



Exploring the diversity of the Malagasy *Ponera* (Hymenoptera: Formicidae) fauna via integrative taxonomy

Sándor Csősz^{1,2} · Ana C. Loss³ · Brian L. Fisher⁴

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Abstract

The genus *Ponera* includes over 60 extant species worldwide. These tiny, endogeic predator ants are predominantly distributed in the Indomalaya and Australasia regions, with a few additional Holarctic species. Herein, we explore and describe the diversity of the Malagasy *Ponera* fauna through an integrative taxonomic approach. We obtained our morphological species hypotheses from multivariate analyses of ten continuous morphometric characters. Species boundaries and reliability of morphological clusters were tested via confirmatory Linear Discriminant Analysis (LDA), cross-validation (LOOCV), and analyses of a mitochondrial COI gene fragment. According to the combined application of the analyses, altogether, three species are inferred in the Malagasy region, *Ponera petila* Wilson (1957), *P. swezeyi* Wheeler (1933), and *P. adumbrans* Csősz & Fisher sp. n. *Ponera petila* and *P. swezeyi* belong to the Indo-Australian *Ponera tenuis* group; the third species, *P. adumbrans* sp. n., is morphologically similar to the Papua New Guinean *P. clavicornis* Emery (1900). Furthermore, Linear Discriminant Analysis classified the type specimens of *P. bableti* Perrault (1993), along with a *P. petila* cluster with posterior $p = 1$. Therefore, we propose the new junior synonymy of *P. bableti* with *P. petila*. Madagascar's extant biodiversity is predominantly explained by colonization events from the African continent across the Mozambique channel via rafting. However, since no native *Ponera* species are known from the Afrotropical continent, and the closest congeners have an almost exclusively Indo-Australian distribution, the likelihood of an Indo-Australian origin of the Malagasy *Ponera* fauna is implied.

Keywords Biodiversity · Colonization · Biological invasion · Morphometry · Phylogeny

Introduction

The Malagasy region, i.e., Madagascar and the surrounding smaller islands, harbors unique fauna. Madagascar's fauna is taxonomically imbalanced (Samonds et al., 2013). Many lineages of its extant terrestrial and freshwater assemblages can be characterized by high levels of species endemism (Goodman & Benstead, 2005). Within some other clades,

Madagascar's fauna is species-poor. This is true for ants as well, with the exception of some hyperdiverse genera, e.g., *Tetramorium* (Hita-Garcia & Fisher, 2014), *Camponotus* (Rakotonirina et al., 2016; Rasoamanana et al., 2017), and *Pheidole* (Salata & Fisher, 2021). Other genera, such as the genus *Ponera*, are represented by only a few species in the region. The genus *Ponera*, encompassing tiny, endogeic predator ants, is primarily distributed in the Indomalaya and Australasia regions, with a few additional Holarctic species (Leong et al., 2019; Taylor, 1967; Wilson, 1957) and currently contains 60 extant and five fossil species worldwide (<https://www.antcat.org/>). The taxonomy of the handful of *Ponera* species in the Malagasy region has never constituted the basis of focused research before.

We, for the first time, revise the Malagasy *Ponera* species via an integrated taxonomic protocol incorporating morphometrics, morphology, and DNA sequence data. Quantitative analyses of continuous morphometric traits were performed via NC-PART clustering as described by Csősz and Fisher (2016a, b). The clusters returned by this protocol

✉ Sándor Csősz
sandorcsosz2@gmail.com

¹ ELKH-ELTE-MTM Integrative Ecology Research Group, Pázmány Péter ave 1/C, Budapest 1117, Hungary

² Department of Systematic Zoology and Ecology, Institute of Biology, ELTE-Eötvös Loránd University, Budapest, Hungary

³ National Institute of the Atlantic Forest, Av. José Ruschi, 4, Santa Teresa, ES 29650-000, Brazil

⁴ Entomology, California Academy of Sciences, 55 Music Concourse Drive, San Francisco, CA 94118, USA

are considered species unless this hypothesis conflicts with other biological information. Species boundaries and reliability of morphological clusters were tested via confirmatory Linear Discriminant Analysis (LDA), cross-validation (LOOCV), subjective evaluation of qualitative morphological characteristics, and analyses of a mitochondrial COI gene fragment. According to the combined application of the analyses, altogether, three species are inferred, *Ponera petila* Wilson (1957), *P. swezeyi* Wheeler (1933), and *P. adumbrans* Csősz & Fisher sp. n.

Two of the three known Malagasy *Ponera* species, *P. petila* and *P. swezeyi*, belong to the Indo-Australian *Ponera tenuis* group; the third species, *P. adumbrans* sp. n., is morphologically similar to the Papua New Guinean *P. clavicornis* (Emery, 1900), that further strengthens the Indo-Australian connection hypothesis. Beyond *Ponera*, five other Malagasy ant genera, *Adelomyrmex*, *Aphaenogaster*, *Chrysapace*, *Eurhopalothrix*, and *Vollenhovia*, exhibit this biogeographic pattern, i.e., are not present in Africa but are distributed in India or the Indo-Australian regions (Fisher & Bolton, 2016).

Madagascar's extant biodiversity is predominantly explained by colonization events from the African continent across the Mozambique channel via rafting once lower sea levels allowed for the journey in the Cenozoic era (Ali & Huber, 2010; Godfrey et al., 2020; Masters et al., 2020). However, the occurrence of Malagasy *Ponera* species raises issues about the colonization route of the Malagasy *Ponera*, because (i) there are no known native extant or extinct *Ponera* species known from the Afrotropical continent, and (ii) both the *tenuis* group and *clavicornis* group, to which the Malagasy *Ponera* belong, almost exclusively exhibit an Indo-Australian distribution. Therefore, an Indo-Australian origin of the Malagasy *Ponera* fauna should be considered.

Material and methods

Ant samples used in this study comply with the regulations for export and exchange of research samples outlined in the Convention on Biological Diversity and the Convention on International Trade in Endangered Species of Wild Fauna and Flora. For fieldwork conducted in Madagascar, permits to research, collect, and export ants were obtained from the Ministry of Environment and Forest as part of an ongoing collaboration between the California Academy of Sciences and the Ministry of Environment and Forest, Madagascar National Parks and Parc Botanique et Zoologique de Tsimbazaza. Approval Numbers: N° 0142N.EA03.MG02, N° 340N-EV10.MG04, N° 69 du 07.04.06, N° 065N-EA05.MG11, N° 047N-EA05.MG11, N° 083N-A03.MG05, N° 206 MINEN-VEF.SG.DGEF.DPB.SCBLF, N° 0324N.EA12.MG03, N° 100 MEF.SG.DGEF.DADF.SCBF, N° 0379N.EA11.MG02,

N° 200N.EA05.MG02. Authorization for export was provided by the Director of Natural Resources. For research in the Seychelles, permits were obtained from the Seychelles Bureau of Standards, Ministry of Environment, and National Park Authority. Material Transfer Agreement was provided by the Seychelles Ministry of Environment.

In the present study, 10 continuous morphometric traits were recorded in 154 worker individuals belonging to 90 nest samples from the Malagasy, Indo-Australian, and Pacific regions.

The material is deposited in the following institutions, abbreviations after Evenhuis (2013): CASC (California Academy of Sciences, San Francisco, California, USA), MCZC (Museum of Comparative Zoology, Cambridge, Massachusetts, USA), MNHN (Muséum National d'Histoire Naturelle), PSWC (Phil S. Ward's collection, University of California Davis, Davis, California, USA), BPBM (Bernice P. Bishop Museum, Honolulu, Hawaii, USA), HNHM (Hungarian Natural History Museum). Type material and samples that were morphometrically investigated are presented in the "Type material investigated" and "Material examined" sections in the format as follows: CASENT code, collection code, verbatim locality, longitude, latitude, elevation in meters, collector, date (number of individuals measured, abbreviation of depository). Additional information on habitat and microhabitat, whenever available, is given in brackets. For the full list of material, see Supplementary Table S1. All images of specimens are available online on AntWeb (<https://www.antweb.org/>). Images are linked to their specimens via the unique specimen identifier affixed to each pin CASENT0159966. Digital color montage images were created using a JVC KY-F75 digital camera and Syn-croscopy Auto-Montage software (version 5.0), or a Leica DFC 425 camera in combination with the Leica Application Suite software (version 3.8). In verbatim descriptions of the Malagasy *Ponera* species, we follow terminology and character states established by Bolton and Fisher (2011).

Morphometric character recording

All measurements were taken by the first author using a Leica M165C stereomicroscope equipped with an ocular micrometer at a magnification of $\times 120$. Body size dimensions are expressed in μm . Due to the abundance of worker specimens relative to queen and male specimens, the present revision is based on the worker caste only, which can further be justified by the fact that the name-bearing type specimens in this study belong to the worker caste. For the definition of morphometric characters, earlier protocols (Csősz & Seifert, 2003) were considered. Explanations and abbreviations for measured characters are given in Table 1. Morphometric data for each measured specimen are given in Supplementary Table S2.

Table 1 Abbreviations (Abbr.) for morphometric characters, and the protocol used to measure traits

Abbr.	Measurement protocol
CL	Cephalic length; maximum median length of head capsule. The head must be carefully tilted so the maximum length is positioned in the measuring plane.
CWb	Maximum cephalic width in full-face view. Measurement of the real cuticular surface and not of the diffuse pubescence surface.
CS	Absolute cephalic size. The arithmetic mean of CL and CWb.
FRS	Maximum width of frontal lobes.
ML	Diagonal length of the mesosoma in profile. Measured in lateral view from the anteriormost point of the anterior pronotal slope to the caudalmost point of the lateral metapleural lobe.
MW	Maximum width of pronotum.
NOH	Petiole node height; measured in a right angle from a reference line beginning at the transition point between the caudal node profile and the caudal petiolar neck and ending at the most dorsal point of node corner.
PEH	Maximum height of the petiole; measured from the subpetiolar toothlike process to the top of the petiole.
PEL	Petiole length; horizontal distance from the tip of the frontolateral node corner to the caudalmost point of the petiole.
PEW	Maximum width of the petiole in dorsal view.
SL	Maximum straight-line scape length excluding the articular condyle.

Further indices that have consistently been applied by earlier researchers (Perrault, 1993; Taylor, 1967; Wilson, 1957) are also provided in this work in order to improve integration of the new findings to earlier species concepts:

CI Cephalic index ($CWb/CL \times 100$)

SI Scape index ($SL/CWb \times 100$)

PNI Petiolar node index ($PEW/MW \times 100$)

Statistical framework—hypothesis formation and testing

Generating prior species hypotheses via the combined application of NC clustering and PART. This method searches for discontinuities in continuous morphometric data, sorts all similar cases into the same cluster (see Seifert et al., 2014), and determines the ideal number of clusters. The present statistical framework follows the procedure applied in Csősz and Fisher (2016a, b). Advantages and limitations of the present procedure are discussed there.

Arriving at final species hypothesis using confirmatory LDA and LDA ratio extractor. To provide increased reliability of species delimitation, hypotheses for clusters and classification of cases via exploratory processes were confirmed by LDA leave-one-out cross-validation (LOOCV). Analyses were done in R (R Core Team, 2020).

DNA sampling

We sequenced 658 base pairs (bp) of the mitochondrial cytochrome oxidase I (COI) gene from 182 Malagasy *Ponera* specimens. DNA extraction and COI sequencing were performed at the University of Guelph (Ontario, Canada) following

the protocol described in Fisher and Smith (2008). An additional 60 *Ponera* sequences from different biogeographic regions, including Australasia and Indomalaya, and 3 outgroup sequences (*Diacamma*, *Parvoponera*, and *Pseudoponera*) were obtained from GenBank. Outgroup selection was based on *Ponera* phylogeny from Branstetter and Longino (2019). GenBank accession code and additional information for all sequences are given in Supplementary Table S3.

Molecular phylogenetic inference

We used molecular phylogenetic inference to confirm species taxonomic identification. Sequences were aligned using the Geneious Alignment algorithm implemented in Geneious 6.1.8 (Biomatters Ltd.). We included only unique haplotypes from each locality, resulting in a final alignment with 86 sequences. Phylogenetic analyses were performed using Bayesian inference in Mr. Bayes 3.2.7a and Maximum Likelihood using RAxML-NG online platform (<https://raxml-ng.vital-it.ch/>). The best-fit model of molecular substitution was estimated using PartitionFinder2 (Lanfear et al., 2016) under the Bayesian Information Criteria and linked branch lengths. The best partitioning scheme selected has 3 subsets, partitioning by codon position, as follows: GTR + G for first position, F81 for second position, and TRN + G for third position. Maximum likelihood bootstrap consensus tree was estimated using SumTrees 4.0.0 (Sukumaran & Holder, 2010, 2015). For Bayesian inference, two independent analyses of Metropolis coupling MCMC algorithm (MC³) ran for 10⁶ generations, sampling every 500 generations, resulting in 1500 trees after a burn-in of 25%. Each analysis ran with 4 chains, 1 cold and 3 heated at a temperature of 0.1 and chain swap frequency of 1 each generation. Convergence of independent runs was assumed when the standard deviation of split frequencies reached values below 0.01.

Species concept

Integration of the evidence provided by different lines of evidence into an integrative species hypothesis is performed based on the principles advocated by Schlick-Steiner et al. (2010) and “integration by congruence” protocol introduced by Padial et al. (2010). In addition, we employ the universal GAGE species concept formulated by Seifert (2020), which represents the most widely used and accepted theoretical and practical indications in ant taxonomy.

Results

The exploratory NC-clustering, in combination with two hypothesis-free partitioning methods “hclust” and “kmeans,” concordantly returned three completely separated clusters (Fig. 1) in Malagasy *Ponera* samples based on 10 morphometric characters. The confirmatory LDA including the pessimistic LOOCV confirmed this pattern showing complete classification success. The type specimens (four syntype specimens of *P. swezeyi* are nested in one of the clusters, while the holotype of *P. petila* and the type series of *P. bableti* are nested together in another). These types have also been classified along with the relevant cluster with posterior $p = 1$, when type specimens were set to wildcard

in LDA, i.e., no grouping label was added for type specimens. The third cluster endemic to Seychelles is considered a new species. Though this species clearly belongs to the *P. clavicornis* group, no similar available species could be identified using the literature (Leong et al., 2019; Perrault, 1993; Taylor, 1967; Wilson, 1957). This species is named *Ponera adumbrans* sp. n. reflecting its restriction to Seychelles islands.

Phylogenetic analyses corroborate this finding, showing three independent and monophyletic clusters from the Malagasy region (Fig. 2). We found complete agreement between monophyly of clusters in the molecular phylogeny and the quantitative morphology-based cluster delimitation protocols. Taking all of these lines of evidence into account, we consider the three clusters to be three species.

The three *Ponera* species distributed in the Malagasy region also differ in qualitative diagnostic features and body ratios (for body ratios, see Table 2).

Synopsis of Malagasy *Ponera* species

adumbrans Csősz & Fisher sp. n. (Fig. 3A–C)

petila Wilson, 1957 (Fig. 4A–C)

swezeyi (Wheeler, 1933) (Fig. 5A–C)

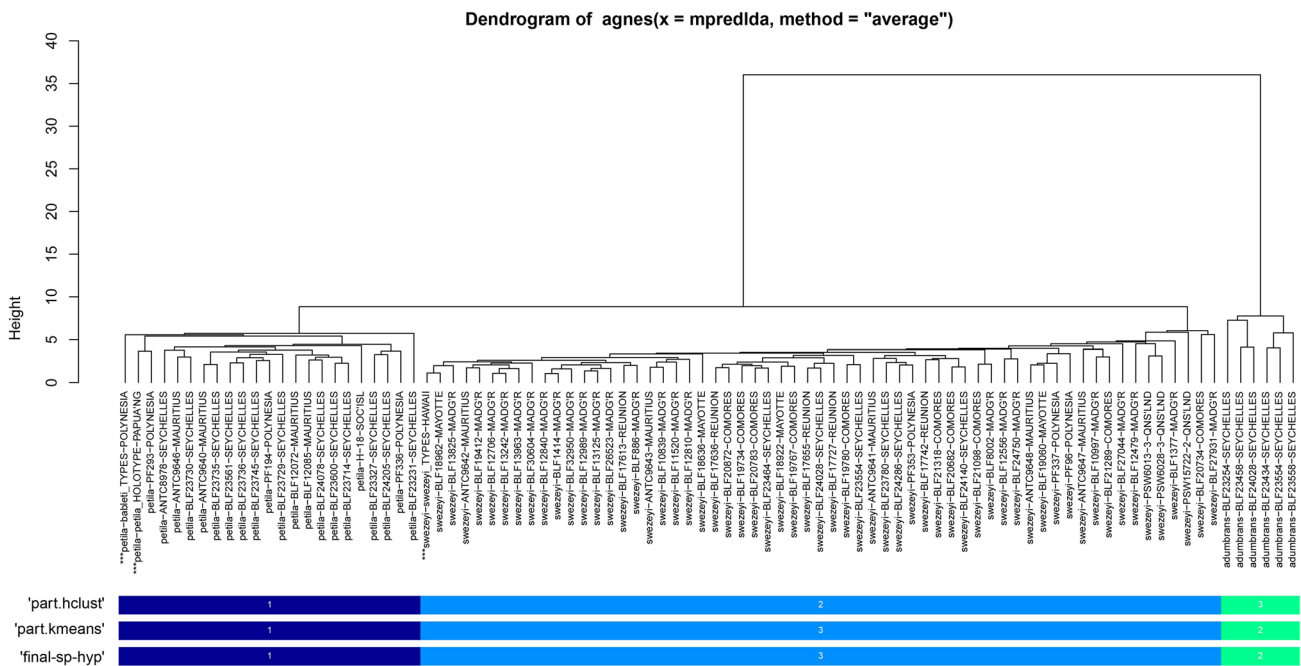


Fig. 1 Dendrogram solution for Malagasy *Ponera* species. Sample information in the dendrogram follows this format: final species hypothesis and CASENT number as a unique sample identifier followed by the capitalized geographic region separated by a hyphen. Two out of three columns of color bars represent prior species hypothesis resulted by method PART using two cluster methods “part.hclust” and “part.kmeans” (for additional

information see text). Final species hypothesis bar shows classification of samples after confirmation by cross-validated LDA. Different colors distinguish species. *Ponera petila*: dark blue, *P. adumbrans* sp. n.: green, *P. swezeyi*: light blue. Types are marked by asterisks

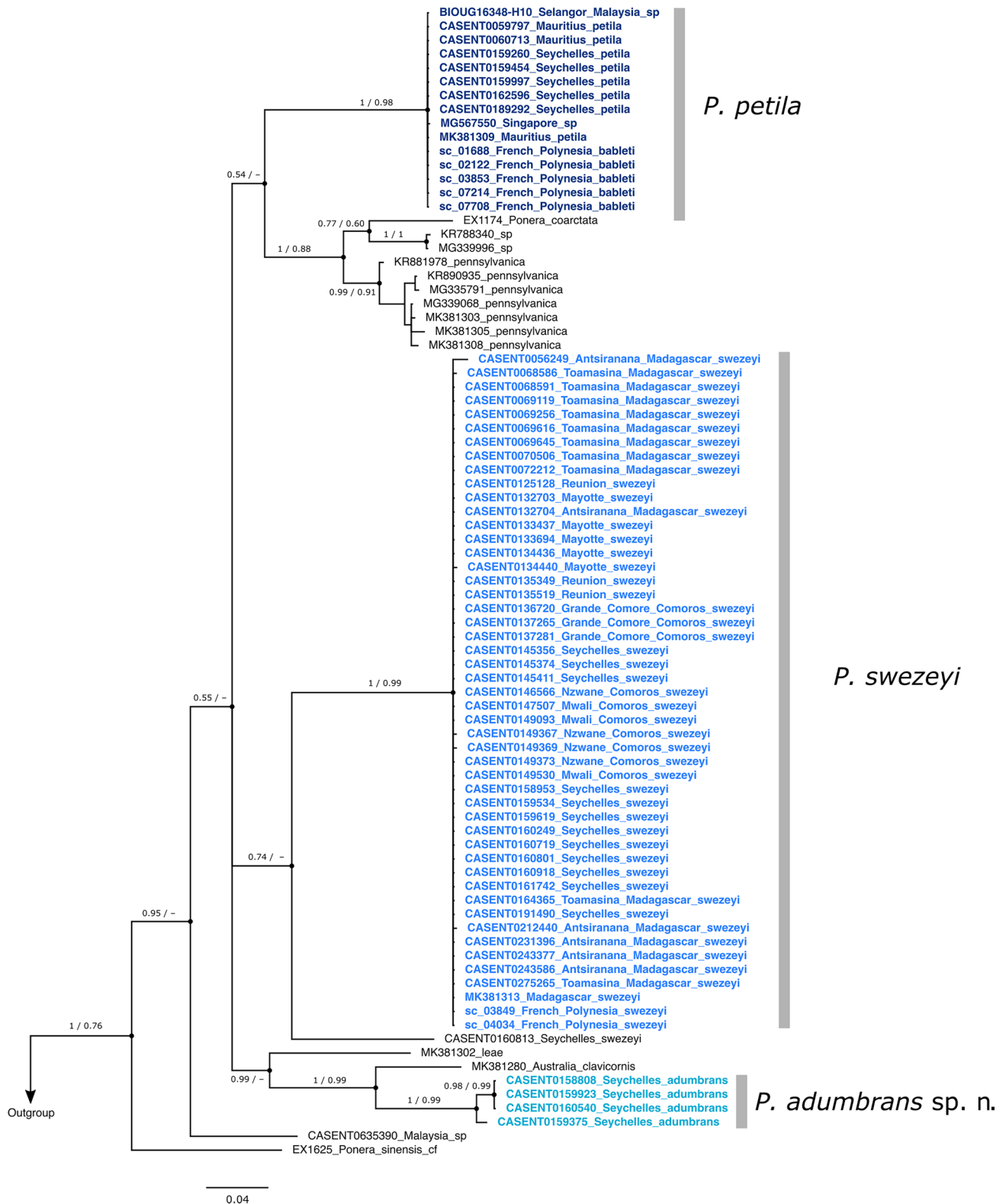


Fig. 2 Phylogeny of the Malagasy *Ponera* fauna. Bayesian inference phylogeny of *Ponera* COI sequences. Numbers associated to branches indicate Bayesian posterior probabilities and maximum likelihood bootstrap values. Only values greater than 50% are shown. Scale bar represents the

number of substitutions per site. Different colors distinguish species. *Ponera petila*: dark blue, *P. adumbrans* sp. n.: green, *P. swezeyi*: light blue

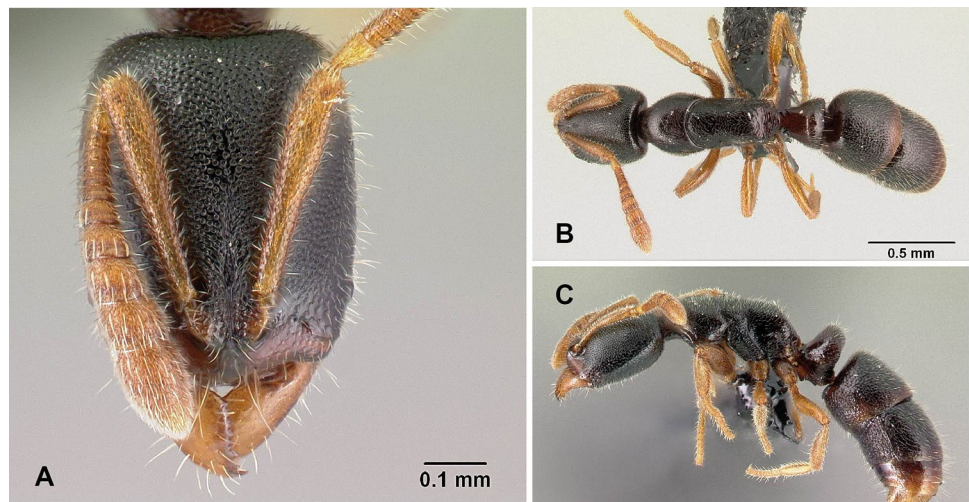
Table 2 Mean of morphometric ratios calculated for each species on individual level. Morphometric trait ratios and \pm SD are provided in the upper row, and minimum and maximum values are given in parentheses in the lower row

	<i>adumbrans</i> (n=8)	<i>swezeyi</i> (n=105)	<i>petila</i> (n=27)
CS	533 \pm 10.4 [518, 550]	358 \pm 7.2 [343, 376]	386 \pm 4.7 [372, 393]
FL.CS	0.25 \pm 0.01 [0.24, 0.26]	0.21 \pm 0.01 [0.19, 0.23]	0.22 \pm 0.01 [0.20, 0.24]
SL.CS	0.80 \pm 0.02 [0.79, 0.83]	0.71 \pm 0.01 [0.68, 0.74]	0.74 \pm 0.01 [0.72, 0.76]
MW.CS	0.69 \pm 0.01 [0.67, 0.70]	0.63 \pm 0.02 [0.58, 0.67]	0.65 \pm 0.01 [0.64, 0.68]
PEW.CS	0.55 \pm 0.02 [0.52, 0.58]	0.47 \pm 0.01 [0.43, 0.51]	0.50 \pm 0.01 [0.48, 0.53]
ML.CS	1.52 \pm 0.02 [1.51, 1.57]	1.47 \pm 0.02 [1.41, 1.57]	1.44 \pm 0.03 [1.39, 1.51]
PEH.CS	0.70 \pm 0.02 [0.68, 0.74]	0.68 \pm 0.02 [0.63, 0.72]	0.68 \pm 0.02 [0.65, 0.72]
NOH.CS	0.38 \pm 0.02 [0.37, 0.41]	0.34 \pm 0.01 [0.31, 0.37]	0.35 \pm 0.01 [0.32, 0.36]
PEL.CS	0.41 \pm 0.01 [0.39, 0.43]	0.40 \pm 0.01 [0.36, 0.42]	0.37 \pm 0.01 [0.36, 0.39]
CI	77 \pm 0.01 [75, 78]	74 \pm 0.02 [70, 79]	76 \pm 0.02 [74, 77]
SI	93 \pm 2.4 [90, 96]	83 \pm 1.6 [79, 87]	85 \pm 1.8 [82, 88]
PNI	80 \pm 2.5 [77, 83]	75 \pm 2.9 [67, 84]	77 \pm 2.3 [71, 81]

Key to workers of the Malagasy *Ponera* species

1. Dark brown species (Fig. 3A–C). The largest species in the region, head width (CWb): $> 400 \mu\text{m}$ [447, 478].
...*adumbrans* sp. n.

Fig. 3 A–C *Ponera adumbrans* sp. n. paratype worker (CASENT0159966). Head in full-face view (A), dorsal view of the body (B), lateral view of the body (C)



- Workers yellow to light brown (Figs. 4A–C and 5A–C). Small species (CWb): $< 400 \mu\text{m}$ [291, 339]...2
- 2. Anterolateral and posterior surface of petiolar node meet in rounded transition (Fig. 4C). Larger species (CWb): $333 \mu\text{m}$ [323, 339]. Scape longer (SL/PEL): 1.99 [1.86, 2.09], [5–95% percentiles: 1.92, 2.08] ...*petila* Wilson (1957).

Posterior surface of petiolar node bearing a ridge or fine vertical carina demarcating the posterior and lateral surfaces (Fig. 5C). Smaller species (CWb): $306 \mu\text{m}$ [291, 323]. Scape shorter (SL/PEL): 1.79 [1.68, 1.91], [5–95% percentiles: 1.71, 1.89]...*swezeyi* (Wheeler, 1933).

Ponera adumbrans Csósz & Fisher sp. n.

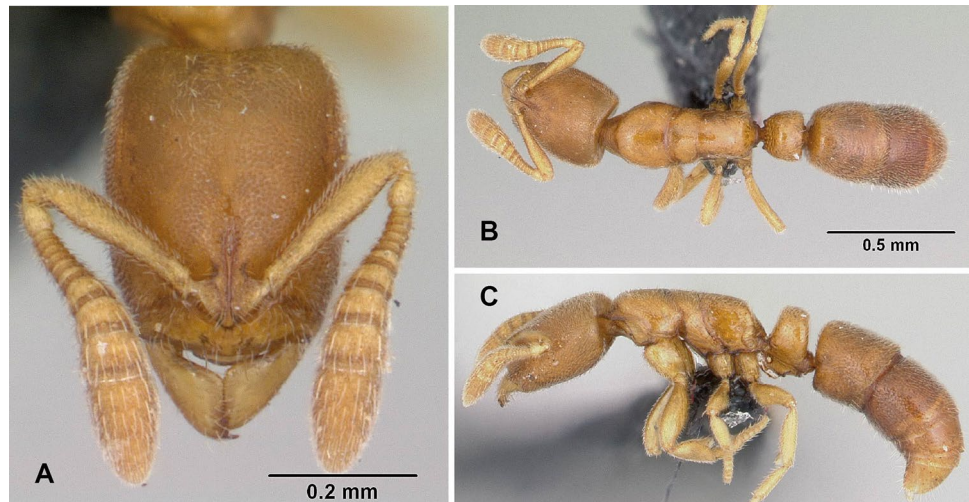
(Fig. 3A–C, Table 2)

Type material investigated

Holotype Seychelles: CASENT0160838, collection code: BLF23554, Silhouette Island, on ridge toward Mont Corgat, [forest, sifted litter, leaf mold, rotten wood], -4.49537, 55.23946, alt. 445 m, B.L. Fisher et al., 2010.01.28, (3w, CASC);

Paratypes Six workers with the same label data as the holotype under CASENT codes: **Seychelles:** CASENT0160837, collection code: BLF23554, (1w, CASC); CASENT0160840, collection code: BLF23554, (1w, CASC); CASENT0159923, collection code: BLF23458, (1w, CASC); CASENT0159088, collection code: BLF23558, (1w, CASC); CASENT0159966, collection code: BLF23434, (1w, CASC); CASENT0158808, collection code: BLF23254, (1w, CASC); CASENT0159375, collection code: BLF24028 (1w, CASC).

Fig. 4 A–C *Ponera petila* non-type worker (CASENT0059796). Head in full-face view (A), dorsal view of the body (B), lateral view of the body (C)



Etymology

The specific name "adumbrans" is a Latin singular present active participle in the nominative case that means "obscuring or silhouetting." It refers to the obscure origin of this species in the Malagasy region and the type locality, Silhouette Island.

Description of workers

General appearance Small species, absolute cephalic size (CS): 533 μm [518, 550]. Body color brown to black. Body concolorous, with antennae, funiculus, and legs lighter.

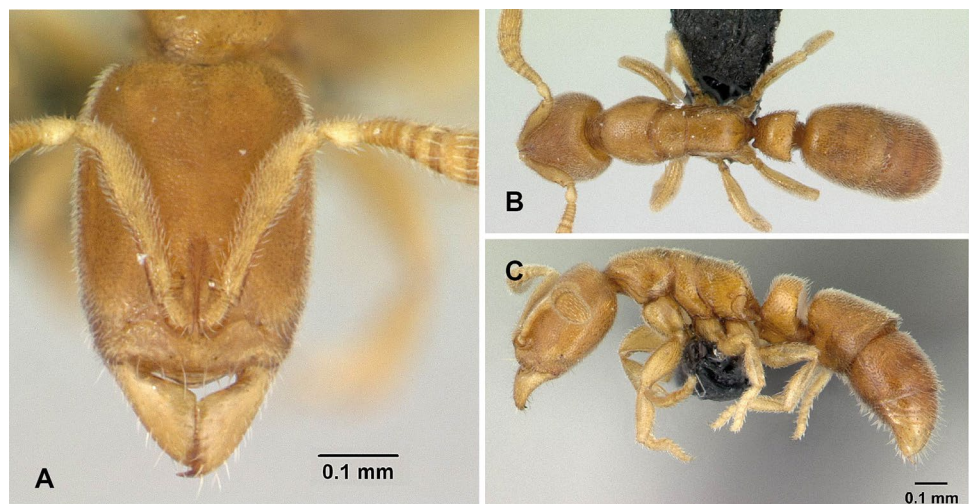
Head Small, Head width (CWb): 462 μm [447, 478]; and conspicuously longer than broad, Cephalic index (CI): 77 [75, 78]. Frontal lobe distance vs. absolute cephalic size (FRS/CS): 0.25 [0.24, 0.26]. Head dorsum coarsely punctate. Anterior clypeal margin with an inconspicuous median indentation or notch. Eyes absent, occasionally 1 to 3 facets

are visible. Scape longer, Scape index (SI): 93 [90, 96]; when laid straight back from its insertion the apex falls near, but does not reach, the midpoint of the posterior margin in full-face view. Mandibles with 3 distinct apical teeth occupying half of the masticatory border.

Mesosoma Pronotal-mesonotal articulation present and developed across dorsum of mesosoma; always a strongly defined groove that conspicuously interrupts the surface. Metanotal groove absent. Dorsal surface of mesosoma coarsely punctate, dull. Lateral surface of pronotum finely punctate or areolate, dull. Mesopleural sculpture punctate to areolate, dull; meta-pleural sculpture longitudinally rugulose, partly shiny.

Petiole Petiolar node squamiform, large; petiole width vs. absolute cephalic size (PEW/CS): 0.55 [0.52, 0.58]; broader than long, (PEW/PEL): 1.34 [1.31, 1.41]; anterior face of node widely rounded, the sides usually very divergent posteriorly. Petiolar node in profile relatively high and moderately

Fig. 5 A–C *Ponera swezeyi* non-type worker (CASENT0135035). Head in full-face view (A), dorsal view of the body (B), lateral view of the body (C)



long, with a rather short and weakly convex dorsum. Dorsal region of petiole smooth to finely punctate, shiny. Anterior and posterior faces of node usually clearly convergent dorsally. Lateral surface of node meets the posterior surface in a rounded transition without a cuticular ridge or transverse carina. Subpetiolar process absent to moderately developed, if present, its apex forms a right angle.

Differential diagnosis

This species cannot be confused with other Malagasy *Ponera* species due to its dark brown color in contrast to the yellowish brown color of *P. petila* and *P. swezeyi* and its larger body size relative to the latter two species. Worldwide, this species most resembles the Indo-Australian *P. clavicornis* Emery (1900), but *P. adumbrans* sp. n. clearly differs from its congener in shape, color, and size characteristics. The two species exhibit non-overlapping ranges of morphometric ratios: *P. adumbrans* has a shorter head (cephalic index, CI: 77 [75, 78]) than *P. clavicornis* (CI: 81–85, see Taylor, 1967), and the scape of *P. adumbrans* is longer (SI: 93 [90, 96]) than that of *P. clavicornis* (SI: 80–89, see Taylor, 1967).

Distribution

Known only from the Seychelles islands Silhouette and Mahé.

Ponera petila Wilson (1957)

Ponera petila Wilson (1957: 368)

Ponera bableti Perrault (1993: 334 **syn. nov**)

(Fig. 4A–C, Table 2)

Type material investigated

Holotype of *Ponera petila* Papua New Guinea: MCZ-ENT00030124, lower Busu River, Huon Pen. N.G., [lower rainforest,], V-/0-55 #999, E.O. Wilson, (1w, MCZ, MCZ-ENT00030124) [examined].

Holotype and paratypes of *Ponera bableti* Fangataufa, “mers” 1987 “unintelligible text”, *Ponera bableti* Perrault (1993) det T. Ramage, (6w, MNHN, holotype: EY25377, paratypes EY25378, EY25379, EY25327, EY25328, EY25329) [Examined, measured by Quentin Rome based on the same character recording protocol defined in this paper, MNHN].

Other material morphometrically examined

Mauritius (8w), Seychelles (17w), Society Islands, French Polynesia (1w). For the full list of material, see Supplementary Table S1.

Description of workers

General appearance Small species, absolute cephalic size (CS): 386 μ m [372, 393]. Body color yellow to light brown. Body concolorous.

Head Small, Head width (CWb): 333 μ m [323, 339]; and conspicuously longer than broad, Cephalic index (CI): 76 [74, 77]. Frontal lobe distance vs. absolute cephalic size (FRS/CS): 0.22 [0.20, 0.24]. Head dorsum coarsely punctate. Anterior clypeal margin lacks a median indentation or notch. Eyes absent, occasionally 1 to 3 facets are visible. Scape short, Scape index (SI): 85 [82, 88]; when laid straight back from its insertion the apex falls far short of the midpoint of the posterior margin in full-face view. Mandibles with 3 distinct apical teeth occupying half of the masticatory border.

Mesosoma Pronotal-mesonotal articulation present and moderately developed across dorsum of mesosoma; sometimes a weakly defined shallow groove visibly interrupts the surface. Metanotal groove inconspicuous, rarely absent. Dorsal surface of mesosoma punctate, slightly shiny. Lateral surface of pronotum finely punctate, ventral area smooth and shiny. Mesopleural sculpture inconspicuous, medial and ventral part smooth and shiny; metapleural sculpture inconspicuously punctate, centrally smooth and shiny.

Petiole Petiolar node not squamiform, moderately large; petiole width vs. absolute cephalic size (PEW/CS): 0.50 [0.48, 0.53]; broader than long, (PEW/PEL): 1.35 [1.28, 1.45]; anterior face of node bluntly rounded, the sides usually slightly divergent posteriorly. Petiole node in profile moderately low and long, with a relatively short and weakly convex dorsum. Dorsal region of petiole smooth to finely punctate, shiny. Anterior and posterior faces of node usually weakly convergent dorsally, sometimes nearly parallel. Lateral surface of node meets the posterior surface in rounded transition without cuticular ridge or transverse carina. Subpetiolar process developed, its apex forms a right angle (rarely acute angle).

Differential diagnosis

In the Malagasy region, *P. petila* can only be confused with *P. swezeyi*; relevant diagnostic characters are discussed

under diagnosis of the latter species. Worldwide, *P. petila* workers differ in size from all other species but *P. szentivanyi* Wilson, (1957). The latter species is reported to have a wider head (CWb: 340 μ m, see Wilson, 1957) than *P. petila* (CWb: 333 μ m [323, 339]), but the scape index (SI) of the *P. szentivanyi* is considerably larger (SI: 94) than that of *P. petila* (SI: 85 [82, 88]).

Distribution

In the Malagasy region, this species is distributed in Mauritius and Seychelles. Outside the region, it is known from Papua New Guinea and the Society Islands (French Polynesia).

Ponera swezeyi (Wheeler, 1933)

Pseudocryptopone swezeyi Wheeler (1933: 16)

Ponera swezeyi: Wilson (1957)

(Fig. 5 A–C, Table 2)

Type material investigated

Syntypes Hawaii, USA: CASENT0249111, Is. Oahu, R. H. Van Zwaluwenburg, (1w, BPBM, CASENT0249111) [examined]; **MCZ-ENT00593416**, Is. Oahu, R. H. Van Zwaluwenburg, (3w, MCZC, MCZ-ENT00593416) [examined];

Other material morphometrically examined

Comores (16w), Madagascar (47w), Mauritius (5w), Mayotte (8w), Queensland, Australia (5w), Réunion (7w) Seychelles (11w). For the full list of material, see Supplementary Table S1.

Description of workers

General appearance Small species, absolute cephalic size (CS): 358 μ m [343, 376]. Body color yellow to light brown. Body concolorous.

Head Small, Head width (CWb): 306 μ m [291, 323]; and conspicuously longer than broad, Cephalic index (CI): 74 [70, 79]. Frontal lobe distance vs. absolute cephalic size (FRS/CS): 0.21 [0.19, 0.23]. Head dorsum coarsely punctate. Anterior clypeal margin lacks a median indentation or notch. Eyes absent, occasionally 1 to 3 facets are visible. Scape short, Scape index (SI): 83 [79, 87]; when laid straight back from its insertion the apex falls far short of the midpoint of the posterior margin in full-face view. Mandibles with 3 distinct apical teeth occupying half of the masticatory border.

Mesosoma Pronotal-mesonotal articulation present and moderately developed across dorsum of mesosoma; sometimes a weakly defined shallow groove visibly interrupts the surface. Metanotal groove inconspicuous, rarely absent. Dorsal surface of mesosoma punctate, slightly shiny. Lateral surface of pronotum finely punctate, ventral area smooth and shiny. Mesopleural sculpture inconspicuous, medial and ventral part smooth and shiny; metapleural sculpture inconspicuously punctate, centrally smooth and shiny.

Petiole Petiolar node not squamiform, moderately large; petiole width vs. absolute cephalic size (PEW/CS): 0.47 [0.43, 0.51]; slightly broader than long, (PEW/PEL): 1.20 [1.08, 1.35]; anterior face of node bluntly rounded, the sides usually slightly divergent posteriorly. Petiole node in profile relatively low and long, with a moderately long and weakly convex dorsum. Dorsal region of petiole smooth to finely punctate, shiny. Anterior and posterior faces of node usually weakly convergent dorsally, sometimes nearly parallel. Posterior surface of node bearing cuticular ridge or a fine vertical carina demarcating the posterior and lateral surfaces. Subpetiolar process well developed, its apex forms an acute angle.

Differential diagnosis

In the Malagasy region, this species can only be confused with *P. petila*, but petiole characters help distinguish these two species: in *P. swezeyi*, the anterolateral and posterior surfaces of petiolar node meet in sharp vertical ridges, while the lateral and posterior petiolar surfaces of *P. petila* meet in a rounded transition. Size of workers may also provide clues for identification: head width (CWb) of *P. swezeyi* is smaller 306 μ m [291, 323]) than that of *P. petila* (333 μ m [323, 339]). Worldwide, *P. swezeyi* is the smallest species within the *tenuis* group. Size variance detected in a relatively large pool ($n=105$) of *P. swezeyi* workers was found negligible (CWb: 291 μ m to 323 μ m) allowing for separation of *P. swezeyi* workers from that of other species. Only one species, *P. szaboi* Wilson, (1957) overlapping with *P. swezeyi* (CWb: 306 μ m [291, 323]), but the petiole of *P. swezeyi* species is considerably wider (PNI: 77–83) than that of *P. szaboi* (PNI: 61–65, see Taylor, 1967).

Distribution

Ponera swezeyi is known to occur in the eastern coast of Madagascar, Comoros, Mauritius, Mayotte, Reunion, and Seychelles in the Malagasy region. Moreover, populations are known from Queensland (northeastern Australia), Hawaii, Christmas Island, Japan (Fiji Island), Hawaii, Japan (Ogasawara Islands), Samoa, Solomon Islands, and Tanzania (Fisher & Bolton, 2016; Leong et al., 2019).

Discussion

Altogether, three *Ponera* species were recognized in the Malagasy region via an integrative taxonomic protocol, *P. petila* Wilson, 1957, *P. adumbrans* sp. n., and *P. swezeyi* (Wheeler, 1933). Two of them (*P. petila* and *P. swezeyi*) belong to the predominantly Indo-Australian *Ponera tenuis* group. The third species, *P. adumbrans* sp. n., is morphologically close to the Indo-Malayan *P. clavicornis*. The two *tenuis* group species occupy both natural and semi-natural environments from low to high altitudes. The two *tenuis* group species occupy both natural and semi-natural environments from low to high altitudes. *Ponera swezeyi* is found widely in Madagascar and adjacent islands, while *P. petila* is known only from Mauritius and Seychelles. The *P. adumbrans* is endemic to Seychelles and typically occurs in higher altitudes.

The Indo-Australian origin of each Malagasy *Ponera* species has been supported by two lines of evidence: (a) Africa does not harbor native *Ponera* fauna (Fisher & Bolton, 2016), and no fossil records are known from the continent; (b) two out of the known three Malagasy *Ponera* species belong to the Indo-Australian *Ponera tenuis* group and are identical with two recent species, *P. petila* and *P. swezeyi*, while the third species, *P. adumbrans*, is morphologically closely related to the Papua New Guinean *P. clavicornis* Emery (1900), which further strengthens the Indo-Australian connection.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s13127-023-00610-1>.

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Data availability All data generated or analyzed during this study are included in this published article (Supplementary Tables S2 and S3).

Declarations

Conflict of interest The authors declare no competing interests.

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