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INGOLFIELLA BEATRICIS, NEW SPECIES (AMPHIPODA: INGOLFIELLIDAE) FROM SUBTERRANEAN WATERS OF SLOVENIA

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A B S T R A C T

A new species of ingolfiellid Amphipoda, *Ingolfiella beatricis*, collected from groundwater in a cave near Ljubljana in Slovenia is described. It is the first ingolfiellid, possessing developed "ocular lobes", found in inland fresh groundwater and is therefore of biogeographic interest. Its morphological characters suggest a marine origin, a supposition that is strengthened by the marine relationships of accompanying fauna.

On the Balcan Peninsula four species of the genus *Ingolfiella* (*sensu lato*) are known today. They all occur on the southern part of the peninsula: *I. acherontis* (S. Karaman, 1933) from the groundwater in the vicinity of Skopje (Macedonia); *I. petkovskii* S. Karaman, 1957, Elkhovo (Bulgaria) and Keramou (Euboea, Greece); *I. macedonica* S. Karaman, 1959, in the underflow of the river Pcinja (surroundings of Skopje, Macedonia); *I. vandeli* Bou, 1970, in groundwater near river sediments of Aitolia and the Peloponnisos, Greece (see Bou, 1970).

According to the systematics proposed by Ruffo and Vigna Taglianti (1989), those four species can be placed in the subgenera *Balcanella* S. Karaman, 1933 (*I. acherontis, I. macedonica*) and *Gevgeliella* S. Karaman, 1959 (*I. petkovskii, I. vandeli*), both of which are characterized by the absence of "ocular lobes", as in all other ingolfiellids known from strictly freshwater habitats.

During the collection of aquatic fauna from a cave in Slovenia, conducted by Beatrice Sambugar (Verona) and Fulvio Gasparo (Triest), a single specimen of *Ingolfiella* was found. Subsequently this cave has been visited several times, but despite meticulous search no other specimens were found.

The locality of this cave is particularly interesting from a biogeographic viewpoint because it is situated on the outer eastern borders of the Julian Alps and in the hydrographical basin of the Sava River, the right affluent arm of the Danube.

Although based only on one specimen, we have decided to describe this species because it is the first record of *Ingolfiella* in the Alpine region and the first ingolfiellid with well-developed "ocular lobes" to be found in underground freshwater.

MATERIALS AND METHODS

The specimen examined was collected in a small stream in the cave of Pajsarjeva Jama, Pajsar, Vrhnika, Slovenia (45°59′51″N, 14°16′15″E). Collection was accomplished by upturning the sand and lime sediment upstream of a handheld net which was to filter out the animals carried downstream by the flowing water. The temperature of the water was 9.9°C and the conductivity microSiemens 240/cm. Accompanying fauna consisted of Oligochaeta (Tubificidae Phallodrilinae gen. sp., Parvidrilidae gen. sp.); Gastropoda (*Graziana pupula*); Copepoda Harpacticoida (*Bryocamptus* (*Limocamptus*) *dacicus* s.l., *Bryocamptus* (*Rheocamptus*) *balcanicus, Ceuthonectes serbicus, Elaphoidella cvetkae, Elaphoidella jeanneli, Parastenocaris gertrudae*); Copepoda Cyclopoida (*Diacyclops belgicus*); Amphipoda (*Gammarus* sp., *Niphargus* cfr. *minor, Niphargus stygius, Synurella ambulans*), and some unidentified Rotifera and Nematoda.

The specimen was first immersed in glycerin and drawn *in toto*, then dissected and permanently mounted in Faure's medium on glass slides. Drawings of the separate appendages were made with a camera lucida attached to a phase contrast compound microscope.

SYSTEMATICS

Family Ingolfiellidae
Ingolfiella beatricis, new species
Figs. 1–3

Material Examined.—Holotype (MVRCr 387), probably female (2 mm) collected 16 May 1998 in Pajsarjeva Jama, Pajsar, Vrhnika, Slovenia. The dissected holotype (slides 4225, 4226) is deposited in the Museum of Natural History, Verona, Italy.

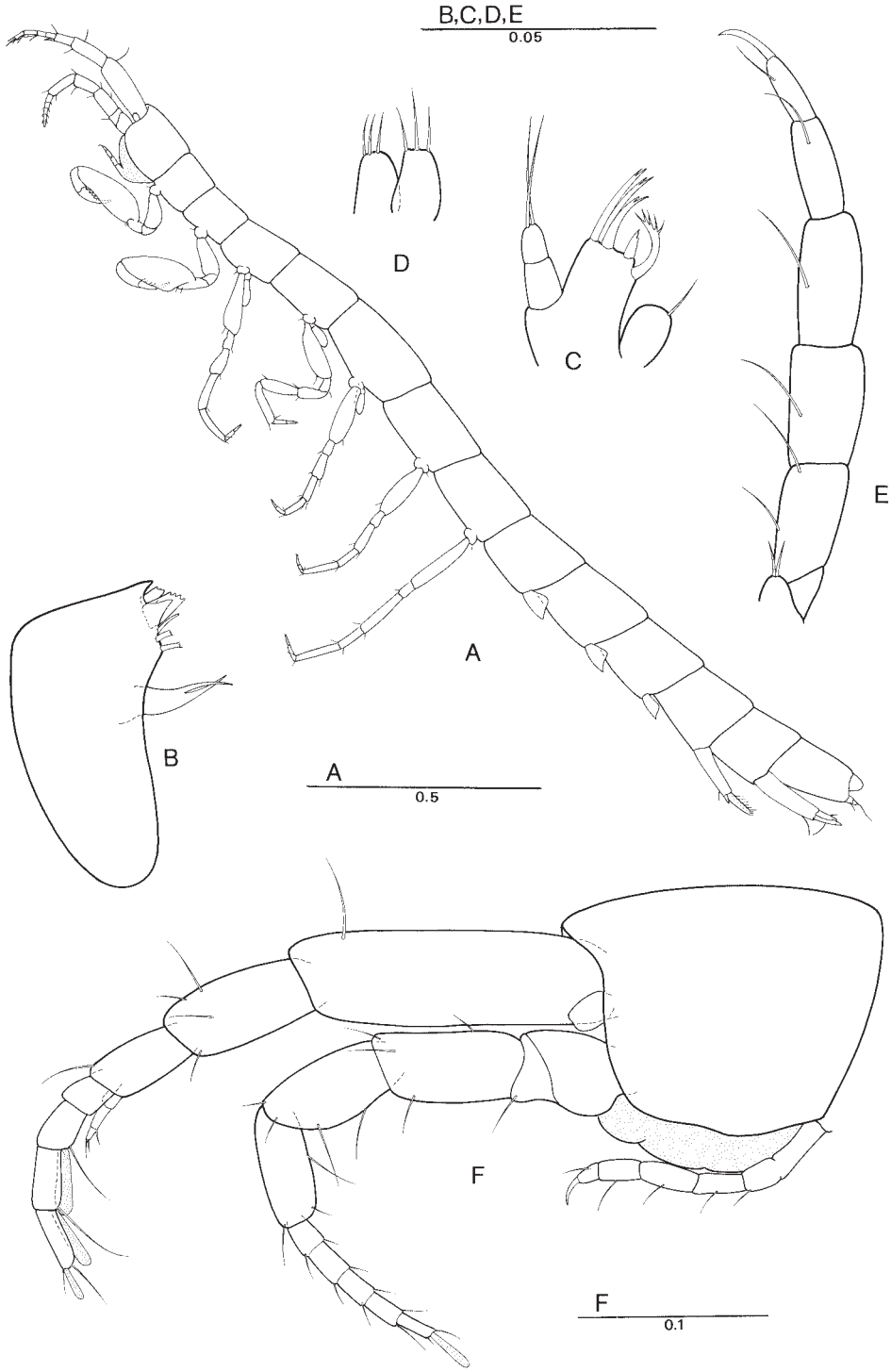

Fig. 1. *Ingolfiella beatricis* n. sp. (?female), Pajsarjeva Jama (Slovenia). A, habitus; B, mandible; C, D, maxilla 1, 2; E, maxilliped; F, head with antennae 1, 2. (Scales in mm.)

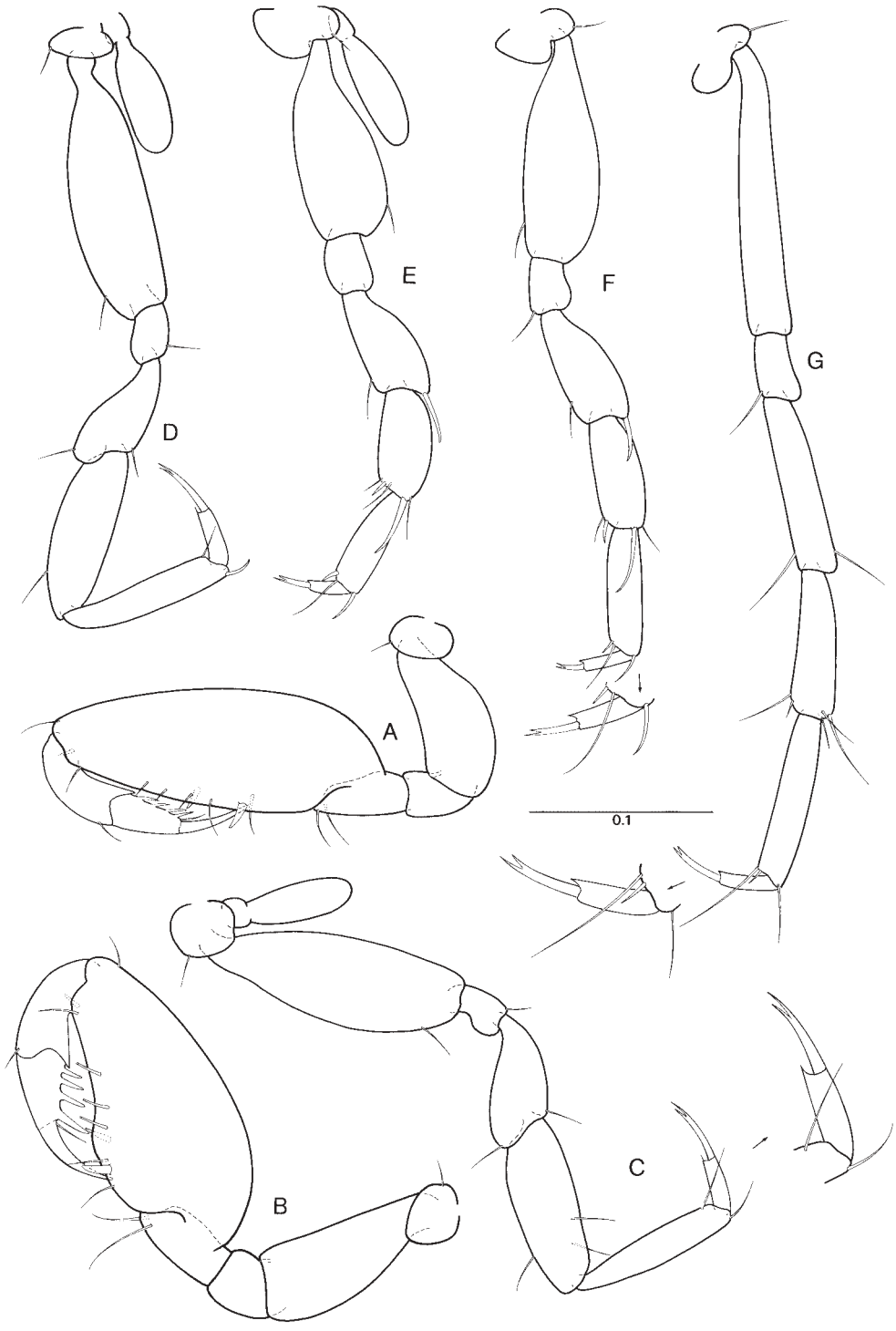

Fig. 2. *Ingolfiella beatricis* n. sp. (?female), Pajsarjeva Jama (Slovenia). A, B, gnathopods 1, 2; C, D, pereiopods 3, 4; E–G, pereiopods 5–7. (Scale, all figures 0.1 mm.)

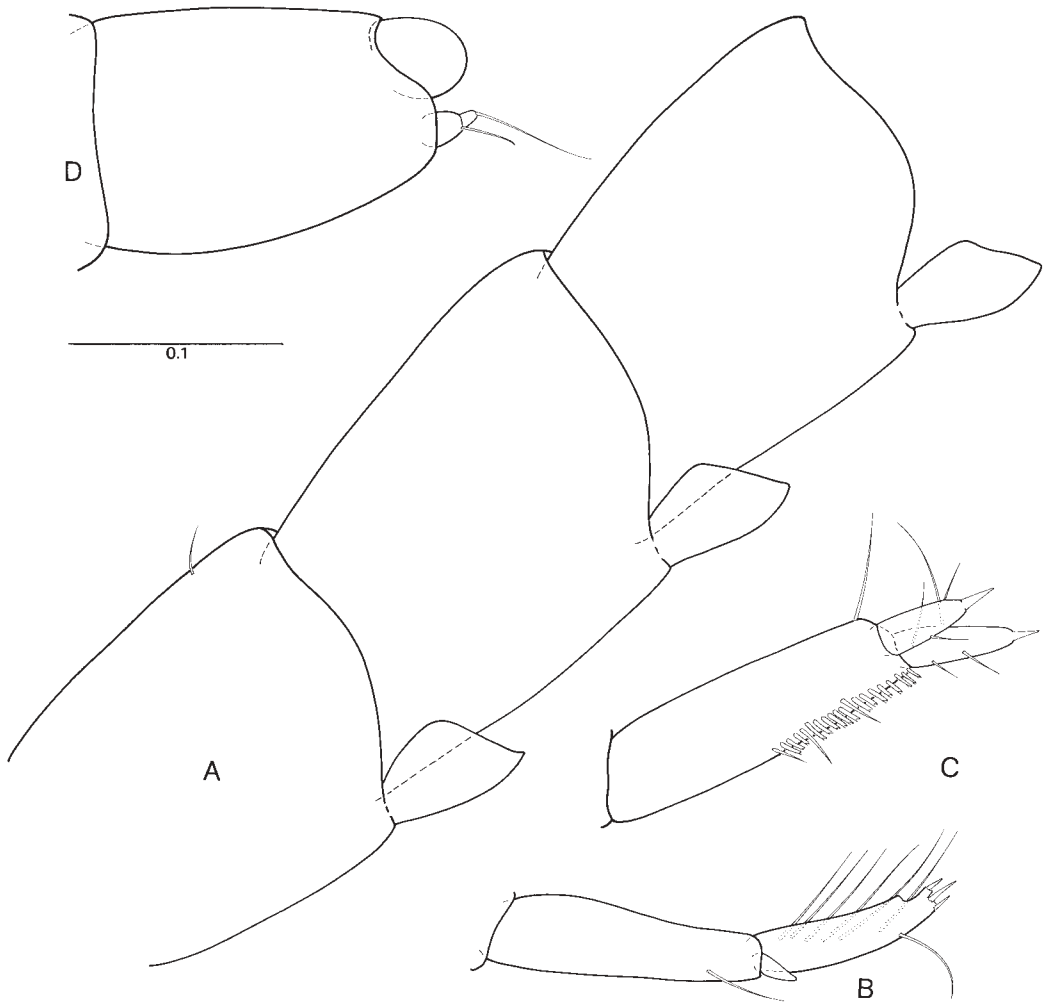

Fig. 3. *Ingolfiella beatricis* n. sp. (?female), Pajsarjeva Jama (Slovenia). A, pleosom with pleopods 1–3; B, C, Uropods 1, 2; D, urosomite 3 with uropod 3 and telson. (Scale, all figures 0.1 mm.)

Diagnosis.—Ingolfiellidae, ocular lobes developed. Maxilla 1, outer plate with 5 spines. Oostegites not observed. Gnathopods 1, 2 with smooth palm, dactyli with 4 elongate spiniform processes. Pereiopods 3–7 with dactyli similar, with proximal part ending in an interior acutely pointed spur, claw slender, finely bifid distally. Pleopods 1–3 present, subtrapezoidal, similar. Uropod 1 with outer ramus about ¼ length of inner ramus, uropod 2 peduncle long, with 3 diagonal rows of short distally bifid spines.

Description.—Female (?) 2 mm. Body very elongate, all segments laterally compressed (Fig. 1A). Head with weakly protruding rostrum, apically rounded, lateral margin rounded, "ocular lobes" developed, suboval. Pereionite 1 shorter than head, pereionites 2–7 increasing in length, pereionites 5–7 markedly longer than deep; pleonites 1–3 and urosomites 1, 2 subtrapezoidal, urosomite 3 subcylindrical, markedly longer than deep.

Antenna 1: peduncular (Fig. 1F) article 1 longer than articles 2+3; flagellum 4-articulate, shorter than peduncle, articles 2–4 with 1 aesthetasc, accessory flagellum 3-articulate, shorter than flagellar articles 1+2. Antenna 2 (Fig. 1F) subequal in length to antenna 1, flagellum 5-articulate, article 5 with 1 apical aesthetasc.

Mandible (Fig. 1B) without palp, incisor with teeth, lacinia mobilis distally denticulate, spine row with 3 denticulate spines, molar formed as long, pointed, partially bifid process. Maxilla 1 (Fig. 1C) palp 2-articulate, subequal in length to outer plate, with 2 distal setae; outer plate with 5 spines, inner spine strongly curved, distally denticulate, second strong, short and bifid, third to fifth spines elongate, with subapical denticle; inner plate rounded quadrangular, with 1 distal seta. Maxilla 2 (Fig. 1D) with 3 distal spiniform setae on every plate. Maxilliped (Fig. 1E) basally fused, only basal lobes present, with 2 distal setae; palp with 5 articles, article 1 with 2 elongate setae, articles 2–4 each with single elongate seta, dactylus with long, falcate claw.

Coxal plates 1–4 small (Fig. 2A–D), subquadrate, distally rounded, with 1 anterior seta; coxal plates 5–7 lobed, posterior lobe shorter than anterior one, with 1 posterior seta. Gills on pleon segments 3–5. Oostegites not observed (absent?).

Gnathopod 1 (Fig. 2A) carposubchelate, carpus strong, ovato-elongate, palm feebly convex, smooth, with 3 setae finely bifid apically, and 1 submedial spine on inner face, palm having 1 strong spine and 1 short triangular process on inner face, dactylus with 4 slender spiniform processes, claw slender, elongate. Gnathopod 2 (Fig. 2B) carposubchelate, dissimilar from gnathopod 1, carpus subpiriform, stronger than in gnathopod 1, palm oblique, distally concave, smooth, with 3 setae finely bifid apically and 1 submedial spine on inner face, palm defined by 1 strong spine, dactylus stronger than in gnathopod 1, with 4 elongate spiniform processes, claw elongate, falciform; c/p index (see Stock, 1977) = 1.9.

Pereiopods 3, 4 (Fig. 2C, D) similar, dactylus longer than ½ propodus, basal part ending in acutely pointed internal spur, claw slender, as long as basal part of dactylus, finely bifid distally. Pereiopods 5–7 (Fig. 2E–G) progressively longer. Pereiopods 5, 6 (Fig. 2E, F) basis piriform-elongate, merus, carpus, and propodus with 1 distoposterior curved spine, merus and carpus inflated, dactylus similar to those of pereiopods 3, 4, but relatively shorter, claw bifid distally, shorter than basal part of dactylus. Pereiopod 7 distinctly longer than pereiopods 5, 6, basis elongate, linear, merus and carpus not inflated, dactylus similar to those of pereiopods 5, 6, but longer, claw bifid distally, as long as basal part of dactylus.

Pleopods 1–3 (Fig. 3A) subtrapezoidal, with disto-anterior corner finely pointed, without setae, similar, but pleopod 3 slightly narrower than pleopods 1–2.

Uropod 1 (Fig. 3B) peduncle longer than inner ramus, with 1 subdistal ventral seta; outer ramus very short, about ¼ length of inner ramus, without setae; inner ramus subdistally emarginate, distally truncated, with 3 distal spines and row of 6 long setae on inner face. Uropod 2 (Fig. 3C) as long as urosomite 3, peduncle length about twice the rami, with 1 long distoventral seta and with 3 diagonal rows of strong spines, truncated and shortly bifid, on inner face, every row preceded and finished by 1 normal elongate spine; outer ramus slightly shorter than inner, rami distally constricted and apically pointed. Uropod 3 (Fig. 3D) very short, peduncle with 1 distal seta, ramus shorter than peduncle, with 1 long distal seta. Telson fleshy, subglobular, apparently without setae.

Etymology.—The new species is named after Dr. Beatrice Sambugar, one of the collectors of the specimen during her research on subterranean Oligochaetes.

Remarks.—*Ingolfiella beatricis* is thought to have a marine origin. It was reported by B. Sambugar that in the same cave where *I. beatricis* was found, two species of Tubificidae Phallodrilinae occurred, whose relatives are in the marine environment (Sambugar *et al.*, 1999). It seems that the aquatic fauna of the Pajsarjeva Jama is a mixture of freshwater and marine elements. This may be confirmed by the observation that *I. beatricis* is similar to a group of species with oligohalyne occurence, e.g., the subgenus *Antilleella* Ruffo and Vigna Taglianti, 1989, that is present in the Atlantic, the Antilles, Canary Islands, and Madeira Island, with a probably tethian distribution (see Discussion).

Discussion

Ruffo and Vigna Taglianti (1989) divided the genus *Ingolfiella* Hansen, 1903, into seven subgenera. Three of them (*Ingolfiella* s. str.; *Hansenliella* Stock, 1981; *Tethydiella* Ruffo and Vigna Taglianti, 1989), containing 16 true marine species, all with developed "ocular lobes", are contrasting with three other sub-

Table 1. Characters of *Ingolfiella* (*Antilleella*) species (from *putealis* to *unguiculata*) and *Ingolfiella* (*Tethydiella*) species (from *xarifae* to *longipes*).

	putealis	*fontinalis*	*tabularis*	*margaritae*	*similis*	*unguiculata*	*beatricis*	*xarifae*	*kapuri*	*grandispina*	*quadridentata*	*canariensis*	*fuscina*	*longipes*
ocular lobes	vestigial	reduced	reduced	?	?absent	vestigial	developed	developed	developed	developed	developed	developed	developed	developed
Gn1 dactylus inner teeth	3	3	3	3	3	3	4	3	4	3	4	4	4	4
Gn2 dactylus inner teeth	3	3	3	3	3	3	4	4	4	3	4	4	4	4
Gn2 palmar margin	serrated	serrated	serrated	serrated	serrated	serrated	smooth	smooth	serrated	smooth	serrated	serrated	feebly serrated	serrated
Gn2 c/p index (female)	1.8–1.9	2.1–2.4	2.1	unknown	2.3	2.2	1.9	2	1.9	2	2	2	2	1.9
P3, 4 dactylus disto-internal spur	present	present	present	present	present	present	present	present	absent	present	absent	absent	absent	absent
P5–7 dactylus disto-internal spur	present	present	present	present	present	present	present	absent	absent	absent	present only in P7	absent	absent	absent
P3, 4 dactylus claw	thin, bifid	thin, bifid, or simple	thin, bifid, or simple	thin, bifid	thin, bifid	thin, bifid	thin, bifid	thin, trifid	thin, bifid	thin, simple	thin, simple	thin, trifid	thin, trifid	thin, bifid
P5–7 dactylus claw	thin, bifid	thin, bifid, or simple	thin, bifid, or simple	thin, bifid	thin, bifid	thin, bifid	thin, bifid	robust, bifid	robust, bifid	robust, bifid	robust, bifid	robust, bifid	stout, bifid	robust, bifid
U1 exop./endop. ratio	0.5/1	0.5/1	0.6/1	0.5/1	0.5/1	0.35/1	0.25/1	0.5/1	0.6/1	0.8/1	0.5/1	0.5/1	0.55/1	0.6/1
Distribution	Bonaire	Bonaire	Curaçao	Margarita Isl. (Venezuela)	Canary Isl.	Madeira Isl.	Slovenia	Maledive	Andaman Isl.	Curaçao	Curaçao	Canary Isl.	W Atlantic coast off South Carolina, Gulf of Mexico	Bermuda Isl.
Ecology	anchialine	?fresh water	anchialine	fresh water	?fresh water	anchialine	fresh water	marine	marine psammal	anchialine	submarine sands	marine psammal	marine (17–151-m depth)	marine

genera (*Gevgeliella* S. Karaman, 1959; *Balcanella* S. Karaman, 1959; *Tyrrhenidiella* Ruffo and Vigna Taglianti, 1989) containing 9 freshwater subterranean species, without "ocular lobes." The subgenus *Antilleella* Ruffo and Vigna Taglianti, 1989, has an intermediate position between the two groups described above and is composed by 6 anchihalyne species which live in fresh- or brackish water, but always in close vicinity to marine coasts. These species have their "ocular lobes" more or less reduced, sometimes vestigial or lacking.

Ingolfiella beatricis is similar to all species of the subgenus *Antilleella* (see Table 1), especially in the pereiopods 3–7 dactyli, which are all equal, with one distointerior spur and a thin bifid claw, furthermore for the presence of pleopods 1–3 (but because the male is unknown, it is impossible to say if the pleopod 1 male has 2 distal setae, like all the species of *Antilleella*). From all these species *I. beatricis* is clearly different by: 1) presence of developed "ocular lobes" (reduced or probably quite absent in the *Antilleella* species; 2) gnathopods 1 and 2 dactyli with 4 inner teeth (always 3 in the *Antilleella* species); gnathopod 2 palmar margin smooth (always serrated in the *Antilleella* species); 3) outer ramus of uropod 1 very short, ¼ length of inner ramus (½ in *Antilleella,* except *I. unguiculata* ⅓ of inner ramus); 4) gnathopod 2 female c/p index (for definition see Stock, 1977) = 1.9, while it is more than 2 in species of *Antilleella* except *I. putealis* with c/p = 1.8–1.9.

Ingolfiella beatricis has some characters that are similar to species of the subgenus *Tethydiella* (see Table 1): 1) "ocular lobes" developed as in all *Tethydiella* species; 2) gnathopods 1 and 2 dactyli with 4 inner teeth (as in *Tethydiella* species except *I. grandispina* with 3 teeth and *I. xarifae* with 3 teeth in gnathopod 1 and 4 teeth in gnathopod 2); 3) c/p index = 1.9, as in all *Tethydiella* species (1.9–2). *Ingolfiella beatricis,* however, is different from *Tethydiella* by the similar dactyli of pereiopods 3–7 (like *Antilleella*), with a distointernal spur (absent in *Tethydiella,* except *I. xarifae* and *I. grandispina* with distointernal spur only in pereiopods 3 and 4, and *I. quadridentata* with distointernal spur present only in pereiopod 7).

We did not include in this discussion the mouthparts, as they are not always, and then often insufficiently, described for the difficulty of their preparation. But what is known does not indicate significant differences. For *I. beatricis* we want to underline the structure of mandibular molar, which is elongate, pointed, and bifid (as in *I.* (*Antilleella*) *similis* Rondé-Broekhuizen and Stock, 1987), the spination of maxilla 1 inner plate with 5 spines (as in *I.* (*Antilleella*) *unguiculata* Stock, 1992, the inner strongly curved as in some species of *Tethydiella*). Also we did not take into consideration the characters of sexual dimorphism, especially for gnathopod 2, pleopods 1–3, uropod 2, because *I. beatricis* is represented by only a single female specimen.

In conclusion it appears that *Ingolfiella beatricis* has an intermediate position between the true marine species (especially *Tethydiella*) and those anchihalyne ones, which are inhabiting fresh- interstitial or phreatic waters, in close vicinity to marine coasts, especially the subgenus *Antilleella,* to which our species has its closest affinities. The discovery of this species in subterranean freshwater, some 50 km inland from the Adriatic coast seems therefore quite significant for the evolution and biogeography of the genus *Ingolfiella.*

The subdivision in subgenera (Stock, 1976; Ruffo and Vigna Taglianti, 1989) which has been already discussed (Dojiri and Sieg, 1987; Lowry and Poore, 1989) should be revised with more complete character sets, as soon as one will have further material of the known species and hopefully also additional new discoveries in the future.

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