ROLES OF DIETARY PHYTOESTROGENS IN VARIOUS HUMAN DISORDERS

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Summary

Phytoestrogens are estrogenic compounds which occur naturally in many plants and fungi and which are biologically active in humans and animals. But the dietary availability of phytoestrogens from these sources is small. However, since epidemiological studies indicated that a high consumption of soy-based or phytoestrogen containing foods may decrease the risk of certain hormone-dependent cancers (e.g. breast and prostate), of osteoporosis and to reduce menopausal symptoms, cognitive functions. This review examines the evidence for a possible role of dietary phytoestrogen. Thus phytoestrogen rich foods or purified isolated compounds (genistein, daidzein, biochanin A, formononetin, matairesinol, secoisolariciresinol and coumestrol,) have been used as dietary supplements and health claims are made for these compounds, which are primarily evidence for beneficial effects based on epidemiological findings. There is increasing evidences that the health beneficial effects of phytoestrogens like isoflavonoids and lignans have been postulated as breast cancer, menopausal disorders, atherosclerosis, various cardiovascular diseases, Alzheimer's disease, anxiety and various other physiological conditions. Conflicting and controversial data exist about the functional effects of high-dosed phytoestrogens. Well designed studies investigating dose-response relationships are generally lacking and for specific cell or organ functions, it is still not clear at which concentrations phytoestrogens exert anti- or proestrogenic activity. Considering all these uncertainties and the lack of evidence from well designed and conducted studies it is not justified to recommend the regular consumption of high-dosed dietary phytoestrogen supplements. For the development of functional foods it is essential to perform human dietary intervention studies and to use well defined and validated biomarkers to assess the potential health benefits of dietary phytoestrogens. The present review throw light on the facts and deficiency about role phytoestrogens in various human disorders

Keywords: Phytoestrogens, Isoflavons, lignans, Diabetes, osteophoresis, Breast cancer, cardiovascular disordes, cognitive fuctions.

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Introduction

Phytoestrogens are estrogenic compounds which occur naturally in many plants and fungi and which are biologically active in humans and animals. These compounds can be defined as plant substances that are structurally or functionally similar to estradiol. They consist of a number of classes, mainly isoflavones, lignans, and coumestans (1–3). Soy products and legumes are the main sources of isoflavonoid phytoestrogens, such as genistein and daidzein. Genistein and daidzein have estrogenic, antiestrogenic, and anticarcinogenic activity. Lignans, such as enterolactone and enterodiol, are mostly found in linseed, whole cereals, legumes, and berries and, to a lesser extent, in other fruit and vegetables (4–6). Limited quantitative data are available for the concentrations of isoflavones and lignans in plant foods (7–9).

In recent years, phytoestrogens have attracted increased attention among the public and in the medical community. Accumulated evidence from a large body of literature suggesting that consumption of plant-based foods rich in these phytochemicals may benefit human health. Substantial data from epidemiologic surveys and nutritional intervention studies in humans and animals suggest that dietary phytoestrogens have protective effect against menopausal symptoms and a variety of disorders, including cardiovascular disease, cancer, hyperlipidemia, osteoporosis, and various forms of chronic renal disease (10–14). Phytoestrogens may have beneficial as well as adverse health effects. The ability of phytoestrogens to interrupt reproductive function is well established in a number of species (15,16). Several studies in humans and animals have shown that soy protein reduces plasma total cholesterol and LDL cholesterol. Evidence is also emerging that consumption or supplementation in food which is rich in phytoestrogens may have a beneficial effect on diabetes mellitus and obesity in animals and humans. Common phytoestrogen sources include soybeans, soy products, alfalfa fodder, flaxseed, **(Table no.1)** and over-the-counter dietary supplements. This review summarizes the evidences available regarding obesity and diabetes, Menopausal symptoms, the bone-altering effects, cognitive functions in humans and animals, it is aimed at providing snapshot of present staus of phytoestrogens and their potential applications in therapy, and other uses.

Biochemistry of Phytoestrogen

Phytoestrogens are a group of biologically active plant substances with a chemical structure that is similar to that of estradiol, an endogenous estrogen. **(Figure. 1)** The structural similarity accounts for the ability of these compounds to bind to estrogen receptors(ER) in various cells and exert estrogenic or antiestrogenic effects (17–21). Due to their abundance and (anti)-estrogenic potencies, the soy-derived isoflavones, coumestrol, resveratrol, would appear to have the potential for effectively functioning as endocrine disruptors(22).Single plant normally contains more than one class of phytoestrogens. The three major classes of phytoestrogens are isoflavones, lignans, and coumestans. The major bioactive isoflavones are genistein and daidzein, which are derived from the precursors biochanin A and formononetin, respectively due the action of intestinal glucosidases. Daidzein is further metabolized to equol **(Figure 2)** and O-dismethylangiolensin (O-DMA).

Genistein and diadzein having higher affinity to ER alpha than ER Beta, ER alpha are mainly found in bladder, uterus ovary, prostate, brain, lungs and testis. Lignans are constituents of many plants and form the building blocks for the formation of lignin in the plant cell wall (24). They are more common in the plant kingdom than are isoflavones. The two major lignans, enterolactone and enterodiol, are produced from matairesionol and secoisolariciresinol, respectively. Coumestrol is the most important form of coumestan consumed in the form of foodstuff for human.

Recently, number of studies reported various phytoconstituensts like 8- prenylnargenin isolated from hops *Humulus Lupulus* (25); ginsenoid Rg1 isolated from *panax notoginseng*; 8-phenenylnarginin and resveratrol isolated from grapes (26); lindleyin isolated from *Rhei rhizima*.(27) These phytoestrogens are stated to have estrogenic receptor modulating activity hence these constituents can be used in various human disease conditions.

Table: 1. Various sources of phytoestrogens and its active molecules

Source	Class	Phytoestrogen molecule
Soy Peanut Sunflower seeds Walnut	**Isoflavones**	Biochanin A, Genistein, Genistin, Diadzein, Diadzin, Formononetin.
Flax seeds Cereals Grains Fruit Vegetables	**Lignans**	Secoisolariciresinol, Matairesionol, Enterodiol, Enterolactone.
Soy sprout Alfalfa sprout Clover	**Coumestans**	Coumestrol

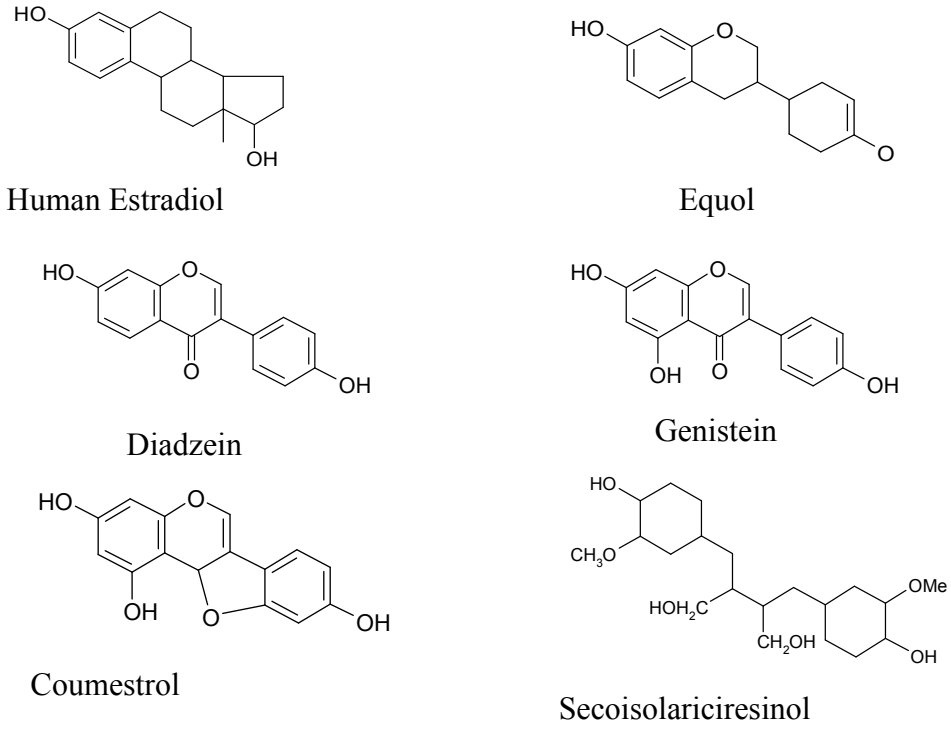

Figure 1. Structural similarity between the human estradiol and phytoestrogens.

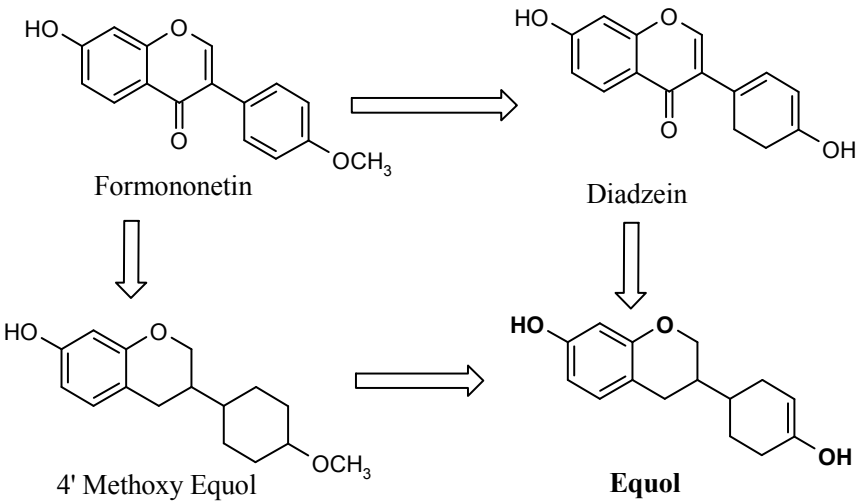

Figure 2. Metabolism of Equol

Phytoestrogens in Diabetes and Obesity

Metabolism of glucose and lipids is a complex process highly regulated by both peptides and steroid hormones and is influenced by diet. Obesity and diabetes mellitus have become major public health concern in industrialized countries. It is not only because of increasing prevalence in epidemic proportions but also because of their frequent association with major cardiovascular risk factors like dyslipidemia, atherosclerosis, and coronary artery disease, which are responsible for excess morbidity and mortality. Obesity is a disorder of energy balance and is associated with hyperinsulinemia, insulin resistance, and abnormalities in lipid metabolism.

The results of a very recent study (Lei Guan *et al,* 2006), showed that the ovariectomized hamsters orally given soybean and kudzu phytoestrogen extracts had significantly decreased serum total cholesterol (TC) and non-high-density lipoprotein cholesterol (non-HDL-C) with HDL cholesterol (HDL-C) being unaffected. It was concluded that both soybean and kudzu phytoestrogens could modify favorably lipoprotein profiles in ovariectomized hamsters. Many other studies also reported effect of phytoestrogens in diabetes and obesity (28). Tsai *et al* reported soy polysaccharides reduce postprandial blood glucose and serum triacylglycerol in obese and type 2 diabetes persons. (29) Vedavanam *et al* suggested that soy isoflavones may be beneficial for diabetic subjects because of their estrogenic activity and their ability to prevent glucose-induced lipid peroxidation and inhibit intestinal glucose uptake by decreasing sodium-dependent glucose transporter, which results in a reduction in postprandial hyperglycemia. (30) In another study Nogowski *et al* studied the effect of coumestrol on carbohydrate metabolism in ovariectomized rats. Coumestrol had no significant effect on plasma insulin or glucagon concentrations, but it decreased muscle glycogen and inhibited insulin binding to muscle membrane. Thus, the effect of coumestrol on carbohydrate metabolism appears to be via changes in insulin receptors. Whether these actions of phytoestrogens on skeletal muscle have an effect on overall glucose disposal in vivo is not known (31). Persaud *et al* suggested that, in isolated rat adipocytes, genistein inhibited de novo lipid synthesis from acetate and glucose but stimulated lipolysis (32). Thus, genistein appears to have direct effects on lipid metabolism in liver and adipose tissue by affecting both lipogenesis and lipolysis. In skeletal muscle cells, genistein was recently shown to inhibit glucose uptake stimulated by uncoupling protein (33). Coumestrol also affects lipid metabolism. In chicks, dietary coumestrol decreased plasma cholesterol concentrations in a dose-dependent manner (34). Szkudelska K *et al* reported that, in cultured islets of Langerhans, genistein (at a concentration of 100 mol/L) was shown to increase basal insulin secretion, but this dose of genistein also reduced islet cell proliferation. In additional studies, genistein was shown to inhibit islet tyrosine kinase activities and glucose- and sulfonylurea-stimulated insulin release without affecting glucose metabolism (35). Thus, phytoestrogens appear to have favorable biological actions on glucose and lipid metabolism that may explain their potential to benefit obesity and diabetes.

Phytoestrogens in Symptoms of Menopause and Breast Cancer

Various studies regarding action of phytoestrogens in pre or post menopausal symptoms were carried out and concluded the application of phytoestrogens. A descriptive study by Adlercreutz *et al*, reported in 1982, was the first to detect lower urinary excretion of enterolactone, enterodiol, and equol in postmenopausal breast cancer patients compared with healthy postmenopausal omnivorous and vegetarian American women (36). In two recent case-control studies (37), urinary levels of isoflavones (genistein, daidzein, equol, and glycitein) and lignans (enterolactone, enterodiol, and matairesinole) were used as an exposure index instead of dietary questionnaires. Both studies used a mixed pre- and postmenopausal population, and both observed an inverse association between higher urinary excretion of certain isoflavones and lignans and breast cancer risk (36). However, in these retrospective case-control studies, urinary phytoestrogen values were measured in breast cancer patients after diagnosis and, therefore, might have been influenced by metabolic consequences of the disease rather than be its possible causes.

Several other studies addressed the issue of intake of isoflavones in premenopausal women. Their main findings [soy protein daily intake significantly increased follicular phase length, suppressed luteinizing hormone and follicular stimulating hormone mid-cycle surges, and delayed menstruation by 1–5 days , as well as reduced circulating ovarian steroids and adrenal androgens] suggested that dietary estrogens may play a role in protecting women against breast cancer in several ways: (*a*) increased menstrual cycle length and thus reduced lifetime exposure to estrogens; (*b*) increased follicular phase length which eventually results in a reduced lifetime exposure to luteal phase in which mitotic rate of breast tissue is almost 4-fold greater (compared with the follicular phase) and, therefore, the risk for a carcinogenic transformation is greater; (*c*) an antiestrogenic effect of reduced proliferation of breast epithelial cells caused by phytoestrogens; and (*d*) decreased circulating ovarian steroid levels caused by phytoestrogens (37, 38, 39). Postmenopausal women apparently respond differently to a soy-supplemented diet. Measured luteinizing hormone, follicular stimulating hormone, and endogenous estrogen levels were not significantly changed by it (40). So phytoestrogens might have a protective role in premenopausal but not in postmenopausal women. It should also be noted that, despite the previously detected inverse association between soy product exposure and breast cancer risk, phytoestrogens are weak estrogens and might stimulate cell proliferation and estrogen-dependent gene expression (41-45).

Phytoestrogens and Cardiovascular System

Relation of menopause with cardiovascular diseases is well known and attribute to the protective role of estrogen in atherosclerosis. Raines *et al* studied In Vitro, Genistein inhibit the proliferation of many cells and inhibit angiogenesis which is an important process in atherosclerosis (46). As previously discussed, ER alpha and beta are expressed in vascular endothelial, smooth muscles cell and myocardial cells. Zhu *et al* suggested that there was abnormal vascular function and hypertension in mice deficient with ER beta. This result can be concluded as, the ER beta specific ligands might act as a cardio-vascular protective (47). Reports of various clinical trials stated that consuming 20-25 mg/day of soy protein is safe and effective in the reduction of cholesterol by 4 to 8 %

(48). In a recent study, Vander Schow *et al* (49) reported that consumption of higher than the usual dietary intake of phytoestrogens have protective effect in the atherosclerosis and arterial degeneration. Recently in 2003, Zhang *et al* (50) reported strong evidence that soy diet might reduce the risk of coronary heart disease in women. Beneficial and associated effects of phytoestrogens related to cardiovascular diseases need to be studied in detail.

Phytoestrogens and Osteoporosis

In the recent study by Zhang J *et al*,(2004) phytoestrogens could increase the bone mineral density (BMD) level, serum estradiol and inorganic phosphorus content, and uterine index, tartrate-resistant acid phosphatase (TRAP), alkaline phosphatuse ALP) and interleukin-6 levels. Pathological examinations showed that in the model group, the cortex of bone thinned, bone trabecula thinned also, with poor integrity, distorted, broken and decreased in size, while in the Phytoestrogens groups, the above-mentioned changes were significantly alleviated. Phytoestrogens are effective in preventing and treating osteoporosis in ovariectomized rats (51). In another study Lee YB *et al* (52) reported that supplementation with isoflavone prevented the losses of bone density and mineral content caused by ovariectomization. Although both isoflavone and 17beta-estradiol exhibited similar bone-sparing ability on the ovariectomize -induced bone loss, the effect of isoflavone was not the same as that of 17beta-estradiol on the serum ALP and TRAP, body weight increase, and uterine weight change. They concluded that dietary supplementation with soybean isoflavone can prevent postmenopausal bone loss via a different mechanism of estrogen in ovariectomized rats. Though these studies stated the effectiveness of phytoestrogens in animals and humans, there is still a problem of inconsistency in positive results which exists for menopausal symptoms and breast cancer. In light of better established drug products, phytoestrogens should not be recommended at this time as first-line therapy for prevention or treatment of osteoporosis in men or women because questions regarding the product selection and the degree of efficacy and safety (53) remain unanswered. Now it's a need of time to target the key research and continue to provide important patient education regarding the therapeutic uses and safety of phytoestrogens for osteoporosis.

Phytoestrogens and Cognitive Function

Epidemiological data from retrospective and case-control studies have indicated that estrogen replacement therapy (ERT) can decrease the risk of developing Alzheimer's disease. In addition, ERT has been found to promote cellular correlates of memory and to promote neuronal survival both in vivo and in vitro. Phytoestrogens have been proposed as potential alternatives to ERT (54). Various investigations of neuroprotective and neurotrophic efficacy of phytoestrogens were conducted which report that six phytoestrogens, genistein, genistin, daidzein, daidzin, formononetin, and equol, were tested for their neuroprotective efficacy against two toxic insults, glutamate excitotoxicity and ß-amyloid. Neuronal membrane damage was quantitatively measured by lactate dehydrogenase (LDH) release, and neuronal mitochondrial viability was determined by 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyl tetrazolium bromid (MTT) assay.

Results of these studies demonstrated that all phytoestrogens induced a modest but significant reduction in LDH release following exposure to glutamate and ß-amyloid. In contrast, none of phytoestrogens induced a significant increase in reduced MTT levels, which occurred in the presence of a full estrogen agonist, 17ß-estradiol (55-64). Lephart ED *et al* (65) reported the major results indicate that consumption of dietary phytoestrogens resulting in very high plasma isoflavone levels (in many cases over a relatively short interval of consumption in adulthood) can significantly alter sexually dimorphic brain regions, anxiety, learning and memory. The findings of these studies identify the biological actions of phytoestrogens, specifically isoflavones and their metabolites, found in animal soy-containing diets. Effect on brain and behavior implicate the importance of phytoestrogens as the recognized significance of estrogens in brain and neural disorders, such as Alzheimer's disease, especially in women. On the other hand, set of studies indicating that high concentrations of genistein caused cytotoxicity and DNA ladder formation in primary cultures of cortical neurons and chronic administration of genistein at high doses may induce cytotoxicity and apoptosis in the rat brain (66). Kreijkamp-Kaspers S *et al* reported that they carried out double-blind randomized trial which does not support the hypothesis that the use of soy protein supplement containing isoflavones improves cognitive function, bone mineral density, or plasma lipids in healthy postmenopausal women when started at the age of 60 years or later (67). In a control study it reported that long-term consumption of dietary antioxidants and phytoestrogens such as genistein, daidzein, biochanin A, formononetin, matairesinol, secoisolariciresinol and coumestrol, may reduce the risk of adult glioma(68). The results of these investigations suggest merits and demerits of phytoestrogens in cognitive function. According to these reports, it should be considered preliminary and need to be verified in larger, in deep and prospective studies.

Conclusion

According to the present reviewed data it is suggested that phytoestrogen rich diet can have beneficial effects on many aspects of diabetes and obesity. The dietary components responsible for the beneficial effects have yet to be determined. Definitive studies should shed more light in this area. Phytoestrogens plays a vital role for reduction in insulin resistance and adiposity by inhibiting insulin secretion from the pancreatic cells. Isoflavones and lignans improve glucose tolerance and insulin sensitivity. The exact mechanism of action is yet not clear, though there are some evidences of in vivo or vitro reports made. Long-term controlled trials on the safety and effectiveness of phytoestrogen on the development and progression of diabetes and obesity and their complications in patients with diabetes mellitus and obesity are overdue.

Several studies addressed the uses of isoflavones in premenopausal women where in it significantly increased the follicular phase length, suppressed luteinizing hormone and follicular stimulating hormone mid-cycle surges, and delayed menstruation by 1–5 days. It is also noticed that dietary estrogens may play a role in protecting women against breast cancer in several ways

In the cardiovascular disorders, phytoestrogens inhibit cell proliferation and avoid atherosclerosis as well as act as cardiovascular protective by providing ER ligand. In various studies phytoestrogens play a key role in association of coronary heart diseases.

Phytoestrogens could increase the the bone mineral density (BMD) level, serum estradiol and inorganic phosphorus content, and uterine index, tartrate-resistant acid phosphatase (TRAP), alkaline phosphatase (ALP). Several animal studies reveal that these compounds can prevent postmenopausal bone loss and osteophorosis.

There is controversy in the reports of phytoestrogens for its use in CNS disorders where in most of preclinical or clinical studies suggested that it improves the cognitive conditions in Alzhemiers disease and has benifecial effects on dimorphic brain regions, anxiety, learning and memory. However some other randomized studies reported that phytoestrogens do not improve cognitive function, bone mineral density, or plasma lipids in healthy postmenopausal women. Though phytoestrogens have been proved to have conventional therapeutic or beneficial effect, a detailed indepth study from basic etiology to epidemiologic studies are required to pave way for authenticating the efficacy and safety of phytoestrogens.

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