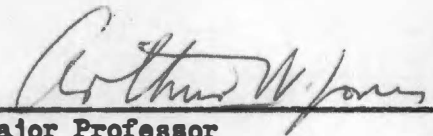


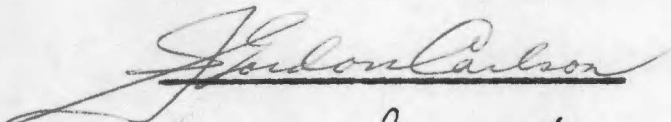
August 8, 1950

To the Committee on Graduate Study:

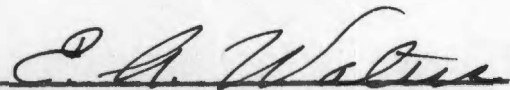
I am submitting herewith a thesis written by Theodore Gates Brown, Jr. entitled "The Head Organs of Cleidodiscus mirabilis (THEMATODA, MONOGENEA)." I recommend that it be accepted for nine quarter hours credit in partial fulfillment of the requirements for the degree of Master of Science, with a major in Zoology.


Major Professor

We have read this thesis
and recommend its acceptance:


Geo. K. Schweitzer

Accepted for the Committee:


Dean of the Graduate School

HEAD ORGANS OF CLEIDODISCUS MIRABILIS (TREMATODA, MONOGENEA)

A THESIS

Submitted to
The Committee on Graduate Study
of
The University of Tennessee
in
Partial Fulfillment of the Requirements
for the degree of
Master of Science

by

Theodore Gates Brown, Jr.

August 1950

ACKNOWLEDGMENT

The author wishes to express his sincere appreciation to Dr. Arthur W. Jones for his encouragement and assistance, and to Dr. John D. Mizelle of Notre Dame University for verifying the identification of the specimens.

T. G. B.

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INTRODUCTION

Little or no detailed description has been heretofore attempted on Cleidodiscus, the work to date having been confined to taxonomy and general morphology (Mueller, 1934 and 1937; Mizelle, 1938; Mizelle and Gromin, 1943).

The head organs and cephalic glands were chosen for this study because of their conspicuous nature in the living animal and, at the same time, their confinement to a relatively small anatomical area. Since no minute description of these structures is known, an attempt was made to discover their exact histologic nature. The methods to be used were to include well known techniques of fixation, sectioning, and staining, but, in addition, an attempt was to be made to try to evaluate the usefulness of the phase-contrast microscope as a supplementary tool especially for examination of living material.

MATERIALS AND METHODS

Specimens were collected from the gill filaments of two species of catfish; the Yellow Cat (Pilodictis olivaris Rafinesque) and the Blue Cat (Ictalurus furcatus Cuvier and Valenciennes). All hosts were taken from Fort Loudon Lake, Tennessee River at Knoxville, Tennessee, and Lenoir City, Tennessee. Some of the specimens were not removed from the host until several hours after it had been taken out of the water. In such instances all the specimens were found to be dead when examined. The majority of the hosts were examined for parasite infestation within thirty minutes after removal from the lake. In such instances most of the specimens were still alive when examined under the wide-field microscope. Some of these living specimens were immediately removed from the gill filaments. The remaining specimens were left attached to the gill filaments and kept alive by placing the filaments in an aerator. The aerator consisted of a one-liter suction flask two-thirds filled with water. A single-holed rubber stopper was placed in the top of the flask and a piece of glass tubing run to the bottom of the flask. Air was then drawn into the flask by suction and allowed to bubble up through the water. The gill filaments were then suspended about one inch below the surface of the water in the path of the rising air bubbles. Specimens attached to the gill filaments were kept alive by this method four to six hours.

The dead specimens were fixed in Carnoy's and mounted as whole mounts. Some were stained with Delafield's hematoxylin before mounting

them in Piccolyte¹ and others were mounted unstained in glycerine chromate gel (Yetwin, 1944). The living specimens were fixed in Carnoy's, Zenker-formol, or Flemming's solutions. Zenker-formol proved to be the most satisfactory fixative among those tried. Specimens fixed with Zenker-formol showed less shrinkage and more cellular detail than those fixed in either of the other fixatives used. Specimens for sectioning were dehydrated and imbedded in paraffin. Sections were cut at ten micra mainly in transverse planes although some were sectioned in sagittal or frontal planes. All sections were stained with Heidenhain's iron hematoxylin and destained with picric acid, then mounted in Piccolyte.

Specimens for examination alive were mounted in water on slides, under cover slips ringed with vaseline. The cover slip was pressed gently upon the slide so that the specimen had a minimum of vertical space in which to move about. In this way activity was reduced without crushing the specimen. The specimens remained alive, in most cases, for two to three hours. Examination of the living specimens thus prepared was made with the phase-contrast microscope. Best examination results were obtained with Spencer dark contrast-medium 4 mm. objective and the dark contrast-medium 1.8 mm. oil immersion objective. Details seen with the phase-contrast microscope were so numerous that there was a tendency during first observations to confuse many of the structures. This tendency was easily overcome once one became accustomed to using the phase technique. Photomicrographs were made of the head region of living whole mounts (to

¹A synthetic resin obtainable from the General Biological Supply Company, Chicago, Illinois.

be compared with similar photomicrographs of fixed whole mounts). See Plate I. The pictures were made with the phase-contrast microscope using the 4 mm. dark contrast-medium objective. Exposures were made at ten inches from the eyepiece for one-half second with the Bausch and Lomb research ribbon filament lamp at nine-tenths greatest intensity. Eastman Tri-X Pan film was used, and it was developed in Eastman D. K. 76 developer.

Semi-diagrammatic drawings were made of the head region showing the cephalic glands, ducts, and head organs in their relationship to the pharynx (see Plate II, Figure 1); and of the general anatomy of a whole specimen (see Plate II, Figure 2). Camera lucida drawings were made of typical transverse sections of the cephalic glands (see Plate II, Figure 3).

OBSERVATIONS

The parasites used in this study are moderately large dactylogyrids, with a smooth cuticula. The body is flattened, has an elliptical shape and a hexagonal opisthohepator. Two pairs of eyespots are present. Four anchors are present on the opisthohepator, similar in shape and size. Two non-articulate bars are present, each of which connects the base of a pair of anchors. There is a single testis and ovary. The ovary is anterior to the testis and two or three times larger. The copulatory complex is situated just posterior to the esophageal bifurcation. Vitellaria are present in two lateral bands along the body. The pharynx is well developed and the mouth is located in the mid-ventral region near the level of the anterior eyespots. The short esophagus bifurcates to form two lateral intestinal caeca which unite posteriorly. For a more complete description of the species studied see Mueller (1937) and Mizelle and Cronin (1943).

The head organs of the probpator are situated on the antero-lateral margins of the head. There are usually four to six pairs. Although in a few instances a seventh organ was found on one side, never were less than four pairs observed. The organs are thickened elliptical tubes of from 0.009 to 0.015 mm. in length, and 0.002 to 0.006 mm. in width. They are connected at their proximal end to a duct which runs posteriad to the cephalic glands. The head organs were seen extruding the mucoid secretion of the cephalic glands from varying pairs of the organs at different times, indicating the possible presence of some control mechanism.

When examined with the phase-contrast microscope, the head organs were seen to contain a thread-like material, similar to the contents of the rhabdite-glands of certain tubellaria. In fixed specimens the organs showed a granular composition. This granular material is probably the result of fixing the rhabdite filaments seen in the living specimens.

The cephalic glands are situated just anterior, lateral, and just posterior to the pharynx. They are composed of several lobed structures which when examined in serial sections proved to be syncytial masses each with several prominent nuclei. The lobes are usually 0.007 to 0.014 mm. in diameter. The nuclei were all approximately the same size. They were about 0.002 mm. in diameter and several times larger than the granules in the cell, which are stained dark like the nuclei. When observed with phase the living animal showed these same characteristics.

The glands are all connected by ducts with the head organs. The ducts seem to be extensions of gland cells. The glands on each side of the animal supply their respective head organs.

DISCUSSION

In the Monogenea the anterior adhesive apparatus or prohaptor may take one of several forms (Dawes, 1945). In some the mouth is encircled by an oral sucker, in others there is a pair of suckers which may be closely associated with the mouth or situated some distance from it. Frequently the prohaptor is ill-defined, not sucker-like, or absent. In a few instances there are two grooves which undoubtedly serve a suctorial purpose. Sometimes, instead of the grooves, there are lateral expansions of this region of the head called head lappets, or there may be papilla-like outgrowths of more compact form which are called head organs. Associated with the head organs are multi-cellular cephalic glands and ducts by which a sticky substance is formed and passed out onto the surface of the prohaptor. Head organs may vary in number and are important taxonomic characters. In some monogenea neither head lappets nor head organs occur, but antero-lateral glandular areas coexist with anterior suckers. In rare instances the mouth is encircled by a somewhat membranous structure called a pseudosucker.

Whatever its structure, the primary function of the prohaptor is to apply the anterior tip of the animal to the substratum during the feeding operation. It is also used, as the author repeatedly observed, in locomotor function, being capable of preserving attachment to the host when the animal is seeking a fresh hold with the opisthohaptor. The alternate action of these two sets of organs, together with muscular movements of the body, may produce a rudimentary looping movement, although

the animal probably does not move far once it has established itself on the host.

The general structure and anatomy of the prohaptor of Cleidodiscus mirabilis is certainly not unusual or strikingly different from many other forms of Monogenea (such as Ancyrocephalus and Tetraonchus). Similar structures with similar functions are to be found in certain of the Turbellaria (Rhynchomesostomum).

Almost all of the details of structure of these animals could be seen in living specimens with the phase-contrast microscope. As a check on the structures thus seen the known methods of examination were used, i.e., fixation, sectioning, and staining. The latter techniques corroborated the findings made with the phase-contrast microscope especially for examination of living material.

SUMMARY

1. Specimens of Cleidodiscus mirabilis were obtained from the catfish and the finer structure of the head organs studied.
2. Fixation in Zenker-formol proved best for sectioned material and in Carnoy's for whole mounts.
3. The head organs were found to consist of four to six pairs of ducts opening to the antero-lateral margins of the probaptor.
4. The head organs are connected to synctial masses forming the cephalic glands.
5. The cephalic glands are lobate in outline and are situated near the pharynx.
6. The cephalic glands produce a mucoid secretion of rhabdite nature which aids in attachment of the probaptors for feeding and locomotion.
7. Examination of living material with the phase-contrast microscope aided materially in this study.

BIBLIOGRAPHY

BIBLIOGRAPHY

- Dawes, Ben. 1946—~~The~~ Trematoda. University Press, Cambridge, XVI + 644.
- Mizelle, John D. 1938—Comparative studies on trematoda (Gyrodactyloidea) from the gills of North American fresh-water fishes. Illinois Biol. Monogr. 17(1):1-81.
- Mizelle, John D. and John P. Cronin. 1943—Studies on monogenetic trematodes. X. Gill parasites from Reelfoot Lake fishes. Amer. Midl. Nat. 30(1):196-222.
- Mueller, J. F. 1934—Parasites of Oneida Lake fishes. Part 4. Additional notes on parasites of Oneida Lake fishes, including descriptions of new species. Roosevelt Wildlife Annals. 3(4):335-373.
- , 1936—New gyrodactyloid trematoda from North American fishes. Trans. Amer. Micros. Soc. 55(4):457-464.
- Yetwin, I. J. 1944—A simple permanent mounting method for *Necator americanus*. J. Parasitol. 30:201.

CRANESEST CREST

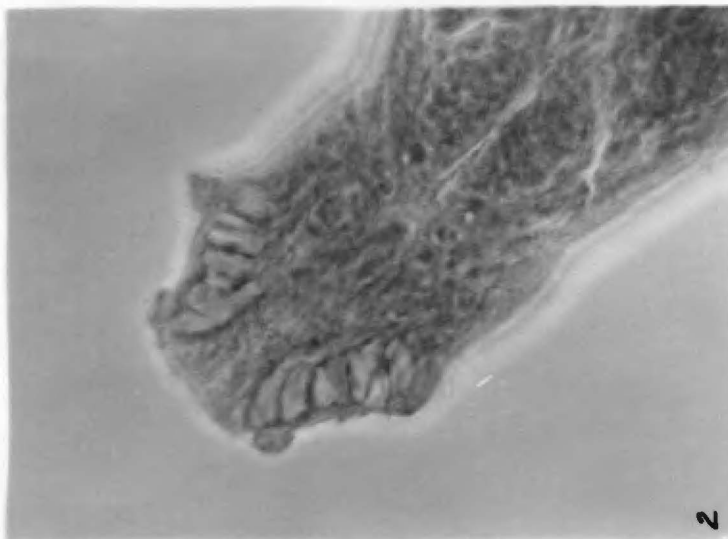
PLATES

CRANESEST CREST

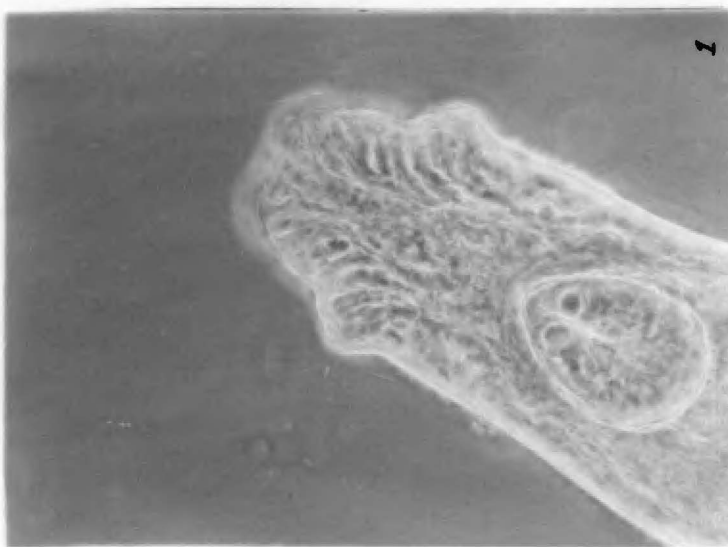
Plate I

Figure 1. Photomicrograph of living specimen showing head region. X 500.

Figure 2. Photomicrograph of fixed specimen showing head region. X 470.



2



1

Plate I

Plate II

Figure 1. Semi-diagrammatic drawing showing head region. X 620.
Abbreviations:

ho - head organ	p - pharynx
cg - cephalic gland	d - duct
e - eye	l - lens of eye

Figure 2. Semi-diagrammatic drawing showing whole mount. X110.
Abbreviations:

hg - head organ	o - ovary
c - cirrus	ce - caeca
ep - egg pore	t - testis
sv - seminal vesicle	vt - vitellaria
sr - seminal receptacle	v - vagina

Figure 3. Camera Lucida drawings showing outline of cephalic gland lobes with the nuclei.

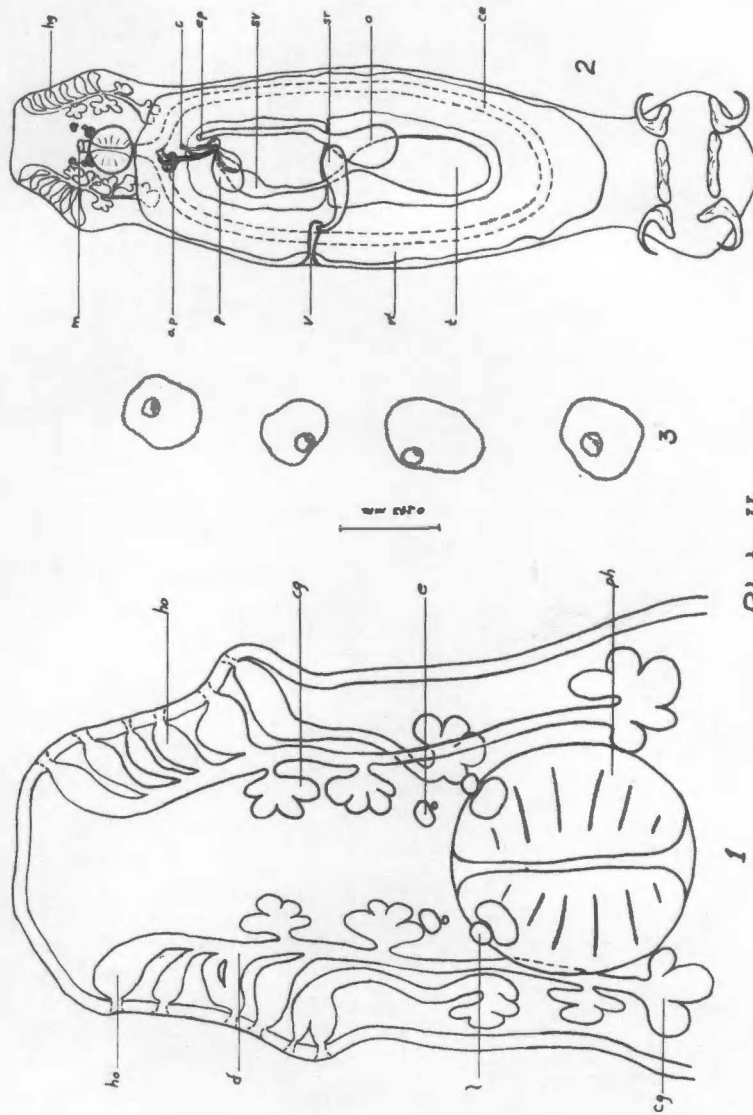


Plate II