HETEROPHYID (TREMATODA) PARASITES OF CATS IN NORTH THAILAND, WITH NOTES ON A HUMAN CASE FOUND AT NECROPSY

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INTRODUCTION

Due to their tolerence of a broad range of hosts, heterophyid flukes not uncommonly are able to develop to maturity in man. Little is known of the life histories of most heterophyids in their snail hosts. Most undergo the metacercarial stage in marine and fresh-water fish which are ingested by the definitive hosts, a variety of birds and mammals (Yamaguti, 1958; Pearson, 1964). Human infection can occur wherever fish are eaten raw or partially cooked. In Thailand, Manning et al., (1971) reported finding Haplorchis yokogawai and H. taichui adults in several human autopsies in Northeast Thailand. The intermediate hosts were not determined. There are no other published reports of human heterophyid infections in mainland Southeast Asia, although both of the above parasites and Heterophyes heterophyes have been reported in dogs, and the latter in cats also (Segal et al., 1968). A number of human cases involving both intestinal infections and ectopic deposition of heterophyid eggs (and rarely adults) in the spinal cord and heart have been reported from the Philippines (Africa and Garcia, 1935; Africa et al., 1935 a, b, 1936, 1937; Africa, 1937). Heterophyes heterophyes, Metagonimus yokogawai, Haplorchis yokogawai and H. pumilio are known to be endemic in certain areas of Asia, and at least six other heterophyids occasionally have been recovered from man (Faust et al., 1971).

Stellanthchasmus falcatus adults have been recovered from cats, dogs, mice, birds and

man in the Asian Pacific region, the Middle East and Australia (Noda, 1959; Alicata, 1964; Pearson, 1964) and were first described from man by Africa and Garcia (1935) in the Philippines and later by Alicata and Schattenburg (1938) in Hawaii. Ching (1961) examined stools of 1,380 persons in Hawaii and found 7.6% of Filipinos and native Hawaiians to be infected with S. falcatus. As the ova of heterophyid flukes superficially resemble those of Opisthorchis, and Clonorchis. many heterophyid infections have been assigned erroneously to the common liver flukes. Despite numerous stool surveys, S. falcatus has not been previously detected in Thailand in man or animals. The present paper reports the finding of S. falcatus, Haplorchis yokogawai and H. taichui infection in fish and cats in Northern Thailand and discusses a possible human infection with the latter fluke discovered at necropsy in Chiang Mai.

MATERIALS AND METHODS

During extensive helminth surveys in North Thailand (Ratanasritong and Kliks, 1972) a number of different metacercariae were recovered from the flesh of local fish. Cysts removed from the fins of *Dermogenys pusillus*, were fed to one previously uninfected cat, laboratory rats, mice and chickens. Eggs of a fluke appeared in the faeces of the cat after 13 days and numerous adults were recovered from the ileum at autopsy after 40 days. No other flukes were present in the cats and none of the other animals exposed were

found infected. Adult specimens were sent to J.C. Pearson of the University of Queensland who confirmed the diagnosis of *Stellanthchasmus falcatus*. More recently, a domestic cat was found to be naturally infected with both *Haplorchis yokogawai* and *H. taichui*. Metacercariae of these flukes have been frequently observed in *Puntius orphoides*, *P. leicanthus*, and *P. gonionotus*, all of which are consumed raw or marinaded, by the local population.

Surveys of local snails have uncovered two very similar ocelate parapleurolophocercous cercariae, resembling those of Opisthorchis. These were shed in the morning by 65 of 567 Melanoides tuberculata (11.5%) collected from the same areas as the infected fish. Upon exposure in glass finger bowls the cercariae were observed to penetrate readily and encyst in the fins and flesh of either Puntius species, producing H. taichui and H. yokogawai cysts, and in Dermogenys, resulting in S. falcatus. A definitive description of S. falcatus adults was published by Pearson (1964); Vazquez-Golet and Africa described the metacercariae, and Martin (1958) and Noda (1959) described the cercariae. Hsu (1951) made studies on larval stages of heterophyids. Likewise, descriptions of the adults of Haplorchis taichui and H. yokogawai, have been made by Manning et al., (1971) and Pearson (1964).

While examining necropsy sections of ileum from a patient who died suddenly of heart failure following routine appendectomy, sections of an adult trematode and numerous eggs were noted. The patient did not harbor *Opisthorchis* infection in the liver. Further sections from the same block revealed no additional parasite material. Autopsy permission was not given and other organs were not available for study. A segment of ileum had been removed at necropsy.

DESCRIPTION OF THE PARASITES

Stellanthchasmus falcatus (Figs. 1, 2)

All measurements are given in microns unless otherwise stated.

Adult: (8 specimens, fixed in 10% formalin, stained in carmine, mounted in permount). Small, pyriform, body length 313 to 468 (381), forebody narrower than hindbody, maximum width 126 to 162 (150) across testes (Fig. 1). Cuticle covered with scale-like spines, 3.2 to 4.0 long, curved posteriad and diminishing in size and number

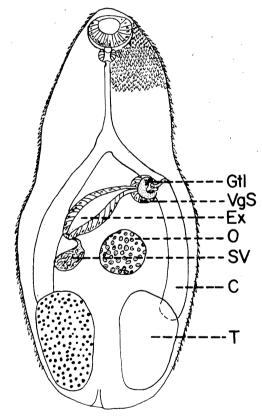


Fig. 1—Diagramatic sketch of Stellanthchasmus falcatus, adult; dorsal view showing spination pattern, genital apparatus and major organs. Uterus, eggs and vitellarica omitted: T, testes; O, ovary; SV, seminal vesicle; VgS, ventrogenital sac; Gtl, gonotyl; Ex., expulsor; C, caeca. (Actual size, 381 by 150 microns average).

in posterior fourth. Oral sucker subterminal, 25 to 36 (34) deep by 29 to 40 (33) wide; prepharynx 10 to 16 (12) long; pharynx 18 to 28 (23) long by 18 to 29 (24) wide; oesophagus 90 to 144 (114) long; bifurcation of caeca at midbody, caeca extending to near posterior edge of testis. Testes large, ovoid, opposite, posterior, 72 to 90 (80) long by 36 to 61 (47) wide. Seminal vesicle thin-walled, 63 to 58 (60) long by 30 to 32 wide, to left of ovary, connected to muscular expulsor lying dorsal to left caecum. Ovary submedian, to left of ventral sucker, 34 in diameter in one specimen. Uterus extensive, coiled in posterior third of body; terminal portion with thickened muscular wall; masses of sperm often seen in coils. Vitelline glands large, follicular, dorsal, extending from level of ovary to posterior extremity. Eggs (10 uterine) 18 to 22 (19) long by 8 to 14 (11) wide, shell smooth, thinwalled, rounded and slightly thickened at the abopercular end, without a distinct knob; tapering from middle to small operculum, 4 to 5 across; without obvious shoulders or neck; (10 faecal, in 10% formalin) 18 to 21.6 (21.2) by 10 to 12 (10.6) (Fig. 2). Gonotyl small, unarmed, arising from left side of ventrogenital sac. Ventrogenital sac median to submedian, on right, posterior to bifurcation of caeca, lined by thick cuticle; contains ventral sucker, gonotyl, male and female genital pores. Ventral sucker indistinct, armed with minute spines, 1 to 2 long, mostly

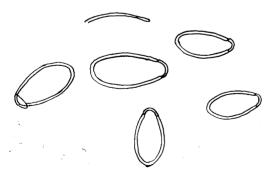


Fig. 2—Characteristic eggs of S. falcatus from cat faeces (X 2000)

distributed over rim in two dense, raised masses.

Host: Domestic cat, natural infection; experimental infection after feeding cysts from fins of *Dermogenys pusillus*.

Habitat: Ileum.

Metacercaria: (10 specimens, alive in wet Cysts spherical to saline preparation). ovoid, 184 to 259 (221) by 151 to 234 (192); wall double, inner 3.6 to 4.8 (4.2), thick, outer delicate, often missing. Organism usually folded once, actively moving; residual eyespot pigment occasionally seen in anterior region. Cuticle heavily spined. Distribution of organs similar to adult: oral sucker 43 to 54 (48) by 26-61 (41), ventrogenital apparatus 25 to 43 (33) by 18 to 34 (24); ventral sucker possesses minute spines, as in adult. Excysted metacercaria active, 270 to 410 (318) long by 120 to 180 (132) wide.

Host: Dermogenys pusillus.

Habitat: Skin of fins.

Haplorchis taichui and H. yokogawai

Adult: The specimens recovered from cats in Chiang Mai were similar in every way to those described by Manning *et al.*, (1971).

Metacercaria: The precocious metacercariae of these two species are similar to the adults and the species can be readily distinguished by the presence of very large spines on the ventral sucker of *H. taichui* and minute ones on that of *H. yokogawai*. Both species active within the cyst; cuticle heavily spined; vestiges of eyespot pigment frequently retained; discoidal granules seen inside intestinal caeca. Encysted *H. taichui* measured 180 to 252 (200) long by 108 to 198 (170); excysted specimens were 220 to 306 (263) long by 160 to 216 (145) wide. Encysted *H. yokogawai* measured 180 to 234 (210) long by 162 to 198 (175) wide; excysted, 360 to 410 (385) long by

110 to 180 (145) wide. Metacercariae of both species appeared to be fully developed as soon as 5 days after exposure of fish to cercariae shed by *M. tuberculata*.

Host: Natural infections in *Puntius leicanthus*, *P. gonionotus*, *P. orphoides*; experimental infection in *P. gonionotus* after exposure to cercariae from *M. tuberculata*.

Habitat: Skin and muscles.

The snail *Melanoides tuberculata* is viviparous; however, those shedding heterophyid cercariae were noted to have ceased production of young, possibly because of parasitic castration.

NOTES ON A HUMAN CASE

Clinical History

A 33-year-old Thai male from Chiang Dao, a rural district of Chiang Mai Province in Northern Thailand, was admitted to the surgical ward of Nakorn Chiang Mai Hospital with a complaint of abdominal pain (which began around the umbilicus) and distension of 3 days duration. Seven days prior to admission, pain, which was not relieved by analgesic drugs, developed in the left shoulder. A history of bloody, mucus diarrhoea during the month prior to admission was given. Nausea and vomiting were not reported.

Physical examination on admission revealed an alert, restless, weak, moderately dehydrated and hyperpneic man. A systolic murmur was detected; lungs clear; abdomen protruding with guarding and rigidity, positive shifting dullness, and decreased bowel sound; liver and spleen not palpable. Temperature 37°C, pulse 120/min, respiration 28/min, blood pressure 130/90 mm Hg. Haemoglobin 15.0 gm%; haematocrit 46%, WBC 20,000/c.mm, with lymphocytes 13%, neutrophils 86%, and eosinophils 1%. Stool

examination positive for hookworm ova; occult blood 4+, numerous white cells and mucus.

Exploratory laparotomy revealed a lesion localized in the right iliac fossa; terminal ileum covered by mucofibrinous exudate; appendix slightly congested, 5.5 cm long by 0.8 cm in diameter; resected. No evidence of perforation. Diagnosis of acute pancreatitis considered.

Post-operatively patient fed by retained nasogastric tube. Haemoglobin 12.5 gm%, haematocrit 38%; WBC 9880/c.mm with monocytes 35%, neutrophils 60%, lymphocytes 35%, eosinophils 2%. Patient had fever and oozing of yellow fluid through operative wound due to peritonitis; died on second post-operative day. Probable cause of death, heart failure. Permission to autopsy denied. Only the appendix and a short segment of inflammed terminal ileum were removed at necropsy and were available for study.

Necropsy Findings

Macroscopic: A 8 cm long unfixed specimen of terminal ileum; serosal surface covered by a patchy layer of purulent exudate; lumen opened along mesenteric border appeared normal; mild hyperaemia of mucosa; no ulcerations or perforations seen.

Microscopic: Haematoxylin and eosin stained sections of terminal ileum showed hyperplasia of mucosal epithelium, pseudoatrophic macrovilli, moderate infiltration of mononuclear cells in lamina propria (Fig. 3); cross sections of a fluke and *Enterobius vermicularis* observed in crypts. Muscularis mucosae moderately infiltrated by eosinophils; serosal surface covered by thick layer of fibrinopurulent exudate. Sections of appendix showed chronic periappendicial inflammation; no parasites seen.

Parasitological: Sections of ileum stained in haematoxylin and eosin; each contained

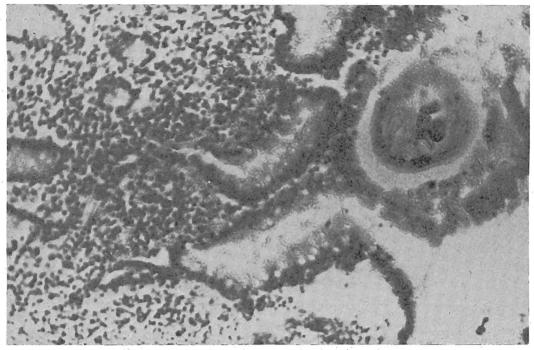


Fig. 3—Ileum with cross section (I) of spined fluke in crypt; infiltration of lamina propria by monocytes; hyperplasia of mucosal epithelium, H. & E. (X 200).

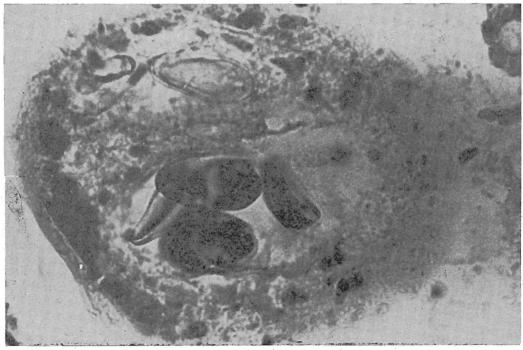


Fig. 4—Heterophyid fluke; cross section showing imbricate spines on cuticle (right); elongate eggs in uterus; H. & E. (X 900).

one cross section of an adult fluke deep crypts; eggs seen inside worm and free in tissues. Largest cross-section measured 111 by 130 in maximum diameter (Fig. 4): integument 3.2 to 4.8 thick; spines scale-like, imbricated, largest 4.5 long by 3.2 wide, arranged in well-spaced rows of 40 to 60 spines each, those projecting from lateral edge of sections appeared to be true spines. Parenchyma vacuolar, parenchymal nuclei uniform, vesicular, 7 to 8 in diameter. Uterine tube thin-walled, filled with elongate embryonating eggs, 32 to 34 long by 13 to 15 wide, containing 6 to 10 pyknotic nuclei and amorphous, acidophilic material; egg shell 1.0 to 1.6 thick with salient operculum and abopercular knob; operculum set off from body by distinct neck (Fig. 5).

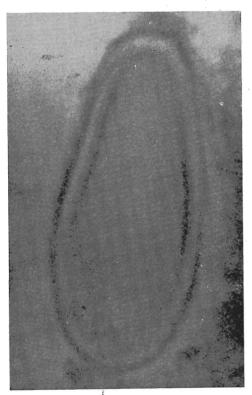


Fig. 5—Detail of fig. 4 showing elongate egg with neck, operculum and abopercular knob; in uterus, H. & E. (X 3500).

Smaller cross section, 126 by 126, tissues degenerate; cuticle inflated, spines clearly visible around entire circumference. Uterus visible in 6 sections, 24 to 32 in diameter, containing eggs with pyknotic nuclei. One section each of seminal vesicle 24 by 54, containing sperm, and ovary, containing vesicular and pyknotic nuclei (Fig. 6). Several cross sections of pinworm, *Enterobius vermicularis*, were also present in the specimen of ileum.

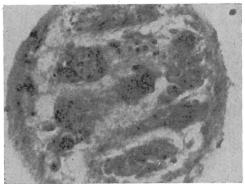


Fig. 6—Heum with cross section (II) of spined fluke in crypt; sections of uterus, ovary and seminal vesicle: H. & E. (X 1000).

Diagnosis: A mature heterophyid fluke, probably *Haplorchis taichui*.

DISCUSSION

Infections with heterophyid flukes in man, cats, and fish-eating animals are known to be widespread throughout Asia but the real prevalence is frequently masked by the simultaneous occurrence in the faeces of ova of *Opisthorchis* and other liver flukes. Heterophyid infection in man, therefore, are demonstrated more readily at autopsy, necropsy or in tissue sections, than by stool examination (Africa *et al.*, 1935 a, b, 1937; Faust *et al.*, 1971). Only after long and careful study were Manning *et al.*, (1971) able to differentiate the ova of *Haplorchis yokogawai* and *H. taichui* from those of *Opisthorchis* in human stools in Northeast Thailand.

The prevalence of human *Opisthorchis* infection in villages in the vicinity of Chiang Mai was reported to vary from 8 to 54% (Cunningham *et al.*, 1970). At the time of the present survey no other trematode eggs had been discerned in human faeces in North Thailand and no special effort had been made to find them.

Collections of more than 13,500 Bithynia snails, the known intermediate host for Opisthorchis in this area, revealed prevalence of 0.02 to 0.3% (Thirachantra and Khamboonruang, 1971). Furthermore, 11.5% of Melanoides tuberculata snails examined in the present work were found to shed at least two types of very similar, typical heterophyid cercaria, while none of several hundred Bithynia shed any cercaria.

Virtually all of the species of cyprinoid fish examined from canals, fields and streams in the Chiang Mai area were heavily infected with the characteristic precocious heterophyid metacercariae. However, *Opisthorchis*-type metacercariae were not found and no adult liver flukes were produced when the cysts were fed to cats.

The present study confirmed the presence of Haplorchis taichui, H. yokogawai and Stellanthchasmus falcatus in naturally-infected cats in Northern Thailand. Of 48 cats whose faeces were examined in a recent survey for coccidian parasites, one was naturally infected with H. yokogawai and H. taichui (Wimonwattrawatee, 1974). In routine helminth faunal surveys by the Department of Parasitology from 1971 to 1974, 3 of 4 autopsied cats harboured adults of Opisthorchis and one was infected with Stellanthchasmus falcatus.

Given the paucity of *Opisthorchis* larval stages in the snail and fish intermediate hosts and the prevalence of reported opisthorchiasis in the human and feline population, it is possible that a significant proportion of the human cases, as diagnosed by stool examina-

tion, may be attributed to one of several heterophyid flukes. Evidence of this derives from frequent finding at autopsy of small, spined flukes deep in the intestinal mucosa in virtually all areas of the world where fish are consumed raw (Alicata, 1964).

In the present case where both *Haplorchis* species and *S. falcatus* constitute well-established enzoonoses in wild and domestic piscavores, the human population must also be at considerable risk.

While it is not possible to positively identify the fluke seen in the necropsy sections of ileum on the basis of size and morphology of the ova, it is believed that H. taichui is the most likely candidate. Differences in fixation and preparation notwithstanding, the maximum cross-section diameter of the fluke in the sections of ileum was 130 which is similar to that of stained and mounted specimens of S. falcatus, (126 to 162), but smaller than H. taichui (210 to 340, Pearson, 1964) and H. yokogawai (195 to 276; Pearson 1964). The ova of S. falcatus, however, are very much smaller, both in utero in mounted specimens (average 19 by 11), and in formalinfixed faeces (21.2 by 10.6) than those seen in the sections (32 to 40 by 13 to 15). The ova of both H. taichui (24 to 28 by 12 to 15; Pearson, 1964; 23 to 32 by 10 to 16; Manning et al., 1971) and H. yokogawai (28 to 30 by 13 to 17; Pearson, 1964) are similar in size. Ova of the latter species are rather rounded, lack a distinct shoulder and operculum and have only a slight abopercular thickening of the shell. H. taichui ova, like those seen in sections (Fig. 5), possess a distinct abopercular knob, and a salient operculum set off from the body by a well-defined neck. Unlike the rounded ova of H. yokogawai those of both H. taichui and those seen in section were elongate, appeared lopsided and slightly asymmetric.

The patient in the present human case, as in many of those recorded by Africa and his

associates (1935 a, b, 1936), apparently suffered from a pathological cardiac condition. probably in the mitral valve. While Africa was able to demonstrate heterophyid ova in sclerosed mitral valves and in myocardium, permission to autopsy in the current case was denied and cardiac involvement with deposited heterophyid ova could not be verified. It should be noted that while hookworm ova were found in the patient's stool, no trematoda ova were seen. Further studies of sedimented mucosal scrapings in local autopsies, particularly in persons positive for trematode ova in the stool, or with cardiac symptoms, will be required in order to determine the importance of heterophyid flukes as agents of human disease in Northern Thailand.

SUMMARY

Adult flukes recovered from the ileum of a cat at autopsy, 40 days after being fed with cysts obtained from local fish was identified as Stellanthchasmus falcatus. A brief description of adult, metacarcaria, host and habitat was given. Haplorchis taichui and H. yokogawai were also recovered from natural and experimentally infected cats and fish. Necropsy sections of ileum from a patient revealed a matured heterophyid fluke probably H. taichui.

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