

**A New Family of Arthrotardigrada
(Tardigrada: Heterotardigrada) from the
Atlantic Coast of Florida, U.S.A.¹**

REINHARDT MØBJERG KRISTENSEN² AND ROBERT P. HIGGINS

Department of Invertebrate Zoology, National Museum of Natural History,
Smithsonian Institution, Washington, D.C. 20560, U.S.A.

Abstract. A new interstitial arthrotardigrade, *Renaudarctus psammocryptus* n. gen., n. sp., is described from high-energy marine beaches of Fort Pierce and Boca Raton, Florida. It is found in sediment consisting of stratified layers of coarse quartz sand and shell hash. The tardigrade is distinguished particularly by its dorsal cuticular plates, toes, and claw structure. The Renaudarctidae is established as a new family in the order Arthrotardigrada. A recently described tardigrade, *Neostygarctus acanthophorus* from a marine cave in Italy, originally placed in the family Stygarctidae, is included in the Renaudarctidae based on toe and claw structure. The phylogenetic relationships of Renaudarctidae to other families in Heterotardigrada are discussed. Seven other species of tardigrades found with the new species included: *Parastygarctus sterreri*, *Stygarctus goubaultae*, *Halechiniscus remanei*, *Raiarctus colurus*, *Raiarctus* sp., *Tanarctus tauricus*, and *Batillipes bullacaudatus*. Only *B. bullacaudatus* and *H. remanei* have been reported from the U.S.A. previously. The vertical and horizontal distribution of the tardigrades in the beaches appears to be related to granulometry more than to other physical or chemical factors, including depth in the beach.

Since the discovery of the first marine tardigrade, *Microlyda* Dujardin, 1851, most marine biologists have considered these microscopic metazoans to be a rare component of the marine ecosystem. The first monograph on this phylum (Marcus, 1929) recorded only six marine species. Schulz (1935, 1951, 1955) described several new species. Since 1955, the most significant taxonomic contribution has been that of Renaud-Mornant, who has produced 32 publications and described 9 genera and 27 species. In a recent review of the marine Tardigrada (see Renaud-Mornant, 1982), 22 genera of Heterotardigrada were noted. These animals, a total of 56 species, have been found in a wide variety of marine habitats from the deep sea to tropical beaches.

¹ The material was obtained through the kind cooperation of the Smithsonian Marine Station at Link Port, Florida. We especially acknowledge the technical assistance of William Lee, of the Smithsonian Marine Station at Link Port, who assisted with sampling and processing; the cooperation of Carolyn Gast, of the Department of Invertebrate Zoology, Smithsonian Institution, who prepared the illustrations; and the aid of Marie Wallace, also of the Department of Invertebrate Zoology, who assisted with sample processing and in the preparation of the manuscript. Our appreciation is also expressed to Horton H. Hobbs, Jr., who critically reviewed this manuscript and made many valuable suggestions. This research was supported by the Danish Natural Science Research Council (Grant No. 11-3558), a NATO Science Fellowship and a Smithsonian Postdoctoral Fellowship to the senior author, and funds made available to the junior author through the Smithsonian Marine Station.

² Institute of Cell Biology and Anatomy, University of Copenhagen, Universitetsparken 15, DK-2100 Copenhagen Ø, Denmark.

In the United States, the majority of marine tardigrade species have been reported from intertidal sandy beaches of the East Coast (Hallas & Kristensen, 1982; Lindgren, 1971; McGinty & Higgins, 1968; McKirdy, 1975; Pollock, 1970a,c, 1979). Although the marine tardigrades from intertidal, mostly high-energy, marine beaches of North America are relatively well known, little is known about these animals from other marine habitats. Renaud-Mornant (1982) predicted a remarkable increase in the number of species once deep-sea, coral sand, and other substrates were sampled. Indeed, five species of arthrotardigrades were described recently from the subtidal sediment of marine caves along the Italian coast (Grimaldi de Zio et al., 1982a,b), and the first semipelagic tardigrade, *Tholoarctus natans*, was found in subtidal coarse sand and shell gravel collected along the northern coast of France (Kristensen & Renaud-Mornant, 1983).

In this investigation, a relatively new technique of momentarily subjecting large quantities of coarse sediment to freshwater (Kristensen & Higgins, 1984) is used. Furthermore, we have taken samples deeper than usual, to 1.8 m below the beach surface. Our research was directed toward substrate choice of the different species in a stratified coarse quartz sand habitat with layers of shell hash. In this paper, the first of two on tardigrades from coarse sediment, a new family of Arthrotardigrada is described. A future paper will include descriptions of tardigrades found in the same kind of sediment located subtidally, about 10 km off the coast of Fort Pierce, Florida.

MATERIALS AND METHODS

Samples were collected from two different beaches located on the East Coast of Florida, Pepper State Park Beach at Fort Pierce and Red Reef Park Beach at Boca Raton. During the period of investigation, from 5 October 1982 to 9 April 1983, almost monthly samples were taken within 1 h of low tide along a transect of four stations, extending from mid- to low-tide regions, perpendicular to the shore. A single hole was dug at each station to ground-water level. The ground-water salinity, measured with an American Optical refractometer, and the temperature were recorded. From each hole, about 30 kg of sediment were placed in a plastic bucket and saturated with seawater.

Extraction of organisms from the sediment was accomplished by transferring small amounts (a few handfuls) of sediment to a bucket containing about 10 liters of freshwater. The osmotic shock causes the tardigrades (and other interstitial organisms) to release their otherwise tenacious hold on the sand grains. The sediment was agitated gently and the water decanted through a 62- μ m mesh net. After repeating this process once with the same sediment, the sediment was discarded and a unit of untreated sediment was used to repeat the process. After the second unit had been processed, the contents of the net were washed into a container with 2–3% formalin buffered with Borax, or fresh seawater was used to wash the animals into a watch glass for live observation. (The reintroduction of seawater allowed most of the meiofauna to recover from the osmotic shock.) The animals were observed with the aid of 50 \times magnification of a stereomicroscope and by 1,250 \times oil immersion phase-con-

trast microscopy. Sorted animals later were transferred with a drop of 2% formalin to microslides and covered with coverslips. The formalin preparation was infused with a 10% solution of glycerin in 96% ethyl alcohol and allowed to evaporate to glycerin over a period of several days; the resulting whole-mount preparation was sealed by Murrayite. The holotype, allotype, and one paratype were illustrated by aid of a camera lucida. One specimen was pressed tightly between the coverslip and slide in order to make certain critical observations. Zeiss differential interference-contrast optics (Nomarski) were used to make photomicrographs while whole-mount specimens were still in 2% formalin.

Measurements are given in micrometers (μm) and the resulting data are expressed in a terminology used by Kristensen & Renaud-Mornant (1983) and Kristensen & Higgins (1984). The holotype and allotype of the new family are deposited at the National Museum of Natural History, Smithsonian Institution (USNM), Washington, D.C. (U.S.A.). The squashed preparation and two other paratypes are deposited at the Zoological Museum of Copenhagen, Denmark, and bear the senior author's (RMK) specimen numbers only.

SYSTEMATIC ACCOUNT

Order Heterotardigrada Marcus, 1927
 Suborder Arthrotardigrada Marcus, 1927
 Renaudarctidae n. fam.

Diagnosis. Arthrotardigrades with segmental dorsal plates, both claws and toes present. Two to four accessory claw spines present on each foot. Primary and secondary clavae present along with complete set of cephalic cirri.

Type-genus: *Renaudarctus* n. gen.

Diagnosis. Renaudarctidae with segmental dorsal plates, 2 paired intersegmental plates, 4 dorsal intersegmental ridges, and 3 thin paired ventral plates. Cephalic sense organs complete but reduced; 3 pairs of lateral spines on trunk or leg near each trunk-leg junction of first three trunk segments. Terminal plate (pIV) with 2 pairs of lateral spikes, 1 pair of caudal spikes, and lateral margins continuous with furca-like caudal extensions. Leg 4 with small ovoid papilla. Cirrus E with cirrophore. Female and male with ovoid gonopore papillae connecting to terminal anal system. Female with latero-ventral projections containing seminal receptacle and accessory genital vesicle.

Etymology. This genus is formed from a combination of the surname of Jeanne Renaud-Mornant, and the Greek (Gr.) word *arctos*, "bear."

Type-species: *Renaudarctus psammocryptus* n. sp.
 (Figs. 1-16)

Diagnosis. Same as for genus.

Etymology. The name of this species is derived from *psammos* (Gr.), "sand" and *kryptos* (Gr.), "hidden."

Type-material. Holotypic female, 98 μm long, with two large eggs, gonopore, and seminal receptacle (USNM 81199), collected by R. M. Kristensen

and R. P. Higgins, 5 October 1982. Paratypic young female, 85 μm long, with small genital cells, gonopore, and seminal receptacle (USNM 81200), other data as for holotype. Paratypic female (squashed preparation), 101 μm long (RMK 820510.3; Zoological Museum, Copenhagen).

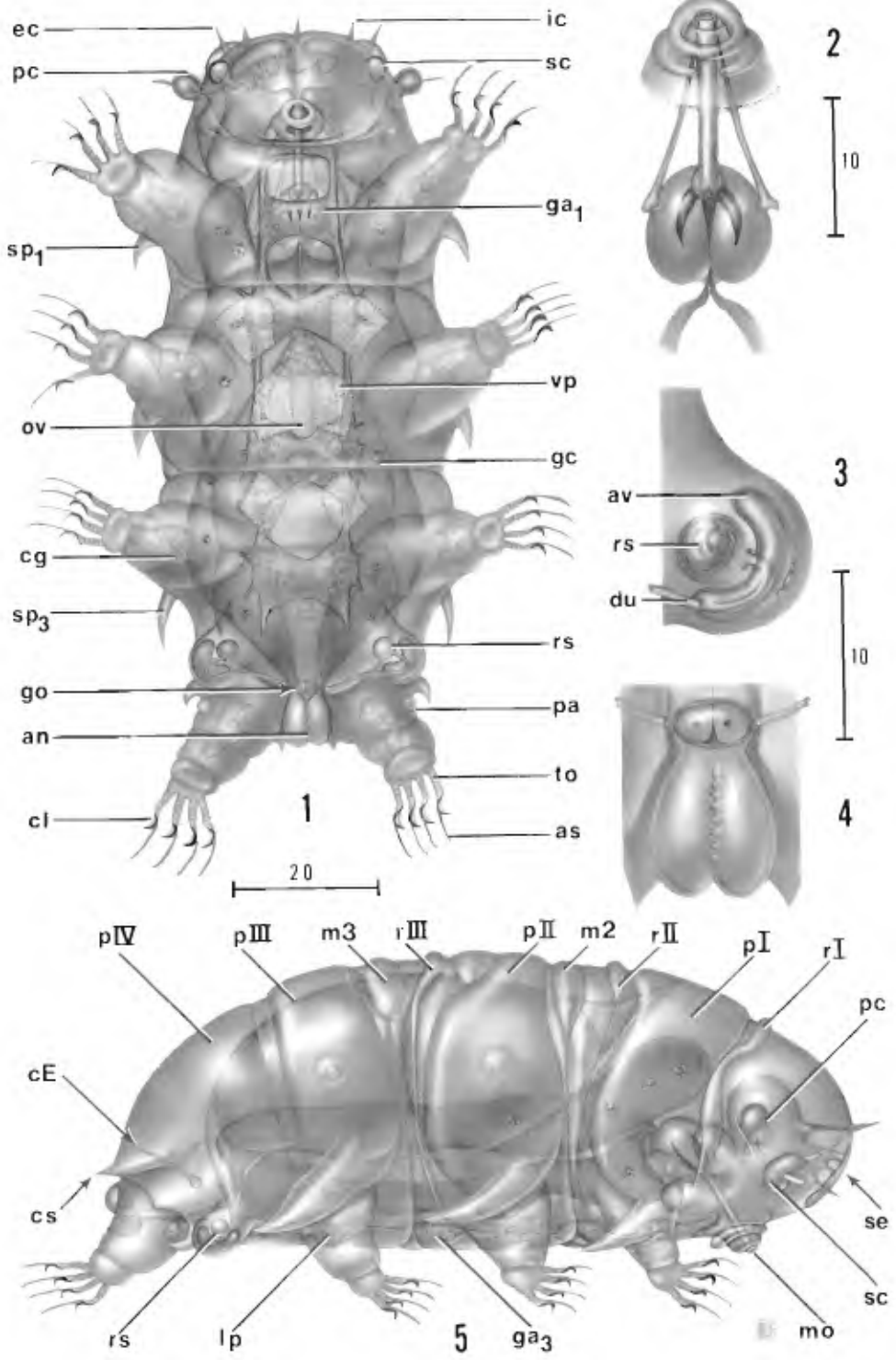
Type-locality. Pepper State Park, Fort Pierce, Florida (27°30.0'N, 80°17.0'W), found 70–100 cm deep in a coarse sand with shell hash taken 2 m above mean low-water level. Temperature, 23°–27°C. Salinity, 35–36‰.

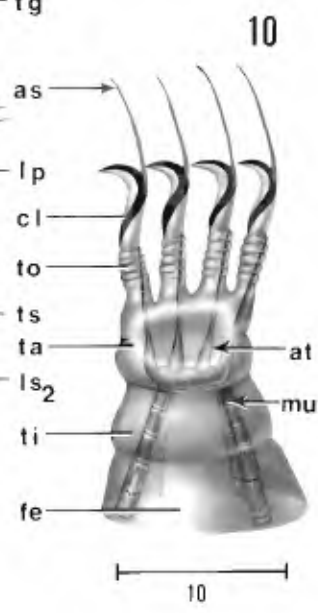
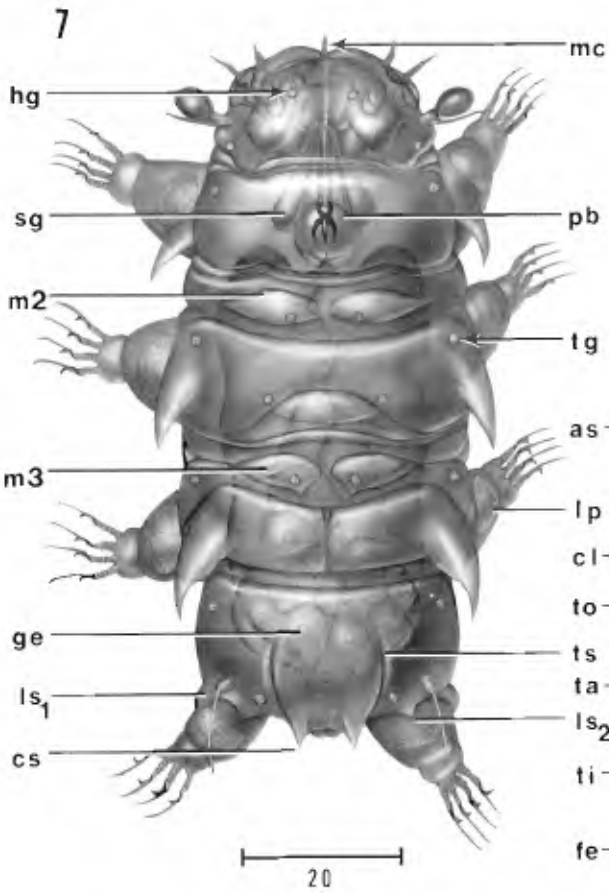
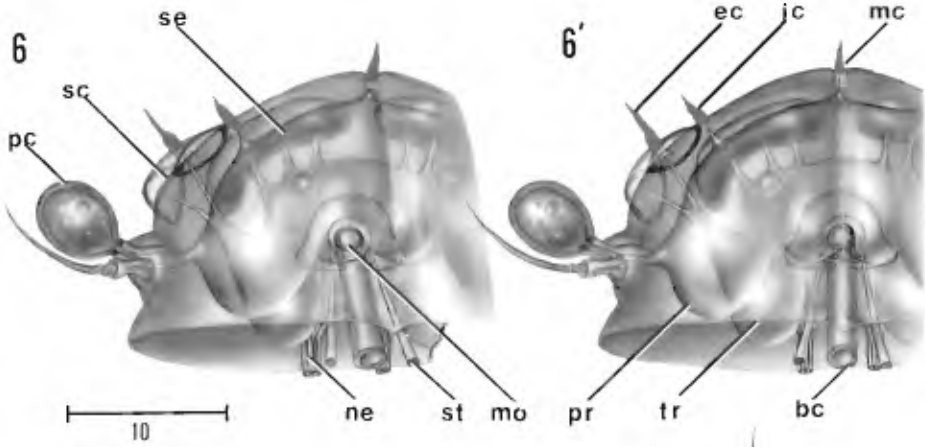
Additional paratypes. One mature female, 97 μm long (RMK 830904.3; Zoological Museum, Copenhagen), and 1 mature male, 87 μm long (RMK 830904.4; Zoological Museum, Copenhagen), from Red Reef Park, Boca Raton, Florida (26°21.0'N, 80°4.0'W), collected by R. M. Kristensen, 9 April 1983.

Description

External structures. The holotypic female (Fig. 5), 98 μm long, has a maximum width of 30 μm . The outer segmentation is distinct and can be observed best in lateral aspect. The dorsal cuticle consists of more or less sclerotized plates as in the family Echiniscidae of the suborder Echiniscoidea. The head segment is covered by the frontal plate and is separated from the trunk segment 1 by a thin cuticular fold or ridge (rI). Three more ridges separate the next three trunk segments (Fig. 11). The posterior margin of the frontal plate has a pair of rounded lobes dorsomesially. The first trunk plate or "scapular plate" is undivided and has prominent spinose, posteriorly curved flanges situated on leg 1, slightly below the trunk-leg junction and between the scapular plate and the undivided segmental plate 2 (pII). Two small median intersegmental plates (m2) are present. Segmental plate 2 has a pair of lateral spinose flanges homologous with those situated on the first leg. An indistinct single median plate, separated from the posterior edge of segmental plate 2, can be seen when animals are squashed. Two more intersegmental plates (m3) are present on each side of the dorsal midline between trunk segments 2 and 3. Paired segmental plates 3 are divided by a thin median suture, not always evident, and bear a pair of large lateral spinose flanges (sp₃). The ridge (rIV) between segments 3 and 4 is nearly plane and relatively broad. Segmental plate 4 or the "terminal plate" has two large indentations or "sutures" and

FIGS. 1–5. *Renaudarctus psammocryptus* n. gen., n. sp. Fig. 1. Paratypic female, ventral view. FIGS. 2–4. Detail from same animal after squashing. Fig. 2. Mouth opening and buccal apparatus. Fig. 3. Ventral projection containing seminal receptacle and accessory genital vesicle. Fig. 4. Female gonopore with two genital ducts and anus with two anal plates. Fig. 5. The holotypic female, lateral view. Scales in μm . an, anus; as, accessory hook of claw; av, accessory genital vesicle; cg, claw gland; cl, claw; cs, caudal spike; du, genital duct; ec, external cirrus; ga₁, ventral ganglion 1; ga₃, ventral ganglion 3; gc, gut crystal; go, gonopore; ic, internal cirrus; lp, leg plate; m2, paired median intersegmental plate 2; m3, paired median intersegmental plate 3; mo, mouth opening; ov, ovary; pI, segmental plate I; pII, segmental plate II; pIII, segmental plate III; pIV, segmental plate IV; pa, papilla of leg 4; pc, primary clava; rI, ridge I of dorsal cuticle; rII, ridge II of dorsal cuticle; rIII, ridge III of dorsal cuticle; rs, seminal receptacle; sc, secondary clava; se, frontal sense organ; sp₁, lateral flange (spine) 1; sp₃, lateral flange (spine) 3; to, toes; vp, ventral plate.





three pairs of spinous projections. The middorsal pair of caudal spikes (cs) is easy to see, but the second laterocaudal spikes (ls_2) are difficult to see because of their smaller size. Laterocaudal spikes 1 (ls_1) are hook-shaped. Ventral segmentation in the cuticle is not so distinct because it is on the dorsal surface; the segmental limits are best developed between trunk segments 1 and 2 and segments 2 and 3. Four small midventral plates (Fig. 1) are present on trunk segment 2 and a single pair is present on trunk segment 3. Legs have a small plate (lp) on the femur.

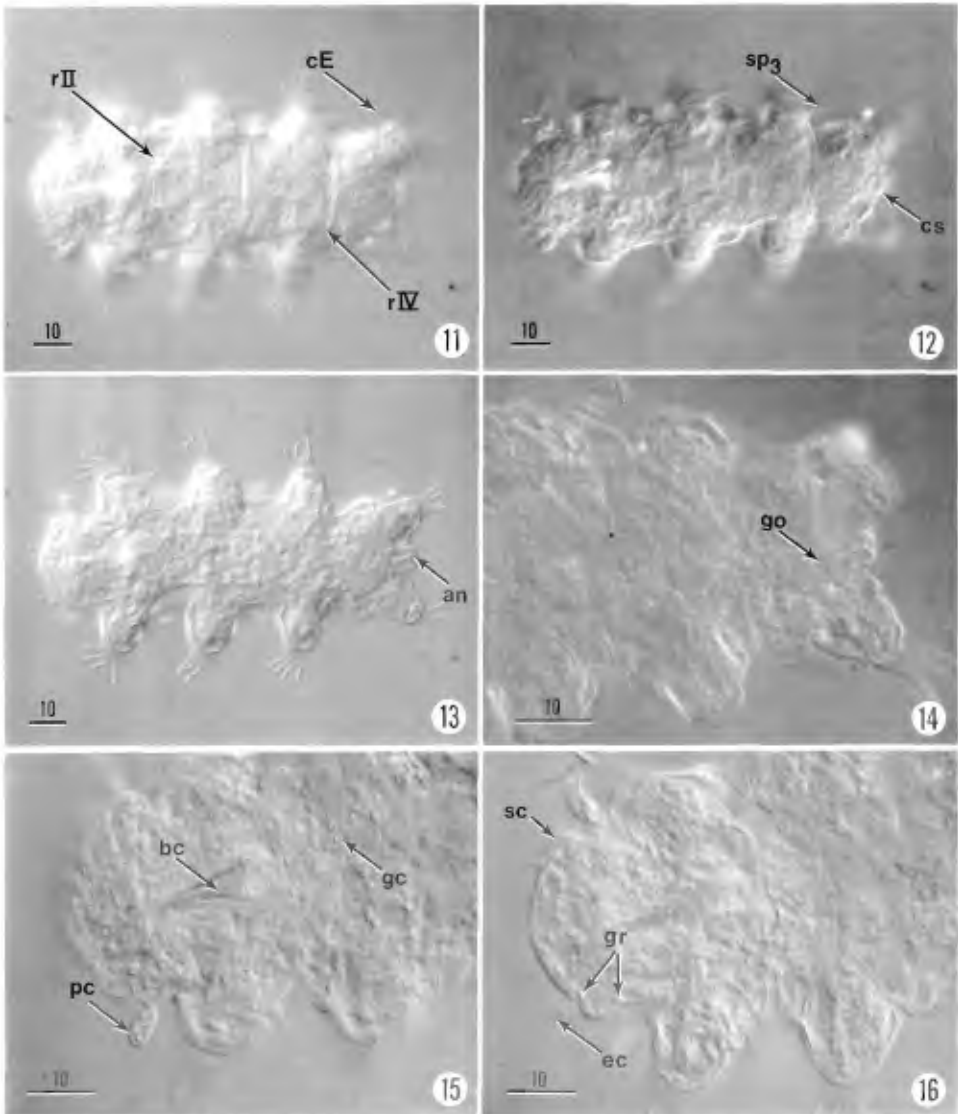
A complete set of small cephalic sense organs is present (Figs. 1, 5, 6). The holotype has ovoid primary clavae ($7\ \mu\text{m}$) and lateral cirri ($8\ \mu\text{m}$) inserted on a conic pedestal ($5\ \mu\text{m}$). The secondary clavae ($8\ \mu\text{m}$) are nearly sausage-shaped and surround the base of the external cirrus ($6\ \mu\text{m}$ long). Under interference phase-contrast optics, the secondary clavae appear dome-shaped in dorsal optical perspective, and as bulbous projections with two granulae in ventral optical perspective. The internal cirri ($5\ \mu\text{m}$) and the single median cirrus ($4\ \mu\text{m}$) are located on the anterior margin of the head. All cephalic cirri consist of 3 segments, the cirrophore, the scapus, and the flagellum; this segmentation is most obvious in the lateral cirri. Visible from ventral view are a pair of sensory plates which are innervated by nerves from the tritocerebrum; they probably are equivalent to what otherwise would be called tertiary clavae. A small round papilla ($3\ \mu\text{m}$) is present on leg 4 (Fig. 9). The only cirri on the trunk are those designated cirri E ($13\ \mu\text{m}$); each consists of a bell-shaped cirrophore, an accordion-like scapus, and a thin flagellum (Fig. 8).

Each of the stumpy legs consists of a coxa, femur, tibia, and tarsus of the echiniscid type. When the holotype was seen alive, only the tarsus was retracted telescopically within the tibia. The hand-shaped tarsus has four cuticular muscle-attachment fibers, each of which functions as a tendon. The four toes ("digits") of each leg are well developed, and when fully extended, they become as much as $5\ \mu\text{m}$ in length (Fig. 10). The toes appear to be shortened when the leg muscles are retracted. This may explain why 4–5 wrinkles may be visible on the toes. The claw ($4\ \mu\text{m}$) consists of a strongly cuticularized portion and a thin, more translucent laminar portion. All claws have a remarkably thin accessory spine, $5\text{--}7\ \mu\text{m}$ long.

Morphometric measurements are given in Table I.

Internal structure. The nervous system (Fig. 1) consists of a very large three-lobed brain and a small subpharyngeal ganglion, both located in the head, and

FIGS. 6–10. *Renaudarctus psammocryptus* n. gen., n. sp. Paratype young female. Fig. 6, 6'. Stereoscopic drawings of the head. Fig. 7. Dorsal view. Fig. 8. Cirrus E. Fig. 9. Leg 4 papilla. Fig. 10. Ventral view of right leg. Scales in μm . as, accessory hook of claw; at, muscle attachment to claw; bc, buccal canal; cl, claw; cs, caudal spike; ec, external cirrus; fe, femur; ge, genital cell; hg, head gland; ic, internal cirrus; lp, leg plate; ls_1 , laterocaudal spike 1; ls_2 , laterocaudal spike 2; m2, median intersegmental plate 2; m3, median intersegmental plate 3; mc, median cirrus; mo, mouth opening; mu, muscle; ne, ventral nerve; pb, pharyngeal bulb; pc, primary clava; pr, protocerebrum; sc, secondary clava; se, frontal sense organ; sg, stylet gland; st, stylet; ta, tarsus; tg, trunk gland; ti, tibia; to, toe; tr, tritocerebrum; ts, terminal extension.



FIGS. 11-16. *Renaudarctus psammocryptus* n. gen., n. sp. (Interference-contrast micrographs.) FIGS. 11-13. Paratyptic female before squashing. Fig. 11. Optical view, dorsal level. Fig. 12. Optical view, middle level. Fig. 13. Optical view, ventral level. FIGS. 14-16. Paratyptic young female. Fig. 14. Ventral view of caudal region, showing gonopore. Fig. 15. Optical view, middle level of anterior part of animal. Fig. 16. Optical view, ventral level of anterior part of animal. Scales in μm . an, anus; bc, buccal canal; cE, cirrus E; cs, caudal spike; ec, external cirrus; gc, gut crystal; go, gonopore; gr, granula in secondary clava; pc, primary clava; rII, ridge II of dorsal cuticle; rIV, ridge IV of dorsal cuticle; sc, secondary clava; sp₃, third lateral flange (spine).

TABLE I

Comparative features of the five specimens of *Renaudarctus psammocryptus* (measurements in μm)

Characteristic	Pepper State Park			Red Reef Park	
Body length	98	101	85	97	87
Width between legs III-IV	30	35	30	32	30
Buccal canal	15	13	14	15	12
Pharynx bulb	8	10	10	10	9
Placoid	7	6	6	8	6
Stylet	18	15	17	18	15
Median cirrus	4	3	4	4	3
Internal cirri	5	4	4	4	5
External cirri	6	6.5	6.5	7	6
Lateral cirri	8	7	7	8	8
Primary clavae	7	6	6	7	8
Secondary clavae	8	6	6	8	8
Cirri E	13	16	14	15	13
Leg 4 papillae	3	3	3	3	3
Lateral flanges 3	10	12	11	10	10
Lateral flanges 2	9	11	9	8	7
Lateral flanges 1	8	7	6	7	7
Caudal spikes	4	5	6	4	4
Laterocaudal spikes 2	2	3	3	2	2
Laterocaudal spikes 1	4	3	4	3	3
Sex	♀ ^a	♀	♀ ^b	♀	♂

^a Holotype.^b Immature female.

a large ventral ganglion in each of the four trunk segments. The protocerebrum, the large dorsal lobe, innervates the lateral cirri and the primary clavae. The deutocerebrum, the small dorsal lobe, innervates the internal cirri and perhaps also the median cirrus. The tritocerebrum (lateral lobe) innervates the secondary clavae, external cirri, and the two ventroposterior sensory plates.

Two pairs of refractive unicellular glands are present in the head and on each trunk segment. Only in one paratype, a young female (Fig. 7), were they easily seen. Each gland has several thin, radiating cellular strands giving the appearance of sun-rays.

The reproductive system (Fig. 5) of the holotypic female consists of two large ova in a single ovary. The gonopore complex consists of a simple oval papilla anteriorly adjacent to the anus; all other heterotardigrade females have six rosette cells surrounding the gonopore. Heterotardigrade males, however, have almost the same kind of gonopore complex as that present in the females of *Renaudarctus psammocryptus*. The female gonopore is divided internally by a septum; a small gland empties into the resulting two portions of the genital opening. Two seminal receptacles are present, one situated in a lateral projection on each side of the body. In squashed preparations, sperm was seen inside the seminal receptacle and genital duct. Each genital duct communicates with

the gonopore and, for a short distance from the seminal receptacle, is surrounded by a portion of a sausage-shaped accessory vesicle (Fig. 3) prior to its junction with the seminal receptacle. Young females (Figs. 7, 14) without well-developed gonads already have a gonopore, genital ducts, and seminal receptacles, an unusual occurrence in tardigrades.

The mouth opening is subterminal on a small cone surrounded by a sense organ. The buccal canal is narrow ($1\ \mu\text{m}$) and short ($15\ \mu\text{m}$). The stylets are thin with large furcae (Fig. 2); no stylet supports are present. The pharyngeal bulb is small, only $8\ \mu\text{m}$ in diameter, with three very robust but small placoids. A short esophagus leads to the three-lobed gut, which contains a white amorphous material and many gut crystals (Figs. 5, 15). The anal complex is located posteriorly so that a portion protrudes from the animal (Fig. 13) and can be seen in a dorsal view. The anus is a zig-zag slit surrounded by two bulbous anal plates.

No secondary sexual dimorphism was seen; the male has the same kind of gonopore as the female.

Phylogenetic Relationships in Heterotardigrada (Fig. 17)

The marine Arthrotardigrada are considered the most ancestral suborder within the phylum Tardigrada (see Kristensen, 1976; Renaud-Mornant, 1982). Until recently, the marine family Halechiniscidae Thulin, 1928 (not Puglia, 1959) was considered to exhibit the greatest number of plesiomorphic characters (Greven, 1980; Ramazzotti, 1962, 1972), but Renaud-Mornant (1982, p. 161) postulated that *Parastygartus* (family Stygarctidae Schulz, 1951) should be considered as the ancestral tardigrade.

The presence of toes with claws, in addition to the presence of cuticularized dorsal plates, suggests that the Renaudarctidae are intermediate between the Stygarctidae and the Halechiniscidae. The Stygarctidae exhibit well-developed, elongate claws, often with 2–4 filiform accessory hooks, directly inserted on the foot. The insertion of claws directly on the foot is a plesiomorphic condition. The filiform accessory hooks also are found in *Renaudarctus* and, although not so elongate, they appear to be present on the mesial claws of *Neostygartus* (see Grimaldi de Zio et al., 1982a, fig. 1). The presence of filiform accessory hooks, whether plesiomorphic or apomorphic, is a character shared only among the Stygarctidae and Renaudarctidae. A second plesiomorphic character shared by both families is that of the cuticularized dorsal plates. The Halechiniscidae lack these plates but have claws inserted on well-developed toes, an apomorphic condition. The new family, Renaudarctidae, shares with the Stygarctidae the plesiomorphic condition of cuticularized dorsal plates, but the claws, as in the Halechiniscidae, are inserted on well-developed toes.

The toes are developed secondarily from a stygarctid-like foot with elongate claws. The toes lose their claws in the Orzeliscidae and each elongated toe is modified as a sucking organ. In the Batillipedidae, only the distal end of each toe is a sucking disc; the proximal portion becomes a narrow supporting structure.

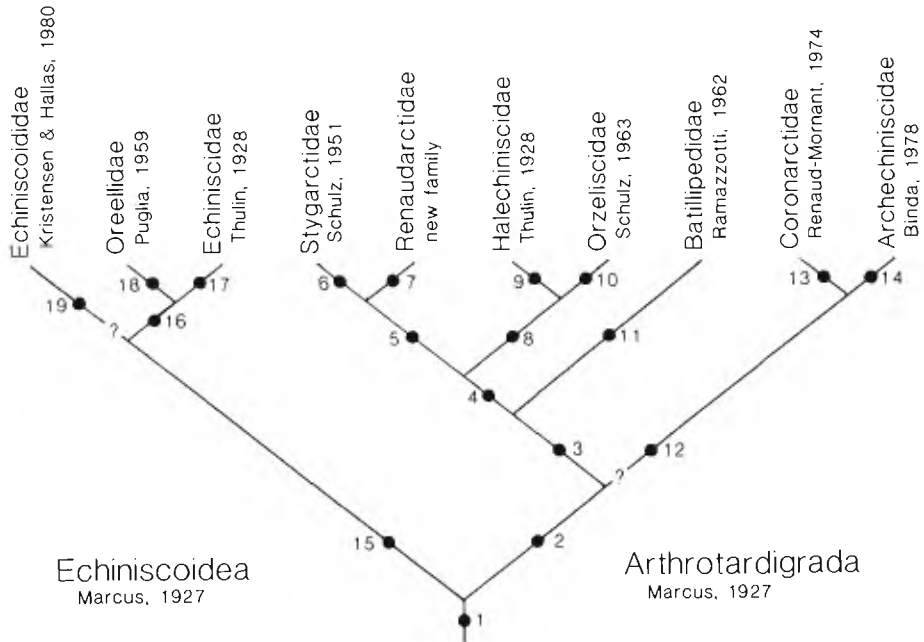


FIG. 17. Cladogram of Heterotardigrada. Numbers refer to apomorphic characters in text.

McKirby et al. (1976) noted that the stygarctid genus *Megastygarcoides* has short pedestals from which each claw originates. This is the first suggestion of toe development. *Neostygarcus*, like *Renaudarctus*, has less pronounced toes than those found in the Halechiniscidae. Therefore, the Renaudarctidae provide an important link in an evolutionary sequence: *Parastygarcus*—*Stygarcus*—*Mesostygarcoides*—*Neostygarcus*—*Renaudarctus*—*Halechiniscus*—*Orzeliscus*—*Batillipes*.

Considering the development of toes and adhesive discs, the Halechiniscidae, Orzeliscidae, and Batillipedidae must be considered apomorphic families within the Arthrotardigrada. The lack of dorsal, as well as ventral cuticular plates, also is an apomorphic feature. Therefore, the family Stygarctidae, together with its sister group Renaudarctidae, can be recognized as the most plesiomorphic group.

Renaudarctus and *Neostygarcus* are considered to be members of the same family, the Renaudarctidae, because they are the only two genera that have both toes on the feet and cuticular dorsal plates. Furthermore, both have dome-shaped secondary clavae. However, in other characters, such as cirrus E and the shape of the cephalic plate, *Neostygarcus* also is similar to Stygarctidae as Grimaldi de Zio et al. (1982a) already have indicated. Among the arthrotardigrades, *Neostygarcus* is unique in possessing middorsal spines. Within the Heterotardigrada, only the genus *Hypechiniscus* Thulin, 1928 (suborder Echiniscoidea) has one or two middorsal spines. We believe that the presence

of middorsal spines, as in the case of the median cirrus on the head, is a plesiomorphic character, and the ancestral heterotardigrade had these characters, as well as dorsal and ventral plates. A lack of the median cirrus in the suborder Echiniscoidea is almost the only character distinguishing them from members of the suborder Arthrotardigrada. Renaud-Mornant (1975) and Kristensen & Hallas (1980) already have indicated that this character should be used carefully because the median cirrus can be reduced or secondarily lacking, as in the family Archechiniscidae Binda, 1978, which was formerly placed in the suborder Echiniscoidea (see Schulz, 1963). The same kind of character reduction occurs in the genus *Angursa* within the subfamily Styraconyxinae, family Halechiniscidae (see Kristensen & Renaud-Mornant, 1983). One species, *Angursa bicuspis* Pollock, 1979 lacks or nearly lacks the median cirrus, but the second member of this genus, *A. lanceolata* Renaud-Mornant, 1981b, has a distinct median cirrus. According to available descriptions, only two other arthrotardigrades, *Tanarctus tauricus* Renaud-Debyser, 1959 and *Halechiniscus gutteli* Richters, 1908 lack the median cirrus.

Renaudarctus psammocryptus has a distinct median cirrus and feet with toes. Thus, by definition, the species is assigned to the Arthrotardigrada. Until now, however, the presence of both ventral and dorsal plates has been seen only in members of the suborder Echiniscoidea, where the ventral plates are best developed in the semiterrestrial species *Echiniscus spitzbergensis*. Dastych (1973) has studied the variation of these plates in different populations of *Echiniscus spitzbergensis*. They are always strongly sculptured, as are the dorsal plates. Thin ventral plates are reported in at least one species of *Bryodelphax*; i.e., *B. sinensis* Pilato, 1974. The presence of ventral plates in *Renaudarctus* must be considered a plesiomorphic character shared with members of the suborder Echiniscoidea, but the pattern of the dorsal plates of *Renaudarctus* and *Bryodelphax* (family Echiniscidae) are nearly the same. Thus, in this respect, *Renaudarctus* is intermediate between marine families and the semiterrestrial family Echiniscidae (Fig. 17).

The cephalic sense organs of *Renaudarctus* are the most complex among tardigrades. Both primary and secondary clavae are present, but in addition, there are two large flat sacs on the frontal part of the head, which may be the equivalent of a third pair of clavae (tertiary clavae). Also present is another sense organ that nearly surrounds the mouth cone and resembles the secondary clavae of the genus *Halechiniscus*.

The only comment we can make on the abundance of sense organs in the head of *Renaudarctus* is that it is possible that the ancestral tardigrade had three pairs of clavae corresponding to the three pairs of cephalic cirri, and, in turn, three trunk segments became fused with the original head segment.

Evidence that *Renaudarctus* belongs to an ancestral group of Heterotardigrada is indicated also by the genital organs. Both the female and male gonopores are oval and not separated from the anal complex. All other females in Heterotardigrada have a gonopore complex consisting of six or seven rosette cells, and males have an oval gonopore papilla. When Pollock (1970b) published his account of the reproductive anatomy of tardigrades, it was not known that the females of *Orzeliscus*, *Styraconyx*, and *Oreella* also have the rosette

cells surrounding the gonopore. *Renaudarctus* is the first heterotardigrade with a female gonopore of the male type. In exhibiting the combination of a seminal receptacle associated with an accessory vesicle, and genital ducts from the two seminal receptacles opening near the gonopore-anus complex, *Renaudarctus* nearly has a cloaca as found in all Eutardigrada.

We conclude, therefore, that because *Renaudarctus* shares such a strange combination of apomorphic and plesiomorphic characters with *Neostygarctus*, a combination not demonstrated by other genera of Arthrotardigrada, the erection of a new family is justified. We include *Neostygarctus* in the Renaudarctidae because of its claw with a thin membrane, toes, dorsal plates, and dome-shaped secondary clavae. New species of *Neostygarctus* perhaps will require that this genus be placed in its own family.

The different evolutionary lines that we postulate for the order Heterotardigrada Marcus, 1927 are shown in Fig. 17. The numbers on the various branches of the cladogram indicate the apomorphic characters for each branch beginning with "1," which represents the autapomorphic characters of the Heterotardigrada pertaining to the Mesotardigrada Rahm, 1937 and Eutardigrada Marcus, 1927, the two remaining orders of Tardigrada. Relevant to the families in Heterotardigrada, however, this same number "1" represents the most plesiomorphic characters. Other numbers (2-18) relate to the apomorphic characters for the different branches of the cladogram, the highest number representing the most apomorphic. The numbered character states are as follows:

1. Segmental plates both ventral and dorsal. Cephalic cirri, clavae, and body or leg spines present. Seminal receptacles opening separately, on either side of female rosette-shaped gonopore. Stylet support present. Claws inserting directly on legs.
2. Median cephalic cirrus, three pairs of lateral cephalic cirri, two or three pairs of clavae. Cirrus E with cirrophore.
3. Cephalic cirri long, consisting of two or three segments.
4. Two to four claws or modified rod-like appendages, primary and secondary clavae not similar in shape.
5. Stylet support absent.
6. Toes absent. Dorsal segmental plates sometimes with lateral flanges or spine-shaped appendages. Pharynx bulb with internal stylet support.
7. Four claws inserting via toes on foot. Thin ventral plates sometimes present.
8. Cuticular plates absent. Toes always present.
9. Two to four toes with terminal claws. Secondary clavae variously reduced.
10. Four toes of similar size each with elongate suction discs. Seminal receptacles in each of two lateroventral projections. Secondary clavae flattened on ventral surface.
11. Six toes each with suction disc (= "adhesive organs"). Claws absent. Secondary clavae dome-shaped. Seminal receptacle single or absent.
12. Dorsal segmental plates absent. Leg spines of first three legs absent.

- Cephalic cirri short and reduced. Secondary clavae flat, not papillate. Stylet supports absent. Cirrus E without cirrophore.
13. Four long spurred claws connected to foot by membrane. Secondary clavae forming corona around cephalic cirri.
 14. Median cephalic cirrus absent or vestigial. Two median toes with claws, two lateral claws inserting directly on foot.
 15. Median cephalic cirrus absent. Leg papillae reduced or absent. Cirrus E absent or without cirrophore. Toes absent.
 16. Four claws on each leg (*Carphania* Binda, 1978 not considered member of Oreellidae). Only legs 1 and 4 with sense organs.
 17. Variably sclerotized dorsal plate. Ventral plate sometimes present. Both secondary and primary clavae papillate or dome-shaped. Body appendages present in many species, but never in form of cirri with cirrophores. Seminal receptacles absent.
 18. Dorsal plates absent or indistinct. Long primary clavae. Short secondary clava. Seminal receptacles opening directly without genital ducts. Cirrus E absent.
 19. Dorsal plates absent. "Eutardigrade" (simple) cuticle. Three to 13 claws on each leg. Leg 4 sometimes with one claw less than on legs 1-3. Leg spine reduced or absent. Stylet support lacking. Cirrus E present.

Two question marks in Fig. 17 indicate that the systematic position of the three families Coronarctidae Renaud-Mornant, 1974; Archechiniscidae Binda, 1978; and Echiniscoididae Kristensen & Hallas, 1980 is problematic; however, it is highly likely that the Echiniscoididae and Archeschiniscidae are related. The Coronarctidae also share some apomorphic characters with Echiniscoididae. Future new species of marine tardigrades perhaps will require that these three families be included in a separate suborder.

Ecological Remarks

Renaudarctus psammocryptus was a rare tardigrade in both sample sites. Of 24 samples containing several thousand tardigrades, only three contained representatives of this new species. The five specimens of *R. psammocryptus* were found from low- to mid-tide regions of the beaches, both from strata 25-100 cm deep. There was no clear vertical or horizontal distribution of any of the seven species of tardigrades encountered: *Parastygarctus sterreri* Renaud-Mornant, 1970; *Stygarctus goubaultae* Renaud-Mornant, 1981a; *Halechiniscus remanei* Schulz, 1955; *Raiarctus colurus* Renaud-Mornant, 1981c; *Raiarctus* sp., *Tanarctus tauricus*, Renaud-Debyser, 1959; and *Batillipes bullacaudatus* McGinty & Higgins, 1968. *Raiarctus colurus*, *Raiarctus* sp., and *Tanarctus tauricus* were found together with *Renaudarctus psammocryptus* in shell hash from the same hole. *Stygarctus goubaultae*, the most abundant species, was found from low- to high-tide regions, but only in coarse quartz mixed with small amounts of coralline sand. At the high-tide region, *S. goubaultae* was found in strata as deep as 180 cm, a record in sediment depth for

a tardigrade. *Batillipes bullacaudatus*, the second most abundant species, was found at or near the surface in the low-tide region, but was more abundant in the 10–30-cm strata at the mid-tide region.

Halechiniscus remanei was common only in the mid-tide region, in quartz sand, at 30–50 cm depth. In subtropical, high-energy, tidal beaches such as Pepper State Park Beach and Red Reef Park Beach, the salinity and temperature are relatively constant; however, especially in the winter, the entire beach can be changed by storm waves as was the case during our investigation. The shell hash horizon was found only at the low-tide region 5 October 1982. Subsequently, a severe storm rearranged the granulometric configuration and the shell hash was found slightly above the mid-tide region, overlain by a layer of quartz sand 100 cm deep. The tardigrade fauna in this deep horizon of shell hash had a very high concentration of *Raiarctus* sp., and a few *Parastygarctus sterreri*, *Tanarctus tauricus*, and *Renaudarctus psammocryptus*. The same kind of shell hash is common offshore; the tardigrade fauna of such a substrate in the beach ecosystem perhaps indicates that this habitat may be merely an extension of the subtidal shell hash habitat which extends at least 10 km offshore to depths of at least 20 m.

Most ecological studies of intertidal tardigrades (Crisp & Kristensen, 1984; Lindgren, 1971; Pollock, 1970c; Renaud-Debyser, 1963; de Zio, 1965) have considered water circulation, salinity, and temperature as the principal factors determining the distribution of tardigrades. Furthermore, these investigations have noted a significant variation in the vertical and, to a lesser extent, horizontal distribution of different species. Our results indicate that vertical and horizontal distribution of the eight species of tardigrades was clearly related to the physical nature of substrate; i.e., granulometry, more than to other apparent physical and/or chemical factors. Depth of a given stratum of sediment in the beach appeared to be insignificant, and three of the eight species were found 10 km offshore in the same shell hash sediment as sampled in our two intertidal stations. Recently, Rodrigues et al. (1982) came to the same conclusion in their study of subtidal meiobenthos from the northeastern Bay of Bengal; meiofaunal abundance was related more closely to the texture of the sediment than to depth.

Zoogeographically, we find it very interesting to note that only *B. bullacaudatus* and *H. remanei* have been reported previously from the United States. *Stygarctus goubaultae* and *Raiarctus colurus* were described from the Caribbean island of Guadeloupe (Renaud-Mornant, 1981a,c). *Parastygarctus sterreri*, which was described from the Mediterranean Sea (Renaud-Mornant, 1970), also has been found in Bermuda (Renaud-Mornant, 1971) and the Galapagos (McKirdy et al., 1976). The intertidal tardigrade fauna from the two beaches investigated clearly differs from that of more northern localities such as Woods Hole, Massachusetts (Pollock, 1970c) and North Carolina (Lindgren, 1971). The influence of the Gulf Stream could be a contributing factor, but more probably, it is the coarse calcium carbonate sediment that offers a significantly different kind of habitat from that occurring in more northern climes.

LITERATURE CITED

- BINDA, M. G. 1978. Ristestemazione di alcuni Tardigradi con l'istituzione di un nuovo genere di Oreellidae e della nuova famiglia Archechiniscidae. *Animalia*, 5(1/3): 307-314.
- CRISP, M. & KRISTENSEN, R. M. 1984. A new marine interstitial eutardigrade from East Greenland, with comments on habitat and biology. *Vid. Medd. Dansk. Naturh. Foren.*, 143. (in press)
- DASTYCH, H. 1973. Systematic studies on Tardigrada. II. *Echiniscus clavisetosus* Mihelčić 1958, a synonym of *Echiniscus spitzbergensis* Scourfield 1897. *Bull. Soc. Amis Sci. Lett. Poznan*, 14: 89-99.
- DUJARDIN, F. 1851. Sur les Tardigrades et sur une espèce a longs pieds vivant dans l'eau de mer. *Ann. Sci. Nat. Zool.*, Ser. 3, 15: 160-166.
- GREVEN, H. 1980. Die Bärtierchen. *Die Neue Brehm Bücherei*, 537: 1-101.
- GRIMALDI DE ZIO, S., D'ADDABBO GALLO, M. & MORONE DE LUCIA, R. M. 1982a. *Neostygarcus acanthophorus* n. gen. n. sp., nuovo Tardigrado marino del Mediterraneo. *Cah. Biol. Mar.*, 23: 319-323.
- GRIMALDI DE ZIO, S., D'ADDABBO GALLO, M., MORONE DE LUCIA, R. M., VACCARELLA, R. & GRIMALDI, P. 1982b. Quattro nuove specie di Halechiniscidae rinvenute in due grotte sotto marine dell'Italia meridionale (Tardigrada: Heterotardigrada). *Cah. Biol. Mar.*, 23: 415-426.
- HALLAS, T. E. & KRISTENSEN, R. M. 1982. Two new species of the tidal genus *Echiniscoides* from Rhode Island, U.S.A. (Echiniscoididae, Heterotardigrada). In Nelson, D., ed., *Proc. Third Int. Symp. Tardigrada, Johnson City, Tennessee, U.S.A. 1980*, East Tenn. Univ. Press, pp. 179-192.
- KRISTENSEN, R. M. 1976. On the fine structure of *Batillipes noerrevangi* Kristensen, 1976. I. Tegument and moulting cycle. *Zool. Anz.*, 197: 129-150.
- KRISTENSEN, R. M. & HALLAS, T. E. 1980. The tidal genus *Echiniscoides* and its variability, with erection of Echiniscoididae fam. n. *Zool. Scripta*, 9: 113-127.
- KRISTENSEN, R. M. & HIGGINS, R. P. 1984. Revision of *Styraconyx* (Tardigrada: Halechiniscidae) with description of two new species from Disko Bay, West Greenland. *Smithson. Contrib. Zool.* (in press)
- KRISTENSEN, R. M. & RENAUD-MORNANT, J. 1983. Existence d'Arthrotardigrada semi-benthiques de genres nouveaux de la sous-famille des Styraconyxinae subfam. nov. *Cah. Biol. Mar.*, 24: 337-353.
- LINDGREN, E. W. 1971. Psammolittoral marine tardigrades from North Carolina and their conformity to worldwide zonation patterns. *Cah. Biol. Mar.*, 12: 481-496.
- MARCUS, E. 1927. Zur Anatomie und Ökologie mariner Tardigraden. *Zool. Jahrb. (Abt. Syst.)*, 53: 487-588.
1929. Tardigrada. In Bronn, H. G., ed., *Klassen und Ordnungen des Tierreichs*, 5 (sec. 4, book 3): 1-608.
- MCGINTY, M. M. & HIGGINS, R. P. 1968. Ontogenetic variation of taxonomic characters of two marine tardigrades with the description of *Batillipes bullacaudatus* n. sp. *Trans. Am. Microsc. Soc.*, 87: 252-262.
- MCKIRDY, D. J. 1975. *Batillipes* (Heterotardigrada): comparison of six species from Florida (USA) and a discussion of taxonomic characters within the genus. In Higgins, R. P., ed., *Int. Symp. Tardigrades, Pallanza, Italy, June 17-19, 1974*. *Mem. Ist. Ital. Idrobiol.*, 32 Suppl.: 177-223.
- MCKIRDY, D. J., SCHMIDT, P. & MCGINTY-BAYLY, M. 1976. Interstitielle Fauna von Galapagos. XVI. Tardigrada. *Microf. Meeresb.*, 58: 409-449.
- PILATO, G. 1974. Tre nuove specie di Tardigradi muscicoli di Cina. *Animalia*, 1(1/3): 59-68.
- POLLOCK, L. W. 1970a. *Batillipes dicrocercus* n. sp., *Stygarcus granulatus* n. sp. and other Tardigrada from Woods Hole, Massachusetts, U.S.A. *Trans. Am. Microsc. Soc.*, 89: 38-52.
- 1970b. Reproductive anatomy of some marine Heterotardigrada. *Trans. Am. Microsc. Soc.*, 89: 308-316.
- 1970c. Distribution and dynamics of interstitial Tardigrada at Woods Hole, Massachusetts, U.S.A. *Ophelia*, 7: 145-165.

1979. *Angursa bicuspis* n. g., n. sp., a marine arthrotardigrade from the western North Atlantic. *Trans. Am. Microsc. Soc.*, 98: 558-562.
- PUGLIA, C. R. 1959. Some aspects of the taxonomy, ecology and distribution of the tardigrades with emphasis on the tardigrades of east central Illinois. Unpublished Ph.D. Thesis, Univ. Illinois, Urbana (Micro. N. 60-228). 125 pp.
- RAHM, G. 1937. Eine neue Tardigraden-Ordnung aus den heissen Quellen von Unzen, Insel Kyushu, Japan. *Zool. Anz.*, 120: 65-71.
- RAMAZZOTTI, G. 1962. Il Phylum Tardigrada. *Mem. Ist. Ital. Idrobiol.*, 14: 1-595.
1972. Il Phylum Tardigrada, 2nd ed. *Mem. Ist. Ital. Idrobiol.*, 28: 1-732.
- RENAUD-DEBYSER, J. 1959. Études sur la faune interstitielle des îles Bahamas. III. Tardigrades. *Vie Milieu*, 10: 297-302.
1963. Recherches écologiques sur la faune interstitielle des sables. Bassin d'Arcachon, île de Birmini, Bahamas. *Vie Milieu*, suppl. 15: 1-157.
- RENAUD-MORNANT, J. 1970. *Parastygarctus sterreri* n. sp., Tardigrade marin nouveau de l'Atlantique. *Cah. Biol. Mar.*, 11: 355-360.
1971. Tardigrades marins des Bermudes. *Bull. Mus. Natl. Hist. Nat. Paris*, 2nd series, 42: 1268-1276.
1974. Une nouvelle famille de Tardigrades marins abyssaux: les Coronarctidae fam. nov. (Heterotardigrada). *C. R. Acad. Sci. Paris*, 278: 3087-3090.
1975. Deep-sea Tardigrada from the "Meteor" Indian Ocean Expedition. "Meteor" *Forschungsergeb.*, ser. D, 21: 54-61.
- 1981a. *Stygarctus goubaultae* n. sp. un nouveau Tardigrade marin (Arthrotardigrada) de la Guadeloupe. *Bull. Mus. Natl. Hist. Nat. Paris*, 4th ser. 3 (section A, no. 1): 175-180.
- 1981b. Deux nouveaux *Angursa* Pollock, 1979, du domaine abyssal (Tardigrada, Arthrotardigrada). *Tethys*, 10: 161-164.
- 1981c. *Ratarctus colurus* n. g., n. sp., et *R. aureolatus* n. sp., Tardigrades (Arthrotardigrada) marins nouveaux de sédiments calcaires. *Bull. Mus. Natl. Hist. Nat. Paris*, 4th series, 3 (section A, no. 2): 515-522.
1982. Species diversity in marine Tardigrada. In Nelson, D., ed., *Proc. Third Int. Symp. Tardigrada*, Johnson City, Tennessee, U.S.A. 1980, East Tenn. State Univ. Press, pp. 149-178.
- RICHTERS, F. 1908. Marine Tardigraden. *Zool. Anz.*, 33: 77-85.
- RODRIGUES, C. L., HARKANTRA, S. N. & PARUSEKAR, A. H. 1982. Sublittoral meiobenthos of the Northeastern Bay of Bengal (India). *Indian J. Mar. Sci.*, 11: 239-242.
- SCHULZ, E. 1935. *Actinarctus doryphorus* nov. gen. nov. spec., ein merkwürdiger Tardigrad aus der Nordsee. *Zool. Anz.*, 111: 285-288.
1951. Über *Stygarctus bradypus* n. g. n. sp., einen Tardigraden aus dem Küstengrundwasser und seine phylogenetische Bedeutung. *Kiel. Meeresforsch.*, 8: 86-97.
1955. Studien an marinen Tardigraden. *Kiel. Meeresforsch.*, 11: 74-79.
1963. Über die Tardigraden. *Zool. Anz.*, 171: 3-12.
- THULIN, G. 1928. Über die Phylogenie und das System der Tardigraden. *Hereditas*, 11: 207-266.
- ZIO, S. DE. 1965. Distribuzione del mesopsammon in rapporto alla distanza della linea di riva e alla distanza da un corso di acqua salmastra. *Boll. Zool.*, 32: 525-537.

10

11

12

13