Processing Effect of Nutritional and Anti-Nutritional Content of African Locust Bean Seeds (*Parkia biglobosa* Benth)

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African Locust Bean (*Parkia biglobosa*) is among the leguminous plants used by man particularly in some African countries for the production of local condiment. African locust bean seeds are rich in protein and usually fermented to a tasty food condiment called *daddawa* which is used as a flavour intensifier for soups and stews and also adds protein to a protein-poor diet. However, the use of African locust bean seeds and other legumes as protein source is limited by the presence of anti-nutritional factors which are a diverse range of naturally occurring compounds in many tropical plants. This study sought to investigate the nutrients and anti-nutrients content of locust bean. The result of nutrient factors were analyses and it can be seen that the carbohydrate had highest percentage occurrence with 70.72% followed by Moisture with 11.5% and lipid with 10% and protein with 6.78% then ash with 4%. The results of anti-nutritional factor on locust bean can be seen that the Nitrate had a highest percentage occurrence with 82.8% followed by Tannins with 33% and Cyanide with 4.1% and Phytate with 2.94% and Oxalate with 0.0027%. Therefore this present work show that locust bean contain high nutritional value and less anti-nutritional value.
INTRODUCTION

African locust bean seeds are rich in protein and usually fermented to a tasty food condiment called *dawadawa* which is used as a flavour intensifier for soups and stews and also adds protein to a protein-poor diet (Ikenebomeh and Kok, 1984; Odunfa, 1986; Dike and Odunfa, 2003). However, the use of African locust bean seeds and other legumes as protein source is limited by the presence of anti-nutritional factors which are a diverse range of naturally occurring compounds in many tropical plants. The anti-nutritional factors cause poor protein digestibility in man and animals and are capable of precipitating other deleterious effects. Manifestations of toxicity from the consumption of legumes containing anti-nutritional factors range from severe reduction in food intake and nutrient availability or utilization, to profound neurological effects and even death (Osagie, 1998). To improve the nutritional quality and organoleptic acceptability of leguminous seeds, processing techniques have been employed to reduce or destroy the anti nutrients present in them. Some of the commonly used processing techniques include soaking in water, boiling at high temperatures in water, alkaline or acidic solutions, sprouting, autoclaving, roasting, dehulling, microwave treatment, steam blanching and fermentation. Little information is available on the effect of processing on the anti nutritional factors of African locust bean. This study was therefore undertaken to investigate the effects of processing on some nutrients and anti-nutritional factors in African locust bean seeds and the extent of reduction of these following processing.

METHODOLOGY

Sample collection

Locust bean (dawdawa) were obtained from sellers at central market (Kofargawo) Sokoto South L/G Sokoto State were then stored in polything bag and brought to MCB lab Sokoto State University and use for analysis.

Determination of nutrients in fermented locust bean

Moisture content

An appropriate amount of fermented locust bean (ml) equivalent to 2g of dried residue was measured into an empty disc which has been cleaned and dried in an oven at 80°C for about 30 minutes and weight (W₁), the crucible plus fermented locust bean sample was then weight (W₂). The sample in the crucible was dried in an oven at temperature of 105°C for 24 hours. The crucible was removed from the oven, cool in a desiccator for 20 minutes and it was weighed (W₃). The procedure was repeated, drying for about 3 hours for each subsequent drying until a constant value is obtain. The differences in weight after heating gives the moisture content and is determine by subtracting the dry weight from the wet-weight (AOAC, 1995).

\[
\% \text{ Moisture} = \frac{\text{Loss in weight due to drying} \times 100}{\text{Weight of fresh sample}}
\]

\[
\% \text{ Moisture} = \frac{W₁ - W₃ \times 100}{W₂ - W₁}
\]

Ash content

A clean crucible was ignited in a hot furnace for one minute. The crucible was then removed and cools in a dessicator and weighed (W₁). Two gram of the fermented locust bean residue was place into the crucible and then weighed (W₂) the crucible containing the sample was place in a muffle furnace and heat at 600°C for five hours. After ashing, the crucible would be removed, cool in a dessicator and weighed (W₃) (AOAC, 1995).

\[
\% \text{ Ash} = \frac{\text{weight of Ash} \times 100}{\text{Weight of sample}}
\]

\[
\% \text{ Ash} = \frac{W₃ - W₁ \times 100}{W₂ - W₁}
\]

Note that: W₁ = weight of empty crucible  
W₂ = weight of sample and crucible  
W₃ = weight after ashing

Lipid content

The free thimble was weighed (W₁) Two grams (2g) of the dried sample was weighed and transferred into a thimble which has been dried and weighed. The thimble containing the powdered sample was weighed (W₂) and the mouth porous thimble was covered with fat free absorbent cotton wool in order to distribute the draping petroleum ether. The thimble was then placed in a soxhlet extractor fitted to a round bottom flask containing 150cm of petroleum ether. The apparatus was switched on for 5 to 6 hours at 50°C. After this, the thimble was removed from the soxhlet and weighed (W₃) the flask was removed with care and the weighed to know the content of crude lipid (AOAC, 1995).

\[
\% \text{ Crude lipid} = \frac{\text{weight loss by thimble} \times 100}{\text{Weight of sample}}
\]

\[
\% \text{ Crude lipid} = \frac{W₃ - W₁ \times 100}{W₂}
\]

Crude protein

Three steps are involved in this analysis. They are digestion, distillation and filtration.
a. Digestion

Two gram of sample was weighed and transferred into a 500ml kjeldahl digestion flask, 20ml of concentrated H\textsubscript{2}SO\textsubscript{4} was added and mix gentle by swirling under tap water. 10g of anhydrous Na\textsubscript{2}SO\textsubscript{4} and 1g of CuSO\textsubscript{4} was mixed together and 3g of this catalyst was introduced into the flask. Anti-burning chips was added into the mixture. The content was boiled gently in a fume cupboard until charred particles disappear and a clear green solution obtained, the digested mixture was make-up to 100ml with distilled water (AOAC, 1995).

b. Distillation

Forty millimeters (40ml) of 2% boric acid was measured into 250ml beaker and 2 drops of indicator was added. Appropriately 10ml of digestive sample was pour into the distillation flask and the apparatus was set up. The heating system was switch on for 25 minutes; the receiver beaker was then removed (AOAC, 1995).

c. Titration

The collected distillate was cooled and titrated against 0.1 N HCl acid to an end point (indicates by change in colour from gray to purple) (AOAC, 1995).

\[
\% \text{Nitrogen} = \frac{\text{Titer values} \times \text{Nitrogen} \times 6.25}{\text{Weighed of sample (w)} \times \text{mls of Aliquot}}
\]

Where

\[
\text{N} = \text{Normality} = 0.1
\]

\[
\text{Tv} = \text{Titer values}
\]

\[
\% \text{Crude protein} = \text{Nitrogen} \times \text{conversion factor (6.25)}
\]

Carbohydrates content

\[
\% \text{Carbohydrates} = 100 - (\% \text{Moisture} + \% \text{Ash} + \% \text{Lipid} + \% \text{protein}) \quad (\text{Hunt et al., 1987})
\]

Determination of antinutrients in fermented locust bean

Nitrate

Nitrate was determined using method of (Joslyn, 1970). 0.1g of powder sample was added into 100ml conical flask, 10ml of distilled water was added and boil for 30 minutes, filter using filter paper. Mix and to make the volume of 50ml of distilled water DH\textsubscript{2}O, and incubate for 20 minutes at room Temperature and measure the absorbance at 760nm.

\[
\text{Absorbance of sample} \times \text{concentration of standard}
\]

Cyanide

Cyanide was determined by the method reported by (Bohm and Kocipai, 1994). 0.5g of powder sample was measure into 100ml of conical flask and 50ml of distilled water DH\textsubscript{2}O was added and boil for 30 minutes and filter using filter paper. Mix and boil at 90\textdegree for 5 minutes. Cool and measure the absorbance at 490nm.

\[
\text{Absorbance of sample} \times \text{concentration of standard}
\]

Oxalate

Oxalate was determined by the method reported by (Jrand and Underwood, 1986). One gram (1g) of the sample was added to 75ml of 15% H\textsubscript{2}SO\textsubscript{4} the solution was carefully stirred intermittently with magnetic stirred for 1 hours and filtered using filter paper, the filtrate (25ml) was then collected and titrated against 0.1N M\textsubscript{2}O\textsubscript{4} solution till a faint pink colour appeared that persisted for 30 seconds. 1cm\textsuperscript{3} of 0.1N KM\textsubscript{2}O\textsubscript{4} = 0.0045g of oxalic acid.

\[
\% \text{o}xalate \text{g} \% = \text{Titer value} \times 0.0045.
\]

Phytate

The phytate of each sample was determined through phytic acid determination using the procedure described by (Ajayi, 2011). 4g of the sample was soaked in 100ml of 2% HCl for 3 hours, and filtered. 25ml of the filtrate, 5ml of 0.3\%NH\textsubscript{4}SCN, and 53ml of distilled water DH\textsubscript{2}O were mixed together and titrate against 0.01N standard ferric chloride fecl\textsubscript{3} solution containing 0.00195g/ml until a brownish yellow colour persisted for 5 minutes.

\[
\% \text{Phy}tate \text{mg} \% = \text{Titrat value} \times 1.19 = \text{Phytin Phosphorus}
\]

Phytate content = Phytin Phosphorus \times 3.55

RESULTS

The result of various nutrients determination carried out on locust bean which include: moisture content, protein content, lipid, ash content and carbohydrates. And it can be seen in (table 1 and figure 1) that the carbohydrate had highest percentage occurrence with 70.72% followed by Moisture with 11.5% and lipid with 10% and protein with 6.78% then ash with 4%. 

\[
\text{Titrat value} \times 1.19 = \text{Phytin Phosphorus}
\]

Phytate content = Phytin Phosphorus \times 3.55
The results of anti-nutritional factor on fermented locust bean were analyses. From the result it can be seen that the Nitrate had a highest percentage occurrence with 82.8% followed by Tannins with 33% and Cyanide with 4.1% and Phytate with 2.94% and Oxalate with 0.0027% are presented in table 2 and figure 2.

| Sample | Oxalate(%) | Phylate(%)Mg | Cyanide(%)Mg | Tannins(%)Mg | Nitrate(%)Mg |
|--------|------------|--------------|--------------|--------------|--------------|
| A1     | 0.0022     | 2.94         | 4            | 33           | 77.6         |
| A2     | 0.0027     | 0.31         | 4.1          | 32.7         | 80.5         |
| A3     | 0.0022     | 0.31         | 4            | 32.9         | 82.8         |
| Mean   | 0.0035     | 1.1          | 6.0          | 32.8         | 80.3         |
DISCUSSION

The carbohydrate content gave an indication that the bean studied here can be considered as a rich source of energy and was able for supplying the daily energy requirements of the body in children and adult. Most of the condiments have high moisture content, particularly the soya beans and locust bean daddawa, which can encourage microbial growth and enhance spoilage by organisms such as bacteria and fungi, if not properly stored; the high moisture content may make the bean highly susceptible to microbial attack. Lipids are found to have 10% which are probably the most important source of derived flavors and also lipids are important precursors of volatile flavors. Protein are found to have 6.78% protein value for fermented locust bean is high when compared with those protein – rich foods such as groundnut, pigeon peas, bambara groundnut and some oil seeds which contain less protein value. Fermented locust bean and could therefore be used as an alternative source of protein in the diet/protein supplement especially in nation like Nigeria where the majority of the populace lives on starchy food and cereals. Ash content are found to have 4% this is an indication that locust bean is potential good source of minerals require by the body.

The anti-nutrients factors determine were presented in table 4, which include Oxalate, Tannins, Phytate, Cyanide, and Nitrate. It can be seen that Nitrate had higher percentage with 82.8%, Cyanide 4.1%, Tannins 33%, Phytate 2.94%, Oxalate 0.0027%. Nitrates are found to have 2.8%, but various studies suggest that nitrate is harmless and rather beneficial. It has been postulated as a useful nutrient (Dykhuizenet al., 1996). Bjorne et al. (2004) in this study, our result revealed the presence of low antinutrients value in cyanide, phytate, tannins and oxalate. Although antinutrient was removed after defatting process with butanol. The values detected are at a safe level that poses no danger in diets. It is believed that fermentation reduces antinutritional factors; therefore, the processing method may account for the result obtained. This is in agreement with what was reported previously for African locust beans (Ijarotimi and Keshinro 2012). Antinutrients are natural compounds that interfere with the bioavailability of nutrients by interfering with their absorption in the gastrointestinal tract. Phytate and oxalate are examples of antinutrients that interfere with some mineral components such as calcium, iron, zinc, and magnesium by forming insoluble complexes (Ijarotimi and Keshinro 2012). Oxalates may be present in plants as soluble salts such as potassium, sodium or ammonium oxalate. Phytate content phytic acid, a hexaphosphate or inositol is an important storage form of phosphorus in plants. It is insoluble and cannot be absorbed in the human intestines. Phytic acid has 12 replaceable hydrogen atoms with which it can form insoluble salts with metals such as calcium, iron, zinc, and magnesium. The formation of these insoluble salts renders the metals unavailable for absorption into the body. (Aregheore and Agunbiade, 1991; Akpabioet al,2012) reported that, cooking does not destroy anti-nutritional factors which are toxic to health and make dietary minerals available for absorption. Tannins form insoluble enzyme resistance complex with proteins thereby reducing their digestibility and protein quality. Tannin causes decreased feed consumption in animals, binds dietary protein and digestive enzymes to form complexes that are not readily digestible (Aletor, 1993). They also cause decreased palatability and reduced growth rate (Roeder,
1995). These reduction in antinutrients may be attributed to the processing techniques involved in the fermentation of the raw African locust bean seed which include dehulling, soaking, boiling which helps to leach away antinutrients. Enjuigga and Ayodele-Oni, (2003) this confirms an earlier suggestion that the traditional methods employed in processing the seeds namely soaking, hydrothermal treatment, and fermentation would considerably reduce the levels of antinutritional factor.

CONCLUSION

Fermented locust bean and could therefore be used as an alternative source of protein in the diet/protein supplement especially in nation like Nigeria where the majority of the populace lives on starchy food and cereals. Ash content are found to have 4% this is an indication that locust bean is potential good source of minerals require by the body. While reduction in antinutrients may be attributed to the processing techniques involved in the fermentation of the raw African locust bean seed which include dehulling, soaking, boiling which helps to leach away antinutrients. This confirms an earlier suggestion that the traditional methods employed in processing the seeds namely soaking, hydrothermal treatment, and fermentation would considerably reduce the levels of antinutritional factor

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