



## DISTRIBUTION OF *DREISSENA POLYMORPHA* IN HUMAN- MADE LAKES ALONG THE LESNOVSKA RIVER (TRIBUTARY OF ISKAR RIVER, DANUBE DRAINAGE BASIN)

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**Abstract:** The distribution of invasive zebra mussel *Dreissena polymorpha* in seven sand-pit lakes and two reservoirs was studied. The water bodies are located along the Lesnovska River (tributary of Iskar River, Danube drainage basin). The occurrence of the species in the Chepintsi Lake was confirmed. The species was newly recorded in the Negovan Lakes (small and big), in the Kazichene Lake and in the Ognyanovo Reservoir. Morphology and quantitative characteristics of invasive populations were studied. The highest quantitative parameters were recorded in Chepintsi Lake, which was probably the earliest infested. Potential threats as a result of further spread of zebra mussel along the Iskar River were mentioned.

**Key words:** *Dreissena polymorpha*, invasion, abundance, biomass, sand-pit lakes, reservoirs

### INTRODUCTION

Zebra mussel *Dreissena polymorpha* (Pallas, 1771) is an invasive species that may cause serious ecological and economic problems (LUDYANSKIY *et al.*, 1993, NALEPA & SCHLOESSER, 1993). It is considered a native invasive species to Bulgaria. It was reported initially in the Danube River, the lowest reaches of its tributaries and the Black Sea coastal lakes and rivers (KREGLINGER, 1870, WOHLBEREDT, 1911, DRENSKY, 1947, VALKANOV, 1957, RUSSEV *et al.*, 1994, ANGELOV, 2000). In recent years, however, the species spread rapidly into the Bulgarian inland water bodies (HUBENOV, 2002, 2005, TRICHKOVA *et al.*, 2007, 2008). Some of the infested reservoirs, such as Ogosta, Rabisha and Ovcharitsa supported abundant zebra mussel populations (TRICHKOVA *et al.*, 2008). The infestations had strong impact on the infested reservoirs affecting different industries such as electricity production (damages on Thermoelectric Power Plants, HUBENOV, 2002), irrigation, aquaculture, etc.

The first record of zebra mussel in Sofia region was reported in the sand excavation lake Chepintsi (HUBENOV, 2005). The goal of the present study was to monitor this and other human-made lakes in the drainage basin of the Lesnovska River for the occurrence and abundance of zebra mussel.

## MATERIALS AND METHODS

Seven sand-pit lakes and two reservoirs were studied. They are situated in the drainage area of the Lesnovska River - the biggest right tributary of the Iskar River in the region of Sofia (Fig. 1). The Iskar River is the biggest tributary of the Danube in the Bulgarian part.

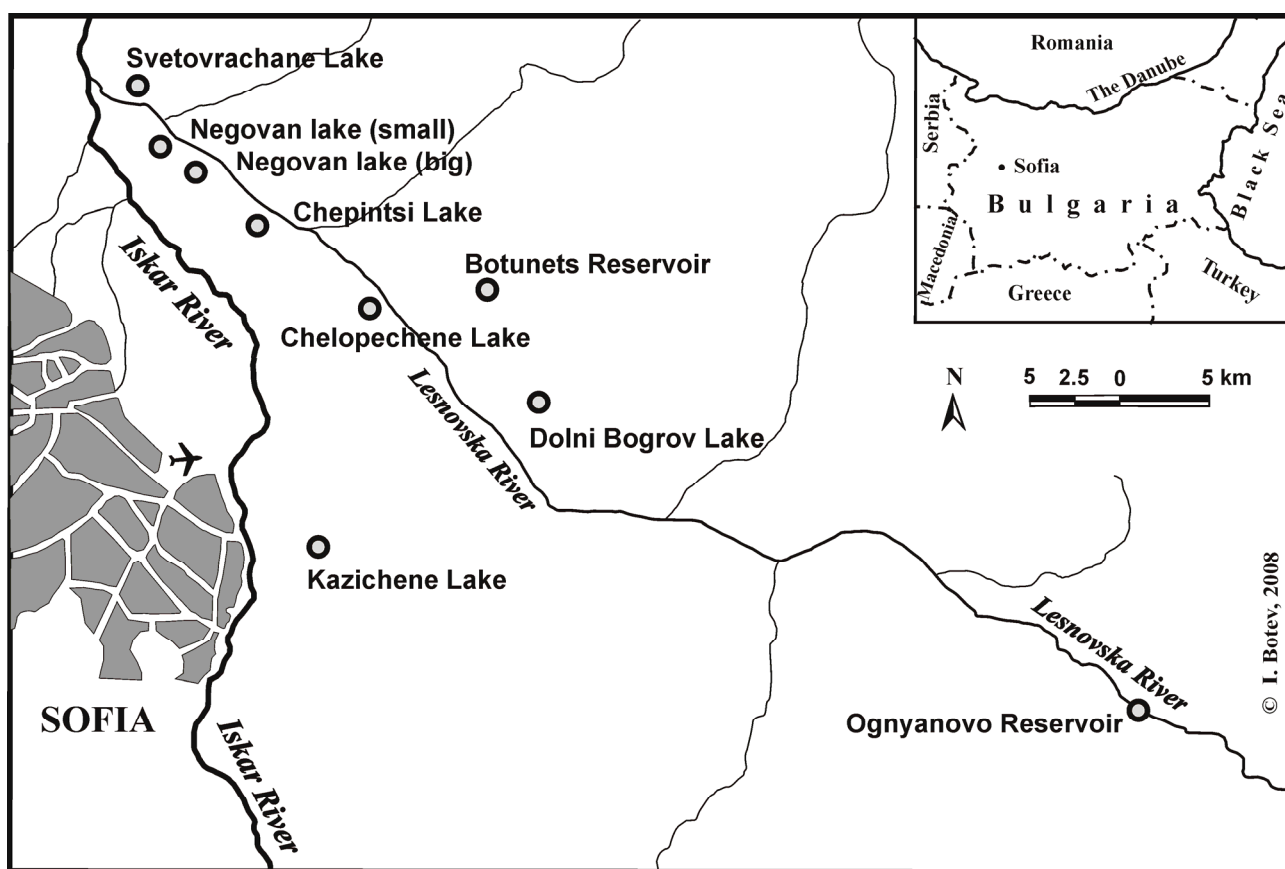


Fig. 1. Survey lakes and reservoirs within the Lesnovska River basin.

The study water bodies are very different in surface area and depth (Table 1). The largest one is Ognyanovo Reservoir and the smallest Svetovrachane Lake. They are situated to the North, North-East and to the East of Sofia at altitudes from 519 m a.s.l. (Svetovrachane Lake) to 630 m a.s.l. (Ognyanovo Reservoir). The lakes were artificially made for excavation of sand and gravel (Table 1). Some of them are currently not in use. The reservoirs Ognyanovo and Botunets are used for water supply of Kremikovtsi Metal-Processing Plant. At present, all water bodies are used for recreational fishing.

**Table 1.** *Surface area, maximum depth and use of the sampled water bodies.*

Water body	Surface Area, km <sup>2</sup>	Maximum Depth, m	Use
Ognyanovo Reservoir	4.16	47	Industrial water supply; Irrigation; Recreational fishing
Botunets Reservoir	0.42	5.5	Industrial water supply; Recreational fishing
Kazichene Lake	0.31	6.5	Former sand-pit lake; Aquapark; Recreational fishing
Dolni Bogrov Lake	0.37	4.5	Former sand-pit lake; Irrigation; Recreational fishing
Chelopechene Lake	0.82	15	Former sand-pit lake; Recreational fishing
Chepintsi Lake	0.24	19	Sand-pit lake; Recreational fishing
Negovan Lake (big)	0.64	15	Sand-pit lake; Recreational fishing
Negovan Lake (small)	0.10	2.8	Former sand-pit lake; Recreational fishing
Svetovrachane Lake	0.04	2	Former sand-pit lake; Recreational fishing

The lakes were visited in two periods: 17-18 May 2008 and 2-5 July 2008. Biological samples were collected in May and July, while physico-chemical samples in July 2008 (Table 2).

**Table 2.** *Physico-chemical parameters of the water bodies sampled in July 2008.*

Water body	Sampling Depth, m	Water Temp., °C	Dissolved Oxygen, mg/dm <sup>3</sup>	pH	Electroconductivity, µS/cm	Secchi Disk Transp., cm	Ca, mg/ dm <sup>3</sup>
Ognyanovo Reservoir	0	25.3	7.57	8.6	283	530	34.07
Ognyanovo Reservoir	15	18.0	3.53	7.8	240		34.07
Botunets Reservoir	0	26.0	10.9	8.9	355	55	-
Botunets Reservoir	4	26.2	7.28	8.6	365		40.08
Kazichene Lake	0	27.0	8.16	8.9	270	140	26.05
Kazichene Lake	6	20.5	0.52	7.6	279		40.08
Dolni Bogrov Lake	0	27.3	7.81	8.1	519	200	-
Dolni Bogrov Lake	3	27.1	5.60	8.0	520		24.05
Chelopechene Lake	0	24.4	11.54	8.3	592	62	-
Chelopechene Lake	6.2	19.6	1.04	7.5	454		42.08
Chepintsi Lake	0	25.8	8.04	8.3	414	245	18.04
Chepintsi Lake	11	23.2	4.17	7.8	425		38.08
Negovan Lake (big)	0	27.0	8.45	8.5	554	145	42.08
Negovan Lake (big)	11	19.7	3.35	7.9	503		38.08
Negovan Lake (small)	0	30.0	9.10	8.2	952	45	40.08
Svetovrachane Lake	0	29.2	8.20	9.9	378	40	24.05

Transparency was measured by Secchi disk. Water temperature, dissolved oxygen, pH and electroconductivity were measured on-site using portable oxygen, pH and conductivity meters - Schott GMBH and Hanna. Half liter water samples were taken from every water body using water sampler according to Hydrobios PVS 416865, then stored in plastic bottles and transported to the laboratory in a cooler with ice for determining calcium concentrations.

Quantitative samples of adult zebra mussels were collected by Petersen bottom sampler of medium size (17.0x16.5 cm) and by hand bottom dredge (pole scraper) with 17 cm length of the scraper, 500  $\mu\text{m}$  mesh size and 1.70 cm length of the rod. The samples were fixed first in formaldehyde and later at the laboratory, in alcohol.

The following morphometric parameters of zebra mussels were measured: shell length (SL) - the maximum anteroposterior dimension of the shell; shell height (SH) – the maximum dorsal-ventral dimension of the shell; and shell width (SW) - the maximum lateral dimension with valves closed. These measurements were made on 30 specimens from each water body where zebra mussel was found. For the measurements, a calliper and electronic balance to the nearest 0.1 mm and 0.1 g, respectively were used.

Absolute abundance and total biomass of zebra mussel populations were calculated per square meter according to the standard hydrobiological methods – ISO 8265/1988.

## RESULTS

The water temperature in all study lakes was in the range from 18°C to 30°C (Table 2). Straight stratification of the water was registered in the deeper water bodies such as Ognyanovo Reservoir, Negovan big lake, Chepintsi, Chelopechene and Kazichene lakes. The other lakes are shallow holo-polymictic water bodies with no significant differences in temperature registered.

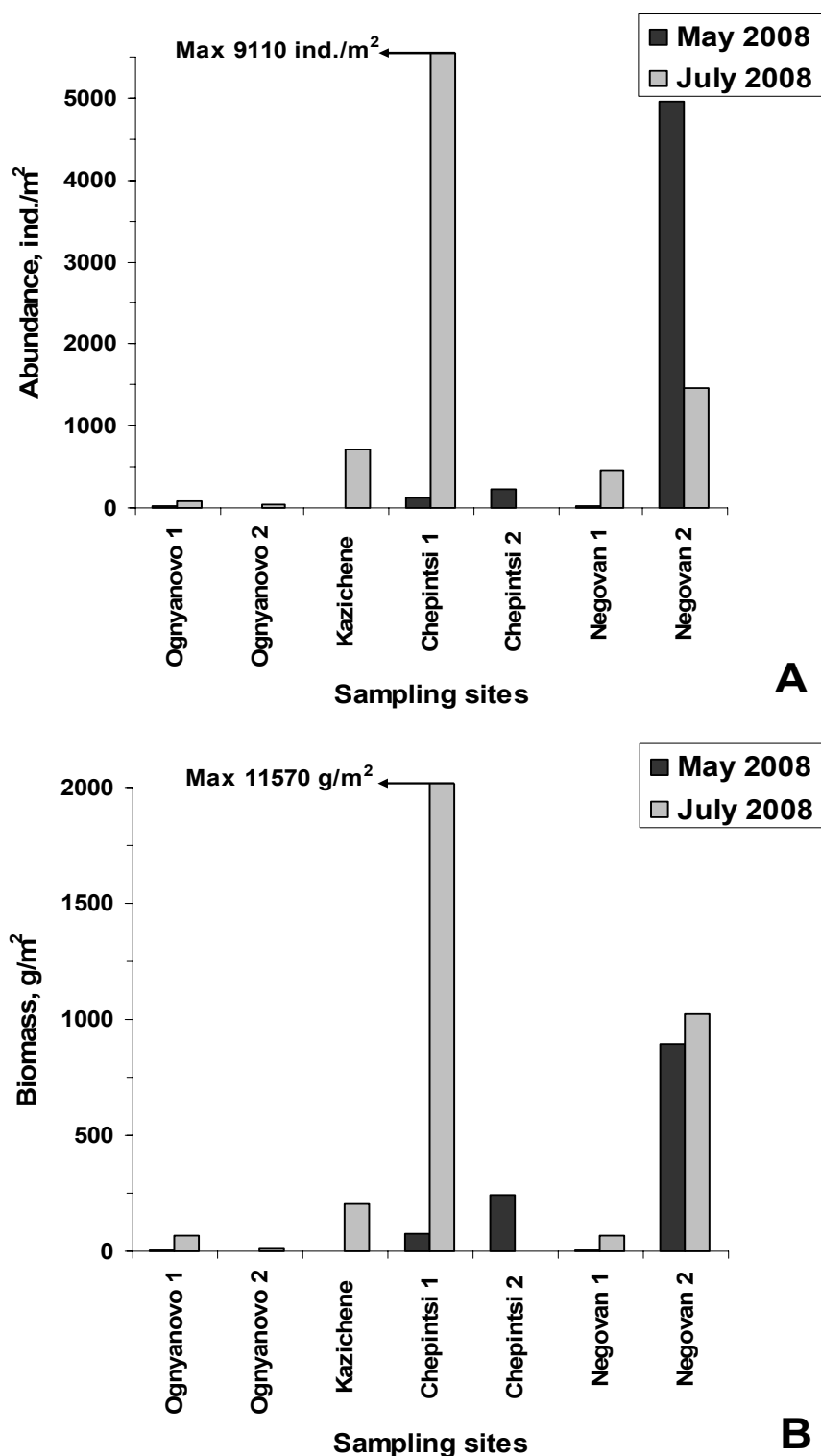
The values of pH ranged from 7.5 (at the bottom of Chelopechene) to 9.9 (in Svetovrachane) and indicated from slightly to strongly alkaline lakes.

The oxygen concentration at the bottom ranged from 0.52 mg/dm<sup>3</sup> (Kazichane) to 7.28 mg/dm<sup>3</sup> (Botunets); and oxygen concentration at the surface from 7.57 mg/dm<sup>3</sup> (Ognyanovo) to 11.54 mg/dm<sup>3</sup> (Chelopechene) (Table 2).

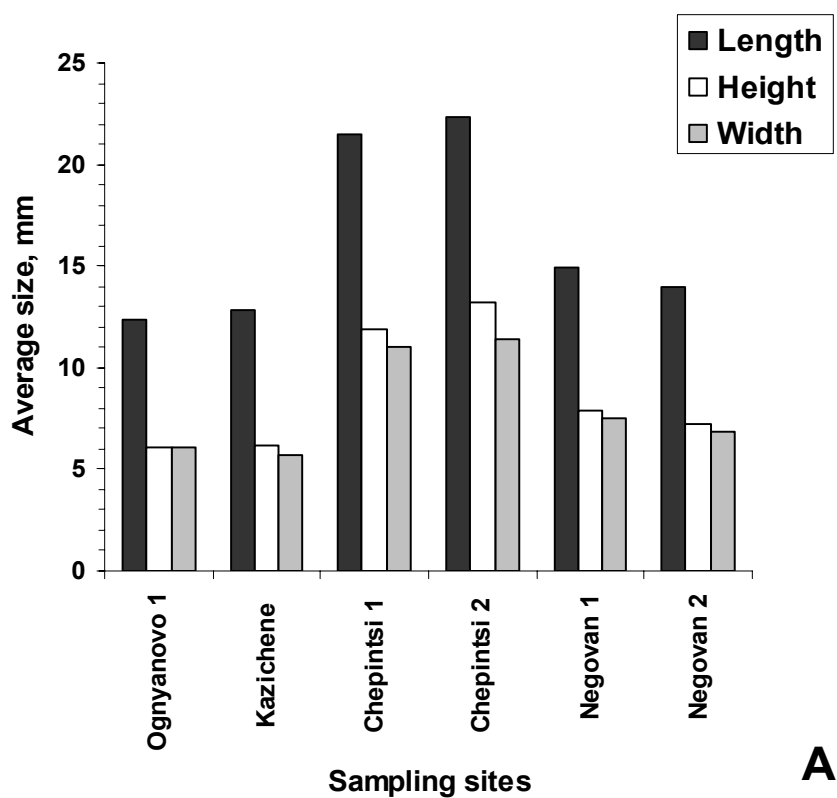
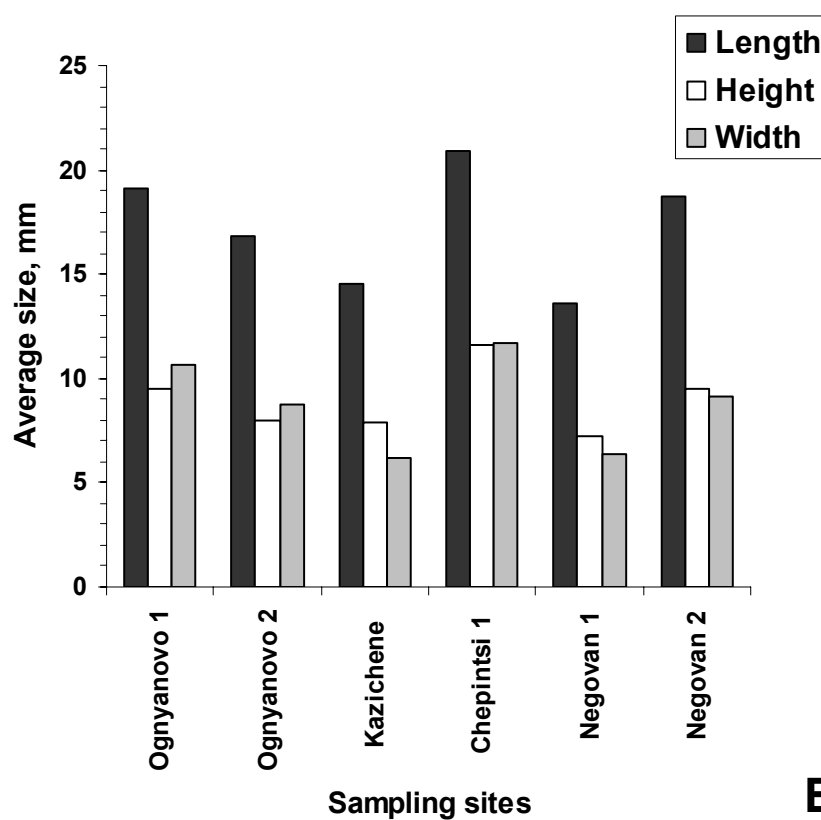
The electroconductivity in the lakes ranged from 240  $\mu\text{S}/\text{cm}$  (Ognyanovo) to 952  $\mu\text{S}/\text{cm}$  (Negovan small); and the calcium concentration from 18.04 mg/dm<sup>3</sup> (Chepintsi) to 42.08 mg/dm<sup>3</sup> (Table 2). The highest value of water transparency was measured in the Ognyanovo Reservoir (530 cm) and the lowest in the lakes Negovan small and Svetovrachane (45 and 40 cm, Table 2).

The zebra mussels were recorded in five of the nine study water bodies, as follows: established populations in lakes Chepintsi, Negovan (big lake), Kazichene and Ognyanovo Reservoir; and a single specimen in Negovan small lake (found in May 2008). Most abundant was the population in Chepintsi Lake reaching 9110 ind./m<sup>2</sup> in July, followed by big Negovan Lake with 4955 ind./m<sup>2</sup> recorded in May (Fig. 2A). The highest biomass (11570 g/m<sup>2</sup>) was recorded again in Chepintsi Lake (Fig. 2B). The

mean shell lengths of zebra mussels collected ranged from 12.4 mm (Ognyanovo) to 22.3 mm (Chepintsi); the mean shell heights from 6.1 mm to 13.2 mm (Chepintsi); the mean shell widths from 5.7 mm (Kazichene) to 11.7 mm (Chepintsi) (Fig. 3A,B).



**Fig. 2.** Abundance (A) and biomass (B) of zebra mussel populations in water bodies along the Lesnovska River sampled in May and July 2008. (1, 2): Numbers of sampling sites.

**A****B**

**Fig. 3.** Mean lengths, heights and widths of zebra mussels in water bodies along the Lesnovska River sampled in May 2008 (A) and July 2008 (B). (1, 2): Numbers of sampling sites.

## DISCUSSION

Recently, a rapid spread of zebra mussel in the Bulgarian inland water bodies has been observed. The species was reported in the Reservoirs Rabisha, Ogosta, Kovachitsa and Shishmanov Val (Archar and Ogosta river basins), Ovcharitsa, Zhrebchevo, Pyasachnik and Malko Sharkovo (Maritsa River basin), Ticha (Black Sea basin), as well as the Lake Chepintsi (Iskar River basin) (Hubenov 2005, Trichkova et al., 2007, 2008). Our results confirmed the occurrence of zebra mussel in the Chepintsi Lake. Furthermore, the species was newly recorded in the Negovan Lakes (small and big), in the Kazichene Lake and in the Ognyanovo Reservoir.

The highest abundance and biomass of zebra mussel populations were found in Chepintsi Lake in July 2008 (Fig. 2A,B). In this lake, the mean sizes of individuals were also the highest – mean shell lengths of 22.3 and 20.9 mm, mean shell heights of 13.2 and 11.6 mm and mean shell widths of 11.4 and 11.7 mm (Fig. 3A,B). This is probably the earliest infested lake in the region.

In the big sand-pit lake Negovan, also comparatively high quantitative parameters of zebra mussel population were registered: the abundance was 4955 ind./m<sup>2</sup> in May and 1463 ind./m<sup>2</sup> in July; and the biomass reached 1024 g/m<sup>2</sup> in July. Similar to Chepintsi Lake, these parameters were recorded only in the littoral zone. Zebra mussels in the two lakes occurred at depths from 0.50 to 2.20 m attached to stones, sunk tree roots, branches, etc. This distribution differs from previously reported in the Rabisha and Ogosta Reservoirs, where zebra mussels were found at depths from 3.5 to 10 m (TRICHKOVA *et al.*, 2008). One factor which may limit the distribution of zebra mussels in the lakes is the type of substrate. Usually, zebra mussels preferably colonize hard substrate and their density was determined by substrate size (KARATAYEV *et al.*, 1998, JONES & RICCIARDI, 2005). Other molluscs and macrophytes are also common substrates for zebra mussel colonization (RICCIARDI *et al.*, 1996, DIGGINS *et al.*, 2004). The bottom substrate in Chepintsi Lake was dominated by clay and sand overgrown with *Myriophyllum* spp. and *Potamogeton* spp. in the littoral. In the Negovan big lakes, clay and coarse sand dominated at shallow part and thick clay, mud and stones at deeper part. So the possible reasons for the distribution of zebra mussels in the two lakes can be, on the one hand, the prevalence of soft substrates in deeper parts, and on the other hand, the intensive excavation work carried out especially in the Chepintsi Lake.

In the Kazichene Lake, the bottom substrate was mainly sand and gravel at shallow part and muddy-clay and sand at deeper part. Zebra mussels were again found only in the littoral zone: juveniles attached to stems of *Myriophyllum verticillatum*, and adults attached to a concrete facility at a depth of about 1 m.

The lowest abundance and biomass of zebra mussels were recorded in the Ognyanovo Reservoir (e.g. 80 ind./m<sup>2</sup> and 65 g/m<sup>2</sup> in July). The conditions in this reservoir appeared to be favorable for the development of zebra mussels: the bottom substrate in the littoral was mostly hard (rocks, stones, sunk trees, concrete blocks, etc.); the oxygen concentration was favorable (Table 2); the waters of the Lesnovska River in the upper reaches where the reservoir is located were not polluted. Therefore, a possible reason for the low quantities of zebra mussels are the significant

fluctuations in the reservoir water level every summer and autumn (with 3-10 m approximately). This is because of the use of the reservoir for industrial water supply to Kremikovtzi Metal-Processing Plant. The regular drainage of the parts with most preferable depths to zebra mussel, most likely, influenced negatively its development in the reservoir.

According to our previous study in the North-West Bulgaria, zebra mussels preferably colonized reservoirs with moderate amount of nutrients (TRICHKOVA *et al.*, 2007). Our present results to some extent confirm this assumption. In both cases we use electrical conductivity as indicator of nutrients content. It appeared that in lakes with the highest electrical conductivity, such as Negovan small lake, Dolni Bogrov and Chelopechene (Table 2) established populations of zebra mussels were not found. In the small Negovan Lake, where the highest value of electrical conductivity was measured, only one zebra mussel specimen was recorded in May. This shows that the species was introduced to the lake, but because of unfavorable conditions was not able to establish abundant population. In Botunets Reservoir and Svetovrachane Lake, where zebra mussels were not found as well, the values of electrical conductivity were lower than other non-infested lakes (Table 2). The Botunets Reservoir collects water for industrial supply and Svetovrachane Lake is the most downstream located lake near the confluence of Lesnovska and Iskar rivers. The bottom substrate of these lakes was thick anaerobic mud. It is possible that pollutants from the Lesnovska River which is heavily loaded with different xenobiotics in its lower reaches to have negative impact on these lakes which can restrict a potential zebra mussel invasion.

All lakes, where zebra mussel was not found, except Dolni Bogrov, were characterized with low values of Secchi disk transparency (from 40 to 62 cm, Table 2). This shows phytoplankton blooms which is also indication of advanced eutrophication. On the other hand, as efficient filter feeders, zebra mussels are responsible for the considerable increase in Secchi disk transparency in the infested water basins (FAHNENSTIEL *et al.*, 1995, MACISAAC, 1996). In our previous study, the zebra mussel abundance showed a statistically significant strong positive correlation with Secchi disk transparency (TRICHKOVA *et al.*, 2007). The present results do not confirm this correlation, but they also show that the transparency was much higher in the infested lakes (Chepintsi, Negovan big, Kazichene, Ognyanovo Reservoir, Table 2). The highest value was recorded in the Ognyanovo Reservoir, which was less disturbed and as already mentioned characterized with clean waters.

As potential zebra mussel dispersal mechanisms in the North-West Bulgaria were identified the transports of larvae or adult individuals with fishing equipment, boats and fish stocking material from the Danube or already infested water bodies (TRICHKOVA *et al.*, 2007). Most likely, similar mechanisms operated in the lakes along the Lesnovska River. Probably the Chepintsi Lake was the earliest infested and the infestation process in some of the other lakes started from this lake. The big Negovan Lake is located about 4 km far from Chepintsi and different fishing equipment and boats can be easily transported between them. Both lakes are easily accessed and used also for recreation and sports fishing. Kazichene Lake is also



easily accessed and popular as aquapark as well as a place for sports fishing. The Ognyanovo Reservoir was built in the early 1990s. Ten years after its storage there were strict restrictions for its use for fishing. After 2000 these restrictions were removed and maybe in the last 5 years the zebra mussel was transferred to the reservoir by some fishing equipments.

## CONCLUSION

The obtained results showed that four of the study water bodies along the Lesnovska River were infested by zebra mussel and supported abundant populations. In the other lakes the advanced eutrophication process suppressed the development of the species. The degree of infestation in the Chepintsi Lake was the highest.

The spread of zebra mussel in the basin of the Lesnovska River was most likely due to the transport of larvae or adult individuals with the help of fishing equipment and boats. There is a potential threat of introducing of the species to the big reservoirs along the Iskar River, such as Iskar, Pancharevo and Pasarel in the near future. Therefore, further studies including regular monitoring and risk assessment together with control measures on recreation, fishing and industrial activities in the lakes must be initiated.

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## **РАЗПРОСТРАНЕНИЕ НА *DREISSENA POLYMORPHA* В ИЗКУСТВЕНИ ЕЗЕРА ПО ПОРЕЧИЕТО НА Р. ЛЕСНОВСКА (ПРИТОК НА Р. ИСКЪР, ДУНАВСКИ БАСЕЙН)**

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### **(Резюме)**

Изследвано е разпространението на инвазивния вид мида зебра *Dreissena polymorpha* в седем кариерни езера и два язовира, разположени по поречието на р. Лесновска (приток на р. Искър, Дунавски басейн). Потвърдено е присъствието на мидата в ез. Чепинци. Видът е установен за пръв път в голямото и малкото езера Негован, в ез. Казичене и в яз. Огняново. Изучени са морфологията и количествените параметри на инвазивните популации. Най-голяма численост и биомаса има популацията в ез. Чепинци, където вероятно инвазията се е осъществила най-рано. Споменати са потенциалните заплахи от по-нататъшното разпространение на мидата в басейна на р. Искър.