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Ecological Aspects of *Proctoeces ichiharai* (Trematoda: Digenea) Parasitic in *Batillus cornutus* (Gastropoda)

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ABSTRACT: Digenetic trematodes *Proctoeces ichiharai* (Fellodistomidae) in the kidney of top shells *Batillus cornutus* from Misaki, Japan, were examined from February 1974 to October 1975. The incidence of infection increased with shell height; all hosts over 85 mm high were infected. The intensity of infection was low in small top shells; hosts less than 45 mm high harbored at most only 2 parasites. In each host group over 55 mm high, the mean intensity of infection reached 5.0–7.4, the heaviest infection being 45. Parasites of 4–5 mm length always dominated in number, irrespective of host size. As hosts increased in size, parasites of 5–6 mm length became more, those of 3–4 mm less frequent. Consequently, mean parasite body length tended to increase in larger hosts. No marked seasonal variation was recognized in either incidence and intensity of infection or size-frequency distribution. *P. ichiharai* was found to be able to survive for at least 14 months in the renal coelom of the top shell.

INTRODUCTION

The genus *Proctoeces* comprises 15 known species. However, owing to intraspecific morphological variations and inadequate recognition of taxonomic characteristics, questions have been raised regarding the validity of some species described on the basis of some slight morphological differences in a few specimens (Hanson, 1950; Freeman and Llewellyn, 1958; Freeman, 1959; Manter and Pritchard, 1962; Stunkard and Uzmann, 1962; Prévot, 1965). Loos-Frank (1969) and Shimazu (1972) point out that certain ecological aspects, such as life cycle, geographical distribution, and host-parasite relationship should also be considered for specific identification in addition to morphological variations.

Ecological information is usually hard to obtain when working on fish trematodes, because it is usually not possible to collect a large sample. However, in the case of molluscan hosts, relatively large numbers of metacercaria and adult stages can be found. Stunkard and Uzmann (1959) investigated the life cycle of *Proctoeces maculatus* in *Mytilus edulis* from the Woods Hole – New York region. Lang and Dennis (1976) elucidated the seasonal incidence of *P. maculatus* infection in *M. edulis*. White (1972) conducted an ecological investigation of *P. subtenuis* in *Scrobicularia plana* from the Thames Estuary and revealed its localized distribution, level of infection and a relationship between host size and number of parasites. Ichihara (1965) dissected top shells *Turbo (Batillus) cornutus (= B. cornutus)* collected from 13 localities on the Japanese coast and found a localization of *Protoeces* sp., Shimura and Egusa (1979a, b) identified this parasite as *P. ichiharai* and reported its development in *B. cornutus*. Some other ecological notes are discussed by Fujita (1925), Dollfus (1964), Sakaguchi *et al.* (1970a, b).

The top shell *Batillus cornutus* lives in rocky coastal waters facing the open sea and feeds mostly on algae. It is a common marine gastropod of commercial importance in Japan. For some time, various control methods have been adopted to prevent overfishing; now research is also underway on its propagation.

The present paper deals with the population biology of *Proctoeces ichiharai* in the top shell with special reference to its regional occurrence, host-parasite relationship and survival period.

MATERIAL AND METHODS

The material was collected either by diving or with a lobster net at three localities outside Tokyo Bay in Misaki, Chikura and Kominato between February 1974 and October 1975 (Fig. 1). After measuring shell height, the shells were cracked carefully so as to avoid injuring the soft body, and the parasites were picked

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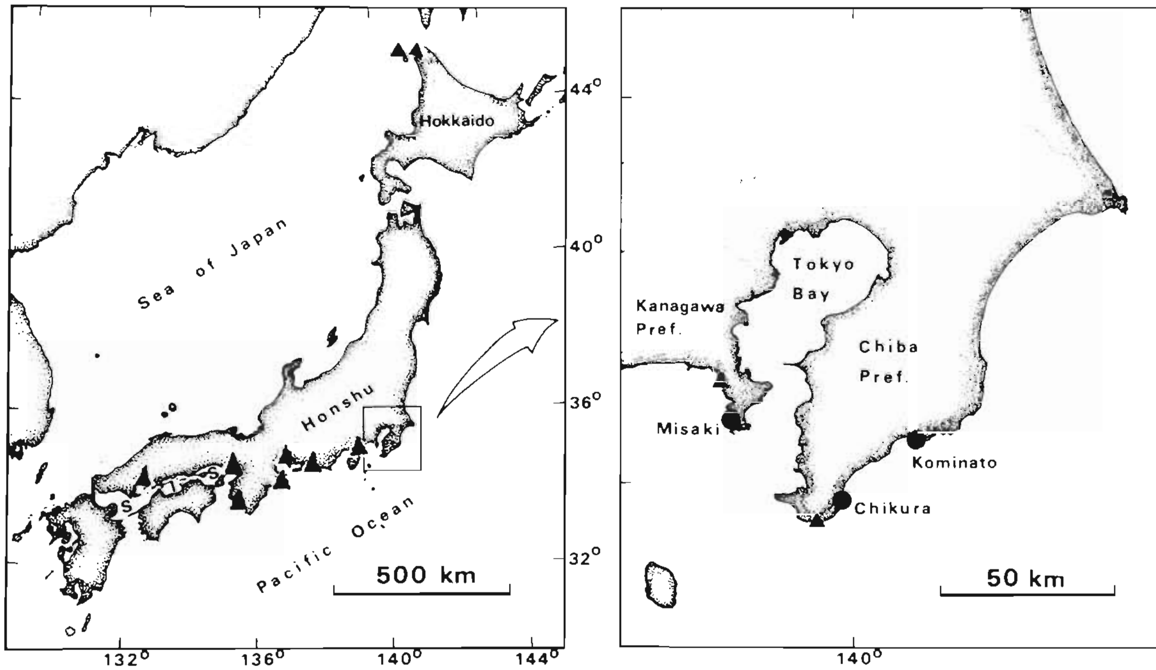

Fig. 1. Map of Japan showing distribution of *Proctoeces* species in Japan based on literature records (▲) and the three sampling locations of the present study (●). SIS: Seto Inland Sea

from the incised renal coelom. In order not to overlook the small specimens, the kidney was taken out, cut into small pieces, pressed between two slides and then examined under a microscope. When fixed in hot formalin, parasites tended to shrink to a definite form. Their body length was measured after such fixation.

RESULTS

The highest incidence of infection was found in hosts taken from Misaki region (81.7 %, 188/230), followed by those from Chikura (29 %, 8/28). No parasite was found in the material from Kominato (0/60), although this location is only about 30 km away from Chikura (Fig. 1). The following results are, therefore, based entirely on material from the Misaki region.

Incidence and Intensity

The incidence and intensity of infection are given in Table 1. The incidence increased in proportion to shell height; all hosts more than 85 mm in height were parasitized. The intensity of infection was low in small top shells less than 55 mm high; hosts under 45 mm high harbored at most only 2 parasites. In larger hosts over 55 mm in height, the mean intensity reached 5.0–7.4, although the number of parasites per host showed a wide range of variation ranging from 0 to 45. Hosts with a heavy infection of more than 10, 20 or 30 parasites occurred at 13.0 % (34 hosts), 3.1 % (8) or 0.8 % (2), respectively. One host of 59.9 mm in height had the heaviest infection with 45 parasites.

Table 1. *Proctoeces ichiharai*. Incidence and intensity of infection as a function of shell height of *Batillus cornutus*

Shell height (mm)	Incidence (%)	Intensity mean	Intensity range	No. of hosts examined
25– 35	31	0.5	0– 2	13
35– 45	44	0.4	0– 1	9
45– 55	67	1.5	0– 9	21
55– 65	86	5.7	0–45	50
65– 75	88	5.4	0–28	94
75– 85	98	7.4	0–38	59
85– 95	100	7.1	1–17	13
95–105		5	1 and 9	2

By taking into consideration of the influence of host size on the level of infection shown in Table 1, top shells ranging from 55 to 85 mm in height were divided into three size groups. Their seasonal incidence and intensity of infection are shown in Table 2. Throughout the year the incidence was higher than 70 % in all cases and especially higher (above 95 %) in the 75–85 mm group. The intensity of infection varied from 2.0 to 10.4 without marked seasonal fluctuation.

Table 2. *Proctoeces ichiharai.* Seasonal incidence and intensity of infection in three size groups of *Batillus cornutus*

Date examined	Shell height (mm) 55–65 Incidence (%)	Intensity (mean)	No.	65–75 Incidence (%)	Intensity (mean)	No.	75–85 Incidence (%)	Intencity (mean)	No.
Apr. 1974	100	6.7	9	89	6.0	9	–	–	0
Aug. 1974	–	–	0	100	6.2	10	100	6.4	11
Oct. 1974	76	7.2	21	86	4.9	21	96	6.0	23
Feb. 1975	100	4.3	3	100	7.2	13	100	10.4	16
Apr. 1975	86	3.9	14	89	5.0	27	100	9.0	1
July 1975	100	2.0	3	71	4.4	14	100	6.5	8

Effect of Host Sex

The effect of host sex on the infection was examined in 94 top shells with mature gonads and free of larval trematodes of unknown species. The incidence of infection was 86 % (36/42) in females and 87 % (45/52) in males. The mean intensity was 4.1 (n=14) in females and 4.2 (n=14) in males of the 65–75 mm size group; it was 7.2 (n=9) in females and 7.4 (n=11) in males of the 75–85 mm group. Therefore, it is concluded that host sex exerts no effect on the level of infection.

Body Length of Parasite

The mean body length of parasites tends to increase in proportion to host size, the correlation coefficient being 0.347 and significant at the 95 % level. The linear relationship of parasite body length and host shell height was determined by the equation

$$Y = aX + b$$

where Y = number of estimated parasites per host, X = shell height in mm, and a and b = constants. The regression equation representing the least square line of relationship was

$$Y = 0.1572X + 3.113$$

Furthermore, body length of parasites was calculated for each host-size group (Table 3). In host groups ranging from 55 to 95 mm high, the mean body length of parasites increased from 4.02 to 4.60 mm in proportion to shell height.

Table 3. *Proctoeces ichiharai.* Body length as a function of shell height of *Batillus cornutus*

B. cornutus Shell height (mm)	No.	*P. ichiharai* Body length (mm) mean	range	SD	No.
25– 35	5	3.81	2.96–5.31	0.72	7
35– 45	1	3.43	–	–	1
45– 55	3	3.60	2.80–4.12	0.43	7
55– 65	14	4.02	2.23–5.51	0.59	121
65– 75	35	4.11	1.78–5.81	0.79	178
75– 85	29	4.30	1.04–5.68	0.62	232
85– 95	8	4.60	2.08–5.80	0.76	47
95–105	1	4.58	3.69–5.03	0.45	9

The size-frequency distribution of *Proctoeces ichiharai* in four host-size groups is shown in Figure 2.

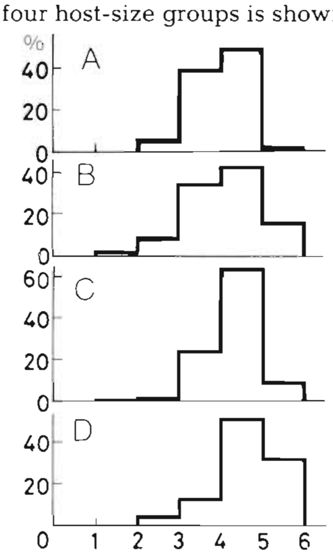

Fig. 2. *Proctoeces ichiharai.* Size-frequency distributions in four size groups of *Batillus cornutus.* A: 55–65 mm in shell height, n = 121; B: 65–75 mm, n = 179; C: 75–85 mm, n = 234; D: 85–95 mm, n = 47

Parasites 4–5 mm long consistently showed a high frequency in each group. In proportion to increased shell height, the frequency decreased for 3–4 mm long parasites and increased for those 5–6 mm long. Parasites smaller than 3 mm were least abundant in each group.

The seasonal pattern of size-frequency distribution of *Proctoeces ichiharai* is shown in Figure 3 based on parasites taken from hosts 55 to 85 mm in height. No distinctive variation was recognized in the histograms throughout the year. Larger specimens were always present as the major constituent and the modes were located at the range 4.2–5.0 mm. Consequently, it is assumed that top shells are infected infrequently but are always exposed to infection.

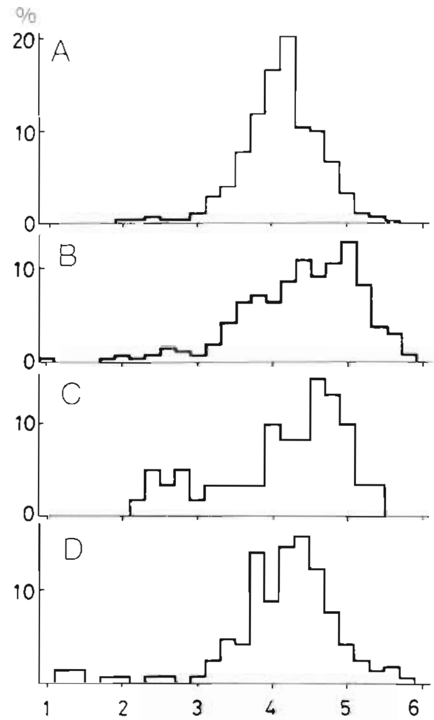

Fig. 3. *Proctoeces ichiharai.* Seasonal size-frequency distributions. A: October 1974; n = 272; B: February 1975, n = 265; C: April 1975, n = 61; D: July 1975, n = 172

Host Range

The following invertebrates collected from the Misaki region were examined for parasites: Polychaeta (2 spp., n = 21), Gastropoda (13 spp., n = 109), Pelecypoda including 118 *Mytilus edulis* (5 spp., n = 154), Asterioidea (4 spp., n = 15) and Urochordata (1 sp., n = 20). However, metacercariae of the genus *Proctoeces*, apparently *P. ichiharai*, were found only in Gastropoda in the kidney and buccal cavity of *Sulculus diversicolor aquatilis* (2/12), in the kidney of *Chlorostoma argyrostoma lischkei* (2/12) and in the kidney and papillary sac of *Marmarostoma stenogyrum* (3/10). The parasites were small, 1.4–3.9 mm long in whole-mount preparations.

DISCUSSION

All specimens of the genus *Proctoeces* found in Japan were taken from hosts at the Pacific coast of Honshu and at Seto Inland Sea, except two cases from Hokkaido as shown in Figure 1 (Fujita, 1925; Yamaguti, 1934, 1938, 1940; Manter and Pritchard, 1962; Ichihara, 1965, 1968; Sakaguchi et al., 1970a, b; Sakaguchi, 1972; Shimazu, 1972, 1979; Shimura and Egusa, 1979a). Although the host *Batillus cornutus* is widely distributed, Ichihara (1965) reported that *Proctoeces* sp. (= *P. ichiharai*) was confined to the coastal region of Chiba and Kanagawa Prefecture. The more distinct localization was reported on *P. subtenuis* in *Scrobicularia plana* in the Thames Estuary (White, 1972). Such localization may be ascribed to the result of the passive mode of dispersal of the parasites, and the inactive intermediate and final hosts that are largely benthic and sedentary animals.

The period of survival of *Proctoeces* species in fish hosts is limited to less than 1 month under experimental conditions (Freeman, 1962; Sakaguchi et al., 1970b) and is very likely the same in natural infections (Freeman, 1962). However, *P. ichiharai* survived at least 14 months in the renal coelom of the top shell in the following experiment: 4 samples of top shell collected from Misaki (67–77 mm high) and 5 from Kominato (51–72 mm high) were tagged and kept in the same cage hanging in running water in an aquarium at the Misaki Marine Biological Station in August 1974. Marine algae such as *Eisenia bicyclis* and *Ecklonia cava* were fed at certain intervals. All top shells were dissected and examined for parasites after 14 months in October 1975. Since no parasite was found in the top shells from Kominato, the possibility of infection during this period of experiment is excluded. Each top shell collected from Misaki harbored 1 or 2 parasites. The parasites were large and measured 4.6–6.0 mm (n = 7) after being fixed in hot formalin. This scarcity of parasites may indicate that the fully matured worms had died during the experiment and the immature or maturing parasites grew and survived for 14 months. Some dead, preserved specimens of *P. subtenuis* were found in the kidney region of *Scrobicularia plana* (Freeman and Llewellyn, 1958; White, 1972) and dead *P. maculatus* were also recovered from *Mytilus edulis* (Lang and Dennis, 1976), although no dead parasite was found in the renal coelom of any top shell examined. Dead or moribund worms may have left through the external orifice or decomposed in the coelom.

The ecological analysis of the present study suggests a long life for *Proctoeces ichiharai*. From Table 1 is apparent that the larger top shells must have been exposed to infection for a longer period of time and accumulated parasites one after another in their renal cavity resulting in heavier infection. Moreover, the parasites in the renal cavity showed a high frequency of larger worms as a result of long-term continuous growth (Table 3, Fig. 2). In addition, the presence of few juveniles and a large number of mature worms throughout the year (Fig. 3) indicates that the period between larval and mature stage must be short compared with that between mature and 'post-adult' stage.

This agrees with the observation and the consideration by Lang and Dennis (1976).

In this study, *Proctoeces ichiharai* showed no seasonality in the incidence and intensity of infection and in size-frequency distribution. *P. subtenuis* parasitic in *Scrobicularia plana* in the Thames Estuary also revealed no seasonal fluctuation in the level of infection (White, 1972). But Lang and Dennis (1976) reported that the incidence of infection of adult *P. maculatus* in *Mytilus edulis* peaked in early winter and rapidly declined with no worms being recovered by late May. According to Loos-Frank (1969), *P. scrobiculariae* taken from *S. plana* in the Weser Estuary apparently reduced their sexual activity, since diminished testes and absence of embryonated eggs in the latter half of the winter were noted. However, almost all specimens of *P. ichiharai* examined were fully mature throughout the season.

Sakaguchi (1972) reported that the metacercaria of *Proctoeces ostreae* infected not only pearl oysters but also almost all benthic bivalves and gastropods in and around the pearl-oyster beds were affected by the trematode. On the other hand, Loos-Frank (1969) recorded a high host-specificity in *P. scrobiculariae* which infected only *Scrobicularia plana*. As far as inferable from the present investigation, *P. ichiharai* infects only archaeogastropods; it therefore might have a relatively high host-specificity.

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