International Journal of Chemical Science
Online ISSN: 2523-2843, Print ISSN: 2523-6075
www.chemicaljournals.com
Volume 1; Issue 1; September 2017; Page No. 33-36

Pharmacognostic study of the leaf of *Plantago rugelii* Decne (Plantaginaceae)

***[1] Ogbiko Cyril, [2] Dabai Musa Usman, [3] Abubakar Sani Yelwa, [4] Hassan Usman, [5] Bisiriyu Muhammad Taoheed**

[1,] Department of Pure and Applied Chemistry, Usmanu Danfodiyo University Sokoto, Sokoto State, Nigeria

[2, 3] Department of Pure and Applied Chemistry, Usmanu Danfodiyo University Sokoto, Sokoto State Nigeria

[4] Department of Chemistry, College of Preliminary Studies, Yelwa-Yauri, Kebbi State Nigeria

[5] Department of Chemistry, Federal University of Technology Minna, Niger State Nigeria

Abstract
Medicinal plants are rich source of medicines for the treatment of various diseases. *Plantago rugelii* has been used in treatment of microbial infections, open ulcer wound ulcertation, pains amongs others. However, it has not yet been studied pharmacognostically. The study investigates the macroscopy and microscopy, phytochemical and ash analysis of the leaf of *P. rugelii* using standard procedures. The phytochemical screening revealed the presence of alkaloids, saponins, carbohydrate, reducing sugars, deoxy sugars, phytosterols, protein and flavonoids. The detailed pharmacognostic account reported will be helpful for the correct botanical identification of the drug. In addition, the values of percentage extractive and ash analysis will be helpful for the standardization and quality control of herbal drugs. The study scientifically validates the use of plant in traditional medicine.

Keywords: *Plantago rugelii*, pharmacognosy, plantaginaceae, physico-chemical

1. Introduction
Species of the genus *Plantago* (commonly known as plantain) have been used extensively for medicinal purposes [1]. *Plantago rugelii* (Plantaginaceae) is found all over the world including Asia, Australia, New Zealand, Africa and Europe. The plant is 3 – 12 inches tall and often found in disturbed areas, along roadsides, near buildings, in sidewalks, and other urban areas. It is not a good competitor with taller plans possessing more highly developed root systems, so it is often one of the first plants to grow in a disturbed area. It blooms during summer and the flowers split open, releasing 2-9 seeds. Pollination is done by wind. The seeds become sticky when they get wet, and stick to things like leaves and small animals. When the leaves are blown around by the wind, the seeds spread. During droughts, seeds can remain viable for years until conditions are more favorable. Squirrels and some birds eat the seeds. Caterpillars of *Junonia coenia* and several moth species eat the leaves [2]. The leaves of *P. rugelii* have been utilized as topical for wounds, bites, stings, bronchial infection, hepatitis, and jaundice among others.

Several researches have shown that herbal medicines have promising choice over modern synthetic drugs since they have been shown to have bearable side effects, cost effective and easy accessibility. Herbal formulations involve generally the use of fresh or dried plant parts and the knowledge of such crude drugs is important in its preparation, safety, and efficacy. Pharmacognosy hence, offers a simple, comprehensive and reliable tool by which the complete information of the crude drug can be studied [3]. Medicinal plants being natural offer preventive and curative therapies from diseases which could be useful in achieving the goal of "Health for all" in a cost effective manner [4]. They form a large group of economically important plants that provide the basic raw materials for indigenous pharmaceuticals. [5, 6]. While the World Health Organization (WHO) estimated that 80% of the population in developing countries rely mostly on traditional medicine like plant drugs, for their primary health care needs, modern pharmacopoeia also contains about 25% drugs derived from plants with many others being synthetic analogues of prototypic plant compounds [7]. Due to increase in demand for plant based crude drugs supplied to pharmaceutical, phytochemical and perfumery industries in both developing and developed countries, there is a tendency for adulteration by foreign organic matters resembling the standard drugs or substituted by inferior quality of crude drugs. Hence, the study was aimed at establishing the pharmacognostic characters of the leaf of *P. rugelii* towards monograph development.

2. Materials and methods
2.1 Collection of plant leaves and authentication
Fresh *P. rugelii* plant leaves collected in June 2012 when the plant will start flowering for the correct botanical identification from a forest in Owerre Olubor in Ika North East Local Government of Delta State Nigeria. Efforts were made to collect the plant when they start flowering and fruiting for the correct botanical identification. The plant material was identified and authenticated by Dr E.O. Aigbokhan, Department of Plant Biotechnology, University of Benin, Benin City Edo State Nigeria. The herbarium specimen has been deposited at Forest Research Institute of Nigeria (FRIN) Ibadan Nigeria where a specimen number FHI 109775 was issued The leaves were washed under running tap water followed by Bavistin (Fungicide) (15% W/V) solution for 5

min. They were finally rinsed with sterile distilled water. The fresh leaves were used to study the macroscopic and microscopic parameters; whereas shade dried powdered leaf material was used for the physico-chemical, pharmacognostic and preliminary phytochemical investigations.

2.2 Macroscopic and Organoleptic studies
The macroscopic study was conducted so as to aid in its identification as well as in its standardization. The fresh leaves were subjected to macroscopic studies which comprised of organoleptic characters *viz*., color, odour, appearance, taste, texture among others were documented by direct observation methods (naked eye observation) of the dried samples.

2.3 Microscopy
The outer epidermal membranous layer (in fragments) were cleared in chloral hydrate, mounted with glycerin and observed under a compound microscope. The presence/absence of the following was observed: epidermal cells, stomata (type and distribution) and epidermal hairs (types of trichomes and distribution). The transverse sections of the fresh leaves through the lamina and the midrib as well as a small quantity of the powdered leaves were also cleared, mounted and observed [8].

2.4 Chemomicrocsopic study
Examination of the powder for starch grains, lignin, mucilage, calcium oxalate crystals, cutin and suberin were carried out using standard techniques [9].

2.5 Quantitative investigation
Quantitative leaf microscopy to determine palisade ratio, stomata number, stomata index, vein – islet number and veinlet termination number were carried out on epidermal strips according to standard procures [10].

2.6 Phytochemical and Physicochemical parameters
The phytochemical screening for important secondary metabolites and various physicochemical parameters which include the determination of ash contents (total ash, water soluble and acid insoluble), extractive values, moisture content was performed according to standard procedures [11, 12, 13].

3. Results & Discussion
3.1 Macroscopic and Organoleptic Studies
Macroscopic study helps in rapid identification of plant material and is the primary step in characterization of crude drug. *P. rugelii* leaves are simple, entire and are oppositely arranged. The leaves are oval shaped which taper to points with three veins. They are reddish at the base and all grow out of the center. Leaves are about 6" long and have long petioles. It lacks leaf blades. The organoleptic studies revealed that the leaves are dark green in color on dorsal surface, light green in color on ventral surface with characteristic pungent odour. The powdered leaf material is light green in color, coarse in texture, slightly pungent in odour with bitter taste. This macroscopic study is important because it helps in the rapid identification of plant material and it is often the primary step in characterization of crude drug [4].

3.2 Microscopic and Chemomicrocsopic Studies
The result of the microscopic investigation of the leaves of *P. rugelii* is presented in Figures 1 (a – c).

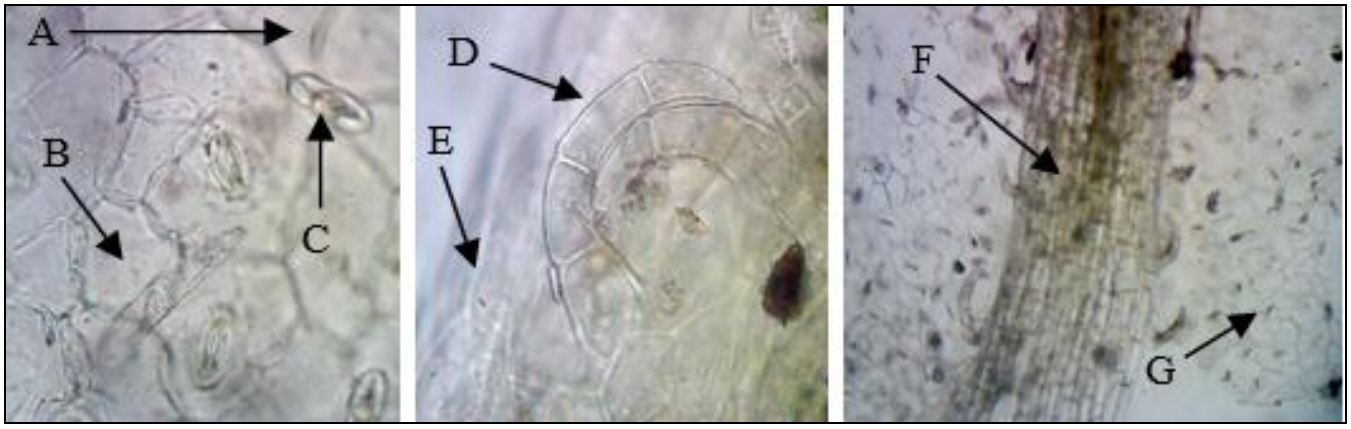

Fig 1a-c: Microscopic photographs of the leaves of *P. rugelii*. A: Epidermis; B = Stoma; C = Stomata; D = Trichome; E = Cuticle; F = Vascular bundle; G = Parenchyma cells.

The microscopic study of a plant material (leaf, stem, bark or root) serves as an important diagnostic tool in differentiating and identifying a particular plant species. The transverse section of the leaf of the plant showed a dorsiventral nature. The upper and lower surface is covered by single layer of epidermis. The upper epidermis showed the presence of well-developed cuticle. The stomata appeared on the upper epidermis with trichomes. The stomata are fairly distributed. While the mesophyll showed the presence of distinct palisade and spongy parenchyma, the midrib showed the presence of 4-7 vascular bundles. Each vascular bundle is surrounded by sclereides on all sides. The chemomicroscopic studies of the leaves revealed the presence of lignin, starch, mucilage and calcium oxalate crystals. The establishment of the microscopic features of the plant is a good aid in identifying the plant and guiding against adulteration [14].

3.3 Quantitative Investigation
The numerical values for the quantitative investigation of the leaves of P. rugelii is presented in Tables 1

Table 1: Numerical data of *P. rugelii*

Parameters	Mean ± SD
Palisade ratio	7.25 ± 0.41
Stomata number: Upper surface	8.30 ± 0.19
Stomata number : Lower surface	8.80 ± 0.15
Stomata index: Upper surface	25.13 ± 0.14
Stomata index: Lower surface	22.47 ± 0.20
Vein islet number	1.15 ± 0.08
Veinlet termination number	1.45 0.03

3.4 Phytochemical and physicochemical parameters

The phytochemical screening revealed the presence of alkaloids, saponins, carbohydrate, reducing sugars, deoxy sugars, phytosterols, protein and flavonoids. Tannin, anthraquinones and phenolic compounds were revealed to be absent. Determination of phytochemical profile of medicinal plants is an indication of the class of compound present in the plant. These phytochemicals are responsible for the various pharmacological activities expressed by medicinal plants. The physico-chemical parameters with reference to herbal Pharmacopeia (Ayurveda and African) [15, 8] is represented in Tables 2 to 7.

Table 2: Moisture content of *P. rugelii*

Reference	Observed value ± SD (% w/w)	Reference value
British Pharmacopeia	12.46 ± 0.08%	Not more than 14%

Table 3: Total ash value of *P. rugelii*

Reference	Observed value ± SD (% w/w)	Reference value
Ayurveda Pharmacopeia	17.22 ± 0.20%	Not more than 17%

Table 4: Acid Insoluble ash value of *P. rugelii*

Reference	Observed value ± SD (% w/w)	Reference value
Ayurveda Pharmacopeia	4.93 ± 0.36%	Not more than 5%

Table 5: Water soluble extractive ash value of *P. rugelii*

Reference	Observed value ± SD (% w/w)	Reference value
Ayurveda Pharmacopeia	6.27 ± 0.18%	Not less than 12%

Table 6: Alcohol extractive index of *P. rugelii*

Reference	Observed value ± SD (% w/w)	Reference value
Ayurveda Pharmacopeia	0.98 ± 0.04%	Not less than 2%

Table 7: Water extractive index of *P. rugelii*

Reference	Observed value ± SD (% w/w)	Reference value
Ayurveda Pharmacopeia	0.25 ± 0.06%	Not less than 2%

Key: SD = Standard Deviation; w/w = weight by weight; % = percentage

Moisture content in plants assists in maintaining the protoplasmic contents of cells but make herbs perishable and susceptible to microbial degradation during storage [16]. The moisture content is lower than the 14% value set by British Pharmacopoeia [10], hence suggests that *P. rugelli* leaves will not be prone to microbial attack and will have a fairly stable shelf life. Ash may be derived from the plant tissue itself (i.e. physiological ash) or from the extraneous matter, especially sand and soil adhering to the surface of the drugs (i.e. non-physiological ash), and this kinds of ash are determined together, therefore it is referred to as total ash [8]. Although, the total ash value is higher than that set by the Ayurveda Pharmacopeia, it is however noted that plants with ash content above 8.8% are useful health wise with a high deposit of mineral elements, which may be nourishing and suitable for consumption [17]. The level of contamination or adulteration by sand (silicious earth) can be detected by the level of acid insoluble ash. Acid insoluble ash of 4.93 % was obtained and with reference to the Ayurveda Pharmacopeia standard [15], the crude powdered drug is free from silicate adulteration. It also reveals that a small portion of the ash content is acid soluble as well as its digestibility when eaten or consumed [18, 19]. The alcohol extractive value was observed to be higher than the water extractive value indicating that water could be a better solvent for the extraction of the plant.

4. Conclusions

The findings of this research revealed the presence of various important bioactive compounds which could be attributed to the diverse pharmacological applications of the plant and this proves that the plant leaves are medicinally important. The physico-chemical results ware within limits of relevant

pharmacopoeia and WHO guidelines. These results will be required in the preparation of a monograph on the plant and may help in standardization, identification and in carrying out further research with *P. rugelii* leaf based drugs which are used in Nigeria and modern pharmacopoeia.

5. Acknowledgment
The authors like to thank the staff of the Department of Pharmacognosy, Faculty of Pharmacy, University of Benin, Benin City Edo State Nigeria for their assistance in carrying out this research.

6. References

1. Blumenthal M, Busse WR, Goldberg A, Gruenwald J, Hall T, Riggins CW, *et al.* Complete German Commission E Monographs−Therapeutic guide to herbal medicines In: Klein S, Rister, RS Eds. American Botanical Council, Boston, Integrative Medicine Communications, 2000, 43-304.
2. David BL. Medicine at your feet: healing plant of the Hawaiian kingdom *Plantago*. Journal of Ethno pharmacology. 2006; 76(1):59-64.
3. Gokhale SB. Textbook of Pharmacognosy, Nirali Prakashan, 1979, 55-58.
4. Srikanth K, Vikram G, Archana P, Rajinikanth M, Rama SN. Pharmacognostic and Phytochemical Investigations in *Strychnos potatorum* Linn. F. Journal of Pharmacognosy and Phytochemistry. 2013; 2(4):46-51.
5. Aiyelaagbe O. Antibacterial activity of *Jatropa multifida* roots. Journal of Fitness. 2001; 72(5):544-546.
6. Augusti KT. Therapeutic values of onion and garlic. Indian Journal of Experimental Biology. 1996; 34(3):634-640.
7. Nathiya S, Santhi N, Kalaiselvi S. A comparative study on ontogenic expression of antioxidants and secondary metabolites in *Withania somnifera.* International Research Journal of Pharmacy. 2012; 3(1):210-215.
8. African Pharmacopoeia. General methods for Analysis. OAU / STRC Scientific Publications, Lagos. 1986; 2(2):01-5, 137-149, 223-237.
9. Evans WC. Trease and Evans Pharmacognosy. WB Saunders Ltd, 14th Edition, London, 1996, 119-159.
10. British Pharmacopoeia. Ash value, Acid insoluble, Water soluble extractive and Alcohol extractive. Appendix XII Majesty Stationary office London. 1980; 2:1276-1279.
11. Stalh E. Drug analysis by chromatography and microscopy. A Practical Supplement to Pharmacopoeias. 1st ed. Ann Arbor, Michigan, USA, 1973, 219-224.
12. Sofowora A. Screening plants for bioactive agents. In medicinal plants and traditional medicine in Africa. Spectrum Books Ltd, Ibadan, Nigeria, 1982, 289.
13. Trease EA, Evans WC. Pharmacognosy. 11th ed. Churchill Livingstone Harcourt Health Service, London, 1996, 60-75.
14. Dinesh K, Karunesh K, Sunil K, Tarun K, Ajay K, Om P. *et al.* Pharmacognostic evaluation of leaf and root bark of *Holoptelea integrifolia* Roxb. Asian Pacific Journal of Tropical Biomedecine. 2012; 11(3):169-175.
15. Indian Pharmacopeia. Controller of Publication, 1996; A-1:16, 17.
16. George PM. Encyclopedia of Food. Humane Press, Washington, USA, 2008; 1:526.
17. Antia BS, Akpan EJ, Okon PA, Umoren IU. Nutritive and Anti-Nutritive Evaluation of Sweet Potatoes *Ipomoea batatas* Leaves. Pakistan Journal of Nutrition. 2006; 5:166-168.
18. Okhale SE, Odiniya EO, Kunle OF. Preliminary Phytochemical and Pharmacognostical Investigation of Pediatrics Antimalarial *Laggera Pterodonta* DC Sch. Bip: Asteraceae of Nigeria Origin. Journal of Ethnobototanical Leaflets. 2010; 14:457-466.
19. Ibrahim J, Ajaegbu VC, Egbarevba HO. Pharmacognostic and Phytochemical Analysis of *Commenlina benghalensis* L., Journal of Ethnobototanical Leaflets. 2010; 14:610-615.