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BRIEF COMMUNICATION

Infections with Digenean Trematode Metacercariae in Two Invasive Alien Fish, *Micropterus salmoides* and *Lepomis macrochirus*, in Two Rivers in Chungcheongbuk-do, Republic of Korea

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Abstract: Present study was performed to survey infection status of digenetic trematode metacercariae in 2 alien fish species, *Micropterus salmoides* (largemouth bass) and *Lepomis macrochirus* (bluegill), in 2 rivers draining Chungcheongbuk-do, Republic of Korea. A total of 107 largemouth bass and 244 bluegills were caught in Daecheong-ho (ho=lake) and Musim-cheon (a branch of Geum-gang), in Chungcheongbuk-do April-July 2015. Additionally, 68 native fish of 5 species, i.e., *Zacco platypus, Hemibarbus longirostris, Carassius auratus, Pseudogobio esocinus* and *Puntungia herzi*, were caught from the same water bodies. All of the fish collected were examined by artificial digestion method. The metacercariae of *Centrocestus armatus, Clinostomum complanatum, Metagonimus* sp. and *Diplostomum* spp. were detected from 4 out of 5 native fish species in Daecheong-ho. However, any metacercariae were not found from 87 *M. salmoides* and 177 *L. macrochirus* in Daecheong-ho. In Musim-cheon, metacercariae of *Exorchis oviformis* and *Metacercaria hasegawai* were detected from 78% *Z. platypus* and 34% *L. macrochirus*, but any metacercariae not found in *M. salmoides*. We report here that the 2 alien fish species were less infected with the metacercariae than the native ones. Surveys on the metacercariae in the alien fish species in geographically various rivers should be undertaken for better understanding on the role of alien fish species in the trematode infections in Republic of Korea.

Key words: Metacercariae, *Micropterus salmoides*, *Lepomis macrochirus*, alien fish, Geum river

Freshwater fish have been recognized as the main source of trematode infections in the Republic of Korea. Although other kinds of factors can also act as an infection source of zoonotic parasites, freshwater fish are considered comparatively more important than other factors [1]. Thus the prevalence of fish-borne zoonotic helminthes has been endemically maintained in Korea whereas that of soil-transmitted helminthes has been dramatically decreased [2-4]. A recent survey showed a higher prevalence of fish-born trematodes, i.e., *Clonorchis sinensis* and *Metagonimus* spp. (8.4% and 1.0%, respectively), than soil-transmitted helminthes such as *Ascaris lumbricoides* and *Trichuris trichiura* (0.01% and 0.1%, respectively) in residents along 5 major rivers in Korea [5]. On the other hand, many Korean researchers have surveyed the infection status of digenetic trematode metacercariae (DTM) in freshwater fishes from various localities to evaluate the endemicity of subjected trematodes in the surveyed areas [6-12]. Especially, lots of studies were conducted for one of the most important zoonotic parasites, *C. sinensis* in Korea. Kim et al. [6]. investigated 34 localities including the Geum-gang (gang=river) basin, which was similar region with the present study for understanding the infection status of *C. sinensis* metacercariae (CsMc) using 21 species of fishes. Connectively, Cho et al. [7] surveyed 3 latitudinal regions of Korean Peninsula in 2008, and later Cho et al. [8], also investigated the infection status of DTM in 32 freshwater fish species (n=865) from Gangwon-do (do=province). Additionally, Sohn et al. [9-11] performed some epidemiological works on the infection status of DTM in freshwater fish from some rivers, i.e., Imjin-gang, Hantan-gang, Seomgjin-gang, Wi-cheon (cheon=stream) and Tamjin-gang [12] in Korea. Although many studies focused on native freshwater fishes, parasites of invasive alien species are poorly known in Ko-

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rea. Two invasive alien fish species, namely, the largemouth bass *Micropterus salmoides* and the bluegill *Lepomis macrochirus*, were introduced to Korea from the U.S.A. as a food resource and for economic reasons. However, the business failed and they were released or escaped from fish farms. The alien fishes are now distributed widely throughout the Korean ecosystem due to their rapid growth, high reproduction rate, predatory instinct against native species, and the almost complete absence of an enemy [13-15]. They currently seem to have adapted successfully to the Korean environment and are occasionally eaten by humans in addition to the native fishes. It appears that parasitic helminthes of alien fish species are poorly investigated until present study in Korea. The aims of the present study were to investigate the parasitic helminthes of 2 invasive alien fish species in Korea, intensively, and to compare their infection status with that of native fishes.

A total of 107 largemouth bass (155-477 mm in length and 65-1,360 g in weight) and 244 bluegills (80-180 mm in length and 16-71 g in weight) were purchased from a local fisherman and/or caught by the lure-fishing in the upper stream region of Daecheong-ho (ho = lake) in Hoenam-myeon (myeon = township), Boeun-gun, and the upper stream region of Musim-cheon in Jangam-dong (dong = village), Cheongju-si (si = city), Chungcheongbuk-do (do = province) from April to July 2015. Additionally, total 68 native fishes in 5 species (n = 69), i.e., *Zacco platypus* (49), *Hemibarbus longirostris* (5), *Carassius auratus* (8), *Pseudogobio esocinus* (3) and *Puntungia herzi* (3), were also collected in the same regions (Table 1). The fishes were measured and then careful observations were made of the body surface and fins with the naked eye to find large-sized metacercariae. Eye balls were separated from the body and observed under a stereomicroscope for detecting diplostomid metacercariae. All body contents including gills and internal organs were then blended and digested by an artificial digestion method using pepsin and HCl [1] for the detection of metacercariae and other larval parasites. These procedures were conducted individually on each fish. The identifications of DTM detected were done with the aid of the differential keys in Sohn [1].

Despite the small numbers of native fishes examined, 4 species of trematode metacercariae, namely, *Centrocestus armatus*, *Clinostomum complanatum*, *Metagonimus* sp. and diplostomula, were detected from 4 native fish species, *Zacco platypus*, *Hemibarbus longirostris*, *Carassius auratus* and *Pseudogobio esocinus*, in the Daecheong-ho region. Metacercariae of *C. armatus* were only found from *Z. platypus* with a prevalence as high as 87% (27 of 31 fishes). Diplostomula was detected from the eye balls of 8 *Z. platypus* (26%), 5 *H. longirostris* (100%), 4 *C. auratus* (50%) and 1 *P. esocinus* (33%). Metacercariae of *C. complanatum* were found from 5 *Z. platypus* (16%) and a *C. auratus* (12.5%). Metacercariae of *Metagonimus* spp. were found only from a *Z. platypus* (3%). On the other hand, the 2 species of alien fishes examined from the same region were all negative

Table 1. Infection status with digenetic trematode metacercariae (MC) in fishes from 2 different regions in Chungcheongbuk-do, Korea

Locality	Species of fish	No. examined	No. (%) infected	No. (%) of MC detected	Range	M ± SD
Daecheong-ho (lake)	Native species					
	Zacco platypus	31	28 (90)	*Centrocestus* spp. 27 (87)	7-432	133 ± 104
				Clinostomum spp. 5 (16)	1-15	5 ± 6
				Diplostomula 8 (26)	2-32	10 ± 10
				Metagonimus spp. 1 (3)	32	32
	Puntungia herzi	3	0 (0)	-	-	-
	Hemibarbus logirostris	5	5 (100)	Diplostomula 5 (100)	1-17	6 ± 7
	Carassius auaratus	8	4 (50)	*Clinostomum* spp. 1 (12.5)	3	3
				Diplostomula 4 (50)	3-5	4 ± 1
	Pseudogobio esocinus	3	1 (33)	Diplostomula 1 (33)	5	5
	Alien species					
	Micropterus salmoides	87	0 (0)	-	-	-
	Lapomis macrochirus	177	0 (0)	-	-	-
Musim-cheon (stream)	Native species					
	Zacco platypus	18	14 (78)	*Exorchis oviformis* 9 (50)	3-107	46 ± 38
				Metacercaria hasegawai 7 (39)	2-88	17 ± 31
	Alien species					
	Micropterus salmoides	20	0 (0)	-	-	-
	Lapomis macrochirus	67	23 (34)	*Exorchis oviformis* 22 (33)	3-22	8 ± 5
				Metacercaria hasegawai 7 (10)	1-10	6 ± 3

for metacercariae infection. Only 2 species of digenetic trematode metacercariae were discovered from fishes obtained from the Musim-cheon. *Exorchis oviformis* metacercariae and *Metacercaria hasegawai* were found from *Z. platypus* (9 and 7 fishes, respectively). Bluegills were infected with both trematode species (Fig. 1). The infection status of alien and native fishes in the present study is detailed in Table 1.

Although the 2 species of alien fishes had been investigated for DTM before, it was sporadic survey which a part of large scaled survey on all kinds of freshwater fish investigation. Sohn et al. [11] examined total 10 *M. salmoides* and 4 *L. macrochirus* from Wicheon-stream in Gunwi-gun (gun = county), Gyeongsangbuk-do, and they found total 6 CsMc from 2 largemouth bass. More recently, Yoon et al. [12] investigated 8

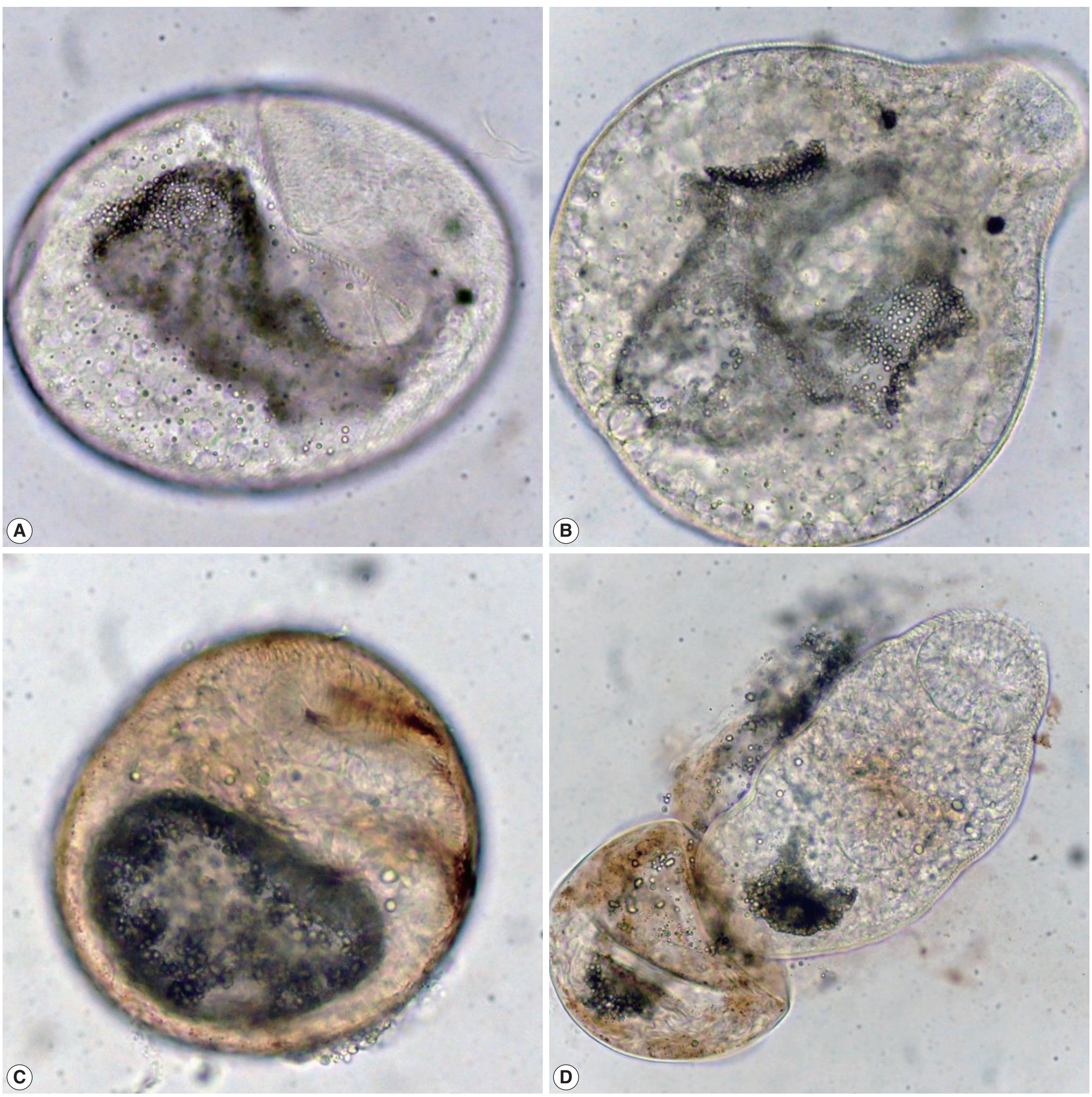

Fig. 1. *Exorchis oviformis* metacercariae (A, B) and *Metacaercaria hasegawai* (C, D) collected from the bluegill *Lepomis macrochirus*. (A, B) Metacercariae of *E. oviformis* which had V-shaped excretory bladder with a pair of eye spots. (C, D) *Metacercaria hasegawai*, the excretory bladder located in posterior body. (A, C) Encysted larvae. (B, D) Excysted larvae.

largemouth bass but they didn't find CsMc from the fishes. A bluegill was examined by Sohn et al. [10] from Seomjin-gang, but it was negative for CsMC. In the present study, alien fishes were poorly infected with metazoan parasites compared to native fishes. Although we found over 6 species of metacercariae from native fishes, results for largemouth bass were all negative. Only 2 native parasites in Korea, *E. oviformis* and *Metacercaria hasegawai*, were found in the alien species bluegill, with a prevalence of 33% and 10%, respectively. We did not find any alien helminthes in both native and alien fishes.

Differences in the prevalence of parasitic infection between native and alien populations have been reported for other regions of the world. Dove [16] reported the poor parasitic prevalence of alien freshwater species in eastern Australia, and most of the shared parasitic species originated from native fish species. Lymbery et al. [17] also described the reduced prevalence of parasitic infection in alien species in Australia. This phenomenon is not only limited to freshwater fishes, but is also present in a wide range of alien species including mammals, birds and invertebrates [18-21]. In our case, we observed a poor parasitic infection in alien fishes compared with that of native populations. Even though the numbers of native fish examined were heavily outnumbered by the alien fishes (50 vs 351), the infection rates were much higher in native fish than in alien fishes (76.5 vs 6.6%).

Unlike the infection status in the present study, both alien fish species in their original habitat have exhibited parasitic infections with a relatively rich parasitic fauna. Largemouth bass in their native habitat are infected with metazoan parasites belonging to a number of phyla [22-25], and helminth parasites of bluegills have also been investigated [26-28]. It has been known that 2 fish species in their original habitat are infected with several species of digenetic trematode metacercariae including *Clinostomum complanatum*, *C. marginatum*, *Diplostomum* sp., *Neascus* sp., *Posthodiplostomum minimum*, strigeids and *Uvulifer ambloplitis*. However, we did not find any alien parasite species that might be introduced with alien fishes. In fact, we did not find any metazoan parasite species with the exception of an anchor worm species in bluegill (unpublished data).

Lower parasitic prevalence of alien species and an absence of their own parasites may result from several reasons, such as host specificity, life-cycle stages, trophic categories, and methods of introduction [19]. Parasites like trematodes need 2 or more host species to maintain their life-cycle. If the introduced parasites fail to find suitable hosts, they will not survive to successfully invade the ecosystem [18,19]. In particular, it may be more difficult to survive for parasites and to find their way as metacercariae in alien fishes. They have to overcome several barriers for a successful invasion. Among the introduced populations, infected fish has to be chosen as a suitable host for successful growth to the adult stage in the host. Although they may overcome the initial progress, they still have to pass the steps concerning adaptation to the environment and the first intermediate hosts.

Consequently, we could not find any helminth parasites from largemouth bass, and only 2 non-zoonotic parasites, *E. oviformis* and *Metacercaria hasegawai*, were found from bluegills. As with previous studies in other regions of the world, the 2 alien fishes of this study were poorly infected with parasites. However, this study did not cover the whole country and every ecosystem in Korea, and was limited to 2 restricted areas. In addition, we did not find the most important DTM in Korea, *C. sinensis*, in either native or alien fishes. These results may therefore not ensure the food safety of raw fish consumption in the regions, and thus additional surveys are needed on alien fish populations distributed widely in Korea.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest with this article.

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