THE LIFE HISTORY OF PLAGIORCHIS JAENSCHI, A NEW TREMATODE FROM THE AUSTRALIAN WATER RAT

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Summary

1. An account of the anatomy of *Plagiorchis jaenschi* from the Australian water rat, *Hydromys chrysogaster* Geoff., var. *fulvolateralis* Gould, is given.
2. Two apparently distinct forms are shown to be the same, the differences being attributable to the state of preservation of the material.
3. Infection of *Lymnaea lessoni* was accomplished experimentally. This is believed (but not proved) to follow ingestion of the eggs.
4. The various stages in the life cycle, excluding the miracidium, are described.
5. The cercaria encysts in mosquito larvae, and also in crustaceans, *Daphnia*, *Chiltonia* and *Cherax*, of which the last-named, the yabbie, is believed to be the normal secondary intermediate host. Similar cysts have been found in yabbies from Tailem Bend. The cercaria also encysts, though rarely, in the liver of the host snail.
6. Natural occurrences of the cercaria are recorded from Wood's Flat and Bow Hill; but it has not been found at Tailem Bend.

The type as well as other representatives of the adult and larval stages have been deposited in the South Australian Museum. Acknowledgment is made of the generous assistance rendered by Messrs. G. G. and B. Jaensch of Tailem Bend and J. Brook of Bow Hill. The investigation was carried out in connection with the Commonwealth Research Grant to the University of Adelaide.

Plagiorchis jaenschi has been found in the lower part of the small intestine of six out of seven water rats, *Hydromys chrysogaster* Geoffr., var. *fulvolateralis* Gould, trapped by Messrs. G. G. and Bryce Jaensch along the banks of the River Murray at Tailem Bend—May 1938, April 1945, March 1947, May 1948, December 1949 (in one of two taken) and April 1950; also in all three *Hydromys* obtained for us by Mr. J. Brook from Bow Hill, near Mannum, also on the Murray, in April 1950. A water rat from Berri, May 1948, and one from the River Torrens, Adelaide, in July 1923, did not contain this trematode. The maximum number obtained on any one occasion was eighty, the worms being in various stages of growth. Fragments of the Murray prawn, *Palaemon australis*, and the yabbie, *Cherax destructor*, were usually present in the digestive tract. Because of unavoidable delay in examining the dead rats, the worms had in some cases become rather macerated before being collected, but these relaxed parasites permitted a more ready examination of the reproductive system. In two cases the parasites, when found, were much smaller and very strongly contracted, so that the reproductive organs were crowded into a small space. These latter worms were well preserved. Because of the marked dissimilarity in size and appearance it is proposed to refer to the two forms separately, the relaxed worms being described first. The type specimen belonging to this latter group is deposited along with paratypes in the South Australian Museum.

Egg-bearing specimens with pigmented vitellaria ranged from ·85 to 1·43 mm. in length and ·23 to ·37 mm. in maximum breadth, most of them being between ·93 and 1·0 mm. long and about ·3 mm. wide. Worms which reached

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the egg-bearing stage but which still contained very few eggs (1 - 20), measured from ·70 to ·77 mm. in length and ·2 to ·24 mm. in breadth. Nearly all relaxed specimens under ·74 mm. in length had not yet become ovigerous, although a worm ·71 mm. by ·22, and another ·73 by ·22, both possessed yolk follicles almost as numerous as they were in the smallest egg-bearing parasites. A worm ·72 mm. long possessed a single egg and its vitellaria were scanty, linear in arrangement and devoid of pigment. The worms were broadly rounded anteriorly, widest at the level of the oesophagus, and tapering very gradually to the level of the posterior testis, when they narrowed rather rapidly to terminate in a rounded tip.

The ventrally-directed oral sucker in ovigerous worms is approximately spherical, ·16 - ·17 mm. in diameter, and the acetabulum ·125 - ·15 mm. in diameter, though sometimes both suckers may be slightly wider than long or longer than wide. The oral sucker is thus somewhat larger than the ventral. The front end of the latter in mature worms is at about one-third of the body length. In immature relaxed worms the lengths of the anterior and posterior suckers are in the ratio of about 4 : 3, but in ovigerous specimens it is about 6 - 7 : 5. In worms ·36 to ·54 mm. long the oral sucker is about ·1 mm. in diameter and the acetabulum ·075 mm. These dimensions become increased (·175 by ·125 - ·15 mm.) as the worms become ovigerous, but do not increase further though the parasites may become much longer. It is more particularly the postacetabular region which increases in length as growth proceeds. In a worm ·36 mm. long the length of that region is 35% of the total body length; in those ·43 - ·47 mm. long, 39%; in a worm ·54 mm. long, 43%; in a worm ·73 mm. long and almost ovigerous, 47%; in a worm ·74 mm. long and just ovigerous, 59%; in a worm 1·43 mm., 58%. The sex pore is at about ·35 mm. from the head end, and close behind the point of bifurcation of the intestine. Spination, though evident in contracted worms, had disappeared from all relaxed specimens. Young worms may show the typical Y-shaped excretory bladder in preserved material.

The prepharynx is very short and lies above the hind edge of the oral sucker. The pharynx is spherical (·08 mm. diameter) or slightly elongate. The narrow oesophagus is very short, measuring about one-quarter or one-fifth of the length of the pharynx. The crura diverge almost at right angles to the longitudinal axis of the body and then curve posteriorly to lie more or less parallel with the edge of the body, their course being slightly sinuous. They terminate beside the posterior part of the stem of the excretory bladder.

The testes are each about ·17 mm. in diameter, though the posterior one may be rather narrower. The cirrus sac is relatively large and takes its origin at about the ovarian level, but on the opposite side. It then curves around and above the right side of the acetabulum, then inwardly and downwards, and approaches the metraterm as the two organs pass forwards to the genital pore. The sac passes directly ventrally to enter the latter. The length, measured along the curve, is about ·45 mm. and its breadth about ·07 mm. The posterior half or two-thirds of the organ serves as a seminal vesicle. Occasionally the sac lies in a more median, sinuous position, crossing above the acetabulum. The cirrus, when at rest, is closely folded in the anterior part of the sac. The fully extruded unarmed cirrus is about ·46 - ·53 mm. in length, and its width 15 - 17 μ except in the vicinity of its base where the breadth is about 42 μ.

The rounded ovary, ·08 by ·1 mm., is situated on one side a short distance behind the acetabulum, portion of the cirrus sac lying between the two organs. The oviduct passes inwards and posteriorly to meet the common yolk duct in the vicinity of the shell gland, the latter lying between the ovary and anterior testis. The uterus soon bends posteriorly and travels in a sinuous course between the testes, reaching almost to the level of the ends of the crura before returning

as the ascending limb which lies beside the descending limb. About the level of the shell gland, the ascending duct is thrown into a few loops as it makes its way to the left side of the worm. It passes below the inner end of the cirrus sac and then forwards in a ventral position beside (or sometimes above) the acetabulum. The metraterm is thin-walled. The yolk glands are extensive, covering the crural region and almost reaching the lateral edges of the worm. They extend from the level of the oesophagus to the free ends of the crura, terminating just behind, or just in front of, the posterior loops of the uterus. The follicles are irregularly rounded (12 - 15 μ diameter) or elongate. The two fields remain separate though some scattered follicles may occur in the oesophageal region between the lateral fields. The latter may approach very closely at their posterior ends. If we recognise the subgenus *Multiglandularis*, which is based on the distribution of the vitellaria, then our species would be *P. (M.) jaenschi*. The main yolk ducts lie dorsally and transversely just in front of the anterior testis. There is a definite yolk reservoir. Eggs are 30 - 37 μ long by 17 - 22 μ wide, most of them being 32 - 34 μ by 17 - 20 μ.

Strongly contracted worms taken in June 1941 were, as stated above, well preserved when compared with those just described. The spines were short but abundant on the anterior region, becoming very low and scarcely recognisable under high power behind the level of the acetabulum. Similar spination was seen on metacercariae. Egg-bearing worms measured ·21 - ·56 mm. long by ·145 to ·25 mm. broad, the smaller individuals tending to be relatively wider than the longer. Those which were not ovigerous ranged from ·14 to ·31 mm. in length and ·088 to ·22 mm. in width. All strongly contracted worms were plump, with broadly rounded extremities and with a more or less marked ventral concavity involving the acetabulum and the region in front of it, sometimes extending to the oral sucker. Two cysts found in one host animal containing contracted worms measured ·11 by ·087 mm. and ·15 by ·112 mm., the latter having the stylet still in situ. A metacercaria freed from a cyst was ·137 by ·12 mm. These sizes fall approximately within the range of dimensions of cysts obtained from experimentally infected crayfish (120 μ by 98 μ to 173 by 158 μ; average 150 by 124 μ).

In the larger strongly contracted worms the suckers tend to be rather broader than long, the oral sucker up to ·075 mm. long and ·12 mm. broad, and the acetabulum ·07 by ·095 mm., the latter tending to become nearly as wide as, but shorter than, the oral sucker. In smaller mature worms the dimensions of the suckers are less than those just given. The pharynx may overlap both suckers in very small worms, but in those which are somewhat larger the organ lies above the posterior end of the oral sucker.

The various organs in these strongly contracted worms occupy the same relative position as in relaxed specimens, but the testes (and to a less extent, the ovary) are markedly compressed to become transversely elongate. Eggs in such worms fall within the range found in the more elongate material (30 - 32 μ by 19 - 20 μ).

Our species is closely related to *P. muris* (Tanabe 1922) from rats and mice in Japan. Tanabe's Japanese paper was translated by Dollfus (1925), who reproduced the figures also. Yamaguti (1933, 106) added further details. Hirasawa and Asada (1929, 507) dealt with its life cycle. Olsen 1937, pl. v. 73) republished Dollfus' figure of the adult worm. McMullen (1937a, 113) reported experimental infection of rats, mice, pigeons and man with *P. muris*. The vitelline follicles are indicated in Dollfus' figure as extending a little further forward than in McMullen's (1937 b) figure. Tanabe stated that the intermediate hosts in Japan were *Lymnaea pervia* and Chironomid larvae. McMullen (1937 b, 239) briefly described the species from North American material, as also did Cort and Ameel (1944, 37 - 48). Ishii (1935, 629) in an extensive survey of the rats

(varieties of *Rattus norvegicus*) occurring in the drains and rivers of Tokyo, found four species of trematodes; three of them were echinostomes and the fourth was *Clonorchis sinensis*, but *Plagiorchis muris* was not mentioned.

If Olsen's key (1937) to the species of *Plagiorchis* be used, our parasite would be accommodated beside *P. muris*. The latter was placed by Schulz and Skworzow (1931, 773) in their subgenus, *Multiglandularis*, and Olsen has followed them.

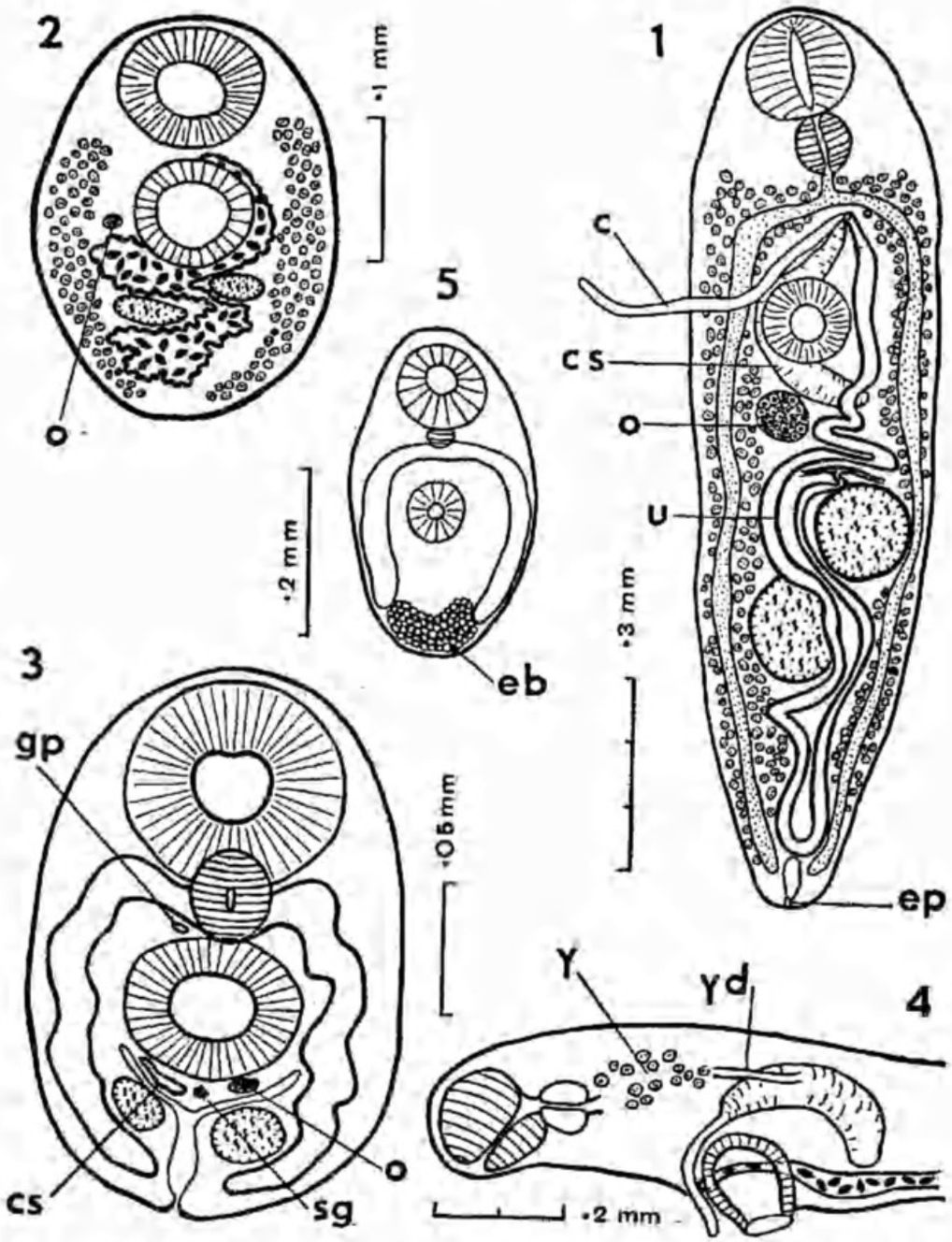

Fig. 1, adult; 2, strongly contracted adult; 3, very young, contracted worm; 4, anterior end, side view; 5, metacercaria, unfixed, somewhat compressed (expressed from a cyst from *Lymnaea lessoni*).

REFERENCE TO LETTERING

al, alimentary caecum; c, cirrus; cs, cirrus sac; eb, excretory bladder; ep, excretory pore; gp, genital pore; o, ovary; ov, probably ovary and shell gland complex; r, refractile granules; sg, shell gland; t1, t2, testes; u, uterus; y, yolk field; yd, vitelline duct.

P. (M.) jaenschi, in its relaxed condition, differs from *P. (M.) muris* in the following features: much shorter and narrower dimensions; much smaller oral and ventral suckers, testes and ovary; genital pore close to the intestinal bifurcation; ovary nearer the acetabulum; posterior testes more remote from the end of the worm; and yolk follicles less abundant at the posterior end where the two fields are almost separated.

Experimental Infections

Lymnaea lessoni[1] has been infected with eggs of *Plagiorchis jaenschi* in three separate experiments. The eggs were dissected out from the adult worms and placed in contact with various molluscs, after varying periods. A summary of all experiments performed follows:—

Experiment A. 26/3/47. Eggs (dissected out from a number of adults from Tailem Bend) put in a petri dish containing snails, including 8 *Lymnaeae*. Snails isolated over 24-hour period once a week from eighth week onwards; after 15 weeks one of two surviving *Lymnaeae* emitting large numbers of cercariae. The second snail died in 21 weeks; and although no cercariae had been recovered during the weekly testings, the liver was found to contain many sporocysts and some cercariae which were probably immature. (The six other *Lymnaeae* died after two to nine weeks; none showed sporocysts on dissection.)

Experiment B. 20/12/49. Eggs (Tailem Bend) dissected out and placed on lettuce in dish containing 6 *Lymnaeae* and other snails. The 6 *Lymnaeae* died within 10 days. Sectioning revealed no evidence of development, or even of ingestion of the eggs.

Experiment C. 14/4/50. Eggs teased from 8 adults (Tailem Bend); kept in water for four days, then fed on lettuce to various snails. Of these, the four *Lymnaeae* died in 4, 16, 17 and 21 days; they were dissected carefully, but no signs of infection were noted.

Experiment D. 17/4/50. Eggs from eight adults (Tailem Bend) teased out; kept in water for nine days (26/4/50) before feeding on lettuce to snails. Snails isolated over 24-hour period once per week from 24/5/50. *Lymnaea* (1) emitted cercariae on 14/6/50. (Infection period thus between six and seven weeks.) The three remaining snails were then kept isolated in tubes (being fed on lettuce). *Lymnaea* (2) gave cercariae on 15/6/50; and *Lymnaea* (3) on 21/6/50. *Lymnaea* (4) died on 7/7/50 (*i.e.*, 72 days after infection); on dissection it was found to be harbouring cercariae and a few sporocysts.

Experiment E. 27/4/50. About 18 adults (Bow Hill) were teased up (in the dish in which they had been lying for several days, and which therefore probably contained some naturally laid, *i.e.*, mature, eggs as well). On 4/5/50 the eggs were pipetted on to lettuce in a dish with 6 *Lymnaeae* and other snails. The snails were isolated once per week from 24/5 to 21/6/50, and thereafter examined daily. Three of the *Lymnaeae* became infected; two gave off cercariae after 67 and 74 days respectively, the infection being very light in both cases. The third snail died after 104 days; it had never given off cercariae, but on dissection was found to contain a number of immature sporocysts.

It is believed that infection of the snails occurs by ingestion of the eggs, as is the case with many Plagiorchids. In Experiment D, *Lymnaea* (1) emitted cercariae six to seven weeks after it had been placed in contact with eggs. This is regarded as a comparatively short period for development at this time of the year (April to June). If a free-swimming miracidium were produced, this period would necessarily be shortened.

[1] The Lymnaeae were known to be uninfected, having been hatched and reared in the laboratory.

According to McMullen (1937 b) Tanabe reported that the fully developed miracidium of *Plagiorchis muris* was produced in 96 hours. Dollfus, 1925, reported that the egg when laid contained an egg-cell and 5 - 6 yolk-cells, and that development of the miracidium was completed in four days at 37°. He did not state whether the miracidium hatched at this time, however. McMullen (1937 b) kept eggs of the American species, from man, at the prevailing August and September temperatures in Michigan, and noted that they did not produce miracidia under 24 days. Our experiments did not show what period after the egg is laid, if any, is required before the miracidium becomes viable.

Other molluscs used in the experiments were *Amerianna* spp., *Planorbis isingi*, and *Notopala hanleyi*. None of these became infected.

C. Plagiorchis jaenschi had not been identified as a natural infection of *Lymnaea lessoni* until April 1949, when it was found in 5 of 45 *Lymnaea*, from Wood's Flat. It was found at Bow Hill in February and April 1950, in 1 of 1,072 and 1 of 460 of the snails, respectively. It has never been recorded from Tailem Bend, where it would not be expected to be a common form, in view of the few *Hydromys* which now frequent the swamps in that region; though cysts, believed to belong to *Plagiorchis jaenschi*, have been found twice in yabbies from Tailem Bend. It should be noted that once or twice we have noted "stylet cercariae" from *Lymnaea*, without further identification; on the other hand, whenever these cercariae were examined more closely, they proved to be a form with stylet 24 μ long. This latter form is the only common xiphidiocercaria found by us in *Lymnaea lessoni*.

The Sporocyst

Sporocysts were present in the livers of the snails in large numbers in the original infection, but less in the later experimental hosts. They are comparatively small, each containing from 2 - 4 cercariae. Average of 7 (not fixed) 645 by 202 μ; range 450 to 825 μ by 150 to 240 μ.

The cells of the sporocyst wall, and the contained cercariae, stained well with a weak solution of basic fuchsin in normal saline. No germ balls were seen; and no details of the excretory system were determined. Tiny green refractile droplets were present throughout the sporocyst wall.

The Cercaria

In general, cercariae emerge from the snail in the early hours of the morning, mostly before 9 a.m. (On one occasion, when no cercariae had been given off earlier in the day, quite a large number emerged between 1 and 2 p.m.)

For the rest of the morning, the cercariae remain suspended in the water, swimming only occasionally. After this they become concentrated at the bottom of the test tube, some of them fastened by the suckers to the tube; sometimes they appear to favour the side nearest the light, but at other times there is no obvious phototactic response. At the end of the day they lie on the bottom of the tube; some of them even attempt to encyst. (It is doubtful whether such encystation preserves their lives, as there was no movement in those "metacercariae" which were removed from the rather ragged, gelatinous coating. Cyst formation was observed several times on the slides—adverse conditions, such as drying out, or perhaps the pressure to which the cercaria was exposed under a coverslip, appearing to stimulate it.) The length of free life may, however, be longer than in many cercariae, for, from the first snail to be infected, a number of cercariae over 48 hours old successfully encysted in *Daphnia* sp.

Measurements of formalinised specimens were subject to some variation, due to the varying states of expansion in which they were fixed. A series of 20 which had been killed at 5 p.m. by adding boiling 10% formalin to an equal

volume of a very weak solution of neutral red, in which they had been placed for ten minutes, ranged from 133 to 187 μ in body length, and from 94 to 148 μ in breadth; the average size being 155 by 120 μ. Ten cercariae similarly treated, but killed at 11 a.m., ranged from 150 to 188 μ in length and 98 to 128 μ in breadth; the average size being 173 by 113 μ. (That is, early in the day the cercariae appear to be fixed in a more extended condition.) The tail measurements are only approximately accurate, due to the fact that the tails were seldom fixed in a perfectly straight condition. They ranged in length from 102 to 162 μ, and in breadth from 26 to 36 μ, an average of 24 specimens (from cercariae fixed in both morning and afternoon) being 132 μ by 29 μ. The measurements of four obviously extended specimens were:—body 255 to 285 μ long and 82 to 90 μ wide (average 270 by 86 μ); tail 158 to 180 μ long and 26 to 32 μ wide (average 172 by 29 μ).

The oral sucker was larger than the ventral, approximate measurements for the former being 41 μ long by 53 μ wide (average of 20 specimens); and for the latter 32 μ long by 38 μ wide (average of 10 specimens).

The stylet (fig. 3) is 34 μ long; 9·5 μ across the widest point, and 6·5 μ across the base. The thickened rim is about 3 μ deep. It does not extend right round the stylet, being incomplete on one face. It starts 10 μ from the tip and from 13 μ the remainder of the "collar" narrows in gradually to a point 15 μ from the base, from which the width remains uniform.

About 20 μ of the length of the tail is enclosed within the body in a caudal pocket which is lined with spines. The pocket may be well retracted into the body, or may be opened out so that the hinder part of its side walls comes to lie on the posterior end of the body. In this region of the pocket the spines, probably about ten on each side, are much longer than elsewhere, and may be seen even in the retracted condition. In the everted position of the pocket they appear as short bristles. After prolonged exposure to a neutral red solution so dilute that it was almost colourless, the walls of the pocket were consistently stained an orange colour, though no other parts of the body were affected by the stain in this dilution.

The general body surface is covered with extremely minute spines which are more prominent anteriorly, and cease to be really obvious at about the acetabular region. Greenish refractile granules, which are mostly fairly large, are more or less evenly distributed throughout the body, except in the region anterior to the pharynx, which is free of them (fig. 7).

There is a mass of gland cells extending from about mid-way between the two suckers to just beyond the posterior border of the acetabulum (fig. 7). Intravitam staining was not of much assistance in determining the number of these cells, which is thought to be at least 10 pairs, and possibly a number more. With neutral red, the three or four large posterior cells show very fine granulation, while the anterior mass takes up the stain quite deeply. The nuclei appear quite clear, and very faintly pink, after basic fuchsin and normal saline. The ducts of the gland cells are often bent upon themselves, or dilated here and there in their course. Some of these, also, show a fine granulation. They run forward together, becoming narrower as they do so, to open near the mouth. Methylene blue stained the cercaria a uniform, very faint, blue, but showed no differentiation at all.

The prepharynx is short and very inconspicuous; the pharynx quite well defined and muscular, but the oesophagus and alimentary crura so inconspicuous that in living specimens there is rarely any indication of their presence. In stained, fixed, specimens they can be distinguished as very narrow structures, one cell in thickness, so that even the existence of a lumen is problematical. The caeca appear to diverge abruptly from the oesophagus.

The excretory system is seen best with the aid of basic fuchsin in normal saline. Cercariae will live all day in good condition for examination in very dilute solutions. Horse serum in addition is also helpful. The posterior part of the Y-shaped excretory bladder opens into the vestibule, from which the excretory pore opens on to the surface of the body. The main excretory tubes open into the cornua of the bladder at the tips, but we had considerable trouble in determining

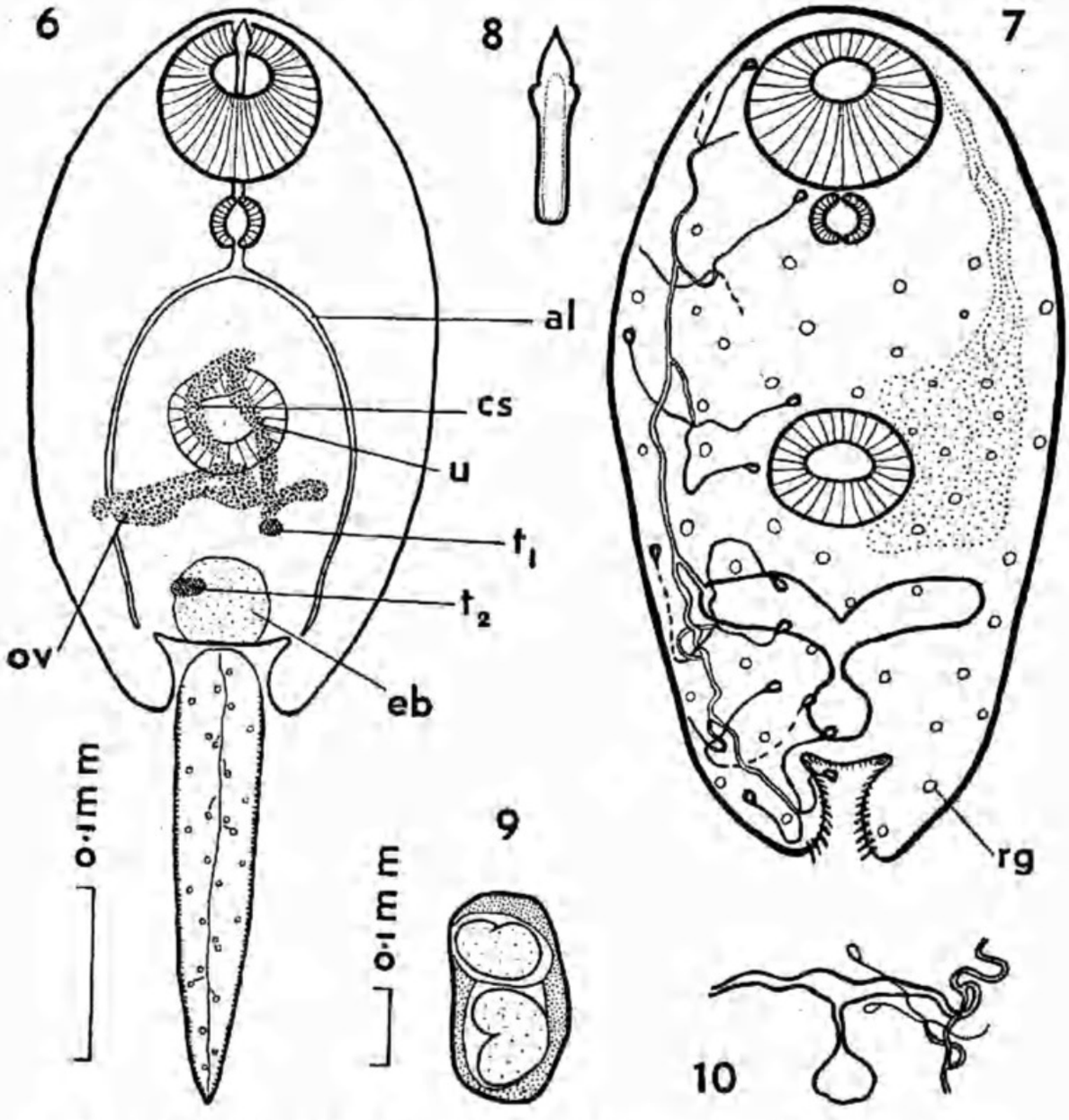

Fig. 6, 7, cercaria: 6, fixed specimen stained with borax carmine in lacto-phenol; 7, details from living specimens. Fig. 8, stylet; 9, two cysts in common envelope; 10, one of the variations in the shape of the bladder.

Fig. 6, 7, to same scale; 8, sketch; 10, sketch.

this because of the presence of cystogenous cells and refractile granules in the area, the twisting of the main and accessory excretory tubes, often over the end of the cornua, and the variability in shape and definition of the arms of the bladder. We mention this to illustrate that in some cercariae, without prolonged study too much reliance cannot be placed on the supposed point of entry of the main excretory tubes.

The canals divide into anterior and posterior collecting tubules at the level of the anterior borders of the excretory bladder. There are three groups of three flame cells connected with each of the anterior and posterior collecting tubules. The excretory formula is thus 2 ($(3+3+3)+(3+3+3)$). Although the figure (fig. 7) shows that in some of the groups the whole six elements (three tubules and three flame cells) were not seen in the cercaria, the determination of the excretory formula was made with additional information supplied by a study of a metacercaria seven days old, in which no less than 27 of the 36 flame cells were seen, the positions of these filling in the gaps left in the excretory picture of the cercaria.

The genital primordium consists of a collection of quite undifferentiated cells, but from the relative positions and arrangement of these cells the anlagen of the adult organs can be diagnosed (fig. 6).

The Cyst

The cercariae have been found, experimentally, to encyst in *Daphnia* sp.; the amphipod, *Chiltonia subtenuis;* the yabbie, *Cherax destructor,* and in mosquito larvae; but not in the molluscs, *Amerianna* spp., *Planorbis isingi, Lymnaea lessoni, Platiopsis tatei* and *Hyridella australis,* or in tadpoles of *Limnodynastes* sp. or the fish, *Gambusia affinis.* In the liver of one of the *Lymnaeae* naturally infected with the cercaria (from Bow Hill) , three cysts which fell within the size range for *Plagiorchis jaenschi,* and contained stylets of the same size, were found; these were obviously cysts of *Plagiorchis jaenschi.* The metacercaria expressed from one of them is figured in fig. 5. McMullen (1937 a, b,) reported that cercariae of *P. muris* and *P. proximus* encysted within the sporocyst. We have not observed this with *P. jaenschi.*

Among the four arthropods, encystation takes place most readily in the yabbie, and this is probably the natural secondary intermediate host, forming, as it does, a large part of the food of *Hydromys.* On at least two occasions (in April 1939 and February 1940) we have found cysts in the gills of yabbies from Tailem Bend. These resemble those of *P. jaenschi* in general appearance and size, and in the site which they occupy.

The cysts were found occasionally in the joint region of the legs, but were predominantly in the main stem of the gills. Sometimes two, three or four were enclosed in series in the same piece of integument, though each had its own cyst wall (fig. 9). The cysts vary in shape from spherical to elliptical. In a series of 15 from the original infection the size range was from 120 by 98 μ to 173 by 158 μ; cysts of the naturally occurring form from Bow Hill ranged from 120 by 98 μ to 210 by 128 μ, and even to 200 by 180 μ in one cyst from a mosquito larva.

The Metacercaria

A metacercaria seven days old was excysted successfully; as mentioned in the description of the cercaria, the excretory formula was thought to be no further advanced. The stylet was still in position in the oral sucker, as it appeared to be in two other cysts of the same age. Metacercariae which were liberated from cysts three months old were relatively small and showed no very great advance in development in those features which could be studied in stained specimens. The genital primordium did not show up even as clearly as in the cercaria, but appeared as a U-shaped string of cells situated in the region dorsal to the acetabulum; this is probably the primordium of the cirrus sac and metraterm. In an average of three stained specimens the oral sucker measured 53 μ long by 68 μ wide, and the acetabulum was 27 μ long by 39 μ wide. We were unable to excyst these metacercariae in sufficiently good condition for studying the excretory system.

In the metacercariae obtained from the host *Lymnaea* (of which, of course, the age is not known) the alimentary system had developed considerably and closely resembled that of the adult. Although the metacercariae were motile, we were unable to determine any excretory detail.

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