GNATHOSTOMIASIS IN THAILAND : A SURVEY ON INTERMEDIATE HOSTS OF *GNATHOSTOMA* SPP. WITH SPECIAL REFERENCE TO A NEW TYPE OF LARVAE FOUND IN *FLUTA ALBA*

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Abstract. To clarify current status of gnathostomiasis in Thailand, a survey on intermediate hosts has been carried out at various localities since 1987. It was found that *Fluta alba* (Fresh water eel) as well as *Channa striata* (snake-headed fish) might be important in playing a role of transmitting the infection either among humans or reservoir animals. During the three years from 1987 to 1989, larvae of *Gnathostoma spinigerum* were found in 80-100% of *F. alba* obtained from markets in Nakhon Nayok, with a maximum recovery of 2,582 larvae per eel. Among larvae found in these eels, five were peculiar in possessing four rows of hooklets with complicated branches at the base. Epithelial cells of the intestine of these larvae contained 1-2 nuclei. These observations indicate that the larvae are different from those of reported species of *Gnathostoma* from Thailand including *G. spinigerum*, suggesting a possibility of the advanced third-stage larvae of *G. malaysiae*.

INTRODUCTION

Gnathostomiasis is one of the most important parasitic diseases in Thailand. More than 800 suspected cases per year visited gnathostomiasis clinics at two major hospitals in Bangkok from 1985 to 1988 (Setasuban, 1990). *Gnathostoma spinigerum* is the only species causing human infection of the five species of *Gnathostoma* reported from Thailand (Daengsvang, 1980). In addition to this particular species, cases of infection with *G. hispidum* (Tsushima *et al,* 1980; Nishimura *et al,* 1981; Akahane *et al,* 1986), *G. doloresi* (Ogata *et al,* 1988), and *G. nipponicum* (Ando *et al,* 1988) were reported from Japan. Therefore, possible human infections, not only with *G. hispidum* or *G. doloresi* (Dissamarn *et al,* 1966) but with the other species reported from Thailand, such as *G. vietnamicum* (Daengsvang, 1973) or *G. malaysiae* (Kamiya *et al,* 1987) should be carefully studied. To clarify the current status of the disease, a survey on intermediate hosts has been carried out at various localities in Thailand since 1987. It was found that *Fluta alba* (fresh water eel) as well as *Channa striata* (snake-headed fish) might be important in transmitting the infection either among humans or reservoir animals. Moreover, among larvae obtained from eels, five were found to be peculiar in possessing four rows of hooklets with complicated branches at the base and intestinal epithelial cells containing 1 - 2 nuclei. The observation indicates that the larvae are different from those of reported species of *Gnathostoma* from Thailand, including *G. spinigerum*, suggesting a possibility of the advanced third-stage larvae of *G. malaysiae*.

MATERIALS AND METHODS

During the period October 1987 to August 1989, various number of three species of freshwater fish, *Channa striata* (snake-headed fish), *Clarias batrachus* (catfish) and *Fluta alba* (fresh water eel),were obtained from markets in Bangkok, Nakhon Nayok, Prachin Buri, and Chiang Mai. Edible frogs, *Rana rugulosa*, were purchased at a market in Nakhon Nayok. Advanced third-stage larvae were collected from the muscle and viscera

of the freshwater fish or frogs already known as the second intermediate host of *Gnathostoma spinigerum* in Thailand (Daengsvang, 1980). The larvae were used for morphological observations and experimental infections in cats and dogs.

RESULTS

Infection rates of *Gnathostoma* larvae among second intermediate hosts are summarized in Table 1. *Channa striata* showed a stable infection rate of around 70% irrespective of the lacality from which the snake-headed fish was obtained. On the other hand, 80-100% of *Fluta alba* from Nakhon Nayok and Prachin Buri were found to be infected with *Gnathostoma* larvae, showing a higher infection rate than in eels from the other localities. Nearly half of *Rana rugulosa* from Nakhon Nayok was also infected.

Infection rates and the number of recovered worms per fish were examined according to the size of fish obtained from Nakhon Nayok. The longer the body length the higher the infection rate observed, but there was no tendency that recovered worms increased in number among fish with a larger size (Tables 2 and 3). Similar results were obtained for fish purchased in the other places.

Frequency distribution of *Fluta alba* harboring different numbers of *Gnathostoma* larvae showed a good correlation with negative binomial distribution (Fig 1).

Most (80-90%) *Gnathostoma* larvae were found in the muscle in *Channa striata* and *Rana rugulosa*, whereas in *Fluta alba*, 53-65% of the larvae (depending on the year of the examination) were recovered from the muscle, indicating a relative concentration of larvae in the liver, if compared with their tissue distribution in the two former intermediate hosts. Typical results are shown in Fig 2. Thus, the larvae were found mainly in the abdominal side and in the central one-third of the muscle of freshwater eels (Fig 3).

Morphological observations revealed that all of the recovered worms, except five as mentioned below, were advanced third-stage larvae of *G. spinigerum*. This was further confirmed by recovering adult worms of this species from experimen-

Table 1

Infection rate of *Gnathostoma* larvae among second intermediate hosts in Thailand.

Host species and localities	Date of examination	No. examined	No. positive (%)
Channa striata			
Bangkok	1987.10	7	5 (71.4)
Nakhon Nayok	1987.11	29	21 (72.4)
	1988.8	19	13 (68.4)
Clarias batrachus			
Bangkok	1987.10	5	0 (0)
Fluta alba			
Bangkok	1988.8	15	4 (26.7)
	1989.8-9	102	23 (22.5)
Nakhon Nayok	1987.11	4	4 (100.0)
	1988.8	30	24 (80.0)
	1989.9	33	27 (81.8)
Prachin Buri	1989.9	13	13 (100.0)
Chiang Mai	1989.8	26	8 (30.8)
Rana rugulosa			
Nakhon Nayok	1988.8	24	11 (45.8)

Table 2

Infection rate and the number of *Gnathostoma spinigerum* larvae per fish according to size of snake-headed fish, *Channa striata**

Body length (cm)	No. fish examined	No. positive	Infection rate (%)	Mean no. larvae/fish
31-35	11	5	45.5	2.2
36-40	13	11	84.6	6.8
41-45	3	3	100.0	6.0
46-50	2	2	100.0	51.0

* Obtained from Nakhon Nayok in 1987.

Table 3

Infection rate and the number of *Gnathostoma spinigerum* larvae per fish according to size of fresh water eel, *Fluta alba**

Body length (cm)	No. fish examined	No. positive	Infection rate (%)	Mean no. larvae/fish
45-60	8	7	87.5	27.7
61-65	9	7	77.8	66.9
66-70	19	14	73.7	43.8
71-75	17	14	82.4	15.1
76-80	6	5	83.3	12.8
81-95	4	4	100.0	43.8

* Obtained from Nakhon Nayok in 1988 and 1989.

tally infected dogs and cats (data not shown). In addition to *G. spinigerum* larvae, we found five larvae distinct from any of already known third-stage larvae of four species of *Gnathostoma* (*G. spinigerum, G. hispidum, G. doloresi, G. vietnamicum*) distributed in Thailand in terms of morphology and number of hooklets. These larvae were obtained from *Fluta alba* purchased in Nakhon Nayok in 1988. Their hooklets showed complicated branching at the base. The mean number of hooklets in the fouth row was 55.5, whereas that of other species ranged from 35.7 to 49.8. Hooklets in the first to third row of these newly found larvae were also larger in number than those of the other known species. Cross sections of the larvae revealed that epithelial cells of the intestine contained 1-2 nuclei.

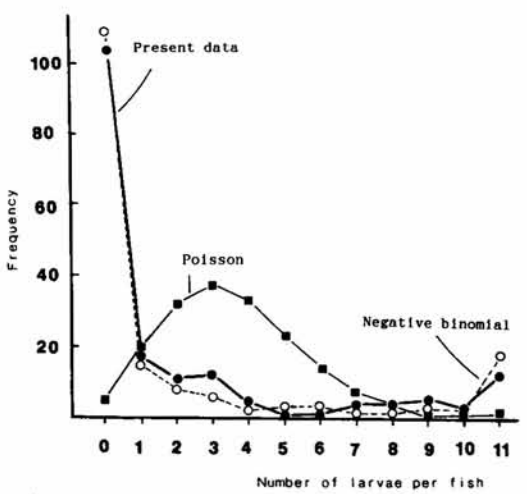
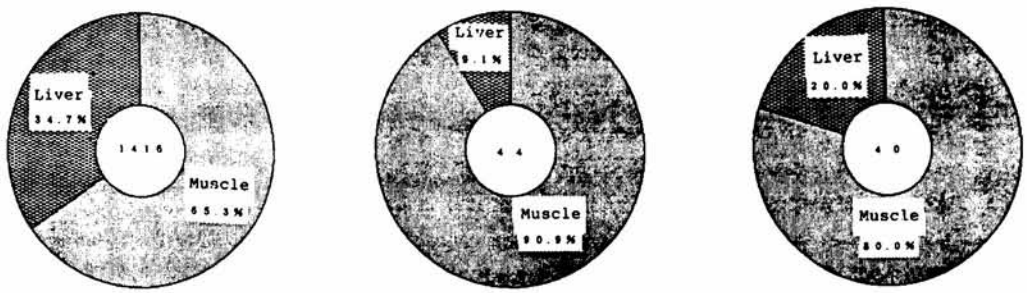


Fig 1—Frequency distribution of *Fluta alba* harboring different numbers of *Gnathostoma* larvae. Standard curves for Poisson distribution and negative binomial distribution are also shown in the figure.

DISCUSSION

The present results indicate that *G. spinigerum*



Fluta alba *Channa striata* *Rana rugulosa*

Fig 2—Comparison of the tissue distribution of *Gnathostoma* larvae in the second intermediate hosts. Intermediate hosts were obtained from Nakhon Nayok in 1988.

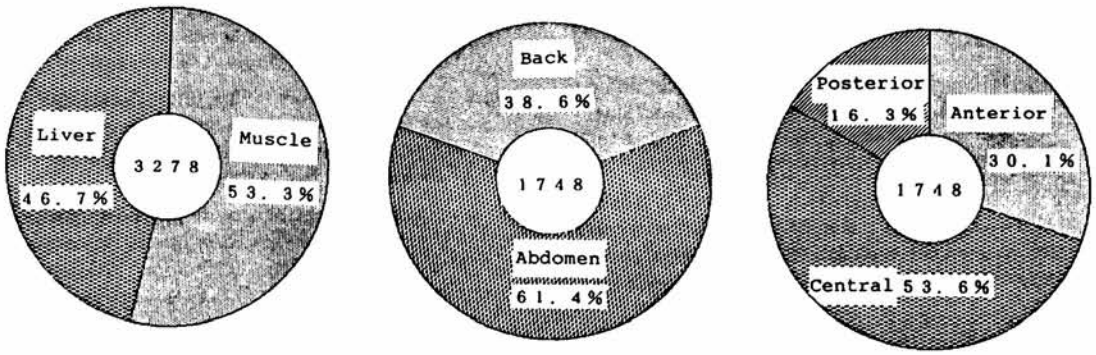


Fig 3—Tissue distribution of *Gnathostoma* larvae in *Fluta alba*. Fresh water eels were obtained from Nakhon Nayok in 1987. Two diagrams on the right side demonstrate distribution of the larvae in the muscle according to body regions.

has been widely distributed throughout Thailand. *Fluta alba*, as well as *Channa striata*, might be very important in transmitting the infection either among humans or reservoir animals (Table 1). It seems likely that the transmission pattern of this parasite in Nakhon Nayok is different from that in other localities, because a higher infection rate of *Gnathostoma* larvae was observed among eels from Nakhon Nayok.

The infection rate increased according to size of fish whereas the number of worms did not increase (Tables 2 and 3), suggesting that the infection of fish might occur at any special spot but not uniformly. This was also confirmed by the fact that the frequency distribution of *Fluta alba* in relation to the number of recovered larvae was well correlated with negative binomial distribution (Fig 1).

Since *Gnathostoma* larvae were found to be concentrated in the liver of fresh water eels (Fig 2), the disposal of viscera should be carefully controlled in order to avoid transmitting a new infection to dogs and cats.

Finally, five larvae of unknown species were obtained from eels purchased in Nakhon Nayok. The larvae were peculiar in possessing hooklets with complicated branches at the base. The larvae were also unique in the fact that the hooklets were larger in number in the fourth row on the head bulb than in any of the third-stage larvae of the species reported from Thailand. Moreover, epithelial cells of the intestine of the newly found larvae contained 1-2 nuclei, which were distinguishable from *G. spinigerum* larvae containing 3-7 nuclei in an epithelial cell (Akahane *et al*, 1986). Thus, it is possible that the newly found larvae may be those of *G. malaysiae* (Nuamtanong *et al*, 1989), because adult worms of this particular species were

described from *Rattus surifer* caught in Khao Yai National Park close by Nakhon Nayok (Kamiya *et al*, 1987). However, this does not exclude another possibility that these larvae may belong to an undescribed new species. Further studies are necessary to clarify these points.

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