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Tinogullmia lukyanovae sp. nov.—a monothalamous, organic-walled foraminiferan from the coastal Black Sea

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Recent studies indicate that monothalamous (single-chambered) foraminifera are an important component of meiobenthic communities in the brackish, coastal waters of the Black Sea. The dominant taxa include *Psammophaga* and *Vellaria*, both of which are common in estuarine settings in other parts of the world. Here, we describe *Tinogullmia lukyanovae* sp. nov. from the Crimean and Caucasus regions of the Black Sea. The new species is several hundreds of microns in size. It is characterized by a more or less symmetrical, spindle-shaped to cylindrical, organic-walled test with two terminal apertural structures, usually in the form of rounded or more elongate, nipple-like projections. The protoplasm is finely granular and a nucleus is not visible. The new species typically occurs in well-oxygenated water at depths of a few tens of metres or less. A specimen from a 250-m deep site off the Caucasus coast, tentatively assigned to *T. lukyanovae* sp. nov., is of particular interest because it occurs at a depth where the bottom water is anoxic and sulphidic.

INTRODUCTION

Monothalamous (single chambered) foraminifera include species in which the test wall is composed either of organic material or incorporates agglutinated particles. In some cases, the test wall is soft and flexible. A general lack of distinctive morphological features makes the systematics of monothalamous foraminifera problematical. Most recent classifications are based on test characteristics (e.g. Loeblich & Tappan, 1987; Sen Gupta, 1999; Lee et al., 2000) and place organic-walled species in the Order Allogromiida and those with agglutinated walls in the Order Astrorhiziida. Molecular studies, however, suggest that classifications based on wall type and test morphology are unreliable (Bowser et al., 1995; Pawlowski et al., 2003). In particular, phylogenetic clades derived from small subunit ribosomal RNA (SSUrRNA) gene sequences often include a mixture of agglutinated and organic-walled species, suggesting that there is no fundamental distinction between allogromiids and astrorhiziids (Pawlowski et al., 2002a, 2002b, 2003).

Biologists often overlook smaller, soft-walled monothalamous foraminifera because they are unfamiliar and difficult to identify. At the same time, they have almost no fossilization potential and are therefore of little interest to geologists. As a result, relatively few species have been described scientifically. The recent European Register of Marine Species checklist of described marine species from around the European coastline (http://erms.biol.soton.ac.uk) includes 1122 benthic foraminiferal species of which only 31 are organic-walled allogromiids. Yet monothalamous foraminifera are often common and diverse in European coastal and deeper-water habitats (Gooday, 2002), making this a key group to target in biodiversity surveys.

In order to advance our knowledge of these poorly-known taxa, it is important to study samples from different parts of the world. Organic-walled allogromiids are common in the coastal waters of the Black Sea. Two species assigned to the genus *Lagynis*, regarded as an allogromiid foraminiferan by Loeblich & Tappan (1987), were either reported (*L. baltica*) or described (*L. pontica*) from the Black Sea (Golemansky, 1999 and references therein). Most of our information, however, comes from recent meiofaunal studies conducted round the Crimean coast (reviewed by Sergeeva, 2003; Revkov & Sergeeva, 2004). Monothalamous foraminifera were first mentioned by Sergeeva & Kolesnikova (1996). Anikeeva & Sergeeva (2001) and Anikeeva (2003, 2005) briefly discussed the occurrence of *Psammophaga* and other monothalamous taxa off the Crimean coast. Sergeeva & Anikeeva (2004) reported another genus, *Vellaria,* from the Sevastopol area of the Crimea. Finally, a species of *Tinogullmia*, which is closely similar to the type species, *T. hyalina* Nyholm, 1954, occurs at a site south-west of the Crimean Peninsula (Sergeeva et al., 2005). In general, however, Black Sea monothalamous foraminifera are poorly documented and few species are described, particularly when compared to calcareous and other hard-shelled species (Yanko & Vorobjeva, 1990, 1991).

The main aim of the present paper is to describe a new species of the organic-walled genus *Tinogullmia*, collected from coastal sites off the Crimean Peninsula and in the eastern part of the Black Sea off the Caucasus coast. We have focused on this relatively uncommon species because of its distinctive morphology. In describing it, we wish to draw attention to the occurrence of monothalamous foraminifera in coastal waters around the Crimean Peninsula as well as in deeper, anoxic layers of the Black Sea.

Table 1. *Locality information. N=number of specimens of the new species. ND=no data. The Caucasus samples were taken during the 102nd cruise of research vessel 'Akademik Kovalevskij'.*

Locality (year of collection)	Latitude (°N)	Longitude (°E)	Depth (m)	Sampler	Bottom-water oxygen	Salinity	Sediment	N
Crimean coast								
Kazach'ya Bay Station 4 (2003)	44°34.40′	33°24.25′	3.5	Petersen grab	6.4 ml l^{-1}	18.0‰	Quartz sand with shell debris and plant detritus	2 (types)
Kazach'ya Bay Station 5A (2003)	44°34.42′	33°24.27′	12	Petersen grab	5.9 ml l^{-1}	18.0‰	ND	2
Sevastopol Bay Station 2	44°38.0′	33°35.0′	12	Petersen grab	ND	17.4‰	Fine silt with plant detritus	2
Caucasus coast								
Station 54 (1986)	44°00.5′	39°08.6′	30	Okean grab	6.6 ml l^{-1}	18.3‰	Silty sand with shell debris	3
Station 62 (1986)	43°59.0′	39°05.7′	110	Okean grab	2.7 ml l^{-1}	19.6‰	Dark silt with phaseolinic silt	1
Station 66 (1986)	43°58.2′	39°05.0′	250	Okean grab	0 ml l^{-1}	19.8‰	Fine sand with shell debris	1

ENVIRONMENTAL CHARACTERISTICS

The Black Sea is the largest permanently anoxic basin, as well as the largest inland water body, in the world. Its greatest depth is 2245 m. A broad shelf is developed to the west of the Crimean Peninsula, but in other areas, including off the Caucasus coast, the shelf is generally very narrow (~10 km off Tuapse). Anoxia is maintained by a strongly developed density stratification (Zenkevitch, 1963; Tolmazin, 1985). The well-oxygenated surface waters (upper 100–150 m) are of low salinity due to the discharge of large volumes of fresh water from several major central and eastern European rivers, notably the Danube, Dnieper and Dniester, which empty onto the wide north-eastern shelf. According to Zenkevitch (1963, figure 186 therein), the salinity of the upper water layers in our study areas ranges from 17.6–18.2‰ (west of the Crimean Peninsula) to 17.4–17.8‰ (off the Caucasus coast). Very little mixing occurs between this upper layer and the denser, more saline, anoxic, water masses below. This leads to the development of an intense bottom-water oxygen gradient from the well-oxygenated surface waters (upper 70–80 m) to anoxic and highly sulphidic conditions below 150–200 m water depth (Zenkevitch, 1963; Tolmazin, 1985; Luth & Luth, 1997).

MATERIALS AND METHODS

Samples were collected from the Crimean and Caucasus regions of the Black Sea using either a Petersen grab (surface area 0.025 m^2) or an Okean grab (surface area 0.25 m^2). Station data are summarized in Table 1; station positions are shown in Figure 1. Subsamples (18.1 cm^2, 0–5 cm depth) for meiobenthic studies were taken using an aluminium tube ('meiobox'). The entire subsample was preserved in 75% alcohol. In the laboratory, the subsamples were washed through sieves of 1 mm and 64 μm mesh size. The 64–1000-μm fraction was stained for a period of 6–24 h with rose Bengal and examined under a binocular microscope. All stained foraminifera were picked out with a glass pipette and placed in a cavity slide with a mixture of glycerol (50%) and water (50%).

Photographs were taken either using a Nikon CoolPix 4500 digital camera fitted to an Olympus BX51 compound microscope equipped with phase contrast optics, or on black and white film using an Olympus BH2 photomicroscope equipped with Nomarski interference optics. In the latter case, the negatives were scanned into a computer.

SYSTEMATICS

Studies beginning in the 19th Century, based on light microscopy, revealed a diversity of monothalamous foraminiferal species (e.g. Rhumbler, 1904). Recent molecular genetic data, however, suggest that test morphology alone does not provide an adequate foundation for the classification of these taxa. Species placed in the same family based on morphological criteria may be unrelated according to analyses of SSU rRNA genes sequences (Pawlowski et al., 2003). Molecular data for the new species are not available and so we base our description on test characteristics and the general appearance of the cytoplasm. The morphology-based suprageneric classification given below is based on Loeblich & Tappan (1987), Sen Gupta (1999) and Lee et al. (2000), although for reasons mentioned above, we recognize that some of the higher taxa may not represent natural groupings.

Class FORAMINIFERA Eichwald, 1835
Order ALLOGROMIIDA Loeblich & Tappan, 1961
Family ALLOGROMIIDAE Rhumbler, 1904
Subfamily SHEPHEARDELLINAE Loeblich & Tappan, 1984

Genus *Tinogullmia* Nyholm, 1954

Type species: *Tinogullmia hyalina* Nyholm, 1954

Tinogullmia lukyanovae Gooday, Anikeeva, Sergeeva sp. nov.
(Figures 2–4)

Derivation of name

In honour of Mrs Ludmilla F. Lukyanova, Department of Shelf Ecosystems, Institute of Biology of the Southern

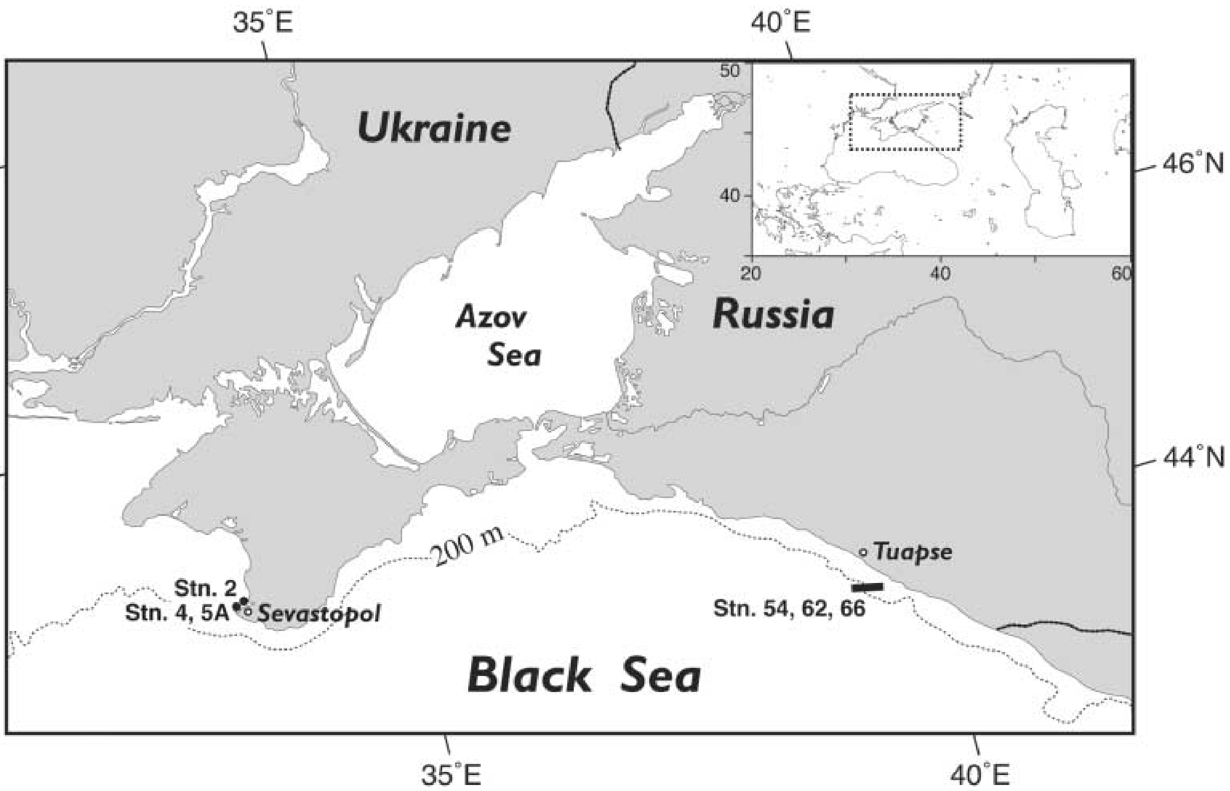

Figure 1. Map showing location of sampling sites in the northern and eastern Black Sea.

Seas, Sevastopol, Ukraine, who has been responsible for sorting many allogromiid foraminifera from Black Sea samples.

Type material

Holotype: complete specimen preserved in 4% formalin, illustrated in Figure 2A–D (Station 4, Kazach'ya Bay, Crimean coast of Black Sea; 44°34.40′N 33°24.25′E; 3.5 m water depth; quartz sand with shell debris and plant detritus). [Micropalaeontology Unit, Palaeontology Department, Natural History Museum, London, UK, registration no. ZF 5164].

Paratype: complete specimen preserved in 4% formalin, illustrated in Figure 2E–H. [Micropalaeontology Unit, Palaeontology Department, Natural History Museum, London, UK, registration no. ZF 5165].

Comparative material examined

Station 5A in Kazach'ya Bay, Crimean coast of Black Sea (44°34.42′N 33°24.27′E, 12 m water depth), 2 specimens; Station 2 in Sevastopol Bay, Crimean coast of Black Sea (44°38.0′N 33°35.0′E, 12 m water depth), 2 specimens; Station 54, Caucasus coast of Black Sea (44°00.5′N 39°08.6′E, 30 m water depth), 3 specimens. Station 62, Caucasus coast (43°59.0′N 39°05.7′E, 110 m water depth), 1 specimen. Station 66, Caucasus coast (43°58.2′N 39°05.0′E, 250 m water depth), 1 specimen. All specimens are housed in the Department of Shelf Ecosystems, Institute of Biology of the Southern Seas, Sevastopol, Ukraine.

Diagnosis

Test 220–550 μm long and 120–160 μm wide with a more or less circular cross-section; lateral outline rather variable, ranging from oval to spindle-shaped to cylindrical. Identical terminal apertures located at ends of rounded or more elongated, nipple-like projections. Test wall organic, surface smooth but not clearly reflective. Cytoplasm yellowish-brown in colour and finely granular without visible nucleus.

Description of type specimens

The holotype is approximately spindle-shaped with the widest point slightly off-centre and tapering towards the ends. Both ends are produced into nipple-like apertural projections. These are about 25 μm long, but are difficult to measure accurately because they merge smoothly with the rest of the test (for this reason, they are termed projections rather than tubes). Test length including apertural projections 404 μm, length excluding projections approximately 350 μm, width 134 μm; length:width (L:W) ratio is 3.0 (including projections), 2.6 (excluding projections). The paratype is rather more cylindrical in shape than the holotype and the ends are more broadly rounded. It measures 373 μm long (with apertural projections), 336 μm long (without projections) and 134 μm wide; L:W ratio 2.8 (including projections) and 2.5 (excluding projections). The apertural projections are approximately 20 μm and 15 μm long. The cross-section of the test is more or less circular in both specimens.

The test wall appears rather thick under a compound microscope and the surface is almost devoid of attached particles. The unstained cytoplasm is opaque and light brown in colour when specimens are viewed in water under reflected light. A relatively thick strand of cytoplasm occupies the axis of the apertural projections. A nucleus is not visible in either specimen.

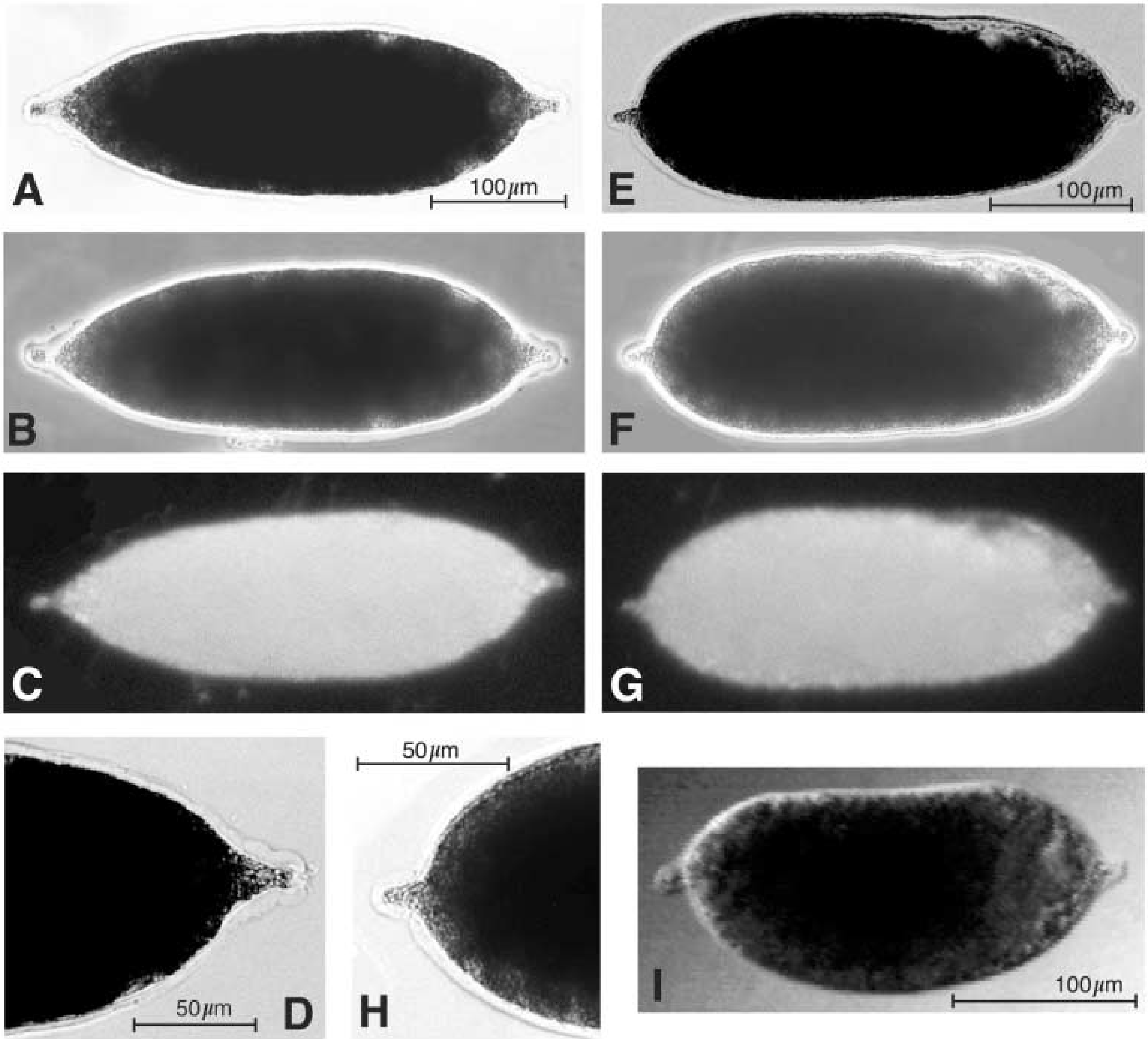

Figure 2. *Tinogullmia lukyanovae* sp. nov. Photographed using Nomarski Interference contrast (A,D,E,H), phase contrast (B,F), bright field (I) and reflected light (C,G). (A–C) Holotype, registration number ZF 5164, from Kazach'ya Bay Station 4, entire test; (D) holotype, apertural projection; (E–G) paratype, registration number ZF 5165, entire test; (H) paratype, apertural projection; and (I) specimen from Kazach'ya Bay Station 5A. All specimens photographed in water in an uncovered cavity slide except for (I) which was photographed in glycerol in an uncovered cavity slide.

Description of other material

A specimen from Station 5A in Kazach'ya Bay, very close to the type locality, has a relatively shorter lateral outline than the types (Figure 2I). One side is almost straight and the other gently curved; the nipple-like apertural projections (about 14 µm long) extend from the broadly rounded ends of the test which has the following dimensions: length (without terminal projections) 230 µm, width 125 µm, L:W ratio 1.8. Two specimens from Station 2 in Sevastopol Bay (illustrated in Figure 3A–E) are mounted under an unsupported cover glass and are therefore somewhat flattened and distorted. They measure 228 µm long and 123 µm wide (L:W ratio 1.85) and 274 µm long and 127 µm wide (L:W ratio 2.15). The apertural projections are rather less prominent than in the type specimens. Specimens from Station 54 (30 m water depth) on the Caucasus margin are generally similar to those from the type locality. The specimen illustrated in Figure 3F–G, also mounted under an unsupported cover glass, has the following dimensions: length 456 µm, width 160 µm, L:W ratio 2.7; the apertural projections are 17 µm long.

Two individuals from deeper water off the Caucasus coast are assigned provisionally to the new species. (i) A specimen from Station 62 (110 m water depth) has a somewhat asymmetrical, spindle-shaped test, 354 µm long and 157 µm wide (L:W ratio 2.2) (Figure 4). The bluntly pointed ends are drawn out into short apertural projections, each about 12 µm long. The wall has some adhering debris. (ii) A specimen from Station 66 (250 m) is relatively slender and more or less cylindrical in form, measuring 410 µm long and 125 µm wide (L:W ratio 3.2). The apertural structures, rather than being nipple-like projections, are clearly tubular, about 28 µm long, and have fine, axial threads of cytoplasm. The test wall is thin with no adhering particles and is separated from the cytoplasm by a distinct space. It is not clear whether this space is a natural feature or an artefact. Deep-water tinogullmid

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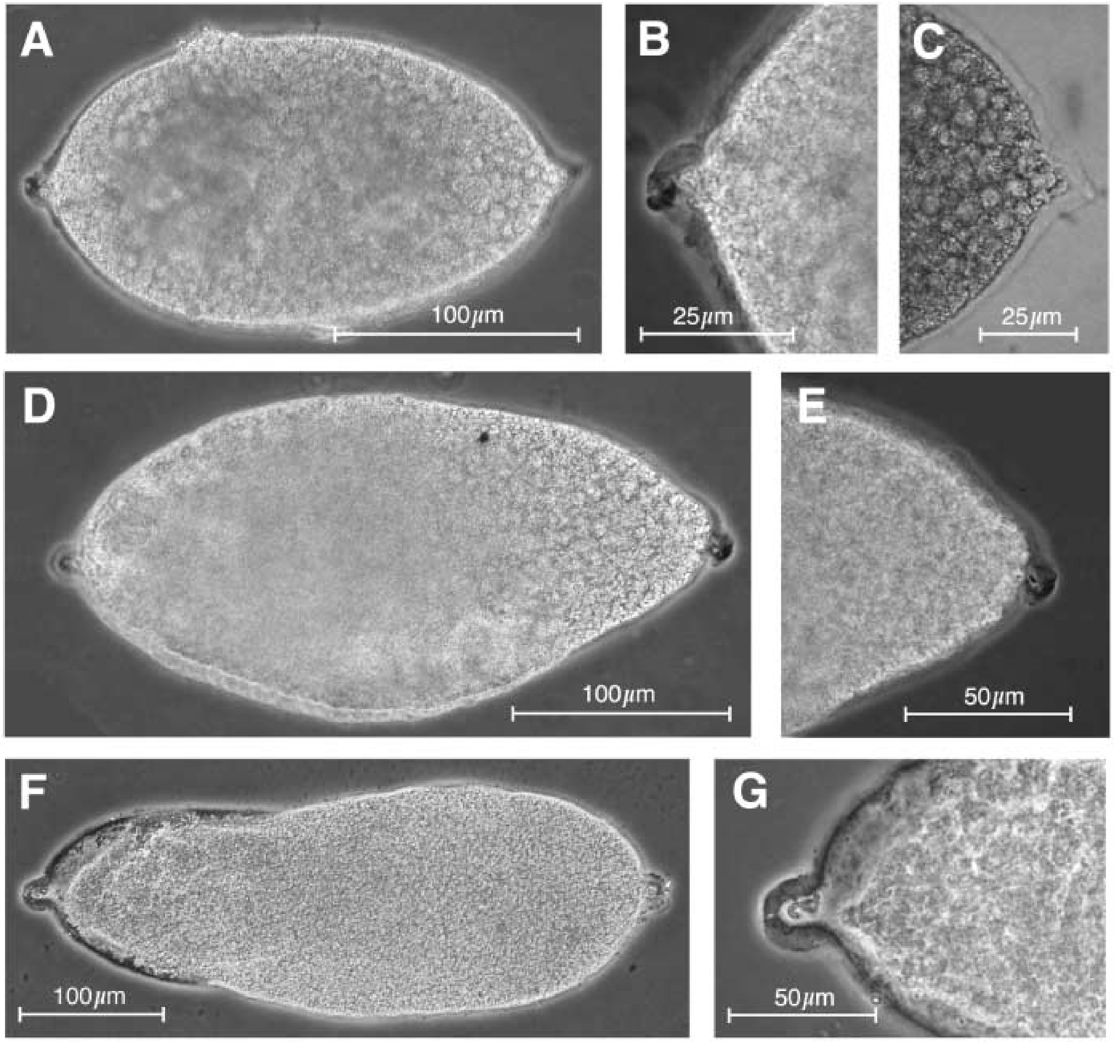

Figure 3. *Tinogullmia lukyanovae* sp. nov.; photographed using phase contrast (A,B,D–G) and Nomarski Interference contrast (C) optics. (A–C) Specimen from Sevastopol Bay, Station 2, complete test and details of terminal apertural structures (note granular nature of cytoplasm in (C)); (D–E) specimen from Sevastopol Bay, Station 2, complete test and details of terminal apertural structures; and (F–G) specimen from Caucasus Station 54, complete test and detail of terminal apertural structure. All specimens were mounted under an unsupported cover glass and are therefore somewhat flattened.

allogromids exhibit a similar space between the cytoplasm and test wall (Gooday, 1990).

Remarks

The new species resembles members of Loeblich & Tappan's (1987) allogromiid subfamily Shepheardellinae in the presence of two terminal apertures. It is much smaller than *Shepheardella taeniformis* Siddall, 1880, which is typically >1 mm in length and has a thread-like morphology. It is also much smaller than *Phainogullmia aurata* Nyholm, 1955 and lacks the reflective, agglutinated surface of this species. The new species most closely resembles *Tinogullmia*, although we place it in this genus with some hesitation. Compared to the type species *Tinogullmia hyalina* Nyholm, 1954 from the Gullmar Fjord on the Swedish west coast, and a very similar form from 78 m water depth off the Crimean Peninsula (Sergeeva et al., 2005), the test is less elongate and straight rather than curved. The surface of the wall is also not shiny and hyaline as in *T. hyalina*. In general form, *T. lukyanovae* resembles the deep-sea species *T. riemanni* Gooday, 1990, but the test is more nearly symmetrical about the long axis. A characteristic feature of both *T. hyalina* and *T. riemanni* is the presence of terminal, aperture-bearing tubes with a central thread of cytoplasm. In *T. lukyanovae*, the terminal structures are best described as elongate, nipple-like projections with a central core of cytoplasm.

The available material of *T. lukyanovae* exhibits some morphological variability. For example, the lateral outline of specimens from Kazach'ya Bay ranges from spindle-shaped to oval (Figure 2). Nevertheless, we feel confident that the Crimean (Kazach'ya Bay and Sevastopol Bay) material, and the specimens from Station 54 off the Caucasus coast, represent the same morphospecies. The specimen from Caucasus Station 62 (Figure 4) is also similar to those from the Crimean coast, except that the test wall appears to be thinner and the terminal apertural structures are rather shorter than in the types. The individual from the deepest Caucasus site (Station 66), however, is rather different and possibly represents a distinct species. It has relatively long, tubular apertural structures, each with an axial thread of cytoplasm resembling those

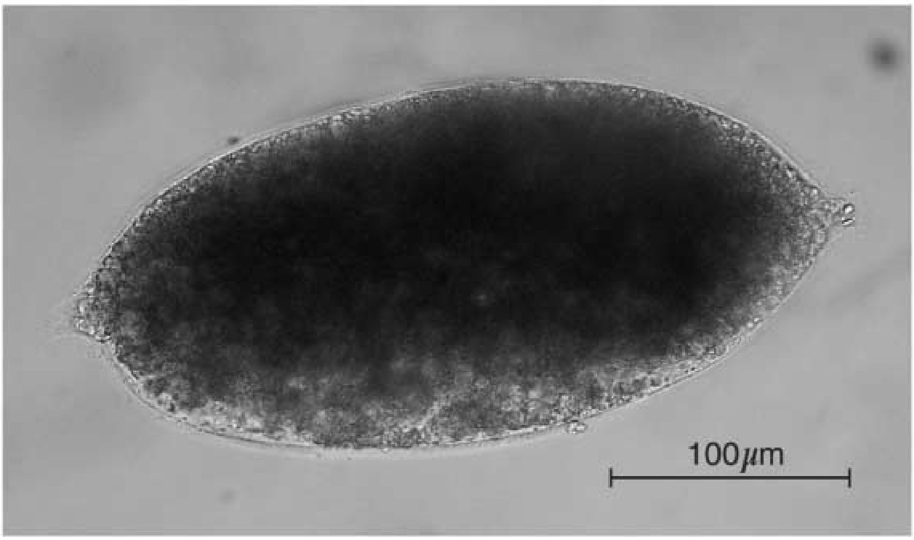

Figure 4. *Tinogullmia lukyanovae* sp. nov., specimen from Caucasus Station 62 photographed using bright field illumination.

developed in *Tinogullmia hyalina*. Problems in determining the morphological limits of species within the genus *Tinogullmia* were also apparent in the case of *T. riemanni*. Individuals of this deep-water species exhibit variation in the shape of the test (symmetrical to asymmetrical) and in the length and position of the apertural structures (Gooday, 1990). It is possible that more than one species is represented among the specimens assigned to *T. riemanni* by Gooday (1990).

DISCUSSION

Monothalamous foraminifera are abundant in coastal Black Sea habitats where they reach average densities of hundreds of individuals per 10 cm^2 (Sergeeva & Anikeeva, 2004). According to Revkov & Sergeeva (2004), the group is represented around the Crimean Peninsula by 20 species. The low salinity of the coastal waters of the north-eastern Black Sea (Zenkevitch, 1963) is reflected in the taxonomic composition of the monothalamous assemblages, which are dominated by species of *Vellaria* (Sergeeva & Anikeeva, 2004) and *Psammophaga* (Revkov & Sergeeva, 2004; Anikeeva, 2005). In other parts of the world, these genera are characteristic of estuarine and intertidal environments (Gooday & Fernando, 1992; Gooday, 2002; Larkin & Gooday, 2004). For example, a *Psammophaga*-like saccamminid species is abundant in the inner part of the Ob estuary, western Siberia (Korsun, 1999 and in Gooday, 2002) and *Vellaria* was first described from the Vellar estuary, south-eastern India (Gooday & Fernando, 1992).

Like *Psammophaga* and *Vellaria*, the new species is an inhabitant of brackish coastal waters. However, the specimens that we assign to *T. lukyanovae* also occur over a considerable range of water depths, from 3.5 m at the type locality off the Crimean coast to 250 m at Station 66 on the Caucasus margin. At our sampling localities, bottom-water oxygen concentrations approached or exceeded 6 $ml\,l^{-1}$ at depths of 30 m and above (Crimean and Caucasus margins), declining to 2.7 $ml\,l^{-1}$ at 110 m and 0 $ml\,l^{-1}$ at 250 m depth (Caucasus margin) (Table 1). The occurrence of a single specimen resembling *Tinogullmia lukyanovae* sp. nov. at 250 m depth, where the bottom water should be anoxic and sulphidic, is remarkable, particularly since organic-walled, monothalamous foraminifera are rarely reported in dysoxic settings (Bernhard & Sen Gupta, 1999). Along transects off the Turkish and Romanian coasts, metazoan meiofauna declined dramatically in abundance below 150 m water depth (Luth & Luth, 1997).

It is possible that this single specimen originated from shallow, well-oxygenated waters and was transported to our deepest station by a process such as the downslope movement of sediment. However, this was not the only monothalamous foraminiferan species to occur at the 250-m site. Station 66 also yielded ten individuals of the saccamminid *Psammophaga simplora* and a variety of other monothalamous foraminifera have been found at similar depths in other parts of the Black Sea (Anikeeva & Sergeeva, unpublished observations). 'Soft-shelled' (monothalamous) foraminifera are common at 260 m water depth off the south-western Crimean coast (Revkov & Sergeeva, 2004). Bernhard et al. (2005) recently discovered saccamminids living in anoxic, sulphidic sediments in the Santa Barbara Basin off the coast of California. The occurrence of a tinogullmid foraminiferan at Station 66 is also consistent with evidence that the deep Black Sea is not as lifeless as was once believed. Recent studies have revealed the existence of a diverse assemblage of meiofauna, including nematodes, acari, harpacticoid copepods and other crustaceans at depths of 400 m to 2250 m (Sergeeva, 2003, 2004). Some species appear to be endemic to anoxic parts of the Black Sea while others are more typical of shelf environments. The occurrence in these hostile environments of monothalamous foraminifera, whether transported or indigenous, merits further study.

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