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Original Scientific Paper

First record of the genus *Decussata* (Patrick) Lange-Bertalot (Bacillariophyta) in Serbia – distribution of the rare species *D. hexagona* (Torka) Lange-Bertalot

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ABSTRACT: The aim of this paper is to present the distribution of *Decussata hexagona*, a new species in diatom flora of Serbia. Seventy-two diatom samples were collected from six localities along the Vrla and Rasina Rivers during 2011 and 2012. *Decussata hexagona* was recorded with single specimens or low relative abundance (0.3%) from epilithic communities in seven samples from the Vrla River and in one sample from the Rasina River. It was recorded in unusual epilithic habitats of running neutral to low-alkaline waters with low ion content.

Keywords: Bacillariophyta, *Decussata hexagona*, distribution, autecology, Vrla River, Rasina River

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INTRODUCTION

The diatoms (Bacillariophyta) are a successful group of eukaryotic, photosynthetic microorganisms. With over 64,000 named entities, they are one of the most diverse groups of algae (Kociolek *et al*. 2015). A review of the literature reveals the presence of 661 taxa from subclass Bacillariophycidea D.G.Mann within 65 genera in Serbia (Krizmanić *et al*. 2011). Recently published new records of diatom taxa from Serbia (Andrejić *et al*. 2012; Vidaković *et al*. 2014; Krizmanić *et al*. 2015; Vidaković *et al*. 2016) expand the diversity of known species, but not the number of genera.

The genus *Decussata* (Patrick) Lange-Bertalot was raised from subgenus level (subg. *Decussata* Patrick) in a group including *Navicula placenta* Ehrenberg and similar species (Lange-Bertalot 2000). It was described entirely on the basis of valve characters. *Decussata* is characterised by solitary living cells with broadly elliptical or linear valves and rounded or rostrate ends. The most distinctive characteristic of this genus is the arrangement of areolae in its members. Areolae are formed from three systems of striae which cross at 60-80° to formi a regular decussate (quincunx) pattern (Lange-Bertalot 2001) that is completely unlike the one seen in *Navicula* sensu stricto taxa. Edlund *et al*. (2006) placed the genus *Decussata* in the family Mastogloiaceae and the order Mastogloiales. This genus is hard to confuse with any other taxon.

The genus *Decussata* consists of three taxa: *D. placenta* (Ehr.) Lange-Bertalot & Metzeltin, *D. placenta* var. *obtusa* (Meister) Lange-Bertalot and *D. hexagona* (Torka) Lange-Bertalot (Lange-Bertalot 2000). *Decussata hexagona* was originally described as *Navicula hexagona*, from Kreise Neustadt (Oberschlesien, Germany) (Torka 1933). Krasske described two taxa, *Navicula placenta* var. *minor* Krasske (Krasske 1923) and *N. placenta* var. *parallela* Krasske (Krasske 1925), which Lange-Bertalot (2000) considered to be synonymous with *D. hexagona*.

Decussata hexagona is a rare species which grows in populations with small individual numbers. It is found only in Europe, in peat bogs and intermittently wet bryophytes (Torka 1933; Lange-Bertalot 2001), contrary to *D.*

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placenta, which has a cosmopolitan distribution (Patrick & Reimer 1966; Mölder & Tynni 1967; Krammer & Lange-Bertalot 1986; Lange-Bertalot 2000, 2001).

Herein, we present the first record of *D. hexagona* in Serbia, based on examination of algal samples from the Vrla and Rasina Rivers, together with new habitat details and environmental data.

MATERIALS AND METHODS

The Vrla River is located in the southeastern part of Serbia (Fig. 1). It empties into the Southern Morava River in the center of Vladičin Han at an elevation of 323 m, and belongs to the Black Sea drainage basin. The springs that are the source of the Vrla River are located on Mt. Vardenik. The river's total length is around 28 km and its drainage basin has an area of about 500 km^2. The Vrla River belongs to the Vlasina's hydro system due to the close proximity of its source springs to the Vlasina River and Lake Vlasina (Đeković *et al*. 2010).

The Rasina River is located in the central part of Serbia (Fig. 1). It empties into the Western Morava River 5 km downstream from the city of Kruševac, and (like the Vrla) belongs to the Black Sea drainage basin. Arising on slopes of the mountains Goč and Željin, the Rasina River is formed by the confluence of the Velika River and the Burmanska River at an elevation of 1340 m. It measures 92 km in length and has a drainage basin with an area of about 981 km^2. The drainage basin of the Rasina River has a distinctly asymmetrical shape, with all right-hand tributaries, except for the Zagrža River, which is a left-hand tributary (Gavrilović & Dukić 2002).

A total of 72 samples were collected during six sampling periods (May, July, September and November of 2011; March and May of 2012) at six localities along both rivers (Fig. 1).

Water temperature, pH and conductivity were measured using a WTW Multi 340i instrument. Amonium ions, nitrates, oxygen, total P, chlorides and sulphates were gauged with a Lovibond MultiDirect photometer. These parameters were measured *in situ,* except in the case of BOD_5, which was measured in the laboratory according to APHA protocols (1998). Epilithic diatoms were collected from boulders and cobbles by brushing stones with a toothbrush during the sampling period. At least five stones were randomly collected at each sampling site and brushed, after which the resulting suspensions were pooled to form a single sample that was put in a labeled plastic bottle. The material was immediately fixed with formaldehyde and preserved in a final concentration of 4%.

For preparation of permanent slides, algal samples were treated by the standard method with cold acid (Krammer & Lange-Bertalot 1986). Permanent slides were mounted using Naphrax medium. At least 400 valves were counted on each slide in random transects in order to calculate the relative abundance (%) of each taxon. Light microscopy was performed using a Zeiss AxioImager M1 instrument with DIC at 1000x magnification. Images were obtained using an AxioCam MRc5 digital camera and AxioVision4.8 software. The terminology of valve morphology used and identification were according to Lange-Bertalot (2001).

Aliquots of algal samples and permanent slides were deposited in the diatom collection of the Institute of Botany and Botanical Garden "Jevremovac", Faculty of Biology, University of Belgrade.

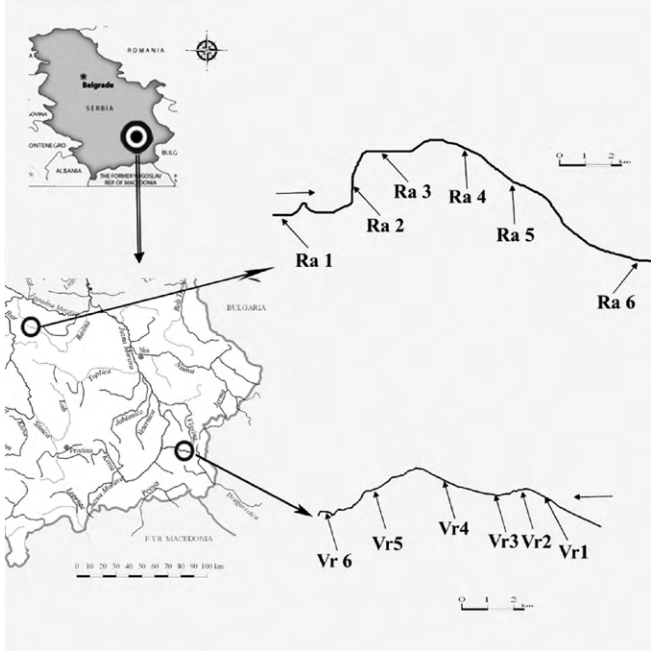

Fig. 1. Sampling sites along the Vrla (Vr) and Rasina (Ra) Rivers.

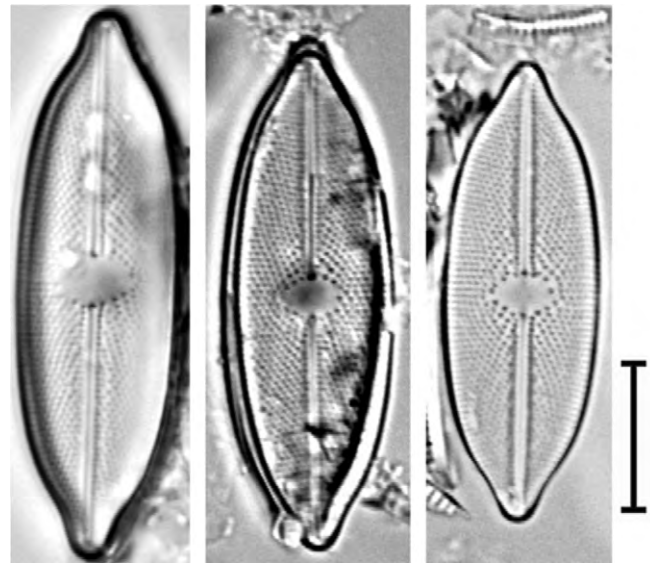

Fig. 2. LM micrographs of *D. hexagona*; scale bar = 10 μm.

RESULTS

Decussata hexagona was recorded in seven samples from the Vrla River and in one sample from the Rasina River.
Class: Bacillariophyceae
Subclass: Bacillariophycidae
Order: Mastogloiales
Family: Mastogloiaceae
Genus: *Decussata*
Species: *Decussata hexagona* (Torka) Lange-Bertalot 2000: 671 (Lange-Bertalot 2000) (Fig. 2)
Basionym: *Navicula hexagona* Torka (1933)

Valves linear-elliptical with narrow ends which are finally obtusely rounded, 29.9 – 38.0 µm long, 10.3 – 11.9 µm wide (Fig. 2). Valve face flat. Raphe filiform. Axial area linear, central area transapically elliptical. Length/width ratio ranging from 2.9 to 3.5. Striae from parallel to slightly radial, 24 – 26 in 10 µm. It is difficult to confuse this species with any other.

It was identified from single specimens (+), except in one sample from the Vrla River, where it was present with low relative abundance (0.3%) (Table 1).

Physicochemical characteristics of the studied rivers are given in Table 2. Sites were located at an elevation of 1400 m a.s.l. (above sea level) to 988 m a.s.l. along the Vrla River; and at 680 m a.s.l. along the Rasina River.

DISCUSSION

The gross valve morphology of *D. hexagona* from the Vrla and Rasina Rivers in Serbia agrees with the range of dimensions given in the literature (length 25 – 44 µm, breadth 9 – 13 µm according to Lange-Bertalot 2001). Although *D. hexagona* has the central and axial area, striae and puncta as in the close species *D. placenta*, these two species clearly differ in outlines, breadth of valves and shape of the central area (Stancheva & Temniskova 2006). In *D. hexagona* the valves are linear to linear-elliptical with a breadth of 9 – 13 µm, and the central area is irregularly outlined with isolated puncta.

This species is known only from Europe (Krasske 1923, 1925; Torka 1933; Lange-Bertalot 2001; Levkov *et al*. 2001; Levkov *et al*. 2005; Stancheva & Temniskova 2006; Hafner *et al*. 2008; Kapetanović *et al*. 2011). Sawai & Nagumo (2003) listed *D. hexagona* as a diatom present in the Alsea Bay salt marsh on the Pacific Coast of the USA, but, fig. 20 on page 39 does not show *D. hexagona*. The valves are elliptical, with rostrate-apiculate apices. The axial area is narrow and expanded at the center to form a transapically oval to circular central area. The external proximal raphe ends are prominent and dilated. To judge from all features, it represents *D. placenta,* a cosmopolitan species (Kociolek 2011).

Decussata hexagona develops best in moss-turfs and bog-like habitats (Lange-Bertalot 2001). In Macedonia, *D. hexagona* was found in small peat bogs in the Nidze Mountains and in the Shara Mountains, at an elevation of over 1400 m (Edlund *et al*. 2006). The ponds were almost without water and had a large quantity of detritus. In addition to this, *D. hexagona* was found in the central Sredna Gora Mountains (Bulgaria) in Holocene sediments of mountain peat bogs at elevations of 1000 – 1500 m (Stancheva & Temniskova 2006). The fen Bijambare Protected Landscape in Bosnia and Herzegovina, where this species was also found, is situated at an elevation of 930 m (Kapetanović *et al*. 2011). We found *D. hexagona* within epilithic communities, in contrast to published data. Our material was found at elevations that correspond well with published figures: the taxon was recorded at sites located at elevations ranging from 680 m to 1400 m.

Decussata hexagona is elsewhere mentioned as being infrequent and occurring only in the guise of single or sparse specimens (Lange-Bertalot 2001), as in the case

Table 1. Abundance of *D. hexagona* in epilithic communities from the observed rivers.

River	Vrla				Rasina
Locality	May 2011	July 2011	Sep. 2011	May 2012	May 2012
2		+			
3	+				+
4		+	+		
5	+		+	0.3%	

(+) single specimens; relative abundance (%). For the river locality numbers, see Fig. 1.

Table 2. Physicochemical parameters of water in the Vrla and Rasina Rivers.

Parameters/Rivers	Vrla	Rasina
Water temperature (°C)	10-15.2	9.2
pH	7.01-8.01	7.66
Conductivity (µS/cm)	62.1-98.1	140.1
O_2 (mg/l)	9-10.61	10.2
Total phosphorus (mg /l)	0.0328-0.0911	0.0149
Ammonium ions (mg /l)	<0.0189-0.188	0.0875
BOD_5 (mg/l)	<0.2-8.6	5.6
Nitrates (mg/l)	1.5-2.1	2.4
Chlorides (mg/l)	0.865-1.315	0.831
Sulphates (mg/l)	5.321-7.832	8.452

of our observations (sporadic or with very low relative abundance, 0.3%). The relative abundance of *D. hexagona* from a small fen habitat in Bosnia and Herzegovina was also low (0.2-0.3%) (Kapetanović *et al*. 2011). Populations from small peat bogs in the Nidze Mountains of Macedonia were with a relative abundance of less than 1% (Edlund *et al*. 2006).

Data about the habitat preferences of rare species are often insufficient, so that fundamental research is still necessary (Wojtal 2001, 2004). The scantiness of algological investigations of marshes, peat bogs and similar habitats in Serbia is possibly the reason for the absence of *D. hexagona* records so far.

Most of the peat bogs with *D. hexagona* in Macedonia were neutral to slightly alkaline environments with low conductivity (Edlund *et al*. 2006). The pH values for sediments of the mountain peat bogs in Bulgaria (5.63 – 6.23) (Stancheva & Temniskova 2006) and for the fen in Bosnia and Herzegovina (4.54 – 6.56) (Kapetanović *et al*. 2011) were lower. Conductivity at the sampling sites from the fen in Bosnia and Herzegovina (37 – 125 μS cm^{-1}) was higher than at our sites, but concentrations of nutrients were similar to our data.

The dominant taxa at the river localities of *D. hexagona* in Serbia were *Achnanthidium minutissimum* (Kützing) Czarnecki, *A. subatomus* (Hustedt) Lange-Bertalot, *Nitzschia abbreviata* Hustedt and *Cocconeis placentula* var. *lineata* (Ehrenberg) Van Heurck. Their abundance varied from 15 to 24%, depending on the season and locality. The investigated peat bogs in Macedonia were characterised by the following dominant taxa: *Achnanthes* sensu lato, *Navicula* Bory and *Placoneis* Mereschkowsky species (Edlund *et al*. 2006). Species from the genera *Pinnularia* Ehrenberg, *Eunotia* Ehrenberg, *Navicula* Bory and *Cymbella* C.Agardh were dominant at sites from the mountain peat bogs in Bulgaria (Stancheva & Temniskova 2006). Acidobiontic and acidophilous species such as *Eunotia exigua* (Brébisson in Kutzing) Rabenhorst, *E. glacialis* F. Meister, *E. paludosa* Grunow and *Pinnularia rhombarea* Krammer in Metzeltin & Lange-Bertalot were dominant at the fen in Bosnia and Herzegovina (Kapetanović *et al*. 2011). In our study, the dominant species are mainly ones that have wide ecological valences, unlike the dominant species in other habitats.

CONCLUSION

The genus *Decussata*, with the rare species *D. hexagona*, was recorded for the first time in Serbia. So far, the species *D. hexagona* is known only in Europe, as a bryophytic diatom in moss-turfs and peat bog-like habitats. In the Vrla and Rasina Rivers, it was recorded in unusual epilithic habitats of running neutral to low-alkaline waters with low ion content. These findings contribute to knowledge of the diatom flora of Serbia and expand the limits within which the species was previously recorded.

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Botanica SERBICA

REZIME

Prvi nalaz roda *Decussata* (Patrick) Lange-Bertalot (Bacillariophyta) u Srbiji – rasprostranjenje retke vrste *D. hexagona* (Torka) Lange-Bertalot

Jelena Krizmanić, Olga Jakovljević, Danijela Vidaković i Jelena Jovanović

Epilitske silikatne alge sakupljene su sa po 6 lokaliteta duž reka Vrle i Rasine tokom 2011. i 2012. godine. Analizom ukupno 72 algološka uzorka zabeleženo je prisustvo novog roda *Decussata* za floru silikatniih algi Srbije, na 5 lokaliteta u 8 uzoraka. Identifikovana vrsta, *D. hexagona*, je retka evropska vrsta sa malom brojnošću na tipičnim lokalitetima, močvarama i tresetištima. Po prvi put, vrsta je zabeležena u epilitskim zajednicama malih reka, u neutralnoj do slabo alkalnoj vodi sa niskom koncentracijom jona.

Ključne reči: Bacillariophyta, *Decussata hexagona*, rasprostranjenje, epilitska zajednica, reka Vrla, reka Rasina.