



**Citation:** Mohd N. Ghazalli et al. (2020) *Nepenthes latiffiana* and *N. domei* (Nepenthaceae), two new species of pitcher plants from Terengganu, Peninsular Malaysia. *Webbia. Journal of Plant Taxonomy and Geography* 75(1): 5-28. doi: 10.36253/jopt-7950

**Received:** January 29, 2020

**Accepted:** April 30, 2020

**Published:** June 30, 2020

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**Data Availability Statement:** All relevant data are within the paper and its Supporting Information files.

**Competing Interests:** The Author(s) declare(s) no conflict of interest.

**Editor:** Riccardo M. Baldini, University of Florence, Italy

## *Nepenthes latiffiana* and *N. domei* (Nepenthaceae), two new species of pitcher plants from Terengganu, Peninsular Malaysia

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**Abstract.** Two new species of *Nepenthes* from Terengganu, Peninsular Malaysia, *N. latiffiana* M. N. Faizal, A. Amin & N. Dome and *N. domei* M. N. Faizal, A. Amin & A. Latiff, are described and illustrated.

**Keywords:** pitcher plants, carnivorous plants, *Nepenthes*, *Nepenthes domei*, *Nepenthes latiffiana*, Terengganu, Peninsular Malaysia.

### INTRODUCTION

In Peninsular Malaysia, a total of eleven species of pitcher plants (*Nepenthes*) or locally known as “periuk kera” have been recorded (Jebb and Cheek 1997; Cheek and Jebb 2001; Clarke 2001; McPherson 2009). The first taxonomic account of the genus in Peninsular Malaysia was given by Ridley (1924) who recorded a total of ten species, namely *N. ampullaria* Jack., *N. albomarginata* Lobb., *N. gracillima* Ridl., *N. alba* Ridl., *N. ramispina* Ridl., *N. sanguinea* Lindl., *N. macfarlanei* Hemsl., *N. gracilis* Korth., *N. rafflesiana* Jack and *N. phyllamphora* Willd. (= *N. mirabilis* (Lour.) Druce). After Ridley’s

treatment, there have been some further researches on the genus by Holttum (1940), Kiew (1990), Turner (1995), Clarke (2002), Adam et al. (2005) Latiff et al. (2011), Clarke and Lee (2012) and Latiff and Norsiah (2016) regarding the diversity, distribution, and ecological study of *Nepenthes* species and natural hybrids. Another taxonomic study of the Peninsular Malaysian taxa was carried out by Rohana in 1988 that covers basic anatomical features and systematic study of the species and hybrids. Turner (1995) listed ten species, which instead included two natural hybrids, namely *N. × hookeriana* and *N. × trichocarpa*, with *N. alba* and *N. ramispina* being excluded. Clarke (1999) added *N. benstonei*, a species then known from Bukit Bakar, Kelantan, to the list and the latest was Adam and Hamid (2007) who described *N. sharifah-hafsahii* which happens to be a natural hybrid between *N. gracilis* and *N. mirabilis* rather than a true species (McPherson 2009).

The most comprehensive and well accepted taxonomic enumeration works of the genus were given by Jebb and Cheek (1997), Cheek and Jebb (2001) and Clarke (2001) who recognised eleven species. In the course of continuous field trips to the state of Terengganu, the authors had encountered several populations of pitcher plants in Setiu whose features did not match the currently described taxa of Peninsular Malaysia. From this finding, we further analysed the morphological, anatomical and micromorphological characteristics and have discovered two distinct new species (as described in this article). The descriptions of *N. domei* and *N. latiffiana* herein can be considered as a comprehensive work in the taxa identification as the sheet contains the sterile/fertile parts and pitchers, anatomical and micromorphological evidences, and enumeration of prominent plant characteristics. We also conducted a comparative molecular study on *N. domei* using internal transcribed spacer (ITS) DNA region to further differentiate it from a closely related species.

## MATERIALS AND METHODS

Analyses of morphological characteristics on new *Nepenthes* specimens were based on living plants observed *in situ* and also from herbarium type specimens (MDI12424 and MDI12423) deposited at MARDI Herbarium (MyGenebank™ Complex MARDI Serdang).

For leaf anatomy analysis, collected and described *Nepenthes* leaves were fixed in AA solution (1 Acetic acid : 3 Alcohol). Investigated leaf parts including midrib, lamina and margin were sectioned in a range of thickness from 15-30 µm using sliding microtome. The leaves

then were cleared using bleaching agent, washed in various alcohol series and stained in Safranin and Alcian blue for 5 minutes each before proceeded for dehydration stage through an ascending alcohol series (50-100%). During the final dehydration stage, the leaf specimens were differentiated in 70% alcohol with a drop of hydrochloric acid (HCL) before mounted in Euparal. Specimen slides were kept in the oven for two weeks at about 60-70°C. Photographs of the investigated sections were taken using Olympus SZH40 microscope and the images were processed using CellSens Image Analysis software.

For micromorphology study, *Nepenthes* epidermis preparative fragments about 3 mm long were excised from the middle portion of approximately three mature leaves of each species. Cuticle parts were prepared by soaking leaf blade fragments in 5-10% aqueous chromium trioxide until all organic material except the cuticle was dissolved. Targeted leaf cuticles were mounted on aluminium stubs with double-sided adhesive tape and proceeded for air drying. The stubs were then sputter-coated with pure gold to a maximum thickness of 15 nm and examined with LEO (Model 1450 SEM) - Field-Emission Scanning Electron Microscope (FESEM). The remaining cuticles were soaked in 5% ammonia and mounted on microscope slides in Canada balsam.

Results of the adaxial and abaxial epidermal layers under scanning electron microscope (SEM) and leaf anatomical study of *N. domei*, *N. latiffiana*, *N. benstonei* and *N. sanguinea* are shown in Figure 5 to 12 and summarised in Table 2 and 3. These results are combined with transverse section of the leaf lamina and margin showing epidermal cells, mucilaginous idioblast, glandular and simple trichomes, solitary crystals observation on the parenchyma and along a vein, and significant venation characters. The systematic significant of leaf anatomy and micromorphology evidences differentiating these investigated *Nepenthes* species are presented in Table 2 and 3.

Finally, for molecular study, all genomic DNA purifications were done according to DNeasy Plant Mini Kit (Qiagen) manufacturer's protocol and matured leaves (100 mg per sample) from *N. domei* sp. nov. (Terengganu), *N. benstonei* (Bukit Bakar, Kelantan) and *N. sanguinea* (Fraser's Hill, Pahang) were used. The internal transcribed spacer (ITS) partial gene (~610bp) was PCR-amplified from *Nepenthes* DNA using universal ITS1-2 primers (ITS\_F: 5'-AGGAGAAGTCGTAACAAGGTT; ITS\_R: 5' GATGCAACCTTGGCCTT) and Q5 High-Fidelity DNA polymerase (New England Biolabs, UK). The parameters for thermocycler were set as follows: initial denaturation at 98°C for 30 s, denaturation at 98°C

for 10 s, annealing at 61°C for 15 s, extension at 72°C for 20 s, and final extension at 72°C for 2 min. The amplicons were sent for Sanger sequencing and the sequencing data was trimmed and aligned using BioEdit Sequence Alignment Editor. Phylogenetic inference of nine peninsular *Nepenthes* species (including *N. domei* sp. nov.) was reconstructed in Molecular Evolutionary Genetics Analysis software (MEGA5) using Neighbor-Joining (NJ) method (1000 bootstraps) based on the ITS partial sequences isolated in this study as well as sequences obtained from the NCBI database (Bunawan et al. 2017; Alamsyah and Ito 2013; Renner and Specht 2011).

#### TAXONOMIC TREATMENT

***Nepenthes latiffiana*** M. N. Faizal, A. Amin & N. Dome, sp. nov. (Figures 1 & 2)

Type: Malaysia, Peninsular Malaysia, Terengganu, Setiu, 30 April 2019, MNFG751 (MDI!), MDI12424, *Mohd. Norfaizal, Amin Asyraf, Dome Nikong, Muhamad Ikhwan, Edward Entalai & Anuar Rasyidi*, [holotypus, MDI]

#### Diagnosis

*Nepenthes latiffiana* differs from *N. sanguinea* in peristome morphology which is considerably developed, loosely cylindrical, with expanded outer margin part towards both sides of the mouth forming flap-like structure which is especially prominent in upper pitcher (vs. simple, expanded, outer margin usually markedly sinuate where the peristome is widest); climbing stem with simple hairs, cross section cylindrical (vs. glabrous, sharply angular) and lid margin slight wavy to wavy that retains its morphology in dried and wet preserved specimens (vs. flat).

#### Description

Terrestrial climber to 0.5-1.5 m tall. *Climbing stems* cylindrical to rounded, particularly towards the leaf nodes, 0.8-1.2 cm in diameter. *Internodes* 2.2-3.5 cm long. *Leaves* coriaceous, sessile to less amplexicaul, linear-lanceolate, apex obtuse to emarginated, 15-20 cm long, 5-6 cm wide; base clasping stem for about ½ of its circumference; longitudinal veins 1-3 on each side of the midrib, pennate nerves conspicuous; tendril with slender formation in the middle, 10-14 cm long, with minute hairs. *Lower pitchers* up to 18-26.5 cm long, 4.9-8 cm wide, fleshy coriaceous texture, arising abruptly from the tendril; broad cylindrical at the lower part of the pitcher, ½ upper of the pitcher slightly swollen with hip formation, 9 cm length, up to 7 cm above of the hip

till nearly closer to the mouth; cylindrical above, with slight widening towards the mouth, surface of the hip smooth; two fringed wings up to 0.9-1.0 cm wide, runs along the pitcher and widest at the mouth, with multicellular hair elements up to 0.3-0.6 cm long; spur with branched form, reaching 1.0 cm long; mouth rounded; peristome loosely cylindrical but widening or expanded towards lateral sides of the pitcher mouth (ca. 1.5 cm wide each side), outer surface smooth, reaching 1.1 cm wide (in front of the mouth), ribs (at the part of the peristome) reaching 1.6 cm wide; lid rounded, up to 6.5 cm long and 6 cm wide, base retuse, without appendages or trichomes on the lower surface of the lid, ~105-126 dotted nectar glands superfluous and dense at the center and base of the lid. *Upper pitchers* up to 20-23.5 cm long, 5-7.5 cm wide, coriaceous texture, arising slight abruptly from the tendril; broad cylindrical at the ¼ lower part of the pitcher, ¾ upper of the pitcher cylindrical and slight narrowing towards the mouth; with minute wings; mouth ovoid to sub ovoid, peristome cylindrical but with expanded lateral sides (1.3-1.4 cm wide each side) and flattened towards the front of the mouth part, outer surface smooth, reaching 9mm wide, ribs reaching 1.3 mm wide; lid rounded, up to 4.8-5.2 cm long and 5.5 cm wide, base cordate, without appendages or trichomes on the lower surface of the lid. *Male inflorescence* a raceme, peduncle up to 16 cm, rachis up to 40 cm, partial peduncles single-flowered, bracts absent, pedicel 10-13 mm long, tepal ovate, up to 2-5 mm, staminal column 2.5-6 mm long, anther head 2-3.75 mm. *Colour of the living plants*: Lower pitcher combination of green-yellowish-brown hues with several red blotches formation on the 1/3 of the upper part of the pitcher body. Upper pitcher light green with fade specks. Pitcher interior green-yellowish to reddish. Leaves green. Stem green to dark green. *Colour of the dried specimen* light brown – pale brown (lamina, stem and pitcher).

#### Etymology

Named in honour of Emeritus Professor Dato' Dr. A. Latiff Mohamad, a prominent figure in Malaysian Botany Field from Universiti Kebangsaan Malaysia, Bangi Campus, Malaysia.

#### Distribution and habitat

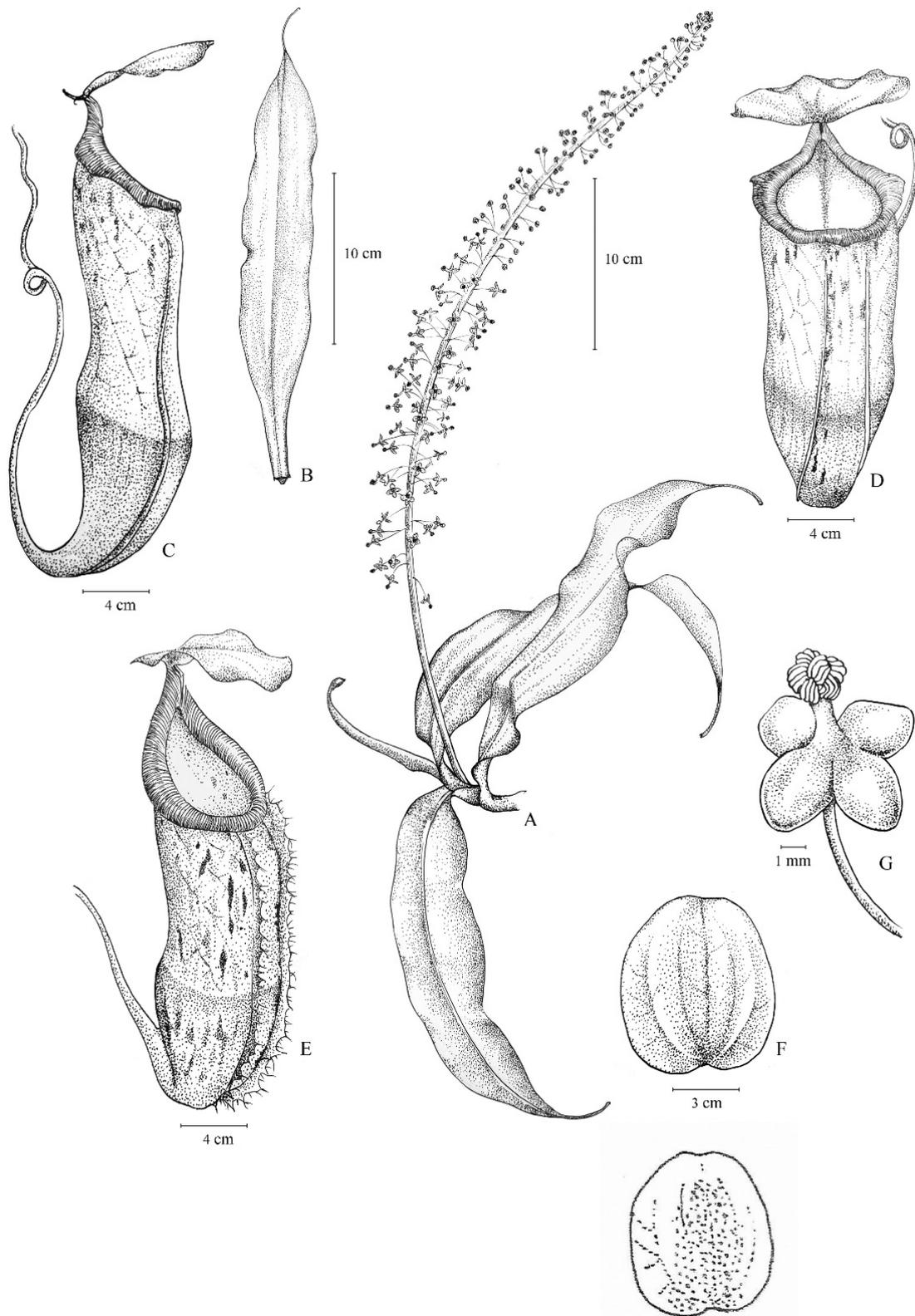
*Nepenthes latiffiana* is currently known only from the restricted area at the type locality in Setiu, Terengganu.

#### Ecology

*Nepenthes latiffiana* occurs as terrestrial climbers on hilly terrains with slight shaded, upper hill forest habitats, at an altitude of c. 1000-1100 m.



**Figure 1.** *Nepenthes latiffiana*, in situ: (A) Lower pitchers. (B) Upper pitcher. (C) Immature upper pitchers. (D) Lid, peristome and waxy inner surface of lower pitcher. (E) Habit showing the lower and upper pitchers. Photographs by Mohd Norfaizal Ghazalli and Amin Asyraf Tamizi.



**Figure 2.** Line drawing of *Nepenthes latiffiana*: (A) Habit, with inflorescence. (B) Leaf blade. (C) Upper pitcher, viewed from side with spur. (D) Upper pitcher frontal view. (E) Lower pitcher. (F) Adaxial and abaxial side of the lid. (G) Male flower. Based on 12424 (MDI). All drawn by Khalid Hashim.

***Nepenthes domei*** M. N. Faizal, A. Amin, & A. Latiff, **sp. nov.** (Figures 3 & 4)

Type: Malaysia, Peninsular Malaysia, Terengganu, Setiu, 30 April 2019, MNFG750 (MDI!), MDI12423, *Mohd. Norfaizal, Amin Asyraf, Dome Nikong, Muhamad Ikhwan, Edward Entalai & Anuar Rasyidi* [holotypus, MDI].

#### Diagnosis

*Nepenthes domei* differs from *N. benstonei* in several lower pitcher characteristics that include thick-leathery coriaceous texture (vs. coriaceous texture), broad rounded formation in the lower 1/3 or 1/2 part of the lower pitcher (vs. broad ovoid – ovoid in the lower part), two fringed wings 5.5-7 mm wide narrowest at the base and widest at the mouth (vs. less than 4mm runs the whole length of the pitcher body), multicellular fringe elements up to 2-4 mm long (vs. overall shorter, 1-3 mm long). The lower pitchers of the new species frequently burrow into humus, a feature that has not observed in *N. benstonei* from Bukit Bakar. The lower pitchers exhibit whitish red to dark ruby red colouration with the majority of individuals sparsely overlain with or entirely absent of specks on the inner and outer wall (vs. brownish red-dark red, inner wall heavily overlain with specks). The inflorescence rachis length 28-35 cm (vs. less than 30-31 cm). Leaves and stems without waxy cuticle (vs. covered with waxy cuticle).

#### Description

Terrestrial climber to 3.5 m tall. *Stem and climbing stems* slight rounded in cross section, up to 0.7-0.9 cm in diameter. *Internodes* 4-8 cm on climbing stems and 1.3-2.5 cm on rosette stems. *Leaves* of lower stem coriaceous, lanceolate, sessile, 12-13 cm long and 4-5 cm wide, sheathing the stem up to ¼ of its circumference. Leaf blades of the climbing stems narrowly linear-lanceolate, sessile, 25-32 cm long, 5-8 cm wide, base contracted into the stem up to ½-¾ of the stem, apex narrow acuminate, tendril insertion simple. Longitudinal nerve 1-2 on each side of the midrib, pennate nerves spreading towards the leaf margins. Leaves of climbing stems with slight differentiation to those of the lower stems which is bigger in size. *Tendrils* coiled, up to 13-37 cm long. *Rosette/lower pitchers* up to 15-17 cm long, 4.4-6.5 cm wide; thick-leathery coriaceous texture; arising slight gradually from the tendril; broad rounded formation at the lower part ca. 1/3 with slight pronounced hip, straight cylindrical towards above, widening slightly towards the mouth. Inner surface of the portion below the lid throughout surfaces above the hip smooth and waxiness, light green in colour; two fringed wings up to 7 mm wide (narrowest at the base and widest at the mouth), run from the

top of the pitcher to the bottom of the pitcher, consist of multicellular fringe elements up to 2-4 mm long; mouth ovoid-rounded and oblique, concave, rising at the rear of the pitcher towards the lid; peristome slight cylindrical, front up to 6 mm wide, sides up to 4 mm, up to 1.2 cm wide at the rear; outer surface with minute unicellular trichomes, lid ovate, up to 2.7 cm long and 2.4-2.5 cm wide, base generally rounded (not cordate), no hairs or appendages observed on the lower surface. ~60-87 dotted - rounded nectar glands scattered on the lower part of the lid, and concentrated at the base of the lid. *Upper pitchers* 10-12 cm long, 1.5-4 cm wide, thin coriaceous; arising gradually from the tendril; slightly infundibular to cylindrical in the lower part, upper part cylindrical and slight widening towards the mouth; peristome similar to the lower pitchers, differs in mouth shape (wide and length of the mouth and peristome); lid narrow ovate, up to 1.8 cm long and 2.3 cm wide, base rounded (not cordate), no hairs or appendages on the lower surface of the lid; spur simple, unbranched less than 0.5-0.6 cm. *Male inflorescence* a raceme, peduncle up to 13 cm, rachis up to 35 cm, partial peduncles single-flowered, bracts absent, pedicel 11-14 mm long, tepal wide ovate, up to 4-6 mm, staminal column 3-4 mm long, anther head 2-2.5 mm. *Colour of living plants* lower pitcher whitish red to dark ruby red throughout or green-red-dish, sometimes with a few red-purple blotches on the inner surfaces and lid, peristome dark red to green with varying degrees of green and red. Leaves and stems without waxy cuticle. Stems reddish-green to green. Leaves dark green. *Colour of dried specimens* dark brown (lamina, stem and pitcher).

#### Etymology

Named in honour of the first person who discovered the population, Mr. Dome Nikong.

#### Distribution

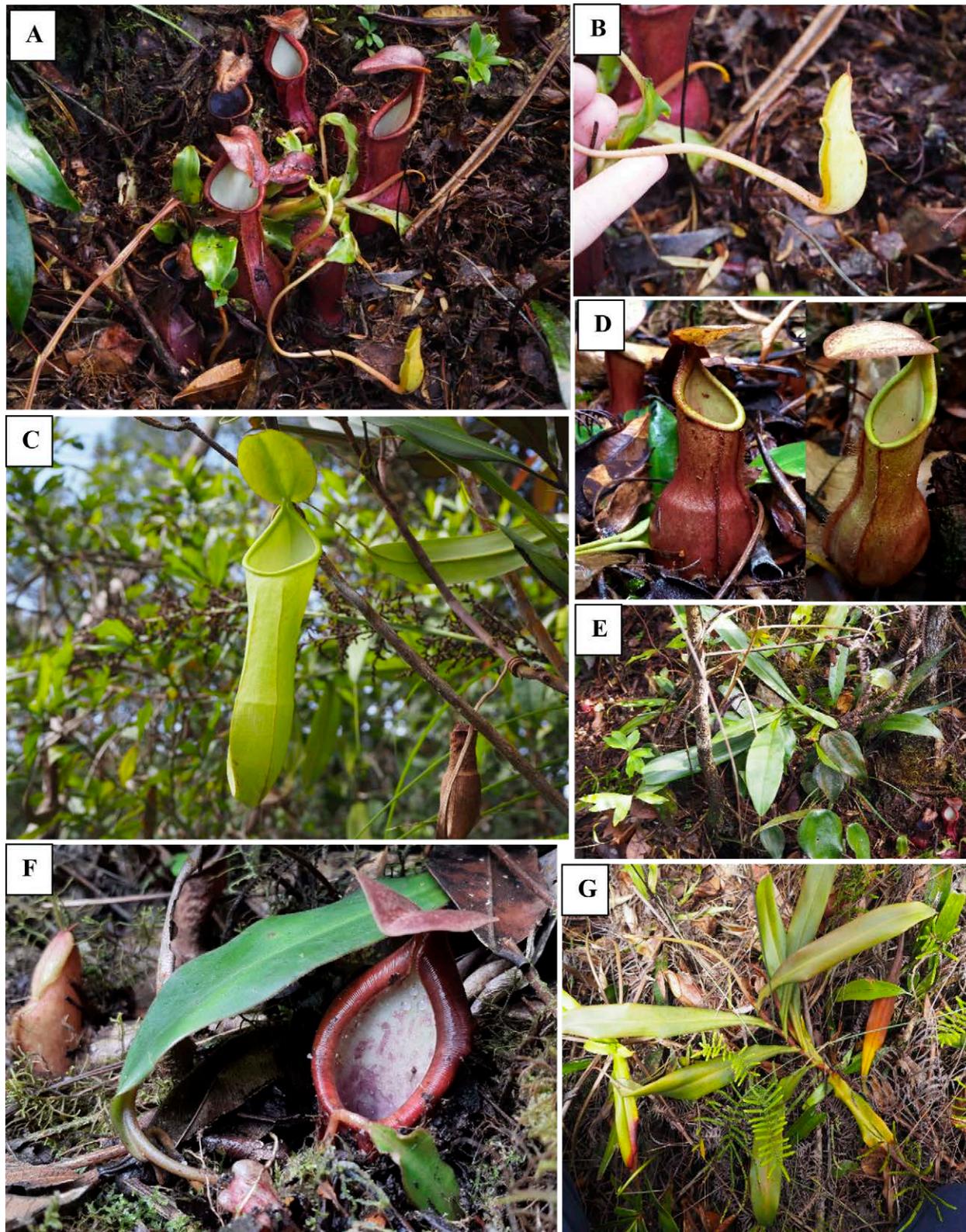
*Nepenthes domei* is currently known only from the restricted area at the type locality in Setiu, Terengganu.

#### Ecology

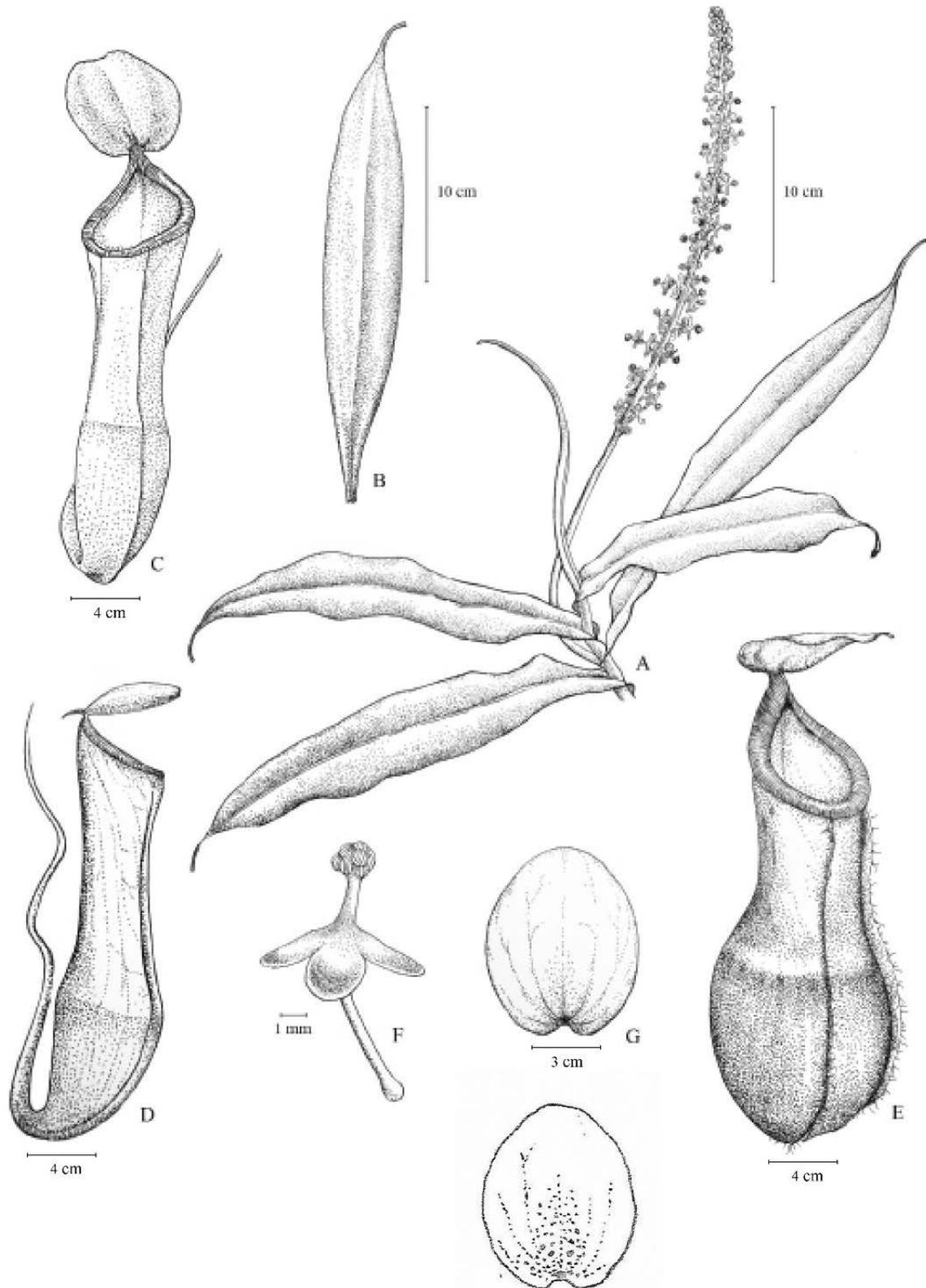
Terrestrial climber in humus rich soil, immature and mature lower/rosette pitchers tend to burrow in the soil rich with humus up to 1/3 – 1/2 of the pitcher. Upper hill forest habitats, at an altitude of c. 850-1000 m.

#### Conservation notes

*Nepenthes domei* and *N. latiffiana* are terrestrial climbers; with *N. latiffiana* observed growing as understorey



**Figure 3.** *Nepenthes domei*, in situ: (A) Unearthed rosette pitchers with rosette leaves. (B) Immature rosette pitcher pulled out from humus rich soil. (C) Upper pitcher. (D) Lid and inner surface of lower pitchers. (E) Leaves from lower stem. (F) Rosette/lower pitchers almost entirely buried in the soil. (G) Habit, showing stem and leaves. Photographs by Mohd Norfaizal Ghazalli and Amin Asyraf Tamizi.



**Figure 4.** Line drawing of *Nepenthes domei*: (A) Habit, with inflorescence. (B) Leaf blade. (C) Upper pitcher. (D) Upper pitcher, viewed from side with spur. (E) Lower pitcher. (F) Male flower. (G) Adaxial and abaxial point view of the lid. Based on 12423 (MDI). All drawn by Khalid Hashim.

plants on a hill slope with rocky substratum at an elevation about c. 1100 m. The vegetation of the locality consisted of various montane species dominated by *Dicranopteris linearis* (Gleicheniaceae), *Dipteris conjugata* (Dipteridaceae) and *Machaerina maingayi* (Cyperaceae), thriving together with rattans, *Pandanus* sp. and *Leptospermum flavescens*. Some climbing stems of *N. domei* were also observed scrambling over *Pandanus* tree. Based on our observation, more than five individuals of *N. domei* were seen at three scattered locations, while only one to two individuals of *N. latiffiana* were recorded at a single location. Both species are known to be restricted in a single mountain since it was discovered. Thus, we assign *N. domei* and *N. latiffiana* in this article as *Vulnerable* under criterion - D2 of IUCN (2001). The habitat had experienced logging in the past; hence, this could explain the limited number of the individuals discovered in the area.

#### Additional specimens examined

MALAYSIA, Kelantan, Machang, Bukit Bakar, 27.9.2006. Yao, T.L. et al. FRI53169 (*N. benstonei*); Malaysia, Terengganu, Gunung Tebu, 31.5.1974. Mohd. Shah et al. FRI13147 (*N. benstonei*); Malaysia, Terengganu, Gunung

Padang, 21.3.2010. Mohd Hairul, M.A. et al. FRI 70887 (*N. sanguinea*); Malaysia, Pahang, Cameron Highlands, Ulu Bertam FR, 26.4.2017. Imin, K. et al. FRI87122 (*N. macfarlanei*).

#### DISCUSSION AND ADDITIONAL TAXONOMIC NOTES

##### 1) Anatomical descriptions

##### *Nepenthes domei*

Holotype: MDI12423

**Leaves anatomy** lamina thickness 292.48-315.76  $\mu\text{m}$  (Figure 9G), **Hypodermis** 1-3 layers on adaxial, thickness 41.52-55.37  $\mu\text{m}$ , 1 layer on abaxial, thickness 39.56-49.73  $\mu\text{m}$ . Mesophyll 227.38-239.78  $\mu\text{m}$ ; palisade parenchyma 1-3 layers with total 157.29-163.21  $\mu\text{m}$  thick; spongy parenchyma total 74.85-89.63  $\mu\text{m}$ , with plenty extracellular space in between of spongy parenchyma (Figure 9G). **Midrib** outline Type I (Figure 9A), 267.89-290.66  $\mu\text{m}$  thick, bigger towards apex leaves; vascular system Type I, sclerenchyma ring not observed, 482.64-515.16  $\mu\text{m}$  diameter, broader towards leaves apex; vascular bundles (8 VB) (Figure 9A & B); 21.28-35.23  $\mu\text{m}$  diameter; midrib epidermis single layer, 11.35-15.45  $\mu\text{m}$

**Table 1.** Morphological comparison between *Nepenthes domei*, *N. latiffiana*, *N. benstonei* and *N. sanguinea* (Figure 1-4).

Character	<i>N. domei</i>	<i>N. latiffiana</i>	<i>N. benstonei</i>	<i>N. sanguinea</i>
Altitudinal range	850-1000 m	1000-1100 m	800-1350 m	300-2000 m
Habitat	Terrestrial in rich humus soil, rosette/lower pitchers tend to burrow in the soil up to ½ of the pitcher body.	Terrestrial climber in upper montane forest.	Terrestrial in secondary vegetation and hill slopes.	Terrestrial in shrubland, rocky areas (eg. inland cliffs, mountain peaks), forest/terrestrial.
Tendrils of the rosette and climbing leaves	Slight coiled, up to 13-37 cm long.	With slender coiled formation in the middle; up to 10-14 cm long, covered with minute hairs,	Slightly coiled, less than 60 cm long.	Tendrils slight coiled, less than 30 cm.
Colour of lower pitchers	Whitish red to dark ruby red with a few red-purple blotches on the inner surfaces and lid.	Green-yellowish-brown hues with several red blotches on the 1/3 of the upper part of the pitcher body.	Reddish-brown.	Brown to reddish.
Lid of lower pitchers	Narrow ovate; lid base rounded.	Rounded; lid base retuse-cordate.	Ovate; lid base sub-ovate.	Generally ovate; base usually slight cordate.
Upper pitchers	Up to 10-12 cm high $\times$ 1.5-4 cm wide.	Up to 20-23.5 cm high $\times$ 5-7.5 cm wide.	Less than 15 cm high $\times$ and 3 cm wide.	Less than 35 cm high $\times$ less than 7 cm wide.
Colour of upper pitchers	Green.	Light green.	Pale green.	Slight yellowish – green, with several blotches on 1/3 upper part of the pitcher.
Lid of upper pitchers	Narrow ovate to elliptical.	Rounded.	Ovate.	Generally ovate, usually slight cordate at the base.
Colour of dried specimen	Dark brown.	Pale brown.	Brown.	Brown.

at the abaxial and 13.53-20.53  $\mu\text{m}$  at the adaxial, irregular between each other, stomata not observed; trichome or appendages not observed (Figure 9A-F); crystal sand and solitary sand crystals occurring in parenchyma cells of the midrib (Figure 9C & D). **Margin** bluntly pointed below, 75-80 degree pointed below, with epidermis thickness 29.57  $\mu\text{m}$ , margin thickness 139.21  $\mu\text{m}$ , overall blade thickness 291.02-284.75  $\mu\text{m}$  (Figure 9H). **Adaxial epidermis** straight – straight to slightly curve, crystal sand not observed, 2.8-4.5  $\mu\text{m}$  thick; epidermis 1 layer, 2-4 sides, 14.6-29.4  $\mu\text{m} \times 7.7$ -27.9  $\mu\text{m}$ ; stomata absent; sessile glands rounded up to 7 cells, rounded, trichome absent (Figure 9M). **Abaxial epidermis** straight – straight to slightly curve, 2.2-3.8  $\mu\text{m}$  thick; epidermis 1 layer, irregular, more than 4-5 sides, 17.6-18.3  $\mu\text{m} \times 5.6$ -19.5  $\mu\text{m}$ ; stomata anomocytic/ranunculaceous (Figure 9L, M & N), sessile glands rounded to flower-like in 7-8 cells, orbicular to elliptical, 42.3-59.6  $\mu\text{m} \times 40.2$ -51.8  $\mu\text{m}$ ; trichome absent (Figure 9L). **Marginal venation** incomplete type, uni-veinlet (Figure 9I). **Areolar/lamina venation** incomplete (Figure 9J), type of veinlets simple, uni-veinlet (linear-curved) (Figure 9K).

*Nepenthes latiffiana*

Holotype: MDI12424

**Leaves anatomy** lamina thickness 788.68 -794.50  $\mu\text{m}$  (Figure 10E & H), **Hypodermis** 1-2 layers on adaxial, thickness 152.89-162.33  $\mu\text{m}$ , 1 layer on abaxial. Mesophyll 296.56-390.76  $\mu\text{m}$ ; palisade parenchyma 2-3 layers 342.44-387.64  $\mu\text{m}$  thick; spongy parenchyma 564.59-576.54  $\mu\text{m}$ , with plenty extracellular space in between of spongy parenchyma (Figure 10F & H). **Midrib** outline Type IV, adaxial with slightly flat and abaxial prominent broad 'U' curve (Figure 10A), 669.63-689.45  $\mu\text{m}$  thick, smaller towards apex leaves; vascular system Type VI, schlerenchyma ring orbicular upwards, slightly curved at adaxial, outer ring present, central or medullary vascular bundle present (3 VB), additional vascular bundle present (several smaller size vascular bundles observed near abaxial epidermis, 380.23-395.56  $\mu\text{m}$  diameter (Figure 10A-D); up to 50.67  $\mu\text{m}$  diameter; irregular between each other; stomata not observed; trichome appendages not observed (Figure 10A); crystal sand - miniature solitary crystals occurring in parenchyma cells of the midrib (Figure 10B & D). **Margin** sharp pointed below up to 85 degree, with epidermis thickness 13.35-21.58  $\mu\text{m}$ , margin thickness 136.78-146.75  $\mu\text{m}$ , overall blade thickness 335.65-380.76  $\mu\text{m}$  (Figure 10F). **Adaxial epidermis** straight – straight to curve, druses observed on the adaxial surface, 3.7-6.5  $\mu\text{m}$  thick; epidermis 1 layer, 4-5

sides, 15.1-38.7  $\mu\text{m} \times 5.9$ -22.3  $\mu\text{m}$ ; stomata absent; sessile glands irregular in rounded up to 6 cells, elliptical-orbicular, trichome absent (Figure 10L). **Abaxial epidermis** straight – straight to curve, 2.4-4.1  $\mu\text{m}$  thick; epidermis 1 layer, irregular, more than 4 sides, 14.32-28.54  $\mu\text{m} \times 43.5$ -32.6  $\mu\text{m}$ ; stomata anomocytic/ranunculaceous (Figure 10M & N), rounded, 108.25-122.645  $\mu\text{m} \times 118.1$ -119.4  $\mu\text{m}$ , sessile glands not observed, trichome absent. **Marginal venation** closed type, none veinlet (Figure 10K). **Areolar/lamina venation** incomplete (Figure 10I), type of veinlets simple uni-bi veinlet (curved) (Figure 10J).

*Nepenthes benstonei*

**Leaves anatomy** lamina thickness 252.96-266.43  $\mu\text{m}$  (Figure 11G), **Hypodermis** 1-2 layers on adaxial, thickness 25.67-35.94  $\mu\text{m}$ , 1 layer on abaxial, thickness 16.26-27.38  $\mu\text{m}$ . Mesophyll 159.17-172.86  $\mu\text{m}$ ; palisade parenchyma 1-3 layers with total 67.65-84.15  $\mu\text{m}$  thick; spongy parenchyma total 106.97-111.25  $\mu\text{m}$ , with plenty extracellular space in between of spongy parenchyma (Figure 11G & H). **Midrib** outline Type I, adaxial very slightly concave, abaxial prominent 'U-V' Shape (Figure 11A), 380.81-391.71  $\mu\text{m}$  thick, vascular system Type VI, outer ring present (rectangular in shape), central vascular bundle present (3 vascular bundles), additional vascular bundle present (several smaller size vascular bundle near abaxial epidermis) and 2 vascular bundle present on the adaxial side of outer ring vascular bundle at the left and right side, midrib epidermis single layered, 13.26-17.83  $\mu\text{m}$  at the abaxial and 15.59-16.42  $\mu\text{m}$  at the adaxial, irregular between each other; stomata not observed; trichome appendages not observed (Figure 11A & I); crystal sand - solitary sand crystals occurring in masses specifically in parenchyma cells of the midrib (Figure 11B-D), druses observed (Figure 11E), starch grains observed. (Figure 11B). **Margin** sharply pointed below 20-25 degree, with epidermis thickness 8.81-13.25  $\mu\text{m}$ , margin thickness 68.51-82.83  $\mu\text{m}$  (Figure 11H). **Adaxial epidermis** straight to wavy, crystal sand observed on the adaxial surface, 3.5-4.9  $\mu\text{m}$  thick; epidermis 1 layer, 4-6 sides, 15.6-34.5  $\mu\text{m} \times 8.4$ -9.3  $\mu\text{m}$ ; stomata absent; sessile glands irregular in flower-like in 6-7 cells, elliptical, trichome absent (Figure 11J). **Abaxial epidermis** sinuous, 2.5-3.8  $\mu\text{m}$  thick; epidermis 1 layer, irregular, more than 4 sides, 18.42.87  $\mu\text{m} \times 6.5$ -36.9  $\mu\text{m}$ ; stomata anomocytic/ranunculaceous (Figure 11K), kidney-like, trichome absent. **Marginal venation** closed type, none veinlet (Figure 11M). **Areolar/lamina venation** incomplete (Figure 11L), type of veinlets simple veinlet (linear-curved) 1-2 branched (Figure 11L).

*Nepenthes sanguinea*

**Leaves anatomy** lamina thickness 224.20-235.23  $\mu\text{m}$  (Figure 12E), **Hypodermis** 1 layer on adaxial, thickness 20.54-37.65  $\mu\text{m}$ , 1 layer on abaxial, thickness 29.15-32.52  $\mu\text{m}$ . Mesophyll 145.87-166.45  $\mu\text{m}$ ; palisade parenchyma 2 layer with total 53.06-63.32  $\mu\text{m}$  thick; spongy parenchyma total 90.71-94.99  $\mu\text{m}$ , with plenty extracellular space in between of spongy parenchyma (Figure 12E & F). **Mid-rib** outline Type I, adaxial slightly flattened and abaxial arched, (Figure 7a), 383.37-445.89  $\mu\text{m}$  thick, vascular system Type VI, sclerenchyma ring orbicular upwards, slightly curved at adaxial, outer ring present, central or medullary vascular bundle present (4 VB), 1 additional vascular bundle present (smaller size vascular bundles observed near abaxial epidermis), 367.11-413.32  $\mu\text{m}$  diameter (Figure 7A-C); diameter up to 40.35  $\mu\text{m}$ ; irregular between each other; stomata not observed; trichome

appendages not observed (Figure 12A); crystal sand - solitary crystals and druse occurring in parenchyma cells of the midrib (Figure 12B-D). **Margin** sharp tip pointed below up to up to 80 degree, roughly crease surface, with epidermis thickness 6.85-9.41  $\mu\text{m}$ , margin thickness 86.76-92.23  $\mu\text{m}$ , overall blade thickness 89.05-106.97  $\mu\text{m}$  (Figure 12F). **Adaxial epidermis** straight – slight curve, druses not observed on the adaxial surface, 3.8-7.8  $\mu\text{m}$  thick; epidermis 1 layer, 3-5 sides; stomata absent; sessile glands irregular in rounded up to 5 cells, elliptical, trichome absent (Figure 12I). **Abaxial epidermis** straight – slight curve, 2.4-4.9  $\mu\text{m}$  thick; epidermis 1 layer, irregular, more than 4-6 sides; stomata anomocytic (Figure 12J), 95.47-117.74  $\mu\text{m} \times$  105.54-147.65  $\mu\text{m}$ , sessile glands not observed, trichome absent. **Marginal venation** incomplete type, uni-veinlet (Figure 12H). **Areolar/lamina venation** incomplete (Figure 12I), type of veinlets – simple uni veinlet (slight curved) (Figure 12I).

**Table 2.** Comparison of leaf micromorphological observation between *Nepenthes domei*, *N. latiffiana*, *N. benstonei* and *N. sanguinea* (Figure 5-8).

	<i>N. domei</i>	<i>N. latiffiana</i>	<i>N. benstonei</i>	<i>N. sanguinea</i>
Cuticular striation	<b>Adaxial:</b> smooth-coarse; anticlinal wall not clear; periclinal wall sunken, cuticular striation not obscure and both anticlinal and periclinal walls cannot be differentiated clearly. Epidermal cell outline obscured by appendages. <b>Abaxial:</b> smooth surface, with granular waxes; both anticlinal and periclinal walls cannot be differentiated clearly.	<b>Adaxial:</b> coarse; anticlinal wall clear; periclinal wall sunken, cuticular striation obscure and both anticlinal and periclinal walls cannot be differentiated clearly. Epidermal cell outline obscured by high density of waxes. <b>Abaxial:</b> coarse; with granules waxes; both anticlinal and periclinal walls can be differentiated clearly.	<b>Adaxial:</b> coarse; anticlinal wall not clear; periclinal wall sunken, cuticular striation obscure and both anticlinal and periclinal walls can be differentiated. Epidermal cell outline obscured by high density of waxes. <b>Abaxial:</b> coarse; with flaked waxes; both anticlinal and periclinal walls cannot be differentiated clearly due to high density of flakes and appendages.	<b>Adaxial:</b> coarse; anticlinal wall not clear; periclinal wall sunken, both anticlinal and periclinal walls can be differentiated clearly. Epidermal cell outline not obscured by high density of waxes. <b>Abaxial:</b> medium coarse; both anticlinal and periclinal walls can be differentiated.
Epicuticular waxes	i. Granules present on both adaxial and abaxial surfaces, tubular wax on abaxial.	i. Flakes (adaxial) ii. Granules (abaxial)	i. Flakes (adaxial) ii) Granules (abaxial)	Flakes present on both adaxial and abaxial epidermises.
Stomata features	Amphistomatous, sunken, scattered mainly on the abaxial surface, sparsely distributed on the adaxial surface, rounded shape.	Hypostomatic, superficial, restricted to and sparsely scattered on the abaxial epidermis surface, broad oval shape.	Hypostomatic, superficial, scattered to and sparsely scattered on the abaxial epidermis surface, broad oval shape.	Amphistomatous, sunken, scattered mainly on the abaxial surface, sparsely distributed on the adaxial surface, rounded-oval shape.
Types of trichomes	<b>i.</b> Simple unicellular (long, pointed tip, with slight echinate ornamentation). <b>ii.</b> Capitate glandular trichome (multicellular terminal). <b>iii.</b> Capitate glandular trichome (unicellular terminal). <b>iv.</b> Peltate, Scale trichome.	<b>i.</b> Simple unicellular (long, pointed tip, slight echinate ornamentation). <b>ii.</b> Simple unicellular short, blunt tip, without echinate ornamentation <b>iii.</b> Capitate glandular trichome (multicellular terminal).	<b>i.</b> Simple unicellular (long, pointed tip, smooth). <b>ii.</b> Capitate glandular trichome (unicellular terminal). <b>iii.</b> Capitate glandular trichome (multicellular terminal).	<b>i.</b> Capitate glandular trichome (multicellular terminal).

**Table 3.** Comparison of leaf anatomical features between *Nepenthes domei*, *N. latiffiana*, *N. benstonei* and *N. sanguinea* (Figure 9-12).

	<i>N. domei</i>	<i>N. latiffiana</i>	<i>N. benstonei</i>	<i>N. sanguinea</i>
Leaf margin	Bluntly pointed below 75-80°.	Sharply pointed below up to 85°.	Sharply pointed below 20-25°.	Sharply pointed below up to 80°, with roughly crease surface.
Midrib	Outline Type I.	Outline Type IV.	Outline Type I.	Outline Type I.
Vascular bundle arrangement	Arrangement Type I.	Arrangement Type VI.	Arrangement Type VI.	Arrangement Type VI.
Marginal venation	Incomplete type, uni-veinlet.	Closed type, none veinlet.	Closed type, none veinlet.	Incomplete type, uni-veinlet.
Areolar venation	Incomplete, type of veinlets simple, uni-veinlet (linear-curved).	Incomplete, type of veinlets simple uni-bi veinlet (curved).	Incomplete; type of veinlets simple veinlet (linear-curved) 1-3 branched.	Incomplete, type of veinlets-simple uni-veinlet (slightly curved).
Pattern of anticlinal cells	<b>Adaxial:</b> straight – straight to slightly curved. <b>Abaxial:</b> straight – straight to slightly curved.	<b>Adaxial:</b> straight – straight to curved. <b>Abaxial:</b> straight – straight to curved.	<b>Adaxial:</b> straight to wavy, crystal sand observed on the adaxial surface. <b>Abaxial:</b> sinuous.	<b>Adaxial:</b> straight –slightly curved. <b>Abaxial:</b> straight – slightly curved.
Stomata feature	Anomocytic (type of stomata-without subsidiary cells); Homostomatic (only one type of stomata present); Amphistomatous (stomata present on both epidermis surfaces).	Anomocytic; Homostomatic; Hypostomatic (stomata only present on abaxial epidermis).	Anomocytic; Homostomatic; Hypostomatic.	Anomocytic; Homostomatic; Amphistomatous.

Results of the study revealed a number of interesting features with some characters which could serve as taxonomic and diagnostic value, while assist in giving additional evidences in describing *N. domei* and *N. latiffiana* against related species. Anatomical diagnostic characters that can be used in directly differentiating and possessed taxonomic value are sclerenchyma ring type and shape, and marginal venation and lamina venation that enumerate significant taxon separation between *N. domei* and *N. benstonei*, and *N. latiffiana* and *N. sanguinea*. Supportive micromorphology and anatomy characters which are also useful in assisting these taxa identification (diagnostic characters) in new taxon descriptions are noted as below, with identification key of the species:

## 2) Diagnostic characters of *Nepenthes domei* and *N. latiffiana* based on leaf anatomy and micromorphology

Anticlinal walls of lamina epidermal cells under light microscope (LM)

According to Stace (1969), the lamina surface has been the focal subject of investigations (as compared with other plant surfaces) that some of studied taxa showed consistent characteristics which often serve as a

taxonomic application. From observations of the lamina surface in studied *Nepenthes*, the anticlinal wall of *N. domei* is straight to slight curve for both abaxial and adaxial epidermis surfaces while *N. benstonei* showed straight to wavy adaxial epidermis surface and sinuous abaxial epidermis surface. On the other hand, *N. latiffiana* showed straight to straight curve epidermis wall for both of its surfaces as compared to *N. sanguinea* which possessed straight – slightly curve and straight for its abaxial epidermis. Therefore, these characters appear to be diagnostic for those two new species, respectively.

## Margin and Midrib Transverse Section (TS)

Studied *Nepenthes* species showed interspecies variations in lamina margin transverse section (TS) which can serve as a useful characteristic for species identification. The outline of the TS of the margin is bluntly rounded (pointed 75-80°) for *N. domei*, sharply pointed downwards that reached 85° for *N. latiffiana*, sharply pointed with 20-25° curve for *N. benstonei*, while *N. sanguinea* having up to 80° and rough creased abaxial and adaxial surfaces. Midrib outline for *N. domei*, *N. sanguinea* and *N. benstonei* is characterised as **Type I** (abaxial with prominent U-V shape and adaxial with very

slight concave shape) while *N. latiffiana* having **Type IV** (abaxial with prominent V shape and adaxial with prominent wide V shape) midrib outline. For midrib TS, the variations are as follows: *N. domei* is having **Type I** (adaxial vascular bundle in stacks, medullary vascular bundle absent, additional vascular bundle present in several smaller size nearby the abaxial epidermis) vascular bundle arrangement, while *N. latiffiana*, *N. benstonei* and *N. sanguinea* are having **Type VI** vascular bundle arrangement (outer ring present, elliptic; central medullary vascular bundle present in scattered formation; additional vascular bundle present in several smaller size nearby the adaxial side of outer ring at the left and right side). These observations (disparity in vascular bundle arrangement combined with midrib outline) agree with some studies conducted by Metcalfe and Chalk (1950) who demonstrated the two characteristics could greatly serve for diagnosis in certain species, as recorded and showed in some *Parashorea* species.

#### Marginal and areolar venation

The variation of leaf venation patterns of angiosperms was extensively studied and classified by Hickey (1979) and Sun et al. (1997) and this further addresses the importance of comprehensive treatment in taxon differentiation. Leaf architecture is primarily used for classification which includes the leaf shape, leaf margin structure and other possible characters. Indeed, the characterisation of an angiosperm leaf venation pattern starts by observing the primary veins and then proceeded to the branching secondary veins. Combination of marginal venation and areolar venation patterns for every taxon are unique, even for those taxa classified under the same genus (Inamdar et al. 1983). In studied *Nepenthes* species, marginal and lamina/areolar venation features showed diagnostic characteristics that are useful in species differentiation and evaluation. *Nepenthes domei* and *N. sanguinea* showed incomplete uni-veinlet, while *N. benstonei* and *N. latiffiana* are characterised with closed type, with none veinlet. Another feature analysed is the areolar / lamina venation, in which all taxa showed incomplete areolar venation but varied in type of venation ending as follows: *N. domei* showed simple veinlet with uni-veinlet (linear to curved), while *N. latiffiana* and *N. benstonei* shared the same feature of uni-bi veinlet (curved) that slightly differs in *N. benstonei* venation ending with linear-curved (1-3 branched) and *N. sanguinea* characterised with uni-veinlet (slight curved). These variable patterns of leaf venation are taxonomically significant in these pitcher plants species as

they can be used as additional evidences for species differentiation.

Cuticular wax and epidermal sculpturing under scanning electron microscopy (SEM)

Cuticular sculpturing also holds considerable diagnostic values as reported by Wilkinson (1979) and Wu et al. (2005). In this study, the epidermal surfaces revealed a number of important micromorphological characters, exhibited interesting interspecies variations that are significant for species identification. Leaf epidermis surfaces (abaxial and adaxial) of the investigated *Nepenthes* species that were viewed under the SEM (low to high magnifications) gave certain features – distribution of idioblastic elements such as trichomes, glands and stomata, combined with the appearance of epicuticular wax. In this study, the appearance of epicuticular wax on both of the leaf surfaces could be tubular granular or flaked, or a combination of different types of wax. *Nepenthes domei* differed from others for having granular wax on both abaxial and adaxial epidermis surfaces, and tubular wax only on abaxial surface (Figure 5). Meanwhile its closely related species, *N. benstonei*, showed numerous wax flakes (Figure 7) for its adaxial epidermis surface and granules for its abaxial surface. It seems the presence of these numerous flakes supports Clarke's (2001) observation saying that the leaves and stems of *N. benstonei* were covered with thick waxy cuticle. *Nepenthes latiffiana* showed similar features as that of *N. benstonei*, while *N. sanguinea* had wax flakes on both abaxial and adaxial surfaces. Stomata features also implicate a significant diagnostic value for our species of interest. *Nepenthes domei* and *N. sanguinea* leaves are amphistomatous (stomata on both surfaces); however *N. benstonei* and *N. latiffiana* are hypostomatous (stomata only on the lower surface). While hypostomatous stomata feature is common for the majority of species for monocotyledons and dicotyledons (Meidner and Mansfield 1968), amphistomatous feature is much rare (Drake et al., 2018). Drake et al. (2018) further points out that amphistomaty may help in better CO<sub>2</sub> absorption but with higher rate of transpiration and this discovery of two different groups of leaves (hypostomatous vs. amphistomatous) within the same genus is truly intriguing. The indumentum feature in dicotyledons was reported to consist of simple unicellular and various glandular trichomes (Metcalfe and Chalk, 1950). In this study, glandular trichomes was present in all species but *N. domei* showed diagnostic character with the observation of scale-type trichomes, known as peltate,

on its abaxial epidermis surface that did not exist in other investigated species. Simple unicellular trichomes are common and present in all species of studied *Nepenthes* except in *N. sanguinea* that also can be postulated as a diagnostic characteristic that differentiates *N. latiffiana* from *N. sanguinea*. This trichome type can be long or short, and has either thick wall ornamentation or smooth.

#### Identification key based on anatomy and micromorphology characters

- 1 a. Midrib outline Type I; vascular bundle arrangement Type I, with peltate trichome.....*N. domei*  
 b. Midrib outline Type IV; vascular bundle arrangement type VI..... 2
- 2 a. Margin transverse section sharply pointed over 20-25° 3  
 b. Margin transverse section sharply pointed below up to 85° .....*N. latiffiana*
- 3 a. Adaxial epidermal cell outline obscured by high density of waxes ..... *N. benstonei*  
 b. Adaxial epidermal cell outline not obscured by high density of waxes..... 4
- 4 Simple unicellular trichome absent, only peltate glandular trichome (multicellular terminal) observed ...*N. sanguinea*

#### 3) Molecular analysis of ITS sequence to infer the relationship between *N. benstonei* and other species

The ITS is a nuclear DNA region which is highly conserved and it has been used successfully to discriminate different plant species including *Nepenthes* (Giudicelli et al. 2015; Gogoi and Bhau 2018). Based on nucleotide search on the public NCBI database (BLASTn), we noted partial ITS region (~610bp) is more than 95% conserved among the majority species in the *Nepenthes* genus and the ITS may not be sensitive to differentiate sub-populations under the same species. However, we tested the degree of ITS sequence conservation from a few different populations of *N. sanguinea* in Peninsular Malaysia and it is proven the partial ITS sequence is 100% conserved within this particular species (data not shown), saying that ITS intraspecific variation of a closed population should be close to 0.

In order to further establish *N. domei* as a separate taxon from its most closely related species *N. benstonei*, we conducted a molecular analysis by comparing the partial ITS sequence from both plants (Figure 13)

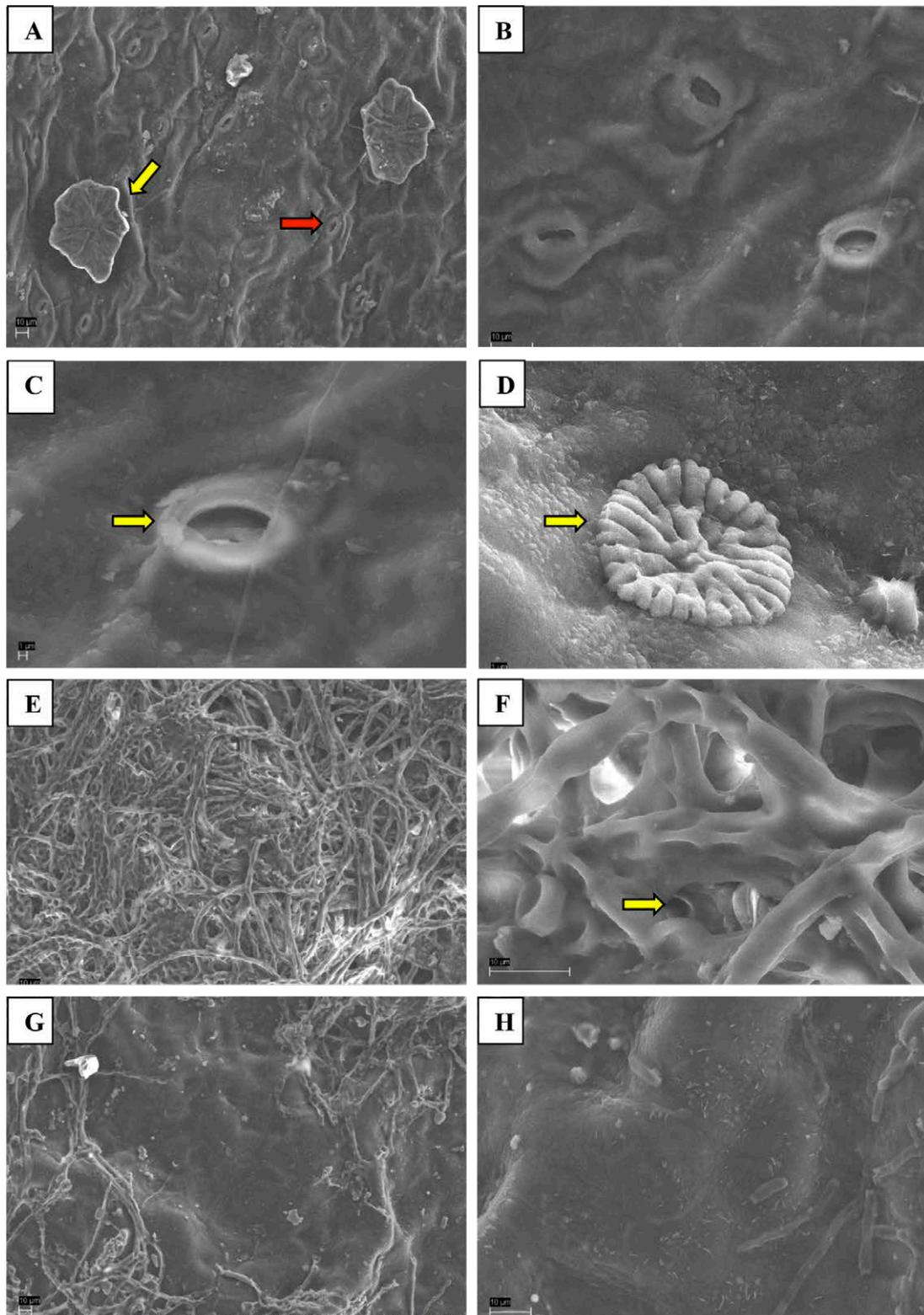
and then inferred phylogenetic relationship (NJ method) of the new species with nine peninsular *Nepenthes* species (Figure 14). Based on sequence alignment, *N. domei* sp. nov shares up to 97.2% ITS nucleotide identity to *N. benstonei*, and the 2.8% nucleotide dissimilarity – a figure significant enough to differentiate *Nepenthes* species in this study – is due to nucleotide polymorphisms that occur at 15 different positions, denoted as non-coloured letters (Figure 13). In addition to this, the phylogenetic inferring has positioned *N. domei* on a separate branch from *N. benstonei* in the highland/intermediate highland clade (Figure 14). This molecular scrutiny – on top of morphology, leaf anatomy and micromorphology – serves as an addition evidence that has positioned *N. domei* as a distinct species from *N. benstonei*.

#### CONCLUSION

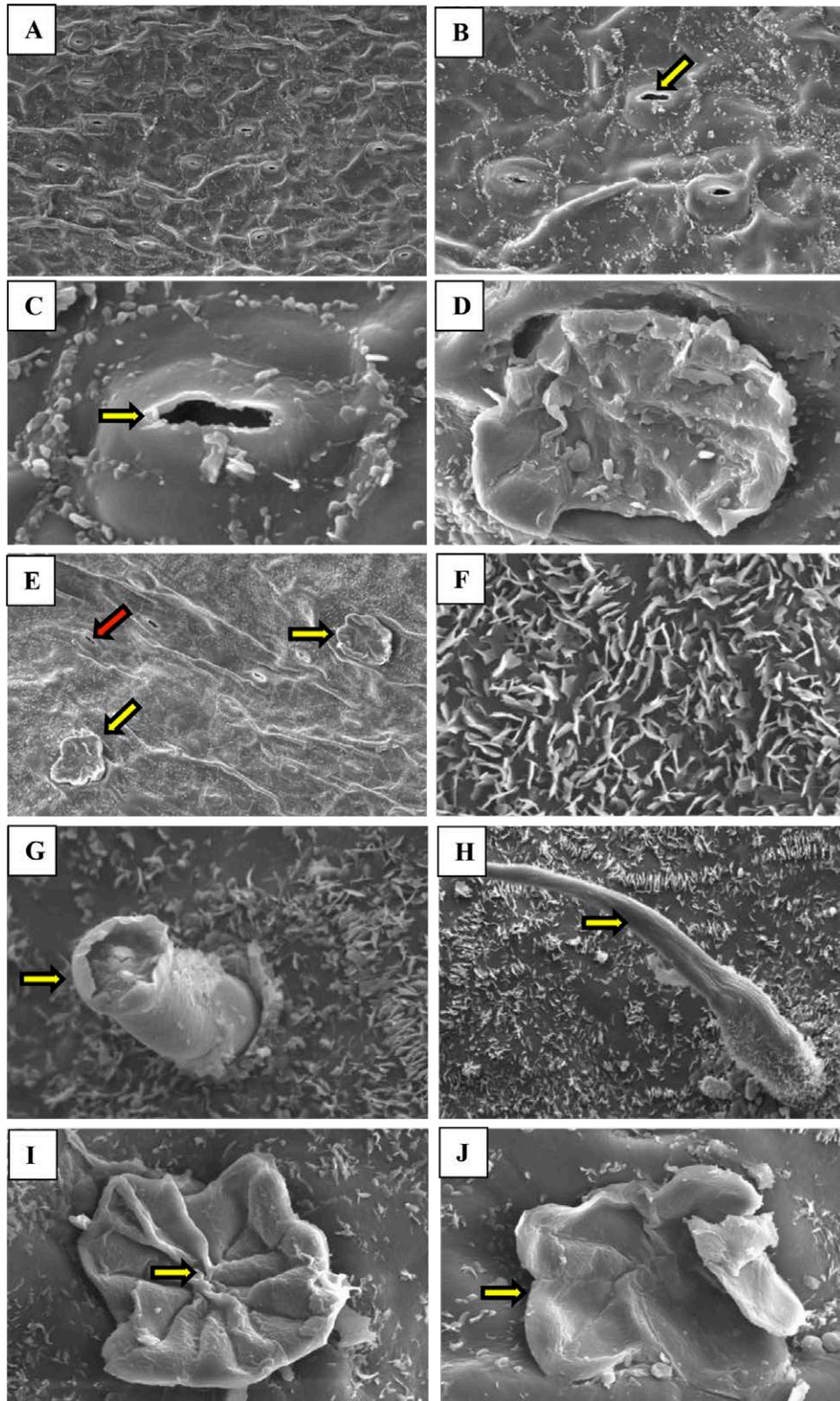
The combination approach of morphology-anatomy-micromorphology analyses used in this comparison study is useful in providing as much aspects as possible in distinguishing plant characteristics of *N. domei* and *N. latiffiana* against their congener species, *N. benstonei* and *N. sanguinea*. As a whole, pitcher morphology of both new species combined with detailed descriptions of the midrib, lamina and epidermis have addressed comprehensive taxonomic resolution in defining these new *Nepenthes* species. Also, the phylogenetic inference has provided an additional supportive evidence for the placement of *N. domei* as a new species. Hence, *N. domei* and *N. latiffiana* described in this paper are confidently positioned as two new species discovered from eastern part of Peninsular Malaysia.

#### ACKNOWLEDGEMENTS

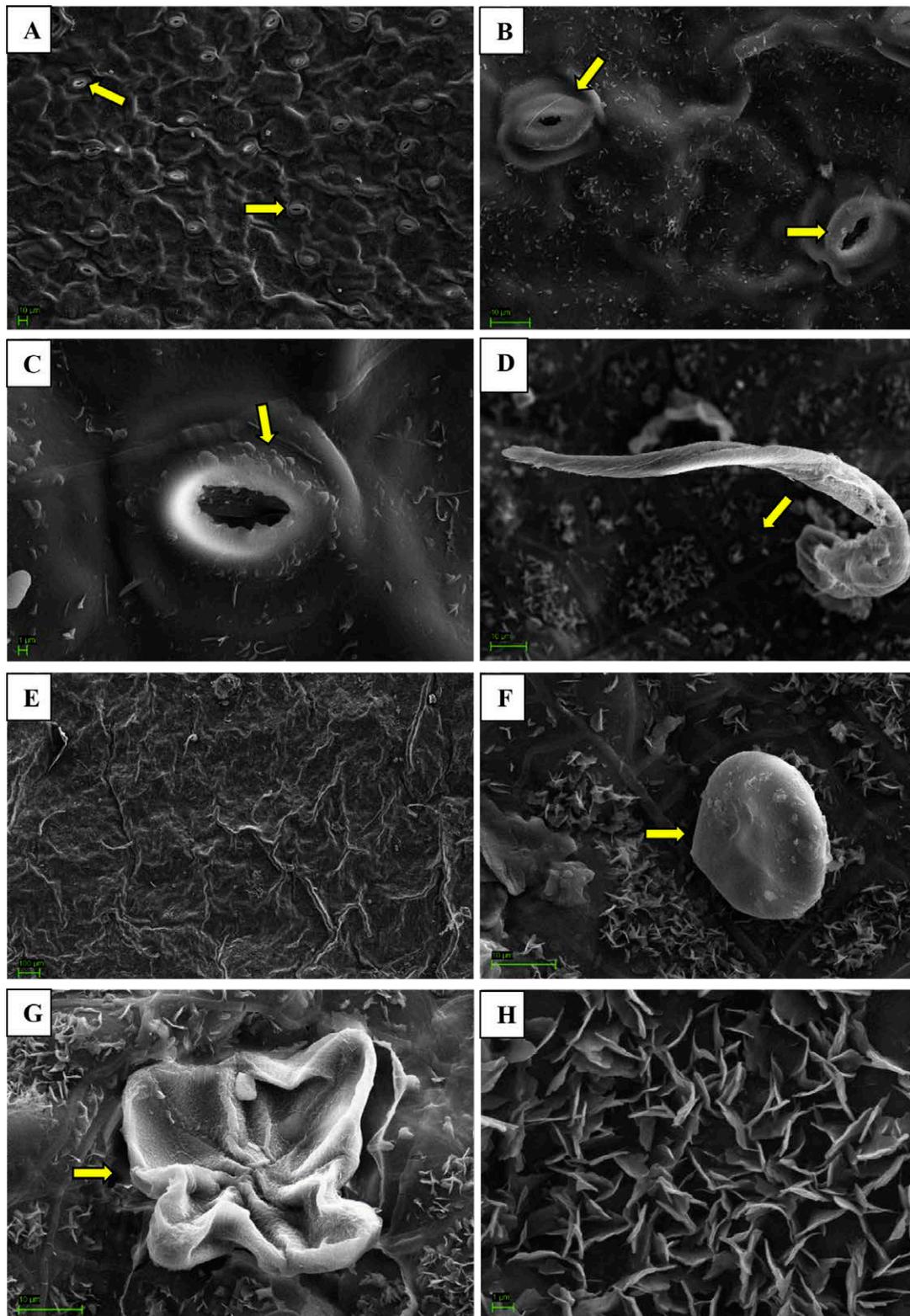
This study was carried out under Forestry Department of Peninsular Malaysia (Ministry of Energy and Natural Resources) permit ref. JH/100 Jld. 22 (41). We are grateful to Dr. Richard Chung Cheng Kong (Forest Research Institute Malaysia, FRIM) for his guidance and help in obtaining permission involving herbarium records and observation of the pitcher plants herbarium specimens (KEP). Special thanks go to Madam Salmaniza Salleh and Miss Nurshahidah Mohd Rusli for their technical assistance in the anatomical and palynological works as well as in herbarium curations. We also extend our gratitude to Muhammad Ikhwan Afandi Md Daud for his assistance during the field work.



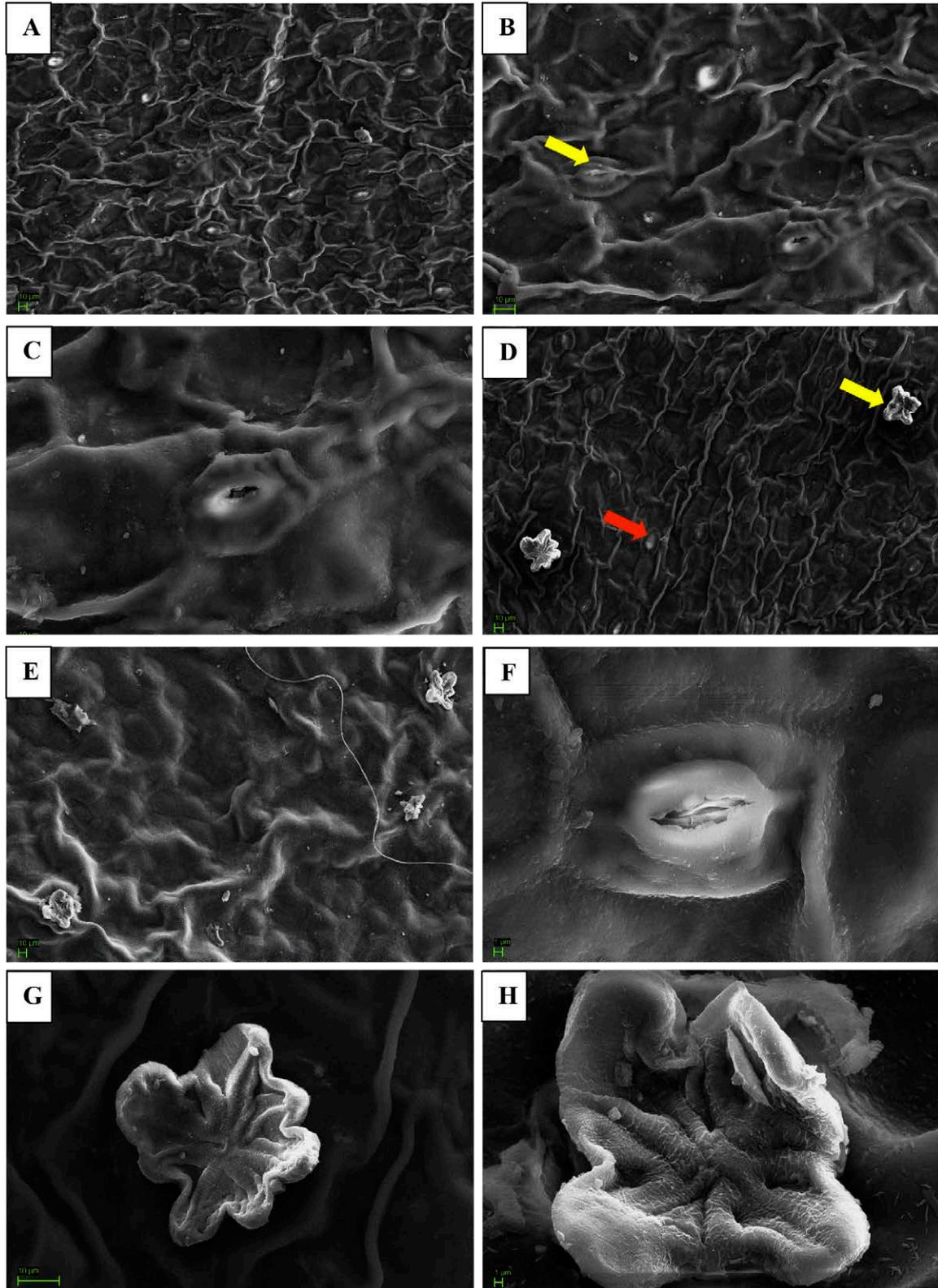
**Figure 5.** *Nepenthes domei*: (A) Abaxial epidermis; stoma (red arrow) and peltate trichome (yellow arrow). (B) Stomata and epicuticular wax. (C) Stoma (arrow). (D) Peltate (scale) trichome. (E) Adaxial epidermis with tubular wax. (F) Stoma deep embedded into tubular wax (arrow) observed on the adaxial epidermis. (G) Granular wax mixed with tubular. (H) Cuticular striation. SEM images by by Mohd Norfaizal Ghazalli and Ahmad Zaki Zaini.



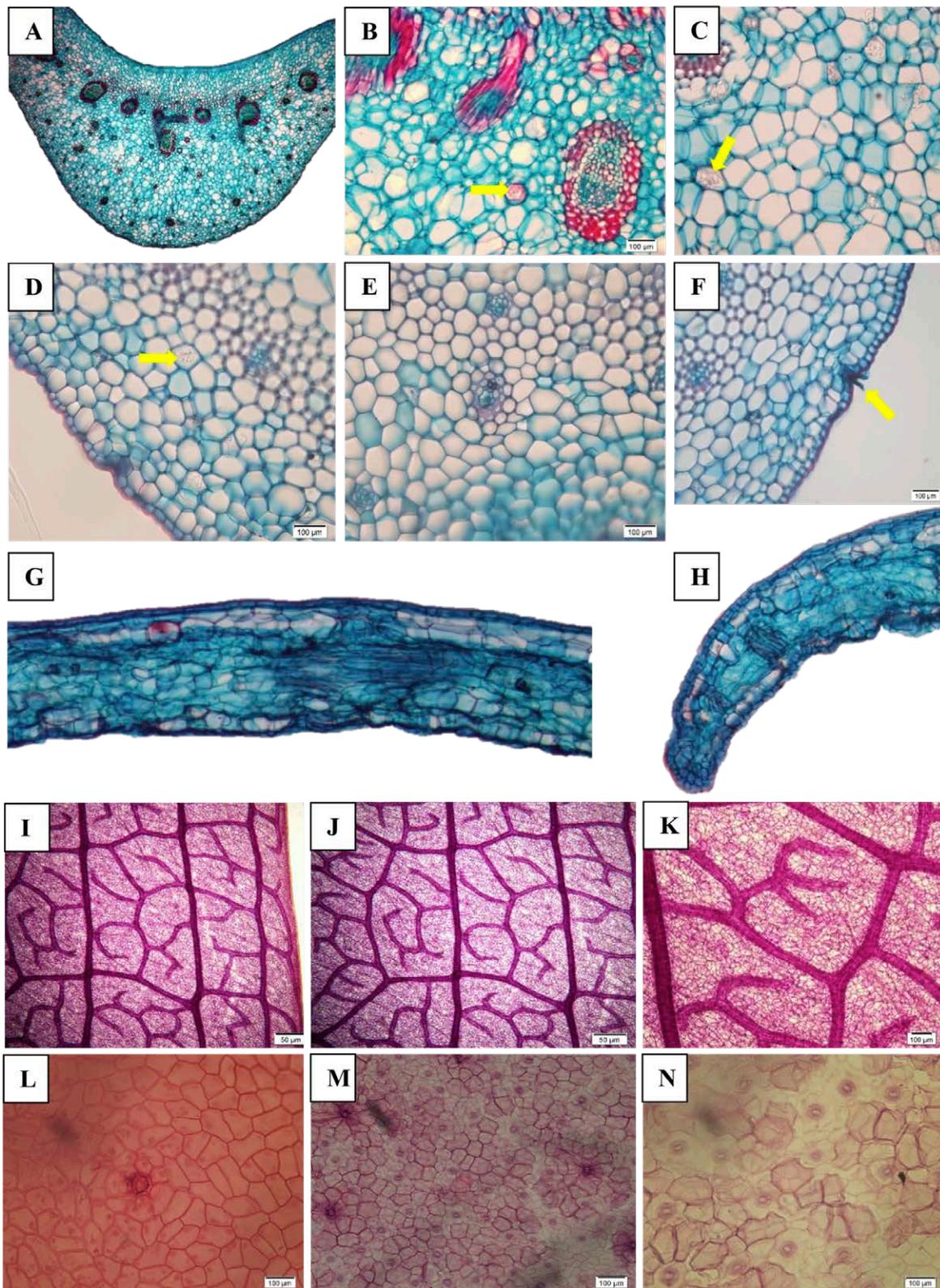
**Figure 6.** *Nepenthes latiffiana*: (A) Abaxial epidermis adorned with numerous stomata. (B) Abaxial stomata (arrow). (C) Stomata (arrow) on abaxial surface with wax granules. (D) Peltate glandular trichome. (E) Peltate glandular trichomes (yellow arrows) and stoma (red arrow). (F) Adaxial epidermis with wax flakes but without stomata. (G) Capitate glandular trichome (arrow). (H) Simple, unicellular trichome, long pointed (arrow). (I & J) Variation of multicellular peltate glandular trichome (arrow). SEM images by Mohd Norfaizal Ghazalli and Ahmad Zaki Zaini.



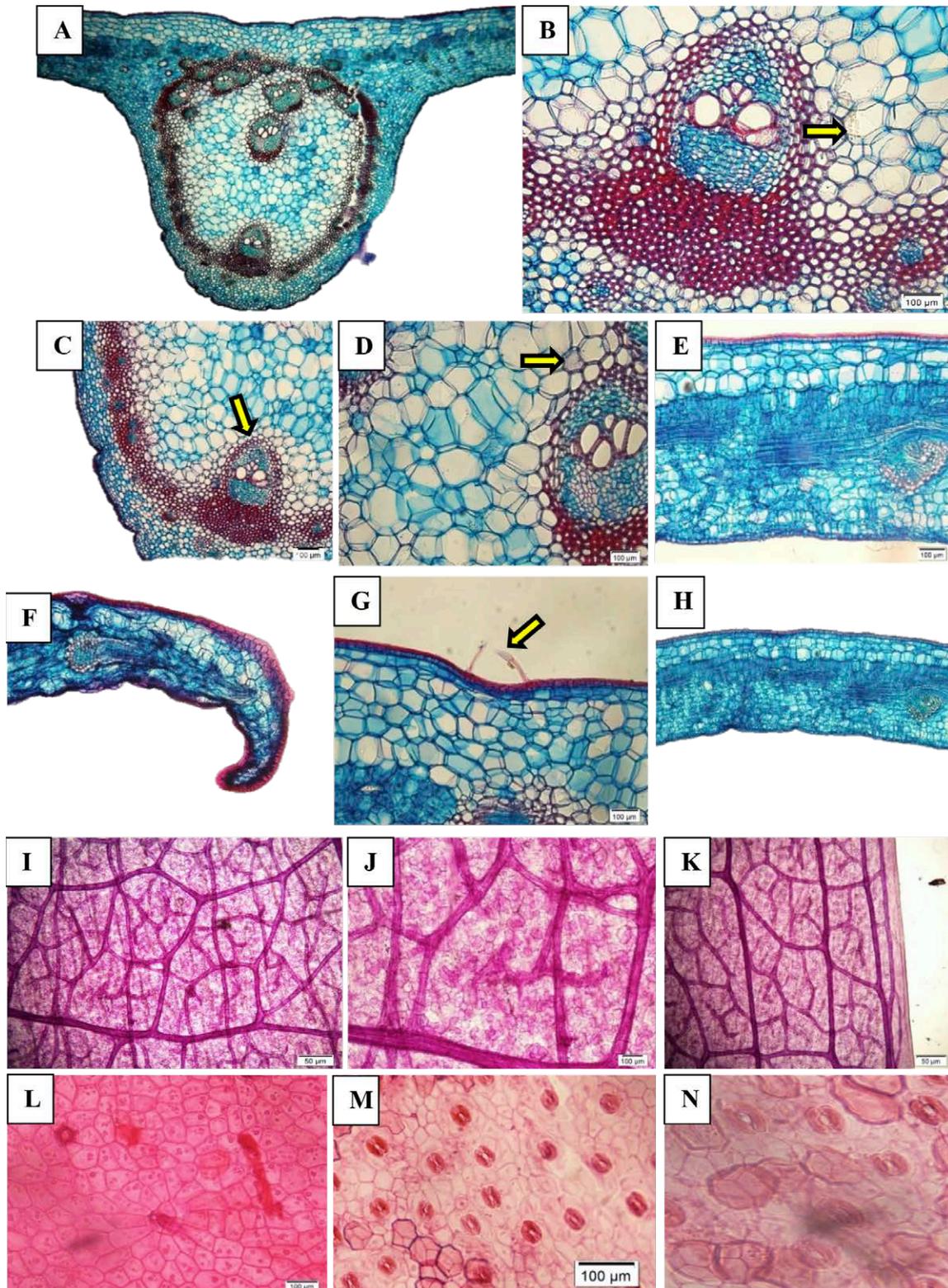
**Figure 7.** *Nepenthes benstonei*: (A & B) Abaxial epidermis with stomata (arrows). (C) Abaxial stoma surrounded by wax granules (arrow). (D) Simple, unicellular trichome, long pointed (arrow). (E) Adaxial surface absent of stomata. (F) Capitate glandular trichome (arrow) surrounded by wax flakes. (G) Multicellular peltate glandular trichome (arrow). (H) Flakes observed on the adaxial epidermis. SEM images by Mohd Norfaizal Ghazalli and Ahmad Zaki Zaini.



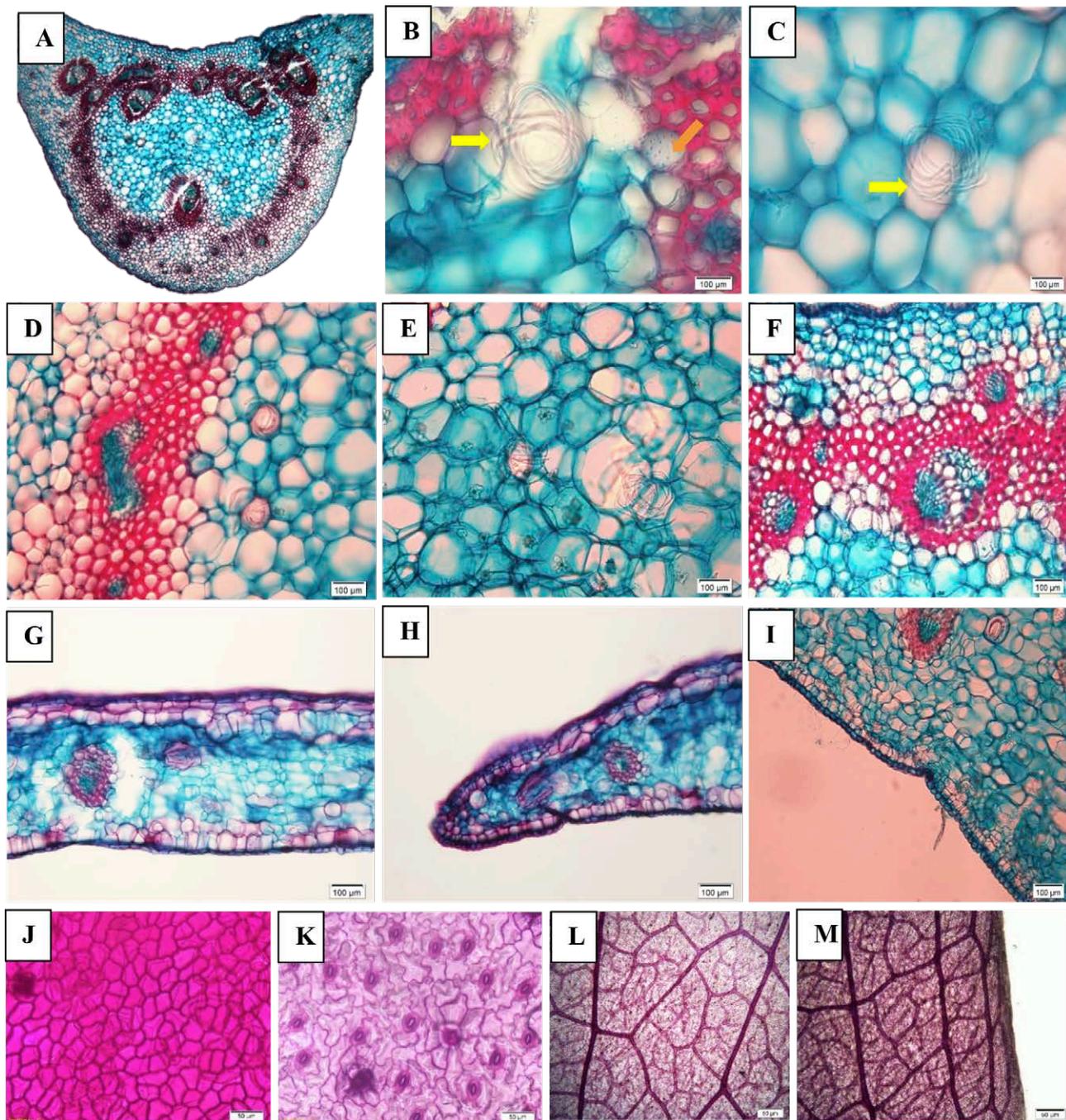
**Figure 8.** *Nepenthes sanguinea*: (A) Abaxial epidermis. (B) Stomata (arrow). (C) Stoma. (D) Adaxial epidermis with stoma (red arrow) and peltate glandular trichome (yellow arrow). (E) Adaxial epidermis surface. (F) Stoma. (G & H) Variation of multicellular peltate glandular trichomes observed on the adaxial epidermis surface. SEM images by by Mohd Norfaizal Ghazalli and Ahmad Zaki Zaini.



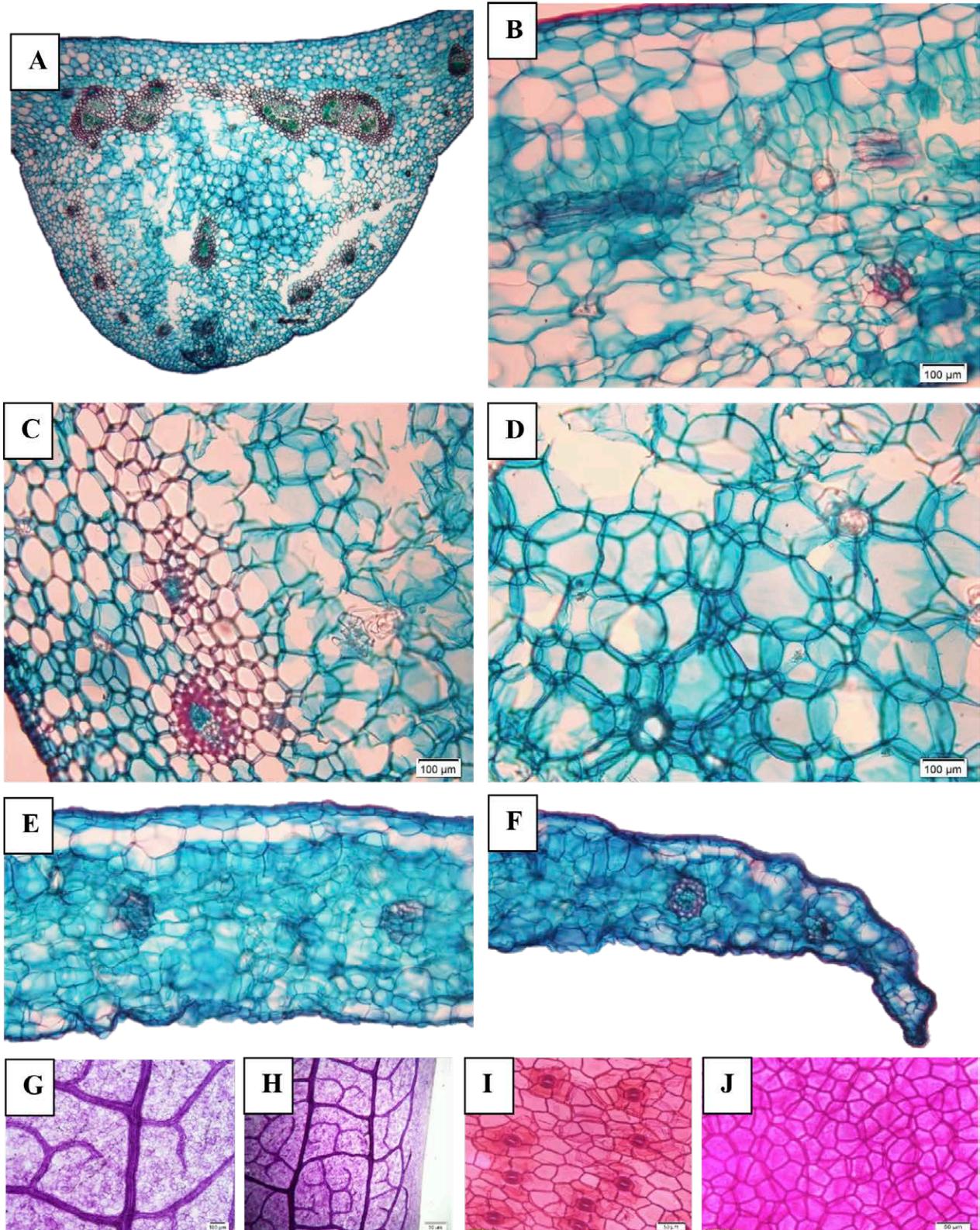
**Figure 9.** *Nepenthes domei*: (A) Midrib TS. (B, C & D) Solitary sand crystal (arrows). (E) Parenchyma cells. (F) Simple, unicellular trichome (arrow). (G) Lamina TS. (H) Margin TS. (I) Margin venation. (J) Lamina venation. (K) Type of veinlet. (L) Adaxial epidermis showing a stoma. (M & N) Abaxial epidermis with stomata. Photographs by Mohd Norfaizal Ghazalli and Amin Asyraf Tamizi.



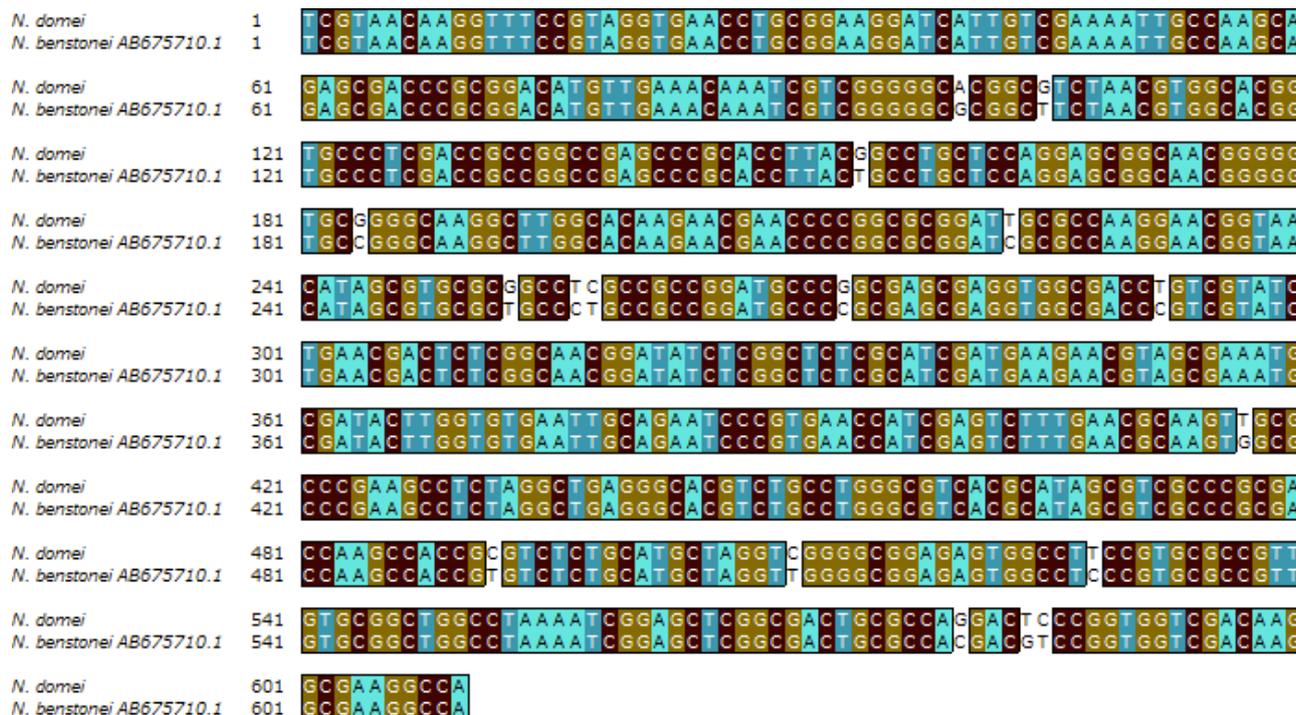
**Figure 10.** *Nepenthes latiffiana*: (A) Midrib TS. (B) Solitary crystal (arrow). (C) Vascular bundle (arrow). (D) Solitary sand crystal. (E) Lamina TS. (F) Margin TS. (G) Simple, unicellular trichomes (arrow). (H) Lamina TS. (I) Lamina venation. (J) Type of veinlet. (K) Margin venation. (L) Adaxial epidermis. (M) Abaxial epidermis with stomata. (N) Stomata. Photographs by Mohd Norfaizal Ghazalli and Amin Asyraf Tamizi.



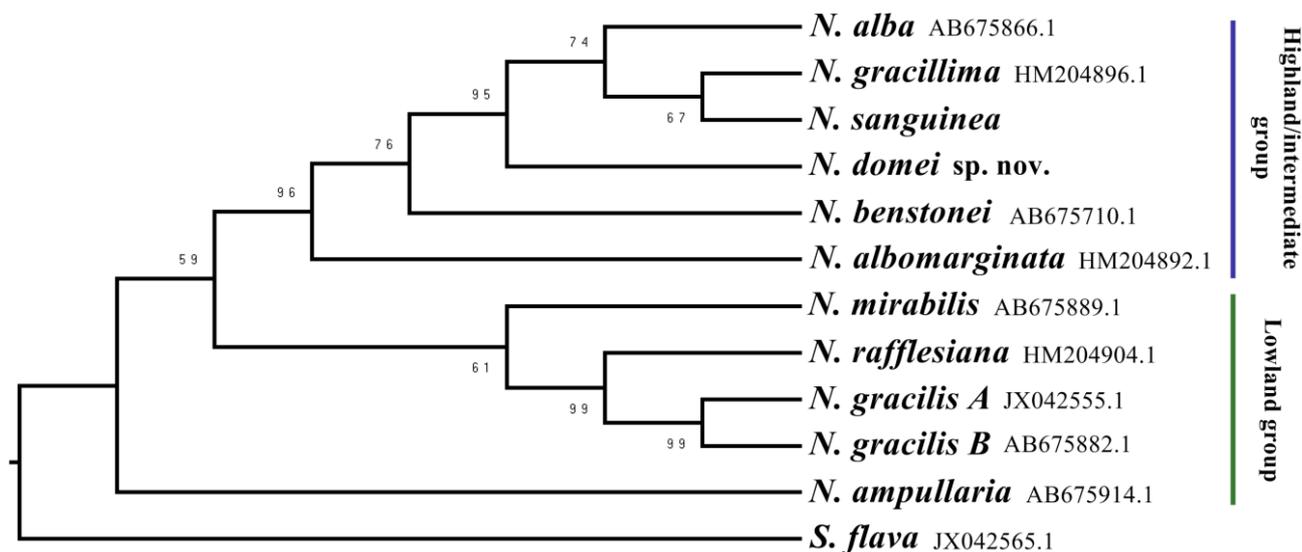
**Figure 11.** *Nepenthes benstonei*: (A) Midrib TS. (B) Observation of solitary crystal (yellow arrow) and starch grain (orange arrow). (C) Solitary crystal (arrow). (D) Solitary crystal. (E) Druses. (F) Vascular bundle. (G) Lamina TS. (H) Margin TS. (I) Simple, unicellular trichome. (J) Adaxial epidermis. (K) Abaxial epidermis with stomata observation. (L) Lamina venation. (M) Margin venation. Photographs by Mohd Norfaizal Ghazalli and Amin Asyraf Tamizi.



**Figure 12.** *Nepenthes sanguinea*: (A) Midrib TS. (B) Solitary crystal (arrow). (C) Solitary crystal (arrow). (D) Solitary sand crystal. (E) Lamina TS. (F) Margin TS. (G) Lamina venation. (H) Marginal venation. (I) Abaxial epidermis. (J) Adaxial epidermis. Photographs by Mohd Norfaizal Ghazalli and Amin Asyraf Tamizi.



**Figure 13.** Pairwise alignment (optimal Global alignment) of partial ITS sequences (610 bp) from *N. domei* sp. nov. and *N. benstonei* (AB675710.1) using BioEdit Sequence Alignment Editor. Unmatched nucleotides are not coloured and matching (conserved) nucleotides are represented by coloured blocks.



**Figure 14.** Phylogenetic tree (NJ method) reconstructed using ITS sequences obtained from the public database (NCBI) and ITS isolated in this study. Bootstrap values are as indicated above relevant branches and an American carnivorous pitcher plant (*Sarracenia flava*) serves as an outgroup.

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