GNATHOSTOMA INFECTION IN FISH CAUGHT FOR LOCAL CONSUMPTION IN NAKHON NAYOK PROVINCE, THAILAND I. PREVALENCE AND FISH SPECIES

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Abstract. Between August 2000 and August 2001, 12,216 fish of 73 species were purchased from several local markets in Nakhon Nayok Province, Thailand, and examined for the presence of *Gnathostoma* larvae. Almost all species were fresh-water fish that had grown naturally, rather than raised commercially. Eight species were found to be infected with gnathostome larvae. The overall prevalence was 5.1% (626/12,216) and a total of 5,969 larvae was recovered. The highest rate of infection (30.1%) was found in *Monopterus albus* (swamp eel). The rates in the remaining infected fish were as follows: *Anabas testudineus* (climbing perch) 7.7%, *Channa striata* (striped snake-head fish) 7.4%, *Clarius macrocephalus* (Gunther's walking catfish) 6.7%, *Channa micropeltes* (giant snake-head fish) 5.1%, *Channa lucius* (blotched snake-head fish) 4.0%, *Clarius batrachus* (Batrachian walking catfish) 1.4%, and *Ompok krattensis* (butter sheatfish) 0.6%. The mean number of larvae/fish was highest in swamp eels (10.0 larvae/eel), and the maximum number of 698 larvae was recovered from one eel. The body sizes of the recovered *G. spinigerum* advanced third-stage larvae were 2.70-5.10 mm in length (average, 3.97 ± 0.50 mm) and 0.29-0.60 mm in width (average, 0.40 ± 0.04 mm). The average number of cephalic hooklets of the larvae from rows 1 to 4 were 41.8 ± 0.5 (range, 40-43), 43.6 ± 0.6 (range, 42-45), 46.1 ± 0.9 (range, 44-48) and 49.3 ± 0.7 (range, 48-51), respectively.

INTRODUCTION

Gnathostomiasis, caused by roundworms of the genus *Gnathostoma*, is a serious helminthic disease that has posed major health problems among the Thai people for a long time. This is not only because of the constantly high incidence of infection, but also the ineffective results of medical treatment (Daengsvang, 1980, 1986; Chitchang, 1987; Setasuban, 1990; Miyazaki, 1991; Suntharasamai *et al*, 1992; Kraivichian *et al*, 1992). The infection is primarily acquired by eating raw or improperly cooked meat, especially fish, leading to the clinical syndrome, cutaneous and/or visceral larva migrans. Cases of human gnathostomiasis have been suspected in different parts of the country, but mostly from central Thailand (Daengsvang, 1986; Rojekittikhun, 2002a).

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Gnathostomiasis is considered an important re-emerging parasitic disease across the continent (Bravo and Sanchez, 2003). Not only has it occurred in Asian countries, especially Thailand and Japan (Daengsvang, 1980; Miyazaki, 1991; Nawa, 1991), and in Latin America, *eg* Mexico and Ecuador (Ollague *et al*, 1984; Diaz-Camacho *et al*, 1998, 2003; Ogata *et al*, 1998; Rojas-Molina, 1999; Almeyda-Artigas, 2000; Nawa, 2002), sporadic cases have been reported in many other countries. Infected people going to live/work abroad (Hira *et al*, 1989; Grobusch *et al*, 2000; Germann *et al*, 2003), emigrants acquiring the infection in endemic countries (Chai *et al*, 2003) and overseas travelers suffering from the disease after returning home (Rusnak and Lucey, 1993; Jelinek *et al*, 1994; Chappuis *et al*, 2001; de Vries *et al*, 2001; Del Giudice *et al*, 2001; Lo Re and Gluckman, 2002; Puente *et al*, 2002; Gorgolas *et al*, 2003; Hale *et al*, 2003; Menard *et al*, 2003; Moore *et al*, 2003), were found to be increasing.

In Thailand, there are five species of *Gnathostoma*, of which mature worms have been natu-

rally recovered from the stomachs and urinary systems of animals. These are: 1. *G. spinigerum* Owen 1836 (Prommas and Daengsvang, 1933), 2. *G. hispidum* Fedtschenko 1872 (Dissamarn *et al*, 1966), 3. *G. doloresi* Tabangui 1925 (Dissamarn *et al*, 1966), 4. *G. vietnamicum* Le-Van-Hoa 1965 (Daengsvang, 1973), and 5. *G. malaysiae* Miyazaki and Dunn 1965 (Kamiya *et al*, 1987). However, only *G. spinigerum* has been proven to cause human gnathostomiasis in Thailand (Daengsvang, 1980, 1986; Radomyos and Daengsvang, 1987; Sirikulchayanonta and Viriyavejakul, 2001; Rojekittikhun, 2002a).

G. spinigerum is a three-host parasite; its life cycle involves three essential hosts: first intermediate hosts (mainly fresh-water cyclops), second intermediate hosts (sources of infection for the definitive host and man), and definitive hosts (normally cats and dogs). In Thailand, the second intermediate (and/or paratenic) hosts are various species of vertebrates, 48 species of which have been reported to be naturally infected with *G. spinigerum* advanced third-stage larvae. These include 20 species of fish, two species of amphibians, 11 species of reptiles, 11 species of avians and four species of mammals (Daengsvang, 1980; Rojekittikhun *et al*, 1989a,b, 2002b; Rojekittikhun, 2002b).

Although there have been several reports concerning the prevalence and intensity of *Gnathostoma* infection in swamp eels in some provinces of Thailand (Setasuban *et al*, 1991; Nuamtanong *et al*, 1998; Rojekittikhun *et al*, 1998a,b, 2001, 2002b; Saksirisampant *et al*, 2002a,b; Sugaroon and Wiwanitkit, 2003), there is only one report on the infection in various species of fish sold in local markets in Bangkok (Rojekittikhun *et al*, 1989a). The purpose of this study is, therefore, to investigate the prevalence and intensity of the infection in any species of fish caught for local consumption, and also to identify the fish species that serve as intermediate/paratenic hosts of the worm in Nakhon Nayok Province, central Thailand, where gnathostomiasis is endemic.

MATERIALS AND METHODS

Fish

All species of fish and eels (mostly fresh-water) that had grown naturally, rather than reared commercially, were purchased 2-3 times a month from several local markets in Nakhon Nayok Province, between August 2000 and August 2001. After purchase, live fish were kept in a container containing an appropriate amount of water; dead fish were separated and kept in an icebox. They were immediately transported to the laboratory of the Department of Helminthology, Faculty of Tropical Medicine, Mahidol University, in Bangkok. Live fish were then transferred into an aquarium; dead ones were kept in a freezer. Before being subjected to the next preparation, each fish was identified to species, weighed, measured, and recorded.

Press preparation (compression) method

The visceral organs of big fish or eels were taken out. Only the liver was cut into small pieces and firmly pressed between two thick glass plates, then examined under a dissecting microscope or a large hand lens for the presence of gnathostome larvae. All muscles were cut and scraped out of bones and scales (or skins), and examined as was done for the liver. For tiny fish, the whole body was pressed and examined, as described above.

Gnathostoma larvae

The recovered gnathostome larvae were collected, cleaned, counted, and fixed in 70% ethanol. Their lengths and widths were measured using a camera lucida. Vital statistics were also calculated.

RESULTS

Twelve thousand, two hundred and sixteen fish of 73 species were purchased and examined (Table 1). Their scientific names, their Thai names, together with the range and mean ± SD of fish lengths and widths, are shown in Table 1. Apart from swamp eels (*Monopterus albus*, up to 97.0 cm in length) and armed spiny eels (*Mastacembelus armatus*, up to 68.0 cm in length), the largest examined fish was *Channa striata* (striped snakehead fish, 78.0 cm in length and 3,600 g in body weight). However, on average, the longest fish was the four *Labeo rohita* (rohu, 48.3 ± 12.6 cm) and the heaviest was the five *Osphronemus goramy* (giant gourami, 760.0 ± 163.6 g). Except for swamp eels, which will be reported in detail elsewhere, the highest number of examined fish (778) was a

tiny *Brachydanio albolineata* (blue danio). There were nine species, for each of which only one specimen was available, and three species with two specimens (Table 1).

Eight species were found to be infected with gnathostome larvae. The overall prevalence was 5.1% (626/12,216) and a total of 5,969 larvae was recovered (Table 2). The highest rate of infection, 30.1% (555/1,844), was found in swamp eels (*M. albus*). The rates in the remaining infected fish were as follows: climbing perch (*Anabas testudineus*) 7.7% (28/365), striped snake-head fish (*C. striata*) 7.4% (17/231), Gunther's walking catfish (*Clarius macrocephalus*) 6.7% (11/165), giant snake-head fish (*C. micropeltes*) 5.1% (5/98), blotched snake-head fish (*C. lucius*) 4.0% (7/175), Batrachian walking catfish (*Clarius batrachus*) 1.4% (2/137), and butter sheatfish (*Ompok krattensis*) 0.6% (1/165) (Table 2). The mean numbers of larvae per fish of the seven infected species are shown in Table 2. The highest number of larvae (698) was recovered from one swamp eel (average, 10.0 larvae/eel) and the lowest (only one) was observed in *O. krattensis*.

The body sizes of the recovered *G. spinigerum* advanced third-stage larvae are shown in Table 3. They (n = 100) measured 2.70-5.10 mm in length (average, 3.97 ± 0.50 mm) and 0.29-0.60 mm in width (average, 0.40 ± 0.04 mm). The number of cephalic hooklets of the larvae is shown in Table 4, and the frequency and relative frequency distributions of hooklet numbers are shown in Table 5. The average number of hooklets from rows 1 to 4 were 41.8 ± 0.5 (range, 40-43), 43.6 ± 0.6 (range, 42-45), 46.1 ± 0.9 (range, 44-48) and 49.3 ± 0.7 (range, 48-51), respectively. The highest frequency of hooklet numbers in rows 1 to 4 was between 40-42 (98.0%), 43-45 (96.0%), 46-48 (76.0%) and 49-51 (89.0%), respectively. No larva having more than 45 hooklets in row 1 or having less than 46 hooklets in row 4, was found (Table 5).

DISCUSSION

Nakhon Nayok Province has a total area of 2,122 km^2, located in central Thailand, about 100 km northeast of Bangkok. This province is a paddy field area with rainfall 6-7 months of the year, and a very hot and dry season during the other 5-6 months (Environment and Sanitation Section, Nakhon Nayok Province Health Office). Some reports have indicated that Nakhon Nayok is an endemic area for gnathostomiasis. Between 1987-1989, the prevalence of infection in *M. albus, C. striata* and *Rana rugulosa* (common lowland frog) were 80.0-100%, 68.4-72.4% and 45.8%, respectively (Setasuban *et al*, 1991). The rates in eels were 68.7% in 1992 (Nuamtanong *et al*, 1998) and 38.1% in 2000 (Rojekittikhun *et al*, 2002b). However, the prevalences in cats were 1.9% in 1989, and 2.9% in 1996 (Rojekittikhun *et al*, 1996), and in dogs 1.2% (Rojekittikhun *et al*, 2000).

In the present study, 73 species of fish were obtained for study, implying the natural fertility of Nakhon Nayok Province. The overall prevalence was only 5.1%; this may be because large numbers of many uninfected fish species were caught from rivers and canals. According to our experience, fish from swamps or ponds, especially in paddy fields, tend to be infected with *Gnathostoma* at much higher rates and with greater intensity. A more confined area, where the host and agent were readily there, might enhance infection.

Eight species of fish in Nakhon Nayok, which were found to be infected with *Gnathostoma*, have already been seen on the list of natural second intermediate hosts of the worm (Daengsvang, 1980; Rojekittikhun, 2002b). All species are common and favorite food fish for native Thais. However, in terms of availability in almost all markets, the two most common species are considered to be *C. striata* and *C. macrocephalus*.

The prevalence in swamp eels was still the highest. This is similar to previous reports, although the infection intensity was much lower (Daengsvang *et al*, 1964; Rojekittikhun *et al*, 1989a; Setasuban *et al*, 1991). Among the eight infected species, the butter sheatfish feeds on both live and decaying animals and plants. Eels, snake-head fish and catfish are carnivorous, and the chance of becoming more and heavily infected is, therefore, increased (Rojekittikhun *et al*, 2002a). The second highest prevalence was found in *A. testudineus*. We surprisingly obtained one

Table 1
Seventy-three species of fish examined for *Gnathostoma* larvae.

No.	Fish scientific name (Thai name)	No. examined	Length (cm)		Weight (g)	
			Range	Mean ± SD	Range	Mean ± SD
1	*Anabas testudineus* (Pla mo Thai)	365	5.0 - 15.0	9.1 ± 2.3	10 - 120	50.2 ± 23.3
2	*Bagriichthys macropterus* (Pla khayaeng mu)	13	8.0 - 13.0	10.5 ± 2.2	45 - 140	98.5 ± 40.9
3	*Barbodes altus* (Pla taphian thong)	81	2.5 - 21.0	8.3 ± 8.0	8 - 100	36.0 ± 36.0
4	*Barbodes gonionotus* (Pla taphian khao)	284	2.5 - 25.0	6.1 ± 6.7	10 - 110	26.2 ± 36.2
5	*Barbodes schwanenfeldi* (Pla krahae thong)	10	10.0 - 25.0	17.0 ± 4.7	10 - 120	75.5 ± 38.3
6	*Boesemania microlepis* (Pla ma)	1	36.5		750	
7	*Botia modesta* (Pla mu khao)	1	8.0		20	
8	*Brachydanio albolineata* (Pla siu bai phai)	778	3.0 - 5.0	4.3 ± 1.0	1 - 2	1.8 ± 0.5
9	*Catlocarpio siamensis* (Pla kraho)	2	30.0 - 45.0	37.5 ± 10.6	350 - 400	375.0 ± 35.3
10	*Channa lucius* (Pla krasong)	175	21.0 - 66.0	28.3 ± 11.5	70 - 1,800	148.7 ± 185.3
11	*Channa micropeltes* (Pla chado)	98	21.0 - 75.0	34.0 ± 14.7	70 - 3,200	259.9 ± 385.1
12	*Channa striata* (Pla chon)	231	16.0 - 78.0	33.0 ± 14.5	50 - 3,600	254.6 ± 427.8
13	*Chitala ornata* (Pla krai)	36	28.0 - 66.0	36.2 ± 3.5	90 - 2,900	355.0 ± 625.3
14	*Cirrhinus microlepis* (Pla nuan chan nam chut)	2	25.0 - 30.0	27.5 ± 3.5	300 - 450	375.0 ± 106.1
15	*Clarius batrachus* (Pla duk dan)	137	20.0 - 35.0	26.7 ± 3.3	150 - 220	186.8 ± 23.4
16	*Clarius macrocephalus* (Pla duk ui)	165	19.0 - 34.0	25.5 ± 3.9	150 - 220	185.8 ± 21.3
17	*Coilia lindmani* (Pla hang kai)	1	18.0		150	
18	*Cosmochilus harmandi* (Pla takak)	1	30.0		280	
19	*Cyclocheilichthys apogon* (Pla sai tan ta daeng)	677	8.0 - 16.0	10.3 ± 2.5	50 - 100	66.2 ± 16.5
20	*Cyclocheilichthys enoplos* (Pla takok)	30	25.0 - 30.0	27.6 ± 1.9	240 - 300	258.0 ± 22.7
21	*Cyclocheilichthys repasson* (Pla sai tan ta khao)	113	8.0 - 18.0	11.6 ± 3.1	50 - 120	73.1 ± 19.6
22	*Cynoglossus microlepis* (Pla yot muang nam chut)	3	13.0 - 16.0	14.3 ± 1.5	80 - 100	86.7 ± 11.5
23	*Hampala macrolepidota* (Pla krasup khit)	113	10.0 - 38.0	21.1 ± 6.8	50 - 180	103.6 ± 43.5
24	*Helicophagus waandersii* (Pla sawai nu)	1	42.0		450	
25	*Helostoma temmincki* (Pla mo tan khieo)	16	15.0 - 19.5	16.1 ± 1.9	55 - 130	74.3 ± 33.2
26	*Hemibagrus nemurus* (Pla kot luang)	146	12.0 - 22.0	15.4 ± 2.7	60 - 200	107.8 ± 41.6
27	*Hemipimelodus bicolor* (Pla uk)	98	10.0 - 22.5	18.1 ± 2.2	60 - 200	106.5 ± 46.2
28	*Henicorhynchus siamensis* (Pla soi khao)	352	10.0 - 16.0	11.1 ± 1.4	59 - 120	72.6 ± 15.6
29	*Heterobagrus bocourti* (Pla khayaeng thong)	81	8.0 - 25.0	16.1 ± 4.9	45 - 200	147.5 ± 45.7
30	*Heteropneustes kemarattensis* (Pla chit)	3	25.0 - 30.0	27.7 ± 2.5	50 - 86	65.3 ± 18.6
31	*Kryptopterus cheveyi* (Pla pik kai)	16	13.0 - 15.0	13.6 ± 0.7	50 - 80	57.5 ± 10.0
32	*Labeo rohita* (Pla yisok thet)	4	30.0 - 58.0	48.3 ± 12.6	300 - 980	732.5 ± 298.1
33	*Labiobarbus siamensis* (Pla sa)	191	10.0 - 16.0	11.2 ± 1.4	60 - 120	72.6 ± 16.2
34	*Lates calcarifer* (Pla kapong nam chut)	9	30.0 - 52.0	39.7 ± 9.1	350 - 560	453.3 ± 80.1
35	*Leiocassis siamensis* (Pla khayaeng hin)	13	6.0 - 9.0	7.8 ± 1.6	40 - 100	71.5 ± 25.1
36	*Luciosoma bleekeri* (Pla siu-ao)	1	10.0		40	
37	*Macrognathus semiocellatus* (Pla lot lai)	109	15.0 - 25.0	16.9 ± 2.4	20 - 98	27.1 ± 15.0
38	*Macrognathus siamensis* (Pla lot chut)	50	15.0 - 20.0	16.7 ± 1.9	20 - 35	24.7 ± 6.1
39	*Mastacembelus armatus* (Pla krathing dam)	73	20.0 - 68.0	41.9 ± 11.4	90 - 920	380.4 ± 203.9
40	*Micronema apogon* (Pla nam ngoen)	19	23.0 - 35.0	27.6 ± 2.7	80 - 100	92.1 ± 6.1
41	*Micronema bleekeri* (Pla daeng)	1	24.0		80	
42	*Micronema micronema* (Pla daeng hai)	38	23.0 - 30.0	26.3 ± 1.3	80 - 100	90.3 ± 3.4
43	*Monopterus albus* (Pla lai na)	1,844	20.0 - 97.0	58.3 ± 9.2	140 - 1,200	389.2 ± 92.1
44	*Morulius chrysophekadion* (Pla ka dam)	46	10.0 - 35.0	21.5 ± 8.4	45 - 200	116.7 ± 53.5
45	*Mystus mysticetus* (Pla khayaeng khang lai)	488	8.0 - 16.0	11.0 ± 2.8	45 - 160	83.8 ± 45.4
46	*Mystus singaringan* (Pla khayaeng bai khao)	165	8.0 - 16.0	10.7 ± 2.5	45 - 180	80.0 ± 44.5
47	*Nandus oxyrhynchus* (Pla sua pru)	24	2.0 - 2.5	2.3 ± 0.2	5 - 8	7.0 ± 1.4
48	*Notopterus notopterus* (Pla salat)	273	11.0 - 33.0	25.1 ± 4.9	40 - 310	189.5 ± 68.9
49	*Ompok krattensis* (Pla nua on)	165	10.0 - 18.0	12.3 ± 3.4	25 - 80	44.0 ± 21.8
50	*Oreochromis niloticus* (Pla nin)	9	15.0 - 36.0	26.1 ± 8.3	54 - 910	378.7 ± 396.3

Table 1 (cont)
Seventy-three species of fish examined for *Gnathostoma* larvae.

No.	Fish scientific name (Thai name)	No. Examined	Length (cm)		Weight (g)	
			Range	Mean ± SD	Range	Mean ± SD
51	*Osphronemus goramy (*Pla raet)	5	35.0 - 52.0	43.4 ± 7.3	550 - 950	760.0 ± 163.6
52	*Osteochilus hasselti* (Pla soi nok khao)	332	10.0 - 18.0	11.0 ± 1.3	58 - 110	71.0 ± 13.8
53	*Osteochilus melanoplera* (Pla phorm hua men)	1	23.0		450	
54	*Oxyeleotris marmoratus* (Pla bu sai)	162	13.0 - 36.0	23.7 ± 7.6	80 - 800	319.6 ± 260.8
55	*Pangasianodon hypophthalmus* (Pla sawai)	4	40.0 - 55.0	48.0 ± 6.8	200 - 300	245.0 ± 52.6
56	*Pangasius macronema* (Pla sangkawat luang)	123	10.0 - 18.0	11.8 ± 2.1	40 - 80	45.6 ± 8.8
57	*Paralaubuca harmandi* (Pla paep)	189	5.0 - 15.0	8.0 ± 3.5	20 - 40	25.0 ± 7.1
58	*Parambassis siamensis* (Pla paen kaeo)	403	3.0 - 5.0	3.5 ± 0.8	10 - 20	14.0 ± 5.2
59	*Parambassis wolffii* (Pla paen kaeo yak)	13	3.0 - 5.0	3.8 ± 3.7	40 - 70	56.9 ± 11.1
60	*Plotosus canius* (Pla duk thale)	3	23.0 - 35.0	30.7 ± 6.7	60 - 190	146.7 ± 75.0
61	*Polynemus longipectoralis* (Pla nuat phram)	54	10.0 - 15.0	10.5 ± 1.2	60 - 100	68.0 ± 15.7
62	*Pristolepis fasciatus* (Pla mo chang yiap)	497	3.5 - 14.0	8.4 ± 2.0	10 - 100	58.2 ± 17.9
63	*Puntioplites proctozysron* (Pla kramang)	199	8.0 - 23.0	16.0 ± 4.3	40 - 120	76.5 ± 28.2
64	*Puntius brevis* (Pla taphian sai)	517	2.5 - 21.0	4.7 ± 5.2	10 - 120	19.3 ± 23.6
65	*Rasbora argyrotaenia* (Pla siu khwai khang ngoen)	458	5.0 - 7.0	5.8 ± 1.0	4 - 5	4.2 ± 0.5
66	*Systomus orphoides* (Pla kaem cham)	227	2.5 - 21.0	7.1 ± 7.1	10 - 120	30.6 ± 32.4
67	*Systomus partipentazona* (Pla sua sumatra)	1	2.5		8	
68	*Thynnichthys thynnoides* (Pla soi klet thi)	191	10.0 - 16.0	11.0 ± 1.3	60 - 100	70.9 ± 14.3
69	*Trichogaster microlepis* (Pla kradi nang)	179	7.0 - 15.0	10.7 ± 2.0	15 - 36	25.9 ± 6.3
70	*Trichogaster pectoralis* (Pla salit)	348	12.0 - 27.0	16.9 ± 3.0	60 - 490	76.5 ± 25.9
71	*Trichogaster trichopterus* (Pla kradi mo)	639	6.0 - 15.0	10.7 ± 2.1	10 - 38	26.1 ± 6.3
72	*Wallago attu* (Pla khao khao)	2	34.0 - 42.0	38.0 ± 5.7	120 - 180	150.0 ± 42.4
73	*Xenentodon cancila* (Pla krathung heo)	117	15.0 - 18.0	16.3 ± 0.8	10 - 40	15.9 ± 7.9
	Total	12,216				

Table 2
Eight species of fish found to be infected with *Gnathostoma* larvae.

No.	Fish scientific name (Thai name)	No. examined	No. positive	% positive	Total no. of larvae recovered	Mean no. of larvae/fish (range)
1	*Monopterus albus* (Pla lai na)	1,844	555	30.1	5,532	10.0 (1-698)
2	*Anabas testudineus* (Pla mo Thai)	365	28	7.7	143	5.1 (1-14)
3	*Channa striata* (Pla chon)	231	17	7.4	143	8.4 (1-21)
4	*Clarius macrocephalus* (Pla duk ui)	165	11	6.7	67	6.1 (1-16)
5	*Channa micropeltes* (Pla chado)	98	5	5.1	36	7.2 (3-12)
6	*Channa lucius* (Pla krasong)	175	7	4.0	38	5.4 (1-12)
7	*Clarius batrachus* (Pla duk dan)	137	2	1.4	9	4.5 (3-6)
8	*Ompok krattensis* (Pla nua on)	165	1	0.6	1	1.0
			626		5,969	

batch of climbing perch with a maximum infection rate as high as 64.1% (25/39) in February 2001. This batch was caught from a pond in a ricefield, indicating a site with a high risk of infection.

Rojekittikhun *et al* (1998b) reported that the average size of *G. spinigerum* advanced third-stage larvae recovered from eels was 4.2 ± 0.7 x 0.4 ± 0.04 mm (range, 2.1-6.2 x 0.3-0.6 mm). A similar result, 3.97 ± 0.50 x 0.40 ± 0.04 mm (2.70-

Table 3
The body size of *G. spinigerum* advanced third-stage larvae (from swamp eels, n = 100).

Body size (mm)	Range	Mean ± SD
Length	2.70 - 5.10	3.97 ± 0.50
Width	0.29 - 0.60	0.40 ± 0.04

Table 4
Numbers of cephalic hooklets of *G. spinigerum* advanced third-stage larvae (from swamp eels, n = 100).

Row no.	No. of hooklets	
	Range	Mean ± SD
Row 1	40 - 43	41.8 ± 0.5
Row 2	42 - 45	43.6 ± 0.6
Row 3	44 - 48	46.1 ± 0.9
Row 4	48 - 51	49.3 ± 0.7

Table 5
Relative frequency distribution of cephalic hooklet numbers of *G. spinigerum* advanced third-stage larvae (from swamp eels, n = 100).

No. of hooklets (range)	Relative frequency (%)			
	Row 1	Row 2	Row 3	Row 4
40 - 42	98	4	0	0
43 - 45	2	96	24	0
46 - 48	0	0	76	11
49 - 51	0	0	0	89

5.10 x 0.29-0.60 mm), was observed in the present study. Methods of fixation and relaxation, together with the equipment used for measurement may have caused the differences in body sizes of the worms between each report.

Daengsvang (1980) reported that the number of cephalic hooklets of the encysted larvae, obtained from the muscle of infected white mice, normally increased posteriorly from rows 1 to 4, averaging 41.5, 43.3, 46.6, and 49.2, respectively. Those of the hepatic-stage larvae from laboratory-infected mice, 2-5 weeks post-infection, were 42.8 ± 2.6, 45.3 ± 2.8, 46.9 ± 2.8, and 50.2 ± 2.9, respectively (Rojekittikhun and Pubampen, 1998); and of the encysted larvae from swamp eels were 42.2 ± 3.0, 44.6 ± 3.1, 46.8 ± 3.1, and 50.0 ± 3.0, respectively (Rojekittikhun *et al*, 1998b). The result of the present study showed an almost identical pattern, *ie*, 41.8 ± 0.5, 43.6 ± 0.6, 46.1 ± 0.9, and 49.3 ± 0.7, respectively.

Fresh-water fish are still considered the main source of infection for humans. If a great number were to be subjected to careful examination, it would be highly likely that new species would find their way onto the list of naturally-infected second intermediate hosts of *Gnathostoma*.

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