Proximate and Mineral Composition of Some Leafy Vegetables Sold in Farin Gadan Market in Jos, Plateau State, Nigeria

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Authors’ contributions
This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT
This study was carried out to determine the mineral concentration and proximate composition of four different leafy vegetable collected in from Farin Gadan Market Jos Plateau State Nigeria and extracted using double acid extraction method. Heavy metals concentration was performed using atomic absorption spectrophotometer and proximate composition was determined by Association of Official Analytical chemistry (AOAC, 1999) method. Results showed that the highest level of lead was found in Jute (0.045 ±0.004) while Lettuce (0.001 ±0.007) had the lowest level of lead. The leafy vegetables in this study may have been polluted with lead through pollutants in irrigation water, farm soil or due to pollution from the highway traffic. Chromium and cadmium were observed to be low in all the leafy vegetables analyzed, which is still within the safe limit set by FAO/WHO. Zinc had the highest concentration (2.802± 0.012) in bitter leaf, proximate analysis indicated significant variability in the constituents contained in the leafy vegetables studied (Carbohydrate, Ash content, crude protein, crude fats, crude fibre and moisture content in descending order). The highest moisture content was found in Lettuce (7.791%) and the highest crude protein in Bitter leaf.
The concentrations of the toxic heavy metals are within the tolerable limits of some regulatory authorities. However, there is need for continuous monitoring of heavy metal and proximate analysis in leafy vegetables because leafy vegetables are the main sources of food in many parts of the world and are considered as bio-indicators of environmental pollution.

Keywords: Vegetables; heavy metal; proximate; AAS.

1. INTRODUCTION

Vegetable is an herbaceous plant or portion of a plant that is eaten whole or part, raw or cooked, generally with an entrée or in a salad but not as a dessert [1]. Fresh vegetables are of great importance in the diet because of the presence of vitamins and mineral salts. In addition, they contain water, calcium, iron, Sulphur and potash [2]. They constitute an important part of human diet since they contain carbohydrates, proteins, vitamins, minerals and fibres. They are very important foods useful for the maintenance of health and the prevention and treatment of various diseases. However, the plants contain both toxic and essential metals over a wide range of concentrations [3].

All societies and ethnic groups eat vegetables because they are essential for maintaining human health in order to meet basic nutritional needs. Vegetable is the part of a plant that is consumed by humans as food as part of a meal, used to increase the quality of soups and also for their dietary purposes [4]. They are rich in nutrients required for human health and are important sources of carbohydrates, proteins, vitamins, minerals and fibre [5].

Vegetables play an important role in human nutrition as most are low fats and calories and contain vitamins such as vitamin A, C and E and mineral salts. They also serve as neutralizing agent for acidic substances formed during digestion [5], they are found to be having reduction in the case of cancer, stroke, cardiovascular diseases and other chronic ailments. Leafy vegetables are widely used for culinary purposes [4]. They are made up of mainly a cellulose, hemi-cellulose and pectin substance that gives them their texture and firmness [4]. In addition, fresh vegetables contain water, calcium, iron, Sulphur and potassium [2].

1.1 African Spinach (Amaranthus hybridus)

*Amaranthus hybridus* is an annual herbaceous plant of 1 – 6 feet high. The leaves are alternate petioled, 3 – 6 inches long, dull green and rough, hairy, ovate or rhombic with wavy margins. The flowers are small with greenish or red terminal panicles. Taproot is long, fleshy red or pink. The seeds are small and lent cellular in shape; with each seed averaging 1 – 1.5mm in diameter and 1000 seeds weighing 0.6 – 1.2g. It is rather a common species in waste places, cultivated fields and barn yards. In Nigeria, *Amaranthus hybridus* leaves combined with condiments are used to prepare soup [6].

As cited in [7], *Amaranthus hybridus* has been shown to contain large amount of squalene, a compound that has both health and industrial benefits. Raw spinach is 91% water 4% carbohydrate 3% protein and contain negligible fat. It has a high nutritional value, especially when fresh, frozen, steamed quick boiled. It is a rich source of vitamins A, C, K, E and B riboflavin and B6. It also contains magnesium, manganese, iron, calcium, potassium, folate and dietary fibre.

1.2 Lettuce (Lactuca sativa)

Lettuce is an annual or biennial plant that has been cultivated for centuries. It was served to Persian kings about 600 years before Christ. The Romans grew the romaine or Cos type as early as the beginning of the Christian era. The Chinese have grown the crop since the fifth century A.D., and by the sixteenth century it had been taken to central and south America [8]. Cultivated lettuce is closely related to the common wild or prickly lettuce (L. Serriola) weed. The many different kinds of lettuce are often grouped into three types. The most common is head lettuce of which there are crisphead (or iceberg) and butterhead cultivars. The romaine of cos type forms loose upright heads. The leaf is none heading and loose – leaved.

Lettuce is, by far, the most importance salad crop. It is grown commercially in about 15 states in the United States and in home gardens in most of the others. California produces by far the largest amount of crisphead lettuce in both area and dollar value.
Lettuce is grown in Texas, New Mexico, New York, New Jersey, Washington and Michigan. Production in leaf and romaine types has increased recently because of the increased popularity of tossed green salads.

Lettuce is a cool-season crop and grows best at a relatively cool temperature of about 12°C to 15°C (55°F to 60°F). It is grown during the summers in the northern states at higher altitudes, or along the coast (e.g., Salinas valley) in California, in Oregon and Washington. In Florida, Texas, Arizona New Mexico and the desert valleys of California (Imperial Valley) lettuce is grown during the winter months. Heading is prevented by seed stalk formation when temperature reaches 21°C to 26°C (70°F to 79°F); also, the leaves develop a bitter taste. Some slow bolting cultivars have been developed to delay premature seed stalk formation. Lettuce seedling tolerate some slight freezing but the larger, more mature leaves are injured. Seeds germinate best at soil temperatures from 5°C to 25°C (41°F to 77°F) [8].

Plant breeding programs conducted jointly by the USDA and some university experiment stations have been responsible for the release of lettuce cultivars adapted to particular environmental conditions and location or resistant to diseases and pests [8].

Lettuce grows well on a wide variety of soils provided the climate requirements are met, especially temperature – lettuce grows on much soils in Florida, Sandy loams in Texas New Jersey, New Mexico and Arizona and on clay or clay loam soils in the Salinas valley of California. Fertile soils high in organic matter are preferred by most farmers. If high temperature is a problem, remember that heavier soils are often cooler than light sandy soils. Soils with high water holding capacity and good drainage are better for the lettuce root system [8].

1.3 Bitter Leaf (Vernonia amygdalina)

The herb known as bitter leaf (Vernonia amygdalina) is a shrub or small tree that can reach 23 feet in height when fully grown. Bitter leaf has a gray or brown coloured bark, which has a rough texture and is flaked. The herb is an indigenous African plant, which grows in most parts if sub-Saharan Africa. The East African Country of Tanzania is traditionally linked to this plant and can be found growing wild along the edges of agricultural fields.
It is a medicinal plant and fresh bitter leaf is of great importance in human diet because of the presence of vitamins and mineral salts [4]. It is very important protective food and useful for the maintenance of health and prevention and treatment of various diseases. Some principal chemical constituents found in bitter leaf herb are a class of compounds called steroid glycosides – type vernonioside β1 – these chemical substances possess a potent anti-parasitic, anti-tumor and bactericidal effect. The bitter leaf is mainly employed as an agent in treating schistosomiasis, which is a disease caused by parasitic worms. It is also useful in the treatment of diarrhea and general physical malaise.

Remedies made from bitter leaf are used in treating 25 common ailments in sub-Saharan African; these include common problems such as fever, and different kinds of intestine complaints, as well as parasite – induced diseases like malaria. Bitter leaf, also helps to cleanse such vital organs of the body like the liver and the kidney. Bitter leaf is also used in the treatment of skin infections such as ringworm, rashes and eczema. However, bitter leaf and other vegetables contain both essential and toxic metals over a wide range of concentrations [3].

1.4 Jute Leaves (*Corchorus olitorius*)

Jute leaves are also known as saluyoi, ewedu or Lalo. They are also known for their medicinal properties. It may prevent conditions like arthritis, acne, asthma, cold. Jute leave may not be known to many but they are a part of the jute
Fig. 4. Jute Leaves (*Corchorus olitorius*)

Jute leaves are used as a food source in these regions. In fact, they are said to add a distinct flavor to food and also act as thickeners in soups, stews and sauces. Jute leaves are also known as soluyot, ewedu or lalo, depending on the region they are cultivated or cooked in. The leaves have slightly toothed edges. When harvested young, jute leaves are generally flavorful and tender on the other hand, older leaves tend to be fibrous and woody. Scientifically known as *Corchorus olitorius*, parts of Jute plants are used in many ways, while Jute stems are used to make rope, paper and a variety of other products. Jute leaves are not just for culinary uses but are known for their medicinal properties.

Heavy metals are non-biodegradable and are persistent environmental contaminants which may be deposited on the surfaces and then absorbed into the tissue of vegetables. Plants take up these heavy metals by absorbing them from deposit on the parts of the plants exposed to the air from pollutants as well as from contaminated soil [9]. Contamination of vegetables with heavy metals as Cadmium (Cd), Lead (Pb), Mercury (Hg), Copper (Cu), Chromium (Cr), Cobalt, Nickel (Ni) and Arsenic (As) may be due to irrigation with contaminated water and metal-based pesticides, industrial emissions, transportation of the harvested products, storage and at the point of sales [9].

According to the definition, a general term “heavy metal applies to the group of metals and metalloids with atomic density greater than 4g/cm, or is 5 times or more, greater than water density. In this group Lead (Pb), Cadmium (Cd), Nickel (Ni), Cobalt (Co), Iron (Fe), Zinc (Zn), Chromium (Cr), Silver (Ag), Arsenic (As), and the platinum group elements are included. There is also an alternative classification of metals which is based on their coordination chemistry. This classification categorizes heavy metal as class B metals that are non—essential trace element and are highly toxic elements which include Mercury (Hg), Silver (Ag), Lead (Pb), Cadmium (Cd) and Nickel (Ni) [10].

Heavy metal poisoning is the accumulation of heavy metals in toxic amount in the soft tissue of plants or animals’ body. The accumulation of these heavy metals in high concentration in the body cause poisoning and serious damage may occur.

Although these metals have crucial biological function in plants and animals, these metals bind with proteins sites which are not made for them by displacing original metals from their natural binding sites causing malfunctioning of cells and ultimately toxicity. Previous research has found that oxidative deterioration of biological macromolecules is primarily due to binding of heavy metals to the DNA and nuclear proteins [11].

2. MATERIALS AND METHODS

2.1 Reagents and Equipment

H₂SO₄, Na₂SO₄, NaOH, HCl, HNO₃, CuSO₄, SeO₂, Boric acid, Soxhlet extractor, Petroleum ether, Conical flask, weight balance, Desiccator, Kjeldal apparatus, Methyl orange, Crucibles, Muffler furnace and AAS model VG 210. All the reagent are of analytical grade obtained from
Sigma Aldrich Chemicals, Nigeria office in Lagos.

2.2 Sample Collection and Treatment

The vegetables sampled include African spinach (Amaranthus hybridus), Lettuce (Lactuca sativa), Bitter leaf (Vernonia amygdalina), and Jute leaves (Corchorus olitorius). These vegetables were bought from Farin Gada market in Jos North Local Government Area of Plateau State, Nigeria. Part of the fresh samples were sliced and used for the determination of proximate content. The rest were, sliced and dried in the oven at 80°C for 24hrs. these were then grinded to fine powder using a blender and stored in a clean and dry polyethylene container for digestion to determines the mineral component of the samples.

2.3 Digestion and Analysis of the Metals in Vegetables Samples

The dried samples were grinding using mortar and pestle and 1.0g of each powdered sample were weighed using electronic balances and put into digestion flask mixed with 10cm³ of concentrated hydrochloric (HCl) acid and nitric acid (HNO₃) in the ratio 1:1 (10ml:10ml) respectively. The digestion flask was heated at 70°C for 40 minutes then the heat was increased to 120°C. The digestion was, completed when the solution became clear / transparent and white fumes appear. The digest was diluted with 20cm³ of distilled water and boiled for 15 minutes. This was then allowed to cool and was transferred into 100cm³ volumetric flask and diluted to the mark with distilled water. The sample solution was then filtered through a filter paper and the filtrate was analyzed for metals using AAS model VGP 210 model [12].

2.4 Determination of Moisture Content

The air-dried vegetable samples were grinding using mortar and pestle and 5g of each powdered sample were weighed in duplicate each in a pre-weighed petri-dish and placed in hot air oven maintained at 105°C for 24 hours, they were removed and placed in a desiccator to cool down to room temperature, and reweighed. The loss in weight after drying was expressed as percentage of moisture content. The results were calculated as follows:

Weight of empty Petri dish = X

Weight of Petri dish + sample before drying = y

Weight of sample + silk cloth after extraction = p

Percentage fat content = \( \frac{p - r}{q} \times 100 \) [13] (3)

2.5 Determination of Ash Content

Each selected vegetable sample was weighed 2g in duplicate in a pre-weighed porcelain crucible. The crucibles were then placed in a muffle furnace maintained at 600°C for 8 hours. The crucibles were removed and put in a desiccator to cool. They were then weighed and the weight of the ash expressed as percentage ash content as shown below;

Percentage Ash = \( \frac{weight of crucible + ash - weight of the crucible}{weight of sample} \times 100 \)

Weight of ash + crucible – weight of crucible = ash(g)

Then, percentage \( \frac{ash \times 100}{weight \ of \ sample} \) [13] (2)

2.6 Determination of Crude Fat

Moisture free selected vegetable samples were weighed 5g in a pre-weighed fat free white silk cloth in duplicate. The cloth was tied and placed in the Soxhlet extractor fitted to the receiving flask containing petroleum ether (60°C – 80°C) and condenser.

The samples were then extracted for 8hours on a heating mantle. After this period the samples were removed from the Soxhlet extractor, dried in an oven maintained at 105°C for 24hours and re-weighed. The loss in weight of the samples were expressed as fat content of the samples as shown below;

Weight of the sample = q

Weight of sample + silk cloth before extraction = p

Percentage fat content = \( \frac{p - r}{q} \times 100 \) [13] (3)

2.7 Crude Fibre Determination

i. Preparation of digestion mixture: The digestion reagent was prepared by mixing thoroughly 45cm³ of glacial acetic acid, 50cm³ of distilled water, 5ml of concentrated nitric acid in 100ml volumetric flask.

ii. Determination – 0.1 of dried selected vegetable samples were accurately weighed and quantitatively transferred to a
150cm$^3$ quick fit conical flask. 20ml digestion mixture was added and the mixture refluxed for 45 minutes with constant shaking. They were then filtered through ash less filter papers previously dried using gentle suction. The residue was then washed with 100ml of distilled water and 50cm$^2$ of ethanol followed by 50ml of petroleum ether (60°C – 80°C). It was then dried in an oven at 100°C, allowed to cool and weighed.

The filter paper and the content were transferred into pre-weighed crucible and in muffle furnace for six hours at 600°C and then re-weighed. The loss in weight on ignition was expressed as percentage crude fibre. Duplicate determinations were carried out and results expressed as below:

Weight of sample = a
Weight of filter paper (ashless) = x
Weight of filter paper + direct sample after filtering = y
Weight of empty crucible = z
Weight of ash + crucible q

Percentage crude fibre
= \frac{(y - x) - (q - z) \times 100}{a} \quad [13] \quad (4)

### 2.8 Crude Protein Determination

#### 2.8.1 Digestion of sample

Moisture free of each selected vegetable sample was accurately weighed 0.5g into kjeldah digestion flask o.2g catalyst made of anhydrous sodium sulphate, cupper (II) sulphate, and selenium di oxide in ratio 98:1:1 was then added. 10ml of concentrated Sulphuric acid was added and the moisture digested by heating until the solution became clear. The solution was allowed to cool after digestion. A blank consisting of the digestion reagents only was set up and also digested. The solutions were transferred into 100ml volumetric flask and diluted to 100ml.

#### 2.8.2 Distillation of sample

10ml of the solution was put in the Markham still and 20ml of 40% NaOH was added, the mixture was then steam distilled into 20ml of 2% boric acid containing 4 drops of screened methyl orange indicator. A pale green solution was obtained. After about 70m/s of the distillate was collected, the process was stopped.

#### 2.8.3 Titration

The distillate was titrated against 0.05MH$_2$SO$_4$ which was standardized with 0.0MNa$_2$CO$_3$. pink colour was obtained at end point. The percentage protein content of the sample was determined from the titre values of both the samples and the blank. Duplicate determinations were carried out for all the samples and blank.

\[ \text{% protein} = \left( \frac{(100 \times T - B \times C_A \times TV_S) \times 6.25}{1000 \times W} \right) \times \frac{1000}{V_S} \quad (5) \]

W = Weight of sample digested
T = Titre value of sample
B = Titre value of Blank
C$_A$ = Concentration of acid
TV$_S$ = Total volume of sample solution after digestion
V$_S$ = Volume of sample used for distillation

### 2.9 Carbohydrate Determination

Carbohydrate was determined by difference. The results were calculated as follows.

\[ \text{Carbohydrate} = (100 - \text{moisture content} + \text{Ash} + \text{Fat} + \text{Fibre} + \text{Protein}) \quad (6) \]

### 3. RESULTS AND DISCUSSION

The results of the proximate analysis of the vegetables from Farin Gada market in Jos North Area of Plateau State, Nigeria is shown in

|                   | Bitter leaf | Lettuce | Jute  | African spinach |
|-------------------|-------------|---------|-------|-----------------|
| Moisture          | 6.263       | 7.791   | 7.430 | 5.970           |
| Fats              | 13.840      | 15.010  | 10.480| 6.491           |
| Crude fibre       | 9.990       | 8.910   | 7.740 | 13.930          |
| Crude protein     | 12.880      | 1.770   | 15.500| 15.630          |
| Ash content       | 10.930      | 15.257  | 11.422| 19.860          |
| Carbohydrate      | 46.097      | 51.262  | 47.428| 38.119          |

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The crude protein content ranged from 5.97% in African Spinach to 7.79% in Lettuce. The percentage moisture content varies from 5.97% in African Spinach to 7.79% in Lettuce.

Table 1 and that of heavy metals which is obtained by taking the average of each element analyzed three time using AAS model VG 210 is indicated in Table 2.

The results of proximate analysis (%) of the selected leafy vegetable are shown in table 1. The percentage moisture content varies from 5.97% in African Spinach to 7.79% in Lettuce. The moderate moisture content provides for moderate activity of water-soluble enzymes and co-enzymes needed for metabolic activities of these leafy vegetables [14]. African spinach had (19.86%) of ash content when compare with bitter leaf with (10.93%) which is the least of the leafy vegetable investigated. This confirm that there are more minerals in African Spinach and Lettuce than Bitter leaf, the values of Ash content obtained are comparable to those reported for some leafy vegetable such as A. hybridus, C. peps and E. Africana [14]. The percentage of fat content ranged from 6.49% – 15.01% in African Spinach and Lettuce respectively. These values were fairly high when compared with values reported in some other leafy vegetable such as ocimum bassilium, ocimum viride and piper guineens [15].

The crude protein content ranged between 15.63% to 1.77%. The crude protein was similar to what was reported for some other leafy vegetable such as Momordica balsamina (11.29%) Moringa Oleifra (20.72%), Lesianthera africaciana leaves (13.10% - 14.90%) and Leptadenia hastate (19.10) [16]. Plant foods that provide more than 12% of their calorific value from protein have been shown to be good source of protein [17]. This shows that all the leafy vegetables investigated are all good sources of protein. The crude fibre content ranged from the reported values (8.50% – 20.90%) for some Nigerian vegetables [18]. Dietary fibre helps to prevent constipation, bowel problems and piles. The total carbohydrate content ranged between 38.119% – 51.262%. The values obtained for all the leafy vegetables under this study fell within amount of carbohydrate for momdrica balsamina (39.05%).

Table 2 shows the mineral composition of leaf vegetables, nutritional significant of element when compared with the standard recommended dietary allowance. The content of potassium, calcium, sodium and magnesium were of comparable values; the values were moderate when compared with standard dietary allowance. The content of potassium, calcium, sodium and magnesium are important intracellular and extracellular cations respectively. Sodium is involved in the regulation of plasma volume, acid – base balance, nerve and muscle contraction [19]. The magnesium content ranges between 0.146± 0.180mg/kg. lettuce had the highest magnesium content, which is in agreement with the previous work carried out by [7] of Lettuce. However, the levels obtained in this study is low to meet the recommended daily allowance of 400mg/day for men, 19 – 30 years old and 310mg/day for women 19 – 39 years old.

Iron is an important trace element in the human body, it plays crucial roles in haemopoietic, control of infection and cell mediated immunity [20]. The deficiency of iron has been described as the most prevalent nutritional deficiency and iron deficiency anemia is estimated to effect more than one billion people worldwide [21]. The consequences of iron deficiency include reduced work capacity, impairments in behaviour and intellectual performance and decrease resistance
to infection [22]. The level of zinc in all the leafy vegetable samples were moderate, the value obtained ranged between 0.394±0.011 and 2.802±0.012 and this value are similar to ones reported [16 & 23].

Zinc is an essential micronutrient for human growth and immune functions [24]. An estimated 20% of the world population is reported to be at risk of inadequate zinc in-take [25]. Studies on Nigeria shows that zinc deficiency affects 20% of children less than five years; 28.1% of mothers and 43.9% of pregnant women [22] a balance proportion of calcium is needed in the body. Deficiency of calcium balance result in osteoporosis, arthritis, pyorrhea, rickets and tooth decay.

4. CONCLUSION

The present study has shown that the leafy vegetables examined have moderate amounts of crude protein, crude fat, crude fibre and ash contents, the vegetables also contained good minerals with abundance of them in calcium, magnesium, sodium and potassium while they were least in nickel and manganese.

The result suggests that the vegetables if consume in sufficient amount would contribute greatly towards meeting human nutritional requirements for normal growth and adequate protection against diseases arising from malnutrition.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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