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Proximate, phytochemical screening and mineral analysis of *Crescentia cujete L.* leaves

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Background: One of the uses of *Crescentia cujete* leaves is in the treatment of cardiovascular diseases, which necessitated the evaluation of its chemical composition.

Aim: The aim of the study was to investigate the chemical composition of *C. cujete* leaves.

Setting: Fresh, mature leaves were harvested from the Herbal Garden, Forestry Research Institute of Nigeria, Jericho Hill, Ibadan, Nigeria. The plant samples were identified at Forest Herbarium, Ibadan, Nigeria, sorted to eliminate extraneous matter and then air-dried on a cabinet dryer for five days at ambient temperature and oven-dried at 40°C for five minutes. The dried leaves were milled to powder using a medium-sized milling machine and stored in an air-tight container until needed for analysis.

Methods: The proximate composition, phytochemical screening and mineral analysis were determined using standard analytic methods. Minerals such as iron, zinc, calcium, manganese and magnesium were determined by the atomic absorption spectrophotometer; sodium and potassium by the flame photometer and phosphorus by the spectrophotometer.

Results: The proximate analysis results revealed that the samples contained carbohydrate (40.46%), crude protein (16.15%), crude fibre (19.71%), crude fat (2.91%), ash (9.77%) and moisture (11.03%), while the phytochemical screening confirmed the presence of cardiac glycosides, saponins, tannins, anthraquinones and flavonoids. The mineral compositions (mg/g) decreased in the order Ca (6.0) > K (5.0) > Mg (2.2) > P (1.5) > Na (0.2) > Fe = Zn (0.6) > Cu = Mn (0.03).

Conclusion: These results conceivably indicate that *C. cujete* is a good source of essential minerals and phyto-nutrients which possess strong pharmacological activities, providing scientific credence for its therapeutic usage in folklore medicine.

Introduction

Medicinal plants are the source of many important drugs of the modern world. Many indigenous medicinal plants are also used as spices in food (Okwu 1999, 2001). In developing countries, medicinal plants are cheaper with lower incidence of adverse effects after use and more accessible to most people than orthodox medicine which might be reasons for their worldwide attention and use as reported (Sofowora 1993). The phyto-constituents (phenols, anthraquinones, alkaloids, glycosides, flavonoids and saponins) found to be distributed in plants are antibiotic principles of plants, but these compounds were not well established owing to lack of knowledge and techniques as reported (Hafiza et al. 2002).

Crescentia cujete, commonly known as the calabash tree, is a flowering plant and native to Central and South America. It is an evergreen tropical tree belonging to the Bignoniaceae family, which can grow up to a height of 7.62 m and produce fruits up to 25 cm in diameter. It has simple leaves and the bark of the tree is rough. The fruit is large, hard and green in colour, which takes about six months to ripen (Gentry 1996). The seeds of the fruit are flat, small and are embedded in the pulp (Elias & Prance 1978). The pulp of the fruit has medicinal properties and acts as a remedy for respiratory problems such as asthma and cough. The syrup prepared from the pulp is used as a medicine for relieving disorders of the chest or respiratory tract and also to cure dysentery and stomach ache. The leaves of the calabash tree are used to lower blood pressure, while decoction of the tree bark is used to clean wounds and also to treat haematomas and tumours. The fruit pulp contains crescentic, tartaric, citric, tannic, chlorogenic and hydrocyanic acids, and the seed oil contains oleic acid (Gentry 1996). The seed can be eaten when cooked and can also be used to make syrup and beverage. According of Ejelonu et al. (2011), the fruits of *C. cujete* can make useful contributions to human and animal health as a result of the presence of phytochemicals observed

in the fruit sample. Other parts of *C. cujete* tree have been reported to have medicinal applications but that of its leaves has not been well-documented, and hence the need for investigation of its chemical compositions.

Materials and methods
Sample collection and preparation

Fresh and matured leaves of *C. cujete* were harvested from the Herbal Garden, Forestry Research Institute of Nigeria, Jericho Hill, Ibadan, Nigeria. The plant samples were identified by Mr Chukwuma at the Forestry Herbarium Ibadan (FHI), Nigeria. The samples were sorted to eliminate all extraneous matter and then air-dried on a cabinet dryer for five days at room temperature to drastically reduce their moisture content. After five days of room drying, samples were further oven-dried at 40°C for 5 min to further reduce the moisture content. The dried leaves were then milled to powder using a medium-sized laboratory milling machine and stored in an air-tight container until needed for both quantitative and qualitative analyses.

Proximate analysis

The powdered samples of *C. cujete* were analysed for moisture, protein, fat, ash and fibre by standard analytical methods (AOAC 2005) while carbohydrate was calculated by difference (100 - [% moisture + % protein + % fat + % ash + % fibre]).

Determination of moisture

Moisture was determined by oven drying, where 2 g of well-mixed sample was accurately weighed in a clean and dry crucible (W_1). The crucible was placed into an oven at 105°C for 12 h until a constant weight was achieved. Then, the crucible was placed in a desiccator for 30 min to cool. After cooling, it was weighed again (W_2). The percentage of moisture was calculated in equation 1:

$$\% \text{ Moisture} = \frac{W_1 - W_2 \times 100}{\text{Weight of sample}} \qquad \text{[Eqn 1]}$$

where W_1 (initial weight of crucible + sample) and W_2 (final weight of crucible + sample)

Determination of ash

Empty crucibles were cleaned and then heated in a muffle furnace at 600°C for 1 h. The crucibles were cooled in a desiccator and then reweighed as (W_1). Sample (1 g) was weighed into crucible (W_2); the crucible was then placed in muffle furnace at 550°C for 24 h. The appearance of grey white ash indicated complete oxidation of all organic matter in the sample. After ashing, the crucible was cooled and weighed (W_3). Percentage of ash was calculated in equation 2:

$$\% \text{ Ash} = \frac{W_3 - W_1 / W_2 - W_1 \times 100}{\text{Weight of sample}} \qquad \text{[Eqn 2]}$$

$W_1 - W_3$ = Difference in weight of ash

Determination of crude protein

Protein in the sample was determined by the Kjeldahl method where powdered sample (1 g) was taken into a digestion flask containing 10 mL of concentrated H_2SO_4 and 8 g of digestion mixture (K_2SO_4, $CuSO_4$ [8: 1]). The flask was swirled for 2 h in order to mix the contents thoroughly and then placed on a heater to start digestion till the mixture became clear. The digest was cooled and transferred to a 100 mL volumetric flask and volume was made up to mark by the addition of distilled water. Distillation of the digest was performed in Markam Still Distillation Apparatus for at least 10 min. The digest (10 mL) was introduced into the distillation tube and then 10 mL of 0.5 N NaOH was gradually added in the same way. The NH_3 produced was collected as NH_4OH in a conical flask containing 20 mL of 4% boric acid solution with few drops of modified methyl red indicator. During distillation, a yellowish colour appeared because of the NH_4OH. The distillate was then titrated against 0.1 N HCl standard solution until the solution turned pink. A blank was also run through all steps as above. The percentage of crude protein content of the sample was calculated using equations 3 and 4:

$$\% \ N = \frac{(S - B) \times N \times 0.014 \times D \times 100}{\text{Weight of sample} \times V} \qquad \text{[Eqn 3]}$$

% Crude Protein = 6.25* × % N (*, correction factor)   [Eqn 4]

where S is the sample titration reading, B is the blank titration reading, N is the normality of HCl, D is the dilution of sample after digestion, V is the volume taken for distillation and 0.014 is the milliequivalent weight of nitrogen.

Determination of crude fat

Crude fat was determined by ether extract method using Soxhlet apparatus where powdered sample (1 g) was wrapped in filter paper, placed in a fat-free thimble and then added to the extraction tube. The weighed, cleaned and dried receiving beaker was filled with petroleum ether and fitted into the apparatus and then water and heater were turned on to start the extraction. After 6 rounds of siphoning, ether was allowed to evaporate and the beaker was disconnected before the last siphoning. The extract was then transferred into a clean glass dish where the ether was washed and evaporated on the water bath. The dish was then placed in an oven at 105°C for 2 h and cooled in a desiccator. The crude fat was determined using equation 5:

$$\% \text{ Crude fat} = \frac{\text{Weight of ether extract} \times 100}{\text{Weight of sample}} \qquad \text{[Eqn 5]}$$

Determination of crude fibre

This was done on moisture-free, ether-extracted powdered sample where 0.2 g of the sample was weighed (W_0) and transferred to a porous crucible with the crucible then placed into a Dosi-Fibre Unit with the valve kept in the 'OFF' position. After that, 150 mL of preheated H_2SO_4 solution was added and

some drops of acetone (foam suppresser) were added to each column. Then, the cooling circuit was opened and the heating elements turned on (power at 90%). When it started boiling, the power was reduced to 30% and left for 30 min. Valves were opened for drainage of acid and rinsed with distilled water thrice to completely ensure the removal of acid from the sample. The same procedure was used for alkali digestion by using KOH instead of H_2SO_4. The sample was dried in an oven at 105°C for 1 h until a constant weight was attained. Then, the sample was allowed to cool in a desiccator and weighed (W_1). The sample crucibles were ashed in a muffle furnace at 550°C for 4 h. After ashing, samples were removed from the furnace and cooled in a desiccator and weighed (W_2). Calculation was done by using equation 6:

$$\% \text{ Crude Fibre} = \frac{W_1 - W_2 \times 100}{\text{Weight of sample}} \qquad \text{[Eqn 6]}$$

Determination of nitrogen-free extract
Carbohydrate was calculated by difference after analysis of all the other items using the formula: NFE = 100 - (% moisture + % crude protein + % crude fat + % crude fibre + % ash).

Qualitative screening of phytochemical constituents of *Crescentia cujete* leaves
Phytochemical screenings were carried out on the powdered sample using standard procedures to confirm the presence of constituents (alkaloids, anthraquinones, flavonoids, saponins, tannins, steroids, cardiac glycosides and total phenolic compounds) as described by Harborne (1993), Trease and Evans (1989) and Sofowora (1993).

Test for saponins: Powdered sample (1 g) was boiled with 10 mL of distilled water in a bottle bath for 10 min. The mixture was filtered while hot and allowed to cool. The following tests were then carried out:

- Demonstration of frothing: 2.5 mL of filtrate was diluted to 10 mL with distilled water and shaken vigorously for 2 min (frothing indicated the presence of saponins in the filtrate).
- Demonstration of emulsifying properties: Two drops of olive oil were added to the solution obtained from diluting 2.5 mL of filtrate to 10 mL with distilled water (as above), then shaken vigorously for a few minutes. The formation of persistent foam was evidence of the presence of saponins.

Test for alkaloids: Sample (1 g) was stirred in 10 mL of concentrated HCl on a steam bath followed by filtration. Filtrate (1 mL) was mixed with two drops of Wagner's reagent, then two drops of Dragendorff's reagent were added to another 1 mL of the filtrate, and the mixtures were then observed for turbidity.

Test for tannins: Powdered sample (1 g) was boiled with 20 mL of distilled water in a water bath and was filtered while hot. Cooled filtrate (1 mL) was distilled to 5 mL with distilled water and two to three drops of 10% ferric chloride were added and observed for any formation of precipitates and any colour change. A bluish-black or brownish-green precipitate indicated the presence of tannins.

Test for flavonoids: Sample (1 g) was boiled with 10 mL of ethanol, and two drops of ferric chloride were added to 5 mL of the extract. The mixture was observed for a dusty green colouration as positive result.

Test for free anthraquinones: Sample (0.5 g) was shaken with 5 mL of chloroform for 10 min, filtered and 5 mL of 10% ammonium solution was added to the filtrate. The mixture was shaken and the presence of a pink, red or violent colour in the ammonia phase indicated the presence of free anthraquinones.

Test for cardiac glycoside: Sample (1 g) was extracted with 10 mL of 80% ethanol for 5 min on a water bath. The extract was filtered and diluted with equal volume of distilled water and two drops of lead acetate solution were added, shaken and filtered after standing for few minutes. The filtrate was then extracted with aliquots of chloroform, and the extract was dissolved in 2 mL of glacial acetic acid containing one drop of $FeCl_3$ solution in a clean test tube. Concentrated H_2SO_4 acid (2 mL) was then poured down the side of the tube so as to form a layer below the acetic acid. The formation of a reddish-brown or brown ring at the interface and a green colour in the acetic layer was taken for a positive result.

Test for total phenolics: Sample (1 g) was soaked in 25 mL of 2% of HCl for 1 h and then filtered through a 10 cm Whatman No. 1 filter paper. Plant extract (5 mL) was mixed with 1 mL of 0.30% ammonium thiocyanate solution and three drops of ferric chloride solution. A brownish-yellow colour indicated the presence of phenols.

Mineral analysis
Mineral contents of *C. cujete* were determined by atomic absorption and flame spectrometry according to standard methods (Harborne 1993). Wet digestion of sample was done by taking 1 g of the powdered sample in a digesting glass tube, to which 12 mL of HNO_3 was added, and keeping the mixture overnight at room temperature. Perchloric acid (4 mL) was then added to this mixture and digested in a fume block. The temperature was increased gradually, starting from 50°C to 300°C, where the appearance of white fumes indicated the completion of digestion. The mixture was left to cool down and the contents of the tubes were transferred to 100 mL volumetric flasks and the volumes of the contents were increased to 100 mL by adding distilled water. The wet digested solution was transferred to labelled plastics. The digest was stored and used for mineral determination. The determination of iron (Fe), zinc (Zn), calcium (Ca), manganese (Mn) and magnesium (Mg) was analysed using atomic absorption spectrophotometer (Hitachi model 170-10) in the Department of Bioscience, FRIN, Jericho Hills, Ibadan, Nigeria. Different electrode lamps were used for each mineral. The equipment was run for standard solutions of each mineral before and during determination to check if it

was working properly. The dilution factor for all minerals except P and Mg was 100. For determination of Mg, the original solution was further diluted using 0.5 mL original solution and adding distilled water to obtain a volume of up to 100 mL. For the determination of Ca, 1 mL of lithium oxide solution was added to the original solution to unmask Ca from Mg. The concentrations of minerals recorded were converted to mg of the minerals by multiplying the absorbency (in ppm) with a dilution factor and dividing by 1000 as in equation 7:

$$MW = \frac{\text{Absorbency (ppm)} \times \text{Dry weight} \times D}{\text{Weight of sample} \times 100}$$ [Eqn 7]

Note: Dilution factor for phosphorus is 2500, for magnesium 10 000 and for other minerals, including calcium, iron, potassium, sodium, manganese and chromium, is 100.

Determination of sodium (Na) and potassium (K) was done by flame photometry. Standard solutions of 20, 40, 60, 80 and 100 mg/mL were used both for Na and K. The calculations for the total mineral intake involve the same procedure as given in atomic absorption spectrophotometry above. Phosphorus (P) in the sample was determined by spectrophotometry by mixing 12 g of ammonium molybdate with 250 mL of distilled water in a beaker (solution A). Antimony potassium tartrate 0.2 g was taken and dissolved in 500 mL H_2SO_4 solution in a volumetric flask. Enough distilled water was added to make the solution up to 1000 mL (solution B). The two solutions were mixed in a 2000 mL volumetric flask to get the mix reagent. The volume of the mix reagent was increased to 2000 mL by adding distilled water. Ascorbic acid (0.7 g) was mixed with 140 mL of the mix reagent in a beaker and left until dissolved to make the colour reagent. Wet digested sample (1 g) was taken in a plastic bottle labelled properly and 4 mL of distilled water was added to make a diluted volume of 5 mL. Colour reagent (5 mL) was added to this volume and the total volume of this mixture (final solution) was increased to 25 mL. The dilution factor of this solution was 2500. After some time, the colour of this final solution turned blue. Sample from the final blue solution was taken in a cuvette and read using a spectrophotometer. The readings of phosphorus were recorded in ppm. The calculations for the total mineral intake involve the same procedure as given in atomic absorption spectrophotemtry.

Statistical analysis

Data were analysed using the descriptive statistical analyses where means and standard deviation (SD) were obtained using Microsoft Excel 2013 version. Results were expressed as mean ± SD. A value of $p < 0.05$ was used to denote statistical significance.

Results

The analyses of proximate composition of *C. cujete* leaves as shown in Table 1 revealed that carbohydrate was the most abundant (40.48 ± 0.11) followed by crude protein (16.08 ± 0.17), while crude fat was the least (2.92 ± 0.02).

TABLE 1: Proximate composition of *Crescentia cujete* leaves on dry basis.

Parameters	Quantity (%)
Moisture	11.04 ± 0.01
Crude protein	16.08 ± 0.17
Crude fibre	19.72 ± 0.03
Crude fat	2.92 ± 0.02
Ash	9.77 ± 0.01
Carbohydrate	40.48 ± 0.11

Values are expressed as mean ± how many SD in triplicates.

TABLE 2: Qualitative analysis of phytochemical composition of *Crescentia cujete* leaves on dry basis.

Components	Tests for phytochemical components	Observation	Inference
Alkaloids	1 g of sample + 10 mL of HCl + heat + filter + 2 drops of Dragendorff's reagents.	The resulting solution was not turbid.	Absent
Anthraquinones	0.5 g of sample + 5 mL of chloroform + filter + 5 mL of 10% NH_4 solution + shaken for 10 min.	No pink, violet, red solution was observed.	Absent
Cardiac glycosides	1 g of sample + 10 mL of 80% ethanol + heat + filter + 2 drops of lead acetate solution + filter + 2 mL of chloroform + 2 drops of glacial acetic acid + $FeCl_3$ solution + 2 mL conc H_2SO_4.	A brown ring at the interface and green colour in the acetic layer were observed.	Present
Flavonoids	1 g of sample + 10 mL of ethanol + heat + filter + 2 drops of $FeCl_3$ solution.	A dusty green colour was observed.	Present
Phenolic compounds	1 g of sample + 25 mL of 2% HCl + filter + 5 mL of extract + 1 mL of 0.3% NH_4 thiocyanate solution + 3 drops of $FeCl_3$ solution.	Brownish-yellow colouration was observed.	Present
Saponins	1 g of sample + 10 mL of distilled H_2O + heat + filter + 10 mL of distilled H_2O in 2.5 mL filtrate + 2 drops of olive oil + vigorous shaking for 2 min.	Persistent foam was observed.	Present
Tannins	1 g of sample + 20 mL of distilled H_2O + heat + filter + 5 mL of distilled H_2O + 2 drops of 10% $FeCl_3$ solution.	A brownish green precipitate indicated presence of tannins.	Present

TABLE 3: Mineral composition of *Crescentia cujete* leaves on dry basis.

Elements	Quantity (mg/100g)
Calcium	613 ± 0.012
Magnesium	207 ± 0.012
Phosphorus	150 ± 0.001
Potassium	513 ± 0.012
Sodium	21 ± 0.001
Copper	3 ± 0.001
Iron	61 ± 0.001
Manganese	3 ± 0.001
Zinc	61 ± 0.001

Values are expressed as mean ± SD in triplicates.

The result of phytochemical screening as shown in Table 2 confirmed the presence of cardiac glycosides, flavonoids, phenolic compounds, saponins and tannins, while alkaloids and anthraquinones were confirmed to be absent in *C. cujete* leaves.

Results for the mineral composition of *C. cujete* leaves shown in Table 3 revealed that the leaves contain minerals in trace amounts as evidenced by the concentrations obtained.

Discussion
Proximate analysis

The proximate analysis shows that moisture content (11.04% ± 0.01%) found in *C. cujete* leaves was higher than the values

reported for *Gnetum africanum* (9.18%) and *Telfairia occidentalis* (8.64%) by Dike (2010). However, the moisture contents obtained in this study for *C. cujete* leaves were far lower than the values obtained for *C. cujete* fruits (84.92%) by Ejelonu et al. (2011) and *Ficus capensis* leaves (28.80%) by Uzoekwe and Mohammed (2015). The low moisture content helps to prevent the leaves from spoilage by microorganism (Adeyeye & Adejuyo 1994). The ash content value of the leaves of *C. cujete* (9.2% ± 0.01%) was lower compared with the value for the *Momordica charantia* leaf (15.42%) as reported by Bakare et al. (2010). However, the value was higher than the values of 5.54% and 6.14% for *Urera trinervis* and *Hippocratea myriantha*, respectively (Andzouana & Mombouli 2012). The high ash content is an indication of the level of inorganic minerals and organic matter present in the leaves. The crude fat content of *C. cujete* (2.92% ± 0.02%) was lower than the amount found in *Costus afer* (15.0%) and *Cedrela odorata* (11.0%) as reported by Asekun, Asekunowo and Balogun (2013). The fat content of the leaves was low, and it can therefore be recommended as part of weight-reducing diets. Gordon and Kessel (2002) reported that low-fat foods reduce cholesterol level and obesity. The leaves of the plant investigated contained a high amount of crude fibre (19.72% ± 0.03%). The values were comparably higher than the 8.61% reported for *Amaranthus hybridus* (Akubugwo et al. 2007) and 7.63% reported for *Vernonia calvaona* (Ayoola & Adeyeye 2010). The crude fibre quantity in these leaves is desirable because adequate consumption of dietary fibre may aid digestion. Fibre softens stools and therefore prevents constipation (Ayoola & Adeyeye 2010). Dietary fibre is also important for lowering serum cholesterol level and reduces the risk of diseases such as coronary heart diseases, hypertension, diabetes and breast cancer (Ishida et al. 2000). The crude protein content obtained in the study (16.08% ± 0.17%) was high compared with the 1.98% reported for *Securinega virosa* leaves (Danlami, David & Thomas 2012), but considerably lower than some other vegetables like *Ocimum gratissimum*, *Gnetum africanum* and *Vernonia amygdalina* (Dike 2010). The value obtained for carbohydrate in *C. cujete* (40.48% ± 0.11%) was higher than the values obtained for *Urera trimervis* (6.07%) and *Mucuna poggei* (11.55%) (Oko et al. 2012; Sofowora 1993), but was lower when compared with the 54.71%, 55.75%, 59.70%, 53.30% and 54.72% obtained for *Morinda lucida, Landolphia hirsuta, Elaeis guineensis, Pterocarpus soyauxii* and *Vernonia amygdalina,* respectively (Dike 2000). The results obtained indicate that the leaves of *C. cujete* are a good source of fibre, minerals (ash), protein and energy, but not a good source of edible fat.

Phytochemical screening

The presence of phytochemicals such as saponins, cardiac glycosides, flavonoids, phenols and tannins in the leaves of *C. cujete* reported in this study suggests its use as a medicinal plant, which is in consonance with the medicinal properties of secondary plant metabolites (Banso & Adeyemi 2007; Gill 1992; Oyeyemi, Arowosegbe & Adebiyi 2014; Temitope & Omotayo 2012). The phytochemical profile obtained in this study was similar to the reports of Annapoorani and Manimegalai (2013) and Santhi et al. (2011), but differs from their reports by the absence of alkaloids and anthraquinone in *C. cujete*. Sodipo, Akiniyi and Ogunbamosu (2000) reported that saponins lower cholesterol level and act as an immune booster and anti-carcinogenic agent. However, it was also reported that high level of saponins may cause gastroenteritis (Awe & Sodipo 2001). Saponins play a major role in the treatment of inflamed tissues as well as in the prevention of cancer (Okwu & Eminike 2006).Tannins are known to have antiviral, antifungal, antibacterial and anti-tumour properties (Sofowora 2006); hence, the presence of tannins in *C. cujete* strongly supports its use in wound treatments, tumours, malaria and virginal discharge (Farquar 1996; Taylor 2005). The presence of flavonoids in the leaves of *C. cujete* also indicates that the leaves of the plants have biological functions such as being anti-oxidant and anti-tumour (Okwu 2004). In addition, this supports the anti-inflammatory activity of these plants (Lipkin & Newmark 1999), and hence their use in the treatment of wounds, haemorrhoids, liver inflammation, rheumatism and pains. Cardiac glycosides act on the heart muscles and increase renal flow (Naga Raju et al. 2013). The results obtained in this study corroborate the work of Ejelonu et al. (2011) which confirmed *C. cujete* fruits as potential sources of phytochemicals.

Mineral analysis

The mineral content in the *C. cujete* leaves showed that calcium had the highest value of 6.13 mg/100 g ± 0.012 mg/100 g. Calcium is an essential structural and functional element in living cells. The National Research Commission (1989) reported that calcium helps in building and maintaining bone mass and strength. Similarly, according to the report of Hassan, Abdulrahaman and Zuru (2004), the intake of calcium has been found to be very important and safer for cancer patients than some chemotherapeutic agents that can cause osteopenia and osteoporosis. Phosphorus content was found to be 1.50 mg/100 g ± 0.001 mg/100 g. Phosphorus plays an important role in normal kidney functioning, transfer of nerve impulse and ionic balance in the blood and tissues as reported by Ahmed and Chandhary (2009). Magnesium content of the leaves was found to be 2.07 m/100 g ± 0.012 mg/100 g, which was relatively lower than 56.05 mg/100 g ± 0.012 mg/100 g reported by Oko et al. (2012) for *Mucuna poggei* leaves and 2.56 mg/100 g for *Diospyros mespiliformis* (Hassan et al. 2004). Magnesium is an activator of coenzymes in carbohydrates and protein metabolism as claimed by Vormann (2003). The value of sodium in the sample, which is required in the body only in a small quantity, was 0.21 mg/100 g ± 0.001 mg/100 g. A recent report by Shomar (2012) indicated that sodium and potassium found in the intracellular and extracellular fluid in humans help to maintain electrolyte balance and membrane fluidity. Ghani et al. (2012) reported that manganese acts as a catalyst and co-factor in many enzymatic processes like muco-polysaccharide and glycoprotein syntheses, involved in the synthesis of fatty acids and cholesterol. Zinc is an essential mineral element for growth, development and for the function of the cells of the immune system (Deshpande,

Joshi & Giri 2013). It is used in the prevention and treatment of diarrhoea, pneumonia, cold, respiratory infections and malaria. According to McLaughlin, Parker and Clarke (1999), copper is an essential nutrient required for a wide range of biological functions such as enzymatic and redox reactions.

Conclusion

This study revealed the presence of phytochemical constituents in the leaves of *C. cujete* which are known to be responsible for the strong medicinal values providing scientific credence for its therapeutic usage in folklore medicine. The proximate and elemental analyses afford them interesting nutritional properties; hence, the plant leaves can contribute to the nutritional and energy requirements of humans.

Acknowledgements

Competing interests

Authors' contributions

M.B.O. designed the study, coordinated plant material storage, carried out all the experiments and drafted the manuscript. I.O.L. was responsible for the collection of plant materials and identification, supervised the laboratory experiments and made substantial contributions to revise the manuscript critically. A.A.O. performed data analysis and edited the manuscript. All authors read and approved the final manuscript.

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