

Investigating the Dietary Antioxidant and Mineral Constituents of Three Commonly Consumed Powder Vegetables Sold in Lapai Market, Lapai - Niger State, Nigeria

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Abstract

*Antioxidant compounds in food play an important role in health protecting factor. Plant sourced food antioxidants like vitamin C, vitamin E, carotenes, phenolic acids, and phytoestrogens have been recognized as having the potential to reduce the risk of chronic diseases. This study therefore investigated some antioxidant and mineral constituents of three commonly consumed powder vegetables (*Adansonia digitata*, *Abelmoschus esculentus* and *Annona senegalensis*) sold in Lapai markets, Lapai – Niger state of Nigeria. The concentration of ascorbic acids in the powder vegetables were determined by titrimetric method. Total carotenoids, β-carotene and lycopene in the vegetables were analyzed using spectrophotometric method. The mineral content was determined using Atomic Absorption Spectrophotometer while sodium and potassium was determined using*

*Flame photometer. The result of this investigation revealed that there was no significant difference ($p>0.05$) in the mean values of ascorbic acid for the three vegetables analyzed. The β -carotene concentration of the vegetables decreases as *A. digitata* > *A. esculentus* > *A. senegalensis*. The result also shows that *A. digitata* was significantly ($p<0.05$) higher in β -carotene concentration when compared to the two other vegetables. Concentration of total carotenoid in the three vegetables revealed that *A. digitata* had the highest concentration while *A. esculentus* recorded the lowest value, but these values were significantly different from each other. Lycopene contents of the vegetables were similar to that of the total carotenoid but *A. digitata* was significantly different ($p<0.05$) from the two other vegetables. The mineral compositions of the vegetables showed that calcium concentration of *Adansonia digitata* was significantly ($p<0.05$) higher when compared to the two other vegetables. *Abelmoschus esculentus* had the highest concentration for Mg, Cu, Fe, Zn, Na and K, followed by *Annona senegalensis* except for magnesium while *Adansonia digitata* recorded the least values for all the elements analyzed apart from calcium and magnesium content. The consumption of these vegetables can significantly contribute to the needed dietary antioxidant and mineral intake that can reduce the risk of chronic diseases and nutrient deficiency in the body.*

Keywords: Vegetables, Dietary antioxidant, Mineral, *Adasonia digitata*, Total carotenoids and Ascorbic acid.

Introduction

Antioxidant compounds in food play an important role as a health protecting factor. They are our first line of defense against free radical damage and are critical for maintaining optimum health and wellbeing. Scientific evidence suggests that antioxidants reduce the

risk of chronic diseases such as cancer and heart disease. Primary sources of naturally occurring antioxidants are whole grains, fruits and vegetables. Plant sourced food antioxidants like vitamin C, vitamin E, carotenes, phenolic acids, and phytoestrogens have been recognized as having the

potential to reduce disease risk (Dembinska-Kiec, 2008). Most of the antioxidant compounds in a typical diet are derived from plant sources and belong to various classes of compounds with a wide variety of physical and chemical properties.

Antioxidants are known mainly for their ability to trap free radicals. Highly reactive free radicals and oxygen species are present in biological systems from a wide variety of sources. These free radicals may oxidize nucleic acids, proteins, lipids or DNA and can initiate degenerative disease. Antioxidant compounds like phenolic acids, polyphenols and flavonoids scavenge free radicals such as peroxide, hydroperoxide or lipid peroxyld and thus inhibit the oxidative mechanisms that lead to degenerative diseases. There are a number of clinical studies suggesting that the antioxidants in fruits, vegetables, tea and red wine are the main factors for the observed efficacy of these foods in reducing the incidence of chronic diseases including heart disease and some cancers. The free radical scavenging activity of antioxidants in foods has been substantially investigated and reported in the literature by Miller *et al.* (2000a and 2000b).

Vitamin C, vitamin E, and beta carotene are among the most widely studied dietary antioxidants. Vitamin C is considered the most important water-soluble antioxidant in extracellular fluids. It is capable of neutralizing reactive oxygen species (ROS) in the aqueous phase before lipid peroxidation is initiated. Vitamin E, a major lipid-soluble antioxidant, is the most effective chain-breaking antioxidant within the cell membrane where it protects membrane fatty acids from lipid peroxidation. Vitamin C has been cited as being capable of regenerating vitamin E (Sies *et al.*, 1992). Beta carotene and other carotenoids are also believed to provide antioxidant protection to lipid-rich tissues. Research suggests that beta carotene may work synergistically with vitamin E (Jacob, 1995; Sies and Stahl, 1995). A diet that is excessively low in fat may negatively affect beta carotene and vitamin E absorption, as well as other fat-soluble nutrients. Fruits and vegetables are major sources of vitamin C and carotenoids, while whole grains and high quality, properly extracted and protected vegetable oils are major sources of vitamin E.

Minerals are essential nutrients found in many different types of plant and animal-based foods. Macro-minerals or those require in greater amounts, include calcium, potassium, sodium, phosphorus, magnesium, chloride, and

sulfur. Trace minerals, or those needed in smaller amounts, include iron, zinc, selenium, manganese, copper, iodine, cobalt, and fluoride. Both types of minerals support a wide variety of bodily functions, ranging from building and maintaining healthy bones and teeth to keeping your muscles, heart and brain working properly (Nadia, 2018). It has been suggested that an inadequate dietary intake of these trace minerals may compromise the effectiveness of these antioxidant defense mechanisms (Duthie and Brown, 1994). Research indicates that consumption and absorption of these important trace minerals may decrease with aging (Duthie and Brown, 1994). Intensive agricultural methods have also resulted in significant depletion of these valuable trace minerals in our soils and the foods grown in them.

Vegetables are important in human health because of their vitamins, minerals, phytochemical compounds and dietary fibres (Musa *et al.*, 2014). They are rich in minerals such as potassium, sodium, calcium, iron, zinc and phosphorus. According to George (2003), the potassium content of leafy vegetable is good in the control of diuretic and hypertensive complications. The nutritive value of vegetable is increased greatly because of the presence of minerals and vitamins. They constitute essential diet components by contributing proteins, vitamins, iron, calcium, and other nutrients that are in short supply. Mammals rely on nutrients such as amino acids, fatty acids, vitamins, and minerals from ingested food to maintain an adequate nutritional status for the regulation of metabolic, physiological, and neuronal homeostasis, as well as for the prevention of diseases (Trumbo 2008; Stover *et al.* 2017).

Many scientists believe that the Recommended Dietary Allowance (RDA) for specific antioxidants may be inadequate and, in some instances, the need may be several times the RDA (Percival, 1998). As part of a healthy lifestyle and a well-balanced, wholesome diet, antioxidant supplementation is now being recognized as an important means of improving free radical protection. It is important to know the antioxidant and mineral content of vegetables, in addition to knowing the basic nutritional information such as the protein, fiber, fat and vitamin contents, thus this study investigates some dietary antioxidant and mineral constituents of three commonly consumed powder vegetables (*Adansonia digitata*, *Abelmoschus esculentus* and *Annona senegalensis*) sold in Lapai markets, Lapai – Niger state of Nigeria.

MATERIALS AND METHOD

Study Area

The study was conducted in Lapai town, Niger State, Nigeria. Lapai is situated between the latitudes 09°04' N and 09°05' N and longitudes 06° 34' E and 06°35'E of the equator. The major economic activity of the people of Lapai is farming with specialization in crops, animal husbandry and fishing. The analysis was carried out at Biochemistry Department, Ibrahim Badamasi Babangida University, Lapai.

Sources of Samples

The powder vegetables of *Adansonia digitata* (kuka), *Abelmoschus esculentus* (tsuku) and *Annona senegalensis* (nungbere) were bought from three different markets, Lapai town, Lapai Local Government of Niger state. The powders were sieved and used for the analysis.

Chemical Analysis

Triplicate sample of each powder vegetable was used to determine ascorbic acid, β-carotene, total carotenoid, lycopene and mineral contents.

Ascorbic Acid (Vitamin C)

The vitamin C content of the sample was determined using the method described in AOAC (2005). Exactly 0.05g of 2, 6-dichloroindophenol dye was dissolved in distilled water and make up to 100ml then filtered. Also, 0.05g of pure ascorbic acid standard was dissolved in 60ml of 20% glacial acetic acid and diluted to 250ml using distilled water. Then 10ml of standard ascorbic acid was pipetted into a conical flask and titrated with the dye solution until a faint pink colour persisted for 15 second. The concentration was expressed as mg ascorbic acid equivalent to 1ml of the dye solution. Accurately ten (10) grams of the powder was homogenized with 100ml of glacial acetic acid. The suspension was filtered through a Whatman filter paper No. 1 and 10ml of the filtrate was pipetted into a conical flask containing 2.5ml acetone and titrated with the dye solution until a faint pink colour persisted for 15 seconds.

Ascorbic acid was calculated as mg per 100g food sample as

Titre x N x dilution factor

N = mg ascorbic acid standard equivalent to 1ml dye solution.

Determination of β -Carotene

The β -carotene of the sample was determined using the method described by Musa *et al.*, (2010). Exactly 2.0g of Na₂SO₄ was added to 10g of the powder samples and homogenized. The homogenized sample was extracted with 100ml of hot 95% ethanol for 30 minutes in hot water bath. The extract obtained was decanted, filtered, measured and distilled water was added to the extract to bring the percentage of the ethanol extract to 85%; then cooled in a cold water bath for 10 minutes. After cooling, the ethanol extract was transferred to separating funnel with 30ml of petroleum ether added and the mixture shaken. The separating funnel was clamped to the retort stand and the mixture was allowed to settle into two layers. The bottom layer containing the ethanol was collected into a beaker while the top layer (petroleum layer) was stored in 250ml conical flask and covered with aluminium foil. The ethanol layer in the beaker was re-extracted twice with 10ml of petroleum ether. The ether layers of re-extraction were added to the original petroleum extract in the conical flask and re-extracted with 50ml of 85% ethanol in order to remove any xanthophylls which may be present. The top petroleum ether layer which contained the β -carotene was collected, measured and volume recorded. The absorbance of the final petroleum ether extract was determined at the wavelength of 450nm with spectrophotometer (UV 3100 PC) using petroleum ether as the blank.

Calculation: The concentration of β -carotene was calculated thus:

$$A = \epsilon\% \times C \times l$$

Where A = Absorbance of the sample

C = Concentration

$\epsilon\%$ = Extinction coefficient of β -carotene

l = Path length (usually 1.0cm)

Determination of Total Carotenoids and Lycopene

Total carotenoids and lycopene were estimated by the method described by Zakaria *et al.* (1979). The experiment was carried out in the dark to avoid photolysis of carotenoids once the saponification was complete. The sample (0.5g) was homogenized and saponified with 2.5ml of 12% alcoholic potassium

hydroxide in a water bath at 60°C for 30 minutes. The saponified extract was transferred to a separating funnel containing 15ml of petroleum ether and mixed well. The lower aqueous layer was then transferred to another separating funnel and the upper petroleum ether layer containing the carotenoids was collected. The extraction was repeated until the aqueous layer became colourless. A small amount of anhydrous sodium sulphate was added to the petroleum ether extract to remove excess moisture. The final volume of the petroleum ether extract was measured and noted. The absorbance of the yellow colour was read in a spectrophotometer (UV 3100PC) at 450nm and 503nm using petroleum ether as blank. The amount of total carotenoids and lycopene were calculated using the formulae,

$$\frac{A_{450} \times \text{Volume of the sample} \times 100 \times 4}{\text{Weight of the sample}}$$

$$\text{Amount of total carotenoids} = \text{Weight of the sample}$$

$$\frac{3.12 \times A_{503} \times \text{Volume of the sample} \times 100}{\text{Weight of the sample}}$$

$$\text{Amount of lycopene} = \text{Weight of the sample}$$

The total carotenoids and lycopene were expressed as mg/g of the sample.

Determination of Mineral Content

Mineral elements of the samples were determined according to AOAC (2000). Exactly 1.0g of each sample was ash in a muffle furnace (SXL-1008) at the temperature of 550°C and the ashes of various samples were dissolved in 10ml of 0.1M HCl, filtered and made up with distilled water to the mark in a 100ml volumetric flask. The mineral elements Mg, Ca, Zn, Fe, Cu, were determined using Atomic Absorption Spectrophotometer (AAS) (AA500 spectrophotometer, Pg Instrument) while Na and K were determined using Flame photometer (Jenway, PFP 7).

Statistical Analysis

The data obtained were subjected to ANOVA using IBM SPSS statistics 23 package and significant difference was accepted at $p < 0.05$. Values are given as Mean \pm S.D.

RESULTS

Antioxidant Constituents of Powder Vegetables

The ascorbic acid contents of the three vegetables show that *Adansonia digitata* has the highest content (0.67 ± 0.29 mg/100g), followed by *Annona senegalensis* (0.50 ± 0.00 mg/100g) and *Abelmoschus esculentus* (0.39 ± 0.10 mg/100g) recorded the lowest concentration. Although all the mean values were not significantly ($p>0.05$) different from each other (fig. 1). The β -carotene of the vegetables ranged from 38.67 ± 24.38 to 375.44 ± 81.13 μ g/100g. *Adansonia digitata* recorded the highest concentration while *Annona senegalensis* has the lowest concentration (fig. 2). Total carotenoid of the powder vegetables indicate that *Adansonia digitata* concentration was statistically higher (1702.13 ± 190.36 mg/g) than the other two vegetables (fig. 3). Lycopene contents show similar result with that of total carotenoid, with *Adansonia digitata* having the highest concentration (9920.77 ± 2302.18 mg/g), followed by *Annona senegalensis* (2289.04 ± 159.83 mg/g) and *Abelmoschus esculentus* (1488.87 ± 185.75 mg/g) recorded the lowest contents. Concentration of lycopene in *Adansonia digitata* was significantly ($p<0.05$) higher when compared with the other two vegetables (fig. 4).

Mineral Contents of Powder Vegetables

The calcium concentration of the powder vegetables shows that *Adansonia digitata* had the highest concentration (1.95 ± 2.64 mg/Kg), followed by *Annona senegalensis* (0.50 ± 0.08 mg/Kg) while *Abelmoschus esculentus* had the lowest concentration (0.40 ± 0.18 mg/Kg). The calcium concentration of *Adansonia digitata* was significantly ($p<0.05$) higher than the other two vegetables. The magnesium contents ranged from 1.07 ± 0.08 - 1.13 ± 0.03 mg/Kg, with *Abelmoschus esculentus* recording the highest value while *Annona senegalensis* had the lowest content. Copper content indicates that *Abelmoschus esculentus* (0.49 ± 0.19 mg/Kg) had the highest value, followed by *Annona senegalensis* (0.41 ± 0.04 mg/Kg) while *Adansonia digitata* (0.27 ± 0.00 mg/Kg) had the least value. There was no significant ($p>0.05$) difference in the copper concentration amongst the three samples. Level of iron content in the samples revealed that *Abelmoschus esculentus* had the highest values (0.63 ± 0.05 mg/Kg) while *Adansonia digitata* recorded the least mean value (0.49 ± 0.03 mg/kg). The iron

content in *Abelmoschus esculentus* was significantly ($p<0.05$) higher when compared with the two other samples. The highest concentration of zinc was found in *Abelmoschus esculentus* (0.40 ± 0.05 mg/Kg), followed by *Annona senegalensis* (0.36 ± 0.07 mg/Kg) while the lowest concentration was found in *Adansonia digitata* (0.28 ± 0.06 mg/kg). Among the studied powder vegetables, *Abelmoschus esculentus* had the highest concentration of sodium (8.57 ± 0.39 mg/Kg) while *Adansonia digitata* recorded the least value (4.78 ± 0.41 mg/kg). Similarly, the same trend was observed for potassium content with *Abelmoschus esculentus* recording the highest mean value (153.20 ± 15.29 mg/Kg) and *Adansonia digitata* had the lowest concentration (68.73 ± 1.96 mg/kg). There was significant ($p<0.05$) difference in potassium content amongst the three samples (Table 1).

Table 1: Mineral Contents of Powder Vegetables (mg/Kg)

Vegetables	Ca	Mg	Cu	Fe	Zn	Na	K
<i>Adansonia digitata</i>	1.95 ± 2.64 b	1.10 ± 0.09 a	0.27 ± 0.00 a	0.49 ± 0.03 a	0.28 ± 0.06 a	4.78 ± 0.41 a	68.73 ± 1.96 a
<i>Abelmoschus esculentus</i>	0.40 ± 0.18 a	1.13 ± 0.03 a	0.49 ± 0.19 a	0.63 ± 0.05 b	0.40 ± 0.05 a	8.57 ± 0.39 b	153.20 ± 15.29 c
<i>Annona senegalensis</i>	0.50 ± 0.08 a	1.07 ± 0.08 a	0.41 ± 0.04 a	0.55 ± 0.03 a	0.36 ± 0.07 a	7.72 ± 0.74 b	121.00 ± 17.65 b

Values are Mean \pm S.D of triplicate determinations. Values with different superscripts down the column are significantly different ($p<0.05$).

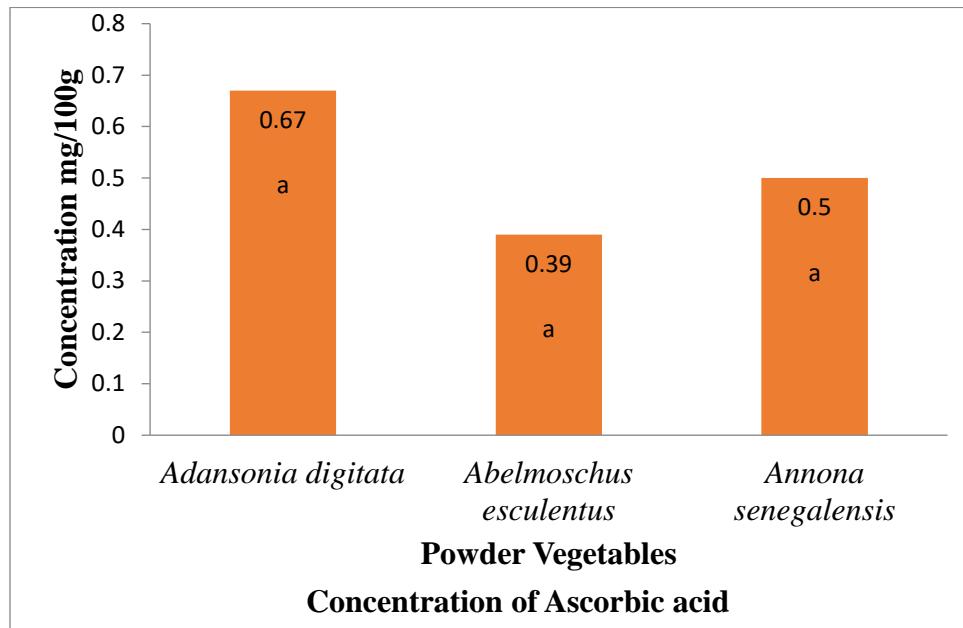


Figure 1: Concentration of Ascorbic acids in three powder vegetables. Bar with the same alphabets are not significantly different from each other ($p>0.05$).

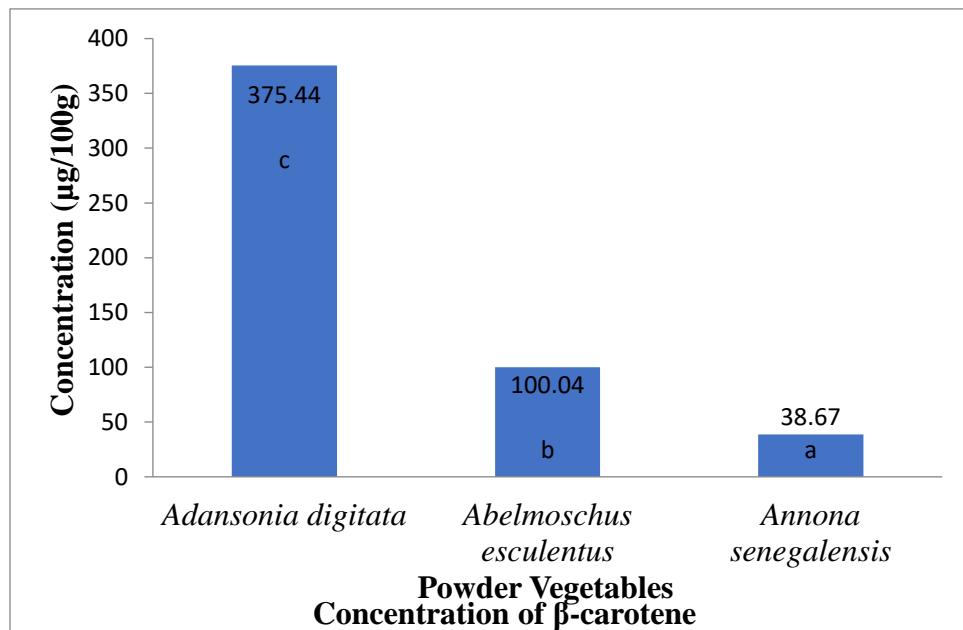


Figure 2: Concentration of β-carotene of three powder vegetables. Bar with the same alphabets are not significantly different from each other ($p>0.05$)

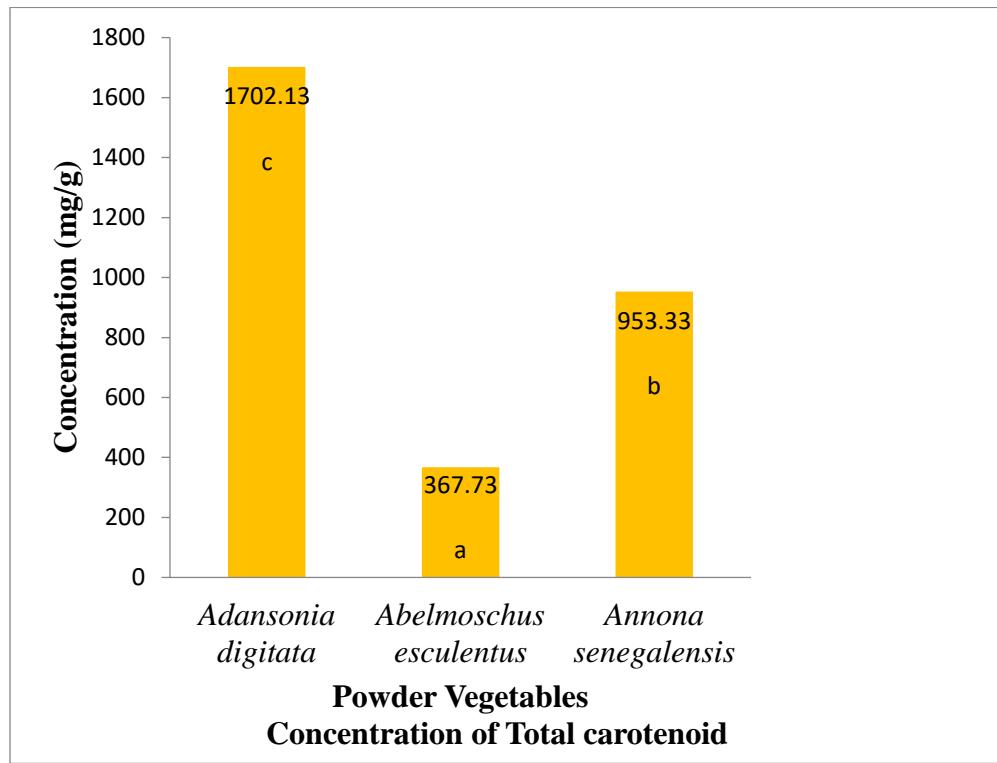


Figure 3: Concentration of Total carotenoid of three powder vegetables.
 Bar with the same alphabets are not significantly different from each other ($p>0.05$)

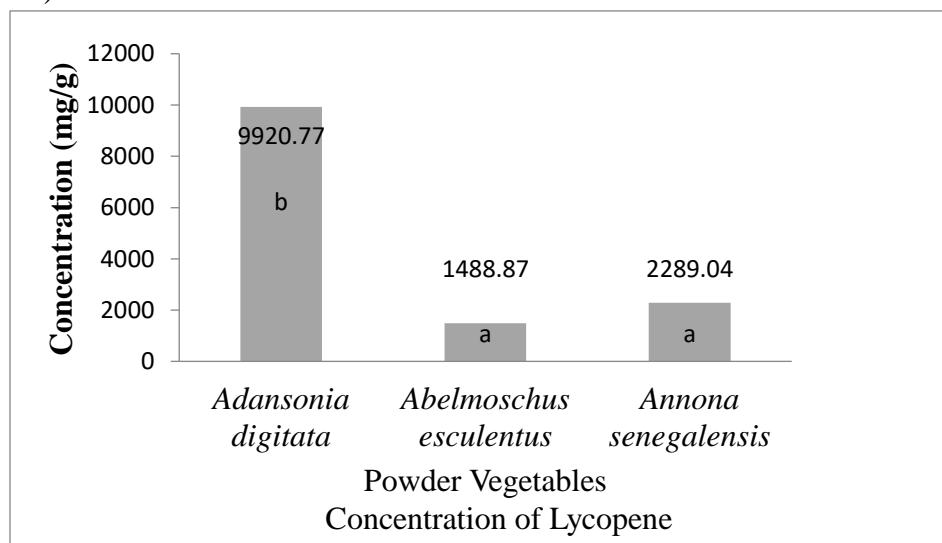


Figure 4: Concentration of lycopene in three powder vegetables. Bar with the same alphabets are not significantly different from each other ($p>0.05$)

DISCUSSION

Antioxidants are important in preventing degenerative diseases such as cardiovascular disease, stroke and cancers. The differences in concentration of ascorbic acids in the three vegetables were not statistically significant; however ascorbic acids or vitamin C is considered the most important water-soluble antioxidant in extracellular fluids. It is capable of neutralizing reactive oxygen species (ROS) in the aqueous phase before lipid peroxidation is initiated. Vitamin A is an essential micronutrient required for vision and a variety of metabolic functions in the body. In developing countries, carotenoids present in plant foods supplied more than 80% of the dietary vitamin A consumed by the people (Bhaskarachary *et al.*, 1995)). The most predominant and active carotenoid in these foods is β -carotene. Bhaskarachary *et al.*, (1995) reported that Green leafy vegetables (GLV) were found to be the best source of pro-vitamin A and also yellow fruits such as mango and papaya. The diets of peoples in the tropical world rarely contains large amounts of milk, eggs, liver, which are the rich sources of preformed vitamin A; therefore, there is a great deal of dependence on carotenoids, particularly from leafy vegetables, as a source of vitamin A (Tee, 1995).

In this study, *Adansonia digitata* has the highest concentration of all antioxidants investigated in the powder leafy vegetables. This may be responsible for the use of the plant in the treatment and management of so many health challenges. High level of lycopene in the vegetables investigated suggests that consumption of the vegetables may be helpful in the treatment and prevention of cancer. This is in tandem with the suggestion of Clinton (1998) that lycopene helps in the prevention of cardiovascular disease and cancers of the prostate or gastrointestinal tract. The three investigated leafy vegetables showed richer levels of lycopene as compared to fruits such as tomatoes, apricot, grapefruit, guava, water melon and papaya which were investigated by Clinton (1998).

Also, the presence of high level of carotenoids and β -carotene may be responsible for the anticancer potential associated with these vegetables. This is in line with the works of Ziegler (1989) and Mayne (1996), both of whom reported that carotenoids reduce the risks of cancer development and beta-carotene prevent diseases in human respectively. Although, Greenberg *et. al.*

(1990) reported that β -carotene has no preventive effects on skin cancer. Also, administration of β -carotene in high risk populations showed an increased rate of lung cancer (Greenberg *et. al.*, 1990). However, a protective effect against oral leukoplakia was reported by Garewal (1995) and pre-malignant lesions of the cervix can be prevented by β -carotene supplementation (Meyskens and Manetta, 1995).

Mineral Compositions

The mineral compositions of the three powder vegetables indicate that *Adansonia digitata* had the highest calcium content and this indicates that it is a better source of calcium than the two other vegetables. *Adansonia digitata* with high content of calcium may help in bone formation and blood coagulation whose deficiency may contribute to rickets, curvature of the spine, pelvic and thoracic deformities (Wardlaw and Smith, 2006). Apart from calcium content, *Abelmoschus esculentus* had the highest concentrations for all the mineral elements analyzed and followed by *Annona senegalensis*. Magnesium concentrations in the vegetables were lower than the values reported by Okerulu & Onyema (2015) for *Gnetum africanum* leaves (12.00 mg/kg) and a value of 240 mg/kg for *A. senegalensis* (Yisa *et al.*, 2010). The presence of magnesium in these leaves may prevent cardiomyopathy, impaired spermatogenesis and bleeding disorders (Chaturvedi *et al.*, 2004). *A. esculentus* could provide more copper contents to the body than the two other vegetables and this is similar to the result reported for *G. africanum* leaves (Okerulu & Onyema, 2015). Copper is essential for red blood cell formation and an enzymatic element for normal plant growth and development but can be toxic at excess dose (Levetin, 2008). Diagnosed deficiency is rare but when it becomes deficient, it may lead to anemia, impaired immunity and bone diseases (Levetin, 2008). *A. esculentus* could contribute to iron contents and iron is an essential component of human hemoglobin and helps to facilitate the oxidation of carbohydrates, protein and fat and hence help to regulate body weight, which is a very important factor in diabetes.

The zinc contents in this study are far below those reported for local okra variety of 12.9 – 13.7 mg/kg (Adetuyi *et al.*, 2011) and 2.40 mg/kg for *G. africanum* leaves (Okerulu & Onyema, 2015). Zinc is an essential trace element needed

for healthy plant growth and plays an important role in biochemical and physiological processes including normal growth, brain development, behavioural response, bone formation and wound healing. Zinc is involved in RNA and DNA synthesis, which influences cell division, repair and growth. It may enhance cell growth and multiplication (Rosenkranz *et al.*, 2017). Zinc deficiency in diabetic patient could impair power to perceive odour and loss of sensation to the skin. Deficiency of zinc can lead to recurrent infections, lack of immunity and poor growth, male hypogonadism, skin changes, poor appetite and metal lethargy (Prasad, 2020).

The sodium concentration of the powder vegetables showed that *A. esculentus* had the highest content but not significantly ($p>0.05$) different from each other. Similar trend was observed for potassium, where *A. esculentus* recorded the highest concentration and followed by *A. senegalensis*. But the concentrations are significantly ($p<0.05$) different from each other. Potassium and sodium are essential electrolytes found in biological fluids and are central to the maintenance of cellular membrane potential, osmoregulation as well as transmit nerve impulse and muscle cells. Their deficiency has been associated with impaired renal function, alterations of gastric secretions and intestinal motility (Princewill-Obgonna *et al.*, 2019). Potassium also influence glucose and lipid metabolism. Increase intake of potassium can lower blood pressure and may help prevent strokes. However excess potassium intake (hyperkalemia) may lead to heart failure and death. Similarly, high sodium intake has been associated with high fluid retention, leading to hypertension and heart failure.

CONCLUSION

Adansonia digitata had better antioxidant constituents amongst the vegetables and *Abelmoschus esculentus* had more of the mineral elements analyzed in the vegetables. Therefore the regular consumption of these vegetables can significantly contribute to the needed dietary antioxidant and mineral deposit in the body that can reduce nutrient deficiencies and risk of chronic diseases in life.

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