

Elemental composition, Mineral safety index, mineral bioavailability phytochemical and non-starch polysaccharides content of fourteen leafy vegetables consumed in Ekiti State, Nigeria

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Abstract

Fourteen leafy vegetables sold in Ekiti-State major markets, Nigeria were investigated for mineral composition, mineral safety index, mineral bioavailability, phytochemicals and non-starch polysaccharide contents using standard analytical procedures. The results (mg/kg) were: minerals: Na (17.8-47.4), K (146-435), Ca (20.8-53.0), Mg (99.0-283), Cu (0.015-0.083), Co (0.00012-0.03), Fe (2.6-8.07), Zn (0.671-1.52), Se (0.0001-0.002) and P (1.63-6.31). Mineral safety index showed that all the examined minerals were positive and would not overload the body of consumers. Phytochemical constituents (mg/100g) were (mg/100g): tannins (0.10- 0.390), phenolics (0.03-250), alkaloids (0.011-2.25), saponin (0.6-3.88), flavonoids (0.380-0.950), phytates (4.95-7.93), oxalates (0.230-0.590) and phytin phosphorous (1.39-2.23). [Ca]/[Phy], [Ca]/[Phy]/[Zn] and [Phy]/[Fe] were: 5.42-13.2, 0.17-1.32 and 0.63-1.73 respectively. Non-starch polysaccharide levels (%): NDF (4.65-11.2), ADF (3.33-9.31), lignin (1.40-4.26), cellulose (1.44-3.76) and hemicellulose (1.32-1.93). Significant differences existed among the Na, K, Ca, Mg, Ca/P, K/Na, Zn/Cu and milliequivalent ratio at $P = 0.05$ (Chi square, χ^2 test) across the fourteen

vegetable samples. Generally, they could be considered as potential sources of nutrients in human nutrition.

Keywords: Bioavailability, fibre fractions, phytochemicals, mineral safety index, vegetables

Introduction

One of the major challenge worldwide is malnutrition and the number of people suffering from chronic undernourishment and malnutrition has been on the increase daily (FAO, 2016). About 30 % of the world population suffers from some form of malnutrition and of these lot, over 2 billion suffer micronutrient deficiencies comprising about 52 % pregnant women and 39% children less than five years of age (FAO, 2016). Zinc, iron and other micro minerals deficiencies have been reported to be responsible for series of severe health problems which include impairments relating to physical growth, the immune system, learning abilities and high risks of infections mostly in the developing countries (WHO, 2002).

Gibson et al. (1997) reported that complementary foods based almost exclusively on plants are often major source of energy and nutrients from non-milk foods for many infants and young children living in resource-poor households in low-income countries and consumption of animal-source foods is often low due to economic or religion concern. These plant-based complementary diets are frequently associated with micro-nutrients deficits, especially iron, zinc and calcium (WHO, 2006), this has actually been impacted by poor bioavailability, especially when the diets are based on unrefined cereals and legumes owing to high phytate contents which may compromise the bioavailability of minerals such as Zn, Fe, Ca, and consequently affects growth, health and cognitive development in children (Rosalind et al., 2010).

Vegetables constitute the most important daily diets in many of the households globally (Kacholi and Sahu, 2018). Leafy vegetables consumption has been reported to increase in the urban community as they provide minerals, vitamins, antioxidants, lipids, amino acids, fibre and dietary nitrates (Yusuf et al., 2003; Ramulu and Udeyasekara, 2004; Govenjak and Cencic, 2013; Adeyeye et al., 2016; Adesina and Adeyeye, 2013). Dietary fibre has gained importance in the past two decade because of its beneficial roles in human nutrition. Fruits and vegetables contribute both soluble and insoluble fibres to human diets. Insoluble fibre including lignin, cellulose and hemicelluloses are found in the cell wall and the skin of vegetables while soluble fibres like, pectin, gums and mucilages are mostly found in the skin (Sarker and Rahman, 2017). Health benefits of

fibre-rich foods range from prevention and treatment of obesity, reduction of blood glucose and cholesterol levels, glycemic regulation and prevention of intestinal diseases like, constipation, hemorrhoids, diverticular disease and colon cancer (Marlett, et al., 2002). Howarth et al. (2001) had reported appreciable decrease in energy density and weight loss (10 % and 1.9 kg respectively) over 3.8 months as a result of 14g/day additional fibre consumption.

According to a 2007 World Health Report, unbalanced diets with low vegetable intake and low consumption of complex carbohydrates and dietary fibre are estimated to cause 2.7 million deaths each year and were among the top 10 risk factors contributing to mortality (Dias, 2011; 2012). Though the exact mechanism by which vegetable consumption reduces human diseases have not been fully understood, but the general consensus among physicians and nutritionists is that phytochemicals and other micro-nutrients in vegetables are responsible for mitigating many diseases (Dias, 2012). Some phytochemicals of vegetables are strong antioxidants and are thought to reduce the risks of chronic diseases by protecting against free radical damage by modifying metabolic activation and detoxification of carcinogens, or even influencing processes that alter the course of tumor cells (Graig and Beck, 1999; Wargovich, 2000; Mullie and Clarys, 2011).

Considering the fact that there are paucity of information on the mineral safety index, mineral bioavailability and non-starch polysaccharide content of the selected vegetables, hence, the need to investigate fourteen commonly sold and consumed leafy vegetables in Ekiti State for minerals contents, mineral ratios, mineral safety index, Zn, Ca, and Fe-phytate interrelationships, phytochemicals and insoluble fibre fractions (non-starch polysaccharides).

Experimental

Sample collection and treatment

Samples of fourteen fresh, healthy and disease free vegetables used for this research were purchased from ten different vendors from major markets in Ado-Ekiti and Iworoko-Ekiti, Ekiti State, Nigeria. The vegetables were identified at the herbarium section of the Department of Plant Science and Biotechnology, Ekiti State University, Nigeria. Edible parts of the plants were separated, thoroughly washed under running tap water, drained and air-dried at room temperature to a constant weight. The dried samples were then pulverized to powder using an electric stainless steel Excella-Mixer grinder (3 S.S. Jars Model, India). The powdered samples were stored in air-

tight plastic containers and refrigerated pending further chemical analysis. The details of the investigated vegetables, their names and sample identities were elaborated in Table 1.

Chemical Analysis

Elemental determination

The minerals were analyzed from the solution obtained by initially dry ashing the samples at 550°C. Filtered solutions were used to determine Na, K, Ca, Mg, Zn, Fe, Mn, Cu, Pb, Co, Cd, Ni and Se by means of atomic absorption spectrophotometer (Buck Scientific Model- 200A/210, Norwalk, Connecticut 06855). Phosphorus was determined colorimetrically by Spectronic 20 (Gallenkamp, UK) using the phosphovanado molybdate method (AOAC, 2006). All chemicals used were of British Drug House (BDH, London, UK) analytical grade. Earlier, the detection limits for the metals in aqueous solution had been determined using the methods of Varian Techtron Varian (1975). The optimal analytical range was 0.1-0.5 absorbance units with coefficients of variation from 0.9 % to 2.21 %. From the mineral elements determined, further calculations were made.

Colorimetric determination of phosphorus

Phosphorus was determined colorimetrically using a Spectronic 20 (Gallenkamp, London, UK) instrument, with KH_2PO_4 as a standard.

Mineral Ratios

Ratios of Ca/Mg, Na/K, Ca/P, Na/Mg, Zn/Cu, Ca/P and $[\text{K}/(\text{Ca} + \text{Mg})]$ were all calculated (Hatcock, 1985; Watts, 2010; ARL, 2012).

Mineral Safety Index (MSI)

The mineral safety index (MSI) (Hatcock, 1985) of Na, P, Ca, Fe, Se, Zn and Cu were calculated using the formula:

$$\text{Calculated MSI} = \frac{\text{MSI} \times \text{Research data result}}{\text{RAI}} \quad (1)$$

Where, MSI = mineral safety index Table (standard), RAI = recommended adult intake.

Phytochemical analysis

The phytochemical components of the vegetable samples were analyzed as follow:

Determination of phytic acid and phytin phosphorus

4 g of the sample was soaked in 100 mL 2 % HCl for 3 hours and then filtered. 25 mL of the filtrate was placed in a 100mL conical flask and 5 mL of 0.03 % NH_4SCN solution was added as indicator. 50 mL of distilled water was added to give it the proper acidity (pH 4.5). This was

titrated with ferric chloride solution which contained 0.005 mg of Fe per mL of FeCl_3 used until a brownish yellow colour persisted for 5 min. Phytin phosphorus (Pp) was determined and the phytic acid content was calculated by multiplying the value of Pp by 3.55 (Young and Greaves, 1940). Each milligram of iron is equivalent to 1.19 mg of Pp.

Determination of tannin

200 mg of the sample was weighed into a 50 mL sample bottle. 10 mL of 70 % aqueous acetone was added and properly covered. The bottles were put in an orbital shaker and shaken for 2 hours at 30°C. Each solution was then centrifuged and the supernatant stored in ice. 0.2 mL of each solution was pipetted into test tubes and 0.8 mL of distilled water was added. Standard tannic acid solutions were prepared from a 0.5 mg/mL stock and the solution made up to 1 mL with distilled water. 0.5 mL folin reagent was added to both sample and standard followed by 2.5 mL of 20 % Na_2CO_3 . The solutions were then vortexed and allowed to incubate for 40 minutes at room temperature after which absorbance were taken against a reagent blank concentration of the sample from a standard tannic acid curve (Makkar and Goodchild, 1996).

Determination of oxalate

1g of the sample was weighed into 100 mL conical flask. 75mL of 1.5 N NH_2SO_4 was added and the solution was carefully stirred intermittently with a magnetic stirrer for about 1 hour and then filtered using Whatman filter paper. 25mL of sample filtrate was collected and titrated hot (80-90°C) against 0.1 M KMnO_4 solution to the point when a faint pink colour appeared that persisted for at least 30 seconds (Day and Underwood, 1986).

Determination of alkaloid

Alkaloid determination was carried out following the procedure of Harborne (1973). 5.0 g of the sample was weighed into a 250 mL beaker and 200 mL of 10 % acetic acid in ethanol was added and covered and allowed to stand for 4 hrs. This was filtered and the extract was concentrated on a water bath to one quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The residue is the alkaloid which was dried and weighed.

Determination of saponin

The method used was that of Obadoni and Ochuko (2001). 5 g of the sample was put into a conical flask and 100 mL of 20 % aqueous ethanol were added. The sample was heated over a

hot water bath for 4h with continuous stirring at about 55 °C. The mixture was filtered and the residue re-extracted with another 200 mL 20 % ethanol. The combined extracts were reduced to 40mL over water bath at about 90 °C. The concentrate was transferred into a 250 mL separating funnel and 20 mL of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. 60 mL n-butanol was added. The combined n-butanol extracts were washed twice with 10 mL of 5 % aqueous sodium chloride. The remaining solution was heated in a water bath after evaporation; the sample was dried in the oven to a constant weight. The saponin content was calculated as percentage.

Determination of flavonoid

The method of Boham and Kocipai-Abyazan (1974) was followed in the determination of flavonoid. 5 g of the sample was extracted repeatedly with 100 mL of 80 % aqueous methanol at room temperature. The whole solution was filtered through whatman filter paper (125 mL). The filtrate was later transferred into a crucible and evaporated into dryness and weighed to a constant weight.

Calculation of the mole ratios

The [phytate]:[Zn], [Ca]:[phytate], [phytate]:[Fe] and [Ca][phytate]:[Zn] mole ratios were calculated as previously described (Watts et al., 1994; IZINCG, 2004).

$$[\text{Phytate}]:[\text{Zn}] = \frac{\text{phytate } (\frac{\text{mg}}{100\text{g}})/660}{\text{Zinc (mg/100)}/65.38} \quad (2)$$

$$[\text{Ca}]:[\text{Phytate}] = \frac{\text{calcium } (\frac{\text{mg}}{100\text{g}})/40.08}{\text{phytate } (\frac{\text{mg}}{100\text{g}})/660} \quad (3)$$

$$[\text{Phytate}]:[\text{Fe}] = \frac{\text{phytate } (\frac{\text{mg}}{100\text{g}})/660}{\text{Iron (mg/100)}/55.85} \quad (4)$$

$$[\text{Ca}][\text{phytate}]:[\text{Zn}] = \frac{(\text{phytate } (\frac{\text{mg}}{100\text{g}})/660) (\text{calcium } (\frac{\text{mg}}{100\text{g}})/40.08)}{\text{Zinc (mg/100)}/65.38} \quad (5)$$

Determination of fibre fractions

Determination of acid detergent fibre (ADF)

0.5 g of the sample was weighed into a Berzelius beaker. 50 mL cold Acid Detergent Solution (ADS) was added and boiled for 5-10 min. It was filtered on a previously weighed Gooch crucible. The residue was washed twice with hot water (90 – 100 °C) and washed repeatedly with acetone until it was colourless. The crucible together with the fibre was dried at 105 °C overnight and hot weighed and cooled (15 °C). 72 % H₂SO₄ was added and stirred with a glass rod to a smooth paste. The crucible was kept at 20-30 °C. The crucible was about half filled with tetraoxosulphate (VI) acid and stirred. The glass rod still remaining in the crucible, it was refilled with 72 % H₂SO₄ and stirred at hourly intervals as acid drained away. After 3 h, it was filtered to drain as much acid as possible using a vacuum pump. The content was washed with hot water until it was free from acid (by testing with litmus paper), rinsed and the stirring rod removed. The crucible was dried and hot weighed. The crucible was ignited in a muffle furnace at 550 °C for 3 hrs, cooled to 105 °C and hot weighed. ADF - Ash was reported as the difference between last weight and the original weight of the crucible (Van Soest and Robertson, 1980).

Determination of neutral detergent fibre (NDF)

0.5 g of the sample was weighed into a 600 mL Berzelius beaker. 50 mL cold neutral detergent solution (NDS) was added and boiled on a refluxing unit. The heat was adjusted in the process to even boiling and avoiding foaming, keeping the sample particles suspended. It was refluxed for 60 minutes from onset of boiling. It was filtered to a previously weighed crucible using light suction. The residue was washed twice with hot water, twice with acetone and dried using suction. Acetone washing was continued until a clear solution was obtained. The lumps were broken so that the solvent could come in contact with all particles of fibre. The crucible was dried in air for 10-15 minutes (to drain part of the acetone) and oven dried for 8 hours at 105 °C. It was hot weighed to obtain yield of cell wall. The crucible was ashed at 500 °C for 8 hours, removed from furnace, put in oven (set at 100 °C) and hot weighed. The loss in weight was the ash free cell wall (Van Soest and Robertson, 1980). Lignin (%) = $\frac{\text{weight after oven drying} - \text{weight after furnace ashing}}{\text{weight of sample}} \times 100$.

Determination of cellulose

2.0 g of the sample was weighed into a 250 mL conical flask and 2 drops of paraffin oil, 25 mL of 8 % (v/v) acetic acid, 1L of concentrated nitric acid and 4 glass beads were added. It was refluxed for 20 min on a refluxing apparatus. The digest was washed into 500 mL centrifuge tube

with hot 95 % ethanol and centrifuged at 1800 r. p. m. for 5 minutes. The liquid was decanted and 95 % ethanol was added, stirred and centrifuged for another 5 minutes. Liquid was decanted and sample washed twice with 95 % ethanol, hot benzene and once with petroleum ether into 25 mL crucible. The crucible was placed in the oven at 100 °C for 1 hour and later placed in the desiccator to cool and it was then weighed. The crucible was later placed in the furnace at a temperature of 600 °C for 4 hours, cooled in a desiccator and weighed for ash content. The base weight less ash weight is the weight of cellulose (Usoro et al., 1982).

Determination of hemicellulose

2 g of sample was weighed into two different conical flasks (A and B). 5 % (v/v) KOH was added to the sample in flask A while 24 % (v/v) KOH was added to the sample in flask B and both samples were allowed to stand for 2 hours. The suspension was filtered using filter paper, washed with additional KOH solution and the filtrate was received into 2 different flasks (A and B) containing excess of glacial acetic acid. The hemicellulose was then quantitatively precipitated by the addition of ethanol. The precipitated hemicelluloses was isolated by centrifuging for 10 minutes and was washed with ethanol and filtered. The samples were oven dried for 2 hours at 80 °C and transferred into a desiccator and allowed to cool. Percentage hemicellulose was then calculated (Usoro et al., 1982).

Statistical Analysis

Descriptive statistics (mean, standard deviation and coefficient of variation) (Chase, 1976) were determined and all the data were subjected to Chi-square (χ^2) test to determine significant differences among the results (Oloyo, 2001).

Results and discussion

Mineral compositions (mg/kg) of the analyzed vegetables were presented in Table 2. Minerals analyzed include: Na, K, Ca, Mg, Cu, Co, Fe, Zn, Se, and P. In all the fourteen vegetables investigated, these minerals were present in varying concentrations. The range of the contents, mean and CV % were as follow: Na (17.8 – 47.4, 28.5, 34.2), K (146 – 435, 301, 34.8), Ca (20.8 – 61.0, 40.2, 31.4), Mg (92.9 – 283, 151, 38.9), Cu (0.015 – 0.092, 0.056, 48.2), Co (0.00012 – 0.003, 0.001, 68.1), Fe (2.60 – 8.27, 4.69, 35.9), Zn (0.558 – 1.52, 0.893, 39.5), Se (0.0001 – 0.003, 0.001, 87.1) and P (1.63 – 6.31, 3.43, 37.2). Na was of highest concentration in *Vernonia*

aamygdalia (47.4 mg/kg) and least in *Launaea taraxacifolia* (17.8 mg/kg), K was highest in *Celosia agentea* (401 mg/kg) and of lowest concentration in *Gnetum africanum* (146 mg/kg), *Celosia agentea* also had the highest concentration of Fe (8.07 mg/kg) and Se (0.002 mg/kg) whereas the least concentrations of Fe and Se occurred in *Amaranthus spinosis* (2.60 mg/kg) and in *Amaranthus viridis* (0.0001 mg/kg). The highest concentration of Ca was observed in *Solanecio bialfrae* (61.0 mg/kg) and lowest in *Amaranthus spinosis* (20.8 mg/kg), Mg was most concentrated in *Ocimum gratissimum* (283 mg/kg) and of least in *Asytasia gangetica* (92.9 mg/kg). Phosphorus had the highest level in *Amaranthus caudatus* (6.31 mg/kg) and the lowest value in *Cnidoscollous aconitifolius* (1.63 mg/kg). The levels of Cu, Co and Se in all the fourteen sampled vegetables were lower than 1.00 mg/kg.

Minerals are inorganic nutrients usually required in small quantities from less than 1.00 to 2500 mg per day depending on the mineral. Just like vitamins and other essential food nutrients, mineral requirements vary with age (Soetan et al., 2010). Nutrient minerals, being elements cannot be synthesized biochemically by living organisms, plants get minerals from soil and most of the minerals in human diets come from eating plants and animals or drinking water (MIC, 2016). They are present in all body tissues and fluids and their presence is necessary for the maintenance of certain physicochemical processes which are essential to life (Soetan et al., 2010; Berdamier et al., 2016).

The levels (mg/kg) of Na and K in the present report across the fourteen vegetables ranged from 17.8 – 47.4 and 146 – 435 respectively. Sodium and potassium are important extracellular and intracellular cations respectively and are involved in the regulation of plasma volume, acid-base balance, nerve and muscle contraction (Araseretnam et al., 2018), Soetan et al., 2010). The values of sodium and potassium in the present report were comparably higher than those reported for *P. latifolia*, *C. halicacabium*, *M. pentaphylla*, *D. elata*, *A. pomacea* and *P. grandis* (13.1 – 19.0 and 10.1 – 12.7 mg/kg) respectively (Araseretnam et al., 2018); *Grewia sapida* (Islary et al., 2016); *Alternanthera sessilis* (Gotruvalli et al., 2016); Whole seeds, testa and dehulled seeds of bambara groundnut (Olaleye et al., 2013). Increased level of sodium in the serum is called hypernatraemia and this occurs in cushion's disease, administration of adrenocorticotrophic hormone, administration of sex hormones, diabetes insipidus and after active sweating (Malhotra, 1998). Excessive intake of sodium may cause hypertension in susceptible individuals and increased level

of potassium may bring Addison's disease, advanced chronic renal failure and dehydration (Soetan et al., 2010).

Calcium functions as a constituent of bones and teeth (Adeyeye et al., 2020; Maqsood et al., 2019). In blood coagulation, calcium activates the conversion of prothrombin to thrombin and also takes part in milk clotting (Soetan et al., 2010). It plays a vital role in enzyme activation, example is the activation of enzymes such as adenosine triphosphatase (ATPase), succinic dehydrogenase etc. The levels of Ca in the present report (20.8 – 53.0 mg/kg) would definitely contribute significantly in achieving the daily requirements in human diets. However, the levels compared favourably with those of *Sagittaria trifolia* (Maqsood et al., 2019), Kikishi (Adeyeye et al., 2020) but lower than the values observed in bambara groundnut samples (35.2 – 82.2 mg/100g) (Olaleye et al., 2013).

In children, calcium deficiency causes rickets due to insufficient calcification by calcium phosphate of the bones during growth, in adults; it causes osteomalacia, a generalized demineralization of bones. It may also contribute to osteoporosis (Malhotra, 1998). The levels of calcium in these 14 vegetables could actually support the meeting of daily requirements of Ca by all age groups and this in turn will circumvent the effect of its deficiencies in human nutrition.

Magnesium is widely distributed in plants and animal foods. Geochemical and other environmental variable rarely have a major influence on its contents in foods. Most green vegetables, legumes, seeds, peas, beans and nuts are rich in magnesium as are some shell fish, spices and soya flour all of which usually contain more than 500mg/kg fresh weight (Mohammed and Sharif, 2011). According to FAO/WHO (2002), soft tissue magnesium functions as a co-factor of many enzymes involved in energy metabolism, protein synthesis, RNA and DNA synthesis and maintenance of the electrical potential of nervous tissues and cell membranes. Of particular importance with respect to the pathological effects of magnesium deficiency is the role of this element in regulating potassium fluxes and its involvement in the metabolism of calcium (FAO/WHO, 2002; Soetan et al., 2010).

As shown in Table 2, all the fourteen vegetables samples analyzed contained very good amount of magnesium (92.9-28.3 mg/kg). These values were comparably higher than those

reported for *Sagiharia trifolia* (<1.0 mg/100g) (Maqsood et al., 2019), Kilishi (Adeyeye et al., 2020), *Fiscus capensis* leaves (1.92 %) (Achi et al., 2017).

Copper is a constituent of enzymes like cytochrome c. oxidase, amine oxidase, catalase, peroxidase, ascorbic acid oxidase etc. and it plays a vital role in iron absorption (Chandra, 1990). It is an essential micro nutrient necessary for the haematologic and neurologic systems and its deficiency has been associated with cardiac hypertrophy and sudden cardiac failure (Tan et al., 2006, Murray et al., 2000). Although the levels of copper in the vegetables investigated were fairly low (0.015- 0.092 mg/kg) as presented in Table 2, however, the values were comparably higher than those reported for some leafy vegetables consumed in Kano, Nigeria (Mohammed and Sherif, 2011) but lower than the levels in two temperate *Astragalus* species: *A. glycyphyllos* (0.272 – 0.596 mg/100g) and *A. cicer* (0.328 - 0.548 mg/100g) (Batkute et al., 2018).

Iron (Fe) is important in the diet, especially for pregnant and nursing mothers as well as infants. It is also needed by the convalescents and the elderly to reduce cases of diseases associated with deficiency of iron such as anemia (D' Millo, 2003; Soetan, 2010). In Table 2, the highest iron content was in *Celosia agentea* leaves (8.07mg/kg) with other samples contained between 2.60 and 6.95 mg/kg). The levels of Fe in the vegetables were high enough to support good health (FAO/WHO, 2002). The observed levels in the current report were comparably higher than those reported for *C. tridens* (0.213mg/100g), *H. cannabinus* (0.260 mg/100g) (Mohammed and Sharif, 2011) and lower than those reported for *A. glycyphyllus* and *A. cicer* leaves (14.1 – 65.3 mg/100g and 6.78 - 30.2 mg/100g) respectively (Butkute et al., 2018). It is worthy to those that all the vegetables investigated in this report would simply meet the recommended daily allowance of Fe: 10-15 mg/day (FAO/WHO, 2002).

The levels of Zn, Co and Se in all the samples of vegetable analyzed were in the following ranges (mg/kg) Zn (0.558 - 1.52), Co (0.00012-0.003) and Se (0.0001- 0.002). These values, though, fairly low but fell within the range of values earlier reported for some leafy vegetables: Five leafy vegetables consumed in Kano (<0.3 mg/100g) (Mohammed and Sherif, 2011), *A. glycyphyllus* and *A. acer* (1.88-3.47mg/100g) (Butkute et al., 2018), *Ficus carpensis* (2.84mg/100g) (Achi et al., 2017); *Sagitharia trifolia* (Maqsood et al., 2019), Zn and Co in Bambara groundnut (ND-11.2mg/100g) (Olaleye et al., 2013); *Grewia sapida* (1.318 mg/100g) (Islary et al., 2016).

Zinc is important for nerve function and male fertility. It is important for normal sexual development, especially for the development of testes and ovaries and for reproduction (Ayoola et al., 2010), healthy functioning of the heart and normal growth (Elizabeth, 1994). Adequate and regular inclusion of these vegetables in human diets would assist in preventing the adverse effect of zinc deficiency which include stunted growth and delayed sexual maturation because of its role in nucleic acid metabolism and protein synthesis (Barminas et al., 1998; Zoroddu et al., 2019). Cobalt is required as a constituent of vitamin B₁₂ and its metabolism is the same as for vitamin B₁₂. In addition to its role in Vit B₁₂, cobalt is a co-factor of enzymes involved in DNA biosynthesis and amino acid metabolism (Arinola et al., 2008). Deficiency disease is manifested in Vit B₁₂ deficiency (FAO/WHO, 2002, Zoroddu et al., 2019). To this end, it is opined that regular consumption of these vegetable would help in alleviating Vit B₁₂ deficiency diseases.

Selenium (Se) on the other hand is a constituent of glutathione peroxidase (Murray et al., 2000) and a constituent element of the defense system that protects the living organisms from harmful action of free radicals. Organic and inorganic selenium compounds function in preventing certain disease conditions that have in the past been associated with vitamin E deficiency (Soetan et al., 2010). Consuming these vegetables as part of daily diets by humans would readily assist in attaining the daily requirements of Se for all categories of people (from children to adults (Males, Females: pregnant and lactating mothers) (15-70 µg/day) (DRI, 1997).

Table 3 depicted the results of computed mineral ratios of the selected analyzed vegetables. The mineral ratios are often more important than the individual mineral levels themselves and this had been illustrated by the following statements by Vitale *et al* as reported by Adeyeye et al. (2020). Knowing the individual levels of each mineral in foodstuff is not as important as the determination of the nutritional interrelationships although, dietary significance is better explained from the individual elemental composition vis-à-vis the effects of their excesses and deficiencies (Watts, 2010). In the present report, important ratio computed includes Ca/Mg, Ca/P, Na/K, Na/Mg, K/Na, Zn/Cu and milliequivalent ratio of Magnesium (K/Ca+Mg) and the results were in the following ranges: Ca/Mg (0.179-0.506), Ca/P (3.74-30.5), Na/K (0.042-0.230), Na/Mg (0.117-0.391), K/Na (4.34-24.0), Na/Mg (0.117-0.391), K/Na (4.34-24.0), Zn/Cu (8.16-101) and ME* (1.66-6.70) with CV % ranging from 32.3-109. From the CV% values, it is evident that the ratios

were closely varied among the results of Ca/Mg, Ca/P, Na/K, Na/Mg, K/Na and ME* but widely varied for the Zn/Cu among all the fourteen vegetable samples with 109% of CV.

Na/K ratios in the samples (0.042 – 0.230) were good as they were lower than the 0.6 requirement to avoid high blood pressure (Adeyeye and Adamu, 2006). The K/Na ratios were high at 4.34 – 24.0, meaning that more sodium may be required to maintain the 1:1 ratio. K/Na is implicated in the enhancement of salt balance of the body fluid (Adeyeye, 2011).

Table 3 also contained the available reference balance ideal (RBI) and acceptable ideal range (AIR) for some selected ratios (Adeyeye et al., 2020). From the results, it was observed that for Ca/Mg ratios, none among the vegetable had its value within the acceptable ideal range of 3.0-11.0, only *Amaranthus spinosis* leaves had its Ca/P ration agreed with the 1.5-3.60 acceptable ideal range, other vegetables had values well above the green range of ideal. Na/K and Na/Mg had values less than both the reference balance ideal and acceptable ideal range above the given references. The implication of these is that for a better nutritional advantage, these vegetables might require supplementation or consumption of foodstuff rich in the minerals alongside them which would adequately complement the mineral sources and availability especially P, Mg, and K. The Milliequivalent ratios of K, Ca and Mg agreed perfectly with the required reference balance ideal (2.2). This showed that adequate and regular consumption of these vegetables or their inclusion in daily diets would prevent deficiency symptoms such as hypomagnesaemia (Watts, 2010; Adeyeye et al., 2020).

The mineral safety index (MSI) values of the analyzed vegetables were shown in Table 4. For easy verification and understanding of the calculations the following information were provided: recommended adult intake (RAI), standard/table value (TV) of the MSI and the calculated value (cV) of the selected vegetables. The MSI values represented in Table 4 were for those elements whose standard comparisons were available from literatures. For better understanding of the explanation, let us take as an example, the recommended adult intake of P is 1200mg, its minimum toxic dose is 1200 mg or 10 times the recommended daily average (RDA). The same explanation holds for all other elements in the table. For all the vegetables analyzed, none of the minerals had negative difference (MSI standard – MSI calculated), these showed that none of the minerals could constitute mineral overload or become toxic to the sample consumers.

The phytochemical/anti-nutrient constituents of the fourteen vegetables were shown in Table 5. The results (mg/100g) were in the following ranges: tannin (0.100-0.390), phenolics (0.05-0.25), alkaloids (0.11-2.25), saponin (0.6-3.88), flavonoids (0.33-0.950), oxalate (0.230-0.590), phytates (4.95-7.93) and phytin phosphorus (1.39-2.23) with CV% values between 18.2 and 130. Results of alkaloids across all the fourteen vegetables were widely varied at 130% coefficient of variation whereas the values of tannin, phenol, saponin, flavonoids, phytates oxalates and phytin phosphorus had closely varied levels across all the vegetables as shown in their CV% values of 16.5 to 44.2.

The presence of tannins (0.100-0.39 mg/100g) in the vegetables analyzed confers them to be good plant sources for the treatment of wounds emanating from disease condition such as varicose ulcers and hemorrhoids (Njoku and Akumufula, 2007). According to Saxena et al. (2013), plants that contain tannins are used as astringents against diarrhea, as diuretics against stomach and duodenal tumors. However, the levels of tannins obtained on the present report were comparably lower than those reported for *Ficus capensis* leaves (687 mg/100g) (Achi et al., 2017) but fell within the values reported for testa, dehulled and whole seed samples of Bambara groundnut (0.09-0.84 mg/100g), (Olaleye et al., 2013) (0.35-0.85 mg/100g) (Adeyeye, 2011), *Canavalia ensiformis* and *Mucuna puriens* (0.87-7.8 mg/100g) (Agbede and Aletor, 2005). Tannins have been reported to bring about their anti-nutritional influences (especially in the monogastric animals) largely by precipitating dietary proteins and digestive enzymes to form complexes which are not readily digestible (Aletor, 1993). Saponins can either be beneficial or deleterious. There are suggestions that saponin consumption be encouraged because of their hypocholesterolemic activity, forage saponins have been reported by Cheeke et al. (1978) to cause toxic and anorexia effects in the rats and swine thereby limiting the feeding value of high saponin animal feeds such as alfalfa. Alkaloids have been reported to have analgesic properties.

Flavonoids in plants possess medicinal benefits which includes antioxidant and anti-inflammatory activities (Saxena et al., 2013). They have ability to scavenge hydroxyl radicals, super oxide anions and lipid peroxy radicals (Okwu and Josiah, 2006). The presence of flavonoids in the considered vegetables could be of help to the consumers against diseases such as cancer, inflammation and atherosclerosis (Onyeka and Nwambekwe, 2007). While the levels of flavonoids observed in the report was comparably lower than that of *Ficus capensis* (1367 mg/100g) (Achi et al.,

2017), it agreed well with those reported for various anatomical parts of bambara groundnuts (0.34-0.79mg/100g) (Olaleye et al., 2013) and for raw and processed *Treculia africana* seeds (Adesina and Adeyeye, 2015).

The presence of alkaloids in the present samples of vegetables could encourage their antibacterial activities. Alkaloids have been reported to possess various pharmacological activities such as antihypertensive effects, antiarrhythmic effect, anti-malaria and anticancer activities (Saxene et al., 2013). As an important dietary supplements and are known to exhibit antimicrobial activities and protect plants from microbial pathogens (Szczekowski et al., 1998), they could be beneficial in modulating blood lipids, lower cancer risks and improve blood glucose response as well as antioxidant activities (Igidi and Edene, 2014). Leaves of plants such as *Ficus capensis* have been reported to possess antimicrobial activities linked to the presence of saponins on them (Ogundare and Akinyemi, 2013; Igwe et al., 2016). The levels of saponins in the present report were however, lower than that of *Ficus capensis* (Achi et al., 2017) but agreed well with those reported for Bambara groundnut samples (1.01-1.38 mg/100g) (Olaleye et al., 2013).

Phytates and oxalates are antinutritional factors capable of interfering with nutrient bioavailability and utilization (Wyatt and Triana-Tejas, 1994; Adesina and Adeyeye, 2015; Adeyeye, 2011). In the present samples, phytates and oxalate levels (mg/100g) were generally low (4.95-7.93 and 0.23-0.59 respectively). Phytate levels in the present report fell below those reported for 13 spices obtained in Nigeria (390-6210 mg/100g) (Adeyeye and Fagbohun, 2005), seven varieties of Nigerian garden egg fruits (507-2788 mg/100g) (Adeyeye and Fagbohun, 2006), Many Nigerian foods such as legumes (14-344 mg/100g) and cereals (112-287 mg/100g) (Adeyeye et al., 2000) but were within the ranges reported for *Canavalia ensiformis* and *Mucuna pruriens* seed flours (6.0-15 mg/100g) (Agbede and Aletor, 2005). The presence of oxalate negatively affects the absorption and utilization of calcium. Oxalates combines with calcium to form calcium oxalate which passes through the intestine without being absorbed (Olaleye et al., 2013). Calcium oxalate is responsible for most of the kidney stone formation. Formation of these stones frequently reflects chronic alkalinity of the bladder and renal pelvic urine caused by infection with bacteria and hydrolyse urea, releasing ammonia (White et al., 1973). Of note is that the levels of oxalate in the fourteen vegetables were generally low to cause any disease condition or interference with calcium absorption and utilization. The phytin phosphorous (pp) levels (1.39-

223 %) recorded for the vegetables in the present report fell below 9.43-10.7 mg/100g) observed earlier for groundnut flours (Adeyeye, 2011) but higher than those reported for 17 wild leguminous crop seeds (Balogun and Fetuga, 1986) and *Mucuna pruriens* (Agbede and Aletor, 2005). Phytin phosphorus as % of P represents how much P is bond to Pp and abnormal change in levels will affect the utilization of divalent minerals and also will render some essential amino acids unavailable (Olaleye et al., 2013).

Calcium-, zinc-, iron- and phytate interrelationships in the fourteen vegetables were shown in Table 6. The values were in ranges as follows: [Phy]:[Zn] (3.23-11.1), [Ca]:[Phy] (5.42-19.7), [Ca][phy]:[Zn] (0.42-1.32) and [Phy]:[Fe] (0.63-1.73) with CV% values ranging from 27.1 to 45.3. These CV% values showed that the results were closely varied. It has been reported in literatures that food with a molar ratio [Phy]:[Zn] less than 10 showed adequate availability of Zn and when the ratio is greater than 15, there were problems (Oberleas and Horlard, 1981). [Phy]:[Zn] molar ratios of 15:1 had been reported to bring about reduced zinc bioavailability (Turnland et al., 1984). Also, WHO (1996) described phytate: Zinc molar ration as an index of zinc bioavailability, a view which was established by Gargari et al (2007). According to WHO (1996), the level of [Phy]:[Zn] in the present report (4.07-11.1) indicated a moderates Zn bioavailability, except *Amaranthus spinosis* leaves at 3.23.

However, the levels of [Phy]:[Zn] observed in the present report (3.23 – 11.1) compared favourably with the values reported for dehulled and whole seed flour of Bambara groundnut (Olaleye et al., 2013), some Nigeria food samples (Adeyeye et al., 2000), four lesser known African seeds (*Citrullus colocynthis*), *Cucumeropsis edulis*, *Ricinus communis* and *Prosopis africana* (3.35-11.81) (Igwe et al., 2013).

A [Ca]:[Phy] molar ratio lower than 6:1 makes phytate precipitation incomplete so that some of the dietary Zn remain in solution. The proportion remaining in solution increases with decreasing [Ca]:[Phy] molar ratio (Wise, 1983). From the forth going, Ca has a sparing effect on Zn and at critical [Ca]:[Phy] molar rations of >6:1, phytate is completely precipitated from the solution. Interestingly all the vegetables analyzed had their [Ca]:[Phy] ratios greater than 6:1 except *Amaranthus blitum* leaves. Thus Zn is available in solution and for absorption (Ojiako et al., 2010). The implication of this is that both the solubility of phytate and Zn availability in the intestine is dependent on dietary calcium levels (Ellis et al., 1987). It was this ascertainment that led to

the idea that $[Ca]:[Phy]:[Zn]$ is a better indicator of Zn bioavailability than either of $[Ca]:[Phy]$ or $[Phy]:[Zn]$ and noted that if the value is greater than 0.5 mol kg^{-1} , then there would be inference with Zn availability. In the present report, the following vegetables had their $[Ca]:[Phy]$ values agreed with the critical value of 0.5 mol kg^{-1} *Launaea taraxacifolia* (0.54), *Amaranthus spinosis* (0.17), *Amaranthus caudatus* (0.54), *Amaranthus blitum* (0.42), *Basella rubra* (0.49) and *Talinum triangulare* (0.57) while others had their ratios fairly above the critical value at $0.66\text{--}1.32 \text{ mol kg}^{-1}$. Besides the $[Phy]:[Zn]$, also calculated was the $[Phy]:[Zn]$ ratio for all the vegetables. Interestingly, the values obtained were low at $0.63\text{--}1.23$ showing that Fe in the samples would also be moderately available. However, these values were comparably lower than those reported for grains obtained from full and reduced irrigation processes ($12.1\text{--}20.4$) (Magallenes-Lopez et al., 2017), durum wheat grains ($15.5\text{--}31.3$) (Salunke et al., 2014) and two bread wheat whole-meal flour samples (12.0) (Ealing et al., 2014).

The non-starch polysaccharide (WSP) contents (%) of the fourteen vegetables analyzed were shown in Table 7. In all the fourteen vegetables, the results are: neutral detergent fibre (NDF) ($4.65\text{--}11.2$), acid detergent lignin (ADL) ($1.90\text{--}4.26$), cellulose (CEL) ($1.44\text{--}3.76$) and hemicellulose (HMC) ($1.32\text{--}1.93$) and CV% values ranged between 13.9 and 42.8 indicating that the values were not widely varied among the vegetables.

The results obtained in the present report for all forms of fibre fractions in the fourteen vegetables compared favourably with the values (%) recorded for dehulled and whole seeds flour of bambara groundnuts ($1.36\text{--}7.13$ and $0.84\text{--}8.16$ respectively) (Olaleye et al., 2013), also with respect to ADL and NDF; wild and cultivated rice (Lowry et al., 1994). Acid detergent fibre has been described as a portion of the plant fibre (Van soest and Robertson, 1980). Other components include the cellulose, and lignin from cell wall and variable of xylans and others. Neutral detergent fibre is considered to be the entire fibre fraction of the feedstuff, but it is known to underestimate cell wall concentration because most of the pectic substances in the cell wall are solubilized (Van Soest and Robertson, 1994). On this basis, NDF is considered a poor estimate of cell wall content ratios for the pectin-rich legumes.

Summary of the statistical (Chi-square (χ^2)) analysis of all the results were shown in Table 8. The numerous variations observed among the results obtained for the fourteen vegetables could possibly be attributed to locations and environmental factors soil, water and farming practices

available for their production. The following parameters had their values significantly different at $P=0.05$; Na, K, Ca, Mg, Ca/P, K/Na, Zn/Cu, ME*. However, the results of Cu, Co, Fe, Zn, Se, P, phytochemicals, mineral bioavailability and non-starch polysaccharides showed no significant differences at $p=0.05$, $v=n-1=13$.

Conclusion

The study assuredly demonstrated the potential of plant materials from fourteen leafy vegetables sold and commonly consumed in Ekiti State as a valuable sources of essential minerals (Fe, Zn, Ca, Mg, P, K, Na, Cu and Co) capable of alleviating micronutrient malnutrition, phytochemicals such as flavonoids, alkaloids, tannins and dietary fibre fractions. Furthermore, the research has also shown the basic interaction of phytate with certain minerals such as Zn, Ca, and Fe as it affects their bioavailability. The investigated plant materials may be considered as potential source of dietary fibre fractions. The [Phy]:[Zn], [Ca]:[Phy], [Ca][Phy]:[Zn] and [Phy]:[Fe] could also be used to make the right choice of plant based products rich in the desired minerals to overcome deficiencies in population groups suffering the hidden hunger related issues of micronutrients bioavailability.

Conflict of interest

The authors declare that there is no conflict of interest.

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Table 1: Description of the studied vegetables sold in popular markets in Ado-Ekiti and Iworoko-Ekiti, Ekiti State, Nigeria

SN	Local Name	Common Name	Scientific Name	Sample ID	Edible part
1	Yanrin	Wild lettuce	<i>Launaea taraxacifolia</i>	A	Leaf
2	Eru, Okazi, Afang	Wild spinach	<i>Gnetum africanum</i>	B	Leaf
3	Peke	Chinese yellow	<i>Asytasia gangetica</i>	C	Leaf
4	Tete elegun	Spiny amaranth, spiny pigweed	<i>Amarathus spinosis</i>	D	Leaf
5	Tete Abalaye	Slender amaranth, green amaranth	<i>Amaranthus viridis</i>	E	Leaf
6	Tete arowojeja pupa	Pendant amaranth, foxtail amaranth	<i>Amaranthus caudatus</i>	F	Leaf
7	Efo Iyana-Ipaja	Tree spinach	<i>Cnidoscollous aconitifolius</i>	G	Leaf
8	Tete Elewe wewe	Purple amaranth	<i>Amaranthus blitum</i> L.	H	Leaf
9	Egbeje, Elegede	Pumpkin leaf, Squash	<i>Cucurbita maxima</i>	I	Leaf
10	Sokoyokoto	Lagos spinach, green	<i>Celosia agentea</i>	J	Leaf
11	Efinrin	Scent leaf, Nchanwu, Daidoya	<i>Ocimum gratissimum</i>	K	Leaf
12	Amunututu, Alaari	Malaba spinach, Vine spinach, Ceylon spinach	<i>Basella rubra</i>	L	Leaf
13	Worowo	Sierra leone Boloji	<i>Solanecio biafrae</i>	M	Leaf
14	Gure, Gbure	Water leaf	<i>Talinum triangulare</i>	N	Leaf

Table 2: Mineral compositions (mg/kg) of the selected leafy vegetables analyzed

Minerals	A	B	C	D	E	F	G	H	I	J	K	L	M
Na	17.8	26.1	20.5	18.1	35.5	24.2	29.7	47.4	41	23.5	19.5	38.1	20
K	225	146	188	435	380	167	292	398	178	401	343	410	38
Ca	28.9	45.3	31.3	20.8	40.7	25.8	49.7	22.6	53.0	45.3	50.6	37.5	61
Mg	105	114	92.9	109	141	99.0	253	121	161	170	283	139	12
Cu	0.092	0.015	0.083	0.015	0.037	0.046	0.064	0.042	0.073	0.081	0.051	0.08	0.0
Co	0.0006	0.0002	0.002	0.0002	0.00012	0.001	0.001	0.0021	0.003	0.002	0.00212	0.001	0.0
Fe	4.02	2.86	3.05	2.60	3.11	3.26	6.01	5.14	5.11	8.07	6.95	5.15	4.3
Zn	0.771	0.579	0.677	1.52	0.546	0.623	0.558	0.88	0.716	1.02	0.995	1.47	0.6
Se	0.001	0.0012	0.0001	0.002	0.0001	0.00013	0.002	0.00021	0.001	0.002	0.00013	0.001	0.00
P	2.42	2.69	4.35	5.57	2.85	6.31	1.63	2.72	3.24	2.52	3.05	4.14	3.5

Table 3: Computed mineral ratios of the selected leafy vegetables

Ratios	A	B	C	D	E	F	G	H	I	J	K	L	M	N	Mean
Ca/Mg	0.275	0.397	0.337	0.191	0.289	0.261	0.196	0.186	0.329	0.267	0.179	0.271	0.506	0.242	0.28
Ca/P	12.0	16.8	7.20	3.74	14.3	4.09	30.5	8.32	16.4	17.9	16.6	9.05	17.4	17.3	13.7
Na/K	0.079	0.179	0.109	0.042	0.093	0.145	0.102	0.119	0.230	0.059	0.057	0.093	0.052	0.147	0.11
Na/Mg	0.170	0.229	0.221	0.166	0.252	0.244	0.117	0.391	0.255	0.139	0.069	0.275	0.166	0.181	0.21
K/Na	12.6	5.59	9.17	24.0	10.7	6.90	9.83	8.40	4.34	17.1	17.6	10.8	19.4	6.82	11.7
Zn/Cu	8.38	38.6	8.16	101	14.8	13.5	8.72	21.0	9.81	12.6	19.5	18.4	8.83	69.6	25.2
ME*	3.36	1.83	3.03	6.70	4.18	2.68	1.93	5.54	1.66	3.73	2.06	4.66	4.27	1.99	3.40

ME*= milliequivalent ratio of $[k/(Ca+Mg)]$, RBI= reference balance ideal, AIR = acceptable ideal range

Table 4: Computed mineral safety index (MSI) of selected minerals from leafy vegetables analyzed.

		A	B	C	D	E	F	G	H	I	J	K
Fe	TV	6.7	6.7	6.7	6.7	6.7	6.7	6.7	6.7	6.7	6.7	6.7
	cV	1.80	1.28	1.36	1.16	1.39	1.46	2.69	2.30	2.28	3.61	3.10
	D	4.90	5.42	5.34	5.54	5.31	5.24	4.01	4.40	4.42	3.09	3.60
Na	TV	4.8	4.8	4.8	4.8	4.8	4.8	4.8	4.8	4.8	4.8	4.8
	cV	0.17	0.25	0.20	0.17	0.34	0.23	0.29	0.46	0.39	0.23	0.19
	D	4.63	4.55	4.60	4.63	4.46	4.57	4.51	4.34	4.41	4.57	4.61
Ca	TV	10	10	10	10	10	10	10	10	10	10	10
	cV	0.241	0.377	0.261	0.173	0.339	0.215	0.414	0.188	0.442	0.377	0.421
	D	9.76	9.62	9.74	9.83	9.66	9.79	9.59	9.81	9.56	9.62	9.58
P	TV	10	10	10	10	10	10	10	10	10	10	10
	cV	0.020	0.022	0.036	0.046	0.024	0.053	0.014	0.023	0.027	0.021	0.025
	D	9.98	9.98	9.96	9.95	9.98	9.95	9.99	9.98	9.97	9.98	9.97
Zn	TV	33	33	33	33	33	33	33	33	33	33	33
	cV	1.70	1.27	1.49	3.34	1.20	1.37	1.23	1.94	1.58	2.24	2.19
	D	31.3	31.7	31.5	29.7	31.8	31.6	31.8	31.1	31.4	30.8	30.8
Cu	TV	33	33	33	33	33	33	33	33	33	33	33
	cV	0.138	0.023	0.1245	0.0225	0.0555	0.069	0.096	0.063	0.110	0.122	0.0765
	D	32.9	33.0	32.9	33.0	32.9	32.9	32.9	32.9	32.9	32.9	32.9
Mg	TV	15	15	15	15	15	15	15	15	15	15	15
	cV	3.94	4.28	3.48	4.09	5.29	3.71	9.49	4.55	6.04	6.36	10.6
	D	11.1	10.7	11.5	10.9	9.71	11.3	5.51	10.5	8.96	8.64	4.40
Se	TV	14	14	14	14	14	14	14	14	14	14	14
	cV	0.2	0.24	0.024	0.4	0.02	0.026	0.4	0.042	0.2	0.4	0.026
	D	13.8	13.8	13.98	13.6	13.98	13.97	13.6	13.96	13.8	13.6	13.97

TV= table value, cV= calculate value, D= TV-cV

Table 5: mg/100g levels of phytochemicals present in the analyzed leafy vegetables

Phytochemical	A	B	C	D	E	F	G	H	I	J	K	L	M
Tannin	0.27	0.27	0.27	0.02	0.17	0.22	0.28	0.37	0.100	0.35	0.39	0.36	0.29
Phenol	0.14	0.13	0.13	0.14	0.05	0.13	0.17	0.23	0.06	0.23	0.25	0.20	0.15
Alkaloids	0.21	0.20	0.15	0.17	0.11	0.25	0.15	2.21	0.03	2.18	2.25	2.20	0.21
Saponin	1.85	1.88	1.90	0.60	1.38	1.80	2.10	3.62	2.00	3.50	3.88	3.85	2.30
Flavonoids	0.53	0.56	0.58	0.35	0.56	0.6	0.8	0.91	0.33	0.76	0.95	0.93	0.90
Phytates	5.88	5.84	5.86	4.95	6.10	5.10	6.07	6.75	7.93	6.71	7.93	7.90	5.01
Oxalate	0.24	0.25	0.24	0.23	0.28	0.26	0.39	0.59	0.36	0.58	0.38	0.35	0.27

**Table 6: Concentrations of phytate, Zn, Ca, Fe and calculated [Phy]/[Zn], [Ca]/[Phy], [Phy]/[Fe] and [Ca][Phy]
Mole ratios of the vegetables analyzed**

Parameters	A	B	C	D	E	F	G	H	I	J	K	L	M
Phytates	5.88	5.84	5.86	4.95	6.10	5.10	6.07	6.75	7.93	6.71	7.93	7.90	5.01
Calcium	2.89	4.53	3.13	2.08	4.07	2.58	4.97	2.26	5.30	4.53	5.06	3.75	6.10
Zinc	0.0771	0.0579	0.0677	0.152	0.0546	0.0623	0.0558	0.088	0.0716	0.102	0.0995	0.147	0.0623
Fe	0.402	0.286	0.305	0.260	0.311	0.326	0.601	0.514	0.511	0.807	0.695	0.515	0.402
[Phy]/[Zn]	7.56	10.0	8.58	3.23	11.1	8.11	10.9	7.60	9.29	7.70	7.90	5.33	7.56
[Ca]/[Phy]	7.95	12.5	8.64	6.80	10.8	8.19	13.2	5.42	12.8	9.24	10.3	7.68	19.9
[Ca][Phy]/[Zn]	0.54	1.12	0.66	0.17	1.11	0.52	1.32	0.42	1.22	0.86	0.99	0.49	1.0
[Phy]/[Fe]	1.24	1.73	0.63	1.61	1.66	1.32	0.854	1.11	1.31	0.700	0.965	1.30	1.0

Values of phytates and selected minerals used were in mg/100g

Table 7: Non starch polysaccharide (NSP) components (%) of the vegetable samples analyzed

Parameters	A	B	C	D	E	F	G	H	I	J	K	L	M	N
NDF	5.6	5.62	5.65	5.59	4.98	7.21	5.56	5.58	7.25	8.21	4.65	8.3	7.8	11.22
ADF	3.98	3.99	4	3.96	3.54	5.83	3.8	3.84	5.88	6.3	3.33	6.37	6.52	9.31
ADL	2	2.01	2.03	2	2.02	3.17	1.92	1.95	3.2	4.22	1.9	4.26	4.5	5.55
CEL	1.98	1.98	1.97	1.96	1.52	2.66	1.88	1.89	2.68	2.08	1.443	2.11	2.02	3.76
HMC	1.62	1.63	1.65	1.63	1.44	1.38	1.76	1.74	1.37	1.91	1.32	1.93	1.28	1.91

NDF=neutral detergent fibre, ADF= acid detergent fibre, ADL = acid detergent lignin, CEL= cellulose, HMC= Hemicellulose

Table 8: Results of Chi-square test (χ^2) on the results from Tables 2, 3, 5 and 6

Mineral	χ^2	Mineral Ratio	χ^2	ANFS	χ^2	Interrelationship	χ^2
Na	43.4 ^a	Ca/Mg	0.380 ^b	Tannin	0.582 ^b	Phytates	2.22 ^b
K	473 ^a	Ca/P	46.7 ^a	Phenol	0.298 ^b	Calcium	51.5 ^a
Ca	51.5 ^a	Na/K	0.343 ^b	Alkaloids	16.3 ^b	Zinc	1.81 ^b
Mg	298 ^a	Na/Mg	0.400 ^b	Saponin	5.823 ^b	Fe	7.84 ^b
Cu	0.168 ^b	K/Na	37.4 ^a	Flavonoids	0.853 ^b	[Phytate]/[Zn]	1.18 ^b
Co	0.008 ^b	Zn/Cu	388 ^a	Phytates	2.216 ^b	[Ca]/[Phytate]	8.93 ^b
Fe	7.84 ^b	ME*	17845869 ^a	Oxalate	0.535 ^b	[Ca][Phytate]/[Zn]	16.8 ^b
Zn	1.81 ^b					[Phytate]/[Fe]	2.10 ^b
Se	0.008 ^b						
P	6.17 ^b						

^a results significantly different at $p=0.05$, $v=n-1=13$, ^b results not significantly different at $p=0.05$, $v=n-1=13$, AFNS= anti-nutritional factors, CEL= cellulose, HMC= hemicellulose