SPECIES COMPOSITION OF TESTATE AMOEBAE IN SUOI SIM RESERVOIR, KHANH HOA, VIETNAM

TRAN QUOC HOAN

1. INTRODUCTION

Testate amoebae are a group of rhizopoda, which is an important group of protist. They are observed in very different environments from soil to rivers, lakes, ponds, marsh, and even in the sea. They play an important role in the transfer of matter and energy through the ecosystem because they are component of "microbial loop" [5].

Species composition and distribution of testate amoebae in Vietnam have been still poorly studied. Currently, there are only five works about testate amoebae in Vietnam [1, 2, 6, 9, 11] but research areas are still small and scattered. However, the results of their works are not bad with revealing approximately 286 species, varieties and forms of testate amoebae in Vietnam [11]. Further works on testate amoebae in different regions in Vietnam not only could contribute new data about this group in the country, but also add new information for science.

Suoi Sim reservoir is important water body for agriculture in Ninh Sim commune (Khanh Hoa province). It is the main water resource for all living activities of local residents, including sugar-cane and tobacco irrigation. Despite that this reservoir is surrounded by farmlands, it is still kept in natural state. However, by the planning of province, Suoi Sim reservoir may be exploited, upgraded, renovated and expanded and its current state will be severely affected. This may lead to the change of testate amoeba community structure in the reservoir and the extinction of rare or new for science testate amoeba species. Thus, the study on species composition of testate amoebae in Suoi Sim reservoir is needed.

2. MATERIALS AND METHODS

Suoi Sim reservoir, which is about 50km northwest of Nha Trang city, covers approximately an area of 50 hectares. The climate of the studied region is tropical monsoon with mean annual temperature approximately 26°C. The dry season starts in January and lasts until August, while rainy season lasts from September to December.

Seven water and eight sediment samples were collected from Suoi Sim reservoir in 9th April 2016 (fig. 1). At each sampling point, both water and sediment samples were collected, excluding one point without water. Water samples were taken around aquatic plants, as water hyacinth, water spinach and water primrose (or water dragon). Geographical coordinates of sampling points were determined by Garmin GPSMAP 78sc and presented in table 1. Hydrochemical characteristics in sampling points were measured by Hanna HI 9828 (tab. 1).

All samples were fixed in 4% formalin and stored in the laboratory [4, 7]. Seven to ten replicates, 1 ml each, were analyzed in Petri dishes from each sample in order to study testate amoebae species composition. Testate amoebae were identified in drops of an aliquot of samples mixed with glycerol at a magnification of x200 or x400 using a light microscope Olympus CX41 equipped with Cannon EOS 650D. Identification of testate amoebae was carried out based on [3, 8, 10].

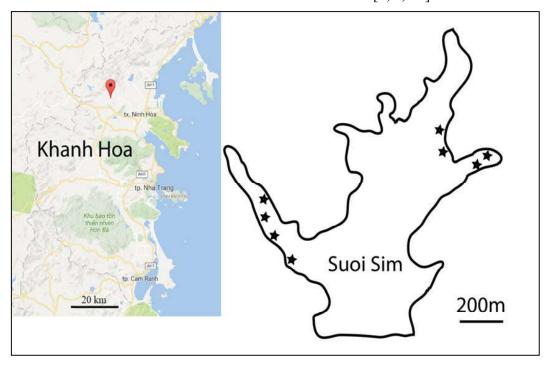


Figure 1. Location of studied site: black star - sampling points

The frequency of occurrence of testate amoebae was estimated as the percentage of samples in which particular species was found to the total number of samples from the reservoir. Data analysis was performed by MS Excel 2010.

3. RESULTS AND DISCUSSIONS

Water pH of the sampling points is relatively neutral. Dissolved oxygen (DO) and electrical conductivity (EC) are both low. The reason of low DO is that the samples were taken at near-shore points and in the dry season, so water transfer is low. However, DO seems to be not a limited factor for testate amoeba distribution, because they can distribute in deep-water sediment and in the soil where DO has low value.

Table 1. Basic hydrochemical characteristics of sampling points

№	Coordinates	Water temperature (°C)	pН	Dissolved oxygen, ppm	Electrical conductivity, µS/cm	Salinity,
1	N 12° 34.54' E 109° 01.00'	28.3	6.47	0.30	97	0.04
2	N 12° 34.58' E 109° 01.02'	29.8	6.79	2.22	73	0.03
3	N 12° 34.49' E 109° 01.03'	30.6	6.62	1.46	38	0.02
4	N 12° 34.44' E 109° 01.08'	31.5	6.73	1.71	39	0.02
5	N 12° 34.64' E 109° 01.58'	29.1	6.48	1.31	41	0.02
6	N 12° 34.69' E 109° 01.50'	32.6	6.95	4.26	44	0.02
7	N 12° 34.74' E 109° 01.49'	32.5	6.98	5.75	42	0.02
8	N 12° 34.69' E 109° 01.59'	Not measured	Not measured	Not measured	Not measured	Not measured

The total list of testate amoebae identified from 15 samples in Suoi Sim reservoir includes 46 species, varieties and forms from 4 families and 7 genera (tab. 2). Two testate amoebae were not identified to species level. They may be new species or new varieties for science and further works should be done on them to reveal more information. The species diversity of *Difflugia* was the highest with 24 taxa. The species diversity of genera *Arcella* and *Centropyxis* also was high with 10 and 7 taxa, respectively. A number of testate amoeba planktonic species is clearly higher than the number of benthic species. It may be due to that benthic samples were taken in clay-rich or gravel sediments with low concentration of organic matter, where testate amoebae are not often observed.

Centropyxis aculeata, Difflugia corona, Difflugia elegans, Difflugia elegans angustata, Difflugia lobostoma, Difflugia sp.2 and Lesquereusia spiralis were the most common testate amoeba planktonic species in Suoi Sim reservoir. Other species, as Arcella discoides, Arcella vulgaris penardi, Centropyxis ecornis, Difflugia capreolata, Difflugia levanderi, Difflugia limnetica, Difflugia parva and Difflugia penardi, were also observed with high occurrence (57.1%). By contrast, Centropyxis aerophila sphagnicola, Centropyxis constricta and Difflugia sp.1 were not found in the water column, but observed in sediment. Centropyxis aculeata and Difflugia corona were popular in sediment. Some rare species were observed in the reservoir: Difflugia levanderi, Difflugia serbica, Cucurbitella modesta. Therein, two last species were found in Vietnam for the first time. Some other species were also identified in Vietnam for the first time: Difflugia gramen, Difflugia leidyi, Difflugia lingula regularis, Difflugia urceolata lageniformis, Difflugia venusta.

Table 2. The frequency of occurrence (%) of testate amoebae from benthic and planktonic samples of Suoi Sim reservoir

No	Species	Benthos	Plankton
	Arcellidae Ehrenbeg, 1843		1 1001110011
1	Arcella discoides Ehrenberg, 1843	0	57.1
2	Arcella discoides pseudovulgaris Deflandre, 1928	0	14.3
3	Arcella discoides scutelliformis Playfair, 1918	0	42.8
4	Arcella hemisphaerica Perty, 1852	0	28.6
5	Arcella intermedia (Deflandre, 1928) Tsyganov, Mazei, 2006	25	14.3
6	Arcella intermedia laevis (Deflandre, 1928) Tsyganov, Mazei, 2006	0	28.6
7	Arcella mitrata spectabilis Deflandre,1928	0	14.3
8	Arcella vulgaris Ehrenberg, 1830	0	14.3
9	Arcella vulgaris penardi Deflandre 1928	0	57.1
10	Arcella vulgaris wailesi Deflandre 1928	0	14.3
	Centropyxidae Jung, 1942		
11	Centropyxis aculeata (Ehrenberg, 1832) Stein, 1857	50	100
12	Centropyxis aerophila sphagnicola Deflandre, 1929	12.5	0
13	Centropyxis cassis (Wallich, 1864) Deflandre, 1929	0	14.3
14	Centropyxis constricta (Ehrenberg, 1841) Deflandre, 1929	12.5	0
15	Centropyxis ecornis (Ehrenberg, 1841) Leidy, 1879	12.5	57.1
16	Centropyxis laevigata Penard, 1890	0	28.6
17	Cyclopyxis kahli Deflandre, 1929	25	14.3

	Difflugiidae Wallich, 1864		
18	Difflugia ampullula Playfair, 1918	0	42.8
19	Difflugia brevicolla Cash et Hopkinson, 1909	0	14.3
20	Difflugia capreolata Penard, 1902	0	57.1
21	Difflugia corona Wallich, 1864	37.5	71.4
22	Difflugia curvicaulis Penard, 1899		14.3
23	Difflugia difficilis Thomas, 1954	0	14.3
24	Difflugia elegans Penard, 1890		71.4
25	<i>Difflugia elegans angustata</i> Gauthier-Lievre et Thomas, 1958	0	100
26	*Difflugia gramen Penard, 1902	0	42.8
27	Difflugia lanceolata Penard, 1890	0	28.6
28	*Difflugia leidyi Wailes, 1912	0	42.8
29	Difflugia levanderi Playfair, 1918	0	57.1
30	Difflugia limnetica Penard, 1902	37.5	57.1
31	*Difflugia lingula regularis Gauthier-Lievre et	0	14.3
22	Thomas, 1958	2.5	100
32	Difflugia lobostoma Leidy, 1879	25	100
33	Difflugia oblonga Ehrenberg, 1838	0	14.3
34	Difflugia parva (Thomas, 1954) Ogden, 1983	0	57.1
35	Difflugia penardi Hopkinson, 1909	0	57.1
36	Difflugia schurmani van Oye, 1932	25	28.6
37	Difflugia serbica Ogden et Zivkovic, 1983	12.5	28.6
38	Difflugia sp.1	25	0
39	Difflugia sp.2	0	85.7
40	Difflugia urceolata lageniformis	0	42.8
41	*Difflugia venusta (Penard, 1902) Ogden, 1983	0	14.3
42	*Cucurbitella modesta Gauthier-Lievre et Thomas, 1960	0	14.3
	Lesquereusidae Ogden, 1979		
43	Lesquereusia modesta Rhumbler, 1895	0	57.1
44	Lesquereusia spiralis (Ehrenberg, 1840) Butschli	12.5	71.4
45	Netzelia oviformis (Cash, 1909) Ogden, 1979	0	42.8
46	Netzelia wailesi (Ogden, 1980) Meisterfeld, 1984	0	14.3
	Number of samples examined	8	7
	Number of species, varieties and forms identified	15	43
	Total observed species, varieties and forms in this work	46	

Note: * - new taxa for Vietnam

Like Bau Sen and Bau Trang lake [11] (Binh Thuan province, Vietnam), in Suoi Sim reservoir, the frequency of occurrence of *Centropyxis aculeata* was the highest. In other work [2], the frequency of occurrence of this species in Bau Sau lake was also high. Due to that these lakes and reservoir are located in different regions, it indicates that *Centropyxis aculeata* is most popular in water bodies in Vietnam.

Suoi Sim reservoir has own testate amoeba composition. Many species of the reservoir have been not observed in Bau Sau, Bau Sen, and Bau Trang lakes. In addition, the occurrence of testate amoeba species in the reservoir is also differed from the studied lakes. Because there are only few samples taken, the identified species of Suoi Sim reservoir may be only a small part of the unknown composition of testate amoeba community. Further works should be done to reveal more testate amoeba species in this reservoir.

It is worth to mention, that most observed testate amoeba species in Suoi Sim reservoir were in dead form. It indicates that in other time, they have lived here, but the change of habitats may lead to their death. The change of the reservoir environments might be occurred due to some activities of local residents. In the agricultural activity, they use many fertilizers and chemical compounds which in the end are discharged into the reservoir. Moreover, the reservoir upgrade and renovation also lead to the change of natural habitats for testate amoebae.

4. CONCLUSIONS

- Forty-six species, varieties and forms from 4 families and 7 genera were identified from Suoi Sim reservoir, Khanh Hoa, Vietnam. Two testate amoebae were not identified to species level.
- The species richness of *Difflugia, Arcella* and *Centropyxis* were the highest. Many species were observed with high occurrence. Some species which are global rare and new for Vietnam were found.
- More intensive investigations of testate amoebae should be expected to reveal more species in Suoi Sim reservoir.

Acknowledgements

The author would like to thank Dr Denis V. Tikhonenkov for fruitful discussions, Dr Elena Malysheva and Mr. Ferry Siemensma for their help in the testate amoeba identification, and Vu T. Dao, Dien D. Tran for their help in the material collection. This study was supported by the Vietnamese-Russian Tropical Centre (Grant number 302/QD-CNVB).

REFERENCES

- 1. Balik V., Testate amoebae (Protozoa: Rhizopoda) from a primary mountain rain forest in the Tam-Dao region (Vietnam), Acta Soc. Zool. Bohem, 1995, **59**:1-16.
- 2. Bobrov A. A., Mazei Y. A., Tiunov A. V., *Testate amoebae of a monsoon tropical forest of South Vietnam*, Acta Protozool., 2010, **49:**311-325.
- 3. Charman D. J., Hendon D. and Woodland W. A., *The identification of testate amoebae (Protozoa: Rhizopoda) in peats*, QRA Technical Guide No. 9, Quaternary Research Association, London, 2000, 147 p.
- 4. Davidova R., Biometry of three rare testate amoebae species (Arcellinida and Euglyphida) from freshwater and moss biotopes in Bulgaria, Protistology, 2012, 7(2):63-70.
- 5. Gilbert D., Amblard C., Bourdier G., Francez A. J., *The microbial loop at the surface of a peatland: structure, function, and impact of nutrient input*, Microbial Ecology, 1998, **35**(1):83-93.
- 6. Golemansky V., Psammobiotic thecamoebas of the Vietnamese coast of the China sea with description of Cryptodifflugia brevicolla new species Rhizopoda Arcellinida, Acta Protozoologica, 1979, **18**(2):285-292.
- 7. Malysheva E. A., Mazei Yu. A., Yermokhin M. V., *Testate Amoebae Community Pattern in Different Types of Boundary Structures at the Water-Land Contact Zone*, Biology Bulletin, 2013, **40**(10):823-831
- 8. Mazei Yu., Tsyganov A., *Freshwater testate amoebae*, Moscow, KMK Sci Press, 2006, 300 p. (in Russian).
- 9. Nguyen V. H., Bernard N., Mitchell E. A. D., Cortet J., Badot P. M., Gilbert D., Relationship between testate amoeba (Protist) communities and atmospheric heavy metals accumulated in Barbula indica (Bryophyta) in Vietnam, Microbial Ecology, 2007, 53:53-65.
- 10. Siemensma F. J., *Microworld, world of amoeboid organisms*, World-wide electronic publication, Kortenhoef, the Netherlands, 2016, http://www.arcella.nl.
- 11. Tran Q. H., Nguyen T. H. T., *Testate amoeba in Bau Sen and Bau Trang lakes, Binh Thuan province, Vietnam.* International Scientific Forum «PROTIST-2016». Protistology. An International Journal, 2016, **10**(2):83.

TÓM TẮT

THÀNH PHẦN LOÀI TRÙNG CHÂN GIẢ CÓ VỎ TẠI HỒ CHỨA SUỐI SIM, KHÁNH HÒA, VIỆT NAM

Thành phần loài trùng chân giả có vỏ tại hồ chứa Suối Sim (tỉnh Khánh Hòa, Việt Nam) lần đầu tiên được tiến hành nghiên cứu. 46 loài, thứ và dạng trùng chân giả có vỏ thuộc 4 họ và 7 giống đã được phân loại từ các mẫu nước và nền đáy. Số lượng loài trùng chân giả có vỏ dạng phù du cao hơn rõ rệt số lượng loài trùng chân giả có vỏ dạng sống đáy. Đa dạng loài của giống Difflugia là cao nhất với 24 taxa. Các loài Centropyxis aculeata, Difflugia corona, Difflugia elegans, Difflugia elegans, Difflugia lobostoma, Difflugia sp.2 và Lesquereusia spiralis là những loài thường gặp trong số các loài trùng chân giả có vỏ dạng phù du. Đồng thời, Centropyxis aculeata và Difflugia corona cũng phổ biến trong nền đáy. Trong nghiên cứu này, một số loài hiếm gặp và một số loài mới đối với Việt Nam đã được phát hiện. Những nghiên cứu tiếp theo cần được tiến hành để tìm ra thêm các loài trùng chân giả có vỏ trong hồ chứa này.

Từ khóa: Trùng chân giả có vỏ, hồ chứa Suối Sim, Arcella, Centropyxis, Difflugia, Testate amoeba, Suoi Sim reservoir, Arcella, Centropyxis, Difflugia.

Nhận bài ngày 11 tháng 10 năm 2016 Hoàn thiện ngày 03 tháng 11 năm 2016

Chi nhánh Ven biển, Trung tâm Nhiệt đới Việt - Nga