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Quantitative efficacy of soy isoflavones on menopausal hot flashes

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WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT

- Menopause hot flashes can be effectively treated with hormone replacement therapy (HRT). However, possible adverse effects of long term HRT have promoted the use of complementary therapies by menopausal women.
- There has been debate on the benefits of soy isoflavones in the treatment of menopausal hot flashes.

WHAT THIS STUDY ADDS

- The effects of soy isoflavones on menopausal hot flashes was quantitated adequately by the model based meta-analysis.
- The developed model provided the robust evidence that soy isoflavones have slight and slow effects in attenuating menopausal hot flashes compared with estradiol.

AIM

This study aimed to quantitate the efficacy of soy isoflavones in the treatment of menopausal hot flashes.

METHODS

Model based meta-analysis (MBMA) was used to quantitate the efficacy of soy isoflavones. We conducted a systemic literature search to build a time–effect model for placebo and soy isoflavones in treating menopausal hot flashes. Studies were identified, subjected to inclusion and exclusion criteria, and reviewed.

RESULTS

From 55 articles, 16 studies of soy isoflavones met the inclusion criteria, and contained 65 and 66 mean effect values in placebo and soy isoflavone groups, respectively, from about 1710 subjects. Interestingly, the developed model was found to describe adequately the time course of hot flashes reduction after administration of placebo and soy isoflavones. Using this model, we found that the maximal percentage change of hot flashes reduction by soy isoflavones was 25.2% after elimination of the placebo effect, accounting for 57% of the maximum effects of estradiol ($E_{max\text{-}estradiol}$ = 44.9%). However, a time interval of 13.4 weeks was needed for soy isoflavones to achieve half of its maximal effects, much longer than estradiol, which only required 3.09 weeks. These results suggest that treatment intervals of 12 weeks are too short for soy isoflavones, which require at least 48 weeks to achieve 80% of their maximum effects.

CONCLUSIONS

Soy isoflavones show slight and slow effects in attenuating menopausal hot flashes compared with estradiol.

Introduction

Menopause, the time in a woman's life when the ovaries lose their reproductive function, is often accompanied by a range of symptoms. However, only hot flashes and vaginal dryness are consistently associated with this stage of life [1]. Fortunately, hot flashes can be effectively treated with hormone replacement therapy (HRT) [2]. However, possible adverse effects [3] of long term HRT have promoted the use of complementary therapies by menopausal women. Recently, soy isoflavones, black cohosh and red clover have been reported to be widely used to relieve menopausal hot flashes [4], but their efficacies remain largely unknown. A meta-analysis published in JAMA in 2006 showed that no definitive evidence existed to support the efficacy of those plant medicines in treating menopausal hot flashes [5]. Nevertheless, this work presented some limitations. First, the efficacy data obtained at different time points were combined, neglecting the effect of time on treatment efficiency. Then, only a small number of studies were included, with most using small sample sizes. The efficacy data of hot flashes reduction usually display a large variability. Therefore, the conclusions need to be re-examined.

An approach recently described as model-based meta-analysis (MBMA) has been developed and used to estimate the comparative efficacy of different treatments [6]. MBMA can be used to distinguish inter-trial, inter-treatment arm and inter-individual variability. It also can test the impact on efficacy of different factors, including dose, duration, drug formulation and other variables. Therefore, compared with the conventional meta-analysis, MBMA is more powerful in identifying treatment effects.

Soy isoflavones are widely present in soy products, which are convenient to take, with little adverse effects. However, it is still unclear how much soy isoflavones should be taken and how long they need to be used. This study used the MBMA method to evaluate quantitatively the efficacy of soy isoflavones on hot flashes, comparing these compounds with estradiol, which has proven efficacy on hot flashes. This work provides useful information for a deeper understanding of the efficacy of soy isoflavones on menopausal hot flashes.

Method

Search strategy

A comprehensive literature search in the PubMed database was performed from January 1990 to January 2014, using the terms 'hot flash' or 'hot flush', and 'soy' or 'genistein' or 'daidzin' or 'glycitein'. Additionally, our search was limited to clinical trials and articles published in English. Inclusion criteria were (1) placebo controlled clinical trial, (2) longitudinal data of frequency or decreased rates of hot flashes provided in tables or graphics and (3) if the study was a crossover design, only data from the first period were analyzed.

Data extraction

A Microsoft Excel database was created to catalogue the relevant features of various studies. The information collected from each selected clinical study included authors, year of publication, sample size, dosage, study duration, subject category and mean percentage of hot flashes reduction at each time point. The mean percentage change in hot flashes from baseline was used as the evaluation index for modelling, thus eliminating the potential baseline effects on the evaluation of treatment efficiency.

Digitizing software Engauge Digitizer (version 4.1, 2002 by Mark Mitchell) was used to extract the graphical data. If only frequency of hot flashes was reported in a study, the mean percentage change in hot flashes from baseline was estimated as follows:

$$E\% = \frac{E_t - E_{baseline}}{E_{baseline}} \cdot 100\%, \quad (1)$$

where E_t is the frequency of hot flashes at time t and $E_{baseline}$ represents the frequency of hot flashes at baseline.

All data were extracted independently by two researchers (LL and LX), and any disagreement between them was resolved by discussion. Data extraction errors between the two researchers should not exceed 2%. Mean values were considered final data.

Model development

We hypothesized that placebo and soy isoflavone effects would vary with time and reach a plateau. Therefore, the effect profiles for placebo and soy isoflavones were described with the sigmoid E_{max} model, with time considered as an independent variable. The basic model was described as follows:

$$E_{i,j} = E_{placebo,i,j} + E_{soy,i,j} + IAV/sqrt(No_{i,j}) + \varepsilon_{i,j}/sqrt(No_{i,j}) \quad (2)$$

where

$$E_{placebo,i,j} = -\frac{(E_{max-placebo} + \eta_{1,i}) \times T_j}{ET_{50-placebo} + T_j} \quad (3)$$

$$E_{soy,i,j} = -\frac{(E_{max-soy} + \eta_{2,i}) \times T_j}{ET_{50-soy} + T_j} \quad (4)$$

In equation 2, $E_{i,j}$ is the observed mean effect in i^{th} study at time j, $E_{placebo,i,j}$ represents the predicted mean placebo

effect in i^{th} study at time j, $E_{soy,i,j}$ is the predicted mean effect of soy isoflavones in i^{th} study at time j, $No_{i,j}$ is the sample size in i^{th} study at time j, IAV represents the inter-treatment arm variability, assumed to be normally distributed with a mean of 0 and variance of $\omega_{arm}^2/No_{i,j}$ and $\varepsilon_{i,j}$ is the residual error in i^{th} study at time j, assumed to be normally distributed with a mean of 0 and variance of $\sigma^2/No_{i,j}$. IAV and $\varepsilon_{i,j}$ are weighted by the sample size.

In equations 3 and 4, $E_{max\text{-}placebo}$ is the maximal effect of placebo, $E_{max\text{-}soy}$ represents the maximal effect of soy isoflavones, $ET_{50\text{-}placebo}$ is the time to achieve 50% of $E_{max\text{-}placebo}$, $ET_{50\text{-}soy}$ represents the time to achieve 50% of $E_{max\text{-}soy}$, T_j is the time j, $\eta_{1,i}$ represents the inter-trial variability of $E_{max\text{-}placebo}$ and $\eta_{2,i}$ is the inter- trial variability of $E_{max\text{-}soy}$. $\eta_{1,I}$; $\eta_{2,i}$ are assumed to be normally distributed with a mean of 0 and variance of ω_1^2 and ω_2^2.

Once the basic model was established, the dose of soy isoflavones was considered a covariate to be added into $E_{max\text{-}soy}$. A difference in objective function value (OFV) of 6.63 (χ^2, $\alpha = 0.01$, d.f. = 1) was considered statistically significant in the covariate model building process.

Model validation

The accuracy of the model fit was evaluated by graphic assessment. Monte Carlo simulations were performed 1000 times to predict 90% confidence intervals of the effects of placebo and soy isoflavones.

The final model was also evaluated by the leave-one-out cross validation method. Briefly, values from one trial were sequentially dropped from the full data set, and the final model was applied. The parameter estimates obtained from each data set were compared to investigate stability of the final model.

Software

The model estimation and simulation were performed using NONMEM 7 (Level 1.0, ICON Development Solutions, USA). Diagnostic graphics and visual predictive check were performed using the R software (version 3.0.1, The R Foundation of Statistical Computing).

Results

Characteristics of the selected studies

A total of 55 studies were assessed for inclusion in the analysis, and 39 were excluded based on study design that did not meet our inclusion criteria. The remaining 16 studies [7–22] containing 66 and 65 mean effect data of soy isoflavones and placebo, respectively, were included for model building (Figure 1, Table 1).

The included studies were published between 1998 and 2012 in some professional journals, such as Menopause and Maturitas. All the studies taken together comprised 1710 subjects, ranging from 24 to 236 individuals per report (median 90). Study duration ranged between 4 weeks and 2 years, with a median of 12 weeks (Table 2).

Model establishment

The parameter estimates in the basic model are shown in Table 2. The typical value of $E_{max\text{-}soy}$ was estimated at 25.2%,

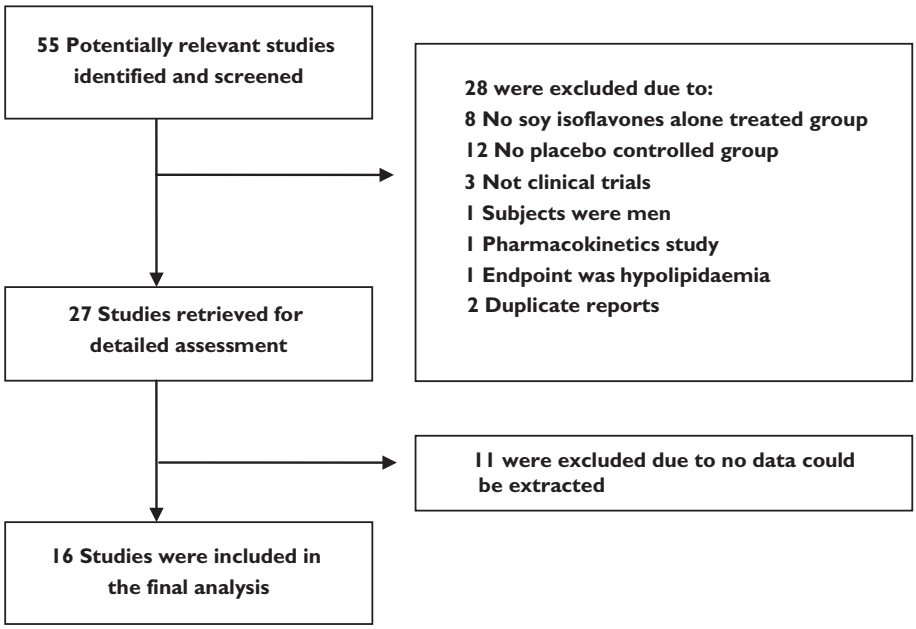

Figure 1
Flowchart for screening relevant articles

Table 1

Summary of included placebo-controlled trials of soy isoflavones

Source	Number of participants	Sample	Therapy	Study duration	Study design	Baseline of hot flashes/results
Albertazzi *et al*. [7]	104	Post-menopausal women; ≥7 hot flashes per day; average age, 52–53 years; Italy.	Isolated soy protein, 76 mg of isoflavones (aglycone units)	12 weeks	Parallel	The baseline frequency: 11.4(10.7–12.7) per day for the soy group, 10.9(10.2–11.8) per day for the placebo group; Reduced frequency (45% *vs.* 30%, $P < 0.01$) with soy *vs.* placebo.
Quella *et al*. [8]	177	Women had breast cancer; ≥14 hot flashes per week; over 18 years of age; United States	Soy tablet, 150 mg daily (with 40–45% genistein, 40–45% daidzein, and 10–20% glycitein)	4 weeks	Crossover	The baseline frequency: 7 ± 4.5 per day; No differences in frequency or severity score between groups, 36% of placebo *vs.* 24% of soy tablet.
Scambia *et al*. [9]	39	Post-menopausal women; average age, 53–54 years; Italy	Soy tablet, 50 mg daily of genistein and daidzein	6 weeks	Parallel	The baseline frequency: 33 ± 5.1 per week for the soy group, 27 ± 5.1 per week for the placebo group; Reduced frequency (40% *vs.* 25%, $P < 0.05$) with soy tablet *vs.* placebo.
Upmalis *et al*. [10]	177	Healthy post-menopausal women; ≥5 hot flashes per day; aged 50 years or older; American	Soy tablet, 50 mg daily of genistein and daidzein	12 weeks	Parallel	The baseline frequency: 8.8 ± 6.2 per day for the soy group, 9.4 ± 6.0 per day for the placebo group; No difference in frequency between groups.
Knight *et al*. [11]	24	Post-menopausal women; ≥3 hot flashes per day; aged 40–45 years; Australia.	Soy powder, 134.4 mg daily of genistein, daidzein and glycitein compounds.	12 weeks	Parallel	The baseline frequency: 50.2 ± 13.6 per week for the soy group, 56.2 ± 26.5 per week for the placebo group; Reduced frequency (43% *vs.* 20%, $P = 0.32$) with soy *vs.* placebo
Faure *et al*. [12]	75	Post-menopausal women; ≥7 hot flashes per day; average age, 53–54 years; France	Soy capsule, 70 mg daily of genistein and daidzein	16 weeks	Parallel	The baseline frequency: 10.1 ± 6.4 per day for the soy group, 9.4 ± 3.4 per day for the placebo group; Reduced frequency with soy *vs.* placebo (61%*vs.* 21%, $P = 0.01$).
Van Patten *et al*. [13]	157	Women had breast cancer; ≥10 hot flashes per week; average age, 55–56 years; England.	Soy beverage, 90 mg of isoflavones	12 weeks	Parallel	The baseline frequency: 7.1 ± 4.3 per day for the soy group, 7.4 ± 6.4 per day for the placebo group; No differences in frequency between groups. Reduced frequency (25.4% *vs.* 33.8%) with soy *vs.* placebo.
Penotti *et al*. [14]	62	Post-menopausal women; ≥7 hot flashes per day; aged 45–60 years; Italy	Soy tablet, 72 mg daily isoflavone	6 mo	Parallel	The baseline frequency: 9.5 ± 3.4 per day for the soy group, 8.8 ± 1.4 per day for the placebo group; 40% reduction in frequency in both groups; No between-group differences.

Crisafulli *et al*. [15]	90	Post-menopausal women; aged 47–57 years; Italy.	Genistein, 54 mg daily;	1y	Parallel	The baseline frequency: 4.6 ± 3.2 per day for the genistein group, 4.7 ± 3.2 per day for the placebo group; Reduced frequency for genistein *vs.* placebo (24% mean difference, $P < 0.01$).
Campagnoli *et al*. [16]	36	Healthy post-menopausal women; ≥5 hot flashes per day; aged 45–58 years; Italy	200 mg standardized soy extract, (with15% daidzein, 15% genistein, 20% saponin)	12 weeks	Crossover	The baseline frequency: 38 per week; No differences in frequency between groups. Reduced frequency (26.5% *vs.* 24.3%) with Soy *vs.* placebo.
Levis *et al*. [17]	99	Post-menopausal women; aged 45–60 years; Canada	Muffins with 42 mg of isoflavones	16 weeks	Parallel	The baseline frequency: 4.1 ± 2.4 per day for the isoflavones group, 4.7 ± 3.0 per day for the placebo group; Reduced frequency (17.4% *vs.* 19.7%) with soy *vs.* placebo.
Nahas *et al*. [18]	80	Healthy post-menopausal women; ≥5 hot flashes per day; aged 45 years or older; Brazil	Soy capsule, 100 mg daily of isoflavone	10 mo	Parallel	The baseline frequency: 9.6 ± 3.9 per day for the soy group, 10.1 ± 4.9 per day for the placebo group; Reduced frequency (3.1 ± 2.3 and 5.9 ± 4.3 per day, $P < 0.001$) with soy tablet *vs.* placebo.
D'Anna *et al*. [19]	236	Post-menopausal women; average age, 54–55 years; Italy	Genistein tablet, 54 mg daily of total isoflavone	24 mo	Parallel	The baseline frequency: 4.4 ± 3.4 per day for the genistein group, 4.2 ± 3.7 per day for the placebo group; In the genistein group, there was a significant decrease in the mean number (–56.4%) of the hot flushes.
Ferrari [20]	180	Post-menopausal women or 6 weeks after bilateral oophorectomy; ≥5 hot flashes per day; aged 40–65 years; Italy	Soybean extract, 80 mg daily isoflavones (with 60 mg genistein and genistin, 16 mg daidzein and daizin, 3 mg glycitein and glycitin).	12 weeks	Parallel	The baseline frequency: 8.0 ± 3.3 per day for the isoflavones group, 7.5 ± 2.8 per day for the placebo group; Reduced frequency (41.2% *vs.* 29.3%, $P = 0.023$) with soy *vs.* placebo at 12 weeks.
Evans *et al*. [21]	84	Healthy post-menopausal women; ≥40 hot flashes per week; aged 40–65 years; Canada	Capsules, 30 mg genistein	12 weeks	Parallel	The baseline frequency: 9.4 ± 3.8 per day for the genistein group, 9.9 ± 3.9 per day for the placebo group; Reduced frequency (51.2% *vs.* 29.8%, $P = 0.046$) with genistein *vs.* placebo at 12 weeks.
Ye *et al*. [22]	90	Post-menopausal women; aged 45–60 years; China	Soy germ isoflavone extract powder, low dose group, 84 mg daily isoflavones; high dose group, 126 mg daily isoflavones (with 52% daidzin, 15% genistin, and 33% glycitin aglcone equivalents)	24 weeks	Parallel	The baseline frequency: 20 ± 11.0 per week for the genistein group, 21 ± 12.5 per week for the placebo group; Reduced frequency (44.3% *vs.* 48.5% *vs.* 27.8, $P < 0.01$) with isoflavone low dose group *vs.* isoflavone high dose group *vs.* placebo.

Table 2
Parameter estimation of soy isoflavones and placebo

Parameters	Value	RSE (%)	95% CI
$E_{max\text{-}placebo}$, %	35.8	11.3	27.9, 43.7
$E_{max\text{-}soy}$, %	25.2	26.9	11.9, 38.5
$ET_{50\text{-}placebo}$, week	2.99	31.6	1.1, 4.8
$ET_{50\text{-}soy}$, week	13.4	30.1	5.5, 21.3
$\eta(E_{max\text{-}placebo})$, %	13.1	48.9	0.5, 25.7
$\eta(E_{max\text{-}soy})$, %	24.4	43.0	3.8, 45.0
Correlation coefficient $[\eta(E_{max\text{-}placebo}) - \eta(E_{max\text{-}soy})]$	0.424	–	–
IAV, %	21.0	23.5	11.3, 30.7
ε, %	26.5	121.6	0, 89.7

Table 3
Parameter estimation of estradiol and placebo

Parameters	Value	RSE (%)	95% CI
$E_{max\text{-}placebo}$, %	55.6	5.4	49.7, 61.5
$E_{max\text{-}estradiol}$, %	44.9	7.0	38.7, 51.1
$ET_{50\text{-}placebo}$, weeks	1.96	11.4	1.5, 2.4
$ET_{50\text{-}estradiol}$, weeks	3.09	19.1	1.9, 4.2
$\eta(E_{max\text{-}placebo})$, %	12.4	26.6	5.9, 18.9
$\eta(E_{max\text{-}estradiol})$, %	12.3	39.1	2.9, 21.7
Correlation coefficient $[\eta(E_{max\text{-}placebo}) - \eta(E_{max\text{-}estradiol})]$	0.453	–	–
IAV, %	0, Fixed	–	–
ε, %	46.2	14.4	33.2, .59.2

and the time to achieve 50% of $E_{max\text{-}soy}$ was 13.4 weeks. Meanwhile, the typical value obtained for $E_{max\text{-}placebo}$ was 35.8% and 2.99 weeks were needed to achieve 50% of $E_{max\text{-}placebo}$. Inter-trial variability of the $E_{max\text{-}soy}$ showed moderate correlation with that of the $E_{max\text{-}placebo}$, with a correlation coefficient of 0.424.

The predicted $E_{max\text{-}soy}$ value was not significantly correlated with the dose of soy isoflavones (Figure 2D). After the dose factor of soy isoflavones was introduced into the $E_{max\text{-}soy}$ parameters, the OFV was not significantly decreased. Therefore, the basic model established above was considered final.

Model evaluation

Figure 2A depicts the application of the final model to each individual trial, showing that the data observed in trials were consistent with the predicted values. The goodness-of-fit plots for the final model are presented in Figure 2B. Generally, there was good accordance between observed (OBS) and population model-predicted (PRED) effects, and between OBS and individual model-predicted (IPRED) effects. The WRES magnitude was small and randomly distributed around a straight line through 0, and located within ± 4 from the centre.

Monte Carlo simulations (1000 times) showed that the 90% CI of the 5%, median and 95% predicted percentiles covered the corresponding observed data, which indicated that the model had an adequate prediction capability (Figure 2C).

In order to test the impact of each individual study on the model parameters, leave-one-out cross validation was conducted. The results (Figure 3) showed that expect for ε, the distribution of most parameters was stable and only slightly affected by the individual trial.

Efficacy comparison with estradiol

The efficacy of estradiol on menopausal hot flashes was also evaluated by MBMA (Supply materials) [15, 23–50]. The results showed that the typical value of $E_{max\text{-}estradiol}$ was estimated at 44.9%, which is about 1.8 times that of $E_{max\text{-}soy}$, and the time to achieve 50% of $E_{max\text{-}estradiol}$ was only 3.09 weeks, much shorter than that of soy isoflavones (Table 3). It should be noted that the $E_{max\text{-}placebo}$ in estradiol trials was 55.6%, overtly higher than the value obtained from trials with soy isoflavones.

Due to the large difference in ET_{50} between soy isoflavones and estradiol, estradiol reached 80% of its E_{max} after 12 weeks treatment, while soy isoflavones only reached 47% of E_{max}. It was only after at least 48 weeks that treatment with soy isoflavones resulted in 80% of E_{max} (Figure 4).

Discussion

Soy isoflavones are phytoestrogens with potential hormonal activity due to their structural similarity with 17-β-estradiol. The increasing availability of soy isoflavones in food and through the use of supplements has prompted extensive research on biological benefits to menopausal women in chronic disease prevention and health maintenance. It has been reported that approximately 70–80% of USA women of menopausal and peri-menopausal age experience hot flashes, in comparison with approximately 10–20% of Asian women [51]. Interestingly, the average blood concentration of the soy isoflavone genistein in Asian women is approximately 25 ng ml^{-1}, while only 2 ng ml^{-1} is found in USA women [51]. The sharp contrast between frequency of hot flash symptoms and soy genistein concentrations has led researchers to assess soy isoflavones for their potential to prevent hot flashes.

Unfortunately, according to the published meta-analyses, there is still an ongoing debate on the efficiency of soy isoflavones in treating menopausal hot flashes. In the Forest plot [1], it seemed that the treatment efficiency in patients administered soy isoflavones was superior to those treated with placebo. However, no statistical difference ($P > 0.05$) was observed due to a wide confidence

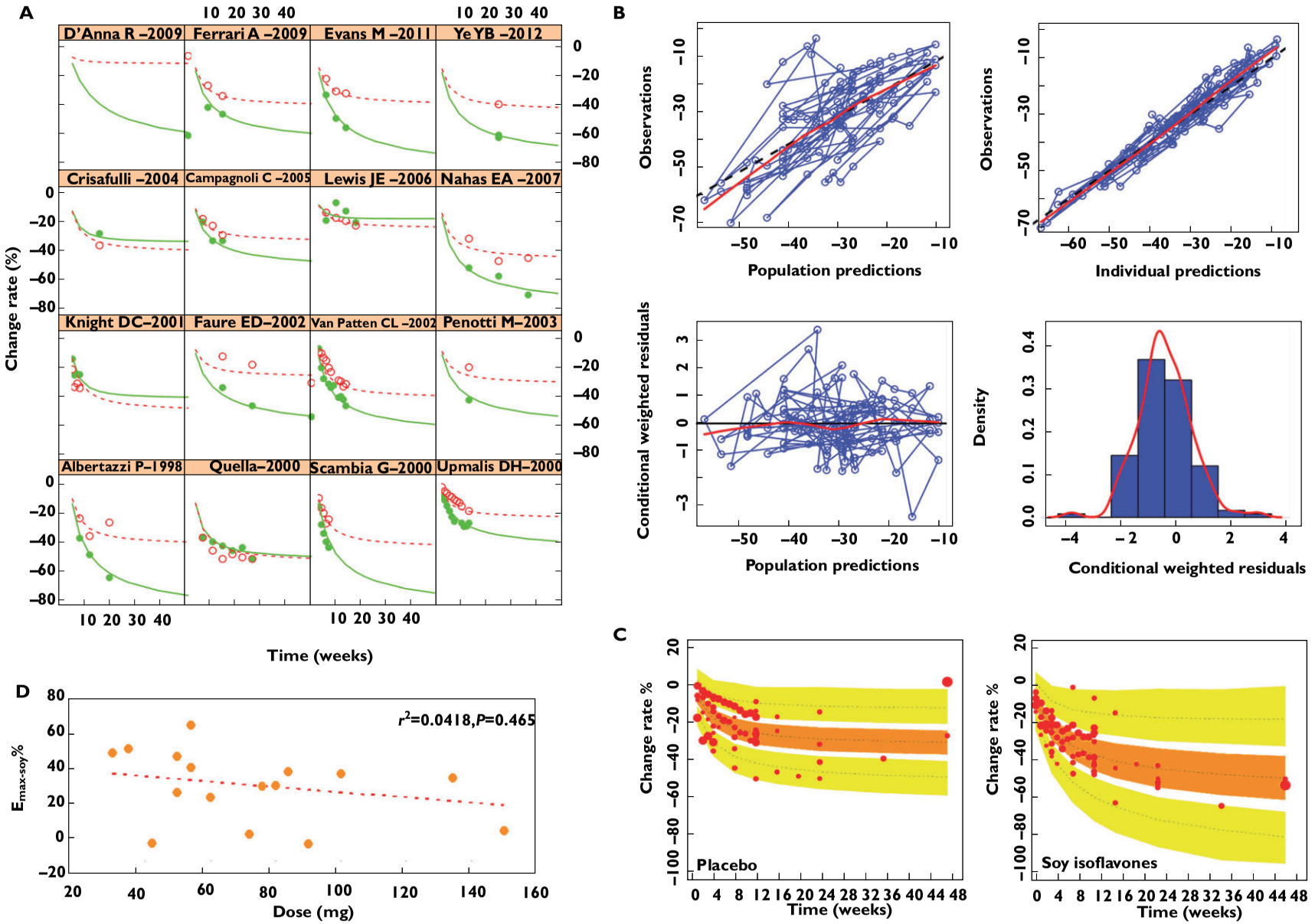

Figure 2

Model evaluation. (A) Time course of mean % change of hot flashes from baseline for each individual study. (B) Model diagnosis graph. (C) Prediction corrected visual predictive check of the model. The dashed lines are the 5th, 50th and 95th percentiles of observed data. The shaded areas are the corresponding confidence intervals of the simulated data. The solid points represent observed data, and the symbol size is proportional to the number of subjects in each study. (D) The relationship between the dose of soy isoflavones and the predicted $E_{max\text{-}soy}$ value. ●, observed value-soy isoflavones; ○, observed value-placebo; ——, predicted value-soy isoflavones; ·····, predicted value-placebo

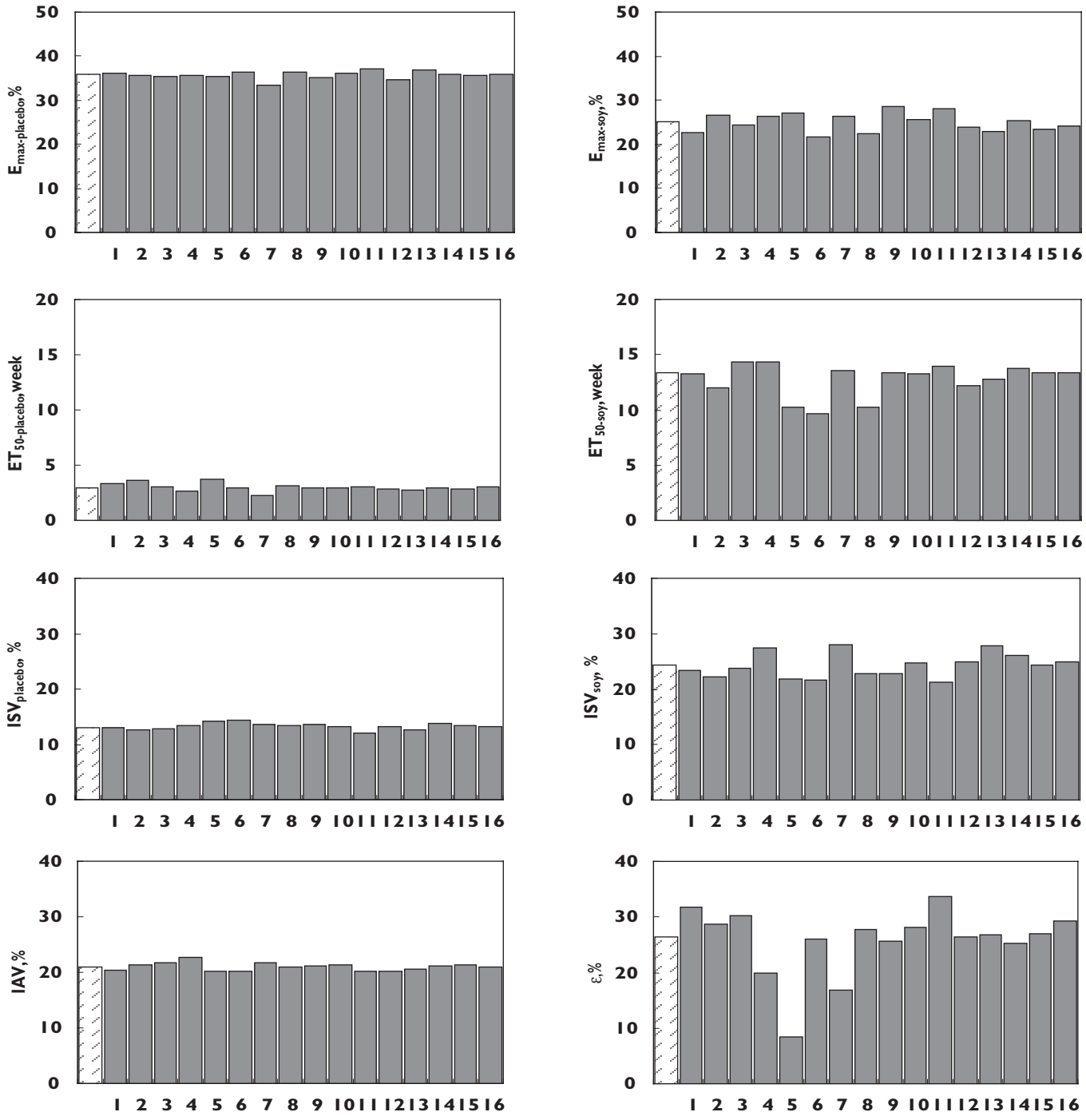

Figure 3

Bar charts of the typical parameter values obtained from the final model in a leave-one-out method. The abscissa represents the study number dropped from the full data and the ordinate, the typical value of parameters. The left shadow bar represents parameters obtained from full data. Study 1: Faure *et al.* [12], 2: Penotti *et al.* [14], 3: Scambia *et al.* [9], 4: Upmalis *et al.* [10], 5: Quella *et al.* [8], 6: Crisafulli *et al.* [15], 7: Albertazzi *et al.* [7], 8: Knight *et al.* [11], 9: Van Patten *et al.* [13], 10: Campagnoli *et al.* [16], 11: Levis *et al.* [17], 12: Nahas *et al.* [18], 13: D'Anna *et al.* [19], 14: Ferrari [20], 15: Evans *et al.* [21], 16: Ye *et al.* [22]

interval, which can be explained by the following: 1) the evaluation of treatment efficiency was relatively subjective as the daily frequency of hot flashes was reported by the patients themselves. The accuracy was therefore not guaranteed with a large variation. The relative standard deviation (RSD) in most studies was in the range of 50 ~100%, 2) most clinical studies were of small sample size, precisely enrolling less than 60 subjects in each treatment group and 3) the time points for final observation varied by trial. Despite these fluctuations, the results obtained were

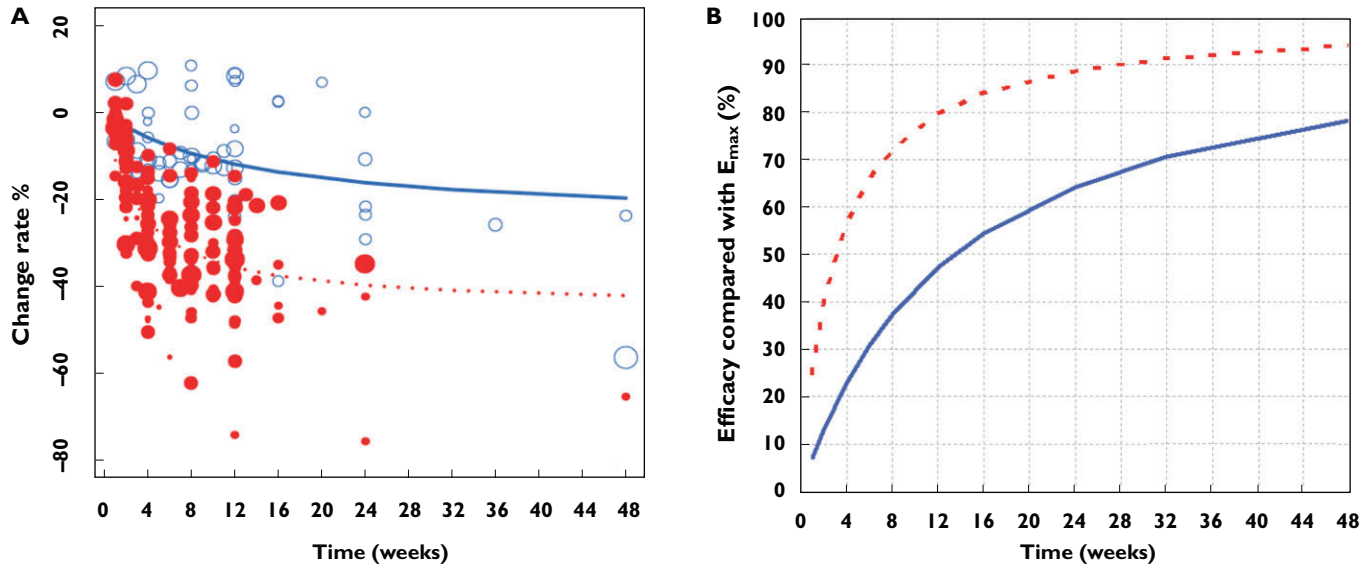

Figure 4

Typical predicted effect (lines) and observed data (points) of soy isoflavones and estradiol after elimination of the placebo effect (A). The corresponding efficacy ratio compared with the E_{max} value is presented in Figure 4B. - - -, estradiol; ——, soy isoflavones

analyzed in a combined manner in the meta-analysis, which neglected the effects of time on treatment efficiency. All these contributed to errors, which may affect the evaluation of treatment efficiency.

Unfortunately, these errors could not be distinguished using conventional meta-analysis. However, MBMA overcomes the disadvantages of conventional meta-analysis through detailed description of efficacy data using a mathematical model, as well as separation of inter-trial variation, inter-treatment arm variations and residual errors. Therefore, accurate evaluation of the efficiency of soy isoflavones was carried out using MBMA.

To eliminate the potential effects of baseline frequency on the evaluation of treatment efficiency, the evaluation index was defined as the descending ratio of frequency of hot flashes after different treatments compared with the baseline frequency. Significant placebo effects were observed with a maximal value of 35.8% in the soy isoflavones trial, and much higher in the estradiol trial (maximal value of 55.6%). This might be attributed to the fact that most subjects were aware that the effects of soy isoflavones were definitely not superior to those of estradiol, and the lower expectation in the effects of soy isoflavones resulted in the lower placebo effects observed in the soy isoflavone trials. After eliminating the placebo effect, the maximum effects of soy isoflavones was 25.2%, accounting for 57% of the maximum effects of estradiol ($E_{max\text{-}estradiol} = 44.9\%$). However, soy isoflavones need 13.4 weeks to achieve half of the peak activity, much longer than estradiol, which only requires 3.09 weeks. Therefore, after 12 weeks treatment, estradiol achieved 80% of its maximum effects, while the effects of soy isoflavones only reached 47% of maximum effects. Of note, 12 weeks is the recommended treatment interval in the FDA guide for estrogen and estrogen/progestin drug products for treatment of vasomotor symptoms [52]. This time frame is very reasonable for estradiol, but too short for soy isoflavones. After 48 weeks of treatment, the effects of soy isoflavones were just close to 80% of ceiling effects.

In the 16 reports included in this MBMA, the dose of isoflavones ranged from 30 to 200 mg day^{-1}. Meanwhile, the active components of various isoflavone preparations were different, most of which were isoflavone extracts of soybean containing genistein, diadzein, and glycitein. Genistein monotherapy was performed in three studies [15, 19, 21]. In our study, we attempted to adjust the parameter $E_{max\text{-}soy}$ by dosage of soy isoflavones. However, no significant correlation was observed between the inter-trial variations of dosage and $E_{max\text{-}soy}$. Finally, dose was not introduced in this model. In this study, we could not evaluate the efficacy of a single active component in treating hot flashes, as isoflavone components varied by study. Therefore, the large inter-trial variances observed among these studies may be partly induced by the dose and type of soy isoflavones.

In conclusion, this study quantified the efficacy of soy isoflavones in treating menopausal hot flashes for the first time. Important parameters, such as the maximum effect and time associated with an effect equal to 50% of E_{max}, were obtained by the MBMA. These results demonstrate that soy isoflavones have slight and slow effects in attenu-

ating the menopausal hot flashes, and provide a valuable reference for a rational use of soy isoflavones.

Competing Interests

All authors have completed the Unified Competing Interest form at http://www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author) and declare no support from any organization for the submitted work, no financial relationships with any organizations that might have an interest in the submitted work in the previous 3 years and no other relationships or activities that could appear to have influenced the submitted work.

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Supporting Information

Additional Supporting Information may be found in the online version of this article at the publisher's web-site:

Figure S1
Goodness-of-fit plots for the model. A) Population predicted effect data *vs.* observed effect data; B) individual predicted effect data *vs.* observed effect data; C) conditional weighted residuals *vs.* population predicted effect data; D) Density distribution of conditional weighted residuals. The black and red lines in A and B represent identity and regression lines, respectively, whereas in C the black line is the position where conditional weighted residual equals 0 and the red lines are the nonparametric regression lines. The red line in D is density distribution line

Figure S2
Prediction corrected visual predictive check of the final model. The dash lines are the 5th, 50th and 95th percentiles of observed data. The shaded areas are the corresponding confidence intervals of the simulated data. The solid points represent observed data, and the symbol size is proportional to the number of subjects in each study

Table S1
Summary of included placebo-controlled trials of estradiol