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Studies on Intestinal Trematodes in Korea

XVI. Infection Status of Loaches with the Metacercariae of *Echinostoma hortense*

Jong-Yil Chai, Sung-Jong Hong, Woon-Mok Sohn, Soon-Hyung Lee and Byong-Seol Seo

Department of Parasitology and Institute of Endemic Diseases, College of Medicine, Seoul National University, Seoul 110, Korea

INTRODUCTION

Echinostoma hortense, one of the fluke family Echinostomatidae, was first discovered by Asada (1926) from the intestine of albino rats experimentally infected with the metacercariae from the frogs. Thereafter its natural infection in the domestic or wild animals such as the rats (Yamaguti, 1933; Park, 1938; Seo *et al.*, 1964 & 1981), the dogs (Asada, 1927; Cho *et al.*, 1981) and the weasles (Yamaguti, 1939; Kamiya *et* Ishigaki, 1972), has been reported in Korea, Japan and Manchuria. Since human infection was not known before the report of Tani *et al.* (1974), it was only recently that this fluke began to draw medical attentions. At present, however, more than 20 human cases are known in the literature (Makino *et al.*, 1982; Seo *et al.*, 1983). Major clinical symptoms were gastrointestinal troubles such as abdominal pain and diarrhea in experimentally infected human volunteers (Arizono *et al.*, 1976; Tani, 1979).

The amphibia such as the frogs with their tadpoles (Asada, 1926) and the larvae of salamander (Mori, 1935), and the freshwater fishes such as the loaches (Ono, 1930) and the Japanese bitterlings (Tani, 1976b) are the second intermediate hosts of *E. hortense*. In the human cases reported in Japan, the source of infection was suggested to be the loach (Tani, 1976a; Arizono *et al.*, 1976). In Korea, however, there has been few study on intermediate hosts of this fluke. As to the loach, there is only one paper on the detection of the metacercariae of *E. hortense* (Seo *et al.*, 1980). The infection rate of the loaches, the metacarial density, *etc.* are not known so far. In this respect, this study was undertaken to evaluate the status of metacerarial infection in the loaches of several localities in Korea.

MATERIALS AND METHODS

A total of 154 loaches, *Misgurnus anguillicaudatus*, were purchased from 4 local markets and the infection rate, density and location of the metacercariae of *E. hortense* were examined. The fish body was divided into head and gill, muscle, scale and fin, proximal and distal intestines, and visceral organs. Each portion was examined either by compression method between two slide glasses or by peptic digestion technique. The compression method was applied to the intestine, which was cut into many pieces from the proximal to distal portions with the attached mesentery and peritoneum. Other portions, after peptic digestion, were filtered through mesh to remove the large tissue debris, and the precipitates were examined for the metacercariae.

The metacercariae of *E. hortense* were identified morphologically by the size and shape of their cyst, and by the characteristic features of the internal organ structures. Especially those with head crown bearing a total of 26~28 spines or bearing 4 end group ones were regarded as *E.*

hortense. These metacercariae were finally identified by the adult worms obtained after experimental infection to albino rats, of which the details are described in a separate paper (Seo *et al.*, 1985).

RESULTS

The metacercariae of *E. hortense* from the loaches were round to elliptical in shape (Fig. 1 & 2), and 160~175μm long and 140~155μm wide. In encysted state, dorsally uninterrupted collar spines (arrow heads; Fig. 3) and 4 end group spines on each side were easily seen (Fig. 4), however, in some cases, thorough counting of the whole collar spines was difficult. The excysted metacercariae were, under slight pressure, 315~431μm long and 103~120μm wide (Fig. 5). The oral sucker was 57~62μm and the ventral sucker 60~70μm in diameter. The number of total collar spines was counted to be about 26~28 (Fig. 6 & 7). All other morphological features were identical to those described by Saito *et* Tani (1982).

Out of 154 loaches collected at 4 areas, 64 (41.6%) were found infected with the metacercariae of *E. hortense* (Table 1). From the infected loaches, a total of 516 metacercariae (1~29 per loach) were collected and the average number per loach was 8.1. The infection rate and the metacercarial density of the loaches were much different in 4 areas. The highest infection rate and density were obtained from the loaches collected at Kangjin-gun, Jeonranam-do and the lowest metacercarial density from Naju-gun, Jeonranam-do.

The metacercariae of *E. hortense* revealed peculiar distributions in the body of the loach (Table 2). They were most frequently found from the wall of the distal intestine including the adjacent mesentery and perianal tissue. As much as 372 (72.1%) metacercariae were detected from these areas. Almost all of others (27.5 %) were found from the tissues of head and gill. In comparison, the muscles, proximal part of the intestine, and other visceral organs revealed no metacercariae at all. The scales and fins revealed only two metacercariae in number, which seem to have been from the skin near the perianal region.

Table 1. Intfection status of the loaches with the metacercariae of *E. hortense*

Locality of loaches	No. exam.	No. posit. (%)	No. metacer. collected	
			Total (*)	Range/loach
Kangjin-gun (Jeonranam-do)	50	37 (74.0)	380 (10.3)	1-24
Sooyoo Market (Seoul)	54	12 (22.2)	74 (6.2)	1-29
Kimhae City	40	12 (30.0)	58 (4.8)	1-22
Naju-gun (Jeonranam-do)	10	3 (30.0)	4 (1.3)	1-2
Total	154	64 (41.6)	516 (8.1)	1-29

* Average/infected loach

Table 2. Location of *E. hortense* metacercariae in body of the loaches

Body portions	No. metacercariae (%)
Head and gill	142 (27.5)
Muscle	0
Scale and fin	2 (0.4)
Proximal intestine*	0
Distal intestine* and perianal tissue	372 (72.1)
Visceral organs	0
Total	516

* Including the adjacent mesenteries

DISCUSSION

The first report on the detection of the metacercariae of *E. hortense* in loaches was made in Japan by Ono (1930), under the name of *Echinostoma campi*, which is regarded as a synonym of *E. hortense*. Much later than that, Okahashi (1966) reported 45.0% positive rate of the loaches collected from Okayama Prefecture. In other reports made in Japan, the infection rate of loaches was 23.8% among 42 purchased at the markets in Osaka City (Arizono *et al.*, 1976), 49.2% among 120 collected from 6 areas in Akita Prefecture (Tani, 1976) and 38.8~

44.1% among 543 from two areas in Hokkaido (Miyamoto *et al.*, 1983). According to these reports, the average metacercarial density ranged 2.5~6.8 per loach. In the present study, the infection rate and metacercaial density of 154 loaches collected at 4 areas were 41.6% and 8.1 respectively, so that it is suggested that *E. hortense* infection in the loaches in Korea should not be lower than in Japan.

Fortunately, however, the Koreans generally do not eat the loaches under raw condition, and instead, they prefer the boiled soup of the ground loaches. This may be one the reasons for rare occurrence of human *E. hortense* infection in Korea; only 4 cases (Seo *et al.*, 1983; Chai, 1984; Ryang *et al.*, 1985). In Japan where the raw loaches are preferred by some of the people, at least 20 human cases have been reported (Makino *et al.*, 1982).

It is, however, noteworthy that some of the freshwater fishes other than the loach may take the role of the source of human infection. Tani (1976b) reported that the Japanese bitterlings, *Acheilognathus moriokae*, also carried the metacercariae. Ryang *et al.* (1985) detected the metacercariae from another kind of the freshwater fish, *Moroco oxycephalus*, and it was regarded as the source of human infection of two cases found by them. Furthermore, it is also notable that Ono (1930) succeeded in infection of the silver carp and the goldfish with the cercariae which were emerged from the snails, *Lymnaea* sp. In practice, the first human case reported in Korea (Seo *et al.*, 1983) had the history of eating various kinds of freshwater fishes but not the loach. In this respect, further studies are needed to search for possible sources of human infection in Korea.

On the location of the metacercariae of *E. hortense* in body of the loaches, there have been some disagreements among the investigators. Ono (1930) and Arizono *et al.* (1976) found the metacercariae from the soft tissues adjacent to gill, while Okahashi (1966) detected them from the mesentery. On the other hand, Saito *et* Tani (1982) collected more metacercariae from the perianal skin as well as the head tissue than other portions including the distal intestine and the mesentery. In the present study, the metacercariae were most frequently found from the wall of the distal intestine and the perianal tissue, followed by the head tissue. This result is most similar to that of Saito *et* Tani (1982). As to the other freshwater fishes, the metacercarial density and their location in the fish body have not been sufficiently studied.

SUMMARY

The infection stasus of the loaches, *Misgurnus anguillicaudatus*, with the metacercariae of *Echinostoma hortense*, was studied in Korea. A total of 154 loaches purchased at 4 local markets (Seoul, Kimhae, Naju-gun and Kangjin-gun) were examined their infection rate as well as the density and location of the metacercariae in the fish body.

The results are as follows:

1. The loaches carrying the metacerariae of *E. hortense* were 64 (41.6%) in total number and the metacercarial density ranged 1-29 per infected loach with an average value of 8.1. The highest infection rate and metacercarial density were obtained from the loaches purchased at Kangjin-gun, Jeonranam-do.

2. The metacercariae of *E. hortense* were chiefly distributed in the distal intestinal wall and the adjacent mesentery, the perianal tissues, and the head and gill of the loaches examined.

From the results, it is concluded that the loach is one of the important second intermediate hosts of *E. hortense* in Korea, and their infection rate and metacercarial density are considerably high.

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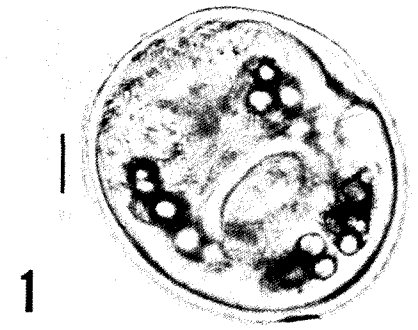

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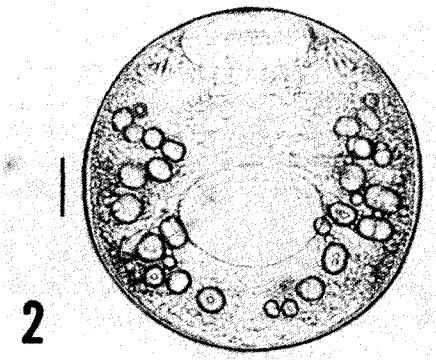

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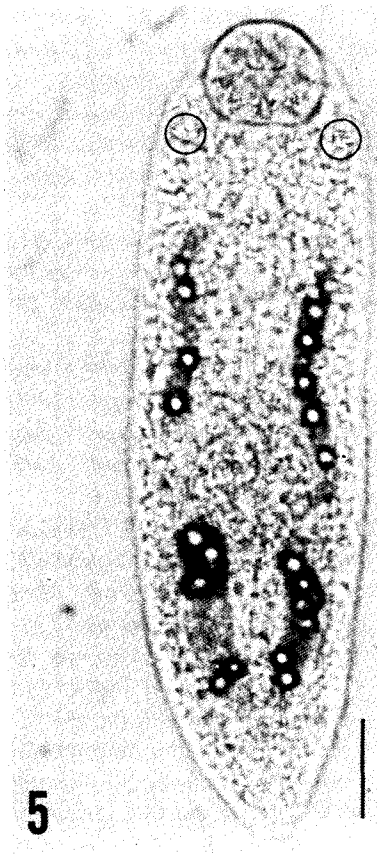

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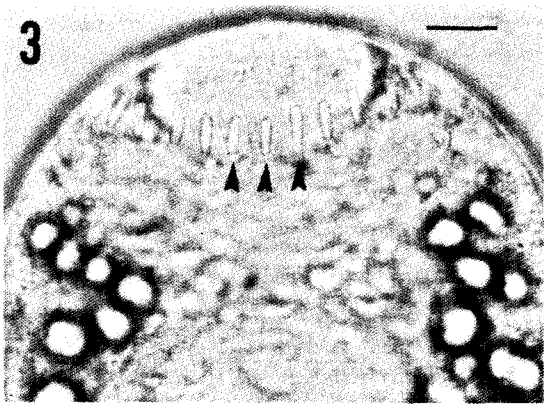

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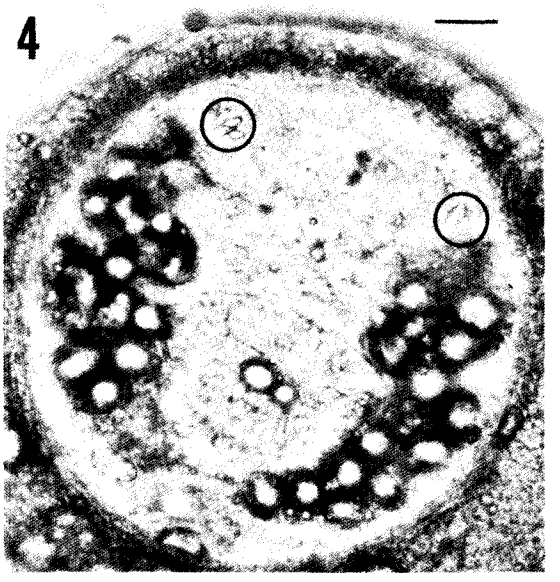

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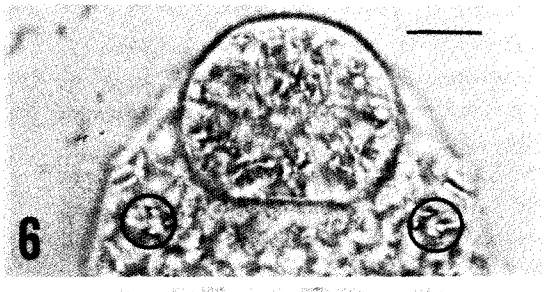

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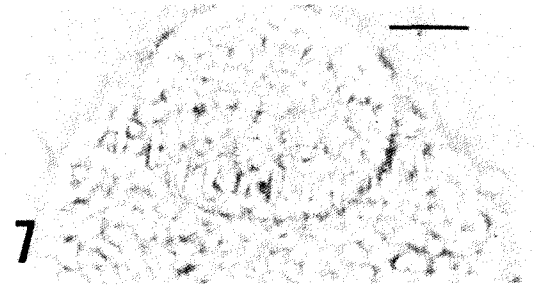

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=국문초록=

韓國의 腸吸蟲에 관한 硏究

XVI. 미꾸리의 호르덴스棘口吸蟲 被囊幼蟲 感染狀況

서울大學校 醫科大學 寄生蟲學敎室 및 風土病硏究所
蔡鍾一・洪性琮・孫運睦・李純炯・徐丙尙

미꾸리(*Misgurnus anguillicaudatus*)의 호르덴스棘口吸蟲 被囊幼蟲 感染狀況을 조사하였다. 전국 4個 地域(서울, 김해, 나주군 및 강진군)에서 총 154마리의 미꾸리를 구입하여 被囊幼蟲 感染率, 感染量 및 미꾸리 體內 寄生部位를 조사한 바 그 결과는 다음과 같다.

1. 호르덴스棘口吸蟲 被囊幼蟲이 검출된 미꾸리는 총 64마리로 41.6%의 陽性率을 보였고 마리당 1~29個(平均 8.1個)가 검출되었다. 가장 높은 被囊幼蟲 陽性率을 보인 地域은 전남 강진군이었다.

2. 被囊幼蟲은 주로 미꾸리의 腸管 말단부 및 腸間膜, 肛門周圍組織, 頭部 및 아가미에서 검출되었고 근육 등에서는 전혀 검출되지 않았다.

이상의 결과로, 우리나라의 미꾸리는 호르덴스棘口吸蟲의 중요한 第二中間宿主임이 판명되었으며 感染率 및 感染量도 비교적 높음을 알 수 있었다.

EXPLANATIONS FOR FIGURES

Fig. 1. Encysted metacercaria of *E. hortense*, unpressed (scale: 30μm).

Fig. 2. Another metacercaria showing the characteristic structures of collar spines, suckers and excretory bladder with large corpuscles, pressed specimen (scale: 30μm).

Fig. 3. Magnification of Fig. 2. The dorsally uninterrupted collar spines (arrow heads) are seen (scale: 20μm).

Fig. 4. Another matacercaria in the perianal tissue of a loach. Note 4 end group spines (circles) at both sides of the oral sucker (scale: 20μm).

Fig. 5. An excysted metacercaria showing the oral sucker with collar spines, ventral sucker and excretory bladder. Note 4 end group spines (circles) (scale: 50μm).

Fig. 6. Magnification of Fig. 5, showing the spines of end groups (circles) and several lateral ones (scale: 20μm).

Fig. 7. Dorsal view of the oral sucker of another metacercaria showing the dorsally uninterrupted collar spines (scale: 20μm).