



## Subterranean Campodeidae fauna from Sicily (Diplura): its biogeographical interest with the description of a new species of *Plusiocampa*

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### Abstract

A description is given of a biogeographically interesting new species of Campodeidae (Diplura), *Plusiocampa (Plusiocampa) tinoamorei* sp. nov., a troglolithic species found in the Sicilian Villasmundo and Scrivilleri caves. A second subterranean species already known from three caves in northwest Sicily near Palermo, *Campodea (Campodea) majorica sicula* Condé, 1957, also was studied. Both species were characterized with SEM. Each species belongs to a different monophyletic group: *Plusiocampa* s. str., with thoracic medial posterior macrosetae, and the group related to *Campodea (Campodea) grassi* Silvestri, 1912. Both groups are widely distributed on almost all of the large western Mediterranean islands. Nevertheless, although these two monophyletic groups have a different origin both could be dispersed via land connections between the mainland and the islands during the Messinian Age. This new discovery shows the great value of Sicilian caves that hold species with unique features and of high biogeographic significance.

**Key words:** *Plusiocampa tinoamorei*, *Campodea majorica sicula*, cave fauna, troglomorphy, paleobiogeography, Western Mediterranean

### Introduction

Although Sicily has a speleological variety of caves excavated or formed within limestone, volcanic or gypsum rocks and large karstic areas, its subterranean fauna is still poorly known (Cobolli *et al.* 1994). This is the case for Diplura, an invertebrate group well represented in terrestrial cave fauna around the Mediterranean region (Condé 1955; Sendra 2003; Sendra *et al.* in press). In Sicily subterranean diplurans were represented by a single species, *Campodea (Campodea) majorica sicula* and as Condé (1957) pointed out, it is closely related to other cave-dwelling species in the western Mediterranean. Three troglomorphic species of the *Campodea (Campodea) grassi* Silvestri 1912 group live in subterranean spaces on all of the large islands in the region, except Menorca and Ibiza (Condé 1955) in addition to a small karstic area in the east of the Iberian Peninsula (Sendra & Moreno 2004).

Recent collections in two caves from Siracusa in southeastern Sicily resulted in the discovery of a new species of *Plusiocampa*. This discovery gives a more understandable scenario of the historical biogeography of subterranean campodeids in the western Mediterranean.

### Material and methods

**Sampling methods.** Collections were carried out in four caves: Villasmundo, Scrivilleri, Abisso del Gatto and Abisso della Pietra Selvaggia. The specimens were collected manually using a soft tweezer. They were directly transferred to 96% ethanol in vials immediately after collection, labeled and sealed for transport. In Villasmundo cave temperature and humidity was also measured with a 175–H2 Datalogger.

**Material processing and identification.** The specimens were washed with distilled water and placed between slides and glass coverslips to be examined under a phase-contrast optical microscope (Leica DMLS) using Marc André II solution for permanent slides. The illustrations were made with a drawing tube and the measurements were taken with an ocular micrometer. For measuring the length of the body, the specimens were mounted 'in toto' and were measured from the base of the frontal process distal macrochaetae to the abdomen's supra-anal valve. Two paratypes were coated with palladium-gold used for scanning electronic microscopic photography (Hitachi S-4100) and measurement of the sensilla.

The morphological description and abbreviations used in this paper follow Condé (1956). We use the term gouge sensilla for the concave-convex sensilla located on the antennae (Bareth & Condé 1981) and rosette gland formations for the epicuticular glands described in several Campodeinae species (Bareth & Juberthie-Jupeau 1996). For the position of macrosetae, we follow the terms of Condé (1956): *ma*, medial anterior macrosetae; *la*, lateral anterior macrosetae; *lp*, lateral posterior macrosetae; *mp*, medial posterior; *post*, posterior.

**Karstic Map.** Recent karstic maps (GIS version) produced under the WOKAM concept, method and map of Europe (Chen *et al.* 2017) were used to produce the article's distributional maps.

## Results

### *Plusiocampa (Plusiocampa) tinoamorei* Sendra & Nicolosi sp. nov.

Figs. 1–23; Tables 1, 2.

**Etymology.** This species is dedicated to Prof. Concetto Amore (1940–2018), known as Tino Amore, geologist and associate professor of Sedimentology and Coastal Dynamics at the University of Catania. An expert in karstic processes, he made the census of the Hyblean caves in the province of Syracuse. Prof. Amore was a highly respected authority in the study of erosion phenomena of the coasts and watersheds. As Director of CUTGAN in the period 2007–2010, he worked to protect the Sicilian natural heritage.

**Type material.** Female holotype labelled ♀1 from Villasmundo cave, Melilli, Siracusa, Sicily, Italy (15°06'22.3"E, 37°13'17.2"N), 29<sup>th</sup> July 2018, Giuseppe Nicolosi leg. One male and two juveniles paratypes labelled ♂1, juv1 and juv2, same cave, date and collector. One juvenile paratype labeled juv3, 5 July 2018, same cave and collector. One female and one male paratypes labelled ♀2 and ♂2 from Scrivilleri cave, Priolo Gargallo, Siracusa, Sicily, Italy (37°08'28.9"N 15°09'34.5"E), 30 June 2018, Giuseppe Nicolosi leg. All type material mounted in Marc André II solution, deposit in first author's private collection.

**Other studied material.** Six paratypes from Villasmundo cave, Melilli, Siracusa, Sicily, Italy, 11<sup>th</sup> October 2018, Giuseppe Nicolosi leg., mounted on two separate aluminum stages and coated with palladium-gold.

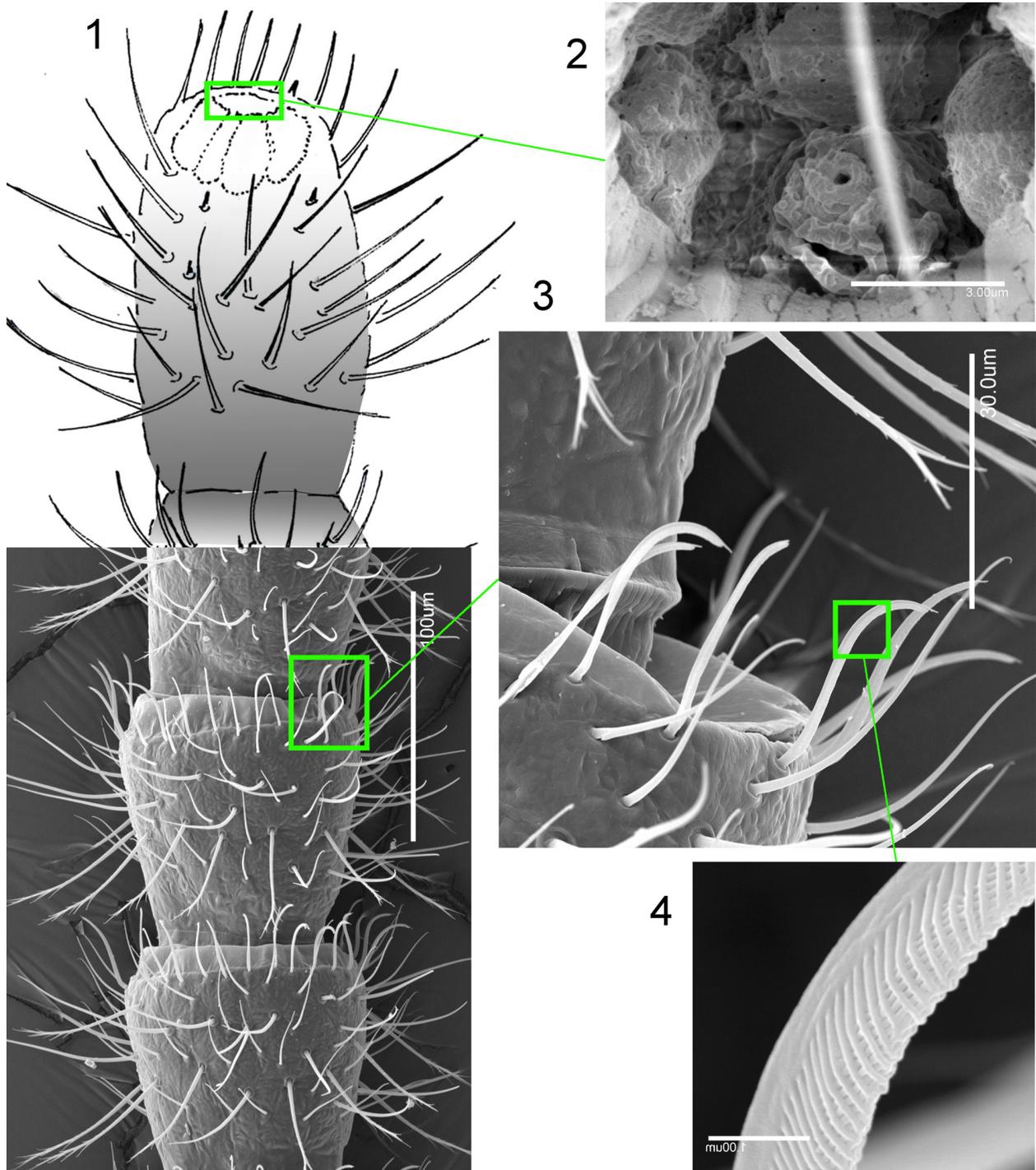
**Description.** Body length 3.0 and 4.5 mm (males), 3.4 and 4.2 mm (females) and 2.7–3.0 mm (juveniles, n= 3). Epicuticle smooth under optical microscopy but weakly reticulated in high magnifications as roundish polygonal structures of variable size (Fig. 7); body with a small number of thin clothing setae with thin distal barbs.

Antennae with 37 to 41 antennomeres; antennae approximately as long as the body length (1–0.9) with medial antennomeres 1.1–1.3 times longer than wide, apical antennomere 2 times longer than wide. Cupuliform organ occupying 1/5 to 1/4 of the total length of the apical antennomere, with about eight large complex olfactory chemoreceptors, each one a multiperforated fold in a rose-form structure, all tightly packed in the narrow open space of the cupuliform organ (Figs. 1, 2). Distal and central antennomeres with two whorls of smooth or distal barbed macrosetae and scattered smooth setae in addition to a single distal whorl of 8–10 moderately long gouge sensilla 26–28 µm long (Figs. 1, 3, 4) and two very short, grooved bud-like sensilla 5 µm long; bud-like sensilla, more than one, also present in the apical antennomere. Proximal antennomeres with typical trichobothria plus a small sensillum on third antennomere in ventral position.

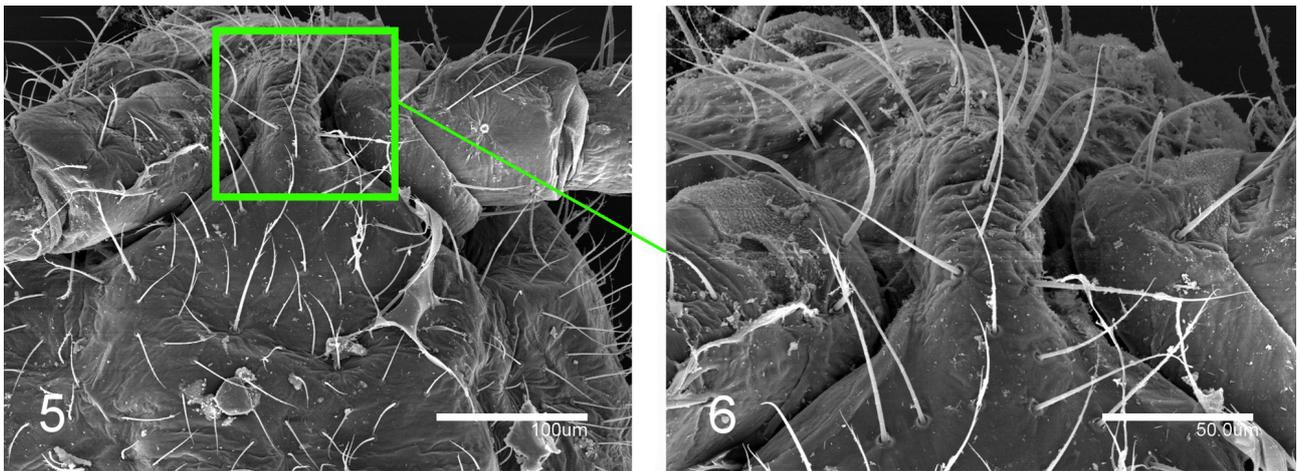
Very slight protrusion of frontal process with slightly differentiated smooth macrosetae (Figs. 5, 6). The three macrosetae along each side of the line of insertion of antennomere and *x* setae with thin distal barbs; length ratios of *a/i/p/x* as 15/20/17/20 (Fig. 12). Suboval labial palps with small latero-external sensillum, two guard setae, up to six setae on anterior border and up to 60 neuroglandular setae.

Thoracic macroseta distribution (Fig. 9): pronotum with 1+1 *ma*, 4+4 *la*, 2+2 *lp* macrosetae and 1+1 slightly differentiated interior *ma* setae; mesonotum with 1+1 *ma*, 3+3 *la*, 2+2 *lp*<sub>1,3</sub>, 1+1 *mp* macrosetae; metanotum with 1+1 *ma*, 1+1 *la*, 2+2 *lp*<sub>2,3</sub>, 1+1 *mp* macrosetae. All macrosetae long with thin barbs along basal half to two-thirds of

each seta; thin marginal setae double the length of clothing setae, and both with thin distal barbs (Figs. 7–9). Legs elongated, metathoracic legs reaching abdominal segments IX or X. Tibia slightly longer than femur and tarsus, which are similar in length (Table 1, Fig. 10). Femora I–III each with one long dorsal macrosetae well differentiated with thin long barbs on the distal half (Fig. 10). Calcars with long, thin barbs throughout (Fig. 15). Tibia I–III with two or three well barbed ventral macrosetae (exceptionally four on tibia III) (Fig. 10). Two or three dorsal, lateral and ventral tarsal setae similar to clothing setae but much longer (Fig. 11, 14). Claws: posterior slightly longer (1.08–1.12: ratio posterior/anterior); large lateral crests; ventral side of the claws noticeably ridged and covered with a micro-granulated surface; and posterior claw with large backward overhang (Figs. 11–14).



**FIGURES 1–4.** *Plusiocampa (Plusiocampa) tinoamorei* sp. nov., paratype. 1) Distal antennomeres. 2) Olfactory chemoreceptors within the cupuliform organ. 3) Gouge sensilla from the distal whorl of a medial antennomere. 4) Closeup of portion of gouge sensillum.



**FIGURES 5–6.** *Plusiocampa (Plusiocampa) tinoamorei* sp. nov., paratype. 5) Frontal process. 6) Closeup of frontal process.

**TABLE 1.** *Plusiocampa (Plusiocampa) tinoamorei* sp. nov., dimensions (mm) and number of antennomeres.

Specimen	Body length	Antennomeres	Antennae length	Femur	Tibia	Tarsus
Paratype, male 2	4.5	-	-	0.70	0.89	0.75
Holotype, female 1	4.2	38	3.7	0.57	0.63	0.46
Paratype, female 2	3.4	41	3.5	0.54	0.61	0.50
Paratype, male 1	3.0	-	-	0.58	0.65	0.55
Paratype, juv. 1	3.0	37	2.7	0.33	0.40	0.37
Paratype, juv. 2	2.9	40	2.6	0.31	0.38	0.36
Paratype, juv. 3	2.7	-	-	0.39	0.41	0.39

Distribution of abdominal macrosetae on tergites (Fig. 16): 1+1 *post*<sub>1</sub> on I–III; 1+1 *la*, 3+3 *lp*<sub>1,2,3</sub> on IV; 1+1 *la*, 5+5 *lp*<sub>1,2,3,4,5</sub> on V–VII; 6+6 (5+6) *lp*<sub>1,2,3,4,5,6</sub> on VIII; and 8+8 *lp*<sub>1,2,3,4,5,6,7,8</sub> on IX abdominal segment. All tergal abdominal macrosetae long and well differentiated with thin barbs along the distal third to half.

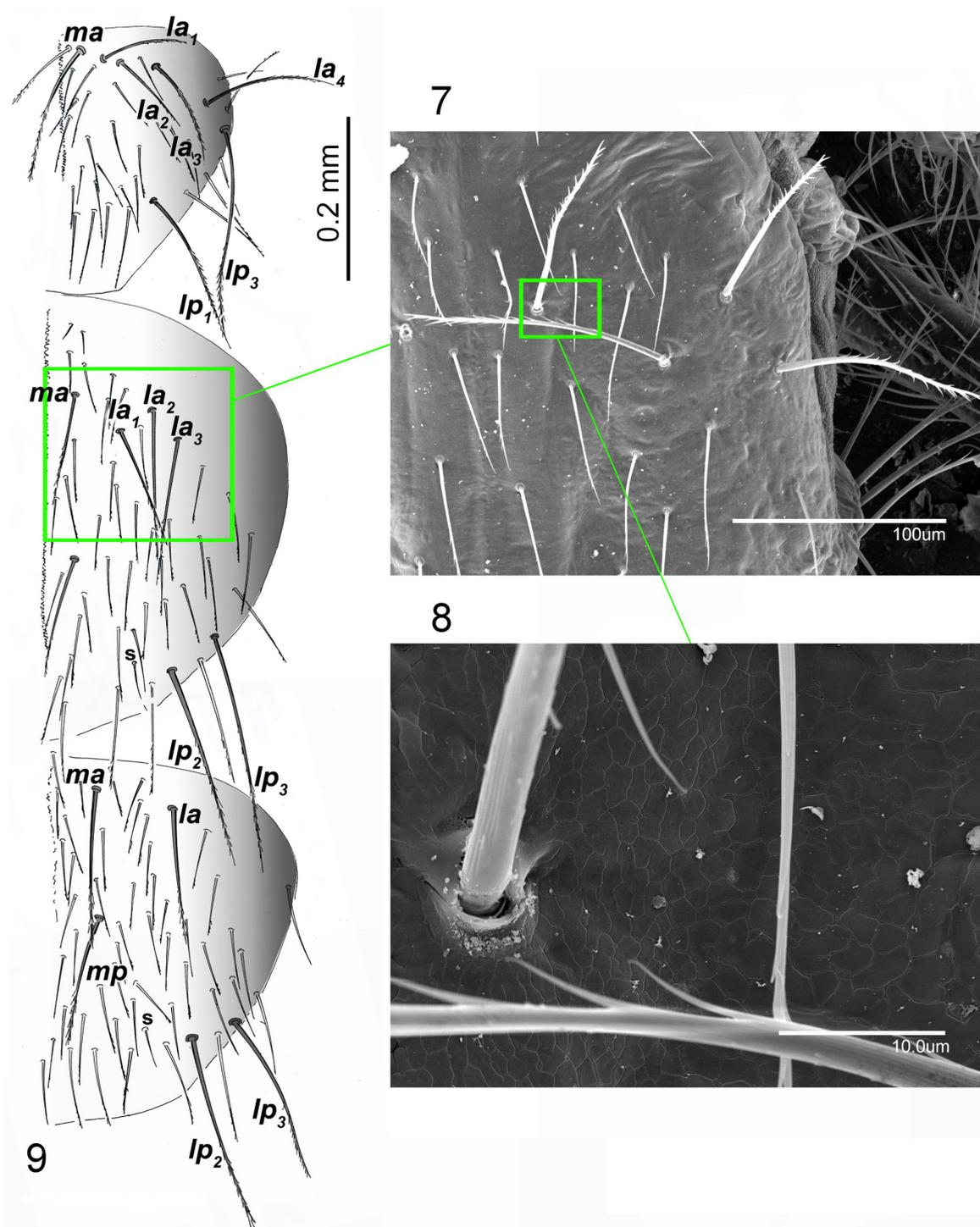
Urosternite I with 7+7 macrosetae (Fig. 17–18); urosternites II to VII with 5+5 macrosetae; urosternite VIII with 2+2 macrosetae; urosternal macrosetae of medium length or longer, with long barbs. Stylus with a long apical seta with two to three thin barbs and with at least one basal tooth; subapical and ventromedial setae with four to six longer barbs (Fig. 23). Cerci almost double the body length, 1.53 and 1.95 times longer than body in the two apparently intact cerci; with six and five primary articles not counting the basal article (Figs. 19–22, Table 2). Length of the cerci increasing slightly from the proximal to distal articles; in addition, the number of whorls of macrosetae macroseta number increasing from about six to more than twenty. All primary articles with a distal thin, barbed whorl seta (Fig. 22).

**TABLE 2.** *Plusiocampa (Plusiocampa) tinoamorei* Sendra & Nicolosi sp. nov., cercal article length and total length (mm) including number of articles of each cercus.

Specimen, body length	Lengths of cerci and articles							
	Base (secondary article)	1st	2nd	3rd	4th	5th	6th	Cercus length
Paratype, ♀2, 3.4 mm	1.00	0.70	0.74	1.20	1.32	1.65		6.61
Paratype, ♂1, 3.0 mm	0.55	0.45	0.48	0.54	0.69	0.87	1.00	4.58

Female urosternite I with short subcylindrical appendages, each bearing up to 10 glandular *a*<sub>1</sub> setae in a distal field.

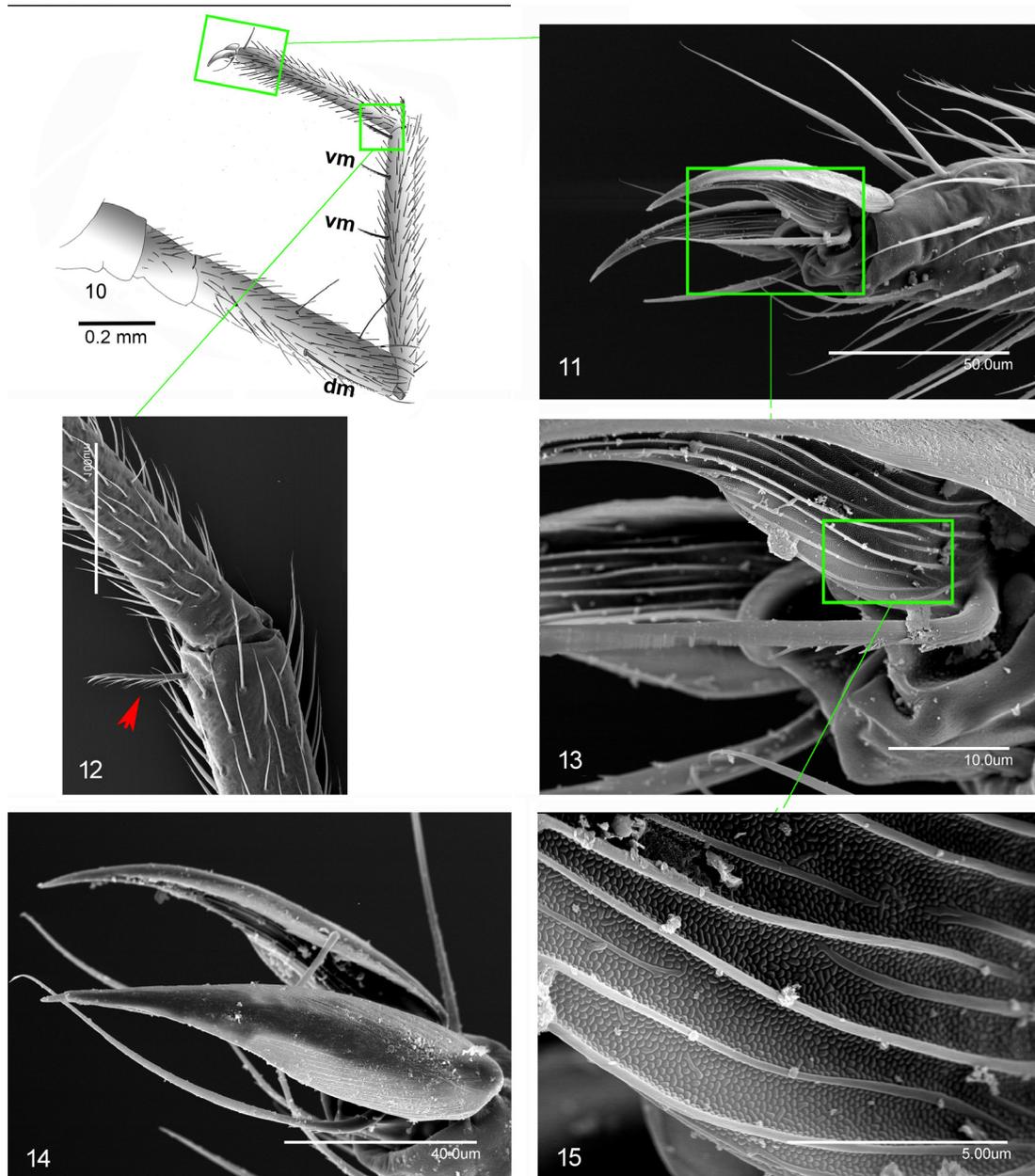
Male urosternite I with short, slightly thickened subcylindrical appendages, each bearing about 14 glandular *a*<sub>1</sub> setae in a distal field; posterior edge of the first urosternite with a glandular field of up to 108 glandular *g*<sub>1</sub> setae, two to three rows of which overlap the posterior margin of the first urosternite (Figs. 17, 18).



**FIGURES 7–9.** *Plusiocampa (Plusiocampa) tinoamerei* sp. nov. 7) Epicuticle surface on mesonotum, paratype. 8) Epicuticle surfaces on metanotum including the *mp* macrosetae, paratype. 9) Pro-, meso- and metanotum, right side, holotype.

**Remarks.** *Plusiocampa (Plusiocampa) tinoamerei* sp. nov. belongs to the group of *Plusiocampa* s. str. with thoracic medial posterior macrosetae. This group appears to be a monophyletic group well represented in European karstic regions including most of the Mediterranean islands (Sendra *et al.* 2003, in press). Focusing on the Western Mediterranean Islands, this group of *Plusiocampa* species with thoracic *mp* macrosetae inhabit subterranean habitats in Mallorca (*Plusiocampa (Plusiocampa) fagei* Condé 1955), Ibiza and Formentera (*Plusiocampa (Plusiocampa) breuili* Condé 1955) and Sardinia (*Plusiocampa (Plusiocampa) sardiniana* Condé 1981 and *Plusiocampa (Plusiocampa) socia* Condé, 1983). They also occur in soil environments of Corsica and the Pontine Islands (*Plusiocampa (Plusiocampa) cyrnensis* Condé 1953 and *Plusiocampa (Plusiocampa) notabilis* Silvestri 1912), as well

as the deep subterranean ecosystem in karstic regions of southern Sicily (*P. (P.) tinoamorei* **sp. nov.**) (Condé 1946a, 1947a, 1953, 1955, 1956, 1978, 1983; Sendra *et al. in press*). In the Western Euro-Mediterranean inland karstic regions, this monophyletic group has a fragmented representation in the Italian Peninsula up to the Rhône Valley (*Plusiocampa (Plusiocampa) bonadonai bonadonai* Condé 1948, *Plusiocampa (Plusiocampa) bonadonai lanzai* Condé 1961, *Plusiocampa (Plusiocampa) bonadonai pavani* Condé & Poivre 1982, *Plusiocampa (Plusiocampa) magdalenae* Condé 1957, *Plusiocampa (Plusiocampa) provincialis provincialis* Condé 1949, *Plusiocampa (Plusiocampa) provincialis praedita* Condé 1949 and *Plusiocampa (Plusiocampa) romana* Condé 1954) and throughout the Baetic Mountains (*Plusiocampa (Plusiocampa) lagari* Sendra & Condé 1987 and *Plusiocampa (Plusiocampa) lucenti* Sendra & Condé 1986) (Condé 1954, 1961, 1981; Condé & Poivre 1982; Sendra & Condé 1986, 1987; Sendra *et al.* 2004). Of these taxa, *P. (P.) tinoamorei* **sp. nov.** seems to be most closely related to *P. (P.) bonadonai*, a species well distributed throughout the Western Alps. *Plusiocampa (P.) tinoamorei* **sp. nov.** has 3+3 posterior macrosetae on the fourth urotergite, whereas *P. (P.) bonadonai* has 5+5. In addition, *P. (P.) tinoamorei* **sp. nov.** has larger and more unequal claws (1.08–1.12) than *P. (P.) bonadonai* (1.05) and more numerous antennomeres (37–41 in *P. (P.) tinoamorei* **sp. nov.**, 31–37 in *P. (P.) bonadonai*).



**FIGURES 10–15.** *Plusiocampa (Plusiocampa) tinoamorei* **sp. nov.** 10) Metathoracic leg, holotype. 11) End of the tarsus and pretarsus, paratype. 12) Detail end of tibia with one calcar indicated with a red arrow, paratype. 13) Claw and lateral processes, lateral side, paratype. 14) Pretarsus, dorsal view, paratype. 15) Detail posterior claw, lateral side, paratype.

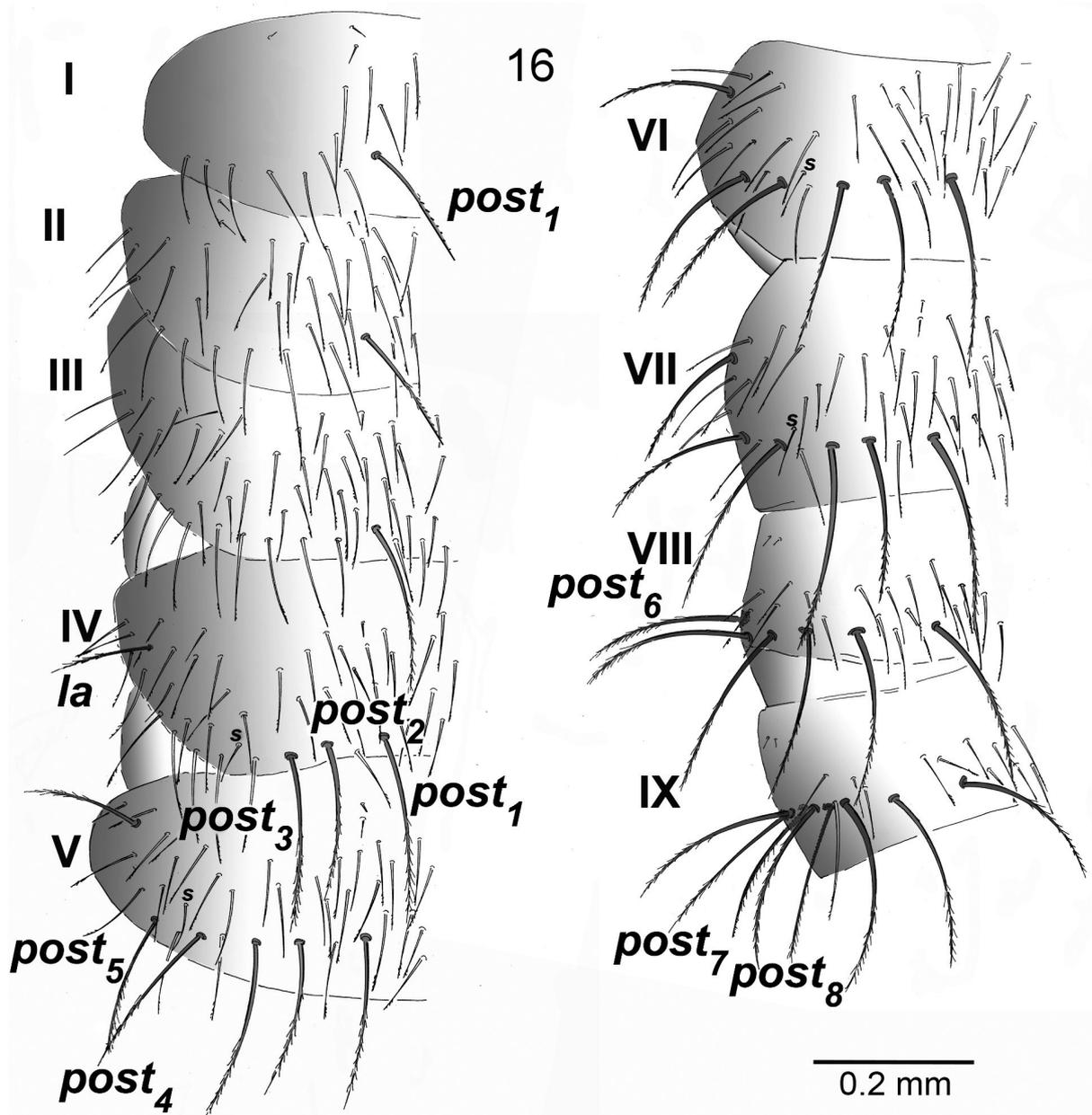
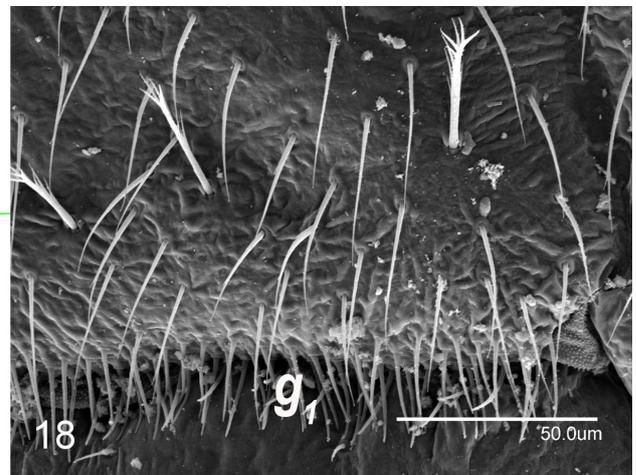
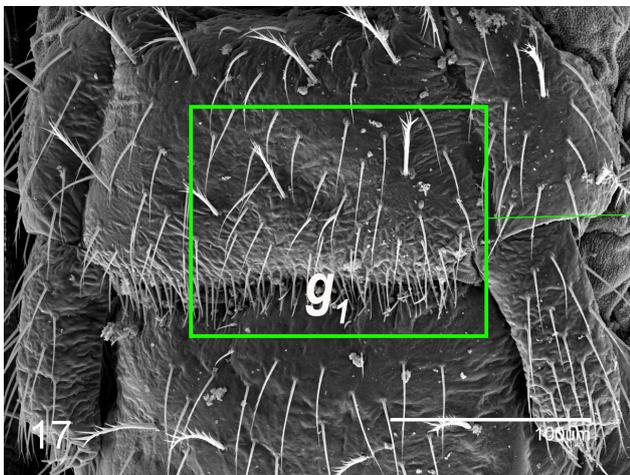
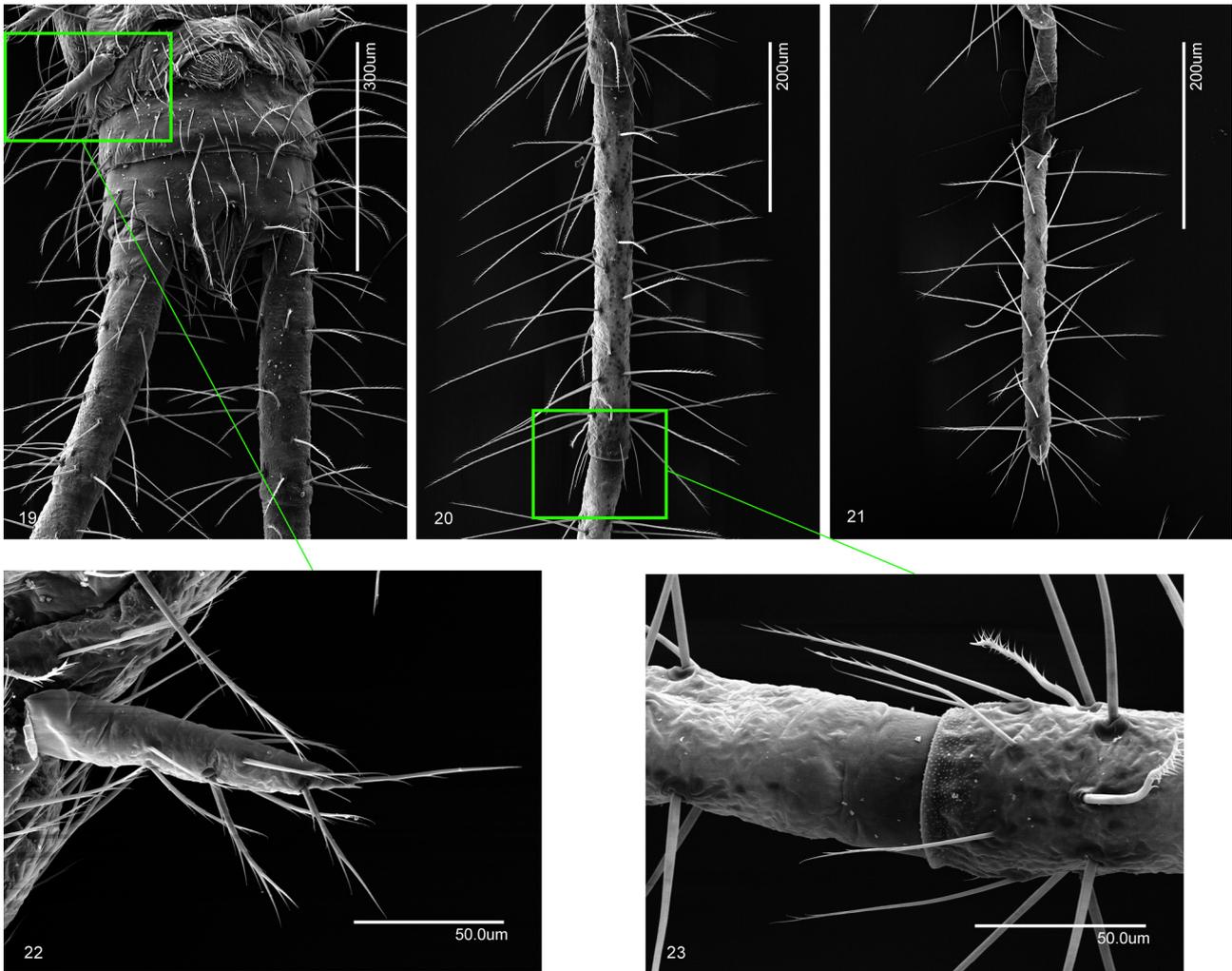


FIGURE 16. *Plusiocampa (Plusiocampa) tinoamorei* sp. nov. Urotergites I-IX, left side, holotype.



FIGURES 17-18. *Plusiocampa (Plusiocampa) tinoamorei* sp. nov., ♂ paratype. 17. First urosteronite of male. 18. Closeup of male first urosteronite with  $g_1$ -glandular.



**FIGURES 19–23.** *Plusiocampa (Plusiocampa) tinoamorei* sp. nov. male paratype. 19. Posterior end of abdomen, ventral view, including proximal parts of cerci. 20. Fourth primary cercal article. 21. Apical cercal article. 22. Distal portion of the fourth primary article. 23. Stylus of the sixth urosternite.

***Campodea (Campodea) majorica sicula* Condé, 1957**

Figs. 24–48, Table 3

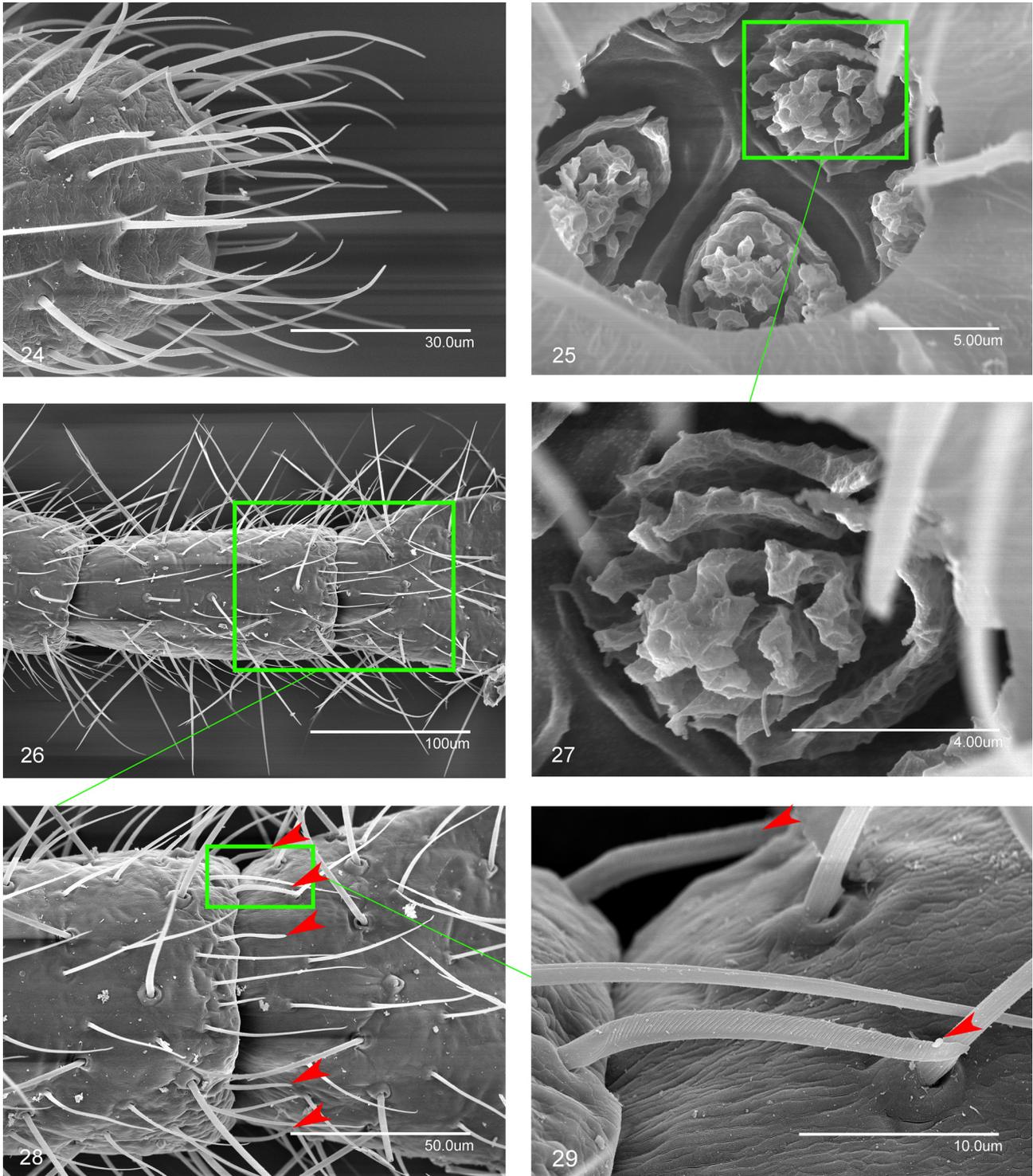
**Studied material.** 5 females, 2 males and 11 unsexed specimens, Abisso del Gatto, Cefalù, Palermo, Sicily, Italy, 10 June 2017, Giuseppe Nicolosi leg; 2 ♀♀, 1 juvenile, Abisso della Pietra Selvaggia, Palermo, Sicily, Italy, 17 November 2018. Giuseppe Nicolosi leg.

**Additions to original description.** Body length 6.4–8.6 mm (females), 5.2–5.3 mm (males); antennae length similar to body length with 44 and 46 antennomeres; medial antennomeres two times longer than wide. Epicuticle of dorsal sclerites of the body and legs covered with microdenticles visible with optical microscopy in addition to a reticulated surface only observed at high magnifications (Figs. 30–36, 42); ventral sclerites smooth under optical microscope but surface reticulated at high magnifications (Figs. 43, 45); rosette gland formations present along scattered on the body (Fig. 32). Dorsal sclerites of body with abundant short, slightly thickened, smooth clothing setae (Figs. 30–36); ventral sclerites with longer and thinner clothing setae (Fig. 43).

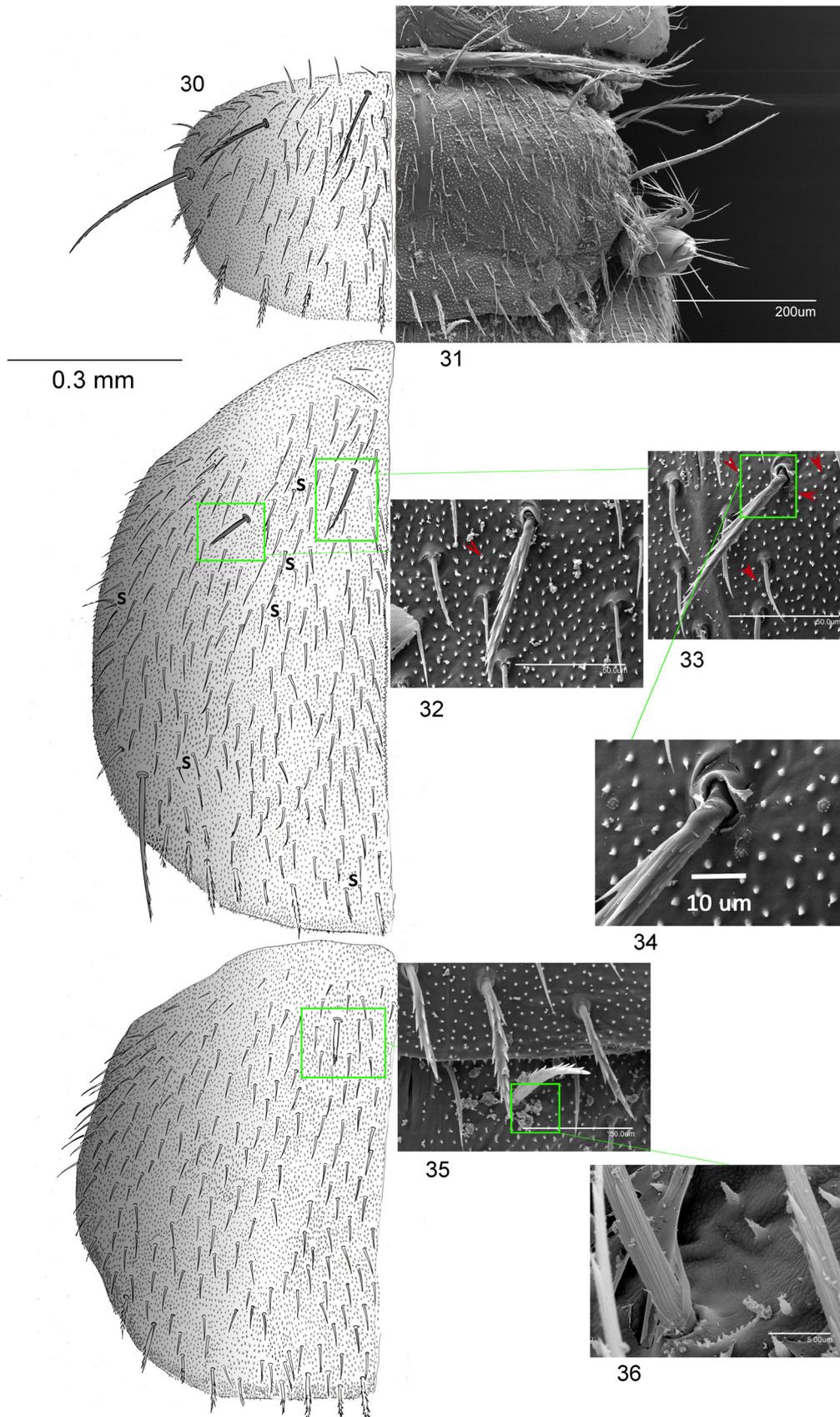
Cupuliform organ occupying 1/8 of the total length of the apical antennomere with about 8 complex olfactory chemoreceptors. Each chemoreceptor is made up of both loosely and tightly packed, multi-perforated folds forming a concentric cupuliform structure with a rim edge. The rim and folds are of varied height and there is a characteristic raised central area, the central pore. All the chemoreceptors are joined by being held within individual cells of a single form (Figs. 24–26). Distal and central antennomeres with three whorls of smooth macrosetae smooth or with

one or two distal barbs, up to three whorls of smooth setae, and a single distal whorl of 10–12 gouge sensilla 18–22  $\mu\text{m}$  long. Apical antennomere with similar setal arrangement (Figs. 27–29). Proximal antennomeres with typical bothriotricha; third antennomere with small subcylindrical ventral sensillum.

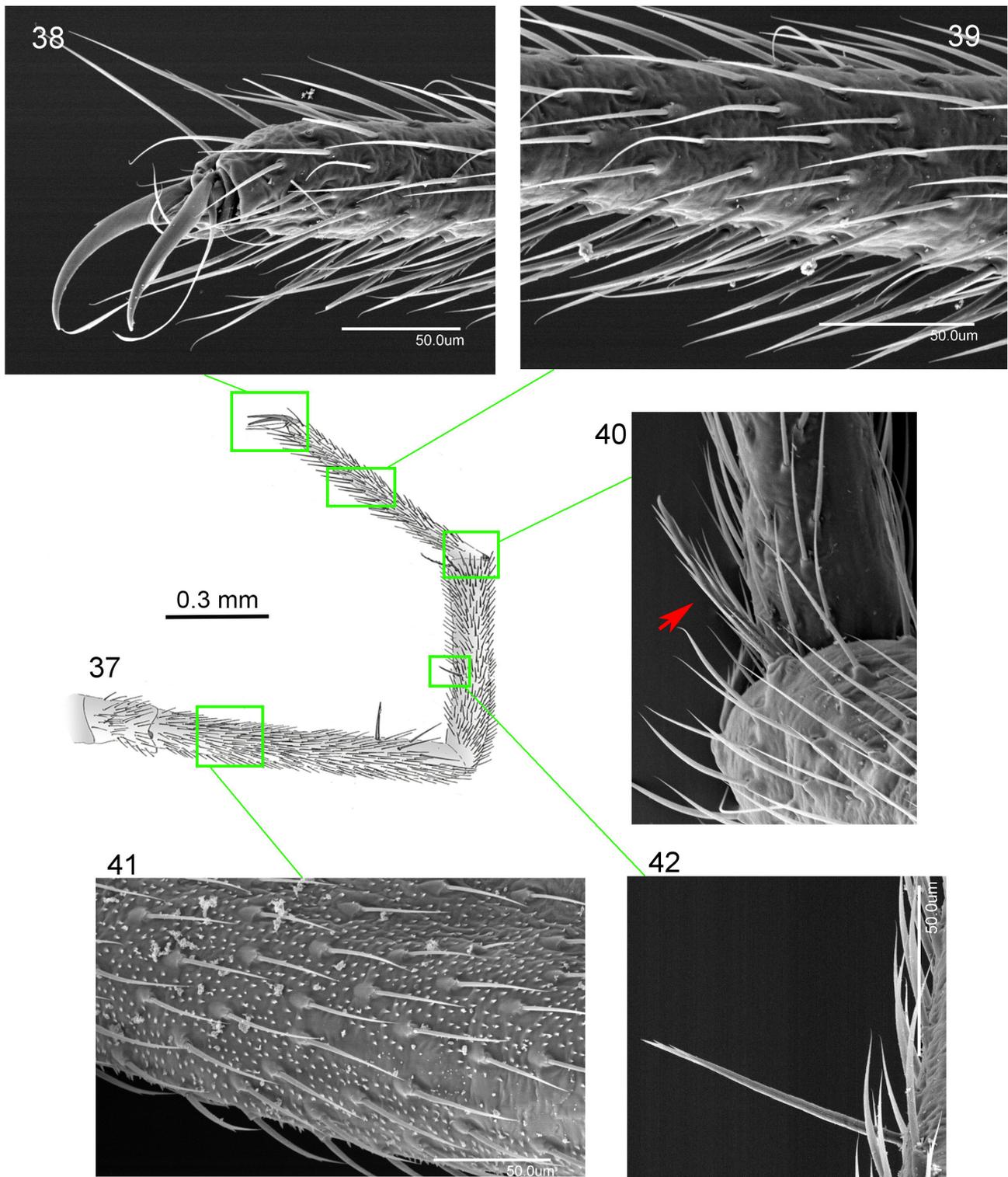
Head subtrapezoidal, slightly protuberant on lateral posterior angles. Simple frontal process with three frontal macrosetae and three thicker, longer and well barbed macrosetae along each side of the line of insertion distinguishable.



**FIGURES 24–29.** *Campodea (Campodea) majorica sicula* Condé, 1957. Specimens from Abisso della Pietra Selvaggia. 24. Distal portion of the last antennomere. 25. Olfactory chemoreceptors within the cupuliform organ. 26. Medial antennomeres. 27. Detail of an olfactory chemoreceptor. 28. Distal portion of a medial antennomere. 29. Gouge sensillum from the distal whorl of a medial antennomere (red arrows indicate gouge sensilla on antennae in Figs. 28–29; f: fold; ca: central pore).



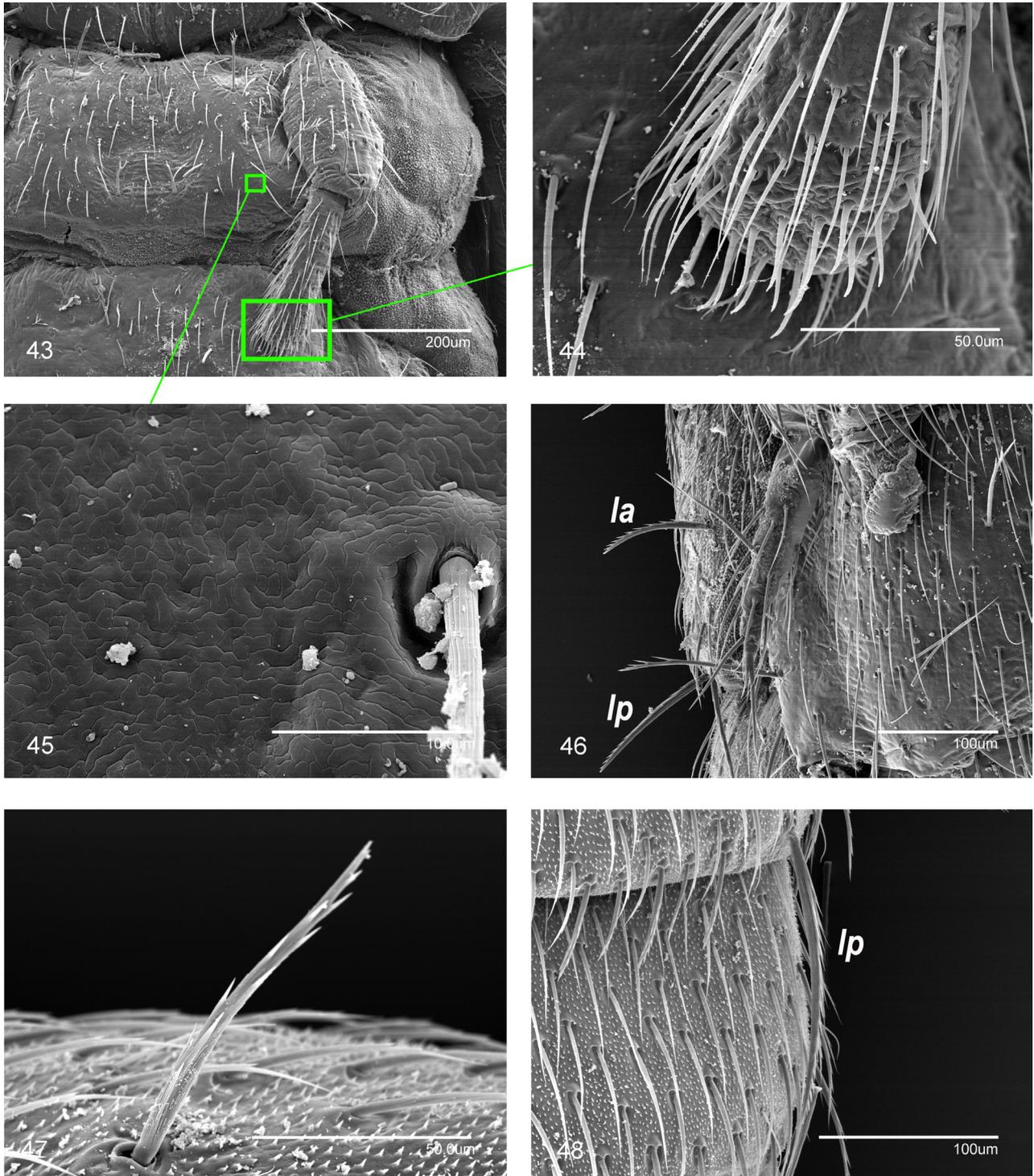
**FIGURES 30–36.** *Campodea (Campodea) majorica sicula* Condé, 1957. Specimens from Abisso della Pietra Selvaggia. 30. Pronotum, mesonotum and metanotum, left side; scale bar = 0.3 mm. 31. Pronotum, right side. 32. Lateral anterior macroseta on mesonotum. 33. Medial anterior macroseta on mesonotum. 34. Detail of rosette gland formations. 35. Medial anterior macroseta on metanotum. 36. Surface of metanotum (red arrows indicate rosette gland formations on mesonotum in Figs. 32 and 33).



**FIGURES 37–42.** *Campodea (Campodea) majorica sicula* Condé, 1957. Specimens from Abisso della Pietra Selvaggia. 37. Metathoracic leg; scale bar = 0.5 mm. 38. Pretarsus and apical portion of tarsus. 39. Medial portion of tarsus. 40. Tibio-tarsal joint with calcar, indicated by red arrow. 41. Tibial macroseta. 42. Femur surface.

Thoracic macrosetal distribution (Figs. 30–36): 3+3 *ma*, *la*, *lp* macrosetae on pronotum and mesonotum, and 1+1 *ma* on metanotum; *ma* and *la* macrosetae well differentiated from clothing setae, slightly thick and with moderately thick barbs; *lp* macrosetae long with thin, short barbs along their entire length. Marginal setae well differentiated from clothing setae, thick and with thick barbs. Urotergal macrosetal length, thickness and number variable, 1+1 *la* and 1+1, 0+1 or 0+0 *lp* in sixth urotergite (Figs. 46–48).

Legs elongate (Fig. 37), metathoracic legs reaching the abdominal segments 8 or 9; tibia slightly longer than femur and tarsus, femur and tarsus + pretarsus of similar length (Table 3). Calcars with barbs of variable length beginning near their base (Fig. 40). Tibiae I–III each with one short, smooth ventral macroseta sometimes possessing one minute distal barb (Fig. 41). Each tarsus with three smooth, subapical macrosetae, a double ventral row of thicker, weakly barbed ventral setae, and a few setiform sensilla (Fig. 39). Subequal slightly curved claws subequal, with setiform lateral processes with a slight apical laminar expansion (Fig. 38).



**FIGURES 43–48.** *Campodea (Campodea) majorica sicula* Condé, 1957. Specimens from Abisso della Pietra Selvaggia. 43. First urosternite of female, left side. 44. Distal portion of the left appendage from the first urosternite of female. 45. Reticulated surface of first urosternite. 46. Latero-ventral view of the seventh abdominal segment; lateral anterior (*la*) and lateral posterior (*lp*) urotergal macrosetae. 47. Lateral anterior macroseta on sixth urotergite. 48. Lateral posterior (*lp*) macroseta on the sixth urotergite.

**TABLE 3.** *Campodea (Campodea) majorica sicula* Condé 1957, dimensions (mm) and number of antennomeres and cercal primary articles.

Specimen	Body length	Antennomeres	Antenna length	Femur	Tibia	Tarsus	Cercal articles	Cercus length
Abisso della Pietra Selvaggia, juvenile	3.8	44	4.25	0.48	0.55	0.52	-	-
Abisso del Gatto, male	5.2	-	-	0.70	0.80	0.65	-	-
Abisso del Gatto, male	5.3	-	-	0.85	0.95	0.75	-	-
Abisso del Gatto, female	6.4	-	-	0.80	0.95	0.70	-	-
Abisso della Pietra Selvaggia, female	7.2	46	8.40	1.05	1.25	0.90	14	11.5
Abisso della Pietra Selvaggia, female	8.6	44	8.00	1.03	1.18	0.90	-	-

Urosternal macrosetae with a few long distal barbs; 6+6 macrosetae on first urosternite (Fig. 43), 4+4 on second to seventh urosternites and 1+1 on eighth urosternite. Stylus setae smooth with two long teeth on apical seta (Fig. 46). Cerci 11.5 mm long on 7.2-mm female; cercal base divided in three followed by 14 primary articles (Table 3). Proximal latero-internal macrosetae with 0–1 distal barbs in the preserved cerci of Abisso del Gatto specimens and with 3–8 barbs in Abisso della Pietra Selvaggia specimens.

Male urosternite I with a posterior field of glandular  $g_1$  setae, and subtrapezoidal appendages with a distal field of glandular  $a_1$  setae: Female subcylindrical appendages with glandular  $a_1$  setae (Figs. 43, 44).

**Remarks.** *Campodea (Campodea) majorica sicula* belongs to a distinct monophyletic group closely related to the soil-dwelling species *C. (C.) grassi* Silvestri 1912 living in the Western Mediterranean region from the Iberian to the Italian Peninsulas, southern France, northern Algeria, and a few locations in Corsica, Pontine Islands, Sardinia and Sicily (Condé 1947a, 1947b, 1955, 1978; Condé & Mathieu 1957; Sendra 1989; Sendra & Jiménez 1986; Sendra & Moreno 2004; Sendra *et al.* 2017; Silvestri 1912, 1932). Condé (1955) suggested that this monophyletic group was composed of three others already described species and six subspecies (*C. majorica sicula* included), all inhabiting the deep subterranean spaces and found in caves. They occupy the main Western Mediterranean Islands: Corsica (*Campodea (Campodea) cyrnea cyrnea* Condé 1946; *Campodea (Campodea) cyrnea alethae* Condé 1948; *Campodea (Campodea) cyrnea virgola* Condé 1948 and *Campodea (Campodea) blandinae blandinae* Condé 1948), Mallorca (*Campodea (Campodea) majorica majorica* Condé 1955 and *Campodea (Campodea) majorica interjecta* Condé 1955), Sardinia (*Campodea (Campodea) blandinae ichnusa* Condé 1957) and Sicily (*C. (C.) majorica sicula*); in addition to an inland subspecies from the extreme east of the Pre-Baetic Mountains in the Iberian Peninsula (*Campodea (Campodea) majorica valentina* Sendra & Moreno 2004) (Condé 1946b, 1948, 1955, 1957; Marletta *et al.* 2015; Sendra & Moreno 2004). The similarities among the taxa of the *C. (C.) grassi* monophyletic group are based on the pattern of macroseta distribution, their robust dorsal clothing setae, marginal setae and macrosetae, and the micro-denticles on the cuticle surface. The species and subspecies of this group are difficult to separate in spite of their isolated environments and insular distributions. All taxonomic features suggested in the descriptions of the taxa should be reviewed. In fact, the variability observed in the shape of the dorsal thoracic and abdominal macrosetae and in the presence or absence of macrosetae on the sixth urotergite, seen in the studied material of *C. (C.) majorica sicula*, cast doubt on the validity of the species and subspecies of this monophyletic group.

Scanning electron microscopy observations were used in this study for better examination of taxonomic features, and for future comparison with other taxa. In the future it will be valuable to use molecular data to better understand the current taxa separation and their validity.

### Description of type localities and other sampled caves and their fauna

Villasmundo cave (cadastral number SISR7032, 15°06'22.3"E, 37°13'17.2"N), the type locality of *Plusiocampa (Plusiocampa) tinoamorei* sp. nov., is part of the Carbonate cave system called Villasmundo-Alfio. The cave is included within the "Complesso Speleologico Villasmundo-S. Alfio" Regional Nature Reserve, managed by "Centro Universitario per la Tutela e la Gestione degli Ambienti Naturali e degli Agroecosistemi" of Catania University.

Villasmundo cave develops in the upper part of the Monti Climiti Formation (Lower-Middle Miocene 25–12 Ma), consisting of calcarenites to calcirudites with detritic-organogenic characteristics (Amore 1998). The cave is located within the territory of Melilli municipality at an elevation of 130 m along the banks of Cugno di Rio stream and, with a total extension of 2.5 kilometers, it represents the longest cave known in the Hyblean district so far. The cave was predominantly formed in vadose conditions and it is still affected by two permanent streams that converge into a terminal lake with a depth of 56 m. However, other streambeds are dry and show a varied presence of speleothems of great scientific value. The entrance tunnel can be flooded in the case of heavy rains, allowing the entry of a large amount of organic matter, which represents the main food sources within the cave. Cracks are also present favoring the entry of roots and other organic matter. Temperature and humidity were, respectively, 18.1°C and 95.3% within the sampling area.

Unlike other cavities in the Hyblean area, Villasmundo cave has never been subjected to faunal investigations; Caruso 1998 reported the presence of only one trogloden species, the isopod *Armadillidium decorum* Brandt 1833, emphasizing the importance of further investigation into a cave of great value due to its wide variety of environments. Currently the cave is under investigation and preliminary observations have allowed the observation of a varied fauna that includes both accidental and adapted species to the subterranean ecosystem. *Plusiocampa (Plusiocampa) tinoamorei* **sp. nov.** was easily spotted and collected under rocks on the clay walls that run along the main gallery, approximately 1.5 meters from the floor, between the entrance tunnel and the section affected by the internal stream waters. They were abundant near biological baits previously placed into the cave, to which they were attracted. Baits containing cheese have been placed within the cave, as a long-term monitoring of the cave is ongoing.

Scrivilleri cave (cadastral number SISR7003, 37°08'28.9"N 15°09'34.5"E) is set in the municipality of Priolo Gargallo (SR), about 10 kilometers from the Villasmundo cave. The entrance to the cavity opens at the base of a paleo-cliff leading to the Eastern Climiti Mountains, consisting of Oligocene macroforaminifera limestones, near the stratigraphic passage with the Monti Climiti Formation (Cavallaro 1998). The cavity has been known since the first half of the last century to have 229 meters of galleries, but recent explorations allowed the discovery of over 2 km of additional underground environments with a predominantly horizontal trend with a great variety of speleothems, that have now made it one of the longest cavities in Sicily.

Scrivilleri cave is located inside the homonymous "Masseria", between two limestone quarries. Thanks to the presence of a farm and the sensitivity of the owner, Pasquale Nicita, the cave has been protected from excavation activities (Bernabei 2018). Until now, only five species have been known of in the cave (Caruso 1998), and none of particular faunistic relevance. *Plusiocampa (P.) tinoamorei* **sp. nov.** was collected near the cave entrance, among stones and stalagmites.

The "Abisso del Gatto" (cadastral number SIPA245, 14°02'54"E, 37°59'37"N) is located in the municipality of Cefalù, Palermo, on the western slope of Cozzo Carzarello, elevation 380 m, within the boundaries of the "Parco delle Madonie". It was excavated in the platform of outcropping limestones in the Madonie Mountains and, together with the "Abisso del Vento", represents the best-developed cave in the Sicily region (Biancone 1994; Macaluso *et al.* 1994). Reaching a total depth of 327 m, it is the deepest cave in the Sicily region, and consists of 1100 m of narrow and meandering galleries connected by a sequence of shafts up to 60 m deep, which are affected by water flow only in the case of prolonged rainfall (Di Maggio *et al.* 2012). No studies related to the subterranean fauna inside the "Abisso del Gatto" are present so far, probably due to its complexity, which makes it accessible only to more experienced speleologists. *Campodea (C.) majorica sicula* was easily spotted along the meander, under rocks and debris, especially in the inner parts of the cave.

The "Abisso della Pietra Selvaggia" (cadastral number SIPA50, 38°9'33.20"N 13°21'25.70"E) is included within the "Monte Pellegrino" Regional Nature Reserve managed by "Rangers d'Italia". It is set on the south side of Pellegrino Mountain, a carbonate massif (606 m elevation) originating in shallow seas from Trias to Eocene, at the Northern side of Palermo. The cave opens at an elevation of 425 m and, with a predominantly vertical development, and consists of a sequence of four shafts of respectively 31 m, 6 m, 38 m, 62 m, and reaching a total depth of 171 m (Mannino 1985). The cave is mainly dry except for the lower part, which is one of the few areas affected by a constant dripping of water and where *Campodea (Campodea) majorica sicula* was easily detected. The presence of this species was already reported by Marletta *et al.* (2015) for the "Abisso della Pietra Selvaggia" and, besides the "Addaura Caprara" cave, located at the opposite slope (north-east) of Mount Pellegrino, it represents the second report for a cave of Monte Pellegrino.

## Notes on conservation status of the Sicilian hypogea

Sicily, with an area of 25,426 km<sup>2</sup>, is the largest Italian region and the largest island in the Mediterranean Sea. It represents a unique area due to its complex geological constitution in which carbonate, evaporite and volcanic terrains coexist simultaneously. This mix has permitted the establishment of a great variety of interesting surface forms and a considerable number of hypogean environments (Panzica La Manna 1993).

Karstic caves are distributed throughout the island, especially in the northwestern Apennine chain (Trapani, northern Palermo, and eastern Madonie Mountains) and the Hyblean plateau in the southeastern part of Sicily. Gypsum caves mostly occur in the central and southern areas of the island, though evaporite landscapes are also present in the northern and western parts of Sicily (Abbate 2003; Di Maggio *et al.* 2012). Over 200 lava caves occur in the east sector of the island along the volcano Etna slopes, in the province of Catania, but are also present in the circum-Sicilian volcanic islands.

Caves in Sicily represent environments of great value, not only for the variety of geological formations but also because they contain a great diversity of habitats that hold species with unique features and high biogeographic significance. The first species for a cave in Sicily dates back to 1776 when the presence of the orthopteran *Dolicopoda palpata* Sulz 1776 was reported within the "Latomie" of the Ear of Dionysus in the Syracuse area (Caruso 1982). This was followed by other sporadic reports and descriptions of a few new species, but it is only more recently that investigations in the subterranean environment have intensified in the island. Of significance is the reporting of two Diplura, *Campodea majorica sicula* within the Addaudo Caprara (Palermo) and *Plusiocampa (Plusiocampa) suspiciosa* Condé & Mathieu 1958 in a small unidentified cave of the Messina province, although this last report is still doubtful due to dubious labeling.

Systematic investigations have been conducted only since 1960 by the University of Catania (e.g. Caruso 1982, 1995, 1998; Caruso & Costa 1978) allowing a more realistic view of Sicilian cave fauna. However, there are still many caves that have not been investigated so far, especially the lava tubes, where the fauna is still poorly known. A greater knowledge of the existing cave fauna and a greater sampling effort in the Sicilian caves would be, therefore, desirable, not only for a better understanding of the processes of biogeographic dispersion but also to favor a more incisive conservation action of the hypogean environments.

The protection of these environments has significantly improved their status. The enactment of Regional Laws 98/1981 and subsequent No. 14/1988 defined the organization of the system of protected natural areas of the Region, favoring the creation and conservation of several natural areas (Dimarca 2004), and recognizing the great value of the geological characteristics of the territory, including caves.

However, despite the conservation level of the natural areas in many cases being satisfactory, there are still many management problems, especially because there are often no specific laws for the protection of hypogean habitats from soils to deep subterranean environments, or they do not refer to the most recent scientific knowledge.

A limit to the optimal protection of the hypogea environments is the establishment of the surrounding protected areas. In Sicily, in fact, the areas of maximum protection do not often extend beyond a limited buffer zone surrounding the cave entrance, often excluding the projection of the cave on the surface, which are frequently subject to human activities. For instance, grazing and agriculture are often allowed in the nearest areas, seriously threatening the delicate balance of these ecosystems and their faunal assemblage, as suggested by several authors (e.g. Hobbs & Gunn 1998; Poulson & Lavoie 2000).

In addition, many other karst or volcanic caves of the island are not included within parks or reserve boundaries. In the territory surrounding the Etna volcano area, for example, the high degree of building development has led to the partial or total disappearance of numerous lava tunnels. Furthermore, the negative effects resulting from unregulated frequency by visitors is undoubted (Baker & Genty 1998).

There are also caves on private lands, not accessible and which are excluded from protected boundaries, whose conservation needs to be ensured. Many of them have never been subjected to scientific investigations or have been irreparably damaged.

*Plusiocampa (Plusiocampa) tinoamorei* **sp. nov.** was detected in Villasmundo (SISR7032) and Scrivilleri caves (SISR7003). The first, being located within a Strict Nature Reserve since 1998, is accessible just for scientific purpose and by experienced speleologists. The establishment of a protected area together with the presence of a rather narrow entrance tunnel, which prevents access to people without technical training, has favored a high degree of conservation of the cave. A long-term study is currently underway and the presence of other troglobitic species has

been documented, including a new Pselaphinae (Sabella *et al.* 2019). Although the cave remains well preserved, heavy grazing impacts occur in the surrounding areas.

Scrivilleri cave has been known since the early twentieth century, as evidenced by the presence of numerous damage such as graffiti and damage to stalactites and stalagmites. Access to the cave is through an artificial well, which can only be accessed via a speleological ladder. The presence of some very narrow passages has prevented access to the innermost portions of the cave, which is still very well preserved. Currently, the cave is located within a private farm and this has allowed its conservation and protection from the advance of two neighboring quarries, one of which is about 150 meters away from the cave entrance.

*Campodea majorica sicula* was detected in the “Abisso del Gatto” and “Abisso della Pietra Selvaggia”, both included in the boundaries of protected areas. The first is located within the “Parco delle Madonie”, managed by the “Ente Parco delle Madonie”. The “Abisso della Pietra Selvaggia” included within the Reserve “Monte Pellegrino”, is managed by the “Rangers d'Italia”. Both caves have a good state of preservation, also thanks to technical difficulty, which allow access to expert speleologists only.

## Biogeographical considerations

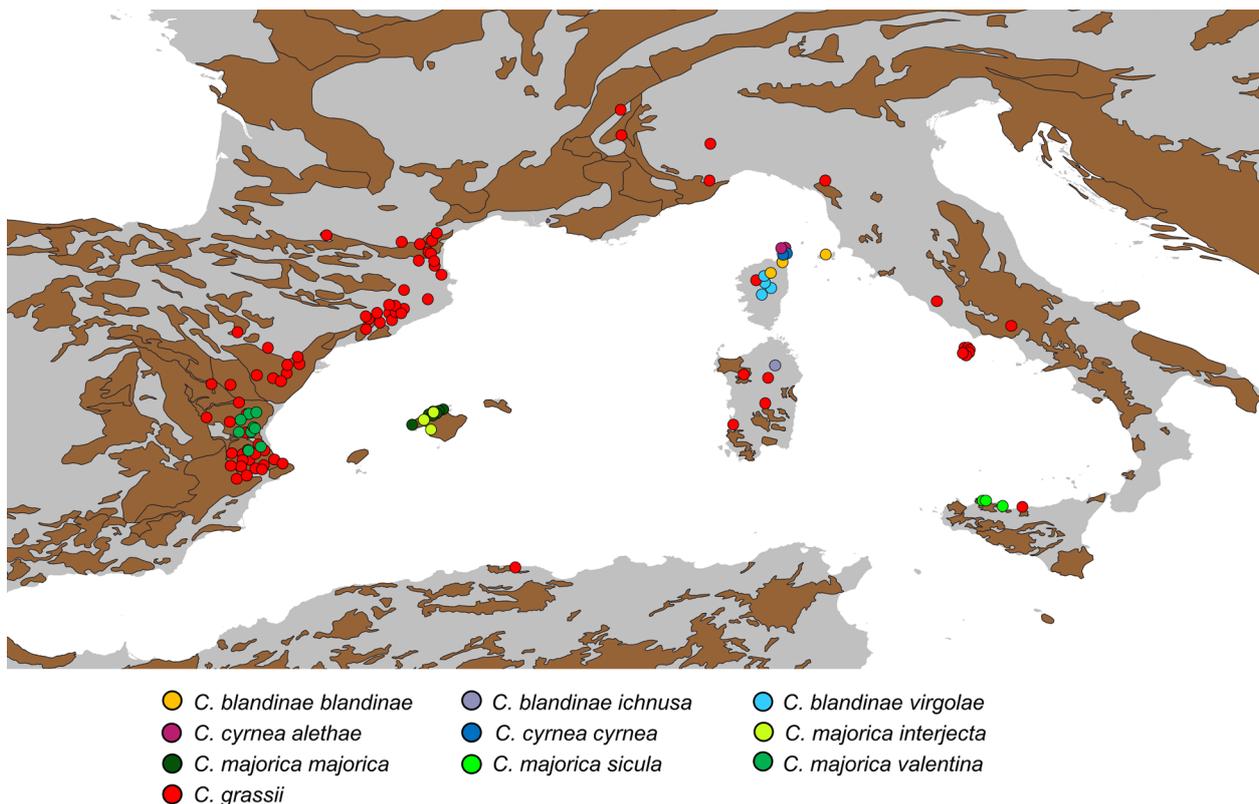
Sicily is located a short distance from the Italian peninsula, Sardinia and the Tunisian coasts. Links between the island and surrounding regions have allowed dispersion during the Cenozoic mainly during the Quaternary epoch, thanks to the presence of relatively low waters, and the continuous phenomena of regression and glacial eustatism. The opening of the Strait of Messina occurred rather recently, about 20–30 thousand years ago, when the condition of Sicily corresponded little to its current state and new faunistic inputs would not have been possible (La Greca 1957). Nevertheless, there are other older connections; according to Furon (1950), the Paleogene Sicily would have been part of a large emerged land called Tirrenide, which would have occupied the area of the current Tyrrhenian. This hypothesis is corroborated by the numerous faunal affinities existing between Sardinia, Corsica, southern France, Balearic Islands and Iberian Peninsula on one side; between North Africa and the Iberian Peninsula, as well as between North Africa and Sicily on the other (La Greca 1958, 1961, 1983). Furthermore, the closure of communications between the Atlantic and the Mediterranean, and the desiccation of the latter, during the Messinian, would have facilitated faunistic and floristic exchanges between North Africa and Sicily (Giachino *et al.* 2011).

Despite the knowledge of Sicilian fauna in some cases appearing to be still limited, the presence and distribution of some groups seems to confirm the evidence of a past territorial connection of Sicily with the rest of the Mediterranean basin (Giachino & Vailati 2007). Giachino (2011) identified three models of populations on the basis of the study of the Sicilian Cholevidi: the presence of taxa that are distributed in Europe or Italy which, through the Apennine chain, penetrate towards Sicily; Maghreb elements which penetrate from the south and, finally, Balkan elements that arrived through trans-Adriatic or trans-Ionian penetrations. Moreover, analysis of the cave populations of species suggests their mainly Tyrrhenian origin (La Greca 1958), as evidenced by the presence of different genera with a Mediterranean-western distribution, locally present in Morocco, Algeria, Northern Sahara, Balearic Islands, Southern Spain, Sicily, and the circum-Sicilian Islands (e.g. Caruso 1982; Pezzino 2014). Such distribution is particularly evident for some taxa, such as Isopoda of the genus *Spelaeoniscus*, which shows a southwestern Mediterranean distribution, confirming a remarkably old phylogeny of the taxa that probably originated in North Africa (Caruso *et al.* 2017).

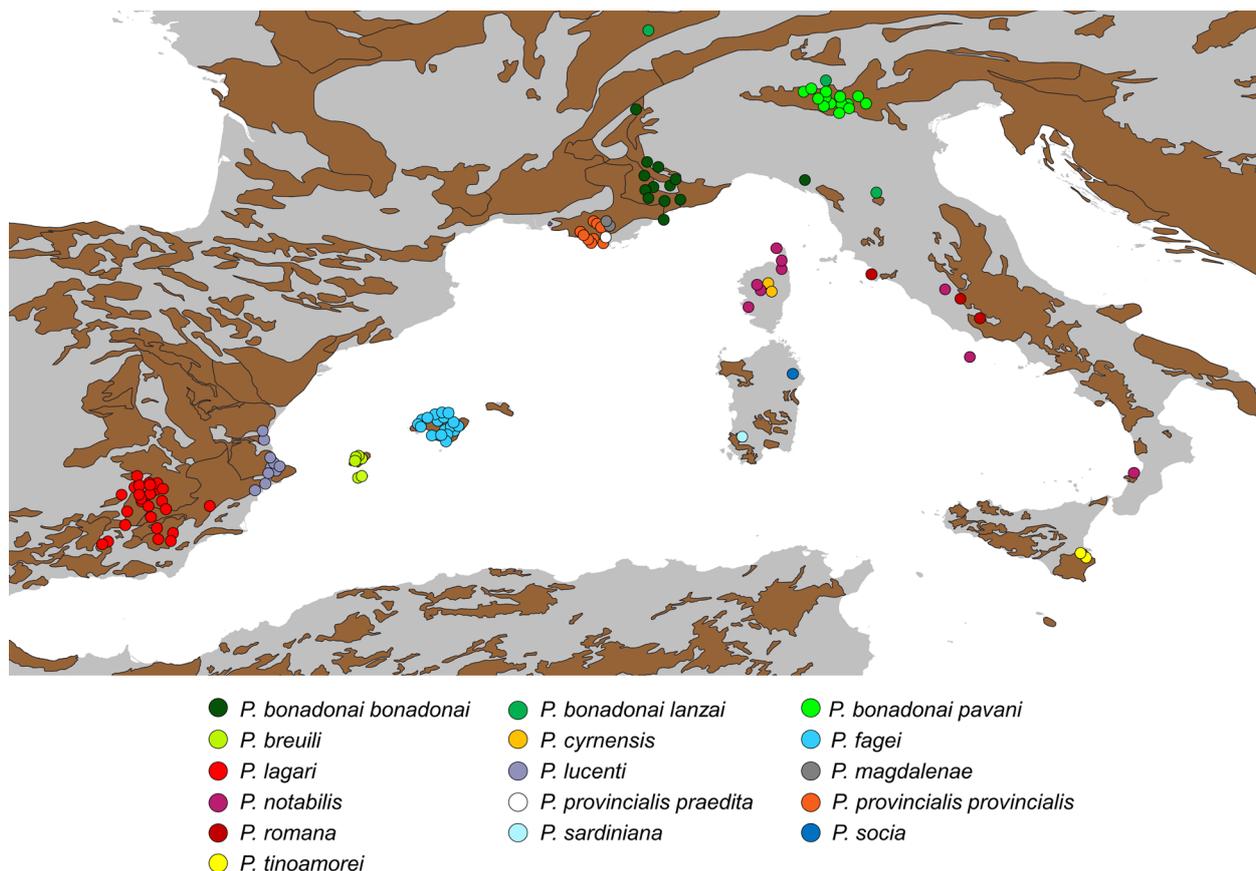
The distribution of soil and subterranean Sicilian campodeids fits with this paleobiogeographical Cenozoic scenario and with the dispersal hypothesis suggested for Cholevidi and the isopod *Spelaeoniscus*. Focusing on the two phyletic groups present in subterranean habitats of Sicily—*Plusiocampa* s. str. with thoracic medial posterior macrosetae and the *C. (C.) grassi* group—both have a large distribution range around the Western Mediterranean karstic regions, in islands and mainland areas. Furthermore, this distribution area extends throughout the Mediterranean basin in the case of *Plusiocampa* s. str. that possess thoracic medial posterior macrosetae (*mp*). The distinguishing character state of these groups is their troglomorphy due to their hypogean life in the deep subterranean ecosystem, where they thrive with great success. In both groups the land connection of mainland and islands during the Messinian Age is a suitable explanation for such faunal affinities (Sendra 2003; Sendra *et al.* 2004). Nevertheless, both monophyletic groups seem to have a different history. The *C. (C.) grassi* group belongs to the Campodeinae, which has a worldwide distribution and seems to have inhabited the older European microplates probably during the Meso-

zoic or a much older era (Sendra 2003). Four current species and six subspecies of the *C. (C.) grassi* monophyletic group (*C. (C.) blandinae blandinae*, *C. (C.) blandinae ichnusa*, *C. (C.) cyrnea alethae*, *C. (C.) cyrnea cyrnea*, *C. (C.) cyrnea virgolae*, *C. (C.) majorica interjecta*, *C. (C.) majorica majorica*, *C. (C.) majorica sicula*, *C. (C.) majorica valentina*) show a more recent and complete distribution in the regions around the Western Mediterranean. *Plusiocampa* s. str. (*mp*) and Plusiocampinae in general seem to be relatively recent arrivals to the Mediterranean basin (Sendra *et al.* 2004). It is suggested that *Plusiocampa* s. str. (*mp*) could disperse in the late Miocene from Anatolia along the Miocene Alpine range. This connection was open throughout Europe during this epoch (Rögl & Steiniger 1984) and could have allowed dispersal through the new emerged lands in Balkan Peninsula and Aegean Islands, Italian Peninsula, Rhône valley, and Western Mediterranean, including the Mediterranean islands and Pre-Baetic Mountains in the south of the Iberian Peninsula. When these areas emerged from the late Tethys Sea or Paratethys, these campodeids were able to inhabit the Western Mediterranean. The vestiges of this dispersal are currently 11 species and three subspecies of *Plusiocampa* s. str. (*mp*) newcomers: *P. (P.) fagei* and *P. (P.) breuili* from the Balearic islands; *P. (P.) lucenti* and *P. (P.) lagari* from the Pre-Baetic Mountains; *P. (P.) sardiniana* and *P. (P.) socia* from Sardinia, *P. (P.) cyrnensis* and *P. (P.) notabilis* from Corsica, *P. (P.) bonadonai bonadonai*, *P. (P.) bonadonai lanzai*, *P. (P.) bonadonai pavani*, *P. (P.) magdalenae*, *P. (P.) provincialis provincialis*, *P. (P.) provincialis praedita* and *P. (P.) romana* from the Italian Peninsula up to the Rhône Valley); and *P. (P.) tinoamorei* **sp. nov.** from Sicily.

The fragmented distribution of this *Plusiocampa* s. str. (*mp*) group around the Western Mediterranean karstic regions reveals the difficulty encountered in dispersal and establishment. These regional gaps (Provence, Catalonia, Sardinia, Kabyliès and the rest of the Alboran plate in the Interior Zone of Baetic Mountains) were presumably already occupied by the monophyletic group of *Plusiocampa* that lack thoracic medial posterior macrosetae (Sendra *et al.* 2004).



**FIGURE 49.** Map of karstic areas (in brown) of Western Mediterranean with the distribution of taxa of the *Campodea (Campodea) grassi* Silvestri, 1912 monophyletic group.



**FIGURE 50.** Map of karstic areas (in brown) of Western Mediterranean with the distribution of taxa of the monophyletic group *Plusiocampa* s. str. with thoracic medial posterior macrosetae.

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