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Effect of Multi-processing Techniques on the Chemical Composition of Horse Eye Bean (*Mucuna urens*)

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ABSTRACT

Horse Eye Bean Meal (HEBM) is one of the lesser-known legumes with a great potential. Though promising, the beans contain anti-nutritional factors. Eliminating the anti-nutritional factors by single processing method only gave partial detoxification. This experiment was therefore designed to evaluate the effect of multi-processing techniques on the chemical composition of the horse eye bean meal. The horse eye beans were cracked, divided into three groups and soaked in water at room temperature for 24, 48 and 72 h, respectively. Soaked beans in each group were peeled, divided into two sub-groups each and cooked for 60 and 90 min, respectively at 100°C. The cooked beans were sun-dried, toasted on open fire at 100°C, milled and individually stored in air-tight containers prior to the commencement of the experiment. The proximate composition, mineral, amino acid profile and concentration of some important anti-nutritional factors were determined. The experimental design used was completely randomized design. Result of the proximate composition showed that the processing methods significantly ($p<0.05$) reduced the percentage crude protein with the least value (21.62%) in treatment E. The crude fat and ash contents were significantly ($p<0.05$) increased with highest (10.16%) and (4.98%) in treatments D and F, respectively. Anti-nutritional factors were significantly ($p<0.05$) reduced. Treatment F caused greater reduction in the composition of Tannins (80.0 mg 100 g^{-1}), total phenols (10.38 mg/100 g) and hydrogen cyanide (15.20 mg/100 g). Except for calcium and magnesium, the rest of the minerals were significantly ($p<0.05$) influenced by processing. Different processing methods caused substantial reduction of some of the essential amino acids. It was concluded that 72 h soaked, 90 min cooked and toasting (treatment F) was most effective method of processing horse eye bean meal.

Key words: Horse eye bean, soaking, cooking, toasting, anti-nutritional factors, multi-processing techniques

INTRODUCTION

Legumes are important sources of proteins, carbohydrates, dietary fibre and minerals consumed worldwide (Osman, 2007). They are generally well adapted to a wide range of climates and environmental conditions. Of the thousand known legume species, only few have been extensively promoted and used. Many other potentially useful legumes are marginally known (Emenalom and Udebibie, 2005). These potential legumes might be of great importance in many zones of developing countries where there is a pressing need for food sources of high energy and good protein quality.

Horse eye bean is one of the lesser-known legumes with a great nutritional potential. Studies on the nutrient composition showed that the bean is a good source of protein (23-35%),

carbohydrate (50-56%) and fat (8-11%) (Umoren *et al*., 2007). Osaniyi and Eka (1978) reported that when the amino acids of the horse eye bean were scored against the reference protein (egg amino acids), the chemical scores were so close to each other.

Though promising, the horse eye bean contains Anti-Nutritional Factors (ANFs) such as trypsin inhibitor, lectins, phytates, phenols, cyanogenic glycosides, tannins and L-3, 4 dihydroxy phenyl alanine. These ANFs negatively affect the nutritive value of bean through direct and indirect reactions: They inhibit protein and carbohydrate digestibility, induce pathological changes in the intestine and liver tissue thus affecting metabolism (Bressani, 1993). These effects limit the use of the raw horse eye bean meal, although various processing technique like cooking and heating (toasting) tends to reduce the ANFs. Vidal-Valverde *et al*. (1997) observed that processing the beans by single method only gave partial detoxification and that the use of one method of processing may not effect the desired removal of the anti-nutritional factors. Combination of two or more methods is therefore required.

According to Emenalom and Udebibie (2005), soaking the seed in water prior to cooking was more effective in improving the nutritional value of the mucuna bean than cooking alone. They reasoned that soaking prior to cooking may have opened up more surface area for heat penetration. There is therefore, need to seek methods which when combined will effectively eliminate anti-nutritional factors and toxic substances in the horse eye bean. The study was design to evaluate the effect of combining soaking for 24, 48, 72 h, cooking at 60 and 90 min and toasting as processing methods on the chemical composition of the horse eye bean.

MATERIALS AND METHODS

Commencement of the study: The horse eye beans used for the study were bought from local farmers in Akamkpa Local Government Area of Cross River state.

The study commenced on the 25th November, 2009 and lasted for three months.

Processing of the horse eye bean meal: The raw beans obtained for this study were cracked using stone and processed on group basis as follows:

- In the first group, the cracked beans were soaked in water at room temperature for 24 h, peeled, rinsed in fresh clean water and thereafter divided into two (2) sub groups and cooked at 100°C for 60 and 90 min, respectively on open fire
- In the second and third groups, beans were soaked in water at room temperature (37°C), for 48 and 72 h, respectively, peeled, rinsed in fresh clean water and thereafter divided into two (2) sub-groups each and cooked at 100°C for 60 and 90 min. Timing started from the point of boiling. After cooking, the beans were rinsed again in clean water and sun-dried
- Soaked and cooked beans under the six sub-groups were subjected to further processing by toasting in frying pot, on open fire until the beans turned brown
- The processed beans were individually milled on 4mm screened hammer mill and stored in air tight containers prior to the commencement of the analyses

The experimental treatments were thus as follows:

Control : Raw
Treatment A : 24 h soaked, 60 min cooked and toasted
Treatment B : 24 h soaked, 90 min cooked and toasted

Treatment C : 48 h soaked, 60 min cooked and toasted
Treatment D : 48 h soaked, 90 min cooked and toasted
Treatment E : 72 h soaked, 60 min cooked and toasted
Treatment F : 72 h soaked, 90 min cooked and toasted

Proximate analysis: Samples of raw and the six processed Horse Eye Bean Meal (HEBM) groups were analyzed for their proximate composition. The percentage crude protein, crude fibre, ether extract, total ash and nitrogen free extracts were determined, according to the methods of AOAC (1995).

Amino acids determination: The amino acids profile in raw and each of the processed HEBM groups were determined on the amino acids analyzer-Technicon TSM 1(model-DNA0209) using methods described by Spackman *et al.* (1958).

Mineral analysis: Atomic Absorption spectrophotometer (Unicam-939) was used in the determination of calcium, manganese, magnesium, zinc, iron and copper while potassium and sodium were determined by the use of flame photometer. The molybdovanadate method (AOAC, 1990) was used to determine the total phosphorus.

Anti-nutritional factors determination: Concentrations of some important anti-nutritional factors present in the raw and processed HEBM were determined. Phytic acid content was determined by the method described by Wheeler and Ferrel (1971), using two grams of each dry samples. Quantitative estimation of tannins was carried out using the modified vanillin-HCl method of Price *et al.* (1978). Oxalates were determined by method out lined by Dye (1956). Cyanogenic glycoside was estimated by determining the amount of hydrogen cyanide released on hydrolysis (AOAC, 1990).

Statistical analysis: The experimental design used was completely randomized design. Data collected was subjected to analysis of variance procedures (AOAC, 1995). When significant differences were observed between treatments, the means were separated using Duncan's multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

The result of the effect of different processing techniques on the proximate composition of horse eye bean meal is presented in Table 1.

All the processing techniques caused significant ($p<0.05$) reduction in the protein content of the Horse Eye Bean (HEBM). The least protein value of 21.62% was observed in the HEBM under Treatment E and highest (27.15%) in the raw sample. The reduction in crude protein contents agree with earlier reports on Jack bean (Udedibie *et al.*, 1994) and velvet bean (Emenalom and Udebibie, 2005; Tuleun and Patrick, 2007) and this could possibly be, due to leaching and vaporization of some nitrogenous compound during processing (Osman, 2007; Yagoub *et al.*, 2008). Processing methods caused significant ($p<0.05$) increase in crude fat content. The value increased from 6.22% in the raw bean to 10.16% in HEBM under Treatment D (Table 1) soaking and cooking of the sample might have led to the cleavage of the protein-lipid or carbohydrate-lipid linkages, thereby facilitating the easy extraction of the oil by the extracting solvent

Table 1: Effect of processing on the proximate composition of processed horse eye bean meal (%dry matter)

	Treatment groups							
Constituents	Raw	A	B	C	D	E	F	SEM
Crude protein	27.15^{a}	22.25^{c}	23.65^{b}	23.65^{b}	23.83^{b}	21.62^{c}	22.04^{c}	±0.65
Crude fat	6.22^{c}	9.02^{b}	9.04^{b}	10.14^{a}	10.16^{a}	9.22^{b}	8.94^{b}	±0.46
Crude fibre	5.04	4.08	4.12	4.10	4.14	4.04	4.80	±0.14
Ash	3.28^{b}	4.68^{a}	4.74^{a}	4.82^{a}	4.80^{a}	4.92^{a}	4.98^{a}	±0.21
Nitrogen free extract	51.37	55.51	53.01	51.61	51.87	55.54	55.25	±0.67
Dry matter	93.06	94.54	94.56	94.54	94.58	95.72	95.01	±0.01

Means with different superscripts within the same row are significantly different ($p<0.05$). Means are values of triplicate determinations
SEM: Standard error of mean

(Esenwah and Ikenebomeh, 2008). Processing did not significantly ($p>0.05$) influenced the composition of crude fibre in the HEBM. Values however range from 4.04% in Treatment E to 5.04% in the raw bean (Table 1). The value obtained was lower than 5.6% reported for soybean meal (Obioha, 1992). The low crude fibre observed gives the bean an advantage in term of monogastric animal feeding. Processing significantly ($p<0.05$) increase the level of ash from 3.28% in the raw bean to 4.98% in Treatment F (Table 1). Increase in the composition of ash, in all the processing groups may be attributed to the destruction of the anti-nutritional factors, specifically phytates which are thought to be responsible in mineral bio- availability (Osman, 2007). Composition of the nitrogen free extracts was not significantly ($p>0.05$) influenced by the processing method. The average carbohydrate value of 53% obtained in this experiment is quite high and superior to those of soybean (22.12%) (Ezeagu *et al*., 2003) and most common legumes, suggesting that HEBM could be suitable an alternative protein source in monogastric diets.

The effect of different processing techniques on the concentration of some important Anti-Nutritional Factors (ANFs) in the horse eye bean is presented in Table 2.

The phytic acid content varied from 380 mg/100 g in treatment E to 820 mg/100 g in the control. The decrease in the phytic acid content by soaking and cooking of pre-soaked bean may be due to leaching out of this compound in water. The high level of phytate present in the raw HEBM might decrease the bioavailability of minerals (Ca^{2+} and Fe^{2+}) (Akpattu and Nelligaswatta, 2005). Phytate has also been implicated in decreasing protein digestibility by forming complexes and also by interacting with enzymes such as trypsin and Pepsin (Reddy and Pierson, 1994). Tannins reduced significantly ($p<0.05$) from 122 mg/100 g in raw sample to 80 mg/100 g, representing 34.43% in treatment F. Loss of tannin may be due to its solubility in water and its sensitivity to heat during boiling and toasting (Ahmed *et al*., 2006; Esenwah and Ikenebomeh, 2008). Igboeli *et al*. (1997) reported 35.9 to 43.6 percentage reduction in tannin content during the processing of lablab seed (*Adansonia digitata*) by dehulling, cold water, hot water, hot alkali and acid treatments which agree with the findings of this study. Esenwah and Ikenebomeh (2008) reported higher reduction level of 59.8% for tannin content of processed Locust bean. El-Adawy (2002) reported 48 to 50% reduction during processing of chick pea by different processing methods. The percentage reduction in the composition of tannins reported by this author is higher than the range obtain in this experiment and could be attributed to the processing method employed. Tannin inhibits the activities of some enzymes like trypsin, amylase and lipase by forming insoluble complexes with protein (Griffiths, 1979) and divalent ions such as Fe^{2+} and Zn^{2+}, thereby reducing their absorption in the body (Elegbede, 1998). Total phenol was reduced from

Table 2: Effect of processing on anti-nutritional factors of the horse eye bean (*Mucuna urens*) meal

	Treatment groups							
Anti-nutritional factors	Raw	A	B	C	D	E	F	SEM
Phytate (mg/100 g)	820.00^{a}	540.00^{b}	460.00^{b}	480.00^{b}	420.00^{b}	380.00^{b}	420.00^{b}	±52.12
Tannins (mg/100 g)	122.00^{a}	96.00^{b}	92.00^{b}	92.00^{b}	86.00^{b}	82.00^{b}	80.00^{b}	±4.90
Total phenols (mg/100 g)	38.42^{a}	32.16^{c}	33.44^{b}	21.23^{d}	15.36^{e}	13.22^{f}	10.38^{g}	±3.90
Hydrogen cyanide (HCN) (mg/100 g)	56.30^{a}	49.24^{b}	48.66^{b}	25.40^{c}	21.48^{d}	15.20^{e}	15.20^{e}	±5.74
Oxalates (mg/100 g)	33.00^{a}	16.80^{b}	16.40^{b}	16.30^{b}	76.00^{d}	6.20^{d}	6.72^{d}	±3.39

Means with different superscripts within the same row are significantly different ($p<0.05$). Means are values of triplicate determinations
SEM: Standard error of mean

38.4% in control group to 10.38% in treatment F, representing 72.98% reduction. The rate of reduction increased significantly ($p<0.05$) with increasing duration of soaking and cooking. This indicates that polyphenols are heat labile. Total poly-phenol concentration recorded in the raw bean sample was similar with 40.17 mg GAE g^{-1} reported by Ogbunugafor *et al.* (2011). Iyayi *et al.* (2005) reported a higher percentage (67-79) reduction in polyphenols of *Mucuna pruriens* toasted at 100°C for 1 h. Hydrogen Cyanide (HCN) was significantly ($p<0.05$) reduced from 56.30 mg/100 g in the raw sample to 15.20 mg/100 g in Treatments E and F, representing 73% reduction. Reduction in the values of HCN with processing can be explained by the report of Okolie and Ugochukwu (1989). The authors stated that processing drastically reduces the cyanide levels in foods. Onwuliri and Obu (2002) also noted that prolonged processing like water soaking; cooking, discarding water used and removal of testa could reduce HCN (Omafuvbe *et al.*, 2004). Processing caused significant ($p<0.05$) reduction in the concentration of oxalate. It was reduced from 33.0% in the raw bean meal to 6.20% treatment E (Table 2). Oxalate and phytates combine with phosphorus and calcium, respectively to form complexes and render them available for absorption (Abdulrashid and Agwunobi, 2009).

Results of the mineral composition of the raw and processed horse eye bean are presented on Table 3.

Comparatively, all treatments had higher values for phosphorous, zinc and manganese, copper and iron than the raw sample. Treatment C retained more copper (3.42 mg/100 g) and Zinc (4.12 mg/100 g) while Treatment E retained more manganese (12.53 mg/100 g) and phosphorous (330 mg/100 g) than the raw bean sample. This implies that processing improved the concentrations of these minerals. However, raw bean contained higher values of potassium (650.0 mg/100 g) and sodium (8.00 mg/100 g), showing that some quantities may have been lost during processing (Table 3). The reduction in mineral content during soaking might be due to the leaching out of minerals in the soaking water (Rani and Hira, 1998; Samantray *et al.*, 1989).

The reduction in the mineral contents with toasting might be attributed to loss of nutrients while treating at high temperature (Malik *et al.*, 2002; Rani and Hira, 1998).

The results of the amino acids composition of the raw and processed HEBM are presented in Table 4.

Glutamic acid, aspartic acid and leucine were the major amino acids in the raw HEBM and had values of 13.24, 8.69 and 9.75 mg100 protein, respectively.

Relative to FAO reference protein (egg protein) pattern (FAO/WHO, 1990), the limiting amino acids were found to be the sulfur amino acids (methionine + cystine). The lysine content of the raw horse eye bean meal was 6.11 mg/100 g protein which is slightly higher than 5.8 mg/100 g protein reported for the FAO reference protein (FAO/WHO, 1990).

Table 3: Mineral composition of the differently processed horse eye bean (*Mucuna urens*) meal (m g/100)

	Treatment groups							
Elements	Control	A	B	C	D	E	F	SEM
Calcium	240.00	240.00	230.00	220.00	240.00	240.00	240.00	±2.75
Magnesium	90.00	80.00	70.00	80.00	70.00	90.00	80.00	±2.64
Potassium	650.00^{a}	350.00^{b}	300.00^{b}	120.00^{e}	100.00^{e}	50.00^{d}	50.00^{d}	±10.98
Sodium	8.00^{a}	7.00^{ab}	6.00^{b}	4.00^{e}	3.00^{e}	2.00^{cd}	3.00^{e}	±0.80
Phosphorus	266.00^{e}	202.00	242.00^{f}	320.00^{b}	330.00^{d}	330.00^{a}	310.00^{c}	±16.11
Zinc	3.21^{b}	3.31^{b}	3.83^{a}	4.12^{a}	2.71	2.29^{c}	2.44^{c}	±0.24
Copper	11.31^{b}	12.83^{a}	3.08^{a}	3.42^{a}	2.28^{b}	1.94	8.62^{d}	±0.75
Iron	6.98^{d}	9.04^{c}	6.86^{f}	7.84^{e}	9.63^{b}	11.31^{a}	9.08^{c}	±0.16
Manganese	2.74^{b}	2.92^{ab}	3.08^{a}	3.42^{a}	2.28^{b}	12.53^{b}	8.62^{d}	±0.19

Means with different superscripts within the same row are significantly different ($p<0.05$). Means are values of triplicate determinations
SEM: Standard error of mean

Table 4: Effect of multi-processing on the amino acids profile of the horse eye bean (*Mucuna urens*) meal (g/100 g)

	Treatment groups							
Amino acids	Raw	A	B	C	D	E	F	SEM
Lysine*	6.11	5.19	6.00	6.51	6.00	6.24	5.56	±0.15
Histidine*	2.37	2.02	2.05	1.70	1.99	2.02	1.83	±0.07
Arginine	6.21	4.32	5.01	6.04	5.52	5.87	4.49	±0.26
Aspartic acid	8.69	5.58	6.58	6.51	6.20	7.01	6.82	±0.34
Threonine*	3.20	1.93	2.59	4.08	2.65	3.53	1.99	±0.28
Serine	2.61	1.85	1.75	2.53	2.05	2.69	1.61	±0.16
Glutamic acid	13.24	15.11	14.20	13.75	14.66	13.97	15.68	±0.29
Proline	2.65	2.03	1.83	3.05	1.83	3.26	2.24	±0.14
Glycine	4.04	2.11	3.05	3.41	3.10	3.56	2.35	±0.24
Alanine	3.91	3.34	4.03	3.84	4.37	3.19	2.70	±0.20
Cystine*	0.53	0.53	0.53	0.66	0.66	0.69	0.69	±0.06
Valine*	4.19	3.93	3.99	3.21	4.19	4.53	3.47	±0.16
Methionine*	1.07	0.83	0.99	0.73	0.68	0.70	0.57	±0.06
Isoleucine*	3.83	2.32	2.79	3.04	2.70	3.11	2.57	±0.17
Leucine*	9.75	7.36	8.59	10.16	7.52	8.04	10.05	±0.86
Tyrosine*	2.84	3.00	2.53	3.32	3.32	2.37	3.00	±0.13
Phenylalanine*	5.14	4.46	4.97	4.28	4.37	4.62	4.62	±0.11

*Essential Amino acids, SEM: Standard error of mean

Different processing methods caused substantial reduction of some of the essential amino acids. For instance, Histidine value was reduced from 2.37% in the raw bean to 1.70% in treatment C. While threonine was reduced from in control group to 1.99% in treatment F. Methionine and isoleucine were reduced from 1.07 and 3.83 mg/100 g in the raw bean sample to 0.57% and 2.57 mg/100 g, in Treatment F, respectively. Reduction of methionine and other amino acids during prolonged cooking (90 min) was in agreement with the report of Ziena *et al.* (1991) and Siddhuraju and Becker (2005). The authors attributed the changes in amino acids profile of the HEBM to Transamination and deamination reactions.

CONCLUSION

From the results of this study, it was concluded that 72 h soaking, 90 min cooking and toasting was most effective method in reducing the anti-nutritional factors in the HEBM, followed by 48 h

soaking, 90 min cooking and toasting 24 h soaking, 60 min cooking and toasting was least effective in reducing the anti-nutritional factors in the HEBM. Marginal loss in the relevant nutrients were observed in 72 h soaked, 90 min cooked and toasted HEBM and 48 h soaked, 90 min cooked and toasted HEBM relative to HEBM in other treatments.

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