Contributions to the Morphology of Cyclostomata.

II.—The Development of Pronephros and Segmental Duct in Petromyzon.[1]

By

S. Hatta,
Professor in the College of Peers, Tōkyō.

With Plates XVII-XXI.

The following pages contain the second of a series of studies on the later stages in the development of *Petromyzon*, the first having already been published some time since in this journal ('97, vol. x, pp. 225-237).

Our knowledge of the earliest development of the excretory organs in the lampreys is still somewhat incomplete. This circumstance is, I believe, mainly due to the want of recent investigations upon the subject. Since the appearance of the works by MÜLLER ('75), SCOTT ('82), SHIPLEY ('87), GOETTE ('88), KUPFFER ('90), and others, ten years or more have

1) It was my intention to publish this paper shortly after the appearance of my preliminary notice in 1897, (Annot. zool. Jap., vol. I. pp. 137-140) but various unavoidable circumstances have combined to cause the delay. Meanwhile I have had opportunities of renewing my study on various points and the results here given are different from those of the preliminary paper in several important respects.

elapsed, and, so far as I am aware, no important fact has been added during the interval by any renewed researches. I need not, therefore, apologise for the publication of the present paper which embodies the results of my study on the subject during the last few years.

The investigation of a longitudinally stretched, or a metamerically arranged, organ-system such as the neural canal, the chorda, the pronephros, &c., is rendered peculiarly difficult in *Petromyzon* by the fact that the longitudinal axis of the embryo in early stages describes a semi-circle. Some sections in a series of cross-sections of such an embryo are therefore unavoidably cut in planes which meet the longitudinal axis of the embryo in variable degrees of inclination ; consequently a structure stretching in the direction of this axis is cut through obliquely, as, for instance, the neural cord shown in figs. 2 and 3, Pl. xvii. The vertical dimension of the cord is not in reality as long as is represented in these figures. To gather accurate notions of the form, the position, &c., of a given structure, therefore, it is necessary to compare series of sections of two or more embryos of as nearly the same age as possible. Further, the difficulty of observation is greatly increased by the crowd of yolk-granules in cells, especially by their reaction against staining fluids. Certain fluids such as haematoxylin, borax-carmine, &c., either stain diffusely all the parts, or act on the granules more intensely than on the other contents of cells, so that we can not discriminate different kinds of tissues. This difficulty was, however, obviated by employing picro-carmine. The embryos were stained *in toto* in this fluid, decolorized to the proper degree in acid-alcohol, and then washed in 90% alcohol. In the sections of specimens thus prepared, the histological struc-

tures are distinguishable very clearly, being almost entirely discolored in all parts except nuclei which are stained intensely.

I wish here to express my warmest thanks to my former teachers, PROF. MITSUKURI and PROF. IJIMA, for much invaluable advice and for their kindness in looking through the manuscripts and the proof-sheets of this paper.

To avoid confusion the present paper will be divided into two sections, the first of which will contain mere descriptions, while in the second will be given a historical review and conclusions.

I. Descriptive.

A.—The Pronephros.

The youngest stage of the embryo which I have to deal with in the present investigation, is only a little advanced beyond an ellipsoidal gastrula; it is intermediate between Stage I and II of the list given in my first contribution above referred to (fig. 1, A and B, of that article). The head-fold forms a pointed protuberance at one pole of the ellipsoid, while the blastopore is plainly visible at the opposite pole. The prominent neural ridge extends longitudinally from the anterior end of the head-protuberance to the dorsal lip of the blastopore.

For the general relation of the germinal layers at this stage, I refer to fig. 1 which is drawn from the same series as the section represented in fig. 18 of my former work.[1] It represents a transverse section though the dorsal region of an embryo in the stage above described. As seen in this figure, the solid

1) S. Hatta, On the form. of the germ. lay. in Petr.: this Journal, vol. V, 1891.

neural cord is interposed in the median line between the right and left halves of the mesoblast, underneath the epiblast composed by of a single row of columnar cells. The epiblast is always limited with a sharp contour against the mesoblast. The latter consists, on each side, of a dorsal (*d.*), a ventral (*v.*) and a median (*m.*) row of cells. The dorsal row represents the parietal, and the median and ventral rows together form the visceral, layer. Both the visceral and parietal layers of the mesoblast show, in the proximal portion, a regular arrangement of a high columnar epithelium, while distally this arrangement is more or less disturbed.

The above structures are localized in a small portion on the dorsal aspect of the ovoid embryo, the remaining larger part being taken up by yolk-cells compactly loaded with yolk-granules.

In this early stage, we can detect, therefore, neither in the epiblast nor in the mesoblast any structure whatever which is to be regarded as the rudiment of the pronephros.

Period 1.

In the next following stage, which corresponds approximately to the early part of Stage II (*loc. cit.*, fig. 1, B), certain alterations are met with in the mesoblast. The most important of these is its metameric segmentation. This process first begins at the neck[1] and proceeds both forwards and backwards. At the present stage, there are found 16 or more mesoblastic somites.[2] At about the time when this process has extended to the anteriormost section of the mesoblast a second change arises in the mesoblast, *viz.* the first appearance of the pronephros.

1) The term *neck* is used for the sake of convenience to designate the slender region where the head-fold passes over into the hind globular part.

2) The exact number of the somites can not be reckoned, for the metameres become indistinct posteriorly.

Fig. 2 represents a section through the middle of the fifth somite[1] of the embryo mentioned above. The general features of the germinal layers and of other primitive organs are essentially the same as before. The epiblast (*ep.*) is a single row of columnar cells and is sharply bounded from the structures beneath it; the neural cord (*n.*) remains still solid.[2] In the mesoblast, however, two portions are distinguishable: the proximal portion composed of high columnar cells (*mt.V* and *a.pn.2*) which undergoes metameric segmentation, and the distal portion consisting of a loose group of somewhat irregularly shaped cells (*lm.*) which remains unsegmented and constitutes the lateral plate. It is noteworthy that the former takes up the largest portion of the mesoblast, while the latter is represented by a small portion; these two portions represent respectively the parts of the same name in the mesoblast of *Amphioxus*. However, between them there exists no distinct limit in the lamprey; the one passes gradually over into the other. Although the visceral layer shows no sign of constriction, the parietal layer is notched at about the middle of the proximal segmented portion (*x*). The parietal layer distal to this notch is composed of a regular cylindrical epithelium (*a.pn.2*), which is slightly arched against the epiblast, so as to cause an indentation in the latter, while the visceral layer of the corresponding portion consists of a more or less disturbed row of high columnar cells. As the subsequent history teaches, the proximal half of this extent (*mt.V*) represents the myotome[3]

1) The somites are reckoned from the anterior end. The first, *i.e.* the foremost lies immediately behind the auditory vescicle when the vescicle comes into view.

2) The vertical diameter of the neural cord in figs. 2 and 3 is shown greater than it really is, the sections passing obliquely owing to the bending of the longitudinal axis of the embryo, as noted in the introduction (p. 312).

3) This term here means the Sclero-myotom of German authors.

and the distal portion (*a.pn.2*) constitutes the *Anlage of the pronephros*—the name I assigned to the same in my preliminary paper ('97). To avoid tiresome reiteration, I shall often speak of them in the following pages simply as the " Anlage " and when it is necessary to refer to special ones, as the Anlage first, the Anlage second, etc. in the order of their position in the series of mesoblastic somites, beginning from the anterior end.

In the somite next following, *i.e.* the sixth (fig. 5), the mesoblast shows almost the same condition as that already described; but in the somite preceding the fifth, *i.e.* in the fourth, the Anlage of the pronephros is a little more advanced in development (fig. 3). On the left side of fig. 3, the fourth myotome is sliced only at its hind wall (*mt.IV*), while, on the right, it is cut through in the middle (*mt.IV*). On the right half of the section, no marked progress is visible in the mesoblast except the separation of the myotome, which shows a pentagonal outline (*mt.IV*) and consists of high cylindrical cells from the lateral plate formed of a loose mass of cells (*lm.*). In the left half, however, the state of things is quite otherwise: the Anlage of the pronephros (*a.pn. 1*) together with the corresponding visceral layer is entirely constricted off from the myotome (*mt.IV*), although it is still connected with the lateral plate (*lm.*). The cells composing the Anlage are compactly set together and arranged more or less in a radial manner; the Anlage itself is rounded off at the proximal end. The lateral plate, on the other hand, still consists of loosely grouped cells of variable shape.

The Anlage is thus always (before and after its separation from the myotome) histologically very distinct from the lateral plate; one might therefore often be misled to suppose that there is no organic connection between these two structures.

Fig. 4 represents a section passing between the two somites above mentioned (the fourth and fifth) and is much magnified (Zeiss, $E \times 2$) to illustrate the finer structure of this portion. The structural cells are all loaded with an enormous quantity of ovoid corpuscles or yolk-granules. The epiblast (*ep.*) consists of a single row of cubical cells and shows a sharp limit against the structures inside it. The irregularly polygonal mass of cells (*mt.V*) is the anterior wall of the fifth myotome. Two rows of variously shaped cells (*lm.*) constitute the lateral plate which is histologically quite like that in the somitic portion, being composed of irregularly quadratic cells and tapering towards the distal (ventral) extremity (compare with the lateral plate, *lm.*, in figs. 2 and 3). However, in the proximal portion, where the Anlage of the pronephros consisting of a regular row of tall columnar cells would be found in the somitic portion, we see here a group (x) of a few cells of faint appearance, forming the proximal edge of the lateral plate. By a comparative study of two or more series of sections, it is easily demonstrated that these cells are a piece of the somite lying in front and have nothing to do with the Anlagen. To elucidate this point still further, I have drawn fig. 7 which represents a section through the intersomitic plane between the sixth (fig. 5) and the seventh somite (fig. 6). In this part the Anlage of the pronephros is developed still more weakly, and the mesoblast remains in a more primitive state. In the proximal edge of the lateral plate (x), no special structure is detected, but the edge fades away without a distinct limit. By comparison with Fig. 4, we can not find any marked difference; thus, here likewise, *there is no cellular connection between the Anlagen in the two succeeding somites.*

From the fifth somite backwards for 9 or 10 somites, the

mesoblast presents almost the same feature of the Anlage as in the fifth somite mentioned above. Fig. 5 represents a section through the sixth somite, next behind the fifth ; when compared with fig. 2 no marked difference is detected in regard to the structure of the mesoblast. But in some segments the development of the Anlage is somewhat weaker than in others, as seen in fig. 6, which shows a section through the seventh somite ; while in a segment posterior to this somite, we find the Anlage as much pronounced as in the sixth somite. However, generally speaking, the Anlage of the pronephros in an anterior somite develops further than that in a posterior. It must be remembered that the somite in which the Anlage has already become expressed does not pass over suddenly into the somite in which no trace of it is to be seen ; but its development gradually grows less and less distinct from the anterior to the posterior part, until finally no trace of it is perceived.

In the present stage, therefore, the Anlage of the pronephros is detected in more than 4 somites but is completely separated from the myotome only in one segment, viz. the fourth somite[1], *and it has no genetic connection either with the Anlage in the next following somite or with the epiblast ; and it must be noticed that we find the foremost Anlage not exactly beneath the fourth myotome, but always underneath its hind border.*

Figs. 8-17 represent sections through a still older embryo of this stage, having about 20 somites. The epiblast (*ep.*), the neural cord (*n.*), and the chorda (*ch.*) are essentially the same as before. Being cut through somewhat obliquely, the myotomes

1) Such a case is very rare. In most specimens examined, the Anlage separated from the myotome is found in many segments, so that we can hardly decide in which segment the separation takes place first.

on the two sides do not exactly correspond. On the right side of fig. 8, the hind border of the fourth myotome (*mt.IV*) is cut through; the Anlage of the pronephros (*a.pn.1*) presents in section an oval shape, consisting of columnar cells radially arranged and containing a cavity of an irregular form. The histological structure of the Anlage is as compared with that in fig. 3, more or less loose[1], and the Anlage itself is thereby also distended. The lateral plate (*lm.*) shows, however, no marked progress. The left side of this figure and the right of figs. 11 and 12 represent the section through the fifth myotome (*mt.V*) and the Anlage of the pronephros (*a.pn.2*) for that somite. The Anlage presents almost the same development as that just described. The left half of fig. 12 and the right half of fig. 13 shows the sixth somite (*mt.VI*) and the Anlage belonging to it (*a.pn.3*). It can be inferred from the arrangement of its component cells that the Anlage has been just constricted off from the myotome, as is shown by the fact that the cells at the point marked with *x* of the visceral and parietal layers are not yet rearranged to form a continuous layer,—a condition which is observed not infrequently in younger embryos. Fig. 14 shows on the right side a section through the hind wall of the sixth myotome; the Anlage beneath it (*a.pn.3*) is, therefore, the hind part of that represented on the right side of fig. 13: it is entirely cut off from the myotome (*mt.VI*), and the two layers at this point have completely fused together, enclosing a comparatively wide cavity. The same condition is observed in the

1) When the pronephric Anlagen are cut off from the myotome, their structure is at first loosened, that is, their component cells become loosely set together. Later the cells multiply themselves, and are again compressed by mutual pressure; giving a compact structure to the Anlage—probably the same condition observed by VAN WYHE in Selachian embryos ('89, p. 476).

section through the anterior border of the somite. This phase of constriction is doubtless earlier than that shown in fig. 3. The right half of fig. 16 and the left of fig. 14 is from the section through the mid-plane of the next following somite, *i.e.* the seventh. The Anlage of the pronephros (*a.pn.4*) is not yet cut off from the myotome (*mt.VII*), but the process is beginning as shown by a slight constriction and an inclination to arch out, while the suddenly weakened lateral plate (*lm.*) forms the distal (ventral) continuation of it. This feature of the mesoblast reminds us of the youngest stage of the Anlage described above (compare with figs. 2 and 5). In the section passing through the anterior or the posterior border of the somite too, the same condition of the Anlage, as on the right side of fig. 14, is observed.

From the facts mentioned above, it is easily understood that the separation of an Anlage from a myotome begins with the constriction which takes place at the anterior and the posterior border of the somite, and that the middle portion is the last to be cut off, so that the cavity of the myotome (myocoelome) communicates with the peritoneal cavity, during some time, through a narrow passage at the middle point.

Myotomes when cut off from the Anlage of the pronephros assume a pentagonal form (see fig. 3) constructed of a dorsal, a lateral, a ventral, and two median sides, each of which is composed of a regular row of tall cylindrical cells. The dorsal and lateral rows of cells constitute the parietal layer of the myotome, while the three other sides represent the visceral layer (compare with the description on p. 314).

For about ten segments behind the seventh somite, the Anlage of the pronephros shows the same condition as that seen on the right side of fig. 16. Fig. 17 represents a section

through the twelfth somite; we can find no marked difference between the Anlage in the seventh somite and in this. The segments lying still farther backwards are not cut through exactly transversely in this same series of sections, owing to the cause stated above (pp. 312 and 315), so that we can not trace the differentiation of the mesoblast from the anterior to the posterior part in this one series. But I could demonstrate from several other series of sections that the Anlage of the pronephros is, in the present stage, found in no less than 15 somites.

Figs. 9-11 represent the contiguous sections through the intersomitic portion, on the right side, between the first and second Anlagen, *i.e.* between that of the fourth, and that of the fifth, somite. Fig. 9 is from the section next behind that shown in fig. 8; the portion (*cd.*) lying proximal to the lateral plate (*lm.*) presents no longer a weak appearance as in younger embryos (see the statement on p. 317 and figs. 4 and 7), but is occupied by a compact cellular structure (*cd.*) which suddenly passes over into the loosely composed lateral plate (*lm.*). Fig. 10 is from the section next posterior to fig. 9 and next anterior to the second Anlage represented in fig. 11 and shows almost the same condition as in fig. 9, with respect to the structure in the proximal portion of the lateral plate. In other words, in the intersomitic portion between the first and second Anlagen, a cellular cord has become established, which connects these two Anlagen. It is this cord which gives rise to the collecting duct or *Sammelrohr* of RÜCKERT ('88), putting all the pronephric tubules in communication.

On the left side of figs. 9, 10, and 11, the contiguous sections through the intersomitic portion between the Anlagen second and third, are represented. In figs. 9 and 11, the condition of

the structure (*cd.*) at the proximal portion of the lateral plate is almost the same as that on the right side just described, although it is here somewhat weaker in development than there. The section represented in fig. 10 intervenes between the two mentioned above; in this section, the structure in question (*cd.*) is weakest in development, consisting of four or five cells only. In the left half of fig. 15 which represents the section through the intersomitic portion between the sixth and seventh somites, there is found no structure to be compared with the cord mentioned above, the proximal edge of the lateral plate (x) being of the same condition as that in figs. 4 and 7. *In fact, the cord appears after the complete separation of the Anlage from the myotome, and when it is first established, the nearer the plane of a section to the Anlage either anterior or posterior, the thicker the cord.* For instance, of the above three sections (the left side of Figs. 9-11), the middle (fig. 10) is the weakest. But this unequal development of the cord is soon made even by its growth as seen in the case of the cord between the Anlagen first and second (the right side of figs. 9 and 10).

The history of this cord as given above shows that it has doubtless the genetic relation with the Anlage of the pronephros. In early stages, no such structure is found in the intersomitic portion, but it becomes established one after the other with the development of the Anlagen. The cord is in section, thickest near the Anlage and weakest in the midway between two consecutive Anlagen, when it is first established. These facts give naturally an impression that it is growing out of the two consecutive Anlagen backwards and forwards and these two growing ends meet at some point in the midway between these two Anlagen, finally to fuse together. This point of meeting is, I think, indicated by

the part where the duct has been described above as weakest. It is also the fact that repeated cell-multiplication takes place at the outer rim of each Anlage of the pronephros. One might suppose that the product of the cell-division would contribute only to the growth of the Anlage itself and has nothing to do with the cord; but this is not the case: the Anlage does not grow at the outer (lateral) end, as it might seem, but by cell-division within its own structure. I have never observed any case of cell-proliferation along the dorsal edge of the lateral plate in an intersomitic portion, although the cords appear, in later stages, to have some connection with that edge, when they are fully established (see the right side of figs. 9 and 10); this connection thus is not primary, but secondary. The epiblast has, from the first, no share in the formation of the cord, always showing a sharp contour against the mesoblast below.

There is thus no difficulty in accepting the view that the connecting cord is formed of the intersomitic cell-outgrowths which are budded out of the anterior and posterior rims of each Anlage of the pronephros and are subsequently fused together. The cord is, therefore, originally brought about by the confluence of the free extremities of the Anlagen.

Further development of the Anlage of the pronephros may be intelligible by refering to fig. 18 which represents a section through a little older embryo of Stage II. The epiblast (*ep.*) consists of a single layer of cubical cells as before; the neural cord (*n.*) is still solid. On the left side of the figure, the hind border of the fourth myotome (*mt.IV*) is cut, while on the right, the mid-plane of the fifth myotome is met with. A comparison with the corresponding parts in the younger stages (figs. 3 and 8) will

plainly demonstrate a progressive change undergone by the pronephric Anlage. The Anlage on the right side (*a.pn.2*) presents a feature much like that seen in fig. 3, notwithstanding some points of progress. The Anlage on the left side (*a.pn.1*), however, shows a considerable progress; it has become much more compact by the active multiplication of its component cells. Owing to mutual pressure, the cells are compressed and their nuclei are regularly arranged, describing together an ellipsoidal figure. The inside of the ellipse encloses a comparatively large lumen, which is standing in connection with the body-cavity represented, at the present stage, only by the boundary line of the parietal and the visceral layer of the lateral plate (*lm.*).

In a little more advanced embryo, the cross-sections of which are represented in figs. 20-31, the neural cord (*n.*), the myotomes (*mt.*), and the Anlage of the pronephros show some progress as compared with those described in the preceding pages. The epiblast (*ep.*) consists, as in the embryo just described, of a single layer of cubical cells and is limited by a sharp line against the structures below. The component cells of the neural cord[1] become arranged in two layers, leaving, in the anterior section of the cord, a vertical fissure-like lumen in the median line of the cord, which represents the beginning of the central canal (figs. 20-21, &c.). The posterior part of the cord is still solid, although the position of the central canal is marked by a vertical line produced by cell-boundaries (fig. 26) just as described in the foregoing pages. The myotomes are, in the anterior region, likewise enlarged, probably owing partly to multiplication of component cells and partly to

1) Owing to the same cause as the sections represented in Figs. 2 and 3, the vertical diameter of the neural cord in Figs. 20-23 is shown somewhat longer than it is in reality.

the loosening of the composition of the tissue[1] and assume the shape of a scalene triangle (figs. 20-23); the median side of the triangle (*mus.*) represents the visceral, and the two other sides (*cut.*) the parietal, layer of the myotome. In the posterior region, they are yet of a compact structure of a pentagonal form, enclosing a cavity (figs. 25-31, *mt.VII-X*).

The anteriormost Anlage of the pronephros is found as before under the hind part of the fourth myotome, the section of which is represented in fig. 20 (*a.pn.1*). It shows a considerable development: the component cells, which are of high columnar character are no longer compressed, but the tissue is more or less loosened. Thus the Anlage itself is distended, and its upper (dorsal) angle becomes acute and grows in between the epiblast and the myotome. The internal cavity of the Anlage also becomes conspicuous. The Anlage of the pronephros under the next posterior myotome (the fifth) is not so advanced as in the last somite (the fourth). In fig. 22 is shown the section through the hind part of this somite and of the pronephric Anlage belonging to it (*mt.V* and *a.pn.2*), a section through the mid-plane being unfortunately wanting in this series of sections. The next posterior Anlage is found just under the sixth myotome and represented in fig. 24 (*a.pn.3*) together with the hind border of the myotome (*mt.VI*). The Anlage shows a compact structure which is probably due to a rapid multiplication of the constituent cells. The next following Anlage of the pronephros is found beneath the seventh myotome (fig. 26, *a.pn.4*). It shows no further development than the separation of it from the myotome and the fusion at the retrenched ends of the two layers of mesoblast: it is in the same stage of constriction as that in the

1) See the foot note in p. 319.

right of fig. 13 which represents the Anlage third in a younger embryo.

The Anlagen above referred to are connected with each other by the solid connecting cords (figs. 21, 23, and 25, *cd.*), which are found between these Anlagen just as described in the younger stages. Of these connecting cords, that between the Anlagen second and third (fig. 23, *cd.*) is the thickest, while that between the Anlagen third and fourth (fig. 25, *cd.*) is the weakest in development, owing probably to its having been just established. The cord between the Anlagen first and second (fig. 21, *cd.*), however, is rather weaker as compared with that in the next posterior intersomitic plane (fig. 23, *cd.*). Such a case is occasionally met with; but this is doubtless not normal; in most cases examined, the cord is thickest in anterior segment and decreases in thickness gradually posteriorly as seen in the last example (see pp. 321-322 and figs. 9-11).

The Anlage of the pronephros belonging to the eighth somite and that of the ninth somite are not completely cut off from the myotome to which these respectively belong (figs. 28 and 30). They are, however, constricted already at the anterior and posterior borders of the segment: fig. 27 shows the posterior part of the Anlage fifth, and fig. 29 represents the anterior part of the Anlage sixth respectively. Such a case is observed in the younger stage described in the foregoing pages (pp. 319-320). Compare these two figures with the right half of Fig. 14 and the description on p. 319: here the phase of constriction is a little more advanced than there. The anterior part of the Anlage fifth and the posterior part of the Anlage sixth, the figures of which are omitted, have features much like those seen in figs. 27 and 29. In these two segments, the central portion of the Anlage is

just in the process of being constricted off from the myotome, and we can not decide by this case alone which segment (whether the anterior or the posterior) is the further developed; a comparative study of other examples shows that the separation in the posterior segment follows that in the anterior. The state of the mesoblast in the next posterior segment, *i.e.*, the tenth segment (fig. 31), is quite different from that just described; it is in a more primitive condition of development. The Anlage of the pronephros (*a.sd.*) presents only an indication of constriction,—a feature which we have observed repeatedly in embryos of younger stages (compare with figs. 2, 5, 6, 14, and 16). From this segment backwards, a few segments show almost the same condition. Still further posteriorly, the structure of the mesoblast can not be readily observed, since the planes of sections incline by degrees in the cranio-caudal direction, owing, as above stated, to the bending of the longitudinal axis of the embryo.

In all the segments mentioned above, the lateral plate (*lm.*) consists of a loose tissue of cells of variable shape, and the Anlage passes over suddenly into the lateral plate just as in the embryos described in the foregoing pages.

In this stage, therefore, the Anlage of the pronephros is completely separated from the myotome in 4 somites, i.e., from the fourth to the seventh inclusive; and these are connected with one another by the intersomitic solid cord. In the following 4 or 5 somites, the constriction is just going on, while in a few of still more posterior somites it is indicated merely by a slight depression in the parietal layer of the mesoblast.

Period 2.

In the embryos which belong to Stage III, we observe a decided advance in several respects. Figs. 32-50 represent a series

of cross-sections through one of these embryos which has about 25 somites. The neural cord (*n.*) is reduced in size and in the anterior part has a conspicuous canal. The myotomes which showed before a pentagonal outline, in the anterior part of the body have now assumed an elongated lozenge-shape[1] and is composed of an outer, and an inner, layer of long cells which have begun to diffentiate themselves. The inner layer (*mus.*) represents the median and ventral rows of the pentagonal myotome mentioned on p. 320 and, therefore, corresponds to the visceral layer of the myotome; the outer layer (*cut.*) is the product of the dorsal and lateral layers and constitutes the parietal layer. The pronephric Anlage is composed of high columnar cells which are plainly distinguishable from the much shorter elements of the lateral plate (*lm.*). The component cells of the Anlage of the pronephros which we generally found to be compressed in the foregoing stage (pp. 324 and 325), are now more or less loose, and the internal cavity of the Anlage is somewhat widened, being distended by the loosening of the cells.[2]

The peritoneal cavity is, at the present stage, still represented merely by the boundary-line of the parietal and visceral layers of the lateral plate.

In the present stage, the foremost Anlage of the pronephros is, as before, found under the hind part of the fourth myotome (figs. 32-35, *a.pn.1*). The Anlage shows, in section, a circular outline and is composed of high columnar cells arranged in a radial manner. The internal cavity of it is confined no longer to one section, but it is observed in three or more sections; it is most spacious in the hind part of the fourth somite (fig. 33) or in that part where the cavity is visible from an early period.

1) A few myotomes in the anterior somites tend to assume this shape already in the last stage (see figs. 20, 21, and 22).

2) See the foot-note on p. 319.

From this part backwards it gradually decreases in width until no space is perceptible. Anteriorly the cavity is also somewhat narrowed, but not as much as in its posterior continuation, and ends blindly rather suddenly at its anterior end (fig. 32). The anterior portion of the Anlage forms a blunt conical tube (Fig. 32, *a.pn.1*) projecting anteriorly and lying between the dorsal edge of the lateral plate (*lm.*) and the lower surface of the fourth myotome (*mt.IV*). The existence of this conical tube[1] gives us a strong impression that originally there must have been present an Anlage of the pronephros in the anterior segment which was connected by a connecting cord with the Anlage belonging to the fourth somite, but had disappeared during the phylogeny and that this conical tube is the remnant of this connecting cord.[2]

The next posterior Anlage, which is found under the fifth myotome (figs. 37-39, *a.pn.2*) and shows an outline much resembling that represented in figs. 32-35, has an internal cavity of irregularly triangular form, extending through three sections, of which the foremost section contains the most spacious cavity, while in the others the lumen grows smaller and smaller. The pronephric Anlage in the next following somite (figs. 41-43, *a.pn.3*) has an outline much like that shown on the left side of Figs. 18 and 24, being in the same phase of development, that is, it is of the form of an isosceles triangle whose two basal angles touch the myotomes. This Anlage is found under the sixth

1) The internal cavity of this conical tube is not entirely closed, but there is clearly seen a small canal (*x*) directed towards the median side and opening below the myotome. I can not decide, at present, whether this canal is normal or abnormal; for I can not make out the corresponding structure on the opposite side and have no other embryo of exactly the same stage, in which the structure in question would probably be found, if it be of some definite meaning; I also can not detect any trace of such a canal in embryos of advanced or younger stages.

2) See the description under Period. 4.

myotome and contains the internal cavity extending likewise for three sections, of which however the hindmost contains the widest cavity, while it is diminished in width anteriorly : in other words, the width of the cavity enlarges in inverse direction as compared with that in the preceding two somites. The Anlage fourth under the seventh myotome (figs. 45 and 46, *a.pn. 4*) has an oval outline like that shown in fig. 26 and encloses an internal cavity, which covers two sections and is anteriorly wide and posteriorly narrow. The two following Anlagen which are detected under the eighth and ninth myotomes respectively (fig. 48, *a.pn.5* and fig. 50, *a.pn. 6*) show almost the same condition of development as in the somite just described ; the internal cavity which they contain is likewise extended into two sections ; the width of the cavity is about the same in these two sections, being of a fissure-like form.

The solid cord which is observed in the embryos of the last stage connecting the Anlagen with one another, is also found here. The cord in the intersomitic plane between the Anlagen first and second (fig. 36, *cd.*), that between the Anlagen second and third (fig. 40, *cd.*), and that between the Anlagen third and fourth (fig. 44, *cd.*) are all comparatively short, so that they are in each stretch confined to only one section, while that in the two posterior intersomitic planes, *i.e.* between the Anlagen fourth and fifth, and between the Anlagen fifth and sixth, the cord is extended in each case into four sections. In this latter part, the cord is in a primitive condition ; the component cells are actively multiplying. Hence these four sections all show similar features. I have endeavoured to show in fig. 47 one of these sections which is taken from one of the four sections between

the Anlagen fourth and fifth, and in fig. 49, one between the Anlagen fifth and sixth.

This inequality in the length of the intersomitic solid cord is, I believe, due to differences in the degree to which the canalization within the Anlage has extended into the connecting cord. In the anterior section of the pronephros, this process has already proceeded to some extent into the interior of this cord, while in the posterior, the cavity is still confined entirely within the Anlage itself. The whole system of the pronephros at the present condition may be compared to a bamboo-cane with nodes and internodes; in the anterior section of the system, the nodal septum has become very thin, while it has a considerable thickness in the the posterior. As will be shown further on, all these septa entirely disappear later when the collecting duct is fully established.

From the fact mentioned above, it will be easily seen that the process of canalization in the pronephric system of *Petromyzon* begins in the internal cavity of the pronephric Anlage in each segment and is extended into the intersomitic connecting cord. The direction in which this process proceeds seems, generally speaking, to be from the anterior section to the posterior; for in most cases, not only the internal cavity in each Anlage is spacious anteriorly and narrowed posteriorly, but the cavity in anterior somites is extended more, or canalization goes on further, than in the posterior section of the system; although the progress in the opposite direction is occasionally met with.

From the tenth somite backwards, five or six segments show the same condition of the mesoblast as in the eighth and ninth somites, after which the series can not be studied, owing to the inclination of the planes of sections, referred to above.

In all the segments above referred to, the lateral plate of the mesoblast shows the same condition as in the foregoing stages, but has become more distinct from the Anlage of the pronephros.

In the present stage of development, then, the Anlage of the pronephros is cut off from the myotome in more than 10 segments, and the canalization has advanced in the anterior section of the system, to a state just ready to put the Anlagen in the succeeding somites in communication with one another, although the inter-somitic connecting duct in the posterior part remains still solid.

Figs. 51-58 were drawn from a series of sections through one of the older embryos in this stage. The internal structures are developed much more than in the embryo just described. The cells forming the visceral layer of the myotome have been differentiated into the muscle-plates, while the parietal layer is composed of cubical cells. The Anlagen of the pronephros have acquired, in most cases, a tubular structure and have grown dorsally, being folded out from the body-cavity ; I will accordingly call them the pronephric tubules.

On the right side of fig. 51, the foremost tubule (*pt. 1*) is visible, which no longer contains the internal cavity but is converted into a solid mass of cells occupying the space beneath the fourth myotome. This consolidation is not due to retrogressive changes, but is effected by very active cell-multiplication which takes place within the tissue. The cross-section of the collecting duct seen on the left side of fig. 52 (*cd.*) which represents the third section behind the last, is likewise solid. The tubule on the right side of this figure (*pt.2*) and that on the left side of the third section posterior to it (fig. 53, *pt.2*) are respectively the second tubule of the right and left side found

under the fifth myotome; both are of a triangular form and contain a very spacious internal cavity of the same shape. On the right side of fig. 53, the sixth myotome and the third tubule are shown. The section next posterior to fig. 53 (fig. 54) shows the cross-section of the collecting duct (*cd.*) on the right side and a slice of the hind wall of the second tubule on the left (*pt.2*). The cells composing the duct are closely set together, although arranged more or less radially, acquiring a tubular form. As has been repeatedly mentioned above, the epiblast is, as in the foregoing stage, marked off from the mesoblast as well as from the Anlage; but at the present stage, the second tubule (figs. 54, *pt.2*) pushes against the epiblast, probably in consequence of an enormous multiplication of its component cells, so as to cause the latter to be a little elevated externally. It must be remarked here that the Anlagen, especially the first and the second, when they first assume the tubular form, are brought into an intimate relation with the epiblast, striking against it. In some of my sections, a mitotic figure is seen at that point of the epiblast[1] (fig. 54, *x*). This might lead some to assume a genetic connection between the epiblast and the pronephros in *Petronyzon*; but there is, I believe, in reality no such relation. If the epiblast gives some cells destined to build the pronephros or a part of it, cell-proliferation or some other mode of cell-production would necessarily be observed in the epiblast in the preceding stages or at least, in the stage here spoken of. In the foregoing stages, the epiblast had, as has been repeatedly mentioned above, a sharp limit against the structures inside it. At the present stages also, it is marked off by the boundary-line of cells from the tissue of the tubule,

1) In the series of sections, from which fig. 54 is drawn, I observe mitotic figures at that point in several sections.

showing no structural alteration. Mitotic figures are met with not infrequently in that part of the epiblast (fig. 54, *x*); their axis lies, however, in all the cases examined parallel to the plane of the epiblast, giving us an impression of the resulting cells contributing to the formation of no other part than the epiblast itself; on the contrary, within the structure of the tubule the cells are rapidly multiplying (figs. 51, *pt.1* and fig. 54, *pt.2*), showing that the growth of the tubule is actively going on. In fact, the connection, or rather the intimate contact, of the pronephric tubule with the epiblast is a temporary condition; the separation follows immediately afterwards, and the tubule returns soon into a state similar to that seen in fig. 52 (*pt. 2*).

According to RÜCKERT ('88), a similar case is observed in Selachian embryos: the tubules become connected *secondarily* with the epiblast—what caused him to believe that the latter might give some constituent elements to the tubules.

The third section behind that represented in fig. 54 (fig. 55) shows, on the right side, the fourth (*pt.4*) and, on the left, the third tubule (*pt.3*) respectively. The latter is not so far developed as its counterpart on the opposite side (fig. 53, *pt.3*), while the former presents a great progress: it consists of a definite epithelium and contains a distinct cavity of triangular shape, although the corresponding tubule on the opposite side (fig. 56, *pt.4*) which is found in the third section behind the last, is much less advanced in development. The fifth tubule, the tubule on the right side of fig. 56 (*pt.5*), is somewhat more developed than that which belongs to the anterior somite (the fourth tubule on the opposite side); but it has a feature much resembling the fourth tubule on the same side (fig. 55, *pt.4*) and the second on the

opposite side (fig. 53, *pt.2*). In short, in this series of sections, the tubules on the right side, are all more advanced than those on the opposite side. The sixth is very primitive in development; fig. 57 represents the section, on the left side, through the anterior part of the ninth somite and, on the right, the posterior part of it. The left tubule is sliced at its anterior wall, but the right tubule is cut through in its mid-plane. It is composed of two layers of columnar cells, but no cavity has yet appeared in the interior.

From the tenth somite backwards, the Anlagen are cut off from both the myotomes and the lateral plate, and constitute *the segmental duct* or the posterior continuation of the collecting duct, which is distinctly traceable for 7-8 somites. Not infrequently, however, a somite is met with, in which the segmental duct is not yet cut off from the lateral plate at the time when the separation is finished in a majority of somites, as seen in fig. 58 which represents a section through the twelfth somite. The left half of the figure shows the duct entirely cut off from the lateral plate, while the right exhibits the state not yet separated. The same structure is made out in two contiguous sections, so that one might mistake it for a pronephric tubule. This point will be described further on.

The relation of the pronephric tubule and the peritoneal cavity is not so simple as in the last specimen; besides the pronephic tubule, there is seen another structure which projects out of the inner angle of the peritoneal cavity (figs. 52, 53, 55, and 56, *c.p.*). This projection is originally a fold of the peritoneal wall and gives rise, as subsequent history shows, to the radix of the mesentery, whence the gonads and the mesonephric tubules are derived. It will here be called briefly the "*coelomic projection.*"

At that point of the visceral layer of the mesoblast, where the Anlage of the pronephric tubule passes over to the lateral plate, it is always many cells deep (figs. 55 and 56, *c.p.*), and the projection in question is brought about by repeated division of these cells. The projection formed is consequently seen in each somite and thus shows a *segmental arrangement.* Its component cells are soon re-arranged into an epithelium, and the pouches thus formed push their way between the myotome and the hypoblast.

The coelomic projection appears, at first sight, to be homologous with the coelomic pocket described by PRICE ('97) in *Bdellostoma.* The coelomic pocket is, however, according to PRICE, the product of both the parietal and visceral layers of the lateral plate and is afterwards converted into the cavity between the glomerulus and BOWMAN'S capsule of the Malpighian corpuscle; the floor of the pocket forms BOWMAN'S capsule, and its roof together with a part of the pronephric tubule is transformed into the cover of the glomerulus ('97, p. 213). The coelomic projection in *Petromyzon* is, on the contrary, formed out of the visceral layer of the distal half of the somite and gives rise, as just stated, to the radix of the mesentery, from which partly the mesonephric tubule and partly the gonads are formed.[1]

Figs. 59-63 are from a series of sections through an older embryo of the same stage. In this series of sections, a further development of the coelomic projections is clearly seen; the first figure (fig. 59) shows the section through the second tubule, fig. 60 through the third, and so forth. In the first 3 figures and on the right of fig. 62, the coelomic projection (*c.p.*) presents an

1) I will not here further discuss this structure, as I intend to do so in a future paper in which the development of the mesonephros in *Petromyzon* will be delt with.

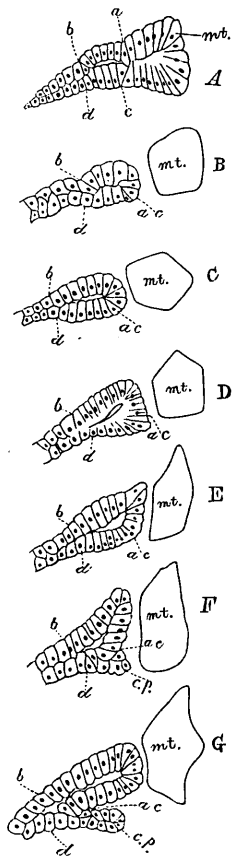
epithelial structure, forming the continuation of the peritoneum and folding out from the peritoneal cavity. Beneath the first tubule, there is found no rudiment of the projection; under the second (fig. 59) it is very weak, while beneath the third (fig. 60), fourth (fig. 61), and fifth (fig. 62), tubule, respectively it is most vigorously developed. But on the left side of figs. 61 and 62 it is again in a primitive condition, just as in the last series of sections (figs. 52, 53, 55, and 56).

The coelomic projections are not confined to the anterior region where the pronephric tubules are found, but it is found likewise in the posterior part where only the segmental duct develops. Fig. 63 shows the section through the thirteenth somite; on this section, the duct is cut off from the myotome and a well developed coelomic projection (*c.p.*) is observed; I will return once more to this subject further on.

Leaving the coelomic projection in this stage of development, I will return to the origin of the Anlage of the pronephros and give somewhat more exact details on the subject. Since the piece of the mesoblast called above the Anlage of the pronephros forms for a time the proximal portion of the lateral plate, one might presume that its whole mass will be transformed into the pronephric tubule and will not partake in the formation of the peritoneal membrane. I was at first of this opinion, but a careful observation of sections through the embryos in each stage showed my error.

To illustrate this point satisfactorily, I have given, in the annexed wood-cut (Wood-cut 1), a series of semi-diagramatic figures, which show the successive phases of the changes going on in the structure. A shows the first indication of the Anlage of the pronephros before the separation of it from the myotome;

a-b indicates the extent of the Anlage; *c-d* shows that of the coelomic projection. When the myotome is cut off, the point of the parietal layer indicated by *a* becomes fused with the point *c* of the visceral layer (*B*, *ac*). This piece of the mesoblast assumes an ellipsoidal shape (*C*). The component cells of this ellipsoid are multiplied by active cell-divisions, and the piece almost loses its lumen and gets a compact consistence (*D*). Meanwhile the cells in the space *d-c* remain inactive. Consequently the piece acquires a triangular form (*E*), whose upper sharp angle, together with the two sides enclosing this angle, gives rise to the pronephric tubule. The lower (median) obtuse angle now begins to grow by cell-mulplication (*F*) and produces a small knob (*F*, *c.p.*), which grows further and pushes in between the myotome and the hypoblast (the upper wall of the enteric canal). This cellular projection is that which has been called the coelomic projection. It is reduced into a thin plate of epithelial cells (*G*, *c.p.*) and assumes then the form of



Wood-cut 1.—Semidiagramatic figures to illustrate the successive phases of the evolution of the nephrotome.

A. from the right side of fig. 16.
B. from fig. 3.
C. from the right side fig. 18.
D. from the left side of the same.
E. from the left side of fig. 53.
F. from the right side of fig. 55.
G. from the left side of fig. 61.

a true fold of the visceral layer of the lateral plate. At the same time, the upper angle or the pronephric angle develops further and assumes a tubular form composed of a single layer of columnar cells (*G*).

The peritoneal cavity begins, therefore, at the point, from which the coelomic projection starts, and the part of the layer dorsal to this point is all appropriated to the formation of the pronephric tubule. The nephrostome will be found, therefore, by the point where the tubule passes over to the projection.

I will add a few words on the differentiation of the myotome, so far as concerns the topographical relation of it to the Anlage of the pronephros. The myotome consists, at the present stage (Stage III), of the inner and outer layers which constitute respectively the *Muskelblatt* and the *Cutisblatt* of German authors (fig. 59 and 60, *mus.* and *cut.*). The cells composing the *Muskelblatt* (*mus.*) are, simply differentiated into a transverse row of the muscle-plates. The outer layer (*cut.*) undergoes, however, subsequently a series of interesting changes: it folds in, just as the *Sklerablatt* or sclerotome described by HATCHEK in *Amphioxus* ('88) between the *Muskelblatt* and the chorda and the neural tube.[1] As is well known, RABL ('88) has homologised HATCHEK's *Sklerablatt* with his *Sclerotomdivertikel* of Selachian embryos, which is the evagination of the ventral part of the visceral layer of the mesoblastic somite. This part of the somite (the selerotome) corresponds, I believe, exactly to the ventral row of the pentagonal myotome in my embryo (see pp. 314 and 320), which comes afterwards to form the ventral part of the cutis-layer (see figs. 21, 22, 23, 36, 37, 43, 49, 59, 60, &c.). When the myotome is not yet separated from the rest of the mesoblast (fig. 2), this part of the

1) This subject will be treated of in an independent article.

selerotomic layer forms, as has been seen above, a direct continuation of the visceral layer giving rise to the coelomic projection. (The successive changes of the myotome are seen in figs. 1, 2, 3, 18, 21, 22, 43, 49, 60, &c.)

From the above account, it can be inferred that the ventral half of the mesoblastic somite in *Petromyzon*, which gives rise to the pronephic Anlage and the coelomic projection, is doubtless homologous with the "*intermediate cell-mass*" of BALFOUR described by him in Selachia and, therefore exactly coincides with the "*Nephrotom*" of RÜCKERT.[1] So far as concerns its future destination, however, the results arrived at by me slightly deviate from their views.

A series of sections through the oldest embryo of this stage is represented in figs. 64-76. The epiblast has undergone no histological change, but remains, as before, one cell deep. Many structures, however, exhibit a remarkable progress. The muscle-layer (*mus.*) of the myotome is, for instance, further differentiated, now consisting of a transverse row of long muscle-cells, although the cutis-layer (*cut.*) is still composed of short cubical cells. In the anterior region, the true coelome (*pp.c*), becomes conspicuous enclosed by the parietal (*m.p.*) and the visceral (*m.v.*) layers of the lateral plate, both of which consist of a single row of cubical cells. The ventral edges of the lateral plates on both sides do not, however, yet meet in the ventral median line. The walls of the enteric canal too are, in the anterior region, reduced into a single cell layer.

A great alteration is met with in the pronephric tubules. They have assumed a cylindrical form composed of tall columnar epithelium and have grown dorsally, pushing in between the myotome and the epiblast, causing the latter to be elevated a

1) In spite of the discussion by RÜCKERT ('89, pp. 19-20) on the inexactness of the expression "intermediate cell-mass," I homologise, with many authors, these two terms with each other.

little. The internal lumen of the tubules are put not only in wide communication with the peritoneal cavity, but also in direct continuation with one another through the collecting duct, which consists of a regular columnar epithelium-cells arranged radially and now encloses a conspicuous lumen.

In the foremost of these twelve sections (fig. 64), we notice that a structure (*pt.1*) consisting of a few cells projects at the outer corner of the proximal edge of the lateral plates and lies in contact with the outer wall of the myotome on either side. This structure is found under the anterior border of the fifth myotome and I infer that it is a remnant of the first pair of the pronephric tubules which begins to decline in the present stage. The reason why it is found not under the fourth myotome as in all the stages hitherto described but beneath the anterior border of the fifth myotome, consists probably in its shifting backwards; for we find, in this series of sections, another pair of the tubules under this same fifth myotome. A comparison of this figure with fig. 65 representing the next posterior section will make the matter clear. On the left side of fig. 65, the same remnant structure (*pt.1*) together with the collecting duct (*cd.*), which connects the first and the second tubules can be observed, while the cross-section of the collecting duct in the corresponding intersomitic plane is seen on the opposite side (*cd.*). The next following section is shown in fig. 66; the tubules (pt.2) on both sides communicate freely with the peritoneal cavity; these are found beneath the hind part of the fifth myotome and are the second pair of the tubules; the openings (*nst.2*) to the peritoneal cavity are, therefore, the second nephrostome of the pronephros. The tubule on the left side is weaker than that on the right, since a larger part of the left tubule is visible on the section next posterior which is represented in fig.

67 (*pt.2*). The shape, which the tubules of the second pair (fig. 66, *pt.2*) assume at about this stage, is a characteristic triangle, whose two angles, the one directed dorsally and the other directed medially, are acute and whose outer (lateral) angle is obtuse (see the left side of fig. 59 and the right side of fig. 66, *pt.2*) ; so much so that we can easily determine by this feature the fact of their being the second pair. This peculiar shape of the second tubule is retained for a considerable time as will be seen further on. On the right side of fig. 67, the collecting duct (*cd.*) is cut through transversely ; on the left, the same duct (*cd.*) and the hind part of the second tubule (*pt.* 2) are seen.

At the point where the nephrostome opens to the peritoneal cavity, the visceral peritoneum at the median corner of the latter projects out between the myotome and the hypoblast (figs. 66 and 67, *c.p.*) ; beneath the collecting duct, however, no such structure is detected (see the right side of figs. 65 and 67). Such a pouch is repeated in each nephrotome (see figs. 66-75, *c.p.*) and is what has been called above the coelomic projection.[1]

The next following sections shown in figs. 68 and 69 show the third pair of the tubules (*pt.3*) to be of the same structure. In these two sections the tubules are cut through lengthwise, and the nephrostomes (*nst.3*) on the two sides come into view in symmetrical manner. The tubules are so simple as to need no further explanation. Fig. 70 represents a section through the intersomitic plane between the sixth and the seventh somites, and next posterior to fig. 69. It shows on either side the cross-section of only the collecting duct (*cd.*), consisting of radially arranged cells. Fig. 71 is from a section through the seventh somite and is the third

1) In this series of sections, we often see the coelomic projection on sections passing through the intersomitic plane; but this is the piece of it belonging to either the anterior or the posterior somite.

behind the section shown in fig. 70; the tubules of the fourth pair (*pt.4*) show themselves symmetrically on both sides; they are somewhat less developed as compared with those of the last pair. Fig. 72 is the section next behind fig. 71; it shows on both sides the collecting duct (*cd.*) together with the coelomic projection (*c.p.*) which is a part of that of the anterior segment. Fig. 73 is the third section posterior to that just described; the blastoderm becomes more flattened than in the foregoing sections; it shows on both sides the tubules of the fifth pair (*pt.5*); the condition of the tubules and nephrostomes (*nst.5*) is much like that in fig. 71. Fig. 74 is from the fourth section posterior to fig. 73; the right tubule (*pt.6*) of the sixth pair and its nephrostome (*nst.6*) are visible on the right side, while the collecting duct (*cd.*) is cut through on the left. The sixth tubule and nephrostome on the opposite side are observed in the next anterior section which is not figured. The segments back of the ninth somite have no trace of the tubule, but the cross-sections of the posterior continuation of the collecting duct, the segmental duct, are repeated in each section. Fig. 75 represents a section through the sixteenth somite; the cross-section of the segmental duct (*sd.*) on either side is seen; it always occupies the space where, in the anterior region, the tubules or the collecting duct is found.

This condition, however, is not continued to the dorsal lip of the blastopore. As I have stated in my previous paper ('91), many processes of development are much delayed in the hind region, so that we are here reminded of what were seen in the anterior region of the younger stages. Fig. 76 represents the fifteenth section from the dorsal lip of the blastopore and passes through about the twenty-third somite. In this comparatively late stage, in which many mesoblastic organs have developed in the anterior

region, the neural cord (*n.*) is still solid; the mesoblast (*ms.*) is many-cell-layered and its metameric segmentation is still going on. On the right side of the figure, the section passes through the mid-plane of the myotome showing no sign of its separation from the rest of the mesoblast, while on the left, which shows the intersomitic portion, the process of separation (*mt.* and *lm.*) is going on. On both sides, however, there is no structure that can be recognised as the Anlage of the segmental duct.

The six pairs of pronephric tubules observed in this stage are the maximum number for Petromyzon; this stage ought, therefore, to be regarded as the highest point of development with reference to the pronephros. Even in the present stage, the foremost tubules show a tendency to degenerate.

Period 3.

The embryos of Stage IV, which have about 35 mesoblastic somites, present a remarkable progress. The head-fold is much prolonged; in older embryos of this stage, it begins to twist ('97, fig. 1, *D*). Figs. 77-91 represent sections through one of these embryos. In some myotomes, the sclerotomic fold goes deeper between the muscle-layer and the chorda. The parietal layer of the lateral plate (*m.p.*) is much lessened in thickness, so that it is reduced, in the dorsal region (the posterior two thirds of the pronephric extent), into a thin epithelial lining of the body wall (see figs. 79-85). The coelomic projection is likewise reduced into a thin plate (figs. 82-86, *c.p.*) except in the anterior two segments of the pronephic region, in which it still keeps the characters of the younger stages (figs. 77-81, *c.p.*), only folding in deeper than in the foregoing stages. The visceral layer (*m.v.*) of the

peritoneum still consists of a cubical or rather cylindrical epithelium. The pronephric tubules are, in general, much prolonged and begin to coil in the dorso-lateral direction, so as to cause an elevation in the epiblast. The walls of the tubules consist of a regular row of cylindrical cells, which passes over suddenly into the thin peritoneum (figs. 77-86), except in the region of the second pair of the tubules, where the parietal layer (*m.p.*) of the lateral plate still retains the character of the younger stages, being composed of cylindrical epithelium like the tubules themselves (figs. 77-79). At some regions, even a few mesenchyma-cells (*mch.*) appear,—for instance, beneath the chorda (see figs. 80, 82, and 84), in the median ventral space (see figs. 81 and 82), and also inside the lateral epiblast (see figs. 77 and 80).

Fig. 77 shows a section through the fifth somite and therefore corresponds to fig. 66, which represents the section through the same plane of an embryo at a younger stage. The longitudinal section of the second tubule (*pt.2*), together with the corresponding nephrostome (*nst.2*), is seen on the left side of the figure, greatly resembling the tubules of the same pair in the younger stage (compare with fig. 66). On the right side, the nephrostome (*nst.2*) alone is observed; the tubule proper is to be seen in the two following sections which are represented in figs. 78 and 79.

Beneath the myotome anterior to the one just described, there is found neither a tubule nor any structure that may be regarded as the remnant of it. In the space between the epiblast, the myotome and the lateral plate, however, a few scattered cells (fig. 77, *mch.*) are found. I at first supposed that these might be disconnected component cells of the first pair of tubules; but, as free cells of quite the same character are found

in other places, for instance, in the space between the lateral plate and the epiblast (figs. 79-81, *mch.*), I have been compelled to conclude that they have no genetic relation with the pronephros, but are mesenchymatous cells which are destined to form the blood-vessels and corpuscles.

As the embryo was somewhat twisted, the sections did not pass through the lateral walls of the body in an exactly transverse plane, but unavoidably obliquely, on either side, as the continuous serial sections represented in figs. 79-86 show.

While on the left side of fig. 78 the posterior portion of the second tubule (*pt.2*) is seen, the second nephrostome (*nst.2*) is observed on the right together with a cross-section of a tubular structure (*cd.*). This latter might be taken as a slice of the anterior border of the second tubule, but is, in my opinion, the remnant of the collecting duct which once connected the second tubule with the first and forms, at present, a tubercle in front of the second tubule, the first tubule having disappeared ; for the second tubule on that side is observable in the next following section represented in fig. 79 (*pt.2*) showing its characteristic features stated above (p. 342).

On the left side of fig. 79 and on the right side of fig. 81, we see the anterior half of the third tubule (*pt.3*) and nephrostome (*nst.3*) of each side, their respective posterior half being found on the left side of fig. 80 and on the right side of fig. 82 (*pt.3* and *nst.3*) ; the tubules are bent laterally and dorsally, probably caused by the prolongation of their tubular portion ; for their nephrostomal part and dorsal blind end retain their original position. This is the first step in the convolution of the pronephric tubule.

As seen on the left side of figs. 79 and 80, the tubule (*pt.3*)

of the third pair shows a new character: the dorsal blind end and the nephrostomal portion of the tubule are more or less expanded, while these two portions are united by a slender middle trunk. When compared with the tubule of the same pair on the opposite side represented in figs. 81 and 82 (*pt.3*), this character of the tubule will be understood more clearly: the dorsal expansion is seen in fig. 81, while the nephrostomal widening is observed in fig. 82. The tubules of the following two pairs show the same feature. On the right side of fig. 80, only the collecting duct between the second and third tubules is found. The left tubule of the fourth pair is shown on the left side of figs. 81 and 82 (*pt.4*); fig. 83 shows a cross-section through the collecting duct (*cd.*) between the third and fourth tubules and a slice of the anterior wall of the right tubule (*pt.4*) of the fourth pair which is prolonged and bent like the tubules of the last pair. The fifth pair of tubules is seen on the left half of fig. 84 on one side (*pt.5*) and on the right of fig. 85 on the other (*pt.5*). It is not developed as much as the more anterior pairs, but shows considerable progress as compared with the tubules in fig. 73 which represents the younger stage of the same pair.

These four pairs of the tubules (from the second to the fifth) contain a spacious lumen and stand in wide communication with the peritoneal cavity, which becomes, at the present stage, conspicuous from this region forwards.

In fig. 86 which shows the fifth section behind the section shown in fig. 85, the space on the left side which is occupied, in the more anterior region, by the tubule or the collecting duct, is replaced by the cross-section of a duct (*sd.*) with an oval outline and an ovoid lumen. This is the segmental duct under the tenth myotome (*mt.X*). On the right side of the figure, however,

besides the cross-section of a duct (*cd.*), there is seen a pronephric tubule (*pt.6*), the long axis of which is directed vertically to the inner surface of the epiblast. It is found just beneath the ninth myotome where the sixth tubule should be found. Although the parts of it are also to be seen in two consecutive sections (the one represented in fig. 86 and another preceding it), the communication of its lumen with the collecting duct is not to be found anywhere. In some embryos, the tubule loses the connection with both the body-cavity and the duct. The structure (*pt.6*) in question is, I believe, nothing else than the remnant of the sixth tubule which is in a stage of degeneration, and the duct (*cd.*) is doubtless the collecting duct between the tubules of the fifth and sixth pairs. Compare the segmental duct (*sd.*) on the left side with the collecting duct (*cd.*) on the right side just described ; the latter has a wide circular lumen, whilst in the former it is slender and compressed. This difference of character between these two ducts is noticeable for some time in the younger stages.

To sum up the results obtained in this stage the tubules of the third to the fifth pairs are vigorously developed, while the second is very weak, the sixth retrograding, and the first has entirely disappeared.

In the present stage, a peculiar structure is observed inside the walls of the body-cavity (figs. 77-85, *pp.1-3*). At some points of the peritoneum, a thin plate which consists, in cross-section, of one, two, or three cells, projects from the peritoneal wall into the body-cavity ; it will be called here shortly the "*peritoneal partition.*" A peritoneal outgrowth is found at the level where the coelomic projection passes over to the visceral layer of the lateral plate ; at the same level, another peritoneal outgrowth starts up out of the parietal layer. These two outgrowths meet at midway

and cut off a long chamber from the body-cavity along the openings of the nephrostomes (figs. 77-85, *pp.1*). This longitudinal chamber communicates anteriorly as well as posteriorly with the body-cavity below, which is represented, in those parts, by the boundary of the parietal and visceral layers of the lateral plate. This is the uppermost partition. The second partition is weaker in development and is detected a little more ventrally, projecting likewise from the parietal and the visceral layers of the lateral plate. It is most obvious in the region beneath the third and fourth nephrostomes (81-84, *pp.2*). We find the third partition still more ventrally, which is weakest in development; its extent is almost the same as the second (figs. 81-84, *pp.3*).

These partitions disappear after a short existence; in a little older embryo, none of them is detected, as will further be seen. This is probably the same structure as the "*peritoneale Scheidewände*" or "*Peritonealbrücke*" described by GOETTE in *Petromyzon fluviatilis* ('90). As to the meaning of the structure I have nothing to say.[1]

It is important here to illustrate the topographical position of the pronephros and the relation of it to other parts; for these become definite for the first time in the present stage. For this purpose, a series of sagittal sections is instructive (figs. 112-114).[2] A few anterior myotomes (*mt.II-V*) are seen in fig. 112, which represents the section nearest the median line. In the posterior part of these myotomes, four cell-layers are distinguishable; the outmost layer (*ep.*) is the epiblast; the cell cord (*cd.*) inside the epiblast is the longitudinal section of the collecting duct, and

1) See the historical review under *Petromyzon*.

2) The embryo, from which these figures are drawn, is a little younger than that just spoken of.

its caudal continuation (*sd.*) is the segmental duct ; while the inner two layers (*m.v.* and *m.p.*) present respectively the parietal and visceral layers of the lateral plate. Below these structures, the roof of the enteric canal covers the fore-gut (*fg.*) and the commencement of the mid-gut which forms the passage of the enteric cavity from the anterior slender portion to the posterior wide cavity. In fig. 113 which represents the section next outside the last, the lateral walls of the five myotomes from the first to the fifth (*mt.I-V*) are noticed ; the cell-mass (*au.*) seen next anteriorly to the first myotome (*mt.1*) is a slice of the wall of the left auditory pit. The cross-sections of the pronephric tubules from the second to the fourth (*pt.2-4*) follow immediately behind the fifth myotome ; an oblique section of the fifth tubule (*pt.5*) and the nephrostomal part of the sixth tubule (*pt.6*) are also obvious behind the fourth tubule. The nephrostomes of the second (*nst.2*) and fifth (*nst.5*) tubules are seen only in part, while a larger part of the third, fifth and sixth nephrostomes is seen in the next figure (fig. 114, *nst.3*, *5*, and *6*) which represents a section still further outside. The sixth tubule, which is of weak development and has a wide nephrostome, is visible in these two sections (figs. 113 and 114, *pt.6* and *nst.6*). Except the first tubule which has already disappeared without leaving any trace, the five tubules are all thus seen in the dorso-lateral aspect of the hind section of the fore-gut and the commencement of the mid-gut.

The pronephros is thus situated in the neck which connects the head-protuberance with the globular abdominal portion. Below the pronephros, the narrow passage of the fore-gut passes through to unite the fore-gut with the wide space of the mid-gut, where afterwards the liver (*l.*) is found. Underneath the passage of the fore-gut, a group of mesenchymatous cells (*mch.*), which

constitutes the earliest fundament of the heart, is detected. *The present position of the pronephros,—dorsal to the heart, anterior and dorsal to the liver, and along either side of the chorda,—is retained by it for a comparatively long period* (see fig. 97); in later stages, the liver is somewhat shifted backwards, so that the pronephros now comes entirely in front of it (see fig. 115).

In an older embryo of this stage (figs. 92-96) the median folds of the coelomic projection, the component cells of which are very much flattened out, go in deeper towards the median line to meet with its counterpart on the opposite side. The second tubule (figs. 92 and 93, *pt.2*) has become weaker, as a comparison of these figures with figs. 77 and 79 will show. On the contrary, the tubule of the next pair (fig. 94, *pt.3*) has much elongated and is bent considerably in dorso-lateral direction, so that we can no longer observe the nephrostome together with the tubule itself on the same section. The following tubule, the fourth (fig. 95, *pt.4*), is likewise well developed; the fifth (fig. 96, *pt.5*) is more or less weak in development as compared with the tubules of the two foregoing pairs. In short, these three pairs (from the third to the fifth) make parallel progress with the development of other structures, for instance, the mesenterial fold or the muscle-segments. This is a fact that is to be observed too in the younger embryo of this stage, as above described. At the present stage, we can find no trace of the tubules beneath the ninth myotome, where the tubules of the sixth pair ought to be found, but only the cross-sections of the collecting duct or the anteriormost part of the segmental duct are seen.

Thus, the tubules of the third, fourth, and fifth pairs continue to grow, while the first pair has disappeared in the early part of the present stage (or at the end of the foregoing stage); the sixth has already commenced retrogression and the second is also growing weaker and weaker.

In the oldest embryo of this stage, there is to be seen no marked change in the pronephros, but the peritoneal lining is reduced into a very thin plate of a definite epithelium everywhere except at the pericardial portion, where the cells still have a columnar shape. The mesenchymatous cells accumulated on the median ventral line of the body are arranged in a certain order to be transformed into the cardiac tube. The third, fourth, and fifth tubules are also markedly prolonged and project into the body-cavity so as to cause the parietal layer of the peritoneum to fold between the epiblast and the body of the tubule (see this Journal: vol. x, Pt. XVIII, figs. 8, 9, and 10). In some of the embryos, the tubules of the second pair undergo degeneration. I have met with, in this series of sections, the same condition of the sixth tubule as on the right of fig. 86, the right tubule having entirely disappeared.

Period 4.

The embryos in the next advanced stage (Stage v) are much diminished in size, assuming a form of a retort or of a pistol ('97, fig. 1, *F*). Figs. 98-106 represent sections through an embryo of this stage. The posterior larger section of the foregut comprising the pronephric region, has been reduced into a slender tube (*fg.*) which is bounded by almost a single layer of high cylindrical cells. The parietal layer of the peritoneum as

well as the coelomic projection (*r.m.*) is very much decreased in thickness and encloses the peritoneal cavity (*pp.c.*) that has now become spacious, while the visceral peritoneum is still thicker than other parts. The mesenchymatous cells found in the foregoing stage on the median ventral line are transformed into a thin layer of the endocardium of the heart and its anterior continuation (*h.* and *tr.a.*) which are suspended by the dorsal and the ventral mensenteries, and enclosed in the thick pericardial coat still forming a continuation of the peritoneum. I can detect, however, none of the traces of the peritoneal partition which was developed so markedly in the last stage that one could not possibly overlook them. I have endeavoured to trace the mode of disappearance of this structure, but have only found that in one lot of embryos the whole set of the structure was present while in the other no trace of it was perceptible. Unfortunately I have found no embryo in an intermediate condition.

The few cells observed from the last stage beneath the chorda, and also in the space outside the pronephric tubules on both sides are more or less multiplied. As the former group is transformed finally into the dorsal aorta, and the latter into the anterior cardinal vein of either side, I shall call them the tract of the dorsal aorta and of the anterior cardinal veins respectively.

Fig. 98 represents the section through the hind border of the branchial region. On either side of the enteric tube a small space (*pp.c.*) of the body-cavity is surrounded by the peritoneal epithelium, still consisting, in this part, of somewhat cubical cells. The ventral edges of the peritoneal membrane of both sides are just meeting at the median ventral line. A few mesenchymatous cells (*tr.a.*) found in the space between this meeting point and the ventral wall of the enteric canal, are destined to

form the anterior continuation of the cardiac tube or the *truncus arteriosus*. An irregular cell-structure (x) is seen on either side above the dorsal corner of the body-cavity and inside the tract of the anterior cardinal vein. It is this structure about which I could not at first decide with certainty whether it was a slice of the hind wall of the branchial chamber or a part of the pronephric tubule. All the cases examined, however, point towards its being a part of the tubule ; the structure is detected in the anteriormost part of the body-cavity which wedges in, at about this stage, to the branchial region with a sharp angle (see fig. 97). The narrow space (fig. 98, *pp.c.*) found intervening between the structure and the peritoneal walls is a part of this cavity. One might suppose that the space may be the coelomic cavity of the branchial region ; but, the space between the parietal and the visceral peritoneum of the branchial region is consolidated already in the preceding stage, being filled up with variously shaped cells of mesenchymatous nature (see fig. 97).

In the next following section shown in fig. 99, a tubular structure (*pt.2*) with an oval outline is seen on either side at the place where the pronephric tubule ought to be found. Its long axis is directed just like a tubule (compare with figs. 101, 102, &c.). This is doubtless a part of a pronephric tubule ; but the corresponding nephrostome which ought to be found either in the section in front (fig. 98) or behind (fig. 100), can not be detected in either of them. The nephrostome must, therefore, be looked upon as having degenerated ; and since this pair of the tubules is, in fact, detected underneath the fifth myotome, it must be identified as the second pair of the tubules. The section represented in fig. 100 shows on both sides the cross-sections of the collecting duct (*cd.*). On the left side, a cellular structure connects the

peritoneum and the collecting duct; it is the posterior wall of the tubule in figs. 99. Fig. 101 represents the section through the axial plane of the third tubule, the nephrostomes of which are recognized more clearly in the section behind it (fig. 102, *nst.3*). The tubules of this pair are comparatively not long. The fourth pair of the tubules and their nephrostomes are obvious in fig. 104 (*pt.4* and *nst.4*) which represents the third section behind that of fig. 102; the tubules much resemble those of the pair in front, showing the same convolutions as these. It is a peculiarity of the present stage that the aperture of the nephrostomes of the third and the fourth pair is not so wide open as in the last stage or as in more advanced stages! It is always nearly closed and slit-like, so that we can hardly trace the communication between the lumen of the tubule and the body-cavity.

Fig. 103 represents the section intervening between the sections shown in figs. 102 and 104. On the right side, the collecting duct alone, and on the left side, the duct together with a small part of the fourth tubule, is shown. The peritoneal membrane on the dorsal end of the body-cavity is folded far into that cavity (fig. 103, *bs.*). This fold is traceable from the anterior part of the third tubule to the hind part of the fourth (figs. 100-104, *bs.*). The space enclosed in this fold communicates freely with both the tract of the dorsal aorta under the chorda and the tract of the anterior cardinal vein outside of the pronephros and contains a number of mesenchymatous cells which probably wander in from the tract of the aorta and the anterior cardinal vein. As subsequent history shows, this structure constitutes the beginning of *the glomerulus of the pronephros.*

Figs. 105 and 106 represent two contiguous sections immediately posterior to the section shown in fig. 104. In fig. 105 we

observe on either side the cross-section of the collecting duct (*cd.*) together with a part of the fourth tubule (*pt.4*) ; the longitudinal section of the fifth tubule (*pt.5*) is seen on the right side of fig. 106, standing in wide communication (*nst.5*) with the body cavity. This is the hindmost tubule. In the sections lying behind this, the cross-section of only the segmental duct is repeated.

Thus the tubules of the second pair undergo, at the present stage, complete degeneration. This process begins, in this case, as above seen, at the nephrostome and proceeds upwards to the collecting duct,—a process which is just the reverse of what is observed in the reduction of the tubules of the sixth pair and probably also of the first pair, in both which cases the tubules are first cut off from the collecting duct and the separation from the peritoneal cavity follows afterwards.

Period 5.

In the Stage VI, embryos have developed so far that all the organs have received their definite forms and proper position with the exception of the middle and the hind portion of the gut, whose development is much delayed on account of the yolk-mass. Having absorbed the yolk-granules, the component cells of most organs are much diminished in size.

Figs. 107-110 have been drawn from a series of sections through an embryo in this stage. The enteric canal (*fg.*) is much diminished in diameter, presenting, in section, an elongated heart shape. The peritoneum becomes very thin in all its parts with the exception of the pericardium and the coat of the *truncus arteriosus*, in which its component cells are of cylindrical or cubical shape.

The peritoneal membrane lining the enteric canal immediately behind the branchial region is also thicker as compared with other parts (fig. 107), being composed of a single layer of cubical cells,—a peculiarity observed since the last stage (compare figs. 98-99 with fig. 107).

The pronephric tubules as well as the collecting duct are composed of a regular epithelium of cylindrical cells; the former, moreover, are much prolonged and, in some parts (fig. 108), much coiled, so that the peritoneal cavity which was almost a hollow space in the last stage, is filled up with the tubules and the cardiac tube.

Fig. 107 represents the section through the hind part of the sixth myotome; a pair of the tubules (*pt.3*) is hanging down in the body-cavity immediately behind the hind wall of the branchial chamber. On the right side, the axial plane of the tubule is cut through, while, on the left, the anterior wall of it is sliced; these are the tubules of the third pair. They show no bending in the antero-posterior direction, but are curved laterally and ventrally. The component cells are, in the nephrostomal portion, taller in comparison with those in other parts of the tubule or the collecting duct. The fourth tubule and nephrostome are seen on the right side of fig. 108, while on its left side, the communication of the corresponding tubule on the opposite side with the collecting duct is recognizable. The left nephrostome is found in the third section behind this, which is not figured. This pair of the tubules exhibits, in section, constrictions at two or three points owing to their curving somewhat in the antero-posterior direction (see the tubule on the right side of fig. 108). Fig. 109 is the section immediately behind the last and shows the cross-sections of the collecting duct (*cd.*) and a piece of the left fourth tubule (*pt.4*).

A pair of the glomeruli (figs. 108 and 109, *gl.*). is seen adhering on the median side of the tubule on each side and lined with the visceral peritoneum. The glomerulus represented in the last stage by a folding of the peritoneum which covers the tubules from the third pair to the fourth[1], is reduced, at present, into a pair of sacs of this membrane projecting on each side between the fourth and the fifth tubules; the other part of the folded membrane becomes adhered firmly to the walls of either the tubules or the body-cavity leaving no space of sacculation,—in short, a pair of long folds, extending from the anterior part of the third tubule to the fifth in the last stage, is reduced into a pair of sacs found in the position just mentioned. The inside of the sacs is compactly filled up with free-cells and communicates with the aorta tract and with the space outside the pronephros, where free-cells to be afterwards transformed into the anterior cardinal vein have been observed already from the foregoing stage.

The section represented in fig. 110 fortunately passes symmetrically through a pair of the nephrostomes (*nst.5*) and of the tubules (*pt.5*) hanging down in the peritoneal cavity. This is the fifth or the hindmost pair of the pronephric tubules in the present stage. The communication of the tubules with the collecting duct is seen in the section behind this. The tubules present also some antero-posterior bendings. Posterior to this, no tubule is found.

The pronephric tubules in the present stage are, therefore, reduced into the minimum number, i.e., three pairs[2], *all of which are retained so long as the organ functions as the excretory apparatus*

1) See p. 355.

2) We occasionally find the four tubules to persist, and the additional tubule is the sixth.

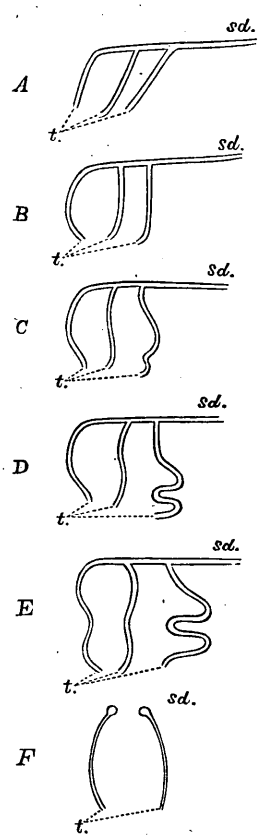
during the larval life of Petronyzon. Especially it must be noticed that the foremost pair of the persistent tubules (the third pair) is in close contact with the hind border of the hind wall of the branchial chamber where, in the foregoing stage, the second pair of the tubules was found, this latter having disappeared in the course of the last stage. It follows that the two somites, to which the first and the second pair of the tubules have belonged, have now entered into the formation of the branchial region.

The development of the pronephros after this consists only in the prolongation and the convolution of the tubules, no further change taking place with reference to the number of the tubules or to their histological structure, until the system undergoes degeneration to be replaced by the mesonephros, which functions as the excretory organ for the whole subsequent life of *Petromyzon.*

The convolution of the tubules is hard to make out. I have reconstructed them from a number of sections; some of these are diagramatically given in the annexed woodcut (Woodcut 2).

With the growth of the muscle-segments the collecting duct is prolonged, so that the points of connection of the tubules with that duct become farther apart from one another, while the nephrostomal portions of the tubules retain more or less their original positions; in this wise, the tubules are laid in oblique positions directed anteriorly and posteriorly (*A*) and have no other curvature than the ventro-lateral bending (the frontal projection of the curvature is shown in *F*). Then the antero-posterior bending begins to take place. The foremost tubule is curved forwards in its whole length, while a small curvature in the distal

(nephrostomal) portion of the two following pairs is directed backwards. The nephrostomes retain their first position (*B*). Now the secondary curvatures take place (*C*). The nephrostomal part of the foremost pair is crooked just like that of the two hind pairs in *B*; the middle tubule is bent forwards like the foremost tubule. The hindmost tubule makes a small forward curvature and a large backward bending. In the next stage, *D*, the foremost and the middle undergo no marked change, but the secondary curvatures of the hindmost tubule are much more strongly expressed. In *E* the foremost receives a secondary curvature directed backwards at the middle part; the middle acquires a curvature in opposite direction; the hind tubule undergoes no marked change except in the increased degree of the original curvatures. It seems that the subsequent bendings always take place in the curved portion until there arises a system of complexly convoluted tubules filling up the chest cavity.



Wood-cut 2.—Diagrams showing the convolutions of the tubules in later stages.
t. pronephric tubules.
sd. segmental duct.

As has been said, throughout these phases the positions of the nephrostomes are not markedly changed, retaining the same condition for a considerable period. The bendings of the tubules

are caused, therefore, by the growth of the tubule at the point of bending.

The curvature in the ventro-lateral direction is very simple and undergoes no remarkable change; its projection is shown in *F*.

B.—The Segmental Duct and the Genital Cells.

For the sake of simplicity, the development of the segmental duct and of the vascular system in the pronephros has been entirely put aside in the description given above.

As already alluded to, the origin of the segmental duct in *Petromyzon* is extremely difficult to make out, because its formation goes on rapidly at a comparatively young stage. The early process of its formation is essentially the same as in the pronephric tubules. In the anterior region, the *intermediate cell-mass* or the *nephrotome* (see p. 340) behaves itself in precisely the same manner as in the Anlage of the pronephric tubules; the difference is that it is cut off from the lateral plate and is transformed into the duct, while in the case of the tubule it retains the continuity with the lateral plate. If fig. 31, which represents the section through the tenth somite (*i.e.* the somite, from which backwards the Anlagen are converted to the segmental duct) be compared with the left half of figs. 2, 5, 6, 14, and the right half of fig. 16, in which the Anlagen all develop to the pronephric tubules, it will be found that there is no difference between them; in fact, they are morphologically equivalent to one another. Such an Anlage is, posterior to the pronephros, not confined to the tenth somite, but, as has been already repeatedly said (pp. 317, 320, and 327), is observed for some segments further backwards (see fig. 17).

The Anlage thus pronounced in each somite soon assumes a characteristic oval form, being completely cut off from the myotome to which it belongs (compare the right side of fig. 3 with the right side of fig. 58 and see the description on p. 335). The mode of constriction is also the same as in the case of the pronephric tubules; the indentation begins at the anterior and posterior borders of the somite, and the middle portion is cut off last (compare with the explanation on p. 320).

Here also, the coelomic projection is formed in the same mode and at the same point as in the case of the pronephric tubules (see left side of fig. 63, *c.p.*).

Up to about this time, the Anlage shows a feature much resembling that of the tubule, so that one who has not followed its further history might mistake it for a pronephric tubule (compare the left side of fig. 63 with figs. 67-74). But cell-mutiplication which occurs almost invariably in the case of the pronephric tubules, is not observed in the Anlage of the segmental duct which is soon cut off from the lateral plate (including the coelomic projection) and assumes a characteristic tubular structure composed, in cross-section, of radially arranged cells of columnar shape. Its position is always on the parietal aspect of the dorsal (proximal) angle of the peritoneum where the coelomic projection passes over into the lateral plate (see fig. 75). This separation of the Anlage of the duct from the lateral plate goes on, it seems to me, on the whole from the anterior part to the posterior, but often irregularly ; for not infrequently, the duct in some anterior somite is connected with the lateral plate, while it is already cut off completely in posterior somites. In fact, there are some somites in which the separation is very much delayed and I have often been surprised to find what appeared like a pronephric tubule in a

somite (see fig. 63) far backward of the posteriormost tubule which is found in the ninth somite.

The segmentally arranged Anlagen of the segmental duct are secondarily united with one another just as in the case of the collecting duct in the pronephric region. This union seems to take place during the separation of the Anlage from the myotome and is finished before it is separated from the lateral plate; for, when the Anlage first comes into view, there is no intersomitic cord as in the case of the pronephric tubules and the duct is seen already consisting of radially arranged cells (fig. 58) when it is cut off from the lateral plate. When established, the duct is the same in structure in both the somitic and intersomitic spaces; a cross-section of such a duct in the intersomitic portion is shown on the right side of fig. 75, while that in the somitic portion is seen on the left of the same figure.

This condition of the duct is already traceable, in Stage III, for no fewer than 10 somites from the hindmost pronephric tubule backwards, and it forms a direct posterior continuation of the collecting duct. The duct remains awhile as a solid cord of cells arranged radially in cross-section, but it soon acquires a lumen (figs. 75, 86, and 87, *sd.*). The further development of the duct goes on more promptly than that of the tubules in the hind part, and therefore, the embryos at such a stage (Stage III) have a well developed duct and more or less primitive tubules (compare fig. 74 with fig. 75).

In the hind region, where yolk-cells are crowded, the process is much delayed and more or less modified. Instead of the differentiation of the cells *in situ*, it seems to me, a few cells are detached from the nephrotome; a number of cells is produced by repeated division of these cells (fig. 19, *a.sd.*) and becomes ar-

ranged as in the Anlagen in the anterior region. Fig. 89 represents the section through the twenty-eighth somite in the series of sections shown in figs. 77-86 ; it is the hindmost section in this series of sections, in which the cells just spoken of are detected ; there are found a few cells (*a.sd.*) of this kind which show no definite structure, but are scattered. In the next anterior section (fig. 88) the cells are arranged more or less radially. In the sections lying further anteriorly to this a perfect tube is formed as seen in fig. 87 (*sd.*) which shows the frontal section through the seventeenth to twenty-third somites in the same series as the above two figures.[1] In what somite this modified mode of the formation of the duct begins I can not tell with exactness, but it is certain that the duct arises by the differentiation of the nephrotomic cells *in situ* more than 10 segments back of the hindmost pronephric tubule. I have considered it possible that these cells (*a.sd.*) might be epiblastic in origin, but I can not find that the cells composing the epiblast over this cell-group show any sign of mutiplication ; while on the other hand, the cells on the dorsal edge of the lateral plate (which corresponds to the nephrotome in the anterior part) are very active. I see, therefore, no escape from the conclusion that these cells are mesoblastic in origin.

Also in the anterior part of the body, the epiblast consists throughout these phases of development always of a single layer of cubical cells and shows a sharp contour against the structure inside it, being, in most cases, intervened by a space. Naturally, mitotic figures are observed at several points, but the products of these cell-divisions contribute only to the extension of the epiblast itself, as may be inferred from the direction of

1) By the bending of the body-axis, some sections in a series of cross-sections are unavoidably cut through frontally.

the spindles, the long axes of which are directed always parallel to the surface of the layer. *I have nowhere observed any trace of either the proliferation or of the casting off of cells from the epiblast to give rise to the segmental duct.*

In the cloacal region, the formation of the segmental duct goes on a little earlier than in the region next anterior to it. In spite of much effort, I failed to observe the very beginning of the formation at the cloacal opening, and I have nothing to tell of its earliest stage. In the series of sections from which figs. 77-89, are drawn, I can not yet find in the adjacent part of the cloaca any trace of the duct; but in the section represented in fig. 90 which passes through the dorsal lip of the blastopore of an embryo with about the same number of the mesoblastic somites (34 or 35) as the one just referred to, the duct already breaks through into the cloacal cavity (*co.sd.*)[1]. Fig. 91 represents the next ventral section which passes through the dorsal part of the blastopore (*bp.*). As seen in these two sections, immediately inside of the blatlopore (*bp.*), where the hypoblast passes over into the epiblast, the walls of the cloacal cavity send out, right and left, a symmetrical pair of diverticula[2] (*c.dv.*), forming an acute angle, the inner side of which is a part of the enteric wall, while its outer side is the direct continuation of the epiblast. The walls of this diverticulum pass over into the segmental duct (*sd.*). *The communication of the segmental duct with the cloacal cavity is found, therefore, at the point where the epiblastic layer of the lip is reflected inside and passes over into the hypoblast.* This point of communication is, however, shifted far inside and

1) This opening is found in the same vertical plane as the 34th or 35th somite.

2) The right diverticulum only is seen in figs. 90 and 91, the left one being observed in another section which is unfigured.

dorsally when the development proceeds further (fig. 111, *co.sd.* and *c.dv.*).

I have also met with two cases (figs. 90 and 111), in which I have observed some epiblastic cells of the external lateral walls of the blastopore multiplying actively and having mitotic spindles (x) with axes directed perpendicularly to the plane of the epiblast, while the duct comes in firm connection with that point of the epiblast,—the connection is so firm that the duct and the epiblast appear to form one and the same tissue. At this point, thus, there is every appearance of epiblastic cells partaking in the construction of the segmental duct.

The collecting duct pertaining to the ninth somite forms the segmental duct in that segment, having lost the connection with the tubule.

Up to Stage II, the duct is represented by the segmental Anlagen in about 8 segments back of the ninth somite; in Stage III, these Anlagen are converted into the duct in about 10 anterior segments; while in the course of Stage IV it opens out into the cloacal cavity.

From the above account, it is easily conceivable that *the Anlage of the segmental duct and that of the pronephric tubule are perfectly homologous, and that the duct is a continuation of a series of abortive pronephric tubules in the hind region.*

Underneath ten and more myotomes lying posterior to about the fifteenth somite the proximal portion of the lateral plate, which corresponds to the nephrotome, contains peculiar large cells (figs. 87, 88, and 89, *gc.*) loaded with an enormous quantity of yolk-granules; the other mesoblastic cells in this part, being much flattened out, form a thin layer over these cells. These

peculiar cells are, I think, the equivalent of the primitive genital cells found in the corresponding part of the Amphibian and Selachian body.

Up to Stage III, these cells can not be distinguished from other mesoblastic cells which are equally rich in yolk-granules. In Stage IV, they become conspicuous; and in Stage V, again indistinguishable from other constituent cells of this part.

C.—The Vascular System in the Pronephros.

In early stages, no trace of the vascular system is perceived in the pronephros. What is recognisable as a fore-runner of the vessel is represented by mesenchymatous cells scattered in the space between the primary germinal layers (figs. 77 and 82, *mch.*). These free cells are detected, during Stage IV,[1] in three tracts, viz., beneath the chorda, beneath the ventral wall of the enteric canal and outside the pronephric tubules on either side (figs. 77, 79, 80, 81, and 82, *mch.*). In Stage V, or at the end of Stage IV, the cells below the hind section of the fore-gut are converted into the endothelium of the heart and of the vessels which are its direct continuations. The cells beneath the chorda are destined to be transformed into the dorsal aorta, and the cells on either side of the pronephros constitute the first indication of the cardinal veins. It is these three vessels—the aorta and the two cardinal veins—which come in relation with the pronephros.

In the embryos in which the degeneration of the tubules is still going on, there is no special vessel supplying the pronephros; but when the process is over, a pair of long blood-spaces (figs. 100-104, *bs.*) is found in communication with the aorta-tract.

1) A few of them are observed here and there already in Stage III.

They are the spaces formed by the slackening and folding of the median peritoneum which coats the pronephric tubules, as above stated (p. 355). The fold, i.e. the space, extends throughout almost the whole length of the pronephros (figs. 100-104) and contains numerous free-cells. But, as the peritoneum finally adheres to the median walls of the tubules in each nephric segment, the space becomes divided into three pairs : the anterior pair, which soon disappears, is found between the tubules of the second and third, the middle is detected between the third and fourth, and the posterior between the fourth and fifth tubules. These spaces communicate directly,—medially with the aorta tract and externally with the tracts of the anterior cardinal veins, which emerge, in later stages, in the pronephros. They are the blood-spaces which, I believe, correspond with the intersomitic arteries demonstrated by PAUL MAYER and others in Selachia.

When the tubules develop further, the arterial portion of these blood-spaces disappear except the middle portion where it is sacculated and filled up with the mesenchymatous cells (figs. 108-109, *gl.*). This portion is the structure which is called the glomerulus of the pronephros ; it is found one on each side (see pp. 355 and 358).

Having followed, in the foregoing pages, the successive processes which take place in the development of the pronephros in *Petronyzon*, step by step, I will give a short resumé of the facts.

1. In the earliest part of Stage II, the mesoblast consists simply of the parietal (dorsal) and the visceral (median and ventral) layers. The proximal portion of the mesoblast is dis-

tinguished, first of all, in the histological structure from the distal portion: the former is composed of columnar cells, and the latter of irregularly shaped cells. Only the proximal portion which occupies the largest part of the mesoblast undergoes the metameric segmentation and gives rise to the scleromyotome and the nephrotome (in the sense of RÜCKERT); the distal smaller portion remains unsegmented and is later converted into the flattened epithelium of the peritoneum.

2. The earliest traces of the pronephros are noticeable in exceedingly young stages, that is, in the early part of Stage II, in which the embryo has about 16 somites.

3. They are expressed in the form of a diverticulum of the parietal layer of that section in each mesoblastic somite, which forms the ventral half of the segmental part of the mesoblast and is called the nephrotome. This is the Anlage of the pronephric tubule and not of the segmental duct.

4. The pronephric diverticulum or the Anlage is brought about by the evagination of the parietal layer in each nephrotome, enclosing a part of the primary coelomic cavity.

5. The nephrotome is separated from the proximal portion of the segmented mesoblast and forms awhile the proximal portion of the unsegmented mesoblast or the lateral plate. The separation begins with an indentation in the anterior and posterior borders of the mesoblastic somite; the myocoelome communicates for some time by a narrow passage with the general coelomic cavity.

6. The Anlage has no histological connection either with the preceding or the following Anlage or with the other germinal layers; it is, therefore, segmental in origin and myomeric in position.

7. The Anlagen are developed, in Stage II, in about 12

segments and are cut off from the scleromyotome in 4 segments. In Stage III, the separation of the Anlage from the myotome goes as far backwards as the sixteenth or seventeenth segment.

8. The anteriormost Anlage is found in the hind part of the fourth somite and is the first to arise; the second follows it, and so forth.

9. The Anlagen in each somite are secondarily united with one another by the solid cellular cord which is budded out of the anterior and posterior rims of the Anlagen themselves; thus the collecting duct (*Sammelrohr* in the sense of RÜCKERT) is established. This process is originally to be looked upon as the coming together of the ends of the tubules.

10. The canalization of the collecting duct begins within each Anlage and proceeds, generally speaking, posteriorly, until the Anlagen in front and back are put in free communication.

11. Each Anlage grows dorso-laterally and acquires a tubular form. The collecting duct is shifted gradually in a dorso-median direction; finally it comes to lie between the myotome, the mesentery, and the *chorda dorsalis*.

12. The tubules open in the coelomic cavity at the lateral angle of the dorsal corner of that cavity.

13. In the somites posterior to the ninth, the tubules are, during Stage III, cut off also from the lateral plate and establish a long duct running, on each side, along the dorsal aspect of the lateral plate where originally the tubules opened. This is the segmental duct; the tubules and the collecting duct in the somites anterior to this constitute the glandular part of the pronephros.

14. The glandular part, or the pronephros proper comprises six somites, from the fourth to the ninth. The maximum number

of the pronephic tubules which is attained by the embryo in Stage III, is, therefore, six pairs.

15. The tubules of the first and second pairs come, in Stage III, temporarily in close contact with the epiblast, but do not receive cells from it; they soon return to their original condition.

16. The anterior extremity of the system shows, from the first, degenerating features. The first, second, and sixth of the tubules begin, during Stage III, to decline; and at the end of Stage IV, or the beginning of Stage V, the tubules are reduced into the minimum number, which consists of three pairs from the third to the fifth. These three pairs function as the actual excretory organ for a considerable length of time.

17. Retrogression is first met with in the first pair of the tubules, which decline probably without further development, soon after their separation from the myotome is completed; they seem to atrophy from the free end. The next pair degenerating is the sixth, which is at first cut off from the collecting duct and remains for a short time, but soon disappears without leaving a trace. The second pair persists for some time seemingly to function as the excretory organ, but it atrophies already in the early part of Stage V, the communication with the coelomic cavity being first obliterated; and in Stage VI, none of the structure remains to be recognized.

18. The foremost pair of the persistent tubules comes to lie in close contact with the hind wall of the branchial chamber. The two mesoblastic somites which correspond to the first and second nephromeres should therefore be looked upon as having entered into the formation of the branchial region.

The stages in which the tubules appear and abort in different somites are shown in the annexed table.

	Som. I	Som. II	Som. III	Som. IV	Som. V	Som. VI	Som. VII	Som. VIII	Som. IX	Som. X	Som. XI	Som. XII
Stage II				Anl. 1	Anl. 2	Anl. 3	Anl. 4	Anl. 5	Anl. 6	Anl. 7	Anl. 8	Anl. 9
Stage III				Tub.1	Tub.2	Tub.3	Tub.4	Tub.5	Tub.6	Segmental duct.		
Stage IV					Tub.2	Tub.3	Tub.4	Tub.5	Tub.6	Segmental duct.		
Stage V					Tub.2	Tub.3	Tub.4	Tub.5	Segmental duct.			
Stage VI						Tub.3	Tub.4	Tub.5	Segmental duct.			

19. In older embryos of Stage III, the visceral layer of the nephrotome is folded out, and is called the coelomic projection which resembles the coelomic pocket described by PRICE for *Bdellostoma*; however, in *Bdellostoma*, the fold is derived from *the parietal and visceral layers of the lateral plate* and is afterwards converted into the BOWMAN'S capsule, whereas the coelomic projection is the *product of only the visceral layer of the nephrotome*; it gives rise to the radix of the mesentery which offers materials to the mesonephric tubules and to the gonads.

20. The topographical position of the pronephros becomes first definite in Stage IV. It is situated in the chest cavity, dorsolateral to the heart, forward of and dorsal to the liver, extending along either side of the chorda. This position is somewhat changed as the development proceeds; the pronephros comes, in later stages, in front of the liver.

21. During Stage IV, a structure, which I have called above the *peritoneal partition*, is observed as an outgrowth of the peritoneal wall and disappears during the same stage.

This horizontal partition is found at three levels. The most dorsal is well developed, the ventral is a mere trace, and the middle is intermediate between the above two. I can not state at present anything definite as regards the significance of this structure.

22. The convolution of the pronephric tubule takes place in Stage IV. With the growth of the myotome, the collecting duct is prolonged; consequently the connecting points of the tubules with the duct are farther removed from one another than before, whilst the nephrostomes retain their original position; so that, the two posterior pairs of the tubules are placed in an oblique direction from dorsal and caudal to ventral and cranial. Each tubule is, then, convoluted in a cranio-caudal direction between the heart and the lateral peritoneal wall. In older stages, the tubules are coiled in all directions, until the chest cavity becomes filled up with the convolution of the tubules.

23. Up to Stage VI, the nephromeres and the myomeres exactly coincide one above the other in position. This period is very long in comparison with other Craniota. As the development proceeds further, the pronephic tubules are however shifted gradually backward, so that, in Ammocoetes 10 mm. long, the myotomes are already not situated upon the tubules pertaining to each of them. In later stages, nothing of the relation can be traced.

24. The segmental duct is looked upon as being brought about by the union of a series of the abortive pronephric tubules in about 12 somites lying posterior to the eighth somite[1]. The Anlage is laid in the parietal layer of the nephrotome in exactly the same manner as in the pronephric tubules of the glandular part.

1) In the 9th somite, the aborted tubule actually forms the duct in the segment.

The difference is, that the tubules in the posterior region are soon cut off from the lateral plate and become the duct.

25. Between the epiblast on one hand, and the Anlage or the duct on the other, there exists always a space, and the duct has no connection with the epiblast except at its posteriormost end where the epiblastic cells might, as judged from the mitotic figures, contribute to the formation of the duct.

26. In the somites posterior to about the twentieth somite, the Anlage of the duct is represented by a few cells in each segment probably detached from the dorso-lateral angle of the nephrotome. These cells multiply and are transformed into the segmental duct in the posterior part.

27. In Stage II, the Anlagen of the segmental duct are cut off from the mother layer in a few somites; in Stage III, the duct is formed as far as about the eighteenth somite, while in Stage IV, it breaks out into the cloacal cavity. The cloacal opening of the segmental duct is found at a point where the hypoblastic cloacal wall is reflected into the epiblast, these two layers forming a diverticulum on either side.

28. In stage IV, the primitive genital cells become apparent in the nephrotomes of the posterior 10 or more somites; they can not be discriminated from other mesoblastic cells in the next advanced Stage.

29. The blood-vessels, which specially supply the pronephros, acquire definite form in comparatively later stages, viz., at about Stage V. The dorsal aorta pours out the blood into two pairs of the blind vessicles, which are formed by the folding of the parietal peritoneum and are found between the first and second pairs, and between the second and third pairs, of the persistent tubules

respectively. The venous blood is carried away through the anterior cardinal veins which penetrate the pronephros.

29. These blood-spaces are thus segmental in arrangement and *intersomitic* in position. The two anterior pairs of them soon undergo atrophy, but the posteriormost pair persists, becoming enlarged and sacculated at the distal extremity. This sacculated part of the vessel is filled up with free-cells and is called the glomerulus of the pronephros, and, therefore, there is only a pair of glomeruli in *Petromyzon*.

II. Historical Review and Conclusions.

As is well known, MAX SCHULTZE ('56) was the first who discovered the pronephros in *Petromyzon*. Having investigated the larvæ of *P. planeri*, the author describes the structure as "Drüsenanlage" and homologised it with the "Urnieren (Wolf'sche Körper)" of the frog's larva. His statements on this body are as follows: "Nicht lange nach der Bildung dieser Drüse (Thymus) entsteht die Anlage einer zweiten, aus dem unter der Chorda dorsalis angehäuften Blastem über dem Herzen. Aus der durch Pigmentanlagelungen früh schon sehr undurchsichtig werdenden Masse wachsen nämlich nach unten, gegen das Herz zu, 3 oder 4 kurze Fortsätze hervor, welche eine eigenthümliche Wimperung zeigen" (p. 30).

The stage spoken of probably corresponds to Stage V, or VI, of my embryo.

Our knowledge on this subject received important additions by the noted investigations of W. MÜLLER and MAX FÜRBRINGER. MULLER ('75) noticed the first traces of the pronephros in a very young embryo, which had yet only four pairs

of gill-slits. This Anlage gives rise to a much coiled gland, which opens into the body-cavity, at first through only one ciliated funnel, but afterwards through four. The gland passes over posteriorly to a pair of ducts, which run along the chorda on either side and open into the cloaca. MÜLLER has homologised the structure with the "Vorniere" of *Myxine* and called the duct "Urnierengang" (pp. 121-122). He found a pair of glomeruli projected on the median surface of the gland and lined with the peritoneal epithelium.

MAX FÜRBRINGER ('78) studied the larvæ of *Ammocoetes planeri*, which varied from 4.5 to 180 mm. in length. His statements essentially confirm MULLER'S. In his account we find the following sentences: "Die auf allen Präparaten ausgebildete Vorniere, die ich im Wesentlichen ganz wie MÜLLER fand, bildet einen nahmentlich bei den mittleren Stadien voluminösen und durch 4-5 Myokommata erstreckten Complex von Windungen, die vorn durch mehreren Peritonealcanäle (Wimpertrichter) in Bauchhöhle münden und hinten in den Vornierengang übergehen. Diese auf die 2-3 ersten Myokommata beschränkten Trichter ragen in unregelmässiger Folge bald ventral-medial, bald ventral-lateral in die Bauchhöhle vor und wurden (von CARLBERLA und mir) meist zu fünf gefunden. Die von rundliche Epithelzellen bekleidete Glomerulus verhielt ganz wie MÜLLER beschreibt" (p. 42).

The larvæ of *Ammocoetes* in question seems to correspond probably to Stage v, or later stages of my list; in such a stage, I could not find more than three (or rarely four) pairs of the tubules, or of the nephrostomes.[1)]

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1) See the foot-note on p. 358.

The authors who have investigated the development of *Petromyzon* embryos step by step, are W. SCOTT, GOETTE, SHIPLEY, and v. KUPFFER. Their opinions are, however, somewhat divergent. SCOTT ('82) derives the pronephric tubules from the segmental duct which is, according to him, brought about by the differentiation *in situ* of the cells forming the proximal margin of the lateral plate. The process takes place in the whole extent at the same time. At certain points (segmental?) of the duct thus formed, evaginations are produced out of it; these evaginations subsequently open into the body-cavity and establish the nephrostomes which are, according to SCOTT, found from two to three pairs in number. At about the stage in which the funnels are formed, he observed a pair of glomeruli.

"In most respects," SHIPLEY's observations ('87) "confirm his" (SCOTT's). But "on the origin of the ciliated funnels, the results differ from SCOTT's" and agree with those of FÜRBRINGER (Amphibian pronephros?). According to SHIPLEY, "in the region of the heart, where the body-cavity has already appeared, its origin (*i.e.*, of the segmental duct) seems to be somewhat different. The lumen of the segmental duct here becomes continuous with a groove in the parietal peritoneum, lying near the angle where the somatopleure and the splanchnopleure diverge. When this groove closes it leaves four or five openings which persist as the openings of the ciliated funnels" (p. 20).

v. KUPFFER[1] ('88) observed, in *P. planeri*, the three pairs of the tubules arising from three distinct evaginations of the parietal

1) I know this paper only by the abstract in: Jahresbericht ü. die. Fortschr. d. Anat. u. Physiol., Bd. 17. 1889.

layer of the lateral plate; the segmental duct is looked upon as of the epiblastic origin.

GOETTE ('90) worked the development of *Petromyzon fluviatilis*; his results with respect to the pronephros show some agreement with mine, especially those concerning the later stages. The author derives also the whole system of the pronephros (including the segmental duct) solely from the mesoblast. But we diverge in some important points from each other; he has found the earliest traces of the structure at a time when the rudiment of the heart first becomes apparent (his VI. Periode) (p. 64). From the account given in the foregoing pages it is clear that this period belongs to a later stage in which the pronephros has already made a considerable progress in development; his figures 99, 103, &c., which are spoken of as representing the first appearance of the structure, approximately correspond with my figures 82, 83, &c., and with those of even older stages.

The pronephros is, according to GOETTE, not of a separate Anlage in its first appearance, but arises in a form of a longitudinal furrow formed, on each side, by an evagination of the parietal layer of the mesoblast; the lips of the furrow being fused at certain points, there remain three openings; these are converted afterwards into three tubules and ciliated funnels. The tubules are added by stages until there are usually five, or more rarely four or six; but how these are multiplied, he can not say with certainty. The tubules have, it seems to him, no relation to the metameres of the body; for 3 to 5 tubules are found in the extent of 2 to 3 metameres (*loc. cit.*, pp. 64-65).

The segmental duct originates, according to GOETTE, in precisely the same way as the pronephros proper; the only difference is the complete constriction of it from its mother-layer just as

I have made out. From the region of the liver-anlage backward the development of the duct is irregular; he says: "Auf der einen Seite zeigt sich seine Anlage noch rinnenförmig, während sie auf der andern Seite schon vollkommen röhrenformig abgeschnürt ist. Endlich wechselt dies Verhalten auch auf derselben Körperseite, so dass derselbe Gang, von der Lebergegend rückwärts verfolgt, bald rinnen-, bald röhrenförmig, geschlossen oder mit offener Lichtung sich darstellt" (*loco cit.*, p. 56). The hind end of the duct opens in the cloaca (Afterdarm) by the fusion of their walls and by the communication of the lumen of the duct and the diverticulum of the cloaca. I have not observed in any stage of my embryos examined the numerous convolutions of the segmental duct demonstrated by GOETTE in the region immediately behind the "ursprüngliche Kopfniere."

GOETTE has made out the three "peritoneale Scheidewände," as he calls them: two respectively in the anterior and the posterior end of the pronephros, and the third on either side of the liver. Later, the first contributes, according to him, to the formation of the hind wall of the branchial pouch (Kiementasche); the second is converted into "eine Venenbrücke zwischen dem Sinus venosus and der Leibeswand," while the third disappears without leaving a trace. They are, according to GOETTE, homologous with the "Schlussplatte" of the pronephros in Teleostei; considered phylogenetically, nevertheless, they have no intimate relation to the pronephros in *Petromyzon* (*loco cit.*, pp. 56-61). This structure is, as stated on p. 349, doubtless the same as the uppermost peritoneal partition which I have found in my embryos. I have nothing to communicate on its significance; but I feel sure that his statement is not accurate when he says the structure appears earlier than the pronephros; for his figs.

96 and 97, to which his statement refers, represent a stage considerably later than the first formation of the pronephros itself. And the peritoneal partition is not confined to these three points, but is continuous throughout the whole extent of the pronephros; moreover, beside the "peritoneale Scheidewände," there are found two other partitions of a similar character as above stated. Also, as to the fate of the structure my results differ from his: I have not been able to observe at all any such contribution to the formation of the hind wall of the branchial chamber and of the "Venenbrücke," as is affirmed by GOETTE.

RABL ('96) says in his recent extensive work on the Selachian nephric organ, that in quite young larvæ of *Petromyzon fluviatilis* the pronephros also begins in the seventh somite, in which the first of the four ostia are found, as in *Pristiurus*.[1] His larvæ are, however, 501 hours or 20 days and 21 hours old; such larvæ correspond to my embryos in Stage VI, and upwards, in which anteriorly two pairs, and posteriorly, one pair of the tubules disappeared and only three persistent tubules are seen. His first nephrostome represents the foremost of the persistent nephrostome.

The accounts cited above all agree with the results given in the present paper in deriving both the pronephros and the segmental duct from the mesoblast alone, with the single exception of v. KUPFFER who assumes the epiblastic origin of the segmental duct. They differ from the account given in the foregoing pages in the mode of the formation and in the number of the tubules formed. The first point of difference is due to the

1) See the reference under Selachia (p. 390).

fact that the authors probably overlooked the earliest phases of formation, which take place, as shown above, in a stage very young, but not younger in comparison than that in other Anamnia; for the formation follows *the metameric segmentation of the mesoblast* in the anterior region. In later stages, the tubules and the intersomitic portion of the collecting duct repeated in sections of a series appear, indeed, like the cross-sections of a longitudinal furrow or groove of the lateral plate, the lips of which are fused at certain points, as described by SHIPLEY and GOETTE (see my figs. 66-74).

The number of the tubules and nephrostomes varies according to the stages of development. And if some stage or stages are overlooked, it must necessarily lead to an erroneous conclusion. This is the probable reason why the statements of the writers with reference to the number differ.

Indeed, the anterior extremity of the pronephros has already, from the first appearance, the features of a rudimentary organ; the first pair of the tubules can not be observed at the same time with the following five pairs, except by extremely good luck. In some embryos of Stage III, we see occasionally the collecting duct alone in front of the first tubule, so that we are led to infer that there were some pairs of tubules in front of the present first pair, which have degenerated during the course of the ancestral history.[1]

As is seen above, all investigators who have been occupied with the study of the development of *Petromyzon* agree in describing only one pair of glomeruli. SHIPLEY says "there is only one glomerulus on each side, stretching on each side of the

1) I have stated above that in the earliest part of Stage III, the anterior extremity of the left collecting duct presents a conical protuberance (see the foot-note on p. 329).

alimentary canal extending through about the same space as the glandular part of the kidney. Each glomerulus is a diverticulum of the peritoneum, which generally becomes sacculated;....." (p. 21). The statements by GOETTE confirm SHIPLEY's, and my results also agree with theirs. However, this is not all of the vascular system of the pronephros but represents a posterior portion of it, the anterior part having disappeared entirely (see p. 368).

No previous writer on *Petromyzon* has described such early stages as given above in the development of the pronephros, nor has any one remarked the temporary existence of the pronephric tubules in the branchial region as well as in the region of the segmental duct. I will, therefore, extend the comparison over the allied groups such as Myxinoids and *Amphioxus*, and higher Craniota to verify the new facts.

With reference to the development of the nephric organ in Myxinoids, there is a great deal of information which we owe to the unwearied labors of W. MÜLLER, SEMON, WELDON, and others[1]. They had, however, no opportunity to observe the earliest stage of the embryos. Recently our knowledge on this subject has been greatly augmented by the new works of PRICE, DEAN, and MAAS.

PRICE ('97) worked out the early development of the pronephros observed in a few embryos at different stages of *Bdellostoma stouti*. According to him "the first indication of the system occurs here in the eleventh segment (of spinal ganglion), and consists of a simple thickening of the somatic layer of the coel-

1) I have not seen the paper by J. MÜLLER.

omic epithelium, which extends through seven sections,..... the thickening has not been caused by a proliferation of cells, but certain cells having assumed the form of columnar epithelium, while the adjoining cells retained the form of flat epithelium. .....later an evagination will here take place, to form a segmental tubule" (p. 209). These evaginations are connected with one another "by a streak of columnar epithelium, which in transverse section resembles the first tubule anlage, except that there is no concavity on the lower surface; this is the segmental (collecting) duct." "The union between the duct and tubules is," in another place he says, "primary and not secondary" (*loco cit.*, p. 210).

The pronephros in *Bdellostoma* comprises, according to PRICE, 69 segments (spinal ganglions). As it begins at the transverse plane opposite the eleventh spinal ganglion, it is inferred that the pronephros in *Bdellostoma* is extended over the whole length of the branchial region. But "the excretory system disappears through the greater part of this region before the gills are formed" (*loco cit.*, p. 217).

The segmental duct (in *s. str.*) is, according to the author, brought about by the rudiments of the hinder 20 degenerated tubules (in his Stage C); the number of the declining tubules increases by stages: in Stage A, there are two; in Stage B, nineteen; and in Stage C, twenty.

This account is thus in close agreement with that given in the present work, excepting a slight difference as to the origin of the collecting duct and as to the number of the tubules. In *Bdellostoma*, the collecting duct develops out of the Anlagen independent from that of the tubules, while in *Petromyzon*, as stated in the foregoing description, the Anlage in a mesoblastic

somite develops solely into the tubule, and by the secondary union of the tubules' ends, the collecting duct is brought about.

As regards the number of the tubules, there are, in *Petromyzon*, only two pairs in the branchial region instead of twenty in *Bdellstoma*. The number is, however, of secondary importance; it varies with the stages of embryos and possibly with individuals, and naturally more with the embryos of different families. This numerical variation is readily explained by the degenerating tendency of the tubules.

PRICE has made out the segmental evaginations of the dorsal corner of the coelomic cavity corresponding to the nephromeres; they are called by him the "coelomic pockets." In *Petromyzon*, I have found a series of solid knobs on the visceral layer of the *intermediate cell-mass*, which are transformed into the segmental folds of epithelium, forming then the direct continuation of the peritoneum. Thus the coelomic pocket in *Bdellostoma* and the coelomic projection in *Petromyzon* are apparently very similar structures; the two, however, differ from each other in origin and in fate. The former (coelomic pocket) is constructed by the parietal and visceral layers of the *lateral plate*, while the latter (coelomic projection) is the product of only the visceral layer of the nephrotome, the ventral half of the *segmented part of the mesoblast*. The coelomic pockets become the Malpighian body, and the coelomic projections give origin to the radix of the mesentery, from which the gonad-cells and the mesonephric tubules are derived. Nevertheless, these two structures are, I believe, homologous. PRICE's statements on the derivation of the coelomic pocket from the two peritoneal layers, are not as clear as is desirable, and its partition from the body-cavity might, it seems to me, represent the uppermost peritoneal partition which

soon disappears, in *Petromyzon*, without any definite significance. At any rate, the structure represents "parts of the original segmental coelome, that is, the nephrotome," an unmistakable fact which is denied by PRICE.

The embryos of *Myxine* which formed the materials of the valuable works by MAAS are too old to be compared with those of *Petromyzon* used in the present work. But the results obtained by the author differ from those of PRICE in an important point, namely, in the derivation of the mesonephros.

The pronephros and mesonephros are, according to PRICE, different parts of the same organ. "If the organ in question could only be a pronephros alone, or mesonephros alone," says PRICE "I should unhesitatingly pronounce in favour of its being a pronephros" (*loco cit.*, p. 120). And he proposes to call "the entire embryonic kidney *holonephros*." With RABL, MAAS, and others, I hesitate to accept PRICE's conclusion; for there are, as may be inferred from his statements, great gaps not only between the Stages B and C, but also between Stage C and the adult. The formation of the mesonephros takes place in *Petromyzon* only at a stage much advanced, in which the processes of the formation and degeneration of the pronephros go on in much the same manner as in *Bdellostoma*, and it is open to doubt if the mesonephros might not appear in later stages which were lacking among PRICE's materials.

Up to the oldest embryo observed by PRICE there were neither glomeruli nor bloodvessels of a definite form, although there were found in the splanchnopleure some vessels whose position seemed to suggest their corresponding to the glomeruli of Selachia and *Amphioxus*; "but they do not have any relation to the openings of the tubules, nor have they any direct connection

with the aorta" (*loco cit.*, p. 213). The glomerulus figured in his Taf. 17, fig. 12 (*gl*) corresponds, I think, with a part of the glomerulus in *Petromyzon*.[1)]

As is very well known, the independent studies of WEISS ('90) and BOVERI ('92) on the branchial chamber of *Amphioxus* gave a new direction to the morphological investigation of this field. A number of external openings of ciliated tubes is found at the dorsal corner of the peribranchial chamber of *Amphioxus*. Through the morphological study and physiological experiments this organ-system is demonstrated to be the excretory apparatus, or "Nierencanälchen," as BOVERI calls them, of *Amphioxus*.

BOVERI counted 91 "Nierencanälchen" in an individual 4 cm. in length and possessing 183 gill-bars on the right side. In the adult he counted about 180 of the "Nierencanälchen"; the number is, however, by no means constant, but varies within a certain limit.

In the middle region of the branchial chamber, a "Nierencanälchen" has 3 or 4 "Seitentrichter," and 2 "Endtrichter;" such is the most complete one. It becomes gradually simplified both anteriorly and posteriorly, until it is at last represented by a short single tubule, as seen in Taf. 33, figs. 9 and 13, given by BOVERI. The tubules in the anterior and posterior part of the system thus show a sign of degeneration, as in the case of the pronephros of Cyclostomata.

1) DEAN has published two papers on the development of the Californian Hag ('98 and '99); these excellent works contain merely the general account of the course of the development in surface view. We may expect that the full account will throw much light on the ontogeny of Craniota. There stand, in the account given by him in these works, the important facts that the "pronephric tubules are apparent in connection with all the mesoblastic somites" ('98, p. 274) and that the pronephros is extended far backwards, beyond the anal region, into the tail ('99, p. 272). It would be highly desirable to observe the pronephric tubules of the Hag in relation to the myomere, and not to the spinal ganghia alone, as PRICE has done.

These nephric tubules receive, according to BOVERI, the blood from the aorta, which gives two branchlets to each nephric segment. These branchlets form in each segment a network in the neighbourhood of, and winding around, the nephric tubule; it is this network that BOVERI calls *glomerulus*.

From the structure, the position, the segmental arrangement, the physiological function, and the relation of the blood-vascular system to this system of organs, BOVERI regards the latter as a primitive form of Vertebrate nephric organ and homologised it particularly with the pronephros of Craniota. The points of difference which exist between the "Nierencanälchen" of *Amphioxus* and the pronephros of Craniota, have been smoothed away by the author's masterly arguments. The first of these points is the want of the segmental duct in *Amphioxus*; but this is represented, according to BOVERI, by a part of the peribranchial chamber. The second is the relation of the nephric segments to other systems of organs. The "Nierencanälchen" is branchiomeric while the pronephros of Craniota is myomeric, in arrangement. But this difference is looked upon by him as only apparent; for the number of gill-slits first formed agrees with that of the muscle-segments in the same region; this is sufficiently demonstrated by the figure given by WEISS (*loco cit.*, fig. 3).

Thus the author has brought the "Nierencanälchen" of *Amphioxus* into perfect harmony with the pronephros of Craniota. Some additional light is now, I believe, thrown from the side of Craniota by the facts obtained in Cyclostomata, the lowest class of Craniota. This harmony will be brought out more in discussing the development of the pronephros in Selachia, Teleostei, and Amphibia, which will be treated further on.

Thanks to the labors of many eminent investigators, the early development of the Selachian pronephros has been, as is well known, fully studied, so that the facts gathered from this field are well adapted to be compared with those from other groups. I have found, in the present investigation, many important points running parallel with the development of the Selachian pronephros. I may then be allowed to compare my own results in *Petromyzon* with those already arrived at in Selachia. Reference will, however, be limited to those works which are sufficient to verify the points I wish to bring out.

Through the excellent work of RUCKERT ('88) we can best learn the origin of the pronephros in Selachia. "Die erste Anlage der Vorniere" is recognised "in Form einer gegen den Ectoblast gerichteten Vorbuchtung des parietalen Mesoblasts." This Anlage is first brought about by the thickening of the parietal layer of the mesoblast, which is found "in den Bereich des segmentirten Mesoblasts, d.h. Somiten" (p. 209); this thickening is called by the author "Segmentalwulst." The foot-note also runs as follows: "Der Ursprung des Segmentalwulstes reicht ventral bis zu der Stelle herab, wo die Somiten in den unsegmentirten Mesoblast der Peritonealwand übergehen" (p. 209). The "Segmentalwulst" is so called because it is noticed as the segmental thickening of the parietal mesoblast of which RUCKERT recognised, in his Stad. II[1], six for *Torpedo* and four for *Pristiurus*, stretching over a corresponding number of the myotomes. The first indication of the pronephros is expressed, in Selachia also, segmentally in the segmental part of the mesoblast at the stage in which the metameric segmentation of the mesoblast is still going on, and

1) The embryos in the stage have 25-27 somites.

the myotome is not yet cut off from the lateral plate, just as in *Petromyzon*. The foremost of them is found in the hind part of the third or fourth body-somite. The development of the Anlage in each segment agrees also with that in *Petromyzon*; for he says: "Der Segmentalwulst zeigt in vorliegende Stadium (Stad. 1) regelmässig die stärkste Entwicklung in seiner mittleren Abschnitt, also ungefähr im Bereich des dritten des ihm gehörigen Somiten, und verjüngt von da allmählich nach seinem vorderen und hinteren Ende zu....." (*loco cit.*, pp. 210).

The account given by RÜCKERT is essentially confirmed by later investigators such as VAN WYHE ('89)[1] RABL ('96), and others, although they differ from one another in the interpretation of the facts and in some unimportant points. RABL looks upon the Anlage of the pronephros (his Vornierenwulst) as the ventral portion of the somite just as RÜCKERT does, while it is, according to VAN WYHE, the product of the lateral plate (his Hypomer). This is, as it seems to me, not a contradiction in the facts, but in the terms used; for VAN WYHE states: "Da nun der Pronephros, wie spätere Entwicklungsstadien zeigen, ein Produkt der Seitenplatte ist, während der unmittelbar dorsal davon liegende Theil des Mesoderms zur Mittelplatte gehört, ist die Segmentirung des Mesoderms bei Selachiern also nicht auf die Myotomplatte beschränkt, sondern erstreckt sich auch auf die Mittelplatte und den dorsalen Theil der Seitenplatte" (*loco cit.*, pp. 474-475). The fact is, therefore, no other than that the portion of the mesoblast dorsal to the ventral limit of the Anlage of the pronephros undergoes segmentation, and the portion ventral to this point remains unsegmented, constituting the lateral plate. I will, in

1) The embryo, in which the first traces of the pronephros is seen, is, according to VAN WYHE, in a stage with 27 somites, whereas RABL has seen in an embryo of *Pristiurus* with 25 somites.

this place, not go further, but return in future pages to the discussion of this point. It is, however, safe, I believe, to regard this portion of the mesoblast as a part of the somite.

VAN WYHE found the foremost pronephric segment in the third body-somite (his Rumpfsegment), and RABL states that the Vornierenwulst begins in the seventh somite formed (his Gesammtsegment). According to RABL, however, VAN WYHE's third Rumpfsegment corresponds to his seventh Gesammtsegment. To verify this fact RABL has extended the comparison over *Petromyzon*, and found that in this case also the pronephros begins in the seventh somite ; but the pronephric tubule in that somite is, as noticed above (p. 380), not the anteriormost of the tubules in his sense, but of the persistent tubules.

VAN WYHE noticed five of the pronephric segments for *Raja*, and three for *Scyllium* and *Pristiurus* ; while RABL counted eight Vornierenwülste for *Raja*, and four for *Pristiurus*. The results in *Petromyzon*, therefore, best agree with those made out by RÜCKERT in *Torpedo*.

The authors agree in deriving the collecting duct from the lateral extremities of the pronephric Anlagen, where they become confluent.

RÜCKERT has observed, in *Pristiurus*, as well as in *Torpedo*, the *secondary connection* of the Segmentalwulst with the epiblast, which has led him to believe in some contribution of epiblastic cells to the formation of the pronephros, while VAN WYHE and RABL deny this. I have found the same connection in *Petromyzon*, but I have found no sign of the contribution of epiblastic cells to the formation of the pronephros. The phenomenon is temporary in both Selachia and *Petromyzon* ; it takes place in Selachia, according to RÜCKERT, in his Stad. II,

and already in his Stad. III, a space is seen between these two structures.

The degeneration of the tubules in Selachia runs a course parallel with that mentioned under *Amphioxus* and Cyclostomata. As seen above, the Anlagen of the pronephros are developed most vigorously in the middle part of the pronephros, as in the case of *Amphioxus* and Cyclostomata; and degeneration begins at the cranial and caudal extremities as there.

VAN WYHE says that the degeneration consists in a confluence (Verschmelzung) of the ostia. According to RUCKERT, the Vornierenfalte becomes simply flattened out in the cranial part of the pronephros. The reduction in the caudal part is noteworthy: the Anlagen are here constricted off from the mesoblast and converted into the anteriormost section of the segmental duct. Only the middle (the third) diverticulum (in *Torpedo*) persists in communicating with the body-cavity and becomes the *ostium abdominale.*

In *Petromyzon,* I have unfortunately failed to observe accurately the manner of degeneration of the tubule in the cranial part. It is however probable that it begins either from the blind tip of the tubule (the first tubule), or by obliteration of the nephrostome (the second tubule). In the caudal part, the collecting duct is constricted off from the lateral plate by obliteration of the tubule and constitutes the foremost section of the segmental duct, in precisely the same manner as in Selachia. The difference is: in *Petromyzon* the communication with the body-cavity is retained by the three middle nephrostomes, while in Selachia, it is through only the middle one, that is, the *ostium abdominale.*

The segmental duct becomes apparent in an embryo with

35 (VAN WYHE), or 34 to 35 (RABL) somites. The anterior small section of the duct is formed, as just stated, in the same manner in *Petromyzon* and Selachia. The mode of formation of its posterior larger portion in Selachia differs from that of *Petromyzon*. RÜCKERT ('88) and VAN WYHE ('88, '89, '98) believe that it is the product of the epiblast[1], while RABL maintains its purely mesoblastic origin. At any rate, the posterior tip of the duct or the cord is sharply pointed and connected firmly with the epiblast throughout its growth until it opens into the cloacal cavity, which is effected, according to VAN WYHE and RABL, in the embryo with 83 to 84 somites. It can be inferred from VAN WYHE's figs 7*a* and 7*b*, that this communication is found in a plane vertical to the thirty-eighth Rumpfsegment[2]. In *Petromyzon*, the duct, being formed of a series of abortive pronephric tubules, has no genetic relation to the epiblast except in the cloacal region where the duct seems actually to receive cells from the epiblast, as fully stated above (p. 366).

The nephric arteries of Selachia which were discovered by PAUL MAYER without reference to their relation to the pronephros, were studied by RUCKERT and their true nature was pointed out by him. There are six of them in *Torpedo* corresponding to the number of the nephric segment; they are, however, not somitic but intersomitic in position. The vessels not only pass through the nephric fold, but throw a solid process, the interior of which consists of round or spindle-shaped cells. This is, according to RÜCKERT, the equivalent of the pronephric glomerulus of Amphibia described by FÜRBRINGER. The development and decline of these vessels go on parallel with those of the pronephric diver-

1) I will return to this point again in future pages.

2) According to RABL's counting, this somite corresponds to his forty-second somite.

ticula. The vessels in the cranial as well as in the caudal part of the pronephros are weaker than those in the middle (the third and fourth); only the latter vessels develop further and become the vitelline artery. VAN WYHE confirms RÜCKERT's account and has described three vessels in *Pristiurus*. In addition to these, VAN WYHE has pointed out the very small segmental vessels on the left side, which go not to the intestine, but to the body-wall. They are not equivalent to the intestinal vessels on the opposite side. One of them gives a branchlet to the glomerulus which sends out, in its turn, a branchlet to the cardinal vein. The homologous vessels on the right side are to be seen coming out of the root of the vitelline artery. BOVERI remarks that the vessels of PAUL MAYER present many points of harmony with the branchial vessels in *Amphioxus*. RABL agrees essentially with the account given by RÜCKERT and VAN WYHE, but denies the existence of a glomerulus. According to RABL, the structure called the glomerulus by RÜCKERT does not fulfil the conditions of being a glomerulus; he says: "Eine einfache Ausbuchtung einer Arterie ist noch keine Gefässschlinge, geschweige denn ein Glomerulus" (*loco cit.*, p. 668).

Most of the early investigators, who observed the development of the Teleostian pronephros, believe it to be mesoblastic in origin. There are very few writers as RYDER ('87), and BROOK ('88), who derive the segmental duct from the epiblast. According to OELLACHER ('73), GOETTE ('75 and '88), FÜRBRINGER ('78), and HOFFMANN ('86), the first Anlage of the pronephros is brought about by the evagination of the parietal layer of the mesoblast at the level of the junction of the somite with the lateral plate, forming thus a longitudinal groove on each side,

which is subsequently constricted off from the body-cavity. This takes place at first in the middle region of the body, whence it proceeds both anteriorly and posteriorly.

OELLACHER observed that the Anlage is converted into a longitudinal canal or the segmental duct, being completely shut off from the body-cavity in both the anterior and posterior parts. The anterior section of the duct is much swollen and transformed into the pronephric chamber. From the dorsal aorta, a pair of branches is given off which pushes into the pronephric chamber, pressing against its median wall and giving rise to a pair of the glomeruli. This portion of the duct becomes coiled up and constitutes the pronephros.

GOETTE's view somewhat differs from the account given above: the anterior end of the longitudinal groove is not completely closed from the body-cavity, but leaves awhile the communication with the latter, which is, according to GOETTE, the morphological equivalent of the nephrostomes of the Amphibian and *Petromyzon* pronephros. Opposite this nephrostome, he says, the glomerulus is formed by evagination of the visceral peritoneum and projects freely into the body-cavity. This portion of the peritoneum together with the nephrostome is constricted from the rest of the peritoneum; the coelomic cavity thus shut off is converted into the pronephric chamber.

This view is essentially confirmed by subsequent writers such as FURBRINGER ('78), HOFFMANN ('86), and others, although HOFFMANN differs in his view of the mode of the formation of the glomerulus.

According to the results recently arrived at by FELIX[1] in

1) I know his paper only by the abstract in the Jahresberichte über die Fortschritte der Anatomie und Physiologie, N.F. Bd. III. '97.

the embryos of Salmonidæ, the earliest traces of the pronephros consist, in embryos with 11 pairs of the somites, of five solid proliferations of the lateral plate which is already cut off from the somite. These proliferations, being coincident with the caudal half of the third to seventh somites, are strictly metameric in arrangement and are regarded by the author as the rudimentary pronephric tubules. These tubules soon become confluent with one another to form a single outgrowth of the lateral plate, which is called by the author the "primäre Vornierenfalte." The "primäre Vornierenfalte," which passes over into the parietal and visceral layers of the lateral plate, undergoes a longitudinal constriction (the "sekundäre Vornierenfalte") by which it is divided into the dorsal and ventral parts. From the former, the anterior section of the segmental duct originates, while the latter is transformed into the pronephric chamber. By stages, the dorsal part wanders laterally, and the ventral part travels medianwards. At the same time, these parts are separated from each other, leaving the communication at only one point, which is called the "Pseudonephrostom."

This phase of the development of the pronephros observed by FELIX is, as I believe, undoubtedly earlier than that looked upon by the previous authors as the earliest indication of the pronephros.

At the time when the Anlagen of the pronephros are converted into the "Vornierenfalte," the Anlage of the caudal continuation of the segmental duct, becomes apparent in the eighth to the tenth somite; it is brought about by the division of the primary lateral plate (lateral plate in the ordinary sense) into (1) the secondary lateral plate (lateral), (2) the segmental duct (middle), and (3) the Anlage of the "Stammvenen" (median). This pro-

cess proceeds posteriorly until the duct comes to lie close to the rectum (Enddarm).

FELIX thus observed the segmental Anlage of the pronephros in its glandular part, and derives the rest of the system from the proximal margin of both the parietal and visceral layers of the secondary lateral plate; he has observed neither the posterior growth, nor the epiblastic origin, of the segmental duct.

The pronephric chamber which results from the confluence of the five pronephric tubules, is not homologous, according to FELIX, with that of Amphibia, in which the chamber should be a constricted part of the body-cavity into which the tubules open.

Quite recently, SWAEN and BRACHET ('99) have published a paper on the early development of the mesoblastic organs in Salamonidæ. Although my manuscripts were nearly finished, when I saw this interesting paper, I must here refer in a few words to it[1].

The authors found the first traces of the pronephros under the fifth somite, of two embryos, one of which was in the stage of 11 somites, and the other of 13 somites. It is not the product of the parietal layer of the lateral plate only, but is formed, as FELIX believes, by the proximal portion of both the parietal and visceral layers of the secondary lateral plate (l'extremé interne de la plaque latérale secondaire[2]). The internal cavity enclosed by the pronephros is, therefore, not the diverticulum, but a part of the body-cavity.

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1) I am much indebted to my friend, DR. A. OKA, who read the paper for me.

2) According to the authors, the "plaque latérale primitive" is divided into the "plaque latérale secondaire" and the "masse intermédiaire;" therefore, the "plaque latérale secondaire" corresponds to the lateral plate itself of *Petromyzon*.

The Anlage of the pronephros is laid in exactly the same manner from the fourth somite to the cloacal region. Under the anterior three somites from the fourth to the sixth, the Anlagen are developed into the pronephric chamber; the Anlagen posterior to these are all transformed into the "canal excréteur," as they call the segmental duct, and they have come to the conclusion that the "canal excréteur" of the pronephros has the morphological value of a rudimentary pronephric chamber.

The facts given in the last two papers, are thus in close accordance with one another as well as with those given by myself in the foregoing pages. Differences between their results and mine are that the authors derive the system from the lateral unsegmented mesoblast, and that both the parietal and visceral layers of it partake in the formation of the system. As has been stated in the descriptive part, this derivation is only apparent; a little further study shows that only the parietal layer gives rise to the system, and this part of the layer belongs to the somite. Indeed, this part appears to form, for some time, the proximal portion of the lateral plate, being early cut off from the rest of the somite. It must be remembered that this separation is not the separation of the lateral plate from the somite, but that of the Anlage of the pronephros from the rest of the somite; or, the result of the development of the pronephros. It is merely for a physiological reason that this development or separation of the pronephric Anlage goes on earlier than, for instance, in Selachia, it performing in Teleostei the actual excretory function. This will be understood easily, when a comparison with other groups is made further on.

It has been a well known fact that the development of

Amphibia shows, in several respects, a parallel course with that of *Petromyzon*. Careful observations on the development of Amphibian pronephros, adduced by recent investigators, have intensified this similarity with the exception of a few points which are, however, probably of secondary importance.

Most authors who have worked on the Amphibian development agree in deriving the entire system of the pronephros from the parietal layer of the mesoblast only, and in regarding it as arising originally as a common pouch, the anterior part of which is divided secondarily, by a partial closure of the peritoneal communication, into a number of the pronephric tubules.

This view has been advanced by earlier authors such as W. MÜLLER ('75), GOETTE ('75), FÜRBRINGER ('78), HOFFMANN ('86), and others. The stage at which the pronephros appears coincides exactly with that in *Petromyzon*, as MAX FÜRBRINGER says in his well known work: "Die erste Entwicklung der Vorniere und ihres Ausführungsganges findet hier nach der Scheidung des Mesoderms in Urwirbel und Seitenplatten statt und folgt unmittelbar der beginnenden Sonderung der ersten in einzelne Urwirbel amd der Spaltung der letzteren in Haut- und Darmfaselplatten. Embryonen von *Rana temporaria* von *circa* 2.5 Mm. Länge und von *Triton alpestris* von *ca.* 2.0 Mm. L. entsprechen diesen Stadium" (p. 3)[1]

MOLLIER ('90) has made out the segmental Anlage of the Amphibian pronephros, having worked with the embryos of *Triton*,

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1) The nephrostomes are found, according to the author:

2 in *Salamandrina maculata*,	3 in *Rana temporaria*,
2 in *Triton alpestris*,	3 in *Bombinater ignens* (GOETTE), and
2 in *Siredon pisciformis*,	4 in *Coecilia rostrata* (SPENGEL).

Bufo, and *Rana*. His accounts confirm, as a whole, those given by RÜCKERT for Selachia above referred to, but differ somewhat from those of most other authors who have worked on Amphibian pronephros. MOLLIER states as follows: "Wir sehen hier ebenfalls zuerst eine solide, von dem Mesoblast ausgehende Anlage, deren Structur anfänglich schwer zu erkennen ist und erst mit dem Hohlwerden, wie bei den Selachiern, klar hervortritt. Dann finden wir, dass hier zwei resp. drei getrennte Canälchen vorhanden sind, die von den Somiten in convergenter Richtung ausgehen und erst nachträglich untereinander vereinigen zu einem Längscanal, von dem aus die Vornierentrichter in die Leibeshöhle führen" (*loco cit.*, p. 229).[1] The author derives in this wise the pronephric tubules, exactly as in the case of *Petromyzon*, from the segmented part of the mesoblast only.

MOLLIER's accounts are for the most part in close accord with the results given by FIELD ('91, p. 282), who, one year later independently of MOLLIER, began with Anura, and extended the work over Urodele Amphibia. In one point, their results differ widely; but "the difference is," it seems to FIELD, "apparent rather than real." According to MOLLIER, the nephrostomes communicate with the cavity of the myotome, the myocœlome of VAN WYHE; this is denied by FIELD, who believes that "the pronephric tubules have to do with the ventral segment of the mesoderm" (*loco cit.*, p. 283). It seems to me that this "ventral segment of the mesoderm" corresponds to the pronephrotome of VAN WYHE in Selachia or to "l'extremé interne de la plaque latérale secondaire" of SWAEN and BRACHET in

1) It seems that the earliest traces of the pronephros are perceived in an embryo younger than that with 7 somites.

Teleostei, and I agree with the view of RUCKERT, here represented by that of MOLLIER.

MOLLIER and FIELD agree with each other in assigning three pairs of the tubules for *Rana* and *Bufo*, extending from the second to the fourth somite, and two pairs for *Triton* (MOLLIER) and *Amblystoma* (FIELD), covering the third and fourth somites.[1] In addition to these, MOLLIER observed occasional occurrences of the third tubule in *Triton*, which is, according to FIELD, not equivalent, as MOLLIER maintains, to the third tubule in *Bufo* and *Rana*, because the additional third tubule in *Triton* is found in the fifth somite, while the third tubule in *Rana* and *Bufo* is under the fourth somite. According to SEMON, there are ten pairs of the tubules on either side of the body in *Ichthyophis*.

A pair of glomeruli has been made out in Amphibia; the structure is connected by special vessels with the dorsal aorta on one hand and with the cardinal vein on the other. This branch of the aorta is believed by FIELD to correspond to a part of MAYER's vessels in Selachia. Beside these, there is no vessel arranged segmentally or otherwise.

The section of the body-cavity corresponding to the pronephric stretch is gradually expanded, and is shut off temporarily from the rest of the cavity by a close contact of the parietal and visceral layers of the cœlome; this part of the cavity is, according to GOETTE, homologous with the pronephric chamber in Teleostei and with the homologous structure in *Petromyzon*, which is called by him the "peritoneale Scheidewände."

The so-called ventral portion of the Amphibian pronephros is, according to MOLLIER, brought about by the separation of

1) According to FIELD, MOLLIER's first body-segment in *Triton* corresponds to his third somite in *Amblystoma*.

the ventral portion of the pronephric Anlagen from the dorsal, which latter is differentiated into the tubules and constitutes the dorsal portion of the pronephros. The ventral part of the Anlagen separated from the dorsal retains anteriorly its connection with the anteriormost tubule and posteriorly with the segmental duct. It is prolonged and bent out anteriorly in front of the dorsal part. "MOLLIER's description is," FIELD says, "substantially in accord with my own observation,".....(*loco cit.*, p. 286). This feature of the duct shows, it seems to me, a close resemblance to the anteriormost section of the Teleostean segmental duct which is, as above referred to, bent in the same fashion.

The segmental duct arises, according to previous writers, as a longitudinal common furrow oft he parietal peritoneum, which furrow is later constricted off from the mother-layer and becomes converted into a long canal. MOLLIER has observed the segmental duct transformed directly from the mesoblast, just like the glandular part of the pronephros, in the two somites behind the pronephros. Whether the greater remaining part of the duct is formed likewise by differentiation of the mesoblast, or by a backward growth of the hind end of the duct first formed, he could not decide with certainty; but the observations of FIELD elucidate this point.

"The segmental duct arises," FIELD says, "throughout its entire length by a proliferation *in situ* of the somatopleure" (*loco cit.*, p. 223). The author has observed neither its epiblastic origin nor a free growth of its posterior end, except in the cloacal region where it "grows across the cloaca free from adjacent tissue" (*loco cit.*, p. 223). In Stage v, the cloacal opening is seen. This opening is found, in *Rana* and *Bufo*, in the vertical plane with the middle of the twelfth somite, whereas it is below the twentieth somite in *Amblystoma* (FIELD).

The duct is segmental in origin. FIELD says: "I believe I am justified in concluding that the segmental duct between Somites V, and IX, arises *in situ* from a thickening of the somatopleure serially equivalent to that from which in the anterior region the pronephros is developed" (*loco cit.*, p. 219). There are no other Vertebrata which agree more with *Petromyzon* with reference to the development of the segmental duct, than Amphibia. Indeed, here as there, the segmental duct is of segmental origin and is to be looked upon, as seen in *Petromyzon*, as the continuation of a series of abortive pronephric tubules in the posterior region.

Authors who have observed the epiblastic origin of the segmental duct in Amphibia are very few. VON PERENJI ('87) has published the results of his study on *Rana esculenta*, but his note is unfortunately very short[1]. This view is opposed, so far I am aware, by almost all recent observers. After bringing the results by him into harmony with those by RÜCKERT in Selachia, MOLLIER says: "Im einen Punkte weichen die Amphibia von Selachiern ab, dass die Vorniere mit dem Ektoblast in keine nähere Beziehung tritt. Allerdings heftet sie besonders in den Stadien, in welchen sie voluminöser erscheint, dem Ektoblast oft in affallend inniger Weise an. * * * Doch lässt sich stets eine scharfe Grenze beiderlei Blätter ziehen, wenigstens bei *Bufo*, wo die Ektoblastelemente durch ihren Pigmentgehalt deutlich gekennzeichnet sind" (*loco cit.*, p. 229).

The historical review undertaken in the foregoing pages shows the agreement to a large extent of the results arrived at in several groups of Anamnia. Some points of disagreement are naturally met with; but these are, I believe, only apparent.

1) I have not seen the paper by Brook.

In the groups above referred to, the first indication of the excretory system becomes apparent at a stage in which some mesoblastic somites are formed and the metameric segmentation of the mesoblast is going on. This is the Anlage not of the segmental duct, but of *the pronephros*. A single exception is found in *Bdellostoma*, in which the early traces of the system become visible, as we learn from PRICE, at a stage much more advanced than in other Anamnia, that is, at the stage in which the sclero-myotome is cut off from the rest of the mesoblast and mesenchymatous cells fill up the spaces between organs and organ-systems.

I have endeavoured to reconcile the points, in which the views of the previous authors diverge from one another, under the following three headings:—

A.—The Anlage of the Pronephric Tubule is the Product of the Mesoblastic Somite and not of the Lateral Plate.

The view that derives the pronephros from a single common groove formed either of only the parietal, or of both the parietal and visceral layers of the unsegmented mesoblast (the lateral plate), is advocated by most of the authors who have worked on the development of *Petromyzon*, Teleostei, and Amphibia. This is due probably to the early separation of the sclero-myotome from the rest of the mesoblast in these groups. In them the Anlagen of the pronephros (or the nephric segments) together with the lateral plate are cut off from the sclero-myotome and form, for some time after this separation, the proximal portion of the lateral plate. It must be borne in mind that this separation is not the separation of the lateral plate, but of the nephrotome, from the sclero-myotome. This is, therefore, a

step in the differentiation of the mesoblastic somite, and because the distal (ventral) portion of the latter happens for a time to be continuous with the lateral plate, we are not justified in concluding that it is derived from the lateral plate, which, as we know, never undergoes segmentation.

It is a significant fact that in Selachia and Amniota, in which the pronephros does not function as the actual excretory organ, this separation of the mesoblast into the sclero-myotome and the nephrotome is not effected so early as in the above groups, but takes place only at later stages, with the first differentiation of the mesonephros. This consideration makes it reasonable to conclude that the early separation of the mesoblastic somite into the proximal and distal portions is caused by physiological necessity and has no morphological significance[1].

The case of *Lacerta agilis* is very instructive. According to HOFFMANN ('89), the Anlagen of the pronephros in this animal are, in the most anterior segment, cut off from the myotome (sclero-myotome) and remain connected with the lateral plate just as in *Petromyzon*, Teleostei, and Amphibia; whilst in all the following portion, they are the actual diverticula formed segmentally in the parietal layer of the lower part of the somite, as in other Reptilia (pp. 264 and 265). We thus see the two modes of separation in one and the same animal.

All recent authors agree in thinking that the Anlage of the pronephros is expressed in itself segmentally and is strictly myomeric. Now the question arises: How many parts are to be distinguished in the mesoblast, and to what part of it does the Anlage of the pronephros belong?

VAN WYHE ('89) has distinguished, in Selachia, three por-

1) This view is grounded upon the suggestion of PROF. MITSUKURI.

tions of the mesoblast which are called by him the "Epimer," "Mesomer," and "Hypomer" respectively. The epimere of VAN WYHE corresponds solely to the myotome; his mesomere comprises the Anlage of the mesonephros and the sclerotome; and the hypomere consists of the Anlage of the pronephros, the genital gland, and the lateral plate. The epimere, mesomere, and the dorsal part of the hypomere undergo the metameric segmentation, while the remaining portion of the hypomere remains unsegmented. According to VAN WYHE, the dorsal segmented part of the hypomere is, therefore, the product of the lateral plate (see p. 389). It seems to me that this division of the mesoblast does not agree with the facts observed in *Petromyzon* and the other Anamnia above referred to; for the mesoblast in these groups consists, in early stages, of two portions: (1) the segmented, and (2) the unsegmented, and of nothing more, just as RÜCKERT ('88) and RABL ('88, '96) have remarked. According to RÜCKERT and RABL, the segmented portion—the somite—comprises the myotome and the sclerotome; the pronephros and the mesonephros are derived from its ventral (distal) portion, which is called by RUCKERT the "Nephrotom" ('88, p. 272).

In *Petromyzon*, these two portions of the mesoblast, the segmented and the unsegmented, are, in early stages, clearly distinguished, being histologically different (see p. 315). The mesoblast in such an undifferentiated state is almost entirely occupied by the segmented portion, while the unsegmented portion is very small, being represented by the loose tissue of a few cells. Such a mesoblastic segment exactly corresponds to the somite of RÜCKERT and RABL. The proximal half of the segmented portion coincides with the sclero-myotome of those authors. It consists not only of the myotome, but also includes the sclerotome. And it is the

distal half of this segmented portion which folds out in each segment to give rise to the Anlage of the pronephric tubule on one hand and to the cœlomic projection on the other, and, therefore, corresponds to the "Nephrotom" of RÜCKERT.

The nephrotome, therefore, constitutes, in both *Petromyzon* and Selachia, precisely the same part of the mesoblast, viz. the distal (ventral) portion of the somite, through which the sclero-myotome is connected with the lateral plate.

The above early stage in the differentiation of the mesoblast, in *Petromyzon* corresponds also to the "Ursegment" of *Amphioxus* in HATSCHEK's sense ('88). By further development of it the unsegmented mesoblast is brought into light, and we can then distinguish the "Urwirbel" and the "Seitenplatte" of HATSCHEK ('88). And the ventral half of the Urwirbel constitutest in *Petromyzon*, the connecting canal between the unsegmented cœlomic cavity and the sclero-myotome, that is to say, the nephrotome. Let us now examine what part of the Ursegment of *Amphioxus* represents the nephrotome of the Craniota.

In his excellent work on "Die Nierencanälchen des Amphioxus," BOVERI ventures to solve this important question. After a discussion he comes to the conclusions:

(1) That the "Gononephrotom" in Craniota must correspond to a part of the "Urwirbel" of HATSCHEK;

(2) That the "Gononephrotom" of Craniota is homologous with the genital chambers of the adult *Amphioxus*.

But these chambers "sind ursprünglich die segmentale Verbindungscanäle zwischen der unsegmentirten Leibeshöhle und der Sclero-Myotom gwesen" ('92, p. 493). I can, therefore, ascribe no other significance to the ventral half of the segmented

mesoblast in *Petromyzon* than that it is the morphological equivalent of the "segmentale Verbindungscanäle."

It thus follows that the distal half of the segmented mesoblast in *Petromyzon* undergoes exactly the same fate as that in *Amphioxus*: it is transformed into the pronephros and the cœlomic projection or the "dorsal segmental cœlome," which latter gives rise, just as BOVERI suggests in *Amphioxus*, to the mesonephric tubules and, in the hinder region, to the genital gland.

As has been pointed out in the historical review, the relation of the Anlage of the pronephric tubule to the mesoblastic somite is the same for Teleostei and Amphibia, as in *Petromyzon*.

It may, therefore, safely be stated, that the segmented portion of the mesoblast constitutes in these groups a single integral structure until the separation of the nephrotome in continuo with the lateral plate from the sclero-myotome. This separation is, as above stated, not the separation of the somite from the lateral plate, but the differentiation of the somite into the sclero-myotome and the nephrotome, preparatory to the development of the urogenital system. The reason why the separation takes place earlier in some groups than in others, rests only on physiological grounds.

B.—The Whole System of the Pronephros of Cyclostomata, Teleostei, and Amphibia is Homologous with the Nierencanälchen of Amphioxus (BOVERI) *and not perfectly Homologous with the Selachian Pronephric System.*

I have already stated above (pp. 386 and 387) that BOVERI has brought the pronephric system of Craniota in harmony with the system of the "Nierencanälchen" of *Amphioxus*, basing his arguments on the structure, the position, the myomeric arrange-

ment, the physiological function, and the relation of the vascular system to the organ. His comparison is, however, almost entirely limited to Selachia on the side of Craniota, owing perhaps to the scantiness of the literature at that time. I accept in the main this homology, and I may perhaps extend this comparison a little further.

I will begin with the homology of the pronephros of Cyclostomata with the "Nierencanälchen" of *Amphioxus*.

It is well known that the starting point of the hepatic diverticulum from the enteric canal demarcates, in the Chordata, the respiratory section of the canal from the nutritious section of it; and, as GEGENBAUR ('78, pp. 563—581), BALFOUR[1] ('85), and others affirm, the œsophagus and stomach in the higher forms are a part of the former section, which is called the fore-gut. And the homology of the hepatic cœcum of *Amphioxus* with the liver of the Craniota, has been much strengthened by recent morphological studies and physiological experiments[2]. The results of my present study also confirm this view. I will use, therefore, this fixed point as the landmark of comparison of the two organ-systems, the pronephros and the "Nierencanälchen," and of the pronephros in different groups of Craniota.

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1) From the account of BALFOUR, I will cite the following lines:—

'In Amphioxus the respiratory region extends close up to the opening of the hepatic diverticulum, and therefore to a position corresponding with the commencement of the intestine in higher types. In the craniate Vertebrata the number of the visceral clefts has become reduced, but from the extension of the visceral clefts in Amphioxus, combined with the fact that in the higher Vertebrata the vagus nerve, which is essentially the nerve of the branchial pouches, supplies, in addition the walls of the œsophagus and stomach, it may reasonably be concluded, as has been pointed out by Gegenbaur, that the true respiratory region primitively included the region which in the higher types forms the oesophagus and stomach' (Vol. II, p. 758).

BALFOUR has also shown that the solid cord of the œsophagus in Elasmobranchii and Teleostei, is the remanent of the gill-rudiments in the ancestry (*loco cit.*, pp. 61 and 78).

2) J. A. HAMMAR, '99, '98, and GUIDO SCHNEIDER, '99.

The "Nierencanälchen" of *Amphioxus*, according to BOVERI ('92), extends over and is limited to, the whole extent of the branchial region, the posterior larger part of which covers the hepatic cœcum. The pronephros of Cyclostomata extends from the anterior body-somite to the cloaca. The anterior section of the system constitutes afterwards the glandular part represented by the pronephric tubules and is found in front of, and over, the Anlage of the liver, or in the region of the fore-gut; a certain number (two in *Petromyzon*, twenty in *Bdellostoma*) of the anterior nephric segments are found in the branchial region, and the posterior one or two segments of the glandular part (*Petromyzon*) cover the liver-Anlage. It follows that the six[1] to twenty or more pronephric tubules correspond to as many "Nierencanälchen" in about the middle one third[2] of the branchial region of *Amphioxus*, and that the "Nierencanälchen" lying posterior to this point are represented, in Cyclostomata, by a number of the rudimentary tubules which are converted into the segmental duct.

The "Nierencanälchen" are not put in communication with one another by the collecting duct, as in the pronephros of Cyclostomata, but open to the exterior segmentally. I have stated in the descriptive part (p. 333) that the free extremities of the pronephric tubules in *Petromyzon* are brought into close contact with the epiblast, so that the latter is pressed out by the enormous growth of the tubule and that this is especially the case in the first and second tubules. This fact throws light upon the homology of the pronephric tubules in *Petromyzon* with the "Nierencanälchen" of *Amphioxus*: in other words, the condition

1) The glandular part of the pronephros in *Petromyzon*, are represented by the six pronephric tubules.

2) The branchiomeres in the posterior section of the gill-basket of *Amphioxus* are afterwards added (see pp. 410-411).

seen in the "Nierencanälchen" would be brought about, if the tubules in *Petromyzon* came to open to the exterior, boring through the epiblast by the further growth of their free extremity. This intimate contact of the tubule-end with the epiblast takes place, as above mentioned, in the middle of Stage II, where the tubules have developed a little beyond the mere Anlage.

The pronephric tubules of *Petromyzon* are, in this stage, already united with one another by the intersomitic solid cord; this union is, however, not primary, but secondary. This stage presents, I think, the phylogenetic stage, in which the "Nierencanälchen" with separate external segmental openings, and the pronephric tubules with the collecting duct, diverge from each other.

By this assumption, it is not meant that in the ancestry of Chordata the tubules were closed blindly inside the epiblast; for the Anlage of the pronephric tubule might have been, in the ancestral form too, brought about by the folding of the mesoblast, to break out finally to the exterior. This perforation would become unnecessary when the secondary union of the tubules had been acquired.

Since a certain number of the "Nierencanälchen" in front of the base of the hepatic cœcum, is represented by the pronephric tubules of the glandular part in Cyclostomata, those lying over it will be homologous with the pronephric tubules which are found over and posterior to the hepato-pancreatic Anlage and converted into the anterior section of the segmental duct, being secondarily united with one another by the confluence of the free extremities of the tubules.

There is not to be seen the post-hepatic "Nierencanälchen" in *Amphioxus*. We learn from LANKESTER ('89) and WILLEY ('91) that in *Amphioxus*, the new branchial slits are added, by stages,

to the posterior end of the pharynx, so that, in later stages, the coincidence of the number of the slits with that of the myotomes is lost; and that this addition of the slits continues throughout life. The nephrotomes in these new slits have been, I think, originally coincident, in each segment, with the myotomes, from which they were cut off in early stages and have remained undeveloped until the new appearance of the added slits. It seems, therefore, probable that the branchial region of *Amphioxus* once extended over the largest portion of the enteric canal, while a very small section in the posterior part of the canal performed the nutritious function, as is seen now in the Ascidian[1]. The "Nierencanälchen" in this hinder part may represent the pronephric tubules in the post-hepatic section of the segmental duct of Cyclostomata[2].

The pronephric system of *Petromyzon* comes to have the same relations with the epiblast as the "Nierencanälchen" of *Amphioxus* at three different points: the free ends of the two anterior pronephric tubules and the hind end of the segmental duct (probably the hindmost pronephric tubule). Whilst in the greatest section of the system the communication with the exterior has been lost, these three points might have preserved it to a considerably later phylogenetic stage: the two anterior tubules playing the same physiological part as the "Nierencanälchen", of *Amphioxus*, and the posterior being employed as the only excretory pore of the system secondarily established by the union of the tubules.

In main points (with exception of the presence of the tubules in the branchial region, of the contact or connection of

1) Balfour says: "In Ascidians the respiratory sack is homologous with the respiratory tract of Amphioxus" (*loco cit.*, p. 758.)

2) See p. 108.

them with the epiblast, &c.), the pronephric system of Teleostei and Amphibia shows, as stated in the historical review, the same characters as that of Cyclostomata, so that the facts established in Cyclostomata have the same significance for the Teleostei and Amphibia.

Although such is the case in those Craniota and *Amphioxus*, the pronephros of Selachia is quite otherwise: the Anlagen of the pronephros are here formed in the mesoblastic somites posterior to the Anlage of the liver (see below), and only one or two segments of them are converted into the segmental duct (RÜCKERT). *These pronephric Anlagen in Selachia are, therefore, the morphological equivalent, not of the glandular portion of the pronephros, but of those which are converted into the segmental duct in the Craniota just mentioned.*

C.—The Segmental Duct in Selachia is not the Morphological Equivalent of the Duct of the Same Name in Cyclostomata, Teleostei, and Amphibia.

Contradictory views are met with in the derivation of the segmental duct. The results arrived at in Cyclostomota, Teleostei, and Amphibia, well agree in making it of the mesoblastic origin; there are a few authors who believe in the epiblastic origin of the duct in these groups, but their papers are not more than mere notes. In Selachia, the circumstance is reversed; I am not aware of any recent author other than RABL, who advocates the mesoblastic origin of the Selachian segmental duct. The facts given by RABL are, however, not the same as those observed in the groups just referred to. In these, as stated above, the duct is

differentiated, so to speak, *in situ* from the mesoblast in its whole length, and as recent authors agree, is composed of a series of the abortive tubules formed in each nephrotome. This is not the case in Selachia; here it is brought about, as RABL states, by the posterior growth of the collecting duct which is formed by the confluence of the lateral extremities of the pronephric Anlagen. It is not easy to bring these two widely divergent modes of formation into harmony with each other.

A few morphological considerations, however, would, I believe, enable one to derive one type of the system from the other. I may be permitted to state here some of these considerations.

I will start with the question: Is the segmental duct of Selachia the morphological equivalent of that of *Petromyzon*, Teleostei, and Amphibia? I believe the question can be answered safely in the negative, if we consider (1) the position of the pronephros first formed, and (2) the origin of the duct.

In the first place, the pronephros in Selachia appears, as we learn from RABL, in the mesoblastic somites lying *posterior* to the Anlage of the liver; thus the Anlage of the liver lies under the fourth and fifth somites, and that of the pancreas under the sixth, while the pronephros covers the seventh to tenth somites ('96, p. 667). On the contrary, in *Petromyzon*[1] the pronephros originates in the mesoblastic somites *anterior* to the hepato-pancreatic Anlage, only the posterior one or two nephrotomes covering the liver. Such being the case, the pronephric segments in Selachia correspond to the same number of the abortive tubules in *Petromyzon* and the other Craniota above

1) As we learn from GOETTE ('75) and Oellacher ('73), the anterior section of the pronephric system in Amphibia and Teleostei, is also found in the mesoblast opposite to the posterior section of the fore-gut.

mentioned, which are converted into the anterior section of the segmental duct in these groups. It follows that the segmental duct in this part of *Petromyzon* and the two other groups, is not the morphological equivalent of the duct of the same name, but of the pronephros itself, in Selachia.

In the second place, let us consider the mode of growth of the segmental duct. Whichever view may be taken of its origin, whether epiblastic or mesoblastic, this duct in Selachia does not arise segmentally as in the other Craniota just referred to. It is a backward growth produced either by delamination from the epiblast, as RÜCKERT and VAN WYHE affirm, or by cell-multiplication within the structure of the mesoblastic collecting duct itself, as RABL states. Hence it can not be homologous with the duct of the same name in *Petromyzon* and the two other groups, which is derived segmentally from the rudimentary pronephric tubules. The Selachian segmental duct is, in its whole length, represented, as I believe, by the posterior small section of the segmental duct in *Petromyzon* and Amphibia.

In *Petromyzon*, the hind end of the duct comes into an intimate connection with both the epiblast and the lateral diverticula of the cloaca, filling up the space between them, and fusing with both of them. We may suppose that the direct communication of the duct with the exterior, if such truly existed in the ancestral history, may have been at this point of the epiblast. This fused condition of the duct and the epiblast reminds us of the early stages of the Selachian duct at the stage when it has been produced only a little posteriorly from the pronephric region. I believe that if the duct is to be compared in *Petromyzon* and Selachia, a stage such as the above ought to be taken. The largest part of the Selachian duct is represented by a free hind-

ward growth formed after such a condition is passed, and its homologue can not be found anywhere in *Petromyzon*. I believe that the same can be stated of Amphibia which is, to judge from FIELD's account, very much like *Petromyzon* in this respect (see p. 401).

According to VAN WYHE and RABL, the duct in Selachia appears in the seventh to the tenth (in RABL's sense) somites of embryos with 34-35 somites and, when it is later connected with the cloacal wall, the connection is found,—as can be inferred from fig. 7*b* of VAN WYHE,—in the thirty-eighth (forty-second of RABL) Rumpfsegment (or further backwards) of *Pristiurus* embryos with 80 (VAN WYHE) to 87 (RABL) mesoblastic somites. There is found in Selachia, therefore, a number of the mesoblastic somites in the region back of the pronephros, which do not give rise either to the pronephric tubules or to the segmental duct, and the duct grows backwards, free from the mesoblast inside, during the period in which the somites increase from 34-35 to 80-87, and for the space reaching from the eleventh or twelfth to the forty-second (in the sense of RABL) somite. Such a considerable prolongation of the duct during this period is not observed in *Petromyzon*[1] and in the two other groups of Craniota mentioned.

And furthermore, it is questionable whether, during this posterior growth, the duct in Selachia receives the constituent cells from the epiblast along its whole length, as RÜCKERT and VAN WYHE believe; or only at the point of the epiblast overlying the hind end of the pronephros, with which the duct is connected, and posteriorly to this point grows free from both the epiblast

1) At about this stage (Stage IV), there is no space left behind the pronephric system segmentally formed; for the embryo of *Petromyzon*, is retort-shaped and has the anus situated in the ventral median line of the bulb of the retort.

and the mesoblast. The latter view seems probable to me. The figures ('89, figs. 5 *a-c*) given by VAN WYHE to illustrate his view of the epiblastic origin of the duct, are from the vertical plane of the eighth Rumpfsegment of a *Scyllium* embryo with 37 somites, which corresponds to the twelfth Gesammtsegment of RABL. The figures ('96, figs. 9A, 9B, 10A, and 10B) given by RABL to the negation of VAN WYHE's view, are from the vertical plane of the twenty-second Gesammtsegment of an embryo with the 63 somites. These two cases are, I suppose, the two ends of the same duct in different stages; the anterior end, being the equivalent of the hind end of the segmental duct in *Petromyzon*, actually receives cells out of the epiblast, as the figures by VAN WYHE show; the other end, which is seen in RABL's figures, is the point of mere contact with the epiblast, along which it is shifting backwards.

If the above comparison be correct, the segmental duct in Selachia is, except the anterior very small section which is formed directly of the abortive tubules, not homologous with the duct of the same name in Petromyzon, but is a structure secondarily acquired.

From the above account, it may be safely concluded that in its primary phylogenetic stage, the pronephric system of the Craniota above referred to consisted of a number of segmentally arranged tubules, which were directly formed, in each mesoblastic segment, from the distal (ventral) portion of the mesoblastic somite, and opened independently to the exterior; that the lateral extremities of these tubules were afterwards secondarily united with one another, thus constructing the collecting and segmental duct, the hind end of which opened directly to the exterior; and that the acquisition of an opening of the duct into the cloaca

was the tertiary stage of changes in the system. Such a course of the phylogenetic development of the system is, however, no other than that advanced by RÜCKERT ('88, p. 265).

In the present paper, the historical comparison will be limited to the groups of Vertebrata stated above; the review of Amniota and some other theoretical considerations will be reserved to a future paper, in which I propose to deal with the further fate of the pronephros and the development of the mesonephros in *Petromyzon*.

Having compared the results arrived at in the present work, with those in different classes of Anamnia, I may be justified in drawing the following conclusions.

In *Petromyzon*, the first indications of the pronephros becomes apparent at a stage earlier than those hitherto regarded as the starting point, that is, at a stage in which the mesoblast in the anterior region has undergone the metameric segmentation but the lateral plate is not yet cut off from the somite.

The tissue giving rise to the pronephros is the parietal layer of a small section of the mesoblast, which forms the distal (ventral) half of the mesoblastic somite. This section of the mesoblast exactly corresponds to the "Nephrotom" of RÜCKERT in Selachia.

The Anlage of the pronephros in all the groups of Vertebrata above referred to is produced by the evagination of the parietal layer of the nephrotome which theoretically ought to contain a part of the cœlomic cavity. In Cyclostomata, such a cavity is

actually present[1]: in other groups, the Anlagen are mere thickenings.

As the pronephric tubules are derived, in each segment, from the distal (ventral) half of each mesoblastic somite, the pronephros is, from the first, of a segmental arrangement, being strictly *myomeric*. In fact, the separation of the sclero-myotome from the lateral plate is effected on account of the differentiation of the Anlage of the pronephros or of the nephrotome.

In *Petromyzon*, the pronephric tubules which constitute the glandular part of the system and the anterior section of the segmental duct, are formed in the region of the fore-gut and some of them are detected in the region where the gill-pouches are afterwards formed ; these latter disappear entirely before the gills come into view.

The segmental Anlagen of the pronephric tubules are secondarily connected by the duct formed out of two adjacent pronephric Anlagen and put in communication with one another.

The degeneration of the pronephric tubules takes place from both the cranial and caudal extremities of the system. In the cranial part, the tubules disappear without leaving any trace ; while in the caudal, they are converted into the anterior section of the segmental duct. The remaining part of the system functions for some time as the excretory organ.

The pronephric Anlagen in the hinder region do not develop beyond a certain point, but are employed solely to give rise to the segmental duct just as in the somites having degenerated tubules.

From what has been said, it is, I venture to think, no rash conclusion to regard the pronephric tubules in Petromyzon as having once extended over the body-segments from the branchial region

1) According to SWAEN and BRACHET, the same fact is seen in Teleostei.

to the cloacal part, and as having been, in the anterior region, replaced by gills and, in the posterior, converted into the segmental duct.

In the two anterior segments which belong to the branchial region, the free ends of the tubules are brought into close contact with the epiblast but this germinal layer has, in this region, no share in the formation of the system. The hind extremity of the segmental duct, however, strikes against the epiblast and has every appearance of receiving some cells out of it. *These facts allow us to infer that all the tubules once had each an independent external opening until they were secondarily united with one another by the intersomitic duct.*

The visceral layer of the nephrotome becomes evaginated medianwards and forms a series of segmental pouches on either side of the subchorda; but this feature is temporary, and the structure is soon smoothed by their becoming confluent with one another. This series of pouches is, I believe, the remnant of the *primitive segmental cœlome,* and gives rise to the gonads and the mesonephros.

If the accounts given above be correct, the primary mesoblast is, during early development, divided into two distinct portions: (*a*) *the larger proximal portion which is segmented, and* (*b*) *a small distal portion which is unsegmented. The former is differentiated into the sclero-myotome and the nephrotome, and the latter forms simply the peritoneal linings.*

The pronephric vessels acquire their definitive form in much later stages; when established, they are intersomitic in position. The posterior part is transformed into a pair of the glomeruli of the pronephros.

Petromyzon has for a long time been looked upon as being

peculiar and standing apart from other Vertebrata in the development of the pronephros. But the results brought out in the present work speak for a complete parallelism between this genus and the representatives of other classes of Vertebrata.

Biological Laboratory,
The College of Peers, Tokyo.

November, 1899.

Postscript.

By the kindness of PROF. WATASÉ, I have been enabled to look through DR. WHEELER'S paper on "The Development of the Urogenital Organs of the Lamprey"[1] which has just been published. I find a general agreement of his results with mine. The most important point is the discovery of the earliest traces of the pronephros as given in the foregoing pages. As to the formation of the segmental duct, his views are somewhat different from mine; this and some other points of divergence are, as I believe, due to gaps in his materials. Thus, his fig. 1, which represents section through an embryo in his Stage 1, corresponds to my fig. 1, while the next older stages (Stages 2 and 3) in his series, spoken of as representing GOETTE'S fig. 9 where the heart is already formed, coincide with the oldest embryo of my Stage IV. As seen in the foregoing description, most of the important processes in the

1) Zool. Jahrbücher, Abtheil. für Anat. and Ontog., Bd. XIII, '99.

development of the pronephros and the segmental duct take place in this interval of time, which WHEELER has unfortunately omitted to study. But in the main, his results confirm mine. This agreement arrived at independently naturally affords a good evidence of the correctness of the facts given.

Literature.

('79) BALFOUR :—A Monograph on the Development of Elasmobranch Fishes.

('85) ——— :—A Treatise on Comparative Embryology.

('87) BEARD:—On the Origin of the Segmental Duct in Elasmobranchs: Anat. Anz., Bd. II.

('92) BOVERI :—Die Nierencanälchen des Amphioxus: Zool. Jahrb., Bd. V.

('88) BROOK :—Note on the Epiblastic Origin of the Segmental Duct in Teleostean Fishes and in Birds: Proceedings of the Royal Society of Edinburgh, Vol. XIV.

('98) DEAN :—On the Development of the Californian Hagfish (Bdellostoma stouti Lockington): Quart. Journ. Micr. Sc., N. S., Vol. XL.

('99) ——— :—On the Embryology of Bdellostoma stonti: Festschr. z. siebenzigsten Geburtst. v. C. v. Kupffer.

('82-'90) DOHRN :—Studien zur Urgeschichte der Wirbelthirkërpers: Mittheil. a. d. Zool. Stat. z. Neapel.

('91) FIELD :—The Development of the Pronephros and Segmental Duct in Amphibia:—Bullt. Mus. Comp. Zool. Harv. Coll., Vol. XXI.

('86) FLEMMING :—Die ektoblastische Anlage des Urogenitalsystems bei Kaninchen: Arch. f. Anat. u. Physiol., Anat. Abtheil.

('97) FELIX :—Die PRICE'sche Arbeit „Development of the Excretory Organs of a Mixynoid (Bdellostoma stouti)" und ihre Bedeutung für die Lehre von der Entwickelung des Harnsystems: Anat. Anz., Vol. XIII.

('78) FÜRBRINGER :—Zur Vergleichenden Anatomie und Entwicklungsgeschichte der Excretonisorgane der Vertebraten: Morph. Jahrb., Bd. IV.

('78) GEGENBAUR :—Grundriss der Vergleichenden Anatomie, 2. Auflage.

('75) GOETTE :—Die Entwicklungsgeschichte der Unke, Leipzig.

('88) ——— :—Ueber die Entwickelung von P. planeri: Zool. Anz., Jahrg. VI.

('90) ——— :—Entwicklungsgeschichte des Flussneunauges (P. fluviatilis), Hanburg und Leipzig.

('97) GREGORY :—Origin of the Pronephric Duct in Selachians: Zool. Bullet. Vol. I.

('81) HATSCHEK :—Studien über Entwickelung des Amphioxus: Arb. a. d. Zool. Inst. d. Univ. Wien., Tom. IV.

('98) HAMMAR:—Zur Kenntniss der Leberentwickelung bei Amphioxus: Anat. Anz., Bd. XV.

('97) ———:—Ueber einige Hauptzüge der ersten embryonalen Leberentwickelung: Id., Bd. XIV.

('88) HATSCHEK:—Ueber den Schichtenbau von Amphioxus: Id., Bd. III.

('91) HATTA:—On the Formation of the Germinal Layers in Petromyzon: Jour. Coll. Sc. Imp. Univ., Jap. Vol. V.

('97) ———:—Contributions to the Morphology of Cyclostomata. I. On the Formation of the Heart in Petromyzon: Id., Vol. X.

('97) ———:—Preliminary Note on the Development of the Pronephros in Petromyzon: Annot. Zool. Jap., Vol. I.

('90) HERTWIG:—Lehrbuch der Entwicklungsgeschichte des Menschen und der Wirbelthiere, 3. Auflage.

('86) HOFFMANN:—Zur Entwicklungsgeschichte der Urogenitalorgane bei den Anamnien: Zeitschrift f. wiss. Zool., Bd. XLIV.

('89) ———:—Zur Entwicklungsgeschichte der Urogenitalorgane bei den Reptilien: Id., Bd. XLVIII.

('88) v. KUFFER:—Ueber die Entwickelung der Neunaugen: SB. d. k. bayer. Akad. Wis. zu München, Bd. I. (Ref. in Jahresb. Fortschr. Anat. Phys., '89).

('90) ———:—Die Entwickelung von P. planeri: Arch. f. mikrosk. Anat., Bd. XXV.

('89) LANKESTER:—Contributions to the Knowledge of Amphioxus lanceolatus Yarrell: Quart. Journ. Micr. Science, N. S., Vol. XXIX.

('90) LANKESTER & WILLEY:—The Development of the Atrial Chamber of Amphioxus: Id., XXXI.

('97) MAASS:—Ueber die Entwicklungsstadien der Vorniere und Urniere bei Myxine: Zool. Jahrb., Bd. X.

('88) MARTIN:—Ueber die Anlage der Urniere beim Kaninchen: Arch. f. Anatomie and Entwicklungsgeschichte.

('87) MAYER, (P.):—Ueber die Entwickelung des Herzens und der grossen Gefässstämme bei Selachiern: Mittheil. Zool. Stat. Neapel, Bd. VII.

('88) MITSUKURI:—The Ectoblastic Origin of the Wolffian Duct in Cheolonia (Prelim. Notice): Zool. Anz. Jahrg. XI.

('90) MOLLIER:—Ueber die Entstehung des Urogenitalsystems bei Amphibien: Arch. Anat. Phys., Anat. Abth., Bd. III.

('75) MULLER (W.):—Das Urogenitalsystem des Amphioxus und Cyclostomen: Jen. Zeitschr. Naturw., Bd. IX.

('72 & '73) OELLACHER :—Beiträge zur Entwicklungsgeschichte der Knochenfische : Zeitsch. f. wis. Zool., Bd. XXII u. Bd. XXIII.

('70) OWSJANNISKOW :—Die Entwicklungsgeschichte der Flussneunaugen : Melanges Biologie, Tom. VII.

('87) v. PERENJI:—Die ectoblastische Anlage des Urogenitalsystems bei Rana esculenta und Lacerta viridis : Zool. Anz., Jahrg. X.

('97) PRICE :—Development of the Excretory Organs of a Myxinoid (Bdellostoma stonti Lockington): Zool. Jahrb. Bd. X.

('88) RABL:—Ueber die Differenzirung des Mesoderms: Anat. Anz., Bd. III.

('87) ———:—Theorie des Mesoderms : Morph. Jahrb., Bd. XV.

('96) ———:—Ueber die Entwickelung des Urogenitalsystems der Selachier (zweite Fortsetzung der " Theorie des Mesoderms "): Id., Bd. XXIV.

('88) RÜCKERT :—Ueber die Entstehung der Excretionsorgane bei Selachiern: Arch. Anat. u. Phys., Anat. Abth.

('89) ——— :—Zur Entwickelung des Excretionssystems der Selachier; eine Erwiederung an Herrn van Wyhe : Zool. Anz., Jahrg. XII.

('82) RYDER :—A Contribution to the Embryography of Osseous Fishes with Special Reference to the Development of the Cod : Report U. S. Fishcommission.

('99) SCHNEIDER, (GUIDO) :—Einiges über Resorption und Excretion bei Amphioxus lanceolatus : Anat. Auz., Bd. XVI.

('56) SCHULTZE, (MAX) :—Die Entwicklungsgeschichte von Petromyzon planeri, Haarlem.

('82) SCOTT :—Beiträge zur Entwicklungsgeschichte der Petromyzonten. Morph. Jahrb., Bd. VII.

('81) SEDGWICK :—On the Early Development of the Anterior Part of the Wolffian Duct and Body in the Chick : Quart. Journ. Micr. Sc., N.S., Vol. XXI.

('96) SEMON :—Das Excretionssystem der Myxinoiden in seinen Bedentung für die morphologishe Auffassung (Ref. Jahresber ü. d. Fortsch. d. Anat. u. Phys., N. F., Bd. II).

('97) ———:—Das Excretionssystem der Myxinoiden : Anat. Anz., Bd. VIII.

('97) ———:—Vorniere und Urniere, Id., Bd. XIII.

('75) SEMPER :—Das Urogenitalsystem der Plagiostomen und seine Bedeutung für das der übrigen Wirbelthiere : Arbeiten a. d. zool.-zootom. Inst. in Würzburg, Bd. II.

('87) SHIPLEY:—On Some Points in the Development of Petromyzon fluviatilis : Quart. Journ. Micr. Sc., N.S., Vol. XXVII.

('95) SOBOTA :—Die Entwickelung der Vorniere der Salmoniden: Anat. Anz., Bd. VI.

('84) SPEE, (GRAF):—Ueber directe Betheiligung des Ektoderms an der Bildung der Urnierenanlage des Meerschweinchens: Arch. f. Anat. und Phys., Anat. Abth.

('99) SWAEN et BRACHET :—Entude sur les premiéres phases du dévelopment des organs dérivés du mésoblaste chez les poissons téléostéeus: Arch. de Biologie, Tom. XVI.

('90) WEISS :—Excretory Tubules in Amphioxus lanceolatus: Quart. Journ. Micr. Sc., N. S., Vol. XXX.

('84) WELDON:—On the Head-kidney of Bdellostoma, with a Suggestion as to the Origin of the Suprarenal Bodies: Quart. Journ. Micr. Sc., N.S., Vol. XXIV.

('83) WIEDERSHEIM :—Lehrbuch der vergleichenden Anatomie der Wirbethiere.

('91) WILLEY :—The Later Larval Development of Amphioxus: Quart. Jour. Micr. Sc., N.S., Vol. XXXI.

('94) ——— :—Amphioxus and the Ancestry of Vertebrates, New York and London.

('86) v. WYHE:—Die Betheiligung des Ectoderms an der Entwickelung des Vernierenganges: Zool. Anz. Bd. IX.

('89) ——— :—Ueber der Mesodermsegmente des Rumpfes und die Entwickelung des Excretionssystems bei Selachiern : Arch. f. mikr. Anat., Bd. XXXIII.

('98) ——— :—Ueber die Betheiligung des Ektoderms an der Bildung des Vornierengang bei Selachiern: Verhandl. d. Anat. Gesetsch. auf. d. zwölften Vorsamml. in Kiel.

('88) ZIEGLER :—Der Ursprung der mesenchymatischen Gewebe bei den Selachiern: Arch f. mikr. Anat., Bd. XXXII.

PLATE XVII.

Plate XVII.

[The magnification is the same for all figures: $C \times 2$, with the single exception of fig. 4 which is $E \times 2$.]

a.pn.1-6, Anlagen of pronephric tubules from the first to sixth.
a.sd., Anlage of segmental duct.
cd., collecting duct.
ch., chorda dorsalis.
cut., cutis-layer of myotome.
d., dorsal row of mesoblast.
ep., epiblast.
hy., hypoblast.
l.m., lateral plate of mesoblast.
m., median row of mesoblast.
m.p., parietal layer of mesoblast.
mes., mesoblast.
mt.I, II, &c., the first, second, &c. myotome.
mus., muscle-layer of myotome.
m.v., visceral layer of mesoblast.
n., neural cord or canal.
v., ventral row of mesoblast.

Fig. 1. A transverse section through the dorsal region of an embryo intermediate between Stages I and II.

Fig. 2-7. From a series of transverse sections through a younger embryo of Stage II.

Figs. 8-17. From a series of transverse sections through an older embryo of Stage II.

Figs. 18 and 19. Two sections from a series of cross-sections through a little more advanced embryo than the last.

Figs. 20-29. From serial cross-sections through the most advanced embryo of Stage II.

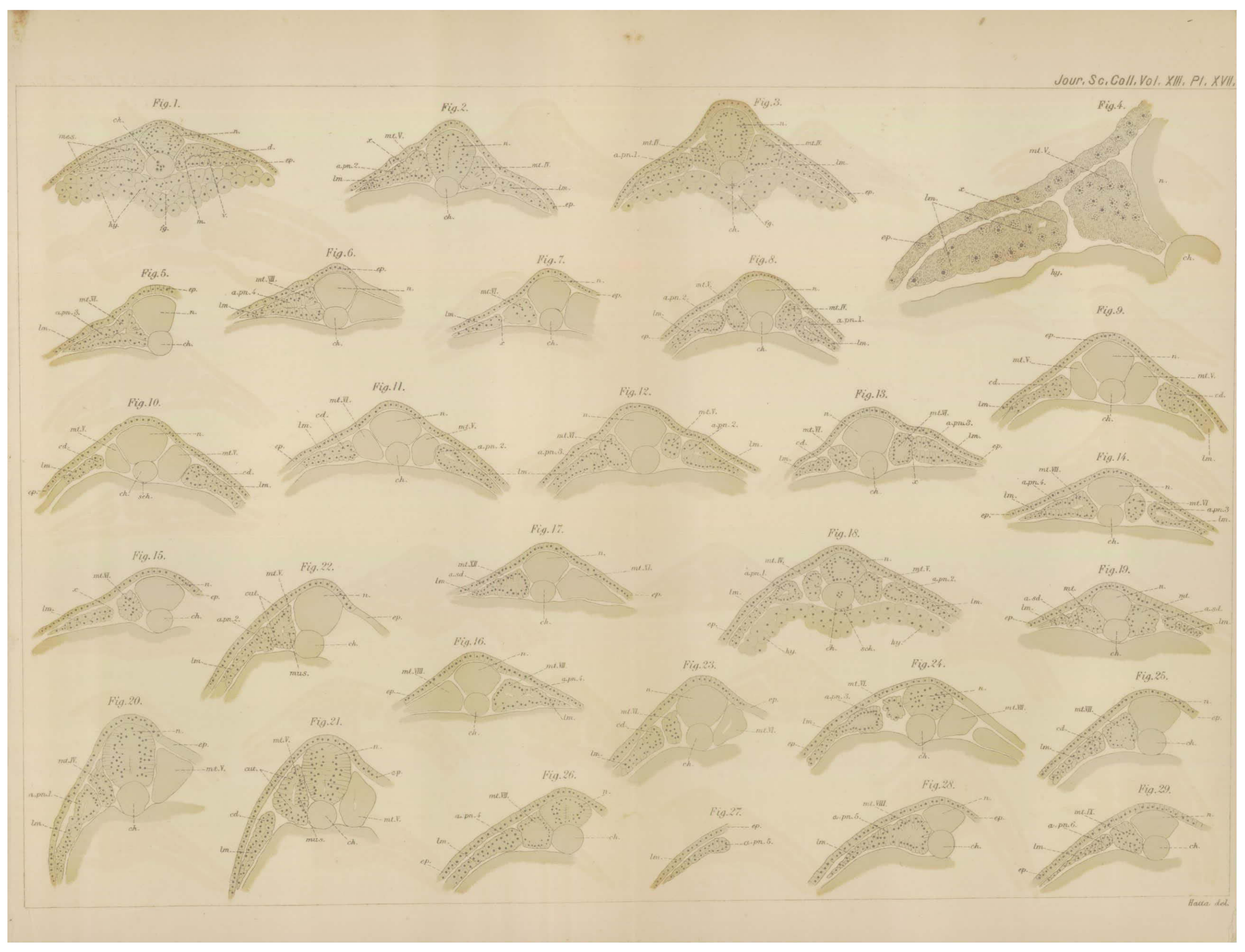


Fig. 1.
ch. mes. n. d. ep. hy. fg. m. v.
Fig. 2.
mt.V. x. n. a.pn.2. mt.IV. lm. lm. ep. ch.
Fig. 3.
n. mt.IV. mt.IV. a.pn.1. lm. ep. ch. fg.
Fig. 4.
mt.V. n. x. lm. ep. ch. hy.
Fig. 5.
ep. mt.VI. n. a.pn.3. lm. ch.
Fig. 6.
ep. mt.VII. n. a.pn.4. lm. ch.
Fig. 7.
n. mt.VI. ep. lm. x. ch.
Fig. 8.
mt.V. n. a.pn.2. mt.IV. a.pn.1. lm. ep. lm. ch.
Fig. 9.
ep. n. mt.V. mt.V. cd. cd. lm. ch. lm.
Fig. 10.
mt.V. n. cd. mt.V. lm. cd. ep. ch. sch. lm.
Fig. 11.
mt.VI. n. cd. mt.V. lm. a.pn.2. ep. ch. lm.
Fig. 12.
n. mt.V. a.pn.2. mt.VI. a.pn.3. lm.
Fig. 13.
n. mt.VI. mt.VI. a.pn.3. cd. lm. lm. ep. ch. x.
Fig. 14.
mt.VII. a.pn.4. n. lm. mt.VI. a.pn.3. ep. lm. ch.
Fig. 15.
mt.VI. n. x. ep. lm. ch.
Fig. 22.
mt.V. n. cut. ep. a.pn.2. ch. lm. mus.
Fig. 17.
n. mt.XII. mt.XI. a.sd. lm. ep. ch.
Fig. 18.
mt.IV. n. a.pn.1. mt.V. a.pn.2. lm. lm. ep. hy. ch. sch. hy.
Fig. 19.
mt. n. a.sd. mt. lm. a.sd. ep. lm. ch.
Fig. 16.
n. mt.VIII. mt.VII. a.pn.4. ep. lm. ch.
Fig. 23.
n. ep. mt.VI. cd. mt.VI. lm. ch.
Fig. 24.
mt.VI. n. a.pn.3. mt.VII. lm. ep. ch.
Fig. 25.
n. mt.VII. ep. cd. ch. lm.
Fig. 20.
n. ep. mt.IV. mt.V. a.pn.1. lm. ch.
Fig. 21.
mt.V. n. cut. ep. cd. mt.V. mus. ch. lm.
Fig. 26.
mt.VII. n. a.pn.4. ch. lm. ep.
Fig. 27.
ep. a.pn.5. lm.
Fig. 28.
mt.VIII. n. a.pn.5. ep. lm. ch.
Fig. 29.
mt.IX. n. a.pn.6. lm. ch. ep.
Hatta del.

PLATE XVIII.

Plate XVIII.

a.pn.1-6, Anlagen of pronephric tubules from the first to sixth.
a.sd., Anlage of segmental duct.
cd., collecting duct.
ch., chorda dorsalis.
c.p., coelomic projection.
cut., cutis-layer of myotome.
fg., fore-gut.
ep., epiblast.
hy., hypoblast.
l.m., lateral plate of mesoblast.
m.p., parietal layer of mesoblast.
ms., mesoblast.
mt.I, II, &c., the first, second, &c. myotome.
mus., muscle-layer of myotome.
m.v., visceral layer of mesoblast.
n., neural cord or canal.
pp.c., pleuroperitoneal cavity.
pt.1-6, pronephric tubules from the first to sixth.
sch., subchorda.
sd., segmental duct.

Figs. 30-31. From the same series as figs. 20-29 of the last plate.

Figs. 32-50. From a series of transverse sections through a younger embryo of Stage III.

Figs. 51-58. From a series of transverse sections through an embryo of Stage III.

Fig. 59. A section through an older emboyo of Stage III, the posterior continuation of which is shown in the next following plate (figs. 60-63).

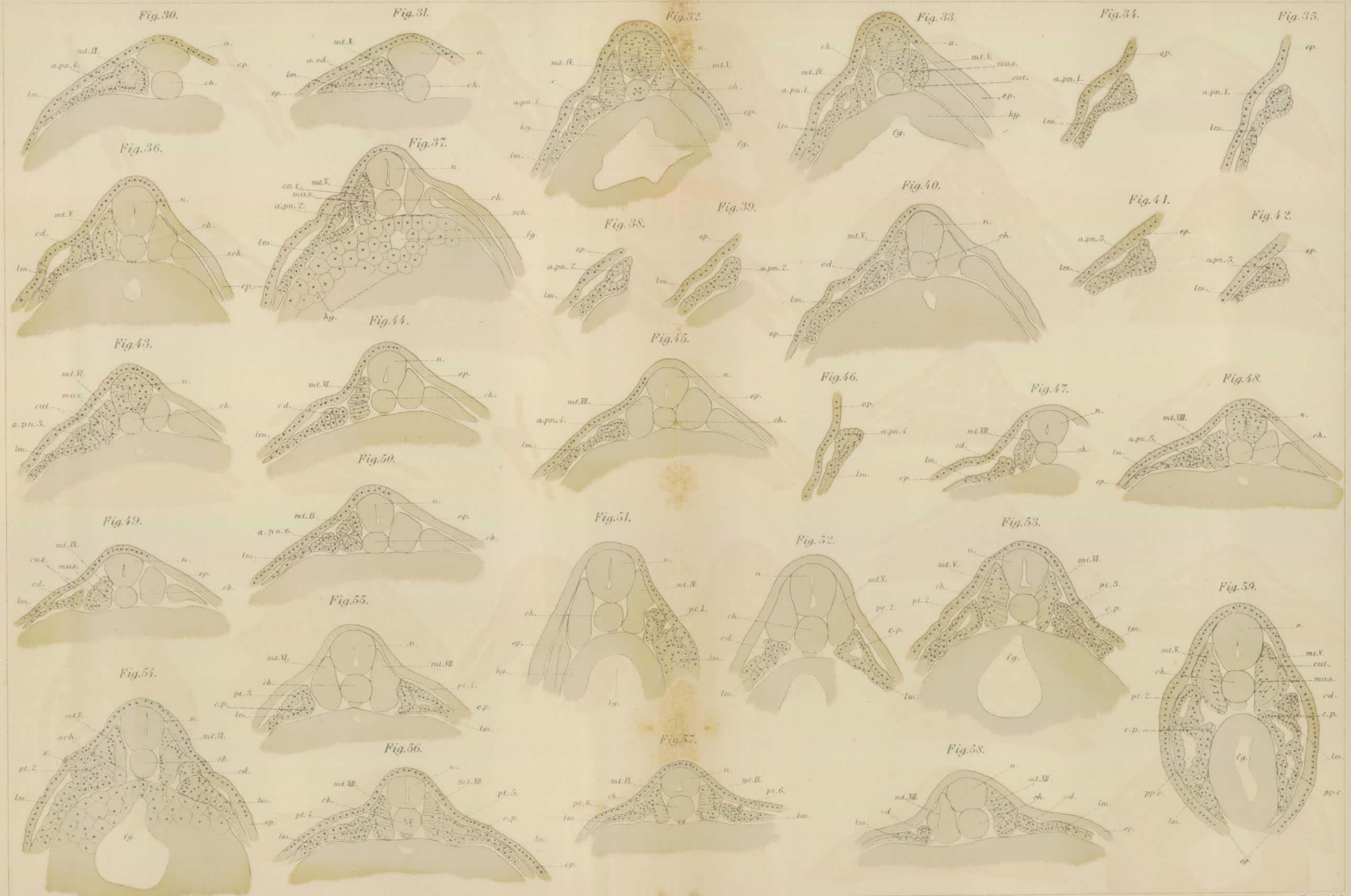


Fig. 30.
mt.IX.
a.pn.6.
n.
ep.
ch.
lm.
Fig. 31.
mt.X.
a.sd.
lm.
ep.
n.
ch.
Fig. 32.
n.
mt.IV.
mt.V.
x.
ch.
a.pn.1.
ep.
hy.
fg.
lm.
Fig. 33.
a.
ch.
mt.V.
mus.
mt.IV.
cut.
a.pn.1.
ep.
hy.
lm.
fg.
Fig. 34.
ep.
a.pn.1.
lm.
Fig. 35.
ep.
a.pn.1.
lm.
Fig. 36.
n.
mt.V.
ch.
cd.
sch.
lm.
ep.
Fig. 37.
n.
cu.t.
mt.V.
mus.
ch.
a.pn.2.
sch.
lm.
fg.
hy.
Fig. 38.
ep.
a.pn.2.
lm.
Fig. 39.
ep.
a.pn.2.
lm.
Fig. 40.
n.
mt.V.
ch.
cd.
lm.
ep.
Fig. 41.
a.pn.3.
ep.
lm.
Fig. 42.
a.pn.3.
ep.
lm.
Fig. 43.
mt.VI.
n.
mus.
cut.
ch.
a.pn.3.
lm.
Fig. 44.
n.
mt.VI.
ep.
cd.
ch.
lm.
Fig. 45.
n.
mt.VII.
ep.
a.pn.4.
ch.
lm.
Fig. 46.
ep.
a.pn.4
lm.
Fig. 47.
n.
mt.VII.
cd.
ch.
lm.
ep.
Fig. 48.
mt.VIII.
n.
a.pn.5.
ch.
lm.
ep.
Fig. 49.
mt.IX.
cut.
mus.
n.
ep.
cd.
ch.
lm.
Fig. 50.
n.
mt.IX.
ep.
a.pn.6.
ch.
lm.
Fig. 51.
n.
mt.IV.
pt.1.
ch.
ep.
hy.
lm.
fg.
Fig. 52.
n.
mt.V.
pt.2.
ch.
cd.
c.p.
lm.
lm.
Fig. 53.
n.
mt.V.
mt.VI.
ch.
pt.3.
pt.2.
c.p.
lm.
fg.
Fig. 54.
mt.V.
n.
sch.
mt.VI.
x.
ch.
pt.2.
cd.
lm.
lm.
ep.
fg.
Fig. 55.
n.
mt.VI.
mt.VII.
ch.
pt.3.
pt.4.
c.p.
c.p.
lm.
lm.
Fig. 56.
n.
mt.VII.
mt.VII.
ch.
pt.5.
pt.4.
c.p.
lm.
lm.
ep.
Fig. 57.
n.
mt.IX.
mt.IX.
ch.
pt.6.
pt.6.
lm.
lm.
Fig. 58.
n.
mt.XII.
mt.XII.
ch.
sd.
cd.
lm.
lm.
ep.
Fig. 59.
n.
mt.V.
mt.V.
cut.
ch.
mus.
pt.2.
cd.
c.p.
c.p.
fg.
lm.
pp.c.
pp.c.
lm.
ep.

Hatta del.

PLATE XIX.

Plate XIX.

cd., collecting duct.
ch., chorda dorsalis.
c.p., coelomic projection.
cut., cutis-layer of myotome.
d., dorsal row of mesoblast.
ep., epiblast.
fg., fore-gut.
l., Anlage of liver.
hy., hypoblast.
l.m., lateral plate of mesoblast.
m., median row of mesoblast.
mch., mesenchymatous cells.
ms., mesoblast.
m.p., parietal layer of mesoblast.
mt.I, II, &c., the first, second, &c. myotome.
mus., muscle-layer of myotome.
m.v., visceral layer of mesoblast.
n., neural canal.
nst.2-3, nephrostome the second and third.
pp.1-3, peritoneal partition.
pp.c., pleuroperitoneal cavity.
pt.1-6, pronephric tubules from the first to sixth.
sch., subchorda.
sd., segmental duct.
sg., spinal ganglion.
v. ventral row of mesoblast.

Figs. 60-63. From the same series as, and the posterior continuation of, fig. 59.

Figs. 64-76. From a series of transverse sections through an oldest embryo of Stage III.

Figs. 77-81. From a series of transverse sections through an embryo of Stage IV; hence the body of embryo in the present stage is twisted, the sections pass through unavoidably oblique planes.

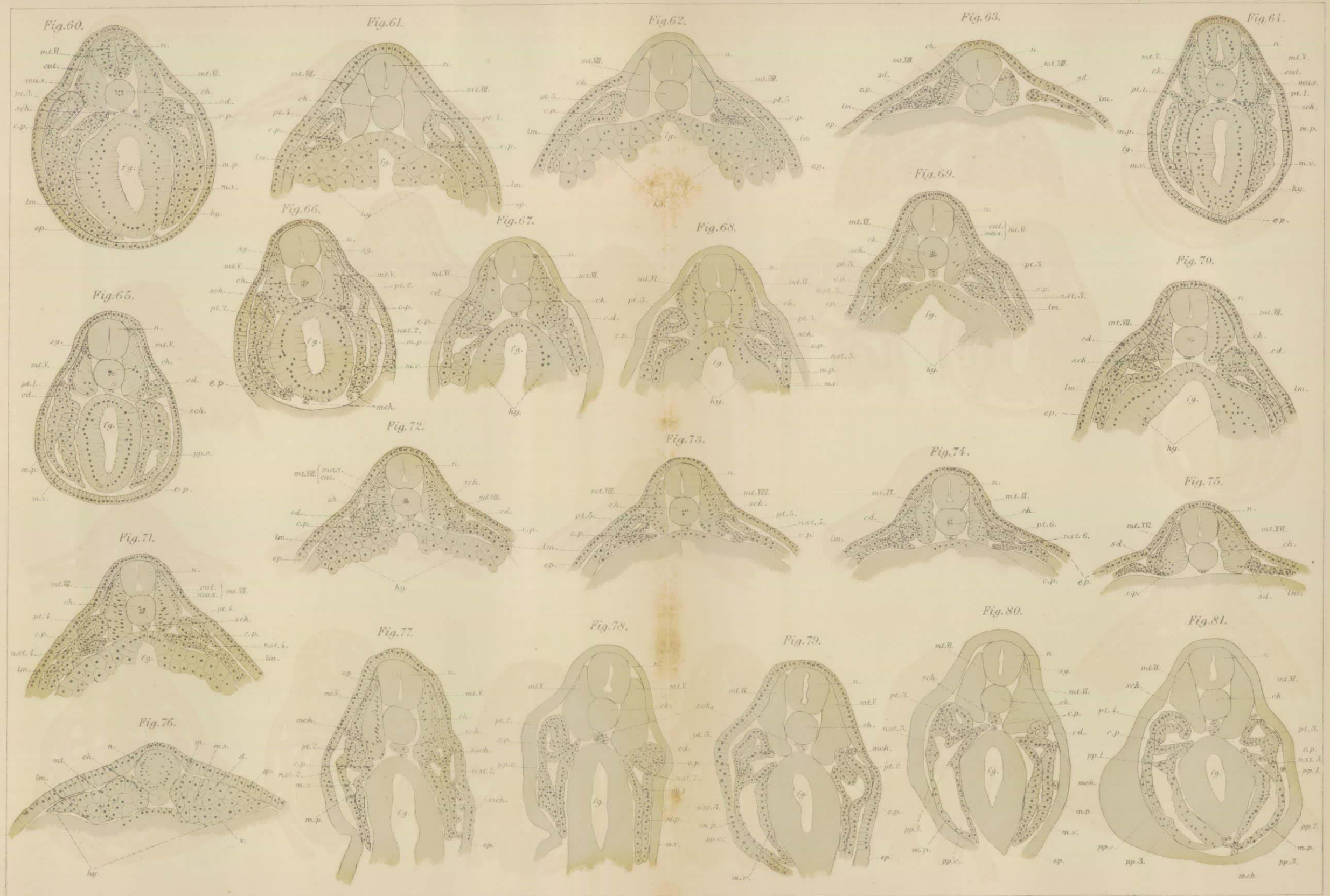


Fig. 60.
Fig. 61.
Fig. 62.
Fig. 63.
Fig. 64.
Fig. 65.
Fig. 66.
Fig. 67.
Fig. 68.
Fig. 69.
Fig. 70.
Fig. 71.
Fig. 72.
Fig. 73.
Fig. 74.
Fig. 75.
Fig. 76.
Fig. 77.
Fig. 78.
Fig. 79.
Fig. 80.
Fig. 81.

Hatta del.

PLATE XX.

Plate XX.

a.sd., Anlage of segmental duct.
bp., blastopore.
brg., branchial region.
cc., cloacal cavity.
cd., collecting duct.
ch., chorda dorsalis.
c.dv., diverticulum of cloacal cavity.
co.sd., cloacal opening of segmental duct.
c.p., cœlomic projection.
dl.bp., dorsal lip of blastopore.
fg., fore-gut.
ep., epiblast.
gc. genital cells.
hy., hypoblast.
int., intestine.
l., Anlage of liver.
l.m., lateral plate of mesoblast.
mch., mesenchymatous cells.
ms., mesoblast.
m.p., parietal layer of mesoblast.
mt.I, II, &c., the first, second, &c. myotome.
m.v., visceral layer of mesoblast.
n., neural canal.
nst.5, fifth nephrostome.
perit., peritoneal membrane.
pp.1-3, peritoneal partition.
pp.c., pleuroperitoneal cavity.
pt.1-6, pronephric tubules from the first to sixth.
sch., subchorda.
sd., segmental duct.
yc., yolk-cells.

Figs. 82-89. Sections from the same series as fig. 81, lying posterior to it. The section shown in fig. 87 passes through somewhat frontally owing to the bending of the body-axis of the embryo; the neural canal, which is bent in the same manner as the axis meets with two time in section.

Figs. 90 and 91. Two sections passing through in the same way as in fig. 87; in fig. 90 the dorsal lip of the blastopore, and in fig. 91, the upper (dorsal) portion of it, is cut through.

Figs. 92-96. From a series of transverse sections through a little older embryo than the last; the embryo is twisted in the same way as it.

Fig. 97. Frontal section through an embryo about the same stage as the last, the body of which has been straightened before cut through.

Fig. 82.

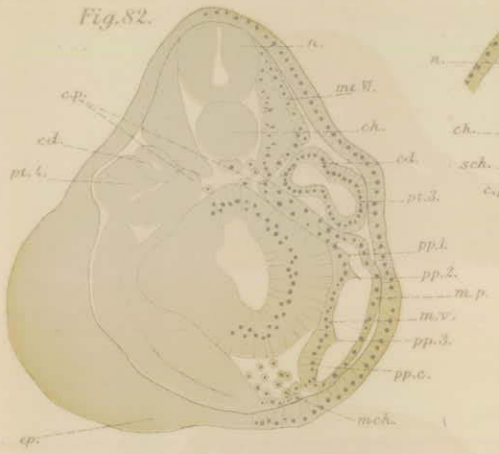
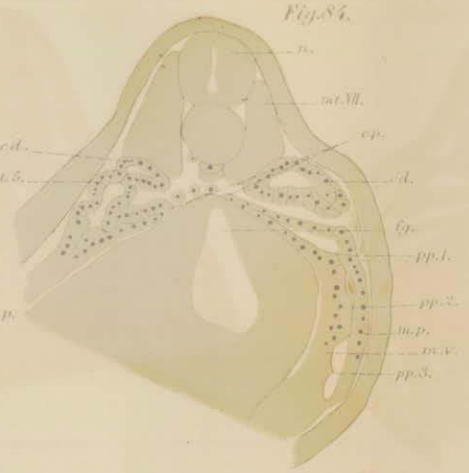


Fig. 83.

Fig. 84.



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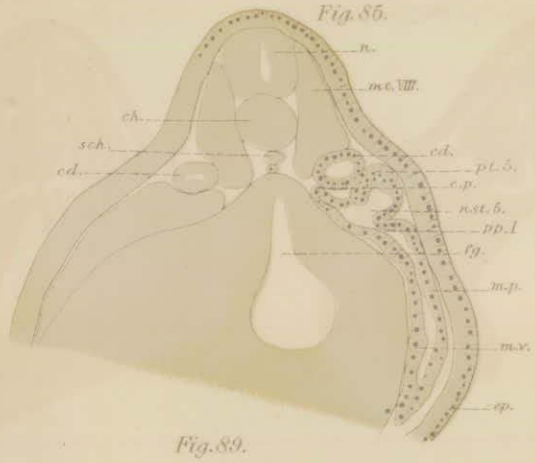

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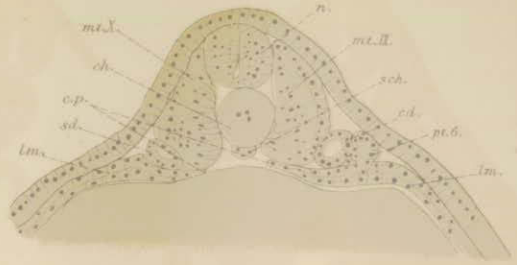

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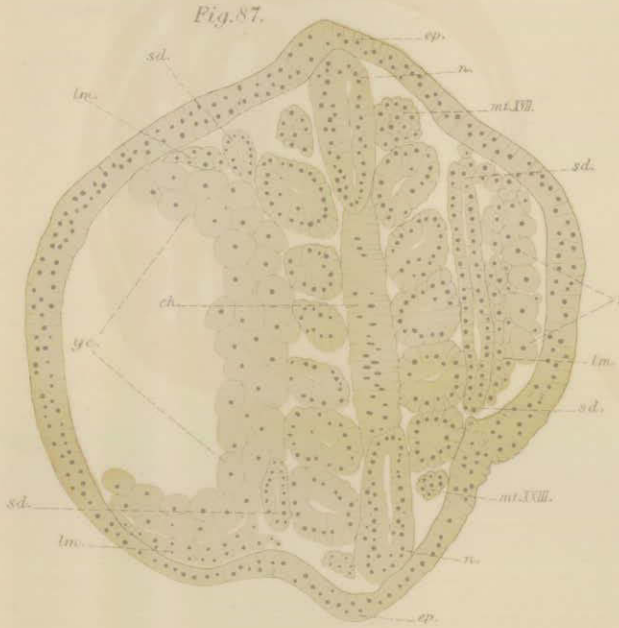

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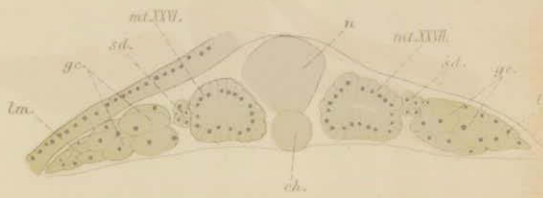

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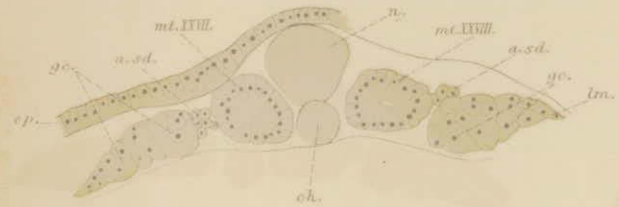

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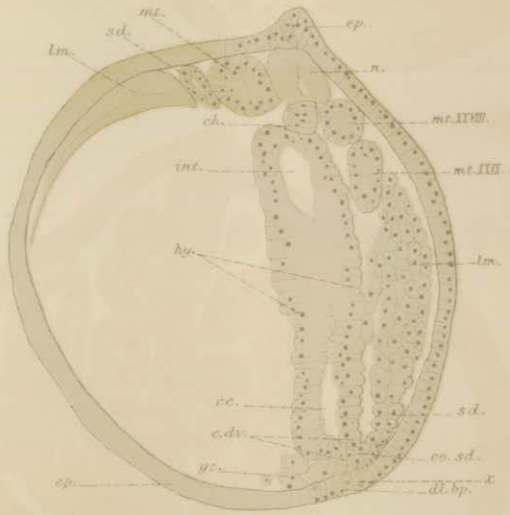

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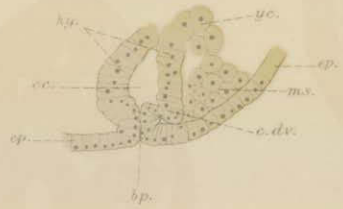

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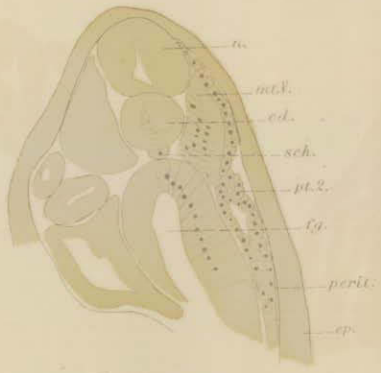

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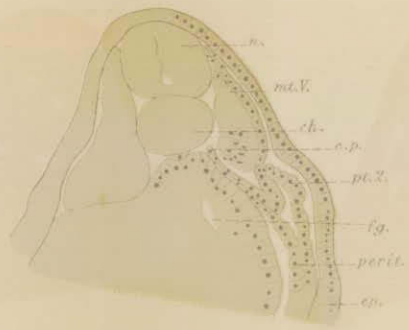

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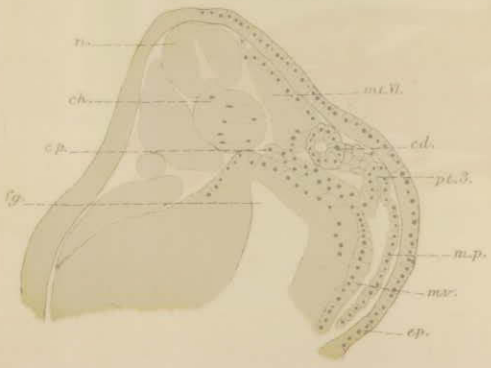

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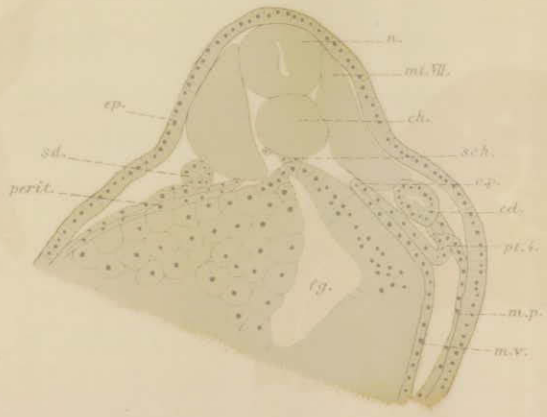

Fig. 96.

Fig. 97.

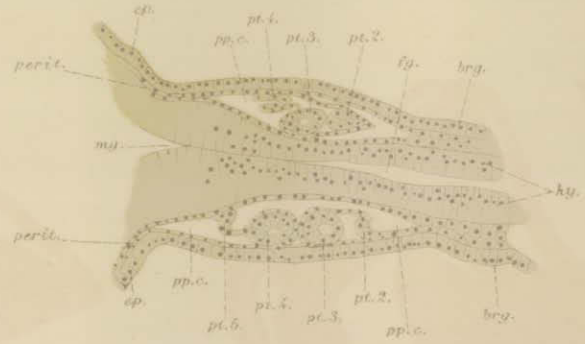

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PLATE XXI.

Plate XXI.

a.cv., anterior cardinal vein.
au., auditory pit.
bp., ventral lip of blastopore.
brg., branchial region.
bs., blood space.
cd., collecting duct.
ch., chorda dorsalis.
cc., cloacal cavity.
c.dv., diverticulum of cloacal cavity.
co.sd., cloacal opening of segmental duct.
cw., wall of cloaca.
ep., epiblast.
gl., glomerulus of pronephros.
f., dorsal fin.
fg., fore-gut.
h., heart.
hy., hypoblast.
l., liver, or Anlage of liver.
l.m., lateral plate of mesoblast.
mch., mesenchymatous cells.
m.p., parietal layer of mesoblast.
perit., peritoneal membrane.
mt.I, II, &c., the first, second myotome, &c.
m.v., visceral layer of mesoblast.
n., neural canal.
nst.2-6, nephrostomes from the second to sixth.
pp.c., pleuroperitoneal cavity.
pt. 2-5, pronephric tubules from the first to fifth.
r.m., radix of mesentery.
sch., subchorda.
sd., segmental duct.
t.a, tract of aorta.
t.ac. tract of anterior cardinal vein.
tr.a. truncus arteriosus.

Figs. 98-106. From a series of transverse sections through an embryo of Stage V.

Figs. 107-110. From a series of transverse sections through an embryo of Stage VI.

Fig. 111. Transverse section through the cloacal region of an older embryo of Stage VI.

Figs. 112-114. A series of sagittal sections through a younger embryo in Stage IV.

Fig. 115. A frontal section through an embryo a little more advanced than that of Stage VI.

Fig. 98.

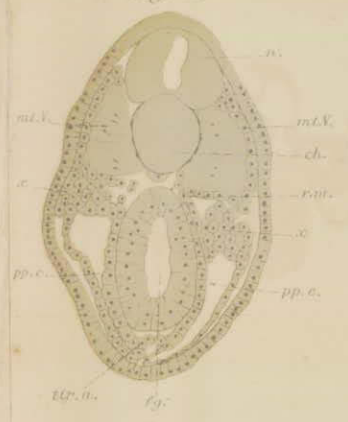


Fig. 99.

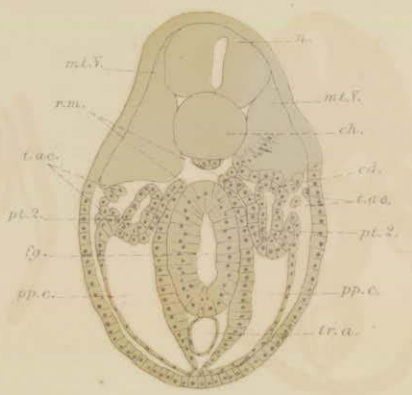


Fig. 100.

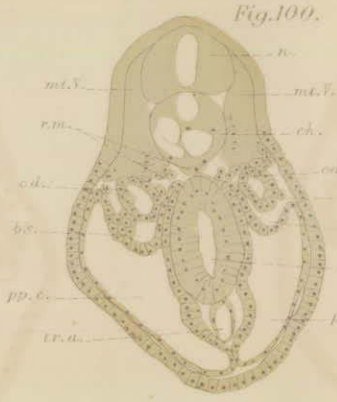
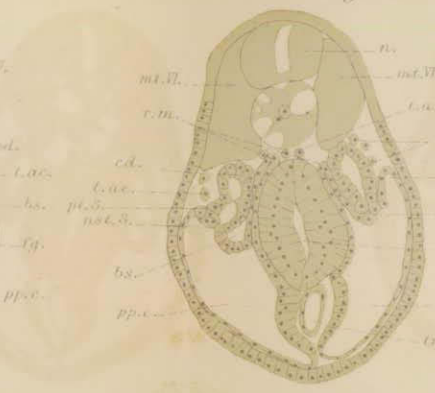


Fig. 101.



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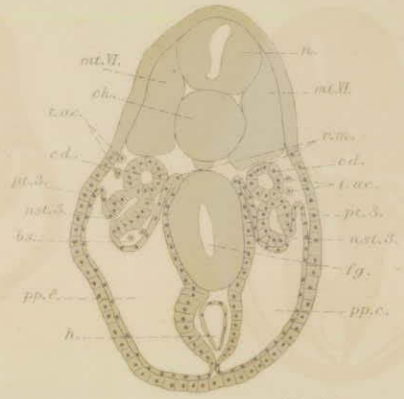

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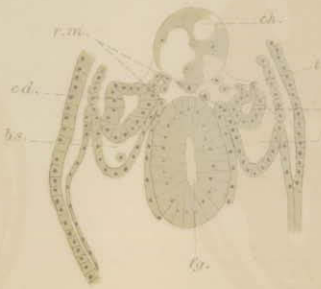

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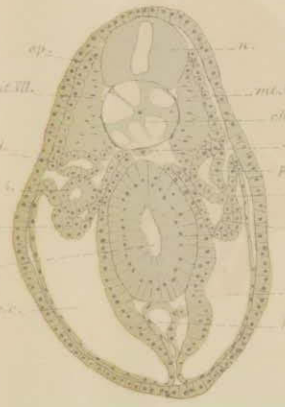

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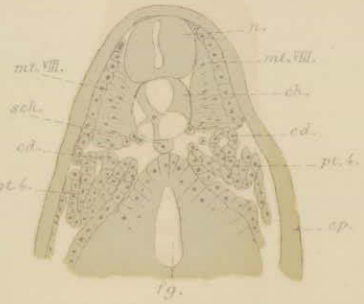
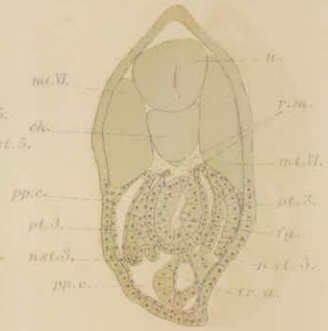


Fig. 106.

Fig. 107.



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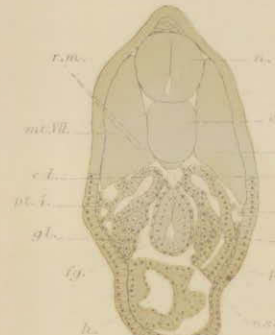


Fig. 109.

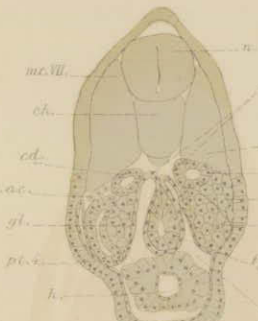


Fig. 110.

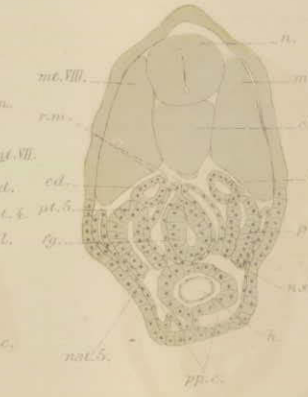


Fig. 111.

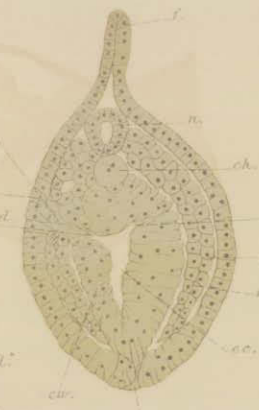


Fig. 112.

Fig. 113.

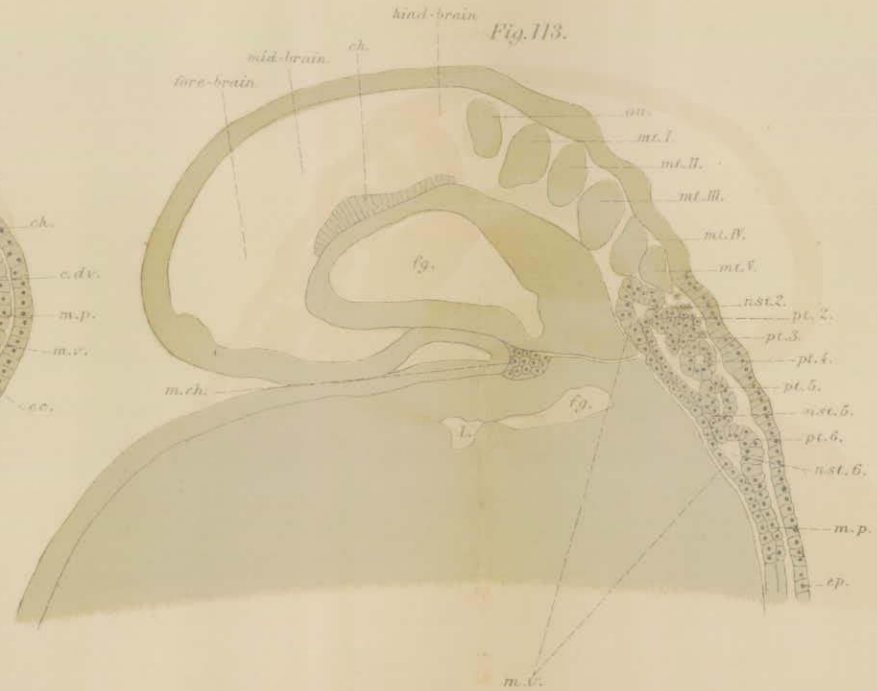
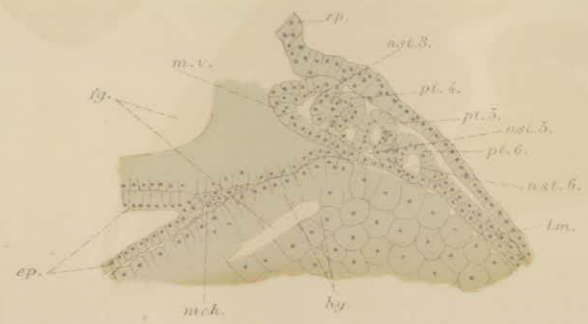


Fig. 115.

Fig. 114.



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