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Full Length Research Paper

Mineral contents and antioxidant potential of selected legumes of Pakistan

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Legumes are an important component of the common mans' diet in Pakistan and various species of legumes are grown throughout Pakistan. Little work exists on determination of metal contents and antioxidant activity of pods and leaves of food legumes. Current research is designed to quantify metal contents of leaves and pods of selected legumes and to establish antioxidant profiles of crude methanolic extracts of leaves and pods of said species. Presence of higher amounts of all necessary mineral contents indicates that said legumes may find their place in livestock feed as well as human diet. On the basis of employed analytical methods, the tested legume extracts may be considered easily accessible natural sources of antioxidants and valuable additions to forage for grazing animals due to mineral contents present in their leaves and pods.

Key words: Legumes, pods, leaves, metals, antioxidant, Pakistan.

INTRODUCTION

Leguminosae is 3rd largest family of flowering plants containing 727 genera and 19325 species (Lewis et al., 2005). Legumes grow in varied agrogeoeclimatological conditions and comprise of grain legumes; oilseed, forage, ornamental and medicinal crops as well as many agroforestry species. Legumes are present in human food, animal feed and commercial applications such as soap, paints, resins, coatings, linoleum, cosmetics, pharmaceutical products, timber, tannins, gums, insecticides and molluscicides (Singh et al., 2007). Reasonable consumption of legumes may help to prevent cardiovascular disease, Parkinson's, Alzheimer's, and Huntington's diseases, liver ailments, cancer and diabetes (Andersen et al., 1984; Grusak, 2002; Madar and Stark, 2002; Jenkins et al., 2003; Singh, 2005, 2007). Legumes contain high amounts of protein and dietary fibers, and fewer amounts of cholesterol and saturated fat. One-third of all dietary protein and processed vegetable oil for human consumption is obtained from legumes (Graham and Vance, 2003). Legumes and cereals complement each other in human food and agriculture system in Pakistan particularly in efficient use of water and land resources. Chickpea (desi and kabuli), mash bean, mung bean, lentil, and pea are among major legumes grown in Pakistan. Oxygen is vital for the survival of life, however, during the process of its utilization in normal physiological and metabolic processes, about 5% of it gets reduced to oxygen derived free radicals like superoxide, hydrogen peroxide, hydroxyl and nitric oxide radicals (Halliwell and Gutteridge, 1988; Yu, 1994). These radicals, frequently known as reactive oxygen species (ROS) exert oxidative pressure on human body cells (Lata and Ahuja, 2003). When excess amount of these radicals is not countered by human body defense system, these radicals attack

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Table 1. Mineral contents of leaves.

Plants	Na (g/kg)	Ca (g/kg)	P (g/kg)	K (g/kg)	Mg (g/kg)	Fe (mg/kg)	Cu (mg/kg)	Zn (mg/kg)
Desi chickpea	1 ± 0.03^{b}	16 ± 0.11^{a}	5 ± 0.04^{a}	31 ± 0.07^{a}	4 ± 0.06^{a}	133 ± 0.34^{a}	8 ± 0.45^{a}	124 ± 0.48^{a}
Kabuli chickpea	2 ± 0.07^{ab}	13 ± 0.18^{a}	4 ± 0.01^{b}	37 ± 0.05^{a}	3 ± 0.01^{a}	145 ± 0.63^{a}	10 ± 0.34^{a}	103 ± 0.55^{d}
Lentil	3 ± 0.02^{a}	14 ± 0.05^{a}	5 ± 0.03^{a}	23 ± 0.02^{b}	2 ± 0.04^{b}	121 ± 0.44^{b}	7 ± 0.41^{b}	99 ± 0.74^{d}
Mung bean	4 ± 0.06^{a}	15 ± 0.24^{a}	7 ± 0.11^{a}	27 ± 0.30^{b}	4 ± 0.05^{a}	109 ± 0.25^{c}	6 ± 0.12^{b}	107 ± 0.27^{c}
Mash bean	2 ± 0.05^{ab}	10 ± 0.03^{a}	6 ± 0.17^{a}	25 ± 0.15^{b}	5 ± 0.07^{a}	139 ± 0.21^{a}	11 ± 0.05^{a}	113 ± 0.12^{b}
Pea	5± 0.03^{a}	12 ± 0.02^{a}	4 ± 0.20^{b}	19 ± 0.12^{c}	7 ± 0.03^{a}	139 ± 0.18^{a}	13 ± 0.12^{a}	101 ± 0.05^{d}

Data are expressed as mean ± standard deviation (n = 3), values marked by the different letters in same column differ significantly (P ≤ 0.05).

Table 2. Mineral contents of pods.

Plants	Na (g/kg)	Ca (g/kg)	P (g/kg)	K (g/kg)	Mg (g/kg)	Fe (mg/kg)	Cu (mg/kg)	Zn (mg/kg)
Desi chickpea	19 ± 0.12^{c}	207 ± 0.33^{a}	152 ± 0.05^{b}	196 ± 0.45^{a}	167 ± 0.25^{a}	8 ± 0.09^{a}	11 ± 0.04^{a}	24 ± 0.22^{b}
Kabuli chickpea	23 ± 0.29^{b}	198 ± 0.27^{b}	164 ± 0.23^{a}	173 ± 0.63^{b}	135 ± 0.12^{d}	9 ± 0.12^{a}	13 ± 0.06^{a}	39 ± 0.47^{a}
Lentil	31 ± 0.22^{a}	189 ± 0.18^{c}	157 ± 0.66^{ab}	182 ± 0.88^{a}	149 ± 0.08^{c}	6 ± 0.04^{a}	17 ± 0.11^{a}	29 ± 0.44^{b}
Mung bean	17 ± 0.09^{c}	204 ± 0.56^{a}	155 ± 0.32^{b}	187 ± 0.74^{a}	154 ± 0.33^{b}	9 ± 0.23^{a}	16 ± 0.29^{a}	27 ± 0.07^{b}
Mash bean	26 ± 0.38^{b}	210 ± 0.47^{a}	160 ± 0.45^{a}	176 ± 0.66^{b}	163 ± 0.26^{a}	7 ± 0.16^{a}	12 ± 0.07^{a}	33 ± 0.21^{b}
Pea	15 ± 0.04^{c}	212 ± 0.11^{a}	166 ± 0.31^{a}	169 ± 0.38^{c}	157 ± 0.11^{b}	10 ± 0.05^{a}	14 ± 0.10^{a}	31 ± 0.29^{b}

Data are expressed as mean ± standard deviation (n = 3), values marked by the different letters in same column differ significantly (P ≤ 0.05).

biomolecules like proteins, lipids, DNA and carbohydrates leading to a number of physiological disorders. Antioxidants defend living systems against harmful effects of these radicals. Similarly, abundance or shortage of certain metals present in plants may be harmful to human health. Presence of these metals up to acceptable limits is essential for normal human physiobiochemical functions. Usually presence of metals in plants depends on cultivars, agrogeoclimatological conditions, maturity and collection time as well as plant tissue, soil, water and fertilizers compostion along with permissibility, selectivity and absorbility of plants for the uptake of these metals. The focus of the scientific community has been directed to antioxidant components and various constituents such as amino acids, proteins, oil constituents, trace elements, and sugars of legumes (Zia-Ul-Haq et al., 2007a, b, 2008a, b, 2009, 2010, 2011a, b, c, d, e, 2012). We have evaluated metal contents and employed a battery of antioxidant capacity assays to evaluate the antioxidant potential of some commonly consumed legumes in Pakistan.

MATERIALS AND METHODS

Plant material (leaves and pods) of desi and kabuli chickpea (*Cicer arietinum* L.), lentil (*Lens culinaris* Medik), mung bean (Vigna radiate (L.) Wilczek), mash bean (*Phaseolus mungo*) and pea (*Pisum sativum* L.) was obtained from Department of Agronomy, Bahauddin Zakariya University, Multan, Pakistan.

Determination of metal contents

Plant material (leaves and pods) was washed with tap water, followed by distilled water containing a nonionic detergent, and finally three rinses of distilled water, and then were oven-dried at 60°C for 48 h. It was then milled, ground, ashed at 45°C, and digested in 10 ml 1 mol/L HCl. Among major minerals, K was measured by flame photometry and P colorimetrically by the molybdovanadate method (Kitson and Mellon 1944). Ca, Mg and trace minerals (Fe, Cu and Zn) were measured by atomic absorption spectrometry (AA680, Shimadzu, Japan). Standardized procedures for measuring the mineral element concentrations followed the guidelines of the (AOAC, 1990). The concentrations are expressed on dry matter basis (Ayaz et al., 2007; Ibrikci et al., 2003) (Tables 1 and 2).

Extraction

The plant material (leaves and pods) was crushed to coarse powder separately with help of pestle and mortar, and macerated with aqueous methanolic mixture (80:20; v/v, 1 L) at room temperature for fifteen days with occasional shaking. The extracts obtained were filtered through filter paper under vacuum and concentrated under reduced pressure in a rotary evaporator (model Q-344B – Quimis, Brazil) using a warm water bath (model Q-214M2 - Quimis, Brazil) to obtain a thick gummy mass, which was further dried in a desiccator and stored in air-tight vial till further use. Methanol, catechin, gallic acid, Folin and Ciocalteau's reagent, ferric chloride, 2,2-diphenyl-1-picrylhydrazyl radical (DPPH$^{\bullet}$), butylated hydroxyl toluene (BHT), and L-Ascorbic acid were obtained from Sigma (St Louis, MO), USA. All the chemicals used were of analytical grade.

Determination of total phenolic (TPC) and flavonoid (TFC) contents

Extracts were assayed for total phenolic (TPC) and flavonoid (TFC) contents by following previously reported methods (Jia et al., 1999;

Table 3. Total phenol content and total flavonoid content of legume leaves.

Plant	TPC (mgGAE/g)	TFC (mgCAE/g)
Desi chickpea	2.91 ± 0.34[a]	0.99 ± 0.39[a]
Kabuli chickpea	1.52 ± 0.59[c]	0.65 ± 0.51[a]
Lentil	1.04 ± 0.18[d]	0.36 ± 0.39[b]
Mung bean	2.17 ± 0.35[a]	0.89 ± 0.16[a]
Mash bean	2.65 ± 0.17[a]	0.68 ± 0.19[a]
Pea	1.79 ± 0.09[b]	0.74 ± 0.15[a]

Data are expressed as mean ± standard deviation (n = 3), values marked by the different letters in same column differ significantly (P ≤ 0.05).

Table 4. Total phenol content and total flavonoid content of legume pods.

Plant	TPC (mgGAE/g)	TFC (mgCAE/g)
Desi chickpea	3.51 ± 1.62[a]	1.29 ± 0.19[a]
Kabuli chickpea	2.09 ± 0.59[b]	1.07 ± 0.23[b]
Lentil	1.79 ± 0.11[b]	1.43 ± 0.41[a]
Mung bean	2.91 ± 0.53[b]	1.62 ± 0.13[a]
Mash bean	3.89 ± 0.91[a]	1.58 ± 0.27[a]
Pea	2.62 ± 0.64[b]	1.72 ± 0.11[a]

Data are expressed as mean ± standard deviation (n = 3), values marked by the different letters in same column differ significantly (P ≤ 0.05).

Heimler et al., 2005; Zia-Ul-Haq et al., 2008a) respectively and results were expressed as gallic acid equivalents mg (GAE)/g and as mg catechin equivalents (CAE)/g (Tables 3 and 4).

DPPH radical scavenging assay

This spectrophotometric assay uses the stable radical 2, 2-diphenyl-1-picrylhydrazyl (DPPH) as a reagent (Amarowicz et al., 2004). An aliquot of the sample (100 µl) was mixed with ethanol (1.4 ml) and then added to 0.004% DPPH (1 ml) in ethanol. The mixture was shaken vigorously and then immediately placed in a UV-Vis spectrophotometer (UNICO, Shanghai, China) to monitor the decrease in absorbance at 517 nm. Monitoring was continued for 70 min until the reaction reached a plateau. The radical-scavenging activities of samples, expressed as percentage inhibition of DPPH, were calculated according to the formula:

$$\text{Inhibition (\%)} = \frac{A_0 - A_1}{A_0} \times 100$$

where A_0 = absorbance of the control and A_1= absorbance of the extract. Scavenging activity was compared with natural antioxidants like ascorbic acid (Yen and Duh, 1994) (Tables 5 and 6).

β-Carotene-linoleic acid test

Antioxidant activity of the samples was determined using the β-carotene-linoleic acid test (Taga et al., 1984). β-carotene (10 mg) was dissolved in chloroform (10 ml). The carotene–chloroform solution (0.2 ml) was pipetted into a boiling flask containing linoleic acid (20 mg) and Tween-40 (200 mg). Chloroform was removed using a rotary evaporator at 40°C for 5 min, and distilled water (50 ml) was added to the residue slowly with vigorous agitation, to form an emulsion. A portion of the emulsion (5 ml) was added to a tube containing the sample solution (0.2 ml) and the absorbance was immediately measured at 470 nm against a blank, consisting of an emulsion without β-carotene. The tubes were placed in a water bath at 50°C and the oxidation of the emulsion was monitored spectrophotometrically by measuring absorbance at 470 nm over a 60 min period. Control samples contained 200 µl of water while BHT was used as a reference. The antioxidant activity was expressed as inhibition percentage with reference to the control after a 60 min incubation using the following equation:

$$AA = 100 \times \frac{DRC - DRS}{DRC}$$

where AA = antioxidant activity; DRC = degradation rate of the control; DRS = degradation rate in presence of the sample (Wu et al., 2009) (Tables 5 and 6).

Superoxide radical scavenging potential

1 ml of 156 mM nitro blue tetrazolium (pH = 7.4), 1 ml of 468 mM NADH solution (pH = 7.4), and 1 ml sample solution of extract were mixed. To initiate reaction 100 ml of 60 mM phenazine methosulfate (pH 7.4) were added to the mixture. The reaction mixture was incubated at 25°C for 5 min and the absorbance was measured at 560 nm against blank sample (NBT and NADH) and compared with standards. Decreased absorbance of reaction mixture was indicative of increased superoxide anion scavenging activity. Following formula was used to calculate % inhibition of superoxide anion generation:

Table 5. *In vitro* antioxidant activity of leaves by various assay IC_{50} (µg/ml).

Plants	DPPH radical scavenging activity	β-carotene-linoleic acid test	Superoxide radical scavenging activity
Desi chickpea	407.05 ± 1.25[b]	423.25 ± 1.16[c]	168.39 ± 1.12[b]
Kabuli chickpea	490.11 ± 1.86[b]	503.04 ± 1.23[a]	206.02 ± 1.37[a]
Lentil	539.31 ± 1.04[a]	561.67 ± 1.39[a]	239.14 ± 1.34[a]
Mung bean	456.71 ± 1.62[b]	479.27 ± 1.55[b]	196.07 ± 1.52[a]
Mash bean	432.89 ± 1.73[b]	461.68 ± 1.81[b]	177.35 ± 1.43[b]
Pea	507.44 ± 1.03[a]	529.84 ± 1.73[a]	247.63 ± 1.88[a]
Ascorbic acid	192.13 ± 1.38[c]	-	99.04 ± 1.17[c]
BHT	-	175.27 ± 2.33[d]	-

Data are expressed as mean ± standard deviation (n = 3), values marked by the different letters in same column differ significantly (P ≤ 0.05).

Table 6. *In vitro* antioxidant activity of pods by various assays.

Plants	DPPH radical scavenging activity	β-carotene-linoleic acid test	Superoxide radical scavenging activity
Desi chickpea	367.21 ± 1.09[a]	341.36 ± 1.33[c]	137.12 ± 1.44[c]
Kabuli chickpea	432.108 ± 1.32[a]	466.23 ± 1.84[a]	176.33 ± 1.06[b]
Lentil	465.51 ± 1.24[a]	503.44 ± 1.39[a]	205.05 ± 1.01[a]
Mung bean	389.22 ± 1.31[a]	417.16 ± 1.03[b]	175.04 ± 1.62[b]
Mash bean	401.44 ± 1.85[a]	404.73 ± 1.98[b]	163.17 ± 1.22[b]
Pea	457.13 ± 1.21[a]	477.42 ± 1.54[a]	203.52 ± 1.29[a]
Ascorbic acid	192.13 ± 1.38[b]	-	99.04 ± 1.17[d]
BHT	-	175.27 ± 2.33d	-

Data are expressed as mean ± standard deviation (n = 3), values marked by the different letters in same column differ significantly (P ≤ 0.05).

$$\text{Inhibition (\%)} = \frac{A_0 - A_1}{A_0} \times 100$$

where A_0 = absorbance of the control (blank, without extract) and A_1 = absorbance in the presence of the extract (Ilhami et al., 2005) (Tables 5 and 6).

RESULTS AND DISCUSSION

Besides their nutritional repute, metals have both healing and protective role. Metals investigated in pods and leaves indicated that calcium, potassium, phosphorus and magnesium were present in highest contents while iron and zinc were major trace elements present in both leaves and pods of investigated species. Sodium and potassium endure acid-base balance and nerve transmittance. Calcium and phosphorus are necessary for development and appropriate functioning of bones muscles and teeth. Iron is compulsory for hemoglobin and myoglobin synthesis (Saleh-e-in et al., 2008). Sodium, potassium and calcium also impact heart functions. Calcium increases heart shrinkage (Rajurkar et al., 1997). Many metabolic processes require magnesium, iron and potassium. Zinc plays central role in many biochemical and immunological functions (Fell and Lyon, 1994; Dell and Sunde, 1997). It is apparent that the investigated species are meaningful sources of all necessary elements in the human diet. Our results (Tables 1 and 2) are close to those reported earlier for related species (Ibrikci et al., 2003; Mateos-Aparicio et al., 2010). Phenolic acids presence in food and feed commodities was considered as negative earlier as they were doubted to decrease nutrients availability de-creasing nutritional importance. However, now it is proved that polyphenolics presence in food is important for oxidative stability and antimicrobial properties (Carbonaro et al., 2002; Floridi et al., 2003). Phenolics possess a wide spectrum of biochemical activities like anticancer, antioxidant and gene-modifying capacities. Similarly, several studies indicate a significant correlation between high dietary intake of flavonoids and reduction of cardiovascular and carcinogenic risk (Marinova et al., 2005). Our results indicated that all tested extracts had sufficient amount of phenolic and flavonoid contents. Highest phenolic contents were observed in mash bean

Table 7. Correlation coefficient of various antioxidant assays.

Assay	TPC	DPPH radical scavenging activity	β-carotene-linoleic acid test	Superoxide radical scavenging activity
TPC	-	0.242	0.361	0.451
DPPH radical scavenging activity	0.242	-	0.987	0.946
β-carotene-linoleic acid test	0.361	0.987	-	0.971
Superoxide radical scavenging activity	0.451	0.946	0.971	-

(3.89a ± 0.91), desi chickpea (3.51a ± 1.62) and mung bean (2.91b ± 0.53) pods. Similarly, pods of these three species also contained highest amounts of flavonoids. To gauge antioxidant potential, battery of assays is commonly used. In the current study, we also used three top-benched antioxidant assays, DPPH radical scavenging activity, β-carotene-linoleic acid test and superoxide radical scavenging activity to evaluate antioxidant potential of these legumes. IC_{50} values observed indicated both organs, that is, pods and leaves as sufficient sources of antioxidants. Superoxides are produced from molecular oxygen due to oxidative enzymes of body as well as via non-enzymatic reaction such as auto-oxidation by catecholamines (Hemmani and Parihar, 1998). The results (Tables 5 and 6) showed that the potency of legume leaves and pods extract against superoxide radical scavenging activity in the model system. There are similar reports for other plants that are carried out in different systems (Akinmoladun et al., 2010). The apparent mechanism of scavenging the superoxide anions may be due to the occurrence of phenolic compounds that are present in the extracts and their uptake of generated superoxide in *in vitro* reaction mixture.

The results of the different antioxidant assays used in the present study of different extracts were compared and correlated with each other and results represented in Table 7. The content of total phenolics (TFC) showed good correlation with most of the antioxidant assays, such as DPPH radical scavenging activity (r = 0.242), β-carotene-linoleic acid test (r = 0.361), Superoxide radical scavenging activity (r = 0.451). Many scientists have reported outstanding linear correlations between antioxidant activity tests and total phenolic content (Sultana et al., 2007; Oliveira et al., 2009). There was also good relation among different antioxidant assays. DPPH radical scavenging assay showed close relation with superoxide anion scavenging activity (r = 0.946). This may be due to the reason that many other compounds such as carotenoids, tocopherol and vitamin C other than total phenols and total flavonoids also contribute to antioxidant activity (Mccune and Johns, 2002).

REFERENCES

Akinmoladun AC, Obuotor EM, Farombi EO (2011). Evaluation of antioxidant and free radical scavenging capacities of some Nigerian indigenous medicinal plants. J. Med. Food, 13(2): 444-51.

Amarowicz R, Pegg RB, Moghaddam PR, Barl B, Weil JA (2004). Free-radical scavenging capacity and antioxidant activity of selected plant species from Canadian prairies. Food Chem., 84: 551-562.

Andersen JW, Story L, Sieling B, Chen WJL, Petro MS, Story J (1984). Hypocholesterolemic effects of oat-bran or bean intake for hypercholesterolemic men. Am. J. Clin. Nutr., 40: 1146-1155.

Ayaz FA, Torun H, Ayaz S, Correia PJ, Alaiz M, Sanz C, Grúz J, Strnad M (2007). Determination of chemical composition of anatolian carob pod (*Ceratonia siliqua* L.): sugars, amino and organic acids, minerals and phenolic compounds. J. Food Qual., 30: 1040-1055.

Carbonaro M, Mattera M, Nicoli S, Bergamo P, Cappelloni M (2002). Modulation of antioxidant compounds in organic vs conventional fruit (peach, *Prunus persica* L., and pear, *Pyrus communis* L.). J. Agric. Food Chem., 50: 5458-5462.

Dell BL, Sunde RA (1997). Handbook of nutritionally essential mineral elements. Marcell Dekker Inc: New York, USA, pp. 68.

Fell GS, Lyon TDB (1994). Trace element analysis in biological specimens. In: Herber R. F. M. and M. Stoeppler (Eds.) Amsterdam: Elsevier, pp. 541.

Floridi S, Montanari L, Marconi O, Fantozzi P (2003). Determination of free phenolic acids in wort and beer by coulometric array detection. J. Agric. Food Chem., 51(6): 1548-1554.

Graham PH, Vance CP (2003). Legumes, importance and constraints to greater use. Plant Physiol., 131: 872–877.

Grusak MA (2002). Phytochemicals in plants: genomics-assisted plant improvement for nutritional and health benefits. Curr. Opin. Biotechnol., 13: 508-511.

Halliwell B, Gutteridge JM (1988). In Free radicals in biology and medicine. 2nd ed, Clarendon Press, Oxford, UK.

Heimler D, Vignolini P, Dini MG, Romani A (2005). Rapid tests to assess the antioxidant activity of *Phaseolus vulgaris* L. dry bean. J. Agric. Food Chem., 53: 3053-3056.

Hemmani T, Parihar MS (1998). Reactive oxygen species and oxidative DNA damage. Indian J. Physiol. Pharmacol., 42: 440.

Ibrikci H, Sharon JB, Knewtson, Grusak MA (2003). Chickpea leaves as a vegetable green for humans: evaluation of mineral composition. J. Sci. Food Agric., 83: 945-950.

Ilhami GI, Haci AA, Mehmet C (2005). Determination of *in vitro* antioxidant and radical scavenging activities of Propofol . Chem. Pharm. Bull., 53: 281-285.

Jia Z, Tang M, Wu J (1999). The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. Food Chem., 64: 555-559.

Jenkins DJA, Kendall CWC, Marchie A, Jenkins AL, Augustin LSA, Ludwig DS, Barnard ND, Anderson JW (2003). Type 2 diabetes and the vegetarian diet. Am. J. Clin. Nutr., 78: 610S-616S.

Lata H, Ahuja GK (2003). Role of free radicals in health and disease. Indian J. Phys. Allied Sci., 57: 124.

Lewis G, Schrire MB, Lock M (2005). Legumes of the world. Royal Botanical Garden, Kew Richmond, Surrey, UK.

Madar Z, Stark AH (2002). New legume sources as therapeutic agents. Br. J. Nutr., 88: S287-S292.

Marinova D, Ribarova F, Atanassova M (2005). Total phenolics and total flavonoids in bulgarian fruits and vegetables. J. Univ. Chem. Tech. Met., 40(3): 255-260.

Mateos-Aparicio I, Redondo-Cuenca A, Villanueva-Suárez M, Zapata-Revilla M, Tenorio-Sanz M (2010). Pea pod, broad bean pod and

okara, potential sources of functional compounds. LWT - Food Sci. Tech., 43:1467-1470.
Oliveira I, Lopes VP, Andrade R, Bento PB, Seabra RA, Pereira JA (2009). Phytochemical characterization and radical scavenging activity of *Portulaca oleracea* L. leaves and stems. Microchem. J., 92(2): 129-134.
Rajurkar NS, Damame MM (997). Elemental Analysis of some herbal plants used in the treatment of cardiovascular diseases by NAA and AAS. J. Radioanal. Nucl.Chem., 219: 77-80.
Saleh-e-in MM, Sultana A, Hossain MA, Ahsan M, Roy SK (2008). Macro and micro elemental analysis of *Anethum sowa* L. (Dill) stem by X-ray fluorescence spectrometry. Bangladesh. J. Sci. Ind. Res., 43: 483-494.
Singh RJ, Chung GH, Nelson RL (2007). Landmark research in legumes. Genome, 50: 525-537.
Singh R. J (2005). Landmark research in grain legumes. In: Genetics resources, chromosome engineering and crop improvement series: grain legumes, Vol. 1. Edited by Singh RJ, Jauhar PP. CRC Press, Inc., Boca Raton, Fla. pp. 1-9.
Singh RJ (2007). Landmark research in oilseed crops. In: Genetics resources, chromosome engineering and crop improvement series: oilseed crops, Vol. 4. Edited by R. J. Singh. CRC Press, Inc., Boca Raton, Fla. pp. 1-12.
Sultana B., Anwar F, Przybylski R (2007). Antioxidant activity of phenolic components present in barks of *Azadirachta indica*, *Terminalia arjuna*, *Acacia nilotica*, and *Eugenia jambolana* Lam. trees. Food Chem., 104: 1106-1114.
Taga MS, Miller EE, Pratt DE (1984). Chia seeds as a source of natural lipid antioxidant. J. Am. Oil Chem. Soc., 61: 928-9.
Wu N, Fu K, Fu Y, Zu Y, Chang F, Chen Y, Liu X, Kong Y, Liu W, Gu C (2009). Antioxidant activities of extracts and main components of pigeonpea [*Cajanus cajan* (L.) Millsp.] leaves. Molecule, 14: 1032-1043.
Yen GC, Duh PD (1994). Scavenging effect of methanolic extracts of peanut hulls on free-radical and active-oxygen species. J. Agric. Food Chem., 42: 629-632.
Yu BP (1994). Cellular defenses against damage from reactive oxygen species. Physiol. Rev., 74: 139.
Zia-Ul-Haq M, Iqbal S, Ahmad S, Imran M, Niaz A, Bhanger MI (2007a). Nutritional and compositional study of desi chickpea (*Cicer arietinum* L.) cultivars grown in Punjab. Pakistan. Food Chem., 105: 1357-1363.
Zia-Ul-Haq M, Ahmad M, Iqbal S, Ahmad S, Ali H (2007b). Characterization and compositional studies of oil from seeds of desi chickpea (*Cicer arietinum* L.) cultivars grown in Pakistan. J. Am. Oil Chem. Soc., 84: 1143-1148.
Zia-Ul-Haq M, Iqbal S, Ahmad M (2008a). Characteristics of oil from seeds of 4 mungbean (*Vigna radiata* L. wilczek) cultivars grown in Pakistan. J. Am. Oil Chem. Soc., 85: 851-856.
Zia-Ul-Haq M, Iqbal S, Ahmad S, Bhanger MI, Wiczkowski W, Amarowicz R (2008b). Antioxidant Potential of Desi Chickpea varieties commonly consumed in Pakistan. J. Food Lipid, 15: 26-342.
Zia-Ul-Haq M, Ahmad S, Ahmad M, Iqbal S, Khawar KM (2009). Effects of cultivar and row spacing on tocopherol and sterol composition of chickpea (*Cicer arietinum* L) seed oil. Tarim Bilim. Der., 15: 25-30.
Zia-Ul-Haq M, Ahmad S, Chiavaro E, Mehjabeen, Ahmed S (2010). Studies of oil from cowpea (*Vigna unguiculata* (l) walp.) cultivars commonly grown in Pakistan. Pakistan. J. Bot., 42(2): 1333-1341.
Zia-Ul-Haq M, Ahmad S, Shad MA, Iqbal S, Qayum M, Ahmad A, Luthria DL, Amarowicz R (2011a). Compositional studies of some of lentil cultivars commonly consumed in Pakistan. Pakistan. J. Bot., 43(3): 1563-1567.
Zia-Ul-Haq M, Ahmad M, Mehjabeen, Jehan N, Ahmad S, Qayum M, Marwat IK (2011b). Antimicrobial screening of selected flora of Pakistan**.** Arch. Biol. Sci., 63(3): 691-695.
Zia-Ul-Haq M, Nisar M, Shah M R, Akhter M, Qayum M, Ahmad S, Shahid S A, Hasanuzzaman M (2011c).Toxicological screening of some selected legumes seed extracts. Legume Res., 34(4): 242-250.
Zia-Ul-Haq M, Ćavar S, Qayum M, Imran I, Feo V (2011d). Compositional studies: antioxidant and antidiabetic activities of *Capparis decidua* (Forsk.) Edgew. Int. J. Mol. Sci., 12(12): 8846-8861.
Zia-Ul-Haq M, Ahmad S, Iqbal S, Luthria DL, Amarowicz R (2011e). Antioxidant potential of lentil cultivars. Oxid. Commun., 34: 819-831.
Zia-Ul-Haq M, Shahid SA, Ahmed S, Ahmad S, Qayum M, Khan I (2012). Anti-platelet activity of methanolic extract of *Grewia asiatica* L. leaves and *Terminalla chebula* Retz. Fruits. J. Med. Plant Res., 6: 139-143.