

MAIN NUTRITIONAL AND ANTINUTRITIONAL COMPOUNDS OF BEAN SEEDS – A REVIEW*Urszula Krupa**Division of Food Science, Institute of Animal Reproduction and Food Research of the Polish Academy of Sciences, Olsztyn*

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Bean seeds have a unique nutritive value as they deliver valuable proteins, saccharides, minerals and vitamins, and dietary fibre. Besides, they contain a wide range of bioactive compounds that cannot be considered as nutrients, however they exert physiological effects on humans. The purpose of the present review is to describe the main nutritional and antinutritional compounds of bean seeds as well as to demonstrate their positive and adverse effects on human health.

INTRODUCTION

The increasing number of chronic diseases and growing costs of health care have prompted researchers to revise the current guidelines concerning nutrition. Nowadays, nutrition as a science needs to expand above its basic functions, like the prevention of dietary deficiency and the establishment of nutrition standards and dietary guidelines, into the new concept focused on the support of the state of well-being and health, and minimizing the risk of diet-related diseases associated with either excess or deficiency of some nutrients. Facing the growing expectations of consumers interested in benefits resulting from nutrition, expected to support the disease control and prevention, a variety of concepts of innovative food products with specific nutrient properties have appeared worldwide. The “functional food” concept was developed in Japan at the early 1980s and as “food for specified health use (FOSHU)” was established in 1991 [Diplock *et al.*, 1999]. Functional food, defined as “any food or ingredient that has a positive impact on an individual’s health, physical performance, or state of mind, in addition to its nutritive value” [Goldberg, 1994], must satisfy the following conditions: should be naturally occurring, can be consumed as part of the daily diet, and when ingested should enhance or regulate a particular biological process or mechanism to prevent or control specific diseases. Soon, the term “functional food” was accompanied by numerous related expressions such as “nutraceuticals”, “pharmafood”, “medifood”, “vitafood” *ect.* Although all these terms, derived from a field of nutrition science, emphasize the beneficial effects of food components and their interactions with body functions and/or pathological processes, though they are intrinsically dissimilar in meaning, besides they are often misused for nutrients or nutrient-enriched food that can prevent or treat diseases.

The term “nutraceutical” was defined by the Foundation for Innovation in Medicine [Pszczola, 1992] as “substances considered a food or part of a food, that provide medical and/or health benefits, including the prevention and treatment of diseases”, in order to distinguish between functional food and drugs. Nutraceuticals are clearly not drugs, which are pharmacologically active substances, but surely are components that not only maintain, support, and normalize any physiological or metabolic function, but can also potentiate, antagonize, or otherwise modify physiological or metabolic functions [Hardy, 2000]. This statement suggests that any single naturally occurring ingredient in the form of powder or tablet, not necessarily a complete food, could be recognized as a nutraceutical. Therefore, the use of the term “nutraceutical” and other related definitions, nevertheless conceptually different, in relation to food components involved in disease prevention or treatment should be reserved only to cases demonstrating a biological activity, examined in the course of the intricate physiological processes proceeding in human body. Duranti [2006] suggested that the attribution of specific functionalities to a food or a food component is not always scientifically demonstrated since the cause/effect relationships of single food components are difficult to prove, specially in men.

Legumes, including beans, occupy an important place in human nutrition as in many countries they are one of the staple food. Bean seeds have unique nutritive value. Besides being a cheap source of valuable proteins, saccharides, and several micronutrients including minerals and vitamins, they are known as rich in dietary fibre and low in fat [Sgarbieri, 1989; Soral-Śmietana *et al.*, 2002]. The contribution of legumes in the daily diet has many beneficial physiological effects as it allows to prevent common metabolic diseases, such as diabetes mellitus, coronary heart disease (CHD) and cancer [Bassano *et al.*, 2001; Champ, 2001; Mathers, 2002].

A regular intake of beans, or any other pulse, may contribute to the lowering of the plasma cholesterol level [Leterme, 2002]. In addition, beans, together with peas, lentils and chickpeas, are also shown as the best sources of folate, the vitamin that lowers the blood level of homocysteine. Therefore, their consumption is supposed to have a positive correlation with reducing the CHD death [Mann & Chisholm, 1999]. Legumes contain a wide range of biologically-active microconstituents that cannot be considered as nutrients, however possess many beneficial properties, like antioxidative, anti-inflammatory, detoxicating, which may be useful in certain diseases prevention. Of particular interest are resistant starch, enzyme inhibitors, lectins and polyphenols, therefore their role as preventive agents in diets of persons suffering from metabolic disorders is gaining attention. The term "antinutritional compounds" (ANCs) will be used in the present paper in regard to bioactive compounds of bean seeds.

The consumption of grain legumes in Europe is lower than in other regions of the world, however, there are variations between countries and generally a slight increase has been observed in recent years [Schneider, 2002]. Although legumes are a valuable food supply, in most countries of West European the consumption of legume seeds, especially beans, diminished considerably [Champ, 2001]. The factors limiting the consumption of legumes are mainly: an inadequate level of innovation for developing the legume products adapted to modern life, a small home supply of legumes and competition from cheaper low-quality imports [Schneider, 2002]; moreover gastric problems following their consumption, a low sensory value and a long time of preparation of legume-based dishes.

The purpose of this review is to describe the main nutritional and antinutritional compounds of bean seeds, to demonstrate their positive and adverse effects on human health and to assess the influence of technological processing on their activity.

BEAN SEEDS COMPOSITION

Cultivated under Polish climatic conditions bean varieties are rich in nutritional components, especially proteins and starch, but also in valuable non-nutritional components such as resistant starch and dietary fibre [Soral-Šmietana *et al.*, 2002].

Starch content in legumes is high and ranges from 22% to 45%, in bean seeds it reaches above 40% [Hoover & Sosulski 1991; Hedley, 2001]. Depending on botanical origin, legume starch occurs in a granular form of various shapes and sizes [Tharanthan, 1995; Tester *et al.*, 2004] and bean starch has kidney-like or oval shape. In a single starch granule, around *hilum* interior exocentric or concentric arrangement of layers can be observed, arising probably as a consequence of uneven starch hydration during starch granule formation [Gallant *et al.*, 1997]. Chemically, starch is a mixture of two glucose polymers: linear amylose and branched amylopectin, and a minor third component known as the intermediate fraction, which is neither amylose nor amylopectin in its primary structure [Tharanthan & Mahadevamma, 2003; Tester *et al.*, 2004]. From the nutritional point of view, starch is a heterogeneous component and can be divided into easily digested starch (DS) and digestion-resistant fraction (RS) [Soral-Šmietana,

2000]. Dry bean seeds are rich in dietary fibre and resistant starch fraction [Soral-Šmietana & Krupa, 2005]. Substantial quantities of starch not hydrolysed in the small intestine reach the large bowel and can be fermented in the colon [Gordon *et al.*, 1997]. Cassidy *et al.* [1994] suggest a strong negative correlation between the starch intake and the risk of colorectal cancer. These authors hypothesized that it is the resistant starch which provides the protection.

In comparison with cereals, in which the content of proteins varies from 5 to 15%, bean seeds are a valuable source of proteins, containing from 17 up to 39% d.m. [Bressani, 1993; Krupa & Soral-Šmietana, 2003; Soral-Šmietana *et al.*, 2003]. Most of them are devoid of any catalytic activity nor play any structural role in the cotyledonary tissue. These proteins, termed as storage proteins, are stored in membrane-bound organelles, storage vacuoles or protein bodies, in the cotyledonary parenchyma cells, survive desiccation in seed maturation and undergo proteolysis at germination, thus providing free amino acids, as well as ammonia and carbon skeletons to the developing seedling [Duranti, 2006]. Despite bean seeds storage proteins, similarly to other legumes storage proteins, are relatively low in methionine and tryptophan, yet they are high in lysine. For this reason they are known as a dietary supplement of cereal proteins, generally lacking for this amino acid. Amino acids profile is important in predicting the potential value of proteins, however the main determinants of their nutritive value is their digestibility and availability [McNab, 1994]. The factors limiting the biological value of bean proteins are few. One of them is the resistance to digestion [Deshpande & Nielsen, 1987; Nielsen *et al.*, 1988; Melito & Tovar, 1995]. Generally, plant proteins have been reported to be less susceptible to proteolytic breakdown *in vivo* than animal proteins [Fridman, 1996]. Deshpande & Damodaran [1989], analysing the heat-induced conformational changes of phaseolin, the main fraction of bean seeds storage proteins, suggest that the disruption of its tertiary and quaternary structure following heating is the crucial step to enhance its susceptibility to trypsin. Microwave cooking appreciably improved the digestibility of moth bean (*Phaseolus aconitifolius* Jacq) protein [Negi *et al.*, 2001]. The *in vitro* experimental approaches demonstrated that the resistance of bean proteins to proteolysis is related to their structure stabilized by S-S bonds and carbohydrate moiety [Piecyk *et al.*, 2000]. Besides, the proteolytic resistance has been also attributed to the presence of antinutritional compounds which affect proteins digestibility alone or together with other components [Liener, 1989, 1994; Periago *et al.*, 1996]. Improving the nutritional value of legume proteins requires the application of various methods of technological processing. The physicochemical processing of legume seeds can diminish the negative influence of ANCs on their proteins digestibility [Klepacka *et al.*, 1997; Carbonaro *et al.*, 2005].

MAIN ANTINUTRITIONAL COMPOUNDS OF BEAN SEEDS

Bean seeds contain a number of antinutritional compounds which can be of proteinous or non-proteinous nature. They are difficult to classify as their structure and physi-

ological effects are very diverse. Several ANCs are ubiquitous, like proteinase inhibitors, lectins, phytates, polyphenols, other are more specific, as some complex glycosides. Most of food antinutrients have an impact on the digestive system, like the inhibition of digestive enzymes (*e.g.* protease inhibitors), impairment of hydrolytic functions and of transport at the enterocyte site (lectins), formation of insoluble complexes which cannot be adsorbed, decrease of bioavailability of some nutrients (phytates, polyphenols), and the increase of the production of gases in the colon (α -galactosides) [Thompson, 1993]. Bean seeds antinutrients are considered to limit protein and carbohydrate utilization. However, negative effects of several of these compounds, manifested in human and animal bodies, are only observed after the consumption of raw and unprocessed seeds or flour, as normally heat denaturation inactivates ANCs which are sensitive to high temperature.

Protease inhibitors are proteins of low molecular weight forming stable complexes with digestive enzyme, irreversibly inhibiting their activity. The most characterised protein inhibitors of legume seeds are trypsin inhibitor of both, Bowman-Birk type and Kunitz type, and α -amylase inhibitors. The presence of protease inhibitors in food decreases the apparent nutritional quality of proteins in the diet by affecting the ability of body digestive enzymes to degrade dietary protein, and thus limiting the intake of amino acids needed to construct new proteins. However, in certain situations the effects of inhibitors on protein digestion might be advantageous, *e.g.* by improving the intact absorption of some therapeutic proteins such as orally delivered insulin [Yamamoto *et al.*, 1994]. Moreover, the control of proteases activity, considered to play a decisive role in a wide range of biological processes and malfunctioning related to cancer progression, may be considered as anticarcinogenic mechanism [Clemente & Domoney, 2001]. Several *in vitro* and *in vivo* studies have provided evidence that certain protease inhibitors of legume seeds are effective at preventing or suppressing carcinogen-induced transformation [Troll *et al.*, 1984; Kennedy, 1993; Banerji & Fernandes, 1994; Fernandes & Banerji, 1995, 1997]. Steinmetz & Potter [1991] in their extensive review, concluded that "consumption of higher levels of vegetables and fruits is associated consistently, although not universally, with a reduced risk of cancer at most sites, and particularly with epithelial cancer of the alimentary and respiratory tracts". Most studies on the health-promoting properties of legume protease inhibitors have used the Bowman-Birk inhibitor (BBI) from soybean. However, other grain legume seeds are rich sources of protease inhibitors as well [Clemente *et al.*, 2004]. A bean seed amylase inhibitor, which is composed of two glycopolyptide subunits, alpha and beta, was well characterised [Lajolo & Finardi Filho, 1985; Gibbs & Alli, 1998], its complete amino acid sequence was established by Kasahara *et al.* [1996]. Amylase inhibitor can reduce starch digestion. A partially purified amylase inhibitor derived from white beans slows dietary starch digestion *in vitro*, rapidly inactivates amylase in the human intestinal lumen, and, at acceptable oral doses, may decrease intraluminal digestion of starch in humans [Layer *et al.*, 1985]. Besides, it significantly decreases duodenal, jejunal, and ileal intraluminal amylase activity, reduces the early postprandial plasma glucose and eliminates

the late postprandial glucose, and abolishes postprandial plasma concentrations of insulin, C-peptide, and gastric inhibitory polypeptide [Layer *et al.*, 1986]. A general claim on the anti-diabetic role of α -amylase inhibitors has already been published [McCarty, 2005] and some patents concerning the use of food preparations containing suitable amounts of α -amylase inhibitors for the obesity control and the prevention and treatment of diabetes have appeared [Suzuki *et al.*, 2003; Muri *et al.*, 2004]. These findings confirmed the potential of α -amylase inhibitors to be used as a nutraceutical compound. However, the positive or negative effect of all enzyme inhibitors depends on their level in different legumes and on the dose and frequency of consumption.

Legumes are the main sources of lectins in ordinary human food. Most of bean species seem to be a good source of lectins, but their content depends on bean variety [Bond & Duc, 1993]. Lectins are glycoproteins of non-immune origin that are capable of the recognition and reversible binding to carbohydrate moieties without the alteration of the covalent structure of the recognized glycosyl ligands [Vasconcelos *et al.*, 2004]. They exhibit specific carbohydrate binding activities and many of them have haemagglutinating activity. The toxicity of lectins is characterised by growth inhibition in experimental animals, and by diarrhoea, nausea, bloating and vomiting when injected in humans [Liener *et al.*, 1986]. Lectins display a variety of biological activities, including anti-tumor [Abdullaev & Mejia, 1997], immunomodulatory [Rubinstein *et al.*, 2004], anti-fungal [Herre *et al.*, 2004], and insecticide [Macedo *et al.*, 2003]. Literature data reported the inhibitory effect of lectins on human immunodeficiency virus type 1 reverse transcriptase (HIV-1 RT) [Ye & Ng, 2001; Wong & Ng, 2003; Barrientos & Gronenborn, 2005]. Red kidney beans contain lectin phytohaemagglutinin (PHA) [Shi *et al.*, 2007] which possess a potential for cell agglutination and mitogenic activities. These authors demonstrated that extracts from raw red kidney bean and canned red kidney bean contained bioactive compounds capable of inhibiting HIV-1 RT *in vitro*.

Bean seeds contain a number of non-protein ANCs with different chemical structure and properties, like phenolic compounds, saponins, alkaloids, phytates *etc.* that impair the biological utilization of their nutrients. Among polyphenols, tannins will be extensively described in this review. The content of tannins in dry bean seeds varies from 0.00 to 0.93% [Deshpande *et al.*, 1986]. They are compounds of intermediate to high molecular weight (up to 30,000 Da). In bean seed, the major amount of tannins are located in the seed coat, with low or negligible amounts located in the cotyledons. Tannins are known to interact with proteins forming complexes which, in turn, decrease the solubility of proteins and make protein complexes less susceptible to proteolytic attack than the same proteins alone [Reddy *et al.*, 1985; Carbonaro *et al.*, 1996]. Besides, they impair starch and disaccharide assimilation [Carmona *et al.*, 1996], and interact with proteolytic enzymes inhibiting their activity. Other toxic effects of tannins can be categorized as: depression of food intake, inhibition of digestive enzymes, increased excretion of endogenous protein, digestive tract malfunctions and toxicity of absorbed tannin or its metabolites [Jansman & Longstaff, 1993]. However, tannins and other plant polyphenols (anthocyanins, flavonoids)

are receiving growing interest due to their potential role as protective factors against free radical mediated pathologies, such as cancer and atherosclerosis, in humans [Kehrer, 1993]. Bawadi *et al.* [2005] demonstrated that water-soluble condensed tannins isolated from black beans inhibited the growth of Caco-2 colon, MCF-7 and Hs578T breast, and DU 145 prostatic cancer cells. Other findings, associating polyphenols to free radicals scavenging and metal chelating activities, suggested their potential beneficial implications in the treatment and prevention of cancer [Gali-Muhtasib *et al.*, 2001; Gomez-Cordoves *et al.*, 2001; Hangen & Bennink, 2002]. Literature data suggest that, in spite of the known adverse action on protein digestibility, legume seed tannins might exert a beneficial antioxidant activity and contribute to diseases prevention.

EFFECT OF PROCESSING ON ANTINUTRITIONAL COMPOUNDS OF BEAN SEEDS

In general, legume seeds are consumed after technological treatments leading to changes in the internal arrangement of cotyledon structure and modification of the main polymer, starch and proteins, properties. Different processing methods such as boiling [Jood *et al.*, 1985], hydration and germination [Matella *et al.*, 2005] have been used to increase the utilization of bean seeds. Technological processing may evoke positive effects like proteins coagulation, starch swelling and gelatinisation, texture softening and formation of aroma components however, the conditions applied may induce some undesirable modifications like a loss of vitamins and minerals, the formation of indigestible aggregates and changes in their conformation [Ohlsson, 2002; Krupa *et al.*, 2007]. Inactivation and/or removal of undesirable components is essential in improving the nutritional quality and organoleptic acceptability of beans and, in turn, helps to effectively utilize their potential as human food and animal feed. Decrease of ANC's may occur either by their physical elimination or by heat inactivation, as many of them, especially the proteinous ANC's, are thermally-sensitive. Removing the seed coat, which constitutes about 10% of the dry bean seeds [Soral-Šmietana *et al.*, 2002], may influence the concentration of some antinutrients on a unit weight basis [Deshpande *et al.*, 1982], thus evoking changes in proteins digestibility. Thermal treatment may improve the nutritional value of food by reducing proteinase inhibition, thus increasing the availability of lysine and other amino acids. Proteinase inhibitors are sensitive to physical treatment, and can be denatured by heat, however the degree of the inactivation depends on their thermal stability and seed variety. In general, thermal processing decreases the activity of trypsin inhibitors to a harmless level [Liener, 1989]. Inactivated by heat, protease inhibitors play a positive nutritional role due to a high content of sulfur-containing amino acids, as compared to the majority of plant seed proteins [Ryan, 1990]. Besides, heat processing may reduce the toxicity of lectins [Reddy & Pierson, 1994], but low temperature or insufficient cooking may not completely eliminate their toxicity [Franz, 1991]. However, care must be taken to avoid using excessive heat, as it can impair the nutritional value of dietary proteins by causing crosslinking reactions or amino acids racemization [Garcia-Carreno, 1996]. The impact of phytic acids and tan-

nins can also be reduced/eliminated through the thermal processing [Alonso *et al.*, 2000; Habiba, 2001]. Several studies demonstrated that soaking, cooking, and fermentation of legume seeds reduced the contents of phytic acid, tannins, phenols, α -amylase and trypsin inhibitors [Vidal-Valverde *et al.*, 1992; Abd El-Hady & Habiba, 2003]. Shimelis *et al.* [2007] investigating the effects of different processing and their combinations on the content of ANC's of *Phaseolus vulgaris* varieties, indicated that germination significantly reduced certain heat-stable antinutrient components, whereas cooking of pre-soaked beans appeared as an adequate method for reducing heat-sensitive antinutrients. The authors concluded that no single method can remove or eliminate most of these toxic factors, therefore they suggested a combination of autoclaving followed by germination as the most promising method of ANC's reduction/elimination.

CONCLUSIONS

Unbalanced, high-fat diet aggravated by physical inactivity is widely believed to be a major contributing factor in metabolic diseases (obesity, type 2 diabetes, CHD and cancer). Therefore, it becomes urgent to promote an increased contribution of legumes, including beans, in the diet in order to take advantage of their components that are nutritious and provide most of the ingredients that help to improve health. Generally, the beneficial effect of legumes on human health, when consumed in significant amounts, is attributable to their nutritive compounds, although it is likely that the bioactive ANC's present in the legumes play an important role as well.

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