

## *Stictodora fuscatum* (Heterophyidae) metacercariae encysted in gobies, *Acanthogobius flavimanus*

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**Abstract:** The metacercariae of the genus *Stictodora* encysted in gobies, *Acanthogobius flavimanus*, purchased from a local market of Haenam-gun, Chollanam-do, were identified as those of *S. fuscatum*. A total of 1,437 metacercariae were collected from 30 gobies and some of them were orally fed to cats in order to obtain the adult worms. The metacercariae were 0.194-0.301 mm by 0.168-0.270 mm in size, yellowish brown, round to elliptical, and with a thin and transparent cyst wall. The excysted larvae were 0.280-0.423 mm long and 0.150-0.184 mm wide, beset with numerous tegumental spines on the anterior half of the body, and equipped with a characteristic ventrogenital sac. Total 21 adult worms were recovered from 2 cats 7 days after infection. The adult worms were small, leaf-like, 0.514-0.918 mm long and 0.235-0.377 mm wide. The ventrogenital sac was elliptical and its gonotyl was armed with 13-18 spines which were radially arranged and appeared as fan-shape. It has been confirmed that *A. flavimanus* is a second intermediate host of *S. fuscatum* in Korea.

**Key words:** *Stictodora fuscatum*, *Acanthogobius flavimanus*, metacercariae, ventrogenital sac, gonotyl spines, cat

### INTRODUCTION

*Stictodora fuscatum* is a small intestinal fluke belonging to the family Heterophyidae. It was originally described as *Cornatrium fuscatum* by Onji and Nishio (1916) from cats which were experimentally fed with the mullet, *Mugil cephalus*, in Japan. Later Yamaguti (1939) transferred it into the genus *Stictodora* Looss, 1899. In the genus *Stictodora*, more than 20 species including the type species *Stictodora sawakiensis* have been described in the literature (Yamaguti, 1958; Velasquez, 1973; Kinsella and Heard, 1974). However, in

Korea, only 2 species have been reported. One species is known as *Stictodora fuscatum* which was recovered from a human infection case by Chai *et al.* (1988). The other is *S. lari* from kittens and puppies experimentally infected with the metacercariae collected from the gobies, *Acanthogobius flavimanus* (Chai *et al.*, 1989).

Up to present, 8 species of heterophyid flukes, *i.e.* *Metagonimus yokogawai*, *M. takahashii*, *Heterophyes nocens*, *Heterophyopsis continua*, *Pygidiopsis summa*, *Stellantchasmus falcatus*, *Centrocestus armatus* and *Stictodora fuscatum*, have been reported from humans in Korea (Chai and Lee, 1990). Among them, *S. fuscatum* was the most recent one and the only known species infecting humans among the genus *Stictodora* (Chai *et al.*, 1988).

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Since the first human case infected with *S. fuscatum* was reported, thirteen additional human cases were found from a seashore village in a southwestern coastal area of Korea (Chai and Lee, 1990). The gobies and mullets are known to harbour the metacercariae of this fluke in Japan (Onji and Nishio, 1916). However there were no reports on the intermediate hosts of this fluke in Korea. This study was undertaken to know whether gobies serve as a second intermediate host of and the source of human infection with *S. fuscatum*.

## MATERIALS AND METHODS

### 1. Collection of the metacercariae and infection to cats

A total of 30 gobies, *Acanthogobius flavimanus*, were purchased from a local market in Haenam-up, Haenam-gun, Chollanam-do on August and November, 1990. They were brought to the laboratory under refrigeration, and artificially digested in pepsin-HCl solution after dissected into 3 parts of head, viscera and muscle. The digested materials of each part were filtered with a sieve ( $1 \times 1$  mm mesh) and washed several times in 0.85% saline. Metacercariae were collected from the sediment under a stereomicroscope. Some of the collected metacercariae were observed and measured microscopically and some were artificially excysted. The excysted larvae were fixed in AFA-solution under the cover slip and stained with Semichon's acetocarmine. In order to obtain adult worms, 5 cats were orally fed with 200 metacercariae each.

### 2. Recovery of the adult worms

A cat was necropsied on the day 1 postinfection (PI), and 4 cats were sacrificed on the day 7 and day 9 PI. Their small intestines were resected, opened longitudinally, and washed in 0.85% saline. Worms were collected from the washed intestinal content under the stereomicroscope. They were fixed and stained as the excysted larvae above.

## RESULTS

### 1. Metacercariae of *S. fuscatum*

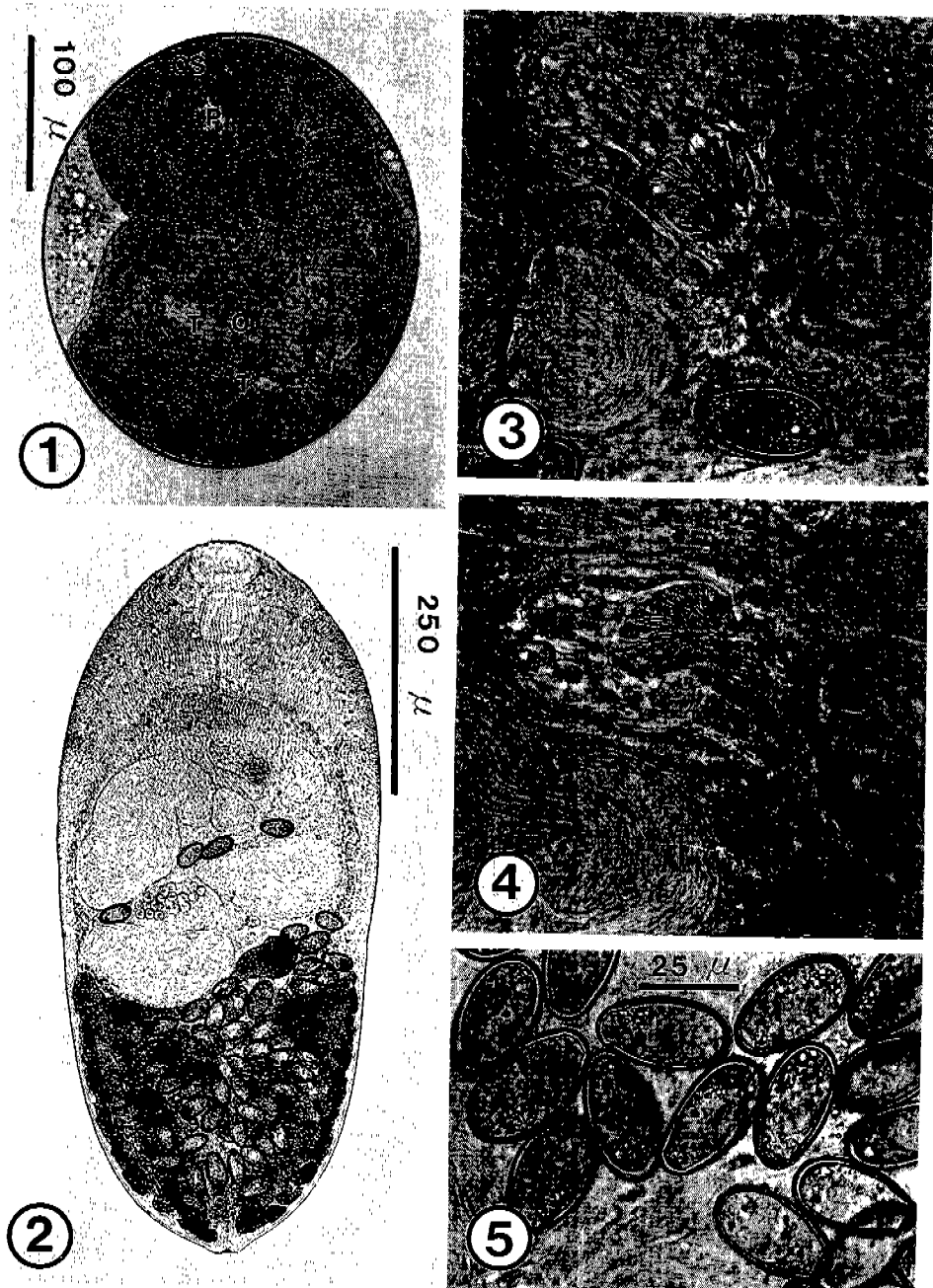
Total 1,437 metacercariae of *Stictodora* sp. were collected from 30 *A. flavimanus*, with the average number per fish of 48. They were found in the muscle (51.1%), head portion (27.7%) and viscera (21.2%).

**Description:** The metacercariae were 0.194-0.301 (0.264) by 0.168-0.270 (0.226) mm in size, yellowish brown, and round to elliptical in shape, with a thin and transparent cyst wall (Fig. 1). The excysted larvae were 0.280-0.423 (0.358) mm long, 0.150-0.184 (0.166) mm wide and beset with numerous tegumental spines on anterior half of the body. They were equipped with the oral sucker ( $0.041 \times 0.045$  mm), prepharynx (0.026 mm), pharynx ( $0.027 \times 0.025$  mm), esophagus (0.025), ovary ( $0.026 \times 0.012$  mm) and 2 testes ( $0.033 \times 0.038$  mm), and characterized by the presence of a ventrogenital sac ( $0.050 \times 0.038$  mm) and metraterm armed with small spines.

### 2. Adults of *S. fuscatum*

Total 9 worms of *S. fuscatum* were recovered from a necropsied cat on the day 1 PI, and 21 worms from 2 cats sacrificed on the day 7 PI. However, no worms were recovered from 2 cats sacrificed on the day 9 PI. Among the recovered worms, 10 of 7 days PI were measured and observed.

**Description:** Body small, leaf-like, 0.514-0.918 (0.714) mm long and 0.235-0.377 (0.313) mm wide, and beset with minute spines on the surface of anterior 2/3 (Figs. 2 & 6). Oral sucker round, 0.041-0.064 (0.056)  $\times$  0.058-0.086 (0.068) mm in size. Prepharynx short, 0.011-0.051 (0.034) mm. Pharynx well developed, 0.046-0.056 (0.050)  $\times$  0.033-0.046 (0.041) mm in size. Esophagus 0.030-0.069 (0.049) mm long. Ceca thick walled and terminated at posterior extremity of the body. Acetabulum slightly embedded in parenchyma, under gonotyl. Gonotyl elliptical, 0.058-0.109 (0.072)  $\times$  0.031-0.051 (0.043) mm in size and armed with 13-18 spines (Fig. 3 & 4). Seminal vesicle constricted into 3 parts and located between the ventrogenital sac and ovary. Ovary



**Fig. 1.** A metacercaria of *S. fuscatum* obtained from a goby showing the oral sucker (OS), pharynx (P), ovary (O) and testes (T). **Fig. 2.** An adult worm of *S. fuscatum* recovered from a cat on the day 7 postinfection. **Fig. 3.** Ventrogenital sac and its gonotyl of the worm in Fig. 2. **Fig. 4.** Ibid, another worm. Note the number and arrangement of gonotyl spines. **Fig. 5.** Intrauterine eggs of *S. fuscatum*, with a thick-shell.

semilunar shape, located between the two testes and 0.025-0.076 (0.044) × 0.064-0.102 (0.082) mm in size. Testes bean-shaped,

obliquely tandem in middle part of the body and 0.031-0.079 (0.052) × 0.076-0.140 (0.109) mm in size. Seminal receptacle small

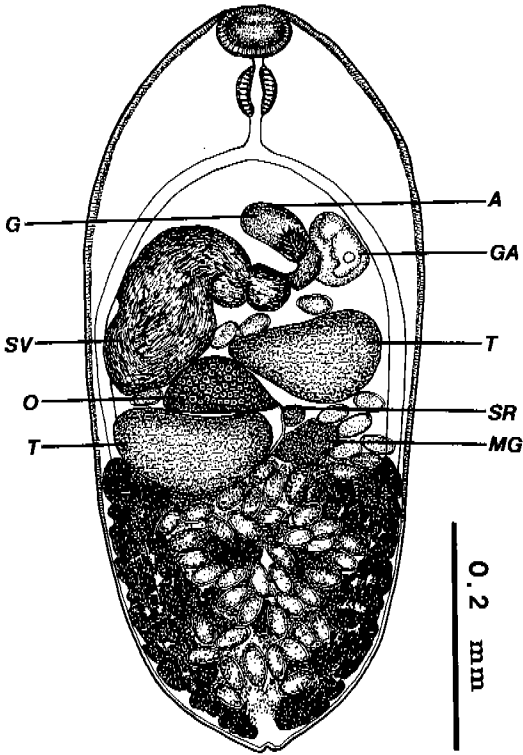


Fig. 6. Schematic drawing (ventral view) of *S. fuscatum* showing the acetabulum (A), gonotyl (G), genital atrium (GA), seminal vesicle (SC), ovary (O), testis (T), seminal receptacle (SR) and Mehlis' gland (MG).

and round, and located between the two testes. Uterus occupied most of the posterior part of the body. Eggs oval, dark brown, thick shelled, 0.032~ 0.038 (0.035) mm long and 0.019~0.023 (0.020) mm wide (Fig. 5). Vitellaria distributed posterior to the testes.

**Specimen deposition:** The Korean Society for Parasitology Helminthological Collection, No. 94-07 and 94-08.

### DISCUSSION

By this study, it was first confirmed that *A. flavimanus* is the second intermediate host of *S. fuscatum* in Korea. According to Onji and Nishio (1916), the metacercariae of *S. fuscatum* were found from the mesentery, adjacent fat tissues and muscles of the mullets and gobies. They were in three size ranges of 0.38-0.52 mm, 0.25-0.36 mm, and 0.16-0.19 mm in

diameter. However, the metacercariae found in this study were chiefly from the muscles, head portions and viscera, and they were  $0.264 \times 0.226$  mm in the average size. The measurements of the metacercariae of *S. fuscatum* by Onji and Nishio (1916) were excessively broad and not specific. Although metacercariae are able to differ in size by the encysted sites, they do not remarkably grow after cercarial penetration into the skin of fish host and cyst formation. Therefore, the metacercariae described by Onji and Nishio (1916) may be more than two kinds of *Stictodora* spp. including *S. fuscatum*.

The morphology of the ventrogenital sac, especially the number and arrangement of chitinous spines on its gonotyl, is regarded as the most important key among the differential points of each species in the genus *Stictodora* (Chen, 1951; Bearup, 1961). The ventrogenital sac in our specimen is elliptical, and its gonotyl is overlapped with the genital sinus and armed with 13-18 spines which are radially arranged and appeared as fan-shape. Considering the morphology of the ventrogenital sac and the number of gonotyl spines, our specimen is compatible with *S. fuscatum* and *S. manilensis* Africa and Garcia, 1935. However, as Chai *et al.* (1988) suggested, *S. manilensis* should be the same species as *S. fuscatum*, and we identified our specimen as *S. fuscatum*.

Five out of 8 species of human-infecting heterophyid flukes in Korea, *H. nocens*, *H. continua*, *P. summa*, *S. falcatus* and *S. fuscatum*, are infected by eating raw flesh of the brackish water fish. As their second intermediate hosts 4 kinds of brackishwater fishes, shad (*Konosirus punctatus*), perch (*Lateolabrax japonicus*), mullet (*Mugil cephalus*) and goby (*A. flavimanus*) have been reported (Chun, 1960 & 1963; Seo *et al.*, 1980, 1981 & 1984; Chai and Sohn, 1988). Among them, the goby has been known as the second intermediate host of *H. nocens*, *H. continua* and *P. summa* (Chun, 1960; Seo *et al.*, 1981 & 1984). By this study, *S. fuscatum* is added to the group of heterophyid flukes contracted from the raw flesh of the gobies.

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=국문초록=

문절망둑(*Acanthogobius flavimanus*)에서 획득한 *Stictodora fuscatum*의 피낭유충

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1990년 9월과 11월에 전남 해남군에서 구입한 문절망둑에서 *Stictodora*속 흡충의 피낭유충을 검출하였으며 이들을 고양이에 실험감염시킨 후 성충을 회수하였던 바, *Stictodora fuscatum*으로 동정되었다. 피낭유충은 30마리의 문절망둑에서 총 1,437개가 근육(51.1%), 두부(27.7%) 및 내장(21.2%) 등에서 검출되었고 0.194~0.301(평균 0.264) x 0.168~0.270(평균 0.226) mm 크기 이었으며 타원형이었다. 5마리의 고양이에 각각 200개씩의 피낭유충을 감염시키고 7일과 9일 후에 충체를 회수하였던 바, 감염 7일된 2마리의 고양이에서 총 21마리의 성충이 회수되었다. 성충은 체장이 0.514~0.918(평균 0.714) mm이었고 체폭은 0.235~0.377(평균 0.313) mm이었으며 나무잎 모양이었다. *Stictodora*속 흡충의 종 동정에 있어서 중요한 열쇠가 되는 ventrogenital sac은 타원형이었고 gonotyl과 genital sinus가 겹쳐 있었으며 gonotyl에는 13~18개의 소극이 부채살 모양으로 배열되어 있었다. 자궁내 충란은 긴 난원형으로 0.032~0.038(평균 0.035) x 0.019~0.023(평균 0.020) mm 크기이었다. 이 연구로 문절망둑이 *S. fuscatum*의 제 2 중간숙주 및 인체 감염원임을 확인하였다.

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