Rapid Communication

First records of largemouth bass *Micropterus salmoides* (Lacépède, 1802) from Bulgaria (Balkan Peninsula)

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Abstract

The largemouth bass *Micropterus salmoides* (Lacépède, 1802), a North American fish from the family Centrarchidae, is reported for the first time from Bulgaria. In April–June 2018, adult largemouth bass (2+ years) were captured in two localities in the Struma River Basin (south-west Bulgaria). In May 2018, individuals from several schools of recently hatched *M. salmoides* were collected. The presence of multiple-age classes suggests that *M. salmoides* survives and reproduces naturally in Bulgarian freshwaters. The increasing number of reports of captured specimens along the Struma River suggests the species may be widespread. The largemouth bass was probably imported and dispersed in Bulgarian freshwater bodies by anglers without a prior risk assessment or authorization by the responsible institutions. In accordance with nature conservation efforts, to prevent further dispersal of the largemouth bass in Bulgaria effective management policies should urgently be developed and implemented by the appropriate authorities.

Key words: Pisces, non-indigenous species, Centrarchidae, Struma River

Introduction

Invasive species are one of the major factors for biodiversity loss, particularly in freshwater ecosystems (Clavero and García-Berthou 2005; Cox and Lima 2006). Over the last five decades, more than 20 fish species have been introduced into Bulgarian inland waters (Yankova 2016). With a few exceptions, all these fishes were imported intentionally to diversify aquaculture production, for ecological manipulation and bio-control of unwanted organisms, and/or to enhance commercial and recreational fisheries (Uzunova and Zlatanova 2007). However, most of these introductions were made without a prior risk assessment and, in some cases, even illegally. Several species have been released directly into the environment, while others have escaped from fish farms. Many introduced fishes remain unnoticed in the wild for long periods of time and their detection and registration is commonly accidental, often after negative consequences of invasion have occurred (Nehring 2005).

Largemouth bass, *Micropterus salmoides* (Lacépède, 1802), is a member of the family Centrarchidae and native to eastern North America from southern Canada to northern Mexico, and from the Atlantic coast to the central region of the United States (Scott and Crossman 1973; Warren 2009). Largemouth bass is one of the most popular and valuable freshwater sport-fish (Lasenby and Kerr 2000). Since the beginning of the 20th century, it has been introduced into more than 70 countries in South America, Africa, Europe, Asia, the Pacific islands, as well as areas within North America (e.g. Takamura 2007; Brown et al. 2009; Warren 2009; Leunda 2010; van Rensburg et al. 2011; Hargrove et al. 2017). Largemouth bass has broad ecological and habitat tolerance and inhabits a wide range of slow-flowing to still freshwater ecosystems and brackish waters with salinity up to 13‰ (Kottelat and Freyhoff 2007; Brown et al. 2009; Warren 2009). Largemouth bass is an opportunistic top predator, exploiting prey from the bottom to the surface in a given water body. Adults feed primarily on fishes, but also consume crayfish, amphibians and aquatic macroinvertebrates (Warren 2009). In both native and non-native environments, largemouth bass displays substantial trophic plasticity and can easily adjust its diet according to pray availability and abundance(Huskey and Turingan 2001; Almeida et al. 2012). As a result, *M. salmoides* can alter native fish assemblage structures by reducing the abundance and diversity of small-bodied fishes, modifying planktonic and benthic communities and changing food webs and habitat complexity (e.g. De Moor and Bruton 1988; Godinho and Ferreira 1998; García-Berthou and Moreno-Amich 2000; Gratwicke and Marshall 2001; MacRae and Jackson 2001; Azuma 2002; Jackson 2002; Takamura 2007; Shelton et al. 2008; Tweddle et al. 2009; Weyl et al. 2010; Trumpickas et al. 2011; Tsunoda and Mitsuo 2012; Hossain et al. 2013; Kimberg et al. 2014; Han et al. 2016). Ecological risk assessments revealed that *M. salmoides* is a high-risk invasive species for several European countries (e.g. Almeida et al. 2013; Perdikaris et al. 2016; Piria et al. 2016; Tarkan et al. 2017) and moderately high for others (Simonovic et al. 2013). Because of its negative impacts on native biodiversity, *M. salmoides* is considered one of the world's most malignant invasive alien species (Welcomme 1992; Lowe et al. 2000).

In the past two years, we have observed several captures of the largemouth bass in Bulgarian waters posted by recreational fishermen on Internet forums, a source that in many cases is found to be a faster method of detection for new invaders than regular monitoring programs (Kalous et al. 2018). However, these records are still unconfirmed and there remains no official report for this species distribution in Bulgaria. This study presents, for the first time, data confirming the presence of largemouth bass in Bulgaria. The possible implications of the establishment of self-sustaining populations of *M. salmoides* and mitigation measures are discussed.

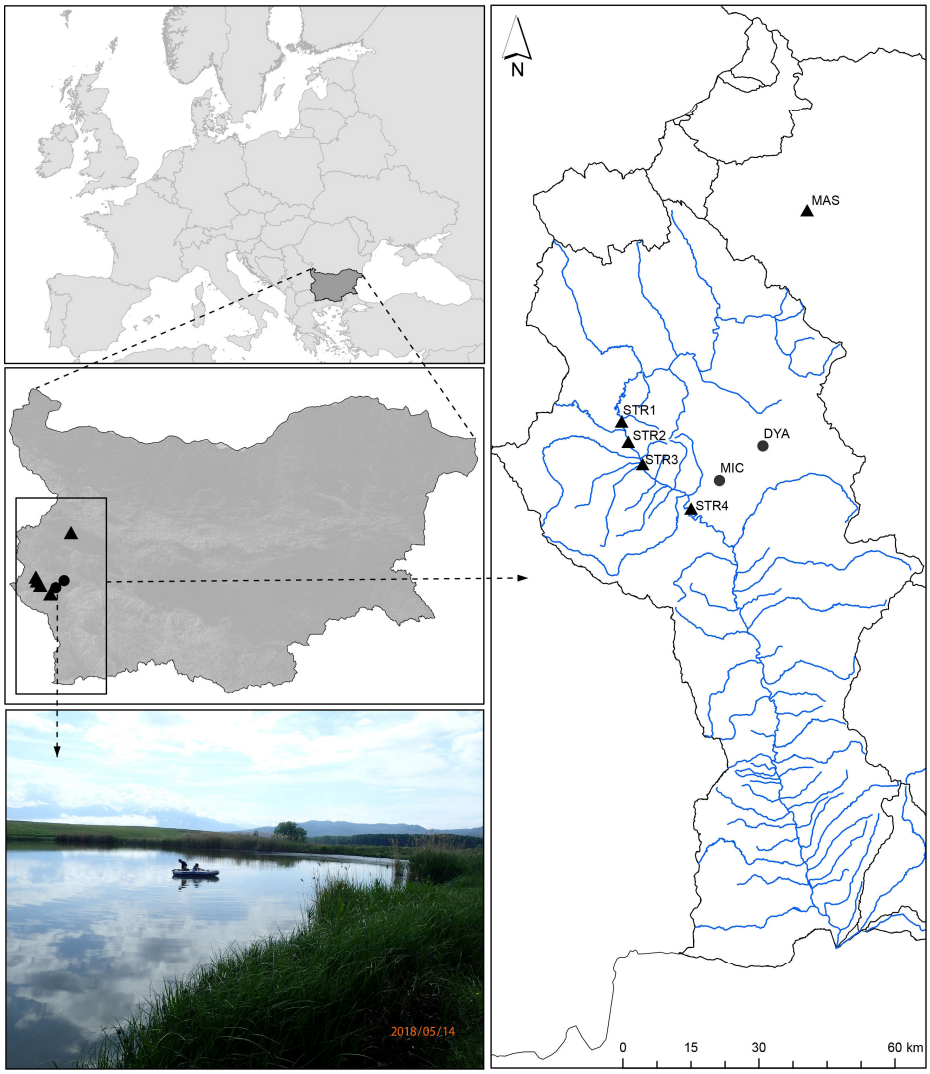

Figure 1. Map with the records of *Micropterus salmoides* in Bulgaria. Black circles indicate the locations of origin of the specimens stored in the GAH Department's fish collection. Black triangles show records provided by fisheries control authorities. The photo shows the shoreline of the Micro Reservoir 1 where small fish were caught. For the meaning of the site codes see Supplementary material Table S1. Photo by Eliza Uzunova.

Materials and methods

On the 29th of April 2018, one *M. salmoides* specimen was captured by fishing rod in the Dyakovo Reservoir (south-western Bulgaria; Figure 1, Supplementary material Table S1). The fish was caught by the second author, brought to the laboratory of the Department of General and Applied Hydrobiology (GAH), Faculty of Biology, Sofia University and deposited in the Department's fish collection (catalogue number GAH 1823551; Figure 2). Dyakovo Reservoir is located on several tributaries of the Struma River (Aegean Basin). The reservoir was built in 1975 for water supply purposes and has a maximum surface area of 2000 km^2 and maximum water volume of 35.400×10^6 m^3. On 14 May 2018, to verify information from Internet forums for sport fishermen, we conducted a survey in a reservoir with an area of 50 ha located in the same watershed

Figure 2. The specimen Cat. N GAH 1823551, *M. salmoides*, 0.34 m TL, 635 g captured in the Dyakovo Reservoir on April 29th 2018 (on the left); specimen GAH 1874557, 0.19 m TL, 120 g weight captured in Micro reservoir 1 on 20th June 2018 (in the middle); specimen GAH 1874553, 0.023 m TL, 0.18 g weight, captured in the Micro Reservoir 1 on May 14th 2018 (on the right). Photographs by Eliza Uzunova.

(Micro reservoir 1, Figure 1, Table S1). This water body is used for irrigation and recreational fishing. A significant proportion (about 60%) of its coastal area is covered by a reed belt with a width of 2–3 m. The bottom substrate is dominated by mud (Figure 1). Using a boat and pond nets (0.6 m diameter of the head and 4 mm mesh size), dozens of small *M. salmoides* were caught (GAH collection nos. 187452–56). On 20 June 2018, two adult specimens (GAH nos. 187457–58) were captured by fishing rod in the same water body. All captured fish were retained with no individuals being released alive, in accordance with nature conservation efforts to prevent alien species establishment. Fish were preserved in 4% formaldehyde with the exception of the first specimen from Dyakovo Reservoir (cat. N GAH 1823551) which is stored frozen at −20 °C.

Data on the records of *M. salmoides* caught by anglers in the Struma River were obtained from the Executive Agency of Fisheries and Aquaculture (EAFA) and the Executive Agency of Forestry (EAF). To verify this information, a survey via electrofishing was carried out at three sites along the Struma River (Figure 1, Table S1). A minimum of 150 m long transect was applied per site. The river sections were selected to include riffle and pool mesohabitats, the last being with a maximum depth of 1.2 m, formed downstream of river fragmentation structures (weirs, sluices). One run of electrofishing without block nets was conducted at each site (SAMUS-725G, DC, 30 cm diameter ring anode, average voltage 200–350 V, operating at an average of 3–8 A depending on water conductivity). Information for a *M. salmoides* caught in the Maslovo Reservoir (Iskar River Basin) was obtained from a representative of the municipality of Kostinbrod (Figure 1, Table S1).

Each adult largemouth bass and recently hatched individual was photographed with a Canon EOS Digital 350D camera and the photos were used for morphometric measurements (Digimizer Version 4.6.1 MedCalc Software). The measurement of the plastic and meristic characteristics was according to the scheme proposed by Yokogawa (2014). The identification keys of Scott and Crossman (1973) and Warren (2009) were used for

species determination. All specimens were weighed (W) with an accuracy of 0.1 g. We measured eviscerated weight (W*e* ± 0.1 g), and gonad weight (W*g* ± 0.1 g) for adult specimens. The gonado-somatic index (GSI) was calculated from the equation GSI % = 100 (W*g*/W*e*). The age of adult fish was determined using whole sagittal otoliths immersed in water and observed under a stereo microscope. Under reflected light, the alternation of opaque and hyaline zones formed one annulus.

Results and discussion

We collected largemouth bass from two sites and additional information for another five records was provided by representatives of the fisheries control agencies in Bulgaria (EAFA and EAF; Figure 1, Table S1). Data on fish catches are given in Table S1. The three adult specimens from Dyakovo and Micro 1 Reservoirs had elongated body with irregular bars forming a strip along the sides. Top of the head and back dark to light green; underside of head and belly lighter. Mouth large, terminal; upper and lower jaws extending past the back edge of the eye. Dorsal fin rays X+13; anal fin rays III+11; pectoral fin rays 13–14; pelvic fin rays I+5; lateral scales (62)61 to 63; rows above lateral line 7 to 8; rows below lateral line 13 to 14. Standard length 1.15 times total length; greatest depth 33% of standard length; preorbital length 3.3 times into head length. Notch between anterior and posterior dorsal fins deep (Figure 2, Table 1).

The morphological features and body coloration of the captured individuals led to the unequivocal identification of the fish as *M. salmoides*. The total body mass of all collected individuals ranged from 0.17 g to 635 g, and total body length ranged from 22 mm to 340 mm (Table 1). The average GSI for adult fish was 0.47%. The biggest specimen (GAH 1823551) was a male of 340 mm TL and body weight of 635 g (Figure 2).

The age of the specimens GAH 1823551, 1874557 and 187458 were estimated to be 2+ years. We suspect that differences in the diet are a likely cause for the pronounced size difference between fish at the same age from Dyakovo Reservoir and Micro Reservoir 1.

The presence of numerous shoals of small fish, guarded by males, was recorded in the coastal areas of Micro Reservoir 1 (personal observation). This gives evidence for natural reproduction and establishment of this species in Bulgarian waters.

Since the first catch in Dyakovo Reservoir, surveys were conducted among local and regional authorities responsible for fisheries control in Bulgaria. Results showed that in 2017 and 2018, *M. salmoides* were caught by anglers at three sites in the Struma River (Table S1). The presence of largemouth bass in the Struma River was not verified during this study. Electrofishing may not be the most appropriate method for catching largemouth bass in the pools formed downstream of the barrages (Ribeiro

Table 1. Morphometric measurements and meristic counts for the six specimens of *Micropterus salmoides* stored in the GAH Department's fish collection. N/A indicates that the scale counts for small fish were "not applicable".

Characters	*Micropterus salmoides* specimens					
	GAH 1823551	GAH 1874557	GAH 1874558	GAH 1874552	GAH 1874553	GAH 1874556
Total length, cm	33.97	19.17	18.51	2.49	2.41	2.36
Standard length, cm	29.74	16.61	16.09	2.15	2.05	2.07
In % of the standard length						
Pre-anus distance	66.49	61.15	63.30	59.82	60.58	56.57
Pre-dorsal fin length	44.36	42.25	40.63	44.29	44.81	44.06
Body depth	33.15	33.33	31.35	26.21	25.38	25.00
Caudal peduncle depth	13.09	14.04	13.26	10.95	11.11	11.82
Caudal peduncle length	23.77	24.77	25.80	26.07	24.39	25.14
First dorsal fin length	6.49	8.43	7.59	5.66	6.01	6.14
Second dorsal fin length	13.10	13.53	11.55	13.74	11.91	11.02
Length of anal fin	11.17	13.88	14.04	12.29	12.37	12.01
Length of pectoral fin	16.44	16.06	12.49	9.72	11.37	8.03
Length of pelvic fin	14.37	12.59	15.46	8.00	8.22	9.05
Postdorsal distance	21.65	24.35	23.28	27.78	25.94	26.85
Length of head	34.72	31.02	33.69	36.84	35.72	34.26
In % of the head length						
Preorbital length	27.93	25.10	27.74	27.18	27.28	28.83
Horizontal diameter eye	11.47	16.54	14.35	24.59	27.60	25.28
Postorbital length	61.91	60.57	59.40	48.51	47.07	47.32
Maxilla length	45.78	50.78	52.61	35.85	37.85	30.37
Mandibula length	58.54	59.00	56.72	44.89	37.28	36.21
Interorbital distance	24.20	27.46	25.83	23.87	15.98	27.01
Meristic characters						
Anal-fin spines and rays*	III 11	III 11	III 11	III 11	III 11	III 11
Dorsal-fin spines and soft rays*	X 13	X 13	X 13	X 13	X 13	X 13
Pectoral fin soft rays	14	13	13	13	13	13
Pelvic fin spines and soft rays	I 5	I 5	I 5	I 5	I 5	I 5
Pored scales on lateral line	61	63	62	N/A	N/A	N/A
Scales above lateral line	8	7	7	N/A	N/A	N/A
Scales below lateral line	14	14	13	N/A	N/A	N/A
Body weight [g]	635	120	118	0.22	0.18	0.18

* Roman numerals indicate spines, Arabic – soft rays

et al. 2015). It is possible that standing water bodies with self-sustaining populations, such as Dyakovo Reservoir and Micro Reservoir 1, serve as nursery ponds and facilitate the spread of *M. salmoides* in the adjacent river system (Ribeiro et al. 2009). In addition, river regulation by weirs favours invasion by providing both suitable food and habitat resources (Almeida et al. 2012).

We suspect that largemouth bass was introduced in Bulgaria for sport fishing and a major role in bass distribution is played by fishermen. However, this introduction was done without risk assessment analysis or authorization by the competent authorities. The species was probably imported from neighboring countries where it has already spread in nature. Largemouth bass is established in the wild in most Mediterranean countries (Kottelat and Freyhof 2007). According to an anonymous source, the species was imported into Bulgaria from Cyprus. Considering the age of the older fish and the existence of mature breeding fish, we estimated that *M. salmoides* was introduced into Bulgaria at least 3–5 years ago.

Naturalization of largemouth bass in Bulgaria raises the question of consequences for freshwater ecosystems, and which native organisms will be at risk. Previous research has shown that predation by introduced largemouth bass has resulted in decreased abundance of native fish, crustaceans and insects in Asia and Africa (e.g. Maezono and Miyashita 2003; Yonekura et al. 2004; Han et al. 2016). The impacts of invasive populations of *M. salmoides* appear to be greatest for small fish living in both lakes and rivers (Jackson 2002). In the Struma River, it is expected that several highly vulnerable endemic fish species such as *Oxynoemacheilus bureschi* (Drensky, 1928), *Cobitis strumicae* (Karaman, 1955), and *Barbus strumicae* (Karaman, 1955) will be most negatively affected by direct predation of *M. salmoides*. In addition, largemouth bass can become a food competitor for autochthonous predators such as pike (*Esox lucius* L.). A possible negative impact could be expected also for fish species listed in Annex 2 of the Council Directive 92/43/EEC on the Conservation of natural habitats and of wild fauna and flora, such as *Sabanejewia aurata* (De Filippi, 1863), *Rhodeus amarus* (Bloch, 1782), *Barbus plebejus* (Bonaparte, 1839), *Cobitis taenia* (Linnaeus, 1758), and *Aspius aspius* (Linnaeus, 1758). The Struma River runs through several protected areas with high conservation value included in the European Natura 2000 network (e.g. Zemen BG 0001012, Skrino BG 0001013, Kresna-Ilindentsi BG 0000366), as well as Lake Kerkini (Greece), where the presence of these species has been recorded (Vassilev and Pehlivanov 2002). Furthermore, *M. salmoides* is not the only exotic Centrarchid fish that has invaded the Bulgarian freshwater ecosystems. The pumpkinseed *Lepomis gibbosus* (L.) has established abundant populations in various types of water bodies throughout the country (Uzunova et al. 2012). The pumpkinseed forms a significant share in the diet of largemouth bass (> 200 mm); it can be assumed that *M. salmoides* will find a suitable nutritional base in those water bodies where the abundance of the pumpkinseed is high (Godinho and Ferreira 1998; Almeida et al. 2012).

Detection of alien species in the early stages of an invasion process and application of mitigation measures is crucial for their future impacts (Copp et al. 2005). It is now essential, in accordance with nature conservation efforts, to develop and implement effective management policies with the appropriate authorities in order to stop the dispersal of the invasive *M. salmoides* in Bulgaria. Therefore, the Ministry of Agriculture, Food and Forestry and the Executive Agency of Fishery and Aquaculture were immediately notified and emergency measures to prevent *M. salmoides* (re)introductions or further natural spread were recommended. Information bulletins and posters containing a description of the fish and expected consequences of its invasion will be sent to all regional directorates of the EAFA with the aim to disseminate information to fishpond owners, sport

fishermen, fishing clubs and societies and local populations regarding the potential spread, impacts and biosecurity issues associated with *M. salmoides*. A newsletter will be distributed among anglers and pet shops containing strong exhortation not to use this species as bait or aquarium fish. Strict control on restocking activities and prevention of fish escapes from farm facilities are also urgent measures that should be implemented. An analysis of the parasitic fauna for the possible transfer of parasites along with the imported specimens is forthcoming. Water bodies that could potentially provide suitable habitat conditions for *M. salmoides* should be examined periodically according EU Directives. As a precautionary measure, it is suggested that keeping or breeding of *M. salmoides* outside of RAS systems should be prohibited in Bulgaria.

We also emphasize that implementation of measurements for the control of *M. salmoides* distribution, where necessary and possible, is likely to be the most cost effective strategy. Otherwise, it will be only a matter of time before this invasive alien species spreads throughout the country.

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Supplementary material

The following supplementary material is available for this article:

Table S1. Records of *Micropterus salmoides* in Bulgaria.

This material is available as part of online article from:
http://www.reabic.net/journals/bir/2019/Supplements/BIR_2019_Uzunova_etal_Table_S1.xlsx