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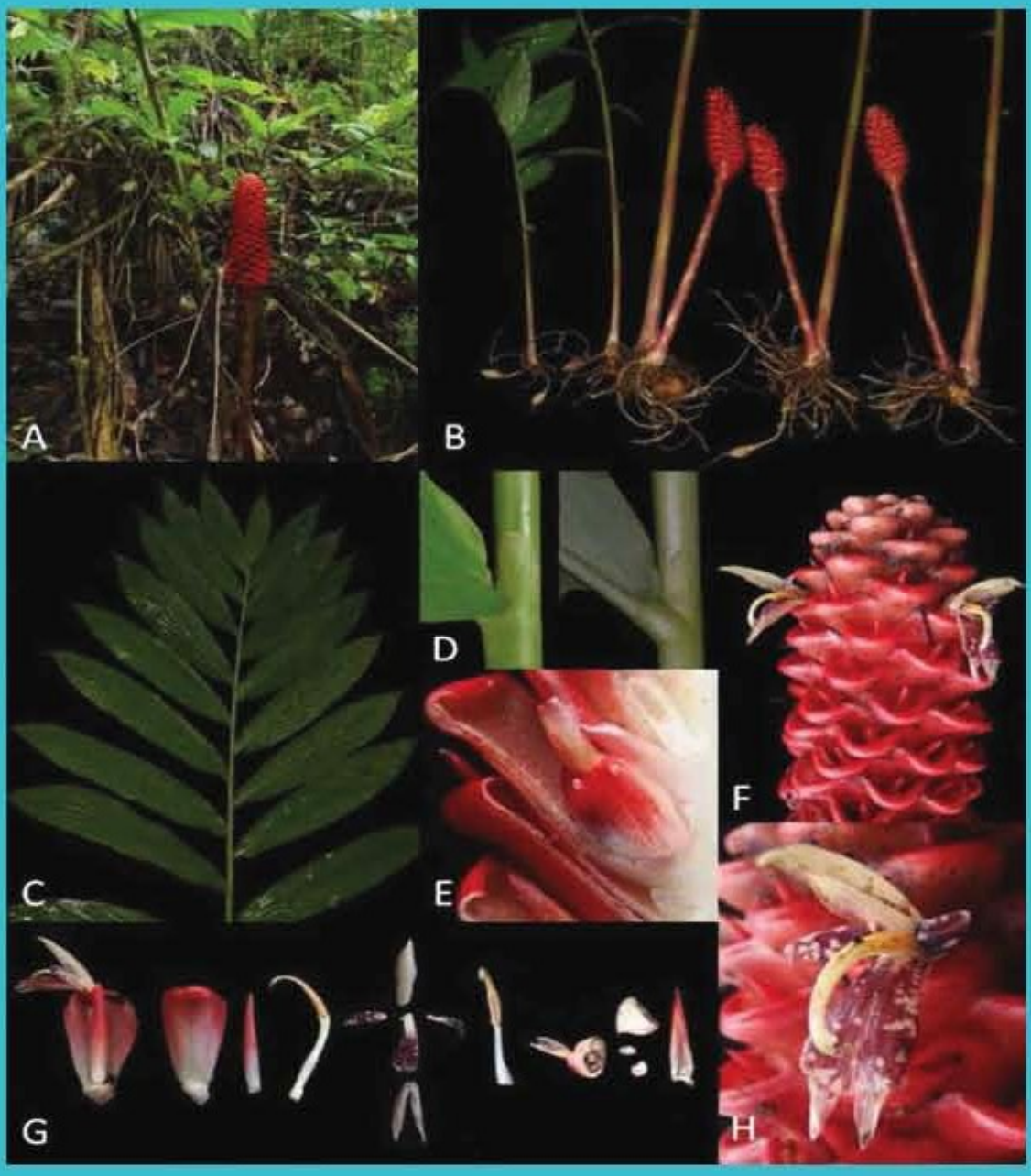

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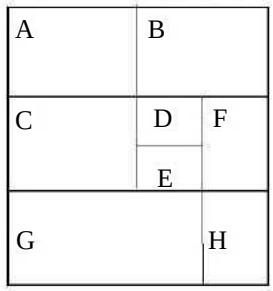

Cover images: *Zingiber engganoensis* Ardiyani. A. Habit B. Leafy shoot and the inflorescence showing rhizomes, roots and root-tuber C. Leaves D. Ligule and swollen petiole E. Dissection of inflorescence showing fruit F. Spike and flowers G. Dissection of flowers and fruits showing bract, bracteole, two lateral staminodes, two petal lobes, labellum, and the four appendages of the anther H. Flower. Source of materials: E190 (BO). Photo credits: B, C, D by Arief Supriatna. A, E, F, G, H by Marlina Ardiyani.

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NOTES ON MORPHOLOGICAL CHARACTERISTICS OF *EURYCOMA* SPP. AND ITS STATUS IN PENINSULAR MALAYSIA

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ABSTRACT
TAN, A. L., KAMAL, N. M., TAN, H. P. & ROSLAN, I. 2015. Notes on morphological characteristics of *Eurycoma* spp. and its status in Peninsular Malaysia. *Reinwardtia* 14 (2): 259 – 263. — A study had been carried out on the genus *Eurycoma* Jack that aimed to ascertain the diagnostic characteristics of the two species that occur in Peninsular Malaysia. Samples were collected from 15 localities comprising of forest reserves and plantations throughout Peninsular Malaysia covering the 5 regions *i.e.* northern, western, eastern, southern and central. The sampling was done to capture the morphological variations from different habitat. In general, morphologically both species were very similar. They could be clearly distinguished using their fertile parts. *Eurycoma longifolia* Jack had long, drooped inflorescences while in *E. apiculata* A.W. Benn was usually short, pointed upwards. Small differences were also noted on the leaflet of *E. apiculata* where the apex of the leaflet was often abruptly pointed while its base was rounded with conspicuous petiolule. On contrary, *E. longifolia* leaflet apex was usually subacute with its base asymmetrical and decurrent to its petiolule. In addition, anatomical transverse section of *E. longifolia* leaflet midrib outline and margin were dissimilar with *E. apiculata* by having convex abaxial surface and blunt tip margin compared to the slightly arc-shape abaxial and tapered margin in *E. apiculata.* The abundancy of *Eurycoma* spp. had decreased.

Key words: *Eurycoma,* morphological characters, Peninsular Malaysia.

ABSTRAK
TAN, A. L., KAMAL, N. M., TAN, H. P. & ROSLAN, I. 2015. Catatan karakteristik morfologi *Eurycoma* spp. dan statusnya di Semenanjung Malaysia. *Reinwardtia* 14 (2): 259 – 263. — Penelitian marga *Eurycoma* Jack telah dilakukan untuk memastikan karakter diagnostik dua jenis *Eurycoma* yang ditemukan di Semenanjung Malaysia. Sampel telah dikoleksi dari 15 lokasi yang meliputi hutan alam dan perkebunan di seluruh Semenanjung Malaysia. Lokasi tersebut mencakup 5 kawasan yaitu bagian utara, timur, barat, selatan dan tengah. Pengambilan sampel ini dilakukan untuk merekam variasi morfologi yang tampak pada habitat berbeda. Secara umum, kedua jenis tersebut mempunyai karakter morfologi yang hampir sama. Keduanya hanya bisa dibedakan dengan menggunakan bagian fertilnya. *Eurycoma longifolia* Jack mempunyai perbungaan yang panjang dan menjulur ke bawah sedangkan perbungaan *E. apiculata* A.W. Benn pendek dan tegak ke atas. Perbedaan kecil lainnya dapat dilihat pada anak daunnya. Anak daun *E. apiculata* melancip secara mencolok dengan pangkal yang membulat dan tangkai anak daun yang jelas. Sebaliknya, ujung anak daun *E. longifolia* pada umumnya agak lancip dengan pangkal yang asimetris dan melanjut hingga ke tangkai anak daun. Selain itu, anatomi dari sayatan melintang permukaan tulang daun dan tepi anak daun *E. longifolia* berbeda dengan *E. apiculata.* Hasil pengamatan anatomi dari sayatan melintang permukaan tulang daun dan tepi anak daun *E. longifolia* menunjukkan permukaan bawah yang cembung dan tepi yang tumpul, sedangkan pada *E. apiculata* permukaan bawah tulang daunnya hampir berbentuk seperti busur dengan tepi yang runcing. Penelitian ini juga mencatat bahwa keberadaan *Eurycoma* spp. telah berkurang.

Kata kunci: *Eurycoma*, karakter morfologi, Semenanjung Malaysia.

INTRODUCTION

The root of *Eurycoma longifolia* Jack, international trade name Longjack or Malaysian ginseng is a very popular medicinal herb that is claimed to be able to increase the libido and virility of men. In Malaysia, it is locally known as tongkat ali, setunjang bumi or pasak bumi. Traditionally, its root decoction had also been used as a febrifuge while the pounded root poultice can be pasted on wounds, ulcers and sores (Uji, 1999). In Brunei, the leaves are eaten raw to relieve stomach ache while its decoction is used for washing itchiness (Kochummen, 1983).

The genus of *Eurycoma* is confined to tropical South-East Asia which consists of 3 species. Only 2 species can be found in the Malesian region; namely the quite variable, widespread *E. longifolia* occurring from Myanmar through Indo-China and Thailand to Peninsular Malaysia, Sumatra, Borneo and the Phillipines; and *E. apiculata* A.W. Benn which is confined to Peninsular Malaysia and Sumatra (Uji, 1999). *Eurycoma* is a common understorey plant occurring from beach forest to lower montane forest (Kochummen, 1983). Since both species were very similar in habit and could occur simultaneously in primary and secondary forests, microscopic characteristics had been incorporated for additional assurance to distinguish them especially when the plants were sampled sterile. Besides that, the diagnostic anatomy characteristics of *Eurycoma* species might be able to help in confirming the presence of *Eurycoma* root in products with ground *Eurycoma* root. Thus, it was timely to establish the diagnostic morphological (macro and micro) characteristics of *E. longifolia* and *E. apiculata*.

MATERIALS AND METHODS

Morphological characterization

Collections of *Eurycoma* spp. were sampled from forest reserves and plantations covering the five regions (northern, southern, western, eastern and central) in Peninsular Malaysia. A total of 16 localities had been sampled. An average of 5 individuals sampled in each localities were used for morphologically characterization and anatomical analysis.

The leaves and fertile materials found were sampled for morphological characterization and preparation of herbarium specimens for species identification. Notes on the habitat and morphological characters of each sampling sites were made. The fertile specimens representative from the localities collected were then deposited at KEP while the sterile specimens were kept at Natural Products Division, FRIM specimen room. Furthermore, diagnostic characteristics of both *E. longifolia* and *E. apiculata* were determined. Specimens of *Eurycoma* species from FRIM herbarium (KEP) had been also examined to have a better understanding on the species that occur in Peninsula Malaysia.

Specimens collected from field during this study.

E. apiculata: TAA-BK-001 (Batu Kurau, Perak); TAA-ERS-001 (Endau Rompin NP (Selai, Segamat, Johor), TAA-BLG-002 (Bukit Lagong F.R., Selangor), TAA-TEB-001 (Taman Etnobotani, FRIM).

E. longifolia: TA-SGJ-001 (Labis F. R., Segamat, Johor), TA-SGJ-011 (Moakil F. R., Comp 293, Segamat, Johor), TA-LG-001 (Gunung Raya F. R., Lubuk Semilang, Langkawi, Kedah), TA-LG-011 (Pulau Singa Besar F. R., Langkawi, Kedah), TA-BLR-003 (Bukit Larut F. R., Taiping, Perak), TA-BK-002 (Pondok Tanjung F. R., Batu Kurau, Perak), TA-ERS-001 (Endau Rompin NP (Selai), Segamat, Johor), TA-BH-001 (Bukit Hari, FRIM), TA-MRN-004 (Stesen Penyelidikan FRIM Maran, Pahang), TA-STU-003 (Stesen Penyelidikan FRIM Setiu, Terengganu), TA-STU-013 (Hutan bris Setiu, Terengganu), TA-PAP-0012 (Pantai Acheh F. R., Pulau Pinang), TA-PJP-005 (Pulau Jerejak F. R., Pulau Pinang), TA-SM-007 (Semangkok F. R., Selangor), TA-PSH-003 (Pasoh F. R., Negeri Sembilan).

KEP specimens examined:

E. apiculata: 5887 (Sungai Kahang, Johor), 11602 (Ulu Kenderong, Gerik, Ulu Perak), 12707 (Maxwell Hill, Perak), 20210 (Rotan Tunggal F. R., Raub, Pahang), 21054 (Weld Hill F. R., K. L.), 22743 (Sungai Lalang F. R., Kajang, Selangor), 24175 (Bukit Enggang F. R., Kajang, Selangor), 33862 (near boundary Lagong F. R., Selangor), 36212 (Cameron Highland, Valley of Bertam), FRI 1951 (Ulu Gombak V. J. R., Selangor), FRI 2903 (Maxwell Hill, Tea Garden, Perak), FRI 3860 (Peta border, Ulu Endau, Pahang/Johore), FRI 6109 (Sungai Wang, Bubu F.R., Perak), FRI 8686 (banks of Sg. Kahang, Kluang Forest V. J. R., Johore), FRI 11892 (Ulu Sg. Pukin, Lesong F. R., SW Pahang), FRI 56627 (Jeram Toi, Berembun F. R., Jelebu, Negeri Sembilan), KEP 94643 (Bukit Lagong F. R., Selangor), SK 511 (Kuala Depang F. R., Perak), KL 425/KL 1443/KL 147 (K. Pansom, Bukit Tangkol, Ulu Langat, Selangor), KL 1255 (Kg. Lui, Ulu Langat, Selangor).

E. longifolia: FRI 23666 (Sg. Pinang F. R., Pulau Pangkor, Perak), 75m68 (Bukit Bauk), FRI 4723 (Taman Negara Pahang), FRI 7641 (Tg. Penawar, Johor Coast), 104600 (Trolak F. R.), 2641 (Weld Hills, K. L.), FRI 13963 (Bubu F. R., Perak).

Plant anatomy

Leaflet anatomy

Fixation, embedding and sectioning were made following Johansen (1940) and Sass (1958) with suitable modifications. Fresh leaf materials were

fixed in AA (1:3), of 25 % acetic acid and 70 % ethanol. Leaf transverse section of the specimens were sectioned with a sliding microtome at 20–30 µm thickness and stained in 1 % Safranin in 50 % alcohol and 1 % Alcian Green in 100 ml purified water with three drops of acetic acid. Sections were made from the middle and marginal parts of the leaflets lamina using a Reichert sliding microtome. All slides were mounted in Euparal after dehydration using alcohol series 50 %, 70 %, 95 % and 100 %. Finally, digital photos of the slides were taken for image analysis.

RESULTS AND DISCUSSION

Collections of *Eurycoma* species were sampled from 16 localities comprising of forest reserves and plantations throughout Peninsular Malaysia covering the 5 regions *i.e.* northern, western, eastern, southern and central. *Eurycoma longifolia* had been sampled from 14 localities while *E. apiculata* had been sampled from 3 localities *i.e.* Bukit Lagong F.R. (BLG), Pej. Renjer Sg. Sega, Batu Kurau (BK) & Endau Rompin NP (Selai), Segamat (ERS) (Table 1).

Both *Eurycoma* species were noted to be small to big treelet, dioecious, with spiral, imparipinnate/ paripinnate leaves, condensed on the top. The stems were usually covered with large rounded leaf scar, not branching when young to many branching on large treelet. The leaflets were opposite to subopposite, elliptic to lanceolate with very short petiolule (nearly sessile), with lobed secondary veins (Kochummen, 1983). The inflorescences are of axillary panicles, pubescent with many small pedicellate valvate flowers with 5-6 pubescent flower lobes. The male flower consists of 5-6 stamens with yellow anther while the female flowers with 5-6 adnate green carpels with reddish peltate, 5 lobed stigmas. Meanwhile, the fruits consist of 2-5 drupe nutlets on a stalk, green when young and turning red when riped.

Eurycoma longifolia could be differentiated from *E. apiculata* using its fertile material (Table 2). The inflorescences of *E. longifolia* were usually of long green (young) or red (mature) panicles, drooping (Fig. 1A & 1B) as opposed to *E. apiculata* with short or slightly compact green (young) or maroon (mature) panicles that were always pointed upright (Fig. 2A & 2B). The petals of *E. longifolia* were red, small, lanceolate or ovate-lanceolate, the opening rather constricted, puberulous on both lobe surfaces (Fig. 1A inset). On the other hand, the petals of *E. apiculata* were pinkish-cream, slightly bigger and longer, linear or oblong, the opening reflexed, puberulous on the outer lobes but glabrous inside (Fig. 2A). Meanwhile, the young fruit nutlets of *E. longifolia* were usually light green in comparison to *E. apiculata* which were usually yellowish green (Fig. 2B). These characteristics also coincide with the findings by Nooteboom (1972).

Table 1. Sampling localities of *Eurycoma* spp. in Peninsular Malaysia

No	Locality (Code)	Region	Remarks
1	Bukit Hari Research Plot, FRIM (BH)	Central	Planted
2	Pantai Acheh F.R., Pulau Pinang (PAP)	Northern	Wild
3	Pulau Jerejak F.R., Pulau Pinang (PJP)	Northern	Wild
4	Stesen Penyelidikan FRIM Setiu, Terengganu (STU)	Eastern	Planted & Wild
5	Pasoh F.R., Negeri Sembilan (PSH)	Central	Wild
6	Endau Rompin NP (Selai), Segamat, Johor (ERS)	Southern	Wild
7	Labis F.R., Segamat, Johor (SGJ)	Southern	Wild
8	Moakil F.R., Comp 293, Segamat, Johor (SGJ)	Southern	Wild
9	Gunung Raya F.R., Lubuk Semilang, Langkawi, Kedah (LG)	Northern	Wild
10	Pulau Singa Besar F.R., Langkawi, Kedah (LG)	Northern	Wild
11	Bukit Larut F.R., Taiping, Perak (BLR)	Western	Wild
12	Pondok Tanjung F. R., Batu Kurau, Perak (BK)	Western	Wild
13	Stesen Penyelidikan FRIM Maran, Pahang (MRN)	Eastern/ Central	Planted
14	Bukit Lagong F.R., Selangor (BLG)	Central	Wild
15	Pej. Renjer Sg. Sega, Batu Kurau, Perak (BK)	Western	Planted
16	Semangkok F.R., Kuala Kubu Baru, Selangor (SM)	Western	Wild

Table 2. Diagnostic morphological characteristics fertile parts and supporting characters that could differentiate both *Eurycoma* spp. that occur in Peninsular Malaysia

Characteristics	*E. longifolia*	*E. apiculata*
Inflorescence type	Long, rather complex, drooped panicle	Short, usually compact, pointed upright panicle
Flower: Petal	Opening rather constricted, ovate-lancolate, puberulous on both lobes surfaces	Opening reflexed, linear or oblong, puberulos on outer lobes, glabrous on the inside lobes surface
Colour	Red	Pinkish cream or reddish cream
Fruit colour	Light green turning red to maroon when ripen	Yellowish green turning red to maroon when ripen
Leaflet: Apex	Subacute or acute to acuminate	Abruptly pointed or acuminate
Base	Asymmetrical, cuneate, often decurrent to petiolule; petiolule not conspicuous	Rounded/ obtuse, occasionally asymmetrical, not decurrent with conspicuous petiolule

Table 3 Anatomical characteristics noted that could be used to distinguish both *Eurycoma* spp. in Peninsular Malaysia in the absence of fertile materials

Characteristics	*E. longifolia*	*E. apiculata*
Midrib outline	Convex abaxial surface	Slightly arc-shape abaxial surface
Margin shape	Blunt tip, slightly pointed downwards	Tapered tip, pointed downwards
Lamina trichomes (unicell)	Absent / Few present	Present in abundance
Petiolule trichomes (unicell)	Absent / Few present	Present in abundance

Fig. 1. *E. longifolia* Jack A. Drooping inflorescences, flower petals rather constricted opening, pubescent on both surfaces (flower inset-scale 5 mm) and B. Drooping infructescences; leaflet with acute to acuminate apex and asymmetrical base that tapered towards petiolule (B inset).

Fig. 2. *E. apiculata* A.W. Benn. A. Pointed upright inflorescences, flower petals reflexed, pubescent on outer surface, glabrous inside and B. Pointed upright infructescences, leaflets with abruptly acuminate apex and rounded base with conspicuous petiolule (marked with red rings).

Vegetatively, both species were very similar. However, certain characteristics on the leaflet could be used to distinguish these two species during the absence of fertile materials. The leaflet apex of *E. longifolia* was often subacute or acute to acuminate where the leaflet base was usually asymmetrical, cuneate, decurrent with very short petiolule (1−2 mm) (Fig. 1B inset). In comparison, the leaflet apex of *E. apiculata* was always abruptly pointed/acuminate while the leaf base was rounded, seldom asymmetrical, not decurrent with conspicuous short petiolule, 1–2 mm (Fig. 2B – red rings). The leaflet of *E. apiculata* also tend to be larger and wider, while the leaf was shorter compared to *E. longifolia* (Kochummen, 1983). However, these characteristics have to be used with caution as the leaves of *E. longifolia* could be short when they were young/cultivated and may varies in different habitats and the juvenile leaves of *E. longifolia* might be very large and wide with rather rounded basal.

Microscopically, the presences of simple, unicell trichomes were noted at the leaflet midrib and petiolule of both *Eurycoma* spp. (Table 3). Abundance of foliar sclereids were also found at the leaflet lamina tranverse section. In addition, the leaflet midrib transverse section also showed the presence of sclerenchyma sheath at the vascular bundles. These findings coincide with those reported by Khatijah (2006). Both species could be distinguished using the outline of the leaflet midrib and margin transverse section. *E. longifolia* midrib had convex abaxial (Fig. 3A) compared to slightly arc-shape abaxial (Fig. 3C) in *E. apiculata*. Moreover, *E. longifoila* had blunt tip, slightly pointed downwards (Fig. 3B) margin as opposed to *E. apiculata* with tapered, pointed downwards (Fig. 3D) margin. Meanwhile, the presence of unicell trichomes (Fig. 3E) in abundance at leaflet lamina, midrib and petiolule were noted in *E. apiculata.* This characteristic was absent in *E. longifolia* samples.

Preliminary findings showed that the abundancy of *Eurycoma* spp. had greatly decreased due to overharvesting. Much of the populations of *Eurycoma* at the forest edges from the localities visited were noted to be gone or with few individuals left. Therefore, some of the *Eurycoma* spp. could only be found at limited isolated area in the forest (*e.g.* Pondok Tanjung F. R., Bukit Larut F. R., Endau Rompin (Selai) National Park). Several localities (*i.e.* Bukit Larut F. R., Pulau Jerejak F. R.) where *E. apiculata* were sighted and collected in the past based on the herbarium records (KEP specimens) were no longer found in the area during our sampling visits. On the other hand, some robust populations of *E. longifolia* were also noted at Pulau Singa Besar F. R.,

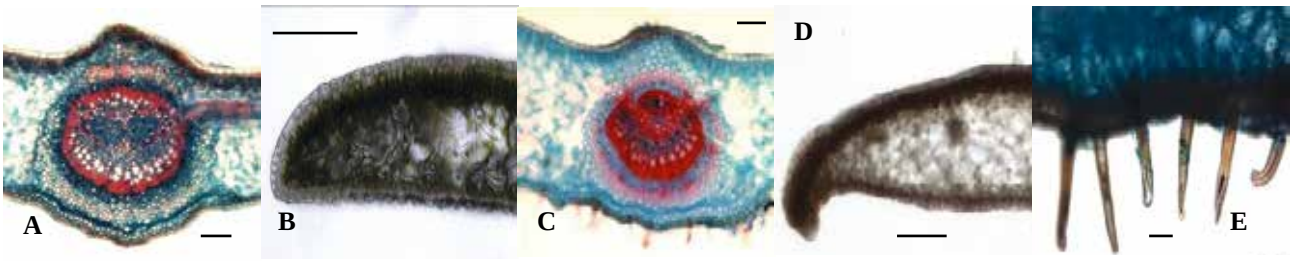

Fig. 3. Transverse Section (TS) of *E. longifolia* Jack A. Midrib with convex abaxial (scale 50 µm) and B. Margin with blunt tip, slightly pointed downwards (scale 50 µm). TS of *E. apiculata* A.W. Benn. C. Midrib with slightly arc-shape abaxial (scale 50 µm) and D. Margin with tapered tip, pointed downwards (scale 50 µm). E. The presence of simple, unicell trichomes (scale 20 µm) in abundance was noted at lamina, petiolule and midrib of *E. apiculata* leaflet.

Gunung Raya F. R., Pantai Acheh F. R. (Teluk Bahang National Park) and Moakil F. R.

CONCLUSIONS

In conclusion, both macroscopic and microscopic characteristics could be used in combination to distinguish both *Eurycoma* species especially when fertile materials were unavailable. Both species could be morphologically distinguished using the fertile materials (inflorescences) and microscopically distinguished using the characteristics of the leaflet midrib and margin outline. Preliminary findings showed that the population of *Eurycoma* spp., especially *E. apiculata* had greatly decreased.

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Map/line drawing illustration/photograph. Map, line drawing illustration, or photograph preferably should be prepared in landscape presentation to occupy two columns. Illustration must be submitted as original art accompanying, but separated from the manuscript. The illustration should be saved in JPG or GIF format at least 350 pixels. Legends or illustration must be submitted separately at the end of the manuscript.

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