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▣ BRIEF COMMUNICATION

# Infection Status of *Isthmiophora hortensis* Metacercariae in Dark Sleepers, *Odontobutis* Species, from Some Water Systems of the Republic of Korea

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**Abstract:** Present study was performed to survey on infection status of *Isthmiophora hortensis* (formerly *Echinostoma hortense*) metacercariae (IhMc) in dark sleepers, *Odontobutis* spp., from some water systems of the Republic of Korea. A total of 237 *Odontobutis* spp. was collected in the water systems of 5 rivers, i.e., Mangyeong-gang (gang means river), Geum-gang, Tamjin-gang, Seomjin-gang, and Nakdong-gang. They were all examined with artificial digestion method for 5 years (2013-2017). A total of 137 (57.8%) *Odontobutis* spp. were infected with 14.8 IhMc in average. The prevalence was the highest in Nakdong-gang areas (62.9%) and followed by in Mangyeong-gang (57.1%), Geum-gang (56.3%), Tamjin-gang (54.8%), and Seomjin-gang (53.9%) areas. Metacercarial densities were 28.1 (Geum-gang), 13.9 (Mangyeong-gang), 13.3 (Nakdong-gang), 13.1 (Tamjin-gang), and 2.3 (Seomjin-gang) per infected fish. Especially, in case of Yugucheon (cheon means stream), a branch of Geum-gang, IhMc were detected in all fish (100%) examined and their density was about 48 per fish. By the present study, it was confirmed that the infection status of IhMc is more or less different by the surveyed areas and the dark sleepers, *Odontobutis* spp., are suitable fish hosts of *I. hortensis*.

Key words: *Isthmiophora hortensis*, dark sleeper, *Odontobutis* spp., metacercaria

*Isthmiophora hortensis* (Digenea: Echinostomatidae) was redescribed by Kostadinova and Gibson [1] with a new combination from originally described as *Echinostoma hortense* in Japan [2]. Kostadinova and Gibson [1] clarified the validity of genus *Isthmiophora*, which had long been in the obscure taxonomic position in relation to the genus *Euparyphium* and additionally made a differential key to the species of *Isthmiophora* with the morphological characteristics of related worm samples. To support the taxonomic validity of this fluke, Sohn et al. [3] made the differential indices with variety of worm samples from the Republic of Korea (Korea). Since the original description from rats in Japan, this species of echinostome has been discovered in rats, dogs, cats, weasels, raccoons, striped field mice, a raccoon dog and a wild boar, which were naturally infected in Korea, Japan, China and Vietnam [3-12]. Human infections with this fluke were also reported in Japan, Korea and China [13-19]. Especially in Korea, this echinostome is a dominant species with clinical importance [20-28].

Several species of freshwater fishes, i.e., *Misgurnus anguillicaudatus, Misgurnus mizolepis, Odontobutis obscura interrupta* (*O. interrupta*), *Moroco oxycephalus* (*Rhynchocypris oxycephalus*), *Coreoperca kawamebari* and *Squalidus coreanus* (*S. japonicus coreanus*), yellowfin goby, *Acanthogobius flavimanus*, and tadpoles of *Rana nigromaculata* were reported as the second intermediate hosts and/or the source of human infection in Korea. Most of these reports were performed in limited areas and with small number of animals [29-35]. Therefore, we examined the dark sleepers, *Odontobutis* spp., broadly collected from 13 sites in 5 rivers, i.e., Mangyeong-gang, Geum-gang, Tamjin-gang, Seomjin-gang and Nakdong-gang, of Korea.

Total 237 *Odontobutis* spp. were collected from 13 sites in 5 rivers, i.e., Mangyeong-gang (n=49), Geum-gang (n=32), Tamjin-gang (n=73), Seomjin-gang (n=13) and Nakdong-gang (n=70), and examined all with the artificial digestion method for 5 years (2013-2017) (Fig. 1). The number of fish examined by the surveyed sites was detailedly revealed in Table 1.

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The metacercariae of *I. hortensis* (IhMc) were globular or elliptical, 143-165 (154) by 128-158 (144) μm in size, and have a double layered cyst wall, 27 collar spines including 4 end group ones in each side of the head crown, a ventral sucker transversely elliptical, 2-fold as larger as the oral sucker, and granules in the excretory bladder arranged in 2 rows of tube (Fig. 2).


**Fig. 1.** Two dark sleepers, *Odontobutis interrupta*, from Yugucheon in Gongju-si, Chungcheongnam-do, and Wicheon in Gunwi-gun, Gyeongsangbuk-do. The second intermediate host of *I. hortensis* identified in this study.

A total of 137 (57.8%) *Odontobutis* spp. were infected with 14.8 IhMc in average. The prevalence was most high in Nakdong-gang areas (62.9%) and followed by it in Mangyeong-gang (57.1%), Geum-gang (56.3%), Tamjin-gang (54.8%) and Seomjin-gang (53.9%) areas. Metacercarial densities were 28.1 in Geum-gang, 13.9 in Mangyeong-gang, 13.3 in Nakdong-gang, 13.1 in Tamjin-gang and 2.3 per fish infected in Seomjin-gang respectively. Especially, in case of Yugucheon, a branch of Geumgang, IhMc were detected in all fish (100%) examined and their density was about 48 per fish. The infection status with IhMc in fish from 13 surveyed sites was detailedly designated in Table 1.

In the present study, it was confirmed that 2 species of dark sleepers, *O. platycephala*, and *O. interrupta*, are suitable fish hosts of *I. hortensis* although the infection status of IhMc is more or less different by the surveyed areas. Moreover, the prevalences and metacercarial densities of this study were much higher than those of previous studies. Ahn et al. [30] detected total 32 IhMc in 10 (27.8%) out of 36 dark sleepers, *O. interrupta*, from Seom-gang, a branch of Namhan-gang, in

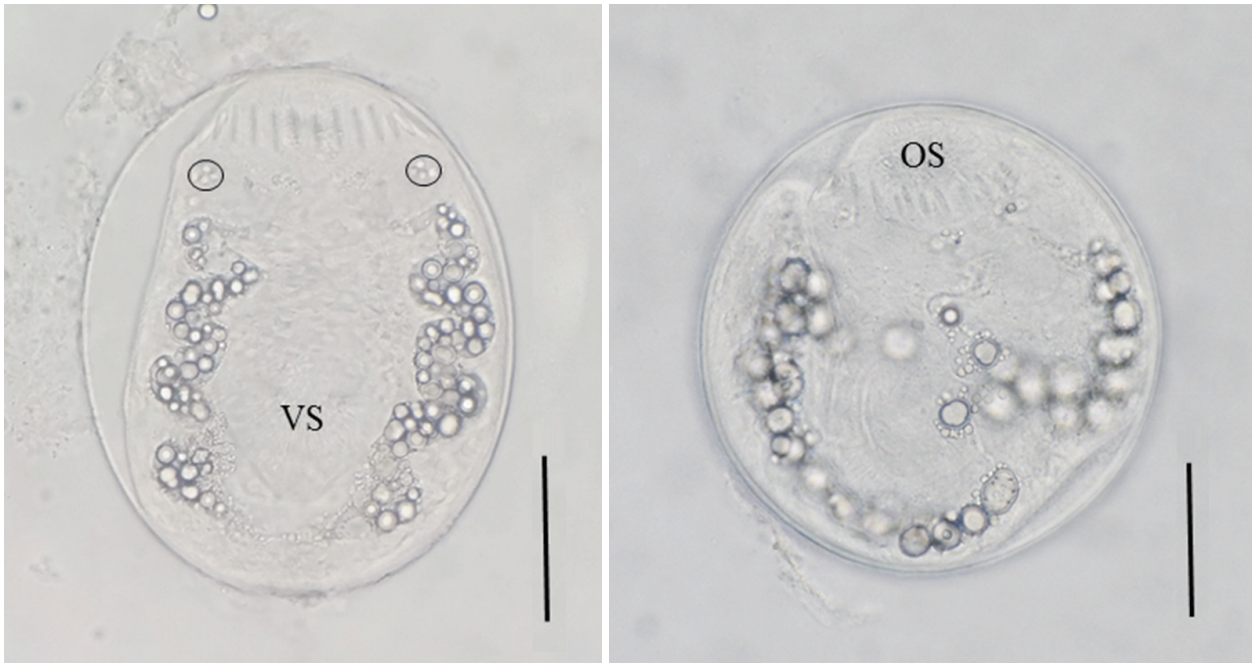


**Fig. 2.** Two metacercariae of *I. hortensis* collected from Korean dark sleeper, *O. platycephala*. They characteristically have an oral sucker (OS), ventral sucker (VS), 27 collar spines including 4 end group ones (encircled) in each side of the head crown and granules in the excretory bladder arranged in 2 rows of tube. Scale bar is 50 μm.

**Table 1.** Infection status of *Isthmiophora hortensis* metacercariae in dark sleepers, *Odontobutis* spp., from some water systems of Korea

| Locality surveyed | No. of fish examined | No. of fish infected | No. of metacercariae detected | |
|---|---|---|---|---|
| | | | Range | Average |
| Soyangcheon[a] | 49 | 28 (57.1) | 1-54 | 13.9 |
| Geum-gang | 32 | 18 (56.3) | 1-123 | 28.1 |
| Cheonnae-gang (Geumsan) | 22 | 8 (36.4) | 1-13 | 3.5 |
| Yugucheon (Gongju) | 10 | 10 (100) | 5-123 | 47.7 |
| Tamjin-gang | 73 | 40 (54.8) | 1-132 | 13.1 |
| Middle reaches (Jangheung) | 49 | 23 (46.9) | 1-132 | 20.3 |
| Lower reaches (Gangjin) | 24 | 17 (70.8) | 1-7 | 3.3 |
| Seomjin-gang | 13 | 7 (53.9) | 1-4 | 2.3 |
| Seomjin-gang (Sunchang) | 9 | 6 (66.7) | 1-4 | 2.3 |
| Seomjin-gang (Gurye) | 4 | 1 (25.0) | - | 2.0 |
| Nakdong-gang | 70 | 44 (62.9) | 1-95 | 13.3 |
| Wicheon (Gunwi) | 11 | 3 (27.3) | 2-29 | 12.0 |
| Banbyeoncheon (Yeongyang) | 4 | 3 (75.0) | 2-21 | 8.7 |
| Yangcheon (Sancheong) | 22 | 8 (36.4) | 1-12 | 3.6 |
| Taehwa-gang (Ulju)[b] | 9 | 7 (77.8) | 1-91 | 29.4 |
| Hyeongsan-gang (Gyeongju) | 21 | 20 (95.2) | 1-95 | 12.9 |
| Osipcheon (Yeongdeok) | 3 | 3 (100) | 3-25 | 10.7 |
| Total | 237 | 137 | 1-132 | 14.8 |

[a]A branch of Mangyeonggang located in Wanju-gun, Jeollabuk-do.
[b]6 fish collected from Cheokgwacheon (a branch of Taehwagang) located in Ulju-gun, Ulsan Metropolitan City.

Wonseong-gun (gun = county), Gangwon-do (do = province). Ahn and Ryang [22] also detected IhMc in 4 (22.2%) out of 18 dark sleepers from Namhan-gang. Lee et al. [23] found IhMc in only 1 (2.3%) out of 44 dark sleepers from Cheongsong-gun, Gyeongsangbuk-do. Ryang [20] detected IhMc in 11 (20.3%) out of 54 dark sleepers from Chungju-ho (ho means lake) and the upper streams of Namhan-gang. In case of suitable fish host, muddy loach (*Misgurnus anguillicaudatus*), Chai et al. [29] reported that total 64 (41.6%) out of 154 loaches were infected with 1-29 (8.1 in average) IhMc per fish infected. Ahn and Ryang [22] detected IhMc in 106 (34.1%) out of 311 loaches from Namhan-gang. Lee et al. [23] found IhMc in 2 (10.5%) out of 19 loaches from Cheongsong-gun, Gyeongsangbuk-do. Ryang [20] detected IhMc in 30 (40.5%) out of 74 loaches from Chungju-ho and the upper streams of Namhan-gang. In another fish hosts of *I. hortensis*, *M. mizolepis*, *R. oxycephalus*, *C. kawamebari*, *S. japonicus coreanus* and *A. flavimanus*, small number of IhMc were detected in Korea [29-35].

Some endemic foci of *I. hortensis* infections have been reported in Korea [20-23]. Especially in the villages of Cheongsong-gun in Gyeongsangbuk-do, 59 (22.4%) out of 263 residents were positive with echinostome eggs and 1-649 (51 in average) adult worms of *I. hortensis* were collected from 35 egg-positive cases in the worm recovery after praziquantel treatment and $MgSO_4$ purgation. As the source of human infections, the dark sleeper is most convincing among the second intermediate hosts in these endemic areas. It has been known that residents in inland of Korea frequently eat raw flesh of freshwater fishes such as Mandarin fish (*Siniperca scherzeri*), Korean aucha perch (*Coreoperca herzi*) and dark sleepers (*O. platycephala* and *O. interrupta*) [20-23,30]. These fishes are powerful predators in the ecosystems of freshwater and presently eaten in raw by residents living in riverside areas of Korea. Therefore, Korean peoples pay attention to the raw consumption of dark sleepers, *Odontobutis* spp., to prevent the infections with this species of echinostomid fluke.

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## CONFLICT OF INTEREST

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

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