# The Little-Known Free-Living Ciliates from the Soils of Pirgulian State Reserve (Eastern Azerbaijan)

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**Abstract:** The species diversity of the soil free-living ciliates of 3 parts of Pirgulian State Reserve was investigated. In total, 120 species of free-living ciliates were found. Five of them (Enchelys vermiformis F., 1986, Epispathidium terricola F., 1986, Drepanomonas sphagni Kahl, 1931, Frontonia terricola F., 1986, and Cirrophrya terricola F., 1986) are new for the Caucasus fauna. The description of 5 species are based on the observations of living specimens and the analysis of total slides impregnated by silver nitrate

Key Words: Fauna, soil ciliates, impregnation, Azerbaijan

### Introduction

Pirgulian State Reserve includes 3 parts, consisting in all of 1520 ha. The highest part (Arakhchian, 29% of the area) is located on the forest level at 1600-2000 m above sea level (a.s.l.). Some of the lower part (1000-1500 a.s.l.) (Pirgulian, 34.9%) is located on the middle of the forest level, and at 800-1000 m a.s.l., there is the lowest part (Janginian, 36.1%) of the State Reserve.

Considering the poor knowledge of the soil ciliates (especially in the South Caucasus), we carried out an investigation of the free-living ciliate fauna in the soils of all 3 parts of Pirgulian State Reserve, located near the city of Shemakha (Eastern Azerbaijan).

### **Materials and Methods**

The investigated material was collected seasonally from January 2004 to October 2005. In all, 80 soil samples were taken. Twenty samples were taken from the highest (Arakhchian) part, located on the border of subalpine and forest landscapes, 30 samples were taken from the Pirgulian part with forest landscape, and 30 samples were collected from lowest (Janginian) part with forest landscape.

The low and middle parts of Pirgulian State Reserve have brown mountain-forest soils and are covered with beech, hornbeam and oak trees. The highest part has turf alpine-grasslands soils and is covered with sparse growth of bushes and grass.

Freshly collected soil samples (horizon 0-20 cm) were delivered to the Protistology Laboratory of the Institute of Zoology of the National Academy of Sciences of Azerbaijan. Altogether 80 soil samples were placed in petri dishes (diameter 10 cm). To 1 g of soil was added 10 ml of distilled water with a small quantity of dry peptone. The soil samples run with water were kept at room temperature. The topology of kinetomes was determined by a method of silver nitrate and silver proteinate impregnation (Chatton and Lwoff, 1930; Alekperov, 1992). The nuclei were stained with the Feulgen nuclear reaction. The ciliates were investigated with a light microscope. All the measurements were made on no less than 15 specimens of each species in vivo and total impregnated slides.

### **Results and Discussion**

Family Enchelyidae Ehrenberg, 1838

1. Enchelys vermiformis Foissner, 1986 (Figure 1, A-B)

This species was found at the first time in the mountain soils not far from Salzburg (Austria) by Foissner (1986).

The size of living cells is 120-200  $\mu m$  x 30-50  $\mu m$ , after fixation 100-150  $\mu m$  x 20-35  $\mu m$ . Body elongated worm-like shape. Apical cytostome with 8-11 spiral nematodesmal elements. Below cytostome 3 short rows of dorsal brosse, consisting of double kinetosomes.

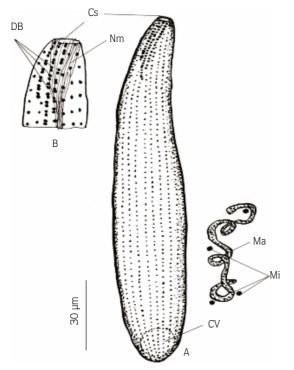


Figure 1. Enchelys vermiformis. A – general view, B – oral zone, Cs – cytostome, DB – dorsal brosse, CV – contractile vacuole, Nm – nematodesmata (silver impregnation), Ma – macronucleus, Mi – micronucleus (Feulgen reaction).

Somatic ciliature consists of 21 rows of kinetosomes, of which 3 unipolar ones begin at dorsal brosse.

Endoplasm dark brown, without inclusions. Nuclear apparatus consisting of a long band-like macronucleus and 3-5 micronuclei. Contractile vacuoles located at the caudal end.

Our specimens of *E. vermiformis* has a larger number of somatic kineties, contrary to the first description (Foissner, 1986) (21 rows versus 17 rows). It is a rare species in the study area, being registered only the highest (Arakhchian) part of Pirgulian State Reserve.

## Family Spathidiiade Kahl in Dofflein and Reichenow, 1929

2. *Epispathidium terricola* Foissner, 1986 (Figure 2, A-B)

This species was found for the first time in the soils in Austria (Foissner, 1986). Size of living cells 100-130  $\mu m$  x 20-45  $\mu m$ , fixed cells up to 90  $\mu m$ . Body lanceolate, flattened dorsoventrally. Oral slit encircled by a well-marked lip. Cytostome with numerous thick

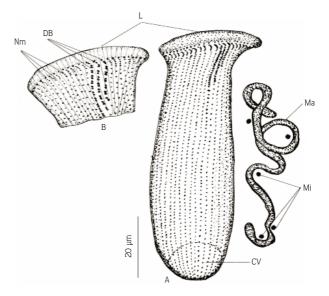


Figure 2. Epispathidium terricola. A – general view, B – oral zone, L – lip, DB – dorsal brosse, CV – contractile vacuole, Nm – nematodesmata (silver impregnation), Ma – macronucleus, Mi – micronucleus (Feulgen reaction).

nematodesmata. Dorsal brosse located at the anterior part of body, contains 3 relatively short (15  $\mu$ m) rows, consisting of double kinetosomes and continuing posteriorly as normal somatic kineties.

Somatic ciliature is formed of 38-40 kineties, beginning at anterior and ending at posterior end of body. Contractile vacuole located at the caudal end.

Endoplasm transparent with numerous food vacuoles. Nuclear apparatus consisting of a long band-like macronucleus and 5 micronuclei.

Our specimens of *E. terricola* show morphological characteristics that correspond to the species described in the first description (Foissner, 1986), and differ only by the smaller cell size (our specimens 100-130 x 20-45  $\mu$ m versus 120-160 x 35-60  $\mu$ m).

### Family Microthoracidae Wresniowski, 1870

3. Drepanomonas sphagni Kahl, 1931 (Figure 3, A-B)

This is a small species of ciliate. Size of living cells 25-35  $\mu m$  x 10-15  $\mu m$ , after fixation up to 20  $\mu m$ . Body flattened laterally with anterior 4 teeth on the left edge with short (6-3) kinetic rows. On the right side a small cytostome with 5-7 nematodesmata and paroral membrane. On the right body side 3 incomplete somatic rows, consisting of dikinetids lying to the right of

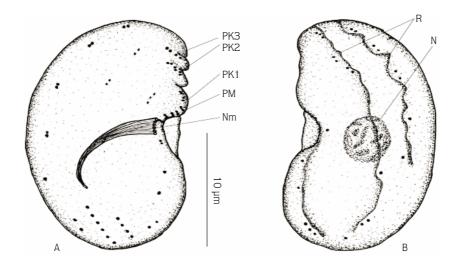


Figure 3. *Drepanomonas sphagni*. A – right lateral side, Nm – nematodesmata, PM – paroral membrane,  $PK_1$ ,  $PK_2$ ,  $PK_3$  – paroral kinetosomes, B – left lateral side, R – ribs, N – nuclei (silver impregnation).

C)

cytostome and one lying to the left of cytostome. On the left lateral body side 3 tortuous ribs with randomly scattered kinetosomes.

Endoplasm transparent, without inclusions. Contractile vacuole located below cytostome. Nuclei placed in center of the cell and consisting of a spherical (5.2  $\mu$ m) macronucleus and a single micronucleus.

From the first description (Kahl, 1931) our specimens differ by the smaller size (our specimens 25-25 x 10-15  $\mu m$  versus 30-45 x 20-25  $\mu m$ ) and stronger ribs on the left lateral body side.

It is a rare species in the study area, being registered only in the highest (Arakhchian) part of Pirgulian State Reserve.

### Family Frontoniidae Kahl, 1926

4. Frontonia terricola Foissner, 1986 (Figure 4, A-C)

This species was found and described for the first time from the soils near the Vienna suburbs (Austria) by Foissner (1986).

Size of living cells 80-140 x 50-75  $\mu$ m, after fixation up to 100  $\mu$ m. Body oval. Ventral cytostome located in the anterior part of the body. To the right of the cytostome there is an undulating membrane and 5 perizonal (vestibular) rows of closely adjacent kinetosomes. Three of them (to the right) are

uncompleted and shorter than the others. Two long perizonal rows begin at the front of the preoral unciliated suture and end at the ventral suture, dividing the posterior part of the ventral side in two.

To the left of the cytostome there are 3 strong peniculuses, consisting of several rows of cilies, 6-7 postoral rows of kineties start below the peniculuses and reach the ventral suture. Somatic ciliature consists of 55-60 rows. Endoplasm bright brown, with several food vacuoles. Nuclear apparatus represented by bean-like macronucleus with single micronucleus. On the center of dorsal side there is 1 contractile vacuole with 5 excretory pores.

Our specimens of *F. terricola* show morphological characteristics that correspond to the species described in the original description (Foissner, 1986), differing only in the smaller somatics (our specimens 55-60  $\mu$ m versus 70  $\mu$ m) and postoral rows of kinetics (our specimens 6-7 versus 8).

This species is common in the whole study area.

### Family Platyophryidae Puytorac et al., 1979

5. Cyrrophrya terricola Foissner, 1986 (Figure 5, A-

This species was found and described for the first time from the soils of Austria (Foissner, 1986).

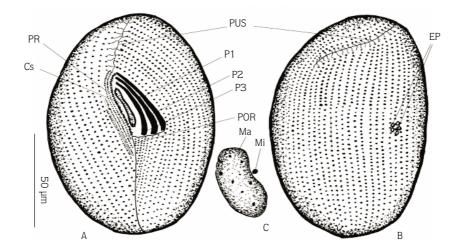


Figure 4. Frontonia terricola. A – ventral side, POR – postoral rows; PR – perizonal rows, PUS – preoral unciliated suture,  $P_1$ ,  $P_2$ ,  $P_3$  – peniculuses, Cs – cytostome, B – dorsal side, EP – excretore pores (silver impregnation), C – nuclei, Ma – macronucleus, Mi – micronucleus (Feulgen reaction).

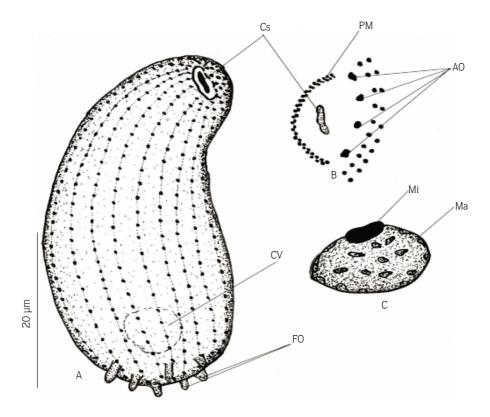


Figure 5. *Cyrrophrya terricola*. A – general view, Cs – cytostome, FO - finger-like organelles, CV – contractile vacuole, B – Oral area, PM – paroral membranes, AO – adoral organelles (silver impregnation), C – nuclei, Ma – macronucleus, Mi – micronucleus (Feulgen reaction).

Size of living cells 40-5 x 15-20  $\mu$ m, after fixation up to 45 x 12  $\mu$ m. Body elongated, contractile and curved to the left anterior end. There are 5-7 finger-like organelles on the posterior end. Its function is to attach to an animal for substrate. Subapical cytostome bordered to the left with 4 adoral organelles, to the right C-like paroral membranes.

Somatic ciliature is formed of 15-18 rows of dikinetids, which commence from the anterior edge and end at posterior edge of the body.

Endoplasm transparent, bright yellow. Nuclear apparatus consisting of a spherical macronucleus with single elliptical micronucleus. Contractile vacuoles located on the posterior part.

Our specimens differs from the first description of  $\it C.$  terricola by a lower number of somatic rows (our specimens 15-18 rows versus 20 rows).

It is a rare species in the study area, being registered only once in the Pirgulian part of the State Reserve.

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