

LIGHT AND SCANNING ELECTRON MICROSCOPY OF *STELLANTCHASMUS* SP (TREMATODA : HETEROPHYIDAE)

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Abstract. Light and scanning electron microscopy of heterophyid fluke, *Stellantchasmus* sp were studied. The adults were obtained from *Rattus norvegicus*, 7 days after experimental infections with the metacercariae from the body cavity of *Dermogenus pusillus*. Living specimens and stained flukes were examined by light microscope. The present worm differed from *S. batillans* having a single testis while testes of *S. gallinte* were tandem. It is closed to *S. falcatus* and *S. aspinosus* but can not be identified to species because of acetabular spines. The tegumental surface by SEM was investigated on encysted metacercariae and adults. It was nearly smooth with one end shallow wide groove of the metacercarial surface. The entire surface of adult worms were almost covered with scale-like spines, pectinate, 7-9 teeth. The size of scale was largest in the middle of body and decreased both anterior and posterior. No spines were around the excretory pore but has a wrinkled surface appearance. The dorsal surface appeared to have scale-like spines and pectination on the ventral surface. The papillae consisted of a single club-like cilium, solidary or grouped on the anterior part of the body especially anterior of oral sucker. Acetabulum located slightly right of the middle and invagination.

INTRODUCTION

The *Haplorchis*-group of the Heterophyidae flukes, include the genera *Haplorchis*, *Procerovum* and *Stellantchasmus* (Pearson and Ow-Yang, 1982). The subfamily Metagoniminae included the genera *Stellantchasmus*, *Dexiagonimus* and *Metagonimus* (Yamaguti, 1958). The other genera of Heterophyidae were *Haplorchoides* (Pande and Shukla, 1976), *Heterophyopsis* (Hong *et al.*, 1991), *Heterophyes* (Chai *et al.*, 1992) and *Plagiorchis* (Radomyos *et al.*, 1992). *Stellantchasmus* is one of the small heterophyid flukes reported from the cat, rat, mouse, dog, bird and man (Pearson, 1964; Kliks and Tantachamrun, 1974; Tantachamrun and Kliks, 1978; Pearson and Ow-Yang, 1982). There are 4 species, *S. falcatus*, *S. aspinosus*, *S. gallinae* and *S. batillans*. Taxonomically the species is difficult because the worms are very small (Pearson and Ow-Yang, 1982). In Northern Thailand adult *S. falcatus* was found in the human ileum (Tantachamrun and Kliks, 1978) and in Northeastern of Thailand adults were recovered from two human cases (Radomyos *et al.*, 1990).

The scanning electron microscopy has been studied on *Metagonimus yokogawai* (Lee *et al.*, 1984), *Haplorchis pumilio* (Srisawangwon *et al.*, 1989), *Stellantchasmus falcatus* (Radomyos *et al.*, 1990), *Heterophyopsis*, *continua* (Hong *et al.*, 1991) and

Heterophyes nocens (Chai *et al.*, 1992). This paper presents studies in both light and SEM (JEOL-JSM 840A) of *Stellantchasmus* sp from experimental rat, Chiang Mai, Thailand.

MATERIAL AND METHODS

Adults of *Stellantchasmus* sp were obtained from *Rattus norvegicus*, 7 days after experimental infections with the metacercariae from the body cavity of *Dermogenus pusillus*. The worms were fixed in 5% formalin overnight without depression, stained with Borax's carmine, dehydrated through alcohol series, mounted in permount for light microscopy. The species identification referred to Yamaguti (1958); Pearson (1964); Kliks and Tantachamrun (1974); Pearson and Ow-Yang (1982); Radomyos *et al.* (1990). Specimens for SEM were prepared by washing the worms several times with 0.85% NaCl, fixed in cold 4% paraformaldehyde in phosphate buffer pH 7.4 overnight without depression, washed several times in phosphate buffer, post fixed in 1% osmium tetroxide for 1 hour, washed in phosphate buffer, dehydrated through graded series of ethanol, transferred to acetone and dried in critical point dryer, mounted on stubs under stereo microscope, coated with gold and examined with JEOL-JSM840A scanning electron microscope. The ultrastructure surface of adult worms re-

ferred to Lee *et al* (1984); Srisawangwong *et al* (1989); Radomyos *et al* (1990); Sugiyama *et al* (1990); Hong *et al* (1991); Chai *et al* (1992); Sobhon *et al* (1994) and Pakdeenarong *et al* (1995).

RESULTS

Light microscopic observation

Adults *Stellantchasmus* sp obtained from rat (*Rattus norvegicus*), 7 days postinfection (Fig 1-2), 15 were stained and measurements were given in microns. The body is pyriform, body length 426 (300-500), forebody narrower than hindbody with maximum width 236 (190-290) across testes. Almost the entire surface was covered with scale-like spines, 3 (2.5-5) long. Oral sucker subterminal, 30 (20-40) long by 35 (20-50) wide; prepharynx 12 (7-15) long; pharynx 24 (20-30) long by 26 (20-37) wide; esophagus 67 (42-115) long with bifurcation extending to anterior border of testis. Testes large, ovoid, opposite, 100 (30-162) long by 65 (25-92) wide. Seminal vesicle thin-walled to left of rounded ovary, connected to expulsor lying dorsal to left cecum. Expulsor with muscle fibers, 55 (32-80) long by 30 (22-37) wide, opened into ventrogenital sac dorsally on left side and anterior to ovary. Rounded ovary slightly right and anterior to testes, 54 (25-77) in diameter. Uterus coiled in posterior part of the body, filled with operculated eggs. Vitelline glands dorsal to ovary and extending between testes. Eggs 21 (20-22) long by 13 (10-15) wide, thin-walled and smooth shell, a small operculum at one end.

Ventrogenital sac is slightly submedian to right of the body, posterior to cecal bifurcation, contains acetabulum, gonotyl, male and female genital pore. Acetabulum is smaller than the oral sucker, 30 (27-37) in diameter.

Scanning electron microscopic observation

The surface of encysted metacercariae by SEM was covered with mesentery in body cavity of *Dermogenus pusillus* (Fig 3). The surface without mesentery was smooth and a shallow wide groove (arrow) was present, cytoplasmic projection was not seen. The adult worm was pyriform, tapering at anterior (Fig 4). Body covered with scale-like spines except around the excretory pore but with a wrinkled surface (Fig 5). The spines are pectinate and arrange in transverse rows, alternate between a row. Most of the spines are pectinate with 7-9 teeth but the size was largest at the middle of the body. The oral sucker was

larger in size than acetabulum and distinct details. Acetabulum located slightly right and invaginated with unknown acetabular spine, arrow (Fig 6). Scale-like spines were decreasing size both anterior and posterior part of the body.

Papillae were single or grouped especially the anterior part of oral sucker. Each papilla was a hollow tube and a single club-like cilium was arising from the hollow (frame) on the surface at anterior part of oral sucker and between spines on the ventral surface (Fig 7).

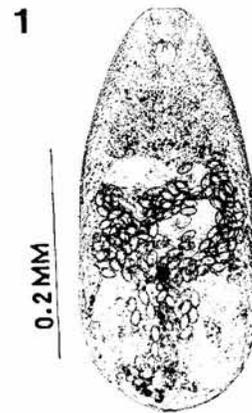


Fig 1 - A living adult of *Stellantchasmus* sp (dorsal).

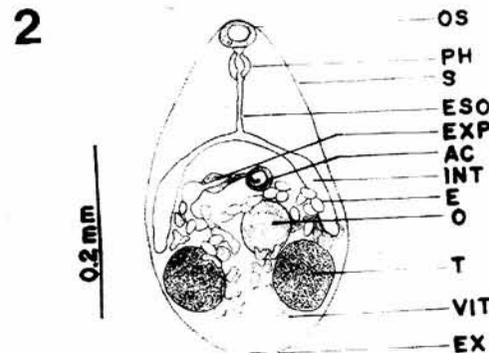


Fig 2 - A drawing of adult *Stellantchasmus* sp, Camera lucida.

AC = acetabulum, E = eggs, ESO = esophagus, EX = excretory pore, EXP = expulsor, INT = intestinal ceca, O = ovary, OS = oral sucker, PH = pharynx, S = spines, T = testes, VIT = vitelline gland



Fig 3 - SEM micrograph had been used to investigate encysted metacercariae from body cavity of *Dermogenus pusillus* (X 285, scale bar = 100 μ).

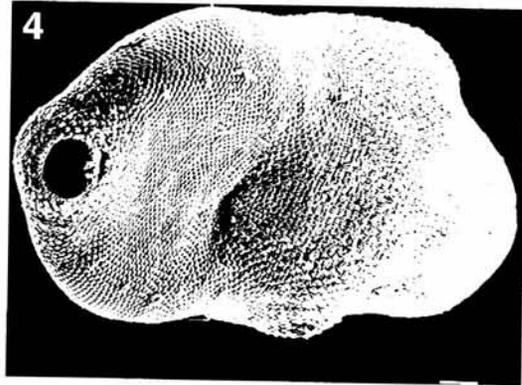


Fig 4 - The entire adult of *Stellantchasmus* sp (X 900, scale bar = 10 μ).

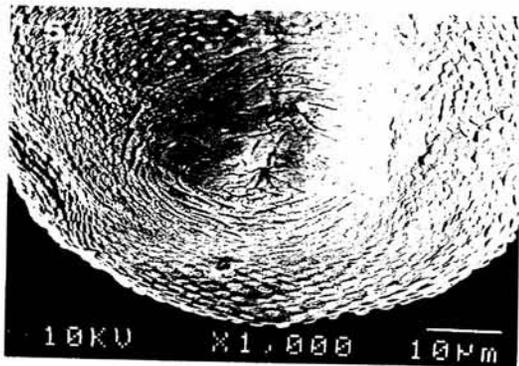


Fig 5 - Excretory pore was shown no spines but wrinkled (X 1,500, scale bar = 10 μ).



Fig 6 - The scale-like spines on the middle of the body around the acetabulum, arrow (X 9,000, scale bar = 10 μ).

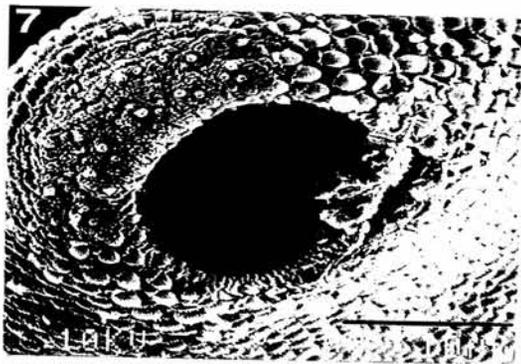


Fig 7 - The oral sucker, abundant papillae on the anterior part (X 3,450, scale bar = 10 μ); frame, club-like papillae.

DISCUSSION

The present *Stellantchasmus* sp differed from *S. batillans* having a single testis while *S. gallinae* testes were tandem (Pearson, 1964; Pearson and Ow-Yang, 1982). The body, testes, expulsor and acetabulum of *S. aspinosus* was shown to be smaller in range of the present fluke (Pearson and Ow-Yang, 1982). The size of the body and organs of the present worms were smaller and wider than the range of *S. falcatus* except the length of prepharynx. *S. falcatus* may reach 47 (Tantachamrun and Kliks, 1974; Pearson and Ow-Yang, 1982; Radomyos *et al.*, 1990). For species identification the acetabular spines would have to be sectioned through the genital complex.

The metacercariae of the present flukes were col-

lected from the body cavity of *Dermogenus pusillus*. The eggs of the adult worms appeared in feces of *Rattus norvegicus*, 7 days postinfection while metacercaria reported by Tantachamrun and Kliks (1974) were recovered from flesh of *Dermogenus pusillus* and the eggs of the fluke appeared in the feces of the cat after 13 days.

SEM observation of *S. falcatus* was first studied in Thailand by Radomyos *et al* (1990) who reported cuticular spines prominent over the whole body but no other details were available. In this study revealed that the tegument of *Stellantchasmus* sp was covered with scale-like spines, similar to *Metagonimus yokogawai* (Lee *et al*, 1984), *Haplorchis pumilio* (Srisawangwong *et al*, 1989), *Heterophyopsis continua* (Hong *et al*, 1991) and *Heterophyes nocens* (Chai *et al*, 1992). Tegumental surface of adult worms differed from *Metagonimus yokogawai*, the whole surface was armed with scale-like spines; 7-8 points on anterior body and 2-3 points on posterior body (Lee *et al*, 1984). The oral sucker spines had 4-5 teeth on *Haplorchis pumilio* whereas 7-9 teeth on this report (Srisawangwong *et al*, 1989). The other similar characteristics of heterophyid fluke showed type of club-like papillae (Lee *et al*, 1984; Srisawangwong *et al*, 1989). The differences of papillae and spines from another family were dome-shaped papillae of *Paragonimus heterotremus* (Sugiyama *et al*, 1990); each spine has serrated edge with 16-20 sharp points, the necklace of papillae around the acetabulum and dome shaped with nipple like tip on the lateral surface of *Fasciola gigantica* (Sobhon *et al*, 1994); the nipple-like tip papillae were observed on the middle part of the body of *Diplodiscus* sp (Noppakun *et al*, 1995).

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