Pak. J. Bot., 57(3): 1065-1074, 2025. DOI: http://dx.doi.org/10.30848/PJB2025-3(10)

TAXONOMIC AND ECOLOGICAL SIGNIFICANCE OF FOLIAR EPIDERMAL CHARACTERS IN THREE TAXA OF EUPHORBIACEAE IN NIGERIA

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Abstract

Foliar epidermal features of three taxa of the Euphorbiaceae: *Bridelia ferruginea* Benth., *Hura crepitans* L. and *Ricinodendron heudelotti* (Baill.) Pierre ex Heckle in Nigeria were studied to extract their taxonomic and ecological significance. An ecological survey design was used in the study of the three plant taxa from three locations with ten replicas each from each location. The study was carried out in the Department of Plant Science and Biotechnology, University of Nigeria, Nsukka between January and December. Leaves were plucked from freshly collected twigs from the Tropical Rainforest Vegetation (TRFV), Derived Savannah Vegetation (DSV) and Guinea Savannah Vegetation (GSV) within the study period. Anatomical slides were prepared using standard procedures (i.e. clearing method) and observed by light microscopy. The stomata sizes, density and indices significantly differed at $p>0.05$. The leaves of *B. ferruginea* were hypostomatic and the stomatal type was anomocytic while *H. crepitans* and *R. heudelotti* were amphistomatic, sharing anomocytic type of stomata in common. Paracytic types were also observed in the abaxial (lower) leaf surface of *H. crepitians*; polygonal epidermal cells on both surfaces with scanty uniseriate trichomes. Anisocytic types were also observed in abaxial leaf surface in *R. heudelotti*, irregular and variously elongated epidermal cells on adaxial (upper) and abaxial surfaces respectively which were glabrous. The highest stomatal density (166.18 ± 3.70 µm) on the abaxial leaf surface was recorded in *R. heudelotti* from DSV, while *B. ferruginea* from GSV recorded the lowest (100.00 ± 2.40 µm). Uniseriate glandular trichomes were in abundance in *B. ferruginea* and scanty in *H. crepitans*. Epidermal features recorded in this study could be utilized in the delimitation of the plant taxa. The occurrence of smaller apertural and stomatal density in samples from more xeric zones, no doubt justifies the low rates of transpiration found in these taxa.

Key words: Leaf epidermal characters; Stomata; Taxonomy; Ecology; Trichome; Euphorbiaceae; Nigeria.

Introduction

The three genera: Bridelia ferruginea Benth. Hura crepitans L. and Ricinodendron heudelotti (Baill.) Pierre ex Heckle belongs to the Euphorbiaceae (Keay, 1989; Adesina & Akomolafe, 2014; Vassallo *et al*., 2020; Yeboah *et al*., 2022; Yembeau *et al*., 2022). However, the tribe Bridelieae placed in the fasciculate clade of the new family segregate, Phyllanthaceae of Euphorbiaceae and its members including *B. ferruginea* possess axillary, fasciculate clusters of flowers and absence of tannins in the epidermal cells in their leaves (Wurdark *et al*., 2004; Bouman *et al*., 2020). Angiosperm Phylogeny Group also recognized Phyllanthaceae as one of the five segregates of Euphorbiaceae sensu lato (APG II, 2003; Bouman *et al*., 2020). The Euphorbiaceae has about 322 genera and 8910 species found in varied habitats, in many areas of the tropical and subtropical regions (Webster, 1987; Thakur & Patil, 2011; Bahadur *et al*., 2022). It consists of trees, shrubs, but are rarely woody climbers and is characterized as follows: flower hypogynous, actinomorphic, mostly unisexual; perianth rarely double; usually simple; androecium 1-∞ ovary of 3 carpels, trilocular with 1-2 suspended ovules in each cell; micropyle directed upwards and outwards and covered with a fleshy outgrowth (caruncle). Fruit almost invariably a schizocarp-capsule, splitting into carpels (Judd *et al*., 2007; Takhtajan, 2009; Yakoub-Zokian, 2011).

Bridelia ferruginea is the most available Bridelia species of the savannah woodland region of Africa, distributed across Tropical Africa and Asia (PROTA, 2007; Jonathan *et al*., 2014; Afolayan *et al*., 2019). It is a small non-laticiferous tree or shrub with a wide range of economic values which include medicinal, ecological and industrial utilization of its wood. The leaves, fruits, roots and stembarks have antimicrobial, laxative, anti-diabetic, anti-oxidant and anti-inflammatory activities (Adebayo & Ishola, 2009; Kayode and Jose; 2009; Omotade & Seun, 2012; Taiwo *et al*., 2012; Afolayan *et al*., 2019). It has also been reported that the bark extract has the potential for water treatment (Kolawole & Olayemi, 2003; Kolawole *et al*., 2007; Yeboah *et al*., 2022). *B. ferruginea* is monoecious, ebracteate and the staminate flowers are pedicellate and puberulous with polyandrous stamens. The pistillate flower is subsessile with 2 locular ovaries and the fruit is a drupe (Keay, 1989).

The genus Hura L. is represented by two species, *Hura crepitans* L. introduced to tropical regions of Africa and Asia and *Hura polyandra* Bail., restricted to South America (Kadiri & Adeniran, 2016). The tree is planted as a shade and ornamentals in villages and towns and the seeds yield non-edible oil used in the production of biodiesel and soap. Oleic acid is the major fatty acid and because of its ratio to linoleic acid, it is used in various industries for possessing long shelf-life. (Abdulkadir *et al*., 2013). *H. crepitans* are monoecious, with aculeate stem and branches; monochlamydeous staminate flowers, bracteates, multilocular ovary and dehiscence explosive fruits (Keay, 1989; Oliveria *et al*., 2013). Medicinal properties of *Hura crepitans* have been reported by Vassallo *et al*., (2020).

Ricinodendron heudelotii is characteristic of equatorial hot and secondary Rainforest in the tropical zone of Africa. The generic name is based on the Greek word for tick and tree because the seeds were thought to resemble ticks (Tchoundjeu

& Atangana, 2006). The tree, which is sometimes buttressed, starts bearing fruits at 7 to 10 years of age in open light spaces (Shiembo *et al*., 1997). With regards to Ricinodendron and intra-specific taxa, it is generally agreed that it comprises a single species (Tchoundjeu & Atangana, 2007). However, ICRAF (2004) noted two varieties: var. heudelotii and var. africanum, which other researchers named sub-species. Subspecies heudelotti has 3-5 leaflets per leaf, mostly 3 lobed fruits whereas africanum has (3-) 5-7 (-8) leaflets per leaf and 2-lobed fruits and are more common (Tchoundjeu & Atangana, 2007). The plant is distinctively valued for its seeds, which are used as spice and thickening agents in foods in Nigeria. Mapongmetsem & Tchiegang (1996) opined those soaps, cooking oil, pharmaceuticals and margarine could be produced from the fruits because of their high oil content and could also be used in cosmetics and paint production because of high values of saponin and iodine (Ekam, 2003). *R. heudelotti* is deciduous, dioecious, bracteates with pentamerous calyx and corolla; polyandrous stamens; ovary 2-locular and hypogynous (Keay, 1989).

The use of data generated from leaf epidermal surfaces in resolving the taxonomy of taxa has gained much recognition for a very long time (Aworinde *et al*., 2009). It was reported by Evert (2006) that different developmental sequence usually results in different configurations of stomatal complexes, which may occur only on one surface, either the upper (epistomatic) or more commonly on the lower (hypostomatic) or both surfaces (amphistomatic). There are variations of stomatal types in vascular plants, for example, diacytic type, paracytic type, anisocytic type and anomocytic type, commonly found in Euphorbiaceae, cyclocytic, parallelocytic type and actinocytic (Evert, 2006; Raju & Rao, 2008). Aworinde *et al*., (2009) stated that foliar epidermal patterns could serve as important tools in the taxonomic study of various plant taxa. Since stomata are homologous structures and of universal occurrence in plants, systematic studies of their types, distribution and ontogeny are of great relevance in taxonomy and phylogenetic inference could be made from such studies (Metcalfe & Chalk, 1979; Rasmusen, 1981).

Nigeria is located between Latitude 4º and 14º N of Longitudes 2º and 15º E, having Rainforest, Savannah and Montane as three major vegetation zones. The three locations for the collection of plant samples were: Obudu in Cross-River State (Latitude 6º67: N and Longitude 9º16′E), TRFV; Nsukka in Enugu State (Latitude 6º86′N and Longitude 7º39′E), DSV and Suleja in Niger State (Latitude 9º29′N and Longitude 7º17′E), GSV (Iloeje, 1999). The average annual rainfall in Obudu is 1,873 mm; 1,579 mm in Nsukka and 1,328 mm in Suleja, while the mean annual temperature in Obudu is 26.4ºC, varying by 3.5ºC throughout the year; 25ºC to 27ºC in Nsukka and 26.3ºC in Suleja, varying by 4.5ºC throughout the year (Climate data.org, 2019).

The quantitative leaf epidermal features, particularly, the stomatal parameters have proved useful in plant taxa delineation and the most widely utilized parameters are stomatal number, stomatal size and stomatal index. This work was aimed at presenting more precise characters of leaf epidermal features of the three taxa of the Euphorbiaceae and also to assess the impact of climatic factors on them across the three ecological zones.

Material and Method

Study locations: This study was conducted in the Plant Taxonomy/Anatomy Laboratory, Department of Plant Science and Biotechnology, University of Nigeria, Nsukka. The field collection and experiments were carried out between January and December 2019. Leaves of *B. ferruginea*, *H. crepitans* and *R. heudelotii* were freshly collected from the plant taxa growing in their natural regions of provenance. The collections were made from Obudu; Nsukka and Suleja representing three ecological zones of Tropical Rainforest Vegetation, Derived Savannah Vegetation and Guinea Savannah Vegetation in Nigeria respectively. Identification of the exomorphological features of the three taxa was observed and recorded in the field. The specimens were identified and authenticated by Prof. Dr. M. O. Nwosu of the Department of Plant Science and Biotechnology, University of Nigeria, Nsukka and also by comparing them with the Type specimens accessed from the Royal Botanic Gardens, Kew, England.

Leaf epidermal structures: Micromorphological studies of the foliar epidermises were carried out by clearing method as described by Ogbonna *et al*., (2018). Fresh leaves from the three taxa were soaked in commercial bleach (3.5% sodium hypochlorite) for about 12-20 hours depending on the nature of the samples. The leaves were rinsed two to three times with water. A sharp razor blade was used in scraping the leaf epidermises, which were rinsed in tap water using a camel's hairbrush. Samples of both upper (adaxial) and lower (abaxial) leaf surfaces were mounted on glass slides and stained with two to three drops of Safranin solution. They were mounted in glycerine and covered with coverslips. To prevent desiccation, a nail hardener was used to seal the edges of the coverslips.

Microscopy: The mounted strips were viewed at x40, x100 and x400 magnifications using a light Olympus microscope (Japan CX31) and observations were made on both qualitative and quantitative micro-morphological leaf epidermal variables. Photomicrographs were taken from good preparations with a Motic B3 compound Microscope (Carlsbad, USA), to which a Moticam 2.0 image system and software were attached. The means of quantitative parameters were worked out from 10 observations made from each of the samples following that of Aworinde *et al*., (2009). The following observations and assessments were made:

(a) The type and number of epidermal cells were counted and recorded.
(b) The stomatal complex types were recorded.
(c) The stomatal length and breadth were measured in a total of 10 fields of view for each sample using the Moticam 2.0 image system and software and their areas worked out.
(d) The stomatal densities were determined by the number of stomata per field of view.
(e) The stomatal index (SIs) were determined as follows;

$$Stomatal\ Index\ (\%) = \frac{Sn}{Sn + En} \times 100$$

Where: Sn represents the number of stomata in a field of view and En represents the number of epidermal cells in the same field of view.

Statistical analysis

The data collected from the study were used to quantify the relationship between the three taxa and the ecological significance of the three study locations. The collated data were subjected to one-way Analysis of Variance (ANOVA) and to Least Significance Differences (LSD) to compare their means and for multiple separations of means respectively to determine their differences. Means were tested for significance at $p \leq 0.05$.

Results

Leaf exomorphology: The leaves were simple in *B. ferruginea* and *H. crepitans*, digitate in *R. heudelotii*; petiolate, alternate, with reticulate pinnate venation (Plate 1); broadly elliptic lamina in *B. ferruginea*, cordate in *H. crepitans* while the leaflets of *R. heudelotii* were obovate-to-lanceolate (Plate 2). The leaf margin of *B. ferruginea* is undulate and the base is rounded with abruptly acuminate apex; *H. crepitans* has serrate to the entire margin; cordate base and acuminate apex while *R. heudelotii* has entire margin, cuneate base and attenuate apex (Plate 2). Prominent stipules, fan-like in shape with toothed margin were present at the leaf bases of *R. heudelotii*, particularly in young stems and branches (Plate 1d). The adaxial and abaxial surfaces of the three taxa were light and dark green respectively, pubescent in *B. ferruginea* and *H. crepitans* and glabrous in *R. heudelotii.*

Epidermal cells: The shapes of the epidermal cells were polygonal and irregular in the adaxial leaf surface of the three taxa. On their abaxial leaf surfaces, *B. ferruginea* and *H. crepitans* were polygonal in outline and B. heudelotii was conspicuously elongated variously. The anticlinal cell walls in *B. ferruginea* and *H. crepitans* were thicker on their adaxial leaf surfaces than on the abaxial and less undulating (Plates 3 and 4) while that of *R. heudelotii* were thin on both the adaxial and abaxial leaf surfaces and undulating (Plate 5). The adaxial leaf surfaces, *B. ferruginea* recorded the highest epidermal cell number (108.00 ± 0.71; 113.75 ± 1.38 and 91.50 ± 1.71 µm) across the three ecological zones (Table 3).

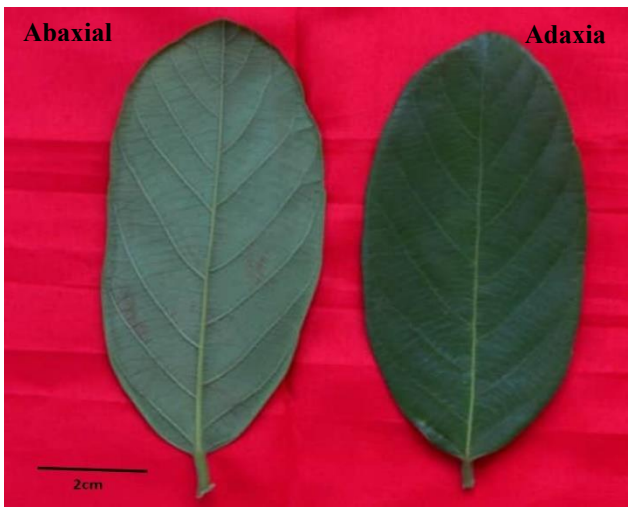
a: A twig of *B. ferruginea*

c: A twig of *R. heudelotii*

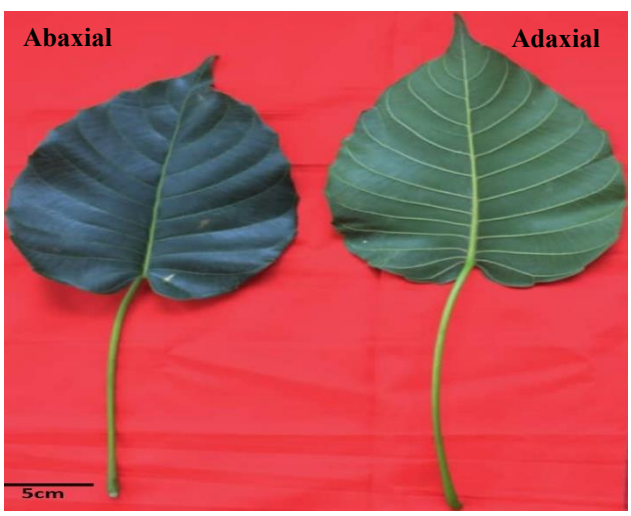
b: A twig of *H. crepitans*

d: A young twig of *R. heudelotii* showing prominent stipules (x $^1/_4$)

Plates 1 a-d: Photographs of young twigs of *B. ferruginea, H. crepitans, R. heudelotii and* a young twig of *R. heudelotii* showing alternate leaf arrangement, prominent stipules (fan-shaped).



a: Adaxial and abaxial leaf surfaces of *B. ferruginea* showing broadly elliptic lamina, rounded base and abruptly acuminate apex.



b: Adaxial and abaxial leaf surfaces of *H. crepitans* showing cordate lamina and base and acuminate apex.

c: Adaxial and abaxial leaf surfaces of *R. heudelotii* showing obovate-to-lanceolate leaflets with cuneate base and attenuate apex.

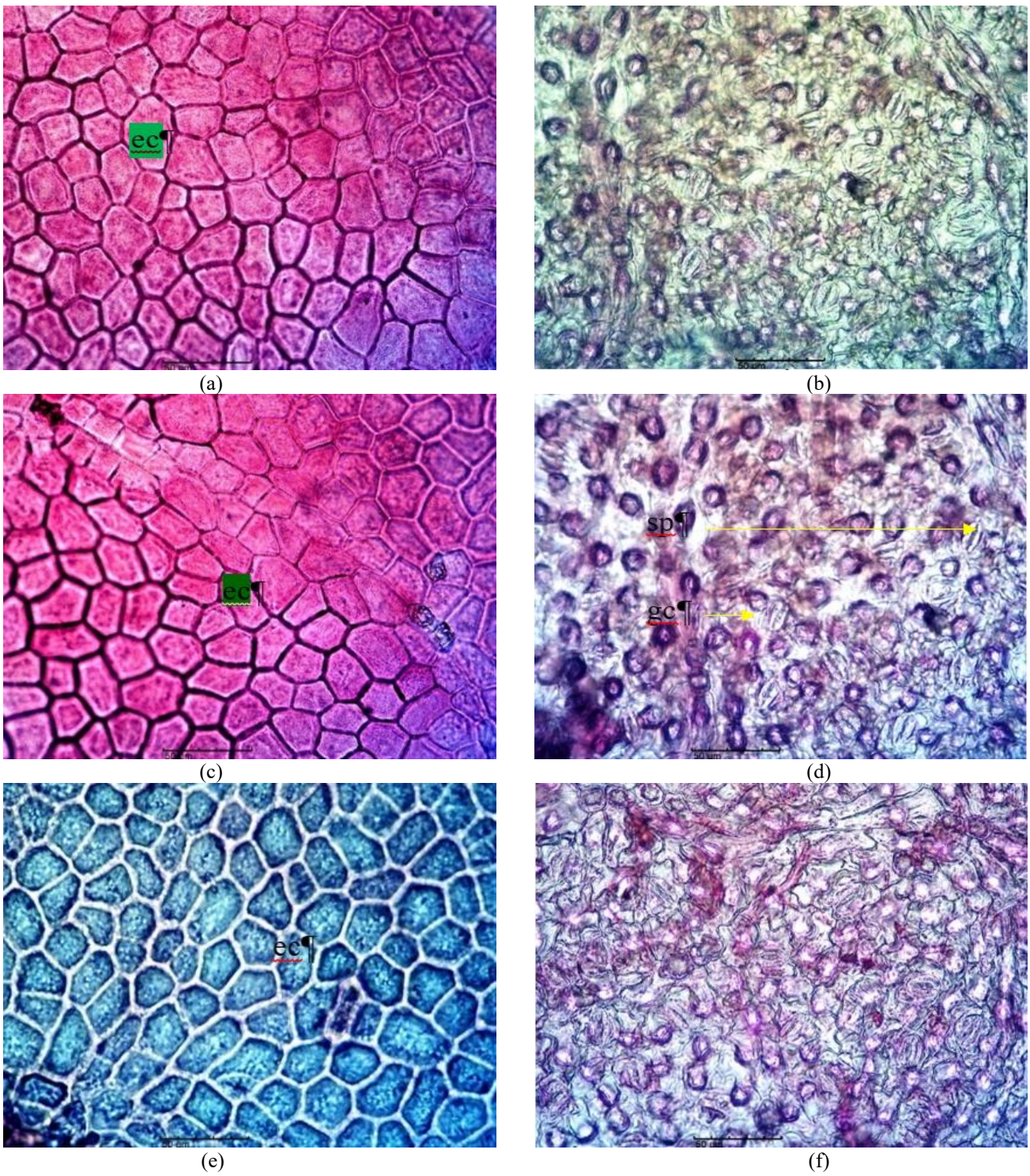
Plates 2a-c: Adaxial and abaxial leaf surfaces of *B. ferruginea, H. crepitans,* and *R. heudelotii.*

Stomata features: The result of the stomatal anatomy of the three taxa showed that the leaves of *B. ferruginea* were hypostomatic (i.e. stomata occur only on the lower surfaces), showing anisocytic type of stomata on the abaxial leaf surface and the absence of stomata on the adaxial surfaces. In *H. crepitans* and *R. heudelotii*, leaves were amphistomatic (i.e. stomata occur on both the upper and lower surfaces of the leaf), showing anomocytic stomata on the abaxial and adaxial surfaces of their leaves. In addition, paracytic and anisocytic stomata were also shown on the abaxial surface of *H. crepitans* and *R. heudelotii* (Plates 4 and 5; Table 1). The stomatal features of the three taxa varied greatly and were significantly different (p<0.05) for all the parameters measured (Table 2 and 3). Rossette crystals were present on the abaxial and adaxial leaf surfaces of the three taxa. The highest stomatal density (166.18 ± 3.70) was recorded in the abaxial leaf surfaces of R. heudelotii from Derived Savannah, which also had the highest in the two remaining ecological zones. The Guinea Savannah recorded the least stomatal density for the three taxa (Table 3). The abaxial leaf surfaces in *B. ferruginea* recorded the highest stomatal indices (61.40 ± 0.53; 65.59 ± 0.74 and 52.41 ± 0.34 µm) across the three ecological zones (Table 3).

Discussion

Foliar epidermal features from the three taxa showed that the shapes and nature of the anticlinal cell walls were highly variable in their outlines. The epidermal cell walls were irregular and polygonal on the adaxial leaf surfaces on the three taxa and their abaxial leaf surfaces. In *B. ferruginea* and *H. crepitans*, epidermal cells were predominantly polygonal and variously elongated in *R. heudelotii*. These findings were in agreement with those of Ahmed *et al*., (2009), who reported that among various taxa and even within the same taxon, the shapes of the epidermal cells and anticlinal cell wall outlines vary greatly. Irregular, polygonal or variously elongated epidermal cells were also reported by Raju & Rao (2008), Kadiri *et al*., (2009) and Thakur & Patil (2011) in their studies of variations in the development and structure of stomata in the Euphorbiaceae.

Uniseriate glandular trichomes were in abundance in *B. ferruginea*, scanty in *H. crepitans* and absent in *R. heudelotii*. The above features could be utilized in the delimitation of the taxa. This feature was also observed by Pandey (2004), who reported that members of a particular family may be identified by the occurrence of one or more types or by the presence or complete absence of one type of trichome or the other. On the other hand, the qualitative leaf micromorphological features of the three taxa were virtually similar across the three ecological zones inferring that leaf epidermal cell features were not significantly affected by the environment. It could also be inferred that since the above qualitative morphological features were shared by the three taxa across the three ecological zones, these features are predetermined genetically and are taxonomically very useful.

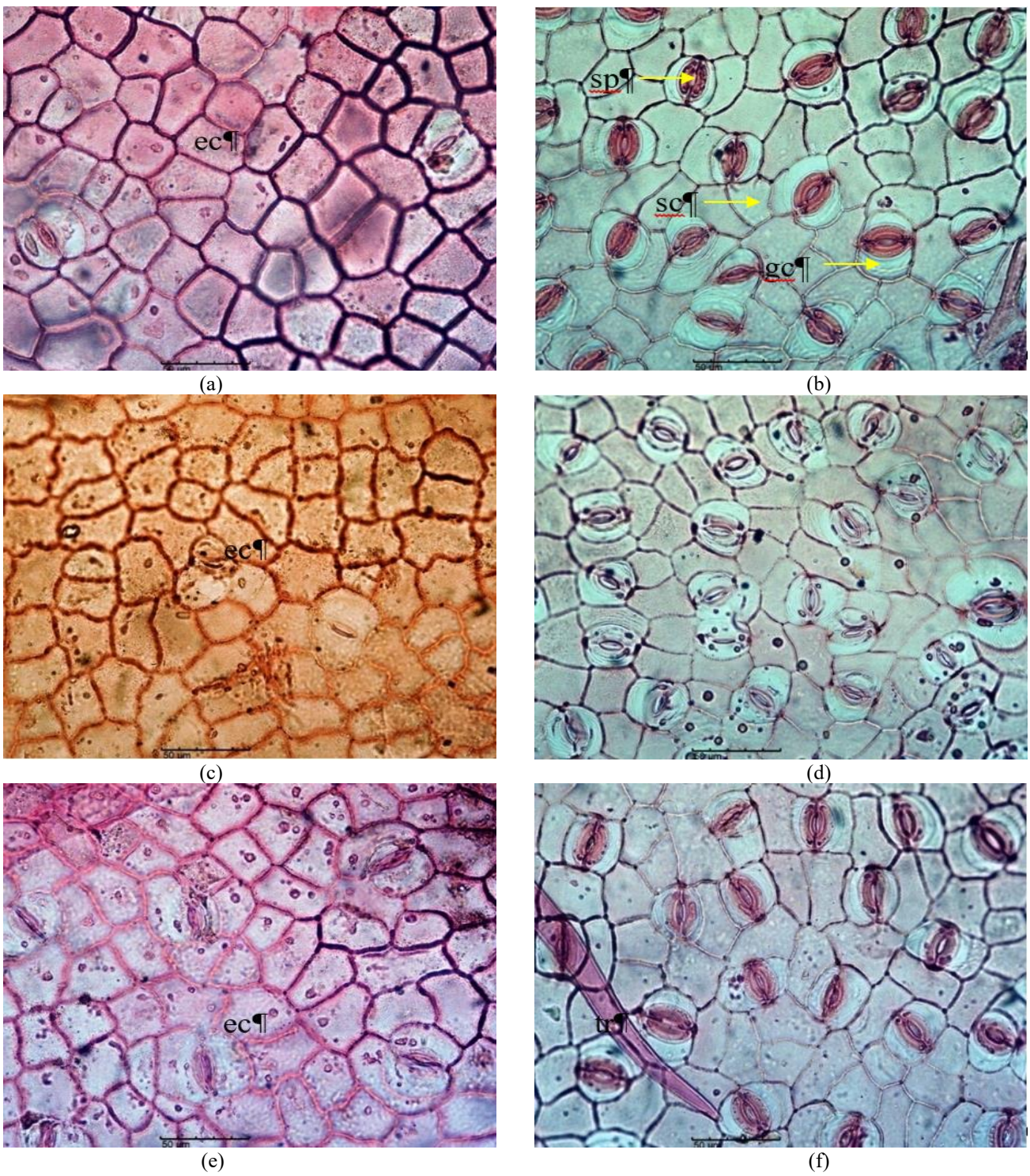


Plates 3a-f: Adaxial and abaxial surfaces of the leaf of *Bridelia ferruginea* showing anisocytic stomata (x 400) a and b = TRFV; c and d = DSV; e and f = GSV. ec = epidermal cell; sp = stomata pore; gc guard cell.

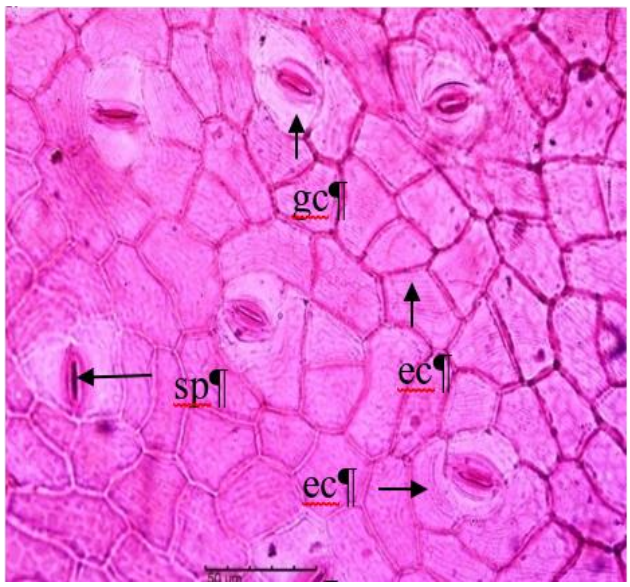
The use of data generated from leaf epidermal surfaces in resolving the taxonomy of taxa has gained much recognition for a long time (Aworinde *et al*., 2009). It was reported in Evert (2006) that different development sequences usually result in different configurations of stomatal complexes and that in leaves, stomata may occur on both surfaces (amphistomatic leaf) or only one, either the upper (epistomatic leaf) or more commonly on the lower (hypostomatic leaf). The leaves of *B. ferruginea* were found to be hypostomatic while those of *H. crepitans* and R. heudelotii were amphistomatic. The appearance of stomata on the abaxial leaf surfaces only in *B. ferruginea* in this work goes further to support the recent review and splitting of Euphorbiaceae sensu lato into five families: Pandaceae, Picrodendraceae, Putranjivaceae, Phyllanthaceae and Euphorbiaceae sensu stricto in the work of Wurdack *et al*., (2004). The presence or absence of stomata on the upper surfaces is often a good diagnostic feature (Sonibare & Adeniran, 2014; Kolawole *et al*., 2017). Three basic types of stomata were encountered in this study namely: anisocytic in *B. ferruginea*, anomocytic and paracytic in *H. crepitans* and anomocytic and anisocytic in *R. heudelotii*. These findings were in agreement with those of Metcalfe & Chalk (1950), who reported the presence of anomocytic (i.e. Ranunculaceous type) and paracytic (i.e. Rubiaceous type)

stomata across the Euphorbiaceae and also of Aworinde *et al*., (2009), who reported that the most common types of stomata in the family are the anomocytic and anisocytic. Our findings are also slightly different from those of Aworinde *et al*., (2009), who reported the presence of parallelocytic stomata on the abaxial leaf surfaces of *H. crepitans* and that of Kadiri & Adeniran (2016); who reported the presence of paracytic and amphibrachy paracytic stomata on the adaxial leaf surfaces of *H. crepitans*.

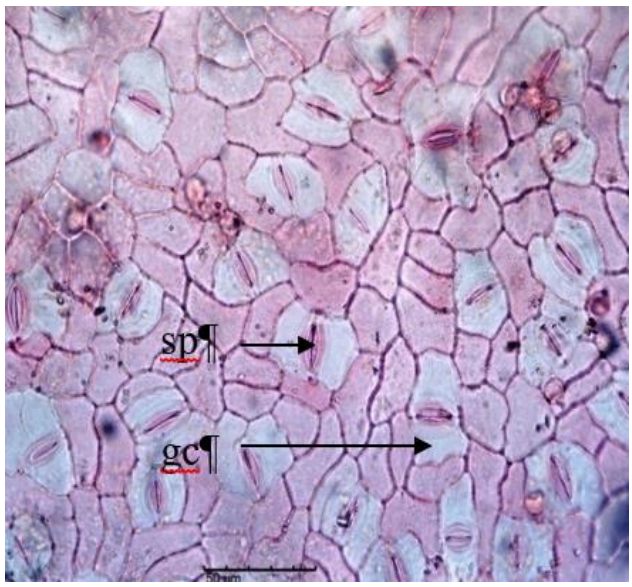
Besides the stomatal index value of 26.79 ± 0.89 µm recorded in *H. crepitans* from GSV, stomatal density and indices in TRFV and DSV (mesic zones) had higher values than that of GSV (xeric zone) in the abaxial leaf surfaces of the three taxa (Table 2). This could be attributed to adaptation to changes in climatic conditions. Thakur & Patil (2001) and Petrova (2012) noted that plants with small leaf sizes, thick cuticles and/or pubescent are associated with small stomata. They equally noted that atmospheric humidity, CO_2; temperature and light intensity can influence stomatal density. The stomatal density is also correlated to the CO_2 uptake in that the higher the density, the more the carbon dioxide that can be taken up and subsequently more water transpiration pull (Kolawole *et al*., 2017).



Plates 4a-f. Adaxial and abaxial surfaces of the leaf of *Hura crepitans showing* anomocytic and paracytic stomata (x 400) a and b = TRFV; c and d = DSV; e and f = GSV. ec = epidermal cell; sp = stomata pore; gc guard cell, sc = subsidiary cell, tr = trichome.



(a)



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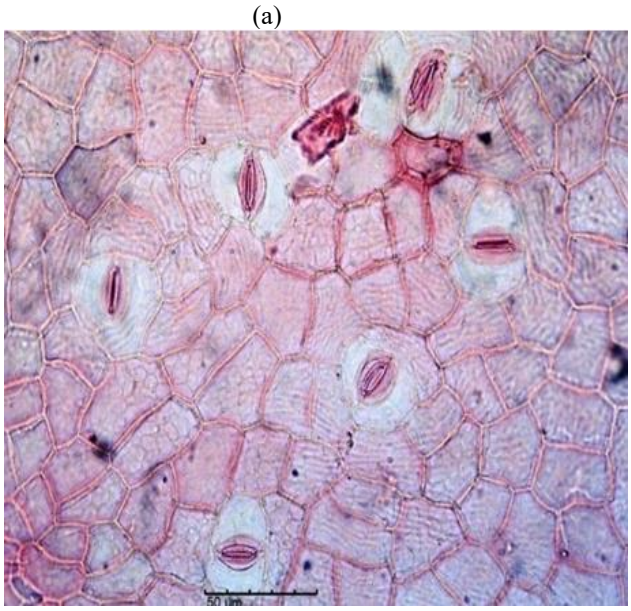

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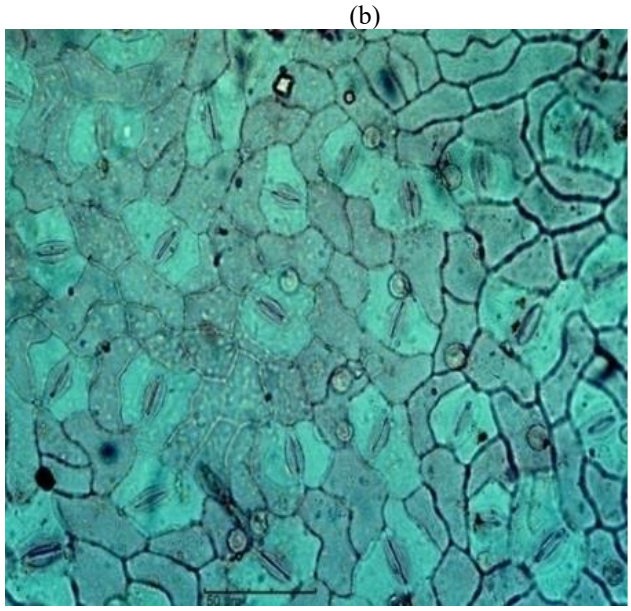

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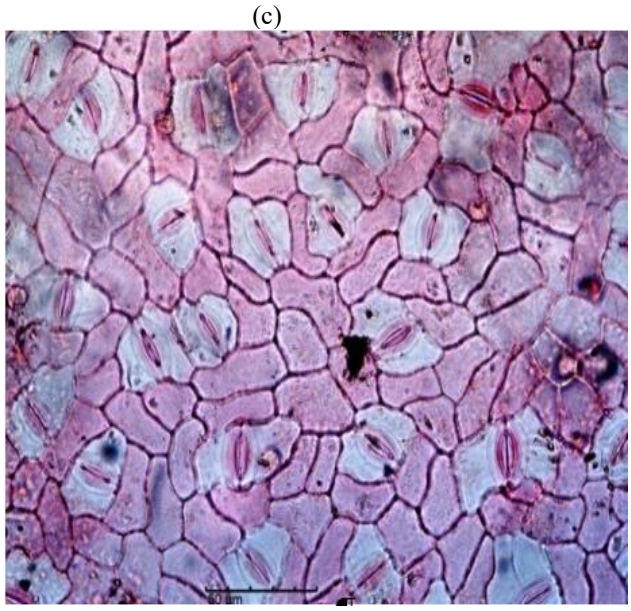

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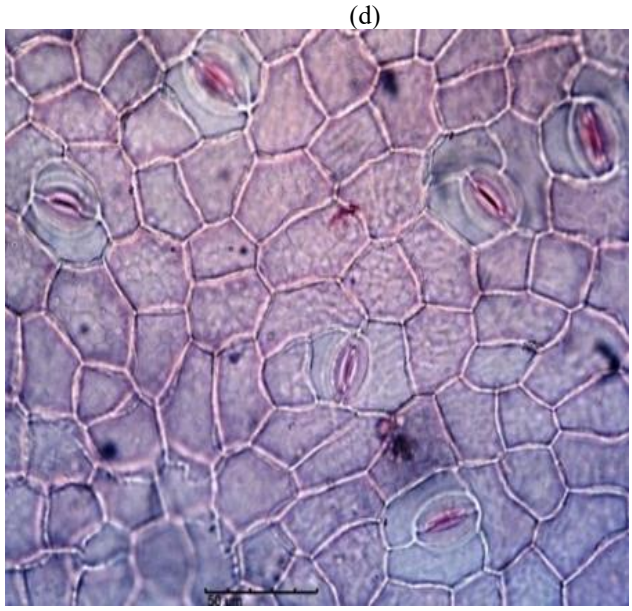

(f)

Plates 5a-f: Adaxial and abaxial surfaces of the leaf of *Ricinodendron heudelotii* showing anomocytic and anisocytic stomata (x 400) a and b = TRFV; c and d = DSV; e and f = GSV, ec = epidermal cell; sp = stomata pore; gc guard cell, sc = subsidiary cell.

Table 1. Qualitative values of stomatal features of the three genera.

Genera	Stomata			Trichome Nature	Nature of epidermal wall	
	Distribution	Adaxial	Abaxial		Adaxial	Abaxial
B. ferruginea	Hypostomatic	Absence	Anisocytic	Uniseriate	Polygonal, Irregular	Polygonal
H. crepitans	Amphistomatic	Anomocytic	Anomocytic, Paracytic	Uniseriate (scanty)	Polygonal	Polygonal
R. heudelotii	Amphistomatic	Anomocytic	Anomocytic, Anisocytic	Glabrous	Irregular Polygonal	Variously elongated

Table 2. Leaf epidermal variables (Abaxial) in *B. ferruginea, H. crepitans* and *R. heudelotii* from the three ecological zones (µm).

Genera	Tropical Rainforest	Derived Savannah	Guinea Savannah
	Stomata length (F-LSD = 4.32)		
B. ferruginea	$19.94 \pm 0.62^{b,1}$	$19.63 \pm 0.30^{b,1}$	$20.02 \pm 0.79^{b,1}$
H. crepitans	$27.69 \pm 0.42^{a,12}$	$25.27 \pm 0.43^{a,2}$	$30.17 \pm 0.49^{a,1}$
R. heudelotii	$26.10 \pm 1.16^{a,1}$	$25.69 \pm 0.77^{a,1}$	$26.02 \pm 1.07^{a,1}$
	Stomata breadth (F-LSD = 5.75)		
B. ferruginea	$12.50 \pm 2.30^{b,1}$	$10.92 \pm 0.27^{b,1}$	$12.18 \pm 0.75^{b,1}$
H. crepitans	$19.77 \pm 1.23^{a,1}$	$18.26 \pm 0.41^{a,1}$	$20.10 \pm 0.29^{a,1}$
R. heudelotii	$15.22 \pm 0.52^{ab,1}$	$15.22 \pm 0.31^{ab,1}$	$16.98 \pm 0.77^{ab,1}$
	Stomata size (F-LSD = 170.84)		
B. ferruginea	252.90 ± 54.88^{b}	214.43 ± 6.10^{b}	244.72 ± 21.64^{b}
H. crepitans	548.87 ± 41.12^{a}	461.62 ± 15.27^{a}	606.61 ± 17.08^{a}
R. heudelotii	397.51 ± 24.73^{ab}	391.20 ± 17.03^{a}	443.18 ± 34.12^{a}
	Stomata number (F-LSD = 2.60)		
B. ferruginea	$21.50 \pm 0.29^{a,2}$	$24.50 \pm 0.65^{b,1}$	$17.00 \pm 0.41^{b,3}$
H. crepitans	$20.00 \pm 0.41^{a,12}$	$24.00 \pm 0.41^{b,1}$	$17.50 \pm 0.29^{ab,2}$
R. heudelotii	$22.50 \pm 0.29^{a,2}$	$28.25 \pm 0.63^{a,1}$	$19.75 \pm 0.25^{a,3}$
	Stomata density (F-LSD = 14.53)		
B. ferruginea	$126.47 \pm 1.70^{ab,2}$	$144.12 \pm 3.80^{b,1}$	$100.00 \pm 2.40^{b,3}$
H. crepitans	$117.65 \pm 2.40^{b,2}$	$141.18 \pm 2.40^{b,1}$	$102.94 \pm 1.70^{ab,2}$
R. heudelotii	$132.35 \pm 1.70^{a,2}$	$166.18 \pm 3.70^{a,1}$	$116.18 \pm 1.47^{a,3}$
	Epidermal cell number (F-LSD = 7.67)		
B. ferruginea	$79.50 \pm 1.19^{b,2}$	$75.50 \pm 0.87^{b,2}$	$90.75 \pm 1.11^{a,1}$
H. crepitans	$55.00 \pm 1.47^{c,2}$	$73.75 \pm 1.65^{b,1}$	$48.00 \pm 2.04^{b,2}$
R. heudelotii	$84.00 \pm 1.08^{a,2}$	$94.00 \pm 1.29^{a,1}$	$97.75 \pm 0.48^{a,1}$
	Stomata index (F-LSD = 3.30)		
B. ferruginea	$61.40 \pm 0.53^{a,2}$	$65.59 \pm 0.74^{a,1}$	$52.41 \pm 0.34^{a,3}$
H. crepitans	$26.70 \pm 0.86^{b,1}$	$24.56 \pm 0.21^{b,1}$	$26.79 \pm 0.89^{b,1}$
R. heudelotii	$21.13 \pm 0.34^{c,1}$	$23.10 \pm 0.28^{b,1}$	$16.81 \pm 0.15^{c,2}$

Data are presented with mean ± standard error; means with different alphabets on each vertical array of each parameter represents significant differences among the genera, while means with different number along each horizontal array represent significant difference across the ecological zones

Table 3. Leaf epidermal variables (Adaxial) in *B. ferruginea, H. crepitans* and *R. heudelotii* from the three ecological zones (µm).

Genera	Tropical Rainforest	Derived Savannah	Guinea Savannah
	Stomata length (F-LSD = 5.14)		
B. ferruginea	$0.00 \pm 0.00^{b,1}$	$0.00 \pm 0.00^{b,1}$	$0.00 \pm 0.00^{b,1}$
H. crepitans	$26.38 \pm 1.06^{a,1}$	$24.67 \pm 0.34^{a,1}$	$27.79 \pm 0.54^{a,1}$
R. heudelotii	$24.89 \pm 0.49^{a,2}$	$22.90 \pm 2.09^{a,2}$	$30.14 \pm 1.23^{a,1}$
	Stomata breadth (F-LSD = 2.77)		
B. ferruginea	$0.00 \pm 0.00^{b,1}$	$0.00 \pm 0.00^{b,1}$	$0.00 \pm 0.00^{b,1}$
H. crepitans	$15.83 \pm 0.55^{a,2}$	$14.55 \pm 0.04^{a,2}$	$18.76 \pm 0.71^{a,1}$
R. heudelotii	$14.90 \pm 0.38^{a,2}$	$13.18 \pm 0.50^{a,2}$	$21.90 \pm 0.81^{a,1}$
	Stomata size (F-LSD = 142.56)		
B. ferruginea	$0.00 \pm 0.00^{b,1}$	$0.00 \pm 0.00^{b,1}$	$0.00 \pm 0.00^{b,1}$
H. crepitans	$419.27 \pm 31.27^{a,12}$	$358.89 \pm 5.99^{a,2}$	$521.34 \pm 23.66^{a,1}$
R. heudelotii	$371.03 \pm 13.86^{a,2}$	$304.21 \pm 37.34^{a,2}$	$662.30 \pm 47.66^{a,1}$
	Stomata number (F-LSD = 1.45)		
B. ferruginea	$0.00 \pm 0.00^{c,1}$	$0.00 \pm 0.00^{c,1}$	$0.00 \pm 0.00^{b,1}$
H. crepitans	$2.75 \pm 0.25^{b,2}$	$2.25 \pm 0.25^{b,2}$	$6.25 \pm 0.25^{a,1}$
R. heudelotii	$5.75 \pm 0.25^{a,1}$	$6.25 \pm 0.25^{a,1}$	$5.25 \pm 0.25^{a,1}$
	Stomata density (F-LSD = 7.02)		
B. ferruginea	$0.00 \pm 0.00^{c,1}$	$0.00 \pm 0.00^{c,1}$	$0.00 \pm 0.00^{b,1}$
H. crepitans	$16.18 \pm 1.47^{b,2}$	$13.23 \pm 1.47^{b,2}$	$36.76 \pm 1.47^{a,1}$
R. heudelotii	$33.82 \pm 1.47^{a,1}$	$36.76 \pm 1.47^{a,1}$	$30.88 \pm 1.47^{a,1}$
	Epidermal cell number (F-LSD = 7.05)		
B. ferruginea	$108.00 \pm 0.71^{a,2}$	$113.75 \pm 1.38^{a,1}$	$91.50 \pm 1.71^{a,3}$
H. crepitans	$69.25 \pm 1.11^{b,12}$	$74.25 \pm 0.85^{c,1}$	$63.50 \pm 1.32^{b,2}$
R. heudelotii	$73.50 \pm 0.87^{b,2}$	$87.50 \pm 1.32^{b,1}$	$64.25 \pm 1.25^{b,3}$
	Stomata index (F-LSD = 1.74)		
B. ferruginea	$0.00 \pm 0.00^{c,1}$	$0.00 \pm 0.00^{c,1}$	$0.00 \pm 0.00^{b,1}$
H. crepitans	$3.83 \pm 0.35^{b,2}$	$2.94 \pm 0.31^{b,2}$	$8.97 \pm 0.42^{a,1}$
R. heudelotii	$7.25 \pm 0.26^{a,1}$	$6.68 \pm 0.32^{a,1}$	$7.55 \pm 0.24^{a,1}$

Data are presented with mean ± standard error; means with different alphabets on each vertical array of each parameter represents significant differences among the genera, while means with different number along each horizontal array represent significant difference across the ecological zones

Conclusion

The study has contributed to the taxonomic understanding of the Euphorbiaceae sensu lato. The presence of the anisocytic, anomocytic and paracytic stomata types has supported the findings of earlier researchers and reconfirmed the usefulness of leaf epidermal features in the delimitation of plant taxa. The present study also indicated that plants in more xeric zone tend to have reduced stomatal density, which in no doubt, results in low rates of transpiration thereby conserving water.

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(Received for publication 8 May 2023)