Morphology and Ontology

The Skeletomuscular System of the Mesosoma of *Formica rufa* Workers (Hymenoptera: Formicidae)

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Subject Editor: Elizabeth Jockusch

Received 2 September 2021; Editorial decision 14 December 2021

Abstract

The mesosoma is the power core of the ant, containing critical structural and muscular elements for the movement of the head, legs, and metasoma. It has been hypothesized that adaptation to ground locomotion and the loss of flight led to the substantial rearrangements in the mesosoma in worker ants and that it is likely the ant mesosoma has undergone functional modifications as ants diversified into different ecological and behavioral niches. Despite this importance, studies on the anatomy of the ant mesosoma are still scarce, and there is limited understanding of important variation of internal structures across the ant phylogeny. Recent advances in imaging techniques have made it possible to digitally dissect small insects, to document the anatomy efficiently and in detail, and to visualize these data in 3D. Here we document the mesosomal skeletomuscular system of workers of the red wood ant, *Formica rufa* Linnaeus, 1761, and use it to establish a 3D atlas of mesosomal anatomy that will serve as reference work for further studies. We discuss and illustrate the configuration of the skeletomuscular components and the function of the muscles in interaction with the skeletal elements. This anatomical evaluation of a ‘generalized’ ant provides a template for future studies of the mesosoma across the radiation of Formicidae, with the ultimate objective of synthesizing structural, functional, and transformational information to understand the evolution of a crucial body region of ants.

Key words: ant, mesosoma, thorax, internal morphology, anatomy

Ants are among the most abundant and ecologically important insect groups on Earth. The ubiquity, diversity, and cooperation of ants have inspired intense scientific interest in their sociality (e.g., Wilson 1971, Nowak et al. 2010, Boomsma and Gawne 2018), as well as ecology and evolution (e.g., Hölldobler and Wilson 1990, Lach et al. 2009, Ward 2014, Branstetter et al. 2017a). Although the foundation of ant classification and systematics is comparative morphology (e.g., Forel 1878, Brown 1954, Baroni Urbani et al. 1992, Bolton 2003), there remains a considerable amount to learn about anatomy and functional morphology. This is especially true because advances in imaging technologies (e.g., Beutel et al. 2014, Friedrich et al. 2014) and molecular phylogenetics (e.g., Branstetter et al. 2017b, Borowiec et al. 2020) enable explicit and detailed hypothesis testing of ancestral states and macroevolutionary patterns. The powerful tools of micro-computed tomography (μ-CT) and 3D modeling in particular have provided new vigor to the study of ant anatomy and functional morphology. Using μ-CT data, special focus has thus far been paid to the head (e.g., Larabee et al. 2017, Gibson et al. 2018, Khalife et al. 2018, Klunk et al. 2021), including a series of detailed anatomical works that have established a baseline for the documentation of variation across the ant phylogeny (Richter et al. 2019, 2020, 2021). In comparison, the mesosoma is understudied (Liu et al. 2019, Peeters et al. 2020), while metasoma anatomy has not been analyzed in detail using μ-CT technology.

The mesosoma is the central power core of the ant body (Peeters et al. 2020), governing flight, locomotion, and the movement of the head and the petiole. The origin of winged-wingless polyphenism in females was arguably the seminal evolutionary event in the history of ants, allowing for the optimization of the legs and wingless mesosoma for ground labor (Boudinot 2015, Boudinot et al. 2020b, 2020c).
Peeters et al. 2020), and potentially for the origin of specialized worker castes (Rajakumar et al. 2018) and of eusociality itself (Hanna and Abouheif 2021). Furthermore, given the broad ecological and behavioral diversity across the ants, as well as considerable structural variation (e.g., Bolton 1994, 2003), we expect the structures of the mesosoma and its limbs to have important functional traits that co-occur with ant performance in different contexts (e.g., Sosiak and Barden 2021). However, before such a phylogenetically broad analysis, we need a comprehensive anatomical foundation to map out relevant structures and resolve inconsistencies in nomenclature and homology.

Our knowledge of the internal mesosomal anatomy of ants across their phylogeny remains limited. Detailed studies treating the internal structures of the mesosoma are available for only four ant species, with early studies of exceptional quality but limited utility (e.g., Lubbock 1881, Nasonov 1889, Janet 1897, 1898, Emery 1900). Each of these prior works differs in the applied techniques and terminology, the latter being an issue that remains unresolved (e.g., Markl 1966, Liu et al. 2019). More generally, the terminological conflict between the systems of myrmecologists and other entomologists is widespread because of the systematic emphasis on the highly modified worker caste. Such conflict can obscure the literature, cause misinterpretation, and imply falsified or otherwise incorrect homology. Efforts to establish a universally understood anatomical nomenclature bridging ants, other Hymenoptera, and insects more broadly are incomplete, but address wing venation (Brown and Nutting 1950, Yoshimura and Fisher 2009), the alate mesosoma and proximal leg articulations (Boudinot 2015), the male genitalia and surrounding structures (Yoshimura and Fisher 2007, 2011; Boudinot 2013, 2018), the head (Richter et al. 2019, 2020, 2021), and traditionally recognized structures more broadly (Keller 2011). As we progress in the documentation of anatomy, so will our language change to better reflect the observed and inferred evolutionary patterns.

Prior studies on the mesosomal anatomy of ants have focused on Formica Linnaeus, 1758 and Myrmica Latreille, 1804 queens (Janet 1898), Lasius Fabricius, 1804 (Lubbock 1881, Nasonov 1889), Formica (Markl 1966), Camponotus Mayr, 1861 (Saini et al. 1982), Myrmecia Fabricius, 1804 (Liu et al. 2019), and various external structures (e.g., Emery 1900, Tulloch 1935). In a comprehensive study, Reid (1941) compared the mesosoma of winged and wingless species of different groups of Hymenoptera, including Formicidae. However, he only addressed external features. Janet’s (1898) treatment of the queen of a Myrmica species is the most detailed study of winged ant mesosomal anatomy to date and is meaningfully comparable to the highest quality studies on workers, namely Lubbock (1881), Markl (1966), and Liu et al. (2019). The very detailed work of Markl (1966) was dedicated to the mesosoma of Formica, in this case only covering the muscles and nerves. Skeletal elements were shown in the illustrations but not explicitly described in the text of that study. A comparative study on alates and workers of Camponotus also covers the muscles but is very general and does not address the anatomy in detail (Saini et al. 1982). The most recent treatment of an ant mesosoma is that of Liu et al. (2019), wherein Myrmecia nigrocincta Smith F., 1858 was studied with modern techniques for the first time. Although this work provided a detailed description of muscles, the endoskeletal elements were not described in detail, and many questions of homology remain unanswered. Moreover, the focus of Liu et al. (2019) was on an ant with specialized jumping capacity; thus, there is a degree of uncertainty about whether the skeletomuscular arrangement of this species can be generalized to other ants.

Considering the existing gaps in knowledge and the lack of consistency of morphological approaches and terminology, our primary aim is to provide a detailed documentation of the mesosoma of a well-known and ecologically generalized ant species. We focus on Formica rufa, a mound-building ant, which is not only the type species of the family Formicidae, but also occupies a wide range of climatic zones and is one of the most dominant and widespread ant species in Eurasia. Critically, from the morphological perspective, Formica are not specialized for hypogaeic or arboreal lifestyles, nor do they have specialized locomotory capacities (such as jumping, e.g., Musthak Ali et al. 1992). Formica is thus expected to be a reasonable comparison point for other ants. Due to the abundance and ecological importance of F. rufa in sustaining forest health and biodiversity, this species has been extensively studied (e.g., Dlussky 1967, Collingwood 1979, Gosswald 1989). Although workers of this species are not divided into discrete subcastes, they are highly polyphenic, with the body size ranging from 4.5 to 9 mm long (Collingwood 1979). Formica rufa, therefore, may also be used as a model for allometric variation of musculature and endoskeletal structures.

The combination of µ-CT, computer-based reconstructions, and 3D modeling, in particular, represents a cutting-edge methodology for the generation of high-resolution, virtual 3D models of anatomical tissues, organs, or whole-body parts (e.g., Friedrich et al. 2014, Wippler et al. 2016). The resulting 3D models can be easily and virtually manipulated, thus enabling highly detailed and comprehensive analyses of a variety of problems. Moreover, 3D imagery provides novel opportunities for the interactive presentation and dissemination of anatomical structures as 3D videos (e.g., Lauridsen et al. 2011, Faulwetter et al. 2013, Sarnat et al. 2016, Hita Garcia et al. 2017a), 3D PDFs (e.g., Berry and Ibbotson 2010, van de Kemp et al. 2014, Hita Garcia et al. 2017b), or embedded Sketchfab (https://sketchfab.com/) for interactive exploration of 3D surface models (Hita-Garcia et al. 2019, Sarnat et al. 2019). However, although it is now possible to create interactive, detailed 3D atlases of insect anatomy, almost all studies present the resulting 3D data in 2D (e.g., Rybak 2012, Rother et al. 2021), with very few exceptions, such as Berry and Ibbotson (2010).

Here, we employ a multi-modal approach to document the external and internal anatomy of the mesosoma of F. rufa. The objectives of our study are 1) to catalog all sclerites and muscles of the mesosoma, 2) to establish skeletomuscular homologies of ant mesosoma with those of other Aculeata, and 3) to unify mesosomal terminology based on these broader comparisons. To accomplish the second goal, we compare our observations to the results of other contributions on the ant mesosoma (e.g., Lubbock 1881, Janet 1898, Saini et al. 1982, Keller 2011, Boudinot 2015, Liu et al. 2019), and especially an earlier study on the nervous system and musculature of a Formica species (Markl 1966). Our overarching goal is to establish a starting point and reference for a larger effort to characterize the groundplan and evolution of mesosomal anatomy across the ants.

Material and Methods

Material

Most of the data presented in the current study are based on adult workers of F. rufa preserved in ethanol (collection code FHG01185: Germany, Hessen, Darmstadt, Prinzenberg, 49.836656°, 8.664995°, 240 m, forest edge and open area, 20.IV.2016, leg. F. Hita Garcia). Two F. rufa specimens (unique specimen identifiers: CASENT0741316 and CASENT0741323) were used for µ-CT scanning of mesosoma and the middle leg. In order to achieve a higher quality scan, the middle leg was detached from the body and scanned individually.
The specimens were stained in iodine solution 1 wk prior to the scanning date. Unfortunately, no *F. rufa* specimens were left for SEM, and instead, two specimens initially assigned to *Formica polyctena* Foerster, 1850 were used (collection code ANTC37555: Croatia, Lika-Senj, northwest of Konjisko, 44.531833°, 15.131778°, 870 m, pine forest, meadows, road margin, 19.IX.2018, leg. A. Richter). Both species, *F. rufa* and *F. polyctena*, are closely related, almost identical morphologically, and have the ability to interbreed successfully. In fact, based on seta counts of our specimens, the samples we used are likely to represent *Formica polyctena × rufa* hybrids according to the classification of Seifert (2021), which was used for species-level identification.

**Scanning Electron Microscopy**

Two specimens of *F. polyctena* (unique specimen identifier: CASENT0753212) were dissected to a differing degree (removal of legs on one side of the body or additionally removal of head and gaster). All samples were transferred to 100% ethanol (70, 80, 90, 95, 100%) and dried at the critical point in liquid CO₂ and gaster). All samples were transferred to 100% ethanol (70, 80, 90, 95, 100%) and dried at the critical point in liquid CO₂. Using a rotatable specimen holder with an Emitech K 850 Critical Point Dryer (Sample Preparation Division, Quorum Technologies Ltd., Ashford, England). The samples were glued on the tip of minute needles (either on the gaster or division, Quorum Technologies Ltd., Ashford, England) and gaster). All samples were transferred to 100% ethanol (70, 80, 90, 95, 100%) and dried at the critical point in liquid CO₂ and gaster). All samples were transferred to 100% ethanol (70, 80, 90, 95, 100%) and dried at the critical point in liquid CO₂. Using a rotatable specimen holder (Pohl 2010), SEM micrographs were taken with a Philips ESEM XL30 (Philips, Amsterdam, Netherlands) equipped with Scandum FIVE software (Olympus, Münster, Germany).

**Micro-CT Scanning and 3D Reconstruction**

The µ-CT Scanner used was a Zeiss Xradia 510 Versa 3D X-ray microscope operated with the Zeiss Scout-and-Scan Control System software (version 14.0.14829.38124) at the Okinawa Institute of Science and Technology Graduate University, Japan. The scanning parameters chosen consisted of a 40 kV (75 μA)/3 W beam strength with 4-s exposure time for the thorax and 7 s for the leg under a 4× magnification, which resulted in a voxel size of 2.97 and 1.40 μm, respectively. 3D reconstructions of the resulting scan projection data were done with the Zeiss Scout-and-Scan Control System Reconstructor (version 14.0.14829.38124) and saved in txf file format. Postprocessing of txf raw data was done with Amira 2019.2 software (Visage Imaging GmbH, Berlin, Germany) in order to segment individual structures into discrete materials. The segmented materials were then exported with the plugin script ‘multiExport’ (Engelkes et al. 2018) in Amira 2019.2 as 2D Tiff image stacks. VG-Studio 3.4 (Volume Graphics GmbH, Heidelberg, Germany) was used to create volume renders out of the Tiff image series.

**Image Processing**

SEM images were edited with Adobe Photoshop CC 2021 (Adobe System Incorporated, San Jose, CA). Adobe Illustrator CC 2021 (Adobe Systems Incorporated) was used to arrange figures into plates and label the figure plates.

**Terminology**

The terminology, particularly for musculature, is mainly based on Friedrich and Beutel (2008) and Beutel et al. (2014). We integrate this Hexapoda-wide terminology with the general terminology of ants (Bolton 1994) and the specific revisions thereof provided by Keller (2011), Boudinot (2015), and Serna and Mackay (2010), including the latter’s revision (Serna et al. 2011). Overall, our terminology is compatible with the Hymenoptera Anatomy Ontology (Yoder et al. 2010; HAO), unless otherwise noted. Of the presently used terms, 48 are not defined in the ontology; see Supp Table S3 (online only) for a complete list of term equivalencies between the present study and the HAO. A number of specific terms are used from various other sources: the sculptural term ‘imbricate’ is used following Harris (1979), with each cell of the imbrication referred to as a ‘scute’ following Mikó et al. (2016); the profurcal orifice through which the ventral nerve cord passes is referred to as the ‘neural foramen of the profurca’ (Porto et al. 2017); we refer to ‘upper’ and ‘lower mesopleural areas’ (e.g., Gibson 1986) instead of the traditional ‘aneisternum’ and ‘katepisternum’ (Bolton 1994; for our reasoning, see Discussion). The ‘posteroventral pronotal process’ is an acute yet rounded posteroventral corner of the pronotum.

We employ standard orientational terminology, recognizing the anteroposterior, dorsoventral, and lateromedial axes as well as the sagittal, frontal, and transverse planes. In the present work, we refer to objects situated close to the sagittal plane of the body as ‘mesial’, following vertebrate anatomical practices; these structures may also be labeled as ‘medial’. The only exception to the standard orientational terminology is in the description of the foreleg telopod (trochanter–tarsus), which is modified in ants such that it is bent anteriorly and oriented in the sagittal plane (Boudinot 2015). We refer to the apparent lateral and mesial surfaces of the foreleg telopod as the ‘posterior’ and ‘anterior surfaces’, respectively.

Note that we use the term ‘fusión’ to broadly refer to the lack of developmental differentiation between two or more sclerites that are freely articulating in other taxa, including other castes and/or other Auleata or Hymenoptera. ‘Fused’ sclerites are otherwise ‘sutured’ sensu (Snodgrass 1962). In this manner, we consider the dorsoventrally situated line between the mesopleural and metapleural regions the ‘mesometapleural sulure’ as it is unfused in other Auleata. Likewise, we label grooves situated ectad internal ridges as ‘sulci’, such as the epistomal sulcus of the head (e.g., Richter et al. 2019, 2020, 2021) or the ‘probisisternal sulcus’, i.e., the median and longitudinal discrimin sulcus of the prothorax. In some cases, especially for alates, there are externally visible lines that are weakly impressed. We refer to these simply as ‘lines’, such as the transscutal line which divides the mesoscutum from the mesoscutellum, or ‘sina’ in the case of muscle attachment scars, such as the parapsides and the ventrolateral mesoscutal (‘subpleural’) sina (e.g., Daly 1964).

**Homology**

We subscribe to the Remanean criteria of homology recognition, namely that candidate homologs correspond in position relative to other structures, have specific shared details, are connected by morphological intermediates, and are shared among closely related taxa, particularly for simple and serially repeated structures. See Boudinot (2018) and Boudinot et al. (2021) for greater detail and discussion of the process of homology induction and the testing of deductive predictions. Because our work does not directly incorporate comparisons with other Formicidae or Hymenoptera, nor does it include explicit phylogenetic modeling of character transformation, the homologies drawn in the present study should be considered novel or revised primary homology hypotheses. Initial homology hypotheses are drawn from the myrmecological studies cited in the Terminology section. See our Discussion for our rationale for key interpretations and term applications.

**Data Availability**

All specimens used in this study have been databased, and the locality and collection data are freely accessible on AntWeb (http://www.antebook.org). Each specimen can be traced by a
unique specimen identifier included in the preservation vial. The original µ-CT scans are available in txm format at the Dryad Digital Repository (https://doi.org/10.5061/dryad.pnvx0k6nw). In addition to the raw data at Dryad, we also provide freely accessible 3D models of all treated species on Sketchfab (https://sketchfab.com/arilab/collections/formica-mesosoma-anatomy).

**Results**

**Mesosomal Skeleton**

The mesosoma is composed of the three thoracic segments, their appendages, and the tergum of abdominal segment I (propodeum); it is elongate, circa (ca.) 0.4 x as long as the entire body and slightly more than 2 x as long as the greatest width of the pronotum. The cuticular surface is finely imbricate, with the microscopic form of the scutes varying considerably (Fig. 1); it is covered by a dense pubescence that comprises two seta classes, including fine, appressed setae (type 1, ca. 0.05 mm long), and sparse longer, standing setae (type 2, ca. 0.15 mm long; Figs. 1 and 2); the longer setae are relatively shorter and flatter on the pronotum and longer elsewhere; they are present on the pronotum, the prothorax, the mesonotum, the mesepisternum, the coxa, the femora, the tibia, and the tarsi; the longer tibial setae are arranged in two ventral rows, while the tarsal setae are complex of fused segments, including the mesothorax, metathorax, and propodeum. The prothorax comprises the pronotum, the paired anteroventral propleura, the ventral and posteromesially located prothorax anteriorly, but it is fully fused with the propodeum. The metathorax freely articulates with the furcasternum. The mesosoma posterior to the prothorax comprises a complex of fused segments, including the mesothorax, metathorax, and propodeum (Figs. 1 and 2), the erstwhile mesonotomesopectal suture is faint and curved, running from the anterior mesonotal margin to the posterior margin of the mesoscutum, the mesoscutellum, nor a mesosternum (Fig. 1E); the prophragma is not developed, rather the anterior mesonotal margin bears a short articulatory surface of smooth cuticle devoid of pilosity; the parapsides, nota, transcutal line, scutoscutellar sulcus, axillae, and mesoscutellum are not developed. The mesonotomesospectral suture is faint and curved, running
between the meso- (Sp₂) and metathoracic (Sp₃) spiracles (Fig. 1A); it partially demarcates the mesonotum from the mesopectus. The mesopectus is not differentiated into mesopleurae and mesosternum and lacks the ventral paramedian ‘subpleural’ signa of alates; anteromesially, it bears a transverse sulcus, the anterior mesopectal groove, which subtends the anterior mesopectal flange; the groove

Fig. 1. SEM images of mesosomal exoskeletal elements of *Formica polyctena*. (A) Mesosoma, lateral posterodorsal oblique view. (B) Metathoracic spiracle. (C) Propodeal spiracle. (D) Metapleural gland opening. (E) Metanotum, dorsal view. Cx1/2/3—pro-/meso-/metacoxa; P1/2/3—pro-meso-/metapleuron; IT—propodeum; N1/2/3—pro-/meso-/metanotum; Isp—propodeal spiracle; Pl3G—metapleural gland opening; sp2/3—meso-/metathoracic spiracle; PL—pronotal lobe.
forms an additional area for muscle attachment, while the flange is posteriorly margined by a transverse carina and lacks muscle attachment. The mesopleural area is divided into the upper and lower mesopleural areas by the weakly demarcated transepisternal line, which originates anteriorly at the mesothoracic spiracle, beneath the pronotal lobe; the lower mesopleural area continues anteriorly where it transitions to the mesosternal area; the posterolateral region of the mesopleural area is bulged and the cuticular thickness is

Fig. 2. SEM images of mesosomal exoskeletal elements of Formica polyctena. (A) Prothorax, lateral view. (B) Mesosoma excluding prothorax, lateral view. (C) Notum, dorsal view. (D) Mesosoma excluding pro-/mesothorax and petiole, dorsal view. (E) Prothorax, ventral view. (F) Pterothorax, ventral view. Cx1/2/3—pro-/meso-/metacoxa; Pl1/3—pro-/metapleuron; IT—propodeum; N1/2/3—pro-/meso-/metanotum; Isp—propodeal spiracle; PI3G—metapleural gland opening; sp2/3—meso-/metathoracic spiracle; a—mesometapleural suture; b—bulla of mesocoxal foramen; Pl—pronotal lobe.
decreased opposite the mesocoxal insertion relative to the rest of the mesopleural area, thus forming the bulla of the mesocoxal foramen. The mesepimeron is considerably reduced, being only represented by the metaspiracular operculum (Fig. 1B). The ventral mesoplectal area is dominated by broad, longitudinally oriented impressions, the epicnemial depressions; the epicnemial carinae are absent (note that, when present in Formicidae, the epicnemial carinae delimit the epicnemium laterally, rather than posteriorly as defined by the HAO); mesiad and posteromesiad the depressions is the mesosternal area, which is marked by the attachment of IIscm1, thus is triangular with the apex at the anterior end of the mesodiscrimen and the bases at the posteroventral margin of the mesothorax. The mesodiscrimen is represented by a faintly impressed longitudinal line on the ventral mesoplectal surface and corresponds to the internal mesodiscmenal lamella (Fig. 5C); it ends posteriorly at the mesofurcal pit, which corresponds internally to the mesofurca, which is less strongly developed than the profurca. The mesofurcal arms connect with the pleural wall through either an apodeme or a muscle (due to the low resolution of the µ-CT scan, it is not clear whether mesofurcal arms in Formica connect with the pleural wall via an apodeme or a muscle IIspm2 [Snodgrass 1942: 79]); they are connected to each other by the transverse mesofurcal bridge, creating a mesofurcal

Fig. 3. 3D reconstruction of propleuron of Formica rufa. (A) Mesosoma, side view. (B) Sagittal view. (C) Lateral view. (D) Dorsal view, anterior to the top. (E) Ventral view, anterior to the top.
neural foramen below it; the bridge forms a thin horizontal lamella anteromesially that serves as a site for muscle attachment.

**Metathorax**

The metanotum is present only as a shallow transverse groove with a small, raised strip in between the mesonotum and propodeum (Fig. 1E); it is delimited laterally by the metathoracic spiracles, which are enclosed by the distinctly elevated and circular metaspiracular operculum. The metapleural area is not differentiated into dorsal and ventral areas and is entirely fused with the propodeum posteriorly. The ventral surface of the metapectus, the metasternal area, is shorter than that of the mesopectus; it is marked anteriorly by a very indistinct line, the ventral mesometapetal lamella suture, which corresponds to the lateral portion of the paracoxal ridge (the internal and transverse metapetal lamella); mesially the ridge is wide, but it is gradually narrowed toward the lateral margin. The metadiscrimen is not marked externally, although the metadiscriminal lamella is distinct; it is shorter than its mesothoracic equivalent; dorsally and posteriorly, it bears the long, wide, and curved metafurcal arms. The metafurcal arms connect on their dorsal ends to the metapleural wall through the pleural apophysis, leaving marked endophragmal pits externally; metafurcal arms are not connected by a transverse bridge. Their distal portions are wider compared to the mesofurcal arms, thus providing a wider attachment area for muscles; each of their distal ends (apices) bears a short dorsal process. The posterior area of the

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**Fig. 4.** 3D reconstruction of profurca of *Formica rufa*. (A) Ventral view, anterior to the top. (B) Side view, anterior to the left. (C) Anterolateral view, anterior to the left. (D) Posterior view. (E) Lateral view, anterior to the left. (F) Posteroventral view.
metapleuron bears the metapleural gland bulla (Pl3G); externally, the bulla opens to the metapleural gland evaporatorium, which is a longitudinally oriented sulcus that is overhung by the posteriorly lobate longitudinal metapleural flange; the ventral metapleural gland flap is not developed.

**Propodeum**

The propodeum is distinctly delimited by the metapleural sulcus, which is in the form of a parabolic furrow; it is not subdivided by sutures, and it is completely fused with the metathoracic region; the lateral propodeal surface (lateropropodeum) bears the slit-like and slightly curved propodeal spiracles that are enclosed by a distinct oval bulge; the posterior propodeal surface (posteropropodeum, propodeal declivity) is slightly concave and glabrous where it is contacted by the petiole. The propodeopetiolar articulation comprises a socket, the propodeal foramen; it is margined by the propodeal flange, which bears a pair of ventromesially directed teeth that articulate on each side of the petiolar levator process, the propodeal condyles. Ventrally the metasternal area and the petiole are connected by a long intersegmental membrane. The metacoxal foramina are closed by the lateral propodeal bridges and thus independent of the propodeal opening.

**Legs**

The prothoracic legs differ from the meso- and metathoracic legs mainly in length, the form of the coxae, trochanters and their articulations, and the presence of a complex antenna-cleaning device, the strigil, which comprises the protibial calcar and probasitarsomeral notch and comb. The lengths of the fore and middle legs are about equivalent (Supp Table 1 [online only]); the procoxa is longer than the other two pairs of coxae, while the metacoxa is longer than the mesocoxa; the mesotibia is slightly longer than the protibia; the hind legs are distinctly longer than the other two pairs; the length ratio of the tarsi from anterior to posterior is 2.3:2.5:3.4 (Supp Table 1 [online only]). Nearly the entire surfaces of the brownish legs are covered by a dense vestiture of very fine appressed setae of ca. 0.025 mm length (type 1 setae), locally interspersed with some longer setae with a length of about 0.1 mm (type 2); thicker spine-like setae (type 3, ‘traction setae’ or ‘chaetae’) of different length and shape are concentrated on the apical parts of the tarsomeres and also inserted along the anterior and posterior edges and on the ventral side.

The procoxae are elongate, about twice as long as their maximum width; they are divided into proximal and distal regions by a distinct proximal inflection, the basicoxal sulcus. The prodisticoxal regions are cylindrical, moderately narrowing toward their apices; they are evenly convex anteriorly and laterally, whereas the posterior surfaces are almost flat; they are about twice as long as those of the middle coxae but with similar diameter; the prodisticoxal foramina are completely enclosed by a sclerite, more-or-less circular and oriented in the sagittal plane; proximad the foramina, the apicolateral and mesial disticoxal margins are tightly approximated, forming transverse fissures ventrad the disticoxal foramina. The prodisticoxal regions are very short dorsoventrally, anteriorly, mesially, and posteriorly, forming a short rim that is constricted in diameter relative to the prodisticoxal region; they are flange-like laterally, forming the lateral proximal processes; the mesial and lateral prothoracicoxal articulations are between the propleuron and the flanged lateral prodisticoxal rim, respectively. Type 2 setae are mainly developed anteriorly.
The protrochanters are slightly less than 1/3 as long as the procoxae (Supp Table 1 [online only]); they are divided into proximal and distal portions by a very distinct constriction. The proximal protrochanteral articulations are borne on the probasitrochanters, which are offset from the prodistitrochanters at a distinct angle; only one of the two proximal articulations is developed, which articulates with the corresponding flange-like and single prodisticoxal condyle; the posterior disticoxal articulation is not developed in ants. The prodistitrochanters are elongate-cup-shaped and, in lateral view, their narrow bases are rounded, with the anterior side slightly concave, the posterior side distinctly convex, and the apical edge nearly straight; they are apicodorsally produced, with the process in the form of a narrow wedge; their posterolateral apices are desclerotized and flexible.

The profemora are about 1.8 times as long as the procoxae (Supp Table 1 [online only]); their dorsal and ventral margins are very slightly sinuate; their greatest width is in their proximal 1/3; they narrow very slightly toward the base and apex; a very indistinct bulge is present distad the apical trochanteral process; their apicodorsal and apicoventral margins are emarginate, with the dorsal
emargination being narrower than the ventral; their anterior and posterior apices are produced ventrally as rounded lobes, the distal femoral flanges; each lobe internally bears a tibial acetabulum of the femur. The protrochantella (proprefemora) are distinctly developed ventrally but not dorsally, and thus appear wedge-shaped in lateral or posterior view. A few type 2 setae are inserted posteroventrally on the anterior half.

The protibia is slightly shorter than the profemur (Supp Table 1 [online only]); it is slender basally and widens toward its apex; it is trilobate proximally, with the paired anterior and posterior femoral condyles of the tibia articulating with the corresponding tibial acetabula of the femur, and the dorsomedian lobe fitting into the apicodorsal femoral notch; its proximal portion is slightly curved in posterior (lateral) view and narrows proximad the chordotonal bulge; the distal tibial foramen is uneven in contour because of the apicoventral tibial notch that surrounds the single apical spur, the calcar; the notch and calcar are subtended posteriorly by a brush of spatulate or paddle-shaped setae, the posteroventral apical tibial brush. The calcar is proximally associated with a very small flexible pad; it is 0.56 mm long and appears straight in ventral view but sinuate viewed from laterally; it has a single pointed apex; along its inner margin it bears a straight, very dense comb of flexible microtrichia (ca. 0.03 mm) which is subtended by a narrow strip of smooth cuticle; it lacks a transparent lamella, the ‘velum of the calcar’, along its ventral margin; its posterior surface is covered by a brush of very short, fine, and apically pointed microtrichia; its anterior surface is covered by a dense brush of longer, spatulate microtrichia; a ventral row of teeth is absent. In addition to the ventral double row, some type 2 setae are developed posteroventrally at the tibial apex.

The five-merous protarsus (five tarsomeres) is slightly longer than the profemur (Supp Table 1 [online only]); the tarsomeres are slender and nearly cylindrical. The probasitarsomere is more than 1.3x as long as the remaining tarsomeres combined (Supp Table 1 [online only]); its proximal portion, articulating with the tibial apex, is curved. The tarsal part of the strigil comprises the basitarsal notch and comb, which are situated in the proximal third of the basitarsomere, with the notch forming a shallow ventral concavity (ca. 0.4 mm long) and the comb being a straight and very dense line of flexible microtrichia (ca. 0.02 mm); a slightly irregular row of strong setae (0.1–0.12 mm) is present, along the posterior edge, whereas only few thinner setae are inserted on the anterior side; the anteroventral side of the tarsomere bears a dense field of paddle-shaped tenent setae (ca. 0.1 mm), which are slightly extended, thus spatulate apically; several strong setae (ca. 0.15 mm) are present anteriorly and posteriorly on the apical region, and an additional pair is inserted subapically on the dorsal side; the apex of the probasitarsomere is truncated; tarsal plantulae are lacking as on all other tarsomeres. Protarsomere 2 is about 1/4 as long as 1 (Supp Table 1 [online only]) and moderately widening toward the apex; its proximal portion, articulating with the probasitarsomere is shaped like a narrowed peduncle; several pairs of spine-like setae (type 3) are inserted between the fine setae on the ventral surface; the apical armature of setae is similar to that of tarsomere 1. Protarsomere 3 is slightly shorter than 2 (Supp Table 1 [online only]); it is also articulated with a short peduncle and distinctly widening distally; the apex is emarginated; two pairs of slightly flattened spines with a pattern of longitudinal ruffles are inserted at the slightly extended anterior and posterior apices, and an additional pair between them on the

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**Fig. 7.** Muscles of prothorax of *Formica rufa*, dorsal view, anterior to the top. (A) The simplified diagram of skeletomuscular system redrawn from the work of Markl (1966: Fig. 18). (B) The 3D reconstruction of the prothoracic muscles.

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ventral side; two groups of spine-like setae are present on the ventral surface, four closer to the anterior edge and three closer to the posterior margin. Protarsomere 4 is shorter than 3 (Supp Table 1 [online only]) and widening strongly toward the extended anterior and posterior apical edges, which are separated by a distinct emargination. Protarsomere 5 is about twice as long as 4 (Supp Table 1 [online only]) and similar in shape to tarsomere 2; the stronger apical and ventral setae are longer and thinner than those of tarsomeres 1–4; the apical margin displays rounded lobes separated by a median emargination; a pair of long setae is inserted close to the distal edge.

The pretarsus comprises the manubrium dorsally, the paired claws that articulate with the unguitractor, the arolium with the arcus, and the planta on the ventral side proximad this median adhesive lobe; the glabrous proximal end of the manubrium is inserted in the apical emargination of protarsomere 5; a pair of long setae (ca. 0.1 mm) is inserted on its surface; its distal part reaches deeply into the arolium. The unguitractor plate is well-sclerotized, rectangular with rounded corners, and largely covered by a scaly surface pattern; a vestiture of relatively short and thin setae is present on most areas of the surface. The planta is distally adjacent to the unguitractor plate; it bears a pair of setae (ca. 0.08 mm) laterally; additional shorter setae are distributed over the entire surface. Auxiliary sclerites are not recognizable. The well-developed pretarsal claws bear a dense pattern of very short microtrichia on their basal portion and an array of setae of different thickness and length; a tooth is lacking; a groove is present mesally on the distal half. The apically lobed arolium appears smaller compared to the claws; it is ca. 0.07 mm long and 0.05 mm wide near its base; a dense pattern of papillae with pointed spines is present dorsally and at the apical margin, whereas the ventral surface is smooth. The arcus is a thin, sclerotized structure and not visible externally.

The mesocoxa and metacoxa are very similar to one another in shape, differing primarily in size; they are about half as long as the procoxa and appear more globular, with the mesocoxa being slightly smaller than the metacoxa; they are closely approximated laterally and mesially. The meso- and metatibial basical sulci divide the coxae into their respective basicoxal and disticoxal regions; the sulci are disc-like, slightly impressed around their respective basicoxae, and more-or-less oriented in the frontal plane of the body. The meso- and metadisticoxal regions are small and ball-like, with their dorsoventral longitudinal axes at a distinct angle to the longitudinal axes of their respective disticoxae; their rims are torqued and bear the insertions of the coxal muscles. The meso- and metadisticoxal regions are more or less water-balloon-shaped in anterior view; their disticoxal foramina are open, and they are anteroposteriorly narrow and dorsoventrally long and margined by a distinct carinal rim; their trochanterial acetabula are located anteriorly and posteriorly, and are associated with longitudinal fissures, similar to the distal, closed rim of the procoxa. The meso- and metatrochanters are similar to one another in shape and are very distinct from the protrochanters; they are approximately bell-shaped, with anterior and posterior condyles; their proximal portion is narrow and neck-like but not set at a distinct angle. The remaining leg segments are similar to those of the foreleg with the following exceptions: a bastarsomeral strigil is not developed; the ventroapical spur is simple and straight, being lanceolate in form with only a finely barbirulate inner comb; the ventroapical tibial notch is sclerotized, i.e., there is a sclerotized bridge that separates the ventroapical spur from the distal tibial foramen.

List of Mesosomal Muscles of Ant Workers
(Summary is given in Table 1 and Supp Table 2 [online only])
Based on Lubbock (1881), Janet (1898), Snodgrass (1942), Saini et al. (1982), Liu et al. (2019), and especially Markl (1966). Comments on shape and size and sites of origin and insertion apply to Formica (Markl 1966) unless otherwise noted. The nomenclature follows Friedrich and Beutel (2008) and Beutel et al. (2014), where Roman numerals refer to the segment (e.g., I = prothorax). The numbers used by Snodgrass (1942) for the muscles of the honey bee are given in brackets or if the muscle is lacking in the honey bee, the number used by Markl (1966) for Formica.
In this section, we present the descriptions of the skeletomuscular elements and the functional hypotheses together to improve readability and to enhance the interpretability of the functions.

**Prothoracic Muscles**

**Pronoto-cephalic Muscles**

*Levators of the Head.* Idlm1, *M. prophragma-occipitalis* (40 + 41); an extrinsic head muscle (cervical muscle), converges toward its insertion on the head as a single slender bundle (Figs. 6 and 7; Models 1, 4). **Functional note:** Levator of the head if symmetrically contracted or rotator if asymmetrically contracted.

Idvm9, *M. profurca-occipitalis* (43); a cervical muscle with a relatively broad origin, converges anteriorly on a narrow tendon (Figs. 7 and 9; Models 1 and 4). **Functional note:** Levator or rotator, as for Idvm9.

**Propleuro-cephalic Muscles**

*Levators of the Head.* Itpm1, *M. pleurocrista-occipitalis* (42a); an extrinsic head muscle with a fairly broad area of origin (Fig. 8; Models 1 and 4). **Functional note:** Levator or rotator, as for Idvm9.

Itpm2, *M. propleuro-occipitalis* (42b & c); an extrinsic head muscle with a fairly broad area of origin (Fig. 8; Models 1 and 4). **Functional note:** Levator or rotator, as for Idvm9.

**Profurco-cephalic Muscles**

*Depressor of the Head.* Ivlm3, *M. profurca-tentorialis* (44); a large, tripartite extrinsic head muscle (Figs. 7 and 9; Models 1 and 5). **Functional note:** Depression of the head, with a strong tendon that appears hollow with minute filaments in the lumen.

**Pronoto-propleural Muscles**

**Pronoto-propectal Stabilizers.** Itpm3, *M. pronoto-pleuralis anterior* (48); moderately sized conical, vertical muscle (Figs. 7 and 9; Models 1 and 4). **Functional note:** Pulls the cervical apodeme and anterior propleuron toward the pronotum (Duncan 1939).

**Functional note:** Interpretation from Markl (1966: 48).

Itpm4, *M. pronoto-apodemalis anterior* (49); a band-shaped muscle composed of two parallel horizontal subcomponents (Figs. 7 and 9; Models 1 and 4). **Functional note:** Anterolaterally on the pronotum; I: apex of the profurcal arm and the propleural apophysis (pleural arm), i.e., a mesially directed projection of the posterior portion of the propleuron. **Functional note:** Interpretation from Markl (1966); according to Snodgrass (1942), it is a protractor of the propectus.

**Propleural Levator/Rotator.** Idvm5, *M. pronoto-cervicalis anterior* (Markl 1966: 46 + 47); strongly developed and fan-shaped, with separate innervation of its two bundles (Markl 1966; Figs. 7 and 9; Models 1 and 4). **Homology note:** We remain uncertain about the correspondence of this muscle in *Apis*.

**Pronoto-profurcal Muscle**

*Profurcal Stabilizer.** Itpm5, *M. pronoto-apodemalis posterior* (Markl 1966: 50); band-shaped and vertically oriented (Fig. 7; Models 1 and 4). **Homology note:** We remain uncertain about the correspondence of this muscle in *Apis.*
Table 1. Skeletomuscular correspondences among *Formica* and previously studied taxa, adapted and modified from Liu et al. (2019, table 1). References: *Vespula* (Duncan 1939), *Apis* (Snodgrass 1942), *Myrmecia* (Li et al. 2019), *Myrmica* (Janet 1898), *Camponotus* (Saini et al. 1982), *Lasius* (Lubbock 1881), *Formica* (Markl 1966). Reference was also made to Friedrich and Beutel (2010) to understand the holometabolan pterothoracic musculature. Note that, for muscles specific to winged individuals, a number of homologies remain uncertain.

| Family   | Vespidae | Apidae | Formicidae |
|----------|----------|--------|------------|
| Genus    |          |        |            |
|          | *Vespula* | *Apis* | *Myrmecia* | *Myrmica* | *Camponotus* (w, aq) | *Lasius* (w, aq) | *Formica* (w) |
| Muscles of the prothorax | | | | | | |
| Idlm1    | –        | 40 + 41 | +         | M. 35 | 1 | a | 40 and 41 |
| Idlm5    | Is1[50] and Is2[51] | 45 | +         | – | – | – | 45 |
| Idvm5    | Ipm1[37] and Ipm2[38] | 47 | +         | M. 42 | 6 | c | 46 and 47 |
| Idvm9    | Os2[33]  | 43 | +         | M. 38 | 2 | a | 43 |
| Idvm18   | Ilm6[48] | 55 | +         | M. 51 | – | – | 55 |
| Itpm1    | O1[32]   | 42a | +         | M. 37 | – | b | 42/1 |
| Itpm2    | O1[32]   | 42b and c | + | M. 36 | – | c1 and c2 | 42/2 |
| Itpm3    | Ipm3[39] and Ipm4[40] | 48 | + (misidentified as Itpm5) | M. 45/ | – | f | 48 |
| Itpm4    | Ipm5[41] | 49 | +         | M. 48 | 10 | e | 49 |
| Itpm5    | Ipm6[42] | 50 | + (misidentified as Itpm6) | M. 48 | – | g | 50 |
| Ipcm2    | Ilm7[49] | + (over-looked, present in *Apis*) | + (missed, present in *M. croslandi*) | M. 40 | – | d1 | mcr |
| Ipcm4    | Ilm2[44] | 53 | +         | M. 43/44 | – | h | 53 |
| Ipcm8/Iscm6 | Ilm3[45] | 61 | +         | M. 47/ M. fäch. fm. | – | k and l | 61 |
| Ivlm1    | Ifp[36]  | 51 | +         | M. 41 | – | d | 51 |
| Ivlm3    | Os3[34] and Os4[35] | 44 | +         | M. 39 | 3 | b | 44 |
| Ivlm7a   | Is4[53]  | 52 | +         | M. 53 | 19 | m | 52/1 |
| Ivlm7b   | Is5[54]  | 52 | + (missed, present in *M. croslandi*) | M. 36 | 18 | n | 52/2 |
| Iscm1    | Ilm1[43] | 54 | +         | M. 46 | – | i | 54/2 |
| Iscm3    | Ilm5[47] | 54/57 | + (missed, present in *M. croslandi*) | M. 36 | 18 | n | 52/2 |
| Iscm4/Iscm2? | Ilm4[46] | 56 | +         | M. ext. cx. | – | j | 56 |
| Muscles of the mesothorax | | | | | | |
| Illdm1   | Illd1[56] | 71 | –         | M. vib. l. | 20 | β | – |
| Illdm1   | Illd4[57] | 72 | –         | M. vib. t | 21 | 0 | – |
| Illdm6   | Ilpm6[56] | 82 | –         | M. 85? | 23 | – | – |
| Illdm8   | Illd2[58] | 78 | –         | M. 81, M. 82? | – | – | – |
| Ilspm1   | Ilpm1[59] | 77 | –         | M. 90 | 22 | – | – |
| Iltpm5/6 | Ilpm4[62] | 75 | –         | M. 91 | 25 | – | – |
| Iltpm7   | m3Ax[63] | 76a | –         | M. 92 | 24a | – | – |
| Iltpm9   | Ilpm[60] and Ilpm3[61] | 76b/c | – | M. 93 | 24b | – | – |
| Ilpspm1  | 2osp    | 73 | + (missed, present in *M. croslandi*) | M. ferm. st 1 | – | – | 73 |
| Ilpspm1  | Ilsp2[73] | – | + (missed, present in *M. croslandi*) | – | – | – | 79 |
| Ilpspm2  | Ilp1[70] | 79 | + (missed, present in *M. croslandi*) | M. 83? | – | – | – |
| Ilpspm1  | 3osp    | – | + (missed, present in *M. croslandi*) | – | – | – | – |
| Ilscl1   | Il1m1[65] | 81 | +         | M. 58 | 28? | s | 81 |
| Ilscl2   | Ilpm4[68] | 83 | + (missed, present in *M. croslandi*) | M. 61 | 29 | r | 83 |
| Ilscl3   | Ilpm2[66] | 83 | +         | M. 60 | 27 and 31? | q and t | 83 |
| Ilscl3/4 | Ilpm5[41] | 80 | + (misidentified as Ilscl4) | M. 57 | 30 | p | 80 and 82 |
| Ilscl6/Ilpcm5/Idvm7 | Ilm3a&b[67] | 86 | + (missed, present in *M. croslandi*) | M. 62 | 26? | o | 86 |
| Genus | Vespula | Apis (f) | Myrmecia (w) | Myrmica (aq) | Camponotus (w, aq) | Lasius (w, aq) | Formica (w) |
|-------|---------|----------|-------------|-------------|------------------|--------------|-------------|

**Muscles of the metathorax**

| Muscles | Genus | Length (mm) | Comments | Length (mm) |
|---------|-------|-------------|----------|-------------|
| IIIdvm6 | IIpm5[80] | 105 | - | M. 86? | 37 | - | - |
| IIIdvm1 | IIpm1[76] | 101 | - | ? | 36 | - | - |
| IIIdvm6 | IIpm3a&b[78a&b] | 102 | - | M. 87 | 39 | - | - |
| IIIdvm7 | IIpm2a&b[77a&b] | 100 | - | M. 88 | 38a | - | - |
| IIIdvm9 | IIpm2a&b[77a&b] | 100 | - | M. 89 | 38b | - | - |
| IIIscm1 | IIIm1[81] | 104 | + | M. 64 | 41 | z | 104 |
| IIIscm2 | IIIm2[82]? | 106 | + (missed, present in M. croslandi) | M. 66b | - | z1 | 106 |
| IIIscm3 | IIIm2[82]? | 106 | + (missed, present in M. croslandi) | M. 66a | 43 | x1 | 106 |
| IIIpcm3/4 | IIIm4[84] | 103 + 105? | + (misidentified as IIIdvm4 and IIIpcm5) | M. 65 | 42 | y and y1 | 103 and 105? |
| IIIscm6/IIIpcm5/IIIdvm7 | IIIm3[83] | 109 | + (missed, present in M. croslandi) | M. 63 | 40 | x | 109 |
| IIIspm2 | - | - | + (missed, present in M. croslandi) | M. 84 | - | - | - |
| IIIvlm3 | IIIis3[87] | 119 | + (misidentified as IIIvlm2) | M. 69/M. v. a. 4 | 33 | u1 | 119 |
| IIIvlm2 | IIIis1[85] | 118 | + (misidentified as IIIvlm7) | M. 68/M. v. m. 4 | 35 | v | 118 |

**Muscles of the abdomen**

| Muscles | Genus | Length (mm) | Comments | Length (mm) |
|---------|-------|-------------|----------|-------------|
| IA1 | Iadl1[88] | 120 | + | M. 75 | 34 | u | 120 |
| IA2 | Iadl2[89] | 121 | + | M. 67 | 32 | w | 121 |
| IAAspim1 | ospI[90] | 122 | + (missed, present in M. croslandi) | M. ferm | - | - | 122 |
| IAAspim2 | dpsI[91] | 123 | + (missed, present in M. croslandi) | M. ouv | - | - | 123 |

**Intrinsic muscles of the legs**

| Muscles | Genus | Length (mm) | Comments | Length (mm) |
|---------|-------|-------------|----------|-------------|
| I-AI-IIltkm1 | n.c. | 59, 84, 107 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltkm2 | n.c. | 60, 85, 108 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltkm3 | n.c. | 62, 87-88, 110 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltfm1 | n.c. | 63, 89, 111 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltfm2 | n.c. | 64, 90, 112 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltfm1 | n.c. | 64, 90, 112 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltfm2 | n.c. | 65, 91, 113 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltfm1 | n.c. | 69b, 93, 117b | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltfm2 | n.c. | 67, 93, 115 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |
| I-AI-IIltfm3 | n.c. | 68, 94, 116 | n.c. | n.c. | n.c. | n.c. | n.c. | n.c. |

+ = present; - = not observed or recorded; ? = uncertain; n.c. = not covered.
Propleuro-profurcal Muscle

Profurcal Stabilizer. Ivlm1, *M. profurca-cervicalis* (51); relatively small cervical muscle (Figs. 6 and 7; Models 1 and 5). O: Medially on the anterior surface of the profurcal arm and the anterior profurcal lamella; I: posterior apex of the cervical prominence. **Functional note:** Also stabilizes the propleuron. **Homology note:** As for Idvm5, this muscle also attaches to the cervical prominence of the propleuron.

Procoxal Muscles

Remoters of the Procoxa. Idvm18, *M. pronoto-coxalis lateralis* (55); a moderately sized extrinsic coxal muscle that is slender and conical (Figs. 8 and 12; Models 1 and 4). O: Dorsolaterally on the posterior portion of the pronotum; I: posterolaterally on the lateral procoxal process. **Functional note:** Lateral remotor.

Iscm4, *M. profurca-coxalis lateralis* (56); a very large and bipartite extrinsic coxal muscle (Figs. 7 and 12; Models 1 and 3). O: Posterior surface of the profurcal arm, reaching the posterior apex, and on a median ridge of the posterior profurcal surface; I: posterolaterally on the lateral procoxal process, slightly ventrad the insertion of Idvm18. **Functional note:** Lateral remotor. **Note:** Based on our scan data, we cannot resolve whether this muscle attaches solely to sclerite or also on a membrane to some degree. Also, in the
future study, it may be necessary to recognize this as two distinct muscles.

Rotators of the Procoxa. Ipcm2, M. procoxa cervicalis transversalis (Markl 1966: mcr, M. cruciatus); a slender pair of extrinsic procoxal muscles that cross over one another in the midline (Figs. 8 and 12; Models 1 and 3). O: Posterior apex of the cervical prominence on the opposite side of the body; I: anterolaterally on the lateral procoxal process. Functional note: Also stabilizes the prothorax (Markl 1966).

Iscm1, M. profurca-coxalis anterior (Markl 1966: 54/2); a triangular, fan-shaped extrinsic coxal muscle (Figs. 8 and 12; Models 1 and 3). O: Mesially on the basisternal prodiscrimenal lamella; I: anterolaterally on the lateral procoxal process, mesial to the insertion of Ipcm2. Functional note: Rotates the procoxa anterolaterad Markl (1966). Homology note: The insertion site of this muscle does not match with muscle 54 in Apis (Snodgrass 1942); additional examinations are required to resolve the homology of this muscle. Markl (1966) suggested that Iscm1 and Iscm3 together could correspond to muscle 54 in Apis (Snodgrass 1942).

Promotors of the Procoxa. Iscm3, M. profurca-coxalis medialis (Markl 1966: 54/1); a very large extrinsic coxal muscle (Figs. 7 and 12; Models 1 and 3). O: Posterior surface of the profurcal arm and profurcal stem; I: mesially on the procoxal rim. Functional note: Mesial promotor. Homology note: The homology of this muscle with muscle 54 in Apis is uncertain (Markl 1966).

Ipcm4, M. propleuro-coxalis superior (53); an elongated and triangular extrinsic coxal muscle that has a broad attachment area (Figs. 7 and 12; Models 1 and 3). O: Ventral surface of the dorsal propleural ridge; I: anterolaterally on the lateral procoxal process. Functional note: Lateral promotor.

Protrochanteral Muscles

TrochanteralDepressors. Ipcm8, M. propleuro-trochanteralis and/or Iscm6, M. profurca-trochanteralis (61); a long and well-developed conical extrinsic leg muscle (Figs. 7 and 12; Models 1 and 3). O: On two sclerites: posteriorly on the ventral surface of the dorsal propleural ridge and laterally on the posterior surface of the profurcal arms; I: protrochanteral depressor apodeme. Functional note: According to Markl (1966), this muscle is also an important remotor of the proleg and also a depressor of the trochanter. Homology note: Because this single muscle has origins on two unfused sclerites, its precise homology with either M. propleuro-trochanteralis or M. profurca-trochanteralis is uncertain; alternatively, it may be homologous with both.

Ictm3, M. procoxa-trochanteralis medialis (62); the largest muscle originating in the coxa (Fig. 12; Models 1, 2, and 6). O: Anterior, posterior, and mesial procoxal surfaces, covering up to ¾ of the inner surface area; I: depressor apodeme of the trochanter, together with Ipcm8 (61).

Fig. 10. 3D reconstruction of meso- and metathoracic muscles of Formica rufa; anterior to the left. (A) Lateral view. (B) Sagittal view. (C) Dorsal view. (D) Ventral view.
Trochanteral Levators. Ictm1, *M. procoxa-trochanteralis anterior* (59) anterodorsal trochanteral muscle (Fig. 12; Models 1, 2, and 6). O: Anterodorsal procoxal surface; I: base of the trochanter. Functional note: Anterior levator.

Ictm2, *M. procoxa-trochanteralis posterior* (60); posterodorsal trochanteral muscle (Fig. 12; Models 1, 2, and 6). O: Posterodorsally on the procoxal wall; I: base of the trochanter, with its tendon close to that of Ictm1. Functional note: Posterior levator.

Intersegmental Muscles

Prothoracic Depressors. Idlm5, *M. pronoto-phragmalis anterior* (45); a fan-shaped intersegmental muscle with two bundles that converge toward their insertion (Fig. 6; Models 1 and 4). O: Lateral surface of the pronotum at about midlength; I: anterior mesonotal margin, distinctly lateral Idlm1 (40 + 41). Functional note: In addition to depressing the pronotum, this muscle also stabilizes the connection with the mesonotum (Snodgrass 1942, Markl 1966).

Ivlm7a, *M. mesofurca-profurcalis* (52/1); a large ventral intersegmental muscle that connects the mesofurca with the profurca (Figs. 6 and 7; Models 1 and 5). O: Along the mesofurcal arm and on the upper mesopleural area below spiracles 2; I: posterolaterally on the dorsal margin of the furcasternum. Functional note: A depressor of the prothorax according to Lubbock (1881, Plate XI, fig. 2: m), and possibly also a retractor of the profurca (von Kéler 1955: 74. Musculus mesofurcaprofurcalis).

Ivlm7b, *M. profurca-mesofurcalis* (52/2); a slender ventral intersegmental muscle between the profurca and the mesofurca (Figs. 6 and 7; Models 1 and 5). O: Laterally on the posterior surface of the profurcal arm; I: triangular mesofurcal process. Function: Stabilizes the connection between the pro- and mesothorax, retractor of the prothorax; elevator of the prothorax according to Lubbock (1881, Plate XI, fig. 2: n).

Mesothoracic Muscles

Mesocoxal Muscles

Promotors of the Mesocoxa. Ilpcm3/4, *M. mesanepisternalis-coxalis* (80); a large, lateral coxal muscle (Figs. 10 and 12; Models 1 and 3). O: On the anteroventral region of the mesopectus, including a low transverse groove along the anterior edge and lower lateral wall (anterior mesopectal groove); I: anterolaterally on the mesocoxal rim. Function: Interpreted as a possible mesocoxal levator in addition to its promotor role by Markl (1966). Homology note: According to Markl (1966), this muscle could be a product of a fusion of muscles 80 and 82 of *Apis* (Snodgrass 1942). Ilpcm3 or Ilpcm4 (80) was misinterpreted as Ilscm4 in Liu et al. (2019).

Ilscm1, *M. mesofurca-coxalis anterior* (Markl 1966: 81); one of the three furca-coxal muscles (Figs. 10 and 12; Models 1 and 3). O: Ventrally on the mesodiscrimenal lamella of the ventral mesopectal region; I: anterolaterally on the mesocoxal rim. Functional note: Interpretation from Snodgrass (1942).

Remotors of the Mesocoxa. Ilscm2, *M. mesofurca-coxalis posterior* (83); a fan-shaped muscle that originates ventrad Ilscm3 (Figs. 10 and 12; Models 1 and 3). O: Ventrally on the mesodiscrimenal lamella and on the mesoventral portion of the mesopectal region; I: posteromesially on the mesocoxal rim.

Ilscm3, *M. mesofurca-coxalis medialis* (83); a very large muscle that originates dorsad Ilscm2 (Figs. 10 and 12; Models 1 and 3). O: Medially on the mesofurcal arm, the mesodiscrimenal lamella, and the ventral mesopectal region; I: mesially on the mesocoxal rim.

Mesotrochanteral Muscles

Depressors of the Trochanter. Ilscm6, *M. mesofurca-trochanteralis* and/or Ilpcm5, *M. mesanepisterno-trochanteralis* (86); a trochanteral muscle that originates outside of the coxa (Figs. 10 and 12; Models 1 and 3). O: Laterally on the mesofurcal arm and on the anterodorsal mesopleural region; I: apodeme of the trochanter. Functional note: Depressor of the trochanter, together with Ilctm3 and Ilpcm5, Markl (1966) interpreted this muscle as a remotor. Homology note: The specific homology of this muscle relative to other Hymenoptera is uncertain.

Ilctm3, *M. mesocoxa-trochanteralis medialis*. (87, 88); a large muscle composed of two branches (Figs. 12; Models 1, 2, and 6). O: Medial mesopectal wall, covering up to ¾ of the internal surface; I: on the depressor apodeme of the trochanter, together with Ilscm6 and Ilpcm5.

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Fig. 11. Muscles of metathorax and propodeum of *Formica rufa*, sagittal view, anterior to the left. (A) The simplified diagram of skeletonmuscular system redrawn from the work of Markl (1966: Fig. 23). (B) The 3D reconstruction of the metathoracic and propodeal muscles.
Levators of the Trochanter. Ilctm1, M. mesocoxa-trochanteralis anterior (84); a fan-shaped anterior levator (Fig. 12; Models 1, 2, and 6). O: Anterior mesocoxal wall; I: anterior trochanteral process according to Markl (1966: Trochanterzapfen), but this structure is not clearly recognizable in µ-CT scans.

Ilctm2, M. mesocoxa-trochanteralis posterior (85); a fan-shaped posterior levator (Fig. 12; Models 1, 2, and 6). O: Posterior mesocoxal wall; I: posterior trochanteral process, with its tendon inserting close to Ilctm1.

Intersegmental Muscles
Ilvlm3, M. mesofurca-metafurcalis. (Markl 1966: 79); extremely thin and short (Fig. 6; Model 1). O: Lateral arm of the mesofurca near the dorsal apex; I: lateral arm of the metafurca. Function: Stabilizes the furcae and the connection between pterothoracic segments. Homology note: According to Markl (1966), the homology with muscle 79 in Apis (Snodgrass 1942) is uncertain.

Muscles of the Metathorax and Propodeum
Metacoxal Muscles
Promotors of the Metacoxa. IIIpcm3/4, M. metanepisternalis-coxalis (103); large, lateral coxal muscle (Figs. 10–12; Models 1 and 3). O: Laterally and ventrally on the metasternal plate, extending to the lateral wall of the propodeum; I: laterally on the metacoxal rim. Functional note: Markl (1966) suggested a levator function, but this appears rather unlikely. Homology note: According to Markl (1966), this muscle could be a product of a fusion of muscles 103 and 105 in Apis (Snodgrass 1942).

IIIpcm5, M. metanepisterno-trochanteralis (106); a posterior trochanteral depressor. O: Laterally on the metacoxal rim; I: trochanteral apodeme. Functional note: Depressor of the trochanter, together with IIIctm3 and IIpcm5. Markl (1966) remarked that this muscle also functions as a remotor, which we find unlikely because the metatrochanter is dicondylic. Homology note: The homology is uncertain. Markl (1966) recorded this muscle as originating on the propodeum.

Depressors of the Trochanter. IIIpcm1, M. metacoxal-trochanteralis medialis (110); a large coxal muscle (Figs. 10–12; Models 1 and 3). O: Metathorax and propodeum; I: trochanteral apodeme. Functional note: Depressor of the trochanter, together with IIIctm3 and IIpcm5. Markl (1966) remarked that this muscle also functions as a remotor, which we find unlikely because the metatrochanter is dicondylic. Homology note: The homology is uncertain. Markl (1966) recorded this muscle as originating on the propodeum.

Remotors of the Metacoxa. IIIscm1, M. metafurca-coxalis posterior (106); relatively small furca-coxal muscle (Figs. 10–12; Models 1 and 3). O: Metadiscal lamella ventrad IIIscm3; I: posteromesially on the metacoxal rim.

IIIscm2, M. metafurca-coxalis medialis (106); a large coxal muscle (Figs. 10–12; Models 1 and 3). O: Posterior and lateral surface of the metathorax region, metadiscal lamella, and transverse metapleural lamella of the metapleural region; I: mesially on the metacoxal rim.

Metatrochanteral Muscles
Depressors of the Trochanter. IIIscm6, M. metacoxal-trochanteralis and/or IIIpcm5, M. metanepisterno-trochanteralis (109); posterior trochanteral depressor (Figs. 10–12; Models 1 and 3). O: Laterally on the metathorax and on the anterodorsal area of the metathoracic region; I: trochanteral apodeme. Functional note: Depressor of the trochanter, together with IIIctm3 and IIpcm5. Markl (1966) remarked that this muscle also functions as a remotor, which we find unlikely because the metatrochanter is dicondylic. Homology note: The homology is uncertain. Markl (1966) recorded this muscle as originating on the propodeum.

IIIctm3, M. metacoxal-trochanteralis medialis (110); a large coxal muscle with two branches (Fig. 12; Models 1, 2, and 6). O: Ventromesial metacoxal wall, covering up to ⅔ of the surface; I: trochanteral depressor apodeme, together with IIIscm6.

IIIctm1, M. metacoxal-trochanteralis anterior (107); fan-shaped anterior levator (Fig. 12; Models 1, 2, and 6). O: Anterior...
metacoxal wall; I: anterior trochanteral process (Markl 1966: Trochanterzapfen).

IIIctm2, M. metacoxa-trochanteralis posterior (108); fan-shaped posterior levator (Fig. 12; Models 1, 2, and 6). O: Posterior metacoxal wall; I: posterior trochanteral process via a tendon, insertion close to IIIctm1.

Spiracular Muscles

Occlusors of the Spiracles. IIpspim1, M. mesanepisterno-spiracularis (73); a slender muscle (Figs. 6 and 9; Models 1 and 2). O: Anterior margin of the upper mesopleural area; I: small plate close to the orifice of metathoracic spiracle.

IIIpspim1, M. mesopleura-spiracularis (Markl 1966: Ɛ); a small closing muscle of the metathoracic spiracle (Fig. 6; Models 1 and 2). O: Ventrally on the corner of the peritreme of the metathoracic spiracle; I: small plate close to the orifice of metathoracic spiracle.

IAspim1, M. spiracularis I superior (122); a short muscle (Fig. 11; Models 1 and 2). O: Sclerotized bar above the propodeal spiracle; I: sclerotized bar below the propodeal spiracle.

Dilator of the Propodeal Spiracle. IAspim2, M. spiracularis I posterior (123); very thin and long (Figs. 6 and 11; Models 1 and 2). O: propodeal bridge; I: sclerotized bar below the propodeal spiracle.

Depressor of the Petiole. IIIvlm2, M. metafurca-abdominosternalis superior (118); a slender longitudinal muscle (Figs. 6 and 11; Models 1 and 2). O: Posteriorly on the upper portion of the metatarsal arm and the dorsal region of propodeum (Markl 1966: epinotum); I: mesioventrally on the anterior margin of the petiole.

Intrinsic Leg Muscles Distad Coxa (Not Covered in Friedrich and Beutel 2008). The intrinsic muscles of the leg are highly conserved (e.g., Snodgrass 1935, Larsen 1966). The muscles listed in the following are probably generally present in ants like in other groups of insects.

Femoral Muscle

Iftm1/IItfm1/IItfm1, M. trochantero-femoralis (63, 89, 111); long and fan-shaped, with a very broad attachment area (Fig. 13; Model 6). O: Dorsally on the trochanteral wall; I: proximal rim of the femoral foramen. Function: Reductor of the femur.

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Fig. 13. 3D reconstruction of the midleg muscles of Formica rufa. (A) Dorsal view, anterior to the bottom. (B) Lateral view, anterior to the front. (C) Ventral view, anterior to the top. (D) Pretarsal claw retractor muscle (IIfpm1) in lateral view, anterior to the front.
Tibial Muscles
Itf1m/IIft1m/IIIftm1, M. femuro-tibialis dorsalis (64, 90, 112); bipinnate (Fig. 13; Model 6). O: Dorsal wall of the femur; I: axial tendon or a membrane on the proximal end of the tibia. Function: Extensor of the tibia.

Itf2m/IIftm2/IIIftm2, M. femuro-tibialis ventralis (65, 91, 113); large and bipinnate (Fig. 13; Model 6). O: Ventral wall of the femur; I: genuflexor plate (HAO: sclerite to which tibial muscle inserts). Function: Flexor of the tibia.

Pretarsal Muscles
Itfpm1/IItfpm1/IIItfpm1, M. femuro-pretarsalis (69a, 95a, 117a; Fig. 13; Model 6); fan-shaped, with an extremely long tendon inserted on the pretarsal unguitractor plate (Fig. 11). O: Proximoventral femoral lobe; I: unguflexor plate. Function: Flexor of the pretarsal claws.

Itipm1/IItipm1/IIItipm1, M. tibio-pretarsalis (69b, 95b, 117b; Fig. 13; Model 6); bipinnate, with a long tendon fused with the tendon of Itfpm1 (M. femuro-pretarsalis; Fig. 13). O: Ventral tibial wall; I: unguflexor plate. Function: Retractor of the pretarsus.

Basitarsal Muscles
Ibhm1/IIbhm1/IIIbhm1, M. tibio-basitarsalis ventralis (67, 93, 115); flat and fan-shaped (Fig. 13; Model 6). O: Ventral tibial wall; I: posteriorly on the tibial articular process, opposite tibm2. Function: Retractor of the tarsus in the fore and mid legs, levator in the hind leg.

Ibhm2/IIbhm2/IIIbhm2, M. tibio-basitarsalis anterior (66, 92, 114); a flat muscle (Fig. 13; Model 6). O: Lateral tibial wall; I: anteriorly on the articular process of the tibia, opposite tibm1. Function: Promotor of the tarsus.

Ibhm3/IIbhm3/IIIbhm3, M. tibio-basitarsalis posterior (68, 94, 116); a flat conjunctival muscle (Fig. 13; Model 6). O: Lateral tibial wall; I: ventral conjunctival membrane at the distal end of the tibia. Function: Depressor of the tarsus. This muscle lacks an antagonist, the opposite movement—elevation—is produced by the elastic conjunctival membrane.

Discussion
Our analysis provides a comprehensive atlas of the skeletomuscular elements of the Formica mesosoma and a baseline for further comparative work on anatomy and functional morphology across the ants. Overall, in the worker of E. rufa, we identified and described 49 muscle pairs, of which 38 originate in the trunk of the mesosoma (prothorax 20, mesothorax 8, and metathoracicopropodeal complex 11) and 11 are intrinsic to each leg (coxa 3, trochanter 1, femur 3, and tibia 4). As these counts match those previously observed for the worker of E. polyctena (Markl 1966) and M. nigrocincta (Liu et al. 2019; L.A., unpublished data), we expect that the muscles described herein are plesiomorphic for the genus Formica and lineages within “formicoid” clade. From the phylogenetic-comparative perspective, the taxon pair Myrmecia and Formica span the crown node of the nondoryline ‘formicoids’; thus, equivalent muscle counts in these two taxa provide a reasonable groundplan hypothesis for this clade (for phylogeny, see, e.g., Branstetter et al. 2017b). Prior reports for Lasius (Lubbock 1881) and Myrmica (Janet 1898) include far fewer muscles, despite the presence of wing muscles in the latter, indicating that these studies were incomplete. Direct comparison of a winged ant mesosomal muscle apparatus with Apis (Snodgrass 1942) and other aculeates is expected to resolve issues of muscle presence/absence and may improve nomenclature, particularly as observations of alate Hymenoptera were used to construct the HAO (Yoder et al. 2010).

Skeletomusculature
Key Functional Findings
We observed a series of key features are found in the formicid worker prothorax that are linked with specific functions exercised by ants. The head is not only prognathous, as generally recognized, but has a postocciput that is derived relative to other Aculaeta and which fits into the prothorax like the ball of a ball-and-socket joint, thus allowing for greater cranial mobility. Combined with the reduction of the flight apparatus, the cranial and notopleural muscles are proportionally large; thus, we agree with Keller et al. (2014) that this condition provides increased power for both lifting and a wide range of motion. The elongated procoxae and their derived coxotrochanteral articulations likely result in highly efficient locomotion on the ground or other surfaces (Boudinot 2015) and may have enhanced the capacity to carry loads with the mandibles (Boudinot 2015, Boudinot et al. 2020b). The meso- and metacoxal foramina are, moreover, directed ventrally, and the coxae are bent at an approximate right angle, allowing these joints to serve as support columns of the body while maximizing stride length by keeping the legs in the frontal plane (Boudinot et al. 2020a).

The structure of the posterior mesosoma of workers is obviously affected by loss of the flight function, having a distinctly reduced inventory of muscles, particularly the absence of both the large indirect flight muscles and the smaller, complex direct flight muscles (Table 1). We postulate a trade-off between intramesosomal flexibility, which is a necessary prerequisite of a functional flight apparatus, and mechanical rigidity, which may improve strength, protection, and perhaps be a safety factor under load. Reduced degrees of freedom within the mesosoma likely allows for a simplification of the muscle apparatus, as it has also been described from the strongly sclerotized Coleoptera (Beutel and Haas 2000, see also Beutel et al. 2021). These modifications and those of the head will be valuable to consider together with the derived conditions of the metasoma (e.g., Rasnitsyn 1980, Dlussky and Fedoseeva 1988, Hashimoto 1996, Perrault 2004), which has yet to be studied in totality or using μ-CT technology. One feature of note, however, is the paired propodeal condyles (‘propodeal teeth’), which pinch the articular process of the petiole; this modifies the range of metasomal motion and has been considered to be phylogenetically important among the Apocrita (e.g., Rasnitsyn 1980, Rasnitsyn and Quicke 2002).

Leg Musculature and Endoskeleton
Two special elements of our study are the treatment of leg musculature and endoskeletal structures. Previous studies on ants (and almost all other groups of insects) have not addressed intrinsic leg muscles, presumably due to the widely held assumption that these muscles are highly conserved. With these muscles enumerated and depicted, however, future studies can focus on differences that may be of potential functional value, such as transformations of the proximal leg articulations (e.g., Boudinot 2015) or variation in muscle volume, pectination, and orientation. Regarding the endoskeleton, very few efforts have been made to date to operationalize these structures in ants for comparative study, although they have been illustrated previously to some degree (e.g., Lubbock 1881, Markl 1966, Liu et al. 2019). A few notable exceptions focus on the prosternum, which is known to display variation of phylogenetic importance in bees and wasps (e.g., Mikó et al. 2007, Vilhelmsen et al. 2010, Porto et al. 2017, Meira and Gonçalves 2018, Boudinot et al. 2020a), and is expected to be informative for ants (e.g., Lattke 1994, 2004, B.E.B., unpublished data).

Because of the limited focus on the prosternum, we consider a number of observations worthy addressing in the hope of...
encouraging future studies. In addition to clarifying the identity of the basisternum and furcasternum for Formicidae and providing the most detailed illustration of these structures for Hymenoptera to date (Figs. 2E, 3A, D, E, and 4), we observe, for the first time in ants, that the profurcal pits of the prothorax are paired and located at the extreme dorsal end of the furcasternum (Fig. 4D and F). We expect that the paired condition is plesiomorphic for the Aculeata, as this has been recorded for Platygastroidea, Chalcidoidea, Trigonalidae, and Bethylidae, while it is unpaired in Mymarommatidae and most Cynipoidea (Vilhelmsen et al. 2010). A potential synapomorphic condition of the Anthophila or within the Apoidea more broadly is implied by the reports of Porto et al. (2017) and Meira and Gonçalves (2018), who recorded a single profurcal pit for the Apidae and Halictidae, respectively. However, we observe that the unpaired ‘profurcal pit’ indicated by these authors corresponds internally to the prodissinonal lamella, while the actual paired branching structure (i.e., the furca) and its paired pits are located posterior to this and are unlabeled (see, e.g., fig. 1 in Porto et al. 2017; compared with our Fig. 4). We note that this misapplication of the term ‘profurcal pit’ can be traced to Michener (1944), who labeled the median unpaired groove of the basisternum as the ‘apophyseal pit’ (e.g., his fig. 26) and left the true profurcal pits unlabeled. The internal structures of ant and bee prothorax are, otherwise, highly distinct and deserve greater scrutiny, particularly with extended sampling within the Formicoidea, Apoidea, and among other vespid aculeates.

Within the Formicidae, the only study to our knowledge to adequately illustrate the internal anatomy of the prothorax is that of Lattke (2004), who included three SEMs for Heteroponerinae Mayr, 1887 and two for Gnamptogenys Roger, 1863, both being genera of the Ectatomminae. A clear distinction between Formica and the genera sampled by Lattke (2004) is the form of the furcasternum, which is roughly triangular in the former and roughly rectangular in the two ectatommine genera. Among these ants, we also observe that the profurcal arms of all three taxa are lamellate, i.e., dorsi-ventrally broad, and are fused medi ally, forming both the neural bridge and the anterior and posterior profurcal lamellae. From the presently available sampling, some degree of homoplasy is indicated for the neural bridge, as the profurcal arms are also lamellate and medially fused in at least some Figitidae (Vilhelmsen et al. 2010: fig. 41E), Evanidae (Vilhelmsen et al. 2010: fig. 10C), Gasteruptiidae (Vilhelmsen et al. 2010: fig. 10D), Halictidae (Meira and Gonçalves 2018), and Apidae (Snodgrass 1935, 1942, Porto et al. 2017), but these arms are much narrower and do not form a neural bridge in at least some Tenthredinidae (Weber 1927), Stephanidae (Vilhelmsen et al. 2010: fig. 41A), Ichneumonidae (Vilhelmsen et al. 2010: fig. 41C), Scelionidae (Mikó et al. 2007: fig. 7), Proctotrupidae (Vinuelas et al. 2010: fig. 40D), and apparently the Scoliidae (Betrem 1928). We are certain that further comparison will reveal additional variation to characterize within the Formicidae and other Aculeata and Hymenoptera more broadly.

Among the other endoskeletal structures, we observe an internal, transverse ridge at the boundary of the meso- and metasternal regions. Such a ridge was not remarked upon for Myrmecia (Liu et al. 2019), in which it appears to be highly reduced, possibly in association with saltatorial specialization. The only prior work on ants to explicitly label this structure is that of Janet (1898), who referred to it as ‘la lame transverse’. Its presence in Formica and Myrmica suggests that this ridge may be a consistent structure among ants. Upon broader comparison, we propose the homology of this structure with the ‘paracoxal ridge’ of the symphytan grade of Hymenoptera (Vilhelmsen 2000).

Positionally, the transverse ridge of Formica and Myrmica corresponds in location to the paracoxal ridges of other Hymenoptera, being situated between the meso- and metacoxae. Furthermore, it is also in contact with the anterior base of the metacoxa in Formica and the ‘sawflies’. With respect to musculature, the paracoxal ridge bears the origin of a basalar muscle in winged Hymenoptera (Friedrich and Beutel 2010: IIIspm1, Beutel et al. 2014, Mikó et al. 2016: pl13a-ba, Willsch et al. 2020). Although the basalareas are not developed in worker ants, the ridge of Ampulex and Scelipteron also bears the origin of the lateral metacoxo-metacoxal muscle (Willsch et al. 2020: fu3l-cx3), a condition that is shared with Formica (Illsm2m, M. metafurca-coxalis posterior; see Table 1 for extended muscular correspondences). Given the present sampling, Formica differs in that the ridge is not clearly marked by an external ‘paracoxal sulcus’ (e.g., Vilhelmsen 2000, Lanes et al. 2020). Because the paracoxal ridge of Formicidae has not been recorded for over a century, our findings here bring to light further new characters of interest for the study of ant evolution and functional morphology.

Terminological Considerations

From the terminological perspective, we break with myrmecological tradition (e.g., Tulloch 1935, Bolton 1994, Serna and Mackay 2010) in that we do not recognize a distinct katepisternum of the meso- and metapleurites. The katepisternum in other groups of holometabolous insects is a small transverse sclerite located anterior to the pterothoracic coxae and articulating with them mesially (Vilhelmsen 2000: fig. 1A, B, Friedrich and Beutel 2010). In very clear contrast, the lower mesopleural region of ants is nearly vertical, and the lateral coxal articulation is immovably fused to the mesoscutum. Therefore, rather than an upper ‘anepisternum’ and lower ‘katepisternum’, we recognize ‘upper’ and ‘lower mesopleural regions’, which are divided by a longitudinal line or groove that corresponds internally to a ridge which divides the natal and coxal muscles in winged individuals (Janet 1898). Furthermore, we discourage the use of ‘pleuron’ and ‘sternum’ for the meso- and metathoracic segments, as the ancestral pleurae and sternae are fused, forming the meso- and metaplecta. Instead, we recommend recognizing ‘pleural and sternal regions’ of their respective segments.

Following the above provision regarding the misapplication of ‘katepisternum’ and ‘anepisternum’, we emphasize our usage of ‘mesopectus’ and our new application of the ‘epicnemium’ concept. In Hymenoptera, the pleural and sternal regions are undifferentiated (Snodgrass 1910), and the sternum of Holometabola is largely internalized (the ‘cryptosternite’ theory; see, e.g., Weber 1928, Ferris 1940, Compere 1962, Matsuda 1970), likely forming the discernible (e.g., Gibson 1993). For this reason, we refer to these ‘fused’ (undifferentiated) pleurosternal sclerites as the mesopectus, as done for alate ants (Boudinot 2015), with the addition of the mesonotum which is also immovably connected to the dorsal pleural region in workers and ergatoid gynes.

With respect to the epinotum, we recognize that the mesopectus of all ant castes bears a set of consistent landmarks (B.E.B., unpublished data), specifically an anterolateral carina that extends from the mesothoracic spiracle to the posterolateral corner of the mesoscutum, and a transverse flange subtended by a posterior groove at the anteroventral mesopleural margin. This carina laterally delimits the procoxal contact surface of the mesoplectal venter, thus functionally corresponding to the ‘epicnemial carina’ of other Hymenoptera, while the flange lacks a specific term in other hymenopteran groups (see HAO term equivalences, Supp Table 3 [online only]). Although it would be tempting to refer to this flange and groove as the epicnemium, it is apparent through comparison
across the Hymenoptera that the epicenium, or ‘coxal scrobe of the mesoscutus’, is present in addition to these structures and is variable in shape and size, depending on the relative lengths and shapes of the procoxae and mesoscutus (Boudinot et al. 2020a, B. E. Boudinot et al., in preparation).

In many hymenopteran taxa, such as the Ichneumonidae and Tiphiiidae, the epicenium is a small zone restricted to the anterior portion of the mesoscutus, whereas in others, e.g., Chrysididae and ‘Crabronidae’, it is a much larger region. The major, pertinent distinction between Formicidae and other hymenopteran clades is the length of the procoxae, and the fact that the epicenial carina of ants does not delimit the epicenial area posteriorly, which is otherwise a definitional condition of the carina in the HAO. Furthermore, in Formica, other Formicinae, and various Dolichoderinae, the epicenial carina is absent entirely (Figs. 1 and 2), and the epicenial depressions are in the form of small impressions on the otherwise convex ventral surface of the mesoscutus (Fig. 2). We anticipate that refined comparison of the mesopetal contact surfaces and their carinate or ecarinate margination will reveal diagnostic apomorphies for the Formicidae and clades within.

Application of µ-CT Methods
As noted above, even though µ-CT and computer-based 3D reconstructions open up numerous possibilities for in-depth analyses of morphology, previous studies using 3D models of ants actually present the data in 2D in the form of colored illustrations based on snapshots taken from renderings of the reconstructed and segmented 3D models (e.g., Khalife et al. 2018; Richter et al. 2019, 2020, 2021; Peeters et al. 2020). When morphological data have been presented in 3D, they have been in the form of 3D surface models of entire ants but with very few or no details about internal anatomy (e.g., Sarnat et al. 2016; Hita Garcia et al. 2017a, b). Furthermore, despite the widespread use of 3D atlases in neuroanatomy, the presentation in the actual publication is almost always in 2D (e.g., Rybak 2012, Rother et al. 2021). For the first time in ants, we provide 3D models stored on the Sketchfab platform (https://sketchfab.com/arilab/collections/formica-mesosoma-anatomy) that are directly embedded in this publication, in addition to 2D illustrations of our 3D reconstructions. The 3D models serve as an additional data source to be used together with the anatomical descriptions and 2D illustrations above since they are completely interactive in that they can be rotated, scaled, and manipulated. In addition, we have annotated the most important structures.

We believe that by providing 3D interactive models our data allow a higher level of reproducibility compared with previous studies and will encourage further interest for the study of insect/ant anatomy. Learning and understanding morphology is time-consuming, often requires experience with manual dissection or histology, and the development of considerable expertise for the anatomical structures of interest. However, our well-segmented and annotated skeletonmuscular system, presented in 3D, has the potential to accelerate this learning process since interested users can study the anatomy of the mesosoma of Formica in higher detail and can easily compare it with physical specimens of other ants and insect taxa. Furthermore, we aim to show that it is possible to create interactive, detailed 3D atlases of ant/insect anatomy to be used as baseline for future studies of other ants and Hymenoptera. Finally, since the interactive 3D models are openly and freely available on Sketchfab, we hope to stimulate general attention to entomology and insect morphology in particular.

Conclusion
Our study contributes to the knowledge of ant mesosomal anatomy, which remains very limited at present. Effectively, our work is a modern revision of Markl’s (1966) treatment of Formica that is done using nondestructive technology, and in the broader context of comprehensive studies for non-formicid groups of Hymenoptera (Snodgrass 1942, Mikó et al. 2007, Vilhelmsen et al. 2010, Porto et al. 2017, Meira and Gonçalves 2018), and other insect orders, such as Coleoptera (Larsén 1966, Baehr 1975, Beutel and Komarek 2004, Friedrich et al. 2009), and Antliophora (e.g., Friedrich and Beutel 2010), among others. In the present study, we provide a detailed documentation of the mesosoma of a well-studied and ecologically generalized ant species, F. rufa, address certain homology issues, and suggest functional interpretations for each of the muscles. We hope that this work will form a template for future comparative works focused on the elucidation of mesosomal evolutionary transformations and the functional–ecological diversification of ants. Future studies will benefit from expanded phylogenetic sampling, such as the ‘poneroid’ clade and Leptanillinae, as well as focused documentation of the skeletonmusculature of winged mesosomata.

Supplementary Data
Supplementary data are available at Insect Systematics and Diversity online.

Supplemental Table S1. The measurements of the leg segments of Formica rufa (unique identifiers: CASENT0741323 and CASENT0741316) in mm.

Supplemental Table S2. Summary of muscle origins, insertions, and functions in Formicidae.

Supplemental Table S3. Tabulation of presently used terms versus Hymenoptera Anatomy Ontology (HAO) preferred terms and their unique record identifiers (URIs, http://purl.obolibrary.org/obo/HAO_#####). HAO terms are bolded when an alternative preferred label is available. Presently used terms are organized by order of appearance for structures of the trunk and muscles but are organized proximal to distal for the legs.

Acknowledgments
We thank the Okinawa Institute of Science and Technology Graduate University (OIST) Imaging Section for providing access to the Zeiss Xradia micro-CT scanner and Shinya Komoto for general support. This work was supported by several Japan Society for the Promotion of Science (JSPS) grants-in-aid KAKENHI grants [no. 21J20268 to L.A.; no. 17K15180 to E.P.E.; no. 18K14768 and 21K06326 to F.H.G.], and a grant from the Japan Ministry of the Environment (Environment Research and Technology Development Fund no. 4-1904 to E.P.E.). In addition, A.R. was supported by a scholarship of the Evangelisches Studienwerk Villaest eV, and B.E.B. is supported by an Alexander von Humboldt Research Fellowship. This work was also supported by subsidy funding to OIST.

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