Fauna and Systematics



UDC 593.121:477.42 SPECIES OF NAKED AMOEBAE (PROTISTA) NEW FOR THE FAUNA OF UKRAINE

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Species of Naked Amoeba (Protista) New for the Fauna of Ukraine. Patsyuk, M. K. — The species *Rhzamoeba* sp., *Thecamoeba quadrilineata* Carter, 1856, *Thecamoeba verrucosa* Ehrenberg, 1838, *Flamella* sp., and *Penardia mutabilis* Cash, 1904 are first reported in the fauna of Ukraine and described based on original material.

Key words: fauna, Zhytomir Polissya, Volyn Polissya, naked amoebae.

Новые находки голых амеб (Protista) фауны Украины. Пацюк М. К. — Представлены сведения об обнаружении новых для фауны Украины голых амеб: *Rhizamoeba* sp., *Thecamoeba quadrilineata* (Carter, 1856), *Thecamoeba verrucosa* (Ehrenberg, 1838), *Flamella* sp., *Penardia mutabilis* Cash, 1904.

Ключевые слова: фауна, Житомирское Полесье, Волынское Полесье, голые амебы.

Introduction

Naked amoebae are unicellular eukaryotic organisms that are capable of amoeboid movement. This name characterizes morphologically and ecologically similar but not related organisms. According to the current system of eukaryotes (Adl et al., 2012), most of the amoeboid organisms are incorporated into three molecular clusters of unclear taxonomic position.

Naked amoebae are among the most important components of aquatic and soil ecosystems. Due to numerous difficulties in species identification, the fauna of these protists remains poorly studied in Ukraine. Previous studies (Patsyuk, 2010, 2011 a, 2011 b, 2012 a, 2012 b, 2014 a, 2014 b; Patcyuk, Dovgal, 2012) recorded 45 species of this group on the territory of Ukraine. Herein, we present morphological characteristics of several new for the Ukrainian fauna species of naked amoebae.

Material and methods

Material was collected in 2009–2014 in water reservoirs of different types in Zhytomir and Volyn Polissya. The samples of water and bottom sediments were gathered in 0.5 L glass containers and transported to laboratory. Amoebae were cultivated in Petri dishes (100 mm diameter) on non-nutrient agar in accordance with F. C. Page (Page, 1988; Page, Siemensma, 1991). Observations on the protozoa and photomicrography were carried out under light microscope Zeiss Axio Imager M1 equipped with differential interference contrast optics at the Center of collective use of scientific equipment "Animalia" (Schmalhausen Institute of Zoology NAS of Ukraine). Species identification was conducted at two stages: firstly their morphotype was determined according to relevant sources (Smirnov, Goodkov, 1999; Brown, Smirnov, 2004; Smirnov, 2008). Then, if possible, taxonomy-based identifying keys (Page, 1988; Page, Siemensma, 1991) and the latest publications on naked amoebae systematics (Smirnov, Goodkov, 1993; Smirnov et al., 2011; Smirnov, 2008) were used.

Results and discussion

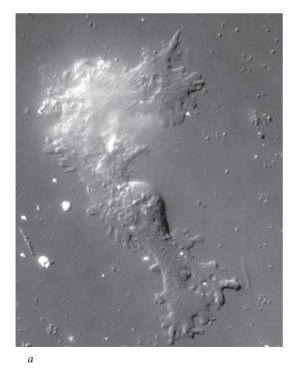
In Ukraine, publications on naked amoebae contain no species characteristics obtained by contemporary research methods. For this reason, we present here descriptions of naked amoebae found in water reservoirs of Zhytomir and Volyn Polissya. All essays are based on original studies, with literature data taken into consideration.

Class TUBULINEA Smirnov, Nassonova, Berney, Fahrni, Bolivar et Pawlowski, 2005 Order LEPTOMYXIDA Pussard et Pons, 1976 Family LEPTOMYXIDAE Pussard et Pons, 1976 Genus Rhizamoeba Page, 1972 Rhizamoeba sp. (fig. 1)

The species belongs to the branched morphotype. Slowly moving amoeba with flattened cell body. Under bright lighting it elongates and forms numerous cytoplasmic branches. Small but discernible contractile vacuoles are present. No crystals were observed in the cellular cytoplasm. Cell length is $110-170 \mu m$, width is $50-80 \mu m$, L/B ratio is 2.5-4.6. Single nucleus, up to $12 \mu m$ in diameter is present.

Distribution: species of the genus *Rhizamoeba* occur in Europe, Australia, North America, Netherlands, Sweden, France (Page, Siemensma, 1991).

Habitats: we found the individuals of the species in bogs (Kharytonivka village, Korostyshevshky District and Martynivka village, Chervonoarmiysky District of Zhytomir Region).





b



Fig. 1. *Rhizamoeba* sp., locomotory forms (a, b, c). ×1240.

Class DISCOSEA Cavalier-Smith, Chao et Oates, 2004 Subclass LONGAMOEBIA Smirnov, Nassonova, Chao et Cavalier-Smith, 2011 Order THECAMOEBIDA Smirnov, Nassonova, Chao et Cavalier-Smith, 2011 Family THECAMOEBIDAE Schaeffer, 1926 Genus Thecamoeba Fromentel, 1874 Thecamoeba quadrilineata (Carter, 1856) (fig. 2)

The species belongs to the striate morphotype. The amoeba is elongated oval. There are several parallel dorsal folds (4–5) that extend along the cell from end to end. Hyaloplasm is undulating dorso-ventral crescent that makes up almost ³/₄ of the cell. There is singular large contractile vacuole of inconstant form. Cytoplasmic crystals are absent. The length of amoeba is $40-82 \mu m$, the width is $20-40 \mu m$, L/B ratio = 1.3-2.2. The diameter of a single nucleus is $8-12 \,\mu\text{m}$.

Remarks: the species can be distinguished from other species of *Thecamoeba* genus by the shape and length of the cell, and vesicular nucleus with central nucleolus. The thin inner nuclear lamina is absent. The nucleus and nucleolus are distorted (Smirnov, Goodkov, 1993; Page, 1988; Page, Siemensma, 1991).

Distribution: T. quadrilineata occurs in Asia, Europe, and North America (Page, Siemensma, 1991).

Habitats: we the species in lakes (near Zoriane and Chornorudka villages, Ruzhynsky District of Zhytomir Region; also near Tsegiv village, Gorokhivsky District and Bubniv village, Lokachivsky District of Volyn Region).

Thecamoeba verrucosa (Ehrenberg, 1838) (fig. 3)

The species belongs to the rugose morphotype. Hyaloplasm forms numerous folds. Uroid is visible in the narrowed end of the amoeba body. Cell length is $150-250 \mu m$, width is 80–120 µm, L/B ratio is 1.5–2.3. Single nucleus is 10–18 µm in diameter.

Remarks: the species can be distinguished from other species by shape and proportions of the cell, also by two tightly adjacent nucleoli in the nucleus (Smirnov, Goodkov, 1993; Page, 1988; Page, Siemensma, 1991).

Distribution: *T. verrucosa* occurs in the most parts of Europe (Page, Siemensma, 1991).

Habitats: we found the species in a lake near Bubniv village of Lokachivsky District, Volyn Region.







Fig. 2. *Thecamoeba quadrilineata*, locomotory forms (*a*, *b*). x1240.

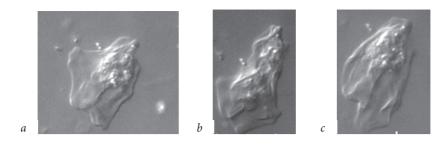


Fig. 3. Thecamoeba verrucosa, locomotory forms (a, b, c). x1240.

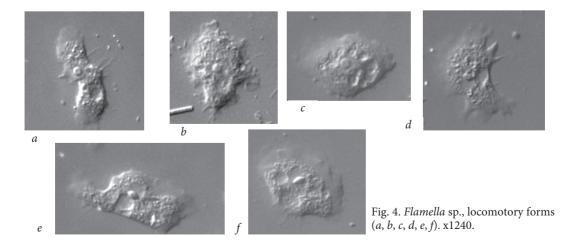
Class VARIOSEA Cavalier-Smith, Chao et Oates, 2004 Order VARIOPODIDA Cavalier-Smith, Chao et Oates, 2004 Family FILAMOEBIDAE Cavalier-Smith, Chao et Oates, 2004 Genus Flamella Schaeffer, 1926 Flamella sp. (fig. 4)

The species belongs to the flamellian morphotype. Locomotive shape is flattened and semicircular. Anterior edge of hyaloplasm is uneven and chaotically forms numerous short subpseudopodia, thin or conical. Lateral hyaloplasm also can produce subpseudopodia. Cytoplasm contains several contractile vacuoles. This amoeba is capable of rapid locomotion. Posterior part of the cell is as a rule concave, rarely convex, usually with numerous short filaments. Amoebae of the genus *Flamella* are able to encyct (Page, 1988; Page, Siemensma, 1991) though we did not observe this in the cultures. Cell length is 30–55 μ m, width is 50–70 μ m, L/B ratio is 0.4–1.2. There is a single nucleus 4.8–9.0 μ m in diameter.

Assignment of observed amoebas to the genus *Flamella* is confirmed by the complex of above mentioned morphological features, the presence of the semi-circular locomotor form, the wide frontal zone of hyaloplasm, which creates subpseudopodias, and formation of the adhesive filaments on the posterior end of cell (Michel, Smirnov, 1999); for the definitive identification electron microscopy and cyst survey are needed.

Distribution: species of the genus *Flamella* occur in Europe and North America (Page, Siemensma, 1991).

Habitats: we found representatives of the genus in the rivers (River Guyva of Zhytomir and Andrushivka cities; River Kam'yanka near Zhytomir city, Lopatychi village, Olevsky District, and Velyky Lug village, Chervonoarmiysky District of Zhytomir Region; the river near Perga village, Zdolbunivsky District and near Groziv village, Ostrozky District of



Zhytomir Region), lakes (Pedynka village, Lyubarsky District of Zhytomir Region; Chevel village, Staro-Vyjivsky District of Volyn Region), bogs (Berezivka village, Zhytomir District; Galiyivka village, Vel. Korovytskogo District of Zhytomir Region; Chevel village, Staro-Vyzhyvsky District of Volyn Region) and drainage canal near Ostrog city of Rivno Region.

Class PROTEOMYXIDEA Lankester, 1885 Order ACONCHULINIDA De Saedeleer, 1934 Family BIOMYXIDAE Loeblich et Tappan, 1961 Genus Penardia Cash, 1904 Penardia mutabilis Cash, 1904 (fig. 5)

This is a large naked free-living amoeba. Cell shape is variable, spread if adhered to a substrate. The locomotion is relatively rapid. In motion, the amoeba changes its shape, branching in a reticulate net. Pseudopodia can reach 35 μ m in length, growing out of a peripheral hyaline fringe of the cytoplasm. Cell length is 300–540 μ m, cell width is 80–110 μ m, L/B ratio is 3.5–4. The nuclei are 5–7.5 μ m in diameter.

Remarks: the species can be distinguished from closely related species of the genus *Biomyxa* Leidy, 1875 by cytoplasm's rapid movement and greenish color (the *Biomyxa* amoebae have uncolored clear cytoplasm) (Page, Siemensma, 1991).

Distribution: P. mutabilis occurs in Europe (Page, Siemensma, 1991).

Habitats: we found the species in lake Svitiaz (Shatsky National Natural Park, Volyn Region) (Patcyuk, Dovgal, 2012).

Thus, as a result of the research, 5 amoebae species were found in Ukraine for the first time. According to latest views on the system of naked amoebae (Smirnov et al., 2011; Bass et al., 2009), they belong to 4 genera from 4 classes, 4 orders and 4 families. Four of the species are lobosea naked amoebae (*Rhizamoeba* sp., *T. quadrilineata*, *T. verrucosa*, *Flamella* sp.), one is a naked filosea amoeba species (*P. mutabilis*). Also, *P. mutabilis* ecologically belongs to limnic species complex of naked amoebae that is characteristic of Shatsk lakes (Patcyuk, Dovgal, 2012).

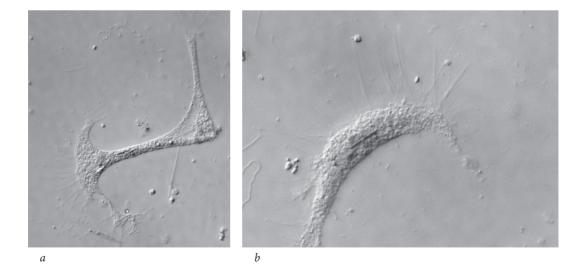


Fig. 5. Penardia mutabilis, locomotory forms (a, b). x1240.

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References

- Adl, S. M., Simpson, A. G., Lane, C. E. et al. 2012. The Revised Classification of Eukaryotes. J. Eukaryot. Microbiol., 59 (5), 429-493.
- Bass, D., Chao, E., Nikolaev, S. et al. 2009. Phylogeny of Novel Naked Filose and Reticulose Cercozoa: Granofilosea cl. n. and Proteomyxidea Revised. *Protistogy*, **160**, 75–109.
- Brown, S., Smirnov, A. 2004. Diversity of Gymnamoebia in grassland soil southern Scotland. *Protistology*, **3**,191–195.
- Michel, R., Smirnov, A. V. 1999. The Genus *Flamella* Schaeffer, 1926 (Lobosea, Gymnamoebia), with Description of Two New Species. *Europ. J. Protistology*, **35**, 403–410.
- Page, F. C. 1988. A new key to freshwater and soil gymnamoebae. *Freshwater Biological Association*. Ambleside, Cumbria, UK, 1–122.
- Page, F. C., Siemensma, F. J. 1991. Nackte Rhizopoda und Heliozoea (Protozoenfauna; Bd 2). Gustav Fischer Verlag, Stuttgart; New York, 3–170.
- Patsyuk, M. K. 2010. Naked Lobose Amoeba (Lobosea, Gymnamoebia) of Some Reservoirs of Fence Surrounding Villages of Radomishl'. Scientific Notes of Ternopyl' National Pedagogical University. Ser. Biology, 2 (43), 390–395 [In Ukrainian].
- Patsyuk, M. K. 2011 a. Biotopic Allocation of Naked Amoebas in the Waters of Ukrainian Polessje. IV International Symposium "Ecology of Free-Living Protists in Terrestrial and Aquatic Ecosystems: Book of Abstracts" (October 17–21, 2011, Togliatty, Russia), 53 [In Russian].
- Patsyuk, M. K. 2011 b. The First Finds of Naked Amoebas in the Lake Svityaz. Scientific Notes of Ternopyl' National Pedagogical University. Ser. Biology, 3 (48), 27–30 [In Ukrainian].
- Patsyuk, M. K. 2012 a. New Gymnamoebae species (Gymnamoebia) in the fauna of Ukraine. *Vestnik Zoologii*, **46** (2), 105–111.
- Patsyuk, M. K. 2012 b. The Taxonomical Composition of Naked Amoebas in the Lakes of Shatsk. *In:* Zuzuk, F. V., ed. *The Nature of Western Polessje and Its Neighborhoods*. Volyn National University, Lutsk, 9, 177–180 [In Ukrainian].
- Patcyuk, M. K., Dovgal, I. V. 2012. Biotopic distribution of naked amoebes (Protista) in Ukrainian Polissya area. *Vestnik Zoologii*, **46** (4), 355–360.
- Patsyuk, M. K. 2014 a. Naked Amoebas of the Lakes of Shatsk. In: Zuzuk, F. V., ed. The Nature of Western Polessje and Its Neighborhoods. Volyn National University, Lutsk, 11, 239–243 [In Ukrainian].
- Patsyuk, M. K. 2014 b. Naked Amoebas in the Fauna of Kyiv Woodland Region. Scientific Notes of Ternopyl' National Pedagogical University. Ser. Biology, 2 (59), 49–52 [In Ukrainian].
- Smirnov, A. V., Goodkov, A. V. 1993. Paradermamoeba valamo gen. n., sp. n. (Gymnamoebia, Thecamoebidae) — freshwater amoeba from bottom soil. Zoologicheskii Zhurnal, 72 (2), 5–11 [In Russian].
- Smirnov, A. V., Goodkov, A. V. 1999. An illustrated list of basic morphotypes of Gymnamoebia (Rhizopoda, Lobosea). *Protistology*, **1**, 20–29.
- Smirnov, A. V. 2008. Amoebas, Lobose. In: Schaechter M., ed. Encyclopedia of Microbiology. Elsevier, Oxford, 558–577.
- Smirnov, A. V., Chao, E., Nassonova, E. S., Cavalier-Smith, T. 2011. A Revised Classification of Naked Lobose Amoebae (Amoebozoa: Lobosa). *Protistology*, 162, 545–570.

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