Equol Improves Menopausal Symptoms in Japanese Women*1,2

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Abstract

It has been well documented that the frequency of vasomotor menopausal symptoms, such as hot flashes and night sweats, of Japanese menopausal women is less than that of Western women. High intake of soy isoflavones in the traditional Japanese diet has been postulated as the possible explanation of the difference. Epidemiological studies have reported that the content of equol, which is a biologically active metabolite of the isoflavone, daidzein, is lower in the women who complain of severe vasomotor symptoms. To investigate the involvement of equol in the manifestation of menopausal symptoms, especially vasomotor symptoms, and the possible therapeutic role of a supplement containing equol (natural S-equol developed by Otsuka Pharmaceutical) on the menopausal symptoms of Japanese women, 3 randomized clinical trials were conducted. The studies indicated that a daily dose of 10 mg of natural S-equol improved menopausal symptoms. In the confirmation study, menopausal women who were equol nonproducers who consumed 10 mg/d of natural S-equol for 12 wk had significantly reduced severity and frequency of hot flashes as well as a significant reduction in the severity of neck or shoulder stiffness. The equol-ingesting group also showed trends of improvement in sweating and irritability and a significant improvement in the somatic category symptoms. Thus, it is concluded that the supplement containing natural S-equol, a novel soybean-derived functional component, has a promising role as an alternative remedy in the management of menopausal symptoms. J. Nutr. 140: 1386S–1389S, 2010.

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principal isoflavones found in soybeans and most soy foods (6). It was reported that approximately one-half of the Japanese population and 70–80% of the U.S. population cannot produce equol after ingesting soy foods or consuming isoflavones directly (7–9). One-half of middle-aged Japanese women (n = 95) excreted detectable equol levels in 24-h urine, but ~70–80% of the younger generation of women (n = 68) and men (n = 75) showed undetectable levels [10 µg/L (41.3 nmol/L) of equol] was the detectable limit by HPLC in the current study (unpublished data). The inability of some humans to produce equol has been attributed to individual differences in gut microflora (10). A study on urinary isoflavone excretion of Japanese women (n = 106, aged 29–78 y) reported that 24-h urinary equol excretion ranged from 0.4 to 123.6 µmol (11).

Uchiyama et al. (12) investigated the relationship between urinary excretion of isoflavones and menopausal symptoms in peri- and postmenopausal Japanese women. Surveys were mailed to 116 dietitians (aged 40–60 y) living in Fukuoka prefecture located in Kyushu Island in southwest Japan, and 108 participated in the study (93.1% response rate). The 24-h urine samples were collected for the measurement of isoflavonoids by HPLC using methods reported elsewhere (13) and the simplified menopausal index (SMI) (14) was used to assess 10 menopausal symptoms. Urinary excreted amounts of total isoflavonoids, genistein, daidzein, and equol of all participants were 38.9 ± 29.2, 19.6 ± 15.1, 10.0 ± 8.9, and 9.3 ± 14.1 µmol/24 h (mean ± SD), respectively. Genistein and daidzein were detected in all samples, but equol was only detected in the urine of 51.6% of participants.

From the total participants, 46 postmenopausal women (whose last menstrual period had occurred >6 mo prior) were selected for the analysis of menopausal symptoms using SMI (14). The participants were divided into 2 groups according to their SMI score: a high score group (SMI score >15, daily life was disturbed, n = 23) and a low score group (SMI score <15, daily activities were not disturbed, n = 23). Urinary excretion of genistein and daidzein for women in the high and low score groups did not differ; however, the amount of equol excreted in the urine was lower in the high score group than in the low score group (P < 0.05) (Fig. 1). These results indicate that women who can metabolize daidzein into equol by their own intestinal bacteria report milder menopausal symptoms.

![FIGURE 1](https://academic.oup.com/jn/article-abstract/140/7/1386S/4689030)

**FIGURE 1** Relation between urinary equol excretion and menopausal symptoms in Japanese women. Data are means ± SEM, n = 23. Low: SMI score <15; daily activities were not apparently disturbed. High: SMI score >15; daily life was disturbed. Statistical analysis was carried out using Wilcoxon’s Rank Sum test. *Different from low group, P < 0.05. Adapted from Uchiyama et al. (12) with permission from The Japan Menopause Society.

**Development of natural S-equol supplement**

To access the efficacy of equol as an alternative treatment for menopausal symptoms, a natural S-equol supplement was developed. Equol exists as an enantiomer, S-isomer and R-isomer, but only the S-isomer is biologically produced in vivo. A lactic acid bacterium, *Lactococcus garvieae* (*Lactococcus 20–92 strain*), with equol-producing capabilities was identified and isolated from human feces (15,16). Recently, a standardized product of natural S-equol supplement was developed by Otsuka Pharmaceutical (17). The supplement was formed from fermentation of a soy germ solution by *Lactococcus 20–92*. The concentration of S-equol in the supplement ranged from 0.5 to 0.8% of natural S-equol and the supplement does not contain the R-equol isomer. The micronutrient content of the supplement was not significantly different from the original material. Fermentation with *Lactococcus 20–92* revealed that except for isoflavones and some amino acids, the levels of protein, fat, carbohydrate, and other components were not changed by fermentation. The levels of protein, fat, and carbohydrate in the supplement were 38.3, 14.5, and 26.8%, respectively (17). The safety of the supplement was confirmed in genotoxicity, acute and subchronic toxicity, and reproductive and development toxicity tests (17,18).

**Clinical trials on the effects of natural S-equol on menopausal symptoms of Japanese women**

Three randomized clinical studies (pilot study, dose-finding study, and confirmation study) were conducted to investigate the ability of the natural S-equol supplement to relieve menopausal symptoms in Japanese women. In this section, the results of 3 clinical trials are discussed. The protocols were approved by the Institutional Review Board of each site and the studies were carried out according to the guidelines of the Declaration of Helsinki. All women provided written informed consent to participate in the study. The first study has been previously published (13) and the other studies will be submitted for publication separately.

**Pilot study of natural S-equol supplement**

Ishiwata et al. (13) conducted a randomized, double-blind, placebo-controlled trial to investigate the effects of the natural S-equol supplement on menopausal symptoms and mood states in pre-, peri-, and postmenopausal Japanese women. Each package of supplement contained 10, 0.8, and 2.0 mg of equol, daidzein, and genistein, respectively. A total of 134 healthy Japanese female participants (aged 40–59 y) were divided into 3 groups: EQ-1 (n = 44, 10 mg of equol/d), EQ-3 (n = 46, 30 mg of equol/d), and placebo (n = 44). The women were allowed to consume a maximum of 20 mg soy isoflavones from their diet each day. Menopausal symptom score and mood score were self-reported by filling out a menopausal symptom scale and the Profile of Mood States questionnaires. The 24-h urine samples were collected for measurement of isoflavones. Physical examinations and biochemical analyses were conducted at baseline and at the end of the 12-wk period.

It was described in the report (13) that 127 participants (94.8%) completed the trials. Participants who excreted <10 µg/L (41.28 nmol/L) of equol (HPLC detection limit) in 24-h urine samples were defined as equol nonproducers. At baseline, equol producers (n = 46) had fewer total menopausal symptoms (P < 0.05) than nonproducers (n = 81). Somatic symptom scores were reduced in the EQ-3 group compared with the placebo group of postmenopausal equol nonproducers (P < 0.05) (Fig. 2). Postmenopausal nonproducers also reported beneficial effects on...
Japanese women. Values are means ± SD, n = 18 or 19. * * Different from wk 0, P < 0.05 and P < 0.01 (Wilcoxon’s Rank Sum test). a Different from placebo in wk 12 changes, P < 0.05 (Mann-Whitney test). Adapted from Ishiwata et al. (13) with permission from Wolters Kluwer/Lippincott Williams & Wilkins.

**FIGURE 2** Effects of natural S-equol on the menopausal symptom total score (A) and somatic score (Greene climacteric scale) (B) in Japanese women. Values are means ± SD, n = 18 or 19. * * Different from wk 0, P < 0.05 and P < 0.01 (Wilcoxon’s Rank Sum test). a Different from placebo in wk 12 changes, P < 0.05 (Mann-Whitney test). Adapted from Ishiwata et al. (13) with permission from Wolters Kluwer/Lippincott Williams & Wilkins.

depression and fatigue following the ingestion of 30 mg of equol/d. Neither serious adverse events nor abnormal laboratory tests were observed. The authors concluded that natural S-equol supplementation reduced menopausal symptoms in Japanese women who were postmenopausal equol nonproducers.

**Dose-finding study of natural S-equol supplement**

This study aimed to determine the optimal dose of equol for clinical use. A double-blind, placebo-controlled, parallel-group, interventional trial was conducted to investigate the physiological effects of natural S-equol supplementation on Japanese menopausal women who were equol nonproducers. The healthy Japanese menopausal women of 105 were divided into 4 groups: EQ-2 group (n = 26, 2 mg equol/d), EQ-6 group (n = 25, 6 mg equol/d), EQ-10 group (n = 27, 10 mg equol/d), and placebo group (n = 27). Each supplement tablet contained 1.0 mg equol, 0.1 mg daidzein, and 0.1 mg genistein and 2, 6, and 10 tablets/d were ingested orally in the EQ-2, EQ-6, and EQ-10 groups for 12 wk, respectively.

A total of 99 participants (94% response rate) completed the study. Daily consumption of 10 mg of S-equol improved menopausal symptoms of Japanese postmenopausal equol non-producing women, especially shoulder stiffness at 6 wk, compared with the placebo group (P = 0.034). The measurement of uterine endometrial thickness by transvaginal ultrasonography and cytological analysis of vaginal epithelium revealed no biological effect of the supplement on reproductive organs. Thus, we concluded that 10 mg/d of natural S-equol was the effective dose for improving Japanese women’s menopausal symptoms.

**Confirmation study of natural S-equol supplement**

To confirm the physiological effects of the natural S-equol supplement in menopausal Japanese women who were equol nonproducers, an intervention trial (randomized double-blind, placebo-controlled, parallel-group) was conducted. This study consisted of a screening period, ingestion period for 12 wk, and follow-up period for 6 wk. One hundred and sixty postmenopausal women with normal BMI who reported having hot flashes at least once per day were enrolled in the study.

Before enrollment to the trial, all participants were asked to take a placebo for 4 wk (screening period). Participants who showed placebo effects of 50% reduction of SMI after the period were excluded from the final enrollment. During the ingestion period, participants were divided into placebo (n = 60) and equol (n = 66, 10 mg equol/d) groups. The supplement containing equol (5 mg), daidzein (1.2 mg), and genistein (1.4 mg) was orally ingested twice per day (morning and evening) for 12 wk.

The baseline frequencies of hot flashes in the placebo and equol groups were 2.9 ± 2.1 and 3.2 ± 2.4/d (mean ± SEM), respectively. The difference between the 2 groups was not significant. Women who ingested 10 mg/d of natural S-equol for 12 wk had reductions in the frequency and severity of hot flashes compared with those in the placebo group (P = 0.0092 and P = 0.0154, respectively). A greater reduction of the severity of neck or shoulder stiffness was observed for the equol group than that of placebo group, when evaluated using both the questionnaire (P = 0.0149) and the visual analogue scale method (P = 0.0070). After the 6-wk follow-up, the ingested supplement was washed out and rebound responses were observed in hot flashes and neck or shoulder stiffness. No serious adverse events were reported. The results of vital sign and clinical laboratory tests indicated no changes (P > 0.10) after ingestion of 10 mg equol/d for 12 wk.

**Discussion and conclusion**

Because the mental and physical conditions of women in menopause are complicated, health care in this life stage has to be done holistically. By the systemic evaluation of these conditions, the personal management for each woman should be selected. Lifestyle improvements, including diet habits, exercise, and relaxation, should be conducted before medical treatment.

Among the health problems in menopause, menopausal symptoms, especially vasomotor symptoms like hot flashes and sweating, disturb daily activities and quality of life seriously in the great portion of women in Western societies and also for a certain number of Japanese women. In the majority of such women, the symptoms cannot be controlled by nonmedical management and need to be treated by some medical interventions.

Although the mechanism underlying vasomotor symptom has not been fully clarified, the results of various basic and clinical studies have indicated that dramatic changes in the hormonal environment, especially the sharp decline of estrogen, during menopause play key roles. Hormone replacement therapy (HRT) has been widely adapted as a potent evidence-based medicine for menopausal symptoms. The report of a large-scaled study in the United States, the Women’s Health Initiative published in 2002 (19), indicated increasing risks associated with a kind of HRT regimen after long-term use. It is, however, still convincing that the first choice of medical treatment for severe vasomotor symptom is tailor-made HRT under appropriate diagnostic and follow-up medical care.

At the same time, the development of medical treatments for the women suffering from health problems caused by estrogen
deficiency including severe menopausal symptoms who cannot use HRT has been a great issue in clinical practice. It is also expected in menopausal health care that a medical intervention will improve the quality of life in the majority of menopausal Japanese menopausal women who complain of moderate- to mild-grade menopausal symptoms that are not severe enough to be treated by HRT.

In the clinical trial in which a subjective score is used as the end point of evaluation, how to obtain solid data showing the efficacy of a product is particularly critical. The severity of menopausal symptoms is one of the typical cases, which is easily influenced by nonspecific factors inducing placebo effects.

The series of studies described in this paper were attempts to investigate whether natural S-equol supplementation can be a new alternative. To confirm the efficacy of the compound strictly in clinical trials, the confirmation study was designed to include 3 periods. The screening period aimed to exclude the placebo effects of the intervention. At the same time, equol nonproducers were selected by the results of 3 separate measurements of urinary equol contents. Furthermore, special attention was paid to minimizing the bias caused by evaluation methods of interviewers who assessed changes of symptoms. In the follow-up period, it was predicted that a rebound response should appear if the ingested natural S-equol supplement had actual biological effects suppressing menopausal symptoms. As the result, such rebound patterns were observed in the responses of hot flashes and neck or shoulder stiffness. It is indicative that these procedures for standardizing the enrolled population and arrangements adapted in the study made certain contributions to clarifying the importance of equol’s effects on certain menopausal symptoms.

This study confirmed the positive effects of equol on menopausal symptoms and was the first report, to our knowledge, based on a pausal symptoms. clarifying the importance of equol’s effects on certain meno-

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**Literature Cited**

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