

The Diversity of Subterranean *Dorylus* Ants in the Agricultural Regions of Northern Thailand

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ABSTRACT. – Thailand, primarily an agricultural nation, faces significant crop yield losses due to various pests. Members of the *Dorylus* Fabricius, 1793 ant genus are significant pests of both vegetables and cash crops. Despite extensive research on ant taxonomy in Thailand, a comprehensive understanding of ant diversity remains incomplete, with many species are undiscovered. This study examines the diversity of subterranean *Dorylus* ants in the agricultural regions of Northern Thailand, predominantly occupied by hill tribes. Ant surveys were carried out in organic vegetable farms located in the Chiang Mai and Mae Hong Son provinces of Northern Thailand, by using palm oil baits in sieve buckets. The worker ants were identified through their morphological and molecular characters derived from the cytochrome oxidase subunit I (COI) region of the mitochondrial gene. Our findings revealed that two species, *Dorylus laevigatus* and *D. orientalis*, are major *Dorylus* ants that infest Brassicaceae and Fabaceae vegetable crops. This is the first record of these species infesting Chinese radish (*Raphanus sativus* L.). In addition, we present the first documentation of *D. laevigatus* in Pak choi (*Brassica chinensis* L.), and peanut (*Arachis hypogaea* L.) in Thailand. These pests burrow into the subterranean parts of crop roots, resulting in reduced growth yield. The damage manifests as an unhealthy appearance in the above-ground parts of the crops.

KEYWORDS: food security, Chinese radish, pak choi, peanut, *Dorylus* pest

INTRODUCTION

Thailand, predominantly an agricultural nation, agriculture significantly contributes to the country's economic growth (Thepent, 2009; Suwannarat, 2011; Panuwet et al., 2012; Krailertrattanachai et al., 2019). However, the agriculture sector is continually threatened by pests, leading to a heavy reliance on pesticides for crop protection and enhanced land productivity (Jaipieam et al., 2009; Panuwet et al., 2012). This excessive and improper pesticide usage poses substantial health and environmental risks (Praneetvatakul et al., 2013; Tawatsin et al., 2015; Riwthong et al., 2017).

Ants are recognized as the primary urban pests by the structural pest control industry (Field et al., 2007; Gebreyohanes et al., 2007). Army ants, among the most extensively studied ant species, are distinguished by their behaviors of colony migration and mass raiding (Berghoff, 2002). They are prominent arthropod predators, particularly in tropical forests, and play a crucial role in shaping these ecosystems (Hölldobler and Wilson, 1990; Lach et al., 2010; Ohyama et al., 2020). The earliest documentation of

ant fauna in Thailand was reported by Bingham (1903), who identified 39 species in the southern regions of Songkhla, Pattani, and Narathiwat. Jaitrong and Nabhitabhata (2005) expanded this knowledge by reporting 247 ant species across 55 genera and 9 subfamilies in Thailand. More recently, Khachonpisitsak et al. (2020) provided an updated checklist, recording 529 valid species and subspecies within 109 genera across ten subfamilies of Thai ant fauna. Of these, 81 species are native to Thailand, with 20 species endemic and 14 exotic (Deyrup et al., 2000; Wetterer and Vargo, 2003; Heterick and Shattuck, 2011; Vonshak and Ionescu-Hirsch, 2009; Wetterer, 2009; 2010a; 2010b; 2010c; Yamane and Jaitrong 2011; Wetterer 2013; Wetterer et al., 2015). Additionally, more than 200 species remain unidentified, including several potentially new to science (Khachonpisitsak et al., 2020). Despite these advancements, information on ants in Thailand remains dispersed and not comprehensively compiled.

The army ant genus *Dorylus*, belonging to the subfamily Dorylinae within the order Hymenoptera, is widely distributed, with members recorded from Sub-Saharan Africa through North Africa to Asia,

predominantly in the Afrotropics (Niu et al., 2010; Borowiec, 2016). The worker ants of *Dorylus* are identifiable by several distinct features: a well-developed promesonotal suture, a propodeal spiracle positioned high on the propodeum without propodeal lobes, a single waist segment, a large pygidium with a flattened surface featuring two cuticular projections, and simple pretarsal claws (Borowiec, 2016). Additionally, the males of this genus are characterized by flattened femora, which are noticeably broader and more compressed compared to the tibiae and tarsi (Borowiec, 2016). *Dorylus* ants, recognized for their sharp cutting mandibles and large colonies, are known predators of vertebrates (Gotwald, 1982), they primarily hunt and nest in soil, rarely surfacing (Schöning et al., 2005). *Dorylus* species are categorized based on their foraging habits, including subterranean, leaf litter, and surface foragers (Schöning and Moffett, 2007). The genus is often noted for its yellowish-brown *Dorylus* and is frequently studied due to its conspicuous appearance and predatory behavior on other arthropods in the Afrotropics (Borowiec, 2016). In Thailand, three subterranean ant species within the *Dorylus* genus have been documented: *D. laevigatus* (Smith, 1957), *D. orientalis* Westwood, 1835, and *D. vishnui* Wheeler, 1913. *D. orientalis*, a common pest, is found in the northern, northeastern, and central regions of Thailand, while *D. vishnui* is reported in the eastern and southern parts. *D. laevigatus*, on the other hand, has a nationwide presence (Jaitrong et al., 2011). Sakchoowong et al. (2008) identified a species of Dorylinae in forests and agricultural lands in Northern Thailand, though its precise species-level identification remains unclear. *Dorylus* ants are frequently observed in both primary and secondary forests, as well as agricultural areas across Thailand (Jaitrong et al., 2011).

Previous researches indicate that *Dorylus* pest species are most active from December to April, with their population growth favored by dry conditions and high temperatures (Hussain et al., 2019; Saikia and Nath, 2017). These ants primarily damage plant stems and tubers by burrowing holes (Hussain et al., 2019). A notable example is *D. orientalis*, a red soil ant, which significantly impacts economically vital vegetable and cash crops such as coconut palm, citrus, and sugarcane (Roonwal, 1976; Dash et al., 2013; Borkakati et al., 2020). Native to various Asian countries, *D. orientalis* is typically managed through soil pesticide applications (Dash et al., 2013; Riaz et al., 2019). Morphologically, it features nine-segmented antennae, an absence of eyes, a rectangular head, deeply emarginate occiput, an elongated thorax constricted at the pro-mesonotal suture, short robust legs, and a single-node pedicel

(Korgaonkar et al., 2020). Another pest species, *D. laevigatus* from Malaysia, exhibits a hypogenic lifestyle and is less extensively studied than *D. orientalis* (Berghoff, 2002). Despite its local abundance, the biology of *D. laevigatus* remains largely unexplored.

In this study, we aim to explore the diversity of subterranean *Dorylus* ants in the agricultural areas of Northern Thailand and to classify their species using morphological and molecular traits. Ant sampling was carried out using palm oil baits in sieve buckets across five districts in two provinces of Northern Thailand.

MATERIALS AND METHODS

Study Area and Surveying of Ants (Fig.1)

The study was carried out across five organic farms, including a pak choi farm in Chom Thong district and a Chinese radish farm in Mae Wang district of Chiang Mai province, as well as three peanut farms in the Khun Yuam, Muang, and Pang Ma Pha districts of Mae Hong Son province. Sampling took place from March to October in 2018 (Fig. 1A and G). In each farm, 15 traps were randomly installed, each consisting of a palm oil bait in a sieve bucket, following the methodologies of Weissflog et al. (2000) and Berghoff et al. (2002a). To set up each trap, a 30 cm deep hole was dug, where a sieve bucket containing the bait was placed. Each trap used 80 ml of palm oil as bait to attract subterranean ants. Then the bucket is filled with soil (Fig. 1L). The traps were checked within five days of installation, and ants attracted to the trap (Fig. 1H and L) were collected.

Morphological Analysis

Specimens collected from field surveys were pinned on triangular cards and photographed using a ZEISS Axiocam 305 color Microscope Camera mounted on a ZEISS Stemi 508 Stereo Microscope. Identification was carried out using key characters as described in the literature by Wilson (1964), Jaitrong (2011) and Jaitrong et al. (2011). Image processing, including combination and modification, was done using Adobe Photoshop CS6 version 10.0. The preparation of specimens followed the methods outlined by Boudinot et al. (2002) and they were subsequently deposited at the Natural History Museum of the Natural Science Museum in Pathum Thani, Thailand (THNHM).

Molecular Analyses

DNA was extracted from ant specimens using the standard Chelex_100 (BioRad, Hercules, CA) resin protocol. Sequences were successfully obtained from



FIGURE 1. Experimental sites and ant species: (A–G) Experimental sites; (H–L) *Dorylus* ants on soil.

D. laevigatus and *D. orientalis*, utilizing primers under PCR conditions as recommended by Wang and Hua (2020). The DNA sequences generated in this study were then subjected to BLAST searches on the NCBI database (<https://www.ncbi.nlm.nih.gov>) to select appropriate taxa for phylogenetic analysis.

All available COI sequences of the genus *Dorylus* and some related taxa were chosen for analysis and

downloaded from GenBank (Table 1). The COI sequences were unambiguously aligned using MUSCLE algorithm in MEGA X. The final dataset contained a total of 744 positions. The alignment was checked for stop codons by translating it to amino acids using MEGA X (Kumar et al., 2018). Best-fit models of molecular evolution were determined for each codon position under the Bayesian information

TABLE 1. List of the species, locality and GenBank accessions used for molecular phylogenetic analyses.

Voucher number	Species	Locality	GenBank accession no.	References
DLCM-01	<i>Dorylus laevigatus</i>	Ban Muaeng Ang, Chiang Mai province, Thailand	OQ396675	This study
DLMHS-01	<i>Dorylus laevigatus</i>	Khun Yuam, Mae Hong Son province, Thailand	OQ396676	This study
DOCM-01	<i>Dorylus orientalis</i>	Khun Yuam, Mae Hong Son province, Thailand	OQ396677	This study
1	<i>Dorylus wilerthi</i>	Kibale (Uganda)	EF413837	Kronauer et al. (2007)
2	<i>Dorylus terrificus</i>	Kibale (Uganda)	EF413842	Kronauer et al. (2007)
3	<i>Dorylus molestus</i>	Mt. Kenya (Kenya)	EF413809	Kronauer et al. (2007)
4	<i>Dorylus molestus</i>	Mt. Kenya (Kenya)	EF413836	Kronauer et al. (2007)
5	<i>Dorylus burmeisteri</i>	Comoé (Ivory Coast)	EF413808	Kronauer et al. (2007)
6	<i>Dorylus mayri</i>	Bossou (Guinea)	EF413844	Kronauer et al. (2007)
7	<i>Dorylus arcens</i>	Taï (Ivory Coast)	EF413829	Kronauer et al. (2007)
8	<i>Dorylus sjoestedti</i>	Ndoki (DR Congo)	EF413834	Kronauer et al. (2007)
9	<i>Dorylus rubellus</i>	Gashaka (Nigeria)	EF413833	Kronauer et al. (2007)
10	<i>Dorylus emeryi</i>	Taï (Ivory Coast)	EF413810	Kronauer et al. (2007)
11	<i>Dorylus gerstaeckeri</i>	Bossou (Guinea)	EF413812	Kronauer et al. (2007)
12	<i>Dorylus opacus</i>	Kibale (Uganda)	EF413813	Kronauer et al. (2007)
13	<i>Dorylus kohli</i>	Kibale (Uganda)	EF413814	Kronauer et al. (2007)
14	<i>Dorylus nigricans</i>	Taï (Ivory Coast)	EF413841	Kronauer et al. (2007)
18	<i>Dorylus helvolus</i>	Cullinan (South Africa)	EF413832	Kronauer et al. (2007)
19	<i>Dorylus braunsi</i>	Kakamega (Kenya)	EF413835	Kronauer et al. (2007)
21	<i>Dorylus spininodis</i>	Gashaka (Nigeria)	EF413826	Kronauer et al. (2007)
22	<i>Dorylus gribodoi</i>	Taï (Ivory Coast)	EF413817	Kronauer et al. (2007)
23	<i>Dorylus affinis</i>	Mt. Kenya (Kenya)	EF413846	Kronauer et al. (2007)
27	<i>Dorylus laevipodex</i>	Mt. Kenya (Kenya)	EF413840	Kronauer et al. (2007)
29	<i>Dorylus laevigatus</i>	Poring (Malaysia)	EF413819	Kronauer et al. (2007)
34	<i>Dorylus conradti</i>	Kakamega (Kenya)	EF413820	Kronauer et al. (2007)
35	<i>Dorylus cf. vishnui</i>	Poring (Malaysia)	EF413821	Kronauer et al. (2007)
BTN17xii08-20	<i>Dorylus orientalis</i>	Binh Chau-Phuoc Buu Nature Reserve, Ba Ria-Vung Tau Province, Vietnam	AB767278	Eguchi et al. (2014)
BTN17xii08-20	<i>Dorylus orientalis</i>	Binh Chau-Phuoc Buu Nature Reserve, Ba Ria-Vung Tau Province, Vietnam	AB767279	Eguchi et al. (2014)
BTN17xii08-20	<i>Dorylus orientalis</i>	Binh Chau-Phuoc Buu Nature Reserve, Ba Ria-Vung Tau Province, Vietnam	AB846958	Eguchi et al. (2014)
Outgroups				
39	<i>Aenictus aratus</i>	Tawau Hills (Malaysia)	EF413824	Kronauer et al. (2007)
40	<i>Aenictus laeviceps</i>	Poring (Malaysia)	EF413823	Kronauer et al. (2007)

criterion using KAKUSAN4 (Tanabe, 2011): HKY85 with gamma distribution (+G) for the first codon position and GTR+G for the second and third positions. Phylogenetic relationships were assessed by Bayesian inference using MrBayes v3.2.7a (Ronquist and Huelsenbeck, 2003) as implemented in the CIPRES Science Gateway (Miller et al., 2010). Searches were performed in two parallel runs with eight chains each for ten million generations, sampled every 100th generation. After the first 25 % of the sampled trees were discarded, the final topologies were consented following the 50 % majority rule. Maximum likelihood (ML) analysis was conducted with IQ-TREE 2.1.2 (Nguyen et al., 2015; Minh et al., 2020), using the option “complete bootstrap” and 1,000 nonparametric replicates. The phylogenetic tree was drawn by FigTree v.1.4.3 (<http://tree.bio.ed.ac.uk/software/figtree>), then edited in Microsoft PowerPoint version 2013 and Adobe Photoshop CS6 version 10.0.

RESULTS AND DISCUSSION

Damages in vegetable crops by ants (Fig. 2)

During our survey in organic vegetable fields, we observed withered Pak choi seedlings (Fig. 2A, B, C, D, and E). The damage caused by ants was evident upon uprooting the plants, revealing a congregation of ants at the roots (Fig. 2D and E). In the case of Chinese radish, the root tubers showed holes attributed to ant activity (Fig. 2F, G and H), leading to reduced productivity. Similarly, in peanut crops, holes were observed on the nut shells, caused by ants (Fig. 2I). The interior of these nuts was often filled with soil, which negatively impacted the yield (Fig. 2J).



FIGURE 2. Damages caused by ant species in vegetable crops: (A–E) Withered Pak choi leaves; (D, E) Root hole made by ants in Pak choi; (F–H) Damaged Chinese reddish; (I) Upper part of the damaged ground nut; (J) Inner surface of the damaged ground nut.

Morphological Analysis and Identification

Taxonomy

Order Hymenoptera Linnaeus, 1758

Family Formicidae Latreille, 1802

Genus *Dorylus* Fabricius, 1793

Type-species.— *Dorylus helvola* (= *Vespa helvola* Linnaeus, 1764)

Dorylus has a long, confusing history. The genus was divided into six subgenera namely *Alaopone*, *Anomma*, *Dichtadia*, *Dorylus* sensu stricto, *Rhogmus*, and *Typhlopone* (Schöning et al., 2005). However, later

molecular study didn't support this subgeneric classification whereas the most speciose subgenera such as *Dorylus*, *Anomma* and *Dorylus s. str.* resulted as polyphyletic (Kronauer et al., 2007). Thus, Borowiec (2016) proposed seven informal groups for the subgenera namely, 1) *Dorylus fimbriatus*-group (equivalent of *Rhogmus*), 2) *Dorylus fulvus*-group (equivalent of *Typhlopone*), 3) *Dorylus helvolus*-group (equivalent of *Dorylus s. str.* but excluding species of politus-group and including two species previously assigned to *Anomma*), 4) *Dorylus laevigatus*-group (equivalent of *Dichthadia*), 5) *Dorylus nigricans*-group (equivalent of *Anomma* excluding *emeryi* and *kohli*), 6) *Dorylus orientalis*-group (equivalent of *Alaopone*) and 7) *Dorylus politus*-group (species excluded from *Dorylus s. str.* based on phylogeny in Kronauer et al. (2007). The workers of *Dorylus* are differ in size continuum whereas large workers differ from small workers by possessing falcate, sharply pointed mandibles which appear to be well suited for piercing skin or cuticle, but less so for other tasks (Braendle et al., 2003).

Dorylus laevigatus (Smith, 1857)

(Fig. 3)

Material examined.— Northern Thailand, Chiang Mai province, Chom Thong district, Ban Muaeng Ang, IV.2018, 124 workers (DLCM-01), P. Suttiprapan leg.; Northern Thailand, Chiang Mai province, Mae Wang district, Ban Thung Luang, VIII.2018, 76 workers (DLCM-02), P. Suttiprapan leg.; Northern Thailand, Mae Hong Son province, Khun Yuam district, Ban Huai Tong Jing, 15 September 2018, 57 workers (DLMHS-02), P. Suttiprapan leg.; Northern Thailand, Mae Hong Son province, Khun Yuam district, 13 August 2018, 62 workers (DLMHS-01), P. Suttiprapan leg. (GenBank numbers DLCM-01: OQ396675; DLMHS-01: OQ396676).

Worker.— Major worker: Head and antennae shiny, not much hairy, top of the head has long hair, reddish brown. Antenna with 12 segments, second segment is longer than 3rd and 5th segments. The last or 12th segment is longer than 9th and 11th segments. Inner edge absents of teeth, inner edge of the mandible sharp, pointed. Petiole waist similar to width and height, from the top view the front is narrower than footer. Thorax has no hair. Minor worker: Head shiny but no hair. Clypeus distantly curve. Antennae with 10 segments, second segment longer than 3rd and 5th segments, 10th segment equal length with 5th and 9th segments. Mandible, sharp, pointed. Inner edge with one tooth. short and fine hair covers the body. *Middle worker:*

moderate size, Inner edge with one tooth. Antenna with 10 to 12 segments (Fig. 3).

Remarks.— *D. laevigatus* has a long history for more than 150 years with worldwide distribution and a single excavated colony of *D. laevigatus* contained about 300,000 workers (Berghoff et al., 2002b). However, colony composition or nesting habits are largely unknown (Berghoff et al., 2002b). Nevertheless, *D. laevigatus* had been abundant in Thailand, its host distribution is unknown (Jaitrong et al. 2011) and this study provides the first report of the species from Chinese radish and peanut. *D. laevigatus* shows hypogaecic lifestyle which makes difficulties to investigate its biology (Berghoff et al. 2002a).

Dorylus orientalis Westwood, 1835

(Fig. 4)

Material examined.— Northern Thailand, Chiang Mai province, Mae Wang district, Ban Thung Luang, 17.IV.2018, 77 workers (DOCM-01), P. Suttiprapan leg.; Northern Thailand, Mae Hong Son province, Muaeng Mae Hong Son district, 14 October 2018, 50 workers (DOMHS-01), P. Suttiprapan leg.; Northern Thailand, Mae Hong Son province, Pang Mapha district, 22 September 2018, 53 workers (DOMHS-02), P. Suttiprapan leg. (GenBank numbers DOCM-01: OQ396677).

Worker.— Head rectangle, narrow, has hair holes, oblong head, longer than wide, distinguishable, reddish brown, for minor worker yellowish brown, head is darker than body, sides are quite straight and parallel to the edge of the cranial ridge. Body shiny skin. Occipital margin concave. Clypeus shorter. Lateroclypeal teeth absent. Tentacle are the same length as or close to 9th segment, the base of the tentacles is very short, more than half of the header length. Antennae with nine segments, apical antennal segment enlarged and longer, broader and longer than two preceding segments combined, antennal scape shorter, second and the rest of the segments are similar in size, last segment same length as segments 5–8, lined up, narrow jaw with pointed tip Inner edge has two large teeth. Mandible narrow, sharp, pointed. Thorax square-shaped, flattened. Propodeal flattened. Petiole is the same length as or close to the height, top is rounded, with the footer slightly higher than the front. Sub petiolar large, semi-triangular in shape, tip pointing down (Fig. 4).

Remarks.— Ek-Amnuay (2010) reported *D. orientalis* on peanuts and water melon from Thailand. The pest also caused 70–90% damage at harvest in potatoes in farmers' fields in Bihar, India (Kishore et al., 1990) and other vegetable crops. The species known to have wide



FIGURE 3. *Dorylus laevigatus* (A) lateral view of major worker; (B) lateral view of minor worker; (C) dorsal view of major worker; (D) dorsal view of minor worker; (E) lateral view of head of major worker (F) dorsal view of head of major worker; (G) dorsal view of head of minor worker; (H) front view of head of major worker; (I) front view of antennae of major worker (J) front view of antennae of minor worker (K) lateral view of thorax of major worker; (L) dorsal view of thorax of major worker; (M) dorsal view of thorax of minor worker; (N) lateral view of gaster of major worker; (O) dorsal view of gaster of major worker; (P) dorsal view of gaster of minor worker.

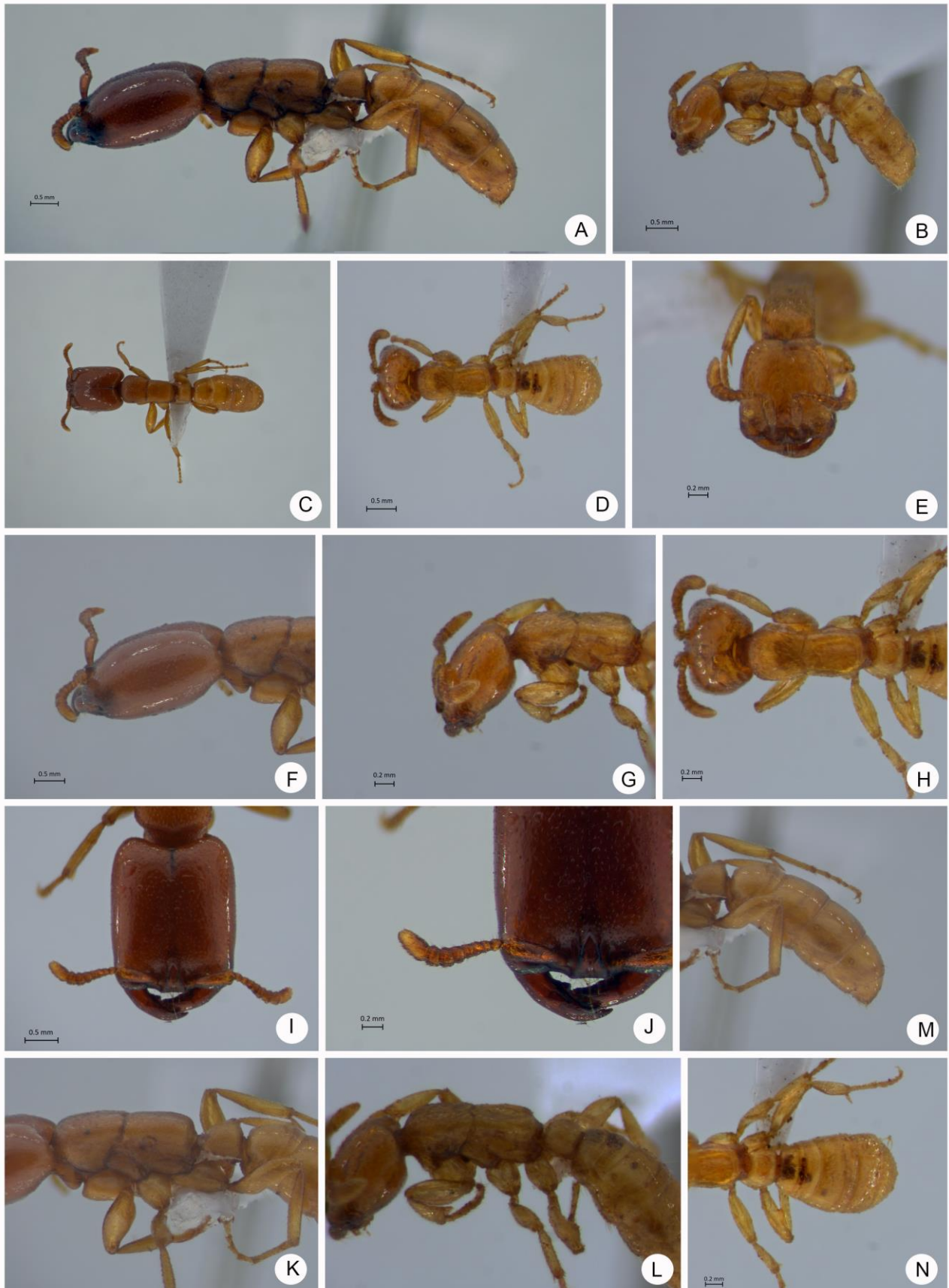


FIGURE 4. *Dorylus orientalis* (A) lateral view of major worker; (B) lateral view of minor worker; (C) dorsal view of major worker; (D) dorsal view of minor worker; (E) front view of head of minor worker (F) dorsal view of head of major worker; (G) dorsal view of head of minor worker; (H) lateral view of head of minor worker; (I) front view of head of major worker (J) front view of antennae of major worker (K) lateral view of thorax of major worker; (L) dorsal view of thorax of minor worker; (M) lateral view of gaster of minor worker; (N) dorsal view of gaster of major worker.

range of food habit especially in agricultural area and no record was available of attacking Chinese radish and is initially reported in the present study. The workers are commonly identified with narrow, sharp, and pointed 9-segmented mandibles. Mukerjee (1930) stated that the mandibles of *D. orientalis* were structured to be carnivorous. The workers of *D. orientalis* have caused damages in February and April in India and May and September in Sri Lanka. They formed tunnels and galleries through the soil to reach the plant parts (Roonwal, 1976) (Fig. 1).

Molecular Analyses

All COI sequences from the genus *Dorylus*, including our newly generated sequences were retrieved as a significantly supported clade by phylogenetic analyses (Supplement materials Table S1; Fig. 5) (1 bpp for BI and 99% bootstrap values for ML). Our two specimens of *D. laevigatus* (DLMHS-01 and DLCM-01) were retrieved as a sister clade to *D. laevigatus* (EF413819) from Malaysia with strong support (1 bpp for BI and 100% bootstrap values for ML). Similarly, the specimen *D. orientalis* (DOCM-01) belongs to the same clade with other *D. orientalis* specimens (AB767278, AB767279, and AB846958) from Vietnam (1 bpp for BI and 98% bootstrap values for ML). *D. cf. vishnui* formed a sister group to *D. orientalis* with strong support (1 bpp for BI and 98% bootstrap values for ML), thus the taxonomic placement of *Dorylus* is compatible with the study of Kronauer et al. (2007). The interspecific divergence of the COI uncorrected p-distance among these 23 *Dorylus* species was found to be rather high (range: 4.82–23.56%, average: 13.49%; Supplement materials Table S1), whereas this analysis demonstrated that the intraspecific divergences of both *D. laevigatus* and *D. orientalis* populations were rather low (2.8% and 1.76 %, respectively). This ensures that the molecular data considerably supports our ability to identify new specimens as belonging to these two species.

The taxonomy of *Dorylus* has a confusing history and the modern sub-generic division of *Dorylus* was stabilized by Emery (1895, 1910). The genus has also been investigated with molecular data in recent decades. However, the most speciose subgenera *Anomma* and *Dorylus sensu stricto* were found to be not monophyletic (Kronauer et al., 2007) including in the present study. The surface and leaf litter-foraging species have been collectively referred to as ‘driver ants’ (Savage, 1847) and are traditionally classified in the subgenus *Anomma* (Borowiec, 2016). Kronauer et al. (2007) estimated the crown group age of *Dorylus* to be between 30 and 64 million years but later study

suggested it to be much younger at about 22 million years (Brady et al., 2014).

The Brassicaceae family comprises a range of economically significant crops cultivated globally (Aires, 2015; Subramanian et al., 2023). These plants are adaptable to various ecological conditions and are among the earliest known cultivated plants (Ahuja et al., 2011; Koyyati et al., 2016). The genus *Brassica*, the largest and most diverse within this family, includes vegetables and is also utilized for producing canola oil and mustard condiments (Jeon et al., 2018). Members of the Brassicaceae family are vulnerable to pests, which predominantly impact growth and crop yield (Ahuja et al., 2011; Fourie et al., 2016; Zalucki et al., 2021). Ant species such as bigheaded ants, carpenter ants, *Camponotus compressus* (Fabricius), *Pheidole* sp. (Fabricius), and *Solenopsis invicta* (Boren) have also been observed in Brassicaceae crops (Debbarma et al., 2017).

Chinese radish (*Raphanus sativus* var. *longipinnatus*), an organic root vegetable belonging to the Brassicaceae family, is cultivated globally, especially in Asia (Aires, 2015; Subramanian et al., 2023). Renowned for its nutritional and medicinal values, it is often used to treat hyperlipidemia, coronary heart diseases, and cancer (Curtis, 2003). In this study, we report the presence of both *D. laevigatus* and *D. orientalis* in Chinese radish for the first time.

Another Brassicaceae vegetable, *Brassica rapa* subsp. *chinensis*, known as Chinese cabbage, Bok choy, Pak choy, or Buk choy, is a popular green leafy vegetable in Asia (Jeon et al., 2018; Managa et al., 2020). In Thailand, it is commonly referred to as Pak Choi or Pak-Kad Hong-Te, this vegetable is rich in nutrients, vitamins, minerals, and unique secondary metabolites like glucosinolates, carotenoids, chlorophylls, and phenolic compounds such as flavonoids and hydroxycinnamic acids (Heinze et al., 2018; Managa et al., 2020; Zou et al., 2021). McDonald and Westerveld (2008) noted that the soil-borne protist *Plasmodiophora brassicae* (Woronin) causes damage to Pak choy. This study marks the first record of *D. laevigatus* in Pak choy.

The Fabaceae family, known for its significant ecological potential in nitrogen fixation and soil conservation, also plays a crucial economic role through the production of carbohydrates, colorants, forage, fuel, gums, oils, pesticides, proteins, and timber (Rodriguez-Riano et al., 1999). Peanuts (*Arachis hypogaea* L.), a member of the Fabaceae family, are globally popular due to their nutritional value, taste, and affordability (Okello et al., 2010; Toomer, 2018). While Ek-Amnuay (2010) reported the presence of *D. orientalis* in Thai peanuts, this study provides the first

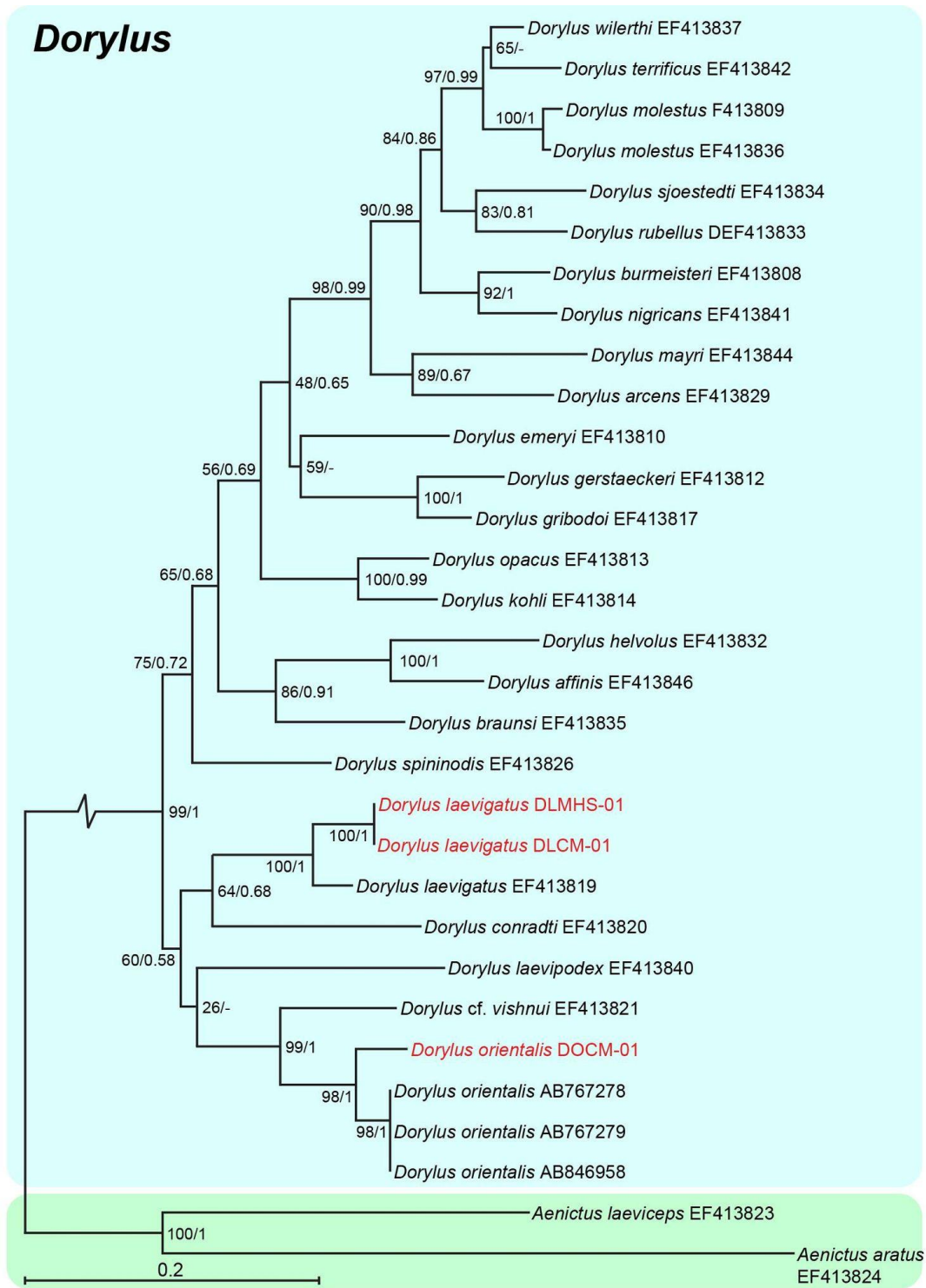


FIGURE 5. Maximum likelihood tree (ML) of subterranean *Dorylus* species and some related taxa based on a total of 744 bp of COI genes. Terminals in red indicate newly generated sequences for this study. Numbers on nodes are bootstrap values (bs) from ML analysis and bipartition posterior probability (bpp) from Bayesian inference analysis (BI) and are shown as ML/BI.

record of *D. laevigatus* being found in peanuts in Thailand.

Vegetable crops infested by *Dorylus* ants revealing severe root damage characterized by holes in the ground cover. Symptoms were also visible in the upper parts of the plants, where affected crops exhibited

reduced growth yield and an unhealthy appearance. Therefore, effective pest management in the field is crucial to control these damages (Pathak and Chilluwal, 2014). Despite Thailand being a biodiversity hotspot, ant research in the country is limited, with data largely scattered (Jaitrong and

Nabhitabhata, 2005; Khachonpisitsak et al., 2020). Our study areas, near natural forests in the mountainous cities of Chiang Mai and Mae Hong Son, are regions where agriculture is widely practiced by the "hill tribes" of Northern Thailand (Sakchoowong et al., 2008). Modern agricultural practices pose threats to natural ecosystems and their native biodiversity (Matson et al., 1997; Foley et al., 2005; Pacheco et al., 2013). The conversion of forest land to farming also affects the abundance and diversity of terrestrial ants (Andersen, 1995; Sakchoowong et al., 2008). Therefore, the species reported in our study may have migrated from nearby natural forests. Previous studies have also reported the presence of both *D. orientalis* and *D. laevigatus* in the Chiang Mai province of Thailand (Jaitrong and Nabhitabhata, 2005; Jaitrong, 2011; Suwannaphak et al., 2016; Khachonpisitsak et al., 2020).

In conclusion, this study highlights the significant impact of *Dorylus* ants, particularly *D. laevigatus* and *D. orientalis*, on agricultural crops in Thailand. We report the first presence of these species in Chinese radish and peanuts, noting their contribution to root damage and reduced yields. This emphasizes the need for effective pest management in Thai agriculture, especially in regions where hill tribes practice farming. Additionally, our findings contribute to the understanding of ant diversity in Thailand's biodiversity hotspots and suggest a shift in ant populations due to changing land use and agricultural practices. Therefore, sustainable agricultural methods are vital for maintaining ecological balance and protecting crop productivity.

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