European Journal of Taxonomy 989: 94–118
https://doi.org/10.5852/ejt.2025.989.2887
ISSN 2118-9773
www.europeanjournaloftaxonomy.eu
2025 · Johnston N.P. *et al.*

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Research article

urn:lsid:zoobank.org:pub:C2687BEC-66D1-4516-B847-56977033AA49

A species-group framework to unravel blowfly diversity: integrative revision of the *Calliphora clarki*-group (Diptera: Calliphoridae)

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Abstract. The Australian Calliphorinae represent a diverse division of the global blowfly fauna and a broad range of life histories. Past research focused on the carrion-breeding species and there remains a paucity of research on species with other life histories. This study is the first in a series of revisionary works that combine morphology with mitochondrial DNA to revise the entire Australian Calliphorinae. A new species-group system for the subfamily is proposed, along with descriptions and revisions of species within the newly established *Calliphora clarki*-group. Three new species are described: *C. ampyx* sp. nov., *C. ignicera* sp. nov. and *C. niveata* sp. nov. Keys are provided to the Australian Calliphorinae species groups and to species within the *clarki*-group. Species concepts are supported by molecular delimitation analysis using the mitochondrial cytochrome oxidase subunit 1 gene.

Keywords. Blowfly, biodiversity, cytochrome oxidase subunit 1, sequence analysis, species delimitation.

Johnston N.P., Szpila K., Pape T., Meiklejohn K.A., Foley L.B. & Wallman J.F. 2025. A species-group framework to unravel blowfly diversity: integrative revision of the *Calliphora clarki*-group (Diptera: Calliphoridae). *European Journal of Taxonomy* 989: 94–118. https://doi.org/10.5852/ejt.2025.989.2887

Introduction

The Australian Calliphorinae Brauer & Bergenstamm, 1889 (Diptera: Calliphoridae) are best known for the carrion-breeding behaviour of some of their constituent species, particularly of the genus *Calliphora* Robineau-Desvoidy, 1830. Due to this life history, many species of *Calliphora* are of agricultural, medical and forensic importance. A large number of other Australian calliphorines from the genera *Calliphora*, *Onesia* Robineau-Desvoidy, 1830 and *Bellardia* Robineau-Desvoidy, 1830 are not carrion-breeding and instead likely to exhibit a parasitic or predatory life history, with some species reported to parasitise earthworms (Fuller 1933). Beyond the original descriptions, research on these non-carrion-breeding species has been piecemeal and consists of the treatment of only a few species (Malloch 1927; Fuller 1933; Kurahashi 1971; Norris 1973, 1994, 1996, 2000, 2002), resulting in an incomplete and inconveniently fragmented knowledge of the Australian fauna.

The Australian Calliphorinae

Currently there are six accepted calliphorine genera recorded from the Australasian region: *Aphyssura* Hardy, 1940 (Yan *et al.* 2021), *Bellardia*, *Calliphora*, *Onesia*, *Ptilonesia* Bezzi, 1927 and *Xenocalliphora* Malloch, 1924 (Kurahashi 2016). However, it is difficult to accurately attribute species to *Calliphora*, *Onesia* and *Bellardia* based on adult features due to a lack of strong, consistent and diagnostic synapomorphic character states. The most promising sources of characters are larval morphology and the shape of the male terminalia, but larval morphology is known for only a few species as larvae are difficult to obtain. The two other prominent genera in the Australasian region, *Ptilonesia* and *Xenocalliphora*, are morphologically distinct as adults (separated from the other genera by the setose subcostal sclerite) but are generally restricted to New Zealand and its surrounding islands (Dear 1985). The few records of the species of these genera in Australia are from coastal areas adjacent to cities with large shipping ports, where they have likely been found because of anthropogenic introductions (Dear 1985; Norris 1997).

Because of the lack of strong synapomorphies in adult morphology at the generic level, the Australian Calliphorinae have been subject to several different and conflicting taxonomic treatments, and the scientific justification for these genera and the species division between them has been a topic of debate over the last century. Malloch (1927) highlighted that the characters separating the genera *Calliphora* and *Onesia* in the Northern Hemisphere (namely the curvature of the "apical section of the fourth wing vein", length of the postpedicel and number of postsutural intra-alar setae) could not be used to separate the Australasian species nor do they constitute the criteria for generic monophyly. As such, he treated all Australian *Calliphora* and *Onesia* under the genus name *Calliphora.* Hardy (1930, 1932, 1947), noting the difficulty of delineating Australian *Calliphora* compared to the European and North American species, also treated all Australian calliphorine species under the genus *Calliphora* and considered *Onesia* to be a subgenus.

Kurahashi (1970, 1971), in his circumscription of the Oceanian Calliphorini, divided all calliphorine genera into three genus groups, *Melinda*-group, *Calliphora*-group and *Onesia*-group, separating these genus-groups based on their life histories and morphology of the male and female terminalia. However, Kurahashi also highlighted two core difficulties in defining these groups: (1) the lack of a reliable criterion to separate *Melinda* Robineau-Desvoidy, 1830 and *Onesia*, and (2) the combination of (or intent to combine) *Onesia* and *Calliphora* in Australia by several taxonomic authorities including

Malloch (1927) and Hardy (1930, 1947). It should be noted that Kurahashi's (1970, 1971) concept of *Melinda* is not applicable to the West Palaearctic species of *Melinda*. In the latter region, species of *Melinda* can be morphologically separated from *Onesia* by the absence of setulae on the dorsal surface of the lower calypter, the long oviscapt in females, and their oviparous reproductive strategy (Schumann 1964; Rognes 1991, 1998).

Rognes (1991) suggested that the Australian *Onesia accepta* Malloch, 1927 might in fact belong to *Bellardia* based on similarities between the male terminalia illustrated by Malloch (1927) and the general terminalia morphology of *Bellardia*. Verves (2004) agreed with the assessment of Rognes and formally moved all species of *Onesia* with "2 pairs of postsutural intra-alar setae and well developed massive apical hook of paraphallus" into *Bellardia*. He further reiterated his reasoning by defining *Onesia* as species with "3 pairs of postsutural intra-alar setae and poorly developed apical hook of paraphallus".

The Australian genus *Aphyssura* has been recently placed under Calliphorinae as a result of the phylogenomic analysis by Yan *et al*. (2021) based on 2221 single-copy nuclear genes. This phylogenetic hypothesis resolves (Aphyssurinae + Calliphorinae + Melanomyinae) + Toxotarsinae as sister to the Luciliinae, and provides evidence supporting the placement of Aphyssurinae with its single constituent genus *Aphyssura* in the Calliphorinae.

A species-group system for the Australian Calliphorinae

Several authors attempted to subdivide the Australasian *Bellardia*, *Calliphora* and *Onesia* into species-group systems based on morphology. Patton & Cushing (1934) described three groups based on the male terminalia: the *augur*-group, *canimicans*-group and *erythrocephala*-group [*C. erythrocephala* (Meigen, 1826) was later synonymised under *C. vicina* Robineau-Desvoidy, 1830]. Patton & Cushing (1934) did not provide any strong synapomorphies to define these groups but rather utilised a polythetic combination of many character states from the male terminalia.

Kurahashi (1971) expanded upon Patton's system, further dividing *Calliphora* into five sub-genera: *Neocalliphora* Brauer & Bergenstamm, 1891 (containing *erythrocephala*-group, in part, sensu Patton), *Papuocalliphora* Kurahashi, 1971, *Paracalliphora* Townsend, 1916 (containing *augur*-group sensu Patton), *Australocalliphora* Kurahashi, 1971 (containing *canimicans*-group sensu Patton) and *Calliphora* s. str. Kurahashi (1971) then suggested that *Onesia* would be sister to all of *Calliphora*. Kurahashi's subgeneric groupings also lacked strong, unifying synapomorphic character states and were instead defined by polythetic combinations of many character states from the general adult morphology and male terminalia. Further complicating the widespread adoption of Kurahashi's (1971) subgeneric system were the many exceptions inherent in his diagnoses, such as for the subgenus *Neocalliphora*: "eyes in both sexes with quite dense, erect yellow hairs or very sparsely and indistinctly haired, but quite bare in some cases of the flies having abdomen tessellated". Furthermore, Kurahashi (1971) provided no formal systematic analysis to support his system.

To simplify the taxonomy of the large and diverse Australasian Calliphorinae and to facilitate revising the Australian species, we have placed species of *Bellardia* and *Onesia* in *Calliphora* s. lat. and used the adult morphology (including terminalia), with a focus on practical and easily identifiable character states, to divide the species into groups of convenience. These groups represent our assessment, based on a subjective similarity criterion, of practical major species assemblages under a concept of strict monophyly as supported by phylogenetic relationships based on mitochondrial genomes (mtgenomes) (Johnston *et al.* in prep.). We hope that the groupings will prove useful not only by simplifying the identification and ongoing documentation of the constituent species, but also as a means of communicating about subsets of the large genus *Calliphora*.

The most practical diagnostic characters for broadly categorising the Australasian Calliphorinae into subgroups are (a) the colouration and pattern of abdominal microtomentum, (b) the setulation of eyes and subcostal sclerite, and (c) the colouration of legs, calypters and thoracic spiracles. The male terminalia also hold diagnostic power, but their features are more difficult to deal with as they mostly relate to subtle differences in shapes and sclerotisation patterns and require expertise for preparation and examination.

The present study is intended as part of a revision of the entire Australian Calliphorinae, and it is the first in a new series of revisionary works on this subfamily. This paper also outlines a new species-group system for the Australian Calliphorinae and provides a key and diagnoses to each of those species groups. Additionally, a taxonomic key is provided for all members of the proposed *Calliphora clarki*-group, along with their diagnoses and descriptions.

Material and methods

Specimen information

Museum specimens were studied from the Australian National Insect Collection (ANIC). Fresh material was collected during the Austral summers of 2021/2022 and 2022/2023 using standard sweep netting.

Label data for specimens analysed during this study are given in a standardised form providing location, collection date and collector, and using the standard abbreviations for the Australian states: ACT = Australian Capital Territory, NSW = New South Wales, TAS = Tasmania, VIC = Victoria, WA = Western Australia. Label data are grouped together by state then listed from the oldest to most recently collected.

Morphological analysis

Stacked photographs of specimens were taken with a Leica MZ16 A imaging microscope and a DFC295 Camera (Leica Microsystems, Melbourne, VIC, Australia). Photographic stacking was performed using Zerene Stacker (Zerene Systems, LLC, Richland, WA, USA) and the number of photographs optimised for each individual stacked figure (ranging from 50–200 photographs per stack). Stacked images were processed using Adobe Photoshop (Adobe, San Jose, CA, USA). Dissected male terminalia were cleared in cold potassium hydroxide (10% w/v) overnight, rinsed in demineralised water and stored in glycerol. Photography of terminalia mounted in a mix of glycerol and gel hand sanitiser then followed the same imaging protocol.

To reduce the length and repetition of the morphological descriptions, only the first species in this species-group, *C. clarki* Malloch, 1927, is described in detail. All other species are described based on differences from this species. Terminology for morphological characters follows Cumming & Wood (2017), with additional characters relevant to calyptrate male terminalia from Rognes (1991).

Molecular analysis

As this project forms part of a larger systematic revision of the entire Australian Calliphorinae, the molecular methods detailed below outline the preparation of entire mitochondrial genomes (mtgenomes). For the present paper we utilise only a part of these data – the barcoding region of cytochrome oxidase subunit 1 gene (COX1) – for the purposes of species delimitation.

DNA extraction

Prior to DNA extraction, each dry leg sample tissue (specimen list available in Supp. file 1: Table S1) was washed in a 15% bleach solution followed by two washes in distilled water. Total genomic DNA was extracted from a single leg of each specimen using a DNeasy Blood & Tissue kit (Qiagen, Germantown, MD USA) following the manufacturer's instructions for insects. Following extraction, the total isolated genomic DNA was determined using the Qubit 4 Fluorometer (Invitrogen, Waltham, MA, USA) and the Qubit dsDNA kit (Invitrogen). A subset of DNA samples representing a range of species and specimen

ages was also analysed using an Agilent TapeStation automated gel electrophoresis system (Agilent Technologies, Santa Clara, CA, USA) to determine whether DNA fragmentation was needed prior to library preparation (it was deemed unnecessary).

Mtgenome capture and next generation sequencing

The protocol for mtgenome capture, sequencing and bioinformatic processing broadly followed Scheible *et al.* (2024). In brief, DNA libraries were prepared using the KAPA Hyper Prep Kit (Roche, Basel, Switzerland) and KAPA Illumina compatible unique dual-indexed adapter kit (Roche) following the manufacturer's recommendations with one exception: library preparation was completed in half-reactions (all volumes divided by two). Following library preparation and indexation, samples were pooled equimolar, and mitochondrial DNA enrichment was completed using a Daicel Arbor Biosciences Custom MyBaits Kit (Daicel Arbor Biosciences, Ann Arbor, MI, USA) with probes designed from a mtgenome of *C. vicina* (GenBank accession NC_019639.1). Hybridisation capture was completed using a reaction temperature of 60°C following the manufacturer's instructions with one exception: 96 samples were pooled into a single hybridisation reaction. The enriched libraries were purified using 1.5X KAPA Pure beads (Roche) prior to sequencing on an Illumina MiniSeq (Illumina, San Diego, CA, USA) using a Mid Output Kit (2 ×150 paired-end sequencing).

Following sequencing, single reads were assessed for quality using FastQC (Andrews 2010), then assembled in CLC Genomics Workbench (Qiagen, Hilden, Germany) using a reference-guided approach with the mtgenome of *C. vicina* (NC_019639.1) as a reference. Assembled sequences were then annotated in Geneious Prime ver. 2023.2.1 (Dotmatics; https://www.geneious.com/) and single COX1 sequences were extracted and used for subsequent delimitation analyses in this revision. The final list of taxa and relevant COX1 accession numbers are available in Supp. file 1: Table S1.

Molecular species delimitation

To confirm that our morphological species hypotheses were supported by molecular data, we analysed the COX1 dataset using the software Assemble Species by Automatic Partitioning (ASAP) (Puillandre *et al*. 2020) methods. First, we aligned all COX1 genes using the L-INSI algorithim in MAFFT (Katoh & Standley 2013) then trimmed the alignment to the 658 bp COX1 barcoding region fragment in Geneious Prime ver. 2023.2.1. The aligned barcodes were then submitted to the ASAP web server (https://bioinfo.mnhn.fr/abi/public/asap/asapweb.html, accessed 8 Feb. 2024) with the split groups parameter set to 0.01 and using the simple distance method that was used previously to analyse barcoding sequences derived from closely related species of flies (Johnston *et al*. 2023). To support our ASAP analysis, we also completed an analysis of intra- and interspecific distances using the adegenet ver. 2.1.10 (Jombart & Bateman 2008) and BarcodingR ver. 1.0.3 (Zhang *et al.* 2017) packages in R ver. 4.3.2 (https://www.r-project.org/) inside R studio ver. 2023.12.0+369 (Posit, Boston, USA).

Results

Molecular analysis

Despite the low number of input sequences (n = 14), the ASAP delimitation analysis identified the best partition scheme for our COX1 barcode data to be five groups (ASAP-Score = 1; p = 0.22; w = 0.0017 and threshold distance = 0.010), and these aligned with the species identified by their morphology. The second-highest ranked partition identified four groups (ASAP-Score = 1; p = 0.77; w = 0.0014 and threshold distance = 0.018), combining what is treated as *C. xanthocera* Malloch, 1927 and *C. ampyx* sp. nov. below. The ASAP analysis is summarised in Supp. file 1: Table S2. Analysis of the ASAP identified putative species groups using the R package BarcodingR revealed the minimum interspecific distance of the COX1 barcode region to be 1.67% and the maximum intraspecific distance to be 0.61%, leading to a barcoding gap of ~1% between species. Overall, the mean interspecific distance was 3.51%,

while the mean intraspecific distance was 0.21%. The distance matrix for all input sequences is available in Supp. file 1: Table S3.

Taxonomy

Class Insecta Linnaeus, 1758
Order Diptera Linnaeus, 1758
Family Calliphoridae Brauer & Bergenstamm, 1889
Genus *Calliphora* Robineau-Desvoidy, 1830

Diagnosis of genus *Calliphora*

The genus *Calliphora* shares a combination of (a) abdomen ground colour metallic blue, purple or green, mottled gold or silver or entirely orange, and (b) stem vein bare dorsally with other genera of subfamily Calliphorinae, but differs by the combination of (c) lower calypter setose dorsally, and (d) subcostal sclerite not setose (pubescent).

Key to genera of Australian Calliphorinae and species-groups of *Calliphora*

1. Subcostal sclerite setulose (Supp. file 1: Fig. S1F) 2
– Subcostal sclerite pubescent or bare (Supp. file 1: Fig. S1E) 3

2. Eye densely and uniformly setulose (Supp. file 1: Fig. S1B) ..... ***Ptilonesia*** Bezzi, 1927
– Eye bare or with only sparse setulae (Figs 1B, D, 2B, D) ..... ***Xenocalliphora*** Malloch, 1924

3. Male abdominal sternite 5 with postero-median margin extended into down-turned subapical spine; lower calypter bare dorsally (see Norris 1999: fig. 1h–i) ..... ***Aphyssura*** Hardy, 1940
– Male abdominal sternite 5 with postero-median margin unmodified; lower calypter with setae dorsally ..... 4 (***Calliphora***)

4. Eye densely setulose over entire surface (Supp. file 1: Fig. S1B) ..... ***ochracea***-group
– Eye without setulae 5

5. Femora and tibiae at least partially orange or yellow (Supp. file 1: Fig. 1A); abdominal tergites 1+2–5 with microtomentum mottled golden at least in the lateral 0.3 (Supp. file 1: Fig. S1D), some species with contrasting median metallic patches dorsally on T1+2–T5 (Supp. file 1: Fig. S1C) ..... ***stygia***-group
– Femora and tibiae dark brown or black (Figs 1C, 2C); abdominal tergites 1+2–5 with microtomentum mottled golden, silver or absent (Supp. file 1: Fig. S1D) 6

6. Anterior spiracle reduced in size, smaller in width at widest part than proepisternum (Supp. file 1: Fig. S1G; Figs 3D, 5E) 7
– Anterior spiracle regular in size, equal to or larger in width at widest part than proepisternum (Supp. file 1: Fig. S1H) 8

7. Postpedicel at least partially orange (Figs 2B, 3B); upper calypter with dark orange or brown fringe (Figs 1E, 4E); male terminalia with cercus hooked with a weak concave region in basal half and weak to no sclerotisation in distal half of mesohypophallus (Fig. 4F, H) ..... ***clarki***-group
– Postpedicel orange, brown or black (Supp. file 1: Fig. S1I, M); upper calypter with yellow or hyaline fringe (Supp. file 1: Fig. S1G); male terminalia with weakly curved cercus (lateral view) and mesohypophallus with distinct break or very weak region of sclerotisation medially (Supp. file 1: Fig. S1J) ..... ***flexipenis***-group

8. Male terminalia with mesohypophallus strongly curved (c-shaped) in distal half, without break in sclerotisation (Supp. file 1: Fig. S1K) ..... ***sternalis*-group**
– Male terminalia with mesohypophallus sclerotised and straight or gently curved in distal half 9

9. Epandrium elongated, twice length of cercus from base to insertion point of cerci; acrophallus ~2 × as long as paraphallus; cercus broad in basal half, at least twice width at tip; surstylus distally with swollen lobe (Supp. file 1: Fig. S1N) ..... ***tibialis*-group**
– Epandrium not elongated, similar in length to cercus; acrophallus equal to length of paraphallus at most extending slightly beyond tips; cercus slender for entire length, similar in width to surstylus; surstylus distally without a lobe (Supp. file 1: Fig. S1L) ..... ***typica*-group**

Diagnosis of the *clarki*-group

The *clarki*-species-group can be separated from all other Australian Calliphorinae by the presence of (1) a reduced anterior spiracle, smaller at widest part than width of proepisternum (Fig. 3C, D); (2) hyaline or lightly coloured upper calypter with contrasting dark brown or orange fringe in combination with an infuscated lower calypter (Figs 1E, 4E); (3) postpedicel at least partially orange (Figs 1B, 2B, 3B); and (4) male terminalia with weak or no sclerotisation in distal part of mesohypophallus (Figs 2H, 4F, 5H). The species of this group were previously placed under the genera *Onesia* and *Calliphora.*

Key to species of the *clarki* species-group

Abbreviations for states and territories are provided within couplets for species that are restricted to specific regions of Australia.

1. Basicosta ground colour black (Figs 4E, 5E) 2
– Basicosta ground colour brown, orange or yellow (Figs 1E, 2E) 4

2. Arista plumose, 1.5 × postpedicel length (Figs 4C, 5C); NSW 3
– Arista almost bare, with only a few very short and sparse setulae near base, 2.0 postpedicel length (Fig. 3C); TAS ..... ***C. ignicera*** sp. nov.

3. Acrostichal setae 2+2 (rarely 2+3); abdominal tergites ventrally with very weak or no microtomentum; pleural suture between anepisternum and anepimeron not pronounced, similar in colour to surrounding structures (Fig. 4E); *males only*, cercus appears hook-shaped in lateral view, with only very slight concavity in its basal half (Fig. 4F); NSW ..... ***C. niveata*** sp. nov.
– Acrostichal setae 2+3 (Fig. 5A); abdominal tergites ventrally with grey microtomentum that changes colour with viewing angle; pleural suture between anepisternum and anepimeron pronounced, convex and dark red-brown, distinct from ground colour of pleura (Fig. 5E); *males only*, cercus with strong concavity in its basal half (lateral view) (Fig. 5F); NSW, VIC ***C. xanthocera*** Malloch, 1927

4. *Males only*, fronto-orbital and parafacial plate mottled gold at the height of antennal insertion (Fig. 2A–B); ACT, NSW, VIC ..... ***C. ampyx*** sp. nov.
– *Males only*, fronto-orbital and parafacial plate grey, silver, white or black at the height of antennal insertion (Fig. 1A–B) 5

5. Pleural suture between anepisternum and anepimeron pronounced and dark red-brown, distinct from ground colour of pleura (Fig. 5E); NSW, VIC ..... ***C. xanthocera*** Malloch, 1927
– Pleural suture between anepisternum and anepimeron not pronounced and similar in colour to surrounding structures (Fig. 1E); WA ..... ***C. clarki*** Malloch, 1927

Calliphora clarki Malloch, 1927
Fig. 1

Diagnosis

This species can be distinguished from other members of the *clarki*-group by the unique combination of the basicosta orange/brown and fronto-orbital and parafacial plates silver/grey at the height of the antennal insertion.

Type material

Holotype

AUSTRALIA – **WA** • Perth; 1917; J. Clark leg.; ANIC.

Other material examined

AUSTRALIA – **WA** • 3 ♂♂; Circular Pool, Nornalup National Park; 9 Oct. 1970; D.H. Colless leg.; ANIC • 2 ♂♂; Albany; 29 Oct. 1969; K.R. Norris leg.; ANIC • 1 ♂; 16 km North East of Albany; 30 Dec. 1970; P. Ferrar leg.; ANIC • 1 ♂; Yallingup; 9 Nov. 1958; E.F. Riek leg.; ANIC • 1 ♀; Shannon River; 14 Sep. 1974; N. Monzu leg.; ANIC • 1 ♂, 1 ♀; Inlet River; 14 Sep. 1974; N. Monzu leg.; ANIC • 1 ♀; Augusta; 3 Oct. 1970; D.H. Colless leg.; ANIC.

Description

Male

Head. Fronto-orbital plate with light grey microtomentum along entire length (viewed dorsally), microtomentum highly variable, appearing black laterally. Fronto-orbital plate setose with row of ~10 setae, additional weaker setulae also present. Ocellar triangle with many strong black ocellar setae, no outer vertical setae, one pair of inner vertical setae. Median occipital sclerite black with dull grey microtomentum, one pair of black post ocellar setae, one pair of paravertical setae; postocular setae black. Occiput with grey microtomentum, with intermixed black and yellow setulae. Postocular area with grey microtomentum. Parafacial plate with silvery/grey microtomentum, ground colour black, with several irregular rows of black setae. Frontal stripe dark brown, tapering, decreasing from ~5 × width of anterior ocellus to almost obliterated (lunule to height of anterior ocellus). Face with grey microtomentum. Facial ridge setose with irregular row of black setae. Scape and pedicel dark brown. Postpedicel ~2 × length of pedicel, black with basal regions of orange occupying ~0.25 of flagellomere, faint white microtomentum (most visible when viewed dorsally). Arista plumose dorsally and ventrally, black. Gena and postgena black with grey microtomentum and black setulae. Postgena concolorous with gena, with black setulae. Genal groove dark brown, without microtomentum, broader than width of parafacial plate, without setulae. Vibrissae strong, black, decussate. Frontoclypeal membrane black, pronounced anteriorly to posterior margin of postpedicel, bare. Palpus orange and straight. Proboscis dark brown, glossy.

Thorax. Ground colour metallic dark blue with weak grey microtomentum most visible presuturally. Postpronotal lobe concolourous with mesonotum but completely covered in grey microtomentum. Mesonotum with 4 weak vittae, most visible presuturally, varying in width and visibility depending of viewing angle. Scutellum concolourous with mesonotum. Proepisternum, anepisternum, katepisternum, anepimeron, meron, anatergite and katatergite all black, with weak grey microtomentum. Greater ampulla dark brown with dense short yellow setulae; lesser ampulla orange/brown with dense short yellow setulae. Haltere brown. Anterior spiracle orange/brown, reduced (smaller than proepisternum). Posterior spiracle complete, dark brown, darker than anterior spiracle.

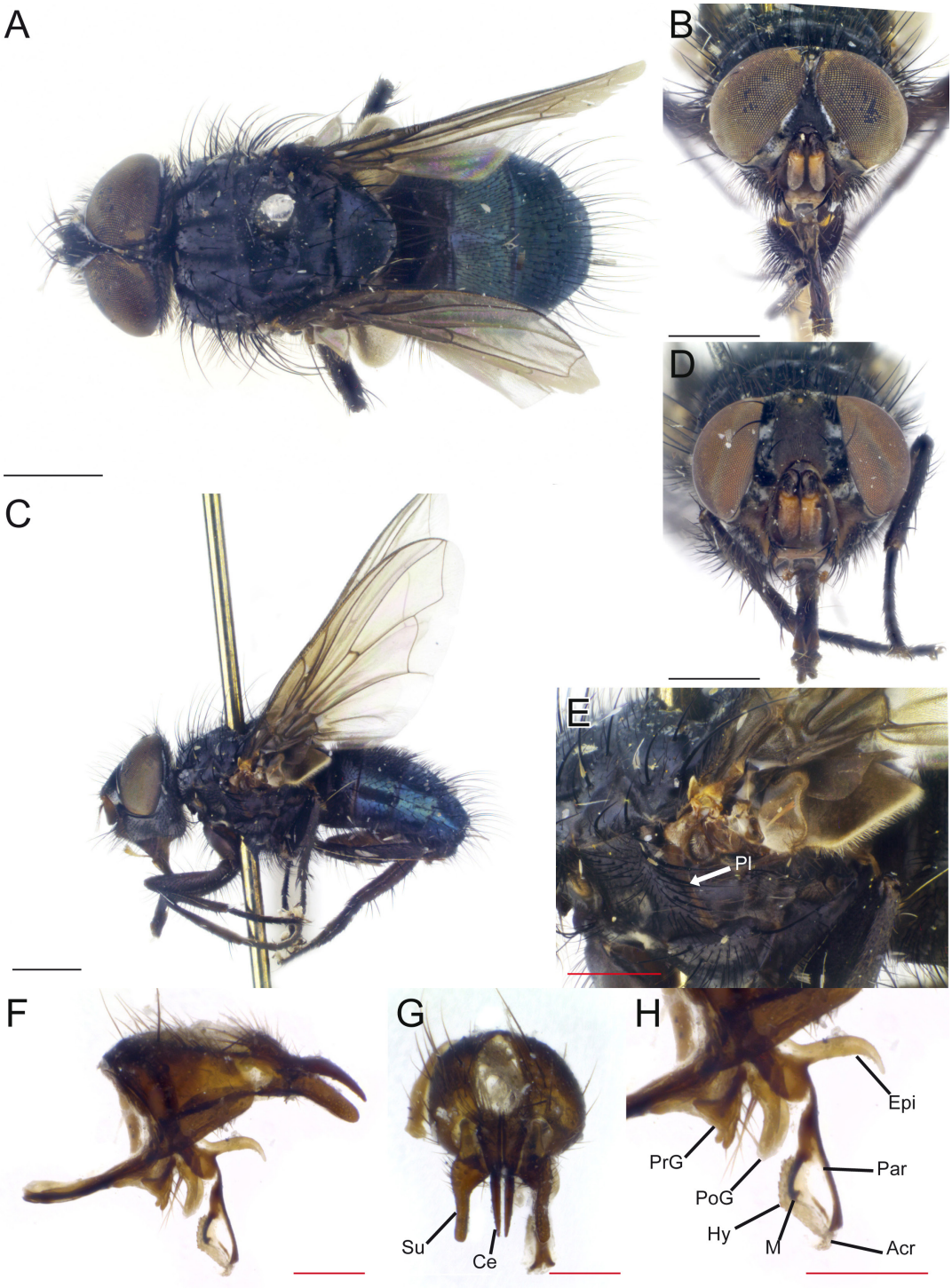

Fig. 1. A–C. *Calliphora clarki* Malloch, 1927, ♂, Yallingup, WA, 9 Nov. 1958, E.F. Riek leg. (ANIC). **A**. Habitus, dorsal view. **B**. Habitus, lateral view. **C**. Head, anterior view. **D–E**. Female, Augusta, WA, 3 Oct. 1970, D.H. Colless leg. (ANIC). **D**. Head, anterior view. **E**. Calypter, lateral view. **F–H**. Male terminalia and phallus, Circular Pool, Normalup WA, 9 Aug. 1970, D.H. Colless leg. (ANIC). **F**. Lateral view. **G**. Posterior view. **H**. Phallus, lateral view. Abbreviations: Acr = acrophallus; Ce = cercus; Epi = epiphallus; Hy = hypophallic lobe; M = mesohypophallus; Par = paraphallus; Pl = pleural suture; PoG = postgonite; PrG = pregonite; Su = surstylus. Black scale bars = 1 mm; red scale bars = 500 µm.

Chaetotaxy. Acrostichal setae: 2+3 (presutural + postsutural); dorsocentral setae: 3+3; intra-alar setae: 3+2; supra-alar setae: 2+3; 2 notopleural setae; no anterior postpronotal seta; 3 strong basal postpronotal setae; 2 postalar setae; 3 pairs of marginal scutellar setae, 1 pair of strong discal scutellar setae, additional long setulae near discal setae almost equal in length; katepisternal setae 2+1. Katepisternum with numerous short black setulae half of katepisternal setae length. All major pleural setae black. Anepisternum, anepimeron, meron, anatergite and proepisternum with black setulae. Katatergite with short dense yellow setulae.

Wing. Weakly infuscated in basal half appearing pale brown. Basicosta orange/brown. Tegula black. Subcostal sclerite brown with short dense orange setulae. Upper calypter weakly infuscated light brown with distinct dark brown margin, dorsal surface bare, brown setulae on margin particularly in fold between upper and lower calypters. Lower calypter infuscated brown with pale yellow margin, dorsal surface with black setulae, entire margin with pale yellow setulae.

Legs. Fore, mid and hind coxa, trochanters, femora, tibia and tarsi black with grey microtomentum and black setulae. Tarsal claws slightly shorter than tarsomere 5, dark brown. Pulvilli brown, each pulvillus equal in width to tarsomere 5 and equal in length to tarsal claws.

Abdomen. Abdominal tergites 1+2 (T1+2)–5 with ground colour metallic blue green with weak grey microtomentum covering all tergites (variable with viewing angle), with black setulae. T1+2 with lateral black marginal setae. T3–5 with a row of marginal black setae. Sternites 1+2–5 black with weak grey microtomentum and black setulae. S5 cleft into two lobes.

Male terminalia. *Cercus*. Equal in length to phallus; in lateral view, hooked with a weak concave region in basal half; in posterior view, fused in proximal third. *Surstylus*. Equal in length to cercus; setose; in lateral view, curved slightly downwards, in posterior view, curved towards phallus. *Phallus*. In lateral view, epiphallus equal in length to post-gonite, hooked in distal 0.1. Pre-gonite half of post-gonite length, triangular. Post-gonite curved, with a broad rounded tip. Hypophallic lobe serrated on anterior margin and sclerotised for entire length. Paraphallus weakly curved, not reaching posterior margin of hypophallic lobe, tip of paraphallus hooked. Acrophallus straight; mesohypophallus weakly or not sclerotised in distal half, sclerotised in proximal half.

Female

As for male, except in the following respects:

Head. Frontal stripe broad for entire length, one pair of outer vertical setae present.

Abdomen. S5 uncleft.

Ecology

Unknown.

Distribution

AUSTRALIA: WA.

Remarks

Label data indicate that a specimen was collected from a flowering shrub (♂; 16 km North East of Albany; 30 Dec. 1970).

Calliphora ampyx sp. nov.
urn:lsid:zoobank.org:act:063B0134-5BAA-41DD-B41D-A8F12A670870
Fig. 2

Diagnosis

This species can be distinguished from other members of the *clarki*-group by the unique combination of the basicosta ground colour not black, males with fronto-orbital and parafacial plates mottled gold at height of antennal insertion, and the cercus strongly hooked and with a pronounced concave region in the basal half.

Etymology

The species epithet '*ampyx*' is a Greek word for a 'diadem' or 'headband', often made from metal and worn around the front of the head. This name refers to the diagnostic golden microtomentum on the fronto-orbital plates of this species.

Type material

Holotype

AUSTRALIA – **NSW** • ♂; Kosciuszko National Park; 36°21′00.7″ S, 148°33′51.9″ E; 29 Jan. 2023; N.P. Johnston, J.F. Wallman, A. Grzywacz and K. Szpila leg.; ANIC.

Paratypes

AUSTRALIA – **ACT** • 2 ♀♀; Jervis Bay; 17–18 Sep. 1951; K.R. Norris leg.; ANIC • 1 ♀; Canberra; Oct. 1982; M. Whitten leg.; ANIC • 63 ♂♂, 5 ♀♀; Canberra; 4 Mar. 1984–20 Jan. 1993; K.R. Norris leg.; ANIC • 1 ♀; Black Mountain; 26 Sep. 1984; L. Liepa leg.; ANIC • 2 ♂♂, 13 ♀♀; Mt Stromlo; 20–26 Oct. 1993; D.K. Yeates leg.; ANIC. – **NSW** • 1 ♀; Barrington Tops; 6 Apr. 1949; S.J. Paramonov leg.; ANIC • 1 ♀; Depot Beach; 22 Dec. 1967; I.F.B. Common leg.; ANIC • 7 ♂♂, 16 ♀♀; Kosciuszko National Park; 36°21′00.7″ S, 148°33′51.9″ E; 29 Jan.–4 Feb. 2023; N.P. Johnston, J.F. Wallman, A. Grzywacz and K. Szpila leg.; ANIC • 10 ♂♂, 1 ♀; Ulladulla; F.H. Taylor leg.; ANIC • 1 ♂; Wentworth Falls; F.H. Taylor leg.; ANIC. – **VIC** • 1 ♀; Victorian Border, Princes Highway; 30 Dec. 1955; C. Fuller leg.; ANIC • 7 ♂♂, 1 ♀; Urquhart Bluff; 19 Nov. 1975; K.R. Norris leg.; ANIC • 1 ♂; Young's Creek; 9 Nov. 1976; D.H. Colless and L. Liepa leg.; ANIC • 2 ♂♂; Omeo Highway; 30 Nov. 1979; K.R. Norris leg.; ANIC.

Description

This species is morphologically similar to *C. clarki*, except for the following character states.

Head. Fronto-orbital plate (viewed dorsally) with gold microtomentum along entire length, microtomentum highly variable when viewed laterally. Parafacial plate black, with gold microtomentum. Frontal stripe dark brown, tapering, decreasing from ~5 × to 1 × width of anterior ocellus (lunule to height of anterior ocellus). Face with brassy microtomentum. Postpedicel twice length of pedicel, orange, some specimens with anterio-lateral black regions that occupy 0.25–0.50 of flagellomere, with very weak yellow microtomentum (most visible when viewed dorsally). Gena black with brassy microtomentum and black setulae. Postgena concolorous with gena, with black setulae.

Thorax. With weak grey microtomentum, only visible presuturally. Proepisternum, anepisternum, katepisternum, anepimeron, meron, anatergite and katatergite all black, with weak brassy or grey microtomentum. Anterior spiracle dark brown. Posterior spiracle complete, dark brown concolourous with anterior spiracle.

Chaetotaxy. One pair of discal scutellar setae; katepisternal setae 1+1.

Wing. Upper calypter white with contrasting brown margin.

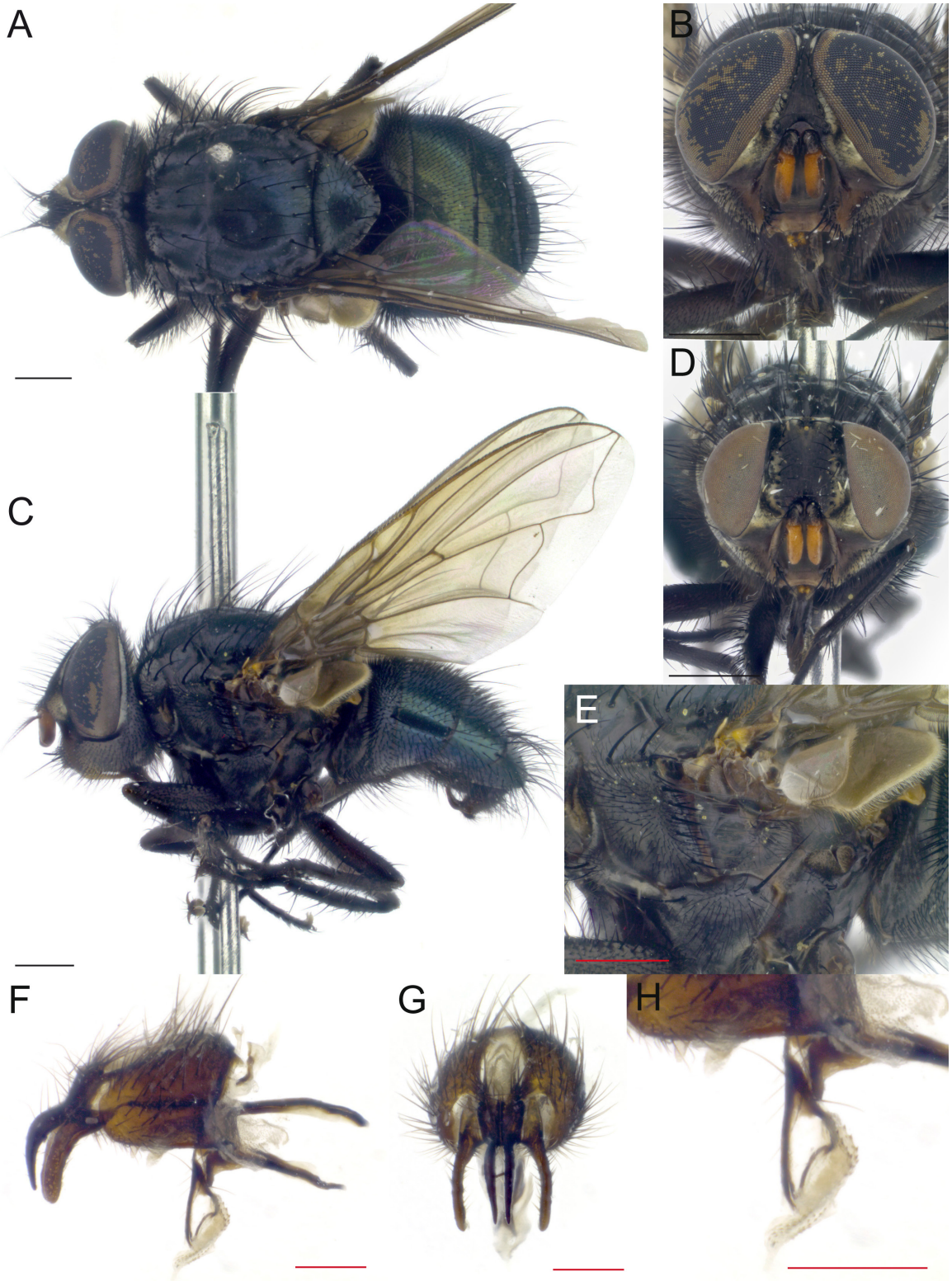

Fig. 2. A–C, F–H. *Calliphora ampyx* sp. nov., paratype, ♂, Canberra, ACT, 6 Mar. 1984, K.R. Norris leg. (ANIC). **A**. Habitus, dorsal view. **B**. Habitus, lateral view. **C**. Head, anterior view. **D–E**. Paratype, ♀, Mt Stromlo, ACT, 26 Oct. 1993, D.K. Yeates leg. (ANIC). **D**. Head, anterior view. **E**. Calypter, lateral view. **F**. Terminalia, lateral view. **G**. Terminalia, posterior view. **H**. Phallus, lateral view. Black scale bars = 1 mm; red scale bars = 500 µm.

Abdomen. T1+2–T5 ground colour metallic green, with no microtomentum.

Male terminalia. *Cercus*. Equal in length to phallus, hooked with a pronounced concave region in basal half; in posterior view, fused in proximal 0.25, separated by greater than width of one cercus for remaining 0.75. *Surstylus*. Slightly longer than cercus, setose; in lateral view, curved slightly downwards; in posterior view, straight. *Phallus*. In lateral view, epiphallus as long as post-gonite, hooked in distal 0.1. Pre-gonite length half of post-gonite; triangular. Post-gonite curved, with broad rounded tip. Hypophallic lobe serrated on anterior margin, sclerotised for entire length. Paraphallus straight, not reaching posterior margin of hypophallic lobe. Acrophallus straight, mesohypophallus weakly sclerotised in distal half, with small break, then sclerotised in proximal half.

Female

The external morphology of the female is identical to that of the male, except in the following respects:

Head. Fronto-orbital and parafacial plate with gold microtomentum, one pair of outer vertical setae.

Abdomen. S5 uncleft.

Ecology

Unknown.

Distribution

AUSTRALIA: ACT, NSW and VIC.

Remarks

Label data indicate that this species was seen emerging from a garden lawn in large numbers (♀; Canberra; Oct. 1982; M. Whitten leg.; ANIC). It was also observed on dog faeces (♀, 8 Apr. 1984; K.R. Norris) and caught at light traps (♀; Black Mountain; 26 Sep. 1984; L. Liepa).

Calliphora ignicera sp. nov.

urn:lsid:zoobank.org:act:0FE45501-BA3B-4933-9CE1-443CB8A44A00

Fig. 3

Diagnosis

Females of this species can be distinguished from all other female members of the *clarki*-group by the unique combination of the basicosta black, arista with only short, sparse setulae near its base and twice length of the postpedicel.

Etymology

The species epithet '*ignicera*' comes from the combination of the Latin '*ignis*' meaning 'fire' and the suffix '*-cera*' meaning 'horned'. The name refers to the bright orange antennae of this species.

Type material

Holotype

AUSTRALIA – **TAS** • ♀; Flinders Is.; 3 Nov.1989; C. Tann leg.; ANIC.

Paratypes

AUSTRALIA – **TAS** • 1 ♀; Cradle Valley; 16 Jan. 1923; A. Tonnoir leg.; ANIC • 10 ♀♀; Meredith River; 5–6 Jan. 1954; T.G. Campbell leg.; ANIC • 6 ♀♀; Pieman River; 5 Jan. 1954; T.G. Campbell leg.; ANIC • 11 ♀♀; Flinders Is.; 3 Nov. 1989–20 May 1991; C. Tann leg.; ANIC.

Description

This species is morphologically similar to *C. clarki*, except for the following character states.

Female

Head. Fronto-orbital plate with brassy microtomentum. One pair of outer vertical setae. Parafacial plate with brassy microtomentum. Frontal stripe ~8× width of ocellus for entire length. Arista almost bare, only a few very short and sparse setulae near base, twice length of postpedicel.

Thorax. Ground colour metallic dark green with grey microtomentum that is most visible presuturally.

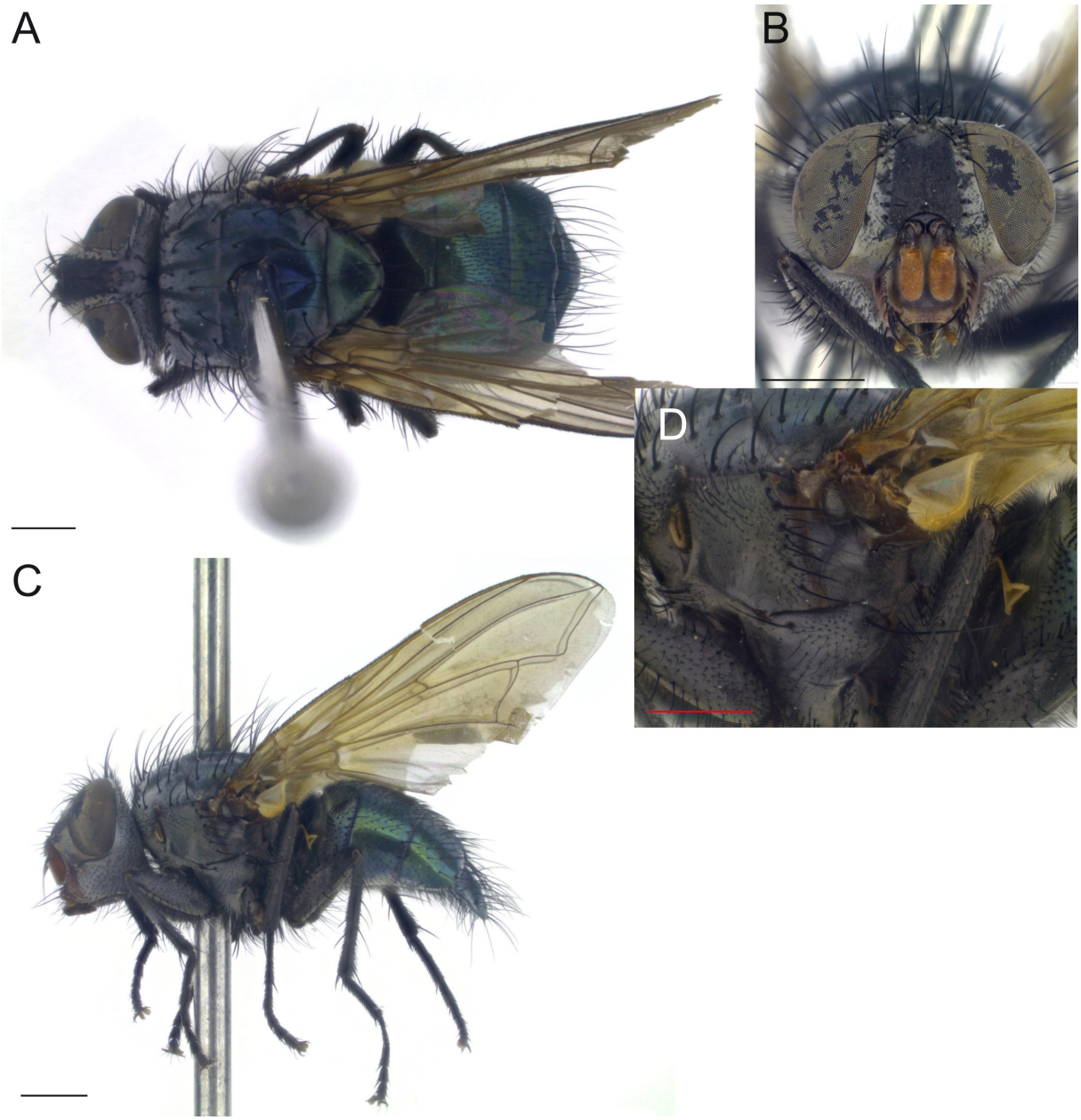

Fig. 3. *Calliphora ignicera* sp. nov., paratype, ♀, Flinders Is., TAS, 20 May 1991, C. Tann leg. (ANIC). **A**. Habitus, dorsal view. **B**. Head, anterior view. **C**. Habitus, lateral view. **D**. Calypter, lateral view. Black scale bars = 1 mm; red scale bar = 500 µm.

Chaetotaxy. 1 pair of discal scutellar setae.

Wing. Basicosta black. Upper calypter white with contrasting brown margin, yellow setulae on margin particularly in fold between upper and lower calypters. Lower calypter weakly infuscated brown with pale yellow margin, dorsal surface with yellow setulae, entire margin with pale yellow setulae.

Abdomen. S5 uncleft.

Ecology

Unknown.

Distribution

AUSTRALIA: TAS.

Remarks

No male specimens were available.

Calliphora niveata sp. nov.
urn:lsid:zoobank.org:act:35535CA1-25C3-4F40-952F-C590CA45C308
Fig. 4

Diagnosis

This species can be distinguished from all other members of the *clarki*-group by the unique combination of the basicosta black, arista plumose, abdominal tergites with weak or no microtomentum, and males with the cercus almost straight in the lateral view, without a concave region in the basal half.

Etymology

The species epithet *niveata* stems from the Latin '*nivis*' meaning 'of snow' and alludes to the habitats of the Australian Snowy Mountains from which this species has been collected.

Type material

Holotype

AUSTRALIA – **NSW** • ♂; Guthega, Kosciuszko National Park; 1 Feb. 1974; K.R. Norris leg.; ANIC.

Paratypes

AUSTRALIA – **NSW** • 3 ♂♂; same data as for holotype; ANIC • 8 ♂♂, 5 ♀♀; Rainbow Lake Walking Track, Kosciuszko National Park; 36°22′08″ S, 148°28′30″ E; 30 Jan. 2023; N.P. Johnston, J.F. Wallman, A. Grzywacz and K. Szpila leg.; ANIC.

Description

This species is morphologically similar to *C. clarki*, except for the following character states.

Male

Head. Occiput with grey microtomentum, with black setulae only. Parafacial plate black, with brassy/grey microtomentum, with several irregular rows of black setae. Face with brassy/grey microtomentum. Facial ridge setose, with irregular row of black setae for ~0.70× length. Postpedicel twice length of pedicel, dark brown with small orange region in basal 0.20, with white microtomentum (most visible when viewed dorsally).

Thorax. Greater ampulla dark brown, with dense short yellow setulae; lesser ampulla brown, with dense short yellow setulae. Haltere brown. Anterior spiracle dark brown. Posterior spiracle complete, dark brown, concolourous with anterior spiracle.

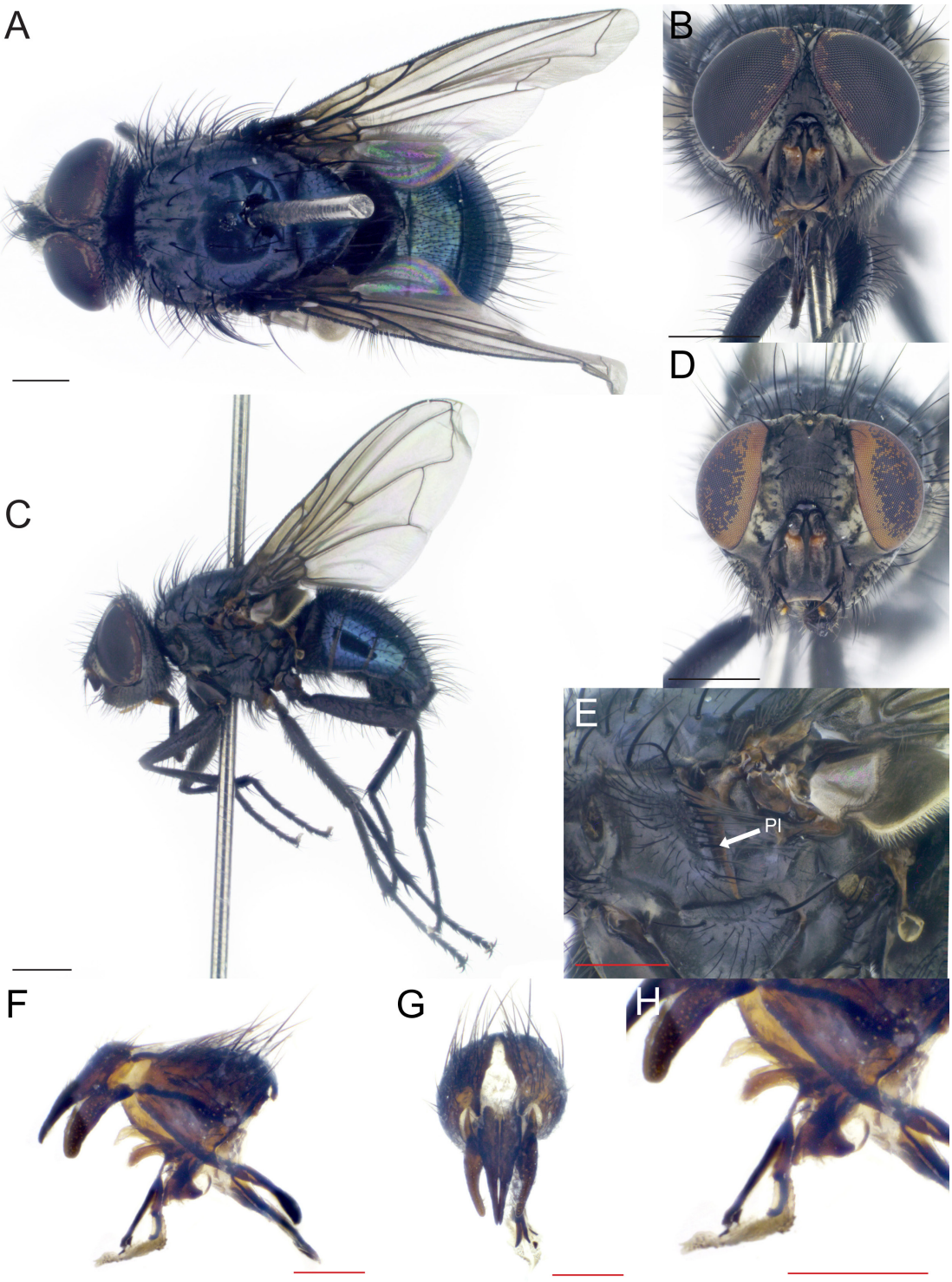

Fig. 4. A–C. *Calliphora niveata* sp. nov., paratype, ♂, Rainbow Lake Walking track Kosciuszko, NSW, 30 Jan. 2023, N.P. Johnston, J.F. Wallman, A. Grzywacz and K. Szpila leg. (ANIC). **A**. Habitus, dorsal view. **B**. Habitus, lateral view. **C**. Head, anterior view. **D–E**. Paratype, ♀, Rainbow Lake Walking track Kosciuszko, NSW, 30 Jan. 2023, N.P. Johnston, J.F. Wallman, A. Grzywacz and K. Szpila leg. (ANIC). **D**. Head, anterior view. **E**. Calypter, lateral view. **F–H**. Paratype, ♂, Guthega, NSW, 4 Apr. 1984, K.R. Norris leg. (ANIC). **F**. Terminalia, lateral view. **G**. Terminalia, posterior view. **H**. Phallus, lateral view. Abbreviation: Pl = plural suture. Black scale bars = 1 mm; red scale bars = 500 µm.

Chaetotaxy. Acrostichal setae: 2+2 (presutural + postsutural; rarely 2+3); 2 notopleural setae; no anterior postpronotal seta; 3 strong basal postpronotal setae; 1 pair of discal scutellar setae.

Wing. Basicosta black. Upper calypter white, with contrasting brown margin.

Abdomen. T1+2–T5 metallic green, without obvious microtomentum.

Male terminalia. *Cercus*. Equal in length to phallus; in lateral view, appearing slightly curved downward; in posterior view, fused in proximal half, angled towards each other, touching in distal 0.25. *Surstylus*. Equal in length to cercus, setose; in lateral view, curved slightly downwards; in posterior view, curved towards phallus. *Phallus*. In lateral view, epiphallus equal in length to post-gonite, hooked in distal 0.1. Pre-gonite half of length of post-gonite, triangular. Post-gonite curved, with broad rounded tip. Hypophallic lobe serrated on anterior margin, sclerotised for entire length. Paraphallus straight, not reaching posterior margin of hypophallic lobe. Acrophallus straight, mesohypophallus weakly or not sclerotised in distal half, sclerotised in proximal half.

Female

As for male, except in the following respects:

Head. Frontal stripe broad for entire length, one pair of outer vertical setae.

Abdomen. S5 uncleft.

Ecology

Unknown.

Distribution

AUSTRALIA: NSW.

Remarks

Label data indicate that this species has been collected above 1600 m altitude.

Calliphora xanthocera Malloch, 1927
Fig. 5A–H

Diagnosis

This species can be distinguished from other members of the *clarki*-group by the unique combination of the basicosta dark brown or black; arista plumose, 1.5 × length of postpedicel; acrostichal setae 2+3; abdominal tergites with very weak or no microtomentum; pleural suture between anepisternum and anepimeron pronounced, dark red-brown, distinct from the ground colour of the pleura.

Type material

Holotype

AUSTRALIA – **NSW** • ♂; Kosciuszko National Park; 5 Dec. 1921; ANIC.

Other material examined

AUSTRALIA – **NSW** • 1 ♀; Wee Jasper; 29 Jan. 1933; C. Fuller leg.; ANIC • 1 ♀; Mt Kosciuszko; 29 Jan. 1933; I. Mackerras leg.; ANIC • 1 ♀; Tin Mine Creek; 30 Nov. 1959; D.F. Waterhouse leg.; ANIC • 1 ♂, 3 ♀♀; Kosciuszko; 27 Jan.–25 Mar. 1965; D.E. Havenstein leg.; ANIC • 2 ♂♂, 6 ♀♀; Tinmine Hut Track, Snowy Mountains; 20 Mar. 1971; K.R. Norris leg.; ANIC • 1 ♀; Tinmines, Snowy

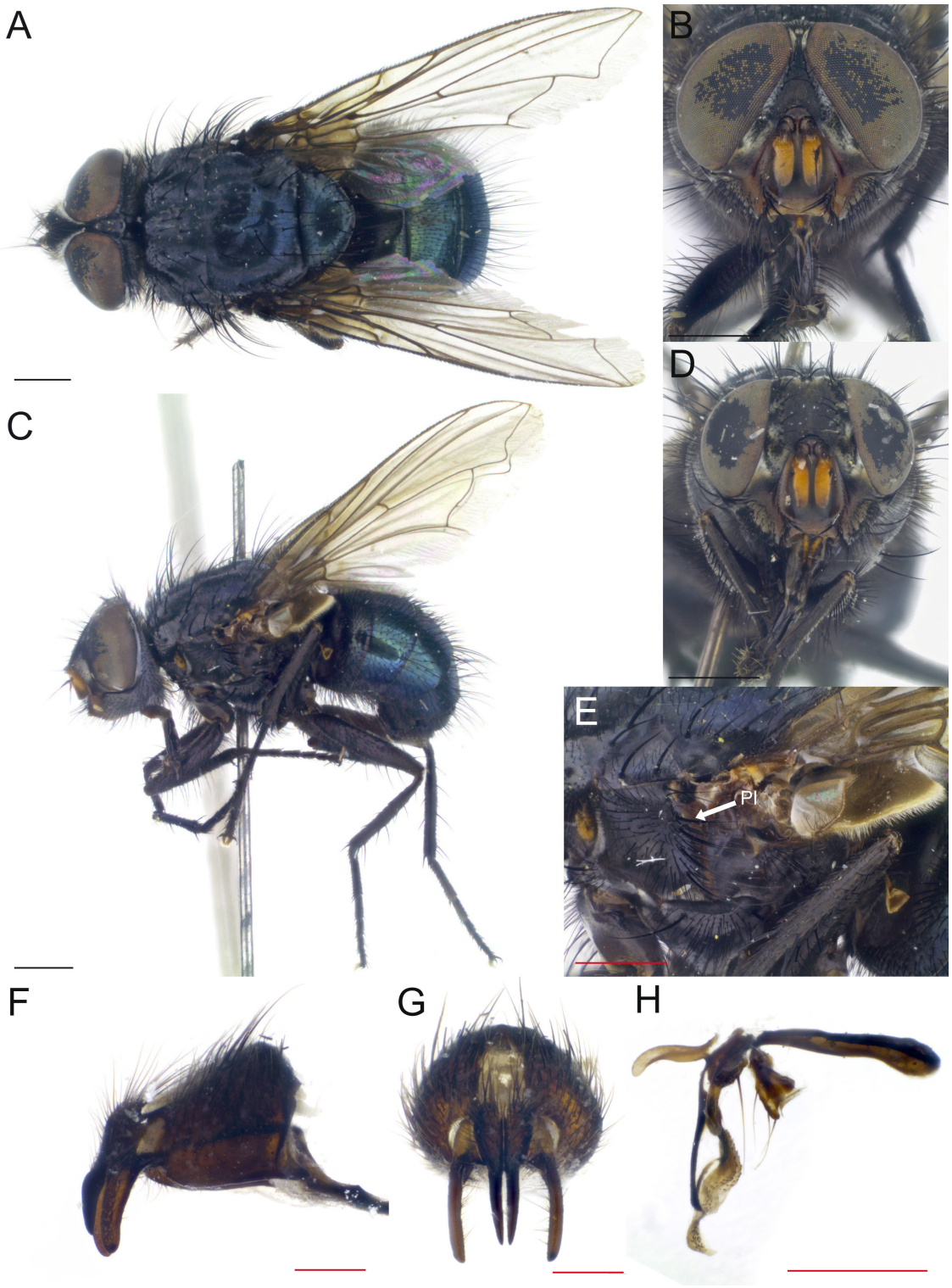

Fig. 5. A–C. *Calliphora xanthocera* Malloch, 1927, ♂, Tinmine Hut Track, Snowy Mountains, NSW, 20 Mar. 1971, K.R. Norris leg. (ANIC). **A**. Habitus, dorsal view. **B**. Habitus, lateral view. **C**. Head, anterior view. **D–E**. Female, Tinmine Hut Track, Snowy Mountains, NSW, 20 Mar. 1971, K.R. Norris leg. (ANIC). **D**. Head, anterior view. **E**. Calypter, lateral view. **F–H**. Male, Kosciuszko, NSW, 25 Mar. 1965, D.E. Havenstein leg. (ANIC). **F**. Terminalia, lateral view. **G**. Terminalia, posterior view. **H**. Phallus, lateral view. Abbreviation: Pl = pleural suture. Black scale bars = 1 mm; red scale bars = 500 µm.

Mountains; 20 Mar. 1971; K.R. Norris leg.; ANIC • 1 ♀; Mt Tingaringy; 25 Feb. 1972; K.R. Norris leg.; ANIC • 1 ♀; Charlotte Pass; 22 Jan. 1986; K.R. Norris leg.; ANIC • 2 ♂♂; Rainbow Lake Walking Track, Kosciuszko; 36°22′08″ S, 148°28′30″ E; 30 Jan. 2023; N.P. Johnston, J.F. Wallman, A. Grzywacz and K. Szpila leg.; ANIC. – **VIC** • 1 ♂; Quambat Flat; 20 Mar. 1971; K.R. Norris leg.; ANIC.

Description

This species is morphologically similar to *C. clarki*, except for the following character states.

Male

Head. Scape and pedicel dark brown. Postpedicel twice length of pedicel, orange with slight darkening in anterio-lateral 0.25, with white microtomentum (most visible when viewed dorsally).

Thorax. Anterior spiracle orange, reduced, smaller than proepisternum. Posterior spiracle complete, dark brown, contrasting in colour to anterior spiracle. Pleural suture between anepisternum and anepimeron pronounced, dark red-brown, distinct from ground colour of pleura.

Chaetotaxy. 4 strong basal postpronotal setae; 1 pair of discal scutellar setae.

Wing. Basicosta black or dark brown. Upper calypter white, with contrasting brown margin.

Abdomen. T1+2–T5 metallic green, with no microtomentum.

Male terminalia. *Cercus*. Equal in length to phallus; in lateral view, hooked with weak concave region in basal half; in posterior view, fused in proximal 0.30. *Surstylus*. Equal in length to cercus, setose; in lateral view, curved slightly downwards; in posterior view, curved slightly towards phallus. *Phallus*. In lateral view, epiphallus equal in length to post-gonite, hooked in distal 0.10. Pre-gonite half of length of post-gonite, triangular. Post-gonite curved, with broad rounded tip. Hypophallic lobe serrated on anterior margin, sclerotised for entire length. Paraphallus weakly curved, not reaching posterior margin of hypophallic lobe, tip of paraphallus hooked. Acrophallus straight; mesohypophallus weakly sclerotised in distal half, with small break, then sclerotised in proximal half.

Female

External morphology of the female is identical to the male, except in the following respects:

Head (Fig. 5D). Fronto-orbital and parafacial plate with gold microtomentum, one pair of outer vertical setae.

Abdomen. S5 uncleft.

Ecology

Unknown.

Distribution

AUSTRALIA: NSW and VIC.

Discussion

Diagnostic morphology, sexual dimorphism

This paper describes a newly established species-group within the Australian *Calliphora*, containing five species, two (*C. clarki* and *C. xanthocera*) previously described by Malloch (1927) and three (*C. ampyx* sp. nov., *C. ignicera* sp. nov. and *C. niveata* sp. nov.) new to science. The most important morphological

characters uniting species of the *clarki*-group are the colouration of the antennae, size of the anterior spiracle, colouration of the upper and lower calypters and unique shape of the male terminalia (in particular the cercus and mesohypophallus). Indeed, while these species are grouped together here for the first time, their similarities, particularly in the male terminalia, can in fact be observed in Malloch's original illustrations (1927: figs 8, 12). Morphological variation between species is more subtle and tends to be related to the colouration on the head and thorax and the general chaetotaxy of the thorax. Interestingly, all species of the *clarki*-group exhibit sexual dimorphism (beyond the typical separation of the eyes observed in many female calliphorids). Females of the *clarki*-group all exhibit strong golden microtomentum on the fronto-orbital plates compared to the males, which are typically duller and brassier. Sexual dimorphism in colour is well documented across the calyptrate flies (Butterworth *et al.* 2020; White *et al.* 2020; Johnston *et al.* 2021) and may be related to sexual selection or mate attraction (White *et al.* 2020).

Molecular delineation

While DNA barcoding has been observed to be ineffective in separating some groups of calliphorid species (Wallman & Donnellan 2001; Wells *et al.* 2004; Whitworth *et al.* 2007; Sonet *et al*. 2012; Williams *et al.* 2016), all species in the *clarki*-group were delineated using COX1 alone with a barcode gap of ~1%. However, it should be noted that the minimum interspecific distance between species was ~1–3% lower than what has been observed in other subfamilies of Calliphoridae (Nelson *et al.* 2007) and other non-carrion-breeding calyptrate flies in Australia (Johnston *et al.* 2020a, 2020b) and abroad (Szpila *et al.* 2023). The intraspecific variation is consistent with that observed by other authors (e.g., Johnston *et al.* 2020a, 2020b; Szpila *et al*. 2023).

Biology and biogeography

Very little is known about the biology of species in the *clarki*-group. However, based on their habitat and the lack of attraction to rotten meat-baits (N.J. pers. obs.), it can be hypothesised that these species are predatory or parasitic and may feed on earthworms, as observed in other Australian species of *Calliphora* such as *C. accepta* Malloch, 1927 (Fuller 1933). In addition, label data indicate that adults of the *clarki*-group are attracted to flowering plants, most likely for carbohydrates. Species of the *clarki*-group appear to share their pattern of distribution with the broader Australian *Calliphora*, showing a preference for temperate mesic environments (Norris 1965). Interestingly, there also appears to be an east-west separation of species, with *C. clarki* occurring only in Western Australia and *C. ampyx* sp. nov., *C. niveata* sp. nov. and *C. xanthocera* restricted to south-eastern Australia (NSW and VIC), whereas *C. ignicera* occurs only in Tasmania. This pattern is observed in other Australian *Calliphora*, with the well-known carrion-breeding species: *C. albifrontalis* Malloch, 1932 and *C. varifrons* Malloch, 1932 restricted to Western Australia, and *C. stygia* Fabricius, 1782 and *C. hilli* Patton, 1925 occurring in eastern Australia (Wallman 2001).

Future prospects

This paper marks the first of a series of targeted revisions of the Australian Calliphorinae. In this paper, we establish a new species-group system for Australian *Calliphora* and circumscribe the *clarki*–group. This species-group and its constituent species have remained unstudied for almost a century due to the cryptic biology of the non-carrion-breeding species, subtle morphological characters and relative difficulty in sampling these species in large numbers. By integrating morphology and molecular data, we have been able to strengthen the evidence for species delineation in this group. Based on the material examined during this revisionary work, the *clarki*-group represents the smallest species-group of *Calliphora* in Australia, while still containing three new species. It is expected that massive calliphorine diversity is yet to be discovered in Australia, but the integrative framework established in this paper should greatly accelerate its delineation and description.

Acknowledgements and funding

This work would have been impossible without the tremendous efforts of the late entomologists K.R. Norris and S.J. Paramonov, who both provided extensive notation on the specimens that they studied in the Australian National Insect Collection (ANIC). We also thank David Yeates (ANIC) for his generous arrangement of access to and loan from the Collection of thousands of blowfly specimens for this project, and Melissa Scheible for her assistance with generating mtgenomes. This project was supported through government funding from the Australian Biological Resources Study (grant ID: 4-G046X5I). KS was supported by Polish National Science Centre project 2018/31/B/NZ8/02113.

Data availability statement

All molecular sequences used in this manuscript are deposited in the GenBank genetic sequence database (available at: https://www.ncbi.nlm.nih.gov/genbank/). Accession numbers for specific sequences are available in Supp. file 1: Table S1.

Conflict of interest

The authors declare no conflict of interest.

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Manuscript received: 27 June 2024
Manuscript accepted: 26 November 2024
Published on: 29 April 2025
Topic editor: Tony Robillard
Section editor: Torbjørn Ekrem
Desk editor: Pepe Fernández

Printed versions of all papers are deposited in the libraries of four of the institutes that are members of the *EJT* consortium: Muséum national d'Histoire naturelle, Paris, France; Meise Botanic Garden, Belgium; Royal Museum for Central Africa, Tervuren, Belgium; Royal Belgian Institute of Natural Sciences, Brussels, Belgium. The other members of the consortium are: Natural History Museum of Denmark, Copenhagen, Denmark; Naturalis Biodiversity Center, Leiden, the Netherlands; Museo Nacional de Ciencias Naturales-CSIC, Madrid, Spain; Leibniz Institute for the Analysis of Biodiversity Change, Bonn – Hamburg, Germany; National Museum of the Czech Republic, Prague, Czech Republic; The Steinhardt Museum of Natural History, Tel Aviv, Israël.

Supp. file 1. https://doi.org/10.5852/ejt.2025.989.2887.13091

Table S1. Specimen label and accession data for molecular analyses.

Table S2. Assemble Species by Automatic Partitioning (ASAP) analysis based on COX1 data (658 bp fragment).

Table S3. Cytochrome oxidase subunit, genetic pairwise distance matrix.

Fig. S1. Calliphorinae Brauer & Bergenstamm, 1889 diagnostic morphology. **A**. *Calliphora hilli* Patton, 1925, forelegs. **B**. *C. quadrimaculata* (Swederus, 1787), head lateral view, highlighting setulation setulation. **C**. *C. dubia* (Macquart, 1855), abdomen. **D**. *C. stygia* (Fabricius, 1781), abdomen. **E**. *C. minor* Malloch, 1927, lateral view of thorax highlighting subcostal sclerite. **F**. *Xenocalliphora* sp., lateral view of thorax highlighting subcostal sclerite. **G**. *C. minor*, lateral view of thorax highlighting anterior spiracle and upper calypter. **H**. *C. minor*, lateral view of thorax highlighting anterior spiracle and upper calypter. **I**. *C. minor*, head anterior view highlighting postpedicel. **J**. *Calliphora flexipenis*-group male terminalia highlighting mesohypophallus. **K**. *Calliphora sternalis*-group male terminalia highlighting mesohypophallus. **L**. *Calliphora typica*-group male terminalia highlighting mesohypophallus. **M**. *C. apicalis* Malloch, 1927, head anterior view highlighting postpedicel. **N**. *Calliphora tibialis*-

group male terminalia highlighting mesohypophallus and sclerotised mesohypophallus. **O**. *Calliphora ampyx* sp. nov., male terminalia posterior view, highlighting diagnostic characters. Abbreviations: Hy = hypophallic lobe; M = mesohypophallus; Par = paraphallus. Black and white scale bars: = 1 mm; red scale bars = 500 μm.