A New Digenetic Trematode, Amblosoma suwaense sp. nov., the Morphology of its Adult and Metacercaria (Trematoda: Brachylaimidae)*

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In the course of studies on the helminth fauna of Lake Suwa, Nagano Prefecture, carried on at odd times since 1970, a brachylaimoid metacercaria has been found parasitizing the fresh-water snail, *Sinotaia quadrata* (Benson). The metacercaria was already reported briefly in a preliminary note by Shimazu and Koh (1973).

The experimentally raised adults were successfully obtained by inoculating the metacercariae on the chick chorioallantois, although the natural adults have not been got as yet. The adult and metacercarial stages obtained proved to differ morphologically from those of the only affined species, *Amblosoma exile* Pojmańska, 1972 (Brachylaimidae: Leucochloridiomorphinae).

Materials and Methods

The metacercaria was found unencysted in the space between the digestive gland and the shell, usually in its top region, of both the sexes of the fresh-water snail, *Sinotaia quadrata* (Benson) (Viviparidae), in Lake Suwa and its neighbouring waters, Nagano Prefecture.

Many metacercariae taken out of the snails collected in the lower Funato were washed in three changes of sterile Hanks' solution containing penicillin (300 units/ml) and streptomycin (300 μ g/ml), and then inoculated, ten to fifteen per egg, on the chorioallantois just under the air chamber of 4- or 7-day-old

fertile hens' eggs. The inoculated eggs were maintained at 34 to 39°C and examined at various intervals of one to twelve days after inoculation. The flukes appeared almost all in the albumen of the eggs but very rarely on the chorioallantois at the examination. Several worms were found still alive in the addled eggs. The 15-day-old chorioallantoic specimens wers also prepared through once a subinoculation at another time.

These chorioallantoic trematodes were flattened under the moderate coverslip pressure, fixed in AFA fixative, stained with Heidenhain's iron haematoxylin, and mounted in Canada balsam. Similarly, the whole mounts were made of the metacercariae from the snails collected in the lower Funato during 1970 to 1974.

It was found that the metacercariae when cultured in the fertile egg at 34 to 39°C developed to the adult stage and began to produce eggs at the earliest at five days after inoculation. Accordingly, the description of the adult stage is based on 29 of the fully ovigerous 5- to 15-day-old chorioallantoic specimens. But the reproductive system in the adult stage was made detailed on much vounger mature worms. The description of the metacercarial stage is based on 24 whole mounts. All whole mounts were measured by means of a screw-micrometer eye-piece. Drawings were made with the aid of a camera lucida.

^{*} Supported in part by the "Nagano-ken Kagaku Shinkô Kai", Nagano Prefecture, in 1971-1973.

Amblosoma suwaense sp. nov.

Adult

(Figs. 1-3)

Host and habitat: Chick chorioallantois (experimental).

Date: November 1973—February 1974.

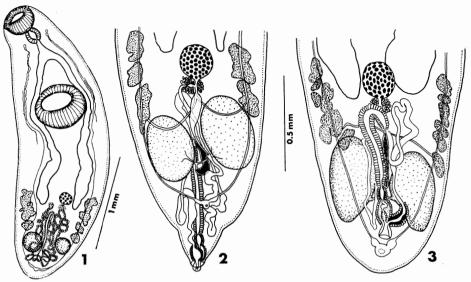
Specimens: Holotype and paratypes (in part) deposited in National Scientific Museum, Tokyo, Coll. No. NSMT-PL-1587; another paratypes in author's collections.

Description: Body fairly small, oval, elongated, 1.79-3.51 mm in length by 0.56-1.16 mm in maximum width at midlevel of body, reddish in colour when alive. Cuticle thick, without spines. Oral sucker 0.21-0.31 mm long by 0.24-0.37 mm wide, more thickened in dorsal brim than in ventral, with mouth subventral in position. Prepharynx very short. Pharynx barrel-shaped, 0.10-0.17 mm in diameter. Oesophagus short, extending backwards at most to midway between pharynx and ventral sucker. Intestinal caeca undulating, reaching two-hirds of hindbody. Ventral sucker round, 0.35-0.60 mm long by 0.35-0.68 mm wide, slightly anterior to midlevel of body; sucker width ratio 1:1.42-1.89.

Reproductive system present within posterior half of hindbody. Testes ellipsoidal, usually symmetrical, sometimes displaced diagonally with constriction of body, situated a little behind posterior ends of intestines; right testis 0.15-0.48 mm long by 0.12-0.26 mm wide, left testis 0.15-0.42 mm long by 0.13-0.28 mm wide. Vasa efferentia short. Vas deferens swollen to act as a seminal vesicle, 0.21-0.42 mm long, ascending just dorsally or laterally to pars prostatica. Pars prostatica tubular, straight or winding with constriction of body, 0.20-0.50 mm long, bending proximally, behind or slightly overlapping ovary, descending longitudinally between testes, surrounded tightly by welldeveloped prostate gland cells. Male duct between pars prostatica and cirrus pouch muscular, thick-walled, surrounded by gland cells, sometimes expanded to preserve spermatozoa. Cirrus pouch ovoid, 0.04–0.13 mm long by 0.04–0.08 mm wide, usually behind level of posterior border of testes, containing winding ejaculatory duct and short cirrus armed with small hooks. Genital atrium funnel-shaped, small. Terminal genitalia sometimes held in a cylindrical or cone-like protrusion from ventral surface of body. Genital pore median, opening between testes and excretory pore (or occasionally transferred to behind posterior end of body, when located on top of extending protrusion of body).

Ovary globular to oval, 0.10-0.25 mm in diameter, pretesticular, almost median, situated between posterior ends of intestines. Oviduct very short. Ootype a little behind ovary; Mehlis' gland compactly massed, free in parenchyma. Laurer's canal swollen proximally to act for a seminal receptacle, descending longitudinally, opening outside between testes on dorsal surface of body. Uterus folded, restricted almost to area enclosed with gonads and genital pore, rarely extending forwards very slightly beyond anterior border of ovary. Metraterm globular, thick-walled, 0.08-0.27 mm long by 0.05-0.15 mm wide, situated on left of cirrus pouch, surrounded by gland cells. operculated, dark brown in colour, 0.016-0.018 by 0.035-0.039 mm (ten in holotype in balsam), and 0.015-0.018 by 0.029-0.032 mm (ten in one of paratypes); embryo not worked out. Vitellaria follicular, extracaecal, shortly extending mostly from midlevel of hindbody to in front of testes, rarely from level of ovary to midlevel of testes.

Excretory vesicle not epithelial, saccate, small, 0.20–0.29 mm in diameter. Each main collecting tube almost extracaecal, expanded in proximalmost portion, ascending to level of pharynx, turning posteriorly to form a ciliated descending limb, and then dividing into anterior and posterior collecting tubes at level of ventral sucker. Excretory pore dorsal, near posterior end of body.



Figs. 1-3 Adults of Amblosoma suwaense sp. nov.

- Fig. 1 Holotype, ventral view.
- Fig. 2 Reproductive system in one of younger adults, dorsal view.
- Fig. 3 Reproductive system in another younger adult, ventral view.

Metacercaria

(Figs. 4-9)

Host: Sinotaia quadrata (Benson) (Viviparidae).

Habitat: Between digestive gland and shell.

Locality: Lake Suwa and its neighbouring waters, Nagano Prefecture.

Specimens: Coll. No. NSMT-PL-1588.

The metacercariae so far obtained are separated into two groups for convenience on the basis of the degree of development of the reproductive organs: The Group I includes 14 specimens with the differentiated gonads and genital ducts but without the developed vitellaria; and the Group II has 10 specimens with the fully formed genitalia including the vitellaria.

The shape of the body and the anatomy and relative situation of each internal organ except the very weekly formed cirrus pouch and metraterm in the metacercarial stage were basically much the same as those in the adult stage described above. The external structure of the cuticle of the metacercariae

was noticeable and different from that of the adults. The cuticle was very thick, much rugged, especially in the forebody, and sometimes covered throughout its surface thinly with the slender spines of about 0.019 mm in length (Fig. 6).

Group I (Figs. 7-9): Not encysted. Body usually swollen, 1.14-2.37 mm long by 0.44-1.00 mm wide, reddish in colour when alive. Both suckers often transformed into a plate. Oral sucker 0.13-0.25 mm long by 0.17-0.25 mm wide. Ventral sucker in anterior onethird of body, 0.20-0.34 mm long by 0.23-0.40 mm wide; sucker width ratio 1:1.14-1.64. Pharynx 0.09-0.14 mm in diameter. Oesophagus short. Intestinal caeca usually expanded broadly, sometimes filled with food, ending in front of ovary. Reproductive system confined within posterior tenth part Testes symmetrical, right testis 0.04-0.08 mm in diameter, left testis 0.04-0.09 mm in diameter. Pars prostatica 0.06- $0.10 \, \mathrm{mm}$ long. Ovary 0.03-0.07 mm in diameter. Vitellaria not yot differentiated.

Group II (Figs. 4-6): Not encysted. Body 2.09-3.68 mm long by 0.66-1.09 mm wide,

reddish in colour when alive. Oral sucker 0.20–0.32 mm long by 0.25–0.34 mm wide, with thickening of dorsal brim more prominent than that in adult. Pharynx 0.11–0.18 mm in diameter. Intestinal caeca generally empty, ending in front of ovary. Ventral sucker 0.26–0.49 mm long by 0.32–0.56 mm wide; sucker width ratio 1:1.29–1.67.

Reproductive system fully formed, confined within posterior one-third of body. Testes symmetrical, right testis 0.06–0.21 mm long by 0.06–0.15 mm wide, left testis 0.07–0.19 mm long by 0.06–0.14 mm wide. Vasa efferentia and vas deferens completely formed. Pars prostatica 0.13–0.21 mm long, far behind ovary, lying longitudinally between both testes. Cirrus pouch still tubular, surrounded by gland cells; ejaculatory duct and cirrus not yet differentiated. Genital atrium funnel-shaped, small. Genital pore median,

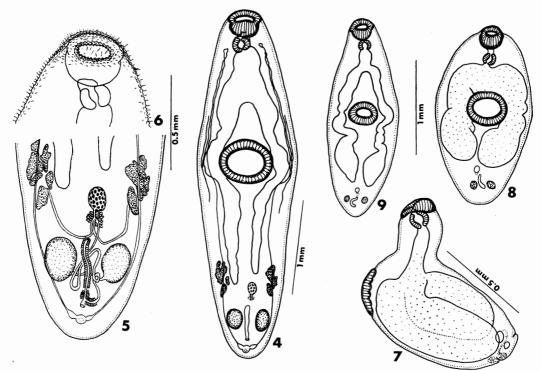
in front of excretory pore.

Ovary 0.05-0.10 mm in diameter, median, behind posterior ends of intestines. Ootype-Mehlis' gland complex and Laurer's canal differentiated. Uterus fully formed, folded between both testes. Metraterm tubular, short, surrounded by gland cells. Vitellaria shortly extending, lateral to posteriormost region of intestines.

Excretory system similar to that in adult stage. Flame cell formula too intricate to permit revealed.

Discussion

The present adult and metacercarial specimens oviously belong to the genus *Amblosoma* Pojmańska, 1972 of the subfamily Leucochloridiomorphinae Yamaguti, 1958, the family Brachylaimidae Miller, 1936. In the genus *Amblosoma* has been so far



Figs. 4-9 Metacercariae of Amblosoma suwaense sp. nov.

Fig. 4 Fully formed metacercaria, ventral view.

Fig. 5 Reproductive system of specimen of Fig. 4, ventral view.

Fig. 6 Anterior portion of metacercaria, ventral view.

Figs. 7-9 Younger metacercariae.

described the type and only species, A. exile Pojmańska, 1972. The metacercaria of A. exile occurs on* the hepatopancreas of the fresh-water snail, Viviparus viviparus L. (Viviparidae), and its adult parasitizes the cloaca of the water bird, Aythya fuligula L. (Anatidae), in Poland (Pojmańska, 1972).

The present trematode fairly resembles A. exile in morphology and in biology. But it differs from the latter mainly in possessing the larger body size, the smaller cirrus pouch and the smaller egg size in the adult stage, and further in having the larger body size and the less developed cirrus pouch and metraterm in the metacercarial stage.

The parthenita(e), cercaria and natural adult worm of *Amblosoma suwaense* sp. nov. have not been found up to now. Studies on the full life-cycle and ecology of the new species in Lake Suwa and its neighbouring waters are in progress.

Summary

The morphology of the adult and metacercarial stages of a new digenetic trematode, Amblosoma suwaense sp. nov. (Brachylaimidae: Leucochloridiomorphinae), is described and illustrated. The metacercariae were found unencysted in the space between the digestive gland and shell of the freshwater snail, *Sinotaia quadrata* (Benson) (Viviparidae), in Lake Suwa and its neighbouring waters, Nagano Prefecture. The adults could be obtained experimentally by inoculating the metacercariae on the chick chorioallantois. The new species differs from the only affined species, *A. exile* Pojmańska, 1972, in possessing the larger body size, the smaller cirrus pouch and the smaller egg size in the adult stage, and further in having the larger body size and the less developed cirrus pouch and metraterm in the metacercarial stage.

Acknowledgement

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^{*} Pojmańska (1972), in the diagnosis of the genus (p. 43), erroneously located the habitat of the metacercaria "in", in spite of "on", the hepatopancreas of the snail host.

新しい吸虫 *Amblosoma suwaense* sp. nov. の成虫とメタセルカリア について (Trematoda: Brachylaimidae)

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諏訪湖と近接水域に 棲息するヒメタニシ Sinotaia quadrata (Benson) の消化腺と貝殻との間の空所から、非被嚢性メタセルカリアを見出した. メタセルカリアを鶏卵漿尿膜上で培養して、成虫をえることができた. この吸虫は近縁種 Amblosoma exile Pojmańska, 1972に比較して、成虫期では体全体が大きいこと、陰茎嚢が小

さいこと、そして卵が小さいことが、またメタセルカリア期では体全体が大きいこと、そして陰茎嚢とメトラテルムの発育が劣ることが認められた。したがつて、この吸虫を新種 Amblosoma suwaense sp. nov. として、成虫とメタセルカリアの形態を記載した。