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HALIPEGUS DUBIUS KLEIN, 1905 (TREMATODA, HEMIURIDAE): A REDESCRIPTION, WITH NOTES ON THE WORKING OF THE OVARIAN COMPLEX

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A redescription of Halipegus dubius *Klein, 1905, of the Brazilian frogs* Leptodactylus pentadactylus *and* L. ocellatus, *is presented. The parasite was found exclusively in the buccal cavity, in numbers varying from 1 (in most instances) to 3, and exceptionally 25 (one instance). Morphological data were based on 40 whole-mounted specimens and 4 serially sectioned ones. Larval development takes place in planorbid snails (*Biomphalaria glabrata *and* B. tenagophila*) and in an undetermined species of cyclops. Working of the ovarian complex was followed by microscopical observation of live specimens.*

Contrasting with the similarity of the four American species of Halipegus *currently recognized as valid, they can be easily separated by the characters of their cercariae.*

Key words: Trematoda – *Halipegus dubius* – taxonomy – working of ovarian complex

In the course of observations on transmitting sites of schistosomiasis in Belo Horizonte, Minas Gerais State, in 1943, I found numerous specimens of *Biomphalaria glabrata* infected with large, very mobile orange-colored rediae producing cysticercous cercariae like those depicted in Figs 2, 3 and 20. Ten years later, again in Belo Horizonte, I found the adult fluke in the buccal cavity of frogs from several of the previously examined breeding places with infected *B. glabrata.* I then spent some time on studying the adult worm and attempting to follow its life cycle, but soon after I gave up that project to become deeply involved in a program of investigation on the taxonomy of planorbid snails.

Owing to their incompleteness, the notes and documentation accumulated in 1953 were set aside. The celebration of Professor Lauro Travassos' centenary leads me to recover from oblivion those notes which deal with one of his favorite subjects of investigation, the trematodes. The following text, written in 1953, is now published with added references to findings and opinions of some subsequent authors.

The occurrence of the genus *Halipegus* in the Neotropical region was first recorded by Lühe (1900) under equivocal circumstances. According to the label accompanying the specimens deposited in the Museum of Berlin, they were found in the Brazilian snake, *Coluber olivaceus,* and were identified by Lühe as *"Halipegus* n. sp.?". Only a single species, the European *H. ovocaudatus,* described by Vulpian (1859) from *Rana esculenta* (referred to as *rana viridis*) and found by Sonsino (1893b) in that frog and *Rana temporaria,* was then known. No description of the new material was produced by Lühe, who confined himself to measuring the specimens and stating that they were indistinguishable from *H. ovocaudatus.* Considering the possibility of mislabeling, he even doubted to regard the Brazilian snake as a new host of *H. ovocaudatus.*

Coluber olivaceus is an enigmatic taxon not recorded in Sherborn's Index Animalium, and unknown to several consulted Brazilian herpetologists.

Klein (1905), describing *Halipegus longispina* of *Rana hexadactyla* from India, referred to Lühe's specimens, naming them *Halipegus dubius,* to be distinguished by the length of the egg filament. He mentions a letter from Lutz to Lühe, in which the former suggested that the Brazilian *Halipegus* should have been originally a parasite of frogs swallowed by the snake and reported having found an identical *"Distomum"* in the frog *Cystignathus ocellatus* (=*Leptodactylus ocellatus*) from Rio de Janeiro, Santos, São Paulo and Montevideo.

Lutz (1928: 109) pointed out the likeness between the developing stages os *H. ovocaudatus* and of the Brazilian species and, seemingly unaware of Klein's proposal, named the latter *Halipegus similis*, though neither describing it nor indicating the differences between the two species.

The first description of *H. dubius* was given by Cordero (1942), who presented figures of an adult specimen and two eggs. His material was taken from the pharynx of the frog *Leptodactylus ocellatus* from Montevideo, Uruguay, and his figure and description were reproduced by Travassos et al. (1969: 659-660). He admitted the validity of the species based on zoogeographic data and chiefly on the egg-filament length ratio.

Ruiz (1952) described a redia-derived cercaria parasitizing *Australorbis* sp. (=*Biomphalaria tenagophila*) from Santos, São Paulo state, naming it *Cercaria hemiura* and considering it very probably to belong to *H. dubius*.

A redescription of this species was published by Kohn & Fernandes (1988).

MATERIALS AND METHODS

This study is based on 52 specimens of *H. dubius* collected from frogs in Belo Horizonte city and adjacencies.

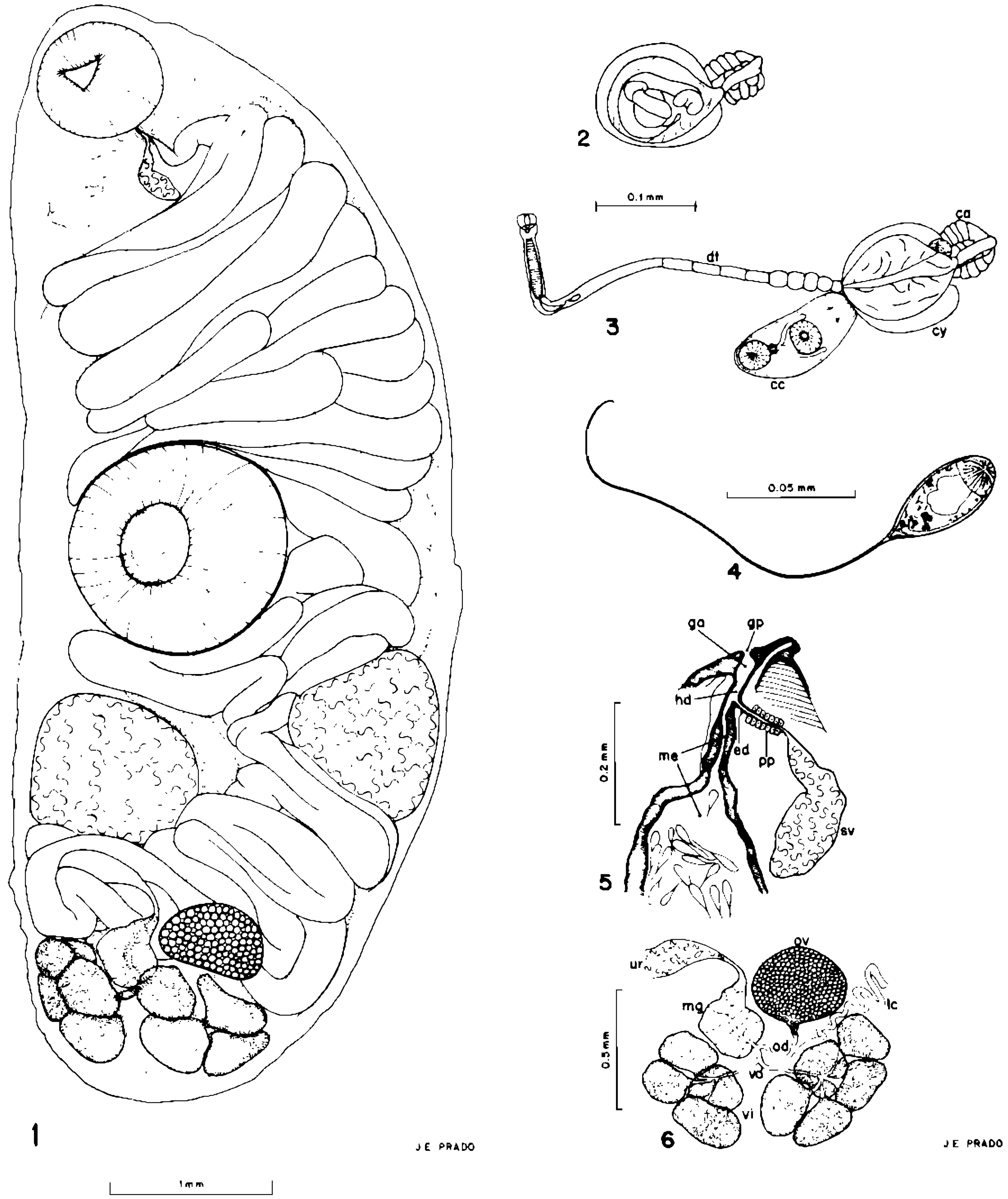
The flukes were present in 15 of 64 *Leptodactylus pentadactylus* (23.4%) and 7 of 41 *L. ocellatus* (17.1%). All of them were situated under the tongue, attached to a side of the mouth floor and showing a marked predilection for the left side. Thus, in *L. pentadactylus* they were on the left in 10 frogs, on the right in 4, and on both sides in 1; in *L. ocellatus* they were on the left in 6 frogs and on the right in 1. Most frogs – 11 *L. pentadactylus* and 5 *L. ocellatus* – harbored a single fluke. Two *L. pentadactylus* had each 2 flukes on the left, one had 3 flukes (1 on the left, 2 on the right), and one had 25 gathered on the left. One *L. ocellatus* had 2 flukes on the left, and another had 1 on the left and 1 on the right.

Forty adult specimens were fixed, after compression, in Railliet-Henry's fluid (formalin 5 ml, glacial acetic acid 2 ml, normal saline 93 ml), stained in bulk with alcoholic HCl carmine, cleared in creosote and mounted in Canada balsam. Measurements were made on camera lucida drawings. Four specimens were fixed in Bouin's fluid, embedded in paraffin, serially sectioned (sagittal sections in 2, frontal sections in 2), and stained with hematoxylin-eosin for microanatomic observation.

The activity of the ovarian complex was observed under the microscope in live specimens mounted in saline under a large cover glass on which weights of adequate heaviness were placed so as to obtain a favorable degree of transparency of the fluke's body.

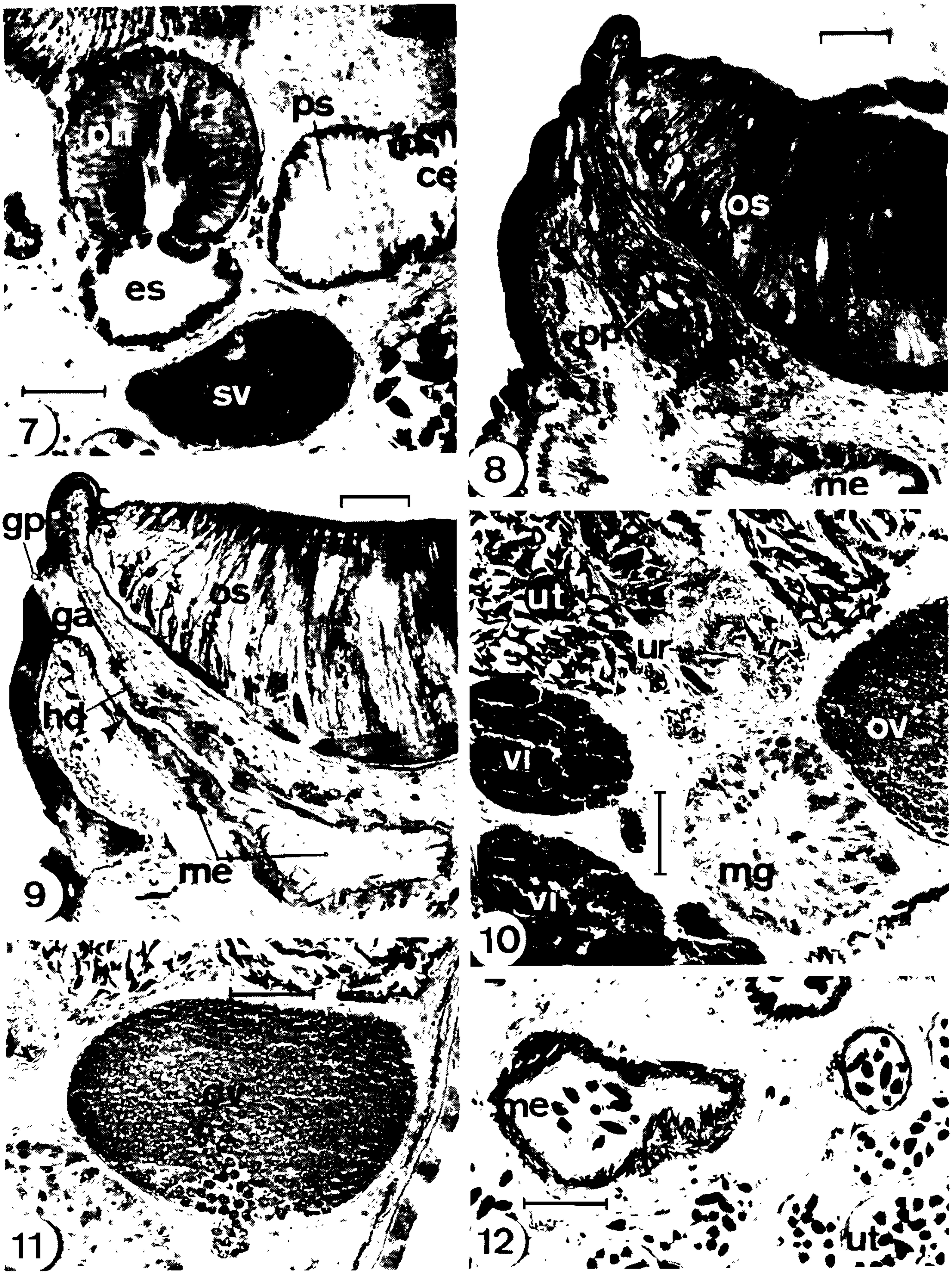
DESCRIPTION

Body medium-sized (Fig. 1 shows an adult specimen 7 mm long), a little constricted near the middle in most specimens. Body surface smooth. Oral sucker subterminal. Absence of prepharynx. Pharynx bulbous, emptying into an extremely short esophagus which has a cuticularized lining (Fig. 7, es). Cecal bifurcation behind the pharynx. Ceca initially wide (prececal sacs), smooth-walled, lined with long-ciliated cells (Fig. 7, ps), then plicate (Fig. 7, ce), lined with digestive epithelium, descending dorsally to the other organs and ending blindly near the caudal extremity. Ventral sucker near the middle of the body, larger than the oral one. Testes ovoidal, preovarian, moderately oblique, sometimes symmetrical, immediately or shortly caudal to the ventral sucker; either testis, right or left, may be larger than or caudal to the other. Seminal vesicle (Figs 5, 7, sv) saclike, free in the parenchyma, about on the midline and near the pharynx. Pars prostatica (Fig. 5, pp) short, tubular, surrounded by non-delimited gland cells (Fig. 8, pp), followed by a still shorter ejaculatory duct (Fig. 5, ed) which joins the metraterm to form a short hermaphroditic duct (Fig. 5, me, hd). Gential atrium small (Figs. 5, 9, ga). Sinus-sac and sinus-organ absent. Genital pore close to the caudal border of the oral sucker (Fig. 9, gp). Ovary egg-shaped, nearly median, dorsal (Fig. 11), just anterior to the vitellaria and separated from the testes by uterine loops, with the long axis at about right angle, seldom oblique, to the long axis of the body. Mehlis' gland (Fig. 10, mg) postovarian (in about 20% of specimens on the right of the ovary, Figs. 1; 6, ov). Laurer's canal (Fig. 6, lc) opening dorsally in the midline. Vitellarium (Fig. 6, vi) comprising two groups of 4 and 5 (exceptionally 3) ovoid follicles with smooth or slightly lobed surface; same number on both sides never observed. Uterus beginning as a uterine semi-

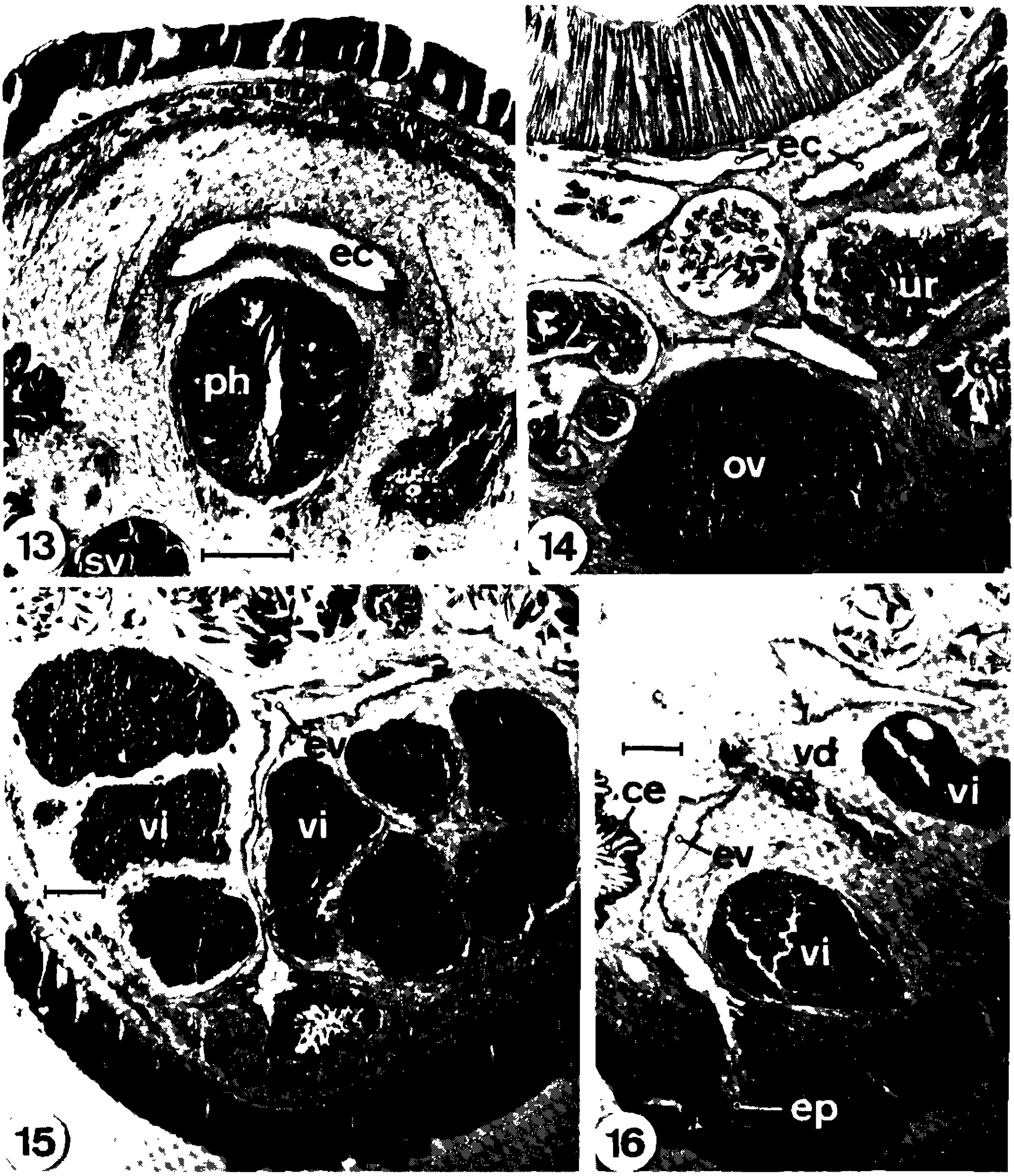


Halipegus dubius – Fig. 1: adult specimen. Fig. 2: encysted cercaria. Fig. 3: cercaria and delivery tube excysted. Fig. 4: egg. Fig. 5: terminal genitalia reconstructed from serial sections. Fig. 6: ovarian complex, from specimen of Fig. 27.

nal receptacle (Figs 6, 10, ur) and extending, packed with eggs, in transverse S-shaped coils from the vitellaria to the cecal bifurcation. Metraterm muscular, not surrounded by gland cells (Figs 5, 12, me) and with ciliated distal portion (Fig. 9, me). Two lateral ciliated excretory canals united dorsally (Fig. 13, ec) behind the oral sucker, each of them running down extracecally toward a lateral side of the ventral sucker; there each canal curves around the corresponding half of the caudal side of the sucker (Fig. 14, ec) to join its counterpart into the Y-shaped excretory vesicle. The latter is situated between the ovary and the vitellaria,



Halipegus dubius – Fig. 7: peripharyngeal region. Fig. 8: oblique section of pars prostatica. Fig. 9: part of terminal genitalia; arrow indicates point where metraterm (me) joins ejaculatory duct (present in other sections) to form hermaphroditic duct (hd). Fig. 10: section through ovarian complex. Fig. 11: oviduct arising from ovary. Fig. 12: metraterm and uterine coils. Bar = 100 μm (Figs 7, 10, 11, 12) and 50 μm (Figs 8, 9).

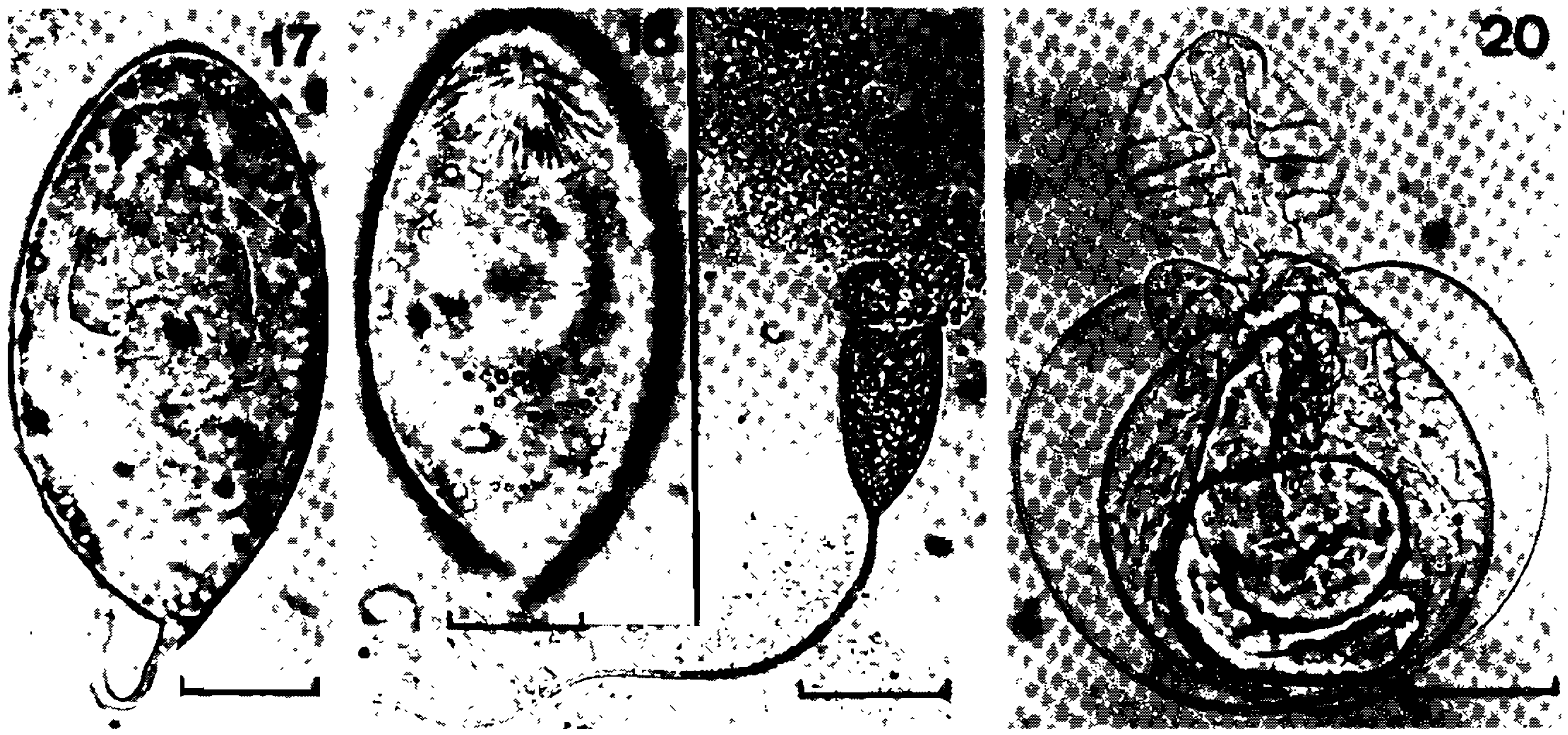


Halipegus dubius – Fig. 13: origin of excretory system (ec). Fig. 14: right and left excretory canals (ec) converging below ventral sucker (vs). Fig. 15: point of convergence (ev) of right and left excretory canals to form excretory vesicle. Fig. 16: excretory vesicle (ev) and excretory pore (ep). Bar = 100 µm.

and its long stem runs between the two clusters of vitelline follicles (Fig. 15, ev) to discharge through the excretory pore (Fig. 16, ep).

Eggs (Figs 4, 17, 18, 19) operculate, with long filament at the anopercular pole, about 1.5-2.7 times as long as the egg in most specimens, rarely more as in Fig. 4 (3.36), and tapering to fine tenuous recurved tip; fully embryonated when laid; miracidium with cephalic crown of penlike spines and covered with minute bristles.

Larval development in snails *Biomphalaria glabrata* (my material) and *B. tenagophila* (Ruiz, 1952), producing cystocercous cercariae (Figs 2, 3, 20). The latter, ingested by cyclops (undetermined species), migrate through the gut wall into the hemocoel. Development of metacercaria not followed.



Halipegus dubius – Fig. 17: egg, focus on apical gland of miracidium. Fig. 18: egg, focus on crown of penlike spines. Fig. 19: egg with detached operculum. Fig. 20: cercaria. Bar = 10 µm (Figs 17, 18) and 25 µm (Figs 19, 20).

Adult in the buccal cavity of frogs *Leptodactylus pentadactylus* and *L. ocellatus* from Belo Horizonte, Minas Gerais state, Brazil.

Measurements in Table.

TABLE

Measurements in mm of body, organs and eggs of *Halipegus dubius* from *Leptodactylus* frogs from Belo Horizonte, Brazil. *N* = 40, except pharynx (37), seminal vesicle (28) and Mehlis' gland (33)

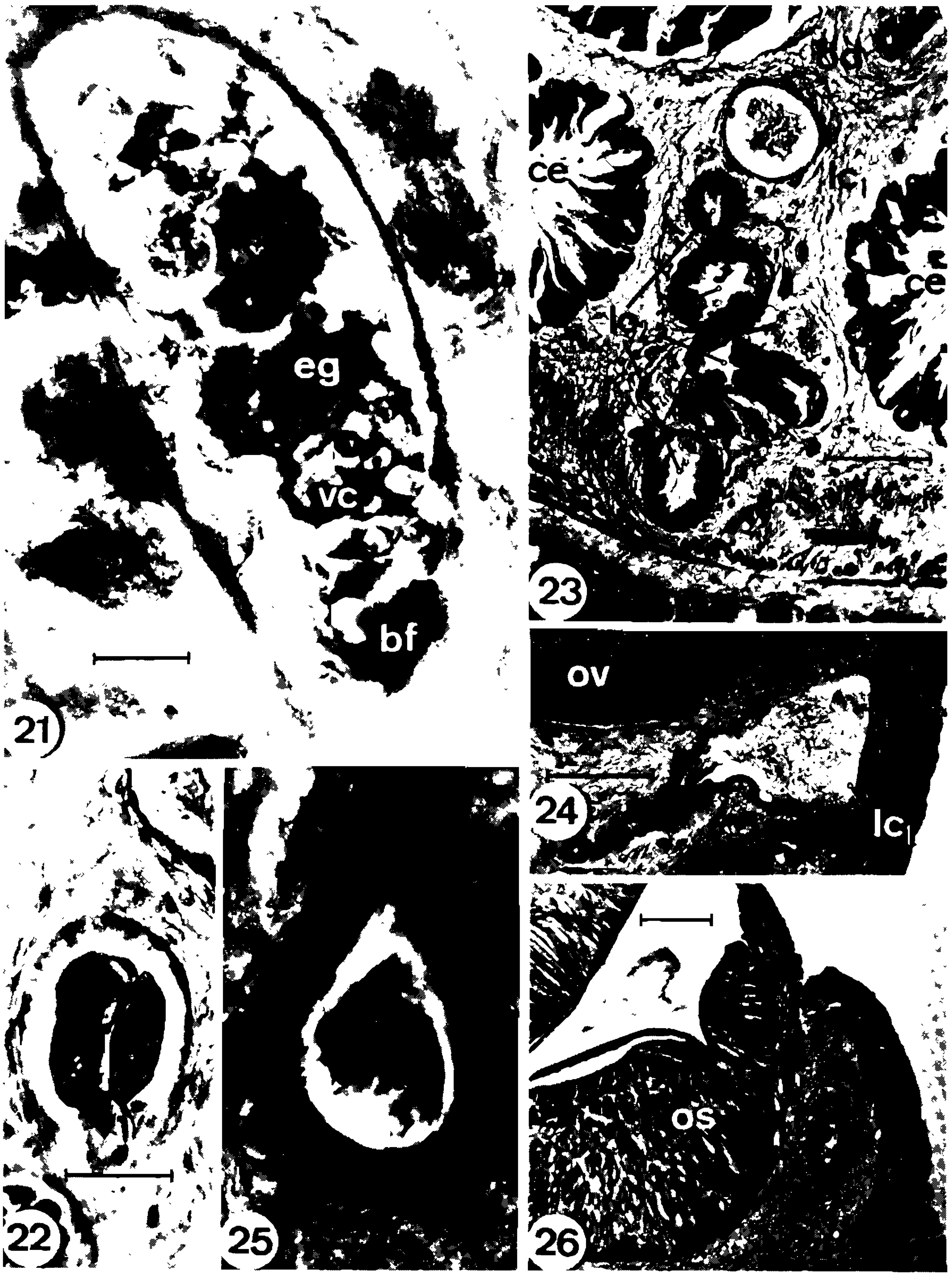
	Range	Mean	± s.d.
Body length	5.00-8.40	6.50	0.92
Body width	1.30-3.40	2.23	0.34
Oral sucker (l)	0.50-0.93	0.73	0.09
Oral sucker (w)	0.52-0.79	0.65	0.08
Ventral sucker (l)	0.84-1.46	1.22	0.12
Ventral sucker (w)	0.88-1.46	1.20	0.12
Pharynx (l)	0.17-0.33	0.28	0.04
Pharynx (w)	0.15-0.34	0.27	0.04
Seminal vesicle (l)	0.10-0.57	0.27	0.14
Seminal vesicle (w)	0.05-0.40	0.19	0.09
Right testis (l)	0.40-1.17	0.76	0.22
Right testis (w)	0.31-1.10	0.77	0.22
Left testis (l)	0.36-1.12	0.77	0.23
Left testis (w)	0.28-1.22	0.78	0.23
Ovary (l)	0.45-0.90	0.64	0.12
Ovary (w)	0.28-0.84	0.42	0.10
Mehlis' gland (l)	0.21-0.53	0.36	0.09
Mehlis' gland (w)	0.15-0.43	0.27	0.07
Egg (l)	0.046-0.052	0.049	0.001
Egg (w)	0.015-0.021	0.018	0.001
Filament (l)	0.083-0.117	0.097	0.010
Ratio filament: egg[a]	1.69-2.54	1.98	0.22

(l) = length; (w) = width
a: Egg of Fig. 4: exceptional ratio of 3.36.

Material deposited in the Helminthological Collection of Instituto Oswaldo Cruz (CHIOC no. 32815 a-q).

NOTES ON THE WORKING OF THE OVARIAN COMPLEX

The ovary is surrounded by a fibrous capsule which contains thin muscle fibers. Mature egg cells accumulate in a funnel-shaped projection of the ovary (Fig. 11), continuous with the oviduct, that has no suction chamber or ovicapt. They line up in the proximal portion of the oviduct, moving to and fro until the foremost one reaches the oviductal opening of Laurer's canal. That portion of the oviduct is lined with non-ciliated squamous epithelium coated with very thin fibers and has no contractile wall, there seeming that the egg cells are propelled by the *vis a tergo* imparted by weak contractions of the ovarian capsule and the flux of their newly matured fellows. The distal portion of the oviduct, extending from the Laurer's canal opening to the ootype, is ciliated and has a thicker wall. On reaching the oviductal opening of Laurer's canal the egg cell stops for a while, and is soon driven by a peristaltic wave, originated at that point, in the direction of the ootype. After traveling most of the distal portion of the oviduct it stops exactly at the opening of the common vitelline duct. Just then a small group of vitelline cells (usually 4) approach the egg cell and adhere to its side. Sometimes the egg cell enters the duct's outlet to leave it with the vitelline cells attached.



Halipegus dubius – Fig. 21: egg in the making in the ootype. Fig. 22: two newly formed uterine eggs, egg cell at the opercular pole. Fig. 23: Laurer's canal showing thin-walled proximal portion (lc_1) and thick-walled distal portion (lc_2). Fig. 24: proximal portion of Laurer's canal distended with leftover spermatozoa. Fig. 25: distal portion of Laurer's canal with leftover spermatozoa and vitelline granules. Fig. 26: section through convergence of metraterm and ejaculatory duct showing a double lumen (arrow). Bar = 10 μm (Figs 21,25), 25 μm (Figs 22, 23), 50 μm (Figs 24, 26).

Impressive is the precision of the mechanism that converges egg cell and vitelline cells to a definite spot. When the fluke gets wearied from prolonged compression of its body the movements of the egg cell in the proximal portion of the oviduct begin to fail; it moves to and fro but does not reach the opening of Laurer's canal. At the same time the vitelline cells also move to and fro in the vitelloduct without reaching its outlet. So perfect a synchronism suggests the operation of a kind of reflex arc (afferent branch in the oviduct, effector in the vitelloduct).

The egg cell, now with the vitelline cells attached to its posterior half, moves through the short remaining portion of the oviduct and enters the ootype. In contact with the secretion of Mehlis' gland most granules of the vitelline cells rapidly disappear leaving vacuoles in their place, at the same time that around the group of vacuolate vitelline cells there appears a thin, transparent, colorless, refractile, deformable pellicle that is destroyed by Bouin's fixative. A conical extension appears at the posterior end (pointing to the oviduct) of that pellicle, corresponding to the base of the egg filament (Fig. 21, bf). As to the filament proper, I was unable to witness its formation in the ootype, owing perhaps to the transparency of the latter being diminished by interposition of the Mehlis' gland wall. At this stage there is no sign of operculum, the forming eggshell resembling a goblet full of vitelline-cells (Fig. 21, vc) surmounted by the egg cell which surpasses the goblet's brim (Fig. 21, eg).

These observations show that, according to several authors (review by Smyth & Clegg, 1959), the primordial eggshell (the above-mentioned pellicle) originates from the granules of the vitelline cells, for its appearance follows disappearance of the granules and it encloses just the group of vitelline cells. It thus constitutes a matrix on which the eggshell is built.

At the outlet of the ootype the unfinished egg is impelled by peristaltic waves through a narrow duct which gradually widens to empty into the first uterine coil, usually filled with spermatozoa (uterine seminal receptacle, Figs 6, 10, ur). That coil also contains refractile granules which adhere to the above-mentioned egg pellicle, and only then does an operculum become visible. The late appearance of the operculum in the first portion of the uterus was observed by Linstow (1890) in *Haplometra cylindracea*. It seems plausible that the two parts, destined to be later separated, should have separate origin. The sequence described above of egg formation in *H. dubius* is paralleled by that of *Clonorchis sinensis*, as observed by Ujiie (1936b): "there appears first the peripheral wall, next the covering of the posterior end, and finally the operculum". As the formed egg progresses along the first uterine coils (Fig. 22) its shell gradually takes on a brownish color.

The presence of spermatozoa being rejected through Laurer's canal (see below) indicates that they may be found in the distal portion of the oviduct and in the ootype, where they can fertilize the egg cells. So, the late appearance of the operculum, leaving the egg cell long exposed ahead of the forming egg, suggests that such a disposition ensures egg cell fertilization in case it meets no competent spermatozoa during its travel along the oviduct and ootype.

The convoluted Laurer's canal (Figs 6, lc; 23, lc_1, lc_2) is lined with an epithelium that shows rare nuclei. The lining cells are flat and ciliated in the proximal half (Fig. 23, lc_1), higher and without cilia in the distal portion (Fig. 23, lc_2) whose muscular coat is more developed. *In vivo* observation shows that the cilia of the oviduct beat from the ootype toward Laurer's canal. In the ootype and distal oviduct the ciliary movement is perceptible when the lumen is open, what occurs periodically between passing egg cells.

When the egg cell, moving along the proximal oviduct, reaches the Laurer's canal opening, the latter suddenly closes, and at that point a peristaltic wave originates, sweeping down the duct. This wave counteracts the effect of the ciliary current, propelling the egg cell toward the ootype, under whose contractions it keeps on moving forward, now with the attached vitelline cells.

The ciliary current drains to the exterior sperm, egg cells, vitelline cells and free granules, excess fluids and all other leftovers from egg formation. Those materials are brought to the proximity of Laurer's canal. From tine to time the canal opening dilates and those materials are suddenly projected into the canal, what can be seen when, after prolonged obser-

Halipegus dubius – Fig. 27: adult specimen, caudal half. Bar = 1 mm.

vation, the egg cells arrived at the ootype, being unable to advance owing to the failure of the ootype wall to contract, are sent back by the ciliary current together with the vitelline cells, and instead of passing into the proximal oviduct they are swallowed by Laurer's canal.

The proximal half of Laurer's canal contains a liquid with suspended sperm and, in less amount, other materials from among the just mentioned (Figs 23, 1c$_1$; 24, 1c$_1$), permanently whirling under ciliary action. Therefore that segment frequently distends (Fig. 24), and owing to the presence of sperm such transient swelling may be mistaken for a seminal receptacle.

Intermittent peristaltic rushes simultaneously sweep several portions of the canal, pushing its contents toward the outlet (Fig. 25), through which they are ejected from time to time into the surrounding salt solution.

REMARKS

The structure of the terminal genitalia in *Halipegus* has been diversely described by several authors. Creutzburg (1890) and Sonsino (1893a) made mention of a cirrus and a penial pouch, respectively, in the type species, *H. ovocaudatus.* Looss (1894), however, asseted that there is no sign of such organs in that species. He described a pear-shaped seminal vesicle, free in the parenchyma, which continues into a short muscular ejaculatory duct surrounded proximally by scanty prostate glands and emptying into the genital atrium; and a short, little strong vagina well proportionate to the short ejaculatory duct. Hamann's (1986) description of the terminal genitalia of the holotype of *H. ovocaudatus* does not differ essentially from Looss'.

As to *H. dubius,* Cordero (1942) gives no details of the terminal genitalia, saying that "the seminal vesicle is well developed and the genital pore opens in front of the pharynx", and that the uterine coils follow "a tortuous course to discharge with the deferens through the genital pore". According to Kohn & Fernandes (1988), this species has neither sinus-sac nor sinus-organ, its seminal vesicle continues into a pars prostatica, and the uterus opens through a muscular metraterm into the genital atrium; they make no reference to either ejaculatory or hermaphroditic duct. As seen above, under "Description", the terminal genitalia of the specimens herein studied agree with those of *H. ovocaudatus* as described by Looss (1894) and Hamann (1986), except for the presence of a hermaphroditic duct in *H. dubius.* In Fig. 9 an arrow indicates the transition from the metraterm to the hermaphoditic duct, and in Fig. 26 the convergence of the metraterm and the ejaculatory duct appears as a double lumen in cross-section.

The role of the vitelline cells in eggshell formation has been investigated by a number of workers since Leuckart (1886) and Henneguy (1902, 1906) drew attention to the subject (see review by Smyth & Clegg, 1959). The above account brings no addition to current knowledge except for minor details. The number of vitelline cells associated with each egg cell, which is typical for the various species studied – 3-4 in *Haplometra cylindracea* (Linstow, 1890), about 30 in *Fasciola hepatica* (Henneguy, 1902), 2 in *Zoogonus mirus* (Goldschmidt, 1905), 3-7 (usually 4) in *Collyriclum faba* (Tyzzer, 1918), 5-7 in *Clonorchis sinensis* (Ujiie, 1936b), 18 or more in *Hexacotyle extensicauda* (Dawes, 1940), 30-40 in *Schistosoma mansoni* (Gönnert, 1955), etc. – is usually 4 in *H. dubius.*

The association between the egg cell and the vitelline cells has been described somewhat differently by the authors. There follow some examples:

– in trematodes generally: the egg cell gets in touch in the ootype with the vitelline cells which, subjected to lively contractions, gather together around the egg cell (Leuckart, 1886);

– in *Diplodiscus subclavatus:* the egg cell arrives at the uterus and a number of vitelline cells enter after it, arranging themselves into a kind of ellipsoid with the egg cell at a pole (van Beneden, 1870);

– in *Fasciola hepatica:* the vitelline cells are poured into the region of Mehlis' gland when the egg cell arrives there, no mention being made of their kind of association (Sommer, 1880; Schubmann, 1905); Henneguy (1906) admitted the likelihood of this process, and stated that the egg cell and the vitelline cells did not group but could be seen very irregularly distributed in the uterine ootype, their groupment occurring farther on in the uterus. "A number of vitelline cells and an ovum group together after passing through the uterine valve" (Smyth & Clegg, 1959);

– in *Haplometra cylindracea:* after fertilization of the egg cell in the ootype by refluent sperm from the proximal region of the uterus (uterine ootype) 3 or 4 vitelline cells enter the ootype (Linstow, 1890);

– in *Echinochasmus japonicus:* "the ovum and the yolk masses meet in the initial portion of the oviduct, and the ovum becomes partly immersed into the anterior end of the vitellogen mass" (Ujiie, 1936a);

– in *Clonorchis sinensis:* the vitelline cells "enter like droplets into the axial cavity of the ootype"; "when a matured egg-cell has arrived in the initial portion of the ootype from the oviduct, it is followed by 5-7 yolk cells from the yolk reservoir" (Ujiie, 1936b). From this description it seems that the egg cell and the vitelline cells are not joined together at the ootype;

– in *Parorchis acanthus:* the vitelline cells "follow the oocytes closely, but do not become applied to them immediately. The application of yolk cells takes place at the first loop of the uterus" (Rees, 1939);

– in *Dolichosaccus trypherus:* the egg cell enters the ootype and moves toward the entrance of the yolk duct (which opens into the ootype); "mostly the ovum moves up to that point and away again several times, but, finally, a number of yolk-cells (generally four) enter the ootype, and at once become fastened to one side of the ovum, so that this cell now forms the apex of a little group of five cells" (Johnston, 1912).

The last example is the one that most closely agrees with my observations in *H. dubius.* In both cases live specimens were extendedly watched.

It seems reasonable to assume that a reflex arc mechanism converges an egg cell and a species-specific number of vitelline cells just to the doorstep of the ootype, the site of eggshell elaboration; and that surface tension forces (van der Waals attractive forces) ensure a firm adherence of the vitelline cells to each other and to the egg cell, so that they can withstand, without separating, the contractions of the ootype wall until reaching as a whole the uterine seminal receptacle where eggshell formation is completed.

As mentioned above, Lühe (1900) treated the Brazilian trematode collected by Olfers and Sello as *"Halipegus* sp.?" and found it identical with *H. ovocaudatus;* Klein (1905) considered it a good species, naming it *Halipegus dubius;* and Cordero (1942) confirmed its validity, giving as diagnostic characters the proximity of the testes to the ventral sucker, their being larger than the ovary, and chiefly the filament: egg length ratio (3.0-4.0). My specimens are much larger (5.0-8.4 mm) than Cordero's (3.3-4.0 mm). Those of Kohn & Fernandes (1988) also are smaller (2.06-4.14 mm). Such differences result from intraspecific variation. *H. ovocaudatus,* for instance, varies in length from 4-5 mm (Sonsino, 1893a), 5-7 mm (Creutzburg, 1890) and 6-7 mm (Vulpian, 1859) to 12-13 mm (Loss, 1894). Examples of intraspecific variation in size, body shape and structure of the reproductive system in trematodes are adduced by Dogiel (1964: 445).

The same holds true for differences in filament: egg length ratio. Exemplifying again with *H. ovocaudatus,* this ratio varies from 1.0-1.5 (Creutzburg, 1890; Looss, 1894; Lühe, 1900) and 1.45 (Vulpian, 1859) to 4.0-6.0 (Sonsino, 1893b). The available values for *H. dubius* are

1.93 (Fig. 4 of Kohn & Fernandes, 1988), 2.27 (Lühe, 1900), mostly 1.7-2.5 but up to about 3.3 (Paraense, this paper) and 3.0-4.0 (Cordero, 1942).

Notwithstanding the similarity of the four American species of *Halipegus* recognized as valid, they differ significantly in some characters of their cercariae. As shown in Figs. 2, 3 and 20, the cercaria of *H. dubius* has a caudal appendage (handle) shorter than the cyst (tail or bulb). In *H. occidualis* Stafford, 1905 the caudal appendage is proportionally slenderer and about as long as the cyst (Krull, 1935; Goater et al., 1990). In *H. eccentricus* Thomas, 1939 the caudal appendage is shorter than the cyst (as in *H. dubius*) and there are two ribbon-like streamers, one on either side of the caudal appendage. And in *H. amherstensis* Rankin, 1944 there is no caudal appendage and the delivery tube (which is normally folded up within the cyst in the other species) is permanently everted.

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ABBREVIATIONS IN FIGURES

bf = base of egg filament, ca = caudal appendage, cc = cercaria, ce = cecum, cy = cyst (cercarial tail), dt = delivery tube, ec = excretory canal, ed = ejaculatory duct, eg = egg cell, ep = excretory pore, es = esophagus, ev = excretory vesicle, gp = genital pore, hd = hermaphroditic duct, lc_1 = Laurer's canal, proximal portion, lc_2 = Laurer's canal, distal portion, me = metraterm, mg = Mehlis' gland, od = oviduct, os = oral sucker, ov = ovary, ph = pharynx, pp = pars prostatica, ps = prececal sac, sv = seminal vesicle, ur = uterine seminal receptacle, ut = uterus, vc = vitelline cells, vd = vitelline duct, vi = vitellarium, vs = ventral sucker.