THE ALIMENTARY TRACT OF THE MARGINED BLISTER BEETLE, EPICAUTA CINEREA MARGINATA FAB. (COLEOPTERA—MELOIDAE)[1]

RAY THOMAS EVERLY
Holmesville, Ohio

During the Fall of 1927, while engaged on field work for the United States Department of Agriculture in Meigs and Monroe counties in southeastern Ohio, several heavy infestations of adult blister beetles of the species *Epicauta cinerea* Forst. variety *marginata* Fab., (the margined blister beetle), were observed upon the flowering spikes of ragweed, (*Ambrosia elatior* L.), and several hundred specimens were collected. These were fixed in Kahle's solution and stored in 70% alcohol.

Sections were made of the internal organs and stained with haemalum and fast green. Repeated trials proved that over-staining in haemalum, 2 to 3 hours, and very short staining in the fast green, 8 to 10 seconds, gave the best differentiation of the tissues and cells. All sections were cut 10 microns in thickness.

The investigation was carried on under the direction of Dr. C. H. Kennedy, to whom appreciation is expressed, and to fellow students in the graduate department of Entomology, in particular to Mr. R. H. Davidson, Mr. P. E. Schaefer, and Mr. F. B. Whittington, and to Mrs. Laura Wadsworth Everly for assistance with the manuscript.

THE GROSS ANATOMY OF THE DIGESTIVE TRACT

The alimentary tract in general, is similar to that found in other coleopterous insects. It consists of three main divisions, the fore-intestine, (stomodeum), the mid-intestine, (mesenteron), and the hind-intestine, (proctodeum), and is approximately one and one-third times the body length, (Plate I, Fig. 3). The fore and hind-intestines are ectodermal in origin, whereas the mid-intestine originates from the mesoderm.

THE FORE-INTESTINE

The fore-intestine occupies approximately one-fourth the alimentary tract and consists of the oesophagus, a long narrow tube showing but little variation in diameter until just before reaching the mid-intestine,

[1]Thesis presented as partial fulfillment of the requirements for the degree of Master of Science.

where it abruptly widens into the gizzard. There is no external constriction at the oesophageal valve. Sections of the gizzard show very characteristic foldings of the intima, (INT, Plate I, Fig. 1). The oesophageal valve is definitely a part of the fore-intestine, as a tension placed upon the alimentary tract separates the fore and mid-intestines, the oesophageal valve remaining attached to the former. A gross examination discloses this valve to consist of four finger-like projections, which extend into the mid-intestine about one-fifth its length. These projections are attached at the base and are free the greater portion of their length. They appear to be of similar shape and size, (Plate II, Fig. 4). The outer edges are heavily chitinized on the basal third and gradually less so to the apex. This chitin is pigmented with a yellow color, which is retained in sections and does not stain.

THE MID-INTESTINE

The mid-intestine occupies about one-fourth the entire alimentary tract. It is a large and broad organ, filling the greater portion of the abdomen. Externally it appears folded and covered with minute papillae, the latter being the infolding of the epithelial tissue, which is apparent through the thin translucent connective tissue surrounding the entire mid-intestine.

THE HIND-INTESTINE

The mid-intestine narrows more or less abruptly into the hind-intestine. At this point the six malpighian tubules first enter the tract. Posterior to their point of entrance, the ileum turns abruptly anteriorly. Along this curvature striations or ridges appear externally, which are apparently due to the peculiar arrangement of the circular muscles and the heavier longitudinal muscles at this point, (C M, L M, Plate III, Fig. 8). This part of the intestine appears dark and heavy walled, attributable to the heavy musculature of this part of the hind-intestine. The ileum proceeds anteriorly to about the midpoint of the mid-intestine, at which point the six malpighian tubules enter the tract the second time. At this point of entrance they do not penetrate into the lumen, but remain just beneath the connective tissue. Here the ileum once more turns and then proceeds posteriorly, gradually widening into the colon, and then, more or less abruptly, into the short, broad, bulbous-appearing rectum. The hind-intestine occupies about one-half of the alimentary tract.

THE HISTOLOGY OF THE ALIMENTARY TRACT

THE FORE-INTESTINE

A histological examination of the fore-intestine shows it to consist of the following tissues, the connective tissue, (often misnamed the peritoneal membrane), the circular muscles, the longitudinal muscles, the hypodermal cells, and the intima, (Plate I, Figs. 1 and 2); Plate II, Fig. 5).

The connective tissue is a very thin membranous-appearing layer of

cells, entirely surrounding the alimentary tract and its appendages. In the fore-intestine it retains its membranous appearance the entire length.

Immediately within the connective tissue is the layer of circular muscles, consisting of a relatively few fibers in the anterior portion of the oesophagus, (C M, Plate I, Fig. 2), to a layer four or five fibers in thickness around the gizzard, (C M, Plate I, Fig. 1), and the oesophageal valve, (C M, Plate II, Fig. 5).

Just within the layer of circular muscles are the longitudinal muscles, which nearly fill the folds of the intima in the anterior portion of the fore-intestine, (L M, Plate I, Figs. 1 and 2), and apparently are entirely absent at the oesophageal valve, (L M, Plate II, Fig. 5). These muscle fibers are relatively short.

At the base of the intima and lying next to the longitudinal muscles is the hypodermal tissue. In the fore-intestine these cells are relatively small, flattened, brick-like in appearance. Between this tissue and the muscle tissues is the basement membrane, which, however, could not be demonstrated in the fore-intestine, either in longitudinal or cross sections.

The intima, which lines the lumen of the fore-intestine, is secreted by the hypodermal cells and is homologous with the chitin of the body wall. However, it does not appear to be pigmented, with the exception of the oesophageal valve, and stains with fast green. The outer layer of primary intima stains a much deeper color than the inner or secondary intima. However there is no definite line of demarcation between the two layers, the color gradually increasing in intensity as the surface is approached. At the base of the oesophageal valve, the intimal layer, just prior to its end, shows projections of the base of the hypodermal cells into the secondary layer, (HYP, INT, Plate II, Fig. 5). Here the hypodermal cells are elongated and slender, projecting into the open space within the finger-like projection of the valve itself.

In the anterior portion of the oesophagus, the folds of the intima projecting into the lumen, are irregular in size, shape, and number, (INT, Plate I, Fig. 2). However in the gizzard, just anterior to the oesophageal valve, these folds assume a more or less definite and regular arrangement and shape, (INT, Plate I, Fig. 1). This consists of four folds, mushroom-shaped in cross section, of thick, heavy intima. Arising just above the base of these folds are two shorter folds, finger-like in cross section, one on each side. Between the large mushroom-shaped folds is one long finger-like fold.

A longitudinal section of the fore-intestine demonstrates a ringed or ridged characteristic structure of the surface of the intima of the posterior portion of the oesophagus and of the gizzard. From the summits of these ridges, spines of chitinous material project in a curved direction, mesad and caudad, into the lumen. These spines do not stain and are about equal in length to the thickness of the intima. As the oesophageal valve is approached, these spines become rapidly shorter, appearing as tooth-like ridges upon the throat of the valve and rapidly becoming obsolescent posteriorly, (INT, Plate II, Fig. 5). These spines appear also in the hind-intestine, immediately posterior to the entrance of the malpighian tubules into the lumen of the intestine, but they do not

occupy as extended an area, also the ringed character of the intima is entirely lacking in the hind-intestine.

Between the fore and mid-intestines, enclosed within the connective tissue, is a wedge-shaped collar of tissue, (F T, Plate II, Fig. 5). This tissue stains a deep green and the cell walls cannot be demonstrated. It appears filled with small globules of oil and is possibly a fatty tissue. It entirely surrounds the oesophageal valve.

THE MID-INTESTINE

The mid-intestine occupies the greater portion of the abdomen and in its entirety is of uniform appearance, both externally and histologically. Circular and longitudinal muscles may be present, but in very small numbers, and could not be identified in most of the sections. Surrounding the mid-intestine is the connective tissue, which is contiguous with that of both the fore and hind-intestines. The tissues within the connective tissue is chiefly composed of epithelial cells, whose function is the excretion of digestive fluids. This epithelial layer of cells is invaginated to form large folds of epithelial tissue, which project well into the lumen of the mid-intestine. At intervals along the basement membrane are located ovoid, dark-stained masses, which are probably the nidi from which the epithelial cells arise. (C T, EPI, NI, Plate I, Fig. 5; Plate III, Fig. 8). These epithelial cells are formed in definite groups, surrounded by a membrane. Around the inner or mesal edge of the epithelial tissue is another membrane, which is probably the peritrophic membrane or at least functions in that capacity. It follows closely the foldings of the epithelial tissue, and its origin could not be determined.

THE HIND-INTESTINE

At the posterior end of the mid-intestine, just prior to the first entrance of the malpighian tubules, is a layer of intima and hypodermal cells which have a "fluffy" appearance and project slightly into the lumen of the mid-intestine. This is the only indication of a pyloric valve. Here as in the finger-like projections of the oesophageal valve, the hypodermal cells are much elongated and slender, and they have projecting attachments to the secondary intima which stain a very deep green. The basement membrane is in decided evidence, and there are a few fibers of circular muscles present about the valve, (INT, HYP, C M, Plate III, Fig. 8).

The first point of entrance of the malpighian tubules is taken as the point of demarcation between the mid and hind-intestine, which is supported histologically by a decided differentiation between the tissues of each of the parts of the tract. The connective tissue, which is present as a thin membrane, anterior and posterior to this point, becomes much thickened, although definite cell separations and nuclei could not be demonstrated, and separates from the other enclosed tissues to form a separate surrounding envelope about the entrance of the tubules, (C T, Plate III, Fig. 6). Anteriorly from this point, along the tubules, the connective tissue once more becomes intimately associated with the tubules, (C T, Plate III, Fig. 7). Any extension of the intima of the hind-intestine and pyloric valve into the malpighian tubules could not

be shown. The extent and paths of the tubules in the body cavity could not be determined, as they became very brittle in the preserved material and were intertwined with the other tissues in the abdomen of the insect. However, they are evidently very extensive, as the body cavity was quite filled with the tubules.

Immediately posterior to the entrance of the tubules is the hind-intestine. The tissues of this part of the alimentary tract are very similar in appearance and location to those of the fore-intestine, substantiating the evidence that these portions of the alimentary tract are both of ectodermal origin, the chief difference noted being the relationship between the muscle tissues. In the fore-intestine the circular muscles entirely surrounded the longitudinal muscles, whereas in the hind-intestine, the longitudinal muscles are isolated strands of fibers located at more or less regular intervals outside the circular muscles.

The distal ends of the malpighian tubules re-enter the tract at a point about midway along the ileum. They do not penetrate into the lumen but remain just beneath the connective tissue that surrounds the digestive tract. Here they begin a series of convolutions and eventually form a complete sheath about the hind portion of the ileum and the entire colon, and fore-part of the rectum, (M T, Plate III, Fig. 9; Plate IV, Fig. 10). It was not determined whether the tubules divide, however they become much smaller, both the tubes and the cells composing them, and, either the tubes divide, or, they dwindle in size due to the convolutions, which occur in a very short distance, to form a complete layer of tubules about the tract. Just prior to the anus, the tubules end blindly in the space between the outer sheath of connective tissue and the circular muscles.

The longitudinal muscles are very small and few in number, and are located outside the circular muscles at points about equidistant about the digestive tract. They are most numerous just posterior to the pyloric valve, but can scarcely be demonstrated in the rectum. The longitudinal muscle fibers lie in constrictions of the circular muscles in each part of the hind-intestine although in the rectum these constrictions are scarcely evident.

The entire hind-intestine, with the exception of the posterior portion of the rectum, shows a well-developed circular muscle system. These muscles first appear just prior to the first entrance of malpighian tubules, (C M, M T, Plate III, Fig. 8), and surround the pyloric valve. In the forepart of the ileum they are a thick heavy layer of tissue and are "tied-in" at six points to the hypodermal layer. This same "Tieing-in" is apparent throughout the entire hind-intestine, but less noticeably so in the posterior portion of the rectum, (C M, Plate IV, Fig. 11). At each of these points the hypodermal layer and intima are in close association with the muscle layer. As the posterior portion of the ileum is approached, the thickness of the circular muscle tissue decreases notably. This decrease continues until in the colon only two or three very elongate fibers remain, and in the forepart of the rectum, only one fiber surrounds each portion of the tract, (C M, Plate IV, Fig. 10). From this point there is a sudden increase in the fibers to just prior to the anus, where the circular muscle tissue becomes many fibers thick, (C M, Plate IV, Fig. 11).

In contrast to the fore-intestine, the basement membrane in the hind-intestine is very evident its entire length.

The hypodermal layer of tissue is much more in evidence in this portion of the alimentary tract than in the fore-intestine. Here the cells are much larger and nearly square in outline. At the point where the hypodermal cells appear, just prior to the first entrance of the malpighian tubules, (HYP, Plate III, Fig. 8), the cells are much elongated and have dark-staining attachments to the secondary intima, as was noted in the oesophageal valve. In the ileum the cells of this tissue are much smaller and increase in size as the posterior part of the hind-intestine is approached, (HYP, Plate III, Fig. 9; Plate IV, Figs. 10 and 11). As these cells secrete the intima, they are very closely associated with it and lie in folds projecting into the lumen of the tube.

As stated previously, the intima shows the spiny character exhibited in the oesophagus and gizzard of the fore-intestine, but here the area is not as extended, (INT, Plate III, Fig. 8). There is no ringing or striation of the intima in the hind-intestine. The intima at the pyloric valve is of a "fluffy" appearance, which character is maintained for a short distance posteriorly. It is very thick at this point, gradually becoming less so as the narrower part of the ileum, (INT, Plate III, Fig. 9), is approached, and then becoming increasingly thick toward the rectum, (INT, Plate IV, Figs. 10 and 11). In the colon the intima is quite thick, and in the forepart of the rectum lies in pad-like arrangement, (INT, Plate IV, Fig. 10). There are six of these pads, whose delineations, however are shown only by the attachment of the circular muscle fibers and the thinning of the intima at these points. In the posterior portion of the rectum, the intima projects into the lumen in numerous folds, which nearly fill the lumen of the rectum, (INT, Plate IV, Fig. 11.) Anterior to the rectal pads, the folds of the intima nearly fill the lumen of the tube, becoming gradually less elongate until a short distance posterior to the pyloric valve, where they become more nearly as in the anterior portion of the rectum.

SUMMARY

This paper is a study of the morphology and histology of the alimentary tract of the blister beetle, *Epicauta cinerea* Forst., variety *marginata* Fab., collected in southeastern Ohio.

The fore-intestine consists of the oseophagus, gizzard, and oesophageal valve. The latter is highly developed, consisting of four finger-like projections, which extend into the mid-intestine about one-fifth its length.

The mid-intestine lacks any well-developed muscle tissues. The epithelial tissue is invaginated to form folds, which project well into the lumen of the intestine. This tissue possibly forms the peritropic membrane.

Just prior to the entrance of the malpighian tubules, there is a ring of circular muscles. At this point the intima and

hypodermal cells project slightly into the lumen of the tract. This is the only indication of a pyloric valve.

The hind-intestine consists of the ileum, the colon, and the rectum. The circular muscles are well-developed in the ileum, and posterior portion of the rectum. In the colon and anterior portion of the rectum, they consist of only one or two strands. The longitudinal muscles are few in number, and arranged in groups of six, approximately equi-distant about the circular muscles, which exhibit a "tieing-in" at these points.

The malpighian tubules are six in number and enter the tract at two points, just posterior to the pyloric valve, where they penetrate into the lumen of the alimentary tract, and midway of the ileum, where they remain beneath the connective tissue, and through increasing convolutions and decreasing size form a sheath of small tubules about the posterior portion of the hind-intestine.

BIBLIOGRAPHY

Auten, Mary. 1933. The Structure of the Digestive System in *Boliotherus cornutus*. Ohio Journal of Science 33 (4): 280–286, 2 plts. (Good bibliography.)

Becton, Edward M., Jr. 1930. The Alimentary Canal of *Phaenaeus vindex* Macl. (Scarabaeidae). Ohio Journal of Science 30 (5): 315–323, 2 plts.

Bess, Henry A. 1935. The Alimentary Canal of *Calasoma sycophanta* Linnaeus Ohio Journal of Science 35 (1): 54–64, 3 plts.

Bigham, J. T., Jr. 1931. The Alimentary Canal of *Asaphes memonius* Hbst. Ohio Journal of Science 31: 386–395, 2 plts.

Brubaker, R. W. 1934. The Alimentary Canal of *Penthe pimelia* Fabr. (Coleoptera: Dacnidae). Ohio Journal of Science 34 (1): 46–56, 2 plts.

Burgess, Emory D. 1932. A Comparison of the Alimentary Canals of the Active and Hibernating Adults of the Mexican Bean Beetle, *Epilachna corrupta* Muls. Ohio Journal of Science 32 (3): 249–261, 2 figs., 2 plts.

Cecil, Rodney. 1930. The Alimentary Canal of *Philaenus leucophthalmus* L. Ohio Journal of Science 30 (2): 120–130, 2 plts.

Comstock, J. H. 1924. An Introduction to Entomology.

Davidison, Ralph Howard. 1931. The Alimentary Canal of *Criocerus aspargi* Linn. Ohio Journal of Science 31 (5): 396–405, 2 plts.

Dean, R. W. 1932. The Alimentary Canal of the Apple Maggot, *Rhagoletis pomonella* Walsh. (Dipt.: Trypetidae). Annals Entomological Society of America 25 (1): 210–223, plts. I–IV.

Dean, R. W. 1933. The Morphology of the Digestive Tract of the Apple Maggot Fly, *Rhagoletis pomonella* Walsh. Technical Bulletin 215, New York Agricultural Experiment Station, Geneva, N. Y., 3 plts.

Dunham, W. E. 1927. The Alimentary Tract of the Carpenter Bee (*Xylocopa virginica*). Master of Science Thesis, Ohio State University.

Fletcher, Fred Walker. 1930. The Alimentary Canal of *Phyllophaga gracillis* Burm. Ohio Journal of Science 30 (2): 109–119, 2 plts.

Kershaw, J. C. 1914. The Alimentary Canal of a Cercopid. Psyche 21: 65–72, 1 plt.

Imms, S. D. 1929. A General Textbook of Entomology.

Ishimori, Naoto. 1924. Distribution of the Malpighian Vessels in the Wall of the Rectum of Lepidopterous Larvae. Annals Entomological Society of America 17: 75–86, 8 figs., 2 plts.

Jahn, L. A. 1930. The Internal Anatomy of the Mydas Fly. Ohio Journal of Science 30 (2): 85–97, plts. I–III.

Lewis, Harold C. 1926. The Alimentary Canal of Passalus. Ohio Journal of Science 26 (1): 11–24, 2 plts.

Mansour, K. 1927. The Development of the Larval and Adult Midgut of *Calandra oryzae* (Rice Weevil). Quarterly Journal of Microscopical Society of London, New Series, 71: 313–352, 1 fig., plts. 29–33.

Miller, W. C. 1931. The Alimentary Canal of *Mercantha contracta* Beauv. (Tenebrionidae). Ohio Journal of Science 31 (3): 143–156, 2 plts.

Neiswander, C. R. 1925. Additions to our Knowledge of the Anatomy of Ranatra (Heteroptera). Transactions American Entomological Society 51 (879): 311–320, 1 plt.

Potts, S. F. 1927. The Alimentary Canal of the Mexican Bean Beetle. Ohio Journal of Science 27 (3): 127–137, 2 plts.

Schaefer, Paul Everett. 1931. The Alimentary Canal of *Sphaeroderus nitidicollis* Chev. var. *Schaumi* Chd. (Coleoptera). Ohio Journal of Science 31 (5): 406–415, 3 plts.

Swingle, M. C. 1930. Anatomy and Physiology of the Digestive Tract of the Japanese Beetle. Journal of Agricultural Research 41 (3): 181–196, 4 plts.

Talbot, Mary. 1928. The Structure of the Digestive System in *Creophilus villosis* (Grav.) (Staphylinidae). Ohio Journal of Science 28: 261–266, 2 plts.

Whittington, F. B. 1935. The Alimentary Canal of *Harpalus pennsylvanicus* DEG. (Carabidae: Coleoptera). Ohio Journal of Science 35 (2): 131–138, 3 plts.

Woods, William Colcord. 1916. The Malpighian Tubules of *Haltica bimarginata* Say (Coleoptera). Annals Entomological Society of America 9 (4): 391–406, 1 plt.

Woods, William Colcord. 1918. The Alimentary Canal of *Altica bimarginata* Say. Annals Entomological Society of America 11 (3): 283–314, plts. 26–29.

ABBREVIATIONS USED ON PLATES

B M—Basement Membrane.
C M—Circular Muscles.
COL—Colon.
C T—Connective Tissue.
EPI—Epithelial Tissue.
F T—Fat Tissue.
GIZ—Gizzard
HYP—Hypodermal Tissue.
ILL—Ileum.
INT—Intima.
L M—Longitudinal Muscles.
LU—Lumen.
MI—Mid-intestine.
M T—Malpighian Tubules.
NI—Nidus.
OES—Oesophagus.
O V—Oesophageal Valve.
P V—Pyloric Valve.
REC—Rectum.

EXPLANATION OF PLATES

PLATE I

Fig. 1. Cross section through the gizzard.
Fig. 2. Cross section through the oesophagus.
Fig. 3. Dorsal view showing the gross dissection of the alimentary tract.

PLATE II

Fig. 4. Gross drawing of one of the finger-like projections of the oesophageal valve.
Fig. 5. Longitudinal section through the oesophageal valve, showing the posterior portion of the oesophagus, the gizzard, and the anterior portion of the mid-intestine.

PLATE III

Fig. 6. Cross section through a malpighian tubule just prior to first entrance into the alimentary tract at the pyloric valve, showing the surrounding envelope of connective tissue.
Fig. 7. Cross section through a malpighian tubule in the body cavity.
Fig. 8. Longitudinal section through the pyloric valve, showing the posterior portion of the mid-intestine, the entrance of the malpighian tubules into the lumen of the alimentary tract, and the anterior portion of the ileum.
Fig. 9. Cross section through the ileum immediately posterior to the second entrance of the malpighian tubules into the alimentary tract, showing the tubules beneath the connective tissue.

PLATE IV

Fig. 10. Cross section through the anterior portion of the rectum, showing the surrounding envelope of malpighian tubules immediately beneath the connective tissue.
Fig. 11. Cross section through the posterior portion of the rectum.

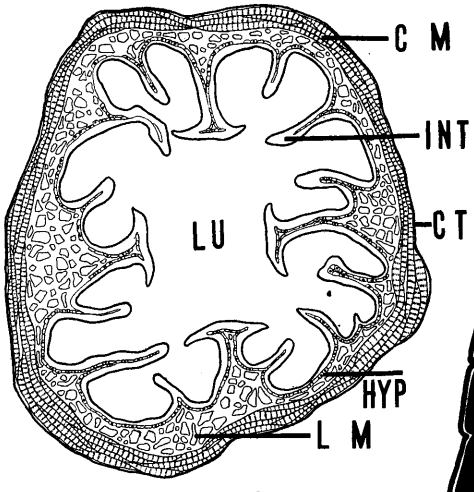

FIG 1

FIG 2

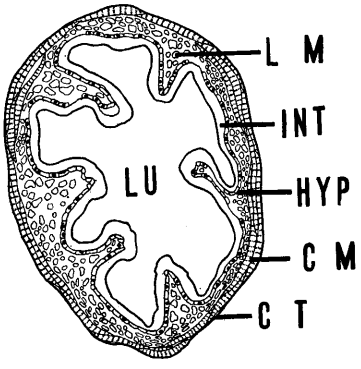

FIG 3

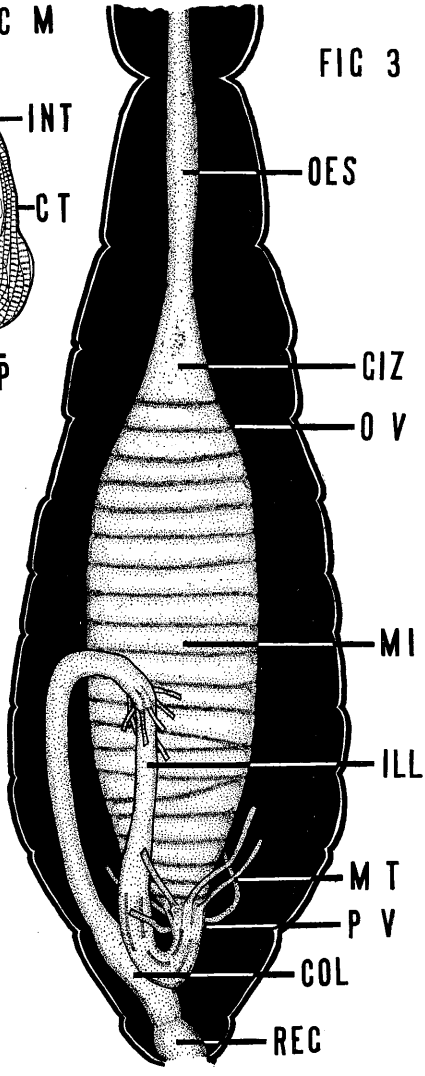

FIG 4

FIG 5

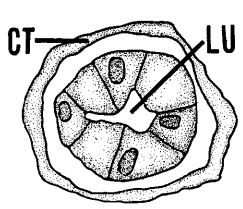

FIG 6

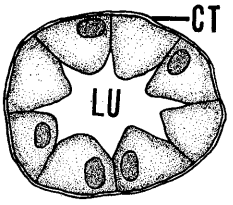

FIG 7

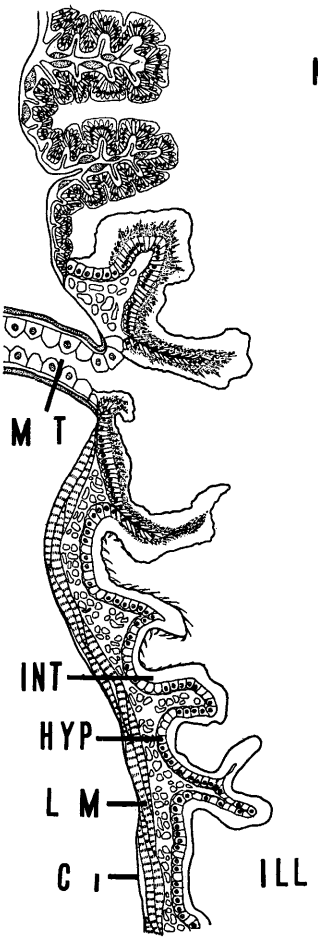

FIG 8

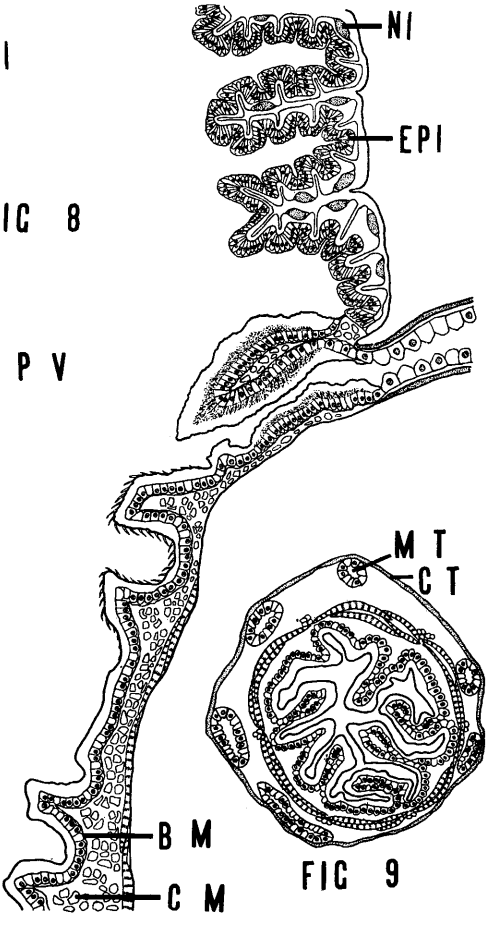

FIG 9

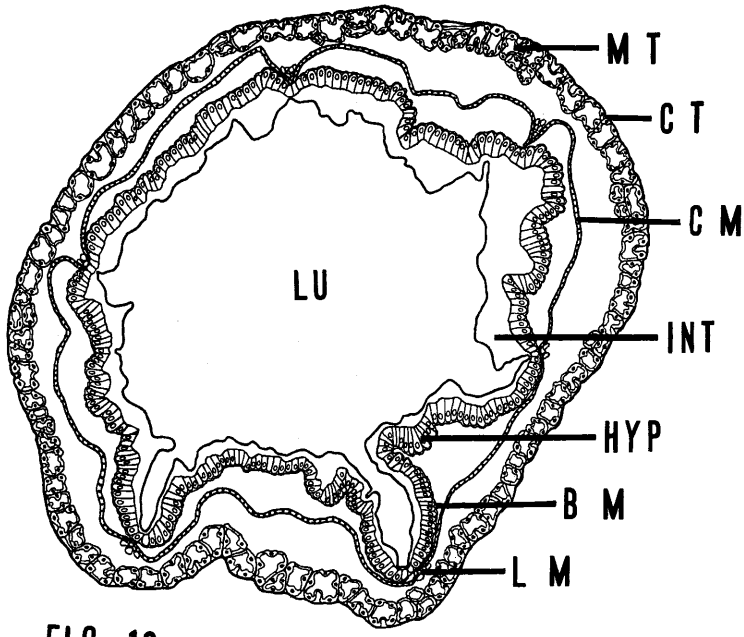

FIG 10

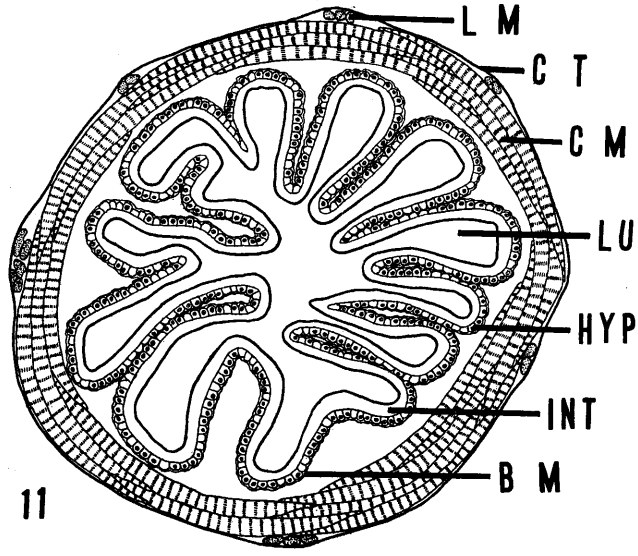

FIG 11