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Anatomical Characterization of Leaves of Some Medicinal Plants of The Family Euphorbiaceae in Nigeria

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Abstract

Medicinal plants, which are valued assets and local heritage to any country, are necessary to be collected, preserved, characterized, scientifically analyzed and used in treating different diseases of humans and animals. A lot of important medicinal plants from different parts of the world have not been indexed, characterized and tested for their medicinal values. To avoid adulteration, proper scientific characterization of specific plant parts of each is important. Members of the family Euphorbiaceae are seen as one of the top twenty-five economically important plants, existing in different variety of forms which has great ethnomedicinal value. Based on the medicinal importance of the family Euphorbiaceae, seven species were selected for study from four genera which includes: genus Acalypha – *A. hispidia* and *A. wilkesiana*, genus Euphorbia – *E. heterophylla* and *E. hirta*, genus Jatropha – *J. curcas* and *J. gossipifolia* and the genus Manihot – *M. esculenta*. The main aim of this research was to taxonomically characterize and validate the classification of selected members of the family Euphorbiaceae by looking at the anatomical features of the transverse sections of the leaves using light microscopy. Results from the leaf anatomical study showed that they all have a uniseriate epidermis with small intercellular spaces. Cortical tissue of all species studied was found to be made up of collenchyma and parenchyma cells. Variations were however observed in midrib shape, arrangement, number, nature and shape of vascular bundles as well as in the presence or absence of trichomes etc. The standardized anatomical characters assisted in the detection and diagnosis of the particular medicinal plants species.

Keywords: *Acalypha*, Anatomy, *Euphorbia*, Euphorbiaceae, *Jatropha*, *Manihot*, Medicinal plants.

Introduction

One of the factors influencing human civilization is the interaction between humans and plants [1]. Through ethnobotanical studies, there has been documentation of medicinal uses of plants and this has enable the development of contemporary drugs and treatments as well as plant conservation [2,3]. Many ethnobotanical studies around the world, report the use of herbal plants for healing process, which has been in used for several generations in their respective societies [4,5].

Traditional medicine has become an integral part of culture and plants have been used as a source of medicine from time immemorial to treat different ailments [6]. These traditional medical practices and remedies are recorded in oral tradition and in early medico-religious manuscripts and traditional pharmacopoeias, which, according to the estimates of some historians, date back to the 15th century AD [7].

The family Euphorbiaceae, also called the spurge family, is a large family of flowering plants with 300 genera and around 7,500 species. Most spurges are herbs, but some, especially in the tropics, are shrubs or trees. Some are succulent and resemble cacti. This family occurs mainly in the tropics, with the majority of the species in the Indo-Malayan region and tropical America. A large variety occurs in tropical Africa, but they are not as abundant or varied as in these two other tropical regions. However, *Euphorbia* also has many species in non-tropical areas such as the Mediterranean Basin, the Middle East, South Africa, and southern USA [8]. .

The leaves are alternate, seldom opposite, with stipules. They are mainly simple, but where compound, are always palmate, never pinnate. Stipules may be reduced to hairs, glands, or spines, or in succulent species are sometimes absent. The radially symmetrical flowers are unisexual, with the male and the female flowers usually occurring on the same plant. As can be expected from such a large family, there is a wide variety in the structure of the flowers. They can be monoecious or dioecious. The stamens (the male organs) can number from one to 10 (or even more). The female flowers are hypogynous, that is, with superior ovaries [8].

Plant anatomical studies as systematic line of evidence in Plant Taxonomy are used with other systematic lines to achieve good taxonomic decision making [9]. The reason is because anatomical characters are conserved and stable and so are employed as taxonomic characters in plant systematic and taxonomic studies [10]. The anatomical character, which includes root anatomy, trichomes, stem anatomy, stomata and epidermal, wood anatomy, nodal anatomy, sclereids and fibres, cambium and leaf anatomy are adopted in Biosystematics and taxonomic studies to identify plants, establish genetic relationships and solve taxonomic disputes [10]. 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 in nature, hence limiting their availability for study and proper identification [11]. Currently, there are calls for some novel protocol for proper and easy identification of plant at any season of the year [12]. Plant anatomical studies of the vegetative organs like leaves, root and stem which may be found at all seasons seems to be a solution such to challenges [12]. Hence the adoption of plant anatomy in this research studies.

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 much regions of Nigeria and West Africa. As a result of the economical value of these species of Euphorbiaceae in Nigeria, it becomes expedient that investigative study be made using anatomical line of evidence with the sole aim of characterizing, identifying and validation of existing classification of these taxa.

Materials and Methods

Collection and Identification of Plant Materials

Fresh and healthy samples of selected plant species from four different genera in the family Euphorbiaceae were collected in separate bags from Rivers, Delta, Abia, Bayelsa and Imo States all from the Niger Delta of Nigeria. The various useful parts like leaves were separated and preserved for the study. Identification of various plants species done by comparing with authenticated herbarium specimens, later confirmed with the help of diagnostic keys and morphological description given in various floras. Identified pressed plant samples were deposited at the University of Port Harcourt (UPH) and Rivers State University (RSU) Herbarium for reference and further studies.

Anatomical study of part used

The leaves of all the seven species studied were fixed in F.A.A. (e. Formalin acetic acid-alcohol, 1:1:18) after trimming them to correct dimensions. Hand sections of fresh leaves were cut using a sharp blade. Thin transverse sections were stained in safranin and then fast green, passed through alcohol grades for dehydration, and then mounted in D.P.X. Observations were taken from these sections using light microscope. These sections were also photomicrographed using XSZ-N107 Microscope with (MA88-900) camera. Special identifying features of the plant part(s) were studied and identified.

Results

Midrib Anatomy

A summary of the observable midrib anatomical characters showing differences and similarities among the seven species of the family Euphorbiaceae studied is shown in Plate l and Table 1**.**

The results showed that all the species have a uniseriate upper and lower epidermis, covered by waxy cuticle. Trichomes were found on the epidermis of *E. hirta* only. Immediately below the epidermal cells are collenchyma cells, then parenchyma cells in varying number of layers. The central portion of the midrib is occupied by well developed crescent shaped vascular bundles, composed of the xylem and phloem tissues. The xylem is positioned towards the lower or abaxial side of the midrib, while phloem is found towards the adaxial surface. There is great variation in the number of vascular bundles in the different species studied. Also, the vascular bundles lack cambium.

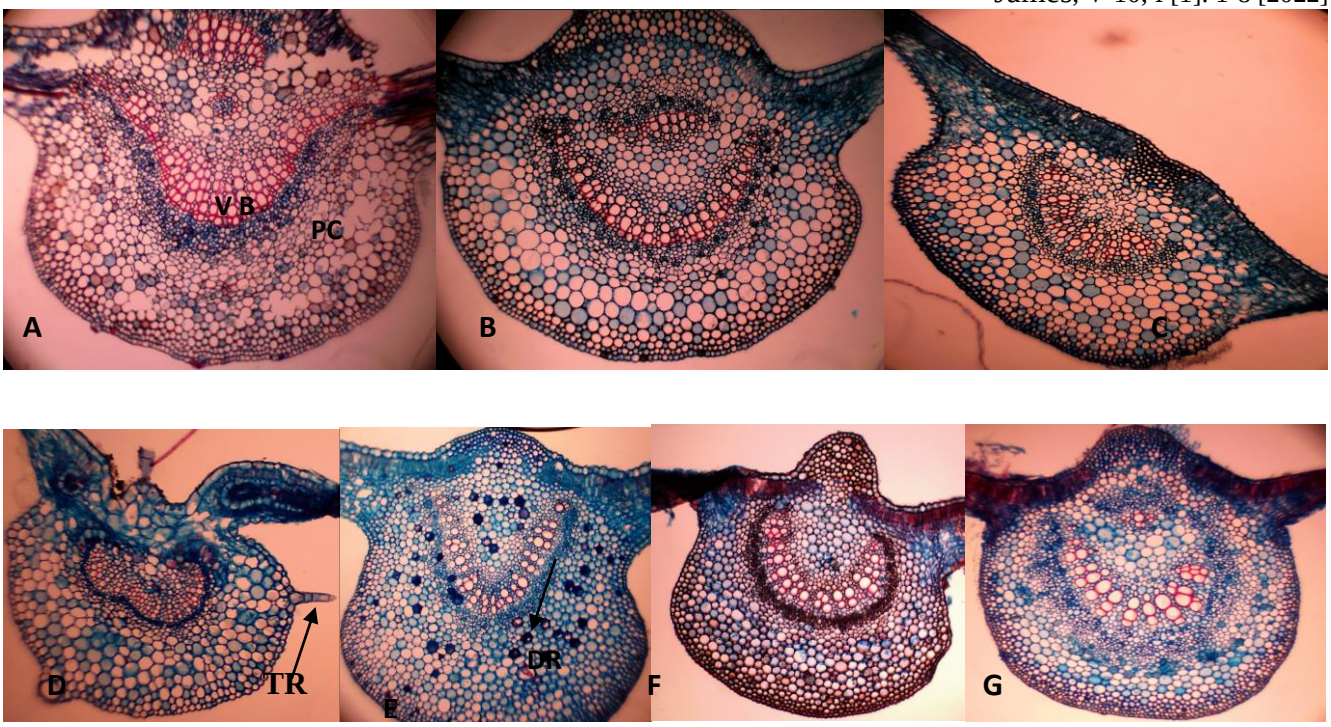

Plate 1. Transverse section of the Midrib of A- *A. hispidia*, B- *A. wilkisiena*, C- *E. heterophylla*, D- *E. hirta*, E- *J. curcas*, F- *Jatropha gossypifolia* and G- *Manihot esculenta*; VB – Vascular bundle; PC- Parenchyma cells; DR- Druses; TR- Trichomes (X40)

Table 1: Midrib and Lateral Anatomical Characters of the Species of Euphorbiaceae Studied

S/N	Character	*A. hispida*	*A. wilkesiana*	*E. heterophylla*	*E. hirta*	*J. curcas*	*J. gossypifolia*	*M. esculenta*
1	Epidermis	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate	Uniseriate
2	Collenchyma cells	4-5 layers	4-5 layers	3-4 layers	3-4 layers	3-4 layers	3-4 layers	3-4 layers
3	Parenchyma cells	7-8 layers	6-7 layers	6-7 layers	7-8 layers	7-8 layers	8-9 layers	6-7 layers
4	Number of Vascular Bundles	2 (Big and small)	2 (Big and small)	2 (Big and small)	1 (Big)	1 (Big)	1 (Big)	2 (Big and small)
5	Shape of Vascular bundles	Crescent	Crescent	Crescent	Crescent	Crescent	Crescent	Crescent
6	Druses and tannins in midrib	Present	Absent	Absent	Absent	Present	Present	Present
7	Midrib shape in Transverse section	Arc	Arc	Rounded	Rounded	Rounded	Arc	Crescent
8	Trichomes in midrib	Absent	Absent	Absent	Present	Absent	Absent	Absent
9	Adaxial Protrusion of midrib	Absent	Absent	Absent	Absent	Present	Present	Present
10	Shape and Nature of Palisade mesophyll	Cylindrical without intercellular spaces	Cylindrical without intercellular spaces	Cylindrical without intercellular spaces	Cylindrical without intercellular spaces	Cylindrical without intercellular spaces	Cylindrical without intercellular spaces	Cylindrical without intercellular spaces
11	Shape and Nature of spongy mesophyll	Rounded with intercellular spaces	Rounded with intercellular spaces	Rounded with intercellular spaces	Rounded with intercellular spaces	Rounded with intercellular spaces	Rounded with intercellular spaces	Rounded with intercellular spaces

Discussion

Some anatomical traits are very diagnostic. Thus, they are frequently used in routine identification. Since the leaf is regarded as the most varied organ of the angiosperms, taxonomic studies of various taxa were carried out on the basis of leaf anatomy [13-17]. These studies present many anatomical characteristics of potential taxonomic significance [15].

Comparison of the anatomic features of the seven species in this research was made according to the "Anatomy of the Dicotyledones"[18]. Anatomical characters of species were observed to have been similar to usual features of Euphorbiaceae family. It was determined that the most important anatomical differences among the seven species studied were in the layers of collenchyma, parenchyma, number of vascular bundles, cell inclusions, trichomes and adaxial protrusion of midrib.

The observation of the transverse section of the leaves of the species studied showed bilateral symmetry. The epidermal cells which are protective cells against physical aggressions and pathogens were thick-walled and tightly packed in a single layer. This thickening is as a result of the deposit of a waxy substance secreted by the epidermis called cuticle. The characters of leaf epidermal surfaces in resolving the taxonomy of taxa has obtained numerous confession for a long time [19-25].

Collenchyma cells were 4-5 layers in *A. hispida* and *A. wilkesiana*, while the cells are 3-4 layers in *E. hirta, E. heterophylla, J. curcas, J. gossypifolia*, and *M. esculenta*. Collenchyma cells are found just beneath the epidermis and generally they are elongated and their walls are pliable in addition to being strong. Collenchyma is the primary supporting tissue in stems, leaves and floral parts of dicotyledons. The design and function is to build and maintain the special unevenly thick primary cell wall. The cells are also typically quite elongate. The role of this cell type is to support the plant in areas still growing in length. The primary wall lacks lignin that would make it brittle, so this cell type provides what could be called plastic support. Stretchable support (without elastic snap-back) is a good way to describe what collenchyma does.

The layers of parenchyma cells also varied. Parenchyma cells are 7-8 layers in *A. hispida, E. hirta* and *J. curcas,* but 6-7 layers in *A. wilkesiana, E. heterophylla* and *M. esculenta. J. gossypifolia* has 8-9 layers of parenchyma cells. Apart from the xylem and phloem in their vascular bundles, leaves are composed mainly of parenchyma cells. Some parenchyma cells, as in the epidermis, are specialized for light penetration and focusing or regulation of gas exchange, but others are among the least specialized cells in plant tissue, and may remain totipotent, capable of dividing to produce new populations of undifferentiated cells, throughout their lives.

Vascular system were represented by two vascular bundles in the mid rib region of *A. hispida, A. wilkesiana, E. heterophylla* and *M. esculenta*; the upper one is bigger than the lower kidney shaped one The number of vascular bundles were one each in *E. hirta, J. curcas* and *J. gossypifolia.* On the ventral side, the xylem allowing the transport of water and mineral salts; it consists of conductive vessels. But, on the dorsal side, the sieve tubes ensure the circulation of prepared food. The companion cells provide an energetic role and the parenchymatous cells serve for the storage and the mobilization of the reserves.

The Plate 2 showed cells of palisade parenchyma and spongy parenchyma. The palisade parenchyma cells are elongated and oriented perpendicular to the leaf surface, and are the site of photosynthesis. While, spongy parenchyma cells are composed of irregularly shaped cells and large intercellular spaces, and mainly function as the site of gaseous exchange.

Cells containing tannin were present very common in the unlignified tissues of *A. hispida, J. curcas, J. gossypifolia* and *M. esculenta* investigated [18,26]. [27] stated that tanniniferous idioblasts and druses occur in many families including Euphorbiaceae. Druses crystals were very common in *A. hispida, J. curcas, J. gossypifolia* and *M. esculenta* also, in the mesophyll and cortex of the leaves. According to [27], the presence and location of crystals may be distinctive and useful in taxonomic classification. In this study, druse crystals were found in the mesophyll and also in the midrib region.

Trichomes are very diverse in Euphorbiaceae species, such as unicellular glandular or non-glandular and stinging types [18,28]. Multicellular uniseriate trichomes with pointed apices were found on both surfaces of *Euphorbia hirta* [20]. Many studies have revealed the taxonomic value of trichomes in angiosperms [29-31].

Conclusions

The anatomically study of the structure of midrib, are taxonomically and diagnostically significant at the species level. Some differences were determined in the anatomical properties of all studied taxa. Some anatomical characteristics of the leaves were found to be of diagnostic importance, such as the presence of trichomes, tannins, druses, layers of collenchyma, parenchyma, number of vascular bundles and adaxial protrusion of the midrib. All of these characteristics are environmentally influenced, and future studies analyzing plants from several localities are needed; nevertheless, they can be very useful in the delimitation of species.

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