

# Four Freshwater Eutardigrades from Korea with Description of *Isohypsibius brevitubulatus* n. sp.

Hyun Soo Rho, Seung Yeo Moon<sup>1</sup>, Cheon Young Chang\*, and Won Kim<sup>2</sup>

Department of Biology, College of Natural Sciences, Taegu University, Kyungpook 712-714, Korea:

<sup>1</sup>Station Biologique BP 74, 29682 Roscoff Cx, France:

<sup>2</sup>Department of Molecular Biology, Seoul National University, Seoul 151-742, Korea

Key Words:

Tardigrada

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As a result of examining the specimens collected from various freshwater bodies since 1995, four eutardigrades are added to Korean fauna: *Isohypsibius brevitubulatus* n. sp., *I. baldii* (Ramazzotti), *I. marcellinoi* (Binda and Pilato), and *Diphascion* (*Diphascion*) *higginsii* Binda. *I. brevitubulatus* n. sp. differs from its congeners by having a combination of the following characteristics: smooth cuticle, rather big body size, short and broad buccal tube, presence of lunules, and pharynx bearing 3 rod-like macroplacoids without microplacoid. All the species are illustrated and briefly commented on their habitats with the associated fauna.

Since Iharos (1971) first reported eight eutardigrade species from North Korea, Dastych (1974) examined 27 species found by "The Expedition of the Institute of Systematic and Experimental Zoology of the Polish Academy of Sciences in Kraków, 1971", and made a checklist on 33 North Korean species including 25 eutardigrade ones.

Thereafter, a series of investigations from South Korea (Moon and Kim, 1988; Moon et al., 1989; Moon and Kim, 1991; Moon et al., 1992) added ten eutardigrades to the Dastych's list. Recently Moon et al. (1994) and Chang and Rho (1996) newly described *Doryphoribius koreanus* and *Isohypsibius granditintinus*, respectively.

As a result of examining specimens deposited in Department of Biology, Taegu University, we report four eutardigrades including a new species. Therefore, a total of 41 eutardigrade species of 11 genera in 4 families are now known from the Korean Peninsula.

## Materials and Methods

The materials examined in this study were collected from submerged sand bottoms of various freshwater bodies during the period of March 1995 to April 1997. Sands including the specimens were dredged into polyethylene plastic bag, and then the specimens were extracted by the anaesthetization (using MgCl<sub>2</sub>)-decantation technique (Hulings and Gray, 1971) at the laboratory. The extracted specimens were fixed by 5% formalin.

Specimens were drawn and measured in lactophenol

on Cobb's aluminium hole slide, and also observed and photographed under differential interference microscope. Figures were made with the aid of a drawing tube. Specimens for SEM examination was fixed with hot (about 80°C) ethanol immediately after extraction, and fixed again for overnight at 4°C in a 2.5% glutaraldehyde, then followed by postfixation with 1% cold osmium tetroxide. After dehydration through a graded series of ethanol (50%, 60%, 70%, 80%, 90%, 100%, 100%) for 30 minutes each, the material was critical point dried, and coated with gold-palladium in a high evaporator, and then examined in a Hitachi S-520 scanning electron microscope operated at 20 KV.

## Systematic Accounts

Class Eutardigrada Marcus, 1927  
Order Parachela Schuster, Nelson, Grigarick and Christenberry, 1980  
Family Hysibiidae Pilato, 1969  
Subfamily Hysibiinae Pilato, 1969  
Genus *Isohypsibius* Thulin, 1928

*Isohypsibius brevitubulatus* Rho, Chang and Kim, n. sp.  
(Figs. 1, 2A-D)

Material examined: 20 individuals (including 2 exuviae), submerged sand bottom of Sömjin River under Kurye Bridge (35° 09' 43" N, 127° 27' 11" E), 20 May 1995, H. S. Rho and G. H. Rho. All are mounted in lactophenol. Holotype and three paratypes will be deposited in the Zoologisches Museum, Universität Hamburg, Germany. Other paratypes (16 individuals including 2 exuviae) are kept in the collection of the authors.

\* To whom correspondence should be addressed.

Tel: 82-53-850-6454, Fax: 82-53-850-5834

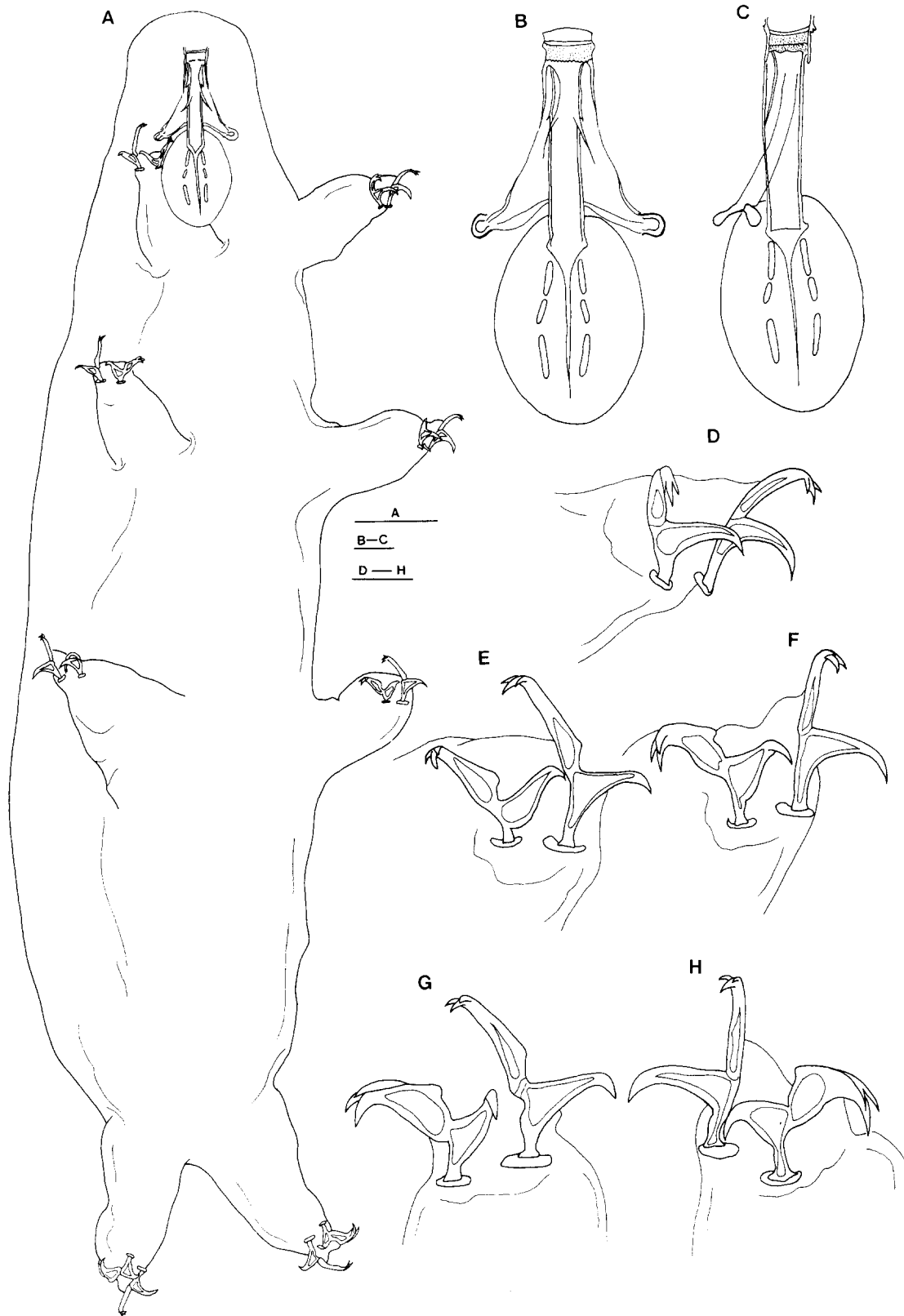


Fig. 1. *Isohypsibius brevitubulatus* n. sp. A, Habitus, ventro-lateral view. B, Bucco-pharyngeal apparatus, ventral view. C, Bucco-pharyngeal apparatus, lateral view. D, Claws on leg I. E, Claws on leg II. F, Claws on leg III. G-H, Claws on leg IV. Scale bars=0.05 mm (A) and 0.01 mm (B-H).

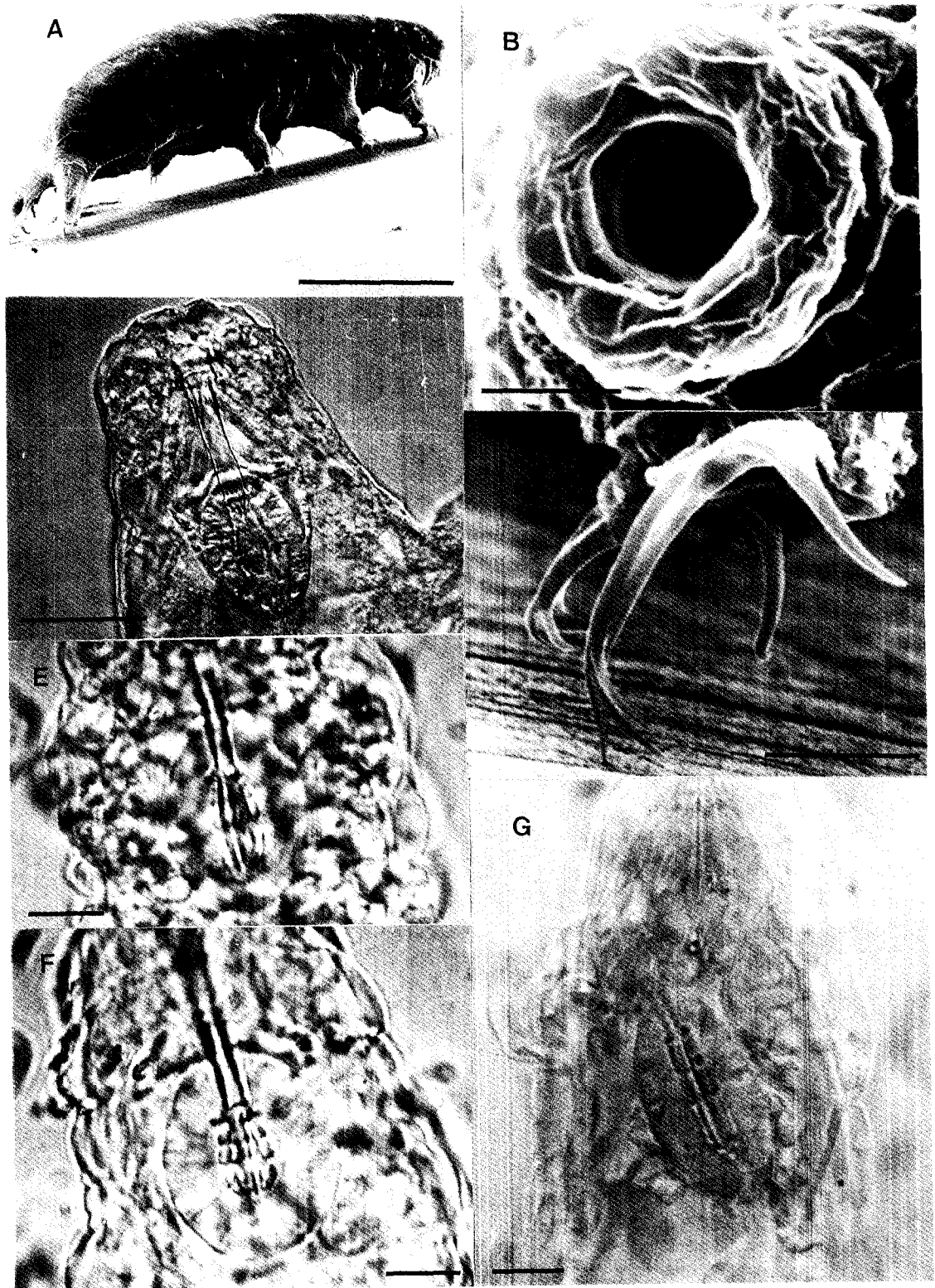


Fig. 2. *Isohypsibius brevitubulatus* n. sp. A, Habitus. B, Mouth opening with peribuccal area. C, Claws of the second pair of legs. D, Bucco-pharyngeal apparatus, ventral view. E-H, Bucco-pharyngeal apparatus, ventral view. E, *Isohypsibius baldii* (Ramazzotti). F, *Isohypsibius marcellinoi* (Binda and Pilato). G, *Diphascocon* (*D.*) *higginsii* Binda. Scale bars=0.01  $\mu$ m (E-H), 0.05  $\mu$ m (D), 7.5  $\mu$ m (B), 8.6  $\mu$ m (C), and 120  $\mu$ m (A).

Table 1. Character measurements ( $\mu\text{m}$ )

Character	Specimen									Mean	<i>I. myrops</i>
	*H	**P1	P2	P3	P4	P5	P6	P7			
Body length	600	500	501	496	471	893	790	961	652	560	
Macroplacoids I:II:III	0.8:0.4:1 (10.5:5.8:13.9)	0.7:0.5:1 (5.4:4.1:8.2)		0.8:0.6:1 (6.1:4.4:7.7)		0.7:0.3:1 (10.4:6:14.1)			0.75:0.45:1 (8:4.7:10.9)	0.75:0.6:1 (6:5:8)	
Length of buccal tube /body length	97%	84%	84%	89%	85%	73%	74%	71%	82%	155%	
Length /width ratio buccal tube	8.3 (58/7)	7 (42/6)	7 (42/6)	7.3 (44/6)	6.7 (40/6)	7.2 (65/9)	7.3 (58/8)	6.8 (68/10)	7.1 (52/7.3)	21.75 (87/4)	
Width of buccal tube /length of pharyngeal bulb	12%	15%	16%	15%	16%	14%	14%	15%	14.6%	8.69%	
Length/width ratio of pharyngeal bulb	1.5 (58/39)	1.4 (40/29)	1.4 (38/27)	1.4 (40/29)	1.5 (38/26)	1.4 (63/46)	1.4 (58/41)	1.3 (65/50)	1.41 (50/43.3)	1.31 (46/35)	

\*H: holotype, \*\*P: paratype.

Additional material examined: 3 individuals, collection site same as type locality, May 12, 1996, H. S. Rho and G. H. Rho; 5 individuals, collection site same as type locality, August 21, 1996, J.-L. Cho and H. S. Rho, of which 3 specimens are gold-coated on the aluminium stub for SEM preparation.

Diagnosis: cuticle smooth; body very big; buccal tube short and broad; pharynx with 3 rod-shaped macroplacoids without microplacoid; lunules present.

Holotype: Body length up to 600  $\mu\text{m}$ , and width 137  $\mu\text{m}$ . Body elongated and yellowish. Eye pigments absent. Cuticle completely smooth. Legs also smooth without any kind of humped projection. Mouth opening located in a front-ventral position, with peribuccal lobes and mucrons and without lamellae; buccal armature resembling other species of *Macrobotus* type; width of mouth opening 13.2  $\mu\text{m}$ . No apophyses in pharynx; internal diameter of buccal tube 7  $\mu\text{m}$  at stylet support insertion site, extraordinarily wide as against an *Isohypsibius* species; 51  $\mu\text{m}$  long from mouth to point of entry to pharyngeal bulb. Stylet support present. Pharyngeal bulb oval (length 58  $\mu\text{m}$ , width 39  $\mu\text{m}$ , length to width ratio 1.49), ahead of first leg pair, with 3 large rod-shaped macroplacoids aligned, which are not constricted in their middle: size arrangement of macroplacoids III > I > II (I : II : III = 10.5  $\mu\text{m}$  : 5.8  $\mu\text{m}$  : 13.9  $\mu\text{m}$  = 0.8 : 0.4 : 1). No microplacoid. Paired buccal glands elongated, present outside of pharyngeal bulb.

Legs with claws of "Isohypsibius type": Double claws each on legs I-IV, external claw and internal one different in size, with claw sequence 2-1- 2-1 (secondary-primary-secondary-primary). Length ratios of primary branch to secondary one on external claw of leg I-IV 1.32, 1.49, 1.08, 1.03, respectively, with 2 accessory points at apical part of lateral margin.

Examined under scanning electron microscope, shapes of accessory points of external claw and internal one resembling each other, as shown in Fig. 2C. Primary branch on external claw of each leg pair diverging from

secondary one with an right angle or more. Basal branch expanded. All claws bearing very large lunule, but no cuticular bar.

Measurements and variations: Measurements based on eight specimens (holotype and seven paratypes) are shown in Table 1. Size arrangement of 3 rod-shaped macroplacoids is (I : II : III = 0.75 : 0.45 : 1) and the length to width ratio of pharyngeal bulb shows rather high consistency. The width of buccal tube is extraordinarily short and broad [length to width ratio of buccal tube is about 7.1 : 1, while about 21.75 : 1 in *I. myrops* (Bois-Reymond Marcus)]. Any cuticular ornamentation and eyespot were not found in all specimens examined. Exuviae contained 6-7 smooth eggs, diameter of which up to 65  $\mu\text{m}$ .

Etymology: The specific name, *brevitubulatus* (brevis, L.: short; *tubulus*, L.: tube; -atus, provided with) is taken from the short buccal tube, which is the most characteristic feature of this species.

Remarks: The present new species, *I. brevitubulatus*, may be easily distinguished from the other species of genus *Isohypsibius*, for it has a very large body and wide buccal aperture, as shown in many species of *Macrobotus*. In general shape of body and buccal apparatus, this species is most similar to *I. myrops* (Bois-Reymond Marcus), which was recorded only from Brazil (Bois-Reymond Marcus, 1944) and Mexico (Nelson and Schuster, 1982). However, as shown in Table 1, the present new species is clearly discernible from *I. myrops* in having the following combinations of characteristics: (1) body rather longer, up to mean 652  $\mu\text{m}$  (ranging 471-961  $\mu\text{m}$ ), (2) presence of lunules, (3) the percentage of width of buccal tube to length of pharyngeal bulb 14.6%, while 8.69% in *I. myrops*, (4) length to width ratio of buccal tube 7.1, while 21.75 in *I. myrops*, (5) size arrangement of three macroplacoids (I : II : III) is 8  $\mu\text{m}$  : 4.7  $\mu\text{m}$  : 10.9  $\mu\text{m}$  (0.75 : 0.45 : 1), while 6  $\mu\text{m}$  : 5  $\mu\text{m}$  : 8  $\mu\text{m}$  (0.75 : 0.6 : 1) in *I. myrops*.

Habitat and associated fauna: This species was collected from submerged sand bottom with rich organic detritus of Sömjin River, and co-occurred with the following aquatic invertebrates mostly known as indexing  $\beta$ - to  $\alpha$ -mesosaprobic waters in Korea: Cladocera - *Chydorus sphaericus* (O. F. Müller); Copepoda - *Elaphoidella grandidieri* (Guerne et Richard), *Parastenocaris* sp., *Eucyclops serrulatus* (Fischer), *Paracyclops fimbriatus* (Fischer), *Microcyclops varicans* (Sars).

*Isohypsiobius baldii* (Ramazzotti, 1945)  
(Figs. 2E, 3)

*Hypsiobius (Isohypsiobius) baldii* Ramazzotti, 1945 (cited from Bertolani and Balsamo, 1989); Ramazzotti, 1972 (p. 483, fig. 270).

*Isohypsiobius baldii*: Bertolani, 1982 (p. 65, fig. 35); Ramazzotti and Maucci, 1983 (p. 589, fig. 355); Bertolani and Balsamo, 1989 (p. 85, figs. 1-7).

Material examined: 2 specimens, Pongrae Fall, Ullüng I., March 4, 1995 (H. S. Rho and J. Y. Rho).

Remarks: This is the first record since Bertolani and Balsamo (1989) redescribed the present species after reconfirmation from the type locality to compensate the poor original description. Our specimens fit well with Bertolani and Balsamo's redescription except some minor discrepancies in the absences of eyespot and the size of cuticular granules, but these do not nevertheless represent the genuine species or subspecies distinction but may be attributed to the improper preparation or intraspecific deviation, and so are recognized as the same species with *I. baldi* (Ramazzotti).

Habitat and associated fauna: our specimens were collected from the submerged sand bottom covered with some fallen leaves and snows around a fall in Sönginbong Mt., Ullüng I., the East Sea of Korea. The associated fauna co-occurred were: Gastrotricha - *Chaetodontus laroides* (Marcolongo), *C. maximus* (Ehrenberg); Copepoda - *Canthocamptus* sp., *Maraenibiotus brucei* (Richard), *Diacyclops languidoides* (Lilljeborg), which are the representative species from cold and oligotrophic waters, especially frequent in mountain waters in early spring of Korea. This species is considered to be rare, and to be restricted to cool mountain waters.

Distribution: Italy and Korea.

*Isohypsiobius marcellinoi* (Binda and Pilato, 1971)  
(Figs. 2F, 4)

*Hypsiobius (Isohypsiobius) marcellinoi* Binda and Pilato, 1971 (p. 906, fig. 50).

*Isohypsiobius marcellinoi*: Pilato, 1974 (p. 242); Bertolani, 1982 (p. 77, fig. 42); Ramazzotti and Maucci, 1983 (p. 636, fig. 396); Pilato and Binda, 1988 (p.

52, fig. 2a-c); Biserov, 1991 (p. 203); McInnes, 1994 (p. 308).

Material examined: 1 specimen, small fountain near the top of Hyangchökpöng Hill (1,614 m), Tökyu Mt. (water temperature 9.2°C, pH 6.9), Oct. 13, 1995 (H. S. Rho and K. H. Lee); 25 specimens, Wölpochön Stream, Yöngil-kun (water temperature 19.3°C, salinity 1‰), Oct. 6, 1995 (H. S. Rho and K. H. Lee).

Remarks: we could not find any morphological differences between the specimens from a spring at hilltop of Tökyu Mt. and ones from an estuary, and both of them were fully coincided to the original description of Binda and Pilato (1971).

Habitat and associated fauna: This species was originally recorded from mosses in Sicily, Italy and later in the Simento River, Sicily (Pilato, 1974). Our specimens were collected from a mountainous spring, and from about 50 m upstream from a mouth of small stream draining into the East Sea of Korea. The former was cool and clear, and surrounded by mossy rocks with coarse sand bottom, and inhabited by the copepod *Acanthocyclops* sp. and *Canthcamptus* sp., while the latter was brackish water with fine sands submerged, and flourished by  $\beta$ - to  $\alpha$ -mesosaprobic cladocerans [*Chydorus sphaericus* (O. F. Müller), *Simocephalus vetulus* (O. F. Müller)] and copepods [*E. serrulatus*, *Megacyclops viridis* (Jurine), *Mesocyclops* sp.] between littoral macrophytes. So, this species seems to adapt to wide range of water bodies including brackish waters with low salinity.

Distribution: Italy (Sicily), Russia, U.S.A. (East Tennessee), and Korea.

Subfamily Diphasconinae Dastych, 1992  
Genus *Diphascon* Plate, 1889  
Subgenus *Diphascon* Pilato, 1987

*Diphascon (Diphascon) higginsi* Binda, 1971  
(Figs. 2G, 5)

*Diphascon higginsi* Binda, 1971 (p. 761, figs. A-C); Pilato, 1974 (p. 76, figs. 1, 2); Pilato, 1975 (p. 285, fig. 2); Binda and Guglielmino, 1982 (p. 209); Weglarska and Korecka, 1983 (p. 89); Ramazzotti and Maucci, 1983 (p. 283, fig. 119); Maucci, 1987 (p. 205); Dastych, 1988 (p. 188, figs. 125 D-G).

*Hypsiobius (Diphascon) higginsi*: Hallas, 1977 (p. 176). *Diphascon (Diphascon) higginsi*: Pilato, 1987 (p. 350); Ito, 1995 (p. 23, fig. 3).

Material examined: 1 specimen, a bog beside Yulsansöwön, Kyöngsan city, June 8, 1995 (C. Y. Chang, J. M. Lee, and H. S. Rho); 12 specimens, same locality (water temperature 12.0°C, pH 8.2), Apr. 15, 1997 (C.

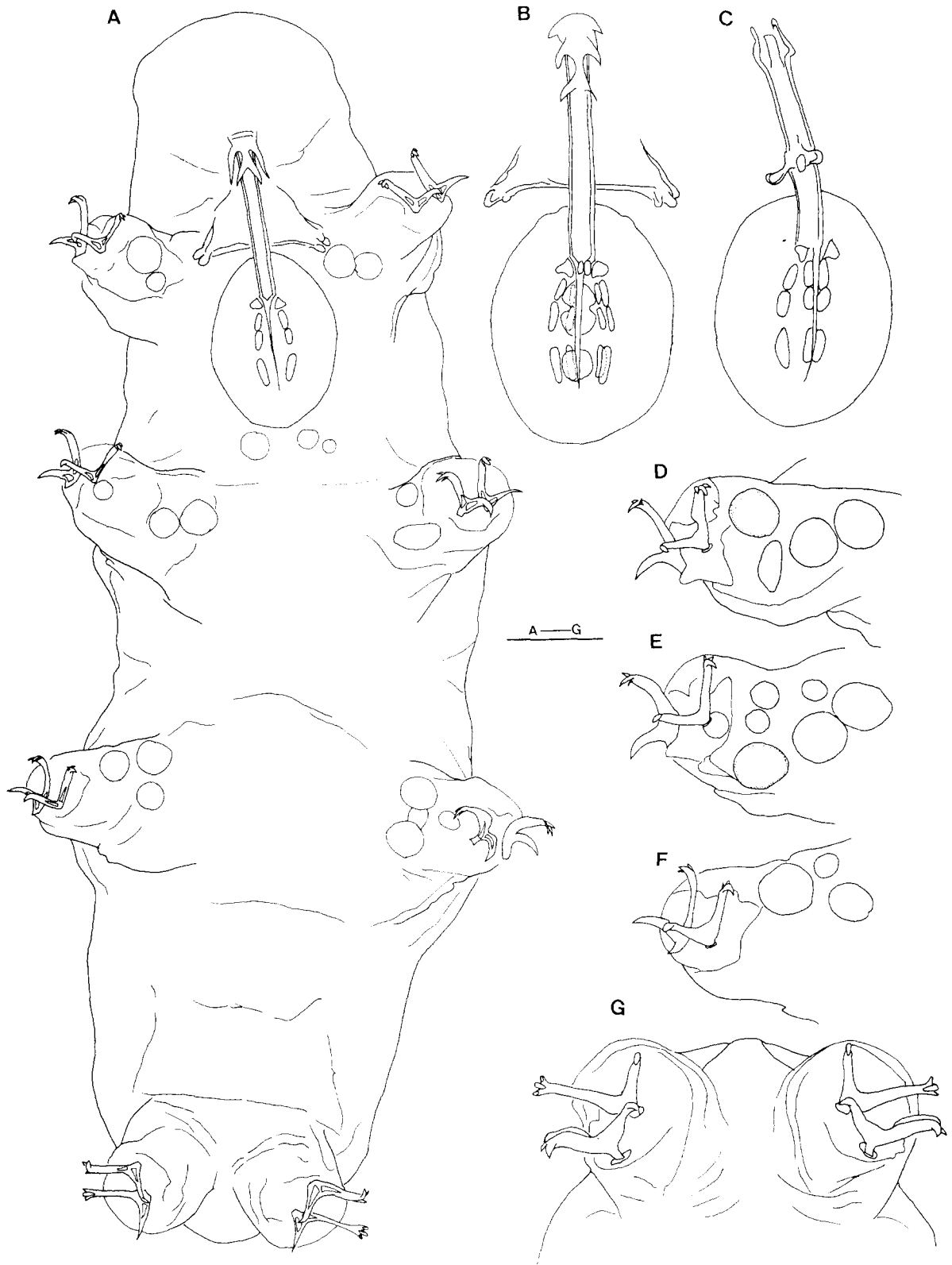


Fig. 3. *Isohypsibius baldii* (Ramazzotti). A, Habitus, ventral view. B, Bucco-pharyngeal apparatus, ventral view. C, Bucco-pharyngeal apparatus, lateral view. D, Claws on leg I. E, Claws on leg II. F, Claws on leg III. G, Claws on leg IV. Scale bar=0.01 mm.

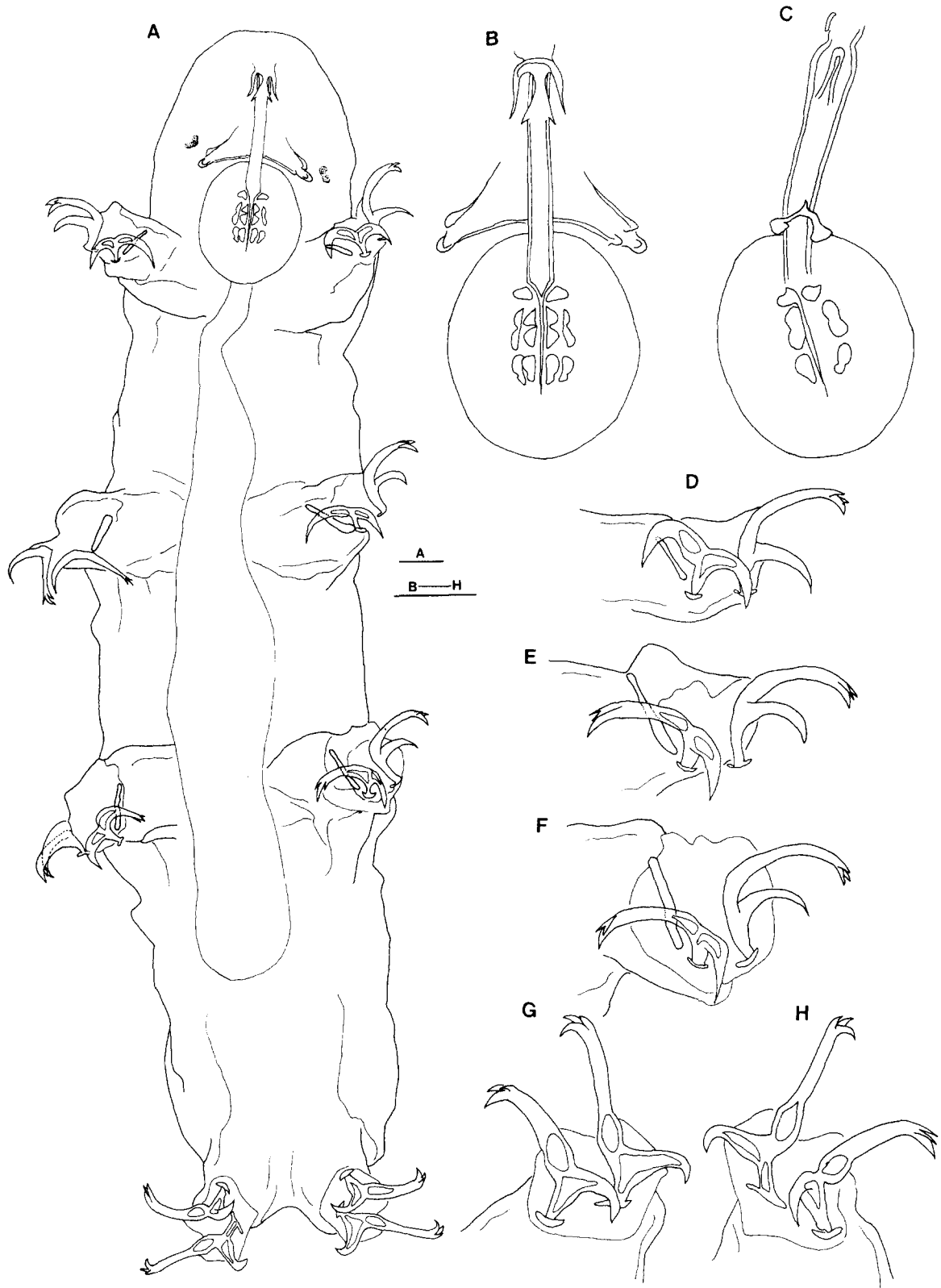


Fig. 4. *Isohypsibius marcellinoi* (Binda and Pilato). A, Habitus, ventral view. B, Bucco-pharyngeal apparatus, ventral view. C, Bucco-pharyngeal apparatus, lateral view. D, Claws on leg I. E, Claws on leg II. F, Claws on leg III. G-H, Claws on leg IV. Scale bars=0.01 mm.

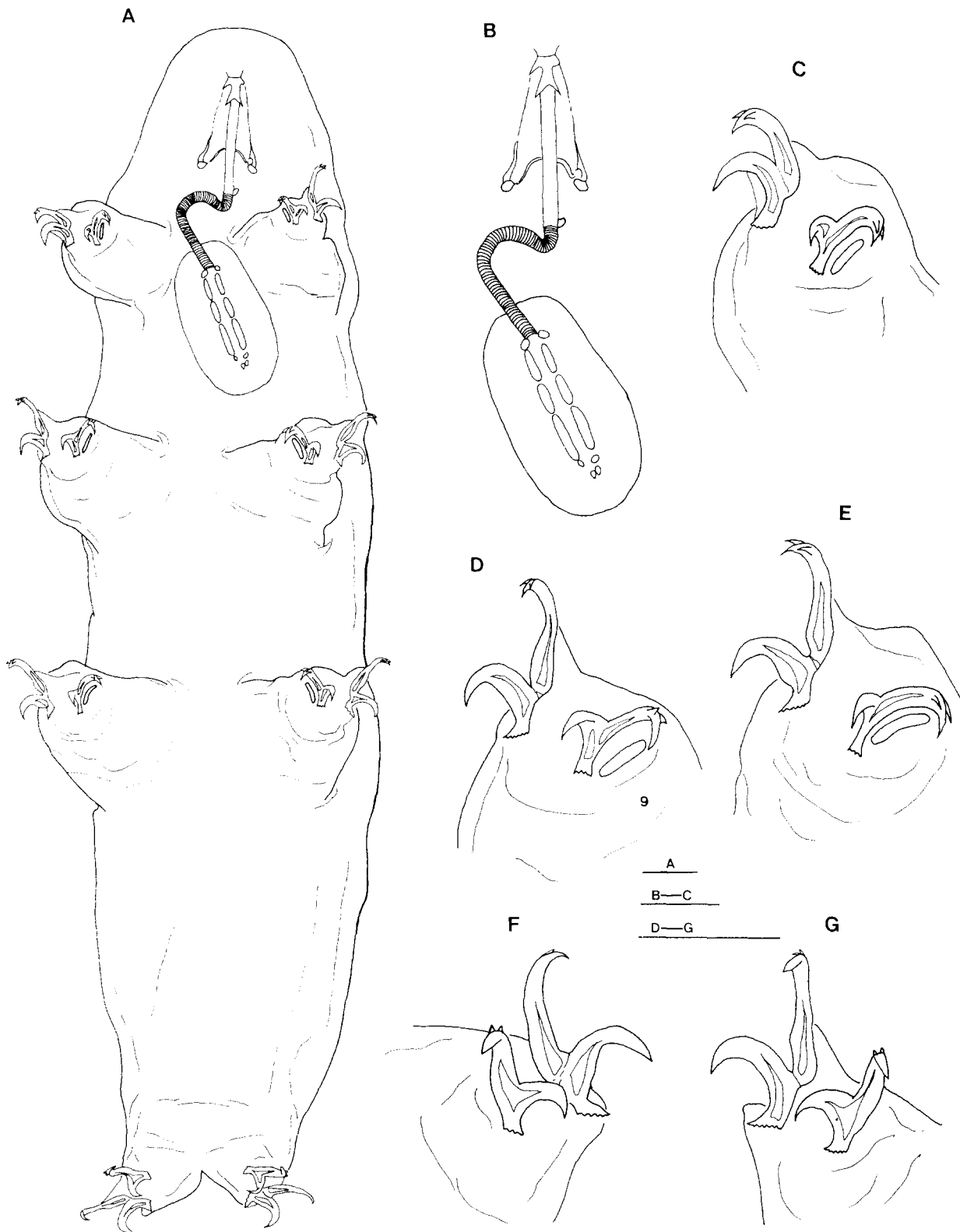


Fig. 5. *Diphascocon (Diphascocon) higginsi* Binda. A, Habitus, ventral view. B, Bucco-pharyngeal apparatus, ventral view. C, Claws on leg I. D, Claws on leg II. E, Claws on leg III. F-G, Claws on leg IV. Scale bars=0.01 mm.



Y. Chang, and H. S. Rho).

Remarks: The presence of drop-like thickening on the wall of buccal tube was not mentioned in the description of Binda (1971), and also in Pilato (1975). Later, after the observation of the type specimens, Pilato (1987) confirmed its presence and attributed it to the subgenus *Diphascoen* (Ito, 1995). Our specimens bear also the same structure as shown in Fig. 5B. Other characteristics are also quite well fitted to the original description as well as Dastyh's (1988) and Ito's (1995).

Habitat and associated fauna: This species has been reported worldwide usually from terrestrial or aquatic mosses. Our specimens were collected from a stagnant bog with plentiful of organic detritus, and co-occurred with the aquatic invertebrates which usually frequent in the eutrophicated swamps or transient pools in Korea: Rotifera - *Polyarthra major* (Burckhardt); Gastrotricha - *Polymerurus* sp.; Cladocera - *Chydorus sphaericus* (O. F. Müller); Copepoda - *Macrocyclus albidus* (Jurine), *Eucyclops serrulatus* (Fischer), *Diacyclops thomasi* (Forbes), *Megacyclops viridis* (Jurine).

Distribution: Morocco, Poland, Italy, Finland, Russia, New Zealand, U.S.A. (Tennessee), Japan and Korea.

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