FOLIAR EPIDERMAL STUDY OF SOME SPECIES OF *AGLAONEMA* Schott (ARACEAE) IN NIGERIA

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ABSTRACT

The adaxial and abaxial epidermal surfaces of six species of *Aglaonema* Schott were studied in order to document characters of taxonomic importance among them. The species are *Aglaonema brevispathum* Schott (Engl.) Engl., *Aglaonema commutatum* Schott, *Aglaonema costatum* Schott, *Aglaonema crispum* Schott, *Aglaonema pictum* Schott and *Aglaonema rotundum* Schott. Epidermal peels of both the adaxial and abaxial surfaces of the species were made following standard procedures. Qualitative and quantitative characters were observed and measured respectively. Photomicrographs of the surfaces were also taken. Generic characters revealed include brachyparacytic stomata complex, elliptic stomata shape, polygonal to irregular epidermal cell shape as well as straight to wavy anticlinal cell wall pattern on the abaxial surface. Additional anisocytic and anomocytic stomata complexes were present in some of the species. The hypostomatic nature of *A. rotundum* was documented. This clearly separated it from the other species in this study which are amphistomatic in nature. Anticlinal wall pattern varies from wavy to undulating to sinuous on the adaxial surface and straight to wavy on the abaxial surface of the species. Irregular epidermal cell shape on the adaxial surface separated *A. costatum* and *A. pictum* from the other species which have polygonal to irregular epidermal cell shape. Highest epidermal cell size and stomata size was encountered on both surfaces of *A. pictum*. Druses and raphides of calcium oxalate were present in *A. commutatum*, *A. costatum* and *A. crispum* which delimited them from the other species. The study reveals key diagnostic features among the species studied. Further research work from other fields of evidence is recommended in order to shed more light on the taxonomy of the species.

Key words: Araceae, *Aglaonema*, Taxonomy, Epidermis, Brachyparacytic

INTRODUCTION

The Araceae is a family of herbaceous monocotyledonous flowering plant in which flowers are borne on a type of inflorescence called a spadix. The family is a large family of more than 105 genera and 3300 species (Mayo *et al.*, 1997) of tropical and temperate distribution (Vargas *et al.*, 2004; Mora *et al.*, 2006). Members of the family Araceae, also known as aroids, are of diverse growth forms and ecological groups (Croat, 1992). Aroids contain oxalic acid, most of which is deposited in plant tissues as crystals of calcium oxalate. Calcium oxalate crystals can form in any organ or tissue within the plants as they are found on leaves, stems, petiole, corm and root of Araceae plants (Middendorf, 1982).

Aglaonema Schott, tribe Aglaonemateae, is a genus of tropical aroids of about 21 species native to Southern Asia from northeastern India across Southern China and Indonesia through New Guinea They are grown for their ornamental foliage in homes, greenhouses, and in the outdoors (Nicolson, 1969). Species of *Aglaonema* are herbaceous perennial plants growing to 20-150 cm in height. The leaves are several, alternate on the stems, lanceolate to narrowly ovate, dark to medium green, 10-45 cm long and 4-16 cm broad, depending on the species. *Aglaonema* inflorescence, spadix, consists of an upright central axis covered with several minute petal-less flowers. The flowers are relatively inconspicuous, white or greenish-white spathes that can give way to red berries. Staminate (male) flowers cover the upper half of the spadix and pistillate (female) flowers are located on the basal half (Henny, 1983).

The sap of *Aglaonema* species is poisonous to health. It causes skin irritation. If ingested, the sap causes irritation of the mouth, lips, throat and tongue (Bown, 2000). Stem is erect to decumbent, internodes green, becoming brown with age, smooth, often rooting at nodes when decumbent. They are easy to propagate from cuttings, which will root even in a glass of water. These plants survive well in moist, but well drained, heavy soil. They are among the best plants for poorly lighted

situations, though bright, indirect sunlight or partial shade is best (Henny, 1988). Chromosome number reported for the genus by Mayo *et al.* (1997) is 2n = 40, 60, 80, 100, 120.

Anatomical method has been a widely used tool in the Plant Taxonomy. Many authors have employed plant anatomical methods in resolving identification problems among groups of plants. Such studies include the works of Adedeji (2004) on species of *Emilia* Cass., Adedeji and Illoh (2004) on some species of *Hibiscus* Linn., Green and Oguzor (2009) on four selected species from genera *Xanthosoma*, *Dieffenbachia* and *Colocasia*, Adedeji (2012) on the species of *Stachytarpheta* Vahl., Ina and Eka (2013) on some species in the genera *Alocasia*, *Colocasia* and *Remuatia*, Oladipo and Ayo-Ayinde (2014) on genera *Aneilema* and *Commelina*, Osuji and Nwala (2015) on some cultivars of *Xanthosoma* and *Colocasia*.

Some research works have been carried out on the genus *Aglaonema*. Nicolson (1969) did a revision of the genus. Benedetto *et al.* (2005), Benedetto *et al.* (2006) and Chao-Yi and Der-Ming (2008) worked extensively on the physiology of *Aglaonema commutatum* as well as that of other Araceae members. Mohamed *et al.* (2006) established a positive correlation between the chromosome number and the protein electrophoretic band patterns in two species of *Aglaonema* as well as some other Araceae members. There is no known published work on the foliar epidermal morphology of species in the genus *Aglaonema* in Nigeria.

MATERIALS AND METHODS

The six species of the genus *Aglaonema* used for this work are *Aglaonema brevispathum* Schott (Engl.) Engl., *Aglaonema commutatum* Schott, *Aglaonema costatum* Schott, *Aglaonema crispum* Schott, *Aglaonema pictum* Schott and *Aglaonema rotundum* Schott. They were collected from Rainbows Q Gardens (N07°24.148' E003°57.726'), Ibadan, Oyo State and Parks and Gardens Unit (N07°31.335' E004°31.829'), Obafemi Awolowo University, Ile-Ife, Nigeria. The species were identified at the Forestry Research Institute of Nigeria (FHI), Ibadan. Voucher specimens were deposited at the Obafemi Awolowo University (IFE) herbarium. The scrape technique of Metcalfe (1960) was used in getting the epidermal peels of both the adaxial and abaxial surfaces of their fresh leaves. The median portion of well expanded leaves were peeled by placing the desired epidermal surface face down on a glass slide and then scraping off with a sharp blade all tissues above the required epidermis. The epidermal peels were then stained in 1% aqueous Safranin O and mounted in 25% glycerin for microscopic examination.

The epidermal tissue of each species was carefully studied in search of constant micromorphological characters and features. The long and short axes of 50 epidermal cells and the length and width of 50 stomata were measured at random using an ocular micrometer. Epidermal cell area was calculated by multiplying the length of the long and short axes of the epidermal cells, while stomata area was calculated by multiplying the length and width of the stomata. Also calculated was the Stomata Index (I) for the two surfaces, that is, the adaxial and abaxial surfaces, using the formula below according to Metcalfe and Chalk (1979).

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

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

RESULTS

The summary of the important qualitative and quantitative foliar epidermal features of the adaxial and the abaxial surfaces of the six species of *Aglaonema* studied are shown on Tables 1 and 2 respectively. The photomicrographs of the adaxial and the abaxial surfaces taken with the aid of an AmScope camera attached to a light microscope are shown in Figures 1 (A - N) and Figures 2 (A-L) respectively. Detailed description of the epidermal surfaces is presented as follows.

A. brevispathum

Adaxial Surface

Epidermal cells are polygonal to irregular with wavy to undulating anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 816.0 μm^2 - 2570.4 μm^2 with mean value of 1593.92 μm^2. Amphistomatous,

restricted to the non-venous regions; stomata complex is brachyparacytic, occasionally anomocytic and anisocytic, stomata shape is elliptic, occasionally circular **(**Figures 1 A - C). Stomata size ranges between 435.2 μm^2 - 816.0 μm^2 with mean value of 671.70 μm^2 and stomata index ranges between 2.5 - 3.03% with mean value of 2.70%.

Abaxial Surface

Epidermal cells are polygonal to irregular with straight to slightly wavy anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 550.8 μm^2-1955.0 μm^2 with mean value of 1181.16 μm^2. Amphistomatous, restricted to the non-venous regions; stomata complex is brachyparacytic, occasionally anomocytic and anisocytic, stomata shape is elliptic **(**Figure 2 A - B). Stomata size ranges between 489.6 μm^2 - 693.6 μm^2 with mean value of 599.62 μm^2 and stomata index ranges between 7.69 - 14.29% with mean value of 11.14%.

A. commutatum

Adaxial Surface

Epidermal cells are polygonal to irregular with wavy anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 442.0 μm^2-2356.2 μm^2 with mean value of 1379.52 μm^2. Amphistomatous, restricted to the non-venous regions; stomata complex is brachyparacytic, stomata shape is elliptic, occasionally circular **(**Figure 1 D). Stomata size ranges between 326.4 μm^2 -561 μm^2 with mean value of 447.30 μm^2 and stomata index ranges between 2.27 - 5.0% with mean value of 2.99%.

Abaxial Surface

Epidermal cells are polygonal to irregular with straight to wavy anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 489.6 μm^2- 2448.0 μm^2 with mean value of 1433.1 μm^2. Amphistomatous, restricted to the non-venous regions; stomata complex is brachyparacytic, occasionally anisocytic, stomata shape is elliptic, occasionally circular **(**Figures 2 C - E). Stomata size ranges between 459.0 μm^2 - 652.8 μm^2 with mean value of 549.64 μm^2 and stomata index ranges between 6.67 - 11.11% with mean value of 8.58%. Druses are present.

A. costatum

Adaxial Surface

Epidermal cells are irregular with undulating to sinuous anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 578.0 μm^2- 2169.2 μm^2 with mean value of 1170.43 μm^2. Amphistomatous, though sparsely distributed and restricted to the non-venous regions; stomata complex is brachyparacytic, occasionally anomocytic and anisocytic; stomata shape is elliptic, occasionally circular **(**Figures 1 E - I). Stomata size ranges between 353.6 μm^2 – 612.0 μm^2 with mean value of 481.83 μm^2 and stomata index ranges between 1.54 – 5.08% with mean value of 2.73%. Druses and raphides are present.

Abaxial Surface

Epidermal cells are polygonal to irregular with straight to wavy anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 408.0 μm^2- 2550.0 μm^2 with mean value of 1261.54 μm^2. Amphistomatous, restricted to the non-venous regions; stomata complex is brachyparacytic, occasionally anisocytic or anomocytic; stomata shape is elliptic, often circular **(**Figure 2 F). Stomata size ranges between 408.0 μm^2 - 652.8 μm^2 with mean value of 534.55 μm^2 and stomata index ranges between 4.0 – 10.20% with mean value of 7.48%.

A. crispum

Adaxial Surface

Epidermal cells are polygonal to irregular with wavy to slightly undulating anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 635.8 μm^2- 2907.0 μm^2 with mean value of 1687.28 μm^2. Amphistomatous, though sparsely distributed and restricted to the non-venous regions; stomata complex is brachyparacytic, occasionally anomocytic; stomata shape is elliptic, occasionally circular **(**Figure 1 J - L). Stomata size ranges between 380.8 μm^2 – 809.2 μm^2 with mean value of 545.7 μm^2 and stomata index ranges between 2.17 – 5.13% with mean value of 3.03%. Druses are present.

Abaxial Surface

Epidermal cells are polygonal to irregular with

straight to wavy anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 856.8 μm^2- 3672.0 μm^2 with mean value of 1757.26 μm^2. Amphistomatous, restricted to the non-venous regions; stomata complex is brachyparacytic, occasionally anomocytic; stomata shape is elliptic, occasionally circular **(**Figure 2 G - I). Stomata size ranges between 459.0 μm^2 -751.4 μm^2 with mean value of 594.18 μm^2 and stomata index ranges between 7.14 – 13.33% with mean value of 10.60%. Druses and raphides are present.

A. pictum

Adaxial Surface

Epidermal cells are irregular with wavy to undulating anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 897.6 μm^2- 3019.2 μm^2 with mean value of 1768.82 μm^2. Amphistomatous, sparsely distributed on this surface and restricted to the non-venous regions; stomata complex is brachyparacytic, stomata shape is elliptic **(**Figure 1 M). Stomata size ranges between 646.0 μm^2 - 904.4 μm^2 with mean value of 786.53 μm^2 and stomata index ranges between 2.86 - 3.70% with mean value of 3.20%.

Abaxial Surface

Epidermal cells are polygonal to irregular with straight to wavy anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 952.0 μm^2-2992.0 μm^2 with mean value of 1880.40 μm^2. Amphistomatous, restricted to the non-venous regions; stomata complex is brachyparacytic, occasionally anomocytic, stomata shape is elliptic, occasionally circular **(**Figure 2 J). Stomata size ranges between 751.4 μm^2 - 969.0 μm^2 with mean value of 856.53 μm^2 and stomata index ranges between 7.14 - 12.5% with mean value of 9.98%.

A. rotundum

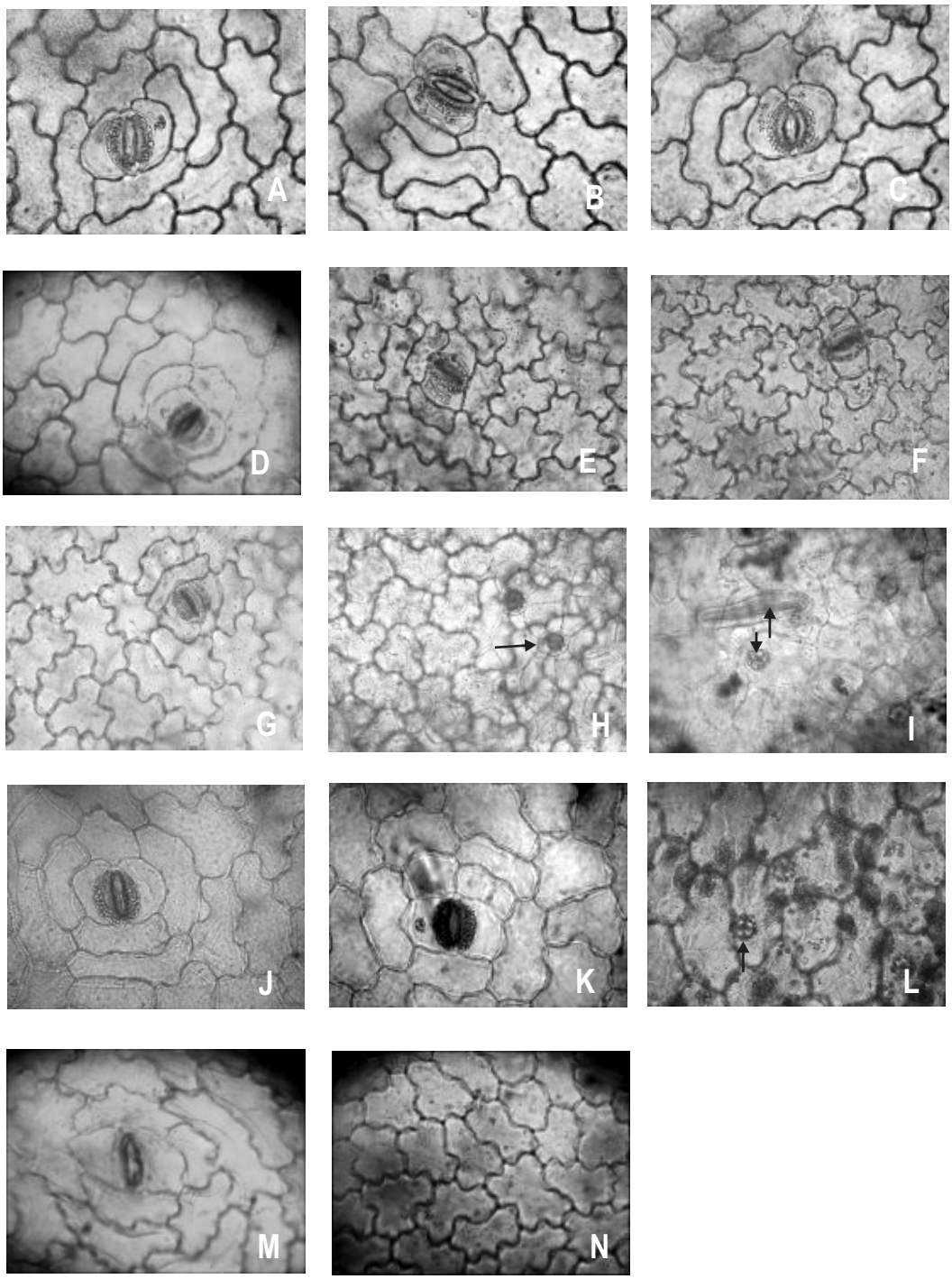
Adaxial Surface

Epidermal cells are polygonal to irregular with wavy to slightly undulating anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 612.0 μm^2- 1570.8 μm^2 with mean value of 945.61 μm^2. Stomata absent **(**Figure 1 N).

Abaxial Surface

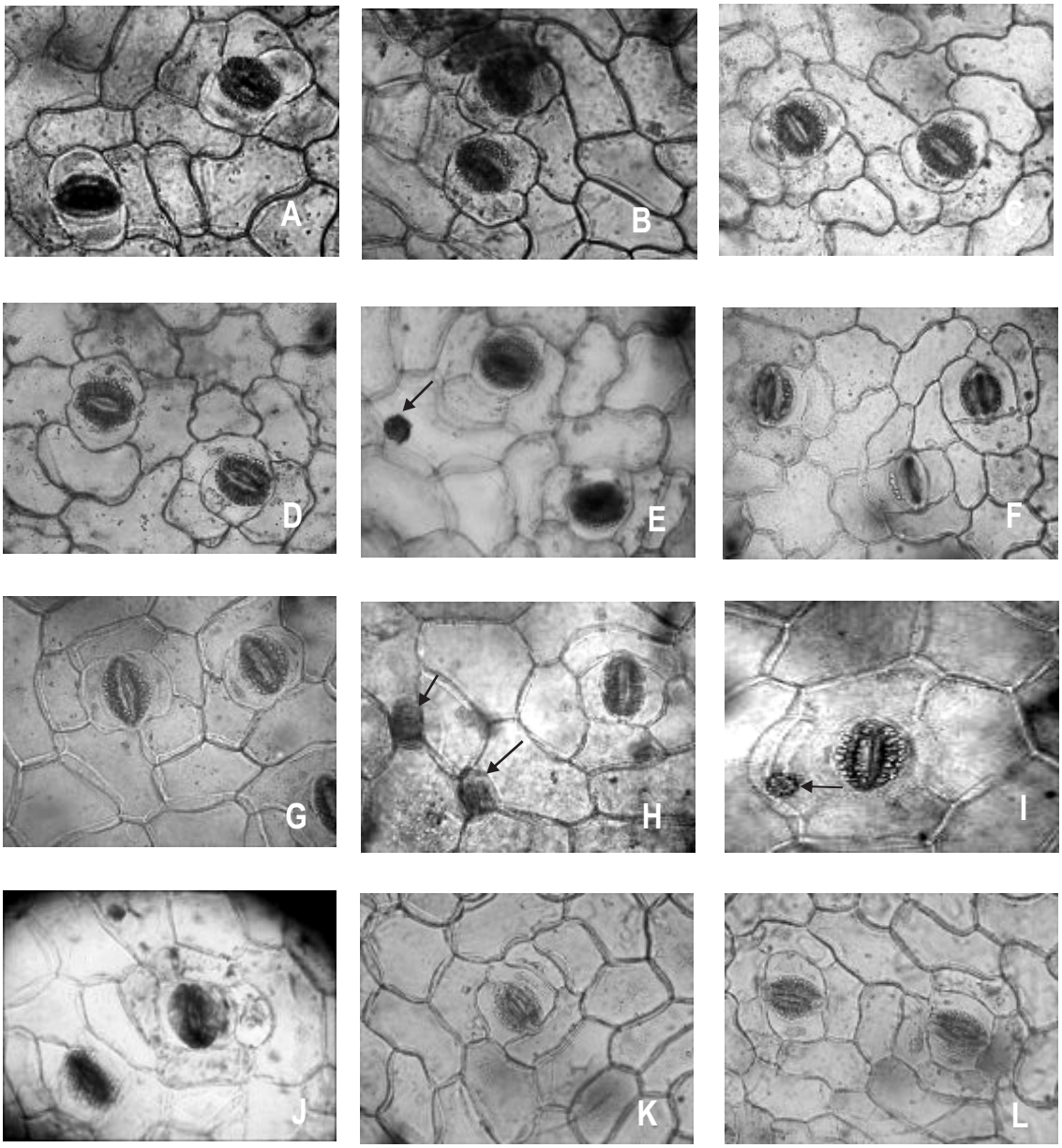
Epidermal cells are polygonal to irregular with straight to wavy anticlinal wall. They vary in size, shape and arrangement. Epidermal cell area ranges between 333.2 μm^2- 1700.0 μm^2 with mean value of 1100.51 μm^2. Leaf is hypostomatous, restricted to the non-venous regions: stomata complex is brachyparacytic, occasionally anomocytic and anisocytic, stomata shape is elliptic **(**Figure 2 K - L). Stomata size ranges between 510.0 μm^2 – 663.0 μm^2 with mean value of 588.0 μm^2 and stomata index ranges between 4.76 - 11.29% with mean value of 7.88%.

Table 3 is showing the Eigenvalue and the percentage of total variation accounted for by the first three components axes of ordination of the *Aglaonema* species. Figure 3 shows the clustered diagram of the species while Figure 4 is the graph of the species using the first two components from the Principal Components Analysis.



Figures 1 (A – N): Photomicrographs of adaxial lamina surfaces of *Aglaonema* species showing stomata types/morphology.

A. *Aglaonema brevispathum* (brachyparacytic), **B**. *A. brevispathum* (anomocytic), **C**. *A. brevispathum* (anisocytic), **D**. *A. commutatum* (brachyparacytic), **E**. *A. costatum* (brachyparacytic), **F**. *A. costatum* (anomocytic), **G**. *A. costatum* (anisocytic), **H**. Druses (Arrowed) in *A. costatum*, **I**. Raphide (arrowed up) and Druse (arrowed down) in *A. costatum*, **J.** *A. crispum*, (brachyparacytic) **K.** *A. crispum* (anomocytic), **L**. Druses (arrowed) in *A. crispum*, **M**. *A. pictum* (brachyparacytic), **N**. Epidermal surface of *A. rotundum*. All magnifications at X400.



Figures 2 (A – L): Photomicrographs of abaxial lamina surfaces of *Aglaonema* species showing stomata types/morphology.

A. *A. brevispathum* (brachyparacytic and anomocytic), **B**. *A. brevispathum* (anisocytic), **C.** *A. commutatum* (brachyparacytic and anomocytic) **D**. *A. commutatum* (brachyparacytic and anisocytic), **E**. Brachyparacytic stomata and druse (arrowed) in *A. commutatum*, **F.** *A. costatum* (brachyparacytic, anomocytic and anisocytic), **G**. *A. crispum* (brachyparacytic and anomocytic), **H**. Brachyparacytic and Raphides (arrowed) in *A. crispum*, **I**. Brachyparacytic stomata and druse (arrowed) in *A. crispum*, **J**. *A. pictum* (brachyparacytic and anomocytic), **K.** Anisocytic stomata in *A. rotundum,* **L**. *A. rotundum* (brachyparacytic and anomocytic). All magnifications at X400.

Table 1: Important Foliar Epidermal Features of the Adaxial Surfaces of the Species of *Aglaonema* Studied

Species studied	Important characters of the foliar epidermal tissue							
	Epidermal cell shape	**Anticlinal wall pattern**	**Stomata Shape**	**Stomata type**	**Cell inclusion**	**Mean Epidermal Area (µm^2)**	**Mean Stomata Area (µm^2)**	**Mean Stomata Index (%)**
Aglaonema brevispathum	Polygonal to Irregular	Wavy to undulating	Elliptic, Circular	Brachyparacytic, Anomocytic, Anisocytic	Nil	1593.92	671.70	2.70
Aglaonema commutatum	Polygonal to Irregular	Wavy	Elliptic, Circular	Brachyparacytic	Nil	1379.52	447.30	2.99
Aglaonema costatum	Irregular	Undulating to sinuous	Elliptic, Circular	Brachyparacytic, Anomocytic, Anisocytic	Druses, Raphides	1170.43	481.83	2.73
Aglaonema crispum	Polygonal to Irregular	Wavy to undulating	Elliptic, Circular	Brachyparacytic, Anomocytic	Druses	1687.28	545.7	3.03
Aglaonema pictum	Irregular	Wavy to Undulating	Elliptic	Brachyparacytic	Nil	1768.82	786.53	3.20
Aglaonema rotundum	Polygonal to Irregular	Wavy to slightly undulating	Nil	Nil	Nil	945.61	Nil	Nil

Table 2: Important Foliar Epidermal Features of the Abaxial Surfaces of the Species of *Aglaonema* Studied

Species studied	Important characters of the foliar epidermal tissue							
	Epidermal cell shape	**Anticlinal wall pattern**	**Stomata Shape**	**Stomata type**	**Cell inclusion**	**Mean Epidermal Area (µm^2)**	**Mean Stomata Area (µm^2)**	**Mean Stomata Index (%)**
Aglaonema brevispathum	Polygonal to Irregular	Straight to wavy	Elliptic	Brachyparacytic, Anomocytic, Anisocytic	Nil	1181.16	599.62	11.14
Aglaonema commutatum	Polygonal to Irregular	Straight to wavy	Elliptic, Circular	Brachyparacytic, Anisocytic	Druse	1433.1	549.64	8.58
Aglaonema costatum	Polygonal to Irregular	Straight to wavy	Elliptic, Circular	Brachyparacytic, Anomocytic, Anisocytic	Nil	1261.54	534.55	7.48
Aglaonema crispum	Polygonal to Irregular	Straight to wavy	Elliptic, Circular	Brachyparacytic, Anomocytic	Druses, Raphides	1757.26	594.18	10.60
Aglaonema pictum	Polygonal to Irregular	Straight to wavy	Elliptic, Circular	Brachyparacytic, Anomocytic	Nil	1880.40	856.53	9.98
Aglaonema rotundum	Polygonal to Irregular	Straight to slightly wavy	Elliptic	Brachyparacytic, Anomocytic, Anisocytic	Nil	1100.51	588.0	7.88

Table 3: Eigenvalue and the percentage of total variation accounted for by the first three components axes of ordination of the *Aglaonema* species.

Principal components axis	Eigenvalue	Percentage of total variation	Cummulative percentage
I	192724	82.33	82.33
II	23951.3	10.23	92.56
III	17058.9	7.29	99.85

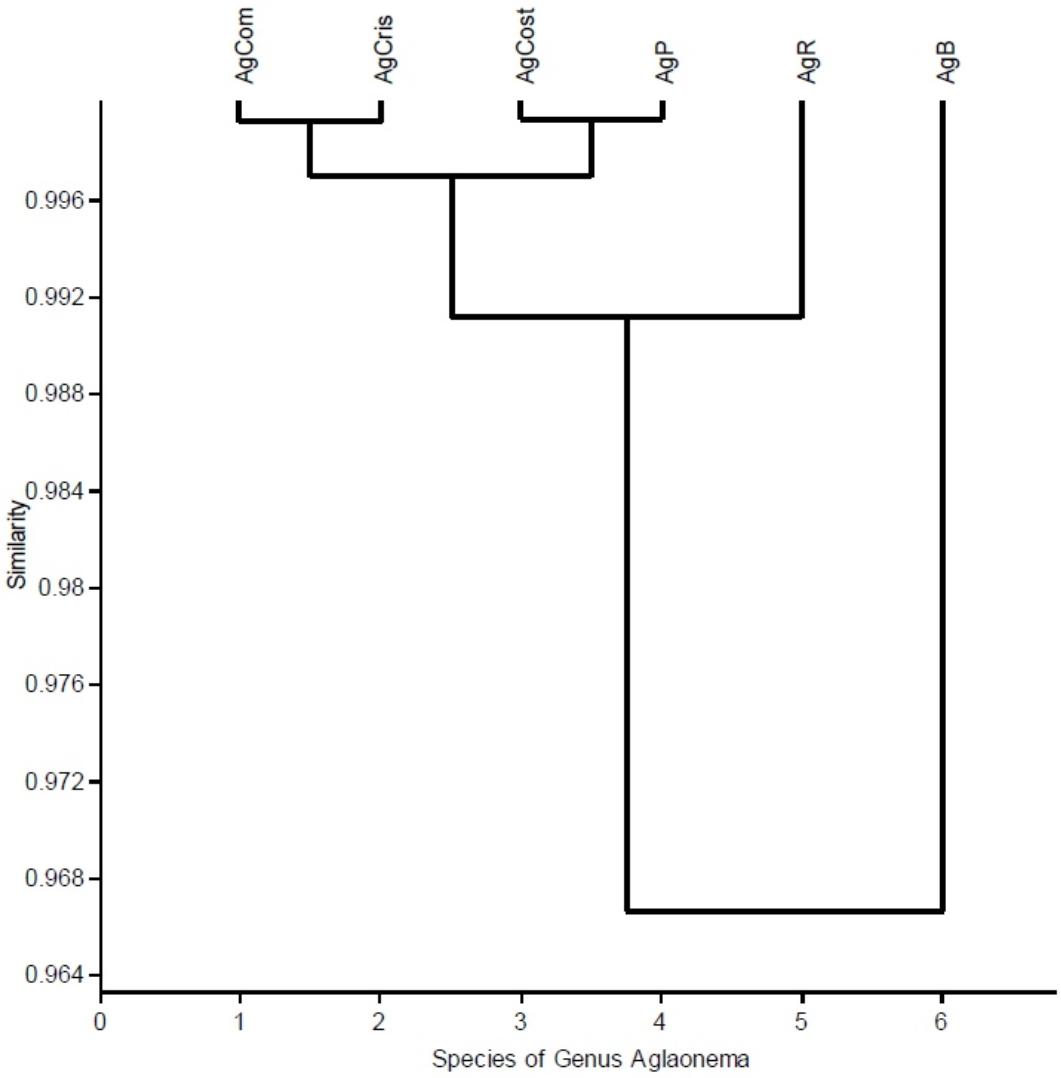


Figure 3: **Single Linkage Cluster Analysis (SCLA) Dendrogram of the species of *Aglaonema* Studied Using Six Mean Anatomical Quantitative Data.**

Legend: **AgB** – *Aglaonema brevispathum*; **AgCom** – *Aglaonema commutatum*; **AgCost** – *Aglaonema costatum*; **AgCris** – *Aglaonema crispum*; **AgP** – *Aglaonema pictum*; **AgR** – *Aglaonema rotundum*.

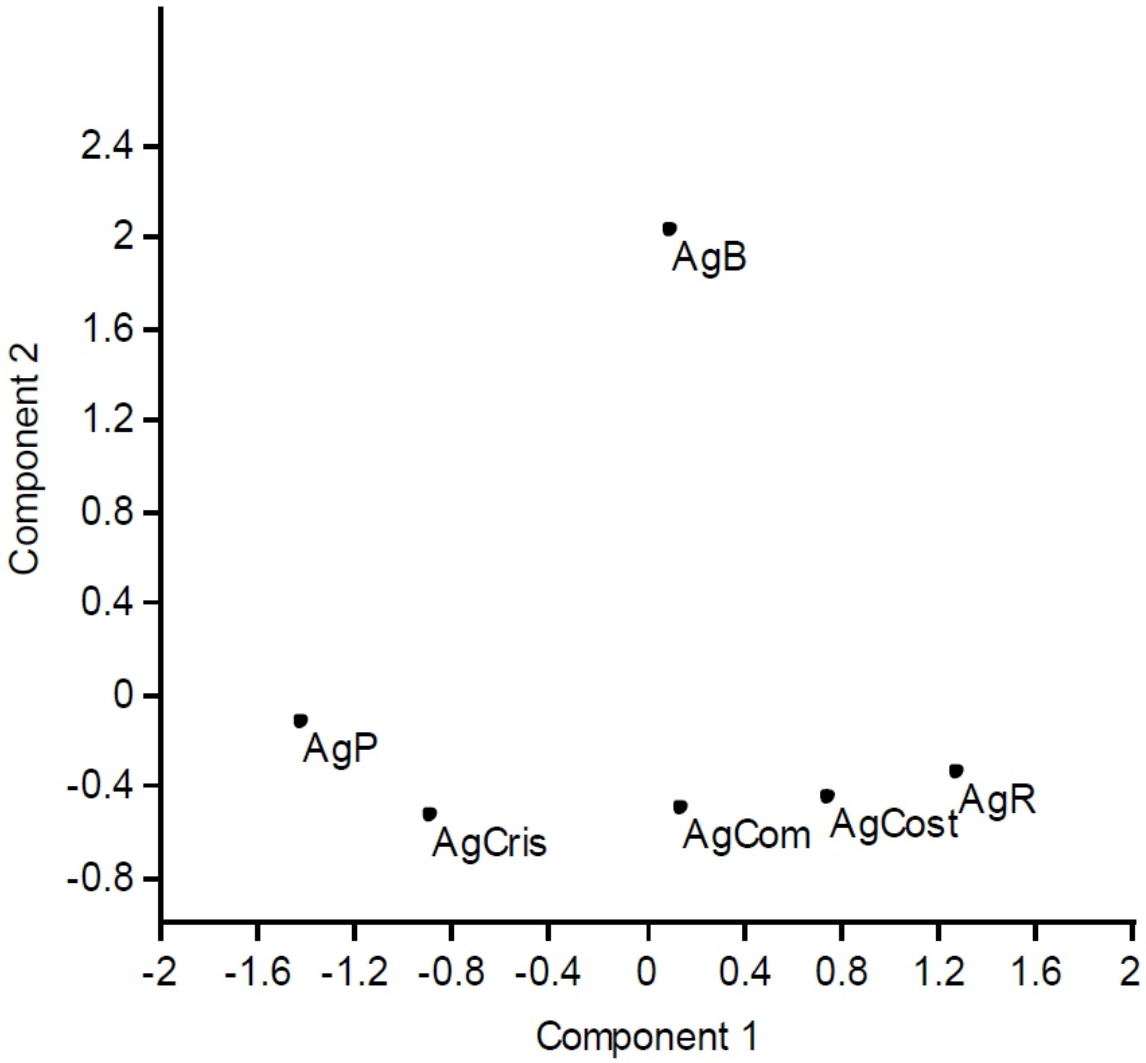


Figure 4: **Principal Components Analysis showing the relationship of *Aglaonema* species based on Six Foliar Quantitative Anatomical Characters.**

Legend: **AgB** – *Aglaonema brevispathum*; **AgCom** – *Aglaonema commutatum*; **AgCost** – *Aglaonema costatum*; **AgCris** – *Aglaonema crispum*; **AgP** – *Aglaonema pictum*; **AgR** – *Aglaonema rotundum*.

DISCUSSION

Similarities and differences occur among the six species of *Aglaonema* studied. The similarities affirm their generic grouping while the differences enhance their specificity. Epidermal cell area and stomata size are interestingly higher on the abaxial than the adaxial surfaces of all the species except in *A. brevispathum* where the adaxial values are higher than those of the abaxial. This can be used in delimiting the species of *Aglaonema*. The least epidermal cell size was encountered on the adaxial and abaxial surfaces of *A. rotundum* while the highest values were encountered on the adaxial and abaxial surfaces of *A. pictum*. Oladipo and Ayo-Ayinde (2014) delimited some species of *Aneilema* and *Commelina* using epidermal cell size.

The species studied can be separated based on the epidermal cell shape on their adaxial surfaces. Irregular epidermal cell shape on the adaxial surfaces of *A. costatum* and *A. pictum* delimits them from the other species. Anticlinal wall pattern is another character of taxonomic importance

among the species. Although the pattern on the abaxial surfaces of all the species is similar, they can still be separated based on the pattern on their adaxial surfaces. Sinuous anticlinal wall pattern clearly separates *A. costatum* from the other members of the genus in this study. Some species of the family Solanaceae have been separated based on anticlinal wall pattern (Adedeji *et al.,* 2007).

The hypostomatic nature of *A. rotundum* was revealed in this study as opposed to the amphistomatic nature of all the other species. This is quiet diagnostic. Brachyparacytic stomata complex is common to all the species studied. Saadu *et al.* (2009) reported brachyparacytic stomata for *Xanthosoma sagittifolium* and *Colocasia esculenta* which are also in the family Araceae. However, additional anisocytic and anomocytic stomata complexes were encountered in some of the species. Elliptic stomata shape unifies all the *Aglaonema* species in this study. Some occasional circular-shaped stomata were however encountered in *A. commutatum*, *A. costatum*, *A. crispum* and *A. pictum*.

Stomata size and index are quite diagnostic and of great taxonomic importance in plant species (Adedeji *et al.*, 2007 and Thair and Rajput, 2009). The highest stomata size is present on the adaxial and abaxial surfaces of *A. pictum* while least stomata size was encountered on the adaxial surface of *A. commutatum* and the abaxial surface of *A. costatum*. Stomata index was generally higher on the abaxial surfaces than on the adaxial surfaces of all the species in this study. This agrees with the findings of Osuji and Nwala (2015) in their work on some cultivars of *Xanthosoma* and *Colocasia*. The reason for the higher stomata index on the abaxial surfaces is probably to reduce transpiration rate from the surfaces of the leaves because they are shade-loving plants (Nicolson, 1969).

Aroids are known to contain calcium oxalic acid usually deposited as calcium oxalate crystals in any organ or tissue within the plant (Middendorf, 1982). The distribution of these crystals in the species studied is useful and relevant to their classification. They are encountered in the form of druses and raphides in just three of the six species studied, which are *A. commutatum*, *A. crispum* and *A. costatum*.

The cluster analysis separates the species into two main groups. *A. brevispathum* clearly separates from the other species in the first cluster. In the second cluster, *A. rotundum* separates from the other species while the third main cluster groups *A. commutatum* and *A. crispum* together on one side and *A. costatum* and *A. brevispathum* on the other side. This outcome is similar to the result of the Principal Components Analysis. The first three components accounted for 99.85 % of the total variation among the species. From the PCA loadings, the characters responsible for the separation of the species are the adaxial epidermal cell area and the abaxial epidermal cell area. Swanson (1968) ascertained that increase in cell size is the most immediate and universal effect of polyploidy. Polyploidy occurs in the genus *Aglaonema* as reported by Mayo *et al.*, 1997. A research into the ploidy levels of these species will therefore be needful so as to ascertain the evolutionary lineage or descent of the species.

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