Screening for Minerals and Anti-minerals Composition of

Gongronema latifolium (Utasi) Leaf

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7 ABSTRACT

8 **Aims:** The study was conducted to determine the biochemical composition and nutritional value of 9 *Gongronema latifolium* leaf locally cultivated in Mkpat Enin, Akwa Ibom State, Nigeria.

10 **Study design:** The study involved sampling, identification of the plant, sample preparation , analyses

11 and results evaluation/comparison with existing data.

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

21 Results: The results of the analyses obtained for Moisture content, crude protein, crude fiber and 22 carbohydrate were: 60.91± 2.09 %, 22.07 ± 0.04%, 4.96 ± 0.11% and 2.33 ± 0.09% respectively. 23 Among the micronutrients determined were potassium, $471 \pm 12.08 \text{ mg/kg}$; zinc, $0.397 \pm 0.07 \text{ mg/kg}$; 24 sodium, 143.8 ± 8.13 mg/kg; calcium, 130 ± 7.45 mg/kg; magnesium, 133 ± 5.02 mg/kg and iron, $1142 \pm$ 25 14.21 mg/kg. The calorific value determined for the leaf was 129.3±10.04 J/kg. The highest 26 concentration of 471 ±12.08 mg/kg was obtained for potassium, and the lowest mineral content was 27 0.397± 0.07 mg/kg for zinc. The concentrations of anti-nutrients were generally low with phytate having 28 the highest value of 8.24± mg/kg. Other toxicants had the following values, lead, 0.16±0.009mg/kg; 29 cadmium, 0.13±0.003mg/kg; hydrogen cyanide, 0.173±0.043 mg/kg; oxalate, 0.88 ±0.056 mg/kg; tannins, 0.11± 0.005 mg/kg. The calorific value of the leaf was determined to be 129.3± 10.04 J/kg . 30 31 Conclusion: The results showed enhanced contents of micronutrients and protein in Utasi leaf, which 32 are a health boast for consumers, mostly pregnant women and children. With the low levels of 33 anti-nutrients and toxicants in Gongronema latifolium leaf, it is a potential source of food supplements 34 and an immune boaster in diets.

35 Keywords : Anti-nutrients, Gongronema latifolium leaf, Green leafy vegetable, Medicinal plant,

36 INTRODUCTION

Green leafy vegetables constitute an indispensable constituent of 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 in terms of structures and 40 biological properties [2]. In recent years, the prevention of cancer and cardiovascular diseases has 41 been associated with the ingestion of spices, fresh fruits, vegetables or teas rich in natural 42 anti-oxidants [3]. Plants provide raw materials for body buildings, manufacture of biofuels, dyes, 43 perfumes, pesticides, absorbents, treatment of diseases, and also serve as valuable starting materials 44 for drug development[4]. Gongronema latifolium is an herbivorous, non-woody plant from the family of 45 Asclepiadaeceae. It is a leafy green vegetable that has been widely accepted as a dietary constituent 46 and medicinal plant among peasants in Nigeria and it is more popular in Southeastern States of Nigeria. 47 In South- Eastern and South -Western Nigeria, Gongronema latifolium is commonly called utazi and 48 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 alkaloid, flavonoid, tannins and phenolic compounds[8]. The phytochemicals are responsible for the colour, flavor, 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 act 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 medicinal plant in traditional folk medicine. It can be consumed fresh, cooked or dried and applied as powdery spices. Whichever ways it carries a moderate 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 greenish yellow flowers.
It occurs in deciduous and secondary forests, and also in 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 bush buck.

72 Plant bark contains much latex and has been used in fork medicine for maintaining healthy blood 73 glucose levels. Leaves have been found to be very efficacious as an anti-diarrhea [17]. Gongronema 74 latifolium is believed to carry powerful medicinal gualities used for amelioration of malaria, diabetes, 75 hypertension among others. It has been reported to inhibit α -glucosidase in experimental animals 76 induced with diabetes [18]. Leaf extracts of this plant were reported by Iwaala et al. [19], to exhibit 77 strong inhibitory activity on human lung carcinoma and human breast adenocarcinoma. Leaf extracts 78 also exhibited free radical scavenging activity against 1, 1- diphenyl - 2 - picrylhydrazyl (DPPH) [20], 79 [21]. Elevinmi [8] reported that the methanolic extract of the plant leaf showed inhibitory activity 80 against Salmonella enteritidis, Salmonella cholerasius. Sertyphimunium, Pseudomonas acruginosa 81 and Listeria monocytogenes while the aqueous extract showed inhibitory activity against E. coli and P. 82 aeruginosa. Edim et al. [16] gave a reviewed reports on inhibitory effects of Gongronema latifolium 83 plant extracts on Staphylococcus aureus. The inhibitory action of essential oil from Utasi leaf on 84 bacteria isolated from HIV patients in Lagos, Nigeria has been reported [22]. The inhibitory effects 85 were comparable to those of Ampicillin but less than those of ciproflaxacin and chloramphenicol 86 reported in the study [22].

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

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100 2. MATERIALS AND METHODS

101 **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 deionized water was used

109 for preparation of solutions of reagents.

110 **2.2 Proximate Analyses:**

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

113 **2.3 Minerals and anti-nutrients analysis**

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

118 3. RESULTS AND DISCUSSION

119 **3.1 Proximate Analysis**

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

123 Table 1: The results of proximate composition

Proximate composition	<i>Utasi</i> 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

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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 mostly 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 it contains [11]. The value of moisture content obtained in this study compares with the literature report for leafy medicinal plants such as *Solanum nigrum* (68.0 ± 3.0 %) *Leonotis leonorus* (58.9 ±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 Elevinmi [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 defense. They are also enzymes that direct and accelerate biochemical reactions, provide structural support and serve as 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 protein [30]. Comparatively, this plant has far higher protein content than most other crops reported.

146 Carbohydrate content of this plant was determined as 2.33 %. Carbohydrates provide energy to the

147 body, particularly through glucose, a simple sugar that is found in many basic foods. All vegetables and 148 fruits contain some carbohydrates [32]. Carbohydrates contains soluble and insoluble elements; the 149 insoluble part is known as fiber, which promote regular bowel movement, regulate the rate of 150 consumption of blood glucose, and also helps to remove excess cholesterol from the body. In addition, 151 carbohydrate containing foods are vehicles for important micronutrients and photochemicals. . Unlike 152 fat and protein, a high level of dietary carbohydrates provided it is not obtained from a variety of 153 sources, is not associated with adverse health effects. Also, diets high in carbohydrate as compared to 154 those high in fat reduce the likelihood of developing obesity and its co-morbid conditions [33]. The 155 result obtained from this work is within the range of 3.92±0.23 % reported in the literature [34]. The 156 calorific value of the leaf has value of 129.3 ± 10.04 J/kg, which could be a good source of energy in 157 delicacies.

158 **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 \pm 12.08 mg/kg, 143.8 \pm 8.13 mg/kg, and 130 \pm 7.45 mg/kg respectively, while magnesium, zinc and iron content were 133 \pm 5.02 mg/kg, 0.397 \pm 0.07mg/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 crucial role in human nutrition, including the prevention and treatment of various diseases and conditions as well as the optimization 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 adequacy of Na, K, Mg and Zn.

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	

170 **Table 2:** Mineral contents of *Gongronema latifolium* leaf

Zinc, Zn	0.397.3± 0.07
Iron, Fe	1,142± 14.21

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172 **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 anti-nutritional 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**.

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	

178 **Table 3:** Anti-nutrients and toxic metals level in *Utasi* leaf

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180 The oxalate content of Utasi leaf obtained in this study is 0.88 ± 0.02 mg/kg. Oxalate is a naturally 181 occurring molecule found in abundance in plants and humans. It is not a required nutrient in human 182 nutrition, and too much of it can lead to kidney stones. In plants, oxalate helps to get rid of extra 183 calcium by binding with it [29]. That is why so many high oxalate foods are from plants. In humans, it 184 may work as "probiotic" feeding good bacteria in the gut. Some of the anti-nutritional and off-flavour 185 problem (bitter and astringency taste and scratches in the mouth and throat) associated with cocoyam 186 are caused by calcium oxalate [29]. Dietary oxalate has been known to complex with calcium, 187 magnesium and iron leading to the formation of insoluble oxalate salts and resulting in oxalate stone 188 [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, chymotropsin, 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 acid 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. 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].

213 Cadmium content obtained in this research was 0.003 ± 0.001 mg/kg, at trace level, whilst a cadmium 214 level of 0.35 ug/100g was reported elsewhere [43]. Cadmium is a toxic metal that occurs naturally in 215 the environment. Humans are exposed to cadmium mostly through plant-derived food. There is no 216 safe margin of cadmium exposure and the need to lower human exposure is desperate [42]. The 217 cadmium metal produces number one health problems and is a known carcinogen. Cadmium is of no 218 use to the human body and is toxic even at low levels of exposure. The negative effects of cadmium on 219 the body are numerous and can impact nearly all systems in the body including cardiovascular, 220 reproductive, the kidneys, eyes and even the brain. It affects blood pressure, prostrate function and

testosterone levels [44]. It induces bone damage (Itai–Itai) [12]. Exposure to cadmium can affect renal
 and dopaniunergic systems in children [44].

4. CONCLUSION

Grongronema latifolium leaf has been recognized 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 significant nutritional contribution to the diet of the populace because of its high nutrient and phytochemical contents.

229 From the foregoing, it could be concluded that the plant has a high concentration of nutritional 230 important minerals and vitamins and as such its use in diet formulation. It also contains substances that 231 are of great pharmacological and biochemical values. The consumption of this plant should be 232 increased imperatively for all in both rural and urban setting due to its nutritional and medicinal 233 potentials. The levels of toxicants like lead (Pb) and cadmium (Cd) determined in the leaf of 234 Grongronema latifolium are at levels that do not raise safety concern to the potential consumers. The 235 leaf can be pulverized and added to children's meals who often do not consume vegetables as adults, 236 as this plant is highly enriched in calcium, iron, potassium, sodium, zinc and calcium from the results of 237 this study and other related studies. More so, the plant under study is rich in protein, and highly 238 recommended for consumption for pregnant women, children and as an immune boaster in all 239 humans.

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