THE INTERNATIONAL JOURNAL OF SCIENCE & TECHNOLEDGE

Proximate and Anti-Nutritional Studies of Two Varieties of Germinated Acha Seed (Digitaria Exilis)

Ijeoma Lawretta Princewill-Ogbonna
Lecturer, Department of Food Science and Technology,
Michael University of Agriculture, Umudike, Abia, Nigeria

Abstract:
The aim of the study was to determine the effect of germination on the proximate and anti-nutritional composition of the two varieties of acha (Digitaria exilis). Germination increased the moisture content (9.70±0.04 to 10.45±0.04 and 9.80±0.02 to 10.63±0.01) for the white and yellow varieties respectively. There was significant reduction (P<0.05) in the ash content (1.92±0.00 to 1.61±0.02 and 1.90±0.02 to 1.84±0.01), crude fibre (2.41±0.01 to 2.22±0.01 and 2.38±0.01 to 2.14±0.01), crude fat (2.60±0.01 to 1.88±0.02 and 2.75±0.02 to 2.19±0.00), during the period of germination for white and yellow varieties respectively. There was complete reduction in the tannin content of the two varieties, hydrogen cyanide of the yellow variety after 48 hours of germination. Furthermore, germination reduced the tannin, phytate, hydrogen cyanide and saponin content of the white and yellow varieties of acha.

1. Introduction

Acha (*Digitaria exilis*) also known as fundi, hungry rice and Asian millet (N.R.C., 1996) is a nutritious cereal consumed mainly by Africans and Asians. The statistics in 2003, revealed that out of the 347,380 hectares of land devoted to Acha production in Africa, Nigeria alone provided almost half of the areas (FAOSTAT, 2003).Despite this high production rate of acha, it is still classified as Underutilized crop of Africa (Jideani et al 1996) Recent discoveries revealed that forming is cultivated and used as food and forage in the Dominican Republic of Island of South Africa.

The protein content and protein quality of *Digitaria exilis* is higher than the rest of the cereals (wheat, maize, millet, sorghum, barley, oat, etc.). Its protein content is high in leucine (9.8%), methionine (5-6%) and valine (5-8%). Jideani and Jideani, (2011) repoted that acha contains resistant starches. Resistant starches have shown promise in the management or prevention of certain diseases or health conditions. Acha has been used health maintanace such as the treatment of diabetics in some parts of Africa, control of diabetics (Jideani and Jideani 2011) Despite its nutritional quality, Acha is considered as one of the lost crops of Africa (Philip and Itoto, 2006).

Acha is consumed in diverse ways it can be consumed as a whole grain, porridge and cous cous, ground and mixed with other flours to make used for pastries. It has also been successfully used in beer production and beverage consumption.

Antinutrients are chemicals which have been evolved by plants for their own defense, among other biological
functions and reduce the maximum utilization of nutrients especially proteins, vitamins, and minerals, thus preventing optimal exploitation of the nutrients present in a food and decreasing the nutritive value.(Habtamu and Negussie 2014). Some the anti nutrients found in creals include phytic acid, lectins, tannins, saponins, amylase inhibitors and protease inhibitors . These anti nutrients have been shown to reduce the availability of nutrients.

Germination is receiving attention because of its probability to improve flavor and nutritional qualities of food (Hwei-ming *et al.*, 1997) During germination, polymers such as starch is broken into simple units (glucose) making it easier for the young rapidly growing plant to meet its energy requirements which in turn induces an increase in free limiting amino acids and available vitamins. Consequently, germinated food materials often have modified functional properties (Elin *et al.*, 2004; Kinsella, 1979)..

Germination has been shown to reduce anti-nutritional factors in legumes resulting in improved digestibility and utilization (Uwaegbute, et al., 2000). Mohammed *et al.* (2007) reported that germination increased the nutritive value of cereals, legumes while a decrease in the anti-nutrients was observed.

The study aims at evaluating the effects of germination on the proximate and anti-nutritional composition of two varieties (white and yellow) of Acha (*Digitaria exilis*).

2. Methodology

Two varieties of Acha (white and yellow) were purchased from Jos in Plateau State, Nigeria. The acha grains used in this study were fully mature and free of insect infestation.

2.1. Sample Preparation

The white and yellow varieties were sorted, cleaned to remove immature and extraneous matter. The seeds were soaked with water for 6 hours and germinated using a decom. The seeds were germinated for 24h, 48h washed, dried to stop sprouting. The dried seeds were de-hulled with a mortar and pistle. The de-hulled seed were ground into flour using hammer mill and sieved using a sieve size of 1mm aperture. Flour samples were packaged in a ziplock bag and stored for proximate and anti-nutrient analysis.

2.2. Determination of Proximate Analysis

2.2.1. Determination of Moisture Content

Exactly two (2) grams of each of the samples was weighed out with the aid of an analytical balance into dried, cooled and weighed dish in each case. The samples in the dishes were then put into a Genlab moisture extraction oven set at 1050c and allowed to dry for 3 hours. When this time elapsed, the samples were then transferred into a desiccator with the aid of a laboratory tong and then allowed to cool for 30 minutes. After cooling in the desiccator, they samples were weighed again and their respective weights recorded accordingly. The above processes were repeated for each sample until a constant weight was obtained in each case. The difference in weight was calculated as a percentage of the original sample.

Percentage moisture content $= \frac{W2 - W3 \times 100}{W2 - W1 \quad 1}$

Where W1 = Initial weight of the empty dish, W2 = Weight of the dish + undried sample and W3 = Weight of the dish + dried sample.

2.2.2. Determination of Protein

The micro-Kjeldahl method was employed to determine the total nitrogen and the crude protein (N×6.25) (AOAC, 2000).

2.2.3. Determination of Ash Content

Exactly two (2) grams of each of the samples was weighed out with the aid of an analytical balance into a dried cooled and weighed crucible in each case. The samples were then charred by placing them on a Bunsen flame inside a fume cupboard to drive off most of the smoke for 30 minutes. The samples were thereafter transferred into a pre-heated muffle furnace already at 5500C with the aid of a laboratory long. They were allowed to stay in the furnace for 3 hours until a white or light grey ash resulted. Samples that remained black or dark in color after this time had elapsed were moistened with small amount of water to dissolve salts, dried in an oven and then the ashing processes repeated again. After ashing, the crucibles were then transferred into a desiccator with a laboratory long. When they cooled, they were each weighed again and recorded accordingly.

Percentage Ash Content $= \frac{W3 - W1 \quad x \quad 100}{W2 - W1 \quad 1}$

Where W1 = weight of empty crucibles, W2 = weight of crucible + food sample before ashing and W3 = weight of crucible + ash

2.2.4. Crude Fibre Determination

The crude fiber was determined by the Weende method as described by James (1995). Exactly 2g of each sample was defatted. The defatted sample was boiled in 200ml of 1.25% Tetra Oxo Sulphate (vi) solution under reflux for 30 minutes. After that the sample was washed with hot water, using a two-food muslin cloth to trap the particles, the washed sample was transferred quantitatively back to the flask and boiled again in 200ml of 1.25% sodium hydroxide solution for 30 minutes and washed before it was transferred to a weighed porcelain crucible and dried in the oven at 105ºC for three hours. After cooling in a desiccators, it was re-weighed.

The percentage crude fiber was calculated as follows

$$\% \text{ crude fibre} = \frac{W2 - W3 \times 100}{W1}$$

Where:

W1 = weight of sample

W2 = weight of sample + crucible

W3 = weight of crucible + ash

2.2.5. Determination of Fat Content

Two hundred and fifty milliliters (250ml) of boiling flasks were washed with water, dried in a Genlab oven set at 1050c for 30 minutes, cooled in a desiccator and then used for each sample. The flasks were firstly labeled, weighed with an analytical balance and then filled with 300ml of petroleum ether in each. Then, five grams of each of the samples was weighed out with an analytic balance into a correspondingly labeled thimble. The extraction thimbles were in each case tightly plugged with cotton wool. The soxhlet apparatus was then assembled and allowed to reflux for 6 hours.

When this time elapsed, the thimble was removed and the petroleum ether was collected in each case in the top of the container in the set up and drained into another container for re-use. The flasks were removed in each case and then dried in a Genlab oven at 1050c for 1 hour. After drying, they were transferred into a desiccator and allowed to cool and weighed. The percentage fat was calculated for each sample thus

Percentage Fat $= \frac{C - A \quad x \quad 100\%}{B \qquad 1}$

Where A = weight of empty flask, B = weight of the sample and C = weight of oil after drying.

2.2.6. Determination of Carbohydrate Content

The carbohydrate content of the sample was determined by estimation using arithmetic difference.

% CHO = 100 - (% moisture+ %protein % ash+ %fat content+ % Crude fiber)

2.3. Determination of Anti-nutrients

2.3.1. Determination of Saponin

The spectrophotometric method of Bruner (1984) was used for saponin determination. Exactly 2g of the sample was put into a 250ml beaker and 100ml of ISO butyl alcohol added. A shaker was used to shake the mixture for 5 hours to ensure uniform mixing. The mixture was then filtered using the No.1 Whatman filter paper into a 100ml beaker containing 20ml of 40% saturated solution of magnesium carbonate ($MgC0_3$). The mixture obtained was filtered again through No1 filter paper to obtain a clean colorless solution was taken to a 500ml volumetric flask using a pipette made to mark with the distilled water. It was allowed to stand for 30minutes for the need color to develop. The absorbance was read after the color development on the spectrophotometer at 350nm.

$$\text{Saponin} = \frac{\text{Absorbance of sample}}{\text{Conc. of sample}} \times \text{Absorbance of Stan.}$$

2.3.2. Tannin Determination

The method of Pearson (1976) was used. Exactly 1g of each sample was weighed into a centrifuge tube with 2ml of distilled water. It was centrifuged at 1500rpm for 10 minutes. The centrifuge samples were then poured out into a beaker and the supernatant (extract) dispersed. One ml of NaCo3 and Folin Denis reagent was added in the beaker and allowed to settle. Therefore, the readings were taken using a spectrophotometer. Tannin could be calculated as follows:

$$\%\ \text{Tannin} = \frac{A_n}{A_s} \times \frac{C}{W} \times 100 \times \frac{V_F}{V_a}$$

A_n = absorbance of test sample

As = absorbance of standard sample

C = concentration of standard solution.

2.3.3. Determination of Hydrogen Cyanide

The method of Onwuka (2005) was used for this determination. Exactly 5g of the sample was made into paste by dissolving in 50ml of distilled water in a conical flask. The mixture was allowed to stay over-night. The solution was filtered. Two milliliters of the filtrate was mixed with 4ml of alkaline pyrate solution and incubated in a water bath for 5mins for color development (reddish brown) and absorbance was at 490nm. A blank was prepared using 2ml distilled water. The cyanide content was extrapolated ajamide standard curve.

$$\text{Calculate: HCN} = \frac{Vf \times 1 \times 100 \times 100}{Va \quad 100 \quad W \quad 1}$$

Where Vf = total volume of extract

Va = volume of extract used

W = weight of sample used.

2.3.4. Determination of Percentage Phytate

A known weight of each ground sample was soaked into 100ml of 2% HCl for 5 hours and filtered. Twenty-five cubic centimetres (25cm3) of the filtrate was taken into a conical flask and five cubic centimetres (5cm3) of 0.3% ammonium thiocyanate solution was added. The mixture was titrated with a standard solution of FeCl3 until a brownish yellow colour persisted for 5 minutes.

The concentration of the FeCl3 was 1.04% W/V

The Calculation mole ratio of Fe to phytate = 1:1

$$\text{Concentration of phytate phosphorous} = \frac{\text{Titre value x } 0.064}{1000 \text{ x Weight of sample}}$$

Phytic acid content was calculated on the assumption that it contains 28.20% phosphorus by weight. (Reddy *et al*., 1982)

3. Results

Table 1 shows the proximate composition of the Acha flour samples. The moisture content varied from 9.70 – 10.63%. The yellow variety germinated for 48 hours had the highest moisture content (10.63%) which deferred significantly from the rest of the samples. The dry matter, content, ash, crude fat, crude fibre, protein content of the un-germinated samples were significantly higher than that of the other samples. This implies that germination significantly ($P< 0.05$) decreases the dry matter content (90.20 – 89.39%) and (90.23 – 89.54%), ash (1.90 – 1.65% and 1.92 – 1.61%), crude fibre (2.38 – 2.14% and 2.41 – 2.22%) and crude fat (2.75 – 2.19 and 2.60 – 1.88%) for the yellow and white variety respectively. The carbohydrate content of the samples varied from 73.52 – 74.85% and 73.43 – 75.83%) for a yellow and white variety respectively.

The results of the anti-nutritional composition of the samples are presented in Table 2. Germination decreased the tannin content in the two varieties of Acha from 0.12 – 0.00 and 0.14 – 0.00 for the yellow and white variety respectively. There was no significant difference ($P<0.05$) in the tannin content of the yellow and white variety.

There was no significant difference in the phytate content of the yellow and white variety. Germination also decreased the phytate content of the samples from 0.95 – 0.23 and 1.06 – 0.12 for white and yellow variety respectively. There was no significant difference ($P<0.05$) in the phytate content of the yellow and white variety.

A total reduction in the hydrogen cyanide content of the yellow variety was observed after 48 hours of germination. There was difference among the samples. The hydrogen cyanide content (0.15 +0.01) of the yellow variety was significantly ($P<0.05$) higher than that of the white variety (0.12+ 0.01)

There was no significant difference ($P<0.05$) in the saponin content of the yellow and white variety after 24 hours of germination. However, a significant difference ($P<0.05$) was observed after 48 hours of germination from 0.30 – 0.13 and 0.26 – 0.15 for white and yellow variety respectively.

4. Discussion

The increase in the moisture content of the germinated samples could be as a result of water sprinkled on the seeds during germination. However, a decrease was observed in the dry matter content of the germinated samples. The breaking down of seed reserves (Vanderstuep, 1951) and enzyme activity which occurs during germination could lead to a loss of total dry matter and increase in total protein. (Lorenz, 1980). The decrease in the carbohydrate content of the germinated samples could be as a result of α – amylase activity (Maneemegala and Mandakumar, 2011). The α – amylase break down complex carbohydrate to simpler sugars which were utilized by the growing seedling in the initial stages of germination (Onwuka *et al*, 2009).

Many researchers had reported a decrease in the fat (lipid) content of germinated seeds (Chandiasiri *et al.*, 1990). As germination progresses, half of the tri-acylglycerol acts as the major carbohydrate source for seed growth (Bai *et al.*, 1997). Decrease in fiber content may be due to low hydrolysis of complex carbohydrate to higher absorbable sugar to liberate low fiber (Sigah and Ali, 2000).

Reduction in the phytate content of cereals and legumes during germination have also been reported in literature. The decrease could be attributed to a large increase in phytase activity (Bai *et al.*, 1997). Phytate plays a vital role in mineral availability (Philippy and Johnson, 1985). Phytate decrease the availability of zinc, manganese, copper, calcium, magnesium, iron as well as protein (Maga, 1982, Beleia *et al.*, 1993). The phytate levels obtained in the untreated samples of the yellow and white varieties were higher than those reported for bambara groundnut, 0.29%, pigeon pea, 0.2% (Igbedioh et al., 1994); wild yam tubers, 0.18 to 0.36% (as phytic acid) (Bhandari and Kawabata, 2004); yam 0.47%, cassava, 0.4% and maize 0.16% (Adeyeye et al., 2000). Tannins inhibit the digestibility of protein (Umoh, 2013).

The lethal dose range for humans, of HCN ingested is estimated to be only 0.5 to 3.5mg/kg – body weight (Brandbury, 1991). According to Makkar et al. (1998), FAO/WHO recommended safe limit of cyanide for human consumption is 10 mg·100g-1.

The knowledge of cyanogenic glycosides content of food is vital because cyanide being an effective cytochrome oxidase inhibitor interferes with aerobic respiratory system (Umoh, 2013).

5. Conclusion

Germination increased the moisture and protein content of the two varieties of acha flour, however a significant reduction was observed in the fat, crude fidre and ach content of the flour samples. Furthermore germination reduced the tannin, hydrogen cyanide and phytate and saponin content.

Sample	Dry matter%	Moisture %	Ash %	Crude Fiber %	Crude Fat %	Crude Protein %	CHO%
Control W	90.23^{a}± 0.04	9.70^{d}±0.04	1.92^{a}±0.00	2.41^{a}±0.01	2.60^{b}±0.01	8.63^{e}±0.02	73.54^{e}±0.04
Control Y	90.20^{a}±0.01	9.80^{cd}±0.01	1.90^{a}±0.02	2.38^{a}±0.01	2.75^{a}±0.02	8.72^{d}±0.01	73.52^{f}±0.02
Y24	89.59^{b}±0.09	10.40^{b}±0.09	1.84^{b}±0.01	2.16^{d}±0.01	2.21^{c}±0.02	9.33^{b}±0.02	74.85^{b}±0.05
Y48	89.37^{d}±0.01	10.63^{a}±0.01	1.65^{c}±0.00	2.14^{e}±0.01	2.19^{d}±0.00	9.65^{a}±0.01	73.86^{d}±0.01
W24	90.18^{a}±0.02	9.81^{c}±0.02	1.72^{d}±0.03	2.22^{c}±0.01	1.88^{f}±0.02	9.17^{c}±0.01	75.83^{a}±0.02
W48	89.54^{c}±0.04	10.45^{b}±0.04	1.61^{e}±0.02	2.26^{b}±0.01	1.93^{e}±0.02	9.75^{a}±0.00	74.43^{c}±0.05

Table 1: Proximate Compositions of Samples of Acha Flour

Values are means ± standard deviation of three determinations.

Means with different superscripts within each column are significantly different ($p<0.05$)

Note:

- Control W represents control for white acha
- Control Y represents control for yellow acha
- Y24 represents yellow acha germinated for 24hrs
- Y48 represents yellow acha germinated for 48hrs
- W24 represents white acha germinated for 24hrs
- W48 represents white acha germinated for 48hrs

Sample	HCN (mg/100g)	Phytate (mg/100g)	Tannin (mg/100g)	Saponin (mg/100g)
Control W	0.12^{d}±0.01	0.95^{a}±0.00	0.14^{a}±0.00	0.30^{a}±0.01
Control Y	0.15^{c}±0.01	1.06^{a}±0.00	0.12^{a}±0.01	0.26^{a}±0.01
Y24	0.70^{a}±0.01	0.25^{c}±0.00	0.096^{b}±0.00	0.20^{b}±0.00
Y48	ND	0.12^{e}±0.00	ND	0.15^{c}±0.00
W24	0.16^{b}±0.01	0.34^{b}±0.00	0.06^{c}±0.00	0.20^{b}±0.00
W48	0.08^{e}±0.01	0.23^{d}±0.00	ND	0.13^{d}±0.00

Table 2: Values for the Anti-Nutritional Properties of Acha Flour Samples

Values are means ± standard deviation of two determinations.
Means with different superscripts within each column are significantly different ($p<0.05$)
NOTE:

- Control W represents control for white acha
- Control Y represents control for yellow acha
- Y24 represents yellow acha germinated for 24hrs
- Y48 represents yellow acha germinated for 48hrs
- W24 represents white acha germinated for 24hrs
- W48 represents white acha germinated for 48hrs
- HCN represents Hydrogen cyanide

6. References

i. Philip T. and Itodo F., (2006). Acha (Digitaria spp); a rediscovered indigenous crop of West Africa. Agricultural engineering International: the CIGR journal limited overview. No. 23, Vol. VII, December, 2006.
ii. FAOSTAT, (2003) Database. Food and Agriculture organization of the United Nations.
iii. N.R.C., 1996. Lost crops of Africa; Volume 1; Grains National research council. National academy press, Washington D.C. pp 59 – 75.
iv. Ogbonnaya C. and Abdul-kadir (2008). Proximate chemical composition of Acha (Digitaria exilis and Digitaria iburua). Grains Journal of food technology 6(5): 214 – 216.
v. Inyang C. U. and zakari U. M. (2008). Effect of germination of pearl millet on proximate, chemical and sensory properties of instant fura. Nigerian cereal food. Pak J. Nutr. 7. 9 – 12.
vi. Mohammed E. M, Amro B. H., Mashier A. S., Elfadil E. B., 2007. Effect of processing, followed by fermentation on anti – nutritional factora content of pearl millet (Rennisetum glaucum L.) cultivars. Pak. J. Nutr. 6: 463 – 467.
vii. Vanderstoep J. 1981. Effect of germination on the nutritive value of legumes. Food technol. 38: 33 -85.
viii. Lorenz K., 1980. Cereal sprouts: composition nutritive value, food application. Crit. Rev. Food. Sci. Nutri. 13: 353 – 385.
ix. Onwuka C. F., Ikewuchi C. C., Ikewuchi C. J., Ayalogu O. E., 2009. Investigation on the effect of germination on the proximate composition of African yam bean (Sphenostylis stenocarpa Hochst ex A Rich) and cluted pumpkin (Telfera accidentalis) J. Applied Sci. Environ Mange, 13: 59 – 61.
x. Elin, H., Senol, I., Ainsworth, P. (2004). Effect of fermented/germinated cowpea flour addition on the rheological and baking properties of wheat flour. Journal of Food Engineering, 63, 177–184.
xi. Uwaegbute, A. C., Iroegbu, C. U., & Eke, O. (2000). Chemical and sensory evaluation of germinated cowpeas (Vigna unguiculata) and their products. Food Chemistry, 68, 141–146.
xii. Kinsella, J. E. (1979). Functional properties of soy protein. Journal of the American oil Chemists Society, 56, 242–257.
xiii. Makkar, H.P., Beckar, K. (1998). Plants toxins and detoxification methods to improve feed quality of tropical seeds. Asian-Australian Journal of animal science, 12, 467-480.
xiv. Umoh, E. O. 2013. Anti-nutritional factors of false yam (Icacina tricachantha) flour. Internet Journal of Food Safety, Vol.15, 2013, pg.78 – 82.
xv. Bau, H., Villaume, C., Nicolas, J., & Mejean, L. (1997). Effect of germination on chemical composition, biochemical constituents and antinutritional factors of soybean (Glycine max) seeds. Journal of the Science of Food and Agriculture,73, 1–9.
xvi. Hwei-Ming B, Christian V, Jean-Pierre N, Luc M (1997). Effect of germination on chemical composition, biochemical constituents and antinutritional factors of soya beans (Glycine max) seeds J. Sci. Food Agric. 73: 1-9.
xvii. Reddy NR, Sathe SK, Salunkhe DK (1982). Phytates in legumes and cereals. Adv. Food Res. 28: 1-92.
xviii. Chandrasiri, V., Bau, H. M., Villaume, C., Giannangeli, F., and Mejean, L. (1990): Effect of germinated and heated soybean meals on plasma cholesterol and triglycerides in rats. Report Nutr Dev, 30, 611 – 618.
xix. Singh, V., & Ali, S. Z. (2000). Acid degradation of starch. The effect of acid and starch type. Carbohydrate Polymers, 41, 191–195.
xx. Maga, J. 1982. Phytate: Its chemistry, occurrence, food interactions, nutritional significance and methods of analysis. Journal of Agricultural and Food Chemistry 30: 1-9
xxi. Beleia, A., Thu Thao, L.T. and Ida, E.I. 1993. Lowering phytic phosphorus by hydration of soybean. Journal of Food Science 58: 375-377

xxii. Igbedioh, S.O., Olugbemi, K.T. and Akpapunam , M.A. (1994) Effects of Processing methods on Phytic acid level and some constituents in Bambara Groundnut (Vigna subterranean) and pigeon pea (Cajanus cajan). Food Chem. 50: 149 –151
xxiii. Brandbury, J.H. (1991) Properties and Analysis of Antinutritional factor in Foods. ASEAN Food Journal, 6 (4) : 123-128.
xxiv. Onwuka, G. I. (2005) Food Analysis and Instrumentation: Theory and Practice. Naphtahli Prints, Lagos. Nigeria.
xxv. Pearson, D. A. (1976) Chemical Analysis of Foods. (7th edn) Church Hill, Living stone, Edinburgh.
xxvi. Brunner JH (1984). Direct Spectrophotometric determination of saponin. Anal Chem. 1984;34:1314– 1326.
xxvii. Bhandari, M.J. and Kawabata, J. 2004. Organic acid, phenolic content and antioxidant activity of wild yam (Dioscorea spp.) tubers of Nepal. Food Chemistry 88: 163-168
xxviii. Adeyeye, E.I., Arogundade, L.A., Akintayo, E.T., Aisida, O.A., and Alao, P.A. (2000). Calcium, zinc, and phytate interrelationships in some foods of major consumption in Nigeria. Food Chemistry, 71: 435-441.
xxix. James, C.S, (1995). Experimental procedures, analytical chemistry of foods. Chapman and hall, New York.
xxx. Jideani I. A. and Jideani V. A. Developments on the cereal grains Digitaria exilis (acha) and Digitaria iburua (iburu) (2011) . J Food Sci Technol . 48(3):251–259.
xxxi. Habtamu F. G, and Negussie R.(2014). Antinutritional Factors in Plant Foods: Potential Health Benefits and Adverse Effects.
xxxii. International Journal of Nutrition and Food Sciences. . 3, (4). 284-289. doi: 10.11648/j.ijnfs.20140304.18