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Infection status of dragonflies with *Plagiorchis muris* metacercariae in Korea

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Abstract: *Plagiorchis muris* has been found in both house and field rats as well as in humans. The infection status of the second intermediate hosts of *P. muris* is prerequisite in understanding their biological features in an ecosystem. Six species of dragonflies were caught in a wide range of areas in Korea; and they were *Sympetrum darwinianum, S. eroticum, S. pedomontanum, S. infuscatum, Pantala flavoscens, Calopteryx atrata,* and *Orthetrum albistylum speciosum*. The occurrence of *P. muris* metacercariae in dragonflies was nationwide with various infection rates. The metacercarial burden of *P. muris* in the surveyed areas was the highest in *S. eroticum* followed by *S. darwinianum, S. pedomontanum,* and *C. atrata*. The highest infection rate by *P. muris* metacercariae was found in *S. darwinianum* followed by *S. eroticum*. The metacercarial burden was particularly heavy in the dragonflies found in Hamyang-gun and Kosong-gun, Kyongsangnam-do. It is, therefore, likely that dragonflies play a significant role as the second intermediate host in the life cycle of *P. muris* in Korea.

Key words: *Plagiorchis muris*, metacercaria, dragonfly, *Sympetrum* spp.

INTRODUCTION

Flukes of the genus *Plagiorchis* have been found in the small intestine of mammals, including humans, birds, reptiles, amphibians, and fish (Radomyos et al., 1989). The second intermediate hosts of the flukes are aquatic insects, such as mosquito larvae, insect naiads, freshwater snails, and freshwater fishes (Tanabe, 1922; Asada et al., 1962; Komiya, 1965; Hong et al., 1996).

In Korea, of the genus *Plagiorchis, P. koreanus, P. orientalis, P. corpulentus, P. magnacotylus, P. vespertilionis, P. rhinolophi,* and *P. kyushuensis* were reported from bats (Park, 1939a, 1939b; Sogandares-Bernal, 1956; Kifune et al., 1983). *Plagiorchis muris* was collected from wild or house rats captured in Yongin-gun and Pochon-gun, Kyonggi-do, and in Chorwon-gun and Yangyang-gun, Kangwon-do, and from house rats in Hadong-gun, Kyongsangnam-do, and in Seoul (Seo et al., 1964, 1981; Lee et al., 1990). Moreover, human infection by *P. muris* was reported in Hamyang-gun, Kyongsangnam-do (Hong et al., 1996). From these reports, the distribution of *P. muris* is considered to be nationwide in Korea. The second intermediate hosts, such as dragonflies and freshwater fishes were found to be infected by the metacercariae of *P. muris* (Hong et al., 1998). In albino rats, the meta-

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cercaria of *P. muris* grew rapidly to an ovigerous adult on day 4 post-infection (Hong et al., 1998).

The dragonfly is one of the most predominant insects found in Korea during summer. Dragonflies, naiads, and chironomids, all with the metacercariae of *P. muris*, are easy targets of prey for fish, birds, and other mammals (Tanabe, 1922; Komiya, 1965). In this context, it is likely that dragonflies play a role in the life cycle of *P. muris* in Korea. There have not been any studies regarding the second intermediate host of the genus *Plagiorchis*. This survey was, therefore, conducted to elucidate the infection status of the dragonflies with *P. muris* metacercariae in Korea.

MATERIALS AND METHODS

Dragonflies were caught near ricepaddies, canals, streams, and/or villages in 20 local areas in Korea from May 1993 to August 1994 (Table 1). Captured dragonflies were identified according to Shin's method (1993) and examined individually or as a whole for the metacercariae of *P. muris* by artificial digestion method. As a way of estimating the metacercarial burden in the dragonflies in a given area, dragonflies of the same species were digested as a whole. On the other hand, in order to get an information on the infection rates of the dragonflies with *P. muris* metacercariae, a dominant species was subjected to the individual examination. The dragonflies were digested for 1 hr in artificial digestion juice (pepsin 5-6 g (Sigma, St. Louis, USA), 10 ml HCl in 1 L H_2O) at 37°C. The digest was washed and sedimented in physiological saline several times to clarity. The metacercariae of *P. muris* were collected from the sediment of digest under the dissecting microscope and identified under the light microscope (Hong et al., 1998). The data of individual examination was not included to those of the whole examination. The metacercariae collected were grouped into 50 to 100 in number and fed to albino rats with a gastric needle. The flukes were recovered from the small intestine of albino rats up to 28 days after the metacercarial infection. All of the flukes recovered were stained with Semichon's acetocarmine and identified microscopically to *P. muris* (Hong et al., 1998).

RESULTS

The metacercaria

The metacercaria of *P. muris*, 0.169 (0.165-0.174) mm by 0.158 (0.148-0.170) mm, was elliptical to spherical in shape with a hyaline thin wall (Fig. 1). The oral sucker was subterminal and armed with a short flame-shape stylet. The acetabulum was round, smaller than the oral sucker and was located in the middle of the body. The excretory bladder was Y-shape and filled with refractile granules. No primordium of genital organs was found in the metacercaria.

Infection status of the dragonflies

A total of 4,172 dragonflies was caught from 20 different areas through this survey. The species of captured dragonflies consisted of *Sympetrum darwinianum*, *S. eroticum*, *S. pedomontanum*, *S. infuscatum*, *Pantala flavoscens*, *Calopteryx atrata*, and *Orthetrum albistylum speciosum*. Of the species, *S.*

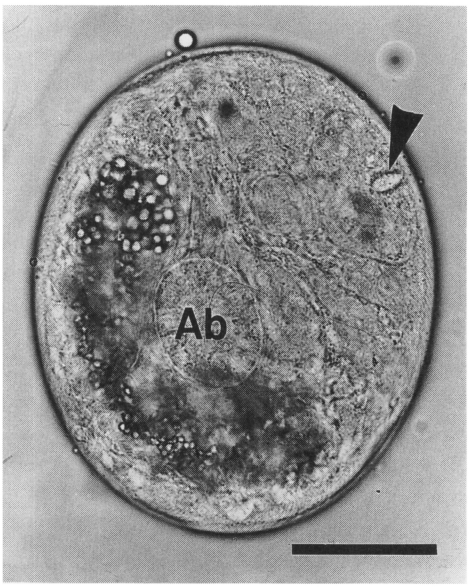

Fig. 1. A *Plagiorchis muris* metacercaria collected from a dragonfly, *Sympetrum darwinianum*. A stylet (arrowhead) is in the oral sucker which is larger than the acetabulum (Ab). Bar=50 μm.

Table 1. Infection status of *Plagiorchis muris* metacercariae in dragonflies captured from 20 areas in Korea

Area surveyed	Dragonflies[a)]		No. of metacercariae
	Species[b)]	No. of examined	
Kyonggi-do			
Yongin-up, Yongin-gun	*S. eroticum*	74	78
	S. pedomontanum	33	3
	S. darwinianum	4	0
Yongjong-myon, Pochon-gun	*S. darwinianum*	5	2
	S. eroticum	68	0
	S. pedomontanum	11	0
	P. flavoscens	11	0
Kangwon-do			
Tongri-dong, Taebaek-shi	*S. darwinianum*	188	27
Sangseo-myon, Hwachon-gun	*S. eroticum*	102	1
	S. darwinianum	17	5
	P. flavoscens	33	1
	S. infuscatum	7	0
Yopo-dong, Chunchon-shi	*P. flavoscens*	137	1
Kirin-myon, Inje-gun	*S. darwinianum*	107	15
Chungchongbuk-do			
Angsong-myon, Chungju-shi	*S. darwinianum*	62	1
	S. pedomontanum	10	0
	S. infuscatum	6	0
	P. flavoscens	9	0
	O. albistylum speciosum	9	0
Chollabuk-do			
Mupung-myon, Muju-gun	*S. darwinianum*	163	75
	S. infuscatum	10	0
Chollanam-do			
Changan-myon, Sungju-gun	*S. darwinianum*	99	1
	S. infuscatum	6	0
Kwayog-myon, Kohung-gun	*S. darwinianum*	268	0
	S. eroticum	4	0
Pulgap-myon, Yonggwang-gun	*S. darwinianum*	63	37
	S. eroticum	20	5
Kyongsangbuk-do			
Kajin-myon, Koryong-gun	*S. darwinianum*	88	25
	S. pedomontanum	30	0
	S. eroticum	30	0
Sanyang-myon, Mungyong-gun	*S. eroticum*	8	13
	S. pedomontanum	8	1
	S. darwinianum	26	0
	C. atrata	45	2
Chinbo-myon, Chongsong-gun	*S. darwinianum*	42	2
	S. eroticum	20	4

(continued)

Table 1. continued

Area surveyed	Dragonflies[a]		No. of metacercariae
	Species[b]	No. of examined	
Kyongsangnam-do			
Hamyang-up, Hamyang-gun	*S. darwinianum*	124	236
	S. infuscatum	15	16
	C. atrata	46	10
	S. eroticum	40	2
Yongo-myon, Kosong-gun	*S. eroticum*	851	967
	S. pedomontanum	540	159
	C. atrata	133	100
	S. darwinianum	50	5
	P. flavoscens	32	0
	O. albistylum speciosum	5	0
Popsu-myon, Haman-gun	*S. eroticum*	59	1
	P. flavoscens	13	0
Hwagae-myon, Hadong-gun	*S. darwinianum*	109	0
Myongsok-myon, Chinju-shi	*S. darwinianum*	242	0
	S. eroticum	23	0
Ihyon-dong, Chinju-shi	*S. darwinianum*	39	0
	S. eroticum	18	1
	S. infuscatum	2	0
	O. albistylum speciosum	7	0

[a]Dragonflies caught in a designated area were examined as a whole rather than by individual by the artificial digestion method. [b]For full names, see the text of Results.

Table 2. Infection rates of dragonflies with *Plagiorchis muris* metacercariae by individual examination

Area surveyed	Species[a] of dragonflies	No. of examined	No. of possitive (%)	No. (mean) of metacercariae
Yongin-up, Yongin-gun	*S. eroticum*	20	6 (30.0)	10 (1.7)
Kirin-myon, Inje-gun	*S. darwinianum*	20	4 (20.0)	4 (1.0)
Mupung-myon, Muju-gun	*S. darwinianum*	20	6 (30.0)	10 (1.7)
Pulgap-myon, Yonggwang-gun	*S. darwinianum*	20	7 (35.0)	7 (1.0)
Kajin-myon, Koryong-gun	*S. darwinianum*	20	2 (10.0)	2 (1.0)
Hamyang-up, Hamyang-gun	*S. darwinianum*	10	8 (80.0)	55 (6.9)
Yongo-myon, Kosong-gun	*C. atrata*	20	6 (30.0)	9 (1.5)
	S. eroticum	20	14 (70.0)	21 (1.5)

[a]For full names, see the text of Results.

darwinianum caught from 18 local areas was the most predominating species followed by *S. eroticum*, *S. pedomontanum*, and *P. flavoscens* in descending order (Table 1).

When estimated as a whole, the burden of the dragonflies with *P. muris* metacercariae was the highest in *S. eroticum* by 0.8 per head followed by *C. atrata*, *S. darwinianum*, *S. pedomontanum*, and *S. infuscatum*. Geographically, the metacercarial burden in the dragonflies was very heavy in two areas, Hamyang-gun and Kosong-gun, Kyongsangnam-do; moderate in seven areas, Yongin-gun, Taebaek-shi, Inje-gun, Muju-gun, Yonggwang-gun, Koryong-gun and Mun-gyong-gun; and very low in the rest of the surveyed areas (Table 1).

The infection rate with the metacercaria was

44.0% and the mean metacercarial burden was 3.2 for 90 individually examined *S. darwinianum*. The highest value of the metacercarial infection rate was found from those collected in Hamyang-up, Hamyang-gun, Kyongsangnam-do (Table 2). The metacercaria, however, was not found in *S. darwinianum* caught from six areas.

Sympetrum eroticum, the second frequent species, was caught from 13 areas and was the dominant species in four areas. Through the individual examination, the highest infection rate was found in *S. eroticum* from Yongo-myon, Kosong-gun, but the the heaviest metacercarial burden was detected from Yongin-up, Yongin-gun (Table 2). On the other hand, through the whole examination, the metacercarial burden of the species was the heaviest from Yongo-myon, Kosong-gun (Table 1). The metacercaria was not detected from *S. eroticum* caught from four areas.

Sympetrum pedomontanum and *S. infuscatum* were caught from six areas, and their metacercarial burden was 0.3 each. *Sympetrum pedomontanum* was more abund-ant in numbers than *S. infuscatum* in the surveyed areas. The metacercaria of *P. muris* was found only from 2 out of 235 *P. flavoscens* dragonflies caught from six areas (Table 1).

Calopteryx atrata were caught along the canals and/or streams in three areas. The infection rate of *C. atrata* was 30.0%. The mean metacercarial burden was 1.5 for *C. atrata* captured from Yongo-myon, Kosong-gun (Table 2). The dragonfly, *O. albistylum speciosum*, was very rare and was captured from only three areas. The metacercaria was not detected in this dragonfly (Table 1).

DISCUSSION

The dragonflies, *Sympetrum* spp., were caught at ricepaddies where they emerge from pupae and mature into adults. It was easy to catch dragonflies in the early morning or on rainy days. *Calopteryx atrata* were abundant along canals and streams, and it seemed that they preferred such places. *Pantala flavoscens* were caught from a swarm of dragonflies flying in the air (Shin, 1993).

The size and shape of the cyst and the features of somatic organs of the metacercaria agreed well with the characters of *P. muris* metacercaria (Komiya, 1965; Hong et al., 1998).

The adult *P. muris* was collected from field rats captured from Chorwon-gun and Pochon-gun (Seo et al., 1964), and from house rats found in Yongin-gun and Hadong-gun (Seo et al., 1981). The metacercarial burden in the dragonflies in these areas appeared not to be closely related to the occurrence of the adult flukes in the rat, even though there was a big difference between the two surveys. In Yongin-gun, it was suggested that the occurrence of adult *P. muris* in house rats could be related to the metacercariae in dragonflies, because field rats could feed on dead dragonflies.

Seven species of *Plagiorchis* have been recorded from Korean bats (Park, 1939a, 1939b; Sogandares-Bernal, 1956; Kifune et al., 1983). Bats are nocturnal animals with a precision ultrasonar system. It is likely that bats feed on the insects such as dragonflies, mosquitos, and chironomids which are known to be the second intermediate host of *P. muris* (Tanabe, 1922; Komiya, 1965).

The aquatic insect larvae have been suggested to be a source of human infection of *P. muris* in Japan (Asada et al., 1962). The surface water of springs, wells, ponds, streams and rivers contain a wide variety of aquatic insects and/or insect larvae which might be infected with *P. muris* metacercariae as shown in the present survey. By drinking the surface water without a proper treatment, human could be infected by *P. muris* in Korea as was in Japan. In this respect, it is likely that the larvae, unlikely the adults, of dragonflies are a possible source of human plagiorchiasis.

As far as the metacercarial burden in the dragonflies is considered, the insectivorous diurnal birds can be treated as the final hosts of *P. muris* in Korea. To broaden our knowledge on *P. muris*, further researches must be done on the metacercarial infection rate in mosquitos and chironomids. Furthermore, adult flukes found in the birds deserve further investigation.

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