



Cernosvitoviella cryophila sp. n. (Oligochaeta, Enchytraeidae) from Alpine streams

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Abstract: A new species, *Cernosvitoviella cryophila* sp. n. (Enchytraeidae) was described from materials collected in the late 1990s in krenal and kryal streams in the Italian Alps (Adamello Brenta Natural Park, Trentino). The most distinctive morphological features of *C. cryophila* sp. n. are the unique shapes of the sperm funnel (cylindrical, about ten times longer than wide) and the sperm duct (long, with an expanded proximal third and abrupt width change between both duct sections). Seminal vesicles are absent and the spermatheca is very simple, with an oval ampulla and an ectal duct of nearly the same length. In the samples collected in the late 1990s, *C. cryophila* sp. n. co-occurred with seven other species of *Cernosvitoviella* but it has not been found again in materials collected at the beginning of the 21st century. The cause of its apparent disappearance remains unclear and needs further examination.

Key words: Carè Alto Glacier, Italian Alps, glacier-fed stream, new species, spring-fed stream, taxonomy

Introduction

Over the past 30 years, numerous papers concerning the distribution, ecology and monitoring value of headwater invertebrates have been published (e.g. Lods-Crozet et al. 2001, Maiolini & Lencioni 2001, Maiolini et al. 2006, Oertli et al. 2008, Lencioni & Spitale 2015, Lencioni 2018, Boggero et al. 2019, Becquet et al. 2022, Boggero et al. 2023, Cesarini et al. 2025), including their responses to global warming and glacier retreat (e.g. Debiasi et al. 2022, Lencioni et al. 2022, Wilkes et al. 2023). Several papers have focused specifically on oligochaetes (Anneli-

da, Oligochaeta) that colonise all types of mountain freshwater habitats: from springs (Sambugar 2007, Stoch et al. 2011) to the extreme upper reaches of glacier-fed and non-glacial streams, both in the benthic and hyporheic zones (Malard et al. 2001, Lencioni et al. 2004, Dumnicka et al. 2023) and in alpine lakes (Dumnicka & Boggero 2007, Dumnicka et al. 2015).

In particular, the genus *Cernosvitoviella* is well represented in Alpine waters (Dumnicka & Boggero 2017). According to gbif.org (2025), 23 species are currently assigned to this genus (excluding synonyms proposed by Schmelz & Collado 2010).

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However, the status of two synonymised species (*C. microtheca* Rota and Healy, 1999 and *C. goodhui* Healy, 1975) requires clarification and in this paper, as well as in Dumnicka & Boggero (2007) and Timm (2009), they are still treated as valid species. The majority of species is known from Europe, but this genus is cosmopolitan (gbif. org. 2025).

Sixteen species of *Cernosvitoviella*, including two previously described from the Rhaetian Alps, Italy (Dumnicka 2004, 2010), have been reported from Alpine freshwater habitats (Dumnicka & Boggero 2017). This high diversity may partly reflect the intensive research effort in the region associated with various scientific programs. Moreover, the genus is known to prefer cold waters. In addition to Alpine and other high mountain habitats, some species occur in other cold environments such as Icelandic springs (Klinth et al. 2019) and streams in North-Eastern Europe (Baturina 2012). Species of *Cernosvitoviella* also frequently inhabit the hyporheic zone, caves (Dumnicka et al. 2020, Creuzé des Châtelliers et al. 2021) and springs (Stoch et al. 2011), underscoring their thermophobic tendencies. Moreover some species occur in bogs, moorlands (Healy 1987, Dózsa-Farkas et al. 2017) and even wet soils.

This paper describes a new species of *Cernosvitoviella* collected in the 1990s from several glacial catchments of the Carè Alto-Conca system (Adamello-Brenta Natural Park, Adamello Mt.), Trentino Province (NE Italy).

Materials and Methods

The study was conducted in the north-east part of Italy in the Trentino Province (46°N, 10°E: Fig. 1). Sampling focused on the glacier-fed stream originating from the Conca Glacier, on the Mt. Carè Alto (Conca Stream) and its main non-glacial tributary. Specifically, nine 15-m sites were surveyed during the 1996-1997 growing seasons: sites C0, C1, C2 and C3 on the glacial reach (kryal); C6 and C7 on its main non-glacial tributary (krenal) and C4, C5 and C8 downstream of their confluence (mixed reach=glacio-rhithral). All sites were sampled during five-day periods on six occasions (late June, early August and mid-September of both 1996 and 1997), except for C1 and C5, which were sampled only in 1996. At each site and date, ten 0.1 m² areas (samples Q1-Q10) were sampled using a 0.30 x 0.30 m pond net with a 250- μ m mesh. Almost 500 samples were collected during the two years. Detailed descriptions of the study area and methods were published by Maiolini & Lencioni (2001). All invertebrates, including oligochaetes, were preserved in 75% ethanol until



Fig. 1. Study area and pictures of three of the localities where *Cernosvitoviella cryophila* sp. n. was detected. Site C3 – type locality; sites C2 and C3 – on the glacier-fed stream; site C7 – on the non-glacial tributary.

microscope identification. The same protocol was repeated at the same sites (C0 – C8) 25 years later (in 2022). All oligochaetes collected in both campaigns were examined. Permanent whole-mount slides were prepared in Canada balsam and identified following Timm (2009), Schmelz & Collado (2010) and original papers describing or comparing species of *Cernosvitoviella* (Healy 1975, Rota & Healy 1999, Dózsa-Farkas et al. 2017, Dózsa-Farkas 2019). Light micrographs were taken with a Nikon Eclipse 80i microscope equipped with a Nikon DS-Fi 1 camera.

Results

Characteristics of the studied streams

The specimens of *Cernosvitoviella cryophila* sp. n. were found at sites from C2 to C8 and characteristics of these sites are given in Table 1. Almost all of the studied sites (except for C8) were located above 2000 m a.s.l. but the average water temperature during the growing seasons varied considerably among sites (Table 1). The lowest pH values were found at the highest sampling site (5.7), after which the value increased slowly. We recorded very low conductivity values (maximum 11.3 μ S.cm⁻¹ in the krenal sector) and varying concentrations of suspended sediments (Table 1). The bottom substrata of the streams consisted mainly of stones of various sizes with some gravel and sand, while silt was found only sporadically (Table 1).

Table 1. Coordinates and main physico-chemical features of the sampling sites (mean \pm SD) where *Cernosvitoviella cryophila* sp. n. was recorded in 1996-1997.

Sampling sites	C2	C3	C4	C5	C6	C7	C8
Stream type	kryal	kryal	mixed	mixed	krenal	krenal	mixed
Latitude N (DD)	46.10135	46.09877	46.10062	46.10372	46.08333	46.09813	46.10357
Longitude E (DD)	10.61940	10.62222	10.62848	10.63389	10.62050	10.62472	10.66107
Altitude m a.s.l.	2507	2252	2115	2100	2285	2170	1300
Mean temp. (°C)	7.5 \pm 2.5	11.0 \pm 3.6	11.1 \pm 2.5	6.9 \pm 2.6	6.3 \pm 2.5	4.9 \pm 1.3	11.1 \pm 2.7
Discharge (m ³ .sec ⁻¹)	0.017 \pm 0.01	0.038 \pm 0.025	0.29 \pm 0.195	0.2 \pm 0.1	0.028 \pm 0.02	0.008 \pm 0.006	1.09 \pm 0.69
pH	5.7 \pm 0.5	5.9 \pm 0.5	6.1 \pm 0.4	6.4 \pm 0.1	6.0 \pm 0.4	6.1 \pm 0.4	6.0 \pm 0.5
Conductivity (μ S.cm ⁻¹)	6.8 \pm 2.7	7.9 \pm 3.2	10.0 \pm 1.0	10.2 \pm 1.2	10.7 \pm 0.9	11.3 \pm 0.8	10.3 \pm 2.1
Suspended sediments (mg.dm ⁻³)	3.7 \pm 1.7	9.2 \pm 9.5	6.2 \pm 10.8	2.7 \pm 1.8	1.6 \pm 1.3	3.0 \pm 2.6	4.2 \pm 2.4
Sediment type:							
boulder (%)	52	47	23	45	51	53	49
cobbles (%)	24	23	34	24	8	24	27
gravel (%)	14	17	19	14	34	12	14
sand (%)	10	12	24	17	7	11	10
silt (%)	-	-	1	-	-	-	-

Legend: DD – decimal degree

Substrate types: boulders >20 cm; cobbles 5 - 20 cm; gravel 0.2 – 5 cm; sand 0.01 – 0.2 cm; silt <0.01 cm

Description of *Cernosvitoviella cryophila* sp. n. Dumnicka

Holotype: A whole mature specimen mounted in Canada balsam, kept in the collection of the Institute of Nature Conservation, Polish Academy of Sciences (PAS), Kraków, Poland.

Type locality: Conca Stream (site C3, sample Q3), Adamello Mountain Group, Trentino, Italy (46.09877 N, 10.62222 E), 2252 m a.s.l., collected on 24 September 1996, leg. Lencioni V. and Maiolini B.

Paratypes: Five mature specimens from the same locality and date as the holotype. Three are stored in the Institute of Nature Conservation, PAS, Kraków, Poland. Two specimens (MUSE-INV-c001 000422 e MUSE-INV-c001 000423) have been deposited in the Natural Science Museum of Trento (now Museum of Science – MUSE), Italy.

Additional specimens: Mature specimens from a tributary of the Conca Stream (site C7, sample Q4), 2170 m a.s.l. (46.09813 N, 10.62472 E), collected on 1 August 1996 by Lencioni V. and Maiolini B., housed in the MUSE, Trento. Numerous other specimens were found in the studied streams. The new species was more abundant in samples collected in September 1996.

Etymology: The name refers to the species' occurrence in cold headwaters.

Description: length 1.8–3.3 mm, number of segments 23–28. Chaetae sigmoid with nodulus, in anteclytellar segments 27–38 μ m long, in postclytellar ones slightly shorter (24 – 34 μ m). Chaetal formula as follow: 5-7 (8) – (4) 5-6 : 6-8 – 5-7. No chaetae in segment XII. Saddle-shaped clitellum on segments XII-1/2XIII, weakly developed with small glandular cells, irregularly distributed dorsally and laterally. Coelomocytes not numerous, round (13–18 μ m diameter) and oval (15–25 μ m length), usually containing dark granules; sometimes granules in their peripheral part only. Some individuals exhibit an accumulation of coelomocytes in caudal segments (Fig. 2A).

Prostomium without internal papillae (Fig. 2B), brain twice as long as wide, with deep posterior incision (Fig. 2B). Two pairs of pharyngeal glands present: small, rounded primary glands (not united dorsally) and slightly larger, elongated secondary glands (Fig. 2C). Dorsal vessel originating from segment XII or anterior region of XIII. Large chloragogen cells originating from segment V and forming dense layer from segment VI onward; partially filled with small granules or nearly empty.

Unpaired nephridia from segments 6/7 to 8/9, middle one sometimes absent. Anteseptal part comprising only a small funnel; postseptal part elon-

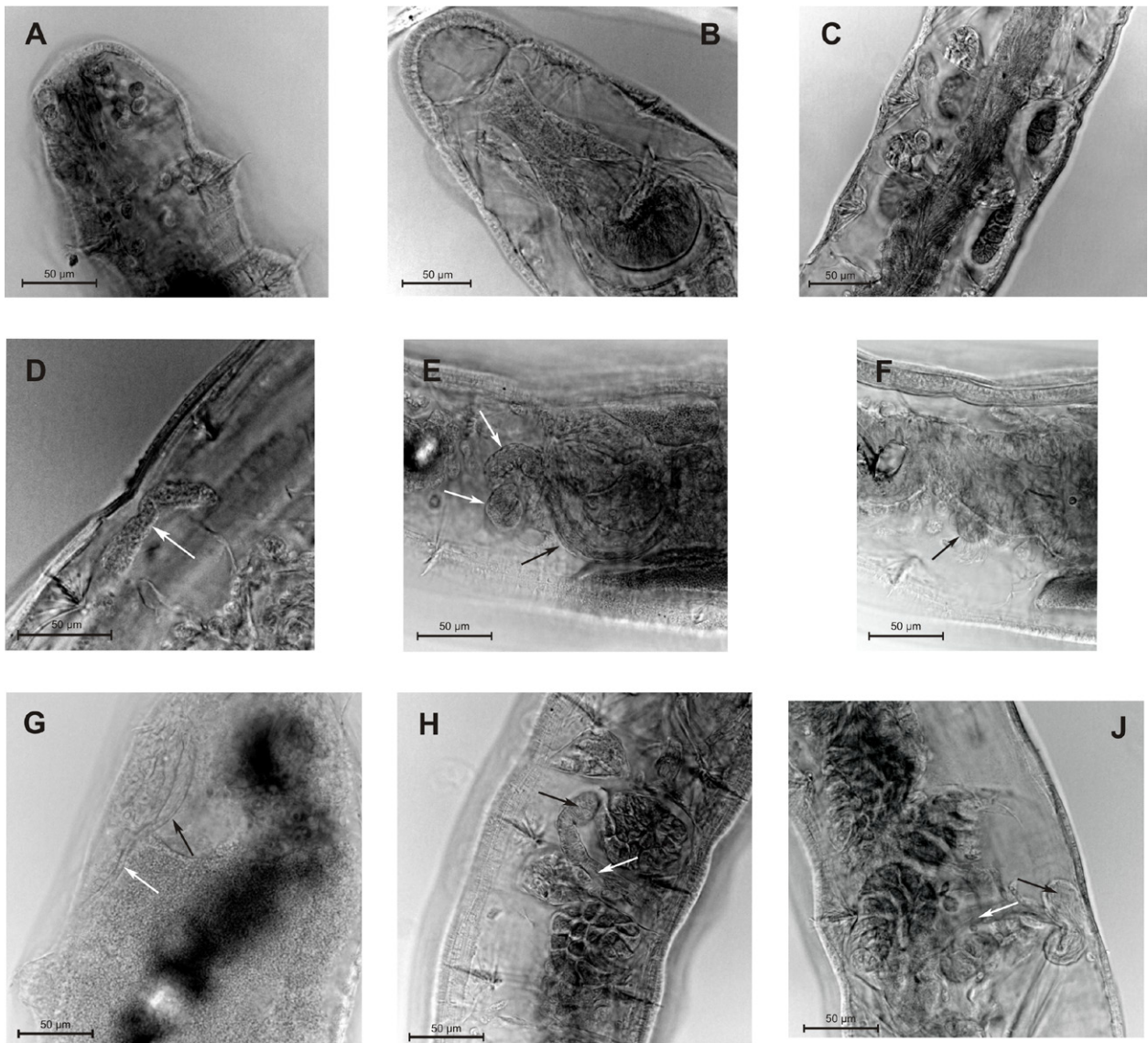


Fig. 2. Light micrographs of *Cernovitoviella cryophila* sp. n.

(A) Posterior body region with rounded coelomocytes (non-type material); (B) Anterior part with brain and pharynx (holotype); (C) Segments IV and V showing pharyngeal glands (paratype); (D) Nephridium (white arrow) (holotype). (E) Coiled sperm funnel (white arrows) and enlarged proximal part of sperm duct (black arrow) (holotype); (F) Collar outline (black arrow) and spermatozoa (non-type material); (G) Wide (black arrow) and narrow (white arrow) parts of sperm duct and penial bulb (paratype); (H) Spermatheca; ectal duct (white arrow), ampulla (black arrow) (holotype); (J) Coiled ampulla (black arrow) and ectal duct (white arrow) (non-type material).

gated (Fig. 2D, white arrow) with short terminal efferent duct.

Seminal vesicle absent. Sperm funnel long (90–140 μm), cylindrical, 9–10 times longer than wide (10–13 μm) (Fig. 3), usually twisted (Fig. 2E, white arrows), making precise measurements difficult. Collar distinct (Fig. 2F, black arrow), 15–20 μm high, slightly wider than funnel. Sperm duct long (250–300 μm), with proximal part widened. Widening starting just after funnel (Fig. 2E, black arrow), occupying about one-third of sperm duct length (Fig. 3). Wide section about twice as wide as narrow section; change

of width between sections abrupt (Figs. 2E, 2G, black arrows). So called “penial bulb” inconspicuous, formed by fused glands (Figs. 2G, 3).

Spermatheca simple (Figs. 2H, 2J), consisting of short ectal duct (50–60 μm) (Figs. 2H, 2J, white arrows) without glands at ectal orifice, with slightly longer ampulla (65–75 μm), diameter same or slightly larger than duct (Fig. 2H, black arrow). Ampullae sometimes coiled inside segment (Fig. 2J, black arrow), with loosely arranged sperm. Spermathecae usually in segment V, occasionally reaching segment VI.

Differential diagnosis

Cernosvitoviella cryophila sp. n. is distinguished primarily by the unique shapes of its sperm funnel and sperm duct. Although several species (e.g. *C. carpatica* Nielsen & Christensen, 1959, *C. crassoducta* Dózsa-Farkas, 1990, *C. minor* Dózsa-Farkas, 1990, *C. immota* (Knöllner, 1935), *C. aggtelekiensis* Dózsa-Farkas, 1970, *C. goodhui*, *C. parviseta* Gadzińska, 1974, *C. longiducta* Dumnicka, 2010) have cylindrical sperm funnels, in those species the funnels are no more than three times as long as wide (Schmelz & Collado 2010) (Table 2). *Cernosvitoviella palustris* Healy, 1979 is an exception, with sperm funnels 5–7 times as long as wide, but it differs from *C. cryophila* sp. n. in the shape and position of the spermathecae, which have a long ectal duct with an ectal widening and long ampullae reaching segment VII (Schmelz & Collado 2010) (Table 2).

Several species of *Cernosvitoviella* have sperm ducts with widened portions, but in most cases the widening is located ectally and comprises one third to one half of the duct length (e.g. *C. aggtelekiensis*, *goodhui*, *C. immota*, *C. microtheca* Rota & Healy, 1999, *C. omodeoi* Rota, 1995, *C. palustris*, *C. parviseta*) (Table 2). Additionally, short proximal widening occurs in *C. carpatica* and *C. bulboducta* Martínez-Ansemil & Collado, 1996. Besides the new species, only *C. crassoducta* has a widened proximal part of the sperm duct, but in that species this widening is longer and distinctly larger than the narrow part (Schmelz & Collado 2010). Furthermore, *C. crassoducta* differs by having a small, pear-shaped sperm funnel and a distinctly shorter whole sperm duct (Dózsa-Farkas et al. 2017; Table 2). The gradual widening of medium or proximal part of the sperm duct was also observed in some populations of *C. atrata* (Bretscher, 1903) (Schmelz & Collado 2010, Dózsa-Farkas 2019), but in *C. cryophila* sp. n. this widening finishes abruptly. Moreover *C. atrata* has funnel-shaped sperm funnel, only two times longer than wide (Table 2).

Discussion

The richness and distribution of oligochaete fauna in alpine freshwater habitats in several countries (Italy, Switzerland and France) is well understood, owing to numerous studies conducted mainly in the 21st century (summary in Dumnicka & Boggero 2017). Most species of the genus *Cernosvitoviella*, the only typically aquatic taxon among Enchytraeidae, are found in the Alps, but only *C. atrata* is widespread (Dumnicka & Boggero 2017). The remaining spe-

Table 2. Structure of the reproductive organs in species of *Cernosvitoviella* having cylindrical sperm funnels (SF) and/or sperm duct (SD) with widened section.

Species	Seminal vesicle	Sperm funnel			Sperm duct		Spermatheca		
		shape	length: width	SD : SF	wide section	in segments	shape of ampulla	ampulla: ectal duct	
<i>C. cryophila</i> sp. n.	-	cylindrical	9-10 : 1	2.5-3 : 1	proximal	V (VI)	cylindrical	1.5 : 1	
<i>C. aggtelekiensis</i>	-	cylindrical	2.5-3 : 1	3-3.5 : 1	ectal half	V - VI	oval	1 : 2.5	
<i>C. atrata</i>	-	funnel-sh.	1.5-2 : 1	~8 : 1	not or prox.	V	oval	1 : 1	
<i>C. bulboducta</i>	small	funnel-sh.	~2 : 1	~8 : 1	prox. bulb	VII - VIII	long, narrow	3.5-4 : 1	
<i>C. carpatica</i>	-	cylindrical	1.5-2 : 1	3-3.5 : 1	ectal, small	V	sphaerical	1 : 1.5-2	
<i>C. crassoducta</i>	-	funnel-sh.	1.5-2 : 1	2.5 - 3.5	proximal	V	oval	1 : 1.5-2	
<i>C. goodhui</i> (syn.)*	-	cylindrical	2-3 : 1	3-5 : 1	ectal 1/4	V - VI	cylindrical	1 : 1-1.5	
<i>C. immota</i>	large	cylindrical	2-3 : 1	~3 : 1	ectal 1/3	V	cylindrical	1 : 1.5	
<i>C. longiducta</i>	small	cylindrical	1-1.5 : 1	12-15 : 1	-	V	oval	1 : 1.5	
<i>C. microtheca</i> (syn.)*	-	funnel-sh.	2 : 1	7-8 : 1	ectal 1/3	V	sphaerical	1 : 1	
<i>C. minor</i>	-	cylindrical	~2.5 : 1	2.5-3 : 1	-	V	sphaerical	1 : 2.5-3	
<i>C. omodeoi</i>	-	barrel-sh.	~2 : 1	~4 : 1	ectal half	V (VII)	cylindrical	3-4 : 1	
<i>C. palustris</i>	-	cylindrical	5-7 : 1	1-1.5 : 1	ectal 1/3	VII	long, narrow	1 : 2	
<i>C. parviseta</i>	-	cylindrical	~3 : 1	2.5 - 3 : 1	ectal half	V - VI	cylindrical	1 : 2	

*According to Schmelz & Collado (2010), *C. goodhui* is synonymised with *C. aggtelekiensis* and *C. microtheca* – with *C. atrata* (more explanations in the text).

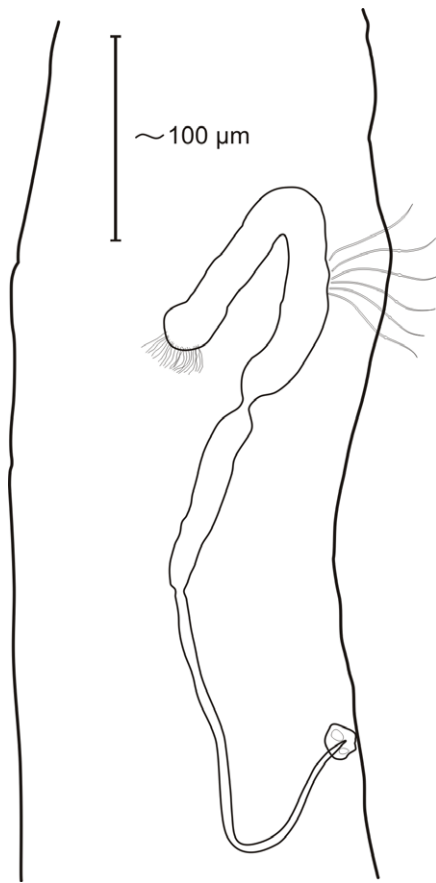


Fig. 3. Scheme of the male reproductive organ of *Cernovitiella cryophila* sp. n.

cies have been found in fewer of the studied regions: from four to one among the nine studied, which proves their patchy distribution.

Cernovitiella cryophila sp. n. was recorded exclusively in zoobenthos samples collected in the Conca Stream and its tributary in 1996–1997. The same system was resampled 25 years later, in 2022, but this species – abundant in the samples from 1996–1997 – was not found again (unpublished data). During this period, the glacier shrank from approximately 18 ha to roughly 9 ha and the Conca Stream lengthened by approximately 300 m (Smiraglia & Diolaiuti 2015, Tridentine Mountaineers' Society - pers. comm.). The mean of maximum daily water temperatures at site C0 increased from 3.1 °C in August 1996–1997 to 7.8 °C in August 2022 (unpublished data).

In the late 1990s, *C. cryophila* sp. n. co-occurred with seven other species of *Cernovitiella* in the Conca Stream (*C. ampullax* Klunghand & Abrahamsen, 1981, *C. atrata*, *C. goodhui*, *C. immota*, *C. palustris*, *C. tridentina* Dumnicka, 2004, *C. longiducta*). The latter two species were originally described based on 21st-century samples from other Alpine streams (Careser and Noce Bianco) (Dum-

nicka 2004, 2010) but were also found in recently analysed samples. All these species persisted in Alpine freshwater habitats at least into the early 21st century (Dumnicka & Boggero 2017), but *C. cryophila* sp. n. has not been found again. Some species of *Cernovitiella* seem to be restricted to the uppermost sectors of glacier-fed streams (Dumnicka et al. 2023) and they could be threatened with extinction due to the warming of alpine waters. Nevertheless, the causes of apparent disappearance of *C. cryophila* sp. n. remain unclear and further studies are needed in order to understand true species distribution and its ecological requirements.

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