Assessment On Nutritional Profile Of Ethanol Leaf-Extract Of Phyllanthus Reticulatus (Poir) From Abakaliki In Ebonyi State Of Nigeria

Ominyi¹, Matthias C; Ali¹, Fredrick U; Ebenyi¹, Lilian N; Nwali², Basil U; Uraku², Anayo J; Ibina¹, Agatha N.

Department Of Biotechnology, Ebonyi State University, P.M.B. 053 Abakaliki, Nigeria. Department Of Biochemistry, Ebonyi State University, P.M.B. 053 Abakaliki Nigeria.

Abstract

This study was carried out to evaluate the nutritional composition of ethanol leaf-extract of Phyllanthus reticulatus collected from Abakaliki in Ebonyi State of Nigeria. The leaves were cleaned, dried and processed for analysis. Exactly 500 g of the sample was immersed in 1500 mL of absolute ethanol for 3 days and filtered afterwards. The extract was concentrated to dryness using rotary evaporator and then analyzed for proximate, vitamins and mineral compositions by standard methods. The results of proximate composition showed significantly (P < 0.05) higher percentage of carbohydrate ($67.63^{a}\pm0.20$ %) followed by crude fats ($11.11^{b}\pm0.34$) and proteins ($9.84^{c}\pm0.18$). Vitamin A was significantly (P < 0.05) high ($394.58^{a}\pm0.04$ mg/100g) followed by vitamin C ($84.05^{b}\pm0.03$) and then vitamin E ($83.87^{c}\pm0.05$) while vitamins B_{2} , B_{3} and B_{1} were significantly low at P < 0.05, ($3.27^{d}\pm0.06$ mg/100g), ($2.92^{e}\pm0.04$ mg/100g) and ($2.83^{f}\pm0.001$ mg/100g) respectively in a descending order. Magnesium was significantly (P < 0.05) most abundant mineral ($47.99^{a}\pm0.14$ mg/100 g) followed by calcium ($29.52^{b}\pm0.02$) then zinc ($24.4^{c}\pm0.10$) while copper was the least in concentration ($0.014^{h}\pm0.002$ mg/100g). This study has shown that Phyllanthus reticulatus ethanol leaf-extract is a good source of essential nutrients that can serve as food supplement.

Keywords: Phyllanthus reticulatus, nutrients, mineral, vitamins, proximate.

Date of Submission: 06-08-2024

Date of Acceptance: 16-08-2024

I. Introduction

Plants are the major sources of essential biomolecules needed in human diet. Such molecules constitute the nutritional requirements of the body, which include carbohydrates, proteins and lipids. These plants do not only contribute to the major energy requirement but also serve as supplementary sources of other essential nutrients such as vitamins and minerals. These nutrients are indispensable requirement of the body which help to maintain the proper physiological homoeostasis of the body [27]. Currently, many researches are directed on plants as source of food [26] and medicine, for the treatment and management of various ailments like diabetes, jaundice, wounds, cancer and others [18]. World Health Organization reported in 2003 that about 80 % of the developing countries depend on herbs for the treatment their diseases [11]. Even in the developed world, plants are also used for the treatment of some health issues due to their phytochemicals, which are less in side effects and affordability [23].

Phyllanthus reticulatus is a climbing shrub with branches and attains a height of 0.5 to 4.5 metres [1]. The plant is commonly known as black-honey shrub, is a pubescent shrub with smooth or lenticellate branches; leaves elliptic to oblong and the fruit is a purplish black berry [6]. The plant is common in Africa and India and other tropics in the world, usually in hedges or waste places. The plant is astringent, sweet, cooling, diuretic and constipating [6]. *Phyllanthus reticulatus* is used as a remedy to treat various ailments such as diarrhea, syphilis, bleeding gums, smallpox and asthma [33]. The leaves are used to treat diabetes and also as cooling drug [33]. Some herbs in the *Phyllanthus* genus are used by many tribes in Africa to treat malaria and wound tetanus infection [31].

Despite the important literature on research pertaining the medicinal and therapeutic applications of *Phyllanthus reticulatus*, this study finds it necessary to research more on the nutritional composition of the plant to explore more on its potentials to act as food supplements and other nutritional applications for healthier living.

II. Materials And Methods

Collection of plant material and ethanol extract preparation

The leaves of *P. reticulatus* were collected from Abakaliki Local Government Area of Ebonyi State and was identified and authenticated by a taxonomist in the Department of Applied Biology, Ebonyi State University, Nigeria. The plant leaves were washed with clean running water and allowed to air-dry for 21 days under room temperature. The sample was then crushed into fine texture using mechanical grinder and sieved to remove the chaff. The ground leaves were extracted using 98% absolute ethanol for 3 days. The ethanol extract was concentrated to dryness using a rotary evaporator.

Determination of Proximate Compositions

The protein, crude fat, crude fibre and ash contents of the extact were determined using the methods of Association of Official Analytical Chemists [4]. Moisture content was determined using the official method of analysis of the Association of Official Analytical Chemists [3]. Percentage nitrogen free extract (NFE) or carbohydrate was calculated according to Pearson (1976). Thus, the carbohydrate content of the sample was determined by taking the sum of ash, protein, moisture, crude fibre, fat and oil from 100.

Determination of Vitamin Contents

Vitamin A (carotene) content of the extract was determined using spectrophotometric method of Jakkutowicz *et al.* [13]. Vitamin B₃ (niacin) and vitamin C contents were determined by Association of Official Analytical Chemists [5] method while vitamin B₁ (thiamin), B₂ (riboflavin) and B₆ (pyridoxine) contents were determined as described by Association of Official Analytical Chemists [3]. The vitamin E (tocopherol) content of the extract was determined using the method of Rutkowski *et al.* [24] and vitamin B₉ (folate) was determine according to the method of Association of Vitamin Chemists (1966).

Determination of Mineral Content of the Extract

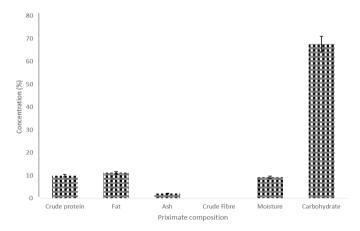
The levels of manganese and selenium in the extracts were determined using Atomic Absorption Spectrophotometer (AAS) method described by Maida *et al.* [17]. Potassium and sodium contents were determined by running the extract solution filtrate in a flame photometer with the standard of potassium and sodium respectively according to the method of Association of Official Analytical Chemists [5].

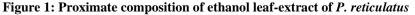
Magnesium and calcium components were determined by EDTA titration method of Association of Official Analytical Chemists [5]. While manganese and zinc were determined by applying EDTA complexometric titration methods of Association of Official Analytical Chemists [5]. Copper content was determined by ferrocyanide method of Association of Official Analytical Chemists [3] and iron was determined using the Orthophenothrolin method described by Association of Official Analytical Chemists [5]. Lastly, the concentration of phosphorus in the extract was determined by the ascorbic method of Association of Official Analytical Chemists [5].

III. Results

Proximate composition of ethanol leaf extract of P. reticulatus

The proximate composition results of ethanol leaf extract of *P. reticulatus* showed that the extract has high carbohydrate content (67.63 ± 0.20 %) but moderate fat (11.18 ± 0.33 %), crude protein (9.92 ± 0.24 %) and moisture content (9.18 ± 0.15 %). The result also revealed that the extract has low ash content (2.09 ± 0.03 %) but has no crude fibre as presented in Figure 1.





Vitamin contents of ethanol leaf-extract of P. reticulatus

The result of the vitamin content of the ethanol leaf-extract of *P. reticulatus* showed that vitamins A, B₁, B₂, B₃, B₆, B₉, C and E were present in the extract. The result showed that among the vitamins identified, the most abundant was vitamin A ($394.58\pm0.04 \text{ mg}/100\text{g}$) followed by vitamin E ($84.05\pm0.03\text{ mg}/100\text{g}$), vitamin C ($83.87\pm0.05 \text{ mg}/100\text{g}$), vitamin B₉ ($7.77\pm0.04 \text{ mg}/100\text{g}$), vitamin B₆ ($7.11\pm0.04 \text{ mg}/100\text{g}$), vitamin B₂ ($3.27\pm0.06 \text{ mg}/100\text{g}$), vitamin B₃ ($2.92\pm0.04 \text{ mg}/100\text{g}$) while vitamin B₁ recorded the lowest in concentration ($2.83\pm0.00 \text{ mg}/100\text{g}$) as shown in Figure 2.

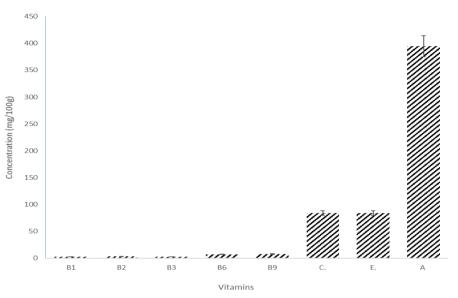


Figure 2: Vitamin content of ethanol leaf-extract of P. reticulatus

Mineral composition of ethanol leaf-extract of P. reticulatus

The results of the mineral content of *Phyllantus reticulatus* showed that the most abundant mineral in the sample was magnesium (47.99 \pm 0.14 mg/100g) followed by calcium (29.52 \pm 0.02 mg/100g), zinc (24.4 \pm 0.10 mg/100g), manganese (20.46 \pm 0.07 mg/100g) and selenium (10.21 \pm 0.06 mg/100g). Others include sodium (5.3 \pm 0.05 mg/100g), potassium (4.44 \pm 0.05 mg/100g) while iron, phosphorus and copper were present at trace levels of 0.072 \pm 0.001 mg/100g, 0.045 \pm 0.001 mg/100g and 0.014 \pm 0.002 mg/100g respectively as presented in Figure 3.

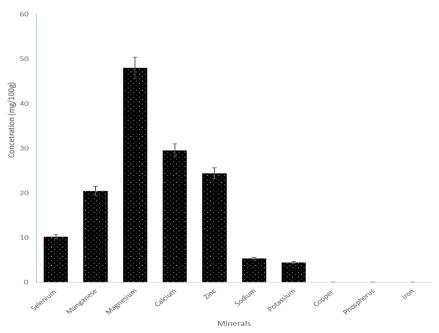


Figure 3: Mineral content of ethanol leaf-extract of *Phyllantus reticulatus*

IV. Discussion

This study has revealed the constituents of both macro and micronutrients available in the ethanol leafextract of *Phyllanthus reticulatus*. The high percentage composition of carbohydrate in the extract was an indication that *P. reticulatus* leaf extract could be a good source of energy and can be useful as food supplement for animals and human diets, since carbohydrates are the major source of energy for the body [30]. A similar finding was observed by Kumar *et al.* [15] who also reported high carbohydrate content in the extract of *P. reticulatus*. The second most abundant nutrients of the plant extract was crude fat. Fat is one of the three main macronutrients, along with carbohydrate and protein [21]. Fat is an important foodstuff for many forms of life, and serve both structural and metabolic functions. Hence, this extract can supply human body with fraction of the fat needed for normal body function and healthy living. Each gram of fat when burned or metabolized releases about 37 kJ = 8.8 kcal [21]. This result revealed that *Phyllanthus reticulatus* ethanol leaf extract has moderate content of crude protein and could serve as a good source of protein. The crude proteins are good source of amino acids utilized by the cells of the body to synthesize all the numerous proteins required for the function of the cells and also to furnish energy [2].

The study recorded less ash content of the ethanol leaf-extract of *P. reticulatus* when compared to earlier report on *Phyllanthus amarus* which is a closely related species [9]. The difference in the ash composition might be attributed to the analyzed state of the analytes involved. The researcher of *Phyllanthus amarus* used the whole leaves while this study analyzed the leaf extract. The similar reason could have also affected the results of the crude fibre content as reported by Ekaete *et al.* [9].

Meanwhile, the result of the vitamin content of the extract showed highest concentration of vitamin A followed by vitamin E and C at a closely comparative manner. Since, vitamins A, C and E are well known for their scavenging properties against reactive oxygen species (ROS) [8], it is therefore inferred that the ethanol leaf-extract of *P. reticulatus* might be a potential source of antioxidants in the body. Other vitamins found in the extract include vitamins B₉, B₆, B₃, B₂ with the lowest amount of vitamin B₁. Furthermore, it has been established that vitamin A, a group of unsaturated nutritional organic compounds that includes retinol, retinal, retinoic acid and several provitamin A carotenoids, has multiple functions as it is important for growth and development, for the maintenance of the immune system and good vision [10, 28]. Hence, the plant could as well serve as food supplement needed for good vision.

The presence of vitamins B_3 , B_6 and B_9 (niacin, pyridoxal phosphate and folic acid respectively) in the extract has made the plant more important for consumption. Niacin is mostly required in the synthesis of NAD (nicotinamide adenine dinucleotide) and NADP (nicotinamide adenine dinucleotide phosphate) which are important biological coenzymes, and also participated in DNA repair [14]. NAD converts to NADP by phosphorylation in the presence of the enzyme NAD⁺ kinase. Nicotinamide adenine dinucleotide phosphate (NADP) and nicotinamide adenine dinucleotide NAD are coenzymes for many dehydrogenases, participating in many hydrogen transfer processes [29]. The presence of vitamin C in the extract has opened for more applications of the plant extract. Scurvy is a disease due to lack of vitamin C, can be prevented and treated with foods or dietary supplements which contains vitamin C [12]. Vitamin C is also an essential nutrient involved in the repair of tissue and the enzymatic production of certain neurotransmitters [32]. It is also acts as coenzyme for several enzymes and required for the maintenance of immune system [16].

The results of mineral analysis in this study showed that the extract had high quantities of magnesium, calcium, zinc and manganese. This was in agreement with report of Ekaete et al. [9] who observed the presence of iron, copper, zinc, magnesium, calcium, manganese, potassium, sodium, phosphorus and chromium in Phyllanthus amarus. Magnesium is an essential mineral present in all human tissues, especially in bone. It is needed for several body functions such as activation of many enzymes of DNA replication and RNA transcription; parathyroid hormone secretion and for muscle and nerve functions [25]. Calcium (Ca), on the other hand, is the most abundant mineral in the body and is essential for a number of vital functions. The body needs adequate dietary calcium (alongside vitamin D and several other nutrients such as vitamin K) to develop and maintain healthy bones and teeth. Calcium also plays a vital role in many systems including intracellular signaling to enable the integration and regulation of metabolic processes, the transmission of information via the nervous system, the control of muscle contraction (including the heart) and blood clotting [25]. The content of Zinc and manganese shows more nutritional relevance of the extract. The major function of zinc in human metabolism is as a cofactor for numerous enzymes. In addition, zinc is required for the functioning of the immune system and in the structure and function of the skin, and hence plays a vital role in wound healing [19]. While manganese is required for bone formation and for energy metabolism [7]. It is also a constituent of an antioxidant enzyme, which helps prevent free radical-mediated damage to cells [7].

Also, the result showed that selenium, sodium and potassium were available at moderate level where as iron, copper and phosphorus were present in trace quantities. The main function of selenium is as a component of some of the important antioxidant enzymes (e.g. glutathione peroxidase), and therefore to protect the body against oxidative damage [22]. Sodium, on the other hand, is responsible for regulating body water

content and electrolyte balance. The control of blood sodium levels depends on a balance between sodium excretion and absorption at the kidneys, which is regulated by nerves and hormones. Sodium is also required for the absorption of certain nutrients and water from the gut [25]. Iron is essential for the formation of haemoglobin in red blood cells; haemoglobin binds oxygen and transports it around the body. Iron is also an essential component in many enzyme reactions and has an important role in the immune system. In addition, it is required for normal energy metabolism and for the metabolism of drugs and foreign substances that need to be removed from the body [20].

V. Conclusion

This study has shown that ethanol leaf-extract of *Phyllanthus reticulatus* is a potential source of vital nutrients such as carbohyrates and proteins. It could also be a good source of important vitamins and minerals which might be of high nutritional and health benefits. Therefore, this extract could be included in dietary supplements to augment for some vital nutritional requirements in the body.

Acknowledgements

The authors gratefully acknowledge the authorities of Ebonyi State University, Abakaliki, Nigeria for providing the laboratory facilities. This research did not receive any grant from funding agencies in the public and commercial sectors.

References

- Abhijit, S., Siddharth, B. And Pushpendra, A. Phyllanthus Reticulatus- An Important Medicine Plant And Its Pharmacological Properties. International Journal Of Pharmaceutical Research And Applications, 8(2): 648-653, 2023.
- [2] Adamu, A. U., Paul, E. D., Gimba, C. E. And Ndukwe, I. G. Phytochemical And Proximate Analysis Of Aspillia Kotschyi (Sch.Bipex, Hochst) Oliv. Nigerian Journal Of Technology, 36(4): 1135 – 1137, 2017
- [3] Aoac (Association Of Official Analytical Chemists). Official Methods Of Analysis Of Aoac, International Association Of Official Analytical Chemists. Washington, Dc, Usa, 2005.
- [4] Aoac (Association Of Official Analytical Chemists). Official Methods Of Analysis. 18th Edn. Association Of Official Analytical Chemists Inc., Washighton, Usa. Pp. 1153-169, 2006.
- [5] Aoac (Association Of Official Analytical Chemists). Official Method Of Analysis Of The Aoac (Ed.): Horwitz, W. 20th Edn. Association Of Official Analytical Chemists Inc., Washighton D.C., Usa. Pp. 1321-1337, 2010.
- [6] Aswatha, R. H.N., Shreedhara, C.S., Gajera, F. P. And Zanwar, S. B. Phamacognostical Evaluation Of Phyllanthus Reticulatus Poir. Pharmacognosy Magazine, 4(18):176-182, 2009.
- Blevins, D. G., Lukaszewski, K.M. Functions Of Boron In Plant Nutrition. Annual Review Of Plant Physiology And Plant Molecular Biology. 49(1): 481–500, 2008.
- [8] Duerbeck, N. B., Dowling, D. D., Duerbeck, J. M. Vitamin C: Promises Not Kept. Obstetrical And Gynecological Survey, 71(3):187-93, 2016.
- [9] Ekaete, D.U., Ukana, D.A. And Itoro, E.U. Phytochemical Screening And Nutrient Analysis Of Phyllanthus Amarus. Asian Journal Of Plant Science And Research, 3(4):116-122, 2013.
- [10] Fennema, O. Fennema's Food Chemistry. Crc Press, New York, Usa. Pp. 454–455, 2008.
- [11] Goyal, B.R., Goyal, R.K. And Mehta, A. A. Phyto-Pharmacognosy Of Archyranthesaspera: A Review. Pharmacognosy Research, 1: 1-12, 2008.
- [12] Hemilä, H. And Chalker, E. Vitamin C For Preventing And Treating The Common Cold. The Cochrane Database Of Systematic Reviews, 1: 98-110, 2013.
- [13] Jakkutowicz, K., Tomick, Z. I. And Leokadia, L. Determination Of Total Plasma Tocopherol In The Presence Of Carotenes. Polskie Archiwum Waterynryjne, 20: 45-57, 1977.
- [14] Kennedy, D. O. B Vitamins And The Brain: Mechanisms, Dose And Efficacy-A Review. Nutrients, 8 (2): 68, 2016.
- [15] Kumar, S., Kumar, D., Deshmukh, R.R., Lokhande, P.D., More, S.N. And Rangari, V.D. Antidiabetic Potential Of Phyllanthus Reticulatus In Alloxan-Induced Diabetic Mice. Fitoterapia, 79(1): 21-23, 2008.
- [16] Maggini, S., Wenzlaff, S. And Hornig, D. Essential Role Of Vitamin C And Zinc In Child Immunity And Health. Journal Of International Medical Research, 38: 386 – 414, 2010.
- [17] Maida, A., Farooq, A., Raziya, N., Umer, R., Kazi, T.G. And Hadeem, M. Mineral Composition Of Moringa Oleifera Leaves And Pods From Different Regions Of Pujab, Pakistan. Asian Journal Of Plant Sciences, 4 (4): 417-421, 2005.
- [18] Mir, M.Y. Documentation And Ethnobotanical Survey Of Wild Edible Plants Used By The Tribals Of Kupwara, International Journal Of Herbal Medicine, 2 (4): 11-18, 2014.

[19] Mitsuo, K., Toshiro, S. And Masayoshi, Y. Aspects Regarding Fluoride Treatment For Reinforcement And Remineralization Of Apatite Crystals. Journal Of Hard Tissue Biology, 21(3): 475–476, 2012.

- [20] Momcilović, B. A Case Report Of Acute Human Molybdenum Toxicity From A Dietary Molybdenum Supplement-A New Member Of The Lucor Metallicumfamily. Archives Of Industrial Hygiene And Toxicology, 50(3): 289–297, 2009.
- [21] Mozaffarian, D., Katan, M.B., Ascherio, A., Stampfer, M.J. And Willett, W.C. Trans Fatty Acids And Cardiovascular Disease. New England Journal Of Medicine, 354(15): 1601–1613, 2006.
- [22] Nielsen, F.H. Minerals In Human And Animal Nutrition. Plant And Soil, 193(2): 199–208, 2007.
- [23] Padma, T.V. India Ayurveda. Nature, 25: 436-486, 2005.
- [24] Rutkowski, M., Grzegorczyk, K. And Paradowski, M.T. Kolorymetryczna Metoda Oznaczania Całkowitej Witaminy E W Osoczu Krwi – Modyfikacja Własna Metody Tsena (Colorimetric Method Of Blood Plasma Total Vitamin E Determination – The Own Modification Of Tsen Method). Diagnostic Laboratory, 41:375-379, 2005.
- [25] Schlenker, E. And Gilbert, J.A. Williams' Essentials Of Nutrition And Diet Therapy. Elsevier Health Sciences, 16: 162–167, 2014.
- [26] Seal, T., Chaudhuri, K., Pillai, B. Nutraceutical And Antioxidant Properties Of Cucumis Hardwickii Royle: A Potent Wild Edible Fruit Collected From Uttarakhand. Journal Of Pharmacognosy And Phytochemistry, 6 (6): 1837-1847, 2017.