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Nutrient and Phytochemical Composition and Effects of Boiling on *Amaranthushybridus* Seeds

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Abstract:
Introduction: This study evaluated the nutrient and chemical value of Amaranthushybridus seeds. The study was aimed to: determine the nutrient and photochemical content of the seeds as well as to determine the effects of boiling on the nutrient composition of A. hybridus seeds.
Methodology: Standard analytical methods were used to assess the chemical composition of the seeds. The seeds were treated as boiled and raw samples. These samples were milled into fine flour and used for the analysis.The statistical analysis was done in triplicate and the mean, standard deviation was calculated, the T-test value was used to compare the means.
Result:The proximate analysis showed the percentage (%) moisture, ash, crude protein, Crude fibre, fat, and carbohydrate content of the seed as 12.7, 7.2, 12. 87, 12.7.7, 5.6. and 48.93 respectively for the raw sample while there was a little reduction in the boiled. Elemental analysis mg/100g indicated that the seed contained zinc (6.45), lron (2.42), calcium (2.50) Iodine (9.03ųg/l00g). The vitamin composition of the seed in mg/100g was vitamin A (9.6RE), Ascorbic Acid (0.56), Riboflavin 6.05 and thiamin 2.28. The antinutrient composition in mg/100g was tannin 3.98, oxalate 5.25 and phytate 3.78. The phytochemical content of the seed was high and they are saponin 23.00 flavonoid 2.00, phenol 0.87, Alkaliod 6.00 and carotenoid 9.05 all in mg/100g.
Conclusion:The data of this project work proved that the seed contains appreciable amount of proteins, fat, crude fibre, vitamins, minerals and phytochemicals. The high phytochemical contents show that the seed could be useful medically and in pharmacological industries.

1. Introduction

Most developing countries depend on starch based foods as the main staple food for the supply of both energy and protein. This accounts in part for the deficiency which prevails among the population as recognized by food and Agricultural Organization (1). The continuous increase in population has brought about decrease in available food resources thus leading to global malnutrition especially Protein Energy Malnutrition (PEM) and micronutrient deficiency. In most areas, the causes of malnutrition in Nigeria are ignorance, taboos, cultural and religious beliefs.

Our indigenous crops are being neglected simply because there are no best techniques for processing and preserving these food crops. This has led to low consumption level of these readily available and cheap foods. In Nigeria, as in most other tropical countries of Africa, the daily diets are dominated by starchy staple foods. Vegetables act as the cheapest and most readily available source of important vitamins, minerals and essential amino acids, because they are affordable, available and acceptable (2 - 3)But many of the local vegetables and seed materials are under-exploited because of inadequate scientific knowledge of their nutritional potentials (2). Though several works has been done on the nutrient composition and functional properties of various types of edible plants crops in use in developing countries(4 -9). Much still needs to be done on the use of the seeds of these plants. According to Ihekoronye and Ngoddi(10), malnutrition can only be curbed through improvement in diversification and variation of diet, indigenous food production capacity and knowledge of their nutrition value. So to reduce the prevalence of energy, protein and micronutrient deficiency in Nigeria, strategies such as dietary diversification, fortification are needed (3). Dietary diversification means choosing a number of different foods within any given food group rather than eating the same kind of food day after day (3). So in order to diversify our food base many researchers have gone into researching on a promising traditional food that can be integrated or re-integrated into modern diet to improve human health (11). One of these traditional plants is*Amaranthushybridus*.

Amarathushybridus is one of the under-utilized crops which belongs to the Amaranth family. It is popularly known as ‘pigweed’ and commonly called ‘Green or African spinach’. It is a herbaceous plant growing up to 6-feet high. The leaves of this crop are commonly consumed in different parts of the world. In Nigeria, the leaves are combined with condiments used to prepare soup ‘vegetable soup’ (9). In Congo the leaves are eaten as spinach or green vegetables (12). The leaves are boiled and mixed with groundnut source and eaten as salad in mozambique and West Africa [13]. It has been proposed as an inexpensive natural crop that could be cultivated by indigenous people on rural area for several reasons which includes: easily cultivated andproduces a lot of seeds used as grain, highly tolerant to weather condition and large amounts of protein and essential amino acids such as lysine in the leaf.

According to Railey[14], it is also recommended for people with low blood count. It goes with all Nigerian carbohydrate dishes and its known in Yoruba language as 'arowojeja' (we have left over money for fish). Railey[14]also stated that this food has the potential to improve nutrition, boost food security, foster rural development, support land care and diversify our food base.

Besides, plants produce a wide variety of compounds called phytochemicals. They are found in fruits, vegetables, beans, grains and other plants. They are promoted for the prevention and treatment of many health conditions, including cancer, heart disease, diabetes and high blood pressure. There is some evidence that certain phytochemicals may help prevent the formation of potential carcinogens, block the action of carcinogen on their target organs or tissue, or act on cells to suppress cancer development[15].There are several major groups of phytochemicals. The polyphenols include a large subgroup of chemicals called flavonoids. These flavonoids are studied to find out whether they can prevent chronic disease such as cancer and heart disease. Polyphenol also acts as antioxidants that rid the body of harmful molecules known as free radicals. Carotenoids, which gives yam, squash, apricots etc. their orange colour are also promoted as anticancer agents [15].Another group of phytochemicals, called allyl sulfides are found in garlic and onions and may help to stimulate enzymes that help the body get rid of harmful chemicals. They may also help to strengthen the immune system.

Craig, [16] states that these phytochemicals when consumed in a healthy diets are likely to be helpful and are unlikely to cause any major problems, so a balanced diet that includes 5 or more servings a day of fruits and vegetable, along with foods from a variety of other plant source such as seeds, nuts, whole grain cereals and beans is likely to be more effective in reducing cancers risk than consuming one particular phytochemical in large amounts.

Based on this, it is imperative to conduct a study on the nutrients contents and phytochemical properties of *Amaranthushybridus*seed so as to help the populace in their food selection, combination and consumption thereby reducing the effect of malnutrition.

2. Material and Methods

2.1. Sample Collection and Treatment

The sample of a hybridus seed were obtained from a household garden in Nsukka. The seeds were collected together with the flower, it was then winnowed manually to separate the seed from the flower and then winnowed again to remove the chaffs.

The sample was then divided into two portions and treated differently. One portion was boiled and the other raw, this was to see the effect of boiling on the sample. For the boiled portion 50g of the sample was collected boiled for 30 minutes until it was tender under a temperature of 120^{O}C. It was then sun dried for 45hrs and milled with Glen Creston electric miller type DFH 48 into fine flour. The raw portion was also milled into fine flour. The weighing balance used to measure the samples for this analysis was Mettler'sUSA.

2.2. Proximate Analysis

Moisture and Ash content of the sample was determined using the method described by AOAC [17].Crude protein was determined using micro-kjeldahl method as described by Pearson[18]. The fat content was analyzed using the soxhlet method described by Pearson, (1976). Crude fiber as determined using weaned method as described by AOAC [17]. Carbohydrate was calculated by difference that is the quantity of moisture, ash, crude protein, fiber and fat was subtracted from 100 and the reminder was value of carbohydrate.

2.3. Vitamin Analysis

The quantity of vitamin A in *A.hybridus* was determined using Haborne method as described by Pearson [18].Determination of thiamin was done using the method described by Pearson [18].Vitamin B2Riboflavindetermination was conducted using method described byonwuka[19].Folic acid determination was done using Association of vitamin chemist, *Method of vitamin Assay*manual [20].

2.4. Mineral Determination

Iron determinationby phenanthrolinemethod asdescribedbyPearson [18].Zinc determination using dithizone method as described by Pearson [18].Calcium determination was done using AOACmethod[17].

2.5. Antinutrients Determination

Tannin, Phytateand Oxalate determinations were done by methods describes by[17].

2.6. Phytochemical Analysis

The following phytochemicals were analysed in this work; phenols, flavonoids,saponin, alkaloid and caroteniod.Total phenols using spectrophotometric method by Obadori*et al.*,[21], photoelectric colorimeter model AE-11D China was used.Saponinwasdeterminedusingmethod by Obadori*et al*[21], Alkaloid wasdeterminedusingmethod byHarborne[22].Caroteniodwas determined using spectrophotometer described by Havalampaand Kard[23].

2.7. Statistics Analysis

The statistical analysis was done in triplicate and the mean, standard deviation was calculated, the T-test value was used to compare the means [24].

3. Results

Parameter	Treatment		Test value
	Raw (%)	Boiled (%)	
Moisture	12.7±1.5	14.13±0.51	0.133
Ash	7.20±1.5	4.60±1.2	0.002
Protein	12.87±0.1.5	12.62±1.45	0.693
Fat	5.6±2	5.50±2.0	0.013
Crude fibre	12.7±0.3	9.20±1.03	0.14
Carbohydrate	48.93±0.7	47.41±1.2	0.843

Table 1: Proximate composition of raw and boiled A. hybridusseeds
Mean ± S.D of triplicate determination

Decision rule: If the difference between the two means is greater or equal (≥) to the "T test" value, then the treatment means are significantly or statistically different.

Vitamin	Raw	Boiled	T test value
Ascorbic acid (c)	0.56±0.3	0.41±0.4	0.122
Thiamine (B_1)	2.28±0.75	1.74±0.4	0.159
Riboflavin (B_2)	6.05±1.04	4.07±2.0	0.07
Vitamin A (RE)	9.06±0.4	16.9±1.7	0.21

Table 2: The vitamin composition of boiled and raw of Amaranthushybridus seeds (mg/100g)
Mean ± S.D of triplicate determination

Decision rule: If the difference between the two means is greater or equal (≥) to the "T test" value, then the treatment means are significantly or statistically different.

Table 1 presents the proximate values of processed (boiled) and unprocessed (raw) *A. hybridus* seeds samples. Processing had varied effect on the moisture content of the two samples. The raw sample had 12.7% while the boiled sample had 14.13%. The ash content of the samples differed significantly ($P<0.05$). The raw sample had 7.2% while boiled had 4.6%. There was a slight difference in the protein content of the seeds from (12.87%) in the raw to (12.62%) in the boiled. The difference was not significant ($P<0.05$). The effect of boiling on the fat content was insignificant, it reduced from (5.6%) to (5. 50%). The crude fibre values also differed. The boiled sample had less fibre 9.20% compared to 12.7% of the raw samples. The mean difference was significant ($p< 0.05$). The carbohydrate content of raw sample was also higher (48.93%) compared to (47.41%) in the boiled sample, there was a significant difference between the two samples ($P< 0.05$).

Table 2 above presents the vitamin composition of raw and boiled *A. hybridus* seeds. Boiling decreased the ascorbate value (0.4mg), the boiled sample had higher value (0.56mg). The thiamin value (1.7mg) of the boiled was lower than the value (2.28mg) for the control. There was reduction in the riboflavin content. The boiled sample had lower value of 4.01 when compared to the raw sample. There was a wider variation in the vitamin A content of the samples. The raw had 9.06 RE while the boiled had 16.9 RE.

Mineral element	Raw	Boiled	T-test value
Zinc (Zn)mg/100g	6.45±1.04	2.86±1.0	0.023
Iron (Fe) mg/100g	2.42±0.42	1.60±0.7	0.037
Calcium(Ca)mg/100g	2.50±1.61	0.03±0.01	0.078
Iodine (I)mg/100g	9.0±1.28	3.94±0.4	0.02

Table 3: mineral composition of raw and boiled Amaranthushybridus seeds
Mean ± S.D of triplicate determination

Decision rule: If the difference between the two means is greater or equal (≥) to the "T test" value, then the treatment means are significantly or statistically different.

Antinutrients	Treatments		T-test value
	Raw(mg/100g)	Boiled(mg/100g)	
Tannin	3.98±1.15	4.84±1.2	0.877
Oxalate	5.75±1.6	5.01±2.02	0.757
Phytate	3.78±0.85	3.46±0.3	0.716

Table 4: Antinutrient of composition of raw and boiled Ahybridus seeds
Mean ± S.D of triplicate determination

Decision rule: If the difference between the two means is greater or equal (≥) to the "T test" value, then the treatment means are significantly or statistically different.
Table 3 above presents the effect of boiling on the mineral composition of *A. hybridus* as against the raw sample. The zinc value varied. Boiled sample had lower Zinc value (2.86mg/100g) than the raw sample (6.45mg). The difference was significant ($P<0.05$). It also reduced the iron content from (2.42mg/100g) in the raw sample to (1.60mg/100g) in the boiled sample. The iodine value varied greatly with 3.94µg/100g in the boiled sample compared to 9.03µg/100g in the raw. The differences between the calcium content were not comparable. The raw sample had 2.50mg/100g and the boiled sample had 0.03mg/100g. The difference between the means of the two samples shows that the zinc, iron calcium and iodine contents of the raw was statistically different ($P<0.05$).
Table 4 above presents the effects of the processing method (Boiling) on the antinutrient composition of the seed samples. Boiling the seed increased the tannin content from (3.98mg/100g) to (4.84mg/100g) in the raw and boiled respectively and the difference (0.86) between the means which is less than the T-test value (0.82) shows that the contents of tannin in the two samples is comparable.
But oxalate reduced from (5.7mg/100g) to (5.01mg) in the raw and boiled respectively. Boiling had the same effect on phytate composition, it reduced from (3.78mg/100g) to (3.46mg/100g) respectively.

Phytochemical	Treatment (mg/100g)		T-test value
	Raw	Boiled	
Sapaonin	23.00±0.45	1.00±0.75	6.2
Flavoniod	2.00±1.01	1.01±0.25	0.153
Phenol	0.87±0.17	0.00±0	0..013
Alkaliod	6.00±1.03	6.00±1.05	1
Carotenoids	9.05±1.6	7.04±1.4	0.00

Table 5 Phytochemical composition of raw and boiled Amaranth's hybridus seeds
Mean ± S.D of triplicate determination

Decision rule: If the difference between the two means is greater or equal (≥) to the "T test" value, then the treatment means are significantly or statistically different.
Table 5 above shows the effect of boiling on the phytochemical content of the seed as compared with the boiled. There was a drastic decrease in saponin content of the seed from 23mg/100g to 1.00mg/100g. The content of saponin in the raw was statistically higher than that of the boiled ($P<0.05$). The flavoniod content also reduced with (2.00mg/100g) in the raw and (1.0mg/100g) in the boiled. The same effect was found in phenol and carotenoid. Phenol content reduced from (0.87mg/100g) to (0.00) while caroteniod reduced from 9.05mg/100g to 7.04mg/100g. Their content in the raw sample was statistically higher than that of the boiled sample ($P<0.05$). But the Alkaloid content did not decrease, it remained the same for the two samples (6.00mg/100g) and (6.00mg/100g) respectively. This shows that the content of alkaloid in the two samples were comparable.

4. Discussion

4.1. Proximate Composition

Moisture content of the boiled sample was higher than that of the raw samples. This may be attributed to absorption of much water by the seeds during boiling. But the moisture content of the seed (12.7%) was low when compared to that of the leaves (83.48%) [11]. The high ash content of the raw sample was expected, this may be because of the dry nature of the seed which concentrated the dry matter. Then the lower content of ash in the boiled sample may be due to leaching of ash into the boiling water. This ash content of this seed (7.20%) contrast the (2.85%) ash content found in *Amaranthus*specie planted in Uganda [25]. There was reduction in the protein content of the boiled sample which could be that the boiling temperature denatured the protein in the seed. The protein content (12.87%) was comparable to the protein content (12.37%) in amaranth specie planted in Uganda, there was no significant difference[25]. The protein content was high compared to 1.88g in mustard seeds. The low fat content of the boiled sample is interesting. It shows that the boiled sample may keep for a longer period than the raw sample. This fat content of the seed (5.65%) was higher when compared to (4.65%) fats content in *A. hybridus*leaf[11]. So in terms of fat contents the seed is more nutritious than the leaf.
The high crude fibre in the raw sample is not a surprise. This is because the raw sample is more concentrated in dry matter of which crude fibre is included. The boiled sample has absorbed water which might have reduced the crude fibre content. The crude fibre content of the seed (12.7%) contrast the quantity of crude fibre (6.3%) found in amaranth specie planted in Uganda [25]. Fibre helps in lowering serum cholesterol level, reduces risk of heart disease, constipation, cancer etc. [25]. Boiling reduced the carbohydrate content of the seed which may be due to leaching into boiling water. The carbohydrate content of the seed (48.93%) was not comparable to (63%) carbohydrate found in the specie planted in Uganda.

4.2. Vitamin Composition

Vitamin composition of *A.hybridus* seed presented in (Table 2) shows that seed contains appreciable amount of riboflavin (6.05mg/100g), which was far greater than (0.23mg/100g) in the specie planted in Uganda. But boiling lowered riboflavin content of the seed which could be attributed to the fact that it is a water soluble vitamin and might have leached out into the boiling

water. Boiling had the same effect on the thiamin content of the seed. Comparing the thiamin content (2.28mg/100g) of the seed to (0.1mg/100g) of the seed of amaranth specie planted in Uganda, they are not comparable, and there was a significant difference between the two.
The lower ascorbate of the boiled sample is expected. It is a water soluble vitamin and as such may have evaporated with steam. There was an increase in vitamin A content of the seed from (9.06R.E) to (15.6 R.E). This could be that boiling exposed the surface area of the seed and more vitamin A was released.

4.3. Mineral Composition

The mineral composition of *A.hybridus*seeds on Table 3 shows that boiling reduced the contents of the mineral elements analyzed in this seed. This could be attributed to loss of these minerals during boiling. The quantity of Iron (2.50mg/100g) in the seed is low when compared to (17.4mg/100g) found in the seed of amaranth specie planted in Uganda. This variation could be due to changes in environmental factors or the laboratory used for the analysis. The iron content (2.50mg) of *A.hybridus* seeds was higher than (0.76mg) in mustard seeds [(26)].Mustard seeds with this value was rated as a good source of iron[(27)], so *A. hybridus* could also be rated as a good source of iron. Iron is essential for normal heamoglobin formation, normal functioning of the central nervous system and oxidation of carbohydrate, protein and fat [(3)]. The zinc content (6.45mg.100g) of the seed was high compared to (3.80mg/1000g) found in the leaf, the zinc content of the seed compared well with the quantity (0.44mg) found in mustard seeds rated as a good source of zinc. Zinc is involved in normal functioning of the immune system [(11)].
The calcium content (2.42mg/100g) of the seed was very low when compared to (175mg/100g) found in the seeds of the specie planted in Uganda. Calcium is associated with growth and maintenance of bones, teeth and muscle[(28)]. There was a drastic reduction in iodine content of the seed from 9.03mg/100g to 3.94mg/100g. Iodine is necessary for normal functioning of thyroid gland.

4.4. Antinutrientcomposition

The results of antinutrient composition of the seed (Table 4) were high when compared to the quantity found in the leaf. But according to Akubor[(29)], the safe level of phytate consumption is (5.0g/100g) and oxalate (2-5g/100g). The level of phytate (5.75mg/100g) and tannin (3.98mg/100g) in the seed falls within the safe level so the seed may be safe for human consumption. These antinutrients also have health benefits, examples phytate reduce the Iron in the body before it could cause damages to the colon [(30)]. Tannin suppresses tumor growth. It is also a phenolic compound antioxidants and radical scavenging properties as well as anticarcinogenic effects [(31)].

4.5. Phytochemicalcomposition

The phytochemical compositions of the seed were high. The saponin content of the seed (23mg/100g) was far greater than (1.68mg/100g) found in the leaf [(4)]. Saponin is used in various drug preparations, controlling blood cholesterol level, bone health and building of immune system (Okwu, 2003). The alkaloid content of the seed (6.00mg) was higher than (3.54mg) content of the *A. hybridus*leaf. Alkaloid plays some metabolic roles and control development in living organisms [(7)]. The phenol content of the seed (0.89mg) was comparable to the phenol content of the leaf (0.35mg), there was no significant difference. Phenols have disease preventive effects and are used for its medicinal purposes.
The flavonoid content of the seed was reduced during boiling. The flavonoid content (2.00mg/100g) was higher than (0.8mg/100g) found in the leaf. Flavonoid helps modulate brain function, inhibit cancer cell growth and heart disease [(32)]. The carotenoid content of the leaf was reduced with boiling from (9.05mg/100g) to (7.04mg/100g). Carotenoids offers protection against some cancers, macular degeneration, cataracts and other health conditions linked to oxidative or free radical damage (Susan, 2006). This high phytochemical content of the seed suggests that is may be useful for human consumption just like the leaf.

5. Conclusion

The data of this project work proved that the seed contains appreciable amount of proteins, fat, crude fibre, vitamins, minerals and phytochemicals. The high phytochemical contents show that the seed could be useful medically and in pharmacological industries. The results of the processing method applied in this study caused a reduction in the nutrient contents of the seed even the antinutrient except tannin showing that the seed may be better used in the raw form. Furthermore, considering the protein, Iron, Zinc riboflavin and thiamin contents of the seed we could ascertain that the seed may be useful in the treatment of malnutrition in children.

6. Recommendation

Having successfully concluded this study the researcher makes the following recommendations:

- The seed may be incorporated into existing diet because of its nutrient potentials.
- Further study should be done on the method of preparation, processing and preservation of this seed.
- Chemical analysis should not be the sole criterion for assessing nutritional value of plant seed but it is necessary to consider other aspects such as biological evaluation of the nutrient content of the seed in order to determine the bioavailability of the nutrients.
- Efforts should be made to educate the public on preservation of this seed to avoid wastage.

7. References

1. Ladeji, O., Okoye, Z.S., Ojobe, T. (1995).Chemical Evaluation of the nutritive value of leaf of fluted pumpkin. (TelferiaOccidentalis). Chem. 53: 353-355.
2. Okafor, J.C. (1983). Horticultural Promising Indigenous wild plant Species of Nigerian Forest zone Acta. Hort. 123,pp165-176.
3. Wardlaw, G.M. and Kessel, M. (2002). Perspective in Nutrition (5thed) McGraw. Hill, Boston, Pp. 271-274.
4. Akubugwo, I.E.,Obasi, N.A., Chinyere, G.C. and Ugbogu, A.E. (2007).Nutritional and chemical value of Amaranths hybridus leaves from Afikpo, Nigeria.Africa Journal of Biotechnology,6(24), pp 2833-2839.
5. Akindahunsi, A.A andSalawu, S.O. (2005).Phytochemical screening and nutrient-antinutrient composition of selected tropical green vegetables.Afr. J. Biotech. 4: 497-501.
6. Lockeett, C.T., Calvert, C.C., Grivetti, L.E. (2000).Energy and Micronutrient composition of dietary and medicinal wild plants consumed during drought: study of Rural Fulani, Northeastern Nigeria.Int. J. Food sci.Nutr. 51: 195-208.
7. Edeoga, H.O., Omosun, G.,Uche L.C. (2006). Chemical composition of Hyptissauveolens and Ocimum,gratissiumfrom hybrids NigeriaAfr. J. biotechnol. 5(10): 892-895.
8. Hassan, L.G., Umar K.J. (2006).Nutritional value of Balsam. Apple (MomordicabalsaminaL.) leaves. Pak. J.Nutr. 5(6): 522-529.
9. Mepha, H.D., Eboh L., Banigho, D.E.B. (2007). Effects of processing treatment on the Nutritive composition and conscience acceptance of some Nigerian edible leafy vegetables.Afr. J. Food Agric. Nutr. Dev. 7(1): 1-18.
10. Ihekoronye, A.I. and Ngoddy, P.O. (1983).Integrated food Science and technology for the tropics. Macmillan Publishers Ltd. London, Pp 236-254.
11. Akubugwo, I.E.,Obasi, N.A., Chinyere, G.C. and Ugbogu, A.E. (2007).Nutritional and chemical value of Amaranths hybridus leaves from Afikpo, Nigeria.Africa Journal of Biotechnology,6(24), pp 2833-2839.
12. Dhellot, J.R., Matouba, E., Maloumbi, M.G., Nzikou, J.M.,Safou-Ngoma, D.G., Linder, M., Desobry S., Parmentier, M.(2006). Extraction, chemical composition and nutrition characterization of vegetable oils: Case ofAmaranthusHybridus (Var 1 and2) of Congo Brazzaville. Afr J. Biotechno 5(11): 1095-1101.
13. Martin, F.W. and Telek L. (1979).Vegetables for the hot humid part 6: Amaranth and celosia..U. S.Dept of Agric. New Orleans LA. PP 156-163.
14. Railey, R. (2008). Amaranth.A healthy Grain for vegetarian recipe.http://chel.day.com/amaranth.html.
15. Gruenwald, J.(2004).PDR for Herbal medicines. 3rd ed. MontvaleNJ: Thomson PDR.
16. Craig, W.J. (1999).Health-Promoting properties of common herbs. Am J. ClinNutr. 70: 491S-499S
17. AOAC (1995).Method of Analysis of Association of Chemist (16thed) Washington D.C 600-792.
18. Pearson, D.A (1976).The chemical Analysis of food (7thed) New York: Churchill and Livingstone.
19. OnwukaG.I. (2005).Food analysis and instrumentation theory and practice Napthali prints Lagos, Nigeria.
20. Association of vitamin chemist, Method of vitamin Assay, Third Edition,Inc, pp. 287-344.
21. Obadori, B.O. and Ochuko, P.O. (2001).Phytochemical Studies and comparative efficacy of the guide extracts of some haemostatic plants in Edo and Delta state, journal of pure and applied science page 203-208.
22. Harborne, J.B. (1973).Phytochemical methods Chapman and Hall London pp: 110-113
23. Havalampl and Kard (1997).Determination of Total plasma tocopherol in the presence of carotenes Pol. Arch Walter.45-47.
24. Udom, A.U. (2005)Essential of statistics, magnet business enterprise, Enugu, Nigeriapp 30-35.
25. John, H. M.,Nabakaya, D., Nakimbugwe, D. N. andMaisinde, D.(2008). Efforts to promote Amaranth.Production and consumption in Uganda to fight malnutrition MakereUniversity, KampalaUganda
26. Wood and Rebeccca, (1988).The Whole Food Encyclopedia.New York, NY: Prentice-Hall Press.
27. Ensminger A.H., Esminger, M.K.J.(1988). Food for Health: A Nutrition Encyclopedia. Clovis, California: Pegus Press.
28. Turan, M. Kordali S. Zergin H. Dursun A. Sezen Y. (2003). Macro and Micro- Mineral Content of some wild edible leaves consumed in Eastern Anatolia. Acta Agric. Scand. Sect. B. Plant Soil Sci. 53. 129-137.
29. Akubor, P.I. (2006). Processing, Utilization and Storage properties of locust beans (Pakiabiglobosa) fruit pulp.Deparemnt of Fd. Sc. and Tech.M.Sc.ThesisUniversity of Nigeria Nsukka.
30. James, P. (2008). Phytates.Challenges and Possibilities of inositol HexaphosphateSUITE 101.
31. Marienfeld, C. (2003). Inhibition of Cholangicarcinoma growth by tannic and Hepatology. 37(5): 1097-104.
32. Lotito,S.B.andFrei B. (2006).Consumption of flavonoid-rich foods and increased plasma antioxidant capacity in humans: cause, consequence, or epiphenomenona Free Radic, Boil. Med. 4: (2): 1727-46.