International Journal of Life Sciences

(A peer reviewed International Journal)

The Anti nutritional properties of African Yam Bean *(Sphenostylis sternocarpa)* as affected by chemical treatment

International Journal of Life Sciences, Vol. 1 No. 3. pp. 74-81 2277-193x. 2012

ISSN *2277 – 193x*

Article type *Full Length Research Article*

Submission date *28 June 2012*

Acceptance date *10 July 2012*

Publication date *15 July 2012*

Article URL *http://www.crdeep.com/category/ijls*

Authors ***NWOSU, J.N, Owuamanam, C.I, Onuegbu, N, Ogueke, C.C., and Ojukwu, M.***

CRDEEP Head Office: 315/10, Kaulagarh Road, Rajendranagar, Indervihar, Dehradun, 248001, Uttrakhand, India.

International Journal of Life Sciences Vol.1 No.3. 2012. Pp. 74-81

Full Length Research Paper

The Anti nutritional properties of African Yam Bean *(Sphenostylis sternocarpa*) as affected by chemical treatment

***NWOSU, J.N, Owuamanam, C.I, Onuegbu, N, Ogueke, C.C., and Ojukwu, M.**

Department of Food Science and Technology, Federal University of Technology P.M.B 1526, Owerri, Imo State, Nigeria

****Corresponding Author***

Abstract

The effect of Alum and Trona on the antinutritional properties of African Yam Bean seeds was studied. African yam seeds were separately soaked in solutions of alum and trona with varying concentrations (0.25%, 0.50% and 1.0%) and allowed to stand for 24h and 48h before being processed into flour. The phytochemical analysis of the alum and trona treated AYB and the untreated (control) showed significant variations in their antinutritional properties. Relative to the raw (untreated) AYB, the 24h soaked seeds showed reduction in the alkaloid content (0.37% - 0.34%), though not significantly different. Tannin content (0.41% - 0.19%) and Saponin (0.57% - 0.34%). Similarly, oxalate and HCN reduced significantly. Flavonoids and phenols were not affected by the alum treatment .Results also showed that soaking in alum for 48h had higher reduction effect on Saponin (0.57%-0.31%) and Oxalate(0.36%-0.23%) Soaking for 48h in trona was found to have higher reduction effect on all the antinutrients except saponin which rather increased from (0.57-0.73) and oxalate. In addition ,both flavonoids and phenols were reduced by trona treatment. Comparatively, antinutrient reductions were higher in trona than in alum solutions while the 48hours soaked samples had higher effect than 24h soaked sample in both alum and trona solutions,except for Saponins.

Keywords: *Anitnutritional properties, steeping, Alum, Trona*

Introduction

African Yam Bean (*Sphenostylis stenocarp*), also known as wild yam bean is an annual crop that belongs to the leguminous family and sub-family of *papilonacea sp.* (Ihekoronye and Ngoddy, 1985). In Igbo speaking parts of Nigeria, it is commonly known as "Agwa". Also, in some parts of Igboland for instance in Abia State, it is known as "Odudu", Urhobo in Delta state calls it "Ekpakpani" while Edo calls it 'illoloegwa', etc. African yam bean is a climbing plant which twines and climbs to a height of about 3M and requires staking, with each pod containing several edible seeds (Wikipedia, 2005). African yam bean is grown mostly in the eastern part of Nigeria. e.g. Ebonyi, Abia and Enugu States, etc.

The nutritional composition of African yam bean is similar to that of most edible legumes. It contains about 21.10% of protein, 5.70% of Crude fibre, 74.10% of carbohydrate, 3.20% of Ash, 8.5% of moisture and 8.25% of fat (Nnayelugo, *et al:* 1995). They are good sources of vitamins A and C, folate, magnesium, manganese, riboflavin, phosphorus and potassium (Wikipedia, 2005). Thus the major nutrient is protein and carbohydrate, of which its variability in protein is influenced by genotype as well as environmental factors. (Bliss, 1995).

African Yam bean is consumed in different forms, mostly in the eastern part of Nigeria and the whole country at large. Various types of products are traditionally produced from it through soaking, dehulling, grinding, boiling, steaming and frying or by combination of any two or more of these methods.

The tender crisp pods are eaten both fresh and when cut into short sectors ,used for cooking purposes. Wikipedia, (2005), found out that they are best if picked for vegetable use before they reach full maturity, though the mature seeds can be utilized as cooked beans or converted into paste or flour for subsequent use in 'Akara' (by frying) or moi-moi (by steaming) (Uzuegbu and Eke, 2000). Many authors have stressed the important roles beans play in the diets of many populations in countries where protein is deficient. This has promoted research on various species and aspects of the bean utilization. William, (1984) studied the various qualities that determine consumer preference and identified the following in descending order of priority: ability to swell when cooked, good bonding properties and desirable sensory properties such as flavor and texture, as the major factors. As a result of economic recession, the majority of Nigerians now derive protein mainly from bean species, because the country is faced with acute shortage of animal protein, which is often beyond the reach of an average Nigerian (Henshaw and Sanni, 1995). The choice of bean by Nigerian women is guided predominantly by the cooking time, swelling capacity, taste and colour of the bean (Hussein, *et al*, 1984).

The matured seed of African yam bean has been found to have limitations such as long cooking time, reduced swelling ability, and production of black liquid during cooking. Also dehulling of the dry and soaked seeds is of great difficulty to the traditional man. This has seriously affected its use for such

food products that require dehulling e.g moi-moi and akara. Other problems associated with African yam bean are flatulence and beany off flavor. These have affected the consumption rate and acceptability of African yam bean for subsequent processes.

The objectives of this research work therefore is

(i) to investigate the effect of soaking African Yam bean in different chemical (trona/Alum) on the antinutritional properties of the bean.

(ii) to know the effect of soaking with these chemicals on the proximate composition of these seeds and to select the best treatment method that can be used to reduce these antinutrients. It is hoped that this will help to expose the functionality of African Yam bean and its increased utilization in food formulations and processing.

Materials and Methods

Source of Materials:

The dry seeds of African yam bean (AYB) (*Sphenostylis, sternocarpa*) were purchased from a local market in Aba, Abia State of Nigeria.

Chemicals

The Chemicals and equipment used were of analytical grade from the Department of Food Science and Technology and Department of Crop Science and Technology of the Federal University of Technology Owerri, Imo State Nigeria.

Sample Preparation

The seeds were first examined and sorted to remove extraneous materials such as dirt, dust, etc as well as shriveled and decreased (pest infected) ones. The wholesome ones were used for the work.

The wholesome seeds were weighed out in portions, each of which was 500g (i.e. 0.5kg). A total of twenty (20) portions were weighed out. The weighed portions were further grouped in three sub-groups of two (2). The groups were labeled, A, B, C which was for control (ie Raw samples) ;Alum treated and Trona treated respectively.

The Alum treated group was labeled in accordance with the treatments which were $A0.25_a$, $A0.50_a$, $A0.75_a$, $A1.0_a$, for 24hours soaking and $A0.25_b$, $A0.50_b$, $A0.75_b$, $A1.0_b$, for 48hours soaking. The samples in each labeled portion were eventually soaked separately in Alum solution containing 0.25%, 0.50%, 0.75% and 1.0% respectively. The ones with subscripts (a) were allowed to soak for 24hours only while those with subscript (b) were soaked for 48hours.

Exactly the same treatments that were given to the Alum treated samples were also given to the Trona treated samples. However, they were soaked in Trona solutions in place of Alum solution. Hence they were labeled from T0.25 up to $T1.0_a$ and $T0.25_b$ to $T1.0_b$.

The treated samples (soaked in various concentrations of Alum and Trona) were allowed to stand at room temperature for 24h and 48h according to the treatments. At the end of each soaking period, the soaked seeds were brought out, washed in running water (tap) and rinsed in distilled water (this process removes residual treatment salts from the surface of the seeds).

The seeds were then dried in the oven (Carbolite, England) at 60^0C for 8hours and then milled to powder with the aid of an electric laboratory mill (Arther Thomas, USA).After milling, the ground seeds were sieved through 1mm test sieve to obtain processed powered sample which was used for the analysis. The milling machine was cleaned well after milling each sample before grinding the next sample. All the ground samples were put in well labeled screw capped glass ample bottles. Analysis was done on the samples within a period of 96hours (4days)

Determination of the Anti-Nutritional factors

Determination of Tannin

The tannin content of the sample was determined by Folin Denms Colometric method (Harborne, 1993). A measured weight of the processed sample (5.0g) was mixed with distilled water in the ratio of 1:10 (w/v). The mixture was shaken for 30min at room temperature and filtered to obtain the extract.

A standard tannin acid solution was prepared, 2ml of the standard solution and equal volume of distilled water were dispersed into a separate 50ml volumetric flask to serve as standard and reagent blank respectively. Then 2mls of each of the sample extracts was put in their respective flasks and labeled.

The content of each flask was mixed with 35ml distilled water and 1ml of the Folin Denms reagent was added to each. This was followed by 2.5mls of saturated $N_{a2}CO_3$ solution. Then each flask was diluted to the 50ml mark with distilled water and incubated for 90min at room temperature. Their absorbance was measured at 760nm in a colorimeter with the reagent blank at zero. The tannin content was calculated as shown below

% tannin = 100/w x au/as x c/1000 x vt/va

w = weight of sample

au = absorbance of test sample

as = absorbance of standard tannin solution

c = concentration of standard tannin solution

vt = total volume of extract

va = volume of extract analyzed

Determination of Saponin

This was done by the double solvent extraction gravimetric method (Harborne, 1993). 5.0g of the processed sample was mixed with 50mls of 20% aqueous ethanol solution and incubated for 12h at a temperature of 55^0C with constant agitation.

After that, the mixture was filtered through what man No 42 grades of filter paper. The residue was re-extracted with 50ml of the ethanol solution for 30min and the extracts weighed together.

The combined extract was reduced to about 40mls by evaporation and then transferred to a separating funnel and equal volume (40mls) of diethyl ether was added to it. After mixing well, there was partition and the outer layer was discarded while the aqueous layer was reserved. This aqueous layer was re-extracted with the ether after which its pH was reduced to 4.5 with drop wise addition of dilute NaOH solution.

Saponin in the extract was taken up in successive extraction with 60ml and 30ml portion of named butanol. The combined extract (ppt) was washed with 5% NaCl solution and evaporated to dryness in a previously weighed evaporation dish. The saponin was then dried in the oven (at 60^0C removes any residual solvent); cooled in a desiccators and re-weighed. The saponin content was calculated as shown below;

$$\% \text{ saponin} = \frac{W_2 - W_1}{W}$$

W = Weight of sample used
W_1 = Weight of empty evaporation dish
W_2 = Weight of dish + Saponin extract

Determination of Alkaloids

The alkaline precipitation gravimetric method (Harbone, 1993) was used.

A measured weight of the processed sample (5g) was dispersed in 100mls of 10% acetic acid in ethanol solution. The mixture was well shaken and allowed to stand for 4 hours at room temperature being shaken every 30min. At the end of this period, the mixture was filtered through what man No 42 grade of filter paper.

The filtrate (extract) was concentrated by evaporation; to a quarter of its original volume. The extract was treated with drop-wise addition of concentrated NH_3 solution to precipitate the alkaloid. The dilution was done until the NH_3 was in excess.

The alkaloid precipitate was removed by filtration using weighed what- man No 42 filter paper. After washing with 1% NH_4OH solution, the precipitate in the filter paper was dried at 60^0C and re-weighed after cooling in a desiccators. The Alkaloid content was calculated as shown below:

$$\% \text{ alkaloid} = \frac{W_2 - W_1}{\text{Wt of sample}} \times \frac{100}{1}$$

Where W_1 = Weight of empty filter paper
W_2 = Weight of filter + Alkaloid ppt

Determination of Phenols

This was determined by the Folin – Ciosptean spectrophotometer (AOAC 1990). The total phenol was extracted in 200mg of the sample with 10ml concentrated methanol. The mixture was shaken for 30min at room temperature.

The mixture was centrifuged at 500rpm for 15min and the supernatant (extract) was used for the analysis.

1ml portion of the extract from each sample was treated with equal volume of Folin-Ciosptean reagent followed by the addition of 2mls of 2% Na_2CO_3 solution. Meanwhile, standard phenol solution was prepared and diluted to a desired concentration.

1ml of the standard solution was also treated with the F-D reagent .Blue colouration was measured (absorbance) in a colour meter at 560nm wavelength. Measurement was with a reagent blank at zero.

The phenol content was calculated using the formula below:

$$\%\text{phenol} = \frac{100 \times au}{W} \times \frac{c}{as} \times \frac{Vt}{100\ Va}$$

Where W = Weight of sample
au = absorbance of test sample
as = absorbance of standard phenol sample
c=concentration of standard phenol sample
vt = total extract volume
va = volume of extract analyzed

Determination of Flavonoids

Flavonoid was determined using the method described by Harborne (1993).

A measured weight of the processed sample (5g) was boiled in 100mls of 2MHCL solution under reflux for 40min. It was allowed to cool before being filtered. The filtrate was treated with equal volume of ethyl acetate and the mixture was transferred to a separation funnel. The flavonoid extract (contained in the ethyl acetate portion) was received by filtration using weighed filter paper. The weight was obtained after drying in the oven and cooling in desiccators. The weight was expressed as a percentage of the weight analyzed. It was calculated as shown below:

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

Where W_1 =Weight of filter paper x Flavonoid precipitate
W_2 = Weight of filter paper alone

Determination of Hydrogen Cyanide (HCN)

This was determined by alkaline pikrate colorimetric method by Balagopalan *et al,* (1998) .2g of the sample was dispersed in 50ml of distilled water in a 25ml conical flask. An alkaline pikrate paper was hung over the sample mixture and the blank in their respective flasks.

The set up were incubated overnight and each pikrate paper was eluted or dipped into a 60ml of distilled water. A standard cyanide solution was prepared and diluted to a required concentration. The absorbance of the eluted sample solutions were measured with colorimeter at 540nm wavelength with the reagent blank at zero.

The cyanide content was determined by the formular shown below.

$$\text{HCN mg/kg} = \frac{100 \times au}{W} \times \frac{C}{as} \times D$$

Where W =weight of sample analyzed
au = absorbance of test sample
as = absorbance of standard HCN solution
C = concentration of the standard in mg/d
D = dilution factor where applicable

Determination of Phytate

Phytate in the sample was determined using the Biphyrimidine colorimeter method described by Onwuka (2005). A weighed sample (2g) was soaked in 50ml of 0.2N HCl solution and shaker for 30min in a machine shaker. It was filtered to obtain the extract. A portion of the extract (0.5mls) was dispensed into a test tube and 1ml of acidified ferrous ammonia sulphite solution was added to it. The tube was stoppered and boiled in water bath for 30min. It was then cooled in ice water for 15min and allowed to reach room temperature. The mixture was centrifuged at 3000 rpm for 5min and the supernatant was collected for analysis. 1ml of the supernatant was mixed with 1.5ml of 2.2 Biphrimidine solutions. Meanwhile a standard solution of phytate was prepared and diluted to a chosen concentration. 1ml of the standard solution was treated the same way as the sample extract as described above. The absorbance of the standard and the sample were read in a spectrophotometer at a wavelength of 519nm.

A reagent blank was used to set the instrument at zero. The formular below was used to calculate the phytate content.

$$\%\text{phytate} = \frac{100}{W} \times \frac{au}{as} \times \frac{c}{100} \times \frac{Vt}{Va}$$

Where au = Absorbance of sample

as = Absorbance of standard solution

c = Concentration of the standard

Vt = Total volume of extract

Va = Volume of extract analyzed.

Determination of Oxalate

5g of the sample was weighed into a 100ml beaker, 20ml of 0.30N HCl was added and warmed from 40 – 50^{0}C, using magnetic hot plate and stirred for one hour. It was extracted three times with 20ml of 0.30N HCl and filtered into a 100ml volumetric flask. The combined extract was diluted to 100ml mark of the volumetric flask.

The oxalate was estimated by pipetting 5ml of the extract into a conical flask and made alkaline with 1.0ml of 5N ammonium hydroxide. A little indicator paper was placed in the conical flask to enable us know the alkaline regions. It was also made acid to Phenolphalein (2 or 3 drops of this indicator added, excess acid decolourizes solution) by dropwise addition of glacial acetic acid. 1.0ml of 5% C_aCI_2 was then added and the mixture allowed to stand for 3h after which it was then centrifuged at 3000rpm for 15min. The supernatants were discarded and the precipitates washed 3 times with hot water with thorough mixing and centrifuging each time. Two milliliters of 3N H_2SO4 was added to each tube and the precipitates dissolved by warming in a water bath (70 – 80^{0}C). The content of all the tubes was carefully poured into a clean conical flask and titrated with freshly prepared 0.05M $KMnO_4$ at room temperature until the first pink colour appeared till the solution became colourless. The solution was then warmed to 70 – 80^{0}C and titrated until a permanent pink colour that persisted for at least 30 seconds was attained.

Determination of Trypsin Inhibitor

This was done using the spectrophotometric method, described by Amtfield *et al*; (1985).

A measured weight (2g) of the test sample was dispersed in 50ml of 0.5M NaCl solution and stirred for 30min at room temperature. It was centrifuged and the supernatant filtered through Whatman No 42 filter paper. The filtrate was used for the assay.

Standard trypsin was prepared and used to treat the substrate solution (N-benzoyl – D1 – arginine – p – anilide; BAPA). The extent of inhibition was used as a standard for measuring the trypsin inhibitory activity of the test sample extract. Into a test tube containing 2ml of extract and 10ml of the substrate (BAPA) 3ml of the standard trypsin solution was added. Also 2ml of the standard trypsin solution was added in another test tube containing only 10ml of the substrate. The latter served as the blank/control.

The content of the tubes were allowed to stand for 30min and then the absorbances of the solution measured spectrophotometrically at 410nm wavelength.

One trypsin activity unit inhibited is given by an increase of 0.01 absorbance unit at 410nm.

Trypsin unit inhibited

$$= \frac{Au}{As} \times 0.01 \times F \times \frac{Vt}{Va} \times \frac{1}{w}$$

Where Au = Absorbance of test sample

As = Absorbance of standard (uninhibited) sample

F = Experimental factors given as

Vt = Total volume of extract

Va = Volume of extract analyzed

W = Weight of sample analyzed.

Results and Discussion

Effects of chemical (alum) treatment, soaking time and concentration on the antinutritional properties of african yam bean.

Table 1 shows the antinutritional factors of African yam bean (AYB) soaked with different concentrations of alum. For 24h alkaloid showed no significant difference ($P \geq 0.05$) with different concentrations of alum. Hydrogen cyanide (HCN) was highest in the untreated AYB (9.89mg/kg) and lowest in that of 1.00% (7.51mg/kg). There was a significant difference ($P \leq 0.05$) in the HCN contents of the AYB samples soaked with different concentrations of alum for 24h. Tannin of untreated AYB was highest (0.41%) while AYB soaked with 1% concentration of alum was lowest (0.194%). There was no significant difference in the tannin contents of AYB (0.5, 0.75 and 1.0% concentrations of alum. Saponin was highest in the untreated AYB (0.5%) and lowest in AYB soaked with 1.0% concentration of alum (0.33%).There was a significant difference ($P \leq 0.5$) in the saponin contents of the AYB samples. Flavanoid showed no significant difference ($P \geq 0.5$) in the untreated AYB and AYB soaked with different concentrations of alum. Flavanoids ranged from 0.40 – 0.41%. Total titratable acidity (TTA) was highest in the untreated AYB (2.5%) and lowest in that of 1.0% (1.3%). However, there was no significant difference ($P \geq 0.05$) in TTA of the treated and untreated AYB samples. Phytate was highest in the untreated AYB and AYB soaked with 0.25% concentration of alum (0.32%) and lowest in 1.0% concentration of alum (0.30%). There was no significant difference ($P \geq 0.05$) in the phytate content of the untreated AYB and that soaked with diffused concentration of alum . Oxalate was highest in the

untreated AYB (0,36%) and lowest in AYB soaked with 0.75% and 1.0% concentrations of alum with a value of 0.25

There was no significant difference (P≥ 0.05) in the oxalate content of AYB soaked with 0.75% and 1.0% concentrations of alum. Phenol was highest in AYB soaked with 1.0% of concentration of alum and lowest in other concentrations and the untreated AYB. There was no significant difference (P≥0.05) in the phenols of the treated AYB and
the untreated AYB. From this result, it could be inferred that the soaking of AYB with alum helped to reduce their antinutritional properties, but for the concentration of phenols.

Table 1: Mean values of the antinutritional properties of African yam bean soaked for 24h and 48h with different concentrations of alum

TRT (24H)	ALK %	HCN mg/kg	TANNIN %	SAPONIN %	FLAV %	TTA %	PHYTATE %	OXALATE %	PHENOL %
Control	0.37^{a}	9.89^{a}	0.41^{a}	0.57^{a}	0.41^{a}	2.5^{a}	0.322^{a}	0.36^{a}	0.16^{a}
0.25	0.37^{a}	9.80^{a}	0.28^{b}	0.50^{b}	0.41^{a}	1.5^{a}	0.320^{a}	0.33^{b}	0.16^{b}
0.50	0.37^{a}	7.73^{c}	0.20^{c}	0.43^{c}	0.40^{a}	2.0^{a}	0.313^{a}	0.27^{c}	0.16^{b}
0.75	0.34^{a}	7.61^{d}	0.20^{c}	0.39^{d}	0.41^{a}	1.8^{a}	0.312^{a}	0.25^{d}	0.16^{b}
1.00	0.34^{a}	7.51^{e}	0.19^{c}	0.33^{e}	0.41^{a}	1.3^{a}	0.307^{a}	0.25^{d}	0.50^{a}
LSD (0.05%)	0.03	0.02	0.02	0.03	0.02	0.02	0.02	0.02	0.02
48H									
Control	0.37^{a}	9.89^{a}	0.41^{a}	0.57^{a}	0.41^{a}	2.5^{a}	0.322^{a}	0.36^{a}	0.16^{a}
0.50	0.35^{b}	7.07^{c}	0.187^{c}	0.41^{b}	0.41^{a}	2.0^{c}	0.315^{b}	0.25^{c}	0.16^{a}
0.75	0.33^{c}	7.01^{d}	0.183^{d}	0.39^{b}	0.41^{a}	1.8^{d}	0.312^{c}	0.24^{cd}	0.16^{a}
1.00	0.31^{d}	6.95^{e}	0.180^{e}	0.31^{b}	0.41^{a}	1.7^{c}	0.306^{d}	0.23^{d}	0.50^{a}
LSD (0.05%)	0.02	0.02	0.001	0.10	0.01	0.08	0.02	0.02	0.02

Mean values down the columns with the same letters are not significantly different at (p ≥ 0.05). Key: control = untreated AYB 0.25, 0.50, 0.75 and 1.0% = different concs alum, ALK = alkaloid, HCN = Hydrogen Cynide, TTA = total titratable acidity, LSD = least significant difference.

The effect of alum treatment on the antinutritional properties of ayb for 48h

The antinutritional properties of AYB soaked with different concentration of alum for 48h is also shown in Table 1. Alkaloid was highest in the untreated AYB (0.37%) and lowest in that of AYB soaked with 1.0% concentration of alum (0.31%). There was no significant difference (P ≥0.05) in the AYB samples soaked with 0.5% and 0.25% concentrations of alum. HCN was highest the untreated AYB (9.89mg/kg) and lowest in AYB soaked with 1.0% concentration of alum (6.95mg/kg). There was a significant difference (P≤ 0.05) in the HCN of the AYB samples.Tannin was highest in the untreated AYB (0.41%) and lowest in the AYB soaked with 1.0% concentration of alum (0.180%). There was a significant difference (P≤0.05) in tannin contents of the untreated and treated AYB samples. Saponin was highest in the untreated AYB (0.57%) and lowest in AYB soaked with 1.0% concentration of alum (0.31%). There was no significant difference (P≥0.05) in the saponin of the untreated AYB and AYB soaked with 0.25% concentration of alum. Also, there was no significant difference (P≥0.05) in the saponins of AYB soaked with 0.75%, 0.5% and 1.0% concentration of alum. There was no significant difference (P≥0.05) in the flavanoids of the untreated and treated AYB samples. TTA^{s} was highest in the untreated AYB (2.3%) and lowest in AYB soaked with 1.0% concentration of alum (1.70%). There was a significant difference (P≤0.05) in the total titratable acidity of the untreated and treated AYB samples. Phytate was highest in untreated AYB (0.322%) and lowest in AYB soaked with 1.0% concentration of alum (0.306%). There was no significant difference (P≥0.05) in the AYB soaked with 0.25% and 0.5% concentration of alum. Oxalate was highest in the untreated AYB (0.360%) and lowest in that of AYB soaked with 1.0% concentration of alum (0.23%). Phenol showed no significant difference (P≥0.05) in the untreated and treated AYB samples.

Effect of soaking with trona on the antinutritional properties of ayb soaked for 24h.

The antinutritional properties of African yam bean (AYB) soaked for 24h with different concentrations of trona is shown in table 2. The Alkaloid content was highest in untreated AYB (0.37%) and lowest in AYB soaked with 1.0% concentration of trona (0.31%). There was no significant difference (P≥0.05) in the alkaloids of AYB soaked with different concentration of trona but they differed with the untreated (control) sample. Hydrogen cyanide (HCN) was highest in the untreated AYB (9.89%) and lowest in AYB soaked with 1.0% concentration of trona (3.15%). There was a significant difference (P≤0.05) in the HCN contents of untreated AYB and different concentration of trona and between each concentration level.

Tannin was highest in the untreated AYB (0.197%) and lowest in AYB soaked with 1.0% concentration of trona (0.185%). There was no significant difference (P≥0.05) in the tannins of AYB soaked with 0.5% and 0.75 concentration of trona though there were significant difference between them. Saponin was highest in AYB soaked with 0.75% and 1.00

concentration of trona (0.63%) and lowest in the untreated AYB (0.56%). There was no significant difference (P≥0.05) in the saponin contents of AYB treated with different concentrations of trona. This result indicates that increase in the trona concentration increased the saponin content and therefore is not recommended for use in food formulations. Flavanoid was highest in untreated AYB (0.41%) and lowest in AYB soaked in 1% concentration of Trona (0.37%). There was no significant difference that teated with 0.25% Trona. There was no significant difference (P≥0.05) in the flavonoid contents of AYB soaked with 0.25%, 0.75% and 1% concentration of Trona.Total titratable auldity (TTA) was highest in untreated AYB (2.50%) and lowest in AYB treated with 1.0% concentration of trona 1.70. There was no significant difference (P≥0.05) in the TTA of AYB soaked with 0.5%, 0.75% and 1.0% concentrations of trona. Phytate was highest in the untreated AYB (0.322%) and lowest in AYB soaked with 1.0% concentration of trona (0.193%). There was a significant difference (P≤0.05) in the untreated AYB and AYB soaked with different concentrations of trona. Oxalate was highest in the untreated AYB 0.36% and lowest in AYB soaked with 1.0% concentration of trona (0.290%). Phenol was highest in the untreated AYB (0.16%) and lowest in AYB soaked with 1.0% concentration of trona (0.09%). There was no significant difference (P≥0.05) in phenol at 0.75% and 1.0% concentration of trona. Alkaloids of the soaked AYB reduced with increase in the concentrations of alum and trona for 24h and 48h. Hydrogen cyanide in the soaked AYB also showed reduction with increase in the concentrations of alum and trona for 24h and 48h.

Soaking of AYB with different concentration of alum and trona for 24h and 48h gave significant reduction. However, some trace element of tannins recorded in the study could be of health advantage. Recent studies have demonstrated that products containing chestnut tannins included at low dosages (0.15-0.2%) in the diet can be beneficial (Ologhobo *et al*, 1993). Some studies suggest that chestnut tannins have been shown to

Table 2: Mean values of the antinutritional properties of African yam bean soaked for 24h and 48h with different concentration of trona

TRT (24h)	ALK %	HCN mg/kg	TANNIN %	SAPONIN %	FLAV %	TTA %	PHYTATE %	OXALATE %	PHENOL %
Control	0.37^{a}	9.89^{a}	0.197^{a}	0.57^{b}	0.41^{a}	2.50^{a}	0.322^{a}	0.36^{a}	0.16^{a}
0.25	0.33^{b}	6.54^{b}	0.191^{b}	0.61^{a}	0.40^{ab}	2.20^{b}	0.307b	0.33^{b}	0.13^{b}
0.50	0.33^{b}	5.40^{c}	0.187^{c}	0.61^{a}	0.39^{bc}	1.80^{c}	0.281^{c}	0.32^{bc}	0.13^{b}
0.75	0.31^{b}	3.85^{d}	0.187c	0.63a	0.38^{cd}	1.80^{c}	0.253	0.31^{c}	0.10^{c}
1.0	0.31^{b}	3.15^{c}	0.185^{c}	0.63^{a}	0.37^{d}	1.70^{d}	0.193^{c}	0.29^{d}	0.09^{c}
LSD (0.05%)	0.02	0.02	0.002	0.03	0.02	0.14	0.003	0.02	0.02
48H									
Control	0.37^{a}	9.89^{a}	0.197^{a}	0.57^{b}	0.41^{a}	2.50^{a}	0.322^{a}	0.36^{a}	0.16^{a}
0.25	0.30^{b}	4.00^{b}	0.189^{b}	0.63^{b}	0.35^{b}	1.80^{b}	0.285^{b}	0.288^{b}	0.12^{b}
0.50	0.29^{bc}	3.06^{c}	0.186^{c}	0.65^{b}	0.33^{bc}	1.70^{b}	0.272^{c}	0.280^{b}	0.08^{c}
0.75	0.29^{bc}	3.03^{c}	0.182^{d}	0.71^{a}	0.331^{c}	1.20^{c}	0.222^{d}	0.26^{b}	0.07^{c}
1.0	0.27^{c}	0.51^{c}	0.179^{c}	0.73^{a}	0.31^{c}	1.20^{c}	0.187^{c}	0.27^{b}	0.07^{c}
LSD	0.02	0.81	0.003	0.05	0.03	0.26	0.002	0.02	0.01

Mean values down the columns with the same letters are not significantly different at (p≥0.05). Key: control = untreated AYB; 0.25, 0.50, 0.75 and 1.0% = different concs of alum, ALK = alkaloid, HCN = Hydrogen cynide, TTA = total titratable acidity, LSD = least significant difference.

have positive effects on silage quality in the rand bale silages, in particular reducing non protein nitrogen (NPNs) in the lowest wilting level (Tobacco *et al*, 2006). Also improved ferment ability of soya meal nitrogen in the rumen has also been reported by Mathew and Jouany (1993).

Effects of trona treatment on the anti-nutritional properties of ayb soaked for 48h.

The anti-nutritional properties of AYB soaked for 48h with diffuse concentration of trona is shown in Table 2. Alkaloid was highest in the untreated AYB (0.37%) and lowest in AYB soaked with 1.0% concentration of trona (0.27%). There was no significant different (P≥0.05) in the alkaloids of AYB soaked with 0.25% 0.50% and 0.75% concentration of trona. Also, there was no significant difference (P≥0.05) in that of 0.75% and 1.0% concentration of trona. HCN was highest in untreated AYB (9.89%) and lowest in AYB soaked with 1.00% concentration of trona (0.51). There was no significant difference (P≥0.05) in the HCN of AYB soaked with 0.5%, 0.75% and 1.0% concentration of trona. Tannin was highest in the untreated AYB (0.197%) and lowest in AYB soaked with 1.0% concentration of trona (0.179%). There was a significant difference (P≤0.05) in the tannin contents of AYB soaked with difference concentration of trona and untreated AYB. Saponin was highest in AYB (0.57%). There was no significant difference (P≥0.05) in the saponin of AYB soaked with 1.0% and 0.75% concentrations of trona though it was in an increasing trend. Flavanoid was highest in the untreated AYB (0.41%) and lowest in AYB soaked with 1.0% and 0.75% concentration of trona (0.31%). There was no significant difference (P≥0.05) in the flavanoid contents of AYB soaked with 0.5%, 0.75% and 1.0% concentrations of trona. TTA was highest in the untreated AYB (2.50%) and lowest AYB soaked with 0.75% and 1.0% concentration of trona (1.2%). There was no significant different (P≥0.05) in the TTA of AYB soaked with 0.25% and 0.5% concentrations of trona; 0.75%

and 1.0% concentrations of trona. Phytate was highest in the untreated AYB (0.322%) and lowest in 1.0% concentration of trona (0.187%). There was a significant difference ($P \leq 0.05$) in the phytate contents of untreated AYB and AYB soaked with different concentration of trona. Oxalate was highest in the untreated AYB (0.36%) and lowest in AYB soaked with 0.75%) concentrations of trona (0.26%).

There was no significant difference ($P \geq 0.05$) in the oxalate contents of AYB treated with different concentrations of trona. Phenol was highest in untreated AYB (0.16%) and lowest in AYB soaked with 0.75% and 1.0% concentrations of trona (0.07%). There was no significant difference ($P \geq 0.05$) in the phenols of AYB soaked with 0.50%, 0.75% and 1.0% concentrations of trona.

Saponins present in the AYB reduced to mineral levels when soaked with different concentrations of alum and trona for 24h and 48h. The result confirms with the report of Onimawo and Akubor. (2005), that alkaline washing or dry scouring and abrasive dehulling have been suggested as techniques for saponins reductions in legumes. Saponins are not destroyed during cooking. However trace elements of saponins are nutritionally beneficial because of their hypocholestorolemic activity (cholesterol lowering) (Pnimawo and Akubor, 2005).

Reduction in phytate, typsin inhibitors and tannins in the soaked AYB with different concentrations of alum and trona for 24h and 48h are in agreement with the report of Mubarak (2005) on the nutritional compensation and anti-nutritional factors of mung bean seeds (*phaseolus aurew*) as affected by some home traditional processes. Reduction of phytate using different chemicals and soaking time also confirms with the work of Wikipedia, (2005) report that home food preparation techniques can reduce phytic acid by simply cooking to some degree. More effective methods are soaking on and acid medium, lactic acid fermentation and spronting. Reduction in typsin inhibitor to minimal levels in the soaked AYB with alum and trona for 24h and 48h is in line with the previous work reported by Eicher and Satterlee (1988) that a substantial amount of typsin inhibitors have been reported to leach out of great Northern beans by soaking in acidic or alkaline solutions.

Fernandez *et al.,* (1993) observed that soaking fava beans in 0.7% sodium bicarbonate solution was more effective in decreasing typsin inhibitor activity than soaking in 0.1% citric acid solution probably due to the stability of inhibitor in acidic $P^{H.}$

Soaking of AYB with different concentrations of alum or trona for 24h and 48h gave oxalate values below the lowest published lethal dose of 600mg/kg in humans (SOPC, 2005).

Conclusion

It could therefore be concluded, based on the findings from this research work, that the use of alum and trona reduced the antinutrient properties of AYB seeds. Most antinutrients like alkaloid, saponin, HCN, tannin, phytate and oxalate significantly reduced by soaking in alum for 24h and 48h respectively. However phenols and Flavanoid were not affected by soaking in alum. Soaking in trona however caused reduction in flavanoid and phenol of which were stable to alum effect. It was also observed that on the whole, trona had more effect on the antinutritional properties than alum and this was attributed to the softening effect of trona on the cotyledon which probably had an increased solubility and leaching of the antinutrient into the soak water.

References

Amtfield, S.D; Ismond, M.A.H and Murray, E.D. (1985). The fate of antinutritional factors during the preparation of fava bean protein Isolate using a micclization technique. Canadian Institute of Fd. Sc. Tech. J. 18 (2): Pg 137-143.

A.O.A.C. (1990) Official method of analysis (11th edn). Association of official Agriculture Chemists, Washington. SC, USA.

Balagopalan, C; Padmaja, G; Nanda, S.K and moorthy, S.W. (1988). Cassava in food, feed and industry. CRC Press Inc. Florida Pg 187-189

Bliss, F.A. (1995). Cowpea in Nigeria. In proceeding of a symposition on Nutritional improvement of legumes by breeding 3-5 July 1995 New York, Nations, proteins Advisory group Pg 71-82.

Eggum, B.O. (1984). Factors affecting the protein quality of pigeon pea (Cajanus Cajan) as influenced by seed polyphenols. Plant foods for human nutrition. 43:171-179.

Eicher, N.J and Satterlee, L.D (1988). Nutritional quality of Great Northern bean proteins processed at barying pH. J. Food Sci, 53: 1139-1143

Enwere, N.J. (1986). Effect of temperature and drying on selected legumes. Msc. Thesis Department of Food Science and Technology. University of Nigeria, Nsukka. 21:337-294.

Fernandez, N.M. Aranda, P, Lopez Mrado, M. Urbano, G. Estrella, I. Sotonmayor, C; Diaz, C; Prodanor, M; Frisc, J. and Bidal-Balverde, C. (1993). Effect of processing on some nutritive factors of fava beans: Influence on protein digestibility and food intake in rats. In: Recent advances of research in antinutritional factors in Legume seeds; Wageningen Per: Wageninge, The Nether lands, Pg 467-471

Gbadge, P, N, Vairagar, P.R and Prasad, K (2008), Physical properties of click pee split (CicerarietiumL) Agricultural Engineering International: The CIGR Ejournal, Manuscript FP 07 039. Vol. x.

Gonzadez, S., pebon, ML, and Carulla, J (2002). Effects of tannins on the invitro ammonia release and dry matter degradation of soybean meal. Arch. Latinoam. Prod. Anim. 10 (2): 97-101.

Harborne J.B. (1993) Photochemical methods Chapman and Hall, New York

Henshaw F.O. and Sanni S.A. (1995): The effects of seed physical properties and chemical composition on the cooking properties of seven cowpea (vigna unguiculataa) varieties. Nigeria fd. 13:53-63

Hussein M.A, Akinyele I.O. Omololu A, and Akinlosotu A. (1984). Medical problems associated with consumption of cowpea, perception of Nigeria woman paper presented at the world cowpea Research conference, IITA, Ibadan November 4-9

Kumar, S. Singh, G.K. Kumar, R, Bhatia, N.K and Awasthi. C.P. (1984). Variation in quality traits of Pigeon pea (Cajanus Cajan L. Mill SP.) varieties. Journal of food science and Technology. 28:173-174

Lu, A.K. and Jorqunun, C. (1982). Education of quality legumes in Asia-journal of food science.

Mathew, F and Jouany, J.P (1993). 'Effect of chestnut tannin on the ferment ability of soybean meal nitrogen in the rumen 'Ann Zootech. 42:127

Mubarak, A.E (2005). Nutritional composition and antinutritional factors of mung bean seeds (phaseolus auureus) as affected by some home traditional processes J. food chemistry 89: Pp 489-495

Ngoddy, P.O, Enwere, N.J. and Onuora, V.I. (1986). Cowpea flour performance in akara and moi.moi preparations. Tropical Science 21:337-294

Ologhobo A.D., Apata, D.F. and Oyejide, A.(1993). Utilization of raw jack bean (Canavalia ensiformis) and jack bean fractions in diets for broiler chicks. Br.Poult. Sci., 34:323-337.

Onwuka, G.I (2005). Corporative studies on the winning potentials of black tamarind, local grape fruit and exotic apple. Department of Food Science and Technology, Michael Okpara University of Agriculture. Journal of Food Technology 4 (4):350-353.

Osagie, A.U. (1998). Post harvest research unit. Department of Biochemistry, University of Benin Pg 221-244

Prakash, D. and Misra, P.S. (1987), Protein and Amino acid composition of some leguminous seed. Plant food for human Nutrition. 37:29-32

SOPC(2005) Safety Officer in Physical Chemistry. Safety (MSDS) data for oxalic acid dehydrate. Oxford University.

SAS (2000) SAS Users Guide. (Version 8). Cary, NC: SAS Institute Inc.

Somari, R.O. and Balogh, E. (1993). Effect of soaking on the oligosaccharide conent of cowpea flour Journal of science food Agriculture. 61:339-343

Tobacco. E., Borreshi, Crobetto, G.M, Galassi, G., Colonbo, D and Cavallarin, L (2006). Effect of chestnut tannin on fermentation quality, proteolysis, and protein rumen degradability of alfalfa silage. J. Diary Sci. 89(12): 473-46.

Wikipedia C. Yard (2005) long bean In ;Free encyclopedia; http://www.buytikitorches.com Accessed,2005 Feb.,2007.