TAS Trends in Agricultural Sciences

OPEN ACCESS

Anatomical Characterization of Stems of Some Medicinal Plants of the Family Euphorbiaceae in Nigeria

O.E. James, B.O. Green and M.G. Ajuru and V. Wilson

Department of Plant Science and Biotechnology, Rivers State University, Port Harcourt, Rivers State, Nigeria

ABSTRACT

Background and Objective: Detailed knowledge of plant anatomical characters and their variation among closely related species is paramount to understanding their evolution and function. Plants belonging to the Euphorbiaceae family are widely used in Nigeria since it has important secondary metabolites like tannins, alkaloids, flavonoids, phenols, triterpenes, polysterols, etc. Euphorbiaceae is an important family which contains numerous medicinal plants. Based on their medicinal importance, seven species were selected for study from four genera which include: Genus Acalypha-*A. hispida* and *A. wilkesiana*, genus Euphorbia-*E. heterophylla* and *E. hirta*, genus Jatropha-*J. curcas* and *J. gossypifolia* and the genus Manihot-*M. esculenta*. **Materials and Methods:** This study characterizes and compares the anatomical characteristics of the stem of these seven species of Euphorbiaceae occurring in some parts of the Niger Delta Region of Nigeria, as a means of providing information for quality assessment of the herbal industry. Three individuals of each species were collected, fixed, stored and prepared following usual anatomy techniques, for subsequent observation using light microscopy techniques. **Results:** The observations made of the transverse sections of the stem revealed structural arrangements and variations of the epidermis, hypodermis, cortex, vascular tissues and pith. The cell wall contours have been described in detail. The occurrence of cell inclusions was also noted in different tissues of the stem-axes. **Conclusion:** The observable characters in the seven species studied are diagnostic and taxonomically significant enough to separate and distinguish them. These features are helpful when conducting a quality control process.

KEYWORDS

Acalypha, stem anatomy, *Euphorbia*, euphorbiaceae, *Jatropha*, *Manihot*, taxonomy, inflorescences-cyathium

INTRODUCTION

The family Euphorbiaceae, commonly called the spurge family, is one of the largest families of flowering plants comprising of plants with over 300 genera and 8,000 species[1,2]. They contain a large variety of phytotoxins, a toxic substance produced by plants, A milky latex is a characteristic of the Euphorbiaceae. The family is very diverse in range, composed of all sorts of plants ranging from monoecious trees, shrubs, herbs, twiners and sometimes succulent and cactus-like[3]. Life span is perennials and annuals. This family

ISSN: 2957-9449 (Online)
ISSN: 2957-9430 (Print)
https://doi.org/10.17311/tas.2022.87.94

Received: 15 Dec. 2021
Accepted: 13 Mar. 2022
Published: 01 Oct. 2022

is easily recognized by their inflorescences-cyathium. The plant is distributed worldwide in varied environmental conditions ranging from desert to tropical rainforest. To adapt, it produces a variety of medicinally important secondary metabolites.

Acalypha hispida Burm. f. has its common name in English as Philippines Medusa, foxtail and the red hot cat's tail. *Acalypha* is the fourth largest genus of the Euphorbiaceous family and contains many species that are found in the tropical regions of the world. Its cultivation is widespread, especially as a houseplant and this has earned it, the Royal Horticultural Society's Award of Garden Merit[4].

Acalypha wilkesiana Mull. Arg. is commonly called copperleaf and Jacob's goat[5]. It grows better with light and well-drained soil and is suited to a protected shady position. Biogeographical study of *A. wilkesiana* showed that it has a wider range of distribution as it is found in the USA, Vanuatu, Papua New Guinea, Thailand, Zimbabwe, Uganda, Vietnam, Polynesia, Tanzania, Pacific, PNG, Norfolk Island, Botswana, South Africa, Zambia, Asia, The Bahamas, Nigeria, Brazil, Tonga, Bermuda, South Africa, Asia, Fiji, Hawaii, Indochina, Malaysia and Kenya[5].

Euphorbia heterophylla Linn. is known as painted plants, Mexican fire plants, fire on the mountain etc., *E. heterophylla* is a widespread weed in the tropics and has adapted to subtropical conditions. It originates from tropical and subtropical America but it is now distributed throughout the tropics and many herbicides fail to control it, hence, the rapid spread in many parts of the word[6]. Its distribution is frequent and usually abundant in Benin, Côte d'Ivoire and Tanzania; frequent but not abundant in Burkina Faso, Chad, Kenya, Mali and Nigeria, rare and not abundant in Ghana and Senegal, rare but abundant when present, in Uganda[7].

Euphorbia hirta Linn. commonly called asthma plant, garden spurge and red euphorbia is a pantropical weed possibly native to tropical and subtropical America and is widespread as a weed in the tropical and subtropical regions of the world[8,9]. Usually, they can be very invasive and can as well spread rapidly[10]. *Euphorbia hirta* thrives well in a much-brightened environment that meets so well on dry grounds[9,10]. It prefers the sandy ground or with gravel and is found in sunny to a slightly shaded environment that is not too wet premises, grassy sites, along roadsides, usually between stones[9,10].

Jatropha curcas Linn. is commonly called Barbados nut, nutmeg plant, physic nut and purging nut[11]. The origin of *J. curcas* is controversial because it is found in many countries of both central and North America which remain the only area where it was found and collected from undisturbed vegetation[11]. It is native to Central America but now grows in many tropical regions of the world naturally, including Nigeria[12].

Jatropha gossypifolia Linn. is commonly called American purging nut, figus nut, black physics nut, castor bean, wild cassava, bellyache bush etc. *J. gossypifolia* is widely cultivated in tropical countries, though it is native to the new world[13]. It is found in North America, mainly in Mexico, Hawaii, USA and Florida[14]. It is also found in African countries like South Africa, Mozambique, Zambia, Zimbabwe, Kenya, Senegal, Nigeria, Chad, Cameroon and Ghana[15]. It is found in Asia and the Pacific, particularly in Singapore, Cambodia, Indonesia, New Guinea, New Caledonia, India and La Reunion[16,17].

Manihot esculenta Crantz is known as cassava. It is native to South America where it is widely cultivated. It is found in the tropics and sometimes in subtropics of the old and new world as a very important food that provides starch. It is widely distributed in many countries like Brazil, Mexico, Central America, India, Philippines Islands, Thailand, Jamaica, Colombia, South Africa, Madagascar, Nigeria, Ghana, Federated Malay States, Indonesia, Sri Lanka, Paraguay and Fiji. Cassava grows well in well-drained soil of tropical and subtropical regions of the world where the climate is warm and humid.

Plant anatomical studies as a systematic line of evidence in plant taxonomy are used with other systematic lines to achieve good taxonomic decision making[18]. The reason is that anatomical characters are conserved and stable and so are employed as taxonomic characters in plant systematic and taxonomic studies[19]. The anatomical character, which includes root anatomy, trichomes, stem anatomy, stomata and epidermal, wood anatomy, nodal anatomy, sclereids and fibres, cambium and leaf anatomy is adopted in biosystematics and taxonomic studies to identify plants, establish genetic relationships and solve taxonomic disputes[19]. Over the years, many plants are classified based on their flower and fruits which are considered as external morphological structures whose production are usually seasonal, hence limiting their availability for study and proper identification[20]. Currently, there are calls for some novel protocol for proper and easy identification of plants at any season of the year[21]. Plant anatomical studies of the vegetative organs like leaves, roots and stems which may be found at all seasons seem to be a solution to such challenges[21].

Members of the family Euphorbiaceae are of great economic importance to our country Nigeria, as they are found useful in terms of medicine, formation of hedges, landscape and beautification, production of timber, provision of food and fodder etc. They are found in the Niger Delta Region and the entire southern part of Nigeria. Its distribution is spread across many regions of Nigeria and West Africa. As a result of the economical value of these species of Euphorbiaceae in Nigeria, it becomes expedient that this study is undertaken to provide a vivid anatomical description that will help the taxonomical characterization and the identification of the plant species even in fragmentary conditions.

MATERIALS AND METHODS

Study area: The study was carried out in the Department of Plant Science and Biotechnology Research Laboratory, Rivers State University, Port Harcourt, Rivers State, Nigeria, from June, 2020 to August, 2021. The summary of vital data obtained in the field during collection and identification of samples of studied plant species. Fresh and healthy samples of selected plants from four different genres in the family Euphorbiaceae were collected in separate bags from Rivers, Delta, Abia, Bayelsa and Imo state all from the Niger Delta of Nigeria. The collection site's ecological conditions like altitude, longitude and latitude were recorded. Other information taken includes the name of the collector, collection site, collection number and date of collection. The plant specimens were identified at the herbarium of the Department of Plant Science and Biotechnology, University of Port Harcourt, Rivers State (UPH). Identified pressed plant samples were deposited at the University of Port Harcourt (UPH) and Rivers State University (RSU) Herbarium for reference and further studies (Table 1).

Sample collection: Fresh and healthy samples of selected plants from four different genera in the family Euphorbiaceae were collected in separate bags from Rivers, Delta, Abia, Bayelsa and Imo state all from the Niger Delta of Nigeria. The plant specimens were identified at the herbarium of the Department of Plant Science and Biotechnology, University of Port Harcourt, Rivers State (UPH). Identified pressed plant samples were deposited at the University of PortHarcourt (UPH) and Rivers State University (RSU) Herbarium for reference and further studies.

Anatomical study: Stems of the plant samples studied were gotten and immediately fixed in FAA (Formalin, Alcohol and Glacial Acetic Acid) in the ratio of 1:3:1 for 24 hrs and preserved in 70% alcohol[22]. Free-hand sections of the stem were made with a safety blade and dropped into various Petri dishes which were filled with water. The sectioned stem of the plant was placed on separate slides, stained in 1% safranin and 1% fast green and dehydrated following the method[22]. Few drops of glycerol were applied and covered with coverslips which were later mounted under the light microscope for examination and viewed under ×10 and ×40 objective lens. Photomicrographs were taken using XSZ-N107 Microscope with (MA88-900) camera.

Table 1: Data collection from some states in the Niger Delta Area of Nigeria

Herbarium ID	Names of plant	Collection location	Latitude	Longitude	Altitude	Date of collection
RSU JO 01	*A. hispida*	Ofrima Building-Uniport 'PH, Rivers State	4°54'3.6"N	6°55'22.8"E	13.20 m	14th December, 2018
RSU JO 02	*A. wilkesiana*	Umuezike Ofeme-Umuahia, Abia State	5°39'57.6"N	7°25'19.2"E	71 m	15th December, 2018
RSU JO 03	*E. heterophylla*	Ihube-Okigwe, Imo State	5°50'58.28"N	7°22'56.93"E	203 m	15th December, 2018
RSU JO 04	*E. hirta*	College of Health Sciences NDU, Bayelsa State	4°58'7.9"N	6°5'40.5"E	3 m	13th December, 2018
RSU JO 05	*J. curcas*	Oleh, Delta State	5°29'2.219"N	6°12'19.9"E	4 m	13th December, 2018
RSU JO 06	*J. gossypifolia*	Oyigbo, Rivers State	4°52'52"N	7°7'94"E	11.07 m	13th December, 2018
RSU JO 07	*M. esculenta Crantz*	Agricultural Farm, Uniport, PH Rivers State	4°54'14.4"°N	6°55'22.8"E	11.40 m	12th July, 2018

Table 2: Stem anatomical characters of interest of species of family Euphorbiaceae studied

Character	*A. hispida*	*A. wilkesiana*	*E. heterophylla*	*E. hirta*	*J. curcas*	*J. gossypifolia*	*M. esculenta*
Shape of stem tranverse section	Rounded	Rounded	Notched, not fully rounded	Notched, not fully rounded	Rounded	Rounded	Rounded
Epidermal cells	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate
Hypodermis	Collenchymatous (3-4 layers)	Collenchymatous (5-6 layers)	Collenchymatous (3-4 layers)	Collenchymatous (3-4 layers)	Collenchymatous (5-6 layers)	Collenchymatous (2-3 layers)	Collenchymatous (2-3 layers)
Cortex	Parenchymatous (4-5 layers)	Parenchymatous (4-5 layers)	Parenchymatous (5-6 layers)	Parenchymatous (5-6 layers)	Parenchymatous (6-7 layers)	Parenchymatous (7-8 layers)	Parenchymatous (4-5 layers)
Cell inclusion	Present	Present	Absent	Absent	Present	Absent	Absent
Pith cavity	Parenchymatous	Parenchymatous	Hollow	Parenchymatous	Parenchymatous	Parenchymatous	Parenchymavous
Trichomes	Multicellular, glandular trichomes	Unicellular, non-glandular trichomes	Multicellular, glandular trichomes	Multicellular, glandular trichomes	Absent	Absent	Absent
Endodermis	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate
Pericycle	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate
Medullary rays	Parenchymatous	Parenchymatous	Parenchymatous	Parenchymatous	Parenchymatous	Parenchymatous	Parenchymatous
Number of collateral vascular bundles in stem	19-21	24-26	22-24	23-24	24-26	23-24	20-22

RESULTS

Stem anatomical structure: A summary of the stem anatomical characters showing differences and similarities of the seven species of family Euphorbiaceae studied is shown in Fig. 1a-g and Table 2. Results from the stem anatomical study showed that they all have similar characteristics with little variations observed in their quantitative characters, such as the number of layers of cortical collenchyma and parenchyma cells as well as in the number of observed vascular bundles. Variations were also observed in trichomes of the stem, as trichomes were found present in all species of the genus *Acalypha* and *Euphorbia* studied but was absent in species of the genus *Jatropha* and *Manihot* studied.

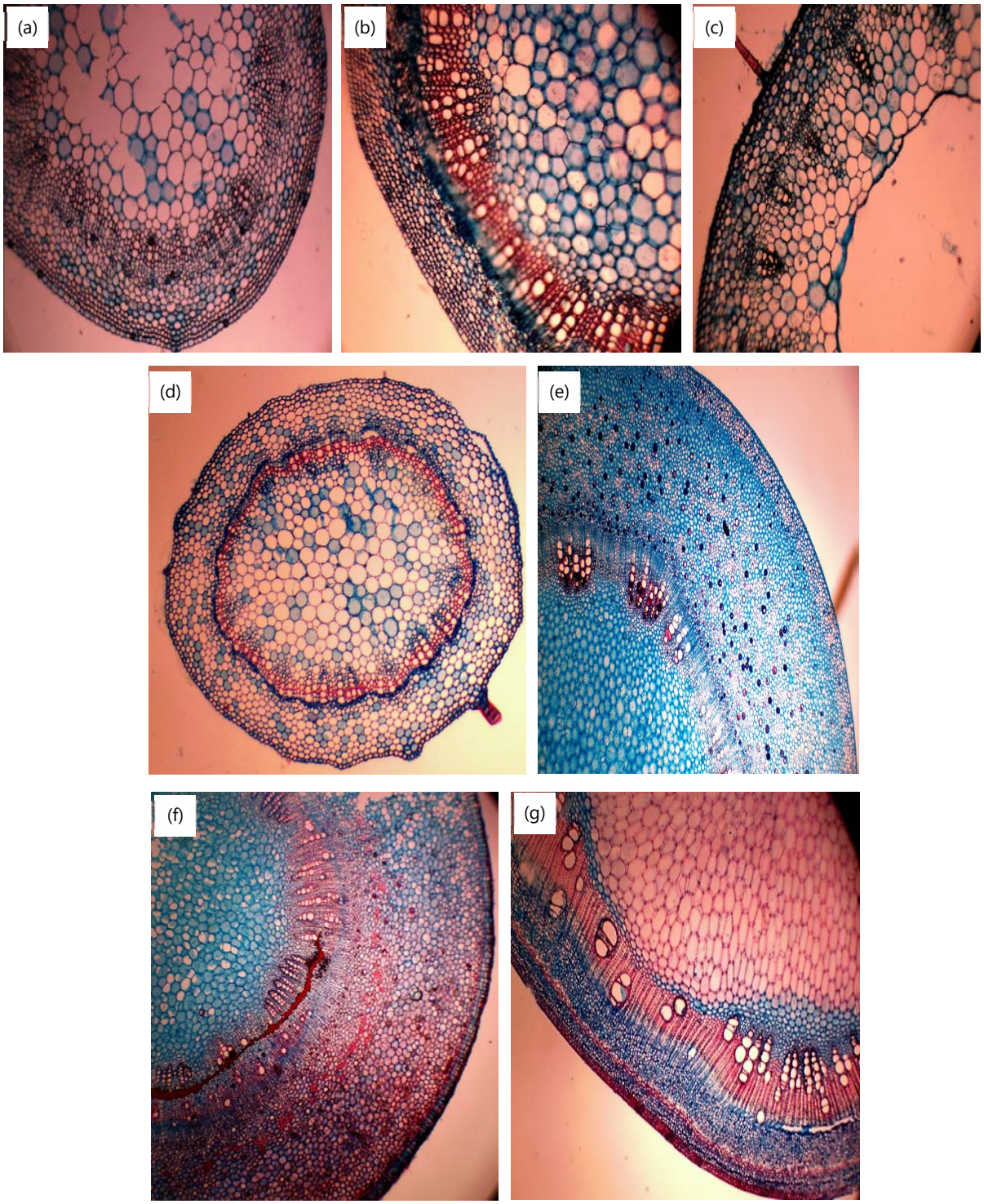

Fig. 1(a-g): Transverse sections of the stem of different varieties, (a) *A. hispida*, (b) *A. wilkesiana*, (c) *E. heterophylla*, (d) *E. hirta*, (e) *J. curcas*, (f) *M. esculentus* and (g) *J. gossypifolia*

DISCUSSION

Anatomical features of the species studied revealed the structural diversity of this family. Taxonomic uses of the anatomical characters in this family are reported elsewhere[23,24]. The most important anatomical characters from studied species are trichomes, nature of cortex and hypodermis, nature of pith, the shape of a transverse section of stem and the number of observed vascular bundles[25].

Results from the transverse section of the stem anatomical structures showed that the stem is circular or oval in outline in all species, except *E. heterophylla*, where the stem-axis is conspicuously notched. The outermost delimiting layer is the epidermis, which is generally uniseriate and composed of barrel-shaped, small or medium-sized cells. They are thick-walled, covered by a thick cuticle. Trichomes were present on the epidermis in some species such as *A. hispida*, *E. heterophylla* and *E. hirta* where there were glandular, multicellular trichomes and unicellular, non-glandular trichomes present in *A. wilkesiana*. Trichome characters are now very significant in taxonomic studies. Trichomes are useful in plant adaptations to varying ecological factors and for the prevention of herbivory[25-27].

The epidermis is followed by the hypodermis, which is usually collenchymatous. It is 3-4 layers in *A. hispida*, *E. heterophylla* and *E. hirta*, 5-6 layers in *A. wilkesiana* and *J. curcas*, while it is 2-3 layers in *J. gossypifolia* and *M. esculenta*. The hypodermis is then followed by a few to the many-layered cortex. The cells are parenchymatous, rounded or polygonal and moderately thickened. Some of the species exhibits 4-5 layered cortex (*A. hispida*, *A. wilkesiana* and *M. esculenta)*, whereas others show 5-6 layered broad cortex (*E. heterophylla* and *E. hirta)*, while there are 6-7 layers in *J. curcas* and 7-8 in *J. gossypifolia*. This is followed by the endodermis and pericycle which are uniseriate in all the species. This is closely followed by the vascular tissue that extends in stem-axes in the form of a continuous ring and is collateral and open. The constitution of vascular tissue is, however, basically similar to each other.

It is a pith that occupies the central region of the stem axis. The pith cavity is filled with thin-walled and parenchymatous cells, except in *E. heterophylla*, which has a hollow pith. Different types of crystals have been recorded in the members of Euphorbiaceae[28]. The granular matter is observed in the cortical cells in *A. hispida*, *A. wilkesiana* and *J. curcas*. All anatomical characters can be employed in the taxonomy of the family.

There were several druses in the ground parenchyma and druses are cluster crystals formed by aggregates and they include blocky, tabular, styloid and tetrahedral crystals[29].

Presently in Nigeria, there is a need for plant taxonomists to employ the system of molecular identification of every plant to establish a standard genetic library. This will help to curtail the confusion created by ambiguous identification. Moreover, it will also aid to review obsolete literature in the field of taxonomy because recent findings will eradicate wrong information on overlapping species identification, nomenclature and classification.

CONCLUSION

Many of the characteristics presented in this work corroborate the descriptions that had already been reported for other species of Euphorbiaceae. Some anatomical features can be used as diagnostic, allowing for the segregation of the species studied, such as the presence of trichomes, druses and the number of layers of hypodermis and cortex. The important microscopic features of the stem of the species studied may all serve as a useful diagnostic tool, thus, being a basis for proper authentication of the species.

SIGNIFICANCE STATEMENT

Most research works involving this taxon: Euphorbiaceae are focused mainly on the morphological and anatomical study of the leaves and roots of members of this family, with little research work on cytological and phytochemical investigation across members of the genera in the family with the sole aim of validating or invalidating existing classification. As a result of the medicinal and economical value of these species of Euphorbiaceae in Nigeria, it becomes expedient that investigative study is made with the sole aim of characterizing, identifying and validating the existing classification of these taxa to avoid adulteration during its usage for medicinal purposes. Hence, the need for this research work.

REFERENCES

1. Mwine, J.T. and P. van Damme, 2011. Why do Euphorbiaceae tick as medicinal plants? A review of Euphorbiaceae family and its medicinal features. J. Med. Plants Res., 5: 652-662.
2. Kumar, S., R. Malhotra and D. Kumar, 2010. *Euphorbia hirta*: Its chemistry, traditional and medicinal uses, and pharmacological activities. Pharmacogn. Rev., 4: 58-61.
3. Fayed, A.A., M. Soliman, A. Faried and M. Hassan, 2019. Taxonomic evaluation of *Euphorbiaceae sensu lato* with special reference to Phyllanthaceae as a new family to the flora of Egypt. Biol. Forum: Int. J., 11: 47-64.
4. Bramwell, D. and Z.I. Bramwell, 2001. Wild Flowers of the Canary Islands. 2nd Edn., Editorial Rueda SL, Madrid, Spain, ISBN: 9788472071292, Pages: 437.
5. Betancur-Galvis, L.A., G.E. Morales, J.E. Forero and J. Roldan, 2002. Cytotoxic and antiviral activities of Colombian medicinal plant extracts of the *Euphorbia* genus. Mem. Inst. Oswaldo Cruz, 97: 541-546.
6. Ennos, R.A., G.C. French and P.M. Hollingsworth, 2005. Conserving taxonomic complexity. Trends Ecol. Evol., 20: 164-168.
7. Williams, D.M., 2010. Plant taxonomy: The systematic evaluation of comparative data. Syst. Biol., 59: 608-610.
8. Johnson, D.E. and R.J. Kent, 2002. The impact of cropping on weed species composition in rice after fallow across a hydrological gradient in West Africa. Weed Res., 42: 89-99.
9. Evert, R.F., 2006. Esau's Plant Anatomy: Meristems, Cells, and Tissues of the Plant Body: Their Structure, Function, and Development. 3rd Edn., John Wiley & Sons, United States, ISBN: 9780471738435, Pages: 601.
10. Raven, P.H., R.F. Evert and S.E. Eichhorn, 2005. Biology of Plants. 7th Edn., W.H. Freeman and Company Publishers, New York, USA, Pages: 686.
11. Kumar, A. and S.K. Tewari, 2015. Origin, distribution, ethnobotany and pharmacology of *Jatropha curcas*. Res. J. Med. Plants, 9: 48-59.
12. Sultana, R.S., 2017. Anatomical characteristics of stem and leaf in *Euphorbia hirta* L. Plant, 5: 9-12.
13. Ahmad, M.P., A. Hussain, H.H. Siddiqui and S. Wahab, 2012. Macroscopical, anatomical and physico-chemical studies of *Euphorbia hirta* Linn. growing widely on Eastern Uttar Pradesh region of India. Int. J. Biomed. Adv. Res., 3: 541-545.
14. Jepsen, J.K., R.K. Henning and B. Nyathi, 2008. Generative propagation of *Jatropha curcas* L. on Kalahari sand. Jatropha J.,
15. Brundu, G. and I. Camarda, 2004. The exotic flora of chad: A first contribution. Weed Technol., 18: 1226-1231.
16. Ajuru, M.G. and B.E. Okoli, 2013. Comparative vegetative anatomy of some species of the family Cucurbitaceae Juss in Nigeria. Res. J. Bot., 8: 15-23.
17. Dayrat, B., 2005. Towards integrative taxonomy. Biol. J. Linn. Soc., 85: 407-415.
18. Awomukwu, D.A., B.L. Nyananyo, C.J. Uka and C.U. Okeke, 2015. Identification of the genus *Phyllanthus* (family Phyllanthaceae) in Southern Nigeria using comparative systematic morphological and anatomical studies of the vegetative organs. J. Plant Sci., 3: 137-149.

19. Akcin, O.E. and T.B. Ozbucak, 2006. Morphological, anatomical and ecological studies on medicinal and edible plant *Malva neglecta* Wallr. (Malvaceae). Pak. J. Biol. Sci., 9: 2716-2719.
20. Abdulrahaman, A.A., F.O. Egbedo and F.A. Oladele, 2009. Stomatal complex types, stomatal density and the stomatal index in some species of *Dioscorea*. Arch. Biol. Sci., 61: 847-851.
21. Ozimede, C.O., G.C. Obute and B.L. Nyanayo, 2019. Morphological and anatomical diversity study on three species of *Amaranthus* namley; *A. hybridus* L., *A. viridis* L. and *A. spinosusfrom* Rivers State Nigeria. J. Appl. Sci. Environ. Manage., 23: 1875-1880.
22. Gagliardi, K.B., A. Rosado, L.A. de Souza, I.S. Moscheta and A.L.M. Albiero, 2012. Structure and development of fruits and seeds of weed species of Euphorbiaceae. Acta Bot. Bras., 26: 38-45.
23. Jafari, A. and Y. Nasseh, 2009. An internal structure investigation on *Euphorbia L*. species in North-East of Iran. Asian J. Plant Sci., 8: 86-88.
24. Aworinde, D.O., D.U. Nwoye, A.A. Jayeola, A.O. Olagoke and A.A. Ogundele, 2009. Taxonomic significance of foliar epidermis in some members of euphorbiaceae family in Nigeria. Res. J. Bot., 4: 17-28.
25. Lekshmi, S., R.D. Raj, A. Ramachandran and A. Sivaprasad, 2017. Anatomical and molecular characterization of selected species of *Euphorbia* L. (euphorbiaceae) from Kerala, India. Int. J. Adv. Res., 5: 332-341.
26. Sereena, K. and T.A. Shahida, 2014. Comparative anatomical and histochemical studies of *Euphorbia hirta* L. and *Euphorbia thymifolia* L. (stem). Int. J. Pharm. Sci. Res., 6: 772-777.
27. Sultana, R.S., 2016. Stem and leaf anatomy of *Lantana camara* L.-A plant of the Verbenaceae family. Int. J. Curr. Res. Biosci. Plant Biol., 3: 27-31.
28. Onyeweaku, G.C., B.L. Nyananyo and C.O. Ozimede, 2020. Taxonomic studies on the genus *Cissus* L. (Vitaceae) present in Obio/Akpor local government area of Rivers State, Nigeria. J. Appl. Sci. Environ. Manage., 24: 139-145.
29. He, H., T.M. Bleby, E.J. Veneklaas, H. Lambers and J. Kuo, 2012. Morphologies and elemental compositions of calcium crystals in phyllodes and branchlets of *Acacia robeorum* (Leguminosae: Mimosoideae). Ann. Bot., 109: 887-896.