Screening for Minerals and Anti-minerals Composition of *Gongronema latifolium* (Utasi) Leaf

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Original Research Article

ABSTRACT

Aims: The study was conducted to determine the biochemical composition and nutritional value of *Gongronema latifolium* leaf locally cultivated in Mkpat Enin, Akwa Ibom State, Nigeria.

Study Design: The study involved sampling, identification of the plant, sample preparation, analyses and results evaluation/comparison with existing data.

This vegetable is consumed generally due to its nutritional and medicinal potentials.

Place and Duration of Study: The study was carried out in the Chemistry Laboratory of Akwa Ibom State University and Ministry of Science and Technology Laboratory, Uyo. The study was conducted for six months from July 2017 to January 2018

Methodology: Proximate analyses were done using standard analytical methods of the Association of Analytical Chemist (AOAC) 2000 edition. Micronutrients and trace metals were by spectroscopic, using atomic absorption spectrometer (ATI UNICAM, 939) Standard analytical methods. Of

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Association of Analytical Chemists (AOAC, 2000) were used for the determination of anti-oxidants: tannin, oxalate , cyanide and phytate.

Results: The results of the analyses obtained for Moisture content, crude protein, crude fibre and carbohydrate were: $60.91\pm 2.09 \%$, $22.07 \pm 0.04\%$, $4.96 \pm 0.11\%$ and $2.33 \pm 0.09\%$ respectively. Among the micronutrients determined were potassium, $471 \pm 12.08 \text{ mg/kg}$; zinc , $0.397 \pm 0.07 \text{ mg/kg}$; sodium, $143.8\pm 8.13 \text{ mg/kg}$; calcium, $130\pm 7.45 \text{ mg/kg}$; magnesium, $133\pm 5.02 \text{ mg/kg}$ and iron, $1142 \pm 14.21 \text{ mg/kg}$. The calorific value determined for the leaf was $129.3\pm10.04 \text{ J/ kg}$. The highest concentration of $471 \pm 12.08 \text{ mg/kg}$ was obtained for potassium, and the lowest mineral content was $0.397\pm 0.07 \text{ mg/kg}$ for zinc. The concentrations of anti-nutrients were generally low with phytate having the highest value of $8.24\pm \text{ mg/kg}$. Other toxicants had the following values, lead, $0.16\pm0.009 \text{ mg/kg}$; cadmium, $0.13\pm0.003 \text{ mg/kg}$; hydrogen cyanide, $0.173\pm0.043 \text{ mg/kg}$; oxalate, $0.88\pm0.056 \text{ mg/kg}$; tannins, $0.11\pm 0.005 \text{ mg/kg}$. The calorific value of the leaf was determined to be $129.3\pm10.04 \text{ J/ kg}$.

Conclusion: The results showed enhanced contents of micronutrients and protein in *Utasi* leaf, which are a health boost for consumers, mostly pregnant women and children. With the low levels of anti-nutrients and toxicants in *Gongronema latifolium* leaf, it is a potential source of food supplements and an immune booster in diets.

Keywords: Anti-nutrients; Gongronema latifolium leaf; green leafy vegetable; medicinal plant.

1. INTRODUCTION

Green leafy vegetables constitute an indispensable constituent of the human diet, especially in local delicacies. It is estimated that over sixty species of green leafy vegetables are used as food [1]. Plants exhibit important sources of active natural products, which differ widely regarding structures and biological properties [2]. In recent years, the prevention of cancer and cardiovascular diseases has been associated with the ingestion of spices, fresh fruits, vegetables or teas rich in natural anti-oxidants [3]. Plants provide raw materials for body buildings, manufacture of biofuels, dyes, perfumes, pesticides, absorbents, treatment of diseases, and also serve as valuable starting materials for drug development[4]. Gongronema latifolium is a herbivorous, non-woody plant from the family of Asclepiadaceae. It is a leafy green vegetable that has been widely accepted as a dietary constituent and medicinal plant among peasants in Nigeria and it is more popular in the Southeastern States of Nigeria. In South-Eastern and South -Western Nigeria. Gongronema latifolium is commonly called utazi and arokeke respectively [5], [6].

Leaves of this plant belong to the class of medicinal plants beneficiary for prevention and treatment of certain diseases and ailments that are detrimental to human health. It can be chewed, infused or used for cooking and mainly used in Western part of Africa for nutritional and medicinal purposes [7]. The most important phytochemicals of these plants are an alkaloid,

flavonoid, tannins and phenolic compounds[8]. The phytochemicals are responsible for the colour, flavour, smell, and texture of the plant and they also work to affect anti-oxidant activity, hormonal action, stimulation of enzymes and antibacterial effect among others [9].

This plant also acts as spices, the story of spices and other flavourings materials is one of the most interesting in the history of vegetable products [10]. This enables the plant to be therapeutically useful in the management of convulsion, leprosy, stomachache, inflammation and/or rheumatoid pains, cough and loss of appetite [11], [12].

Utasi leaf is used in many different ways in different places, as spices and vegetable for preparation of delicacies in homes and as a medicinal plant in traditional folk medicine. It can be consumed fresh, cooked or dried and applied as powdery spices. Whichever ways it carries a moderately bitter taste that contributes tremendously to its flavour. *Gongronema latifolium* leaf contains nutrients such as Potassium (K), Calcium (Ca), Iron (Fe), Phosphorus(P), Sodium (Na), Magnesium (Mg), etc. and trace amounts of anti-nutrients such as oxalate, proteinase inhibitor, phytates, tannins, alkaloids, steroids and cyanogenic glycosides [13].

It is a climber with woody hollow glamorous stems below and characterized by greenishyellow flowers. It occurs in deciduous and secondary forests, and also in the destined roadside forest [14]. *Gongronema latifolium* is commonly known by the Ikales of Ondo State of Nigeria as *Iteji* [15]. The Igbos called it *Utazi*, the Efik/Ibibio called it *Utasi* while Yoruba called it *Arokeke* or *Madumaw* [16].. But the common name for the plant is amaranth globe while the English name is bushbuck.

Plant bark contains much latex and has been used in folk medicine for maintaining healthy blood glucose levels. Leaves have been found to be very efficacious as an anti-diarrhoea [17]. Gongronema latifolium is believed to carry powerful medicinal qualities used for amelioration of malaria, diabetes, hypertension among others. It has been reported to inhibit α-glucosidase in experimental animals induced with diabetes [18]. Leaf extracts of this plant were reported by Iwaala et al.[19], to exhibit strong inhibitory activity on human lung carcinoma and human breast adenocarcinoma. Leaf extracts also exhibited free radical scavenging activity against 1, 1- diphenyl - 2 - picrylhydrazyl (DPPH) [20], [21].. Elevinmi [8] reported that the methanolic extract of the plant leaf showed inhibitory activity Salmonella enteritidis, Salmonella against cholerasius. Sertyphimunium, Pseudomonas acruginosa and Listeria monocytogenes while the aqueous extract showed inhibitory activity against E. coli and P. aeruginosa. Edim et al.[16] gave a reviewed reports on inhibitory effects of Gongronema latifolium plant extracts on Staphylococcus aureus. The inhibitory action of essential oil from Utasi leaf on bacteria isolated from HIV patients in Lagos, Nigeria has been reported [22]. The inhibitory effects were comparable to those of Ampicillin but less than those of ciproflaxacin and chloramphenicol reported in the study [22].

Oral administration of aqueous and ethanolic leaf Gongronema latifolium extracts of to streptozotocin-induced diabetic rats significantly raised the activity of superoxide dismutase, glutathione reductase, glutathione peroxidase and glucose - phosphate dehydrogenase (G6PD) thereby acting as antidiabetic agent [5]; [6]. Also, Sylvester et al.[23] observed a decrease in the blood glucose of streptozotocin-induced diabetes mellitus rats by 66.34% when treated with Gongronema latifolium leaf extracts.

Gongronema latifolium leaf is used by Ikales/Efiks in Nigeria and other West African countries to treat malaria, nausea, diabetes, hypertension, constipation, cough, intestinal worms, dysentery, dyspepsia and anorexia [24]. Although the cultivation and consumption of *Utasi* leaf have been widespread in Nigeria, the mineral and anti-nutrient composition of the leaf has not been thoroughly investigated. This study was therefore conducted to provide more information to consumers, regarding the biochemical composition of *Gongronema latifolium* leaf

2. MATERIALS AND METHODS

2.1 Plant Collection and Preparation

Fresh plant materials of *Gongronma latifolium* were collected in June 2017 from the farm at Ekim Town, Mkpat Enin Local Government Area of Akwa Ibom State, Nigeria. The Department of Botany, Akwa Ibom State University, Ikot Akpaden, authenticated the species. A voucher specimen was prepared and deposited in the herbarium of the Department of Botany. The plant material was allowed to air-dry at ambient temperature and then milled. The powdered sample was stored in an airtight plastic container for subsequent analysis.

All the reagents used for the analyses were of analytical grade (Analar), and deionised water was used for the preparation of solutions of reagents.

2.2 Proximate Analyses

Recommended methods of the Association of Official Analytical Chemists AOAC, 2000 [25] were used for the determination of moisture, ash, crude lipid, crude fibre, carbohydrate and crude protein content.

2.3 Minerals and anti-nutrients Analysis

The elements comprising of sodium, potassium, magnesium, iron, zinc, lead and cadmium were determined based on the method described in AOAC, 2000 [25], using atomic absorption spectrophotometer (UNICAM 939). While the anti-nutrients oxalate, tannins, phytate and cyanide were determined following the procedure described by Onwuka [26].

3. RESULTS AND DISCUSSION

3.1 Proximate Analysis

Proximate analysis of the plant determined in the study was; moisture content, ash (mineral), crude fibre, crude fat (lipid), crude protein, calorific value and carbohydrate. The results of the proximate composition are presented in Table 1.

Table 1. The results of proximate composition

Proximate composition	Utasi leaf
Moisture content (%)	60.91± 2.09
Ash content (%)	3.16± 0.042
Crude Protein (%)	22.07± 3.07
Crude Fat (%)	3.57± 0.15
Crude Fiber (%)	4.96± 0.11
Carbohydrate (%)	2.33± 0.09
Calorific Value (J/Kg)	129.3± 10.04

The moisture content was obtained as 60.91 %. Moisture content is significantly used for the determination of the stability and quality of foods. Materials with less moisture content stay longer than those with high moisture content [26]; [27]. The determination of moisture content in food samples is most important and most widely used measurements in the processing and testing of foods since the amount of dry matter in a food is inversely related to the amount of moisture content obtained in this study compares with the literature report for leafy medicinal plants such as *Solanum nigrum* (68.0 \pm 3.0 %) *Leonotis leonorus* (58.9 \pm 1.7 %) [28].

The ash content and crude fibre were determined as 3.16 % and 4.96 % respectively. Eleyinmi et al. [11], determined the ash and crude fibre content of this plant to be 11.6 % and 10.8 % respectively, while a value of 19.81 % ash content was reported elsewhere [29]. The low values of ash content and crude fibre obtained in this study compared to other reported values could be attributed to geographical location and maturity level of the plant.

The protein content of *Utasi* was determined as 22.07 %. This value is relatively high compared to literature value of 0.67 % [2], however, the value for protein determined in this study was close to the value of 27.2 % reported by Eleyinmi [8].

Crude proteins are all the proteins that can be found in a plant or sample. Protein encountered in living organisms has diverse functions such as catalysis, structure and defence. They are also enzymes that direct and accelerate biochemical reactions, provide structural support and serve as a reserve of essential nutrients [30]. The protein content of foods varies from 0.2 - 80 g per 100 g, but all foods do contain some protein as their building materials [31]. Roots and tubers are estimated to contain 8% of the protein [30]. Comparatively, this plant has far higher protein content than most other crops reported. Carbohydrate content of this plant was determined as 2.33 %. Carbohydrates provide energy to the body, particularly through glucose, a simple sugar that is found in many basic foods. vegetables and fruits contain some All carbohydrates [32]. Carbohydrates contain soluble and insoluble elements; the insoluble part is known as fibre, which promotes regular bowel movement, regulate the rate of consumption of blood glucose, and also helps to remove excess cholesterol from the body. In addition. carbohydrate-containing foods are vehicles for important micronutrients and phytochemicals. Unlike fat and protein, a high level of dietary carbohydrates provided it is not obtained from a variety of sources, is not associated with adverse health effects. Also, diets high in carbohydrate as compared to those high in fat reduce the likelihood of developing obesity and its co-morbid conditions [33]. The result obtained from this work is within the range of 3.92±0.23 % reported in the literature [34]. The calorific value of the leaf has a value of 129.3 ± 10.04 J/kg, which could be a good source of energy in delicacies.

3.2 Micronutrients

The results of the mineral contents of *G*. *latifolium* are presented in **Table 2**. Potassium, sodium and calcium content were 471.3 ± 12.08 mg/kg, 143.8 ± 8.13 mg/kg, and 130 ± 7.45 mg/kg respectively, while magnesium, zinc and iron content were 133 ± 5.02 mg/kg, 0.397 ± 0.07 mg/kg and $1,142 \pm 14.21$ mg/kg respectively. Enhanced levels of potassium, calcium and magnesium in *Gongronema latifolium* leaf are comparable to values reported by Offor *et al.* [35]

Micronutrients play a crucial role in human nutrition, including the prevention and treatment of various diseases and conditions as well as the optimisation of physical and mental functioning. They are critical for anyone seeking to maintain or improve his or her health. Food containing many micronutrients are considered nutrient dense. Minerals are important in human nutrition. It is well known that enzymatic activities, as well as electrolyte balance of the blood fluid, are related to the adequacy of Na, K, Mg and Zn.

3.3 Anti-nutrients

The contribution of fruits, seeds and vegetables of some plants in Nigeria to minerals, vitamins and amino acids in human nutrition is limited due to the presence of anti-nutrients which render some of the nutrients and protein unavailable for human nutrition [27]. The most common antinutritional factors in fruits and vegetables are oxalic acid, tannins, phytic acid and hydrocyanic acid [36]. The result of anti-nutrients determined in *Utasi* leaf is presented in **Table 3**.

Table 2. Mineral contents of Gongronema latifolium leaf

Mineral composition	Concentration (mg/kg)
Potassium, K	471.3± 12.08
Sodium, Na	143.8± 8.13
Calcium, Ca	130 ± 7.45
Magnesium, Mg	133.1± 5.02
Zinc, Zn	0.397.3± 0.07
Iron, Fe	1,142± 14.21

Table 3. Anti-nutrients and toxic metals level in Utasi leaf

Anti-nutrient	Content (mg/kg)
Oxalate	0.88± 0.02
Tannins	0.11± 0.005
Hydrogen Cyanide	0.173± 0.043
Phytate	8.24± 0.056
Lead Pb	0.006 ± 0.001
Cadmium Cd	0.002 ± 0.001

The oxalate content of Utasi leaf obtained in this study is 0.88 ± 0.02 mg/kg. Oxalate is a naturally occurring molecule found in abundance in plants and humans. It is not a required nutrient in human nutrition, and too much of it can lead to kidney stones. In plants, oxalate helps to get rid of extra calcium by binding with it [29]. That is why so many high oxalate foods are from plants. In humans, it may work as "probiotic" feeding good bacteria in the gut. Some of the antinutritional and off-flavour problem (bitter and astringency taste and scratches in the mouth and throat) associated with cocovam are caused by calcium oxalate [29]. Dietary oxalate has been known to complex with calcium, magnesium and iron leading to the formation of insoluble oxalate salts and resulting in oxalate stone [37].

The tannins level is 0.11±0.005 mg/kg, as presented in Table 3. The presence of tannins could be partly responsible for the bitter taste associated with the raw inflorescence and its use in treating wounds [1]. Tannins are present in plants as phenolic compounds that are soluble in water and have a molecular weight between 500 and 3000 Daltons. Tannins inhibit the activities of some enzymes such as trypsin, chymotrypsin, amylase and lipase [38]. It also interferes with

dietary iron absorption [39]. Tannins cause browning or other pigmentation problems on both fresh food and processed products [26].

Hydrogen cyanide content of the plant leaf was 0.173±0.043 mg/kg as presented in **Table 3**. Hydrocyanic acid does not occur free but combines with sugars to form a non-toxic compound known as cyanogenic glycoside [26]. A lot of hydrocyanic acids is lost during soaking and cooking so that its content in the vegetables poses no danger of toxicity [27]. Cyanide is produced in the human body and exhaled in extremely low concentrations with each breath. It is also produced by over 1000 plant species including sorghum, bamboo and cassava. The relatively low concentration of cyanide can be highly toxic to man and wildlife.

The phytate content of *Utasi* leaf was 8.24 ± 0.056 mg/kg. The value of phytate content obtained in this study is low compared to 127.82 mg/100g reported elsewhere [8]. This low phytate content makes the plant save for human consumption without attendant health problems.

Toxicants such as lead and cadmium were indicated and had values of 0.006 ± 0.001 mg/kg and 0.003 ± 0.001 mg/kg respectively (**Table 3**).

Lead content obtained in this study which was $0.006 \pm 0.001 \text{ mg/ kg}$, at trace level and lower than that reported elsewhere [40], as. lead has gained considerable attention as a toxic pollutant of concern, partly because it has been prominent in the debate concerning the growth of anthropogenic pressure on the environment [41]. Lead safe-level of 5 ug/dl, was thought to be a safe level, may be associated with decreased intelligence in children behavioural difficulties and learning problems [42].

Cadmium content obtained in this research was 0.003 ± 0.001 mg/kg at trace level, while a cadmium level of 0.35 ug/100g was reported elsewhere [43]. Cadmium is a toxic metal that occurs naturally in the environment. Humans are exposed to cadmium mostly through the plant-derived food. There is no safe margin of cadmium exposure and the need to lower human exposure is desperate [42]. The cadmium metal produces number one health problems and is a known carcinogen. Cadmium is of no use to the human body and is toxic even at low levels of exposure. The negative effects of cadmium on the body are numerous and can impact nearly all systems in the body including cardiovascular,

reproductive, the kidneys, eyes and even the brain. It affects blood pressure, prostate function and testosterone levels [44]. It induces bone damage (Itai–Itai) [12]. Exposure to cadmium can affect renal and dopaminergic systems in children [44].

4. CONCLUSION

Grongronema latifolium leaf has been recognised to share a basic two principal capacities as a popular vegetable and as a medicinal plant. It has been thoroughly investigated to identify pharmacologically active principles which form the basis for its medicinal values. This vegetable could make a significant nutritional contribution to the diet of the populace because of its high nutrient and phytochemical contents.

From the foregoing, it could be concluded that the plant has a high concentration of nutritionally important minerals and vitamins and as such its use in diet formulation. It also contains substances that are of great pharmacological and biochemical values. The consumption of this plant should be increased imperatively for all in both rural and urban setting due to its nutritional and medicinal potentials. The levels of toxicants like lead (Pb) and cadmium (Cd) determined in the leaf of Grongronema latifolium are at levels that do not raise safety concern to the potential consumers. The leaf can be pulverised and added to children's meals who often do not consume vegetables as adults, as this plant is highly enriched in calcium, iron, potassium, sodium, zinc and calcium from the results of this study and other related studies. More so, the plant under study is rich in protein and highly recommended for consumption for pregnant women, children and as an immune booster in all humans.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

 Okafor JC. Conservation and Use of Traditional vegetables from woody forest species in Sotheastern Nigeria: In the International Plant Genetics Resources Institute Workshop on Genetics Resources of Traditional Vegetables in Africa Conservation and Use. ICRAF- HQ; Nairobi, Kenya.1995

- Uhegbu FO, Emeka EI, Kanu I. Studies on the chemical and anti-nutritional content of some Nigerian spices. *Int. J. Nut. Meta*; 2011; 3(6): 72 – 76.
- Virgili F, Scaccini C., Packer L. Rimbach G. Cardiovascular disease and nutritional phenolics. In: Pokomy, J., Yanishlleva, N., Cordon, M. (Eds). *Antioxidants in Food*. Wood Head Publishing Ltd. Cambridge. 2001; 87 – 99.
- 4. Ajibesin KK. *Dacryodes edulis*: A review on its medicinal, phytochemical and economical properties. *Res. J. Med. Pla.* 2011; 5: 32-41.
- 5. Uqochukwu NH Babady ME. Antihyperglycemic effect of aqueous and ethanolic extracts and Gongronema latifolium leaves on glucose and glycogen metabolism in livers of normal and streptozotcinic induced diabetic rats. Lif. Sci.. 2002; 73(15): 1925-1938.
- 6. Ugochukwu NH, Babady ME. Antioxidant effects of *Gongronema latifolium* in hepatocytes of rat models of non-insulin dependent diabetes mellitus. *Fitoterapia*.2003; 73(7-8): 612-618.
- Apori SO, Long RJ, Castro FB, Ørskov ER. Chemical composition and nutritive value of leaves and stems of tropical weed Chromolaena odorata. Gra. For. Sci 2000; 55(1):77–81.
- 8. Eleyinmi AF. Chemical composition and antimicrobial activity of *Gongronema latifolium. J. Zhejiang Univ. Sci. B. 2007; 8* (5): 352–358.
- Corlett JL, Clegg MS, Keen CL, Grivetti LE. Mineral content of culinary and medicinal plants cultivated by Hmong refugees living in Sacramento, California. *Int J Food. Sci Nutr.* 2002; 53(2):117–128.]
- 10. Obadoni BO,Ochuko PO. Phytochemical studies and comparative efficacy of the crude extracts of some homeostatic plants in Edo and Delta States of Nigeria. *Glo.J. P. Appl. Sci.* 2002; 8: 203-208.
- Eleyinmi AF, Bressler DC, Amoo IA, Sporns P, Oshodi AA . Chemical composition of bitter cola (Garcinia kola) seed and hull. *Polish J Food Nutr Sci.2006:* 15/56(4):27–32.
- Ciobanu L, Olivier R, Lynn U, Bechir J, Denis LB. Effects of Anesthetics Agents on Brain Blood Oxygenation Level Revealed with Ultra –High Field MRI. https//doi.org/10.1371/Journal. 2012; Pone.0032645.

- 13. Ahamefule FO, Obua BE, Ibeawuchi JA, Udosen NR. The nutritive value of some plants browsed by cattle in Umudike, Southeastern Nigeria. Pakistan .1 Nutr. ;5(5):404-409.
- 14. Chattopadhyah R.R. A comparative evaluation of some blood sugar lowering agents of plant origin. J. Ethnopharmaco 1999; 67: 367-372.
- 15. Morebise O, Fafunso M.A., Makinde JM, Olajide OA. Evaluation of the bioactivity of Gongronema latifolium leaf extract in rodents. Sci. Foc. 2006; 11(1): 27-30.
- Edim EH., Egomi UG., Ekpo UF 16. Archibong EU. A review on Gongronema latifolium (Utasi): A novel antibiotic against Staphylococcus aureus related infections. Int. J. Biochem. Biotech. 2012; 1(8): 204-208.
- 17. Sofowara EA. Medicinal plants and traditional medicine in Africa. Spectrum Books Ltd, Ibadan, Nigeria. 1982.
- Nnodim J, Emejulu A, Ihim A, Udujih HI. 18. Influence of Gongronema latifolium on some biochemical parameters in alloxan induced diabetes. IJAPBS. 2012; 1(1): 13 - 17.
- 19. Iweala EEJ, Liu F, Cheng R, Li Y. Anticancer and free radical scavenging activity of some Nigerian food plants in Vitro. Int. J. Can. Res.2015; 11(1): 41 – 51.
- Sun J, Chu YF, Wu X, Liu RH. Antioxidant 20. and Antiproliferative Activities of Common Fruits. J Agric Food Chem. 2002; 50: 7449 - 7454.
- 21. Liu RH. Health Benefits of Fruits and Vegetables are from Additive and Synergistic Combinations of Phytochemicals. Am J Clin Nutr. 2003; 78: 517 - 520.
- Adeleve IA, Omadime ME, Daniels FV . 22. Antimicrobial activity of essential oil and extracts of Gongronema latifolium on bacterial isolates from bloodstream of HIV infected patients. J Pharmacol Toxicol. 2011 ; 6(3):312 - 320.
- 23. Sylvester EG, Israel EU, Olajumoke AD. The effect of Gongronema latifolium leaf extract on blood biochemical assay in diabetic rats. J. Sci Res. Rep. 2015; 6(7):514 - 522.
- Edet EE, Akpanabiatu, M.I, Uboh, F E., 24. Edet, T.E., Eno, A.E., and Itam, E.H. Gongronema latifolium crude leaf extract reverses alterations in haematological indices and weight-loss in diabetic rats. J. Pharmacol Toxico. 2011; 6 (2):174-181.

- 25. Association of Official Analytical Chemist (AOAC). Official Methods of Analysis of AOAC International. 18th Edition. Maryland, USA. AOAC International 2005.
- Onwuka G I. Food Analysis and 26. Instrumentation - Theory and Practice. Naphthali Prints, Surulere, Lagos, Nigeria. 2005: 210.
- 27. Isaac IO, Ekpa OD. Minerals and antinutrients in two varieties of African pear (Dacryodes edulis). J. Food Techn. 2009; 7(4): 106 - 110.
- Jimoh FO, Adedapo AA, Afolayan AJ. 28. Comparison of the nutritional value and biological activities of the acetone, methanol and water extracts of the leaves of Solanum nigrum and Leonotis leonorus. F. Chem. Toxicol.2010; 48: 964 - 971.
- 29. Iwuoha C I, Kalu F A. Calcium oxalate and Physiochemical Properties of Cocovam (colocasia esculenta and xanthosoma sagittifolium) Tuber flours as affected by Processing . F. Chem. 1995 ; 54.61-66
- 30. Schneider C, Rotscheidt K, Breitmaier E. A pregnane glycosides from new Gongronema latifolium (Asckepiadaceae) Liebias Annalen Der Chemie. 1993:10:1057-1062.
- Ross I A. Medicinal Plants of the World. 31. Chemical Constituents, Traditional and Modern Uses. Totowa NJ 07512: Humane Press; 2001.Vol.2: p. 487.
- 32. Morebise O, Fafunso MA. Antimicrobial and phytotoxic activities of saponin extracts from two Nigerian edible medicinal plants. Biokemistri. 1998; 8(2): 69-77.
- World Health Organization :. Energy and 33. Protein Requirements.Report of a Joint FAO/WHO/UNU Expert Consultation. WHO Technical Report Series, No. 724.
- 34. WHO World Health Report (1998). Life in the 21st Century. A vision for .all.1998.
- 35. Offor CE, Agidi JU, Egwu CO, Ezeani N, Okechukwu PCU. Vitamin and Mineral Contents of Gongronema latifolium Leaves. W. J. Med. Sci.2015; 12 (2): 189-191.
- Akwaowo EU, Ndon BA, Etuk EU. 36. Minerals and antinutrients in fluted pumpkin (Telfaria occidentalis Hook F.). F. Chem.2000; 70: 235 - 240.
- 37. Valko M, Leibfritz D, Moncol J, Cronin MTD , Mazur M, Telser J. Free radicals and anti-oxidants in normal physiological functions and human disease . Int. J Biochem. Cell Biol.2007; 39 (1), 44 - 84 38.
 - Hernandaz N E, Tereschuk N L, Abdala L

R. Anti-microbial activity of Flavonoids in Medicinal Plants from Tafidel Valle (Tucumon Argentina). J. Ethnopharmacol. 2000; 73 (1-2), 317 – 322.

- Ogundipe OO, Moody JO, Akinyemi TOL, Raman A. Hypoglycemic Potentials of Methanolic extracts of selected Plants Food in Alloxanized Mice. Plant Foods Hum. Nutri.2003; 58 (3), 1 – 7.
- 40. Ajayi IA, Oderinde RA, Kajogbola DO, Ukponi JU. Oil content and fatty acid composition of some underutilized legumes from Nigeria. Food Chem. 2006; 99(1):115–120.
- 41. Aletor MVA. Adeogun OA. Nutrient and Anti- nutrient Components of Some

Tropical Leafy Vegetables. F. Chem. 1995; 53,375-379.

- 42. WHO World Health Statistics. Monitoring health for the Sustainable Development Goals targets with thirty-five indicators, as well as data on life expectancy. 2017
- 43. Glew RS, VanderJagt DJ, Huang YS, Chuang LT, Bosse R, Glew RH. Nutritional analysis of the edible pit of Sclerocarya birrea in the Republic of Niger (daniya, Hausa). *J Food Comp Anal.2004;* 17(1):99–111.
- 44. Pizent A, Tarila B, Zivkovic T. Reproductive toxicity of metals in MenArhHigRada Toksikol Suppl. 2012; 63 (1): 35 - 46