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Original Article

Leaf blade anatomy characteristics of the genus *Amorphophallus* Blume ex Decne. in Thailand

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ABSTRACT

Twenty-three species of *Amorphophallus* Blume ex Decne. were collected from several areas of Thailand between November 2008 and May 2012, and grown under greenhouse conditions with 70% sunlight. Leaf blade anatomy characteristics were studied using free hand section and epidermal peeled slides before being observed using a light transmission microscope from May 2011 to November 2013. The results showed the different anatomical characteristics of each species. The midribs in cross section were curved, or had 5, 6, 7, 8 or 12 lobes. Vascular bundles numbered 5, 6, 7, 8, 9, 10, 13, 15, 16 or 23. The upper and lower epidermal cell walls had three subtypes—straight-sided, undulate or sinuous anticlinal. Both sides of the epidermal cell wall can be similar or can vary in each species. There were 1, 2, 3, 4 or 6 subsidiary cells along both sides of paired guard cells and the stomatal type was paracytic and stomatal subtypes varied from species to species, being brachyparacytic, hemiparacytic, amphibrachyparacytic, paratetracytic or parahexacytic. The stomatal number was 16–104/mm^2 of leaf area and varied with the leaf gloss and leaf texture of each species.

Introduction

The genus *Amorphophallus* Blume ex Decne. consists of 205 species and is distributed widely in the tropics from West Africa eastward into Polynesia, while in Thailand, 58 species have been reported (Hetterscheid, 2012). The Thai vernacular name for this genus is “Buk” and this varies with species or location (Chayamarit et al., 2014). This genus is monocotyledonous and is classified in the family Araceae, subfamily Aroideae and tribe Thomsonieae, with most species being endemic (Hetterscheid, 1992; Mayo et al., 1997; Hetterscheid, 2009).

Hetterscheid (2009) provided a general description of *Amorphophallus* species as being herbs with an underground storage organ called a tuber. One single leaf emerges from the tuber consisting of a vertical petiole and a horizontal leaf blade; it is rare to see few leaves on the same plant. The leaf is dissected into a few or numerous small leaflets. Keating (2003) described the leaf anatomy of *Amorphophallus* as dorsiventral, with leaf sharpness and midrib shapes being useful to differentiate between species. Leaf venation has a marginal or submarginal collector vein and a lamina space between the lateral secondary veins which are higher order reticulate.

Hay and Mabberley (1991) reported that the inflorescence may develop when plants are mature or replace the leaf in one season, and consists of a spathe, which usually envelops the spadix. Hetterscheid (1992) reported that most parts of the spathe will wilt and drop off, the individual female flowers develop into fruit, of a berry type, containing 1–3 seeds after successful pollination. These berries are usually green when young and change to red or orange-red, blue, white, or yellow-and-white when ripe. The corm or tuber has a dormancy period in the arid season and grows well in the rainy season.

For a long time, Thai people have used the underground stem, the petiole and the young inflorescence of many species in this genus for cooking native dishes. They have local wisdom in the

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selection of non-poisonous or less-poisonous species as edible vegetables (Nguanchoo et al., 2014). Medicinal application includes the use of *Amorphophallus* starch, glucomannan, to reduce cholesterol and blood sugar levels (Sukumolnondha, 2005). For these reasons, *Amorphophallus* underground stems are collected widely from the forest and used as food and in medicinal processing (Independent News Network, 2015). In addition, many attractive species are used for decoration (McPherson and Hetterscheid, 2011). Although *Amorphophallus* specimens in Thailand have been surveyed and classified to the species level (Hetterscheid, 2012), the systematic study of this genus still needs revision. More data and information about their characteristics and species biodiversity should be obtained through systematic studies of *Amorphophallus* by collecting living specimens, examining herbarium specimens, cultivating reproductive parts and studying the botanical characteristics and ecological habitats for the growth of each species.

This study compared the morphological and anatomical characteristics of leaves that could be used for species identification at any time of the year where the leaf morphology of species is similar, especially where the inflorescence has not yet emerged from tubers or corms.

Materials and methods

Living *Amorphophallus* specimens were collected from protected areas with appropriate authorization and from natural areas in Kanchanaburi, Prachuap Khiri Khan, Surat Thani, Lop Buri, Sa Kaeo, Chachoengsao and Nakhon Ratchasima provinces, Thailand from November 2008 to May 2012. The habitat and environmental characteristics of each sample were recorded. Living specimens were grown under conditions of 70% sunlight in a greenhouse of the Department of Botany, Faculty of Liberal Arts and Science, Kasetsart University, Kamphaeng Saen campus, Nakhon Pathom province.

The leaf and leaflet morphology of the samples were studied. The anatomical characteristics of three fresh leaflets, with five replications of each species, were studied using the freehand section method (Johansen, 1940). The upper epidermis and lower epidermis were peeled onto separate glass slides and mounted using 10% glycerine for the observation of epidermal cell types, shapes and numbers of subsidiary cells surrounding pairs of guard cells and stomatal numbers per leaf area before being observed under a light microscope (model CX31; Olympus; Tokyo, Japan) and recorded using a digital camera (model DP20; Olympus; Tokyo, Japan) at 40×, 100× and 400× magnifications (Johansen, 1940). This research was undertaken in the laboratory and greenhouse from May 2011 to November 2013.

Results and discussion

Around 180 samples of living specimens of *Amorphophallus* spp. were collected from November 2008 to May 2011. The habitats of species varied with some being consistent while others differed. The habitats were in bamboo forest, deciduous dipterocarp forest, dry evergreen forest and mixed deciduous forest. Many species grew in soil pH 7–8, but some species grew in gaps of limestone. Almost all species were growing under the shade of trees, cliffs or big rocks, except for *Amorphophallus paeoniifolius* (Dennst.) Nicolson and *Amorphophallus tenuispadix* Hett. which were growing in both shade and full sunlight.

Leaf morphology

In this study, 115 living specimens of 23 species were identified by the morphology of their tubers, leaves, petioles and inflorescences after being grown under greenhouse conditions—*Amorphophallus albispathus* Hett., *Amorphophallus amygdaloides* Hett. & Sizemore, *Amorphophallus asterostigmatus* Bogner & Hett., *Amorphophallus bulbifer* (Roxb.) Blume, *Amorphophallus cruddasianus* Prain ex Engl., *Amorphophallus curvistylis* Hett., *Amorphophallus flotoi* (S.Y. Hu) ined, *Amorphophallus koratensis* Gagnep., *Amorphophallus krausei* Engl., *Amorphophallus lacourii* Linden & André, *Amorphophallus longituberosus* (Engl.) Engl. & Gehrm., *Amorphophallus macrophyllus* (Gagnep. ex Serebryanyi),

Table 1
Midrib shape, number of vascular bundles, upper and lower epidermal cell shapes, number of subsidiary cells, stomatal types and stomatal number per leaf area of *Amorphophallus* spp.

Species	Midrib shape	Number of vascular bundles	Upper epidermal cell shape	Lower epidermal cell shape	Number of subsidiary cells	Stomatal type[a]	Stomatal number/leaf area (mm^2)
Amorphophallus albispathus	Curved	13	UN	SI	2	BP	28
A. amygdaloides	8 Lobes	16	ST	UN	1, 2	HP, BP	71
A. asterostigmatus	Curved	13	UN	SI	2	BP	20
A. bulbifer	7 Lobes	9	UN	SI	4	AP	68
A. cruddasianus	6 Lobes	8	ST	SI	2	BP	43
A. curvistylis	5 Lobes	8	ST	UN	2, 4, 6	BP, TP, XP	45
A. flotoi	Curved	10	ST	UN	2, 4	BP, AP	74
A. koratensis	8 Lobes	15	ST	UN	2	BP	47
A. krausei	7 Lobes	10	SI	SI	2	BP	43
A. lacourii	Curved	5	ST	ST	2	BP	41
A. longituberosus	7 Lobes	12	ST	UN	2, 4	BP, TP	37
A. macrophyllus	Curved	6	ST	UN	2	BP	29
A. macrorhizus	12Lobes	23	ST	SI	2	BP	54
A. maxwellii	8 Lobes	13	SI	SI	2	BP	47
A. muelleri	6 Lobes	10	UN	SI	2, 4	BP, AP	104
A. operculatus	Curved	9	ST	ST	4, 6	TP, XP	40
A. paeoniifolius	7 Lobes	10	ST	UN	2	BP	69
A. scutatus	5 Lobes	8	ST	SI	6	XP	16
A. tenuispadix	Curved	13	ST	UN	4, 6	AP, XP	30
Amorphophallus sp.1	Curved	8	ST	UN	1, 2, 4	HP, BP, AP	34
Amorphophallus sp.2	6 Lobes	7	UN	UN	2	BP	100
Amorphophallus sp.3	5 Lobes	7	UN	SI	2	BP	47
Amorphophallus sp.4	5 Lobes	8	ST	SI	2	BP	42

[a] ST = straight-sided anticlinal walls, SI = sinuous anticlinal walls, UN = undulate anticlinal walls, HP = hemiparacytic, 1 subsidiary cell, BP = brachyparacytic, 2 subsidiary cells, AP = amphibrachyparacytic, 4 subsidiary cells, TP = paratetracytic, 4 subsidiary cells, XP = parahexacytic, 6 subsidiary cells.

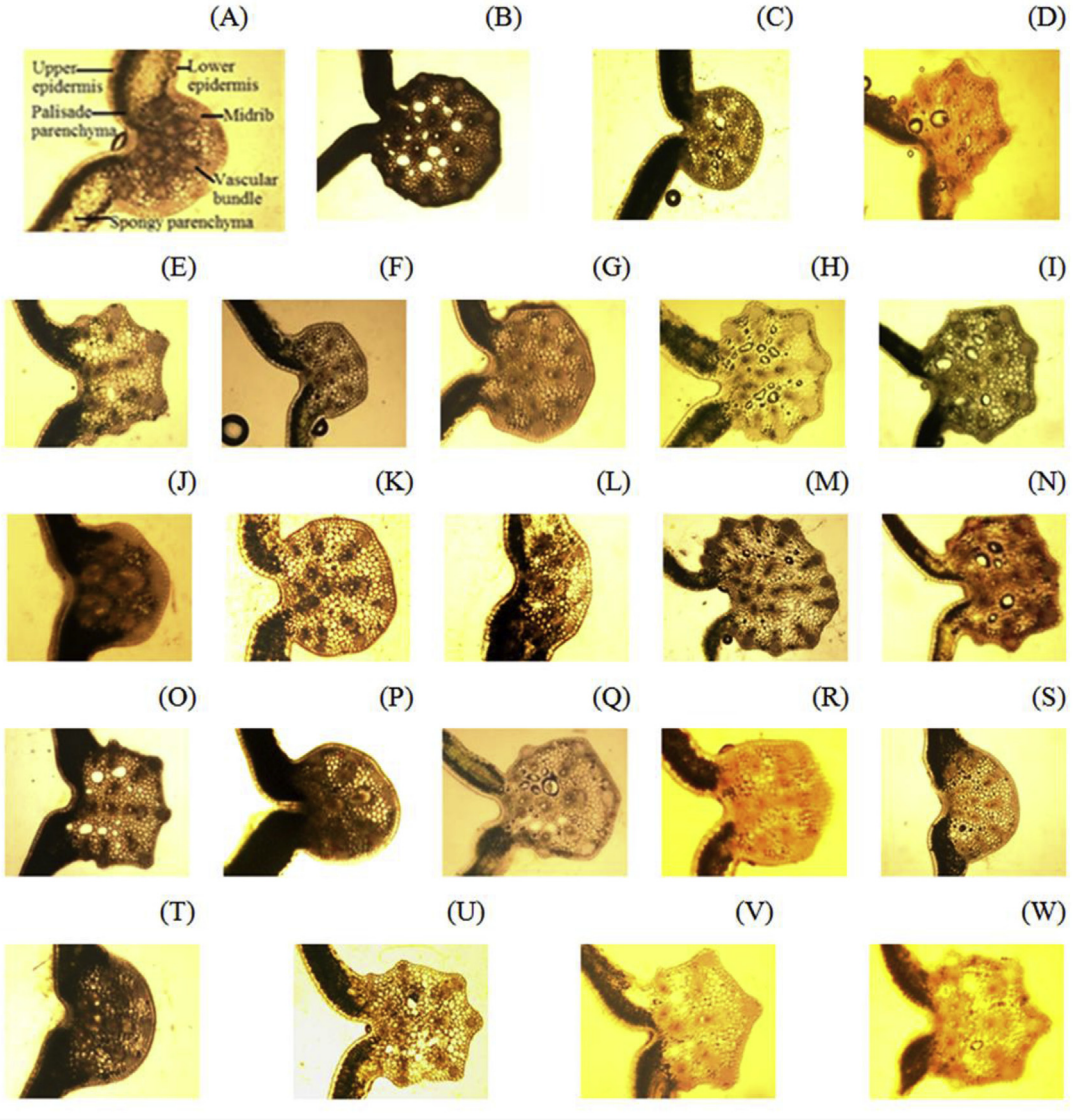

Fig. 1. Midrib of leaflet cross section (40×): (A) *Amorphophallus albispathus* Hett.; (B) *A. amygdaloides* Hett. & Sizemore; (C) *A. asterostigmatus* Bogner & Hett.; (D) *A. bulbifer* (Roxb.) Blume; (E) *A. cruddasianus* Prain ex Engl.; (F) *A. curvistylis* Hett.; (G) *A. flotoi* (S.Y. Hu) ined; (H) *A. koratensis* Gagnep.; (I) *A. krausei* Engl.; (J) *A. lacourii* Linden & André; (K) *A. longituberosus* (Engl.) Engl. & Gehrm.; (L) *A. macrophyllus* (Gagnep. ex Serebryanyi); (M) *A. macrorhizus* Graib; (N) *A. maxwellii* Hett.; (O) *A. muelleri* Blume; (P) *A. operculatus* Hett. & Sizemore; (Q) *A. paeoniifolius* (Dennst.) Nicolson; (R) *A. scutatus* Hett. & T.C.Chapm.; (S) *A. tenuispadix* Hett.; (T) *Amorphophallus* sp.1; (U) *Amorphophallus* sp.2; (V) *Amorphophallus* sp.3; (W) *Amorphophallus* sp.4.

Amorphophallus macrorhizus Graib, *Amorphophallus maxwellii* Hett., *Amorphophallus muelleri* Blume, *Amorphophallus operculatus* Hett. & Sizemore, *A. paeoniifolius* (Dennst.) Nicolson, *Amorphophallus scutatus* Hett. & T.C. Chapm., *A. tenuispadix* Hett., *Amorphophallus* sp.1, *Amorphophallus* sp.2, *Amorphophallus* sp.3 and *Amorphophallus* sp.4.

The leaves of these species differed in the color, shape, size, latter and number of petiolules (decompound, trisect, pinnatisect, bipinnatisect or dichotomously divided) and the number of leaflets. The results could be classified by comparison with herbarium specimens and literature cited (Hetterscheid, 2012; Royal Botanic Garden, Kew, 2013).

Leaf morphology is an important character for the classification and identification of plant species but many species in the Araceae still needed to be identified using the inflorescences and fruits, for example, *Alocasia* (Boyce and Sookchaloem, 2012), *Cryptocoryne* (Jacobsen et al., 2012), *Epipremnum* (Boyce, 2012), *Typhonium* (Hetterscheid and Sookchaloem, 2012).

Leaf anatomy

After cultivation in the greenhouse under 70% natural sunlight, the leaflet anatomy was studied and compared among species. Leaflet cross sections and epidermal layers were studied using a light microscope. Lamina or leaf blades were dorsiventral, with the upper mesophyll or palisade parenchyma being different from the lower mesophyll or spongy parenchyma. Leaflet tissues were composed of: 1) an upper (adaxial) epidermis which lacks stomata; guard cells and subsidiary cells; 2) palisade parenchyma elongate and green cells compact under the upper epidermis; 3) spongy parenchyma, with cells arranged loosely under the palisade parenchyma; 4) a lower (abaxial) epidermis with stomata and epidermal cells; and 5) vascular bundles distributed in the mesophyll and having many sizes in the midrib, veins and veinlets. Species could be identified by the shapes of the midrib (Table 1 and Fig. 1), stomatal types and the shapes of upper epidermis and lower epidermis (Table 1 and Figs. 2–4).

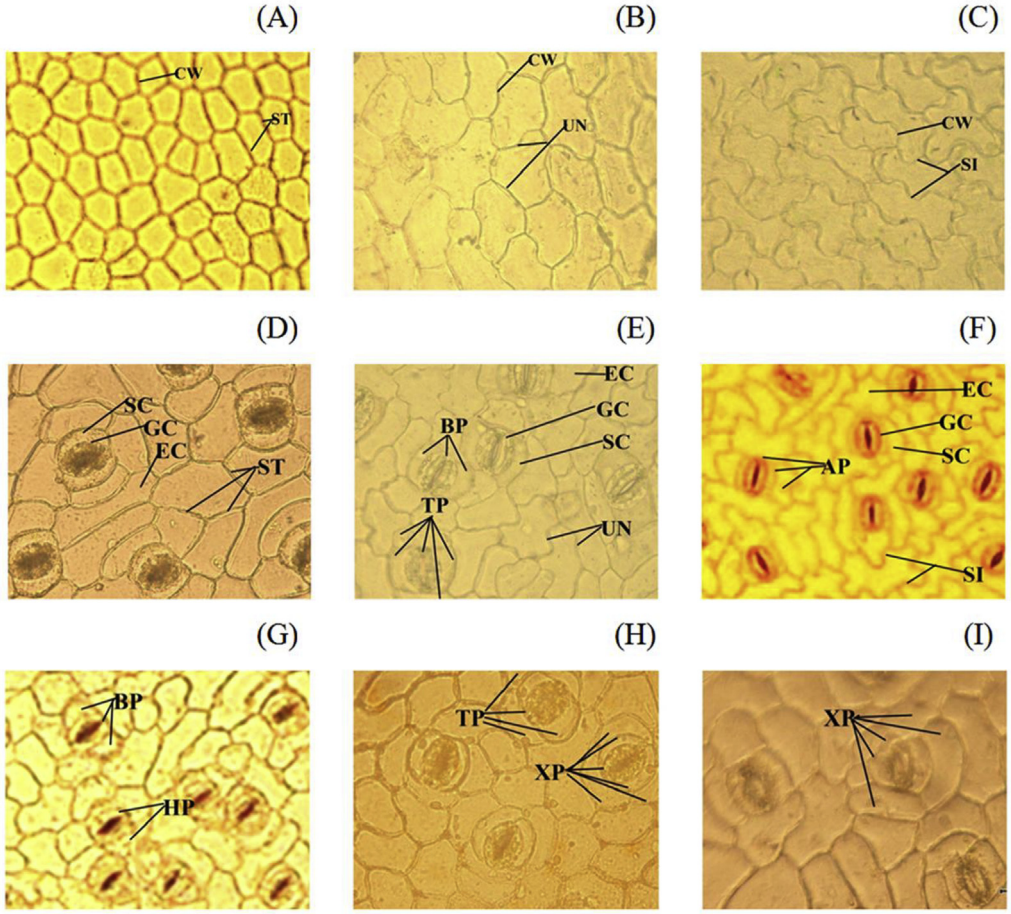

Fig. 2. Upper epidermis of leaflet (100×): (A) Straight-sided anticlinal walls (ST) of *Amorphophallus koratensis*; (B) Undulate anticlinal walls (UN) of *A. asterostigmatus*; (C) Sinuous anticlinal walls (SI) of *A. krausei*; (D) Lower epidermis of leaflet, straight-sided anticlinal walls (ST), *Amorphophallus* sp.1, stomatal subtype BP; (E) Undulate anticlinal walls (UN), *A. longituberosus*, stomatal subtype BP and TP; (F) Sinuous anticlinal walls (SI), *A. bulbifer* stomatal subtype AP; (G) *A. amygdaloides* (40×), stomatal subtype HP and BP; (H) *A. operculatus*, stomatal subtype TP and XP; (I) *A. curvistylis* stomatal subtype XP. Lower and upper epidermal cells labels are: cell wall (CW), straight-sided anticlinal walls (ST), sinuous anticlinal walls (SI), undulate anticlinal walls (UN), guard cells (GC), subsidiary cells (SC), epidermal cell (EC). Stomata types labels are: hemiparacytic (HP) with one subsidiary cell, brachyparacytic (BP) with two subsidiary cells, amphibrachyparacytic (AP) with four subsidiary cells, paratetracytic (TP) with four subsidiary cells and parahexacytic (XP) with six subsidiary cell arrangements in the paracytic types which varied among species in this study.

In their natural environment, *Amorphophallus* spp. tubers are always dormant in the soil in the arid season and may not show any inflorescences when the tubers are still young and even in mature tubers if growing under unsuitable conditions. It is difficult to identify species by comparing only the morphological characters of the tuber or leaf and leaflet in the growing season. Thus, the study of stem and leaf anatomical characteristics is one of the methods to classify and identify species when plant specimens have no flowers or fruits.

Midrib

There were six types of midrib cross section shapes (Table 1 and Fig. 1): 1) a curved shape occurred in eight species—*A. albispathus*, *A. asterostigmatus*, *A. flotoi*, *A. lacourii*, *A. macrophyllus*, *A. operculatus*, *A. tenuispadix*, *Amorphophallus* sp.1 (Fig. 1A, C, G, J, L, P, S and T, respectively); 2) a five lobed shape was present in four species—*A. curvistylis*, *A. scutatus*, *Amorphophallus* sp.3 and *Amorphophallus* sp.4 (Fig. 1F, R, V and W, respectively); 3) a six-lobed shape was found in three species—*A. cruddasianus*, *A. muelleri* and *Amorphophallus* sp.2 (Fig. 1E, O and U, respectively); 4), a seven-lobed shape was found in four species—*A. krausei*, *A. longituberosus*, *A. bulbifer*, and *A. paeoniifolius* (Fig. 1D, I, K and Q, respectively); 5) an eight-lobed shape was present in three species—*A. koratensis*, *A. amygdaloides* and *A. maxwelli* (Fig. 1B, H and N, respectively); and 6) a twelve lobed shape was found only in *A. macrorhizus* (Fig. 1M).

Furthermore, the number of vascular bundles scattered in the midrib was also different with 5, 6, 7, 8, 9, 10, 12, 13, 15, 16 or 23 vascular bundles (Table 1). Five bundles occurred in *A. lacourii* (Fig. 1J), six bundles in *A. macrophyllus* (Fig. 1L), seven bundles in *Amorphophallus* sp.2 and *Amorphophallus* sp.3 (Fig. 1U and V, respectively), eight bundles in five species—*A. cruddasianus*, *A. curvistylis*, *A. scutatus*, *Amorphophallus* sp.1 and *Amorphophallus* sp.4 (Fig. 1E, F, R, T and W respectively), nine bundles in *A. bulbifer* and *A. operculatus* (Fig. 1D and P), 10 bundles in four species—*A. flotoi*, *A. krausei*, *A. muelleri*, and *A. paeoniifolius* (Fig. 1G, I, O and Q, respectively), 12 bundles in *A. longituberosus* (Fig. 1K), 13 bundles in four species—*A. albispathus*, *A. asterostigmatus*, *A. maxwellii* and *A. tenuispadix* (Fig. 1A, C, N and S, respectively), 15 bundles in *A. koratensis* (Fig. 1H), 16 bundles in *A. amygdaloides* (Fig. 1B) and 23 bundles in *A. macrorhizus* (Fig. 1M).

Upper epidermis and lower epidermis

The type and shape of the upper epidermis and lower epidermis of the leaflet were different in each species. There were no guard

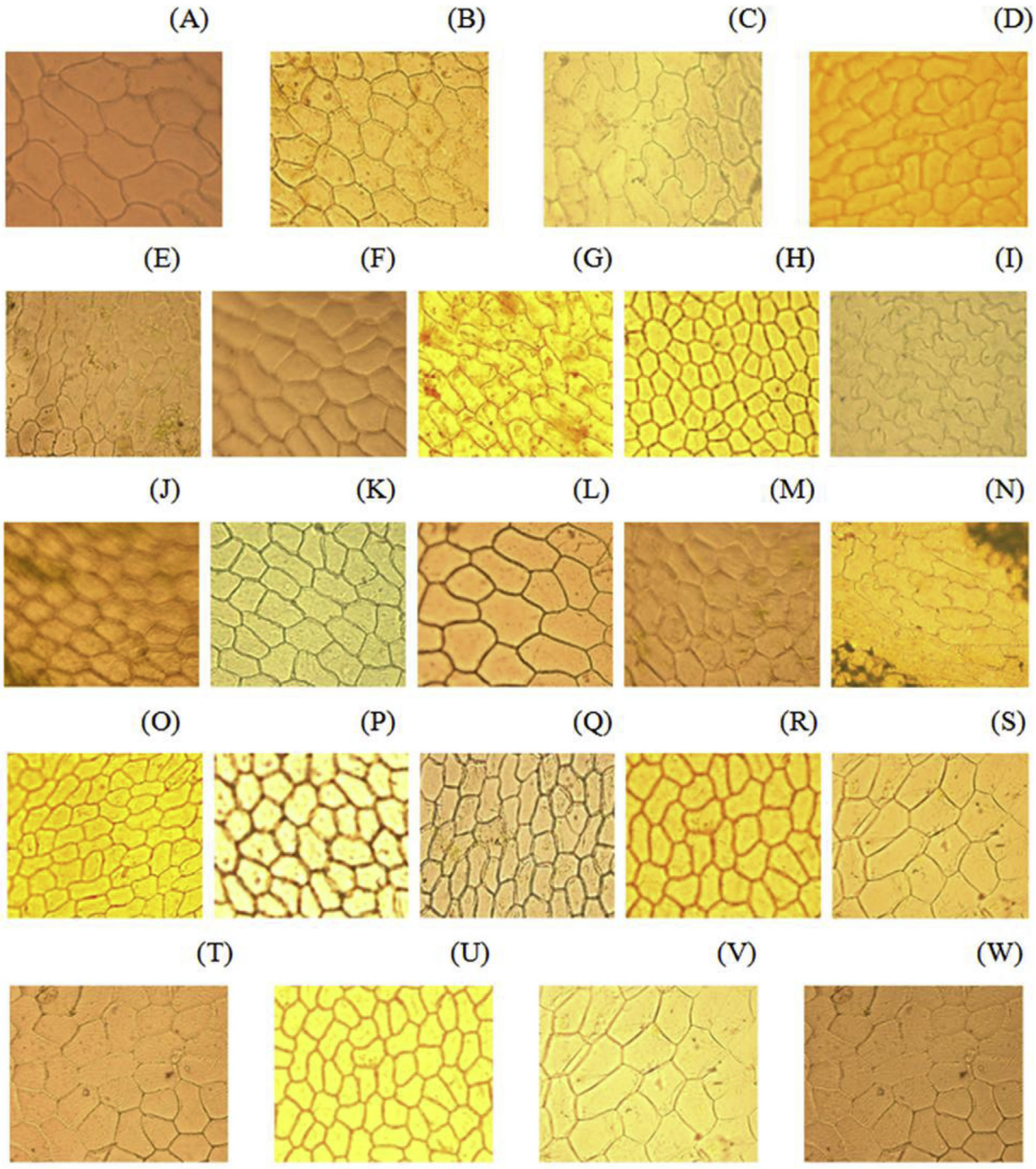

Fig. 3. Upper epidermis of leaflet (100×): (A) *Amorphophallus albispathus*; (B) *A. amygdaloides*; (C) *A. asterostigmatus*; (D) *A. bulbifer*; (E) *A. cruddasianus*; (F) *A. curvistylis*; (G) *A. flotoi*; (H) *A. koratensis*; (I) *A. krausei*; (J) *A. lacourii*; (K) *A. longituberosus*; (L) *A. macrophyllus*; (M) *A. macrorhizus*; (N) *A. maxwellii*; (O) *A. muelleri*; (P) *A. operculatus*; (Q) *A. paeoniifolius*; (R) *A. scutatus*; (S) *A. tenuispadix*; (T) *Amorphophallus* sp.1; (U) *Amorphophallus* sp.2; (V) *Amorphophallus* sp.3; (W) *Amorphophallus* sp.4. Straight-sided anticlinal walls, ST (B, E–M, P–T, W), sinuous anticlinal walls, SI (I, N), undulate anticlinal walls, UN (A, C, D, O, U, V).

cells, while the subsidiary cells were composed of stomata on the upper epidermis or adaxial epidermis. These cells were found only on the layer of the lower epidermis or abaxial epidermis. Three shapes of epidermal cells were separated by anticlinal walls—straight-sided anticlinal walls (ST), sinuous anticlinal walls (SI) and undulate anticlinal walls (UN) (Fig. 2A–C). The upper and lower epidermal cell shapes of four species were the same, but in many species they were different. The upper and lower epidermal cells of *A. krausei* (Figs. 2C, 3I and 4I) and *A. maxwellii* (Figs. 3N and 4N) had sinuous anticlinal walls, *Amorphophallus* sp.2 (Figs. 3U and 4U) had undulate anticlinal walls, *A. lacourii* (Figs. 3J and 4J) and *A. operculatus* (Figs. 2H, 3P and 4P) had straight-sided anticlinal walls (Table 1). Different shapes of the upper epidermis and lower epidermis were found in the other 19 species.

The shapes of the epidermal cells could be separated into three groups. In the first group, the upper epidermal cells had undulate anticlinal walls (UN) but the lower epidermal cells had sinuous anticlinal walls (SI)—*A. albispathus* (Figs. 3A and 4A), *A. asterostigmatus* (Figs. 2B, 3C and 4C), *A. bulbifer* (Figs. 2F, 3D and 4D), *A. muelleri* (Figs. 3O and 4O) and *Amorphophallus* sp.3 (Figs. 3V and 4V). In the second group, the upper epidermal cells had straight-sided anticlinal walls (ST) and the lower epidermal cells had undulate anticlinal walls (UN)—*A. amygdaloides* (Figs. 2G, 3B and 4B), *A. curvistylis* (Figs. 2I, 3F and 4F), *A. koratensis* (Figs. 2A, 3H and 4H), *A. longituberosus* (Figs. 2E, 3K and 4K), *A. paeoniifolius* (Figs. 3Q and 4Q), *A. tenuispadix* (Figs. 3S and 4S), *Amorphophallus* sp.1 (Figs. 2D, 3T and 4T), *A. flotoi* (Figs. 3G and 4G) and *A. macrophyllus* (Figs. 3L and 4L). In the third group, the upper epidermal cells had

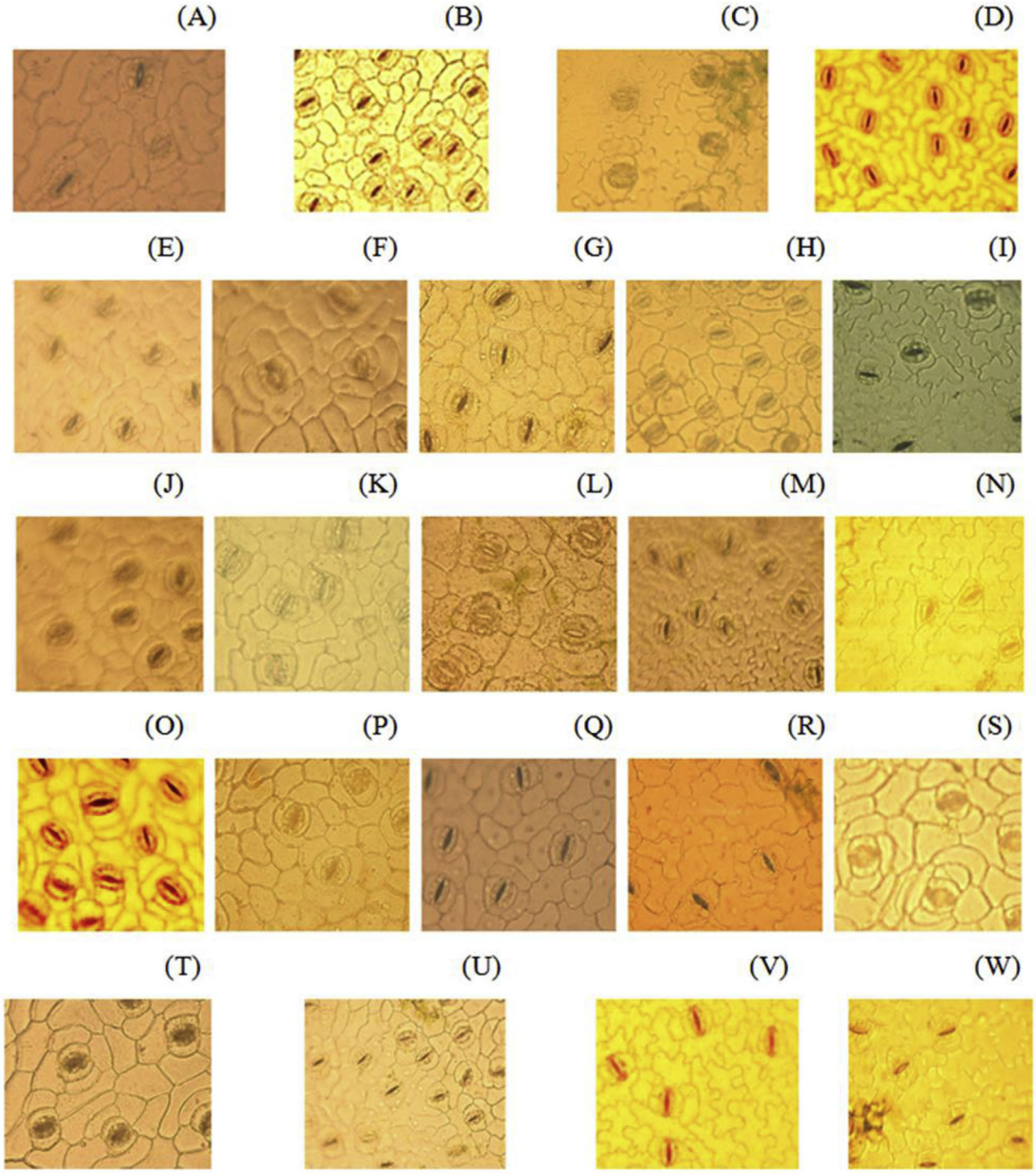

Fig. 4. Lower epidermis and stomata types of leaflet (100×): (A) *Amorphophallus albispathus*; (B) *A. amygdaloides*; (C) *A. asterostigmatus*; (D) *A. bulbifer*; (E) *A. cruddasianus*; (F) *A. curvistylis*; (G) *A. flotoi*; (H) *A. koratensis*; (I) *A. krausei*; (J) *A. lacouri*; (K) *A. longituberosus*; (L) *A. macrophyllus*; (M) *A. macrorhizus*; (N) *A. maxwellii*; (O) *A. muelleri*; (P) *A. operculatus*; (Q) *A. paeoniifolius*; (R) *A. scutatus*; (S) *A. tenuispadix*; (T) *Amorphophallus* sp.1; (U) *Amorphophallus* sp.2; (V) *Amorphophallus* sp.3; (W) *Amorphophallus* sp.4. ST = straight-sided anticlinal walls (Figures J and P), SI = sinuous anticlinal walls (Figures A, C, D, E, I, M, N, O, R, V, W), UN = undulate anticlinal walls (Figures B, F, G, H, K, L, Q, S, T, U).

straight-sided anticlinal walls (ST) and the lower epidermal cells had sinuous anticlinal walls (SI)—*A. cruddasianus* (Figs. 3E and 4E), *A. macrorhizus* (Figs. 3M and 4M), *A. scutatus* (Figs. 3R and 4R) and *Amorphophallus* sp.4 (Figs. 3W and 4W). The cell sizes of each species were not equal at the same magnification (Figs. 3 and 4).

Stomatal types in lower epidermis

The lower epidermis had guard cells, subsidiary cells and epidermal cells. Two guard cells were composed as a stoma which was also different in its size, shape and density in each species. Subsidiary cells were different in their size, shape, number and arrangement and could be divided into stomatal subtypes. There were 1, 2, 3, 4 and 6 subsidiary cells and five subtypes of stomatal.

The paracytic types were characterized by lateral subsidiary cells which were oriented parallel to the sides of the guard cells (Dilcher, 1974; Prabhakar, 2004; Carpenter, 2005); the hemiparacytic (HP) subtype consisted of only one lateral subsidiary cell on one side, the brachyparacytic (BP) subtype consisted of two lateral subsidiary cells, the amphibrachyparacytic (AP) subtype consisted of four lateral subsidiary cells, the paratetracytic (TP) subtype consisted of two lateral subsidiary cells and two short cells oriented in the upper and lower guard cells, but the parahexacytic (XP) subtype had four lateral subsidiary cells and two short cells oriented in the upper and lower guard cells and was called parahexacytic (XP) (Fig. 2H and I).

Thirteen species had only two subsidiary cells and brachyparacytic (BP) stomata (Table 1)—*A. albispathus* (Fig. 4A),

A. asterostigmatus (Fig. 4C), *A. cruddasianus* (Fig. 4E), *A. koratensis* (Fig. 4H), *A. krausei* (Fig. 4I), *A. lacourii* (Fig. 4J) and *A. macrophyllus* (Fig. 4K). *A. macrorhizus* (Fig. 4M), *A. maxwellii* (Fig. 4N), *A. paeoniifolius* (Fig. 4Q), *Amorphophallus* sp.2 (Fig. 4U), *Amorphophallus* sp.3 (Fig. 4V) and *Amorphophallus* sp.4 (Fig. 4W).

A. scutatus had six parahexacytic (XP) subsidiary cells (Fig. 4R). *A. amygdaloides* had one hemiparacytic (HP) subsidiary cell and two brachyparacytic (BP) subsidiary cells (Figs. 2G and 4B). *A. bulbifer* had four amphibrachyparacytic (AP) subsidiary cells (Figs. 2F and 4D). *A. longituberosus* had two brachyparacytic (BP) subsidiary cells and four paratetracytic (TP) subsidiary cells (Figs. 2E and 4K). *A. flotoi* (Fig. 4J) and *A. muellerii* (Fig. 4O) had two brachyparacytic (BP) subsidiary cells and four amphibrachyparacytic (AP) subsidiary cells. *A. operculatus* (Figs. 2H and 4P) had four paratetracytic (TP) subsidiary cells and six parahexacytic (XP) subsidiary cells. *A. tenuispadix* (Fig. 4S) had four amphibrachyparacytic (AP) subsidiary cells and six parahexacytic (XP) subsidiary cells. The other two species—*A. curvistylis* (Figs. 2I and 4F) and *Amorphophallus* sp.1 (Figs. 2D and 4T)—had 1, 2, 4 or 6 subsidiary cells but had three stomatal subtypes, being BP, TP, XP and AP, BP, HP, respectively.

The epidermal cells of *Amorphophallus* had three shapes—straight-sided anticlinal walls, undulate anticlinal walls and sinuous anticlinal walls. Stomata have been reported to be brachyparacytic with six subsidiary cells (Keating, 2003). The result was different in the current study, where the leaflet anatomy of *Amorphophallus* spp. was different in each species. The subsidiary cell numbers were 1, 2, 4 or 6 cells and the stomatal subtypes were hemiparacytic, brachyparacytic, amphibrachyparacytic, paratetracytic and parahexacytic. Ground tissues had small cavities, vascular bundles were small, having no trichosclereid, non-anastomosing laticifers and secretory ducts were lined without epithelial cells (Keating, 2003). Prismatic crystals, druse crystals and raphide crystals were found in wide, elongated, or tubular cells, with two or more raphide bundles in a single cell; individual cells were articulated end to end, forming articulated raphide tubes, with thin-walled, spindle-shaped raphide cells (Sungkajanttranon and Sookchaloem, 2014).

The stomatal numbers per leaf area were different in each species, ranging from 16/mm^2 to 104/mm^2 (Table 1 and Fig. 4). Numbers were low in the glossy leaves of *A. albispathus*, *A. asterostigmatus*, *A. macrophyllus*, *A. tenuispadix* and *A. scutatus*, while the large, thin and dull leaflets of *A. muelleri* and *Amorphophallus* sp.2 had many stomata per leaf area, with 104/mm^2 and 100/mm^2, respectively (Table 1 and Fig. 4). Nevertheless, the shape, size and arrangement of stomata also varied among species.

Epiphytic and hemiepiphytic stem anatomical studies have been reported for 14 species of *Philodendron* (Tenorio et al., 2012) and 11 genera of Pothoideae in the Araceae (French and Tomlinson, 1981). The results showed vascular patterns of the stem in six groups, but identification was not possible to the species level. Stem anatomical study of *Amorphophallus* is very difficult to use to identify to the species level because the underground stems are large and filled with glucomannan, water, anthocyanin, laticifer and there are many vascular bundles scattered inside (Sungkajanttranon et al., 2010). The results of the leaflet anatomy of *Amorphophallus* spp. in the current study provide confidence in microscopic studies of plant structure to classify and identify species, rather than depending solely on leaf morphology.

Nevertheless, under higher or lower light intensity, soil moisture content, temperature and relative humidity conditions, if these species were grown under both greenhouse conditions and in natural areas, the leaf morphology and leaf anatomy could be different from the current study. The effect of light intensity and ambient CO_2 has been reported on different levels of photosynthesis in hybrid Araceae for interior landscaping using *Aglaonema commutatum* 'Sylver Queen', *Anthurium andreanum* 'Dakota', *Dieffenbachia picta* 'Camilla', *Philodendron erubescens* 'Red Emerald', *Spathiphyllum wallisi* 'Mauna Loa' and *Syngonium podophyllum* 'Maya Red' (Giorgioni and Neretti, 2010). High light intensity at 20% can also affect the leaf thickness of *Alocasia macrorrhizos* by more than 1% (Sim and Pearcy, 1992), but Fan et al. (2012) reported low light intensity can also affect the leaf thickness and number of stomata per unit of leaf area of young tomato leaves, with amounts under 300 μmol/m^2 s being higher than for 450–550 μmol/m^2 s (1882.64/mm^2 and 1603.69–1678.28/mm^2, respectively). Further study of the characters of leaflets and other organs is required for species identification.

Leaf morphology and leaflet anatomy characteristics can be used to identify *Amorphophallus* species if the specimens are reproductively sterile. Leaf morphology and leaflet anatomy characteristics could be applied for collection and cultivation using simple methods and provide confidence to be able to accurately identify to the species level. Tuber and leaf morphology alone are insufficient to distinguish among species. The anatomical characters of leaflets could help to identify species more correctly than using only morphological characters.

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