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Species of the *Paramecium aurelia* Complex in Russia: New Stands and Overall Distribution

Alexey POTEKHIN, Ewa PRZYBOŚ, Irina NEKRASOVA, Varvara YASHCHENKO and Maria RAUTIAN

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New stands of *Paramecium biaurelia, P. triaurelia, P. tetraurelia, P. pentaurelia, P. novaurelia*, and *P. dodecaurelia* were recorded in Russia. Especially interesting is the record of *P. novaurelia* in Vladivostok, Russian Far East, as it is a very rare species outside of Europe. The distribution of species of the *Paramecium aurelia* complex in Eurasia with emphasis on findings in Russia is discussed.

Key words: *Paramecium aurelia* species complex, distribution of species, species expansion, biogeography of protists.

Alexey POTEKHIN, Irina NEKRASOVA, Varvara YASHCHENKO, Maria RAUTIAN, Faculty of Biology and Soil Science, St. Petersburg State University, Oranienbaumskoye shosse 2, 198504 St. Petersburg, Russia.
E-mail: Alexey Potekhinspbu-tox@yandex.ru
Ewa PRZYBOŚ, Department of Experimental Zoology, Institute of Systematics and Evolution of Animals, Polish Academy of Sciences, Slawkowska 17, 31-016 Kraków, Poland.
E-mail: Przybos@isez.pan.krakow.pl

Debates on the concept of protist biogeography are ensuing in the field of microbial ecology (FINLAY & FENCHEL 2004; FOISSNER 2006; BASS *et al*. 2007). Neither of two opposing views – "ubiquitous dispersal hypothesis" or "endemism hypothesis" at the moment can gain an advantage, as currently available information on the distribution of free-living protozoan species worldwide remains scarce. The lack of data is to a great extent due to uneven sampling in different parts of the world, as well as to difficulties in the identification of many unicellular organisms. Thus, extensive sampling of protozoa readily identified to the species level seems to be one of the promising approaches in protist zoogeography. One of the best candidates for such an investigation is *Paramecium* – a genus of ciliates which includes a number of well-known morphological species; they are subdivided into syngens, which in some cases are equivalent to genetic species, such as sibling species of the *Paramecium aurelia* complex (SONNEBORN 1975). Because one of the speciation mechanisms by the presence of geographic barriers, the geographic distribution of evolutionary young sibling species of the *P. aurelia* complex is of great interest.

At present 15 species of the *P. aurelia* complex are known worldwide (SONNEBORN 1975; AUFDERHEIDE *et al*. 1983). Some are cosmopolitan, while others are limited to certain regions or are just extremely rare, known from single populations (cf. SONNEBORN 1975; PRZYBOŚ & FOKIN 2000; PRZYBOŚ 2005; PRZYBOŚ *et al.* 2008a). Most of the data on frequency and overall distribution of *P. aurelia* species were collected in the last 50 years, mainly in North America (SONNEBORN 1975) and in Europe (PRZYBOŚ *et al*. 2008a). The third continent of the Northern hemisphere – Asia – remained much less sampled: extensive sampling has been performed only in Japan (KOŚCIUSZKO & KOIZUMI 1984; PRZYBOŚ & FOKIN 2001b; PRZYBOŚ *et al*. 2003). Some findings of *P. aurelia* species were scattered episodically in different parts of this enormous territory – Southeast Asia (Thailand, Vietnam, India, China), Central Asia (Turkmenistan), and the Near East (Israel, Lebanon, Turkey) (see PRZYBOŚ & FOKIN 2001a for a summary; PRZYBOŚ *et al*. 2007c). Russia, with European and Asian parts to its territory, until recently was a

large white spot on the map of the *P. aurelia* species distribution in the Northern hemisphere. All data on the occurrence of *P. aurelia* species in the Asian part were scarce and random, as only some places were sampled in Eastern Siberia and the Russian Far East, and the presence of *P. primaurelia* and *P. biaurelia* (PREER *et al*. 1974; DAGGETT 1978; KOŚCIUSZKO 1985; PRZYBOŚ & FOKIN 1996), and *P. dodecaurelia* (PRZYBOŚ *et al*. 2008b), was revealed. Recently, the West Siberian Lowland, the Altai Mountains, and the Altai Foreland were representatively sampled, and the presence of *P. primaurelia*, *P. biaurelia*, *P. triaurelia*, and *P. pentaurelia* was recorded (POTEKHIN *et al*. 2006). The European part of Russia has been studied more extensively, especially in recent years: the presence of ten of fifteen species of the complex (*P. primaurelia*, *P. biaurelia*, *P. triaurelia*, *P. tetraurelia*, *P. pentaurelia*, *P. sexaurelia*, *P. septaurelia*, *P. novaurelia*, *P. decaurelia*, *P. dodecaurelia*) was recorded (cf. KOMALA & DUBIS 1966; KOŚCIUSZKO 1985; FOKIN & OSSIPOV 1986; PRZYBOŚ & FOKIN 1996; PRZYBOŚ *et al*. 2004, 2005a; PRZYBOŚ *et al*. 2006; PRZYBOŚ *et al*. 2007b; PRZYBOŚ *et al*. 2007/2008; PRZYBOŚ *et al*. 2008b ; POTEKHIN *et al*. 2008). Still, different regions were unevenly sampled: random sampling was carried out near St. Petersburg, Kaliningrad, Vladimir, Moscow, Belgorod, and in the Black Sea region. Complex studies, when more than a hundred water samples were taken from one geographical territory, were carried out in the Upper Volga region – in the Vologda region, Yaroslavl region, Moscow region (POTEKHIN *et al*. 2008) and in the Lower Volga regions: the Astrakhan Nature Reserve and in Volga-Akhtuba flood lands of the Volgograd region (PRZYBOŚ *et al*. 2004, 2005a).

At present new stands of the *P. aurelia* species in European (Kaliningrad, St. Petersburg, Novgorod, Moscow, Yaroslavl regions) and Asian parts of Russia (Central and Southeastern Siberia and the Russian Far East) are described.

Material and Methods

Material

Water samples with plankton were collected through several years in July-September from different kinds of water reservoirs (ponds, small rivers, streams, lakes, ditches, flooded sandpits). The total volume of each sample was 15-40 ml. Paramecia were isolated from the whole sample volume, and clones were established. The collecting sites and indexes for newly isolated strains of the *P. aurelia* complex are presented in Table 1.

Methods

Culture and identification of paramecia were performed according to SONNEBORN (1970). Paramecia were cultivated on a lettuce medium inoculated with *Enterobacter aerogenes*. Species of the *P. aurelia* complex were identified by mating the investigated strains with the mating types of standard strains of particular species of the complex. The following standard strains were used:

P. biaurelia, strain Rieff from Scotland,

P. triaurelia, strain 324 from Florida, USA,

P. tetraurelia, strain from Sydney, Australia,

P. pentaurelia , strain 87 from Pennsylvania, USA,

P. octaurelia, strain 138, from Florida, USA,

P. novaurelia, strain 510 from Scotland, UK,

P. dodecaurelia, strain 246 from Mississippi, USA.

Results and Discussion

New stands of five species of the *P. aurelia* complex were found in Russia (Table 1). The overall records of species of the *P. aurelia* complex in Russia are summarized in Table 2; regions where sampling was performed and any records exist are shown in Figure 1.

P. biaurelia was recorded in the environs of St. Petersburg and Moscow as well as in Central Siberia (Kemerovo region). It is a cosmopolitan species and one of the most abundant species of the *P. aurelia* complex.

P. triaurelia was found in habitats situated in Southeastern Siberia (Khabarovsk) and in the Russian Far East, in localities at the shore of the Pacific ocean, the so called Primorye (Vladivostok and Slavyanka). This is the most eastern record of this species – previously it was known to occur in Eurasia all over Europe and in Central Siberia (Krasnoyarsk).

P. tetraurelia was recorded in one population sampled in a pond in Kaliningrad. This species is very rare in Russia, as it was earlier known only from the Black Sea region (Novorossiysk). In the same sample (designated KD) another species – *P. pentaurelia* was found. The latter species was also registered in Staraya Russa near Novgorod.

The finding of a new stand of *P. novaurelia* in Vladivostok, Russian Far East is of interest. *P. novaurelia* is the most common species in Europe and for a long time it was thought to be restricted to this continent (SONNEBORN 1975). Then a single population was found in Turkey (PRZYBOŚ 1998), which nominally belongs to Asia; this finding was not unexpected, as this territory is geographically

Table 1

New stands of the *Paramecium aurelia* species in Russia

Strain index	Geographic origin	Kind of habitat	Identified as *P. aurelia* species
KD-7	*Kaliningrad*	*Town pond*	*P. pentaurelia*
KD9	Kaliningrad		*P. tetraurelia*
KD5	Kaliningrad		*P. tetraurelia*
KD8	Kaliningrad		*P. tetraurelia*
SPb 35-2	St. Petersburg	Small river	*P. biaurelia*
SPb 35-3	St. Petersburg		*P. biaurelia*
PA-1	St. Petersburg	Pond in the park	*P. biaurelia*
PA-4	St. Petersburg		*P. biaurelia*
PA-5	St. Petersburg		*P. biaurelia*
LR 7-5	Luga, St. Petersburg region	Roadside pool	*P. biaurelia*
NR 28-5	Staraya Russa, Novgorod region	Pond	*P. pentaurelia*
Mya 156-5	Myshkin, Yaroslavl region	Small river	*P. dodecaurelia*
TD 77-4	Dmitrov, Moscow region	Small river	*P. biaurelia*
TD 77-5	Dmitrov, Moscow region	Small river	*P. biaurelia*
TD 80-3	Dmitrov, Moscow region	Small river	*P. biaurelia*
TD 80-4	Dmitrov, Moscow region		*P. biaurelia*
TD 80-5	Dmitrov, Moscow region		*P. biaurelia*
Ant 101-2	Antibes, Kemerovo region, Central Siberia	Flooded sandpit	*P. biaurelia*
Ant 101-6	Antibes, Kemerovo region, Central Siberia		*P.biaurelia*
Ant 101-13	Antibes, Kemerovo region, Central Siberia		*P. biaurelia*
ShKm 1	Shestakovo, Kemerovo region, Central Siberia	River Kiya	*P. biaurelia*
ShKm4	Shestakovo, Kemerovo region, Central Siberia		*P. biaurelia*
PrS 142-2	Slavyanka, Primorye	Stream flowing to the Pacific Ocean, close to the mouth	*P. triaurelia*
PrS 142-4	Slavyanka, Primorye		*P. triaurelia*
PrS 142-5	Slavyanka, Primorye		*P. triaurelia*
PrS 142-6	Slavyanka, Primorye		*P. triaurelia*
PrS 142-7	Slavyanka, Primorye		*P. triaurelia*
Vv 171-5	Vladivostok, Primorye	Pond in the park	*P. triaurelia*
Vv171-18	Vladivostok, Primorye		*P. novaurelia*
Hb 179-5	Khabarovsk, South-Eastern Siberia	City pond	*P. triaurelia*

Mentioned in the text

Gaq2	Gelendzhik	Pet shop, aquarium	*P. octaurelia*
Gaq7	Gelendzhik	Pet shop, aquarium	*P. octaurelia*

Fig. 1. Map of sampled territories of Russia. Regions where sampling was performed are marked with rectangles. 1 – Northwestern region (St. Petersburg region, Kaliningrad region, Novgorod region, Vologda region, Komi Republic); 2 – Central Russia (Moscow region, Vladimir, Tver region, Yaroslavl region); 3 – Southern Russia (Belgorod region; Black Sea region); 4 – Lower Volga region (Volgograd region, Astrakhan region); 5 – Western Siberia (Novosibirsk, Omsk) and Altai; 6 – Central Siberia (Kemerovo region, Krasnoyarsk region); 7 – Baikal and Transbaikalia (Irkutsk, Ulan-Ude); 8 – Russian Far East (Khabarovsk, Primorye, Sakhalin, Kamchatka). Bold line shows the border between European and Asian parts of Russia.

Table 2

Occurrence of species of the *P. aurelia* complex in Russia

Part of Russia	*P. aurelia* species	Region	Climate (temperature zone)	Number of populations	Reference
European	*P. primaurelia*	St. Petersburg Moscow Astrakhan region Volgograd region Black Sea region	Cold Cold Moderate Moderate Warm	3 5 5 1 1	KOMALA & DUBIS, 1966; PRZYBOŚ *et al*. 2004, 2005a; PRZYBOŚ *et al*. 2007/2008
	P. biaurelia	St. Petersburg St Petersburg region Tver region Moscow Moscow region Yaroslavl region Vologda region Komi republic Astrakhan region Volgograd region	Cold Cold Cold Cold Cold Cold Cold Cold Moderate Moderate	4 2 3 1 3 3 1 1 1 1	KOMALA & DUBIS, 1966; PREER *et al*. 1974; PRZYBOŚ & FOKIN 1996; PRZYBOŚ *et al*. 2005a; POTEKHIN *et al*. 2008; current paper
	P. triaurelia	Vologda region Astrakhan region Volgograd region	Cold Moderate Moderate	1 1 3	KOŚCIUSZKO 1985; PRZYBOŚ *et al*. 2004, 2005a; POTEKHIN *et al*. 2008
	P. tetraurelia	Kaliningrad region Black Sea region	Cold Warm	1 1	PRZYBOŚ *et al*. 2007/2008; current paper
	P. pentaurelia	Kaliningrad region Novgorod region Belgorod region Astrakhan region Volgograd region Black Sea region	Cold Cold Moderate Moderate Moderate Warm	1 1 1 5 1 1	FOKIN & OSSIPOV 1986; PRZYBOŚ *et al*. 2004, 2005a; PRZYBOŚ *et al*. 2007/2008; current paper
	P. sexaurelia	Astrakhan region	Moderate	5	PRZYBOŚ *et al*. 2004, 2005a
	P. septaurelia	Astrakhan region Volgograd region	Moderate Moderate	11 1	PRZYBOŚ *et al*. 2004, 2005a
	P. novaurelia	St. Petersburg St Petersburg region Moscow Kaliningrad region Vladimir Astrakhan region Volgograd region	Cold Cold Cold Cold Cold Moderate Moderate	3 1 4 2 1 1 1	KOMALA & DUBIS, 1966; KOŚCIUSZKO 1985; PRZYBOŚ *et al*. 2005a; PRZYBOŚ *et al*. 2006
	P. decaurelia	Yaroslavl' region Volgograd region	Cold Moderate	5 1	PRZYBOŚ *et al*. 2007d; POTEKHIN *et al*. 2008
	P. dodecaurelia	Yaroslavl' region Vologda region	Cold Cold	2 1	PRZYBOŚ *et al*. 2008b; current paper
Asian	*P. primaurelia*	Omsk region Kamchatka	Cold Cold	2 1	DAGGETT 1978; POTEKHIN *et al*. 2006
	P. biaurelia	Altai Mountains Kemerovo region Krasnoyarsk Irkutsk Primorye Sakhalin	Cold Cold Cold Cold Moderate Moderate	2 2 2 1 1 1	PREER *et al*. 1974; KOŚCIUSZKO 1985; PRZYBOŚ & FOKIN 1996; POTEKHIN *et al*. 2006; current paper
	P. triaurelia	Krasnoyarsk Khabarovsk Primorye	Cold Moderate Moderate	3 1 2	POTEKHIN *et al*. 2006; current paper
	P. pentaurelia	Altai Mountains Altai Foreland Novosibirsk	Cold Cold Cold	1 2 1	POTEKHIN *et al*. 2006
	P. novaurelia	Vladivostok	Moderate	1	current paper
	P. decaurelia	Altai Mountains	Cold	1	PRZYBOŚ *et al*. 2007b
	P. dodecaurelia	Ulan-Ude	Cold	1	PRZYBOŚ *et al*. 2008b

very close to Europe and shares climate peculiarities with the countries of Southern Europe, and is not separated by any natural barrier. Recently, this species was recorded in one population in North America (USA, Boston; PRZYBOŚ *et al*. 2007a); and here we report its first stand in Eastern Asia.

The presence of *P. dodecaurelia* in the Upper Volga region (Myshkin) (cf. PRZYBOŚ *et al.* 2008b) was confirmed at present.

Some interesting tendencies may be observed if the data on the *P. aurelia* species distribution and frequency in Russia are included into the total distribution of these sibling species in Eurasia. Among the most abundant species, *P. primaurelia* is extremely widespread and does not demonstrate defined preferences for climate type, as specimens were recorded all over Europe, in different regions of Russia, in Japan, Vietnam, Turkmenistan, Israel. *P. biaurelia* was considered by SONNEBORN (1975) as a "cosmopolitan, chiefly in moderate to cold climates, where it is commonly found", and the recent data support this conclusion. Although this species is the most frequent in habitats with a chilly, sometimes even rigorous climate, it becomes rarer in warmer environments. *P. novaurelia*, very common all over Europe, is also regularly observed in the European part of Russia, but there are only two findings of this species to the east of the Ural Mountains (Russian Far East, this paper) or in other continents (USA, PRZYBOŚ *et al.* 2007a). This situation remains enigmatic.

As to the species which are less frequent in nature, *P. triaurelia* is distributed in all climatic zones, although its occurrence in Vologda and Krasnoyarsk regions with inclement winter does not argue against SONNEBORN's (1975) inference that "this species seems to be mainly confined to the temperate and cold regions". *P. tetraurelia* has been previously found in Russia only in the Black Sea region (PRZYBOŚ *et al*. 2007/2008), and here we report it from Kaliningrad; it is a common European and American species, known also from Peru and Australia; in Asia it was revealed in Japan, India, and Israel (PRZYBOŚ & FOKIN 2000; PRZYBOŚ *et al*. 2009a) but it was never found in territories with a continental climate, from Central Russia to Eastern Siberia, or, i.e., in Canada (SONNEBORN 1975). Two new records of *P. pentaurelia* in the Novgorod region and in Kaliningrad are the first records of this species at such high latitudes – previously it was collected mostly from territories with a warm climate or at least with very hot summers and severe but short winters (i.e. Astrakhan region). Nevertheless, several strains of *P. pentaurelia* were also isolated earlier from samples taken in Western Siberia, which is characterized by a continental climate with a long and cold winter; thus, new findings support the viewpoint that *P. pentaurelia* is not really a species with exclusive temperature-limited occurrence. The distribution of *P. sexaurelia* by SONNEBORN (1975) "…appears prevailingly to be tropical but extends into the temperate zone (in the USA)" in Europe this species was recorded as far north as Germany, in Russia – in Astrakhan and Volgograd regions with a relatively warm climate, and in Asia it is known only from the southeast – India, Thailand, Japan (PRZYBOŚ & FOKIN 2001a) and China (PRZYBOŚ *et al*. 2007c). So this species, in our opinion, can be considered as temperature-restricted, as well as the next two species: *P. septaurelia* and *P. octaurelia*. *P. septaurelia* was for a long period of time considered as only American (found in Florida and Alabama, SONNEBORN 1975) until it was found by us in the Lower Volga region of Russia (PRZYBOŚ *et al*. 2004, 2005a) and in one locality in Germany (PRZYBOŚ *et al*. 2005b). Still, this species is very rare in nature and seems to be restricted even to specific habitats in the warm zone. *P. octaurelia* is a species rarely found in different parts of the world, known before mainly from the USA, and also single strains from Panama and Uganda (SONNEBORN 1975); later it was recorded in Israel (PRZYBOŚ *et al*. 2002), and recently in Europe in Germany (PRZYBOŚ *et al*. 2009). Now we recovered *P. octaurelia* (Table 1) in the water samples collected in an aquarium in a pet shop in Gelendzhik (Black Sea region, Russia); unfortunately, we cannot put the origin of these strains on a map, but perhaps the ciliates were transferred to the aquarium with water plants or with tropical aquarium fishes. According to SONNEBORN (1975), "This species is .. common in the tropical and subtropical Americas and may be so around the world".

Two more species rarely recorded in Russia are *P. decaurelia* and *P. dodecaurelia*. *P. decaurelia* is a very rare species, single populations were found in Florida, USA (SONNEBORN 1975), Japan (PRZYBOŚ *et al*. 2003) and in two regions of Russia, both characterized by a cold climate, Yaroslavl region and Altai Mountains (PRZYBOŚ *et al*. 2007b). Thus, it seems that the rarity of this species is not connected with its climatic preferences. *P. dodecaurelia* was recorded in the Yaroslavl and Vologda regions, and in Transbaikalia of Eastern Siberia (PRZYBOŚ *et al*. 2008b). According to recent data, this species is scattered all over Eurasia, and is known from southern North America and Hawaii (PRZYBOŚ *et a*l. 2008b), so it can be considered as cosmopolitan.

All these facts seem to support the conclusion of BASS *et al*. 2007: "geographic dispersal in macroorganisms and microbes is not fundamentally different: some taxa show restricted and/or patchy

distributions while others are clearly cosmopolitan". Still, patterns of distribution of microorganisms reported to date are literally "state-of-the-art", as they can be changed cardinally with any new findings in less sampled territories. Vast territories of Russia still remain totally unsampled (see Fig. 1) and very promising for further research.

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