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## NAKED LOBOSE AMOEBAE OF THE GENUS *MAYORELLA* (AMOEBOZOA, DISCOSEA, DERMAMOEVIDA) IN UKRAINIAN WATER BODIES

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**Naked Lobose Amoebeae of the Genus *Mayorella* (Amoebozoa, Discosea, Dermamoebida) in Ukrainian Water Bodies.** Patsyuk, M. K. — In Ukrainian water bodies, the genus *Mayorella* Schaeffer, 1926 is represented by ten species: *Mayorella cantabrigiensis* Page, 1983, *Mayorella vespertiliooides* Page, 1983, *Mayorella bigemina* Schaeffer, 1918, *Mayorella leidyi* Bovee, 1970, *Mayorella penardi* Page, 1972, *Mayorella viridis* Leidy, 1874, *Mayorella* sp. (1), *Mayorella* sp. (2), *Mayorella* sp. (3), *Mayorella* sp. (4). The most widely distributed are *M. cantabrigiensis*, *M. vespertiliooides*, *Mayorella* sp. (1), the least observed are *M. leidyi*, *M. penardi*, *M. viridis*, *Mayorella* sp. (4). The distribution of amoebae is influenced by abiotic environmental factors.

**Key words:** naked amoebae, *Mayorella*, Ukrainian water bodies, abiotic factors.

### Introduction

Schaeffer erected the genus *Mayorella* in 1926 with the type species *Mayorella bigemina* (*Amoeba bigemma*) Schaeffer, 1918 (Glotova et al., 2018). In the current system of naked lobose amoebae (Smirnov et al., 2011), this genus belongs to the class Discosea Cavalier-Smith et al., 2004, order Dermamoebida Cavalier-Smith, 2004, family Mayorellidae Schaeffer, 1926. The amoebae have mayorellian morphotype (Smirnov & Goodkov, 1999), generally with undivided fingerlike hyaline subpseudopodia of approximately equal length produced out of hyaline cytoplasm; in some species, subpseudopodia may be temporarily absent during locomotion. The length of moving mayorellian amoebic body exceeds its width. The floating amoeba has acute pseudopodia spreading out of irregularly shaped central cell mass, vesicular nucleus, and one big nucleolus (Goodkov & Buryakov, 1987). *Mayorella*-like amoebae are widely distributed in soils and fresh and sea waters. Based on light microscopy and electron microscopy of the cellular surface, eight species are recognized: fresh-water *Mayorella cantabrigiensis* Page, 1983, *Mayorella vespertiliooides* Page, 1983, *Mayorella penardi* Page, 1972, *Mayorella viridis* Leidy, 1874, and seawater *Mayorella kuwaitensis* Page, 1983, *Mayorella gemmifera* Schaeffer, 1926, *Mayorella dactylifera* Goodkov and Buryakov, 1988, and *Mayorella pussardi* Hollande, Nicolas & Escaig, 1981 (Glotova et al., 2018).



Fig. 1. Sampling localities (Ukraine).

There are reports, based on light and electron microscopy and molecular biology methods, according to which the genus *Mayorella* is richer than the described eight species (Glotova et al., 2018). There is almost no data on naked amoebae fauna of the habitats with differing environmental conditions and of remote locations. That is why it is necessary to sample in distant habitats and compare findings with local faunas (Smirnov et al., 2011). Since naked amoebae remain poorly studied in Ukraine, we investigated the *Mayorella* species isolated from Ukrainian water bodies.

#### Material and methods

Material was collected in 2011–2018 in various water bodies of Ukraine (fig. 1), resulting in 360 samples from 48 locations. We employed modern light microscopy methods, including DIC, to study c. 120 *Mayorella*-like amoebae. It should be noted that a full range of approaches and methods (including ultrastructural and molecular biological) are currently used for exact identification of naked amoebae. Therefore, our taxonomic identification does not pretend to unambiguity. The morphological data are used for the identification of naked amoebae in many faunistic studies.

The samples of water and disturbed bottom sediment were manually collected into glassware of up to 500 ml and transferred to the laboratory. Amoebae were isolated from samples containing the upper layer of bottom soil and a small quantity of bottom water. They were cultured on Petri dishes of 50 mm diameter on non-nutrient agar (Page & Siemensma, 1991) at 15 °C. We used the light microscope Zeiss AxioImager M1 at the Centre for collective usage of scientific equipment “Animalia” of I. I. Schmalhausen Institute of Zoology with differential contrast optics for observations of living cells in water droplets on the slides.

We recorded the temperature, dissolved oxygen and organic matter concentration (by permanganate oxidation) of the studied water bodies during sampling (Stroganov & Buzinova, 1980).

We used the Chekanovsky-Sørensen index to compare the faunistic lists. The clustering was bootstrapped and the multi-dimensional scaling was done in PAST 1.18 (Hammer et al., 2001).

#### Results and discussion

In this study we identified 10 amoeba species of the genus *Mayorella*: *M. cantabrigiensis*, *M. vespertiliooides*, *M. bigemma*, *Mayorella leidyi* Bovee, 1970, *M. penardi*, *M. viridis*, *Mayorella* sp. (1), *Mayorella* sp. (2), *Mayorella* sp. (3), *Mayorella* sp. (4). (fig. 2, table 1) (Patsyuk, 2012, 2013, 2014, 2016 a, b, 2018; Patsyuk & Dovgal, 2012).

We established the occurrence frequency of amoeba, since the absolute quantities could not be calculated for this group. The most common species were *M. cantabrigiensis*

**Table 1. Characters of amoebae of the genus *Mayorella* from Ukrainian water bodies**

No	Amoebae species	Cell length, $\mu$	Cell breadth, $\mu$	Length to breadth ratio, L/B	Nucleus diameter, $\mu$
1.	<i>M. cantabrigiensis</i>	90–110	45–60	2.5–3.6	3.0–3.6
2.	<i>M. vespertiliooides</i>	85–100	50–75	1.3–1.8	10.0–15.0
3.	<i>M. bigemma</i>	110–285	100–85	2.5–3.5	11.0–11.8
4.	<i>M. leidyi</i>	105–185	60–100	2.8–3.0	11.0–11.5
5.	<i>M. penardi</i>	43–50	18–24	3.2–4.0	4.0–6.0
6.	<i>M. viridis</i>	45–55	26–28	1.6	5.0–9.0
7.	<i>Mayorella</i> sp. (1)	100–130	50–70	2.1–3.7	8.0–12.0
8.	<i>Mayorella</i> sp. (2)	50–95	25–40	2.0–2.5	6.5
9.	<i>Mayorella</i> sp. (3)	62–120	30–84	1.8–3.2	5.8–11.0
10.	<i>Mayorella</i> sp. (4)	40–60	15–20	4.8	6.2–6.8

(60.20 %), *M. vespertiliooides* (50.32 %), *Mayorella* sp. (1) (50.03 %), the rarest were *Mayorella* sp. (4) (0.25 %), *M. leidyi* (0.9 %), *M. penardi* (0.5 %), *M. viridis* (1.3 %). The frequency of *M. bigemma* in Ukrainian water bodies was 30.25 %, *Mayorella* sp. (2) 31.01 %, and *Mayorella* sp. (3) 28.32 %. We analyzed the effect of temperature, levels of oxygen and organic matter (by permanganate oxidation) on most common *Mayorella* species: *M. cantabrigiensis* was an eurythermal organism, found at +2 to +26 °C; *M. vespertiliooides* and *Mayorella* sp. (1) were collected mostly in warm season at +16 to +26 °C. *Mayorella* sp. (1) tolerated high oxygen concentrations (4.35 to 31.94 mg/l) and was euryoxicid; *M. cantabrigiensis* and *M. vespertiliooides* were recorded at oxygen levels under 17.84 mg/l meaning that they were stenooxidic. *M. cantabrigiensis* was found at the highest organic matter concentration (50.05 mg O<sub>2</sub>/l), probably the most favorable to its development. *M. vespertiliooides* (24.45 mg O<sub>2</sub>/l) and *Mayorella* sp. (1) (28.53 mg O<sub>2</sub>/l) were sensitive to dissolved organic matter levels.

The distribution of the *Mayorella* species in various types of Ukrainian water bodies was analyzed according to a hydrobiological classification of continental waters (Konstantinov, 1986). Most amoebae were found in rivers (eight species) and riparian basins (seven), only two were discovered in channels, and three were collected in swamps. Four species of amoebae were found in lakes (table 2).

Only *Mayorella* sp. (1) was recorded in all types of water bodies and can be considered eurytopic. Three species were found only in lakes or rivers: *M. penardi* and *M. viridis* were observed only in lakes, and *Mayorella* sp. (2) was collected in rivers because of hydrochemical and trophic specifics of the waters. The other six species were found in water bodies of

**Table 2. Distribution of naked amoebae of the genus *Mayorella* in various water bodies of Ukraine**

No	Amoeba species	Water basin type				
		river	swamp	channel	riparian basin	lake
1.	<i>M. cantabrigiensis</i>	+	+	-	+	-
2.	<i>M. vespertiliooides</i>	+	-	-	+	+
3.	<i>M. bigemma</i>	+	-	-	+	-
4.	<i>M. leidyi</i>	+	-	-	+	-
5.	<i>M. penardi</i>	-	-	-	-	+
6.	<i>M. viridis</i>	-	-	-	-	+
7.	<i>Mayorella</i> sp. (1)	+	+	+	+	+
8.	<i>Mayorella</i> sp. (2)	+	-	-	-	-
9.	<i>Mayorella</i> sp. (3)	+	+	+	+	-
10.	<i>Mayorella</i> sp. (4)	+	-	-	+	-
Total		8	3	2	7	4

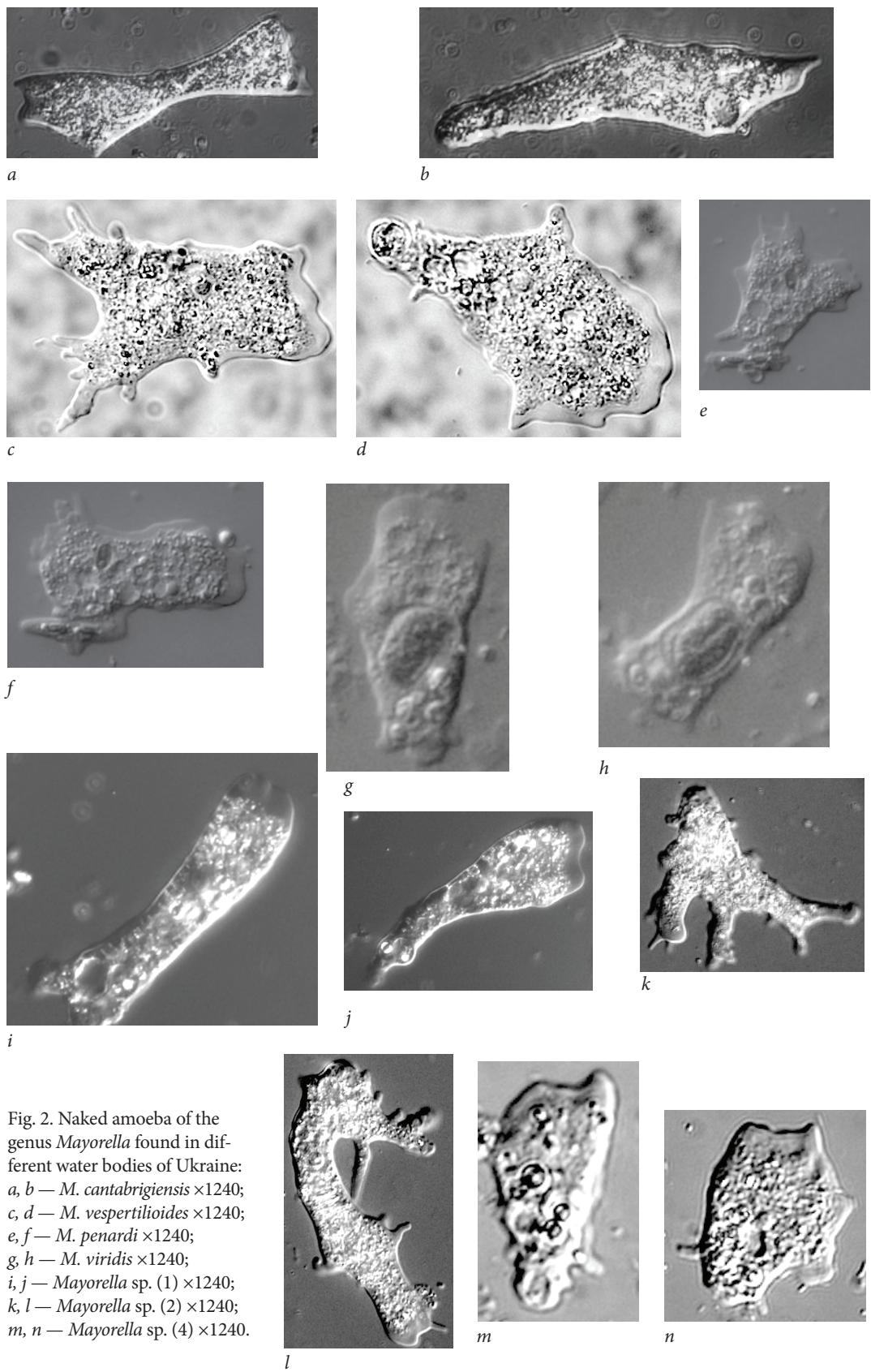


Fig. 2. Naked amoeba of the genus *Mayorella* found in different water bodies of Ukraine:  
 a, b — *M. cantabrigiensis*  $\times 1240$ ;  
 c, d — *M. vespertiliooides*  $\times 1240$ ;  
 e, f — *M. penardi*  $\times 1240$ ;  
 g, h — *M. viridis*  $\times 1240$ ;  
 i, j — *Mayorella* sp. (1)  $\times 1240$ ;  
 k, l — *Mayorella* sp. (2)  $\times 1240$ ;  
 m, n — *Mayorella* sp. (4)  $\times 1240$ .

two or more types, possibly indicating tolerance to wide environmental ranges (table 2).

According to the Chekanovsky-Sørensen index, the most similar assemblages of *Mayorella*-like amoebae were those of rivers and riparian basins (0.93), and the least common assemblages were from swamps and lakes (0.28). Only 0.36 of species were common for lakes and other types of water bodies. According to the cluster analysis (fig. 3), amoebae species lists formed a lake species complex (Shatsky Lakes) and a cluster of riparian basins, rivers, swamps, and channels species complex. The probability of existence of the two clusters was bootstrapped (1000 permutations) and equaled 100 % and 64 %, respectively.

The first cluster of amoebae arose from specific conditions in lakes (mostly sand-silt bottom, calcium-hydrocarbonate water with low minerals levels); and the second cluster ("riparian species complex") formed in basins connected to rivers and thus they are interlinked.

Besides that, *Mayorella* species complexes in Ukrainian waters were influenced by temperature as well as dissolved oxygen and organic matter concentrations. Figure 4 shows that the riparian species complex of *Mayorella*-like amoebae is formed at relatively higher temperatures and higher concentrations of organic matter compared to that of the lakes. The lake complex was defined by low temperature and low organic matter levels. Oxygen levels had a weak effect on the lake and riparian species complexes. This factor will be studied in more detail in our further work.

Therefore, there are 10 amoeba species of the genus *Mayorella* in lakes of Ukraine. In relation to abiotic factors, *M. cantabrigiensis* is eurythermal; *Mayorella* sp. (1) is euryoxicidic; *M. cantabrigiensis* and *M. vespertiliooides* are steno-oxicidic; *M. cantabrigiensis* tolerates high organic matter content, and *M. vespertiliooides* and *Mayorella* sp. (1) do not have tolerance to such conditions. Species

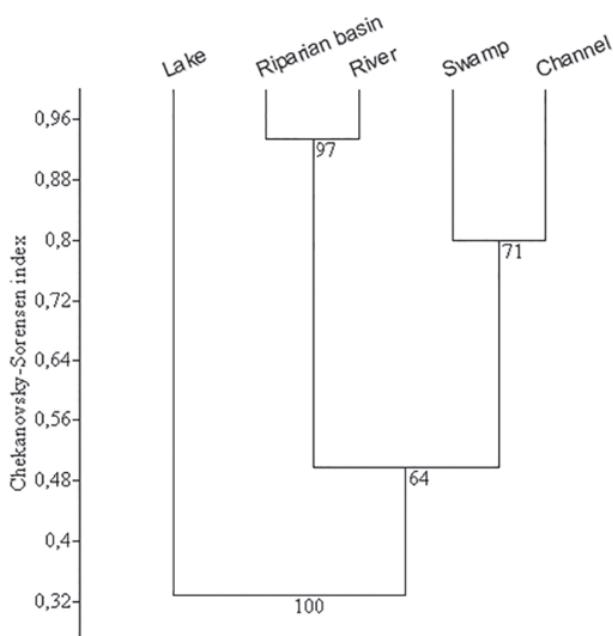


Fig. 3. Similarity of *Mayorella* amoebae complexes in various types of water bodies in Ukraine according to the Chekanovsky-Sørensen's index (numbers in the nodes show the % probabilities of clusters at 1000 bootstrap permutations).

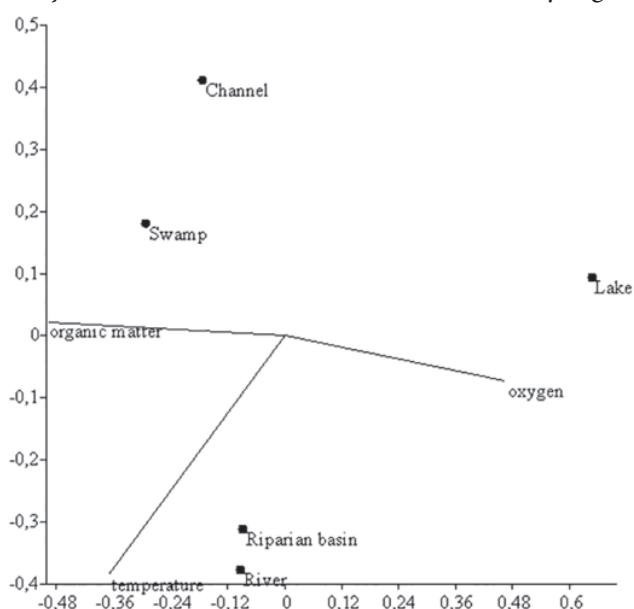


Fig. 4. Ordination of *Mayorella*-like amoebae species complexes in various types of water bodies by environmental factors (according to non-parametric multi-dimensional scaling).

of *Mayorella* form a lake species complex and a riparian complex, both tied to certain abiotic factors.

Notably, *M. cantabrigiensis* was isolated from a remote fresh-water basin in Germany, meaning that this protist can be widely distributed. The Ukrainian fauna of mayorellians is probably not confined to ten species. Small number of species we have so far found can be an artifact of sample transportation, culturing, and species identification. Further research is necessary to determine the distribution of these protists using methods of light and electron microscopy and molecular biology.

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