Delimitation of tribes in the subfamily Leptanillinae (Hymenoptera: Formicidae), with a description of the male of Protanilla lini Terayama, 2009

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Abstract

The subfamily Leptanillinae Emery, 1910 (Hymenoptera: Formicidae) is a clade of cryptic subterranean ants, which is restricted to the tropics and warm temperate regions of the Old World. Due to acquisition bias against the minute and hypogaecic workers, most known leptanilline specimens are male, with four genera described solely from males. The sexes have been associated in only two out of 69 described species, meaning that redundant naming of taxa is likely. Herein, the male of Protanilla lini Terayama, 2009 is associated with corresponding workers collected on Okinawa-jima, Japan, by means of genome-scale data, allowing the first published description of male ants belonging to the Anomalomyrmini Taylor, 1990, one of the two established tribes within the Leptanillinae. The first male-based diagnoses of these tribes are provided, based on a phylogeny of the Leptanillinae inferred from ultra-conserved elements using maximum-likelihood and Bayesian inference, along with a dichotomous key to all described male-based species within the Leptanillinae and to undescribed male morphospecies sequenced in this study. With molecular data enabling the association of separately collected sexes and phylogenomic inference contextualizing morphological observations, the parallel taxonomy that afflicts this enigmatic group of ants can begin to be resolved.

Key words: Leptanillinae, Anomalomyrmini, male morphology, phylogenomics.

Introduction

The ant subfamily Leptanillinae (Hymenoptera: Formicidae) Emery, 1910 is a group of small, hypogaecic ants largely restricted to the Old World tropics and subtropics. Little is known of the biology of these ants; the few species for which detailed behavioral observations exist appear to be specialist predators of geophilomorph centipedes (Masuko 1990, Hsu & al. 2017). Known gynes of Leptanilla Emery, 1870 are dichtadiiform (e.g., Emery 1870, Masuko 1990, López & al. 1994), whereas those of other genera are alate or ergatoid (Bolton 1990a, Baroni Urbani & de Andrade 2006, Borowiec & al. 2011, Billen & al. 2013, Chen & al. 2017, Hsu & al. 2017). The position of the lepantanillines within ant phylogeny has been extensively debated (Moreau & al. 2006, Rabeling & al. 2008, Kück & al. 2011, Moreau & Bell 2013, Borowiec & al. 2019), but molecular evidence indicates that the Leptanillinae are sister to Martialis heureka Rabeling & Verhaagh, 2008, with these two taxa constituting a clade that is sister to all other crown-group ants (Borowiec & al. 2019).

The subfamily Leptanillinae is formally divided into the tribes Leptanillini and Anomalomyrmini (Bolton 1990a), with the monotypic Opamyrrma Yamane & al., 2008 unplaced to tribe and sister to the remainder of the Leptanillini (Ward & Fisher 2016, Borowiec & al. 2019). The Leptanillini Emery, 1910 consist of the genus Leptanilla Emery, 1870 (46 spp.; Bolton 2020), which is known from both sexes, and four genera known only from males: Scyphodon Brues, 1925; Phaulomyrma Wheeler & Wheeler, 1930; Noonilla Petersen, 1968; and Yavnella Kugler, 1987 (Bolton 1990a). Of these male-based genera only Yavnella is not monotypic (2 spp.; Bolton 2020). The Anomalomyrmini Bolton, 1990 include the genera Anomalomyrma Taylor, 1990 (3 spp.; Bolton 2020) and Protanilla Taylor, 1990 (13 spp.; Bolton 2020) (Bolton 1990a, Hsu & al. 2017). Borowiec & al. (2019) extensively sampled molecular data from across the Leptanillinae, attempting to resolve basal divergences within the Formicidae with model-based inference from 11 nuclear loci. Otherwise, only Ward & Fisher (2016) have made explicitly phylogenetic contributions to our understanding of this clade – an understanding hampered by dissociation of male and worker specimens.
Due to collection bias most known leptanilline material is male, including most of the terminals sampled by Borkowec & al. (2019). Males attributable to the Anomalomyrmini by molecular data have been discovered and sequenced but remain undescribed and unassociated with female counterparts (Boudinot 2015, Borkowec & al. 2019). Within the Leptanillini, the only species for which both sexes have been identified is Leptanilla japonica Baroni Urbani, 1977 (Ogata & al. 1995); 28% of all described Leptanilla spp. are known only from males (Bolton 2020). The male of Opamyrrha has also been described (Yamada & al. 2020).

The description of ant taxa based solely upon males results in parallel taxonomy, culminating in taxonomic “confusion rather than enlightenment” (Bolton 1990a). Therefore, any phylogenetic reclassification of the Leptanillinae that clarifies the interrelationships of the male-based and worker-based taxa must necessarily integrate morphological and molecular data for both sexes, preferably with association of sexes for each species. To this end, the aim of the current paper is to 1) describe the male of Protanilla lini Terayama, 2009 – a species heretofore known from workers and queens – from material collected on Okinawa-jima, Japan, associated with the conspecific worker using genome-scale data; and 2) provide the first formal morphological male-based definitions of the Anomalomyrmini and Leptanillini, with these definitions informed by inference from phylogenomic data.

Materials and methods

Material sampled: Morphology. The following material was physically examined: a series of four male Protanilla (M. Yoshimura det.), inferred to be Protanilla lini (see below), collected on Okinawa-jima; a series of four males belonging to an undescribed species of Protanilla, resembling the inferred P. lini, collected in northern Vietnam; and 37 additional male morphospecies attributed to the Leptanillinae according to the definition provided by Boudinot (2015). When necessary, images of male Leptanillinae already available on AntWeb (www.AntWeb.org) were used. A worker representative of P. lini (S. Iiniyama & T. Yoshida det.) collected with a Sea, Air & Malaise (SLAM) trap in 2016 on Okinawa-jima was also examined and used for DNA sequencing, along with one of the males.

All specimens were examined with a Leica MZ75 compound microscope, except for the male of Martialis heureka, for which observations were derived from Boudinot (2015). Specimens were imaged using a JVC KY-F75 digital camera and color photographs were compiled from these with the Syncroscopy AutoMontage Program (v. 5.02.0096). Scanning electron microscopy was undertaken using a Hitachi TM4000 tabletop microscope. Material is deposited in the following repositories: the Okinawa Institute of Science and Technology, Onna, Okinawa, Japan (OIST); the Bohart Museum of Entomology, University Of California, Davis, CA, USA (UCDC); the California Academy of Sciences, San Francisco, CA, USA (CASC); the California State Collection of Arthropods, Sacramento, CA, USA (CSCA); the Lund Museum of Zoology, Lund, Sweden (MZLU); and the Australian National Insect Collection, Canberra, Australia (ANIC).

Phylogenomics. Phylogenomic data were acquired from 39 specimens of Leptanillinae, along with Martialis heureka as an outgroup (Tabs. 1 - 2), using ultra-conserved elements (UCEs) (see below, “Genomic Data Generation & Processing”). This approach was used as an alternative to barcoding methods, the reliance of which upon a single locus (usually cytochrome oxidase c, subunit 1, COI) can produce spurious phylogenetic inferences at a macroevolutionary scale (Talavera & Vila 2011, Caravas & Friedrich 2012, Simon & Hadrys 2013, Chen & al. 2014) unless topologies are constrained by prior inference from other datasets (Zhou & al. 2016). Mitogenomes can be retrieved from raw data generated in UCE sequencing; if needed for this purpose, the raw reads used for this study are publicly available (Tab. 2).

The phylogenomic sampling of the Leptanillinae in this study is the first to encompass both described species of the male-based genus Yavnella (or morphospecies with close affinity to them) and morphospecies attributable to the monotypic male-based genus Noonilla. Noonilla zhg-my04 closely resembles Noonilla copiosa Petersen, 1968 but cannot be identified as such without better intraspecific sampling. The remaining four sampled Noonilla morphospecies diverge from the habitus of N. copiosa in that the mesoscutum is flattened relative to that species, resembling Scyphodon anomalum in this respect; however, their highly distinctive genital morphology conforms to that of Noonilla as described by Petersen (1968), and will be described in future publications (Z. Griebenow, G. Fischer & E. Economo, unpubl.).

Genomic data generation and processing: DNA was extracted non-destructively using a DNeasy Blood & Tissue Kit (Qiagen Inc., Valencia, CA) according to manufacturer instructions. Genomic concentrations were quantified for each sample with a Qubit 2.0 fluorometer (Life Technologies Inc., Carlsbad, CA). Phylogenomic data were generated using the ant-specific version of the UCE probe set hym-v2 (Branstetter & al. 2017), with libraries being prepared and target loci enriched using the protocol of Branstetter & al. (2017). Enrichment success and size-adjusted DNA concentrations of pools were assessed using the SYBR FAST qPCR kit (Kapa Biosystems, Wilmington, MA) and all pools were combined into an equimolar final pool. Depending on the lane in question, the contents of this final pool were sequenced on an Illumina HiSeq 2500 at the High Throughput Genomics Facility, University of Utah, Salt Lake City, UT; or an Illumina HiSeq 4000 at Novogene, Sacramento, CA. Sequence Read Archive (SRA) accession numbers for all raw reads are presented in Table 2.

The FASTQ output was demultiplexed and cleansed of adapter contamination and low-quality reads using illumiprocessor (Faircloth 2013) in the PHYLUCe package. Raw reads were assembled with trinity v. 2013-02-25 (Grabherr & al. 2011) or with SPAdes v. 3.12.0
Tab. 1: Collection data for all terminals sequenced in this study. Taxon names are followed with Ward Laboratory extraction codes.

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<th>Form</th>
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<th>Voucher type</th>
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Tab. 2: Summary statistics for final 368,656-bp UCE alignment, along with iTru primer sequences and Sequence Read Archive (SRA) accession numbers. Taxon names are followed with Ward Laboratory extraction codes.

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(Bankevich & al. 2012). All PHYLUCE commands hereinafter are cited from Faircloth (2016). Species-specific contig assemblies were obtained with the ant-specific hym-vs2 probe set (Branstetter & al. 2017) using phyluce\textunderscore assembly\textunderscore match\textunderscore contigs\textunderscore to\textunderscore probes.py (min\textunderscore coverage=80), with min\textunderscore identity = 90 to minimize the influence of possible contamination; and a list of UCE loci shared across all taxa was generated using phyluce\textunderscore assembly\textunderscore get\textunderscore match\textunderscore counts.py, and separate FASTA files for each locus were created using these outputs. These sequences were aligned separately by locus using MAFFT L-INS-i (Katoh & Tom 2008), rather than the default version of MAFFT implemented in phyluce, implemented with the command phyluce\textunderscore assembly\textunderscore seqcap\textunderscore align.py. These sequences were then trimmed with Gblocks (Castresana 2000) as implemented by the wrapper script phyluce\textunderscore assembly\textunderscore get\textunderscore gbblocks\textunderscore trimmed\textunderscore alignment\textunderscore from\textunderscore untrimmed.py (settings: b1 = 0.5, b2 = 0.5, b3 = 12, b4 = 7). Alignment statistics for the output FASTA files were calculated with phyluce\textunderscore align\textunderscore get\textunderscore align\textunderscore summary\textunderscore data.py. Finally, a dataset that was 90% complete with respect to taxon coverage per locus was generated using the script phyluce\textunderscore align\textunderscore get\textunderscore only\textunderscore loci\textunderscore with\textunderscore min\textunderscore taxa.py, consisting of 580 loci. The final alignment was then concatenated and converted to PHYLP format with phyluce\textunderscore align\textunderscore format\textunderscore nexus\textunderscore files\textunderscore for\textunderscore raxml and was 368,656 bp in length, with 19.07% missing data, 135,832 parsimony-informative sites, and mean locus length being 634 bp. This alignment, along with the partition scheme, is available on Dryad (doi: 10.25338/B8490T). Summary statistics for this alignment were computed with the SWSC partitioning algorithm, consisting of four Metropolis-coupled continuous-time Markov chains (three of them “heated”, with an increment of 0.5). Topology was considered converged once the average standard deviation of split frequencies (ASDSF) equaled 0.05. Apparent convergence of the Markov-chain Monte Carlo (MCMC) with respect to continuous parameters was assessed with Tracer v. 1.7.1 (Rambaut & al. 2018). The output of these Bayesian analyses is available on Dryad (doi: 10.25338/B8490T).

Phylogenetic inference: Evolutionary processes operating on ultra-conserved elements and their flanking regions vary due to differing constraints, both between loci and among sites within a single locus (Tagliacollo & Lanfear 2018). Failing to accommodate this variation (i.e., model misspecification) can result in erroneous inferences (Lanfear & al. 2014; Kainer & Lanfear 2015). Using the corrected Akaike Information Criterion (AICc) Tagliacollo & Lanfear (2018) found that for six empirical phylogenomic datasets, partitioning drastically improved model fit. Therefore the final alignment in this study was partitioned according to, and within each, UCE locus using the analysis.py script of PartitionUCE (available at https://github.com/Tagliacollo/PartitionUCE/tree/master/scripts) using the SWSC partitioning algorithm, with site entropy as the nucleotide property by which data blocks were derived (SWSC-EN; Tagliacollo & Lanfear 2018). Substitution models were then selected for these partitions using ModelFinder (Kalyaanamoorthy & al. 2017) in IQ-Tree v. 1.4.2-beta (Nguyen & al. 2015), with the AICc as test statistic, and only the top 20% of partition schemes considered using the relaxed hierarchical clustering algorithm (Lanfear & al. 2014); all substitution models were considered, with the exception of those with I + G extensions (Yang 1996). The resulting scheme consisted of 1221 partitions.

The phylogeny of the Leptanillinae was inferred using IQ-Tree v. 1.6.10 (Nguyen & al. 2015) on the CIPRES Science Gateway (v. 3.3) (Miller & al. 2010) with 1,000 ultrafast bootstrap replicates (Hoang & al. 2018), both unpartitioned and using the partition scheme (Chernomor & al. 2016) inferred using the SWSC-EN algorithm and ModelFinder. Bayesian inference was performed in ExaBayes (Aberer & al. 2014) using two cloned analyses on the CIPRES Science Gateway with the same partitioning scheme as above, but with GTR + G imposed as the substitution model across all partitions. Each analysis involved two runs, each proceeding for 240,000 generations and consisting of four Metropolis-coupled continuous-time Markov chains (three of them “heated”, with an increment of 0.5). Topology was considered converged once the average standard deviation of split frequencies (ASDSF) equaled 0.05. Apparent convergence of the Markov-chain Monte Carlo (MCMC) with respect to continuous parameters was assessed with Tracer v. 1.7.1 (Rambaut & al. 2018). The output of these Bayesian analyses is available on Dryad (doi: 10.25338/B8490T).

Measurements and indices: Out of all terminals sampled in this study for which males are known, Protanilla zhg-vn01 is the most closely related to P. lini (Figs. 1, 2). Therefore, morphometric comparisons were made between males of these two lineages (Tabs. 3 - 4). Unlike the examined males of P. lini all examined Protanilla zhg-vn01 males were syntopic, and so less morphometric variation among them is expected than in examined P. lini. Protanilla zhg-vn01 material examined. CASENT0106382, CASENT0842613, CASENT0842655-6. Vietnam: Vinh Phúc, Tam Đảo National Park (21.46667° N, 105.65° E), 1200 m elevation, 19 - 22.VI.2014, leg. M. Hauser and N. von Ellenreider, 4 males (CSCA).

Measurements.

DPW = Dorsal Petiole Width, maximum width of the petiole measured in dorsal view

EL = Eye Length, maximum measurable length of compound eye parallel to anteroposterior axis of head

EW = Eye Width, maximum measurable length of eye parallel to dorsoventral axis of head

FrW = Frons Width, the shortest distance between the medial margins of the compound eyes measured in full-face view

HL = Head Length, maximum length of head in full-face view from anterior clypeal margin to posterior head margin between lateral ocelli, ignoring distance which ocelli project

LF1 = First Funicular Segment Length, maximum length of 1st funicular segment (pedicel) in dorsal view (Ward 1985)

LF2 = Second Funicular Segment Length, maximum length of 2nd funicular segment (most basal flagellomere) in medial view

LOD = Lateral Ocellus Length, maximum diameter of lateral ocellus with head oriented such that anterior and posterior lateral ocellus margins are in same plane of focus

MFL = Metafurum Length, maximum length of metafurum in profile view
**Fig. 1:** Maximum-likelihood phylogeny of the Leptanillinae based upon ultra-conserved elements. Non-parametric bootstrap values are given for both unpartitioned and partitioned analyses, respectively, where non-parametric bootstrap values < 100. (A) *Protanilla lini* (CASENT0011097); (B) *Yavnella TH08* (CASENT022755; Shannon Hartman, image courtesy of AntWeb, www.AntWeb.org); (C) *Leptanilla zhg-bt01* (CASENT084612). Bootstrap support = 100 unless otherwise noted.

**Tab. 3:** Metrics useful in distinguishing male *P. lini* and *Protanilla zhg-vn01*, whether directly or via indices derived from them. *P. lini* = pale gray; *Protanilla zhg-vn01* = dark gray. All measurements given in millimeters.

<table>
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<tr>
<th>Specimen identifier</th>
<th>HL</th>
<th>FrW</th>
<th>SL</th>
<th>LF1</th>
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<th>EW</th>
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<table>
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<th>DPW</th>
<th>PTH</th>
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<th>TW4</th>
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<td>N/A</td>
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MOD = Median Ocellus Width, maximum diameter of median ocellus in full-face view
PFL = Profemur Length, maximum length of profemur in profile view
PTH = Petiole Height, maximum width of the petiole in profile view along dorsoventral axis.
PTL = Petiole Length, length of petiole in profile view along anteroposterior axis from inflection point of petiolar presclerites (the articulatory surfaces) to most posterior point of posterior margin
SL = Scape Length, maximum length of scape in medial view, excluding condylar neck

TW3 = Width of Tergite III, maximum width of abdominal tergite III measured in dorsal view orthogonal to dorsal vertex of abdominal tergite III
TW4 = Width of Tergite IV, maximum width of abdominal tergite IV measured in dorsal view orthogonal to dorsal vertex of abdominal tergite IV

Indices.

CS = Cephalic Size (FrW + HL) / 2
FI = Femora Index PFL / MFL × 100
PTI = Petiole Index PTH / PTL × 100
SEI = Scape-Eye Index EL / SL × 100
SI = Scape Index SL / FrW × 100
TI1 = Tergal Index 1 TW3 / TW4 × 100

All continuous morphometric data for \textit{P. lini} and \textit{Protanilla zhg-vn01} that are of discriminatory use are summarized in Tables 3–4.

Terminology. Morphological terminology follows that promulgated by the Hymenoptera Anatomy Ontology project (Yoder et al., 2010), except for terminology for the male genitalia, which follows Boudinot (2018), and terms pertaining to metasomal sclerites, which are derived from Bolton (1990b) and Keller (2011). Hamuli are termed according to the nomenclature of Basibuyuk & Quicke (1997). Terminology for sculpturation and setation follows Harris (1979) and Wilson (1955: p. 23, Fig. 3), respectively.

Results

Phylogenomic inference: The maximum-likelihood (ML) phylogeny of the Leptanillinae presented here (Fig. 1) recovers the same topology whether based upon partitioned or unpartitioned genome-scale UCE alignments, and corroborates the previous molecular phylogenetic studies of the subfamily (Ward & Fisher 2016, Borowiec & al. 2019). Non-parametric bootstrap values are maximal throughout, except for two internal nodes (Fig. 1): 1) \textit{Anomalomyrma boltoni} Borowiec & al., 2011 as sister to the anomalomyrmine subclade containing \textit{P. lini}; and 2) the sister-group relationship of \textit{Leptanilla} GR02 and \textit{Leptanilla zhg-bf01}. \textit{Opamyrna} is recovered as sister to all remaining Leptanillinae, which bifurcate into two well-supported nodes corresponding to the tribes Leptanillini and Anomalomyrmini. Bayesian inference with
ExaBayes under the same partitioning scheme as the ML analyses corroborates the ML phylogeny exactly, but with Bayesian Posterior Probabilities (BPP) maximal across all nodes.

These phylogenies also indicate that the Protanilla lini worker and putative male are conspecific, given the lack of other putative anomalomyrmine male morphospecies on Okinawa-jima (E. Economo, pers. comm.). Such a conclusion is consistent with preliminary maximum-likelihood inference from smaller molecular datasets (E. Economo, pers. comm.). Protanilla JP01, from the northern Ryukyu Islands, is robustly recovered sister to the Okinawan P. lini, and, given its geographical proximity to Okinawa and the brevity of its subtending branch, can be provisionally judged conspecific with P. lini as well. The node corresponding to the Anomalomyrmex itself consists of two major clades, both of which include worker and male material putatively assigned to Protanilla. Anomalomyrma boltoni is sister to the clade including P. lini, but with comparatively weak support, contrasting with the relationship inferred by Borowiec & al. (2019), who recovered A. boltoni sister to the other major anomalomyrmine clade. If the assignment of these undescribed worker and male morphospecies to Protanilla is accurate, then Anomalomyrma renders Protanilla paraphyletic (cf. Borowiec & al. 2019), and should be synonymized with the latter genus. Alternatively, Protanilla in the current sense could be divided into at least two genera (see Discussion).

While sampling of the tribe Leptanillini in this study is more extensive than that of Borowiec & al. (2019), and the loci and inferential framework different, the results of these two studies are largely congruent, with the most conspicuous difference being the placement of Anomalomyrma within the Anomalomyrmini (see Discussion). The two described species of Yavnella are nested within a clade consisting of morphospecies provisionally attributed to Leptanilla, and a single male specimen incorrectly assigned to the male-based genus Phaulomyrma (Z. Griebenow, in review); this clade is distinguishable based upon male morphology (e.g., posterodorsal propodeal outline concave in profile view, noted by Kugler (1986) as contrasting Yavnella with Noonilla), and is hereafter referred to as Yavnella s.l. The sister clade to Yavnella s.l. contains all the morphospecies attributed to Noonilla, itself monophyletic, nested within an assemblage of specimens provisionally identified as Leptanilla; among these is Leptanilla revelieri Emery, 1870, the type species of the genus. This clade is termed Leptanilla s.l. Examination of male morphological diversity across this node, informed by phylogenetic data, is in progress and will clarify the boundaries and status of the four male-based genera currently placed within the Leptanillini.

**Diagnosis of leptaniline tribes based upon the male sex:** Boudinot (2015) acknowledged the existence of male Leptanillinae that were inferred to be anomalomyrmines based upon molecular data, as later published in Borowiec & al. (2019). Due to an error on AntWeb that linked specimens with the wrong images, the male leptanilline in Figs. 6F and 10A of Boudinot (2015), said to be that of Protanilla TH01 (CASENT0119776), is actually that of CASENT0119531 (named Leptanilla TH02 by Borowiec & al. 2019), which closely resembles the male-based genus Yavnella (Leptanillini; Kugler 1986, Borowiec & al. 2019). As noted by Boudinot (2015: p. 14), this morphospecies is notable among the Formicidae for the complete loss of metasomal petiolation.

CASENT0119531 is called Yavnella TH02 in this paper (Tabs. 1 - 2). Examination of CASENT0119776 demonstrated that this specimen in fact closely resembles other male Anomalomyrmini.

Moreover, the specimen in Figure 12D of Boudinot (2015), identified as Protanilla indet. (CASENT0178838) (Fig. 3), is qualitatively like five male morphospecies sequenced in this study (Leptanilla zhg-my02-05, -id01) and robustly recovered within a clade corresponding to the Leptanillini. The rationale for labeling CASENT0178838 as Protanilla is not given (Boudinot 2015), nor was it noted anywhere outside of peer review (B. Boudinot, pers. comm.). Thus, there is no justification for regarding CASENT0178838 as Protanilla. CASENT0178838 and those five male-based morphospecies provisionally assigned to Leptanilla exhibit such peculiar attributes as prothoracic combs of robust setae and ventrolateral setose metasomal processes. The latter are unique among the whole of the Hymenoptera (L. Kimsey & B. Boudinot, pers. comm.), and were previously hypothesized (Boudinot 2015) to be 1) filiform extensions of the gonocoxae sensu Schulmeister 2001) or 2) pygostyles.

Therefore, the known phenotypic diversity of male anomalomyrmines is circumscribed relative to what is implied by Fig. 12 of Boudinot (2015). All 5 anomalomyrmine morphotaxa represented by male material that were sampled in this study, including P. lini, consistently contrast with males of the Leptanillini in several respects. The two previously established tribes and Opamyrma hungvuong Yamane & al., 2008 are diagnosed below.

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**Fig. 3:** CASENT0178838 (*Leptanilla* indet.), image courtesy of AntWeb (www.AntWeb.org), photographer April Nobile.
according to these character states, in addition to a male-based definition of the Leptanillinae relative to other Formicidae; character states that are apomorphic within the Leptanillinae are underlined. *Opamyrma* is left unplaced to tribe because there is no comparative basis for defining such a tribe morphologically, given that said tribe would be monobasic. For taxonomic synopses of *Opamyrma*, Leptanillini, and Anomalomyrmini refer to Bolton (2020).

**Subfamily Leptanillinae** EMERY, 1910


**Male Diagnosis** (modified from Boudinot 2015).

1. Mandibles edentate; minute and nub-like, or hypertrophied and spatulate
2. Frontal carinae and lobes absent
3. Anterior clypeal margin without pegs
4. Antenna 13-merous; funiculus filiform to submoniliform
5. Mesopleural sulcus present or absent
6. Metapleural spiracular plate absent
7. Propodeal lobes inconspicuous or absent
8. Metacoxal cavities closed
9. Tibial spur formula 2s,2s; 1s,2(1s,1p); 1s,2s; or 0,1p
10. Metatarsus lacking posterolateral line of dense differentiated setae
11. Pretarsal claws edentate
12. Wing venation Ogata Type IVb
13. Hindwing venation reduced, at most R+Rs and 1A tubular
14. Jugal lobe absent
15. Petiolar tergum not forming anteroventral collar around sternum
16. Helcium axial or infra-axial
17. Abdominal segment IV not vaulted, as long as, or distinctly longer than, more posterior abdominal segments
18. Abdominal spiracles IV-VIII obscured by preceding tergites
19. Posterior margin of abdominal sternite IX with posteromedian process, entire, or with posterolateral processes
20. Pygostyles (i.e., cerci) absent
21. Cupula present or absent
22. Parossiculus and lateropenite (Boudinot 2018) (i.e., cuspis and digitus) distinct or indistinct

**Opamyrma** YAMANE, BUI & EGUCHI, 2008


**Male Diagnosis**.

1. Ocelli not set on tubercle
2. Four maxillary palpomeres
3. Pronotum not prolonged posteriorly
4. Mesoscutum not prolonged posteriorly
5. Notauli present or absent
6. Rs+M and 1m-cu absent in forewing
7. 1A present in hindwing
8. Pterostigma present
9. Petiole distinct, without tergosternal fusion
10. Postpetiole absent
11. Parossiculus and lateropenite distinct; lateropenite club-shaped

**Tribe Leptanillini** EMERY, 1910


Constituent genera: *Leptanilla; Scyphodon; Phaulomyrma; Noonilla; Yavnella*

**Male Diagnosis**.

1. Ocelli, when present, set on tubercle (with exception of *Leptanilla zhg-my05*)
2. One maxillary palpomere
3. Pronotum prolonged posteriorly, or not prolonged posteriorly
4. Mesoscutum prolonged posteriorly, or not prolonged posteriorly
5. Notauli absent
6. Rs + M and 1m-cu absent in forewing
7. 1A absent in hindwing
8. Pterostigma absent
9. Petiole distinct to absent and with tergosternal fusion
10. Postpetiole absent
11. Parossiculus and lateropenite not distinct

**Tribe Anomalomyrmini** TAYLOR, 1990


Constituent genera: *Anomalomyrma; Protanilla*

**Male Diagnosis**.

1. Ocelli not set on tubercle
2. Four maxillary palpomeres
3. Pronotum not prolonged posteriorly
4. Mesoscutum not prolonged posteriorly
5. Notauli present or absent
6. Rs + M and 1m-cu absent in forewing
7. 1A present or absent in hindwing
8. Pterostigma present
9. Petiole distinct, with tergosternal fusion
10. Postpetiole present or absent
11. Parossiculus and lateropenite distinct

**Discussion**

**Delimitation of major lineages within the Leptanillinae:** According to the admittedly “tentative and unsatisfactory” diagnosis of Bolton (1990a), the males of Leptanillinae are identified by 7 co-occurring character states, most of them vague (e.g., “genitalia large to ... hypertrophied; not retractile”) and so of limited utility. None of these character states are unique to the subfamily. Later male-based revision of the Formicidae (Boudinot 2015) recognized the reduction of the propodeal lobe (Boudinot 2015: Fig. 4G) as diagnostic of leptanilline males in combination with other character states.
Male Leptanillini are far better represented in this study than males of their sister group, and correspondingly exhibit morphologies that are not only more diverse, but qualitatively more disparate, than those of anomalomyrmicine males. Even so, there are two morphological characters that reliably separate males of Anomalomyrmini and Leptanillini: the presence / absence of the pterostigma (Figs. 4A - B) and the presence/absence of an ocellar tubercle or swelling (Figs. 5A - B). While better sampling may bring further male leptonilline morphological diversity to light and so necessitate revision of this morphological diagnosis, it is satisfactory given the material available.

Males of the clade to which CASENT0178838 putatively belongs (consisting of Leptanilla zhg-my02 through -05 and zhg-id01) display what resembles a pterostigma with the confluence of Rf and 2s - rs + Rs + 4 - 6 (Fig. 4C), but this condition is likely homoplasious with that seen in the Anomalomyrmini. Pterostigmal condition remains the most useful character for discriminating males of the two tribes: although there are lineages within the Leptanillini with males that have deciduous forewings (pers. obs.) and therefore for which the condition of the pterostigma is unknown, in no known male Anomalomyrmini are the forewings deciduous. Exceptions to the diagnosis of male Leptanillini as possessing an ocellar tubercle do occur, but these either occupy a unique character state in that they lack ocelli entirely (Fig. 6B; Yavnella TH02 and zhg-bt01) or are distinct from the Anomalomyrmini in that the anteromedian ocellus is not orthogonally dorsal to the compound eyes (e.g., Leptanilla zhg-my05; Fig. 7).
This survey of novel male material attributable to the Leptanillinae refines and extends the previous male-based diagnoses of the subfamily. It is apparent that 1) there are 4 maxillary palomeres in all known anomalomyrmine males (and *Opomyrma hungvuong*, in which the palpal formula is 4·2; Yamada & al. 2020), in contrast with the single maxillary palomere of both males (Fig. 8) and workers (Bolton 1990a, Ogata & al. 1995, Boudinot 2015) of the Leptanillini (Kugler 1986 reported a vestigial constriction of the single worker maxillary palomere in *Leptanilla escheri* Kutter, 1948 and *Leptanilla judaica* Kugler, 1987); 2) the mesoscutum is not always flattened and posteriorly extended in the males of Leptanillinae, with *O. hungvuong*, the Anomalomyrmini and *Yavnella* s.l. being the exceptions; and 3) the propodeal lobe is inconspicuous to absent in all leptanilline male morphospecies examined in this study that were not cited by Boudinot (2015: p. 31) and in *O. hungvuong* (Yamada & al. 2020), corroborating the male diagnosis of the subfamily by Boudinot (2015: p. 29).

In the context of male Formicidae, I infer the anomalomyrmine habitus to be plesiomorphic overall relative to that of previously described male Leptanillinae (Petersen 1968, Kugler 1986, Bolton 1990a, Ogata & al. 1995, Boudinot 2015, Yamada & al. 2020), e.g., in the presence of the pterostigma. Micro-CT scans of *Protanilla zhg-vm01* genitalia (Z. Griebenow, G. Fischer & E. Economo, unpubl.) indicate that a reduced ventral cupula (Schulmeister 2001) is present in that morphospecies, contrasting with all males attributable to Leptanillini in which this character can be observed; while the genitalia of other anomalomyrmine male material were not examined by dissection (virtual or otherwise), I predict that the cupula is present in *Protanilla lini* and the Anomalomyrmini as a whole. Yamada & al. (2020: Fig. 15D) report the presence of a similarly reduced cupula in the male of *Opamyrma hungvuong*, the sister group of the remaining Leptanillinae (Yamada & al. 2020: Fig. 14A), and like the Anomalomyrmini, male *O. hungvuong* exhibit a pterostigma and lack an ocellar tubercle; moreover, Yamada & al. (2020) report lack of tergosternal fusion in the petiole of male *O. hungvuong*, the converse of which is here described in male *P. lini*. Tergosternal fusion is present in all males so far included in phylogenomic analysis, in addition to *Scyphodon anomalum* Brues, 1925 and *Noonilla copiosa* Petersen, 1968. The lack of tergosternal fusion in male *O. hungvuong* is likely plesiomorphic within the Leptanillinae. Conversely, the male genitalia of *O. hungvuong* exhibit derived character states not seen in *P. lini*, including medial gonocoxal fusion (Yamada & al. 2020: Figs. 13A - B), which is otherwise known only in *Yavnella* TH03 and some subclades of *Leptanilla* s.l. The pedunculate lateropenes (Yamada & al. 2020: Figs. 13E - F) and penial “spinose lobes” (Yamada & al. 2020: p. 45, Figs. 13A, G) are unparalleled among male Leptanillinae surveyed in this study.

**Delimitation of anomalomyrmine genera with male morphology:** It would be premature to define *Protanilla* based upon the male sex, since molecular evidence indicates that the boundaries of the anomalomyrmine genera require revision. *Protanilla* was described based upon worker material, whereas *Anomalomyrma* was described from a dealate gyne (Bolton 1990a): the initially unknown female castes of both were described subsequently (Baroni Urbani & de Andrade 2006, Borowiec & al. 2011). Mandibular characters were initially used to distinguish these genera (Bolton 1990a, 1994, Imai & al. 2003), but with the description of the worker caste of *Anomalomyrma* it became arguable that abdominal morphology provided more consistent diagnoses (Borowiec & al. 2011).
The first molecular study to include exemplars of both anomalomyrmine genera (Borowiec et al. 2019) recovered the sole sampled *Anomalomyrma* on a long branch nested deep within two well-resolved clades, both identified as *Protanilla*; and irrespective of dataset or statistical framework weakly recovered *Anomalomyrma boltoni* sister to the clade consisting of *Protanilla* TH03, *Protanilla* VN01, and *Protanilla* VN03. However, the ML and Bayesian UCE-based phylogenies presented herein recover *A. boltoni* as sister to the major sampled clade of Anomalomyrmini, which is here found to include *P. lini*, with strong but sub-maximal support under ML inference (Fig. 1) and maximal support under Bayesian inference. Either result implies that the two anomalomyrmine genera could be synonymized, or *Protanilla* divided into at least two genera. Future phylogenomic work will explore the influence of different methods on inference of basal divergences within the Anomalomyrmini – in particular, the effects of including more UCE loci, and filtering those loci in order to compensate for systematic biases in the data.

*Protanilla* TH03 is conspicuously different from the remaining known anomalomyrmine males in the presence of a postpetiole (Fig. 9B) and apical maxillary palpomeres that are subequal in length (Fig. 10B); this is consistent with its position in a clade sister to that containing all other sequenced anomalomyrmine male material and subtended by a long branch. *Protanilla* TH03 is certainly not a male of *Anomalomyrma* under the present definition of that genus, as this undescribed male is embedded within a clade consisting of morphospecies known only from workers that clearly have little morphological affinity to *Anomalomyrma*, and among described anomalomyrmine species appear closely akin to *Protanilla bicolor* Xu, 2002 and *Protanilla gengma* Xu, 2011. The future status of *Anomalomyrma* will depend on scrutiny of morphological differences between the two major anomalomyrmine clades, with reference to both sexes. Sequencing of and
description of further anomalomyrmine males and worker material will clarify the best course for taxonomic revision.

Note on male morphospecies delimitation in Protanilla: While formally describing males of single species without description of males belonging to similar species is limiting, I conclude that it is warranted in this case. Based on phylogenomic data, the males described herein are indubitably conspecific with worker material collected in sympatry and identified as *P. lini*, justifying the description of the males of those species; doing so expands our holomorphological knowledge of *P. lini* (cf. Terayama 2009, Hsu & al. 2017).

We lack confident associations of the sexes in leptanilline species aside from *O. hungvuong*, *P. lini*, and *L. japonica*, but these species can be discriminated from other male morphospecies included in this study by means of the key that follows. Those undescribed morphospecies that were included in Borowiec & al. (2019) are renamed as needed according to provisional conclusions from this and other phylogenetic studies (see Tables 1 - 2). Some Leptanilla spp. have been described only from males (Santschi 1907, 1908, Wheeler & Wheeler 1930, Smith 1953, Petersen 1968, Dlussky 1969, Baroni Urbani 1977, Kugler 1986), and these are included in the key below based upon published descriptions. Undescribed leptanilline material figured in previous publications but not yet sequenced is excluded from this key (Petersen 1968, Baroni Urbani 1977, Ogata & al. 1995, Scupola & Ballarin 2009).

Key to leptanilline male morphotaxa for which phylogenomic data are available and described species for which male morphology is known:

1. Discal cell present (Fig. 9Ai); parossiculus and lateropenite distinct. .............................................
2. Pterostigma present (Fig. 9Ai-ii), wings never deciduous; ocellar tubercle absent. .............................. 3
3. Notauli present. .......................................................................................................................... 4
4. Notauli scrobiculate; postpetiole present (Fig. 9Bi). ................................................................. 5
5. Styilar (i.e., telomeral sensu Schulmeister 2001) apex pointed (Fig. 9Ci). ................................. 6
6. Anterior face of abdominal sternite II nearly perpendicular to craniocaudal axis in profile view (Fig. 9Di); abdominal tergum III slightly narrower than IV in dorsal view (T1 70-77). ...............................................................
Fig. 12: (A) posterodorsal view of penial sclerites in (i) Noonilla zhg-my02 and (ii) Leptanilla zhg-my05 (CASENT0106432), with phallopertamic setae marked; (B) profile view of male head in (i) Noonilla zhg-my04 and (ii) Noonilla zhg-my01. Abbreviations: di = discal cell; st1 = stylus.

- Anterior face of abdominal sternite II gently sloping relative to craniodorsal axis (Fig. 9Dii); abdominal tergum III much narrower than IV in dorsal view (T1I 50-55). ............................................ Protanilla lini Terayama, 2009
7. Propodeum concave in profile view; pronotum and mesoscutum not posteriorly prolonged (Fig. 11Aii). .......................... 8
- Propodeum not concave in profile view; mesoscutum and pronotum posteriorly prolonged (Fig. 11Aii). .......................... 17
8. Gonocoxae entirely fused medially; posterior margin of abdominal sternite IX with median extension. ........................................... Yavnella TH03
- Gonocoxae partly to fully separate medially; posterior margin of abdominal sternite IX entire. .................. 9
9. Ocelli absent (Fig. 6A). .................. Yavnella zhg-bt01
- Ocelli present (Fig. 6B). .................. 10
10. Gonopodite (i.e., paramere sensu Schulmeister 2001) longer than penial sclerites (i.e., pensivalvae). .............................................. 11
- Gonopodite shorter than, or equal in length to, penial sclerites. .............................................. Yavnella zhg-th01
11. Profemur curved (Fig. 11Bi), constricted basally. .. 12
- Profemur not curved nor constricted basally (Fig. 10Bi). .............................................. 13
12. Volsella bifid (Fig. 11Ci), ventral process bifurcated. ........................................... Yavnella TH02
- Volsella bifid (Fig. 11Cii), ventral process entire. ........................................... Yavnella TH02
13. Stylar apex subtriangular, entire. .......................... Yavnella MM01
- Stylar apex tapering, entire or bifid. ................................ 14
14. Posterodorsal margin of gonopodite with vestiture of sparse setae; stylar apex bifurcated. ........................................... Yavnella TH08
- Posterodorsal margin of gonopodite with vestiture of dense setae; stylar apex entire. ................................ 15
15. Volsella bifid; mandible articulated to gena. .......................... Yavnella TH04
- Volsella entire; mandible fused to gena. .......................... Yavnella TH06
16. Internal margins of apical penial cleft distinctly separated (Fig. 11Di); posteroventral gonocoxal margin entire. .................. Yavnella argamani Kugler, 1987
- Internal margins of apical cleft of penial sclerites subparallel (Fig. 11Di); posteroventral gonocoxal margin sinuate. .................................. 17
- Color flavous to pallid; posterior margin of compound eye convex in profile view (Sri Lanka). .......................... Yavnella cf. indica
18. Dorsolateral carina present on propodeum. ........................... Leptanilla palauensis (M.R. Smith, 1953)
- Dorsolateral carina absent from propodeum. .................. 19
19. Stylus lenticular in outline; penial sclerites with medial conjunctiva (Fig. 11E). .............................................. Leptanilla astylina Petersen, 1968
- Stylus not lenticular in outline; penial sclerites usually without medial conjunctiva. ............................ 20
20. Phalotreme surrounded with dense setae (Fig. 12Ai). .......................... 21
- Phalotreme bare (Fig. 12Aii). .......................... 24
21. Mandals not extending to mandibular apex; antero- median ocellus orthogonally dorsal to compound eye in profile view (Fig. 12Bi). ............................................. Noonilla copiosa Petersen, 1968;
- Noonilla zhg-my04
22. Mandalus extending to mandibular apex; antero- median ocellus positioned posterodorsal to compound eye in profile view (Fig. 12Bii). .......................... 23
- Stylus longer than gonocoxa (Fig. 13Ai). .......................... Noonilla zhg-my01
- Stylus shorter than, or subequal in length to, gonocoxa (Fig. 13Aii). .......................... Noonilla zhg-my06
23. Penial apex entire. .......................... Noonilla zhg-my02
- Penial apex cleft. .......................... Noonilla zhg-my06
24. Dorsal propodeal face long, parallel to craniodorsal axis; protibial comb present. ............................ 25
- Dorsal propodeal face short, with propodeal outline in profile view convex; protibial comb absent. .......... 29
25. Phalotreme at penial apex (Fig. 13Bi). .......................... 26
- Phalotreme basal to penial apex, anatomically ventral (Fig. 13Bii). .......................... 27
26. Penial sclerites dorsoventrally compressed at apex. .................. **Leptanilla zhg-my03**
   - Penial sclerites lateromedially compressed at apex. .................. **Leptanilla zhg-my04**
27. Stylus present, penial sclerites with recurved apical hook (Fig.13Ci). .................. **Leptanilla zhg-id01**
   - Stylus absent, penial sclerites without recurved apical hook (Fig.13Cii-iii). .................. 28
28. Basolateral gonocoxal lamina subulate (Fig.13Cii). .................. **Leptanilla zhg-my02**
   - Basal lateral gonocoxal lamina lanceolate (Fig.13Ciii). .................. **Leptanilla zhg-my05**
29. Mesoscutellum produced into recurved posterior process. .................. **Leptanilla zhg-th01**
   - Mesoscutellum not produced into recurved posterior process. .................. 30

Fig. 13: (A) Profile view of male genitalia of (i) *Noonilla* zhg-my01 and (ii) *Noonilla* zhg-my02, to scale; (B) posterior view of penial sclerites in (i) *Leptanilla* zhg-my04 (CASENT0842553) and (ii) *Leptanilla* zhg-my05 (CASENT0106432); (C) profile view of male genitalia (not to scale) in (i) *Leptanilla* zhg-id01 (hook at penial apex marked), (ii) *Leptanilla* zhg-my02, and (iii) *Leptanilla* zhg-my05, basolateral gonocoxal laminae marked for Cii-iii; (D) ventral view of male genitalia in (i) *Leptanilla* tenuis (after Santschi 1907: Fig. 1C), (ii) *Leptanilla* bifurcata (after Kugler 1986: Fig. 8), and (iii) *Leptanilla* santschii (after Wheeler & Wheeler 1930: Fig. 2D); (E) ventral view of genitalia of (i) *Leptanilla* tanit (after Santschi 1907: Fig. 2B) and (ii) *Leptanilla* israelis (after Kugler 1986: Fig. 14); (F) ventral view of gonopodite in (i) *Leptanilla* israelica (after Baroni Urbani 1977: Fig. 39), (ii) *Leptanilla* australis (after Baroni Urbani 1977: Fig. 38), (iv) *Leptanilla* africana (after Baroni Urbani 1977: Fig. 37), and of the entire genitalia (iii) in *Leptanilla* exigua (after Santschi 1908: Fig. 1A); (G) dorsal view of the male genitalia in (i) *Leptanilla* alexandri (after Dlussky 1969: Fig. 3), and (ii) *Leptanilla* japonica (after Ogata & al. 1995: Fig. 12), with penial sclerites marked in red. Volsellae marked in blue. Abbreviations: pen = penial sclerites; gcx = gonocoxa; stl = stylus.
30. Stylus bifurcated. .......................... 31
   Stylus entire. ............................ 38
31. Petiole without distinct dorsal node. .......................... 32
   ............... Leptanilla minuscula SANTSCHI, 1907
32. Ventromesal gonocoxal margin with sinuate process
   (Fig. 13Eii). .......................... 33
   Ventromesal gonocoxal margin entire (Fig. 13Eii). 33
33. Stylar apex with obtuse tooth subterminating dorsal process.
   ........................................ Leptanilla GR02
   Stylar apex lacking obtuse tooth subterminating dorsal
   process. .................................. 34
34. Ventromedian margin of stylus excavated basal to
   apical furca. ............................ 35
   Ventromedian margin of stylus entire basal to
   apical furca. ............................. 35
35. Dorsal process of stylar apex acuminate. ........................ 36
   ........................................ Leptanilla tenuis SANTSCHI, 1907
   Dorsal process of stylar apex rounded. .................. 36
36. Penial apex entire. .......................... 37
   Penial apex emarginate. ........................ 37
37. Internal margins of apical penial cleft distinctly
   separated, ventral stylar process narrower than
   dorsal process (Fig. 13Dii). ...........................
   ........................................ Leptanilla bifurcata KUGLER, 1987
   Internal margins of apical penial cleft adjacent,
   stylar processes subequal in breadth (Fig. 13Eii).
   ........................................ Leptanilla israelis KUGLER, 1987
38. Stylus not tapered. .......................... 39
   Stylus tapered. ........................... 42
39. Volsella with expanded apex (Fig. 13Dii). ...................
   Leptanilla santschii WHEELER & WHEELER, 1930
   Volsella (when visible) without expanded apex
   (Figs. 13Eii, Fiii-iv, Gi). ........................... 40
40. Stylus with expanded, rounded apex (Fig. 13Fii). ....
   .............................. Leptanilla islamica BARONI URBANI, 1977
   Stylus with apex not expanded (Figs. 13Fii-iv,
   Gi-ii). ......................................... 41
41. Penial outline attenuate in posterodorsal view
   (13Gii). ................................. Leptanilla alexandri BLUSSKY, 1969
   Penial outline elliptic in posterodorsal view
   (Fig. 13Gii). ............................... 42
   ....................................... Leptanilla japonica BARONI URBANI, 1977
42. Stylus ligulate in outline (Fig. 13Fiv). .....................
   ...................................... Leptanilla africana BARONI URBANI, 1977
43. Stylus not ligulate in outline. .......................... 43
44. Stylar apex acuminate (Fig. 13Fii). ........................ 44
   Stylar apex digitate (Fig. 13Fii). ........................ 45
45. Mesopleural sulcus traversing most of mesopleuron;
   abdominal sternite II without ventral projection.
   ............................ Phaulomyrma javana
   Wheeler & Wheeler, 1930
   Mesopleural sulcus traversing posterior 1/3 of
   mesopleuron; abdominal sternite II with ventral
   projection. ............................... Leptanilla zhg-bt01
   Penial sclerites broader than long. ........................ 46
   Penial sclerites longer than broad. ........................ 47
46. Stylus not articulated to gonocoxa. .......................... 47
   .............. Leptanilla exigua SANTSCHI, 1908
   Stylus articulated to gonocoxa. ........................ 47
   Mesopleural sulcus present; Sc+R+Rs tubular. ....
   ........................................ Leptanilla zhg-au01
   Mesopleural sulcus absent; Sc+R+Rs absent. ....
   ........................................ Leptanilla australis BARONI URBANI, 1977

Description of Protanilla lini male

Protanilla Taylor, 1990

SINGAPORE. Type-species: Protanilla rafflesi, by original
designation.

Protanilla lini Terayama, 2009

P. lini Terayama, 2009: 126, Figs. 113 - 118 (worker).

Material examined (4 males): OKENT0027514.
Japan, Okinawa Is.: Ogimi, Hentona High School
(26.70134° N, 128.13156° E), 13 - 27.V.2016, 21 m elevation,
leg. OKEON, SLAM trap (S0015), OK01355 (OIST).
OKENT0028803. Japan, Okinawa Is.: Nago, Nago Central
86 m elevation, leg. OKEON, SLAM trap (S0068), OK01516
(OIST).

OKENT0018456. Japan, Okinawa Is.: Naha, Sueyoshi
Pk. (26.22831° N, 127.71600° E) 1 - 15.VII.2016, 65 m elevation,
leg. OKEON, SLAM trap (S0057), OK01851 (OIST).

OKENT0011907. Japan, Okinawa Is.: Onna, OIST
Campus Forest Site (26.48509° N, 127.84190° E), 17.VI.
- 1.VII.2015, 107 m elevation, leg. OKEON, SLAM trap
(S0008), OK0017 (OIST).

Male description: Head. In full-face view head
slightly broader than long (CS 0.409 - 0.465), excluding
compound eyes (Fig. 15A). Labrum reduced, lateromedial-
ly compressed, bare of apparent vestiture. Mandibles
reduced, tub-like, edentate, articulated to cranium ("mdb"
in Fig. 14); mandalus ("mdl" in Fig. 15C) large, covering
entire anterodorsal mandibular surface in full-face view.
Palpal formula assessed to be 4,1 in situ; maxillary palp

![Fig. 14: Mouthparts of male Protanilla lini, full-face view. Abbreviations: mdb = mandible; gal = galea; gls = glossa; lbp = labial palp; mpx = maxillary palp.](image-url)
Fig. 15: Protanilla lini male. (A) full-face view of head (OKENT0018456); (B) dorsal view of mesosoma (OKENT0011097); (C) profile of head (OKENT0011097); (D) profile of mesosoma (OKENT0028803); (E) profile of metasoma (OKENT0011097).

Abbreviations: pm = pronotum; aas = antero-admedian line; mes = mesonotum; pps = parapsidal line; teg = tegula; axi = axilla; mdp = mandalus; mps = mesopleural sulcus; ump = upper metapleuron; pro = propodeum; prs = propodeal spiracle; ste = abdominal sternite IX; pen = penial sclerites.

("mxp" in Fig. 14) extending past hypostomal margin, covered with dense setae, articulation between palpomeres 1 - 2 indistinct; labial palp ("lbp" in Fig. 14) short and robust, sparsely setose. Premental shield broadly truncate at apex. Galea ("gal" in Fig. 14) simple, sparsely setose, twice the length of mandible in full-face view. Clypeus with medial anteroposterior length about twice the diameter of the torulus, anterior margin entire, posterior margin not produced between toruli (Fig. 15A). Anterior tentorial pits situated directly anterior to antennal toruli, with no part of torular lobe extending anterad of anterior tentorial pit. Ocellar region bulging moderately, but ocelli not set on distinct tubercle; posterior ocellar line longer than lateral ocellar line. Occipital carina present dorsally, not enclosing occiput. Hypostomal carina present, not laminate. Compound eyes wider than long in profile view, slightly convex in full-face view, medial margin slightly convex, all margins entire (Fig. 15A, C). Antennae 13-merous; scape cylindrical, shorter (SL 0.150 - 0.173 mm) than width (EW 0.273 - 0.301 mm) or length (EL 0.206 - 0.234 mm) of compound eye (Fig. 15C); pedicel short, subcylindrical, dilated apically, 2 / 3 length of scape (LF1 0.075 - 0.091 mm); antennomere 3 long, cylindrical, twice the length of pedicel (LF2 0.131 - 0.140 mm); antenna filiform, slightly longer than mesosoma.

Mesosoma. In profile view anterodorsal pronotal face linear, diagonal to craniocaudal axis at ~60° angle; anterior and posterior pronotal margins subparallel in profile view (Fig. 15D). Mesoscutum expanded dorsally, strongly convex ("mes" in Fig. 15B, D); mesoscutal width measured in dorsal view between pronotal lobes subequal to mesoscutal length measured in same view from anterior mesoscutal margin to transscutal line. Notauli absent. Antero-admedian line present on mesoscutum ("aas" in Fig. 15B). Parapsidal signa ("pps", Fig. 15B) present, divergent, slightly impressed. Parascutal carina present. Axillae ("axi" in Fig. 15B) small and well-separated in dorsal view, anteroposteriorly expanded laterally, lateral faces concave. Preaxilla present, impressed. Axillula ("axu" in Fig. 16) impressed into shallow trough; axillular line indistinct. Mesotum longer than tall, dorsoventrally lower than that of mesoscutum, posterodorsal face of mesoscutellum ("msd" in Fig. 16) convex and without posterodorsal process(es); mesoscutellar arm ("msa" in Fig. 16) strongly elevated. Metasoma small and anteroposteriorly narrow, extending posterior to mesoscutellum in dorsal view ("met" in Fig. 16). Metanotal trough deeply excavated, with coarse longitudinal sulci; metascutellar arm ("mta" in Fig. 15) moderately elevated. Mesopleural sulcus ("ols") bisecting mesoscutus ("mps") (Fig. 15D). Longitudinal metapleural sulcus absent. Upper metapleuron ("ump" in Figs. 13 - 14) distinct from propodeum, constricted ventrally in profile view. Lower metapleuron fused insensibly to propodeum. Metapleural gland absent. Propodeum ("pro" in Figs. 15D - 16) parabolic in profile view, with narrow distinct dorsal face; propodeal spiracle ("prs" in Figs. 15D - 16) circular, facing posteriorly, slightly more adjacent to propodeal foramen than to metapleuron; propodeal lobe absent. All pairs of legs with similar proportions; procoxa without anteroventral transverse carina; protrochanters twice as long as wide; profemur not markedly constricted at base, not incrassate, lacking ectoventral flange at apex, carina absent from mesal face; femora moderately anteroposteriorly compressed; dorsoventral protibial width greatest at apex, apex not dorsoventrally flattened; ventral protibial face without distinct margins, convex in cross-section. Mesotibial spur not apparent; pro- and metatibial spurs conspicuous.

Forewing. Tegula small ("teg" in Fig. 15B). Membrane hyaline. Venation of Ogata Type IVb (Fig. 17A). C, Sc + R + Rs, Rf, Mf1, and 1A tubular; M + Cu nebulous at juncture with cu-a, spectral and disappearing basally. Cu-a with weakening adjacent to LA. Pterostigma present, heavily infuscated, with all enclosing abscissae tubular, although RF with weakening basal to pterostigma. 2s-rs + Rs + 4 - 6 tubular and spectral apically, not reaching costal margin (Fig. 17A).

Hindwing. Membrane hyaline. Four distal hamuli present ("ham" in Fig. 17B). Venation reduced: R+Rs tubular, extending < 1 / 5 of distance along costal margin;
A spectral, not extending to anal margin, one quarter of R+Rs length. Jugum absent. Clavus ("clv" in Fig. 1) weakly developed.

Metasoma. Petiole (Fig. 18) anteroposteriorly compressed (DPW 0.164 - 0.199 mm, PTL 0.201 - 0.242 mm), without peduncle, lateral faces subparallel in dorsal view; posterior petiolar foramen elevated dorsad anterior petiolar foramen; tergal spiracle present, situated anteriorly on petiole; longitudinal carinulae of tergum II absent from petiole; petiolar tergite and sternite fused, delimited by faint longitudinal suture ("ls" in Fig. 18) visible in ventral profile view; petiolar tergite with pronounced dorsal node, with dorsal face of node slightly convex; petiolar sternite lateromedially compressed into process occupying the anterior 2/3 of petiole length. Pretergite III with lateral margins diverging dorsally; presternite III with lateral margins converging ventrally; medial anteroposterior length of pretergite III greater than that of presternite III. Dorsomedial length of helcium 1/3 × that of petiole; helcium axial (Fig. 15E). Abdominal segment III distinctly narrower than IV in dorsal view (TW3 0.247 - 0.321 mm, TW4 0.487 - 0.579 mm), with margins of tergite III subparallel in dorsal view; sternite III convex, without prora. Presclerites of abdominal segment IV visible with girdling constriction present but indistinct; presclerites of abdominal

Fig. 16: Male mesosoma of Protanilla lini (OKENT0028803), posterior view. Abbreviations: mes = mesonotum; axu = axillula; msd = mesoscutellar disc; msa = mesoscutellar arm; ump = upper metapleuron; pro = propodeum; prs = propodeal spiracle.

Fig. 17: (A) Forewing and (B) hindwing of Protanilla lini. Abbreviations: ham = hamuli; clv = clavus.

1A spectral, not extending to anal margin, one quarter of R+Rs length. Jugum absent. Clavus ("clv" in Fig. 17B) weakly developed.
segments V - VIII inconspicuous. Abdominal segments III-VIII without tergosternal fusion. Abdominal tergite VIII broader than long, posterior margin somewhat laminate. Abdominal sternite IV moderately convex medially; abdominal sternites IV-VIII unmodified, not convex; all sternites visible in situ. Posterior margin of sternite VIII entire and unmodified. Abdominal sternite IX with margin extended into posterodorsal process ("ste" in Figs. 15E, 19B, D), its anterodorsal surface concave, tip rounded, three times longer than visible lateral length of sternite IX; posterior margin of sternite IX lateral to process with rounded laminae.

Genitalia. Pygostyles absent. Gonopodites partially articulated, with faint ventral articulation discernible in profile view. Gonocoxae ("gcx" in Fig. 19A, D) glabrous, somewhat dorsoventrally compressed; distinct from one another along entire ventromedial length; posterodorsal marginal laminae absent. Styli ("stl" in Fig. 19A, D) not compressed at base, dorsoventrally compressed towards apex with distinct ectal and dorsal faces, all stylar surfaces with posteriorly directed setae, dorsal face slightly convex, medial margin of stylus with dorsal carina along basal 2 / 5 of stylar length; stylar apex blunt. Parossiculus ("prs" in Fig. 19A - B, D) lateromedially compressed towards apex, margins rounded; lateropenite ("ltp" in Fig. 19A - B, D) extending posterad parossiculus, ectal surfaces convex, lateropenital apex ectally recurved. Penial sclerites ("pen" in Fig. 19A - B, D) lateromedially compressed, ectal surfaces convex; medial conjunctiva extending from gonocoxae to penial apex; dorsal margins of valviceps ("vlv" in Fig. 19A) extended into triangular processes, tips curving laterally; ventral margins of valviceps ventrally extended into triangular processes.

Sculpturation. Sculpturation weak to lacking on most sclerites. Fine piligerous punctae present on head, femora, and tibiae; mesosomal and metasomal piligerous punctae, where present, coarser; punctae on mesosoma and petiole sparse by comparison to those on head, abdominal segments III - IX, and limbs; piligerous punctae absent from propleuron, pronotum, and mesoscutum. Sublar areas of upper mesoscutum and upper metapleuron intricately, confusedly striolate; metanotal trough porcate; sclerites otherwise glabrous. Dorso-anterodorsal mesoscutal margin, mesopleural suture, and anterior margin of metapectal-propodeal complex scrobiculate. Irregular denticle-like microsculpture on dorsal-ectal (Fig. 19C) and posterodorsal faces of valviceps.


Setation. Vestiture coarse overall, with most somal and appendicular setae short (~30 μm) to moderate (~80 μm) in length, subrect to decumbent, variable among setae on given sclerites. Pedicel and flagellum densely covered with short decumbent or appressed setae, with longer subdecumbent setae scattered over surface of antennomeres. Ectal pro- and metacoxal surfaces bare; posterior metacoxal surfaces bare; tarsi covered with vestiture like that present on pedicel and flagellum. Head, scape, and mesosoma with moderately dense setae; prosternite, antero-alar region of pronotum, and upper metapleuron with setae sparse, surfaces of these sclerites being almost bare.
Wings covered with short subdecumbent setae. Petiolar setae dense only on anterior face of petiolar node; abdominal segment II otherwise largely bare. Remainder of metasoma covered with dense setae; setal length gradually increasing posteriorly, with those on abdominal sternites VII - IX especially long (0.10 - 0.13 mm; ranges given representing inter-specimen variation in maximum length) and recurved, this trend being most pronounced on posteroventral median process of sternite IX. Genital setae restricted to stylus and basivolsella, except for some basodorsad the gonopodital articulation. Some dorsal stylar setae unusually long (~100 - 115 μm) and recurved.

Conclusions
Ant systematics generally relies upon the worker caste to the exclusion of males: short-lived, male ants are less likely to be collected than their female counterparts, and male morphology has only occasionally (e.g., Wild 2007, La Polla & al. 2012, Barden & al. 2017) been found to contain phylogenetic signal not already provided by workers. The Leptanillinae are unusual in that in their case phylogenetic inference from molecular data and male morphology for the purpose of delimiting subclades of the tribe Leptanillini, and so resolve the status of the many taxa therein that are known only from males.

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