A New Genus and four New Species of semiterrestrial Water-Bears from South Africa (Tardigrada)

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ABSTRACT: Four new semiterrestrial tardigrades, Calcarobiotus filmeri gen. n. sp. n., C. occultus sp. n., Macrobiotus drakensbergi sp. n. and Ramazzottius theroni sp. n., from South African mosses, lichens and soil are described. The new genus closely resembles Macrobiotus, but differs from it by the presence of unique spurs on the bases of the claws.

KEYWORDS: Semiterrestrial tardigrades, South Africa, new taxa.

Introduction

Knowledge of South African water-bears or tardigrades, widely distributed and common but neglected micrometazoans, are fragmentary and restricted to a few contributions. From these papers, the oldest and the most comprehensive (Murray 1907) lists 14 species. A further work by the author (1913) surveying the Central African fauna, reports from South Africa five other tardigrades. DASTYCH (1980, 1992) described two new species and a new genus, BINDA (1984) recorded 11 and PILATO, BINDA & CATANZANO (1991) three taxa from that country. Now 28 nominal tardigrade species are known from South Africa.

In neighbouring Namibia five taxa had been found (MARCUS 1936, PILATO, BINDA & CATANZANO 1991), three other species in Mozambique (BINDA 1984, op. cit.). All these represent however only a tiny fraction of the tardigrade fauna that can be expected in the Southern African Region.

The objective of the present paper is a description of four new South African tardigrades, of which two species also belong to a new genus.

Materials and Methods

The animals and their eggs were extracted from moss-, lichen- or soil samples by the method described by DASTYCH (1984) and mounted on microslides in gum chloral (FAUER's medium). Observations and measurements were carried out in phase or interference contrast. Unless stated otherwise, all measurements are for holotype. Abbreviations used in illustrations (Figs 1-40) are as follow: ar = areoles between processes of egg, ba = transverse cuticular bar, dl = dorso-lateral ridge in mouth cavity, ey = eye spot, in = incision on macroplacoid III, lb = light-refracting unit of primary branch, lu = lunula, p = pores between processes of egg, pb = primary branch of claw, pn = pharyngeal needle, pr = posterior ring of granulation in mouth cavity, s = spur on claw, sb = secondary branch of claw, ss = sensory structure.

The type material is deposited in the Zoologisches Museum, Universität Hamburg, Germany (ZMH) and the Natal Museum, Pietermaritzburg, South Africa (NM).

Descriptions of New Taxa

Calcarobiotus gen. n.

Diagnosis: Semiterrestrial macrobiotids with buccopharyngeal apparatus of Macrobiotus-type. Claws V-shaped, their bases and branches wide, their formula “2112”.
Figs. 1-6. *Calcarobiotus filmeri* sp. n.: 1- whole animal in ventral view; 2 and 3- armature of mouth cavity, dorsal and ventral view, respectively; 4- claws of legs I; 5- internal claw of leg II, lateral view; 6- claws of legs IV (Holotype. All abbreviations are explained in “Materials and Methods”).

A pair of unique symmetrical spurs protrude laterally from the bases of all claws. Transverse cuticular bar on ventrum of legs I to III. Freely layed eggs with processes on shell.

**Type Species:** *Calcarobiotus filmeri* sp. n.

**Etymology:** *Calcar* (L.) = spur, *bios* (Gr.) = life.

**Species List:** *C. filmeri* sp. n., *C. occultus* sp. n.

**Remarks:** The new genus is closely related to *Macrobiotus*, as judged by its type of buccopharyngeal apparatus, claws and egg processes. However, autapomorphic spurs on the bases of its claws, not known in other eutardigrades, and transverse bar on legs I to III separate the new genus *Calcarobiotus* from other genera of the
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Macrobiotidae. The spurs enhance the clinging property of claws during locomotion of animal. Somewhat similar spur-like structures, but convergently evolved, had been described in the marine genus *Anisonychus* Pollock, 1975, of the Class Heterotardigrada. The transverse cuticular bars occur rarely in Macrobiotidae, Eohypsibiidae and Hypsibiidae, but are characteristic for highly derived Milnesiidae.

![Image](image)

**Figs. 7-8. Calcarobiotus filmeri** sp. n.: 7(A) - processes of egg, lateral view, (B) - bases of the processes, dorsal view; **Calcarobiotus occultus** sp. n.: 8(A) - processes of egg, lateral view, (B) - bases of the processes, dorsal view.

**Calcarobiotus filmeri** sp. n.

Figs. 1-7, 9-16, 36-39

**Diagnosis:** Median sized *Calcarobiotus* with anterior eyes. Pharynx with three macroplacoids and a microplacoid. Conical processes of the egg shell with strongly elongated tips, covered with irregular granulation. The shells surface between the processes with small areoles.

**Material examined:** 2 specimens, 2 eggs.

**Description:** Body whitish and 470 μm longs in holotype (paratype: 132 μm). Cuticle smooth, thick and with several small pores. Eyes located anteriorly and small (paratype without them but they are present in embryo). The buccopharyngeal apparatus moderately sized (Fig. 1). Mouth opening surrounded by a ring of 10 peribuccal lamelles. Anterior part of mouth cavity with a ring of dense granulation composed of tiny granules (Fig. 3). Posterior ring with bigger granules, elongated on ventral (Fig. 3), but more rounded on dorsal side (Fig. 2). Dorsal and ventro-lateral ridges well developed, ventro-median ridge composed of 4 granules (2-3 in juvenile forms) (Fig. 3). Mouth tube 56 μm long, 8 μm wide. Stylets relatively robust, with "typical" furca.

Pharynx ovoid (53 x 47 μm) with three macroplacoids and a distinct microplacoid (Figs. 10, 11), the first macroplacoid longest, the second the shortest. The lengths of placoids as follow: I= 7.7, II= 4.4, III= 6.6, microplacoid 5 μm. The third macroplacoid with a small incision in its latero-posterior part (Fig. 11). Juvenile forms without the incision.
Claws of *Macrobiotus*-type, V-shaped. A pair of unique spurs protrude laterally from the claw bases (Figs. 4-6, 12-16). The claw bases are wide, short and provided with their own cuticular walls (Figs. 4, 6, 12, 13). Primary and secondary branches of each claw relatively short and not much differentiated in size, with the primary branch slightly longer (Fig. 13). Accessory spines on the primary branches relatively long but delicately formed. The claw lunules on leg I to III smooth, median sized (Figs. 4, 13), on legs IV large (9 μm wide) and with dentated edges (Figs. 6, 16). At the bases of claws on legs I to III a single elongated cuticular bar, more or less bent (Figs. 1, 4, 13, 15). The length of claws I including lunules, 14 μm, claws IV= 17 μm.
The eggs are whitish, their shells covered with relatively small conical processes, which have strongly elongated tips often ended with 2-3 smaller, irregularly shaped branches (Figs. 7A, 37, 38). Surface of processes with irregular granulation. Base of each process surrounded by a ring of small irregularly shaped meshes, creating a kind of areolation (Figs. 7B, 39). The processes are 12-18 \( \mu m \) long, and their bases 4.5-6.0 \( \mu m \) in diameter. Diameter of areoles 1.0-1.5 \( \mu m \). Eggs 78 and 88 \( \mu m \) in diameter each, excluding the processes.

Holotype: Sex indet., 470 \( \mu m \); 7 May 1988, coll. M. R. FILMER.

Type Locality: Transvaal, Waterriver Valley, between Lydenburg and Bergersdorf, 2000 m a.s.l. On the ground, in a sample of soil, detritus and tiny roots; (holotype and paratype, 2 eggs).

Type Repositories: Holotype, embrionate egg (slide No. 1) and one paratype with egg (Slide No. 2) in the collection of NM.

Etymology: The species is named in honour of its collector, Dr. MARTIN R. FILMER (Johannesburg).

Remarks: Adults of *C. filmeri* sp. n. are extremely similar to the second new species of *Calcarobiotus* gen. n. Distinct difference between both taxa occurs in the morphology of their eggs, as presented below, at the description of *C. occultus* sp. n.

*Calcarobiotus occultus* sp.n.

Figs. 8, 40-42

**Diagnosis:** Median sized *Calcarobiotus* without eyes. Pharynx with three macroplacoids and a microplacoid. Processes of the egg shell bell-shaped, smooth, their tips short and spine-like. Shell surface between processes covered with closely spaced minute pores.

**Material examined:** 9 specimens, 1 egg.

**Description:** Body whitish and 200 \( \mu m \) long, in the holotype measured on a shrunked specimen (paratypes: 126-417 \( \mu m \)). Cuticle smooth, eyes absent.

The buccopharyngeal apparatus as in *C. filmeri* sp. n., ventro-median ridge short, transversal bar. In some specimens the ridge composed of 2-3 granules. Mouth tube 51 \( \mu m \) long, 7 \( \mu m \) wide. Pharynx 37 x 41 \( \mu m \) (squashed), I macroplacoid 6 \( \mu m \), II= 4, III= 6.2 \( \mu m \) long. Length of microplacoid 4 \( \mu m \).

Claws as in *C. filmeri*, transverse cuticular bars on legs I to III weaker developed, compared to those of former taxon. The claws I= 8 \( \mu m \), IV= 11 \( \mu m \) long (including lunules).

The only egg found is whitish and measures 77 x 68 \( \mu m \), excluding the processes on its shell. The processes small, bell-shaped and end usually with a tiny, spine-like structure (Figs. 8A, 42). This spine occasionally absents. About 32 processes on the circumference of the egg. Surface of the processes smooth, i.e. without granules, but with 1-3 indistinct cuticular rings (Fig. 8A). Surface between processes covered with a net of minute and closely spaced pores, each 0.3-0.5 \( \mu m \) in diameter (Figs. 8B, 41). Length of processes, including apical spine, 4-6 \( \mu m \), their width at the base 3.5-4.5 \( \mu m \). Embryo similar to that of *C. filmeri*.

**Holotype:** Sex indet., 200 \( \mu m \). 30 May 1988, coll. H. DASTYCH.

**Type Locality:** Natal, Central Drakensberg. Cathedral Peak Area, 1920 m a.s.l., meadow near artificially planted pine forest; in soil between grass-tussocks (holotype, 5 paratypes, embrionate egg). Other localities: Cathedral Peak Area, the vicinity of the Ukahlamba (the 'Museum's House'), moss from rocks, 1660 m a.s.l., 30 May 1980, coll. H. DASTYCH (1 paratype). Cathedral Peak Area, Tarn Hills, 1940 m a.s.l. Moss from basalt rocks, 9 July 1988, coll. I. PAJOR (1 paratype). Pretoria, Botanical Garden, moss from stone. June 1971, coll. A. SZEPTYCKI (1 specimen).
Figs. 17-23. *Macrobiotus drakensbergi* sp. n.: 17- whole animal in ventro-lateral view; 18- armature of mouth tube, ventral view; 19- buccopharyngeal apparatus, dorsal view; 20- processes of egg, lateral view; 21- processes of egg, dorsal view; 22- external claw of leg I; 23- internal claw of legs IV (Figs. 17, 18: holotype).

**Type Repositories:** Holotype and 3 paratypes (slide No. 1), 2 paratypes and egg (slides Nos 2-4) are deposited in NM, one paratype (coll. I PAJOR: slide No. A5) and the specimen from Pretoria in the collection of ZMH (Reg. No. A5/93).

**Etymology:** *occultus* (L.)= concealed, secret.

**Remarks:** The new species closely resembles *C. filmeri* but can be separated from the latter through the lack of eyes, eye-dots being present in *C. filmeri*. However, in the latter species in one of three specimens examined (including embryo) this character could not be found. Distinct differences between the both taxa occur in the morphology of their shells. In *C. occultus* the processes of the egg shell are bell-shaped, short and smooth, while those of *C. filmeri* are conical, elongated and covered with irregular granulation. Moreover, in the latter taxon, the egg surface between the processes is covered with areoles and in *C. occultus* occurs there tiny and densely distributed pores, similar to those in *Macrobiotus hufelandi*-complex. Thus, identification of the species can only be based with certainty using the morphology of their eggs, a common procedure for the Macrobiotidae. Consequently, the identification of the specimen from Pretoria (i.e. that without eyes) is tentative.
The morphology of egg shell is recognized as a significant taxonomic character for the Macrobiotidae (Pilato 1975). However, a distinct variability might occur within various population of the same species (see, for example, Macrobiotus blocki: Dastych 1984). The descriptions of C. filmeri and C. occultus are based on small samples of specimens and eggs and therefore the intraspecific variability remains unknown. Hence, one cannot exclude that both taxa may turn out in the future to be conspecific. However, against this points the different structural pattern of their egg surface, which is relatively constant and species-specific character in the Macrobiotidae, compared to the at times variable shape and size of the egg processes.

Macrobiotus drakensbergi sp.n.
(Figs. 17-23, 43)

Diagnosis: Median sized Macrobiotus with two macroplacoids and a microplacoid. Claws dumpy, with dentated lunules. Processes of eggs short, smooth and cone-shaped. The egg surface between the processes with densely distributed minute pores.

Material examined: 30 specimens, 21 eggs.

Description: Body whitish, 526 µm long in holotype (paratypes: 188-610 µm). Cuticle, including that on legs, smooth. Eyes distinct and located posteriorly, four specimens without eyes. Legs relatively small, the fourth pair greatly reduced (Fig. 17). The intestine contents green or brownish-green.

Buccopharyngeal apparatus moderately sized (Figs. 17, 43). Mouth opening surrounded by a ring of 10 peribuccal lamelles. Mouth cavity without rings of granules, dorso-median ridge not separated (Fig. 19). Ventro-lateral ridges weakly formed, ventro-median ridge reduced to one small granule (Fig. 18). Larger specimens with 2 or even 3 such granules, two specimens without them. Mouth tube 48 µm long and 5.5 µm wide.

Pharynx oval (39 x 34 µm), with two macroplacoids and a small, thin microplacoid (Figs. 19, 43). Small pharyngeal needles present (Fig. 19). Apophyses relatively small. The first macroplacoid 13 µm long, the second 7 µm. Length of microplacoid 3 µm. The first macroplacoid with small lateral incisions located slightly below its middle.

Legs short, claws relatively small. Claws dumpy, with a rounded base and distinct, dentated lunules (Figs. 22, 23). External claws and their lunules slightly larger than internal ones. Claw branches distinctly sclerotized (Fig. 22). Accessory spines on the main branches small. Lunules on legs I to III with 4-12, on legs IV with 12-16 teeth. The claws I= 12 µm, IV= 13 µm (including lunules).

Eggs whitish, their shells covered with cone-shaped, short and sharply pointed processes (Figs. 20, 21). Surface of the processes smooth, that between the processes with tiny, closely spaced pores about 0.3 µm in diameter (Fig. 21). The processes 5.5-7.0 µm long (usually 5 m), their bases 4.0-5.5 µm in diameter (mostly 4.0-4.5 µm), are rarely bluntly ended (Fig. 20). Diameter of eggs 78-92 m, excluding their processes.

The relatively short, reduced legs make the locomotion of the species somewhat similar to that of Macrobiotus xerophilus (DASTYCH, 1978), which is characteristically a snakelike movement.

Holotype: Sex indet., 526 µm. 9 July 1988, coll. I. PAJOR.
Type locality: Natal, Central Drakensberg, Cathedral Peak Area. Tarn Hills, 1940 m a.s.l., in mosses from rocky (basalt) surface, (holotype, 14 paratypes, 13 eggs: some embryonated). Other locality: Cathedral Peak Area, the vicinity of Ukhahlamba (the 'Museum's House'), 1660 m a.s.l., moss from rocks, 30 May 1988, coll. H. DASTYCH (15 paratypes, 8 eggs).
Type repositories: The holotype and 3 eggs (slide No. 1) and 9 paratypes and 8 eggs (slides Nos 12,13) in ZMH (Reg. No. A6/93), 20 paratypes and 10 eggs (slides Nos 2-11) in NM.
Etymology: The species name refers to the Drakensberg Mts.
Remarks: The new species is similar to Macrobiotus hufelandi (SCHULTZE, 1834) as far as its buccopharyngeal apparatus and the shape and distribution of pores on the egg shell between the egg processes is concerned. On the other hand, it resembles M. crenulatus RICHTERS, 1904 (=M. echinogenitus RICHTERS, 1904, partim: see BINDA 1988), with regards to its distinctly dentated lunules and the cone-shaped processes
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M. drakensbergi can readily be separated from M. hufelandi through its dentated lunules, those of the latter species being smooth or delicately curled. Moreover, the egg processes in M. hufelandi are shaped as inverted egg-cups, while those of M. drakensbergi are cone-like. The new species differs from M. crenulatus chiefly through its smaller apophyses, relatively longer first macroplacoid, the difference in type of lunula dentation (see DASTYCH 1987, Binda 1988), the lack of cuticular granulation on the legs and distinct pores in the cuticle. Furthermore, the network of pores between the egg processes is distinct from the smooth surface of M. crenulatus, while the processes of the latter species, covered with a network of tiny meshes, are bigger and less numerous, compared to those of the new taxon. Also the apophyses are larger and the first macroplacoid is only slightly longer than the second one in M. crenulatus. The above characters place M. drakensbergi between the taxa of the hufelandi- and crenulatus (=echinogenitus)-complex, the position being closer to the former group than to the latter.

Ramazzottius theroni sp.n.
(Figs. 24-35, 44, 45)

Diagnosis: A median sized Ramazzottius with eyes and granulated cuticle. Freely laid eggs, their processes cone-shaped and usually with elongated tips. Surface between the processes distinctly granulated.

Material examined: 83 specimens, 20 eggs.

Description: Body light-pink, 290 μm long (paratypes: 150-320 μm). Some specimens with small clumps of dark-brown pigment. Dorsum, lateral sides of the body and legs sculptured, the sculpturing being composed of hemispherical, polygonal thickenings (Figs. 27, 44, 45) which increase slightly in size posteriorly. The thickenings 2-6 μm wide (usually 3-4 μm). In dorsal view, the polygons internally with closely spaced irregularly shaped tiny granules, about 0.3 μm in diameter. In lateral view these granules resemble minute rods. Eye-dots small, usually located anteriorly and composed of a few dark-brown pigment granules. A pair of small elliptical sensory structures (Fig. 25) dorsally in the head region. Ventral cuticle smooth.

Mouth opening located ventro-anteriorly and surrounded by 6 lobes. Mouth cavity small and without granulation. Mouth tube 30 μm long and 2.8 μm wide. Pharynx almost round (28 x 26 μm), with two small grain-like macroplacoids. No microplacoid. Apophyses relatively large (Fig. 25), 2.2 μm in length. First macroplacoid 3.3 μm long and with hardly discernible lateral incisions in the middle of its length. Second macroplacoid 2.2 μm long, smooth.

Claws moderately sized, their length increases slightly towards the rear of the animal. Accessory spines small. Lunules absent, except in the biggest specimens, where they are then thin, rudimentary structures on the internal claws (Fig. 27). Primary branches of external claws moderately long and with poorly sclerotized base, thus appearing as a short, differently light-refracting basal unit (Fig. 28). External claws on leg IV 20 μm long, their primary branches 14 μm in length.

The eggs are whitish. Egg shell with variably shaped, cone-like processes, each mostly with more or less elongated tips (Figs. 29-35). The processes rather sparsely distributed and usually smooth. Their surface in some eggs is, however, covered with 1-2 tiny, elongated thickenings or irregularly shaped granule(s). Egg surface between the processes distinctly granulated (Figs. 34, 35), the granules being more or less regular in shape, 0.3-0.5 μm in diameter and rather closely spaced. The processes 4-13
μm long (usually 6.5-8.0 μm) and at their bases 2.2-3.3 μm wide. Diameter of eggs 56-78 μm, excluding the processes. Two eggs with embryos.

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Figs. 36-45. Calcarobiotus filmeri sp. n.: 36- egg shell and embryo; 37 and 38- processes of egg; 39- surface of egg; Calcarobiotus occultus sp. n.: 40- distal part of buccopharyngeal apparatus; 41- surface of egg, dorsal view; 42- processes of egg, lateral view; Macrobiotus drakensbergi sp. n.: 43- front of the body with buccopharyngeal apparatus; Ranaazzottius theroni sp. n.: 44- dorsal cuticular sculpture, rear of the body; 45- claws of legs IV (Figs. 36-38, 42: contrast phase; the others: interference contrast).
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Holotype: Sex indet., 290 μm. 5 December 1988, coll. P. D. THERON.
Type locality: SW Cape, about 4 km W of Bonnievale, southern slope of Rooiberg, 195 m a.s.l. Lichens from stems of various Karoo-bushes, (holotype, 75 paratypes and 20 eggs); as above, northern slope (7 paratypes).
Type Repositories: Holotype, 64 paratypes and 15 eggs are housed in ZMH (slides Nos 1-7; Reg. No. A7/93), 18 paratypes and 5 eggs (Nos. 8, 9) in NM.
Etymology: The species is named in honour of its collector, Prof. Dr. PIETER D. THERON (Potchefstroom).
Remarks: The new taxon resembles most closely R. tribulosus BERTOLANI & REBECCHI, 1988, among the 10 nominal species of the genus Ramazzottius. It can easily be separated from that species, however, by the presence of eyes as well as the relatively larger and internally granulated cuticular thickenings on the dorsum and by the granulated surface of the egg shell between the egg processes, the dorsal sculpture of R. tribulosus being composed of smaller and more rounded thickenings deprived from tiny granules, while the surface between the processes is smooth. Moreover, the processes in R. tribulosus have irregular sculptures internally, those in R. theroni being usually smooth.

The presence of eyes, a plesiomorphic and rare character in Ramazzottius, has been reported only in the Argentinian R. saltensis CLASP & ROSSI, 1984. This latter species has 8 transversal rows of tubercles dorsally, however, these being absent in R. theroni.

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References


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