PROXIMATE ANALYSIS, PHYTOCHEMICAL SCREENING, ANTIOXIDANT ACTIVITIES AND POTASSIUM/SODIUM DETERMINATION OF YELLOW *Ipomoea batata* GROWN IN ANKPA, KOGI STATE, NIGERIA

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Abstract: The physical and chemical component, bioactive substance, concentration of sodium & potassium and antioxidant properties of yellow *Ipomoea batata* were examined. Crude fiber, moisture content were present; phytosteroids, phenols, tannins, flavonoids, protein & amino acids were bioactive substances found in both peels and tuber of the plant. High potassium content and low sodium content were also present in the plant. The extracts showed that it contained an antioxidant properties, the presence of this important substances makes this plant a useful neutrocetical therapy for better health management.

Keywords: Antioxidant, phytochemical, *Ipomoea batata*, sodium, potassium

Introduction

Sweet potato (*Ipomoea batata*) is one of the staple food grown in Africa which play a vital role in combating food shortage and malnutrition among the population (Sanoussi *et al*., 2016; Fesco *et al*., 2002) assert that sweet potato is an important food crop of the tropical and subtropical areas which have a nutritional advantage for both rural and urban dwellers of these region by increase in its production and consumption. In comparison with other tubers, Ingabire and Hilda (2011) revealed that sweet potato contain an average amount of protein and carbohydrate. Also, that it contain some free sugar which give the tuber its sweet taste. In addition, Sanoussi *et al.* (2016) and Teow *et al*. (2007) added that sweet potato is a nutritious food, low in fat and protein but rich carbohydrate.

Phytochemicals are bio-active substances (Natural compound) that are present in plants. Phytochemical screening of some plants has revealed the presence of numerous chemicals including alkaloids, tannins, flavonoids, steroids, glycosides, phenolic compounds, saponin, and carbohydrates (Sofowara, 1993). Cowan (1999) revealed that these chemicals contain active ingredients like metabolites that are mostly used as source of medicine. He further affirmed that secondary metabolites of plants serve as defense mechanism against predation by micro-organisms, insects and herbivores. In view of this, Ivanova (2005) stated that various medicinal properties have been attributed to natural herbs which constitute the main source of new pharmaceutical and health care products.

Antioxidants are compounds that inhibit oxidation. Oxidation is a chemical reaction that can produce free radicals which causes damage to the cells. Well known antioxidants include enzymes, vitamins C & E, and beta carotene. These are capable of counteracting the damaging effects of oxidation. Oxidative stress is the imbalance between pro oxidants and antioxidants produced in biological system; these have serious effects in the process of aging and development of chronic disease such as arteriosclerosis, cancer and diabetes. Halliwel (1994) opined that relative oxygen and nitrogen species formed in excess in any biological system destroys cellular components like lipids, protein and DNA or cause a decrease in the capacity of antioxidant defense. Kris-Etherton *et al.* (2002) also revealed that, aside the normal body process that result in the formation of reactive species , dietary , lifestyle and environmental factors like smoke, alcohol consumption and infection , oxidative stress could accelerate the generation of harmful free radical which are dangerous to health. Shobha *et al.* (2015) affirmed that antioxidants inhibit the oxidation of molecules and terminates the chain reactions caused by free radicals. Therefore antioxidants are needed to not only reduce the level of oxidative stress common in many chronic diseases but also to serve as adjuvants to other standard therapies in order to provide a synergistic effect in combating diseases (Halliwel, 1994; Kris-Etherton *et al.*, 2002). Antia *et al.* (2006) further added that both tubers and leaves of sweet potato are good sources of antioxidant, fiber, zinc, potassium iron and vitamin C.

Physiochemical analysis is a method of investigating physic-chemical parameters that makes possible a determination of nature of the interactions between the components of a system through a study of the relations between the system, physical properties and composition. Ncube (2008) added that characterization of physiochemical properties has attained strong interest in pharmaceutical research area. The author further stated that it is one of the key challenges in developing a pharmaceutical active ingredient in drugs.

According to Ikwuchi and Ikwuchi (2009), minerals are naturally occurring chemical elements that the body uses to help perform certain chemical reactions and they are essential for the normal functioning of muscles, heart, nerves and in the maintenance of body fluid composition as well as for building strong bones (Chaney, 2006). Therefore, sodium and potassium play an important role in body regulation. Sodium in the form of sodium chloride is used as flavor in food and also serves as food preservative. This element is required in a small amount in the body to conduct nerve impulses, contract and relax muscles. According to Alinnor and Oze (2011), sodium is an important mineral that assists in the regulation of the body fluid and in the maintenance of electrical potential in the body tissue. Desimane *et al.* (2006) further revealed that increase of sodium intake can cause increased blood pressure which in turn can increase the risk for cardiovascular and renal disease. More so, according to Alinnor and Oze (2011) a food source having Na/K ratio of less than 1 has impact on lowering blood pressure.

Potassium, on the hand, is required by the body to help balance the level of sodium in the body. Good sources of potassium include vegetables, fruits, seafood and dietary. Potassium has been found to play an important role in reducing the risk of hypertension which can lead to stroke. Sanoussi *et al.* (2016), Alinnor and Oze (2011) assert that intake of potassium in the body contributes to the regulation of heart beat, neurotransmission and water balance of the body. In addition, Dietary Guidelines for Americans (2005) recommended intake of potassium for adult and children to range from 3800 – 4700 mg/day.

Materials and Methods

The materials used include pulverized peels &tuber of sweet potato (*Ipomoea batata*) which was extracted using absolute ethanol (99%), sodium hydroxide, ammonia solution,

hydrochloric acid, ferric chloride, conc. H_2SO_4, Nitric acid, Wagner reagent, Benedict solution were standard analytical reagents used for the phytochemical analysis. Muffle furnace and double beam uv/vis spectrophotometer 2700, were the instruments used for physiochemical determination and antioxidant activity.

Collection and sample treatment

The raw sweet potato tubers were purchased from Ankpa main market of Kogi state and the bark was carefully peeled. The tubers were cut into pieces and both the bark and tuber was air dried for five (5) days. The dried tuber and peels were grounded into powder, 200 g each of the pulverized tuber and peels were weighed into 500 cm capacity beaker respectively. 400 ml of absolute ethanol was then added to each beaker and both were covered with aluminum foil and allowed to extract for a week. The extracts was filtered and evaporated to dryness to obtain the solutes. These were used for antioxidant activities and phytochemical screening.

Proximate analysis

Moisture content

This was done by the gravimetric method as described by AOAC (2000). 5 g of the samples each were weighed into a moisture can and the samples were air dried at room temperature for five (5) days. These samples were weighed each day until a constant weight was obtained. Loss in moisture was calculated and expressed as percentage of the weight of sample analyzed using the formula

Moisture content (%) = w3-w1/w2-w1* 100/1

Where: W1 = weight of empty moisture can; W2 = weight of empty can + sample before drying (g); W3 = weight of can +dried sample at constant weight (g)

Crude fiber

Crude fiber was analyzed as described by Ramanyi (2017); 5.0 g of the processed samples were boiled in 150 ml of 1.25% H_2SO_4 solution for 30 min. The boiled samples were then washed in several portion of hot water using a two- fold cloth to trap the particles. It was then returned to the flask and boiled again in 150 ml of NaOH solution for another 30 minutes under the same condition. After washing in several portion of hot water, the sample was allowed to drain dry before being transferred into a weighed crucible where it was ignited in a muffle furnace at 600^0C for 30 min. Samples were then cooled in a desiccator and weighed. Crude fiber was calculated thus:

Crude fiber (%) = w3-w2/w1*100/1

Where: W1 = weight of the sample (g); W2 = weight of the crucible + sample ash (g); W3 = weight of crucible + sample after washing, boiling & drying (g)

Ash analysis

Total ash content

This was done by the furnace incineration gravimetric method described by Chinyere *et al.* (2014). 5 g of the processed samples was weighed into a weighed porcelain crucible. The samples were ignited to ash in a muffle furnace at a temperature of 550^0C. When it has become completely ashed, it was cooled in a dessicator and weighed. The weight of the ash obtained was determined by difference and the result expressed in percentage thus:

Ash content (%) = w2-w1/sample weight * 100/1

Where: W1 = weight of empty crucible (g); W2 = weight of crucible + ash (g)

Water soluble ash value

This was determined in accordance with AOAC (1990) and Adeyeye and Agesin (2007). The ash obtained was boiled with 25 cm^3 distilled water for 30 minutes and the liquid filtered through an ashless filter paper and thoroughly washed with hot distilled water. The filter paper together with the residue was then ignited in the original crucible at a temperature of 550^0C, cooled and water insoluble ash weighed. This is obtained using:

Water soluble ash (%) = Total ash (%) – water insoluble ash (%)

Acid insoluble ash value

The procedure for the determination of total ash was followed as described by AOAC (2005). The ash was boiled with 25 ml dilute HCl acid (10% v/v) for 5 minutes, the liquid was filtered through an ashless filter paper and thoroughly washed with hot water. The filter paper together with the residue was then ignited at 550^0C in muffle furnace in the original crucible. It was then cooled and weighed (Adeyeye and Agesin, 2007). Acid insoluble ash was calculated thus:

Acid insoluble ash (%) = Acid insoluble ash/weight of sample * 100/1

The acid soluble ash value was determined by difference between the % weight of ash and % weight of the insoluble ash value.

Potassium and sodium determination

The Potassium and Sodium were determined using AOAC (2005) wet ashing method. 2 g of the samples each were weighed into separate glass tubes and 5 ml of the digested solution (conc. HNO_3 & perchloric acid in a ratio of 2:1) was added. The tubes were then heated at 200^oC in a water bath until the solution became translucent and brown fumes stop being released. This indicate complete digestion of the organic matter. The digest rate were then filtered using ashless filter paper into 50 ml volumetric flask and make up to the mark using distilled water. The aliquot of the solution were used for analyses. The potassium and sodium were determined using uv/vis spectrophotometer 2700 at wavelength 350 nM

Phytochemical screening

Standard phytochemical screening method was employed for each of the parameters as described by Prashant *et al.* (2011).

Test for alkaloid

Wagner test; 2 g of each extracts was dissolved in dil. HCl and filtered. The filtrates were then treated with 2 ml Wagner reagent. The mixture was then observed for brown/reddish precipitate confirming the presence of alkaloids.

Test for saponin

Foam test; 1 g of each extracts were mixed with distilled water and shaken vigorously and allowed stand for 10 minutes, observation of a stable persistent foam indicates the evidence of saponins.

Test for phytosterols

Salkowskis test; 2 g of the extracts were treated with chloroform and filtered, the filtrate was treated with few drops of conc. H_2SO_4, shaken and allowed to stand for the appearance of golden yellow color.

Test for flavonoids

Alkaline reagent test; 2 g of the extracts were treated with few drops of sodium hydroxide solution and the formation of intense yellow color which become colorless on addition of dilute acid was looked for.

Test for carbohydrate

Benedicts test; 1 g each of the extracts were dissolved in 5 ml of distilled water and filtered. 0.2 ml of the filtrates were each treated with Benedict reagent and heated gently. Formation of orange-red precipitate confirm the presence of carbohydrate.

Test for tannin

Ferric chloride test; 2 g of the sample extracts were boiled in water in a test tube and filtered. Two drops of ferric chloride solution was added to the filtrate, greenish-black precipitate will hydrolysable tannin (Trease and Evans, 2002).

Test for glycosides

Modified Borntragers test; 0.2 g each of the extracts were hydrolysed with dilute HCl and then treated with ferric

chloride solution, it was then immersed in boiling water for 5 minutes. The mixture was cooled and extracted with equal volume of benzene. The benzene layer was separated and treated with ammonia solution. Formation of rose-pink color in the ammoniacal layer indicate the presence of cardiac glycosides.

Test for phenols

Ferric chloride test; 0.2 g of the extracts were treated with 3-4 drops ferric chloride solution. Formation of bluish-black color indicates the presence of phenols.

Test for protein & amino acids

Xanthoproteic test; 1 g of the extracts were treated each with few drops of conc. HNO_3 acid. Formation of yellow color indicates the presence of proteins.

Antioxidants activities

Preparation of standard solutions

1. 0.0011 mol/100ml Ascorbic acid; 0.2 g of ascorbic acid was weighed and dissolved in 100 ml of distilled water. 2. 0.00013 mol/100ml potassium permanganate; 0.02 g of potassium permanganate was also weighed and dissolved in 100 ml of distilled water to make the stock. From the stock 25 ml of $KMnO_4$ solution was acidified by adding 10 ml of 2M H_2SO_4 solution.

Radical scavenging method using acidified potassium permanganate ($KMnO_4$) was used. In this method, ascorbic acid at concentration 2.0 - 0.25 mg/ml (i.e. 2.0, 1.5, 1.0, 0.5 & 0.25 ml) was used as standard antioxidant. 0.5 ml of acidified $KMnO_4$ solution and 0.5 ml of hydrogen peroxide (H_2O_2) was developed as the pro-oxidant. Absorbance of both the $KMnO_4$/ascorbic acid and $KMnO_4$/sample extracts of sweet potato (i.e. tuber & peels) were taken spectrophotometrically at a wavelength of 520 nM after 30 min incubation period. The graph of absorbance against concentration was then plotted. The % KMnO4 scavenging effect was determined using;

$$\% KMnO_4 \text{ scavenging effect} = Ac - At / Ac * 100/1$$

Where: Ac = Absorbance of control; At = Absorbance of the tested sample

Results and Discussion

The proximate analysis of the sweet potato samples (Table 1) shows the values for moisture content and ash content to be 60.5 and 5.4% for peels and 62.1 and 0.6% for tuber respectively. These values are in line with Ingabrie and Hilda (2011). The crude fiber, acid insoluble ash and acid soluble ash are present at 94.3, 1.2, and 4.2% for peels and 87.7, 0.2, and 0.4% for tuber, respectively. This result showed a high values for crude fiber in both the peels and the tuber. This confirmed Olusanya (1991) statement on the same plant that it contains fibers, he further revealed that dietary fiber improves glucose tolerance and is therefore beneficial in treating maturity pre- set diabetes. The high value of moisture content in tuber and crude fiber is also in line with Abubakar *et al.* (2010) and Hiroshi *et al.* (2000). The values of acid insoluble ash and acid soluble ash were found to be low; 0.2 and 4.2% for peels while the tuber has the values of 0.2 and 0.4%, respectively. In the same vein, both the peels and the tuber shows a low value for water soluble ash and water insoluble ash with values 2.4 and 3.0% for peels; 0.2 and 0.4% for tuber, respectively.

The protein values are 0.1519% for peels and 0.1544% for tuber, respectively (Table 1). This shows low protein content in both extract, this is in agreement with Abubakar *et al.* (2010), Ingabrie and Hilda (2011) as they also reported low values of protein on their work on similar plant.

Table 1: Proximate analysis of sweet potato peels & tuber

S/N	Parameter	Peels (%)	Tuber (%)
1	Moisture content	66.53	62.05
2	Ash content	5.4	0.6
3	Crude fiber	94.33	87.67
4	Acid insoluble ash value	1.2	0.2
5	Acid soluble ash value	4.2	0.4
6	Water soluble ash value	2.4	0.2
7	Water insoluble ash value	3.0	0.4
8	Protein value	0.1519	0.1544

Table 2: Phytochemical screening result

S/N	Parameter	Test performed	Peels	Tuber
1	Alkaloids	Wagner test	-	-
2	carbohydrates	Benedict test	-	+
3	Glycosides	Modified Borntragers test	+	-
4	Saponins	Foam test	-	-
5	Phytosterols	Salkowskis test	+	+
6	Phenols	Ferric chloride test	+	+
7	Tannins	Ferric chloride test	+	+
8	Flavonoids	lkaline reagent test	+	+
9	Protein & amino acids	Xantoproteic test	+	+

+ve = present -ve = absent

The result of the phytochemical screening (Table 2) showed the presence of carbohydrate in the tuber but absent in the peels; glycosides was found in the peels but absent in the tuber. phytosterols, phenols, tannins, flavonoids and protein were present in both the peels and tuber of the sweet potato samples. Alkaloids and saponins were absent in both the peels and tuber. This result is similar to the work done on the same plant by Anuselvi and Muthumani (2014). The presence of tannin could be reducing the risk of coronary heart disease as this important effect of tannin was revealed in Auselvi and Muthumani (2014). They also added that many natural antioxidant present in different parts of the plant in the form of phenolic compounds such as flavonoids, phenolic acid and tocophenols are potential antioxidants and are free radical scavengers.

The values for the concentration of potassium and sodium (Table 3) shows potassium value for both peels and tuber as 326 and 310 mg/100g, respectively and the value of sodium as 68.05 and 75.91 mg/100g, respectively. The result showed that the concentration of potassium is high in the peels than the tuber. This is in line with the findings reported by Ellong *et al.* (2014) and Laurie *et al* (2012). The high potassium present could be beneficial to consumers as potassium intake revealed by Naireta (2013) and Adjatin *et al.* (2013) could increase iron utilization which could be beneficial to those taking diuretic to control hypertension.

These values for sodium are within the recommended range of WHO (2013) new guidance on dietary salt. This is in agreement with Sanuonssi *et al.* (2016) where they recommended dietary allowance for children and adult to be 400 and 500 mg/day, respectively.

Table 3: Concentrations of potassium and sodium found in the samples

S/N	Element	Peels	Tuber
1	Potassium	326 mg/100g	310 mg/100g
2	Sodium	68.05 mg/100g	75.91 mg/100g

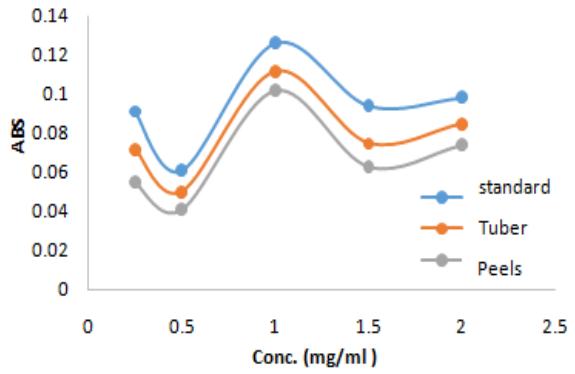

Fig. 1: Antioxidant activities of the extracts using the $KMnO_4$ scavenging effect

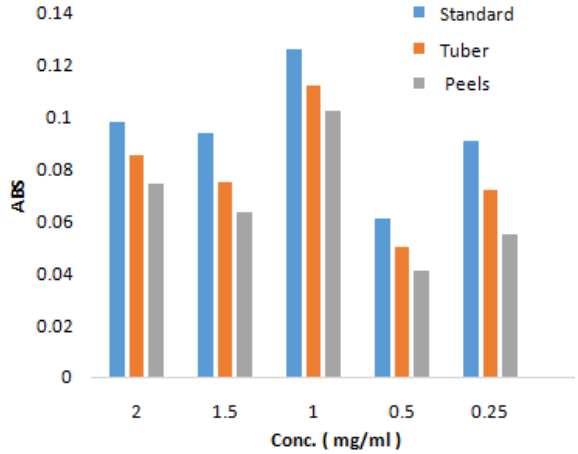

Fig. 2: Bar chat representation of the antioxidant scavenging effect of $KMnO_4$ on the extracts

The antioxidant activities using the $KMnO_4$ scavenging effect (Fig. 1) shows the effective concentration at 50 (EC50) values for both the peels and tuber as 0.775 and 0.675 mg/ml respectively. This shows that both extracts exhibit antioxidant properties as presented in the curves. The presence of various phytochemicals in the extracts may be responsible for it antioxidant properties. This result is in agreement with Kim *et al.* (2012). Where they revealed that sweet potato possess rich antioxidant content especially in the form of phenolic compounds. The % KMnO4 scavenging effect for peels and tuber samples were also found to be 19.1 and 11.1%, respectively.

Conclusion

From our findings we recommend the consumption of yellow sweet potato as it contains antioxidant and important phytochemicals that could fight free radicals in the body. It also contain high potassium content that could be of important in the regulation of heart beat, neurotransmission and water balance of the body. This plant could be a useful nutraceutical therapy for hypertension.

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Conflict of Interest

Authors declare there is no conflict of interest related to this study.

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