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Comparative anatomical study of leaves for twelve Indonesian woody plant species

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Abstract. *Damaiyani J, Fiqa AP, Rindyastuti R, Lestari DA, Rahadiantoro A, Yulistyarini T. 2022. Comparative anatomical study of leaves for twelve Indonesian woody plant species. Biodiversitas 23: 3744-3754.* Foliar epidermal and stomatal features are widely used as plant microscopic traits either from taxonomic or ecological standpoints. The studies on woody plants in the Malesian region which serve as the essential component of tropical ecosystems are still limited. Here we conduct a comparative study on stomatal and epidermal features of twelve important woody plant species from Indonesia using a descriptive method based on the Light Microscope and Scanning Electron Microscope micrographs. The shape, size and stomatal type were revealed in all species studied. Moreover, quantitative features were measured including epidermal cell length and width, solidity (S), aspect ratio (AR), and stomatal index (SI). The study results showed a variation in foliar epidermal and stomatal traits across the species studied. Due to the presence of trichomes and waxes on the abaxial side, not all species can be measured for their stomatal index and epidermal cells. *Syzygium polyanthum* has the highest stomatal index and cells with complex interlocking shapes, which provide an effective strategy for reducing mechanical stress on epidermal cell walls, making this native species predicted to adapt well when it was planted in habitats with similar environmental conditions.

Keywords: Cell solidity, epidermis, leaf trait, micrographs, stomatal

INTRODUCTION

The study of anatomical features is one of the very important investigations in various systematic research and it is also a good model for exploring basic plant developmental processes (Hepworth et al. 2018; Butt et al. 2020). From a biosystematic standpoint, the foliar epidermis is one of the most notable taxonomic features, and the leaf epidermis is used in taxonomic investigations for several plant families. Although taxonomists have only recently recognized the relevance of microscopic epidermal traits, taxonomic monographs are now considered an incomplete method without that matter (Ahmad et al. 2010). Anatomical data has been acknowledged in traditional taxonomy because the varieties within a species, genus, or family are frequently reflected in anatomical traits. Although research on the plant's macroscopic morphology and wood anatomy have been proven as useful tools for identifying plants, such as leaf venation (Scott and Buot 2022), foliar epidermal morphology is an important part of the identification criterion (Ahmad et al. 2010; Akinsulire et al. 2018).

The epidermis is composed of various functionally specialized cells that regulate water loss, defense against harsh sunlight and wind, function in gas exchange, photosynthesis, pollinator attraction, respiration, transpiration, transpiration, flexibility, and mechanical strength (Butt et al. 2020). The epidermic layer derives from many features such as stomata, trichomes, and other anatomical features of the leaf epidermis, which serve as helpful anatomical tools in plants (Ahmad et al. 2010). Taxonomic problems have been widely solved using foliar anatomical characters such as stomata and trichomes (Begum et al. 2014). Moreover, foliar epidermal traits are very important for studying systematics and characterization among subfamilies and tribes (Wetzel et al. 2017).

For several centuries, scientists and botanists have been interested in the variation and development of stomata (Nunes et al. 2020). The features of plant stomata are an essential aspect of plant germplasm resource diversity. Stomata are the critical organs in plant phylogenetic research, and their properties are important indexes in plant origin, evolution, and classification investigations (Hong et al. 2018). Further, stomatal architecture, which includes the size, form, and orientation pattern of stomatal guard cells concerning other epidermal cells (subsidiary cells), is fundamental in taxonomic terms (Das 2002; Shtein et al. 2017). Different plant species contributed to the variation of this trait in different ways (Hong et al. 2018). Guard cells are distinct from regular epidermal cells in that they are specialized structures (Amaral and Mello-Silva 2008; Rahayu et al. 2012). In systematics, the number and arrangement of secondary cells have long been employed as a character (Amaral and Mello-Silva 2008; Shtein et al. 2017). Stomatal pores in the aerial epidermis are not only important ecologically, but they constitute one of the most persistent micromorphological traits in terrestrial plant evolution (Rudall et al. 2013). Moreover, the stomatal complex can vary significantly between taxa. It is further advantageous in taxonomy, ecology, and evolution studies.

The ultrastructure of epidermal cells from Indonesian woody plant species has received less attention, while this plant functional group is essential in composing tropical ecosystems. There have not been any comprehensive morphological studies on the epidermic features of Indonesian woody plant species. The current study is focused on describing leaf epidermal cell features including shape types and size of certain Indonesian woody plant species. We also examined the epidermal features of studied species compared to their generic relatives using previous study results.

MATERIALS AND METHODS

Plant materials

In this study, mature leaves of twelve Indonesian plants were collected from Purwodadi Botanic Garden-National Research and Innovation Agency (BRIN), Pasuruan, East Java (Indonesia), which is located 300 meters above sea level and has temperatures ranging from 22° to 32° C. The coordinates of the sites are latitude 112.4412 and longitude -7.4759. All species are collected in Indonesia; nine species are native to Indonesia, while three species are introduced and are now widely spread in Indonesia (Table 1). Each species was represented by three individuals, and in each individual three leaf samples were taken to be observed.

Stomatal and epidermal measurement

Epidermal and stomatal observations were carried out on the abaxial part, based on the presence of stomata, using both conventional anatomical preparation procedures (LM observation) (Zhu et al. 2018; Salsinha et al. 2021) and scanning electron microscope (SEM) procedures (Akçin et al. 2013). For LM observation, the abaxial surface of the leaf was coated with a thin layer of nail polish or glycerin. The nail polish or glycerin imprint was removed from the leaf surface after drying for around 30 minutes and examined under a light microscope at magnifications of 100X to 400X.

Table 1. Species studied in this research, Family name, local name, and their global distribution

Species	Family	Local name	Distribution	Ref.
Canarium vulgare	Burseraceae	*Kenari*	Java, Lesser Sunda Is., Maluku, New Guinea, Solomon Is., Sulawesi	https://powo.science.kew.org
Syzygium polyanthum	Myrtaceae	*Salam*	Andaman Is., Borneo, Cambodia, Java, Laos, Lesser Sunda Is., Malaya, Myanmar, Nicobar Is., Philippines, Sumatra, Thailand, Vietnam	https://powo.science.kew.org
Cynometra schefferi	Fabaceae	*Pohon sapu tangan*	Sulawesi to the Bismarck Archipelago	https://powo.science.kew.org
Artocarpus heterophyllus	Moraceae	*Nangka*	SW. India introduced to Africa and Southeast Asia, Vietnam, Thailand to Indonesia	https://powo.science.kew.org
Ficus religiosa	Moraceae	*Pohon bodhi*	Andaman Is., Assam, Bangladesh, East Himalaya, India, Malaya, Myanmar, Nepal, Nicobar Is., Pakistan	https://powo.science.kew.org
Garcinia dulcis	Clusiaceae	*Mundu*	Andaman Is., Borneo, Java, Lesser Sunda Is., Malaya, Maluku, Myanmar, New Guinea, Nicobar Is., Philippines, Queensland, Sulawesi	https://powo.science.kew.org
Protium javanicum	Burseraceae	*Trenggulun*	Java to Lesser Sunda Islands	https://powo.science.kew.org
Swietenia macrophylla	Meliaceae	*Mahoni*	Mexico to Bolivia and Brazil	https://powo.science.kew.org
Cinnamomum sintoc	Lauraceae	*Kayu manis*	Borneo, Java, Lesser Sunda Is., Malaya, Sumatra	https://powo.science.kew.org
Dimocarpus longan	Sapindaceae	*Kelengkeng*	Andaman Is., Assam, Bangladesh, Borneo, Cambodia, China South-Central, China Southeast, East Himalaya, Hainan, Laos, Malaya, Maluku, Myanmar, Nicobar Is., Philippines, Sri Lanka, Sulawesi, Sumatra, Thailand, Vietnam	https://powo.science.kew.org
Diospyros celebica	Ebenaceae	*Kayu hitam*	Sulawesi	https://powo.science.kew.org
Durio zibethinus	Malvaceae	*Durian*	Sumatra to Borneo	https://powo.science.kew.org

For scanning electron microscopy, the dissected leaves were extracted from the untreated fresh specimen and placed on a stub with sticky carbon tape. Then, the prepared stubs were coated with gold using a sputter coater for 30 minutes. The foliar epidermis was examined under a scanning electron microscope (SEM) 'FEI type Inspects 25' at magnifications of 500X to 5000X. The microscopic qualitative features have been considered, including epidermal cell shape and cell wall sinuosity, surface, and type of stomata. Quantitative features measured in this study included epidermal cell length and width, solidity (S), aspect ratio (AR), and stomatal index (SI). Solidity was determined by discovering the area of the cell shape (A_C) and dividing it by the area of the convex hull (A_{CH}). To calculate the aspect ratio, cells were oriented according to their longest axis, and the longest cell width (W) was divided by the longest cell length (L) in this orientation (Vofely et al. 2019). A comparison of foliar structures of 12 studied species was also done with their genetic relatives.

The stomatal index (SI) was calculated using the formula of Salisbury (1927). In this study, the stomata index was determined at a 200x200 µm field of view area.

$$\mathbf{SI} = \frac{\mathbf{S}}{\mathbf{S+E}} \times 100$$

Where:

S : Stomata number per leaf area unit

E : Epidermal cell number in the sample leaf unit

Data analysis

Variations of solidity, Aspect Ratio (AR), and stomatal index were analyzed statistically by Analysis of Variance (ANOVA) confidence level of 95%. Further, the post hoc Tukey and Games-Howell test was performed for data grouping using Paleontological Statistics (PAST) 4 (Hammer et al. 2001). While the stomatal type, epidermal shape, and the presence of trichome in each species were described descriptively.

RESULTS AND DISCUSSION

Epidermal leaf traits

This study revealed that the epidermal cells at the abaxial side of all studied species were irregular and isodiametric in outline. Irregular arrangements were found on *C. vulgare, C. schefferi, S. polyanthum, G. dulcis, P. javanicum, F. religiosa,* and *S. macrophylla*. At the same time, isodiametric types of cells were observed in *C. sintoc* and *A. heterophyllus*. According to Song et al. (2020), leaf epidermal cells were arranged isodiametrically or irregularly, and their shapes were penta-, hexa- to polygonal, irregular to polygonal, or irregular. The isodiametric epidermal cells usually had straight or straight to curved anticlinal walls, whereas irregular cells usually had undulous or sinuous anticlinal walls. The results of this study are in line with Song et al. (2020). Cells arranged irregularly in *C. vulgare, C. schefferi, S. polyanthum, G. dulcis, P. javanicum, F. religiosa*, and *S. macrophylla* have undulating margins, but cells arranged isodiametrically in *C. sintoc* and *A. heterophyllus* tend to have straight margins (Figure 1, 2, 3).

The traditional shape descriptors of solidity and aspect ratio were used to quantify epidermal cell shape. According to Vofely et al. (2019), solidity and aspect ratio were good descriptors for margin undulation and base cell shape, respectively. In this observation, we found that solidity values had significant differences among the species. The cell solidity in all samples occupied a range between 0.62 and 0.89. On the abaxial surface, most of the cells from *C. sintoc* and *A. heterophyllus* showed a curved cell wall, so the value of solidity in both species was high and slightly different from each other, which were 0.89 and 0.83, respectively. Epidermal cell walls of most plant species studied displayed margin undulation with the gradation of sinuosity, sinuous in *S. polyanthum, G. dulcis, P. javanicum,* and *S. macrophylla* showing tend to be strong, while slightly sinuous in the *C. vulgare, C. schefferi*, and *F. religiosa*. Most sampled epidermal cells showed some degree of undulation. From this research result, it is shown that there are no species that displayed complex margins (low solidity, S = 0.5). Vofely et al. (2019) discovered that cells with complex margins were the most common in the ferns, with epidermal cell fern margins more undulating than eudicots. Our research of cell aspect ratios revealed most epidermal cells of local woody plant species studied had an elliptical base cell shape (median > 0.5), and showed significant differences among species. The aspect ratio showed a range of 0.6 and 0.7 on average, except *P. javanicum* (AR = 0.53), and neither highly anisotropic nor truly isotropic cells were discovered in our data (Table 2). However, *C. vulgare* showed the largest range of aspect ratio, from 0.42 to 0.93. The research results are in line with Vofely et al. (2019), who claimed that the aspect ratio of dicots is between 0.6 and 0.7, indicating a slightly ellipsoidal base shape norm. In contrast, gymnosperms and monocots have more skewed distributions, with medians below 0.4, indicating a trend towards a more anisotropic base shape in these groups. We found that the average epidermal cell in local woody plants could be best represented by a quite anisotropic cell with weak margin undulation. According to Sapala et al. (2018), cells with complex interlocking shapes, similar to puzzle pieces, which were found in *C. vulgare, C. schefferi, S. polyanthum, G. dulcis, P. javanicum, F. religiosa,* and *S. macrophylla*, provide an effective strategy to reduce mechanical stress on the epidermal cell walls.

The epidermal cell shape of three species observed, namely *D. longan, D. celebica,* and *D. zibethinus,* cannot be described. The epidermis of *D. celebica* and *D. longan* were covered by a meshwork of asterisk-like epicuticular wax on its epidermal abaxial surface. While on *D. zibethinus*, the presence of trichomes on the leaf surface which densely covered the surface area of the leaf causes the epidermal cell shape to be unobservable. However, the SEM method's observations can show the shape, size, and stomatal type in all species studied (Figure 4).

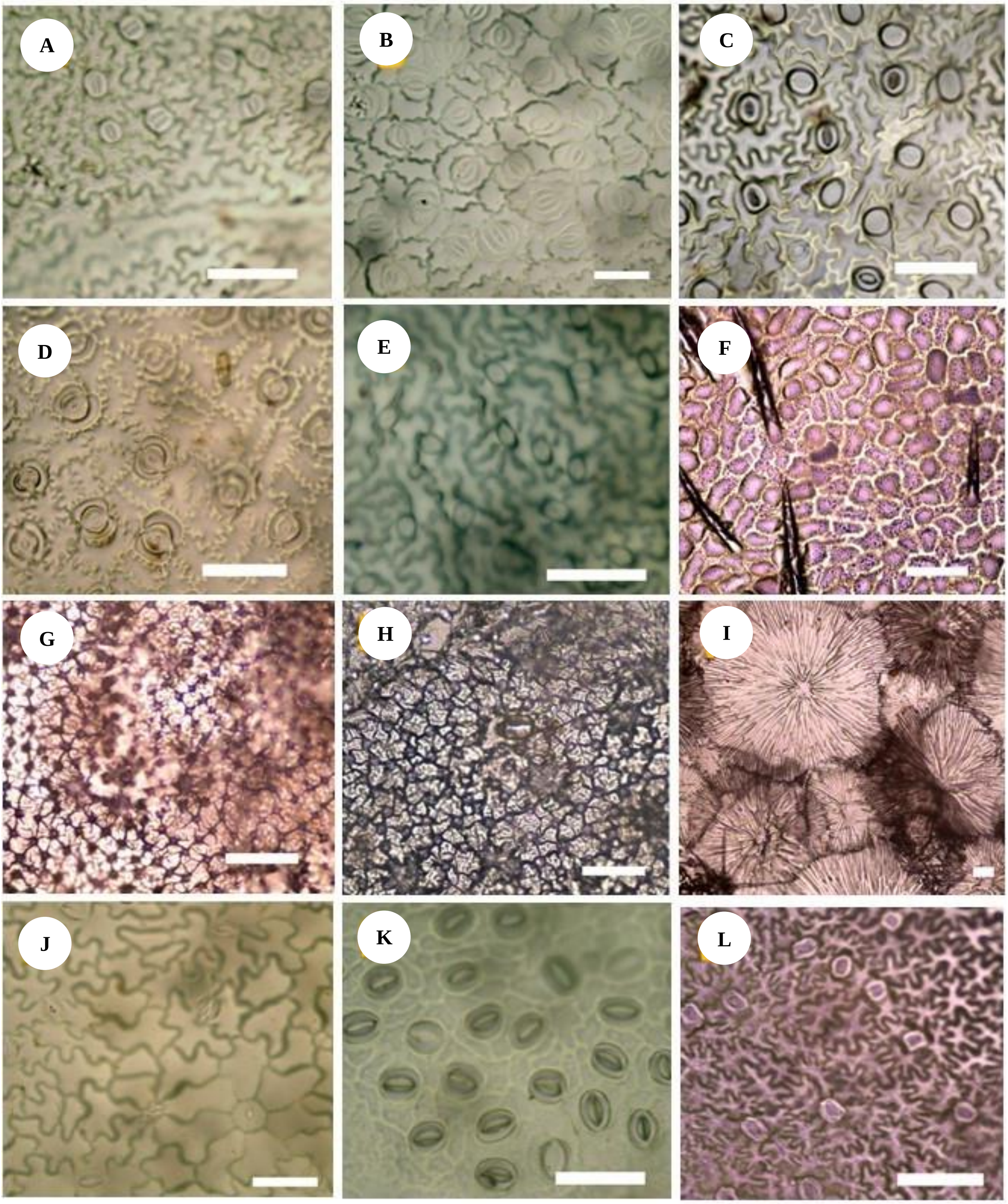

Figure 1. Photographs of leaf epidermal cells on the abaxial surface by light microscope (LM). (A) *Canarium vulgare*, (B) *Cynometra schefferi*, (C) *Syzygium polyanthum*, (D) *Garcinia dulcis*, (E) *Protium javanicum*, (F) *Cinnamomum sintoc*, (G) *Dimocarpus longan*, (H) *Diospyros celebica*, (I) *Durio zibethinus*, (J) *Ficus religiosa*, (K) *Artocarpus heterophyllus*, (L) *Swietenia macrophylla*. Bar = 50 µm

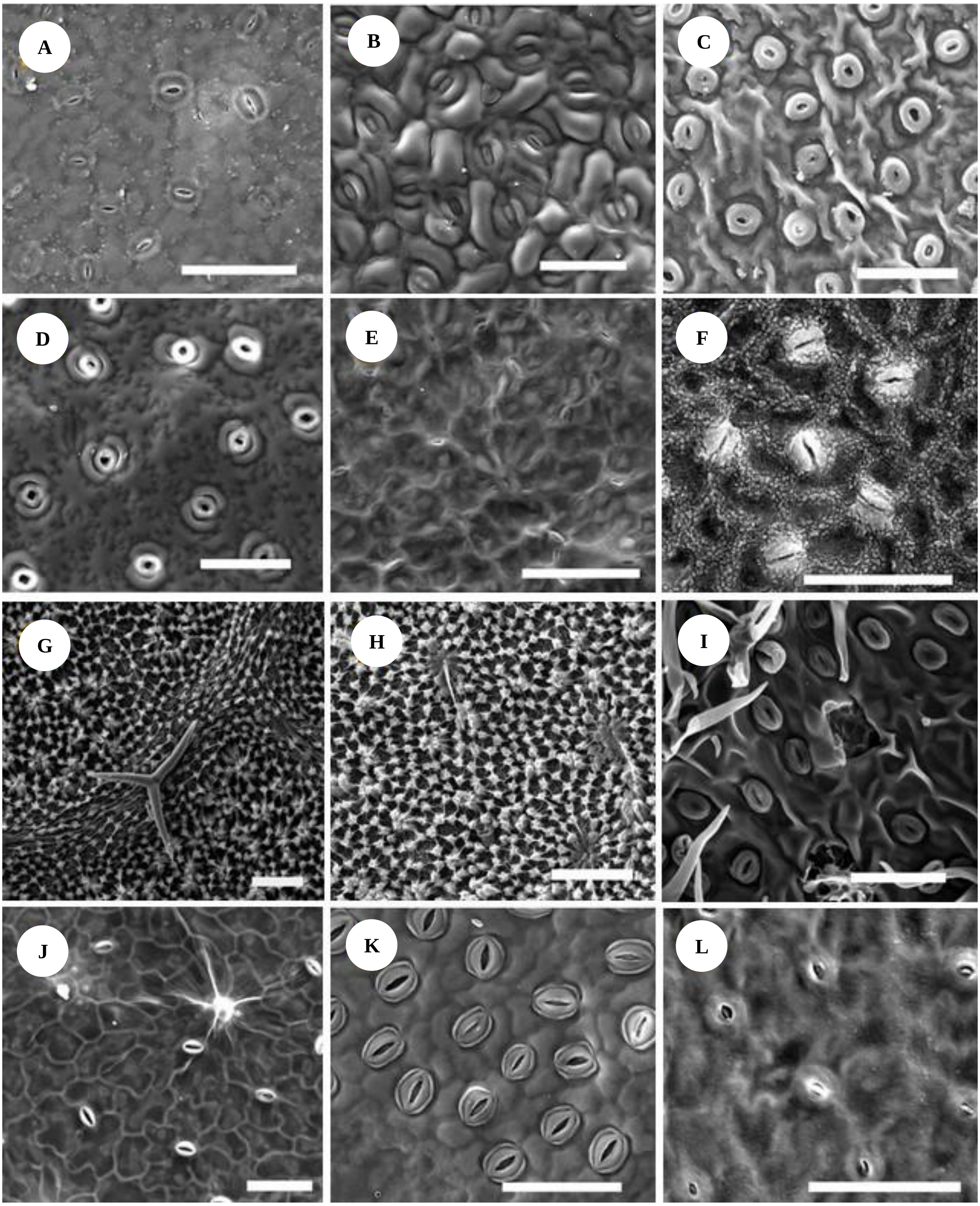

Figure 2. Photographs of leaf epidermal cells on the abaxial surface by scanning electron microscope (SEM). (A) *Canarium vulgare*, (B) *Cynometra schefferi*, (C) *Syzygium polyanthum*, (D) *Garcinia dulcis*, (E) *Protium javanicum*, (F) *Cinnamomum sintoc*, (G) *Dimocarpus longan*, (H) *Diospyros celebica*, (I) *Durio zibethinus*, (J) *Ficus religiosa*, (K) *Artocarpus heterophyllus*, (L) *Swietenia macrophylla*. Bar = 50 µm

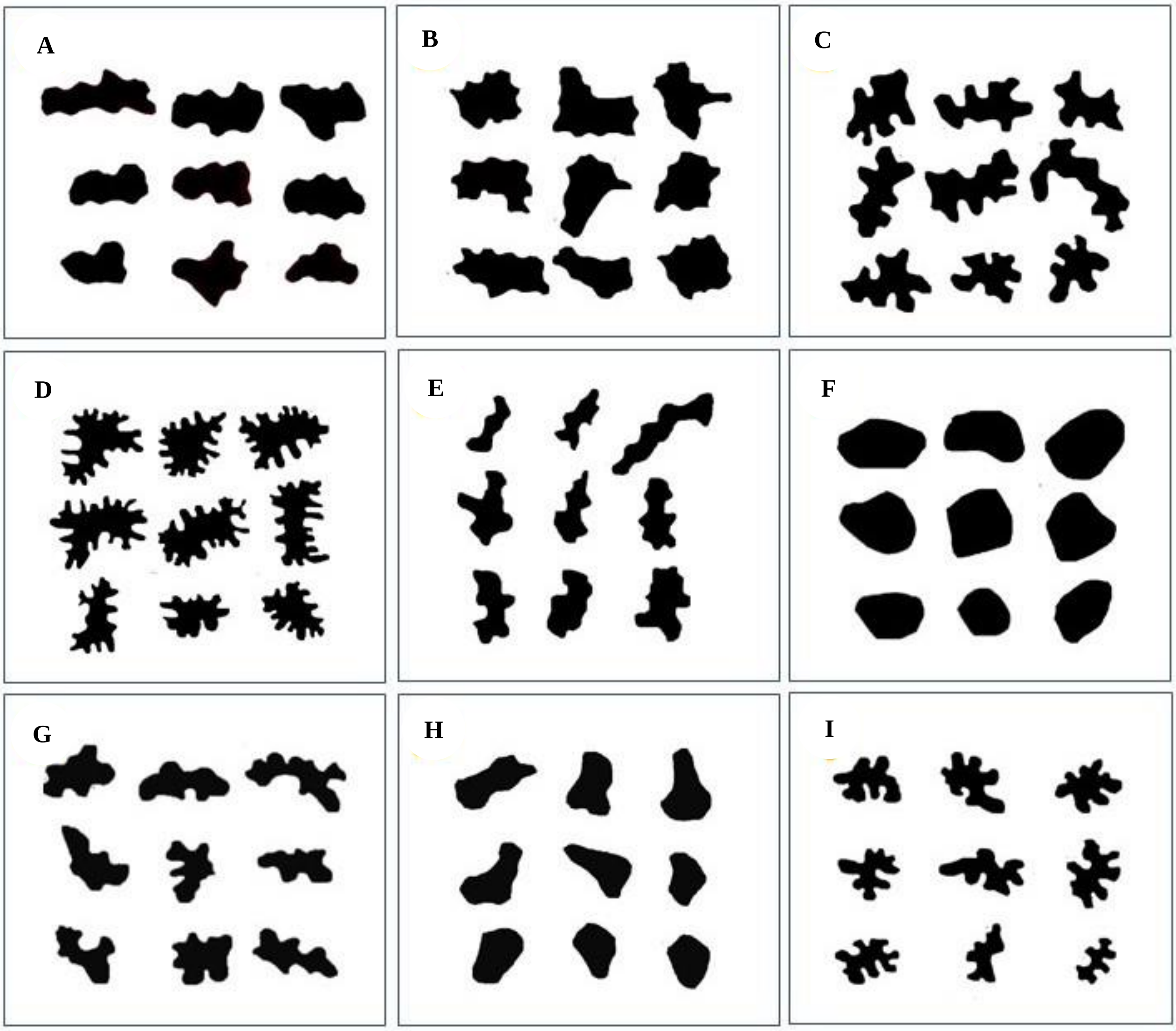

Figure 3. Illustration of leaf epidermal cells on the abaxial surface. (A) *Canarium vulgare,* (B) *Cynometra schefferi,* (C) *Syzygium polyanthum,* (D) *Garcinia dulcis,* (E) *Protium javanicum,* (F) *Cinnamomum sintoc,* (G) *Ficus religiosa,* (H) *Artocarpus heterophyllus,* (I) *Swietenia macrophylla*

Morphological traits of stomata

Based on the microscopic observations, all species studied were composed of two kidney-shaped guard cells, likewise the stomata of other dicotyl plants. In increasing gas exchange capacity, stomata with kidney-shaped guard cells have slower movement than do the dumbbell-shaped stomata (Harrison et al. 2020). Guard cells can show morphological diversities (Kim and Lee 2017). Ultrastructure observation of stomata morphology in the 12 species studied was shown in Figure 4 and Figure 5. Stomata were mostly paracytic type. However, anomocytic, anisocytic and actinocytic stomata had also been documented. Most of the species studied had a paracytic type; they were *C. vulgare*, *S. polyanthum*, *G. dulcis*, *P. javanicum*, *D. longan, D. zibethinus*, and *A. heterophyllus*. In this type, the stoma is surrounded by two subsidiary or accessory cells that are parallel to the longest axis of the pore and the guard cells. In anomocytic type, the stoma was surrounded by a limited number of subsidiary cells which were quite like the remaining epidermal cells found on *C. sintoc* and *F. religiosa*, while in anisocytic type, the stoma was surrounded by three cells, of which one was distinctly smaller than the other two, which was found on *C. schefferi*. Lastly, *D. celebica* and *S. macrophylla* had actinocytic type, whereas stoma remains surrounded by a circle of radiating cells. According to Franks and Farquhar (2007), two species with laterally moving paracytic guard cells must overcome considerable mechanical advantages to fully open their stomata.

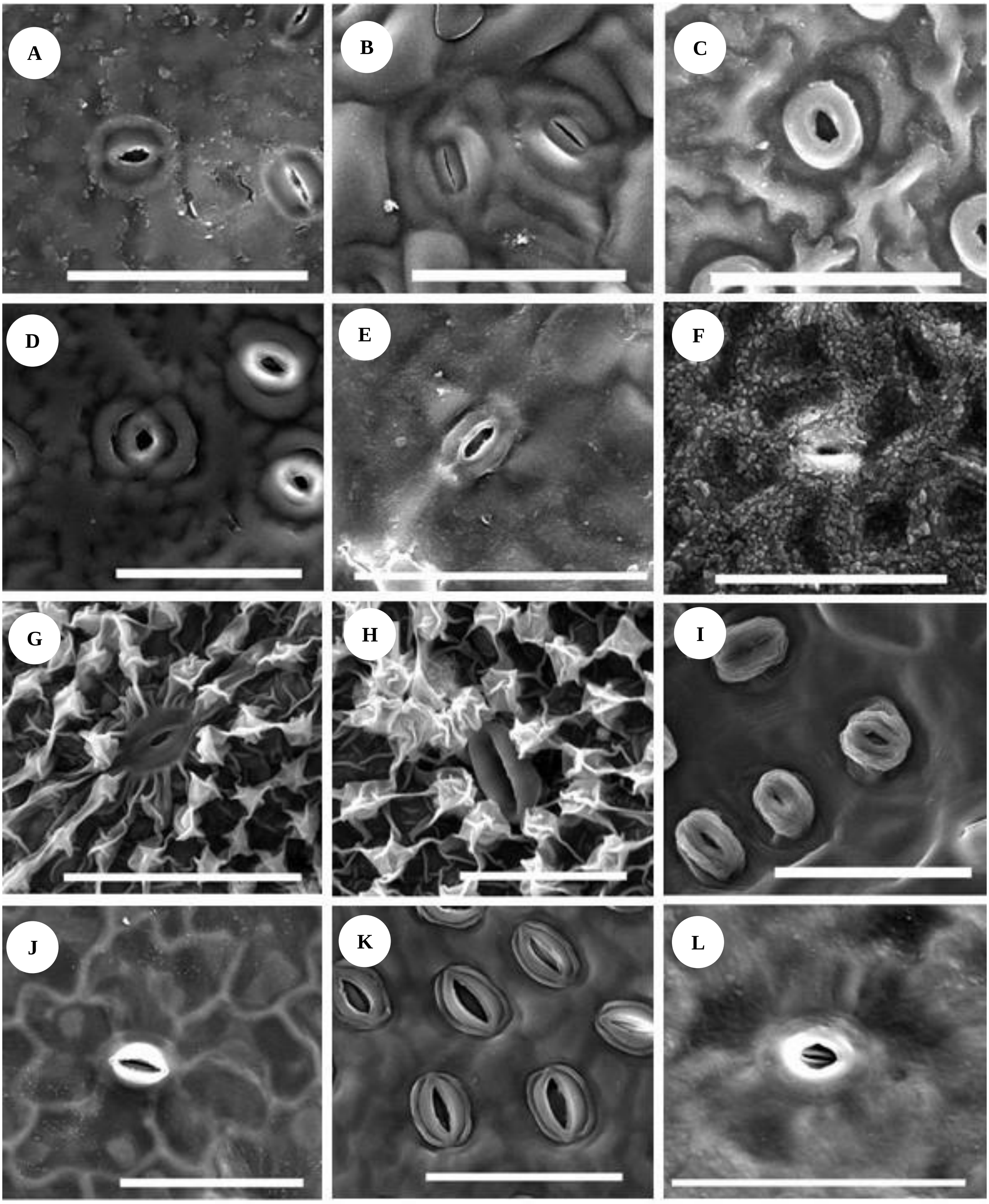

Figure 4. Photographs of stomata on the abaxial surface by scanning electron microscope (SEM). (A) *Canarium vulgare,* (B) *Cynometra schefferi*, (C) *Syzygium polyanthum*, (D) *Garcinia dulcis*, (E) *Protium javanicum*, (F) *Cinnamomum sintoc*, (G) *Dimocarpus longan*, (H) *Diospyros celebica*, (I) *Durio zibethinus*, (J) *Ficus religiosa*, (K) *Artocarpus heterophyllus*, (L) *Swietenia macrophylla*. Bar = 50 µm

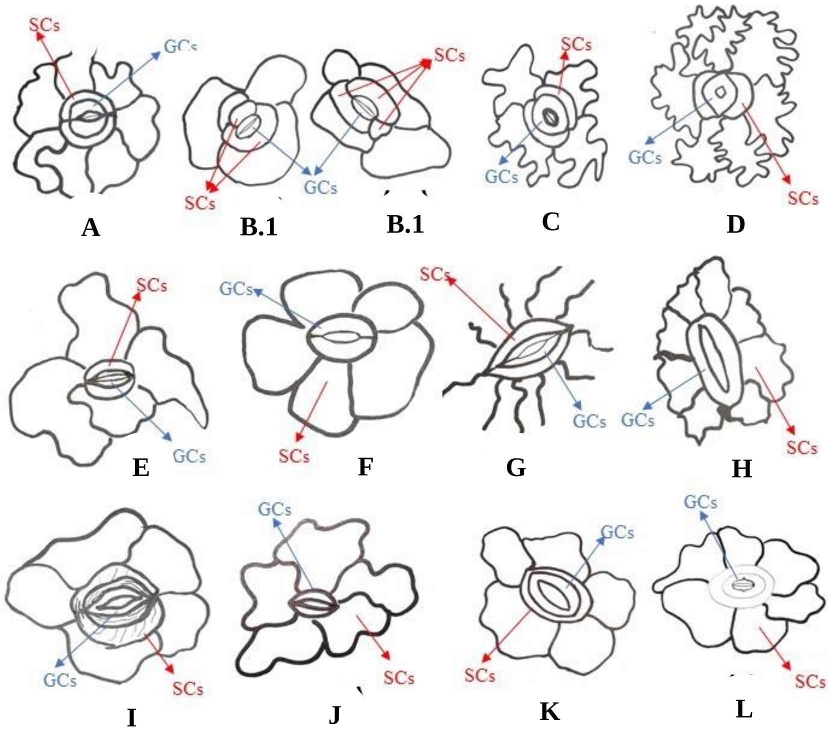

Figure 5. Illustration of stomata on the abaxial surface. A. *Canarium vulgare*, B1, B2. *Cynometra schefferi*, C. *Syzygiumpolyanthum*, D. *Garcinia dulcis*, E. *Protium javanicum*, F. *Cinnamomum sintoc*, G. *Dimocarpus longan*, H. *Diospyros celebica*, I. *Durio zibethinus*, J. *Ficus religiosa*, K. *Artocarpus heterophyllus*, L. *Swietenia macrophylla*. SCs = Subsidiary cells, GCs = Guard cells

In terms of stoma distribution, in all observed species, stomata were irregularly arranged with no fixed direction (Figure 1 and 2). Most of them were distributed on the leaf lamina, except *D. longan*, in which the stomata were only found on the veins (Figure 2G). The stoma of *D. longan* and *D. celebica* was also surrounded by a meshwork of asterisk-like epicuticular wax, as shown in Figure 4G and Figure 4H. Except for *D. zibethinus*, stomata and epidermal cells were in the same plane. Scanning electron microscopy showed sunken stomata in *D. zibethinus*. Plants with sunken stomata are frequent in arid regions but can also be found in humid climates (Gray et al. 2020). Sunken stomata have guard cells that are much lower than the rest of the leaf epidermal cells. This helps protect against the loss of water from the plants. This assumption was supported by a trichome structure in *D. zibethinus*, which covers most of the laminar area (Figure 1I). This trichome's existence, besides as adaptive behavior in the arid environment, also helps them to protect from herbivores (Kariñho -Betancourt et al. 2015).

Variation of a stomatal index from various plant families was presented in Table 1, some species showed a higher stomatal index than the others. The stomatal index on the abaxial surface was found to be high in *S. polyanthum*, with a value of 31.73%. The lowest stomatal index was recorded in *F. religiosa*, with a value of 13.05%. The values of the stomatal index of *A. heterophyllus*, *C. schefferi*, *C. sintoc*, *S. macrophylla*, *G. dulcis*, *P. javanicum*, and *C. vulgare* were 23.51%, 22.16%, 20.53%, 18.59%, 18.52%, 17.62%, 17.06%, respectively. This study could not observe the stomatal index of *D. longan*, *D. celebica*, and *D. zibethinus* because the epidermal cells were covered by a meshwork of asterisk-like epicuticular wax and very dense trichomes. The stomatal index is used to calculate CO_2 levels in the atmosphere. As a result, this symbol represents the amount of pollution in the air. The highest stomatal index indicates transpiration, metabolism, and mineral and water absorption. The stomatal index of species can also be used as a geographical indicator (Munir et al. 2011; Yan et al. 2017).

Anatomical comparison among congeneric relatives

Epidermal structures of observed species were compared with those of their congeneric species in similar genera based on literature studies. The shape of epidermal cells of *D. celebica* was unobserved mainly because of the presence of dense trichomes. However, Paembonan et al. (2021) said that the stomatal density of *D. celebica* leaves in the sun-exposed area was higher than in the shaded area. In addition, many species of *Diospyros* were not covered by the dense trichome such as *D. cauliflora*, *D. javanica* (Rindyastuti et al. 2021), and *D. lanceifolia* (Kalita et al. 2015). Akinsulire et al. (2018) investigated the taxonomic significance of epidermal and venation characters in the genus *Diospyros* L. (Ebenaceae) in Nigeria. Based on this study, the shape of epidermal cells of *Diospyros* spp. are commonly irregular, rectangular, polygonal, and pentagonal. Further, slightly sinuous to sinuous and undulating anticlinal walls are the common configuration observed in this genera.

Table 2. Qualitative and quantitative character of foliar epidermal anatomy yof local woody plant species at the abaxial side

Plant species	Family	Epidermal cell						Stomata		Trichome
		Shape	Anticlinal wall	Abaxial surface	Length x width (µm)	Solidity	AR	Type	SI	
Canarium vulgare	Burseraceae	Irregular	Undulate, slightly-sinuous	Smooth	28.01 (22.77-38.33) × 17.48 (14.06-24.03)	0.81^{bc}	0.64^{ab}	Paracytic	17.06^{ab}	Absent
Cynometra schefferi	Fabaceae	Irregular	Undulate, slightly sinuous	Smooth	37.65 (30.98-46.19) × 27.08 (19.19-37.89)	0.75^{cd}	0.73^{a}	Anisocytic	22.16^{b}	Absent
Syzygium polyanthum	Myrtaceae	Irregular	Undulate, strongly sinuous	Smooth	40.80 (32.65-51.72) × 27.34 (21.05-32.93)	0.62^{f}	0.68^{ab}	Paracytic	31.73^{a}	Absent
Garcinia dulcis	Clusiaceae	Irregular	Undulate, strongly sinuous	Smooth	37.26 (31.26-43.39) × 26.59 (19.54-31.42)	0.63^{f}	0.72^{a}	Paracytic	18.52^{ab}	Absent
Protium javanicum	Burseraceae	Irregular	Undulate, strongly sinuous	Smooth	34.17 (27.49-56.97) × 17.81 (11.43-25.63)	0.66^{ef}	0.53^{b}	Paracytic	17.62^{ab}	Absent
Cinnamomum sintoc	Lauraceae	Isodiametric	Curved	Rough	21.67 (15.05-26.04) × 16.53 (12.93-21.20)	0.89^{a}	0.77^{a}	Anomocytic	20.53^{ab}	Present
Dimocarpus longan	Sapindaceae	-	-	Covered by a meshwork of asterisk-like epicuticular wax	-			Paracytic	-	Present
Diospyros celebica	Ebenaceae	-	-	Covered by a meshwork of asterisk-like epicuticular wax	-			Actinocytic	-	Present
Durio zibethinus	Malvaceae	-	-	Covered by trichomes	-			Paracytic	-	Present
Ficus religiosa	Moraceae	Irregular	Undulate, slightly sinuous	Smooth	67.20 (50.93-87.47) × 38.96 (30.31-46.00)	0.71^{de}	0.6^{ab}	Anomocytic	13.05^{c}	Absent
Artocarpus heterophyllus	Moraceae	Isodiametric	Straight-curved	Smooth	20.36 (16.39-25.76) × 13.95 (11.60-15.81)	0.83^{ab}	0.69^{ab}	Paracytic	23.51^{b}	Absent
Swietenia macrophylla	Meliaceae	Irregular	Undulate, strongly sinuous	Smooth	37.52 (23.57-50.29) × 25.97 (18.56-32.30)	0.65^{ef}	0.7^{ab}	Actinocytic	18.59^{ab}	Absent

Note: Different letters represent significant differences between species according to a post hoc test, with $p < 0.05$

Of the comparison between *Garcinia* species, *G. nigrolineata* showed relatively similar epidermal anatomy to *G. dulcis*, it has intercostal epidermal cells that are irregular with a highly sinuous anticlinal wall (Begum 2020). Whilst, a different character was found in its stomatal type which is hemi-parasitic mixed with few anomocytic stomata. This indicates that stomatal type is important in distinguishing interspecific relationships in *Garcinia*. *Canarium vulgare* has a different epidermal cell shape with its congeneric relative *C. schweinfurthii*. It has polygonal cells with straight cell walls (Ayogu et al. 2020). This shape variation showed that foliar structure in *Canarium* may largely vary across species. Different from other taxa, a previous study in *Syzygium polyantum* indicated the variation of foliar structures among varieties or cultivars, however, the type and presence of stomata were relatively similar (Khandaker et al. 2018; Chatri et al. 2020). Epidermal cells of *S. polyantum* in Abdulrahman et al. (2018) are both regular and irregular shaped, while the specimen of this study revealed irregular cell shape.

There was a broader study of foliar structures in *Ficus* genera. Khan et al. (2011) investigated foliar structures of 22 *Ficus* species. The polygonal shape of epidermal cells is present in *F. palmata, F. religiosa, F. racemosa, F. bengalensis, F. carica, F. nerrifolia, F. hispida, F. auriculata, F. johannis, F. sarmentosa, F. subincisa, F. benjamina, F. tsiela, F. pumila, F. lacor and F. microcarpa,* in *F. foveolata* and *F. virens* adaxial surface is polygonal while those of abaxial surface is irregular. Whilst *F. religiosa* in this study presents an irregular epidermal cell shape with undulate, slightly sinuous anticlinal walls and anomocytic stomatal type at the abaxial surface. Among the species studied by Khan et al. (2011), only *F. nerifolia* showed a similar stomatal type with *F. religiosa* while the others have paracytic configuration. Similar study results were also shown by Shakir and Baji (2016).

Artocarpus heterophyllus has isodiametric epidermic cells and straight-curved anticlinal walls with paracytic stomatal type. Sa et al. (2019) revealed foliar epidermal structures of this species from Brazil. It has uniseriate epidermic cells with sinuous walls on both sides. This comparison showed intraspecific variation in foliar structures in this species, especially on the stomatal type. Moreover, interspecific variation in epidermal structures is also indicated within genera *Artocarpus*. *Artocarpus altilis* has uniseriate, polygonal cells with straight or slightly sinuous walls on both sides and anomocytic and actinocytic stomata, while *A. comunis* has uniseriate with rectangular, oval, or circular epidermal cells and steurocytic stomata (Sa et al. 2019; Akinloye et al. 2015).

Conclusion and advanced study opportunity

This research shows that *Syzygium polyanthum* has the highest stomatal index compared to other species observed and the cells of *S. polyanthum* have complex interlocking shapes that provide an effective strategy for reducing mechanical stress on epidermal cell walls, therefore this species is predicted to be more adaptable to environmental conditions than other species studied. Basically, the study on the stomatal variation of plant species has been frequently and widely conducted. However, there are wider opportunities for studying the relationship between stomatal features with other foliar traits such as epidermis that have never been conducted in Indonesia. With enormous biodiversity, we have more opportunities to carry out this study further on more diverse species to increase our understanding of the morphological and anatomical variation of plant species, and on the ability of plant species to adapt to changing environments. Because they are immediately exposed to the environment, leaf features are frequently influenced by ecosystem variables. The most significant physiological machinery for photosynthesis and transpiration in vascular plants is the stomata of leaves (Tiwari et al. 2013; Auclair 2014; Liu et al. 2019).

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