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SEASONAL DYNAMICS OF FRESHWATER TESTATE AMOEBAE (AMOEBOZOA, ARCELLINIDA) COMMUNITIES IN LANKARAN NATURAL AREA

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Abstract. The aim of the work was to study the species composition, communities structure and main regularities of the seasonal dynamics of testate amoebae. In this regard, the seasonal changes in the total and species number of this group of protozoans with rich species diversity were studied in different water bodies of Lankaran natural area. 126 species belonging to 11 families were recorded. The spring-summer communities of testaceans (May-July) are more unique in terms of species composition and characterized by a complex of dominant species - *Centropyxis aculeata, Difflugia accuminata, D.corona, D.gramen, D.elegans*. In summer, the highest total number of testate amoebae was 7.4 thousand individuals/dm^2, and the lowest was 4.6 thousand individuals/dm^2. In winter communities, the total number of species varied between 0.1-0.9 thousand individuals/dm^2 in different water bodies.

Keywords: *testate amoebae, seasonal dynamics, species abundance, biomass, freshwater.*

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1. Introduction

Testate amoebae are widely distributed in soil, freshwater, marine and wetland biotopes and are important indicators of ecosystem status (Beaulne *et al*., 2018). They are small, diverse and abundant organisms in freshwater basins. These protozoans build their shell of various sizes and shapes from secretion or materials collected from the surrounding environment (Delaine *et al.*, 2017). Studies of species diversity and community structure of testate amoebae are the basis for further functional work and bioindication. At the same time, seasonal changes in communities are a common occurrence for most animals. The study of the seasonal dynamics of a community usually involves analysis of the species composition of the community, the dominant complex of species, the total number of organisms, the presence of rare species in the community, the number of individuals at different stages of the life cycle, etc. A distinctive feature of protozoa communities is the strong variability of the species structure over a wide range, due to the microscopic size and high reproduction rate of organisms (Burkovsky, 1992). Long-term changes are interannual fluctuations, manifested in the recombination of the complex of dominant species (they are usually associated with climatic differences of seasons), or directional changes, manifested in

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the long-term dynamics of the complex of mass species (Burkovsky, 2010). Finally, seasonal dynamics - a consistent irreversible and natural change of one microbial community by another for a certain area of the environment in time. Undoubtedly, seasonal dynamics is the central mechanism that ensures the stability and lability of protozoocenosis. Studying the seasonal rearrangements of protozoan communities is very laborious; it requires considering many environmental factors and regular collection of material for research.

2. Material and methods

Material was collected seasonally during 2013-2020. More than 2100 water samples were collected and processed over the research period. Samples were collected from plankton, peryphiton and benthos using polyethilene containers. To separate the mineral particles from the shell, the surface sediments in the samples were carefully mixed with distilled water in a clean glass beaker for 5 minutes. All specimens were identified to the lowest taxa, except where sufficient taxonomic characters were not observed. Individuals were identified up to species level and counted. Counting the number of all shells gives an adequate idea of the overall composition of biotopes where individuals are distributed. 0.001% neutral red dye solution was used during the counting of individuals. The coloring of the protoplasm indicates that the individuals are alive. This process repeated 5-15 times for each sample and the average number of individuals of species was calculated. In addition, a FlowCam fluid imaging densitometer was used to count the number of testate amoebae. For this purpose, the water sample was filtered and cleaned of soil sediments. Then 2 ml of that water sample added to the densitometer tube. In the software of the device, a numerical analysis was performed by entering a diameter interval of 10-200 μm for the objects to be studied. A flotation method was also used to collect amoeba individuals in benthic samples. This method is usually used for counting the number of psammophilous species living in sand, salt and other mineral substrates (Mahlfeld *et al.*, 2016). Glycerin-gelatin method was mainly used to identify species based on temporary preparations (Alekperov *et al.*, 1996). Olympus light microscope and JCM-6000 scanning electron microscope (SEM) were used for qualitative analysis of the collected material. In the taxonomic classification of the species, we based on the systematics of Adl et al (Adl *et al*., 2012).

Determining the dominant species of amoebas was carried out by calculating the frequency of occurrence. According to the counting method, species are constant (occurrence in more than 50% of the collected samples), secondary (occurrence in 25-50% of the collected samples) and random (occurrence in less than 25% of the collected samples), calculated by the following formula:

$$pF = \frac{m}{n} \times 100\%,$$

where *m* is the number of samples with individuals of a certain species, n is the total number of collected samples.

3. Results and discussion

On the territory of the Lankaran natural area, we began to study the communities of freshwater testate amoebae in 2013. Later, we continued systematic working to

describe the diversity and structure of the community in the following water basins: Khanbulanachay reservoir, water bodies in Gumbashi, Dashtatuk and Azfilial settlements and Gamyshevka river in Lankaran region; Vilash reservoir, water bodies in Rudakanar, Miyanku settlements in Masalli district; Rvarud, Siyov, Piran settlements in Lerik district and Lankaranchay, as well as the right tributary of the river – Basharuchay; Goytepe reservoir, water bodies in Haziabad, Bajiravan settlements in Jalilabad district; water bodies in Avash, Chayuzu, Porsova settlements in Yardimli district; Astarachay, Tangerud river, water bodies in Mashkhan, Pensar, Gilekaran settlements in Astara region (Alekperov *et al*., 2017; Snegovaya & Tahirova, 2015; Tahirova, 2014a, 2014b, 2015).

A total of 126 species of testate amoebae belonging to 11 families were found in the studied water bodies. The number of species by registered families is given in the chart (Figure 1).

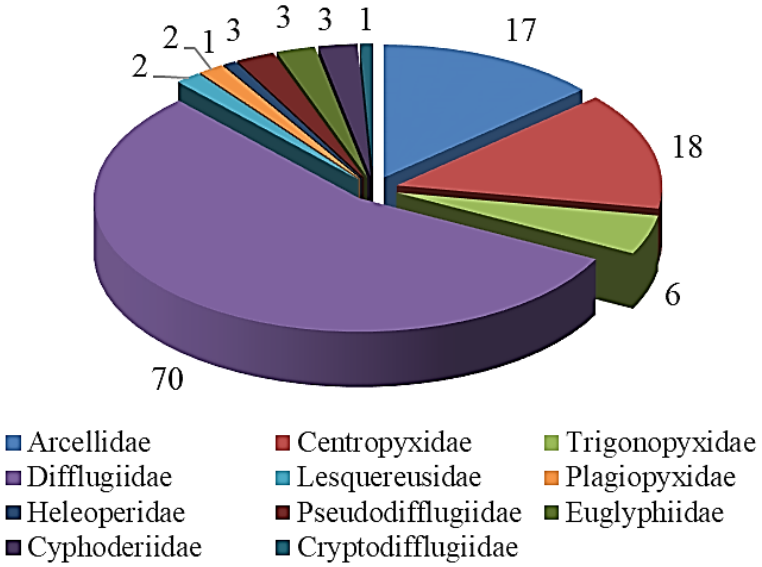

Figure 1. Species number of testate amoebae by the recorded families

The largest number of species among the studied ponds was recorded in the water body in Azfilial settlement (56 species), and among the rivers in Lankaranchay (24 species).

It is known that the difference in the temperature range in which testate amoebae are found affects the regularity of the seasonal distribution of their total number and species composition. In all studied water bodies, the highest quantitative growth of testate amoebae was observed in the warm time of the year.

During the researches in Lankaran region, the largest number of testaceans was noted in the water body near the Azfilal settlement (7.4 thousand ind./dm^2), and the least - in the water body in the Gumbashi region (4.6 thousand ind./dm^2). In all studied basins, the lowest total number was observed in winter, and in this period maximum number of testaceans was recorded in the water basin in Dashtatuk territory (0.9 thousand individuals/dm^2), and the minimum number was in the basin in Gumbashi settlement (0.3 thousand individuals/dm^2). The same regularity was observed in other districts within the Lankaran natural area as well.

In both winter and summer season, *D.gramen* was dominant among recorded

species in terms of number and biomass. In samples collected in winter, the number of this species was on average 16 individuals per 2 ml. It should be noted that although seasonal changes affect the species composition of testaceans, some species were found throughout the year: *C.aerophila, D.gramen, D.corona*.

During all years of the research (2013-2018), the highest biomass of testaceans was recorded in June 2014 (57.1 mg/dm^2), and the lowest in January 2015 (3.2 mg/dm^2).

Figure 2 shows the seasonal dynamics of the total biomass of testate amoebae in some studied water bodies. These data adequately reflect the results for the total number of teatate amoebae.

Average annual biomass of testaceans in the water body of Azfilial settlement of Lankaran region was 41.6 mg/dm^2. Calculations were made in May, July, October and January and the average biomass was determined. The average annual biomass of testaceans in Masalli region was 25.75 mg/dm^2, 24 mg/dm^2 in Lerik, 39 mg/dm^2 in Astara, 27.65 mg/dm^2 in Jalilabad, and 7.8 mg/dm^2 in Yardimli.

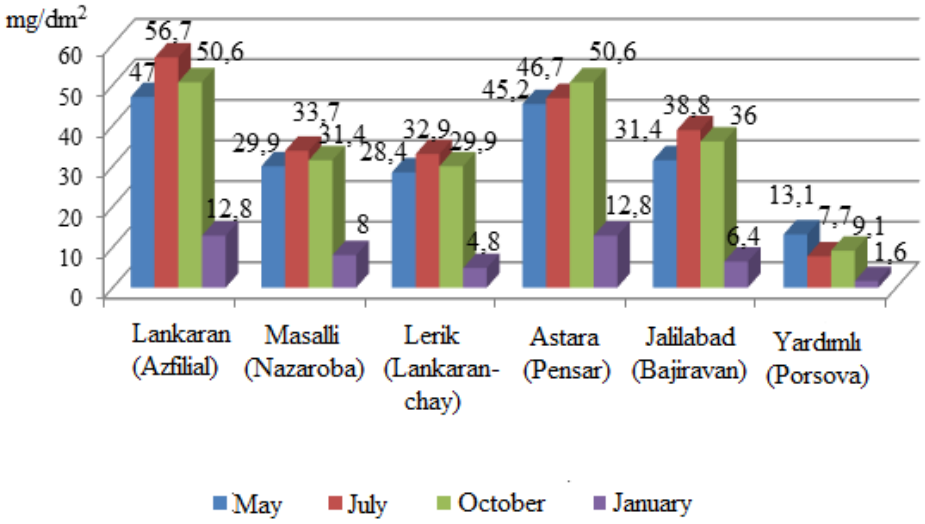

Figure 2. Quantitative indicators of testate amoebae biomass in some water bodies of the Lankaran natural area (2015)

As a result of the conducted research, we distinguished 2 groups according to the frequency of occurrence of species in different freshwater bodies: constant and secondary species. The following table provides information on the frequency of occurrence of the common species in the studied regions (Table 1).

Obviously the number of constant species is approximately the same in all 6 districts. According to our outcome based on the formula given in the "Materials and methods", the constant species are the species found in more than 50% of the collected samples. As follows, a total of 16 permanent species were recorded in various water bodies of Lankaran, Astara, Jalilabad regions, 14 in Masalli region, 13 and 12 in Yardimli and Lerik regions, respectively. The same regularity was observed in relation to secondary species. These species are found in 25-50% of the collected samples. The largest number of species belonging to this group was recorded in Lankaran region - 10 species. The least number of species was registered in Yardimli district with 2 species. In general, according to the compiled table, the total number of constant species is 18, and secondary species is 10.

Table 1. Frequency of occurrence of common species in the studied regions

Species	1	2	3	4	5	6
Constant species (>50%)						
Arcella arenaria	+	+	-	-	-	-
A.discoides	+	-	-	+	+	-
A.vulgaris	+	+	-	-	-	-
Cyclopyxis euristoma	+	+	+	+	+	+
Centropyxis aculeata	+	+	+	+	+	+
C.aerophila	+	-	+	-	+	-
D.acuminata	+	+	+	+	+	+
D.corona	+	+	+	+	+	+
D.difficilis	+	+	+	+	+	+
D.elegans	+	+	+	+	+	+
D.gramen	+	+	+	+	+	+
D.labiosa	+	+	-	-	+	-
D.limnetica	-	+	+	+	+	+
D.linearis	+	+	+	-	+	-
D.litophila	+	+	+	+	+	+
D.penardi	-	+	+	+	+	+
D.urceolata	+	+	+	+	+	+
Pontigulasia compressoidea	+	+	+	+	+	+
Total	16	16	14	13	16	12
Secondary species (=25-50%)						
Arcella dentata	+	-	-	-	-	-
A.gibbosa	+	-	+	-	-	-
Cyclopyxis elongata	+	+	+	+	+	+
C.kahli	+	-	+	-	+	-
D.claviformis	+	+	+	-	+	-
D.giganteacuminata	+	+	-	-	-	+
D.globulosa	+	+	-	-	-	+
D.lanceolata	+	-	+	-	+	-
D.microclaviformis	+	-	+	-	-	-
D.pyriformis	+	+	+	+	+	+
Total	10	5	7	2	5	4

1- Lankaran, 2- Astara, 3- Masalli, 4- Yardimli, 5- Jalilabad, 6- Lerik

The dominant species in the spring-summer communities of testate amoebas are *Centropyxis aculeata*, *Difflugia accuminata*, *D. corona*, *D. gramen*, *D. elegans* and their niche optimum has a wide range interval (Figure 3). However, these species are considered to be stable species that are tolerant to sharp changes in abiotic factors in the communities and reach their maximum in population growth rate and abundance.

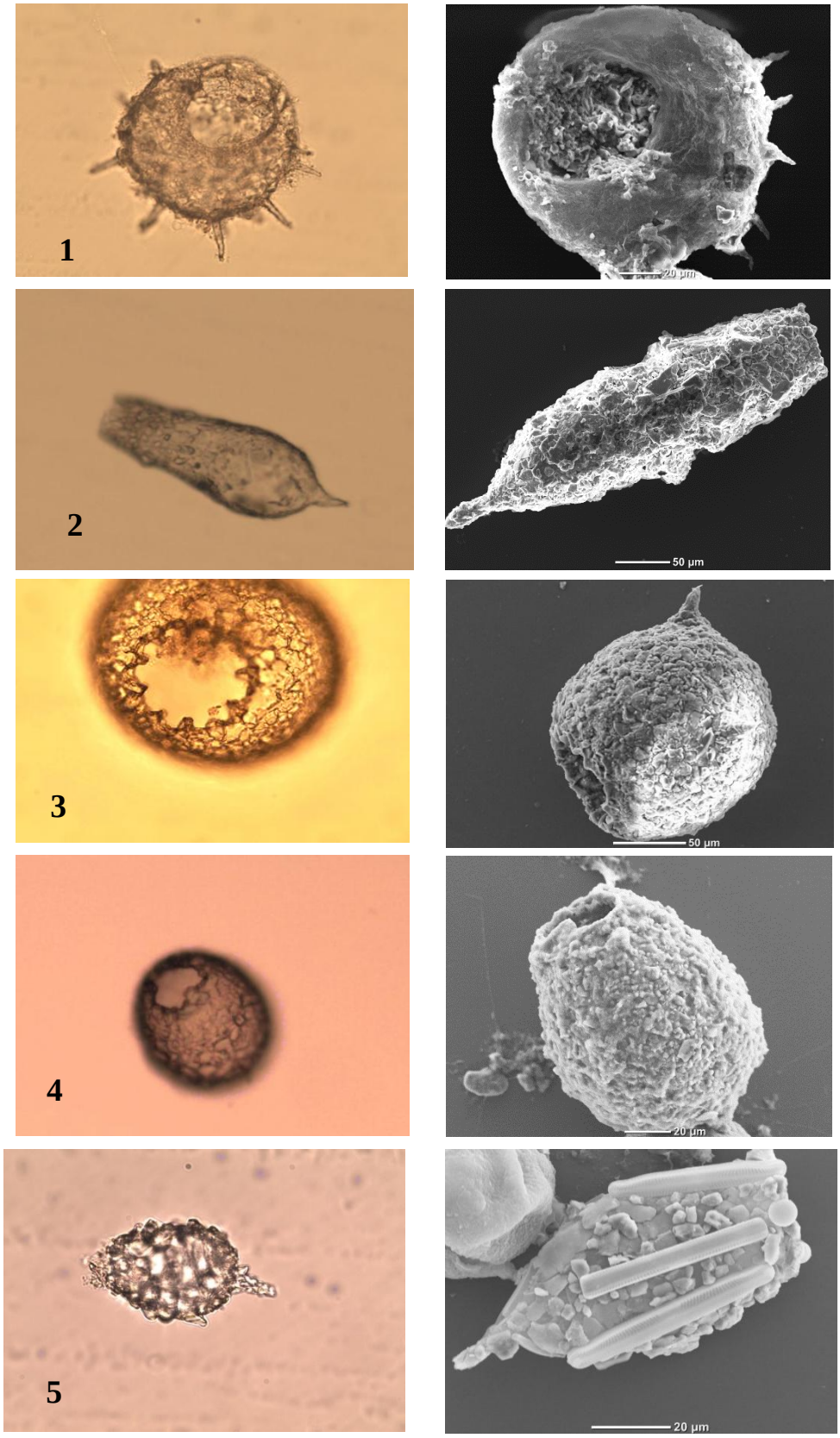

Figure 3. The dominant species in the spring-summer communities of testate amoebas (images obtained through Olympus light microscope and scanning electron microscope-SEM); *1-C.aculeata, 2-D.accuminata, 3-D.corona, 4-D.gramen, 5-D.elegans*

4. Conclusion

In seasonal changes, the spring–summer variant (May–July) of community of testate amoebae is most peculiar, where a complex of dominants is formed, including the species *Centropyxis aculeata, Difflugia accuminata*, *D.corona, D.gramen* and *D.elegans*. The autumn community (October–November) is very homogeneous and is characterized by the predominance of eurybionts.

Completely testacean fauna of freshwater basins of Lankaran region are generally divided into 2 complexes according to the occurrence frequency of species. Besides, the stagnant water ponds are characterized by a higher species diversity than rivers. This is due to the fact that the river biogeocenoses are not favorable for the formation of shells of amoebas and in the rivers floods occur frequently.

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