The Embryology of Patella [1]

by

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I desire to express my thanks to Prof. Claus for the personal kindness and generosity he has shown in placing at the disposal of one who had no claims of nationality or a previous personal acquaintance, the admirable opportunities for original investigation afforded by the Government Zoological Station at Trieste.

I. Preparation of the Embryos.

The ova are 0,12 mm. in diameter, of a bluish green color and perfectly opaque. A preparation of acetic acid and glycerine rendered them sufficiently transparent for a study of the general external characters, while it was found necessary to resort to artificial sections in order to elucidate the more complicated internal changes.

A few drops of concentrated acetic acid were added to a watch glass full of sea water into which the embryos were transferred; if the solution is of the proper strength, which can easily be learned by experience, the color will change to a semi-transparent, dirty yellow, which in turn disappears entirely upon the addition of glycerine, leaving the embryo colorless with well marked nuclei and cell boundaries.

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[1]) During the preparation of this paper only a part of the literature I desired to consult was accessable to me; I have, therefore simply recorded my observations without any attempt to make comparisons with or criticisms upon the works of a similar nature to my own. Theoretical considerations have also been excluded partly for the same reason and partly because I desire to confirm my opinions by further study.

A very small amount of acetic acid is sufficient to kill the embryos instantly; they however retain theircolor and unless placed in glycerine at once, dissolve or fly to pieces in a very remarkable manner. If the acetic acid is too strong the ova become too transparent and the cells lose their sharp outlines. One drawback to the use of acetic acid is that it is liable to lead one into error unless supplementary studies are made upon material which has been prepared in other ways. The cilia of the velum for example are well preserved by acetic acid and thus one is led to suppose that if there were other cilia present they would be equally well perserved, but that is not the case. For the bunch of large and prominent cilia situated at the animal pole, and the long stiff hairs of the anal cells disappear like a flash on the addition of acetic acid and not a trace of them is to be observed upon the otherwise well preserved embryos. These large cilia may be studied in the living embryos or by examining preparations made in osmic acid and glycerine, as in these media all the cilia are well preserved.

The sections represented in the plates were all drawn with the aid of the camera and cut from embryos killed in acetic acid, preserved for some time in alcohol and afterwards stained either in alcoholic borax carmine or Kleinenberg's haematoxylin.

A great many embryos were preserved in osmic acid and others in sublimate, or a mixture of sublimate and picric acid. In the last two cases it was difficult to make the embryos transparent by means of acetic acid and glycerine, although this could be done quite easily when the embryos had been originally killed in acetic acid.

No sections were made of embryos prepared either in osmic acid or sublimate, as those killed in acetic acid could be stained and sectioned with entire satisfaction.

II. Fecundation of the ova.

After many unsuccessful attempts to obtain the ova of Haliotis, Fissurella and Patella, during which my attention was called to the absence of any external sexual organs or albuminous gland to furnish envelopes for the ova, I came to the conclusion that the ova were scattered about singly in the water and there underwent an external fecundation.

The spawning season of Haliotis and Fissurella had passed; I determined to try artifical impregnation on Patella and was delighted to obtain a large number of fecundated eggs at the first attempt. It was not until the first of Nov. that the ova were sufficiently matured to allow its successful performance. The spawning season continued from that time until the middle of January. So far as I know this is the first instance of artificial fecundation[1]) having been successfully tried upon any of the Gasteropods and it was considered of sufficient importance to publish a notice of the same in the „Zoologischer Anzeiger", VIII. Jahrg. April 27th. p. 236.

Unfortunately I was seldom able to keep the larvae alive longer than ten or twelve days, at the end of which time all the thousands that at first were apparently healthy became so abnormal and dwarfed that it was impossible to use them for study. Only once did I succed in obtaining a few embryos with a normally developed nautiloid shell. Pl. V, Fig. 66. Others which had been kept twice as long, or fourteen days, developed no shell at all although the conditions were apparently the same in both cases.

Various unsuccessful attempts were made to obtain larvae by fishing from the surface.

The mature ova of Patella when taken from the ovary are of a bluish green color and measure 0,12 mm. in diameter. They are protected by a very thick transparent chorion whose outer surface is covered with a number of shallow indentations between which is a much greater number of very small dots, which when highly magnified are also seen to be indentations, but smaller and more thickly distributed than the former. The bottom of each of these small pits is continuous with a fine line or canal (?) extending radially from the outer to the inner surface of the chorion. In profile the outer surface has the appearance represented in Pl. I,

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[1]) I believe the ova of Haliotis and Fissurella could also be artificially, fecundated, as the same conditions prevail in them as in Patella, but the season had passed before it occurred to me to try it. It was not until after my experiments upon Patella were completed that I learned indirectly through v. Ihering's paper of W. H. Dall's observations on the „Extrusion of the Sexual Products of Patella" by means of a fusion and subsequent rupture of the walls of the right organ of Bojanus and that of the ovary or testis, during the spawning season, thus affording free communication between the ovary or testis and the outer world.

Fig. 1, while the inner surface is smooth. [1]) The larger pits are simply indentations in the outer surface of the chorion and are not connected with or extended into fine lines or tubes. None of these dots nor the fine lines connected with them have anything in the nature of a micropyle, which is situated at the animal pole and consists of a funnel-shaped projection of the chorion with a large irregular opening at its summit. Within this opening were a number of highly refractive globules, which greatly interfered with the observation of the fecundation and formation of the pole globules.

About ten minutes after removing the ova from the ovaries the pole globules appear as two colorless and transparent prolongations arising from the surface of the ova at the bottom of the funnel shaped micropyle, and extending upward towards its opening. Pl. 1, Fig. 4. They are of enormous size and when living appear structureless, but upon the addition of acetic acid take on the turbid appearance characteristic of protoplasm when treated with this reagent. At first they are of the same shape, long and irregular, Pl. I, Fig. 4; but finally one becomes much larger than the other, its distal extremity increases in size and at the same time assumes a globular form, while the proximal part is reduced to a slender neck. Upon treatment with glycerine and acetic acid a small indistinctly marked nucleus surrounded by radiating protoplasm may be observed in the centre of the swollen end of the pole globule. Pl. I, Fig. 6. The other pole globule at first approximately straight and club shaped, Pl. I, Fig. 4, finally becomes very much curved and twisted; no nuclei could be seen in it. Two pole globules were usually present, but in abnormal cases I have observed as many as four or five, only one of which however had the peculiar globular extremity, while the remaining ones were elongated and more or less curved. By the end of an hour the necks of both pole globules were very much reduced in size and finally became so slender, that a very slight jar was sufficient to break their connection with the ova.

I had often seen these detached polar globules without knowing what they were, and remember distinctly having seen several in division, although unfortunately at the time I failed to make drawings of them.

[1]) On one or two occasions I have observed a very thin, transparent and structureless membrane surrounding the chorion, from which it was separated by a large space. Whether it was always present forming a second chorion and only to be seen when thus distended I am unable to say.

Sections of the ova at this period often showed the presence of a layer of finely granulated protoplasm at the animal pole from which the pole globule arose.

At times deep depressions would be formed in this region to the bottom of which the pole globules were attached. Pl. I, Fig. 5.

After the extremities of the pole globule had fallen off, a small portion still remained attached to the ova, by means of which one was able to distinguish the animal pole as late as the stage with eight segmentation spheres,

III. Segmentation.

Segmentation takes place according to the general Molluscan type and in its earlier stages very much resembles that of Planorbis as shown by Rabl.

The first division is meridional, and in the majority of cases divides the ova into two unequal parts, Pl. I, Fig. 9, although I have often observed this stage when it was impossible to detect any difference in the two products.

The next division is also meridional and at right angles to that of the first, only affecting however the larger of the two segmentation spheres, which it divides into two equal parts. Pl. I, Fig. 10.

The division of the remaining first segmentation sphere takes place some time afterwards, but in the same plane as the second, giving rise to the stage with four segmentation spheres. Pl. I, Fig. 11.

The third segmentation, counting that of the two primary spheres as the second, although the division is not simultaneous, is parallel to the equator of the ova and a little nearer the animal than the vegetative pole. Instead however of dividing all four spheres simultaneously, it acts upon them successively, producing stages with five, six, seven and eight segmentation spheres. Pl. I, Fig. 12, 13 and 14. [1])

The stage with six spheres is characteristic, having three small spheres superimposed upon three larger ones. I was not able to find the pole globule at this stage, therefore the ova can not be accurately oriented.

[1]) I am inclined to think that an inequality of the two primary spheres is the normal condition, for although a difference in size between the first two spheres may not be discernible, a difference of some kind really exists since one invariably divides before the other.

The stage with seven spheres I have not with certainty been able to find, but think that such a stage exists.

The stage with eight spheres is represented in Pl. I, Fig. 14. It will be seen that there is a slight difference in size between the four cells of the two poles of the egg, while otherwise, so far as could be observed, they were of the same color and structure. By a further division of the cells the ova become transformed into a blastosphere with a slightly eccentric and oval segmentation cavity.[1])

In surface views of the vegetative pole of such an embryo, may be observed four large cells, only distinguished from the remaining ones by their coarser structure; in optical section it will be seen that the gradation in size of these four cells toward the smaller ones at the opposite pole is very gradual. Subsequent development shows that those four cells constitute the beginning of the endoderm.

Very early in the segmentation stages the thick chorion falls away from the embryo leaving it free to undergo further development. It is seldom that I have observed embryos at the end of the segmentation still enclosed within the chorion, — never after that period.

IV. Gastrulation.

It is difficult to determine the characters by which one can decide when the gastrulation begins since there is no invagination of the endoderm to determine this point. We will consider the beginning of the gastrulation, and hence the second period of development, as being indicated by the division of the four primitive endoderm cells and their growth inwards to fill up the segmentation cavity. It is also at this period, or possibly a little before, that the rudiments of the velum and apical plate appear.

At the end of the preceding period four long cells, with difficulty to be distinguished from the surrounding ones, were to

[1]) It must be remarked here that owing to the great opacity of the ova, the uniformity in size of the segmentation spheres and their indistinct boundaries, and owing to the artifical condition, which produced a large percentage of abnormal types, in the earlier stages difficult to distinguish from the normal ones, — it was difficult to follow with certainty the rhythm in the segmentation after the eight — cell stage. Hence the succeeding stages as far as the blastosphere have been omitted, since I am unable to say with certainty whether the results were those of a normal development or not.

be seen, Pl. I, Fig. 15 and 16, which during the first of the present period increase rapidly in size growing principally inwards toward the animal pole, while in doing so they encroach more and more upon the segmentation cavity and finally nearly obliterate it.

Pl. I, Fig. 15, shows a surface view of these four cells, while Pl. I, Fig. 16, is a section through an embryo of the same stage. An optical section of the embryo at the end of segmentation shows that these cells are wedge-shaped, whereas at the beginning of the gastrular stage they have increased in size at their inner ends, assuming an oval form, Pl. I, Fig. 17 and Pl. II, Fig. 20, which immediately distinguishes them from the surrounding cells of the ectoderm.

As they continue to increase in length their inner extremities, which are relieved from the lateral pressure of the adjacent cells, expand into peculiar club-shaped cells, which nearly fill the segmentation cavity. Pl. I, Fig. 18.

It is during this increase in size and inward-growth of the four primitive endoderm cells, that at first two and later four or five slightly enlarged cells at the apical pole become furnished with tufts of short cilia, thus forming the beginning of the apical plate; at the same time the velum is established by the appearance of similar tufts of cilia upon each one of a double row of cells extending around the equator of the embryo. Externally this double row of cells cannot be recognised as such, for the cells constituting it are only distinguished from the others by the presence of the cilia, Pl. II, Fig. 21. A longitudinal section of the embryo at this period, Pl. I, Fig. 17 and 18, shows however that in reality the cells in question and their nuclei are considerably larger than those of the remaining ectoderm cells.

The embryo, which at this early stage is already provided with the rudiments of the velum and apical plate, can only roll about on the bottom of the glass or whatever vessel it may be in. It is not able to swim freely in any direction, until the growth of the post-velar portion has carried the centre of gravity out of, and behind the centre of the equatorial plane in which the velum lies.

It is important that we should have a correct idea of the general shape of the embryo and the relation of its main axes, before describing the further changes which take place. The embryo, as we have seen, is spherical. The four endoderm cells are situated at the vegetative pole, and opposite is the animal or

apical pole, at which point we have seen the enlarged ciliated cells which later form the apical plate. The main axis of the embryo therefore passes through the apical plate and the centre of the blastopore.

The velum at first either coincides with, or when parallel to the equator, is situated *nearer the blastopore than the apical pole.* Pl. II, Fig. 22. An indefinite number of radiating axes may be drawn in the equatorial plane having for their point of intersection the point at which the main axis of the embryo is perpendicular to the equatorial plane. The embryo is therefore a pure radial type. But this condition does not last long for two large cells soon appear, one of each side of the four endoderm cells, destroying the previous symmetry and transforming our embryo into a bilateral organism. These two cells may be seen in optical section in Pl. II, Fig. 22 *en.* m^1 and *en.* m^2; and in actual section in Pl. II, Fig. 23. During their present condition they may be called the endo-mesoderm cells since the subsequent division of each of these cells gives rise to two cells, one of which becomes the primitive mesoderm cell, and the other, after remaining in the mouth of the blastopore for some time, finally becomes pushed inwards by the narrowing of the blastopore, and forms one of the endoderm cells lining the cavity of the mesenteron. Bearing these conditions in mind we have only a few points to consider before we reach the next stage.

On examination of Pl. I, Fig. 17 and 18, and Pl. II, Fig. 22, it will be seen that one or at most two cells only, intervene between the velum and the large endoderm cells which fill the mouth of the gastrula. By the elongation and inward-growth of the endoderm cells the body of the embryo loses its spherical shape and becomes somewhat lengthened, to assist which, the ectoderm cells we have just pointed out increase in number and thickness. The cells which constitute the velum are large and wedge-shaped with convex outer surfaces, which later often become somewhat indentated at the point where the cilia originate. Pl. II, Fig. 25.

The embryo-cap, or that portion of the embryo anterior to the velum and including the future apical plate, consists of a small number of wedge-shaped cells decreasing slightly in size towards the summit, which is occupied by four or five cells provided with a few cilia, but otherwise indistinguishable from the surrounding cells. With the increase in length of the embryo, the embryo-cap

cells lose their wedge like form and become somewhat flattened. Pl. II, Fig. 25 and 32.

V. Migration of the Blastopore and Appearance of the Dorso-ventral Axis.

At the end of the preceding stage the four primitive endoderm cells had divided at their inner ends giving rise to a small number of large cells, while the outer ends of the four original cells still filled the mouth of the gastrula.

If, at the beginning of the present stage, we examine the blastopore it will be seen that the four large cells with which it was filled have now increased to seven or eight. In surface views of this part of the embryo, these cells are easily distinguished from the surrounding ectoderm by their finely granular appearance and the absence of nuclei, whereas the ectoderm cells are filled with fat globules and contain well marked nuclei. Pl. II, Fig. 28 and Pl. III, Fig. 43. This difference in the appearance of the two kinds of cells is easily explained by the examination of sections through the blastopore at this stage. Such a section is seen in Pl. II, Fig. 25. In this example the number of free endoderm cells is very small, the majority consisting of elongated cells distended at their inner extremities and closely packed in the gastrula mouth, which they completely fill. The nuclei are situated at the inner ends of the cells a long way removed from the outer surface, thus accounting for their absence in surface views. The outer ends of these cells are usually deeply stained and consist of very fine and uniformly granular protoplasm extending inwards as far as the large vesicular nuclei. Beyond the nuclei the cells consist of protoplasm, which is reduced almost to a network by a great number of cavities, which in the living cells were probably filled with fatty material, now removed by the action of the reagents.

In the same Fig. 25, *en.*[1] is a very large cell almost divided into two portions, the narrowest part connecting the two ends is filled with protoplasm, the granules of which have arranged themselves with their long axes parallel to the long axis of the cell. A second cell *en*[2] may be seen in the same section, that has completely separated itself from the parent cell, which still helps to fill the blastopore.

Finely granular protoplasm may be seen in cell *en*[2] marking the place where it was formerly connected with its parent cell.

Such endodermic cells, which have only recently been separated from those filling the mouth of the gastrula, can always be distinguished by the one sided position of this deeply stained patch of protoplasm, which afterwards becomes uniformly distributed throughout the cell.

About the twentieth hour the blastopore no longer retains its central position at the basal end of the embryo. It gradually moves towards the future ventral surface retaining meantime its size and characteristic appearance. In Pl. II, Fig. 27 is a section of an embryo of twenty-five hours where it will be seen that the centre of the blastopore is not equally distant from the velum on each side, a greater number of ectoderm cells intervening on the dorsal, than on the ventral side. Pl. III, Fig. 40 is also a longitudinal section through the blastopore of an embryo 20 hours old, it will be seen that the gastrula mouth has become more ventrally placed and smaller, while it is filled with only two large cells and the small ends of two others. In Pl. II, Fig. 31, it has become still more ventrally placed and slightly elongated.

After the blastopore has reached about the position indicated in Pl. III, Fig. 34, it decreases in size and occupies the bottom of a V shaped hollow, which increases in depth as it decreases in width. The apex, which is the deepest part of the furrow, is directed towards the velum. As the groove narrows it grows deeper and finally it is impossible to distinguish in surface views the ends of the endoderm cells, which fill the mouth of the gastrula and pave the anterior floor of the deepening groove. Sections show us plainly that the blastopore, which is now very much reduced in size, occupies only a small part. the apex, and very deepest portion of this V shaped furrow, Pl. III, Fig. 41, while the remaining part is occupied by ectoderm cells, which ascend abruptly to the surface in the wall next the velum, while at the opposite side the slope is more gradual. The enormous cells, which formerly filled the blastopore, have become extremely attenuated and, at their outer ends, reduced almost to lines in order to accomodate themselves to the now reduced limits of the blastopore.

Before the pressure upon the surrounding cells has forced the ends of the endoderm cells out of the gastrula mouth and therefore permanently closed the blastopore, the ectoderm cells surrounding the V shaped pit have increased slightly in size and become conspicuous by their regular order, while at the same

time they assume at the apex of the groove a decidedly circular arrangement, Pl. III, Fig. 37 and 38. Meantime the diverging arms of the V have become parallel enclosing a narrow and shallow furrow *which deepens at its widened basal extremity, Pl. III. Fig. 38, forming a round opening similar to the one at the opposite end of the furrow, but smaller and not so distinctly marked.* The furrow connecting the two pits becomes less distinct and soon after the posterior pit m' disappears leaving a long shallow groove to mark its former position, Pl. III, Fig. 37 and 42; the anterior pit however remains and, becoming completely surrounded with prominent wedge-shaped cells, forms the opening of the oesophagus or mouth. At the same time the blastopore closes.

The ectoderm cells, which formed the side walls and floor of the V shaped furrow, now constitute the walls of the stomodaeum or oesophagus.

Just before the blastopore moved from its central position at the basal end of the embryo it was filled with four large cells, which had grown into and almost filled the segmentation cavity, at the same time they underwent a division more or less perfect in a plane perpendicular to the long axis of the cells. The products of this division remain for some time intimately associated with the parent cells, Pl. II, Fig. 22, showing by their configuration and position the source of their origin. As the blastopore however moves towards the ventral surface, the cells in question become separated from the parent cell and assume rounded or somewhat distorted and compressed shapes in order to accommodate themselves to the limits of the segmentation cavity, which they soon completely fill. At the same time the parent cells, which continue to occupy the blastopore, divide parallel to their long axes increasing the number of cells in the mouth of the gastrula to seven or eight. As the blastopore migrates toward the position of the future mouth, the products of the first division of the four endoderm cells become less distorted and, as the embryo increases in length, arrange themselves symmetrically beneath the apical plate. The products of each subsequent cross division of the cells which fill the blastopore become smaller with each succeeding division, owing to the diminishing size of the parent cells. By the time the blastopore has closed, the endoderm cells, which before were irregularly arranged and completely filled the segmentation cavity, have now assumed some order in their distribution. A long

slit-like cavity appears in the centre of the mass of endoderm cells, the dorsal and anterior walls of which are formed by the larger cells, the oldest and largest cells forming the anterior wall. The posterior ventral wall of the cavity is formed by the smallest and youngest cells and also by those cells which, with the closure of the blastopore, were forced out of the mouth of the gastrula into the interior of the embryo.

Before the blastopore has commenced to leave its primitive position opposite the apical pole, the *endo-mesoderm* cells leave their median lateral position and approach each other on the future dorsal side of the embryo, at the same time they become elongated, the inner ends being rounded and somewhat distended, while their outer ends are more or less rod-shaped owing to the pressure of the adjacent cells. When the cells in question have reached a distinctly dorsal position, they divide in a plane perpendicular to about the middle of the long axis of the cells, and thus give rise at their inner ends to the primitive mesoderm cells, which for some time show by their position and contour the source of their origin. *A long time after the production of the primitive mesoderm cells, one may see in sections the stalk-like ends of the endo-mesoderm cells situated on the dorsal edge of the blastopore, by the subsequent closure of which they are forced into the interior of the embryo, and form a part of the endoderm lining of the mesenteron.*

During the first movements of the blastopore I have often noticed two small cells between the primitive mesoderm and the remnants of the endo-mesoderm cells. Pl. III, Fig. 40 *a*. I think that they originated by a second division of the latter cells. I was not able to find them in later stages. Whether they are the products of abnormal development or not, I am unable to say but am confident that they do not take any part in the formation of the mesodermic chords. They disappear soon after their first appearance.

The two primitive mesoderm cells, whose dorsal position now marks a further advance in the differentiation of the simple bilateral into the dorso-ventral type, are large and spherical, but finally become placed so closely to each other, that their point of contact is transformed into a plane, which exactly coincides with the median longitudinal dorso-ventral plane of the embryo. Pl. III, Fig. 36, and Pl. II, Fig. 29, and 30. This partially spherical form soon changes to an oval one that precedes the first division, which takes place at right angles to the long axis of the cell and

nearer the anterior than the posterior end, thus giving rise to two unequal parts. Pl. IV, Fig. 49 and Pl. III, Fig. 37.

Before the nuclear changes which precede division have appeared, the nucleus is surrounded by an envelope of finely granular protoplasm, which, just previous to the formation of the spindle, becomes beautifully radiate sending long thread like arms of protoplasm toward the periphery of the cell, which is filled with the vacuolated protoplasm similar to that seen in the endoderm cells; Pl. II, Fig. 32. Pl. IV, Fig. 49 represents a section through one of the primitive mesoderm cells during its first division; cells m^1 and m^2 and the subsequent cells produced from *pr. m* do not divide themselves until each of the primitive mesoderm cells has given rise to a single row of smaller cells, the two V shaped rows thus formed diverging forward and downward, while the two primitive mesoderm cells form their point of convergence; Pl. IV, Fig. 50. The smaller mesoderm cells now divide giving rise to a double row of cells compactly arranged and extending forward beyond the velum. The primitive mesoderm cells are even then plainly visible as two large cells at the point where the mesoblastic cords meet.

General shape. During the changes just described the general shape of the embryo has undergone some rather important modifications to which we will now turn our attention. The basal end of the embryo was at first round, but by the time the blastopore has reached the edge of the ventral surface, it has become dorso-ventrally compressed; Pl. II, Fig. 30 and 32. As the blastopore encroaches still more upon the ventral surface two swellings may be observed on each side of it; Pl. II, Fig. 31. *As the blastopore moves forward and leaves its intermediate position between the two swellings, they unite and form a median protuberance which developes into the foot, whose dual origin is for a long time indicated by the presence of a small median indentation.*

When the blastopore closes the foot has only attained insignificant proportions.

After having obtained an idea of the general form, and the relation of the internal to the external changes which have taken place, we can more easily comprehend the exact nature of the movements of the blastopore, which have thus far played so important a part and served as a landmark for the study of the other changes which take place. As we might very well imagine, *a priori*, the motive force of the ventral movement of the blasto-

pore does not lie in the cells filling the blastopore itself, but entirely outside of them and indeed in the cells which lie upon the dorsal side of the embryo. An unexpected help in locating and following the growth of these cells was given in the appearance of two large cilia bearing cells, which appear very early on the dorsal side of the blastopore just about the time it begins its ventral movement; Pl. II, Fig. 27 and 32, and Pl. III, Fig. 36. These two cells, whose outer ends are sharply granular and covered with very fine motionless hairs, are situated close together one of each side of the median dorso-ventral plane and at first on the dorsal side. Only one row of cells separates them from the blastopore while two or three intervene between them and the velum. A flattening of these second intervening cells causes an increase in their superficial area, at the same time they increase in number by division. The large velar cells remaining fixed, this extension of the dorsal surface, not being counteracted by a corresponding one on the opposite side, forces the blastopore and the anal cells from their original position towards the ventral surface. This continues until the blastopore has reached the position indicated in Pl. II, Fig. 31 and 32, when the movement is accelerated by growth in another region. By comparing Pl. II, Fig. 25, with Pl. II, Fig. 27 and 32, it will be seen that the number of cells upon the ventral side has remained nearly the same while on the dorsal side they are not so deep, and have plainly increased in number and extent.

Fig 1.

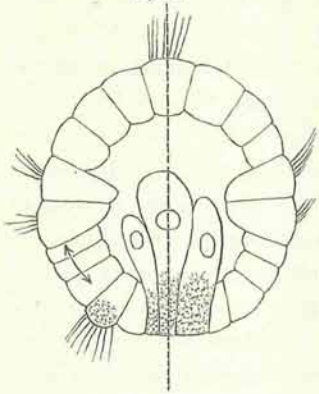


Fig. 2.

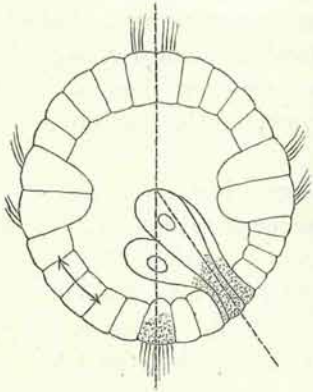
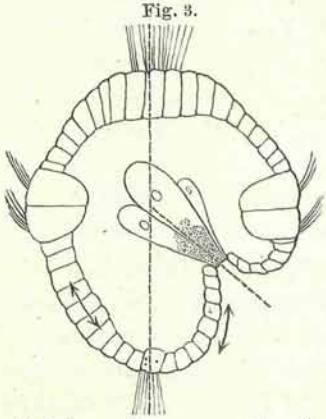


Fig. 3.



After the anal cells have been carried by the growth of the dorsal surface almost to the basal end of the embryo, the cells between the blastopore and the anal

cells rapidly multiply, thus increasing the distance between those two structures while the anal cells remain almost motionless at the basal end of the embryo.

The three adjoining wood cuts will make these changes in the position of the blastopore easily understood.

During the present stage the velum has assumed enormous proportions, and is composed of two rows of large cells encircling the embryo, and situated somewhat nearer the apical than the basal pole. The cilia, which upon their first appearance formed a tuft in the centre of each velar cell, increase in size and arrange themselves in two continous bands passing through the centre of each row of cells. The nuclei are situated in the centre of the cells opposite the cilia, from which they are separated by a band of finely granular protoplasm the width of which equals that of the band of cilia; Pl. II, Fig. 27 and 32. As the embryo grows older, the anterior row of cells decreases in size and becomes hardly distinguishable upon preparation of the whole embryo. They form what we shall call the anterior support cells of the velum. At first they are triangular with their bases resting upon and conforming to the adjacent wall of the large velar cells; later they become much flattened and elongated, but still retain a small band of cilia on their outer surfaces.[1])

The posterior row of velar cells increases in size and forms a single row of very large laterally compressed cells, whose cilia have also increased in size and form a rather broad and continuous band passing through the centre of the cells. This band is very distinct in preparations of the whole embryo owing to the absence of fat globules in the velar cells at the base of the cilia.

A third row of cilia bearing cells is formed on the posterior side of the large cells, and afterwards constitutes the posterior row of support cells. On the dorsal side of the embryo this latter row of cells becomes widened and especially well marked in preparations of the whole embryo. The band is widest on the dorsal side, in the centre of which it becomes slightly indentated. Pl. III, Fig. 36. It gradually decreases in width and distinctness as it passes towards the ventral side, where we are no longer able to

[1]) I have often observed in young specimens that, while the velum consisted of only one row of cells on the dorsal surface, that on each side of the embryo the velum often developed two or even three rows of cells. I have not observed this condition in stages later than the closure of the blastopore.

distinguish it except by means of sections, on examination of which those cells will be readily distinguished as a single row of wedge-shaped ciliated cells closely applied to and supporting the very large quadrate or wedge-shaped velar cells; Pl. IV, Fig. 51 and 52, etc.

The peculiar form and distinctness of this widened dorsal portion of the posterior row of support cells appears soon after the blastopore has begun to move toward the ventral surface and is retained until after the evagination of the shell gland.

The shell gland makes its first appearance shortly before the closure of the blastopore, as a plate of thickened cells; Pl. III Fig. 41 *sh. g.*, situated on and including almost the entire dorsal surface of the embryo posterior to the velum. A flattening in the outer ends of the cells and a large shallow depression in the centre of the plate are the first indications of the invagination which is formed there at the beginning of the next stage.

The cells composing the embryo cap have during this stage increased in number and decreased in size, assuming a more cuboidal shape, only those at the summit bearing cilia. The cells of the apical plate have also increased in number and become much elongated forming a lenticular thickening, the convex side of which is turned inwards. Pl. IV, Fig. 49 and 50. As these cells increase in length their nuclei, increasing in size and containing less stainable substance than before, leave their central position and move towards the inner ends of the cells.

As the apical plate increases in size the outer ends of the cells composing it, which were at first rounded, become flattened and finely granular, while the nuclei, now more deeply located, are no longer visible from the exterior. Meantime two cells, one on each side of the apical plate, have become conspicuous by retaining their rounded outer ends and by an increase in length which causes them to project some distance above the level of the surrounding cells. Pl. III, Fig. 35, 36 etc. *a. c.* The projecting ends become filled with a number of highly refractive granules and covered with extremely fine radiating hairs, which when seen upon the living animal are perfectly straight and motionless.

The remaining cells of the apical plate, and also nearly all the cells of the anterior half of the embryo-cap, have become supplied with short and active cilia, which upon the two or three cells in the centre of the apical plate and exactly at the apical or animal pole, become enormously developed and form a tuft of

from fifteen to twenty long cilia, which, when the embryo is in motion are entirely inactive and either remain straight and motionless or hang gracefully to one side in one or more festoons, which vary in position as the embryo moves about. When the latter is quiet, they are motionless, or at most show only a slight restless or uneasy movement.

At first sight one might attribute to these long cilia the function of regulating the direction in which the embryo moves, a function, which, considering the fact that the apical plate moves first, could be much more easily exercised at the opposite end of the embryo. I am inclined rather to attribute the function above mentioned to the smaller cilia around the apical end of the embryo, the larger and longer ones being especially modified to serve as *sensory hairs*, a supposition the more reasonable when we consider their sluggish motion and intimate connection with the very early developed rudiments of the sopra-oesophageal ganglion. Besides it is not easy to see why the embryo should have two rudders of different lengths at the same pole. It is possible that the long stiff hairs of the anal cells, which in this stage have attained considerable proportions, may assist in guiding the movement of the embryo, through I have never seen them in motion of any kind.

VI. From the Closure of the Blastopore to the Formation of the Nautiloid Shell.

When the closure of the blastopore has taken pace, the body of the embryo has slightly elongated, and the posterior portion becomes somewhat flattened dorso-ventrally, which is however obscured by the ventrally placed foot which is now rapidly increasing in prominence.

The endoderm which at the end of the preceding section had begun to show more regularity in the arrangement of its cells, has ceased to fill so completely as before the body cavity, and with the rapid division of the cells and their consequent reduction in size, has begun to assume the characters which distinguish it during this stage. Just after the closure of the blastopore, the endoderm cells are very large, their irregularity in size and shape still indicating the various modifications between the older and more rounded cells and the long wedge-

shaped and compressed ones, which have only recently freed themselves from the mouth of the closing gastrula. In the midst of the anterior endoderm cells, which are the first to show more regularity in size and a more symmetrical arrangement, appears a longitudinal opening widest anteriorly and gradually decreasing in width towards the opposite extremity. As this slit like opening increases in length, the cells at its posterior extremity, which hitherto have shown very irregular and compressed outlines, also become cuboidal in shape and arrange themselves in a single row around this cavity. This sac, or the mesenteron, now becomes more elongated, its anterior and lateral walls being composed of slightly wedge-shaped cells, those at the anterior end being larger than those at the opposite extremity. The ventral wall is composed of a layer of very much smaller cells, while at first the posterior dorsal wall is very imperfect owing to the want of space caused by the enormous development of the shell gland.

The large endoderm cells, which just before the closure of the blastopore entirely filled the segmentation cavity, have become so reduced in size through the demands made upon them by the active embryo that a large space now exists between the walls of the mesenteron and the ectoderm, which constituted the body cavity.

The further development of the mesenteron consists in the contraction of the elongated sac just described into a smaller and spherical one which assumes a more posterior position on the dorsal side of the embryo; Pl. V, Fig. 58. We have already seen how at the end of the preceding section the V-shaped furrow at the bottom of which was the blastopore became transformed into the oesophagus. As the blastopore was gradually moved towards the ventral side the few ectoderm cells Pl. III, Fig. 41, which intervened on that side between the blastopore and the velum, became subject to a pressure which finally bent them inwards and upwards, where they formed the anterior wall of the apex of the V shaped furrow, and when the blastopore closes they form the anterior wall of the oesophagus. As the oesophagus increases in length a marked difference is seen in the construction of its anterior and posterior wall, the former being composed of small cuboidal cells, while the latter is thick and made up of larger and much longer cells. The oesophagus at first grows directly forwards so that the thin anterior wall comes to lie against the velum. As it increases in length the extremity becomes bent inward towards the centre of the embryo, where it comes

in contact with the anterior cells of the mesenteron; as the latter contracts towards the posterior and dorsal portion of the embryo the oesophagus keeps pace with it and describes in its course a more or less perfect semicircle; Pl. V, Fig. 58. The thickened ventral wall of the oesophagus close to the mouth produces a single median invagination, which probably gives rise to the radula, although I have not found embryos far enough advanced to follow its further changes.

The mesoblastic chords, described in the last section, during the present stage increase in length reaching forward as far as the velum, at which point the cells, as they increase in number by division, no longer retain their former shape and intimate connection with each other but become isolated and at the same time assume elongated forms with pointed ends; Pl. IV, Fig. 52 The posterior portions of the mesoblastic chords remain for a long time intact being composed of two rows of cells anteriorly, while posteriorly they consist of a single row of compressed cells. The primitive mesoblast cells are situated at the very posterior extremity of the chords and are still easily recognized as such by their superiority in size.

As the development of the embryo proceeds, a portion of the mesoderm cells, which have become freed from the anterior extremity of the mesoblastic chord, moves towards the dorsal surface of the mesenteron, there forming a layer of cells which gradually grows toward the ventral side. Half a dozen or more of these cells become greatly elongated and, converging toward a point on the dorsal surface, become attached to the shell and form the muscle cells which serve to draw the embryo into the shell, when the latter has attained sufficient proportions. Another group of mesoderm cells extends forward and ventrally, surrounding the oesophagus and forming a specially large collection of cells above and at the sides of the oesophagus and around the invaginated pits on either side of it, which probably give rise to the auditory sacs. At the same time a few scattered cells arrange themselves around the inner wall of the velum; Pl. V, Fig. 60. A few elongated cells usually connect the mesoderm cells encircling the oesophagus with those upon the velum. A special chord of such cells, distinguished by their greater size is always found on either side connecting the oesophagus with the velum; Pl. V, Fig. 60. Besides these a few free, deeply stained and perfectly spherical cells are always to be found in the spaces between the

outer walls of the embryo and the mesenteron. They probably form the blood corpuscles.

I was not able to determine what became of the primitive mesoderm cells which at the 120th hour, the oldest embryo carefully studied, were still distinguishable from the other mesoderm cells by their greater size.

As soon as the foot becomes hollowed within, it is also filled with scattering mesoderm cells.

The shell gland, which at the end of the preceding stage had only formed a thick plate of cells slightly hollowed in the centre, during the present period assumes such enormous proportions as to change the shape of the whole posterior portion of the embryo and to exert a modifying influence upon the processes which take place within the interior of the embryo.

The cells become very deep as the invagination of the gland progresses. The cavity of the gland is at first shallow and wide mouthed, but gradually decreases in size still retaining its circular opening; Pl. V, Fig. 59. When the latter has reached its smallest limits it loses its circular outline, increases in size and becomes laterally extended, at the same time the cavity itself, which has meantime increased in depth, becomes roughly Y shaped, Pl. IV, Fig. 46 and Pl. V, Fig. 64. The shell gland, the cavity of which is now surrounded with very thick walls, occupies almost the entire dorsal surface of the embryo posterior to the velum, and soon encroaches upon its lateral walls; the floor of the cavity decreases rapidly in thickness while its edges become more prominent.

This decrease in the thickness of the bottom of the cavity is due to its lateral and posterior extension, which in time causes the anterior arm of the Y shaped cavity to disappear. Pl. V, Fig. 57 and 65. The bottom of the cavity is thus entirely exposed to the outer world, the only trace of it which remains is the posterior arm of the Y, which still forms an infolding of the ectoderm at the posterior end of the embryo. The lips of the invagination, which has thus become almost entirely obliterated, still retain their great size and form a gradually widening ring which grows toward and finally encircles the ventral surface of the embryo. This thickened ring forms the edge of the mantle.

The shell itself, which at first formed only a thin membrane over the opening of the gland, increases in thickness and becomes hat-shaped and slightly corrugated at its apex; Pl. V, Fig. 57. The

shell finally becomes nautiloid owing to its more rapid growth on the dorsal side.

The anal cells still occupy the posterior end of the embryo; they are triangular in shape, the bases being directed inwards. The apices of the cells have developed a small plate-like projection from which a bunch of long motionless hairs takes its origen; Pl. V, Fig. 65. The nuclei of the cells are large and lie against the inner walls of the cells, as far as possible from the exterior. A flattened and elongated area of cells provided with a thick coat of fine and active cilia exists between the ones just described and the base of the foot; Pl. V, Fig. 57 and 58. As the edge of the shell gland encroaches upon the ventral surface, this ciliated patch of cells becomes bent forward with its outer surface against, or close to, the base of the foot, while the long stiff hairs of the anal cells are now pointed ventrally instead of posteriorly. The projection at the end of which are situated the anal cells now remains stationary, and as the posterior edge of the shell gland, which at this point is thick and composed of very large cells, continues to grow ventrally, it finally envelops the anal projection, leaving only the ends of the anal cells with their long hairs projecting beyond the edge of the mantle; Pl. V, Fig. 58. The cavity of the shell gland has now entirely disappeared, the former thick floor of the cavity has become very thin and composed of flattened lenticular cells; Pl. V, Fig. 65 and 58.

The foot has increased very much in prominence in the last stage; its flattened anterior surface has approached so close to the velum that it almost entirely closes the opening to the oesophagus and completely conceals the mouth from view.

The posterior surface of the foot, which at first was convex, now becomes flattened and developes a thin horny operculum with which the mouth of the shell can be closed. Even at this late stage, if we observe the foot from above or below, a small indentation will be seen in the median plane marking the place where the blastopore passed between the two lateral swellings, the subsequent fusion of which gave rise to the foot.

The auditory organs appear shortly after the closure of the blastopore as two shallow folds of the ectoderm, one on either side of the oesophagus, just below the velum; Pl. III, Fig. 42. These folds at first lie nearly parallel to the long axis of the

embryo. As they increase in depth, their slit like openings come to lie parallel to and directly below the velum upon the ventral side of the embryo. The invaginations are directed forwards and inwards and form two narrow-mouthed sacs which afterwards lie against the inner wall of the velum. Pl. IV, Fig. 45. On this account it is seldom that the same series of cross sections will show both the invagination and its opening equally well. In Pl. IV, Fig. 53—55, which represent three consecutive sections of the embryo cut not quite parallel to the plane of the velum, the cavity of the auditory sac with its external opening can easily be studied. The inner ends of the invaginated sacs become clothed with mesoderm cells. With the closure of the external opening the sacs become entirely separated from the ectoderm and assume a globular form, while the cavity, which was never very large, is now almost entirely obliterated by the increase in depth of the cells which constitute its walls.

Meantime the auditory sacs have left their primitive position beneath the velum and become permanently located in the cavity of the foot.

The velum has not materially changed those characteristics which we have already described in a previous section. It still consists of a central row of large, laterally compressed cells with a broad central band of cilia, flanked on either side by a row of smaller less uniformly regular cells supplied with a narrow band of much smaller cilia. We have already called them the anterior and posterior support cells of the velum, on account of the manner in which they apply themselves to the walls of the larger cells forming a kind of support for them. The peculiar thickening of the posterior row of cells, Pl. IV, Fig. 46, already described can no longer be observed.

The embryo-cap has become greatly reduced in high during the latter stages, hardly projecting above the level of the velum; at the same time it becomes laterally compressed and assumes an oval form, when seen from above or in cross sections, in order to accommodate itself to the opening of the shell, which has a similar shape.

At the sides of the apical plate and somewhat dorsally placed appear two clear refractive spots formed by the projecting rounded ends of four or five cells of the epidermis; Pl. V, Fig. 57 *e. s.* After about twenty-four hours an irregularly shaped pigment spot appears in the place formerly occupied by these cells, Pl. V, Fig. 66, *e. s.*

The projecting cells covered with fine radiating cilia, observed in the preceding stages, Pl. III, Fig. 34, 35 etc. *a. c.* are no longer distinctly visible. The tuft of long cilia in the centre of the apical plate retains however its former prominence. The apical plate has increased slightly in depth, and consequently the cells of which it is composed have become longer and contain nuclei distinguished from those of the other cells in the embryo-cap by their greater size and small quantity of chromatine. These nuclei are situated at the inner ends of the cells and are partly surrounded with vesiculated protoplasm. The outer ends of the cells are filled with fine granules which extend inward as far as the nuclei. This granular protoplasm contains fine lines continuous with the cilia upon the outer surface and extended inward towards the nuclei.

About the seventy-fifth hour two projections are developed upon the anterior surface of the embryo-cap on each side of the median line, Pl. IV, Fig. 45, they are thin walled and dome-shaped and often filled with fine globules of a refractive substance. I was at first inclined to consider them abnormal productions, but am convinced that such is not the case on account of their symmetrical appearance and regular occurrence at this period. By the end of the hundredth hour they have disappeared.

Unfortunately, as has happened with so many other observers who have studied the development of the Mollusca, I am unable to contribute any definite results as regards the development of the nervous system. This is due to the fact that, when the embryos had reached a stage old enough for the study of these changes, they had become so abnormally developed that any attempt to examine them for such a purpose would have been fruitless. The few normal examples of the oldest stages obtained, although sufficient to give the external form, were not abundant enough to afford material for sectioning. A few fortunate sections at this stage however will be of interest. Pl. V, Fig. 62 is one of these sections and it will be observed that the apical plate is still formed of a layer of long cells, while on each side of it the cells have increased in number and formed two lateral masses. In surface views at the same stage, the embryo cap-is seen to be somewhat depressed at the points where these lateral masses of cells should be while the apical plate has left its primitive position at the apical pole and now lies close to the velum on the ventral side of the embryo. An actual invagination was not seen to occur. Whether these lateral masses of cells increase in size and subsequently fuse to form the

supra-oesophageal ganglion, or whether it arises from a thickening of the apical plate or from a combination of both I am unable to say. A paired thickening of the ectoderm constituting the anterior wall of the foot appears at this time, which I am inclined to believe gives rise to the pedal ganglion.

Explanation of Plates.

an. c. anal cells.
a. p. apical plate.
a. anus.
b. blastopore.
en. endoderm.
In. intestine.
f. foot.
m. l. micropyle.
m. c. mesodermic chords.
m. s. mesoderm.
pr. m. primitive mesoderm cells.
m. n. mesenteron.
o. e. oesophagus.
p. g. pole globules.
r. radula.
s. c. segmentation cavity.
st. stomodaeum.
sh. g. shell gland.
sh. shell.
V. velum.
l. s. lateral swellings.

Plate I.

Fig. 1. Chorion. Fig. 1a. Optical section of the chorion still more highly magnified.

Fig. 2. Ovum just after its removal from the ovary.

Fig. 3. Ovum about ten minutes after its removal from ovaries, showing amoeboid arms.

Fig. 4. Ovum $^1/_2$ hour old, showing two pole globules surrounded by the refractive particles in the cavity of the micropyle.

Fig. 5. Ovum with large depression lined with finely granular protoplasm from which a single pole globule rises.

Fig. 6. Ovum $^3/_4$ hour old showing the two differentiated pole globules.

Fig. 7. Fertilized ova during its first division into two spheres.

Fig. 8. Similar ovum with two equal segmentation spheres.

Fig. 9. Ovum with two unequal segmentation spheres.

Fig. 10. Ovum with three segmentation spheres, seen from the animal pole.

Fig. 11. Ovum with four segmentation spheres.

Fig. 12. Ovum with five segmentation spheres.

Fig. 13. Ovum with six segmentation spheres.

Fig. 14. Ovum with eight segmentation spheres.

Fig. 15. Ovum in blastosphere stage seen from the vegetative pole as an opaque object, showing the four endoderm cells en^1. en^2. en^3. en^4.

Fig. 16. Section of an ovum in the blastophere stage, passing through the endoderm cells en^1. en^2, and the laterally placed endo-mesoderm cells *en.* m^1. and *en.* m^2.

Fig. 17. Section through an embryo which has just hatched, (about 10 hours) showing the two equatorial bands of cilia V^1. V^2. and the cilia upon the apical pole *a. p.* also the two large endoderm cells en^1. en^2.

Fig. 18. Section through an embryo of about 14 hours showing the lengthened and enlarged endoderm cells en^1. en^2. en^3.

Plate II.

Fig. 19. Embryo in the blastosphere stage, seen as a semi-transparent object. (10 hours.)

Fig. 20. Embryo at the end of the blastosphere stage, showing four large endoderm cells en^1. en^2. en^3. en^4. and the laterally placed cells *en. m.* which give rise to the primitive mesoderm cells. (12 hours.)

Fig. 21. Embryo of 14 hours; it has just escaped from the chorion.

Fig. 22. Section of an embryo of 14 hours, showing first cross division of the endoderm cells en^1. en^2.

Fig. 23. Dorso-ventral section through the blastopore of an embryo of 14 hours, showing the primitive mesoderm cell before it has separated itself from the cell *en. m.* on the edge of the blastopore.

Fig. 24. Dorso-ventral section through an embryo of 15 hours, showing the primitive mesoderm cell *p. m.* just after its division from the parent cell *en. m.*

Fig. 25. Longitudinal section through an embryo of 18 hours.

Fig. 26. Embryo of 20 hours, seen as an opaque object.

Fig. 27. Median dorso-ventral section of an embryo of 24 hours. The blastopore has become slightly eccentric.

Fig. 28. Posterior end of an embryo of 14 hours, showing four endoderm cells in the blastopore.

Fig. 29. Posterior end of an embryo of 26 hours with ventrally placed blastopore.

Fig. 30. Posterior end of an embryo of 28 hours with still more ventrally placed blastopore.

Fig. 31. Embryo of 30 hours seen as an opaque object.

Fig. 32. Dorso-ventral section through an embryo of 28 hours with ventrally placed blastopore.

Plate III.

Fig. 34. Embryo of 34 hours seen from the ventral surface showing the blastopore at the apex of the V-shaped furrow.

Fig. 35. Embryo about 2 hours older showing the very small blastopore in the semicircular area on the ventral surface of the embryo.

Fig. 36. Embryo of 34 hours seen from dorsal surface, showing the primitive mesoblast cells and the peculiar conformation of the posterior row of velar cells.

Fig. 37. Embryo of 40 hours seen from the ventral surface, showing a stage in the transformation of the V-shaped furrow into the opening of the oesophagus. The primitive mesoblast cells have given rise to two new mesoderm cells.

Fig. 38. Shows the transformation of the anterior end of the V-shaped furrow into the mouth opening.

Fig. 39. Embryo of 40 hours seen from the dorsal surface.

Fig. 40. Section through the blastopore of an embryo of 30 hours. *a*, cell probably arising from a second division of the parent cells of the primitive mesoblast.

Fig. 41. Section through the blastopore of an embryo of 40 hours, showing the blastopore shortly before it closes.

Fig. 42. Embryo of 44 hours seen from the ventral side; the ventral furrow *v. f.* still marks the place where the V-shaped depression formerly existed.

Fig. 43. Basal end of an embryo of 25 hours.

Fig. 44. Cross section through the shell gland of an embryo of 45 hours.

Plate IV.

Fig. 45. Embryo of 65 hours, seen from the ventral surface.

Fig. 46. Embryo of 55 hours, seen from the dorsal surface.

Fig. 47. Embryo of 55 hours, seen from the side.

Fig. 48. Embryo of 65 hours, seen from the side.

Fig. 49. Lateral section of an embryo of 45 hours, showing the first division of the primitive mesoblasts.

Fig. 50. Similar section of an embryo of 50 hours, showing the development of the mesoblastic chords.

Fig. 51. Similar section of an embryo of 58 hours showing the further development of the mesoblastic chords.

Fig. 52. Lateral section of an embryo of 65 hours showing the disintegration of the apical ends of the mesoblastic chords.

Fig. 53—55. Three consecutive cross sections of an embryo of 50 hours, through the region of the mouth, showing the invagination of the auditory organs.

Fig. 56. Tangential section through an embryo of 40 hours showing arrangement of cells around the blastopore.

Plate V.

Fig. 57. Side view of an embryo 80 hours old.

Fig. 58. Side view of an embryo 100 hours old.

Fig. 59. Diagonal section through an embryo of 50 hours.

Fig. 60. Cross section through the median row of velum cells of an embryo 90 hours old.

Fig. 61. Cross section through the velum of an embryo of 100 hours.

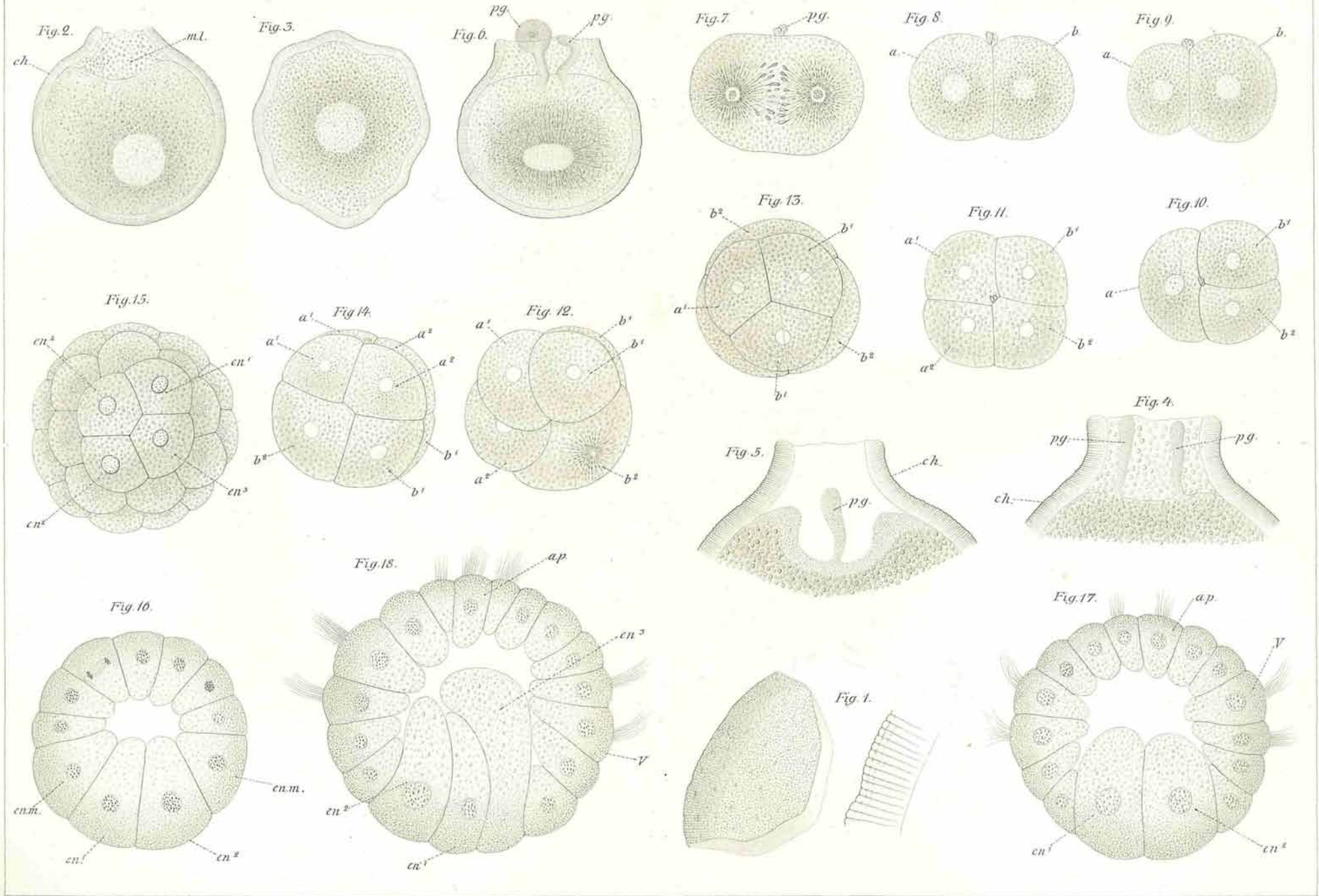
Fig. 62. Median longitudinal section through an embryo of the same age as Fig. 61.

Fig. 63. Tangential section through the ventral surface of an embryo 80 hours old.

Fig. 64. Median longitudinal section of an embryo of 70 hours.

Fig. 65. Median longitudinal section of an embryo of 98 hours.

Fig. 56. Side view of an embryo of 130 hours.



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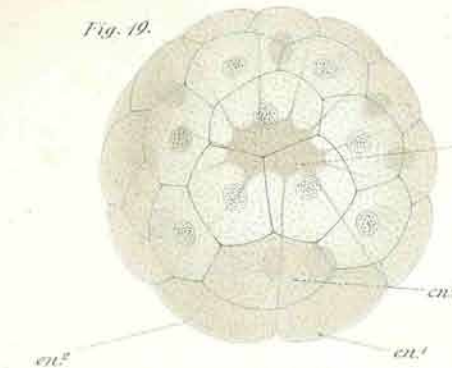
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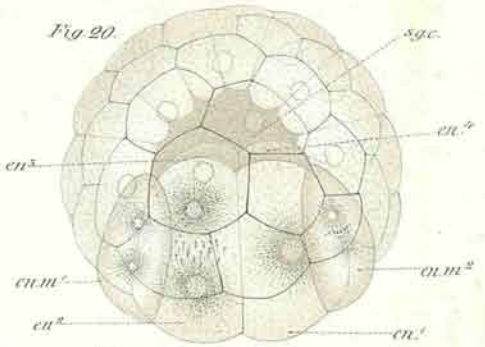
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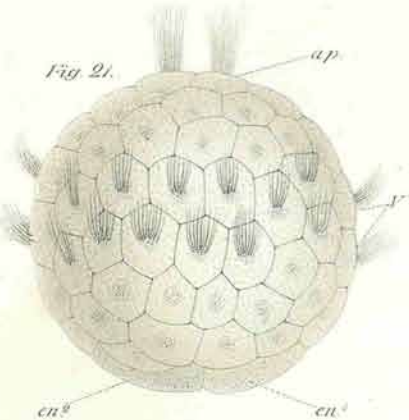
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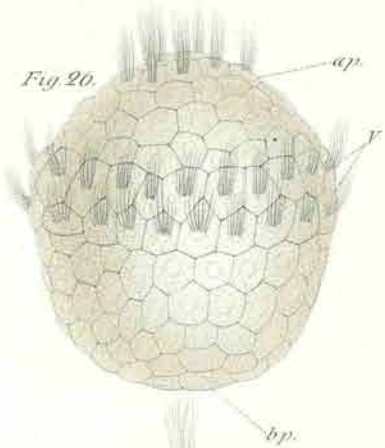
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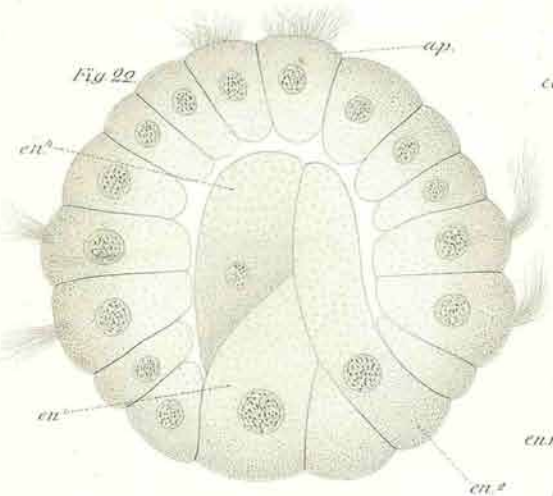
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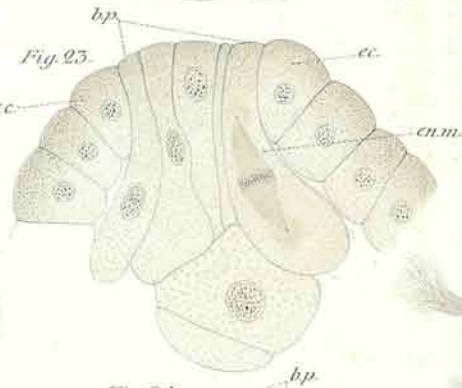
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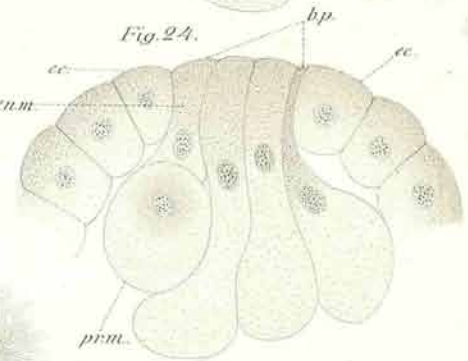
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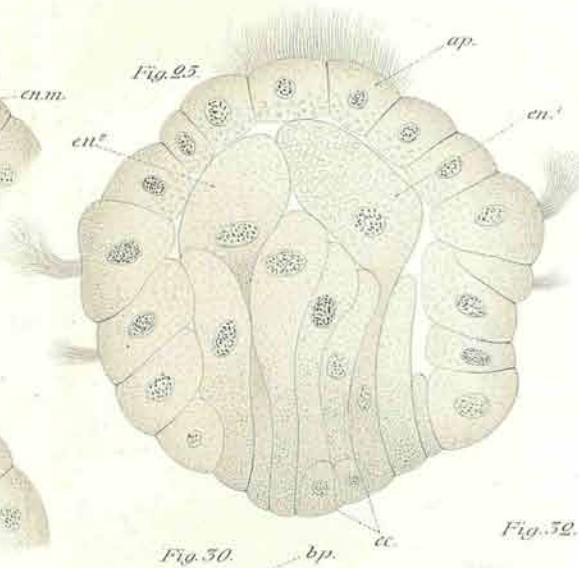
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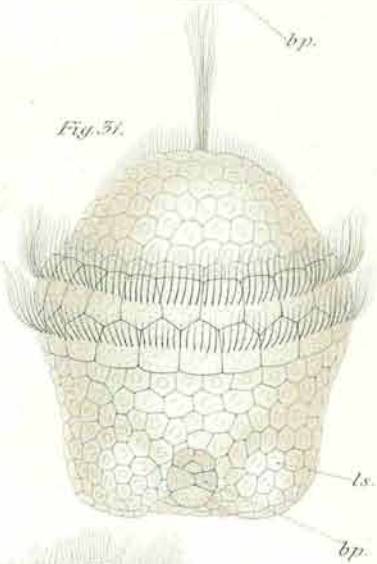
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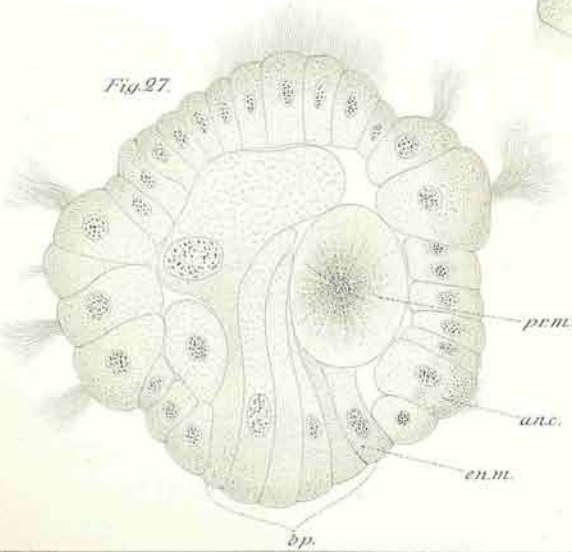
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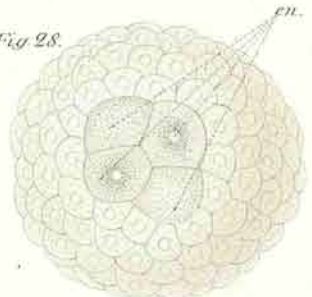
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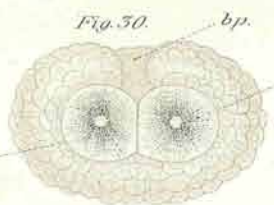
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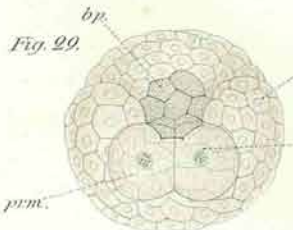
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bp.
ls.
pr.m.
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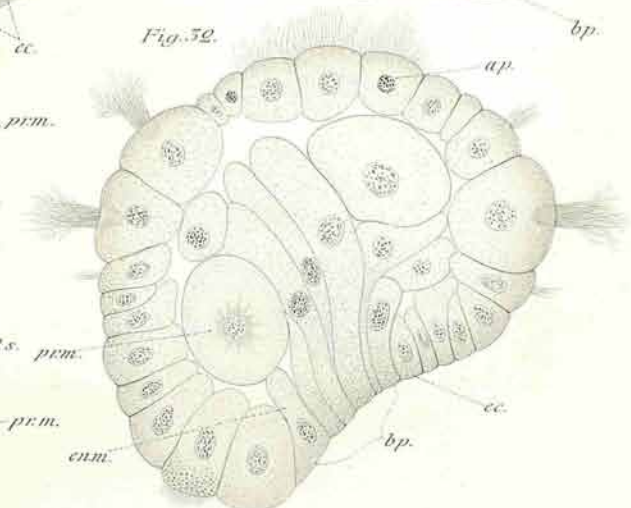
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ap.
pr.m.
ec.
bp.
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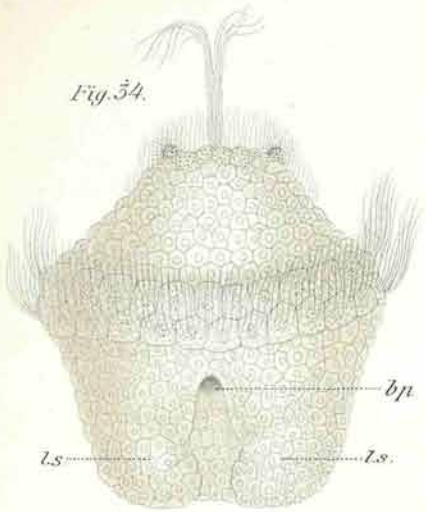
Fig. 34.



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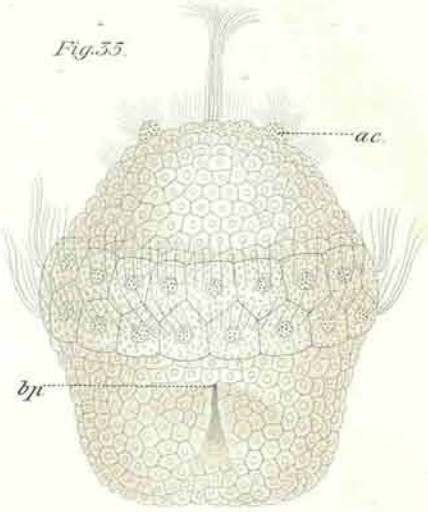

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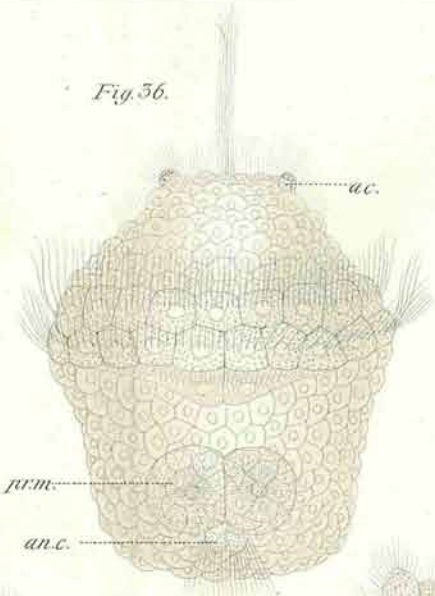

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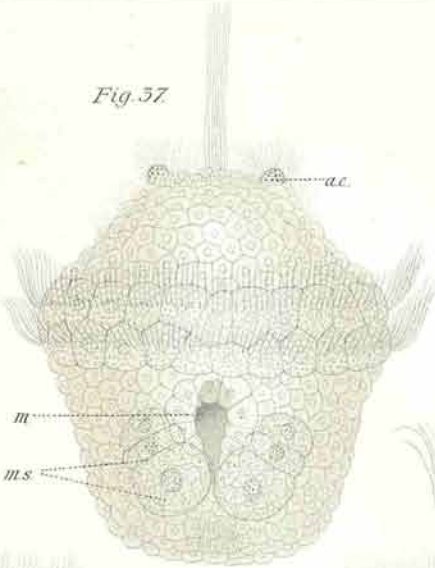
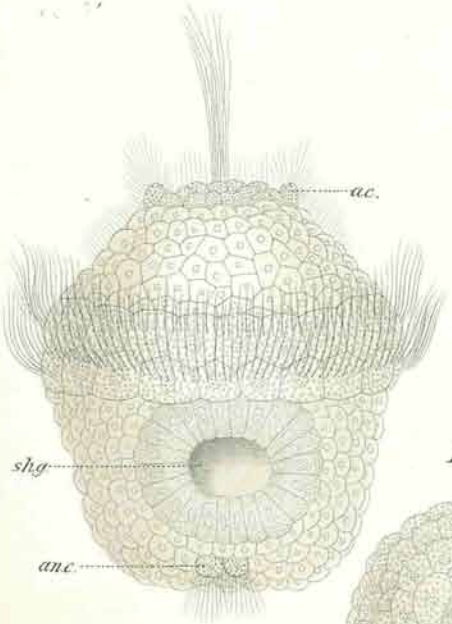


Fig. 38.

Fig. 39.



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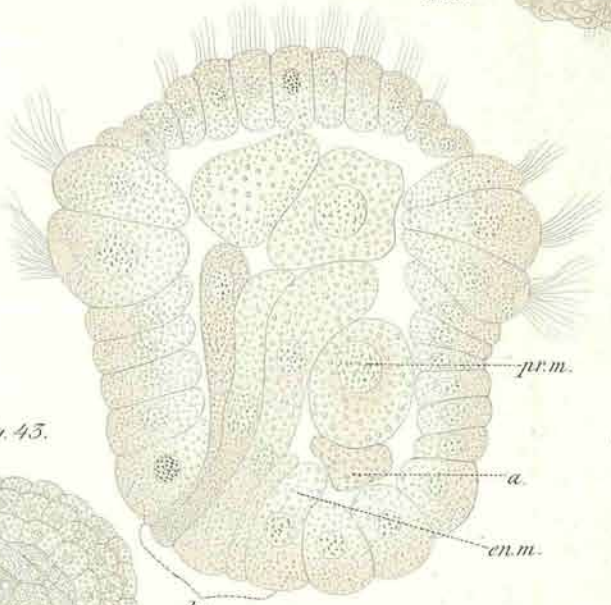

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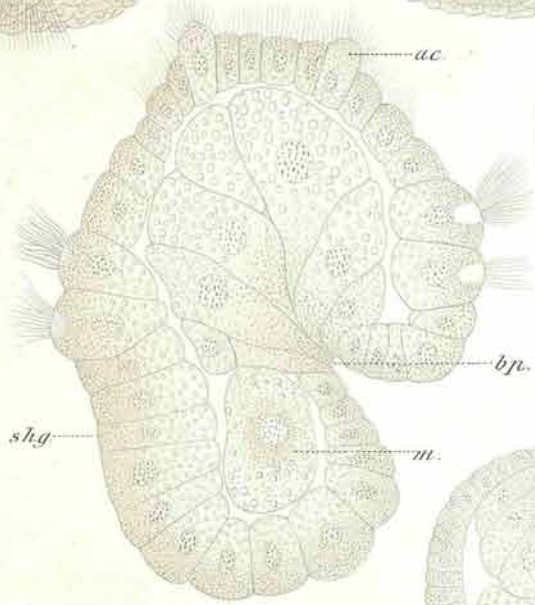

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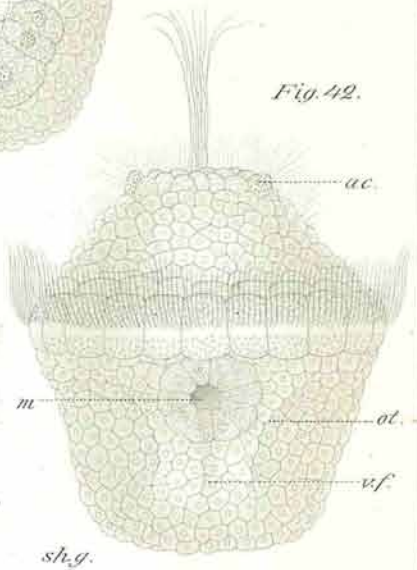

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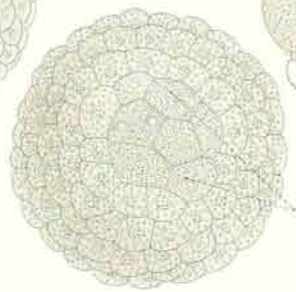

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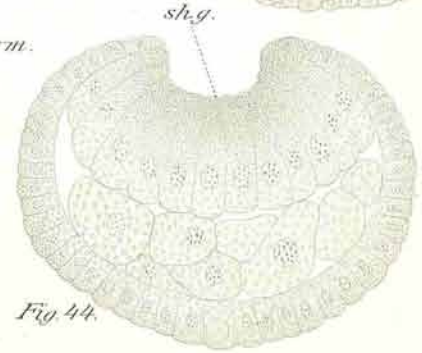

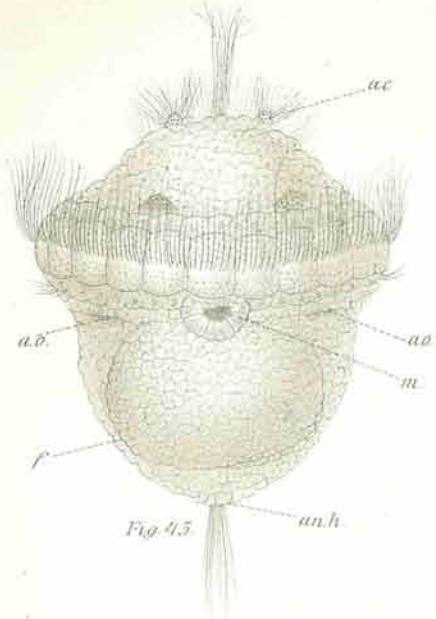

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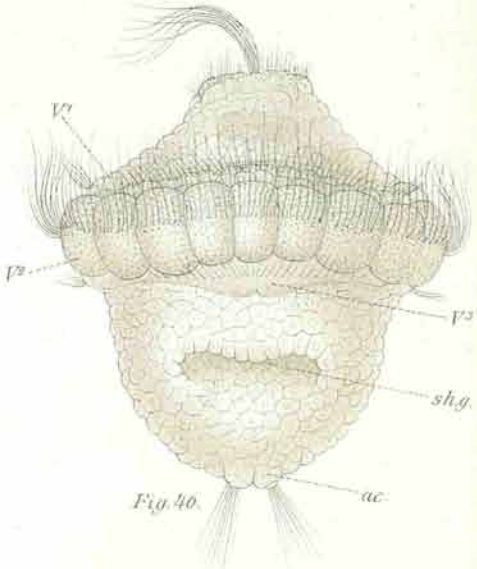

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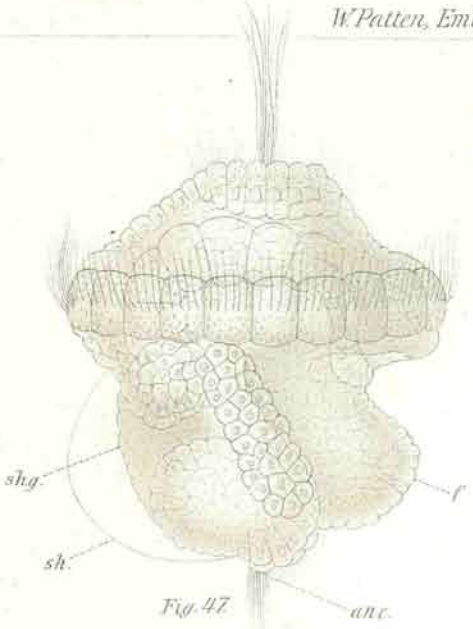

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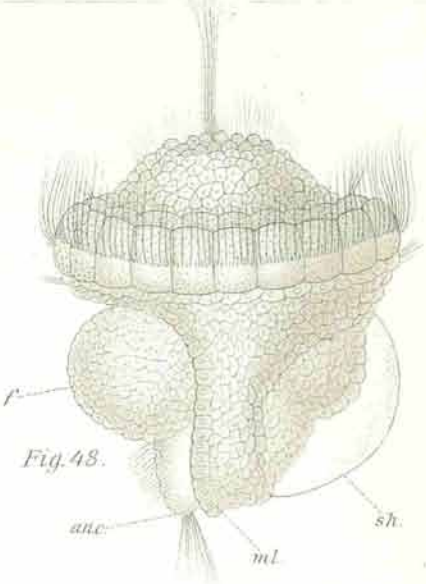

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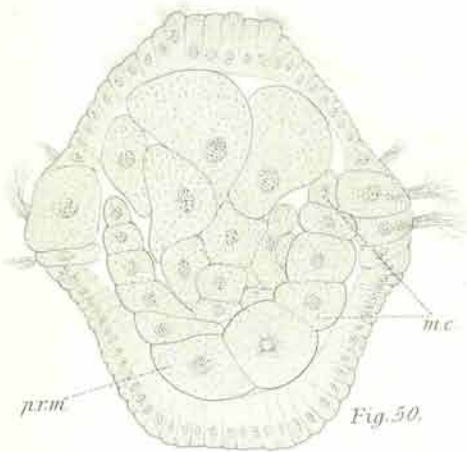

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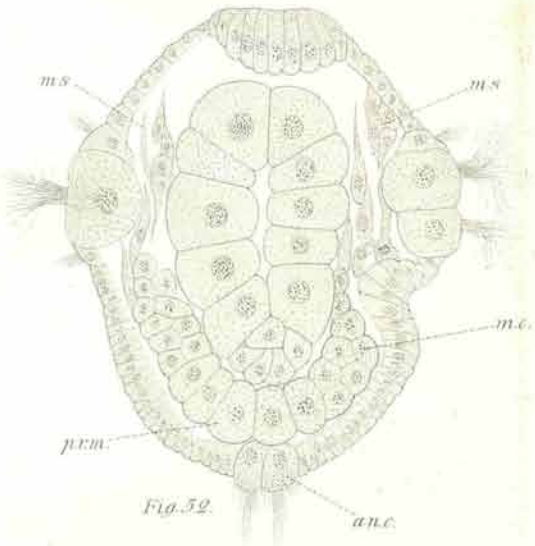

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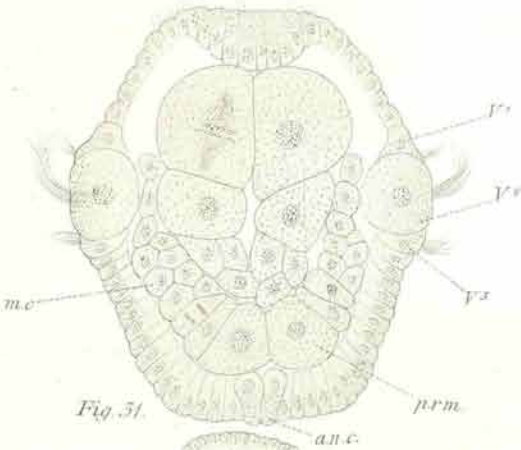

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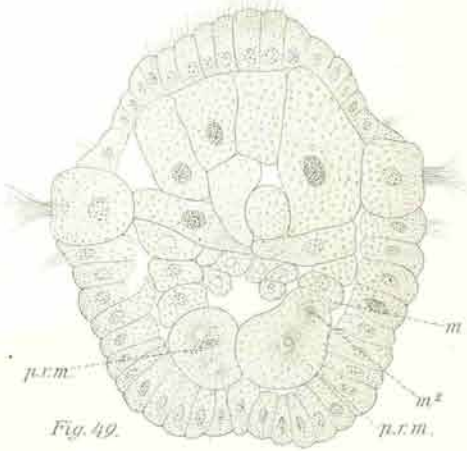

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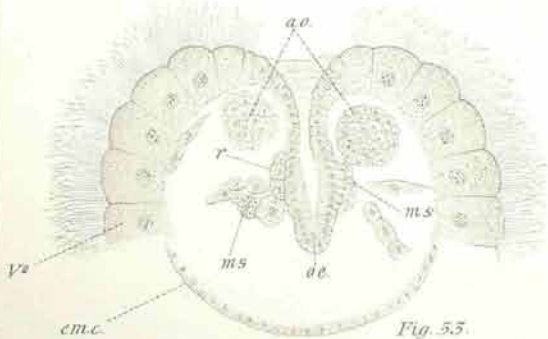

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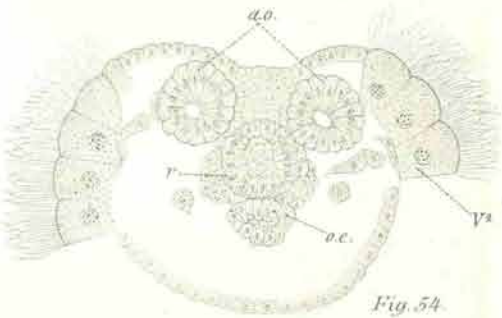

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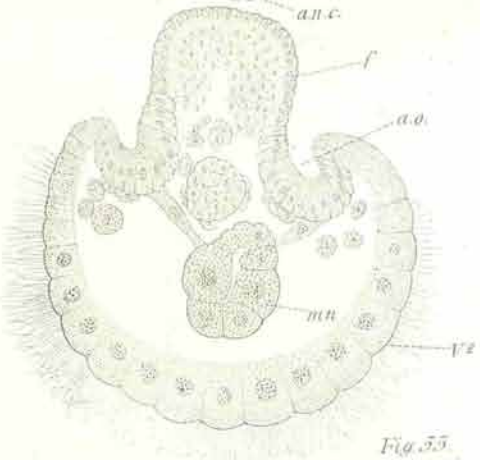

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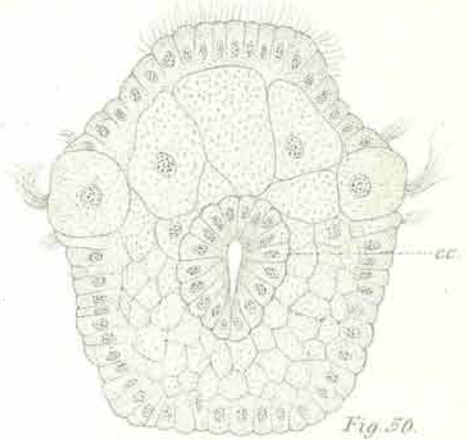

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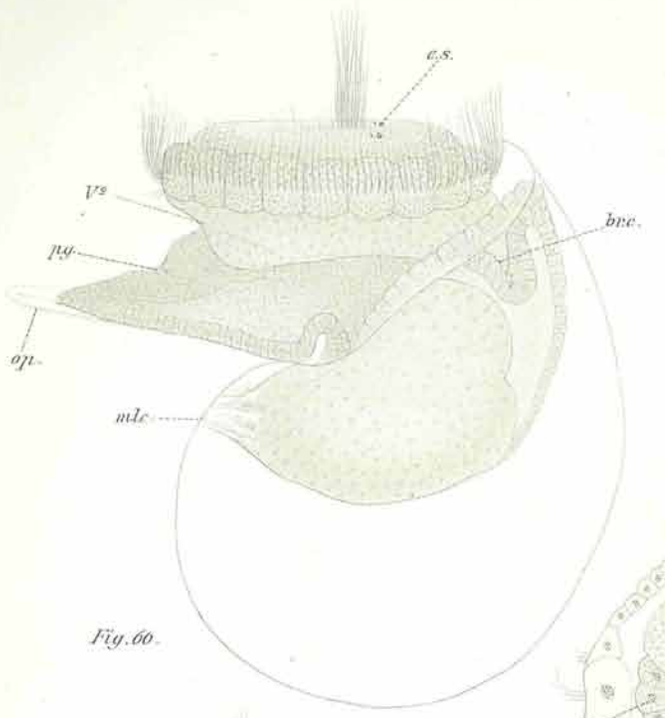

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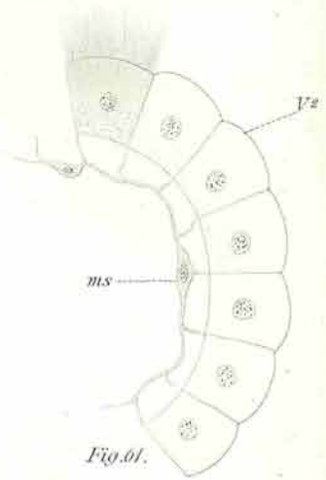

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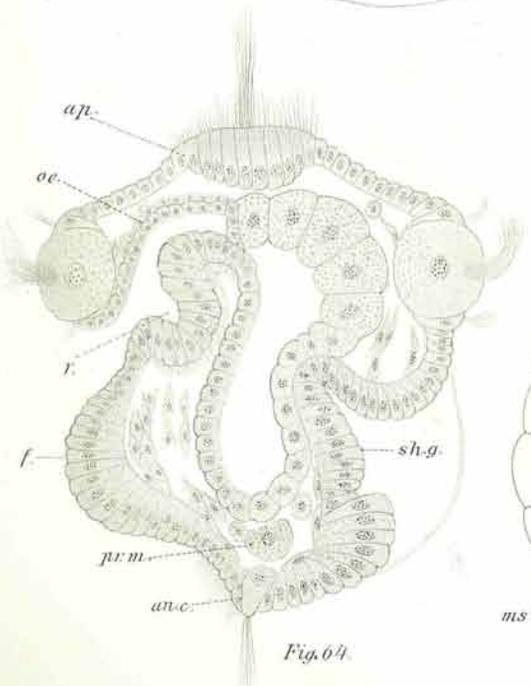

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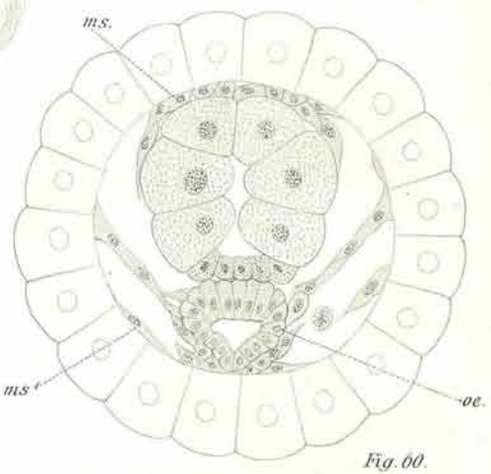

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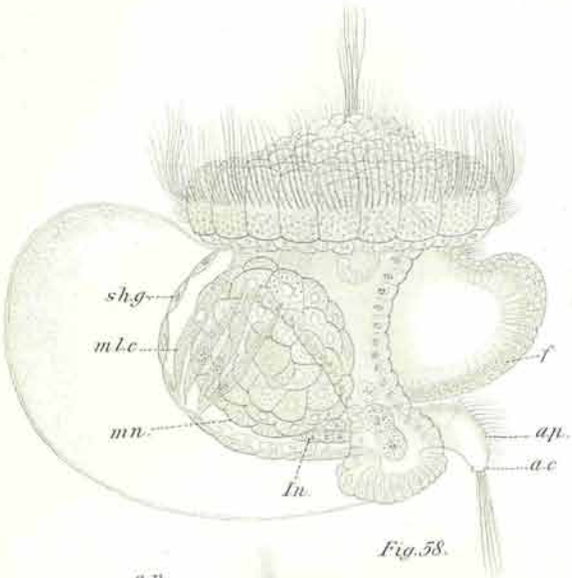

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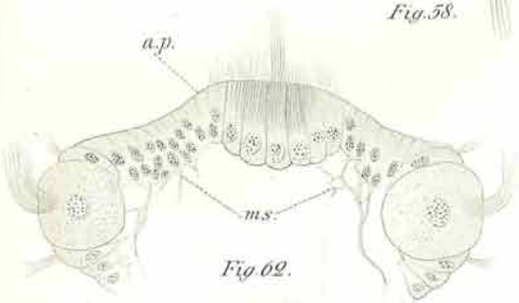

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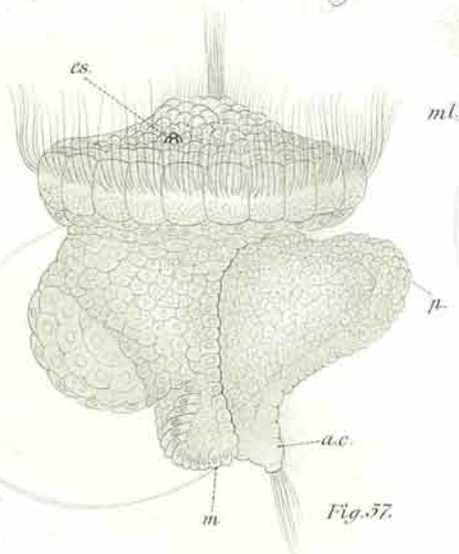

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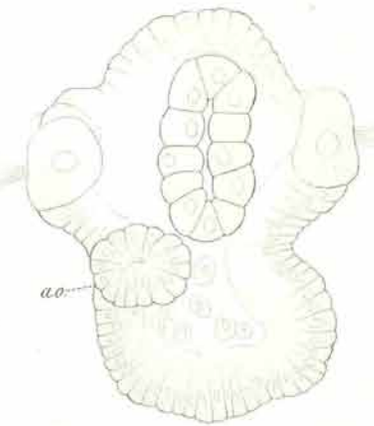

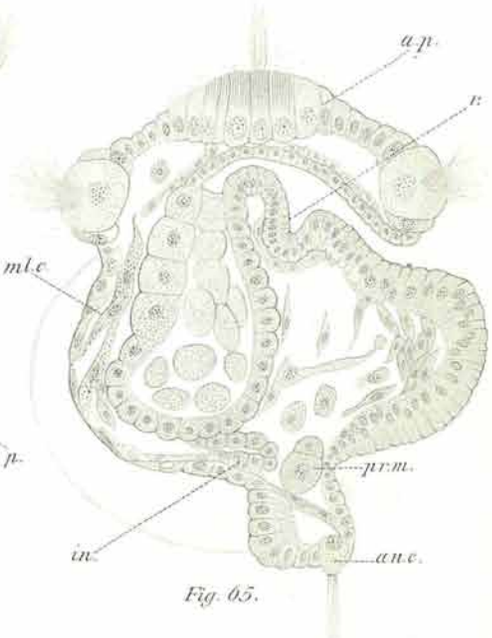

Fig. 65.

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