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# First record of freshwater bryozoans (Bryozoa: Phylactolaemata) in the aquatic invertebrate fauna of Serbia

### Vesna M. Martinovic-Vitanovic\*, Vesna M. Milankov, Vladimir I. Kalafatic

Department of Freshwater Ecology and Water Protection, Institute for Biological Research "Sinisa Stankovic", University of Belgrade, 142 Blvd. Despota Stefana, 11 000 Belgrade, Serbia

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#### Introduction

Bryozoans are sessile invertebrates that grow as a colony of identical zooids attached to underwater objects. Within the Phylum Bryozoa, species of the class Phylactolaemata live exclusively in fresh water habitats. The phylactolaemates include five families (Bushnell 1965; Massard and Geimer 2008a). Phylactolaemate taxonomy has advanced considerably since the publication of the classic works in this field. According to Massard and Geimer (2008a) the latest revision of the systematics of Phylactolaemata is done by Vinogradov (2004) and still waiting for general acceptance.

They often occur on aquatic vegetation, stones, branches and other organic or artificial firm substrata. As suspension feeders they are an important part of biofiltering system in aquatic habitats. Phylactolaemates produce asexual bodies called statoblasts which when the conditions are favourable, can start a whole new colony generation. Morphologically statoblasts are grouped into three categories: sessoblasts, piptoblasts, and floatoblasts (Wöss 1996). Most species produce two types of dormant bodies, sessoblasts and floatoblasts. One species, *Plumatella casmiana* Oka, 1907 forms two distinct types of floatoblasts (Wöss 2002a). With their preferred habitat and growth form bryozoans present a major biofouling nuisance in municipal water

#### ABSTRACT

Species of freshwater bryozoans are newly recognized in the fauna of aquatic invertebrates in Serbia. This first record includes a total of five species of class Phylactolaemata found at eight localities during limnological investigations in the period from 2002 to 2006. Four species are assigned to family Plumatellidae (*Plumatella casmiana, P. emarginata, P. geimermassardi* and *Hyalinella punctata*). Family Cristatellidae is represented by one species (*Cristatella mucedo*). The freshwater bryozoans in the form of intact colonies and especially as dormant bodies were identified to the species level. The identification was done using scanning electron microscopy, analyzing resting bodies' morphological ultrastructure. © 2009 Elsevier GmbH. All rights reserved.

pipes and irrigation pipelines (Wood and Marsh 1999). Pennak (1953) and Wood (2001) give details on bryozoan morphology, anatomy and ecology.

Until now, 19 species of freshwater bryozoans have been recorded in Europe (Massard and Geimer 2005, 2008a).

Freshwater bryozoans have never been previously a target of scientific investigations in Serbia (or in republics of former Yugoslavia). Only one report about this group has been published, dealing with the occurrence of Plumatella repens (L.) and Cristatella mucedo Cuv. in the Skadar Lake in Montenegro (former Yugoslavia's republic) and Albania (Pesic and Dhora 2001). The most probable reason is that these small sessile filter feeders are seldom the main focus of standard limnological investigations. Furthermore, colonies could be very easily misidentified as a mat of moss because of their plant like appearance. In addition, colonies cannot be found through entire year, because of the dormant stage in the life history of freshwater bryozoans. Moreover, at the species level, freshwater bryozoan species are very difficult to identify because of their high morphological variability. The plasticity of morphological features in colonies makes these characters inconsistent distinguishing feature. Analysis of ultrastructure on the surface of resting bodies using scanning electron microscopy (SEM) can provide useful data for positive species identification (Reynolds 2000).

#### Sites and methods

Geographical positions of study sites are given in Fig. 1.



<sup>\*</sup> Corresponding author. Tel.: +381112078369; fax: +381112761433. *E-mail addresses*: vmartino@ibiss.bg.ac.rs, vesna.m\_martinovic@vektor.net

<sup>(</sup>V.M. Martinovic-Vitanovic).

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Fig. 1. Map of the investigated localities in Serbian waters.

Geographical details of locations studied with sampling dates and denotations of site names corresponding with site numbers in Fig. 1, with descriptions of collecting sites are listed below:

- Site Canal/Pond Petra near village Perlez, 24.2 km S from Zrenjanin city as part of 1 the Danube–Tisa–Danube canal network in Central Banat District-Vojvodina Province (N 45°11′41.5″, E 020°23′23.6″). Pond Petra is 18 km long and 10–20 m wide, permanent water body situated on alluvial plane 74 m above sea level in the old abandoned bed of River Tisa. Rich macrophytic submerged and emergent vegetation overgrow the bottom and the banks of the pond at sampling site. Substrate was sand and silt. On May 5, 2003 periphyton samples were taken from submersed and emergent macrophytes and hygrophytes (*Typha latifolia, Polygonum hydropiper, Menta aquatica, Alisma plantago aquatica*) and other underwater substrates such as wood, branches, etc. Water was transparent to the bottom. Open water trophic status of canal/ pond Petra based on qualitative/quantitative analyses of plankton and macrophytic communities is described as oligo-mesotrophic.
- Site Settling basin as part of the Sava Lake reservoir, damned backwater of the 2 Sava River between its right bank and a sandy island Ada Ciganlija (from 9 to 4.5 r-km) is situated in the inner of the Belgrade city area (N 44°46'32.8", E 020°22′09.7″). Laying upstream the Lake, settling basin is connected with the River Sava and the Sava Lake through pumping stations. Settling basin is 0.85 km in length and 230 m in width (maximum width 260 m), covering an area of 19.7 ha. It is a shallow part of the Sava Lake reservoir with a mean depth of about 6.5 m (maximum around 10 m) and situated 78 m a.s.l. Macrophytic submerged (and floating and emergent) vegetation overgrow the banks and the bottom of the basin: Najas marina, N. minor, Potamogeton spp., Ceratophyllum demersum, Elodea canadensis, T. latifolia, etc. Substrate was sand and silt. On November 23, 2006 benthos sample was taken by Van Veen grab. At the sampling spot Secchi disk water transparency was 1.5 m. Settling basin's trophic status in the autumn 2006 was in the mesotrophic zone. Generally, water quality of the Sava Lake reservoir as well as Settling basin's as it's part is under the influence of the Sava River (Martinovic-Vitanovic and Kalafatic 1990; Martinovic-Vitanovic et al. 2008).
- Site Ada Safari Lake is situated on the island Ada Ciganlija in the Sava River 4 km 3 from the Belgrade city centre (N 44°47′33.3″, E 020°24′58.5″). It is a small, irregularly shaped lake on the northern tip of the island Ada Ciganlija 0.4 km long and 120 m wide. Elevation is 73 m a.s.l. It was the last remaining marshy area in that section of the island, overgrown by submerged macrophytes and reeds. This area was turned into a small (covering 6 ha) sports-fishing lake in 1994. On July 16, 2003 periphyton samples were taken from macrophytes and other underwater substrates such as wood, branches, etc. Trophic state is described as meso- to eutrophic.
- Site Navigation canal "Marina" is in Danube permanently flooded area on its left 4 bank (1082.5–1078 r-km) as a part of Mali Rit Marsh (1084–1079 r-km) at Banatska Palanka village 11 km SW from Bela Crkva city (N 44°48'42.7", E 021°16'45.7"). Navigation canal is 4.5 km long and 100 m wide situated 65 m above sea level. Canal is totally overgrown with submerged and floating macrophytes: Potamogeton spp., C. demersum, Myriophyllum spicatum, Trapa natans (as dominant species), N. marina, Salvinia natans, etc. There is a reed belt surrounding water body (Stevanovic et al. 2003). Sampling site is in the middle of the longitudinal profile in front of Hunting House at Hunting Ground "Dragica Hat" (1081.5 r-km). Substrate is silt with coarse detritus. On July 9, 2005, benthos sample was taken by Van Veen grab. Trophic state based on macrophytic vegetation is described as eutrophic.

- Site Labudovo okno (Swan Lake) belonging to the Mali Rit March on the exit of
- 5 the canal "Marina" (1078 r-km) is situated in Dolnice bay as a part of Danube permanently flooded area at Banatska Palanka village 11 km SW from Bela Crkva city (N 44°48'45.7", E 021°16'55.4"). Elevation is 67.7 m above sea level. Labudovo okno (Swan Lake) is a shallow flooded area mosaically overgrown with submerged and floating macrophytic vegetation: Potamogeton spp., S. natans, Lemna spp., with T. natans as dominant species, etc. Among emergent plants the most significant are Phragmithes australis, Typha angustifolia and T. latifolia (Kalafatic and Martinovic-Vitanovic 1999; Stevanovic et al. 2003). Substrate is silt with coarse detritus. On July 12, 2002 periphyton samples were taken from macrophytes using Kick-Net. On September 30, 2006 benthos sample was taken by Van Veen grab. Substrate was greyish-yellow-black very fine silt and detritus. Based on trophic indicators zooplankton and macrophytic vegetation analyses Labudovo okno (Swan Lake) trophic status is described as mesotrophic and eutrophic, respectively (Kalafatic and Martinovic-Vitanovic 1999).
- Site "Canal DTD" on its mouth to Danube (1076.2 r-km at Banatska Palanka
  village 11 km SW from Bela Crkva city) is a part of Danube–Tisa–Danube canal network (N 44°49′41.1″, E 021°19′54.9″). With steeped banks and about 70 m width canal is situated 65.5 m above sea level. Substrate is greyish-black mud with very fine sand particles. Canal is highly eutrophic overgrown with submersed and emergent macrophytes with floating species: *Potamogeton* spp., *C. demersum, C. submersum, M. spicatum, T. natans*, *S. natans, Lemna* spp., *P. australis, T. latifolia, T. angustifolia* and *Scirpus lacustris*. On July 12, 2002 benthos sample was taken by Van Veen grab. Saprobic analyses of plankton and benthic communities indicate II–III class of water quality, i.e. beta-meso- to alfa-mesosaprobity.
- Site Ada Cibuklija is a flooded islet in the Danube course at Banatska Palanka
  7 village 11 km SW from Bela Crkva city. This marshy area has elongated shape (about 4 km in length) and collecting site is on the downstream tip of the flooded islet (1083–1079 r-km) situated 65 m above sea level (N 44°48′14.0″, E 021°18′57.3″). Ada Cibuklija marsh is highly eutrophic overgrown with submersed and emergent macrophytes and floating species: *Potamogeton* spp., C. *demersum, C. submersum, M. spicatum, T. natans, S. natans, Azolla filicauloides, Lemna* spp., *P. australis, T. latifolia, T. angustifolia* and *S. lacustris*. On July 24, 2006 and on August 20, 2006 periphyton samples were taken from the stands of *Potamogeton* spp.
- Site Danube at Donji Milanovac city in Djerdap I reservoir (991 r-km) (N
  8 44°28'12.4", E 022°08'39.6") is situated 64 m above sea level.
  On April 8, 2005 from the middle of the transversal profile (depth 21 m) benthos sample was taken by Van Veen grab. Substrate was of sand and gravel. Based on qualitative and quantitative plankton analysis and the concentration of Chlorophyll *a* (in April maximal annual concentration was 5.62 mg m<sup>-3</sup> oligotrophy (OECD 1982); mean annual concentration 4.0 mg m<sup>-3</sup> mesotrophy (OECD 1982; Felföldy 1974)) the trophic status of Djerdap I reservoir at Donji Milanovac is classified as oligotrophy.

Investigated localities belong to different types of water. They included artificial stagnant water bodies: Site 3 is a sports-fishing lake situated on the island Ada Ciganlija in the Sava River, Site 2 is a settling basin of the Sava Lake an artificial water body (reservoir), and Site 1-canal/pond Petra considered as part of an old water bed of the River Tisa, included in the flow-through Danube-Tisa-Danube canal network. This network also includes Site 6. Sites 4, 5 and 7, belong to wetland "Labudovo okno" - one of the Ramsar's sites in Serbia as part of the permanently flooded area of the Danube River, section from Zilovo islet (1091 r-km) to River Nera mouth into Danube (1075.4 r-km). Site 4 is navigation canal in the part of Mali Rit Marsh (1084-1078 km). This marsh also include Site 5. a shallow flooded lake Labudovo okno (Swan Lake) situated in the bay Dolnice. Site 7 is a flooded islet-marsh in the Danube's main course, now located in a permanently flooded zone of the Danube (flow-through reservoir) resulting from dam constructed in the 1960s. Site 8 is in Djerdap I reservoir (99 r-km).

Material presented in this paper was collected by two different sampling procedures.

The first group of samples involves sampling of intact colonies. The colonies were taken in 2003 and 2006 at Sites 1, 3, and 7. Sampling was carried out in water less than 1 m deep, using scissors to cut the natural substrate for bryozoan's colonies (submerged branches and stems, leaves and roots of aquatic plants). Colonies were narcotized with saturated aqueous solution

#### Table 1

Values of environmental variables measured at Sites 1-8.

Localities/year	t <sub>H₂O</sub> (°C)	pН	
Conductivity (µS/cm)			
1. Canal/Pond Petra-Danube–Tisa–Danube canal network in Vojvodina province near village Perlez, May 2003	20.5	7.4	1,240
2. Settlement basin upstream the Sava Lake reservoir, November 2006	10.6	8.30	400
3. Ada Safari Lake, sports-fishing lake at Ada Ciganlija island in the Sava River, Belgrade Region, July 2003	24.7	7.9	-
4. Navigation canal "Marina" (1082.5–1078 r-km) in Mali Rit March-Danube, permanently flooded area 1081.5 r-km in	25.0	8.3	430
front of Hunting House at Hunting Ground "Dragica Hat" near Banatska Palanka, July 2005			
5. Labudovo okno (Swan Lake) as a part of Mali Rit March on the exit of the canal "Marina" (1082.5–1078 r-km),			
Dolnice–Danube permanently flooded area at Banatska Palanka			
July 2002	24.8	7.9	378
September 2006	20.5	8.45	324
6. Canal Danube–Tisa–Danube at Banatska Palanka, mouth – 1076.2 Danube km, July 2002	25.0	8.1	422
7. Ada Cibuklija – an flooded islet in the Danube, 1083–1079 r-km,			
July 2006	28.8	8.22	380
August 2006	22.8	7.99	370
8. Danube at Donji Milanovac town, Djerdap I reservoir, 991 Danube km, April 2005	11.0	8.05	412

of chloral hydrate [Cl<sub>3</sub> CCH (OH)<sub>2</sub>] and fixed in 10% aqueous solution of 35% formaldehyde. Small pieces of colonies were taken by forceps and statoblasts were removed from the zooids using fine entomological needles.

The second group of samples involves material from benthos samples collected for the purpose of standard limnological investigations in the Serbian part of the Danube. Benthos samples containing bryozoan statoblasts were taken in 2002, 2003, 2005 and 2006. Additionally, statoblasts were also found in benthos samples from the Sava Lake in 2006. Sampling was carried out with Van Veen grab ( $270 \text{ cm}^2$ ). One of the periphyton samples was taken by Kick-Net. Animals were washed through sieve (mesh size  $200 \,\mu\text{m}$ ), fixed and stored in 4% aqueous solution of formaldehyde.

Data from each site included: GPS positioning and altitude, followed by a more detailed description of the site location. Water temperature, pH and conductivity are given in Table 1.

Species identification of collected material was done using statoblasts. Scanning electron microscopy was conducted at the Department of Theoretical Biology, Morphology Section at University of Vienna. Floatoblasts were rinsed with distilled water several times than cleaned with aqueous solution of bleach (1:1) in order to remove as much dirt and microorganisms, as possible. Samples were then chemically dehydrated in series of dehydrating agents: DMP (2,2-dimethoxypropane), water-free acetone and hexamethyldisalazone (HMDS), respectively, and than air dried on the filter paper under the fume hood. After drying, floatoblasts were put on aluminium stabs, mounted with TEMPFIX-thermo glue, gold sputtered coated examined at 10–15 keV in a Philips 20 scanning electron microscope and photographed digitally.

Species identification was done using following keys: Mundy (1980), Geimer and Massard (1986), and Wood and Okamura (2005).

#### Results

A total of five species of freshwater bryozoans were sampled and identified on 10 occasions in the period from 2002 to 2006, at eight investigated localities given in Table 1 with the values of measured environmental variables.

Physical-chemical factors measured *in situ* in the water at sampling sites were in the range:

Water temperature	10.6–28.8 °C
Water reaction (pH)	7.40-8.45
Conductivity	324-1240 µS/cm at 20 °C

Four identified species are assigned to family Plumatellidae and one to Cristatellidae.

Family Plumatellidae Plumatella casmiana Oka, 1907 Plumatella emarginata Allman, 1844 Plumatella geimermassardi Wood and Okamura, 2004 Hyalinella punctata Hancock, 1850 Family Cristatellidae Cristatella mucedo Cuvier, 1798

#### **Family Plumatellidae**

1. Plumatella casmiana Oka, 1907

Sites 1 and 3, colonies-periphyton sample taken from macrophytes and other underwater substrates (wood, branches, etc.); *Plumatella casmiana* Oka, 1907 was recorded examining intact colonies samples. The colony was compact; with short richly branched zooids adhere to the substratum throughout their length. The colony wall was semi-transparent. The lophophore is bearing fewer than 30 tentacles. Two types of free statoblasts were detected – "capsuled" floatoblast, as typical in all plumatellids, and thin walled leptoblast. The presence of the thin walled floatoblast is the unique characteristic for the *P. casmiana*. Leptoblasts (Fig. 2) have a relatively narrow annulus and weak tuberculation. Its valves are not fused together and it germinates immediately when released from colony.

In Fauna Aquatica Austriaca (Wöss 2002b) denote *P. casmiana* as rare in Austria and as a species without saprobiological indication, i.e. without saprobic valence.

2. Plumatella emarginata Allman, 1844

Site 6, floatoblasts only;

Site 7, periphyton sample taken from *Potamogeton* spp. Very dense colonies of *P. emarginata* were found on stems of *Potamogeton perfoliatus* and *P. fluitans* signed as *Potamogetonetum pectinati* prov. (Stevanovic et al. 2003).

*Plumatella emarginata* Allman, 1844 was identified from free statoblasts and colony fragments. The colony had very dense and compact dark brown zooids with pale neck. The number of tentacles on the lophophore was between 38 and 42. Floatoblasts of *P. emarginata* are distinguished by small flat dorsal fenestra with smooth surface (Fig. 3). The ventral valve is slightly larger than dorsal, strongly convex and with very distinct tubercles on the fenestra (Fig. 4).

*P. emarginata* was denoted in Fauna Aquatica Austriaca by Wöss (2002b) as saprobiological indicator with individual saprobic value SI = 2.3, and G-indicator weight value 2. In Joint Danube Survey 2001 Techical Report (JDS 2002) saprobic value s = 2.0.



Fig. 2. Scanning electron micrograph showing leptoblast of Plumatella casmiana.



Fig. 3. Scanning electron micrograph showing dorsal valve of floatoblast of Plumatella emarginata.

*P. emarginata* is known as species tolerant to very nutrient rich (or even polluted) water (Wöss 2005).

- 3. Plumatella geimermassardi Wood and Okamura, 2004
- Site 7, colonies-periphyton sample taken from Potamogeton spp.;

*Plumatella geimermassardi* Wood and Okamura, 2004 has been identified by floatoblasts which have relatively large dorsal fenestra and narrow annulus (Fig. 5). The fenestra is bearing

low, weakly defined tubercles that become less distinct toward the centre.

4. *Hyalinella punctata* Hancock, 1850 Site 8, floatoblasts only

*Hyalinella punctata* Hancock, 1850 have been identified by observing highly defined (determined) tubercles on the surface of floatoblasts. The entire surface of the dorsal valve is covered, both on fenestra and annulus, with evenly distributed and very



Fig. 4. Scanning electron micrograph showing ventral valve of floatoblast of Plumatella emarginata.



Fig. 5. Scanning electron micrograph showing dorsal valve of floatoblast of Plumatella geimermassardi.

distinct tubercles (Fig. 6). The same distribution pattern as well as the same appearance of tubercles is observed on the ventral valve's surface (Fig. 7). A little difference is noticed between the size of fenestra on the dorsal and ventral valve of the floatoblasts. *H. punctata* was denoted in Fauna Aquatica Austriaca by Wöss (2002b) as saprobiological indicator with individual saprobic value SI = 2.2, and G-indicator weight value 3.

#### Family Cristatellidae

5. *Cristatella mucedo* Cuvier, 1798 Sites 2, 4, and 5, floatoblasts only;

*Cristatella mucedo* Cuvier, 1798 was identified from floatoblasts, which are easily recognized by its discoid shape and spines with terminal hooks radiating from the margins of the dorsal fenestra (Fig. 8).



Fig. 6. Scanning electron micrograph showing dorsal valve of floatoblast of Hyalinella punctata.



Fig. 7. Scanning electron micrograph showing ventral valve of floatoblast of Hyalinella punctata.

*C. mucedo* indicate beta-mesosaprobic conditions in the water being saprobiological indicator with individual saprobic value SI = 1.9 and G-indicator weight value 3 (in Fauna Aquatica Austriaca, Wöss 2002b).

This species is present in a great number of smaller water bodies in riverine forests and floodplain areas, with higher abundance in nutrient poor sites, such as gravel ponds (Wöss 2005).

#### Discussion

Although Serbia has a relatively long tradition of limnological investigations, bryozoans were never their main focus. The species presented in this paper are found for the first time in the territory of Serbia. The findings of total of five species are the result of discontinued investigations in the period from 2002 to 2006 in Serbian natural and artificial water bodies.



Fig. 8. Scanning electron micrograph showing dorsal valve of floatoblast of Cristatella mucedo.

There are published data about the presence of freshwater bryozoans in other European countries (Geimer and Massard 1986; Massard and Geimer 2005, 2008a; Økland and Økland 2000, 2003, 2005; Pesic and Dhora 2001; Taticchi et al. 2005; Wood and Okamura, 2004; Wöss 1991, 2002a, 2005; Wöss and Walzl 2006). From the total of 19 evidenced species in Europe, the smallest number of bryozoans was in Slovenia (seven species), while the highest diversity of freshwater bryozoan's species was in Germany, with the number of 17 species (Massard and Geimer 2005).

Comparing the results of bryozoan's distribution in Austrian Danube (Wöss 2005) with the species recorded in Serbian sector of the River Danube, we have noticed certain similarity.

In our previous long-term bottom fauna investigations the Danube in Serbia (river kilometre 1433-845.6) a large lowland river was divided into - I: Upper Part - Riverine zone; II: Middle Part – Transitional zone-flow through reservoir with permanently flooded areas from 1091 km Ada Zilovo down to 1076.2 km Banatska Palanka-Ram; and III: Lower Part - Lacustrine zone (lake part-reservoirs: Djerdap I and II (Martinovic-Vitanovic et al. 2006)). From four species recorded in the Serbian part of Danube, three of them (P. emarginata, H. punctata and C. mucedo) were found in two free flowing stretches of the Austrian Danube sector upstream and downstream from Vienna. The Austrian part of the Danube is alpine in character (Fesl et al. 2005). Nine of the ten species of freshwater bryozoans recorded in Austria (Wöss 2002b) belong to the class of Phylactolaemates (and one to the class Gymnolaemates - Paludicella articulata) were found in the River Danube's main course, in its floodplain areas, backwaters and tributaries (rivers and streams) as well as in the other standing natural (lakes and ponds) and artificial water bodies (Wöss 2005). P. geimermassardi was not recorded in the Austrian Danube. So far P. geimermassardi is reported from England, Ireland, Belgium, southern Norway, northern Germany and Italy (Wood and Okamura 2004). P. casmiana is one of the less common species of the family Plumatellidae in Austria, being exceptionally very abundant in backwater 'Pond at Ringelau', i.e. in the floodplain area on the left bank of the Danube near Linz (Wöss and Walzl 2006).

Five other bryozoan species from nine freshwater bryozoan's taxa (the class of Phylactolaemates) recorded in Austria belong to

three genera: *Fredericella* (*F. sultana*), *Plumatella* (apart already mentioned *P. emarginata* — *P. fungosa*, *P. repens* and *P. fruticosa*), and *Lophopus* (*L. crystallinus*), were observed in the Danube's main channel among them *Fredericella sultana* in the form of colonies and statoblasts being most abundant (Fesl et al. 2005). According to Wood (1973) *F. sultana* shows greater tolerance to low temperatures (average temperature in investigated Austrian Danube stretch was  $10 \,^{\circ}\text{C}$  – Fesl et al. 2005) than does any other freshwater Ectoprocta, and it has a worldwide distribution being a frequent member of the benthos of several Swiss and Italian alpine lakes.

The reason why these (and other) species have not been recorded in Serbia is that many regions in Serbia have never been surveyed for freshwater bryozoans although an actual occurrence in the Danube main stream and floodplain areas seems very probable. For this reason we assume that the list is not yet complete.

According to hydrological and ecological features Serbian sector of the Danube River has a permanently flooded area which belongs to transitional zone of the middle part of Danube River in Serbia (Martinovic-Vitanovic et al. 2006). The Danube's permanently flooded area is showing water level fluctuations during each year due to the seasonal changes in the temperate climate and as a result of hydropower plant operations. The Danube's course in Serbia, especially parts II and III, are within the zone of the modes of operation of the Djerdap I and Djerdap II hydropower plants, with forces oscillations of the water level, changes in the course velocity, etc. (Martinovic-Vitanovic et al. 2006). Nevertheless, the bryozoans species were detected in this fluctuating environment. Localities where intact colonies or resting stages were collected are characterized by rich association of submerged and floating macrophytes: P. perfoliatus L., P. pectinatus L., P. crispus L., P. fluitans Roth, T. natans L., C. demersum L. and S. natans L., etc. Very dense colonies of P. emarginata were found on stems of P. perfoliatus and P. fluitans. Statoblasts of P. geimermassardi and C. mucedo were evidenced from benthos samples (Martinovic-Vitanovic et al. 2006) from this area.

Sampling sites near the town of Donji Milanovac (in the middle of the Danube's sampling transversal profile) differ from other localities (Martinovic-Vitanovic et al. 2006). Only at this locality were the statoblasts of *H. punctata* found. The benthos material where the statoblasts were detected was sampled from the depth of 21 m with substrate of gravel and sand.

Colonies of *P. casmiana* and dormant stages of *C. mucedo* were recorded in investigated artificial standing water bodies (ponds in permanently flooded areas and reservoirs) and in slowly flowing waters (canals).

Physical-chemical factors varied considerably throughout the year. Measured water temperature ranged 10.6 °C (November-April) and 28.8 °C (May, July, August and September). In general, colonies of bryozoans limit their occurrence to the summer months. During the winter, under unfavourable conditions, only resting stages can survive in the water bodies (Wöss 1991, 2002a, 2005). Concentration of H<sup>+</sup> (pH = 7.40–8.45) indicates neutral to weekly alkaline water reaction at investigated localities with electrolytic content varying from low to moderate and high values (conductivity from 324 to 1240  $\mu$ S/cm at 20 °C). All recorded Bryozoa species were found at low elevations up to 200 m a.s.l., mostly about 100 m.

The study of Bryozoa in the Serbian stretch of the Danube continued in 2007 and according to our preliminary results the species of the freshwater bryozoans were found in the benthological samples at the following seven sites (in 10 probes) from June to November 2006 and 2007: in Riverine zone: Ledinci near Novi Sad (right bank) r-km 1258–1260; in Transitional zone-flow through reservoir with permanently flooded areas: Brestovik (right bank) r-km 1127; Smederevo (left bank) r-km 1116.2; Ram (right bank) r-km 1077–1076.2; and in Lacustrine zone (lake part-reservoirs: Djerdap I and II): Veliko Gradiste (right bank) r-km 1059; Donji Milanovac (right bank) r-km 991; and Kladovo (in the middle of the Danube's sampling transversal profile and at right and left bank) r-km 933.8 (author's unpublished data). Further investigations will show which bryozoan's species are present at mentioned sites.

Habitat characteristics at these localities are typical for the bryozoan's settlement due to the great amount of natural submerged substrate that can be colonized including macrophytic vegetation (Wood 1973; Stevanovic et al. 2003).

In investigated stretch of the Danube (1091–1075.4 r-km) the most dominants types of vegetation are communities of rootless (*Salvinio-spirodeletum polyrhizae* and *Lemno minoris-Azolletum filiculoides*) and rooted floating plants (*Ceratophyllo-Trapetum natantis* or pure *T. natans* stands), as well as submerged communities of pondweed and hornworth (*Myriophyllo-Potametum* and mixed or pure stands of *Potamogeton* species). Macrophytic vegetation was characterized through the seven common and four provisionally described macrophytic communities (Stevanovic et al. 2003).

Autochthonous, highly productive macrophyte vegetation action coupled with the allochthonous, potentially eutrophicating (and even polluting) influences on investigated water bodies are considered to be two major factors conditioning the trophic status of an examined ecosystem. Potentially high eutrophicating effects on investigated open water of different types of aquatic ecosystems resulting from acting anthropogenic factor are suppressed by the activities of eutrophic macrophyte vegetation of immediate surroundings as well as of the vegetation of flooded region. This is considered to be due to the huge more efficient uptake of nutrients by macrophytes concurrent to that of phytoplankton as well as to the slow degradation of plant remains resulting in slowing the nutrient circulation in ecosystem (Martinovic-Vitanovic 1996). These interactions are believed to be of prime importance in defining the trophic status of open water estimated upon relevant data on phyto- and zooplankton, and other aquatic components to be of oligo-mesotrophic and mesotrophic character.

According to Wöss (1991), generally the occurrence and abundance of the bryozoans in the riverine forests of the Austrian Danube change from nutrient poor to nutrient rich (back)waters indicating a progression from *C. mucedo, Plumatella fruticosa, Fredericella sultana, H. punctata, Plumatella emarginata, P. repens* to *P. fungosa.* 

Bryozoans species found in Austria are saprobiological indicators mainly indicating beta-meso-saprobity (Wöss 2002b; JDS 2002). *P. casmiana* as rare in Austria, and *P. geimermassardi* (not identified in Austrian fauna; Fauna Aquatica Austriaca, Wöss 2002b) are species whose saprobiological indications, i.e. saprobic valences are unknown.

A large number of waterfowl observed on the Danube in the last 25 years (Ham 1989; Puzovic et al. 1999; Simic 2000; Puzovic 2007) may be related to the appearance and distribution of bryozoans in the investigated stretch of the river Danube and in Djerdap I reservoir since statoblasts can be carried overland by waterfowl (Wöss 2005; Massard and Geimer 2007). According to the International Waterfowl Census on Yugoslav Danube more than 300,000 waterfowl come annually from northern Europe to overwinter on the Danube at towns of Ram (our sites – 4–7 are situated in the area of "*Labudovo okno*" – one of the Ramsar's sites in Serbia), Golubac, Donji Milanovac (in our investigations Site 8), and other locations downstream (Djerdap II and riverine part) up to Bulgarian border.

Based on species lists from elsewhere in the Austrian, Slovakian, and Hungarian Danube region, Austria as well as those from the other European countries, we can expect a more extensive Serbian bryozoans. Our most recent findings of Bryozoa (author's unpublished data from the years 2006, 2007, and 2008 too) and the data from literature (Massard and Geimer 2005, 2008a, b; Wöss 2005; Wöss and Walzl 2006) suggest that our list is not yet definitive.

Future studies will include investigations of the species richness and distributions as well as investigations of ecological requirements of freshwater bryozoans in Serbia in different types of water bodies. We expect to use different sampling methods including plankton and benthos along with periphyton (collecting "aufwuchs" material) sampling. The results described in this paper extend known geographic distribution of bryozoans in Europe.

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