A NEW LIVER FLUKE FROM A MONKEY AND NEW PARASITIC ROUNDWORMS FROM VARIOUS AFRICAN ANIMALS

By J. H. SANDGROUND

Of the Department of Tropical Medicine, Harvard Medical School, Boston, Mass.

The Harvard African Expedition of 1926-27, under the leadership of Prof. Richard P. Strong of the department of tropical medicine, Harvard University Medical School, spent some months in the Republic of Liberia and then crossed Africa from the Belgian Congo on the west to Mombassa on the east coast, making investigations of the natives and the local fauna from the medical and zoological aspects.

In the extensive collection of material brought back to the United States for detailed study are many tubes of parasites from various animals secured in the different localities where halts were made. Many of these parasites were assigned to me for identification and systematic study, and I take pleasure in recording my indebtedness to Professor Strong for this privilege.

The following paper, constituting part of the report on the parasite collection, contains a description of a new species of the trematode genus Dicrocoelium from the liver of a monkey, a new genus of the nematode family Strongylidae, also from a monkey, and a new species of the trichostrongylid genus Oswaldocruzia from a lizard. Nematodirus hopkeni (Leiper 1907) from the hippopotamus was well represented in this collection, and as a result of the reexamination of this parasite it is considered necessary to remove it from the genus Nematodirus and to create a new genus for its reception.

Several other helminths, probably new to science, are also present in this collection, but the inadequacy or the unfavorable condition of some of the material does not permit me to give a complete description and specific determinations of these worms at the present time.

As a matter of record, a list is appended of parasites present in the collection which could be identified generically, and in some instances specifically, with the material available.

An asterisk denotes a new host record of a previously described species of parasite.

TREMATODA

	INEMATODA	
Parasite	Host	Locality
Fasciola hepatica Linnaeus, 1758.	Bubalus caffer	Ituri Forest, Belgian Congo.
Anaplocephala gorillae Nybelin, 1927.		Congo.
Bothridium ovatum Diesing, 1850_		Rutchuru River, Belgian Congo.
	NEMATODA	
Murshidia species	Loxodonta africana	Semliki Valley, Belgian Congo.
Cobboldina vivipara Leiper, 1911_		Congo.
Setaria labiato-papillosa (Alessan- dri, 1838) Railliet and Henry, 1911.		Ruindi Plains, Belgian Congo.
Setaria poultoni Thwaite, 1928	Damaliscus tiang	Ruindi Plains, Belgian Congo.
Setaria hornbyi Boulenger, 1921_	* Cobus defassa	Ruindi Plains, Belgian Congo.
Cylicospirura subaeqalis Molin 1860.	* Felis leo	Rutchuru Plains.
Ophidascaris filaria (Dujardin, 1845) Baylis, 1921	Python species	Rutchuru Plains.
Streptopharagus pigmentatus (Linst, 1897) Railliet and Henry, 1918.		Liberia.
Trichuris trichiura (Linnaeus, 1771) Stiles, 1901.		
Enterobius species	*Cercopithecus dianae	Liberia.
Strongyluris brevicaudata Muller, 1894.		
Saurositus agamae Macfie, 1924.	Agama colonorum	Liberia.

TREMATODA

Family DICROCOELIDAE

The material collected from the liver of a species of Colobus monkey shot in the Ituri Forest, south of Lake Albert, Belgian Congo, consisted of some 15 small trematodes, which had been fixed in Zenker's solution. Of this number only three or four are not contracted and generally distorted to the extent that the structure of the parasites can not be profitably studied. The measurements given in the following description were taken from the most favorable specimens available mounted in gum damar after staining in carmine.

DICROCOELIUM COLOBUSICOLA, new species

Specific diagnosis.—Dicrocoelium: Length 3.6 mm. to 5.2 mm. The greatest width in the largest specimen is 1.9 mm. at a post-equatorial point level with the posterior border of the yolk glands.

The body is thin and flat with weakly developed musculature. The specimen least contracted, and hence chosen to represent the type, is spindle-shaped, the body tapering gradually to a rounded extremity anteriorly. In other specimens the preacetabular region is drawn out into a rather narrow neck. The superficial cuticle is devoid of scales, but the presence of numerous subcuticular cells gives it a granular appearance which to some extent masks the internal structure so that the more delicate details are difficult to determine. Oral sucker circular in outline, from 0.22 mm. to 0.28 mm. in diameter. It is terminal and ventral, with strong muscular rim nearly 90 wide. The oral aperture is subterminal and semilunar in shape. Muscular pharynx, 0.08 to 0.12 mm. in length, followed by a narrow esophageal region, about 0.15 mm. long, which bifurcates to form the intestinal ceca. Intestinal ceca simple, thin-walled, and so narrow that in some specimens their presence is determined with difficulty. In other specimens the ceca are about 0.12 mm. wide and run a slightly undulating course posteriorly on either side of the acetabulum to terminate in a small vesicle, just behind the middle of the body. The course of the intestinal ceca is usually entirely external to the vitellarian fields, but in some specimens the crest of the undulations overlie the vitelline glands. The acetabulum is muscular. In diameter it ranges from 0.20 to 0.28 mm., being slightly smaller in size than the oral sucker. The excretory vessel is a simple narrow tube, the lateral horns of which are not visible in mounted specimens. It opens at the posterior extremity of the body into a slight indentation of the contour of the body.

Male genitalia.—The testes are situated in the second quarter of the body. In the convenient terminology used by Stiles and Goldberger (1910) to describe the topography of the organs of trematodes, both the zones and fields of the two testes would be said to overlap, and the testes abut on their internal borders. In the type specimen the testes are deeply lobed and the area of the anterior testis is perhaps a little smaller than that of the posterior. In other specimens a lobed condition of the testes is not noted. The vasa-efferentia and the vas deferens are presumably too delicate to be observed in totomounts, but a well-developed, although small cirrus, which is somewhat coiled, can be seen. The genital atrium, which receives the cirrus, is situated just posterior to the point of bifurcation of the esophagus.

Female genitalia.—The ovary, measuring about 0.22 mm. by 0.28 mm., occupies a position immediately behind the right testis. It is usually ovoid in shape, sometimes almost spherical. A large receptaculum seminis, about 0.13 mm. in diameter, lies posterior to and in the same longitudinal field as the ovary. Neither Mehlis's gland nor Laurer's canal was observed. The coils of the massive

uterus are so massed together that the usual ascending and descending branches are not distinguishable. The transverse coils of the uterus occasionally extend almost to the margin of the body. The vitellaria consist of large aggregations of glands connected by rather narrow longitudinal ducts forming a moniliform band, which stretches on each side from a point just posterior to the vesicula seminalis to about the equator of the body. The transverse vitelline ducts were not conspicuous. The eggs in the posterior coils of the uterus are of a golden yellow color, but become darker as they advance toward the metraterm. In this part of the uterine tube, which passes directly under the acetabulum, the eggs measure 44μ to 48.2μ in length by 28.3μ in width; they have a slight shoulder, and are operculated.

Host.—Colobus species.

Location.—Liver (bile ducts).

Locality.—Ituri Forest, Belgian Congo, May 21, 1927.

Type.—Cat. No. 8012, U.S.N.M., Helm. Coll.; Cat. No. 8013, U.S.N.M.

Paratypes.—Helm. Coll.

SYSTEMATIC POSITION

The species described above is a typical member of the genus Dicrocoelium Dujardin, 1845, the number of species of which are numerous. They are distributed in a cosmopolitan manner in reptiles, birds, and mammals, usually occupying the bile and pancreatic ducts. The course that the intestinal ceca pursue, external to the line of vitelline glands is rather unusual, but the present species shares this character, at least, with D. hospes Looss, 1907, found in Egyptian cattle, and with D. macrostomum Odhner, 1911, of Numida ptilorhyncha from the White Nile. The present species resembles D. macrostomum to a remarkable extent, the two forms being almost identical with regard to the size and disposition of the organs. The only differences that can be detected from Odhner's description of the species concerns the inconspicuousness or absence of a receptaculum seminis (not mentioned in D. macrostomum) and a slightly greater length of the intestinal ceca relative to the total length of the body. The former difference may be apparent rather than real, depending upon the physiological condition of the genitalia at the time of examination, and the latter difference is so small as to be of doubtful significance. It seems, however, rather unlikely that the present material coming from a monkey is identical with the flukes from a bird derived from a totally different locality. For this reason, it is proposed to credit the two points of morphological difference, noted above, with specific value. The name, D. colobusicola, is provisionally proposed for the material under consideration, pending the opportunity for making a comparative examination with D. macrostomum.

Family TRICHOSTRONGYLIDAE

LEIPERIATUS, new genus

Generic diagnosis.—Trichostrongylidae: Dorsal lobe of bursa reduced. Two spicules, with relatively massive, ridged, proximal proportions, and with flexible, filiform terminal appendages.

Type species.—Leiperiatus hopkeni (Leiper, 1910) new combination.

LEIPERIATUS HOPKENI (Leiper, 1910), new combination

Synonym.—Nematodirus hopkeni Leiper, 1910.

Specific diagnosis.—Leiperiatus: In the preserved state the worms are of a greenish yellow color. The cuticle is finely striated transversely, and there is also a series of a dozen or more longitudinal lines extending the entire length of the body. The cuticle of the head is not inflated. The head is about 24μ wide at the extremity. The mouth cavity is surrounded by four papillae, two subdorsal and two subventral, and a pair of amphids, laterally. The amphids, which are well developed, are considerably larger than the papillae, which are minute and inconspicuous. The buccal cavity is very shallow and into it there projects a very definite, although at times obscured, cuticularised spine, about 6.5µ long, which arises from the floor of the mouth. The esophagus, of the typical trichostrongyle type, widens only slightly posteriad. It is about one-twelfth of the body length in the male and about one-ninth of the body length in the female. The nerve ring embraces the esophagus anteriorly in the first quarter (0.28 mm. to 0.32 mm. from its anterior end), and at the same level the excretory tube opens by a fine duct on the ventral surface. Cervical papillae were not observed in the material at hand. They may be present as minute acicular points, which are stated by Leiper to project 0.4 mm. behind the nerve ring.

Male.—12 to 13.4 mm. long with a maximum thickness of 0.23 mm. The bursa consists of two symmetrical lateral lobes united by a small unindented lobe, dorsally. The lateral lobes are figured, seemingly over the entire internal surface, with macular markings arranged to form a delicate mosaic design, and their posterior margins are finely scalloped by the cuticular striae which are quite conspicuous in this region. The small dorsal lobe is supported by a single dorsal ray, of proportionately reduced dimensions, which bifurcates near the middle of the lobe. Each of the bifurcations terminates in two minute digits which are slightly curved and extend to the margin of the lobe. The lateral lobes are supported by six rays of which the latero-ventral and ventro-ventral, as seen in the normal condition of the bursa, appear to be closely approximated, but when the lobes are spread out it is seen that the tips of these rays are well separated. The three lateral rays are parallel; the postero-lateral is the smallest and the

medio-lateral the stoutest. The externo-dorsal ray, the basal origin of which could not be definitely determined, is very slender. A pair of exceedingly minute prebursal papillae are also present.

The spicules are equal in size, about 0.31 mm. in length, and brown in color. Each is composed of a proximal and terminal portion. The proximal portion, about 0.18 mm. long and 0.025 mm. broad, is adorned with one or two, somewhat twisted, longitudinal crests or ridges, and appears to be tubular in form. The distal portion is filiform and flexible, often being bent, as Leiper described, in the form of an interrogation mark. The tips of the spicules are not united by a membrane. The genital cone lies ventral and just anterior to the dorsal lobe of the bursa. It is ornamented with tuberclelike papillae, but details of this structure could not be seen in the material available.

Female.—18 to 22.0 mm. long with a maximum diameter of about 0.30 mm. near the middle. The terminal part of the intestine is narrowed to form the rectum with cuticularized lumen which opens at a point where the body is 0.12 mm. wide, about 0.37 mm. from the posterior extremity. The body narrows gradually to form a conical tail. The vulva is without salient lips and, in a specimen 19.3 mm. long, is situated 45 mm. from the posterior extremity. The long uteri open through divergent muscular ovejectors into a short vagina. The eggs are ellipsoidal and have thin shells which measure from 66μ to 73μ long by 40μ wide.

Host.—Hippopotamus amphibius.

Location.—Stomach (?).

Locality.—Lake Albert, Central Africa, April 17, 1927.

Specimens.—Males and females, Cat. No. 8014, U.S.N.M., Helm. Coll.

The above description is based upon material collected and preserved separately from two hippopotami. The parasites, of which about 20- worms were collected from each animal, appear to be identical, but the material in one tube only was in reasonably good condition for study; because of the fixation the material from the second animal could not be cleared to show the internal anatomy. The species was first recorded and briefly described by Leiper (1910), who collected it from a hippopotamus shot in the Uganda and named it Nematodirus hopkeni. Leiper's description is unfortunately incomplete, and the diagrams accompanying the description are not accurate enough in certain details to be of service for the indentification of the parasite. The shape of the spicules, however, is so distinctive that although the range of the present material is slightly larger than shown by Leiper's figures, there can be little, if any, doubt that we are concerned with the same species. Leiper described the bursa of the male as being devoid of a posterior or

dorsal lobe and its supporting dorsal ray. This discrepancy from the present description can be accounted for by the fact that this organ, although it must be constantly present, is inconspicuous. was only exposed and visible in two of the four male specimens examined. In preparing the description presented above, a number of significant points of divergence were found distinguishing the species from the 12 or more species of the genus Nematodirus, and calling for a reconsideration of the taxonomic status of the worms. One of the most outstanding differences concerns the spicules, which in their filiform shape and membranous union are a constant feature of all other species of Nematodirus. The inclusion of "N. hopkeni" in the genus breaks the natural homogeneity of the spicule character and, particularly if other significant morphological differences could be found to support the action, it would be advisable to remove the species N. hopkeni from the genus Nematodirus. A comparison of "N. hopkeni" with other described species of Nematodirus provides the following additional points of departure:

	"N. hopkeni"	Other species of Nematodirus
1. Cuticle of head 2. Position of excretory pore 3. Size of eggs 4. Shape of tail in female 5. Dorsal lobe of bursa	At level of nerve ring. 73μ by 40μ Conoid	

These differences are of greater than specific magnitude and warrant the removal of "N. hopkeni" from the genus Nematodirus. I propose creating for its reception the new generic name Leiperiatus in honor of the original describer of the parasite.

Family TRICHOSTRONGYLIDAE

OSWALDOCRUZIA AGAMAE, new species

Specific diagnosis.—Oswaldocruzia.

Male.—Length 7.4 mm.; greatest breadth 0.15 mm. near middle of body from which point there is a gradual tapering toward the anterior end, where the body is 0.03 mm. wide. The head is rounded and bears 4 inconspicuous oral papillae and 2 amphids. The cephalic cuticle is inflated for a distance of about 35μ . The only occurrence of striae on the cuticle is for a short distance behind the cephalic inflation, the cephalic cuticle itself being as devoid of striae as the remainder of the body. The esophagus is 0.34 mm. long, claviform, and encircled by the nerve ring, slightly anterior to its middle. A

minute cervical papilla is found opposite the nerve ring. The excretory pore is situated just anterior to the base of the esophagus.

The bursa is slightly longer than broad, and its comparatively narrow supporting rays, as indicated in the accompanying diagram, are according to the plan characteristic of the genus. A small dorsal lobe is easily distinguishable. The dorsal ray bifurcates near its end, and each branch is split into three rather minute digitations. The spicules are of a light yellow color. They measure about 0.175 mm. in length, and at their broadest point, near the anterior end, measure 17μ wide. They are very slightly ridged, not spirally fluted, and their termini are adorned with a few inconspicuous processes. Gubernaculum absent.

Female.—Length from 11.5 to 12 mm.; greatest breadth 0.19 mm. The esophagus is 0.55 mm. long and the cephalic inflation measures about 0.05 mm. in length. The vulva is situated 7 mm. from the anterior end, dividing the body in the ratio of 3:2. The uteri are divergent, and the anterior ovary extends forward almost to the level of the base of the esophagus where it is reflected backward. The eggs, which become embryonated before oviposition, measure on the average 86μ by 45μ . The body of the female tapers gradually to end in a conoid tail to which a fine acicular process is appended.

Host.—Agama colonorum.

Location.—Intestine.

Locality.—Du River, Liberia.

Type.—Male and female, Cat. No. 8015, U.S.N.M., Helm. Coll. Paratypes.—Male and females, Cat. No. 8016, U.S.N.M., Helm. Coll.

The species described above which appears to be the first member of the genus described from an African reptile, may be differentiated from previously described members of the genus on the basis of (1) its unstriated cephalic cuticular swelling, (2) the narrow and relatively simple spicules, which are also shorter than in other species, and (3) the smaller eggs.

NEMATODA

Family STRONGYLIDAE

COLOBOSTRONGYLUS, new genus.

Generic diagnosis.—Strongylidae: Comparatively large worms with mouth directed straight forward. Buccal capsule infundibular and with thick walls. An external leaf crown with numerous slender elements and an internal leaf crown of minute elements present. Only the amphids, or so-called lateral papillae, are prominent in the circumoral region. Cervical papillae very minute or absent. Anterior portion of esophagus bent away from the main axis of the esophagus;

from its dilated funnel-shaped portion, three narrow teeth project into the mouth capsule. Male bursa with lateral lobes and an inconspicuous dorsal lobe. Ray formula as in genus Oesophagostomum. Spicules long and narrow, with knobbed proximal ends and without a sheath. Gubernaculum present. Tail of female mucronate. Anus and vulva open close together near tail. Vagina long, opening into two kidney-shaped chambers (pars ejaculatrix) which communicate with long ovejectors and parallel uteri.

Type species.—Colobostrongylus strongi, new species.

COLOBOSTRONGYLUS STRONGI, new species

Specific diagnosis. - Colobostrongylus: In the preserved state the worms are girdled either at their anterior or posterior extremities with a belt of brownish-black material resembling clotted blood. They are yellowish in color and robust in build. Body cylindrical, tapering toward the anterior extremity. The cuticle is finely striated, and, as a marking superimposed on the general striation, the cuticle is coarsely crinkled in the anterior and posterior regions of the body. Head region well defined. There are four circumoral papillae and two amphids, or so-called lateral papillae. The submedian, dorsal, and ventral papillae are flat, and visible only when the head is cut off and viewed on end. The amphids are fairly conspicuous, and with a broad base, provided with a terminal sensory filament, Mouth directed straight forward, with comparatively narrow mouth collar. Diameter of mouth, 0.085 mm. Mouth capsule broader than long, measuring 0.12 mm. by 0.11 mm., and with a thick chitinous wall. The dorsal gutter of the esophageal gland was not visible, and is probably absent. There are two leaf crowns, the external composed of 24 narrow elements, which project for a short distance beyond the mouth aperture, and an internal leaf crown consisting of exceedingly minute peglike elements, whose number can not be accurately computed. The esophagus is clavate, the posterior swollen portion being 230 µ wide. Its anterior quarter is bent at an angle from the main axis of the esophagus, and anteriorly it is widened out to form a funnel from the base of which there projects, halfway into the buccal cavity, a chitinous trident composed of three narrow lancets. In some specimens the trident is rather difficult to see on account of débris obscuring the view. Excretory tubule very narrow, opening at a level with the kink in the esophagus, 0.31 mm. from the anterior extremity. Cervical papillae were not visible.

Male.—From 24 mm. to 26 mm. in length with a maximum breadth of 0.47 mm. near the middle. Bursa short, measuring 0.23 mm. long by 0.4 mm. wide. Ray formula similar to that of the genus Oesophagostomum; ventral rays cleft near their base and parallel. Externolateral and other lateral rays arise from a common trunk; the former diverges and its tip does not quite reach to the margin of the bursa,

while the mediolateral and posterolateral rays are parallel, and extend to the margin of the bursa. The externodorsal rays, which are relatively slender, arise from a common trunk with the dorsal ray. The latter divides near its middle, and each branch bifurcates terminally. Spicules equal in length, filiform and unsheathed. They measure 1.23 mm. in length, and their termini are without barbs. A slender, curved gubernaculum, about 0.10 mm. long, is present.

Female.—From 30 to 31 mm. in length, with maximum breadth in the posterior half of the body of 0.78 mm. Esophagus 0.78 mm. long. Measurements of buccal cavity approximate those of the male. The posterior half of the body tapers conically, to terminate in a sharp mucronate tail. Anus opens about 0.15 mm. from tip of the tail, and the vulva, which does not have salient lips, is situated about an equal distance anterior to the anus. Muscular vagina about 0.28 mm. long. It bifurcates into two more or less kidney-shaped chambers, that receive two convergent ovejectors, which are continuous with the parallel uteri. Eggs (in uterus) thin shelled, measuring, on the average, 80μ by 40μ .

Host.—Colobus polykomos ("Black and white" Colobus monkey).

Location.—Small intestine (?). Locality.—Du River, Liberia.

Type.—Male and female, Cat. No. 8017, U.S.N.M., Helm. Coll.

Paratypes.—Cat. No. 8018, U.S.N.M., Helm. Coll.

Nine female and seven male specimens of the parasite described above were found free in the lumen of the small intestine of the host, which, at the time of examination, had been dead for several hours. Cysts, the size of a pea, resembling those produced by *Oesophagostomum* were seen by Dr. Max Theiler, a member of the expedition, on the walls of the cecum, but on dissection of one of these cysts no parasites were found. Because of the generic affinities of the worms, it seems not unlikely that the normal habitat of the parasite is in the large intestine and that, in the present case, post-mortem migration had occurred.

The characters described in the diagnosis can not be reconciled with those of any known genus of the Strongyloidea. On the basis of the shape of the buccal cavity and the associated oral structures, the status of the parasite would seem to fall between the two chief subfamilies, the Strongylinae and the Trichoneminae, but in other characteristics it exhibits affinities with the Oesphagostominae. The shape of the buccal cavity is closer to that of the Strongylinae, but the absence of a dorsal esophageal gland prolonged as a ridge on the dorsal wall of the buccal capsule is more characteristic of the Trichoneminae. If it were not for the lack of a transverse ventral cervical groove and of any semblance of cephalic inflations, an affinity with the Oesophagostominae would be indicated, especially since the

bursal ray formula is practically identical with, and the dental armature of the esophagus resembles these structures in *Ternidens*, a typical genus of the Oesophagostominae. In view of these considerations, the writer has refrained from indicating the subfamily relation of *Colobostrongylus*. It seems, however, that the Oesophagostominae, as at present defined, constitute an artifical group, necessitating the allocation to other subfamilies of several genera (as *Oesophagostomoides* Schwartz, 1928) whose characteristics show a close affinity with the type genus *Oesophagostomum*.

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EXPLANATION OF PLATES

Abbreviations: a. acetabulum; an. anus; c. cirrus organ; d. dorsal ray; exd. externo-dorsal ray; exl. externo-lateral; exv. excretory vessels; g. p. genital pore; gub. gubernaculum; i. intestinal caeca; l. v. latero-ventral; met. metraterm; m. l. medio-lateral; o. ovary; oe. esophagus; os. oral sucker; ph. pharynx; p. l. postero-lateral; pp. prebursal papilla; s. r. seminal receptacle; t1 and t2 anterior and posterior testes; u. uterus; v. vitellaria; vu. vulva; v. v. ventro-ventral.

PLATE 1

Colobostrongylus strongi

- Fig. 1. Anterior extremity showing buccal teeth and kink in esophagus.
 - 2. Posterior extremity of female showing vulva, vagina, etc.
 - 3. Bursa of male flattened out to show distribution of rays.
 - 4. Posterior extremity of male.

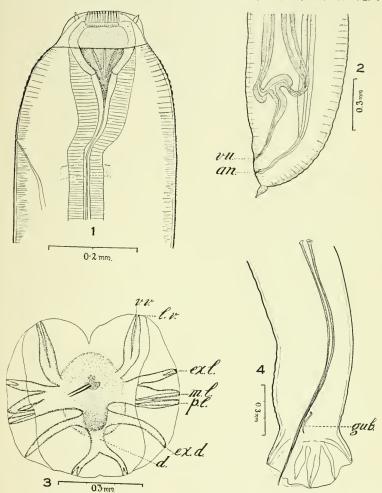
PLATE 2

- Fig. 1. Oswaldocruzia agamae, caudal extremity of male with bursa spread.
 - 2. Oswaldocruzia agamae, anterior extremity of female.
 - 3. Leiperiatus hopkeni, anterior extremity from the dorsal aspect.
 - 4. Leiperiatus hopkeni, caudal extremity of female viewed from the side.
 - 6. Leiperiatus hopkeni, caudal extremity of male with bursa spread.
 - 5. Dicrocoelium colobosicola, ventral view showing topography of organs.



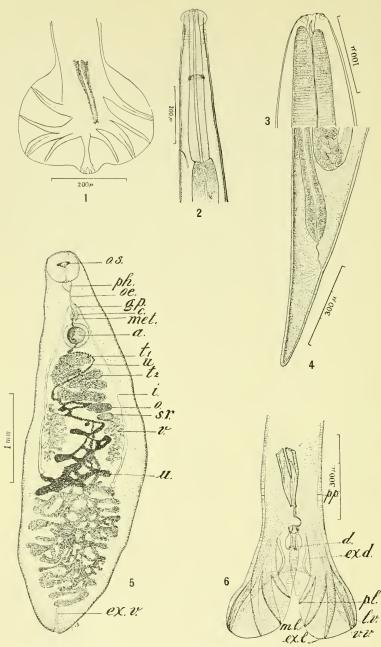
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COLOBOSTRONGYLUS STRONGI

FOR EXPLANATION OF PLATE SEE PAGE 11



OSWALDOCRUZIA AGAMAE, LEIPERIATUS HOPKENI, AND DICROCOELIUM COLOBOSICOLA

FOR EXPLANATION OF PLATE SEE PAGE 11