

Consumption of Soy Isoflavones Does Not Affect Plasma Total Homocysteine or Asymmetric Dimethylarginine Concentrations in Healthy Postmenopausal Women^{1,2}

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ABSTRACT Postmenopausal women are at increased risk for cardiovascular disease because many risk factors are aggravated by menopause. Phytoestrogens may modulate risk factors favorably, involving mechanisms similar to estrogen. The effect of phytoestrogens on the atherogenic amino acids homocysteine and asymmetric dimethylarginine (ADMA) was investigated in a controlled intervention study in healthy postmenopausal women. A multicenter, double-blind, crossover intervention trial in 89 postmenopausal women from Denmark, Germany, and the UK was performed. Subjects consumed fruit cereal bars with or without soy isoflavones (50 mg/d) for 8 wk each with an 8-wk washout period in between. Urinary phytoestrogens increased significantly after isoflavone intervention ($P < 0.001$). Isoflavone supplementation did not affect plasma total homocysteine or ADMA. For homocysteine, changes from baseline were $0.32 \mu\text{mol/L}$ (range: -0.31 – 0.92 ; 95% CI 0.13 – 0.72), and $0.29 \mu\text{mol/L}$ (range: -0.45 – 1.09 ; 95% CI 0.01 – 0.63 , $P = 0.286$) for isoflavone treatment and placebo, respectively. For ADMA concentrations, changes from baseline were $-0.02 \mu\text{mol/L}$ (range: -0.08 – 0.03 ; 95% CI -0.04 – 0.01), and $0.00 \mu\text{mol/L}$ (range: -0.05 – 0.03 ; 95% CI -0.03 – 0.01 , $P = 0.397$) for isoflavone treatment and placebo, respectively. There was no association between plasma total homocysteine and ADMA. Changes from baseline in plasma ADMA and folate were negatively correlated ($r = -0.18$, $P = 0.017$). These results challenge the overall health effect of isoflavone supplementation in healthy postmenopausal women. *J. Nutr.* 136: 100–105, 2006.

KEY WORDS: • total homocysteine • asymmetric dimethylarginine • nitric oxide metabolites • postmenopausal women • cardiovascular disease

Cardiovascular disease (CVD)⁴ is the leading cause of death among women in Westernized societies (1). Cardiovascular disease risk increases strongly after menopause due to diminished estrogen production (2). Several risk factors are influenced by menopause, including a decrease in serum HDL

cholesterol (3), changes in plasma clotting and fibrinolytic factors (4), and an increase in plasma total homocysteine (5). Homocysteine is a nonproteinic amino acid of methionine metabolism, and an elevated plasma concentration is considered to be a strong and independent risk factor for cardiovascular disease in epidemiologic studies (6,7).

Recently, studies revealed that elevated concentrations of asymmetric dimethylarginine (ADMA), another amino acid that occurs in proteins by post-translational modification of L-arginine (8), are associated with an increased cardiovascular disease risk (9). ADMA is a natural competitive inhibitor of endothelial nitric oxide synthase and is related to endothelial dysfunction (10). Interestingly, ADMA and homocysteine metabolism are related because the methyl groups of ADMA are derived from S-adenosylmethionine, thus rendering S-adenosylhomocysteine (11). Another common feature of

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⁴ Abbreviations used: ADMA, asymmetric dimethylarginine; BP, blood pressure; CVD, cardiovascular disease; HRT, hormone replacement therapy; NOx, nitric oxide metabolites; TAG, triglycerides; tHcy, total homocysteine.

both atherogenic amino acids is the potential influence of sex hormones on their plasma levels. Plasma levels of homocysteine are higher in postmenopausal than in premenopausal women (5). Furthermore, hormone replacement therapy (HRT) was effective in lowering plasma homocysteine levels (12,13).

Fewer studies of ADMA are available and data on the effect of menopause are sparse. Recently, it was demonstrated that ADMA was reduced by HRT with conjugated equine estrogen (14).

Isoflavones belong to the group of phytoestrogens with a structure similar to that of 17β -estradiol (15). Specific compounds are genistein and daidzein, which occur naturally in soy, sprouts, peas, and other vegetables (16). The daily isoflavone intake in Asian populations consuming a traditional soy-containing diet (17) is 15–50 mg, compared with <1 mg in Western populations (18). Isoflavones bind with a high affinity to both estrogen receptors α and β (19), and therefore exert hormonal effects depending on the endogenous estrogen level. Although isoflavones compete with estrogen for binding to the estrogen receptor in a high estrogen milieu (e.g., in premenopause) they may act as estrogen mimics at low hormone levels (e.g., in postmenopause). Like estrogen, isoflavones were shown to inhibit the replication and migration of smooth muscle cells in vitro (20) and postinjury neointima development in animals (21). Moreover, in monkeys, estrogen and high soy protein diets reduced atherosclerotic lesions and cholesteryl ester content (22). Concentration-dependent potent vasodilatory properties and improved acetylcholine-mediated vasorelaxation were demonstrated with estrogen (23) and isoflavones (24).

Data on isoflavone consumption and levels of either homocysteine or ADMA are lacking. Recently, a high consumption of soy foods was inversely associated with plasma homocysteine concentrations (25) in an observational study. There is a gap in the knowledge concerning controlled intervention studies with isoflavones and their effect on homocysteine and ADMA. Therefore, the present study was performed to elucidate the effect of dietary phytoestrogens on homocysteine and ADMA plasma levels in healthy postmenopausal women.

SUBJECTS AND METHODS

Study design. This multicenter intervention study was conducted in study centers located in Frederiksberg (Denmark), Potsdam (Germany), and Reading (UK). Healthy postmenopausal women were recruited at each of the centers based on drawings from the central national registers and by advertisement in the local media. Volunteers were 45–70 y old and postmenopausal (defined as no menstrual period for >12 mo and confirmed by follicle stimulating hormone and luteinizing hormone if <36 mo had elapsed since the last menstrual period). Before the study, none of the volunteers had used HRT for 6 mo, or supplements containing fatty acids, isoflavones, vitamins, or minerals for 2 mo, or antibiotics for 3 mo. Volunteers had no history of diabetes, inflammatory diseases, or CVD and did not use antihypertensive, anti-inflammatory, or lipid-lowering drugs on a regular basis. Women who received such drugs during the study were excluded. Renal and liver functions were normal at entry. All volunteers were nonsmokers, had a blood pressure <160/90 mm Hg and a BMI of 20–32 kg/m². A screening blood sample was taken before entry and all subjects had a plasma total cholesterol <8 mmol/L, triglycerides (TAG) 1–3 mmol/L, HDL cholesterol <1.9 mmol/L, hemoglobin >7.4 mmol/L, and fasting glucose <6.5 mmol/L. Of 177 screened volunteers, 99 were included in the intervention study; 10 women withdrew from the study due to health problems, HRT usage or personal reasons; 89 women (88%) completed the intervention study according to the study protocol, with 27 participants from the UK, 34 women from Germany, and 28 study patients from Denmark. Participants were stratified according to BMI, age, and TAG before study entry and randomly assigned to 2 groups; cohort A started with placebo and cohort B with the isoflavone-enhanced fruit cereal bars. Randomization groups (cohort A and B) did not differ in baseline characteristics as demonstrated in **Table 1**. The study protocol complied with the Helsinki Declaration as revised in 1983 and was approved by local Ethics Committees of the countries (Denmark, Research Ethical Committee of Copenhagen and Frederiksberg; UK, University of Reading Ethics Committee; Germany, Ethics Committee of the University of Potsdam). All volunteers signed written informed consent before the start of the study.

The study was performed in a double-blind, placebo-controlled, randomized, crossover manner. Volunteers were obliged to consume 2 fruit cereal bars each enriched with 25 mg isoflavones, with a genistein:daidzein ratio of 2:1 or containing no isoflavones in the placebo arm. The daily dose of 50 mg is in line with the average intake of phytoestrogens in countries with traditional soy intake (17).

TABLE 1

Baseline characteristics of the study population comprised of healthy postmenopausal women¹

	Cohort A ²	Cohort B ²	Total
<i>n</i>	46	43	89
Age, y	59 ± 5	58 ± 6	59 ± 5
BMI, kg/m ²	24.4 ± 3.1	24.4 ± 2.8	24.4 ± 3.0
Systolic blood pressure, mm Hg	124 ± 13	119 ± 21	121 ± 17
Diastolic blood pressure, mm Hg	76 ± 8	73 ± 10	75 ± 9
Fasting plasma glucose, mmol/L	5.2 ± 0.5	5.1 ± 0.5	5.1 ± 0.5
Plasma total cholesterol, mmol/L	6.2 ± 1.2	6.2 ± 1.3	6.2 ± 1.3
HDL cholesterol, mmol/L	1.6 ± 0.4	1.6 ± 0.4	1.6 ± 0.4
LDL cholesterol, mmol/L	3.9 ± 1.1	4.1 ± 1.1	4.0 ± 1.1
Plasma TAG, mmol/L	1.3 ± 0.5	1.2 ± 0.5	1.2 ± 0.5
Plasma tHcy, μmol/L	9.9 ± 1.9	10.6 ± 2.8	10.2 ± 2.4
Plasma folate, nmol/L	22.0 ± 9.7	22.4 ± 12.2	22.2 ± 10.9
Plasma vitamin B-12, pmol/L	434 ± 112	370 ± 142	403 ± 131
Plasma ADMA, μmol/L	0.74 ± 0.22	0.68 ± 0.10	0.71 ± 0.18
Plasma NOx, μmol/L	27.5 ± 11.6	30.9 ± 15.5	29.2 ± 13.6

¹ Values are mean ± SD.

² A and B indicate the treatment order; group A started with placebo and group B with the isoflavone-enhanced fruit cereal bar.

Furthermore, this dosage is effective in exerting hormonal effects in premenopausal women (26,27). Study duration for each intervention was 8 wk with an 8-wk washout period in between. Volunteers were instructed to consume 1 fruit cereal bar during the morning and 1 in the afternoon and to keep daily records of cereal bar consumption and well-being in a study diary. In addition, volunteers were advised to replace snacks with the fruit cereal bars. Each cereal bar (40 g) had a mean nutrient content of energy 652 kJ; protein 2.6 g; carbohydrate 17.3 g; fat 8.5 g; fiber 1.8 g; sodium 0.012 g. The cereal bars were produced in 4 different flavors (apricot and almond, apple and cardamon, hazelnut, or raspberry) (Health & Diet Food). The added isoflavones used are commercially available as "Solgen 40" (Solbar Plant Extracts). The product was tested before packaging and during the study by HPLC (28) as described previously (29) to ensure stability of the isoflavones.

Habitual diet was assessed by a 3-d dietary record at baseline (wk 0) and in the middle of each intervention period (wk 4). Dietary intake was calculated at each study center using local food databases.

Compliance was assessed by measurement of phytoestrogen concentrations in 24-h urine samples (30) and by a record of cereal bar consumption kept by volunteers. Urine was collected for 24 h at the beginning and end of each intervention period. Participants visited the study centers at baseline and at wk 4 and 8 during each intervention period. Each visit involved anthropometric (weight, height, waist:hip ratio) and blood pressure (BP) measurements. Three brachial arterial BP readings were recorded on an automatic BP device (Boso medicus memory, BOSCH + SOHN), with subjects in a sitting position after 10-min of rest, and the readings averaged.

Biochemical assays. Urinary phytoestrogens (genistein, daidzein, equol) were analyzed by a new validated monoclonal antibody based DELFIA (Dissociation Enhanced Lanthanide Fluorescent Immuno-Assay) immunoassay using AutoDelia 1235 automatic immunoassay system (Perkin Elmer) (31). The sensitivity of the time-resolved fluorescence immunoassays was determined to be 3.9 nmol/L (daidzein), 88.8 nmol/L (urinary genistein), 8.7 nmol/L (serum genistein), and 2.2 nmol/L (equol). The intra-assay CV for daidzein concentrations of 157.2 nmol/L was 3% and the inter-assay CV was 0.6%. The intra-assay CV for urinary genistein concentrations of 3700 nmol/L was 3.9% and the inter-assay CV was 2.2%. The intra-assay CV for serum genistein concentrations of 462.5 nmol/L was 4% and the inter-assay CV 0.8%. The intra-assay CV for equol concentrations of 97.5 nmol/L was 2.9% and the inter-assay CV was 1.9%.

Equol is formed by the action of duodenal and bacterial β -glucosidases from daidzein (32). Based on the discovery of equol in amounts >936 nmol/L urine, participants were classified as equol producers or nonproducers (33). Samples for each subject were processed in one batch.

Blood samples were collected from fasting subjects at each visit. Plasma samples for total homocysteine (tHcy), ADMA, nitric oxide metabolites (NOx), and vitamin analysis were centrifuged at 1600 \times g and 4°C for 10 min. Plasma was stored at -80°C and thawed immediately before analysis. The concentration of NOx, the sum of plasma nitrite and reduced nitrate, was measured by a reduced NADPH-dependent nitrate reductase assay (34). The inter-assay CV was <7%. Plasma tHcy, folate, and cobalamin were measured by automated methods on a random access analyzer (AxSYM tHcy, Abbott Laboratories; Elecsys Folate and Elecsys vitamin B-12, Roche Diagnostics). The interassay CV for the assays used was <5%. The concentration of ADMA in plasma was determined using a commercial ADMA-ELISA Kit (DLA Diagnostika). The recovery rate of ADMA was 89–105%, the intra-assay CV was 6% and the inter-assay CV 9.5%. Samples for each subject were processed in 1 batch.

Statistical analysis. SAS 8.4 (SAS Institute) was used for all statistical analyses (PROC MIXED procedure). All data are presented as median and percentiles (25th–75th), except in Table 1 where means and SD are given. Changes from baseline, i.e., wk 8 – wk 0, were used as dependent variables. If original data were approximately normally distributed, then changes from baseline on the original scale were calculated. If necessary, changes from baseline on a log scale were calculated. Subjects were included as a random factor within a linear mixed model. Fixed effects always included in the final model were: baseline values, treatment, center, time, and treatment order. Fixed

effects included in the final model if the effect was significant were: treatment order \times treatment interaction, baseline \times treatment interaction, and center \times treatment interaction. The difference between dietary intakes at mid-isoflavone (wk 4) and mid-placebo (wk 4) of the intervention arms was calculated and compared using the Wilcoxon test. Pearson's coefficient of correlation was calculated for baseline values and for changes from baseline. Cohorts with different treatment order were compared with respect to their baseline characteristics using Student's *t* test. A two-tailed $P < 0.05$ was accepted as the level of statistical significance.

RESULTS

The 2 cohorts of the crossover study did not differ in BMI, age, blood pressure measurements, plasma lipids, and the outcome variables (plasma tHcy, ADMA, NOx, vitamins) at start of the first intervention period (Table 1).

The isoflavone supplements were well tolerated; 49% of all participants reported increased gastrointestinal discomfort both during placebo and isoflavone treatment, probably due to the high fiber content of the fruit cereal bars. Isoflavone supplementation led to a significant increase in the urinary excretion of genistein, daidzein, and equol, as expected. Equol (>936 nmol/L) was detected in appreciable amounts in 33% of all participants with a higher level of equol producers in the German study population compared with the other centers (Germany 56.7%, Denmark 20.0%, UK 23.3%, $P = 0.036$).

Energy intake and anthropometrical measures. Habitual diet as assessed by 3-d dietary records did not differ between the placebo and isoflavone periods (Table 2). There were no relevant changes in BMI or the intakes of energy, fat, carbohydrates, and dietary fiber during the entire 24-wk study. The waist:hip ratio also was unchanged throughout the study period.

Plasma ADMA, total homocysteine, vitamins, NOx. Plasma ADMA, tHcy, plasma folate, and vitamin B-12 were not affected by placebo or isoflavone supplementation (Table 3). Plasma NOx decreased during the placebo period from 24.0 μ mol/L (range: 19.7–32.4) to 20.0 μ mol/L (range: 16.2–26.9) ($P < 0.001$) and did not change during the isoflavone supplementation period [before 23.6 μ mol/L (range: 17.6–33.6); after: 23.8 μ mol/L (range 18.0–33.7) $P = 0.813$], resulting in a significant difference in concentration after the treatment periods ($P < 0.001$). The

TABLE 2

Calculated daily nutrient intakes of postmenopausal women at wk 4 of placebo and isoflavone periods^{1,2}

	Isoflavones	Placebo
Energy, kJ/d	8635 (7389–9577)	8800 (7675–9699)
Protein, g/d	75 (61–85)	73 (66–83)
Fat, g/d	73 (63–89)	77 (64–91)
SFA, g/d	24 (18–35)	26 (20–32)
PUFA, g/d	11 (10–15)	13 (10–15)
MUFA, g/d	25 (20–32)	26 (20–33)
Carbohydrate, g/d	240 (215–280)	240 (210–218)
Total dietary fiber, g/d	25 (19–30)	25 (19–30)
Dietary cholesterol, mg/d	244 (163–340)	258 (190–338)
Folate, μ g/d	269 (230–320)	276 (231–329)
Vitamin B-12, μ g/d	4 (3–5)	4 (3–6)

¹ Values are medians (25th–75th percentiles), $n = 89$. The medians did not differ.

² Abbreviations: SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids.

TABLE 3

Changes (wk 8 – wk 0) in urinary isoflavones, and plasma concentrations of homocysteine, nitric oxide metabolites, asymmetric dimethylarginine, and vitamins in postmenopausal women during the placebo and isoflavone treatment periods¹

	Isoflavone	Placebo	P-value ²
Urine ³			
Genistein, nmol/L	11228 (8155–15762)	–63 (–258–365)	<0.001
Daidzein, nmol/L	9142 (5889–12900)	–0.8 (–196.1–130.5)	<0.001
Equol, nmol/L	201 (90–2253)	1.7 (–43.4–45.6)	<0.001
Plasma			
tHcy, μ mol/L	0.32 (–0.31–0.92)	0.29 (–0.45–1.09)	0.286
Folate, nmol/L	–0.15 (–1.86–1.13)	–0.32 (–1.57–1.45)	0.934
Vitamin B-12, pmol/L	–23.9 (–59.1–21.9)	–10.5 (–56.1–32.45)	0.465
ADMA, μ mol/L	–0.02 (–0.08–0.03)	0.00 (–0.05–0.03)	0.397
NOx, μ mol/L	1.00 (–6.65–7.85)	–2.60 (–8.75–2.25)	0.001

¹ Values are medians (25th–75th percentiles), $n = 89$.

² P-values are shown for the treatment effect within a stepwise-generated general linear mixed model implemented in SAS PROC Mixed. Changes from baseline were used as the response variable.

³ Isoflavones measured in 24-h urine samples.

intervention groups did not differ in baseline plasma TAG, total cholesterol, LDL cholesterol, or HDL cholesterol concentrations. The baseline plasma lipid profile did not influence plasma tHcy, ADMA, NOx, folate, or vitamin B-12.

Correlations between ADMA, total homocysteine, vitamins, and NOx. At baseline, correlations between the variables of interest were generally weak but significant. Plasma tHcy was inversely correlated with plasma vitamin B-12 ($r = -0.25$, $P = 0.001$) and plasma folate ($r = -0.15$, $P = 0.050$). Interestingly, there was a positive association between plasma ADMA and plasma vitamin B-12 ($r = 0.17$, $P = 0.022$) whereas a negative correlation was found for plasma ADMA and folate ($r = -0.18$, $P = 0.021$). A significant negative association for changes from baseline was observed between plasma ADMA and plasma folate ($r = -0.18$, $P = 0.017$). Neither plasma ADMA nor tHcy was correlated with plasma lipids, blood pressure, BMI, the waist:hip ratio, or plasma NOx (data not shown).

Influence of equol status. Equol status did not affect any of the measured variables. Equol producers and nonproducers did not differ in plasma ADMA and tHcy at baseline or in their changes during isoflavone supplementation or placebo periods.

DISCUSSION

This double-blind, placebo-controlled, randomized, cross-over intervention study with 50 mg dietary isoflavones in healthy postmenopausal women did not show an effect on the atherogenic amino acids homocysteine (tHcy) and asymmetric dimethylarginine (ADMA) in plasma. This is an unexpected finding because studies with estrogen in postmenopausal women showed a decrease in plasma ADMA (14,35,36) and also in plasma total tHcy (12,13). Because most of the health benefits of isoflavones are thought to be mediated in part by an estrogenic mechanism, a decrease of plasma ADMA and homocysteine might have been expected in the present study.

Studies investigating the effect of isoflavones on plasma tHcy used isolated soy protein as a vehicle rather than isolated isoflavones. Usually, the soy treatment was compared with casein-supplemented diets. Studies addressing the effect of isolated isoflavones on homocysteine levels are lacking.

Hyperlipidemic men and postmenopausal women investigated by Jenkins et al. (37) received soy protein containing either 10 or 73 mg isoflavones/d for 1 mo. A small but significant decrease in homocysteine was observed with intake of the low-isoflavone diet. Tonstad et al. (38) studied the effect of 30 or 50 g of isolated soy protein containing 111 or 185 mg isoflavones for 16 wk in healthy men and postmenopausal women compared with casein-supplemented diets. Compared with the casein treatment, soy protein decreased plasma tHcy significantly by $-0.8 \mu\text{mol/L}$. In patients with type 2 diabetes mellitus, isolated soy protein providing 165 mg isoflavones/d reduced the plasma tHcy concentration by 14% in contrast to casein (39).

The present study did not use different protein sources, but isoflavone compounds extracted from soy protein, which were given in comparison with placebo. Thus, the present study cannot be compared directly with the other studies. Casein, in contrast to soy protein, is an animal protein with a higher methionine content (40). Studies showed that methionine loading leads to a rapid increase in the plasma homocysteine level in humans (41). Therefore, the effect seen in the other studies was most likely due to the protein source rather than to the isoflavone content.

The present study is the first to address the effect of isoflavones on plasma ADMA concentrations. The decrease in ADMA observed with estrogens is likely due to activation of the ADMA hydrolyzing enzyme, dimethylarginine dimethylaminohydrolase (35); however, a similar effect was not observed in the in vivo situation with healthy postmenopausal women. Two important aspects must be considered. First, the plasma ADMA concentration in our population was well within the reference range compared with other healthy postmenopausal women (36), and greater reductions could have been expected in patients with initially higher plasma ADMA levels. Second, there are no data assessing the minimal length of investigation required for a reduction of plasma ADMA levels. In addition, there are no observational studies comparing ADMA concentrations in populations with a high habitual isoflavone intake and a Western diet. Therefore, no data are available concerning whether high isoflavone or soy protein consumption affect plasma ADMA concentrations in the general population.

Isoflavones may not have affected plasma tHcy and ADMA in this study because the postmenopausal women in the present

group were neither hyperhomocysteinemic nor did they exhibit other major cardiovascular risk factors such as insulin resistance or hyperlipidemia. The importance of the initial risk status of the study population is emphasized by other investigators who found that the tHcy-lowering effect of estrogen or HRT is more pronounced the higher the basal plasma tHcy concentration. A study population comprising postmenopausal women at a higher risk for cardiovascular disease may therefore have had a greater response to cardiovascular risk intervention.

The difference in plasma NO_x concentrations between the treatments is in accordance with reported changes in plasma concentrations of NO_x after pure genistein intake (54 mg/d) (42,43) although this is not consistent in all studies available (44). The hypothesis that isoflavones improve NO bioavailability is also supported by *in vitro* studies (45). The underlying mechanisms are not fully clear at present but it is thought that the antioxidant properties may play an important role in prevention of superoxide-mediated degradation of NO (45). In the endothelial cell, NO is continuously synthesized by constitutively expressed endothelial nitric oxide synthase from the substrate L-arginine. NO is the major vasodilator in the vascular system and exhibits major antiatherosclerotic properties. Thus, reduction of employable NO is considered a driving factor for endothelial dysfunction and a prerequisite for the development of atherosclerosis. In accordance, clinical studies revealed an inverse association between urinary NO_x and the degree of vascular impairment in patients with peripheral arterial disease (9). However, *in vivo* measurement of NO production is difficult due to rapid metabolism. Therefore, NO_x have been accepted as an indirect measure of NO availability even though it is influenced by internal and external factors such as diet and bacterial and enzymatic formation (46). Recently, it was shown that NO_x did not change during activation of the NO pathway, suggesting that our results may reflect solely a reduced degradation by isoflavones rather than a modulation of endothelial nitric oxide synthase activity (47). Additionally, it can be concluded that the differences observed for NO_x are independent of ADMA.

Plasma inflammatory markers and plasma endothelin-1, the major endothelium-derived constricting factor, were also not affected by isoflavone intervention in the same study population (29). These findings rule out a possible link to the results obtained for tHcy and ADMA as well as for changes observed in NO_x.

In conclusion, the findings of the current study challenge the overall health effect of isoflavone supplementation in healthy Western populations. This is in agreement with other findings that could not verify the proposed beneficial effects of isolated soy isoflavones on blood lipids (48), on inflammatory markers (29), and menopausal hot flashes (49). In addition, results from intervention studies with isoflavones on osteoporosis risk (50) and hormone-related cancers (51) remain controversial. Based on the present results, a general supplementation of isolated isoflavones in healthy Western postmenopausal women cannot be recommended.

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