Systematics

Discovery of a New *Crematogaster* Species with 10-Segmented Antennae From the Indochina Region, With Description of the Species and Its Phylogenetic Position (Hymenoptera: Formicidae)

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Abstract

Here we report, for the first time, the occurrence of a *Crematogaster* species with 10-segmented antennae in the Indochina region. Specifically, the new species, *Crematogaster indosinensis* **sp. nov.**, exhibits size polymorphism in the worker caste and was collected in Cambodia. However, it is not currently known whether *C. indosinensis* has an association with ant-plants. The interspecific cytochrome *c* oxidase subunit I divergence for *C. indosinensis* versus *C. borneensis* was 20%, suggesting that the taxa diverged relatively long ago. Molecular phylogenetic analysis using nuclear genes revealed that *C. indosinensis* is sister to the remainders of the *C. borneensis*-group.

Key words: *Crematogaster borneensis*-group, Indochina region, *Macaranga*, molecular phylogeny, polymorphism

In most species of the ant genus *Crematogaster*, workers have 11-segmented antennae, but some species have 10-segmented antennae (Blaimer 2012c). Members of the *C. borneensis*-group have 10-segmented antennae and are obligately associated with the ant-plant genus *Macaranga* (Euphorbiaceae) (Feldhaar et al. 2016). The group is distributed in Peninsular Malaysia, Borneo, and Sumatra, but has not yet been observed in other regions. The establishment of the *Crematogaster borneensis*-group by Blaimer (2012c) is based on molecular phylogeny and morphological assignment. The *C. borneensis*-group consists of eight species (*C. borneensis* André, *C. captiosa* Forel, *C. claudiae* Feldhaar et al, *C. decamera* Forel, *C. hullettii* Feldhaar et al, *C. linsenmairi* Feldhaar et al, *C. maryatii* Feldhaar et al, and *C. roslihashimi* Feldhaar et al) from Peninsular Malaysia, Borneo, and Sumatra (Blaimer 2012c, Feldhaar et al. 2016). Members of the *C. borneensis*-group have been differentiated based on the following characters of the worker caste: 10-segmented antennae; head shape rectangular, longer than wide; petiole suboval, circular, or hexagonal in dorsal view; postpetiole globular, and a faint posterior impression may be present (Blaimer 2012c).

Traditionally, *Crematogaster* species with 10-segmented antennae were placed in the subgenus *Decacrema* (Emery 1922, Wheeler 1922). This subgenus is distributed in the Old World and is composed of Asian and Afrotropical species. Asian members are known to have mutualistic relationships with ant-plants in the genus *Macaranga* (Euphorbiaceae), while the Afrotropical members are free-living (Blaimer 2010). However, Blaimer (2012d) revealed that the subgenus *Decacrema* is not monophyletic and divided it into Asian and Afrotropical clades in her molecular phylogeny, and then established the *C. borneensis*-group to accommodate the Asian members (Blaimer 2012c).

The *C. borneensis*-group (former subgenus *Decacrema* found in Asia) has long attracted the interest of biologists investigating the interactions between ants and plants (Fiala and Maschwitz 1990; Federle et al. 1997; Fiala et al. 1999; Itioka et al. 2000; Inui et al. 2001; Itino and Itioka 2001; Feldhaar et al. 2003; Itino et al. 2003; Quek et al. 2004, 2007; Ueda et al. 2008, 2010, 2015). The host plant *Macaranga* is widely distributed in the Old World (Whitmore 2008), but obligate ant-plants associated with *Crematogaster* are known only from Peninsular Malaysia, Borneo, and Sumatra (Quek et al. 2004, Blaimer 2012c, Fiala et al. 2016). In Asia, no species in the *C. borneensis*-group have been reported from the mainland of the Indochina region.

During our field surveys in the Indochina region, we collected one colony (only four workers) of *Crematogaster* species with 10-segmented antennae from natural forests of Cambodia. The species is morphologically similar to *C. borneensis* and *C. decamera*,

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but apparently new to science and is described here as a new species. The phylogenetic position will provide insight into the biogeography of the *C. borneensis*-group.

The aim of this study was thus to 1) clarify the phylogenetic position of the *Crematogaster* ants with 10-segmented antennae by using molecular phylogenetic analysis, and 2) describe the taxon as a new species.

Materials and Methods

Observations and Measurements and Indices

Four workers of the *Crematogaster* species (SH10-Cam-46) were examined in this study. Most observations were made on a Leica M205C stereomicroscope. Images were taken using a Canon EOS 50D with a Canon MP-E 65 mm 1–5× Macro lens, then processed using Combine ZM. Measurements were made under a Leica M205C stereomicroscope using micrometers. All measurements are expressed in millimeters, recorded to the second decimal place. The measurements for petiole and postpetiole follow Longino (2003). Head Width (HW): Maximum width of head in full-face view, excluding the eyes Head Length (HL): Perpendicular distance from vertex margin to line tangent anteriormost projections of clypeus in full-face view; Cephalic Index (CI): HW/HL × 100; Scape Length (SL): Length of the first antennal segment, excluding the neck and basal condyle; Scape Index (SI): SL/HW × 100; Eye Length (EL): Maximum length of the compound eye; Pronotal Width (PW): Maximum width of the pronotum in dorsal view; Weber's Length of the mesosoma (WL): Diagonal length, measured in lateral view from the anterior margin of the pronotum (excluding the collar) to the posterior extremity of the propodeal lobe; Propodeal Spine Length (PSL): Measured from tip of propodeal spine to closest point on outer rim of propodeal spiracle; Petiole Length (PtL): Length of the petiole in lateral view; Petiole Width (PtW): Maximum width of petiole in dorsal view; Petiole Height (PtH): Height of the petiole in lateral view; Postpetiole Length (PpL): Length of the postpetiole in lateral view (Longino 2003, Fig. 2); Postpetiole Width (PpW): Maximum width of postpetiole in dorsal view, excluding the helcium; Petiole Height Index (PtHI): PtH/PtL × 100; Petiole Width Index (PtWI): PtW/PtL × 100; Postpetiole Width Index (PpWI): PpW/PpL × 100; Waist Index (WI): PpW/PtW × 100.

Depositories of Material

Type specimens were examined and deposited in the collections listed below. Codes for public institutions generally follow those in Brandão (2000): BMNH, Natural History Museum, London, United Kingdom; CASC, California Academy of Science, USA; KUEC, Institute of Tropical Agriculture, Kyushu University, Fukuoka, Japan; THNHM, Thailand Natural History Museum, Technopolis, Khlong Luang, Pathum Thani, Thailand.

Molecular Data Collection

Genomic DNA was extracted from legs using a DNeasy Blood & Tissue kit (Qiagen, Maryland, USA). We sequenced fragments of five nuclear genes: arginine kinase (ArgK, 291 bp), carbamoylphosphate synthase (CAD, 192 bp), long wavelength rhodopsin (LW Rh, 747 bp exon/intron), DNA topoisomerase 1 (Top1, 666 bp), and wingless (Wg, 285 bp), and the mitochondrial genome, 3'region of the cytochrome oxidase I (COI, 711 bp). The total number of base pairs for all nuclear genes was 2,181. Seven new sequences were generated for this study (Table 1), and the remainders were taken from Blaimer (2012d). Nineteen species of *Crematogaster* (17 species of the Australo-Asian *Crematogaster* clade (Blaimer 2012d, Fig. 2. III), *Crematogaster osakensis*, *Crematogaster indosinensis* **sp. nov.**) were selected for molecular phylogenetic analysis (Table 1). Primers, amplification, and sequencing procedures followed Ward and Downie (2005), Ward et al. (2010), Blaimer (2012a, d), and Ward and Sumnicht (2012). The sequence data were deposited at GenBank or DDBJ.

The regions of the nuclear genes were amplified via the polymerase chain reaction (PCR) using primers shown in Table 2. Reactions were carried out at 10 µl volumes in a PCR Thermal Cycler MP (TaKaRa Bio Inc.) under the following conditions: first 40 cycles of 95°C for 30 s, annealing at 50–58°C for 30 s, and 72°C for 90 s, then 1 cycle of 95°C for 1 min, and finally 72°C for 3 min for the nuclear genes (Ward and Downie 2005, Blaimer 2012a).

A 711 bp region of the 3' region of COI was amplified via the PCR using primers "Jerry" and "Pat" (Simon et al. 1994). Reactions were carried out at 10 µl volumes in a PCR Thermal Cycler MP (TaKaRa Bio Inc.) under the following conditions: a first cycle of 94°C for 1 min, followed by 5 cycles of 94°C for 1 min, annealing at 48°C for 90 s, and 72°C for 90 s, then 30 cycles of 94°C for 1 min, annealing at 51°C for 90 s, and finally 72°C for 90 s for the COI.

PCR products were visualized on a 1% agarose E-Gel 96-well system (Invitrogen), and then purified with 1.0 µl of ExoSAP-IT (GE Healthcare Life Sciences). All products were sequenced in both directions using BigDye Terminator v3.1 (Applied Biosystems) on an ABI 3100 Avant DNA Sequencer (Applied Biosystems) at the Faculty of Science, Kyushu University, Fukuoka. Contigs were made using Vector NTI Advance TM ver. 11 (Invitrogen Corp.) and subsequently aligned by eye. Sequence data were assembled and edited in the program Vector NTI Advance Tm ver. 11 (Invitrogen Corp.) and MEGA 5 (Tamura et al. 2011) and subsequently aligned by eye.

Phylogeny was inferred using a Bayesian approach with MrBayes v.3.1.2 (Ronquist and Huelsenbeck 2003) and maximum likelihood with PAUP*4.0b10 (Swofford 2002). Data were partitioned by gene and codon position. We selected nucleotide sequence models for all partitions using the Akaike Information Criterion in the program MrModeltest 2.3 (Posada and Crandall 1998, Nylander et al. 2004) and PAUP*4.0b10 (Swofford 2002). The best-fit models for the nuclear genes were as follows: SYM + I (ArgK); HKY (CAD); HKY + G (LWRh, Wg); GTR + G (Top1).

Bayesian analyses involved Metropolis-coupled Markov chain Monte Carlo (MCMCMC) analysis employing two runs, each with four chains, sampled every 1,000 generations. All runs reached stationarity, as judged by the average standard deviation of split frequencies (ASDSF) approaching 0.01. Approximately 25% of samples were discarded as burn-in before parameters and trees were summarized. A maximum likelihood analysis was conducted using the heuristic search, the GTR + I + G model, and bootstrap search with 100 replicates in PAUP.

Nomenclature

This paper and the nomenclatural act it contains have been registered in Zoobank (www.zoobank.org), the official register of the International Commission on Zoological Nomenclature. The LSID (Life Science Identifier) number of the publication is: urn:lsid:zoobank.org:pub:E2D3F3E7-14F6-4465-91C8-ED3F0B66C9DE

Table 1. Specimens included in the current analysis with their associated GenBank or DDBJ numbers

Taxon name	ArgK	CAD	LWRh	Top1	Wg	COI
Crematogaster indosinensis	KU686885*	KU686886*	LC126692*	LC126019*	KU686887*	LC126018 *
Crematogaster abrupta	JQ326432	JQ326547	JQ326662	JQ326777	JQ326309	N/A
Crematogaster ampullaris	JQ326435	JQ326550	JQ326665	JQ326780	JQ326311	N/A
Crematogaster borneensis	JQ326442	JQ326557	JQ326672	JQ326787	JQ326318	LC126017 * **
Crematogaster coriaria	JQ326447	JQ326562	JQ326677	JQ326792	JQ326323	N/A
Crematogaster dahlii	JQ326449	JQ326564	JQ326679	JQ326794	JQ326325	N/A
Crematogaster decamera	JQ326450	JQ326565	JQ326680	JQ326795	JQ326326	N/A
Crematogaster fruhstorferi	JQ326502	JQ326617	JQ326732	JQ326847	JQ326377	N/A
Crematogaster HFmsp10	JQ326457	JQ326572	JQ326687	JQ326802	JQ326333	N/A
Crematogaster inflata	JQ326459	JQ326574	JQ326689	JQ326804	JQ326335	N/A
Crematogaster mjobergi	JQ326498	JQ326613	JQ326728	JQ326843	JQ326374	N/A
Crematogaster modiglianii	JQ326471	JQ326586	JQ326701	JQ326816	JQ326347	N/A
Crematogaster onusta	JQ326476	JQ326591	JQ326706	JQ326821	JQ326352	N/A
Crematogaster rothneyi	JQ326488	JQ326603	JQ326718	JQ326833	JQ326364	N/A
Crematogaster ss_AUS5	JQ326499	JQ326614	JQ326729	JQ326844	JQ326375	N/A
Crematogaster subcircularis	JQ326532	JQ326647	JQ326762	JQ326877	JQ326407	N/A
Crematogaster tetracantha	JQ326536	JQ326651	JQ326766	JQ326881	JQ326411	N/A
Crematogaster weberi	JQ326538	JQ326653	JQ326768	JQ326883	JQ326413	N/A
Crematogaster osakensis	JQ326483	JQ326598	JQ326713	JQ326828	JQ326359	N/A

It is noted that seven new sequences (*) were generated for this study, and the remainders were taken from Blaimer (2012d).

** Separate individuals from the Peninsular Malaysia have been used in DNA extraction.

Table 2. PCR primers used for the amplification of gene loci

Gene	Primer	Sequence (5′–3′)	Source
ArgK	AK1F2	ATGGTTGAYGCYGCYGTTYTGGA	Ward et al. (2010)
	AK345ERcr	ACTTACGGTGGGGTCGAGATTGC	Blaimer (2012a)
CAD	CD1258Fcr	CAGGCTGGAGAATTYGATTATTCGGG	Blaimer (2012a)
	CD1592R	GCRAAYATYTTYCTRTCYTCRGT	Ward et al. (2010)
LWRh	LR143F	GACAAAGTKCCACCRGARATGCT	Ward and Downie (2005)
	LR639ER	YTTACCGRTTCCATCCRAACA	Ward and Downie (2005)
Top1	TP1339F	GARCAYAARGGACCKGTRTTYGCACC	Ward and Sumnicht (2012)
	TP1793R	TTRCCCATYTTRGGRTGCTCRCCRCG	Ward and Sumnicht (2012)
	TP1987F	GGHGAAAARGAYTGGCARAARTAYGA	Ward and Sumnicht (2012)
	TP2192R	GARCARCARCCYACDGTRTCHGCYTG	Ward and Sumnicht (2012)
Wg	Wg578F	TGCACNGTGAARACYTGCTGGATGCG	Ward and Downie (2005)
	Wg1032R	ACYTCGCAGCACCARTGGAA	Ward and Downie (2005)
COI	Jerry	CAACATTTATTTTGATTTTTTGG	Simon et al. (1994)
	Pat	TCCAATGCACTAATCTGCCATATTA	Simon et al. (1994)

Table 3. Data on number of bases, number of variable characters (VC), and number of parsimony informative characters (PIC)

Gene	No. bases	No. VC	No. PIC
ArgK	291	32	18
CAD	192	12	8
LWRh	747	62	55
Top1	666	60	45
Wg	285	16	14
Total	2181	182	140

Results

The five nuclear gene sequences consisted of 2,181 bp and contained 182 variable characters (VC) and 140 parsimony informative characters (PIC; Table 3). Sequence for COI were generated for *Crematogaster indosinensis* **sp. nov.** and *C. borneensis*, but not forthcoming for other 17 species in this study. The COI sequences consisted of 711 bp and both had uniform sequence lengths.

Molecular phylogenetic analysis using nuclear genes revealed that *Crematogaster indosinensis* is sister to the remainder of the *C. borneensis*-group (100% posterior probability, 100% ML bootstrap; Fig. 1). Since both Bayesian analyses and maximum likelihood produced nearly identical topologies for the *C. borneensis*-group, only the Bayesian analysis is shown.

Discussion

Crematogaster indosinensis shares the following features diagnosed by Blaimer (2012c): 1) 10-segmented antennae; 2) rectangular head shape (but slightly wider than long in *C. indosinensis*); 3) suboval petiole in dorsal view; 4) globular postpetiole, and a faint posterior impression with the *C. borneensis*-group. Based on these morphological characters and molecular phylogenetic analysis, we consider *C. indosinensis* to be a member of the *C. borneensis*-group. *Crematogaster indosinensis* can be distinguished among the other members of the *C. borneensis*-group by the head being wider than long and having a developed subpetiolar process.

Maschwitz and Fiala (1995) reported a *Crematogaster* species associated with the ant-plant *Neonauclea* (Rubiaceae) from Sulawesi. In their molecular phylogeny, estimated using mitochondrial DNA analysis, Quek et al. (2004) proposed that the *Crematogaster* sp. was a sister group to the *C. borneensis*-group associated with *Macaranga* plants, albeit with lower support (88% posterior probability; 67% bootstrap value). However, the *Crematogaster* sp. is morphologically quite different from the *C. borneensis*-group (including *C. indosinensis*), particularly in the shape of antennae, mesosoma, petiole, and postpetiole.

The *Macaranga*-associated *C. borneensis*-group (former subgenus *Decacrema* in Asia) has been known from the Sundaic region (Peninsular Malaysia, Borneo, and Sumatra). The members of this group are considered to have codiversified with the *Macaranga* in the region (Quek et al. 2004). While the position of *C. indosinensis* suggests that the ancestral area for the *C. borneensis*-group is likely to be the Indochina region (Fig. 2), although it is needed to decide the phylogenetic relationship between the *C. borneensis*-group and the *Crematogaster*. sp. associated with ant-plant *Neonauclea* (Maschwitz and Fiala 1995, Quek et al. 2004).

In their phylogeographic analysis of *Macaranga* in Southeast Asia, Fiala et al. (2016) presented a possible scenario for the history of the association between ants and plants. In Peninsular Malaysia and Sumatra, *M. griffithiana* is associated with *Crematogaster borneensis*, but in eastern Thailand and Cambodia, *M. griffithiana* is associated with a *Camponotus* sp. (Formicinae). These authors proposed that possible original ant-associates had gone extinct during the Pleistocene, and that *C. borneensis* subsequently found an empty niche on *M. griffithiana* in Peninsular Malaysia and Sumatra, and also that *C. borneensis* may not have reached eastern Thailand and Cambodia. According to this scenario, *Crematogaster indosinensis* may have already occurred in the Indochina region. In addition, although it is unknown whether *C. indosinensis* has a relationship with ant-plants, their scenario assumes that *Camponotus* species formed an association with *Macaranga* before *C. indosinensis* did (Fig. 2).

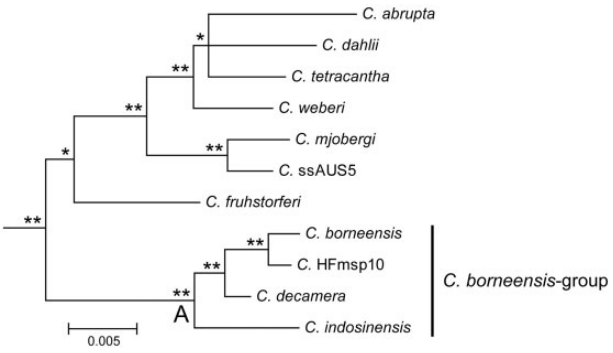


Fig. 1. Bayesian majority rule consensus tree reconstructed for 19 taxa with five genes (ArgK, CAD, LWRh, Top1, and Wg) in a MrBayes analysis. Some outgroups are not shown. Posterior probability values greater than 95% are above the branches (*>95%, **=100%). Data were partitioned by gene and codon position and analyzed with one million generations and a burn-in of 1,000 generations.

Some species of the genus *Crematogaster* show polymorphism in size and sculpture (Longino 2003; Hosoishi and Ogata 2009, 2015; Blaimer 2012b). In general, the *C. borneensis*-group associated with *Macaranga* does not show such polymorphism, but *C. indosinensis* does exhibit distinct polymorphism in size (Fig. 2). The ancestral character state of the *C. borneensis*-group is considered to be polymorphic in size, which then presumably disappeared in the ants associated with *Macaranga* over time.

Interspecific COI sequence divergence was 20.0% between *C. indosinensis* and *C. borneensis*. The substitution rate of mitochondrial DNA has been widely used in divergence time estimation of closely related taxa. Quek et al (2004) employed a lower rate of 1.5% for COI in *Macaranga*-associated *Crematogaster* species, but several studies employed higher rates for COI (3.34% for Coleoptera: Cicindelidae by Pons and Vogler 2005; 3–4% for Orthoptera: Tettigoniidae by Shapiro et al. 2006). The accurate estimation requires careful model choice, biogeographic data, and geological evidence (Papadopoulou et al. 2010). According to their range of the divergence rates (1.5–4% per million years), we estimated that node A in Fig. 1 corresponded to an age of ca. 13.3–5 million years ago. These roughly estimated dates fall close to ones of the mid- to late-Miocene plant dispersal events that occurred ~14 and 9.5 million year ago in the region (Morley 2000). At that time, most members of the *C. borneensis*-group are considered to have undergone drastic diversification with respect to the formation of mutualistic associations with *Macaranga*, which is widely distributed in the Sundaic region; however, whether *C. indosinensis* formed such a relationship is not known.

Taxonomy

Crematogaster indosinensis Hosoishi & Ogata, New Species

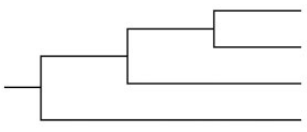
(Fig. 3–8)
(urn:lsid:zoobank.org:act:D559D714-E7DB-4449-89AF-447955D49F36)

Type Material

HOLOTYPE. Worker, labeled: "Cambodia: Permanent Sample Plots (natural forest), Kampong Thom Province, 12°34′N, 105°23′E, 11.i.2010 (SH10-Cam46) (*S. Hosoishi*)" (THNHM: KUMANT034). **PARATYPES.** Three workers, same data as holotype. Deposited in BMNH: KUMANT035, CASC: KUMANT036, KUEC: KUMANT037).

Worker

Measurements: HW = 0.78 (0.62–1.05); HL = 0.75 (0.60–1.00); CI = 104 (102–107); SL = 0.5 (0.42–0.63); SI = 65 (60–69);

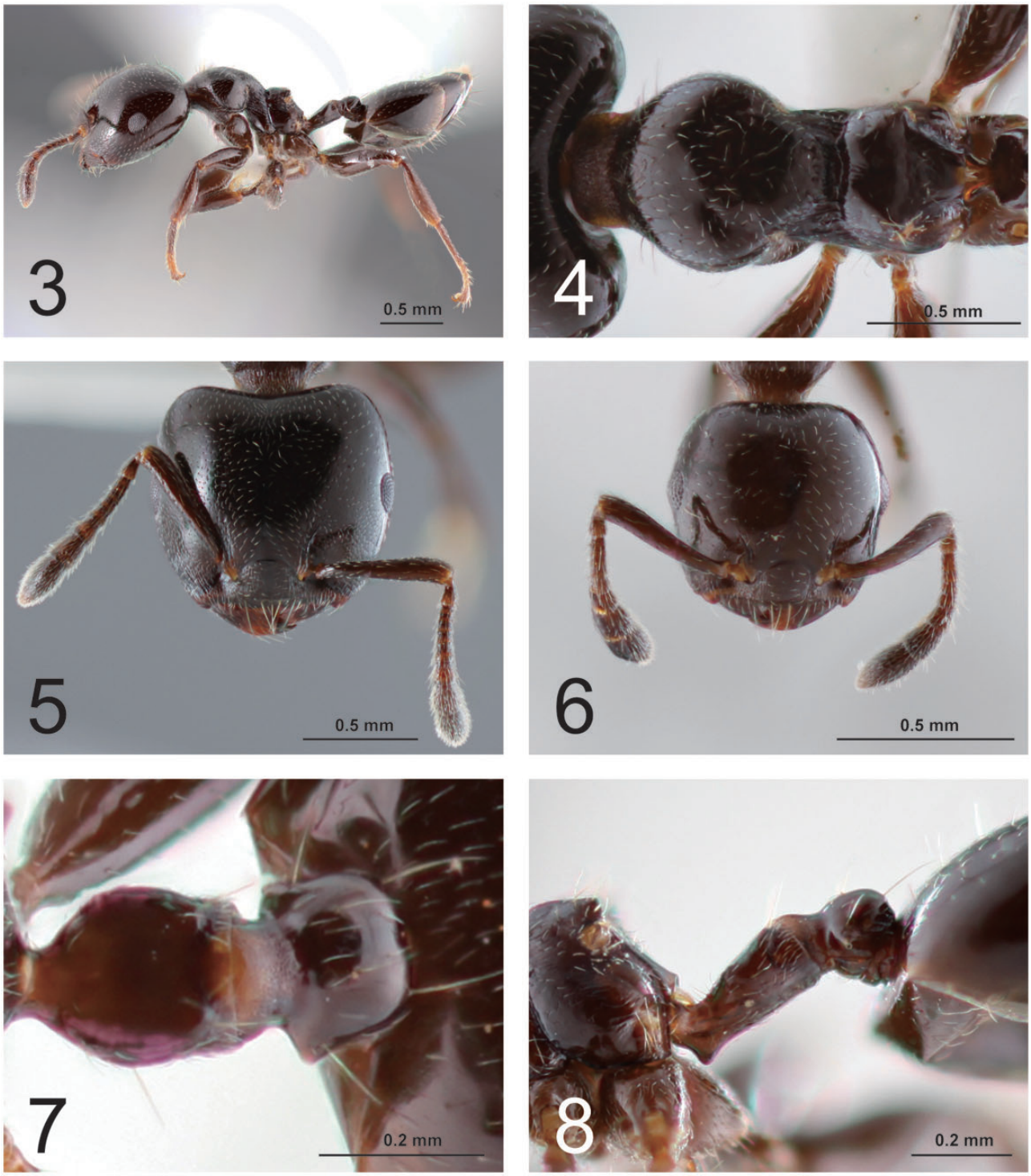


Distribution	Host plant	Polymorphism
Sundaland	*Macaranga*	Absent
Sundaland	*Macaranga*	N/A
Sundaland	*Macaranga*	Absent
Indochina	N/A	Present

Fig. 2. Character states of distribution, host plant association, and size polymorphism in the *Crematogaster borneensis*-group. In this study, the "Sundaland" includes Peninsular Malaysia, Borneo, and Sumatra.

EL = 0.15 (0.12–0.18); PW = 0.44 (0.35–0.57); WL = 0.79 (0.65–1.01); PSL = 0.05 (0.04–0.06); PtL = 0.21 (0.17–0.28); PtW = 0.18 (0.14–0.23); PtH = 0.14 (0.12–0.17); PpL = 0.13 (0.11–0.15); PpW = 0.17 (0.14–0.20); PtHI = 68 (61–71); PtWI = 87 (82–94); PpWI = 131 (127–137); WI = 94 (87–100) (n = 4).

Workers with size polymorphism. Head subquadrate in full-face view, with weakly concave posterior margin, subparallel sides; posterior portion developed in large workers. Occipital carinae developed. Mandible with four teeth, apical and subapical teeth large. Anterior margin of clypeus convex; anterolateral margins of clypeus protruded anteriorly; posterior margin of clypeus rounded between frontal lobes. Frontal carinae almost parallel. Antennae 10-segmented; antennal club 2-segmented. Scape not reaching posterior corner of head. Basal flagellar segment (antennal segment III) slightly broader than long. Compound eyes not projecting beyond lateral margins of head in full face view.



Figs. 3–8. *Crematogaster indosinensis*, worker. 3, Body in lateral view; 4, dorsal view of mesosoma; 5, full-face view of head in the largest worker [HW 1.05; HL 1.00]; 6, full-face view of head in the smallest worker [HW 0.62; HL 0.61]; 7, petiole and postpetiole in dorsal view; 8, petiole and postpetiole in lateral view.

Pronotum and mesonotum almost fused without defined suture, forming same dorsal outline in lateral view. Metapleural gland opening slit-shaped. Propodeal spiracle circular, situated at posterolateral corners, not touching metapleural gland bulla. Metanotal groove convex posteriorly in dorsal view, deep and forming concave region between mesonotum and propodeum. In dorsal view, longitudinal rugulae connecting between mesonotum and propodeum; the boundary distinct. Propodeal spines weakly developed as short process.

Petiole elliptical shaped, but flattened with weakly convex sides, slightly longer than broad; spiracle situated at almost midportion between dorsal and ventral margin of petiole in lateral view, directed laterally. Subpetiolar process developed. Postpetiole without longitudinal median sulcus, but weakly bilobed behind in large workers; spiracle situated anteriorly on lateral surface.

Dorsal surface of head generally smooth; gena with rugulae. Clypeus with longitudinal rugulae in large workers, but weakly punctate in small workers. Promesonotum smooth and shining, but anterior collar sculptured. Lateral surface of pronotum smooth and shining. Mesopleuron generaly smooth, but with longitudinal rugulae on higher portion. Anterodorsal surface of propodeum smooth and shining, but posterior declivity punctate. Lateral surface of propodeum generally smooth and shining. Dorsal and lateral surfaces of petiole smooth and shining. Dorsal and lateral surfaces of postpetiole smooth and shining.

Standing pilosity sparse. Dorsal face of head with some pairs (ca. 6–7) of erect setae and short appressed setae. Clypeus with some pairs (ca. 3–4) of erect setae. Anterior clypeal margin with single median setae and one pair of long setae mixed with short setae laterally. Scape with erect and appressed setae sparse. Promesonotal dorsum with sparse erect setae and appressed setae. Petiole with one pair of suberect setae posteriorly. Postpetiole with three pairs of suberect setae dorsally, laterally, and posteriorly. Fourth abdominal tergite with appressed setae and sparse erect setae. Body color redbrown.

Etymology

The specific name refers to the region from which the material is collected.

Distribution and Biology

This species is known only from the type locality in Cambodia. The type material examined in this study was collected from the permanent sample plots established by the Cambodian Forestry Administration. The author (SH) collected the workers foraging on lower vegetation. It is unknown whether *C. indosinensis* has a mutualistic relationship with ant-plants, because no information on their nesting habits is available. In a list published by Toyama et al. (2013) of the vascular plants found in the permanent plots, *Macaranga griffithiana* was recorded from the edge of a swamp in Kampong Thom Province. Fiala et al. (2016) reported that *Camponotus* sp. was collected from *M. griffithiana* in Kampong Thom Province in Cambodia and Chantaburi Province in Thailand, but they did not collect any *Crematogaster* species from it. It is beyond the scope of this study to infer aspects of the life history of *C. indosinensis*, but further studies on the biology of this species need to be conducted in the future.

Comments

This species is similar to *C. borneensis* and *C. decamera*, but can be distinguished from both of these species by having a developed subpetiolar process (Fig. 8). While our collections of this species are very limited ($n = 4$), but those specimens show distinct size polymorphism. The posterior portion of head is developed in large workers (Fig. 5), but rounded in small workers (Fig. 6). The head in the largest worker (HW 1.05) is ca. 1.7× as wide as that one in the smallest worker (HW 0.62). In general, the *C. borneensis*-group associated with *Macaranga* does not show such polymorphism in size. The relationship between the presence/absence of polymorphism and ant-plant association is currently unclear because little is known about the biology of *C. indosinensis*.

The relative high COI divergence observed between *C. indosinensis* and *C. borneensis* (20% K2P distances) suggests that the taxa diverged relatively long ago, because the mean COI divergence between *C. chhangi* (Cambodia) and *C. fraxatrix* (Peninsular Malaysia and Borneo) was 9.4% (8.1–10.8%) (Hosoishi and Ogata 2014).

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