First record of the enigmatic genus *Redudasys* Kisielewski, 1987 (Gastrotricha: Macrodasyida) from the Northern hemisphere

Tobias KÅNNEBY
Department of Zoology, Swedish Museum of Natural History,
PO Box 50007, SE-104 05 Stockholm (Sweden)
tobias.kanneby@nrm.se

Mary K. WICKSTEN
Department of Biology, Texas A & M University,
TX 77843-3258 College Station (USA)
wicksten@bio.tamu.edu

Kånneby T. & Wicksten M. K. 2014. — First record of the enigmatic genus *Redudasys* Kisielewski, 1987 (Gastrotricha: Macrodasyida) from the Northern hemisphere. *Zoosystema* 36 (4): 723-734.
http://dx.doi.org/10.5252/z2014n4a2

ABSTRACT

Gastrotricha Metschnikoff, 1865 is a small phylum of acoelomatic animals common in both marine and freshwater environments. To date, close to 800 species are known from all over the world. The order Macrodasyida Remane, 1925 [Rao & Clausen, 1970] is almost exclusively marine except for the rare genera *Marinellina* Ruttner-Kolisko, 1955 and *Redudasys* Kisielewski, 1987. In this study, we present morphological and molecular data for a potentially new species of *Redudasys* from the Edwards Aquifer, Texas, USA. It is the first record of the genus from the Northern hemisphere. It has only previously been reported from Brazil. The Texan specimen, designated as *Redudasys* sp., has a single pair of anterior adhesive tubes and is considerably shorter than the specimens reported from Brazil. Molecular data from the 18S rRNA and COI mtDNA genes support a close relationship of *Redudasys* sp. to *Redudasys fornerise* Kisielewski, 1987. Barcodes are provided for both *Redudasys* sp. and *R. fornerise*. The limited material obtained does not justify the description of a new species, but the record is certainly important from a biogeographical standpoint. The colonization and invasion of marine species into freshwater habitats, in particular those of the Edwards Aquifer, is discussed. Although certain marine gastrotrichs appear to have an astonishing ability to adapt to changes in salinity *Redudasys* is likely to be a marine relic.

KEY WORDS
Redudasyidae,
biogeography,
artesian outflow,
Edwards Aquifer,
Texas,
new record.

RÉSUMÉ

Première signalisation du genre énigmatique Redudasys *Kisielewski, 1987 (Gastrotricha: Macrodasyida) dans l'hémisphère nord.*

Les Gastrotriches Metschnikoff, 1865 forment un petit phylum d'animaux acoelomates, communs à la fois dans l'environnement marin et en eau douce. À ce jour, près de 800 espèces sont connues à travers le monde. L'ordre Macrodasyida Remane, 1925 [Rao & Clausen, 1970] est presque exclusivement marin, sauf les genres *Marinellina* Ruttner-Kolisko, 1955 et *Redudasys* Kisielewski, 1987 qui sont rares. Dans cette étude, nous présentons des données morphologiques et moléculaires pour une espèce potentiellement nouvelle de *Redudasys* originaire de l'aquifère d'Edwards, Texas, USA. Il s'agit de la première signalisation dans l'hémisphère nord pour ce genre, précédemment mentionné uniquement du Brésil. Le spécimen du Texas, désigné ici comme *Redudasys* sp. possède une seule paire de tubes adhésifs antérieurs et est considérablement plus petit que les spécimens rapportés du Brésil. Les données moléculaires des gènes 18S rRNA et COI mtDNA supportent l'hypothèse d'une parenté étroite de *Redudasys* sp. avec *Redudasys fornerise* Kisielewski, 1987. Les données barcode sont fournies pour les *Redudasys* sp. et *R. fornerise*. La faible quantité de matériel obtenu ne justifie pas la description d'une nouvelle espèce, mais la signalisation est certainement importante du point de vue biogéographique. La colonisation et l'invasion d'espèces marines dans les habitats d'eau douce, en particulier ceux de l'aquifère d'Edwards, sont discutées. Bien que certains gastrotriches marins semblent avoir une capacité étonnante à s'adapter aux changements de salinité, *Redudasys* est probablement une relicte marine.

MOTS CLÉS
Redudasyidae,
biogéographie,
écoulement artésien,
aquifére d'Edwards,
Texas,
signalisation nouvelle.

INTRODUCTION

Gastrotricha Metschnikoff, 1865 are microscopic aquatic organisms and a common component of the interstitial, epibenthic and periphytic fauna. The group is cosmopolitan with close to 800 species (see Balsamo *et al.* 2009, 2013; Hummon & Todaro 2010; Kolicka 2014 and references therein). The group is classified into the two orders Chaetonotida Remane, 1925 [Rao & Clausen, 1970] and Macrodasyida Remane, 1925 [Rao & Clausen, 1970]. Chaetonotida contains tenpin-shaped species with representatives in both freshwater and marine habitats, while the strap-shaped species of Macrodasyida are almost exclusively marine. Only two nominal species of Macrodasyida, *Marinellina flagellata* Ruttner-Kolisko, 1955 and *Redudasys fornerise* Kisielewski, 1987, have been recorded from freshwater habitats.

Marinellina flagellata was described from the river Ybbs at Lunz in Austria from two, apparently immature, specimens. Attempts at rediscovering the species at the type locality have failed. However, a freshwater macrodasyidan has been found in another Austrian stream but it is not clear if this specimen belong to *M. flagellata* (see Todaro *et al.* 2012). Recently Araújo *et al.* (2013) reported an apparently different species of *Marinellina* Ruttner-Kolisko, 1955 from the state of Minas Gerais, Brazil.

Redudasys fornerise has so far only been reported from the type locality, Represa do Broa, state of Sao Paulo, Brazil (Kisielewski 1987a; Todaro *et al.* 2012). In addition, Garraffoni *et al.* (2010) reported a representative of *Redudasys*, *Redudasys* sp., in high numbers from the Cabral Mountains, state of Minas Gerais, Brazil. It is apparently different from *R. fornerise* but its taxonomic status has yet to be assessed (Todaro *et al.* 2012). Hence, the scarcity of data available to date may give the false impression of a purely tropical distribution of *Redudasys*.

In this study we present morphological and molecular data of a potentially new subtropical species of *Redudasys* from San Marcos, Texas, USA. It is the first record of the genus from the Northern hemisphere.

MATERIAL AND METHODS

Study area

The San Marcos springs are an artesian outflow from the Edwards Aquifer located on the eastern edge of Edwards Plateau, Texas, USA. Springs of the Edwards Aquifer provide most of the water for the San Marcos River that runs for 121 kilometers before it joins the Guadalupe River to the southeast. In 1849, a dam was built just downstream from the springs resulting in Spring Lake, which served as a recreational center and amusement park known as Aquarena Springs during the greater part of the 1900s. During the 1990s, Texas State University purchased the park and the surrounding area, now known as Meadows Center for Water and the Environment.

Collection and documentation

Collection was done by SCUBA on June 16, 2012 at a water temperature of 23-25°C. Samples were collected in 0.5L plastic jars at 3-9 meters by scooping up the top layer of the sediment. Samples contained clean sand, pebbles, algae and organic material. All samples were processed within 24 hours. Subsamples were treated with a 1% solution of MgCl2 to anaesthetize the animals and subsequently transferred to a small petri dish. Individual specimens were picked out with a micropipette under an Olympus LMS225R dissecting microscope. Specimens were mounted alive on a glass slide and subsequently studied and documented under an Olympus BH-2 microscope equipped with DIC optics and a Canon EOS Rebel T1i digital camera. After documentation specimens were transferred to 95% EtOH for further treatment.

The position of certain morphological characters along the body are given in percentage units (U) of the total body length following the convention of Hummon *et al.* (1992).

Sequencing and molecular analysis

Extraction, amplification and sequencing follow Kånneby *et al.* (2012). A complete list of taxa included in the analysis can be found in Table 1. The 18S rDNA and COI mtDNA genes were aligned with MAFFT v.7 (online version). The 18S rRNA gene was aligned using the Q-INS-i algorithm, taking the secondary structure of RNA into account, while COI was aligned using the G-INS-i algorithm (Katoh & Standley 2013). The concatenated dataset (18S rDNA + COI mtDNA) was analyzed with MrBayes v.3.2 (Ronquist *et al.* 2012) under the mixed model setting with estimated proportion of invariable sites and gamma distributed rate variation across sites. To ensure sufficient mixing and a reliable sample from the posterior distribution the dataset was analyzed with 8 MCMC chains for each run and a heating parameter of 0.1. The analysis was run for 20 million generations with default setting of priors. Convergence of the two runs was ascertained by checking the log likelihood graphs, the average standard deviation of split frequencies and the potential scale reduction factor (PSRF). After a burnin of 5 million generations, chains were sampled every 1000th generation. Members of Muselliferidae Leasi & Todaro, 2008 and Xenotrichulidae Remane, 1927 were used as outgroups (Table 1).

Uncorrected pairwise distances were computed with MEGA6 (Tamura *et al.* 2013).

Abbreviations

DLS	dorsolateral sensory hairs;
E	egg;
NA	not available;
PhiJ	pharyngeal intestinal junction;
PhL	length of pharynx;
PP	pharyngeal pores;
TbA	anterior adhesive tubes;
TL	total body length;
TbP	posterior adhesive tubes.

TABLE 1. — Gastrotrich taxa used in the phylogenetic analysis and their respective Genbank accession numbers. Abbreviations: see Material and methods.

Taxon	Origin	Reference	18S/COI Accession number
Macrodasyida Remane, 1925 [Rao & Clausen, 1970]			
Cephalodasyidae Hummon & Todaro, 2010			
Cephalodasys sp.	White Sea, Russia	Petrov *et al.* 2007	AY963691
Dolichodasys sp.	San Isidoro, Italy	Todaro *et al.* 2003	AM231778
Mesodasys laticaudatus Remane, 1951	Albinia, Italy	Todaro *et al.* 2011	JF357657/JF432043
Mesodasys laticaudatus Remane, 1951	Bohuslän, Sweden	Todaro *et al.* 2011	JF357668/JF432050
Mesodasys littoralis Remane, 1951	Bou Ficha, Tunisia	Todaro *et al.* 2011	JF357658/JF432044
Paradasys sp.	Ionian Sea, Italy	Todaro *et al.* 2003	AM231781
Pleurodasys helgolandicus (Remane, 1927)	NA	Paps & Riutort 2012	JF970236
Pleurodasys helgolandicus	Ibiza, Spain	Todaro *et al.* 2012	JN203486
Dactylopodolidae Strand, 1929			
Dactylopodola cf. *baltica* (Remane, 1926)	Ras Alard, Kuwait	Todaro *et al.* 2011	JF357650
Dactylopodola mesotyphle Hummon, Todaro, Tongiorgi & Balsamo, 1998	Punta Ala, Italy	Todaro *et al.* 2011	JF357651/JF432036
Dactylopodola typhle Remane, 1927	Bou Ficha, Tunisia	Todaro *et al.* 2011	JF357652/JF432037
Dactylopodola typhle Remane, 1927	Torre Civette, Italy	Todaro *et al.* 2011	JF357653/JF432038
Macrodasyidae Remane, 1926			
Macrodasys sp. 1	Torre Civette, Italy	Todaro *et al.* 2011	JF357654/JF432040
Macrodasys sp. 2	Bohuslän, Sweden	Todaro *et al.* 2011	JF357670/JF432052
Urodasys sp.	NA	Giribet *et al.* 2004	AY218102
Urodasys sp.	Florida, USA	Sørensen *et al.* 2006	DQ079912
Planodasyidae Rao & Clausen, 1970			
Crasiella sp.	Ilha Bela, Brazil	Todaro *et al.* 2012	JN203488
Megadasys sp.	Grotto del Ciolo, Italy	Todaro *et al.* 2011	JF357655/JF432042
Megadasys sp. 1	Porto Cesareo, Italy	Todaro *et al.* 2011	JF357656/JF432041
Redudasyidae Todaro, Dal Zotto, Jondelius, Hochberg, Hummon, Kånneby & Rocha, 2012			
Anandrodasys agadasys (Hochberg, 2003)	St. John Island, USA	Todaro *et al.* 2012	JN203487
Redudasys sp.	San Marcos, Texas, USA	This study	KJ950121/KJ950123
Redudasys fornerise Kisielewski, 1987	Represa do Broa, Brazil	Todaro *et al.* 2012; This study	JN203489/KJ950122
Thaumastodermatidae Remane, 1927			
Acanthodasys sp. A	Capraia, Italy	Todaro *et al.* 2011	JF357638
Acanthodasys aculeatus Remane, 1927	Capraia, Italy	Todaro *et al.* 2011	JF357639
Diplodasys ankeli Wilke, 1954	Meloria, Italy	Todaro *et al.* 2011	JF357624
Diplodasys ankeli Wilke, 1954	Bohuslän, Sweden	Todaro *et al.* 2011	JF357667/JF432049
Diplodasys meloriae Todaro, Balsamo & Tongiorgi, 1992	Meloria, Italy	Todaro *et al.* 2011	JF357640/JF432031
Diplodasys sp.	Kuwait	Todaro *et al.* 2011	JF357641/JF432032
Oregodasys ocellatus (Clausen, 1965)	Meloria, Italy	Todaro *et al.* 2011	JF357642
Oregodasys ruber (Swedmark, 1956)	Meloria, Italy	Todaro *et al.* 2011	JF357625/JF432020
Oregodasys tentaculatus (Swedmark, 1956)	Meloria, Italy	Todaro *et al.* 2011	JF357628/JF432021
Pseudostomella etrusca Hummon, Todaro & Tongiorgi, 1993	Albinia, Italy	Todaro *et al.* 2011	JF357633/JF432026
Ptychostomella sp. 1	Ilha Bela, Brazil	Todaro *et al.* 2011	JF357643/JF432033
Ptychostomella tyrrhenica Hummon, Todaro & Tongiorgi, 1993	Albinia, Italy	Todaro *et al.* 2011	JF357634/JF432027

TABLE 1. — Continuation.

Taxon	Origin	Reference	18S/COI Accession number
Tetranchyroderma cf. *antenniphorum* Hummon & Todaro, 2010	Kuwait	Todaro *et al.* 2011	JF357645
Tetranchyroderma cirrophorum Lévi, 1950	Capraia, Italy	Todaro *et al.* 2011	JF357635/JF432028
Tetranchyroderma esarabdophorum Tongiorgi & Balsamo, 1984	Mahdia, Tunisia	Todaro *et al.* 2011	JF357627
Tetranchyroderma hirtum Luporini, Magagnini & Tongiorgi, 1973	Capraia, Italy	Todaro *et al.* 2011	JF357628/JF432023
Tetranchyroderma pachysomum Hummon, Todaro & Tongiorgi, 1993	Meloria, Italy	Todaro *et al.* 2011	JF357636/JF432029
Tetranchyroderma papii Gerlach, 1953	Sardegna, Italy	Todaro *et al.* 2011	JF357637/JF432030
Tetranchyroderma quadritentaculatum Todaro, Balsamo & Tongiorgi, 1992	Albinia, Italy	Todaro *et al.* 2011	JF357629
Tetranchyroderma quadritentaculatum Todaro, Balsamo & Tongiorgi, 1992	Punta Ala, Italy	Todaro *et al.* 2011	JF357647/JF432024
Tetranchyroderma thysanophorum Hummon, Todaro & Tongiorgi, 1993	Fautea, Corsica, France	Todaro *et al.* 2011	JF357646/JF432025
Tetranchyroderma sp. 1	Bohuslän, Sweden	Todaro *et al.* 2011	JF357672
Tetranchyroderma sp. 3	Ilha Bela, Brazil	Todaro *et al.* 2011	JF357648/JF432035
Tetranchyroderma sp. 4	Ilha Bela, Brazil	Todaro *et al.* 2011	JF357644
Thaumastoderma moebjergi Clausen, 2004	Bohuslän, Sweden	Todaro *et al.* 2011	JF357671
Thaumastoderma ramuliferum Clausen, 1965	Pumta Ala, Italy	Todaro *et al.* 2011	JF357649
Turbanellidae Remane, 1927			
Paraturbanella dohrni Remane, 1927	Punta Ala, Italy	Todaro *et al.* 2011	JF357659
Paraturbanella pallida Luporini, Magagnini & Tongiorgi, 1973	Capraia, Italy	Todaro *et al.* 2011	JF357660/JF432045
Paraturbanella teissieri Swedmark, 1954	Punta Ala, Italy	Todaro *et al.* 2011	JF357661
Turbanella bocqueti Kaplan, 1958	Tramore, Ireland	Todaro *et al.* 2011	JF357662/JF432046
Turbanella cornuta Remane, 1925	Chioggia, Italy	Todaro *et al.* 2011	JF357663/JF432047
Turbanella lutheri Remane, 1952	Torö, Sweden	Todaro *et al.* 2011	JF357669/JF432051
Xenodasyidae Todaro, Guidi, Leasi & Tongiorgi, 2006			
Xenodasys riedli (Schoepfer-Sterrer, 1969)	St. John Island, USA	Todaro *et al.* 2012	JN203490
Chaetonotida Remane, 1925 [Rao & Clausen, 1970]			
Muselliferidae Leasi & Todaro, 2008			
Musellifer delamarei (Renaud-Mornant, 1968)	NA	Todaro *et al.* 2006	AM231775
Musellifer reichardti Kånneby, Atherton & Hochberg, 2014	Capron Shoal, Florida, USA	Kånneby *et al.* 2014	KF578503
Xenotrichulidae Remane, 1927			
Draculiciteria tesselata (Renaud Mornant, 1968)	Punta Ala, Italy	Kånneby *et al.* 2012	JN185457/JN185541
Draculiciteria tesselata	St. John Island, USA	Kånneby *et al.* 2012	JN185470/JN185549
Heteroxenotrichula squamosa Wilke, 1954	Punta Ala, Italy	Kånneby *et al.* 2013	JQ798542
Xenotrichula intermedia Remane, 1934	Mahdia, Tunisia	Todaro *et al.* 2011	JF357664/JF432047
Xenotrichula punctata Wilke, 1954	Östersidan, Sweden	Kånneby *et al.* 2012	JN185464
Xenotrichula velox Remane, 1927	Åhus, Sweden	Kånneby *et al.* 2012	JN185499
Xenotrichula sp.	NA	Paps & Riutort 2012	JF970234
Xenotrichula sp. 1	St. John Island, USA	Kånneby *et al.* 2012	JN185466/JN185545

Table 2. — Selected characters for species within *Redudasys* Kisielewski, 1987 and *Marinellina* Ruttner-Kolisko, 1955. Abbreviations: See Material and methods. All measurements in micrometers.

	***Redudasys* sp. (Texas)**	***Redudasys fornerise* Kisielewski, 1987**	***Redudasys* sp. (Brazil)**	***Marinellina flagellata* Ruttner-Kolisko, 1955**	***Marinellina* sp.**
TL	242	300 – 414	280 – 462	220	405
Length TbA shorter	Absent	3 – 9	Absent	Absent	Absent
Length TbA longer	9 – 10	9 – 14	Present	10	Present
Length TbP inner	10 – 12	8 – 12.5	7 – 13	20	Present
Length TbP outer	15 – 17	11 – 17	12 – 17.5	20	Present
PhL	92	101 – 154	87.5 – 154	NA	156
PhIJ	U38	U35 – 36	NA	NA	NA
PP	U33	U31	NA	Absent?	NA

SYSTEMATICS

Phylum GASTROTRICHA Metschnikoff, 1865
Order MACRODASYIDA Remane, 1925
[Rao & Clausen, 1970]
Family Redudasyidae Todaro, Dal Zotto, Jondelius, Hochberg, Hummon, Kånneby & Rocha, 2012
Genus *Redudasys* Kisielewski, 1987

Redudasys sp.
(Figs 1-3; Tables 2, 3)

Material examined. — One specimen that is no longer extant. Specimen prepared for sequencing of 18S rRNA and COI mtDNA. GenBank Accession numbers: KJ950121 and KJ950123.

Distribution. — In fine to medium grained sand with some organic content at 3-9 meters. Spring Lake, San Marcos, Texas, USA (29°53’36”N; 97°55’53”W), June 17, 2012.

Description

Redudasys with total body length of 242 µm. Body width at head (U12), neck (U24), trunk (U76) and base of caudal lobes (U94) is, 47 µm, 34 µm, 47 µm and 21 µm, respectively. Head with clearly distinguishable sensory cilia. Slight neck constriction present. Trunk of approximate equal width and not wider than widest part of head, narrowing toward the caudal end which indents at U95. Body surface transparent, without cuticular structures.

Apparently only a single pair of TbA present, 9-10 µm in length, and located at U13. Two pairs of TbP: inner pair 10-12 µm in length and outer pair 15-17 µm in length.

Sensory cilia abundant in anterior head region and reach lengths up to 25 µm. Anteriormost cilia surrounding mouth appear stiffer and are shorter, approximately 10 µm in length. At least seven pairs of dorsolateral to lateral sensorial hairs observed, posteriormost pair inserted just anterior to caudal indent. Ventral ciliation apparently in somewhat regularly spaced tufts from U8 to U50. Posterior to U50, tufts are medial in distribution.

Mouth terminal. Pharynx 92 µm long with pharyngeal intestinal junction located at U38. Pharyngeal pores could not be detected in the live specimen but they were subsequently detected in photos, they are located in the posterior part of the pharynx at approximately U33. Intestine straight, narrowing towards its posterior end, with anus at approximately U90.

One egg, approximately 50 µm in diameter, present dorsally in mid-posterior trunk region. Sperm or accessory reproductive organs not detected.

Remarks

Compared to other specimens of *Redudasys*, the most notable difference of the Texan specimen is its smaller size. The shortest total body lengths reported for *R. fornerise* and *Redudasys* sp. from Brazil are 300 µm and 280 µm, respectively (Kisielewski 1987a;

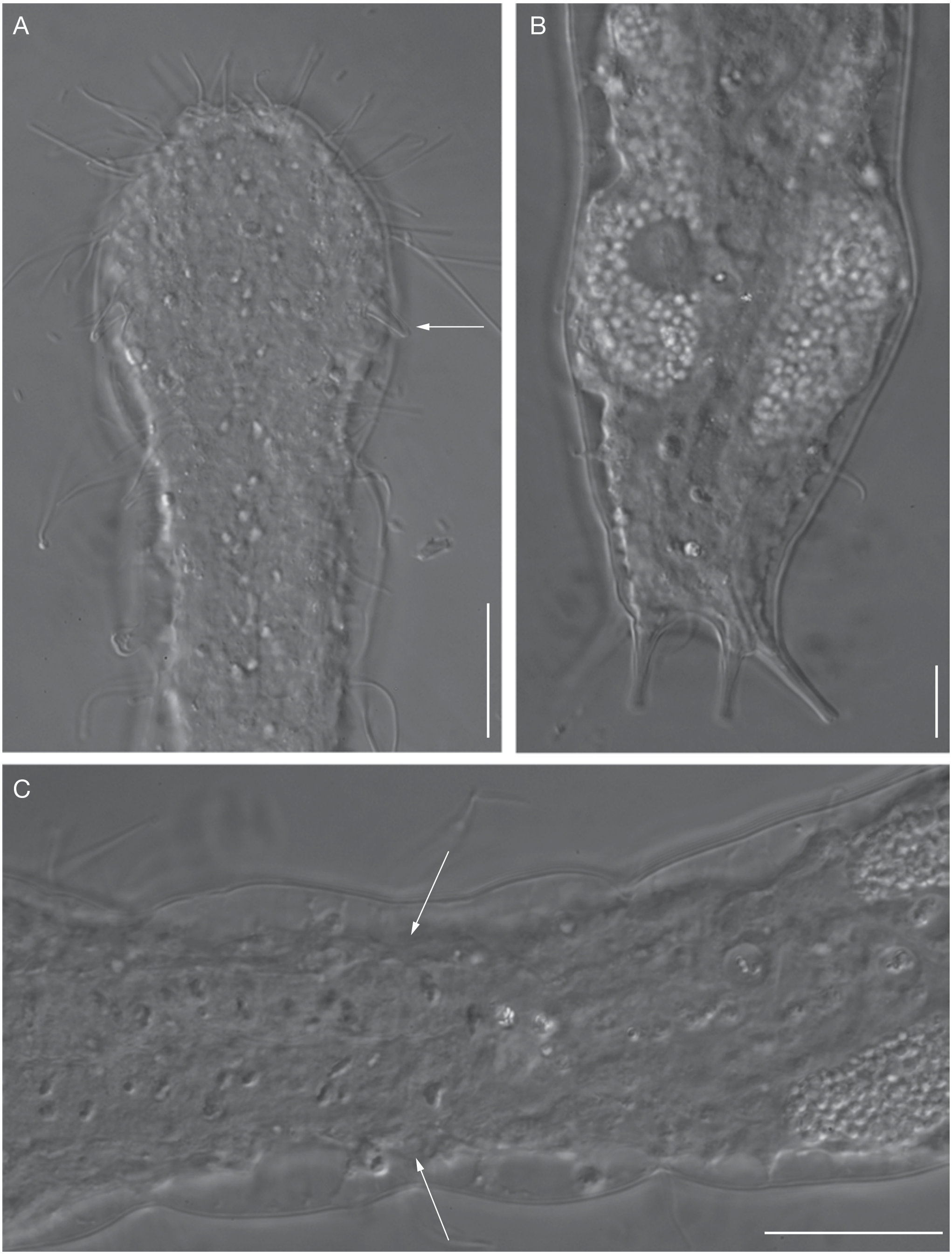

FIG. 1. — *Redudasys* sp. found in San Marcos, Texas: **A**, ventral view of anterior portion of body showing the anterior adhesive tubes (arrow) and the sensorial hairs of the head; **B**, posterior portion of body showing the caudal lobes with posterior adhesive tubes; **C**, posterior part of pharynx showing the position of pharyngeal pores (arrows). Scale bars: A, C, 20 µm; B, 10 µm.

Table 3. — Single pure characters for each species based on an alignment of only the taxa within Redudasyidae. For each gene the unique nucleotide and its position in the alignment are given. The number within parenthesis corresponds to the same position in the original unaligned sequence. Only the ten first unique positions are presented for each species where applicable. Abbreviations: see Material and methods.

Species	Gene	Unique nucleotide with position in alignment and in original sequence
Redudasys sp.	18S	C, 127 (123); T, 134 (130); A, 150 (145); G, 194 (175); T, 208 (189); G, 687 (658); A, 1352 (1316); T, 1449 (1318); A, 1647 (1516); T, 1804 (1673)
	COI	T, 1 (1); T, 10 (10); C, 14 (14); T, 22 (22); G, 34 (34); C, 44 (44); T, 46 (46); C, 48 (48); T, 49 (49); T, 67 (67)
Redudasys fornerise Kisielewski, 1987	18S	A, 124 (122); A, 125 (123); A, 126 (124); T, 129 (127); A, 131 (129); A, 134 (132); T, 150 (147); T, 153 (150); C, 208 (191)
	COI	A, 1 (1); C, 10 (10); T, 14 (14); G, 22 (22); A, 34 (34); T, 44 (44); A, 46 (46); T, 48 (48); C, 49 (49); C, 67 (67)
Anandrodasys agadasys (Hochberg, 2003)	18S	C, 20 (20); T, 21 (21); C, 23 (23); A, 24 (24); C, 28 (28); A, 29 (29); C, 43 (43); A, 56 (56); G, 61 (61); T, 80 (80)
	COI	NA

Garraffoni *et al.* 2010), compared to 242 µm for the Texan specimen (Table 2). However, our specimen could be a young adult, judging from the presence of only a single egg, and it may attain a larger body size as it matures. The Texan specimen can be separated from *R. fornerise* based on the following combination of morphological characters: (i) a single pair of anterior adhesive tubes, and (ii) a shorter body length. Although the second pair of anterior adhesive tubes can be hard to detect, they appear absent in our specimen. The only apparent difference from the Brazilian *Redudasys* sp. reported by Garraffoni *et al.* (2010) is the shorter body length (Table 2). The exact distribution of lateral and/or dorsolateral sensorial hairs cannot be justified from the single specimen obtained in this study. However, since the specimen from Texas is geographically far separated from the Brazilian taxa, it is plausible that it is new to science.

The classification of our specimen within the family Redudasyidae and the genus *Redudasys* is based on the presence of pharyngeal pores in the posterior part of the pharynx and the unequal length of the inner and outer pair of TbP. The Bayesian analysis of the concatenated data set of 18S rDNA and COI mtDNA give maximum support for the inclusion of our specimen within Redudasyidae. The uncorrected pairwise distance based on COI mtDNA, between the Texan specimen and *R. fornerise* is 0.175. In order to diagnose the Texan specimen, we have provided the position of the ten first unique nucleotides for each taxa for which sequence data is available within Redudasyidae (Table 3). However, it should be noted that *R. fornerise* is the only other freshwater macrodasyidan for which sequences are available on GenBank.

DISCUSSION

All in all there are now five taxa of the predominantly marine Macrodasyida that have been reported from freshwater (Table 2). They are all characterized by a reduction of the number of adhesive tubes, only anterior and posterior adhesive tubes are typically present (Kisielewski 1987; Todaro *et al.* 2012). The poor description of *M. flagellata* and the fact that it is based on apparently immature specimens further complicates the morphological separation of *Marinellina* and *Redudasys* (see Kisielewski 1987). However, there are some characters that can be used to distinguish *Marinellina* from *Redudasys*: i) the TbP of *M. flagellata* are of equal length, while in *Redudasys* the inner pair of TbP are shorter than the outer pair; ii) the absence of pharyngeal pores in *M. flagellata*. Ruttner-Kolisko (1955) was well aware of the presence of pharyngeal pores in Macrodasyida and hypothesized that the reduction of these pores were an adaptation to a life in a freshwater environment. However, it should be noted that pharyngeal pores are sometimes difficult to observe and this may be especially true for such a small species as *M. flagellata*. It is also possible that *M. flagellata* is in fact a juvenile form of *Redudasys*. To disentangle such a relationship it would be appropriate to collect and sequence several specimens of *Marinellina* from the type locality in Austria. This however appears a daunting task since all attempts at rediscovering *M. flagellata* at the type locality have failed. The new findings of a hypothesized member of *Marinellina*, designated as *Marinellina* sp., from Brazil by Araujo *et al.* (2013) may shed some light on this issue if it is formally described. This taxon has a much greater body length than *M. flagellata*. Whether pharyngeal pores are absent and the TbP are of unequal length or not is not mentioned in the short description.

Turbanella cornuta Remane, 1925, *Turbanella hyalina* Schultze, 1853 and *Turbanella lutheri* Remane, 1952 have been reported from the Baltic Sea at salinities of 0.2-0.6% (Jansson 1968), although these species are well known from localities with proper marine salinity (Kieneke *et al.* 2012). Kisielewski (1987b) reported *T. lutheri* from freshwater beach seeps in Roscoff, France. Moreover, several marine chaetonotidans have been recorded from brackish environments in the Baltic Sea (e.g., Hummon 2008; Kånneby *et al.* 2013). This suggests that certain taxa of both Macrodasyida and Chaetonotida have a remarkable tolerance to adapt to changes in salinity. It has to be stressed that future genetic studies could demonstrate that these taxa are indeed complexes of cryptic species where each species is adapted to a narrower range in salinity. *Redudasys* on the other hand is likely to be a marine relic. It has strong morphological affinities with marine Macrodasyida, e.g., a strap-shaped body, the inverted Y-shaped pharynx lumen and the presence of pharyngeal pores (Kisielewski 1987a) and presumably was derived from marine ancestors by stranding following the regression of marine embayments. All localities where the taxon hitherto has been found are far away from marine waters (the closest is Spring Lake in this study, approximately 221 km from the Gulf of Mexico).

Holsinger & Longley (1980) noted that there was a “strong possibility” that species of amphipod crustaceans in artesian wells of the Edwards Aquifer were derived from marine ancestors. These ancestors invaded newly opened freshwater habitats during the late Cretaceous, when a large embayment covered central Texas. These authors postulated the existence of marine relic species in groundwater and

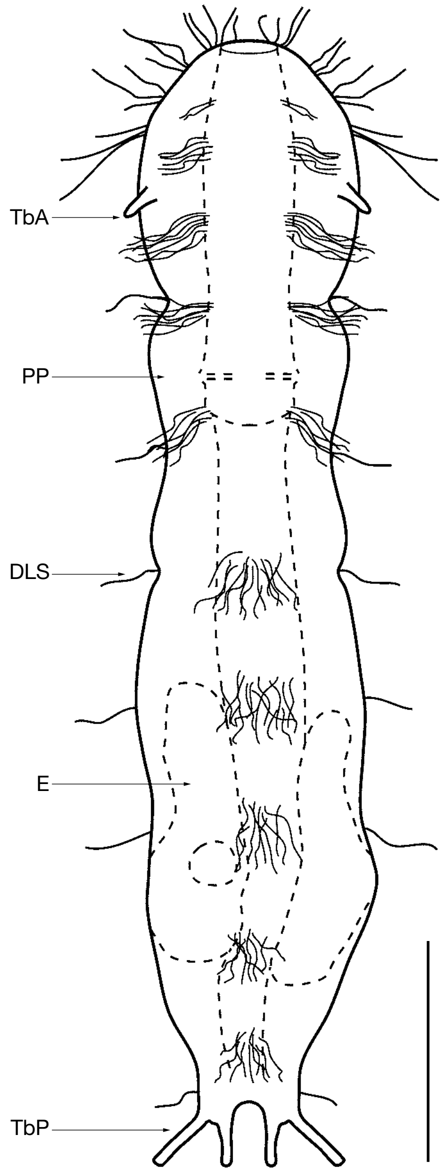

Fig. 2. — Schematic drawing of the ventral side of *Redudasys* sp. found in San Marcos, Texas. Abbreviations: see Material and methods. Scale bar: 50 µm.

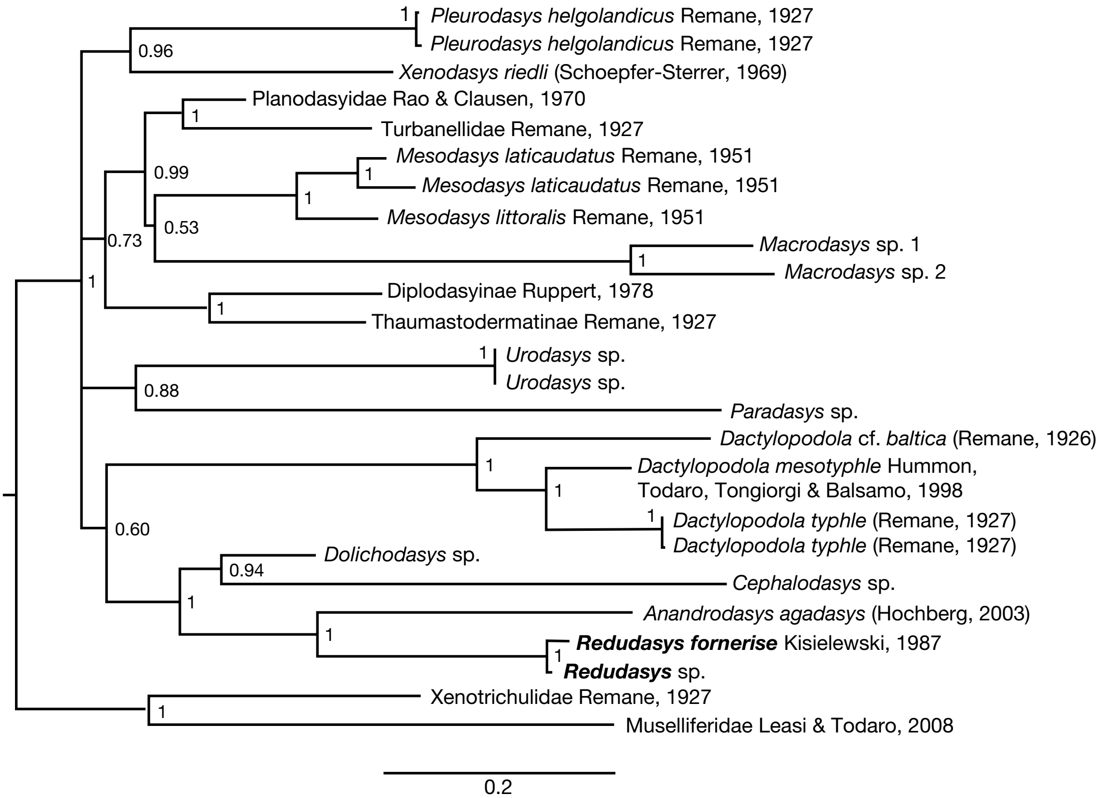

FIG. 3. — Majority rule consensus tree of the phylogenetic relationships of Macrodasyida inferred from Bayesian analysis of the 18S rRNA and COI mtDNA genes. Numbers at nodes represent posterior probability. *Redudasys* sp. from Texas and *Redudasys fornerise* are highlighted in bold font. The outgroup is represented by members of Muselliferidae Leasi & Todaro, 2008 and Xenotrichulidae Remane, 1927 (Chaetonotida).The clades Planodasyidae, Muselliferidae Thaumastodermatidae, Turbanellidae and Xenotrichulidae have been collapsed for clarity. Dactylopodolidae was not collapsed, since it appears close to Redudasyidae.

cavernous limestone buried under late Cretaceous sediments and later exposed by uplift and faulting of the Balcones fault line during the Miocene. Gibson *et al.* (2008) expanded on these ideas, commenting that the rare crustaceans *Ingolfiella* sp., *Tethysbaena texana* (Maguire, 1965), and certain stygiobiontic amphipods, also found in the Edwards Aquifer, appear to have a Tethys Sea origin and colonized the area during regression of a Cretaceous sea.

Acknowledgements

The authors wish to extend their gratitude to Dr Randy Gibson, U. S. Fish and Wildlife Service, San Marcos, Texas, for providing some of the samples and to Dr Ronald Griffin, Department of Agricultural Economics, Texas A & M University, for providing necessary equipment, chemicals and invaluable help. Many thanks also go to Dr Veronica Lundgren, Centre for Ecology and Evolution in Microbial Ecosystems, Linneaus University and Dr Daniel Roelke, Department of Wildlife and Fisheries Sciences, Texas A & M University. Special thanks go to Dr M. Antonio Todaro, Department of Life Sciences, University of Modena and Reggio Emilia, Modena, Italy. We are also grateful to Keyvan Mirbakhsh, Department of Biodiversity Informatics, Swedish Museum of Natural History, Stockholm, Sweden, for technical assistance and to Dr Rick Hochberg, Department of Biology, University of Massachusetts Lowell, Lowell, USA as well as two anonymous reviewers for comments on an early draft of the paper. Emily Dock Åkerman, Department of Zoology, Swedish Museum of Natural History, Stockholm, Sweden is acknowledged for translating parts of this study into French. This study was financially supported by a grant from Lennanders Stiftelse (to TK). The Swedish Taxonomy Initiative is also acknowledged.

REFERENCES

Araújo T. Q., Alcântara F. C. & Garraffoni A. R. S. 2013. — New records of Gastrotricha from Minas Gerais, Brazil. *Studies on Neotropical Fauna and Environment* 48: 68-75.

Balsamo M., d'Hondt J. L., Pierboni L. & Grilli P. 2009. — Taxonomic and nomenclatural notes on freshwater Gastrotricha. *Zootaxa* 2158: 1-19.

Balsamo M., Guidi L. & d'Hondt J. L. 2013. — Phylum Gastrotricha, *in* Zhang Z.-Q. (ed.), Animal Biodiversity: An Outline of Higher-level Classification and Survey of Taxonomic Richness (Addenda 2013). *Zootaxa* 3703 (1): 79-82.

Garraffoni A. R. S., Araújo T. Q., Lourenço A. P. & Balsamo M. 2010. — New data on freshwater psammic Gastrotricha from Brazil. *Zookeys* 60: 1-12.

Gibson J. R., Harden S. J. & Fries J. N. 2008. — Survey and distribution of invertebrates from selected springs of the Edwards Aquifer in Comal and Hays counties, Texas. *The Southwestern Naturalist* 53: 74-84.

Giribet G., Sørensen M. V., Funch P., Kristensen R. M. & Sterrer W. 2004. — Investigations into the phylogenetic position of Micrognathozoa using four molecular loci. *Cladistics* 20: 1-13.

Holsinger J. R. & Longley G. 1980. — The subterranean amphipod crustacean fauna of an artesian well in Texas. *Smithsonian Contribution to Zoology* 308: 1-59.

Hummon W. D. 2008. — Brackish-Water Gastrotricha of the Polish Baltic Coast. *Meiofauna Marina* 16: 109-116.

Hummon W. D & Todaro M. A. 2010. — Analytic taxonomy and notes on marine, brackish-water and estuarine Gastrotricha. *Zootaxa* 2392: 1-32.

Jansson B-O. 1968. — Quantitative and experimental studies of the interstitial fauna of four Swedish sandy beaches. *Ophelia* 5: 1-71.

Kånneby T., Todaro M. A. & Jondelius U. 2012. — A phylogenetic approach to species delimitation in freshwater Gastrotricha from Sweden. *Hydrobiologia* 683: 185-202.

Kånneby T., Todaro M. A. & Jondelius U. 2013. — Phylogeny of Chaetonotidae and other Paucitubulatina (Gastrotricha: Chaetonotida) and the colonization of aquatic ecosystems. *Zoologica Scripta* 42: 88-105.

Kånneby T., Atherton S. & Hochberg R. 2014. — Two new species of *Musellifer* (Gastrotricha: Chaetonotida) from Florida and Tobago and the systematic placement of the genus within Paucitubulatina. *Marine Biology Research* 10: 983-995.

Katoh K. & Standley D. M. 2013. — MAFFT Multiple sequence alignment software version 7: Improvements in performance and usability. *Molecular Biology and Evolution* 30: 772-780.

Kieneke A., Martinez Arbizu P. & Fontaneto D. 2012. — Spatially structured populations with a low level of cryptic diversity in European marine Gastrotricha. *Molecular Ecology* 21: 1239-1254.

Kisielewski J. 1987a. — Two new interesting genera of Gastrotricha (Macrodasyida and Chaetonotida) from the Brazilian freshwater psammon. *Hydrobiologia* 153: 23-30.

Kisielewski J. 1987b. — New records of marine Gastrotricha from the French coasts of Manche and Atlantic I. Macrodasyida, with descriptions of seven new species. *Bulletin du Muséum national d'Histoire naturelle* 9: 837-877.

Kolicka M. 2014. — Gastrotricha from Copenhagen Palm House – contribution to the knowledge of Lepidodermella intermedia Kånneby, Todaro & Jondelius, 2012 (Chaetonotida, Gastrotricha). *Zoosystema* 36 (4): 713-722. http://dx.doi.org/10.5252/z2014n4a1.

Paps J. & Riutort M. 2012. — Molecular phylogeny of the phylum Gastrotricha: new data brings together molecules and morphology. *Molecular Phylogenetics and Evolution* 63: 208-212.

Petrov N. B., Pegova A. N., Manylov O. G., Vladychenskaya N. S., Mugue N. S. & Aleshin V. V. 2007. — Molecular phylogeny of Gastrotricha on the basis of comparison of the 18S rRNA genes: Rejection of the hypothesis of a relationship between Gastrotricha and Nematoda. *Molecular Biology* 41: 445-452.

Ronquist F., Teslenko M., van der Mark P., Ayres D. L., Darling A., Höhna S., Larget B., Liu L., Suchard M. A. & Huelsenbeck J. P. 2012. — MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology* 61: 539-542.

Ruttner-Kolisko A. 1955. — *Rheomorpha neiswestnovae* und *Marinellina flagellata*, zwei phylogenetische interessante Wurmtypen aus dem Süsswasserpsammon. *Österreichische Zoologischer Zeitschrift* 6: 55-69.

Sørensen M. V., Sterrer W. & Giribet G. 2006. — Gnathostomulid phylogeny inferred from a combined approach of four molecular loci and morphology. *Cladistics* 22: 32-58.

Tamura K., Stecher G., Peterson D., Filipski A. & Kumar S. 2013. — MEGA6: Molecular Evolutionary

Genetics Analysis Version 6.0. *Molecular Biology and Evolution* 30: 2725-2729.
Todaro M. A., Littlewood D. T. J., Balsamo M., Herniou E. A., Cassanelli S., Manicardi G., Wirz A. & Tongiorgi P. 2003. — The interrelationships of the Gastrotricha using nuclear small rRNA subunit sequence data, with an interpretation based on morphology. *Zoologischer Anzeiger* 242: 145-156.
Todaro M. A., Telford M. J., Lockyer A. E. & Littlewood D. T. J. 2006. — Interrelationships of the Gastrotricha and their place among the Metazoa inferred from 18S rRNA genes. *Zoologica Scripta* 35: 251-259.
Todaro M. A., Kånneby T., Dal Zotto M. & Jondelius U. 2011. — Phylogeny of Thaumastodermatidae (Gastrotricha: Macrodasyida) inferred from nuclear and mitochondrial sequence data. *PLoS ONE* 6(3): e17892.
Todaro M. A., Dal Zotto M., Jondelius U., Hochberg R., Hummon W. D., Kånneby T. & Rocha C. E. F. 2012. — Gastrotricha: A marine sister for a freshwater puzzle. *PLoS ONE* 7(2): e31740.

Submitted on 12 March 2014;
accepted on 27 June 2014;
published on 26 December 2014.