

## EVALUATION OF THE PHYTOCHEMICAL AND ANTINUTRIENT COMPOSITION OF RAW AND PROCESSED *MUCUNA PRURIENS* (VELVET BEANS)

Anthony Chinedu **Ogudoro**<sup>1\*</sup>, Abubakar Ndaman **Saidu**<sup>1</sup>, Adamu Yusuf **Kabiru**<sup>1</sup>

Biochemistry Department, Federal University of Technology, Minna, Niger State, Nigeria

E-mail\*: [togudoro@gmail.com](mailto:togudoro@gmail.com)

### Abstract

Raw and processed *Mucuna pruriens* were screened for phytochemical constituents and antinutrients composition. This was done using standard analytical procedures. Processing procedures included fermentation, boiling, autoclaving, roasting, microwaving and dehulling. All the phytochemicals screened were present in all the samples and most of them were decreased as a result of processing. Antinutrients evaluated included Tannic acid, Phytate, Saponins, Cyanide, Phenols, Oxalate, Flavonoid and Alkaloids. The phytate concentration of the raw mucuna beans (4.210 g/100g) was reduced to 1.372 g/100g by roasting while it was increased to 6.576 g/100g by dehulling. Also, dehulling (0.0533 g/100g) significantly ( $p < 0.05$ ) reduced the saponin content of the raw mucuna beans. Total phenols content of the raw bean (1.6875 g/100g) was also significantly ( $p < 0.05$ ) reduced by dehulling (0.9296 g/100g). Processing also caused significant reductions in the oxalate composition of the raw *Mucuna pruriens*. Cyanide content of the raw mucuna beans was not affected by processing it. The levels of some of the antinutrients found in the present study suggest that they may be significantly higher than previously shown in earlier ones. This research however suggests that processing has the potential to reduce the antinutrient and phytochemical presence in mucuna pruriens to levels that may be manageable and even beneficial to human and animal health.

**Keywords:** *Mucuna pruriens*; phytochemicals; antinutrient composition; fermentation; microwaving.

Submitted: 09.01.2014

Reviewed: 16.04.2014

Accepted: 14.05.2014

## 1. INTRODUCTION

*Mucuna pruriens* is commonly known as velvet beans or *mucuna* beans in Nigeria. It is a highly productive black seeded tropical legume that is relatively unknown and under-utilized as human and animal feed despite its seeds being a potential source of protein, minerals, dietary fibre and vitamins. The seeds of *Mucuna pruriens* can be used to bridge the protein energy nutrition gap especially in the developing world. However, despite the obvious potential of the beans, there are a variety of antinutrients found in the legume which causes consumption of unprocessed or poorly processed seeds to result in various ailments in humans and animals. The anti-nutritional factors in the legume seeds adversely affect protein digestibility (Gupta, 1987). These substances, unless destroyed by heat or some other suitable treatment, can exert effects that may be physiologically harmful when ingested by man and animals (Liener, 1980). On the contrary, it has been suggested that, consumption of low levels of certain

antinutrients may produce health benefits while avoiding some of the adverse effects associated with their large intake (Thompson, 1988). A clinical study confirmed the efficacy of the velvet bean seeds in the management of Parkinson's disease by virtue of their L-dopa content (Bell et al., 1971; Manyam, 1995). The phytic acid of *Mucuna pruriens* possesses antioxidant, anticarcinogenic and hypoglycemic activities (Graf and Eaton, 1990; Shamsuddin et al., 1997) and they are effective at low concentrations. To increase biological benefits, ease of digestion and decrease antinutrient compounds in legumes, including *Mucuna pruriens*, traditional procedures such as heating or blanching, soaking and fermentation, roasting are generally used. Most of the methods employed are based on the use of water and thermal treatments (Bressani, 2002; Diallo and Berhe, 2003; Gilbert, 2002). *Mucuna pruriens* is one of the plants that have been shown to be active against snake venom and, indeed, its seeds are used in traditional medicine to prevent the toxic effects of snake

bites, which are mainly triggered by potent toxins such as neurotoxins, cardiotoxins, cytotoxins, phospholipase A2 (PLA2), and proteases (Guerranti et al., 2002). The mechanisms of the protective effects exerted by *mucuna pruriens* seed aqueous extract (MPE), were investigated in detail, in a study involving the effects of *Echis carinatus* venom (EV) (Guerranti et al., 2002). In vivo experiments on mice showed that protection against the poison is evident at 24 hours (short-term), and one (1) month (long term) after injection of MPE (Guerranti et al., 2008). This may be further evidence of the presence of phytochemicals in the *mucuna* beans. Again, one important secret of longevity is maintaining optimal levels of key hormones as well as other phytochemical factors in the bloodstream. *Mucuna pruriens* has already been shown to be a natural growth hormone secretagogue (hgh) as well as a booster of other hormones of youth that tend to diminish with time. It has been found that blood plasma levels of some of the top antioxidants such as superoxide dismutase (SOD), catalase (an enzyme that prevents the hair from going gray!), glutathione (GSH) and ascorbic acid (vitamin C) are kept elevated in individuals who take this adaptogenic herbal aphrodisiac.

The present study tries to establish the presence of some phytochemicals in the flour made from raw and processed *mucuna pruriens* and to see the effect of the processing techniques on the levels of these phytochemicals and antinutrients present in the legume.

## 2. MATERIALS AND METHODS

Sample collection: The velvet bean seeds were cultivated at the International Institute of

Tropical Agriculture (IITA) Ibadan, Nigeria and purchased from the commodities store of the same institute.

All the samples were put into sterile, air-tight cellophane bags and stored in the refrigerator prior to their use.

### Phytochemical Screening:

Chemical analyses were done on the aqueous extracts of the powdered samples using standard procedures to identify the constituents as described by Sofowara (1993), Trease and Evans (1989) and Harborne (1973).

Test for tannins: 0.5 g of the dried powdered samples was boiled in 20 ml of water in a test tube and then filtered. A few drops of 0.1% ferric chloride was added and observed for brownish green or a blue-black colouration.

Test for saponin: 2 g of the powdered sample was boiled in 20 ml of distilled water in a water bath and then filtered. 10ml of the filtrate was mixed with 5 ml of distilled water, shaken vigorously and observed for a stable persistent froth. The frothing was mixed with 3 drops of olive oil, shaken vigorously again, and then further observed for the formation of emulsion which confirmed the presence of saponins.

Test for flavonoids: A portion of the powdered plant sample was in each case heated with 10 ml of ethyl acetate over a steam bath for 3 min. The mixture was filtered and 4 ml of the filtrate was shaken with 1 ml of dilute ammonia solution. A yellow colouration was observed indicating a positive test for the presence of flavonoids.

Test for phenol (Lead Acetate test): A total of 1 ml of alcoholic or aqueous extract was diluted to 5 ml with distilled water; a few drops of 1% aqueous solution of lead acetate were added. A yellow precipitate was formed to indicate the presence of phenols.

### Sample preparation

Raw Velvet bean	→ Sun-dried 12H, Ground/Sieved	RwV
	→ Oven roasted, 2H at 100 <sup>0</sup> C, Ground/Sieved	RsV
	→ Boiled, 3H at 100 <sup>0</sup> C, sun-dried, ground/sieved	BoV
	→ Soaked, 72H; water changed every 24H, ground/sieved	FeV
	→ Microwaved for 10min, ground/sieved	MwV
	→ Autoclaved, 2H; sun-dried; ground /sieved	AcV
	→ Dehulled, Sundried; ground/ sieved	DeV

Test for Alkaloid: 0.5g of the extract was stirred with 5cm<sup>3</sup> of 1% aqueous HCl on a steam bath, few drops of picric acid solution was added to 2cm<sup>3</sup> of the extract. The formation of a reddish brown precipitate was taken as a preliminary evidence for the presence of alkaloids.

Test for Glycosides: To 1ml of the test solution, 2 drops of Conc. Sulphuric acid was added and placed in water bath for about 15 minutes. A few drops of 20% KOH were added to make the solution alkaline. To this solution, few drops of concentrated FeCl<sub>2</sub> were added. The formation of a brick red precipitate indicates the presence of glycosides.

#### **Quantitification of Anti-Nutrients**

##### *Determination of Phytic Acid:*

Phytic acid content was determined by a modification of the method described by Wheeler and Ferrel (1971). Phytic acid was extracted from 1g of the sample with 50ml of 3% Trichloro acetic acid (TCA) by shaking at room temperature followed by high speed centrifugation. The phytic acid in supernatant was precipitated as ferric phytate by adding excess ferric chloride and centrifuged. The ferric phytate was converted to ferric hydroxide by adding a few ml of water and 5ml of 1N NaOH to 1ml of the filtrate. The precipitate was dissolved in 4ml of hot 3.2N HNO<sub>3</sub> and the colour read immediately at 480nm. The iron content present in the sample was estimated from a standard solution prepared from Fe(NO<sub>3</sub>) and the Iron content was extrapolated from the Fe(NO<sub>3</sub>) standard curve. The phytate phosphorus was calculated from the iron results assuming a 4: 6 iron: phosphorus molecular ratio. The phytic acid was estimated by multiplying the amount of phytate phosphorus by the factor 3.55 based on the empirical formula C<sub>6</sub> P<sub>6</sub> O<sub>24</sub> H<sub>18</sub>

##### *Determination of Saponins:*

0.5 g of the sample was added to 20 ml of 1N HCl and was boiled for 4H. After cooling it was filtered and 50 ml of petroleum ether was added to the filtrate for ether layer and evaporated to dryness. 5 ml of acetone ethanol was added to the residue. 0.4 mls of each was taken into 2 different test tubes. 6 ml of Ferrous

sulphate reagent was added into them followed by 2 ml of conc H<sub>2</sub>SO<sub>4</sub>. It was thoroughly mixed after 10 min and the absorbance was taken at 490 nm (Oloyed, 2005).

##### *Determination of Oxalate:*

A modification of the titrimetric method of Day & Underwood (1986) was used in the determination of oxalate in the Velvet bean samples. 75 ml of 3 N H<sub>2</sub>SO<sub>4</sub> was added to 1 g of the ground Velvet bean samples and the solution was carefully stirred intermittently with a magnetic stirrer for 60 minutes and filtered using Whatman No 1 filter paper, after which 25 ml of the filtrate was collected and titrated against hot (90<sup>o</sup>C) 0.1 N KMnO<sub>4</sub> solution until a faint pink color appeared that persisted for 30 seconds. The concentration of Oxalate in each sample was obtained from the calculation: 1ml of 0.01M KMnO<sub>4</sub> = 0.0006303g Oxalate.

##### *Determination of Alkaloids*

The quantitative determination of alkaloids was carried out by the alkaline precipitation through Gravimetric method described by Harbone (1973). Two grammes (2g) of the sample was soaked in 20ml of 10% ethanolic -acetic acid. The mixture was allowed to stand for 4 hr at room temperature. Thereafter, the mixture was filtered through Whatman filter paper no. 40. The filtrate (extract) was concentrated by evaporation over a steam bath to a quarter of its original volume. For the alkaloids to be precipitated, concentrated ammonia solution was added in drops to the extract until it was in excess. The resulting alkaloid precipitate was recovered by filtration using a previously weighed filter paper. After filtration, the precipitate was washed with 1% ammonia solution and dried in the oven at 600C for 30min, cooled in a desiccator and reweighed. The experiment was repeated two more times and the average was taken. The weight of alkaloids was determined by difference and expressed as a percentage of the weight of the sample analysed as shown.

$$\% \text{ Alkaloids} = \frac{W_2 - W_1}{\text{Wt of sample}} \times 100$$

W1 = Weight of Filter paper

W2 = Weight of paper + alkaloid precipitate

#### *Determination of Hydrogen cyanide*

The extraction was according to Wang and Filled method as described by Onwuka (2005). A portion (5 g) of sample was made into paste and the paste was dissolved into 50 ml distilled water and left overnight. The extract was filtered and the filtrate was used for cyanide determination. To 1 ml of the sample filtrate, 4 ml of alkaline picrate was added and absorbance was recorded at 490 nm and cyanide content was extrapolated from a standard cyanide (KCN) curve.

#### *Determination of Total Phenols by spectrophotometric method according to the method reported by (Mugendi et al., 2010)*

2 g of the sample were defatted with 100 ml of diethyl ether using a soxhlet apparatus for 2 h. The fat free sample was boiled with 50 ml of ether for the extraction of the phenolic component for 15 min. 5 ml of the extract was pipetted into a 50 ml flask, then 10 ml of distilled water was added. 2 ml of ammonium hydroxide solution and 5 ml of concentrated amylalcohol were also added. The samples were made up to mark and left to react for 30 min for colour development. This was measured at 505 nm. Tannic acid was used to establish the calibration curve.

#### *Determination of Total Flavonoid:*

Total flavonoid was determined using aluminium chloride colorimetric method (Chang et al., 1994) Quercetin was used to establish the calibration curve. Exactly 0.5ml of the diluted sample was added into a test tube containing 1.5ml of methanol. 0.1ml of 10% AlCl<sub>3</sub> solution and 0.1ml sodium acetate (NaCH<sub>3</sub>COO-) were added, followed by 2.8ml of distilled water. After incubation at room temperature for 30 min, the absorbance of the reaction mixture was measured at 415nm. The amount of 10% AlCl<sub>3</sub> was substituted by the same amount of distilled water in blank.

#### *Determination of Tannin*

0.2 g of sample was measured into a 50ml beaker. 20ml of 50% methanol was added and covered with para film and placed in a water bath at 77-80°C for 1 hr. It was shaken thoroughly to ensure a uniform mixture. The extracted was quantitatively filtered using a

double layered whatman No. 41 filter paper into a 100ml volumetric flask, 20ml water added, 2.5ml Folin-Denis reagent and 10ml of 17% Na<sub>2</sub>CO<sub>3</sub> were added and mixed properly. The mixture was made up to mark with water mixed well and allowed to stand for 20min. A bluish-green colour which developed at the end of range 0-10ppm was measured. The absorbances of the tannic acid standard solutions as well as samples were read after colour development on a UV-Vis spectrophotometer model 752 at a wavelength of 760nm (AOAC, 1984).

#### **Statistical analysis:**

The analysis was carried out in duplicates for all determinations and the results of the duplicates were expressed as mean ± SEM. Data was exported from excel and analyzed using the Statistical Package for Social Sciences (SPSS) Version 16 (SPSS Inc., Chicago, IL USA) for windows Computer. The Software Package was also used for the Analysis of Variance (ANOVA). Statistical differences between means were compared using paired Duncan HSD. Differences in means were considered statistically significant at  $p < 0.05$ .

### **3. RESULTS AND DISCUSSIONS**

The result of the phytochemical screening showed that Tannin was highly present in the raw sample. The presence of tannin was however, reduced as a result of the processing techniques applied on the raw velvet beans. Saponin, another phytochemical was also detected in all the samples analysed. While the raw sample had a moderate presence for the phytochemical, the dehulled sample also showed a slight presence for it. The other samples (fermented, boiled, autoclaved, micro-waved and roasted) had slight (+) presence for saponin. Flavonoid was also moderately present (++) in the raw unprocessed velvet beans. The other processing methods reduced the presence of flavonoids to slight (+) in the other samples except the dry heat treatments

which showed moderate (++) presence of flavonoids.

For phenol, there was also a high (+++) presence in the raw, microwaved and roasted samples. The boiled and auto-claved samples had moderate (++) presence while the fermented and dehulled samples had a slight

(+) presence for the phytochemical. Alkaloids were moderately (++) present in the raw, boiled and autoclaved samples. The other treatment and processing methods reduced the alkaloid presence to slight (+) in the other samples. Glycosides are also slightly (+) present in all the samples analysed including the raw sample.

**Table 1. Phytochemical screening of the raw and processed velvet bean samples**

Sample	Tannin	Saponin	Flavonoid	Phenol	Alkaloid	Glycosides
RwV	+++	++	++	+++	++	+
FeV	+	+	+	+	+	+
BoV	+	+	+	+	+	+
AcV	+	+	+	+	+	+
MwV	+	+	++	+++	+	+
RsV	+	+	++	+++	+	+
DeV	+	+	+	+	+	+

KEY: + Low presence. ++ moderately presence. +++ high presence

**Anti-nutrient content in raw and processed velvet bean samples**

**Table 2a: Anti-nutrient content in the Raw and treated Velvet bean samples (g/100g).**

Sample	Tannic	Phytate	Saponin	Cyanide
RwV	0.741 <sup>d</sup> ±0.03	4.210 <sup>e</sup> ± 0.45	0.1335 <sup>d</sup> ±0.012	0.233 <sup>b</sup> ±0.005
FeV	0.280 <sup>a</sup> ±0.06	3.565 <sup>d</sup> ±0.08	0.0975 <sup>b,c</sup> ±0.011	0.231 <sup>b</sup> ±0.001
BoV	0.406 <sup>a,b</sup> ±0.02	2.282 <sup>c</sup> ± 0.08	0.1045 <sup>b,c</sup> ±0.017	0.217 <sup>a</sup> ± 0.008
AcV	0.419 <sup>a,b</sup> ±0.02	1.720 <sup>b</sup> ± 0.16	0.956 <sup>b</sup> ± 0.008	0.216 <sup>a</sup> ±0.005
MwV	0.583 <sup>c</sup> ±0.05	1.744 <sup>b</sup> ± 0.13	0.1093 <sup>b,c</sup> ±0.021	0.217 <sup>a</sup> ±0.010
RsV	0.508 <sup>b,c</sup> ±0.01	1.372 <sup>a</sup> ± 0.13	0.1153 <sup>c,d</sup> ±0.011	0.215 <sup>a</sup> ±0.012
DeV	0.345 <sup>a</sup> ±0.03	6.576 <sup>f</sup> ± 0.26	0.0533 <sup>a</sup> ± 0.011	0.238 <sup>b</sup> ± 0.013

\*\*Results represent mean ± SEM of two replicates. Results in the same column with different letter superscripts are significantly different (p<0.05).

**Table 2b: Anti-nutrient content in the Raw and treated Velvet bean samples (g/100g)\*.**

Sample	Phenols	Oxalate	Flavonoid	Alkaloids
RwV	1.6875 <sup>d</sup> ±0.21	18.50 <sup>f</sup> ±1.10	4.25 <sup>a,b</sup> ±0.0045	9.00 <sup>b</sup> ±1.10
FeV	0.9922 <sup>a</sup> ± 0.23	8.98 <sup>b</sup> ±0.30	2.70 <sup>a</sup> ±0.0025	3.50 <sup>a</sup> ± 0.50
BoV	1.4140 <sup>c</sup> ±0.07	5.71 <sup>a</sup> ±0.30	2.93 <sup>a</sup> ±0.0065	6.00 <sup>a,b</sup> ± 0.50
AcV	1.2423 <sup>b</sup> ±0.12	12.61 <sup>d</sup> ±0.91	2.80 <sup>a</sup> ±0.0020	7.00 <sup>a,b</sup> ±0.50
MwV	1.8007 <sup>d,e</sup> ±0.11	10.84 <sup>c</sup> ±0.60	4.48 <sup>a,b</sup> ±0.0060	3.75 <sup>a</sup> ±1.25
RsV	1.8914 <sup>e</sup> ± 0.14	9.35 <sup>b</sup> ±0.90	5.35 <sup>b</sup> ±0.0155	3.75 <sup>a</sup> ±1.25
DeV	0.9296 <sup>a</sup> ± 0.13	14.12 <sup>e</sup> ±0.72	3.00 <sup>a</sup> ±0.0450	4.75 <sup>a,b</sup> ± 1.25

\*Results represent mean ± SEM of two replicates. Results in the same column with different letter superscripts are significantly different (p<0.05).

The anti-nutrient composition of the samples is shown in table 2a and 2b. There was a significant decrease ( $P < 0.05$ ) in the tannin content of micro-waved sample) 0.583 g/100g and roasted 0.508 g/100g. Phytates was also present in the samples ranging from 6.576 g/100g for the dehulled sample to 1.372 g/100g for the roasted sample. The results for saponin ranged from 0.1335 g/100g to 0.0533g/100g for the raw and dehulled samples respectively. Saponin content of the roasted samples (0.1153 g/100g) showed there was no significant difference ( $P < 0.05$ ) from the control. There was a significant different ( $P < 0.05$ ) in the cyanide content of the boiled (0.217 g/100g), auto-claved (0.216 g/100g), microwaved (0.217 g/100g) and roasted (0.215 g/100g) samples when compared to the control (0.233 g/100g). The alkaloid content was significantly reduced ( $P < 0.05$ ) in the fermented sample (3.500g/100g).

Phytochemical screening is very an important tool in determining some biologically active and medicinal components of vegetables and plants. The results of the phytochemical screening of the raw and processed velvet bean samples indicate that the raw sample had a tannin content showing high presence of the phytochemical (+++). The results also showed that Tannin was slightly present in the other samples (+). Tannins have been known to exhibit anti-inflammatory and anti-ulcer effects in rodents. This suggests it possesses a strong antioxidant and therapeutic uses. Foods that are rich in tannins can be used in the management of HFE hereditary hemochromatosis (a hereditary disease that is characterized by excessive absorption of dietary iron, resulting in an increase in the body's iron stores. Processing the raw *mucuna* decreases the tannin content. The saponin content was also reduced from its moderate presence (++) in the raw sample to slight presence (+) in the processed samples. This is with the exception of the roasted sample that has a moderate presence (++) for saponin. Saponins have been known to negatively interfere with sodium ion ( $\text{Na}^+$ ) efflux by locking the entrance of the  $\text{Na}^+$  out of the cells. This lockage leads to an

increase in the  $\text{Na}^+$  concentration in the cells, leading to an activation of  $\text{Na}^+$ -  $\text{Ca}^{2+}$  anti porter in heart muscles. This increase in  $\text{Ca}^{2+}$  in- flux through this anti porter strengthens the contractions of the cardiac muscles (Schneider and Woliling, 2004). Microwave treatment increased the phenolic content in the germinated seeds of faba bean (Randir and Shetty, 2004). It was suggested that microwave caused acute heat stress in plant cells which stimulated Pentose Phosphate Pathway (PPP) towards the production of more phenolics and L-dopa. Phenolics have been suggested to exhibit health related functional properties such as, anticarcinogenic, antiviral, antimicrobial, anti-inflammatory, hypertensive and antioxidant activity (Shetty, 1997). The flavonoid and alkaloid presence of the *mucuna* bean samples are also shown on table 1. They were both reduced from a moderate presence (++) in the raw sample to slight presence (+) in the processed samples except the roasted and microwaved samples which had moderate presence (++) for the phytochemical. Alkaloids have been used to stimulate the Central Nervous System and as a powerful pain reliever. It possesses anti-puretic activity and has also been used as a topical anaesthetic in ophthalmology (Heikens *et al.*, 1995). Flavonoid may confer protection against some diseases by contributing (with antioxidant vitamins and enzymes) to the total antioxidant defense system of the body. Studies have shown flavonoids and carotenoids intake to be inversely related to mortality from heart diseases and the incidence of heart failures. There was moderate presence (+) of glycosides in all the samples analysed. Glycosides are known to be of varied health benefits including maintenance of cardiac health. Oxidative stresses have long been linked to cancer, ischemic injury and neuro-degenerative diseases such as Parkinson's and Alzheimer's diseases (Paloza, 1998) and these phytochemicals help in the body's immune responses.

The Tannic acid content for the raw sample (0.741g/100g) was higher than the 2.31mg/100g reported by Mugendi *et al.*,

(2010). This may be due to differences in specie and cultivation conditions. The Tannic acid content of the *mucuna* bean samples were generally reduced by processing with Fermentation (0.280 g/100g) causing the most reduction. Dry heating (micro-waving 0.583g/100g and roasting 0.508 g/100g) had the least effect in reducing the Tannic acid content in the *mucuna* bean samples. This result is in accordance with the work done by Kala and Mohan (2012) who postulated the limited effect of dry heating on tannic acid removal in legumes. The consumption of tannins by animals resulted in diminished consumption of feeds by the animals. It also binds dietary protein and some digestive enzymes forming undigestible complexes (Aletor, 1993). Tannins also decreased palatability of animal feed and their growth rate (Roeder, 1995).

Phytate content of 4.210g/100g obtained for the raw *mucuna* sample was also higher than the 0.82 g/100g for raw *mucuna* bean and 1.35 g/100g for dehulled *mucuna* bean (Mugendi et al., 2010). There was also a decreasing trend in the phytate content of the *mucuna* samples as a result of treatments. The roasted and microwaved samples showed the least presence of the phytic acid (1.372g/100g and 1.720g/100g). Decrease of phytic acid content as a result of processing has been attributed to low inositol and inositol phosphate by the action of free radicals generated during roasting and irradiation. Therefore, microwave and roasting treatments proved to be more effective in lowering phytic acid level. But simply dehulling the *mucuna* beans actually increased the phytic acid content (Mugendi et al., 2010). Phytates are known to bind many minerals like calcium, iron, magnesium and zinc, making them unavailable for normal body processes. Saponin content of 0.1333g/100g in the raw sample was however lower than the 1.3% obtained by Siddhuraju and Becker (2005). This may also be a result of the different accessions used for the analyses. Fermentation (0.0975 g/100g) reduced the saponin content the most as shown on table 2a. Wet heating was also discovered to be the

better method of reducing saponin than dry heating. Saponins possess a carbohydrate moiety attached to a tri-terpenoid or a steroidal aglycone. They form a group of compounds, which on consumption causes deleterious effects such as hemolysis and permeabilization of the intestine (Cheeke et al., 1986 ; Price et al., 1987). Excessive saponins caused hypocholesterolaemia in animals by binding cholesterol thereby making it unavailable for absorption. It also caused haemolysis of red blood cells of rats (Johnson et al., 1986).

Cyanide content of the raw velvet beans was shown to be 0.233g/100 of sample. This result was also higher than the 0.24mg/100g reported by Kala and Mohan (2012). All the heat treatments significantly ( $p < 0.05$ ) reduced the cyanide content of the *mucuna* samples except fermentation and dehulling, indicating that the effect of fermentation on the cyanide reduction of the raw sample was minimal. This is unlike legumes and cereals with a softer seed coat as the tough seed coat of the *mucuna* seed may make it difficult for water penetration during fermentation. The observed cyanide level was also above the 36mg/100g generally regarded as safe for consumption in foods. The levels of cyanide in the samples seem to be high when compared with lethal level for hydrogen cyanide which is 36 mg/100 g. Consumption of excess *mucuna* beans may result to ill- health. The toxic effects of the free cyanide are well documented and affect a wide spectrum of organisms since their mode of action is inhibition of the cytochromes of the electron transport system (Laurena et al., 1994). Cyanogenic glucosides are hydrolysed to yield toxic hydrocyanic acid (HCN). The cyanide ions depress several enzyme systems, inhibit growth of animals through interference with several essential amino acids and suppress the optimum utilization of nutrients resulting in acute toxicity, neuropathy and death (Osuntokun, 1972; Fernando, 1987). The total phenolics content of the raw *mucuna* beans was 1.6875g/100g. This was lower than the 4.06g/100g reported by Kala and Mohan (2012) and 7.09g/100g also reported by Mugendi et al.,(2010). The result was also

lower than 3.82% reported for cowpea (Preet and Punia, 2005), and 10.6% reported for pigeon pea (Singh, 1988). The phenolics content of the dry heated (microwaved and roasted) samples were significantly ( $p < 0.05$ ) increased at 1.8007g/100g and 1.8914g/100g respectively. Phenolic compounds decrease digestibility of carbohydrates and availability of vitamins and minerals. They lower the activity of the digestive enzymes such as amylase, trypsin, chymotrypsin, and lipase and may cause damage to mucosa of the digestive tract and reduce absorption of nutrients such as vitamin B12. Phenolics also interfere with digestion and absorption of proteins (Liener, 1994).

Oxalate content of the *mucuna* samples showed a concentration of 18.50g/100g. This result indicates a high oxalate content which was much higher than the 0.12mg/100g reported by Kala and Mohan (2012). The difference in oxalate content could be as a result of specie differences and conditions of cultivation. All the treatment methods reduced oxalate content. Table 2b also indicated that wet heating (boiling and auto claving) and fermentation were most effective in reducing the oxalate content of the sample. Excessive body concentration of soluble oxalate prevents the body's absorption of calcium ions because the oxalate binds the calcium ions to form a calcium-oxalate complex which is insoluble. People with the tendency to form kidney stones are mostly advised to avoid oxalate-rich foods (Adeniyi et al., 2009). Dehulling the *mucuna* bean flour did not have an appreciable reduction in the oxalate content. This suggests that the Oxalate may be concentrated in the seed cotyledon.

The flavonoid content of the raw velvet bean (4.25g/100g) was higher than the 0.42g/100g obtained by Nwaoguikpe et al., (2011). The difference in flavonoid content may also be attributed to differences in seed species and cultivation conditions. Wet heating was effective in removing the flavonoid but dry heating actually increased it. Dry heating (roasting and microwaving) actually releases the aromatic and flavour enhancing compounds

in foods. Dehulling did not have a significant effect on flavonoids removal.

The alkaloids content of 9.00g/100g was higher than the 1.09g/100g obtained by Nwaoguikpe et al., (2011). Fermentation significantly ( $p < 0.05$ ) reduced this alkaloid content to 3.5g/100g. This reduction was the most in the samples evaluated. Dry heat treatments (roasting and microwaving) also significantly reduced the alkaloids contents of the samples. Alkaloids caused gastrointestinal and neuron disorders (Aletor, 1993). The glycoalkaloids present in some root crops like potatoes (Saito et al., 1990; Aletor, 1991) are known to be toxic to fungi and animals. Toxicity expressions of potato glycoalkaloids involve intestinal upsets and neurological disorders. The toxicity is manifested in doses exceeding 20 mg/100g in a sample. This makes it imperative for its reduction from foods before consumption. The alkaloid content for all the samples evaluated showed that they were above this critical limit.

#### 4. CONCLUSIONS

*Mucuna pruriens* contained a variety of healthy phytochemicals which were reduced by processing it. The antinutrients evaluated were also reduced by processing the *mucuna* beans. Dry heating (roasting and microwaving) was more effective in reducing phytates, saponin and alkaloids from the raw *mucuna* beans while fermentation was better for reducing tannin, phenols, and oxalates. A combination of processing techniques may be more useful in eliminating the antinutrients present in *mucuna* beans.

#### 5. REFERENCES

- [1] Adeniyi, S.A., Orjiekwe, C.L., Ehiagbonare, J.E., 2009. Determination of Alkaloids and Oxalates in some selected Food Samples in Nigeria. *African Journal of Biotechnology*. 8(1), 110-112.
- [2] Aletor, V.A., 1991. Anti-nutritional factors in some Nigerian Feedstuffs, Hbage byproducts, crop residues and browse plants. A monograph prepared for the Presidential task force on alternative formulation of livestock feeds; Product



- Development, Quality Evaluation and Health Implications.
- [3] Aletor, V.A., 1993. Allelochemicals in plant foods and feeding Stuffs. Part I. Nutritional, Biochemical and Physiopathological aspects in animal production. *Journal of Veterinary and Human Toxicology*. 35(1), 57-67.
- [4] A.O.A.C., 1984. Official method of analysis, Association of Official Analytical Chemistry Food Composition. Additives natural contaminants. Adrich R.C. (ed.) vol.1&2, 15<sup>th</sup> ed. Association of Official Analytical Chemistry Inc. USA.
- [5] Bell, E. A., Nulu, J.R., Cone, C., 1971. L-Dopa and L-3-carboxy-6,7-dihydroxy-1,2,3,4-tetrahydroisoquinoline, a new imino acid, from seeds of *Mucuna mutisiana*. *Journal of Phytochemistry*. 10, 2191-2194
- [6] Bressani, R., 2002. Factors influencing nutritive value in food grain legumes, *Mucuna* compared to other grain legumes. In, Food and Feed from *Mucuna*, Current Uses and the Way Forward (Editors, Flores B M, Eilittä M, Myhrman R, Carew L B and Carsky R J), Workshop, CIDICCO, CIEPCA and World Hunger Research Center, Tegucigalpa, Honduras (April 26-29, 2000), 164-188.
- [7] Chang, M.J, Collins, J.L, Baily, J.W., Coffey, D.L., 1994 . "Tannins Related to Cultivar, Maturity, Dehulling, and Heating," *Journal of Food Science*, 59 (5), 1034-1036.
- [8] Cheeke, P.R, Grobener, M.A., N.M. Patton., 1986. Fibre Digestion and Utilization in rabbits. *Journal of Applied Rabbit Research*. 9, 25-30.
- [9] Diallo, O.K, Berhe, T. 2003. Processing of *Mucuna* for human food in the Republic of Guinea. Dollery, C (Ed.). 1999. Therapeutic Drugs, 2nd Edition. Churchill Livingstone, New York, p L39-L43.
- [10] Fernando, R. 1987. Plant poisoning in Sri Lanka. In, Progress in venom and toxin research. Procedure of the 1st Asia-Pacific Congress in Animal, Plant and Microbial toxins, pp. 624-627.
- [11] Gilbert, R. 2002. *Mucuna pruriens* in Malawi, A promising legume with a troubled history. In, Food and Feed from *Mucuna*, Current Uses and the Way Forward (Editors, Flores B. M, Eilittä M, Myhrman R, Carew L. B., Carsky R J), Workshop, CIDICCO, CIEPCA and World Hunger Research Center, Tegucigalpa, Honduras (April 26-29, 2000), 48-59.
- [12] Graf, E., Eaton, J. 1990. Antioxidant functions of phytic acid. *Free Radical Biology and Medicine*. 8, 61-69.
- [13] Gupta, Y.P. 1987. Anti-nutritional and toxic factors in food legumes, A review. *Plant Foods for Human Nutrition*. 37, 201-228.
- [14] Harborne, J.B., 1973. Textbook of phytochemical methods, 1st Edn, Champraan and Hall Ltd. London. pp, 110-113.
- [15] Heikens, H., E. Fliers, E. Endert, M. Ackermans, G.Van Mont Frans, 1995. Liquorice-induced hypertension, a new understanding of an old disease, *Journal of Medicine*. 5, 230-234.
- [16] Johnson, I.T., Gee, J.M., Price, K., Curl, C., Fenwick, G.R. 1986. Influence of saponin on gut permeability and active nutrient transport *in vitro*. *Journal of Nutrition*. 116, 2270-2277.
- [17] Josephine, M.R., Janardhanan, K., 1992. Studies on chemical composition and anti-nutritional factors in three germplasm seed materials of the tribal pulse, *Mucuna pruriens* (L.) DC. *Journal Food Chemistry*. 43, 13-18.
- [18] Liener, I.E., 1994. Determination of antitryptic activity of soybean. *Journal of Science and Agriculture*. 16,602-609.
- [19] Liener, I.E. 1980. Heat – liable Anti-nutritional factors. In, Advances in legume science. (Eds.) Summerfield, R.J. and Bunting, A.H. Kew, Richmond, Surrey, UK, Royal Botanic Gardens. pp. 157-170.
- [20] Manyam, B., 1995. An alternative medicine treatment for Parkinson's disease, result of a multicenter for clinical trial. HP-200 in Parkinson's disease study group. *Journal of Alternative Complementary Medicine*, 1, 249-255.
- [21] Mugendi, J.B., Njagi, E.N., Kuria, E.N., Mwasaru, M.A., Mureithi, J.G., Apostolides z, 2010. Effects of processing technique on the nutritional composition and anti-nutrient content of mucuna bean (*Mucuna pruriens* L.). *African Journal of Food Science* Vol. 4(4), pp. 156 – 166.
- [22] Nelson, T.S., Ferrara, L.W., Stover, H.L. 1968. Effect of phytate on the calcium requirement of chicks. *Poultry Sciences*. 47, 185 - 189.
- [23] Nwaoguikpe, R.N., Braide, W., Ujowundu, C.O., 2011. The effects of Processing on the phytochemical Compositions of *Mucuna pruriens* Seeds (Velvet Beans). *Pakistan Journal of Nutrition* 10 (10), 947- 951.
- [24] Oloyed, O.I., 2005. Chemical profile of unripe pulp of *Carica papaya*. *Pakistan Journal of Nutrition*. 4, 379-381.
- [25] Onwuka, G.I. 2005. Food Analysis and Instrumentation theory and Paractice, 1st Edn, Naphthali prints, Lagos, pp, 114-169.
- [26] Osuntokun, B.O., 1972. Cassava diet and cyanide metabolism in Wistar rats. *British Journal of Nutrition*. 24, 797-805.
- [27] Palozza, P., 1998. Pro-oxidant actions of Carotenoids in Biologic Systems. *Nutrition Revolution*. 56, 257-256.
- [28] Preet, K., Punia, D., 2005. Proximate composition, phytic acid, polyphenols and digestibility (*in vitro*) of four brown cowpea varieties. *International Journal of Food Science and Nutrition*. 51, 189-193.
- [29] Price, K.R. & Fenwick, G.R., 1985. Naturally-occurring oestrogens in foods – A Review. *Food Additives and Contaminants*. 2, 73-106.

- 
- [30] Roeder, E. 1995. Pharmazie. *African Journal of Food Science*. 50, 83-98
- [31] Saito, K., Horie, M., Hoshino, Y., Nose, N., Nakazawa, J., 1990. High performance liquid chromatographic determination of glycoalkaloids in potato products. *Journal of Chromatography*. 508, 141-147.
- [32] Schneider, G., Woliling, J., 2004. Synthetic Cardenolides and Related compounds in: *Current Organic Chemistry*, 8. Macmillan, London.
- [33] Shamsuddin, A. M., Vucenic, I., Cole, K. E. 1997. IP6, A novel anticancer agent. *Life Science*, 61, 343-354.
- [34] Siddhuraju, P., Becker, K., Makkar, H.P.S., 2005. Studies on the nutritional composition and antinutritional factors of three different seed material of an under-utilised tropical legume, *Mucuna pruriens* var. *utilis*. *Journal of Agriculture and Food Chemistry*. 48, 6048-6060.
- [35] Singh, U., 1988. Antinutritional factors of chickpea and their removal by processing. *Plant Foods for Human Nutrition*. 38, 251-261
- [36] Trease, G.E., Evans, W.C., 1989. *Pharmacognosy*. 2nd Ed., Braille Tiridel and Macmillan publishers
- [37] Wheeler, E.L., Ferell, R.E. 1971. A method for Phytic acid determination in Wheat Fraction. *Cereals Chemistry* 48,312-320.