

Epidermal studies of three species of *Vernonia* Schreb. in Southern Nigeria

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ABSTRACT

Kemka-Evans CI, Okoli B, Nwachukwu CU. 2014. Epidermal studies of three species of Vernonia Schreber in Southern Nigeria. Biodiversitas 15: 137-141. The leaf epidermal studies of three species of *Vernonia* namely *V. cinerea* (L) Less, *V. amygdalina* Delile. (bitter leaf and non-bitter leaf variety) and *V. conferta* Benth. were undertaken with the aim of revealing their foliar characters which will enhance their identification and determination of their taxonomic relationship. Matured leaves were soaked in Sodium Oxochlorate II for 24 hours to separate the epidermal surfaces. Data from the measurement of stomata and epidermal cells were analyzed. The presence of diagnostic characters such as contiguous stomata and sinuous anticlinal walls on the abaxial leaf surface of non-bitter variety of *V. amygdalina* and on both leaf surfaces (adaxial and abaxial) of *V. cinerea* are of taxonomic importance. The irregular T-shaped trichomes on the leaf surfaces of *V. amygdalina* (bitter leaf) and the cuticular striations on the adaxial surface of the same taxa could be used to delimit the taxa from the other species. The distribution of the stomata show hypoamphistomatic in all the three species studied. Anomocytic stomata occurred on all the taxa studied. Anisocytic stomata were found on the abaxial surface of *V. conferta*. These characters examined revealed interspecies relationship among the three species and also suggest that *V. amygdalina* (non bitter leaf) is a variety of *V. amygdalina* (bitter leaf) and should not be regarded as another species of *Vernonia*. The epidermal leaf characters of *V. amygdalina* (non-bitter leaf) is also been reported for the first time.

Key words: Epidermal, Southern Nigeria, *Vernonia*.

INTRODUCTION

Vernonia Schreber belongs to the tribe Vernonieae of the family Asteraceae (Compositae). The family Asteraceae is the largest family of the flowering plants, comprising 950 genera, and 23,000 species (Gills 1988). Olorode (1984) noted that the family Asteraceae possesses simple leaves with alternate or opposite leaf arrangement. Among the species found in Nigeria, *V. cinerea* (L) Less, *V. amygdalina* Delile and *V. conferta* Benth. form an interesting group to study because *V. cinerea* is a herbaceous weed while *V. amygdalina* is usually treated as a shrub and also occur as bitter leaf and non bitter leaf variety and *V. conferta* is a small tree. The existence of *V. amygdalina* in bitter leaf and non-bitter leaf form, sometimes poses a problem in classification of the species as *V. amygdalina* (non-bitter leaf) is usually regarded as a different species. The use of epidermal characters in general and those of trichomes in particular, have been widely recognized in angiosperm taxonomy. The taxonomic value of the leaf epidermal characters is well documented (Jayeola et al. 2001; Adedeji and Illoh 2004; Adedeji 2004). The epidermal characters in general and those of trichome in particular have been used by many researchers in the study of Angiosperm. Such workers include eight species of *Indigofera* (Leguminosae-Papilionideae) (Nwachukwu and Edeoga 2006), two

species of *Solanum* (Solanaceae) (Mbagwu et al. 2008), three species of *Boerhavia* (Nyctaginaceae) (Edeoga and Ikem 2001) and eight species of *Crassocephalum* (Asteraceae) (Kemka and Nwachukwu 2011). Oladele (1990) found the occurrence and morphology of the irregular T-shaped trichomes on *V. amygdalina* to be diagnostic. Adedeji and Jewoola (2008) noted that the epidermal cells of *V. cinerea* and *V. amygdalina* are slightly irregular to polygonal with wavy or undulating anticlinal walls on the adaxial surface and sinuous anticlinal walls on the abaxial surface. They also observed that the leaf surfaces are amphistomatic. The present work which is epidermal studies on the three species of the genus *Vernonia* is aimed at providing a more detailed information of the epidermal characters which could be used to delimit the species and also document the epidermal characters of *V. amygdalina* (non bitter leaf). It is also aimed to ascertain the taxonomic relationship of the three species considering the fact that they exhibit different habits of growth.

MATERIALS AND METHODS

Sources of plant specimens

The three *Vernonia* species studied namely *V. cinerea* (L) Less (Figure 3), *V. amygdalina* Delile (bitter leaf and

non-bitter leaf variety) (Figures 1 and 2) and *V. conferta* (Figure 4) were collected from Imo, Abia and Rivers, Southern Nigeria. The plant materials were grown at University of Port Harcourt, Rivers State, Nigeria. The specimens collected were authenticated by the curator/plant taxonomist at University of Port Harcourt, Rivers State, Nigeria and the voucher specimen deposited at the herbarium of the same university.

Epidermal studies

Epidermal peels of both adaxial and abaxial surfaces of the four different taxa made following a modified method of Metcalfe and Chalk (1979). The materials fixed in F.A.A. (Formalin acetic acid alcohol mixture) for 48 hours materials were rinsed with distilled water and soaked in commercial bleach (Sodium Oxochlorate (II) (NaClO)) for 24 hours to clear the epidermis and loosen the tissues, The two surfaces were carefully separated using a razor blade and finally stained with safranin-O. The epidermal strips were then mounted temporarily on clean slides in 50% glycerin and covered with cover slips. Photomicrographs were taken from good preparation using a Leitz-habolux – 12 – microscope fitted with WILD-Mps camera. The length and breadth of 30 stomata and epidermal cells were measured on both surfaces. Ten microscopic fields chosen at random were used for enumeration of number of stomata and epidermal cells appearing per field view. The stomatal index and frequencies were calculated according to the formula of Salisbury in Olorode (1990) using the formula $\text{Stomatal Index} = \frac{S}{E+S} \times 100$ where S = number of stomata per unit area, E = number of epidermal cells in the same area.

RESULTS AND DISCUSSION

The epidermal cell and stomatal characteristics of taxa investigated are shown in Tables 1 and 2. The epidermal cell shape is irregular in all the taxa. The anticlinal wall is straight – arcuate on both surfaces of *V. amygdalina* (bitter leaf: Figures 6 and 10), *V. conferta* (Figures 8 and 12) and the adaxial surface of *V. amygdalina* (non-bitter variety; Figure 7). Sinuous anticlinal walls occurred on both the abaxial and adaxial surfaces of *V. cinerea* (Figures 5 and 9) and the abaxial surface of *V. amygdalina* (non-bitter variety: Figure 11). The distribution of the stomata is hypoamphistomatic in all the species (stomata occurring on both the upper and lower surfaces of the taxa studied). Stomatal types are generally anomocytic (stoma lacks morphologically differentiated subsidiary cells) in both the adaxial and abaxial surfaces of all the species but contiguous stomata occurred on the abaxial surfaces of *V. cinerea* and *V. amygdalina* (non-bitter leaf Figures 9 and 11). Only *V. conferta* has anisocytic stomata (stoma surrounded by three subsidiary cells. figures 8 and 12). Radiating cuticular striations appeared on the adaxial surface of *V. amygdalina* (bitter leaf). The highest stomatal length 31.86 ± 1.10 occurred on *V. amygdalina* (non-bitter leaf) and the lowest 18.00 ± 1.90 on *V. conferta*. *Vernonia amygdalina* (bitter leaf) exhibited the highest stomatal width 24.82 ± 1.58 and the lowest 12.15 ± 1.30 occurred on *V. conferta*. T-shaped, short-stalked glandular trichomes were found on both surfaces of the leaves of all taxa and bilobed trichomes were common on the abaxial surface of the species studied. The T-shaped trichomes in *V. amygdalina* are peculiar to it (Figure 10).

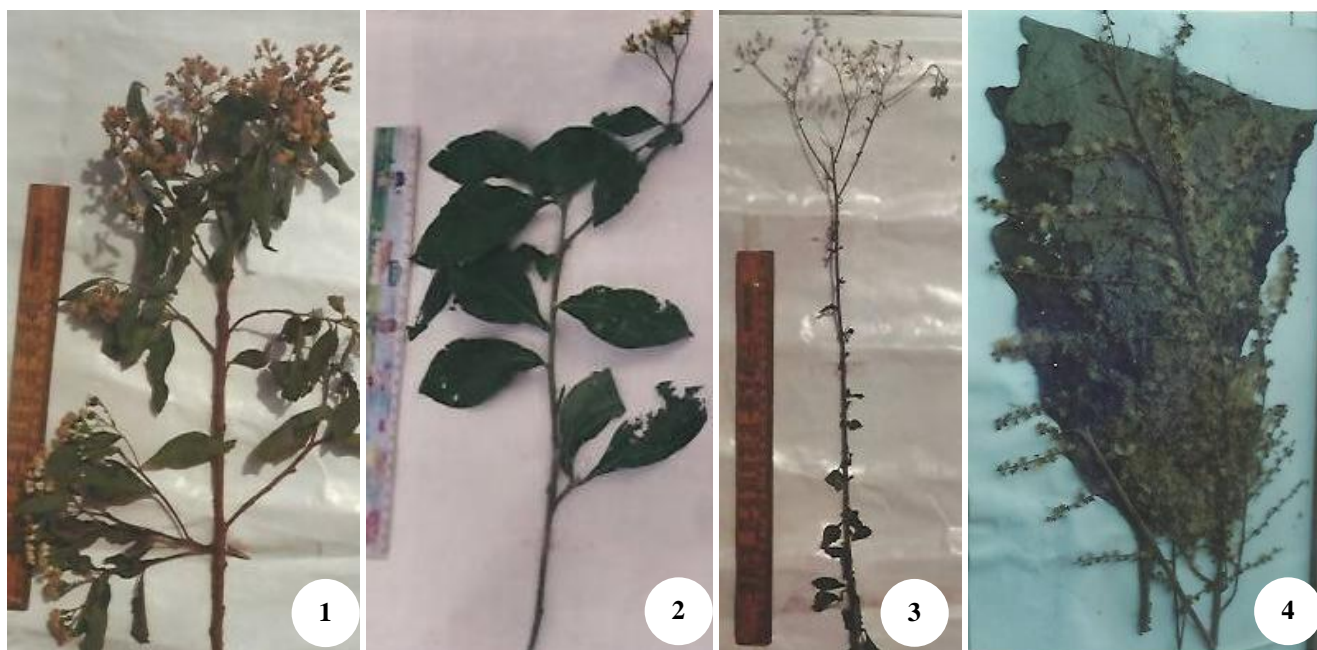


Figure 1. Morphological Features of *Vernonia amygdalina* (bitter leaf), showing leaves and inflorescence.

Figure 2. Morphological Features of *Vernonia amygdalina* (Non-bitter leaf), showing leaves and inflorescence.

Figure 3. Morphological Features of *Vernonia cinerea*, showing leaves and inflorescence.

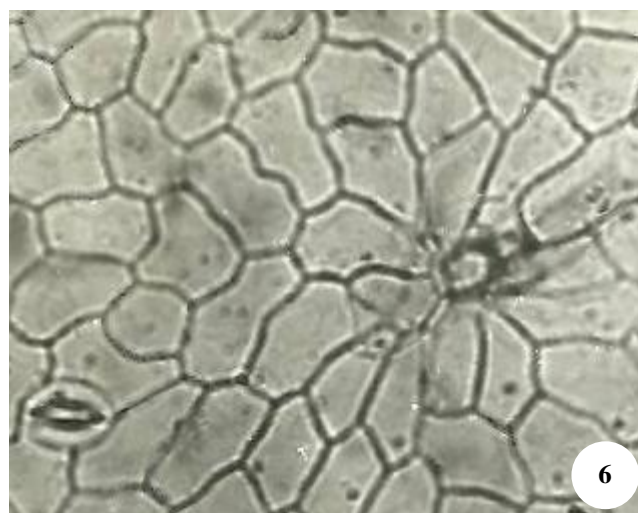
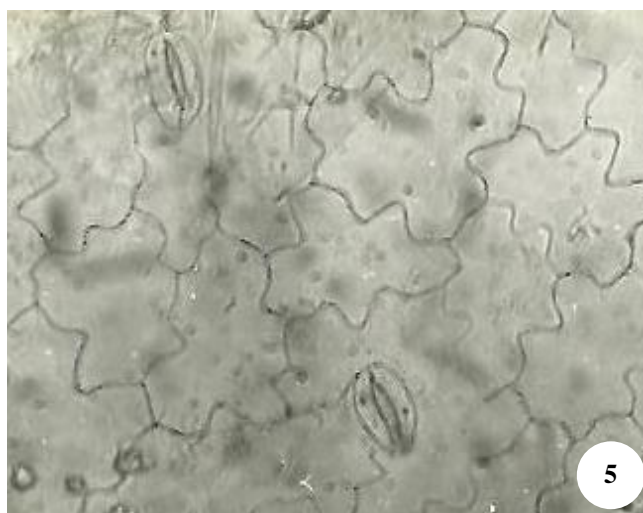
Figure 4. Morphological Features of *Vernonia conferta*, showing leaves and inflorescence.

Table 1. Epidermal cell characteristics of the *Vernonia* species studied

| Species | <i>V. cinerea</i> | | <i>V. amygdalina</i> (bitter-variety) | | <i>V. amygdalina</i> (non-bitter-variety) | | <i>V. conferta</i> | |
|-------------------------------------|-------------------|------------|------------------------------------------|------------|----------------------------------------------|------------|--------------------|------------------|
| | Adaxial | Abaxial | Adaxial | Abaxial | Adaxial | Abaxial | Adaxial | Abaxial |
| Character | Irregular | Irregular | Irregular | Irregular | Irregular | Irregular | Irregular | Irregular |
| Epidermal cell shape | Irregular | Irregular | Irregular | Irregular | Irregular | Irregular | Irregular | Irregular |
| Anticlinal cell pattern | Straight-arcuate | Sinuuous | Straight-arcuate | Sinuuous | Sinuuous | Sinuuous | Straight-arcuate | Straight-arcuate |
| Epidermal cell length (nm) | 56.97±6.74 | 39.81±3.50 | 55.53±1.51 | 41.22±1.85 | 48.40±4.41 | 41.49±6.71 | 36.99±1.84 | 32.04±3.45 |
| Epidermal cell width (nm) | 31.68±5.07 | 29.25±1.22 | 29.07±2.05 | 15.84±0.93 | 20.78±6.90 | 31.41±1.20 | 31.05±3.47 | 19.08±0.69 |
| Number of epidermal cells per sq mm | 5890 | 5300 | 2572 | 1088 | 2750 | 560 | 5334 | 2900 |
| Co-efficient of variation | 11.83 | 9.004.19 | 2.727.05 | 9.335.89 | 2.727.07 | 16.33731 | 5.0411.17 | 10.783.5 |

Table 2. Stomatal characteristics and trichomes of *Vernonia* species studied

| Species | <i>V. cinerea</i> | | <i>V. amygdalina</i> (bitter-variety) | | <i>V. amygdalina</i> (non-bitter-variety) | | <i>V. conferta</i> | |
|---------------------------|----------------------------|----------------------------|------------------------------------------|-------------------------------|----------------------------------------------|---------------------------|-----------------------------|-----------------------------|
| | Adaxial | Abaxial | Adaxial | Abaxial | Adaxial | Abaxial | Adaxial | Abaxial |
| Stomatal type | Anomocytic and contiguous | Contiguous | Anomocytic | Anomocytic | Anomocytic | Anomocytic and contiguous | Anomocytic | Anomocytic |
| Stomatal length | 27.18±4.39 | 27.90±1.29 | 27.45±0.40 | 27.63±1.36 | 26.61±1.32s | 31.86±1.10 | 26.46±1.64 | 18.00±1.90 |
| Stomatal width(nm) | 23.22±2.89 | 16.65±1.25 | 24.82±1.58 | 13.77±0.82 | 19.08±3.68 | 20.97±1.5 | 15.75±1.74 | 12.15±1.30 |
| Co-efficient of variation | 16.07- 12.45 | 4.64- 7.5 | 1.45- 6.36 | 4.92- 5.95 | 4.62- 19.28 | 3.45- 7.15 | 6.19- 11.10 | 10.6- 1128 |
| Stomatal index (%) | 9.84 | 34.88 | 4.84 | 5.35 | 10.44 | 21.61 | 5.32 | 9.38 |
| Stomatal length:width | 1.17 | 1.68 | 1.11 | 2.01 | 1.39 | 1.51 | 1.68 | 1.48 |
| Trichomes | Bilobed glandular trichome | Bilobed glandular trichome | Irregular shaped glandular | T- Irregular shaped glandular | T- T-shaped glandular | Bilobed glandular | T-shaped glandular trichome | T-shaped glandular trichome |



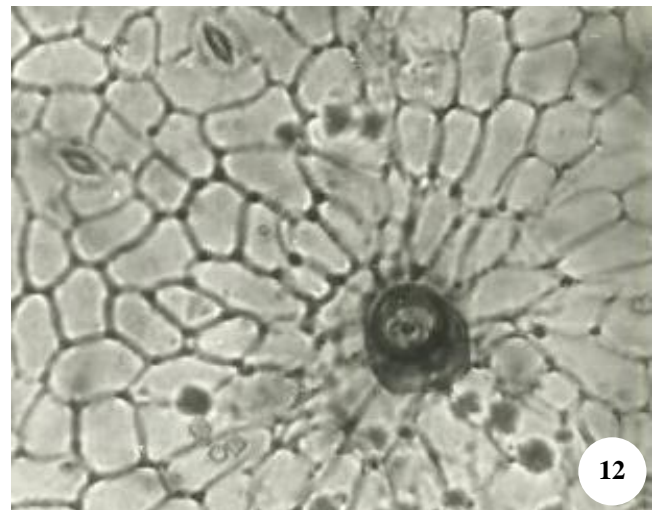
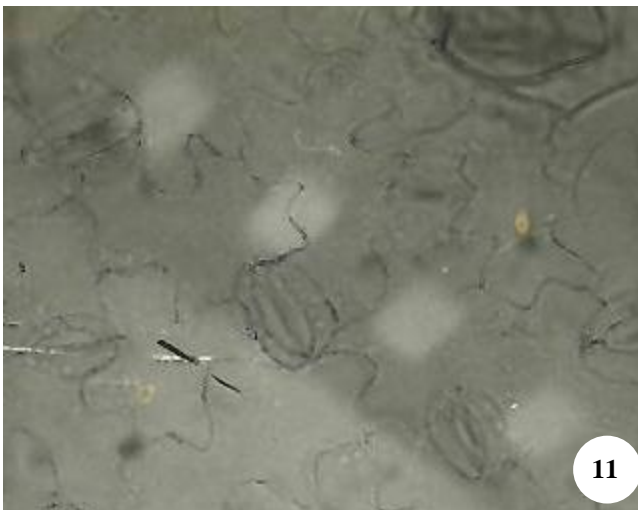
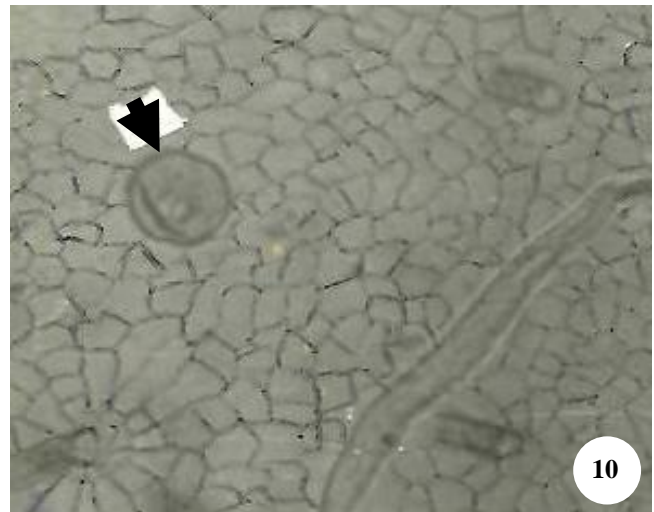
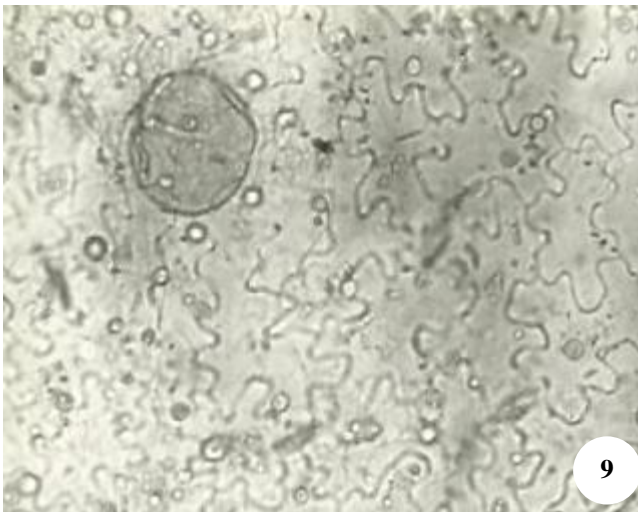
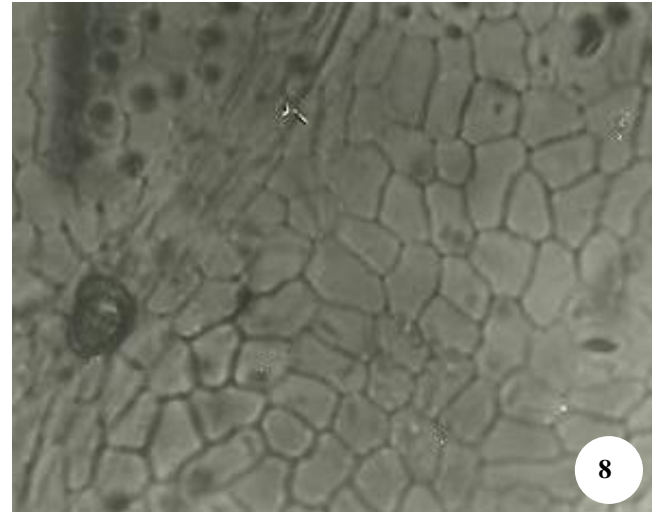
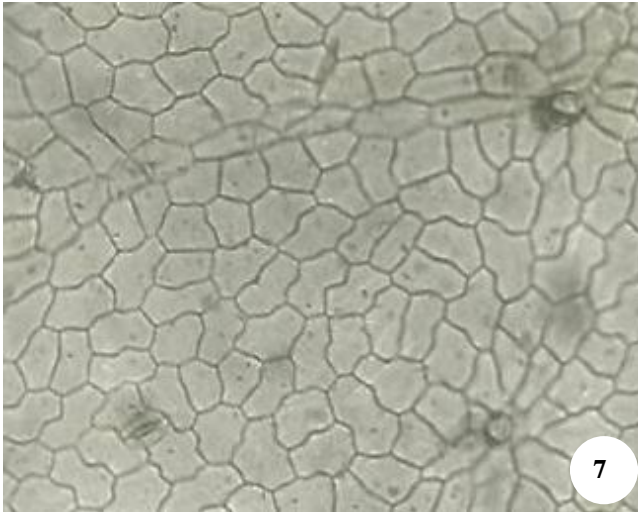


Figure 5. Adaxial epidermis of *Vernonia cinerea* showing sinuous anticlinal walls and anomocytic stomata. X 250

Figure 6. Adaxial epidermis of *Vernonia amygdalina* (bitter leaf) showing straight-arcuate. X 250

Figure 7. Adaxial epidermis of *Vernonia amygdalina* (non-bitter leaf) showing anomocytic stomata. X 150

Figure 8. Adaxial epidermis of *Vernonia conferta*. showing straight-arcuate anticlinal Walls. 150

Figure 9. Abaxial epidermis of *Vernonia cinerea* showing contiguous stomata and sinuous anticlinal walls. X 250

Figure 10. Abaxial epidermis of *Vernonia amygdalina* (bitter variety) arrows show glandular trichomes. X 40

Figure 11. Abaxial epidermis of *Vernonia amygdalina* (non-bitter variety) showing contiguous stomata. X 250

Figure 12. Abaxial epidermis of *Vernonia conferta* showing anisocytic stomata. X 250

The anatomical evidence has provided useful information in the characterization of *V. amygdalina*, *V. cinerea* and *V. conferta*. The epidermal cell shape is irregular in all the taxa. The highest epidermal cell length 56.97 ± 6.74 was observed in *V. amygdalina* (bitter leaf) and the lowest 32.04 ± 3.45 in *V. conferta*. The highest epidermal cell width 31.68 ± 5.07 was also observed in *V. amygdalina* (bitter leaf) and the lowest 19.08 ± 0.69 in *V. conferta*. The anticlinal walls varied from straight–arcuate to sinuous. The sinuous anticlinal walls on the abaxial surface of *V. amygdalina* (non-bitter leaf) could be used to distinguish it from *V. amygdalina* (bitter leaf). The occurrence of the sinuous anticlinal walls and both surfaces of *V. cinerea* could be used to delimit it from the other species. The distribution of stomata is hypoamphistomatic in all the species. The highest stomatal length 31.86 ± 1.10 was recorded on *V. amygdalina* (non-bitter leaf) and the lowest 18.00 ± 1.90 on *V. conferta*. *V. amygdalina* (bitter leaf) exhibited the highest stomatal width 24.82 ± 1.58 and the lowest 12.15 ± 1.30 occurred on *V. conferta*. The presence of bilobed glandular trichomes in all the taxa suggests that the species are related. However the occurrences of irregular T-shaped trichomes in *V. amygdalina* (bitter leaf) agree with the work of Oladele (1990) who found out those irregular T-shaped trichomes could be of diagnostic importance in the three *Vernonia* species studied. The occurrence of anomocytic stomata on all the taxa studied suggests that the species are related. Anisocytic stomata found on the abaxial surface of *V. conferta* are of taxonomic importance. Okoli (1987) found contiguous stomata and cuticular striations to be of useful diagnostic feature on the leaf epidermis of *Telfairia occidentalis* Hook F. The contiguous stomata on the non – bitter leaf of *V. amygdalina* and also *V. cinerea* could be used to delimit the taxa. The presence of cuticular striations on the adaxial surface of *V. amygdalina* (bitter leaf) is also of diagnostic value in delimiting the taxon. The variation in the duration and habit of the various species of *V. amygdalina* is of taxonomic and horticultural importance. *Vernonia amygdalina* (non-bitter variety) which is a perennial plant can be propagated through seedlings and has less longevity compared to that of *V. amygdalina* (bitter leaf). The non – bitter variety also does not need several washing in water to remove the bitter taste before it is used for soup. The two taxa of *V. amygdalina* could be hybridized so as to extend the productivity and longevity of the hybrid. The work through the micro morphological features studied reveal that the three species are related

irrespective of the differences in their habit. The observed micromorphological difference such as the shape of trichomes conform with some classification systems, whereby *V. cinerea* has consistently been put in the section Tephrodes while *V. amygdalina* and *V. conferta* have different times, been put in the section strobocalyx (Isawumi 1993).

CONCLUSION

The overall results from the study show that epidermal characters are of taxonomic importance in the classification and delimitation of the four taxa among the three species of *Vernonia*.

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