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Article

Proximate and Anti-nutritional Analysis of the Leaves of *Vitex doniana*

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Abstract: The search for more nutrition sources among forest products has called for the analysis of nutritional and anti – nutritional composition of *Vitex doniana.* Although, *Vitex doniana* had been known to be very useful among people across tropical Africa and beyond, but there is lack of adequate data on its nutritional composition. The present study designed for determination of proximate, nutritional, and also anti-nutritional analysis of the leaves of V*itex doniana*. Proximate compositions revealed that the leaves of *Vitex doniana* contain moisture content of 4.616±0.005 g, Ash content was 2.027±0.003 g, crude lipids content was3.623±0.168 g, Crude fibre content was 4.991±0.002 g, crude protein 3.423±0.033 g, and carbohydrate content found to be 36.515±0.000g. The anti-nutritional Analysis of *Vitex doniana* leaves revealed that, the level of the oxalate was 19.200 ± 0.280, Nitrate 287.670±6.489, Tannins 91.100±1.100, Cyanide 228.382±0.130, Phytate 20.420±0.140 respectively. It is concluded that the young leaf is highly rich in nutrients and contains the nutrient levels that fall within other popular edible vegetables. It is therefore recommended for human consumption in every household. It is also suggested that further research should be carried out on its economic status and feasibility of the leaves as feed supplement in animal feed.

Keywords: Proximate, Anti-nutritional, Leaves of *Vitex doniana*, Nutritional, and Micronutrients.

1. Introduction

The (*Vitex doniana)* leaves have been recognized as rich sources of micronutrients (minerals and vitamins) and antioxidants (Aberoumand and Deokule, 2009). In most developing nations where food shortage and famine is mostly experienced, green leafy vegetables are the means of livelihood; Niger famine of 2005 is a clear evidence, in which many populace depend on leaves of Anza or Dilo (BosciaSenegegalensis) and Roselle leaves as a means of survival. Vegetables are generally high in carbohydrate and low in proteins. However, their nutritive value depends on the type of soil and climate in which they are grown (Adeyeye, 2012). Vegetables are important food and highly beneficial for the maintenance of health and prevention of diseases. They contain valuable food ingredients which can be successfully utilized to build up and repair the body. They are valued mainly for their high carbohydrate, vitamins and mineral contents. Researchers have repeatedly observed health benefits associated with high fruit and vegetables consumption (Adeyeye, 2012).

Balckplum (*Vitex doniana* sweet) of the family *verbanaceae* is a tree crop that grows in open woodland and savannah regions of tropical Africa; it is the commonest of the *vitex* species in West Africa (Adeyeye, E.I. and Otoketi, 2019). It produces fruits which are plum-like, sweet and edible. The fruit is green when mature and changes to dark brown when fully ripe, with the purple surrounding a hard stone containing one seed (Agbede and Ibitoye, 2007).

2. Materials and Method

2.1. Sample Collection

The sample were obtained from Zuru local government, Kebbi state and was identified by Prof. Dharmendra Sigh of biological department Faculty of Life sciences, Kebbi State University of Science and Technology Aliero (KSUSTA).

2.2. Sample Treatment

The *Vitex doniana* was dried and dried at room temperature and ground into powder using Philips blender. The powdered were kept in air-tight container for further analysis.

2.3. Proximate Analysis

Proximate Analysis is a partitioning of compound in a feed in to six categories based on the chemical properties of the compounds. The six categories are: moisture, ash crude, protein (or Kjaldahl protein). Crude lipid, crude fiber and nitrogen-free extracts (digestible carbohydrates).

2.3.1. Moisture content determination

The moisture content of the all samples were measured as described by AOAC method (AOAC 1980). Crucibles were thoroughly washed and dried in an oven at 100°C for 30min and allow to cool inside desiccators. After cooling, they were weighed as W1. Then, 2.0g of the finely ground samples were put into crucibles and weighed as W_2. Thereafter, the sample plus the crucibles were placed inside the oven and dried at 100°C for 4 hours, cooled and weighed at the same temperature for 30 minutes until constant were obtained as W_3. Then, the moisture content of the sample was calculated.

$$\text{Moisture (\%)} = \frac{W2-W3}{W2-W1} \times 100$$

Where, W_1= Initial weight of empty crucible,
W_2 = Weight of crucible + sample before drying
W_3 = Final weight of crucible + sample after drying.

2.3.2. Ash content determination

Total ash of the sample were determined by Furnace Incineration described by AOAC Method (AOAC, 1980). Out of the finely ground dried samples, 2.0g was weighed into porcelain crucibles and incinerated at 600°C for 6 hours in an ashing muffle furnace (Model 1184A Fisher Scientific, Houston, TX) until ash was obtained. The ash was cooled in a desiccator and reweighed. The ash content was obtained

$$\text{Ash (\%)} = \frac{\text{Weight of Ash}}{\text{Weight of original sample}} \times 100$$

2.3.3. Crude protein determination

The crude protein content of the sample was determined using the Micro Kjeldahl methods of AOAC (AOAC, 1980) which involved protein digestion and distillation. **Protein Digestion**: Out of the rice sample, 0.15g was weighed into a Kjeldahl flask, 0.8g tablets of Kjeldah Catalyst were added, 2 mL concentrated Sulphuric acid was introduced. The whole mixture was subjected to heating in the fume cupboard. The heating was done gently at first and increased with occasional shaking till the solution assumed a green colour. The temperature of the digestion was 420°C for about 30min. The solution was cooled and black particles showing at the neck of the flask were washed down with distilled water. The solution was re-heated gently at first until the green colour disappeared. Then, it was cooled. After cooling, the digest was transferred into a 250 mL volumetric flask. **Protein distillation**: Out of the digested samples, 5.0 mL was pipetted into the body of the apparatus via a small funnel aperture. The digest was washed down with distilled water followed by addition of 15ml of 40% NaOH solution. The digest in the condenser was steamed for 1 -5 minutes after which enough ammonium sulphate was collected. The receiving flask was removed and the tip of the condenser washed down into the flask after

which the condensed water was removed. The solution in the receiving flask was treated with 0.01 m hydrochloric acid. Also, a blank was run through along with the sample. After titration, the % nitrogen was calculated using the formulae below:

$$\text{Nitrogen}(\%) = \frac{V_S\ VB\ M(acid)X0.01401}{W_t} \times 100$$

where, Vs = Volume (ml) of acid required to titrate sample

VB = Volume (ml) of acid required to titrate the blank

M (acid) = Molarity of acid

Wt = Weight of sample (g)

Then, percentage crude protein in the sample was calculated from the Nitrogen as:

Crude protein (%) = % N X F

where, F (conversion factor), is equivalent to 6.25.

2.3.4. Crude fat determination

The Fat content was determined as described by AOAC Method (AOAC, 1980). Cleaned boiling flasks (250 mL) were dried in an oven at 105°C- 110°C for about 30 min and cooled in desiccator. Approximately 2.0g of samples were weighed accurately into labelled thimbles. The dried boiling flasks were weighed correspondingly and filled with about 300mL of petroleum ether (boiling point 40 - 60°C). The extraction thimbles were plugged tightly with cotton wool. After that, the Soxhlet apparatus was assembled and allowed to reflux for 6 hours. The thimble was removed with care and petroleum ether collected from the top container and drained into another container for re-use. After that, the flask was dried at 105 - 110 for 1 hour. After drying, it was cooled in desiccators and weighed. Then, % fat in the sample were calculated.

$$\text{Fat}(\%) = \frac{\text{Weight of fat}}{\text{Weight of sample}} \times 100$$

2.3.5. Crude fibre determination

Crude fibre was determined using the method of AOAC (AOAC, 1980). Out of the sample, 2.0g was hydrolyzed in the beaker with petroleum ether after which it was boiled under reflux for 30 min with 200 ml of a solution containing 1.25% H2SO4 per 100 ml of solution. The solution was filtered through a filter paper, after filtration the samples were washed with boiled water until they were no longer acidic. Then, the residue was transferred onto a beaker and boiled for another 30min with 200ml of solution containing 1.25 % NaOH per 100 ml. The boiled samples were washed with boiled distilled

water. The residues were filtered through Gooch filter crucible, dried at 100°C for 2 hours in an oven, cooled and washed. The percentage crude fibre was calculated.

$$\text{Crude fibre (\%)} = = \frac{\text{Wt.After drying}}{\text{Weight of sample}} \times 100$$

2.3.6. Carbohydrate determination

The total percentage of carbohydrate content in the sample was determined by the difference as reported by Onyeike *et al.* (1995). This method involved adding the total values of crude protein, lipid, crude fibre, moisture and ash constituents of the sample and subtracting it from 100. The value obtained is the percentage carbohydrate constituent of the sample.

Thus: % carbohydrate = 100 (% moisture + % crude fibre + % protein + % lipid + % ash).

2.4. Determination of Anti-Nutrients

The anti-nutritional component: can be defined as those substances generated in natural food substances by the normal metabolism of species and by different mechanism (e.g. inactivation of some nutrients, diminution of the digestive process or metabolic utilization of feed) which exert effect contrary to optimum nutrition. They are: Alkaloid, Tannin, Cyanide, Oxalate and Phytate. Jacobs, M.B (2019).

2.4.1. Determination of nitrate

0.1g of powder sample was added into 100ml conical flask, 10ml of distilled water was added and boil for 30 minutes and filtered using filter paper.

(% Nitrate mg%) = Absorbance of sample x concentration of standard/Absorbance of standard

2.4.2. Determination of tannins

Tannin was determined by the method of (Trease, and Evans 1978). Powdered sample (100g) was put into conical flask, 50ml of distilled water H_2O were added and boil for 30 minutes, in a boiling water and filter paper. The 50ml volume of distil water H_2O was mixed and incubated for 20 minutes at room temperature and the absorbance was measured at 760nm.

(% Tannins mg %) = Absorbance of sample x concentration of standard/Absorbance of standard

2.4.3. Determination of cyanide

0.5g of powder sample was measured into 100ml of conical flask and 50ml of distilled water DH_2O was added and boil for 30 minutes and filtered using filter paper (Trense and Evans, 1978).

(% Cyanide mg %) = Absorbance of sample x concentration of standard/Absorbance of standard

2.4.4. Determination of oxalate

The method of Krishna and Ranjhan (1980) was adopted for the determination of total oxalate.

0.0045 (%Oxalate g%) = Titre value x 0.0045

2.4.5. Determination of phytate

2g of the sample was soked in 100ml of 2% HCl for 3 hours, and filtered, 25ml of the filterate, 5ml of 0.3%NH_4SCN, and 53ml of Distilled water, H_2O were mixed together and titrate against 0.01M standard faric.

3. Results and Discussion

Composition of *Vitex doniana* leaves. The leaves showed moisture content. 4.616±0.005 g, Ash content was 2.027±0.003 g. Crude lipids content 3.623±0.168 g, Crude fibre content. 4.991±0.002 g. Crude protein 3.423±0.033 g, carbohydrate 36.515±0.000g respectively.

The anti-nutritional Analysis of *Vitex doniana* leaves. The level of the oxalate was 19.200 ± 0.280, Nitrate 287.670±6.489, Tannins 91.100±1.100, Cyanide 228.382±0.130, Phytate 20.420±0.140 respectively.

Table 3.1: Represents the result of the proximate, nutritional of leaves of *vitex doniana*

PARAMETER	**RESULTS (%)**
Ash content	2.027±0.003
Moisture Content	4.616±0.005
Lipid content	3.623±0.168
Crude fiber	4.991±0.002
Crude protein	3.423±0.033
Carbohydrate	36.515±0.000

Results are presented as Mean ± Standard error of mean of triplicate results.

Table 3.2: The result Anti-nutritional analysis of leaves of *Vitex doniana*

PARAMETER	RESULTS (mg%)
Oxalate	19.200 ± 0.280
Nitrate	287.670±6.489
Tannins	91.100±1.100
Cyanide	228.382±0.130
Phytate	20.420±0.140

Results are presented as Mean ± Standard error of mean of triplicate results

Moisture content obtained from V. doniana leaves is 4.616% was lower than 16.66% reported in the work Nnamani et al., (2009). Ash content is a measure of the total mineral content of a food. The sample analysed had a value of 2.027%, is low compared to 5.27% reported by Agbede and Ibitoye (2007). The differences could be due to environmental factors which may be due to different of soil. Crude protein of V. doniana was 3.423%, which is lower to 10.0 % reported by Nnamani et al. (2009).

Dreon et al. (1990) showed that most leaves had high carbohydrate content depending on the fruit type, maturity and environment. However V. doniana is on the contrary by having a slightly lower value of 36.515%. The value was also low when compared to 67.0% reported by V. doniana leaves (Nnamani et al., 2009). Crude fibre obtained from V. doniana fruit (4.991%) was grossly lower than 15.0% reported for V. doniana leaves (Nnamani et al., 2009).

Oxalate is an anti-nutrient responsible for kidney stone, electrolyte imbalance and irritation of digestive system in man and animal (Egbuna, and Ifemeje, 2015) dried V. doniana leaves has oxalate (19.200 ± 0.280 mig/iml) which is quite low. Oxalates affect calcium and magnesium metabolism and react with proteins to form complexes which have an inhibitory effect in peptic digestion (Akande et al., 2010). Phytic acid (20.420±0.140 mg/ml). Cyanide in V. doniana leaves (228.382±0.130 mg/ml) is considered to be non-toxic when ingested due to its very small amount. While Tannins in V. doniana leaves 91.100±1.100 and Nitrate in V. doniana leaves 287.670±6.489.

Thus, the results revealed that the astinutrient composition of V. doniana leaves were generally low such that none of the anti-nutrients was above the lethal dosage approved by standard bodies like National Agency for Food and Drugs Administration and Control (NAFDAC) in Nigeria (Dreon et, al 1990).

4. Conclusion and Recommendation

4.1. Conclusion

This research work showed that *Vitex doniana* tender leaves are good sources of minerals which if consumed in sufficient amount could contribute greatly towards meeting human nutritional requirements for normal body growth and adequate protection against diseases arising from malnutrition.

4.2. Recommendation

From the result, *Vitex doniana* leaves are recommended for continued use for nutritional purposes, considering to the amount and diversity of nutrients it contains. Chemical analysis alone however, should not be the exclusive criteria for judging the nutritional significance of a plant parts. Thus, it becomes necessary to consider other aspects such as presence antinutritional/toxicological factors and biological evaluation of nutrients content.

In view of all findings, the antinutritional and toxicological analysis of the leaves are research worthy.

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