(62.35 mg/100g). The antioxidant activity as determined by three different methods showed a wide range of values. DPPH radical scavenging activity (1.07 TE/g), FRAP (373 µmol/g) and reducing power (4.89 mg/g), all three were highest in Rajmash. It was further showed that in pulses the total phenolic content (TPC) was poorly correlated with antioxidant activity (AOA), suggesting thereby that only TPC might not contribute significantly to the antioxidant activity in pulses [37].

Enzyme Inhibitors

Protein inhibitors of hydrolases present in pulses are active against proteases, amylases, lipases, glycosidases, and phosphatases. From the nutritional aspect, the inhibitors of the serine proteases trypsin and chymotrypsin found in plant foodstuffs are the most important [38]. Beans are the second largest group of seeds after cereals reported as natural sources of α-amylase inhibitors [39]. Protease inhibitors isolated from pulses are generally classified into two families, referred to as Kunitz and Bowman-Birk on the basis of their molecular weights and cystine contents. Kunitz type inhibitors have a molecular mass of ~20 kDa, with two disulfide bridges, and act specifically against trypsin. Bowman-Birk type inhibitors with a molecular mass of 8–10 kDa, have seven disulfide bridges, and inhibit trypsin and chymotrypsin simultaneously at independent binding sites. Protease inhibitors interfere with digestion by irreversibly binding with trypsin and chymotrypsin in the human digestive tract. They are resistant to the digestive enzyme pepsin and the stomach's acidic pH. In common bean, lima bean, cowpea, and lentil, protease inhibitors have been characterized as members of the Bowman-Birk family [38;39]. In pea, large genetic variability is available for the activity of Bowman-Birk trypsin/chymotrypsin inhibitor proteins. Protease inhibitor content is moderate in kidney bean and cowpea (8 and 10.6 g of trypsin and 9.2 g of chymotrypsin inhibited kg^{-1}, respectively) [40]. The content of α-amylase inhibitors differs greatly among legumes, with the highest amount found in dry bean.

Lectins

Lectins are proteins or glycoproteins that agglutinate erythrocytes of some or all blood groups *in vitro* depending on their specificity and high binding affinity for a particular carbohydrate moiety on the cell surface [41]. Lectins can reduce the digestibility and biological value of dietary proteins and inhibit the growth of experimental animals [42]. These antinutritional effects are most likely caused by some lectins that can impair the integrity of the intestinal epithelium [43] and thus alter the absorption and utilization of nutrients [44]. The administration of lectins to experimental animals can also alter the bacterial flora [45]. Lectin is one of the major proteins found in lentil (*Lens culinaris*). Studies have suggested that lectins affect the immune response against ovalbumin and may promote the development of food allergy to plants containing lectins. Cooking effectively removes trypsin inhibitor and lectin of vegetable peas and significantly reduces protein and amino acid solubility [46]. Lectin can be completely removed from lentil flour after 72 h fermentation at 42 °C with a flour concentration of 79 g / L [47]. Amount of lectin in pulses vary significantly [48]. High level of lectins has been reported in kidney beans (840×10^{-5}hemagglutinating activity units (HU) /kg) and very low amount in cowpea (3×10^{-5} HU /kg). Dietary lectins have generally been considered to be toxic and anti-nutritional factor. However, many lectins are non-toxic, such as those from lentil, pea, chickpea and faba bean. *Vicia faba* agglutinin (VFA), a lectin present in broad bean, aggregated, stimulated the morphological differentiation, and reduced the malignant phenotype of colon cancer cells [49]. Inclusion of raw kidney bean in the diet of obese rats reduced lipid accumulation that was related to a decrease of insulin level caused by lectins. However, no body or muscle protein losses occurred, even at high doses, as with normal rats, suggesting a possible use of lectins as therapeutic agents to treat obesity [50]. Lectin from kidney bean seeds directly inhibits HIV-1 reverse transcriptase, an enzyme crucial for HIV replication, as well as β-glucosidase, which has a role in HIV-1 envelope protein *gp120* processing, therefore a very potent element of the antiretroviral chemotherapy.

Phytosterols

200 different types of phytosterols have been reported in plant species. In pulses, phytosterols are present in small quantities, and the most common phytosterols are β-sitosterol, campesterol, and stigmasterol [51] (Figure **1**). These compounds are also abundant as sterol glucosides and esterified sterol glucosides, with β-sitosterol representing 83% of the glycolipids in defatted chickpea flour [10]. Total phytosterol content detected in the legumes ranged from 134 mg /100 g (kidney bean) to 242 mg /100 g in pea [52]. Total β-sitosterol content ranged from 160 mg /100 g (chickpea) to 85 mg /100 g (butter bean). Chickpea and pea contained high levels of campesterol

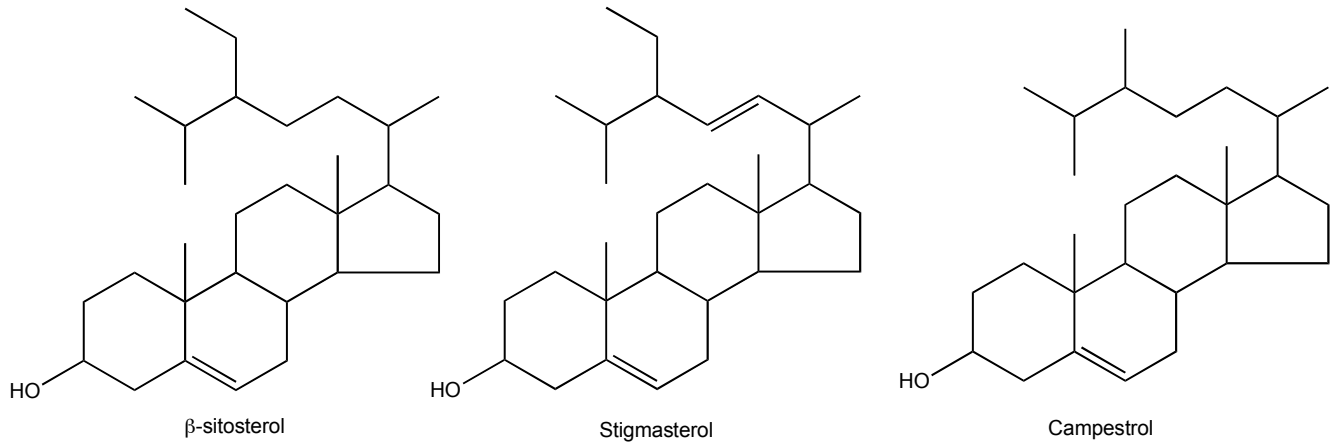

Figure 1: Major sterols reported in Pulse grains.

(21.4 and 25.0 mg /100 g, respectively). Stigmasterol content is higher in butter beans (86 mg /100 g) and squalene content in pea (1.0 mg /100 g). Weihrauch and Gardner (52), reported 127 mg /100 g phytosterol level for kidney bean, with much lower concentration of phytosterols in chickpea (35 mg /100 g). The consumption of pulse grains has been reported to lower serum cholesterol and increase the saturation levels of cholesterol in the bile. A dietary study conducted on humans over a seven week period showed that serum LDL cholesterol was significantly lower during the consumption of a diet consisting of beans, lentil and field pea [53]. The study showed that consumption of pulses lowers LDL cholesterol by partially interrupting the entrohepatic circulation of the bile acids and increasing the cholesterol saturation by increasing the hepatic secretion of cholesterol. The study concluded that other pulse components in the diet may also have contributed to the observed effect; in particular, saponins, which are hydrolyzed by intestinal bacteria to diosgenin, may have exerted a positive effect [54]. Several studies have demonstrated the efficacy of plant sterols and stanols in the reduction of blood cholesterol levels, and plant sterols are increasingly being incorporated into foods for this purpose [55].

Phytic Acid

Phytic acid (myo-inositol hexaphosphate or $InsP_6$), a major phosphorus storage form in plants, and its salts are known as phytates, regulate various cellular functions such as DNA repair, chromatin remodeling, endocytosis, nuclear messenger RNA export and potentially hormone signalling important for plant and seed development [56], as well as animal and human nutrition [57]. It is often regarded as an antinutrient because of strong mineral, protein and starch binding properties thereby decreasing their bioavailability [58]. Phytates play important role in plant metabolism, stress and pathogen resistance in addition to their beneficial effects in human diets by acting as anticarcinogens or by promoting health in other ways such as in decreasing the risk of heart disease or diabetes [59]. Wholegrain cereals and pulses have a high content of phytate [60]. In pulse seeds, phytate is located in the protein bodies in the endosperm. Phytate occurs as a minerals complex, which is insoluble at the physiological pH of the intestine (61). Raw lentil contained 0.3 mmol/kg of $InsP_3$. The most abundant inositol phosphate in raw, dry legume is $InsP_6$, accounting for an average of 83% of the total inositol phosphates, ranging from 77% in chickpea to 88% in black bean. The $InsP_6$ concentration tends to be higher in raw dry bean, blackeye peas, and pigeon peas than in lentils, green and yellow split peas, and chickpeas and ranged between 14.2 and 6 mmol /kg in black beans and chickpeas, respectively [62]. Varietal and agronomic factors, alone and in combination, often result in a wide variation in phytate content of mature legume seeds and cereal grains [63] (Table **3**).

In vivo and *in vitro* studies have demonstrated that inositol hexaphosphate ($InsP_6$, phytic acid) exhibits significant anticancer (preventive as well as therapeutic) properties. It reduces cell proliferation and increases differentiation of malignant cells with possible reversion to the normal phenotype and is involved in host defense mechanism, and tumor abrogation [64]. $InsP_6$ has been suggested to be responsible for the epidemiological link between high-fiber diets (rich in $InsP_6$) and low incidence of some cancers. Phytic acid delays postprandial glucose absorption, reduce the bioavailability of toxic heavy metal such as cadmium and lead, and exhibit antioxidant activity by chelating iron and copper [65]. Dietary and endogenous

Table 3: Trypsin Inhibitor Activity in Grain Legume Seeds

Sr. No.	Crop	Trypsin inhibitor activity (TIU/mg)	Reference
1.	Pea	1-14.6 1.9-6.8 6-15	Bastianelli *et al*., 1998 Gabriel *et al*., 2008 Guillamon *et al*., 2008
2.	Lentil	1-9-2.8 3-8	Wang *et al*., 2009 Guillamon *et al*., 2008
3.	Chickpea (Desi lines) Chickpea (Kabuli lines)	12.7 10.3 15-19	Singh and Jambunathan, 1981 Guillamon *et al*., 2008

phyticacid have protective effects against cancer and heart disease and may be responsible for the cancer-protective effects of high-fibre foods [66]. The anticarcinogenic properties of phytic acid may result from numerous factors, including its ability to chelate metal ions; this depends on the phytate retaining its integrity in the colon [67]. The backbone of most inositol phosphates in cells is *myo-inositol.* Inositol phosphates from seeds are a significant food source of *myo-inositol,* as are the phospholipids and free inositol from many plant- and animal-based foods [68]. *Myo-inositol* has been evaluated for its ability to improve the mental health of patients with various psychiatric disorders [69]. In addition to *myo-inositol,* smaller amounts of *epi-* and *scyllo-inositol are* present in human brains. *Myo-inositol* and $InsP_6$ have synergistic or additive effects in inhibiting the development of cancer [70]. In mice, dietary *myo*inositol has been shown to be effective in preventing cancer of the lung [71], fore stomach [72], liver [73] colon, mammary gland, prostate, and skin [74].

Saponins

Saponins are glycosidic compounds, which are structurally composed of a lipid-soluble aglycon consisting of either a sterol or more commonly a triterpenoid and water soluble sugar residues differing in type and amount of sugars. Their biological activity is closely related to chemical structures that determine the polarity, hydrophobicity and acidity of compounds. Saponins have long been considered undesirable due to toxicity and their haemolytic activity. Although these toxicological properties of plant saponins have long been recognized, there is a renewed interest in these biologically active plant components as recent evidence suggests that saponins possess hypocholesterolemic [75], anti-carcinogenic [76] and immune-stimulatory properties [77]. There is enormous structural diversity within this chemical class, and only a few are toxic [78]. Most of the saponins occur as insoluble complexes with 3-*b*-hydroxysteroids; these complexes interact with bile acid and cholesterol, forming large mixed micelles [75]. In addition, they form insoluble saponin–mineral complexes with iron, zinc, and calcium [79], hence their lower nutrient availability [80]. The most widely studied saponins in legumes include the soyasaponins, which are classified into group A, B, and E saponins on the basis of the chemical structure of the aglycone [81]. Chickpeas contain only one major saponin, belonging to the soyasaponin group B, which is characterized by a reducing sugar 2,3-dihydro-2,5-dihydroxy-6-methyl- H-pyran-4-one (DDMP) moiety on C-22 [82]. The DDMP is a heat-sensitive residue that provides the saponin unique characteristics, including antioxidant capacity, characteristic absorption spectrum and sweet taste. Field peas were initially thought to contain soyasaponin I (S-I) and then soyasaponin VI (S-VI) as the only soyasaponin, but recently field pea extracts were shown to contain dehydrosoyasaponin I (D-I) as a minor component [83]. D-I from pea has insecticidal and anti-feedant properties against stored product insect pests. This triterpenoid Saponin dehydrosoyasaponin I is a natural product present in chickpea and other legumes and is known to be a potent calcium-activated potassium channel opener and as such can be used for treating cardiovascular, urological, respiratory, neurological, and other disorders. Soybean and chickpea constitute major sources of saponins in the human diet [84]. Saponins have been reported in many pulses, lentils [85], and chickpeas [86], as well as in various beans, and peas [78].

Saponin content in chickpea (56 g/kg) is higher than other pulses like green gram (16 g/ kg), lentil (3.7-4.6 g /kg), fababean (4.3 g/ kg) and broadbean (3.5 g/ kg) [87]. Saponin content may vary even among the same species, because of variations in cultivars, varieties, locations, irrigation condition, type of soil, climatic

Table 4: Range of Variation Reported for Phytic Acid in Major Pulses

Sr. No.	Crop	Phytic acid (g/kg)	Reference
1.	Pea	1.3-10.2 0.2-1.3%	Bastianelli *et al.*, 1998 Champ, 2002
2.	Lentil	6.2-8.8 0.15-2.34%	Wang *et al.*, 2009 Champ, 2002
3.	Chickpea (Desi lines) Chickpea (Kabuli lines) Chickpea	7.7-12.3 5.4-11.7 0.4-1.1 %	Chitra *et al.*, 1995 Champ, 2002
4.	Mungbean	10.2-14.8	Chitra *et al.*, 1995
5.	Pigeonpea	6.8-14.9 9.9-16.4	Chitra *et al.*, 1995 Singh, 1999
6.	French bean	0.2-1.9%	Champ, 2002

Table 5: Saponin Content in Major Pulses

Sr. No.	Crop	Saponin (g/kg)	Reference
1.	Pea	1.1 0.3-1.0	Koslovska *et al.*, 2001 Bastianelli *et al.*, 1998
2.	Lentil	1.1	Koslovska *et al.*, 2001
3.	Chickpea	2.3	Koslovska *et al.*, 2001

conditions, and year during which they are grown[88]. Chickpeas, black gram, moth bean, broad beans and peas can contain 3.6, 2.3, 3.4, 3.7, and 2.5 g kg^{-1} dry matter of saponins, respectively [88]. Saponin content in dehulled light and dark coloured peas range from 1.2 to 2.3 g/ kg dry matter [89]. The Saponin content varies from 0.3 to 1.1 g/kg in peas (Table **4**). Some saponin is lost during processing as has been reported in moth beans [88], black gram [90] and pigeon pea [91].

Foods rich in saponins are reported to reduce plasma cholesterol by 16-24 % [92]. The mechanism of cholesterol reduction is by binding to dietary cholesterol or bile acids, thereby increasing their excretion through faeces [93]. β-sitosterol (dominant phytosterol in chickpea) is helpful in decreasing serum cholesterol levels and incidence of coronary heart disease [94]. Recent evidence suggests that legume saponins may possess anti-cancer activity [22] and is beneficial for hyperlipidemia [78]. In addition, they reduce the risk of heart diseases in humans consuming a diet rich in food legumes containing saponins [95]. Epidemiological studies suggest that saponins may play a role in protection from cancer [78]. Metastatic events are critical in cancer proliferation, and there is evidence that glycosylation is an important event in this process. It has recently been demonstrated [96] that soyasaponin I decreases the expression of R-2, 3-linked sialic acid on the cell surface, which in turn suppresses the metastatic potential of melanoma cells. The observed anticancer activity may therefore in part be due to this observed sialyltransferase inhibition activity. Additional mechanistic studies indicate that there is evidence for saponin regulation of the apoptosis pathway enzymes (AKT, Bcl, and ERK1/2), leading to programmed cell death of cancer cells [97]. Research on colon cancer cells suggests that it is the lipophilic saponin cores that may be responsible for the biological activity.

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