Journal of Insect Biodiversity and Systematics
ISSN: 2423-8112
2026 | 12 (03), 633–646
Website: https://jibs.modares.ac.ir
Contents list: https://www.biotaxa.org/jibs
https://doi.org/10.48311/jibs.12.03.633

Original Article

Morphological and Molecular characterization of Mealybugs (Hemiptera: Coccomorpha: Pseudococcidae) Associated with *Hibiscus rosa-sinensis* (Malvaceae)

Sultan Ahmmed [1,3] | **Wei Hong Lau** [1] | **Nur Azura Adam** [1] | **Uma Rani Sinniah** [2]

1 Department of Plant Protection, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia. Emails: sultanbjri1984@gmail.com; lauweih@upm.edu.my; nur_azura@upm.edu.my

2 Department of Crop Science, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia. Email: umarani@upm.edu.my

3 Department of Entomology, Bangladesh Jute Research Institute, Manik Mia Avenue, Dhaka-1207, Bangladesh.

Corresponding authors: Wei Hong Lau | ✉ lauweih@upm.edu.my; Sultan Ahmmed | ✉ sultanbjri1984@gmail.com

https://zoobank.org/urn:lsid:zoobank.org:792FEB38-8A31-4574-97D9-AC0132235B5E

Academic Editor
Mehmet Bora Kaydan

Received
January 28, 2026

Revised
June 09, 2026

Accepted
June 10, 2026

Published online
June 16, 2026

ABSTRACT. Complexities of the identification of mealybugs may compromise effective management practice for these pests. *Hibiscus* species (Malvaceae) are frequently attacked by different mealybug species globally. Nevertheless, little information is available concerning the mealybug fauna associated with *H. rosa-sinensis* in Malaysia. The aim of this study was to identify the mealybug species infesting the *H. rosa-sinensis* by morphological and DNA barcoding. The DNA barcoding study was performed by comparing the sequencing of the universal barcode DNA region, cytochrome c oxidase subunit I (COI), and ribosomal (*18S* and *28S*). The nucleotide sequences confirmed the mealybug species, with the closest BLAST match in the GenBank queries sharing above 97% support. Based on taxonomic characterization by using scanning electron microscopy and DNA barcoding, three mealybug species, *Paracoccus marginatus, Maconellicoccus hirsutus,* and *Phenacoccus solenopsis* were confirmed in this study. DNA barcoding, along with the detailed external ultra-structural analysis of the mealybugs, could add a new dimension for identifying the mealybug species rapidly and more accurately. Ultimately, this study will help in decision-making in management practice for mealybugs.

KEYWORDS: DNA barcoding, Identification, Morphology, Pest management, Ultra-structure

Citation: Ahmmed, S., Wei, H.L., Adam, N.A. & Sinniah, U.R. (2026) Morphological and Molecular characterization of Mealybugs (Hemiptera: Coccomorpha: Pseudococcidae) Associated with *Hibiscus rosa-sinensis* (Malvaceae). *Journal of Insect Biodiversity and Systematics*, 12 (03), 633–646.

INTRODUCTION

Mealybugs (Hemiptera: Coccomorpha: Pseudococcidae) constitute a very diverse group, with 2065 species belonging to 263 genera described worldwide (García Morales et al. 2016). They cause major losses on cultural and ornamental plants worldwide by transmitting viruses and toxins, which cause defoliation, chlorosis, and vigor losses of the plants, and favor the development of sooty mold. They can infest all parts of the plants under a suitable environment. Mealybugs have been reported as a serious pest of *Hibiscus rosa-sinensis* in Pakistan, India, Nigeria, and Malaysia (Akintola & Ande 2008; Hodgson et al. 2008; Sartiami et al. 2016). This plant is one of the most widely cultivated ornamental flower plants throughout the world, and is native to tropical and sub-tropical regions (Magdalita & Pimentel 2010). To date, five mealybug species, namely *Ferrisia dasylirii* Cockerell, 1896; *Maconellicoccus hirsutus* Green, 1908; *Paracoccus marginatus* Williams & Granara de Willink, 1992; *Phenacoccus solenopsis* Tinsley, 1898, and *Pseudococcus jackbeardsleyi* Gimpel & Miller, 1996, have been reported infesting *Hibiscus rosa sinensis* in Malaysia (Kumar et al. 1997; Sartiami et al. 2016).

ISSN 2423-8112 – **Published by** Tarbiat Modares University | **Owner** The Entomological Society of Iran

The identification of mealybugs is complex and limited by a high degree of similarity and polymorphism, especially in the nymphal or egg stages, which poses challenges in the study and management of mealybugs (Pacheco Da Silva et al. 2014). Several taxonomic studies revealed that mealybugs exhibited a huge morphological variation due to environmental change, which led to a huge dilemma in identifying insects (Hodgson et al. 2008; Miller et al. 2014; Singh & Singh 2014). Jhala et al. (2008) reported that the coexistence of *Phenacoccus solani* and *P. solenopsis* on cotton led to issues with identification. Despite the difficulties involved in differentiating the mealybug species, correct identification is essential when dealing with species that are considered pests. Identification of an insect begins with the external morphological characteristics. According to the searched literature, the slide-mounted technique, together with light microscopic examination, has been used for mealybug identification by many researchers (Pacheco Da Silva et al. 2014; Sartiami et al. 2016). A combination of slide-mounted technique and DNA barcoding for mealybug identification has been conducted by Beltrà et al. (2012) and Ricupero et al. (2021). However, this conventional technique has several drawbacks. Firstly, this technique requires expertise in specimen handling, careful preparation of slide-mounted specimens, and expert knowledge of the insect group. It is time-consuming and labor-intensive for accurate identification of mealybugs (Bisevac 1997; Rugman-Jones et al. 2009). Secondly, several environmental factors may induce morphological variation in mealybugs, which in certain cases makes it hard to distinguish between complexes of cryptic species (Charles et al. 2000; Beltrà et al. 2012). These challenges can be addressed by identifying the species through the complementary study of molecular characterization, along with a microscopic study of the morphological features. The external structures such as: ostioles, vulva, claw digitules, cerarii, number of antenna segments, multilocular disc pores, trilocular pores, presence of translucent pores on hind legs, circulus, oral rim tubular ducts, and anal lobe bar of six economically important pest mealybug species (*Coccidohystrix insolita* Green*, Dysmicoccus brevipes, D. neobrevipes Beardsley, Maconellicoccus hirsutus, Phenacoccus solenopsis and Planococcus lilacinus*) in Sri Lanka have been studied by using Scanning Electron Microscope (SEM) (Kumar et al. 1997; Sirisena et al. 2015).

Meanwhile, many researchers have used the DNA barcoding approach to identify Pseudococcidae (Correa et al. 2011; Downie & Gullan 2004; Hardy et al. 2008; Malausa et al. 2011). DNA barcoding can be straightforward for non-experts, particularly those who routinely identify a lot of samples regardless of the stage of development and geographical origin (Sarvananda 2018). Ribosomal DNA, such as subunits 18S and 28S, internal transcribed spacers, and mitochondrial DNA, such as cytochrome oxidase I (*COI*) and *COII,* are commonly used for insect species and subspecies differentiation (Simon et al. 1994; Li et al. 2005; Rung et al. 2008). *COI* gene has been established as the DNA barcode for initial species identification (Hajibabaei et al. 2006; Linares et al. 2009). Nuclear genes are needed for discrimination between species when two groups of organisms share the same DNA barcode(s), but they do not belong to the same species (Hebert et al. 2003). A combination of nuclear and mitochondrial genes is common in mealybug identification. For example, Ashfaq et al.(2010) identified *P. solenopsis* in Pakistan through DNA sequencing using partial nucleotide sequences of nuclear (elongation factor-1α, ribosomal DNA subunits *18S* and *28S*) and mitochondrial (*COI*) genes. Palma-Jimenez and Blanco-Meneses (2016) also used the nuclear and mitochondrial genes for identification of *Pseudococcus* sp. in Costa Rica, whereas Mohammed et al. (2022) used mitochondrial (*COI*) genes for identification of mealybugs and ant species associated with pineapple in Malaysia.

A combination of slide-mounted technique and DNA barcoding for mealybug identification has been conducted by Beltrà et al. (2012) and Ricupero et al. (2021). Nowadays, DNA barcoding has rapidly developed as a powerful tool in taxonomy; however, the accuracy of sequences and related raw information in public repositories is often questionable. Sometimes, drawbacks of DNA barcoding for insect identification include a lack of comprehensive barcode libraries, the difficulty with DNA degradation in old or processed samples, challenges in distinguishing closely related or hybrid species, and a reliance on morphological data to complement its findings. Therefore, the aim of this study was to combine the SEM and molecular approaches as a new dimension to identify the mealybugs found on the *Hibiscus rosa-sinensis*. Report on the ultra-structural study and molecular approaches for the identification of mealybugs associated with *H. rosa-sinensis* plant in UPM, Malaysia, is not available to date and has not been reported yet. They were morphologically different and varied in their body colour. This ultra-structural study, coupled with molecular identification of mealybugs, will provide a more comprehensive characterization of different mealybug species found on the same host plants.

MATERIAL AND METHODS

Collection of specimens. Mealybug specimens associated with *Hibiscus rosa-sinensis* were collected from the vicinity of Universiti Putra Malaysia, Selangor, Malaysia. Three sampling sites within the same geographical region were inspected for mealybugs, with a distance of around 1.5 kilometers between sampling sites located at a latitude of N 02˚59', a longitude E 101˚43', and an altitude of 64 m above sea level. Mealybug specimens with infested twigs were brought to the Laboratory of Insect Pathology, and specimens were stored in ethanol 95% at -20 ˚C separately for microscopic and molecular characterization. The mealybugs were differentiated by their body color upon sampling specimens. Further identification was conducted based on the taxonomy keys of Williams and Granara de Willink (1992), Miller and Miller (2002), and Williams (2004). The adult females were used in the insect identification as they were easily spotted and sampled on the infested plants. Fresh specimens were kept at -5 ˚C prior to structural examination. The body shape, size, colour, and legs of mealybugs (with waxy coating) were examined under a Dino-Eye® eyepiece camera (ANMO® Electronics Corporation, Taiwan), which was connected to a Wild Heerbrugg M3Z Stereo microscope (Leica®, Switzerland). Dino Capture 2.0 software was used to capture photos of mealybugs under the Dino-Eye eyepiece camera. To study the detailed external structure of the mealybugs, Scanning Electron Microscope (SEM) (JEOL® JSM-7600F, JEOL Ltd., Tokyo, Japan) is used following the protocol of Sirisena et al. (2015).

DNA Extraction, amplification, and Sequencing. Total genomic DNA of each single mealybug specimen was extracted using the NucleoSpin® Tissue Total DNA Extraction Mini Kit (Macherey-Nagel, Germany) according to the manufacturer's instructions. The final genomic DNA extract was stored at −20 °C for subsequent PCR amplification. Three pairs of primers were used in the study, namely the *COI*, *18S,* and *28S ribosomal* (*rDNA)* genes (Table 1). The Promega Go Taq® Green Master Mix was used to amplify the *COI*, *18S rDNA,* and *28S rDNA* genes of mealybugs according to the manufacturer's protocol. The reactions and cycling conditions were performed in an automated thermocycler Mastercycler® Pro S. A total volume of 25 µl reaction mixture containing 2.5 µl of 25–30 ng DNA template, 12.5 µl of Green Master Mix, 2.5 µl of the primers (10µM Forward and Reverse each), and 5µl nuclease free water. PCR products were separated on 1% (w/v) agarose gel, and PCR bands were stained with GreenSafe DNA Gel Stain. Positive PCR products were sent to Apical Scientific Sdn. Bhd.Selangor, Malaysia, for nucleotide sequencing. The sequencing results retrieved in FASTA format were analyzed and edited using Sequence Scanner 2, BioEdit program v7.0.5 (Hall 1999). The nucleotide sequences of *18S rDNA, 28S rDNA,* and *COI* genes of mealybug specimens were subjected to a homology search using BLASTn (https://blast.ncbi.nlm.nih.gov/Blast.cgi) and aligned with the ClustalW MEGAX program version 10.1.8 (Kumar et al. 2018). The newly generated nucleotide sequences were compared with each other and the published sequences in the GenBank through the Basic Locus Alignment Search Tool (BLASTn) tool available on the National Center for Biotechnology Information website (https://www.ncbi.nlm.nih.gov/).

Phylogenetic Analysis. Phylogenetic analyses were conducted using MEGAX (Kumar et al. 2018). The Maximum Likelihood method was used for clustering of mealybug specimens among themselves with 1,000 bootstrap replications to evaluate the branching confidence (Felsenstein 1985).

Table 1. Molecular markers used in the identification of mealybugs associated with *H. rosa-sinensis.*

Gene	Primers	Sequence(5′-3′)	PCR conditions	PCR product	Sources
CO1	C1-J-1718-F	GGAGGATTTGGAAATTGATTAGTTCC	94˚C for 2 min; 30 cycles of 94˚C for 30s, 60˚C for 30s, 72˚C for 1m; 72˚C for 5 min	473 bp	Simon et al. (1994)
	C1-N-2191-R	CCCGGTAAAATTAAAATATAAACTTC			
COI	C1-J-2183-F	CAACATTTATTTTGATTTTTTGG	94˚C for 2 min; 35 cycles of 94˚C for 30 s, 45˚C for 1 min; 72˚C for 1 min; 72˚C for 5 min	385 bp	Malausa et al. (2011)
	C1-N-2568-R	GCWACWACRTAATAKGTAT CATG			
28S	28S-F	ACCGTTCGGGGGTAAACGGACAG	94˚C for 2 min; 30 cycles of 94˚C for 1 min, 48˚Cfor 1 min, 72˚C for 1 min 30 s; 72˚C for 5 min	320 bp	Malausa et al. (2011)
	28S-R	TCGGAAAGAGGCCCGAAGATTCG			
18S	18S-2880-F	AGAATTAAGCCATGCATGTCTCAG	94˚C for 2 min; 30 cycles of 94˚C for 30 s; 55˚C for 30 s; 72˚C for 2 min; 72˚C for 5 min	630 bp	Malausa et al. (2011)
	18SB-R	TNGCCCTGCCTAATTGATCCTCG			

RESULTS

Based on external structural features, they were identified as *Phenacoccus solenopsis, Paracoccus marginatus,* and *Maconellicoccus hirsutus* (Fig. 1). Different species of mealybugs exhibited distinct body colors. *M. hirsutus* had a reddish-brown body, while *P. solenopsis* appeared whitish, and *P. marginatus* was yellowish. *Phenacoccus solenopsis* was the largest among them and featured two distinct blackish dorsal stripes, which are absent in *Par. marginatus* and *M. hirsutus*. When soaked in 80% (v/v) alcohol, *M. hirsutus* remained brownish, while *P. marginatus* turned black within days. The adult female of *P. solenopsis* had an oblong, wingless body (3.0–4.5 mm long, 1.2–2.5 mm wide), whitish to light green with blackish stripes along the mid-dorsal line (Fig. 1A). It was well-segmented, covered in wax except at the posterior abdomen, and surrounded by waxy filaments. An egg sac was present beneath the body. *Phenacoccus solenopsis* had three pairs of brownish-red legs and 9-segmented antennae with a long apical segment and conspicuous antennal setae (Fig. 2A). Leg was three pairs and well developed (Fig. 2B), digitules of claw slightly longer than the claw and thicker than the tarsal digitules (Fig. 2C). Ostioles as a single pair and lightly sclerotized (Fig. 2D), possessed 18 pairs of cerarii, each with two conical setae and trilocular pores, with the longest posterior pair on the anal lobe, each with 2 lanceolate setae and a few trilocular pores (Fig. 2E, 2H). The circulus large, oval, and flaccid (Fig. 2F); dorsal multilocular disc pores absent, ventral multilocular disc pores present (Fig. 2G). Ventral multilocular disc pores present in a single row along the posterior edges of abdominal segments VI–VIII, while dorsal multilocular disc pores are absent. Oral collar ducts are not numerous, present on venter only (Fig. 2I).

The adult female of *Maconellicoccus hirsutus* had a soft, elongate-oval, slightly flattened body (2.0–3.0 mm long, 0.8–1.5 mm wide), orange-pink to reddish, with no stripes or blotches and sparsely covered in white mealy wax (Fig. 1B). During maturation, it secreted a sticky, elastic substance and developed a wax filament ovisac covering the entire body. Infested plants showed stunted or bunchy growth. The ventral surface had normal setae (S); trilocular (T) pores (Fig. 3F) and multilocular (M) disc pores (Fig. 3E) appeared evenly on the body (Fig. 3A). Ostioles well-organised (Fig. 3B). The nine-segmented antennae measured 370–450 μm (Fig. 3C), and legs was well developed hindleg without translucent pores (Fig. 3D); cerarii had a pair of short conical setae with large setal sockets. Oral rim tubular ducts are numerous, evenly distributed, and have sclerotized rims with often unclear outer margins (Fig. 3G). In the case of *Paracoccus marginatus*, the adult female had a light yellow to greenish-yellow, waxy, elongated-oval body with light yellow legs. It measured 1.5–2.5 mm in length and 0.8–1.5 mm in width. The dorsum lacked stripes, and a white filamentous wax egg sac extended to three to four times the body length, eventually covering the female (Fig. 1C). The 8-segmented antennae had stout, fleshy setae, while the legs lacked them (Fig. 4A). The dorsal surface had short, slender setae, and there are 15–17 pairs of cerarii, with the posterior pair significantly longer. Translucent pores present only on the coxa of the hind legs (Fig. 4B). Ostioles present (Fig. 4C).

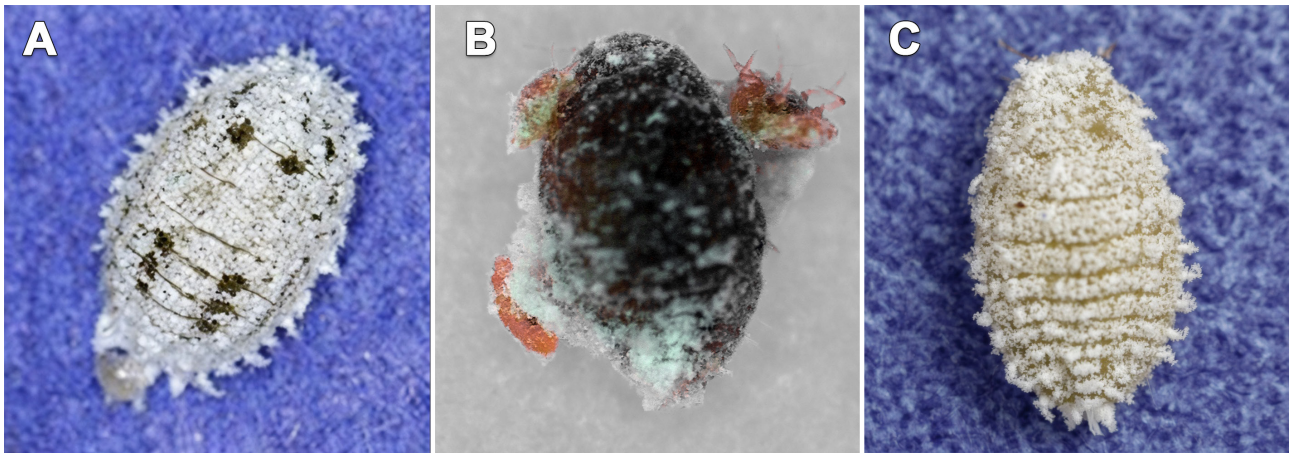

Figure 1. Collected Mealybugs specimens from the *Hibiscus* plant. **A.** *Phenacoccus solenopsis* Tinsley, 1898; **B.** *Maconellicoccus hirsutus* Green, 1908; **C.** *Paracoccus marginatus* Williams and Granara de Willink, 1992.

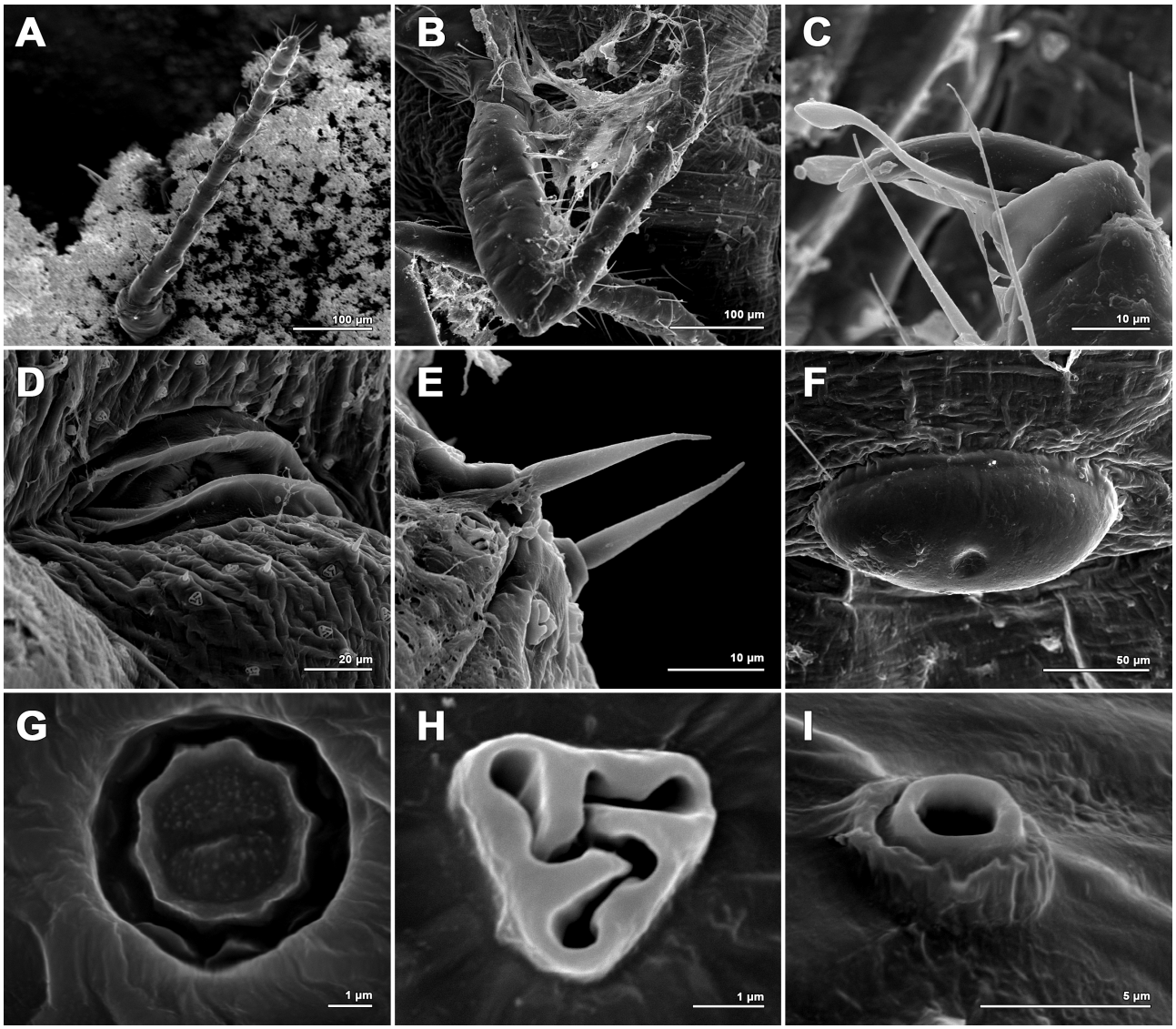

Figure 2. Scanning electron micrographs of *Phenacoccus solenopsis* Tinsley, 1898. **A.** Antenna; **B.** Leg; **C.** Digitules; **D.** Ostiole; **E.** Cerarius with trilocular pore and only two conical setae; **F.** Circulus multilocular pore; **G.** multilocular disc pores; **H.** Tubular duct; **I.** Oral collar ducts.

It normally possessed a pair of claw digitules, knobbed at the apex (Fig. 4D); Tubular ducts with trilocular pores were scattered on the body (Fig. 4F), ventral multilocular pores were lacking on the lateral areas of the abdomen (Fig. 4E), while oral rim tubular ducts were confined to the dorsal and ventral margins (Fig. 4F).

The markers used in this study had successfully generated PCR products of 630 bp (*18S rRNA* gene), 320bp (28S rRNA), 385 bp (*COI* gene), and 473 bp (*COI* gene), respectively. Primer *COI* (1718) did not provide a positive result for *P. marginatus,* while primer *COI* (2183) only showed a positive result for *P. marginatus*. Sequence similarity searches in BLAST revealed a high percentage of sequence homology between the UPM mealybugs and the published sequences of respective mealybug species available in GenBank. Based on the *COI*, *18S* and *28S* genes, the Blast hit result (NCBI) revealed the query sequence of UPM *P. solenopsis* (UPM-COI-MT073415, UPM-18S-MN967099, and UPM-28S-MW035048) were 98-100% homology to the reference sequences of similar species reported in China (COI-MK164397, 28S-MK873095 & 18S-JF965407), USA (28S-MH248360), Pakistan (18S-AB439210), Egypt (28S-MN887777 & COI-MG813769), India (28S-MH248352 & COI-MF770708). The query sequence of UPM *P. maginatus* (UPM-COI-MT309692, UPM-18S-MN967101, and UPM-28S-MW227324) also has 98–100% homology to similar species reported in India (COI-MG833839), China (COI-KJ187495), USA (COI-EU267201, 18S-EU188580).

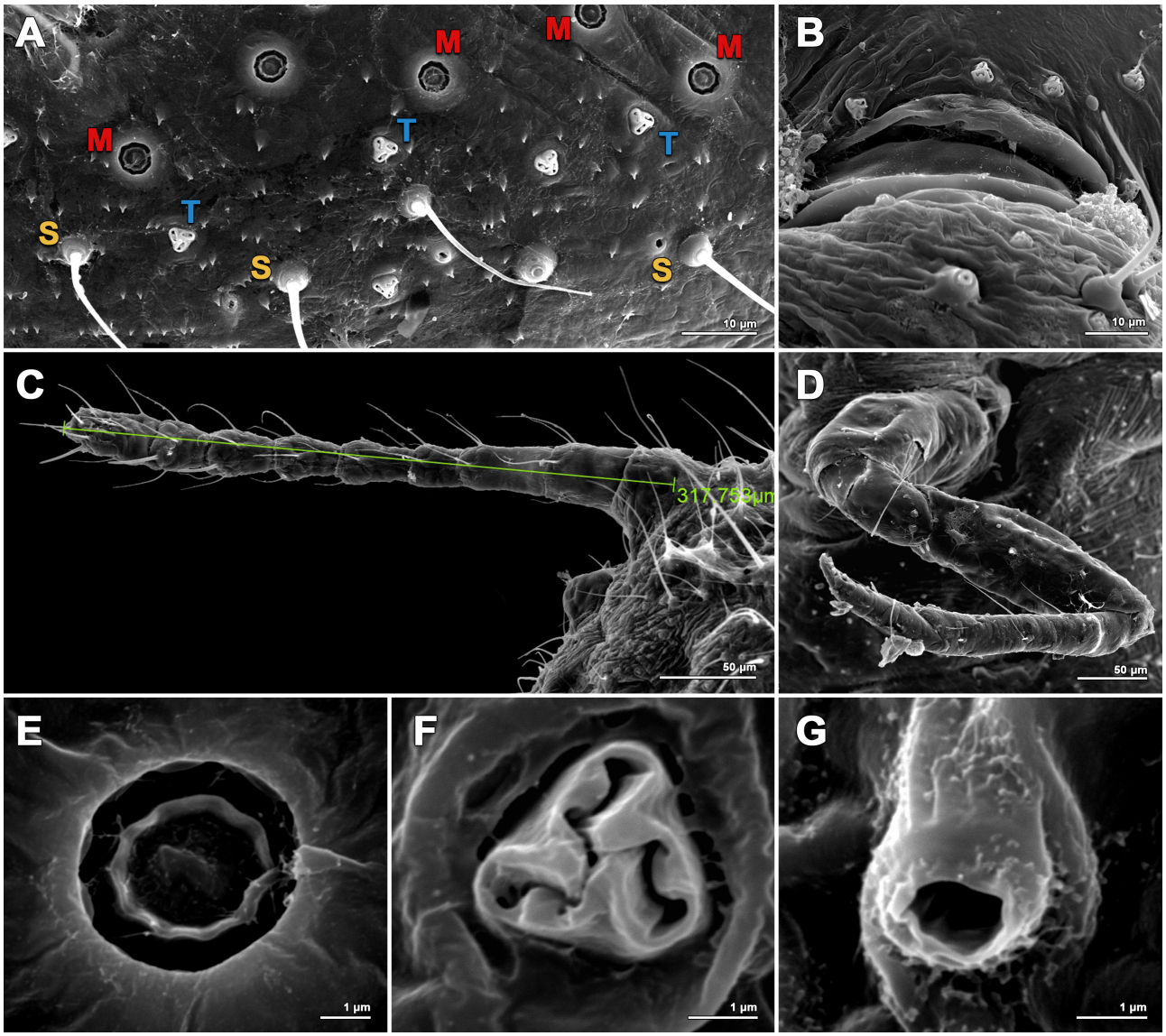

Figure 3. Scanning electron micrographs of *Maconellicoccus hirsutus* Green,1908. **A.** Ventral view of adult female with setae (S), trilocular pores (T), and multilocular disc pores (M); **B.** Ostiole; **C.** Antenna; **D.** Hind leg without translucent pores; **E.** Multilocular disc pore (M); **F.** Trilocular pore (T); **G.** Tubular duct.

The UPM *Maconellicoccus hirsutus* (UPM-COI-MW857557, UPM-18S-MN967100, and UPM-28S-MW227324) scored >/ 95% nucleotide sequence homology to the same species in GenBank from China (COI-KJ187533 and 28S-KY211356), USA (COI-EU267200, COI-AF483207, and 18S-AY426033), Thailand (28S-AY427322), and Egypt (28S-JQ085529). Based on the nucleotide sequence analysis, the mealybug specimens were confirmed as *P. solenopsis* Tinsley, *M. hirsutus* Green, and *P. marginatus* Williams and Granara de Willink.

The phylogenetic tree was constructed using maximum likelihood (ML) methods with the Tamura-Nei model (Tamura & Nei 1993). The topologies obtained based on all the loci and individual loci were mostly similar (Figs 5–7) but varied in resolution at internal nodes. The phylogenetic tree generated from concatenated sequences of UPM *P. solenopsis, M. hirsutus,* and *P. marginatus* from UPM formed a monophyletic clade within the same genus but distinct from other genera in the GenBank database. The concatenated sequences of UPM *P. solenopsis* (UPM-18S-MN967099, UPM-28S-MW035048 and UPM-COI-MT073415) had bootstrap values of 98%, 98%, and 100%, respectively; UPM *P. marginatus* (UPM-18S-MN967101, UPM-28S-MW227324, UPM-COI-MT309692) showed 100%, 89%, and 35–98% bootstrap support, while UPM *M. hirsutus* (UPM-18S-MN967100, UPM-28S-MW227324, UPM-COI-MW857557) had 97%, 99%, and 95% bootstrap support with closely related GenBank sequences.

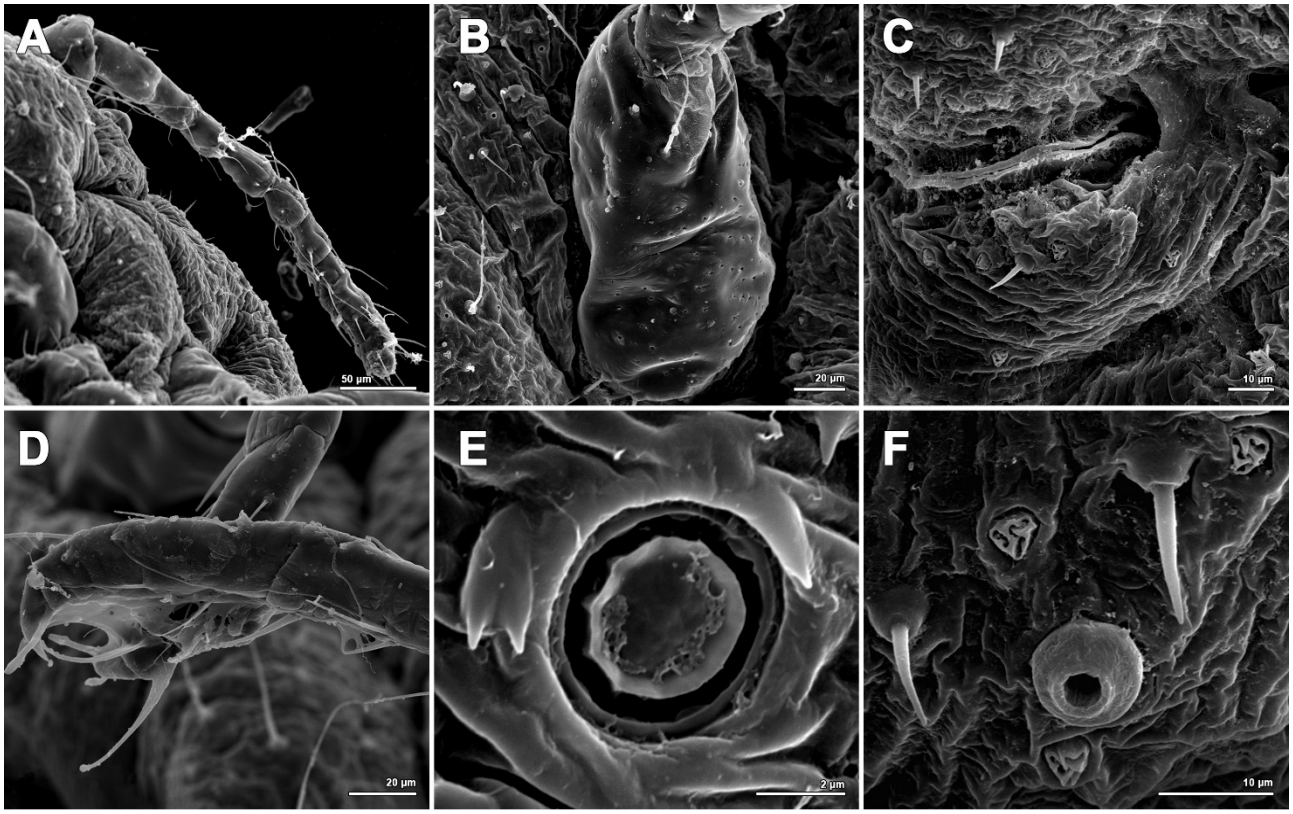

Figure 4. Scanning electron micrographs of *Paracoccus marginatus* Williams & Granara de Willink, 1992. **A.** Antenna; **B.** Translucent pores on the hind coxa; **C.** Ostiole; **D.** Digitules; **E.** Multilocular disc pore; **F.** Tubular duct.

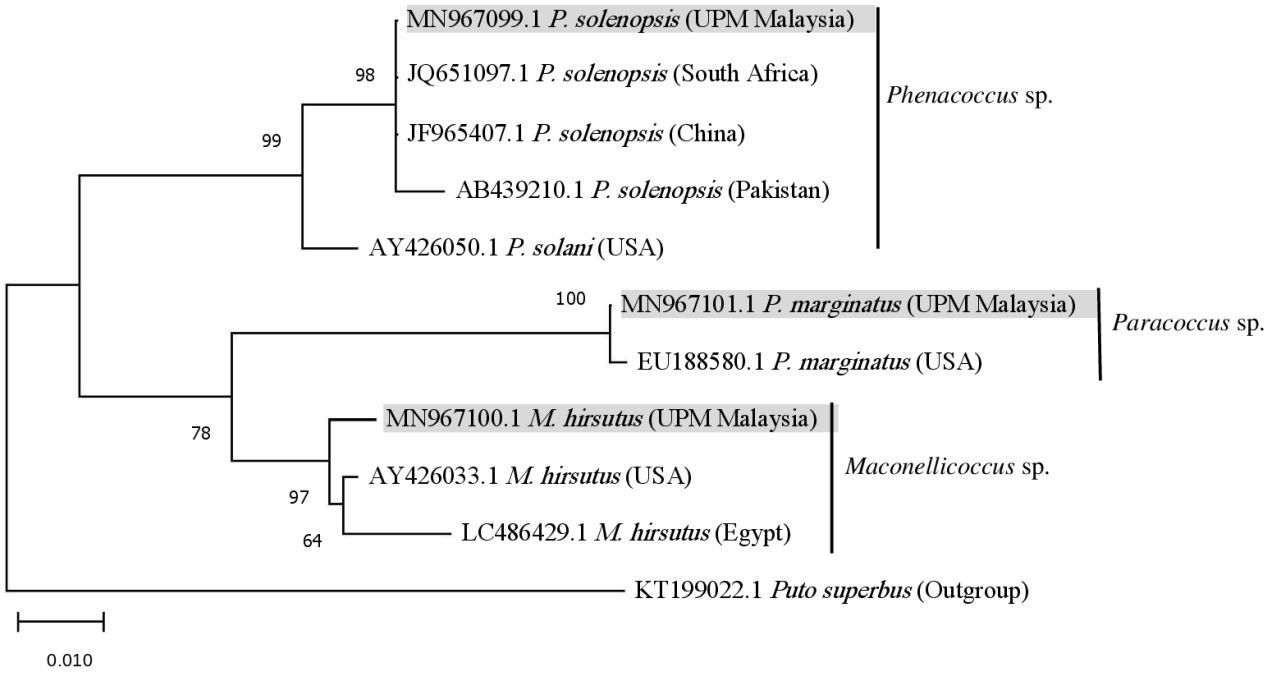

Figure 5. Phylogenetic tree based on 18S ribosomal haplotypes of *M. hirsutus*, *P. marginatus*, and *P. solenopsis* using the Maximum Likelihood method with 1000 bootstrap replicates.

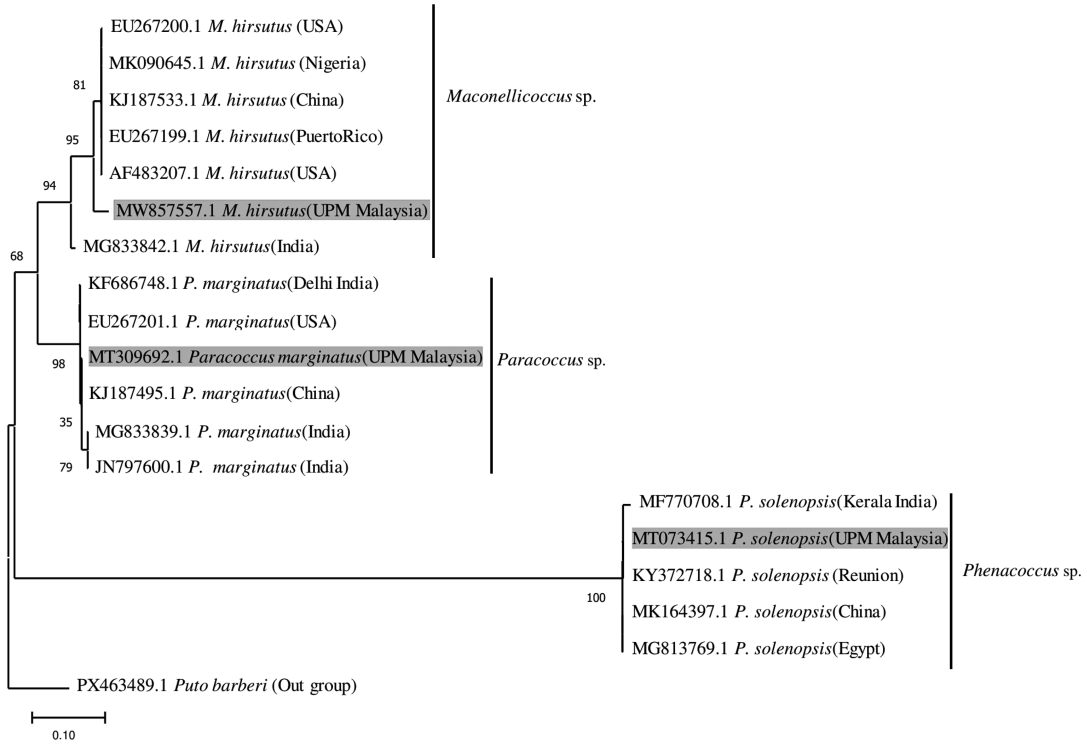

Figure 6. Phylogenetic tree based on *COI* (*mtDNA*) mitochondrial haplotypes of *M. hirsutus, P. marginatus,* and *P. solenopsis* using the Maximum Likelihood method with 1000 bootstrap replicates.

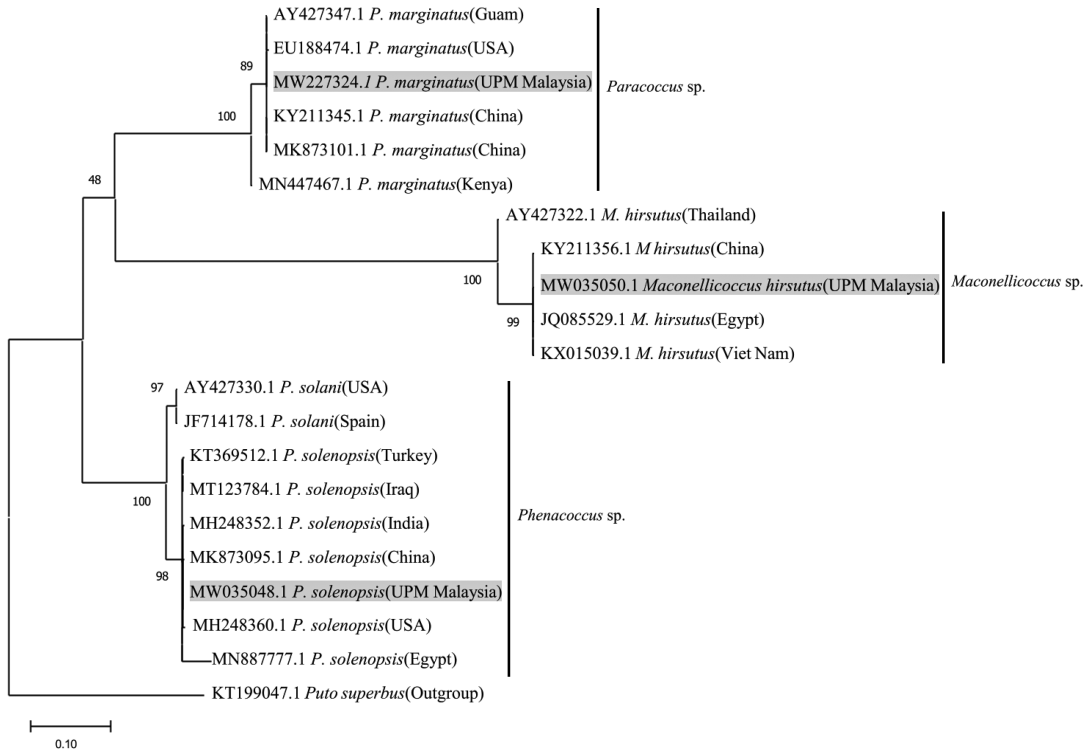

Figure 7. Phylogenetic tree based on *28S* ribosomal haplotypes of *M. hirsutus, P. marginatus,* and *P. solenopsis* using the Maximum Likelihood method with 1000 bootstrap replicates.

This study provides the first molecular data record of these mealybug species on *Hibiscus rosa-sinensis* in UPM, Malaysia (Table 2).

Table 2. Identification of mealybug species collected from *H. rosa-sinensis* in Malaysia.

Gene	Mealybug Species	Voucher accession number
18S	*Phenacoccus solenopsis* Tinsley, 1898	MN967099
	Paracoccus marginatus Williams and Granara de Willink, 1992	MN967101
	Maconellicoccus hirsutus Green,1908	MN967100
COI	*Phenacoccus solenopsis* Tinsley, 1898	MT073415
	Paracoccus marginatus Williams and Granara de Willink, 1992	MT145912
	Maconellicoccus hirsutus Green,1908	MT309692
28S	*Phenacoccus solenopsis* Tinsley, 1898	MW 035048
	Paracoccus marginatus Williams and Granara de Willink, 1992	MW 035050
	Maconellicoccus hirsutus Green,1908	MW 227324

DISCUSSION

Three mealybug species, *Phenacoccus solenopsis, Maconellicoccus hirsutus*, and *Paracoccus marginatus,* were found infesting *Hibiscus rosa-sinensis* at Universiti Putra Malaysia. *Phenacoccus solenopsis* exhibited morphological features consistent with descriptions by Williams (2004), Hodgson et al. (2008), Abbas et al. (2009), and Sirisena et al. (2015), featuring distinct blackish stripes along the mid-dorsal line, a feature absent in *M. hirsutus* and *P. marginatus*. It closely resembles *P. solani* but differs by having 9-segmented antennae, a large circulus, transparent femur pores, and multilocular disc pores on abdominal segments VI–VIII, with segment VII having pores at both ends. In contrast, *P. solani* has 8- or 9-segmented antennae and disc pores on segments IV or V to VIII, with segment VII having pores only at the posterior end. Hodgson et al. (2008) suggested they may be environmentally induced variants of the same species.

Few researchers documented SEM observation on the detailed external structure (ostioles, vulva, claw digitules, cerarii, different types of wax pore, body shape, number of antenna segments, multilocular pore, trilocular pore, posterior legs, the existence of translucent pores, circulus, oral rim tubular ducts and anal lobe bar) of very tiny insects such as scale insect (Zhang et al. 1988), *Maconellicoccus hirsutus, P. solenopsis* (Kumar et al. 1997; Sirisena et al. 2015), *Ferrisia virgata*, *Phenacoccus manihoti* and *Planococcus citri* (Cox & Pearce 1983), *Paracoccus marginatus* (Hazarika et al. 2024). The SEM observation on external structures such as dermal pore patterns, ostioles, vulva, claw digitules, cerarii, different types of wax pore, body shape, number of antenna segments, multilocular pore, trilocular pore, posterior legs, the existence of translucent pores, circulus, oral rim tubular ducts and anal lobe bar of these insect in this study are strongly supported by previous reports (Kumar et al. 1997; Sirisena et al. 2015, Hazarika et al. 2024) and support the current systematic position of *Maconellicoccus hirsutus, P. solenopsis* and *Paracoccus marginatus* as well. Chatzidimitriou et al. (2016) reclassified *P. solenopsis* and *P. solani* as separate species using DNA barcoding. Ahmed et al. (2015) analyzed the *COI* gene to characterize *Phenacoccus* species collected from China, Pakistan, India, and the USA. Their findings grouped *Phenacoccus* spp. into two clusters: the USA group and the Asia group. Phylogenetic analysis based on *COI, 18S*, and *28S* genes showed that UPM *P. solenopsis* is closely related to the same species from Asian countries (China, Pakistan, India, Turkey, Iraq, and Egypt) with high bootstrap support.

Maconellicoccus hirsutus is a destructive polyphagous pest in Southern Asia (Williams 2004), reported on *H. rosa-sinensis* in Kuala Lumpur in 1923, 1932, 1980, and 2016 (Sartiami et al. 2016; Williams 2004). It feeds on over 200 plant genera across 73 families (EFSA Panel on Plant Health et al. 2022). UPM *M. hirsutus* matched descriptions by Williams & Granara de Willink (1992) and OEPP/EPPO (2006). The wingless adult female has an orange-pink to reddish body, sparsely covered with white mealy wax (Williams 1996). SEM analysis confirmed its resemblance to previous studies (Williams & Granara de Willink 1992; OEPP/EPPO 2006; Sirisena et al. 2015). Due to its similarity to other mealybug species, translucent traits are crucial for identification. Preserved in 80% alcohol, UPM *M. hirsutus* remained brownish, while UPM *P. marginatus* turned black, aligning with previous findings

by Williams and Granara de Willink (1992) and OEPP/EPPO (2006). BLASTn (NCBI) and ML phylogenetic analysis confirmed UPM *M. hirsutus* formed a monophyletic clade with specimens from the USA, Egypt, Vietnam, China, Thailand, and Nigeria, with 94% bootstrap support. *Paracoccus marginatus* was first reported in 2009 on cassava, eggplant, jatropha, and hibiscus in Seri Kembangan, Selangor, near the UPM campus (Mastoi et al. 2011). The morphological features of UPM *P. marginatus* matched those described by Williams and Granara de Willink (1992), Miller and Miller (2002), and Wu et al. (2014). Specimens turned black in 80% alcohol, consistent with Wu et al. (2014). The presence of stout fleshy setae on the antennae and translucent pores on the coxa of the hind leg distinguished *P. marginatus* from related species. DNA barcoding, BLASTn results, and phylogenetic analysis confirmed its close relation to *P. marginatus* reported in the USA, Kenya, Guam, China, and India.

Identification of insect species based on *18S rDNA, 28S*, and *COI* genes has been reported by many researchers (Boyer et al. 2011; Liu et al. 2011; McDonnell et al. 2011). The COI region has sites that are evolving at a very fast rate and tend to saturate more quickly, resulting in a small but significant variation in DNA mitochondrial sequences between species (Kondo et al. 2008; Lin & Danforth 2004). On the contrary, the nuclear genes are needed for discrimination between species when two groups of organisms have diverged very recently, may share the same DNA barcode(s), but may not belong to the same species (Hebert et al. 2003; Kaila & Ståhls 2006; Langhoff et al. 2009; Zurovcova et al. 2010). From the phylogeny trees presented in this study, all nodes in the phylogenetic trees were well supported with strong bootstrap values, and similar results were obtained when comparing the phylogenetic relationship of the *18S* ribosomal gene, *28S* ribosomal gene, and *COI* gene. UPM *P. marginatus* and UPM *M. hirsutus* reflected close identity with that of the USA specimen for all genes tested, whereas *P. solenopsis* was closely related to that of Asian specimens. The present findings are in agreement with Kumar et al. (2001), who reported that gene flow between populations was independent of geographic distance except for *P. solenopsis*. The invasive populations might come from the same geographic region in Malaysia from a single introduced population. Molecular characterization using *18S*, *28S* (nuclear genes) and *COI* (mitochondrial) genes, along with ultra-structural study of UPM voucher specimens associated with *Hibiscus rosa-sinensis,* has successfully confirmed three mealybug species, *Par. marginatus, M. hirsutus* and *Ph. solenopsis*. The DNA barcoding approach has further accomplished a more accurate identification of the voucher specimens in this study. This DNA barcoding approach, along with ultra-structural characterization, could be a new approach for more accurate identification of mealybugs associated with *Hibiscus rosa-sinensis* in UPM that can help with decision-making in pest management programs.

AUTHOR'S CONTRIBUTION

The authors confirm their contribution to the paper as follows: S. Ahmmed: collection of the specimens, execution of experiments, design of work, analysis, reviewing and editing the manuscript, interpretation of results, and writing the manuscript. W.H. Lau: analysis, reviewing, editing, and interpretation of results; U.R. Sinnah: review and editing of the manuscript; N.A. Adam: review and editing of the manuscript. All authors read and approved the final version of the manuscript.

FUNDING

This research was supported by the National Agricultural Technology Program-Phase II Project (NATP-2), BARC Component, Bangladesh Agricultural Research Council.

AVAILABILITY OF DATA AND MATERIAL

The specimens listed in this study are deposited in the Laboratory of Insect Pathology (LIP), Department of Plant Protection, Universiti Putra Malaysia (UPM), and are available from the curator upon request. All sequences were made available in the National Center for Biotechnology Information (NCBI)

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This study only included arthropod material, and all required ethical guidelines for the treatment and use of animals were strictly adhered to in accordance with international, national, and institutional regulations. No human participants were involved in any studies conducted by the authors for this article.

CONSENT FOR PUBLICATION

Not applicable.

CONFLICT OF INTERESTS

The authors declare that there is no conflict of interest regarding the publication of this paper.

GENERATIVE AI STATEMENT

The authors declare that no generative AI tools were used in the preparation of this manuscript.

ACKNOWLEDGMENTS

The authors would like to acknowledge the Department of Plant Protection, Universiti Putra Malaysia, for providing the research facilities and support during the study. We are grateful also to the anonymous reviewers and the subject Editor for their critical comments and suggestions.

REFERENCES

Abbas, G., Arif, M.J., Saeed, S. & Karar, H. (2009) A new invasive species of genus *Phenacoccus* Cockerell attacking cotton in Pakistan. *International Journal of Agriculture and Biology*, 11 (1), 54–58.

Ahmed, M.Z., Ma, J.U.N., Qiu, B., He, R., Wu, M., Liang, F.A.N., Zhao, J., Lin, L.I., Hu, X., Lv, L., Breinholt, J.W. & Lu, Y. (2015) Genetic record for a recent invasion of *Phenacoccus solenopsis* (Hemiptera: Pseudococcidae) in Asia. *Environmental Entomology*, 44 (3), 907–918. https://doi.org/10.1093/ee/nvv034

Akintola, A.J. & Ande, A.T. (2008) First record of *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) on *Hibiscus rosa-sinensis* in Nigeria. *Agricultural Journal*, 3 (1), 1–3.

Ashfaq, M., Noor, A.R. & Mansoor, S. (2010) DNA-based characterization of an invasive mealybug (Hemiptera: Pseudococcidae) species damaging cotton in Pakistan. *Applied Entomology and Zoology*, 45 (3), 395–404. https://doi.org/10.1303/aez.2010.395

Beltrà, A., Soto, A. & Malausa, T. (2012) Molecular and morphological characterisation of Pseudococcidae surveyed on crops and ornamental plants in Spain. *Bulletin of Entomological Research*, 102 (2), 165–172. https://doi.org/10.1017/S0007485311000514

Bisevac, L. (1997) A new method for mounting thrips (Thysanoptera) on slides. *Australian Journal of Entomology*, 36 (3), 220. https://doi.org/10.1111/j.1440-6055.1997.tb01457.x

Boyer, S., Blakemore, R.J. & Wratten, S.D. (2011) An integrative taxonomic approach to the identification of three new New Zealand endemic earthworm species (Acanthodrilidae, Octochaetidae: Oligochaeta). *Zootaxa*, 2994 (1), 21–32. https://doi.org/10.11646/zootaxa.2994.1.2

Charles, J.G., Froud, K.J. & Henderson, R.C. (2000) Morphological variation and mating compatibility within the mealybugs *Pseudococcus calceolariae* and *P. similans* (Hemiptera: Pseudococcidae), and a new synonymy. *Systematic Entomology*, 25, 285–294. https://doi.org/10.1046/j.1365-3113.2000.00104.x

Chatzidimitriou, E., Simonato, M., Watson, G.W., Martinez-Sañudo, I., Tanaka, H., Zhao, J. & Pellizzari, G. (2016) Are *Phenacoccus solani* Ferris and *P. defectus* Ferris (Hemiptera: Pseudococcidae) distinct species. *Zootaxa*, 4093 (4), 539–551. https://doi.org/10.11646/zootaxa.4093.4.5

Correa, M., Aguirre, C., Germain, J.-F., Hinrichsen, P., Zaviezo, T., Malausa, T. & Prado, E. (2011) A new species of *Pseudococcus* (Hemiptera: Pseudococcidae) belonging to the "*Pseudococcus maritimus*" complex from Chile: molecular and morphological description. *Zootaxa*, 2926 (1), 46–54. https://doi.org/10.11646/zootaxa.2926.1.2

Cox, J.M. & Pearce, M.J. (1983) Wax produced by dermal pores in three species of mealybug (Homoptera: Pseudococcidae). *International Journal of Insect Morphology and Embryology*, 12 (4), 235–248. https://doi.org/10.1016/0020-7322(83)90020-X

Downie, D.A. & Gullan, P.J. (2004) Phylogenetic analysis of mealybugs (Hemiptera: Coccoidea: Pseudococcidae) based on DNA sequences from three nuclear genes, and a review of the higher classification. *Systematic Entomology*, 29 (2), 238–260. https://doi.org/10.1111/j.0307-6970.2004.00241.x

EFSA Panel on Plant Health (PLH), Bragard, C., Baptista, P., Chatzivassiliou, E., Di Serio, F., Gonthier, P., Jaques Miret, J.A., Justesen, A.F., Sven Magnusson, C., Milonas, P., Navas-Cortes, J.A., Parnell, S., Potting, R., Reignault, P.L., Stefani, E., Thulke, H.H., Van der Werf, W., Civera, A.V., Yuen, J., Zappalà, L., Gregoire, J.C., Malumphy, C., Antonatos, S., Kertesz, V., Maiorano, A., Papachristos, D. & MacLeod, A. (2022) Pest categorisation of *Maconellicoccus hirsutus*. *EFSA journal. European Food Safety Authority*, 20 (1), e07024. https://doi.org/10.2903/j.efsa.2022.7024

Felsenstein, J. (1985) Confidence Limits on Phylogenies: An Approach Using the Bootstrap. *Evolution*, 39 (4), 783. https://doi.org/10.2307/2408678

García Morales, M., Denno, B.D., Miller, D.R., Miller, G.L., Ben-Dov, Y., & Hardy, N.B. (2016) ScaleNet: a literature-based model of scale insect biology and systematics. *Database*, 2016, bav118. https://doi.org/10.1093/database/bav118

Hajibabaei, M., Janzen, D.H., Burns, J.M., Hallwachs, W. & Hebert, P.D.N. (2006) DNA barcodes distinguish species of tropical Lepidoptera. *Proceedings of the National Academy of Sciences*, 103 (4), 968–971. https://doi.org/10.1073/pnas.0510466103

Hall, T.A. (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series*, 41, 95–98.

Hardy, N.B., Gullan, P.J. & Hodgson, C.J. (2008) A subfamily level classification of mealybugs (Hemiptera: Pseudococcidae) based on integrated molecular and morphological data. *Systematic Entomology*, 33 (1), 51–71. https://doi.org/10.1111/j.1365-3113.2007.00408.x

Hazarika, M., Dutta, S.K., Bhattacharryya, B. & Dey, D. (2024) Scanning electron microscopy based morphology of papaya mealybug [*Paracoccus marginatus* Williams and Granara De Wilink (Hemiptera: Pseudococcidae)] on Mulberry plant of Assam, India. *Journal of Advances in Biology & Biotechnology*, 27 (10), 587–595. https://doi.org/10.9734/jabb/2024/v27i101481

Hebert, P.D.N., Cywinska, A., Ball, S.L. & DeWaard, J.R. (2003) Biological identifications through DNA barcodes. P*roceedings of the Royal Society of London*. Series B: Biological Sciences, 270 (1512), 313–321. https://doi.org/10.1098/rspb.2002.2218

Hodgson, C., Abbas, G., Arif, M.J., Saeed, S. & Karar, H. (2008) *Phenacoccus solenopsis* Tinsley (Sternorrhyncha: Coccoidea: Pseudococcidae), an invasive mealybug damaging cotton in Pakistan and India, with a discussion on seasonal morphological variation. *Zootaxa*, 191, 31–35. https://doi.org/10.11646/zootaxa.1913.1.1

Jhala, R.C., Bharpoda, T.M. & Patel, M.G. (2008) *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae), the mealybug species recorded first time on cotton and its alternate host plants in Gujarat, India. *Uttar Pradesh Journal of Zoology*, 28 (3), 403–406.

Kaila, L. & Ståhls, G. (2006) DNA barcodes: Evaluating the potential of COI to differentiate closely related species of *Elachista* (Lepidoptera: Gelechioidea: Elachistidae) from Australia. *Zootaxa*, 1170 (1), 1–26. https://doi.org/10.11646/zootaxa.1170.1.1

Kondo, T., Gullan, P.J. & Williams, D.J. (2008) Coccidology. The study of scale insects (Hemiptera: Sternorrhyncha: Coccoidea). *Ciencia y Tecnología Agropecuaria*, 9 (2), 55–61. https://doi.org/10.21930/rcta.vol9_num2_art:118

Kumar, L.S., Sawant, A.S., Gupta, V.S. & Ranjekar, P.K. (2001) Genetic variation in Indian populations of Scirpophaga incertulas as revealed by RAPD-PCR analysis. *Biochemical Genetics*, 39, 43–57. https://doi.org/10.1023/A:1002797219182

Kumar, S., Stecher, G., Li, M., Knyaz, C. & Tamura, K. (2018) MEGA X: Molecular evolutionary genetics analysis across computing platforms. *Molecular Biology and Evolution*, 35 (6), 1547–1549. https://doi.org/10.1093/molbev/msy096

Kumar, V., Tewari, S.K. & Datta, R.K. (1997) Dermal pores and wax secretion in mealybug *Maconellicoccus hirsutus* (Hemiptera, Pseudococcidae). A pest of mulberry. *Italian Journal of Zoology*, 64 (4), 307–311. https://doi.org/10.1080/11250009709356218

Langhoff, P., Authier, A., Buckley, T.R., Dugdale, J.S., Rodrigo, A. & Newcomb, R.D. (2009) DNA barcoding of the endemic New Zealand leafroller moth genera, *Ctenopseustis* and *Planotortrix*. *Molecular Ecology Resources*, 9 (3), 691–698. https://doi.org/10.1111/j.1755-0998.2009.02537.x

Li, H.-M., Deng, R.-Q., Wang, J.-W., Chen, Z.-Y., Jia, F.-L. & Wang, X.-Z. (2005) A preliminary phylogeny of the Pentatomomorpha (Hemiptera: Heteroptera) based on nuclear 18S rDNA and mitochondrial DNA sequences. *Molecular Phylogenetics and Evolution*, 37 (2), 313–326. https://doi.org/10.1016/j.ympev.2005.07.013

Lin, C.-P. & Danforth, B.N. (2004) How do insect nuclear and mitochondrial gene substitution patterns differ? Insights from Bayesian analyses of combined datasets. *Molecular Phylogenetics and Evolution*, 30(3), 686-702. https://doi.org/10.1016/S1055-7903(03)00241-0

Linares, M.C., Soto-Calderón, I.D., Lees, D.C. & Anthony, N.M. (2009) High mitochondrial diversity in geographically widespread butterflies of Madagascar: a test of the DNA barcoding approach. *Molecular Phylogenetics and Evolution*, 50 (3), 485–495. https://doi.org/10.1016/j.ympev.2008.11.008

Liu, J., Li, Q., Kong, L., Yu, H. & Zheng, X. (2011) Identifying the true oysters (Bivalvia: Ostreidae) with mitochondrial phylogeny and distance based DNA barcoding. *Molecular Ecology Resources*, 11 (5), 820–830. https://doi.org/10.1111/j.1755-0998.2011.03025.x

Magdalita, P.M. & Pimentel, R.B. (2010) *Hibiscus Hybrids and Philippines' Women Achievers*. National Academy of Science and Technology, Bicutan, Taguig City, Metro Manila. 58 pp.

Malausa, T., Fenis, A., Warot, S., Germain, J.-F., Ris, N., Prado, E., Botton, M., Vanlerberghe-Masutti, F., Sforza, R. & Cruaud, C. (2011) DNA markers to disentangle complexes of cryptic taxa in mealybugs (Hemiptera: Pseudococcidae). *Journal of Applied Entomology*, 135 (1–2), 142–155. https://doi.org/10.1111/j.1439-0418.2009.01495.x

Mastoi, M.I., Nur Azura, A., Muhammad, R., Idris, A.B. & Ibrahim, Y. (2011) First report of papaya mealybug *Paracoccus marginatus* (Hemiptera: Pseudococcidae) from Malaysia. *Australian Journal of Basic and Applied Sciences*, 5 (7), 1247–1250.

McDonnell, R.J., Rugman-Jones, P., Backeljau, T., Breugelmans, K., Jordaens, K., Stouthamer, R., Paine, T. & Gormally, M. (2011) Molecular identification of the exotic slug *Arion subfuscus* sensu stricto (Gastropoda: Pulmonata) in California, with comments on the source location of introduced populations. *Biological Invasions*, 13, 61–66. https://doi.org/10.1007/s10530-010-9789-5

Miller, D.R. & Miller, G.L. (2002) Redescription of *Paracoccus marginatus* Williams and Granara de Willink (Hemiptera: Coccoidea: Pseudococcidae), including descriptions of the immature stages and adult male. *Proceedings of the Entomological Society of Washington*, 104 (1), 1–23.

Miller, D.R., Rung, A. & Parikh, G. (2014) Scale Insects, edition 2, a tool for the identification of potential pest scales at USA ports-of-entry (Hemiptera, Sternorrhyncha, Coccoidea). *ZooKeys* 431, 61–67. https://doi.org/10.3897/zookeys.431.7474

Mohammed, M.A., Aman-Zuki, A., Buang, M.G., Hamdan, N.H., Sani, M., King, P.J.H. & Yaakop, S. (2022) DNA barcode of mealybugs (Hemiptera: pseudococcidae) and ants species (Hymenoptera: formicidae) that associated with pineapple in Bintulu, Sarawak, Malaysia. *Serangga*, 27 (2), 38–53.

OEPP/EPPO. (2006) *Maconellicoccus hirsutus*. *EPPO Bulletin*, 36 (1), 167–169. https://doi.org/10.1111/j.1365-2338.2006.00921.x

Pacheco Da Silva, V.C., Bertin, A., Blin, A., Germain, J.F., Bernardi, D., Rignol, G., Botton, M. & Malausa, T. (2014) Molecular and morphological identification of mealybug species (Hemiptera: Pseudococcidae) in Brazilian vineyards. *PLoS ONE*, 9 (7), 1–13. https://doi.org/10.1371/journal.pone.0103267

Palma-Jimenez, M. & Blanco-Meneses, M. (2016) First study about morphological and molecular identification of *Pseudococcus elisae* (Hemiptera: Pseudococcidae) in Costa Rica. *International Journal of Current Research*, 8 (5), 31791–31800.

Ricupero, M., Biondi, A., Russo, A., Zappal, L. & Mazzeo, G. (2021) The cotton mealybug is spreading along the Mediterranean: First pest detection in Italian tomatoes. *Insects*, 12 (8), 675. https://doi.org/10.3390/insects12080675

Rugman-Jones, P.F., Morse, J.G. & Stouthamer, R. (2009) Rapid molecular identification of armored scale insects (Hemiptera: Diaspididae) on Mexican 'Hass' avocado. *Journal of Economic Entomology*, 102 (5), 1948–1953. https://doi.org/10.1603/029.102.0527

Rung, A., Scheffer, S.J., Evans, G. & Miller, D. (2008) Molecular identification of two closely related species of mealybugs of the genus *Planococcus* (Homoptera: Pseudococcidae). *Annals of the Entomological Society of America*, 101 (3), 525–532. https://doi.org/10.1603/0013-8746(2008)101[525:MIOTCR]2.0.CO;2

Sartiami, D., Watson, G.W., Mohamad Roff, M. & Idris, A.B. (2016) Mealybugs (Hemiptera: Coccomorpha: Pseudococcidae) attacking *Hibiscus rosa-sinensis* L. in Malaysia, with two new country records. *AIP Conference Proceedings*. April 13–14, 2016. Faculty of Science and Technology, Universiti Kebangsaan Malaysia; 1784, 060007 pp. https://doi.org/10.1063/1.4966845

Sarvananda, L. (2018) Short introduction of DNA barcoding. International Journal of Research, 5 (4), 673–685.

Simon, C., Frati, F., Beckenbach, A., Crespi, B., Liu, H. & Flook, P. (1994) Evolution, weighting, and phylogenetic utility of mitochondrial gene sequences and a compilation of conserved polymerase chain reaction primers. *Annals of the Entomological Society of America*, 87 (6), 651–701. https://doi.org/10.1093/aesa/87.6.651

Singh, B.B. & Singh, R. (2014) Major rice insect pests in Northeastern UP. *International Journal of Life Sciences, Biotechnology and Pharma Research*, 1 (3), 124–143.

Sirisena, U., Watson, G.W., Hemachandra, K.S., Sage, O. & Wijayagunasekara, H.N.P. (2015) Scanning electron microscopy of six selected mealybug (Hemiptera: Pseudococcidae) species of Sri Lanka. *Tropical Agricultural Research*, 26 (2), 237–247. https://doi.org/10.4038/tar.v26i2.8088

Tamura, K. & Nei, M. (1993) Estimation of the number of nucleotide substitutions in the control region of mitochondrial DNA in humans and chimpanzees. *Molecular Biology and Evolution*, 10 (3), 512–526.

Williams, D.J. (1996) A brief account of the hibiscus mealybug *Maconellicoccus hirsutus* (Hemiptera: Pseudococcidae), a pest of agriculture and horticulture, with descriptions of two related species from southern Asia. *Bulletin of Entomological Research*, 86 (5), 617–628. https://doi.org/10.1017/S0007485300039420

Williams, D.J. (2004) Mealybugs of southern Asia. The Natural History Museum & Southdene, Kuala Lumpur. 896 pp.

Williams, D.J. & Granara, de W. (1992) *Mealybugs of Central and South America*. CAB International, Wallingford. 635 pp.

Wu, F., Liu, Z., Shen, H., Yu, F., Ma, J., Hu, X. & Zeng, L. (2014) Morphological and molecular identification of *Paracoccus marginatus* (Hemiptera: Pseudococcidae) in Yunnan, China. *Florida Entomologist*, 97 (4), 1469–1473. https://doi.org/10.1653/024.097.0422

Zhang, Z.S., Qi, M., Guo, S. & Ma, X. (1988) Study on wax secretion capacity of the white wax insect (*Ericerus pela* Chavannes) in China. *Zoological Research*, 9 (01), 93–97.

Zurovcova, M., Havelka, J.A.N., Starý, P., Vechtova, P., Chundelova, D., Jarosova, A. & Kucerova, L. (2010) " DNA barcoding" is of limited value for identifying adelgids (Hemiptera: Adelgidae) but supports traditional morphological taxonomy. *European Journal of Entomology*, 107 (2), 147. https://doi.org/10.14411/eje.2010.020

تشخیص ریخت‌شناختی و مولکولی شپشک‌های آردآلود (Hemiptera: Coccomorpha: Pseudococcidae) مرتبط با ختمی چینی *Hibiscus rosa-sinensis* (Malvaceae)

سلطان احمد [۱،۳]، وی هونگ لاو [۱]، نور آزورا آدام [۱]، اومارانی سینیا [۲]

۱ گروه گیاهپزشکی، دانشکده کشاورزی، دانشگاه پوترا مالزی، سلانگور، مالزی.
۲ گروه علوم زراعی، دانشکده کشاورزی، دانشگاه پوترا مالزی، سلانگور، مالزی.
۳ گروه حشره‌شناسی، موسسه تحقیقات کنف، بنگلادش.

نویسندگان مسئول: وی هونگ لاو ✉ lauweih@upm.edu.my | سلطان احمد ✉ sultanbjri1984@gmail.com

ویراستار علمی
Mehmet Bora Kaydan

دریافت: ۰۸ بهمن ۱۴۰۴
ویرایش: ۱۹ خرداد ۱۴۰۵
پذیرش: ۲۰ خرداد ۱۴۰۵
انتشار: ۲۶ خرداد ۱۴۰۵

چکیـده: پیچیدگی شناسایی شپشک‌های آردآلود ممکن است مدیریت مؤثر این آفات را به خطر بیندازد. گونه‌های ختمی از خانواده Malvaceae به طور مکرر توسط گونه‌های مختلف شپشک آردآلود در سطح جهانی مورد حمله قرار می‌گیرند. با این حال، اطلاعات اندکی در مورد تنوع شپشک‌های آردآلود مرتبط با ختمی چینی، *H. rosa-sinensis* در مالزی وجود دارد. هدف از این مطالعه، به‌کارگیری روش‌های ریخت‌شناسی و بارکدینگ DNA برای شناسایی گونه‌های شپشک آردآلود که به *H. rosa-sinensis* حمله می‌کنند، می‌باشد. مطالعه بارکدینگ DNA با مقایسه توالی ناحیه بارکد جهانی DNA، زیرواحد I سیتوکروم c اکسیداز (COI) و DNA ریبوزومی (28S, 18S) انجام شد. توالی‌های نوکلئوتیدی گونه‌های شپشک آردآلود را تأیید کردند که نزدیک‌ترین تطابق BLAST در جستجوهای GenBank بالای ۹۷٪ پشتیبانی داشت. بر اساس خصوصیات تاکسونومیک افتراقی مشخص شده در تصاویر میکروسکوپ الکترونی و بارکدینگ DNA، سه گونه شپشک آردآلود شامل *Paracoccus marginatus*، *Maconellicoccus hirsutus* و *Phenacoccus solenopsis* در این مطالعه تأیید شدند. بارکدینگ DNA به همراه تحلیل دقیق فوق‌ساختارهای خارجی، می‌تواند بُعد جدیدی برای شناسایی سریع‌تر و دقیق‌تر گونه‌های شپشک آردآلود ایجاد کند. در نهایت، این مطالعه در تصمیم‌گیری‌های مربوط به مدیریت شپشک‌های آردآلود کمک خواهد کرد.

واژگـان کلیدی: بارکدینگ، شناسایی، ریخت‌شناسی، شپشک آردآلود، مدیریت آفات، فوق‌ساختار