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Effect of Varieties on Bioactive Properties and Mineral Contents of Some Sorghum, Millet and Lupin Seeds

Fahad Al Juhaimi[1]*, Şenay Şimşek[2], Kashif Ghafoor[1], Elfadil E. Babiker[1], Mehmet Musa Özcan[3]*, Isam A Mohamed Ahmed[1], and Omer Alsawmahi[1]

[1] *Department of Food Science & Nutrition, College of Food and Agricultural Sciences, King Saud University, Riyadh, SAUDI ARABIA*
[2] *Department of Plant Sciences, Cereal Science Technology, North Dakota State University, Fargo, US*
[3] *Department of Food Engineering, Faculty of Agriculture, Selçuk University, 42079 Konya, TURKEY*

Abstract: In this study, some physico-chemical properties, amino acids, fatty acids, sugars and mineral contents of sorghum, millet and lupin seeds. Sorghum (red, white and yellow) and millet seeds were purchased from market in Saudi Arabia (Riyadh). Lupin seeds were provided from in Turkey (Konya). Protein contents of seed samples ranged from 8.6% (yellow sorghum) to 37.7% (lutop) ($p < 0.05$). The extractable phenolics contents for gallic acid equivalent (GAE) of grains ranged between 1.43 mgGAE/g (white sorghum) to 8.23 mgGAE/g (red sorghum), and hydrolysable phenolics contents for GAE of grains varied between 1.48 mgGAE/g (white sorghum) to 26.10 mgGAE/g (red sorghum ($p < 0.05$). Total phenol contents of seeds were found between 2769 mg GAE/g (bablon) to 6087 mgGAE/g (yellow sorghum) ($p < 0.05$). Amino acid contents of millet changed between 0.02% (ornithine) and 2.07% (glutamic acid), while amino acid contents of yellow sorghum range from 0.02% (hydroxyproline) to 1.71% (glutamic acid), amino acid values of white sorghum changed between 0.02% (hydroxyproline) and 2.21% (glutamic acid), amino acid values of lutop seed changed between 0.02% (ornithine) and 6.77% (glutamic acid) ($p < 0.05$).While the oleic acid contents change between 25.27% (white sorghum) and 53.50% (Bablone), linoleic acid contents ranged from 14.60% (Bablone) to 42.67% (Millet) ($p < 0.05$). However, the amount of potassium in the seeds varied between 1831.34 mg/kg (while sorghum) and 11895.8 mg/kg (Lutop). Generally, protein, oleic acid, amino acid and mineral contents of lupin varieties were higher as compared to those of millet phenol, anthocyanin and sorghum seeds.

Key words: seeds, bioactive properties, phenolics, amino acid, fatty acid

1 Introduction

Sorghum is one of the most important cereal grain and ranks fifth after wheat, rice, maize and barley. The kernel of sorghum varies in colour, shape, size and certain anatomical components[1)]. The colour of sorghum grains varies from white to dark brown depending on the phenolic pigments present. Anthocyanogens have been detected in yellow millo and red kafir sorghum but not in white waxy or yellow endosperm, varieties[2)]. Nutritionally, sorghum and other cereal proteins are deficient in essential amino acids such as lysine, tryptophan and threonine[1)]. Food grains are the major ingredients used for producing various food products such as oil, starch and glucose[3, 4)]. Lupin belongs to *Leguminosae* family, and is an important legume used for the production of both human and animal foods since the ancient times[5)]. However, a significant reduction in objectionable compounds such as tannins, alkaloids and oligosaccharides is essential before its consumption[6, 7)]. The use of lupin oil as food ingredient is due to the presence of oil in varieties *Lupinus albus* and *Lupinus mutabilis* at levels around 110 and 190 g/kg respectively in the varieties[7)]. Lupin flour is being used as a foaming agent in food instead of egg albumin[8)]. Several studies have reported about the chemical composition and nutritive value of sorghum, millets and lupin seeds originating from different geographical regions around the globe[1, 9)]. The aim of this study was to determine some physico-chemical properties, amino acids, fatty acids, sugars and mineral contents

*Correspondence to: Fahad Y. AL-juhaimi, Department of Food Science & Nutrition, College of Food and Agricultural Sciences, King Saud University, Riyadh, SAUDI ARABIA
E-mail: faljuhaimi@ksu.edu.sa
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of sorghum, millet and lupin seeds.

2 Material and Methods

2.1 Material

Sorghum (red, white and yellow) and millet seeds were purchased from market in Riyadh in Saudi Arabia. Lupin seeds (Bablon, Deşdiğin and Bablone cv) used were supplied by herbarium in the Department of Field Crops, Faculty of Agriculture, Selcuk University in Turkey. The seeds were cleaned, sorted and dried in other to remove all dirts. The pericarps were removed manually from the seeds and the seeds were then packed in polypropylene bags and stored in refrigerator.

2.2 Methods

2.2.1 Proximate analysis

The moisture, protein and ash content of the samples were determined according to AACC methods 44-15.02, 08-01.01 and 46-30.01. The moisture content was determined by heating the samples in a gravity oven for 1 hour at 135℃. The ash content was estimated by heating the sample in a muffle furnace at 525℃ for approximately 16 hours. The crude protein content of the samples were measured using nitrogen combustion method according to AACC International[10] method. Leco combuston analyzer was used to determine the protein content of the samples and conversion factor of 6.25 was used. Total starch was measured using AACC approved method 76-13.01, using an assay kit from Megazyme (Megazyme International, Ireland). The samples were incubated with heat stable fungal α-amylase and amyloglucosdiase to hydrolyze the starch to glucose. The glucose was treated with glucose oxidase/peroxidase (GOPOD) and the absorbance was read at 492 nm. The starch content was calculated based on the glucose concentration[11]. Total fat content was determined according to AOCS method Ba 3-38. Soxhlet apparatus was used to extract fat from the millet sample for 3 hours and n-Hexane was used as the solvent[12].

2.2.2 Extractable polyphenols

The extraction was done by continuous shaking using methanol. Acidified water (50:50 v/v, pH 2, 50 mL/g sample, 60 min, room temperature) with HCL and acetone: water 70:30 v/v, 50 mL/g sample, 60 min, room temperature) were used. Samples were centrifuged (15 min, 25℃, 3000 g) and combined supernatants were analyzed for extractable polyphenols. Ferulic acid and gallic acid were used to prepare standard curves. Extractable polyphenols were determined by the Folin-Ciocalteu procedure[13] and quantified as miligram ferulic acid equivalents per gram (mg FAE/g) and milligram gallic acid equivalents per gram (mg GAE/g).

2.2.3 Hydrolysable polyphenols

Acidic hydrolysis can be used to extract tannis, hydroxycinnamic acids and phenolic acids from food materials composed of polyphenols. Tannis, hydroxycinnamic acids and phenolic acids were extracted from the samples using methanol/H_2SO_4 hydrolysis for 20 h 85℃ using the residues of material used for the determination of soluble polyphenols[14], centrifugation (3000 × g) for 15 min at 25℃ was done after extraction. Folin Ciocalteu method using ferulic acid standard curve were combined with the supernatant[13] was used for the hydrolysable polyphenols analysis which were presented as miligram ferulic acid equivalents per gram (mg FAE/g) and milligram gallic acid equivalents per gram (mg GAE/g).

2.2.4 Sugar composition

The free sugar composition was determined by chromatography technique according to Kakehi and Honda[15].

2.2.5 Amino acid analysis

Amino acid profile analysis was carried out using AOAC Official Method 982.30 E (a,b,c) described by International AOAC *et al.*[16]. Acid hydrolysis, performic acid oxidation followed by acid hydrolysis and alkaline hydrolysis was conducted prior to amino acid analysis. Samples were hydrolyzed with 6M HCl after freezing in a dry ice-alcohol bath under vacuum. The samples were heated at 110℃ for 24 hours before filtering through Whatman No. 1 filter paper. Then the sample is rinsed with water three times and filtered in between each rinse. Cold performic acid was added to a second portion of sample and held overnight at 0-5℃ prior to adding cold HBr and 1-octanol. The sample was mixed in an ice-water bath and evaporated to dryness at 40℃ under vacuum. HCl was then added and hydrolysis was done as stated above. A third portion of sample was added to a tube having a Nalgene polypropylene centrifuge tube liner. Potato starch was added to samples with low starch content, and fresh 4.2M NaOH and 1-octanol was added. The sample was mixed under partial vacuum prior to freezing in a dry ice-alcohol bath. The third portion of sample was then hydrolyzed for 22 hours at 110℃ and then cooled before transferring to a 5 ml volumetric flask containing 6M HCl. Each hydrolysate was analyzed in an amino acid analyzer.

2.2.6 Fatty acid determination

The fatty acid profile determination was done using AOAC methods 996.06, 965.49 and 969.33 as described by International AOAC *et al.*[16]. They were analysed gas chromatography (HP 6890) equipped with flame-ionization detector (FID) and capillary column (60 m × 0.25 mm i.d.; film thickness 0.20 micrometere). The temperature of injection block and dedector was 260℃. It was operated under the following conditions: oven temperature program. 175℃ for 7 min. Raised to 250℃ at a rate 5℃/min and than kept at 250℃ for 15 min); injector and detector temperatures, 250 and 250℃; respectively, carrier gas. nitrogen at

flow rate of 1.51 mL/min; split ratio. 1/50 μL/min.

2.2.7 Total phenolic, flavonoid and anthocyanin contents determination

Quantification of anthocyanins was based on the method of Ticconi *et al.*[17]. Propanol, chlorhydric acid, and water in ratio 18:1:81 in solution was homogenized with 0.5 g of the sample. The homogenates that result were heated in water bath for 3 min, allowed to boil and then incubated for 24 h in a dark room at room temperature. The supernatants (3 mL each) were centrifuged for 40 min at 6500 rpm and the absorbance was recorded at 535 and 650 nm. The phenolic content of the samples were extracted using methanol and quantified with spectrophotometer absorbance at 765 nm using Folin-Ciocalteu reagent (FCR) as described by Madaan *et al.*[18]. Gallic acid standard curve was constructed and used to evaluate the total phenolic, which was expressed using gallic acid equivalent. 10 mg of gallic acid and 100 mL of having 50% methanol μg/mL were mixed, followed by dilution in concentration of 12.5, 25, 50 or 100 μg/mL. An aliquot of 0.076 mL from each of this dilution was kept in a glass tube and diluted further to a final volume of 0.76 mL using distilled water. About 0.12 nL of Folin-Ciocalteu reagent (1 N) was added and the samples were held at room temperature for 5 min incubation after which 0.32 mL of Na_2CO_3 (20% w/w) was added and total volume in test tubes made 2 mL each using distilled water. The samples and standard mixtures prepared in the same manner were vortexed and allowed to stand at room temperature for 30 min, after which the optical activity at 765 nm was measured using UV/VIS spectrophotometer (Schimadzu, Japan) using distilled water as blank. Dilute methanolic extracts of 0.76 mL was used for the estimation of the plant samples in test tubes. The same approach was used for the standard.

The method described by Dewanto *et al.*[19] was used to determine the total flavonoids in the samples. In the procedure, distilled water was thoroughly mixed with methanol extracts, followed by addition of $NaNO_2$ solution was to every test tube. $AlCl_3$ solution was later added after 5 min. The samples were kept for six minutes and 1 M NaOH was added. Total volume of the mixture was made to 5 mL by addition of distilled water and the tubes were vigorously stirred. The resulting solution was pink-colored and its absorbance determined using spectrophotometer at 510 nm against the blank. Catechol was used to construct the calibration curve and the flavonoids in the sample were expressed as mg Catechol equivalents per gram of dry weight (mg CE/g DW).

2.2.8 Mineral determination

Cabinet drier operating at 70℃ was used to dry the seeds to constant weight. Microwave (Cem-MARS Xpress) was used to digest about 0.5 g of the ground seed samples using 5 ml of 65% HNO_3 and 2 ml of 35% H_2O_2. The volume of the digested samples was made to 20 ml using deionized water. The minerals in the sample was then quantified using ICP-AES (Varian-Vista, Australia)[20].

2.2.9 Statistical analyses

Analysis of variance (ANOVA) was conducted using JMP version (SAS Inc., Cary, N.C U.S.A). Analyses were carried out three times and the results are mean ± standard deviation (MSTAT C) of independent several seed samples[21].

3 Results and Discussion

3.1 Proximate composition of millet, sorghum and lupin seeds

The proximate properties of sorghum, millet and lupin seeds are given in Table 1. Moisture contents of samples changed between 5.84% (Bablone) and 8.42% (Yellow Sorghum). The moisture contents showed slight differences which were less than the moisture contents of maize, wheat, rice, pear millet and were determined as 14%, 12%, 9%, 10% and 14% respectively under similar conditions[2]. The ash contents of samples were found between 1.5% (Yellow Sorghum) and 2.2% (Bablone). Protein

Table 1 Proximate analysis of millet, sorghum and lupin seeds (%, Dry weight basis; n:3).

Seeds	Moisture	Ash	Protein	Oil	Starch
Millet	$6.99 \pm 1.01^{*c}$	1.74 ± 0.17^{b}	11.7 ± 1.23^{d}	13.3 ± 1.67^{a}	65.8 ± 2.69^{d}
Yellow sorghum	$8.42 \pm 0.99^{a**}$	1.50 ± 0.21^{e}	8.6 ± 1.12^{f}	9.8 ± 1.74^{d}	74.5 ± 2.85^{a}
White sorghum	7.10 ± 0.87^{b}	1.68 ± 0.32^{c}	10.9 ± 1.28^{e}	11.7 ± 1.89^{b}	70.7 ± 1.69^{bc}
Red sorghum	7.00 ± 0.89^{b}	1.61 ± 0.2^{cd}	12.6 ± 1.72^{d}	9.5 ± 0.79^{e}	71.4 ± 2.93^{b}
Lutop (Lupin)	6.01 ± 0.98^{cd}	1.97 ± 0.21^{a}	37.7 ± 1.28^{a}	8.3 ± 0.76^{f}	0.5 ± 0.09^{e}
Bablone (Lupin)	5.84 ± 0.56^{d}	2.23 ± 0.32^{a}	32.9 ± 1.19^{c}	11.7 ± 1.13^{b}	0.4 ± 0.06^{ef}
Deşdiğin (Lupin)	5.99 ± 0.73^{d}	1.58 ± 0.28^{de}	35.5 ± 2.08^{b}	10.9 ± 1.21^{c}	0.4 ± 0.08^{ef}

*mean ± standard deviation;** Values within each row followed by different letters are significantly different ($p < 0.05$)

content changed between 8.6% (yellow sorghum) and 37.7% (lutop), while oil contents of samples change between 8.3% (Lutop) and 13.3% (Millet). In addition, starch contents of samples ranged from 0.4% (Deşdiğin) to 74.5% (Yellow Sorghum). *Lupinus albus* contained 3% starch[5]. The oil contents of sorghum and millet seeds were determined as 3.95 to 5.63%, respectively[22]. The oil content of sorghum obtained in this study were greater than the values obtained by Osman *et al.*[22]. Several lupin varieties contained 30.6-37.9% protein and 8.54-14.64% oil; 1.21-3.09 g/100 g sucrose, 2.81-4.53 g/100 g starch[23]. The starch content of lupin seeds varied between 12 and 15% and larger amounts of dissolvable non-starch polysaccharides (30-40%)[24]. Some varieties of lupin contained 6.55-7.03% moisture and 29.33-37.07% protein[25]. Martinez-Villaluenga *et al.*[23] and Guemes-Vera *et al.*[25] reported that lupin serves as a good source of vitamins, minerals, lipids, proteins and dietary fibres. Ullah *et al.*[26] reported that proximate compositions of some maize varieties ranged between 9.201-10.90% moisture content, 0.7-1.3% ash, 3.21-7.71% fats, 7.71-14.60% proteins, and 0.80-2.32% crude fibre and 69.659-74.54% carbohydrates. Chukwu *et al.*[1] reported that Brown and white guinea corn contained 5.03% and 3.03% oil, 10.80% and 10.00% (encent yellow sorghum) crude protein and 1.87% and 1.97% ash, respectively. The oil and protein content reported in this study for all samples are higher than those of brown and white guinea corn. However, the ash contents are lower than those of both guinea corns and rice, wheat and maize[1].

3.2 Bioactive compounds in millet, sorghum and lupin seeds

Table 2 shows the extractable and hydrolysable phenolics, total phenolics, total flavonoid and anthocyanin contents of millet, sorghum and lupin seeds. The table showed that the lupin seeds had higher extractable and hydrolsable phenolics than sorghum and millet seeds. While extractable phenolics contents for FAE and GAE of grains change between 1.62 mg FAE/g (White Sorghum) to 12.16 mg FAE/g (Deşdiğin) and 1.43 mgGAE/g (white sorghum) to 8.23 mgGAE/g (Red Sorghum), and hydrolysable phenolics contents for FAE and GAE of grains varied between 1.52 mg FAE/g (White Sorghum) and 12.92 mg FAE/g (Red Sorghum) and 1.48 mgGAE/g (White Sorghum) to 26.10 mgGAE/g (Red Sorghum). It was observed statistically significant differences among bioactive compounds of seed"s ($p<0.05$). While total phenolic contents of seeds change between 2769 mgGAE/g (Bablone) and 6087 mgGAE/g (Yellow Sorghum), total flavonoid contents of seed samples ranged from 3.7 mg catechol/g (Bablone) to 19.0 mg catechol/g (Millet). In addition, anthocyanin contents of seed samples were found between 0.039 μmol/g (Millet) and 1.132 μmol/g (Deşdiğin).

3.3 Amino acid composition of millet, sorghum and lupin seeds

Amino acid contents of millet, sorghum and lupin seeds are given in Table 3. Amino acid contents of millet changed between 0.02% (ornithine) and 2.07% (glutamic acid), while amino acid contents of yellow sorghum range from 0.02% (hydroxyproline) to 1.71% (glutamic acid), amino acid values of white sorghum changed between 0.02% (hydroxyproline) and 2.21% (glutamic acid), amino acid values of lutop seed changed between 0.02% (ornithine) and 6.77% (glutamic acid). While amino acid contents of red sorghum changed between 0.02% (ornithine) to 2.65% (glutamic acid), lupin (Deşdiğin) seed changed between 0.01% (ornithine) and 6.85% (glutamic acid). Glutamic acid was established as the most abundant amino acid in all samples. The amino acids values obtained varied between 8.45% (Yellow Sorghum) to 32.48% (lutop). Chukwu *et al.*[1] reported that nutritionally, proteins in sorghum and other cereals are deficient in some essential amino acids such as lysine, threonine and tryptophan. However, the amino acids in lupin seeds have better amino acid profile[27]. Lupin contains significant amount of lysine, but lack the essential amino acids methionine and cysteine. They are considered

Table 2 Extractable, hydroly sable phenolics, total flavonoid, phenol and anthocyanin contents of millet, sorghum and lupins (n:3).

Samples	Extractable phenolics		Hydrolysable phenolics		Total flavonoids (mg catechol/g)	Anthocyanin (μmol/g)	Total phenol (mgGAE/g)
	(mg FAE/ g)	(mg GAE/g)	(mg FAE/ g)	(mg GAE/g)			
Millet	4.15 ± 0.67*^{b}	3.27 ± 0.57^{b}	3.71 ± 0.51^{c}	3.49 ± 0.63^{d}	19.0 ± 0.1^{a}	0.039 ± 0.002^{f}	4682 ± 191^{d}
Yellow sorghum	1.69 ± 0.12^{d**}	1.49 ± 0.11^{f}	1.59 ± 0.12^{e}	1.54 ± 0.15^{e}	5.0 ± 0.2^{e}	0.043 ± 0.003^{e}	6087 ± 117^{a}
White sorghum	1.62 ± 0.23^{d}	1.43 ± 0.34^{fg}	1.52 ± 0.17^{e}	1.48 ± 0.21^{e}	7.7 ± 0.1^{d}	0.048 ± 0.007^{e}	4409 ± 129^{e}
Red sorghum	2.14 ± 0.43^{c}	1.81 ± 0.26^{d}	1.98 ± 0.28^{d}	1.89 ± 0.19^{e}	7.3 ± 0.1^{de}	0.054 ± 0.005^{d}	4821 ± 130^{c}
Lutop (Lupin)	2.56 ± 0.73^{c}	2.02 ± 0.17^{c}	12.18 ± 1.37^{a}	24.37 ± 2.18^{b}	13.0 ± 0.9^{b}	0.943 ± 0.123^{b}	3756 ± 117^{f}
Bablone (Lupin)	2.16 ± 0.68^{c}	1.68 ± 0.21^{de}	11.52 ± 1.42^{b}	23.41 ± 2.47^{c}	3.7 ± 0.7^{f}	0.548 ± 0.098^{c}	2769 ± 129^{g}
Deşdiğin (Lupin)	12.16 ± 1.23^{a}	8.23 ± 1.16^{a}	12.92 ± 1.28^{a}	26.10 ± 1.69^{a}	9.3 ± 0.8^{c}	1.132 ± 0.056^{a}	5897 ± 130^{b}

*mean ± standard deviation; ** Values within each row followed by different letters are significantly different ($p < 0.05$)

to be a good source of lysine, and are generally poor in the sulfur-containing amino acids (methionine and cystein)[5]. Gross *et al.*[28] that the seed of *Lupinus mutabilis* contained 51.0-52.6% protein and 16.0-16.2% lipids. Also these seeds contained 9.8-10.3 (g amino acid/16 g N) aspartic, 5.1-5.3 serine, 22.8-23.8 glutanic acid, 9.1-9.3 organine, 6.8-6.9 leucinne, 3.9-3.7 glycine, 3.8-3.7 proline and 5.2-5.3 lycine[28]. Our results were found partially different compared to literatüre values. These differences can be peobably due to location, growing conditions, maturation and harvest time.

3.4 Fatty acid composition of millet, sorghum and lupin seeds

Fatty acid compositions of millet, sorghum and lupin seed oils are presented in Table 4. Palmitic, stearic, oleic, linoleic and linolenic acids were major fatty acids of seed samples. Palmitic acid contents of samples ranged from 7.24% (Yellow Sorghum) to 20.04% (white sorghum). While oleic acid contents change between 25.27% (white sorghum) and 53.50% (bablone), linoleic acid contents ranged from 14.60% (Bablone) to 42.67% (millet). Linolenic acid contents of samples were found between 1.50% (Red Sorghum) to 7.35% (Deşdiğin). Generally, oleic and linolenic acid contents of lupin seed oils were found high compared to sorghum and millet seed oils, palmitic and linoleic acid contents were found low. It was observed statistically significant differences among fatty acid compositions of seed oils ($p<0.05$). Unprocessed sorghum oil contains palmitic (12.10 to 13.41%), palmitoleic (0.47 to 1.31%), stearic (1.13 to 1.36%), oleic (33.64 to 40.35%), linoleic (42.33 to 49.94%), linolenic (1.53 to 1.72%), arachidic (0.10 to 0.18%) and eicosenoic acid (0.24 to 0.39% of total lipid)[29]. However, the sorghum grain oils contain palmitic acid (11.73-20.18%) and stearic acid (1.09-2.59%)[30]. Palmitic acid (11.88-14.18%), stearic acid (1.09-1.64%) and arachidic acid (0.12-0.33%) were present in the grain oil of different sorghum varieties[31]. According to Asiedu *et al.*[32] the oil content of sorghum was found to be palmitic acid (13.2%), stearic acid (1.30%) and arachidic acid (0.20%). The content of linoleic acid of all sorghum varieties was between 38.29 and 45.74 where as oleic acid ranged from

Table 3 Amino acid contents of seeds of millet, some sorghum and lupin varieties (%), (n:3).

	Millet	Yellow sorghum	White sorghum	Red sorghum	Lutop (Lupin)	Bablone (Lupin)	Deşdiğin (Lupin)
Taurine	0.08 ± 0.01*^{a}	0.08 ± 0.02^{a}	0.08 ± 0.01^{a}	0.08 ± 0.03^{a}	0.05 ± 0.01^{b}	0.05 ± 0.01^{b}	0.05 ± 0.01^{b}
Hydroxyproline	0.06 ± 0.01^{d}**	0.02 ± 0.01^{e}	0.02 ± 0.01^{e}	– ***	0.15 ± 0.03^{a}	0.13 ± 0.01^{b}	0.10 ± 0.01^{c}
Aspartic Acid	0.90 ± 0.07^{d}	0.59 ± 0.03^{g}	0.74 ± 0.05^{f}	0.80 ± 0.11^{e}	3.53 ± 0.14^{a}	3.23 ± 0.21^{c}	3.41 ± 0.17^{b}
Threonine	0.41 ± 0.03^{b}	0.27 ± 0.02^{d}	0.35 ± 0.01^{c}	0.38 ± 0.03^{c}	1.24 ± 0.09^{a}	1.20 ± 0.07^{a}	1.21 ± 0.11^{a}
Serine	0.47 ± 0.07^{d}	0.36 ± 0.05^{f}	0.43 ± 0.03^{e}	0.51 ± 0.07^{e}	1.59 ± 0.13^{a}	1.49 ± 0.21^{b}	1.57 ± 0.17^{b}
Glutamic Acid	2.07 ± 0.16^{d}	1.71 ± 0.09^{e}	2.21 ± 0.11^{cd}	2.65 ± 0.17^{c}	6.77 ± 0.98^{ab}	6.17 ± 0.63^{b}	6.85 ± 0.71^{a}
Proline	0.70 ± 0.05^{f}	0.70 ± 0.09^{f}	0.86 ± 0.11^{e}	1.01 ± 0.08^{d}	1.27 ± 0.13^{a}	1.20 ± 0.21^{bc}	1.24 ± 0.17^{b}
Glycine	0.41 ± 0.03^{c}	0.30 ± 0.07^{e}	0.36 ± 0.01^{d}	0.37 ± 0.09^{d}	1.39 ± 0.09^{a}	1.33 ± 0.11^{b}	1.38 ± 0.12^{a}
Alanine	0.84 ± 0.11^{d}	0.74 ± 0.13^{e}	0.94 ± 0.09^{c}	1.13 ± 0.05^{a}	1.14 ± 0.07^{a}	1.09 ± 0.23^{b}	1.10 ± 0.06^{b}
Cysteine	0.21 ± 0.03^{d}	0.15 ± 0.01^{f}	0.19 ± 0.01^{e}	0.19 ± 0.05^{e}	0.49 ± 0.03^{b}	0.56 ± 0.07^{a}	0.46 ± 0.03^{c}
Valine	0.59 ± 0.07^{d}	0.42 ± 0.03^{f}	0.52 ± 0.07^{e}	0.59 ± 0.03^{d}	1.39 ± 0.12^{a}	1.35 ± 0.24^{b}	1.37 ± 0.17^{a}
Methionine	0.26 ± 0.01^{a}	0.15 ± 0.01^{f}	0.20 ± 0.03^{e}	0.20 ± 0.05^{e}	0.23 ± 0.01^{bc}	0.24 ± 0.03^{b}	0.22 ± 0.03^{d}
Isoleucine	0.47 ± 0.03^{c}	0.34 ± 0.01^{e}	0.43 ± 0.07^{d}	0.48 ± 0.03^{c}	1.47 ± 0.13^{a}	1.39 ± 0.11^{b}	1.48 ± 0.17^{a}
Leucine	1.07 ± 0.07^{e}	1.06 ± 0.03^{e}	1.39 ± 0.03^{d}	1.66 ± 0.06^{c}	2.51 ± 0.16^{a}	2.40 ± 0.21^{b}	2.41 ± 0.19^{b}
Tyrosine	0.32 ± 0.03^{d}	0.23 ± 0.01^{e}	0.35 ± 0.03^{d}	0.41 ± 0.07^{c}	1.44 ± 0.09^{a}	1.31 ± 0.17^{b}	1.43 ± 0.13^{a}
Phenylalanine	0.56 ± 0.03^{d}	0.44 ± 0.01^{e}	0.57 ± 0.05^{d}	0.66 ± 0.06^{c}	1.32 ± 0.14^{a}	1.24 ± 0.11^{b}	1.30 ± 0.18^{a}
Hydroxylysine	0.04 ± 0.01^{b}	0.03 ± 0.01^{c}	0.05 ± 0.01^{a}	0.04 ± 0.01^{b}	0.03 ± 0.01^{c}	0.03 ± 0.01^{c}	0.04 ± 0.01^{b}
Ornithine	0.02 ± 0.01^{a}	0.01 ± 0.00^{b}	0.01 ± 0.00^{b}	0.02 ± 0.01^{a}	0.02 ± 0.01^{a}	0.01 ± 0.00^{b}	0.01 ± 0.00^{b}
Lysine	0.40 ± 0.03^{c}	0.25 ± 0.01^{e}	0.28 ± 0.05^{d}	0.28 ± 0.01^{d}	1.70 ± 0.21^{a}	1.61 ± 0.19^{b}	1.60 ± 0.23^{b}
Histidine	0.30 ± 0.07^{c}	0.22 ± 0.03^{e}	0.26 ± 0.03^{d}	0.30 ± 0.05^{c}	0.80 ± 0.09^{a}	0.78 ± 0.05^{b}	0.77 ± 0.11^{b}
Arginine	0.52 ± 0.03^{d}	0.31 ± 0.01^{f}	0.41 ± 0.05^{e}	0.42 ± 0.07^{e}	3.67 ± 0.45^{a}	2.94 ± 0.38^{c}	3.14 ± 0.27^{b}
Tryptophan	0.15 ± 0.01^{c}	0.07 ± 0.01^{d}	0.08 ± 0.01^{d}	0.09 ± 0.01^{d}	0.28 ± 0.03^{b}	0.27 ± 0.05^{b}	0.31 ± 0.04^{a}
Total	10.85	8.45	10.73	12.27	32.48	30.02	31.45

*mean ± standard deviation; ** Values within each row followed by different letters are significantly different ($p < 0.05$); *** undetermined

Table 4 Fatty acid composition of seed oils of millet, some sorghum and lupin varieties (%), (n:3).

Fatty acids	Millet	Yellow sorghum	White sorghum	Red sorghum	Lutop (Lupin)	Bablone (Lupin)	Deşdiğin (Lupin)
Myristic	0.21 ± 0.03^{c}*	0.38 ± 0.07^{b}	0.44 ± 0.9^{a}	0.38 ± 0.05^{b}	0.16 ± 0.03^{d}	0.13 ± 0.01^{e}	0.13 ± 0.01^{e}
Palmitic	15.57 ± 0.67**^{c}	15.43 ± 0.33^{c}	20.04 ± 0.78^{a}	17.95 ± 0.91^{b}	8.17 ± 0.11^{d}	7.24 ± 0.13^{e}	8.43 ± 0.21^{d}
Palmitoleic	0.68 ± 0.09^{b}	0.79 ± 0.07^{a}	0.34 ± 0.03^{d}	0.67 ± 0.13^{b}	0.29 ± 0.03^{e}	0.34 ± 0.01^{d}	0.36 ± 0.07^{c}
Stearic	1.98 ± 0.21^{d}	1.71 ± 0.32^{e}	3.92 ± 0.28^{a}	2.12 ± 0.39^{c}	1.39 ± 0.13^{f}	1.84 ± 0.17^{e}	2.45 ± 0.43^{b}
Oleic	33.83 ± 1.24^{f}	37.06 ± 1.09^{d}	25.27 ± 1.32^{g}	35.58 ± 1.48^{e}	43.94 ± 1.26^{c}	53.50 ± 1.35^{a}	50.39 ± 1.63^{b}
Vaccenic	– ***	–	–	–	1.86 ± 0.23^{b}	2.54 ± 0.19^{a}	2.55 ± 0.27^{a}
Linoleic	42.67 ± 0.97^{a}	40.14 ± 0.85^{b}	42.53 ± 0.69^{a}	39.92 ± 0.73^{c}	23.89 ± 1.13^{d}	14.60 ± 0.98^{f}	17.03 ± 0.88^{e}
Linolenic	1.69 ± 0.09^{f}	1.83 ± 0.13^{e}	2.94 ± 0.11^{d}	1.50 ± 0.18^{g}	7.14 ± 0.98^{c}	7.29 ± 0.67^{b}	7.35 ± 0.56^{a}
Arachidic	–	–	–	–	1.00 ± 0.01^{a}	0.9 ± 0.01^{b}	1.01 ± 0.03^{a}
Gonodic	–	0.35 ± 0.12^{d}	–	0.34 ± 0.09^{d}	3.83 ± 0.21^{a}	3.78 ± 0.34^{b}	2.42 ± 0.42^{c}
Behenic	–	–	0.42 ± 0.09^{d}	–	3.0 ± 0.56^{a}	2.85 ± 0.22^{b}	2.81 ± 0.34^{c}
Erucic					1.42 ± 0.32^{a}	1.38 ± 0.27^{b}	0.69 ± 0.11^{c}
Lignoceric	0.34 ± 0.09^{e}	0.35 ± 0.03^{e}	0.40 ± 0.07^{d}	0.41 ± 0.01^{d}	0.79 ± 0.09^{a}	0.62 ± 0.07^{c}	0.73 ± 0.11^{b}

**mean ± standard deviation; ** Values within each row followed by different letters are significantly different ($p < 0.05$); ***undetermined

Table 5 Sugar composition of seeds of millet, some sorghum and lupin varieties (%), (n:3).

Samples	Millet	Yellow sorghum	White sorghum	Red sorghum	Lutop (Lupin)	Bablone (Lupin)	Deşdiğin (lupin)
Fructose	1.26 ± 0.13*^{a}	0.72 ± 0.09^{c}	1.12 ± 0.15^{b}	0.73 ± 0.03^{c}	–	–	–
Glucose	2.90 ± 0.47^{a}**	2.22 ± 0.58^{c}	2.23 ± 0.21^{c}	2.50 ± 0.69^{b}	0.36 ± 0.03^{d}	0.20 ± 0.01^{e}	0.18 ± 0.03^{fe}
Sucrose	– ***	–	–	–	1.57 ± 0.09^{c}	1.87 ± 0.09^{b}	2.66 ± 0.11^{a}
Raffinose	0.03 ± 0.01^{d}	0.04 ± 0.01^{d}	0.01 ± 0.01^{e}	0.01 ± 0.01^{e}	0.61 ± 0.03^{b}	0.65 ± 0.07^{a}	0.56 ± 0.09^{c}
Stachyose	0.01 ± 0.01^{d}	–	0.02 ± 0.01^{d}	0.02 ± 0.01^{d}	2.07 ± 0.21^{c}	2.15 ± 0.17^{b}	2.22 ± 0.13^{a}
Verbascone	–	–	–	–	–	0.12 ± 0.01^{a}	0.05 ± 0.01^{b}

*mean ± standard deviation; **Values within each row followed by different letters are significantly different ($p < 0.05$); ***undetermined

29.15 to 37.98%[22]. However, Mehmood *et al.*[30] reported that polyunsaturated fatty acid contents of sorghum are greater than monounsaturated fatty acids. Oleic acid contents of bitter and sweet lupin oils were found as 52.22% and 44.93, respectively[33]. In addition, bitter lupin oil contained 9.41% palmitic, 20.51% linoleic, 13.30% linoleic and 2.13% arachidic acids, and sweet lupin oil contained 7.71% palmitic, 1.71% stearic, 26.25% linoleic, 15.81% linoleic and 2.74% arachidic acids[33]. Bhardwaj *et al.*[34] reported that lupin oil contained 51% linoleic acid, 23% oleic acid, 10% palmitic acid and 7% linolenic acid. Loredo-Dávila *et al.*[35] reported that lupin seed oil contained 13.12% palmitic, 6.77% stearic, 14.24% oleic 50.59% linoleic and 7.81% linolenic acids. Results showed differences as quantitative vales compared to literature values. But, major fatty acids of seed oils were found similar with literature values.

3.5 Sugar composition of millet, sorghum and lupin seeds

Table 5 shows the sugar content of the seed samples. Glucose contents of samples varied between 0.18% (Deşdiğin) to 2.90% (Millet). While fructose was found in millet and sorghum seeds, sucrose was detected only in lupin seeds. Sucrose amounts in lupin seeds changed between 1.57% (Lutop) and 2.66% (Deşdiğin). Fructose was determined between 0.72% (Yellow Sorghum) and 1.26% (Millet). Generally, stachyose is main sugar of lupin seeds, fructose and glucose were major sugar of millet and sorghum samples. The sucrose content of lupin ranged between 1.5 to 3.5%, why stachyose ranged between 6.0 and 7.5%. Raffinose contents which was found to be 33.2 g/kg was significantly higher than verbascose (8.49 g/kg) in lupin seeds[36]. According to Muzquiz *et al.*[11], lupin (*L. mariae-josephi*) seeds contain 79.1 g/kg oligosaccharides. However, little amount of fructose was found in lupin seeds and *Lupinus albus* seeds contained 1.8% sucrose, 0.4% raffinose, 2.8% stachyose, 0.3% verbascose[5]. Gross *et al.*[28] detected 9.0-9.9% sucrose, 16.9-16.6% raffinose, 68.3-67.7% stachyose and 5.8-5.8% verboscose in seeds of two strains (Inti and line 2150) of *Lupinus metabilis*, respectively.

3.6 Mineral contents of millet, sorghum and lupin seeds

Mineral contents of sorghum, milet and lupin seeds are shown in Table 6. Generally there is a wide variation in mineral contents of sorghum, millet and lupin seeds. While P contents of sorghum ranged between 534.89 mg/kg (Yellow Sorghum) to 3984.60 mg/kg (Bablone), K contents ranged from 1831.34 mg/kg (While Sorghum) to 11895.8 mg/kg (Lutop). Ca contents were found between 112.74 mg/kg (White Sorghum) and 3418.64 mg/kg (Yellow Sorghum). However, Mg contents ranged from 392.17 mg/kg (Red Sorghum) and 1605.38 mg/kg (Bablone). Fe contents ranged from 19.06 mg/kg (Yellow Sorghum) to 64.06 mg/kg (Deşdiğin). Zn contents ranged from 4.33 mg/kg (Red Sorghum) and 36.64 mg/kg (Lutop). Mn was found between 4.18 mg/kg (Red Sorghum) to 2341.40 mg/kg (Lutop). Na contents of samples were found between 18.02 mg/kg (While Sorghum) to 2163.18 mg/kg (Red Sorghum). It was observed statstically significant differences among mineral contents of seed"s ($p<0.05$). In general, lupin seeds contained more minerals than millet and sorghum seeds.

Chukwu *et al.*[1] reported that Brown and white Guinea corn grains contained 0.14 and 0.27% Ca, 0.19 and 0.21% K and 0.16 and 0.12% P, respectively. Previous studies showed that the proximate composition of some maize grains varied between 9.201-10.908% moisture content, (0.7-1.3%) ash, (3.21-7.71%) fat and (7.71-14.60%) protein 26). In maize grains, the amount of sodium is 540.30-620.41 ppm, K (2915-3471 ppm), Ca (410-590 ppm), Fe (38.02-56.14 ppm), Zn (37.05-52.4 ppm), Mg (985.2-1125.3 ppm) and Cu (11.02-14.25 ppm)[26]. Mo contents of samples were found at the low levels (1.92-4.17 mg/kg. There were statistically differences in the mineral contents among the sorghum varieties, lupin and millet grain ($p<0.05$). Results showed differences when compared to results of Ullah *et al.*[26].

Table 6 Mineral contents of seeds of millet, some sorghum and lupin varieties (mg/kg), (n : 3).

	P	K	Ca	Mg	S	Fe	Zn	Mn	B	Cu	Mo	Na	Se
Yellow Sorghun	534.89 ± 10.75^{**g}	6767.77 ± 95.90^{d}	3418.64 ± 10.20^{a}	1338.69 ± 1.02^{e}	637.31 ± 12.47^{d}	20.57 ± 1.38^{f}	11.29 ± 0.79^{e}	17.75 ± 0.51^{d}	9.39 ± 1.67^{d}	4.98 ± 0.21^{d}	0.30 ± 0.04^{f}	55.89 ± 0.22^{e}	0.20 ± 0.02^{e}
Red Sorghun	1021.27 ± 12.79^{f**}	5151.10 ± 68.44^{e}	930.60 ± 3.06^{c}	392.17 ± 1.88^{g}	591.45 ± 8.20^{e}	19.06 ±0.97^{fg}	4.33 ± 0.23^{f}	4.18 ± 0.25^{g}	5.55 ± 1.58^{e}	4.76 ± 0.30^{e}	0.10 ± 0.01^{g}	2163.18 ± 22.08^{a}	0.74 ± 0.01^{b}
White Sorghun	1224.70 ± 3.55^{e}	1831.34 ± 4.59^{g}	112.74 ± 1.99^{g}	754.26 ± 8.68^{f}	470.26 ± 7.89^{g}	21.02 ± 1.30^{e}	7.51 ± 0.52^{e}	6.36 ± 0.21^{f}	2.09 ± 2.01^{g}	2.12 ± 0.06^{g}	0.43 ± 0.07^{e}	18.02 ± 1.03^{f}	1.06 ± 0.02^{a}
Millet	1501.59 ± 10.71^{d}	1965.79 ± 21.62^{f}	125.06 ± 2.38^{f}	931.23 ± 3.65^{e}	507.76 ± 2.87^{f}	26.87 ± 2.30^{d}	10.50 ± 1.00^{d}	9.93 ± 0.89^{e}	3.04 ± 1.03^{f}	3.13 ± 0.04^{f}	0.52 ± 0.1^{d}	11.55 ± 2.01^{g}	1.03 ± 0.02^{a}
Lutop (Lupin)	2986.50 ± 660.95^{b}	11895.8 ± 2366.66^{a}	2755.07 ± 1318.72^{b}	1265.52 ± 267.36^{d}	1881.64 ± 166.81^{b}	56.45 ± 8.03^{c}	36.64 ± 6.57^{a}	2341.40 ± 387.65^{a}	21.75 ± 4.43^{a}	13.32 ± 1.93^{b}	1.92 ± 0.36^{c}	47.18 ± 0.21^{d}	0.17 ± 0.03^{f}
Bablone (Lupin)	3984.60 ± 463.66^{a}	10907.95 ± 784.92^{b}	2616.80 ± 309.06^{c}	1605.38 ± 123.62^{a}	2350.19 ± 246.72^{a}	61.00 ± 1.70^{b}	32.61 ± 4.40^{b}	1668.67 ± 168.97^{c}	16.35 ± 2.54^{c}	11.90 ± 0.94^{c}	2.01 ± 0.34^{b}	58.78 ± 1.17^{b}	0.31 ± 0.07^{c}
Deşdiğin (Lupin)	2016.04 ± 178.21^{c}	7363.66 ± 387.75^{c}	2153.05 ± 598.32^{d}	1411.07 ± 72.24^{b}	1814.56 ± 144.61^{c}	64.06 ± 1.04^{a}	32.54 ± 3.32^{b}	2093.20 ± 46.20^{b}	17.40 ± 2.90^{b}	16.57 ± 8.75^{a}	4.17 ± 0.23^{a}	28.13 ± 2.38^{e}	0.23 ± 0.01^{d}

*mean ± standard deviation; **Values within each column followed by different letters are significantly different ($p < 0.05$)

Conclusion

Sorghum, lupin and millet seeds are important crop and contains important health promoting constituents. Generally, protein, oleic acid, amino acid and mineral contents of lupin varieties were higher as compared to those of millet phenol, anthocyanin and sorghum seeds. The protein contents of lupin seeds were reported higher than those of millet and sorghum seed samples. Glutamic acid was established as the most abundant amino acid in all samples. Generally, amino acid contents of lupin seeds were found higher than thos of amino acid contents of millet and sorghum seeds. Palmitic, stearic, oleic, linoleic and linolenic acids were major fatty acids of seed samples. Generally, stachyose is main sugar of lupin seeds, fructose and glucose were major sugar of millet and sorghum samples.

Generally there is a wide variation in mineral contents of sorghum, millet and lupin seeds. While linoleic acid contents of millet and sorghum seed oils are found higher than those of lupin oils, oleic acid contents of lupin seed oils were determined higher compared to results of millet and sorghum seed oils.

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