PROXIMATE ANALYSIS AND HEAVY METALS DETERMINATION OF AFRICAN SPINACH (*AMARANTHUS HYBRIDUS*) AND AFRICAN EGG PLANT (*SOLANUM MACROCARPON*) IN ILARO, OGUN STATE, NIGERIA.

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ABSTRACT

Vegetable is a vital component of human diet that should be eaten all year bound but scarce during the dry season. In this study proximate composition and heavy metals concentration in African eggplant (Solanum Macrocarpon) and African Spinach (Amaranthus Hybridus) leaves collected from a local farm in Ilaro were investigated. The results of the analysis which were based on dry matter revealed that Solanum macrocapon had a moisture content (71.42± 0.14%), ash (5.96±0.01%), Crude Lipid (2.35to ± 0.00%),Crude fibre (10.10 ± 0.02%), crude protein (18.00+0.05%)

NFE (63.59± 0.11%), and dry matter (28.58 ±0.01%).Similarly, Amaranthus hybridus had moisture content (68.30±0.11%), ash (6.60± 0.02%), crude lipid (4.75±0.03%), crude protein (16.41±0.02%), Nitrogen Free Extract (59.84 ±0.10%), and dry matter (31.77 ± 0.02%).The results above established that both leaves are rich in protein and dietary fibre which maker them useful in diet regulations and maintenance. The concentration of heavy metals (Cu, Zn, Fe, Cd) in solanum macrocarpon and Amaranthus hybridus were0.19±0.02mg/L,0.47+0.01mg/L, 2.17 ± 0.01mg/L, Not detected and 0.69 ± 0.01mg/L, 0.07 ± 0.04mg/L, 2.10 ± 0.01mg/L, Not detected, respectively.

Key words:Amaranthus hybridus,Solanum macrocarpon,Heavy metals,Proximate composition.

INTRODUCTION

Most developing countries depend on starch based foods as the main staple food for the supply of both energy and protein. These accounts in part for protein deficiency which prevaisl among the populace as recognized by food agricultural organization (Ladegi et al., 1995).

In Nigeria, as in most other tropical countries of Africa where the daily diet is domination by starchy staple foods, vegetables are the cheapest and must readily available sources of important proteins, Vitamins, Minerals and essential amino acid (Okafor, 1983).

Vegetables are very important protective food and useful for the maintenance of heat and the prevention and treatment of various diseases (D' Mello, 2003).

Heavy metals have been reported to have positive and negative roles in human life (Dundar and Saglam, 2004). Some like Cd, Pb, Hg and Ar are major contaminants of food supply and may be considered the most important problem to our environment while others like Fe, Zn, and Cu are essential for biochemical reactions in the Body (Zaidi et al., 2005).The term 'Heavy metals'is defined as commonly held for those metals which have specific weights more than $5gcm^{-3}$ and trace elements are those less than $5gcm^{-3}$olleman the wiverd, 1985).

Heavy metal pollution can originate from natural and anthropogenic sources, such as mining and smelting, operation, discharge of sewage sludge, irrigation with animal waste or water waters, unapproved usage of chemical etc. (Zantopoulos et al., 1999).

Egg plant and African Spinach are vegetables that are being consumed as food by man without considering their proximate composition and the environment in which these vegetables are being cultivated which might lead to increase in the amount of trace element and heavy metals, which are not biodegradable by the body digestion metabolism. Most often foods are consumed without considering the way they are affected by excessive intake of heavy metals which could also serve as micro element. The aim of the study is to determine the proximate composition of solanum macro-carpon and Amaranthus Hybridus and also to determine the heavy metals and trace elements present in this vegetables (Cd, Pb, Zn and Cu).

MATERIALS AND METHODS

Fresh samples of the vegetables (Amarantus Hybridus and solanum macrocarpon) were collected from a local farm in Ilaro,Yewa south LGA of Ogun State. The collected samples were placed in dark polyethylene bag and air dried is retain the active components in them. The dried samples were grounded to a fine powder, the powder of each sample was sieved and stored in an air tight cellophane bag as stock required for analysis.

METHODOLOGY

DETERMINATION OF MOISTURE CONTENT.

The method described by AOAC (1990) was adopted 2.0g of fresh sample was weighed and placed in a clean, dried and weighed crucible. This was placed in an oven, dried at 105^0C for three hours. The sample was allowed to cool in a desiccator and then re-weighed. The percentage moisture content was calculated as thus: %Moisture content = W_0 x 100

W1

Where W_0 = loss in weight (g) on drying

W_1= Initial weight of sample.

Determination of Ash content.

The ash content was done using ignition method as described by AOAC (1990).

The crucible was washed and preheated in a muffle furnace at about 500^0 and cooled in a desiccator. 1.0g of the oven dried sample was placed in the crucible and then reweighed. The crucible was covered with its lid, the number noted and then placed in a cold muffle furnace. The temperature was allowed to rise and regulated to 500^0C and the ashing was carried out for 3hours.The crucible removed from the furnace, allowed to cool in a desiccator and reweighed. The percentage ash content was calculated thus.

% Ash content = M_a =MA X 100

Where Ma=mass of ash (g)

Ms=mass f sample used (g)

Crude Protein Determination

The crude protein was done first by determining the total organic nitrogen using the micro-kjeldahl method and multiplying by 6.25 as described by AOAC (1990). This method involved digestion, distillation and titration. 1.0g of the sample was weighed and placed in digestion (Kjehdahl) flask. Few granules of anti-bump and 3.0g of copper catalyst mixture (96% anhydrous sodium sulphate, 3.5% copper sulphate and 0.5% selenium oxide) were added into the flask. Digestion commenced on sample by adding $20cm^3$ of concentrated Sulphuric acid and heating on a heating mantle or digester.

Digestion was continued until a clear solution was obtained and then the flask was allowed to cool. The digest was filtered up to $100cm^3$ with distilled water. The distillation step was done using a rapid distillation unit (Labanco). 10ml of the digest was introduced into the inlet of the distillation machine. Also 10ml of 40% NaOH solution was added to the digest in the inlet and was diluted with 10ml of distilled

water.40ml of 0.5m HCl was pipette into the receiving flask and the adaptor was made to dip into it. The temperature knob was regulated and the sample was distilled to twice the original volume of the content in the receiving flask. The distillate was titrated against 0.5M NaOH solution using methyl orange indicator until the colour changed from pink to colourless.

The titre value was noted and the percentage total organic nitrogen was calculated using the formula

%TON= Titre Value x Mass of Nitrogen x volume of digest x molarity of acid used x100

Mean of sample in mg x mass of digest distilled.

Crude protein = % TON X 6.25

Note: 6.25 is a general factor suitable for products in which the proportions of specific protein are not well defined.

Crude Lipid Determination

The determination of crude lipid content of the sample was done using soxhlet type of the direct solvent extraction method as described by AOAC (1990). The solvent used was n-hexane (boiling range 40^0C to 60^0C) 3.0g of sample was weighed and secured in a soxhlet extraction thimble. The thimble was put into 250cm^3 round bottom flask, weighed and 200cm3 of the 400C to 600C was added to it. The flask was then mounted on the heating mantle and connected to the extractor (with condenser). The condenser and the heating mantle were activated and extraction carried out for 4 hours. At the end of the extraction the solvent was evaporated. The percentage crude lipid calculated using the formula.

$$\%\text{Crude lipid} = \frac{M_{EX}}{M_s} \times 100$$

Where M_{EX} = mass of the extract (g)

M_s = Mass of the sample used (g)

Crude Fibre Determination

The crude fibre content of samples was determined using the acid-base method of AOAC (1999). The defatted sample obtained for the lipid determination was used, lg of the sample was placed in a 250cm^3 flask and weighed as W_1, 200cm^3 of 0.125M H2SO4 was added to the solution boiled. The mixture was b oiled gently for 30minutes with constant rotation of the flask every minute so as to mix the content and remove particles from the sides. The mixture was allowed to cool for some minutes and then filtered through a muslin dot stretched over 9cm Buchner funnel. The flask was rinsed with hot distilled water until acid free. The residue scrapped back into the flask with spatula and 200cm^3 of 0.125M NaOH was added and then the solution was allowed to boil for 30minutes, then the resulting solution was then filtered and thoroughly washed with hot distilled water, filtered twice with petroleum ether. Residue was drained and scrapped into the crucible, dried in the oven at 100^0C cooled in a desiccator and weighed as W_2 then transferred into the muffle furnace at 450^0C for 90 minutes to ash, cooled in a desiccator and weighed as W_3. The percentage crude was detected using the following equation

$$\% \text{ crude fibre} = \frac{W_2 - W_3}{W_1} \times 100$$

Where W_1 = weight of flask + sample

W_2 = weight of crucible + Extract

W_3 = weight of crucible + Ash.

Nitrogen Free Extract Determination

Nitrogen free extract content of each sample was estimated by differences. In this, the sum of the percentage of all the other proximate components was subtracted by 100.

NFE = 100 – (% Ash content + % crude protein + %Crude fat + crude fibre).

Dry Matter Content

Dry matter of the each sample was estimated by 'difference'. In this, the percentage moisture content was subtracted from 100.

% Dry matter = 100 -% moisture content.

Heavy Metal Determination using AAS.

Heavy metal was determined on each sample which was dried for analysis using the over drying method at 105^0C for 24hrs (AOAC, 1990).

About 1.0g of each sample were weighed and digested in a mixture containing 5ml of HCl acid, 2ml of concentrated H_2SO_4 and 20ml of concentrated HNO_3 in a conical flask under a fume hood. The content was mixed and heated gently at 180^0C-2200c for about 30mins on a digester.

The digest was allowed to cool. Make up to mark with distilled water in a 50ml volumetric flask. All reagent used were of analytical grade and heavy metals (Zn, Cd, Cu, and Pb) were determined using AAS buck VCG 210 using air acetylene mixture.

Results and Discussion

Percentage Proximate composition of Amaranthus hybridus and solanum macrocarpon on dry matter basis.

% Proximate constituent	Amaranthus Hybridus	Solanum macrocarpon
Moisture Content	68.30+-0.11%	71.42+0.14%
Ash Content	6.60+0.02%	5.96+0.01%

Crude Fibre	12.40+0.04%	10.10+0.02%
Crude Protein	16.41+0.03%	18.00+0.05%
NFE	59.84+0.10%	63.59+0.11%
Crude Lipid Dry Matter	4.75+0.00% 31.77+0.02%	2.35+0.00% 28.58+0.01%

Mean values are of triplicate determinations ± SD.

Table 2. Shows the results of the trace elements and heavy metals composition of Aamaranthus hybridus and Solanum macrocarpon on dry matter basis.

Element	Solanum macrocarpon(mg/L)	Amaranthus Hybridus(mg/L)	Safe Limit(mg/L)
Zn	0.47+0,01	0.07+0.04	5.0
Fe	2.17+0.01	2.01+0.01	60.0
Cu	0.19+0.02	0.69+0.01	0.3
Cd	ND	ND	0.003

ND=Not Detected

Discussion

The moisture content for A. hybridus and S. macrocarpon were 68.03+0.11% and 71.42+0.14% respectively which is high compared to telferia occidentalis (26.51+0.02%) and Allium sativum (61.2± 0.11%) okoli et al., 2007) which will aid the growth of micro-organisms and the storage life would be low.

The crude protein of the leaves were 16.41± 0.03% and 18.00 ± 0.05%,for A. hybridus and S. macrocarpon respectively. The values were relatively high when compared with Acalypha manginata (13.78%) and moderate when compared with Acalypha racemosa16.19% and Acalypha manginata 18.15 %(Iniaghe et al.,2009). Thus the leaves are only promising a moderate source of dietary protein.

The values of ash content were 6.60 ± 0.02% and 5.96 ± 0.01% for A. hybridus and S. macrocapon respectively. These requires investigation to ascertain the mineral element as they are essential for tissue functioning and a necessity in daily requirements for animal nutrition. The ash content however, compared favourably with leaves of vigna unguiculata (5.97%) and are lesser than leaves of Hibiscus esculentus (8.00%) and occimum graticinum (8.00%) by Akindahunsi and Salawu, 2005). The ash content is the reflection of the mineral content preserved in the food matrices. The results therefore suggest a high deposit of mineral content in the leaves (Antia et al., 2006).

The value of the crude lipid for solanum macrocarpon 2.35% and A. hybridus4.75% which is low compared to Baseila alba(8.71%)but A hybridus is compared favourably with physalis angulata (4.69%) as researched by Ifon and Basshir 1979). Dietary fat function increases in the increase of palatability of food by absorbing and retaining flavour.

The nitrogen Free Extract content was 59. 84±0.10% for A. hybridus and 63.59± 0.11% for S. macrocarpon which high compared to leguminous seed such as Soya bean (26.3%) by Temple et al., 1991 and compared favourably with phaseolus lunatus 67.0%, cajaus cajan 62.2% (Apata and Ologhobo, 1994).

The Zn content for A. hybridus and S. macrocarpon were analysed to be 0.07±0.04mg/L and 0.47±0.01mg/L respectively which is far below the WHO limit of 5mg/L.

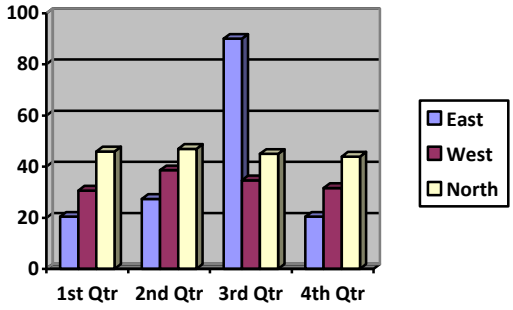
The copper content for A. hybridus and S. macrocarpon were 0.69 ±0.01mg/L and 0.19+0.02mg/L respectively which was low compared to 2.32mg/L found in bitter leaf by Ibrahim et al., 2001 and in some leafy vegetables found in cross Rivers state. Nigeria by Ifon and Basshir, 1979).

Copper is an essential micronutrient which functions as a biocatalyst, required for the body pigmentation in addition to iron, maintains healthy nervous system, prevents

anaemia and interrelated with the function of Zn and Fe in the body (Akinyele and Osibanjo, 1982). From table 2 it was observed that in the two samples cadmium was not detected. Cadmium is non-essential element in foods and natural water and it accumulates principally in the kidney and liver (Divrikli, 2006) various sources of environmental contamination has been implicated for its presence in foods.

Iron is an essential trace element for haemoglobin formation, normal functioning of the central nervous system and in the oxidation of carbohydrates, proteins and fats (Adeyeye and Otokiti, 1999). From the results, S. macrocarpon (2.17mg/L and A. hybridus (2.01mg/L) are high in iron content. Nonetheless, the amount is more than adequate as 1.00mg /day of iron is suitable for adult, human to maintain the daily balance of intake and excretion (Bothwell et al., 1989). The high presence of iron in the sample could be probably the reason for the use of this plant by the lactating mothers to regenerate lost blood.

Fig.1.Chart showing the levels of heavy metals in A.hybridus and S.macrocarpon



The chart comparing the levels of heavy metal A. hybridus and S. macrocarpon revealed that leaves of Zn and Fe were higher in S. macrocarpon while the levels of Cu in A. hybridus was higher.

Fig 2.Chart showing comparing the proximate composition of A.hybridus and S.

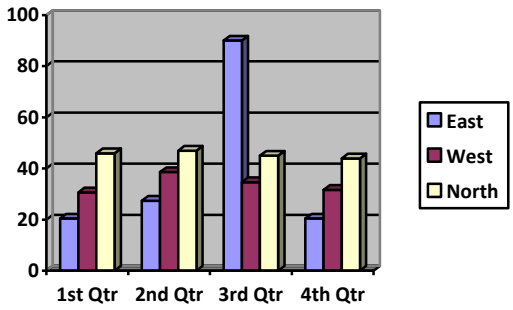


Fig 1. The chart comparing the proximate composition of A. hybridus and solanum macrocarpon revealed that the moisture content and Crude protein were higher in S. macrocarpon while crude fibre and crude fat were higher in A. hybridus. These established that both leaves are rich in protein and dietary fibre which makes them useful in diet regulation and maintenance.

Conclusion

The data from this work has confirmed that the leaves of A. hybridus and S. macrocarpon contained appreciable amount of protein, fats, fibre, carbohydrate, mineral elements, amino acids and generally low levels of toxicants. Thus, it can be concluded that A. hybridus and S. macrocarpon leaves can contributes significantly to the nutrient requirements of man and should be used as a source of nutrients to supplement other food classes.

REFERENCES

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