

# Comparative Foliar and Petiole Anatomy of Some Members of the Genus *Dieffenbachia* Schott in the Family Araceae

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## Abstract

This study provides detailed information of the anatomical attributes of the epidermis and the three regions of the petiole of four members of the genus *Dieffenbachia* Schott. Fresh samples of the leaves of *Dieffenbachia picta* Schott, *Dieffenbachia oerstedii* Schott, *Dieffenbachia senguine* (Jacq) Schott and *Dieffenbachia senguine* cultivar 'Candida' Schott were used. Epidermal peels and transverse sections were made following standard procedures. Generic characters revealed uniform epidermal cell shape on the abaxial surface, wavy to undulating and straight to wavy anticlinal wall patterns on the adaxial and abaxial surfaces respectively, brachyparacytic stomata types, round abaxial petiole outline, the presence of raphides and druses in the petioles of all the taxa. Delimiting features include irregular epidermal cell shape on the adaxial surfaces of *D. senguine* and *D. senguine* cv. 'Candida', additional anomocytic stomata types on the abaxial surfaces of *D. picta*, *D. oerstedii* and adaxial surface of *D. senguine*, cuticular striations on the abaxial surfaces of *D. oerstedii* and *D. senguine* cv. 'Candida', druses and raphide bundle on the epidermal surface of *D. senguine* only, flat adaxial petiole outline and slightly concave adaxial petiole outline in the proximal and median regions of *D. senguine* cv. 'Candida' and the presence of lamellar collenchyma cells in the petiole of *D. picta*. Data for both quantitative and qualitative characters were subjected to Principal Components Analysis and Single Linkage Cluster Analysis. Interestingly, anomocytic stomata complex, cuticular striations, raphide bundles and druses and the adaxial petiole outline separated *D. senguine* and *D. senguine* cv. 'Candida'.

**Keywords:** anatomy, anticlinal wall pattern, brachyparacytic stomata, cuticular striations, *Dieffenbachia*, epidermis, petiole

## Introduction

*Dieffenbachia*, commonly known as dumb cane, is the most poisonous genus in the family Araceae, also known as the aroid family (Bown, 2000). The genus *Dieffenbachia* Schott, tribe Dieffenbachieae, is a small genus of tropical plants that are tall, reaching a height of 6 feet (180 cm) or more and they display fancy, large, more or less variegated leaves. The stems are fleshy, with conspicuous joints. They may be green or somewhat striped with white. The large pointed leaves grow from a central stem, or cane. As the plant grows, loses its lower leaves, receiving a palm-like look (Gary, 2009). Dieffenbachias come in several varieties, one of which is all green (*Dieffenbachia oerstedii*). *Dieffenbachia picta* is the most common species with deep green leaves and white splotches. *Dieffenbachia picta* and *D. senguine* (native to the West Indies), have yielded colourful varieties of horticultural interest which are now widely dispersed throughout the tropics and as house plants in temperate countries (Simson and Ogorzaly, 1995; Johnson, 1999).

The different parts of the *Dieffenbachia* plants contain needle-shaped calcium oxalate crystals called raphides (Franceschi and Nakata, 2005). If a leaf is chewed, these crystals cause a burning sensation in the mouth and throat; swelling can occur along with a temporary inability to speak, hence, the name, dumb cane. Chewing could result in death if swelling of the throat blocks the airway (Gardner, 1994). Slaves, in ancient times, were sometimes punished by having *Dieffenbachia* put into their mouths. Young children (at the age where they regularly put things into their mouths) are at risk of suffocation and death if they eat or chew *Dieffenbachia* leaves (Barnes and Fox, 1955). In spite of its toxicity, *Dieffenbachia* has been a very popular ornamental plant for many years.

Studies of *Dieffenbachia* demonstrated that proteolytic enzymes, as well as other compounds, are responsible for the severe irritation caused by this plant and this has resulted in it being included in many lists of poisonous plants (Perkins and Payne, 1978; Lampe and McCann, 1985; Mulligan and Munro, 1990). The antimicrobial assays of the leaves and stems of *Dieffenbachia picta* have been carried out by Hewitt and

Vincent (1989) and Koneman *et al.* (1997). The genetic relatedness of 42 cultivars of *Dieffenbachia* was studied using Amplified Fragment Length Polymorphism (AFLP) markers (Chen *et al.*, 2004). Colourless essential oils from fresh leaves and stems of *Dieffenbachia picta* have been analysed by gas chromatography (Oloyede *et al.*, 2011). The chromosome number of most *Dieffenbachia* species is  $2n = 34$  (Jones, 1957). Mayo *et al.* (1997) reported the ploidy state of *Dieffenbachia* with chromosome number  $2n = 34, 68$ .

Green and Oguzor (2009) did some anatomical studies on the epidermis of *Dieffenbachia picta* only, in Southeastern part of Nigeria. This paper therefore aims at filling the knowledge gap in the foliar epidermal and petiole anatomy of selected members of genus *Dieffenbachia* species. Detailed description of the anatomy of the epidermis and the three regions of the petiole of the selected taxa in the genus are provided.

## Materials and Methods

### Materials

Four members of genus *Dieffenbachia* were used in this research work. They were *Dieffenbachia picta* Schott, *Dieffenbachia oerstedii* Schott, *Dieffenbachia senguine* (Jacq) Schott and *Dieffenbachia senguine* cultivar 'Candida' Schott. They were collected fresh from the Southwestern part of Nigeria.

### Epidermal studies

The scrape technique of Metcalfe (1960) was used in getting the epidermal peels of both the adaxial and abaxial surfaces of the leaves. The epidermal peels were then stained in 1% aqueous Safranin O and mounted in dilute glycerin for microscopic examination. Photomicrographs of the epidermis were taken for both the adaxial and the abaxial surfaces.

Epidermal cell shape, anticlinal cell wall pattern and stomata types were studied. Epidermal cell area and stomata size were calculated by multiplying the length and width of the epidermal cells and the stomata respectively. Also calculated was the Stomata Index (S.I) for the two surfaces using the formula below:

$$S.I = \frac{S}{S + E} \times 100$$

Where S.I = Stomata Index, S = Number of stomata, E = Number of ordinary epidermal cells plus the subsidiary cells in the same unit area.

### Petiole anatomy

The petiole anatomy of the studied taxa was carried out by cutting transverse section of the petioles of each of the taxa at the proximal, median and distal regions. The transverse sections were cut with the aid of a Reichert sliding microtome at a thickness of 8-15  $\mu\text{m}$ . The sections were stained with Safranin O and counter-stained in Alcian blue, after which they were made to pass through different concentrations of ethanol for differentiation and dehydration. The sections were then mounted in 25% dilute glycerin solution for examination under the microscope. Anatomical descriptions were made according to Fahn (1977) and Adedeji (2004). Photomicrographs of the different sections were made.

### Measurements and data analysis

All microscopic measurements were taken with the aid of an ocular micrometer inserted into the eyepiece of a microscope. These measurements were later multiplied by the ocular constant with respect to the power under which they were taken in order to convert them to micrometer. Qualitative data, which include adaxial epidermal cell shape, adaxial and abaxial stomata shape and type, presence or absence of cuticular striations, druses and raphide bundles on the epidermis, proximal and median petiole outline, type of collenchyma cells as well as presence or absence of starch grains were coded, merged with the quantitative data and subjected to the Principal Components Analysis and Single Linkage Cluster Analysis using Paleontological Statistics software package (PAST).

## Results and Discussion

### Anatomy of the epidermis

The summary of the foliar epidermal features of the adaxial and abaxial surfaces of the studied *Dieffenbachia* taxa is shown on Table 1 while the mean values of the epidermal cell area, stomata area and stomata index on both the adaxial and abaxial surfaces are shown on Table 2. The adaxial and abaxial epidermal surfaces of all the studied taxa were presented in Fig.1. A close observation of the epidermal cells shape reveals that polygonal to irregular epidermal cell shape is common to *D. picta* and *D. oerstedii* on their adaxial surface while *D. senguine* and *D. senguine* cv. 'Candida' have the irregular shape only, thus, adaxial leaf epidermal cell shape can be employed in delimiting the species of *Dieffenbachia*. However, all the taxa have polygonal to irregular epidermal cell shape in common on their abaxial surfaces.

On the adaxial surface, epidermal cell area was of the highest value in *D. senguine* with a minimum value of 1224.0  $\mu\text{m}^2$ , maximum value of 2519.4  $\mu\text{m}^2$  and mean value of 1583.86  $\mu\text{m}^2$ . The least value was in *D. picta*, ranging between 642.6 to 1105.0  $\mu\text{m}^2$  and mean value of 876.93  $\mu\text{m}^2$ . On the abaxial surface, *D. picta* still has the least epidermal cell area ranging from 520.2  $\mu\text{m}^2$  to 1285.2  $\mu\text{m}^2$  with mean value of 891.07  $\mu\text{m}^2$ , while the cultivar, *D. senguine* cv. 'Candida' has the highest value ranging from 918.0 to 3155.2  $\mu\text{m}^2$ . Different plant species have been separated based on the epidermal cell shape and the epidermal cell area (Oladipo and Ayo-Ayinde, 2014; Arogundade and Adedeji, 2016).

Anticlinal cell wall pattern is a generic character among the studied taxa of *Dieffenbachia*. On the adaxial surface, it is wavy to undulating in all the taxa and straight to wavy on their abaxial surfaces, so the taxa in this study cannot be separated based on their anticlinal wall pattern. In some other studies, anticlinal wall pattern has been successfully employed in separating different plant species (Adedeji, 2004; Osuji and Nwala, 2015; Arogundade and Adedeji, 2016).

The leaves of all the studied taxa that are amphistomatous with the stomata restricted to the non-venous regions. Brachyparacytic stomata complex type with elliptic shaped stomata is found on both the adaxial and the abaxial epidermal surfaces of all the studied taxa. The presence of brachyparacytic stomata is a characteristic of the family Araceae as reported by Osuji and Nwala (2015). Although, elliptic shaped stomata are common to all the taxa studied, some of the taxa have

additional circular shaped stomata. Anomocytic stomata type, an additional stomata complex type was present on the adaxial surfaces of *D. senguine* and *D. picta* as well as the abaxial surface of and *D. oerstedii* (Table 1), thus separating *D. senguine* and *D. picta* from the other taxa on the adaxial surface and *D. oerstedii* from *D. picta*, *D. senguine* and the cultivar on the abaxial surface.

Generally, there were more stomata on the abaxial surfaces than on the adaxial surfaces in all the studied taxa. The highest stomata size was encountered on the adaxial and abaxial surfaces of *D. senguine* cv. 'Candida' ranging from 598.4  $\mu\text{m}^2$  to 856.  $\mu\text{m}^2$ , with a mean value of 700.26  $\mu\text{m}^2$  on the adaxial surface and 652.8  $\mu\text{m}^2$  to 904.4  $\mu\text{m}^2$  with a mean value of 802.26  $\mu\text{m}^2$  on the abaxial surface. The lowest stomata size was encountered in *D. picta*. The value ranges from 397.8  $\mu\text{m}^2$  to 612.0  $\mu\text{m}^2$  with a mean value of 520.74  $\mu\text{m}^2$  on the adaxial surface and 442.0  $\mu\text{m}^2$  to 598.0  $\mu\text{m}^2$  with a mean value of 530.94  $\mu\text{m}^2$  on the abaxial surface. This represents another basis of separating the taxa of *Dieffenbachia* in this study. Stomata index was highest on the adaxial surface of *D. picta* with values ranging from 3.13- 7.14%, mean value of 4.95% and the abaxial surface of *D. senguine* cv. 'Candida' with values ranging from 6.67-13.95%; mean value of 10.32% (Table 2).

Ridges or folds of the cuticle establish ornamentations on the epidermal surfaces; these ornamentations largely consist of striae, hence, striated cuticle (Metcalf and Chalk, 1979). According to Solereder (1908), these striations are useful for specific diagnosis and are not always developed in the same way on the two surfaces of the leaf, they are also taxonomically stable. Adedeji and Illoh (2004) used cuticular striations to separate some species of *Hibiscus*. In this study, striated cuticle was found on the abaxial surfaces of *D. oerstedii* and *D. senguine* cv. 'Candida'. The striations were radiating from the guard cells

in the two taxa. Druses of calcium oxalate crystals are present on the adaxial and abaxial surfaces of the epidermis of *D. senguine*. Raphide bundle is also present on the abaxial surface of *D. senguine*, thus, making this species different from the other members of *Dieffenbachia* in this study.

#### Petiole anatomy

The result of the petiole anatomy was shown on Figs. 2 – 13, and provides a wide range of characters with diagnostic value in delimiting the *Dieffenbachia* studied species (Table 4). Thakuri and Patil (2011) separated some species of the family Euphorbiaceae using petiole anatomy. The petiole outline on the adaxial side is generally concave in the proximal, median and distal regions of all the taxa, except on the adaxial surface of the proximal and median regions of *D. senguine* cv. 'Candida' where it is flat and slightly concave respectively (Figs. 12 and 13). The abaxial outline is round for all the three regions of the taxa.

Parenchyma cells with varying number of layers of zero to one were present in the proximal, median and distal regions of the petiole, except in the proximal region of *D. senguine* where two layers of parenchyma were encountered. All the studied taxa have the collenchyma cells as discontinuous bundles but the types and location of collenchyma cells can be used in separating the species. Angular collenchyma cells were found in all the species and cultivar of *Dieffenbachia*. *D. picta* was the only species found to have additional lamellar collenchyma cells (Figs. 2d, 3d and 4d). Air spaces were present in all the taxa. Collateral vascular bundles scattered in the ground tissues were found in the three regions of all the studied taxa and all the xylem cells are surrounded by xylem parenchyma. Vascular bundles are known to be scattered in Monocots, which is one of their major characteristics (Fahn, 1977).

Table 1. Summary of the main qualitative foliar epidermal features of the adaxial and abaxial surfaces of the studied taxa

Species	Surface	Epidermal cell shape	Anticlinal wall pattern	Stomata shape	Stomata type	Other features
<i>Dieffenbachia picta</i>	Adaxial	Polygonal to Irregular	Wavy to Undulating	Elliptic, circular	Brachyparacytic, Anomocytic	Nil
	Abaxial	Polygonal to Irregular	Straight to Wavy	Elliptic,	Brachyparacytic,	
<i>Dieffenbachia oerstedii</i>	Adaxial	Polygonal to Irregular	Wavy to Undulating	Elliptic, Circular	Brachyparacytic	Nil
	Abaxial	Polygonal to Irregular	Straight to Wavy	Elliptic, Circular	Brachyparacytic, Anomocytic	Cuticular striations
<i>Dieffenbachia senguine</i>	Adaxial	Irregular	Wavy to Undulating	Elliptic	Brachyparacytic, Anomocytic	Druses
	Abaxial	Polygonal to Irregular	Straight to Wavy	Elliptic, Circular	Brachyparacytic	Raphide bundles and druses
<i>Dieffenbachia senguine</i> cv. 'Candida'	Adaxial	Irregular	Wavy to undulating	Elliptic	Brachyparacytic	Nil
	Abaxial	Polygonal to Irregular	Straight to Wavy	Elliptic, Circular	Brachyparacytic	Cuticular striations

Table 2. Mean values of the quantitative foliar anatomical features

Species	Epidermal Area Adaxial ( $\mu\text{m}^2$ )	Epidermal Area Abaxial ( $\mu\text{m}^2$ )	Stomata Area Adaxial ( $\mu\text{m}^2$ )	Stomata Area Abaxial ( $\mu\text{m}^2$ )	Stomata Index Adaxial (%)	Stomata Index Abaxial (%)
<i>Dieffenbachia picta</i>	876.93	891.07	520.74	530.94	4.95	10.22
<i>Dieffenbachia oerstedii</i>	1261.67	1422.70	531.49	592.01	2.62	8.87
<i>Dieffenbachia senguine</i>	1583.86	1349.12	566.44	579.77	3.10	9.25
<i>Dieffenbachia senguine</i> cv. 'Candida'	1570.94	1892.44	700.26	802.26	4.20	10.32



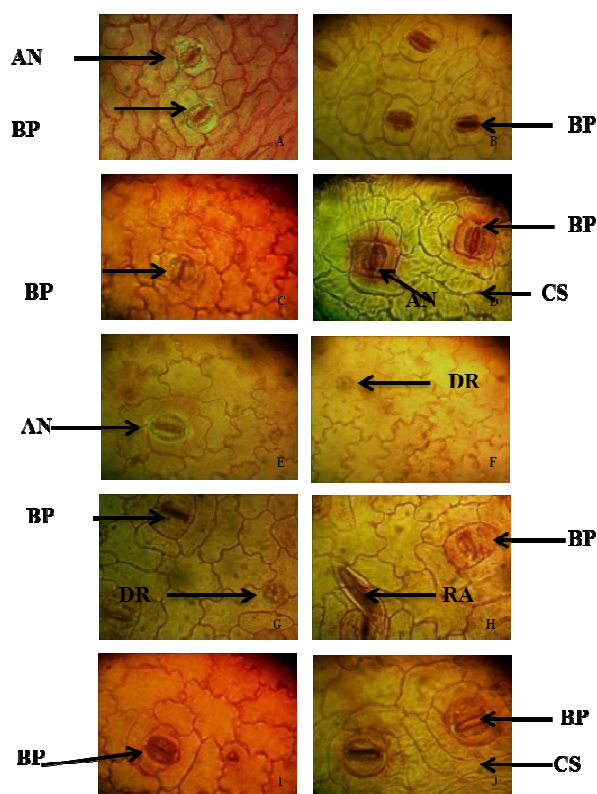


Fig. 1. Leaf epidermal surfaces ( $\times 400$ ). A. Adaxial epidermis of lamina of *D. picta*; B. Abaxial epidermis of lamina of *D. picta*; C. Adaxial epidermis of lamina of *D. oerstedii*; D. Abaxial epidermis of lamina of *D. oerstedii*; E and F. Adaxial epidermis of lamina of *D. sanguinea*; G and H. Abaxial epidermis of the lamina of *D. sanguinea*; I. Adaxial epidermis of lamina of *D. sanguinea* cv. 'Candida'; J. Abaxial epidermis of lamina of *D. sanguinea* cv. 'Candida'. Legend: AN = Anomocytic stomata; BP = Brachyparacytic stomata; CS = Cuticular striations; DR = Druse; RA = Raphides

Cell inclusions in the petioles of the studied species and cultivar include raphides, druses and starch grains. Raphides and druses of calcium oxalate crystals are common occurrences in Araceae species (Middendorf, 1982 and Mayo, *et al.*, 1997). Keating (2004) worked extensively on raphide crystals in Araceae. He described eight types of raphides which are: unmodified, styloids, wide cells (a form of the unmodified raphide crystal), elongated cells, tubular cells, articulated tubes, spindle-shaped and bifurcated raphides. Based on this classification, the raphide types encountered in this study were the wide-unmodified, spindle-shaped and bifurcated types presented in all the studied taxa of *Dieffenbachia*. Keating (2004) did not report the wide cell raphide type for the genus *Dieffenbachia* and in the present study; this type was presented in all the *Dieffenbachia* studied taxa.

Druses were encountered in all the studied taxa. There is no major description for types of druses from the literatures and this was also confirmed by Keating (2004) but in this study some forms of druse appeared more developed than others in terms of size and clarity of outline. Also according to the same author, the understanding and evolution of crystals and their functions remain largely speculative.

#### *D. sanguinea* and its cultivar

Noteworthy in this study is the relationship between *D. sanguinea* and its cultivar *D. sanguinea* cv. 'Candida'. Anomocytic stomata complex, raphide bundles and druses are found occurring in *D. sanguinea* but absent in *D. sanguinea* cv. 'Candida'. Cuticular striations are also found occurring in the epidermis of *D. sanguinea* cv. 'Candida' and not in that of *D. sanguinea*. Flat and slightly concave adaxial petiole outline of the proximal and median regions respectively of *D. sanguinea* cv. 'Candida' also differentiates it from *D. sanguinea* whose proximal and median adaxial outline is concave. The absence of starch grains in the median and distal regions of the petiole of *D. sanguinea* is another delimiting factor. The aforementioned characters can be employed in separating these two taxa. Anatomical characters common to both *D. sanguinea* and *D. sanguinea* cv. 'Candida' are irregular epidermal cell shape on their adaxial surfaces, polygonal to irregular epidermal cell shape on their abaxial surfaces, circular stomata shape on their abaxial epidermal surface, large epidermal cell and stomata areas

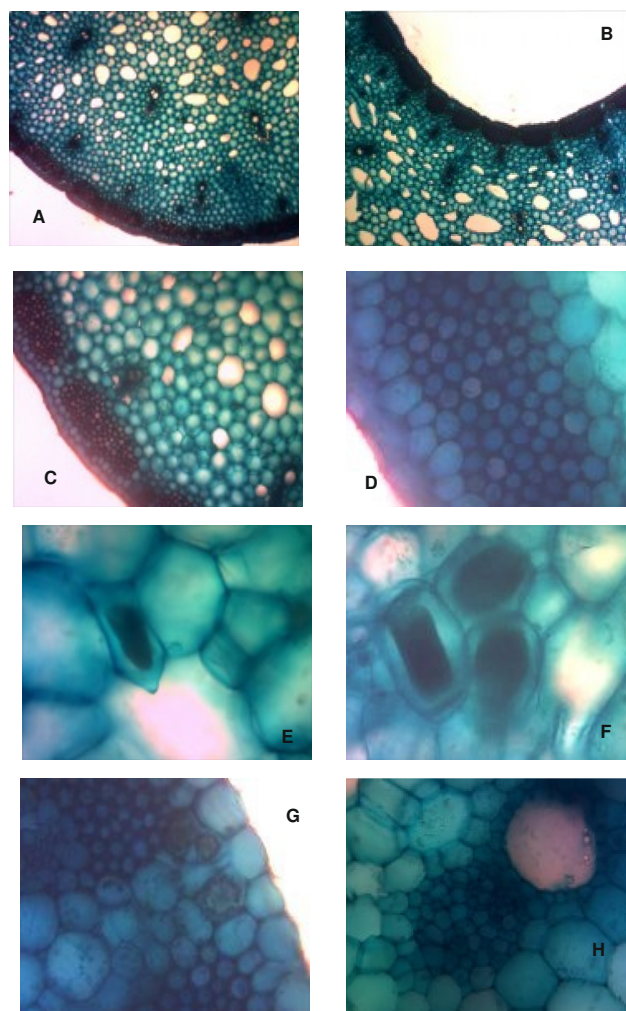


Fig. 2. Transverse section of petiole and cell inclusions in the proximal region of *Dieffenbachia picta*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C.D. Petiole transect ( $\times 100$  and  $\times 400$ ), E.F. Unmodified, wide raphides ( $\times 400$ ), G. Druses in the region ( $\times 400$ ), H. Vascular bundle ( $\times 400$ )

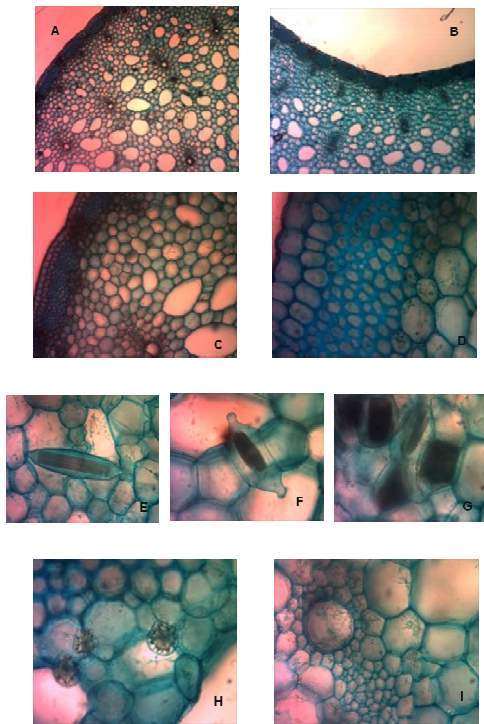


Fig. 3. Transverse section of petiole and cell inclusions in the median region of *D. picta*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$  and  $\times 400$ ), E. F. Spindle-shaped raphides ( $\times 400$ ), G. Unmodified raphides ( $\times 400$ ), H. Druses ( $\times 400$ ), I. Vascular bundle ( $\times 400$ )

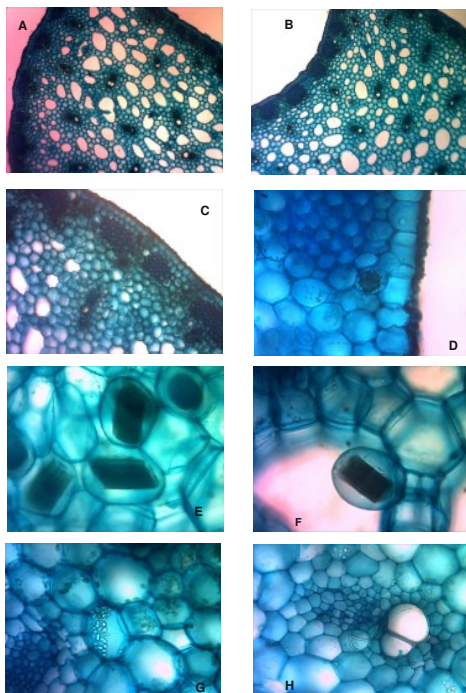


Fig. 5. Transverse section of petiole and cell inclusions in the proximal region of *Dieffenbachia oerstedii*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. Petiole transect ( $\times 100$ ), D. Petiole transect and druse, E. F. Unmodified, wide raphides ( $\times 400$ ), G. Starch grains ( $\times 400$ ), H. Vascular bundle ( $\times 400$ )

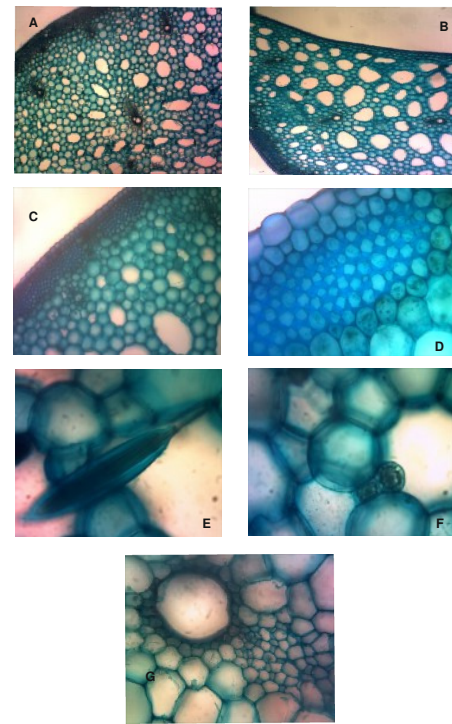


Fig. 4. Transverse section of petiole and cell inclusions in the distal region of *D. picta*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$  and  $\times 400$ ), E. Bifurcated raphide ( $\times 400$ ), F. Druses ( $\times 400$ ) G. Vascular bundle ( $\times 400$ )

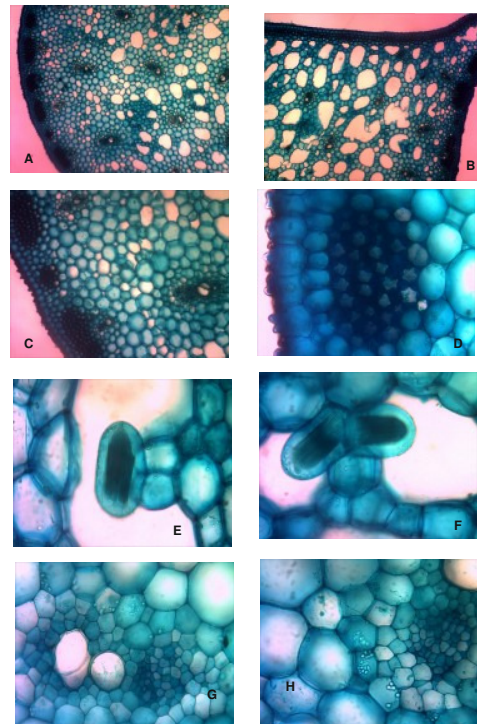


Fig. 6. Transverse section of petiole and cell inclusions in the median region of *D. oerstedii*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$ ,  $\times 400$ ), E. F. Spindle-shaped and unmodified raphides ( $\times 400$ ), G. Vascular bundle ( $\times 400$ ), H. Starch grains ( $\times 400$ )



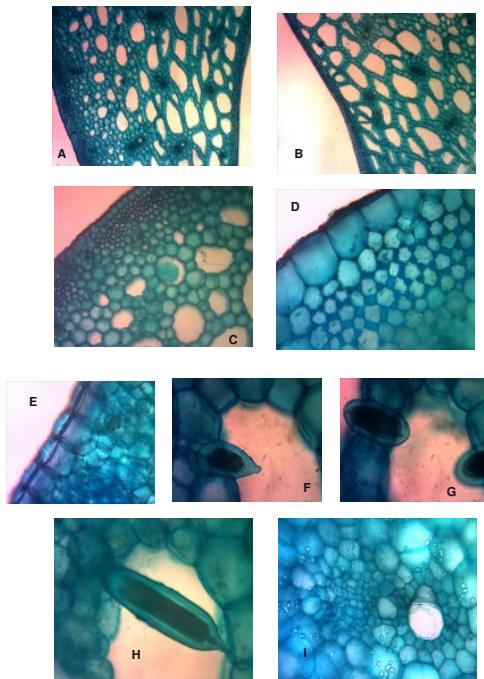


Fig. 7. Transverse section of petiole and cell inclusions in the distal region of *D. oerstedii*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$ ,  $\times 400$ ), E. Druse, F. G. Unmodified and spindle-shaped raphides ( $\times 400$ ), H. Bifurcate raphide ( $\times 400$ ), I. Vascular bundle and starch grains ( $\times 400$ )

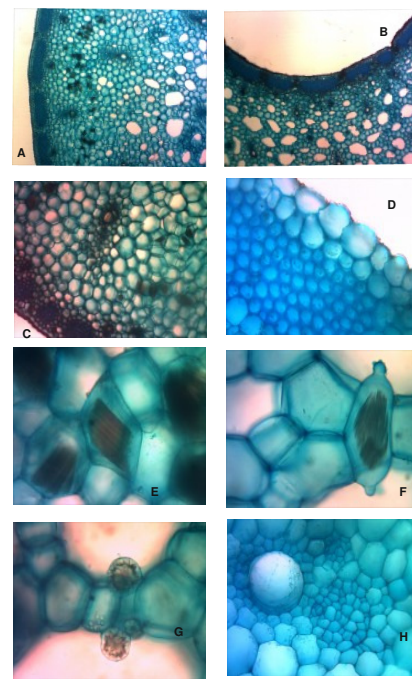


Fig. 8. Transverse section of petiole and cell inclusions in the: Proximal region of *Dieffenbachia senguine*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$ ,  $\times 400$ ), E. Unmodified wide raphides ( $\times 400$ ), F. Spindle-shaped raphide ( $\times 400$ ), G. Druses ( $\times 400$ ), H. Vascular bundle and starch grains ( $\times 400$ )

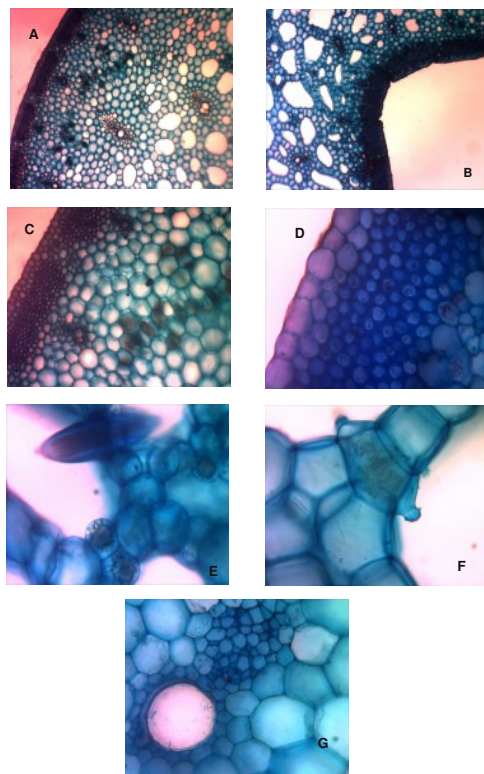


Fig. 9. Transverse section of petiole and cell inclusions in the median region of *D. senguine*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$ ,  $\times 400$ ), E. Spindle-shaped raphide and druses ( $\times 400$ ), F. Unmodified raphide ( $\times 400$ ), G. Vascular bundle ( $\times 400$ )

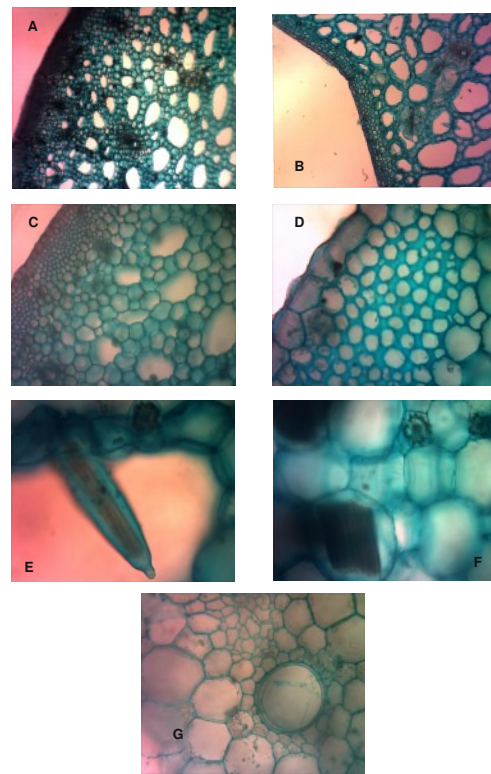


Fig. 10. Transverse section of petiole and cell inclusions in the distal region of *D. senguine*: A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$ ,  $\times 400$ ), E. Bifurcate raphide ( $\times 400$ ), F. Unmodified, wide raphides and druses ( $\times 400$ ), G. Vascular bundle ( $\times 400$ )

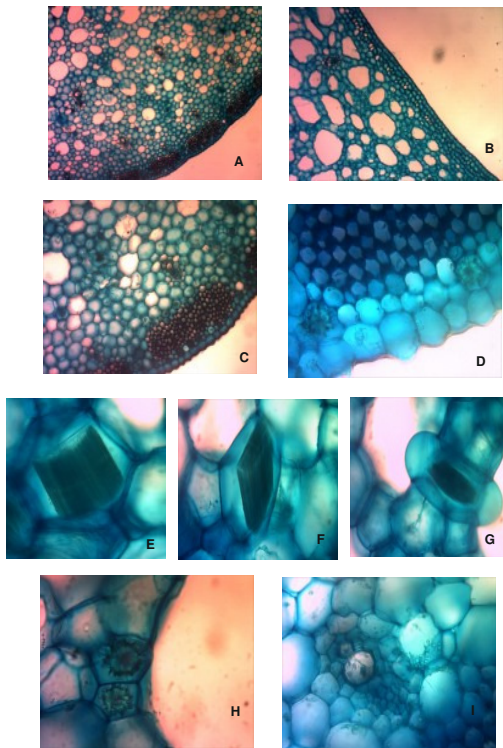


Fig. 11. Transverse section of petiole and cell inclusions in the proximal region of *Dieffenbachia sanguine* cv. 'Candida': A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$ ,  $\times 400$ ), E. F. Unmodified, wide ( $\times 400$ ), G. Spindle-shaped raphides ( $\times 400$ ), H. Druses ( $\times 400$ ), I. Vascular bundle ( $\times 400$ )

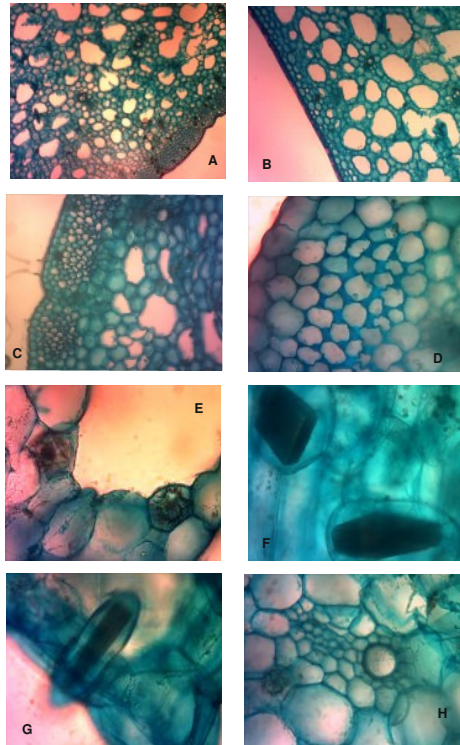


Fig. 13. Transverse section of petiole and cell inclusions in the distal region of *D. sanguine* cv. 'Candida': A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$ ,  $\times 400$ ), E. Druses ( $400\times$ ), F. G. Unmodified and bifurcated raphides ( $\times 400$ ), H. Vascular bundle and Druse ( $\times 400$ )

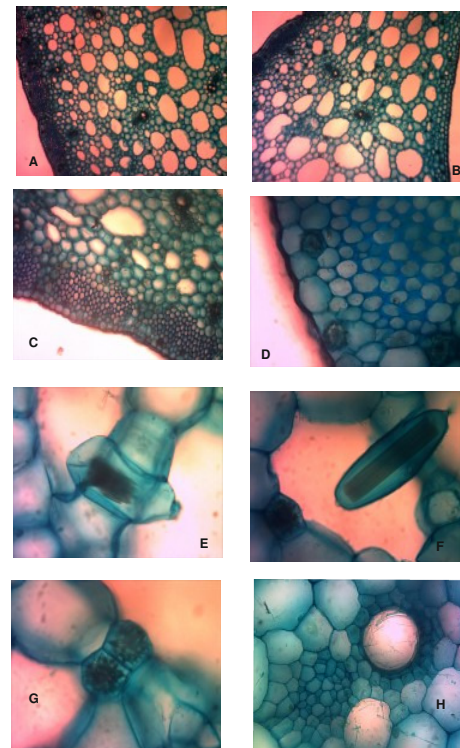


Fig. 12. Transverse section of petiole and cell inclusions in the Median region of *D. sanguine* cv. 'Candida': A. Abaxial outline of petiole ( $\times 40$ ), B. Adaxial outline of petiole ( $\times 40$ ), C. D. Petiole transect ( $\times 100$ ,  $\times 400$ ), E. F. Unmodified and bifurcated raphides ( $\times 400$ ), G. Druses ( $\times 400$ ), H. Vascular bundle ( $\times 400$ )

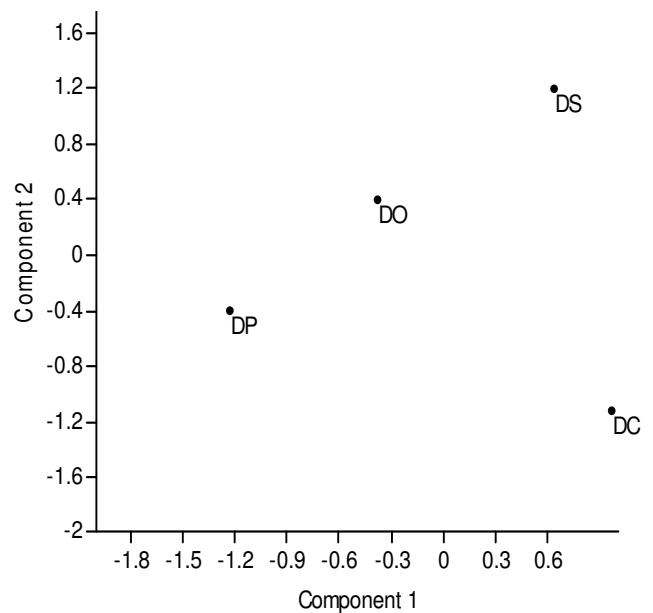


Fig. 14. Principal Components analysis showing the relationship of *Dieffenbachia* taxa based on 23 epidermal and petiole anatomical characters on axis 1 horizontal and axis 2 vertical. Legend: DP - *Dieffenbachia picta*; DO - *Dieffenbachia oerstedii*; DS - *Dieffenbachia sanguine*; DC - *Dieffenbachia sanguine* cv. 'Candida'.

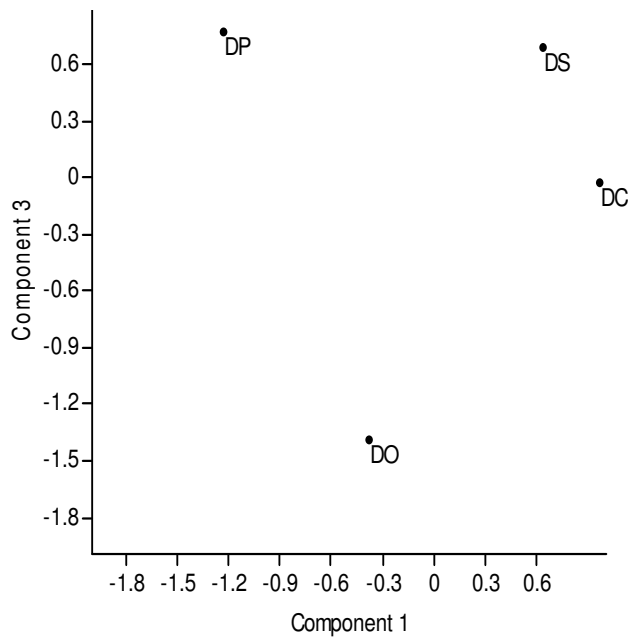


Fig. 15. Principal Components Analysis (PCA) showing the relationship of *Dieffenbachia* taxa based 23 epidermal and petiole anatomical characters on axis 1 horizontal and axis 3 vertical. Legend: DP - *Dieffenbachia picta*; DO - *Dieffenbachia oerstedii*; DS - *Dieffenbachia senguine*; DC - *Dieffenbachia senguine* cv. 'Candida'.

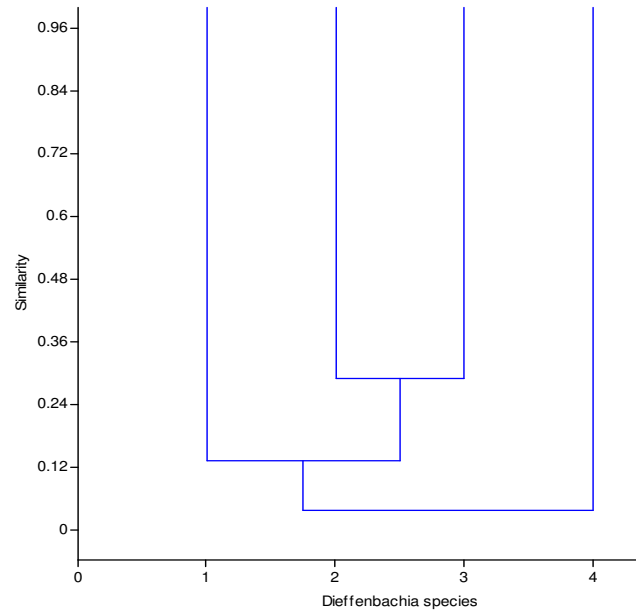


Fig.16. Single Linkage Cluster Analysis (SCLA) dendrogram of the studied taxa of *Dieffenbachia*. Legend: DP - *Dieffenbachia picta*; DO - *Dieffenbachia oerstedii*; DS - *Dieffenbachia senguine*; DC - *Dieffenbachia senguine* cv. 'Candida'.

Table 3. Summary of the proximal, median and distal regions of the petiole anatomy of the *Dieffenbachia* taxa

Characters Species	Petiole Region	Adaxial Outline	Abaxial Outline	Layers of Parenchyma Cells	Type of Collenchyma Cells	Raphide Types Unmodified wide (UW), Spindle shaped (S) and Biforine (B)	Druses (+/-)	Starch Grains (+/-)
<i>Dieffenbachia picta</i>	Proximal	Concave	Round	0-1	Angular and Lamellar	UW	+	+
	Median	Concave	Round	0-1	Angular and Lamellar	UW, S	+	+
	Distal	Concave	Round	0-1	Lamellar	UW, B	+	+
<i>Dieffenbachia oerstedii</i>	Proximal	Concave	Round	0-1	Angular	UW	+	+
	Median	Concave	Round	1	Angular	UW, S	+	+
	Distal	Concave	Round	1	Angular	UW, S, B	+	+
<i>Dieffenbachia senguine</i>	Proximal	Concave	Round	1-2	Angular	UW, S	+	+
	Median	Concave	Round	1	Angular	UW, S	+	-
	Distal	Concave	Round	1	Angular	UW, B	+	-
<i>Dieffenbachia senguine</i> cv. 'Candida'	Proximal	Flat	Round	0-1	Angular	UW, S	+	+
	Median	Slightly Concave	Round	0-1	Angular	UW, B	+	+
	Distal	Concave	Round	0-1	Angular	UW, B	+	+

Legend: + = Present; - = Absent

Table 4. Eigen-value and percentage of variation accounted for by the first three components axes of ordination of *Dieffenbachia* species using their quantitative and qualitative anatomical data

Principal Component (PC)	Eigen-value	Percentage of Variation	Cumulative Percentage
PC1	4.3172	53.41	53.41
PC2	2.3825	29.47	82.88
PC3	1.3836	17.12	100



compared to others, concave adaxial petiole outline of the distal region and the presence of unmodified wide, spindle-shaped and bifurcate raphide types in the three regions of their petiole.

#### Data analysis

The results of the Principal Components Analysis and Single Linkage Cluster Analysis (SLCA) are as shown in Figs. 14, 15 and 16. The graph of the Principal Components Analysis (PCA) based on the qualitative and quantitative anatomical data separated the four taxa when components 1 and 2 (Fig. 14) were used but on the graph of components 1 and 3 (Fig. 15), *D. senguine* and *D. senguine* cv. 'Candida' were grouped together. The first three components of the PCA accounted for the total variation among the studied taxa. From the PCA loadings, it can be gathered that the characters responsible for the separation of the studied taxa from component one are the adaxial and abaxial epidermal cells area as well as the adaxial stomata area, those of component two are abaxial stomata index and number of layers of parenchyma cells on the median and distal regions of the taxa while those of component three are epidermal cuticular striations and adaxial stomata index. The aforementioned are the delimiting characters in the studied species.

The result of the SLCA grouped the taxa into two (Fig. 16); separating *D. picta* from the other three taxa in the first main grouping. In the second main grouping, *D. oerstedii* was separated, leaving *D. senguine* and *D. senguine* cv. 'Candida' clustered together at a higher similarity level. This agrees with the result of the PCA using components 1 and 3.

#### Conclusions

The foliar and petiole anatomy of genus *Dieffenbachia* provided a number of diagnostic features or characters in the genus. These include irregular epidermal cell shape on the adaxial surfaces of *D. senguine* and *D. senguine* cv. 'Candida', the presence of anomocytic stomata type on the abaxial surfaces of *D. picta*, *D. oerstedii* and on the adaxial surface of *D. senguine*, in addition to the brachyparacytic stomata type common to all the taxa, cuticular striations on the abaxial surfaces of *D. oerstedii* and *D. senguine* cv. 'Candida', druses and raphide bundle in *D. senguine* only, flat adaxial petiole outline and slightly concave adaxial petiole outline in the proximal and median regions of *D. senguine* cv. 'Candida' and the presence of lamellar type of collenchyma cells in the petiole of *D. picta*.

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