

**COMPARATIVE PROXIMATE, ANTIOXIDANT VITAMINS AND MINERAL COMPOSITION OF FOUR SELECTED TROPICAL NUTRITIONAL/ MEDICINAL PLANTS NAMELY: *Ocimum gratissimum*, *Piper guineense*, *Gongronema latifolium* and *Vernonia amygdalina*.**

**ABSTRACT**

**Aim:** The aim of the study was to carry out a comparative analysis of the proximate, antioxidant vitamins and mineral composition of the leaves of four selected tropical nutritional/ medicinal plants namely: *Ocimum gratissimum*, *Piper guineense*, *Gongronema latifolium* and *Vernonia amygdalina*. **Methodology:** The macro and micronutrients in the plant leaves were extracted by cold maceration in ethanol and subjected to quantitative proximate, antioxidant vitamins and minerals analysis. **Results:** For all four plants, carbohydrates was the major macronutrient constituents followed by crude fats, crude proteins, ash and fiber in that order. *G. latifolium* had the highest carbohydrate and protein composition while *V. amygdalina* had the highest crude fat composition. Results of Ash analysis of the four leaves showed *P.guineense* to have the highest total mineral content followed by *V. amygdalina*, *O. gratissimum* and *G. latifolium* in that order. *O. gratissimum* and *P.guineense* had the highest composition of crude fiber closely followed by *G. latifolium* and *V. amygdalina*. Vitamin analysis revealed that leaves of the four vegetable plants contained high levels of vitamin C and appreciable quantities of vitamins A and E. *V. amygdalina* leaf contained the highest concentration of vitamin C and A while vitamin E was pretty much the same for the four plants. The mineral assay indicated that the leaves of the plants contain high levels of Magnesium (Mg), Phosphorus (P) and Calcium (Ca) relative to their copper (Cu), Zinc (Zn), Potassium (K) and Sodium (Na) contents. **Conclusion:** In conclusion, these plants were shown to be rich in carbohydrates, proteins and fats, vitamins and minerals justifying their use in diets. The plants were particularly rich in antioxidant vitamins and mineral justifying the therapeutic uses of various preparations of these leafy vegetables, in traditional medicine, for the treatment and management of diseases that have their etiology and pathophysiology in free radical generation and oxidative stress.

**Key words:** Proximate, Vitamins, Minerals, Tropical nutritional plants, *Ocimum gratissimum*, *Piper guineense*, *Gongronema latifolium* and *Vernonia amygdalina*.

**1. INTRODUCTION**

Plants, from prehistoric times, have been used as spices, food and medicine in all cultures [1-3]. More than 70% of people in developing countries depend on plants (vegetables and fruits) for regular dietary needs [4]. It is well known that apart from energy needs, many plants and foods are ingested because of their perceived medicinal and health benefit. Indeed a significant amount of research has shown a correlation between a healthy diet and lifestyle and significant reductions in diseases and associated chronic conditions [5]. Plants are also a rich source of vitamins and minerals. Studies have identified a vast majority of antioxidant vitamins from vegetable plants like vitamins A, C and E [6]. Selective intake of

53 foods containing these antioxidant vitamins, minerals and phytochemicals can prevent the  
54 onset of degenerative diseases like cardiovascular diseases, cancer and diabetes.

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56 Given the plethora of vegetable plants available it becomes difficult to identify which plant  
57 should be added to our diet to address particular nutrient deficiency or ameliorate particular  
58 ailments. Moreover, some plants may contain appreciable levels of anti-nutrients rendering  
59 them unsafe for human consumption. It is with a view to establishing the relative proximate,  
60 antioxidant vitamins and mineral composition in some commonly used vegetable leafs in the  
61 southern region of Nigeria namely *Ocimum gratissimum*, *Piper guineense*, *Gongronema*  
62 *latifolium* and *Vernonia amygdalina*, that the current study is being carried out. The focus on  
63 the four plants stems from their common use as vegetables and spices in soups in the  
64 southern part of Nigeria. The plants have also been employed in ethnobotany for the  
65 treatment of various diseases. A comparative analysis of the proximate, antioxidant vitamins  
66 and minerals composition of the four plants will provide a bio-rational basis for the choice of  
67 the plants for addressing some nutrient deficiency. Earlier work in our laboratory had carried  
68 out a comparative analysis of the phytochemical composition of the four plants [7].

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70 *Ocimum gratissimum* commonly called African basil and belonging to the family *Lamiaceae*,  
71 is a herbaceous perennial flowering plant which is woody at its base. The leaf is called scent  
72 leaf because it possesses a pleasant aroma which is responsible for its use as spice and  
73 condiments in cooking. It is widely distributed in tropical Africa and Asia, especially India.  
74 The plant is economically important for its food flavoring (as spice and condiments) [8] and  
75 essential oil which has been widely used in food industries [9,10].

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77 *Piper guineense* (family *Piperaceace*) is a climbing perennial plant native to the tropical  
78 regions of Central and Western Africa. It is commonly referred to as Ashanti pepper, West  
79 African pepper or African black pepper. *Piper guineense* is economically important for its  
80 culinary uses as well as medicinal, cosmetic and insecticidal uses [11]. It is a highly spicy  
81 plant and the leaves have pungent taste and a pleasant aroma when crushed. It thus imparts  
82 "heat", "pungency" and a spicy aroma to classic West African soups (stews). The plant oils is  
83 used as aromatics in the drink industry [12].

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85 *Gongronema latifolium*, commonly called "utazi," "aroeke" in the South Eastern and South  
86 Western parts of Nigeria respectively, belongs to the family *Asclepiadaceae*. It is primarily  
87 used as spice and vegetable for cooking and in traditional medicine [13]. A non-wood forest  
88 plant, it is native to West Africa and widely distributed elsewhere in tropical Africa and  
89 subtropical Asia.

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91 *Vernonia amygdalina*, popularly called bitter leaf, belongs to the family *Asteraceae*. It is  
92 widely used in the West African sub-region for a number of medicinal and nutritional  
93 purposes [14,15]. It has also been employed as a digestive tonic and appetizer [16].

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## 2. MATERIALS AND METHOD

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### 2.1 Plant Materials

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101 Mature leaf samples of *Ocimum gratissimum*, *Piper guineense*, *Gongronema latifolium* and  
102 *Vernonia amygdalina* were harvested from local farms in Cross River State, Nigeria.

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### 2.2 Methods

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#### 2.2.1 Extraction Procedure

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107 Fresh leaves of each plant were washed and air dried at room temperature (25°C) for two  
108 weeks. The dried leaves were pulverized using a mechanical grinder. A weighed quantity,  
109 200g, of each plant material was extracted by cold maceration in absolute ethanol for 48  
110 hours. The extracts were double filtered, first with a white muslin cloth then with Whatman  
111 no.1 filter paper. The resulting ethanol leaf extracts were concentrated in vacuum using a  
112 rotary evaporator (at temperatures between 40°C and 45°C) to obtain a semi-solid mass.  
113 Weighed quantities of each extract was dissolved in 5% Tween 80 solution for use in the  
114 macro and micro nutrient analysis.

### 115 **2.2.2 Proximate Analysis**

116 Proximate composition of the leaf extracts was determined using methods prescribed by the  
117 Association of Official Analytical Chemists (AOAC) [17] and the Food and Agriculture  
118 organization (FAO) [18].

### 119 **2.2.3 Determination of Mineral Composition**

120 Potassium and sodium were determined by the Flame photometric method while iron,  
121 copper, zinc, calcium and magnesium were determined by atomic absorption  
122 spectrophotometric method as described by James [19] and the Association of Official  
123 Analytical Chemists, AOAC [20]. Phosphorus was determined spectrophotometrically by the  
124 vanadomolybdate yellow method.

### 125 **2.2.4 Determination of Some Antioxidant Vitamins**

126 Vitamin A and E concentration was determined by the spectrophotometric method as  
127 described by Pearson [21]. Vitamin C was determined by the method of AOAC [22].

## 128 **3. RESULTS & DISCUSSIONS**

### 129 **3.1 Proximate Analysis**

130 The result of the proximate composition of the fresh leaves of the four plants is shown in  
131 Figure 1. On balance, for all four plants, carbohydrates was the major macronutrient  
132 constituents followed by crude fats, crude proteins, ash and fiber in that order. *G. latifolium*  
133 had the highest carbohydrate composition followed by *O. gratissimum*, and then  
134 *P.guineense* and finally *V. amygdalina*. Dietary carbohydrate is a major macronutrient for  
135 both humans and omnivorous animals; human adults in the Western countries obtain  
136 approximately half their daily caloric requirements from dietary carbohydrate while it the  
137 major source of energy in other countries [23]. Carbohydrate is stored as glycogen, and  
138 although it is important for short-term energy needs, it is of very limited capacity for providing  
139 for energy needs beyond a few hours.

140 Fats, the second highest macro nutrient in the four plants, constitute the highest energy in  
141 humans. *V. amygdalina* had the highest crude fat composition followed by *O. gratissimum*,  
142 *G. latifolium* and *P.guineense*.

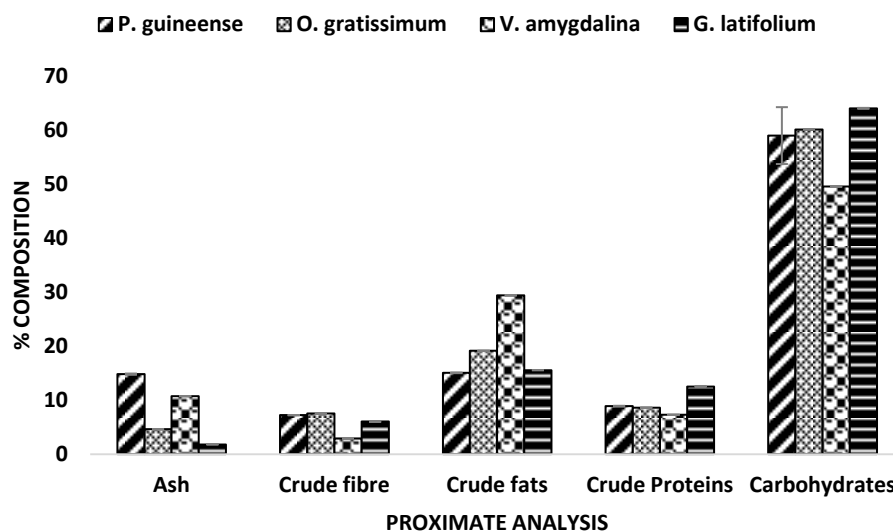
143 Protein is the second largest store of energy in the body after adipose tissue fat stores [24].  
144 The result of macronutrient analysis revealed that all the four plants were a fairly rich source  
145 of protein and may be used as a protein supplement for patients with protein deficiency  
146 diseases. *G. latifolium* had the highest protein composition followed by *P.guineense*, *O.*  
147 *gratissimum* and *V. amygdalina* in that order.

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161 Ash, which refers to the inorganic residue remaining after ignition or complete oxidation of  
 162 organic matter in a food sample, is a measure of the total amount of minerals present within  
 163 the food [25]. Results of Ash analysis of the four leaves shows *P.guineense* to have the  
 164 highest total mineral content followed by *V. amygdalina*, *O. gratissimum* and *G. latifolium* in  
 165 that order.

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 167 Crude fibre is a measure of the quantity of indigestible cellulose, pentosans, lignin and other  
 168 like components in foods. Insoluble fibers can help promote bowel health and regularity. It  
 169 also support insulin sensitivity and may help reduce the risk of diabetes. *O. gratissimum* and  
 170 *P.guineense* had the highest composition of crude fiber closely followed by *G. latifolium* and  
 171 *V. amygdalina*.

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 175 Fig 1: Proximate Analysis of crude leaf extracts of *P.guineense*, *O.gratissimum*, and *V.*  
 176 *amygdalina* and *G.latifolium*. Values are expressed as mean  $\pm$  SEM.

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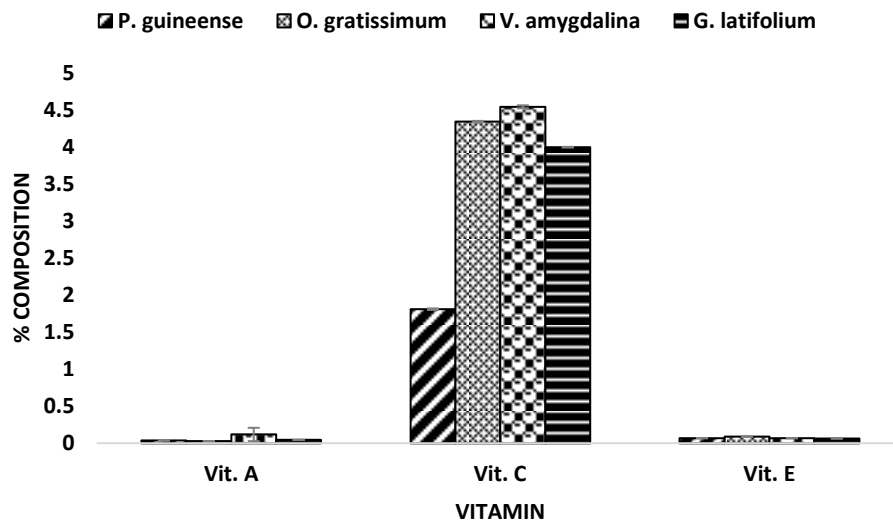
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### 179 3.2 Vitamins

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181 The protective action of fruit and vegetables has been attributed to the presence of  
 182 antioxidants, especially antioxidants vitamins including ascorbic acid,  $\alpha$ -tocopherol and beta-  
 183 carotene [26-28]. The results of this study (figure 2) revealed that leaves of the four  
 184 vegetable plants contain appreciable concentration of vitamin C, vitamin E and beta-  
 185 carotene (vitamin A). *V. amygdalina* leaf contained the highest concentration of vitamin C  
 186 and vitamin A. Other reports have also shown the plant to be rich in Vitamin C and A [28,29].  
 187 The level of Vitamin E was pretty much the same for the four plants with *O.gratissimum*  
 188 having a slightly higher amount. These results seem to suggest that fresh leaves of the  
 189 plants are good sources of antioxidant vitamins. Vegetable leaves/ Spices provide a variety  
 190 of vitamins and minerals as well as macronutrients to the diet [30]. These antioxidant  
 191 vitamins appear to be partly responsible for the antioxidant properties of the leaves. Vitamin  
 192 C is an antioxidant which helps to protect the body against cancer and other degenerative  
 193 diseases such as arthritis and type 2 diabetes mellitus and also strengthens the immune  
 194 system [31]. Vitamin C has also been shown to facilitate iron absorption by its ability to  
 195 reduce inorganic ferric ion to the ferrous form [32]. This suggests that the vegetable leaves  
 196 may be beneficial to people suffering from iron-deficiency anemia. Vitamin E ( $\alpha$ -tocopherol)  
 197 appears to be the most important lipid soluble antioxidant protecting membranes from lipid  
 198 peroxidation by acting as a chain-breaking antioxidant [33]. It also limits the oxidation of LDL

199 cholesterol and may help prevent or delay the development of atherosclerosis and/or  
 200 coronary heart disease (CHD) [34]. This probably explains why high vitamin E intake is  
 201 associated with lower rates of heart diseases. Beta-carotene is a lipid-soluble antioxidant. It  
 202 is the precursor of vitamin A, so it is necessary for the production and re-synthesis of  
 203 rhodopsin. High levels of beta-carotene intake have been correlated with lower risk of lung  
 204 cancer, coronary heart disease, stroke and age-related eye disease [35].  
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206 Fig 2: Quantitative Analysis of some Antioxidant Vitamins in *P.guineense*, *O.gratissimum*,  
 207 and *V. amygdalina* and *G.latifolium*. Values are expressed as mean  $\pm$  SEM.  
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### 210 3.3 Minerals

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 212 The results of the quantitative analysis of mineral elements (Figure 3) indicate that the  
 213 leaves of the plants contain high levels of Magnesium (Mg), Phosphorus (P) and Calcium  
 214 (Ca) relative to their copper (Cu), Zinc (Zn), Potassium (K) and Sodium (Na) contents. In  
 215 addition to the numerous biological roles these minerals play, they also serve as co-factor in  
 216 certain biochemical reactions including those involving antioxidant enzymes. Magnesium  
 217 serves as a co-factor for the enzyme catalase, a primary antioxidant that detoxifies hydrogen  
 218 peroxide by dismutation to water and oxygen. Similarly Copper and Zinc, are vital co-factor  
 219 of the different forms of SOD found in plants and animals [36]. Superoxide dismutase (SOD)  
 220 is a primary antioxidant enzyme that catalyses the dismutation or disproportion of superoxide  
 221 anion radicals ( $O_2^-$ ) to hydrogen peroxide and molecular oxygen [37]. It is therefore  
 222 suggested that these minerals contribute to the antioxidant properties of the plants probably  
 223 by boosting the levels of antioxidant enzymes such as SOD and catalase.  
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225 Phosphorus: Except for *P.guineense*, Phosphorus (P) was the major constituents of the  
 226 mineral elements assayed. *O.gratissimum* had the highest phosphorus content closely  
 227 followed by *V. amygdalina* and *G.latifolium* in that order. Phosphorus is an ubiquitous  
 228 mineral in the human body and has diverse functions ranging from the transfer of genetic  
 229 information to energy utilization [38]. It forms the backbone of DNA and RNA, is an essential  
 230 component of phospholipids that form all membrane bilayers and is an integral component of  
 231 the body's key energy source, adenosine triphosphate (ATP). Phosphorus also plays a vital  
 232 role in the dissociation of oxygen from hemoglobin, is the main intracellular buffer and  
 233 therefore is essential for pH regulation of the human body and is a key component of the  
 234 second messenger molecules such as cyclic adenosine monophosphate (cAMP), cyclic  
 235 guanine monophosphate (cGMP) and inositol polyphosphates. Taken together with the  
 236 equally high level of carbohydrates, the four plants are a very good source of energy.  
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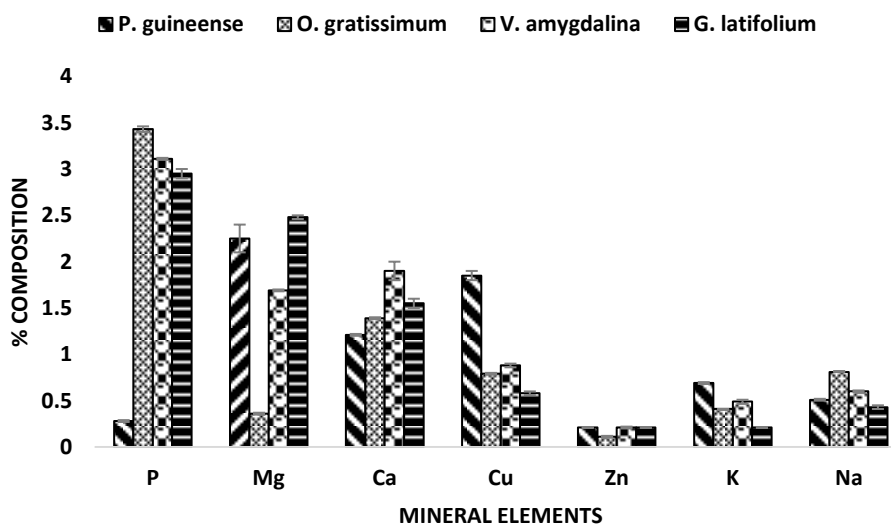
238 Magnesium (Mg): Comparatively *G.latifolium* had the highest Mg content with *O.gratissimum*  
 239 having the lowest. *G.latifolium* is thus the plant of choice to address Mg deficiency. Mg  
 240 plays an essential role in a wide range of fundamental biological reactions. Apart from its  
 241 cofactor role, it is involved in bone mineralization, the building of proteins, muscle  
 242 contraction, nerve transmission and immune system health [4,39]  
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244 Calcium (Ca) is the most tightly regulated ion in the extracellular fluid (ECF). In higher  
 245 mammals, the most obvious role of calcium is structural or mechanical being responsible for  
 246 the mass, hardness, and strength of the bones and teeth [40]. Calcium is also involved in  
 247 cell movement and muscle contraction to nerve transmission, glandular secretion, and even  
 248 cell division where it acts as both a signal transmitter from the outside of the cell to the inside  
 249 and as an activator or stabilizer of the functional proteins involved. Calcium also plays a role  
 250 in the regulatory activities of parathyroid hormone [PTH], calcitonin [CT], and a key activity of  
 251 vitamin D. Ca was more predominant in *V. amygdalina* followed by *G.latifolium*,  
 252 *O.gratissimum* and *P.guineense* in that order.  
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254 Copper is a constituent of many enzymes including superoxide dismutase. It is also required  
 255 for iron metabolism [4,41]. It was more prevalent in *P.guineense*.  
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257 Zinc plays a catalytic, structural, and regulatory role in the body [42]. Zinc is essential for  
 258 general growth and proper development of the reproductive organs and for normal  
 259 functioning of the prostate gland. Apart from SOD, Zinc is a co-factor of over 300 enzymes  
 260 including carbonic anhydrase, which is crucial to maintenance of acid-base balance in the  
 261 blood, and alcohol dehydrogenase that break down alcohol. It is also a component of insulin  
 262 and plays a role in its processing, storage, secretion and action [43]. The Zinc content of *P.*  
 263 *guineense* may be responsible for the observed stimulated sexual behaviors of mature male  
 264 rats fed with extract of *P. guineense* [44]. The level of the mineral was pretty much the same  
 265 in *V. amygdalina*, *G.latifolium* and *P. guineense*. *O.gratissimum* had the lowest level of the  
 266 mineral.  
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268 Sodium (Na) and potassium (K) (and chloride ions Cl<sup>-</sup>) are the major electrolytes located in  
 269 all body fluids. While sodium is extracellular, potassium is intracellular. They are responsible  
 270 for the maintenance of acid/base balance, nerve transmission and muscle contraction and  
 271 regulation of fluid movement in and out of cells [45]. *P. guineense* had the highest amount  
 272 of potassium while *O.gratissimum* had the highest level of sodium.



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 275 Fig 3: Quantitative Analysis of some Minerals in *P.guineense*, *O.gratissimum*, *V. amygdalina*  
 276 and *G.latifolium*. Values are expressed as mean  $\pm$  SEM.

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#### 4. Summary and Conclusion

In summary, the four plants, *P.guineense*, *O.gratissimum*, *V. amygdalina* and *G.latifolium*, have been shown to be rich in carbohydrates, proteins and fats, vitamins and minerals justifying their use in diets. The plants are particularly rich in antioxidant vitamins and mineral. Taken together with our earlier work on the comparative phytochemical analysis of these plants [7], the findings have good correlation with the therapeutic uses of the various preparations of these leafy vegetables in traditional medicine for the treatment and management of diseases that have their etiology and pathophysiology in free radical generation and oxidative stress like diabetes, arthritis, rheumatism, eye problems and infectious diseases such as AIDS. Increased consumption of the leaves of these plants is therefore recommended (especially as they have been shown to contain low levels of anti-nutrients [7]) for optimized health and wellness, and to boost the endogenous antioxidant system and in so doing, help prevent the development of certain free radical related diseases.

#### COMPETING INTERESTS

The Authors declare that no competing interests exist.

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