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ANATOMICAL STUDIES OF LEAF AND STEM OF *EUPHORBIA GRAMINEA (JACQ)* (Euphorbiaceae)

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ABSTRACT

Euphorbia graminea jacq. belongs to the family, Euphorbiaceae. The present investigation was carried out to determine anatomical features of the leaf and stem of Euphorbia graminea.-Fresh leaf and stem of E. graminea were collected at Forestry Research Institute of Nigeria Ibadan. Fresh plant specimens were used for this species. Sample of leaf was macerated in concentrated Trioxonitrate(v) Acid for 2-4 hours. The sample was transferred to water in Petri-dish while the abaxial and adaxial epidermis, the sample were carefully separated using forceps and dissecting needle .The use of light microscope revealed that stomata only occurred on the lower surface of the plant. It also revealed the presence of trichomes on the lower surface of the leaf. The Transverse sections of the stem revealed the structural arrangements in the stem,the root hair, the epidermis, pholem, the xylem, and anticlinical cuticles. The characters observed in E. graminea are diagnostic enough to separate the species and distinguish even when they are sterile and in fragmentary condition. The anticlinical cell walls such as undulate pattern are present.

Keywords: Anatomy, *E. graminea*, Anticlinal cuticles, stomata, Epidermis.

INTRODUCTION

Euphorbiaceae, the spurge family, one of the majorflowering plant families: with 334 genera grouped in 52 tribes and 5 subfamilies, is considered as the sixthlargest family of Angiospermae. The family uphorbiaceae in Nigeria includes about 30 species of the genus *Euphorbia* (Huttchinson and Dalziel,1958). *E. graminea* is a native to northern Mexico, Colombia, and venezula (Yang *et.al.* 2005). *E. graminea* is very morphologically variable, as discussed by McVaugh (1993). It is an annual plant, generally from 15–30 cm tall, sometimes taller, often branching from the base, and dichotomously branched distally. The leaves are alternate; those toward the base of the plant are ovate or oblong with a few large distantly-spaced teeth, while those toward the tops of the plants are elliptical to linear and entire. *E. graminea* is the only grass in the family, Euphorbiaceae. *E. graminea* produces cymes of light-green cyathiae, The University of Ibadan herbarium collections suggest that *E. graminea* weed was present in Nigeria as early as 1993 but only became widely noticeable in the mid 2000s. This thus suggests that the weed was probably introduced into Nigeria in the early 1990s. Among the possible anthropogenic uses of *E. graminea* include treatment of skin infection such as ulcers, cancers, tumor, and other diseases as practiced by Colombian traditional healers (Betancur-Galvis *et .al.* 2002). Alonso-Amelot *et al. (*2004) also reported *E. graminea* to be poisonous to ruminants.

Anatomy or the internal form and structure of plant organs like leaves, stems and nodes is a classical source of data used in plant taxonomy. Anatomical data are often useful in solving problems of relationships because they can suggest with greater confidence, the homologies of morphological character states as well as help in the interpretation of evolutionary directionality (polarity). According to Stace 1984 foliar epidermis is one of the most noteworthy taxonomic characters from the bio-systematic point of view. Therefore, taxonomic studies on Euphorbiaceae have been made based on leaf epidermises. It is sometime held that comparative anatomy has outlived its usefulness and that it is a sterile discipline, bereft of new advances (Stuessy, 2009). This study is undertaken to provide a vivid

anatomical description that will help the taxonomical characterization and the identification of the plant even in fragmentary conditions.

MATERIALS AND METHODS

Sample Collection

Fresh specimens of *E. gramine* used in this study were collected from Forestry Research Institute of Nigeria, Ibadan, Oyo state, where the species are well distributed.

Micro Morphological Study

Epidermal preparation were obtained using the technique of Ugbogu *et. al.* (2016). Fresh plant specimens were used for this study. Sample of leaf was macerated in concentrated Trioxonitrate(v) Acid for 2-4 hours. The sample was transferred to water in Petri-dish while the abaxial and adaxial epidermis, the sample were carefully separated using forceps and dissecting needle. The inner parts (mesophyll tissue) were carefully cleared with Carmel brush; isolated epidermal layers were washed in several changes of water before transferring in 50% alcohol for 1-2 minutes to harden them. The sample was transferred to clear glass microscopic slide and stained (after draining the excess water) with Safranin O for less than 4 minutes and excess stained was washed off using a dropping pipette to add and remove water from tissue. They were later mounted on slide and covered with cover -slip and lady's paint was used to seal to avoid air from entering into it. It was mounted under light microscope and viewed x 10 and x 40 objective lens. The stems of the plant were cut transversely in order to get thin section of the stems. Each of the stems was cut with a safety blade into various Petri dishes which were filled with water. The sectioned stem of the plant were placed on separate slides, containing glycerol droplet. Few drops of Safranin were applied and covered with cover slips which were later mounted under light microscope for examination and viewed under x 10 and x 40 objective lens.

RESULTS

The anatomical section of leaf of *E. graminea* is shown below in plates 1 and 2. On the occurrence of stomata on the leaf surface, the species is said to be hypostomatic (i.e. stomata occurring on abaxial surface only).The leaf has one stomata type which is anisoyctic Qualititative foliar epidermis is shown in table1 below. Anatomical description of the stem showed that the epidermis, cortex and vascular bundle were clearly differentiated (plates 4 and 5.). *Euphorbia graminea* has characteristics of a dicots plant which means *E.graminea* is not a grass.

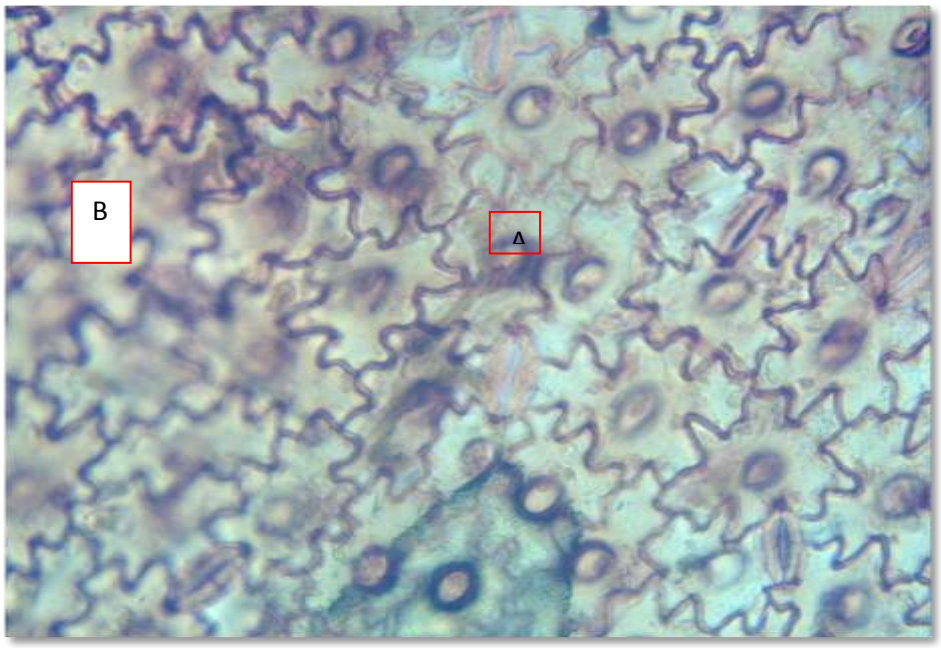

Plate1 a: Abaxial surface of *E. graminea* leaf showing Anisocytic Stomata, Undulate Anticlinical Walls. A-Stomata B-Epidermal cell

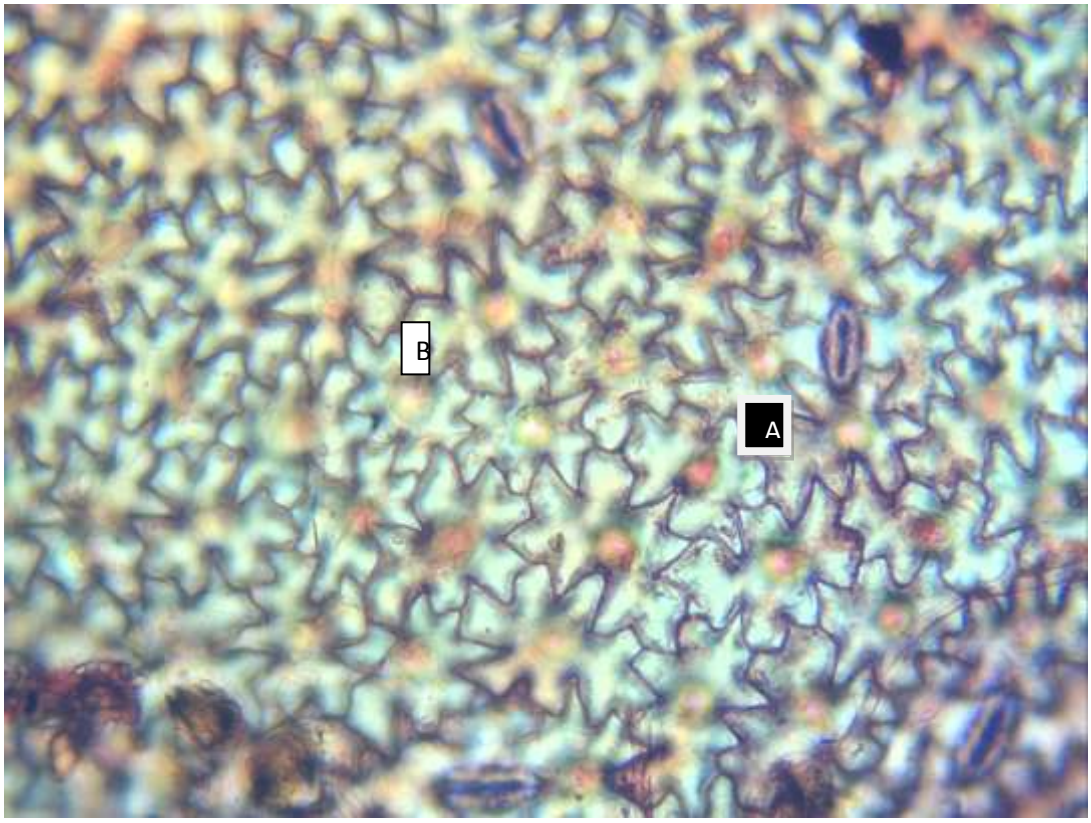

Plate1b: Abaxial Surface of *E. graminea* Leaf

Plate 2: Adaxial Surface of *E. graminea* with Trichomes

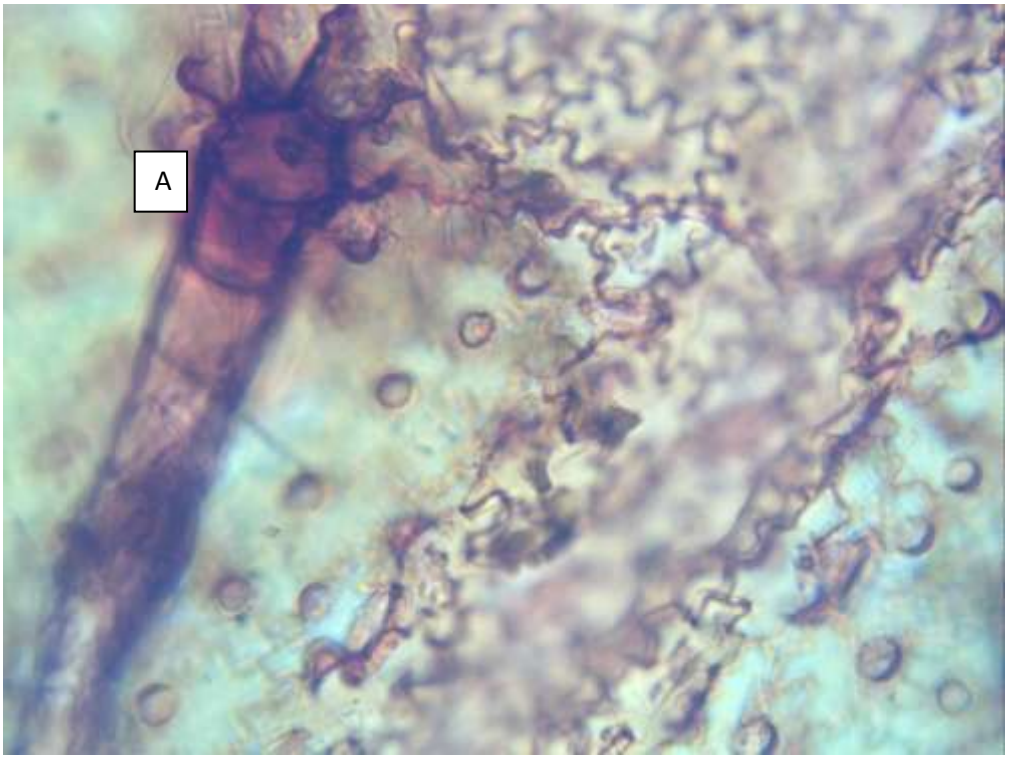

Plate 3: Adaxial of *E. graminea* leaf with Trichome Base.

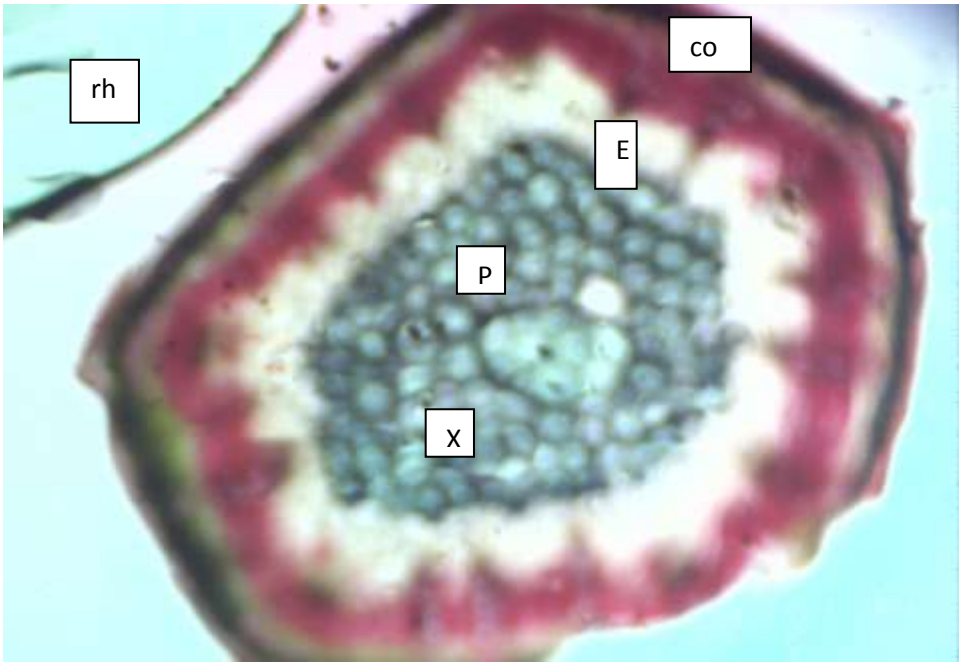

Plate 4: Transverse section of *E. graminea* stem.
Rh: root hair, C:cortex, E:epidermis, P:phloem, X:xylem.

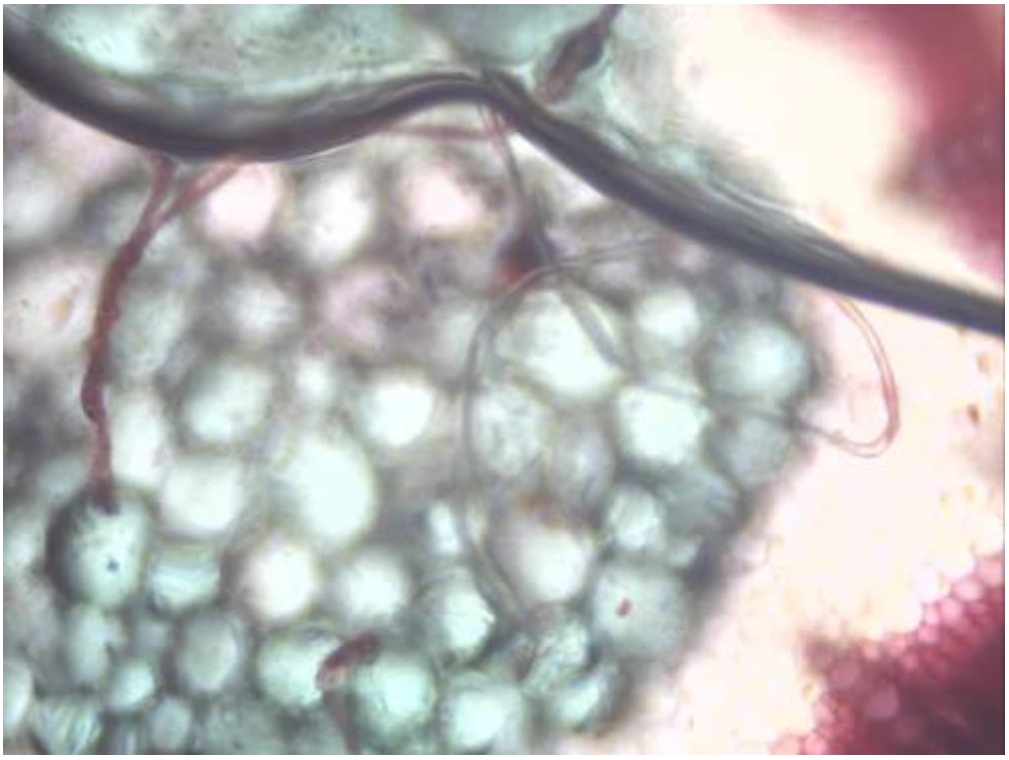

Plate 5: Transverse section of *E. graminea* Stem.

Table 1: Qualitative and Quantitative Foliar Epidermal Eeatures of *E. graminea* Leaf.

S/No.	Basis for comparison	Monocots	Dicots
1	Meaning	Plants with the seed having only one cotyledon are called as monocots, and the plant is called as monocotyledons	Plants with the seed having two cotyledons are called as dicots and plant is called as dicotyledons.
2	Flower	The flower parts are present in multiples of three.	The flower parts are present in multiples of four or five.
3	Leaves	Monocots have stomata on both upper as well as on lower surface of their leaves and so-called as amphistomatous. The venation of the leaf is parallel.	Dicots have stomata only on one surface of their leaves and so-called as hypostomatic . There is the net-like or intersecting type of venation present in the leaf
4	Stem	Vascular bundles in stems are scattered throughout.	Vascular bundles in stems are arranged in a ring-like pattern.

Table 2: Comparative Morphological information to confirm *E. graminea* as a dicot and not a monocot plant

Species Name	Stomata	Stomata Type	Stomata Length (µm)	Stomata Width (µm)	Number of stomata per view	Cell Wall Type	C ell Wall Length (µm)	Cell Wall Width (µm)	Number of cell wall per view	Trichomes
Euphorbia graminea Abaxial	Present	Anisocytic	170.2 2.34±0.74	11.8 2.18±0.69	50 1.42±0.45	Undulate	34.9 7.62±2.41	24.3 4.13±1.32	12.5 4.01±1.27	Absent
Euphorbia graminea Adaxial	Absent	_	_	_	_	Undulate	22.1 5.25±1.66	26.6 6.61±2.09	16.2 6.39±2.03	Present

DISCUSSION

In this study one type of stomata was anatomically identified on the abaxial surface of *E. graminea* ,while there was no stoma on the adaxial surface. Hence, the leaves of *E graminea* are hypostomatic. According to Esau 1965 , stomata found on the lower part of a plant is hypostomatic. Anisoyctic stomata was found, this is in line with the study of other leguminosae family by kathiresan *et al.*, 2011 who reported five species of Bauhinia with anisocytic stomata types as a prominent one in all the species. The cell wall type in *E. graminea* is undulate ,according to ayodele *et al.*, 1997 straight or curve walls ae characteristics of species growing in drier conditions while undulate walls are found mostly in species growing in area of high humidity.

Trichome features are now considered important in taxonomic studies (Leelavathi and Ramayya, 1983). Trichomes observed on the adaxial surface of *E. graminea*, are non-glandular, large in size, but absent on the abaxial surface. Trichomes are use-ful in plant adaptations to varying ecological factors and for prevention of herbivory (Stace 1965; Inam-dar and Gangadhara 1977; Heywood and Moore 1978; Jones and Luchsinger 1986). However, the trichomes bases suggest degeneration from plant surface as it is reported by Theobald *et al*., (1979). Trichomes type has been found to be of little diagnostic value in this species.

From table 1 the study of stomata characters also reveals variations in mean stomata length and width on the abaxial surface. The mean stomata length vary from 2.34±0.74; then the stomata width vary from 2.18±0.69 in the abaxial surface, while stomata was absent in the adaxial. Also as shown in Plate1a, the stomata guard cells found on both abaxial surfaces of the leaf are essential to keep the water inside the cell intact. However, they may also allow the gaseous exchange essential for photosynthetic activity (Abdulrahaman *et al*., 2009). Tabel2 shows that *E. graminea* has characteristics of a dicot plant which means *E.graminea* is not a grass.

From plate 4 and 5, the anatomy of the stem shows Collateral and open vascular bundles in the stem. Stem cells exhibited variable sizes and starch grains. Epidermis, cortex, and vascular bundle were clearly differentiated. Vascular bundles were found to be irregularly-scattered in the ground tissue with phloem and xylem units. Thus, the observed anatomical description for the stem of *E. graminea* may be a unique trait for members of the *Euphorbiaceae* family.

CONCLUSION

The characters observed in *E. graminea* are diagnostic enough to separate the species and distinguish even when they are sterile and in fragmentary condition. The anticlinal cell walls such as undulate pattern are present. The important microscopic features of the leaf and stem of *E. graminea* such as anisocytic stomata types especially in the leaf and may all serve as useful diagnostic tool. Thus, improving or being a basis for proper and authentication of *E. graminea.*

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