



A New Testate Amoeba, *Matsakision ogawaraensis* sp. nov. (Silicofilosea: Incertae sedis Euglyphida) from Lake Shore Sand of Northern Japan

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Abstract. A new species of a testate amoeba, *Matsakision ogawaraensis* sp. nov., is described from Ogawara pond, Aomori Prefecture, Japan. This is the third species in the genus *Matsakision. Matsakision ogawaraensis* sp. nov. is distinguished from two other species by the specific form of the shell in apertural view, which is triangular (the shells of other species are circular or elliptic). In addition, it has a chitinous lip around the aperture. For the first time, long needle-like pseudopodia have been observed.

Keywords: testate amoeba, Matsakision, Rhizaria, lake, Japan

INTRODUCTION

Testate amoebae from terrestrial habitats in Japan are poorly investigated (Bobrov et al. 2012; Shimano

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et al. 2014). Up to now, 45 species and subspecific taxa belonging to 21 genera and 14 families were recorded. Most of them are associated to oligotrophic and acid *Sphagnum* habitats (Shimano *et al.* 2017).

Testate amoebae are a polyphyletic group of at least three phylogenetic clades: Euglyphids, Amphitremids, and Arcellinids (Kosakyan *et al.* 2016). The genus *Matsakision* was Incerta sedis of Cercozoa according to the most recent molecular reconstructions (Kosakyan *et al.* 2016) or Incerta sedis of Euglyphids (Adl *et al.* 2012). This genus was proposed by Bonnet (1967) encom-

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passing two species: *Matsakision cassagnaui* (Bonnet, 1967) described from Greece, and *Matsakision radixicola* (Chardez, 1974) from Belgium. Hence, *Matsakision* is considered as a Laurasian endemic (Foissner 2006).

We found an unknown species of testate amoeba from the littoral habitat of Ogawara pond, Aomori Prefecture, northern Japan, which we describe as a new species of *Matsakision*. This genus is recorded here for the first time in Japan.

MATERIALS AND METHODS

Eight specimens were collected from shore sand of Lake Ogawara (40°44′00"N, 141°16′52"E) Aomori Prefecture, Japan, which is located in the north of Honshu Island. The type series (n = 5) was deposited as permanent slides with the cell numbers MO-001 to MO-005 in one slide (specimen number, TNS-AL-58972) to the microalgal collection of Department of Botany, National Museum of Nature and Science, Tokyo (TNS), and 3 individuals are used for Scanning Electron Microscopy (SEM).

The light micrographs were taken in 5% formaldehyde solution with a Optiphot I microscope (Nikon Corporation, Tokyo, Japan) at Hosei University by S.S., and SEM micrographs were taken with a JSM-6380LV SEM (JEOL Ltd., Tokyo, Japan) at Miyagi University of Education by Y.A. Shells of specimens fixed with a 5% (v/v) formaldehyde solution were washed several times and put on a sample stage with carbon double-sided tape after gold evaporation.

RESULTS

Matsakision ogawaraensis sp. nov.

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Taxonomy:

Rhizaria Cavalier-Smith, 2002

Cercozoa Cavalier-Smith, 1998, emend. Adl et al., 2005 Imbricatea Cavalier-Smith 2003

Order Euglyphida Copeland, 1956, emend. Cavalier-Smith, 1997

Incertae sedis Cercozoan testate amoebae Genus *Matsakision* Bonnet, 1964 *Matsakision ogawaraensis* sp. nov.

Description: Shell is small, nearly colorless and almost transparent, elongated oval (shell width nearly 0.6–0.9 times as long as shell length), subterminal, and truncated at the oval aperture with a clear neck near

the aperture (Figs. 1–3, Table 1). In ventral view, shell is broadly ovoid and bilaterally symmetrical (Figs. 1A, 2A, 2B, 2D, 3A, 3B, Table 1). In the lateral view (Figs. 1B, 2G, 2H, 3D), shell is trapezoid (shell height is approximately 1/2 of shell length; Table 1) with a pointed apex in the anterior region, although smooth in ventral side (Figs. 1A, 3A, 3B). In apertural (Figs. 1C, 3F) and back (Fig. 3E) view, the shell is triangular. The shell is composed mainly of irregularly arranged elliptical scales (Figs. 2E, 3H). Scales of the aperture edge are regularly arranged (Figs. 2F, 3G). The aperture is narrowly circularly rectangle, bordered by a short collar, surrounded by a chitinous lip (Figs. 1C, 2D, 2F, 3F, Table 1). Apertural width is approximately 1/2 of the total shell width (Table 1). Apertural height is about 0.2–0.3 times as long as the total shell height (Table 1). Colorless needle-like pseudopodia of about 20 µm in length from protoplasmic body are present and protrude through aperture (Fig. 2C).

Measurements: Table 1 shows the detailed morphometric characterization of *Matsakision ogawaraensis* sp. nov. Shell length (SL) 40.4–48.7 μm; shell width (SW) 29.6–40.7 μm; shell height (SH) 21.7–28.1 μm. The apertural height (AH) is 6.0–8.2 μm; the apertural width (AW) is 17.0–24.4 μm.

Type locality: Japan, Honshu island, Aomori Prefecture, Lake Ogawara (40°44'00"N, 141°16'52"E).

Type specimen: The holotype MO-001 (Figs. 2D–F) and 4 paratypes MO-002 (Fig. 2G), MO-003 (Fig. 2H), MO-004 (Fig. S1A), and MO-005 (Fig. S1B;) were deposited as one permanent slide (TNS-AL-58972) in the microalgal collection of the TNS.

Etymology: The new species is named after the sampling site, Lake Ogawara.

Related species – similarities and differences: The new species *Matsakision ogawaraensis* sp. nov. is distinguished from two other species *M. cassagnaui* (Bonnet, 1967) and *M. radixicola* (Chardez, 1974) by the specific form of the shell in apertural view (Fig. 3F), which is triangular, although the shell of the other species is circular or elliptic (Table 2). In comparison with the type species, *M. cassagnaui* (Bonnet, 1967), the new species, *Matsakision ogawaraensis* sp. nov. is flattened in front and behind with an elliptical cross section (Fig. 1B), and dorsoventrally pointed in the lateral view

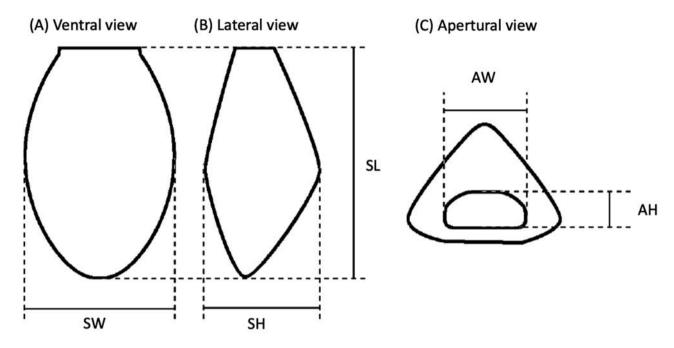


Fig. 1. Outline of Matsakision ogawaraensis sp. nov.: ventral (A), lateral (B), apertural (C) view. SL - shell length, SW - shell width, SH – the longest shell height, AW – apertural width, AH – apertural height.

Table 1. Morphometric characteristics of Matsakision ogawaraensis sp. nov. from Lake Ogawara, Japan. SL - shell length, SW - shell width, SH - the longest shell height, AW - apertural width, AH - apertural height. Features designated in Fig. 1. SW/SL, SH/SL, AW/SW, AH/SH were calculated for in each individual. All measurements in μm.

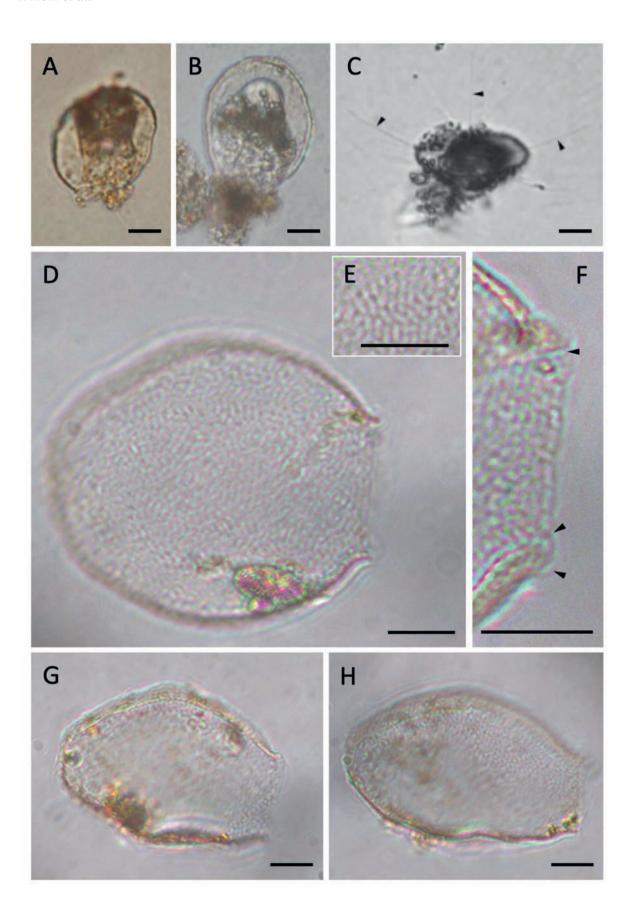
(n = 8)	SL	SW	SH	AH	AW	SW/SL	SH/SL	AW/SW	AH/SH
Mean	44.5	35.8	25.6	7.1	19.3	0.8	0.6	0.5	0.3
Median	44.1	36.0	26.0	7.3	18.8	0.8	0.6	0.5	0.3
Min	40.4	29.6	21.7	6.0	17.0	0.6	0.5	0.5	0.2
Max	48.7	40.7	28.1	8.2	24.4	0.9	0.6	0.6	0.3

(Figs. 1B, 2G, 2H, 3D), and it has a clear light neck and short collar around the aperture (Figs. 1A, 2D, 2F, 3A–C, Table 2). The shell of *Matsakision ogawaraensis* sp. nov. is covered with elliptical scales (Figs. 2D, 2E, 3H), which are observed in two other species (M. cassagnaui and M. radixicola), and parallel to an edge of the aperture, which is observed in M. cassagnaui (Bonnet, 1967) (Table 2). The scales are even (Figs. 2D, 2E, 3H), like them of *M. cassagnaui* (Bonnet 1967), which differ from variable sized scales of M. radixicola (Chardez, 1974) (Table 2). The aperture of Matsakision ogawaraensis sp. nov. is narrowly circular (Figs. 1C, 3F), as the type species (Bonnet 1967), which differs from the circular aperture of *M. radixicola* (Chardez, 1974)

(Table 2). Matsakision ogawaraensis sp. nov. has a chitinous lip (Fig. 2F) and long needle-like pseudopodia (Fig. 2C), absent/not seen in the type species (Bonnet 1967) (Table 2).

DISCUSSION

This is the first record of the genus *Matsakision* in Japan. In the previous reports, all species of the genus were isolated from sphagnum substrates, however, we found *Matsakision ogawaraensis* sp. nov. in shore sand of Lake Ogawara, which is a brackish-water lake facing to the sea. In Japan, numerous testate amoeba species



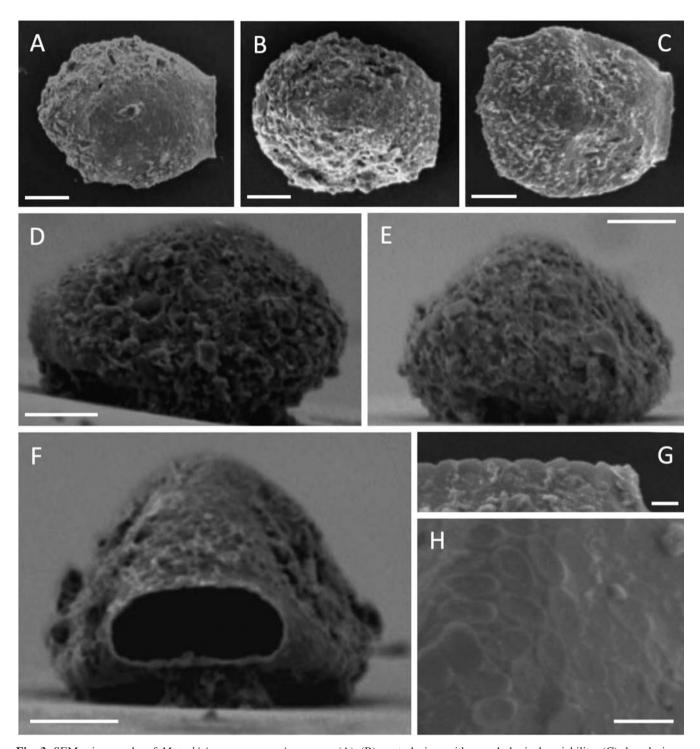


Fig. 3. SEM micrographs of Matsakision ogawaraensis sp. nov. (A)-(B) ventral view with morphological variability. (C) dorsal view. (D) broad lateral view. (E) back view. (F) apertural view. (G) view with emphasis of apertural edge. (H) view with emphasis on shell surface. Scale bars: $A-F-10 \mu m$, $G-1 \mu m$, $H-5 \mu m$.

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Fig. 2. Light micrographs of Matsakision ogawaraensis sp. nov. (A) broad ventral view of a living cell. (B) ventral view of a living cell. (C) back view with emphasis on the pseudopodia. Arrowheads indicate pseudopodia. (D) ventral view of holotype MO-001 (on TNS-AL-58972). (E) view of shell structure when the focal distance is adjusted to the test surface of MO-001. (F) view of MO-001 with emphasis on the apertural edge. Arrowheads indicate a chitinous lip. (G)-(H) lateral view of paratypes MO-002 (on TNS-AL-58972) and MO-003 (on TNS-AL-58972), respectively. Scale bars: 10 μm.

Table 2. Comparison of <i>Matsakision ogawaraensis</i> sp. nov. with two closely related species. All measure	irements in iim
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Characters	Matsakision cassagnaui	M. radixicola	M. ogawaraensis
Lenght	34–38	58-65	40–49
Width	20–24	40–48	30–41
Height	18–22	38–44	22–28
Apertural view	elliptic	circular	triangular
Neck	light	light	light and clear
Collar	short	long	short
Scale	elliptical, even scales and prallel to an edge of the aperture	elliptical, variable sized scales	elliptical even scales and parallel to an edge of the aperture
Aperture	narrowly circular	circular	narrowly circular
Aperture size	$7-8 \times 8-10$	20–24	$6.0-8.2 \times 17.0-24.4$
Chitinous lip	no	unreported	yes
Pseudopodia	unreported	unreported	long needle-like pseudopodia
Sample location	Greece	Belgium	Japan
Data resource	Bonnet 1967	Chardez 1974	present study

were found in a poor fen near the Pacific coast, although these species are, as a rule, associated to oligotrophic/acid sphagnum conditions worldwide (Shimano *et al.* 2017). It is considered that the environmental conditions in soil of a poor fen or lake shore in Japan are similar to these of a sphagnum fen suitable for testate amoebae. In addition, a monospecific testate amoeba, *Porosia paracarinata*, was described from Japan (Bobrov and Kosakyan 2015). Hence, it will be expected that further research in Japan will discover considerably more new or unrecorded testate amoebae.

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Supplementary data

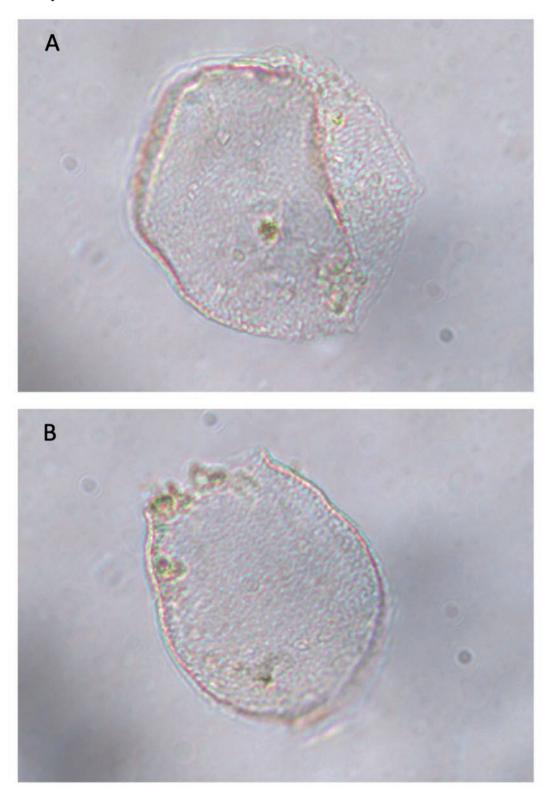


Fig. S1. Light micrograph of *Matsakision ogawaraensis* sp. nov. (A) broad lateral view of paratype MO-004 (on TNS-AL-58972). (B) ventral view of paratype MO-005 (on TNS-AL-58972). Scare bar: 10 μm