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REVIEW

Does equol production determine soy endocrine effects?

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Abstract Isoflavones, a group of phytoestrogens, are selective oestrogen receptor (ER) modulators. They may positively impact endocrine-related conditions but the current evidence is sparse. Equol, a non-steroidal oestrogen, is produced by the metabolism of the isoflavone daidzein by intestinal bacteria. In Western countries, 30–50% of individuals metabolize daidzein into equol and are known as equol producers. Equol production may be the source of benefit from isoflavones in endocrine disease.

Keywords Equol · Phytoestrogen · Isoflavone · Soy · Cardiovascular disease · Osteoporosis · Breast cancer

Introduction

Epidemiological and clinical studies suggest that consumption of isoflavones, a group of phytoestrogens found predominately in soybeans and red clover, reduces the risk of endocrine-related conditions such as osteoporosis, cardiovascular diseases, menopausal symptoms and breast and prostate cancer [1–3]. However, evidence of benefit from intervention studies is not equivocal. Hypothetically, for the health benefits of soy isoflavones, daidzein is converted to equol by gut bacteria in certain individuals, it is the equol produced by these individuals that accounts for some health benefits of soy isoflavones [4]. This review explores equol's involvement in endocrine diseases.

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Phytoestrogens and their isoflavones

Phytoestrogens are plant-derived compounds that structurally and functionally resemble the mammalian oestrogen, 17 β-estradiol [2]. There are four classes of phytoestrogens: isoflavones, stilbenes, lignans and coumestans [2]. The isoflavones, in particular genistein and daidzein, are of interest due to their high concentrations in soy products and the purported health benefits [2, 4].

The molecular structure of phytoestrogens (and isoflavones) includes a phenolic ring that enables these compounds to bind to ERs, in a similar manner to, but with lower affinity than, 17 β-estradiol [5]. Isoflavones are recognized as selective ER modulators with both oestrogenic and anti-oestrogenic properties [2]. They can act as ER agonists in low-oestrogen environments and as ER antagonists in high-oestrogen environments [6]. Anti-androgenic properties of isoflavones, particularly genistein, have also been described [7].

Isoflavones, specifically genistein, have antiproliferative mechanisms; it has been shown that genistein inhibits the growth of breast cancer cells in vitro [8] and antioxidant properties, donating electrons via their hydroxyl groups and scavenging free radicals [9]. These properties, together with their ER modulator and anti-androgenic actions, support the suggestion that isoflavones positively modulate endocrine-related disease.

Metabolism of isoflavones and production of equol

Isoflavones can exist in two forms: glucosides and aglycones [2]. Glucosides (genistin and daidzin) are inactive, glucose-conjugated compounds that are poorly absorbed by the small intestine [6]. Once ingested, glucosides are hydrolysed in the proximal intestine into biologically active aglycones (genistein and daidzein), which are readily absorbed [10]. Daidzein can be further metabolized by specialized intestinal bacteria into equol and O-desmethylangolensin [6, 11, 12].

In vitro incubation of soy germ with faecal bacteria obtained from equol producers results in the production of equol [13], but no equol is seen in the urine of microorganism-free rats given soy [14]. Faecal inoculates from human equol non-producers did not produce equol from daidzein, whereas those from the equol producers did [15], but only when the incubations were carried out under anaerobic conditions. The effects of various antibiotics on the in vitro metabolism of daidzein also revealed interindividual differences. Some antibiotics inhibited the conversion of daidzein to dihydrodaidzein but not the conversion of dihydrodaidzein to equol, suggesting that multiple bacteria are responsible for these biotransformations [15].

Almost every animal species studied has the capacity to uniformly produce equol when fed isoflavone-containing soy products [16, 17]. However, in humans, there is marked interindividual variation, with only 30–50% of individuals in Western countries producing equol after consuming isoflavone-containing soy products [4, 16]. The percentage is higher in vegetarians [18] and Asian populations [19, 20]. Populations can therefore be divided into equol producers and non-equol producers [4]. An equol producer is defined as an individual with fasting serum levels of >20 nM (>5 ng/mL) or a 24-h urine concentration of >82 nM (>20 ng/mL) [18].

Factors that determine equol-producing status

In Japan, China and Korea, up to 80% of individuals are equol producers [21, 22] compared with 25% of individuals in North America and Europe (28). However, the percentage of equol producers in younger men in Japan and Korea is significantly lower than that of older men [22]. It has been suggested that younger generations are consuming few soy products as a result of Western influence which may lead to reduced levels of equol and to an increased risk of prostate cancer and other diseases of the ageing population in the years to come [22].

In a study which evaluated the relationship between equol-producing status and demographic, anthropometric, lifestyle and dietary factors among premenopausal women in the United States, 28% of the women were equol producers [23]. Hispanic and Latino women were more likely to be equol producers. A positive association between equol production and servings per day of vegetables and eggs was observed [23].

It is not clear at what age one becomes an equol producer. There is some evidence that exposure to soy early in life may be required if one is to become an equol producer [24]. In adults, consumption of isoflavones does not convert a non-producer of equol to a producer [25, 26].

It is possible that changes in the bacterial population as a result of diet, antibiotic use and other unknown factors may affect an individual's ability to be a consistent equol producer. Various bacteria have been isolated from faeces of animals which have shown to convert daidzein to equol [27–32]. There are also various Gram-positive, anaerobic, rod-shaped bacteria isolated from human faeces that convert daidzein or dihydrodaidzein to equol [33–44]. One strain of *Lactococci* not normally present in human faeces but found as a fish pathogen was fermented with soy germ such that 50% of the daidzin/daidzein was converted to equol. The resulting nutritional supplement in Japanese women has been shown to relieve menopausal symptoms in a placebo-controlled trial [45].

Equol versus isoflavones

Equol [7-hydroxy-3-(4'-hydroxyphenyl)-chroman] was first identified in the urine of pregnant mares in 1932 [46] and in human urine in the early 1980s [47]. Equol is not classified as a phytoestrogen as it is not derived from plants and is known as a non-steroidal oestrogen [4]. Unlike isoflavones, equol has a chiral centre and can exist as two distinct enantiomers, R-(+)equol and S-(-)equol [47]. In humans, the metabolism of daidzein to equol produces S-(-)equol exclusively and only this enantiomer has a preferential binding affinity to ER-β (comparable to that of genistein). R-(+)equol binds more weakly and has a preference for ER-α [48].

Equol is more bioavailable than daidzein or genistein [49]. Almost 50% of equol circulates in its free unbound form, compared with only 18.7% of daidzein [50, 51], allowing comparatively more equol to bind to available ERs. Equol clears from the plasma at a slower rate than daidzein [4], enabling it to act longer in the body. Unlike its precursor daidzein (but similar to genistein), equol has anti-androgenic properties and may have a positive impact on androgen-mediated pathologies, such as prostate cancer. In vivo experiments in rats demonstrated that equol binds with high affinity to 5α-dihydrotestosterone (DHT), blocking its binding to the androgen receptor (AR), causing reduction in both prostate and epididymal weights [52].

Equol has antioxidant [53] and antiproliferative properties [54], both of which are greater than those of its precursor, daidzein [53, 54]. Recently, equol demonstrated vasorelaxant [55] and anti-inflammatory properties [56], both previously shown in soy isoflavones [57, 58] implicating its protective role in cardiovascular diseases.

Equol production and endocrine-related conditions

Both S-(-)equol and the isoflavone genistein bind to ER-β with high affinity, preventing endogenous mammalian oestrogen from binding. They can act as either ER agonists or ER antagonists depending on environmental circumstances [6]. Numerous epidemiological and clinical studies have examined the involvement of isoflavones and equol in reproductive hormone-dependent conditions, including osteoporosis, cardiovascular diseases, menopause symptoms, and breast and prostate cancer [2, 3]. The findings have not been consistent [59–63] which may be linked to inter-individual differences in equol production [4] or be directly related to the presence of equol itself [45, 54, 63–65].

Osteoporosis

Oestrogen plays an important role in maintaining bone density; its decrease during menopause increases the risk of osteoporosis. It is known that 17β estradiol plays a critical role in the osteoblast to build bone. ERβ is expressed in both osteoblasts and osteoclasts [66]. Postmenopausal hormone replacement therapy (HRT) has been used as a preventive strategy for osteoporosis; however, its popularity has diminished due to associated risk factors, including breast cancer [67]. Isoflavones could potentially replace HRT in postmenopausal women and are marketed widely as such due to their oestrogen agonistic properties.

Isoflavone consumption is associated with bone loss prevention in pre- and postmenopausal women [68] and with increased bone mineral density (BMD), a marker of bone health [69]. A 2-year double-blind clinical trial by Wong et al. and a 3-year randomized controlled trial by Alekel et al. showed that daily supplementation with 120 mg of hypocotyl aglycone isoflavones (compared with a placebo) reduced whole-body bone loss and was protective for femoral neck BMD, but did not slow bone loss at common fracture sites [70, 71]. However, a recent meta-analysis suggested women may need to be equol producers to gain bone sparing benefit from isoflavone consumption [72]. Wu et al. studied the effect of supplementing 75 mg of isoflavones on BMD in one hundred and twenty-eight postmenopausal Japanese women over 24 weeks, demonstrating a significant improvement to BMD (per cent change) following isoflavone consumption in equol producers versus non-equol producers [73, 74]. Similarly, Frankenfeld et al. achieved a 20% reduction in spinal BMD in equol producers following a 3-day soy challenge in ninety-two postmenopausal women, when comparing non-equol producers and producers. The total and site-specific BMD, however, was not influenced by equol status [75].

Equol status has also been shown to be associated with reduced bone resorption. In postmenopausal Japanese women, dietary supplementation of 40 mg of isoflavones for 8 weeks significantly decreased the level of deoxypyridinoline, a marker of bone resorption, especially in equol producers compared with women not consuming isoflavones [76].

Equol production may influence bone remodelling. Equol producers show a higher level of serum osteocalcin (bone formation marker) and urine deoxypyridinoline (bone resorption marker) than non-equol producers, shown in premenopausal women consuming 120 mg of soy isoflavones daily over three menstrual cycles.

The importance of equol for bone protection is still controversial as some studies did not report benefit. Brink et al. did not observe preventive effects on postmenopausal related bone loss and bone turnover in healthy early postmenopausal women with a daily intake of 110 mg soy isoflavone aglycones over 1 year, compared with isoflavone free diets, even when equol phenotype was taken into account [59]. Similarly, daily consumption of 40 mg of isoflavones by postmenopausal Japanese women for 8 weeks had no influence on BMD in equol producers [76]. The variability of isoflavone types, doses and treatment times may contribute to these inconsistencies.

Cardiovascular disease

The risk in women of developing cardiovascular diseases during menopause is associated with elevated cholesterol concentration accompanying the loss of endogenous oestrogen secretion [77]. Oestrogen can affect the cardiovascular system directly by binding to ERs in vascular tissue and indirectly by altering the lipoprotein profile [2]. There is evidence that dietary soy isoflavone consumption may offer cardio-protective benefits by replacing the depleted oestrogen with isoflavones that bind to ERs. Postmenopausal women with a higher dietary intake of the isoflavone genistein had a lower BMI and waist circumference than those with no daily genistein consumption [78]. Soy isoflavone ingestion by males and females was associated with lower levels of total cholesterol and LDL cholesterol [79] and lower systolic blood pressures [80]. Conversely, several studies have not found a significant effect of isoflavones on lipoprotein, cholesterol and triglyceride

concentrations in postmenopausal women [61, 62, 77, 80] and premenopausal women [81]. The discrepancies seen between studies could be related to the production of equol.

Similar to isoflavones, equol production is associated with modifications to various risk factors of cardiovascular diseases. Patients with hypercholesterolaemia who were equol producers had lower total cholesterol, LDL cholesterol, LDL to HDL cholesterol ratio and plasma triglycerides after ingesting pasta enriched with 33 mg of isoflavone aglycones daily for 4 weeks [82]. Menopausal women subjected to dietary supplementation with 40 mg/day of isoflavones for 8 weeks and postmenopausal women consuming tibolone (HRT treatment) without soy supplement, both recorded lower blood pressure in equol producers compared with non-equol producers [76, 83]. Conversely, others did not find that equol excretion had significant effects on LDL cholesterol, HDL cholesterol or total cholesterol [84, 85].

Vasomotor symptoms

The menopausal symptoms of sweating, hot flashes and mood swings are linked to a decline in ovarian oestrogen production. These symptoms can be offset by HRT therapy. Dietary supplements containing isoflavones are widely used as alternatives to hormonal therapies for hot flashes [86]. A recent meta-analysis showed that phytoestrogen consumption by menopausal women was associated with reduction in hot flushes; however, conclusive results could not be established due to variation in type of soy supplement used in the studies [87]. The discrepancies between these studies could be related to the absence of equol or differing equol status of the subjects.

Daily consumption of 135 mg of isoflavones for 1 week by healthy menopausal women led to improved menopausal symptoms in equol producers but not in non-producers [88]. Similarly, daily supplementation with 40 mg isoflavones to menopausal Japanese women for 8 weeks significantly decreased hot flashes in equol producers compared with non-equol producers [76].

Several studies have examined the effect of supplementing equol to the diet of menopausal women. Daily consumption of 10 mg natural S-(-)equol by menopausal Japanese women for 12 weeks reduced the severity and frequency of hot flashes, decreased neck and shoulder stiffness and improved sweating, irritability and somatic symptoms [89]. Similarly, dietary supplementation of 10 mg of equol three times per week for 12 weeks in perimenopausal and postmenopausal Japanese women reduced menopause-related anxiety and depression scores [45].

To determine the selectivity of the genes regulated by S-equol, gene profiling of genes that contain oestrogen-response elements was carried out in lymphocytes isolated from postmenopausal women who were either equol producers or not [90]. In the equol producers, oestrogen-related receptor beta and four orphan nuclear receptors (BROB2, NR2C1, NR2F2 and NR113) were the most overexpressed genes which demonstrating status as an equol producer is associated with selective changes in gene expression [90].

Breast cancer

Epidemiological studies suggest that breast cancer rates are low in populations that consume isoflavone-rich soy products [89, 91–93]. Early exposure to isoflavones is hypothesized to be protective against breast cancer in later life [94, 95]. Higher endogenous oestrogen concentration is associated with a higher risk of breast cancer development. By competing with endogenous oestrogen for ERs, thereby reducing the bioavailability of endogenous oestrogen, isoflavones may hypothetically lower the risk of breast cancer in postmenopausal women [96]. Isoflavones may also increase the serum levels of sex hormone-binding globulin (SHBG) so decreasing the levels of free estradiol [96, 97]. A meta-analysis, however, did not show significant effects of isoflavone consumption on the circulating reproductive hormone concentrations (including oestrogen and SHBG) in pre- and postmenopausal women. In premenopausal women, who are equol producers, the reproductive hormone pattern is consistent with lower risk of breast cancer (independent of isoflavone dose intake) [98]. However, other studies did not replicate these findings [99, 100].

The other hypothesis for breast cancer protection by isoflavones and equol is that both may alter oestrogen metabolism away from the genotoxic oestrogen metabolites (increased risk) toward the protective metabolites (that decrease the risk of breast cancer) [101–105]. Xu et al. showed that a diet rich with isoflavones reduced the synthesis of genotoxic metabolites 16α-OH E1 and 4-hydroxyestrogens, increased the synthesis of the protective metabolite 2-OH E1, and lowered the ratio of 2-OH E1 to 16α-OH E1 (also known to be protective for breast cancer) in both pre- and postmenopausal women [101, 102].

There is evidence that isoflavones lower mammographic breast density [60, 106–109], (the amount of epithelial and stromal tissue in the female breast) which is a biomarker for breast cancer risk [110]. Dense tissue increases the risk of breast cancer four to six times [111], and density is negatively associated with the serum level of free oestradiol in pre- and postmenopausal women [110]. The effect of isoflavones and equol on mammographic density is not well understood. A recent meta-analysis showed that isoflavone intake did not alter mammographic breast density

in post-menopausal women, but induced a small increase in breast density in premenopausal women [112]. Two studies suggested equol production may be necessary to gain benefit from isoflavones on mammographic density. Post-menopausal women with a weekly intake of more than one soy food/supplement and those subjected to a 3-day soy challenge with 10 mg daidzein per day had lower mammographic density in equol producers compared with non-equol producers [107, 109]. Conversely, mammographic density did not change after 1 year of daily supplementation of 99 mg of soy protein containing isoflavones in elderly women even after accounting for equol status [60], and no association between daidzein-metabolizing phenotype and breast density was found in premenopausal women with low dietary soy consumption following a soy challenge [106].

Prostate cancer

Epidemiological studies suggest a correlation between dietary soy consumption and reduced risk of prostate cancer [113, 114], with prostate cancer incidence lower in Asian populations than in Western populations [115, 116]. Anti-androgenic properties of genistein and equol were demonstrated both in vitro and in vivo. Both inhibit 5α-reductase enzyme (which converts testosterone to DHT) in human genital skin fibroblasts [7], and consumption of soy protein isolate by men at high risk of prostate cancer suppresses the expression of the AR [117]. In men, isoflavones concentrate in prostatic tissue and fluid [54, 118]. Several studies concur that isoflavone ingestion is associated with a slower rise in prostatic specific antigen level [119–121].

The protective impact of isoflavones on the reproductive hormones involved in prostate cancer has been shown in animal studies. Rats exposed to a phytoestrogen-rich diet (40 mg/kg or 600 mcg/g of isoflavones) had significantly lower prostate weights and lower concentration of testosterone and andorostenedione, compared with rats fed a phytoestrogen free diet [122, 123]. In men, however, ingestion of isoflavones did not have a significant impact on testosterone, SHBG, free testosterone and free androgen index [124]. There is evidence that isoflavone consumption affects endogenous oestrogen levels (and its metabolites) in men [125, 126]. This is important since prostate cancer pathophysiology is associated with the andropause when there is a rise in the oestrogen to androgens ratio and oestrogen has been associated with carcinogenesis of the prostate [127]. Conversely, Kumar et al. did not demonstrate a significant effect on total oestrodial (among other hormones) following consumption of 80 mg of isoflavones by men with early stage prostate cancer for 12 weeks [128].

It has been hypothesized that the effect of soy isoflavone consumption on prostate cancer protection may be dependent on equol production [129]. Equol production correlates with a lower incidence of prostate cancer in Asian populations [20, 130, 131]. Equol alone has anti-androgenic properties [52] and anti-proliferative properties in the prostate, which may be different to those of isoflavones. Both properties have been demonstrated in vitro and in vivo in animal studies. Lund et al. administered 0.25 mg/kg of equol to male rats for 4–7 days and observed reduced prostate and epididymal weight and increased circulating levels of LH [52]. Equol's action was mediated by binding with high affinity to DHT [7, 52]. The rise in LH observed following administration of equol suggested that equol may block the negative feedback of DHT on the pituitary LH [52]. Equol's anti-proliferative effects in the prostate were demonstrated in vitro. Benign and malignant epithelial cells derived from prostatic fluid of Asian men who regularly consume soy were treated with a mixture of equol [at 10(-6)M and 10(-5)M] and variable concentrations of isoflavones genistein and daidzein. The results showed growth inhibition in the benign prostatic epithelial cells and increased accumulation of cells into Go/G1, the non-division stages of the cell cycle mechanisms [54]. Magee et al. demonstrated that both racemic [R-(+) equol and S-(-)] and S(-)equol mixtures at different concentrations inhibited the growth of human prostate and breast cancer cell lines and blocked their invasiveness. The racemic mixture [unlike S(-)equol] also prevented DNA damage [132]. The relevance of equol production to human prostate cancer needs to be studied in interventional dietary trials.

Conclusion

There is increasing evidence that equol may mediate the endocrine-related benefits associated with soy consumption. The inconsistencies between the soy isoflavone studies may be attributed to the failure to distinguish equol producers from non-equol producers. All the studies to date have measured total equol, i.e., conjugated plus unconjugated equol. Since only free, unconjugated S-equol binds to ERβ with high affinity, total equol does not provide an appropriate measure. S-equol's selectivity for ERβ may provide a better safety profile than those compounds that are selective for ERα. Studies that have considered equol production status are not consistent, the data are difficult to compare due to differing soy preparations and trial designs, together with small sample sizes. Positive effects of equol on several endocrine pathologies have been observed in both in vivo and in vitro studies, but currently it is difficult to say whether these effects are directly related to equol or to other factors. Well-designed clinical trials are needed to

determine whether equol is a potential therapeutic agent in reproductive hormone-dependent conditions.

References

1. Yamori Y (2006) Food factors for atherosclerosis prevention: Asian perspective derived from analyses of worldwide dietary biomarkers. Exp Clin Cardiol 11:94–98
2. Cornwell T, Cohick W, Raskin I (2004) Dietary phytoestrogens and health. Phytochemistry 65:995–1016
3. Cassidy A, Albertazzi P, Lise Nielsen I, Hall W, Williamson G, Tetens I, Atkins S, Cross H, Manios Y, Wolk A et al (2006) Critical review of health effects of soyabean phyto-oestrogens in post-menopausal women. Proc Nutr Soc 65:76–92
4. Setchell KD, Brown NM, Lydeking-Olsen E (2002) The clinical importance of the metabolite equol-a clue to the effectiveness of soy and its isoflavones. J Nutr 132:3577–3584
5. Kuiper GG, Lemmen JG, Carlsson B, Corton JC, Safe SH, van der Saag PT, van der Burg B, Gustafsson JA (1998) Interaction of estrogenic chemicals and phytoestrogens with estrogen receptor beta. Endocrinology 139:4252–4263
6. Izumi T, Piskula MK, Osawa S, Obata A, Tobe K, Saito M, Kataoka S, Kubota Y, Kikuchi M (2000) Soy isoflavone aglycones are absorbed faster and in higher amounts than their glucosides in humans. J Nutr 130:1695–1699
7. Evans BA, Griffiths K, Morton MS (1995) Inhibition of 5 alpha-reductase in genital skin fibroblasts and prostate tissue by dietary lignans and isoflavonoids. J Endocrinol 147:295–302
8. Zava DT, Duwe G (1997) Estrogenic and antiproliferative properties of genistein and other flavonoids in human breast cancer cells in vitro. Nutr Cancer 27:31–40
9. Mitchell JH, Gardner PT, McPhail DB, Morrice PC, Collins AR, Duthie GG (1998) Antioxidant efficacy of phytoestrogens in chemical and biological model systems. Arch Biochem Biophys 360:142–148
10. Day AJ, DuPont MS, Ridley S, Rhodes M, Rhodes MJ, Morgan MR, Williamson G (1998) Deglycosylation of flavonoid and isoflavonoid glycosides by human small intestine and liver beta-glucosidase activity. FEBS Lett 436:71–75
11. Heinonen S, Wahala K, Adlercreutz H (1999) Identification of isoflavone metabolites dihydrodaidzein, dihydrogenistein, 6'-OH-O-dma, and cis-4-OH-equol in human urine by gas chromatography-mass spectroscopy using authentic reference compounds. Anal Biochem 274:211–219
12. Chang YC, Nair MG (1995) Metabolism of daidzein and genistein by intestinal bacteria. J Nat Prod 58:1892–1896
13. Setchell KD, Borriello SP, Hulme P, Kirk DN, Axelson M (1984) Nonsteroidal estrogens of dietary origin: possible roles in hormone-dependent disease. Am J Clin Nutr 40:569–578
14. Axelson M, Setchell KD (1981) The excretion of lignans in rats—evidence for an intestinal bacterial source for this new group of compounds. FEBS Lett 123:337–342
15. Atkinson C, Berman S, Humbert O, Lampe JW (2004) In vitro incubation of human feces with daidzein and antibiotics suggests interindividual differences in the bacteria responsible for equol production. J Nutr 134:596–599
16. Atkinson C, Frankenfeld CL, Lampe JW (2005) Gut bacterial metabolism of the soy isoflavone daidzein: exploring the relevance to human health. Exp Biol Med (Maywood) 230:155–170
17. Setchell KD, Clerici C (2010) Equol: history, chemistry, and formation. J Nutr 140:1355S–1362S
18. Setchell KD, Cole SJ (2006) Method of defining equol-producer status and its frequency among vegetarians. J Nutr 136:2188–2193
19. Song KB, Atkinson C, Frankenfeld CL, Jokela T, Wahala K, Thomas WK, Lampe JW (2006) Prevalence of daidzein-metabolizing phenotypes differs between Caucasian and Korean American women and girls. J Nutr 136:1347–1351
20. Akaza H, Miyanaga N, Takashima N, Naito S, Hirao Y, Tsukamoto T, Fujioka T, Mori M, Kim WJ, Song JM, Pantuck AJ (2004) Comparisons of percent equol producers between prostate cancer patients and controls: case-controlled studies of isoflavones in Japanese, Korean and American residents. Jpn J Clin Oncol 34:86–89
21. Morton MS, Arisaka O, Miyake N, Morgan LD, Evans BA (2002) Phytoestrogen concentrations in serum from Japanese men and women over forty years of age. J Nutr 132:3168–3171
22. Fujimoto K, Tanaka M, Hirao Y, Nagata Y, Mori M, Miyanaga N, Akaza H, Kim WJ (2008) Age-stratified serum levels of isoflavones and proportion of equol producers in Japanese and Korean healthy men. Prostate Cancer Prostatic Dis 11:252–257
23. Atkinson C, Newton KM, Bowles EJ, Yong M, Lampe JW (2008) Demographic, anthropometric, and lifestyle factors and dietary intakes in relation to daidzein-metabolizing phenotypes among premenopausal women in the United States. Am J Clin Nutr 87:679–687
24. Hoey L, Rowland IR, Lloyd AS, Clarke DB, Wiseman H (2004) Influence of soya-based infant formula consumption on isoflavone and gut microflora metabolite concentrations in urine and on faecal microflora composition and metabolic activity in infants and children. Br J Nutr 91:607–616
25. Vedrine N, Mathey J, Morand C, Brandolini M, Davicco MJ, Guy L, Remesy C, Coxam V, Manach C (2006) One-month exposure to soy isoflavones did not induce the ability to produce equol in postmenopausal women. Eur J Clin Nutr 60:1039–1045
26. Mathey J, Lamothe V, Coxam V, Potier M, Sauvant P, Bennetau-Pelissero C (2006) Concentrations of isoflavones in plasma and urine of post-menopausal women chronically ingesting high quantities of soy isoflavones. J Pharm Biomed Anal 41:957–965
27. Minamida K, Tanaka M, Abe A, Sone T, Tomita F, Hara H, Asano K (2006) Production of equol from daidzein by gram-positive rod-shaped bacterium isolated from rat intestine. J Biosci Bioeng 102:247–250
28. Minamida K, Ota K, Nishimukai M, Tanaka M, Abe A, Sone T, Tomita F, Hara H, Asano K (2008) Asaccharobacter celatus gen. nov., sp. nov., isolated from rat caecum. Int J Syst Evol Microbiol 58:1238–1240
29. Yu ZT, Yao W, Zhu WY (2008) Isolation and identification of equol-producing bacterial strains from cultures of pig faeces. FEMS Microbiol Lett 282:73–80
30. Wang XL, Shin KH, Hur HG, Kim SI (2005) Enhanced biosynthesis of dihydrodaidzein and dihydrogenistein by a newly isolated bovine rumen anaerobic bacterium. J Biotechnol 115:261–269
31. Rafii F, Hotchkiss C, Heinze TM, Park M (2004) Metabolism of daidzein by intestinal bacteria from rhesus monkeys (Macaca mulatta). Comp Med 54:165–169
32. Matthies A, Clavel T, Gutschow M, Engst W, Haller D, Blaut M, Braune A (2008) Conversion of daidzein and genistein by an anaerobic bacterium newly isolated from the mouse intestine. Appl Environ Microbiol 74:4847–4852
33. Jin JS, Nishihata T, Kakiuchi N, Hattori M (2008) Biotransformation of C-glucosylisoflavone puerarin to estrogenic (3S)-equol in co-culture of two human intestinal bacteria. Biol Pharm Bull 31:1621–1625
34. Hur HG, Lay JO Jr, Beger RD, Freeman JP, Rafii F (2000) Isolation of human intestinal bacteria metabolizing the natural isoflavone glycosides daidzin and genistin. Arch Microbiol 174:422–428
35. Tamura M, Tsushida T, Shinohara K (2007) Isolation of an isoflavone-metabolizing, Clostridium-like bacterium, strain TM-40, from human faeces. Anaerobe 13:32–35

36. Maruo T, Sakamoto M, Ito C, Toda T, Benno Y (2008) Adlercreutzia equolifaciens gen. nov., sp. nov., an equol-producing bacterium isolated from human faeces, and emended description of the genus Eggerthella. Int J Syst Evol Microbiol 58:1221–1227
37. Wang XL, Kim HJ, Kang SI, Kim SI, Hur HG (2007) Production of phytoestrogen S-equol from daidzein in mixed culture of two anaerobic bacteria. Arch Microbiol 187:155–160
38. Kim M, Kim SI, Han J, Wang XL, Song DG, Kim SU (2009) Stereospecific biotransformation of dihydrodaidzein into (3S)-equol by the human intestinal bacterium Eggerthella strain Julong 732. Appl Environ Microbiol 75:3062–3068
39. Yokoyama S, Suzuki T (2008) Isolation and characterization of a novel equol-producing bacterium from human feces. Biosci Biotechnol Biochem 72:2660–2666
40. Matthies A, Blaut M, Braune A (2009) Isolation of a human intestinal bacterium capable of daidzein and genistein conversion. Appl Environ Microbiol 75:1740–1744
41. Jin JS, Kitahara M, Sakamoto M, Hattori M, Benno Y (2010) Slackia equolifaciens sp. nov., a human intestinal bacterium capable of producing equol. Int J Syst Evol Microbiol 60:1721–1724
42. Tsuji H, Moriyama K, Nomoto K, Miyanaga N, Akaza H (2010) Isolation and characterization of the equol-producing bacterium Slackia sp. strain NATTS. Arch Microbiol 192:279–287
43. Decroos K, Vanhemmens S, Cattoir S, Boon N, Verstraete W (2005) Isolation and characterisation of an equol-producing mixed microbial culture from a human faecal sample and its activity under gastrointestinal conditions. Arch Microbiol 183:45–55
44. Decroos K, Eeckhaut E, Possemiers S, Verstraete W (2006) Administration of equol-producing bacteria alters the equol production status in the Simulator of the Gastrointestinal Microbial Ecosystem (SHIME). J Nutr 136:946–952
45. Ishiwata N, Melby MK, Mizuno S, Watanabe S (2009) New equol supplement for relieving menopausal symptoms: randomized, placebo-controlled trial of Japanese women. Menopause 16:141–148
46. Marrian GF, Haslewood GA (1932) Equol, a new inactive phenol isolated from the ketohydroxyoestrin fraction of mares' urine. Biochem J 26:1227–1232
47. Setchell KD, Clerici C, Lephart ED, Cole SJ, Heenan C, Castellani D, Wolfe BE, Nechemias-Zimmer L, Brown NM, Lund TD et al (2005) S-equol, a potent ligand for estrogen receptor beta, is the exclusive enantiomeric form of the soy isoflavone metabolite produced by human intestinal bacterial flora. Am J Clin Nutr 81:1072–1079
48. Muthyala RS, Ju YH, Sheng S, Williams LD, Doerge DR, Katzenellenbogen BS, Helferich WG, Katzenellenbogen JA (2004) Equol, a natural estrogenic metabolite from soy isoflavones: convenient preparation and resolution of R- and S-equols and their differing binding and biological activity through estrogen receptors alpha and beta. Bioorg Med Chem 12:1559–1567
49. Setchell KD, Zhao X, Shoaf SE, Ragland K (2009) The pharmacokinetics of S-(-)equol administered as SE5-OH tablets to healthy postmenopausal women. J Nutr 139:2037–2043
50. Nagel SC, vom Saal FS, Welshons WV (1998) The effective free fraction of estradiol and xenoestrogens in human serum measured by whole cell uptake assays: physiology of delivery modifies estrogenic activity. Proc Soc Exp Biol Med 217:300–309
51. Fanti P, Sawaya BP, Custer LJ, Franke AA (1999) Serum levels and metabolic clearance of the isoflavones genistein and daidzein in hemodialysis patients. J Am Soc Nephrol 10:864–871
52. Lund TD, Munson DJ, Haldy ME, Setchell KD, Lephart ED, Handa RJ (2004) Equol is a novel anti-androgen that inhibits prostate growth and hormone feedback. Biol Reprod 70:1188–1195
53. Rimbach G, De Pascual-Teresa S, Ewins BA, Matsugo S, Uchida Y, Minihane AM, Turner R, VafeiAdou K, Weinberg PD (2003) Antioxidant and free radical scavenging activity of isoflavone metabolites. Xenobiotica 33:913–925
54. Hedlund TE, Johannes WU, Miller GJ (2003) Soy isoflavonoid equol modulates the growth of benign and malignant prostatic epithelial cells in vitro. Prostate 54:68–78
55. Jackman KA, Woodman OL, Chrissobolis S, Sobey CG (2007) Vasorelaxant and antioxidant activity of the isoflavone metabolite equol in carotid and cerebral arteries. Brain Res 1141:99–107
56. Blay M, Espinel AE, Delgado MA, Baiges I, Blade C, Arola L, Salvado J (2010) Isoflavone effect on gene expression profile and biomarkers of inflammation. J Pharm Biomed Anal 51:382–390
57. Hall WL, Formanuik NL, Harnpanich D, Cheung M, Talbot D, Chowienczyk PJ, Sanders TA (2008) A meal enriched with soy isoflavones increases nitric oxide-mediated vasodilation in healthy postmenopausal women. J Nutr 138:1288–1292
58. Chacko BK, Chandler RT, Mundhekar A, Khoo N, Pruitt HM, Kucik DF, Parks DA, Kevil CG, Barnes S, Patel RP (2005) Revealing anti-inflammatory mechanisms of soy isoflavones by flow: modulation of leukocyte-endothelial cell interactions. Am J Physiol Heart Circ Physiol 289:H908–H915
59. Brink E, Coxam V, Robins S, Wahala K, Cassidy A, Branca F (2008) Long-term consumption of isoflavone-enriched foods does not affect bone mineral density, bone metabolism, or hormonal status in early postmenopausal women: a randomized, double-blind, placebo controlled study. Am J Clin Nutr 87:761–770
60. Verheus M, van Gils CH, Kreijkamp-Kaspers S, Kok L, Peeters PH, Grobbee DE, van der Schouw YT (2008) Soy protein containing isoflavones and mammographic density in a randomized controlled trial in postmenopausal women. Cancer Epidemiol Biomarkers Prev 17:2632–2638
61. Hodgson JM, Puddey IB, Beilin LJ, Mori TA, Croft KD (1998) Supplementation with isoflavonoid phytoestrogens does not alter serum lipid concentrations: a randomized controlled trial in humans. J Nutr 128:728–732
62. Simons LA, von Konigsmark M, Simons J, Celermajer DS (2000) Phytoestrogens do not influence lipoprotein levels or endothelial function in healthy, postmenopausal women. Am J Cardiol 85:1297–1301
63. Nikander E, Kilkkinen A, Metsa-Heikkila M, Adlercreutz H, Pietinen P, Tiitinen A, Ylikorkala O (2003) A randomized placebo-controlled crossover trial with phytoestrogens in treatment of menopause in breast cancer patients. Obstet Gynecol 101:1213–1220
64. Sehmisch S, Uffenorde J, Maehlmeyer S, Tezval M, Jarry H, Stuermer KM, Stuermer EK (2010) Evaluation of bone quality and quantity in osteoporotic mice—the effects of genistein and equol. Phytomedicine 17:424–430
65. Sehmisch S, Erren M, Kolios L, Tezval M, Seidlova-Wuttke D, Wuttke W, Stuermer KM, Stuermer EK (2010) Effects of isoflavones equol and genistein on bone quality in a rat osteopenia model. Phytother Res 24(Suppl 2):S168–S174
66. Braidman IP, Hainey L, Batra G, Selby PL, Saunders PT, Hoyland JA (2001) Localization of estrogen receptor beta protein expression in adult human bone. J Bone Miner Res 16:214–220
67. Chlebowski RT, Anderson GL, Gass M, Lane DS, Aragaki AK, Kuller LH, Manson JE, Stefanick ML, Ockene J, Sarto GE et al

(2010) Estrogen plus progestin and breast cancer incidence and mortality in postmenopausal women. JAMA 304:1684–1692
68. Alekel DL, Germain AS, Peterson CT, Hanson KB, Stewart JW, Toda T (2000) Isoflavone-rich soy protein isolate attenuates bone loss in the lumbar spine of perimenopausal women. Am J Clin Nutr 72:844–852
69. Mei J, Yeung SS, Kung AW (2001) High dietary phytoestrogen intake is associated with higher bone mineral density in postmenopausal but not premenopausal women. J Clin Endocrinol Metab 86:5217–5221
70. Alekel DL, Van Loan MD, Koehler KJ, Hanson LN, Stewart JW, Hanson KB, Kurzer MS, Peterson CT (2010) The soy isoflavones for reducing bone loss (SIRBL) study: a 3-y randomized controlled trial in postmenopausal women. Am J Clin Nutr 91:218–230
71. Wong WW, Lewis RD, Steinberg FM, Murray MJ, Cramer MA, Amato P, Young RL, Barnes S, Ellis KJ, Shypailo RJ et al (2009) Soy isoflavone supplementation and bone mineral density in menopausal women: a 2-y multicenter clinical trial. Am J Clin Nutr 90:1433–1439
72. Ricci E, Cipriani S, Chiaffarino F, Malvezzi M, Parazzini F (2010) Soy isoflavones and bone mineral density in perimenopausal and postmenopausal Western women: a systematic review and meta-analysis of randomized controlled trials. J Womens Health (Larchmt) 19:1609–1617
73. Wu J, Oka J, Higuchi M, Tabata I, Toda T, Fujioka M, Fuku N, Teramoto T, Okuhira T, Ueno T et al (2006) Cooperative effects of isoflavones and exercise on bone and lipid metabolism in postmenopausal Japanese women: a randomized placebo-controlled trial. Metabolism 55:423–433
74. Wu J, Oka J, Ezaki J, Ohtomo T, Ueno T, Uchiyama S, Toda T, Uehara M, Ishimi Y (2007) Possible role of equol status in the effects of isoflavone on bone and fat mass in postmenopausal Japanese women: a double-blind, randomized, controlled trial. Menopause 14:866–874
75. Frankenfeld CL, McTiernan A, Thomas WK, LaCroix K, McVarish L, Holt VL, Schwartz SM, Lampe JW (2006) Postmenopausal bone mineral density in relation to soy isoflavone-metabolizing phenotypes. Maturitas 53:315–324
76. Uesugi S, Watanabe S, Ishiwata N, Uehara M, Ouchi K (2004) Effects of isoflavone supplements on bone metabolic markers and climacteric symptoms in Japanese women. BioFactors 22:221–228
77. Dewell A, Hollenbeck CB, Bruce B (2002) The effects of soy-derived phytoestrogens on serum lipids and lipoproteins in moderately hypercholesterolemic postmenopausal women. J Clin Endocrinol Metab 87:118–121
78. Goodman-Gruen D, Kritz-Silverstein D (2001) Usual dietary isoflavone intake is associated with cardiovascular disease risk factors in postmenopausal women. J Nutr 131:1202–1206
79. Taku K, Melby MK, Takebayashi J, Mizuno S, Ishimi Y, Omori T, Watanabe S (2010) Effect of soy isoflavone extract supplements on bone mineral density in menopausal women: meta-analysis of randomized controlled trials. Asia Pac J Clin Nutr 19:33–42
80. Howes JB, Sullivan D, Lai N, Nestel P, Pomeroy S, West L, Eden JA, Howes LG (2000) The effects of dietary supplementation with isoflavones from red clover on the lipoprotein profiles of post menopausal women with mild to moderate hypercholesterolaemia. Atherosclerosis 152:143–147
81. Samman S, Lyons Wall PM, Chan GS, Smith SJ, Petocz P (1999) The effect of supplementation with isoflavones on plasma lipids and oxidisability of low density lipoprotein in premenopausal women. Atherosclerosis 147:277–283
82. Clerici C, Setchell KD, Battezzati PM, Pirro M, Giuliano V, Asciutti S, Castellani D, Nardi E, Sabatino G, Orlandi S et al (2007) Pasta naturally enriched with isoflavone aglycons from soy germ reduces serum lipids and improves markers of cardiovascular risk. J Nutr 137:2270–2278
83. Tormala RM, Appt S, Clarkson TB, Tikkanen MJ, Ylikorkala O, Mikkola TS (2007) Individual differences in equol production capability modulate blood pressure in tibolone-treated postmenopausal women: lack of effect of soy supplementation. Climacteric 10:471–479
84. Hall WL, Vafeiadou K, Hallund J, Bugel S, Reimann M, Koebnick C, Zunft HJ, Ferrari M, Branca F, Dadd T et al (2006) Soy-isoflavone-enriched foods and markers of lipid and glucose metabolism in postmenopausal women: interactions with genotype and equol production. Am J Clin Nutr 83:592–600
85. Thorp AA, Howe PR, Mori TA, Coates AM, Buckley JD, Hodgson J, Mansour J, Meyer BJ (2008) Soy food consumption does not lower LDL cholesterol in either equol or nonequol producers. Am J Clin Nutr 88:298–304
86. Tice JA, Ettinger B, Ensrud K, Wallace R, Blackwell T, Cummings SR (2003) Phytoestrogen supplements for the treatment of hot flashes: the isoflavone clover extract (ICE) study: a randomized controlled trial. JAMA 290:207–214
87. Bolanos R, Del Castillo A, Francia J (2010) Soy isoflavones versus placebo in the treatment of climacteric vasomotor symptoms: systematic review and meta-analysis. Menopause 17:660–666
88. Jou HJ, Wu SC, Chang FW, Ling PY, Chu KS, Wu WH (2008) Effect of intestinal production of equol on menopausal symptoms in women treated with soy isoflavones. Int J Gynaecol Obstet 102:44–49
89. Aso T (2010) Equol improves menopausal symptoms in Japanese women. J Nutr 140:1386S–1389S
90. Niculescu MD, Pop EA, Fischer LM, Zeisel SH (2007) Dietary isoflavones differentially induce gene expression changes in lymphocytes from postmenopausal women who form equol as compared with those who do not. J Nutr Biochem 18:380–390
91. Wu AH, Wan P, Hankin J, Tseng CC, Yu MC, Pike MC (2002) Adolescent and adult soy intake and risk of breast cancer in Asian-Americans. Carcinogenesis 23:1491–1496
92. Wu AH, Yu MC, Tseng CC, Twaddle NC, Doerge DR (2004) Plasma isoflavone levels versus self-reported soy isoflavone levels in Asian-American women in Los Angeles County. Carcinogenesis 25:77–81
93. Ingram D, Sanders K, Kolybaba M, Lopez D (1997) Case-control study of phyto-oestrogens and breast cancer. Lancet 350:990–994
94. Nagata C (2010) Factors to consider in the association between soy isoflavone intake and breast cancer risk. J Epidemiol 20:83–89
95. Messina M, Hilakivi-Clarke L (2009) Early intake appears to be the key to the proposed protective effects of soy intake against breast cancer. Nutr Cancer 61:792–798
96. Kumar NB, Cantor A, Allen K, Riccardi D, Cox CE (2002) The specific role of isoflavones on estrogen metabolism in premenopausal women. Cancer 94:1166–1174
97. Pino AM, Valladares LE, Palma MA, Mancilla AM, Yanez M, Albala C (2000) Dietary isoflavones affect sex hormone-binding globulin levels in postmenopausal women. J Clin Endocrinol Metab 85:2797–2800
98. Duncan AM, Merz-Demlow BE, Xu X, Phipps WR, Kurzer MS (2000) Premenopausal equol excretors show plasma hormone profiles associated with lowered risk of breast cancer. Cancer Epidemiol Biomarkers Prev 9:581–586
99. Nettleton JA, Greany KA, Thomas W, Wangen KE, Adlercreutz H, Kurzer MS (2005) Short-term soy and probiotic supplementation does not markedly affect concentrations of reproductive hormones in postmenopausal women with and without

histories of breast cancer. J Altern Complement Med 11:1067–1074
100. Bonorden MJ, Greany KA, Wangen KE, Phipps WR, Feirtag J, Adlercreutz H, Kurzer MS (2004) Consumption of Lactobacillus acidophilus and Bifidobacterium longum do not alter urinary equol excretion and plasma reproductive hormones in premenopausal women. Eur J Clin Nutr 58:1635–1642
101. Wangen KE, Duncan AM, Merz-Demlow BE, Xu X, Marcus R, Phipps WR, Kurzer MS (2000) Effects of soy isoflavones on markers of bone turnover in premenopausal and postmenopausal women. J Clin Endocrinol Metab 85:3043–3048
102. Xu X, Duncan AM, Merz BE, Kurzer MS (1998) Effects of soy isoflavones on estrogen and phytoestrogen metabolism in premenopausal women. Cancer Epidemiol Biomarkers Prev 7:1101–1108
103. Atkinson C, Skor HE, Dawn Fitzgibbons E, Scholes D, Chen C, Wahala K, Schwartz SM, Lampe JW (2003) Urinary equol excretion in relation to 2-hydroxyestrone and 16alpha-hydroxyestrone concentrations: an observational study of young to middle-aged women. J Steroid Biochem Mol Biol 86:71–77
104. Nettleton JA, Greany KA, Thomas W, Wangen KE, Adlercreutz H, Kurzer MS (2005) The effect of soy consumption on the urinary 2:16-hydroxyestrone ratio in postmenopausal women depends on equol production status but is not influenced by probiotic consumption. J Nutr 135:603–608
105. Frankenfeld CL, McTiernan A, Tworoger SS, Atkinson C, Thomas WK, Stanczyk FZ, Marcovina SM, Weigle DS, Weiss NS, Holt VL et al (2004) Serum steroid hormones, sex hormone-binding globulin concentrations, and urinary hydroxylated estrogen metabolites in post-menopausal women in relation to daidzein-metabolizing phenotypes. J Steroid Biochem Mol Biol 88:399–408
106. Atkinson C, Newton KM, Aiello Bowles EJ, Lehman CD, Stanczyk FZ, Westerlind KC, Li L, Lampe JW (2009) Daidzein-metabolizing phenotypes in relation to mammographic breast density among premenopausal women in the United States. Breast Cancer Res Treat 116:587–594
107. Fuhrman BJ, Teter BE, Barba M, Byrne C, Cavalleri A, Grant BJ, Horvath PJ, Morelli D, Venturelli E, Muti PC (2008) Equol status modifies the association of soy intake and mammographic density in a sample of postmenopausal women. Cancer Epidemiol Biomarkers Prev 17:33–42
108. Maskarinec G, Williams AE, Carlin L (2003) Mammographic densities in a one-year isoflavone intervention. Eur J Cancer Prev 12:165–169
109. Frankenfeld CL, McTiernan A, Aiello EJ, Thomas WK, LaCroix K, Schramm J, Schwartz SM, Holt VL, Lampe JW (2004) Mammographic density in relation to daidzein-metabolizing phenotypes in overweight, postmenopausal women. Cancer Epidemiol Biomarkers Prev 13:1156–1162
110. Boyd NF, Stone J, Martin LJ, Jong R, Fishell E, Yaffe M, Hammond G, Minkin S (2002) The association of breast mitogens with mammographic densities. Br J Cancer 87:876–882
111. Boyd NF, Lockwood GA, Byng JW, Tritchler DL, Yaffe MJ (1998) Mammographic densities and breast cancer risk. Cancer Epidemiol Biomarkers Prev 7:1133–1144
112. Hooper L, Madhavan G, Tice JA, Leinster SJ, Cassidy A (2010) Effects of isoflavones on breast density in pre- and post-menopausal women: a systematic review and meta-analysis of randomized controlled trials. Hum Reprod Update 16:745–760
113. Nagata Y, Sonoda T, Mori M, Miyanaga N, Okumura K, Goto K, Naito S, Fujimoto K, Hirao Y, Takahashi A et al (2007) Dietary isoflavones may protect against prostate cancer in Japanese men. J Nutr 137:1974–1979
114. Yan L, Spitznagel EL (2009) Soy consumption and prostate cancer risk in men: a revisit of a meta-analysis. Am J Clin Nutr 89:1155–1163
115. Jian L (2009) Soy, isoflavones, and prostate cancer. Mol Nutr Food Res 53:217–226
116. Ozasa K, Nakao M, Watanabe Y, Hayashi K, Miki T, Mikami K, Mori M, Sakauchi F, Washio M, Ito Y et al (2004) Serum phytoestrogens and prostate cancer risk in a nested case-control study among Japanese men. Cancer Sci 95:65–71
117. Hamilton-Reeves JM, Rebello SA, Thomas W, Slaton JW, Kurzer MS (2007) Isoflavone-rich soy protein isolate suppresses androgen receptor expression without altering estrogen receptor-beta expression or serum hormonal profiles in men at high risk of prostate cancer. J Nutr 137:1769–1775
118. Rannikko A, Petas A, Rannikko S, Adlercreutz H (2006) Plasma and prostate phytoestrogen concentrations in prostate cancer patients after oral phytoestogen supplementation. Prostate 66:82–87
119. Pendleton JM, Tan WW, Anai S, Chang M, Hou W, Shiverick KT, Rosser CJ (2008) Phase II trial of isoflavone in prostate-specific antigen recurrent prostate cancer after previous local therapy. BMC Cancer 8:132
120. Hussain M, Banerjee M, Sarkar FH, Djuric Z, Pollak MN, Doerge D, Fontana J, Chinni S, Davis J, Forman J et al (2003) Soy isoflavones in the treatment of prostate cancer. Nutr Cancer 47:111–117
121. Kumar NB, Cantor A, Allen K, Riccardi D, Besterman-Dahan K, Seigne J, Helal M, Salup R, Pow-Sang J (2004) The specific role of isoflavones in reducing prostate cancer risk. Prostate 59:141–147
122. Ohno S, Nakajima Y, Inoue K, Nakazawa H, Nakajin S (2003) Genistein administration decreases serum corticosterone and testosterone levels in rats. Life Sci 74:733–742
123. Weber KS, Setchell KD, Stocco DM, Lephart ED (2001) Dietary soy-phytoestrogens decrease testosterone levels and prostate weight without altering LH, prostate 5alpha-reductase or testicular steroidogenic acute regulatory peptide levels in adult male Sprague-Dawley rats. J Endocrinol 170:591–599
124. Hamilton-Reeves JM, Vazquez G, Duval SJ, Phipps WR, Kurzer MS, Messina MJ (2010) Clinical studies show no effects of soy protein or isoflavones on reproductive hormones in men: results of a meta-analysis. Fertil Steril 94:997–1007
125. Nagata C, Takatsuka N, Shimizu H, Hayashi H, Akamatsu T, Murase K (2001) Effect of soymilk consumption on serum estrogen and androgen concentrations in Japanese men. Cancer Epidemiol Biomarkers Prev 10:179–184
126. Hamilton-Reeves JM, Rebello SA, Thomas W, Slaton JW, Kurzer MS (2007) Soy protein isolate increases urinary estrogens and the ratio of 2:16alpha-hydroxyestrone in men at high risk of prostate cancer. J Nutr 137:2258–2263
127. Farnsworth WE (1996) Roles of estrogen and SHBG in prostate physiology. Prostate 28:17–23
128. Kumar NB, Krischer JP, Allen K, Riccardi D, Besterman-Dahan K, Salup R, Kang L, Xu P, Pow-Sang J (2007) A Phase II randomized, placebo-controlled clinical trial of purified isoflavones in modulating steroid hormones in men diagnosed with localized prostate cancer. Nutr Cancer 59:163–168
129. Tanaka M, Fujimoto K, Chihara Y, Torimoto K, Yoneda T, Tanaka N, Hirayama A, Miyanaga N, Akaza H, Hirao Y (2009) Isoflavone supplements stimulated the production of serum equol and decreased the serum dihydrotestosterone levels in healthy male volunteers. Prostate Cancer Prostatic Dis 12:247–252
130. Akaza H, Miyanaga N, Takashima N, Naito S, Hirao Y, Tsukamoto T, Mori M (2002) Is daidzein non-metabolizer a high

risk for prostate cancer? A case-controlled study of serum soybean isoflavone concentration. Jpn J Clin Oncol 32:296–300
131. Kurahashi N, Iwasaki M, Inoue M, Sasazuki S, Tsugane S (2008) Plasma isoflavones and subsequent risk of prostate cancer in a nested case-control study: the Japan Public Health Center. J Clin Oncol 26:5923–5929
132. Magee PJ, Raschke M, Steiner C, Duffin JG, Pool-Zobel BL, Jokela T, Wahala K, Rowland IR (2006) Equol: a comparison of the effects of the racemic compound with that of the purified S-enantiomer on the growth, invasion, and DNA integrity of breast and prostate cells in vitro. Nutr Cancer 54:232–242