

CHEMICAL ANALYSES AND FOOD PROPERTIES OF LIMA BEAN
(PHASEOLUS LUNATUS L.) SEEDS GROWN IN PLATEAU STATE, NIGERIA

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Abstract

The nutritional composition and functional properties of lima bean were studied using standard analytical techniques. The results gave proximate composition as follows: moisture, ash, crude fat, crude protein, crude fibre and carbohydrate were 3.8, 2.10, 4.0, 1.50, 3.2, 0.50, 2.5, 9.2, 3.0, 2.50, 0.05 and 60.6%, respectively. The results also showed that the predominant mineral was Ca (147.4mg/100g) followed by Mg (142.2mg/100g). Other minerals determined were Fe, P, Mn, Ni, Na, K, Cu, Zn and Cr with concentrations of 56.0, 10.7, 2.6, 1.7, 1.4, 1.3, 1.3, 0.4 and 0.2mg/100g sample, respectively while harmful metals such as Cd and Pb were not at the detectable range of AAS. The sample contained nutritionally useful quantities of most of the essential amino acids with the first limiting amino acid as Met + Cys (0.49g/100g crude protein). Functional properties results were: foaming capacity (6.0, 2.5%), foaming stability (75.0, 1.0%), water absorption capacity (360.0, 6.0%), oil absorption capacity (356.0, 1.5%), emulsion capacity (50.0, 5.0gmL⁻¹), emulsion stability (65.0, 3.0gmL⁻¹), least gelation concentration (12.0, 2.0%) and bulk density (0.39356, 0.5gmL⁻¹). The studied sample could therefore be good sources of protein and supplement cereal diets in raising the biological value significantly.

Keywords: Nutritional composition, functional properties, lima bean

INTRODUCTION

In Africa and other developing countries, there is inadequate supply and shortage of food protein because animal protein such as meat, milk and eggs are expensive and relatively difficult to acquire (Olaofe *et al.*, 1994; Akintayo, 1997). In view of this, it is geared at identifying and evaluating under-utilized protein sources. This development has stimulated research on the utilization of some under-utilized legumes such as African yam bean (Adeyeye, 1996; Oshodi *et al.*, 2005; Adebowale *et al.*, 2004; Amarteifio and Moichubedi, 1997; Enwere and Hung, 1996), mucuna bean (Johnson and Brekke, 1983; Muchuka, 2002), Kersting's groundnut (Aremu *et al.*, 2006a; Mitanga and Sugiyama, 1974), Gila bean (Siddhuraju *et al.*, 2001), faba bean (Kranee *et al.*, 1996), lupin seed (Lqari *et al.*, 2002), cranberry bean (Aremu *et al.*, 2010) and chick pea (Liu and Aung, 1998). The efforts geared towards legumes are predicated on the fact that legumes have been established to be cheap but abundant sources of protein (Aykroyd and Doughty, 1994).

Lima bean (*Phaseolus lunatus*) is referred to as edible seed of leguminous plant belonging to the family of leguminosae or fabaceae. There are more than a hundred named Lima cultivars. Lima beans are most often associated with succotash, a traditional native American dish that combines this delicious bean with corn. Many people think that they are native to the United State where the early European explorers were thought to have first discovered them which is actually reflected in its name "Lima", the capital of the south American country of Peru. The pod of lima bean is flat, oblong and slightly curved, averaging about three inches in length. Within the pod reside two to four flat kidney shaped seeds that are what we generally refer to as lima bean (Labell, 1989). The origin of lima beans cultivation in the middle belt of Nigeria can not be ascertained. Although different varieties in terms of colours such as red, brown and black are grown but nutritional and industrial potential of lima bean are unknown to the host communities.

Therefore this work seeks to evaluate nutritional composition (proximate, minerals and amino acids) and functional properties of lima bean seed flour, with a view to providing useful information towards effective utilization of this legume in various food applications.

MATERIALS AND METHODS

Sample Collection and preparation

Lima bean seeds were directly purchased from farmer in a village close to University of Jos main campus, Plateau State, Nigeria. The seeds were sorted to remove stones and bad ones. Cold water was added on the sample for dehulling. The dehulled seeds were dried in the oven at 45°C and finally ground into fine flour with a small sample mill.

Proximate Analyses

The moisture, ash, ether extract, crude fibre, crude protein (N x 6.25) and carbohydrate (by difference) were determined in accordance with AOAC (1995) methods. All proximate analyses of the legume flour were carried out in triplicate and reported in % sample. All chemicals were of Analar grade.

Mineral Analysis

The minerals were analyzed by dry-ashing the samples at 500°C to constant weight and dissolving the ash in volumetric flask using distilled, deionised water with a few drops of concentrated hydrochloric acid. Sodium and potassium were determined by using a flame photometer (Model 405, Corning, UK) using NaCl and KCl to prepare the standards. All metals were determined by atomic absorption spectrophotometer (Perkin-Elmer Model 403, Norwalk, CT, USA). Earlier, the detection limits of the metals had been determined according to Techtron (1975).

Amino Acid Analysis

Amino acid composition was analyzed by ion exchange chromatography (IEC) (FAO/WHO, 1991) using the Technicon Sequential Multisample (TSM) amino acid analyzer (Technicon Instruments Corporation, New York). The period of analysis was 76min for each sample. The gas flow rate was 0.50mL/min at 60°C with

reproducibility consistent within 3%. The height of each peak produced by the recorder of the TSM (each representing an amino acid) was measured and calculated. The amino acid values reported were the averages of two determinations. Tryptophan was not determined. Norleucine was the internal standard.

Determination of quality of dietary protein isoelectric point and predicted protein efficiency ratio (P-PER)

The quality of dietary protein was measured by finding the ratio of available amino acids in the legume sample compared with needs expressed as ratio (FAO, 1970; Bender, 1992). Amino acid score (AMSS) was then estimated by applying the FAO/WHO (1991) formula;

$$AMSS = \frac{\text{mg amino acid in lg of test protein}}{\text{mg amino acid in lg of reference protein}} \times \frac{100}{1}$$

$$PI_m = \sum_{i=1}^{n-1} p_i X_i$$

Where:

$p_i m$ = the isoelectric point of the mixture of amino acids.

p_i = the isoelectric point of the amino acids in the mixture.

X_i = the mass or mole fraction of the amino acids in the mixture.

The predicted protein efficiency ratio (P-PER) of the sample was calculated from their amino acid composition based on the equation described by Alsmeyer *et al.* (1974) as stated thus;

$$P-PER = 0.468 + 0.454(\text{Leu}) + 0.105(\text{Tyr})$$

Determination of functional properties

Foaming Capacity (FC) and foaming stability (FS), were determined by the method described by Coffman and Garcia (1977). Full experimental details have been reported by Aremu *et al.* (2007). Water and oil absorption capacities (WAC and OAC) were measured by the Beuchat (1977) procedures. Oil emulsion capacity (OEC) was determined by the procedure of Beuchat (1977), modified by Adeyeye *et al.* (1994) and emulsion stability (OES) by the method of Beuchat (1977). Bulk density (BD) was

determined using the procedure of Chou and Morr (1979) as modified by Akpapunam and Markakis (1981); Narayana and Narasinga Rao (1984). Lowest gelation concentration, (LGC) was determined by employing the method of Coffman and Garcia (1977) with slight modification as described by Aremu *et al.* (2007b).

RESULTS AND DISCUSSION

Proximate Composition

Proximate composition of *Phaseolus lunatus* flour is presented in Table 1. The percentage composition of moisture content (3.8 2.10%) was high compared with the value reported for cranberry bean (1.7 0.51%), rare cowpea (1.8 0.25%) and *Prosopis africana* (1.9 0.3%) as reported by Aremu *et al.* (2006b; 2006c). The water soluble ash (4.0 1.50%) is comparable with some reported studies on legumes (Mtanga and Sugiyama, 1974; Kay, 1979; Poulter, 1981; Aletor and Aladetimi, 1989; Aremu *et al.*, 2009). The ether extract (crude fat) value of 3.2 0.50% did not qualify *Phaseolus lunatus* seed as an oil-rich legume when compared with soybean (22.8 23.5%) (Elias *et al.*, 1976; Salunkhe *et al.*, 1985; Aremu *et al.*, 2007b), or other seeds such as pumpkin seed (47.0 49.2%) (Asiegbe, 1989; Fagbemi and Oshodi, 1991) and *Citrullus vulgaris* Schrad. (47.9 51.1%) (Ige *et al.*, 1984; Olaofe *et al.*, 1994; Ogungbenle *et al.*, 2005) grown in Nigeria. Crude protein value (25.9 2.30%) of *Phaseolus lunatus* is comparable with some commonly consumed plant proteins in Nigeria (Akobundun *et al.*, 1982; Ihekoronye and Ngoddy, 1985; Aremu *et al.*, 2007b) and qualified the seeds as protein rich plant food. The recommended daily allowance for protein for children ranges from 23.0 36.0g and for adult, 44 56g (NRC, 1989). It can be evaluated that *Phaseolus lunatus* can supply the recommended daily intake of protein for children. However, about 220g of *P. lunatus* seeds may be required by an adult weighing 70Kg to meet the minimum daily protein requirement of 57g (FAO/WHO/UNU, 1985). This amount cannot lead to dietary stress (Robinson, 1975). *P. lunatus* had crude fibre content (2.5 0.05%) comparable with values of some legumes reported in the literature: cowpea, 2.6% (Aletor and Aladetimi, 1989); bambara groundnut, 2.4 4.1% (Aremu *et al.*, 2006b); pigeon pea, 3.8% (Oshodi and Ekperigin, 1989); African

yam bean, 3.5% (Adeyeye and Aye, 1998). The calculated metabolizable energy and fatty acids values for *Phaseolus lunatus* flour were 1,588.9KJ/100g and 2.56%, respectively. The metabolizable energy in the present study suggests that the flour sample had a concentrated source of energy and was favourably comparable to cereals (Aremu *et al.*, 2006b; Olaofe *et al.*, 2008) while the fatty acids showed that the oils may be both edible and suitable for industrial purposes (Aremu *et al.*, 2007c).

The most abundant minerals were calcium (147.4mg/100g) and magnesium (142.2mg/100g) while the least was chromium (0.4mg/100g) (Table 2). These values compare favourably with some reported values of cowpea varieties and under-utilized legumes (Oyenuga, 1968; Aletor and Aladetimi, 1989; Oshodi *et al.*, 1998; Jirapa *et al.* 2001; Aremu *et al.*, 2006b; Olaofe *et al.*, 1993). The values of sodium, potassium, manganese, chromium, nickel, copper and zinc were low (Table 2). It is interesting to note that harmful minerals such as lead and cadmium are not at detectable range of AAS in the *Phaseolus lunatus* seed powders. Generally, the samples of *Phaseolus lunatus* may be regarded as good sources of essential minerals. Calcium in conjunction with phosphorus, magnesium, vitamin A, C and D, chlorine and protein are all involved in bone formation (Fleck, 1976). Calcium is also important in blood clotting, muscle contraction and in certain enzymes in metabolic processes (Shills, 1973). Magnesium is an activator of many enzyme systems and maintains the electrical potential in nerves (Shills, 1973). Phosphorus assists calcium in many body reactions, although it also has independent functions. Modern diets that are rich in animal proteins and phosphorus may promote the loss of calcium in the urine (Shills and Young, 1992). This has led to the concept of the calcium to phosphorus ratio. If the Ca:P ratio is low (low calcium, high phosphorus intake), more than the normal amount of calcium may be lost in the urine. Food is considered "good" if the Ca:P ratio is above 1 and "poor" if the ratio is less than 0.5, while a Ca:P ratio above 2 helps to increase the absorption of calcium in the small intestine. Result of Ca:P ratio in the studied sample (13.8) was good. Sodium and potassium are required to

maintain osmotic balance of the body fluids, the pH of the body, to regulate muscle and nerve limitability, control glucose absorption and enhance normal retention of protein during growth (NRC, 1989). The ratio of sodium to potassium in the body is of great concern for the prevention of high blood pressure. A Na:K ratio less than 1 is recommended. The value of Na:K in the studied sample was 0.2 (Table 2). This suggests that *Phaseolus lunatus* seed powder may not promote high blood pressure.

The result of amino acid composition is shown in Table 3. Asp and Glu were found to be the most abundant as expected to be present in legumes (Aremu *et al.*, 2010). Asp and Glu together make up to 17.41g/100g crude protein with a percentage of 26.06%. This value is in close agreement with the report of Olaofe and Akintayo (2000); Oshodi *et al.* (1998); Aremu *et al.* (2006a,b); Adeyeye and Aye (1998) who had observed that Asp and Glu were the most abundant in legumes and nuts. Leu was also found to be in abundance with value of 6.17g/100g protein. Table 4 depicts the essential, non-essential, acidic, neutral and sulphur containing amino acids. The total essential amino acids (TEAA) (with His) was 44.0g/100g protein which represents 53.9%. This is comparable with values obtained for selected oil seeds (Olaofe *et al.*, 1994), varieties of *V. subterranean* and *P. coccineus* (Aremu *et al.*, 2006b). Pellet and Young (1977) reported that the nutritive value of a protein depends primarily in its capacity to satisfy the needs for nitrogen and essential amino acid. Tryptophan was not determined. The calculated isoelectric point (pI) was 4.11. This is useful in predicting the pI for protein in order to enhance a quick precipitation of protein isolate from biological samples (Olaofe and Akintayo, 2000). The predicted protein efficiency ratio (PPER) is one of the quality parameters used for protein evaluation (FAO/WHO, 1991). The PPER in this report (2.01) is comparable with the value reported for *Prosopis africana* (2.3) (Aremu *et al.*, 2006a) and *Phaseolus coccineus* (1.91) (Aremu *et al.*, 2007a). Therefore, *Phaseolus lunatus* could be considered as good diets that can provide the required essential amino acids. The observed value (27.3g/100g protein) for essential aliphatic amino acid (EAAA)

which constitutes the hydrophobic region of protein indicates that *P. lunatus* may have better emulsification properties (Oshodi *et al.*, 1998). The total acidic amino acid (TAAA) (36.5g/100g) was found to be greater than total basic amino acid (TBAA) (19.3g/100g), indicating that protein of *P. lunatus* is probably acidic in nature. The essential aromatic amino acid (EArAA) was 9.2g/100g protein. This value falls within the ideal range suggested for infant protein (6.8–11.8g/100g) (FAO/WHO/UNU, 1985).

With the exception of Phe + Tyr in the *P. lunatus* seed flour, the contents of essential amino acids are lower than FAO/WHO (1991) recommendations (Table 5). Thus, for a healthy diet, *P. lunatus* will require supplementation with essential amino acids such as Ile, Leu, Lys, Met + Cys (TSA), Thr and Val. It has been reported that the essential amino acids most often acting in a limiting capacity are Met (and Cys), Lys and Try (Aremu *et al.*, 2010). However, the first limiting amino acids for the studied sample in this report was Thr.

The results of some functional properties are presented in Table 6. *P. lunatus* produced thin foams with medium size air cells and its foaming capacity (FC) of 6.0–2.5% was very low when compared with that of benniseed (18.0%), pear millet and quinoa (19.0%) as reported by Oshodi *et al.* (1999); varieties of legume seeds (14.6–15.5%), Aremu *et al.* (2007d); soybean (66%), Lin *et al.* (1974); great northern bean (32%), Sathe *et al.* (1982) and varieties of African yam bean (54.0–55.0%), Oshodi *et al.* (1997). Foaming stability recorded after 8h was 75.0–1.0%. Water absorption capacity (WAC), 360.0–6.0 is higher than that of sunflower (107%) and soybean (130%) reported by Lin *et al.* (1974) and varieties of lima bean (130–142%) (Oshodi and Ekperigin, 1989). The high WAC recorded in this report suggests that *P. lunatus* may be used in the formulation of some foods such as soups or baked products (Olaofe *et al.*, 1994). The value of oil absorption capacity (OAC) in this report was 356.0–1.5 (Table 6). This value is higher than the reports of Aremu *et al.* (2007d) on varieties of legumes seeds (127.8–172.0%); pigeon pea (89.7%), Oshodi and Ekperigin, 1989 and varieties of cowpeas (281–310%) (Olaofe *et al.*, 1993). OAC is important as oil

acts as a flavour retainer and improves the mouth feel of foods. *P. lunatus* will be good in this respect. For emulsifying activity; *P. lunatus* had a low percentage of emulsion capacity, 50.0 5.0mLg⁻¹ in comparison with pear millet (89.0mLg⁻¹), quinoa (104.0mLg⁻¹) (Oshodi *et al.*, 1974) but higher than value reported for soybean (18mLg⁻¹) (Lin *et al.*, 1974) and pigeon pea (11mLg⁻¹) (Oshodi and Ekperigin, 1989). This indicates that *P. lunatus* might be useful in the production of sausages, soups and cakes (Kinsella, 1979). Emulsion stability (ES) after 24h, which is the volume of water separated is shown in Table 6. The value obtained for *P. lunatus* (65.0 3.0mLg⁻¹) is higher than that of pear millet (34.0mLg⁻¹) (Oshodi *et al.*, 1999). The least gelation concentration (12.0%) is

also shown in Table 6; it is higher than the values reported for some legumes e.g. pigeon pea (10%) (Oshodi and Ekperigin, 1989); and cowpea 10% (Altschul and Wilcke, 1985). The ability of protein to form gels and provide a structural matrix for holding water, flavours, sugars and food ingredients is useful in food application and in new product development, thereby providing an added dimension to protein functionality (Olaofe *et al.*, 1994). The bulk density (BD) was 0.3936 0.5gmL⁻¹) (Table 6). This value is comparable with the values reported for various samples of extrusion texturized soya products with varied protein and soluble sugar contents (0.2382 0.4460gmL⁻¹) (Adeyeye and Adamu, 2003).

Table 1: Proximate composition (%) of lima bean (*Phaseolus lunatus*) flour

Parameter	Concentration (%) ^a
Moisture	3.8 2.10
Total ash	4.0 1.50
Ether extract	3.2 0.50
Crude protein	25.9 2.30
Crude fibre	2.5 0.05
Carbohydrate by difference	60.6
Energy (kJ/100g) ^b	1,588.9
Fatty acids ^c	2.56

^aValues are mean standard deviation of triplicate determinations; ^bCalculated metabolisable energy (kJ/100g) (protein x 17 + fat x 37 + carbohydrate x 17); ^cCalculated fatty acid (0.8 x crude fat).

Table 2: Mineral composition (mg/100g) of lima bean (*Phaseolus lunatus*) flour

Parameter	Concentration (mg/100g sample)
Sodium (Na)	1.7
Potassium (K)	1.4
Calcium (Ca)	147.4
Magnesium (Mg)	142.2
Phosphorus (P)	10.7
Manganese (Mn)	2.6
Chromium (Cr)	0.4
Nickel (Ni)	1.7
Copper (Cu)	1.3
Iron (Fe)	56.0
Zinc (Zn)	1.3
Lead (Pb)	ND
Cadmium (Cd)	ND
Na:K	0.2
Ca:P	13.8

ND = not detected; Na:K = sodium to potassium ratio; Ca:P = calcium to phosphorus ratio

Table 3: Mean amino acid composition (g/100g crude protein) of lima bean (*Phaseolus lunatus*) flour

Amino acid	Concentration (g/100g protein)
Lysine (Lys)*	5.13
Histidine (His)*	2.71
Arginine (Arg)*	5.13
Aspartic acid (Asp)	8.25
Threonine (Thr)*	2.35
Serine (Ser)	2.61
Glutamic acid (Glu)	9.16
Proline (Pro)	2.31
Alanine (Ala)	4.31
Cystine (Cys)	0.91
Valine (Val)*	4.25
Methionine (Met)*	0.82
Isoleucine (Ile)*	3.35
Leucine (Leu)*	6.17
Tyrosine (Tyr)	3.02
Phenylalanine (Pha)	6.17
Isoelectric point (pI)	4.11
PPER	2.01

PPER, predicted protein efficiency ratio

Table 4: Amino acid composition (g/100g crude protein) of lima bean (*Phaseolus lunatus*) flour

Parameter	Concentration (g/100g protein)
Total amino acid (TAA)	66.8
Total non-essential amino acid (TNEAA)	30.0
Total essential amino acid (TEAA) with histidine	44.0
TEAA without histidine	33.0
% TEAA with histidine	53.9
% TEAA without histidine	49.9
Essential alphatic amino acid (EAAA)	27.3
Essential aromatic amino acid (EArAA)	9.2
Total neutral amino acid (ENAA)	24.4
% TNAA	36.5
Total acidic amino acid (TAAA)	36.5
% TAAA	54.6
Total basic amino acid (TBAA)	12.9
% TBAA	19.3
Total sulphur amino acid (TSAA)	1.7
% cystine in TSAA	2.5

Table 5: Amino acid scores of lima bean (*Phaseolus lunatus*) flour based on FAO/WHO (1973) Standards

AAC	Concentration (g/100g protein)		
	PAAESP ^a	EAAC	AMSS
Ile	40	3.4	0.85
Leu	7.0	6.2	0.89
Lys	5.5	4.2	0.76
Met + Cys (TSAA)	3.5	2.7	0.77
Phe + Tyr	6.0	7.9	1.32
Thr	4.0	2.2	0.55
Try	1.0	ND	ND
Val	5.0	4.2	0.84
Total	36.0	31.3	6.08

^aSource: Belschant *et al.* (1975)

AAC, amino acid composition

PAAESP, provisional amino acid (egg) scoring pattern

EAAC, essential amino acid composition (see Table 3)

AMSS, amino acid scores

ND, not determined

Table 6: Functional properties of lima bean (*Phaseolus lunatus*) flour

Property	Value ^a
Foaming capacity (FC)%	6.0 ± 2.5
Foaming stability (FS)%	75.0 ± 1.0
Water absorption capacity (WAC)%	360.0 ± 6.0
Oil absorption capacity (OAC)%	356.0 ± 1.5
Emulsion capacity (EC)mLg ⁻¹	50.0 ± 5.0
Emulsion stability (ES)mLg ⁻¹	65.0 ± 3.0
Least gelation concentration (LGC)%	12.0 ± 2.0
Bulk density (BD) gmL ⁻¹	0.3936 ± 0.5

^aValues are mean standard deviation of triplicate determinations.

CONCLUSION

The results revealed that the lima bean (*Phaseolus lunatus* L.) flour sample was good sources of energy, protein (amino acids) and essential minerals. It was also observed that the foaming capacity and stability in the lima bean samples were higher than some legume flours. Since foam contributes to smoothness, lightness, flavour, dispersions and palatability, hence the results obtained from the present study indicate that the sample would serve as potential replacement of known proteins in food applications requiring high foam ability and stability, for example,

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