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Efficacy and safety of a soy isoflavone extract in postmenopausal women: A randomized, double-blind, and placebo-controlled study

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Abstract

Objective: To investigate the efficacy of soy isoflavone on climacteric symptoms in postmenopausal women.
Design: In this double-blind, randomized, placebo-controlled study, a total of 80 women (mean age = 55.1 years), who reported 5 or more hot flush episodes per day, were randomized to receive either 250 mg of standardized soy extract (Glycine max AT) a total of 100 mg/day of isoflavone ($n = 40$) or placebo ($n = 40$). Exclusion criteria included: contra-indication for hormone therapy (HT), chronic gastrointestinal diseases, and users of HT within the preceding 6-months. For 10-months, climacteric symptoms were evaluated using a score card and the menopausal Kupperman index. Compliance and safety were also assessed. At baseline and the end of the study, lipid and hormonal profiles, as well as vaginal, mammographic and ultrasonographic parameters were measured. The *t*-test, Wilcoxon test and ANOVA were used in the statistical analysis.
Results: At baseline, the mean number of hot flushes was 9.6 ± 3.9 per day in the isoflavone group and 10.1 ± 4.9 in the placebo group ($p > 0.05$). After 10 months, there was a significant reduction in frequency of hot flushes among isoflavone users when compared to those on placebo (3.1 ± 2.3 and 5.9 ± 4.3, respectively) ($p < 0.001$). Kupperman index mean values showed a significant reduction in both groups. However, soy isoflavone was significantly superior to placebo, in reducing hot flush severity (69.9% and 33.7%, respectively) ($p < 0.001$). Endometrial thickness, mammography, vaginal cytology, lipids and hormonal profile did not change in both groups. No serious adverse event related to isoflavone treatment was reported.
Conclusions: The soy isoflavone extract exerted favorable effects on vasomotor symptoms and good compliance, providing a safe and effective alternative therapeutic for postmenopausal women.

Keywords: Hot flushes; Menopause; Isoflavones; Soy; Glycine max AT

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1. Introduction

Hormone therapy (HT) is recommended for the relief of vasomotor symptoms, treatment of vaginal atrophy and prevention of osteoporosis [1]. In spite of these well-known benefits, approximately 70% of the women discontinue HT after the first year of treatment [2]. One of the main causes of this low compliance rate is irregular bleeding. Other reasons include mastalgia, nausea, migraine, weight gain, and hydric retention, besides fear of breast cancer. These effects have led some women to be reserved and opt for natural therapies. Moreover, there are women in whom HT is contraindicated. In these cases, very little can be offered to relieve their climacteric symptoms [3,4]. For the relief of mild vasomotor symptoms, the North American Menopause Society recommends lifestyle changes, either alone or combined with a non-prescription remedy, such as dietary isoflavones, black cohosh or vitamin E. Prescription systemic estrogen-containing products remain the therapeutic standard for moderate to severe menopause-related symptoms in patients without contra indications to HT [5].

Soy foods and soy isoflavones have been adopted by some women as a natural alternative to hormone therapies because the soybean contains nutritionally relevant amounts of isoflavones [6]. Isoflavones belong to a class of compounds called phytoestrogens. The primary isoflavones found in soy are genistein, daidzein and glycitein. These are nonsteroidal compounds, structurally similar to estrogen, that weakly bind to estrogenic receptors (<1% of estradiol binding affinity) [7]. Soy isoflavones preferentially bind to β-estrogen receptors that are found in the central nervous system, bones, vascular walls and the urogenital tract. Unlike estrogens, isoflavones have little affinity with α receptors of breast and uterine tissues [8]. Depending on the concentrations of estradiol, they exert a selective action, i.e., in some tissues they display proestrogenic responses, whereas in others they inhibit estrogenic action [9,10].

Isoflavones have been demonstrated to reduce both the severity and the frequency of menopause-related vasomotor symptoms [5]. Most clinical evidences on the use of isoflavones are epidemiologic and were obtained in areas where soy consumption is high [10]. The weak estrogen-like effects of isoflavones have been proposed as a possible explanation for the low incidence of hot flushes experienced by women in Japan. The association between soy byproducts intake and hot flushes was examined in a cohort of 1106 Japanese premenopausal women, aged 35–54 years, who were followed up for 6 years. After data were controlled for age, total energy intake, and menopausal status, hot flushes were inversely associated with consumption of products containing isoflavones and the occurrence of climacteric symptoms [11].

Soy isoflavones have been investigated, mainly over the past 10 years, because of their potential effects on the health of postmenopausal women. Even though some clinical studies have demonstrated the efficacy of isoflavones in reducing the frequency and severity of hot flushes [3,12–17], others have not found differences between treated and non-treated groups [4,18–20]. These conflicting results may be attributed to the great chemical heterogeneity of the soy products used, exposure length and study design. According to the National Institute of Health State-of-Science Conference Panel on the management of menopausal-related symptoms, trials of soy supplementation are mixed and, because most of these products are not manufactured in a standardized way, they may differ in composition from trial to trial [21].

The aim of this study was to investigate the efficacy of a standardized soy isoflavone extract on climacteric symptoms in postmenopausal women. Lipid and hormonal profiles, as well as vaginal, mammographic and endometrial parameters were evaluated.

2. Methods

2.1. Study design and participants

This clinical randomized, double-blind, placebo-controlled trial included 80 Brazilian women from the Climacterium Outpatient Service of the Department of Gynecology of UNESP-Sao Paulo State University. All subjects included were postmenopausal women aged 45 years or older with good overall health, spontaneous amenorrhea for at least 12 months, follicle-stimulating hormone level greater than 40 mIU/ml, and average of five or more vasomotor symptoms per day. Exclusion criteria included strict vegetarian, high-fiber or high-soy diet, and history of breast cancer, endometrial carcinoma, cardiovascular disease, thromboembolic

disorders, undiagnosed vaginal bleeding, chronic alcoholism, and chronic gastrointestinal diseases. No subject was on hormone therapy or phytoestrogens within the preceding 6 months. Prior to the study, thyroid stimulator hormone (TSH) and free tyroxine (T_4) levels were measured to exclude thyroid dysfunctions that could interfere with the symptoms. Informed consent was obtained from all patients, and the study was approved by the Research Ethics Committee of Botucatu Medical School - UNESP.

The initial evaluation consisted of case history taking, general and gynecological physical examination, oncotic colpocytology, mammography and transvaginal ultrasonography. Data collected included information on age, menarche, time since menopause, parity, HT contraindication, weight, height, and waist circumference. Following a pre-study period, participants were randomly assigned to one of two groups: SI, receiving soy isoflavone extract ($n = 40$) or PL, receiving placebo ($n = 40$). Examiners and subjects had no previous knowledge of group assignment. Placebo and active treatment were identical in appearance. Centralized computerized subject randomization process was conducted using specific software by statistician unaware of the study protocol. The packing of capsules were labeled with cod numbers. The only unblended person was statistician responsible.

Thus, 40 participants were given 250 mg of standardized soy extract (Glycine max AT), corresponding to 100 mg/day of isoflavone, administered twice a day in capsules containing 125 mg of soy extract plus 50 mg of isoflavones each. The standardized extract contained approximately 50% of genistein and 35% of daidzein. The other 40 participants received two lactose capsules/day. All the capsules were identical in appearance. Subjects were instructed to return any unused medication at visits for compliance determination. Follow-up length was 10 months, with evaluations at pre, baseline, 4, 7 and 10 months.

2.2. Study protocol

Vasomotor symptoms were evaluated using a score card and the Kupperman Menopausal Index. All participants were instructed to record the daily number of hot flushes in a diary card and bring it to each visit (pre, baseline, 4, 7 and 10 months). The women experiencing at least five hot flushes per day were included in the study. At the baseline interview and at each follow-up session, Kupperman Menopausal index was obtained. This index is a numerical conversion system that grades 11 of the most common menopausal complaints, namely hot flushes, paresthesia, insomnia, nervousness, melancholia, vertigo, weakness, arthralgia or myalgia, headache, palpitations and stinging. Each symptom was rated on a scale from 0 to 3 for absent, mild, moderate, and severe complaints. To calculate the Kupperman index, symptoms were weighed as follows: hot flush severity (4×), paresthesia (2×), insomnia (2×), nervousness (2×), and all other symptoms (1×). Hot flush severity was separately evaluated, and classified as mild (4), moderate (8), and severe (12), according to self-reported rating.

Weight variation was assessed by the Body Mass Index (BMI = weight/height^2). Height and weight were obtained using a stadiometer and a standard balance beam scale, respectively, with subjects wearing lightweight clothing and no shoes. BMI was classified according to the system used by the World Health Organization (2002): $<18.5\ kg/m^2$ = underweight; 18.5–24.9 kg/m^2 = normal weight; 25–29.9 kg/m^2 = overweight; 30.0–34.9 kg/m^2 = obese class I; 35.0–39.9 kg/m^2 = obese class II; $\geq 40.0\ kg/m^2$ = obese class III. Abdominal fat was indirectly assessed by measuring waist circumference (WC) and was considered high when WC >88 cm.

Vaginal cytology was performed on all participants at baseline and after 10 months. Vaginal smears were collected with a vaginal spatula from the right upper third of the vaginal wall. In a total of 300 exfoliation cells, parabasal cells (P), intermediary cells (I), and superficial cells (S) were manually assessed in Papanicolao stained smears, and results were expressed as Maturation Value (MV). All examinations were interpreted by the same experienced cytopathologist without prior knowledge of the subjects' data. Vaginal pH was measured during the gynecological examination. The pH paper (Merck 0–14, Darmstadt, Germany) was left in direct contact with one-third of external left vaginal wall for 1 min.

Transvaginal ultrasonography was performed to evaluate the endometrial cavity at baseline and at 10 months by the same examiner, using a Toshiba Power Vision 6000 (Japan) with a 7.5 MHz endovaginal transducer. Endometrial thickness, measured between basal layers at sagital view, was considered normal when less

than 5 mm. Mammography was performed using standard techniques with craniocaudal and oblique views, at baseline and after 10 months of treatment.

2.3. Laboratory assessment

Blood samples were collected from each subject after 12-h fasting at pre, 4 and 10 months. A 12-ml blood sample was collected via venipuncture from an antecubital vein. The samples were allowed to clot at room temperature for 10 min, and then centrifuged for 15 min at 0 °C. The serum was then pipetted into polypropylene tubes. Aliquots were frozen at −70 °C for subsequent analysis. Triglycerides (TG), total cholesterol (TC), HDL, and glycemia were measured by the automatic biochemical analyzer RAXT (Technicon, USA). LDL was calculated using the formula of Friedewald et al., where total cholesterol is subtracted from the sum of HDL and triglyceride divided by five. Normality rates were: TC < 200 mg/dl, HDL > 40 mg/dl, LDL < 100 mg/dl, and TG < 150 mg/dl. Follicle-stimulating hormone (FSH), luteinizing hormone (LH) and estradiol levels were assessed at baseline and at 9 months by Immulite System (DPC, USA) automated immunoassay. FSH between 25.0 and 134.8 mUI/ml; LH, 11.3 and 40.0 mIU/ml; estradiol, <30.0 pg/ml were considered as normal menopause values.

At the end of study, the plasma levels of isoflavones were measured to evaluate their bioavailability after the oral administration of the standardized soy extract. The plasma concentrations of genistein and daidzein, the phytoestrogens mostly present in soy, were determined in every participant's sample using reversed-phase high-performance liquid chromatographic (HPLC) assay and ultraviolet scanning (Shimadzu, Japan). Blood samples (0.5 ml) were collected in polypropylene tubes containing 50 μl of heparin (50.000 IU). After centrifugation at 3000 × *g* at 4 °C for 10 min, each sample was stored at −70 °C until analysis. This method for analyzing isoflavone plasma concentrations has been previously described and has a high sensitivity and reproducibility [22]. Plasma genistein and daidzein concentrations were expressed in μmol/dl. The detection limit of the isoflavone assay was <2 μmol/dl.

Safety and tolerability were assessed by means of a symptom questionnaire, adverse events reports, and physical examination during clinical visits every 3 months. Moreover, laboratory analyses, Pap smear, ultrasonographic endometrial thickness and mammography were evaluated.

2.4. Statistical analysis

Statistical analyses were performed using the Statistical Package for the Social Sciences Software (Version 12.0) (SPSS Inc. Chicago, IL, USA). Normally distributed variables were reported as mean ± standard deviations. Group interaction was assessed by an independent *t*-test. Differences between baseline and post-treatment values were analyzed by the paired *t*-test. Timing effect was assessed by one-way repeated measures analysis of variance (ANOVA). When differences were detected, a Tukey's post hoc test was performed to determine pair wise differences. For the variables that showed abnormal distribution, the non-parametric Wilcoxon test was used and the results were expressed as median with 25th and 75th percentiles. Exact *p* values were obtained from the tests employed. Statistical tests were two-tailed and significance was set at 5%.

3. Results

A total of 76 subjects completed the 10-month follow-up, 38 from the isoflavone group and 38 from the placebo group. Their mean age was 55.7 years. In the isoflavone group, two subjects withdrew from the study after 4.5 and 5.5 months of follow-up due to flatulence and epigastralgia, which ameliorated after the medication was discontinued. In the placebo group, two patients withdrew from the study: one after 2 months because of depression and family problems, and the other after 3.5 months for not wishing to take the study medication. Only those subjects who completed 9 months of treatment were included in the efficacy analysis.

Baseline clinical characteristics and laboratorial parameters in the groups on soy isoflavone extract (SI) and on placebo (PL) were statistically compared and are shown in Table 1. There were no statistically significant differences ($p > 0.05$) in all baseline parameters between groups. Participants showing increased body fat percentage with android distribution (WC > 88 cm) were classified as overweight (Table 1). Table 2 shows

Table 1
Baseline clinical characteristics and laboratorial assessment of all subjects

Characteristic	Soy isoflavone ($n=38$)	Placebo ($n=38$)	p-Value*
Age (years)	55.1 ± 6.0	56.2 ± 7.7	0.461
Menopause (years)	48.4 ± 3.7	47.7 ± 3.5	0.258
Time since menopause (years)	6.6 ± 4.8	7.1 ± 4.2	0.356
Parity (children no)	2.9 ± 1.7	2.4 ± 1.8	0.167
Number hot flushes/day	9.6 ± 3.9	10.1 ± 4.9	0.564
Hot flushes severity score/day	9.3 ± 2.5	8.0 ± 2.6	0.189
KMI	23.3 ± 7.0	22.4 ± 7.4	0.580
BMI (kg/m^2)	29.7 ± 5.0	28.5 ± 4.9	0.251
WC (cm)	93.9 ± 12.1	92.0 ± 10.8	0.462
FSH (mUI/ml)	68.1 ± 24.5	66.9 ± 22.9	0.811
TSH (μIU/ml)	2.4 ± 1.3	2.1 ± 1.1	0.158
T_4 (ng/dl)	1.2 ± 0.2	1.3 ± 0.2	0.665

Data are expressed as mean ± standard deviation. KMI, Kupperman menopausal index; BMI, body mass index; WC, waist circumference; FSH, follicle-stimulating hormone; TSH, thyroid stimulator hormone; T_4, free tyroxine.
* Significantly different between groups ($p<0.05$) (independent t-test).

Table 2
Baseline endometrial and vaginal assessment of all subjects

Parameter	Soy isoflavone ($n=38$)	Placebo ($n=38$)	p-Value*
Endometrial thickness (mm)	3.0 (2.2–4.1)	3.7 (2.6–4.1)	0.207
MV	50 (0–52)	50 (2.8–51)	0.667
Parabasal cell (%)	3.5 (0–100)	2.0 (0–95)	0.221
Intermediate cell (%)	76.5 (0–95)	90.0 (0–98)	0.140
Superficial cell (%)	2.0 (0–7)	0.5 (0–6.3)	0.830
Vaginal pH	6 (5.5–7)	6 (5.5–6.5)	0.749

Data are expressed as median with 25th and 75th percentiles in parentheses. MV, Maturation value.
* Significantly different between groups ($p<0.05$) (Wilcoxon Test).

baseline endometrial and vaginal assessment, with no statistically significant differences ($p>0.05$) between groups.

Fig. 1 shows the frequency of hot flushes in both groups throughout the 10 months of the study. At baseline, the mean number of hot flushes was 9.6 ± 3.9 and 10.1 ± 4.9 (mean ± S.D.) per day in the isoflavone group and in the placebo group, respectively. At the end of the study, the participants who were taking soy isoflavone showed a statistically significant reduction in the mean number of hot flushes as compared to those on placebo (3.1 ± 2.3 and 5.9 ± 4.3 per day, respectively; $p<0.001$). The analysis of the Kupperman Menopausal index mean values showed a significant reduction in both groups. When hot flush severity was separately evaluated, a significant reduction in hot flush

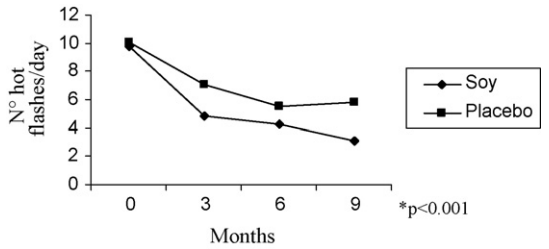

Fig. 1. Mean change in the number of hot flushes from baseline. *Significantly different between groups ($p<0.05$) (independent t-test).

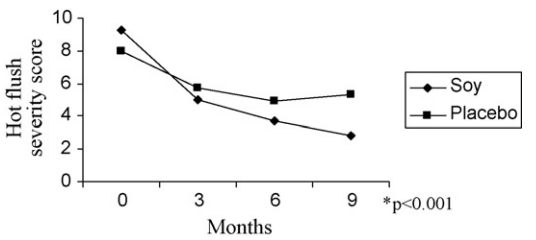

Fig. 2. Mean change in daily hot flush severity score from baseline. *Significantly different between groups ($p<0.05$) (independent t-test).

Table 3
Hormonal profile at baseline and 9 months of intervention (mean ± standard deviations)

Group/parameter	Baseline	9 months	*p*-Values*
FSH (mUI/ml)			
SI	68.1 ± 24.5[a]	66.2 ± 23.7[a]	0.345
PL	66.9 ± 22.9[a]	70.0 ± 20.8[a]	0.080
LH (mUI/ml)			
SI	33.7 ± 11.4[a]	26.5 ± 11.3[a]	0.001
PL	31.5 ± 13.5[a]	25.5 ± 8.8[a]	0.001
E2 (pg/ml)			
SI	21.6 ± 3.3[a]	22.5 ± 4.5[a]	0.320
PL	23.1 ± 5.7[a]	22.6 ± 5.2[a]	0.697

SI, soy isoflavone group; PL, placebo group; FSH, follicle-stimulating hormone; LH, luteinizing hormone; E_2, estradiol.
[a] Without significantly different between groups ($p < 0.05$) (independent *t*-test).
* Significantly different between moments ($p < 0.05$) (paired *t*-test).

severity was observed in the soy isoflavone group as compared with the placebo group (Fig. 2). At baseline, mean daily hot flush severity was 9.3 ± 2.5 in the soy group and 8.0 ± 2.6 in the placebo group, while at the end of the study, it was 2.8 ± 2.4 and 5.3 ± 3.0, respectively ($p < 0.001$). In 36.8% of the subjects (14/38) in the isoflavone group, this symptom completely disappeared, and 10.5% of the participants in the placebo group (4/38) reported total relief from hot flushes.

Table 3 shows FSH, LH, and estradiol average values in both groups at baseline and at 10 months of follow-up. At the end of the study, mean LH values were significantly reduced in both groups ($p < 0.05$). No significant changes in total cholesterol, LDL, HDL, or triglycerides baseline values were found in the isoflavone group. However, in the placebo group, HDL mean values were reduced after 10 months, whereas triglycerides significantly increased ($p < 0.05$). No significant differences in these parameters were observed between groups (Table 4).

Table 5 shows the plasma concentration of daidzein and genistein in all participants after 10 months of intervention. There were statistically significant differences between the groups ($p < 0.05$). The subjects given standardized soy extract showed significantly higher detectable levels of both isoflavones than those in the placebo group.

Table 4
Lipid profile at baseline, 4 and 9 months of intervention (mean ± standard deviations)

Group/parameter	Baseline	4 months	9 months	*p*-Value*
TC (mg/dl)				
SI	215.0 ± 35.6[a]	220.2 ± 36.7[a]	217.3 ± 39.7[a]	0.438
PL	207.7 ± 37.6[a]	209.1 ± 39.5[a]	210.2 ± 37.4[a]	0.801
HDL (mg/dl)				
SI	50.1 ± 10.3[a]	49.5 ± 12.5[a]	52.3 ± 8.3[a]	0.101
PL	52.3 ± 13.3[a]	51.8 ± 12.5[a]	49.7 ± 14.8[a]	0.043
LDL (mg/dl)				
SI	134.2 ± 31.7[a]	137.7 ± 35.4[a]	135.7 ± 34.2[a]	0.330
PL	126.0 ± 31.7[a]	126.4 ± 32.3[a]	127.3 ± 37.9[a]	0.984
TG (mg/dl)				
SI	152.7 ± 64.6[a]	150.4 ± 65.1[a]	138.5 ± 50.4[a]	0.230
PL	147.4 ± 78.2[a]	147.1 ± 75.4[a]	170.5 ± 73.4[a]	0.0059

SI, soy isoflavone group; PL, placebo group; TC, total cholesterol; TG, triglycerides.
[a] Without significantly different between groups ($p < 0.05$) (independent *t*-test).
* Significantly different between moments ($p < 0.05$) (ANOVA–Tukey's).

Table 5
Daidzein and genistein plasma levels at 9 months of intervention (mean ± standard deviations)

Parameter	Soy isoflavone ($n = 38$)	Placebo ($n = 38$)	p-Values*
Daidzein (μmol/dl)	220.4 ± 53.5	125.4 ± 27.9	<0.0001
Genistein (μmol/dl)	144.3 ± 50.5	68.1 ± 19.5	<0.0001

* Significantly different between groups ($p < 0.05$) (independent t-test).

Neither soy extract nor placebo exerted any meaningful effects on vaginal maturation values or vaginal pH. In addition, no estrogenic effect on endometrial thickness was seen by transvaginal ultrasonography. At the end of the study, the median endometrial thickness was 2.4 mm and 2.8 mm in the soy and placebo group, respectively. Body mass index and waist circumference remained unchanged in both groups. No changes on mammographic image were observed in all patients. Seven of the 38 soy group subjects (18.4%) and 4 of the 38 placebo group subjects (10.5%) reported adverse experiences, most frequently in the gastrointestinal system. No serious adverse event related to isoflavone treatment was reported.

4. Discussion

Soy protein or isoflavones dietary supplements are regularly used by millions of women for symptoms associated with menopause or for other purported health benefits [23]. This study showed that the regular consumption of soy isoflavone extract (100 mg) produces a significantly higher reduction in the number of hot flushes than placebo, and has a positive impact quality of life. A reduced frequency of typical climacteric problems was also demonstrated by the significant drop in Kupperman Menopausal index observed in both groups, which suggests a placebo effect. However, when hot flush severity was separately analyzed, a decrease was observed among the women that received soy isoflavone when compared to the placebo group. These results are in agreement with other studies that reported improvement of vasomotor symptoms with standardized soy isoflavone extract in tablets [12–15,24]. Howes et al. [25] conducted a systematic review and meta-analysis of all randomized controlled trials of isoflavone supplementation to determine the efficacy of isoflavone therapy in reducing the number of daily menopausal flushes, and found that isoflavone supplementation was associated with a significant reduction in flushes, even though marked heterogeneity was found among the studies. The percentage reduction in flushes was significantly related to the number of baseline flushes per day and the dose of isoflavone used.

In a previous double-blind, placebo-controlled study of 50 postmenopausal women with contraindication for conventional hormone therapy, we demonstrated alleviation of vasomotor symptoms in 44% of the postmenopausal women using soy germen versus a 10% decrease with placebo [17]. In this present study, total relief of hot flushes occurred in 36.8% of the women on soy isoflavone. This rate is modest when compared to the 77% reduction reported in a meta-analysis of trials using estrogen therapy [26]. Yet, it is greater than the average obtained with placebo in the studies included in the same meta-analysis. Isoflavone supplementation for the treatment of menopausal flushing might, therefore, appear to be less effective than estrogen therapy in suppressing the frequency of menopausal symptoms. However, since many women choose not to undergo hormone therapy, the superiority of isoflavone over placebo may be useful to them [25].

Recent reviews of clinical trials have examined the efficacy of alternative menopause treatments, including soy isoflavones supplements, to alleviate vasomotor symptoms [27,28]. Yet, the efficacy and adverse effects of complementary/alternative and nonhormonal therapies are unclear [28]. Nedrow et al. [27], in a systematic evidence review, evaluated the effectiveness of complementary and alternative therapies in the management of menopausal symptoms. From the 70 randomized controlled trials that met inclusion criteria, 48 studies of phytoestrogens and other biological-base agents showed mixed results. Similarly, Nelson et al. [28] assessed the efficacy and adverse effects of nonhormonal therapies for menopausal hot flushes by reviewing published randomized controlled trials.

From 4249 abstracts, 43 trials met inclusion criteria, including 17 trials of isoflavone extracts. The data collected did not support the efficacy of red clover isoflavone extracts and presented mixed results for soy isoflavone extracts. Data interpretation is further complicated by the nature of research. Currently published trials are generally small, of short duration, and use inadequate methods. The standardization of biological products is poor, making direct comparisons difficult [28]. In fact, besides the large numbers of compounds tested, a comparison of the effects among the different soy products can be complicated by the variability in the concentration of active compounds from product to product [12].

Isoflavones are primarily found in soy products as glycosides and are further conjugated and absorbed in the gastro-intestinal tract. Peak serum isoflavone levels typically occur between 3 and 7 h post-ingestion [29]. Approximately 30–40% of the general population in the United States possess gut microflora that converts the isoflavone daidzein to the more estrogenic dihydroxy isofalvan equol, and this conversion process may provide some added effectiveness from isoflavones for this particular population [30]. There is variation in isoflavone metabolism and absorption among individuals, which can lead to variation in plasma concentrations of parent isoflavones and their metabolites [31]. In this study, a significant increase in genistein and daidzein blood levels was observed in the users of the standardized soy isoflavone extract when compared to the placebo group, showing adequate absorption and compliance to the treatment. The concentrations of the different isoflavone metabolites, as well as their clinical effects, vary widely from individual to individual even when a controlled quantity is administered. Therefore, it is difficult to determine the ideal dosage. Some recommend from 30 mg to 100 mg/day [2,32,33].

Williamson-Hughes et al. [6] evaluated published studies using well-characterized isoflavone-containing supplements to determine whether or not the observed effects were attributable to differences in the composition of isoflavones. In five studies, the product provided more than 15 mg of genistein and a statistically significant decrease in hot flushes was consistently reported. However, of the six studies that provided less than 15 mg, only one reported a significant decrease in symptoms. Reports concluding that isoflavone supplements do not significantly reduce hot flushes may be incorrect. In light of these observations, the evaluation of isoflavone effects should pay greater attention to the specific composition within soy supplements. The standardized soy isoflavone extract used herein contained over 15 mg of genistein, which could account for the beneficial effects observed on climacteric symptoms.

A soy-rich diet may benefit the cardiovascular system due to its favorable effect on lipid profile [10,34]. Some studies have associated isoflavone with a significant reduction in LDL and triglycerides, and increase in HDL [15,17,35]. In our study, no estrogenic activity of soy isoflavone was recognized on lipid profile. However, in the placebo group, the mean values of HDL reduced after 9 months, whereas triglycerides significantly increased. In a recent meta-analysis of the effects of soy protein on lipid profile, Zhan and Ho [36] reviewed 23 trials published from 1995 to 2002, and reported that isoflavone was associated with significant decreases in plasma total cholesterol (3.77%), LDL (5.25%) and triglycerides (7.27%), and increases in HDL (3.03%). These changes were related to the level and duration of intake [36]. It is noteworthy that most of our participants exhibited a normal lipid profile and that the hypocholesterolemic effect of soy intake seems to be significantly related to pretreatment plasma cholesterol [37,38].

Our study also demonstrated that isoflavone had no estrogenic action on the reproductive tract. No change in endometrial thickness, mammography and vaginal maturation resulted from the daily administration of 100 mg of soy isoflavone over 9 months. This finding is in line with other investigations [12,15,24,39]. Previous evaluations of endometrial thickness showed no differences between isoflavone and placebo groups during the course of the trials [28]. Soy isoflavone extract in tablets was generally well tolerated and ingested with a high degree of compliance. No serious adverse event related to isoflavone treatment was reported. For Nelson et al. [28], adverse effects did not differ between isoflavone and placebo groups, although they were not well-characterized in several trials. Gastrointestinal symptoms were generally the most common adverse effects observed with both isoflavone and placebo.

In summary, our results show that 100 mg of the standardized soy isoflavone extract studied was associated with good compliance and safety, and exerted an effect on vasomotor symptoms, decreasing the number

and severity of hot flushes in postmenopausal women. Soy isoflavone extract capsules provide an alternative to the many women who choose to not undergo hormone therapy to relieve menopausal symptoms.

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