

## Trophic ecology of two co-existing Sub-Antarctic limpets of the genus Nacella: spatio-temporal variation in food availability and diet composition of Nacella magellanica and N. deaurata

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#### Abstract

Interactions between algae and herbivores can be affected by various factors, such as seasonality and habitat structure. Among herbivores inhabiting marine systems, species of the order Patellogastropoda are considered key organisms in many rocky coasts of the world. *Nacella* species are one of the most dominant macro-herbivores on the rocky shores of the sub-Antarctic ecoregion of Magellan. However, the importance of its key role must be associated with its trophic ecology. The objective of this work was to evaluate spatial and temporal variabilities in the dietary composition of two intertidal *Nacella* species, considering grazing on macro- (macroalgae) and microscopic (periphyton) food. The composition of periphyton and the availability of macroalgae in the winter and summer seasons were examined at two localities of the Magellanic province, alongside the gut contents of *N. magellanica* and *N. deaurata*. The dietary composition differed between the two *Nacella* species, as well as between seasons and locations. The differences observed in the diet of the two species of *Nacella* may be mainly due to their respective distributions in the

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intertidal zone. Both species presented a generalist strategy of grazing, which is relationed to the seasonality of micro- and macroalgae availability and to the variability of the assemblages between the localities. This research was the first to perform a detailed study of the diet of intertidal *Nacella* species.

#### Keywords

Gastropoda, Magellanic Province, herbivory, macroalgae, Nacellidae, periphyton

#### Introduction

The structure and dynamics of intertidal ecosystems depend on both abiotic factors (e.g., temperature, substrate, and climate) and biological interactions (e.g., predation, competition, recruitment) (Castilla and Duran 1985, Menge 2000, Díaz and McQuaid 2011, Murray et al. 2016). Among the biological interactions, predation through herbivory is one of the most relevant processes, since it helps determine the structure and functioning of ecosystems (Camus et al. 2008). In rocky shores ecosystems, herbivory can modify the spatial and seasonal patterns of algal communities (Aguilera 2011). These changes are not only related to the presence or abundance of grazers but also to the species to which it belongs (species identity) (O'Connor and Crowe 2005). However, interactions between algae and the different species of grazers can be directly or indirectly affected by multiple factors (Iken 1999). Among these factors, seasonality affects the coverage and biomass of micro- and macroalgae (Cubit 1984, Santelices 1987, Iken 1999, Jenkins et al. 2001), and consequently the diet of intertidal herbivorous species (Hill and Hawkin 1991). Seasonality itself may vary depending on the latitudinal range of rocky coasts, since, seasonal fluctuations in biomass of algae communities (micro and macroalgae) over the year increase with the latitude (Hill and Hawkin 1991, Thompson et al. 2000, Jenkins et al. 2001, Ojeda 2013), affecting the availability of food for different species of the food chain, including herbivores (Hill and Hawkin 1991). At the same time, habitat structure and high environmental heterogeneity imply spatial variation in the richness, abundance and structure of algae communities (Hill and Hawkin 1991, Benedetti-Cecchi and Cinelli 1997, Ojeda 2013), thus affecting the diet of herbivores (Branch 1971).

The main investigations on the ecology of herbivore-algae interactions have been carried out on molluscs of the order Patellogastropoda (Rubio et al. 2015). Patellogastropods or 'true limpets' are some of the most ubiquitous mollusks composing the marine littoral hard-substrate communities (Ponder and Lindberg 1997). Most of the 'true limpets' are herbivorous grazers, some on macroalgae, but a majority on sessile aquatic biota, attached to submerged substrate including diatoms, algal spores, detrital deposits and other bottom organism in combination with microbial bio-films (periphyton) (Branch 1981, van Dam et al. 2002). Much of the herbivore-algae interactions have been studied from an experimental point of view, evaluating the effect of exclusion or predation on the colonization of macroalgae and invertebrates (Hill and Hawkin 1991, Díaz and McQuaid 2011, Rubio et al. 2015). Nevertheless, research concern-

ing gut contents is not so extensive within patellogastropod limpets (see Rubio et al. 2015). This information will highlight the roles of species in the functioning of coastal systems (Camus et al. 2008, Aguilera 2011).

Following this, the current paradigm of herbivory in "limpets" has recently changed (Rubio et al. 2015). For example, studies have addressed the role of animal items in some patellogastropod species (Camus et al. 2008), indicating that these limpets actually present omnivorous habits, thus affecting the classical trophic networks in intertidal systems (Camus et al. 2008, Rubio et al. 2015). Small-scale studies of trophic ecology in patellogastropods have highlighted the dietary variability among congeneric species mainly due to their spatial distribution (Santina et al. 1993). Branch (1971), from the study of the trophic ecology of 11 patellogastropods species along the South-African coast, found that the environmental heterogeneity between sites (e.g., different types of substrates, coastal geomorphology, wave exposure) is related to the variability of algal communities, which influences the species diet. Meanwhile, on a seasonal scale, patellogastropods demonstrated a generalist diet that follows the available food in the habitat (Branch 1971, Hill and Hawkin 1991, Jenkins et al. 2001, Aguilera 2011). It is important to note that among patellogastropods with generalist habits, some species are exclusive consumers of periphyton (not macroalgae) (Chapman and Underwood 1992). Therefore, the diet and food preferences of key herbivores like patellogastropods and their temporal and spatial dynamics will help us to better understand the functioning of the rocky coastal environment.

The rocky shores of the channels and fjords of the Magellan Ecoregion present high environmental heterogeneity, influenced by several geomorphological (e.g., type of substrate; Ojeda et al. 2014), oceanographic (e.g., salinity variation; Silva and Calvete 2002), climate (e.g. seasonal variation in solar radiation and temperature; Marambio et al. 2017) and biological (e.g., diversity of biotopes associated with macroalgae; Soto et al. 2012) factors. The richness of patellogastropods in the Magellanic Province is represented by eleven species (Linse 1999), among which the most represented genus is Nacella (Valdovinos and Rüth 2005, Aranzamendi et al. 2009, González-Wevar et al. 2010, González-Wevar et al. 2011), a dominant group among the macro-benthic invertebrates, especially on rocky shores (Guzmán 1978, Ojeda et al. 2014). Along the Strait of Magellan and the sub-Antarctic channels and fjords this group also has an important role in the feeding habits of local communities (indigenous people, fishermen; Ojeda 2013). According to lastest phylogenetic studies of the group, four valid taxonomic units are recognized in the Magellanic Province: Nacella magellanica (Gmelin, 1791), N. deaurata (Gmelin, 1791), N. flammea (Gmelin, 1791) and N. mytilina (Helbling, 1779). Despite the dominance of the four species on the coastal zone, some differences in the distribution are visible; i) only N. deaurata and N. magellanica live in rocky intertidal environments (Aranzamendi et al. 2009, González-Wevar et al. 2011), ii) N. magellanica is more common in the middle intertidal zone, and iii) N. deaurata distributes from the middle and lower intertidal zone to the shallow subtidal (González-Wevar et al. 2011, Rosenfeld 2016). However, related information about the trophic ecology of Nacella species is still under study. The diet of Nacella intertidal species (Andrade and Brey 2014, Andrade et al. 2016) was evaluated upon gut content and stable isotopes but only at a very local level (one time and locality) and evaluating macroscopic food content, however the complete characterization of the diet (microalgae, macroalgae and invertebrates) and the temporal and spatial variations have not yet been addressed. Therefore, the dynamics of the trophic ecology of *Nacella* species remains poorly understood and is essential to comprehend the functioning of the rocky shores ecosystem of the Magellanic channels and fjords. Our objective was to determine the dietary composition of the intertidal species *N. deaurata* and *N. magellanica* across spatial and temporal variations, and evaluate the differences between the two species.

## Material and methods

The present study was carried out in two locations of the Magellanic Province: Puerto del Hambre (53°37'S, 70°54'W), Strait of Magellan and Otway Sound (53°04'S, 71°19'W; Fig. 1). Puerto del Hambre is in the central microbasin of the Strait of Magellan (Valdenegro and Silva 2003), the central microbasin, is filled with Estuarine Water, that is the result of the mixture of Subantarctic Waters from the Pacific with fresh water, coming from the western microbasin, which move towards the east overflowing the Charles III island constriction and sinking into the central basin and filling it, because of its higher density. The entire central micro-basin column is composed of saline estuarine water and average salinity values of 30.8 psu and average temperature of 7 °C (Valdenegro and Silva 2003). The substrate is characterized by intertidal plataforms, constituted by large rocky extensions regularly covered during the tide (Pisano 1980). Otway Sound is approximately 151 km long from west to east and is a postglacial lake (Kilian et al. 2007). Geographically, it presents a connection to the Skyring Sound through the Fitz Roy Channel and to the western side of the Strait of Magellan through the Jerónimo Channel (Valdenegro and Silva 2003). Otway Sound presents less saline superficial water (27.75 psu) that flows into the Strait of Magellan through the Jerónimo Channel (Valdenegro and Silva 2003). The substrate is also made of marine plataforms, similar to those of Puerto del Hambre (Pisano 1980).

#### Composition and abundance of macroalgae

In order to estimate seasonal variability of algal communities, quantitative sampling of macroalgae was carried out three times throughout the winter season in 2014 (June, July and August) and three times in summer in 2015 (Januar, Februar and March). The NaGISA (Natural Geography in Shores Areas) methodology was used for the quantitative sampling at both study sites, so that the biodiversity of coastal communities was quantified (Iken and Konar 2003). The design of NaGISA sampling protocol is intentionally basic and intends to yield baseline data for the sampling sites (Iken and Konar 2003). Three intertidal levels (high, middle and low according to the vertical height



**Figure 1.** Location of study sites, circle = Puerto del Hambre and square = Otway Sound. Abbreviations: **a**, **d** general view of both localities **b**, **e** images of the middle intertidal **c**, **f** images of the lower intertidal.

of the shore were assigned to categories defined by Benedetti-Cecchi and Cinelli's protocol (1997). Sampling was carried out in the two intertidal levels inhabited by *Nacella* species, at middle and lower levels. At each intertidal level, three random quadrats (n = 6) of  $50 \times 50$  cm (area of  $2500 \text{ cm}^2$ ) were analysed per site. Therefore, the sampling design was of factorial type: 6 (Month) × 2 (localities) × 2 (levels) × 3 quadrats: a total of 72 samples. Each macroalgal sample was placed in a plastic bag for further taxonomic identification (Skottsberg 1941, Wiencke and Clayton 2002, Boraso de Zaixso 2004) at the laboratory. The abundance of macroalgae was determined by dry biomass (g. species<sup>-1</sup>.quadrant<sup>-1</sup> in 2500 cm<sup>2</sup>) after approximately 48 h at 60 °C (Rutten 2007) and weighed with an analytical balance RADWAG AS 220/C/2, (± 0.0001 g).

## Composition and richness of periphyton

In parallel, the periphyton was sampled following the protocol of Ocaña and Fa (2003). Using a small quadrant of  $5 \times 5$  cm, a hammer and a chisel, a small area of the rock was extracted. Subsequently, the entire surface of the rock was scraped with a brush and preserved in 10% formalin. To estimate the relative abundance of periphyton in the quadrants, a cross-linked Petri dish with 50 points was used to record the number of points of intersection of each taxon, following the protocol of Aguilera et al. (2013). Periphyton sampling was performed at the same intertidal levels and sites as for the macroalgae.

## Gut contents

In order to evaluate the diet, 10 individuals of each Nacella magellanica and N. deaurata species were collected at each localities from the middle and lower areas of the intertidal levels (Fig. 1a, b, c, d, e, f). In order to determine whether Nacella species have a maximum algal availability in summer compared to winter, this sampling was performed three times during summer and three times during winter. Therefore, the sample design was of factorial type: 6 (Month)  $\times$  2 (localities)  $\times$  2 (species)  $\times$  10 individuals = 240 samples. Each individual collected was injected with 10% formaldehyde to preserve the gut contents and allow the subsequent identification of dietary items. To identify the periphyton composition, 1 ml of gut content was analyzed under an inverted microscope with light contrast (Aguilera et al. 2013). A stereomicroscope was used to analyze the macroscopic gut content. The identification of each dietary item was carried out at the most acurrate taxonomic level possible (genus level and species when it was possible). Organisms for which identification is complex, such as diatoms and some specific macroalgae or invertebrate taxa, were only identified at genus level (Aguilera et al. 2013). In the case of macroscopic items, two classifications were made: i) specific level (e.g., species, genus or family) and ii) functional level. The macroalgae were classified upon the structural hardness of the thallus according to Steneck and Watling (1982) and Santelices et al. (2009).

In the gut content analysis, we used the dietary richness (number of dietary items) of each individual of both *Nacella* species, and we calculated the occurrence frequency (%) of each item, which is the proportion of individuals containing each recorded item. In addition, the relative abundance (%) of each item in the digestive tract was estimated for each individual collected. The relative abundance of periphyton in the gastric contents was estimated using an inverted microscope and a reticulated glass slide with 50 points, thus recording the number of points of intersection of each taxon, as in Aguilera et al. (2013). The relative abundance (percentage value) of the macroscopic content was estimated using a stereomicroscope and a plate with a grid of 30 points of intersection, according to Aguilera (2005).

#### Data analysis

The composition and abundance of micro- and macroalgae in relation to sampling event, species and height on the shore were determined using univariate and multivariate analyses of biodiversity implemented in the program PRIMER 5.0 (Clarke and Warwick 2001). The univariate variables were: a) species richness S (total number of species identified), b) macroalgal abundance N (dry biomass g species<sup>-1</sup> quadrant<sup>-1</sup> in 2500 cm<sup>2</sup>), c) periphyton relative abundance (N% per sample). The univariate (S, N and N %) and multivariate analysis were analyzed with PERMANOVA statistics. Prior to any analyses, the PERMDISP test was performed to assess homogeneity within and between groups (Anderson 2005). The Euclidean distance between pairs of observa-

tions was calculated for the univariate analysis (Claudet et al. 2006). For the multivariate variables, Bray-Curtis dissimilarity was calculated between pairs of observations and the data were transformed to the fourth root. All analyses of PERMANOVA were performed with the FORTRAN program (Anderson 2005). The dietary composition of the different *Nacella* species was evaluated calculating the specific importance of each taxa of periphyton, macroalgae and invertebrates per time (winter months or summer months), locality and species, using the multivariate analysis (SIMPER) (Clarke 1993). In addition, we used an MDS analysis (*non-metric multidimensional scaling*; Kruskal and Wish 1978) using Bray-Curtis similarity matrices in order to compare the assemblages of dietary items among the different species and explore the pattern of spatial ordering.

## Results

## Habitat characterization

A total of 17 microalgae (Suppl. material 1: Table S1) and 63 macroalgae (Suppl. material 1: Table S1) taxa were identified in both localities. Statistical analysis showed that the composition of periphyton changes significantly between intertidal levels, between time and between localities (Suppl. material 1: Table S2, S3). In terms of richness and biomass of macroalgae, the highest values were recorded in the sampling at summer in each localities. The macroalgae assemblages in the middle intertidal zone showed a significant increase of richness and average biomass in summer (Suppl. material 1: Table S4, S5). In addition, the PERMANOVA analysis showed that the composition of macroalgae and periphyton varied significantly between levels (middle and lower), between localities and between the winter and summer months (Suppl. material 1: Table S2, S3, S6, S7).

#### Diet analyses

Throughout the study period, the periphyton assemblages found in the gut contents were composed of 27 taxa, among which were diatoms, cyanophytes and dinoflagellates (Table 1). Among the identified genera, the most common were *Chroococcus*, *Navicula*, *Pinnularia*, *Cocconeis*, *Fragilaria* and *Licmophora*. It should be noted that the dinoflagellates *Dinophysis*, *Protoperidinium* and *Prorocentrum* were registered in the gut contents of *N. deaurata* and *N. magellanica* in summer at Otway Sound. From a spatial point of view, both *Nacella* species showed significant changes in the dietary composition of periphyton between localities in each time (Suppl. material 1: Table S8, S9).

The macroscopic gut contents of both species of *Nacella* were characterized by 39 taxa, including green, brown and red macroalgae, as well as invertebrates such as foraminifera, molluscs and arthropods (Table 1). *Nacella deaurata* contained the highest total richness with 33 taxa, and *N. magellanica* had the highest occurrence frecuency

	P. Hambre				O. Sound				
TAXA	N. deaurata		N. magellanica		N. deaurata		N. magellanica		
	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer	
CYANOBACTERIA									
Chroococcus sp.	+	+	+	+	+	+	+	+	
Oscillatoria sp.	+	+	+	+	+	+	+	+	
BACILLARIOPHYTA	BACILLARIOPHYTA							1	
Melosira sp.	+	+		+	+				
Coscinodiscus sp.	+	+	+	+	+	+	+	+	
Stephanopyxis sp.	+								
Biddulphia sp.		+	+		+	+	+		
Pinnularia sp.	+	+	+	+	+	+	+	+	
Navicula sp.	+	+	+	+	+	+	+	+	
Gyrosigma sp.	+	+	+	+		+		+	
Diploneis sp.	+	+	+	+			+	+	
Diploneis sp2.						+		+	
Plagiotropis sp.	+	+	+	+	+	+	+	+	
Cocconeis sp.	+	+	+	+	+	+	+	+	
Surirella sp.	+	+	+						
Gomphonema sp.			+	+	+				
<i>Cymbella</i> sp.	+	+	+	+	+	+	+	+	
Licmophora sp.	+	+	+	+	+	+	+	+	
Amphora sp.		+	+	+	+	+			
<i>Cylindrotheca</i> sp.		+		+		+		+	
Nitzschia sp.	+		+						
Achnanthes sp.		+	+	+		+	+	+	
Rhabdonema sp.	+	+	+	+		+			
Grammatophora sp.	+	+	+	+	+	+	+	+	
<i>Fragilaria</i> sp.	+	+	+	+		+		+	
MIOZOA									
Dinophysis sp.						+		+	
Protoperidinium sp.						+			
Prorocentrum sp.						+		+	
СНЬООРНУТА									
Spongomorpha pacifica	+	+	+	+	+	+	+	+	
<i>Ulothrix</i> sp.	+		+	+					
Ulva clathrata	+								
Ulva lactuca	+		+				+	+	
Ulva prolifera	+		+						
<i>Ulva</i> sp.	+		+		+			+	
Rhizoclonium sp.	+	+	+						
OCHROPHYTA									
Ectocarpus siliculosus	+	+	+	+				+	
Caepidium antarcticum		+		+					
Adenocystis utricularis		+	+	+		+	+	+	

**Table 1.** Systematic list of items found in the gut contents of *Nacella* species in the winter and summer months, in Puerto del Hambre and Otway Sound, indicating their presence (+).

	P. Hambre				O. Sound			
TAXA	N. deaurata		N. magellanica		N. deaurata		N. magellanica	
	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer
Scytosiphon lomentaria		+		+		+		+
Halopteris funnicularis		+		+	+	+	+	+
Macrocystis pyrifera			+					
RHODOPHYTA								
Acrochaetium sp.	+			+	+	+		+
Nothogenia fastigiata	+	+	+	+	+	+	+	+
Iridaea chordata		+		+				+
Sarcothalia crispata						+		+
Mazzaella laminaroides								+
Grateloupia sp.		+		+				
Ceramium sp.		+			+			
Heterosiphonia sp.				+				
Polysiphonia sp.		+	+		+	+	+	+
Pterosiphonia sp.			+			+		
Ballia callitricha					+		+	
Bostrychia sp.			+		+	+	+	+
Plocamium sp.		+			+			
FORAMINIFERA								
Foraminifera indet	+	+	+	+	+	+	+	
MOLLUSCA								
Margarella violacea	+		+					
Laevilitorina caliginosa	+		+	+				
<i>Eatoniella</i> sp.	+							
Onchidella marginata				+				
Mytilus platensis	+	+	+	+		+	+	+
<i>Lasaea</i> sp.		+		+		+		
ARTHROPODA								
Amphipoda indet		+						
Ostracoda indet		+	+	+	+	+	+	
Notochthamalus				-				
scabrosus				т				
Elminius kingii						+	+	+
Arachnida indet	+	+	+	+				
Halirytus magellanicus	+	+	+	+				+

of invertebrates (57 %). At temporal level, when comparing the dietary composition of the two species within each season separately (e.g., winter 1 × winter 2, winter 2 × winter 3 and summer 1 × summer 2 x, summer 2 × summer 3), there was no significant differences (PERMANOVA, p > 0.05) (Suppl. material 1: Tables S10, S11). However, significant differences (PERMANOVA, p < 0.05) (Suppl. material 1: Table S11) were observed in the dietary composition of both species between the winter and summer months for both species (e.g., winter 1 × summer 1), mainly due to the incorporation of different taxa (e.g., *S. pacifica, S. lomentaria, Bostrychia* sp., *M. platensis*) (Fig. 2) in the gut content during summer. In addition, these species did not present the same gut



**Figure 2.** Percentage contribution SIMPER of items in the gut contents of the *Nacella* species in Puerto del Hambre and Otway Sound for the winter and summer months. The contribution limit was 90% of the total dietary composition. SIMPER analysis shows the dissimilarity between the species of *Nacella* in the two localities (average dissimilarity in bold and on the bar). The contribution limit was 75% of the total dietary composition. M = Macroalgae (with colours) and I = Invertebrates (with grey scale). Structural hardness of the thallus for macroalgae: th = thin filaments, cf = corticated filaments, clf = cylinder-like form and lm= leathery macrophyte. Functional group for invertebrates: s = sessile and m = mobile.



**Figure 3.** Non-metric multidimensional scaling of the dietary composition recorded in the gut contents of the *Nacella* species in Puerto del Hambre (**a**, **c**) and Otway Sound (**b**, **d**). **a**, **b** correspond to the winter months, and **c**, **d** to the summer months. The dashed line indicates the separation between species.

content composition, with significant differences between localities (PERMANOVA, *p* = 0.0001) (Suppl. material 1: Table S11) and mean dissimilarities over 50% (SIMPER). Specifically in the *N. deaurata* diet, the macroalgae *H. funnicularis, Bostrychia* sp., *Rizoclonium* sp., *S. lomentaria, S. pacifica* and *Polysiphonia* sp. contributed significantly to the dissimilarity between the two localities (SIMPER) (Fig. 2). The macroalgae *H. funnicularis, S. lomentaria, Grateloupia* sp., *N. fastigiata* and the invertebrates *M. platensis, E. kingii*, Archnida indet. and *H. magellanicus* contributed significantly to the dissimilarity of the diet of *N. magellanica* between localities (Fig. 2).

Finally, the dietary composition varied significantly between *N. deaurata* and *N. magellanica*, at each localities and each time (See PERMANOVA, p < 0.05, Suppl. material 1: Table S11 and NMDS, Fig. 3), and high dissimilarity averages were observed (> 65 %); SIMPER; Fig. 2). The diet of *N. deaurata* was generally composed of macroalgae taxa such as *H. funnicularis, S. pacifica, Bostrychia* sp., *Rizoclonium* sp.,

*Polysiphonia* sp., *Acrochaetium* sp., *N. fastigiata*, and *Ultothrix* sp. contributed to 90 % of similarity of the group. However, in the diet of *N. magellanica*, we observed a greater number of contributing invertebrates, such as *M. platensis*, *H. magellanicus*, *E. kingii*, Foraminifera indet. and Arachnida indet. (Fig. 2).

## Discussion

This is the first study to perform a temporal and spatial detailed analysis of the diet of intertidal *Nacella* species in the Sub-antarctic Ecoregion of Magellan. In general, both localities had a temporal and spatial variation in the composition of periphyton and macroalgae. In terms of diet, our results showed that in the gut contents of *N. deaurata* and *N. magellanica* we found a great variety of periphyton, macroalgae and some invertebrates. The results also demonstrated a temporal and spatial influence effect in the diet composition of both species. The diet composition between the two species was also different, mainly due to the highest occurrence of invertebrates in the gut content of *N. magellanica* and the highest occurrence of corticated filamentous macroalgae in *N. deaurata*. Here, we discuss how the high temporal and spatial variability of the benthic communities of periphyton and macroalgae affect in the dietary composition of two common grazers that inhabit the Magellan coast.

#### Temporal and spatial variation in micro- and macroalgae assemblages

In general, the average dry biomass of the macroalgae assemblage, per quadrat, at each locality showed a significant increase during the summer months, with the greatest richness and abundance found at the middle intertidal level (Suppl. material 1: Table S4, S5). The multivariate analysis showed differences in the assemblage composition of micro and macroalgae between the two localities at each intertidal level and time. This dynamics of macroalgae assemblages in the channels and fjords of the Magellan Ecoregion was also observed by Ojeda (2013), who observed that the macroalgae assemblages do not present a general spatial structuring pattern, and there may even be differences in the structure of macroalgae assemblages between sites for similar intertidal levels within a same Bay (Ojeda 2013). This author also found that the macroalgae assemblages temporally presented a decrease in wet biomass in winter periods and a considerable increase in summer, showing the marked temporal dynamics of the macroalgae assemblages. These results suggest indetectable general pattern in the vertical zonation of micro- and macroalgae assemblages in these two localities (this study). In the channels and fjords of the Magellan Ecoregion, it has been described that the local environmental heterogeneity likely plays a role in structuring ecological assemblages and communities in Sub-Antarctic marine channels (Ríos et al. 2007, Aldea et al. 2011, Ojeda 2013, Ojeda et al. 2014). Finally, our data suggest an absence of a vertical pattern zonation between the two sites.

## Periphyton in the gut contents of N. deaurata and N. magellanica

In this study, the gut content of both species of *Nacella* presented a great variety of periphyton taxa (27 items), among which the most common were 12 taxa of diatoms and the cyanophyte Chroococcus. The rarest items were the dinoflagellates Alexandrium, Dinophysis and Protoperidinium, whose habits are mainly planktonic (Reguera et al. 2014), suggesting that their consumption was probably accidental. However, dinoflagellates have already been reported in the gastric contents of Patella species (Rubio et al. 2015). In addition, the dinoflagellate Prorocentrum sp. was recorded in summer at the two study locations; but in this case, its consumption cannot be considered accidental, since this dinoflagellate has a benthic habit (Tapia et al. 2002) (Fig. 4). In the present study, Prorocentrum sp. was also recorded in the quadrat samples in both localities. The record of this dinoflagellate is very important, mainly because it is associated with diarrheic toxins (DSP; Tapia et al. 2002), and might impact local people who consumes Nacella species (Ojeda 2013), even if they are not an official fishery in the Magellanic Region. Consequently, further study of Prorocentrum sp. dynamics would be crucial to determine whether this benthic periphyton is commonly found in grazer gastropods, especially Nacella and Fissurella limpets, who are locally consumed.

The abundance and composition of benthic diatoms can vary significantly between different micro-habitats within the rocky shore (Hill and Hawkin 1991, Jenkins et al. 2001). Similar dynamics of the periphyton community have been observed in the present study, since significant differences in the microalgae composition between different intertidal /zonation levels and betwenn, the two localities were observed. Hence, the differences observed for microalgae composition between both *Nacella* species could be associated with its vertical distribution.

Both *Nacella* species presented a shift in dietary composition in microalgae between the winter and summer months. A similar pattern was observed from the quadrats sampling (habitat composition). This shift in dietary composition is related to the temporal dynamics of periphyton communities, mainly due to the incorporation of different microalgae in the diet during summer. In the Northern Hemisphere, the seasonality in the composition of benthic microalgae has already been observed (Attard et al. 2014), and the greatest diversity and abundance were reached during the winter season (Jenkins et al. 2001). Also, studies in high latitudes of the Northern Hemisphere in *Patella vulgata* have shown that this species presents a seasonal change in the dietary composition mainly associated with changes in microalgae composition and abundance during the year (Hill and Hawkin 1991). In this study, the greatest diversity of microalgae was reached during summer; this is probably due to the higher productivity of benthic communities during summer in the Magellan coast (Ojeda 2013).

Analyses of gut contents showed greater richness and relative abundance than those observed in the quadrats. Studies in *P. vulgata* have shown similar results, with a greater variety of diatoms being found in the gut contents (Hill and Hawkin 1991). The authors explained that this is mainly due to these patellogastropods covering greater areas for longer times, and are much more effective when extracting periphyton from rock. The ability



**Figure 4.** Light microscope and stereomicroscope images of microalgae, macroalgae and invertebrates taken from gut contents of *Nacella magellanica*.

of herbivorous molluscs to extract microalgae directly depends on their radular structure (Steneck and Watling 1982, Ocaña and Fa 2003). For example, *Siphonaria* species have a radular structure composed of numerous small non-mineralized teeth, which are better suited for scraping soft foliate macroalgae than benthic diatoms (Ocaña and Fa 2003). On the other hand, the patellogastropod radula is a complex structure with mineralized teeth containing iron and silica, which are able to bore rock and remove microalgae from the substrate (Branch 1981). This feature allows them to highly consume periphyton, such as diatoms, algae spores, detritus and some invertebrates (Branch 1981, Safriel and Erez 1987). From a nutritional point of view, benthic microalgae are very important in the diet of many marine organisms, as they contain a high content of polyunsaturated fatty acids (PUFA) and eicosapentaenoic acid (EPA; Renaud et al. 1999, Vicose et al. 2012). In this sense, *Nacella* species, like other patellogastropods in the world (Branch 1981), present significant consumption of periphyton in the winter and summer seasons. Considering the important reduction of macroalgae cover and biomass during the Magellanic winter (Ojeda 2013, this study), the communities of periphyton constitute an important resource for *Nacella* species.

## Temporal and spatial variation in dietary composition of macroalgae and invertebrates

Variations in the diet of herbivores are generally correlated to food availability (Aguilera 2011). In our study, between winter and summer, both *Nacella* species shifted

December del Hamilian	Macroa	lgae (g)	N. deau	rata (%)	N. magellanica (%)		
Fuerto del flambre	winter	summer	winter	summer	winter	summer	
Thin sheet-like forms	0.64±0.11	1.87±0.51	3±1	8.55±1.9	0.55±0.28	4.55±1.97	
Thin filaments	2.51±1.19	3.95±1.01	11±2.3	21.55±4.6	3.22±1.4	4.55±2.28	
Corticated filaments	1.09±0.3	1.67±0.50	6±2.6	5.11±1.44	2.33±1.3	2.77±2.43	
Cylinder-like forms	1.96±0.65	8±2.36	1±0.3	5.44±1.5	5.88±2.56	9.11±1.57	
Coenocytic forms	0.006±0.0001	0.12±0.12	0	0	0	0	
Cushion-like forms	0.05±0.04	0	0	0	0	0	
Leathery macrophyte	4.34±2.19	4.14±1.70	0	3.33±1.0	0.44±0.30	3.55±1-29	
Otway Sound							
Thin sheet-like forms	2.14±0.66	1.69±0.58	0.22±0.15	0	0.56±0.56	1.22±0.49	
Thin filaments	0.78±0.62	0.42±0.13	8±2	12.56±3.46	1.44±0.73	15.56±3.88	
Corticated filaments	8.99±2.58	2.81±1.04	22.67±3.76	31±4.86	8±2.07	3.33±1.57	
Cylinder-like forms	3.39±1.19	8.59±1.86	1.56±0.84	8.44±2.89	3.33±1.01	7.78±1.85	
Coenocytic forms	0	0.29±0.29	0	0	0	0	
Cushion-like forms	0	0.002±0.002	0	0	0	0	
Leathery macrophyte	3.56±1.23	31.98±7.03	0	1.22±0.52	0	1.78±0.59	

**Table 2.** Average dry biomass (g) of the different functional groups of macroalgae for the winter and summer months in the locality of Puerto del Hambre and Otway Sound and average relative abundance (%) of the different functional groups of macroalgal found in the gut contents of the two *Nacella* species. The values correspond to means ± DS.

their dietary composition. In general, for the winter months in both species and in the two localities, the macroalgae that presented the highest relative abundances in the gut contents corresponded to the filamentous group (thin + corticated) (e.g. S. pacifica, H. funnicularis, Polysiphonia sp.). Similarly, in both locations, the filamentous macroalgae contributed between a 33 (Puerto del Hambre) and a 51 % (Otway Sound) of the total biomass of the assemblages (Table 2). During the summer months, the filamentous macroalgae represented between a 7 (Puerto del Hambre) and a 28 % (Seno Otway) of the total biomass, while the cylinder-like form macroalgae (e.g. A. utricularis, N. fastigiata) increased their abundance, representing between 18 and 40 % of the total biomass of the assemblages. This temporal change in the assemblage was reflected in the dietary composition of both Nacella species, since in both localities, an increased in the relative abundance of cylinder-like form macroalgae in the gut contents were observed (Table 2). Therefore, the temporal variability of the macroalgae abundance was reflected in the diet, thus suggesting a generalist habit for these species. The temporal variation in dietary composition of herbivores also depends strongly on the latitude at which they live. For example, in subtropical rocky coasts, the dietary composition of Megathura crenulata does not vary during the year (Villarreal et al. 2013). In the lower latitudes of the Northern Hemisphere, the gut of Turbo brunneus contains a lot of corallinaceous algae all year without temporal differences (Ramesh and Ravichandran 2008). However, towards higher latitudes, seasonal climatic changes are stronger, and the results differ according to the marked seasonality of micro- and macroalgae assemblages on the coast (Jenkins et al. 2001, Ojeda 2013, this study). Patella vulgata's diet

varies with the composition and abundance of microalgae and filamentous macroalgae (Hill and Hawkin 1991). Seasonal variations in the diet of Chilean herbivorous molluscs have also been described, thus implying a change in food availability between the seasons (Aguilera 2011).

Generalist grazers use an opportunistic strategy in their feeding habits, consuming the most common resources available (Camus et al. 2012). Therefore, the significant differences observed in the dietary composition of *N. magellanica* and *N. deaurata* between the localities of Puerto del Hambre and Otway Sound reflect differences in macroalgae assemblages between sites, and highlight the generalist nature of these species. Similarly, differences in the diet of *M. crenulata*, along different rocky habitats of the west coast of the Baja California Peninsula (Villarreal et al. 2013) probably reflect changes in the benthic community among the localities. In the Magellanic Province, our results suggest that the marked environmental heterogeneity that leads to strong spatial variations in richness, abundance and structure of the benthic communities, plays a key role in the dietary variation of *Nacella* species (Ríos et al. 2007, Mansilla et al. 2013, Ojeda et al. 2014, this study).

Steneck and Watling (1982) classified herbivores molluscs according to their type of radula, and commented that the docoglossa radula of patellogastropods mainly excavates rock and extracts microalgae and filamentous algae, but does not consume leathery macrophytes. However, Raffaelli (1985) found that this pattern of classifying herbivores according to the type of radula may vary according to species characteristics and food availability. Among algal functional groups, the macroalgae with thin and corticated filaments as well as cylinder-like forms were preferentially consumed by *N. magellanica* and *N. deaurata*. Indeed, *N. deaurata* at both localities tends to consume more thin and corticated filamentous macroalga, as is the case in Otway Sound, despite the high abundance of leathery macrophyte species such as *Sarcothalia crispata*. Therefore, this greater tendency to consume more of a certain functional group of macroalgae could be related to the physiological capacity of the radula of these species (Steneck and Watling 1982).

#### Interspecific variations in dietary composition

The diet composition in macroscopic items of both *Nacella* species was varied with a great variety of taxa (37 taxa). Among the most common items ingested by *N. deaurata* were nine species of macroalgae and some invertebrates such as *Laevilitorina caliginosa*, Ostracoda indet and Foraminifera indet, while *N. magellanica* consumed seven species of macroalgae and a large variety of invertebrates, including juveniles of *M. platensis*, larvae of the chironomids *Halirytus magellanicus*, Archnida indet, Ostracoda indet and Foraminifera indet. This is the first report of a nacellid species consuming a chironomid (Fig. 4).

Although *N. magellanica* and *N. deaurata* cohabit the intertidal zone, they differ markedly in their dietary composition, mainly because *N. deaurata* consumes a



**Figure 5.** Light microscope and stereomicroscope images of microalgae, macroalgae and invertebrates taken from gut contents of *Nacella deaurata*.

higher diversity of macroalgae (Fig. 5), whereas N. magellanica feed principally on invertebrates and macroalgae. Andrade and Brey (2014) reported similar results from the diets of 10 individuals of the same two species in Laredo Bay (Strait of Magellan). Their results, analyzing gut contents and stable isotopes, showed that N. deaurata behaves more herbivorous-like by consuming brown and red macroalgae, while N. magellanica consumes more invertebrates, and consequently is more omnivorous-like (by consuming green microalgae, micro-bivalves and foraminifera) (Andrade and Brey 2014). Therefore, this could suggest that the consumption of invertebrates by N. magellanica is not necessarily accidental. Recently in the Strait of Magellan, stable isotope analyses indicated that species of green and brown algae are preferentially consumed by macroherbivores, because they have higher nutritional value than red algae (Andrade et al. 2016). However, the consumption of some macroalgae species by generalist grazers such as Nacella species would more likely be linked to the algal availability in the habitat than to their nutritional profiles (Chapman and Underwood 1992). In this sense, it was observed that the Nacella species usually feed on common and abundant macroalgae such as the filamentous group (thin + corticated), which are fast growing and are available throughout the year (Ojeda 2013, this study) (Table 2), while other abundant groups of macroalgae, such as leathery macrophyte (e.g., Sarcothalia crispata or Iridaea cordata) did not show a high abundance in the gut contents (Table 2). In addition, recent nutritional studies in macroalgae from the Magellanic province (Astorga and Mansilla 2014) show that red algae such as Pyropial Porphyra and Callophylis have higher energy and protein values than brown algae such as Macrocystis pyrifera and

*Durvillaea antarctica*. In this study, two *Porphyra/Pyropia* morphotypes were reported for both localities, however no tissues were found in the gut contents of the *Nacella* species. Therefore, our results suggest that the consumption of groups of macroalgae species could be mainly related to the availability in the habitat and the structural hardness of the thallus. This has been observed for other marine gastropods. For instance, the specie *Turbo sarmaticus* mainly consumes *Ulva rigida* and *Gelidium pristoides* and, although *G. pristoides* presents higher nutritional values, it ingests mainly *U. rigida* because it has greater availability in the habitat than *G. pristoides* (Foster et al. 1999).

The differences observed in the dietary composition of N. deaurata and N. magellanica can also be explained by their vertical distribution in the coastal zone. For example, in the localities of Otway Sound and Puerto del Hambre, N. magellanica presented greater abundance in the middle zone compared to N. deaurata, while N. deaurata presented its highest abundances in the low intertidal zone and in the shallow subtidal (1 meter deep) (Rosenfeld 2016). In this sense, in Otway Sound, N. magellanica mainly consumes juveniles of M. platensis, which populations in the Magellanic Region settle commonly in the middle intertidal zone (Langley et al. 1980, Ojeda et al. 2014). In addition, we observed a marked difference in macroalgal assemblages between the vertical levels at both locations; indeed, the vertical zonation is crucial in the composition, richness, and biomass of macroalgae (Underwood 1980, Ojeda et al. 2014). Therefore, the vertical distribution of herbivores along the rocky shore can be determinant for their diet (Santina et al. 1993, Rubio et al. 2015). Indeed, a study on the dietary composition of the genus Patella showed that P. rustica, which occurs in the upper intertidal zone, only feeds on some species of algae, whereas P. aspera feeds on all types of algae (Santina et al. 1993). In Vancouver, the dietary composition of the species Littorina sitkana and L. scutulata, differs drastically mainly because L. sitkana is very rare in the upper intertidal zone, has low resistance to desiccation and a low capacity to collect food (Voltolina and Sacchi 1990). Finally, many of the differences in dietary composition between congeneric species depend on their zonal distribution on the shore. Consequently, the observed differences between N. magellanica and N. deaurata may be due to their vertical distribution, with N. magellanica being a more common inhabitant of the middle intertidal zone (Rosenfeld 2016).

In this study, how the variability in the composition of micro- and macroalgae among intertidal levels (middle and low) and between localities and time plays a fundamental role in the dietary composition of *N. magellanica* and *N. deaurata* was studied. This work is the first in the Magellanic Region to address the temporal and spatial characteristics of the diet of *Nacella* intertidal species, which provides a more complete listing of periphyton, macroalgae and invertebrates present in the diet of these Sub-Antarctic patellogastropods. Finally, it is important to mention that in this study only gut contents were analyzed. Therefore future research using new techniques such as stable isotopes could give a better resolution on the dietary composition that is effectively assimilated by *Nacella* species.

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This work was conducted using a local species of invertebrates "mauchos" (*Nacella magellanica* and *Nacella deaurata*) as study model, a common limpet species from the southern tip of South America. All applicable international, national, and/or institutional guidelines for the care and use of animals were followed. The Instituto de Ecología y Biodiversidad (IEB/15 2015) and Chilean Fishery Service (SERNAPESCA 429/2015) ethic committees approved sampling protocols and experiments. For this, we complied with local legislation and the Convention on Biological Diversity. The species is not protected by the Chilean Fishery Subsecretary and has not been included in the Chilean fishery statistics. Permission to undertake field studies and to collect specimens was issued by the Chilean Fishery Service Director (Carlos Orellana Céspedes), under the technical memorandum (249/2015).

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## Tables S1–S11

Authors: Sebastián Rosenfeld, Johanna Marambio, Jaime Ojeda, Juan Pablo Rodríguez, Claudio González-Wevar, Karin Gerard, Tamara Contador, Gemita Pizarro, Andrés Mansilla

Data type: Occurence of all algae taxa recorded, and Permutation analysis (PER-MANOVA) of the composition of algae and gut content of *Nacella* species.

- Explanation note: This is a DOC file with all the temporal information of the occurrence of the algae taxa in both localities, and all the information of the PER-MANOVA analyzes used in this study.
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RESEARCH ARTICLE



# A new species of trogloplacine crab of the genus Australocarcinus Davie, 1988 from a freshwater stream in Mahé, Seychelles (Crustacea, Brachyura, Chasmocarcinidae)

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## Abstract

A new species of freshwater chasmocarcinid crab, *Australocarcinus insperatus* **sp. n.**, is described from the Seychelles Islands in the Indian Ocean. This is the first record of the genus and the subfamily Trogloplacinae Guinot, 1986, from the Indian Ocean, with all other members previously recorded from Australia, New Britain, New Caledonia, and Palau in the Pacific Ocean. The disjunct distribution of *Australocarcinus* is unexpected considering all trogoplacines are believed to practice direct development, lacking freeswimming larval stages. The new species is morphologically most similar to *A. riparius* Davie, 1988, from Queensland, Australia, but can be distinguished from its three congeners on the basis of the structures of its carapace, ambulatory legs and male first gonopod.

## Keywords

Chasmocarcinidae, freshwater, Indian Ocean, new species, Trogloplacinae, taxonomy

## Introduction

The Chasmocarcinidae Serène, 1964, is a predominantly marine family, with most of the species occurring in subtidal habitats, and some reaching depths of over 2000 metres (Ng and Castro 2016). One subfamily, the Trogloplacinae Guinot, 1986, however, occurs exclusively in freshwater habitats, sometimes several kilometres from the sea as well as in aquatic inland limestone caves (Davie and Guinot 1996; Ng and Castro 2016). Trogloplacines are also unusual in practising direct development, lacking planktotrophic larvae (Davie and Guinot 1996). Only two genera of Trogloplacinae are known, the monotypic *Trogloplax* Guinot, 1986 (which lives in caves in New Britain, southwestern Pacific), and *Australocarcinus* Davie, 1988 (with three epigeal species from northeastern Australia, New Caledonia and Palau). The first *Australocarcinus* species is reported here from the Indian Ocean, *A. insperatus* sp. n., from southern Mahé in the Seychelles archipelago.

## Materials and methods

Material examined is deposited in the Zoological Reference Collection (**ZRC**) of the Lee Kong Chian Natural History Museum, National University of Singapore. Measurements provided (in millimetres) are of the carapace width and length, respectively. The terminology used follows that in Ng and Castro (2016) and Davie et al. (2015). The following abbreviations are used: **G1** male first pleopod; **G2** male second pleopod.

### **Systematics**

Family Chasmocarcinidae Serène, 1964 Subfamily Trogloplacinae Guinot, 1986

Genus Australocarcinus Davie, 1988

Type species. Australocarcinus riparius Davie, 1988, by original designation.

*Australocarcinus insperatus* sp. n. http://zoobank.org/909E98B1-A957-44F0-BD2E-2C665E67CFB1 Figs 1–3

**Material examined.** Holotype: male  $(10.7 \times 8.6 \text{ mm})$  (ZRC 2017.1072), in shallow stream, ca. 800 m from sea, about 2 km south-southeast of international airport, 4°41'32.42"S, 55°31'2.90"E, Mahé, Seychelles, coll. SR Daniels, May 2010. Paratypes: 1 male (8.5 × 7.2 mm), 1 female (9.5 × 7.8 mm) (ZRC 2017.1073), same data as holotype.

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Diagnosis. Carapace subquadrate, front weakly bilobed, with shallow median concavity (Fig. 1A, B); dorsal surface gently convex (Fig. 1F); dorsal surfaces and margins covered with short uneven tomentum (Fig. 1A, B); anterolateral margins arcuate, with four low teeth: first widest with gently sinuous margin, second lobiform, third wide, fourth (at junction of antero- and posterolateral margins) dentate, directed laterally, protruding beyond margin (Fig. 1B). Posterolateral margin converging towards gently convex posterior carapace margin (Fig. 1B). Epistome compressed, posterior margin with distinct triangular median lobe with median fissure, lateral margins gently sinuous (Fig. 1G). Eye peduncle completely filling orbit, relatively short, mobile; cornea distinct, pigmented (Fig. 1B, F). Third maxillipeds leaving gap when closed; merus quadrate, anteroexternal angle auriculiform; ischium quadrate, slightly longer than merus with very shallow median sulcus (Fig. 1C, D). Chelipeds subequal, relatively stouter in males (Figs 1A, 2E); cutting margins of both chelae with distinct teeth in both sexes, base of fingers with tuft of stiff setae; proximal part of dactylus of right chela with large, triangular tooth directed towards palm (Fig. 2A); ventral surface of cheliped merus with tubercles. Ambulatory legs moderately short; meri unarmed but setose to varying degrees; P2 carpus, propodus and dactylus with very long coarse setae which obscures margins (Figs 1A, 2B); P3–P5 propodus and dactylus setose but setae shorter than on P5 (Fig. 2C); P5 dactylus straight (Fig. 2C). Thoracic sternites 1, 2 fused, broadly triangular, short; separated from sternite 3 by sinuous groove; sternites 3, 4 fused, relatively broad (Fig. 1D). Male pleon with lateral margins of somite 6 and fused somites 3-5 gently sinuous; telson slightly longer than broad (Fig. 1D, E). Sterno-pleonal cavity of male deep, press-button for pleonal holding small, short tubercle posterior to thoracic sternal suture 4/5 near edge of sterno-pleonal cavity. Male thoracic sternite 8 short, rectangular; supplementary plate narrow, wider along outer part (Figs 1E, 2D). G1 stout; basal part truncate; distal part cylindrical, with rounded tip, covered with short spinules (Fig. 3A-D). G2 prominently longer than G1, basal segment curved; distal segment slightly longer than basal segment, apex cup-like (Fig. 3E, F). Somites of female pleon with slightly convex lateral margins; telson wider than long (Fig. 2F). Sterno-pleonal cavity of female moderately deep, with large vulvae distinctly separated from each other, covering most of thoracic sternite 5, ovate, with low raised lip on outer margin, opening slit-like (Fig. 2G).

**Etymology.** From the Latin "*insperatus*" for "unforeseen", alluding to the unexpected discovery of a species of *Australocarcinus* in the western Indian Ocean.

**Remarks.** Davie (1988) originally established *Australocarcinus* for one freshwater species from northern Queensland in Australia, *A. riparius* Davie, 1988. Davie & Guinot (1996) subsequently described two more species, *A. kanaka* Davie & Guinot, 1996, and *A. palauensis* Davie & Guinot, 1996, from New Caledonia and Palau, respectively. Davie & Guinot (1996) showed that *Australocarcinus* was in the same subfamily as the more apomorphic cavernicolous species *Trogloplax joliveti* Guinot, 1986, from New Britain; that it belonged to the family Chasmocarcinidae; and provided evidence that their larval development was truncated with the eggs hatching directly into juvenile crabs or megalopas.

Australocarcinus insperatus sp. n., is morphologically most similar to A. riparius Davie, 1988, in the anterolateral margin possessing four low teeth, the anteroexternal angle of



**Figure 1.** *Australocarcinus insperatus* sp. n., holotype male (10.7 × 8.6 mm) (ZRC 2017.1072), Seychelles. **A** overall dorsal habitus **B** dorsal view of carapace (right side denuded) **C** right third maxilliped (denuded) **D** anterior thoracic sternum and pleon **E** posterior thoracic sternum and pleon **F** frontal view of cephalothorax **G** posterior margin of epistome.

the merus of the third maxilliped is clearly auriculiform and the male telson is relatively longer. *Australocarcinus insperatus* sp. n., however, can easily be separated by possessing a more sub-hexagonal carapace (Fig. 1B) (vs. carapace more subquadrate in *A. riparius*, Fig. 4B); a distinctly convergent posterolateral margin (Fig. 1B) (vs. posterolateral margins subparallel in *A. riparius*, Fig. 4B); the last anterolateral tooth is triangular and protrudes laterally beyond the carapace margin (Fig. 1B) (vs. last tooth truncate and not extending



**Figure 2.** *Australocarcinus insperatus* sp. n. **A–D** holotype male (10.7 × 8.6 mm) (ZRC 2017.1072), Seychelles **E–G** paratype female (9.5 × 7.8 mm) (ZRC 2017.1073), Seychelles. **A** outer surfaces of chelae **B** right first ambulatory leg showing setose posterior margin on propodus and dactylus **C** left fourth ambulatory leg **D** posterior thoracic sternum showing supplementary plate **E** female overall dorsal habitus **F** female posterior thoracic sternum and pleon **G** female sterno-pleonal cavity showing vulvae.

beyond carapace margin in *A. riparius*, Fig. 4B); the ischium of third maxilliped is wider than long (Fig. 1C) (vs. ischium longer than wide in *A. riparius*, Fig. 4C); the ambulatory merus is more elongate and slender (Figs 1A, 2B, C, E) (vs. meri proportionately shorter



**Figure 3.** *Australocarcinus insperatus* sp. n., left G1 and G2; holotype male (10.7 × 8.6 mm) (ZRC 2017.1072), Seychelles. **A** ventral view **B** ventral view **C** distal part (ventral view) **D** distal part (dorsal view) **E** ventral view **F** area between basal and distal segments. Scale bars 0.50 mm (**A**, **B**, **E**); 0.25 mm (**C**, **D**, **F**).

in *A. riparius*, Fig. 4A); and the G1 is relatively more slender (Fig. 3A–D) (vs. G1 stouter in *A. riparius*, cf. Ng and Castro 2016: fig. 98A).

All three specimens of *A. insperatus* sp. n. have a distinct cutting or peeling tooth at the base of the dactylus of the right chela (Fig. 2A), a character which Ng and Tan (1984, 1985) have suggested is used to specially feed on gastropod snails. As most gastropod snails have dextral coiling (opening on the right side when viewed frontally), Ng and Tan (1984, 1985) observed that crabs with the enlarged basal dactylar tooth always have this structure on the right chela to make peeling of the shell more efficient. The other three species of *Australocarcinus* also have this tooth on the right chela (see Ng and Castro 2016: figs 95A, C, E) and on both sexes. This suggests that one of the main food items of *Australocarcinus* are freshwater gastropods.

The discovery of *A. insperatus* sp. n. is surprising as all the members of the Trogloplacinae have been previously found in Australasian and Palau waters. Davie (1988) found juvenile crabs under the pleon of a female *A. riparius*, with ovigerous specimens possessing some 70 large eggs. Davie and Guinot (1996) found megalopa under a female pleon of *A. kanaka*, suggesting that the development was direct, like those in primary freshwater crabs like Potamidae, Potamonautidae and Gecarcinucidae (and some Sesarmidae). All trogloplacines also have large vulvae (Ng and Castro 2016: figs 99B, D, F, H), suggesting the eggs of the other two species, *A. palauensis* and *Trogloplax joliveti* also have large eggs and do not have free-swimming larvae. The vulvae of *A. insperatus* sp. n. are also large (Fig. 2G). If all trogloplacines have abbreviated (or at



**Figure 4.** *Australocarcinus riparius*, male (8.8  $\times$  10.2 mm) (ZRC 2006.167), Australia. **A** overall dorsal habitus **B** dorsal view of carapace (right side denuded) **C** right third maxilliped (denuded) **D** posterior thoracic sternum and pleon **E** anterior thoracic sternum and pleon **F** posterior thoracic sternum showing supplementary plate.

least a semi-abbreviated) development and there are no free-swimming larvae, how did they disperse so widely? Despite hypotheses that primary freshwater crabs may have dispersed through Gondwanic connections (Ng et al. 1995), the available evidence is that they are not old enough to have done so (see Daniels et al. 2006; Cumberlidge et al. 2008; Klaus et al. 2009; Cumberlidge and Ng 2009; Daniels 2011; Cumberlidge and Daniels 2014); and as such, the disjunct distribution of *A. insperatus* sp. n. begs further studies. A complete molecular phylogeny of the Chasmocarcinidae is now being undertaken by L. M. Tsang (Chinese University of Hong Kong) and the results should throw some light on this matter in the future.

**Biology.** The freshwater stream where the specimens were collected was shallow, the water flowing over a sandy bottom, with scattered rocks and construction rubble from past development works in the area. The crabs attempted to bury into the soft sand when disturbed.

#### Amended key to species of Australocarcinus

1	Anterolateral margin entire, without visible lobes or teeth [Palau] A. palauensis
_	Anterolateral margin distinctly dentate
2	Anterolateral margin with 2 low, blunt lobes; anterolateral margin of merus
	of third maxilliped rounded, not auriculiform; male telson relatively short
	[New Caledonia]
_	Carapace anterolateral margin with 4 prominent but low teeth; anterolateral
	margin of merus of third maxilliped expanded, auriculiform; male telson rela-
	tively long
3	Carapace subquadrate, posterolateral margin subparallel; last anterolateral
	tooth truncate, not protruding laterally beyond carapace margin; ischium
	of third maxilliped longer than wide; ambulatory merus relatively shorter,
	stouter; G1 stout [Queensland, Australia] A. riparius
_	Carapace subhexagonal, posterolateral margin gently converging; last antero-
	lateral tooth triangular, distinctly protruding laterally beyond carapace mar-
	gin; ischium of third maxilliped wider than long; ambulatory merus promi-
	nently elongate, slender; G1 relatively more slender [Seychelles]
	A. insperatus sp. n.

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RESEARCH ARTICLE



# Two new species of the genus Aphis (Hemiptera, Aphididae) from Chile on host species of Alstroemeriaceae and Ericaceae

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# Abstract

Specimens from Southern Chile belonging to two undescribed aphid species held at the Natural History Museum in London have been studied. *Aphis luzuriagae* **sp. n.** collected on *Luzuriaga radicans* and on other unidentified *Luzuriaga* species (Alstroemeriaceae) is described from apterous and alate viviparous females. *Aphis gaultheriae* **sp. n.** collected on *Gaultheria mucronata* (Ericaceae) is described from apterous viviparous females. Taxonomic peculiarities of both species are discussed.

# Keywords

Aphididae, aphids, Chile, Gaultheria, Luzuriaga, New species

# Introduction

Specimens belonging to several aphid samples collected by D. Hille Ris Lambers during field work in Chile in 1974 on the very injurious attacks on cereal crops by *Schizaphis graminum* (Rondani, 1852), are preserved in the aphid collection of the Natural History Museum in London. Blackman and Eastop (2017) include in the keys of aphids

living on *Luzuriaga* (Alstroemeriaceae) and on *Gaultheria* (Ericaceae), two unnamed species of *Aphis* collected by Hille Ris Lambers during this field work. In this paper, these two new species are described as *Aphis luzuriagae* sp. n. and *Aphis gaultheriae* sp. n.

#### Material and methods

Studied specimens are mounted on microscopic slides using Canada balsam. Collection data for the studied specimens of the new species are included in the Types section for each new species. For comparison, specimens of other species recorded in South America preserved in the collection of the University of Leon have also been studied. Papers describing these species, and in particular the comparative table of features of South American *Aphis* species presented by López Ciruelos et al. (2017), have been consulted. Measurements of specimens were taken as detailed by Nieto Nafría and Mier Durante (1998). The descriptions of *Aphis vurilocensis* Nieto Nafría, Brown & López Ciruelos, 2017 and of *Aphis cuyana* López Ciruelos & Ortego, 2017 (González Rodríguez et al. 2017; López Ciruelos et al. 2017) have been used as models for the descriptions of the new species. Data on aphid host plants and distribution of plant species have been respectively taken from Blackman and Eastop (2017) and WCSP (2017). Photographs have been taken using an Olympus set: BX61 microscope, DP70 digital camera and DP Manager Software version 2.1.1.163.

# Taxonomy

#### *Aphis (Aphis) luzuriagae* sp. n.

http://zoobank.org/C1F8F288-747A-404A-83F7-E9546F33D568

**Types.** Holotype, apterous viviparous female (measured specimen number 5, mounted with two nymphs): CHILE, Region VII Bio Bio, province Nuble, East of Chillan "fundo Borges", 7 November 1974, on *Luzuriaga radicans*, Hille Ris Lambers *leg.* [867], Natural History Museum collection (BMNH(E) 1984-340). Paratypes: 27 apterous viviparous females [apt] and 5 alate viviparous females [al] mounted on 20 slides, Natural History Museum collection (BMNH(E) 1984-340); same data as the holotype (4 apt); same locality and date and sample number as the holotype, on *Luzuriaga* sp. (1 apt & 1 al); same locality, date and sample number, on *Luzuriaga* sp. [*Luzuriaga falcata* on the label, a name that is unknown to us] (6 apt & 4 al); CHILE, Region IX La Araucania, province Cautin, Temuco "cerro Nielol", 18 November 1974, on *Luzuriaga* sp., Hille Ris Lambers *leg.* [908] (1 apt); CHILE, Region X Los Lagos, province Osorno, Puyehue National Park, 21 November 1974, on *Luzuriaga radicans*, Hille Ris Lambers *leg.* [910] (2 apt); CHILE, Region X Los Lagos, province Chiloe, Huillinco lake, 23 November 1974, on *Luzuriaga* sp., Hille Ris Lambers *leg.* [918] (3 apt) and Tollenaar *leg.* [Hille Ris Lambers 919] (10 apt.).

**Apterous viviparous females.** (Figs 1, 2), from 28 specimens, of which 25 have been measured. Colour when alive unknown. When prepared pale in general, with head, in-



Figure 1. Apterous viviparous females, habitus (in part). Left, *Aphis alstroemeriae*; right, *Aphis luzu-riagae* sp. n., paratype.

cluding clypeus, mandibular and maxillary lames, antennae, rostrum, legs, dorso-thoracic sclerites, spiracular sclerites, genital and anal plates, and cauda tenuously brown pigmented, and siphunculi brown to dark brown. Body 1.37-1.95 mm long, 1.76-2.52 times hind tibia (0.68-1.03 mm) and 4.09-6.29 times siphunculus. Head with a marked ventral protuberance, which is long-elliptical, rugous and carries two setae. Antenna 1.13-1.61 mm and 0.66-0.92 times body length. Antennal segment III 0.25-0.39 mm and 0.68–0.96 times segment VI processus terminalis, dorsally near smooth, and with 6–12 blunt pale setae, 12–23 µm and 0.4–0.8 times subarticular diameter of antennal segment III [D], shorter than those on vertex (22–30  $\mu$ m and 0.8–1.1 times D), which are similar in shape. Antennal segments IV and V respectively 0.16-0.28 and 0.15-0.22 mm. Antennal segment VI processus terminalis 0.32-0.45 mm and 2.67-3.82 times base of segment (0.10–0.13 mm). Rostrum reaching to or slightly beyond the hind coxae; ultimate rostral segment 0.11–0.13 mm, 0.96–1.20 times base of antennal segment VI, 1.00–1.33 times second segment of hind tarsi, relatively broad with convex sides and with 2 long and fine accessory setae. Dorsum of thorax usually with wide but lightly pigmented marginal sclerites. Prothoracic marginal tubercles low and relatively wide, nearly similar in volume to triommatidium. Inside setae on hind trochanter  $32-53 \ \mu m$  and 0.5-1.1 times trochantrofemoral suture; dorsal setae on hind femora  $15-25 \ \mu m$  and 0.5-1.0 times D. Tarsi and apex of tibiae darker than the rest of legs, except the femoro-tibial joint which is nearly black. Apical brown portion of tibiae larger than rest of segment. Tarsi very short, first segments with (2)3.3.3(2) setae. Marginal tubercles on abdominal segments 1 and 7 small, their diameter is shorter than length of closest marginal setae, and sometimes lack-



**Figure 2.** *Aphis luzuriagae* sp. n., apterous viviparous female. **A** end of rostrum **B** end of tibia and tarsus of hind leg **C** siphunculus **D** cauda and anal plate.

ing on some segments and/or sides. Intermediate abdominal segments without marginal tubercles. Dorsum of abdomen without sclerites other than spiracular ones, which are small and brown. Dorsal setae on thorax and abdomen also blunt or slightly pointed; marginal setae on intermediate abdominal segments  $17-25 \mu m$  and 0.6-1.0 times *D*. Abdominal segment 8 with two setae,  $27-35 \mu m$  and 0.8-1.6 times *D*. Siphunculi tapering in general, with apical portion slightly enlarged, poorly ornamented and without flange, 0.26-0.42 mm, 1.93-2.59 times cauda, and (4.33)5.09-6.67(7.10) times its diameter at mid length. Cauda robust, nearly triangular, the proximal part being straight-sided without or with a very small constriction, sometimes more pigmented towards apex, 0.12-0.17 mm and 1.04-1.35 times its basal width, with 4-7 long, delicate and curved setae.

Alate viviparous females. From 5 specimens, all of them measured. Very similar to apterous viviparous females but with darker head, antennae and legs, antennal segment III imbricate-spinulose and brown thorax; two specimens lack dorsal abdominal pigmented sclerites, one specimen has lightly pigmented and spinulose abdominal sclerites: small marginal on segment 1, wide marginal on segments 2-6, and broad transverse on each of segments 7 and 8, and two other specimens exhibit intermediate sclerotization and pigmentation. Secondary sensoria on antennal segments III and IV; 6-10 on III extended along segment and 1-4 on IV. Body 1.50-1.80 mm long, 1.76-1.77 times hind tibia (0.85-1.00 mm) and 5.17-6.04 times siphunculus. Antenna 1.41-1.72 mm and 0.88-1.00 times body length. Antennal segment III 0.31-0.42 mm and 0.71-0.88 times segment VI processus terminalis. Setae on antennal segment III 15-18 µm and 0.5-0.8 times D. Setae on vertex 17-23 µm and 0.6-1.0 times D. Antennal segment IV 0.22-0.29 mm. Antennal segment V 0.18-0.25 mm. Antennal segment VI processus terminalis 0.43-0.47 mm and 3.58-4.33 times segment VI base (0.10-0.13 mm). Ultimate rostral segment 0.11-0.12 mm, 0.92 -1.14 times base of antennal segment VI and 1.15-1.20 times second segment of hind tarsi. Inside setae on hind trochanter 37-40 µm and approximately 0.8 times trochantrofemoral suture. Dorsal setae on hind femora 17-23 µm and 0.6-1.0 times D. Marginal setae on intermediate abdominal segments 20-23 µm and 0.7-1.0 times D. Setae on abdominal segment 8, 27-30 µm and 1.0-1.3 times D. Siphunculus 0.26 -0.30 mm, 2.12-2.41 times cauda, and 4.83-6.78 times its diameter at mid length. Cauda 0.11-0.13 mm and 0.96-1.14 times its basal width, with 4-7 setae.

Sexuales. (Oviparous females and males). Unknown.

Host plant. All collected specimens of *A. luzuriagae* sp. n. have been found on plants of several species of *Luzuriaga* (Alstroemeriaceae). *Luzuriaga* species are distributed in the South of Argentina and in Central and Southern Chile.

**Etymology.** The specific name "*luzuriagae*" is the plant host genus name of the new aphid species, in genitive.

**Taxonomic discussion.** Only two other genera of this plant family have been recorded hosting aphids: *Alstroemeria* and *Bomarea*, but only one species belonging to *Aphis* has been recorded: *A. alstroemeriae* Essig, 1954, on *Alstroemeria* sp. in Chile (Nieto Nafría et al. 2016). The differences between the apterae of this species and *Aphis luzuriagae* sp. n. are very conspicuous and evident (Fig. 1). The new species can be distinguished from other South American *Aphis* species by the combination of the following features: 1) in apterous viviparous females: the absence of dorsal abdominal pigmented sclerotization, even intersegmental, anteroventral cephalic protuberance, very short tarsi, small or absent marginal tubercles on abdominal segments 1 and 7, lack of marginal tubercles on intermediate abdominal segments, and relatively long processus terminalis of VI antennal segment; 2) in alate viviparous females: secondary sensoria extend over all of antennal segment III and present on antennal segment IV, and dorsal abdominal sclerotization (marginal sclerotization 1–6 and transverse bands 7–8) absent, more or less insinuated or conspicuously present.

#### Aphis (Aphis) gaultheriae sp. n.

http://zoobank.org/BBC6836C-6829-48D8-9EFA-BD69CA76C834

**Types.** Holotype apterous viviparous female (measured specimen number 1), CHILE, Region X Los Lagos, province Chiloe, Lake Huillinco, on *Gaultheria mucronata* (L. fil.) E. J. Remy [*Pernettya mucronata* on the label], 24 November 1974, Hille Ris Lambers *leg.* [928], mounted with two nymphs, Natural History Museum collection (BMNH(E) 1984-340). Paratypes: 2 apterous viviparous females collected with the holotype, mounted in two slides with nymphs, Natural History Museum collection (BMNH(E) 1984-340).

Apterous viviparous females. (Figs 3, 4), from 3 specimens, all of them measured. Colour when alive unknown. When prepared, head, including clypeus, mandibular and maxillary lames brown; antennal segments I and II dark brown like segment VI; rostrum brown, coxae, trochanters, and most of femora dark brown (like antennal segment I), tibiae mainly very pale brown (like antennal segments III-V); dorsal thoracic and abdominal sclerites and genital plate brown (lighter than head), siphunculi, anal plate and cauda dark brown. Body 1.85–2.08 mm long, 1.88–2.24 times hind tibia (0.83-1.05 mm) and 5.49-6.27 times siphunculus. Antenna 1.25-1.56 mm and 0.68–0.79 times body length. Antennal segment III 0.35–0.43 mm and 1.32-1.42 times segment VI processus terminalis, dorsally almost smooth, and with 8-10 thick, pointed and very pale setae,  $15-20 \mu m$  and 0.7-1.0 times subarticular diameter of antennal segment III [D], shorter than those on vertex (27–30  $\mu$ m and 1.2–1.5 times D). Antennal segments IV and V respectively 0.18–0.25 and 0.20–0.26 mm. Segment VI processus terminalis 0.25-0.33 mm and 2.13-2.50 times segment VI base (0.12-0.14 mm). Rostrum slightly extending beyond the mid-coxae; ultimate rostral segment long (0.10–0.12 mm, 0.74–0.96 times base of antennal segment VI, 0.80-1.00 times second segment of hind tarsi), straight and with 2 very long and fine accessory setae. Dorsum of thorax with wide marginal sclerites and several pleural and spinal spots, paler than intersegmental sclerites and metathorax/first abdominal segment. Prothoracic marginal tubercles erect, protuberant and smaller than triommatidium. Inside setae on hind trochanter 37-40 µm and 0.67-0.73 times trochantrofemoral suture; dorsal setae on hind femora 25  $\mu$ m and 1.1–1.3 times D. First segment of tarsi with 3 setae. Protuberant marginal tubercles on abdominal segments 1 and 7 delicate, but only those on segment 1 are especially delicate. Intermediate abdominal segments without marginal tubercles. Intersegmental sclerites darker than segmental and spiracular sclerites. Abdominal segments 1-4 with more or less abundant polygonal spinal sclerites that can be coalescent, and usually without marginal sclerites (polygonal cells are present in one specimen); abdominal segments 5 and 6 each with wide transverse spinopleural band, and abdominal segments 7 and 8 with individual narrow transverse stripes; this variability is similar to that presented by A. tehuelchis Nieto Nafría & López Ciruelos, 2016 (López Ciruelos et al., 2016). Dorsal setae on thorax and abdomen also thick and pointed; marginal setae on intermediate



Figure 3. Aphis gaultheriae sp. n., apterous viviparous females; holotype, specimen on the left.

abdominal segments 17–25  $\mu$ m and 0.9–1.3 times *D*. Abdominal segment 8 with 2 setae, 20–22  $\mu$ m and 0.9–1.1 times *D*. Siphunculi tapering on proximal 1/2–2/3 and distally cylindrical, with small flange and ornamentation of spinuled scales, 0.30–0.36 mm, 1.11–1.38 times cauda, and 6.00–6.56 times its diameter at mid length. Cauda robust finger-shaped, 0.26–0.27 mm and 1.93–2.30 times its basal width, with 10–11 long, delicate and curved setae.

Alate viviparous females and sexuales. (Oviparous females and males). Unknown.

**Host plant.** The type specimens of *Aphis gaultheriae* sp. n. were collected on *Gaultheria mucronata* (Ericaeae), which is known from Peru, Bolivia and southern Argentina and Chile (and introduced in Britain and Ireland).

**Etymology.** The specific name "*gaultheriae*" is the plant host genus name of the aphid, in genitive.

**Taxonomic discussion.** Species of 33 genera of Ericaceae have been reported as host plants of aphid species around the World, but only 16 host species of *Aphis*, many of which are polyphagous. Only seven *Aphis* species specializing on plant species be-



**Figure 4.** *Aphis gaultheriae* sp. n., apterous viviparous female. **A** end of rostrum **B** end of tibia and tarsus of hind leg **C** siphunculus **D** cauda.

longing to Ericaceae are known: *A. arbuti* Ferrari, 1872, *A. callunae* Theobald, 1915, *A. madronae* Essig, 1926, *A. multiflorae* Barbagallo & Stroyan, 1982, *A. remaudieri* Börner, 1952, *A. uvaeursi* Ossiannilsson, 1959 and *A. vaccini* (Börner, 1914), which are European in distribution. Apterous viviparous females of *Aphis gaultheriae* sp. n. are recognizable by having spinal abdominal sclerotization, present at least on segments 5 to 8, extensively black femora, broad cauda more or less pigmented like siphunculi and similar to that of *A. spiraecola*, and an ultimate rostral segment no longer than the second segment of the hind tarsi.

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RESEARCH ARTICLE



# First records of the genus *Pelionella* Kaydan, 2015 in East Asia, with description of a new species (Hemiptera, Coccomorpha, Pseudococcidae)

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#### Abstract

Two mealybug species (Hemiptera: Coccomorpha: Pseudococcidae), *Pelionella osakaensis* **sp. n.** and *P. manifecta* (Borchsenius, 1949), are described and illustrated based on adult female specimens collected in Japan, on the Japanese mugwort *Artemisia indica* Willd. var. *maximowiczii* (Nakai) H. Hara (Asteraceae). These are the first records of the occurrence of *Pelionella* species in East Asia. The new species is similar to *P. grassiana* (Goux, 1989) and *P. proeminens* (Goux, 1990), but differs in lacking multilocular pores with double loculi rings on the venter and in possessing dorsal cerarii and a circulus. The Japanese population of *P. manifecta* is morphologically slightly different from the Azerbaijani and French populations in lacking large-type oral-collar tubular ducts associated with clusters formed by multilocular pores and oral-collar ducts on ventral abdominal segments III and IV. A modified key to species of the genus *Pelionella* Kaydan, 2015, is provided.

#### **Keywords**

Artemisia indica, Japanese mugwort, new distribution record, taxonomy

# Introduction

A genus of Pseudococcidae (Hemiptera: Coccomorpha), *Pelionella* Kaydan, 2015, was erected by Kaydan (2015) for a genus related to *Peliococcus* Borchsenius, 1948, based on molecular and morphological analyses. Kaydan (2015) defined the genus *Pelionella* 

based mainly on three diagnostic characters: (i) the presence of a special type of multilocular pore, consisting of double rings of eleven loculi, mainly located within pore clusters on the dorsum; (ii) the absence of a type of dorsal seta similar to cerarian setae that lacks a trilocular pore near the basal socket and is not located on an elevated area; and (iii) the presence of one or two sizes of dorsal oral-collar tubular ducts, and smaller ducts present in the center of clusters of multilocular pores with double rings on the dorsum. To date, eleven species of the genus have been recorded from the Western Palearctic and Eastern Nearctic (García Morales et al. 2016; Kaydan 2015) but none from East Asia, including Japan, although some species of the genus *Peliococcus* have been recorded (Kwon et al. 2003; Tanaka 2017; Tang 1992). *Peliococcus* and *Pelionella* can be clearly recognized by some morphological differences, such as presence or absence of a special type of multilocular pore consisting of double loculi rings, and clusters formed by the pores and several types of oral-collar ducts mainly on the dorsal surface.

Recently, the author examined specimens of two species of *Pelionella* collected from Japan, and recognized among the samples the type species of the genus, *Pelionella manifecta* (Borchsenius, 1949), and a single specimen of an undescribed species. The former showed slight differences from western populations in some morphological character states. This paper describes or diagnoses and illustrates both species collected from Japan based on adult female morphology, and constitutes the first record of the occurrence of *Pelionella* species in East Asia. A modified key to species of the genus *Pelionella* is also given.

This new distribution record for *P. manifecta*, and the description of a new species of *Pelionella* with unique morphological features (i.e., presence of several dorsal cerarii distinctly elevated from dorsal surface) may be useful for understanding and furthering studies on the diversity, morphology, and biogeography of this genus and other related mealybug species.

#### Materials and methods

Examined materials were collected by I. Takahashi, J. Imai, or K. Fujimoto in the fall (from October to November) of 2014. The adopted slide-mounting method is a slight modification of Kawai (1980) which uses lemosol (95% limonen) as a substitute for xylene. Slide-mounted specimens were examined under a phase-contrast compound microscope (Olympus BH2-PH, Tokyo, Japan). The description format and morphological terminology mostly follow Kaydan (2015). The holotype material of the new species and the voucher specimens of *P. manifecta* examined in this study are deposited in the National Museum of Nature and Science, Tsukuba, Japan (**NSMT**). The remaining materials of *P. manifecta* examined and used for description in this study are deposited in the Dr. Kawai scale insect collection in Tokyo University of Agriculture.

#### Taxonomy

### Genus Pelionella Kaydan, 2015

*Pelionella* Kaydan, 2015: 226. *Pelionella*; Danzig & Gavrilov-Zimin, 2014: 449 (Unavailable name).

Type species. Peliococcus manifectus Borchsenius, 1949: 245.

#### Pelionella manifecta (Borchsenius, 1949)

Figure 1

Peliococcus manifectus Borchsenius, 1949: 245: Danzig, 2001: 125.
Peliococcus albertaccius Goux, 1990: 83.
Pelionella manifecta (Borchsenius, 1949); Danzig and Gavrilov-Zimin 2014: 457 (as an unavailable name); Kaydan 2015: 234.

**Material studied.** All three adult females from Japan collected on *Artemisia indica* Willd. var. *maximowiczii* (Nakai) H. Hara (Asteraceae). Osaka-pref., Sennan City, Kansai International Airport, on: 1 adult female, 7.X.2014, coll. I. Takahashi; 1 adult female, 12.X.2014, coll. K. Fujimoto. Hyogo-pref., Kobe City, Chuo-ku, Minato-jima, Naka-machi: 1 adult female, 8.XI.2014, coll. J. Imai (NSMT-I-Ho 00082).

**Description. Slide-mounted specimens of Japanese populations, n = 3.** *Adult female.* Body elongate oval, 1.7–3.1 mm long, 0.9–1.8 mm wide. Eyes on margins, each 31–44 µm in diameter. Antenna 9-segmented, 393–444 µm long; apical segment 58–60 µm long, 22–25 µm wide; with two apical setae each 36–45 µm long, and three fleshy setae 18–30 µm long. Labium 95–110 µm long, 80–88 µm wide. Circulus oval, 95–100 µm wide, situated just anterior to fold between abdominal segments III and IV. Legs well developed; hind legs: coxa 120–140 µm long; trochanter + femur 248–282 µm long; tibia + tarsus 285–306 µm long; claw 30–32 µm long; translucent pores absent. Ratio of lengths of tibia + tarsus to trochanter + femur 1.05–1.18:1; ratio of lengths of tibia to tarsus 2.0–2.41:1; ratio of length of trochanter + femur to greatest width of femur 3.22–4.10:1. Tarsal digitules hair-like, each 23–30 µm long. Claw digitules knobbed, each 25–29 µm long. Claw with well-developed denticle on plantar surface. Anterior ostioles each with a total for both lips of 15–25 trilocular pores and 2–5 setae; posterior ostioles each with a total for both lips of 24–37 trilocular pores and 4–7 setae. Anal ring 73–108 µm wide, bearing 6 setae, each seta 127–190 µm long.

*Dorsum.* Setae spine-like, each 5–15  $\mu$ m long. Cerarii on margin somewhat prominent, slightly sclerotized, numbering 18 pairs; anal lobe cerarii each with 1–2 slender enlarged setae, each 10–22  $\mu$ m long, and one or two spine-like auxiliary setae; other cerarii mostly each with two enlarged setae and several trilocular pores.



**Figure 1.** *Pelionella manifecta* (Borchsenius, 1949) collected in Japan. Adult female. Abbreviations: **ALC**, anal lobe cerarius **ANT** antenna **DC** dorsal cluster **DS** dorsal setae **DMP** Multilocular pore with double rings **LG** leg **MP** multilocular pore **OCD** oral-collar tubular ducts **PC** Penultimate cerarius **QP** quinquelocular pore **TP** trilocular pore **VC** ventral cluster. Scale bars: 200 μm (**ANT, LG**); 50 μm (**ALC, PC**); 10 μm for the others.

Clusters of multilocular pores with double rings present on head and thorax and on abdominal segments as follows: I 9–12, II 12, III 14–15, IV 16–19, V 18–22, VI 10–15, VII 9–11, VIII 0; each cluster containing 1–7 (usually 2-3) multilocular pores with double rings, each pore 6.0–7.1  $\mu$ m in diameter; a small oral-collar tubular duct, 0.5–1.8  $\mu$ m wide; 1–5 large oral-collar tubular ducts, each 2.4–3.4  $\mu$ m wide; and 1–3 minute discoidal pores, each 1.1–1.3  $\mu$ m in diameter. Trilocular pores,

wide; and 1–5 minute discoldal pores, each 1.1–1.5 μm in diameter. Inflocular pores, each 3.2–3.9 μm in diameter, scattered throughout. Minute discoldal pores mainly restricted to within clusters. *Venter.* Setae of two types: (i) slender hair-like setae, each 10–142 μm long, long-

est setae of two types: (1) siender nair-like setae, each 10–142 µm long, longest setae situated medially on head; and (ii) spine-like setae in submarginal areas, each  $4-12 \mu m$  long. Apical setae of anal lobes 198–228 µm long. Multilocular disc pores with single ring, each 5.0–6.8 µm in diameter, present in 15–25 clusters on medial areas of abdominal segments III and IV; each cluster containing 1–5 (usually 2–3) multilocular disc pores surrounding a small oral-collar tubular duct; similar multilocular disc pores present also in single rows on other abdominal segments, as follows: V 7–8, VI 43–47, VII 62–69, VIII + IX 38–46. Multilocular pores with double rings, each 6.6–7.9 µm in diameter, restricted to submarginal areas of head, thorax, and abdomen, usually not arranged in clusters. Quinquelocular pores, each 3.2–5.6 µm in diameter, scattered medially on head, thorax, and medial area of abdominal segments. Trilocular pores, each 2.6–3.2 µm in diameter, scattered throughout. Minute discoidal pores, each 0.8–1.3 µm in diameter, few. Oral-collar tubular ducts of two sizes: small ducts restricted to within clusters; and large-sized ducts, each 2.1–2.9 µm wide, present on body margin and in single rows across posterior abdominal segments; also a few on head, thorax and abdominal segments II and III.

**Distribution.** Armenia, Azerbaijan, Corsica, France, Italy, Kazakhstan, Russia (Krasnodar Territory), Sardinia, Sweden, Turkey (Kaydan 2015), and Japan.

**Discussion.** The Japanese specimens of *Pelionella manifecta* described here differ slightly from the Azerbaijani and French material described by Kaydan (2015) in having many more multilocular disc pores on the venter of the abdominal segments, much smaller tubular ducts and pores (Table 1) and in lacking the large oral-collar tubular ducts associated with multilocular pore clusters on venter of abdominal segments III and IV. However, these morphological differences are herein tentatively regarded as intraspecific variation, because the number of tubular ducts and multilocular pores is known to vary greatly in some mealybug species (Cox 1983; Charles et al. 2000; Chatzidimitriou et al. 2016), considerable geographical morphological variation within *P. manifecta* has been also recorded (Kaydan 2015), and hitherto, the morphological variation of *P. manifecta* and taxonomic significances of the ducts' and pores' sizes have not been sufficiently studied. This description of the Japanese population may be useful for understanding phenotypic variation in *P. manifecta*.

In Japan, this species was collected from Kansai International Airport, one of the largest airports in the country, and from the large sea-port island of Kobe City (Mina-to-jima), both of which are centres of international trade. Furthermore, the species has not hitherto been recorded further east than Kazakhstan. This suggests that the species

Morphological features	Measurements of Japanese specimens (n = 3)	Measurements of Azerbaijani and French specimens (from Kaydan, 2015) (n = 5)
General morphological features		• • • • •
Length of body	1.7–3.1 mm	1.36–1.88 mm
Width of body	0.9–1.8 mm	0.86–1.10 mm
Width of eyes	31–44 μm	47.5–60.0 μm
Lendth of antenna (total)	393–444 μm	410–425 μm
Length of antenna's apical segment	58–60 μm	60 μm
Width of antenna's apical segment	22–25 μm	20–28 μm
Length of antenna's apical setae	36-45 μm	27–45 μm
Length of fleshly setae on antenna's apical segment	18–30 μm	25–33 μm
Length of labium	95–110 μm	135–140 μm
Width of labium	80–88 μm	95 μm
Width of circulus	95–100 μm	65–85 μm
Length of hind coxa	120–140 μm	155–175 μm
Length of hind trochanter and femur	248–282 μm	240–260 μm
Length of hing tibia and tarsus	285–306 µm	260–280 μm
Length of hind claw	30–32 µm	25–30 μm
Ratio of lengths of hind tibia + tarsus to hind trochanter + femur	1.05-1.18:1	1.07-1.23:1
Ratio of lengths of hind tibia to hind tarsus	2.0-2.41:1	2.16-2.41:1
Ratio of length of hind trochanter + femur to greatest width of hind femur	3.22-4.10:1	3.42-4.0:1
Length of hind tarsal digitules	23–30 µm	20–23 µm
Length of hind claw digitules	25–29 µm	20–25 µm
Morphological features on Dorsum		
The number of triolocular pores on anterior ostiole	15-25	21-30
The number of setae on anterior ostiole	2–5	2-4
The number of triolocular pores on posterior ostiole	24–37	32-40
The number of setae on posterior ostiole	4–7	2-4
Width of anal ring	73–108 μm	85–110 μm
Lenth of anal ring setae	127–190 μm	115–145 µm
Length of anal lobe cerarian setae	10–22 µm	17–23 µm
The number of auxiliary setae on anal lobes	1-2	3–4
Length of dorsal setae	5–15 µm	7.5–15 μm
The number of multilocular pore with double rings in clusters	1–7	2–6
Width of multilocular pores with double rings in clusters	6.0–7.1 μm	7.5–10.0 μm
Width of small oral collar tubular ducts in clusters	0.5–1.8 μm	3.0–4.0 μm
Width of large oral collar tubular ducts in clusters	2.4–3.4 μm	4.0–5.0 μm
The number of minute discoidal pores in clusters	1–3	1-4
Width of minute discoidal pores in clusters	1.1–1.3 μm	2 μm
The number of dorsal clusters in abdominal segment I.	9–12	9–11
The number of dorsal clusters in abdominal segment II.	12	10
The number of dorsal clusters in abdominal segment III.	14–15	11–13
The number of dorsal clusters in abdominal segment IV.	16–19	11–15
The number of dorsal clusters in abdominal segment V.	18–22	12
The number of dorsal clusters in abdominal segment VI.	10-15	8-11

**Table 1.** Comparisons of morphometric data between adult females of Japanese and Western Eurasian populations of *Pelionella manifecta*.

Morphological features	Measurements of Japanese specimens (n = 3)	Measurements of Azerbaijani and French specimens (from Kaydan, 2015) (n = 5)
The number of dorsal clusters in abdominal segment VII.	9-11	10-14
Width of triolocular pores	3.2–3.9 μm	3–5 μm
Morphological features on Venter		
Length of slender hair-like setae	10–142 μm	15–88 μm
Length of spine-like setae	4–12 μm	10.0–12.5 μm
Length of apical setae on anal lobes	198–228 μm	145–185 μm
Width of multilocular pores with double rings	6.6–7.9 μm	7.5–10.0 μm
Witdth of multilociular pores with single ring	5.0–6.8 μm	7.5–10.0 μm
The number of clusters on abdominal segments III-IV	15–25	10-14
The number of multilocular pores on abdominal segment V.	7-8	2–3
The number of multilocular pores on abdominal segment VI.	43-47	14–18
The number of multilocular pores on abdominal segment VII.	62–69	34-40
The number of multilocular pores on abdominal segments VIII+IX.	38-46	20–23
Width of quinquelocular pores	3.2–5.6 μm	5.0–7.5 μm
Width of triolocular pores	2.6–3.2 μm	2–3 μm
Width of minute discoidal pores	0.8–1.3 μm	2 µm

might not be truly endemic to Japan, but be a recent introduction. Studies of the detailed distribution of the species in Japan, and the current condition of the species at the sites where it was collected originally, may be important from both biological and plant-quarantine perspectives.

#### Pelionella osakaensis sp. n.

http://zoobank.org/F2AB23EA-CD3B-4CB1-938A-DED7C5F714FE Figure 2

**Holotype.** Adult ♀. Japan, Osaka-pref., Sennan City, Kansai International Airport, 12.X.2014, host plant: *Artemisia indica* var. *maximowiczii*, coll. K. Fujimoto. (NSMT-I-Ho 00081).

**Diagnosis.** Eighteen pairs of cerarii present on body margin. Several slightly elevated dorsal cerarii also present on dorsal surface. Clusters of multilocular pores with double rings present on dorsum; each cluster contains 1–2 multilocular pores with double rings, 0–1 small oral-collar tubular ducts, 0–2 large oral-collar tubular ducts, and 0–3 minute discoidal pores. Multilocular pores with double rings and clusters of multilocular disc pores with single ring and oral-collar tubular ducts absent on venter. Circulus oval, present on posterior part of third abdominal segment of venter. Translucent pores absent on hind legs.

**Description. Slide-mounted specimen.** Adult female. Body elongate oval, 1.7 mm long, 0.9 mm wide. Eyes submarginal, each  $30-32 \ \mu m$  in diameter. Antenna 9-segmented,  $363-387 \ \mu m$  long; apical segment  $53-57 \ \mu m$  long,  $20-25 \ \mu m$  wide; with two apical setae each  $30-38 \ \mu m$  long, and three fleshy setae each  $20-30 \ \mu m$  long. Labium



**Figure 2.** *Pelionella osakaensis* sp. n. adult female (holotype). **ALC**, anal lobe cerarius **ANT** antenna **DC** dorsal cluster **DS** dorsal setae **LG** leg **MP** multilocular pore **OCD** oral-collar tubular ducts **QP** quinquelocular pore **PC** Penultimate cerarius **TP** trilocular pore. Scale bars: 200 μm (**ANT, LG**); 50 μm (**ALC, PC**); 10 μm for the others.

103 µm long, 68 µm wide. Circulus oval, approx. 74 µm wide, situated on posterior part of third abdominal segment. Legs well developed; posterior legs: coxa 82–85 µm long; trochanter + femur 248–250 µm long; tibia + tarsus 275–278 µm long; claw 30–34 µm long. Translucent pores absent. Ratio of lengths of hind tibia + tarsus to trochanter + femur 1.1:1; ratio of lengths of tibia to tarsus 1.4–1.5:1; ratio of length of trochanter + femur to greatest width of femur 3.0:1. Tarsal digitules hair-like, each 20–31µm long. Claw digitules knobbed, each 28 µm long. Claw with well-developed denticle on plantar surface. Anterior ostioles with a total for both lips of 26 to 30 trilocular pores and 2–3 setae; posterior ostioles with a total for both lips of 33–36 trilocular pores and 5–6 setae. Anal ring 85 µm wide, bearing 6 setae, each seta 103–125 µm long.

*Dorsum.* Cerarii on margins slightly prominent but with no sclerotization, numbering 18 pairs; anal lobe cerarii each with 2–3 slender enlarged setae, each 11–21  $\mu$ m long, and 2–3 spine-like auxiliary setae; other cerarii on margins each with 2–3 slender enlarged setae and several trilocular pores. Several dorsal cerarii present on dorsal surfaces as shown in Figure 2, each cerarii with 1–2 relatively small enlarged setae, no auxiliary setae and several triolocular pores; each slightly prominent but with no sclerotization. Setae spine-like, each 5–15  $\mu$ m long; larger setae each with 1–3 trilocular pores near base, sometimes forming dorsal cerarii. Multilocular pores with double rings, each 5.5–6.2  $\mu$ m in diameter, in each cluster present singly or in pairs together with 0–1 small oral-collar tubular ducts, 1.8–2.0  $\mu$ m wide, plus 0–2 large oral-collar tubular ducts, each 2.5–2.8  $\mu$ m wide, and 0–3 minute discoidal pores, each 1.2–1.8  $\mu$ m in diameter, scattered throughout. Several small oral-collar ducts, each 3.2–3.8  $\mu$ m in diameter, scattered throughout. Several small oral-collar ducts, each 1.8–2.0  $\mu$ m wide, each 1.8–2.0  $\mu$ m wide, each 3.2–3.8  $\mu$ m in diameter, scattered throughout. Several small oral-collar ducts, each 1.8–2.0  $\mu$ m wide, each 1.2–1.8  $\mu$ m in diameter, scattered throughout. Several small oral-collar ducts, each 1.8–2.0  $\mu$ m wide, each 1.2–1.8  $\mu$ m in diameter, scattered throughout. Several small oral-collar ducts, each 1.8–2.0  $\mu$ m wide, each 1.2–1.8  $\mu$ m in diameter, scattered throughout. Several small oral-collar ducts, each 1.8–2.0  $\mu$ m wide, each 1.2–1.8  $\mu$ m in diameter, scattered throughout. Several small oral-collar ducts, each 1.8–2.0  $\mu$ m wide, each 1.2–1.8  $\mu$ m in diameter, scattered throughout. Several small oral-collar ducts, each 1.8–2.0  $\mu$ m wide, and minute discoidal pores, each 1.2–1.8  $\mu$ m in diameters.

*Venter.* Setae of two types: (i) hair-like setae, each 14–81  $\mu$ m long, longest present on medial area of posterior abdominal segments; and (ii) spine-like setae, each 5–10  $\mu$ m long, present in submarginal areas. Apical setae of anal lobes each 162–169  $\mu$ m long. Multilocular disc pores, each 5.5–6.5  $\mu$ m in diameter, present in bands on abdominal segments as follows: IV 4, V 0, VI 28, VII 34, VIII + IX 29. Quinquelocular pores, each 3.5–4.5  $\mu$ m in diameter, scattered medially on head, thorax, and first four abdominal segments. Trilocular pores, each 2.8–3.2  $\mu$ m in diameter, scattered throughout. Minute discoidal pores, each 1.0–1.2  $\mu$ m in diameter, few in number. Oral-collar tubular ducts of 1size, each 1.9–2.1  $\mu$ m wide, mostly present in bands across posterior abdominal segments and on medial areas of thoracic segments; a few ducts present in submarginal areas.

**Etymology.** The species is named after the prefecture in Japan where it was collected.

**Discussion.** *Pelionella osakaensis* sp. n. is quite similar to *P. grassiana* (Goux, 1989) and *P. proeminens* (Goux, 1990) in having clusters containing one or two multilocular pores with double rings on dorsum and more than 16 pairs of cerarii. However, *P. osakaensis* differs from the latter species in having a circulus on the posterior part of the third abdominal segment, several slight elevated dorsal cerarii, and in lacking translucent pores on hind legs. Although the presence or absence of a circulus can be variable within a mealybug spe-

cies, it may be a useful, readily observable diagnostic character for *P. osakaensis* given the current status of classification of *Pelionella* species. *Pelionella osakaensis* is also similar to *P. stellarocheae* (Goux, 1990) in lacking translucent pores on hind legs and in having smaller number of multilocular pores with double rings in each cluster on dorsum; however, it clearly differs from *P. stellarocheae* in having 18 pairs of cerarii plus dorsal cerarii. The presence of dorsal cerarii is one of the important features of *P. osakaensis*, although it may appear to conflict with the generic definition of *Pelionella* proposed by Kaydan (2015). Here the importance of the presence of multilocular pores with double rings in the clusters is emphasized, so the new species is considered to belong to the genus *Pelionella*. Further research into the generic definition of the genus *Pelionella* is still much needed.

*Pelionella osakaensis* has only been collected from the site of Kansai International Airport, one of the largest airports in Japan, so it is possible that it is not endemic. A more detailed distributional study of the species and the current population level and distribution of the species at the airport may be important in relation to plant-quarantine measures.

# Key to species of the genus *Pelionella* based on adult female morphology (adopted and partially modified from Kaydan 2015)

1	Clusters on dorsum each with only 1 size of oral-collar tubular duct; mul-
	tilocular pores with double rings around cluster without larger oral-collar
	tubular ducts2
-	Clusters on dorsum each with 2 sizes of oral-collar tubular ducts, with smaller
	ducts in center of each cluster and larger ducts and multilocular pores with
	double rings around cluster (very rarely multilocular pores with double rings
	few on dorsum but ducts still in clusters, i.e., P. glandulifer (Borchsenius) and
	<i>P. tritubulata</i> (Kiritchenko)
2	With 3-6 (generally fewer than 5) multilocular pores with double rings in
	each cluster on thorax and head; each anal lobe cerarius with 4 enlarged cerar-
	ian setae
_	With 4-8 (generally more than 5) multilocular pores with double rings in
	each cluster on thorax and head; each anal lobe cerarius with only 2 enlarged
	setae and 2 smaller auxiliary setaeP. cycliger (Leonardi, 1908)
3	Circulus present
_	Circulus absent
4	Quinquelocular pores numerous on venter; marginal cerarii numbering 13-
	18 pairs
_	Quinquelocular pores absent or, if a few present, mainly around mouthparts;
	fewer than 8 pairs marginal cerarii9
5	Each dorsal cluster with 0-4 multilocular pores with double rings and 0-3
	large oral-collar tubular ducts6
_	Each dorsal cluster with $2-16$ multilocular pores with double rings, and $2-13$
	large oral-collar tubular ducts7

6	Clusters on dorsum few (3-5 in total); each cluster normally without multi-
	locular pores with double rings but with 1–3 large oral-collar tubular ducts
	around a central smaller duct <i>P. tritubulata</i> (Kiritchenko, 1940)
-	Clusters abundant throughout dorsum, each cluster with 1-3 multilocular
	pores with double ring, 0-3 large oral-collar tubular ducts and 0-1 small
	oral-collar tubular duct
7	Each cluster with 5–16 (usually 8–10) multilocular pores with double rings,
	5-13 large oral-collar tubular ducts, and 7-9 minute discoidal pores; quin-
	quelocular pores extremely sparse on venter P. multipora Kaydan, 2015
-	Each cluster with 1-7 (usually 2-4) multilocular pores with double rings,
	1–5 large oral-collar tubular ducts, and 1–4 minute discoidal pores; quinque-
	locular pores common on venter
8	Dorsal cerarii absent. Cerarii numbering 13 pairs
_	Dorsal cerarii present. Cerarii numbering 18 pairs
9	Multilocular pores with double rings on dorsum generally absent; if present,
	very few, restricted to posterior abdominal segments; each cluster with 0 or 2
	(usually 0) multilocular pores with double rings, 1-4 large oral-collar tubular
	ducts, and 2-4 minute discoidal pores P. glandulifer (Borchsenius, 1949)
_	Multilocular pores with double rings present in clusters on dorsum, each
	cluster with 2-5 (usually 3) multilocular disc pores, 2-5 large oral-collar tu-
	bular ducts, and 2-4 minute discoidal pores
10	Marginal cerarii numbering 14–18 pairs; multilocular disc pores restricted to
	abdominal segments VI–VIII11
_	Marginal cerarii numbering fewer than 4 pairs; multilocular disc pores pre-
	sent on abdominal segments IV–VIII
11	Clusters on dorsum common and in distinct rows on each segment; femur
	without translucent pores
_	Clusters on dorsum sparsely distributed on each segment, not forming dis-
	tinct rows; femur with translucent pores

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RESEARCH ARTICLE



# A faunistic study of genus Chasmogenus Sharp, 1882 of China (Coleoptera, Hydrophilidae)

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#### Abstract

*Chasmogenus* Sharp, 1882 is newly reported from the Chinese Mainland. A new species, *C. parorbus* **sp. n.**, is described from China (Yunnan). *Chasmogenus orbus* Watanabe, 1987 is reported from Hong Kong, the first record outside Japan. *Chasmogenus abnormalis* (Sharp, 1890) is reported from the Chinese mainland for the first time. The male genitalia of each species are illustrated. A key to the Chinese species of the genus is provided.

#### **Keywords**

Chasmogenus, China, Hydrophilidae, new record, new species, Oriental region

# Introduction

The genus *Chasmogenus* was erected by Sharp (1882) based on *Chasmogenus fragilis* Sharp, 1882. d'Orchymont (1919) treated it as a subgenus of *Helochares* Mulsant, 1844 until Fernández (1986) separated it as a valid genus. Hebauer (1992) revised all known species of the genus up to that time and recorded 24 species, with Hansen (1999) listing 26 species globally. Since then, new species have been described from the Oriental, Australian, Neotropical and Afrotropical regions (Watts 1998; Garcia 2000; Hebauer 1995, 2001, 2002, 2006; Short 2005, 2010, Short and Hebauer 2006;

Clarkson and Ferreira 2014). A total of 43 species is currently known, of which 18 occur in the Afrotropical region, eleven in the Neotropic, two in the Palearctic, three in the Australian, and five in the Oriental region. *Chasmogenus abnormalis* (Sharp, 1890), known from Cambodia, Indonesia (Borneo, Java, Sulawesi, Sumatra), Sri Lanka, China (Taiwan), Thailand, Vietnam, and the Ryukyu Islands (Hebauer 1992), was considered by Hansen (1999) to occur in the Palearctic region. Based on the fauna of insects, the authors prefer to consider Ryukyu Islands as a part of the Oriental region. Therefore, *C. abnormalis* (Sharp, 1890) should be treated as an Oriental species only.

The genus has been poorly known from China up to now. Only *C. abnormalis* (Sharp, 1890) is known from Taiwan (Watanabe 1987; Hebauer 1992; Hansen 1999) and no species have been reported from mainland China to date. Since 2006, some material of *Chasmogenus* was collected by from southern China, confirming that three species occur in China. In this contribution, a new species is described and two species new for mainland of China are reported.

#### Materials and methods

Specimens of each species were dissected, and the genitalia placed in a drop of glycerol on glass slides. After photography, genitalia were transferred to a plastic mount pinned with the respective specimen. Habitus photographs were taken using an Axioskop 40 compound microscope with AxioCam HRc Rev. 3/3.3v (4164×3120). Photographs of genitalia were taken using an Olympus SZX7 stereomicroscope, and subsequently combined with Auto-Montage software. The SEM photograph was taken using a Phenom Prox scanning electronic microscope. Complete label data are provided for type specimens, exact label data in English being cited for the type material (data in Chinese are translated into English). All specimens used in this study are deposited in the collection of Sun Yat-sun University, Guangzhou, China (**SYSU**).

#### Taxonomy

#### Chasmogenus Sharp, 1882

Chasmogenus Sharp, 1882: 73. Type species: Chasmogenus fragilis Sharp.Crephelochares Kuwert, 1890: 38. Type species: Helochares livornicus Kuwert. Syn.: d'Orchymont 1919: 148; Fernández 1986: 148.

**Diagnosis.** The following character combinations can be used to separate *Chasmogenus* Sharp, 1882 from other genera: 1) maxillary palpi at least as long as width of head; 2) second maxillary palpomeres curved inward, apical segment almost symmetrical, as long as penultimate; 3) clypeus not concealing labrum and not expanded in front

of eyes; 4) mesoventrite with rather strong median carina for entire length; 5) elytra without striae or rows of serial punctures; 6) elytra with sutural stria on posterior half; 7) posterior margin of 5<sup>th</sup> abdominal ventrite with a small apical emargination.

# Chasmogenus parorbus sp. n.

http://zoobank.org/3496519B-82B5-42E1-B6B6-C5A4A069A99F Figs 1–8

**Type material. Holotype: CHINA. Yunnan:** male (SYSU), CHINA: Yunnan Prov., Yingjiang, Tongbiguan, Kaibangyahu, 24.58°N, 97.67°E, 1289m, 25.V.2016, Yu-dan Tang & Rui-juan Zhang leg. [transcribed from Chinese]. **Paratypes** (3 males, SYSU): 2 males, same data as holotype; 1 male., **Yunnan,** Yingjiang, Nabang, 24.75°N, 97.56°E, 239m, 27.V.2016, Yu-dan Tang & Rui-juan Zhang leg. [transcribed from Chinese].

**Diagnosis.** Body oblong (Fig. 1). Black, pronotum and elytra with paler margins. Head, pronotum and elytra with distinct systematic punctures. Ground punctures of pronotum and elytra finer laterally and posteriorly than on disc (Figs 1, 3). Mesoventrite with a longitudinal and moderate high ridge posteromedialy (Fig. 5). Hind femora pubescent on basal four-fifths. Parameres shorter than median lobe, inner margin with a subapical tooth, apex obtuse rounded. Median lobe broad, gradually narrowing from apical fourth to apex, moderately curved on apical fifth and strongly curved subapically, apex truncate (Fig. 8).

**Description.** Length 3.4–3.7 mm, width 1.7–2.0 mm. Body oblong, moderately convex. Dorsum of head, pronotum and elytra black with paler margins. Maxillary and labial palpi, and antennae uniformly yellowish brown (Figs 1–4). Ventral surface and legs reddish brown or dark brown.

**Head.** Labrum sparsely and finely punctate, with slightly emarginate anterior margin and round arcuation on posterior margin. Clypeus emarginate anteriorly (Figs 6–7), systematic punctures distinct, ground punctures fine and sparse, with distance between punctures  $1.5-3.0 \times$  the width of one puncture. Frons with distinct systematic punctures, ground punctures as fine as those on clypeus but a little denser. Mentum transverse, ca.  $2 \times$  as wide as long, rugose, with distinct oblique sculptures and a few strong punctures, strongly depressed anteriorly, anterocentral notch developed (Fig. 4). Maxillary palpi obviously longer than width of head. Antenna with nine antennomeres, pedicel as long as antennomeres 3 to 6 combined.

**Thorax.** Pronotum with distinct systematic punctures, ground punctures on disc as fine as those on frons, with distance between punctures  $1.5-3.0 \times$  the width of one puncture; lateral portions with finer punctures. Prosternite slightly bulged in middle, not carinate, with sparse pubescence, with transverse groove anteriorly (Fig. 4). Elytra with fine punctures similar to those of pronotum, systematic punctures distinct; with distinct sutural stria on posterior three-quarters. Mesoventrite with a longitudinal carina posteromedially, completely fused with mesanepisterna. Metaventrite pubes-



Figures 1–7. *Chasmogenus parorbus* sp. n. I dorsal habitus 2 ventral habitus 3 lateral habitus 4 head and prosternite, ventral 5 meso-, and metaventrite, ventral 6–7 Head, dorsal.

cent with an irregular glabrous area (Figs 2, 5). Meso- and metafemoral pubescence on basal four-fifths.

*Abdomen.* Abdominal ventrites densely pubescent. Apical margin of fifth ventrite with shallow emargination (Fig. 2).

*Aedeagus.* Parameres shorter than median lobe, inner margin with a tooth subapically, apex obtuse rounded. Median lobe broad, gradually narrowing from apical quarter to apex, moderately depressed on apical fifth and strongly curved subapically, apex truncate (Fig. 8).

**Remarks.** This species is close to *C. orbus* Watanabe, 1987. It can be distinguished from *C. orbus* by parameters obtuse apically, subapical tooth sharper; median lobe slender, slightly but distinctly constricted in apical quarter (Fig. 8).

**Etymology.** The species name is combined from Latin "para-", similar, and *orbus*, a species name of the genus.

**Biology.** Aquatic, living in stagnant pools.

Distribution. Only known from type locality.

# Chasmogenus orbus Watanabe, 1987

Fig. 9

Helochares (Crephelochares) orbus Watanabe, 1987: 12 (for detailed description).

**Material examined. CHINA: Hong Kong:** 2 males, 5 females (SYSU), Hong Kong, Rongshu'ao, 22°25.641'N, 114°17.410'E, 10m. 11.vi.2014, Fenglong Jia, Weicai Xie & Jiahuang Chen leg. [transcribed from Chinese].

**Remarks.** Based on the original description of *C. orbus* by Watanabe (1987), the specimens here appear completely identical to this species. The senior author has dissected two males and sent photos of the aedeagus to Dr. Minoshima for comparison with Japanese specimens; he informed the senior author (via email) that he did not see any difference between the photos and the specimens from Japan. Similar to *C. parorbus* sp. n., it can be distinguished from this species by parameres being sharp apically, with the subapical tooth less sharp; and the median lobe broader, not constricted in apical quarter (Fig. 9).

Distribution. China (Hong Kong); Japan. New for China.

#### Chasmogenus abnormalis (Sharp, 1890)

Fig. 10

*Philydrus abnormalis* Sharp, 1890: 351; Hansen 1999: 174 (complete synonymy); Devi et al. 2016: 296.

Material examined. CHINA: Guangdong: 4 males, 6 females (SYSU), Guangdong, Zhuhai, Hengqing island, 10.VII.2006, Fenglong Jia leg. [transcribed from Chinese].



Figures 8–10. Aedeagi (dorsal view): 8 Chasmogenus parorbus sp. n. 9 C. orbus Wlatanabe 10 C. abnormalis Sharp.

Macau: 2 females (SYSU), Cotai Ecosystematic Reserve, part 1, 8.iv.2014, Weicai Xie et Jinwei Li leg. [transcribed from Chinese].

**Remarks.** This species is close to *C. orbus* from which it can only be distinguished by the aedeagus. Parameres without or with small subapical tooth (Devi et al. 2016: figs 10–13), apex broadened inwards (Fig. 10). Median lobe gradually narrowing from apical quarter to apex, moderately constricted in apical fifth and strongly curved subapically, apex truncate (Fig. 10).

**Distribution.** China (Guangdong, Macau); Cambodia, India, Indonesia, Japan, Sri Lanka, Thailand, Vietnam.

#### Key to the species of the Chasmogenus of China

- inwards apically, median lobe widely truncate apically.... *C. abnormalis* (Sharp)
- Parameres with apex obtuse and subapical tooth sharper. Median lobe relatively slender, distinctly constricted in apical quarter (Fig. 8)...... *C. parorbus* sp. n.

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RESEARCH ARTICLE



# Description of a new species of Coelosis Hope from Guajira Peninsula, northern Colombia (Coleoptera, Scarabaeidae, Dynastinae, Oryctini)

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# Abstract

A new species of *Coelosis* is described from the Macuira Mountains, Guajira Peninsula, northern Colombia. A character comparison between this new and other previously known Colombian species in the genus is presented. A key for the identification and distributional map for Colombian species of *Coelosis* is provided, as well as a key for the genera included in the tribe Oryctini in Colombia.

# Resumen

Una nueva especie de *Coelosis* es descrita de la Serranía de la Macuira en la Peninsula de la Guajira, en el extremo norte de Colombia. Se ilustran los caracteres diagnósticos y se compara la nueva especie con las especies previamente reportadas para Colombia. Se provee una clave para la identificación y un mapa de distribución de las especies de *Coelosis* en el país, al igual que una clave para los géneros de la tribu Oryctini en Colombia.

# Keywords

Distribution, Macuira Mountains, Scarabaeoidea, Taxonomy

#### Palabras claves

Distribución, Serranía de la Macuira, Scarabaeoidea, Taxonomía

### Introduction

The genus *Coelosis* is a Neotropical dynastine group of mainly nocturnal forest beetles that can be collected at lights (Endrödi 1985, Iannuzzi and Marinoni 1996, Ratcliffe 2003). The larvae of *Coelosis biloba* (L.) is the only one described in the genus and is known to be associated with the nests of the leaf-cutter ant *Atta cephalotes* (L.) (Hymenoptera: Formicidae) (Pardo-Locarno et al. 2006).

The purpose of this work is to describe a new species of *Coelosis* from a tropical dry forest in the protected area of Macuira, Guajira Pensinsula, to provide new biological data and distribution records for other species *Coelosis*, and to provide diagnostic characters and illustrations for all three Colombian species in the genus *Coelosis* as well as a key to the genera of Oryctini from Colombia.

#### Materials and methods

Internal and external morphological characters were studied using a dissecting microscope (6.5–40.0×). For measurements, an ocular micrometer was used. Internal sclerotized structures were dissected after relaxing the specimen in hot (75 °C) water. Heavily sclerotized parts were soaked in a 15 % solution of potassium hydroxide and neutralized in a 15 % solution of acetic acid. Genitalia were card-mounted or placed in a glycerin–filled vial beneath the specimen.

Specimens were characterized using body length, puncture density, setation, and color as described in Orozco (2012).

#### Material examined

138 specimens were reviewed and label data were obtained from the following Colombian collections except where noted otherwise (curators in brackets):

ANDES	Colección de Entomología, Universidad de los Andes, Bogotá D.C.
	(Oscar Mahecha).
ICN-MHN	Colección de Zoología, Instituto de Ciencias Naturales, Universidad
	Nacional de Colombia, Bogotá D.C. (Germán Amat-García).
IAvH	Colecciones Biológicas, Sección de Entomología, Instituto de Inves-
	tigaciones de Recursos Biológicos Alexander von Humboldt, Villa de
	Leyva, Boyacá.
LGA	Museo de Historia Natural "Luis Gonzalo Andrade". Colección Ento-
	mológica. Universidad Pedagógica y Tecnológica de Colombia, Tunja,
	Boyacá (Fredy Molano).
MEFLG	Museo Entomológico Francisco Luis Gallego, Universidad Nacional
	de Colombia, sede Medellín, Antioquia (Jhon Alveiro Quiroz).

MEUC	Museo de Entomología, Universidad de Cundinamarca, Fusagasugá,
	Cundinamarca.
MLP	Museo de La Plata, Colección de Entomología, La Plata, Buenos Aires,
	Argentina (Nhora Cabrera and Analía Lanteri).
MPUJ	Museo Javeriano de Historia Natural "Lorenzo Uribe", Pontificia Uni-
	versidad Javeriana, Bogotá, D.C. (Dimitri Forero).
MUA	Colección de Ciencias Naturales, Universidad de Antioquia, Medellín,
	Antioquia (Marta Wolf).
MUSENUEV	Museo Entomológico, Universidad del Valle, Cali, Valle del Cauca
	(James Montoya).
UNAB	Museo Entomológico, Facultad de Agronomía, Universidad Nacional
	de Colombia, Bogotá, D.C. (Francisco Serna y Erika Vergara).

Additional locality information was obtained from Sanabria-García et al. (2012). ArcMap 10.0 (ESRI 2011) was used to build the distributional map.

# Results

# Key to the genera of Oryctini from Colombia

1	Labium subtriangular or with bulging lateral margins (Fig. 2E, F, G). Man-
	dibles with 1-2 teeth on external margin (Fig. 2H-J). Second segment of
	maxillary palp longer than first and third segments (Fig. 2A–D)2
_	Labium subrectangular, lateral margins subparallel (Fig. 4C, F, I). Mandibles with
	three teeth on external margin (Fig. 4A, B, D, E, G, H). Second segment of maxillary
	palp similar in length to first and third segment (Fig. 5A, B, C) Coelosis Hope
2	Maxilla with 5–6 teeth (Fig. 2C, D)
_	Maxilla with 1–3 teeth (Fig. 2A, B) <b>5</b>
3	Male with one pronotal horn and one large cephalic horn or tubercle (Fig.
	1F). Female without fovea on pronotum
_	Male with three pronotal horns and/or tubercles and lacking large cephalic
	horn. Female with fovea on pronotum
4	Elytra smooth (Fig. 1E)Strategus Kirby
_	Elytra with rows of deep punctures (Fig. 1B) Gibboryctes Endrödi
5	Protibia tridentate (Fig. 1D). Apex of labium pointed, paraglossa undevel-
	oped (Fig. 2E)
_	Protibia quadridentate (Fig. 1A, B, E, F). Apex of labium blunt, paraglossa
	developed (Fig. 2F, G)
6	Mandibular teeth widely separated at base (Fig. 2H). Males and females with
	cephalic horns (Fig. 1A)
_	Mandibular teeth contiguous or fused at base (Fig. 2I). Cephalic horns pre-
	sent only in males



**Figure I.** Colombian Oryctini (dorsal habitus). **A** *Enema pan* (Fabricius) **B** *Gibboryctes waldenfelsi* (Endrödi) **C** *Heterogomphus chevrolati* Burmeister **D** *Megaceras porioni* Dechambre **E** *Strategus fascinus* Burmeister **F** *Podischnus agenor* (Olivier).



**Figure 2. A, H** Enema pan **B, F, I** Heterogomphus chevrolati **C** Gibboryctes waldenfelsi **D** Podischnus agenor **E** Megaceras morpheus Burmeister **G, J** Strategus fascinus **A–D** maxilla (dorsal view) **E–G** labium (ventral view) **H–J** mandibles (dorsal view).

# Key to adults of the species of Coelosis from Colombia

1	Maxilla without lateral projection (Fig. 5B) Coelosis biloba (I	L.)
_	Maxilla with lateral projection (Fig. 5A, C)	2
2	Galea with two teeth (Fig. 5A). Prohypomeron and metasternum with lo	ng
	setae (Fig. 6F) Coelosis bicornis (Lesk	ke)
_	Galea without teeth (Fig. 5C). Prohypomeron and metasternum with she	ort
	setae (Fig. 6G) Coelosis wayuorum sp.	n.

#### Coelosis wayuorum sp. n.

http://zoobank.org/863A21DA-0365-4404-B1BF-C125EE00B609 Figs 3F–G; 4G–I; 5C, F, I, L; 6C, E, G, J, K, M; 7E–F; 8

**Type material (3)**. Holotype labeled "Colombia, La Guajira, Uribia, PNN La/Macuira Corregimiento Nazareth/Kajashiwoü, 12°11'37.9"N; 71°21'30.1"/W. WGS84, 70 m. Manual.19.ix.2014/C. Medina" [IAvH-E-195379]. Allotype [IAvH-E-195380] and one female paratype [IAvH-E-195381] with the same label data. Types deposited at the Instituto Alexander von Humboldt (IAvH) Villa de Leyva, Boyacá, Colombia.

**Diagnosis.** *Coelosis wayuorum* sp. n. can be separated from the other Colombian *Coelosis* by the following characters: maxilla with lateral sclerite pronounced (Fig. 5C) [similar to *C. bicornis*, but not as in *C. biloba*]; galea without teeth (Fig. 5C) [two teeth in both *C. bicornis* and *C. biloba*]; ventral surface of the mandibles with keels 1 and 2 contiguous (Fig. 4H) [widely separate in *C. bicornis* (Fig. 4B), slightly separate in *C. biloba* (Figs 4E)]; pronotum and elytra strongly punctate, punctures with short, spine-like setae (Figs 6C, E) [spine-like setae absent in both *C. bicornis* and *C. biloba* (Fig. 6A, B, D)]; prohypomeron with spine-like setae [slender and long in *C. bicornis*, slender and short in *C. biloba*]; mesosternum convex as in *C. biloba* [concave in *C. bicornis*]; metasternum covered with short, spine-like setae (Fig 6G) [setae long and slender in both *C. bicornis* and *C. biloba* (Fig. 6J, K) [scarcely punctate in both *C. bicornis* and *C. biloba* (Fig. 6H, I)]. The internal sac is different among the species: in *C. biloba*, the accessorial lamella is short and simple (Fig. 5H), while in *C. bicornis* and *C. wayuorum* sp. n. the lamella is long and complex, although with differences between these two species (Fig. 5G, I).

**Description.** Holotype male (Fig. 3F). Body length 20.2 mm; width 11.00 mm. Color dark reddish brown. *Head*: Frontoclypeal region with small horn, surface rugopunctate, punctures setose, setae spine-like (Fig. 3F). Clypeus strongly rugopunctate, slightly emarginate, apex broad with two reflexed teeth. Mandibles with three conical teeth, subapical notch slightly deep (Fig. 4G, H); labium rugopunctate, with sparse, short, spine-like setae, paraglossa undeveloped, narrow, apex truncate (Fig. 4I); maxilla with lateral projection, galea without teeth (Fig. 5C). *Pronotum*: Surface sparsely punctate; punctures moderately large, umbilicate, setose; setae spine-like (Fig. 6C). Disc with two small, widely separated horns, wide fovea between horns (Figs 3F, 6C).


Figure 3. *Coelosis* species from Colombia (dorsal habitus): A (male major) B (female) C (male minor): *C. bicornis* D (male major): *C. biloba* (L.) E (female): *C. biloba* F Holotype (male): *C. wayuorum* sp. n.
G Allotype (female): *C. wayuorum* sp. n.



Figure 4. A, B, C *Coelosis bicornis* D, E, F *C. biloba* G, H, I *C. wayuorum* sp. n. A, D, G mandibles (dorsal view) B, E, H mandibles (ventral view) C, F, I labium (ventral view).



Figure 5. A, D, G, J *Coelosis bicornis* B, E, H, K *C. biloba* C, F, I, L *C. wayuorum* sp. n. A–C maxilla (ventral view) D–F prosternal process (antero-posterior view) G–I internal sac (copulatory lamellae) J–L genital plate (female).



Figure 6. A, B, D, F, H, I, L *Coelosis bicornis* C, E, G, J, K, M *Coelosis wayuorum* sp. n. A–C pronotum (dorsal view) D, E elytra F, G metasternum H, J mesotibia (dorsal view) I, K metatibia (dorsal view) L, M first metatarsomere (dorsal view).

*Elytra*: Surface with 10 distinct pairs of striae composed of ocellate punctures bearing short, spine-like setae, micropunctures densely intermixed between striae (Fig. 6E). *Py-gidium*: Surface sparsely punctate, punctures small to moderate in size, weakly ocellate,



Figure 7. A, B *Coelosis bicornis* C, D *C. biloba* E, F *C. wayuorum* sp. n. A, C, E parameres (frontral view) B, D, F phallobase (lateral view).

with short spine-like setae, becoming denser at basal angles. Surface regularly convex in lateral view. *Legs*: Protibia tridentate, basal tooth reduced. Meso- and metatibiae with one medial transverse carina, each with short, spine-like setae and one small inner



Figure 8. Distribution of *Coelosis* in Colombia.

tooth (Fig. 6J, K). First metatarsomere apically expanded, apex subtruncate with acute outer projection (Fig. 6M). *Venter*: Prosternal process subtriangular, thick; apex short, parabolic, with process at middle, this process with short, stout setae (Fig. 5F). Mesosternum densely punctate, setose, slightly convex at middle. Metasternum densely

punctate; punctures ocellate, minutely setose; setae spine–like; lateral edge rugopunctate with short, spine-like setae. Abdominal ventrite VIII depressed at middle. *Genitalia*: Parameres as in Fig. 7E, F, internal sac as in Fig. 5I.

**Allotype** (Fig. 3G). Female. Similar to holotype except for the following: body length 22.4 mm; width 12.0 mm, frons and disc of pronotum smooth, without fovea or horns, pygidium slightly concave in lateral view, and genital plate as Fig. 5L.

**Paratype (1).** Similar to allotype except for the following: body length 25.5 mm; width 13.0 mm.

**Etymology.** The specific epithet *wayuorum* refers to the Wayuu indigenous group inhabiting the Guajira Peninsula.

**Distribution.** *Coelosis wayuorum* sp. n. is known only from one locality in Macuira National Park, Colombia (Fig. 8).

**Temporal distribution.** Three specimens collected in September 2014, during the dry season.

Life history. The type material was collected at night with lights during the dry season.

# The following are new biological data and distribution records for *Coelosis* species in Colombia

**Coelosis bicornis**. Vichada, Municipio de Puerto Carreño, Vereda La Esmeralda. El Tomo. 5.554252 Lat. -68.467042 Long. 81 msnm. Trampa de Luz Negra. 31 de marzo a 9 de abril de 2017. J. C. Neita, A. Lopera & J. Cárdenas [IAvH-E-198889 (1♂) and IAvH-E-198890 (1♀)].

Adults of *C. bicornis* have been captured in nests of *Atta laevigata* (Smith, 1858) (Hymenoptera: Formicidae) in the Orinoco basin.

**Coelosis biloba.** Antioquia, Carmen de Viboral, El Porvenir, Finca La Samaria, bosque de restauración, 05°53'15.2"N; 075°11'11.8"W. 1000–1100. WGS84 2016 11 30. A. Lopera & J. Cárdenas [IAvH-E-198891 (13), IAvH-E-198892 (12), IAvH-E-198893 (12)]. **Chocó**, Quibdó, Corregimiento Tutunendó. Finca cerca al pueblo. 5°44,58'N; 76° 32,043'W. 68 m snm. Trampa de Luz Negra. Abril de 2000. J. C. Neita Leg. [IAvH-E-198893 (13), IAvH-E-198894 (13), IAvH-E-198895 (13), IAvH-E-198896 (13), IAvH-E-198897 (12), IAvH-E-198898 (12), IAvH-E-198899 (12)]. **Chocó**, Quibdó, Corregimiento de Pacurita. Pueblo. 5°41'N; 76°40'W. 43 m snm. Trampa de Luz Negra. Febrero de 2001. J. C. Neita Leg. [IAvH-E-198900 (13), IAvH-E-198901(13), IAvH-E-198902 (12)]. **Chocó**, Quibdó. Corregimiento de Tutunendó, Estación Biológica Ambiental IIAP. 5°40'23.04"N; 76°33'42.38"W. 82 m alt. Ene. 2010. J. C. Neita Leg. [IAvH-E-198904 (13), IAvH-E-198905 (13), IAvH-E-198906 (13), IAvH-E-198907 (13), IAvH-E-198908 (12), IAvH-E-198909 (12), IAvH-E-198907 (13), IAvH-E-198908 (12), IAvH-E-198909 (12), IAvH-E-198907 (13), IAvH-E-198908 (12), IAvH-E-198909 (13), IAvH-E-198905 (13), IAvH-E-198901 (13), IAvH-E-198907 (13), IAvH-E-198908 (12), IAvH-E-198909 (12), IAvH-E-198907 (13), IAvH-E-198908 (12), IAvH-E-198909 (12), IAvH-E-198907 (13), IAvH-E-198908 (12), IAvH-E-198909 (12), IAvH-E-198908 (12), IAvH-E-198909 (12), IAvH-E-198910 (12), IAvH-E-198911 (12),].

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RESEARCH ARTICLE



## A new species of the rare, deep-sea polychaete genus Benthoscolex from the Sea of Kumano, Japan (Annelida, Amphinomidae)

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## Abstract

A new species of amphinomid polychaete, *Benthoscolex seisuiae* **sp. n.**, is described from the Sea of Kumano, Japan, from depths of 487–596 m. The species is distinguishable from its congeners by the following features: i) palps 1.8 times as long as lateral antennae; ii) branchiae do not reach to the tip of the notochaetae. This is the first record of *Benthoscolex* from Japan. A partial mitochondrial cytochrome coxidase subunit I gene sequence from the holotype of *B. seisuiae* **sp. n.** is provided for reliable species identification in the future.

### **Keywords**

Amphinomida, deep sea, new genus record, Polychaeta, polychaetes, taxonomy

## Introduction

Marine annelids in the family Amphinomidae are commonly known as fireworms, characterized by having defensive, dorsally-oriented, calcareous chaetae that are thought to be used to inject a venomous substance into predators (Verdes et al. 2017). The family consists of approximately 180 nominal species in 22 genera (Borda et al. 2012; Barroso et al. 2017; Sun and Li 2017), mostly distributed in shallow and tropical waters (Barroso et al. 2017).

One genus, *Benthoscolex* Horst, 1912, is rare and known mainly from deep-sea substrates. Previous collection records of the genus are limited to low latitude areas (Horst 1912; Monro 1937; Hartman 1942; Fauvel 1953; Salazar-Vallejo 1999; Wehe and Fiege 2002). The genus consists of two species: *B. coecus* Horst, 1912 and *B. cubanus* Hartman, 1942. Previous studies have reported *B. coecus* from the Red Sea to the SW Pacific Ocean (Horst 1912; Monro 1937; Fauvel 1953; Wehe and Fiege 2002) and *B. cubanus* from the Caribbean Sea (Hartman 1942). The main diagnostic features of the genus are: 1) caruncle consisting of three ridges and 2) branchiae are absent at least in the first five chaetigers. In Japan, several amphinomids have been reported from the deep sea (e.g., Imajima 2001, 2005, 2006, 2011), but there is no record of *Benthoscolex* species from Japanese waters.

During the research cruise No. 1722 by TRV *Seisui-maru*, we collected three specimens of *Benthoscolex*. We describe the specimens here as a new species and provide a COI sequence as a DNA barcode of the species. This is the first report of *Benthoscolex* from Japan.

## Materials and methods

Fresh specimens were collected by beam trawl from the Sea of Kumano, Japan (34°00.992'N to 33°55.258'N, 136°27.720'E to 136°26.650'E) from 487–596 m depth. The live specimens were fixed in 70% ethanol. After preservation, these specimens were observed with a Nikon SMZ1500 dissecting microscope and OLYMPUS BX51 compound microscope, and photographed with a Nikon D5200 digital camera. All of the material has been deposited in the National Museum of Nature and Science, Tsukuba (**NSMT**). We followed the morphological terminology of Barroso et al. (2017) in the taxonomic description below.

DNA extraction and sequencing for a partial region of mitochondrial cytochrome *c* oxidase subunit I (COI) gene were carried out following the method of Jimi and Fujiwara (2016). The newly obtained sequence data has been deposited in the DNA Data Bank of Japan (**DDBJ**).

## **Systematics**

## Family Amphinomidae Lamarck, 1818

[Japanese name: umikemushi-ka]

### Genus Benthoscolex Horst, 1912

[New Japanese name: mitsu-one-umikemushi-zoku]

**Diagnosis.** Body fusiform, flat. Eyes absent. Caruncle consisting of three ridges without ornamentation. Branchiae absent at least in first five chaetigers, dendritically branched. Dorsal and ventral cirri occur singly on the notopodium and neuropodium.

### Benthoscolex seisuiae sp. n.

http://zoobank.org/BC2C42AA-5761-44F6-89AE-F1D2B7260BBC Figs 1–2 [New Japanese name: Seisui-mitsu-one-umikemushi]

**Material examined.** Holotype: NSMT-Pol H-676, 21 mm long, 5 mm wide (without chaetae, at widest chaetiger), 29 chaetigers, female, the Sea of Kumano, 487–596 m depth, 8 November 2017, collected by NJ (left parapodium of chaetiger 15 was dissected for DNA extraction). Paratypes: NSMT-Pol P-677, two specimens, 19–28 mm long, 4–7 mm wide (without chaetae, at widest chaetiger), 29 chaetigers, female, collection data is the same as that of the holotype.

Sequence. LC360809, COI gene, 507 bp, determined from holotype.

**Description.** Body flat, tapered in anterior and posterior regions, whitish both in life and after fixation; pair of brown longitudinal lines on ventral middle line; no pigmentation on dorsal surface (Fig. 1A). Body surface smooth.

Prostomium triangular; eyes absent. Pairs of lateral antennae and palps present, conical, smooth; palps 1.8 times as long as lateral antennae. Median antenna present, conical, as long as lateral antennae (Figs 1B, 2A). Caruncle consists of three longitudinal ridges, without ornamentation, extends to chaetigers 1–2 (depending on fixation), unattached in posterior part (Fig. 2A). Mouth composed of chaetigers 1–2. Pharynx eversible with black pigmentation.

Parapodia biramous, notopodia and neuropodia clearly separated (Fig. 2B). Dorsal and ventral cirri occur singly on notopodium and neuropodium, conical, whitish, arising from body wall, present in all chaetigers. Branchiae present on chaetiger 6 or 7 and succeeding posterior chaetigers: anterior ones simple, conical lobes; gradually increasing in number and size posteriorly (Fig. 1B, C), branched from base; filaments digitiform, 8–10 filaments per branchia in middle body chaetigers, 15–18 filaments



**Figure 1.** *Benthoscolex seisuiae* sp. n., holotype (NSMT-Pol H-676). **A** whole body, dorsal view **B** anterior end, dorsal view **C** median body, dorsal view **D** posterior end, dorsal view. Scale bars: 5 mm (**A**); 1 mm (**B**); 3 mm (**C–D**).

per branchia in posterior chaetigers; branchiae in posterior chaetigers differ in size between specimens, but never reaching to tip of notochaetae (Fig. 1D).

Notochaetae contain three types: i) harpoon chaetae, with serrations limited to one side (Fig. 2C); ii) bifurcate chaetae with weakly serrated or non-serrated short tip (Fig. 2D); iii) bifurcate chaetae, with long serrated tip (Fig. 2E). Neurochaetae contain two types: i) bifurcate chaetae, with weakly serrated or non-serrated short tip (Fig. 2F); ii) bifurcate chaetae, with long serrated tip (Fig. 2G). Neurochaetae longer than notochaetae.

Anus opening dorsally on terminal chaetiger; anal papilla absent (Fig. 1D).

**Etymology.** The species is named after the TRV *Seisui-maru*. The type specimens from the Sea of Kumano were collected by beam trawl gear of the ship. The specific name is a noun in the genitive case.

**Confirmed distribution.** Only known from the type locality, the Sea of Kumano, Japan, 487–596 m depth.

**Remarks.** *Benthoscolex seisuiae* sp. n. can be discriminated from *B. coecus* and *B. cubanus* by the following features: i) palps 1.8 times as long as lateral antennae (vs. same length as lateral antennae in *B. cubanus*; 2.0 times as long as lateral antennae in *B. coecus*), and ii) branchiae do not reach to tip of notochaetae (vs. extending beyond tip of notochaetae in *B. coecus*; they also do not reach to tip of notochaetae in *B. cubanus*). In addition, the tip of the bifurcate neurochaetae is reportedly serrated in *B. cubanus*,



**Figure 2.** *Benthoscolex seisuiae* sp. n., holotype (NSMT-Pol H-676). **A** anterior end, dorsal view **B** left parapodium of chaetiger 15, posterior view **C** harpoon notochaeta **D** bifurcate notochaeta with short tip **E** bifurcate notochaeta with long tip **F** bifurcate neurochaeta with short tip **G** bifurcate neurochaeta with long tip. Scale bars: 1 mm (**A–B**); 100 μm (**C–G**).

whereas it is only weakly serrated, or not serrated at all, in *B. seisuiae* sp. n., although chaetal serration is known to be variable in *Eurythoe* (Barroso and Paiva 2007).

*Benthoscolex cubanus* is reported to be endocommensal in the body cavity of the bathyal irregular sea urchin *Heterobrissus hystrix* (A. Agassiz, 1880) (Hartman 1942; Emson et al. 1993). *Benthoscolex seisuiae* sp. n. was collected by a beam trawl and found free living. In the same haul, 49 specimens representing five species of irregular sea urchins [*Brisaster latifrons* (A. Agassiz, 1898) (n = 13, NSMT E-10723–10724), *Brissopsis luzonica* (Gray, 1851) (n = 6, NSMT E-10721–10722), *Brissopsis* sp. (n = 1, NSMT E-10727), *Lovenia gregalis* Alcock, 1893 (n = 22, NSMT E-10719–10720), *Schizaster* sp. (n = 7, NSMT E-10725–10726)] were present and some were broken in the net. However, examination of body cavity in all but one specimen (used for spe-



**Figure 3.** Four of the five irregular sea urchin species that were contained in the same haul with *Benthoscolex seisuiae* sp. n., aboral view. **A** *Brisaster latifrons* (A. Agassiz, 1898), NSMT E-10723 **B** *Brissopsis luzonica* (Gray, 1851), NSMT E-10721 **C** *Lovenia gregalis* Alcock, 1893, NSMT E-10719 **D** *Schizaster* sp., NSMT E-10725. Scale bars 1 cm.

cies identification and photography, Fig. 3) for each species revealed no commensal *Benthoscolex* worms (A. Ogawa pers. obs.); *Brissopsis* sp. was not examined because it was represented by only one specimen. Therefore, whether the new species is also endocommensal in sea urchins or not cannot be ascertained at the moment. Future studies are required to confirm the present observations of a free-living lifestyle in the new species.

## Key to species of Benthoscolex Horst, 1912

1	Posterior branchiae extend beyond tip of notochaetae	••••
	B. coecus Horst, 19	12
_	Posterior branchiae do not reach to tip of notochaetae	.2
2	Palps 1.8 times as long as lateral antennae	n.
_	Palps as long as lateral antennae	42

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RESEARCH ARTICLE



## First record of larvae of the water mite Hydrachna processifera Piersig, 1895 from Turkey (Acari, Hydrachnidia, Hydrachnidae)

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## Abstract

Larvae of water mite *Hydrachna processifera* Piersig, 1895 (Acari, Hydrachnidiae) were reported on diving beetles *Dytiscus marginalis* Linnaeus, 1758 (Coleoptera, Dytiscidae) from Turkey. The redescription of the larva was made. Earlier, the larva *H. processifera* was described as *H. inermis*, but it was subsequently synonymized with *H. processifera*. The larva of *H. processifera* is a new record for the Turkish fauna. All larvae of *H. processifera* were found on the mesosternum of the one specimens (prevalence = 16.7%).

## Keywords

Dytiscus marginalis, Hydrachna inermis, parasitism, water beetles, water mites

## Introduction

Mite taxonomy issues continue to pose some difficulties, which causes many synonymic names of particular species. The situation is much more difficult because of the presence of pre-adult stages such as larvae and deutonymphs. For instance, *Hydrach*- *na inermis* Piersig, 1895 has been synonymized with *H. processifera* Piersig, 1895 by Davids et al. (2005, 2007). Larvae of *H. inermis* were described by Sparing (1959) and Wainstein (1980) based on variable and questionable features of adults and these larvae were synonymized with *H. processifera* as well (Davids et al. 2005, 2007). All subsequent information about the parasitic behavior of *H. inermis* on Dytiscidae (Sparing 1959, Zawal 2002) should be recognized as *H. processifera*.

Larvae of water mites of the genera *Hydrachna*, *Eylais*, *Limnochares*, and *Acher-ontacarus* are ectoparasites on aquatic Hemiptera and aquatic Coleoptera (Reilly and McCarthy 1993, Biesiadka and Cichocka 1994, Cichocka 1995, Benfatti and Gerecke 1999, Zawal 2002, 2003a, 2003b, Ihle and McCreadie 2003, Fairn et al. 2008, Zawal et al. 2013, Aykut et al. 2016, Aykut and Esen 2017).

Parasitizing larvae of *H. processifera* (as *H. inermis*) were reported on Dytiscidae and Hydrophilidae in previous studies (Piatakov 1915a, 1915b, Brumpt 1929, Davids 1969, Zawal 2002). Zawal (2002) reported that *H. inermis* occurred as the most frequent parasites of *Dytiscus* (*D. circumcinctus* (Ahrens, 1811), *D. dimidiatus* Bergsträsser, 1778, and *D. marginalis* Linnaeus, 1758). In Turkey, studies on larvae of water mite are not advanced and only several studies were published (İncekara and Erman 2008, Taşar et al. 2012, Zawal et al. 2013, Aykut et al. 2016, Aykut and Esen 2017). In Turkey only six species of the genus *Hydrachna* (*H. conjecta* (Koenike, 1895), *H. globosa* (De Geer, 1778), *H. leegei* (Koenike, 1895), *H. orientalis* (Thon, 1905), *H. processifera* (Koenike, 1903), and *H. skorikowi* (Piersig, 1900)) were previously known (Erman et al. 2010). This study contributes to larval morphology of *H. processifera* and its parasitization on *Dytiscus marginalis*.

## Materials and methods

Parasitized specimens of *Dytiscus marginalis* were collected from a small pond supplied by a small water source in the plateau near Çayıryolu village of Varto district (39°09'23"N, 41°34'56"E; 20.08.2014) in the Eastern Anatolia Region of Turkey (Fig. 1). The coordinates and altitude information of the locality were taken directly from a handheld GPS tool (Magellan Explorist 610). The beetles were collected with a net of mesh size 0.5 mm diameter. Specimens were fixed and preserved in 70% ethyl alcohol solution at the collection site. The clay and muddy substance on their surfaces was brushed off with a small paint brush in the laboratory and each specimen was checked for the presence of water mites under a stereomicroscope. Beetle species were identified according to Friday (1988), Nilsson and Holmen (1995), Pederzani (1995), and Nilsson (1996) and mite larvae according to Wainstein (1980). Photographs were taken using stereo microscope (Z16 APO; Leica, Wetzlar, Germany) equipped with an HD camera (Leica MC170), and with a scanning electron microscope (Quanta 250 FEG; FEI, Eindhoven, Netherlands). The examined material is deposited in the private collection of the first author, at Dicle University, Diyarbakır, Turkey.

The following abbreviations are used: Cx - coxa; L - length; W - width.



Figure 1. Collecting site of Dytiscus marginalis infected with Hydrachna processifera larvae.

## Results

In total, six specimens of *D. marginalis* including four females and two males were collected. Of these, only one female specimen was infected (prevalence = 16.7%). Except *D. marginalis*, a total of 37 specimens belongs to four genera and seven species (*Agabus biguttatus*, *A. bipustulatus*, *A. conspersus*, *Colymbetes fuscus*, *Hydroporus pubescens*, *H. tesellatus*, *Platambus maculatus*). None of them was positive with regards to water mites.

On *D. marginalis*, eleven larvae of *H. processifera* were observed (Fig. 2). All larvae were found on the surface of mesosternum of the beetle body and they were small, 0.15–0.40 mm. The idiosoma are egg-shaped, with the integument striated, and the dorsal shield is very large, covered whole idiosoma, with the integument pointed (Figs 3, 4). There are three pairs of coxal plates located on the proximal half of the idiosoma, and all of they are wider than long. The anterior coxa bears two setae, the medial coxa is without seta, and the posterior coxa has only one seta (Fig. 4). Gnathosomal sucker has a large disk, tibiotarsal claws two in number and the same size, weakly bent, five tibiotarsal spines, three of them roughly barbed (Figs 3, 4). The body sizes of the larvae of *H. processifera* (N = 3) are as follows in  $\mu$ m – idiosoma: L/W 290–300/230–233; dorsal shield: L/W 245–296/170–180; coxal plates: Cx-1 L/W 75–80/38–43, Cx-2 L/W 79–86/33–37, Cx-3 77–84/40–45; gnatosoma; L/W 200–203/148–150; pedipalpal segments: femur L/W 68–70/36–38, genu L 16–17, tibiotarsus L 58–61.



Figure 2. SEM (A, B) and stereomicroscope (C, D) images of *Dytiscus marginalis* infected with larvae of water mites *Hydrachna processifera*.



**Figure 3.** SEM images of larvae of *Hydrachna processifera*. **A** dorsal view **B** ventral view **C** ventral view of gnatosoma **D** lateral view of palp.



**Figure 4.** Larvae of *Hydrachna processifera*: **A** dorsal shield **B** dorsal view of gnatosoma **C** ventral view **D** palp, medial view **E** palp, lateral view **F** I leg, tarsus **G** II leg, tarsus. Scale bars 100 μm.

## Discussion

*Hydrachna inermis* was described from a site in Germany (Piersig 1895, 1897, 1899). Later it was recorded in various parts of Europe but without clearly defined diagnostic features; it was synonymized with *H. processifera* by Davids et al. (2005, 2007). *Hydrachna processifera* is the only species of genus *Hydrachna* which attaches to its hosts on the external integument; all other species attach the under elytra (Zawal 2002). In this study, all larvae of *H. processifera* were found on mesosternum. Zawal (2002) stated that the greatest numbers of larvae were found on the prosternum (40.3%), followed by the mesosternum (20.8%), and the least number on the metasternum (19.5%), and that they infested three species of *Dytiscus (D. circumcinctus, D. dimidiatus, D. marginalis*). For the three species of water beetles, *D. marginalis* was the least infected (prevalence = 0.7%). In the present study, the prevalence (16.7%) and intensity of infestation (11 individuals) of *D. marginalis* was higher than of *D. marginalis* in Zawal's research and similar to prevalence and intensity of infestation of *D. circumcinctus* (Zawal 2002). Of course, the data obtained here should be approached with great caution, as they are based on a very small number of observations. The present study confirms a low prevalence and intensity of infestation of water beetles and water bugs found by other authors (Zawal 2002, 2003b, Biesiadka and Cichocka 1994) compared to dragonflies (Baker et al. 2008, Zawal and Szlauer-Łukaszewska 2012, Zawal and Buczyński 2013, Zawal et al. 2017) but similar to flies and caddisflies (Fairchild and Lewis 1987, Martin et al. 2010, Buczyńska et al. 2015, Stryjecki et al. 2015).

The small size all of water mite larvae of *H. processifera* confirms that the reproductive time is during summer. Wainstein (1980) noted that oviposition of this species takes place in July and developing requires 2–4 weeks in Russia. Zawal (2002) also reported greatest number of small and median sized larvae *H. processifera* in summer-autumn of Poland.

The water mite larvae collected from Muş Province were identified as *H. processifera* on the basis of shape of idiosoma and gnatosoma, and the shape of coxae and its setation. All tibiotarsal claws were large and of equal size. Unlike other larvae of water mites attached under elytra, larvae of water mites *H. processifera* are attached to outer surface of beetle bodies (Fig. 2). The record confirms the presence of this widely distributed, Palearctic species in the south-eastern part of its range (Turkey).

The redescribed of *H. processifera* is identical to Wainstein's (1980) *H. inermis* larva in all features excluding length of pedipalpal femur and genu. The redescription add new features as: striated integument and pointed dorsal shield; and some measurements.

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RESEARCH ARTICLE



## Four new species of Fissocantharis Pic, 1921 (Coleoptera, Cantharidae) from China

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### Abstract

Four new species of *Fissocantharis* Pic, 1921 are described: *F. securiclata* **sp. n.** (China: Zhejiang, Anhui), *F. maculicollis* **sp. n.** (China: Zhejiang), *F. hainana* **sp. n.** (China: Hainan) and *F. laticollis* **sp. n.** (China: Hainan), which are illustrated with the habitus and aedeagus of the male, the abdominal sternite VIII and the internal genitalia of the female. A key to the species from southeast China is provided.

### Keywords

East Asia, description, Cantharinae, taxonomy

## Introduction

The taxonomy of the Chinese species of *Fissocantharis* Pic, 1921 (sensu Yang et al. 2009) was studied mainly by Wittmer in the last half century. Among his publications, the one, in which Wittmer (1988) revised the species from mainland China and adjacent areas, is especially useful for our study of species from southeast China. We recently added some species from Fujian, Zhejiang, Hunan, Anhui, Guangxi, Guangdong and Hainan (Yang and Yang 2009, Yang et al. 2014, 2015a, b). In the present

study, four new species were discovered and described here under the names *F. securiclata* sp. n. (China: Zhejiang, Anhui), *F. maculicolliss*p. n. (China: Zhejiang), *F. hainana* sp. n. (China: Hainan) and *F. laticollis* sp. n. (China: Hainan).

## Material and methods

The material is preserved in the following collections: Institute of Zoology, Chinese Academy of Sciences, Beijing, China (IZAS); Museum of Hebei University, Baoding, China (MHBU); and the private collection of Mr. Andreas Pütz, Germany (CoAP).

Genitalia of both sexes and abdominal sternites VIII of females were dissected and cleared in 10% KOH solution, and female genitalia were dyed with hematoxylin. Habitus photos were taken by a Leica M205A stereomicroscope, multiple layers were stacked using Combine ZM (Helicon Focus 5.3). Line drawings were made using a camera lucida attached to a Nikon SMZ1500 stereomicroscope, then edited in Corel-DRAW 12 and Adobe Photoshop 8.0.1.

Complete label data in Chinese are transliterated for type specimens, and quotation marks are used for labels in English. Body length was measured from the anterior edge of the clypeus to the elytral apex and body width across the humeri of elytra. Morphological terminology of female genitalia follows Brancucci (1980). The specimens are prepared with the antennae extended posterad and photographed in dorsal view (Fig. 1). The lateral edge is the narrow surface of the antenna seen in this position from lateral view.

## Taxonomy

#### Key to the species of Fissocantharis (males) from southeast China

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5	Pronotum wider than long, about 1.2 times as wide as long
-	Pronotum uniformly black 7
_	Pronotum uniformity black marking on disc <i>F. maculicollis</i> sp. n.
7	Head uniformly yellow
8	Maxillary palpomeres II–IV dorsoventrally flattened and widened, II convex at basal part of dorsal side
_	Maxillary palpomeres II–IV slender, not flattened or widened <i>E pallidiceps</i> (Pic. 1911)
9	Middle antennomeres with longitudinal costa-like ridges along lateral edges
-	Middle antennomeres with smooth impressions or scar-like bulges along lateral edges
10	Antennae slightly cylindrically thickened, antennomeres III–IX with longitudi- nal ridges along lateral edges; aedeagus: conjoint dorsal plate of parameres longer than ventral process of each paramere
_	Antennae slightly dorsoventrally flattened, antennomeres III–VIII with lon- gitudinal ridges along lateral edges; aedeagus: conjoint dorsal plate of para- meres shorter than ventral process of each paramere
	<i>F. sexcostata</i> Y. Yang & X. Yang, 2015
11 _	Middle antennomeres with oblong scar-like bulges along lateral edges 12 Middle antennomeres with longitudinal or oblong or round impressions
12	Aedeagus: conjoint dorsal plate of parameres with apical edge rounded in mid- dle, ventral processes abruptly narrowed apically <i>F. gracilipes</i> (Pic, 1927)
_	Aedeagus: conjoint dorsal plate of parameres with apical edge acute in mid- dle, ventral processes evenly narrowed apically
	<i>F. eschara</i> Y. Yang & X. Yang, 2015
13 -	Antennomeres III–X parallel-sided <i>F. sinensomima</i> Y. Yang & X. Yang, 2015 Antennomeres III–X slightly dorsoventrally flattened and obliquely widened apically
14	Aedeagus: conjoint dorsal plate of parameres greatly reduced, with apical edge nearly horizontally aligned with bases of ventral process of each paramere <i>E hasilaris</i> X Yang & X Yang 2015
_	Aedeagus: conjoint dorsal plate of parameres moderately developed, with apical edge distinctly protuberant over bases of ventral process of each paramere
15	Aedeagus: conjoint dorsal plate of parameres about half length of ventral process of each paramere, nearly parallel-sided at basal part
_	<i>F. sinensis</i> (Wittmer, 1988) Aedeagus: conjoint dorsal plate of parameres about one-third length of ven- tral process of each paramere, tapered apically <i>F. hainana</i> sp. n.

16	Antennomeres III-IV or to V deformed, cylindrically thickened or excavate 17
-	Antennomeres III-VIII or to IX, X, XI deformed, at least one antennomere
	not parallel-sided, or excavate or emarginate
17	Maxillary palpomeres II–III excavate18
-	Maxillary palpi not excavate19
18	Antennomere IV with two projections at basal part (Yang et al. 2015b: fig. 7) .
	<i>F. biprojicientis</i> Y. Yang & X. Yang, 2015
_	Antennomere IV unlike above, without projections (Wittmer 1988: fig. 4)
	<i>F. bidifformis</i> (Wittmer, 1988)
19	Antennomere III excavate along the whole length of lateral edge, IV distinctly
	wider than III (Wittmer 1988: fig. 1)F. similis (Wittmer, 1951)
_	Antennomere III excavate at apex, IV as wide as III (Wittmer 1988: fig. 2)
	<i>F. tridifformis</i> (Wittmer, 1988)
20	Antennomere XI knife-like, with lateral edge triangularly protuberant near
	basal one-fourth part, obliquely narrowed apically, acute at apex (Wittmer
	1988: fig. 12)
_	Antennomere XI parallel-sided
21	Antennomeres XI about one-third longer than X
	<i>F. angusta</i> (Fairmaire, 1900)
_	Antennomeres XI about as twice long as X
22	Antennae and legs mostly black; aedeagus: conjoint dorsal plates of parameres
	with apical edge straight
_	Antennae and legs mostly orange; aedeagus: conjoint dorsal plates of para-
	meres with apical edge rounded F. flavimembris (Wittmer, 1951)
23	Antennomere IX distinctly lengthened and widened, about twice as long as
	wide
_	Antennomere IX unlike above25
24	Antennomere IX with lateral edges triangularly protuberant and slightly
	curled dorsally in middle (Yang et al. 2015b: fig. 2A)
	<i>F. novemexcavata</i> (Wittmer, 1951)
_	Antennomere IX parallel-sided (Yang et al. 2015b: fig. 2F)
	F. novemoblonga Y. Yang & X. Yang, 2015
25	Antennomere IV distinctly cylindrically thickened and excavate along the
	whole length of lateral edge, width wider ventrally than dorsally, V–X with api-
	cal angles truncate and finely bordered (Yang and Yang 2009: figs 3-6)
	<i>F. imparicornis</i> (Y. Yang & X. Yang, 2009)
_	Antennomeres IV–X unlike above
26	Antennomeres III–VIII each emarginate at apical part of lateral edge (Witt-
	mer 1988: fig. 16) <i>F. multiexcavata</i> (Wittmer, 1988)
_	Antennomeres III–VIII unlike above27
27	Antennomeres VIII or and VII projecting laterally
_	Antennomeres VII and VIII unlike above

28	Antennomere VIII with apical angle strongly projecting laterally, III-VIII
	minutely serrated along lateral edges (Wittmer 1988: fig. 13)
	<i>F. flavicornis</i> (Gorham, 1889)
_	Antennomeres VII-VIII with apical angles moderately projecting laterally,
	III-VIII not serrated along lateral edges (Wittmer 1988: fig. 14)
29	Antennomeres III-X obliquely widened apically (Wittmer 1989: fig. 8); ae-
	deagus: conjoint dorsal plate of parameres greatly reduced, with apical edge
	nearly horizontally aligned with bases of ventral processes
	<i>F. liuchowensis</i> (Wittmer, 1989)
_	Antennomere III obliquely widened apically, IV–IX securiform, which abrupt-
	ly widened apically and rounded at apical angles (fig. 1); aedeagus: conjoint
	dorsal plate of parameres moderately developed, with apical edge distinctly
	protuberant over bases of ventral processes F. securiclata sp. n.

**Remarks.** In the key, *F. bothridera* (Fairmaire, 1887) (located in Fujian) is not included, because it was described only on the female form, which is difficult to distinguish among other female *Fissocantharis* (Wittmer 1988). A checklist of the species from southeast China is provided after the descriptions, except for those from Guangxi, which have been listed by Yang et al. (2015b).

## Descriptions of new species

*Fissocantharis securiclata* Y. Yang & X. Yang, sp. n. http://zoobank.org/84FD8C2E-9B44-488C-9EBA-2CFF4205E868 Figs 1A, 2A–C, 4A, 5A

**Type material.** Holotype  $\Diamond$  (MHBU): CHINA, Anhui, Jixi, Qingliangfeng, 2–5. VI.2013, leg. J.S. Xu & C.X. Yuan. Paratypes: CHINA:  $1 \heartsuit$  (MHBU): same data as holotype; Zhejiang:  $1\heartsuit$  (IZAS): "Tienmu Shan, 10.6.1936, O. Piel coll.";  $1 \diamondsuit$  (IZAS): "Tienmu Shan, 6.6.1936, O. Piel coll.";  $1 \circlearrowright$  (CoAP) "CHINA: Zhejiang [CH07-39], Hangzhou Pref., Tianmu Shan, 40 km WNW Linan, water reservoir, 30°20'56"N, 119°18'42"E, 300 m, plant refuse, litter from rock edges, 17.VI.2007, leg. A. Pütz".

Distribution. China (Anhui, Zhejiang).

**Description.** *Male* (Fig. 1A). Head orange, darkened at both sides of vertex, mouthparts orange, darkened at mandibular apices, terminal maxillary and labial palpomeres and antennae orange, darkened dorsally at antennomeres III–IX, pronotum, scutellum and elytra black, legs orange, darkened at tarsi, ventral surface of body black, yellow at posterior and sides of abdominal sternites. Body densely covered with short recumbent gray pubescence.



Figure 1. Male habitus (holotype), dorsal view: A *Fissocantharis securiclata* sp. n. B *F. maculicollis* sp. n. C *F. hainana* sp. n. D *F. laticollis* sp. n. Scale bars: 2.0 mm.

Head subquadrate, narrowed posteriorly behind eyes; eyes moderately projecting, head width across eyes greater than that of anterior edge of pronotum; apical maxillary palpomeres nearly long-triangular, widest at apical two-fifths, acute at apices; antennae extending to elytral midlength, antennomeres II about twice as long as wide, III obliquely widened apically, truncate at apical angles, about twice as long as wide, IV–IX securiform, abruptly widened apically and rounded at apical angles, IV and IX slightly longer than wide, V–VIII nearly as long as wide, X and XI nearly parallelsided, XI pointed at apices and about 1.5 times as long as X.

Pronotum subquadrate, about 1.1 times longer than wide, anterior edge arcuate, sides slightly diverging posteriorly, posterior edge nearly straight, anterior angles rounded, posterior angles nearly rectangular, disc slightly convex at posterolateral parts, surface finely and densely punctate.

Elytra about 3.0 times longer than wide, 4.0 times longer than pronotum, width at humeri greater than posterior edge of pronotum, sides nearly parallel, surface slightly more coarsely and densely punctate than pronotum.

Legs with all tarsal claws bifid, each with lower projection as long as upper one.

Aedeagus (Fig. 2A–C): ventral process of each paramere wide, slightly narrowed apically and hooked at apex; conjoint dorsal plate of parameres moderately developed, about one-third length of ventral process of each paramere, slightly narrowed apically, with apex medially concave.

*Female.* Similar to male, but vertex black, antennae black, antenomeres I and II orange, filiform and simple, extending to basal third of elytra. Abdominal sternite VIII (Fig. 4A) nearly straight at posterior edge, lateral angles obtuse.

Internal genitalia (Fig. 5A): vagina abruptly extended apically as a short and thick duct at ventroapical portion; diverticulum and spermathecal duct arising from end of duct of vagina; diverticulum evenly thin, spiraled and long; spermathecal duct slightly shorter and thicker than diverticulum; spermatheca nearly as long as and slightly thicker than diverticulum, with a moderately long thin accessory gland, which is longer than spermatheca.

Body length: 7.9-9.0 mm; width: 1.7-2.0 mm.

**Etymology.** The specific name is derived from Latin *securiclatus* (axe-like), referring to its antennomeres IV–IX securiform in male, which abruptly widened apically and rounded at apical angles.

**Diagnosis.** Head mostly orange, pronotum and elytra uniformly black; male antennomeres IV–IX securiform, abruptly widened apically and rounded at apical angles; aedeagus: conjoint dorsal plate of parameres moderately developed, about one-third length of ventral process of each paramere, slightly narrowed apically, with apical edge medially concave.

**Remarks.** No other species has the male antennomeres IV–IX securiform as in this species. This species is similar to *F. pallidiceps* (Pic, 1911) in the body coloration, but can be easily distinguished by the male antennae, of which the antennomeres III–IX are widened apically, while simple filiform in *F. pallidiceps* (Wittmer 1988: fig. 6); aedeagus: the conjoint dorsal plate of parameres is slightly narrowed apically, with the



**Figure 2.** Aedeagus (**A**, **D** ventral view **B**, **E** dorsal view **C**, **F** lateral view): **A–C** *Fissocantharis securiclata* sp. n. **D–F** *F. maculicollis* sp. n. Scale bars: 1.0 mm. (dp: conjoint dorsal plate of parameres; vp: ventral process of each paramere).

apex medially concave, while distinctly narrowed apically, with the apex straight in *F. pallidiceps* (Wittmer 1988: fig. 26). It also resembles *F. liuchowensis* (Wittmer, 1989), but can be differentiated by the male antennae, of which the antennomeres IV–IX are abruptly widened apically and rounded at apical angles, while obliquely widened apically in *F. liuchowensis* (Wittmer 1989: fig. 8); aedeagus: the conjoint dorsal plate of parameres is moderately developed, while greatly reduced in *F. liuchowensis* (Wittmer 1989: fig. 9).

## Fissocantharis maculicollis Y. Yang & X. Yang, sp. n.

http://zoobank.org/CA4F3AAE-29F9-4AC8-A554-F7853A89AA09 Figs 1B, 2D–F, 4B, 5B

**Type material.** Holotype  $\mathcal{J}$  (MHBU): CHINA, Zhejiang, Qingliangfeng, Longtangshan, 19.V.2011, leg. G.L. Xie. Paratypes: CHINA, Zhejiang:  $2\mathcal{J}\mathcal{J}$ ,  $1\mathcal{Q}$  (IZAS): Xitianmushan, Xianrending, 1500m, 6.VI.1998, leg. H. Wu;  $1\mathcal{J}$  (IZAS): Xitianmushan, Kaishanlaodian, 1050m, 30.V.1998, leg. M.S. Zhao;  $1\mathcal{J}$  (IZAS): Xitianmushan, Dahenglu, 1200m, 6.VI.1998, leg. M.S. Zhao;  $1\mathcal{J}$  (IZAS): Qingyuan, Baishanzu, 20.VIII.1993, leg. H. Wu.

## Distribution. China (Zhejiang).

**Description.** *Male* (Fig. 1B). Head yellow, vertex black, mouthparts yellow, darkened at mandibular apices, antennae black, antennomeres I and II yellow, prothorax yellow, pronotum with a large dark brown marking on disc, marking extending to posterior but anterior edge or sides, and its posterior part wider than anterior part, scutellum and elytra black, legs yellow, slightly darkened at tarsi, ventral surface of body black. Body densely covered with short recumbent gray pubescence.

Head subquadrate, narrowed posteriorly behind eyes; eyes strongly projecting, head width across eyes greater than that of anterior edge of pronotum; apical maxillary palpomeres nearly long-triangular, widest at apical two-fifths, acute at apices; antennae filiform and simple, extending to apical third of elytra, antennomeres II about 1.5 times as long as wide, III about 3.0 times as long as II, IV–X subequal in length, XI pointed at apices and slightly longer than X.

Pronotum subquadrate, about 1.1 times longer than wide, anterior edge rounded, sides slightly diverging posteriorly and sinuate, posterior edge nearly straight, anterior angles rounded, posterior angles nearly rectangular, disc convex at posterolateral parts, surface finely and densely punctate.

Elytra about 3.5 times as long as wide, 5.0 times as long as pronotum, width at humeri greater than posterior edge of pronotum, sides nearly parallel, surface slightly more coarsely and densely punctate than pronotum.

Legs with all tarsal claws bifid, each with lower projection as long as upper one.

Aedeagus (Fig. 2D–F): ventral process of each paramere wide, slightly narrowed apically and hooked at apex; conjoint dorsal plate of parameres moderately developed, about a half length of ventral process of each paramere, with apical edge rounded.

*Female.* Similar to male, but eyes only slightly projecting, antennae narrower and shorter, extending to basal third of elytra, pronotum slightly convex at posterolateral parts. Abdominal sternite VIII (Fig. 4B) widely and shallowly emarginate at middle of posterior edge, bottom of middle emargination slightly roundly protuberant in middle, lateral angles slightly acute.

Internal genitalia (Fig. 5B): vagina abruptly extended apically as a short and thick duct at ventroapical portion; diverticulum and spermathecal duct arising from end of duct of vagina; diverticulum thick at basal portion and thinned apically, spiraled and moderately long; spermathecal duct nearly as long as and thinner than basal portion of diverticulum; spermatheca much longer than and nearly as thick as apical portion of diverticulum, with a moderately long thin accessory gland, which slightly shorter than spermatheca.

**Variation within type series.** Male antennae mostly yellow in some throughout antennomere or ventrally. Body length: 6.2–7.4 mm; width: 1.2–1.6 mm.

**Etymology.** The specific name is derived from the Latin *macula* (marking) and *collum* (neck), referring to its pronotum with a black marking on disc.

**Diagnosis.** Elytra black, prothorax yellow, pronotum with a large dark brown marking on disc; male antennae filiform and simple; aedeagus: conjoint dorsal plate of parameres moderately developed, about a half length of ventral process of each paramere, with apical edge rounded.

**Remarks.** This new species is similar to *F. nigriceps* Y. Yang & Okushima, 2016 (located in Taiwan) in the body shape and male antennae, but can be distinguished from the latter by the following characters: the pronotum has a black marking on the disc and the aedeagus has a wider conjoint dorsal plate of parameres, while in *F. nigriceps*, the pronotum is uniformly orange and the aedeagus has a narrower conjoint dorsal plate of parameres (Li et al. 2016: figs 6G–I, 12A). It is also resembles *F. paulioincrassata* (Wittmer, 1951) in the male antennae, but differs in having a yellow pronotum, with a large black marking on the disc, while uniformly black in *F. paulioincrassata*; aedeagus: the conjoint dorsal plate of parameres is moderately developed, about half length of ventral process of each paramere, while slightly shorter, about one-third length in *F. paulioincrassata* (Wittmer 1988: 353).

## Fissocantharis hainana Y. Yang & X. Yang, sp. n.

http://zoobank.org/471AF09B-DEA7-437C-8CCE-D9D1928DE8A1 Figs 1C, 3A–C, 4C, 5C

**Type material.** Holotype  $\Diamond$  (IZAS): CHINA, Hainan, Diaoluoshan, 928m, 30.III.1980, leg. Y.X. Yang. Paratypes: CHINA, Hainan:  $1\Diamond$ ,  $1\Diamond$  (IZAS): same data as holotype;  $1\Diamond$  (IZAS): same locality, 31.III.2008, leg. L.Y. Jiang;  $1\Diamond$  (IZAS): Qiong-zhong, Wuzhishan, 800m, date and collector unknown;  $1\Diamond$  (IZAS): Qiongzhong, 800m, 5.IV.1980, collector unknown;  $1\Diamond$  (IZAS): Wuzhishan, 936.8m, 18.54°N,



Figure 3. Aedeagus (A, D ventral view B, E dorsal view C, F lateral view): A–C *Fissocantharis hainana* sp. n. D–F *F. laticollis* sp. n. Scale bars: 1.0 mm.

109.40°E, 2.IV.2008, leg. X. L. Chen; 1 $\bigcirc$  (IZAS): Wuzhishan, 708–1206m, 18.89°N, 109.69°E, 9.IV.2010, leg. K.Y. Zhang; 1 $\bigcirc$ , 1 $\bigcirc$  (IZAS): Qiongzhong, Limushan, 840m, 19.17°N, 109.72°E, 6.IV.2010, K.Y. Zhang; 1 $\bigcirc$ , 2 $\bigcirc$   $\bigcirc$  (IZAS): Baisha, Yinggeling, Nankai, 336m, 19.00°N, 109.22°E, 6.IV.2010, leg. M.Y. Lin; 3 $\bigcirc$   $\bigcirc$  (IZAS): "Hainan Prov., Lingshui, Diaoluoshan, Xin-an, 18.72510°N, 109.86861°E, 921m, 2007.03.25, Shi H.L., Yuan F. coll.".

Distribution. China (Hainan).

**Description.** *Male* (Fig. 1C). Body black, except mandibles brown, darkened at apices, prothorax orange, pronotum with a large black marking on disc, marking extending to anterior and posterior edges except at sides, and its posterior part wider than anterior part, elytra dark purple, with metallic reflection.

Head subquadrate, narrowed posteriorly behind eyes; eyes strongly projecting, head width across eyes wider than that of anterior edge of pronotum; apical maxillary palpomeres nearly long-triangular, widest at apical two-fifths, acute at apices; antennae nearly extending to elytral apices, antennomeres II nearly as long as wide, III–IX dorsoventrally flattened and obliquely widened apically, III about 3.0 times as long as II, IV–X subequal in length, X and XI parallel-sided, XI pointed at apices and slightly longer than X, III–XI each with a round or oblong smooth impression near middle of lateral edge.

Pronotum subquadrate, nearly as long as wide, widest near base, anterior edge rounded, sides sinuate, posterior edge nearly straight, anterior angles rounded, posterior angles nearly rectangular, disc convex at posterolateral parts, surface finely and densely punctate.

Elytra about 3.0 times longer than wide, 4.0 times longer than pronotum, width at humeri greater than posterior edge of pronotum, sides nearly parallel, surface slightly more coarsely and densely punctate than pronotum.

Legs with all tarsal claws bifid, each with lower projection as long as upper one.

Aedeagus (Fig. 3A–C): ventral process of each paramere wide, slightly narrowed apically and hooked at apex; conjoint dorsal plate of parameres moderately developed, about one-third length of ventral process of each paramere, with apical edge acute in middle.

*Female.* Similar to male, but eyes only slightly projecting, antennae filiform and simple, extending to midlength of elytra, pronotum moderately convex at posterolateral parts. Abdominal sternite VIII (Fig. 4C) widely and shallowly emarginate at middle of posterior edge, bottom of middle emargination nearly straight, lateral angles rounded.

Internal genitalia (Fig. 5C): vagina abruptly extended apically as a long and thick duct at ventroapical portion; diverticulum and spermathecal duct arising from end of long duct of vagina; diverticulum thin, spiraled and moderately long; spermathecal duct nearly as long as and slightly thicker than diverticulum; spermatheca longer and slightly thicker than diverticulum, with a very long and thin accessory gland, which much longer than spermatheca.




Figure 4. Abdominal sternite VIII of female, ventral view: A *Fissocantharis securiclata* sp. n. B *F. maculicollis* sp. n. C *F. hainana* sp. n. D *F. laticollis* sp. n. Scale bars: 1.0 mm.

**Variation within type series.** Pronotum orange in some, only darkened at anterior and posterior margins, elytra dark blue, or with weakly metallic reflection. Body length: 7.0–11.5 mm; width: 1.3–2.2 mm.

**Etymology.** The specific name is derived from the type locality, Hainan Province, China.

**Diagnosis.** Elytra dark purple, with metallic reflection; male antennae subfiliform, antennomeres III–IX dorsoventrally flattened and obliquely widened apically, III–XI each with a round or oblong smooth impression near middle of lateral edge; aedeagus: conjoint dorsal plate of parameres moderately developed, about one-third length of ventral process of each paramere, with apical edge acute in middle.

**Remarks.** The new species is similar to *F. langana* (Pic, 1923) (located in N. Vietnam), *F. langaniformis* (Wittmer, 1989) (N. Vietnam) and *F. sinensis* (Wittmer, 1988) in the body coloration and male antennae, but can be distinguished by the structure of the aedeagus: the conjoint dorsal plate of parameres is moderately developed, about one-third length of ventral process of each paramere, with the apical edge acute in the middle, while it is greatly reduced in *F. langana* (Wittmer 1988: fig. 29) and *F. langaniformis* (Wittmer 1989: fig. 10), and about half length of ventral process in *F. sinensis* (Yang et al. 2009: fig. 3A–C).

#### Fissocantharis laticollis Y. Yang & X. Yang, sp. n.

http://zoobank.org/573DA6BD-6CE7-4861-B8E5-29D720ABA13D Figs 1D, 3D–F, 4D, 5D

**Type material.** Holotype 3 (IZAS): CHINA, Hainan, Jianfengling, Tianchi, 24.III.2008, leg. Y.X. Yang. Paratypes: CHINA, Hainan: 633, 399 (IZAS): same data as holotype; 533, 19 (IZAS): same locality and collector, 812m, 23.III.2008; 433, 1099 (IZAS): Wenchang, Tongguling, 317m, 18.III.2008, leg. Y.X. Yang; 133, 4999 (IZAS): Wuzhishan, 708–1206 m, 18.89°N, 109.69°E, leg. M.Y. Lin; 19 (IZAS): Jianfeng, Tianchi, 750m, 19.III.1980, leg. S.Y. Wang; 133 (IZAS): Jianfeng, 760m, 20.III.1980, collector unknown; 133 (IZAS): Qiongzhong, Wuzhishan, 800m, 6.IV.1980, leg. S.Y. Wang; 233 (IZAS): Qiongzhong, 1840m, 5.IV.1980, collector unknown.

Distribution. China (Hainan).

**Description.** *Male* (Fig. 1D). Head orange, vertex black, mouthparts orange, darkened at mandibular apices, terminal maxillary and labial palpomeres, antennae black, antennomeres I and II orange at ventral sides, prothorax orange, pronotum with an inverted triangular black marking at middle of anterior part on disc, scutellum and elytra black, legs orange, darkened at tarsi, ventral surface of body black, yellow at posterior edges and sides of abdominal ventrites. Body densely covered with short recumbent gray pubescence.

Head rounded, slightly narrowed posteriorly behind eyes; eyes moderately projecting, head width across eyes slightly wider than that of anterior edge of pronotum; apical maxillary palpomeres nearly long-triangular, widest at apical two-fifths length, acute at apices; antennae filiform and slightly thickened, extending to apical third of elytra, antenomeres II slightly longer than wide, III about twice as long as II, IV–X subequal in length, XI pointed at apices and slightly longer than X.

Pronotum transverse, about 1.2 times wider than long, anterior edge rounded, sides sinuate, slightly converging posteriorly at anterior one-third length, then slightly diverging posteriorly at posterior part, posterior edge slightly arcuate, anterior angles subrounded, posterior angles nearly rectangular, disc slightly convex at posterolateral parts, surface finely and densely punctate.

Elytra about 3.5 times as long as wide, 4.5 times as long as pronotum, width at humeri slightly greater than posterior edge of pronotum, sides nearly parallel, surface slightly more coarsely and densely punctate than on pronotum.

Legs with all tarsal claws bifid, each with lower projection as long as upper one.

Aedeagus (Fig. 3D–F): ventral process of each paramere wide, narrowed apically and hardly hooked at apex; conjoint dorsal plate of parameres reduced, with apical edge deeply emarginate in middle.

*Female.* Similar to male, but eyes slightly projecting, head width across eyes nearly as wide as anterior edge of pronotum, antennae narrower and shorter, extending to midlength of elytra. Abdominal sternite VIII (Fig. 4D) with a pair of small bilobate protuberances in middle of posterior edge, lateral angles obtuse.

Internal genitalia (Fig. 5D): vagina abruptly extended apically as a short and thick duct; diverticulum and spermathecal duct arising from end of duct of vagina;



Figure 5. Female internal genitalia, lateral view: A *Fissocantharis securiclata* sp. n. B *F. maculicollis* sp. n. C *F. hainana* sp. n. D *F. laticollis* sp. n. Scale bars: 1.0 mm. (ag: accessory gland; di: diverticulum; eva: extension of vagina; sd: spermathecal duct; sp: spermatheca; ov: median oviduct; va: vagina).

diverticulum evenly thin, long and spiral; spermathecal duct shorter and thicker than diverticulum; spermatheca nearly as long as and slightly thicker than diverticulum, with a short thin accessory gland, which much shorter than spermatheca.

Body length: 4.0–5.5 mm; width: 0.8–1.1 mm.

**Etymology.** The specific name is derived from the Latin *latus* (wide) and *collum* (neck), referring to its pronotum wider than long.

**Diagnosis.** Elytra black; male antennae filiform and simple; pronotum transverse, about 1.2 times wider than long, sides sinuate, slightly converging posteriorly at anterior one-third length, then slightly diverging posteriorly at posterior part; aedeagus: conjoint dorsal plate of parameres reduced, with apical edge deeply emarginate in middle.

**Remarks.** This species could be distinguished by its pronotum, which is distinctly wider than long, while always longer than wide or subequal in length in other species of *Fissocantharis*. It is more similar to *F. imparicornis* (Y. Yang & X. Yang, 2009) (China: Hainan) in body coloration, but differs from the latter by the following characters: the male antennae are simple and the aedeagus has a reduced conjoint dorsal plate of parameres, while in *F. imparicornis*, the middle antennomeres of the male are deformed and the aedeagus has a well-developed conjoint dorsal plate of parameres.(Yang and Yang 2009: figs 3–6, 8).

# Other species from southeast China (except those from Guangxi)

## Fissocantharis acuticollis Y. Yang & X. Yang, 2014

*Fissocantharis acuticollis* Y. Yang & X. Yang, 2014, Yang et al. 2014: 53, figs 2, 5, 10, 20–22.

Distribution. China (Zhejiang, Fujian, Guangdong, Hunan).

# Fissocantharis bothridera (Fairmaire, 1887)

Rhagonycha bothridera Fairmaire, 1887: 125, 127. Rhagonycha nigricoloriceps Pic, 1928: 55. Synonymized by Wittmer 1988: 344. Micropodabrus bothriderus: Wittmer 1988: 344. Fissocantharis bothridera: Yang et al. 2009: 49.

Distribution. China (Fujian).

#### Fissocantharis flavimembris (Wittmer, 1951)

Podabrus flavimembris Wittmer, 1951: 96, fig. 1. Micropodabrus flavimembris: Wittmer 1988: 358. Fissocantharis flavimembris: Yang et al. 2009: 49.

Distribution. China (Fujian).

# Fissocantharis imparicornis (Y. Yang & X. Yang, 2009)

*Micropodabrus imparicornis* Y. Yang & X. Yang, 2009: 65, figs 1–9. *Fissocantharis imparicornis*: Yang et al. 2009: 49.

**Distribution.** China (Hainan).

#### Fissocantharis novemexcavata (Wittmer, 1951)

Podabrus novemexcavatus Wittmer, 1951: 97, fig. 3.
Podabrus novemexcavatus ab. testaceicollis Wittmer, 1951: 97, fig. 3. Synonymized by Kazantsev and Brancucci 2007: 257.
Micropodabrus novemexcavatus: Wittmer 1988: 359.
Fissocantharis novemexcavata: Yang et al. 2009: 49; Yang et al. 2015b: 363, figs 2A, 3A–C, 5A, 6A.

Distribution. China (Fujian).

## Fissocantharis novemoblonga Y. Yang & X. Yang, 2015

*Fissocantharis novemoblonga* Y. Yang & X. Yang, 2015, Yang et al. 2015b: 371, figs 1D, 2F, 4G–I, 5C, 6C.

Distribution. China (Zhejiang, Anhui).

# Fissocantharis pauloincrassata (Wittmer, 1951)

Podabrus pauloincrassatus Wittmer, 1951: 97. Micropodabrus pauloincrassatus: Wittmer 1988: 353. Fissocantharis pauloincrassata: Yang et al. 2009: 49.

Distribution. China (Fujian).

# Fissocantharis pallidiceps (Pic, 1911)

Rhagonycha pallidiceps Pic, 1911: 271. Micropodabrus pallidiceps: Wittmer 1988: 353. Fissocantharis pallidiceps: Yang et al. 2009: 49.

Distribution. China (Zhejiang).

### Fissocantharis pieli (Pic, 1937)

*Lycocerus pieli* Pic, 1937: 172. *Micropodabrus pieli*: Wittmer 1997: 312, figs 178–180. *Fissocantharis pieli*: Yang et al. 2009: 49; Yang et al. 2014: 47, figs 3, 6, 11–13.

Distribution. China (Zhejiang, Fujian).

#### Fissocantharis similis (Wittmer, 1951)

Podabrus similis Wittmer, 1951: 97. Micropodabrus similis: Wittmer 1988: 348. Fissocantharis similis: Yang et al. 2009: 49.

**Distribution.** China (Fujian).

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RESEARCH ARTICLE



# Contribution to the knowledge of Saprinus Erichson, 1834 of forensic relevance from Lebanon (Coleoptera, Histeridae)

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# Abstract

Many histerid beetles are necrophilous on carrion during both active and advanced stages of decomposition. In this study, 13 species of *Saprinus* were recorded on carrion from Lebanon, containing eight that are new for the Lebanese fauna. The following *Saprinus* species are newly recorded from Lebanon: 1) *Saprinus* (*S.*) *caerulescens caerulescens* (Hoffmann, 1803); 2) *S.* (*S.*) *calatravensis* Fuente, 1899; 3) *S.* (*S.*) *chalcites* (Illiger, 1807); 4) *S.* (*S.*) *godet* (Brullé, 1832); 5) *S.* (*S.*) *maculatus* (P. Rossi, 1792); 6) *S.* (*S.*) *strigil* Marseul, 1855; 7) *S.* (*S.*) *submarginatus* J. Sahlberg, 1913; and 8) *S.* (*S.*) *tenuistrius sparsutus* Solsky, 1876. The peak activity was recorded, key for the species is provided, and habitus images and male genitalia are illustrated in order to facilitate their taxonomic identifications. Saprinus species are diverse and common on animal carcass; they were likewise collected from a human cadaver in Lebanon. Preliminary comments on biology and distribution of the studied species are given. Our paper represents the first faunistic study on Histeridae from Lebanon. A rigorous research program regarding the biology of *Saprinus* in Lebanon and the neighbouring countries would greatly improve the knowledge of the diversity, activity, and possible forensic value of *Saprinus*.

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#### **Keywords**

Carrion, key to species, Lebanon, peak activity, Saprinus

## Introduction

Coleoptera (beetles) have proven to be useful in forensic investigations when fly larvae cannot be obtained or when they left the carcass and only beetles could be sampled (Zanetti et al. 2015). When examining the faunal succession of beetles on carcasses, the carrion beetles (Silphidae) are the first to be attracted, followed by the rove beetles (Staphylinidae) and clown beetles (Histeridae) (Su et al. 2013). Histerid beetles have received rather more attention in both ecological and taxonomic studies (three world catalogues published in the last 40 years, numerous revisions; for the references see Lackner et al. 2015). Regarding their presence on carrion, Histerids have been reported as the second after the Staphylinidae in the number of species on carrion during both the active and advanced stages of decomposition (Majka 2008; Su et al. 2013). Within the carrion community, histerid adults are regular visitors and they have been mentioned in forensic studies that use decomposing pig carcasses as experimental models as well as in real cases on human corpses (Bajerlein et al. 2011; Aballay et al. 2013). Histerids prey predominantly on dipteran eggs and larvae, though certain members of this family were also reported as predators on other beetles' larvae and even to practice cannibalism at low preferred prey densities (Byrd and Castner 2000; Kovarik and Caterino 2016).

Histerids are unusual among beetles in having just two larval instars. In the first instar the head capsule and sclerites harden and the larva becomes capable of killing and feeding on soft-bodied insects. Mandibles and terminal palpal segments darken and become visible through the egg chorion just before eclosion (Kovarik and Caterino 2016). They undergo complete metamorphosis and their larvae are predacious and feed on insect immature stages. The average development of this family from egg to adult is 20 days at 30 °C (Fakoorziba et al. 2017). Our study focused on adults of *Saprinus* and immature stages were unfortunately not collected, albeit we admit that their presence might be informative for the determination of the post-mortem interval. Based on our data collected from pig carcasses, autopsies, and field trips, *Saprinus* species were the most common and diverse within the histerids. This paper aims to clarify the diversity, abundance, and association of *Saprinus* species with decomposing carcasses in Lebanon.

In the Palaearctic Region, 357 species of the Saprininae subfamily have been reported so far (Lackner et al. 2015). With 116 species, members of the genus *Saprinus* Erichson, 1834 are the most numerous. Regarding the countries neighbouring Lebanon, 47 species of the Saprininae are known from Syria, with 29 species recorded from Israel (Lackner et al. 2015). Among Saprininae, *Saprinus* species show similar relative diversities (Syria: 26 species; Israel: 18 species). Lebanon, with only 11 species of reported Saprininae, is the poorest of the three countries. Hitherto, only 5 species attributed to the genus *Saprinus*: *S.* (*S.*) *aegialius* Reitter, 1884; *S.* (*S.*) *magnoguttatus* Reichardt, 1926; *S.* (*S.*) *prasinus prasinus* Erichson, 1834; *S.* (*S.*) *robustus* Krása, 1944

(=S. (S.) vermiculatus Dahlgren, 1964) and S. (S.) subnitescens Bickhardt, 1909 have been confirmed from Lebanon (Lackner et al. 2015). In Lebanon, there is a dearth of faunistic studies on Coleoptera in general, and Histeridae are no exception. In our present study, we focused on the *Saprinus* species that were collected from cadavers, and provide a key to identification of *Saprinus* from Lebanon.

#### Materials and methods

The majority of specimens were collected on a daily basis during the decomposition process (Table 1). They were collected from the soil under five pig carcasses and 14 pitfall traps placed in their vicinity in addition to the sampling of two specimens during autopsy in Beirut, Beirut district, Beirut Governorate [33°53'N, 35°31'E] of a human body brought from Arsal, Baalbek-Hermel Governorate, Baalbek district [34°10'N, 36°25'E]. One pig carcass was placed exposed in each of the five different localities during different seasons (Fig. 1): 1) Badghan, Aley District, Mount Lebanon Governorate [33°46'4.24"N, 35°41'14.78"E] at an altitude of 1174 m during summer (June and July); 2) Fanar, Matn District, Mount Lebanon Governorate [33°52'44"N, 35°34'04"] at an altitude of 250 m during spring (March and April); 3) Deir El-Ahmar, Baalbek-Hermel Governorate, Baalbek District [34°7'30.96"N, 36°7'22.04"E], at an altitude of 1040 m during summer (July) 3) Naas, Bikfaya Matn District, Mount Lebanon Governorate [33°54'42.4"N, 35°40'32.7"E] at the altitude of 1090 m during spring (April and May); and 5) Hasbaya, Natabtieh Governorate, Hasbaya District [33°23'52.35"N, 35°41.6'6.59"E], at an altitude of 750 m (July and August). In addition, a single pig head was placed in Fanar during the autumn season (September and October) and specimens were collected from pitfall traps surrounding it on a daily basis to compare the diversity of Saprinus during different weather conditions. Specimens from Hammana, Baabda District, Mount Lebanon Governorate [33°50'N, 33°44'E] at an altitude 1200 m and Sin El-Fil, Matn District, Mount Lebanon Governorate [33°52'N 35°32'E] at an altitude of 50 m were manually collected during field trips. Insects' activity on the pig carcasses was also reported during the night.

Four stages of decomposition were reported on the carcasses: 1) fresh stage; 2) bloat stage; 3) active decay stage; and, 4) advanced decay stage. Based on Matuszewski et al. (2010), the onset of the bloat stage was the first day when the inflation of the carcass was present. Before the inflation the carcass was considered fresh. The duration of the active decay was associated to the presence of Diptera larvae that fed on the carcasses. The onset of the advanced decay was assigned when Diptera larvae moved away to pupate, and soft tissues were absent on the carcasses (Table 1).

General observation and dissection were carried out using stereomicroscope Nikon SMZ1500. Without genital extraction, the males of *Saprinus* species can be usually recognized through the examination of the anterior tarsal setae, which are expanded and lamellate, whereas they are unexpanded and pointed in female. Often the males possess a longitudinal depression on the metaventrite and occasionally also a single or two tiny tubercles on the apical metaventral margin. Male genitalia were

			Stages of decomposition			
Locality	Coordinates	Season	Fresh	Bloat	Decay	Advanced decay
Badghan	33°46'4.24"N, 35°41'14.78"E	Early summer	Day 1	Day 2-4	Day 5–9	Day 10–30
Fanar	33°52'44"N, 35°34'04"E	Spring	Day 1–4	Day 5–6	Day 7–16	Day 17–30
Deir El-Ahmar	34°7'30.96"N, 36°7'22.04"E	Summer	Day 1	Day 2–3	Day 4–5	Day 6–18
Naas	33°54'42.4"N, 35°40'32.7"E	Spring	Day 1–2	Day 3–6	Day 7–14	Day 15–30
Hasbaya	33°23'52.35"N, 35°41.6'6.59"E	Summer	Day 1	Day 2–3	Day 4–6	Day 7–30
Hammana	33°50'N, 33°44'E	Summer	One day sampling			
Sin El-Fil	33°52'N, 35°32'E	Spring	One day sampling			
Arsal	34°10'N, 36°25'E	Autumn	Autopsy done in Beirut			

Table 1. The sampled localities and the duration of the decomposition stages.



Figure 1. Abundance of Saprinus species in the localities of the present study.

first macerated in 10 % KOH solution for about 3 hours, cleared in 80 % ethanol and macerated in lactic acid with fuchsine, incubated at 60 °C for another 30 minutes, and subsequently cleared in 80 % ethanol and then observed in  $\alpha$ -terpineol in a small dish. Digital photographs of male genitalia were taken by a Nikon 4500 Coolpix camera and edited in Adobe Photoshop CS5. Genitalia drawings based on the photographs or direct observations were produced with the aid of Hakuba klv-7000 light box. Habitus photographs were taken by F. Slamka (Bratislava, Slovakia). Specimens were measured with an ocular micrometre. Due to the lack of phylogenetic information regarding the genus *Saprinus*, the taxa in our present paper are arranged alphabetically. For the morphological terminology the reader is referred to Ôhara (1994) and especially Lackner (2010). The general distribution of *Saprinus* species is extrapolated from Mazur (2011). Whenever Mazur (2011) does not list a given species from Lebanon, while Lackner et al. (2015) report it, we specifically mention it. Specimens were identified using the key of Kryzhanovskij and Reichardt (1976) as well as comparing them with reliably identified voucher specimens deposited in the collection of T. Lackner. All synonymies of species listed here are according to Lackner et al. (2015), with the exception of *Saprinus (S.) certus* Lewis, 1888, which was transferred from the synonymy with *S. (S.) chalcites* to the synonymy with *S. (S.) frontistrius* Marseul, 1885 and *Saprinus (S.) lindrothi* Dahlgren, 1968 that was synonymized with *Saprinus (S.) prasinus* Erichson, 1834 (Lackner and Leschen 2017).

# Results

Our field experiments on pig cadavers in five localities across Lebanon (Fig. 1) resulted in four histerid genera: Saprinus Erichson, 1834 (85.52 %), Margarinotus Marseul, 1854 (14.08 %), Atholus C.G. Thompson, 1859 (0.27 %), and Hypocacculus Bickhardt, 1914 (0.13 %). We collected two species of Margarinotus: Margarinotus (Ptomister) brunneus (Fabricius, 1775) and Margarinotus (Grammostethus) ruficornis (Grimm, 1852); one species of Atholus: Atholus duodecimstriatus duodecimstriatus (Schrank, 1781); and one species of Hypocacculus: Hypocacculus (Hypocacculus) metallescens (Erichson, 1834). The most abundant and diverse was the genus Saprinus of which 13 species were reported. Out of these, eight species are herein reported as new for Lebanon: 1) Saprinus (S.) caerulescens caerulescens (Hoffmann, 1803); 2) S. (S.) calatravensis Fuente, 1899; 3) S. (S.) chalcites (Illiger, 1807); 4) S. (S.) godet (Brullé, 1832); 5) S. (S.) maculatus (P. Rossi, 1792); 6) S. (S.) strigil Marseul, 1855; 7) S. (S.) submarginatus J. Sahlberg, 1913; and 8) S. (S.) tenuistrius sparsutus Solsky, 1876. This paper analyses the diversity of necrophilous Saprinus spp. collected from Lebanon and establishes possible association with the decomposition stages, which could provide more clues for the determination of minimum post-mortem interval (PMI)<sub>min</sub>.

## Diagnosis of the genus Saprinus Erichson, 1834 from the Palaearctic region

The monophyletic (based on the literature references as well as on the on-going molecular studies by T.L.) subfamily Saprininae Blanchard, 1845 is characterised by the presence of distinctive sensory apparatus situated inside their antennal clubs (for more details on this, as well as on the general diagnosis of the subfamily see Lackner 2010) and by the opened antennal cavities, not covered by prosternal 'alae'. Among the Saprininae, the genus *Saprinus* comprises usually ovoid to elongate-oval metallic histerid beetles (although they can also be entirely black, or possess reddish or yellowish maculae on their elytra) of moderate to rather large size (2.50–10.00) for the family. From most other genera, members of *Saprinus* differ usually by widely interrupted frontal stria in the combination of absent prosternal foveae. For the extensive diagnosis of this genus see Lackner (2010).

## Saprinus Erichson, 1834, of Lebanon known hitherto

**1.** Saprinus (Saprinus) aegialius Reitter, 1884 Fig. 2

= Saprinus incognitus Dahlgren, 1964

= Saprinus therondi Auzat, 1931

#### Type locality. Greece.

**Distribution.** Slovakia, Southern Europe, Mediterranean subregion, Armenia, Iran (Mazur 2011). Reported from Lebanon by Lackner et al. (2015).

**Biology.** The biology of *S. aegialius* is not adequately known, but the species is often found on carcasses. This species is often confused with *Saprinus immundus* (Gyllenhal, 1808), and it was absent from our sampling (therefore not shown on a distributional map).

# **2.** Saprinus (Saprinus) caerulescens caerulescens (Hoffmann, 1803) Figs 3, 82, 85a

Hister caerulescens Hoffmann, 1803

= Hister semipunctatus Fabricius, 1792

= Saprinus chobauti Auzat, 1926

Type locality. Germany: Baden-Württemberg.

**Distribution.** Southern Europe, Mediterranean subregion, Portugal (including the Azores Archipelago), Cape Verde Islands, Central Asia, introduced to Peru (Mazur 2011). Newly reported from Lebanon (Fanar, Deir El-Ahmar, Badghan, Naas, Hasbaya; Fig. 82).

**Biology.** This species is found frequently on carrion, with a preference for larger carcasses (e.g., those of dogs, sheep, cattle, camels, etc. – T. Lackner pers. observ.). According to Kryzhanovskij and Reichardt (1976) *S.* (*S.*) *caerulescens caerulescens* was collected on rotten fish, where it preyed upon the beetles of the family Dermestidae. In our samples, we collected 47 specimens. *S.* (*S.*) *caerulescens caerulescens* was abundant during the active decay stage of carcass decomposition and coincides with the presence of Diptera larvae.

**Comment.** This species has another subspecies *S*. (*S*.) *caerulescens punctisternus* Lewis, 1900 that occurs in China, Mongolia, Korean peninsula and Russian Far East (Mazur 2011).

#### 3. Saprinus (Saprinus) calatravensis Fuente, 1899

Figs 4, 15, 16–24, 82, 85b

= Saprinus angoranus Bickhardt, 1911

Type locality. Spain.



Figure 2. Saprinus (S.) aegialius Reitter, 1884 habitus, dorsal view.

**Distribution.** Mediterranean subregion, Arabian Peninsula, Central Asia (Mazur 2011). The distribution of this species is not sufficiently known due to its confusion with the morphologically similar species *S.* (*S.*) *chalcites* (Illiger, 1807) and, mainly *S. georgicus* Marseul, 1862. Newly reported from Lebanon (Deir El-Ahmar, Hasbaya; Fig. 82).

**Biology.** *Saprinus calatravensis* is found on small and medium-sized carrion. According to Kryzhanovskij and Reichardt (1976) it prefers xerophilous landscapes and sandy soils. During our sampling eight specimens were collected of this species.

#### 4. Saprinus (Saprinus) chalcites (Illiger, 1807)

Figs 5, 25, 26–34, 82, 86a

Hister chalcites Illiger, 1807

- = Hister affinis Paykull, 1811
- = Saprinus aerosus Normand & Thérond, 1952
- *= Saprinus melanocephalus* Normand & Thérond, 1952
- = Saprinus prolongatus Normand & Thérond, 1952
- = Saprinus scapularis Normand & Thérond, 1952

#### Type locality. Portugal.



Figure 3. Saprinus (S.) caerulescens caerulescens (Hoffmann, 1803) habitus, dorsal view.



Figure 4. Saprinus (S.) calatravensis Fuente, 1899 habitus, dorsal view.



Figure 5. Saprinus (S.) chalcites (Illiger, 1807) habitus, dorsal view.

**Distribution.** Mediterranean subregion, Africa, Arabian Peninsula, Central Asia, India, Burma, Australia (Mazur 2011). Newly reported from Lebanon (Hasbaya, Fanar, Badghan, Deir El-Ahmar, Naas; Fig. 82).

**Biology.** Saprinus chalcites is a widespread flying predator found on carcasses, rotting vegetable substances, as well as in dung. According to Kryzhanovskij and Reichardt (1976), large numbers of this species have also been found on flowering Stink lily (*Dracunculus vulgaris* Schott, 1832). In our samples, we collected 129 specimens, most abundantly during the active decay stage. Several specimens were likewise collected during the advanced decay stage after the departure of Diptera larvae from the carcasses.

# 5. Saprinus (Saprinus) godet (Brullé, 1832)

Figs 6, 35-43, 82, 86b

Hister godet Brullé, 1832

- *= Saprinus bitterensis* Marseul, 1862
- = Saprinus godetii Marseul, 1857 [emendation]
- = Saprinus pseudolautus Reitter, 1904

Type locality. Greece: Peloponnesus.



Figure 6. Saprinus (S.) godet (Brullé, 1832) habitus, dorsal view.

**Distribution.** Spain, Portugal, southern France, Turkey, Georgia, Kazakhstan, Turkmenistan, Saudi Arabia (Mazur 2011). Reported from Azerbaijan, Greece, Italy, Mongolia, Uzbekistan and southern Russia by Tishechkin and Lackner (2017). Newly reported from Lebanon (Fanar, Badghan, Deir El-Ahmar, Hasbaya, Sin El-Fil; Fig. 82). In our samples 19 specimens were collected.

Biology. Found on carrion (Kryzhanovskij and Reichardt 1976).

#### 6. Saprinus (Saprinus) maculatus (P. Rossi, 1792)

Figs 7, 83

Hister maculatus P. Rossi, 1792

= Saprinus obscuripennis J. Müller, 1899

= Hister personatus Fischer von Waldheim, 1823

= Saprinus pseudocruciata Auzat, 1920

#### **Type locality.** Italy: Etrusca.

**Distribution.** Slovakia, Hungary, south Europe, Turkey, Georgia, Azerbaijan, Cyprus, Iraq, Iran, Turkmenistan, Kazakhstan, Afghanistan (Mazur 2011). Newly reported from Lebanon (Badghan; Fig. 83).

**Biology.** Found chiefly on cadavers, less commonly also in dung or human faeces (Kryzhanovskij and Reichardt 1976). In our samples a single specimen was collected.

# 7. Saprinus (Saprinus) magnoguttatus Reichardt, 1926 Figs 8, 83

#### Type locality. Iran.

Distribution. Turkey, Syria, Iraq, Azerbaijan, Lebanon (Mazur 2011).

**Biology.** Reichardt (1941) reports this species from Azerbaijan as an inhabitant of mountain steppes (500 m); several specimens were also collected by pitfall trapping. In our samples, we collected two specimens from a human corpse that was in the active decay stage (Fig. 83).

**Comment.** Lackner et al. (2015) wrongly attribute this species to G. Müller (1937).

## 8. Saprinus (Saprinus) prasinus prasinus Erichson, 1834

Figs 9, 83, 87a

= Saprinus lindrothi Dahlgren, 1968

#### Type locality. Syria.

**Distribution.** Italy: Sardinia, Greece: Crete, Bulgaria, Turkey, Armenia, Jordan (Mazur 2011). It is reported from Lebanon by Lackner et al. (2015). We collected this species from the following two localities, Fanar and Naas (Fig. 83).

**Biology.** Widespread and frequent East-Mediterranean species found mostly on carrion (Rozner 2010). In our samples, 29 specimens were collected.

**Comment.** Saprinus prasinus has another subspecies S. (S.) prasinus aeneomicans G. Müller, 1960 that occurs in neighbouring Israel and Syria (Lackner et al. 2015). According to Kryzhanovskij and Reichardt (1976) this subspecies differs from the nominotypical one by a distinct bronze hue of the dorsum.

#### 9. Saprinus (Saprinus) robustus Krása, 1944

Figs 10, 44–52, 83, 87b

= Saprinus vermiculatus Dahlgren, 1964

#### Type locality. Turkey: Ankara.

**Distribution.** Greece: Crete, Turkey, Georgia, Syria, Lebanon (Mazur 2011). We report this species from the following two Lebanese localities: Deir El-Ahmar and Hasbaya (Fig. 83).

**Biology.** According to Anlaş et al. (2007) S. (S.) *robustus* is attracted to dung. In our samples, three specimens were collected.



Figure 7. Saprinus (S.) maculatus (P. Rossi, 1792) habitus, dorsal view.



Figure 8. Saprinus (S.) magnoguttatus Reichardt, 1926 habitus, dorsal view.



Figure 9. Saprinus (S.) prasinus prasinus Erichson, 1834 habitus, dorsal view.



Figure 10. Saprinus (S.) robustus Krása, 1944 habitus, dorsal view.

#### 10. Saprinus (Saprinus) strigil Marseul, 1855

Figs 11, 84, 88a

## Type locality. Ethiopia.

**Distribution.** East and central Africa, Saudi Arabia, Turkey, Cyprus, Malta, Syria, Israel (Mazur 2011). Newly reported from Lebanon (Hasbaya, Badghan, Fanar, Deir El-Ahmar, Naas; Fig. 84).

**Biology.** Insufficiently known, according to our observations it is similar to its congeners. In our samples, we collected 149 specimens mainly during the active decay stage and several specimens in other stages of decomposition (Table 1).

## 11. Saprinus (Saprinus) submarginatus J. Sahlberg, 1913

Figs 12, 53–62, 84

#### Type locality. Syria.

**Distribution.** Algeria, Turkey, Israel, Armenia, Azerbaijan, Afghanistan, Iran (Mazur 2011). Newly reported from Lebanon (Badghan).

**Biology.** According to Kryzhanovskij and Reichardt (1976), *S. submarginatus* is a typical inhabitant of xerophilous localities situated in higher elevations. During our sampling, a single specimen was collected. It was collected from altitude of 1174 m, during the summer season (June) from Badghan, Mount Lebanon where the rainfall average was 0 mm and the humidity 55.98 %, which would be in line with the abovementioned observations of Kryzhanovskij and Reichardt (loc. cit.; Fig. 84).

#### 12. Saprinus (Saprinus) subnitescens Bickhardt, 1909

Figs 13, 63–72, 84, 88b

- = Saprinus fagniezi Auzat, 1912
- = Saprinus lecontei Casey, 1916
- = Saprinus meridionalis Ihssen, 1949
- = Saprinus simulans J. Sahlberg, 1913

#### **Type locality.** Hungary, Romania.

**Distribution.** Central and south Europe, North Africa, Spain (including Canary Islands), Portugal (including Madeira), Turkey, central Asia, introduced to north America (Mazur 2011). From Lebanon already reported by Lackner et al. (2015). We herein report this species from the following Lebanese localities: Fanar, Badghan, Naas, Deir El-Ahmar and Hammana (Fig 84).

**Biology.** A typical free-living volant predator found on carrion as well as in dung. In our samples we collected 242 specimens. *Saprinus* (*S.*) *subnitescens* was the most abundant species on the carcasses during both the active and advanced stage of decomposition.



Figure 11. Saprinus (S.) strigil Marseul, 1855 habitus, dorsal view.



Figure 12. Saprinus (S.) submarginatus J. Sahlberg, 1913 habitus, dorsal view.



Figure 13. Saprinus (S.) subnitescens Bickhardt, 1909 habitus, dorsal view.



Figure 14. Saprinus (S.) tenuistrius sparsutus Solsky, 1876 habitus, dorsal view.



Figure 15. Saprinus (S.) calatravensis Fuente, 1899 metaventrite + abdomnen.

# **13.** Saprinus (Saprinus) tenuistrius sparsutus Solsky, 1876 Figs 14, 73–81, 84, 89

#### = Saprinus brunnensis A. Fleischer, 1883

#### Type locality. Uzbekistan.

**Distribution.** Central and South Europe, Central Asia, Mongolia, North China (Mazur 2011). Herein newly reported from Lebanon (Deir El-Ahmar, Hasbaya; Fig. 84).

**Biology.** This species is found predominantly on carcasses. During our sampling eight specimens were collected.

**Comment.** S. (S.) *tenuistrius* has another subspecies, the nominotypical S. (S.) *tenuistrius tenuistrius* Marseul, 1855, which is known from Egypt, north Africa, Ethiopia and Arabian Peninsula (Mazur 2011). According to Kryzhanovskij and Reichardt (1976) the nominotypical subspecies differs from the ssp. *sparsutus* by aciculate punctation on the apical elytral half (the subspecies *sparsutus* lacks this aciculate punctation).



**Figure 16–24. 16** *Saprinus* (*S.*) *calatravensis* Fuente, 1899 male genitalia: 8<sup>th</sup> sternite and tergite, ventral view **17** 8<sup>th</sup> sternite and tergite, dorsal view **18** 8<sup>th</sup> sternite and tergite, lateral view **19** 9<sup>th</sup> + 10<sup>th</sup> tergites, dorsal view **20** 9<sup>th</sup> + 10<sup>th</sup> tergites, lateral view **21** spiculum gastrale (9<sup>th</sup> sternite), ventral view **22** spiculum gastrale (9<sup>th</sup> sternite), lateral view **23** aedeagus, dorsal view **24** aedeagus, lateral view.



Figure 25. Saprinus (S.) chalcites (Illiger, 1807) metaventrite + abdomnen.

# Association of the Saprinus species with the decomposing pig carcasses

*Saprinus* (*S.*) *subnitescens* was collected during the sampling of the months March, April, May, June, July, August, and September. It was collected during three stages of decomposition; bloat, active decay and advanced decay. The maximum amount of specimens was observed during the advanced decay stage of the carcasses. This species was dominant (83 specimens) in Naas (Bikfaya) during spring season at the mean temperature of 16.4 °C. Specimens were collected during both active decay and advanced stages of decomposition from Naas, Badghan, and Fanar, and less frequently from Deir El-Ahmar and Hasbaya, respectively. Moreover, this species was also collected during the bloat stage of decomposition from Badghan (Fig. 88b).

*Saprinus* (*S.*) *strigil* was likewise collected during the sampling events of the months March, April, June, July, August, and September, with the maximum of 104 specimens collected in August from Hasbaya, when the mean temperature was 26.9 °C. The specimens were collected mainly during the active decay stage, less frequent during the advanced decay and minimally present during the bloat stages. Unlike *Saprinus* (*S.*) *subnitescens*, this species was rarely present in Naas. A few specimens were collected during the bloat stage from Badghan (Fig. 88a).



**Figures 26–34.** *26 Saprinus (S.) chalcites* (Illiger, 1807) male genitalia: 8<sup>th</sup> sternite and tergite, ventral view **27** 8<sup>th</sup> sternite and tergite, dorsal view **28** 8<sup>th</sup> sternite and tergite, lateral view **29** 9<sup>th</sup> + 10<sup>th</sup> tergites, dorsal view **30** 9<sup>th</sup> + 10<sup>th</sup> tergites, lateral view **31** spiculum gastrale (9<sup>th</sup> sternite), ventral view **32** spiculum gastrale (9<sup>th</sup> sternite), lateral view **33** aedeagus, dorsal view **34** aedeagus, lateral view.

*Saprinus* (*S.*) *chalcites* was collected during the sampling events of the months of April, June, July, August, September and October. In general, it was dominant during the active decay stage of the carcasses. Peak abundance of 76 specimens was collected in August from Hasbaya during bloat, active decay and advanced decay stages of pig carcass decomposition. Similar amounts were collected during the active and advanced decay stage from Badghan. More specimens were collected during active decay stage than during the advanced decay stage from Deir El-Ahmar. *S.* (*S.*) *chalcites* was present in Naas only during the advanced decay stage (Fig. 86a).

Saprinus (S.) caerulescens caerulescens was captured in March, April, June, July and August; a maximum of 17 specimens was collected during the spring season from



**Figures 35–43. 35** *Saprinus (S.) godet* (Brullé, 1832) male genitalia: 8<sup>th</sup> sternite and tergite, ventral view **36** 8<sup>th</sup> sternite and tergite, dorsal view **37** 8<sup>th</sup> sternite and tergite, lateral view **38** 9<sup>th</sup> + 10<sup>th</sup> tergites, dorsal view **39** 9<sup>th</sup> + 10<sup>th</sup> tergites, lateral view **40** spiculum gastrale (9<sup>th</sup> sternite), ventral view **41** spiculum gastrale (9<sup>th</sup> sternite), lateral view **42** aedeagus, dorsal view **43** aedeagus, lateral view.

Fanar, when the mean weather temperature was 17.6 °C. In general, more specimens were collected during the active decay stages than those collected during the advanced decay ones. Similar amounts were collected during those stages from Badghan. It was collected during the decay stage mainly from Deir El-Ahmar and less frequently from Hasbaya. Regarding the locality Naas, this species was there found to be present mainly during the advanced stage of decomposition and less frequent during the active decay stage (Fig. 85a).



**Figures 44–52. 44** *Saprinus (S.) robustus* Krása, 1944 male genitalia: 8<sup>th</sup> sternite and tergite, ventral view **45** 8<sup>th</sup> sternite and tergite, dorsal view **46** 8<sup>th</sup> sternite and tergite, lateral view **47** 9<sup>th</sup>+10<sup>th</sup> tergites, dorsal view **48** 9<sup>th</sup>+10<sup>th</sup> tergites, lateral view **49** spiculum gastrale (9<sup>th</sup> sternite), ventral view **50** spiculum gastrale (9<sup>th</sup> sternite), lateral view **51** aedeagus, dorsal view **52** aedeagus, lateral view.

*Saprinus (S.) prasinus prasinus –* the peak of its abundance of 27 specimens was likewise in Fanar during spring (March and April). They were mainly captured during the active decay stage and found to be less numerous in the advanced decay stage. This species was likewise collected from Naas during same season during the advanced decay stage only (Fig. 87a).

*Saprinus* (*S.*) *godet* – the peak abundance of this species was in spring in Fanar. It is mainly present during the active decay stage of carcasses decomposition and in lesser numbers during the advanced decay stage. More specimens were collected during the



Figure 53. Saprinus (S.) submarginatus J. Sahlberg, 1913 prosternum.

active decay stage than from the advanced decay stage from Fanar and Badghan. It was only present during the active decay stage in Deir El-Ahmar and during the advanced decay stage in Hasbaya (Fig. 86b).

Both *Saprinus* (S.) *tenuistrius sparsutus* and *Saprinus* (S.) *calatravensis* were collected in July – August. The former was only collected during the active decay stages of Deir El-Ahmar and Hasbaya. It was more abundant in the locality Deir El-Ahmar in July; the mean temperature was 29.7 °C. The latter was mainly present during the active decay stage and less frequently during the bloat and the advanced decay stages (Figs 85b, 89).

*Saprinus* (*S.*) *robustus* was collected from Fanar in March during the active decay stage only, and from Deir El-Ahmar in July during both the active and advanced decay stages. Only a single specimen of *Saprinus* (*S.*) *submarginatus* and of *Saprinus* (*S.*) *maculatus*, respectively, was collected during the summer season from one locality (Badghan); the mean temperature was 20 °C. A male and a female specimen of *Saprinus* (*S.*) *magnoguttatus* were collected from a human corpse that was infested by Diptera larvae (Fig. 87b).



**Figures 54–62. 54** *Saprinus (S.) submarginatus J.* Sahlberg, 1913 male genitalia: 8<sup>th</sup> sternite and tergite, ventral view **55** 8<sup>th</sup> sternite and tergite, dorsal view **56** 8<sup>th</sup> sternite and tergite, lateral view **57** 9<sup>th</sup>+10<sup>th</sup> tergites, dorsal view **58** 9<sup>th</sup>+10<sup>th</sup> tergites, lateral view **59** spiculum gastrale (9<sup>th</sup> sternite), ventral view **60** spiculum gastrale (9<sup>th</sup> sternite), lateral view **61** aedeagus, dorsal view **62** aedeagus, lateral view.

#### Key to Saprinus Erichson, 1834 from Lebanon

This key includes only those species specifically recorded from Lebanon so far. For the *Saprinus* species found in the neighbouring countries (Israel, Palestine, Syria and Jordan), which are likely to occur also in Lebanon (see Lackner et al. 2015 for details), the reader is referred to the key of Kryzhanovskij and Reichardt (1976) that contains most or all of the possible Lebanese *Saprinus* species.

1(4)	Elytra bicolored (Fig. 7)
2(3)	At least the entire lateral elytral margin orange-red, usually most part of the
	elytral disk orange-red with only a short band along the elytral suture black
	(Fig. 7) Saprinus (S.) maculatus (P. Rossi, 1790)
3(2)	Each elytron with a well-defined red macula, never occupying the entire lat-
	eral elytral margin (Fig. 8) Saprinus (S.) magnoguttatus Reichardt, 1926
4(1)	Elytra unicolored, never with red macula (Fig. 2)5
5(6)	Pronotal hypomeron setose, fourth dorsal elytral stria strongly reduced, often absent. A large, usually metallic species (PEL = 5.00–7.50 mm) (Fig. 3)
	Saprinus (S.) caerulescens caerulescens (Hoffman, 1803)
6(5)	Pronotal hypomeron asetose, fourth dorsal elytral stria usually not reduced,
	fully developed. Smaller species (PEL = 2.50–6.50 mm)7
7(8)	Elytra, especially on their apical halves with very dense punctation, punctures
	aciculate and striolate, elytral intervals punctured, third dorsal elytral stria
	well-developed (Fig. 11)Saprinus (S.) strigil Marseul, 1855
8(7)	Elytra with variously dense punctation, but punctures usually not aciculate or
	striolate (some specimens of S. (S.) robustus can have striolate punctures, but
	then the third dorsal elytral stria is always strongly reduced) (Fig. 2)9
9(12)	Elytra with well-defined polished areas 'mirrors', punctation of elytral disk
	very dense, punctures separated by less than their own diameter, third dorsal
	elytral stria reduced to absent (Fig. 2)10
10(11)	Elytral 'mirror' with microscopic scattered punctation, light to dark brown
	species, without greenish or bronze metallic hue, third dorsal elytral stria re-
	duced, but usually discernible; elytral punctation in fourth elytral interval
	reaches elytral half (Fig. 2) Saprinus (S.) aegialius Reitter, 1884
11(10) ł	lytral 'mirror' glabrous, third dorsal elytral stria usually strongly reduced to
	absent, dorsum with distinct greenish or bronze metallic hue; punctation in
	fourth elytral interval does not reach elytral half (Fig. 9)
12(0)	
12(9)	Elytra without well-defined polished areas (mirrors), punctation of the elytral disk
12(1()	less dense, punctures usually separated by their own diameter or more (Fig. 13)13
13(16)	Apices of carinal prosternal striae strongly divergent, laying on lateral sides of the $(\Sigma^2 + C^2)$
1/(15)	prosternal process (Fig. 63); usually larger species (PEL = 3.50–5.30 mm)14
14(15)	Pronotal depressions deep, third dorsal elytral stria usually not reduced, light
	to dark brown species with slight bronze metallic hue (Fig. 13), male with
	deeply depressed metaventrite; male terminalia: apex of 8 <sup>th</sup> sternite (velum)
	asetose, 8 <sup>th</sup> sternite medially not strongly sclerotized (Figs $64-/2$ )
15(14)	Drag and demonstrate shellows third device shere device several demonstrate, 1909
13(14)	Pronotal depressions shallow, third dorsal elytral stria usually strongly re-
	depressed metaventrite, male terminalia, appr of 9th stornite (value) with
	dense tinv setae $8^{th}$ sternite medially strongly seleratized (Figs (4, 52))
	unise my setae, o stermite methany strongry seterouzed (Figs $44-32$ )

- 16(13) Apices of prosternal striae divergent, but never laying on lateral sides of the pronotal process (Fig. 53); usually smaller species (PEL = 2.50–3.90 mm)......17

- 19(20) Entire elytral disk with punctation, punctures separated by twice or more their diameter, dorsal elytral striae thin, impunctate (Fig. 14), antennal club large, light-amber coloured; male terminalia: apices of 8<sup>th</sup> sternite with tiny triangular accessory sclerite furnished with micro-setae, aedeagus short and stout, not dilated apically (Figs 73–81) .....

- 21(22) Apical margin of metaventrite of male without tubercles. Male terminalia: 8<sup>th</sup> sternite with two rows of brush-like setae: one situated approximately medially and another apically, aedeagus constricted before apex; apex rounded (Figs 54–62) (Fig. 12)...... Saprinus (S.) submarginatus J. Sahlberg, 1913
- 22(21) Apical margin of metaventrite of male with two distinct tubercles (Fig. 15) ... 23

# Discussion

In an experiment performed in Al-Baha Province (Kingdom of Saudi Arabia) *Saprinus* species were commonly found on rabbit carcasses during spring, summer, and autumn. In spring, they were found to be abundant during the active decay and the first two days of the dry decay stages. However, in autumn they were common rang-



Figure 63. Saprinus (S.) subnitescens Bickhardt, 1909 prosternum.

ing from the bloat stage to the dry decay stages (Abouzied 2014). Based on Sawaby et al. (2016), in a study that was carried out in Egypt, the authors stated the usefulness of the Histeridae in forensic investigations as they help in time of death estimation. According to Kovarik and Caterino (2016) the majority of histerids are thermophilic, and warmer months of the year correspond to their peak abundance. In general, carcasses decomposition is faster during the summer, due to the large number of insects attracted, whereas rainfall may lead to delayed oviposition and pupation (Abd El-Bar et al. 2016). Based on Bala and Kaur (2014), in an experiment performed on buried pig carcasses in India, Saprinus species and other histerids were common throughout the experiment. Saprinus (S.) semistriatus was previously mentioned in forensic-based experiments as being collected regularly on carrion especially during the active decay stage and useful in post-mortem interval (PMI) estimation (Szelecz et al. 2018). Saprinus (S.) chalcites was recorded on rabbit carcasses during insect succession study in Al-Ahsaa Oasis (Kingdom of Saudi Arabia) in summer, winter, and spring (Shaalan et al. 2017). Saprinus (S.) aeneus was recorded on human cadaver in Italy and utilised to establish the PMI (Introna et al. 1998). Our data concerning carrion-baited Saprinus reflect different peak abundances among species. Generally speaking, the majority of studied species show their peak abundance during the warmer months of the year.

In our survey, *Saprinus* (S.) *caerulescens*, *Saprinus* (S.) *godet*, *Saprinus* (S.) *prasinus*, and *Saprinus* (S.) *robustus* were attracted to cadavers during the both active decay and



**Figures 64–72. 64** *Saprinus (S.) subnitescens* Bickhardt, 1909 male genitalia: 8<sup>th</sup> sternite and tergite, ventral view **65** 8<sup>th</sup> sternite and tergite, dorsal view **66** 8<sup>th</sup> sternite and tergite, lateral view **67** 9<sup>th</sup> + 10<sup>th</sup> tergites, dorsal view **68** 9<sup>th</sup> + 10<sup>th</sup> tergites, lateral view **69** spiculum gastrale (9<sup>th</sup> sternite), ventral view **70** spiculum gastrale (9<sup>th</sup> sternite), lateral view **71** aedeagus, dorsal view **72** aedeagus, lateral view.

advanced decay stages of decomposition, when the Diptera larvae are active and feeding on the carcasses, and were also recovered from the soil after the departure of Diptera larvae to pupate. *Saprinus* (S.) *strigil, Saprinus* (S.) *subnitescens, Saprinus* (S.) *cala*-


**Figures 73–81. 73** *Saprinus (S.) tenuistrius sparsutus* Solsky, 1876 male genitalia: 8<sup>th</sup> sternite and tergite, ventral view **74** 8<sup>th</sup> sternite and tergite, dorsal view **75** 8<sup>th</sup> sternite and tergite, lateral view **76** 9<sup>th</sup> + 10<sup>th</sup> tergites, dorsal view **77** 9<sup>th</sup> + 10<sup>th</sup> tergites, lateral view **78** spiculum gastrale (9<sup>th</sup> sternite), ventral view **79** spiculum gastrale (9<sup>th</sup> sternite), lateral view **80** aedeagus, dorsal view **81** aedeagus, lateral view.

*travensis*, and *Saprinus* (*S.*) *chalcites* were present on the carcasses earlier, in the bloat stage in the summer season, whereas in the spring months (as observed in Fanar and Naas) the arrival of *Saprinus* on the carcass was during the active decay stage (Figs 85, 86, 87, 88). Our data suggest that the active decay stage was reached faster during the summer season than during the spring. The histerid beetles were frequent on all carcasses, but were found to be more diverse during the warm season rather than during autumn. According to our observations, histerids tend to stay in the soil underneath



**Figure 82.** Distribution of S. (S.) *caerulescens caerulescens*, S. (S.) *calatravensis*, S. (S.) *chalcites*, and S. (S.) *godet* in Lebanon.

the carcass during the day and become active during the night. Some are predacious not only on Diptera immature stages but also on the dermestid larvae (Byrd and Castner 2000). Moreover, necrophagous beetles such as Dermestidae are present during the end of the decay process while predators are attracted earlier during the decomposition process (Santos and Santos 2016). This could explain the presence of *Saprinus* during the advanced decay when the dermestids are active. However, the decrease in the abundance of *Saprinus* during the late stages of decomposition coincides with the increase of *Dermestes* immature stages. The other histerid specimens among the genera *Margarinotus, Atholus* and *Hypocacculus* were also reported during the decomposition process of the carcasses, but with lower abundance in comparison with *Saprinus. Mar*-



Figure 83. Distribution of S. (S.) maculatus, S. (S.) magnoguttatus, S. (S.) prasinus prasinus, and S. (S.) robustus in Lebanon.

*garinotus*, *Atholus* as well as *Hypocacculus* are known to be attracted to carrion (Kovarik and Caterino 2016).

It is possible that different patterns of residency on carrion in *Saprinus* result from the differences in release patterns of the volatile organic compounds (VOCs) attracting them (Bajerlein et al. 2011). The decrease in the abundance in late decay stages could be due to the decrease in VOCs. In forensic entomology, Coleoptera is an order of great importance; however, the neglect of this order is due to difficulties regarding taxonomy or its role on the carrion (Almeida et al. 2015). Each biogeographic region has its specific carrion insect fauna (Santos and Santos 2016). Our survey shows the diversity of *Saprinus* in Lebanon, their feeding habits and peak abundances during the warm season in the country.



Figure 84. Distribution of S. (S.) strigil, S. (S.) submarginatus, S. (S.) subnitescens, and S. (S.) tenuistrius sparsutus in Lebanon.

In the present study, 489 specimens of *Saprinus* were recorded, representing 13 species. Out of these, 8 are new records for the Lebanese fauna (Fig. 1). The main aim of this study was to monitor the activity of these beetles on cadavers and identify the species that can be used in later forensic investigations of the country. To ease the identifications, a key for the species is provided, accompanied with illustrations of habitus and male genitalia. More rigorous research concerning both taxonomic identification and the larval development should be conducted to clearly establish the importance of histerid beetles in criminal investigations.



**Figure 85.** Abundance of *Saprinus (S.) caerulescens caerulescens* (85a), and *S. (S.) calatravensis* (85b) during the decomposition stages in different Lebanese localities.



**Figure 86.** Abundance of *Saprinus* (*S.*) *chalcites* (86a) and *S.* (*S.*) *godet* (86b) during the decomposition stages in different Lebanese localities.



Figure 87. Abundance of *Saprinus (S.) prasinus prasinus (*87a) and *S. (S.) robustus (*87b) during the decomposition stages in different Lebanese localities.



Figure 88. Abundance of *Saprinus* (S.) *strigil* (88a) and S. (S.) *subnitescens* (88b) during the decomposition stages in different Lebanese localities.



**Figure 89.** Abundance of *Saprinus (S.) tenuistrius sparsutus* during the decomposition stages in different Lebanese localities.

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