

Identification of *Stictodora lari* (Heterophyidae) metacercariae encysted in the brackish water fish, *Acanthogobius flavimanus*

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Abstract: Metacercariae of the genus *Stictodora* encysted in the head tissue of *Acanthogobius flavimanus* (the gobies) caught at Sachun-gun, Kyongnam Province, were identified to be *Stictodora lari* Yamaguti, 1939 (Trematoda: Heterophyidae), a new parasite fauna in Korea. The metacercariae were 0.39~0.43 mm by 0.32~0.35 mm in size, long elliptical, and with a thin and transparent cyst wall. Total 200 metacercariae were collected from 50 gobies. In order to obtain adult worms two kittens and a puppy were infected each with 34~100 metacercariae, and total 33 adults were recovered between the day 4 and day 8 post-infection. The *S. lari* adults measured 0.95~1.18 mm long and 0.26~0.32 mm wide and the eggs in uteri 0.028~0.033 mm by 0.017~0.020 mm. The most characteristic morphological feature of these flukes was the presence of a gonotyl and gonotyl spines arranged in two groups; densely crowded group of 30~40 spines and linearly-arranged one of 30~40 spines, together of which made a comma (or reversed comma) shape along the lateral margin of the gonotyl. It has been proved by this study that *S. lari* is distributed in southern coasts of Korea.

Key words: *Stictodora lari*, morphology, gonotyl spines, *Acanthogobius flavimanus*, fish, metacercariae, dog, cat

INTRODUCTION

The genus *Stictodora* is a group of minute trematodes belonging to the family Heterophyidae, and parasitizes in the intestine of fish-eating birds or mammals (Yamaguti, 1958). Since this genus was originally established by Looss (1899) with the type species *Stictodora sawakinensis*, more than 20 species have been reported in the world literature (Yamaguti, 1958; Velasquez, 1973; Kinsella and Heard, 1974). They were found from birds and/or mammals,

but had never been from humans. Recently, however, Chai *et al.* (1988) reported a human case of *Stictodora* sp. infection, presumably *S. fuscatum*, based on two adult specimens collected from the diarrheal stool of a 24-year old Korean man after anthelmintic treatment.

Brackish water fishes such as mullets or gobies are known to carry the metacercariae of *Stictodora* flukes (Onji and Nishio, 1916; Yamaguti, 1958). Up to present, however, there were no reports on the intermediate hosts of these flukes in Korea. This study was undertaken to know the possibility of *A. flavimanus* (the gobies)

to serve as a second intermediate host of *Stictodora* spp. in Korea. The present paper describes the discovery of *S. lari* metacercariae from the gobies caught at a southern coastal area.

MATERIALS AND METHODS

1. Collection of metacercariae and infection to animals

Total 50 gobies, *Acanthogobius flavimanus*, were caught from a stream at Sachun-gun (southern coastal area), Kyongnam Province, between July and October, 1987. They were brought to the laboratory under refrigeration. After cut into head and body two parts, they were artificially digested in pepsin-HCl solution and washed several times in physiological saline. Metacercariae of *Stictodora* sp. were collected from the sediment under stereomicroscopic observation. Some of them were mounted on slide glasses, observed and measured. In order to obtain adult worms, two kittens and a puppy were infected orally each with 34, 96 or 70 metacercariae respectively.

2. Recovery of adult worms

Two kittens were sacrificed on the day 4 and day 7 post-infection(PI) and the puppy on the day 8 PI. Their small intestines were resected, opened longitudinally, and washed in saline solution. The worms were collected under stereomicroscopy. They were fixed in 10% neutral buffered formalin under cover glass pressure, and some were stained with Semichon's acetocarmine.

RESULTS

1. Metacercariae of *Stictodora lari*

Total 200 metacercariae of *Stictodora* sp. were collected from the head tissue of 50 *A. flavimanus*, with the average metacercarial density per fish of 4. The metacercariae were 0.39~0.43×0.32~0.35 mm in size, yellowish brown, elliptical, and with a thin and transparent cyst wall. The anterior half of the metacercaria was beset with numerous tegumental spines.

Oral sucker subterminal, and rounded. Prepharynx, pharynx and esophagus present(Fig. 1). Ventrogenital sac and metraterm armed with many small spines(Fig. 2) in mid-portion of the body. However, it was difficult to count the number of gonotyl spines, though they are important keys for the species differentiation. Ovary and testes(Fig. 1) seen in the posterior part of the body.

2. Adults of *Stictodora lari*

Total 33 adult worms of *S. lari* were recovered from three experimental animals; 30 from a puppy, and 2 and 1 from two kittens. Among them, 10 worms of 8 days PI were measured. Mean values are given in parentheses.

Body small, long slender(Fig. 3), 0.95~1.18 mm (1.05) long and 0.26~0.32 mm (0.28) wide. Oral sucker round, 0.050~0.079 mm (0.063) in diameter, and located at subterminal part of the anterior body. Prepharynx 0.030~0.088 mm (0.071) long. Pharynx well developed, and 0.050~0.062 mm (0.053) in diameter. Esophagus slender, and 0.034~0.070 mm (0.057) long. Ceca thick-walled, a little convoluted just after bifurcation, and terminated at the posterior extremity. Ventral sucker slightly embedded in the parenchyma, and 0.061~0.080 mm (0.075) in diameter(Fig. 4). Gonotyl overlaid the ventral sucker, and armed with total 60~80 small spines. The spines arranged in two groups; one densely crowded group of 30~40 spines and the other linearly-arranged one containing 30~40 spines, together of which made a comma (or reversed comma) shape along the lateral margin of the gonotyl(Fig. 4 & 5). Seminal vesicle constricted into 3 or 4 parts, thin-walled, located between ventrogenital sac and ovary, and opened into the genital pore separately from the metraterm. Ovary oval, 0.045~0.076 mm (0.057)×0.065~0.080 mm (0.071) in size, and immediately in front of the right testis. Two testes 0.064~0.099 mm (0.083) long and 0.060~0.108 mm (0.088) wide, and obliquely tandem in middle part of the body. Seminal receptacle 0.025~0.099 mm (0.053) by 0.032~0.062 mm (0.049), and located between two

testes (Fig. 4). Uterus occupied most of the posterior body and reached to the posterior extremity. Eggs oval, dark brown, thick-shelled without muskmelon appearance, 0.028~0.033 mm (0.030) long and 0.017~0.020 mm (0.019) wide (Fig. 6). Vitellaria distributed posterior to the testes. Excretory vesicle Y-shaped.

DISCUSSION

Stictodora lari was first described in Japan from the small intestine of the sea gulls, *Larus crassirostris* (Yamaguti, 1939). Later the metacercariae of *S. lari* were found in several kinds of brackish water fishes including mullets, gobies, leather-jackets, pipefish and toadfish in Australia (Bearup, 1961). In the present study, the gobies (*A. flavimanus*) were found infected with the metacercariae of *S. lari*. According to

Onji and Nishio (1916), the metacercariae of *S. fuscatum* and *S. perpendicularum* were found from the mesentery, adjacent fat tissues and muscle of the mullets and gobies. Interestingly, the metacercariae of *S. lari* were obtained in this study chiefly from the head tissue of the gobies.

The adult worms recovered from the experimental animals revealed several distinct morphological characters, which enabled us to differentiate them from *Galactoscumum*, a morphologically closely related genus to *Stictodora*. The thin-walled and constricted seminal vesicle, post-testicular distribution of the vitellaria, separate opening of the ejaculatory duct and metraterm, and Y-shaped excretory vesicle were all compatible with the genus *Stictodora* (Yamaguti, 1939).

In the genus *Stictodora*, the flukes of different

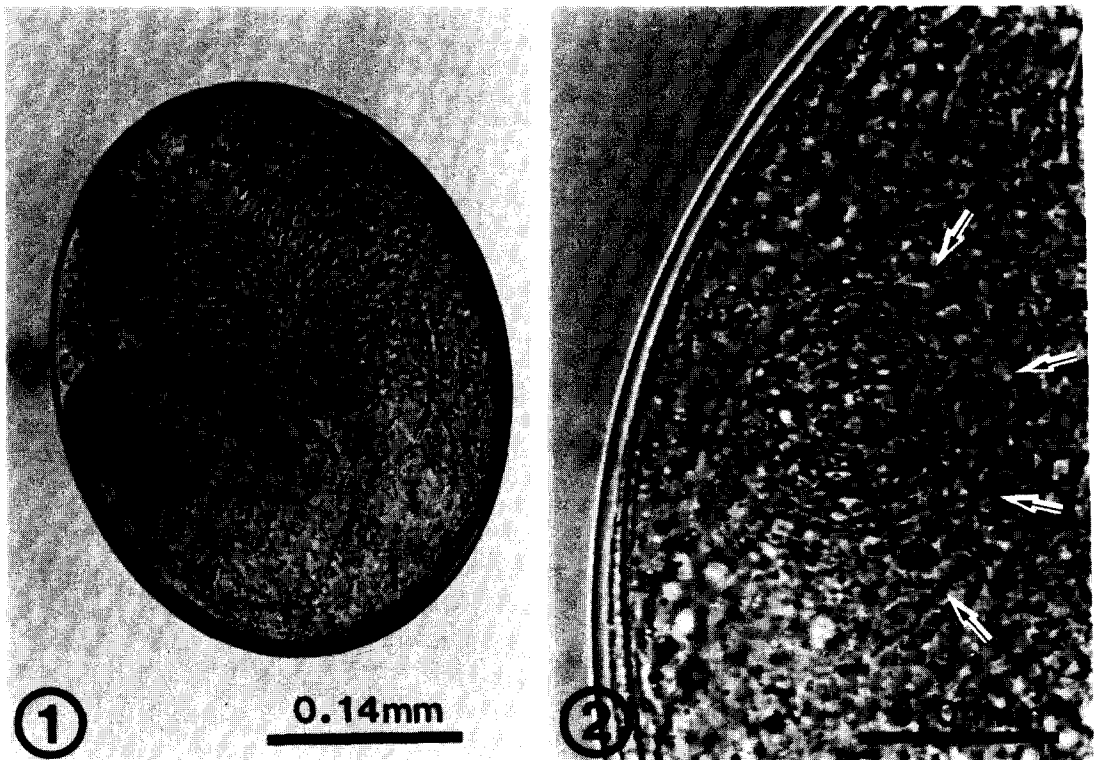
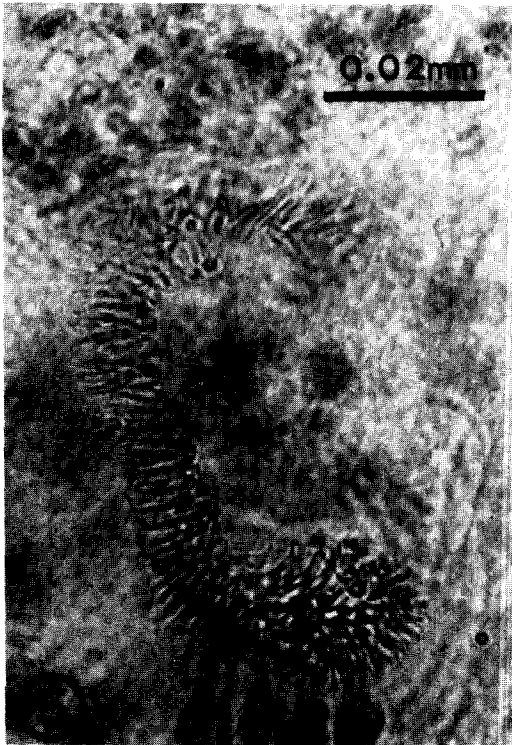
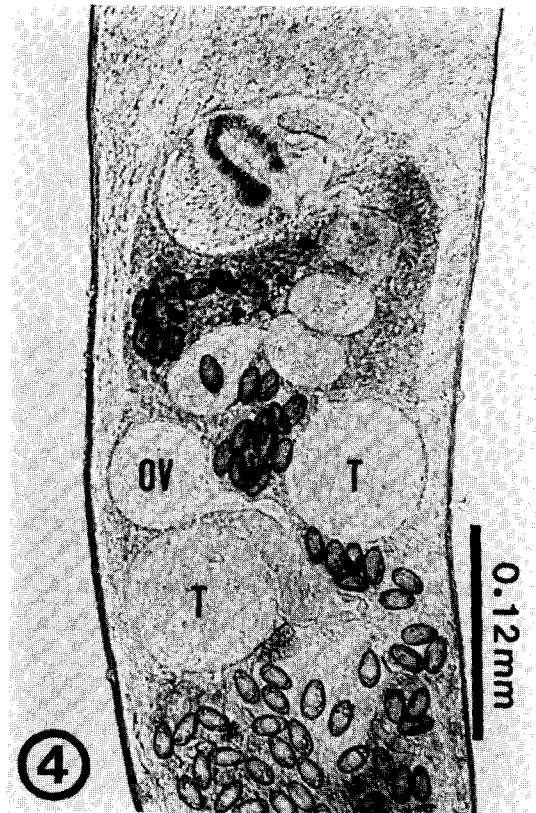
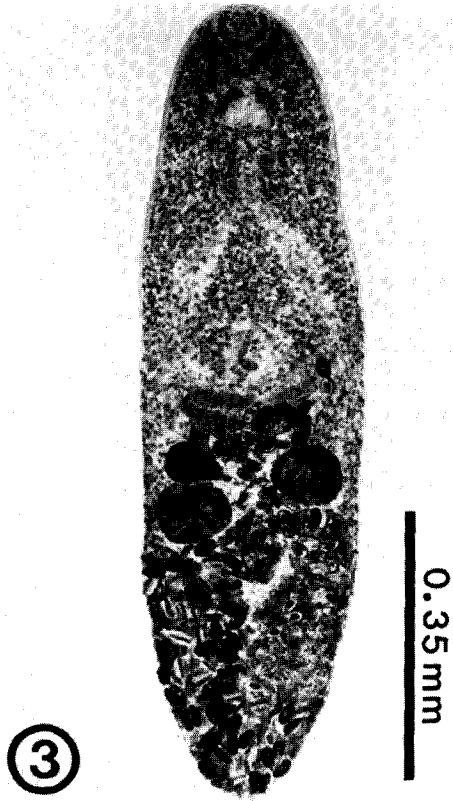


Fig. 1. A metacercaria of *Stictodora lari* obtained from gobies showing oral sucker (CS), pharynx (P), ovary (OV) and testes (T).

Fig. 2. Magnification of the metacercaria showing the ventrogenital sac (arrows), in which the gonotyl spines are not apparent.



species show characteristic features in the morphology of their ventrogenital sac and gonotyl, especially in the number and arrangement of chitinous spines on the gonotyl. For this reason, the gonotyl morphology is regarded as the most important key for the species diagnosis (Chen, 1951; Bearup, 1961).

The gonotyl of *S. sawakinensis* is armed with numerous, up to 78, spines arranged in 6~10 rows (Witenberg, 1929) and that of *S. guerreroi* is armed with 25~28 small circular spinous groups, each composed of about 60~64 spines (Garcia and Refuerzo, 1936). Gonotyl spines of *S. japonica* are in two groups; one containing about 100 small spines and the other containing about 30 slightly larger spines (Yamaguti, 1939). *S. lari* is distinct from all of the above species in having the gonotyl armed with densely crowded group of 40 slightly curved spines, from which 20~30 spines extend in a semi-circular patterns to make a comma-shaped appearance (Bearup, 1961). In our specimens, the morphology of gonotyl spines is highly compatible with *S. lari*, although a slight discrepancy exists in the exact number of the spines.

S. manilensis has a gonotyl armed with 12~15 sclerotized hook-like spines similar to the rostellar hooklets of *Taenia* sp. (Africa and Garcia, 1935; Chen, 1951; Velasquez, 1973). Chai *et al.* (1988) suggested that *S. manilensis* should be the same species as *S. fuscatum* because they each other reveal very similar morphology of their ventrogenital sac and of other genital organs, in spite that the number of gonotyl spines in *S. fuscatum* was not described in the original paper (Onji and Nishio, 1916).

So far, total 7 species of heterophyid flukes including *Metagonimus yokogawai* (Chai *et al.*, 1977 & 1985b; Seo *et al.*, 1981b & 1985),

Heterophyes nocens (Seo *et al.*, 1981a; Chai *et al.*, 1984 & 1985a), *Pygidiopsis summa* (Seo *et al.*, 1981a), *Stellantchasmus falcatus* (Seo *et al.*, 1984a), *Heterophyopsis continua* (Seo *et al.*, 1984b), *Centrocestus armatus* (Hong *et al.*, 1988) and *Stictodora* sp. (Chai *et al.*, 1988) were recorded from humans in Korea. Their intermediate hosts were reported to be fresh water or brackish water fishes.

Considering the popularity of raw fish eating habit among the people in Korea, human infections with *S. lari*, or other heterophyid flukes are expected to occur. Further studies are required on this subject.

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Fig. 3. An adult worm of *S. lari* recovered from a puppy 8 days after experimental infection.

Fig. 4. Mid-portion of an adult *S. lari* showing the ventrogenital sac, seminal vesicle (arrow heads), ovary (OV), testes (T) and eggs in the uterus.

Fig. 5. Magnification of the gonotyl and gonotyl spines. Note reversed comma-shape of the gonotyl spines.

Fig. 6. The uterine eggs of *S. lari*, that are long and ovoid with a thick shell.

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＝국문초록＝

문절망둑(*Acanthogobius flavimanus*)에서 얻은
Stictodora lari 피낭유충의 동정

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채종일 · 박상규 · 홍성중* · 최민호 · 이순형

경남 사천군에서 잡은 문절망둑(*Acanthogobius flavimanus*)의 두부에서 *Stictodora*속 흡충의 피낭유충을 획득하고 강아지와 어린 고양이에게 실험감염시켜 성충을 얻은 바 *Stictodora lari* (Heterophyidae)로 동정되었다.

피낭유충은 문절망둑 50마리에서 총 200개가 검출되었고 크기가 $0.39\sim 0.43\times 0.32\sim 0.35\text{mm}$ 이며 긴 타원형이었다. 강아지 1마리와 어린 고양이 2마리에 34~100개의 피낭유충을 감염시켜 강아지에서 감염 후 8일째에 성충 30마리를, 고양이에서 4일 및 7일째에 각각 성충 2마리 및 1마리를 회수하였다.

성충 10마리(감염 후 8일)를 계측한 바 길이가 $0.95\sim 1.18\text{mm}$ (평균 1.05), 폭이 $0.26\sim 0.32\text{mm}$ (0.28)이었다. 자궁 내 충란은 긴 난원형으로 크기 $0.028\sim 0.033$ (0.030) $\times 0.017\sim 0.020\text{mm}$ (0.019)이었다. 충체의 종 동정(species identification)에서 가장 중요한 특징은 함몰된 복흡반에 겹쳐져 있는 gonotyl의 형태, 특히 소극(small spines)의 배열 및 수이었던 바 gonotyl 외연을 따라 반원 모양으로 둥그렇게 일렬로 배열된 30~40개의 소극과 한쪽으로 휘말려 집괴를 이루고 있는 약 30~40개의 소극, 즉 모두 60~80개의 소극이 ‘콤마’ 형태를 이루고 있는 것이 관찰되었다. 이상의 형태학적 특징으로 이 충체를 *S. lari*로 동정하였다.

이 연구로 우리 나라에 *S. lari*의 생활사가 문절망둑을 중간숙주로 하여 영위되고 있음이 확인되었다.