

New and little known free-living ciliates from the plankton of the Caspian Sea

Ilham Alekperov ¹, Edward Buskey ² and Nataly Snegovaya ¹

¹ *Institute of Zoology NAS of Azerbaijan, Baku, Azerbaijan*

² *University of Texas, Marine Sciences Institute, Port Aransas, USA*

Summary

Two new for science (*Strombidium nabranicum* sp. n. and *Euplotes khazarica* sp. n.) and one little-known (*Euplotes alatus* Kahl 1932) species of free-living ciliates from the plankton of the Caspian Sea are described.

Key words: free-living ciliates, plankton, *Strombidium nabranicum* sp. n., *Euplotes khazarica* sp. n., *Euplotes alatus*, impregnation, Caspian Sea

Introduction

The Caspian Sea is the largest continental water body on the Earth (Dumont, 1998). It is known for its petroleum resources and sturgeon fisheries. This elongate water body is divided into three parts; a shallow northern basin in which its main freshwater inflow, the Volga River, enters, and two deep basins in the central and southern regions. Average salinity is about 12.8‰, and water temperatures exhibit both a north-south gradient and seasonal changes (Dumont, 1998). Vertical mixing keeps the deep basins oxygenated to the bottom, unlike the Black Sea (Kosarev and Yablonskaya, 1994). Nutrient levels and primary production in the Caspian Sea are generally low, although nutrient influx has increased sharply in recent years (Dumont, 1998).

Although this closed salt-water reservoir has no outlets to the ocean, and possesses many attributes of a sea (Zenkevitch, 1963), its legal status as a lake or sea

remains undefined, and this in turn affects how access to resources are regulated between countries (De Mora and Turner, 2004). The Caspian Sea has a highly endemic biota, characterized by their euryhaline tolerances, with both freshwater species that tolerate salinities of up to 13‰, and marine species that tolerate low salinities (Dumont, 1998). During the past few decades the Caspian Sea has undergone a series of serious environmental challenges including pollution, over fishing, introduction of invasive species and changes in water level (Barannik et al., 2004). These environmental changes make it especially important to increase efforts to characterize the biota of the Caspian Sea.

The Caspian Sea has a diverse ciliate fauna with 460 species of ciliates, including 138 planktonic species (Agamaliyev, 1983; Alekperov and Asadullayeva, 1996, 1997, 1999). Planktonic ciliates of the Caspian Sea are represented mainly by marine species (125 species), with only 11 freshwater species, and 2 brackish water species.

In spite of long term investigations of ciliates in the Caspian Sea, several new species have been discovered during a recent sampling program and they are described here. We describe three species of ciliates collected in the near-shore plankton of the central basin of the Caspian Sea in the vicinity of Nabran village (North Azerbaijan). One of them (*Euplotes alatus* Kahl 1932) is new for the Caspian fauna and the others (*Strombidium nabranicum* sp. n. and *E. khazarica* sp. n.) are new for science.

Material and Methods

The topology of kinetomes was determined using the silver impregnation method (Chatton and Lwoff, 1930; Alekperov, 1992). The nuclei were stained with the Feulgen nuclear reaction. All the measurements were made on no less than 10 (usually 15) randomly selected specimens. Morphometric characterization of all described ciliates species is given in the Table 1. The type specimens of the new taxa have been deposited in

the Institute of Zoology, National Academy of Sciences of Azerbaijan, Baku.

Results

DESCRIPTION OF SPECIES

***Strombidium nabranicum* sp. n.** (Table 1; Figs 1, A-B; 5, A-B).

Diagnosis. Size of living cells 60-80 μm , after fixation up 65 μm , acontractile. Somatic ciliature consisting of 18-20 dikinetid rows. Body surface covered with multiple silverline system. There are 15 anterial (collar) and 14 ventral (adoral) membranelles.

Type location. Littoral plankton of the Middle Caspian Sea (near Nabran village, not far from Azerbaijanian border with Russia).

Type specimens. A holotype and a paratype of *S. nabranicum* sp. n. on two slides with silver nitrate impregnated cells have been deposited in the collection of microscope slides of the Laboratory of Protistology

Table 1. Morphometric characterization of described ciliates species (after impregnation)*.

<i>Strombidium nabranicum</i> sp. n.							
Character	Mean	M	Max	Min	SD	SE	n
Length	69	65	80	60	7,66	1,98	15
Width	58	61	65	50	5,86	1,51	15
Collar number of membranelles	15	15	16	14	0,54	0,14	15
Ventral number of membranelles	14	14	14	13	0,46	0,12	15
Somatic rows number	34	35	37	31	1,58	0,41	15
Macronucleus length	32	32	35	28	2,56	0,66	15
Diameter of micronucleus	3,7	3,8	4,0	3,1	0,32	0,81	15
Length of ventral membranelles	9	9	12	6	2,18	0,56	15
Length of collar membranelles	23	24	26	20	1,96	0,62	10
<i>Euplotes alatus</i> Kahl 1932							
Character	Mean	M	Max	Min	SD	SE	n
Length	26	28	30	20	4,20	1,08	15
Width	22	22	30	11,5	5,62	1,45	15
Adoral membranelles number	42	42	45	40	2,03	0,52	15
Adoral zone length	27	27	30	24	1,89	0,49	15
Macronucleus length	21	21	25	17	2,93	0,76	15
Fronto-ventral cirri number	10	10	10	10	0	0	15
Caudal cirri number	4	4	4	4	0	0	15
Dorso-lateral kineties number	8	8	8	8	0	0	15
Number of dorsal bristles in the dorso-lateral rows	14	14	18	12	1,62	0,42	15
<i>Euplotes khazarica</i> sp. n.							
Character	Mean	M	Max	Min	SD	SE	n
Length	30	30	35	22	3,56	0,93	15
Width	28	29	32	20	4,20	1,09	15
Adoral membranelles number	48	48	50	45	2,09	0,54	15
Adoral zone of membranelles, length	26	26	29	23	1,76	0,45	15
Macronucleus length	27	27	29	25	1,1	0,28	15
Fronto-ventral cirri number	11	11	11	11	0	0	15
Caudal cirri number	4	4	4	4	0	0	15
Transversal cirri number	5	5	5	5	0	0	15
Dorso-lateral kineties number	5	5	5	5	0	0	15
Number of dorsal bristles in the dorso-lateral rows	18	18	19	18	0,46	0,12	15

* Measurements in μm . Mean - arithmetic mean, M - Median, Max - maximum, Min - minimum, SD - standard deviation, SE - standard error of mean.

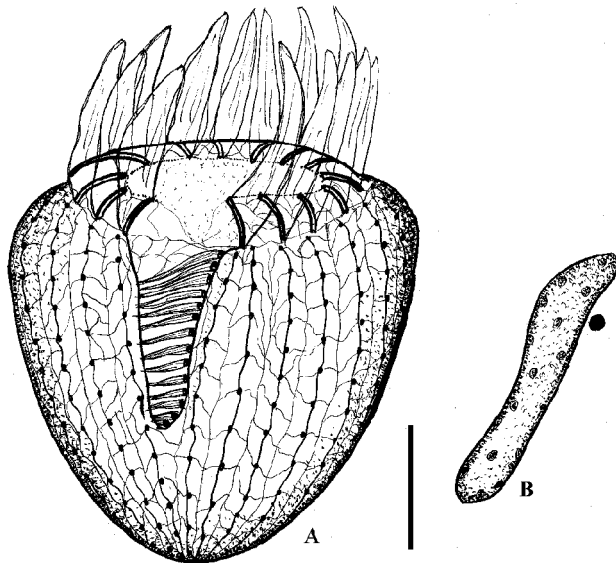


Fig. 1. *Strombidium nabranicum* sp. n. A - general view from ventral side; showing the collar and ventral adoral membranelles; B - nuclei (Feulgen reaction). Scale bar: 15 μ m.

Institute of Zoology National Academy of Sciences of Azerbaijan, Baku city. Accession number Nab. mar. N4.

Dedication. Named after region found.

Description. Size *in vivo* 60–80, after fixation 65 μ m. Body is conical. The peristomal field is confined to the 15 anterior (collar) and 14 ventral membranelles. Anterior membranelles each comprising 3 basal body rows with cilia up to 25 μ m long. Ventral membranelles are distinctly different from anterior membranelles, the longest are about 12 μ m, the other gradually shortened.

The silverline system is unusual for *Strombidium* with a fine net of argyronems. The contractile vacuole is located at the posterior part of the body. Macronucleus is elongated (35 μ m) with numerous nucleoli and a single spherical micronucleus.

Discussion. The *Strombidium* genus has been actively investigated over the past 20 years (Montagnes et al., 1988, 1990; Petz and Foissner, 1992; Alekperov and Mamaeva, 1992; Lynn and Gilron, 1993; Petz, 1994).

Our *Strombidium nabranicum* sp. n. differs from all the remaining species of the genus by its unusual multiple silverline system. It is possible that future examination will show that separation at the genus level is very likely to be appropriate.

***Euplotes alatus* Kahl 1932** (Table 1; Fig. 2, A-C).

This is a very rare species and as far as we know, there was only one modern reinvestigation of *E. alatus* using silver impregnation (Borror, 1968). For the fauna of the Caspian Sea it is its first record.

The size of living specimens is 30–45 μ m; after fixation up to 30 μ m. Body is oval and flattened dorso-ventrally. Adoral zone of membranelles contains 40–45 elements and is located on the left and anterior margins. There are 10 fronto-ventral cirri, five transventral and four caudal cirri. On the dorsal side 8 dorsal kineticies are located. Two of them can be seen from the ventral side. Each dorsal kinetic row contains 12–18 cilia. Dargyrom - type II "eurystomus".

A contractile vacuole is present at about the level of the transverse cirri. Cytoplasm is colorless with some food vacuoles.

There are several *Euplotes* species with dargyrom of "double eurystomus" and 10 frontoventral cirri (Table 2).

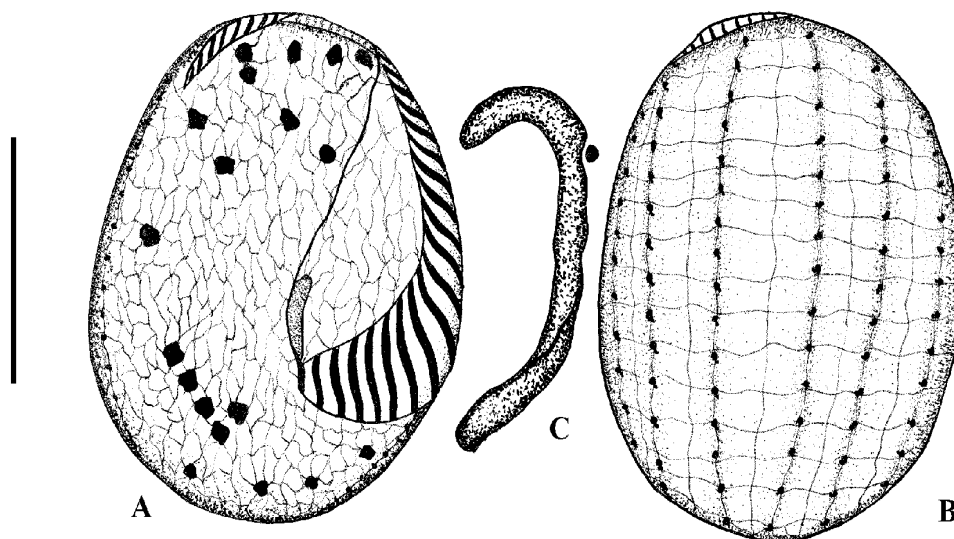


Fig. 2. *Euplotes alatus* Kahl 1932. A - ventral side; B - dorsal side (impregnation by silver nitrate); C - nuclei (Feulgen reaction). Scale bar: 15 μ m.

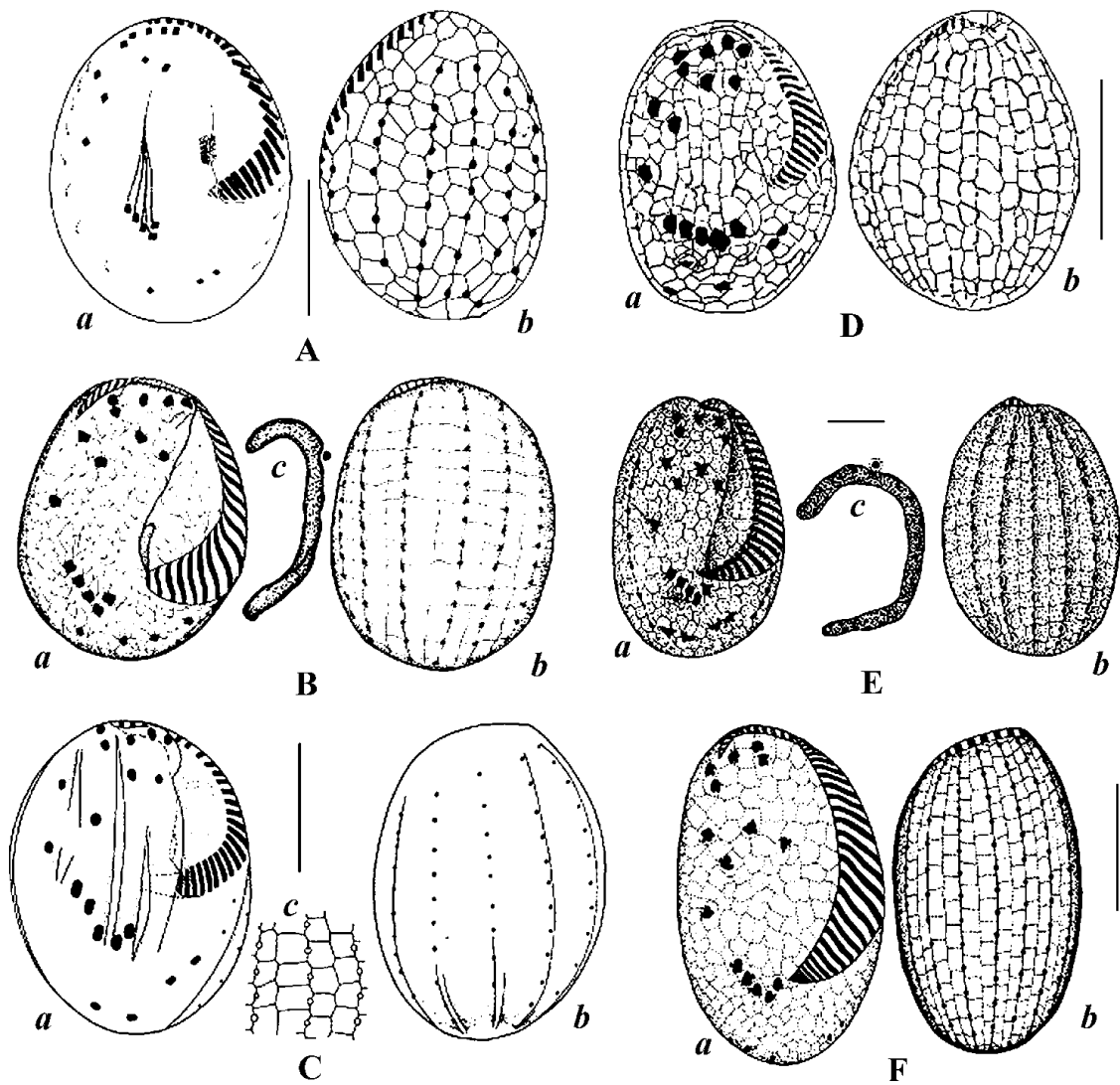


Fig. 3. Comparison of some different populations *Euplotes corsica*, *E. alatus* and *E. balteatus*: A - *Euplotes corsica* (Berger and Foissner, 1989), a - ventral side, b - dorsal side; B - *Euplotes alatus*, Caspian population (present paper), a - ventral side, b - dorsal side, c - nuclei; C - *Euplotes alatus*, Atlantic population (Borrer, 1968), a - ventral side, b - dorsal side, c - "dargyrom"; D - *Euplotes balteatus*, Antarctic population (Song and Wilbert, 2002), a - ventral side, b - dorsal side; E - *Euplotes balteatus*, Caspian population (Agamaliev, 1983), a - ventral side, b - dorsal side, c - nuclei; F - *Euplotes balteatus*, fresh-water population (Alekperov, 1983), a - ventral side, b - dorsal side. Scale bars: A, C, F - 20 µm, B - 15 µm, D - 30 µm, E - 10 µm.

Table 2. Comparison of some small, morphologically-related marine *Euplotes* species with dargyrom of "double eurytomus" and 10 frontoventral cirri.

Species	Body size	AZM number	Fronto-ventral cirri	Transversal cirri	Caudal cirri	Dorsal kineties number	Number of basal body pairs in the dorsal rows	References
<i>E. balteatus</i>	45-50	35-40	10	5	4	7	13-14	Alekperov, 1983
<i>E. balteatus</i>	40-70	27-33	10	5	2+2 *	8-10	11	Song and Wilbert, 2002
<i>E. alatus</i>	20-30	40-45	10	5	4	8	12-15	Present paper
<i>E. alatus</i>	36-43	Probably 30	10	5	4	8	10-11	Borrer, 1968
<i>E. corsica</i>	29-42	22	10	5	3-4	7-8	6-9	Berger and Foissner, 1989

* Song and Wilbert (2002) differs two caudal and two marginal cirri.

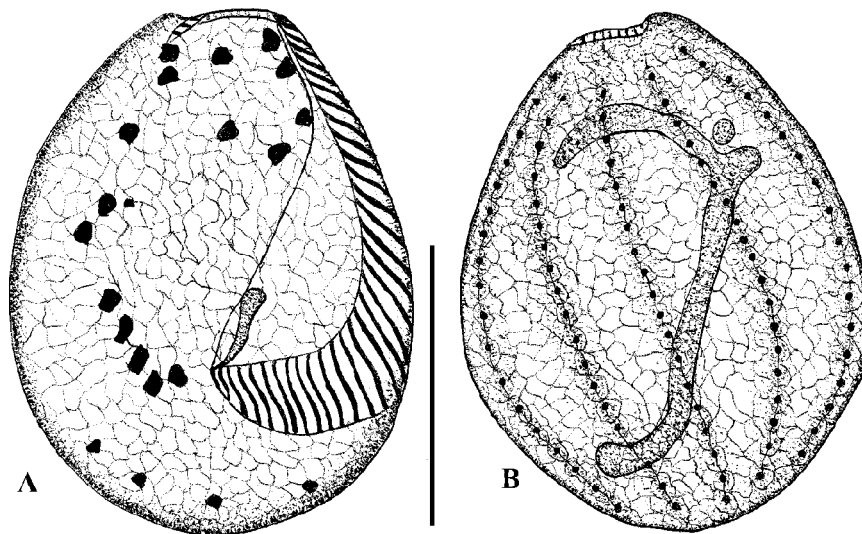


Fig. 4. *Euplotes khazarica* sp. n. A - ventral side; B - dorsal side and nuclei (impregnation by silver nitrate). Scale bar: 25 μ m.

According to Song and Wilbert (2002) *Euplotes alatus* Kahl 1932 is possibly a junior synonym of *E. balteatus* Dujardin 1842.

In Fig. 3 we show 3 populations of *E. balteatus*, described from different regions and different water salinity. In our opinion all this *Euplotes balteatus* populations clearly differs from *Euplotes alatus*, described by Borror (1968) and at the present paper in the shape and size of the body and arrangement of fronto-terminal cirri, especially on the frontal part. That is why we agree with Borror (1968) that *E. balteatus* is really different from *E. alatus* in cirrus pattern and the shape of its buccal overture and that is why *E. alatus* appears to be valid separate species.

In our opinion species *E. corsica* described by Berger and Foissner (1989) is very probably conspecific with the *E. alatus* Kahl 1932 and differs shape and size of the body and the number of dorsal rows. These two species (*E. corsica* and *E. alatus*) has more similarity, than *Euplotes balteatus*.

***Euplotes khazarica* sp. n.** (Table 1; Figs 4, A-B; 5, C-D).

Diagnosis. Size of living cells is 40-55 μ m, after fixation - up to 35 μ m. Body oval, flattened dorsoventrally. Adoral zone of membranelles contains 45-50 elements and occupies the left margin of the ventral side. There are 11 frontoterminal cirri: near two of them which locate upper 5 transversal cirri there is small unciliated argentophilic plate. Four caudal cirri. Ventral argyrom fine with small argyroconts with polygonal shapes. On the dorsal side there are five ribs with 5 dorsal kineties with 18-19 cilia. Dargyrom - type *E. "musvicola"* III.

Endoplasm is clear and uncolored. Macronucleus C-like with small ledge. A spherical micronucleus is located nearby.

Type location. This species was found in the Caspian Sea plankton near Nabran village coast (Not far from Azerbaijan-Russia border).

Type specimens. A holotype and a paratype of *E. khazarica* sp. n. as two slides of silver nitrate impregnated cells have been deposited in the collection of the Institute of Zoology NAS of Azerbaijan. Accession number Nab.mar. N8.

Discussion. There have been many new descriptions and redescrptions of the representatives of *Euplotes* genus published in recent years (Dragesco and Dragesco-Kerneis, 1986; Berger and Foissner, 1989; Hill et al., 1986; Augustin and Foissner, 1989; Agatha and Wilbert, 1990; Valbonesi and Luporini, 1990a, 1990b; Al-Rasheid, 2001; Song and Wilbert, 2002).

According to Borror (1968) the basis for identification of member of *Euplotes* genus are:

1. cortical sculpturing;
2. arrangement of all ciliary organelles, including dorsal and endoral cilia;
3. details of the silverline system.

Euplotes khazarica sp. n. described above differs from other representatives of this genus by its maximal number of frontoventral cirri - 11 and minimal number of dorsal cilia rows. From *E. trisulcatus* Kahl 1932, *E. khazarica* differs in body shape, with a much longer adoral zone of membranelles, smaller on two number of dorsal rows and other type dargyrom.

Dedication. Named after ancestral Caspian Sea name (Khazarian Sea).

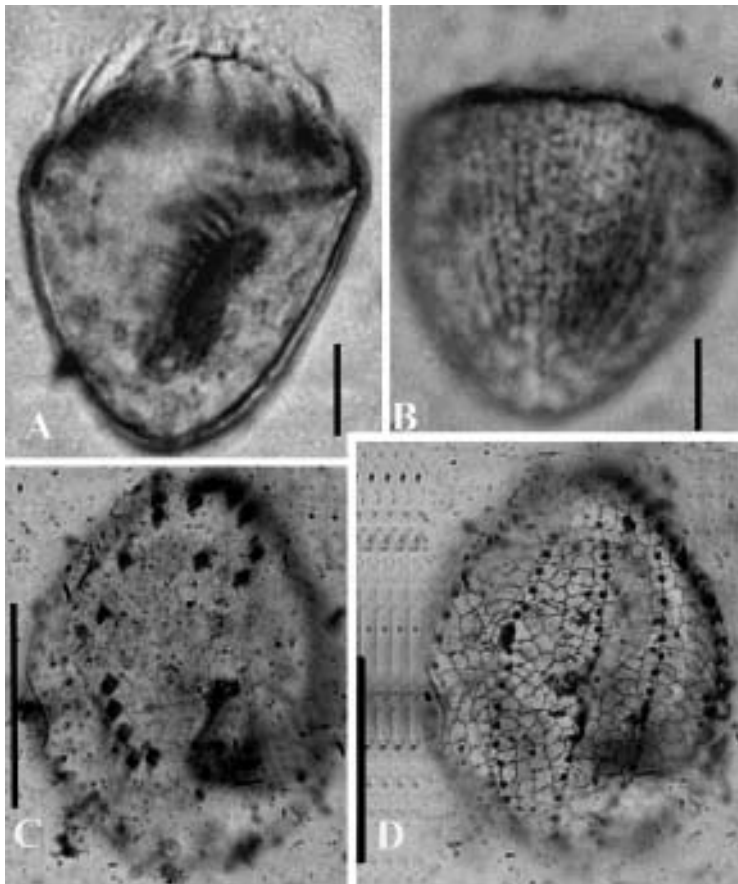


Fig. 5. Silver impregnation of *Strombidium nabranicum* sp. n. and *Euplotes khazarica* sp. n. A - *Strombidium nabranicum*, ventral surface showing ventral adoral membranelles; B - *S. nabranicum*, dorsal surface with somatic kinetic rows and silverline system; C - *Euplotes khazarica*, ventral side; D - *E. khazarica*, dorsal side. Scale bars: A-B - 15 μ m, C-D - 25 μ m.

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References

- Agamaliev F.G. 1983. The Ciliates of the Caspian Sea: systematic, ecology, zoogeography. Leningrad, Nauka (in Russian).
- Agatha S., Wilbert N., Spindler M. and Elbrachter M. 1990. Euplotide ciliates in Sea Ice of the Weddell Sea (Antarctica). *Acta Protozool.* 29, 221-228.
- Al-Rasheid K.A.S. 2001. New records of interstitial ciliates (Protozoa, Ciliophora) from the Saudi coasts of the Red Sea. *Tropical Zoology.* 14, 133-156.
- Alekperov I.Kh. 1983. Morphology of freshwater ciliates of the Azerbaijanian water reservoirs. M., VINITI, N 18391 (in Russian).

Alekperov I.Kh. 1992. New modification of the silver proteinate impregnation method of the infusorian kinetome. *Zool. Zh.* 71, 130-133 (in Russian, with English summary).

Alekperov I. and Mamaeva N. 1992. Planktonic infusoria from the Chuckchee and Bering seas. *Zool. Zh.* 71, 5-14 (in Russian, with English summary).

Alekperov I.Kh. and Asadullayeva E. 1996. New and little-known ciliates from Apsheron coast of the Caspian Sea. Part I. Fam. Prostomatida-Trichostomatida. *Zool. Zh.* 75, 763-769 (in Russian, with English summary).

Alekperov I.Kh. and Asadullayeva E. 1997. New and little-known ciliates from Apsheron coast of the Caspian Sea. Part I. Fam. Nassulida-Oligotrichida. *Zool. Zh.* 76, 1411-1417 (in Russian, with English summary).

Alekperov I.Kh. and Asadullayeva E. 1999. New, little-known and characteristic ciliate species from the Apsheron coast of the Caspian Sea. *Turk. J. Zool.* 23, 275-284.

Augustin H. and Foissner W. 1989. Morphologie einiger Ciliaten (Protozoa: Ciliophora) aus dem Belebtschlamm. *Lauterbornia.* 1, 38-59.

Barannik V., Borysova O. and Stolberg F. 2004. The Caspian Sea region: environmental Change. *Ambio.* 33, 45-51.

Berger H. and Foissner W. 1989. Morphology and biometry of some soil hypotrichs (Protozoa, Ciliophora) from Europe and Japan. *Bull. Br. Mus. nat. Hist. (Zool.).* 55, 19-46.

Borror A. 1968. Systematics of *Euplotes* (Ciliophora, Hypotrichida); toward union of the old and the new. *J. Protozool.* 15, 802-808.

Chatton E. and Lwoff A. 1930. Impregnation, par diffusion argentine, de l'infuciliature des cilies marins et d'eau douce, apres fixation cytologique et sans desiccation. *C. R. Soc. Biol. Paris.* 104, 834-836.

De Mora S.J. and Turner T. 2004. The Caspian Sea: a microcosm for environmental science and international cooperation. *Mar. Poll. Bull.* 48, 26-29.

Dragesco J. and Dragesco-Kerneis A. 1986. Cilies libres de l'Afrique intertropicale. *Faune Tropicale, Paris.* 26, 1-559.

Dumont H. 1998. The Caspian Lake: history, biota, structure, and function. *Limnol. Oceanogr.* 43, 44-52.

Hill B., Small E. and Iliffe T. 1986. *Euplotes iliffei* n. sp.: a new species of *Euplotes* (Ciliophora, Hypo-

trichida) from the marine caves of Bermuda. J. Wash. Acad. Sci. 76, 244-249.

Kosarev A.N. and Yablonskaya E.A. 1994. The Caspian Sea. Netherlands: SPB Academic Publ., The Hague.

Lynn D.H. and Gilron G.L. 1993. Strombidiid ciliates from coastal waters near Kingston Harbour, Jamaica (Ciliophora, Oligotrichia, Strombidiidae). J. mar. biol. Ass. U.K. 74, 47-65.

Montagnes D.J.S., Lynn D.H., Stoecker D.K. and Small E.B. 1988. Taxonomic descriptions of one new species and redescription of four species in the family Strombidiidae (Ciliophora, Oligotrichida). J. Protozool. 35, 189-197.

Montagnes D.J.S., Taylor F.J.R. and Lynn D.H. 1990. *Strombidium inclinatum* n. sp. and a reassessment of *Strombidium sulcatum* Claparede and Lachmann (Ciliophora). J. Protozool. 37, 318-323.

Petz W. 1994. Morphology and morphogenesis of *Strombidium kryalis* nov. spec. (Ciliophora, Strombi-

diidae) from Antarctic Sea ice. Arch. Protistenkd. 144, 185-195.

Petz W. and Foissner W. 1992. Morphology and morphogenesis of *Strombidium caudatum* (Fromentel), *Meseres corlissi* n. sp., *Halteria grandinella* (Muller), and *Strombidium rehwaldi* n. sp., and a proposed phylogenetic system for oligotrich ciliates (Protozoa, Ciliophora). J. Protozool. 39, 159-176.

Song W. and Wilbert N. 2002. Faunistic studies on marine ciliates from the Antarctic benthic area, including descriptions of one epizoic form, 6 new species and, 2 new genera (Protozoa: Ciliophora). Acta Protozool. 41, 23-61.

Valbonesi A. and Luporini P. 1990a. Description of two new species of *Euplotes* and *Euplotes rariseta* from Antarctica. Polar Biol. 11, 47-53.

Valbonesi A. and Luporini P. 1990b. A new marine species of *Euplotes* (Ciliophora, Hypotrichida) from Antarctica. Bull. Br. Mus. nat. Hist. (Zool.). 56. 57-61.

Address for correspondence: Ilham Alekperov. Laboratory of Protistology, Institute of Zoology, Azerbaijan National Academy of Sciences, Baku, 370073 Azerbaijan. E-mail: i_alekperov@yahoo.com

Editorial responsibility: Sergei Fokin