

CERCARIAL INFECTION IN *PALUDOMUS PETROSUS*, FRESH-WATER SNAIL IN PA LA-U WATERFALL

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Abstract. *Paludomus petrosus*, the freshwater snails found in Pa La-U Waterfall, were examined for cercarial infection of trematodes. The snails were collected every other month from April, 2001 to February, 2002. Collections were taken from two sampling stations. The counts per unit of time method was used for collection of the snails. The density of snails was highest in June 2001, and the highest of parasite infection rate was in February 2002. Four types of cercariae were found in the snails: Xiphidiocercariae, Amphistome, Furcocercous cercariae type I, and Furcocercous cercariae type II. Xiphidiocercariae were found in April 2001 to February 2002. Amphistome, Furcocercous cercariae type I and Furcocercous cercariae type II were found in February 2002.

INTRODUCTION

This paper outlines a study of cercariae living on intermediate host snails. One or more species of cercariae, sometimes more than ten, may be found in freshwater and terrestrial gastropods (Ito, 1980). *Paludomus petrosus* (Fig 1) is one of the freshwater snails found in the mountains of Southern Thailand. It inhabits several mountain creeks in the provinces of Chumphon, Ranong, Nakhon Si Thammarat and Phatthalung. This species is common in areas where *Paragonimus westermani*, one of the animal lung flukes that live in carnivores. A number of specimens of this species have been examined for cercariae. No cercariae of infectious trematodes have been reported in man (Brandt, 1974).

Paludomus petrosus was found in Pa La-U Waterfall, located in Hua Hin District, Prachuap Khiri Khan Province (Fig 2). It is one of the tourist attractions in Kaeng Krachan National Park. This complex mountain range is forested at an elevation of 300-900 meters (Tourism Authority of Thailand, 2000). The river from Pa La-U merging with Pran Buri River is an important river for both local residents in Pa La-U and Pran Buri District.

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In this study, *Paludomus petrosus* collected from Pa La-U Waterfall were examined for trematode cercarial infection. The objectives of this study were to investigate: 1). The infection rate of trematodes in *Paludomus petrosus*, and 2). The types of parasites found in *Paludomus petrosus*, in order to control and prevent the risk of infection in the Pa La-U community.

MATERIALS AND METHODS

Determining the sample sites

Pa La-U Waterfall is located in the western area of Kaeng Krachan National Park, which stretches to the Tanaosri Range. Thus, several important creeks originating in the Tanaosri Range flow into the plains area. Pa La-U Waterfall was chosen because it is a popular tourist attraction. The initial survey determined the locations for survey points where we would collect samples of snails and record the geographic position by GPS (Garmin Taiwan model GPS 12 XL). The two sampling sites were: sampling site 1, GPS: 47P 0550597, UTM 1385806; sampling site 2, GPS: 47P 0552003, UTM 1385831.

Collection of snails

The snails were collected every other month from April, 2001 to February, 2002. Collections were taken from two sampling stations at Pa La-U Waterfall. The "counts per unit of time" method, recommended by Olivier and Schneiderman

Table 1
Seasonal distribution of trematode infections in *Paludomus petrosus*.

Month/Year	Sampling site 1		Sampling site 2	
	No. of snails	No. of infected snails (%)	No. of snails	No. of infected snails (%)
April 2001	697	4 (1.08)	686	12 (2.35)
June 2001	1,171	6 (0.93)	1,013	8 (1.50)
August 2001	1,174	6 (0.52)	840	4 (0.48)
October 2001	759	3 (0.40)	1,023	13 (1.27)
December 2001	995	16 (1.61)	1,173	57 (4.86)
February 2002	979	11 (1.12)	1,340	133 (9.93)

(1956), was used in this study. Five collectors hiked upstream, collecting samples of freshwater snails by hand picking for 20 minutes per sampling. The samples were brought back to the laboratory to be examined for cercarial infection.

Examination for parasitic infection

All snails were investigated for parasitic infection. Using the shedding method, each snail was isolated in a bottle about half-filled with dechlorinated water. Cercariae were collected from the water and from crushing the snails, then mounted and noted for their swimming behaviors. The occurrence of sporocysts and/or rediae was examined under dissection microscope.

Study of cercarial morphology

In order to study cercarial morphology, a few cercariae were removed from the water with a pipette and soaked with Schaudinn's fixative for 15 minutes. Then they were washed for five minutes each with solutions of 50% alcohol, 70% alcohol, 0.5% Lugol's iodine in 70% alcohol, 3% sodium thiosulphate and 50% alcohol. The cercariae were dyed with borax carmine for 30 minutes and washed with distilled water. We used 70% alcohol for 10 minutes, with 95% alcohol twice for 20 minutes, each time, to draw out the water. The subject material was made transparent by immersing it in xylene solution for 10 minutes. Finally, the test material was transferred to albumin-coated glass slides, DPX was added and the slides were covered with a cover slip.

RESULTS

Microhabitat of the sampling sites

The first site was a small waterfall approximately 600 meters away from the parking lot. The current is somewhat swift. Medium-sized trees and shrubs grow on the banks and small to medium-sized rocks lie all over the creek bed. The light intensity is about 600-850 lux. This location is about 312 meters above sea level.

The second site was approximately 1,500 meters downstream from the first sampling site. The current is somewhat swift. Dried leaves and small to medium-sized rocks are found everywhere with a few large rocks. Medium-sized trees and shrubs cover the banks. The light intensity is 450-700 lux and the location is about 292 meters above sea level.

The trematode infection rate in the snails

The density of snails was highest in June 2001, and the highest parasitic infection rate was in February 2002 (Table 1).

Identification of cercariae

Four types of cercariae were found in the *Paludomus petrosus* from Pa La-U Waterfall (Fig 3). Morphology was investigated from the body parts of cercariae. In the period April 2001 to February 2002, we found Xiphidiocercariae (Fig 3D). The cercariae have a stylet at the anterior margin of the oral sucker. The cercariae swim rapidly, bending themselves around for about 55 seconds

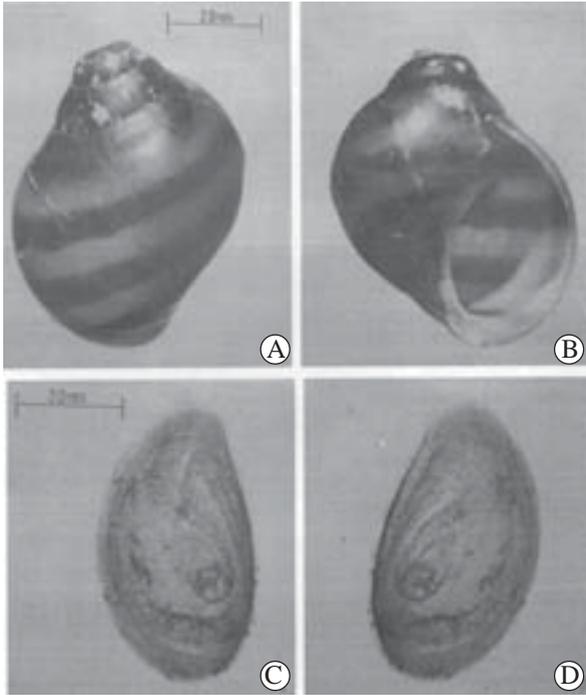


Fig 1—*Paludomus petrosus*, freshwater snail is found in Pa La-U Waterfall.

then stretching out for 2-3 seconds.

The measurements are as follows: body=114-138 (av. 126) x 54-76 (av. 68) μ m, tail=54-82 (av. 76) x 17-20 (av. 18) μ m, oral sucker=17-25 (av. 21) x 10-18 (av. 14) μ m, ventral sucker=17-28 (av. 24) x 12-19 (av. 17) μ m, stylet=10-15 (av. 13) x 4-10 (av. 5) μ m.

In February 2002, we found four types of cercariae: Xiphidiocercariae, Amphistome cercariae, Furcocercous cercariae type I and Furcocercous cercariae type II.

Amphistome cercariae (Fig 3C) possess a large ventral sucker situated at the posterior margin of the body. The oral sucker is small, the body is large and the tail is simple and globular. While swimming, they bend themselves back and forth and keep their tails short. After swimming for about 50-55 seconds, they stretch for 4-5 seconds.

The measurement are as follows: body=208-275 (av. 243) x 67-94 (av. 78) μ m, tail=104-144 (av. 136) x 48-66 (av. 54) μ m, oral sucker=42-56 (av. 45) x 27-37 (av. 32) μ m, ventral sucker=48-60 (av. 53) x 32-41 (av. 38) μ m.

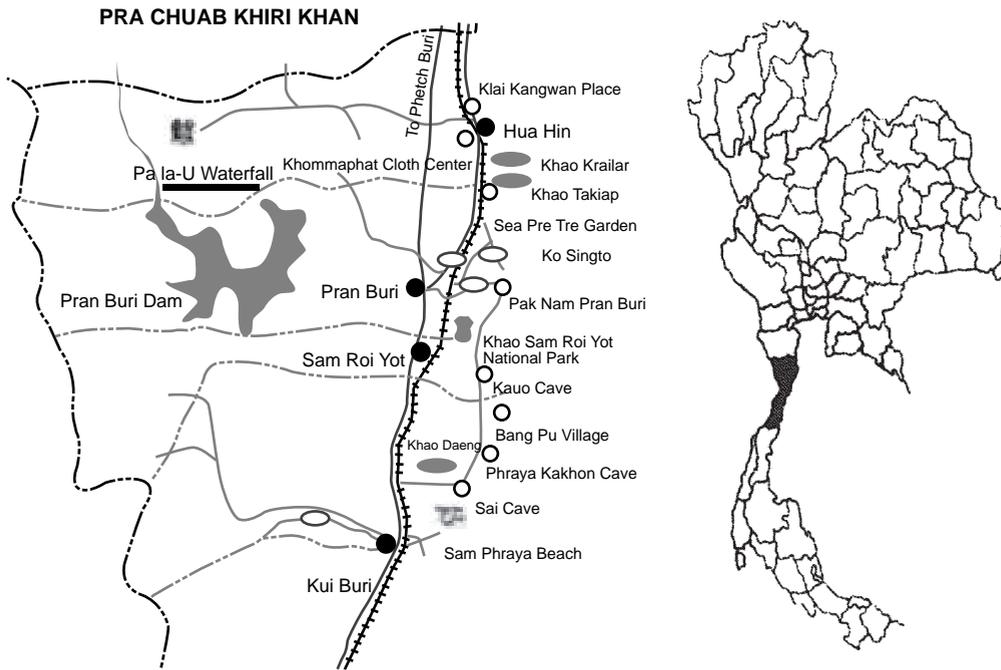


Fig 2—Map of Pa La-U waterfall in Prachuab Kriri Khan Province, Thailand.

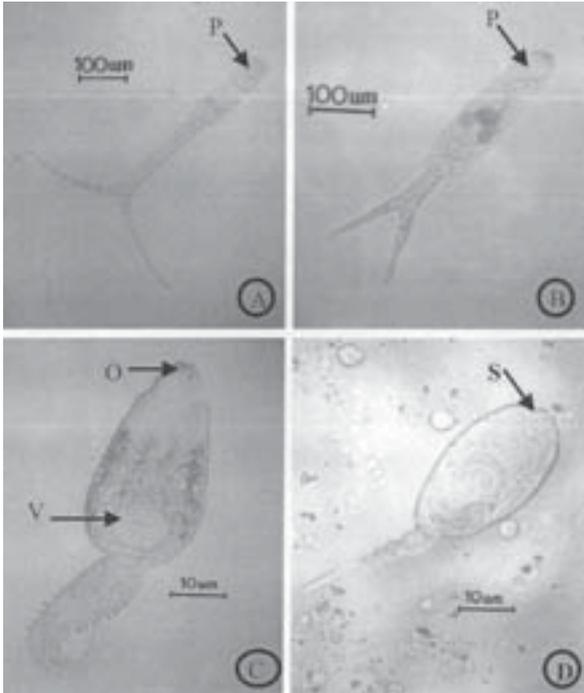


Fig 3—Cercariae are found from *Paludomus petrosus* in Pa La-U Waterfall.

- A. Furcocercous cercariae type I (P = pharynx).
- B. Furcocercous cercariae type II (P = pharynx).
- C. Amphistome cercariae (O = oral sucker, V = ventral sucker).
- D. Xiphidiocercariae (S = stylet).

Furcocercous cercariae (Fig 3A,3B) are forked-tail cercariae with tails terminating as a bifurcation. These cercariae are generally distomate, with two suckers, an anterior oral sucker and a ventral acetabulum. There are several families of trematodes possessing Furcocercous cercariae. Both Furcocercous cercariae types I and type II are strigedae cercariae. These cercariae are distomate and a muscular pharynx is present. We differentiated these two types of cercariae by their fork-tail lengths. Type I Furcocercous cercariae have longer tails (Fig 3A) and are good swimmers. They swim along in the water for 1 minute, then rest for 3-4 seconds.

The measurement are as follows: body=249-297 (av. 278) x 89-132 (av. 117) μm , tail=224-289 (av. 252) x 52-78 (av. 63) μm , fork tail=228-261 (av. 246) x 27-46 (av.38) μm , oral sucker=31-

42 (av. 38) x 20-27 (av. 24) μm , ventral sucker=39-45 (av. 42) x 31-40 (av. 36) μm .

Type II Furcocercous cercariae (Fig 3B) tails are slightly shorter than Type I. These cercariae are characterized by the presence of a muscular pharynx. They swim very fast, for 40-45 seconds, then stretch themselves and float for about 2-4 seconds.

The measurement are as follows: body=312-347 (av. 329) x 129-168 (av. 148) μm , tail=52-68 (av. 61) x 46-55 (av. 49) μm , fork-tail=87-101 (av. 93) x 41-66 (av.57) μm , oral sucker=25-37 (av. 31) x 19-28 (av. 24) μm , ventral sucker=33-47 (av. 44) x 27-36 (av. 32) μm .

DISCUSSION

Few studies of the incidence of cercarial infection in snails have been done, in spite of comprehensive surveys in Thailand, especially of *Paludomus* snails. Pa La-U Waterfall is a tourist attraction; moreover, PaLa-U is an important river for the local residents in Prachuap Khiri Khan Province. These study results can be used as a knowledge base to improve the quality of life for people living in the region.

Though the counts per unit of time method (Olivier and Schneiderman, 1956) measures the density of the snail population in the marked area only, not the total population, it is statistically reliable. The infection rate of parasites was investigated by the shedding and crushing method. One disadvantage of the crushing method, as far as identification is concerned, is that some of the cercariae obtained by this manner may be immature and are therefore unsuitable for identification. This study found infection of trematodes in the snails every season. The infection rate rose to 9.93% during the rainy season in the second sampling site.

A variety of cercariae types exist. A detailed description of the various types can be found in Cheng's report (1973). Some are classified according to the position and number of body suckers. Some are categorized according to the shape and relative size of their tails, while some cercariae are categorized morphologically by specialized body structures like the Xiphidiocer-

cariae, the stylet-bearing cercariae. The various species have one characteristic in common, they all have an anterior margin of the oral sucker. The Xiphidiocercariae are poor swimmers and are often found creeping or attached with their ventral suckers and lashing their tails in all directions (Malex and Cheng, 1974). Xiphidiocercariae found in *Paludomus petrosus* in this study swim rapidly; they have a special swimming pattern. Amphistome cercariae is one of the group of cercariae found in Pa La-U Waterfall. This group possesses a large ventral sucker situated at the posterior margin of the body. They develop in rediae, encyst on aquatic vegetation on the bottom of bodies of water or on the skin of frogs, which almost always possess eyespots (Malex and Cheng, 1974). However, there were no eyespots found on the cercariae in this study. Furcocercous cercariae were differentiated by the length of their fork-tails. The bifurcate tail of Furcocercous cercariae type I is 246 μm , while that of type II is 93 μm ; both are strigeidae cercariae, which distinguishes them from the other group by the presence of a pharynx. Malex and Cheng (1974) described strigeid cercariae as distomate with a muscular pharynx present. Their stout tails terminate as two elongated furcae. When these cercariae enter a suitable second intermediate host, they develop into encysted or nonencysted metacercariae. Encysted metacercariae usually occur in the muscles of fish, while unencysted metacercariae occur in the cranial cavity or eyes of

fishes, amphibians and reptiles.

This is a preliminary study conducted on the morphology of cercariae found in *Paludomus* snails in Thailand. Further study of the flame cell pattern and other morphological features of larval forms is recommended.

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