Pygidiopsis summa (Digenea: Heterophyidae): Status of Metacercarial Infection in Mullets from Coastal Areas in the Republic of Korea

Woon-Mok Sohn1,4, Byoung-Kuk Na1, Shin-Hyeong Cho2, Won-Ja Lee2, Mi-Yeoun Park2, Soon-Won Lee3, Seung-Bong Choi3, Beom-Nyung Huh3, Won-Seok Seok3

1Department of Parasitology and Tropical Medicine, and Institute of Health Sciences, Gyeongsang National University School of Medicine, Jinju 52828, Korea; 2Division of Malaria and Parasitic Diseases, National Research Institute of Health, Centers for Disease Control and Prevention, Osong 28159, Korea, 3Infectious Disease Intelligence Division, Gangwon Institute of Health and Environment, Chuncheon 24203, Korea

Abstract: To know the infection status of zoonotic trematode metacercariae in brackish water fish, we surveyed mullets collected from 18 coastal areas in the Republic of Korea. The metacercariae of Pygidiopsis summa were detected in 236 (68.2%) out of 346 mullets examined. They were found in mullets from 15 areas except for those from Boseong-gun (Jeollanam-do), Pohang-si, and Uijin-gun (Gyeongsangbuk-do). Especially in mullets from Taean-gun (Chungcheongnam-do) and Geoje-si (Gyeongsangnam-do), their prevalences were 100% and 95.5%, and the average metacercarial density was more than 1,000 per fish. They were also detected in mullets from 3 coastal lakes, Gyeongpohoe, Songjiho, and Hwajinpo-ho, in Gangwon-do, and their average densities were 419, 147, and 672 per infected fish, respectively. The metacercariae of 5 other heterophyid species, including Heterophyes nocens, Heterophyopsis continua, Metagonimus sp., Stictodora fuscata, and Stictodora lari, were found in the mullets examined. The metacercariae of H. nocens were detected in 66.7%, 100, 28.6, 81.6, 3.9, 61.5, and 27.3% of mullets from Muan-gun, Shinan-gun, Haenam-gun, Gangjin-gun, and Boseong-gun (Jeollanam-do), Hadong-gun, and Geoje-si (Gyeongsangnam-do), and their metacercarial intensities were 64, 84, 119, 99, 1, 24, and 24 per fish infected, respectively. From the above results, it has been confirmed that P. summa metacercariae are heavily infected in mullets from coastal areas of Korea. It is suggested that residents who frequently consume raw mullet dish can be easily infected with heterophyid flukes.

Key words: Pygidiopsis summa, heterophyid fluke, infection status, metacercaria, mullet, coastal area

INTRODUCTION

Pygidiopsis summa (Digenea: Heterophyidae) is a very small intestinal trematode. This fluke was first found from dogs experimentally fed the metacercariae from mullets, Mugil cephalus, in Japan [1]. Human infections with this fluke were confirmed by detection of eggs in the feces and also adult flukes in Japan [2,3]. In the Republic of Korea (Korea), human cases have been sporadically reported from western and southern coastal areas, i.e., Okgu-gun and Buan-gun (Jeollabuk-do), Shinan-gun, Muan-gun, Gangjin-gun, Haenam-gun, and Yeoungam-gun (Jeollanam-do) [4-11]. Some species of brackish water fish, namely, M. cephalus, Liza haematocheila, and Acanthogobius flavimimus, are known to be the source of human infections and second intermediate hosts of this fluke [12-17].

A total of 11 species in 8 genera have been reported as zoonotic heterophyid flukes (ZHF) in Korea. Among these, 7 including P. summa are contracted by consumption of raw flesh of brackish water fish, including M. cephalus, L. haematocheila, A. flavimimus, Lateolabrax japonicus, Konesirius punctatus, Bolophthalmus pectinirostris, and Scartelaos sp., in Korea [12]. Studies on the source of human infections with ZHF were performed mainly in western and southern coastal areas of Korea. Most of the surveys were carried out in limited areas and examined a small number of fish hosts [13-18]. Especially, Seo et al. [14] surveyed total 59 mullets (1-14; av. 4.2 per locality) from 14 localities, and Guk et al. [17] examined a total of 139 mullets (5-20; av. 12.6) from 11 areas. However, the number of examined fish was not enough for evaluation of the metacercarial endemicity in each surveyed area. Therefore, we examined
mullets broadly collected from 18 coastal areas in 3 Korean seas, Yellow Sea, South Sea, and East Sea, to know the infection status of zoonotic trematode metacercariae.

MATERIALS AND METHODS

Collection sites of mullets

The collection sites of mullets were as follows: 1. Taean-gun, Gyeongsangnam-do: Byeongsulman in Ahnmyeon-eup (latitude: 36.478867; longitude: 126.342750); 2. Gochang-gun, Jeollabuk-do: Gusipo in Sangha-myeon (35.429330; 126.448270); 3. Muan-gun, Jeollanam-do: Haeje-myeon (35.092247; 126.304052); 4. Shinan-gun, Jeollanam-do: Aphae-eup (34.841932; 126.363447); 5. Haenam-gun, Jeollanam-do: Hwansan-myeon (34.596476; 126.460790); 6. Gangjin-gun, Jeollanam-do: Daegu-myeon (34.500880; 126.792037); 7. Boseong-gun, Jeollanam-do: Beolgyo-eup (34.830030; 127.370214); 8. Hadong-gun, Gyeongsangnam-do: Jeygo-myeon (35.009910; 127.918501); 9. Sacheon-si, Gyeongsangnam-do: Gonyangcheon in Gonyang-myeon (35.049422; 127.975090); 10. Goseong-gun, Gyeongsangnam-do: Baedun-myeon (35.048684; 128.372615); 11. Geoje-si, Gyeongsangnam-do: Dunduksamcheon in Dundekmam-do (34.935774; 128.508674); 12. Ulsan Metropolitan City: Taehwagang in Jung-gu (35.547438; 129.297372); 13. Pohang-si, Gyeongsangbuk-do: Hyeongsang-dong: Hyeongsaengang in Yeonil-eup (36.003818; 129.332359); 14. Yeongdeok-gun, Gyeongsangbuk-do: Osipcheon in Ganggu-myeon (36.379421; 129.376851); 15. Uljin-gun, Gyeongsangbuk-do: Wanguicheon in Uljin-eup (36.965826; 129.394991); 16. Gangneung-si, Gangwon-do: Gyepungpo (ho means lake) in

**Fig. 1.** Surveyed areas in South Korea. 1. Taean-gun, Chungcheongnam-do (CN); 2. Gochang-gun, Jeollabuk-do (JB); 3. Muan-gun; 4. Shinan-gun; 5. Haenam-gun; 6. Gangjin-gun; 7. Boseong-gun, Jeollanam-do (JN); 8. Hadong-gun; 9. Sacheon-si; 10. Goseong-gun; 11. Geoje-si, Gyeongsangnam-do (GN); 12. Ulsan Metropolitan City (U); 13. Pohang-si; 14. Yeongdeok-gun; 15. Uljin-gun, Gyeongsangbuk-do (GB); 16. Gangneung-si; 17. Goseong-gun (Songjiho); 18. Goseong-gun (Hwajinpoho), Gangwon-do (GW).
Woonjeong-dong (37.797481; 128.91507); 17. Goseong-gun, Gangwon-do: Songjiho in Jukwang-myeon (38.335426; 128.51339); 18. Goseong-gun, Gangwon-do: Hwajinpoho in Geo-jin-eup (38.466792; 128.442357) (Fig. 1).

Examination of mullets

Total 346 mullets were collected in the above 18 coastal areas for 3 years (2013: 1-4; 2014: 5-11, 16, and 18; 2015: 12-15 and 17). The number of fish collected in each site was designated in Table 1. All collected mullets were transferred to our laboratory (Department of Parasitology and Tropical Medicine, Gyeongsang National University School of Medicine, Jinju, Korea) with ice, and examined by artificial digestion method. Each fish was finely ground with a mortar and pestle or a grinder, the ground fish flesh was mixed well with artificial gastric juice, and the mixture was incubated at 36°C for 2 hr. The digested material was filtered through a 1 × 1 mm of mesh, and washed with 0.85% saline until the supernatant became clear. Trematode metacercariae were collected from sediments under a stereomicroscope, and categorized according to the size and morphological characteristics. The prevalence and intensities of infection were then calculated.

RESULTS

Metacercarial infection in mullets

The metacercariae of *P. summa* were detected in mullets from 15 areas except for 7. Boseong-gun (Jeollanam-do), 13. Pohangsi, and 15. Uljin-gun (Gyeongsangbuk-do). The overall infection rate was 68.2% (236 out of 346 mullets examined). They were found in 76 (93.8%) out of 81 mullets from 4 coastal areas (1-4) of Yellow Sea, 122 (69.3%) of 176 ones from 7 sites (5-11) of South Sea, and in 38 (42.7%) out of 89 mullets from 7 sites (12-18) of East Sea. Their average intensities were 496, 369, and 291 per fish infected, respectively. Especially in mullets from 1. Taean-gun (Chungcheongnam-do) and 11. Geoje-si (Gyeongsangnam-do), their prevalences were 100% and 95.5%, and intensities were 1,116 and 1,444 per fish infected. They were also detected in mullets from 3 coastal lakes, i.e., Gyeongpoho, Songjiho, and Hwajinpoho (‘ho’ means lake), in Gangwon-do, and their average intensities were 419, 147, and 672 per fish infected, respectively. The infection status of *P. summa* metacercariae in mullets by collection sites is shown in Table 1.

Other heterophyid metacercariae

The metacerciae of 5 other species of heterophyids, includ-
of 176 mullets from 7 areas of South Sea, and in 38 (42.7%) out of 89 mullets from 7 sites of East Sea. The average intensities were 496, 369, and 291 metacercariae per fish infected, respectively. Whereas, in Guk et al. [17], the prevalences were 87.0, 54.1, and 4.0% in mullets from the coastal areas of Yellow Sea, South Sea, and East Sea, and their average intensities were 177, 47, and 8 per fish infected, respectively. The trends of metacercarial infection in this study, the higher prevalence and higher intensity in mullets from the coastal areas in Yellow Sea than in other 2 Seas, were similar to those of Guk et al. [17], although each data of this study was much higher. This endemic trend of *P. summa* metacercariae in mullets coincided well with the trend that human cases with this fluke have been mainly reported in the western and southern coastal areas, i.e., Okgu-gun and Buan-gun (Jeollabuk-do), Shinan-gun, Muan-gun, Gangjin-gun, and Yeongdeok-gun (Jeollanam-do), in Korea [4-11]. Among 18 surveyed areas, 5 areas, including Muan-gun, Shinan-gun, Gangjin-gun, Sacheon-si, and Yeongdeok-gun, were the same regions as in Guk et al. [17]. The prevalence and metacercarial density in mullets from these 5 regions were somewhat different between the 2 studies. In the present study, the prevalences were 100, 90.5, 44.7, 94.1, and 37.5% in Muan-gun, Shinan-gun, Gangjin-gun, Sacheon-si, and Yeongdeokgun, respectively, and intensities were 20, 168, 12, 25, and 8 metacercariae per fish infected, respectively. Whereas, in Guk et al. [17], the prevalences were 100, 100, 70, 77, and 0%, and the metacercarial densities were 59, 483, 2, 71, and 0, respectively. These differences were probably due to the collection time and site of mullets.

In previous studies [14,17], *P. summa* metacercariae were rarely detected in mullets from eastern coastal areas. Seo et al. [14] could not find them at all in mullets from Ulsan Metropolitan City, Pohang-si, Yeongdeok-gun (Gyeongsangbuk-do), and Gangneung-si (Gangwon-do). Guk et al. [17] detected total 8 *P. summa* metacercariae in only 1 mullet from Donghae-si (Gangwon-do). They could not detect them in mullets from Yeongdeok-gun (Gyeongsangbuk-do) and Sokcho-si (Gangwon-do) [17]. In the present study, we detected total 11,058 *P. summa* metacercariae (291 in average) in 38 (63.3%) out of 60 mullets from 5 eastern coastal sites, i.e., Ulsan Metropolitan City, Yeongdeok-gun (Gyeongsangbuk-do), and Gangneung-si, Goseong-gun (Songiho and Hwajinpoho, Gangwon-do). Especially in mullets from 3 coastal lakes, Gyeongpoho, Songiho, and Hwajinpoho, in
In the present study, the metacercariae of *H. nocens* were detected in mullets from 7 localities, which included Muan-gun, Shinan-gun, Haenam-gun, Gangjin-gun, and Boseong-gun (Jeollanam-do), Hadong-gun, and Geoje-si (Jeollanam-do), in western and southern coasts of Korea. Their prevalences were 66.7, 100, 28.6, 81.6, 3.9, 61.5, and 27.3%, and the intensities were 64, 84, 119, 99, 1, 24, and 24 metacercariae per fish infected, respectively. In Guk et al. [17], *H. nocens* metacercariae were found in mullets from 7 surveyed areas, i.e., Ganghwa-gun (Gyeonggi-do), Geoje-si (Gyeongsangnam-do), Buan-gun (Jeollabuk-do), Muan-gun, Shinan-gun, Gangjin-gun (Jeollanam-do), and Sacheon-si (Gyeongsangnam-do), in western and southern coasts of Korea. Their prevalences were 42-100%, and the intensities were 4-271 metacercariae per fish infected [17]. On the other hand, it has been known that *H. nocens* is the dominant intestinal fluke species among the residents of western and southern coastal areas, i.e., Buan-gun, Muan-gun, Shinan-gun, Gangjin-gun, and Sacheon-si, in Korea [4-11]. Accordingly, by the present study, it became clear that the endemic areas of *H. nocens* are closely related to the distribution of mullets highly infected with the metacercariae of this species.

More than 18 fish species have been reported as the second intermediate hosts of *H. continua* in Asian countries, such as Japan, China, Korea, and Vietnam [15,16,19-27]. Among them, mullets are most widely distributed and highly important as the source of human infections. Human infection cases have been sporadically reported in western and southern coastal areas, i.e., Gochang-gun (Jeollabuk-do), Shinan-gun, Haenam-gun, Youngam-gun (Jeollanam-do), and Sacheon-si (Gyeongsangnam-do), in Korea [5,6,8-10,22,28]. Although infection rates and densities of *H. continua* metacercariae were relatively low, they were found in mullets from western and southern coastal areas, i.e., Gochang-gun (Jeollabuk-do), Shinan-gun, Haenam-gun, Gangjin-gun, Boseong-gun (Jeollanam-do), Hadong-gun, and Geoje-si (Gyeongsangnam-do) in the present study.

Collectively, heterophyid flukes, including *P. summa*, are prevalent in western and southern coastal areas, which developed the tideland in brackish zones. However, it has been confirmed for the first time that *P. summa* metacercariae are heavily infected in mullets from some lakes in eastern coastal areas of Korea.

**ACKNOWLEDGMENTS**

This study was supported by an Anti-Communicable Diseases Control Program, 2013 (Studies on the biological resources of human infecting trematodes and their larval infections in intermediate hosts), 2014 (Investigation of fishborne parasites and acquisition of their biological resources in the southern regions of Korea), and 2015 (Investigation of fishborne parasites and acquisition of their biological resources in the eastern regions of Korea) of National Research Institute of Health (NRIH), Korea Centers for Disease Control and Prevention (KCDCP). We thank Jung-A Kim and Hee-Ju Kim, Department of Parasitology and Tropical Medicine, Gyeongsang National University School of Medicine, Jinju, Korea, for their help in fish examinations.

**CONFLICT OF INTEREST**

We have no conflict of interest related to this work.

**REFERENCES**

1. Onji Y, Nishio T. On the trematodes whose intermediate host is brackish water fishes. Chiba Igaku Semmon Gakko Zasshi 1916; 81 & 82: 229-249.
2. Takahashi S. On the eggs of *Stellantchasmus falcatus* and *Pygidiopsis summa* found in human stools. Okayama Igakkai Zasshi 1929; 41: 1502-1513.
3. Yokogawa M, Sano M, Itabashi T, Kachi S. Studies on the intestinal flukes II. Epidemiological studies on heterophyid trematodes of man in Chiba Prefecture. Jpn J Parasitol 1965; 14: 577-585.
4. Chai JY, Nam HK, Kook J, Lee SH. The first discovery of an endemic focus of *Heterophyes nocens* (Heterophyidae) infection in Korea. Korean J Parasitol 1994; 32: 157-161.
5. Chai JY, Kim IM, Seo M, Guk SM, Kim JL, Sohn WM, Lee SH. A new endemic focus of *Heterophyes nocens*, *Pygidiopsis summa*, and other intestinal flukes in a coastal area of Muan-gun, Cholla-nam-do. Korean J Parasitol 1997; 35: 233-238.
6. Chai JY, Song TE, Han ET, Guk SM, Park YK, Choi MH, Lee SH. Two endemic foci of heterophyids and other intestinal fluke infections in southern and western coastal areas in Korea. Kore-
11. Cho SH, Cho PY, Lee DM, Kim TS, Kim IS, Hwang EJ, Na BK, Sohn WM. Epidemiological survey on the infection of intestinal flukes in residents of Muan-gun, Jeollanam-do, the Republic of Korea. Korean J Parasitol 2010; 48: 133-138.

12. Chai JY, Lee SH. Food-borne intestinal trematode infections in the Republic of Korea. Parasitol Int 2002; 51: 129-154.

13. Chun SK. A study on some trematodes whose intermediate host are brackish water fish. (I) The life history of Heterophyes continua, the intermediate host of which is Laterolabrax japonicus. Bull Pusan Fish Coll 1963; 5: 1-5 (in Korean).

14. Seo BS, Hong ST, Chai JY, Cho SY. Studies on intestinal trematodes in Korea. IV. Geographical distribution of Pygidiopsis and Heterophyes metacercariae. Seoul J Med 1981; 22: 236-242.

15. Sohn WM, Han GG, Kho WG, Chai JY, Lee SH. Infection status with the metacercariae of heterophyid flukes in the brackish water fish from Haenam-gun, Chollanam-do, Korea. Korean J Parasitol 1994; 32: 163-169 (in Korean).

16. Kim DC, Kim TS, Cho SH, Song HJ, Sohn WM. Heterophyid metacercarial infections in brackish water fishes from Jinju-man (Bay), Kyongsangnam-do, Korea. Korean J Parasitol 2006; 45: 7-13.

17. Guk SM, Shin EH, Kim JL, Sohn WM, Hong KS, Yoon CH, Lee SH, Rim HJ, Chai JY. A survey of Heterophyes nocens and Pygidiopsis summa metacercariae in mullets and gobies along the coastal areas of the Republic of Korea. Korean J Parasitol 2007; 45: 205-211.

18. Cho SH, Kim IS, Hwang EJ, Kim TS, Na BK, Sohn WM. Infection status of estuarine fish and oysters with intestinal fluke metacercariae in Muan-gun, Jeollanam-do, Korea. Korean J Parasitol 2012; 50: 215-220.

19. Kanemitsu T, Akaï T, Otagaki H, Kagi F. Studies on the trematodes of the genus Metagonimus, of which intermediate hosts are brackish water fishes; with additional notes on Heterophyes continua, of which intermediate host is Laterolabrax japonicus. Kyoto Igaku 1953; 6: 296-304 (in Japanese).

20. Chun SK. A study on some trematodes whose intermediate host are brackish water fish. (1) The life history of Heterophyes continua, the intermediate host of which is Laterolabrax japonicus. Bull Pusan Fish Coll 1960; 3: 40-44 (in Korean).

21. Kobayashi H. Studies on the trematoda in Hainan Island, South China and Vietnam (French Indochina). Reports of Scientific works by H. Kobayashi. 1968. pp 155-251.

22. Seo BS, Lee SH, Chai JY, Hong SJ. Studies on intestinal trematodes in Korea XIII. Two cases of natural human infection by Heterophyopsis continua and the status of metacercarial infection in brackish water fishes. Korean J Parasitol 1984; 22: 51-60.

23. Cho SY, Kim SI. 1985. Plecoglossus altivelis as a new fish intermediate host of Heterophyopsis continua. Korean J Parasitol 1985; 23: 173-174.

24. Sohn WM, Na BK, Cho SH. Echinostoma hortense and heterophyid metacercariae encysted in yellowfin goby, Acantohuebies flaviminans, from Shinan-gun and Muan-gun (Jeollanam-do), Korea. Korean J Parasitol 2009; 47: 307-310.

25. Sohn WM, Kim JA, Cho HJ. Two species of goby, Boleophthalmus pectinirostris and Scartelaos sp., as the new second intermediate host of heterophyid flukes in Korea. Korean J Parasitol 2005; 43: 161-164.

26. Vo DT, Murrell D, Dalsgaard A, Bristow G, Nguyen DH, Bui TN, Vo DT. Prevalence of zoonotic metacercariae in two species of grouper, Epinephelus coioides and Epinephelus bleekeri, and flathead mullet, Mugil cephalus, in Vietnam. Korean J Parasitol 2008; 46: 77-82.

27. Chai JY, De NV, Sohn WM. Foodborne trematode metacercariae in fish from northern Vietnam and their adults recovered from experimental hamsters. Korean J Parasitol 2012; 50: 317-325.

28. Hong SJ, Chang CK, Lee DH, Woo HC. One human case of natural infection by Heterophyopsis continua and three other species of intestinal trematodes. Korean J Parasitol 1996; 34: 87-90.