and treatment group for the change in fibrinogen or PAI-1 ($P = 0.63$ and $P = 0.29$, respectively; data not shown).

Blood pressure. There were no treatment effects on systolic or diastolic blood pressures (Table 1), and no interactions between treatment group and menopausal status for these

TABLE 1

Total cholesterol, triglycerides, LDL cholesterol, HDL cholesterol, fibrinogen, PAI-1, and systolic and diastolic blood pressures at baseline and 12 mo in women taking isoflavone or placebo tablets[1]

	Isoflavone		Placebo		Significance of change[2]
	Baseline	12 mo	Baseline	12 mo	*P*-value
Total cholesterol, *mmol/L* (77, 86)[3]	6.34 ± 1.19	6.34 ± 1.14	6.08 ± 1.04	6.24 ± 1.07	0.29
Triglycerides, *mmol/L* (77, 86)	1.24 ± 0.71	1.29 ± 0.62	1.19 ± 0.66	1.22 ± 0.52	0.92
LDL cholesterol, *mmol/L* (75, 86)	4.21 ± 0.94	4.01 ± 1.01	3.88 ± 1.00	3.84 ± 1.02	0.13
HDL cholesterol, *mmol/L* (77, 86)	1.61 ± 0.41	1.81 ± 0.49	1.66 ± 0.48	1.84 ± 0.50	0.56
Fibrinogen, g/L (83, 84)	2.98 ± 0.78	2.96 ± 0.93	2.99 ± 0.79	3.11 ± 0.92	0.37
PAI-1, *IU/L* (83, 87)	16.32 ± 7.79	15.90 ± 8.05	15.89 ± 7.16	16.45 ± 8.56	0.33
Systolic, *mmHg* (74, 79)	128 ± 20	126 ± 17	125 ± 16	127 ± 19	0.10
Diastolic, *mmHg* (74, 79)	81 ± 11	79 ± 11	81 ± 9	78 ± 11	0.28

[1] Values are means ± SD.
[2] *P*-value for difference between treatment groups for changes from baseline to 12 mo.
[3] (number of women in the isoflavone group, number of women in the placebo group).

changes ($P = 0.94$ and $P = 0.46$ for systolic and diastolic blood pressures, respectively).

DISCUSSION

Overall, we did not see a significant effect of the dietary supplement, providing 26 mg biochanin A, 16 mg formononetin, 1 mg genistein, and 0.5 mg daidzein daily for 1 y, on the cardiovascular disease risk factors examined. When grouped by menopausal status, there was a potentially beneficial effect of the isoflavone supplement on the changes in triglycerides and PAI-1 among perimenopausal women, but the sample size was small and a larger study is required to confirm this finding.

In the meta-analysis by Anderson et al. (3), changes in total and LDL cholesterol were directly related to baseline levels. The greatest improvements were seen among those in the upper 2 quartiles of total cholesterol at baseline (>6.71 mmol/L), and the smallest improvements occurred in individuals whose baseline levels of total cholesterol were in the lower 2 quartiles of total cholesterol (<6.60 mmol/L). In comparison with those findings, the mean baseline total cholesterol among women in our study was 6.34 mmol/L in the isoflavone group and 6.08 mmol/L in the placebo group, which may account for the lack of significant effects. Some studies showed little or no effect of soy or isoflavone interventions in normocholesterolemic subjects (44,45); in those that did, the protective effects were relatively modest (46,47). As discussed in the introduction, the effect of isoflavones, either extracted or intact, on cholesterol and LDL cholesterol levels is unclear.

Elevated triglyceride level is a risk factor for coronary heart disease (48–50), and some studies showed increases in triglycerides with conventional hormone replacement therapies (51,52). In contrast, evidence exists that dietary soy protein reduces triglyceride levels (20,53,54). However, the evidence for isoflavone supplements is less convincing (16,17,55). In our study, we observed a potentially beneficial decrease in triglycerides among perimenopausal women taking the isoflavone supplement, but the sample size was small and a larger study is required to confirm this finding.

Although some hypotheses were advanced to explain the lipid-lowering effects of intact soy protein or isoflavones and other bioactive components of soy [reviewed in (56)], the full mechanism of action remains unknown. Our findings suggest that isoflavones alone are not responsible for the well-documented effects of soy protein on blood lipids.

In agreement with others (57–59), we found a significant effect of apoE genotype on baseline levels of total and LDL cholesterol. Some evidence suggests differential responses to interventions according to apoE genotype. Heikkinen et al. (60) reported that women without the apoE E4 allele responded more favorably to hormone replacement therapy than those with the E4 allele, in terms of lowering total and LDL cholesterol. In addition, Loktionov et al. (43) found that study subjects with the apoE E4 allele had the highest PAI-1 activity, and that the E2/E3 group had the lowest activity. We observed interactions that tended to be significant between apoE genotype and changes in total and LDL cholesterol; women with the E2/E3 genotype appeared to respond more favorably to the intervention than women with the E3/E3 or E3/E4 genotypes. This finding is in contrast to those of Gaddi et al. (61) who reported greater reductions in total cholesterol

TABLE 2

Triglyceride and PAI-1 levels in perimenopausal women taking the isoflavone or placebo tablets at baseline and 12 mo[1]

	Isoflavone		Placebo		Significance of change[2]
	Baseline	12 mo	Baseline	12 mo	*P*-value
Triglycerides, *mmol/L* (12, 14)[3]	1.37 ± 1.24	1.17 ± 0.69	1.14 ± 0.45	1.54 ± 0.69	0.02
PAI-1, *IU/L* (12, 12)	16.42 ± 8.76	13.37 ± 5.33	17.28 ± 8.81	22.24 ± 6.94	0.004

[1] Values are means ± SD.
[2] *P*-value for difference between treatment groups for changes from baseline to 12 mo.
[3] (number of women in the isoflavone group, number of women in the placebo group).

TABLE 3

Total and LDL cholesterol levels at baseline and 12 mo in women taking the isoflavone or placebo tablets and stratified by apoE genotype[1]

	Isoflavone		Placebo		Significance of change[2]
	Baseline	12 mo	Baseline	12 mo	*P*-value
Total cholesterol, *mmol/L*					
E2/E3 (8, 16)[3]	6.21 ± 0.79	5.60 ± 1.06	5.23 ± 0.67	5.41 ± 0.95	0.03
E3/E3 (51, 39)	6.36 ± 1.04	6.39 ± 1.11	6.18 ± 0.97	6.41 ± 0.98	0.21
E3/E4 (14, 27)	6.35 ± 1.88	6.64 ± 1.01	6.56 ± 1.00	6.54 ± 1.03	0.47
LDL cholesterol, *mmol/L*					
E2/E3 (8, 16)	3.86 ± 0.71	3.03 ± 1.11	2.98 ± 0.60	2.94 ± 0.76	0.02
E3/E3 (50, 39)	4.20 ± 0.96	4.11 ± 0.93	3.89 ± 0.95	3.99 ± 0.99	0.18
E3/E4 (13, 27)	4.54 ± 1.03	4.30 ± 0.89	4.47 ± 0.89	4.20 ± 0.95	0.88

[1] Values are means ± SD.
[2] *P*-value for difference between treatment groups for changes from baseline to 12 mo.
[3] (number of women in the isoflavone group, number of women in the placebo group).

in response to a soy protein intervention among women with the E3/E3 or E3/E4 genotypes. Another study showed no effect of apoE genotype on response to a soy intervention (62).

In contrast to some studies of soy protein consumption (19–22), we did not see a significant effect of the isoflavone supplement on blood pressure. An isoflavone tablet providing 80 mg isoflavones/d also had no effect on blood pressure in another study (15), but it was relatively short, lasting only 8 wk.

In conclusion, there were no significant effects of the isoflavone supplement, providing 26 mg biochanin A, 16 mg formononetin, 1 mg genistein, and 0.5 mg daidzein daily for 1 y, on the cardiovascular risk factors measured in this population of women as a whole. There were potentially beneficial effects of the isoflavone supplement on triglycerides and PAI-1 among the perimenopausal women, and on total and LDL cholesterol among women with the apoE E2/E3 genotype. However, when grouped by menopausal status or apoE genotype, sample sizes were small and a larger study is required to confirm these findings.

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