MEMOIRS.

A Comparison *of the* Early Stages *in the* Development *of* Vertebrates. By F. M. Balfour, B.A., Fellow of Trinity College, Cambridge. (With Plate X.)

If the genealogical relationships of animals are to be mainly or largely determined on embryological evidence, it becomes a matter of great importance to know how far evidence of this kind is trustworthy.

The dependence to be placed on it has been generally assumed to be nearly complete. Yet there appears to be no *à priori* reason why natural selection should not act during the embryonic as well as the adult period of life; and there is no question that during their embryonic existence animals are more susceptible to external forces than after they have become full grown: indeed, an immense mass of evidence could be brought to show that these forces do act upon embryos, and produce in them great alterations tending to obscure the genealogical inferences to be gathered from their developmental histories. Even the time-honoured layers form to this no exception. In *Elasmobranchs,* for instance, we find the notochord derived from the hypoblast and the spinal ganglia derived from the involuted epiblast of the neural canal, whilst in the higher vertebrates both of these organs are formed in the mesoblast. Such instances are leading embryologists to recognize the fact that the so-called layers are not quite constant and must not be absolutely depended upon in the determination of homologies. But though it is necessary to recognise the fact that great changes do occur in animals during their embryonic life, it is not necessary to conclude that all embryological evidence is thereby vitiated; but rather it becomes incumbent on us to attempt to determine which embryological features are ancestral and which secondary. For this purpose it is requisite to ascertain what are the general characters of secondary features and how they are produced. Many vertebrates have in the first stages of their development a number of secondary characters which are due to the presence of food material in the ovum; the present essay is mainly an attempt to indicate how those secondary characters arose and to trace their gradual development. At the same time certain important ancestral characters of the early phases of the development of vertebrates, especially with reference to the formation of the hypoblast and mesoblast, are pointed out and their meaning discussed.

There are three orders of vertebrates of which no mention has been made, viz., the *Mammals*, the *Osseous* fishes, and the *Reptiles*. The first of these have been passed over because the accounts of their development are not sufficiently satisfactory, though as far as can be gathered from Bischoff's account of the dog and rabbit there would be no difficulty in showing their relations with other vertebrates.

We also require further investigations on Osseous fishes, but it seems probable that they develop in nearly the same manner as the Elasmobranchs.

With reference to Reptiles we have no satisfactory investigations.

Amphioxus is the vertebrate whose mode of development in its earliest stages is simplest, and the modes of development of other vertebrates are to be looked upon as modifications of this due to the presence of food material in their ova. It is not necessary to conclude from this that Amphioxus was the ancestor of our present vertebrates, but merely that the earliest stages of development of this vertebrate ancestor were similar to those of Amphioxus.

The ovum of Amphioxus contains very little food material and its segmentation is quite uniform. The result of segmentation is a vesicle whose wall is formed of a single layer of cells. These are all of the same character, and the cavity of the vesicle called the segmentation cavity is of considerable size. A section of the embryo, as we may now call the ovum, is represented in Plate X, fig. A I.

The first change which occurs is the pushing in of one half of the wall of the vesicle towards the opposite half. At the same time by the narrowing of its mouth the hollow hemisphere so formed becomes again a vesicle.[1]

[1] I have been able to make at Naples observations which confirm the account of the invagination of Amphioxus as given by Kowalevsky, though my observations are not nearly so complete as those of the Russian naturalist.

Owing to its mode of formation the wall of this secondary vesicle is composed of two layers which are only separated by a narrow space, the remnant of the segmentation cavity.

Two of the stages in the formation of the secondary vesicle by this process of involution are shown in Plate X, fig. A II, and A III. In the second of these the general growth has been very considerable, rendering the whole animal much larger than before. The cavity of this vesicle, A III, is that of the commencing alimentary canal whose final form is due to changes of shape undergone by this primitive cavity. The inner wall of the vesicle becomes converted into the wall of the alimentary canal or hypoblast, and also into part or the whole of the mesoblast.

During the involution the cells which are being involuted undergo a change of form, and before the completion of the process have acquired a completely different character to the cells forming the external wall of the secondary vesicle or epiblast. This change of character in the cells is already well marked in fig. A II. It is of great importance, since we shall find that some of the departures from this simple mode of development, which characterise other vertebrates, are in part due to the distinction between the hypoblast and epiblast cells appearing during segmentation, and not subsequently as in Amphioxus during the involution of the hypoblast.

Kowalevsky ('Entwickelungeschichte des Amphioxus') originally believed that the narrow mouth of the vesicle (according to Mr. Lankester's terminology *blastopore*) became the anus of the adult. He has since, and certainly correctly, given up this view. The opening of the involution becomes closed up and the adult anus is no doubt formed as in all other vertebrates by a pushing in from the exterior, though it probably corresponds in position very closely with the point of closing up of the original involution.

The mode of formation of the mesoblast is not certainly known in Amphioxus; we shall find, however, that for all other vertebrates it arises from the cells which are homologous with the involuted cells of this animal.

Since food material is a term which will be very often employed, it will be well to explain exactly the sense in which it will be used. It will be used only with reference to those passive highly refractive particles which are found embedded in most ova.

In some eggs, of which the hen's egg may be taken as a familiar example, the yolk spherules or food material form the larger portion of the ovum, and a distinction is frequently made between the germinal disc and the yolk.

This distinction is, however, apt to lead to a misconception of the true nature of the egg. There are strong grounds for believing that the so-called yolk, equally with the germinal disc, is composed of an active protoplasmic basis endowed with the power of growth, in which passive yolk spherules are embedded; but that the part ordinarily called the yolk contains such a preponderating amount of yolk spherules that the active basis escapes detection, and does not exhibit the same power of growth as the germinal disc.

With the exception of mammals, whose development requires to be more completely investigated, Amphioxus is as far as we know the only vertebrate whose ovum does not contain a large amount of food material.

In none of these (vertebrate) yolk-containing ova is the food material distributed uniformly. It is always concentrated much more at one pole than at the other, and the pole at which it is most concentrated may be conveniently called the lower pole of the egg.

In eggs in which the distribution of food material is not uniform segmentation does not take place with equal rapidity through all parts of the egg, but its rapidity is, roughly speaking, inversely proportional to the quantity of food material.

When the quantity of food material in a part of the egg becomes very great, segmentation does not occur at all; and even in those cases where the quantity of food yolk is not too great to prevent segmentation the resulting segmentation spheres are much larger than where the yolk-granules are more sparsely scattered.

The frog is the vertebrate whose development comes nearest to that of Amphioxus, as far as the points we are at present considering are concerned. But it will perhaps facilitate the understanding of their relations shortly to explain the diagrammatic sections which I have given of an animal supposed to be intermediate in its development between the Frog and Amphioxus. Plate X, fig. B I, represents a longitudinal section of this hypothetical egg at the close of segmentation. The lower pole, coloured green, represents the part containing more yolk material, and the upper pole, coloured yellow, that with less yolk. Owing to the presence of this yolk the lower pole even at the close of segmentation is composed of cells of a different character to those of the upper pole. In this respect this egg can already be distinguished from that of Amphioxus, in which no such difference between the two poles is apparent at the corresponding period (Plate X, fig. A I).

The segmentation cavity in this ovum is not quite so large proportionately as in Amphioxus, and the encroachment upon it is due to the larger bulk of the lower pole of the egg. In fig. B II, the involution of the lower pole has already commenced; this involution is (1) not quite symmetrical, and (2) on the ventral side (the left side) the epiblast cells forming the upper part of the egg are growing round the cells of the lower pole of the egg or lower layer cells. Both of these peculiarities are founded upon what happens in the Frog and the Selachian, but it is to be noticed that the change from the lower layer cells being involuted towards the epiblast cells, to the epiblast cells growing round the lower layer cells, is a necessary consequence of the increased bulk of the latter.

In this involution not only are the cells of the lower pole pushed on, but also some of those of the upper or yellow portion; so that in this as in all other cases the true distinction between the epiblast and hypoblast does not appear till the involution to form the latter is completed. In the next stage, B III, the involution has become nearly completed and the opening to the exterior or Blastopore quite constricted.

The segmentation cavity has been entirely obliterated, as would have been found to be the case with Amphioxus had the stage a little older than that on Plate X, A III, been represented. The cavity marked (*a l*), as was the case with amphioxus, is that of the alimentary canal.

The similarities between the mode of formation of the hypoblast and alimentary canal in this animal and in Amphioxus are so striking and the differences between the two cases so slight that no further elucidation is required. One or two points need to be spoken of in order to illustrate what occurs in the Frog. When the involution to form the alimentary canal occurs, certain of the lower layer cells (marked *h y*) become distinguished from the remainder of the lower layer cells as a separate layer and form the hypoblast which lines the alimentary canal. It is to be noticed that the cells which form the ventral epithelium of the alimentary canal are not so soon to be distinguished from the other lower layer cells as those which form its dorsal epithelium. This is probably a consequence of the more active growth, indicated by the asymmetry of the involution, on the dorsal side, and is a fact with important bearings in the ova with more food material. The cells marked *m* and coloured red also become distinguished as a separate layer from the remainder of the hypoblast and form the mesoblast. The remainder of the lower layer cells form a mass equivalent to the yolk-sac of many vertebrates, and are not converted directly into the tissues of the animal.

Another point to be noticed is the different relation of epiblast cells to the hypoblast cells at the upper and lower side of the mouth of the involution. Above it, on its dorsal side, the epiblast and hypoblast are continuous with one another. On its ventral side they are primitively not so continuous. This is due to the epiblast, as was before mentioned, growing round the lower layer cells on the ventral side, *vide* B II, and merely remaining continuous with them on the dorsal. The importance of these two points will appear when we come to speak of other vertebrates.

The next animal whose development it is necessary to speak of is the Frog, and its differences from the mode of development are quite easy to follow and interpret. Segmentation is again not uniform, and results in the formation of an upper layer of smaller cells and a lower one of larger; in the centre is a segmentation cavity. The stage at the close of segmentation is represented in C I. From the diagram it is apparent that lower layer cells occupy a larger bulk than they did in the previous animal (Plate X, B I), and tend to encroach still more upon the segmentation cavity, otherwise the differences between the two are unimportant. There are, however, two points to be noted. In the first place, although the cells of the upper pole are distinguished in the diagrams from the lower by their colour, it is not possible at this stage to say what will become epiblast and what hypoblast. In the second place the cells of the upper pole or epiblast consist of two layers—an outer called the epidermic layer and an inner called the nervous. In the previous cases the epiblast consisted of a single layer of cells. The presence of these two layers is due to a distinction which, arising in most other vertebrates late, in the Frog arises early. In most other vertebrates in the later stages of development the epiblast consists of an outer layer of passive and an inner of active cells. In the Frog and other Batrachians these two layers become distinguished at the commencement of development.

In the next stage (C II) we find that the involution to form the alimentary canal has commenced (*a l*), but that it is of a very different character to involution in the previous case. It consists in the growing inwards of a number of cells from the point *x* (C I) towards the segmentation cavity. The cells which grow in this way are partly the yellow cells and partly the smaller green ones. At first this involuted layer of cells is only separated by a slit from the remainder of the lower layer cells; but by the stage represented in C II this has widened into an elongated cavity (*a l*). In its formation this involution pushes backwards the segmentation cavity, which finally disappears in the stage C III. The point *x* remains practically stationary, but by the general growth of the epiblast, mesoblast, and hypoblast, becomes further removed from the segmentation cavity in C II than in C I. On the opposite side of the embryo to that at which the involution occurs the epiblast cells as before, grow round the lower layer cells. The commencement of this is already apparent in C I, and in C II the process is nearly completed, though there is still a small mass of yolk filling up the blastopore. The features of this involution are in the main exaggerations of what was supposed to occur in the previous animal. The asymmetry of the involution is so great that it is completely one-sided and results, in the first instance, in a mere slit; and the whole process of enclosing the yolk by epiblast is effected by the epiblast cells on the side of the egg opposite to the involution.

The true mesoblast and hypoblast are formed precisely as in the previous case. The involuted cells become separated into two layers, one forming the dorsal epithelium of the alimentary canal, and a layer between this and the epiblast forming the mesoblast. There is also a layer of mesoblast accompanying the epiblast which encloses the yolk, which is derived from the smaller green cells at *y* (C I). The edge of this mesoblast, *m′*, forms a thickened ridge, a feature which persists in other vertebrates.

It is a point of some importance for understanding the relation between the mode of formation of the alimentary canal in the frog and other vertebrates to notice that on the ventral surface the cells which are to form the epithelium of the alimentary canal become distinguished as such very much later than do those to form its dorsal epithelium, and are derived not from the involuted cells but from the primitive large yolk-cells. It is indeed probable that only a very small portion of epithelium of the ventral wall of the mid-gut is in the end derived from these larger yolk-cells. The remainder of the yolk-cells (C III, and C II, *yk*) form the yolk mass and do not become directly formed into the tissues of the animal.

In the last stage I have represented for the frog, C III, there are several features to be noticed.

The direct connection at their hind-ends between the cavities of the neural and alimentary canals is the most important of these. This is a result of the previous continuity of the epiblast and hypoblast at the point x, and is a feature almost certainly found in Amphioxus, but which I will speak of more fully in my account of the Selachian's development. The opening of the blastopore called the anus of Rusconi is now quite narrowed, it does not become the anus of the adult. It may be noticed that at the front end of the embryo the primitive dorsal epithelium of the alimentary canal is growing in such a way as to form the epithelium both of the dorsal and ventral surfaces of the foregut.

In spite of various features rendering the development of the frog more difficult of comprehension than that of most other vertebrates, it is easy to see that the step between it and Amphioxus is not a very great one, and will very likely be bridged over at some future time, when our knowledge of the development of other forms becomes greater.

From the frog to the Selachian is a considerable step, but I have again hypothetically sketched a type intermediate between them whose development agrees in some important points with that of *Pelobates fuscus* as described by Bambeke. The points of agreement, though not obvious at first sight, I shall point out in the course of my description.

The first stage (D I), at the close of segmentation, deserves careful attention. The segmentation cavity by the increase of the food yolk is very much diminished in size, and, what is still more important, has as it were sunk down so as to be completely within the *lower layer cells*. The roof of the segmentation cavity is thus formed of epiblast and lower layer cells, a feature which Bambeke finds in *Pelobates fuscus* and which is certainly found in the Selachians. In the Frog we found that the segmentation cavity began to be encroached on by the lower layer cells, and from this it is only a small step to find these cells creeping still further up and forming the roof of the cavity. In the lower layer cells themselves we find an important new feature, viz. that during segmentation they become divided in two distinct parts—one of these where the segments owing to the presence of much food yolk are very large, and the other where the segments are much smaller.

The separation between these two is rather sharp. Even this separation was foreshadowed in the frog's egg, in which a number of lower layer cells were much smaller and more active at the two sides of the segmentation cavity than elsewhere. The segmentation cavity at first lies completely within the region of the small spheres. The larger cells serve almost entirely as food yolk. The epiblast, as is normal with vertebrates, consists of a single layer of columnar cells.

In the next stage (D II) the formation of the alimentary canal (*al*) has commenced, but it is to be observed that there is in this case *no true involution.*

As an accompaniment to the encroachment upon the segmentation cavity, which was a feature of the last stage, the cells to form the walls of the alimentary canal have come to occupy their final position during segmentation and without the intermediation of an involution, and traces only of the involution, are to be found in (1) a split in the lower layer cells which passes along the line separating the small and the large lower layer cells; and (2) in the epiblast becoming continuous with the hypoblast on the dorsal side of the mouth of this split. It is even possible that at this point a few cells (though certainly only a very small number) of those marked yellow in D I become involuted. This point in this, as in all other cases, is the tail end of the embryo. The other features of this stage are as follows:—(1) The segmentation cavity has become smaller and less conspicuous than it was. (2) The epiblast cells have begun to grow round the yolk even in a more conspicuous manner than they did in the frog, and are accompanied by a layer of mesoblast cells which again becomes thickened at its edge. The mesoblast cells in the region of the body are formed in the same way as before, viz. by the separation of a layer to form the epithelium of the alimentary canal, the other cells remaining as mesoblast; and as in the frog, or in a more conspicuous manner, we find that the dorsal surface only of the alimentary cavity has a wall formed of a *distinct layer of cells,* but on the ventral side the cavity is at first closed in by the large spheres of the yolk only. The formation of the alimentary canal by a split and not by an involution is exactly what Bambeke finds in *Pelobates.*

The next stage, D III, is about an equivalent age to C III in the frog. It exhibits the same connection between the neural and the alimentary canals as was found there.

The alimentary canal is beginning to become closed in below, and this occurs near the two ends earlier than in the middle. The cells to form the ventral wall are derived from the large yolk-cells. The non-formation of the ventral wall of the alimentary canal so soon in the middle as at the ends is an early trace of the umbilical canal found in Birds and Selachians, by which the alimentary tract is placed in communication with the yolk-sac. The segmentation cavity has by this stage completely vanished, and the epiblast with its accompanying mesoblast has spread completely round the yolk material so as to form the ventral wall of the body.

Though in some points this manner of development may seem to differ from that of the Frog, there is really a fundamental agreement between the two, and between this mode of development and that of the Selachians we shall find the agreement to be very close.

After segmentation we find that the egg of a selachian consists of two parts—one of these called the germinal disc or blastoderm, and the other the yolk. The former of these corresponds with the epiblast and the part of the lower pole composed of smaller segments in the last-described egg, and the latter to the larger segments of the lower pole. This latter division, owing to the quantity of *yolk* which it contains, has not undergone segmentation, but its homology with the larger segments of the previous eggs is proved (1) by its containing a number of nuclei (E I, *n*), which become the nuclei of true cells and enter the blastoderm, and (2) by the presence in it of a number of lines forming a network similar to that of many cells. The segmentation cavity, as before, lies completely within the lower layer cells.

The next stage, E II, is almost precisely similar to the second stage of the last egg. As there, the primitive involution is merely represented by a split separating the yolk and the germinal disc, and on the dorsal side alone is there a true cellular wall for this split, and at the dorsal mouth of the split the alimentary epithelium becomes continuous with the epiblast.

The segmentation cavity has become diminished, and round the yolk the epiblast, accompanied by a layer of mesoblast, is commencing to grow. In this growth all parts of the blastoderm take a share except that part where the epiblast and hypoblast are continuous. This manner of growth is precisely what occurs in the Frog, though there it is not so easily made out; and not all the investigators who have studied the Frog have understood the exact meaning of the appearances they have seen and drawn. This similarity of relation of the epiblast to the yolk in the two cases is a further confirmation of the identity of the Selachian's yolk with the large yolk-spheres of the previous eggs.

The next stage, E III, is in many ways identical with the corresponding stage in the last-described egg, and in the same way as in that case the neural and alimentary canals are placed in communication with each other.

The mode in which this occurs will be easily gathered from a comparison of E II and E III. It is the same for the

Selachians and Batrachians. The neural canal (*n c*) is by the stage figured E III, completely formed in the way so well known in the Bird, and between the roof of the canal and the external epiblast a layer of mesoblast has already grown in. The floor of the neural canal is the same layer marked *ep* in E II, and therefore remains continuous with the hypoblast at *x*; and when by a simultaneous process the roof of the neural canal and the ventral wall of the alimentary become formed by the folding over of one continuous layer (the epiblast and hypoblast continuous at the point *x*), the two canals, viz. the neural and alimentary, are necessarily placed in communication at their hind ends, as is seen in the diagram.

There are several important points of difference between E III and D III. In the first place, owing to the larger size of the yolk-mass in E III, the epiblast, accompanied by mesoblast, has not proceeded nearly so far round it as in the previous case. It is also worth notice that at the right as well as at the left end of the germinal disc the epiblast is commencing to grow round the yolk. The yolk has, however, become surrounded to a much smaller extent on the right hand than on the left. Since, in the earlier stage, the epiblast became continuous with the hypoblast at *x*, it is not from sections obvious how this occurs. I have therefore appended a diagram to explain it (E′). The blastoderm rests like a disc on the yolk and grows over it on all sides, except at the point where the epiblast and hypoblast are continuous (*x*). This point becomes as it were left in a bay. Next the two sides of the bay coalesce, the bay becomes obliterated, and the effect produced is exactly as if the blastoderm had grown round the yolk at the point *x* (corresponding with the tail of the embryo) as well as everywhere else. It thus comes about that the final point where the various parts of the blastoderm meet and completely enclose the yolk mass does not correspond with the anus of Rusconi of the Frog, but is at some little distance from the hind end of the embryo. In other words, the position of the blastopore in the Selachian is not the same as in the Frog.

Another point deserving attention is the formation of the ventral wall of the alimentary canal. This takes place in two ways—partly by a folding-in at the sides and end, and partly from cells formed around the nuclei (*n*) in the yolk. From these a large portion of the ventral wall of the midgut is formed.

The folding-in of the sheet of hypoblast to assist in the closing-in of the ventral wall of the alimentary canal is a consequence of the flattened form of the original alimentary slit which is far too wide to form the cavity of the final canal. In the bird whose development must next be considered this folding-in is a still more prominent feature in the formation of the alimentary canal. As in the last case, the alimentary canal is widely open in the middle to the yolk at the time when its two ends are closed below and shut off from it; still later this opening becomes very narrow and forms the duct of the so-called umbilical cord which places the yolk-sac in communication with the alimentary canal. As the young animal becomes larger the yolk-sac ceases to communicate directly with the alimentary canal, and is carried about by it for some time as an appendage and only at a later period shrivels up.

The mesoblast is formed in a somewhat different way in the sharks than in other vertebrates. It becomes split off from the hypoblast, not in the form of a single sheet as in other vertebrates, but as two lateral sheets, one on each side of the middle line and separated from one another by a considerable interval; whilst the notochord is derived not as in other vertebrates from the mesoblast, but from the hypoblast (*vide* F. M. Balfour, "Development of Selachians," 'Journal of Microscopical Science,' Oct., 1874).

Between the Selachians and the Aves there is a considerable gulf, which it is more difficult satisfactorily to bridge over than in the previous cases; owing to this I have not attempted to give any intermediate stage between them.

The first stage of the Bird (F I) is very similar in many respects to the corresponding stage in the Selachian. The segmentation cavity is, however, a less well-defined formation, and it may even be doubted whether a true segmentation cavity, homologous with the segmentation cavity in the previously described egg, is present. On the floor of the cavity which is the case formed by the yolk are a few larger cells known as formative cells which, according to Götte's observations, are derived from the yolk, in a somewhat similar manner to the cells which were formed around the nuclei in the Selachian egg, and which helped to form the ventral wall of the alimentary canal. Another point to be noticed is that the segmentation cavity occupies a central position, and not one to the side as in the Selachian.

The yolk is proportionately quite as large as in the Selachian's egg, but, as in that case, there can be little or no doubt of its being homologous with the largest of the segmentation spheres of the previous eggs. It does not undergo segmentation. The epiblast is composed of columnar cells, and extends a short way beyond the edge of the lower layer cells.

In the next stage the more important departures from the previous type of development become visible.

The epiblast spreads uniformly over the yolk-sac and not on the one side only as in the former eggs.

This is due to the embryo (indicated in F II by a thickening of the cells) lying in the centre and not at the edge of the blastoderm. A necessary consequence of this is, that the epiblast does not, as in the previous cases, become continuous with the hypoblast at the tail end of the embryo. This continuity, being of no functional importance, could easily be dispensed with, and the central position of the embryo may perhaps be explained by supposing the process, by which in the Selachian egg the blastopore ceases to correspond in position with the opening of the alimentary slit or anus of Rusconi (*vide* E′), to occur quite early during segmentation instead of at a late period of development. For the possibility of such a change in the date of formation, the early appearance of the nervous and epidermic layers in the Frog affords a parallel.

The epiblast in its growth round the yolk is only partially accompanied by mesoblast, which, however, is thickened at its extreme edge as in the frog. Owing to the epiblast not becoming continuous with the hypoblast at the tail end of the embryo, the alimentary slit is not open to the exterior. The hypoblast is formed by some of the lower layer cells becoming distinguished as a separate layer; the remainder of the lower layer cells become the mesoblast.

The formation of the mesoblast and hypoblast out of the lower layer cells has been accepted for the Bird by most observers, but has been disputed by several, and recently by Kölliker. These have supposed that the mesoblast is derived from the epiblast. I feel convinced that these observers are in the wrong, and that the mesoblast is genuinely derived from the lower layer cells.

The greater portion of the alimentary cavity consists of the original segmentation cavity (*vide* diagrams). This feature of the segmentation cavity of Birds sharply distinguishes it from any segmentation cavity of other eggs, and renders it very doubtful whether the similarly named cavities of the Bird and of other vertebrates are homologous. On the floor of the cavity are still to be seen some of the formative cells, but observers have not hitherto found that they take any share in forming the ventral wall of the alimentary canal.

The features of the next stage are the necessary consequences of those of the last.

The ventral wall of the alimentary canal is entirely formed by a folding-in of the sheet of hypoblast.

The more rapid folding-in at the head still indicates the previous more vigorous growth there, otherwise there is very little difference between the forms of the fold at the head and tail. The alimentary canal does not of course, at this or any period, communicate with the neural tube, since the epiblast and hypoblast are never continuous. The other features, such as the growth of the epiblast round the yolk-sac, are merely continuations of what took place in the last stage.

In the development of a yolk-sac as a distinct appendage, and its absorption within the body, at a later period, the bird fundamentally resembles the dog fish.

Although there are some difficulties in deriving the type of development exhibited by the Bird directly from that of the Selachian, it is not very difficult to do so directly from Amphioxus. Were the alimentary involution to remain symmetrical as in Amphioxus, and the yolk-containing part of the egg to assume the proportions it does in the Bird, we should obtain a mode of development which would not be very dissimilar to that of the Bird. The epiblast would necessarily overgrow the yolk uniformly on all sides and not in the unsymmetrical fashion of the Selachian egg. A confirmation of this view might perhaps be sought for in the complete difference between the types of circulation of the yolk-sac in Birds and Selachians; but this is not so important as might at first sight appear, since it is not from the Selachian egg but from some Batrachian that it would be necessary to derive the Reptiles' and Birds' eggs.

If this view of the bird's egg be correct, we are compelled to suppose that the line of ancestors of birds and reptiles did not include amongst them the Selachians and the Batrachians, or at any rate Selachians and Batrachians which develope on the type we now find.

The careful investigation of the development of some reptiles might very probably throw light upon this important point. In the meantime it is better to assume that the type of development of birds is to be derived from that of the Frog and Selachians.

Summary.—If the views expressed in this paper are correct, all the modes of development found in the higher vertebrates are to be looked upon as modifications of that of Amphioxus. It is, however, rather an interesting question whether it is possible to suppose that the original type was *not* that of Amphioxus, but of some other animal, say, for instance, that of the Frog, and that this varied in two directions,—on the one hand towards Amphioxus, in the reverse direction to the course of variation presupposed in the text; and on the other hand in the direction towards the Selachians as before.

The answer to this question must in my opinion be in the negative. It is quite easy to conceive the food material of the Frog's egg completely vanishing, but although this would entail simplifications of development and possibly even make segmentation uniform, there would, as far as I can see, be no cause why the essential features of difference between the Frog's mode of development and that of Amphioxus should change. The asymmetrical and slit-like form of involution on the one side and the growth of the epiblast over the mesoblast on the other side, both characteristics of the present Frog's egg, would still be features in the development of the simplified egg.

In the Mammal's egg we probably have an example of a Reptile's egg simplified by the disappearance of the food material; and when we know more of Mammalian embryology it will be very interesting to trace out the exact manner in which this simplification has affected the development. It is also probable that the eggs of Osseous fish are fundamentally simplified Selachian eggs; in which case we already know that the diminution of food material has affected but very slightly the fundamental features of development.

One common feature which appears prominently in reviewing the embryology of vertebrates as a whole is the derivation of the mesoblast from the hypoblast; in other words, we find that it is from the layer corresponding to that which becomes involuted in Amphioxus so as as to line the alimentary cavity that the mesoblast is split off.

That neither the hypoblast or mesoblast can in any sense be said to be derived from the epiblast is perfectly clear. When the egg of Amphioxus is in the blastosphere stage we cannot speak of either an epiblast or hypoblast. It is not till the involution or what is equivalent has occurred, converting the single-walled vesicle into a double-walled one, that we can speak of these two layers. It might seem scarcely necessary to insist upon this point, so clear is it without explanation, were it not that certain embryologists have made a confusion about it.

The derivation of the mesoblast from the hypoblast is the more interesting, since it is not confined to the vertebrates, but has a very wide extension amongst the invertebrates. In the cases (whose importance has been recently insisted upon by Professor Huxley), of the Asteroids, the Echinoids, Sagitta, and others, in which the body cavity arises as an outgrowth of the alimentary canal and the somatopleure and splanchnopleure are formed from that outgrowth, it is clear without further remark that the mesoblast is derived from the hypoblast. For the echinoderms in which the water-vascular system and muscular system arise as a solid outgrowth of the wall of the alimentary canal there can also be no question as to the derivation of the mesoblast from the hypoblast.

Amongst other worms, in addition to Sagitta, the investigations of Kowalevsky seem to show that in Lumbricus the mesoblast is derived from the hypoblast.

Amongst Crustaceans, Bobretzky's[1] observations on Oniscus ('Zeitschrift fur wiss. Zoologie,' 1874) lead to the same conclusion.

In insects Kowalevsky's observations lead to the conclusion that mesoblast and hypoblast arise from a common mass of cells; Ulianin's observations bring out the same result for the abnormal Poduridæ, and Metschnikoff's observations show that this also holds for Myriapods.

In mollusks the point is not so clear.

In Tunicates, even if we are not to include them amongst vertebrates,[2] the derivation of mesoblast from hypoblast is without doubt.

Without going further into details it is quite clear that the derivation of the mesoblast from the hypoblast is very general amongst invertebrates.

It will hardly be disputed that primitively the muscular system of the body wall could not have been derived from the layer of cells which lines the alimentary canal. We see indeed in Hydra and the Hydrozoa that in its primitive differentiation, as could have been anticipated beforehand, the muscular system of the body is derived from the epiblast cells. What, then, is the explanation of the widespread derivation of the mesoblast, including the muscular system of the body, from the hypoblast?

The explanation of it may, I think, possibly be found, and at all events the suggestion seems to me sufficiently plausible to be worth making, in the fact that in many cases, and probably this applies to the ancestors of the vertebrates, the body cavity was primitively a part of the alimentary.

[1] He says, p. 182: "Bevor aber die Hälfte der Eioberfläche von den Embryonalzellen bedeckt ist, kommt die erste gemeinsame Anlage des mittleren und unteren Keimblattes zum Vorschein."

[2] Anton Dohrn, 'Der Ursprung des Wirbelthieres.' Leipzig, 1875.

Mr. Lankester, who has already entered into this line of speculation, even suggests ('Q. J. of Micr. Science,' April, 1875) that this applies to all higher animals. It might then be supposed that the muscular system of part of the alimentary canal took the place of the primitive muscular system of the body; so that the whole muscular system of higher animals would be primitively part of the muscular system of the digestive tract.

I put this forward merely as a suggestion, in the truth of which I feel no confidence, but which may perhaps induce embryologists to turn their attention to the point. If we accept it for the moment, the supplanting of the body muscular system by that of the digestive tract may hypothetically be supposed to have occurred in the following way.

When the diverticulum or rather paired diverticula were given off from the alimentary canal they would naturally become attached to the body wall, and any contractions of their intrinsic muscles would tend to cause movements in the body wall. So far there is no difficulty, but there is a physiological difficulty in explaining how it can have happened that this secondary muscular system can have supplanted the original muscular system of the body.

The following suggestions may lessen this difficulty, though perhaps they hardly remove it completely. If we suppose that the animal in which these diverticula appeared had a hard test and was not locomotive, the intrinsic muscular system of the body would naturally completely atrophy. But since the muscular system of the diverticula from the stomach would be required to keep up the movement of the nutritive fluid, it would not atrophy, and were the test subsequently to become soft and the animal locomotive, would naturally form the muscular system of the body. Or even were the animal locomotive in which the diverticula appeared, it is conceivable that the two systems might at first coexist together; that either (1) subsequently owing to the greater convenience of early development, the two systems might acquire a development from the same mass of cells and those the cells of the inner or hypoblast layer, so that the derivation of the body muscles from the hypoblast would only be apparent and not real, or (2) owing to their being better nourished as they would necessarily be, and to their possibly easier adaptability to some new form of movement of the animal, the muscle-cells of the alimentary canal might become developed exclusively whilst the original muscular system atrophied.

I only hold this view provisionally till some better explanation is given of the cases of Sagitta and the Echinoderms, as well as of the nearly universal derivation of the mesoblast from the hypoblast. The cases of this kind may be due to some merely embryonic changes and have no meaning in reference to the adult condition, but I think that we have no right to assume this till some explanation of the embryonic can be suggested.

For vertebrates, I have shown that in Selachians the body cavity at first extends quite to the top of what becomes the muscle plate, so that the line or space separating the two layers of the muscle plate (*vide* Balfour, " Development of Elasmobranch Fishes," ' Quart. Journ. of Micro. Science ' for Oct., 1874. Plate XV, fig. 11 *a*, 11 *b*, 12 *a*, *mp*.) is a portion of the original body cavity. If this is a primitive condition, which is by no means certain, we have a condition which we might expect, in which both the inner and the outer wall of the primitive body cavity assists in forming the muscular system of the body.

It is very possible that the formation of the mesoblast as two masses, one on each side of the middle line as occurs in Selachians, and which as I pointed out in the paper quoted above also takes place in some worms, is a remnant of the primitive formation of the body cavity as paired outgrowth of the alimentary canal. This would also explain the fact that in Selachians the body cavity consists at first of two separate portions, one on each side of the alimentary canal, which only subsequently become united below and converted into a single cavity (*vide* loc. cit., Plate XIV ; fig. 8 *b*, *pp*.)

In the Echinoderms we find instances where the body cavity and water-vascular system arise as an outgrowth from the alimentary canal, which subsequently becomes constricted off from the latter (asteroids and echinoids), together with other instances (ophiura, synapta) where the water-vascular system and body cavity are only secondarily formed in a solid mass of mesoblast originally split off from the walls of the alimentary canal.

These instances show us how easily a change of this kind may take place, and remove the difficulty of understanding why in vertebrates the body cavity never communicates with the alimentary.

The last point which I wish to call attention to is the blastopore or anus of Rusconi.

This is the primitive opening by which the alimentary canal communicates with the exterior, or, in other words, the opening of the alimentary involution. It is a distinctly marked structure in Amphioxus and the Batrachians, and is also found in a less well-marked form in the Selachians; in

Birds no trace of it is any longer to be seen. In all those vertebrates in which it is present, it closes up and does not become the anus of the adult. The final anus nevertheless corresponds very closely in position with the anus of Rusconi. Mr. Lankester has shown ('Quart. Journ. of Micro. Science' for April, 1875) that in invertebrates as well as vertebrates the blastopore almost invariably closes up. It nevertheless corresponds as a rule very nearly in position either with the mouth or with the anus.

If this opening is viewed, as is generally done, as really being the mouth in some cases and the anus in others, it becomes very difficult to believe that the blastopore can in all cases represent the same structure. In a single branch of the animal kingdom it sometimes forms the mouth and sometimes the anus: thus for instance in Lumbricus it is the mouth (according to Kowalevsky), in Palæmon (Bobretzky) the anus. Is it credible that the mouth and anus have become changed, the one for the other?

If, on the other hand, we accept the view that the blastopore never becomes either the one or the other of these openings, it is, I think, possible to account for its corresponding in position with the mouth in some cases or the anus in others.

That it would soon come to correspond either with the mouth or anus (probably with the earliest formed of these in the embryo), wherever it was primitively situated, follows from the great simplification which would be effected by its doing so. This simplification consists in the greater facility with which the fresh opening of either mouth or anus could be made where the epiblast and hypoblast were in continuity than elsewhere. Even a change of correspondence from the position of the mouth to that of the anus or *vice versa* could occur. The mode in which this might happen is exemplified by the case of the Selachians. I pointed out in the course of this paper how the final point of envelopment of the yolk became altered in Selachians so as to cease to correspond with the anus of Rusconi; in other words, how the position of the Blastopore became changed. In such a case, if the yolk material again became diminished, the Blastopore would correspond in position with neither mouth nor anus, and the causes which made it correspond in position with the anus before, would again operate, and make it correspond in position perhaps with the mouth. Thus the blastopore might absolutely cease to correspond in position with the anus and come to correspond in position with the mouth.

It is hardly possible to help believing that the blastopore primitively represented a mouth. It may perhaps have lost this function owing to an increase of food yolk in the ovum preventing its being possible for the blastopore to develop directly into a mouth, and necessitating the formation of a fresh mouth. If such were the case, there would be no reason why the blastopore should ever again serve functionally as a mouth in the descendants of the animal which developed this fresh mouth.

On the Anatomy *of the* Border *of the* Posterior Elastic Lamina *of the* Cornea, *in relation to the* Fibrous Tissue *of the* Ligamentum Iridis Pectinatum. By John Denis Macdonald, M.D., F.R.S., Deputy-Inspector General, R.N.; Assistant-Professor of Naval Hygiene, Army Medical School, Netley. (With Plate XI.)

On dissecting the human eye with the view of demonstrating the generally admitted metamorphosis of the whole border of the *posterior elastic lamina* of the cornea into the fibrous tissue from which the little tendons of the *ligamentum iridis pectinatum* arise, I was surprised to find that no such transformation was anywhere to be detected. The two structures, though, indeed, associated in a very intimate and peculiar manner, were nevertheless, perfectly distinct and not to be confounded with one another. This discovery led me to examine the eye of the sheep more minutely in reference to the point in question, as the pillars of the iris in that animal are very distinctly marked, being much less crowded than they are even in the ox. The result of the investigation may be simply stated as follows:

The peripheral tendon-like processes of the pectinated ligament (Fig. 1 and 2 c) were observed to perforate the border, divide dichotomously (e), intercommunicate (f), and break up, on its anterior surface, into a beautiful fibrous plexus (g) with a disposition of its elements in a concentric rather than in a radial direction.

Moreover, the little tendons, on entering the lamina, were enveloped by conical extensions of structureless substance (d), more strikingly resembling a coating of vitreous enamel than the tubular reflections of a membrane as ordinarily understood. I have taken the following extract from Kölliker's 'Microscopic Anatomy' (p. 541), as affording a good statement of the present acceptation of the structure of the