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Macro- and Micro-Morphological Characteristics and Phytochemical Constituents of *Ficus exasperata* Vahl. of Moraceae

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Abstract **Original Research Article**

The research was set to study the macro- and micro-morphological and phytochemical constituents of *Ficus exasperata* Vahl. of the family Moraceae, though it does not produce a milky sap like most members of the family, rather it produces a sticky viscid sap. It is a pan-tropical tree by habit and deciduous perennial dicot that grows up to 12 to 15m in height. It has simple petiolate leaves which are ovate to obovate elliptic with alternate phyllotaxy and cuneate to rounded at base. The margins of the leaves are slightly wavy with very rough lamina when rubbed, commonly called sand-paper plant. The leaves measure up to 27±5cm in length and 22±4cm in width. The flowers are borne in hollow receptacle and the fruits appeared when the tree is leafless, occurring singly or in pairs all along the younger branches. The foliar epidermal studies revealed paracytic stomata and multicellular uniseriate trichomes which are amphistomatic. It has a foliar adaxial index of 3.5% and 13.6% for the abaxial. The qualitative phytochemical investigations conducted showed presence of following metabolites: flavonoids, tannins, saponins, cardiac glycosides and terpenoids while alkaloids, fixed oil components and anthraquinones were absent. The quantitative chemical estimation studied showcased 82.95mg/kg of Sodium (Na), 7816.25mg/kg of Potassium (K) and 391.75mg/kg of Iron (Fe).The investigations conducted here could be used for further delimitation of the species.

Keywords: Morphology, Qualitative, quantitative, Phytochemistry, *Ficus*.

INTRODUCTION

Ficus exasperata Vahl. is a tree by habit which belongs to a genus *Ficus* which contains about 850 species of woody trees, shrubs, vines, epiphytes and hemi-epiphytes in the mulberry family Moraceae [1, 2]. They are generally regarded as fig trees or figs, and are found as native to the tropical and semi-warm temperate zones [3]. The genus *Ficus* as a pan-tropical trees, shrubs and vines which occupy a wide range of ecological niches, most are ever green while some are deciduous [4]. *Ficus* species are characterized by their unique inflorescence and distinctive pollination which utilizes Wasp species for pollination. Figs are relative easy to recognize though the identification of many of the species in Moraceae are difficult [5]. *Ficus exasperata* is commonly called forest sandpaper fig, white fig, or sandpaper leaf tree. It is a deciduous and dioecious species of trees. The flowers are borne in hollow receptacle and the fruits appear when the tree is leafless, occurring singly or in pairs all along the younger branches [6, 7] have also discussed hydathodes in *Ficus*. Previous classifications of the *Ficus* have been largely based on the systematics of pollinating wasps [8, 9].

Worked on the survey of epidermal morphology of the genus *Ficus* and stated that most are of paracytic stomata and that the stomata are restricted to the abaxial surface of lamina (hypostomatic) except in *F. vallis-choudae*, in which leaves are amphistomatic, but stomata more abundant on abaxial surface and that stomata are dense or evenly distributed on the abaxial foliar surfaces in most species, but they are few in *F. abutilifolia, F. capreifolia, F. ottoniifolia, F. platyphylla, F. sansibarica subsp*. macrosperma Stomata are mostly of a paracytic type [10].

The relevance of this studies point to the fact that *Ficus* properties may offer a new source of potential activities against infective bacterial agents to many diseases. Hence the objectives focus on the macro- and micro-morphological characteristics and qualitative phytochemical constituents and quantitative chemical estimation of *Ficus exasperata* Vahl. this belongs to the family Moraceae.

MATERALS AND METHODS

Geographic Location

The location of the parent plant studied was Port Harcourt, Rivers, Nigeria.

Morphological Studies

The meter rule was used to ascertain the plant height from the root-collar to the terminal bud while leaf length from the leaf tip to the petiole base. The leaf width is measured across the leaf lamina, from one margin to another at the widest region.

Micro-morphological (Epidermal) Studies

Fresh leaves and young stem collected for this study were peeled following the method of [11] and subjected to alcohol solutions in the ratio of 30%, 50%, 70%, 95% and absolute alcohol respectively. The cleared epidermal layers obtained were stained with safranin for 5 minutes washed and counter stained with Alcian blue for same time interval, washed and temporarily mounted in aqueous glycerol solution. Photomicrographs were taken from good preparations. The stomatal index [S.I.] was obtained using the formula:

$$S.\ I. = \frac{S}{S + E} \quad X \quad \frac{100}{1}$$

Where S and E are mean numbers of stomata and epidermal cells respectively within the particular area under investigation. Likewise trichome Index (T.I) was obtained using:

$$T.\ I. = \frac{T}{T + E} \quad X \quad \frac{100}{1}$$

Where T and E are trichomes and epidermal cells respectively within the study area.

Qualitative Phytochemical Study

Leaves of *Ficus exasperata* Vahl. studied were sun dried for 72 hours (3 days) and weighed. Fifty grammes (50g) of the dried leaves were macerated in 96% ethanol using a pestle and a mortar. The extract was thereafter filtered and evaporated to dryness (constant weight) using a rotary evaporator set at 45^0C. Residue yields were noted and a portion was used for the phytochemical screening.

Test for Saponins

Frothing tests was done following the method described by [12]. The ability of saponins to produce frothing in aqueous solution and to haemolyse red blood cells was used as screening test for these compounds. 0.5g of the plant extract was shaken with water in a test tube. Frothing which persisted on warming was taken as preliminary evidence for the presence of saponins. The disc was then washed in ether, dried and placed on a 7% blood nutrient agar. Complete haemolysis of red blood cells around the disc after 6 hours was taken as further evidence of presence of saponins.

Test for alkaloids

This was carried out using 0.5g of the plant extract which was stirred with 5ml of 1% aqueous hydrochloric acid on a steam bath; 1ml of the filtrate was treated with a few drops of Mayer's reagent and a second 1ml portion was treated similarly with Dragendorff's reagent. Turbidity or precipitation with either of these reagents was taken as preliminary evidence for the presence of alkaloids in the extract being evaluated [13, 14]. A modified form of the tin-layer chromatography (TLC) method as described by [15] was used. One gramme (1g) of the extract was treated with 40% calcium hydroxide solution until the extract was distinctly alkaline to Litmus paper, and then extracted twice with 10ml portions of chloroform. The extracts were combined and concentrated to 5mls. The chloroform extract was then spotted on thin-layer plates. Four different solvent systems were used to develop each plant extract. The presence of alkaloids in the developed chromatograms was detected by spraying the chromatograms with freshly prepared Dragendorff's spray reagent. A positive reaction on the chromatograms (indicated by an orange or darker colored spot against a pale yellow background) was confirmatory evidence that the plant extract contained alkaloid.

Test for tannins

Five grammes (5g) of each portion of plant extract was stirred with 10ml of distilled water, filtered, and 5% ferric chloride reagent added to the filtrate. A blue-black, green, or blue-green precipitate was taken as evidence for the presence of tannins [16].

Test for anthraquinones

Borntrager's test was used. Five grammes (5g) of each plant extract was shaken with 10ml benzene, filtered and 5mls of 10% ammonia solution added to the filtrate. The mixture was shaken and the presence of a pink, red, or violet color in the ammonia (lower) phase indicated the presence of free hydroxyanthraquinones.

Test for phlobatannins

The deposition of a red precipitate when an aqueous extract of the plant part was boiled with 1% aqueous hydrochloric acid was taken as evidence for the presence of phlobatannins [14].

Test for Flavonoids

Shinoda Reduction Test

5g of the pulverized sample was boiled in 5ml of distilled water for 5 minutes on water bath and filtered while hot. Magnesium (Mg) was added to the filtrate and few drops of conc.H_2SO_4 were carefully introduced into the mixture. The formation of orange, red, crimson or magenta was taken as evidence of preliminary presence of flavonoid.

Lead Acetate Test

5g of pulverized sample was boiled in 5ml of distilled water for 5 minutes in water bath and filtered while hot. 2ml of 10% lead acetate was added to the

filtrate and observed. Yellow precipitate indicated presence of flavonoids.

Test for cardiac glycosides

Lieberman's test was used in which 0.5g of the extract was dissolved in 2ml of acetic anhydride and cooled in ice. One milliliter (1ml) of Sulphuric acid was carefully added in drops until a color change from violet to blue to green indicated the presence of a steroidal aglycone portion of the cardiac glycoside [16].

Steroids and Terpenoids

Salkowski's Test: 2g of plant sample was pulverized and macerated in 5mls of chloroform and filtered. 2mls of H_2SO_4 was carefully added to the filtrate and observed. A reddish brown colour at the interface indicated presence of steroidal substances.

Libermann-Burchard's Test: 2g of plant sample was pulverized and macerated in 5mls of chloroform and filtered. 1ml of acetic anhydride was added to the filtrate followed by 2mls of conc.H_2SO_4 to form a layer. Color change from violet to blue to green at interface showed the presence of terpenoids.

Fixed Oil

About 5 grammes of plant sample was vigorously but carefully rubbed with filter paper. The paper was allowed to air dry for about 5 to 10 minutes and observed. Observation of translucent coloration revealed the presence of fixed oil.

Quantitative Chemical Estimation

IRON ION

Fresh leaves of *Ficus exasperata* Vahl. were collected, air dried and blended. The instrument used for this test was adjusted and its vital settings recommended were put in place, a wave length of 248nm was selected then the air and gas flow was adjusted slit width. Hallow cathode lamp was stabilized and allowed adequate time to energize. The instrument was calibrated with iron ion concentration to obtain a standard plot. 1g of the sample of *Ficus exasperata* was aspirated into the instrument and the concentration of Iron in the sample was obtained extrapolation from the standard iron graph in ppm or mg/L.

POTASSIUM ION

Fresh leaves of *Ficus exasperata* were collected air dried and blended. 1g of the pulverized sample was digested and the solution was made up to 50mls with distilled water. The instrument employed for the test was programmed to the recommended vital settings, air and gas pressure were adjusted and a wave length of 766nm was selected. Standard Potassium ion concentration was aspirated into the instrument "burner chamber" to calibrate the equipment and to plot the graph of the standard ion. The aspirator tubing's system was occasionally flushed with water before the sample was aspirated. The concentration of potassium ion in the sample was automatically displayed on the screen of equipment and also printed out for documentation.

SODIUM ION

Fresh leaves of *Ficus exasperata* were collected air dried and blended. The equipment employed for the test was adjusted to the recommended vital settings, air and gas pressure were adjusted to slit width and a wave length of 589nm was selected. The instrument was calibrated with the standard sodium ion concentration to obtain a standard sodium ion graph. 1g of the sample was aspirated into the instrument and its concentration obtained by extrapolation from the standard graph in mg/L or ppm.

RESULT

The Geographic location

The geographic location of the parent plants is Choba, Port Harcourt, Rivers State, Nigeria.

Macro-morphological Study

The macro-morphological description is shown in Table-1. *Ficus exasperata* is a pan-tropical tree by habit and deciduous perennial dicot that grows up to 12 to 15m in height. It has simple petiolate leaves which are ovate to obovate elliptic with alternate phyllotaxy and cuneate to rounded at base. The margins of the leaves are slightly wavy with very rough surface or lamina when rub commonly called sand plant usually used for wash off charcoal pots after cooking in some localities and measures up to 27±5cm in length and 22±4cm in width. The flowers are borne in hollow receptacle and the fruits appear when the tree is leafless, occurring singly or in pairs all along the younger branches. The fruit is an achene See Plate-1.

Plate-1: *Ficus exasperata* Vahl. 1a: The Canopy of *Ficus exasperata*. 1b: Alternately arranged leaves in a young branch of *Ficus exasperata*, 1c: Adaxial foliar surface, 1d: Abaxial foliar surface

Table-1: Summary of morphological characteristics of *Ficus exasperata*

Characters	*Ficus exasperata*
Habit	Medium Tree
Duration	Perennial and deciduous
Root	Tap Root system
Stem Description	Woody stem that grows up to 12 to 15m in height
Leaf type	Petiolate simple leaves
Leaf organization	Simple
Phyllotaxy	Alternately arranged
Leaf outline or shape	Ovate
Leaf margin	Slightly wavy
Length of leaf (cm) 27cm	
Range	27 to 32cm
Mean	27±5cm
Breadth of leaf (cm) 22cm	
Range	22 to 26cm
Mean	22±4cm
Flower description	Much reduced located in hollow receptacle
Fruit description	The fruits occurring singly or in pairs along the younger branches.

Micro-Morphological Studies

The foliar epidermal studies revealed paracytic stomata and multicellular uniseriate trichomes which are amphistomatic. The foliar adaxial index of 3.5% and 13.6% for the abaxial. Plates 2a, 2b and 2c.

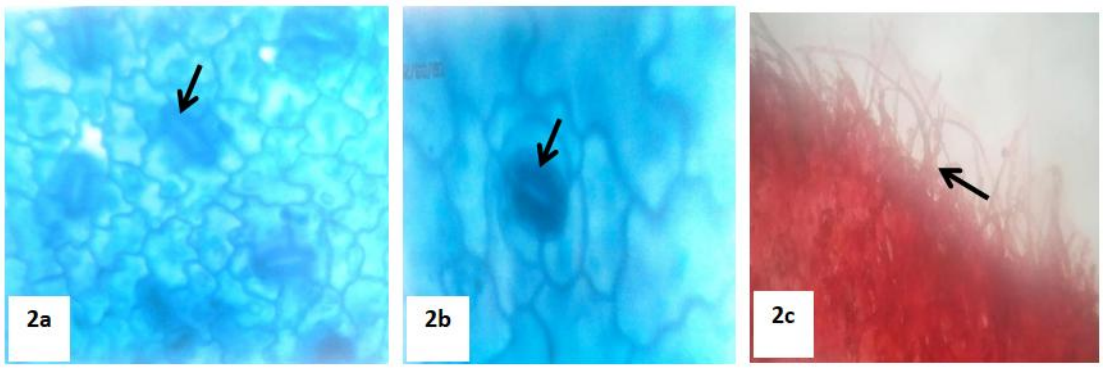

Plate-2: *Ficus exasperata* Foliar Epidermis. 2a: Abaxial surface. 2b: Adaxial surface. 2c: Adaxial region with very numerous Trichomes not branched contributing to the roughness of the surface. Arrow showed paracytic stomata amphistomatic in nature in 2a and b, while in 2c arrow revealed tuft trichomes

Qualitative Phytochemical Studies

The qualitative phytochemical investigations conducted showed presence of following metabolites: flavonoids, tannins, saponins, cardiac glycosides, terpenoids and carbohydrates while alkaloids, fixed oil components and anthraquinones were absent. These are illustrated in Table-2.

Table-2: Qualitative Phytochemical Studies on *Ficus exasperata* Vahl.

Phychemicals tested in *Ficus exasperata*	Result
Saponins	+ve
Alkaloids	-ve
Tannins	+ve
Anthraquinones	-ve
Flavonoids	+ve
Cardiac glycosides	+ve
Steroids	+ve
Terpenoids	+ve
Fixed oil	-ve

Key: '+ve' revealed 'presence' while '–ve' showed 'absence'

Quantitative Chemical Estimations

The quantitative chemical estimation studied showcased 82.95mg/kg of Sodium (Na), 7816.25mg/kg of Potassium (K) and 391.75mg/kg of Iron (Fe) as shown in Table-3.

Table-3: Quantitative Chemical Estimations On *Ficus exasperata* Vahl.

Specimen	Iron (Fe)	Potassium (K)	Sodium (Na)
Ficus exasperata	391.75mg/kg	7816.25mg/kg	82.95mg/kg

DISUSSION

Ficus exasperata Vahl. is a woody tree by habit with smooth trunk or stem, this conforms to the description provided by [1-3]. *Ficus exasperata* is commonly called forest sandpaper fig, white fig, or sandpaper leaf tree. It is a deciduous and dioecious species of trees and fruits are found along younger branches as also support by [6].

The adaxial foliar surface of *Ficus exasperata* has more numerous trichomes than the abaxial foliar surface and hence is more rougher when touched but has very few number of stomata or not all when compared to the abaxial foliar region, which is in line with the work of [10] who worked on the survey of epidermal morphology of the genus *Ficus*. Stomata are of a paracytic type also in concordance to the findings of [10].

CONCLUSION

There is very limited number of research in micro-morphology of *Ficus exasperata*. The proximate analysis, the karyotypes, quantitative aspect of phytochemistry and DNA barcodes may be of future interest.

RECOMMENDATION

The effort of Miss Nwagwu, Cynthia who did the initial field collection of *Ficus exasperata* Vahl. and assisted in some of the laboratory research is immensely commended.

REFERRENCES

1. Hutchinson J, Dalziel JM. Flora of West Africa. Crown Agents for Oversea Government and administrations, 4, Millbank, London, S.W.I. 1953; 600-611.
2. Flaishan VC, Moshe MK. Reconstructing the Phylogeny of figs (Ficus, Moraceae. Journal of Biogeography, 2006;4:521-530.
3. Berges CC, Hijman FY. Ficus: In Flora of Tropical East Africa. 1999;43-86.
4. Haveley BT, Abraham H. Variation in the structure and development of foliar stomata in the Ficus, Moraceae. Bot J Linn Soc. 1999;75:69-97.
5. Quigley MM. Studies of the petiole Anatomy of Moraceae. Indian Natl. Science Academy. 1998;40:56-67.
6. Nyananyo BL. Plants from the Niger Delta. Doval Ventures Limited, 12Ohaeto Street, D/Line, Port Harcourt, Rivers State, Nigeria. 2006;161-162.
7. De-Bary A. Comparative Anatomy of Vegetative Organs of the Phanerogams and Ferns. English Translation by 1884.
8. Corner EJH. An introduction to the distribution of Ficus. Reinwardtia. 1958;4:425-355.
9. Corner EJH. Checklist of Ficus in Asia and Australasia with keys to identification. Gard Bull Singapore. 1965;21**:**1-186.
10. Sonibare MA, Jayeola AA, Egunyomi A, Murata J. A Survey of Epidermal Morphology in Ficus Linn. (Moraceae) of Nigeria. Bot Bull Acad Sin. 2005;46:231-238.
11. Cutler DF. Applied Plant Anatomy. Lib. Of Congr. Cataloguing in Publication Data. William Clowes and Sons Ltd. London. 1978.
12. Wall ME, Eddy CR, McClenna ML, Klump ME. Detection and estimation of steroid Sapogenins in plant tissues. Anal Chem. 1952; 24:1337.
13. Harborne JB. Phytochemical Methods: A Guide to modern Techniques of Plants Analysis. Chapman and Hall London. 1973;279pp.
14. Trease GE, Evans INC. A textbook of Pharmacognosy 3rd ed. Boilliere Tinall. 1989.
15. Farnsworth NR, Euer KL. An Alkaloid screening procedure utilizing thin–layer Chromatography. Lioydia. 1962; 25-186.
16. Shoppe CW. Chemistry of the Steroids, 2nd Ed. Butterworths, London. 1964; 56pp.