CERCARIA SINITZINI[1] N. SP., A CYSTOPHOROUS CERCARIA FROM *PERINGIA ULVAE* (PENNANT 1777).

BY MIRIAM ROTHSCHILD.

(With 31 text-figures.)

IN 1936 I reported the occurrence of a cercaria closely allied to *Cercaria sagittarius* Sinitzin 1911 in *Peringia ulvae.* Since the publication of that note I have found another species pertaining to this group, which, however, is so rare that there seems little if any prospect of solving the life-history. In consequence it is deemed unnecessary to withhold the morphological description of this interesting species until the publication of the writer's fuller paper.

The cystophorous cercariae—the larvae of the **Hemiuridae** Lühe (Sinitzin 1905, Dollfus 1923, Krull 1935)—are among the most extraordinary and beautiful of all known cercariae, and have never failed to excite the enthusiasm of those helminthologists fortunate enough to discover them. Thirteen species have been described, several of which have proved extremely rare, occurring only once or twice in several thousand examples of the host.

Cercaria sinitzini greatly resembles *C. sagittarius* in general appearance and is undoubtedly closely related to it. It can be distinguished instantly from this species by the possession of an additional caudal appendage (see Table I, p. 42).

TECHNIQUE.

These cercariae are exceptional in that they are best studied when fixed and stained, though certain characters, notably the excretory system, are only visible during life. Two rediae and their contents were set aside and studied alive. The remaining rediae were carefully opened and fixed on the slide with Bouin Dubosq. Two slides were stained with Delafield's haematoxylin and another with paracarmine. The extreme anterior portion of the snail's liver was sectioned. The sections were cut at 8 μ, and stained with Delafield's haematoxylin with a counterstain of eosin.

Measurements.—Measurements, unless otherwise stated, were made from preserved material only. The size of the various appendages is very variable. Table II is provided to afford a comparison between them and the developing cercaria.

TABLE I.

C. SAGITTARIUS	C. SINITZINI
1. Rediae not active.	1. Rediae very active.
2. No 5th caudal appendage.	2. 5th caudal appendage present.
3. Terminal portion of Appendage IV (= "Phrygian Cap") not drawn out into long tapering point.	3. Terminal portion of Appendage IV (= "Phrygian Cap") drawn out into long tapering point, longer than the main body of the cap.
4. Linear arrangement of lateral cells of Appendage II (= "Ribbon").	4. Spiral arrangement of central cells of Appendage II (= "Ribbon") and irregular arrangement of lateral cells.
5. Encysted cercaria folded twice inside cyst, with extremities in upper half.	5. Encysted cercaria folded once inside cyst, with extremities in basal half.

[1] I have named this species in honour of D. T. Sinitzin. The type is deposited at the British Museum (Natural History), Cromwell Road, London.

TABLE II.[1]

Measurements of Appendages
in microns.

Body of Cercaria	Tail-vesicle (lateral aspect)	Appendage I	Appendage II	Appendage III	Appendage IV	Appendage V
37 × 31	26 × 23	21 × 6	12 × 10	not developed	not developed	not developed
38 × 31	28 × 26	38 × 5	24 × 10	not developed	not developed	not developed
44 × 31	33 × 31	45 × 5	28 × 9	9 × 10 (at base)	not developed	not developed
44 × 33	31 × 30	38 × 4	28 × 9	not developed	not developed	not developed
45 × 30	43 × 33	79 × 3–5	37 × 9	broken off	26 × 16	not developed
49 × 30	38 × 35	70 × 5	37 × 9	21 long	21 × 16	9 long
51 × 24	38 × 31	107 × 3	40 × 9	38 long	23 × 14	17 long
52 × 31	40 × 35	65 × 7	35 × 9	35 long	23 × 17	14 long
54 × 28	35 × 35	Broken	37 × 9–10	44 long	26 × 12	31 long
56 × 26	44 × 37	87 × 7	44 × 9	40 long	24 × 16	19 long
56 × 38	40 × 35	61 long	43 × 9	17 long	broken	broken
57 × 40	40 × 35	52 long	26 × 9	8 long	broken	broken
59 × 28	40 × 37	115 × 8	58 × 8–9	37 long	23 × 12	17 long
60 × 30	not measured	129 × 2–5 (extended)	45 × 8	40 long	32 × 12	16 long
60 × 30	not measured	113 × 8–9 (not extended)	45 × 8	40 long	32 × 12	16 long
63 × 25	52 × 48	104 × 5	44 × 9	43 long	broken	23 long
72 × 23	49 × 35	122 × 3	45 × 9	49 long	broken	28 long
73 × 23	(ventral aspect) 42 × 31	56 × 7 (telescoped)	49 × 9	31 long	38 × 16	17 long
73 × 23	42 × 31	61 × 10 (telescoped)	48 × 9	30 long	49 × 16	28 long
79 × 18	43 × 35	52 (telescoped)	26 × 9	not measured	not measured	not measured
79 × 23	45 × 40	105 × 7	45 × 9	40 long	38 × 17	17 long
80 × 25	not measured	52 × 10 (telescoped)	50 × 10	30 long	38 × 12	34 × 1·7
82 × 23	50 × 40	51 (withdrawing into tail)	50 × 9	35 long	not measured	17 long
85 × 25	not measured	53 × 11 (telescoped)	48 × 9	35 long	36 × 16	broken
87 × 21	46 × 38	33 (withdrawing into tail)	49 long	35 long	23 long	17 long
87 × 24	52 × 44	58 × 10 (telescoped)	61 × 9	30 long	35 × 13	22 long
Cyst	—	—	Appendage II	Appendage III	Appendage IV	Appendage V
43 × 43 (lateral aspect)	—	—	35 × 6	46 long	not measurable	not measurable
39 × 39 (lateral aspect)	—	—	39 × 6	45 long	not measurable	not measurable
42 × 42 (lateral aspect)	—	—	40 × 6	40 long	not measurable	not measurable

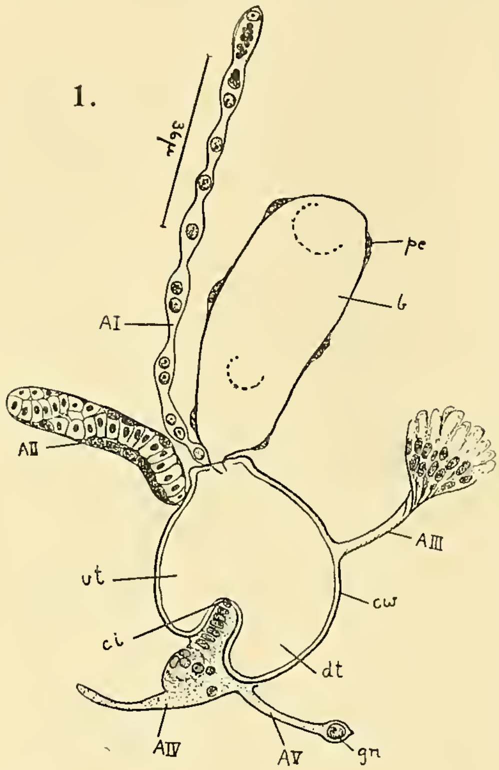
[1] See Fig. 9 for lines of measurements.

Cercaria sinitzini n. sp.

The Redia.—Measurements. Body : 1,000 μ × 200 μ or less.
Intestine : 125 μ ,, ,,
Pharynx : 33 μ × 26 μ ,, ,,

The redia of this species is very distinctive, and can be recognized with the aid of a hand lens. The body is cylindrical and a pearly white in colour. The pharynx is well developed and leads into a short, bright yellow intestine about $\frac{1}{8}$ the length of the body, which is very conspicuous in the living animal. Collar and ambulacral processes are absent. A birth-pore is situated a little below the pharynx. The pharynx is surrounded by digestive glands.

1.



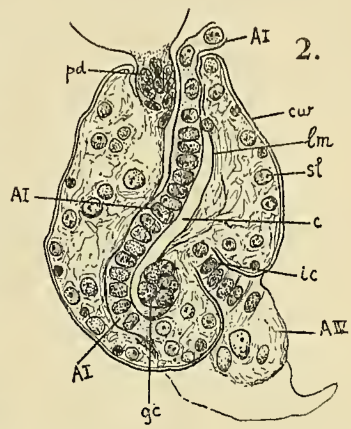
Owing to the scarcity of the material the excretory system was not worked out in detail. It is well developed, and there are over fourteen flame-cells on each side of the body, which appear to be arranged in groups of four.

A very characteristic feature of the redia are its movements, and it is this in conjunction with the pearly white colour which gives these parthenitae such a distinctive appearance. The body wall of the redia is exceptionally muscular. When liberated from the tissues of the snail, the body is violently contracted, first in one place, then in another. Unlike the rediae of *C. sagittarius* these rediae are very active. The body cavity is packed to bursting-point with cercariae in all stages of development, including the encysted forms. Over 100 were counted in one redia. It seems probable that the cysts are expelled through the birth-pore by the contractions of the redia, as they themselves seem incapable of movement. The rediae infest the gonad of the snail. Sections show the liver to be comparatively undamaged. One infection was found in two thousand specimens of *Peringia ulvae* taken from St. John's Lake, Plymouth, in April 1936.

The Cercaria.—Measurements of full-grown specimens :

Body (length)	63 μ–105 μ	(average 85 μ).
Body (width)	12 μ–25 μ	(average 18 μ).
Tail (length without appendages) .	43 μ–52 μ	(average 48 μ).
Tail (width from dorsal surface to ventral surface)	35 μ–42 μ	(average 38 μ).
Oral Sucker	14 μ × 14 μ–15 μ × 14 μ.	
Ventral Sucker	7 μ × 7 μ–9 μ × 9 μ.	
Pharynx	6 μ × 6 μ.	
Peduncle	8 μ–9 μ.	

The Body (fig. 4).—As in most Hemiurid cercariae the body is poorly differentiated and stains in the manner of germ balls and very young individuals of other species of cercariae. The whole animal is very transparent. The oral sucker is markedly subterminal and leads into a pharynx, oesophagus and forked intestine. The caeca reach beyond the ventral sucker posteriorly. The ventral sucker, situated at about the posterior third of the body, is poorly developed and smaller than the oral sucker. The Y-shaped excretory bladder is terminal. The excretory vesicle (fig. 4) is formed by two lateral arms joining the bladder at the anterior cornu, which pass parallel to each other on either side of the ventral sucker and fuse dorsally behind the pharynx. The formation of the excretory vesicle or " closed circuit " occurs after the lateral excretory tubes have turned posteriad (in the region of the ventral sucker) and divided into an anterior-lateral and posterior-lateral branch. Thus the general development of the excretory system resembles that of *C. ephemera* Lebour 1907 (nec Nitzsch) (Rothschild 1936) except in the very important point that the main lateral excretory tubules only grow as far forward as the acetabulum[1] before turning posteriad. Owing to the undifferentiated condition of the cercaria the finer branches of the excretory system are very difficult to follow. If my observations are correct this species (and probably all the **Hemiuridae**) is an exception to Dubois' rule that cercariae with the Mesostoma type of excretory system develop in sporocysts and not in rediae (Dubois 1929).



2.

The cuticle is unarmed and no cystogenous glands were observed.

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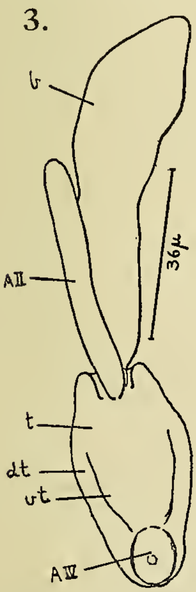

A very striking feature of the body of the cercaria is the investing primitive epithelium (figs. 1, 9, 9*a*). The nuclei are arranged in pairs and are very conspicuous in stained specimens—bulging out at intervals along the edge of the body. This primitive epithelium is retained by the encysted cercaria, and according to Pratt (1898) the metacercariae of Hemiurids found in the body cavity of Copepods also retain it for a considerable period. This fact strongly suggests that any cystogenous material which may be required for closing the orifice of the tail comes from the inside of the tail-vesicle itself, and is not shed by the cercaria as suggested by Sinitzin. A similar condition was recorded for *C. macrura* Faust 1921.

The body of the cercaria is fixed in the anterior aperture of the tail by a thin peduncle, consisting of four rows of single cells. Early in development these arise from the base of the inside of the tail-vesicle, close to the origin of Appendage 1 (fig. 7). The cells forming the original portion of the peduncle degenerate early, so that only a few remain near the aperture of the tail, forming the very short peduncle of the fully-developed cercaria (fig. 2). The cercaria encysts by withdrawing into the tail.

The Tail.—When viewed from the side the main body of the cystic tail is shaped like a jug with a wide mouth (figs. 1, 2, 4, 5). When viewed from directly in front (fig. 3) or behind, it is egg-shaped. Almost all mounted specimens are

[1] In the *Azygiidae* Odhner, the excretory system is of the Stenostoma type, and it seems likely that this pseudo-Mesostoma excretory system of *Cercaria sinitzini* is a later development from the former type—arising by a shortening of the lateral tubules.

seen from the side, as the tail is compressed laterally and tends to assume this position on the slide. The ventral half of the tail is narrower than the dorsal half. The tail carries five appendages (fig. 1, 4).

Appendage I (the "Arrow" of Sinitzin (1911), "Excretory Projection" of Cort and Nichols (1922), Willey (1930), and the "Delivery-Tube" of Krull (1933), arising from the interior floor of the tail and passing out at the aperture, ventral to the body of the cercaria.

Appendage II (the "Ribbon" of Sinitzin), attached externally[1] to the mid-ventral line of the tail, a short distance below the mouth.

Appendage III (the "Sultan's Plume" of Sinitzin), attached externally to the centre of the mid-dorsal line of the tail.

Appendage IV (the "Phrygian Cap" of Sinitzin), forming a prolongation of the terminal portion of the tail, directed ventrad.

Appendage V, a thin outgrowth of the base of Appendage IV, directed distad and posteriad.

The outermost layer of the tail consists of an exceedingly thin transparent membrane which is most probably the primitive epithelium of which the nuclei have degenerated. It invests the vesicle proper and all the appendages, at least in their early stages. Below this is a much thicker, hyaline, cuticular layer staining a faint mauve with Delafield's haematoxylin. This homogeneous layer forms the cyst-wall. Posteriorly it is sharply invaginated (figs. 1, 4) so that in lateral aspect the base of the vesicular portion of the tail is divided into two asymmetrical halves. The dorsal half is larger than the ventral half both in breadth and depth, and shows behind the latter if the tail is viewed from directly in front (i.e. ventrally). This invagination of the cyst-wall seems more pronounced than in *C. sagittarius*. Within the cyst-wall is a layer of single cells with large oval nuclei and granular protoplasm (figs. 2, 5). Quite early in development, before the appearance of the cyst-wall itself, these can be traced clearly outlining its future course, including the posterior indentation, etc. All these three layers thin out, and are obliterated inside the aperture of the tail.

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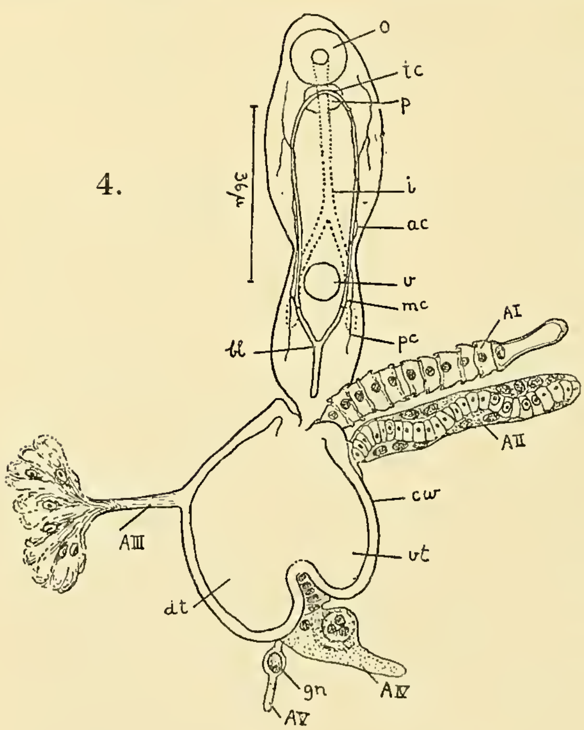


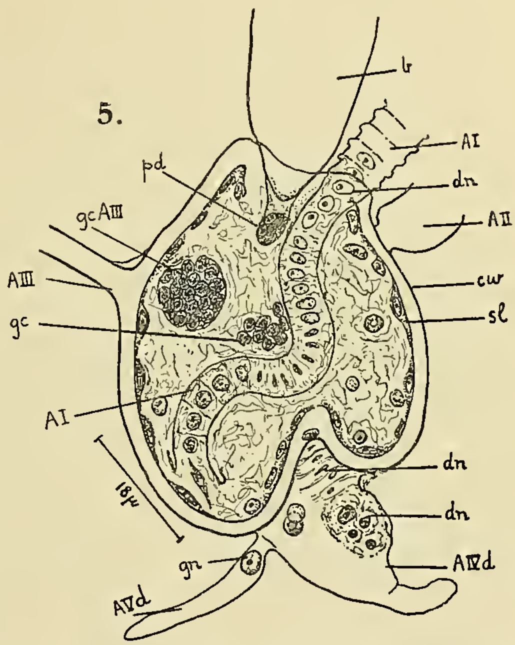
The internal structure of the tail-vesicle is extremely complicated, and I

[1] Prior to encystment Appendages II and III are connected with the outer layers of the tail only, but during some stage in development these appendages also are connected with the inner cell-structure of the tail-vesicle.

have been unable to gain a clear understanding of the whole organ and the function of the various cells. There are several compact groups of cells, each invested in a thin membrane. These give rise to Appendages II, III, the peduncle, etc. Two other groups are placed near the base of Appendage I (fig. 2) and these appear to be intimately connected with the membranous sheath which develops round the base of this appendage after it is withdrawn into the tail-vesicle. In addition there are numerous ungrouped cells, lying in different planes and possessing various-sized nuclei, some of which may provide cystogenous material. The connections between the various appendages and the groups of cells within the tail-vesicle are lost at different times. A gradual degeneration of all these cells takes place, but certain groups disintegrate before others.

By the time the cercaria is ready to withdraw into the cyst the inner structure of the tail has changed considerably. The cyst-wall has thickened, particularly at the aperture of the tail, where it is at least double the thickness of other regions. The nuclei of the granular protoplasmic layer have flattened out. The rest of the cellular contents of the vesicle have shrunk and disintegrated.

5.



While Appendage I (the delivery-tube) is still growing (i.e. before it has attained its maximum length) the tail-vesicle is more or less "solid." The portion of the delivery-tube within the vesicle is, however, surrounded by a narrow space. This cavity is bounded by a stout membrane which can be seen in sections (fig. 2). Although at this stage it is not easy to make out the precise relationships of these parts, the membrane lining the central cavity appears to be continuous with the delivery-tube itself. By the time this appendage has reached the "telescoping" and "withdrawing" stage (see below) the cavity in the tail-vesicle has increased considerably in size. This appears to take place as a result of the disintegration of the internal cell-structure of the tail mentioned above. The innermost membrane expands with the cavity until it forms a second lining to the cyst-wall. Thus when the cercaria withdraws, the tail-vesicle is hollow, and it enters the cavity bounded by this inner membrane.

At this stage the tail-vesicle barely takes up any stain. The cyst-wall and the lining membrane, together with the membranes of two internal groups of cells, are just visible, and here and there the remains of disintegrating nuclei show up.

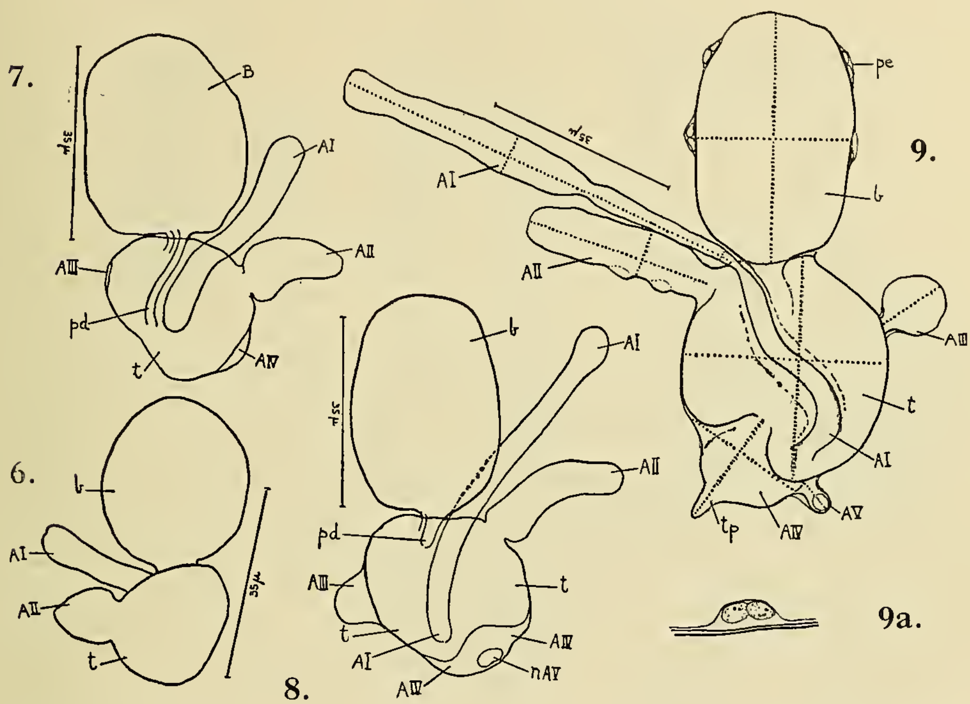
Appendage I (figs. 15–18*a*).—This appendage originates from a group of cells on the floor of the inside of the dorsal half of the tail-vesicle, and, as already mentioned, is homologous with the delivery-tube of Krull. Its function also appears to be identical. It is differentiated at about the same time as the bud of Appendage II, but grows much faster. It arises as a somewhat thick double row of cells, passing upwards and outwards by the future aperture of the tail-vesicle, ventrally to the body of the cercaria. It consists of an inner core of closely-packed cells, with their long axis at right angles to the long axis of the appendage (fig. 15). The cell-walls are never very distinct. When the body of the cercaria measures about 40 μ in length, about thirteen of these cells, counted by their nuclei, can be seen outside the body of the tail. This row of cells terminates in a club-like swelling containing a group of numerous small deeply-staining nuclei. At the extreme terminal portion of the swelling is one giant oval-shaped nucleus. The appendage is invested with a delicate transparent membrane which cannot be detected in later stages except in sections. Rarely, during development, one of the nuclei pertaining to this membrane can be seen bulging out from the surface of the appendage (fig. 15). When the appendage pushes out from the tail-vesicle, it is surrounded by the most external cuticular or epithelial layer of the tail. I have been unable to determine whether this layer is ruptured or whether it grows with the appendage, adhering closely to its sides. The appendage grows by the elongation of these cells outside the body of the tail vesicle. These come to form a long ribbon, of single cells with their long axes now parallel to the long axis of the appendage. The external cells still rarely exceed 13 in number. The nuclei, particularly near the aperture of the tail, tend to be grouped in pairs (fig. 1). At this stage the structure of the inside of the tail-vesicle becomes less dense and the internal course of the appendage can be seen more clearly (fig. 5). Here the long axis of the cells remains transverse. Immediately above the posterior invagination of the tail-vesicle, the appendage bends sharply forward and continues its course upwards in the ventral half of the tail. At a stage when the body of the cercaria measures about 60 μ–70 μ in length, the appendage is capable of considerable elongation and contraction. It then measures 87 μ–129 μ in length and 2 μ–9 μ in width (figs. 16, 17).

When the cercaria reaches a length of approximately 73 μ a marked change occurs in the appendage. A general telescoping of the portion outside the tail-vesicle takes place, so that either the top of one cell fits into the bottom of the next, or the base of one fits into the top of the adjacent cell (figs. 16, 18*a*). This brings the nuclei very much closer together and the appendage now measures only 50 μ–61 μ in length and 10 μ–11 μ in width. The small, deeply-staining nuclei in the terminal bulb disintegrate. The giant terminal nucleus also disappears. The cuticle of the whole appendage thickens somewhat, but particularly that of the terminal bulb. The constriction between it and the rest of the appendage is accentuated, forming a teat-shaped extremity. The entire appendage now stains a pinkish mauve instead of a bluish purple. Vacuoles and fibres become more noticeable in the protoplasm of the cells (fig. 18).

The onset of encystment is marked by the withdrawal of Appendage I into the cavity of the tail. This does not appear to be the result of muscular contraction but due to the mechanical effect of the collapse of the basal, internal portion of the appendage, weakened by the enlarging of the cavity inside the tail-vesicle. The telescoped portion of the appendage is thus pulled down into the inside of the vesicle, within the lining membrane. In *C. sagittarius* it appears that the withdrawal of the appendage and the body of the cercaria coincide, but in the case of *C. sinitzini* complete withdrawal of the appendage takes place first.

It comes to lie in the base of the tail, irregularly coiled up, immediately above and to the sides of the cuticular evagination. Once inside the tail-vesicle, a fairly rapid degeneration of the nuclei and protoplasm of the cells takes place, so that the outline of the whole ribbon can be made out only with difficulty. The cell-walls remain intact and a hollow tube is formed. When Appendage I is thus retracted within the lining membrane of the central cavity of the tail, the whole can be roughly compared with a glove with a single long, inverted finger.

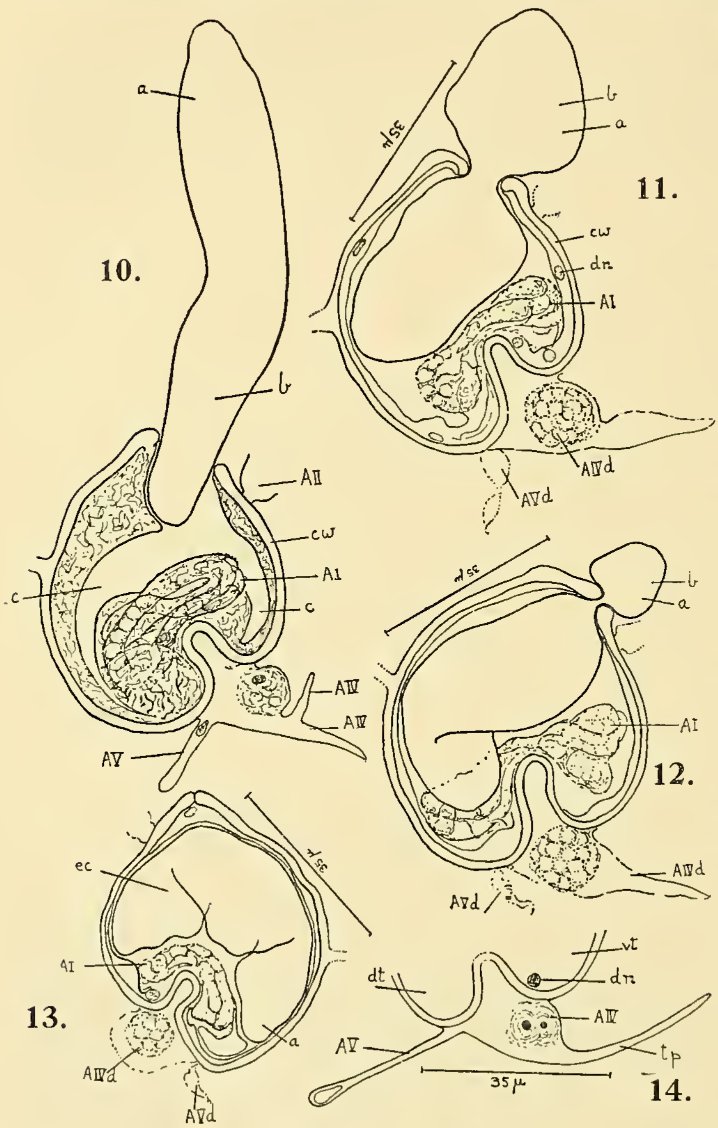
When Appendage I is fully everted during excystment (see below) it will be seen that the basal third is considerably wider than the rest of the appendage, and is invested by a voluminous ribbed sheath (figs. 27–29). This sheath is continuous with the lining membrane and is folded inside the tail-vesicle and cannot be



clearly distinguished prior to excystment. In shape and also in function it is very reminiscent of the " Handle " appendage of both *C. projecta* and the cercaria of *Halipegus occidualis*. It seems possible that is is homologous with that appendage.

Appendage II (figs. 19–21).—The bud of this appendage is differentiated soon after the constriction is formed between body and tail, and is seen as a comparatively large outgrowth on the mid-ventral surface of the tail (fig. 6). With the enlargement of the vesicle it comes to occupy a place nearer the aperture of the tail. The appendage when at the stage of maximum development (fig. 19) is finger-shaped and attached to the tail by a short hyaline stalk, continuous with the hyaline cyst-wall. It then measures 45 μ–60 μ × 8 μ–10 μ. There is a large central cavity, occupied by a column of about 32 closely-packed, single cells, of which the walls and nuclei are very distinct. They are arranged in a loose spiral which is sometimes lost distally, where the column is looped back on itself for lack of space within the central cavity. The long axes of the cells are at right angles to the long axis of the appendage.

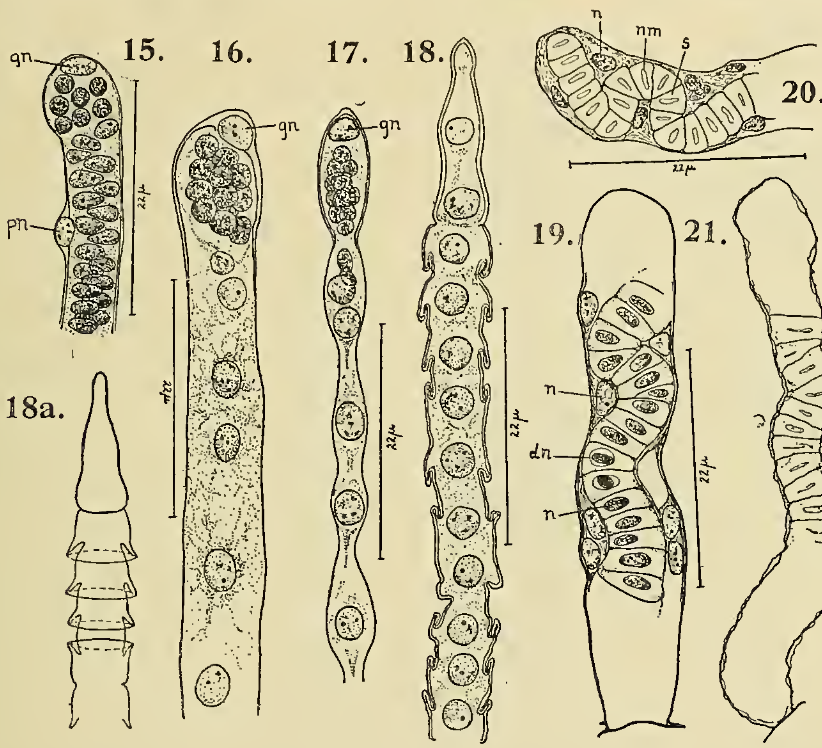
Although the column lies in the centre of the cylinder, it is not closely invested by the membranous outer wall of the appendage. This, unlike the external membrane of Appendage I, appears to be a somewhat rigid structure. Its very conspicuous nuclei, about 13 in number, are scattered irregularly over the appendage, either bulging out from the surface or bulging inwards into the concavities of the spiral core (fig. 19). In *C. sagittarius* it appears that these nuclei are arranged regularly in 2 rows, parallel to the cells of the central column, which here have no spiral twist, but a simple linear arrangement. It is possible that this character is not specific and varies from one infection to another. The whole appendage is invested by the outermost epithelial layer of the tail-vesicle. During the earlier stages of development the core of the appendage is connected with the inside of the tail-vesicle, but this connection is severed when the terminal portion becomes constricted and the short hyaline stalk is formed.



After the telescoping of Appendage I, degeneration of Appendage II commences. The nuclei of the cells of the central core degenerate first (fig. 20). The chromatin of the nucleus shrinks away from the nuclear membrane, forming a dark undifferentiated mass in the centre of the nucleus. It gradually disappears altogether, but even after encystment of the cercaria the faint outline of the collapsed nuclear membrane of these cells can be distinguished in stained specimens (fig. 21). The nuclei of the wall of the appendage subsequently disintegrate completely, and the wall collapses, remaining as a loose crumpled covering to the central column of cells (fig. 21).

The degeneration of the nuclei of all the appendages occurs in a similar manner, but owing to the more definite structure of the cell-walls and nuclei of Appendage II, it is most clearly seen in the case of the latter.

Appendage III (figs. 22–26).—This appendage is differentiated considerably later than I and II, when the body of the cercaria already measures about 40 μ in length (figs. 7, 8). About 16 cells, arising from one of the internal cell-groups of the vesicle, situated in the centre of the mid-dorsal line of the tail, push outwards in a flower-like formation. They are invested by the outer membrane of the tail-vesicle. These cells gradually elongate, each nucleus remaining in the distal portion of the cell. The elongated posterior portions become closely wound together, eventually fusing and forming a homogeneous hyaline stalk continuous



with the thickened cyst-wall. Thus the connection between the appendage and the inside of the tail-vesicle is lost, and the appendage now resembles a flower on a stalk, the petal-like extremities of the cells having apparently broken free from the investing membrane. It measures between 30 μ and 50 μ in length. During and after the encystment of the cercaria the stalk elongates considerably and the nuclei of the cells gradually degenerate. The terminal free ends become ragged, and finally split into a great number of minute threads (figs. 26, 27). In this condition the appendage remains attached to the cyst.

Appendage IV.—This forms a cap at the base of the cystic portion of the tail, 12 μ–17 μ broad at the base × 21 μ–49 μ in length. Its terminal portion is prolonged into a long tapering point, directed towards the ventral side. This tapering point is as long as, or longer than, the upper portion of the cap (figs. 1, 14). There is an internal column of 6–8 cells, in communication with the cells inside the base of the cystic tail. The ventral portion of the cap is enlarged into a hump just before the tapering extremity. A mass of nuclei can be seen inside this portion of the appendage, whereas in the dorsal half no nuclei are visible. The outermost layer of the appendage is continuous with that of the cystic portion of the tail. It is differentiated soon after Appendages I and II. Simultaneously with the appearance of the bud of Appendage III, it can be seen as a hoop of 4–8 cells standing out from the main body of the tail, below Appendage II (fig. 7). Simultaneously with the telescoping of Appendage I, the cells in the cap begin to degenerate. The thickening of the cyst-wall of the tail severs the connection with the central core of cells, which gradually disappear. The cells in the ventral hump break down and form a spherical vacuolated mass with a single disintegrated, deeply-staining nucleus in the centre. The tapering point shrivels up. After the encystment of the cercaria the appendage remains attached to the outside of the cyst, but consists of little more than a structureless mass of tissue.

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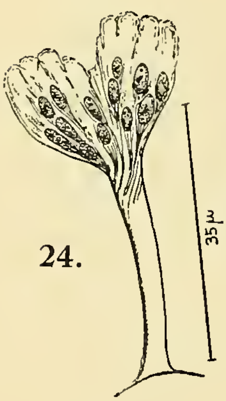

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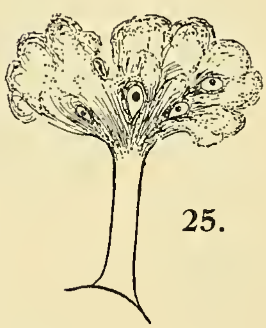


It will thus be seen that Appendage IV of *C. sinitzini* differs considerably from that of *C. sagittarius* (Table I, p. 42). The terminal portion is greatly elongated, being at least twice as long as in *C. sagittarius*. There is also no peripheral layer of cells in the dorsal half of the appendage, but a ventral " hump " which appears to be absent in *C. sagittarius*.

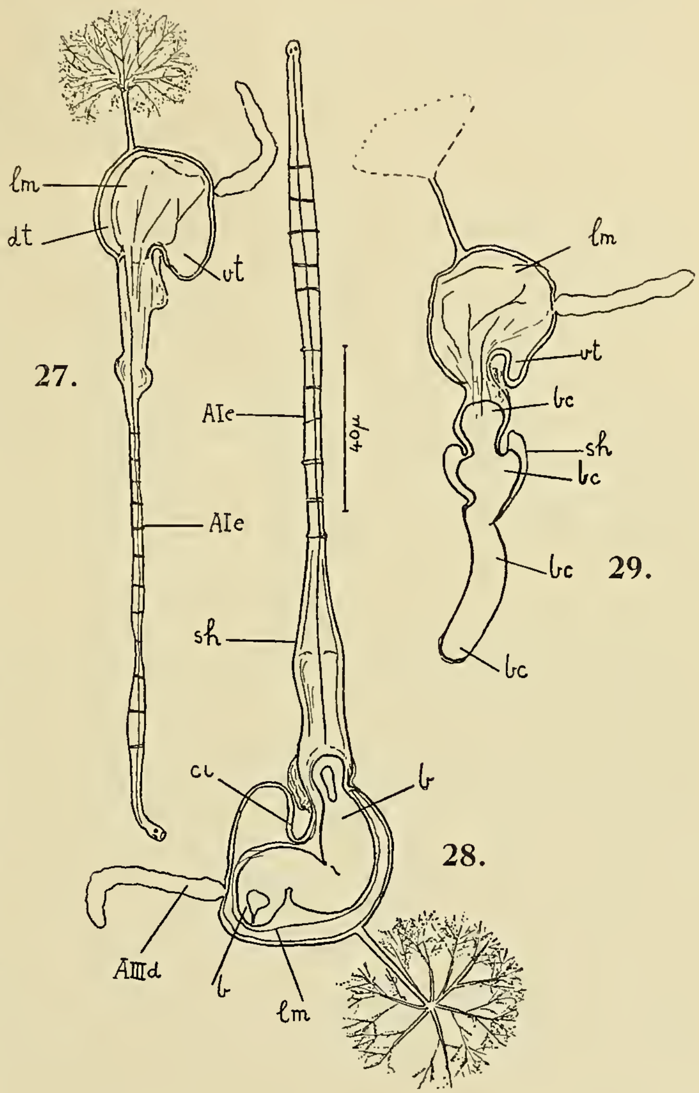
In several specimens of *C. sinitzini*, the terminal portion of the cap is bifurcated (fig. 10) and in other cases it bears a small additional lobe at right angles to the main prolongation.

Appendage V.—This is the last of the appendages to be developed. It is a dorsal outgrowth of the base of Appendage IV (close to the cyst-wall of the tail-vesicle). It is first seen as a giant nucleus pushing out at right angles to Appendage IV (figs. 8, 9). When fully developed it consists of a thin homogeneous hyaline stalk 16 μ–34 μ in length, with a bulbous tip which contains the giant nucleus measuring 4·5 μ × 2·3 μ (fig. 1). It is enveloped in the outermost membrane of the tail-vesicle. After encystment this appendage also undergoes considerable degeneration. The giant nucleus migrates back along the stalk (fig. 5) and finally disintegrates at the base near Appendage IV. The stalk, ragged and distorted in shape, remains attached to the cyst.

In some specimens an interesting abnormality was observed. A second giant nucleus grows out at the base of Appendage V, forming a similar but much smaller appendage at right angles to Appendage V, and parallel to Appendage IV.

Encystment of the Cercaria (figs. 10–13). The cercaria encysts by withdrawing into its tail. As soon as encystment commences, the aperture of the tail-vesicle draws together and it appears as if the body is pulled into the cavity by suction.

The posterior portion of the cercaria descends into the dorsal half of the cyst, rupturing the peduncle (fig. 10). The anterior portion of the body, while still passing through the closing aperture of the tail, undergoes a half twist (fig. 12), and appears to slip through into the central portion of the cyst. However, some vital moment in the process of encystment must have been missed, for when the process of encystment is completed it is the anterior end of the cercaria which almost invariably lies in the dorsal half, and the posterior end in the ventral half of the cyst. During the last stages of withdrawal some fluid material appears to be squeezed out of the tail. This may help to close the cyst, and probably also serves as a lubricant, which assists the cercaria in slipping through the aperture of the tail. The various stages of encystment were followed in preserved material only.



The Cyst (figs. 10–13, 27–29), when fully formed, measures 39 μ × 39 μ–43 μ × 43 μ in lateral aspect. The aperture is completely closed, but its position is marked by a slight thickening of the cyst-wall. The various appendages are still attached to the outside, but in a much reduced or altered condition. All nuclei of Appendage II have degenerated completely. The stalk of Appendage III has elongated and the free ends of the cells are frayed into a great number of separate strands (fig. 27). The nuclei have degenerated. In this condition the spread of the free ends of the appendage is about equal to the width of the entire cyst. Appendages IV and V appear as mere strands of tissue without a trace of nuclei. Appendage I, folded up inside the cyst, is so transparent that it becomes almost invisible. All the cells between the lining membrane and the cyst-wall have degenerated in the fully-formed cyst. The position of the encysted cercaria is different from that of *C. sagittarius*. In the latter species the body is folded twice, so that both extremities come to lie in the top of the cyst. In *C. sinitzini* the body is twisted once only, and both extremities lie in the bottom of the cyst. The anterior end of the cercaria lies in the dorsal half of the cyst. The body is twisted above the cuticular invagination of the cyst-wall, thus bringing the ventral sucker uppermost near the top of the cyst. The posterior end of the cercaria lies in the ventral half of the cyst. It should be remembered that the cyst is not spherical, but strongly compressed laterally.

Excystment of the Cercaria (figs. 28, 29). If light pressure is brought to bear on the cyst, the anterior end of the cercaria is pushed against the base of the dorsal half of the cyst close to the point of origin of the delivery-tube. This releases the delivery mechanism. Appendage I is everted—turned inside out—and thus shot out through the bottom of the cyst at the point of origin of Appendage V. The telescoped walls of the cells of which Appendage I is composed are thus extended once more to their utmost limit (fig. 28). It then measures about $4\frac{1}{2}$ times the length of the cyst itself. Immediately after eversion of Appendage I the cercaria is drawn rapidly down the tube and is projected to the outside.

It was possible to fix and mount specimens in all stages of excystment.

Excysted Cercaria.—When released from the cyst the cercaria moves about sluggishly. The body is still comparatively undifferentiated, but what appears to be the appendiculate portion of the tail has begun to form. In stained specimens, measuring 100 μ or over, two groups of cells in the posterior region stain more deeply than the rest, and probably represent the rudiments of the reproductive organs.

DISCUSSION.

Since the publication of Krull's paper describing the life-history of *Halipegus occidualis* Staff. 1905, the function of some of these extraordinary appendages of Hemiurid cercariae has been demonstrated for the first time. The delivery-tube shoots the encysted cercaria through the intestinal wall into the body cavity of a Copepod, which serves as the second intermediate host. The other appendages are believed to attract the Copepod, which releases the mechanism of the delivery-tube by manipulating, touching or becoming entangled in these appendages, in its endeavour to eat the cyst or its contents.

One of the most interesting aspects of the peculiar caudal appendages of the cystophorous cercariae is their great variety. In this connection it is worth recording that in the case of *C. sinitzini* there appears to be a general tendency for the tail-vesicle to produce accessory appendages. As already noted, a number of abnormal specimens were observed in which the terminal portion of Appendage IV was bifurcated, or else carried a terminal finger placed at right angles to the main prolongation. Other specimens showed a replica of Appendage V, set at right angles to it—formed by the outward migration of a second giant nucleus from the base of Appendage IV. All these appendages may possess some useful or vital function, but it seems quite possible that one or several of them have no function whatsoever. So long as any of the appendages came to serve a useful purpose, the tendency to produce accessory caudal outgrowths might become fixed in the whole group. As would be expected, the delivery-tube, the importance of which is now quite clear, is the least variable of all the appendages. It is fairly constant in structure except in the case of *C. laqueator* Sinitzin 1911, where it is drawn out into an immensely long filament and apparently never withdrawn into the tail-vesicle.

The extraordinary degree of specialisation shown by this group of cercariae is unique, and it is difficult to conceive how the delivery apparatus, with its peculiar function in the life-history of the cercaria, can have arisen. It is worth noting, however, that in the **Azygiidae** Odhner, a family of trematodes almost certainly related to the **Hemiuridae,** the cercariae also encyst by withdrawing into their own tails. Although this was probably the first stage in the evolution of the present peculiar method of excystment of the cystophorous cercariae, it gives us no hint how the extremely complicated and delicately adjusted delivery mechanism first came into being.

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EXPLANATION OF FIGURES.

Fig. 1. *Cercaria sinitzini.* (At this stage the body stains uniformly and the suckers are only indicated.) Tail in lateral aspect with Appendage I expanded. The internal cellular structure of the tail-vesicle is omitted in the figs. except figs. 2 and 5.

Fig. 2. Sagittal section of the tail-vesicle of a fully-developed cercaria before telescoping of Appendage I.

Fig. 3. Outline drawing of *C. sinitzini* from ventral aspect after withdrawal of Appendage I.

Fig. 4. *Cercaria sinitzini*. Body in ventral aspect and tail in lateral aspect after telescoping of Appendage I, of which one cell is already withdrawn into the tail-cavity. The giant nucleus of Appendage V is migrating back along the stalk of the appendage. The other appendages also show signs of degeneration. (Nuclei of primitive epithelium omitted.)

Fig. 5. Optical section of the tail-vesicle of a fully-developed cercaria after the telescoping of Appendage I, showing flattened nuclei of sub-cuticular layer and cell-group giving rise to Appendage III. The central cavity is not visible in all optical sections.

Figs. 6–9. Outline drawings of cercariae in different stages of development. Fig. 9 shows the lines along which measurements were taken for Table II.

Fig. 9*a*. Nuclei of primitive epithelium.

Figs. 10–12. Process of encystment. Appendages II and III have been omitted in figs. 10–13.

Fig. 13. Encysted cercaria. Lateral aspect.

Fig. 14. Outline drawing of Appendage IV, showing maximum development of terminal prolongation.

Figs. 15–18. Terminal portion of Appendage I, showing different stages of development.

Fig. 18*a*. Diagram showing the manner of telescoping of the cells of Appendage I.

Fig. 19. Appendage II at maximum stage of development. A portion of the cellular structure is drawn in to show the spiral arrangement.

Fig. 20. Terminal portion of Appendage II during encystment of cercaria. Nuclei of the spiral have degenerated. External nuclei are shrinking.

Fig. 21. Appendage II after encystment of cercaria.

Figs. 22–26. Appendage III at different stages. Fig. 26 shows the final condition after encystment of the cercaria.

Fig. 27. Empty cyst after projection of cercaria to the outside through the everted delivery-tube (Appendage I).

Fig. 28. Excysting cercaria about to pass down everted delivery-tube.

Fig. 29. Excysting cercaria caught in delivery-tube, which has failed to evert normally beneath a cover-slip.

EXPLANATION OF LETTERING.

A I	Appendage I.
A I*e*	Appendage I everted.
A II	Appendage II.
A III	Appendage III.
A IV	Appendage IV.
A IV*d*	Appendage IV degenerating.
A V	Appendage V.
A V*d*	Appendage V degenerating.
a	anterior end of cercaria.
ac	anterior lateral collecting-tube.

b	body of cercaria.
bc	body of cercaria caught in delivery-tube.
bl	bladder.
c	cavity.
ci	cuticular invagination.
cw	cyst-wall.
dn	degenerating nucleus.
dt	dorsal half of tail-vesicle.
ec	encysted cercaria.
gc	group of cells of unknown function.
gn	giant nucleus.
i	intestine.
ic	internal column of cells of Appendage IV.
lm	lining membrane of cyst.
mc	main collecting-tube.
n	nucleus.
*n*AV	giant nucleus of Appendage V.
nm	nuclear membrane.
o	oral sucker.
p	pharynx.
pc	posterior lateral collecting-tube.
pd	peduncle.
pe	primitive epithelium.
ps	posterior end of cercaria.
sh	sheath of Appendage I after eversion.
sl	subcuticular protoplasmic layer.
t	tail.
tc	tubular circuit of excretory vesicle.
tp	terminal prolongation of Appendage IV.
v	ventral sucker.
vt	ventral half of tail-vesicle.