



***Nitocrellopsis rouchi* sp. n., a new ameirid harpacticoid from phreatic waters in France (Copepoda: Harpacticoida: Ameiridae)**

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Abstract

Ongoing investigation of the interstitial communities of the River Rhône revealed the presence of a new species of Ameiridae Monard, 1927. Both sexes of *Nitocrellopsis rouchi* sp. n. are described herein. Re-examination of type-material (when available) of *Nitocrellopsis ioneli* (Dumont & Decraemer, 1974), *N. petkovskii* (Rouch, 1987), *N. hippocratis* (Cottarelli & Forniz, 1993) and *N. hellenica* (Cottarelli & Forniz, 1993) and other *Nitocrella*-related genera *sensu* Petkovski (1976), revealed differences in cephalic appendage morphology between species which are discussed together with the identification of some minute morphological characters observed for *N. rouchi* with the aid of scanning electron microscopy. A generic diagnosis is given, together with type-species designation to make available the generic name. A key to species of *Nitocrellopsis* gen. nov. is provided.

Abbreviations: Md plp – mandibular palp; Mx1 – maxillule; Mx2 – maxilla; Mxp – maxilliped; Exp – exopod; Enp – endopod; P1-P6 – first to sixth thoracopods

Introduction

Ecological investigations of the River Rhône focused on the surface/groundwater exchanges in the phreatic zone close to the main channel, and to their effects on interstitial community distribution and abundance along both vertical and horizontal profiles (Gibert, 1994). The study site (Grand Gravier area) is located downstream from the city of Lyon, on the right bank of the river. This site is one of the major pumping areas of drinking water for the south-western part of Lyon (13000 m³ d⁻¹, Gibert et al., 1998). Consequently, interstitial flowpaths in this zone are dominated by surface water infiltration. Sampling wells were drilled in the Würm fluvial deposits of the River Rhône. The aquifer, composed of sand and gravel alternations, has an approximate local thickness of 25 m. Hydraulic conductivity is estimated from 1 to 3 10⁻³ m s⁻¹.

Numerous samples taken from the wells at different depths revealed the presence of a new species of *Nitocrellopsis*, described herein as *N. rouchi* sp. n.

The stygobiont genus *Nitocrellopsis* as established by Petkovski (1976) is a poorly diversified genus belonging to the primarily marine family Ameiridae Monard, 1927, at present, known with some 300 species and subspecies (Conroy-Dalton & Huys, 1996).

Ameirid harpacticoids have occasionally successfully invaded freshwater habitats, to which they are especially linked to groundwater. Of the seven ameirid genera recorded from fresh groundwater, with a total of 97 species and subspecies, *Nitokra* Boeck, 1864, *Parapseudoleptomesochra* Lang, 1965 and *Praeleptomesochra* Lang, 1965 are also represented in the marine environment. *Nitocrella* Chappuis, 1923 has only been sporadically recorded from littoral brackish waters, and frequently recorded from both coastal and inland groundwaters. *Stygonitocrella* Petkovski, 1976, *Psammonitocrella* Rouch, 1992 and *Nitocrellopsis* are

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exclusively continental, although some species are known from coastal (not littoral) groundwaters, and some from continental saline and brackish waters. The genus *Nitokra* is common in limnic superficial habitats, but only occasionally radiated in groundwaters, suggesting an intermediate step in the colonisation of continental waters via surface habitats (Rouch, 1986). In contrast, *Nitocrella*, *Parapseudoleptomesochra*, *Nitocrellopsis*, *Stygonitocrella* and *Psammonitocrella* lack such intermediate representatives, and have possibly colonised groundwaters via coastal interstitial and, less frequently, via karstic discontinuities of the carbonatic platform. At present, it is difficult to resolve the sequence of troglobisation and speciation processes of freshwater stygobiont ameirids, as well as patterns of relationship among and within stygobiont genera, due to lack of knowledge on the systematics and phylogeny of the family Ameiridae (Conroy-Dalton & Huys, 1996) as a whole. Mielke (1995) questioned the monophyly of *Parapseudoleptomesochra* as defined by Pesce & Petkovski (1980), and Martínez Arbizu & Moura (1994) questioned the position of *Psammonitocrella* within the Ameiridae. Moreover, the morphological groups recognised within both *Nitokra* and *Nitocrella* genera could represent distinct lineages.

Material and methods

Samples were taken from wells at different depths by using double packer apparatus (Danielopol & Neiderreiter, 1987), with a Bou-Rouch pump (1967), filtered through a 100 μm mesh and preserved in 7% formaldehyde. Dissected specimens were mounted in polyvinyl lactophenol. Drawings and measurements were made using a Leitz Laborlux phase contrast microscope, with the aid of a camera lucida. Because body length measurements are often affected by the telescoping action of body somites, an additional method for measuring the total length of the specimens was used, according to Clément & Moore (1995). Some details gained from scanning electron microscopy (SEM) are added to line drawings. For SEM 20 ♀♀ and 10 ♂♂ were dehydrated in a graded ethanol series, critical point dried in a Balzers Union CPD 020 apparatus and coated with gold in a Balzers Union SCD 040 sputter. Observations were made with a Philips SEM XL30 CP scanning electron microscope. The terminology proposed by Huys & Boxshall (1991) is adopted in the taxonomic description.

Taxonomic account

Order Harpacticoida Sars, 1903
 Family Ameiridae Monard, 1927
 Genus *Nitocrellopsis* gen. nov.

Since Petkovski (1976) establishment of the genus *Nitocrellopsis* was based on the poor descriptions of *N. intermedia* (Chappuis, 1937), *N. elegans* (Chappuis & Rouch, 1959) and *N. ioneli* (Dumont & Decraemer, 1974), an emended diagnosis of the genus is given herein. Moreover, *Nitocrellopsis rouchi* sp. n. is designated as type-species to make available the generic name. The generic name *Nitocrellopsis* is used to maintain the stability of the nomenclature.

Diagnosis. Ameiridae. Body cylindrical, without clear demarcation between prosome and urosome. Hyaline frills of cephalothorax and somites bearing P2–P4 plain or minutely denticulate. Female genital field and first abdominal somite free or fused only ventrally. Anal operculum not prominent, denticulate or smooth. Caudal ramus cylindrical, elongate, or short, as long as wide, with 7 setae in both sexes. Sexual dimorphism in body size, antennule, P1 (inner basal spine), P5, P6 and genital segmentation. Rostrum small, not defined at base. Antennule slender and elongate; 8-segmented in female, with aesthetascs on segments 4 and 8; 10-segmented in male, with geniculation between segments 7 and 8, and aesthetascs on segments 5 and 10. Antenna with basis and proximal endopodal segment separate, with incomplete surface suture on posterior side. Antennary exopod 1-segmented, with 2 apical and 1 subapical setae. Mandibular palp uniramous, 2-segmented, basis with or without seta, endopod with 5 setae. Maxillule: endopod present or absent, if present with 1–2 setae; exopod absent. Maxillary syncoxa with 1–2 endites, proximal endite present or absent, if present with 1–2 setae. Allobasis drawn out into strong unipinnate claw, and 1 lateral accessory seta. Endopod reduced to a minute segment bearing 2 setae. Maxilliped subchelate. Syncoxal seta present or absent. Endopod represented by an unipinnate claw, accompanied by 1 short seta. P1–P3 with 3-segmented rami. P4 with 3-segmented exopod and 2-segmented endopod. P1–P4 exp-2 with 1 inner seta.; P1–P4 exp-3 with 2 outer spines. Armature formula P1–P4 as in Table 1. P5 with separate baseoendopod and exopod in both males and females; fused and not recognizable by setation in *N. intermedia*. Exopod with 4–5 setae (♀) and 4–5 setae (♂); baseoendopod with 2–3–4 setae (♀) and 1–2 setae (♂). Male P6 symmetrical.

Table 1. Armature formula P1–P4 in *Nitocrellopsis* species

	P1		P2		P3		P4	
	Exp	Enp	Exp	Enp	Exp	Enp	Exp	Enp
<i>N. elegans</i>	0.1.122	1.0.120	0.1.122	1.1.020	0.1.122	1.1.120	0.1.222	-1.020
<i>N. intermedia</i>	0.1.122	1.0.120	0.1.022	1.1.120	0.1.022	1.1.120	0.1.022	-1.120
<i>N. ioneli</i>	0.1.022	0.0.120	0.1.022	0.0.020	0.1.022	0.0.020	0.1.122	-0.020
<i>N. petkovskii</i> *	0.1.022	1.0.020	0.1.122	0.0.020	0.1.122	0.0.020	0.1.122	-0.020
<i>N. hellenica</i> #	0.1.022	1.0.120	0.1.022	0.0.010	0.1.022	0.0.020	0.1.122	-0.010
<i>N. hippocratis</i> °	0.1.022	1.0.120	0.1.022	0.0.010	0.1.022	0.0.020	0.1.122	-0.010
<i>N. rouchi</i> sp. n.	0.1.022	0.0.120	0.1.022	0.0.020	0.1.022	0.0.020	0.1.022	-0.020

*Re-examination of the holotype confirms the P1 endopodal setation of the original description (Rouch, 1987).

#Re-examination of the type-material revealed the presence of the short inner seta on P1 endopodal segment 3.

°In the original description (Cottarelli & Forniz, 1993), P1 endopodal segment 3 has been erroneously described with 2 setae and correctly figured with 3.

Type-species Nitocrellopsis rouchi sp. n.

Other species. *N. intermedia* (Chappuis, 1937) comb. nov., *N. elegans* (Chappuis & Rouch, 1959) comb. nov., *N. ioneli* (Dumont & Decraemer, 1974) comb. nov., *N. petkovskii* (Rouch, 1987) comb. nov., *N. hellenica* (Cottarelli & Forniz, 1993) comb. nov., *N. hippocratis* (Cottarelli & Forniz, 1993) comb. nov..

Nitocrellopsis rouchi sp. n.

Material examined. One ♀, holotype completely dissected and mounted in polyvinyl lactophenol; 2 ♂♂, paratypes, Well 2, water depth 350 cm, Grand Gravier (Givors), alluvial plain of the River Rhône (France); coordinates: 45° 35' 90" N, 4° 47' 38" E; altitude: 150 m a.s.l.; temperature: 19 °C; specific conductance: 289 $\mu\text{S cm}^{-1}$; dissolved oxygen: 1.9 mg/l; O₂ (%): 20%; pH: 7.38; phreatic biotope; September 1993; M.-J. Dole-Olivier & M. Creuze Les Chatelliers coll. Other material: One ♀, 2 ♂♂ (paratypes), Well 2, water depth 450 cm; temperature: 19 °C; specific conductance: 290 $\mu\text{S cm}^{-1}$; dissolved oxygen: 1.5 mg/l; O₂ (%): 16%; pH: 7.37. Twenty ♀♀, 10 ♂♂ processed for SEM, Well 3 (close to Well 2), water depth 450 cm, Grand Gravier (Givors), alluvial plain of the River Rhône (France); altitude: 154 m a.s.l.; temperature: 17 °C; specific conductance: 306 $\mu\text{S cm}^{-1}$; dissolved oxygen: 2.9 mg/l; O₂ (%): 30%; pH: 7.29; phreatic biotope; same date and same collectors.

Holotype and one male paratype deposited in the collections of the Natural History Museum, London; remaining material in the personal collection of D. Galassi at the Dipartimento di Scienze Ambientali, University of L'Aquila.

Additional material examined for Nitocrellopsis species and related genera

Nitocrellopsis ioneli. One ♀, 1 ♂, paratypes, personal collection of H. J. Dumont, University of Gent, Belgium. Additional material: 5 ♀♀, 1 ♂, phreatic waters (well) of Aourir, 10 km N from Agadir (Morocco).

Nitocrellopsis petkovskii. One ♀, holotype, ZMA (Zoological Museum, Amsterdam), reg. no. 102.783 (2 slides). Paratypes in the personal collection of R. Rouch no longer available (Rouch, pers. comm.).

Nitocrellopsis hippocratis. Two ♀♀, 1 ♂ paratypes, personal collection of V. Cottarelli, University 'La Tuscia', Viterbo, Italy.

Nitocrellopsis hellenica: One ♀ holotype, 1 ♀, 1 ♂ paratypes, personal collection of V. Cottarelli, University 'La Tuscia', Viterbo, Italy.

Nitocrella stochi. One ♂ holotype, 1 ♀ allotype, personal collection of G. L. Pesce, University of L'Aquila, Italy.

Nitocrella skyrensis. One ♀, 1 ♂, paratypes, personal collection of G. L. Pesce, University of L'Aquila, Italy.

Nitocrella rodiensis. Two ♀♀, paratypes, personal collection of G. L. Pesce, University of L'Aquila.

Nitocrella stammeri. Four ♀♀, Grave Grubbo Cave, Calabria, South Italy, personal collection of D. Galassi, University of L'Aquila, Italy.

Nitocrella kunzi. One ♀, 3 ♂♂ paratypes, personal collection of D. Galassi, University of L'Aquila, Italy.

Nitocrella pescei. Fifteen ♀♀, 20 ♂♂ paratypes, personal collection of D. Galassi, University of L'Aquila, Italy.

Parapseudoleptomesochra italica. One ♀ holotype, 2 ♀♀, 2 ♂♂ paratypes, personal collection of G. L. Pesce, University of L'Aquila, Italy.

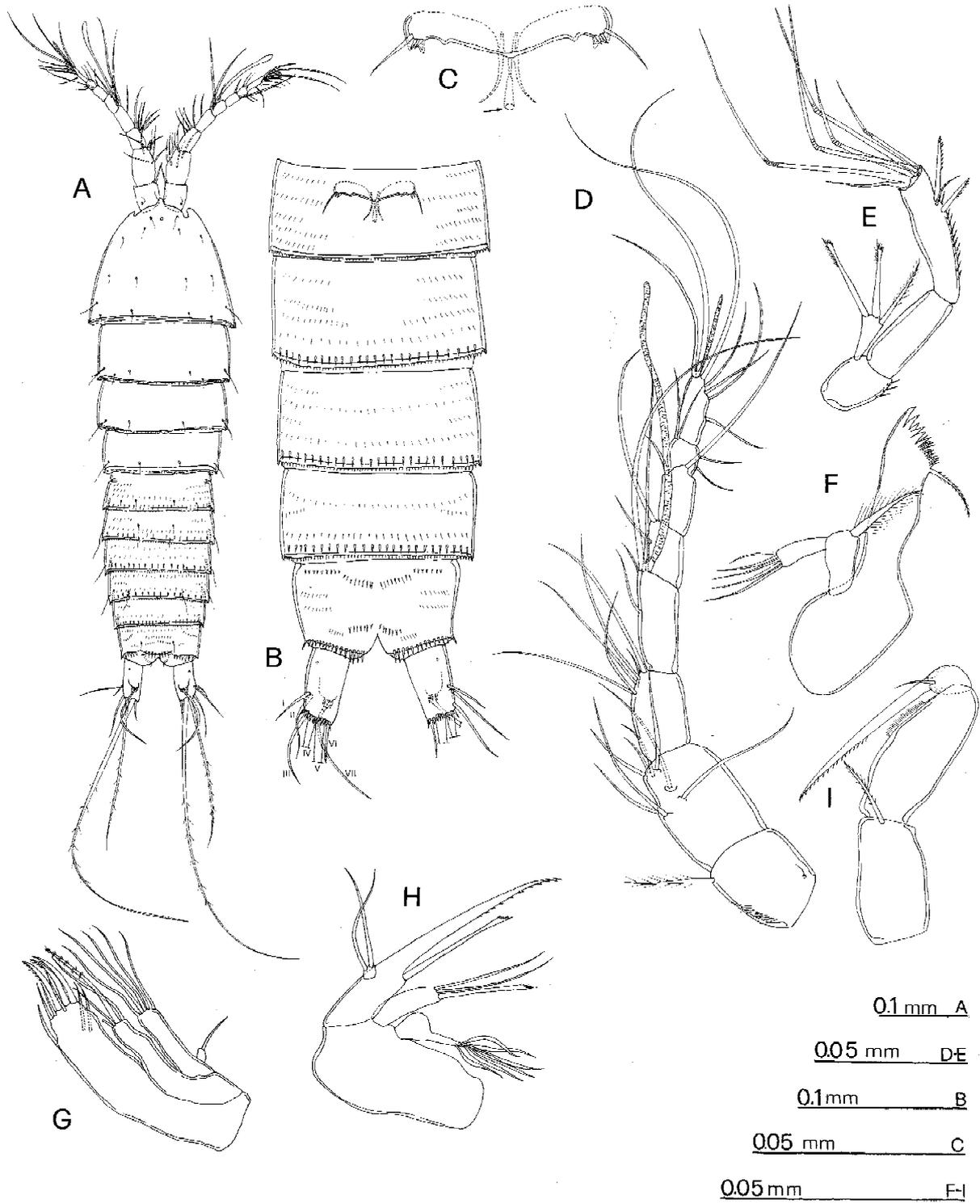


Figure 1. *Nitocrellopsis rouchi* sp. n. (♀ paratype): (A) habitus, dorsal view; (B) abdomen and caudal rami, ventral view; (C) genital field; (D) antennule; (E) antenna; (F) mandible; (G) maxillule; (H) maxilla; (I) maxilliped.

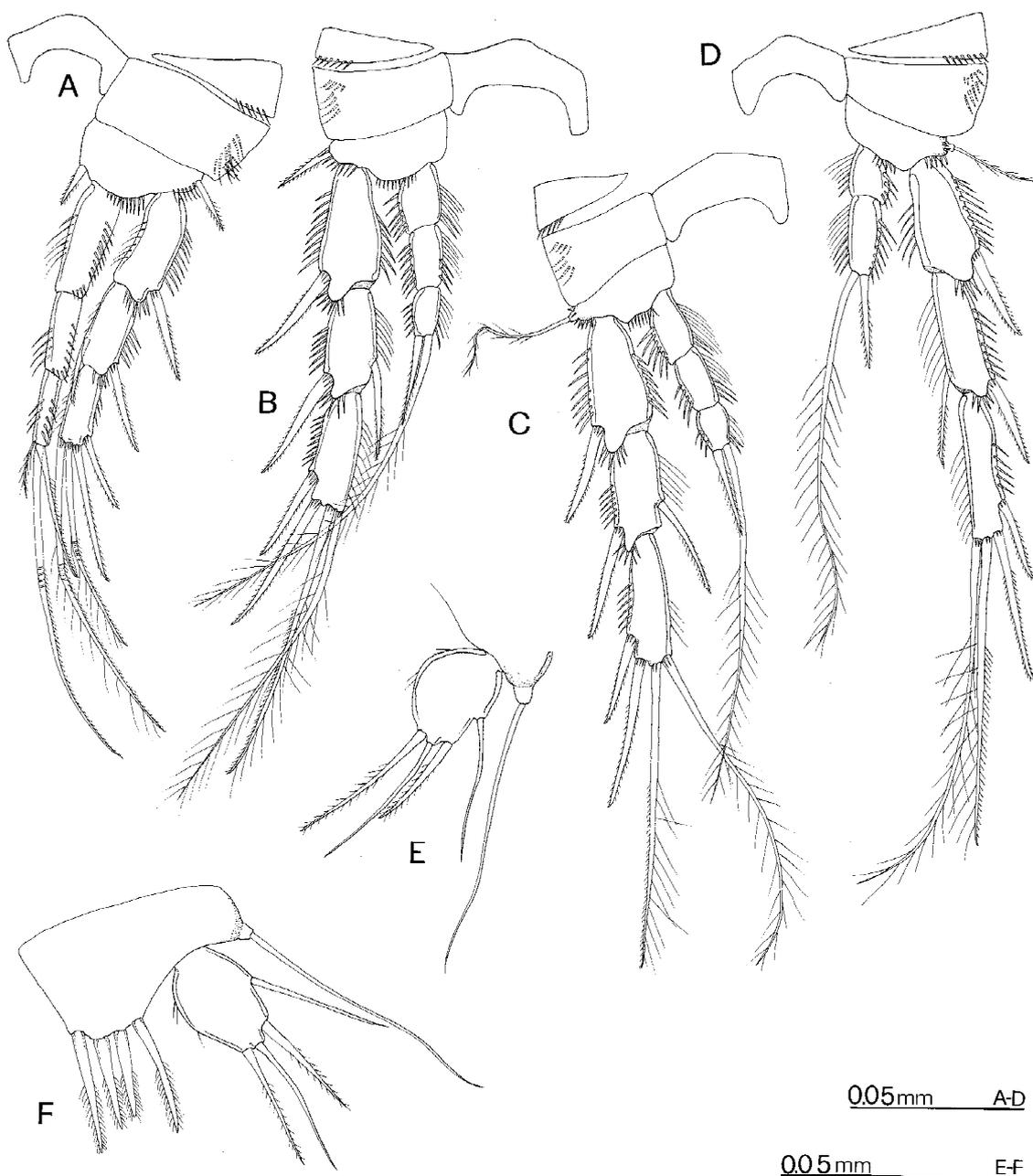


Figure 2. *Nitocrellopsis rouchi* sp. n.: (A) P1; (B) P2; (C) P3; (D) P4; (E) P5 exopod; (F) P5. A–D, holotype, E–F ♀ paratypes.

Nitokra hibernica. Several ♀♀ and ♂♂ from different localities of Italy and France, personal collection of D. Galassi, University of L'Aquila, Italy.

Description: Female (Figures 1, 2, 4 E–F and 5 A). Body length, excluding caudal setae, 708 μm (holotype), ranging from 506 to 708 μm ; by sum

of all body somites, ranging from 605 to 708 μm . Body cylindrical, unpigmented; integument without pitting, not strongly chitinized. Hyaline frill of cephalothorax smooth, minutely denticulate on P2–P4-bearing somites. Cephalothorax and somites bearing P2–P4 without surface ornamentation; sensillae pat-

terns as in Figure 1 A. P5-bearing somite with minute spinule rows both dorsally (Figure 1 A) and ventrally; with large paired pores (Figure 4 B) laterodorsally, and smaller paired pores lateroventrally. Abdominal somites with weakly incised hyaline frill both ventral and dorsally. Genital and first abdominal somites clearly distinct (Figure 1 A–B); ventral and dorsal surfaces of genital somite (Figure 1 A–B) with paired fine spinule rows. Genital field located near anterior margin of genital somite (Figures 1 B–C and 4 E), copulatory pore relatively small. Abdominal somites (Figure 1 A–B) with numerous rows of tiny spinules and continuous row of stronger spinules on posterior margins; with paired setules inserted on dorsal surfaces of all abdominal somites, except third abdominal somite (Figure 1 A–B). Anal somite (Figure 1 A) with dorsal and ventral spinule rows, and paired sensillae dorsally. Anal operculum (Figure 1 A) slightly convex, armed with numerous spinules on free distal margin. Caudal rami (Figure 1 A–B) cylindrical, 1.7 times as long as maximum width; hind margin with spinules ventrally, 1 secretory pore dorsally and 7 setae: seta I short; seta II and III about of the same length; seta IV and V well developed; seta VI minute and bare; seta VII long, arising from a small tubercle, with 3 spinules at its insertion on caudal ramus.

Rostrum (Figure 1 A) small, not defined at base, with 2 dorsal sensillae.

Antennule (Figure 1 D) elongate and slender, 8-segmented. Segment 1 with anterior spinule row and 1 sensilla posterolaterally. Armature formula: 1-[1 plumose], 2-[8], 3-[6], 4-[3 + (1 + ae)], 5-[2], 6-[2], 7-[4], 8-[5 + acrothek]. Apical acrothek consisting of 2 setae of different lengths fused basally to small aesthetasc. Aesthetascs on segments 4 and 8 of different ultrastructure (as for male, see Figure 4 C–D).

Antenna (Figure 1 E): Basis separate, with incomplete surface suture on posterior side; with row of spinules along abexopodal margin. Exopod 1-segmented, bearing 1 subapical plumose seta and 2 apical setae distally serrate. Endopod 2-segmented; segment 1 unarmed; segment 2 with a row of spinules along inner margin with spinule row; lateral armature consisting of 2 unipinnate setae and 1 slender, bare seta; apical armature consisting of 5 geniculate setae with longest, outer seta fused basally to small naked seta.

Mandible (Figure 1 F): Coxal gnathobase elongate, cutting edge with 5 coarse teeth and row of smaller teeth; 1 plumose seta at dorsal corner. Mandibular palp uniramous, 2-segmented; basis with 1 densely

plumose seta; endopod with 5 slender and bare setae apically.

Maxillule (Figures 1 G and 4 F): Praecoxal arthrite well developed, rectangular; with 2 anterior surface setae, 1 lateral and 3 bare apical setae and 3 strong, curved unipinnate spines. Coxal endite with 1 multipinnate and 3 slender setae. Basis with 5 long, slender setae apically. Endopod discrete segment bearing 1 naked seta. Exopod absent.

Maxilla (Figure 1 H): Syncoxa with 2 endites; proximal endite well developed, with 1 modified seta not defined at base, densely filamentous in distal third (Figure 5 A); distal endite with 1 apically serrate and 2 slender, bare setae. Allobasis drawn out into strong unipinnate claw; lateral accessory armature consisting of 1 apically serrate spine. Endopod reduced to minute segment bearing 2 naked setae.

Maxilliped (Figure 1 I) suchelate: Syncoxa with 1 plumose seta. Basis with row of fine spinules along inner margin in distal third. Endopod represented by distally unipinnate claw, accompanied by 1 short seta basally.

P1–P3 (Figure 2 A–C) with 3-segmented rami. P4 (Figure 2 D) with 3-segmented exopod and 2-segmented endopod. Intercoxal sclerites P1–P4 without ornamentation.

P1 (Figure 2A): Coxa with 2 posterior rows of spinules. Basis with 1 outer and 1 inner bipinnate spine with 2 anterior spinule rows. Exopod about as long as endopod; exp-1 and -2 with 1 outer pinnate spine, with spinule row along outer margins and row of long fine spinules along inner margins; exp-2 with 1 inner apically serrate, pinnate seta; exp-3 with 2 outer pinnate spines and 2 geniculate setae apically. Endopod, outer margins of all segments with spinule row, inner margins with long spinules; enp-1 about as long as exp-1; enp-2 unarmed; enp-3 with 2 geniculate setae apically and 1 slender plumose inner seta.

P2–P4 (Figure 2 B–D): Coxae with 2 spinule rows on posterior surface. Bases with 2 distal rows of spinules on anterior surfaces; P2 with outer pinnate spine, P3–P4 with outer plumose seta. P2–P4 exp-1 inner and outer margins with spinule rows; exp-2 and -3 outer margins with spinule rows, inner margins with long fine spinules. Exp-1 with 1 outer spine, exp-2 with 1 outer spine and 1 inner pinnate, apically serrate seta; exp-3 with 2 outer spines, 1 apical seta, pinnate on outer margin and plumose on inner margin, and 1 apical plumose seta. Endopod P2–P3 reaching to middle of exp-2. Each segment with outer row of spinules and inner row of long fine spinules; enp-1 and -2 unarmed,

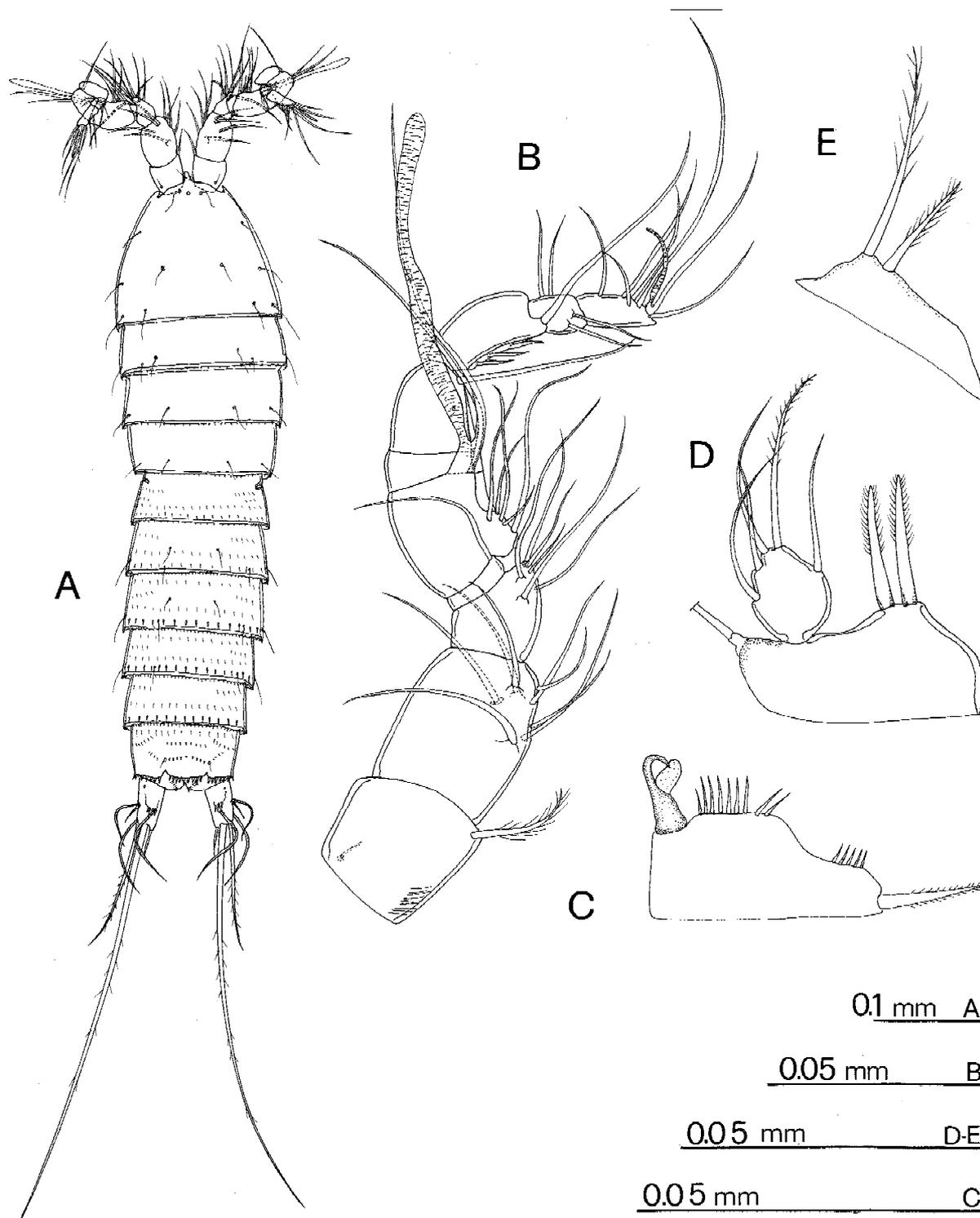


Figure 3. *Nitocrellopsis rouchi* sp. n. (♂ paratype): (A) habitus, dorsal view; (B) antennule (modified setae on segments 5–7 omitted); (C) P1 basis; (D) P5; (E) P6.

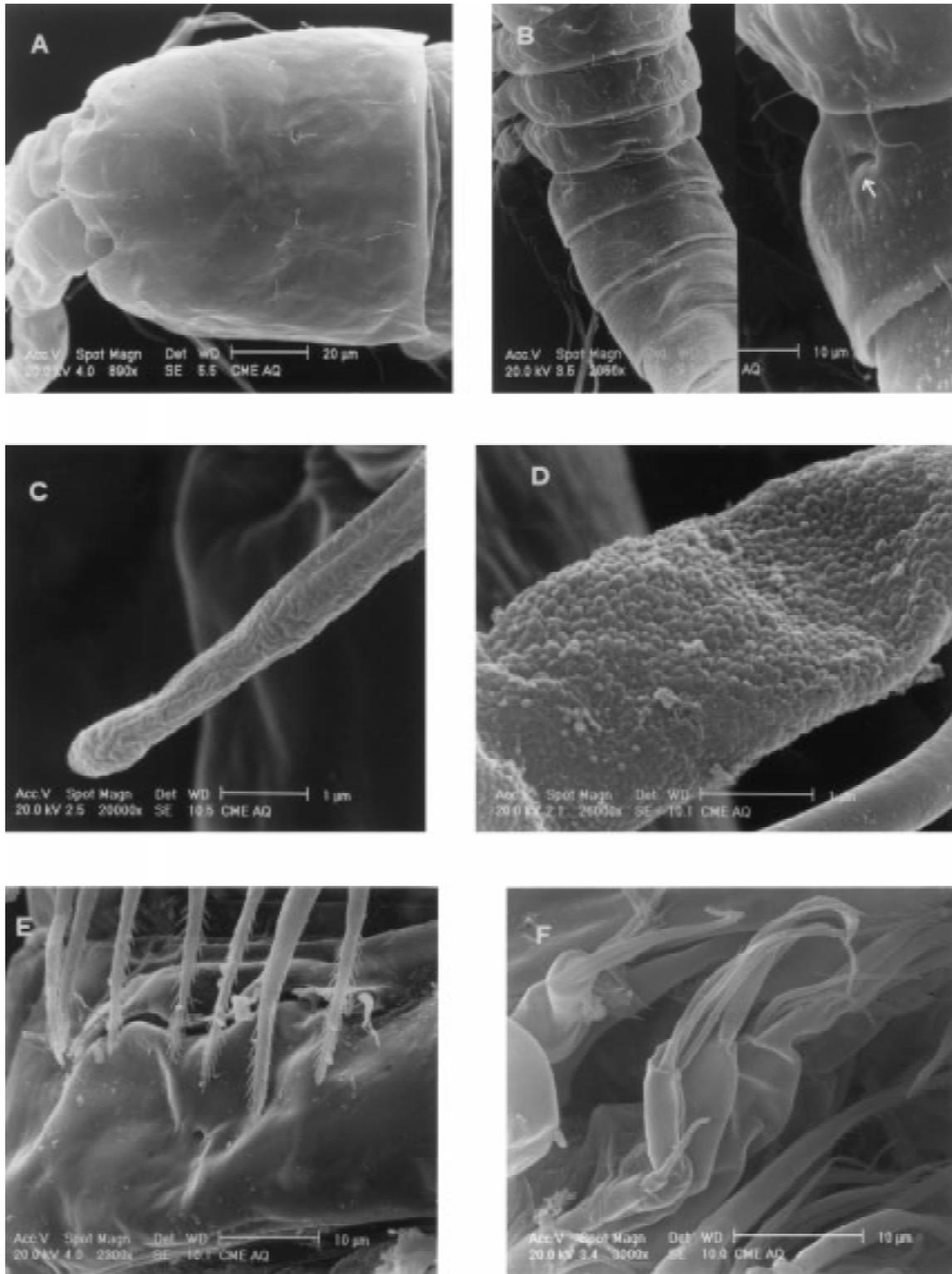


Figure 4. SEM micrographs of *Nitocrellopsis rouchi* sp. n.: (A) cephalothorax, dorsal view (♂); (B) P5-bearing somite (♀) (arrow indicates the dorso-lateral pore); (C) aesthetasc of antennule segment 10 (♂); (D) aesthetasc of antennule segment 5 (♂); (E) genital field; (F) maxillule (♀).

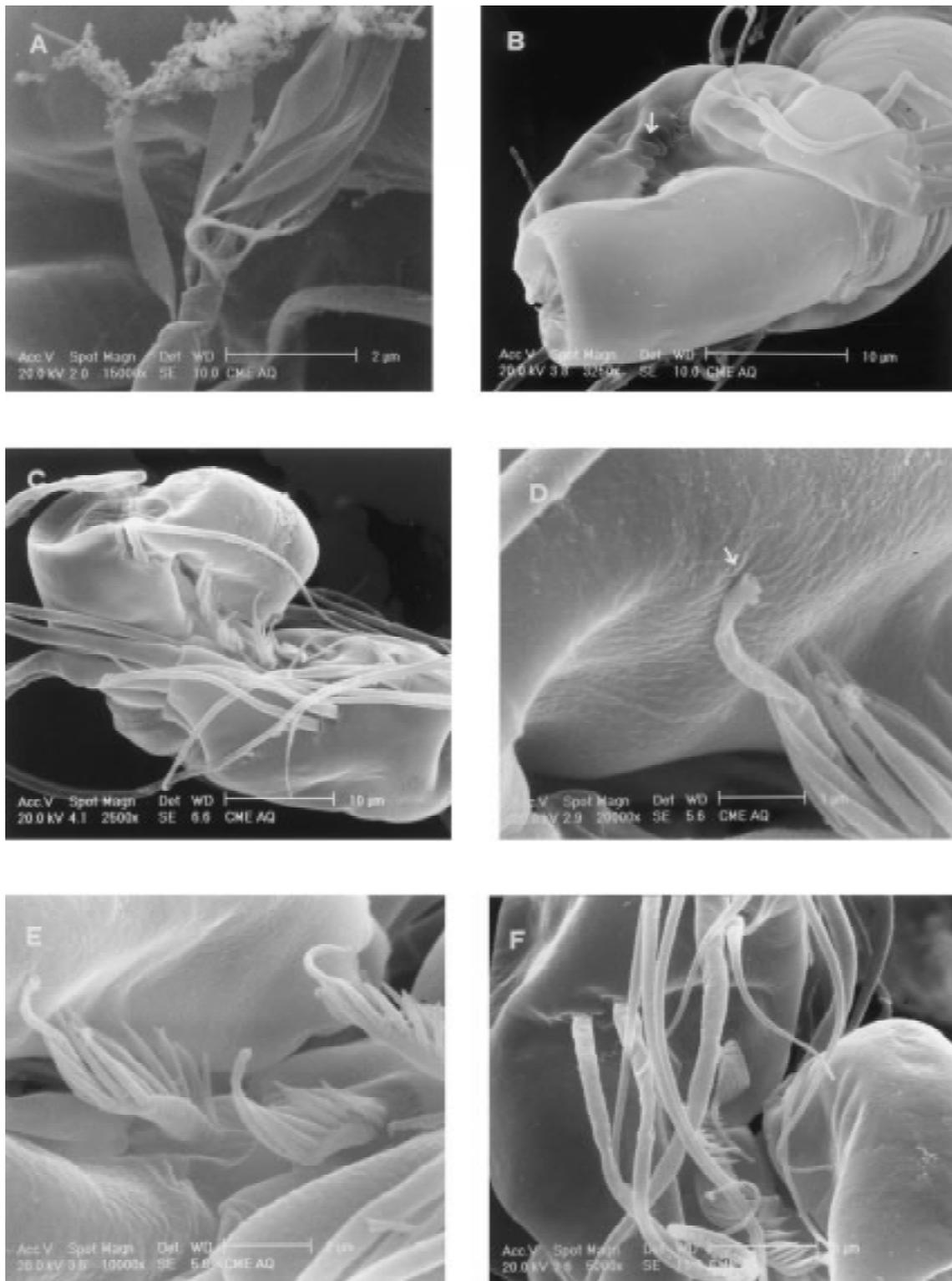


Figure 5. SEM micrographs of *Nitocrellopsis rouchi* sp. n.: (A) maxilla, densely plumose seta of proximal endite (♀); (B) antennule segment 8 (♂) (arrow indicates serrate ridge); (C) modified setae of antennule segments 5, 6 and 7 (♂); (D) antennule, tubular extension of modified seta (arrowed) (♂); (E) modified setae of segments 6 and 7 of antennule (lateral view) (♂); (F) same (frontal view) (♂).

enp-3 with 1 long plumose seta and 1 spine apically. P4 endopod 2-segmented, about as long as exp-1; enp-1 unarmed; enp-2 with 1 long plumose seta and 1 short spine. Detailed ornamentation as for endopods P2–P3. Armature formula P1–P4 as in Table 1.

P5 (Figure 2 E–F): Baseoendopod not prominent, with 4 bipinnate spines; outer basal seta slender and bare. Exopod oval (length/width ratio: 1.35–1.43); some variation in the shape, being the inner margin more or less bulgy (Figure 2 E–F); inner margin with few spinules; with 4 setae, outer margin with 1 slender and 1 pinnate seta, distal margin with 1 bare and 1 bipinnate seta.

P6 rudimentary (Figure 1 B–C), armed with 1 naked seta and 2 spinous processes (not defined at base). Copulatory pore small, arrowed in Figure 1 C.

Male (Figures 3, 4 A–D and 5 B–F): Body length, excluding caudal setae, ranging from 530 to 687 μm . General ornamentation of body somites and caudal rami as in female (Figure 3 A). Sensillae patterns on cephalothorax less dense (Figure 4 A) than in female. P5-bearing somite with large, paired pores, as in female. Sexual dimorphism in body size (although not marked), antennule, P1, P5 and P6.

Antennule (Figure 3 B): Ten-segmented. Segment 1 with 1 anterior spinule row and 1 posterolateral sensilla. Segment 4 represented by small U-shaped sclerite. Armature formula: 1-[1], 2-[8], 3-[6], 4-[1], 5-[1 modified + 6 bare + (1 + ae)], 6-[2 modified], 7-[1 bare + 1 modified], 8-[1 bare + 3 modified], 9-[4], 10-[5 + acrothek]. Aesthetascs on segments 5 and 10 of different ultrastructure (Figure 4 C–D). Modified setae of segments 5, 6 and 7 (Figure 5 C–F) possibly with sensory function, bipinnate with tubular extension. Modified setae on segment 8 fused basally to segment, robust elements, with axial slit. Segments 7 and 8 with lateral series of serrate ridges (Figure 5B) possibly involved with mate grasping.

P1 inner basal spine modified (Figure 3 C). P5 (Figure 3 D) baseoendopod not prominent, with basipodal outer seta slender and bare, and 2 endopodal bipinnate setae. Exopod oval in shape, shorter than in female; with 1 inner seta, 2 apical setae (innermost plumose), 1 subapical and 1 naked outer seta. P6 (Figure 3 E) symmetrical, chitinous lamellar plate with 2 bipinnate setae.

Etymology: The specific name is in honour of Dr Raymond Rouch (Fronton, France) for his invaluable contribution to the systematics, ecology and biogeography of groundwater harpacticoids.

Discussion

The genus *Nitocrellopsis* was established by Petkovski (1976) to accommodate three *Nitocrella* species characterized by 3-segmented endopods P2–P3 and 2-segmented endopod P4. At present, *Nitocrellopsis* comprised of 6 species (excluding *N. rouchi* sp. n.); *N. intermedia* (Chappuis, 1937) from various groundwater habitats of the Balkan Peninsula (phreatic interstitial, karstic springs of Bulgaria and Yugoslav Macedonia); *N. elegans* (Chappuis & Rouch, 1959) from epiphreatic cave waters of the Atlantic Pyrenean Massif (France); *N. ioneli* (Dumont & Decraemer, 1974) from a saline spring, Ziz valley (Morocco); *N. petkovskii* (Rouch, 1987) from phreatic waters near Béchar (Algeria); *N. hellenica* (Cottarelli & Forniz, 1993) from coastal phreatic waters of the island Kos (Greece), and *N. hippocratis* (Cottarelli & Forniz, 1993) from coastal phreatic waters of the islands Kos and Tilos (Greece). Within this genus, the new species can easily be distinguishable by the armature of P5 in both the male and female, and by the unique armature formula of the P1–P4 (Table 1). *Nitocrellopsis rouchi* sp. n. shows some affinities with *N. ioneli* in the identical armature of both rami of P1–P3 and endopod of P4, and P5 baseoendopod of both sexes. The lack of an inner seta on P1 endopod segment 1, together with the presence of 4 elements on female P5 baseoendopod in *N. rouchi* sp. n. are unique characters shared only with *N. ioneli*. The new species differs from *N. ioneli* in the armature of the P4 exopod segment 3 (with 5 elements in *N. ioneli* vs. 4 in *N. rouchi* sp. n.), and in the setation of P5 exopod in both the female (5 in *N. ioneli* vs. 4 in *N. rouchi* sp. n.) and the male (4 in *N. ioneli* vs. 5 in *N. rouchi* sp. n.).

Nitocrellopsis rouchi sp. n. possesses enigmatic large dorso-lateral pores (Figure 4 B) located on the P5-bearing somite. A wider check for the same character in other freshwater ameirids revealed that this feature is currently displayed by freshwater *Nitokra*, *Parapseudoleptomesochra*, *Nitocrella* and *Nitocrellopsis* species, although its role is still unknown. Interestingly these structures have never been described or figured for marine ameirids.

In *Nitocrellopsis rouchi* sp. n., the genital somite is clearly distinct from the first abdominal somite, while in *Nitocrellopsis ioneli* it is fused ventrally only with the first abdominal somite. The genital somite and the first abdominal somite are also completely free in *Nitocrella stammeri* Chappuis, 1938, *N. rhodiensis* Pesce, 1983, *N. pescei* Galassi & De Laurentiis,

1997, *Nitocrellopsis petkovskii*, *N. hippocratis* and *N. hellenica*. *Nitokra hibernica* (Brady, 1880), *Nitocrella kunzi* Galassi & De Laurentiis, 1997 and *Parapseudoleptomesochra italica* Pesce & Petkovski, 1980 also show partial (ventrally only) fusion as in *Nitocrellopsis ioneli*. The phylogenetic significance of the presence vs. absence in harpacticoids of the genital double-somite is difficult to assess (Huys & Boxshall, 1991), and the presence of different conditions within a single genus further compounds the problem. Nevertheless, it is remarkable that some ameirids also possess free genital and first abdominal somites, the condition of a genital double-somite could represent the derived state. It can also be postulated that the 'free' somite condition, primarily present in ancient marine ancestors, has been maintained by some stygobiont descendants, due to the conservative role of the subterranean environment. Moreover, separate somites could facilitate body movements 'between grains' in the same way as the additional 'pseudo-somite' (Huys & Boxshall, 1991; Boxshall et al., 1993) does which is found in some interstitial cyclopoids and harpacticoids. In summary, among ameirids three different states exist: 1. genital and first abdominal somites free; 2. somites fused only ventrally; 3. complete fusion both dorsally and ventrally forming a genital double-somite. With current knowledge, it is difficult to polarize this character. Consequently the hypothesis of post-displacement of fusion between somites, as argued by Fiers (1990) for the Cancrincolidae, and extended by Huys & Boxshall (1991) for the latiremids and a tetragonicipitid is not necessarily applicable to the Ameiridae.

Additional differences between *Nitocrellopsis rouchi* sp. n. and its congeners were also found in the cephalic appendages, such as: segmentation of the male antennule; morphology of the antenna; structure and morphology of the mouthparts. It is difficult to give the correct weight to cephalic appendage characters due to discrepancies in the original descriptions. For example, the male antennule of *N. rouchi* is 10-segmented. Dumont & Decraemer (1974) illustrate the male of *N. ioneli* with an 8-segmented antennule. Examination of type-material of the latter and additional material of *N. ioneli* from Agadir, Morocco (not topotype), revealed the presence of a 10-segmented male antennule. Observations made on other ameirid genera demonstrated the presence of U-shaped antennular segment 4 and hence its frequent omission in many previous descriptions. This is the case for *Nitocrella stochi* Pesce & Galassi, 1986; *Nitocrella*

pescei; *Nitocrella kunzi*; *Parapseudoleptomesochra italica*; *Nitocrellopsis hellenica* and *Nitocrellopsis hippocratis*. The same situation is recognisable also in *Nitocrella skyrensis* Pesce, 1982, although not described and figured in the original description (Pesce, 1982). Another feature that requires attention is the antennary morphology. The incomplete fusion between the basis and first endopod segment can be observed on the posterior surface in both *N. rouchi* sp. n. and *N. ioneli*. Comparative observations of other freshwater stygobiont ameirid species revealed different degrees of fusion between these same segments, but never a typical allobasis (Galassi & De Laurentiis, unpubl.). It appears that stygobiont ameirids tend to maintain the ancestral condition of distinct basis and first endopodal segment, and that ameirids belonging to the same genus sometimes possess a graded series of fusion. For instance, *Parapseudoleptomesochra trisetosa* (Krishnaswamy, 1957) is described and figured by Mielke (1997) with a clear allobasis, while the re-examination of the type-material of *Parapseudoleptomesochra italica* revealed the presence of a distinct antennary basis and first endopodal segment. A similar situation is summarised by Mielke (1997) for the *Nitokra* species, but both *Parapseudoleptomesochra* and *Nitokra* need better definition of their generic boundaries.

With regard to mouthparts, different structures and related armature elements can be recognised within the genus *Nitocrellopsis*, as well as in *Nitocrella* and *Parapseudoleptomesochra*. Characters possibly overlooked and/or misinterpreted in previous descriptions are considered here with caution, and our observations are based on re-examination of type-material whenever possible. The type-material of *Nitocrellopsis elegans* and *N. intermedia* no longer exists (Rouch, in litt.), but the re-examination of the mouthparts from the type-series of *N. ioneli* and additional specimens collected from Morocco and the type-material of *N. petkovskii*, *N. hellenica* and *N. hippocratis*, enabled the original description to be confirmed and/or amended. Several differences in the cephalic appendages between species (Table 2) were also revealed. The significance of these is difficult to assess at present, although Conroy-Dalton & Huys (1996) emphasised the role of these characters in phylogenetic analyses.

Distribution and ecology. The genus *Nitocrellopsis* may have an ancient (Tethyan) origin, showing a distribution confined to the proto-Mediterranean basin. The lack of records from other zoogeograph-

Table 2. Mouthparts structure and setation in *Nitocrellopsis* species

	Md plp basis	Mx1 enp	Mx1 enp setation	Mx2 proximal endite	Mx2 proximal endite setation	Mxp syncoxa
<i>N. elegans</i>	?	?	?	?	?	?
<i>N. intermedia</i>	?	?	?	?	?	?
<i>N. ioneli</i>	no setae	present	2 setae	present	2 setae	1 seta
<i>N. petkovskii</i> *	no setae	absent	/	absent	/	no seta
<i>N. hellenica</i>	1 short seta	present	2 setae	present	2 setae	1 seta
<i>N. hippocratis</i>	1 short seta	present	2 setae	present	?	1 seta
<i>N. rouchi</i> sp. n.	1 long seta	present	1 seta	present	1 seta	1 seta

*The re-examination of the holotype confirms the original description (Rouch, 1987) of mouthparts.

ical regions means that more detailed investigations in groundwaters are needed for these lesser known regions, as for the genus *Nitocrella* (Galassi & De Laurentiis, 1997). *Nitocrellopsis* consists exclusively of stygobiont species, predominantly linked to interstitial phreatic waters. *N. rouchi* sp. n. was found in phreatic waters at depths ranging from 50 cm to 650 cm, with the most consistent distributions at 550–650 cm depth layer. This distribution is consistent with other biological parameters, this depth layer corresponds to the transition zone from coarse to fine-sized sediments, and is a storage zone for organic matter and bacterial activity (Gibert, 1994). Other stygobiont crustaceans of the interstitial community are represented by *Parastenocaris* sp. (Copepoda, Harpacticoida); *Crangonyx subterraneus* Bate, 1859 and *Niphargus* sp. (Amphipoda); and *Proasellus* and *Microcharon* species (Isopoda).

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Key to species of *Nitocrellopsis*: The following key is applicable to both sexes unless otherwise stated.

- | | | |
|----|---|-------------------------|
| 1. | P1-exp 3 with inner seta, with armature formula 122; P3 enp-3 with 3 setae/spines | 2 |
| | P1-exp 3 without inner seta, with armature formula 022; P3 enp-3 with 2 setae/spines | 3 |
| 2. | P5 with fused baseoendopod and exopod, not recognizable by setation | <i>N. intermedia</i> |
| | P5 with distinct baseoendopod and exopod | <i>N. elegans</i> |
| 3. | P2–P3 exp-3 with inner seta; P1 enp-3 with 2 setae/ spines | <i>N. petkovskii</i> |
| | P2–P3 exp-3 without inner seta; P1 enp-3 with 3 setae/spines | 4 |
| 4. | P4 exp-3 with inner seta | 5 |
| | P4 exp-3 without inner seta | <i>N. rouchi</i> sp. n. |
| 5. | P2 enp-3 and P4 enp-2 with 2 setae/ spines; ♀ P5 exopod with 5 setae/spines, baseoendopod with 4 spines | <i>N. ioneli</i> |
| | P2 enp-3 and P4 enp-2 with 1 spine; ♀ P5 exopod with 4 setae/spines, baseoendopod with 3 spines | 6 |
| 6. | Caudal rami cylindrical, about 2 times as long as maximum width | <i>N. hellenica</i> |
| | Caudal rami conical, about 1.3 times as long as maximum width | <i>N. hippocratis</i> |

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