Observations on the Structure of Hydra as seen with the Electron and Light Microscopes

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With 6 plates (figs. 1 to 6)

SUMMARY

Sections of hydra studied with the electron microscope show various structures which have been identified by referring to control histological sections and to previous descriptions. Certain features have also been examined in frozen-dried sections under the light microscope.

In the ectoderm, epithelio-muscular cells contain various organelles, and also smooth longitudinal muscle-fibres with which mitochondria may be associated. The so-called 'supporting fibres' appear to be thin bundles of muscle-fibres. Although points of contact exist between muscle-fibres, there appears to be no cytoplasmic continuity. The muscle-fibres insert on the mesogloea, and appear to be separated from it by two membranes, one belonging to the cytoplasm surrounding the muscle-fibre and the other to the mesogloea.

The mesogloea is extracellular and quite distinct from the intracellular muscle-fibres. It appears granular and sometimes presents an indistinct fibrous background. In frozen-dried material the mesogloea stains blue with Mallory's method, while the muscle-fibres stain red.

Two main types of cells are found in the endoderm. Among these, some of the digestive cells contain transverse muscle-fibres, but they are less distinct than the longitudinal ectodermal fibres. Otherwise the digestive cells vary much in structure, but generally they contain vacuoles and their free surface is thrown into villi covered with small granules. The 'foamy gland cells' are filled with much larger vacuoles containing granular material. The vacuoles are discharged together with portions of cytoplasm, and at this stage lamellated double membranes and mitochondria appear between the vacuoles. Both types of cell possess two flagella, which show a typical ultrastructure and are surrounded by a thick membrane.

Various other cells of the ectoderm are distinguished by their characteristic appearance. Cnidoblasts, for instance, have been found to contain an extensive system of intercommunicating vacuoles bounded by membranes, and do not resemble the interstitial cells. In unexploded penetrant nematocysts the tube is preformed and the butt and stylets can also be seen. The special gland-cells of the pedal disk show large, electron-dense granules which are extruded from the cell without any cytoplasm. A relatively thick homogeneous layer on the surface of the pedal disk is distinguished by the electron microscope.

INTRODUCTION

ALTHOUGH the two-layered anatomy of hydra is well known, the structural units of which it is composed are so small that use of the light microscope has never fully elucidated their arrangement. The far greater resolution afforded by the electron microscope enables some new observations.

to be made on the fine structure of this animal, and provides further information about the nature and distribution of the mesogloea, of the muscle-fibres, and of various kinds of cells composing the ectoderm and endoderm.

Material and Methods

Electron microscopy

A large, colourless variety of *Pelmatohydra oligactis* was employed in the electron microscope study. Animals were fixed for 10 min to 1 h in Dalton’s fixative (Dalton and Felix, 1955), a solution which contains 1% OsO₄, 1% K₂Cr₂O₇ at pH 7.2, and 0.85% NaCl. Pieces of hydra were also fixed in buffered 1% OsO₄ in Ringer’s solution for similar lengths of time. The best results were obtained after short fixation in Dalton’s fluid, and most of the electron micrographs are taken from material treated in this way.

Certain animals were placed directly in the fixative and contracted immediately. These were preserved in a contracted state. Others, which were first anaesthetized with 1/5% chloral hydrate, remained relaxed and were neither greatly contracted nor extended when fixed. Some of the anaesthetized hydra were divided into parts (hypostome, body, pedal disk) before fixing.

After fixation, the specimens were washed in distilled water, placed in 70% alcohol, dehydrated, and then embedded in a partially polymerized mixture of one part of methyl methacrylate to three of butyl methacrylate. This was polymerized at 45°C with benzoyl peroxide as the catalyst. Ultra-thin sections (about 300 Å thick) were cut with a modified Minot rotary microtome equipped with a glass knife, or with the Servall Porter-Blum microtome. Sections were taken in transverse, longitudinal, sagittal, and oblique planes. Most of the observations were made on transverse and longitudinal sections of the body, these being the specimens whose orientation and appearance were simplest to interpret. The sections were mounted on copper mesh grids, which were inserted into a Philips EM 100 or an RCA-EMU type electron microscope, without removing the plastic. Negatives were exposed at a magnification of 1,000 to 6,000 times and enlarged photographically to the desired size. The final magnifications are approximate.

Light microscopy

In order to assist the recognition of structures seen with the electron microscope, control sections were examined regularly under phase contrast. Ordinary histological preparations of white and brown *P. oligactis* were also of help in orientating the electron micrographs.

Frozen-dried sections of a brown variety of *P. oligactis* have been studied with the light microscope. After freezing-drying (see Bell, 1956), specimens were placed in methyl benzoate with 1% celloidin; the normal procedure for double embedding was followed (Pantin, 1948). Section ribbons were flattened over Baker’s formaldehyde-calcium (1944), on which they were left floating overnight at room temperature for post-fixation of the tissues. After two or three changes of distilled water, the ribbons were floated on to albumenized
slides, left to dry overnight, and stained by Mallory's trichrome method (Pantin, 1948). Although post-fixation may cause sections to shrink slightly, they become easier to handle and stain more clearly. It may be noted that freezing-drying, like osmium fixation, does not produce specimens uniformly free from artifact, and that certain regions are much better preserved than others.

RESULTS

It should be borne in mind that all the observations recorded here have been made on fixed material. While the high magnification of the electron microscope allows fine structure to be described far more accurately than is possible with the light microscope, the relation of many structures occurring in fixed material to those found in living hydra has not yet been determined.

The ectoderm

Epithelio-muscular cells. Certain cells of the ectoderm possess large oval nuclei of an even granular texture (fig. 1, A, _nuc_). Their cytoplasm contains a large variety of organelles and inclusions, among which may be found small vacuoles bounded by membranes with apposed granules, structures showing folds extending a short distance into the interior of the organelle and which may be identified as mitochondria of moderate size (fig. 1, A, _mit_), and complex bodies consisting of adjacent smooth membranes, vesicles, and vacuoles. There are several other inclusions, usually seen as electron-dense bodies, such as, for example, a frequently occurring ellipsoidal structure which is distinguished from mitochondria by being smaller and much denser, and without internal folds (fig. 5, F, _org_). Another body, although only moderately dense, is also darker than the surrounding cytoplasm (fig. 1, A, _org_), and is round in outline with a folded or crumpled membrane. Large and small granules, within which further structure may be detected, are seen at the centre. This type of inclusion or organelle, while varying slightly, often occurs in endodermal digestive cells as well (p. 322). A granular material adhering to the outer surface of the ectodermal cells forms a thin coat over the hydra (figs. 1, A; 5, F, _surf_).

Within some of the cells may be seen closely packed bundles of fine fibrils arranged in parallel and running in the general direction of the column axis (fig. 1, A, _mf_). The fibrils are usually accumulated at the base of the cell (compare fig. 2, C, _ect_), but as in fig. 1, A, some may extend upwards above the nucleus. The appearance and disposition of the bundles of fibrils indicate that they correspond to the myofibrils of histologists (see von Gelei, 1924), and the cells in which they are found are therefore epithelio-muscular cells. A detailed consideration of the structure and orientation of these intracellular fibrils further confirms their identity (see p. 322).

Several authors have reported the presence also of supporting or skeletal fibres in the epithelio-muscular cells of hydra, especially in the region of the foot and gonads. They are believed to provide stiffness and elasticity, although
Mueller (1950) has suggested that some of them are in fact muscular. Now the 'supporting fibres' figured by von Gelei (1924) pass from the cell-body into the basal extensions of the epithelio-muscular cell and run alongside the muscle-fibres (compare fig. 6, b). They would thus correspond closely in arrangement to the fibrils which extend into the ectodermal cells as just described. We find that the fine structure of these fibrils is identical with that of the myofibrils at the base of the epithelio-muscular cells. 'Supporting fibres' have not usually been described as being present in the endodermal cells, and correspondingly we find that myofibrils in the endoderm are restricted to the base of the cells. Our observations, therefore, tend to confirm Mueller's suggestion that many 'supporting fibres' form part of the muscular system.

Mueller (1950) agrees with von Gelei (1924) and other authors, however, in believing that a distinct class of intracellular supporting fibres does exist, but our observations so far provide no evidence for this. It must be remembered that other investigators have used different methods from those in the present study, and that under the light microscope fine fibrillar structures are near the limit of resolution and difficult to interpret with certainty.

**Cnidoblasts and developing nematocysts.** Another kind of ectodermal cell occurs in groups, which are sometimes found next to the mesogloea. Each cell contains a large round body which is limited by a relatively thick homogeneous border, and shows dark particles scattered throughout the interior (fig. 4, A, nem). In other cases, these organelles may be even larger, and sections reveal that they are undoubtedly developing nematocysts. The structures in fig. 4, A are therefore immature nematocysts at various stages of development, and the cells containing them are cnidoblasts.

The nucleus of each cell may be displaced by the developing nematocyst. Within the nucleus a very dense organelle showing small clear patches represents the nucleolus. It apparently does not have a thread-like or vesicular structure and is usually found towards the centre of the nucleus (fig. 4, A, n, nuc). Other electron-dense particles are also seen, scattered irregularly within the nuclear membrane.

In these cells numerous vacuoles of a moderate size are dispersed throughout the cytoplasm and intercommunicate with each other, often giving the appearance of channels. The vacuoles generally appear to be empty and are bounded by membranes. At high magnification, numerous fine granules are seen in and around the membranes, and the cytoplasm between two membranes may also appear granular. In some sections the cytoplasm immediately surrounding a nematocyst rudiment is particularly dense and shows a lamellated structure (fig. 4, A, lam). Bodies are also present whose internal folds and structure resemble those of the mitochondria seen in other cells, and they may therefore be identified as such (fig. 4, A, mit).

Although only one nematocyst occurs in each cnidoblast, sections of a group of developing cells show that the cytoplasm of adjacent cnidoblasts is apparently continuous at certain points along their borders (fig. 4, A, con). If this
appearance is not due to fixation artifact, it may indicate that cells in a group of cnidoblasts are in syncytial relation to each other. Such a syncytial arrangement of cnidoblasts containing immature nematocysts might ensure that the batteries of nematocysts in the tentacles developed in a co-ordinated manner. The orderly pattern of mature nematocyst types has been studied by von Gelei (1927) and Semal van Gansen (1951).

**Nematocysts.** An electron microscope study of hydra nematocysts has been made by Semal (1954a), who used whole mounts of discharged capsules. Four kinds of nematocysts are found. The parts characteristic of the stenotele, or penetrant type, are the capsule, leading into the shaft or basal portion of the butt and carrying projecting stylets, the distal part of the butt or conical piece, which may sometimes carry spines, and the elongated filament or tube, which extends from the conical piece and may either carry rows of spines (Hyman, 1940) or be devoid of armature (Semal, 1954a).

In sections through the capsule of a mature undischarged stenotele within its cnidoblast (fig. 3, B), the coiled and elongated filament (t) is found embedded in an electron-dense homogeneous matrix, which is presumably fluid coagulated within the nematocyst capsule. The filament is a narrow tube with homogeneous walls and a central lumen: it appears to be empty and to lack spines. The conical part of the butt is seen in figs. 3, B (b) and 3, D as two serrated structures on whose inner sides numerous dark overlapping spines are attached. Above these in fig. 3, B are the large stylets (s), within the basal part of the butt. The attachment of the butt to the capsule wall cannot be seen in these sections, but in fig. 3, D the distal end of the butt extends as the beginning of the nematocyst thread, which has been cut through.

Sections through capsules of another type of nematocyst also show structures composed of overlapping layers of electron-dense material (fig. 3, C, sp). They apparently represent sections through the barbed thread of a holo-trichous isorhiza or large glutinant nematocyst. These barbs appear similar in structure to the spines on the butt of stenoteles.

The appearance of the tube in sections of undischarged nematocyst capsules shows that the filament is already present before the nematocyst discharges and that it is not formed by extrusion of fluid or magma as suggested by Kepner and his colleagues (1951).

**Interstitial cells.** Groups of small, rounded cells also occur in the ectoderm (fig. 1, c). They are numerous in some areas and absent from others, and appear to be the interstitial cells.

The nucleoplasm of these cells is very light, but it contains granules and an extensive network of dark substance, which may represent nucleolar material. In some nuclei this dense network may surround islands of light nucleoplasm. The cytoplasm is not very dense, and contains granules, mitochondria with internal folds, and round vacuoles of varying sizes (fig. 1, c, vac) to which granules may be apposed. In some sections a complex of smooth membranes and vacuoles is seen.

If the interstitial cells give rise to cnidoblasts (fig. 4, A; see Hyman, 1940),
they must undergo remarkable changes in organization, especially in the case of the nucleolus and of the cytoplasmic vacuoles.

Gland-cells. Groups of secretory cells also occur in the ectoderm of the foot region. Large vacuoles, each of which usually contains a dense ellipsoidal granule, are found towards the periphery of each cell (fig. 5, A, vac, g). There is considerable space between a granule and its vacuole, probably due to shrinkage during preparation of the tissue. The cytoplasm between vacuoles contains mitochondria. The outermost cell membrane is thick and finely granular, and is covered by an external layer of dark amorphous material (fig. 5, A, al).

These cells are apparently ectodermal gland cells of the pedal disk, which are the only kind found in that region and are characteristically filled with coarse granules (Hyman, 1940). Their appearance in frozen-dried material is seen in fig. 6, E, which shows part of a vertical section of the foot (compare fig. 5, A). The peripheral granules are seen again in tangential section in fig. 6, F. The amorphous substance seen in the electron micrographs is probably the adhesive material which cements hydra to the substratum and which is secreted by these gland-cells.

In some electron microscope sections the secretory granules are found outside the foot of the hydra as if they had just been extruded (fig. 5, A): they may rest in a depression in the outer cell-membrane or in the amorphous layer, and give the impression that they have just passed out of the cells and are still clinging to the surface. The extrusion of granules may nevertheless be an artifact, as it is possible that they were forced out of the cell mechanically during preparation of the tissue.

The mesogloea

The mesogloea, which lies between the ectoderm and endoderm (figs. 2, A, B, C; 3, A; 5, E, mes), is non-cellular and usually appears light, although in some sections it is fairly dark. It is usually granular, and the granules may be close together or fairly scattered. In some preparations they seem to be resting on a background of extremely fine longitudinally orientated threads, and it is possible that some of the granules in the mesogloea represent cut fibrils. Other oblique sections reveal very fine threads which appear to be present throughout the mesogloea and give it a fibrillar texture (fig. 5, E, mes).

In frozen-dried preparations also the mesogloea sometimes appears to be fibrous in nature, although when poorly fixed it appears reticulate. If true fibres are present, this is a further likeness between the mesogloea of hydra and that of other coelenterates (see Chapman, 1953). The varying appearance of the mesogloea probably depends on the state of contraction in which the hydra is fixed.

The mesogloea appears to be bounded by two basement membranes, which separate it from the ectoderm and endoderm respectively. These membranes are not always easy to discern but may be seen in fig. 2, c (doub). They will be discussed further in considering the relation of the muscle-fibres and meso-
gloea (p. 324). The mesogloea also shows holes filled with a very light substance (figs. 2, A, c). As will be explained below, these represent oblique sections of cytoplasmic roots, which are extensions of the cytoplasm surrounding the muscle-fibres and penetrate the mesogloea (Semal, 1952).

The endoderm

The endodermal cells of hydra vary in structure and appearance according to the region of the animal as well as to the time since the last meal. Two main types of cell are concerned with the digestion and absorption of food: the gland cell, which pours secretion into the lumen to break down the food and makes its products available for intracellular and perhaps extracellular digestion, and the digestive cell, which absorbs the food products. Both kinds of cell have been observed with the electron microscope.

Gland-cells. Some of the endodermal cells are almost completely filled with large vacuoles, and can be identified as ‘cellules spumeuses’ or foamy cells (fig. 4, c). Semal (1954b) describes them as the most numerous kind of gland-cell, filled with polygonal vacuoles of finely granular content. Their appearances in the light and electron microscopes are very similar and they are thus relatively easy to identify.

Electron micrographs of foamy cells show that the vacuoles appear to be filled with granules, or with a thread-like material, or a mixture of both (fig. 4, c, vac), according to their plane of section. At the base of the cell the vacuoles are smaller than those at the periphery and contain a denser granular material. Small mitochondria with internal folds occur infrequently in the tenuous cytoplasm between the peripheral vacuoles, but they become more numerous towards the base of the cell.

Semal (1954b) has described the way in which foamy cells discharge at the moment the hydra ingests prey, and then undergo a recharging cycle. Fig. 4, c appears to represent part of a discharging cell. Its vacuoles are similar to those of a resting cell, but they may have altered in size as there seems to be more cytoplasm between them. The mitochondria, of moderate size and with short internal folds, are now easier to discern (fig. 4, c, mit). Structures consisting of overlapping or lamellated double membranes may also occur between the vacuoles of a discharging cell, either alone or near mitochondria (fig. 4, c, lam).

The discharged material of the foamy gland cells consists not only of vacuoles and their contents but also of portions of the cytoplasm with its mitochondria and lamellated membranes (fig. 4, c). This type of secretion differs from that of the ectodermal gland-cells of the foot, where only secretory granules formed in the cytoplasmic vacuoles leave the cell.

We have not yet observed the presence of muscle-fibres in these cells, nor seen their attachment to the mesogloea.

Digestive cells. Certain other endodermal cells are more variable in structure and probably represent the digestive cells. These are known to undergo profound changes in shape and organization during the digestion of a meal (Semal, 1954b). Fig. 4, b shows part of one of these cells filled with relatively
small vacuoles (vac): since digestive cells become vacuolated at the beginning of a meal (Semal, 1954b), the state of this cell probably corresponds to an early stage in the digestive process. The peripheral cytoplasm extends into the lumen of the gut as numerous villous processes (fig. 4, B, vp), which are covered with external granules, and are a constant feature of these cells during digestion. In addition the digestive cells, like the foamy gland-cells, each possess two flagella (figs. 1, B; 4, B; 5, B; flag), as described in the next section. Myofibrils are found in the basal cytoplasm of some digestive cells and are considered further on p. 323.

Generally speaking, the cytoplasm of the digestive cells is light (fig. 1, B) and contains vacuoles of varying sizes, with which granules and mitochondria may be associated. Several other organelles or inclusions may be present. Of these, a round body, whose folded membrane encloses a fairly light substance with complex internal granules, much resembles a similar structure found in ectodermal cells (p. 317, fig. 1, A, org).

There is again a complex of membranes associated with vacuoles. Several kinds of inclusions in large vacuoles, smaller vacuoles associated with granules, and dark bodies of variable size whose density is approximately that of lipid material, are among the structures which probably represent stages in the digestion of food products. Similar inclusions have been described by Semal (1954b) and they vary according to the time after a meal and the kind of food ingested.

Flagella. The endodermal cells possess flagella which have been said to number from one to five, with two the most frequent estimate (McConnell, 1931; Mueller, 1950; Semal, 1954b). In our preparations the flagella of both foamy and digestive cells constantly occur in groups of two (figs. 1, B; 5, B, flag). Sections of the flagella show the nine peripheral and two central longitudinal filaments which characterize the cilia and flagella of other animals (Fawcett and Porter, 1954). In addition there is a structureless, relatively thick surrounding membrane, which appears to be closely connected to the flagellum but often becomes detached (figs. 1, B; 4, B; 5, B, c). This thick membrane differentiates the flagella of hydra from those of associated organisms. The numerous cilia shown in fig. 5, D, for example, belong to a protozoon situated on a hydra, and lack the thick sheath which surrounds each hydra flagellum. Part of the protozoon is included in the section.

The muscular system

Since the organization of the muscular system in hydra is as yet incompletely known and it is of considerable functional importance, relevant observations made in this study are discussed together below.

Ectodermal muscle. It has been seen that longitudinal sections of the ectoderm show closely packed bundles of fine fibrils, which are orientated longitudinally at the base of the cells and rest on the mesogloea (p. 317; fig. 1, A). The fibrils are spaced evenly and are not cross-striated. Their bundles form a single-layered network extending over most of the animal. In slightly oblique
transverse sections the bundles of fibrils are seen to form a parallel series above the mesogloea (fig. 2, c, ect). Since the majority of observers find that the ectodermal muscle-fibres of hydra are also longitudinally orientated, and that they occupy the same positions as the bundles of fibrils we have observed, there is strong indication that the two are identical.

The myofibrils are thus found within epithelio-muscular cells, and they extend right into the long cytoplasmic processes forming the base of each cell (see fig. 6, b). Longitudinal sections sometimes show the muscle-fibres surrounded by basal cytoplasm (fig. 2, a, ect, bas), but more often the cytoplasm is so thin that the muscle-fibres seem to rest directly on the limiting membrane of the mesogloea (fig. 3, a, ect). It is interesting that the muscle-fibres are not separated from the cytoplasm by a special membrane, although when the cytoplasm in the basal extensions of the cell becomes very thin, the cell-membrane may appear to belong to the muscle-fibre (fig. 3, a). Mitochondria are often found just outside the myofibrils, and they may then bulge from the surface of the muscle-fibre (figs. 2, a, b; 3, a, mit).

The muscle-fibres are closely applied to the mesogloea and seem either to push into it themselves (figs. 2, b, c, ect, mes), or to be anchored by small pseudopodial processes which penetrate the mesogloea (see mes in figs. 2, a, b, c; 3 a). These appear to be cytoplasmic attachment roots provided by the bases of the epithelio-muscular cells, as described by Semal (1952). Longitudinal sections also show points of contact between the muscle-fibres (fig. 2, b, j). Although the membranes of adjacent fibres are intimately connected they remain distinct and the sarcoplasm and fibrils of neighbouring cells do not fuse.

Endodermal muscle. The bases of digestive cells resting on the mesogloea also contain very fine, closely packed fibrils (figs. 2, a, b; 3, a, end). They are more difficult to discern than the ectodermal myofibrils owing perhaps to the particular state of contraction of the fixed hydra, but their position and structure indicate that they are of a similar nature and they therefore represent endodermal myofibrils. They are orientated at right angles to those of the ectoderm and form a transverse muscular layer: thus when the ectodermal muscle is cut longitudinally, endodermal muscle is cut in cross-section, and conversely.

All the digestive cells which contain muscle-fibres appear to rest upon the mesogloea. As in the ectoderm, the muscle-containing bases of the cells are in contact with each other, but their respective membranes remain independent, as do their sarcoplasm and fibrils. There is little cytoplasm surrounding the muscle-fibres, however, as they are usually close together and almost fill the base of the cell. While the muscle-fibres are closely connected to the mesogloea, they are smaller and penetrate it less deeply than those of the ectoderm, and cytoplasmic roots of attachment are fewer and less robust than those of the epithelio-muscular cells.

The relation of muscle-fibres to the mesogloea. Frozen-dried sections of anaesthetized hydra stained by Mallory’s trichrome method show structures which have been seen by previous authors and also during the present electron
microscope work. The muscle-fibres in particular are very clear. They stain red, as do the muscle-fibres of other coelenterates when coloured by Mallory’s method. Transverse sections show that the fibres, situated within the cytoplasm of the epithelial cells, occupy the positions indicated by electron micrographs. Fig. 6, a illustrates this feature in the ectoderm of a contracted specimen in which the muscle-fibres are especially evident. Their position within a single musculo-epithelial cell may be confirmed by comparing this photograph with fig. 6, b, showing an ectodermal cell obtained by the Hertwigs’ osmic-acetic maceration technique (1879). The muscle-fibre lies in the epithelial cytoplasm at the base of the cell, just above the mesogloea, as all recent workers using maceration methods have been able to demonstrate (Goodrich, 1942; Mueller, 1950; Semal, 1952). This has been shown in the electron micrographs (figs. 2, a, b, c; 3 a), as described above.

In contrast to the red muscle-fibres the mesogloea stains blue with Mallory’s method, and in this respect it resembles the mesogloea of anemones and medusae (Chapman, 1953). The different staining properties of muscle-fibres and mesogloea are illustrated by figs. 6, c, d, which show the same Mallory preparation photographed through blue and red filters. Fig. 6, d shows that the mesogloea forms a distinct layer between the ectoderm and endoderm (see also figs. 2, a, b, c; 3, a; 5, e). It varies in thickness with the state of contraction of the hydra and is best developed in the foot region of this species (Pelmatohydra oligactis); Holmes (1950) found a similar distribution in Chlorhydra viridissima.

As seen above (p. 323), the electron micrographs show that the muscle-fibres are intimately inserted on to the mesogloea, but that they are separated from it by membranes. Oblique sections in particular show that at least two membranes are present: as well as a membrane limiting the cytoplasm which surrounds each muscle-fibre (p. 323), there is a less clearly defined basement membrane belonging to the mesogloea (fig. 2, c, doub). It has long been known, however, that the muscle-fibres on each side of the mesogloea form two systems of open networks, which are orientated longitudinally, in the ectodermal plane, and transversely, in the endoderm (Mueller, 1950). Between adjacent muscle-fibres, undifferentiated epithelial cytoplasm is therefore in contact with the mesogloea, and other types of cell such as cnidoblasts, or digestive cells without muscle-fibres, may also occupy this position.

It has been suggested by Holmes (1950) that the muscle-fibres represent specializations of the mesogloeaal substance and he refers to them as ‘mesogloeaal fibres’. The evidence which has been presented above makes it unlikely that the muscular system of hydra originates from a fluid mesogloea by ‘crystallizing out’ along lines of force, as has been claimed by Holmes. All types of preparation we have studied show that the muscle-fibres occur within epitheliomuscular cells or digestive cells, and that they belong to these cells and not to the mesogloea. It is difficult also to reconcile Holmes’s theory with the observation that distinct membranes are present between the muscle-fibres and mesogloea.
Holmes's explanation (1950) of the organization of the muscular system of hydra, therefore, seems untenable. The problems which it raises, however, as to the nature and origin of the muscle-fibres and mesogloea still remain unsolved. It is to be hoped that future work with the electron microscope will be able to throw light on the much-discussed functional morphology of this small animal.

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REFERENCES

— 1952. Ibid., 38, 642.
— 1954b. Ibid., 85, 217.

EXPLANATION OF FIGURES

All the electron micrographs show sections of hydra, and in these the line represents 1 μ. Magnifications are approximate.

Fig. 1 (plate). A, section through the ectoderm, showing two epithelio-muscular cells and portions of other cells. The ectodermal surface is to the left.
B, section through the endoderm showing the lumen lined by digestive cells, and flagella in cross-section. Each flagellum is surrounded by a membranous sheath, seen most clearly in the middle pair.
C, section through part of a group of interstitial cells in the ectoderm.
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Abbreviations: dig, digestive cell; flag, flagella; mf, myofibrils; mit, mitochondria; nuc, nucleus; org, organelle or inclusion; surf, body surface of hydra; vac, vacuole; x, artifacts in negative.

Fig. 2 (plate). A, B, longitudinal sections through ectodermal muscle-fibres, mesogloea, and endodermal digestive cells with muscle-fibres in their bases.

Abbreviations: bas, basal cytoplasm of epithelio-muscular cell; doub, double membrane between muscle-fibre and mesogloea; dig, digestive cell; ect, ectodermal muscle; end, endodermal muscle; j, junction of muscle fibres; mes, mesogloea.

Fig. 3 (plate). A, longitudinal section through ectodermal muscle-fibre, mesogloea, and endodermal digestive cells with muscle-fibres in their bases. The large space above the longitudinal muscle-fibre is an artifact produced by the tearing away of the muscle from the ectoderm.

b, section through an undischarged stenotele.

c, section through portion of an undischarged holotrichous isorhiza, showing the overlapping layers of electron-dense spines.

d, section through part of an undischarged stenotele, showing overlapping layers of electron-dense bars within the serrated outline of the butt wall.

Abbreviations: b, butt of stenotele; dig, digestive cell; ect, ectodermal muscle; end, endodermal muscle; mes, mesogloea; mit, mitochondrion; oc, outer capsule of nematocyst; s, stylet of stenotele; sp, spines of holotrichous isorhiza; t, tube of stenotele.

Fig. 4 (plate). A, section through the ectoderm showing a group of cnidoblasts with immature nematocysts.

b, section through part of a digestive cell at an early stage in the digestive process. The flagellum is surrounded by a closely applied membranous sheath.

c, section through portion of a discharging foamy gland-cell. Part of the same cell is seen in the bottom left-hand corner of the previous photograph, at higher magnification.

Abbreviations: al, amorphous layer; con, regions of apparent cytoplasmic continuity between adjacent cells; flag, flagella; lam, lamellated membranes; mit, mitochondrion; nuc, nucleus; org, organelle or inclusion; sh, surrounding membrane of flagellum; surf, body surface of hydra; vac, vacuole; vp, villous projection.

Fig. 5 (plate). A, section through ectodermal gland-cells of the pedal disk.

b, section through endoderm showing paired flagella belonging to a foamy gland-cell and to a digestive cell.

c, longitudinal section of a flagellum from an endodermal gland-cell.

d, section through part of a ciliate protozoan found attached to hydra.

e, oblique section showing fibrous mesogloea between ectoderm and endoderm.

f, peripheral part of two ectodermal cells to show the surface.

Abbreviations: al, amorphous layer; ect, ectoderm; end, endoderm; flag, flagella; g, granules; mes, mesogloea; org, organelle or inclusion; sh, surrounding membrane of flagellum; surf, body surface of hydra; vac, vacuole.

Fig. 6 (plate). Light microscope photographs of Pelmatohydra oligactis material. All except B are of frozen-dried sections, treated with Baker's formaldehyde-calcium and stained with Mallory. All are of frozen-dried sections, treated with Baker's formaldehyde-calcium and stained with Mallory.

A, transverse section showing the row of longitudinal muscle-fibres in the body-wall. They lie within the ectoderm cells and above the mesogloea. The lower half of the figure contains endoderm.

b, musculo-epithelial cell from the ectoderm (osmic-acetic maceration / picrocarmine), showing a straplike muscle-fibril within the basal cytoplasm. Note the large nucleus.

c, transverse section of the body-wall, with ectoderm above and endoderm below. The photograph is taken with a blue filter (Wratten C 6 +H) to bring out structures which have stained red. The longitudinal and circular muscle-fibres show darkly on each side of the mesogloea, which is pale.

d, the same section, photographed with a red filter (Wratten A). The mesogloea is now dark since it is stained blue, and the muscle-fibres do not show at all.

e, vertical section through the foot, showing the ectodermal gland-cells. Note distal granules, compact cytoplasm, and basal muscle-fibres.

f, tangential section through distal part of foot ectoderm, showing the distribution of secretory granules.
FIG. 1

A. HESS, A. I. COHEN, and E. A. ROBSON
FIG. 2

A. HESS, A. I. COHEN, and E. A. ROBSON
FIG. 3

A. HESS, A. I. COHEN, and E. A. ROBSON
FIG. 4

A. HESS, A. I. COHEN, and E. A. ROBSON
FIG. 5

A. HESS, A. I. COHEN, and E. A. ROBSON
FIG. 6

A. HESS, A. I. COHEN, and E. A. ROBSON