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Redescription of the cryoconital tardigrade *Hypsibius klebelsbergi* Mihelčič, 1959, with notes on the microslide collection of the late Dr. F. Mihelčič (Tardigrada)

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Abstract: I redescribe and desginate a lectotype of *Hypsibius klebelsbergi* Mihelčič, 1959, a cryoconital tardigrade from an Austrian glacier. The species is similar to *H. janetscheki* Ramazzotti, 1968 from the Himalayas. The presence of active and dormant forms within these cryoconite inhabitants is discussed. The content and the state of preservation of the Mihelčič's tardigrade collection is described.

Keywords: *Hypsibius klebelsbergi,* redescription, cryoconites, Tardigrada.

Introduction

Only a few types of organisms survive and dwell in water-filled small holes that form on surface of glaciers. These peculiar habitats come into being by the melting down of fine dark inorganic and organic debris (dust) into the ice surface under the impact of solar radiation. This wind transported dust is known as cryoconite (e. g. Hamelin & Cook 1967). The cryoconite holes are colonized by some algae, protozoans, rotifers and tardigrades (Steinböck 1957, Mihelčič 1963). Two species of Tardigrada were reported exclusively from this habitat (Mihelčič 1959, Ramazzotti 1968) and, notably, they possess dark pigmentation. One of these species, *Hypsibius klebelsbergi* Mihelčič, 1959 has been described from an Austrian glacier, and the second, *Hypsibius janetscheki* Ramazzotti, 1968, from a Himalayan glacier. Both taxa are known only through the original description. Ramazzotti & Maucci (1983) do not exclude conspecificity between these tardigrades.

Recently I have had the opportunity to examine the remnants of the tardigrade collection of the late Dr. Mihelčič. One microslide of the collection comprised type-series of *Hypsibius klebelsbergi.* Its original description is incomplete and inaccurate and I redescribe this taxon.

Material and Methods

All tardigrades from the Mihelčič's collection are housed in the Tiroler Landesmuseum Ferdinandeum (Innsbruck, Austria) and they are mounted in »Cedax« medium (Dr. Schatz, in litt.). However, no slide was protected by any varnish or seal. Unfortunately, the collection was flooded in 1985 (Dipl.-Biol. Schütz, in litt.) and, as a result, water penetrated under cover-slip of many slides and washed out most of the mounting medium. As water evaporated, remnants of the medium with enclosed tardigrades condensed, darkened and partly cristalized, making majority of animals inaccessible to examination. Several slides lost their cover-slips.

The microslide with four type-specimens of *H. klebelsbergi* was originally labelled: »Ötztaler Alpen/leg. Steinböck« and »*H. klebelsbergi sp. n.*« (in pencil). Today, the animals are damaged,

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their buccal apparatus are deformed and almost indiscernible. To recover the specimens, the slide with its almost separated cover-slip was placed for 12 hours in a Petri dish with distilled water. After the mounting medium had partly dissolved, the cover-slip was removed and the tardigrades were cautiously dissected with a thin needle from the remnants of the mounting substance. Subsequently, the specimens were transferred by a micropippete onto four microslides and mounted in chloral gum (Swan's medium). The observations were carried out using phase- and interference contrast microscopy. Mihelčič (1959) has designated no holotype, thus the specimens had a status of syntypes. A lectotype and paralectotypes are designated in this paper. Unless mentioned otherwise, all measurements refer to the lectotype. The measurements of paralectotypes are given in parentheses.
For comparison , I examined the type-series of another cryoconital tardigrade, *H. janetscheki.* The label reads: »Tipo 178/10 *H. (H.) janetscheki*/Ramazzotti/(Poliv.)« and »Pozzetto/glaciale m 5600/Himalaya, Ghiac/ciao Amar Dablan«. Furthermore, I examined comparative material of active and dormant forms of *Diphascon higginsi* Binda, 1971 with different types of claws. The latter species originated from moss samples collected by Mrs. Ch. Käser in Hamburg.

Redescription

Hypsibius klebelsbergi Mihelčič, 1959
(Figs. 1—9)

Hypsibius (H.) klebelsbergi Mihelčič (Zool. Anz., 1959, 163: 259—261, Fig. 4a—c); *Hypsibius (I.) klebelsbergi:* Mihelčič (1963); *H. klebelsbergi:* Ramazzotti (1962, 1972), Ramazzotti & Maucci (1983).

Material: 4 syntypes of *H. klebelsbergi* (sex undeterminable): lectotype and paralectotypes are designated here. The lectotype and 2 paralectotypes are deposited in the Landesmuseum Ferdinandeum (Innsbruck, Austria), one paralectotype is housed in the Zoologisches Museum, Universität Hamburg (Reg. No. A32/92). Other material: type-series of *H. janetscheki* (10 syntypes, one slide) and 2 additional specimens (Museo Civico di Storia Naturale: Verona, Italy). Four slides with numerous specimens of *D. higginsi* (Zoologisches Museum, Hamburg).

Description of type material: Body 640 (313—536) µm long, 238 (95—218) µm wide and relatively dumpy. Head segment wide. Cuticle smooth. Epidermal cells of all investigated specimens filled with dark-brown pigment giving the animal a unique brown-blackish and non-transparent appearance. Several lesser pigmented transvere and longitudinal stripes occur on dorsum, resembling to some degree the pigmental pattern of the genus *Ramazzottius* Pilato. Eye-dots located »anteriorly«, large, their diameter about 19 µm (Fig. 1). The eyes composed of numerous dark-brown granules, thus difficult to discern against dark pigmented body.
Mouth opening terminal (Fig. 1). The buccal apparatus is mutilated and thus hardly discernible, but was examined in 3 out of 4 specimens. Mouth tube (Fig. 7) 37 µm long, its outer diameter 5 µm. Stylet supports and stylets could not be seen. Poorly defined apophyses and macroplacoids could be seen only in one paralectotype. The apophyses are tiny and about 1.6 µm in diameter. Macroplacoids irregularly shaped (Fig. 8), sharply tipped and the first pair

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distinctly incised in their middle. The macroplacoids are 8—11 µm long. Second pair of macroplacoids with poorer defined lateral incisions, 8—9 µm long. No microplacoids. Strongly squashed pharynx in the above paralectotype measures 63 x 53 µm.

Legs relatively small, particularly their IVth pair (Figs. 1, 9). Hind legs without dorsal hump, although originally described by Mihelčič (1959). Claws small, of *Hypsibius*-type and stumpy (Figs. 2—6). No cuticular bars between the claws nor their bases. Lunules are lacking. Main branches of claws, particularly those of the external ones wide at base and provided with large and flattened accessory spines. These relatively thick and of same size as the apical part of the main branch. The spines are located on lateral sides of main branch (Figs. 2—6). Consequently, the distal parts of claws when seen in the lateral view resemble a wide and flattened structures (Figs. 3, 4). External claw I 12 µm long, with its main branches 10 µm in length and 3.3 µm width. Internal claw IV 14 µm, its main branch 6 µm long. Main branch of the claw 3.5 µm wide. No eggs and exuviae are present.

Remarks: The original description of *H. klebelsbergi* by Mihelčič (1959) disagree in some details with the type-series it was based on. The specimens are relatively plump and their length-for width ratio is 2.7—3.3. Mihelčič (op. cit.) described the animals as »very slim« and reported the ratio as »almost 5:1«. From the quantitative data provided by him that ratio is 4.6 but he gives no information if the measurements were taken from living or mounted animals. The eye dots, though obscured by the dark body pigmentation, are present in all type-specimens, but not reported in the original description. Furthermore, characteristically shaped accessory spines of the main branches on all claws found in type-series »were not seen« by Mihelčič (op. cit.). According to the author, legs in *H. klebelsbergi* are »long and on their IVth pair there is a large hump«. The specimens, however, have the legs, particularly their hind pair, rather small and there is no hump/tubercle on their IVth pair. Ramazzotti & Maucci (1983) reported close resemblance between *H. klebelsbergi* and *H. janetscheki* and they have not excluded the conspecifity between the taxa. For that would also indicate the presence of eyes and the lack of the hump on hind legs in *H. klebelsbergi,* recorded in this study.

In the examined type-series of *H. janetscheki* (10 syntypes) one can distinguish two morphotypes which differ by the degree of the body pigmentation and the shape and size of legs and claws. One group (3 specimens) has the body brown-blackish pigmented, short hind legs and relatively small stumpy claws (Fig. 11). I failed to find any significant differences between these specimens and type-series of *H. klebelsbergi,* with the exception of their slightly longer claw main branches. The remaining morphotype (7 specimens) is characterized by light-brown epidermal pigment, longer hind legs and distinctly longer claw main branches (Fig. 10), compared to those in the dark pigmented forms. That morphological differentiation has been overlooked in the original description of *H. janetscheki.* These morphological differences suggest the presence of two life forms within *H. janetscheki* (and *H. klebelsbergi*?). Dark pigmented specimens with short claws and small hind legs may belong to the dormant/hibernating (?) form. Light pigmented specimens with larger claws and longer hind legs may represent the active form of the species. If this hypothesis is correct, then only one of these forms, the dark pigmented one, has been hitherto described with *H. klebelsbergi.* The question of possible synonymy between the two taxa remains open and can be solved only by the examination of a new material of both species.

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Recently Kristensen (1982) described the first case of cyclomorphosis within tardigrades, based on the marine *Halobiotus crispae* Kristensen, 1982. He recognized, among the other things, distinctly different shape and structures of claws in winter and summer forms. Until his paper, the claw shape and structures were viewed as constant for a particular tardigrade species (Pilato 1975). Consequently, Kristensen (op. cit.) has suggested a possibility of cyclomorphosis for terrestrial eutardigrades, particularly for the genus *Diphascon*. That suggestion appears to be correct for *Diphascon higginsi* Binda, 1971. Presently I have observed two distinctly separated morphotypes within a large population of the species. Both forms differ, apart from buccal apparatus, pigmentation and some other characters, also in the shape and type of claws, i. e. *Hypsibius*-type versus *Calohypsibius/Isohypsibius*-type (comp. Fig. 12 and 13). Detailed description of these specimens is beyond the scope of this paper. However, the morphological differences found in *Diphascon* suggest the presence of similar life history traits in the related genus *Hypsibius* to which both cryoconital species belong. To answer fully the question, more data on almost unknown biology and ecology of these cryoconital species are needed, including information on taxonomically important but insufficiently known buccal apparatus. The latter is damaged and deformed in the type material of *H. klebelsbergi* and poorly visible in that of *H. janetscheki*.

Both species seem to be specifically adapted to the life in cryoconite holes thus they may represent an relic element of cold climate fauna of the Pleistocene. Judging from the morphology of their claws and from what is known of their buccal apparatus, the most like ancestors of *H. klebelsbergi* and *H. janetscheki* are relatives of the forms which gave rise to the recent »*Hypsibius convergens-dujardini*«-complex.

Notes on the microslide collection of the late Dr. Mihelčič

The Mihelčič's tardigrade collection comprises only 35 microslides. Another slide in this collection presumably also contained a type-series of another species, as judging from its label: »*H. (I.) belliformis* n. sp.«. Unfortunately the slide has no cover-slip and I found no tardigrade in the remnants of degenerated mounting medium.

Most slides are seriously damaged (see »Material and Methods«) and, according to their pencilled labels, they should contain the following taxa: *Echiniscus testudo* (Doyère, 1840), *E. trisetosus* Cuènot, 1932, *E. canadensis* Murray, 1910, *Macrobiotus hufelandi* Schultze, 1834, *M. harmsworthi* Murray, 1911, *Dactylobiotus dispar* (Murray, 1907), *Hypsibius pallidus* Thulin, 1911, *H. dujardini* (Doyère, 1840), *Isohypsibius prosostomus* (Thulin, 1928), *I. nodosus* (Murray, 1907), *I. granulifer* Thulin, 1928, *I. schaudinni* (Richters, 1909), *I. hadzi* (Mihelčič, 1938), *Pseudobiotus augusti* (Murray, 1907), *Diphascon scoticum* (Murray, 1905) and *D. arduifrons* (Thulin, 1928). The material originates from Austria, Germany, Norway and Sweden. Unfortunately, most slides are empty and some of species are incorrectly identified. The taxonomically interesting or dubious taxa as *I. nodosus, I. hadzi, I. schaudinni* and *D. arduifrons* are lost as well.

Between 1938 and 1972 Mihelčič described 57 tardigrade species, mainly from Europe, and from Argentina and the United States (Ramazzotti & Maucci 1983, Kofler 1978). These taxa comprises about 7% of all known tardigrades. However, most of these species were never confirmed, and

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if they were, it was by Mihelčič himself. I attribute a low re-discovery rate to the unsufficient and often incorrect original descriptions provided by the above author. As a result, the descriptions have considerably contributed to the taxonomical chaos which prevails at the species level in Tardigrada. The »tuberculatus«-complex of the genus Isohypsibius, where Mihelčič described numerous new taxa can serve as only one example.

Of these 57 taxa only type-series of one species, *H. klebelsbergi,* is present in the collection. Type-series of two *Pseudechiniscus* species described jointly with other authors (Grigarick et al. 1964) are deposited in the collection of Schuster (Davis, California). Thus, one should assume that the type-material of the remaining 54 species has been lost. Consequently, as the original descriptions and illustrations of the species are too inadequate for reliable identification, Mihelčič's species should be recognized as *nomina dubia.* All species described by Mihelčič are included in the recent tardigrade monograph by Ramazzotti & Maucci (1983).

Acknowledgements

I would like to thank Dr. H. Schatz (Institut für Zoologie, Universität Innsbruck) and Dr. G. Tarmann (das Tiroler Landesmuseum Ferdinandeum, Innsbruck) for access to the Mihelčič's tardigrade collection and generous donation fo one paralectotype to the Zoologisches Museum, Universität Hamburg. I am grateful to Prof. Dr. W. Maucci (Verona) for his kind assistance in obtaining access to the type-specimens of *H. janetscheki.* I thank Mrs. Ch. Käser (Hamburg) for samples with *D. higginsi,* Dip.-Biol. G. Schütz (St. Pölten) for all data concerning the Mihelčič's collection and Dr. J. Kolasa (McMaster University, Hamilton) for corrections to the English manuscript.

Zusammenfassung

Es wird ein in den Kryokonitlöchern des österreichischen Gletschers vorkommendes Bärtierchen, *Hypsibius klebelsbergi* Mihelčič, 1959 auf Grund des Typenmaterials nachbeschrieben und ein Lectotypus designiert. Die Art wird mit dem ähnlichen *Hypsibius janetscheki* Ramazzotti, 1968, aus himalayschen Kryokonitlöchern, verglichen und das Vorkommen von aktiven und dormanten Formen bei diesen Bärtierchen wird diskutiert. Inhalt und Zustand der Tardigraden-Sammlung von Mihelčič wird beschrieben.

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Figs. 1—9: *Hypsibius klebelsbergi* Mihelčič: 1 — habitus, dorsolateral view, 2 — external claw I, 3 — internal claw I, 4 — distal unit of the claw main branch IV, 5 — external claw II, 6 — internal claw II, 7 — mouth tube, dorsal view, 8 — macroplacoids, 9 — hind legs, dorsal view (Figs. 1—3, 7 are drawn from lectotype).

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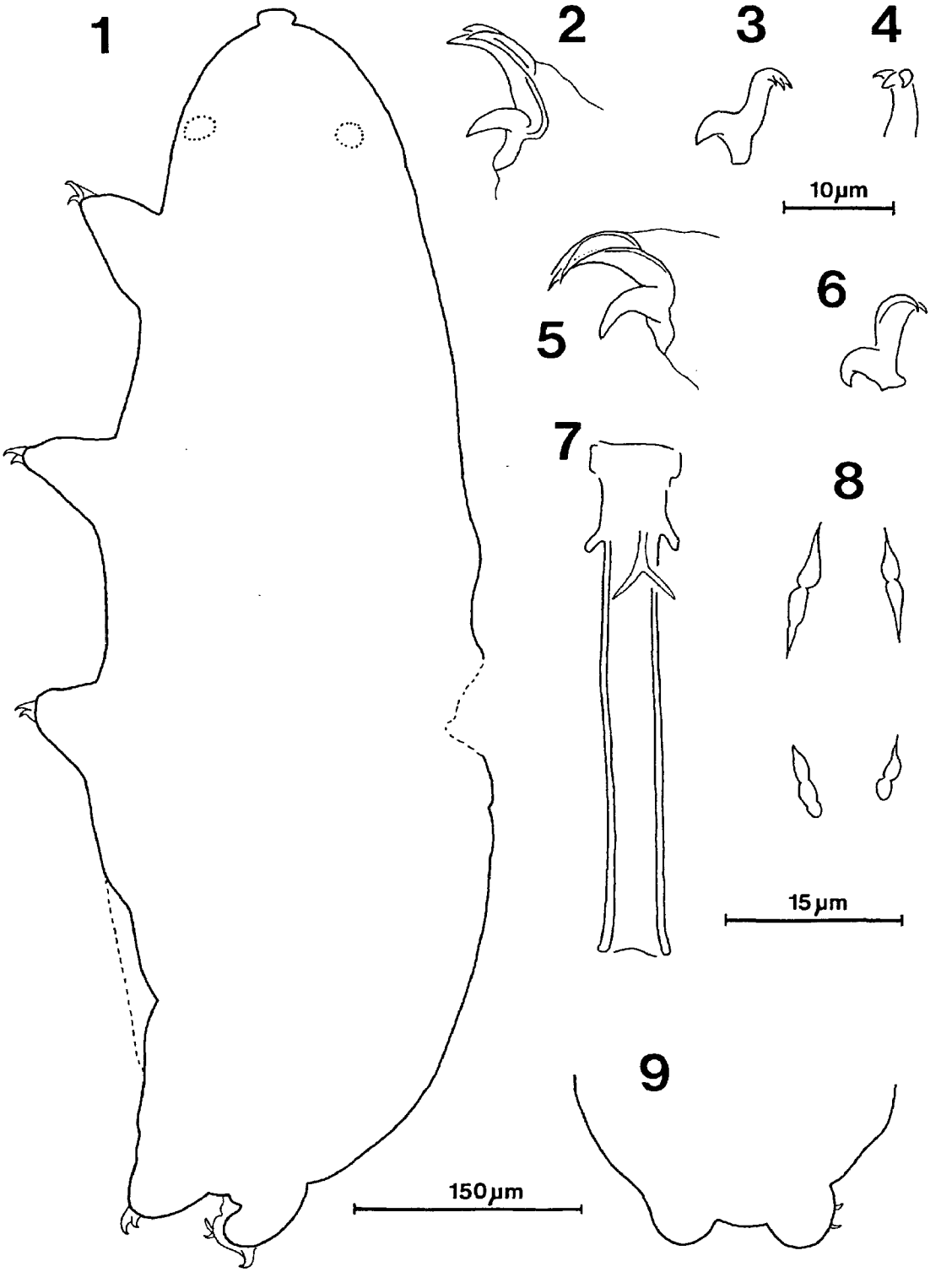

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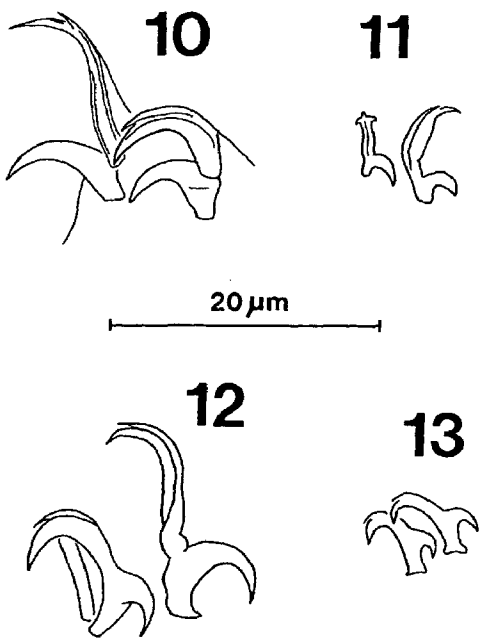

Figs. 10—13: *Hypsibius janetscheki* Ramazzotti: 10 — claws of the IInd pair of legs, active form, 11 — claws of the IInd pair of legs, dormant form; *Diphascon higginsi* Binda: 12 — claws of the IInd pair of legs, active form, 13 — claws of the IInd pair of legs, dormant form.

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Zeitschrift/Journal: Veröffentlichungen des Tiroler Landesmuseums Ferdinandeum

Jahr/Year: 1993

Band/Volume: 73

Autor(en)/Author(s): Dastych Hieronymus

Artikel/Article: Redescription of the cryoconital tardigrade Hypsibius klebelsbergi Mihelcic, 1959, with notes on the microslide collection of the late Dr. F. Mihelcic (Tradigrada). 5-12