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Conqueria laevis gen. and sp. nov., a new soft-walled, monothalamous foraminiferan from the deep Weddell Sea

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Conqueria laevis gen. and sp. nov., a new monothalamous agglutinated foraminiferan, is described from core samples collected in the abyssal western Weddell Sea. The species is characterized by a very elongate, almost cylindrical test that usually follows a more or less curved course and has a single terminal aperture located at the end of a short neck. The wall has a very smooth outer surface and is composed of tiny ($<5\,\mu$m) agglutinated particles. Very similar and presumably congeneric morphotypes occur at northern hemisphere sites, including Arctic fjords around Svalbard and the Porcupine Abyssal Plain. Molecular phylogenetic analyses, based on small subunit rRNA gene sequences, indicate that the new Weddell Sea species forms an independent lineage branching among monothalamous foraminiferans as a sister group to the clade of *Psammophaga*.

INTRODUCTION

This study is based on material collected as part of the ANDEEP (ANtarctic Benthic DEEP-sea Biodiversity) project. The ANDEEP aims to provide the first comprehensive survey of benthic communities living at abyssal and bathyal depths in the western part of the Weddell Sea and the adjoining Scotia Sea and to investigate taxonomic links between the faunas of the Antarctic deep sea and those from other oceanic regions (Brandt et al., 2003). An important component of sediment-dwelling communities living on the ocean floor are the Foraminifera, a group of protists that often dominate both the meiofaunal and macrofaunal size fractions (Tendal & Hessler, 1977; Gooday et al., 1992). Assemblages of deep-sea meiofaunal foraminiferans typically consist of a mixture of taxa including those with: (1) fairly robust calcareous or agglutinated tests; (2) more delicate agglutinated tests (e.g. *Reophax* spp.); and (3) monothalamous (single chambered) forms with soft, flexible test walls composed of either organic or agglutinated material (e.g. allogromids and soft-walled saccamminids in the traditional sense) (Gooday et al., 1998; Gooday, 2002). The soft-shelled foraminiferans within this third category are poorly documented and include many undescribed species and higher taxa. In addition to being an important component of deep-sea biotas, they have considerable phylogenetic significance as representatives of the complex basal radiation from which the much better known multichambered foraminiferans arose (Pawlowski et al., 2003).

The purpose of this paper is to describe a new monothalamous foraminiferal genus and species on the basis of ANDEEP material collected at water depths greater than 4000 m. Similar morphotypes have been observed in material from a wide range of water depths in the northern hemisphere, suggesting that the new genus is widely distributed. The description is based on traditional morphological criteria, mainly test characteristics, combined with molecular phylogenetic data.

MATERIALS AND METHODS

Sampling sites

The samples were collected at four stations in the western Weddell Sea during RV 'Polarstern' Cruise ANT-XIX/4 (ANDEEP II; 28 February to 1 April 2002) (Brandt et al., 2003). Station 134 (4060 m water depth) was situated at the base of the continental slope on the eastern side of the Antarctic Peninsula. Stations 136, 137 and 138 were located on the Weddell Abyssal Plain (4500–4980 m). The sea-floor in the western Weddell Sea is overlain by Weddell Sea Deep Water and Weddell Sea Bottom Water. Bottom-water oxygen concentrations are not available from our sites but visual inspection of core samples suggests that the sediments were well-oxygenated at all stations. Sediment types include clayey silt of pelagic origin at Station 138 and hemipelagic silty clays at Stations 134, 136 and 137 (Howe, 2003).

Shipboard procedures

The material was collected using either a box-corer or a multiple corer equipped with 12 tubes (57 mm internal diameter). As soon as possible after collection, the upper 1 cm or so of sediment was removed and immediately sieved (500 and 125 μm screens) in a cool room set at 2°C. These unfixed residues were stored at this temperature for periods of several hours to several days. Living foraminifera were sorted from the residues under a binocular microscope in a Petri dish of seawater kept cool in a tray of ice. Specimens for morphological study were fixed in 10% formalin buffered with sodium borate and stored in plastic centrifuge vials. Between one and five specimens for molecular analyses were transferred to microtubes containing 60 μl of guanidine extraction buffer.

Morphological study

The tests were photographed either in water using a Wild M440 stereomicroscope (reflected light) or in

glycerol in a cavity slide using an Olympus BH2 photomicroscope (transmitted light). Test measurements (to the nearest 10 μm) were made using an eyepiece graticule fitted to a Wild M50 dissecting microscope.

Molecular analysis

A fragment of the small subunit (SSU) rRNA gene was amplified by polymerase chain reaction (PCR) with the primer pair sl4F3 (5′ACG CA(AC) GTG TGA AAC TTG) and sB (5′TGA TCC TTC TGC AGG TTC ACC TAC), as described in Pawlowski (2000). In all cases, the PCR products were re-amplified using nested primer sl4F1 (5′AAG GGC ACC ACA AGA ACG C). The amplified PCR products were purified using High Pure PCR Purification Kit (Roche Diagnostics) and sequenced directly. Sequencing reactions were prepared by using ABI-PRISM Big Dye Terminator Cycle Sequencing Kit and analysed with an ABI-377 DNA sequencer (Perkin-Elmer), all according to the manufacturer's instructions.

Sequences were aligned manually to the large database of foraminiferan sequences, using the Seaview software (Galtier et al., 1996). Six hundred and fifty-seven sites were selected for analysis, including 253 variable and 184 phylogenetically informative sites. Phylogenetic analyses were performed with the neighbour joining (NJ) method, applied to distances corrected using K2, HKY, LogDet models, and the maximum likelihood (ML) method using tree-building algorithm of FASTDNAML (Olsen et al., 1994). All characters were equally weighted and the transition−transversion ratio was set to 0.794, according to the mean ratio calculated over all sequence pairs. The reliability of internal branches was assessed by bootstrapping (Felsenstein, 1985) with 1000 re-sampling for the NJ and 100 re-sampling for the ML tree respectively. The Phylo_win program (Galtier et al., 1996) was used for distance computations using various models, NJ and ML tree-building and bootstrapping.

SYSTEMATICS

We follow recent suprageneric classifications by placing the new genus in the Order Astrorhizida, Superfamily Astrorhizacea, Family Saccamminidae (Loeblich & Tappan, 1987, 1989). However, we recognize that the monothalamous foraminiferans currently accommodated within the orders Allogromiida (organic walls) and Astrorhizida (agglutinated walls) include a number of phylogenetic lineages that cut across these traditional, morphology-based taxa (Pawlowski et al., 2002b, 2003). Here, we use the Astrorhiziida only as a convenient label.

Class FORAMINIFERA Lee, 1990
Order ASTRORHIZIDA Brady, 1881
Superfamily ASTRORHIZACEA Brady, 1881
Family SACCAMMINIDAE Brady, 1884
Genus *Conqueria* gen. nov.

Derivation of name

In honour of Mr Mike Conquer, photographer at the Institute of Oceanographic Sciences Deacon Laboratory (IOSDL), Wormley, UK, and now at the Southampton Oceanography Centre, UK.

Diagnosis

Test free, monothalamous, elongate, tubular, up to 1000 μm long and 50 to 100 μm wide with a simple aperture at the end of a short terminal neck. Wall very finely agglutinated with a very smooth, non-reflective outer surface. Cytoplasm without stercomata.

Type species

Conqueria laevis sp. nov.

Remarks

Hippocrepinella differs from the new genus in several respects. According to the original description (Heron-Allen & Earland, 1932; Loeblich & Tappan, 1987), the type species (*H. hirudinea* Heron-Allen & Earland, 1932) has a rather irregular test outline and often exhibits transverse wrinkles, a small secondary aperture is sometimes present at the proximal end, and there is no apertural neck. Molecular evidence suggests that species currently assigned to *Hippocrepinella* (including *H. hirudinea* and *H. alba* Heron-Allen & Earland, 1932) belong to evolutionary lineages that are distinct from the clade represented by *Conqueria* (see Figure 5 of this paper and Pawlowski et al., 2003). The absence of an apertural neck, together with its more elongate tubular morphology, also distinguishes *Hippocrepina* (type species *H. indivisa* Parker, 1870) from *Conqueria*. According to the very brief original description (in Dawson, 1870), the aperture of *H. indivisa* has a 'horse-shoe shaped form'.

Conqueria laevis sp. nov.
(Figures 1–4)

Derivation of name

Latin *laevis* meaning smooth, referring to the surface of the test.

Type material

Holotype: complete specimen preserved in dilute formalin, illustrated in Figures 1A & 2A (ANDEEP Station 138; coordinates: 62°58′S 27°54′W; water depth 4500 m). [Forschungsinstitut Senckenberg, Frankfurt am Main, Germany, registration number SMF XXVII 7511].

Paratypes: three complete specimens preserved in dilute formalin, illustrated in Figures 1B–D, 2B–D, 4A–D. [Forschungsinstitut Senckenberg, Frankfurt am Main, Germany, registration number SMF XXVII 7512].

Sixteen additional specimens in A.J. Gooday's collection.

Comparative material examined

ANDEEP Station 134 (65°19.90′S 48°5.55′W; water depth 4060 m), 6 specimens; ANDEEP Station 136 (64°1.67′S 39°7.68′W; water depth 4760 m), 2 specimens.

Diagnosis

As for genus.

Description

The test is elongate, tubular, with a smooth outline uninterrupted by irregularities apart from slight fluctuations in width (Figures 1 & 2). There is a single terminal

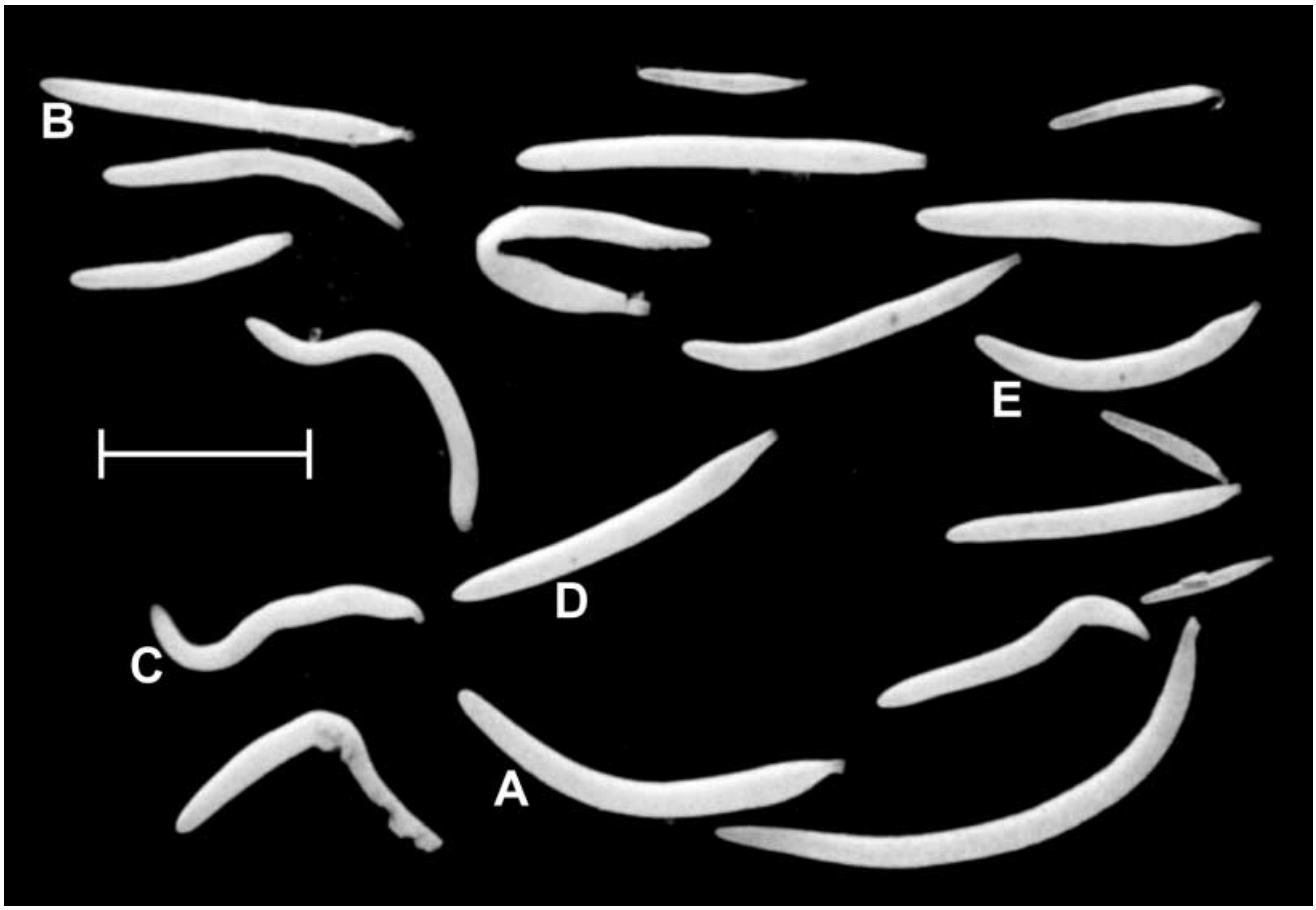

Figure 1. *Conqueria laevis* gen. and sp. nov. Twenty specimens from the type locality (ANDEEP Station 138) photographed in reflected light in water. The holotype (specimen A) and three paratypes (specimens B–D) are illustrated using transmitted light in Figures 2 and 4. Specimen E is illustrated by scanning electron micrograph (Figure 3). Scale bar: 500 μm.

aperture. The test is usually more or less straight or slightly curved, occasionally more strongly curved or following a sinuous course. It ranges from 340 to 1400 μm in length and 50 to 100 μm in width with a L:W ratio of 6.8–15.8 (usually between 8 and 13). The greatest width is located in the anterior half of the test, often fairly close to the apertural end. The posterior part of the test tapers very gradually towards the proximal end which is closed and narrowly rounded or bluntly pointed. The distal end is produced into a short neck, up to 100 μm long and 20–30 μm wide at its termination. The aperture is located at the end of this neck.

The test wall is whitish, more or less opaque in water, with a very smooth outer surface and a slight satin-like sheen. It is composed of very finely agglutinated, angular particles of uniform composition (probably quartz), the vast majority of them $<5\,\mu$m in size and forming a surface pavement that is fairly smooth on a scale of tens of microns. Smaller particles (1 μm or less) are present in the interstices between the larger grains (Figure 3G) and these very fine grains dominate the wall immediately adjacent to the aperture (Figure 3C). The cement is presumably organic, although it is not visible in any of the available scanning electron micrographs. The wall is of fairly even thickness (7–10 μm), except where it becomes thinner in the apertural neck (Figure 4B&D). The lumen of the test usually occupies 65–80% of the total external

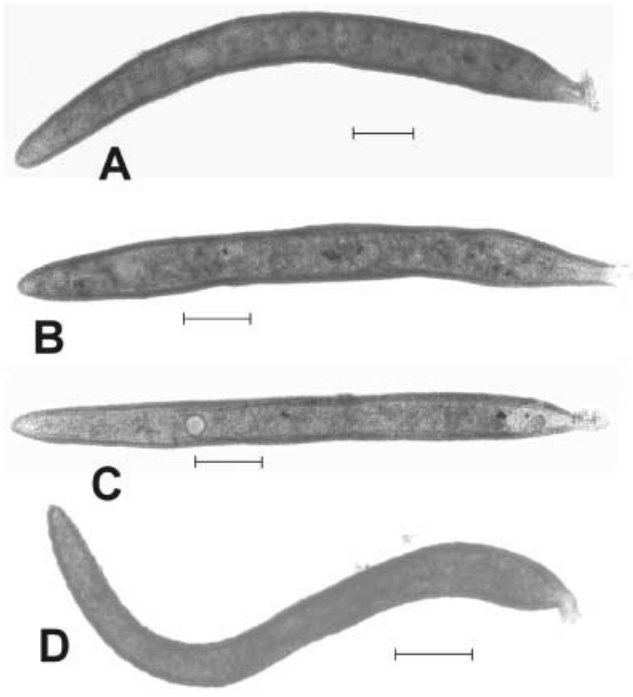

Figure 2. *Conqueria laevis* gen. and sp. nov. Holotype (A) and three paratypes (B) mounted in glycerol and photographed in transmitted light. Scale bars: 100 μm.

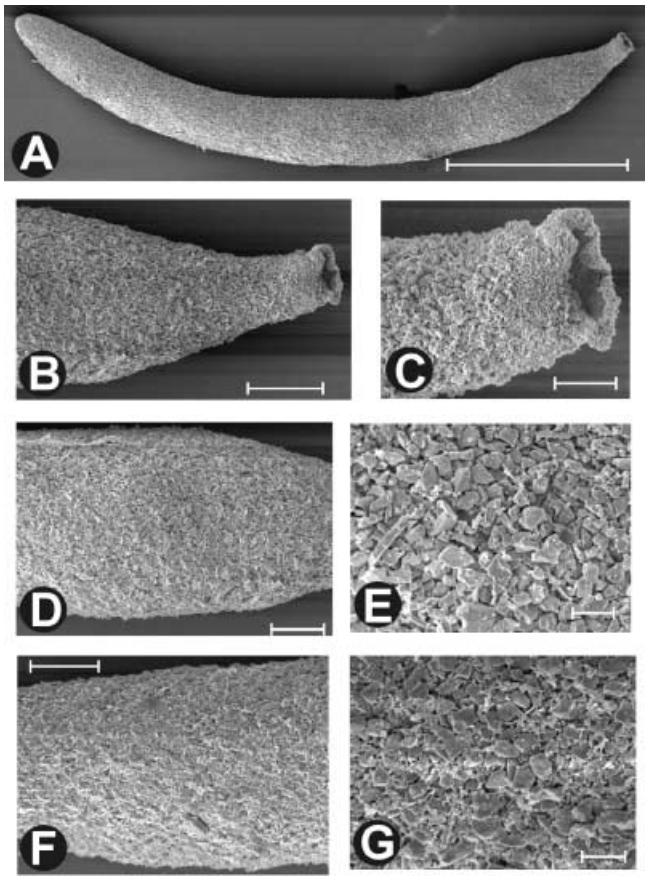

Figure 3. *Conqueria laevis* gen. and sp. nov. Specimens illustrated in Figure 1E: scanning electron micrographs of the test surface. (A) Entire test; (B,C) views of the tubular apertural extension; (D,E) surface near the apertural end of the test; (F,G) surface near the proximal end. Scale bars: A, 200 μm; B, 30 μm; C, 10 μm D&F, 20 μm; E&G, 5 μm.

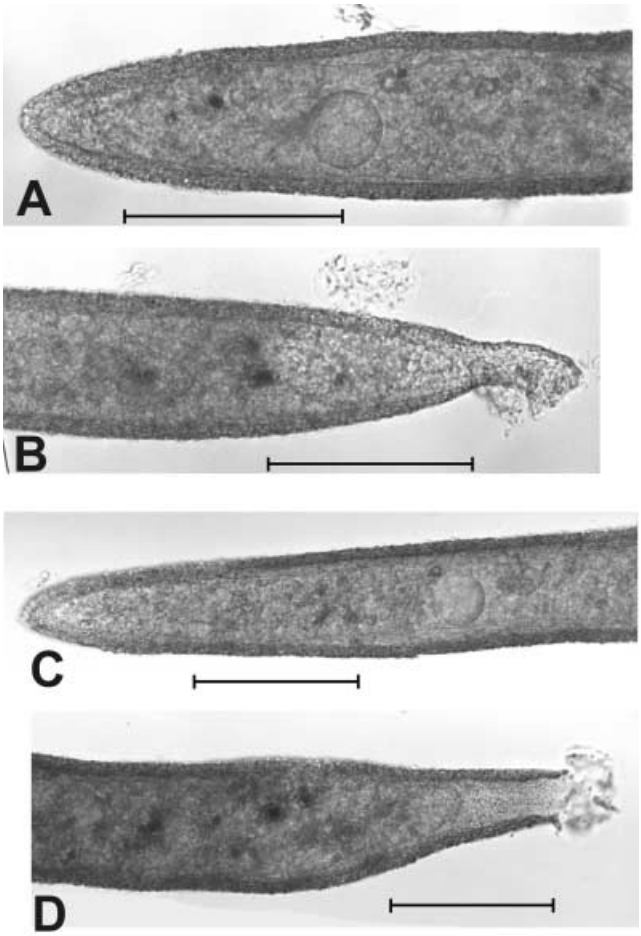

Figure 4. *Conqueria laevis* gen. and sp. nov. Details of two paratypes mounted in glycerol and photographed in transmitted light. (A) Narrowly rounded proximal end of test; (B) apertural end of same individual; (C) proximal end; (D) apertural end of same individual. The nucleus is clearly visible through the test wall near the proximal end in A and C. The dark patches are mineral grains within the cytoplasm. Scale bars: 100 μm.

width. It is completely filled by cytoplasm which becomes more or less constricted into a thread within the apertural tube. The cytoplasm of the four specimens examined in glycerol contained a large (35–45 μm) nucleus (Figure 4A&C). Other inclusions visible in glycerol mounted specimens include scattered mineral grains, some black, others angular transparent particles, presumably quartz.

Molecular characterization

The six sequences obtained group together as an independent lineage among monothalamous foraminifers (Figure 5). It branches as a sister group to the clade that includes species of *Psammophaga*, *Allogromia crystallifera* Dahlgren, 1962 and an undescribed new species of the genus *Vellaria* from Antarctic coastal waters. However, the relations between this clade and *Conqueria* are very weakly supported (23% in ML analysis and 58% in NJ analysis).

Among six sequences, it is possible to distinguish two groups, differing slightly in the variable regions of the SSU rDNA. One group is composed of four specimens from Stations 134, 136, 137, 138 (4060–4980 m water depth); the second group comprises two specimens from Stations 136 and 137. The four sequences in the first group are identical, while the two sequences in the second group differ by 0.35%. The sequence divergence between both groups equals 3.8%, while the divergence between the *Conqueria* and *Psammophaga* clade averages 22.5%.

Distribution

Western Weddell Sea; water depth 4060–4760 m.

Remarks

In general morphology and wall structure, *Conqueria laevis* resembles *Hippocrepinella alba*, particularly the Scandinavian specimens illustrated by Höglund (1948) and Nyholm (1956) which lack the secondary aperture at the proximal end of the test reported by Heron-Allen & Earland (1932). However, the apertural features of the two species are different; *C. laevis* has a simple aperture at the end of a short neck-like extension of the test, whereas in *H. alba* the aperture is constricted by a collar-like structure (Nyholm, 1956). The test of *H. alba* is also larger (up to 2.8 mm long) than that of *C. laevis* and has a much more variable outline. The new species never exhibits the nipple-like projection sometimes developed at the proximal end of *H. alba* (Höglund, 1948). These morphological differences are supported by our molecular evidence which indicates that *C. laevis* and *H. alba* are not closely related

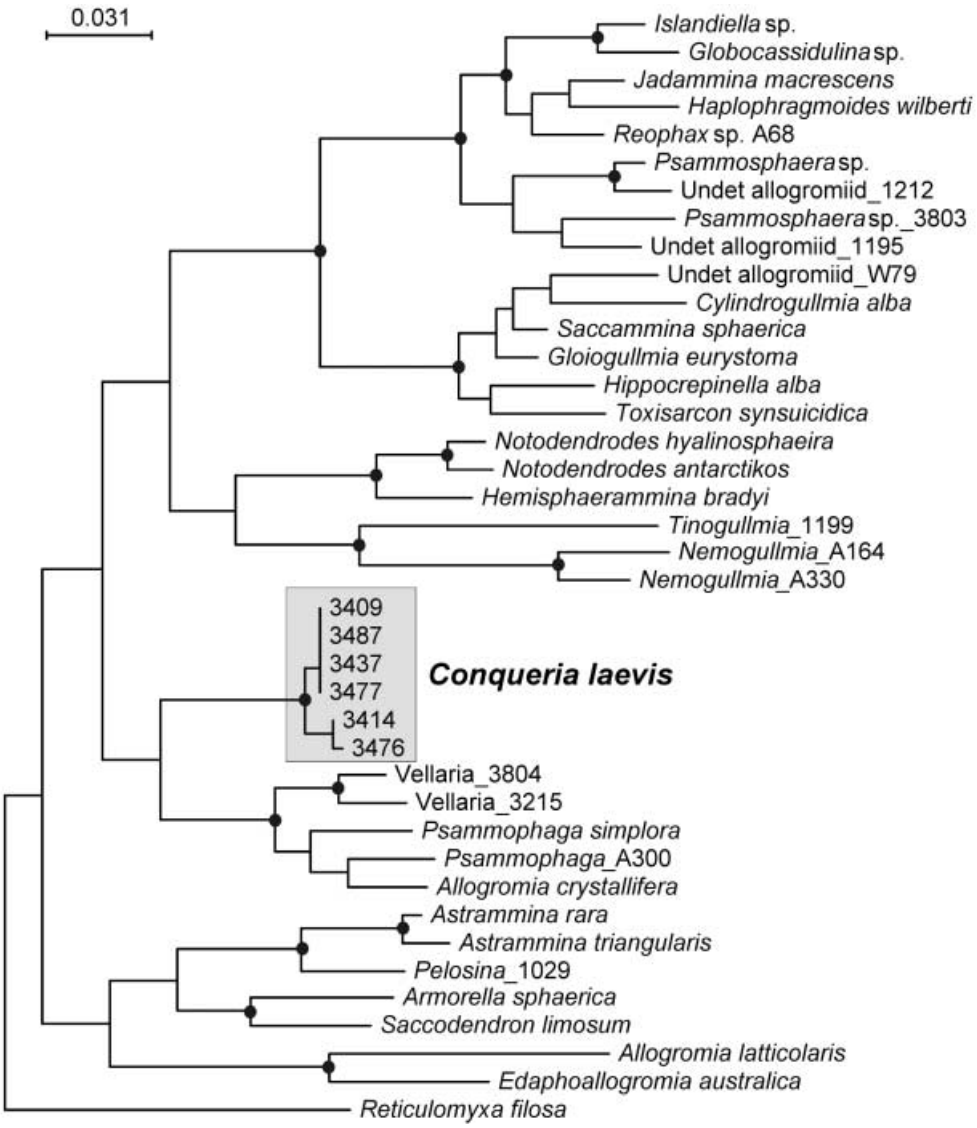

Figure 5. Phylogenetic position of *Conqueria laevis* gen. and sp. nov. inferred from partial SSU rRNA gene sequences (657 sites), using the ML method.

(Figure 5). *Hippocrepina cylindrica* Höglund, 1948 is another similar species. However, the test is less elongate than that of *C. laevis*, the aperture is located at the end of a funnel-shaped structure rather than a tubular extension and the test wall is much thicker (up to 50 μm) and composed of mica particles which impart a silvery lustre to the surface (Höglund, 1948).

DISCUSSION

Based on morphological evidence, we suggest that members of the genus *Conqueria* may be widely distributed in the deep sea and at shallower high latitude sites. In particular, very similar morphospecies are known from 49 m water depth in Kongsfjord and other fjords around Svalbard (J. Gooday & A. Sabbatini, unpublished data) and from much greater depths (4850 m) on the Porcupine Abyssal Plain, north-eastern Atlantic (A.J. Gooday, unpublished data). Whether these morphospecies are also closely related genetically remains to be tested. Nevertheless, the presence of similar morphospecies in the Weddell Sea and Porcupine Abyssal Plains supports other evidence (Cornelius & Gooday, in preparation) for faunal connections between southern and northern hemisphere abyssal plains.

According to molecular data, *Conqueria* constitutes a new lineage of monothalamous (single-chambered) foraminiferans (Figure 5). Traditionally, this group has been divided into two orders distinguished by the character of the wall: the Allogromiida with an organic-walled test and Astrorhizida with an agglutinated test (Loeblich & Tappan 1989). Molecular phylogenetic studies have demonstrated that many lineages of monothalamous Foraminifera include both organic-walled and agglutinated tests, indicating that the composition and structure of the wall is not a good taxonomic marker for evolutionary relationships within this group (Pawlowski et al., 2002b, 2003). The phylogenetic position of *Conqueria*, which has an agglutinated test wall, as a sister group to the organic-walled genus *Vellaria* and the finely agglutinated genus *Psammophaga* (Figure 5), supports this conclusion. Moreover, molecular analyses indicate that *Hippocrepinella alba* and *C. laevis*, which resemble each other in test morphology and wall structure, belong to

different phylogenetic clades. Thus there appears to be considerable potential for morphological convergence between members of different monothalamous foraminiferal clades, as well as considerable morphological variability within particular clades (Pawlowski et al., 2002b).

Our study contributes to the growing appreciation that the diversity of monothalamous foraminiferans is much higher than previously believed. This group has been largely ignored by micropalaeontologists because they are difficult to identify and rarely fossilize (Tappan & Loeblich, 1988). However, several recent morphology-based studies have revealed the high abundance and diversity of monothalamous foraminiferans, particularly in deep-sea and higher-latitude coastal environments (Gooday et al., 1996, 1998; Korsun & Hald, 2000; Gooday, 2002). The high genetic diversity of the group was also demonstrated by a molecular study at a shallow-water Antarctic site (Pawlowski et al., 2002a). A particularly important outcome of recent molecular research has been the recognition of a large number of higher-level phylogenetic lineages which are likely to have evolved in the late Neoproterozoic, well before the first known fossil foraminiferans (Pawlowski et al., 2003). *Conqueria* represents a hitherto unknown lineage within this evolutionary radiation. Based on local molecular clocks calibrated with reference to the Carboniferous diversification of multilocular foraminifera (Pawlowski et al., 2003), we conservatively estimate the origin of the lineage to between 600 and 800 MYA. It is impossible to date it more accurately because of the heterogeneity of evolutionary rates in the foraminiferan SSU rRNA genes (Pawlowski et al., 1997). Interestingly, *Conqueria* is one of a few monothalamous deep-sea species (together with *Vanhoeffenella*) which branches in the basal part of the tree, i.e. it appears to be much older in evolutionary terms than most of deep-sea foraminiferans. The question whether this species is a relict of an ancient deep-sea meiofauna or an example of more recent colonization, will be addressed by studying its phylogenetic relationships with similar shallow-water morphotypes identified in Svalbard fjords.

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REFERENCES

Brandt, A., Broyer, C. de, Gooday, A.J., Hilbig, B. & Thomson, M.R.A., 2003. Introduction to Antarctic benthic deep-sea biodiversity (ANDEEP): colonization history and recent community patterns. *Berichte für Polarforschung*, **470**, 45–49.

Dawson, G.M., 1870. On foraminifera from the Gulf and River St Lawrence. *Canadian Nauralist and Quarterly Journal of Science, Montreal, (New Series)*, **5**, 172–177.

Felsenstein, J., 1985. Confidence limits on phylogenies: an approach using the bootstrap. *Evolution*, **39**, 783–791.

Galtier, N., Gouy, M. & Gautier, C., 1996. SEAVIEW and PHYLO_WIN: two graphic tools for sequence alignment and molecular phylogeny. *Computer Applied Biosciences*, **12**, 543–548.

Gooday, A., 2002. Organic-walled allogromiids: aspects of their occurrence, diversity and ecology in marine habitats. *Journal of Foraminiferal Research*, **32**, 384–399.

Gooday, A.J., Bett, B.J., Shires, R. & Lambshead, P.J.D., 1998. Deep-sea benthic foraminiferal diversity in the NE Atlantic and NW Arabian sea: a synthesis. *Deep-Sea Research II*, **45**, 165–201.

Gooday, A.J., Bowser, S.S. & Bernhard, J.M., 1996. Benthic foraminiferal assemblages in Explorer's Cove, Antarctica: a shallow water site with deep-sea characteristics. *Progress in Oceanography*, **37**, 219–267.

Gooday, A.J., Levin, L.A., Linke, P. & Heeger, T., 1992. The role of benthic foraminifera in deep-sea food webs and carbon cycling. In *Deep-sea food chains and the global carbon cycle* (ed. G.T. Rowe and V. Pariente), pp. 63–91. London: Kluwer Academic Publishers.

Heron-Allen, A. & Earland, E., 1932. Some new foraminifera from the South Atlantic IV. Four new genera from South Georgia. *Journal of the Royal Microscopical Society*, **52**, 253–261.

Höglund, H., 1948. Foraminifera in the Gullmar Fjord and the Skagerak. *Zoologiska Bidrag från Uppsala*, **26**, 1–328.

Howe, J.A., 2003. Recent depositional environments of the north western Weddell Sea and South Sandwich Trench. *Berichte für Polarforschung*, **470**, 124–127.

Korsun, S. & Hald, M., 2000, Seasonal dynamics of benthic foraminifera in a glacially fed fjord of Svalbard, European Arctic. *Journal of Foraminiferal Research*, **30**, 251–271.

Loeblich, A.R. & Tappan, H., 1987. *Foraminiferal genera and their classification*. New York: Van Nostrand Reinhold Company.

Loeblich, A.J.R. & Tappan, H., 1989. Implication of wall composition and structure in agglutinated foraminiferas. *Journal of Paleontology*, **63**, 769–777.

Nyholm, K.-G., 1956. Observations on the monothalamous *Hippocrepinella alba* Heron-Allen and Earland. *Zoologiska Bidrag från Uppsala*, **30**, 475–484.

Olsen, G.J., Matsuda, H., Hagstrom, R. & Overbeek, R., 1994. FastDNAml: a tool for construction of phylogenetic trees of DNA sequences using maximum likelihood. *Computer Applied Biosciences*, **10**, 41–48.

Pawlowski, J., 2000. Introduction to the molecular systematics of foraminifera. *Micropaleontology*, **46**, Supplement 1, 1–12.

Pawlowski, J., Bolivar, I., Fahrni, J., De Vargas, C., Gouy, M. & Zaninetti, L., 1997. Extreme differences in rates of molecular evolution of foraminifera revealed by comparison of ribosomal DNA sequences and the fossil record. *Molecular Biology and Evolution*, **14**, 498–505.

Pawlowski, J., Fahrni, J.F., Brykczynska, U., Habura, A. & Bowser, S.S., 2002a. Molecular data reveal high taxonomic diversity of allogromiid Foraminifera in Explorers Cove (McMurdo Sound, Antarctica). *Polar Biology*, **25**, 96–105

Pawlowski, J., Holzmann, M., Berney, C., Fahrni, J., Cedhagen, T. & Bowser, S.S., 2002b. Phylogeny of allogromiid Foraminifera inferred from SSU rRNA gene sequences. *Journal of Foraminiferal Research*, **32**, 334–343.

Pawlowski, J., Holzmann, M., Berney,C., Fahrni, J., Gooday, A.J., Cedhagen, T., Habura, A. & Bowser, S.S., 2003. The evolution of early Foraminifera. *Proceedings of the National Academy of Sciences of the United States of America*, **100**, 11494–11498.

Tappan, H. & Loeblich, A.R., 1988. Foraminiferal evolution, diversification, and extinction. *Journal of Paleontology*, **62**, 695–714.

Tendal, O.S. & Hessler, R.R., 1977. An introduction to the biology and systematics of Komokiacea. *Galathea Report*, **14**, 165–194.

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