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Assessing Compositional Differences in Soy Products and Impacts on Health Claims

Joyce Boye and Sabine Ribéreau
*Food Research and Development Centre
Agriculture and Agri-Food Canada
Canada*

1. Introduction

In October 1999, the Food and Drug Administration (USA) approved a health claim for soy indicating that consumption of 25 grams of soy protein a day as part of a diet low in saturated fats and cholesterol may help lower the risk of heart disease. Additionally, soy has officially been approved in the UK, Brazil, Malaysia, Japan, Korea, Philippines, Indonesia and South Africa.

Some countries have, however, been slower to approve a health claim and are requiring more substantive evidence. Questions of concern include the impact of processing on the efficacy of soy protein, information on the normal and acceptable levels of isoflavones in soy products, efficacy of soy proteins in which isoflavones have been removed versus soy proteins containing isoflavones in respect to relationship with cardiovascular disease risk reduction, feasibility of consumers to take in 25 g of soy protein a day, effect of antinutritional factors (e.g., trypsin inhibitors, phytates, lectins, lysinoalanine), heat resistance of trypsin inhibitors found in soybeans and the specific effects of soy protein alone compared to soy protein within a matrix (e.g., whole soybean foods and foods made from soy protein ingredients).

These questions are pertinent as there is a wide variety of soyfoods available on the market with different macro and micro nutrient compositions. Answers to these questions will therefore be useful in identifying the specific conditions required for soyfoods to carry a health claim.

Foods prepared from whole soybeans include soymilk and tofu. Soybean derived ingredients include defatted soy flour, soy protein concentrate, soy protein isolate and soy fibre. Soy protein isolates in particular have been very successful commercially and are used extensively today in the production of a large number of foods. Different techniques can be used for making soy protein isolate and the type of processing treatment used can affect the nutrient and physicochemical composition of the isolate (e.g., composition of residual isoflavones, saponins, trypsin inhibitors, phytic acid and minerals). Isoflavones, saponins, trypsin inhibitors and phytic acid are all biologically active molecules and as such variations in their composition can hinder the ability to determine if a reported health benefit is due to the soy protein alone or the presence of these other compounds.

The chapter will provide a brief update on some of the reported health benefits of soy components. An overview of some of the major soyfoods and soy ingredients currently

available on the market will be provided along with a detailed list comparing the effects of processing on the composition of different soy products. Requirements for obtaining health claims using the Canadian example, as well as impacts of compositional differences on potential health benefits are further presented.

2. Soybeans and health

Cardiovascular disease (CVD) is one of the leading causes of death in the world today. According to the World Health Organization by 2030, almost 23.6 million people will die from CVDs, mainly from heart disease and stroke (www.who.int/mediacentre). Elevated low density lipoprotein (LDL) is a major cardiovascular disease risk factor. LDL transports cholesterol and triglycerides from the liver to peripheral tissues and arteries and regulates cholesterol synthesis. Retention of cholesterol in arteries can result in the formation of arterial plaques which increases the risk of atherosclerosis, peripheral vascular disease, strokes and heart attacks. Foods that decrease LDL levels will, therefore, increasingly be of interest as health foods. Several reports have shown a decrease in LDL levels and an increase in HDL after consumption of soy (Nilavsen & Meinertz, 1998; Potter et al., 1998; Merritt, 2004; Zhuo et al., 2004; Sacks et al., 2006; Harlanda & Haffnerb, 2008; Taku et al., 2008). Earlier studies showing similar results formed the basis of the current soy health claim in many jurisdictions.

Due to apparent inconsistencies in findings, in 2008 the Weston Price Foundation submitted a petition to the FDA in response to the FDA's request for public comment on the issue claiming that soy protein products are not safe and have no long history of use in the food supply. The organization also claimed the evidence on soy protein and heart disease was contradictory and inconsistent, and that no standard of scientific agreement had been met (<http://www.physorg.com/news122663958.html>).

A systematic study conducted by the US department of Health & Human Services indicated that while the evidence was weak for other disease outcomes, there is a suggestion of a possible dose-response effect for soy protein for LDL reduction (<http://www.ahrq.gov/Clinic/epcsums/soysum.htm>).

Using predictive equations along with a meta-analysis to determine whether the heart health claim for soy continues to be justified, Jenkins et al. (2010) concluded that low density lipoprotein cholesterol (LDL-C) reduction attributable to the combined intrinsic and extrinsic effects of soy protein foods ranged from 7.9 to 10.3%. They further concluded from their study that soy remains one of a few food components that reduces serum cholesterol (>4%) when added to the diet.

In a more recent study, Onuegbu et al. (2011) fed 500 mL of soymilk daily to 42 apparently healthy young to middle-aged subjects for a period of 21 days and reported that soymilk consumption significantly reduced mean plasma TC by 11% and LDL-C by 25% and increased mean plasma HDL-C by 20%. The authors also concluded that soy drink could be an important non-pharmacological cholesterol-reducing agent.

Furthermore, Bruckert and Rosenbaum (2011) have also recently reported LDL-cholesterol reduction ranging from -3 to -10% for soy protein and have indicated that dietary recommendations may have important impacts on cardiovascular events as they can be implemented early in life and because the sum of the effect on LDL-cholesterol is far from being negligible.

Another major risk factor for developing CVD is elevated blood pressure (EBP). A major contributor to EBP is Angiotensin II which is a potent vasoconstrictor. Vasoconstriction

occurs when renin, an enzyme produced in the kidneys, proteolytically acts on circulating angiotensinogen and converts it to angiotensin I (a decapeptide). In the presence of angiotensin converting enzyme (ACE), angiotensin I is cleaved to the octapeptide, angiotensin II resulting in arterial constriction and EBP. ACE also breaks down bradykinin, a vasodilator, further contributing to the elevation in blood pressure. Inhibition of ACE is, therefore, important for the lowering of blood pressure as this results in a decrease in the concentration of angiotensin II and an increase in the levels of bradykinin (Yang et al., 1970; Erdos, 1975). Various reports have suggested that peptides from soy possess ACE-inhibitory properties (Wu & Ding, 2002; Kuba et al., 2003; Chiang et al., 2006; Hartmann & Meisel, 2007; Yang et al., 2008). These peptides are usually not active when present within the sequence of parent proteins, but are released by enzymatic proteolysis *in vivo* or *in vitro*.

In addition to soy proteins, some reports have attributed the beneficial health effect of soy to phytochemicals found in soybeans such as isoflavones which are naturally occurring non-steroid compounds with weak estrogenic effects and chemical structure similar to estradiol-17 β (Fig. 1). Isoflavones are able to bind to estrogen receptors and are capable of triggering estrogen dependent responses physiologically. The main types of isoflavones found in soy are the aglycones (daidzein, genistein, and glycitein), the β -glucosides (daidzin, genistin, and glycitin) and their 6''-O-malonyl- β -glucosides (6OMalGlc) and 6''-O-acetyl- β -glucosides (6OAcGlc) conjugates.

Taku et al. (2010) conducted a meta-analysis to clarify the effects of soy isoflavone extracts on systolic and diastolic blood pressure (SBP and DBP) in adult humans. They reported that soy isoflavone extracts significantly decreased SBP but not DBP in adult humans, however, no dose-response relationship was observed.

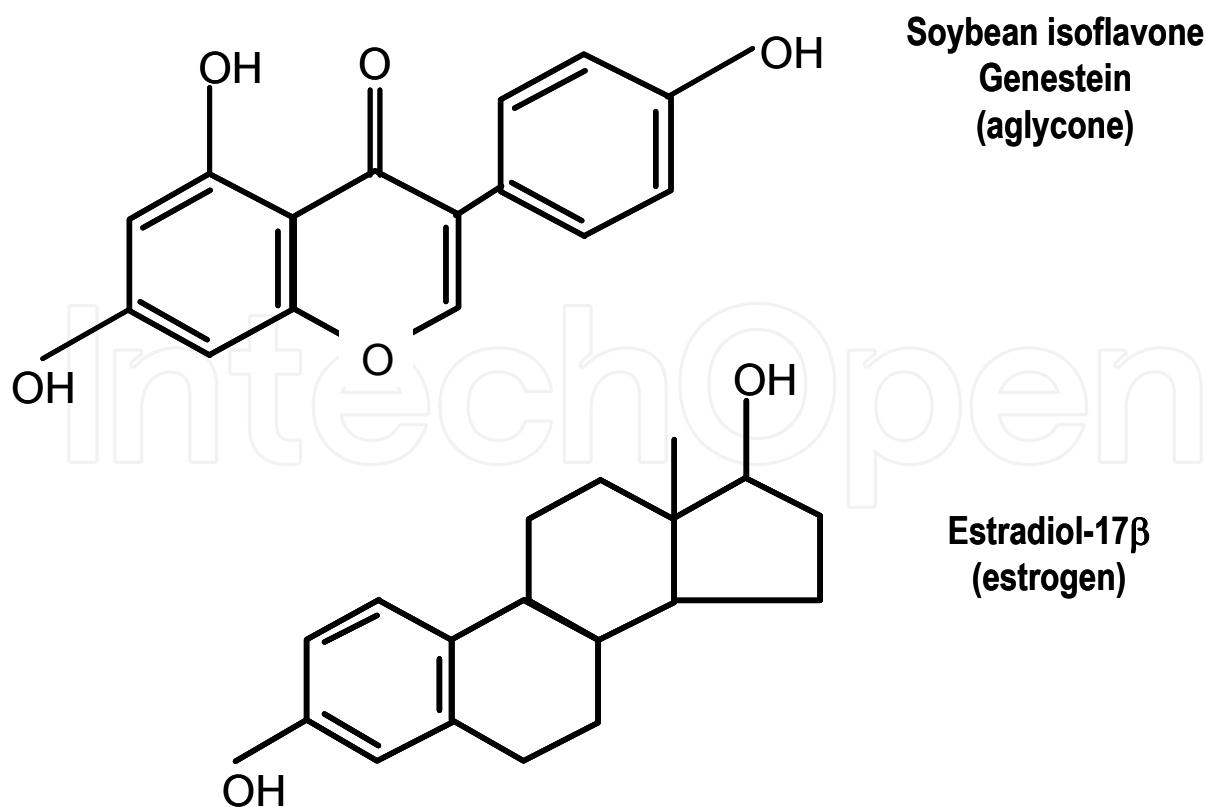


Fig. 1. Molecular structure of soy isoflavone genistein compared to estradiol-17 β .

In other studies, high dietary intakes of soy isoflavones were found to be associated with lower risk of recurrence among postmenopausal patients with breast cancer positive for estrogen and progesterone receptor and those who were receiving anastrozole as endocrine therapy (Kang et al., 2010a, b).

Ogborn et al. (2010) have also recently reported that a soy diet ameliorated renal injury in rats. Soy diets preserved normal renal function and reduced relative renal weight, scores for cystic change, fibrosis, tissue oxidized LDL content, inflammation and epithelial cell proliferation. In this study though, alcohol-extracted slow isoflavone soy protein was found to retain its major protective effects and only subtle differences were attributed to isoflavones.

Consumption of tofu containing high levels of isoflavones reportedly exerted positive effects on verbal memory, although not in older men and women, where no or negative effects of these compounds on brain cells and cognition was observed (Hogervorst et al., 2011).

In regards to other health outcomes, Messina (2010) reported that although recent clinical data have not supported the skeletal benefits of isoflavones, 2 large prospective epidemiologic studies found soy intake to be associated with marked reductions in fracture risk. Additionally, soybean isoflavones modestly alleviate hot flashes in menopausal women.

In addition to isoflavones, soy saponins may also exert bioactive effects. Orally administered commercial purified soy saponin at 80 mg/kg body weight/day to spontaneously hypertensive rats for 8 weeks significantly decreased blood pressure (Hiwatashi et al., 2010). In another study on the effect of soy saponins on the growth of human colon cancer cells, Tsai et al. (2010) reported that intake of soy saponin decreased the number of viable cells in a dose-dependent manner. They concluded that soy saponin may be effective in preventing colon cancer by affecting cell morphology, cell proliferation enzymes, and cell growth.

Additionally, phytic acid which is considered to be an antinutritional component in soybean may possess antioxidant effects (Sakač et al., 2010). Recent research studies further suggest that lunasin, lectins, and trypsin inhibitors may have beneficial health properties. Trypsin inhibitors in soy have been of particular concern because, if not destroyed by heat prior to consumption, they can cause pancreatic hypertrophy/hyperplasia, which ultimately results in growth inhibition (Liener, 1994,1996).

The majority of approved health claim for soy covers only soy proteins as evidence surrounding the effects of isoflavones and other bioactive compounds in soy are more controversial. Overall, although many studies suggest that the beneficial properties of soy may be attributed to the protein fraction, questions remain about potential synergistic or complimentary effects of other soy components.

In the sections below, an attempt will be made to provide a review of some of the different soy products available and their compositional differences and how this could impact their health properties.

3. Commercially available soyfoods and ingredients

Soybean has today become one of the world's most economical and valuable agricultural commodities due to its unique composition. On a wet basis, soybeans contain about 35% protein, 17% oil, 31% carbohydrate and 4.4% ash. The composition varies for different varieties and some cultivars can be found with protein contents of up to 50%. Soy proteins are nutritionally superior among vegetable proteins. Soy protein isolate has a Protein

Digestibility Corrected Amino Acid Score (PDCAAS) of 100% which means that it has all the essential amino acids required to support growth and maintenance. It contains good supplies of essential amino acids, such as lysine, which are normally lacking in other cereals. Fatty acids in soybeans consist of unsaturated fats, such as oleic, linoleic and linolenic acids, which are nutritionally beneficial. Additionally, soybeans also contain fibre and other phytochemicals, such as isoflavones and saponins which may have health benefits.

3.1 Major soyfoods

3.1.1 Soybeans and sprouts

Green soybean and soybean sprouts are two whole soyfoods prepared from soybean seeds. Sprouts are obtained by germinating soybeans for 5-10 days. They may be consumed fresh (e.g., in salads) or used as a vegetable in cooking. Green vegetable soybean, on the other hand, is harvested just before maturity (edamame) and can be cooked and eaten in salads and in soups or as a snack. It is available fresh (in pod or shelled), canned or frozen. The composition of these two products although prepared from the whole seed will vary due to the germination process applied to sprouts.

3.1.2 Soymilk

Soymilk is the liquid extract obtained after cooking, grinding and filtering soybean. It is not a “whole soyfood” per se as the majority of the fibre fraction (okara) is removed during processing. The soymilk extract obtained after filtration has a consistency that is very similar to cow’s milk and is frequently used as an alternative to dairy products. There are four major types of soymilk products available (unsweetened, sweetened, flavoured and low fat). Unsweetened soymilk generally contains only water and soybeans. Sweetened soymilk may be sweetened with rice syrup, honey, corn or barley malt extract. Flavoured soymilk may be sweetened or unsweetened, and is often flavoured with cocoa, vanilla, carob or strawberry. Low fat soymilk may also be sweetened or unsweetened, flavoured or unflavoured, but usually contains less fat. Soymilk is frequently fortified with vitamins and minerals to increase its nutrient value. Blends of soymilk made with soybeans and different cereals or fruits are also available as well as “functional soymilk products” (e.g., with added omega 3 or other functional ingredients). Some manufacturers process soymilk using soy protein isolate rather than starting with the bean.

3.1.3 Tofu

Tofu is a curd made from heated soymilk. It is prepared by adding coagulating agents such as glucono- δ -lactone (GDL) or salts (magnesium chloride, calcium chloride, calcium sulphate) to heated soymilk followed by pressing to remove the whey. The final product is a gel with different textures and degrees of hardness depending on the type and amount of coagulant used and processing method (Fig.2). As with soymilk, tofu is not a “whole food” as the fibre is removed in the process of making the soymilk. The composition is also different from the starting soymilk as much of the whey is removed to concentrate the proteins and facilitate gel formation during pressing. Tofu has a soft white texture which is in some respects similar to cheese. On a wet basis, pressed tofu with a moisture content of about 85% contains 7.8% protein, 4.2% lipid, and 2 mg/g calcium (Wang et al., 1983; Liu, 1997). It is important to mention that in addition to compositional differences due to the type of salts used, health outcomes may also vary due to differences in protein digestibility

resulting from the type of network structure induced by the specific salt used in tofuf-making.

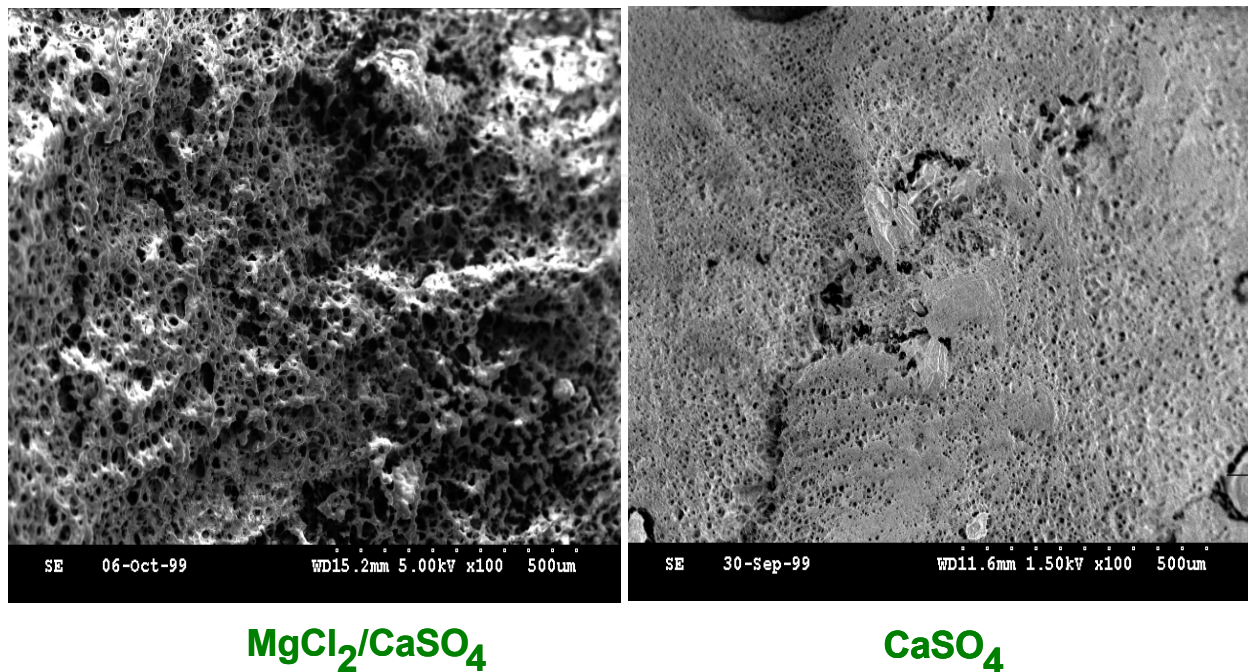


Fig. 2. Microstructure of tofu prepared using different salts.

3.1.4 Other fermented products from soybean

Soy sauce and soy paste (miso) are fermented soy products that are frequently used as condiments and seasoning in foods. Soy sauce and miso are made by fermenting soybean with or without other grains (e.g., wheat, rice, barley) with different types of *Aspergillus*.

Soy sauce is obtained in a liquid form whereas miso is a thick paste. These products are traditionally used in Asian cuisine but have become a mainstay of many modern diets. Tempeh and natto are two other fermented soyfoods but these are less frequently consumed outside of Asia. Tempeh is made by fermenting dehulled and cooked soybeans with *Rhizopus* whereas natto is fermented with *Bacillus subtilis*. The composition of these products will again vary depending on the processing conditions used and the amount of soybean present in the finished product. Furthermore, microorganisms used during fermentation can hydrolyse some phytochemicals such as isoflavones making them more bioactive.

3.1.5 Soy dips/dressings

There is a wide variety of dips/salad dressings etc. made from soybeans, soy flour, tofu, or soy protein isolates available on the market. They contain varying amounts of soy and their composition will similarly vary depending on the other ingredients used in the formulation.

3.1.6 Other soyfoods

The liquid extract during the preparation of soymilk can be further processed into a variety of refrigerated and frozen desserts such as ice cream, soy mousse, and fermented products such as soy yoghurt, soy probiotic beverage, and soy cheese, using processes similar to those

used in the dairy industry. The composition of these products will vary as a function of the ingredients used in processing and the fermentation process including the type of bacteria used during fermentation.

Soybeans can also be roasted in a manner similar to peanuts. The product obtained has a nutlike flavour and a crunchy texture which can be consumed as a snack. As these are made from intact whole bean, the composition will be similar to that of the starting raw material, however, the roasting process can induce changes which may modify digestibility. Roasted soybeans can also be ground to obtain roasted soy flour or roasted soynut butter.

3.2 Major soy ingredients

3.2.1 Soy oil

Today soybean oil is one of the world's leading vegetable oil for human consumption. Soybean oil is extracted from the bean after dehulling and flaking using organic solvents. The extracted oil is downstream processed to obtain a refined oil. Soybean oil is widely used in the manufacture of different foods. It is also frequently used as a salad or cooking oil and in the production of shortening, margarines, mayonnaise and salad dressings. By-products from the processing of soybean oil are also used to produce mono- and diglycerides and lecithin which are commonly used as emulsifying agents in foods.

3.2.2 Soy flour, soy protein concentrates, isolates and hydrolysates

Extraction of oil from soybeans leaves behind the soy meal biomass. Significant effort has been made in the last few decades to process this meal into value-added products. Defatted soy flakes (or flour, grits, meal), soy protein concentrates (SPC) and soy protein isolates (SPI) are the three major products available from soy meal. Additionally a full fat or partially defatted meal or flour can be obtained from the whole soybean.

Defatted soy flakes contain approximately 50% protein while, SPC and SPI generally contain at least 65% and 90% protein on a dry basis, respectively. Full-fat soy flour may be steamed and toasted to inactivate enzymes and enzyme inhibitors or unheated. Unheated, undefatted (or defatted) soy flour gives an enzyme active full-fat soy flour which is used in the bakery industry to bleach flour. Mechanically defatted soy meals with different fat contents are also available and usually sold in the organic category. Micronized soy flour (heat treated with infrared to eliminate anti-nutritional components in soy) are also available as whole beans or ground.

Soy products are widely used in formulated foods, partly because of their nutritional value but especially for the functional properties of the protein, which include gelling, foaming and emulsification which underlie many food sensory attributes. Other food products likely to contain soy ingredients include beverages, nutritional bars, bakery and cereal products, soups, meat products, beverages, confectionery, salad dressings and desserts. A growing use of soy protein concentrates and isolates is in the preparation of texturized food products that are used as meat alternatives or in cereal products. Soy proteins can also be hydrolysed enzymatically or chemically to produce hydrolysed vegetable protein (HVP) which is used as a flavour enhancer in many foods.

3.2.3 Other soy ingredients

In addition to the products listed above, soy dietary fibre, soy hulls, soy isoflavone extracts, vitamin E and soy phytosterols are examples of other ingredients from soybean processing which are used in nutraceutical products.

4. Compositional differences in soy products

The macro and micro nutrient composition of different soy products will vary markedly depending on soybean variety and the attendant biotic and abiotic influences, processing condition, other ingredients used in processing and whether the whole soybean or specific components are used in product development. Tables 1 to 8 provide detailed lists showing some of the compositional differences of a variety of soy products. As can be seen in the tables, the composition of protein, isoflavones, mineral, trypsin inhibitors, phytic acid and saponins can vary markedly for different soy products.

Additionally, it is important to note that even within a particular food category the type of processing technique and conditions used can impact composition and potential health benefits. We have showed in some of our earlier papers that the conditions used for processing of soy protein concentrates and isolates (such as particle size, method of defatting, process pH) can influence final product composition (Russin et al., 2007; L'hocine et al., 2006). Furthermore, Sobral et al. (2010) also reported that methods and conditions of preparation and storage of protein samples and mixtures of proteins were factors that modified their thermal behavior. In some instances higher denaturation temperatures increased thermal stabilization of soybean storage proteins which was attributed to protein-protein interactions occurring during processing. This increased stabilisation can impact digestibility and potentially, bioactivity.

Conditions used during spray drying can affect product composition. An increase of the inlet air temperature during spray drying of fermented soymilk greatly reduced viability and isoflavone aglycone content (Telang & Thorat, 2010). Moreover, denaturation of proteins during processing further reduced product solubility.

Processing of soybeans under severe alkaline conditions could lead to the formation of lysinoalanine, which can have negative impacts on health and decrease the bioavailability of essential amino acids (i.e., lysine). Processing under milder alkaline conditions avoids the formation of lysinoalanine and reduces potential negative side effects (Liener, 1994).

Additionally, the temperature and time used during the germination of soybeans in the production of germinated soybean flour modified the concentrations of bioactive compounds (i.e., isoflavones, saponins, trypsin inhibitors and lectins) (Pauca-Menacho et al., 2010a). At 25 °C, an increase in germination time decreased the concentration of Bowman-Birk inhibitor, lectin and lipoxygenase. Optimal increases in the concentrations of isoflavone aglycones (daidzein and genistein) and saponin glycosides were observed with a 63 h germination time at 30 °C. In a second study the authors found that germination of soybean for 42 h at 25 °C increased lunasin concentration by 62% and decreased the content of lectins by 59% (Pauca-Menacho et al., 2010b). Germination at 25 °C for 42 h resulted in a 32% increase in the concentration of soy saponins.

Similarly, fermentation of soymilk using a variety of probiotic lactic acid bacteria (LAB) resulted in the production of beta-glucosidase which hydrolyzed isoflavone glucosides to the bioactive isoflavone aglycones, genistein and daidzein in the fermented soymilk (Rekha & Vijayalakshmi, 2010a, 2011). Furthermore, decreases in phytic acid and increases in mineral bioavailability (e.g., calcium) were also observed. Huang et al. (2011) prepared sufu, a fermented soybean curd, by ripening salted tofu cubes in *Aspergillus oryzae*-fermented rice-soybean koji mash at 25, 35 or 45 °C for a period of 16 days and found that regardless of temperature, ripening caused a major reduction in the content of β -glucoside and malonylglucoside isoflavones along with a significant increase of aglycone isoflavone

Soy product	Moisture %	Protein %	Fat %	Carb. ⁴ %	Fiber %	Ash %	Reference
Fresh soybean	68	13	6	11	2	2	Snyder & Know, 1987
Soybean (dry)	7.50-10.1	31.1-36.6	16.3-21.3	6.29	22.0	4.69	Souci et al., 2000
Soybean (dry)	db	39.5-40.1	20.4-21.0	14.7-17.2	21.1-21.7	5.3-5.4	Van der Riet et al., 1987
Soybean sprouts	82	8	2	8	1	1	Snyder & Know, 1987
Soy curd	88	6	3	2	0	0.6	Snyder & Know, 1987
Firm tofu	79.3 - 75.5	10.6-14.3					Shurtleff & Aoyagi, 2000
Edam (cheese)	41.0	28.9	25.0	1.4		3.7	Ono, 2003
Ganmodoki (tofu derivative, td)	63.5	15.3	17.8	1.6		1.8	Ono, 2003
Fried bean curd (td)	75.9	10.7	11.3	0.9		1.2	Ono, 2003
Dried frozen tofu (td)	8.1	49.4	33.2	5.7		3.6	Ono, 2003
Tempeh unfermented soybeans ¹	db	48.3	25.7-28.6	2.0-2.8	17.7-19.3	3.0	Van der Riet et al., 1987
Tempeh fermented (24h)	db	48.7-49.7	25.2-27.9	1.9-2.4	18.3-20.2	2.7-2.8	Van der Riet et al., 1987
Tempeh fermented (48h)	db	48.6-49.7	25.2-26.3	1.9-2.2	16.1-19.3	2.7	Van der Riet et al., 1987
Tempeh fermented (72h)	db	49.3	22.9-23.8	1.7-2.3	15.3-15.7	3.0	Van der Riet et al., 1987
Tempeh (FWB) ²	64	18	4	13		1	Snyder & Know, 1987
Natto (FWB) ²	59	17	10	12	2	3	Snyder & Know, 1987
Hamanatto (FWB) ²	36	26	12	14	3	12	Snyder & Know, 1987
Tempeh Gembus (FSP) ³	81	5	2	11		1	Snyder & Know, 1987
Oncom ampas tahu (FSP) ³	84	4	2	8		2	Snyder & Know, 1987
Soy sauce	72	7	0.5	2	0	18	Snyder & Know, 1987
Fermented soy curd	60	17	14	0.1		9	Snyder & Know, 1987
Soy paste	50	14	5	16	2	15	Snyder & Know, 1987
Ko Chu jang (soy paste)	48	9	4	19	4	20	Snyder & Know, 1987

¹Before fermentation; ² FWB-Fermented whole soybean; ³ FSP-Fermented soy pulp; ⁴Carbohydrate.

Table 1. Composition of different soy foods

Soy product	Moisture %	Protein %	Fat %	Carb. ¹ %	Ash %	Reference
Soy milk	94	3	1	1	0.3	Snyder and Know, 1987
Soy milk	88.7	3.20	1.84	5.76	0.48	Souci et al., 2000
Soy milk (full fat) dried	<10	38	18		<7	Garcia et al., 1997
Soy milk (low fat) dried	<5	48	9		<5	Garcia et al., 1997
Soy milk film	9	52	24	12	3	Snyder and Know, 1987
Soy ice cream	91.12 - 92.45	3.12 - 4.08	1.78 - 1.97	1.89 - 2.66	0.46 - 0.85	Sutar et al., 2010

¹Carbohydrate

Table 2. Composition of soymilk and soymilk derived products

Soy product	Moisture %	Protein %	Fat %	Carbohydrate %	Ash %	Reference
Defatted soy meal		53	1	30		Achouri et al., 2005
Soya flour (full fat)	8.94-9.54	35.9-38.8	19.8-22.1	3.10	4.40	Souci et al., 2000
Soy flour (whole)		50				Hoogenkamp, 2001
Soy flour (natural)		40				Shurtleff & Aoyagi, 1983
Soy flour (full fat) dried	<10	42	21		4.7	Garcia et al., 1997
Soy flour (defatted)	6-8	52-54	0.5-1.0	30-32	5.0-6.0	Endres, 2001
Soy flour (defatted)		51				Shurtleff & Aoyagi, 1983
Soy flour (defatted) dried	6-8	42-52	0.5-1.0		5-6	Garcia et al., 1997
Toasted soy flour	5	38	19	32	5	Snyder & Know, 1987
Soya protein concentrate	4-6	62-69	0.5-1.0		3.8-6.2	Garcia et al., 1997
Soy protein isolate	4-6	86-87	0.5-1.0	3-4	3.8-4.8	Endres, 2001
Soy protein isolate	4-6	86-87	0.5-1.0		3.8-4.8	Garcia et al., 1997

¹From defatted soybean

Table 3. Composition of selected soy ingredients

Soy product	wb/db	Daidzein family ($\mu\text{g/g}$)				Genistein family ($\mu\text{g/g}$)				Glycitein family ($\mu\text{g/g}$)			
		DEN	DIN	MDN	ADN	GEN	GIN	MGN	AGN	GEIN	GIIN	MGIN	AGIN
Soy bean	db	—	1294	2510	—	—	966	2755	—	—	205	200	—
	db	0	19-102	107-539	0	0-22	22-209	26-1113	0-11	0	31-126	110-285	0
Roasted soybeans	wb	39	460	45	397	69	551	63	743	52	68	72	102
	wb	44	474	46	40	77	568	65	71	58	70	74	54
Soy flour	wb	5.7	1087	—	—	8.4	1313	—	—	1	211	—	—
	wb	0	122	286	—	24	309	672	0	—	309	55	—
	wb	4	147	261	—	22	407	1023	1	19	41	57	32
Defatted soy flour	wb	80-480	480-770	—	—	40-460	580-1540	—	—	0-30	60-220	—	—
Soy protein concentrate	wb	20-200	30-760	—	—	10-220	40-1910	—	—	0-40	10-220	—	—
	wb	—	—	—	—	—	18	—	1	23	31	—	—
Soy protein isolate	wb	65.1	1326	—	—	47.8	1450	—	—	6	215	—	—
	db	125	44	155	—	246	124	538	61	—	24	34	—
	wb	80-210	140-300	—	—	50-220	550-800	—	—	10-30	30-60	—	—
	wb	11-63	0-133	18-20	6-74	36-136	137-382	88-100	0-215	22-53	34-55	36-39	33-46
Tofu	wb	46	25	159	8	52	84	108	1	12	8	—	29
	wb	116	453	753	54	140	562	788	66	26	130	131	0
	wb	3-22	40-105	64-156	0-18	4-30	64-143	40-158	8-17	2-4	11-29	7-19	0
Tofu yogurt	wb	—	42	61	—	3	80	79	—	5	12	—	—
Tempeh	wb	137	2	255	11	193	65	164	—	24	14	—	—
	wb	318	117	404	68	518	346	750	76	31	34	34	65
	wb	77-85	93-105	64-66	35-49	89-103	206-226	111	46-50	8-9	14	8	0
Miso	wb	135.8	81	—	—	210.0	162	—	—	19	6	—	—
	wb	61	157	1180	0	39	122	979	0	11	37	217	0
	wb	25-38	62-66	14-32	8-14	29-43	89-92	17-37	20-23	12	10-13	0-8	0-9
Soy milk	wb	18	410	690	22	19	710	871	820	10	65	39	89
Soy milk (aseptically processed)	wb	1-2	49-94	6-29	0	1-2	70-130	10-22	0-10	4-11	6-16	0-4	0
Soy milk (pasteurized)	wb	2-3	20-26	38-42	0	2-4	25-34	39-45	4-5	0-1	4-6	5	0
Soy beverage	wb	19.4	218	—	—	24.8	430	—	—	1	30	—	—
Tempeh burger	wb	34	36	25	—	96	158	—	1	18	18	—	—
Vegetable burger	wb	0.4	18	—	—	0.3	25	—	—	0.3	3	—	—
Soy molasses	wb	3.8	280	—	—	3	96	—	—	1	63	—	—
Soy hot dog	wb	8	35	12	—	16	67	42	4	8	15	15	14

Source: Reprinted with permission from Shimoni, 2004.

(DEN - daidzein; DIN - daidzin; MDN - malonyl daidzin; ADN - acetyl daidzin; GEN - genistein; GIN - genistin; MGN - malonyl genistin; AGN - acetyl genistin; GEIN - glycitein; GIIN - glycitin; MGIN - malonyl glycitin; AGIN - acetyl genistin)

Table 4. Isoflavone content of various soy foods

Soy product	Potassium	Phosp. ¹	Calcium	Magn. ¹	Iron	Zinc	Magn. ¹	Sodium	Copper	Ref.
	mg/100g									
Soybean	1693-1739	635-830	187-275	247-282	11.6-11.7	4.5-6.0	2.8-4.1	3.5-9.3	1.7-1.8	2
Tempeh unfermented	393-525	609-627	192-296	168-172	7.2-8.0	4.5-6.2	3.3-3.7	13.8-16.1	1.6-1.9	2
Tempeh fermented soybeans (24h)	222-224	688-731	225-310	193-221	8.7-8.9	5.4-7.1	3.5-4.1	14.2-14.6	1.8-1.9	2
Tempeh fermented soybeans (48h)	220-224	731-742	232-333	193-195	9.0	5.5-7.2	3.5-4.1	14.1	1.8-1.9	2
Tempeh fermented soybeans (72h)	218-219	734-795	248-318	193-201	8.7-8.8	5.4-7.4	3.6-4.2	13.8-15.1	1.8-1.9	2
Defatted soy four	2400-2700	700-900	200-300	200-300	10	5	3-4	3-15	1-2	3
Soy protein concentrate	100-2400	600-900	200-400	300	10-20	5	5	2-1200	1-2	3
Soy protein isolate	100-1400	500-800	100-200	30-90	10-20	4-9	2	40-1200	1-2	3

¹ Phosp. - phosphorous, Magn. - magnesium, Mang. - manganese;

² Van der Riet et al., 1987;

³ Endres, 2001

Table 5. Mineral content of some soy products

content. The highest increase in aglycone content and greatest decrease in malonylglucosides was observed at 45 °C and increasing ripening time further enhanced the changes observed.

The composition and antioxidant property of tofu is also affected by processing and the type of coagulant used (Rekha & Vijayalakshmi, 2010b). Tofu prepared with natural coagulants (*Citrus limonum*, *Garcinia indica*, *Tamarindus indica*, *Phyllanthus acidus* and *Passiflora edulis*) had significantly higher antioxidant activity compared to those prepared with salts. Furthermore, higher total crude protein and fat contents were found in some of the tofu prepared using the plant based coagulants (*G. indica* and *T. indica*).

Differences were reported in the composition and properties of soy whey (aqueous extract of defatted soybean flour) and tofu whey (liquid industrial residue from tofu production) (Sobral & Wagner, 2009), two products that may appear to be similar in properties. Although both byproducts contain primarily carbohydrates, proteins, non-protein nitrogen and salts, tofu whey reportedly had lower amounts of dry matter and proteins, and the antitryptic activity was three times lower than in soy whey. Thermal studies conducted using differential scanning calorimetry (DSC) of soy whey proteins showed endotherms corresponding to lectin and antitryptic factors of Kunitz and Bowman-Birk, whereas the thermogram for tofu whey only showed the presence of antitryptic factors. The authors attributed these differences to the manufacturing processes used. The extent to which these differences can affect health outcomes is unclear.

Soy product	Trypsin inhibitor activity mg/g sample	Reference
Whole soybean	16.7-27.2	Hafez, 1983
Whole soybean	48.2	Miyagi et al., 1997
Raw soy flour	28-32	Rackis et al., 1985
Raw soy flour	52.1	Anderson et al., 1979
Toasted soy flour	7.9-9.4	Rackis et al., 1985
Toasted soy flour	3.2-7.9	Anderson et al., 1979
Soy protein concentrate	5.4-7.3	Peace et al., 1992
Soy protein concentrate	6.3-13.9	Anderson et al., 1979
Soy protein concentrate	4.4-7.3	Peace et al., 1994
Soy protein isolate	1.2-30.0	Peace et al., 1992 Rackis et al., 1985
Soy protein isolate	4.4-11.0	Anderson et al., 1979
Soy based infant formulas	0.3-2.7	Peace et al., 1992
Soy tofu	0.6	Doell et al., 1981
Soy tofu	1.2-3.8	Miyagi et al., 1997
Soy milk	6.3	Miyagi et al., 1997
Soy sauce	0.3	Doell et al., 1981
Soy miso	4.1	Doell et al., 1981
Soy food fiber	6.47	Anderson et al., 1979
Chicken analog*	3.63	Anderson et al., 1979
Ham analog*	5.36	Anderson et al., 1979
Beef analog*	3.42	Anderson et al., 1979
Textured soy flour	5.15	Anderson et al., 1979

* manufactured using soy protein products

Cited from the following sources: Anderson & Wolf, 1995; Snyder and Know, 1987 (calculated on the basis of 1.9TUI = 1 g TI); Gilani et al., 2005.

Table 6. Trypsin inhibitor activity of different soy products

Soy product	Phytic acid % (dry basis)	Reference
Soybean	1.12-1.80	Toda et al., 2006
Soybean	1.00-1.47	Lolas et al., 1976
Soybean	1.32-2.30	Raboy et al., 1984
Whole soybean (dry)	1.07	Sutardi & Buclke,1985a
Soybean, raw	1.41	Sudarmadji & Markakis, 1977
Soaked (24h)	1.69	Sutardi & Buclke,1985a
Soybean, soaked	1.43	Sudarmadji & Markakis, 1977
Soybean, boiled (5 min)	1.68	Sutardi & Buclke,1985a
Soybean, boiled	1.23	Sudarmadji & Markakis, 1977
Soybean soaked (24h)	1.67	Sutardi & Buclke,1985a
Soybean (dehulled)	1.65	Sutardi & Buclke,1985a
Soybean (steamed 30 min)	1.48	Sutardi & Buclke,1985a
Soybean (drained and cooled)	1.47	Sutardi & Buclke,1985a
Full fat soy flour	1.51-1.81	Ranhotra et al., 1974
Defatted soy flour	1.62-1.85	Ranhotra et al, 1974
Defatted soy flour	1.30-1.63	Schuster & Bodwell, 1980
Textured soy flour	1.10-2.02	Davies & Reid, 1979
Textured vegetable protein	0.95-1.63	Harland & Oberleas, 1977
Concentrate	1.25-2.17	Ranhotra et al, 1974
Textured concentrate	1.48-1.50	Harland & Oberleas, 1977
Soy protein isolate	0.97-1.69	Schuster & Bodwell, 1980
Soy protein isolate	1.61-2.00	Honig et al., 1984
Spun isolate fiber	1.48	O'Neill et al., 1980
Soymilk	1.68	Omosaiye & Cheryan, 1979
Soymilk	1.83	Beleia et al., 1990
Tofu	1.5-2.5	Van der Riet et al., 1989
Tofu	1.96-2.88	Schaefer & Love, 1992
Okara (residue from soymilk)	0.5-1.2	Van der Riet et al., 1989
Tempeh	0.69-0.73	Sutardi & Buckle, 1985b
Tempeh unfermented soybeans (before fermentation)	1.0-1.2	Van der Riest et al., 1987
Tempeh fermented soybeans (24h)	0.3-0.6	Van der Riest et al., 1987
Tempeh fermented soybeans (48h)	0.2-0.4	Van der Riest et al., 1987
Tempeh fermented soybeans (72h)	0.1-0.2	Van der Riest et al., 1987
Tempeh (fresh)*	0.68-0.75	Sutardi & Buclke,1985a
Tempeh	0.96	Sudarmadji & Markakis, 1977
Fried fresh Tempeh*	0.35-0.38	Sutardi & Buclke,1985a
Tempeh stored 2wk@5°C*	0.18-0.19	Sutardi & Buclke,1985a
Fried stored tempeh*	0.09-0.10	Sutardi & Buclke,1985a

* Different types of inoculation used in production

Data taken from the following source: Anderson & Wolf, 1995

Table 7. Phytic acid content of different soy products

Soy product	Saponin content %	Reference
Soybean	0.44-0.49	Berhow et al., 2006
Soybean	0.47	Ireland et al., 1986
Soybean	0.225-0.298	Kitagawa et al., 1984
Soybean	5.1	Fenwick & Oakenfull, 1981
Soybean	3.9	Fenwick & Oakenfull, 1983
Soybean	0.46-0.50	Gestetner et al., 1966
Soybean	5.6	Fenwick & Oakenfull, 1981
Toasted, defatted soy flour	0.67	Ireland et al., 1986
Defatted soy flour	2.0	Fenwick & Oakenfull, 1981
Defatted soy flour	0.35	Curl et al., 1985
Defatted soy flour	2.2-2.5	Fenwick & Oakenfull, 1981
Commercial soy flour	0.46	Price et al., 1985
Soya hulls	2.0	Fenwick & Oakenfull, 1981
Full fat, enzyme active soy flour	0.43	Ireland et al., 1986
Full fat, heat treated soy flour	0.49	Ireland et al., 1986
Soya protein isolate	0.76	Ireland et al., 1986
Soy protein isolate : Promine D*	0.3	Fenwick & Oakenfull, 1981
Soy protein isolate : GL-750*	0.8	Fenwick & Oakenfull, 1981
Soy protein isolate : Maxten C*	1.9	Fenwick & Oakenfull, 1981
Soy protein isolate : Maxten E*	2.5	Fenwick & Oakenfull, 1981
Soymilk	0.026	Ireland et al., 1986
Soymilk	0.022	Ireland et al., 1986
Soymilk	0.39	Kitagawa et al., 1984
Yuba (dried soymilk film)	0.41	Kitagawa et al., 1984
Okara (residue of soymilk)	0.10	Kitagawa et al., 1984
Tofu	2.1	Fenwick & Oakenfull, 1981
Tofu	0.30-0.33	Kitagawa et al., 1984
Miso	0.15	Kitagawa et al., 1984
Natto	0.25	Kitagawa et al., 1984
Tonyu (soya milk)	0.047	Kitagawa et al., 1984

*trademark

Table 8. Saponin content of different soy products

In addition to processing, various plant breeding techniques have been used to modify the composition of soybeans for different food applications (Esteves et al., 2010, Brune et al., 2010). Thus depending on the type of variety used, compositional differences can be

expected in macro and micro components (i.e., protein, indispensable, dispensable and total amino acid, lunasin, isoflavones, phytic acid, oxalate and trypsin inhibitors as well as mineral content). The biological activity of soybean products will, therefore, not only depend on the processing technique used but the variety of soybean used.

5. Soyfoods and health claims

An important factor that may contribute to inconsistencies in the evidence of the health benefit of soy may be related to the type of material used for clinical studies. Reinwald et al. (2010) argue the possibility that whole soy may have a more unique effect on health than a select soy component.

Differences in health outcomes related to soy and soy component consumption could be due to varietal and compositional differences, the impact of processing, additive effects of various components (i.e., the whole is greater than the sum of the parts) as well as age and health status. Furthermore, some have argued that the presence or absence of specific gut microflora could also contribute to health outcomes (Patisaul & Jefferson, 2010) as microorganisms may play a critical role in converting physiologically inactive phytochemicals to the bioactive form.

As an example, Reinwald and Weaver (2010) have reported that whereas epidemiological studies in Asia evaluating diets containing traditional whole soyfoods showed a positive association with bone mineral density and fracture protection, smaller scale intervention studies in Western nations mainly using isolated soy protein (SP) and purified or concentrated soy isoflavones (SI) rather than whole soyfoods have produced inconsistent results.

Similarly, Lagari and Levis (2010) found that clinical trials are conflictive regarding the effects of phytoestrogens on bone mineral density and bone turnover markers in premenopausal and postmenopausal women and argue that much of the controversy lies in differences in study design, reporting of results, participants' age and menopausal status, and type and dose of phytoestrogen used.

In Canada, where a health claim for soy has not yet been approved, the ministry responsible for health (Health Canada) requires the following evidence to substantiate health claims:

1. **Causality** - Evidence of high quality and quantity of original research in humans based on randomized controlled intervention and / or prospective observational studies are mandatory to substantiate the health claim of food or food constituents with high level of certainty (statistical significance achieved at $p \leq 0.05$) and the relationship between the amount of food and the health effect.
2. **Generalizability** - The claimed effect of the food or food constituent is biologically / physiologically relevant and expected to benefit the health of the target population, appropriate and validated surrogate marker must be used to ensure the biological relevance and it should be part of the causal pathway between the food and the health outcome.
3. **Quality assurance** - The food is produced according to quality standards and consistently meets predefined specifications.
4. **Safety** - The subject of a health claim application must be for a food approved for safe use; or, if a novel food is the subject of the health claim, a novel food application must

be completed and submitted to Health Canada preceding or concurrent with this application. The adverse effects related to food intake observed in human studies should be addressed and must provide risk management strategies to overcome the adverse effects and or restriction on use of food.

As there is a wide variety of soy products available on the market, the specific type used for clinical studies can impact results.

A major challenge for health claim support studies is the lack of standardized materials and controls for clinical studies as well as the use of appropriate clinical outcomes and surrogate endpoints for different disease risks. Furthermore, very few studies have been conducted to understand what happens to soyfoods and soy components in the gastrointestinal tract and the specific events that occur at the mucosal barrier and how these events influence absorption, distribution, metabolism and excretion. The gastrointestinal mucosa is an interesting and complex system that acts as a barrier between the body and the luminal environment and is selective in that it allows the transfer of selected nutrients across the epithelium while excluding perceived harmful components in the bolus. This complex interplay between bioactives and the intestinal mucosa also needs to be carefully documented to support any eventual health claim. Matrix interactions can influence the bioactivity of the components of interest and the functional properties of the matrix. Furthermore, for double blind placebo controlled studies which are the gold standard in clinical studies, the effect of the matrices used in the clinical trials need to be carefully evaluated.

As policy makers, food regulators, industry and consumers in other jurisdictions demand more evidence to support current and future health claims, some question of interest that remain include the following:

- What is the relationship reported between the exposure to soy and specific health effects from both observational and intervention studies that meet selection criteria?
- Can a clear distinction be made between the effect of soy protein alone and soy protein within a matrix (e.g., whole soybean foods and foods made from soy protein ingredients)?
- What is the efficacy of soy proteins in which isoflavones have been removed versus soy proteins that contain isoflavones in respect to relationship with cardiovascular disease risk reduction and other health claims?
- How are breakdown products absorbed in the gastrointestinal tract and how do they exert physiological effects in the body?
- How do matrix effects influence adsorption, digestion, metabolism and excretion?
- What is the impact of processing on the efficacy of soy protein and the bioactivity of materials used in clinical trials?
- What are the ideal samples and controls that should be used in clinical trial studies for specific clinical outcomes and what surrogate endpoints should be used?
- What is the feasibility for consuming recommended intake levels for health claims especially in populations that do not consume soy frequently (e.g., is the consumption of 20-25 g of soy protein a day feasible?)?
- What is the documented effect of anti-nutritional factors such as trypsin inhibitors, phytates, lectins and lysinoalanine and the extent of heat resistance of trypsin inhibitors found in soybeans?

6. Conclusion

Soy protein has officially been approved in several countries as a “functional food”, making it one of the most valuable vegetable proteins in the world today. As concerns about health, climate change and the impacts of agricultural practices increase, assurance of a diversified sustainable source of nutrition that provides proven health benefits will become increasingly important. Soybeans can contribute to this nutrient biodiversity in an instrumental way. In crop production and rotation, soybeans play a crucial role in nitrogen fixation making them an important component of agricultural sustainability. Furthermore, soy proteins have excellent functional properties that can be exploited in various food applications. Processing technologies are continually being investigated for soy protein fractionation which will allow modifications in protein profile and which could dramatically improve suitability of soy protein products for targeted food, nutraceutical and industrial applications (e.g., enriched 11S, 7S, or 2S soy protein extracts). Breeding efforts to remove or enhance micro nutrients and other bioactive components are also likely to have an impact on improving health outcomes. To support health claims and enhance the benefits of consumption, constant evaluation of the totality of the body of knowledge in regards to potential health benefits and well designed experimental studies using well characterized materials will, therefore, be needed.

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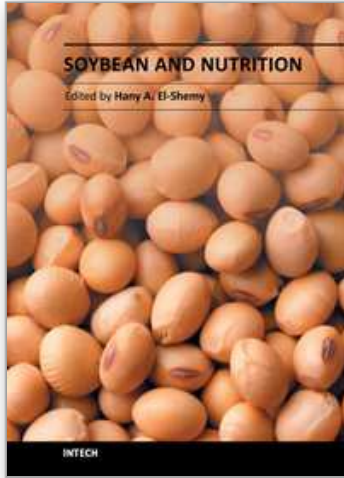
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Worldwide, soybean seed proteins represent a major source of amino acids for human and animal nutrition. Soybean seeds are an important and economical source of protein in the diet of many developed and developing countries. Soy is a complete protein and soy-foods are rich in vitamins and minerals. Soybean protein provides all the essential amino acids in the amounts needed for human health. Recent research suggests that soy may also lower risk of prostate, colon and breast cancers as well as osteoporosis and other bone health problems and alleviate hot flashes associated with menopause. This volume is expected to be useful for student, researchers and public who are interested in soybean.

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University Campus STeP Ri
Slavka Krautzeka 83/A
51000 Rijeka, Croatia
Phone: +385 (51) 770 447
Fax: +385 (51) 686 166
www.intechopen.com

InTech China

Unit 405, Office Block, Hotel Equatorial Shanghai
No.65, Yan An Road (West), Shanghai, 200040, China
中国上海市延安西路65号上海国际贵都大饭店办公楼405单元
Phone: +86-21-62489820
Fax: +86-21-62489821

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