Botanica SERBICA 41 (1): (2017) 65-70

Short Communication

New data on distribution and ecology of *Batrachospermum* (Rhodophyta) in Serbia

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ABSTRACT: The paper describes the distribution and ecology of populations of the genus *Batrachospermum* collected in Serbia from 2006 to 2015 at 10 sites, all of which represent new localities of *Batrachospermum* in Serbia. The records of this genus in the Boračka River and in a spring near Kragujevac (Cerovac) are the first in Central Serbia. *Batrachospermum* was recorded at altitudes of from 235 to 1600 m at localities mainly in partial or full shade on stone, gravel or concrete substrata in cold water (10.50 - 15.40°C) that was neutral to weakly alkaline (pH 7.2 – 8.65) with conductivity of from 55 to 539 μS/cm. Species of the genus *Batrachospermum* in Serbia have to date been recorded in clean, well-aerated waters with a low concentration of biogenic salts and in habitats without anthropogenic impact. Any factors altering abiotic parameters of their habitats can lead to changes and the loss of populations of the sensitive species of this genus.

Keywords: distribution, ecology, *Batrachospermum*, Rhodophyta, Serbia

Received: 01 July 2016 Revision accepted: 16 January 2017

UDC: 582.273:581.5(497.11)
DOI: 10.5281/zenodo.454422

INTRODUCTION

The distribution of red algae in Serbia has been confined to relatively few localities so far, the list of genera recorded to date including *Bangia*, *Batrachospermum*, *Chantransia*, *Hildenbrandia*, *Lemanaea*, *Paralemanea* and *Thorea* (Simić & Ranković 1998; Blaženčić *et al.* 2002; Cvijan 2002; Branković *et al.* 2007; Simić 2007; Simić 2008; Andrejić *et al.* 2010; Simić & Pantović 2010; Simić *et al.* 2010; Simić *et al.* 2014; Simić & Đorđević 2017; Simić *et al.* 2016). Thalli of freshwater algae of the genus *Batrachospermum* (Batrachospermaceae, Batrachospermales) have been found in springs, streams and rivers of Serbia. The following species were recorded and described on the basis of morphological and reproductive characteristics from 1908 to 2002: *Batrachospermum gelatinosum* (Linnaeus) De Candolle; *B. cayannense* Montagne ex Kützing; *B. confusum* (Bory) Hassall; *B. ectocarpum* Sirodot; *B. virgato-decaisneanum* Sirodot; *B. turfosum* Bory; and *Batrachospermum* sp. (Cvijan 2002; Cvijan *et al.* 2003). After that, only individual populations of *B. gelatinosum* and *Batrachospermum* sp. were recorded (Simić 2007; Simić *et al.* 2010; Simić *et al.* 2016).

Modern studies have shown that determination of the species and genus of specimens based on conventional characters is not sufficient, the combination of morphological and molecular analysis being necessary instead. Recently, the new genus *Sheathia* was separated from *Batrachospermum* by Salomaki *et al.* (2014), and many European species have been transferred to the new genus. Salomaki *et al.* (2014) recognised seven species from among the molecular clades. As already indicated above, the combination of morphology and molecular analysis is essential for a more precise determination of the species of individual specimens.

Data on the geographical distribution of species of the genus *Batrachospermum* in Serbia show that from 1908 (Cvijan 2002; Cvijan *et al.* 2003) to 2016 (Simić *et al.* 2010; Simić *et al.* 2016) they were mainly found in the eastern, southern and western parts of the country. In the northern part (Vojvodina), these species were detected only at one locality (Simić *et al.* 2010).

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The habitats of these algae are quite fragile (some of them have even been destroyed since the algae were first collected), so each and every new recording of these species in Serbia is unquestionably significant. All freshwater red algal species, including species of the family Batrachospermaceae, recorded in Serbia are on the list of strictly protected species of plants and animals (SGRS 5/2010).

This paper presents new data on algae of the genus *Batrachospermum*, including their newly recorded localities, the ecological conditions under which they were found there and factors potentially endangering their habitats.

MATERIALS AND METHODS

Thalli of *Batrachospermum* were collected at 10 sites from 2006 to 2015 (Fig. 1).

Site 1. The source of the Crni Timok River (karst spring from which the river flows, broad-leaved surrounding vegetation, stone and gravel substratum, 43°21'000" N 22°14'000" E, elevation 410 m, no anthropogenic impact). **Site 2.** The Grza River 100 m from the source (calcareous spring from which the river flows, broad-leaved vegetation, stone and gravel substratum, 43°53'905" N 21°38'863" E, elevation 432 m, no anthropogenic impact). **Site 3.** The Grza River Reservoir (reservoir, part of the Grza River dammed by aconcrete water gate, 43°53'905" N 21°38'863", elevation 312 m, no anthropogenic impact). **Site 4.** Upper reaches of the Vapa River (river, broad-leaved vegetation, stone and gravel substratum, 43°14'195" N 20°05'751" E, elevation 1030 m, no anthropogenic impact). **Site 5.** Pazarište on the Raška River (river, broad-leaved vegetation, stone substratum, 43°07'879" N 20°24'867" E, elevation 537 m, no anthropogenic impact). **Site 6.** The Boračka River (river, part of it below the dam, broad-leaved vegetation, stone substratum, 43°57'513" N 20°37'190" E, elevation 350 m, no anthropogenic impact). **Site 7.** Upper reaches of the Samokovska River (river, coniferous vegetation, stone and gravel substratum, 43°16'164" N 20°47'063" E, elevation 1600 m, no anthropogenic impact). **Site 8.** Upper reaches of the Barska River (river, coniferous vegetation, stone and gravel substratum, 43°17'136" N 20°43'406" E, elevation 1500 m, no anthropogenic impact). **Site 9**. The source of the Sokobanjska Moravica River (spring, broad-leaved vegetation, gravel substratum, 43°39'350" N 21°46'076" E, elevation 300 m, no anthropogenic impact). **Site 10.** The Cerovac Spring, Kragujevac (capped spring, broad-leaved vegetation, concrete substratum, 44°09'422" N 20°85'007" E, elevation 284 m; water level change). Some of the localities are in parts of rivers under a certain degree of protection (Tables 1, 2).

Specimens were collected in spring, summer and autumn. Algal samples were collected off different types of substratum (calcareous rocks, stones, concrete and tree branches submerged in shallow water). The samples

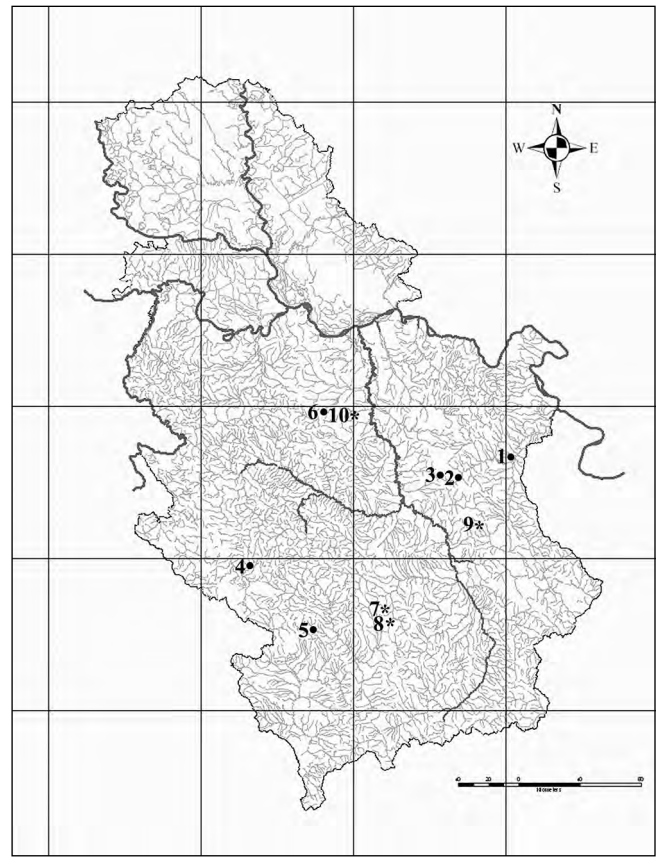

Fig. 1. Distribution of *Batrachospermum gelatinosum* (•) (Sites 1–6) and *Batrachospermum* sp. (*) (Sites 7–10) in rivers of Serbia.

were procured by standard methods (for example, by scraping with a knife and tweezers) and instantly fixed in 4% formaldehyde.

Altitude, latitude, longitude, surrounding vegetation, substratum type**,** depth of water (cm), and degree of shade (+++ full sun, ++ partial sun, + full shade) were determined at each locality. Physical and chemical parameters were analysed according to standard methods (APHA 1995). In determining physical and chemical characteristics of the water at each locality, the following data were recorded: temperature (°C), pH, oxygen concentration (mg/l), saturation (%), conductivity (μs/cm) and hardness (mg/l). Chemical parameters, i.e., phosphate, nitrate and ammonia concentrations (mg/l) were measured with portable instruments (Hanna Instruments). Any possible anthropogenic impact was recorded at each locality.

The analysis of algal material was performed in the Laboratory of Hydrobiology at the Department of Biology and Ecology of the Faculty of Science in Kragujevac. Determination of species was performed using the appropriate morphological and reproductive characteristics of thalli (Kumano 2002; Cvijan *et al.* 2003; Eloranta & Kwandrans 2007; Eloranta *et al.* 2011).

Table 1. Environmental parameters of *Batrachospermum gelatinosum* localities.

Site	Date	Protected areas*	Depth of water (cm)	Degree of shade	Substratum type	Temperature (°C)	pH	Oxygen concentration (mg/l)	Saturation (%)	Nitrate (mg/l)	Phosphate (mg/l)	Ammonia (mg/l)	Hardness (mg/l)	Conductivity (µs/cm)
Site 1	X 2006	No	10-20	++	stone/gravel	15.4	8.2	8.98	109.8	/	/	/	/	440
Site 2	IV 2010	No	10-20	++	stone/gravel	10.5	7.77	9.99	97.9	3.5	3.7	0.21	265	539
Site 3	IV 2010	No	20-30	+++	Concrete	11.5	7.7	9.85	98.5	3.3	3.6	0.18	255	515
Site 4	IX 2006	NR	10-20	++	stone/gravel	13	7.9	/	/	14.2	0.24	0.08	174	346
Site 5	VI 2011	No	10-20	++	Stone	11.8	8.65	10.2	100.6	/	0.1	/	184	379
Site 6	IX 2011	No	10-15	++	Stone	14.6	8.35	8.60	91.2	1.7	0.63	0.04	245	420

*Protected areas: NR - nature reserve.
+++ - full sun, ++ - partial sun, + - full shade; / - no data.

Table 2. Environmental parameters of *Batrachospermum* sp. localities.

Site	Date	Protected areas*	Depth of water (cm)	Degree of shade	Substratum type	Temperature (°C)	pH	Oxygen concentration (mg/l)	Saturation (%)	Nitrate (mg/l)	Phosphate (mg/l)	Ammonia (mg/l)	Hardness (mg/l)	Conductivity (µs/cm)
Site 7	VIII 2005	NP	10-30	++	stone/gravel	10.7	7.25	11.11	105.5	3.4	0.20	0.37	/	55
Site 8	VIII 2010	NP	10-20	+	Stone	12	6.7	9.40	91.5	5.6	1.7	0.31		60
Site 9	IX 2009	No	10-15	+++	stone/gravel	13.2	7.2	9.35	103.7	/	/	/	/	400
Site 10	VII 2015	No	10-20	++	concrete	12	/	/	/	/	/	/	/	/

*Protected areas: NP - national parks.
+++ - full sun, ++ - partial sun, + - full shade; / - no data.

RESULTS AND DISCUSSION

Batrachospermum gelatinosum was recorded at six localities (the source of the Crni Timok River, the source of the Grza River, the Grza River Reservoir, the Boračka River, the Vapa River, the Raška River). At four localities (the Samokovska River, the Barska River, the source of the Sokobanjska Moravica River, the capped spring of Cerovac near the town of Kragujevac), the species of the given genus was not determined due to the lack of reproductive organs in the sampling period. The indicated localities are all newly recorded habitats of *Batrachospermum* in Serbia. The records of this genus at sites 6 and 10 are the first in Central Serbia.

The results of measuring physical and chemical parameters of the water are presented in Tables 1 and 2.

Batrachospermum was recorded at altitudes of from 235 m (Site 10) to 1600 m (Site 7) at localities mainly in partial or full shade on stone, gravel or concrete substrata in habitats surrounded by both broad-leaved and coniferous (Sites 7 & 8) vegetation. Specimens were found in water with temperatures ranging from 10.50°C (Site 2) to 15.40°C (Site 1). The water at the localities with *Batrachospermum* was neutral to weakly alkaline (pH 7.2 – 8.65), seldom weakly acidic (Site 8), had conductivity values ranging from 55 and 60 µS/cm (Sites 7 & 8) to 539 µS/cm (Site 2), and was well-aerated with a low concentration of biogenic salts (Tables 1, 2).

The chemical parameters measured during the sampling time period (concentrations of nitrates, phosphates and ammonia) indicate good-quality water (Tables 1, 2) in watercourses without direct anthropogenic impacts. Some of the locations are protected areas (Tables 1, 2).

Numerous authors emphasise individual ecological factors or a complex of ecological factors as being necessary for the emergence and survival of these algae. All *Batrachospermum* species are described as sensitive and stenovalent with regard to numerous ecological parameters, and every change of habitats can pose a serious threat to the survival of populations of these species (Starmach 1984).

The main factors influencing development, distribution and seasonal dynamics of members of the genus *Batrachospermum* are temperature and light (Starmach 1984; Necchi & Pascoaloto 1993); velocity of the water, its turbidity and the type of substratum (Hambrook & Sheath 1991); the water reaction (Hindak 1978); and the concentration of oxygen and nutrients, the species in question generally prefering waters with low nutrient concentration (Sheath 1984). The results of Brazilian authors (Necchi *et al*. 1999) are consistent with these data.

The genus *Batrachospermum* in Serbia is generally reported to inhabit clear, well-aerated and organic-poor waters (Blaženčić *et al*. 2002). The species *B. gelatinosum* can act as an indicator of xeno- to beta-mesosaprobic and oligotrophic waters (Eloranta & Kwandrans 2004).

Therefore, any factors causing a change of abiotic parameters in habitats are potentially unfavourable. Changes in habitats lead to change and the loss of populations of this sensitive species. Factors having possible indirect impacts at all localities include woodcutting, erosion and change of the water level and river flow speed (caused by construction of mini hydroelectric power plants, high and low rainfall, pollution, etc.). The only habitat of *B. gelatinosum* recorded so far on the territory of Vojvodina – the Batarska River (within the "Zasavica" Nature Reserve) – was completely changed after introduction of the beaver (*Castor fiber*) into this ecosystem. Not only did morphometric characteristics change (slowly flowing water was replaced by standing water with greater depth and width of the riverbed), but changes of hydroecological features were recorded as well (higher average water temperatures, changes in characteristics of the river bottom, slowing of the flow of water, alteration of the oxygen regime, etc.). The changes of ecological conditions endangered the survival of *B. gelatinosum*, and no findings of this species have been confirmed afterwards (Simić *et al.* 2007). Changes in the Krupaj River habitats (low level contaminated water) led to the absence of *Batrachospermum* populations in 2015 (Simić *et al.* 2016).

Based on the criteria of IUCN (2001) and data on the species *B. gelatinosum* available in the BAES - ex situ database, the status of this species in Serbia has been preliminarly estimated as NT (near threatened) (Simić *et al.* 2006). It should also be mentioned that *Batrachospermum gelatinosum* has been included in the regional Red List of Threatened Species, the following categories of endangerment having been determined for it in accordance with international criteria: Poland – VU (vulnerable) (Sieminska 1992); Latvia - R (rare) (Druvietis 1997); Slovakia – CR (critically endangered) http://www.sopsr.sk/webs/redlist/; and Russia – R (rare) (htt://www.econao.ru/lawdoc/reg_23.doc).

CONCLUSION

This paper provides some supplementary information about the genus *Batrachospermum*, and the presented analysis contributes to our knowledge about that rare group of algae. Species of the genus *Batrachospermum* in Serbia have to date been recorded in clean, well-aerated waters and in habitats without anthropogenic impact. Changes in physical and chemical properties of habitats and water caracteristics can lead to the loss of their populations. In order to preserve these strictly protected species in Serbia, it is necessary to register and protect their habitats. Moreover, further morphological and

molecular analysis of the collected material is needed for precise determination of the diversity of this genus in Serbia.

Acknowledgements — This work was supported by grants 43002 and 31011 from the Ministry of Education, Science and Technological Development of the Republic of Serbia. The article is a contribution submitted in recognition of Prof. Dr. Jelena Blaženčić on the occasion of her 80th birthday.

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Botanica SERBICA

REZIME

Novi podaci o distribuciji i ekologiji roda *Batrachospermum* (Rhodophyta) u Srbiji

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U radu su opisani distribucija i ekologija populacija roda *Batrachospermum*, sakupljenih u periodu od 2006. do 2015. godine na deset lokaliteta, pri čemu svi predstavljaju nove lokalitete roda *Batrachospermum* u Srbiji. Lokaliteti u Boračkoj reci i u izvoru kod Kragujevca (Cerovac) su prvi nalazi ovog roda u centralnoj Srbiji. Rod *Batrachospermum* je zabeležen na nadmorskim visinama od 235 m do 1600 m, na mestima koja se uglavnom nalaze u delimičnoj ili potpunoj senci, na kamenitoj, šljunkovitoj ili betonskoj podlozi, u vodi koja je hladna (10.5°C do 15.4°C), neutralna do slabo alkalna (pH 7.2 - 8.65) i u kojoj je elektroprovodljivost bila od 55 µS/cm do 539 µS/cm. Vrste roda *Batrachospermum* u Srbiji su zabeležene u čistim, dobro aerisanim vodama sa malom koncentracijom biogenih soli i na staništima koja su van antropogenog uticaja. Zbog toga, bilo koji faktori koji uzrokuju promenu abiotskih parametara staništa mogu dovesti do promene i nestanka populacija osetljivih vrsta ovog roda.

Ključne reči: rasprostranjenje, ekologija, *Batrachospermum*, Rhodophyta, Srbija