

**EVALUATION OF THE PROXIMATE AND VITAMINS COMPOSITIONS OF *Dioscorea villosa* IN
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Abstract

Dioscorea villosa was gotten from Ifitedunu in Dunukofia LGA of Anambra State. The proximate and vitamin compositions of *Dioscorea villosa* were analyzed. Proximate composition revealed varying quantities of protein, moisture, fats, carbohydrate, ash and fiber content with carbohydrate having the highest value of 70.82% and fibre having the lowest value of 0.56%. While in the vitamin composition, ascorbic acid was the highest with $68.76 \pm 0.00 \text{ mg/kg}$ with pyridoxine having the lowest value of $0.24 \pm 0.00 \text{ mg/kg}$. Conclusively from the finding of our research work, *Dioscorea villosa* should be recommended for daily consumption because of its high level of ascorbic acid in order to prevent scurvy in children and adults.

Keywords: *Dioscorea villosa*, proximate, vitamins, Anambra

INTRODUCTION

The nomenclature Yam applies to members of the *Dioscorea* genus of the *Dioscoreaceae* family within the order Dioscoreales (Alexander *et al.*, 1969). FAO (2001) noted that West Africa alone produces over 90% of the world's yam production with Nigeria being the largest producer producing above 50% of total world production (RMRDC, 1990). *Dioscorea villosa* is a species of twinning tuberous vine which is a native to eastern North America. It is commonly known as wild yam, colic root, rheumatism root, devils bones and four leaf yam (Govaerts *et al.*, 2007). Wild yam species contains diosgenin and have medicinal properties similar to those of other plants. Due to their powerful antifungal properties, they have been traditionally used for the treatment of inflammation, muscle spasms and Asthma. (Tabish, 2008).

Yams are nutritional powerhouse but for a while yam has been downgraded in the health food industry as not good for diabetic patients. Many people do not embrace the consumption of Yam without vegetables due to its belief of being a carbohydrate but they do not realize the widely acclaimed nutritional and health benefits hence the need to ascertain and evaluate the proximate analysis and vitamin analysis.

MATERIALS

Plant Materials

The plant material used for the study was wild yam gotten from Nkwo Ifitedunu in Dunukofia local government area in Anambra State. The wild yam was dried under room temperature, milled into powder and was stored until needed for extraction.

METHOD FOR DETERMINATION OF PROXIMATE COMPOSITION OF THE SAMPLE

Moisture Content (AOAC 2001)

Procedure

A petri-dish was washed and dried in the oven. Approximately 2g of the sample was weighed into petri-dish. The weight of the petri-dish and sample was noted before drying. The petri-dish and sample were put in the oven and heated at 105°C for 2hr. The result noted and heated another 1hr until a steady result is obtained and the weight was noted. The drying procedure was continued until a constant weight was obtained

$$\% \text{ Moisture content} = \frac{W_1 - W_2}{\text{Weight of sample}} \times 100$$

Where W_1 = weight of petri-dish and sample before drying
 W_2 = weight of petri-dish and sample after drying.

Carbohydrate Determination (Differential method)

$$100 - (\% \text{ Protein} + \% \text{ Moisture} + \% \text{ ash} + \% \text{ fat} + \% \text{ fibre})$$

Ash content

Procedure

Empty platinum crucible was washed, dried and the weight was noted. Approximately 1- 2g of sample was weighed into the platinum crucible and placed in a muffle furnace at 550°C for 3 hours. The sample was cooled in a dessicator after burning and weighed (AOAC, 2001)

Calculations

% Ash content =

$$\frac{W_3 - W_1}{W_2 - W_1} \times \frac{100}{1}$$

Where

W_1 = weight of empty platinum crucible

W_2 = weight of platinum crucible and sample before burning

W_3 = weight of platinum and ash.

Crude Fibre

Procedure: Defat about 2g of material with petroleum ether (if the fat content is more than 10%). Boil under reflux for 30 minutes with 200ml of a solution containing 1.25g of H_2SO_4 per 100ml of solution. Filter the solution through linen. Wash with boiling water until the washings are no longer acid. Transfer the residue to a beaker and boil for 30 minutes with 200ml of a solution containing 1.25g of carbonate free NaOH per 100ml. Filter the final residue through a thin but close pad of washed and ignited asbestos in a Gooch crucible. Dry in an electric oven and weigh. Incinerate, cool and weigh

The loss in weight after incineration $\times 100$ is the percentage of crude fibre.

$$\% \text{ crude fibre} = \frac{\text{weight of fibre}}{\text{Weight of sample}} \times 100$$

Crude fat

Soxhlet Fat Extraction Method

Procedure:

Dry 250ml clean boiling flasks in oven at $105 - 110^\circ C$ for about 30 minutes. Transfer into a dessicator and allow to cool. Weigh correspondingly labeled, cooled boiling flasks. Fill the boiling flasks with about 300ml of petroleum ether (boiling point $40 - 60^\circ C$). Plug the extraction thimble lightly with cotton wool. Assemble the soxhlet apparatus and allow to reflux for about 6 hours. Remove thimble with care and collect petroleum ether in the top container of the set – up and drain into a container for re – use. When flask is almost free of petroleum ether, remove and dry at $105^\circ C - 110^\circ C$ for 1 hour. Transfer from the oven into a dessicator and allow to cool, then weigh.

$$\% \text{ fat} = \frac{\text{wt of flask} + \text{oil} - \text{wt of flask}}{\text{weight of sample}} \times 100$$

Crude Proteins

Procedure

Exactly 0.5g of sample was weighed into a 30ml kjedahl flask (gently to prevent the sample from touching the walls of the side of each and then the flasks were stoppered and shaken. Then 0.5g of the kjedahl catalyst mixture was added. The mixture was heated cautiously in a digestion rack under fire until a clear solution appeared. The clear solution was then allowed to stand for 30 minutes and allowed to cool. After cooling was made up to 100ml with distilled water was added to avoid caking and then 5ml was transferred to the kjedahl distillation apparatus, followed by 5ml of 40% sodium hydroxide. A 100ml receiver flask containing 5ml of 2% boric acid and indicator mixture containing 5 drops of Bromocresol blue and 1 drop of methylene blue was placed added under a condenser of the distillation apparatus so that the tap was about 20cm inside the solution and distillation commenced immediately until 50 drops gets into the receiver flask, after which it was titrated to pink color using 0.01N hydrochloric acid. (AOAC, 2001)

Calculations

$$\% \text{ Nitrogen} = \text{Titre value} \times 0.01 \times 14 \times 4$$

$$\% \text{ Protein} = \% \text{ Nitrogen} \times 6.25$$

ESTIMATION OF VITAMINS

Estimation of vitamin A

Procedure

All procedures were carried out in the dark to avoid the interference of light. Sample (1.0g) was mixed with 1.0ml of saponification mixture and refluxed for 20 minutes at 60°C in the dark. The tubes were cooled and 20ml of water was added and mixed well. Vitamin A was sampled twice with 10ml of (40°-60°C) petroleum ether. The two samples were pooled and washed thoroughly with water. Anhydrous sodium sulphate was added to remove excess moisture. An aliquot of the sample (1.0ml) was taken and evaporated to dryness at 60°C. The residue was dissolved in 1.0ml chloroform. Standards (vitamin A palmitate) of concentrations ranging from 0-7.5µg were pipetted out into a series of test tubes. The volume in all the tubes was made up to 1.0ml with chloroform. TCA reagent(2.0ml) was added rapidly, mixed and the absorbance was read immediately at 620nm in a spectrophotometer (Genesys 10UV). The same procedure was repeated for the sample tubes also. Vitamin A content was expressed as µg/g(Onwuka, 2005)

Determination of vitamin B₁ and B₂

Procedure

1g of sample was weighed into a conical flask. This was dissolved with 100ml of deionized water. This was shaken thoroughly and heated for 5 minutes and allowed to cool and filtered. The filtrate was poured into cuvette and their respective wavelength for the vitamins set to read the absorbance using spectrophotometer (Onwuka, 2005)

Vitamin B₁ = 261nm

Vitamin B₂ = 242nm

Calculations:

$$\text{Concentration (mg\%)} = \frac{A \times D.F \times \text{volume of cuvette}}{E} \quad (5)$$

Where A = absorbance

E = extinction coefficient = 25 for B₁ and B₂

DF = dilution factor

Determination of vitamin B₃(Nicotinamide)

Procedure

5g of sample was dissolved in 20ml of anhydrous glacial acetic acid and warmed slightly. 5ml of acetic anhydride was added and mixed. 2 -3 drops of crystal violet solution was added as indicator. Titrate with 0.1M perchloric acid to a greenish blue colour (Onwuka, 2005)

Calculation:

$$\text{Vitamin B}_3 = \frac{\text{titre value} \times 0.0122}{0.1}$$

Determination of vitamin B₆

Procedure

5g of sample was dissolved in a mixture of 5ml of anhydrous glacial acetic acid and 6ml of 0.1M mercury II acetate solution. 2 drops of crystal violet was added as indicator. Titrate with 0.1M perchloric acid to a green colour end point. Calculation: each ml of 0.1M perchloric acid is equivalent to 0.02056g of C₈H₁₁NO₃HCL (Onwuka, 2005)

Determination of vitamin B₁₂

Procedure

25mg of sample was dissolved in 250ml of deionized water

Calculation:

$$\text{Conc Mg\%} = \frac{A \times D.F \times \text{volume of cuvette(s)}}{E}$$

Where A = absorbance

E = extinction coefficient = 25

D.F = dilution factor = 5

Estimation of vitamin C

Procedure

Standard ascorbate ranging between 0.2-1.0ml and 0.5ml and 1.0ml of the supernatant were taken. The volume was made up to 2.0ml with 4% TCA. DNPH reagent(0.5ml) was added to all the tubes, followed by 2 drops of 10% thiourea solution. The contents were mixed and incubated at 37°C for 3 hours resulting in the formation of osazone crystals. The crystals were dissolved in 2.5ml of 85% sulphuric acid, in cold. To the blank alone, DNPH reagent and thiourea were added after the addition of sulphuric acid. The tubes were cooled in ice and the absorbance was read at 540nm in a spectrophotometer (Genesys 10-S, USA). A standard graph was constructed using an electronic calculator set to the line regression mode. The concentrations of ascorbate in the samples were calculated and expressed in terms of mg/g of sample.(Onwuka ,2005)

Estimation of vitamin E

Procedure

Into 3 stoppered centrifuge tubes of 1.5ml of sample, 1.5ml of the standard and 1.5ml of water were pipetted out separately. To all the tubes, 1.5ml of ethanol and 1.5ml of xylene were added, mixed well and centrifuged. Xylene (1.0ml) layer was transferred into another stoppered tube. To each tube, 1.0ml of dipyrindyl reagent was added and mixed well. The mixture (1.5ml) was pipetted out into a cuvette and the extinction was read at 460nm. Ferric chloride solution (0.33ml) was added to all the tubes and mixed well. The red colour developed was read exactly after 15 minutes at 520nm in a spectrophotometer (Genesys 10-S, USA) (Onwuka, 2005) The concentration of tocopherol in the sample was calculated using the formula,
Tocopherols (µg) = Sample A₅₂₀ – A₄₆₀ × 0.29 × 0.15

Standard A₅₂₀

RESULTS

Table 1: Proximate composition of *Dioscorea villosa* used in the study.

Contents	%
Fibre	0.56
Fat	6.54
Ash	3.14
Moisture	7.20
Protein	11.73
Carbohydrate	70.82

Table 2: Vitamins content of *Dioscorea villosa* used in the study

Vitamins	concentration (mg/kg)
Retinol	7.04±0.01
Tocopherol	14.41±0.00
Ascorbic acid	68.76±0.00

Calciferol	3.56±0.00
Thiamine	0.03±0.01
Riboflavin	0.01±0.00
Niacin	0.57±0.01
Pyridoxine	0.24±0.00
<u>Cobalamin</u>	<u>3.42±0.01</u>

DISCUSSION

In Table 4.1, statistical analysis showed significant differences in the means of the proximate composition. The fat content was found out to be 6.54% which is the same with that of Afiukwa *et al.*(2013) who reported that it is higher than 0.17% for yams using USDA National Nutrient Database and many times higher than 0.2-0.4% for common cultivated yams in Nigeria. Because of the very high fat content of *Dioscorea villosa*, there is need to investigate the quality of the fat as part of the safety assessment as compared with earlier researchers. The ash content was found out to be 3.14 % which indicates that the samples could be good sources of nutritionally essential mineral and trace elements (Osagie,1992). The results of our research work showed that the moisture value was found out to be 7.20% which play a very important role in determining the susceptibility of the crops to microbial spoilage and maintaining the shelf life of produce. Thus, species and varieties with low moisture content have longer shelf life and are more suitable for prolonged storage (Polycarp *et al.*, 2012). Fibre content was found out to be 0.57% which plays a vital role in the digestive system of humans as well as animals. The finding is in line with the findings of some earlier researchers. The result of our research showed that fiber increases digestibility of nutrients making it easier for absorption and digestion. Researchers like Dhingra *et al.* (2012) reported that adequate intake of fiber increases water holding capacity, aids in regular bowel movement, fecal bulkiness and less intestinal transit. It also promotes beneficial physiological effects such as reduction of blood sugar and cholesterol level, trapping of toxic substances and encourages the growth of natural microbial flora in the gut. Protein was found out to be 11.73% and is an essential nutrient required for growth and organ development in humans and animals. From the research work, since people of Sub Saharan are basically farmers and consume a lot of *D. villosa* which is majorly their source of energy. So, they were able to increase their productivity annually through manual means of cultivation and our finding is in line with the finding of some authors in Africa which suggest that consumption of starchy staples primarily yam and cassava contributes to a great proportion of protein intake in the region ranging from 5.9% in the Southern and Eastern Africa to 15.9% in west Africa.

The result in Table 2 revealed that yam tuber is a natural source of vitamin B group and could contribute to part of the daily requirements of pyridoxine (vitamin B6), thiamine (vitamin B1), riboflavin (vitamin B₂), cobalamin (vitamin B12) and Niacin (vitamin B3) which are involved in various metabolic routes and could supply approximately 29% of the recommended levels of vitamin C (ascorbic acid) which plays an important role in the reinforcement of immune function , wound healing, bone growth and also slows cell aging (Ramirez *et al*, 2019)

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