# Phylogeny of *Paussus* L. (Carabidae: Paussinae): unravelling morphological convergence associated with myrmecophilous life histories

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**Abstract.** Ant nest beetles (*Paussus* L.) are ecologically fascinating and phenotypically bizarre. Obligate myrmecophiles, *Paussus* have undergone extreme adaptations for life with ants and their profound range of phenotypic diversity has been difficult to reconcile in a systematic framework. We conducted a detailed morphological study of *Paussus* utilizing novel techniques and character systems in order to discover anatomical apomorphies diagnostic of the major clades of *Paussus* strongly supported by molecular data. Bayesian inference (BI) of molecules alone, morphology alone and varying combinations of the two data types reveal that morphology is informative for placing *Paussus* species, despite the extreme phenotypic diversity and convergence prevalent in the group. We propose a new classification for *Paussus* based on this phylogeny. The genus *Paussus* is revised to include all genera, subgenera and species formerly classified as *Paussus* by Nagel (2003) in addition to *Hylopaussus* **syn.n.**, *Hylotorus* **syn.n.** and *Granulopaussus* **syn.n.** The following species are transferred to *Paussus*: *Paussus sebakuanus* (Péringuey) **comb.n.**, *Paussus gracilis* (Reichensperger) **comb.n.**, *Paussus bucephalus* Gyllenhal, *Paussus caroli* (Reichensperger) **comb.n.**, *Paussus uelensis* (Reichensperger) **comb.n.**, *Paussus hottentottus* (Westwood) **comb.n.**, *Paussus blanchardi* (Raffray) **comb.n.**, *Paussus basilewskyi* (Luna de Carvalho) **comb.n.**, *Paussus granulatus* Westwood, *Paussus sankuruensis* Reichensperger, *Paussus leleupi* (Reichensperger) **comb.n.**, *Paussus reichenspergeri* (Luna de Carvalho) **comb.n.** We formally delineate and diagnose the following major subgroups of *Paussus*: *Paussus* I series, comprising the subgenera *Bohemanipaussus* Luna de Carvalho **stat. rev. sensu n.**, *Bathypaussus* Wasmann **stat. rev. sensu n.**, and *Edaphopaussus* Kolbe **stat. rev. sensu n.**; the *Paussus* II series comprising the subgenera *Paussus* L. **stat. rev. sensu n.**, *Klugipaussus* Kolbe **stat. rev. sensu n.**, *Scaphipaussus* Fowler **stat. rev. sensu n.**, *Hylotorus* Dalman **stat.n. sensu n.**, and *Anapaussus* Wasmann **stat. rev. sensu n.**; and the *Paussus* III series comprising the subgenera *Lineatopaussus* Kolbe **stat. rev. sensu n.** and *Shuckardipaussus* Kolbe **stat. rev. sensu n.**

## Introduction

The genus *Paussus* L. is a highly specialized, charismatic group of ground beetles (Carabidae) classified in the subfamily Paussinae (Figs 1, 2). Paussines are collectively known as flanged bombardier beetles due to their ability to explosively discharge defensive benzoquinones at temperatures of between 55 and 100°C (Eisner *et al*., 1977) and to their possessing a unique cuticular flange located at the posterior outer angle of each elytron (flange of Coanda), which functions in directing the flow of discharged defensive blasts (Eisner *et al*., 2000) (though *Metrius* Eschscholtz is a notable exception as species do not possess a cuticular flange). Paussinae are currently divided into four tribes: Metriini, Ozaenini, Protopaussini and Paussini. *Paussus* is the most species-rich paussine genus with *c*. 350 described and at least 40 known, yet undescribed, species distributed in Africa,

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Madagascar, Indo-Malaysia and Southern Palaearctic (Nagel, 2003); all species and most subgeneric clades are endemic to one of these four biogeographic regions (Moore & Robertson, 2014). Adults (Fig. 1) exhibit an extraordinary diversity of unique morphological structures, mostly associated with their obligate symbioses with ants. In fact, the extent of the phenotypic diversity found among *Paussus* species, commonly known as ant nest beetles, greatly exceeds that of any other carabid genus.

All species of *Paussus* are obligate myrmecophiles (associates of ants). In this symbiosis the beetles provide rewarding chemical secretions to their host ants and in return receive protection, a safe place for their vulnerable larvae to develop and a reliable source of protein-rich food: the ants, particularly the brood. It is not clear how *Paussus* enter host ant nests or are initially accepted into the ant colony. Previous observations indicate that different species may vary in the degree to which they integrate into the social structure of their host ants (see Geiselhardt *et al.*, 2007). At least some species are fully integrated into the ant colony – they are not just tolerated roommates/co-inhabitants; rather they are treated as valued guests by the ants. Within the nest, worker ants groom and antennate the beetles and actively lick or palpate all exposed regions of the beetle's body, particularly those rich in trichomes (Maurizi *et al.*, 2012). Worker ants grab adult *Paussus* by their antennae and legs and transport them to new nests (W. Moore, personal observation). One remarkable aspect of this symbiosis is the intimate interaction between beetle and ant. Recent work on the behaviour of *Paussus favieri* and its host ant *Pheidole pallidula* revealed that the adult beetle interacts directly with the queen in her chamber, antennating and rubbing against the queen's body without eliciting any aggressive behaviour from the queen or the workers (Maurizi *et al.*, 2012). Moreover, both *P. favieri* and the host queen stridulate back and forth during this interaction. Indeed, acoustic communication and deception appear to play an important role in their integration into the ant society (Di Giulio *et al.*, 2015). Females lay their eggs in the ant nest and this is where the beetle larvae develop. *Paussus* larvae are physogastric with a reduced head capsule and mouthparts and short, highly modified legs (i.e. trochanter, femur, tibia and tarsus are fused) that function more for support than for walking (Di Giulio *et al.*, 2011). These and other anatomical modifications, along with specialized calling behaviour, are reminiscent of those exhibited by larval ants when soliciting food from workers and suggest that beetle larvae are fed by the ants through trophallaxis (regurgitation of food) (Di Giulio, 2008; Di Giulio *et al.*, 2011). Once integrated into the society, adults feed at will by piercing and holding the ant integument with their mandibles, extracting tissues and fluids through suction (Di Giulio *et al.*, 2012). Adult ants that were observed being preyed upon never reacted aggressively and, when released, remained near the beetle until dying shortly thereafter (see Geiselhardt *et al.*, 2007; A. Di Giulio, personal observation).

As with many other myrmecophilous or termitophilous beetles [e.g. *Eocorythoderus* Maruyama (Scarabaeidae) (Maruyama, 2012); *Diartiger* Sharp, *Colilodion* Besuchet (Staphylinidae: Pselaphinae) (Nomura, 1997; Nomura & Sugaya, 2007); *Gnostus* Westwood (Ptinidae) (Lawrence & Reichardt, 1966; Thomas

*et al.*, 1992)], *Paussus* have undergone extreme phenotypic adaptations for life with ants (see Figs 3–16). Most notably, the antennae of *Paussus* are highly modified; the pedicel is small, ring-like and nearly concealed within the socket of the scape (Fig. 9E), while the flagellum is fused into a single article and occurs in an astonishing diversity of forms (Figs 1, 10). Filled with glands and adorned with associated pores and trichomes, *Paussus* antennae have been co-opted for the production and delivery of appeasement chemicals (Di Giulio *et al.*, 2009, 2012).

While the profound range of phenotypic diversity within *Paussus* is fascinating from evolutionary and ecological standpoints, it has proven difficult to reconcile in a systematic framework (Moore & Robertson, 2014). The plethora of striking morphological features associated with a myrmecophilous lifestyle has obscured the discovery of shared derived character states indicative of a shared evolutionary history.

When forming hypotheses of relationships and proposing classifications within *Paussus*, previous authors focused largely on character systems that are intimately tied to *Paussus*–host ant interactions, such as the highly modified antennae, disc of head and pronotum. While these character systems include the most blatant phenotypic differences between species of *Paussus*, given their key functional roles in symbioses (Maurizi *et al.*, 2012) they are almost certainly under strong selective pressure by host ants; thus the similarity of form between certain species may not reflect evolutionary relationship/history. None of these traditionally used characters for *Paussus* systematics have been investigated in a phylogenetic context. For example, both Darlington (1950) and Nagel (1987) provided qualitative outlines of putative diagnostic character states for some tribes and subtribes within the subfamily Paussinae and select groups within *Paussus*. Nagel (1987) treated species belonging to the *P. armatus* Westwood, *P. inermis* Gerstaecker and *P. laevifrons* (Westwood) groups based on his earlier taxonomic revisions of these taxa (Nagel, 1977, 1980, 1986).

The classification of species of *Paussus* differs amongst authors (e.g. Kolbe, 1930; Jeannel, 1946; Darlington, 1950; Luna de Carvalho, 1989), reflecting their focus on different suites of superficially similar traits among species. For example, both Jeannel's (1946) concept of *Bathypaussus* Wasmann and Luna de Carvalho's (1989) concept of *Spinicoxipaussus* Kolbe were based largely on the presence of a sickle-shaped antennal club. Not surprisingly, there is considerable overlap in the species included in both taxa/classifications (e.g. *Paussus fallax* Péringuey, *P. reichenspergeri* Brauns and Wasmann, *P. cephalotes* Raffray, *P. jeanneli* Reichensperger), but there is also considerable disagreement. Luna de Carvalho (1989) additionally included *P. spinicoxis* Westwood and its allies (ten species) in *Spinicoxipaussus,* while Jeannel (1946) included *P. aldrovandii* Gestro, *P. cultratus* Westwood, *P. granulatus* Westwood and its allies (four species) in *Bathypaussus*. Previous paussine taxonomists divided *Paussus* into about 30 groups, treated as genera or subgenera by different authors. Indeed, the taxonomy of *Paussus* is in a state of turmoil (Moore & Robertson, 2014), due to the lack of identified anatomical character states indicative of shared evolutionary history. Nagel (2003)

**Fig. 1.** Habitus images of *Paussus* illustrating a small glimpse of their phenotypic diversity: (A) *P. sebakuanus*; (B) *P. granulatus*; (C) *P. spinicoxis*; (D) *P. bohemani*; (E) *P. afzelii*; (F) *P. curtisi*; (G) *P.* nr. *klugi*; (H) *P. marshalli*; (I) *P. solidus*; (J) *P. cervinus*; (K) *P.* nr. *pipitzi*; (L) *P. howa*. Scale bars = 1 mm.

synonymized all previously delimited genera and subgenera into a single large genus, *Paussus*, with 350 species.

Recently, Moore & Robertson (2014) used molecular sequence data to infer the evolutionary history of *Paussus*. Their well-supported phylogenetic results revealed striking patterns of rapid adaptive radiation coupled with extreme phenotypic diversity and convergence across geographically disparate lineages. Nearly all *Paussus* subgenera included in their analysis were shown to be polyphyletic. For example, molecular data (Moore & Robertson, 2014) showed both *Bathypaussus sensu* Jeannel (1946) and *Spinicoxipaussus sensu* Luna de Carvalho (1989) to be polyphyletic, demonstrating not only

**Fig. 2.** Habitus images of Paussinae: (A) *Ozaena martinezi*; (B) *Cerapterus* sp.; (C) *Pentaplatarthrus* sp.; (D) *Heteropaussus* sp.; (E) *Platyrhopalopsis picteti*; (F) *Ceratoderus bifasciatus*. Scale bars = 1 mm.

the artificial nature of the current classification but also the astounding degree of convergent (or parallel) evolution of form across disparate lineages of *Paussus*. Moore & Robertson (2014) demonstrate that such phenotypic convergence/parallelism is widespread in *Paussus*. Moreover, the anatomically bizarre paussine genera, *Hylopaussus* Luna de Carvalho and *Granulopaussus* Kolbe, were strongly supported as deeply nested within *Paussus*, closely related to African members of the *P.* (*Cochliopaussus*) *cucullatus* species group (Nagel, 2006). One of the most striking aspects of *Paussus* evolutionary history revealed by molecular data (Moore & Robertson, 2014) is that the anatomically diverse fauna of Madagascar all recently descended from a single common ancestor. In fact, they found

that the area of endemism is generally a better predictor of monophyly for *Paussus* subgroups than current taxonomic designations (Moore & Robertson, 2014). The results of Moore & Robertson (2014) lead to a completely new concept of evolution and relatedness within this phenotypically variable group of myrmecophilous beetles, calling for a complete revision of *Paussus* classification.

In light of the conflicting historical classifications and concepts of relatedness based on morphology and the surprising phylogenetic relationships recovered by molecular data (Moore & Robertson, 2014), we conducted a study to assess the utility of anatomical data in a phylogenetic context given the extreme phenotypic diversity and convergence prevalent in this
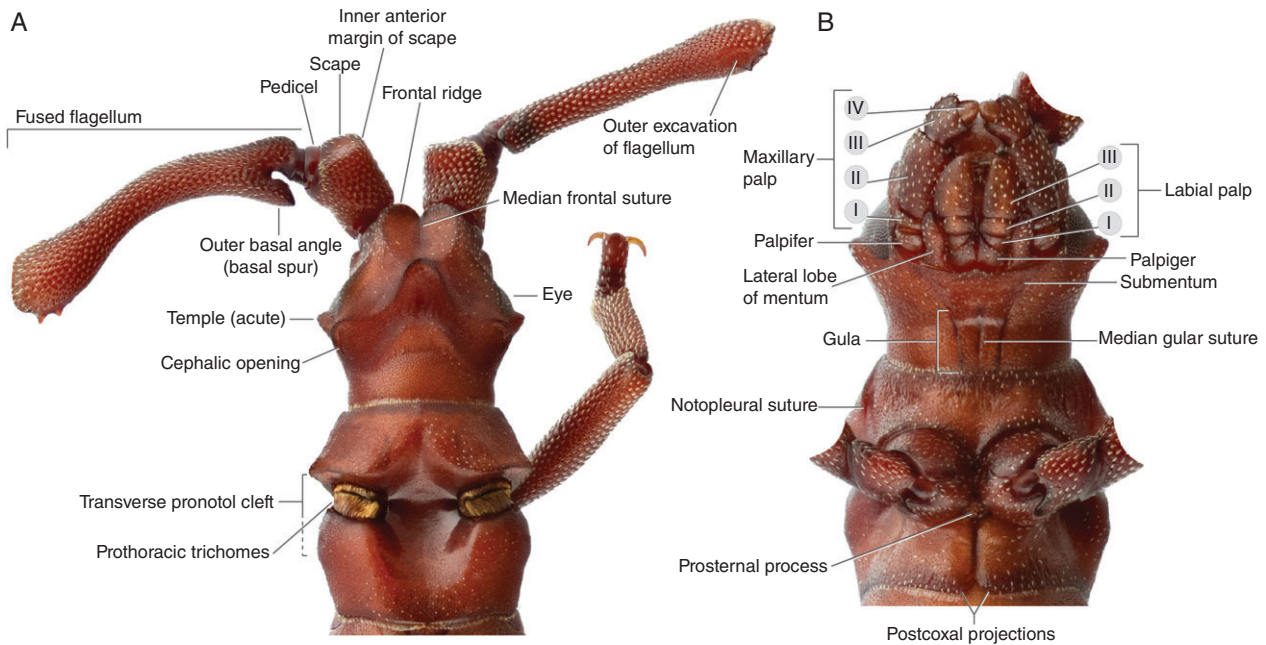
**Fig. 3.** Head and prothorax of *Paussus curtisi* illustrating anatomical features of the genus. (A) Dorsal view with left foreleg digitally removed. (B) Ventral view with antennae and forelegs digitally removed.

adaptively radiating myrmecophilous group. We conducted an in-depth morphological study of *Paussus* utilizing novel techniques and character systems in order to discover anatomical apomorphies of clades strongly supported by molecular data (Moore & Robertson, 2014). Based on our results, we formally delineate and diagnose major subgroups of *Paussus*.

## Materials and methods

### Taxon sampling

In total we included 51 species in our morphological study (Table S1), 40 of which are classified as *Paussus* (Nagel, 2003). We also included the enigmatic genera *Hylopaussus* and *Granulopaussus*, eight outgroup exemplars from Paussini and *Ozaena* Oliver, a more distant taxon within Ozaenini (Fig. 2). Forty-six of the 51 taxa were also included in our previous molecular-based study (Moore & Robertson, 2014), and these were selected to represent all higher-level clades. In addition, we included several taxa for which molecular data are not yet available. These include taxonomically important or systematically enigmatic taxa such as *Paussus* (*Paussus*) *oertzeni* Kolbe, *P*. (*Cochliopaussus*) *solidus* Reichensperger and *P*. (*Edaphopaussus*) *dissimulator* Reichensperger.

### Specimens

*Paussus* specimens are relatively rare, as much of their lives as both adults and larvae are spent within their host ants'

nest. Most of the specimens used in the present study were collected and preserved in 90–100% ethanol (EtOH) by the authors or by our generous colleagues and collaborators (see the Acknowledgements). In most cases, we used the vouchers of Moore & Robertson (2014) to score anatomical character states (see later).

In addition, generous loans of pinned material from the following individuals and institutions served as an invaluable reference collection of *Paussus* that contributed significantly to this study [collection codens largely follow Evenhuis (2008) and the curator responsible for the loan is listed in parentheses]: Albert Dean Allen personal collection, Boise, Idaho, USA (AAC); California Academy of Sciences, San Francisco, California, USA (David H. Kavanaugh) (CAS); Museum and Institute of Zoology, Warszawa, Poland (Wioletta Tomaszewska) (MIZ); Muséum National d'Historie Naturelle, Paris, France (Thierry Deuve) (MNHN); Robert Stephen Copeland personal collection, Nairobi, Kenya (RSC); Iziko Museum of Capetown/South African Museum, Cape Town, South Africa (D. Larsen) (SAMC); South African National Collection of Insects, Pretoria, South Africa (Elizabeth Grobbelaar) (SANC); Senckenberg Deutsches Entomologisches Institut, Müncheberg, Germany (Stephan M. Blank) (SDEI); Wendy Moore Insect Collection. University of Arizona, Department of Entomology, Tucson, Arizona, USA (WMIC); Zoologisches Forschungsmuseum Alexander Koenig, Bonn, Germany (Dirk Ahrens) (ZFMK); Museum für Naturkunde der Humboldt-Universität, Berlin, Germany (Manfred Uhlig) (ZMHB).

For a few taxa, we were unable to use the same species for the morphological data as was sequenced, and for those taxa we
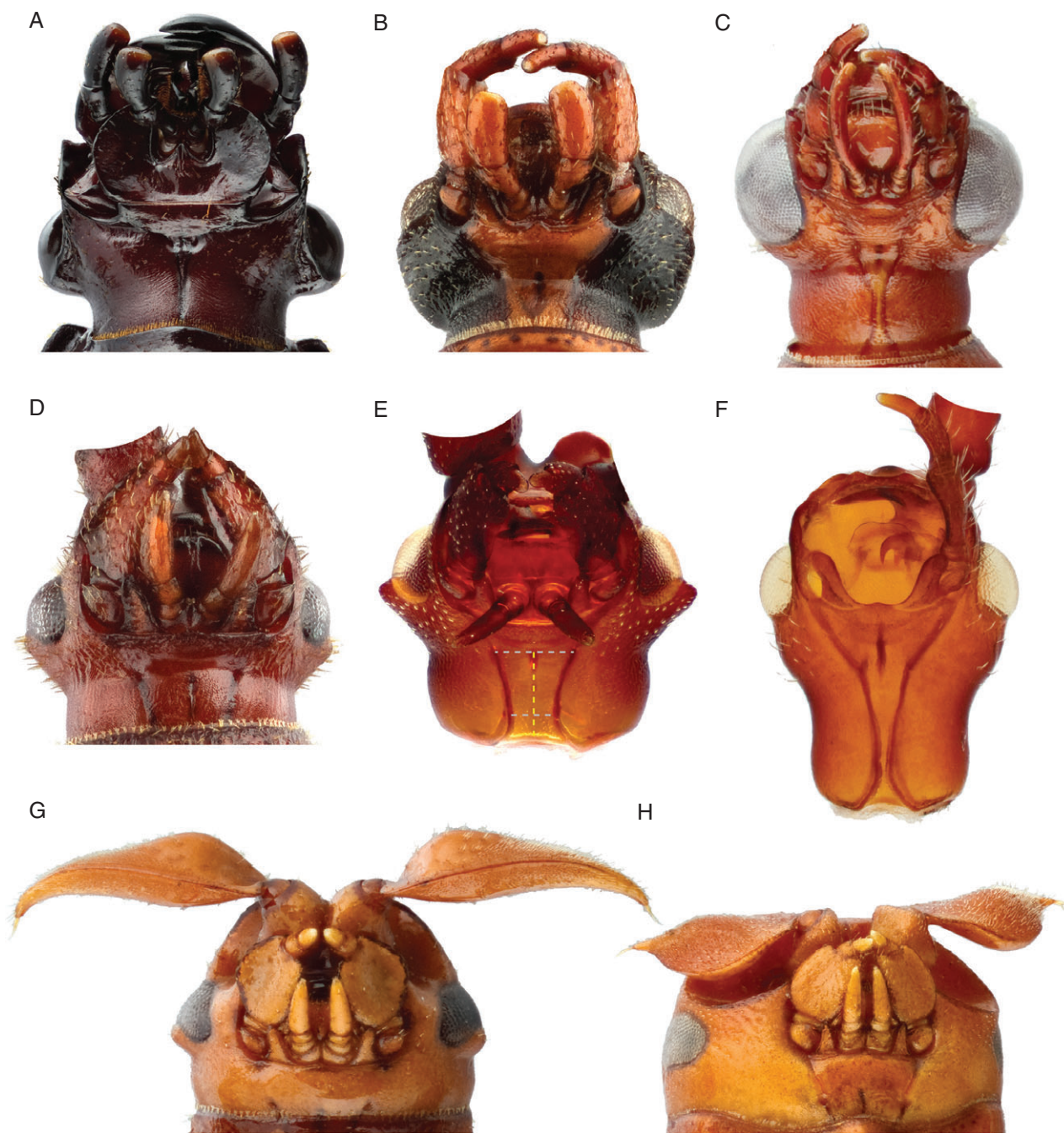
**Fig. 4.** Dorsal aspect of head, with antennae digitally removed (A–J) to show the anterior margin of disc. (A) *Paussus spinicoxis*; (B) *P. bohemani*; (C) *P. signatipennis;* (D) *P. vanrooni*; (E) *P. curtisi*; (F) *P. klugi*; (G) *P. cephalotes;* (H) *P.* nr. *Planifrons*; (I) *P. cervinus*; (J) *P. ghanensis*; (K) *P.* nr. *nudus.*
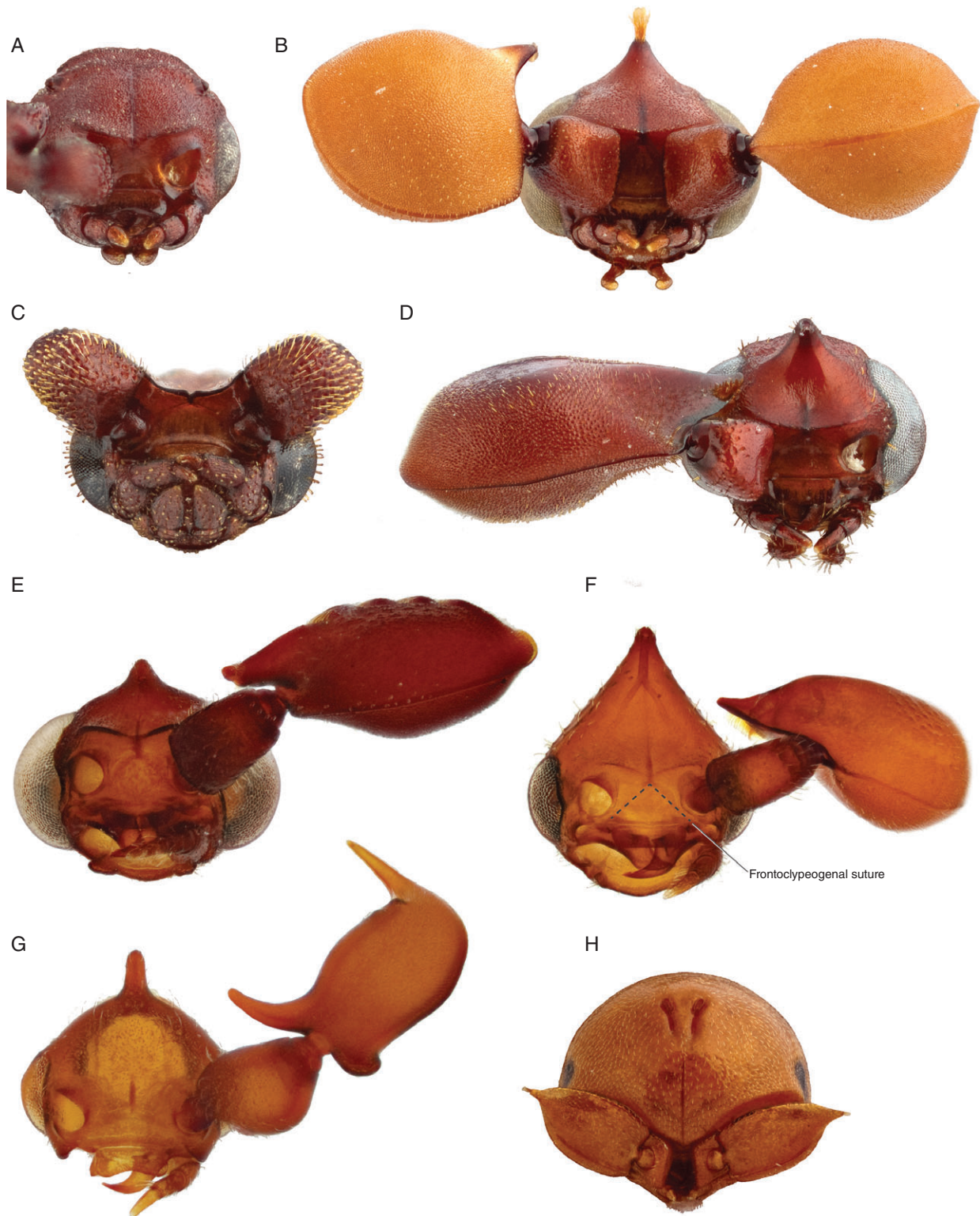
**Fig. 5.** Head, frontal view (cleared specimens: E–G). (A) *Paussus klugi*; (B) *P.* nr. *sphaerocerus*; (C) *P. vanrooni*; (D) *P. spinicoxis*; (E) *P. dohrni*; (F) *P. bohemani*; (G) *P. jeanneli*; (H) *Hylotorus uelensis*.

**Fig. 6.** Ventral aspect of head with (A–F) or without antennae digitally removed (cleared specimens: E–F). (A) *Ozaena* sp.; (B) *Ceratoderus bifasciatus*; (C) *Paussus spinicoxis*; (D) *P. semicucullatus*; (E) *P. solidus* (dashed lines indicate points of measurement for characters 14–15); (F) *P.* nr. *Dama*; (G) *Hylopaussus sebakuanus*; (H) *Hylotorus hottentotus*.

created chimeras of closely related species (e.g. sister species, or species belonging to the same species group) when combining morphological and molecular data matrices. In creating chimeric terminals, we selected closely related species that minimized the amount of missing data (Table S1). Each chimeric terminal comprises species that we expect would be scored identically in our morphological matrix.

*Specimen preparation*

Specimens were cleared using either Proteinase-K at 56°C for 6–14 h or pancreatin (Alvarez-Padilla & Hormiga, 2008) at room temperature for 3–4 days. Once cleared, specimens were disarticulated at the following major junctures: head/prothorax, prothorax/mesothroax and metathorax/abdomen. In addition,
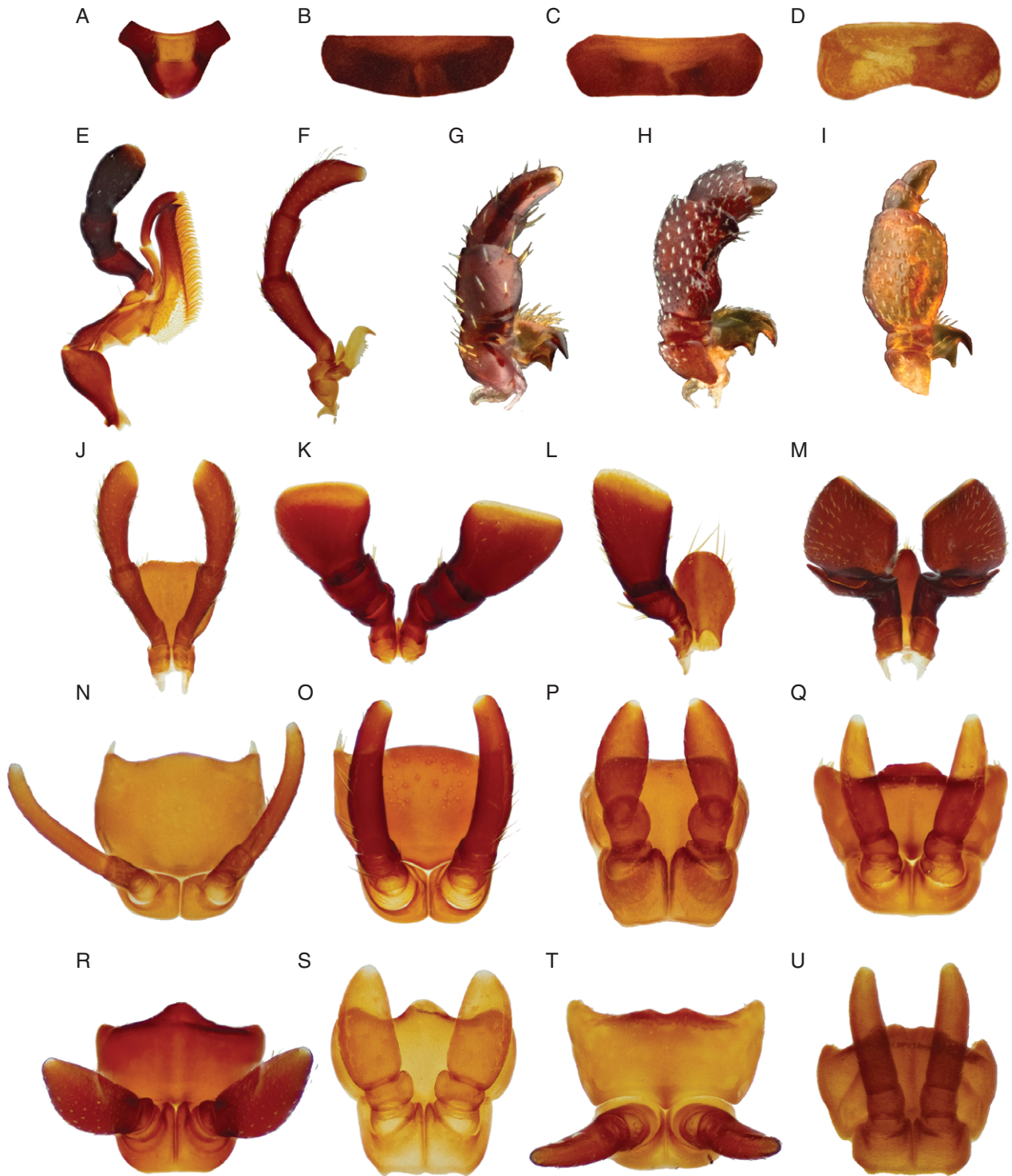
**Fig. 7.** Mouthparts: (A–D) labrum; (E–I) maxilla; (J–U) labium. (A) *Heteropaussus* sp.; (B) *P. dohrni*; (C) *P. bohemani*; (D) *P. jeanneli*; (E) *Ozaena* sp.; (F) *Heteropaussus* sp.; (G) *P. spinicoxis*; (H) *P. curtisi*; (I) *P.* nr. *Fulvus*; (J) *Heteropaussus* sp.; (K) *Arthropterus* sp.; (L) *Cerapterus* sp.; (M) *Homopterus* sp.; (N) *P. bohemani*; (O) *P. humboldti*; (P) *P. vanrooni*; (Q) *P. semicucullatus*; (R) *P. oertzeni*; (S) *P. africanus*; (T) *P. solidus*; (U) *P. pipitzi*.
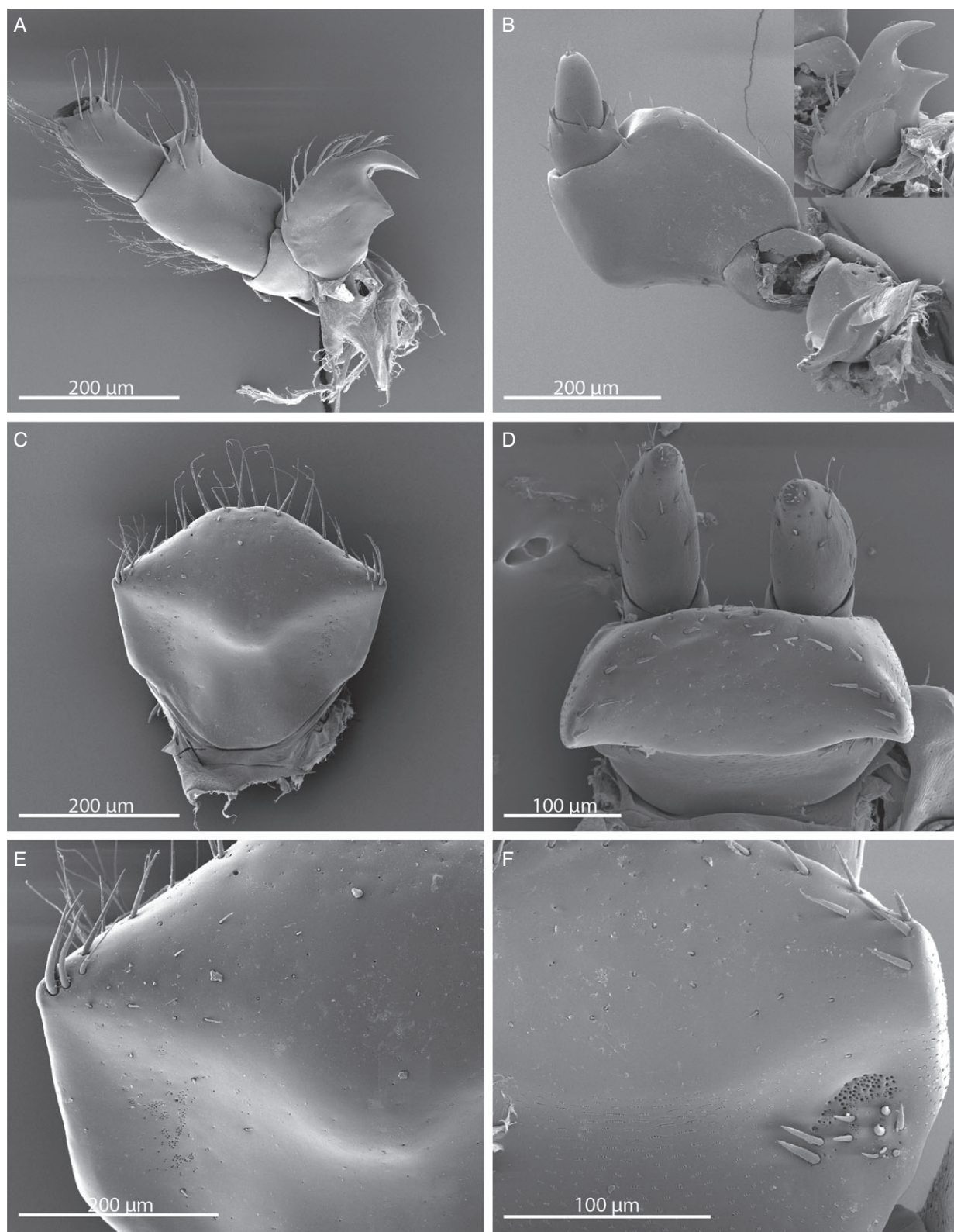
**Fig. 8.** Scanning electron micrograph: (A–B) maxilla; (C–F) prementum. (A, C, E) *Paussus favieri*; (B, D, F) *Hylopaussus* sp.
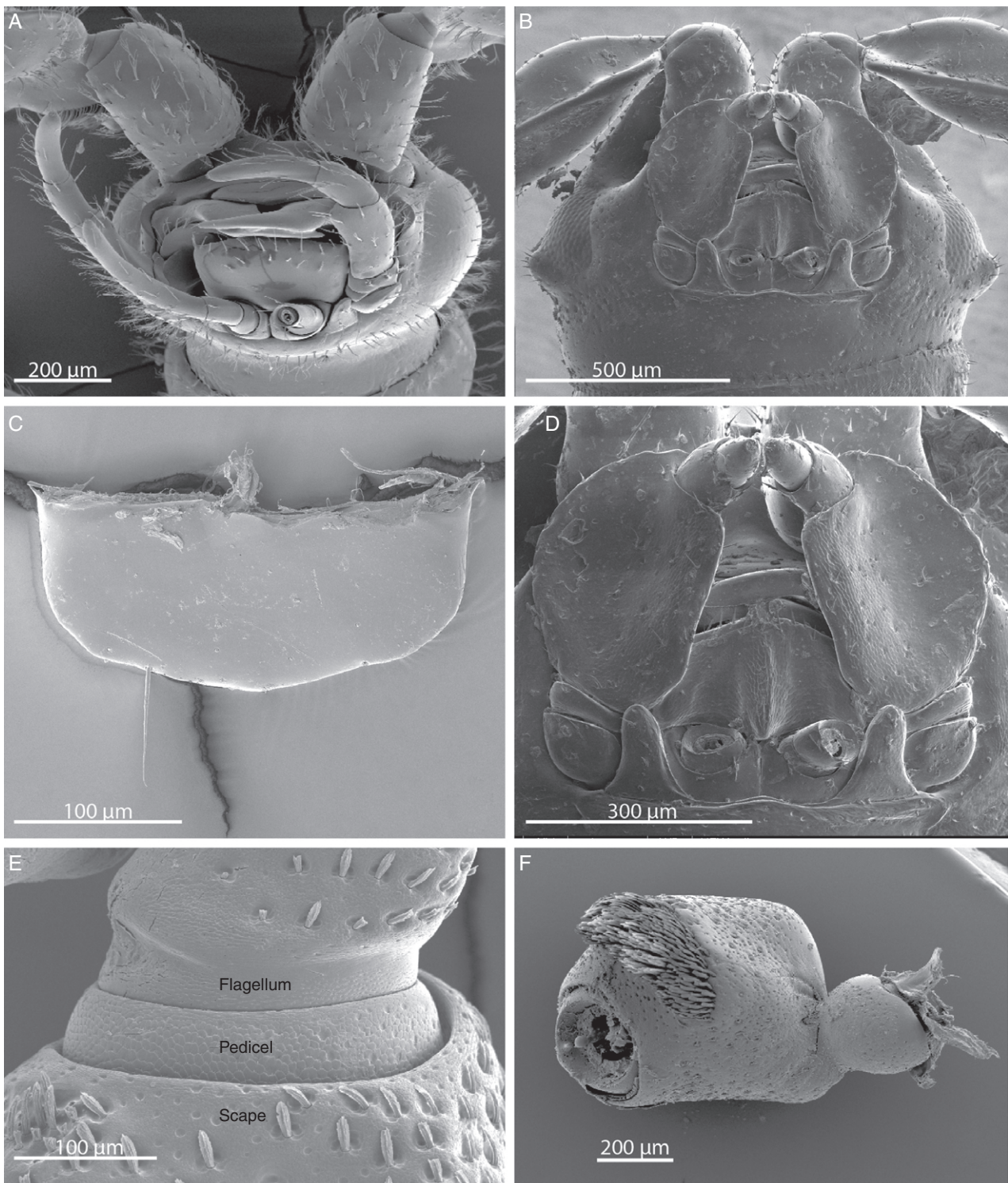
**Fig. 9.** Scanning electron micrograph: (A, B, D) oral cavity; (C) labrum; (E) antennal segments; and (F) scape. (A, C) *Paussus favieri*; (B, D) *Hylopaussus* sp.; (E) *P. curtisi*; (F) *P. oertzeni*.

**Fig. 10.** Dorsal aspect of antenna. (A) *Paussus spinicoxis*; (B) *P.* nr. *sphaerocerus*; (C) *P. oertzeni*; (D) *P. bohemani*; (E) *P.* nr. *mucius*; (F) *P. afzelii*; (G) *P. germari*; (H), *P. jousselini*; (I) *P. curtisi*; (J) *P.* nr. *leechi*; (K) *P. aldrovandii*; (L) *P. planifrons*; (M) *P. cervinus*; (N) *Granulopaussus granulatus*; (O) *P. klugi*; (P) *P. dama*; (Q) *P.* sp.; (R) *P.* sp.; (S) *P. howa*; (T) *P. armicollis*; (U) *P. cucullatus*.

**Fig. 11.** Prothoracic (A–C), mesothoracic (D–F) and metathoracic (G–I) legs. (A) *Paussus* nr. *planifrons*; (B) *Hylotorus hottentotus*; (C) *P. spinicoxis*; (D) *Platyrhopalopsis picteti*; (E) *Paussus spinicoxis*; (F) *P. favieri*; (G) *P. spinicoxis*; (H) *P.* nr. *leechi*; (I) *P. cephalotes*.

the hindwings, antenna (right side only), labial prementum, maxilla (right side only) and mandible (few specimens; right side only) were carefully removed. Each disarticulated specimen was stored in a glycerine-filled concavity slide; hindwings were mounted separately.

Specimens were studied using either a Leica M165 C or Leica M205 C stereomicroscope. Photomicrographs were taken using a Visionary Digital BK system (R. Larimer, http://www.visionarydigital.com). Composite images were assembled using ZERENE STACKER v1.04 (http://www.zerene systems.com/cms/stacker). High-resolution scanning electron

microscope (SEM) images were acquired as follows: (i) dried beetles were rehydrated by boiling them in distilled water for a few minutes; (ii) they were dissected and transferred to Pancreatin solution (at 36°C; see Alvarez-Padilla & Hormiga, 2008) for a time sufficient to properly digest the specimen based on size; and (iii) dissected structures were dehydrated to 100% EtOH by passing through a graded ethanol series, critical point-dried in a Balzer Union CPD 030 unit (Pfäffikon, Switzerland), gold-coated in an Emitech K550 unit (Paris, France), and finally examined using the field emission SEM column (secondary electrons and an operating voltage of 5 kV)
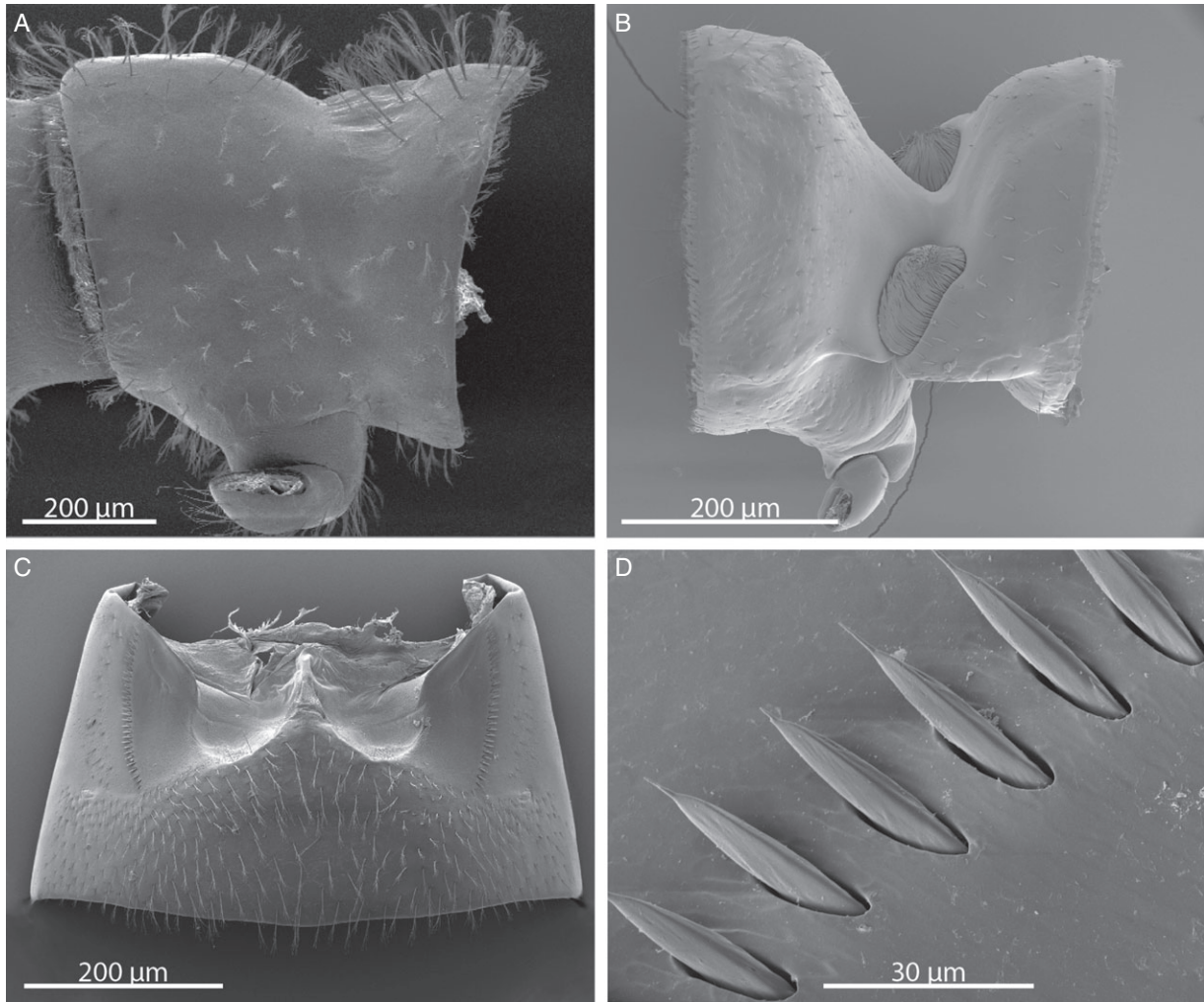
**Fig. 12.** Pronotal disc. *Paussus bohemani* (A); *P. spinicoxis* (B); *P. manicanus* (C); *P.* nr. *nudus* (D); *P. favieri* (E); *P. vanrooni* (F); *P. germari* (G), *P. cephalotes* (H); *P. signatipennis* (I); *P. cucullatus* (J); *Hylotorus hottentotus* (K); *P. solidus* (L).
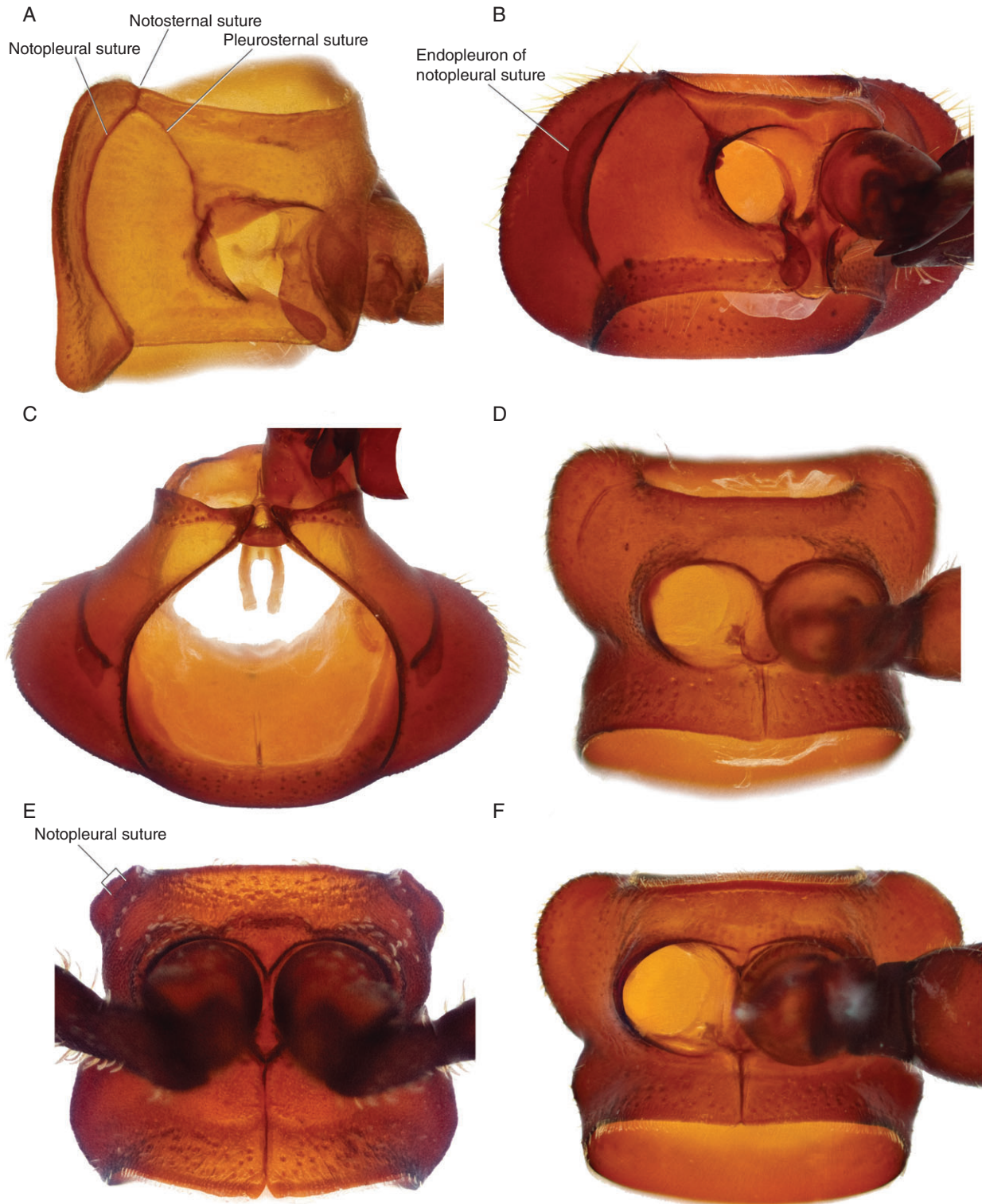
**Fig. 13.** Prothorax: (A, B) posterolateral view; (C) posterior view (D–F) ventral view. (A) *Carabidomemnus* sp.; (B, C) *Cerapterus* sp.; (D) *P. aristoteli*; (E) *P. howa*; (F) *P. laevifrons*.

**Fig. 14.** Scanning electron micrograph. (A, B) Prothorax, lateral view; (C) fused abdominal ventrites 1–3 illustrating the scraper (D) of the stridulatory organ. (A, C, D) *Paussus favieri*; (B) *Hylopaussus* sp.

of the Dualbeam FIB/SEM Helios Nanolab (FEI Company, Eindhoven, the Netherlands) at the L.I.M.E. (Roma Tre University, Rome, Italy). Illustrations and plates were prepared using Adobe ILLUSTRATOR CS6.

*Morphological characters*

Using the cleared, disarticulated collection we discovered a significant number of novel characters for paussine systematics, particularly of the mouthparts, eyes and structural aspects of the head capsule. We also extracted select characters and corresponding states from previous studies (e.g. Darlington, 1950; Nagel, 1987; Di Giulio *et al.*, 2012) and modified them as necessary.

The head capsule of *Paussus* (Figs 1, 4, 5), particularly the disc, often exhibits pores, grooves and openings from which appeasement chemicals are expelled, often placed atop a frontal horn (Fig. 5). These openings are fed via secretory

cells surrounding paired collecting ducts (A. Di Giulio and J. Robertson, personal observation).

The pronotum (Figs 1, 12) is also modified, with a rudimentary to deep transverse excavation usually housing dense trichome brushes. Prothoracic glands feed secretions through a cuticular sieve-like matrix into the cavity or onto the trichomes (Geiselhardt *et al.*, 2007). Trichomes – specialized setae connected to exocrine glands for the release of appeasement chemicals – are known to occur on nearly all body structures but are most common on the antennae, head, prothorax, elytra and pygidium. Trichomes function to hold, spread and prolong the availability of the associated glandular secretion (Di Giulio *et al.*, 2012), functioning similarly to the way a paintbrush holds its medium.

The elytra (Figs 1, 15) have been shown in at least some species to be filled with secretory glands (Yung, 1938; Geiselhardt *et al.*, 2007; J. Robertson, personal observation). Elytral trichomes most commonly occur along the outer margins, spanning the lateral margins and/or at the apices of the elytra.
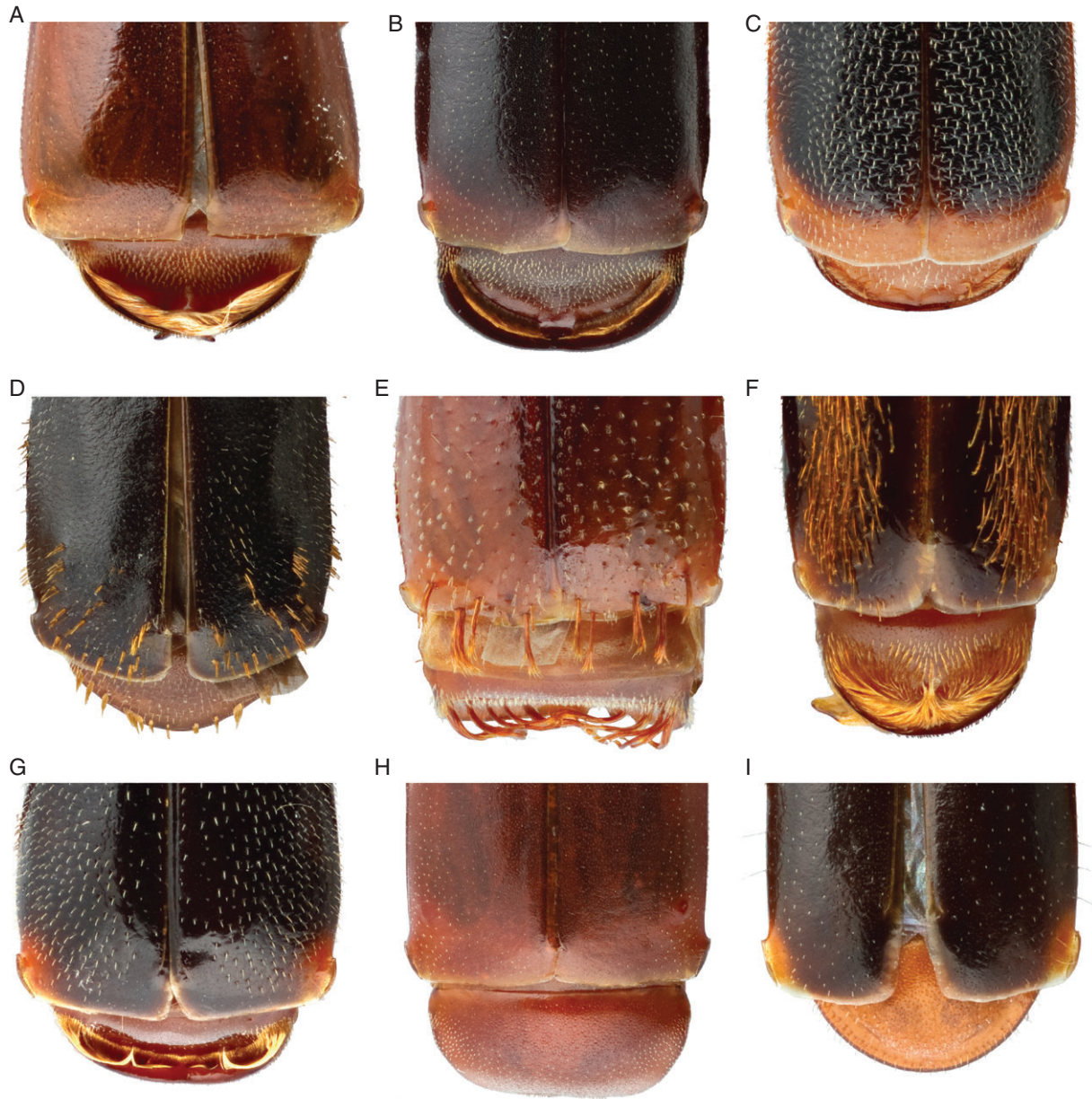
**Fig. 15.** Dorsal aspect of eyltra (apical half) and pygidium. (A) *Paussus cucullatus*; (B) *P. leroyi*; (C) *P. afzelli*; (D) *P.* sp.; (E) *P. armicollis*; (F) *P.* sp.; (G) *P.* sp.; (H) *P. curtisi*; (I) *P.* nr. *planifrons*.

The pygidium (Figs 1, 15, 16) is often modified, with an upturned, explanate outer margin forming a dorsal concavity that may or may not be equipped with trichomes of all varieties and arrangements.

In contrast to previous taxonomic studies of *Paussus* (e.g. Jeannel, 1946; Luna de Carvalho, 1989), we avoided using characters we thought likely to be under strong selective pressure by host ants and that have been shown to be homoplasious, such as the shape of the highly malleable antennal flagellum (see Moore & Robertson, 2014). The resulting morphological dataset includes 88 characters covering the external (most) and internal anatomy of the adult form. Whereas *Paussus* exhibit diverse external anatomy (Figs 3–16), their genitalic characters are relatively homogeneous with the exception of a few discrete characters. MESQUITE 3.04 (Maddison & Maddison, 2015) was used to host these data.

Morphological terminology largely follows recent studies of Paussinae (Nagel, 1987; Di Giulio *et al.*, 2012, 2014) and Coleoptera as a whole (Lawrence *et al.*, 2011). In addition, we introduce multiple new terms (or refine existing ones) to describe novel character states resulting from the present work (Fig. 3) (e.g. median frontal suture, cephalic openings,
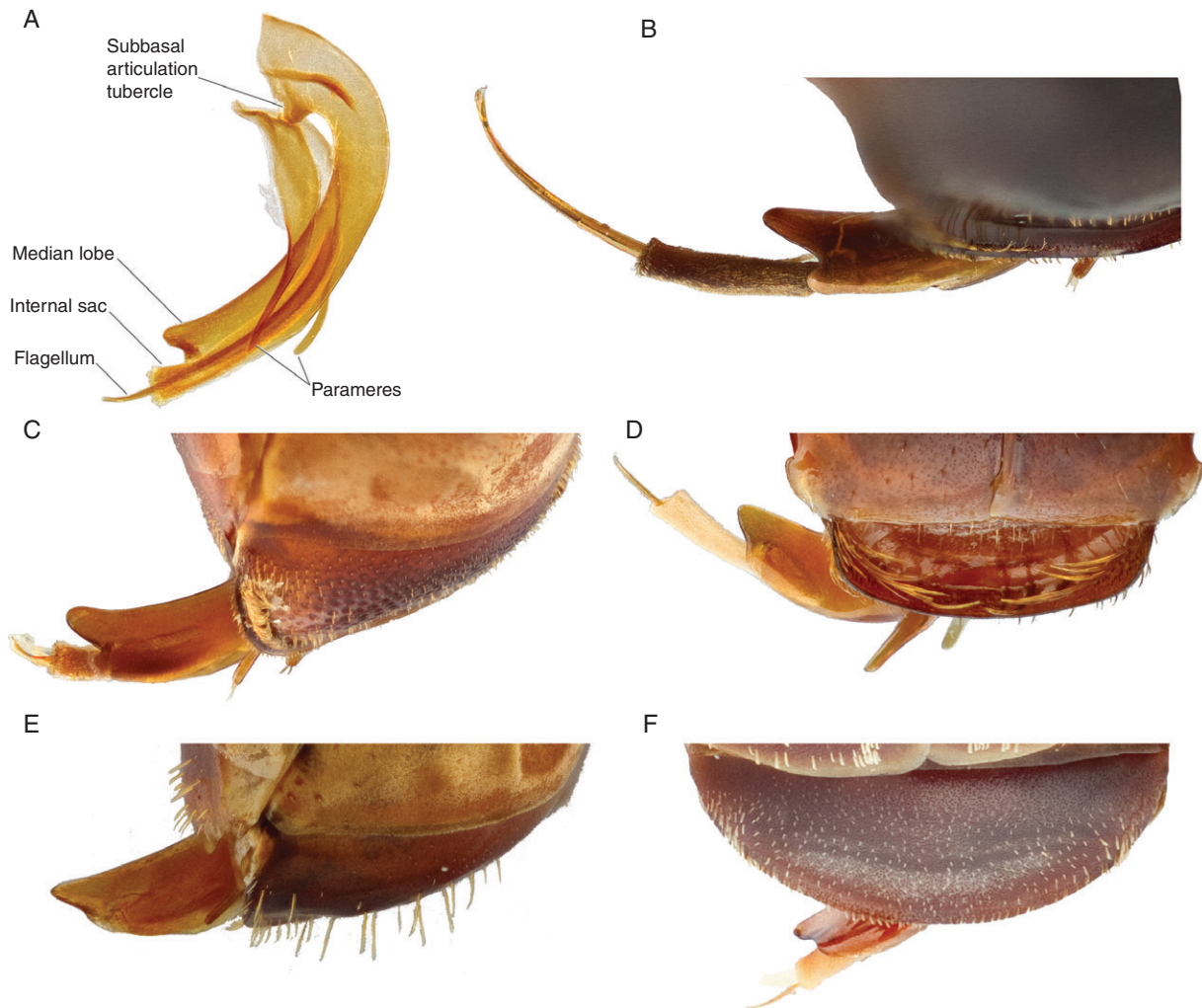
**Fig. 16.** Aedeagus and pygidium (B–F) of male. (A) *Paussus* nr. *serraticornis*; (B) *P. humboldti*; (C) *P. spinicoxis*; (D) *P.* nr. *conradti*; (E) *P.* nr. *dama*; (F) *P. howa*.

tubercle within cephalic opening, admedian setal patches on dorsum of ligula, transverse pronotal cleft, pronotal median endocarina, transverse pronotal endocarina, marginal trichome fringe of pygidium). In the list of characters in Supplementary Information Table S2, we provide an explanation of characters and states; we denote inapplicable characters with 'NA' (not applicable) followed by the relevant character number (e.g. 'C1' = character 1) and state number (e.g. 'S1' = state 1) to which the character/state is inapplicable. Inapplicable characters were scored in the matrix as a '–' and treated as missing data in all analyses.

*Molecular data and phylogeny*

The molecular study of Moore & Robertson (2014) included 88 species representing the taxonomic breadth of the myrmecophilous tribe Paussini. In order to better polarise the anatomical characters states used in the present study, we additionally sampled *Ozaena* as a more distant outgroup within the tribe Ozaenini. Accordingly, we sequenced complimentary molecular markers for *Ozaena* with those used in Moore & Robertson (2014) (*CAD*, *ArgK*, *Wg*, *28S*) following the primers and protocols outlined in that study. We added the *Ozaena* sequences to the molecular matrix of Moore & Robertson (2014) using Opal as implemented in MESQUITE. Sequences new to this study (*Ozaena lemoulti* Bänninger) are deposited in GenBank under accession numbers KU921417–KU921419.

*Phylogenetic analyses*

Phylogenetic analyses are summarized in Fig. 17 and outlined in the following. Bayesian inference (BI) was utilized in the present study to accommodate the statistical phylogenetic analysis of both molecular and morphological data types (see later). The Bayesian analysis of the morphological data alone was implemented in MRBAYES 3.2.2 (Ronquist
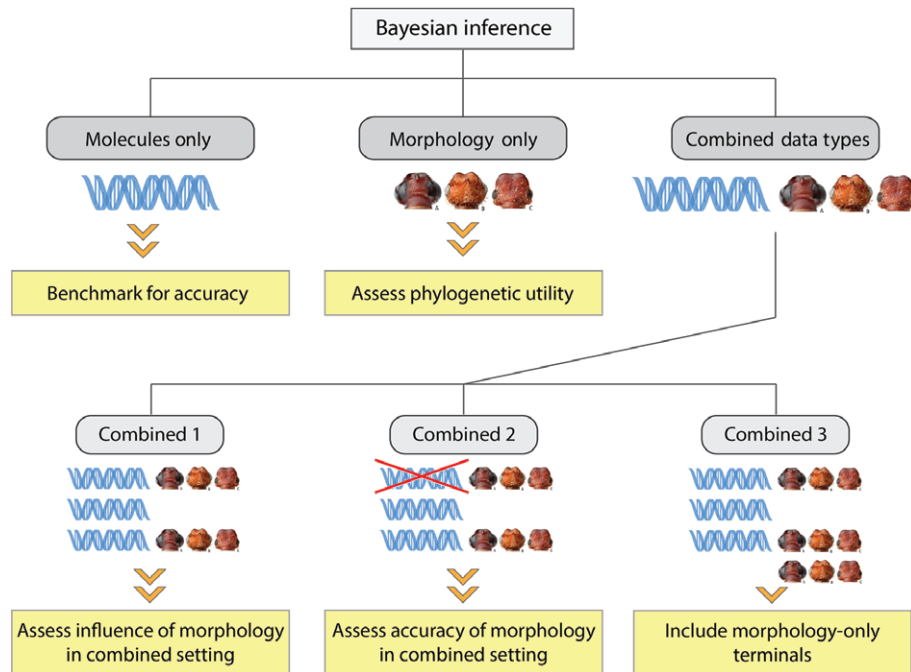
**Fig. 17.** Workflow and summary of phylogenetic analyses. (DNA graphic by Tanya Renner, used with permission.)

& Huelsenbeck, 2003) using a single rate category (Mk1 model; Lewis, 2001) for the entire dataset. BI of the molecular data was achieved using the data subsets and models from Moore & Robertson (2014) (six subsets, GTR + Γ model, generated in PARTITIONFINDER; Lanfear *et al*., 2012). For the combined data type analyses (see later), a mixed-model Bayesian analysis (Nylander, *et al*., 2004) was performed in MRBAYES using the data subsets and models from Moore & Robertson (2014) for the molecular data and a single rate category for the morphology data subset (see earlier). The Bayesian analyses each comprised four separate runs utilizing 20–30 million generations, flat priors, unlinked partitions, four chains (one cold and three hot) and trees sampled every 1000 generations. BI analyses were performed on the Cipres Science Gateway (Miller *et al*., 2010) (http://www.phylo.org/). Convergence and the effective sample size of parameter estimates were checked using the program TRACER 1.5 (Rambaut & Drummond, 2009).

A striking feature of the current suite of molecular markers sequenced for *Paussus* is the high level of concordance observed between the individual gene trees (Moore & Robertson, 2014) (Figure S1). The overwhelming concordance in tree topologies inferred with nuclear protein coding markers, nuclear ribosomal markers, mitochondrial (W. Moore and J. Robertson, unpublished data) markers and the combined molecular data gives us confidence in the resulting pattern of *Paussus* evolutionary history. Thus, we used the tree topology resulting from our combined molecular data (Fig. 18) as a benchmark when assessing the phylogenetic utility and accuracy of our morphological data; it serves to assess the potential of our morphological data to accurately place taxa for which molecular data are not available, including exceedingly rare extant species and fossil taxa.

In order to test the utility of our morphological data for phylogenetic inference and to accurately place taxa for which molecular data are not available, we performed a series of successive combined morphological and molecular data analyses as outlined in the following (see also Fig. 17).

*Combined analysis 1.* For combined analysis 1, we concatenated our morphological data with the molecular data but only included those taxa for which corresponding molecular data were available (some taxa scored for morphology are not represented in Moore & Robertson, 2014). Thus, in combined analysis 1, every terminal included molecular data but not all included morphological data. This resulted in a matrix comprising 89 taxa (same as Moore & Robertson, 2014 + *Ozaena*) and 2644 characters (2556 aligned nucleotide bases + 88 anatomical characters). Note that the molecular dataset of Moore & Robertson contained five species with multiple individuals (two individuals each). In these cases, morphology was added for only a single terminal per species. BI analyses of these data were implemented as outlined earlier. This initial combined analysis allows us to assess the influence of the morphology in a combined data setting; it also serves to establish a comparative baseline for subsequent analyses (see later).

*Combined analysis 2.* To assess the potential of the morphological data to accurately place species of *Paussus* in the phylogeny, we performed a series of combined analyses using the dataset described earlier (combined analysis 1) but selectively removed the molecular data for terminals spanning a diversity
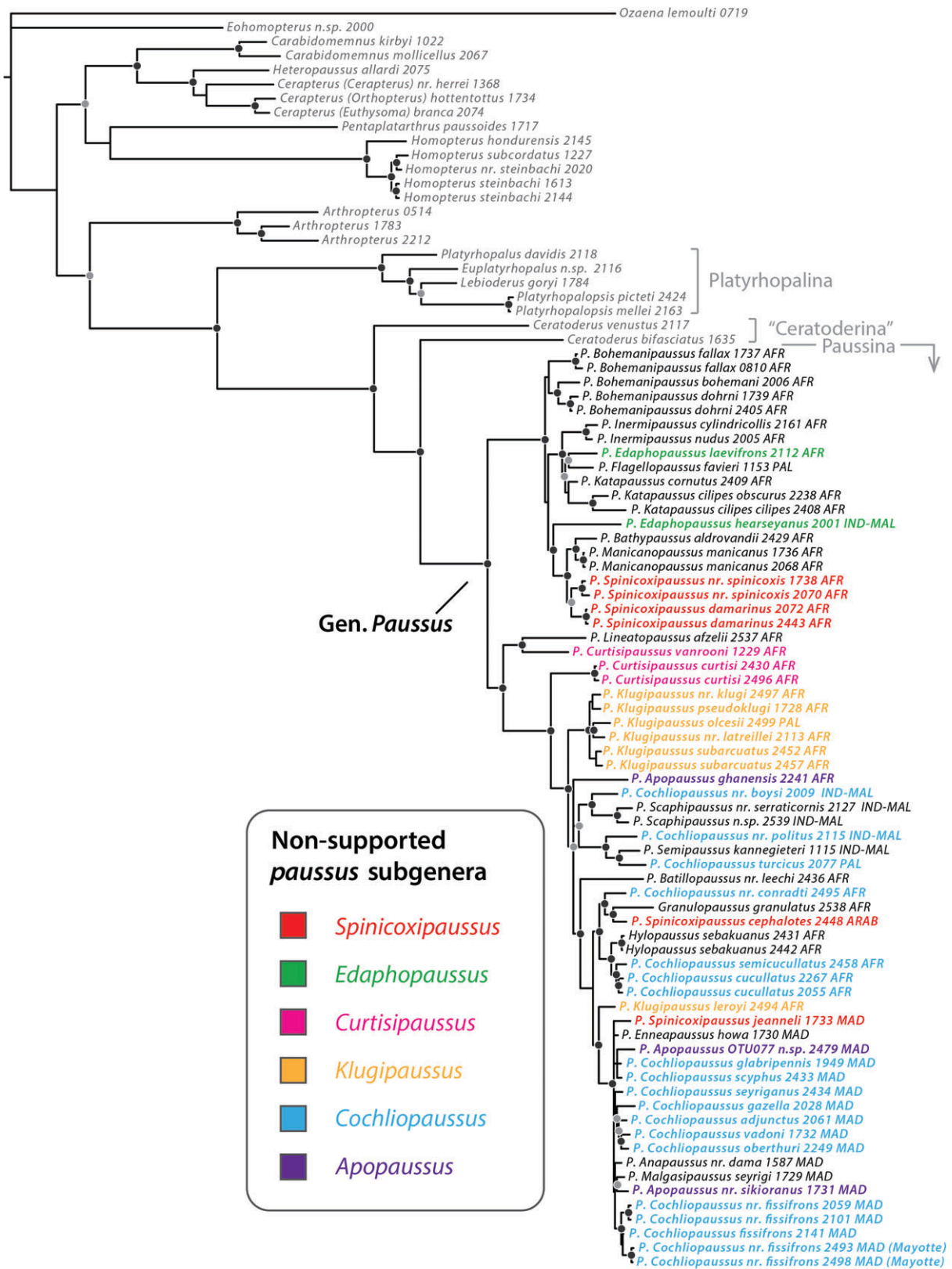
**Fig. 18.** Molecular data *Bayesian inference* tree. Left: terminals coloured by non-monophyletic *Paussus* subgenera; Right: terminals coloured by area of endemism. Dark circles indicate nodes supported with ≥99 Bayesian posterior probability (BPP); light grey circles indicate BPP ≥90 and <99.

of major *Paussus* lineages. Thus for each analysis, the molecular data were removed for a single terminal only – one that originally contained both molecular and morphological data. We attempted to select a range of taxa that we *a priori* predicted to be either likely [high probability placement (HPP) taxa] or unlikely [low probability placement (LPP) taxa] to be accurately placed by morphology alone based on the phenotypic heterogeneity of the terminal species compared with its sister taxon or more inclusive clade. For example, *P. spinicoxis*, *P. nudus*, *P.* nr. *klugi* and *P. cucullatus* are each embedded within or sister to phenotypically similar taxa and were thus predicted to be HPP taxa. Conversely, *P. bohemani*, *P. aldrovandii*, *P. favieri*, *Hylopaussus sebakuanus*, *P. leroyi*, *Granulopaussus granulatus* and *P. kannegieteri* are notably dissimilar in gross morphology compared with their sister taxon or more inclusive clade and were thus considered LPP taxa. Given the extreme phenotypic diversity seen in the Malagasy radiation, we predicted these species would all be LPP taxa. We also included three species that we expected to be moderately difficult to place with morphology alone. Note that the rigour for which the categories were made is not critical for our purposes; we only wanted to ensure that the sampled subset of taxa exhibited a range of probability placement accuracy.

Placement accuracy was assessed by concordance with placement in the combined analysis 1 (molecular + morphological data). Given the significant portion of *Paussus* terminals missing anatomical data (28/65; 43%), we considered taxa as placed accurately if they were recovered ≤3 nodes from their placement in combined analysis 1. The exclusion of the molecular data for select terminals allows us to test the utility of the morphology alone to accurately place taxa in a combined analysis framework. In all, 26 analyses were performed, each with a single, unique terminal having the molecular data removed.

*Combined analysis 3.* If, in general, the morphological data alone could accurately place taxa in a combined analysis (see earlier) (assessed by concordance of combined analysis 1 and combined analysis 2) then we included the taxonomically important or enigmatic taxa for which we scored the morphological dataset but molecular data are not yet available (see earlier; Table S1). The inclusion and placement of *Paussus* (*Paussus*) *oertzeni* Kolbe are of particular interest as its placement bears on nomenclatural and taxonomic actions. Belonging to the phenotypically diverse Malagasy fauna, the placement of *P. solidus* is similarly of great interest.

### Appraisal of anatomical synapomorphies

WINCLADA (Nixon, 2000) and MESQUITE 3.04 (Maddison & Maddison, 2015) were used to optimize morphological characters on the combined data phylogeny (combined analysis 3) using both unambiguous and ACCTRAN optimization. WINCLADA, in particular, lends itself to investigating synapomorphies supporting nodes, as it allows the mapping of all characters and states simultaneously. When discussing anatomical node

support, we use a standard notation, listing the character number followed by a dash and the number for the relevant state. For example, character 3 (median frontal suture) state 1 (absent) would be listed as 3-1. For uncontroverted synapomorphies (i.e. non-homoplasious states), an asterisk follows the character state (e.g. 3-1*). When discussing statistical support for resulting relationships, the Bayesian posterior probability is listed in parentheses following the relationship of interest [e.g. (95)]. When only a specific data type is relevant, it is listed following the support value where 'morph' = morphological data alone, 'mol' = molecular data alone and 'comb' = combined morphological and molecular data [e.g. (95 morph)].

## Results

The scored morphological matrix can be found in Table S3. Taxa listed in bold were used in combined analysis 2. We only had a single specimen of the rare *P. cephalotes* (known only from the Hijaz Mountains, Saudi Arabia) available for study and were not able to fully dissect it. As such, we were unable to observe and score eight characters of the mouthparts.

### Morphological data alone

Bayesian analysis of the morphological data alone (Fig. 19) resulted in a tree with backbone relationships mostly concordant with the molecular results of Moore & Robertson (2014) but with far less resolution. The Paussini genera *Carabidomemnus* Kolbe, *Heteropaussus* Thomson, *Cerapterus* Swederus, *Homopterus* Westwood, *Pentaplatarthrus* Westwood and *Arthropterus* MacLeay were supported as early diverging taxa within Paussinae. *Paussus* was strongly supported (96) as monophyletic only with the inclusion of *Hylopaussus* and *Granulopaussus*, a result consistent with molecular data (Moore & Robertson, 2014). *Ceratoderus bifasciatus* (Kollar) formed the sister group to *Paussus* (86), and *Platyrhopalopsis* Desneux subtends this clade (99). Within *Paussus*, species were largely recovered within two major clades: one comprises species formerly assigned to the subgenera *Edaphopaussus* Kolbe, *Spinicoxipaussus*, *Katapaussus* Wasmann, etc. (95); the other includes taxa previously recognized in the subgenera *Paussus*, *Cochliopaussus* Kolbe, *Klugipaussus* Kolbe, *Scaphipaussus* Fowler, *Apopaussus* Kolbe, etc. (77). *P. afzelii* Westwood was recovered as the sister group to the second major *Paussus* clade and the position of *P. vanrooni* Wasmann was unresolved with respect to these major clades in *Paussus*. *P. oertzeni* was strongly supported (91) as the sister group to *P. curtisi* Westwood and *P. africanus* Luna de Carvalho. *Hylopaussus* was strongly supported (98) in a clade with *P. cucullatus* Westwood, *P. semicucullatus* Wasmann and *P.* nr. *conradti*. *Granulopaussus* was recovered in a poorly supported clade (60) with *P. howa* Dohrn and *P. turcicus* Frivaldsky von Frivald, a result not concordant with molecular data. *P. jeanneli* was recovered as the sister taxon to *P.* nr. *dama*, and *P. solidus* was recovered as the sister group to *P. scyphus* Raffray (63).

*Combined analysis 1*

Combining the morphological data with the molecular dataset, including only terminals from the morphological dataset for which corresponding molecular data was also available (see Fig. 20) (combined analysis 1), resulted in a highly resolved and supported topology that is almost entirely in agreement with the results from the molecular data alone (Moore & Robertson, 2014) (differing only in the resolution of the three main lineages within a main *Paussus* clade comprising *Edaphopaussus* and relatives and the placement of *P. ghanensis* one node away). This is not surprising given both the overwhelming signal contributed by the molecular data and the general concordance of the morphological results (despite the general lack of resolution) with the molecular topology.

*Combined analysis 2*

When the molecular data were removed successively for individual taxa with both data types (combined analysis 2), we found that in the majority of instances the morphology alone was sufficient to accurately place species (Table S4). Indeed, 18 of the 26 species tested (*c.* 70%) were recovered zero to three nodes away from their placement by combined molecules and morphology despite the large portion (43%) of *Paussus* terminals without anatomical data. Seventeen of the 26 species tested (65%) were accurately recovered in their correct subgeneric clade. All five of the HPP taxa, two-thirds of the moderate probability taxa and, surprisingly, 10 of the 18 LPP taxa were accurately recovered in the absence of their molecular data subset. The results of the combined analysis 2 are summarized in Table S4 and discussed below.

*Combined analysis 3*

We included in our morphological study five species for which molecular data were not available*: Paussus oertzeni, P. africanus, P. solidus, P. dissimulator* and *P. humboldti* Westwood. When included in our final combined analysis (combined analysis 3), four out of five of these terminals were recovered in the topology with strong support and precision (Fig. 20). Both *P. oertzeni* and *P. africanus* were strongly supported (100, 100) as forming a clade with *P. curtisi*. The Madagascar endemic, *P. solidus*, was recovered within the Malagasy radiation despite general phenotypic similarities shared with African consubgeners, a result consistent with those of Moore & Robertson (2014). *P. dissimulator* was strongly supported as the sister group to *P. cilipes cilipes* Westwood (100) and nested within a well-supported (100) clade comprising other species formerly assigned to the subgenus *Katapaussus*. In contrast, *P. (Katapaussus) humboldti* was recovered as the sister group to one of the major clades of *Paussus* with strong support (100); however, its exact placement within this series is uncertain as reflected in the poor support spanning the backbone of this series clade. The topologies from combined analysis 1 and combined analysis 3 are entirely concordant.

**Discussion**

*Phylogenetic utility of morphology*

The primary goal of this study was to develop a novel suite of anatomical characters for *Paussus* and test the phylogenetic utility of these data for accurately placing *Paussus* species. Inherent in this objective is the challenge of using morphology in a rapidly, adaptively radiating system with demonstrated phenotypic plasticity, parallelism and ecomorphic evolution (Moore & Robertson, 2014).

Whereas BI analysis of the morphological data alone yielded a topology that was largely concordant with the molecular results (Moore & Robertson, 2014), the data were not particularly decisive at lower levels as evidenced by the general lack of resolution within the major clades of *Paussus* (Fig. 19) and the relatively high degree of homoplasy of anatomical character states in the combined analyses (Figure S2; see below). A few, generally poorly supported, lower-level clades were recovered that are not consistent with the results from either the molecular data alone or the combined data. For instance, *Granulopaussus granulatus* formed a poorly supported clade (60) with *P. turcicus* and Malagasy *P. howa. Granulopaussus granulatus* and *P. howa* are each anatomically bizarre, and phenotypically unique among *Paussus* (Fig. 1B, L).

Interestingly, the African *P. ghanensis* Luna de Carvalho and Malagasy *P.* nr. *sikoranus* were recovered as sister taxa in the morphology-alone analysis with high support (94). These phenotypically similar, yet distantly related species were both classified in the subgenus *Apopaussus* (Kolbe, 1935; Luna de Carvalho, 1989). The results from molecular data (Moore & Robertson, 2014) as well as the combined data unequivocally demonstrate that their similarity is superficial, a result of parallel or convergent evolution. *P.* nr. *leechi*, an additional African species similar in phenotype to *P. ghanensis* and *P.* nr. *sikoranus*, was strongly supported (93) subtending this clade. Anatomical features shared by these three taxa include the mesal anterior margin of scape produced forward, forming a point (52-0), protibiae distinctly dilated and flattened, parallel-sided (64-0), hind tibiae distinctly dilated and flattened, parallel-sided (65-0), pronotum anterad of transverse pronotal cleft distinctly wider than midline and posterior part of pronotum (71-2), and dense marginal trichome fringe of pygidium absent (83-1).

It is noteworthy that several of the above features could be ecotypic for these species (and their shared general phenotype), bearing more ecological than evolutionary significance. Whereas these character states are clearly homoplasious for these phenotypically similar species, they are phylogenetically informative for different taxa at various taxonomic levels. For example, prothoracic tibiae normal, not distinctly dilated and flattened (64-2), lends support for the monophyly of *Paussus*. The form of the hind tibiae (C65) is informative for several well-supported clades, including the lineages comprising *P.* nr. *klugi* and allies (65-1) and *P. conradti, P. cucullatus et al.* (65-1) (both clades treated as subgenera below). Having the mesal anterior margin of scape produced forward, forming a point (52-0) is not informative above the subgenus level but it is probably
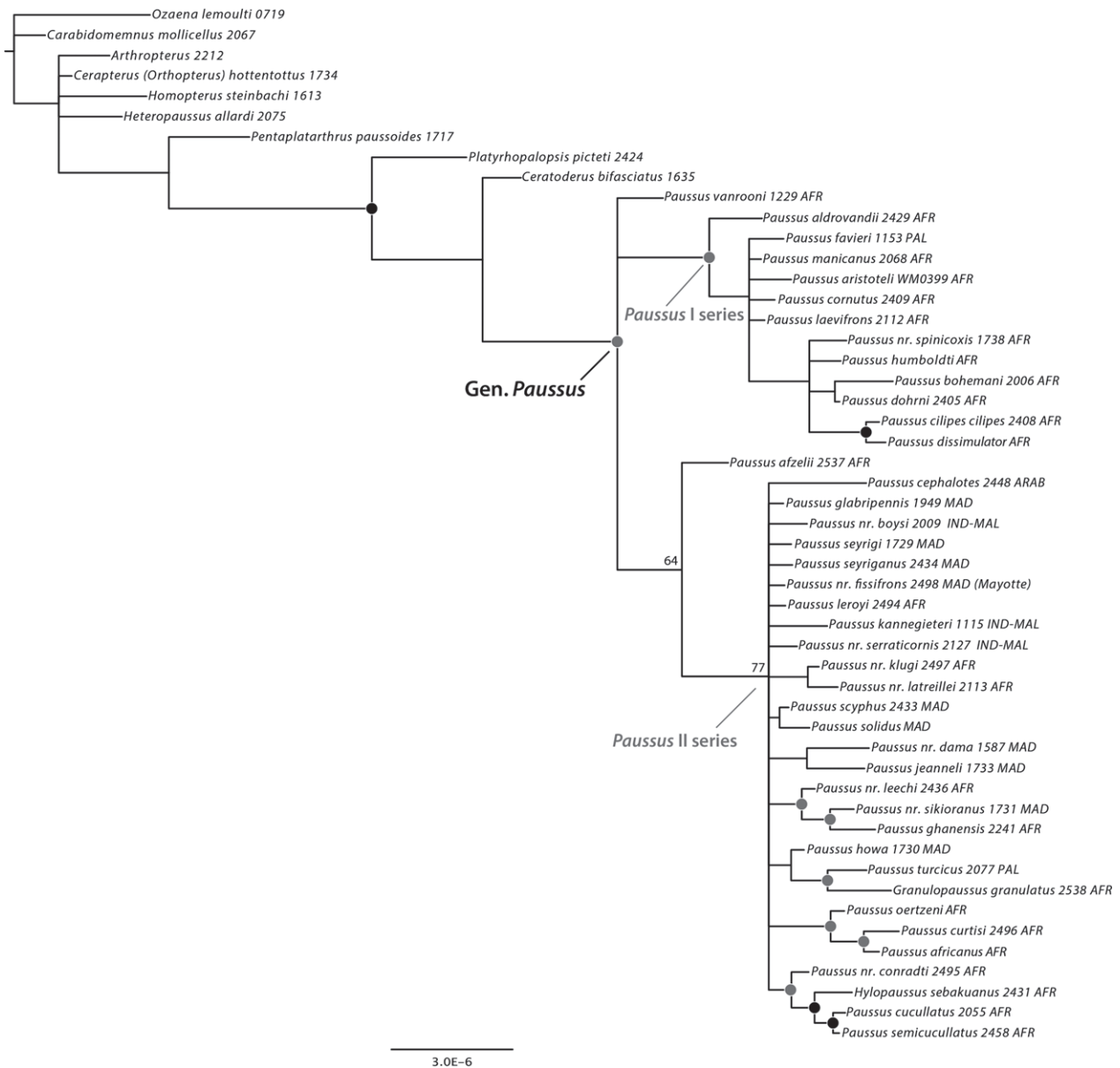
**Fig. 19.** Bayesian inference tree of morphological data. Dark circles indicate nodes supported with ≥99 Bayesian posterior probability (BPP); light grey circles indicate BPP ≥ 90 and <99; BPP is indicated when <90 for select nodes of interest.

informative at lower levels (e.g. species groups) not captured in the present taxonomic sampling [e.g. the Malagasy *P. sikoranus* species group (Jeannel, 1946); J. Robertson and W. Moore, personal observation]. Thus in some cases, whether a given character state is informative or ecomorphic can be relative.

Even so, several of the well-supported *Paussus* clades recovered in the combined data analyses (Figs 19, 20) were recovered with strong support in the analysis of the anatomical data alone, including *P. oertzeni*, sister to *P. curtisi + P. africanus* (91); *Hylopaussus sebakuanus* sister to *P. cucullatus + P. semicucullatus* (100) with *P. conradti* subtending this clade (98); *P. cilipes cilipes + P. dissimulator* (100). The placement of *Hylopaussus*

*sebakuanus* deeply nested within *Paussus*, adjacent to *P. cucullatus*, *P. semicucullatus* was one of the surprising results from Moore & Robertson (2014) and is not consistent with any previous view of paussine relationships and classification (Jeannel, 1946; Luna de Carvalho, 1989; Nagel, 2003). That the morphology alone recovered this clade with high support underscores the phylogenetic power and utility of the morphological dataset.

Results of our combined analyses with selective removal of molecular data for terminals with both molecular and morphological data types (combined analysis 2) demonstrate the utility of the morphology alone in accurately placing taxa in a combined data type context. Not only were the majority of taxa

**Fig. 20.** Combined data type Bayesian inference tree (note that the results of combined analyses 1 and 3 are entirely concordant). Terminals with morphological data are coloured black. Black arrows indicate terminals for which only morphology was available and were included only in combined analysis 3. All terminals except those with black arrows have molecular data. Dark circles indicate nodes supported with ≥99 Bayesian posterior probability (BPP); light grey circles indicate BPP ≥ 90 and <99.

accurately placed with morphology alone, but ten were recovered in the exact same placement as when the molecular data were included for these terminals (combined analyses 1 and 3) (Table S4). (Note that for *P. cephalotes* eight characters of the mouthparts were missing which most likely had a bearing on this taxon not being accurately placed in combined analysis 2).

Moreover, the molecular data included five pairs of conspecific terminals (i.e. five species with two individuals each). Whereas all five of these species were included in our morphological matrix, we only added morphological data in the combined analyses for one conspecific terminal per pair. Of these five species, four were included in the combined analysis 2 such that one conspecific included molecular data only, whereas the other terminal only had the morphological data included. Interestingly, three of these four were accurately placed and surprisingly two conspecific species pairs (*P. cucullatus*, *P. curtisi*) were each recovered in the exact same placement as when the molecular data were included for both terminals (combined analyses 1 and 3), i.e. as monophyletic species.

Whereas in general the morphological data alone were accurate in placing taxa in a combined morphology and molecules setting, combined analysis 2 revealed potential shortcomings of the anatomical data. For example, in a few cases where the *Paussus* phenotypes were either ecomorphic (e.g. *P.* nr. l*eechi*, *P. ghanensis*, *P.* nr. *sikoranus*; see earlier) or extraordinarily unusual (e.g. *P. howa*, *P. granulatus*) the morphology alone was not accurate. Even so, *P. klugi* Westwood and *P. leroyi* Wasmann, *P. bohemani* and *P. spinicoxis*, and *P. cucullatus* and *P. glabripennis* Jeannel represent three additional ecomorphic species pairs, and yet each of these species was accurately placed in combined analysis 2 by morphology alone. Similarly, in contrast to *P. howa* and *P. granulatus*, additional phenotypic anomalies such as *P.* nr. *dama* and *Hylopaussus sebakuanus* were accurately recovered solely by anatomical data in combined analysis 2. This trend is particularly evident with several phenotypically divergent Malagasy taxa that were unexpectedly recovered in the Malagasy clade based on the morphological data alone in combined analyses 2 (Table S4) and 3. An interesting pattern borne out from the morphology alone and in combined analysis 2 is that when phenotypes are particularly divergent, these taxa were often recovered in the Indo-Malayan clade – a lineage strongly supported by molecules but not well characterized from an anatomical standpoint (see later). The Southeast Asian fauna is not particularly well known and our sampling to date is unrepresentative of the actual diversity found there. Perhaps as additional anatomical synapomorphies for this lineage are illuminated and the group becomes better characterized, this phenomenon will dissipate.

While intuitive, it is worth noting that the utility of morphological characters to place taxa phylogenetically relies upon the inclusion of closely related taxa (e.g. within the same subgeneric clade). If the number of terminals for which anatomical data are available is minimal (e.g. 0-1) in a given subgeneric clade then there is very little for the morphology to act on in the analysis. Thus the accuracy of the morphology is dependent upon having morphological data available for a representative sampling within a subgeneric clade for a closely related query species

with scored morphology to be placed accurately. This underscores the ongoing need to expand our taxon sampling for our morphological matrix. Of the five terminals lacking molecular data included in combined analysis 3, all but one were recovered with high topological and anatomical character support at the series, subgenus and lower levels (Fig. 20). The one exception was *P. humboldti*, which was at least strongly supported at the series level, recovered within the *Paussus* I series with maximum support (100) (see later). Taken together, the results of the morphological and combined data analyses reveal the circumstances under which the anatomical data may not be accurate, but overall showcase the power and utility of the morphological data in accurately placing *Paussus* taxa.

Due to their obligate myrmecophily, some *Paussus* species are seldom collected. For example, several *Paussus* species have not been collected more recently than 100 years ago! Therefore, it is unrealistic to hope to include all known species in our molecular dataset. For now, our ongoing goal to infer the evolutionary history of *Paussus*, including all species, must rely to a certain extent upon morphological data. In addition, our ability to accurately place fossil taxa for use as calibration points in divergence time estimation relies upon a phylogenetically informative morphological framework for Paussinae. Despite the misleading past hypotheses of *Paussus* relationships based on gross phenotype (Jeannel, 1946; Luna de Carvalho, 1989; see earlier), we have developed a novel suite of anatomical characters for *Paussus* that are phylogenetically informative. One of the primary goals of this study was to assess the phylogenetic utility of morphology in a group containing several instances of rapid, adaptive radiation with demonstrated phenotypic lability, parallelism and ecomorphic evolution. By generally ignoring the superficial, gross phenotypic features tied to beetle-ant symbioses – the ecotypic/ecological characters (e.g. antennal shape) – and instead delving into novel character systems less likely to be under selective pressure by their host ants, such as the mouthparts, we have illuminated a suite of characters that are informative at multiple levels within Paussini, including across *Paussus* diversity.

### Recovered clades

The results of the present study and of Moore & Robertson (2014) strongly indicate the need for a new subgeneric classification for *Paussus*, one that reflects their evolutionary history. In the following we establish a new classification for *Paussus* based on the combined data results, using strongly supported clades as the framework for our proposed groupings. We implement the following hierarchical, informal and formal ranks subordinate to genus – series > subgenus > species group > species – with most of our classification actions targeting the genus, series and subgenus levels. With the current morphological data we are largely able to place species confidently within *Paussus* series and subgenera (Table S5); classification on a finer scale (e.g. species groups) will, in many cases, require additional data. Because we have not been able to examine or dissect the majority of *Paussus* species, our classification includes some species

that are tentatively treated as *incertae sedis* at the series and/or subgenus (most) levels. Note that our taxonomic actions do not supplant existing taxonomic groups on a finer scale not treated here, but rather complement and allow for stability of existing species groups (e.g. Madagascan species groups, Jeannel, 1946; miscellaneous African species groups: Nagel, 1977, 1980, 1986, 1987, 2006). Indeed, our intent is to provide a robust working scaffold to facilitate the ongoing classification of all *Paussus* species and the revision of historical, and circumscription of new, subgenera and species groups. Our phylogenetic, character-based classification (see Table S5; Fig. 21; Figure S2) as implemented in the following provides the necessary framework for future systematic work.

### Platyrhopalina, Ceratoderina, Paussina

*Platyrhopalopsis*, *Ceratoderus* and *Paussus* represent exemplar taxa of the subtribes Platyrhopalina, Ceratoderina and Paussina, respectively, and together comprise an informal, but long recognized group within Paussini for exhibiting similar behavioural and morphological strategies for integration into the ant society (Nagel, 1987; Geiselhardt *et al.*, 2007). This clade was strongly supported in all our analyses (100) and is supported by several anatomical features, including the median frontal suture present (3-0) (Figs 3–5), frontoclypeogenal suture present (8-1*) (Fig. 5E), oral cavity 'closed' (Nagel, 1987) (20-0*) (Fig. 6B–H), maxillary palpomere II longer than or equal to III and IV combined (24-0) (Figs 7H, I, 8B, 9B) (reversal in *Edaphopaussus* series), posterior margin of lacinia without spines or setae (34-1) (Figs 7G–I, 8A, B) (independently occurs in *Homopterus*), antennal flagellum fused (56-2*) (Fig. 10), antennal flagellum carinate along entire inner margin (59-0) (Fig. 5B, D–F), notosternal sutures absent (73-1) (Fig. 13C, D) (independently occurs in *Homopterus*), pleurosternal sutures absent (76-1*) (Fig. 13D–F), prosternal process entirely obliterated (79-1) (Fig. 13D, F) (widespread subsequent reversals), and postcoxal projections meeting, broadly fused (80-2) (Fig. 13D–F) (independently occurs in *Pentaplatarthrus*). Other character states lend support to this clade [e.g. (74-2)], but these are shared only by the earliest diverging lineages.

### Sister group of Paussus

Given the striking tempo and pattern of *Paussus* diversification (Moore & Robertson, 2014), the sister group of *Paussus* is of great interest. The taxon sampling here does not permit a thorough test of the sister group of *Paussus* as many viable candidates were not included (e.g. *Melanospilus* Westwood, *Leleupaussus* Luna de Carvalho, *Paussomorphus* Raffray) due to their rarity. The genus *Ceratoderus* Westwood as presently constituted is not supported as a monophyletic taxon based on molecular evidence (Moore & Robertson, 2014). Interestingly, Maruyama (2014) recently treated the species of *Ceratoderus* and noted two morphologically cohesive groups in the genus: one comprising species similar to *C. bifasciatus* (four spp. total) and the second similar to *C. venustus* Hisamatsu (two spp. total). In the

present study, we only scored morphology for *C. bifasciatus*, which is recovered as the sister group to *Paussus* with strong support [87 morphological (morph); 100 combined (comb)] with *C. venustus* subtending this clade (100), rendering the genus paraphyletic. However, we note that in contrast to *C. venustus* and the remaining outgroup taxa in our study, *C. bifasciatus* lacks tibial spurs on the mid-tibiae (63-1), a character state that is phylogenetically significant for higher-level Paussini relationships in the present study (see also Darlington, 1950). Anatomical features supporting the monophyly of *C. bifasciatus* + *Paussus* include apex of lacinia bifalcate (30-1*) (Figs 7G–I, 8A, B), median anterior margin of submentum pointed, triangular (36-1*) (Figs 6B–H, 9B), transverse pronotal cleft present (68-0) (Figs 12A–L, 14A, B), pronotal median endocarina absent (77-2) (Fig. 12A–C, E, F, J–L), and sub-basal articulation tubercle of median lobe present (87-0) (Fig. 16A). Additional anatomical features supporting the clade comprising *C. bifasciatus* and *Paussus* but shared primarily by the earliest diverging lineages include (4-1) and (88-0).

### Genus Paussus

Support for the monophyly of *Paussus* is strong across all analyses [96 morph; 100 molecular (mol), comb] but only with the inclusion of *Hylopaussus* and *Granulopaussus*, a result consistent with and initially revealed by molecular data (Moore & Robertson, 2014). Previous authors (Reichensperger, 1930; Jeannel, 1946; Luna de Carvalho, 1989; Nagel, 2003) considered both genera to lie outside of *Paussus*, being sufficiently bizarre anatomically to merit recognition of each at the generic level [though Darlington (1950) is a notable exception)]. However, even morphology alone strongly supports their placement within *Paussus* as they exhibit all synapomorphies of the genus and hierarchically less inclusive clades (see later). The monophyly of *Paussus* is supported by several anatomical features, including anterior (outer) margin of lacinia with row of elongate setae (31-0*) (Figs 7G–I, 8A, B), mentum obliterated medially, reduced to two separated lateral lobes (35-2*) (Figs 6C–H, 9B) (not shown with unambiguous optimization due to missing data in *Heteropaussus*), labial palpomere 3 distinctly longer (more than 1.5×) than 1 and 2 combined (39-0*) (Fig. 7N–U), ligula robust, distinctly thick dorsoventrally (at least 1.5× thickness of terminal palpi) (43-0*) (Fig. 8D), admedian setal patches on dorsal surface of ligula present (51-0*) (Fig. 8E, F), prothoracic tibiae not distinctly dilated and flattened (64-2) (Fig. 11C) (with a few subsequent transitions to alternate states), hind tibiae not distinctly dilated and flattened (65-2) (Fig. 11G) (with multiple subsequent transitions to alternate state), prothoracic trichomes well developed (70-2) (Fig. 12A, B, G, I, J, L) (with secondary reductions in select taxa), scraper of stridulatory organ on abdominal ventrite 1 present (81-1*) (Fig. 14C and D), posterior dorsal margin of pygidium distinctly upturned and explanate (82-0) (Fig. 15A–C, F–G, I), and pygidium with dense marginal trichome fringe present (83-0) (Fig. 15A–G).

Within *Paussus*, we recovered three well-supported and anatomically distinctive clades that we treat at the series rank: *Paussus* I, *Paussus* II and *Paussus* III (see later). It is noteworthy

that in his Hennigian treatment of Paussinae, Darlington (1950) proposed two subtribes, Paussina I and Paussina II, that largely overlap with the *Paussus* I and *Paussus* II clades recovered in the present study. Further, some of the general features that Darlington (1950) outlined supporting these subtribes were found to be informative at this taxonomic level, including the relative size of the eyes, the nature of the prothoracic cleft, the occurrence of tibial spurs, and the presence of apical setae on the male parameres.

### Revised classification

**Paussus L. 1775**
*Hylotorus* Dalman, 1823: 103 **syn.n.**
  *Granulopaussus* Kolbe, 1938: 26 **syn.n.**
  *Falcopaussus* Darlington, 1950: 103.
  *Hylopaussus* Luna de Carvalho, 1989: 984 **syn.n.**

*Type species. Paussus microcephalus* L. 1775.

*Diagnosis.* Adults with median frontal suture present (3-0), frontoclypeal suture absent (7-1), frontoclypeogenal suture present (8-1), often with cephalic openings on disc of head (9-1, 9-2), oral cavity more or less concealed ventrally by the labial prementum and ligula and maxillary palpi (20-0), labrum distinctly wider than long (21-0), galea absent (29-1), apex of lacinia bifalcate (30-1), anterior (outer) margin of lacinia with row of elongate setae (31-0*), mentum obliterated medially, reduced outwardly to two separated lateral lobes (35-2*), median anterior margin of submentum pointed, triangular (36-1), labial palpomere III distinctly longer (more than 1.5×) than 1 and 2 combined (39-0*), ligula robust, distinctly thick dorsoventrally (at least 1.5× thickness of terminal palpi) (43-0*), admedian setal patches on dorsal surface of ligula present (51-0*), pedicel highly reduced and recessed in cavity of scape (55-1), antennal flagellum composed of one free flagellomere (56-2), sutures along entire circumference of fused flagellum clearly demarking previous flagellomeres absent (57-1), prothoracic tibiae not distinctly dilated and flattened (64-2) (with exceptions), hind tibiae not distinctly dilated and flattened (65-2) (with multiple exceptions), transverse pronotal cleft present, including vestige (68-0), prothoracic trichomes well developed (70-2) (with secondary reductions in select taxa), notosternal sutures absent (73-1), notopleural sutures incomplete or absent (74), pleurosternal sutures absent (76-1), postcoxal projections meeting, broadly fused to each other (80-2), scraper of stridulatory organ on abdominal ventrite 1 present (81-1*), posterior dorsal margin of pygidium distinctly upturned and explanate (82-0), and pygidium with dense marginal trichome fringe present (83-0) (exceptions).

*Included taxa.* The genus *Paussus* includes all genera, subgenera and species formerly classified as *Paussus* by Nagel (2003) in addition to *Hylopaussus* **syn.n.**, *Hylotorus* **syn.n.**

and *Granulopaussus* **syn.n.** and their constituent species: *Paussus sebakuanus* (Péringuey) **comb.n.**, *Paussus gracilis* (Reichensperger) **comb.n.**, *Paussus bucephalus* Gyllenhal, *Paussus caroli* (Reichensperger) **comb.n.**, *Paussus uelensis* (Reichensperger) **comb.n.**, *Paussus hottentottus* (Westwood) **comb.n.**, *Paussus blanchardi* (Raffray) **comb.n.**, *Paussus basilewskyi* (Luna de Carvalho) **comb.n.**, *Paussus granulatus* Westwood, *Paussus sankuruensis* Reichensperger, *Paussus leleupi* (Reichensperger) **comb.n.**, *Paussus reichenspergeri* (Luna de Carvalho) **comb.n.**

### Key to *Paussus* groups

Note that many species in our new classification are currently treated as *incertae sedis* due to their uncertain position (see Table S5) and are thus not accounted for in the following key to the *Paussus* groups delineated herein.

1. Mesotibiae with one or two apical tibial spurs (Fig. 11D–F) … . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . .**2**
 1′. Mesotibiae without apical tibial spurs . . . . . . . . . . . . . . . . .**3**
2(1). Maxillary palpomere II not distinctly wider than following palpomere (<2× width of palpomere III) (Figs 7E–H, 8A, 9A) and/or shorter than palpomeres III and IV combined (Figs 7E–G, 8A, 9A); labial palpomere III distinctly narrow, elongate and parallel-sided (Figs 6C, 7N–O); usually with large eyes (representing a significant portion of head capsule) (Figs 4A, D, J, K, 5B–E) and mesal eye emargination present (Fig. 5D, E); lateral apical margins of ligula usually with distinct, tight cluster of setae forming a brush (Figs 7N–O, 8C, E); . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . . **4** (*Paussus* series I).
 2′. Maxillary palpomere II distinctly wider than following palpomere (≥2× width of palpomere III) (Figs 7I, 8B, 9B) and longer than or equal to palpomeres III and IV combined (Figs 7H, I, 8B, 9B); labial palpomere III distinctly wide, flattened and spade-shaped (Fig. 7P, R, S); eyes less prominent, occupying a much smaller portion of head capsule (Fig. 4C, E–I) and lacking mesal eye emargination (Fig. 5F, G); lateral apical margins of ligula with setae not placed in distinct, tight cluster, rather setae spread out (Fig. 7Q and 8D, F); *Paussus* series III: Sg. *Lineatopaussus* **stat. rev. sensu n.**
3(1′). Eyes large (representing a significant portion of head capsule) (Figs 4A, D, J, K, 5B–E); distinct temples absent (see characters 1-2) (Fig. 4A, D, K); maxillary palpomere II shorter than palpomeres III and IV combined (Figs 7E–G, 8A, 9A); labial palpomere III distinctly wide, flattened and spade-shaped (Fig. 7P, R, S); posterior dorsal margin of pygidium not distinctly upturned rather confluent with remaining portion of tergite (Figs 1B, 15H); … *Paussus* series III: Sg. *Shuckardipaussus* **stat. rev. sensu n.**
 3′. Eyes typically less prominent, occupying a much smaller portion of head capsule (Fig. 4C, E–I); temples variable but usually distinct (see characters 1-2) (Fig 4B, C, E–J); maxillary palpomere II distinctly wider than following palpomere (≥2× width of palpomere III) (Figs 7I, 8B, 9B) and/or longer than or equal to palpomeres III and IV combined (Figs 7H, I, 8B, 9B); labial palpomere III variable; posterior dorsal margin of
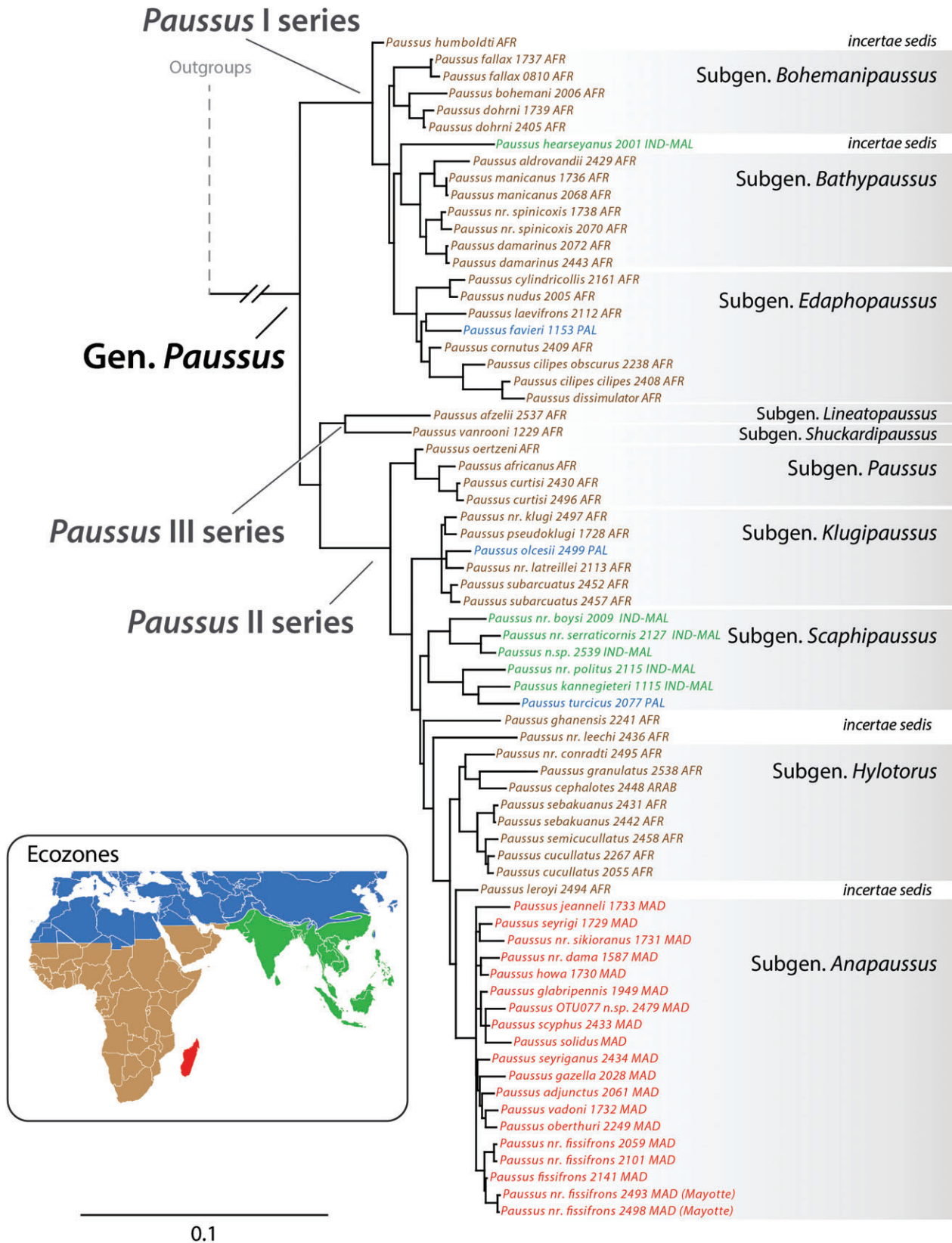
**Fig. 21.** Revised classification of *Paussus* based on the results of combined analysis 3 (Fig. 20). Terminals are coloured by area of endemism.

pygidium often distinctly upturned, explanate (Fig. 15A–C, F, G, I); … **6** (*Paussus* series II).

4(2). Lateral anterior angles of ligula distinctly produced anteriorly (Fig. 7N, O, Q–U); fused flagellum excavated along outer margin (Fig. 10D); … *Paussus* series I: Sg. *Bohemanipaussus* **stat. rev. sensu n.**

4′. Lateral anterior angles of ligula usually not produced anteriorly (Fig. 7J, P); fused flagellum without excavation or distinct concavity (Fig. 10A, B, K, M, N); ................ **5**

5(4′). Transverse pronotal cleft shallow, reduced to a narrow groove or slight constriction (Figs 12D, E, K, 14A); prothoracic trichomes absent (Figs 12D, E, K, 14A); notopleural sutures usually present as a single dot only or apparently absent (Fig. 13F); posterior dorsal margin of pygidium usually not upturned, rather confluent with remaining portion of tergite (Figs 1B, 15H); … *Paussus* series I: Sg. *Edaphopaussus* **stat. rev. sensu n.**

5′. Transverse pronotal cleft variable, shallow to deep and broadly excavated; prothoracic trichomes rudimentary and minute (Fig. 12C, F, H) or well developed (Figs 12A, B, G, I, J, L, 14B); notopleural sutures usually incomplete, extending to anterior margin of procoxal cavities but not beyond posterior margin of coxal cavities (Fig. 13C); posterior dorsal margin of pygidium distinctly upturned, explanate (Fig. 15A–C, F, G, I); … *Paussus* series I: Sg. *Bathypaussus* **stat. rev. sensu n.**

6(3′). Tubercle within and protruding from cephalic openings (Figs 4F, 5A); hind tibiae distinctly dilated and flattened, subtriangular (Fig. 11I); apex of median lobe distinctly emarginate (Fig. 16B, C); antennal club exhibiting a series of tubercles within the outer excavation (Fig. 10O); elytra bearing a dense lateral trichome fringe (Fig. 1G); … *Paussus* series II: Sg. *Klugipaussus* **stat. rev. sensu n.**

6′. Cephalic openings, if present, without internal tubercle; hind tibiae variable; apex of median lobe usually more or less flat, or with indistinct, shallow emargination (Fig. 16D, E); antennal club variable, but not as above; elytra usually not bearing a dense lateral trichome fringe ................... **7**

7(6′). Mesal margin of maxillary palpomere II sinuate, distinctly incised just beyond midpoint (Fig. 7H); maxillary palpomere III distinctly wider than following palpomere, ≥1.5× width of IV (Fig. 7H); labial palpomeres distinctly wide, flattened and spade-shaped (Fig. 7P, R, S); labial palpomere II with socket for palpomere III distinctly shifted to the outer margin of palpomere II (Fig. 7P, R, S); apical median margin of ligula narrowly rounded (Fig. 7R, S); anterolateral margin of scape with trichome brush (Figs 9F, 10C, H, I); … *Paussus* series II: Sg. *Paussus* **stat. rev. sensu n.**

7′. Mesal margin of maxillary palpomere II strongly rounded or more or less straight (Figs 7E–G, I, 8B, 9B); maxillary palpomere III usually not distinctly wider than following palpomere, <1.5× width of IV (Figs 7E–G, I, 8B); labial palpomeres usually only moderately expanded or narrowed (fusiform) (Figs 7Q, T, U, 8D); labial palpomere II with socket for palpomere III positioned/centred more or less along midline (Fig. 7N, O, Q); apical median margin of ligula broadly rounded (Fig. 7N O), truncate (Fig. 7P) or with slight medial invagination

or notch at apex (Fig. 7T, U); trichome brush on anterolateral margin of scape absent ................................ **8**

8(7′). Malagasy species; apical medial margin of ligula often with slight medial invagination or notch (Fig. 7T, U); … *Paussus* series II: Sg. *Anapaussus* **stat. rev. sensu n.**

8′. African, South Palearctic, or Indo-Malayan species; apical medial margin of ligula variable but not notched (Figs 7Q, 8D, 9D) ................................................. **9**

9(8′). African species; width/length ratio of gula at narrowest point between 0.6 and 1.0 (shape of gula quadrate) (Fig. 6D, G, H) or less often between 0.3 and 0.5; anterior margin of lacinia sparsely setose, with six or fewer setae (Fig. 8B); … *Paussus* series II: Sg. *Hylotorus* **stat.n. sensu n.**

9. Indo-Malayan (most) or South Palearctic (one) species; width/length ratio of gula at narrowest point between 0.3 and 0.5; anterior margin of lacinia moderately setose (7–19 setae) (Fig. 8A) or distinctly setose (>20 setae) (Fig. 7G); … *Paussus* series II: Sg. *Scaphipaussus* **stat. rev. sensu n.**

### *Paussus I series (PI)*

In all our analyses we recovered a strongly supported major clade within *Paussus* (95 morph; 100 mol, comb) comprising taxa previously designated in the subgenera *Edaphopaussus*, *Spinicoxipaussus*, *Katapaussus*, *Manicanopaussus*, *Flagellopaussus* and other related taxa. PI comprises mostly African species with relatively few South Palearctic and Indo-Malayan species. Anatomical character states supporting this series include maxillary palpomere III with stout, prominent spiniform setae at apex (27-0) (Figs 7G, 8A), labial palpomere II narrower than palpomere I (41-2) (Fig. 9A), anterior medial margin of ligula broadly rounded (47-0) (Fig. 7N, O), lateral apical margins of ligula with distinct, tight cluster of setae forming a brush (50-0*) (Fig. 7N, O, 8C, E) (with two subsequent transitions to alternate state), dorsal trichome brush on basal spur of fused flagellum (60-0*) (Fig. 5D), and ventral trichome brush on basal spur of fused flagellum (61-0) (Fig. 5F). Many other morphological features characterize PI but they are not unambiguously optimized on this node (or are shared with most outgroups except *Platyrhopalopsis* and or *Ceratoderus*). These include: postocular constriction forming distinct temples absent (1-1) (Fig. 4A, D, K), eyes large, representing significant portion of head capsule (16-0) (Fig. 4A, D, J, K), mesal eye emargination present (mesal margin of eye sinuate) (17-1) (Fig. 5F, G), maxillary palpomere II shorter than palpomeres III and IV combined (24-1) (Figs 7E–G, 8A, 9A), maxillary palpomere II with stout, prominent spiniform setae at apex (26-0) (Figs 7G, 8A), anterior margin of lacinia distinctly setose (32-2) (Fig. 7G) and mid-tibia with two apical tibial spurs (63-0) (Fig. 11D–F).

Although their relative position is strongly supported, both *P. bohemani* and *P. aldrovandii* are in some ways anatomically aberrant for PI, lacking many character states that support and characterize the series. For example both have distinct temples present (1-0) (Fig. 4B, C, E–J), median frontal suture clearly not extending to anterior margin of eye (4-0) (Fig. 4B, C, G), eyes less prominent (16-1) (Fig. 4C, E–I) and labial palpomere

II sub-equal in width to palpomere I (41-1) (Fig. 7N, O, U). Also, although strongly supported as part of PI, the internal placement of *P. humboldti* is uncertain, as indicated by the poor support spanning the backbone nodes comprising the series. At present only morphological data are available for *P. humboldti.* Its weakly supported position forming the sister group to the remaining members of the series may be its rightful placement, but alternatively may indicate that the morphology alone is not sufficiently informative to place this species confidently within a subgeneric clade. We presently refrain from assigning *P. humboldti* to a subgenus but defer such action until its placement is better solidified.

Four major lineages were recovered with moderate to high support within PI. We formally recognize three of these at the subgeneric level as follows: *Bohemanipaussus* **stat. rev. sensu n.**, *Bathypaussus* **stat. rev. sensu n.** and *Edaphopaussus* **stat. rev. sensu n.** (see later). The fourth well-supported lineage is represented by a single terminal only, the Indian *P. hearseyanus* Westwood. Our considerations for its classification are outlined below. Whereas support for these subgeneric lineages is high, the relationship between them remains ambiguous with presently available data.

*Paussus I series: Paussus (Bohemanipaussus) sensu n.*

The subgenus *Bohemanipaussus* **stat. rev. sensu n.** was represented by two terminals in our anatomical study, *P. bohemani* and *P. dohrni*, and is supported by the following character states: disc of head with one cephalic opening (9-1) (Fig. 4A, B) (also present in some *Edaphopaussus* **stat. rev. sensu n.** species), lateral anterior angles of ligula distinctly produced anteriorly (45-1) (Fig. 7N, O, Q–U), fused flagellum excavated along outer margin (58-0) (Fig. 10D), and notopleural sutures nearly obliterated, only present as a short vague line (74-3) Fig. 13E). Note that the fused flagellum in *Bohemanipaussus* **stat. rev. sensu n.** is only weakly excavated.

**Subgenus *Bohemanipaussus* Luna de Carvalho 1982**

*Type species. Paussus bohemani* Westwood 1855.

*Diagnosis.* Disc of head with one cephalic opening (9-1), lateral anterior angles of ligula distinctly produced anteriorly (45-1), fused flagellum excavated along outer margin (58-0) (note: the excavation is weak), and notopleural sutures nearly obliterated, only present as a short vague line (74-3). Anatomical features characteristic of the PI also characterize this group (see earlier).

*Included taxa.* The subgenus *Bohemanipaussus* **stat. rev. sensu n.** includes *P. bohemani, P. dohrni, P. fallax* and *P. massarti* Reichensperger.

*Paussus I series: Paussus (Bathypaussus) stat. rev. sensu n.*

Strongly supported in both the molecular (100) and combined (100) analyses, the subgenus *Bathypaussus* **stat. rev. sensu n.** comprises taxa previously classified in the subgenera *Spinicoxipaussus, Manicanopaussus* and *Bathypaussus.* From an anatomical perspective, both the former *Manicanopaussus* and *Bathypaussus* are distinct and may represent monophyletic taxa. The concept of *Spinicoxipaussus,* however, has varied considerably through time (see earlier). The subgenus *Bathypaussus* **stat. rev. sensu n.** is supported only by a few anatomical character states, including eyes without interfacetal setae (18-1) (Fig. 5D) (reversal in *P. aldrovandii*), lateral lobes of mentum nearly parallel sided (37-0) (Fig. 3) and with pronotum anterad transverse cleft not distinctly widened or prominent (71-0) (Fig. 12B) (indicated with DELTRAN).

**Subgenus *Bathypaussus* Wasmann 1929**
*Cultropaussus* Kolbe, 1938: 26

*Type species. Paussus aldrovandii* Gestro 1901.

*Diagnosis.* Eyes without interfacetal setae (18-1) (reversal in *P. aldrovandii*), lateral lobes of mentum nearly parallel-sided (37-0) and pronotum anterad transverse pronotal cleft not distinctly widened or prominent (71-0). Anatomical features characteristic of PI also characterize the group (see earlier).

*Included taxa.* The subgenus *Bathypaussus* **stat. rev. sensu n.** includes *P. aldrovandii, P. cultratus* Westwood and most species previously treated as *Manicanopaussus* and *Spinicoxipaussus* (see Table S5).

*Paussus I series: P. hearseyanus incertae sedis*

The Indian *P. hearseyanus* was recovered sister to *Bathypaussus* **stat. rev. sensu n.** with moderately high support (89). Given the branch length spanning the two, coupled with distinct biogeographic endemism, it stands to reason that *P. hearseyanus* and the remaining Indo-Malayan *Paussus* I species (not sampled here) represent good candidates for forming an additional subgenus within PI. In fact, Luna de Carvalho (1989) treated the Indo-Malayan *Edaphopaussus* species in the subgenus *Transitupaussus* Kolbe. Although not included in our morphological study, *P. hearseyanus* and its Indo-Malayan allies are anatomically distinct from African *Edaphopaussus* and share some of the anatomical features characteristic of their sister group, *Bathypaussus* **stat. rev. sensu n.** (e.g. form and punctation of elytra and pygidium). However, because *P. hearseyanus* is phenotypically distinct from the otherwise relatively homogeneous remaining taxa previously treated as *Transitupaussus* and the type species of *Transitupaussus* is *P. hardwickii* Hope, we treat *P. hearseyanus* as *incertae sedis* within PI until we can examine and analyse more taxa.

*Paussus I series: Paussus (Edaphopaussus)* **stat. rev. sensu n.**

The subgenus *Edaphopaussus* **stat. rev. sensu n.** was recovered with maximum support in the molecular and combined analyses. This lineage comprises species formerly assigned to the subgenera *Katapaussus*, *Flagellopaussus*, *Edaphopaussus* (part) and *Inermipaussus*. Anatomical character states supporting *Edaphopaussus* **stat. rev. sensu n.** include transverse pronotal cleft shallow, reduced to a narrow groove or slight constriction (69-0) (Figs 12D, E, K, 14A), prothoracic trichomes absent (70-0) (Figs 12D, E, K, 14A) and posterior dorsal margin of pygidium not upturned, rather confluent with remaining portion of tergite (82-1) (Figs 1B, 15H). This suite of features represents a significant departure from other *Paussus* with well-developed prothoracic clefts and trichomes and upturned pygidium and may signify a shift in ecological strategy in this lineage. Additional anatomical features supporting the *Edophopaussus* **stat. rev. sensu n.** clade but reversed to alternate states in subordinate taxa include (18-0), (32-1), (66-1), (71-2), (78-0) and (83-1).

### Subgenus *Edaphopaussus* Kolbe 1920

*Type species. Paussus americanus* Kolbe 1920

*Diagnosis.* Adults with transverse pronotal cleft shallow, reduced to a narrow groove or slight constriction (69-0), prothoracic trichomes absent (70-0) and posterior dorsal margin of pygidium not upturned, rather confluent with remaining portion of tergite (82-1). Anatomical features characteristic of the more inclusive PI also characterize the group (see earlier).

*Included taxa.* The subgenus *Edophopaussus* **stat. rev. sensu n.** includes species formerly assigned to the subgenera *Katapaussus*, *Flagellopaussus*, *Edaphopaussus* (part) and *Inermipaussus* (see Table S5).

*Paussus II series + Paussus III series*

The sister grouping of PII + PIII was recovered with maximum support in the molecular and combined analyses, whereas in the analysis of the morphology alone, *P. vanrooni* was unresolved with PI and the remaining taxa belonging to PII + PIII. This diverse clade of *Paussus* is supported by multiple anatomical character states, including maxillary palpomere II distinctly wider than following palpomere, ≥2× width of palpomere III (23-0) (Figs 7I, 8B, 9B), foramen of scape for reception of pedicel apically toward outer margin (53-1) (Fig. 10G–J) (most), fused flagellum excavated along outer margin (58-0) (Fig. 10C–J, L, O, Q–U), marginal trichome fringe of pygidium long, distinct and well developed (85-1) (Fig. 15A–G) and parameres without apical setae (or rarely one or two hairs) (86-1) (Fig. 16A, D). With a single exception, all species within this clade lack tibial spurs (63-1); at least some members of *Lineatopaussus* **stat. rev. sensu n.**, including *L. afzelii* sampled here, have tibial spurs present. Other characters, including (38-4)

and (48-0), lend support to this clade but are shared primarily by the earliest diverging members.

*Paussus II series*

The *Paussus* II series is a phenotypically diverse lineage that was recovered in all our analyses with moderate (77 morph) to maximum support (100 mol, comb). In terms of the species included, this clade largely overlaps with Darlington's (1950) Paussina II. Despite the breadth of phenotypic diversity exhibited in this species-rich lineage, PII is distinctive from a morphological perspective. Anatomical features that support this series include distinct temples present (1-0) (Fig. 4B, C, E–J), disc of head with two cephalic openings (9-2*) (Fig. 4E, F, H, J), gular width (at narrowest point)/length ratio between 0.3 and 0.5 (15-2), apical margin of labrum broadly emarginate (concave) medially (22-2) (Fig. 7D), lateral angles of ligula distinctly produced anteriorly (45-1) (Fig. 7N, O, Q–U), setae along lateral apical margin of ligula short, stout and spiniform (49-1) (Fig. 7Q, 8D, F), lateral margin of pronotum anterad transverse cleft angulate and prominent (72-0) (Fig. 12G, J, L) and apex of median lobe more or less flat, or with indistinct shallow emargination (88-1) (Fig. 16D–E).

Other features that characterize PII include having the eyes less prominent, occupying a much lesser portion of the head capsule (16-1) (Fig. 4C, E–I), maxillary palpomere II ≥2× width of palpomere III (23-0) (Figs 7I, 8B, 9B), foramen of scape for reception of pedicel apically not along midline, rather toward outer margin of scape (53-1) (Fig. 10G–J), fused flagellum excavated along outer margin (58-0) (Fig. 10C–J, L, O, Q–U), and tibiae without apical tibial spurs (63-1) (Fig. 11A, B, H, I).

Several major clades were recovered with high support within PII and are recognized at the subgenus level as follows: *Paussus* **stat. rev. sensu n.**, *Klugipaussus* **stat. rev. sensu n.**, *Scaphipaussus* **stat. rev. sensu n.**, *Hylotorus* **stat.n. sensu n.**, and *Anapaussus* **stat. rev. sensu n.** (see later). Statistical support for and among these lineages is high (Fig. 20). Three lineages within this clade are for now treated as *incertae sedis* (e.g. *P. ghanensis, P.* nr. *leechi, P. leroyi*) until their respective species constitution is better understood through increased taxon sampling.

*Paussus II series: Paussus (Paussus)* **stat. rev. sensu n.**

One of the most significant taxonomic outcomes of the present study is having placed *P. oertzeni* with unequivocal support with morphology alone in a combined data analysis. The placement of *P. oertzeni* as sister to *P. curtisi* + *P. africanus* was recovered in all analyses that included these taxa. The subgenus *Paussus* **stat. rev. sensu n.**, comprising members of the former subgenera *Paussus* and *Curtisipaussus* (part), is supported by several distinctive anatomical synapomorphies, including mesal margin of maxillary palpomere II sinuate, distinctly incised just beyond the midpoint (25-2) (Fig. 7H) (state shared only with

*Platyrhopalopsis picteti*), maxillary palpomere III distinctly wider than following palpomere, ≥1.5× width of IV (28-0) (Fig. 7H), labial palpomere II with socket for palpomere III distinctly shifted to the outer margin of palpomere II (42-1) (Fig. 7P, R, S), apical median margin of ligula narrowly rounded (47-1) (Fig. 7R, S), and trichome brush on anterolateral margin of scape present (54-0*) (Figs 9F, 10C, H, I). We consider 46-1 a weak synapomorphy for this group, it being shared with many other *Paussus* species. Having the terminal labial palpomeres distinctly wide, flattened and spade-shaped (38-4) (Fig. 7P, R, S) also characterizes this group. Note that when ACCTRAN optimization is used, the character state cephalic openings located on vertex (10-1*) is optimized for the subgenus *Paussus* **stat. rev. sensu n.** when in actuality this state is an uncontroverted synapomorphy for *P. curtisi* + *P. africanus*; the optimization down one node is an artefact of the inapplicable (treated as missing) data for this character for *P. oertzeni* which lacks cephalic openings (9-0).

### Subgenus *Paussus* L. 1775

*Type species. Paussus microcephalus* L. 1775

*Diagnosis.* Adults with mesal margin of maxillary palpomere II sinuate, distinctly incised just beyond midpoint (25-2), maxillary palpomere III distinctly wider than following palpomere, ≥1.5× width of IV (28-0), labial palpomeres distinctly wide, flattened and spade-shaped (38-4), labial palpomere II with socket for palpomere III distinctly shifted to the outer margin of palpomere II (42-1), apical median margin of ligula narrowly rounded (47-1), and trichome brush on anterolateral margin of scape present (54-0*). Anatomical features characteristic of the more inclusive *Paussus* II clade also characterize the group (see earlier).

*Included taxa.* This expanded concept of the subgenus *Paussus* **stat. rev. sensu n.** includes species formerly classified in the subgenus *Paussus* (e.g. *P. microcephalus* L., *P. oertzeni*, *P. affulgens* Reichensperger, *P. barkeri* Péringuey, *P. burmeisteri* Westwood, *P. dissidens* Péringuey, *P. linnaei* Westwood) and *P. curtisi*, *P. africanus*, *P. jousselini* and allies (see Table S5).

*Paussus II series: Paussus (Klugipaussus)* **stat. rev. sensu n.**

The subgenus *Klugipaussus* **stat. rev. sensu n.** includes most species formerly classified in the subgenus *Klugipaussus* and was recovered in all our analyses with weak (morph) to maximum (mol, comb) support. Anatomically, the subgenus is supported by having a tubercle within and protruding from cephalic openings (11-0*) (Figs 4F, 5A), the hind tibiae distinctly dilated and flattened, subtriangular (65-1) (Fig. 11I), and the prosternal process nearly obliterated, consisting of an extremely narrow ridge separating coxae (79-0); note that the latter of these (79-0) is highly homoplasious. The group is further characterized by having the apex of

median lobe distinctly emarginate (88-0) (Fig. 16B, C) (indicated with ACCTRAN optimization), which is relatively rare within PII, and the antennal club exhibiting a series of tubercles within the outer excavation (not considered in present study).

### Subgenus *Klugipaussus* Kolbe 1927

*Type species. Paussus klugi* Westwood 1838.

*Diagnosis.* Adults with a tubercle within and protruding from cephalic openings present (11-0*) (Figs 4F, 5A), the hind tibiae distinctly dilated and flattened, subtriangular (65-1), the prosternal process nearly obliterated, consisting of an extremely narrow ridge separating coxae (79-0), the apex of median lobe distinctly emarginate (88-0), the antennal club exhibiting a series of tubercles within the outer excavation (Fig. 10O), and elytra bearing a lateral trichome fringe (Fig. 1G). Other features characteristic of the more inclusive *Paussus* II clade characterize the group (see earlier).

*Included taxa.* The subgenus *Klugipaussus* **stat. rev. sensu n.** includes most species formerly classified as *Klugipaussus* (see Table S5).

*Paussus II series: Paussus (Scaphipaussus)* **stat. rev. sensu n.**

Analysis of both molecular data alone (Moore & Robertson, 2014) and combined data types consistently recovers a lineage within PII comprising primarily Indo-Malayan taxa with a single Palearctic species, *P. turcicus*. Despite the strong molecular support for the subgenus *Scaphipaussus* **stat. rev. sensu n.** (97 mol, 98 comb), this anatomically diverse lineage was not recovered in the analysis of morphology alone nor is it unambiguously supported by anatomical synapomorphies in the present study. However, there are a few anatomical features that generally characterize the subgenus as indicated with ACCTRAN optimization, including median frontal suture clearly not extending to anterior margin of eye (4-0) (Fig. 4B, C, G), frontal ridges well demarked but not explanate (5-1) (Figs 4A–D, 5B, D–F), mesal margin of maxillary palpomere II strongly rounded (25-0), and ligula without distinct admedian depressions and median keel (48-1) (Fig. 9A). Note that all these features occur in other *Paussus* species, and the first three character states listed are not consistent with *P. kannegieteri*, an endemic to Java.

### Subgenus *Scaphipaussus* Fowler 1912

*Type species. Paussus waterhousei* Westwood 1874

*Diagnosis.* Indo-Malayan (most) and South Palearctic species. Adults generally with median frontal suture clearly not extending to anterior margin of eye (4-0), frontal ridges well demarked but not explanate (5-1), mesal margin of maxillary

palpomere II strongly rounded (25-0), and ligula without distinct admedian depressions and median keel (48-1). Anatomical features characteristic of the more inclusive *Paussus* II clade also characterize the group (see earlier), though it should be noted that this subgenus is not well characterized by anatomical features.

*Included taxa.* With a few notable exceptions (e.g. *P. jousselini* and allies) this expanded concept of *Scaphipaussus* **stat. rev. sensu n.** includes the *Paussus* II Indo-Malayan species and the South Palearctic *P. turcicus* (see Table S5).

### *Paussus II series: P. ghanensis incertae sedis*

The exact placement of *P. ghanensis* within PII is uncertain, though it is strongly supported as being relatively close to *Scaphipaussus* **stat. rev. sensu n.** and the clade comprising *P.* nr. *leechi*, *Hylotorus* **stat.n. sensu n.**, *P. leroyi* and *Anapaussus* **stat. rev. sensu n.** In all analyses it is recovered as forming the sister group to *P.* nr. *leechi* + remaining *Paussus* II clades. However, given the short branch with negligible support subtending *P. ghanensis* and its sister group, support for the alternative hypothesis of *P. ghanensis* forming the sister group to the *Scaphipaussus* **stat. rev. sensu n.** clade is comparable.

*Paussus ghanensis* was formerly classified in the polyphyletic subgenus *Apopaussus* Kolbe (Fig. 18). At present it is not clear what other species and species groups comprise this lineage. What is evident from the current data is that *P. ghanensis* is phylogenetically and anatomically distinct from the remaining *Paussus* sampled here. Until the species membership of this lineage is better known, it is difficult to characterize this group from an anatomical standpoint and we thus refrain from formally recognizing it at the subgeneric level. However, anatomical apomorphies for *P. ghanensis* include eyes large, representing a significant portion of the head capsule anterad the postocular constriction (16-0) (Fig. 4J), anterior margin of lacinia sparsely setose, with six or fewer setae (32-0) (Fig. 8B), median anterior margin of submentum straight, continuous (36-0) (Fig. 6A), lateral lobes of mentum nearly parallel-sided (37-0) (Fig. 3), terminal labial palpomeres distinctly wide, flattened and spade-shaped (38-4) (Fig. 7P, R, S), foramen of scape for reception of pedicel apically along midline of scape (53-0) (Fig. 10D, E, M, N, S), and lateral margin of pronotum anterad of transverse pronotal cleft broadly rounded to straight (72-1) (Fig. 12B–F).

### *Paussus II series: P. nr. leechi incertae sedis*

The placement of *P.* nr. *leechi*, formerly classified as *Batillopaussus* by Luna de Carvalho (1989), is strongly supported in the present study by the molecular (100) and combined (100) data analyses as forming the sister group to a major *Paussus* series clade also recovered with maximum support (100 mol, comb) comprising *Hylotorus* **stat.n. sensu n.**, *P. leroyi* and *Anapaussus* **stat. rev. sensu n.** Until

the species constitution of this lineage is better known it is difficult to characterize this lineage from an anatomical standpoint. However, *P.* nr. *leechi* is supported by the following anatomical apomorphies: frontal ridges well demarked but not explanate (5-1) (Figs 4A–D, 5B, D–F), maxillary palpomere II with one to several stout, but short and indistinct, spiniform setae at mesal apex (26-1) (Fig. 8B), apical medial margin of ligula broadly rounded (47-0) (Fig. 7N, O), and apex of median lobe distinctly emarginate (88-0) (Fig. 16B, C). Whereas we expect all five anatomically similar species previously classified as *Batillopaussus* (Luna de Carvalho, 1989) to comprise this lineage, we refrain from formally recognizing this group at the subgeneric level until its membership is better understood through increased taxon sampling.

### *Paussus II series: Paussus (Hylotorus)* **stat.n. sensu n.**

Including anatomically bizarre taxa such as *P. cephalotes*, those formerly classified as *Hylopaussus*, *Hylotorus* and *Granulopaussus*, and many species fitting the *Cochliopaussus* mould, the subgenus *Hylotorus* **stat.n. sensu n.** comprises a broad spectrum of phenotypic and taxonomic diversity. This African lineage was recovered with maximum support in the molecular and combined data analyses (100 mol, comb) and most of the *Hylotorus* **stat.n. sensu n.** taxa formed a clade in the analysis of morphology alone with high support (98); the enigmatic *P. cephalotes* and *P. granulatus* **comb.r.** (=*Granulopaussus granulatus*) were unresolved or placed elsewhere. Anatomical features supporting the monophyly of this clade include width/length ratio of gula at narrowest point between 0.6 and 1.0 (shape of gula quadrate) (15-3) (Fig. 6D, G, H) (with one subsequent state change), anterior margin of lacinia sparsely setose, with six or fewer setae (32-0) (Fig. 8B), and prosternal process nearly obliterated, barely separating coxae at base (79-0) (occurs in many other *Paussus* series taxa).

The placement of *P. sebakuanus* **comb.n.** (=*Hylopaussus sebakuanus*) as the sister group to *P. cucullatus* + *P. semicucullatus* was strongly supported in all our analyses with maximum support, including the analysis of morphology alone. Although in general appearance these taxa are phenotypically disparate, this sister grouping is supported by several anatomical features, including anterior width/length ratio of gula ≥ 1.2 (14-3*) (Fig. 6D, G, H), apical medial margin of ligula angulate, trapezoidal (47-6) (Fig. 7Q), and pronotum anterad transverse pronotal cleft distinctly wider than at cleft and posterior part of pronotum (71-2) (Fig. 12J–L). This sister grouping underscores the striking degree of phenotypic divergence often present in such closely related species of *Paussus*.

## Subgenus *Hylotorus* Dalman 1823

*Type species. Paussus bucephalus* Gyllenhal 1817.

*Diagnosis.* Adults with width/length ratio of gula at narrowest point between 0.6 and 1.0 (shape of gula quadrate) (15-3),

anterior margin of lacinia sparsely setose, with six or fewer setae (32-0), and prosternal process nearly obliterated, barely separating coxae at base (79-0). Anatomical features characteristic of the more inclusive PII also characterize the group (see earlier).

*Included taxa.* The subgenus *Hylotorus* **stat.n. sensu n.** is taxonomically and phenotypically diverse. Whereas the species constitution of this subgenus remains uncertain, it is clear that *Hylotorus* **stat.n. sensu n.** includes at least *P. hottentottus* Westwood and allies, *P. granulatus* and allies, *P. cucullatus* and allies, and *P. cephalotes*. It is likely that other species groups currently treated as *incertae sedis* (see Table S5) belong to this subgeneric lineage.

*Paussus II series: P. leroyi incertae sedis*

Although the placement of *P. leroyi* is strongly supported in the present study by the molecular (100) and combined (100) data analyses, there are currently no anatomical apomorphies identified uniting it with the Malagasy clade. *P. leroyi* was considered by Luna de Carvalho (1989) to belong to the subgenus *Klugipaussus*, the former bearing several general phenotypic similarities to the latter. However, the results of Moore & Robertson (2014) and the present study clearly show that the similarities linking these taxa are superficial and probably ecotypic in nature. *P. leroyi* lacks the unambiguous synapomorphy of *Klugipaussus* **stat. rev. sensu n.**, namely tubercle within and protruding from cephalic openings present (11-0); in fact, *P. leroyi* lacks cephalic openings altogether. *P. leroyi* further lacks the lateral trichome fringe along the lateral margin of the elytra and series of tubercles within the outer excavation of the antennal club characteristic of the subgenus *Klugipaussus* **stat. rev. sensu n.**

Anatomical apomorphies for *P. leroyi* in the present study include: disc of head without cephalic openings (9-0), mesal margin of maxillary palpomere II strongly rounded (25-0), and mesal anterior margin of scape produced forward, forming a point (52-0) (Fig. 10J, L). In addition to *P. leroyi*, this lineage includes at least *P. viator* (not included in the present study, but anatomically similar to *P. leroyi*). Representing the sister group to the rapid Malagasy radiation of *Paussus*, fleshing out which other species, if any, are included in this enigmatic group, is of great interest.

*Paussus II series: Paussus (Anapaussus)* **stat. rev. sensu n.**

One of the most striking aspects of *Paussus* evolutionary history revealed by molecular data (Moore & Robertson, 2014) is that the phenologically diverse fauna of Madagascar are all recently descended from a single common ancestor. Here recognized as a subgenus within PII, the *Anapaussus* **stat. rev. sensu n.** was recovered with maximum support in all analyses containing molecular data (100 mol, comb). This morphologically diverse lineage is difficult to characterize, but several anatomical features support this subgenus, including maxillary palpomere II

with 1 to several stout, but short and indistinct, spiniform setae at mesal apex (26-1) (Fig. 8B), apical medial margin of ligula with slight medial invagination or notch at apex (47-4*) (Fig. 7T, U), ligula without distinct admedian depressions and median keel (48-1) (Fig. 9A), hind tibiae not distinctly dilated or flattened (65-2) (Fig. 11G). In addition, *Anapaussus* **stat. rev. sensu n.** is characterized by having the labial palpomere II sub-equal in width to palpomere I (41-1) (Fig. 7N, O, U) (shared with several other *Paussus* groups). Note that the slight median notch at the apex of the ligula was found only within Malagasy *Paussus*, but not all in the subgenus *Anapaussus* **stat. rev. sensu n.** have this trait.

**Subgenus *Anapaussus* Wasmann 1929**

*Type species. Paussus dama* H. Dohrn 1890.

*Diagnosis.* Malagasy species. Adults generally with maxillary palpomere II with one to several stout, but short and indistinct, spiniform setae at mesal apex (26-1), apical medial margin of ligula with slight medial invagination or notch at apex (47-4*), ligula without distinct admedian depressions and median keel (48-1), hind tibiae not distinctly dilated or flattened (65-2). Note that this phenotypically heterogeneous subgenus is difficult to characterize and there are many exceptions to the diagnostic characters listed earlier. Anatomical features characteristic of PII also characterize the group (see earlier).

*Included taxa. Anapaussus* **stat. rev. sensu n.** includes all *Paussus* species endemic to Madagascar.

*Paussus III series*

The *Paussus* III series, represented in the present study by *P. afzelii* and *P. vanrooni*, was strongly supported by both molecules alone (100) and combined data types (100) but was not recovered by the morphology alone. This enigmatic clade represents an anatomically intermediate lineage with respect to the *Paussus* I and II series. As a result, this series is not as clearly defined from an anatomical perspective. PIII is supported by having setae along the anterior margin of lacinia recumbent (33-1) (Fig. 7H) and lateral lobes of mentum nearly parallel-sided (37-0) (Fig. 3). The series includes most taxa formerly recognized as *Lineatopaussus* and several species of *Curtisipaussus* (see Table S5). We recognize two subgenera within this series, *Lineatopaussus* **stat. rev. sensu n.** and *Shuckardipaussus* **stat. rev. sensu n.**

*Paussus III series: Paussus (Lineatopaussus)* **stat.n. sensu n.**

The subgenus *Lineatopaussus* **stat. rev. sensu n.**, represented in the present study by *P. afzelii*, includes most taxa formerly

classified as *Lineatopaussus* (e.g. *P. lineatus*, *P. afzelii*, and allies). Until the species membership of this lineage is better known, it is difficult to characterize this group from an anatomical standpoint. However, anatomical apomorphies for *P. afzelii* include eyes with interfacetal setae present (18-0) (Fig. 5C), setation along anterior margin of lacinia sparsely setose (32-0) (Fig. 8B), mesal anterior margin/angle of scape produced forward, forming a point, extending distinctly beyond mesal margin of foramen/socket (52-0) (Fig. 10J, L), foramen/socket of scape for reception of pedicel positioned apically along midline of scape (53-0) (Fig. 10D, E, M, N, S), mid-tibiae with two apical spurs (63-0) (Fig. 11D–F), notopleural sutures nearly obliterated (74-3) (Fig. 13E), and prosternal process nearly obliterated, consisting of an extremely narrow ridge, barely separating coxae at base (79-0).

### Subgenus *Lineatopaussus* Kolbe 1928

*Type species.* *Paussus lineatus* Thunberg 1781.

*Diagnosis.* Eyes with interfacetal setae (18-0), setation along anterior margin of lacinia sparsely setose (32-0), foramen/socket of scape for reception of pedicel positioned apically along midline of scape (53-0), mid-tibiae with two apical spurs (63-0), notopleural sutures nearly obliterated (74-3), and prosternal process nearly obliterated, consisting of an extremely narrow ridge, barely separating coxae at base (79-0). Anatomical features characteristic of the more inclusive *Paussus* III clade also characterize the group (see earlier).

*Included taxa.* The subgenus *Lineatopaussus* **stat. rev. sensu n.** includes most species formerly classified as *Lineatopaussus* (see Table S5).

*Paussus III series: Paussus (Shuckardipaussus)* **stat. rev. sensu n.**

Results of Moore & Robertson (2014) and the present study clearly indicate that *Curtisipaussus sensu* Luna de Carvalho (1989) is not monophyletic. In all analyses *P. vanrooni* was never recovered in a monophylum with *P. curtisi* and *P. africanus*; rather *P. vanrooni* was strongly supported as the sister group to *P. afzelii* (see earlier). The subgenus *Shuckardipaussus* **stat. rev. sensu n.** includes *P. shuckardi* Westwood and allies (e.g. *P. vanrooni*, *P. cylindricornis* Péringuey, etc.) and while it represents an anatomically intermediate lineage with respect to most *Paussus* I and *Paussus* II taxa, it bears particular resemblance to *P. curtisi* and allies.

The uniqueness of *P. vanrooni* is supported by a suite of anatomical apomorphies, including eyes large, representing a significant portion of the head capsule (16-0) (Fig. 4A, D, J, K), maxillary palpomere II <2× width of palpomere III (23-1) (Figs 7E–H, 8A, 9A), maxillary palpomere II shorter than palpomeres III and IV combined (24-1) (Figs 7E–G, 8A, 9A), maxillary

palpomere II with one to several stout, long and prominent, spiniform setae at dorsal, mesal apex (26-0) (Figs 7G, 8A), maxillary palpomere III with one to several stout, long and prominent, spiniform setae at dorsal, mesal apex (27-0) (Figs 7G, 8A), maxillary palpomere III distinctly wider than following palpomere (28-0) (Fig. 7H), labial palpomere II with socket for palpomere III distinctly shifted to the outer margin of palpomere 2, thereby forming a distinct mesal lobe (42-1) (Fig. 7P, R, S), fused flagellum without excavation or distinct concavity (58-1) (Fig. 10A, B, K, M, N), flagellum without carina along entire inner margin (59-1), transverse pronotal cleft shallow, reduced to a narrow groove, suture or slight constriction (69-0) (Figs 12D, E, K, 14A), prothoracic trichomes rudimentary and minute (70-1) (Fig. 12C, F, H), pronotum anterad transverse pronotal cleft not distinctly widened or prominent (71-0) (Fig. 12B), posterior dorsal margin of pygidium not distinctly upturned, rather more or less confluent/even with remaining portion of tergite (82-1) (Figs 1B, 15H), and pygidium without a dense marginal trichome fringe (83-1) (Figs 15H, I, 16F).

### Subgenus *Shuckardipaussus* Kolbe 1938

*Type species.* *Paussus shuckardi* Westwood 1838.

*Diagnosis.* Eyes large, representing a significant portion of the head capsule (16-0), maxillary palpomere II <2× width of palpomere III (23-1), maxillary palpomere II shorter than palpomeres III and IV combined (24-1), maxillary palpomere II with one to several stout, long and prominent, spiniform setae at dorsal, mesal apex (26-0), maxillary palpomere III with one to several stout, long and prominent, spiniform setae at dorsal, mesal apex (27-0), maxillary palpomere III distinctly wider than following palpomere (28-0), labial palpomere II with socket for palpomere III distinctly shifted to the outer margin of palpomere II, thereby forming a distinct mesal lobe (42-1), flagellum without carina along entire inner margin (59-1), posterior dorsal margin of pygidium not distinctly upturned, rather more or less confluent/even with remaining portion of tergite (82-1), and pygidium without dense marginal trichome fringe (83-1).

*Included taxa.* The subgenus *Shuckardipaussus* **stat. rev. sensu n.** includes *P. shuckardi*, *P. vanrooni*, *P. mirei* Luna de Carvalho, *P. recticornis* Raffray, *P. cylindricornis* and *P. telescopifer* Wasmann.

*Summary*

Our current understanding of *Paussus* evolutionary history is quickly expanding with the use of molecular, biogeographic and anatomical data. At present, molecular data provide the greatest accuracy and precision for placing *Paussus* species in a phylogenetic framework. However, both biogeographic and now anatomical data are informative at varying taxonomic levels. For example, all Malagasy *Paussus* belong to a single adaptive radiation; all Indo-Malayan fauna within the *Paussus*

II series form a clade [with the exception of several species of the *P. jousselini* Guerin-Meneville species group (Sg. *Paussus* **stat. rev. sensu n.**) not included in the present study]. By contrast, because *Paussus* originated in Africa with subsequent colonization events of neighbouring continents and islands (Moore & Robertson, 2014), the African fauna cannot be placed by virtue of area of endemism as with the *Anapaussus* **stat. rev. sensu n.** and *Scaphipaussus* **stat. rev. sensu n.** subgenera. It is interesting and, from a practical, systematic standpoint, fortuitous that various African lineages of *Paussus* are relatively well characterized with anatomical data (e.g. Sg. *Paussus* **stat. rev. sensu n.**, *Bathypaussus* **stat. rev. sensu n.**, *Edaphopaussus* **stat. rev. sensu n.**, *Klugipaussus* **stat. rev. sensu n.**, *Hylotorus* **stat.n. sensu n.**), whereas both the *Anapaussus* **stat. rev. sensu n.** and *Scaphipaussus* **stat. rev. sensu n.** subgenera remain difficult to characterize based on morphology but are distinct for their respective endemism, the strongest 'synapomorphy' for these lineages.

Our phylogeny- and character-based classification proposed herein provides the necessary framework for future systematic work and a working scaffold to facilitate the ongoing classification of all *Paussus* species.

## Supporting Information

Additional Supporting Information may be found in the online version of this article under the DOI reference: 10.1111/syen.12205

**Figure S1.** Maximum likelihood trees for the individual genes reconstructed in RAXML (see Moore & Robertson, 2014). All genes are to the same scale; scale bar = 0.1 substitutions per site. *Paussus* lineages are coloured by area of endemism as in Fig. 21. Nodes supported by ML bootstrap value ≥ 90 are indicated by grey circles. Bootstrap support is given for select clades of interest when support ≤ 90.

**Figure S2.** Anatomical characters and states optimized on a reduced taxon tree entirely concordant with the combined data type BI trees (combined analysis 1, 3) including only terminals with morphological data available to facilitate character optimization (terminals with molecular data only were removed to avoid optimization artifacts due to missing data). Characters and states are optimized with ACCTRAN optimization, with those in red indicating characters/states unambiguously optimized.

**Table S1.** Terminal taxa used and material examined in the molecular and anatomical datasets with locality information. Taxonomy follows classification prior to changes introduced in the text. Morphological and molecular chimeric taxa are indicated. Sequences new to this study are deposited in GenBank under accession numbers KU921417–KU921419 (*Ozaena lemoulti*).

**Table S2.** Morphological characters and states.

**Table S3.** Morphological matrix.

**Table S4.** Summary of combined analysis 2 results (combined analyses with selective removal of molecular data for terminals with both molecular and morphological data types). Species were *a priori* rated on the expectation for accurate placement based on morphology alone in a combined data type analysis with 1 = high, 2 = moderate and 3 = low expectation of accurate placement.

**Table S5.** Revised classification of *Paussus* with the following hierarchical, informal and formal ranks implemented – genus > series > subgenus > species group > species – with most of our actions targeting the genus, series and subgenus levels. Species with uncertain phylogenetic position are listed as *incertae sedis*.

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