Ultra-Structure of the Leaf Surfaces of the Family Bignoniaceae Juss. in Nigeria

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Abstract: The leaf surfaces of the Nigerian Bignoniaceae were studied using the Scanning Electron Microscope (SEM). The species in this study were: *Crescentia cujete* Linn. *Jacaranda mimosifolia* D. Don., *Kigelia africana* (Lam) Benth. *Markhamia tomentosa* (Benth) K. Schum., *Newbuldia laevis* (P. Beauv.) Seemann ex Bureau. *Spathodea campanulata* P. Beauv. *Stereospermum acuminatissimum* K. Schum. *Stereospermum kunthianum* Cham. *Tabebuia rosea* (Berthol) D. C. *Tecoma stans* (Linn) H, B &K. and *Tecoma capensis*. The study was conducted using anatomical characters to identify and delimit the Nigerian taxa at the tribal level. Stomata were observed on the abaxial surfaces of all the species studied. Sunken stomata were found in *K. africana* while the others had raised stomata. Peltate trichomes were found on some species like the abaxial surfaces of *C. cujete, J. mimosifolia, M. tomentosa, N. laevis T. stans* and *T. rosea* and on the adaxial surfaces of *C. cujete, M. tomentosa, N. laevis, S.acuminatissimum* and *T. rosea*. *M. tomentosa* had both glandular and non-glandular trichomes on the abaxial surface while *T. capensis* had only non-glandular trichome on the adaxial surface. The non-glandular trichomes of *M. tomentosa* were long and with blunt tip while those of *T. capensis* were short and with pointed tips. Striae were found on the abaxial surfaces of *J. mimosifolia* and *T. stans* and on both the abaxial and adaxial surfaces of *T. capensis*. The genera *Tabebuia* and *Tecoma* are suggested to be retained in the tribe Tecomeae based on striations on their epidermal cells. While other members be assigned to a new tribe 'Spathodeae'.

Keywords: Abaxial, Adaxial, Anatomical characters, Spathodeae, Tecomeae.

INTRODUCTION

The Bignoniaceae, or Trumpet creeper family, is a family of flowering plants comprising of 7-8 tribes [1], and about 650-750 species in 116-120 genera [2]. Members are mostly trees and shrubs, and more rarely lianas and herbaceous plants. Bignoniaceae are "one of the most important families of woody plants and the most important family of lianas in the Central American forest ecosystem'' [3].

Hutchinson and Dalziel [4] recorded five genera in Nigeria*;* these are *Kigelia* Lam*., Markhamia* Seemann. ex. K. Schum. *Newbouldia* Seemann. ex Bureau*. Spathodea* P. Beauv. and *Stereospermum* Cham. Beside these genera, there are also introduced species such as *Crescentia cujete* Linn., *Tabebuia rosea* (Berthol) D. C, *Tecoma stans* (Linn) H, B &K., *Jacaranda mimosifolia* and *Tecoma capensis* in the country.

The family is cosmopolitan, is present in both the old and the new world. The origin of the family is not clear; however, Goldblatt and Gentry [5] speculated that the old World species may be among the most primitive members of the family. Thirteen species in 8 genera (including naturalized) are present in South Africa and 12 genera and 35 species are present, only in mainland states.

Members of this family commonly have pinnately or multiply compound. Palmately compound leaves are rare. Simple leaves are also rare, and when observed, are often dissected in pinatifid or palmatifid fashion. Leaves are typically in opposite but whorled (in *Kigelia)* or spiral arrangement is also found, leaves petiolate and extipulate (but pseudostipules are present in the genus *Markhamia*). Domatia occur in at least 14 genera.

Flowers are bisexual, hypogynous and zygomorphic, either solitary or aggregated in recemes or cymes (dichasium or helicoids cyme). Inflourescence, when it occurs, might be axillary or terminal. Bracts and bracteoles are present and sometimes deciduous. The sepals are fused, forming a synsepalous calyx either entire or toothed in 5 lobes. The petals are equally fused in a sympetalous campanulate corolla that also has 5 lobes. These lobes are often clearly shorter than the tube. The corolla and

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E-ISSN: 2311-858X/14

calyx are distinct from each other. Sometimes, a 2-lipped (bilabiate) corolla is observed. The androecium is didynamous, i.e. there are 4 stemens occurring in 2 pairs of different lengths. The stemen are inserted epigynously and alternate with the corolla lobes. The anthers are often connivent but separated from one another sometimes. One staminode may be present. The gynoecium or stylate are median. It consists of one compound pistil with 2 carpels, a single style, and a superior ovary with typically 2 locules (more rarely 1 or 4) each bearing numerous axile ovules. An annular or capular nectar disk usually occurs around the ovary base. Ovules are anatropous, with microphyles directly downward. Floral formulae: K(5),C(2+3),A2+2,G (2) Capsule.

The fruits are typically elongated dehiscent capsule that opens loculicidally or septicidally. The fruit can sometimes be an indehiscent berry (*Kigelia* and *Crescentia*).

Seeds are numerous, are usually winged and wind dispersed but occasionally also comose and wingless. The winged ones include those of *Jacaranda mimosifolia, Newbouldia laevia, Markhamia tomentosa, Spathodea campanulata, Stereospermum acuminatissimum, Stereospermum kunthianum Tabebuia rosea* and *Tecoma stans* while the wingless includes those of *Kigelia africana* and *crescentia cujete*. Arils are present in all the seeds of the Bignoniaceae studied and the seeds do not have endosperm.

Pollination is either entomophilous (*via* insects), ornithophilous (*via* birds) or cheiropterphilous (*via* bats).

For more than one hundred years the anatomy of plant surfaces has been intensively studied under taxonomic and ecological aspects. The light microscopic and transmission electron microscopic knowledge of epidermis has been summarized by Baker and Holloway [6]. During the last decade the application of scanning electron microscopes has greatly increased our knowledge of plant surfaces. The SEM is a surface microscope and the subject under examination is the natural primary surface of a plant - the epiderm, which covers roots, stems, leaves, flowers, fruits, and seeds. Structurally, epidermis is usually rather thick-walled and stable in a vacuum: this allows quick preparation for SEM examinations, without the need for complicated dehydration techniques. To quite an extent SEM micrographs only serve to illustrate characters well-known by light microscopy.

The study was conducted using anatomical characters to identify and delimit the Nigerian taxa at the tribal level.

MATERIALS AND METHODS

Specimen Collection

Leaves of the species of the Nigerian Bignoniaceae were collected from the field for this study. Portions of the leaves were taken from materials obtained from the field collections.

Scanning Electron Microscopy

The methods of Ayodele and Zhou [7] were adopted. Small pieces (c. 5mm^2) of each leaf were fixed to labeled stubs with double sided adhesive tape. Each sample was coated with 100% Platinum portion in a sputter coating unit, scanned and photographed using the JEOL JSM – 6360A Analytical Scanning Electron Microscope with Energy Dispersive Spectrometer at the Scanning Electron Microscope unit of the Department of Physics, University of Pune, Pune – India.

RESULTS

Table **1** showed the qualitative leaf anatomical characters of the leaf epidermis of the family Bignoniaceae in Nigeria while Plates **1-6** showed the

Table 1: Qualitative Leaf Anatomical Characters of the Species of Bignoniaceae in Nigeria

S/N	Species	Stomata type/position	Type of Trichome	Cuticular Wax	Striae occurrence
1.	*Crescentia cujete* Linn.	Anomocytic/raised	Glandular/Peltate	Present	Absent on both surfaces
2.	*Jacaranda mimosifolia* D.Don.	Anomocytic/sunken	Glandular/Peltate	Present	Present on the abaxial Surface
3.	*Kigelia Africana* (Lam) Benth	Diacytic/sunken	Glandular/Peltate	Present	Absent on both surfaces
4.	*Markhamia tomentosa* (Benth) K. Schum.	Anomocytic/raised	Glandular and Non-glandular	Present	Absent on both surfaces

(Table 1). Continued.

S/N	Species	Stomata type/position	Type of Trichome	Cuticular Wax	Striae occurrence
5.	*Newbouldia laevis* (P. Beauv.) Seemann ex Bureau.	Anomocytic/raised	Glandular/peltate	Present	Absent on both surfaces
6.	*Spathodea campanulata* P. Beauv.	Anomocytic/raised	Absent	Present	Absent on both surfaces
7.	*Stereospermum acuminatissimum* K. Schum.	Anomocytic/raised	Glandular/peltate	Present	Absent on both surface
8.	*Stereospermum kunthianum* Cham.	Anomocytic/raised	Glandular/peltate	Present	Absent on both surfaces
9.	*Tabebuia rosea* (Berthol) D. C.	Anomocytic/raised	Glandular/peltate	Present	Present on abaxial surface
10.	*Tecoma stans* (Linn) H, B &K.	Anomocytic/raised	Glandular/peltate	Absent	Present on abaxial sueface
11.	*Tecoma capensis*	Anomocytic/raised	Non-glandular	Present	Present on both abaxial and adaxial surfaces

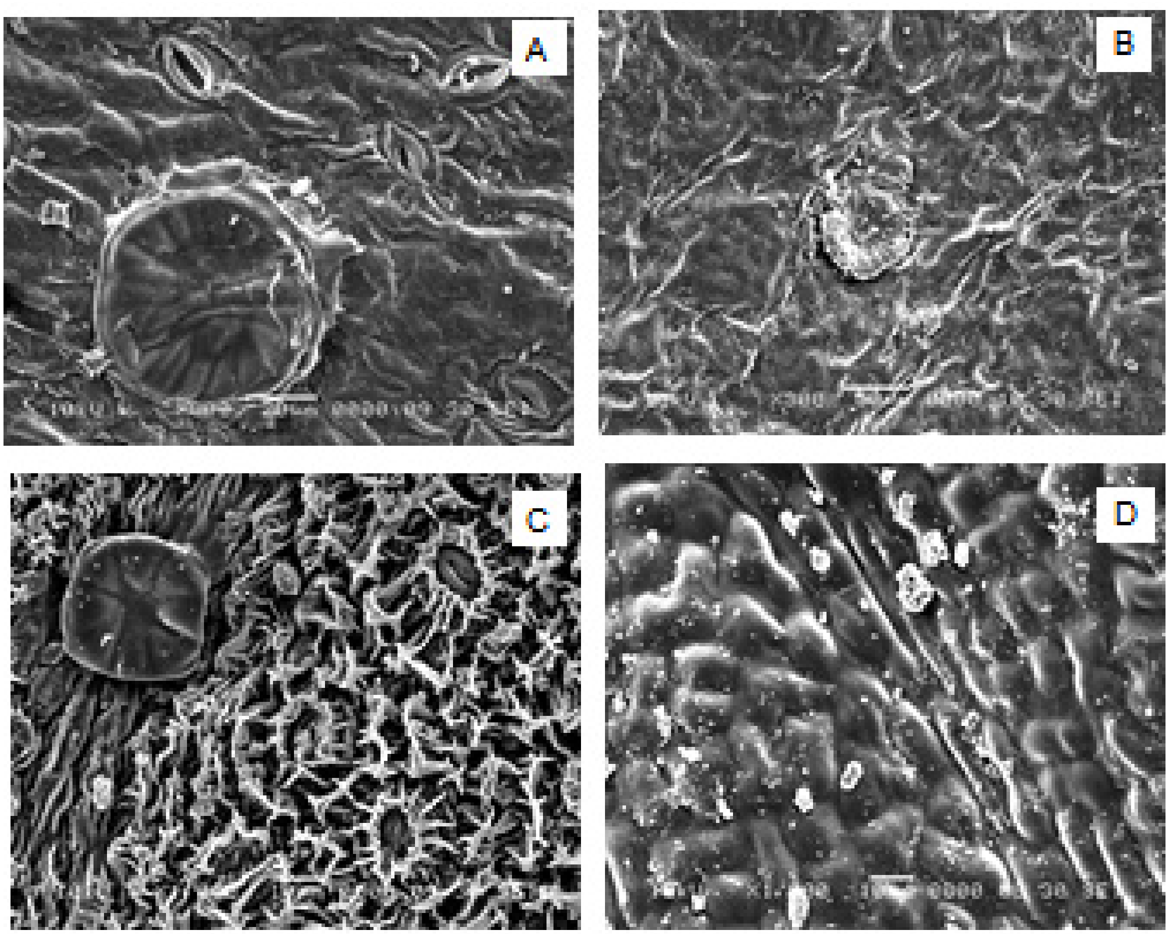

Plate 1: Scanning Electron Micrograph of the leaves of Bignoniaceae in Nigeria.

(**A**). *Crescentia cujete* (lower surface) showing raised stomata (Anomocytic type), peltate trichome and wavy anticlinal cell walls. (**B**). *Crescentia cujete* (upper surface) showing peltate trichome, wavy anticlinal cell walls and no stomata. (**C**). *Jacaranda mimosifolia* (lower surface) showing peltate trichome, sunken stomata and wavy anticlinal cell walls. (**D**). *Jacaranda mimosifolia* (upper surface) showing curved anticlinal walls pattern and no stomata.

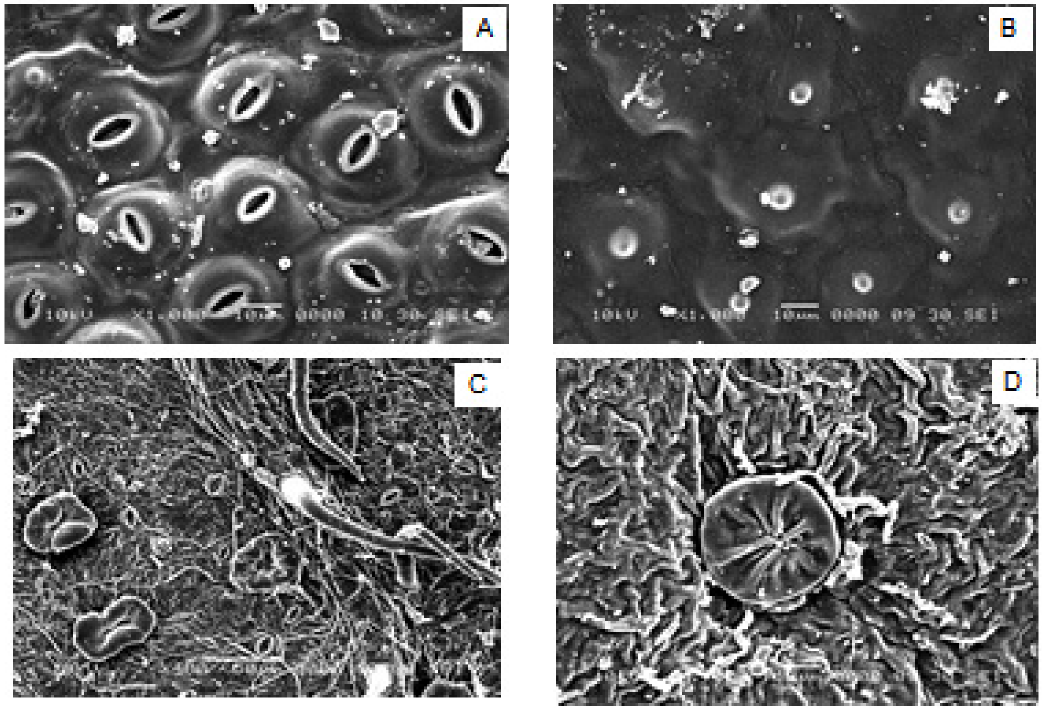

Plate 2: scanning Electron Micrograph of the leaves of Bignoniaceae in Nigeria.

(**A**). *Kigelia Africana* (lower surface) showing raised diacytic stomata and curved cell walls; (**B**). *Kigelia Africana* (lower surface) showing curved anticlinal cell walls and no stomata; (**C**). *Markhania tomentosa* (lower surface) showing long non-glandular trichomes, peltate trichomes raised stomata and wavy cell walls; (**D**). *Markhania tomentosa* (upper surface) showing peltate trichome, and no stomata.

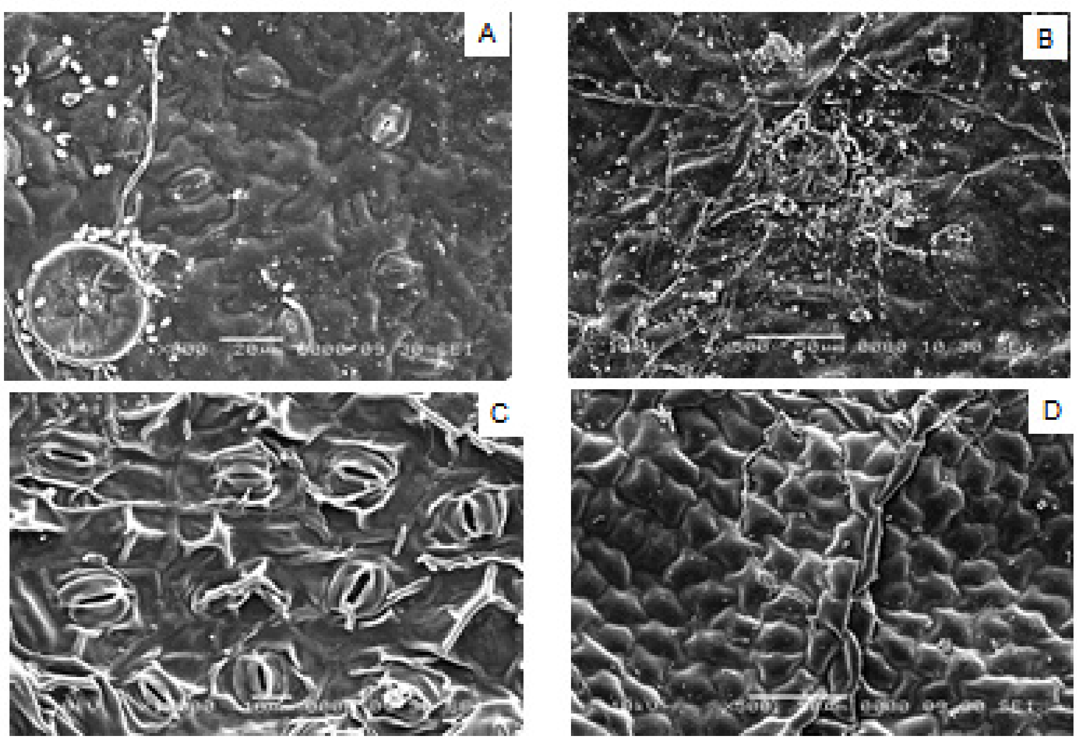

Plate 3: Scanning Electron Micrograph of the leaves of Bignoniaceae in Nigeria.

(**A**). *Newboudia laevis* (lower surface) showing raised stomata, peltata trichomes and curved cell walls; (**B**). *Newboudia laevis* (upper surface) showing peltate trichomes, curved cell walls and no stomata; (**C**). *Spathodea campanulata* (lower surface) showing raised stomata and wavy cell walls, (**D**). Spathodea *campanulata* (upper surface) showing curved cell walls and no stomata.

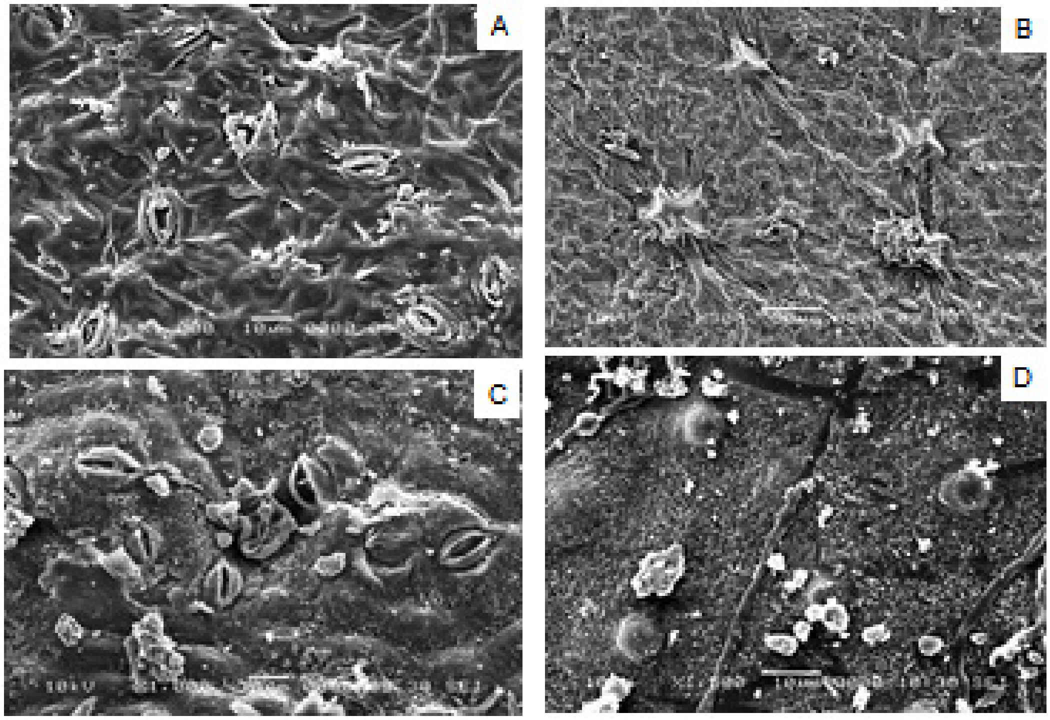

Plate 4: Scanning Electron Micrograph of the leaves of Bignoniaceae in Nigeria.

(**A**). *Stereospermum acuminatissimum* (lower surface) showing raised stomata and wavy cell wall; (**B**). *Stereospermum acuminatissimum* (upper surface) showing peltate trichome, wavy cell walls, and no stomata; (**C**). *Stereospermum kunthianum* (lower surface) showing raised and wax on the leaf, (**D**). *Stereospermum kunthianum* (upper surface) wax on the leafsurface and no stomata.

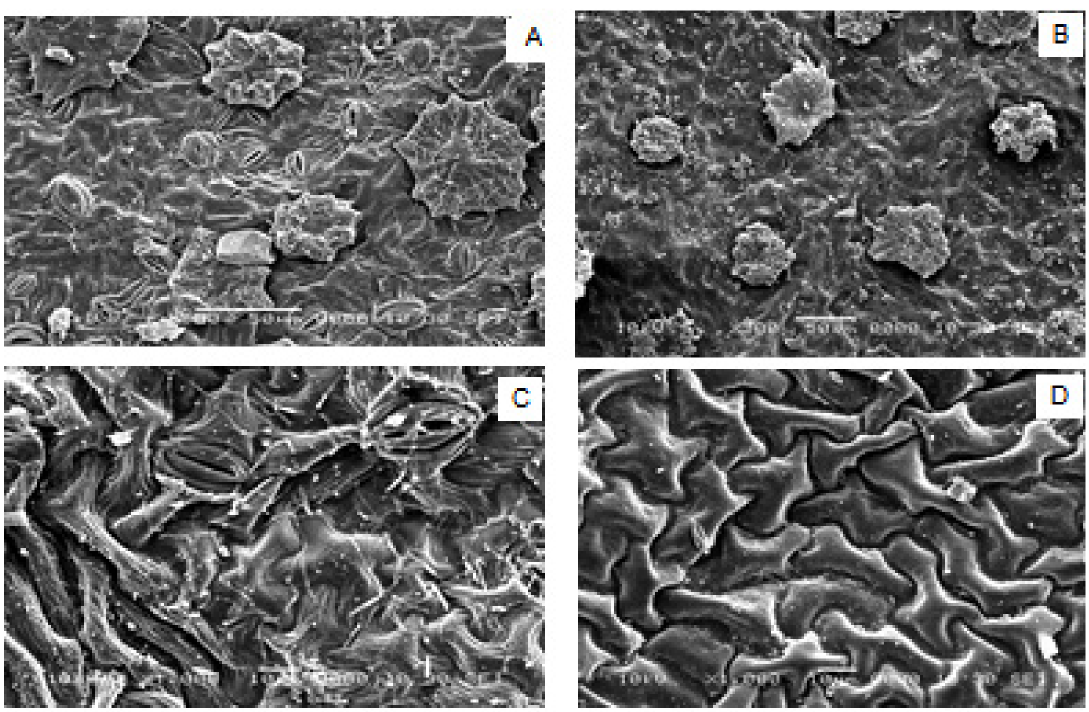

Plate 5: Scanning Electron Micrograph of the leaves of Bignoniaceae in Nigeria

(**A**). *Tabebuia rosea* (lower surface) showing raised, peltate trichome and wavy cell walls; (**B**). *Tabebuia rosea* (upper surface) showing peltate trichome, wax on the leaf surface and wavy cell walls; (**C**). *Tecoma stans* (lower surface) showing raised stomata, wavy cell walls and striations on the epidermis, (**D**). *Tecoma stans* (upper surface) showing curved cell walls and on stomata.

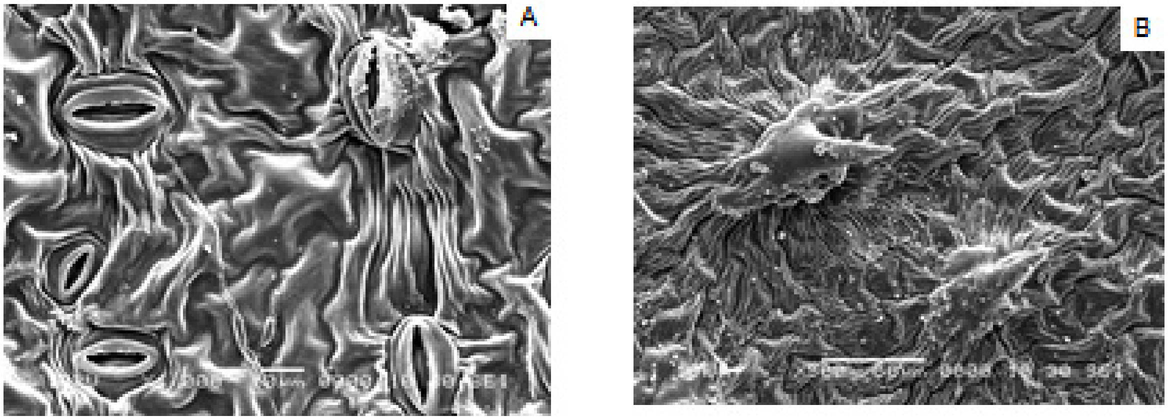

Plate 6: Scanning Electron Micrograph of the leaves of Bignoniaceae in Nigeria.

(**A**). *Tecoma capensis* (lower surface) showing raised stomata curved cell walls and striations on the epidermis; (**B**). *Tecoma capensis* (upper surface) showing non-glandular trichomes, with pointed ends, wavy cell walls and striations on the epidermis.

scanning electron micrographs of all the species studied.

DISCUSSION AND CONCLUSION

The species studied were hypostomatic with stomata restricted to the lower surfaces of the leaves. A stoma (*pl.* stomata) is a microscopic pore on the surface (epidermis) of land plants. It is surrounded by a pair of specialized epidermal cells called guard cells, which act as a turgor-driven valve that open and close the pores in response to given environmental conditions. The presence of countless numbers of stomata is critical for plant function. Typically, the plant epidermis is tightly sealed by wax-coated, interlocking epidermal pavement cells, which protect the plant body from the dry atmosphere and UV-rays. At the same time plants must be able to breathe, or exchange carbon dioxide and oxygen, for photosynthesis and respiration. Stomata act as a gateway for efficient gas exchange and water movement from the roots through the vasculature to the atmosphere. Transpiration *via* stomata supplies water and minerals to the entire plant system. When a plant encounters adverse environmental conditions, such as drought, a plant hormone called abscisic acid triggers stomata to shut tightly in order to prevent plants from dehydration and wilting. Stomata function is important beyond the level of plant physiology and function, and its significance reaches from evolutionary history to atmospheric and environmental sciences. For instance, the acquisition of stomata is considered one of the key developmental innovations that allowed plants to conquer the terrestrial environment, an event that occurred around 400 million years ago [8, 9]. Stomata impact our present global environment as well. Alistair & Woodward [10] estimate that 3 x 10^{18} grams of carbon pass through stomata and fixed into carbohydrate every year, and in addition the total water content of the atmosphere is estimated to be recycled through stomata every half year. Importantly, stomata impact atmospheric environment, and in turn, atmospheric environment drives changes in stomata patterning. It is generally accepted, both from the studies of fossil records and extant plants, that high carbon dioxide condition decreases numbers/density of stomata.

Glandular peltate tichomes were found on the following species: on abaxial and adaxial surfaces of *C. cujete*, *M. tomentosa, N. laevis,* and *T. rosea*; on only the abaxial surfaces of *J. mimosifolia* and *T. stans;* whereas, those of *S. acuminatissimum* have the peltate trichome only on the adaxial surface. *M. tomentosa* had both glandular and non-glandular trichomes on the abaxial surface while *T. capensis* had only non-glandular trichomes on the adaxial surface of the leaf. Trichomes are hair-like appendages that develop from cells of the aerial epidermis and are produced by most plant species. Leaf trichomes can serve several functions including protection against damage from herbivores. Trichomes are unicellular or multicellular outgrowths that originate from the aerial epidermis and which vary in morphological features, location and mode of secretion. A long history of published literature indicates that the type and density of trichomes differ among species and may vary in organs of the same plant [11]. It has been suggested that non-glandular trichomes serve various functions in plants i.e. to reduce the heat load, reflectance of UV light, provide protection from insects and herbivores, increase tolerance to freezing and maintain water balance in leaves [12-14]. The glandular trichomes are termed peltate or capitate, depending on the structure of the secretory head. In general, capitate glandular trichomes have one or two secretory, disk cells, while peltate trichomes may have up to eight cells in the disk. In both cases, the secretory substance is accumulated in a subcuticular space, outside of the cell wall. Peltate trichomes produce most of the essential oil, with terpenes comprising the main component [15, 16]. Along with small amounts of other monoterpenes and sesquiterpenes, the essential oil β-Phellandrene is a main component of the secretory product [17]. In addition to small amounts of essential oil, capitate trichomes normally secrete varying amounts of polysaccharides. Peltate trichomes have short one-celled stalks and large flattened heads of ca. 60–90 μm in diameter, which are formed by four or eight cells arranged in a simple disk or by 12– 18 cells in two concentric circles [18]. Meng-qi Liu and J-feng Liu [19] observed that the secondary metabolites secreted by peltata trichomes of *Isoden rubescens* (Lamiaceae), included terpenoids, flavonoids, carbohydrates, phenolics and alkaloids. Incidentally, all these metabolites were observed in the leaf and stem bark of species of Bignoniacae studied except carbohydrate in the leaf and stem bark of *T. rosea*; and alkaloids were found to be absent on all parts of the species studied.

Based on this study, striations were seen in *Tabebuia rosea, Tecoma stans* and *Tecoma capensis* which happen to belong to the same tribe (Tecomeae) with *M. tomentosa, N. laevis*, *Spathodea campanulata, Stereospermum acuminatissimum*, and *Stereospermum kunthianum*. On the basis of this and the pollen analysis [20], it is suggested that *Tabebuia* and *Tecoma* be in separate tribe. A reclassification of the family is hereby suggested. It is suggested that the genus *Tabebuia* and *Tecoma* etc. be the true *Tecomeae* and the others (*Markhamia*, *Newbouldia, Spathodea* and *Stereospermum* etc.) be put in separate tribe based on the ultra-structure of the pollen grain [20] and the presence of straie on their leaf surfaces. This was also reaffirmed by the phylogenetic analysis where *Tecoma* and *Tabebuia* were found on the lower part of the phylogenetic trees [21]. The tribe 'Spathodeae' is hereby suggested.

Previous Taxonomic Classification

Tribes

1. **Bignonieae** (not represented in Nigeria)
2. **Crescentieae** (represented by: *Crescentia cujete*)
3. **Coleeae** (represented by: *Kigelia africana)*
4. **Eccremocarpeae** (represented by: *Jacaranda mimosifolia)*
5. **Oroxyleae** (not represented in Nigeria)
6. **Tourrettieae** (not represented in Nigeria**)**
7. **Tecomeae** (represented by: Markhamia tomentosa, Newbouldia laevis, Spathodea campanulata Tabebuia rosea, Stereospermum acuminatissimum, S. kunthianum, Tecoma stans and Tecoma capensis)

Family: Bignoniaceae

Suggested taxonomic classification

Tribes

1. **Bignonieae** (not represented in Nigeria)
2. **Crescentieae** (represented by: *Crescentia cujete*)
3. **Coleeae** (represented by: *Kigelia africana)*
4. **Eccremocarpeae** (represented by: *Jacaranda mimosifolia)*
5. **Oroxyleae** (not represented in Nigeria)
6. **Tourrettieae** (not represented in Nigeria)
7. **Tecomeae:** (represented by: Tecoma stans, Tecoma capensis , Tabebuia rosea etc.)
8. **Spathodeae** (Suggested tribe): (represented by: Markhamia tomentosa, Newbouldia laevis, Spathodea campanulata, Stereospermum acuminatissimum, S. kunthianum, etc.

Family: Bignoniaceae

ACKNOWLEDGEMENT

We would like to acknowledge the Director General/CEO NIPRD, Prof. K.S. Gamaniel for his support. We also acknowledge the Head of Department, Physics Department, University of Pune, Pune-India, and especially Mr. Suresh Shinde (the laboratory technologist). We would also want to appreciate the HOD, Microbiology and Biotechnology Department (Dr. Dhamole, P.) and The Principal, SCOE Pune - India for their assistance. We also acknowledge all those that worked with me in the Laboratory (SCOE) Pune - India. The last but not the least we acknowledge Third World Academy of Science (TWAS), FR number: 3240239556 for sponsoring the study.

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Received on 09-04-2014 Accepted on 05-06-2014 Published on 09-09-2014

DOI: http://dx.doi.org/10.12974/2311-858X.2014.02.01.6