

remaining entomeres he calls the "Secondare Darmzellen." The intestine is said to be formed from the small cells C'' and D'' (= 5C and 5D). In the history of the entomeres this is really the only point of difference between Umbrella and Crepidula which cannot be satisfactorily explained.

Neither Wilson nor Lillie have observed the division of the entomeres after the formation of the fourth quartette. Mead ('94), however, has observed the formation of the fifth quartette in Amphitrite, for he says, p. 467: "a⁴, b⁴, c⁴ (= 4A, 4B, 4C) and A, B, C, D form the entoderm, the latter cells each dividing once before the invagination."

VIII. AXIAL RELATIONS OF EGG AND EMBRYO.

1. *The Primary Cleavages.*

In recent years much attention has been paid to the relation of cleavage planes to the future axes of the developing animal. Interest was first awakened in this subject by several observations which tended to show that the first cleavage plane always coincides with the future median plane. Such a relation was found by Agassiz and Whitman ('84) in certain pelagic fish eggs, by Roux ('85) and Pflüger ('85) in the frog, by Van Beneden and Julin ('84) in *Clavellina*, by Watasé ('90) in *Loligo*; and the belief was expressed by some authors that the first cleavage plane would be found to coincide with the median plane in all animals with bilateral symmetry. However, further work on this subject has not justified this opinion.

Hatschek ('80) found that in the case of *Teredo* the first cleavage was transverse to the median plane, and later the same relation was found by Wilson ('90) in *Nereis*, Conklin ('92) in *Crepidula*, Heymons ('93) in *Umbrella*.

In other animals it appeared that the first cleavage lay between the median and transverse planes. Whitman ('78) found in *Clepsine* that one of the first four macromeres is anterior, another posterior, and that both the first and second cleavage planes cross the median plane of the embryo obliquely. Rabl ('79) established such a relation in *Planorbis*. Blochmann

('83) found the same thing true of *Aplysia*, and his figures of the segmenting egg of *Neritina* ('81) show that here also the first two furrows are oblique to the median plane of the embryo. The same seems to be true of Bobretzky's ('77) figures of *Fusus*, and Salensky's ('87) figures of *Vermetus*, though in neither case is this matter mentioned in the text.

In all these gasteropods except *Aplysia* four nearly equal macromeres are formed; but in other cases one macromere is often much larger than either of the other three. Among prosobranchs and lamellibranchs this larger macromere is usually in the median line behind, *e.g.*, *Nassa*, *Illyonassa*, *Urosalpinx*, *Tritia*, *Ostrea*, *Unio*, etc., and in all such cases the plane of bilateral symmetry passes through the middle of the larger macromere. Among opisthobranchs and pteropods one or two of the macromeres are frequently smaller than the others, and these lie at the posterior side of the egg, *e.g.*, *Aplysia*, *Umbrella*, *Cavolina*, etc. In such cases the plane of bilateral symmetry may or may not pass through the smallest macromere.

In still other animals it has been found that the first cleavage apparently bears no definite and constant relation to the median plane, *e.g.*, in the toad fish Miss Clapp ('91) has demonstrated that the axis of the embryo may lie in the direction of the first cleavage furrow, or may vary as much as 70° from it; Morgan ('93) asserts that in *Ctenolabrus* and *Serranus* "there is no relation whatsoever between the cleavage planes of the egg and the median plane of the adult body"; Jordan ('93) has found in the newt that the median plane may vary more or less from the plane of the first cleavage, and Jordan and Eycleshymer ('94) have found the same thing true of *Amblystoma*.

Such a series of discordant phenomena might well cause one to regard the axial relations of the first and second cleavages as of no morphological consequence whatsoever. The fact, however, that in many ova there is an absolutely constant relation between the cleavage planes and the axes of the embryo shows that in these cases the position of the cleavage planes and of the resulting blastomeres is not a matter of chance; and this, taken in connection with many other phenomena of similar character,

leads to the view that in those animals with very definite forms of cleavage the position of the furrows and of the cells is causally related to the future axes and organs.

I think it may fairly be doubted whether in any case of spiral cleavage the first furrow is bilateral with regard to the first two blastomeres, even if it is bilateral with regard to the future animal. In *Crepidula* the first cleavage is radial and the two blastomeres are congruent, not bilateral, antimeres, as is shown by the position of the nuclei and protoplasmic fields subsequent to division, Fig. 6. In all cases the second cleavage is radial, as is shown by the position of the nuclei, and above all by the presence of a polar furrow. Hence, neither of the first two furrows is bilateral with reference to the blastomeres, and therefore neither could be strictly bilateral with reference to the adult. In fact, not a single bilateral cleavage occurs until after the formation of the mesoblast (the cell 4d), and consequently the egg could never, either before or after the formation of 4d, be divided into bilaterally symmetrical halves along the planes of one or any number of cleavages. And yet in many eggs, as has been indicated, the position of the future plane of bilateral symmetry can be determined *approximately* even as early as the first or second cleavage, *e.g.*, *Planorbis*, *Teredo*, *Nereis*, etc. *Such determination, however, is only approximate, since in later stages certain of the micromeres shift across the line of symmetry from one side to the other.*

That the axial relations of the first two cleavages can have no such general significance as was once supposed is beyond question. That they may have, however, very constant and definite relations within or between certain groups of animals, in particular annelids, lamellibranchs, and gasteropods, is the thesis here maintained. At first thought there seem to be many observations opposed even to this limited and modified form of the old doctrine. In fact, among the Mollusca alone three different relations of the first cleavage to the median axis of the embryo are known to exist: (1) it may coincide with that axis, *e.g.*, *Loligo*; (2) it may lie at right angles to it, *e.g.*, *Teredo*, *Crepidula*, *Umbrella*; (3) it may cross it at a definite oblique angle, *e.g.*, *Planorbis*, *Neritina*, *Aplysia*. The

first is found only in a highly modified form of cleavage ; the two latter are characteristic, so far as is certainly known, of all gasteropods, lamellibranchs, and annelids, and if the thesis mentioned above is to be maintained, these differences must be explained. This, I believe, can be done, and in the following manner :

In *Crepidula* and all other animals with spiral cleavage in which there is a marked difference in size between the macromeres and micromeres, the first and second cleavage planes represent in the main the furrows between four entoderm cells. *These cells may have different axial relations in different cases, but so far as known the ectoderm and mesoderm cells to which they give rise always have the same axial relations.*

Thus no exception is known either among mollusks or annelids to the rule that the second and fourth quartettes lie in the future median and transverse planes, and that the first, third, and fifth quartettes lie midway between these planes. *The axial differences, therefore, of the first two cleavage planes, which have been mentioned, are differences merely in the axial relations of the four primary entoderm cells, and do not affect the axial relations of the other cells of the ovum, which are always the same among annelids and mollusks.*¹

That this is really true is further shown by the fact that in all cases in which the first and second furrows coincide with the transverse and median planes respectively, the whole of the ectoblast and mesoblast rotates around the egg axis until the second and fourth quartettes lie in the median and transverse planes; whereas in all forms in which one macromere is anterior, one posterior, one right, and one left, there is no such rotation (*cf.* cross in *Neritina* and *Crepidula*). In all known cases of spiral cleavage, excepting a few sinistral gasteropods, the micromeres are separated from the macromeres in the same

¹ Lillie ('95) has expressed somewhat similar views on this subject. He mentions three cases, *viz.*, *Clepsine*, *Planorbis*, and *Neritina*, in which the macromeres are anterior, posterior, right and left respectively; but he discusses only the first of these cases, and concludes that "at present we are unable to explain why, when widely separated forms agree, nearly related species should show reversed relations." The other two cases, *viz.*, *Planorbis* and *Neritina*, show clearly that these reversed relations concern only the four macromeres.

direction and occupy the same relative positions: *e.g.*, the cross is formed over the middle of each macromere, but since in all cases it occupies the same position relative to the future axes, while the macromeres may occupy different positions, it follows that it must rotate in some cases and not in others.¹

Coenogenetically the macromeres have been much modified, witness their differences in size; and the difference in position is probably another such modification. Whether in later stages even these differences disappear is not certainly known, but even if four cells do occupy different positions in different species, it is a small matter compared with the fact that hundreds of cells forming the most important organs of the body occupy the same positions. These facts show that both phylogenetically and ontogenetically the position of the ectomeres and mesomeres is more fundamental than the position of the entomeres. A proper comparison, therefore, between the axes of different animals in these early stages is to be found in the position of the ectoblast and mesoblast rather than in that of the entoblast. Such a comparison would show, I think, that in all annelids and mollusks (excepting, of course, the cephalopods) the median and transverse axes lie in the same groups of ectomeres and mesomeres, and it might possibly unify the varying results thus far obtained in the study of the cleavage in fishes and amphibians.

2. *Establishment of the Larval Axes.*

Hatschek ('88) has expressed in the terms *Protaxonia* and *Heteraxonia* the fact that in one great group of animals, the Coelenterata, the primary axis of the ovum becomes the chief axis of the larva and of the adult, while in all animals with bilateral symmetry these two axes are not the same. In the typical trochophore larva, *e.g.*, that of *Polygordius*, this change of axis is accomplished by the forward movement of the blasto-

¹ In accordance with the description of these rotations given in the earlier part of this paper, I here regard the macromeres as fixed and the micromeres as rotating. In view of the conclusion here reached it would be much better to reverse the process and consider the micromeres fixed in position and the macromeres changeable.

pore (vegetative pole) to one side of the larva, which then becomes the ventral face. This movement is usually accompanied by the closure of the blastopore from behind forward. In some trochophores, however, especially the larvae of the Mollusca, in which there is a large accumulation of yolk at the vegetative pole, this pole may remain fixed in position while the animal pole is shifted forward. The end result is the same in the one case as in the other. In still other cases the movement takes place at both poles; in fact in all cases it is probable that both poles shift position a little.

The result of this shifting is that the egg axis is bent, the angle growing greater and greater throughout development. This bending of the egg axis caused by the shifting of the poles may all be referred ultimately to the greater and more rapid development of the posterior side of the egg, and this greater growth may be located more specifically in the group of cells derived from the first and second somotoblasts (2d and 4d), and is associated with the formation of the trunk. The continued elongation of the trunk in annelids is accompanied by the development of metameric segmentation; in mollusks it leads to a ventral curvature of the trunk associated with the formation of the shell on the postero-dorsal surface, and in gasteropods this curvature of the longitudinal axis is still further complicated by the laetropic torsion of the trunk region posterior to the foot.

In *Crepidula* the conditions are precisely like those described above for ova with a large amount of yolk. The chief axis of the ovum becomes the principal axis of the gastrula; this axis is at first a straight line, but in later stages its upper end is carried forward through an angle of about 90° . In fact, this bending of the embryonic axis is so great and sharp that it is doubtful whether one should not regard that axis as altogether destroyed. The longitudinal or antero-posterior axis runs from the apical cells in front to the anal cells behind, and is perpendicular to the earlier position of the embryonic axis.

The blastopore closes in the middle of the ventral side, Figs. 65, 66, and its posterior lip does not appear to grow more rapidly than the anterior or lateral lips. This might

seem to indicate that the posterior growth of the embryo was not greater than the anterior or lateral growth, but such is not the case, as is strikingly shown by an examination of the aboral side of the embryo, Figs. 64, 74. It is here seen that while the greater growth of ectoblast at the posterior side of the egg has not changed the position of the blastopore, it has shoved the whole of the ectoblast on the aboral surface forward through an angle of about 90° . In this shifting of the ectoblast the entoblast cells take no part, as is clearly shown by the position of the four macromeres and the polar furrow both before and after this movement of the micromeres, Figs. 64, 74. At this stage the polar furrow marks the middle of the dorsal area, the blastopore lies at the middle of the ventral area, and the apical cells which form the centre of the cross lie on the anterior side of the egg about 90° from either of these points. This forward shifting of the apical pole is due to two causes: first, to the enormous enlargement of the cells of the posterior cell plate (Figs. 74, 77), and second, to the rapid multiplication of cells in the region of the shell gland.

In later stages by the enlargement of the shell gland and the multiplication of cells just ventral to it (the growing-point) the ectoblast of the ventral surface is shifted forward, so that the mouth is carried anteriorly, Fig. 76, until it comes to lie near the apex, as shown in Fig. 78. At the same time the embryo increases greatly in length, chiefly by growth at the posterior end, and marks of the final asymmetry appear. The posterior end of the embryo is marked during all this time by the anal cells and by the distal end of the intestine, Figs. 76-78; the antero-posterior axis is at first a straight line connecting the apical and the anal cells, Fig. 77.

This change of axis is undoubtedly the same phenomenon which has been described by Fol ('75 and '76) in the case of pteropods and heteropods, by Blochmann ('81) in *Neritina*, by Heymons ('93) in *Umbrella*, and by Lillie ('95) in *Unio*. The last-named observer has given such an excellent summary of all these cases except that of *Neritina* that I need not discuss them here.

In one point Blochmann's account of this shifting of the axes in *Neritina* differs from my observations on *Crepidula*.

Blochmann finds the cause of this forward movement of the ectoblast in the formation of an ectodermic invagination at the apex. Apart from the doubtful character of this invagination, to which I have already referred, there is such a mass of evidence in favor of the view that the forward movement is due to the greater growth at the posterior pole that I think the cause assigned by Blochmann may fairly be called in question.

In some gasteropods, in which there is a small quantity of yolk, the change of axis occurs in the method typical for the trochophore, *viz.*, the apical pole does not change position, but the oral pole is shoved forward on the ventral side, probably by the development of a structure homologous with the ventral plate of the annelids. Such a process has been described by Patten ('86) in the case of *Patella*.

These shiftings of the embryonic axes are apparently characteristic of all Mollusca, and they present no essential difference from the methods which Alex. Goette ('82) has shown are characteristic also of turbellarians, nemertians, and annelids.

The question of the origin of bilateral from radial forms, and the consequent establishment of the antero-posterior axis among all bilateria is one which has received much attention from some of the most eminent men of science.

Lankester ('77) long ago suggested that the antero-posterior axis of bilateral animals corresponds to the chief axis of coelenterates, the oral pole of the radiate forming the anterior pole of the bilateral animal, and the aboral part of the coelenterate lengthening to form the trunk of the bilateral animal.

Balfour, on the other hand, held that "the conversion of such a radiate form into the bilateral took place not by the elongation of the aboral surface and the formation of an anus there, but by the unequal elongation of the oral face, . . . while the aboral surface became the dorsal surface." This conclusion was elaborated by Balfour, and after him by Sedgwick. Both supposed the central nervous system of arthropods, annelids, mollusks, and chordates to be derived from a circumoral nerve ring.

After proposing this theory, Balfour says (*Comp. Embryology*, vol. II, p. 379): "The position of the flagellum in *Pilidium*

and of the supraoesophageal ganglion in *Mitraria* suggest a different view of the origin of the supraoesophageal ganglion. . . . The position of the ganglion in *Mitraria* corresponds closely with that of the auditory organ in *Ctenophora*; and it is not impossible that the two structures may have had a common origin. If this view is correct, we may suppose that the apex of the aboral lobe has become the centre of the preoral field of the Pilidium and trochosphere larval forms."

This view has been most carefully and elaborately presented by Alex. Goette in his classical work *Entwicklungsgeschichte der Würmer*. He divides bilateral animals into two groups: (1) the pleurogastric, in which the chief axis of the egg becomes the chief axis of the larva or adult, *e.g.*, *Sagitta* and the echinoderms, and (2) the hypogastric, in which group one of the "cross axes" of the egg becomes the chief axis of the larva or adult, *e.g.*, worms, mollusks, arthropods. Goette has striven to show that in all bilateria the animal pole (*Scheitelpol*) corresponds to the future cephalic pole (*Hirnfeld*), and so far as his hypogastric forms are concerned his views on this subject have found repeated and abundant confirmation in the more minute and exact studies which have been made during the last few years on the development of annelids and mollusks.

3. *Beginnings of Final Asymmetry.*

The development of the characteristic asymmetry of the gasteropod belongs to a later period than is treated of in this paper. However, the beginnings of that asymmetry are clearly marked during the embryonic period, and may be briefly touched upon here.

The first evidence of asymmetry and the first trace of antero-posterior elongation appear at the same time and are apparently due to the same cause, *viz.*, the formation of the fifth quartette. The posterior members of this quartette, 5C and 5D, are cut off from the posterior side of the macromeres C and D (Figs. 57-60), thereby increasing the length of the median axis, and, because 5C is formed earlier and lies nearer the dorsal side than 5D, a certain torsion of the posterior end of the embryo follows. This torsion increases until 5D lies nearly on the mid-ventral

line, while 5C lies on the dorsal side, Fig. 68. At the same time all structures on the ventral side are carried to the right, *e.g.*, the fundament of the intestine, while structures on the dorsal side are displaced to the left, *e.g.*, the fundament of the shell gland, Fig. 74. The ectoderm and mesoderm seem to follow the entoderm cells in this torsion, as if they were being passively shifted by the movements of the latter.

In later stages, with the evagination of the shell gland, Fig. 78, the posterior end, morphologically, is shoved farther and farther toward the ventral side. By the latter movement the distal end of the intestine is carried forward on the ventral side, by the laeotropic torsion it is moved up on to the right side of the embryo, Figs. 80 and 81, until the alimentary canal crosses itself like a figure 8 open at the top. These two motions combined bring about the complicated form of asymmetry characteristic of the adult.

With the evagination of the shell gland, the yolk cells protrude like an immense hernia through the lips of the gland, being covered, however, by an exceedingly thin layer of large ectoderm cells; this portion of the embryo becomes the visceral mass (*Eingeweidesack*). The point at which the shell gland was first formed marks the summit of the spire of the adult shell, and the lips of the invagination become the mantle edge, as is well known.

It is abundantly evident from this account that the asymmetry of the adult *Crepidula* is not produced by the asymmetrical development of the shell gland, as is usually maintained for gasteropods in general. In fact the initial asymmetry of the shell gland is produced by the torsion of the posterior end of the embryo. In *Crepidula* the first recognizable cause of the torsion lies in the asymmetry of the cells 5C and 5D. That this has any profound phylogenetic significance, however, seems to me rather doubtful. The yolk cells, because of their great size, exercise an undue amount of influence upon the shape of the entire embryo. It seems to me that phylogenetically neither the yolk cells nor the shell gland were the source of the torsion; they merely took part in a general twisting of the entire posterior end of the embryo.

Crampton's ('94) observation that in a sinistral gasteropod, Physa, there is a reversal of the usual directions of spiral cleavage, is particularly interesting in this connection. If the initial asymmetry is caused in Physa, as in Crepidula, by the asymmetry of the cells 5C and 5D, then it is easy to see how this reversal of the cleavage stands in a causal relation to the reversed asymmetry of the adult. Both Crampton and Kofoid ('94) call attention to the fact that in Rabl's ('79) figures of the embryology of Planorbis there is a reversal of the usual direction of cleavage, and also that in Haddon's ('82) figures of Janthina a similar reversal is indicated. In Planorbis the asymmetry of the adult is reversed, though this does not seem to be the case in Janthina (*cf.* Kofoid ('95), p. 69).

If it should prove on further investigation that reversed cleavage always leads to reversed asymmetry of the adult, there would be good reason for believing that Crepidula exhibits the usual and perhaps the ancestral method in the establishment of the asymmetry of the gasteropods.

D. GENERAL CONSIDERATIONS.

1. *The Forms of Cleavage.*

Several different kinds of cleavage are commonly recognized: (1) with reference to its extent, cleavage is total (holoblastic) or partial (meroblastic); (2) regarding the relative size of the cell products it is equal or unequal; (3) in the distribution of yolk it is telolecithal or centrolecithal; (4) with reference to the constancy of form, it is regular or irregular; (5) with reference to symmetry it is radial or bilateral. To these five classes I think a sixth should be added, at least for the present, *viz.*, one with reference to the destiny of the cleavage cells and axes, and for which I propose the names determinate and indeterminate. It may be that future work will show that this distinction is not necessary, but in the present state of knowledge such a distinction exists and it is very useful to have a name for it. Attention is directed in this place only to the two