New, World-wide Data on the Distribution of Species of the *Paramecium aurelia* Complex (Ciliophora, Protozoa)

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Accepted May 25, 2010

PRZYBOŚ E., SURMACZ M. 2010. New, world-wide data on the distribution of species of the *Paramecium aurelia* complex (Ciliophora, Protozoa). Folia biologica (Kraków), **58**: 185-188.

This is the first report on the presence of *P. biaurelia* in Tasmania, an island that has probably never been investigated before for the occurrence of the *P. aurelia* species. *P. tetraurelia* was recorded in Brazil, another very poorly investigated country in terms of this species complex. New stands of *P. biaurelia* and *P. tetraurelia* were also recorded in Japan. We present data concerning the occurrence and distribution of the *P. aurelia* species on different continents as a background for the newly described stands of *P. aurelia* spp.

Key words: *Paramecium aurelia* species complex, distribution of species on continents, different levels of sampling.

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The Paramecium aurelia complex is composed of 15 species known world-wide (SONNEBORN 1975; AUFDERHEIDE et al. 1983). Some are considered cosmopolitan such as P. primaurelia, P. biaurelia, P. tetraurelia, and P. sexaurelia, whereas others were recorded only in a few or single habitats (as P. tredecaurelia, P. quadecarelia, P. sonneborni). However, various parts of the world have not yet been studied or sampling was done only very occasionally. In this respect we collected data (SONNEBORN 1975; AUFDERHEIDE et al. 1983; Przyboś & Fokin 2000; Przyboś 2005; PRZYBOŚ et al. 2008a, and other published papers of the senior author, see Table 2c) concerning the occurrence and distribution of the P. aurelia species on different continents as a background for our present findings of new stands of the P. aurelia spp. in Japan, Brazil and Tasmania.

Central and South America, Australia, Africa, as well as several regions of Asia have been studied at a very inconsiderable level (Tables 2a, 2b). In North America only the USA was studied carefully (SONNEBORN 1975) where the majority of species of the complex were recorded (Table 2a). In Asia, more frequent sampling was performed only in Japan and Asiatic Russia, revealing the occurrence of several species (Table 2b). The largest dataset on the distribution and frequency of occurrence of species of the *P. aurelia* complex con-

cerns Europe (Table 2c) in which 531 habitats were studied (PRZYBOŚ *et al.* 2010a). Again, a different number of habitats was studied in particular zones of Europe, i.e. 102 in northern, 55 in southern, and 374 in the central zone, and mainly in Poland (218 habitats among 374) (PRZYBOŚ *et al.* 2010 b).

Material and Methods

Material

Strains identified at present (Table 1) were kindly sent by Prof. Masahiro FUJISHIMA, Yamaguchi University, Japan.

Methods

a. Methods used in strain cultivation and identification

The methods of SONNEBORN (1970) were used for the cultivation of strains, induction of conjugation and autogamy. Paramecia were cultivated on a lettuce medium inoculated with *Enterobacter aerogenes* in a temperature of 27°C. Species of *Paramecium aurelia* were identified by mating the

Table 1
New stands of species of the *Paramecium aurelia* complex

Species	Strain designation	Place of strain origin	Date of collection	Collector's name				
Paramecium tetraurelia	250 (UC 110)	Unknown Japan	Unknown	Toishiro Sugai, Ibaraki University, Japan				
Paramecium biaurelia	Pa 1 Pa 2 Pa3	Yamaguchi Prefecture, Yuu town, Japan	2009	Akiko Sano, Ehime University, Japan				
Paramecium tetraurelia	Cax1 Cax2	Caxambu, Brazil	2007	Masahiro Fujishima, Yamaguchi University, Japan				
Paramecium biaurelia	Tas1 Tas2 Tas3 Tas4	Tasmania Island, Australia	2008	Masahiro Fujishima, Yamaguchi University, Japan				

investigated clones with the mating types of standard strains of known species. The following standard strains were used:

strain 90, Pennsylvania, USA of *P. primaurelia*, strain Rieff, Scotland of *P. biaurelia*, strain 324, Florida, USA of *P. triaurelia*, strain from Sydney, Australia of *P. tetraurelia*, strain 87, Pennsylvania, USA of *P. pentaurelia*, strain 159, Puetrto Rico of *P. sexaurelia*, strain 38, Florida, USA of *P. septaurelia*, strain 138 Florida, USA of *P. octaurelia*, strain 223 Florida, USA of *P. decaurelia*, strain 246, Mississipi, USA of *P. dodecaurelia*.

b. Methods used in strain-crosses

In the strain crosses between the investigated strain and the standard one of the particular species of the P. aurelia complex, the F_1 generation was obtained by conjugation and F_2 by autogamy (using the method of daily isolation lines). The occurrence of the desired stage of autogamy (specimens at a stage of two macronuclear anlagen) was examined on preparations stained with aceto-carmine. The survival of clones in both generations was estimated in 100 clones. According to CHEN (1956), the clones could be categorized as surviving after passing through 6-7 fissions during 72 hours after separation of partners of conjugation or postautogamous caryonids.

Table 2a Occurrence of species of the *Paramecium aurelia* complex in the Americas, Africa and Australia

Continent/coutry	Species of the <i>Paramecium aurelia</i> complex designated by numbers														
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	P.s.*
Northern America	+	+	+	+	+	+	+	+	+	+	+	+			+
Central America	+			+		+		+					+		
Southern America	+	+		+											
Hawaii	+											+			
New Zealand		+													
Australia				+	+									+	
Africa						+		+						+	
Tasmania Island		+													
Tenerife												+			
Madagascar													+		

^{*}P.s. – Paramecium sonneborni.

Table 2b Occurrence of species of the *Paramecium aurelia* complex in Asia

Country		Species of the Paramecium aurelia complex designated by numbers													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	P.s.
Japan	+	+		+		+				+		+			
Vietnam	+														
Thailand						+									
China						+									
Asiatic Russia	+	+	+		+				+	+		+			
Kazakhstan												+			
Turkmenia	+														
Georgia		+													
India	+			+		+									
Turkey									+						
Lebanon		+													
Israel	+	+		+				+					+		

Table 2c Occurrence of species of the *Paramecium aurelia* complex in Europe

Country		Species of the <i>Paramecium aurelia</i> complex, designated by umbers														
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	P.s.	
Iceland	+															
Norway		+														
Sweden		+							+							
Finland		+		+					+							
Russia	+	+	+	+	+	+			+	+		+				
Great Britain	+	+		+					+							
Germany		+	+			+	+	+	+			+				
France	+	+		+					+				+			
Poland	+	+	+	+	+				+			+				
Ukraine	+	+	+						+			+				
Czech Republic		+	+	+					+							
Slovakia	+			+												
Holland				+												
Switzerland	+															
Italy	+	+		+	+							+				
Hungary	+	+			+				+							
Bulgaria	+	+			+				+							
Romania	+	+	+		+				+							
Greece	+						+									
Croatia							+									
Spain	+	+	+	+	+	+			+							

Data in Tables 2a,b,c – from Sonneborn 1975; Aufderheide $et\,al.$ 1983; Przyboś & Fokin 2000; Przyboś $et\,al.$ 2002a,b; Przyboś & Fokin 2003; Przyboś $et\,al.$ 2003b; Przyboś 2005; Przyboś $et\,al.$ 2007; Przyboś $et\,al.$ 2008a,b; Przyboś $et\,al.$ 2009a,b; Potekhin $et\,al.$ 2010.

Results and Discussion

The investigated strains originating from Japan (Yuu in Yamaguchi Prefecture) and Tasmania were identified as *P. biaurelia* based on strong conjugation with the standard strain of the species. Strains from Brazil (Caxambu) and strain of unknown origin (Japan?) were identified as *P. tetraurelia* thanks to strong conjugation with the standard strain of the species (Table 1).

All hybrids from the inter-strain crosses (investigated strain x standard strain of P. biaurelia or P. tetraurelia) showed a high percentage of surviving clones in generations F_1 and F_2 .

The obtained results revealed the first information on the presence of *P. biaurelia* in Tasmania, which has probably never been investigated for the occurrence of the *P. aurelia* species (Table 2a). Similarly, the record of *P. tetraurelia* in Brazil is important as this country has been very poorly investigated (Table 2a). In contrast, several sites in Japan have been sampled before and the presence of *P. primaurelia*, *P. biaurelia*, *P. tetraurelia*, *P. sexaurelia*, *P. decaurelia*, *P. dodecaurelia* (cf PRZYBOŚ & FOKIN 2001; PRZYBOŚ *et al.* 2003a) was recorded (Table 2b), however, new stands of *P. biaurelia* and *P. tetraurelia* confirm the wider distribution of these species in Japan.

Further sampling especially in the southern hemisphere may put forth new data on the occurrence of species of the *P. aurelia* complex. This was the case of *P. quadecaurelia* known for many years only from Emily Gap, Australia (SONNEBORN 1975) but later recorded in Vindhoek, Namibia (PRZYBOŚ *et al.* 2003b). It was identified not only by the mating reaction with the Australian strain according to SONNEBORN's method (1970) but also based on sequences for the cytosol type *hsp70* gene of both strains revealing 99.2% similarity. However, investigations in Africa are still very limited.

Acknowledgements

The authors are very grateful to Prof. Masahiro FUJISHIMA, Yamaguchi University, Japan for sending us the strains from Japan, Brazil and Tasmania.

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