

TAXONOMIC SIGNIFICANCE OF FOLIAR AND STEM ANATOMY OF
PIPTADENIASTRUM AFRICANUM (HOOK.F.) BRENNAN AND *CATHORMION*
ALTISSIMUM (HOOK.F.) HUTCH. & DANDY

ABSTRACT

The current monotypic status of the genus *Piptadeniastrum* Brenan is reasonably challenged as *Cathormion altissimum* is claimed to be closely related to *Piptadeniastrum africanum* – the only species in the genus – by some indigenous people of Bayelsa State. This work is a systematic examination of this claim by studying the foliar and stem anatomy of *P. africanum* and *C. altissimum*. Leaf peels and stem sections were made and studied according to standard procedures. Anatomical characteristics considered include stomata distribution, stomatal Index, type of stomata, the shape of leaf epidermal cells and anticlinal walls, the outline of the stem, and the arrangement of vascular bundles. Results showed that stomatal distribution in both plants was hypostomatic. Stomatal index and stomata type were 21.5% and mainly anomocytic respectively, for *P. africanum* and 14.4% and mainly paracytic, respectively, for *C. altissimum*. Epidermal cell shape is irregular with undulating anticlinal walls in *P. africanum*, while epidermal cell shape is rectangular to polygonal with curved or straight anticlinal walls in *C. altissimum*. The outline of the stem is angular with four pronounced ridges and furrows in *P. africanum* while stem outline is wavy with very mild protrusions in *C. altissimum*. The differences in anatomical characteristics may be justifying the placement of these plants in different genera. *Piptadeniastrum* Brenan is monotypic.

Keywords: *Piptadeniastrum africanum*, *Cathormion altissimum*, anatomical character, anticlinal walls, paracytic, hypostomatic, stomatal index

INTRODUCTION

The species *P. africanum* is a forest tree of the rain forest, published and widely accepted as the only species in the genus *Piptadeniastrum* Brenan, a genus that belongs to the family Fabaceae Lindl, Subfamily Mimosoideae (Airy Shaw, 1985). *P. africanum* (Hook.f.) Brenan is endemic to tropical Africa and occurs in the Niger Delta, especially on riverbanks in the riverine areas of the rainforest (Brenan, 1955; Hutchinson & Dalziel, 1958; Nyananyo, 2006). This forest tree is of great interest due to its wide range of ethnobotanical uses – medicinal, domestic, social, and agricultural applications. Thus, studies have concentrated on its

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chemical constituents and their usefulness to man. Consequently, there are comparatively few taxonomic studies that are necessary to clear the doubts that surround its monotypic taxonomic status. This research gap is evident because names such as *Piptadeniastrum manni* Oliver, with an unresolved status and low confidence level, were placed in the Plant List (2010) and later placed with an accepted status and medium confidence level (The Plant List, 2013), but placed as a species of the genus *Piptadeniastrum* Brenan in TROPICOS (2013). Moreover, reports of varieties of *P. africanum* such as *Piptadeniastrum africanum* var. *africanum* and *Piptadeniastrum africanum* var. *brachysperma* Pellergr. exist in the literature (The Plant List, 2013).

Similarly, the identification, nomenclature, and taxonomic status of this species (whether it is the only species in the genus *Piptadeniastrum* Brenan) among some indigenous people in Bayelsa State is unclear. While most of the local people confidently affirm that there are two types of plants, they disagree on the forms in which they occur in the wild. A survey in the study area showed that one of the trees closely related to *P. africanum* is *C. altissimum* (Youkparigha, Nyananyo, & Oyedeji, 2019). Also of the Fabaceae family, *C. altissimum* is of great ethnobotanical importance. It is used as food spice (Jolaoso, Ajayi, Ogunmuyiwa, & Albert, 2012), medicine (Burkill, 1985; Lemmens, 2006), and wood of choice for carving durable, quality kitchen utensils like mortar, pestle, and spatula that are indispensable in a typical Niger Delta family. These carved utensils serve as valuable traditional gifts to new couples during marriage weddings.

Observation is a critical element of the scientific method. The indigenous people, who are close to the forest and have depended on it for their sustenance over the years, have significant ethnobotanical knowledge about these trees, and the importance of applying appropriate scientific methods to study their observations cannot be overemphasized. The fact that indigenous knowledge, sometimes called Traditional Ecological Knowledge (TEK), has contributed immensely to discoveries in natural sciences and medically necessary substances in plants cannot be controverted (Stephen, 1993; Reyes-Garcia, 2010; Popp, 2018). Moreover, when the identity of even a single plant is not sure and authentic from broad-based, empirical taxonomic evidence, then extant erroneous phylogeny in the relevant taxa is undeniable. This study, therefore, is aimed at describing some leaf epidermal and stem anatomical characteristics that may be significant, at least in serving as supportive taxonomic evidence, in the description and delimitation of *P. africanum* and *C. altissimum* as either belonging to different genera (as they are present) or the possibility of belonging to the same genus and thus establishing their degree of relatedness.

MATERIALS AND METHODS

Study area: Bayelsa State, where this study was carried out, comprises eight Local Government Areas (Figure 1). The state is located within latitudes 04⁰15' North and 05⁰23' south and longitudes 05⁰ 22' West and 06⁰ 45' East. It is bounded by Delta State on the North, Rivers State on the East, and on the west and south by the Atlantic Ocean (Figure 1).

It is one of the youngest states in Nigeria, created along with five others. Consequently, its natural ecosystems are under pressure from overexploitation because of the drive for much-needed infrastructural development. Like all other states in the Niger Delta, Bayelsa state has species-rich ecological systems ranging from mangrove swamps, estuaries, and freshwater swamps to rain forests with an impressive network of rivers and creeks (Izah, 2018; Izah and Seiyaboh, 2018a,b; Izah et al., 2017, 2018; Kigigha, Seiyaboh, Obuah, & Izah, 2017). Apart from the traditional slash and bush agricultural system and intensive commercial lumbering due to roads which have increased the ease of accessing the hinterlands, the crude oil exploration and exploitation activities of multinational companies have led to the pollution of streams and rivers, forest destruction, and biodiversity loss. The ultimate result is a decrease in the carrying capacity of the ecosystems and the attendant delivery of poor-quality ecosystem services. Consequently, the indigenous people who depended on these natural resources for survival are violently stripped of their ancient livelihood and left to pine away in sickness and poverty (Kadafa, 2012; Johnson, 2017). Also, the once abundant prime-quality timber species in forests within the state are now becoming rare due to extensive exploitation with little or no control measures on the part of the state (Youkparigha, Nyananyo & Oyedeji, 2019). Some of the common tree species are *Vitex grandifolia* (Epidi et al., 2016a), *Alstoniaboonei*(Epidi et al., 2016b), *Raphia* spp, *Cleistopholis patens*, *Elaisguineensis*, *Iringiagabonensis*, *Anthocleistaspp*, *Cathormionaltissimum*, *Treculiaafricana*, *Uapacaspp*, *Symphonia globulifera*, *Taminalia* spp, *Harungana madagascariensis*, and *Alchornea cordifolia* (Ihinmikaiye, & Unanaonwi, 2018; Youkparigha, & Patani, 2019).

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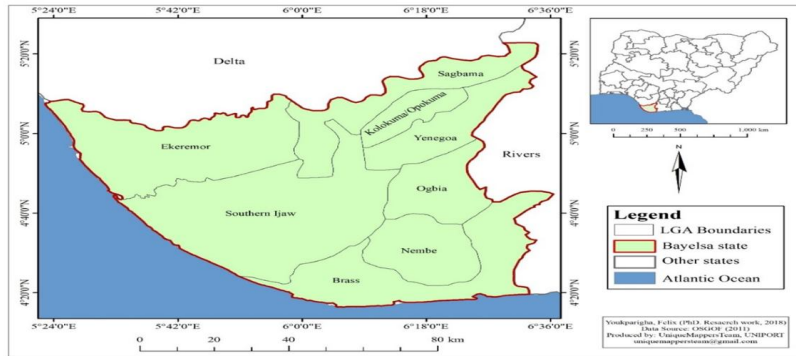


Fig. 1: Bayelsa State, showing the Local Government Areas (Inset: Map of Nigeria showing Study Area)

Sample collection and identification: Matured samples of the plants (Figure 2) were collected from the study area and identified at the Forest Herbarium Ibadan of the Forestry Research Institute of Nigeria (FRIN). Voucher specimens of the plants – *P. africanum* (FHI 110137) and *C. altissimum* (110884) – were deposited in the herbarium for reference and further studies.

(a)



(b)



Fig. 2(a-b): (a) *C. altissimum* (b) *P. africanum*

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Leaf epidermal studies: Cuttings from identical regions were made from fresh leaves of the plants collected from the field. Peels from the adaxial and abaxial surfaces of the leaves were obtained carefully with the aid of pins and forceps. Samples were stained with alcian blue or 1% safranin, rinsed in distilled water to remove excess stain, and mounted on microscope slides with a drop of glycerol. Epidermal features such as the shape of epidermal cells, type of anticlinal wall, stomatal distribution, stomata type, and stomatal index were studied. Stomatal index (SI) was estimated²⁰, while the terminology for stomata type was taken after Metcalfe and Chalk²¹.

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Stem anatomical studies: Matured plants from fresh vegetation were used for this study. Cut sections of the stem were fixed at room temperature (23⁰C – 28⁰C) in Formaldehyde-acetic acid- alcohol (1 part of 40% formaldehyde, 1 part of acetic acid, 18 parts of 70% ethanol) for at least 48 hours. Then, samples were washed with several changes of distilled water, dehydrated through 2-hour incubations in graded alcohol solutions (30%, 50%, 70%, and 100%) at room temperature, and embedded in wax. Hand sections were cut with blades. Thin sections were selected and dewaxed by passing the specimens through a series of 3 hours of incubations in graded ethanol: chloroform solutions (1:3, 1:1, and 3:1), with a final 3-hour incubation in absolute ethanol. Samples were stained with alcian blue, counterstained with 1% safranin, and mounted on microscope slides. Photomicrographs were acquired with a digital camera.

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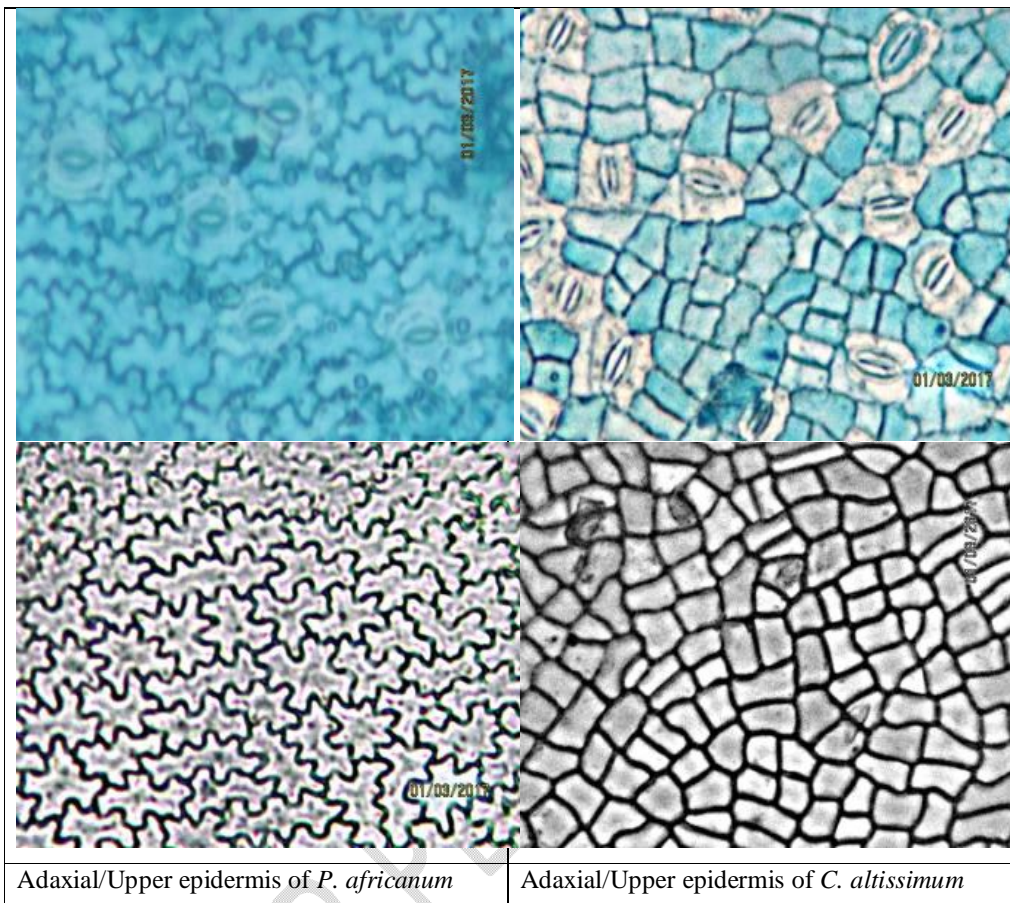
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RESULTS

Leaf epidermal studies: Results of the leaf epidermal studies of the plants are presented in table 1 and figure 3.

Table 1: Leaf epidermal characteristics of *P. africanum* and *C. altissimum*

Characters/ Species	Stomatal Index (%)	Stomata Type	Stomata distribution	Epidermal cell shape	Anticlinal wall
<i>P. africanum</i>	21.5	Mainly anomocytic	Hypostomatic	Irregular	Undulating
<i>C. altissimum</i>	14.4	Mainly paracytic	Hypostomatic	Rectangular/ polygonal	Curved/ straight



Adaxial/Upper epidermis of *P. africanum*

Adaxial/Upper epidermis of *C. altissimum*

Figure 3: Adaxial and Abaxial leaf epidermis of *P. africanum* and *C. altissimum*

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Stem anatomical studies: Result of anatomical studies of the stems of *P. africanum* and *C. altissimum* is shown in Figure 4.

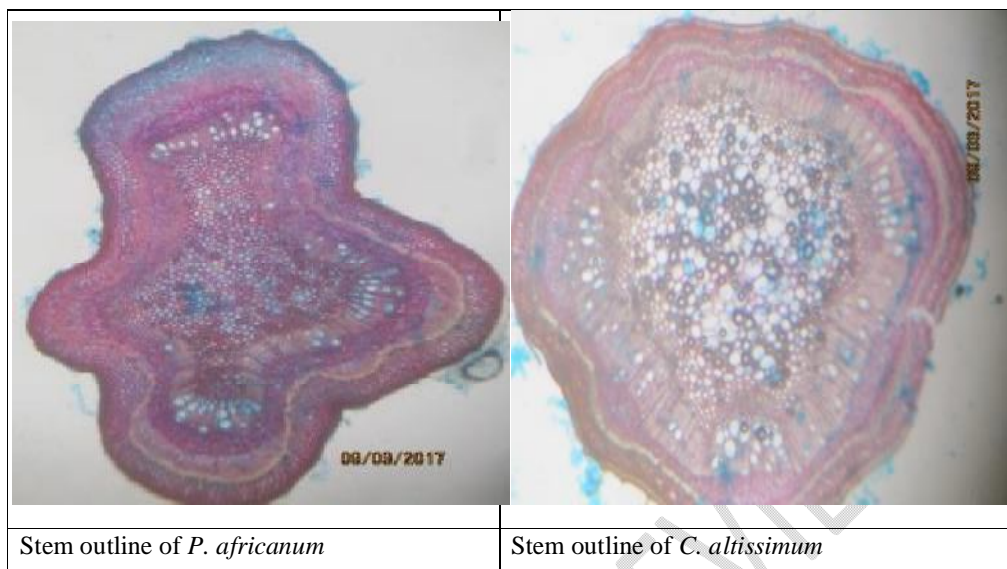


Figure 4: Stem outlines of *P. africanum* and *C. altissimum*

DISCUSSION

Stomatal distribution in both the leaves of *P. africanum* and *C. altissimum* is Hypostomatic i.e. stomata are found only on the abaxial surfaces of the leaves. The epidermal cell shape is irregular with undulating anticlinal walls in *P. africanum*, while the epidermal cell shape for *C. altissimum* is rectangular/polygonal with curved/straight anticlinal walls. The stomatal index is 21.5% in *P. africanum* with mainly anomocytic stomata type, while the stomatal Index for *C. altissimum* is 14.4% with mainly paracytic stomata.

The taxonomical value of foliar epidermal characters such as hairs, stomata, and epidermal cells has been well established. Though epidermal cells vary considerably in size, shape, and outline in different plants, they are affected by environmental factors. Consequently, the shape of epidermal cells should be helpful as confirmatory evidence²¹. However, variations in leaf epidermal characters have been used in the identification and classification of several plant groups because they are primarily controlled by genes and are structurally diverse 22-25. Nurul-Aini *et al*²⁶ showed that leaf micromorphological features such as stomata characteristics and the presence of trichomes were taxonomically significant in some selected taxa of *Acanthaceae*. Researchers have found leaf epidermal micromorphological characteristics such as differences in epidermal cell shape and size and the stomatal index, and the pattern of anticlinal walls of epidermal cells to be of taxonomic significance among 43 species of *Allium*²⁷. It was reported that the pattern of the anticlinal walls of epidermal

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cells was consistent within a species, but there were differences between species. Consequently, micromorphological characteristics of the epidermis of *P. africanum* and *C. altissimum* in this study were of taxonomic significance in their delimitation into different genera.

The stem of *P. africanum* has an angular outline with four pronounced ridges and furrows, while the stem of *C. altissimum* has a wavy outline. Furthermore, although the vascular bundles of both plants are scattered in the ground tissue, they differ in the patterns of grouping and arrangement. In *C. altissimum*, the vascular bundles are arranged in groups of about five, while in *P. africanum*, vascular bundles are concentrated at the ridges or protrusions. These anatomical characteristics may be significant in the delimitation of these plants.

The significance of using stem anatomical features in collaboration with other lines of evidence in taxonomy is well known²⁸⁻³². Cavente, Andreato, and Vieira³³ worked on six species of *Rhipsalis* and found that features of the stem anatomy discriminate between the species. Furthermore, Kaplan & Symoens³⁴ used stem anatomy to distinguish between two confusing African taxa (*Potamogeton schweinfurthii* and *Potamogeton richardii*).

CONCLUSION

This study investigated the degree of relatedness between *P. africanum* and *C. altissimum*, two plants of immense ethnobotanical importance in the tropical rainforest of Bayelsa state in Nigeria. Some foliar and stem anatomical features of these two plants were studied. The results of the studies showed no possible degree of relationship that could warrant the placement of *C. altissimum* under the genus *Piptadeniastrum* Brenan. Consequently, the current monotypic status of the genus *Piptadeniastrum* Brenan is maintained.

SIGNIFICANCE STATEMENT

The nomenclatural conflict between *P. africanum* and *C. altissimum*, arising from the similar morphology of the leaves, was systematically resolved using evidence from foliar and stem anatomy. Furthermore, the information from the results of this study has enriched the existing literature on the taxonomy of plants.

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REFERENCES

1. Airy Shaw, H.K. 1985. *J.C. Willis: A dictionary of flowering plants and ferns* 8th ed. Revised. Cambridge: Cambridge University Press. 1245pp
2. Brenan, J.P.M., 1955. *Piptadeniastrum africanum*. *Kew Bull.*, 179.
3. Hutchinson, J. and J.M. Dalziel, 1958. *Flora of West Tropical Africa*. (2nd ed.). Vol. 1, Part 2. Revised by Keay, R.W.J. London: Crown Agents for Oversea Governments and Administrations.
4. Nyananyo, B.L. 2006. *Plants from the Niger Delta*. Nigeria: Onyoma Research
5. The Plant List 2010. *Piptadeniastrum*. Available from <https://www.theplantlist.org/> Accessed [19-05-2013]
6. The Plant List (2013). *Piptadeniastrum*. Available from <https://www.theplantlist.org/> Accessed [19-05-2013]
7. TROPICOS, 2013. *Piptadeniastrum manni* Oliv. Missouri Botanical Garden. Retrieved from <https://www.tropicos.org/Name/13069459>. Accessed [19-05-2013].
8. Youkparigha, F.O., B.L. Nyananyo, and A.A. Oyedeji, 2019. Morphology and Distribution of *Piptadeniastrum africanum* (Hook.f.) Brenan and *Cathormion altissimum* (Hook.f.) Hutch. & Dandy in Bayelsa State, Nigeria". *IJARB* 5(2), 8-14.
9. Jolaoso, A.A., J.O. Ajayi, J.O. Ogunmuyiwa, J.O. and O.M. Albert, 2012. Changes in Functional Properties as a Measure of Biochemical Deterioration of Oso (fermented seeds of *Cathormion altissimum*). *JETEAS*, 3(4), 608-613.
10. Burkill, H.M., 1985. *The Useful Plants of West Tropical Africa*. Edn. 2, Vol. 3, Families J-l. Royal Botanic Gardens, Kew, Richmond, Surrey, England. 857pp.
11. Lemmens, R.H.M.J. 2006. *Albizia altissima* Hook.f. Record for PROTA4U. Louppe, D. Oteng-Amoako, A.A. and Brink, M. (Editors). PROTA (Plant Resources for Tropical Africa), Wageningen, Netherlands. Retrieved from <https://www.prota4u.org/search.asp>. Accessed [18-04-2018]
12. Stephen, B.B. 1993. Indigenous knowledge of biological resources and Intellectual Property Rights: The role of Anthropology. *Am Anthropol.*, 95(3), 653-671
13. Reyes-Garcia, V. 2010. The relevance of traditional knowledge systems for ethnopharmacological research: theoretical and methodological contributions. *J Ethnobiol Ethnomed*, 6, 32
14. Popp, J. 2018. How indigenous knowledge advances modern science and technology. The Conversation. Available From: <https://theconversation.com/how-indigenous-knowledge-advances-modern-science-and-technology-89351>. Accessed [18-04-2020]
15. Kigigha, L.T., E.I. Seiyaboh, V.J. Obuah, and S.C. Izah, 2017. Contamination of River Nun at Amassoma, Bayelsa State, Nigeria due to microbial diversity in sediments. *Environmental and Toxicology Studies*, 2, 2.
16. Kadafa, A, A. 2012. Environmental Impacts of Oil Exploration and Exploitation in the Niger Delta of Nigeria. *GJSFR*, 12:3
17. Johnson, S. 2017. The difference between tropical monsoon and a tropical rainforest. Retrieved from: <https://www.getawaytips.azcentral.com>
18. Ihinmikaiye, S. and O.E. Unanaonwi, 2018. Tree species structure and diversity in the lowland-rain forest zone of Bayelsa State. *J Ecol & Nat Resour*, 2(2), 126

19. Youkparigha, F.O. and D.E. Patani, (2019). Application of Diversity Indices in the Study of Trees and Shrubs of the Nun River Forest, Nigeria. *IJRSB*, 7 (11), 1-9.
20. Obembe, O.A. 2015. Stomata complex in some shrubs and trees. *GJBAHS*, 4(2), 164-172
21. Metcalfe, C.R. and L. Chalk, 1950. *Anatomy of the Dicotyledons*. Volume 1. Oxford University Press, London. 724p.
22. Pant, D.D., and B.K. Verma, 1974. Taxonomy of the genus Ephedra: Significance of stem and leaf epidermis and cuticle. *Bot. J. Linn. Soc.* 69,287-308.
23. Wilkinson, H.P. 1979. The plant surface (mainly leaf) In Metcalfe, C.R. & Chalk, L. (Eds.). *Anatomy of the Dicotyledons* (2nd ed.). Oxford, England: Oxford Press
24. Adegbite, A.E., 1995. Leaf epidermal studies in three Nigerian species of *Aspilia* (Heliantheae- Asteraceae) and two hybrids. *Niger.J. Bot.*, 8, 25-33.
25. Pandey, B.P. 2004. *Plant Anatomy: Including Embryology and Morphogenesis of Angiosperms* (6th ed.). New Delhi: S. Chand.
26. Nurul-Aini, C.A.C.; T. Noraini, A. Latiff, A.J. Amirul-Aiman, A.R. Ruzi, and S. Idris, 2014. Taxonomic Significance of Leaf Micromorphology in Some Selected Taxa of Acanthaceae (Peninsular Malaysia). AIP Conference Proceedings, 727-733
27. Chen-Yi Lin & T. Dun-Yan, 2015. The taxonomic significance of leaf epidermal micromorphological characters in distinguishing 43 species of *Allium* l. (Amaryllidaceae) from Central Asia. *Pak. J. Bot.*, 47(5): 1979-1988.
28. Wiegleb, G.1990.The importance of stem anatomical characters for systematics of genus *Potamogeton* L. *Flora* 184,197-208.
29. Metcalfe, C.R. 1942. On the taxonomic value of the anatomical structure of the vegetative organs of the dicotyledons: An introduction with special reference to the anatomy of the leaf and stem. *Proceedings of the Linnean Society of London* 155(3): 211-223.
30. Mbagwu, F.N., and C.N. Inyama, 2016. *Stem anatomy of three Chrysophyllum Species (Sapotaceae) and their relevance for taxonomy (Systematic Botany)*.International Conference on Plant Physiology and Pathology. June 09-10, Dallas, USA
31. Aziagba, B.O. and C.U. Okeke, 2017. Taxonomic significance of stem and petiole anatomy of three varieties of *Vigna unguiculata*. *American Journal of Life Sciences Research*, 5(1), 1-5.
32. Goswami, S. 2018. *Anatomy in relation to plant Taxonomy*. Retrieved from <https://www.biologydiscussion.com/plant-taxonomy/30463>. Accessed [13-08- 2013]
33. Cavente, A.M., R.H.P. Andreato, and R.C.Vieira, 2008. Stem anatomy of *Rhipsalis* (Cactaceae) and its relevance for taxonomy. *Plant Syst. Evol.*, 276 (1), 1-7.
34. Kaplan, Z., and J. Symoens, 2005. Taxonomy, distribution and nomenclature of three confused broad-leaved *Potamogeton* species in Africa and surrounding islands. *Bot. J. Linn. Soc.*, 148(3), 329-357.
35. Epidi, J.O, Izah, SC. and Ohimain, EI, Epiidi TT (2016a). Phytochemical, antibacterial and synergistic potency of tissues of *Vitex grandifolia*. *Biotechnological Research*, 2(2): 69-76.
36. Epiidi, J.O, Izah, SC. and Ohimain, EI (2016b). Antibacterial and Synergistic Efficacy of Extracts of *Alstoniaboonei* Tissues. *British Journal of Applied Research* 1(1), 0021-0026.
37. Izah SC (2018). Ecosystem of the Niger Delta region of Nigeria: Potentials and Threats. *Biodiversity International Journal*, 2(4):338–345.
38. Izah SC, Seiyaboh EI (2018a). Challenges of wildlife with therapeutic properties in Nigeria; a conservation perspective. *International Journal of Avian & Wildlife Biology*, 3(4):259–264.

39. Izah S.C., and Seiyaboh E.I., 2018b, Changes in the protected areas of Bayelsa state, Nigeria, *International Journal of Molecular Evolution and Biodiversity*, 8(1): 1-11
40. Izah S.C, Aigberua A.O., and Nduka J.O., 2018, Factors affecting the population trend of biodiversity in the Niger Delta region of Nigeria, *International Journal of Avian and Wildlife Biology*, 3(3):206–214.
41. Izah S.C., Angaye C.N., Aigberua A.O., Nduka J.O., 2017, Uncontrolled bush burning in the Niger Delta region of Nigeria: potential causes and impacts on biodiversity, *International Journal of Molecular Ecology and Conservation*, 7(1): 1-15
- 42.

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