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Supplement: Equol, Soy, and Menopause

Equol, via Dietary Sources or Intestinal Production, May Ameliorate Estrogen Deficiency-Induced Bone Loss[1-3]

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Abstract

Equol, a product of intestinal metabolism of daidzein, is chemically similar to estrogen (without the lipophilic moiety) and has higher estrogen receptor-β binding affinity than its parent precursor. In 2004, a long-term, randomized controlled trial that characterized postmenopausal women by their equol-producing status showed stronger advantages to lumbar spine bone mineral density (BMD) in equol- compared with nonequol-producers. Subsequent studies have related equol status of participants to change in bone turnover markers or BMD in response to soy isoflavone interventions. To our knowledge, we are the first to prescreen women for equol-producing status prior to initiating an intervention. In menopausal Western women, equol status did not affect the modest, but significant, reduction in bone resorption achieved with a soy isoflavone intervention. J. Nutr. 140: 1377S–1379S, 2010.

Introduction

Dietary soy isoflavone supplements have been studied for their effect on ameliorating bone loss associated with menopause. However, studies of dietary equol as a potential dietary supplement are limited. The reported studies of oral and i.v. equol administration on bone in animal models used high doses to inhibit bone loss, which caused adverse effects on reproductive tissues. We conducted a dose-ranging study (0, 50, 100, and 200 mg equol/kg diet) of dietary racemic equol (50% R-equol, 50% S-equol) in 6-mo-old ovariectomized (OVX) Sprague Dawley rats. Doses were selected to achieve similar serum concentrations in postmenopausal women consuming medium and high amounts of isoflavones. We found that tissue (heart, intestine, kidney, and liver) equol concentrations reflected the diet, but only the highest dose studied (200 mg/kg) increased the femoral calcium content. Uterine, but not mammary gland, epithelial tissues were stimulated at this dose. These findings suggested limited benefit with the potential for adverse effects of equol as a dietary supplement.

Equol may be a bone antiresorption agent

Endogenous estrogen helps with maintaining bone mass and its deficiency with transition to menopause has been associated with rapid bone loss (1,2). When circulating levels decrease, bone resorption rates exceed bone formation rates, leading to accelerated bone loss for 3–5 y until bone formation rates catch up (3). Estrogen therapy was a mainstay of osteoporosis prevention in postmenopausal women until adverse cardiovascular and breast cancer health events were reported in women randomized to a conjugated estradiol treatment (premarin) in the Women's Health Initiative trial (4). Replacements for estrogen have been highly sought that have the antiresorption properties of estrogen therapy without the adverse effects on reproductive tissues.

Soy isoflavones have been studied the most for their potential to replace estrogen therapy. These studies have had mixed results (5–15). Equol has received much attention in the last 5 y. Equol, the end product of intestinal metabolism of daidzein, is chemically similar to estrogen (without the lipophilic moiety) and has 80 times more estrogen receptor-β binding affinity than its parent precursor (16). Estrogen receptor-β is dominant in bone and is presumed to be a primary mode of action of estrogen

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and phytoestrogens such as isoflavones (16). Other modes of action, including suppressing osteoclastogenesis, have also been proposed (14,15). Osteoclast formation was inhibited by equol in a dose-dependent manner (10–1000 nmol/L) in a coculture system of mouse bone marrow and primary osteoclast cells (17).

All rodents, but not all humans, have gut microflora that convert daidzein to equol. Many approximate that only 30–50% of the population possesses the gut microflora needed for bacteria intestinal conversion of daidzein to equol (18,19). Here, we explore equol as a dietary supplement on bone properties as well as the effect of equol-producing status on bone antiresorptive responsiveness to soy.

Dietary equol, calcium metabolism, and bone properties in estrogen deplete rodent models

Studies of dietary equol as a potential dietary supplement are limited. Early studies administered equol by i.v. injection or osmotic pump, because it was only recently made affordable by synthesis in adequate quantities for feeding studies. The reported studies of oral and i.v. equol administration on bone in animal models used high doses, which inhibited bone loss (20,21) but caused adverse effects on reproductive tissues (21). Plasma equol levels ranged from 1550 nmol/L (20) to 8433 nmol/L (21). Daily subcutaneous injections of 0.5 mg equol/d in mice (20) as well as oral dosing of 10 μg equol/(g body weight·d) (21) in rats maintained bone mineral density (BMD) of OVX animals to similar levels of SHAM rats. Consumption of 1000 mg/kg dietary equol led to increased uterine epithelial proliferation and plasma equol levels of 6–8 μmol/L (22) in mice. Dietary equol at 400 mg/kg diet attenuated OVX-induced trabecular bone loss at the lumbar spine but had a mild uterotropic activity, decreased weight gain at these concentrations, and circulating equol concentrations of 2460 nmol/L (23). Equol supplementation also improved biomechanical and histomorphometric measures of femurs in OVX rats (24) as well as aided in healing of osteoporotic fractures in OVX rats (25).

We conducted a dose-ranging study (0, 50, 100, and 200 mg equol/kg diet) of dietary racemic equol (50% R-equol, 50% S-equol) in 6-mo-old OVX Sprague Dawley rats. Doses were selected to achieve serum concentrations similar to those of postmenopausal women consuming medium (56 mg/d, 3–39 μmol/L) or high (90 mg/d, 5–49 μmol/L) amounts of isoflavones (26). We found that heart, intestine, kidney, and liver tissue equol concentrations reflected the diet, but only the highest dose studied (200 mg/kg) increased femoral calcium content (27). Calcium absorption and retention tended ($P = 0.07$) to be higher at equol concentrations >100 mg/kg. Mammary gland epithelial tissues were not stimulated, but uterine epithelial tissues were stimulated at this dose. These findings suggest limited benefit with the potential for adverse effects of equol as a dietary supplement.

New method for examining effect of soy phytoestrogens on bone

We have developed a novel method using the rare isotope, ^{41}Ca, to prelabel the skeleton and then monitor bone resorption by measuring urinary ^{41}Ca excretion by accelerator MS (28). The advantages of this method are its extreme sensitivity and specificity compared with traditional methods. We have used this method to study the effect of dose-ranging intervention of soy isoflavones in soy protein isolates (28) as well as to compare several botanical supplements with traditional therapies of osteoporosis (29). Response to the botanical supplements reflected their genistein content. The supplement with the highest genistein content (158 mg/d) was soy derived and suppressed bone resorption about one-third as well as either estradiol (1 mg/d) and mederoxyprogestrone (2.5 mg/d) or a bisphosphate (Actonel, 5 mg/d). We are using this method to determine the response to soy feeding in equol-producers compared with nonproducers and also to determine the effective dose in a dose-response study of purified genistein. This method has promise for determining drug-botanical interactions.

Effect of equol-producing status on response to soy

Recent evidence suggests that the effect of soy isoflavone treatment on health is dependent on the ability to convert daidzein to equol. Soy isoflavone supplementation has a stronger effect on the expression of estrogen responsive genes (genes with estrogen response elements in promoter regions) in equol producers compared with nonequol producers (30). A long-term, randomized controlled trial that characterized postmenopausal women by their equol-producing status showed stronger advantages to lumbar spine BMD after a 2-y intervention in equol (2.4% increase) compared with nonequol producers (0.6% increase) (8). Several subsequent studies have related equol status of subjects to change in bone turnover markers or BMD in response to an intervention containing soy isoflavones (19,31). Equol-producing ability determined by fecal equol levels in 68 of 122 postmenopausal Japanese women was associated with reduced total hip BMD after 24 wk of isoflavone treatment (31). Both urinary equol and O-desmethylangolensin levels indicating the presence of gut metabolizing microflora following 3 d of soy food consumption were related to BMD in 92 postmenopausal women (18). O-desmethylangolensin, but not equol, producers had greater total, leg, and head BMD compared with nonproducers ($P < 0.05$).

Equol metabolism in young adults

The form of equol provided for supplementation also plays a key role in metabolism and could alter potential health benefits of equol. Currently, there is only one pharmacokinetic study to our knowledge that examined the effect of S-equol, R-equol, and racemic equol on circulating equol levels in adults (32). A single oral dose of 20 mg of S-^{13}C-equol, R-^{13}C-equol, or racemic ^{13}C-equol was administered to healthy young men (n = 6) and women (n = 6) and plasma was collected for 48 h. Compared with other phytoestrogens (genistein and daidzein), equol was more bioavailable and rapidly absorbed. Differences in bioavailability and absorption were associated with the form of equol consumed. R-^{13}C-equol had higher bioavailability and fractional absorption than S-^{13}C-equol. However, both enantiomers had higher absorption, plasma concentrations, and bioavailability than the racemic mixture.

Future directions

Although evidence suggests that equol may attenuate estrogen deficiency-related bone loss, there are various factors that could influence the health benefit potential of equol, such as dose, form of equol, and the ability for intestinal production of equol. Other lifestyle traits could also affect the health protective actions of equol. For instance, our dose-response equol study revealed that equol had a detrimental effect on tibia calcium content that could be contributed to the estrogen-like effects during mechanical loading (27). Future research should determine the bone health effect of equol supplementation in physically active postmenopausal women. Overall, a deeper understanding of equol and interaction with lifestyle factors is needed to thoroughly assess the bone health benefit of equol.

Acknowledgments

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