

THE EMBRYOLOGY OF BDELLODRILUS PHILADELPHICUS

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TWENTY-SIX TEXT FIGURES AND EIGHT PLATES

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INTRODUCTION

The Discodrilidae are, in many ways, extremely favorable for the study of annelid development. The material can be readily obtained at almost any season of the year. The development of any one egg can be followed from the time of fertilization to its complete development. The smallness of the eggs and the chitinous-like cocoon are the most objectionable features to contend with. Notwithstanding these facts, this group of annelids has been almost completely neglected by investigators in the study of cell lineage.

Moore, in his paper on "The anatomy of an American Discodrilid (*Bdellodrilus illuminatus*)," refers incidently to his 'embryological studies' in the course of his investigations, but has published no account of the Discodrilid development.

Salensky, in his paper on "The development of *Branchiodella* an European Discodrilid, parasitic on *Astacus fluviatilis*," gives an account of the cleavage, axial relation, origin of the germ layers and the formation of the adult structures. But the points of chief importance are so inadequately described and imperfectly worked out, that his results have no special significance in the study of Discodrilid development.

The development of *Bdellodrilus philadelphicus*, one of the Discodrilidae, has, so far as I can learn, never been worked out. It is in many respects a very important form, not only from the standpoint of development, but from its adult anatomical structure as well.

The more important points in the following paper may be briefly summarized as follows:

1. The cleavage is unequal and regular, but may be variable in some eggs. A very small cleavage cavity is present. The gastrula is formed by the epibolic process. The blastopore occupies a very small area on the ventral surface. Its point of closure corresponds to the median ventral side of the adult worm.

2. The early cleavage planes are definitely related to the future organs of the adult worm; i.e., every cleavage foreshadows

the position of some definite future formation. The large macromere D after the formation of d^3 divides very unequally, the smaller cell becomes the entomere D and the larger cell becomes the mesomere d^4 (M). The entire mesoblast is derived from the large cell M after its equal cleavage. The primary mesoblasts M, M, or meso-teloblasts, are completely grown over by the derivatives of X and the cleavage cells of the third generation of ectomeres. The descendants of the primary mesoblasts are differentiated into two distinct groups of cells. The first group becomes the dorsal mesoderm of the adult worm. The second the mesodermal germ bands, becomes the ventral and lateral mesoderm. The cells of the first group remain undifferentiated until late development. The latter becomes differentiated into muscle tissue much earlier than the former.

3. When completely formed, the germ bands consist of three distinct strata of cells: (a) An outer stratum, ectoblast from one to two cells thick, which is produced by the three generations of ectomeres. This layer persists as the definitive hypodermis and secretes the cuticle; (b) A middle stratum, which gives rise to the nervous system and the nephridia; (c) An inner stratum, mesoblast which gives rise to all of the mesodermal elements, blood vessels, septa, reproductive organs, etc.

4. The middle stratum is composed of eight distinct longitudinal rows of cells, which at first lie at the surface and form part of the general ectoderm (ectoblast), but afterwards become completely covered over by the ectoderm. There are four rows in each germ band, terminating at the posterior end in a large cell or teloblast. The inner or ventral neural row on each side gives rise to the corresponding half of the nervous system. The three remaining rows of cells (nephridial) on either side, give rise to the nephridia. The outer nephro-teloblast often proliferates but few cells.

5. The brain or cephalic ganglia take their origin from the extreme anterior ends of the neural rows and are distinctly independent of the ectoderm.

6. The cleavage of the entomeres A, B, C, and D is continued to the end without delay. The entire digestive tract,

with the exception of the very short stomodaeum and proctodaeum is derived from the entomeres. The proctodaeum is on the dorsal side of the tenth segment. The stomodaeum is formed at the apical pole. The embryo is completely turned on itself, i.e., the extreme anterior and posterior ends are in immediate contact. The outer or curved surface, becomes the ventral side of the future adult worm.

NATURAL HISTORY

The Discodrilids occupy rather a unique position in the annelid group. They resemble the Hirudinea in their parasitism, in their general shape, in the presence of an anterior and posterior sucker and in the existence of chitinous jaws. The last character is not found in any other oligochaet, but occurs in a large number of leeches. These facts, perhaps not important in themselves, are indications of a very close relationship between the Discodrilids and the Hirudinea, a group which they approach, not merely in such habits as the formation of the cocoon in which the eggs are enclosed, but in many other points of internal and external structures. The fundamental differences between the two groups are not numerous and are not of such importance as has been assigned them by different writers. The Discodrilids are classified as a distinct family of the Oligochaeta.

Bdellodrilus philadelphicus occurs very abundantly on *Cambarus virilis*, especially in the early spring and summer months. A few may persist throughout the entire winter in their natural habitat.

For convenience, the animal may be divided into three distinct regions; the head (pharynx), the body proper, and the posterior sucker. The head is much broader than the anterior body segments. The head is composed of four distinct annuli, which perhaps represent distinct segments. The first or peristomal annulus is divided into very mobile dorsal and ventral lobes or lips, which exhibit slight median emarginations, but are otherwise entire. It has sensory hairs and papillae, which are common in this family. The fourth annulus is very narrow.

The middle two appear as muscular rings. The chitinous jaws are triangular, the dorsal with a single tooth, the ventral jaw with a pair of smaller teeth. No lateral mucous glands which are very common in some of the species are present.

The body proper consists of eight strongly bi-annulate somites or rings. The anterior somites are longer and broader than the posterior. When contracted, the minor annuli of the somites are telescoped within the major annuli. The fifth, sixth, and seventh somites are sexual. The first, second, third, fourth and eighth somites are nephridial. The spermatheca is broad, thin walled, and nearly cylindrical. The penis is carried to the exterior by the eversible bursa, into which its projecting end is received. There is a conspicuous prostate in addition to the large glandular sperm sac. These parasitic forms remain attached to the ventral surface of the host throughout their entire life history. The eggs are deposited on the ventral surface of the host, more abundantly where the water is kept in constant motion by the movement of the appendages. Each egg is enclosed in a distinct separate stalked cocoon. The base of the stalk is firmly attached to the host. The deposition of eggs occurs during the entire year, if the parasites be kept in aquaria at room temperature. In their natural habitat eggs are not deposited during the severe winter months.

BRIEF OUTLINE OF DEVELOPMENT

The cleavage of the ovum takes place with considerable precision and regularity. Especially is one impressed with this striking phenomenon, after following the cleavage of many ova. The only perceptible variations being (a) slight differences in the time at which the individual cells divide; (b) slight variations in the size of the same cells in different ova. The rate of cleavage varies somewhat with temperature. Occasionally all the cleavage cells of an individual egg are nearly equal and it is impossible to orient the embryo before the germ bands begin their formation. This, however, is an exception, rather than a usual occurrence.

As development progresses the variations between individual embryos become less apparent and as far as can be recognized, do not affect the final result.

The history of the cleavage is distinguished by three well marked periods, namely: oblique, transitional, and bilateral. In the first period, which extends to the twenty-four-cell stage, the germ layers are differentiated, and the parent cells, which give rise to the future organs are definitely marked out.

The first cleavage is nearly transverse to the median longitudinal axis of the adult worm. The second cleavage plane occurs at an angle of forty-five degrees to the first. The third cleavage plane is horizontal and separates the four ectomeres above from the four macromeres below.

Three generations of four ectomeres each are successively separated from the macromeres A, B, C and D. The first generation of ectomeres (a^1 , b^1 , c^1 and d^1), are formed in a right handed direction. The second generation of ectomeres (a^2 , b^2 , c^2 and d^2), are formed in a left handed direction. The third generation of ectomeres (a^3 , b^3 , c^3 and d^3), are formed in a right handed direction. From these twelve ectomeres the entire ectoderm is formed.

The ectomere d^2 gives rise to all or nearly all the ectoderm of the trunk, to the nervous system and to the nephridia.

The oblique type of cleavage is maintained in the division of macromeres. At the close of the oblique period the embryo consists of twenty-four cells (text fig. 6 and fig. 36). The relation of the cleavage cells to the germ layers is as follows:

4.....	macromeres.....	entoderm.....
1.....	mesomere.....	mesoderm.....
20.....	ectomeres.	$\left\{ \begin{array}{l} 19..... \text{ectoderm} \\ 1 (d^2) .. \text{ectoderm, nervous system, nephridia} \end{array} \right.$

Bilateral division now occurs in some of the ectomeres, while others may continue to divide obliquely. The transitional period shows both types of cleavage. Oblique cleavage persists in some of the cells until the fiftieth or more cell stage.

In the third period, the cleavage becomes essentially bilateral

in most of the cells and the teloblasts of the right and left halves of the embryo are formed. Bilateral symmetry now becomes definitely established and the animal increases in length very rapidly.

CLEAVAGE

1. DESIGNATION OF CLEAVAGE CELLS

In the designation of the cleavage cells, for the sake of uniformity and convenience, I have for the most part adopted the system followed by Wilson in his "Cell lineage of Nereis," and Lillie in his study on "The embryology of the Unionidae." The first four cells (macromeres) are designated by the capital letters A, B, C and D. The generations of micromeres (ectomeres) by the small letters a, b, c, and d. The first index number indicates the generation to which the ectomere belongs. Thus a^1 , $b^{1.2}$ or $c^{1.1.2}$ or $d^{1.1}$ all belong to the first generation; c^2 , b^2 , $d^{2.3}$ belong to the second generation, etc. A, B, C and D correspond to the vegetal pole; a, b, c and d to the apical pole. When a cell divides the products receive the designation of the parent cell with the addition of a further index number; thus $b^2 \begin{cases} b^{2.1} \\ b^{2.2} \end{cases}$

Exceptions to this rule are made only in the case of special cells, which, for convenience, receive special designations: thus d^2 of the second generation of ectomeres becomes the 'first somatoblast' and is designated by (X), and its small derivatives by x^1 , x^2 , x^3 , etc.; d^4 the 'second somatoblast' is designated by (M).

2. TYPES OF CLEAVAGE

a. The oblique period of cleavage: one to twenty-four cells

First cleavage: The first cleavage occurs about five to ten hours after the deposition of the egg. The time varies somewhat with external conditions. The plane of division passes through the area where the polar bodies are formed (fig. 1) and divides the egg into two very unequal parts, AB and CD (text fig. 1 and fig. 2). The smaller of the two cells AB is anterior, and

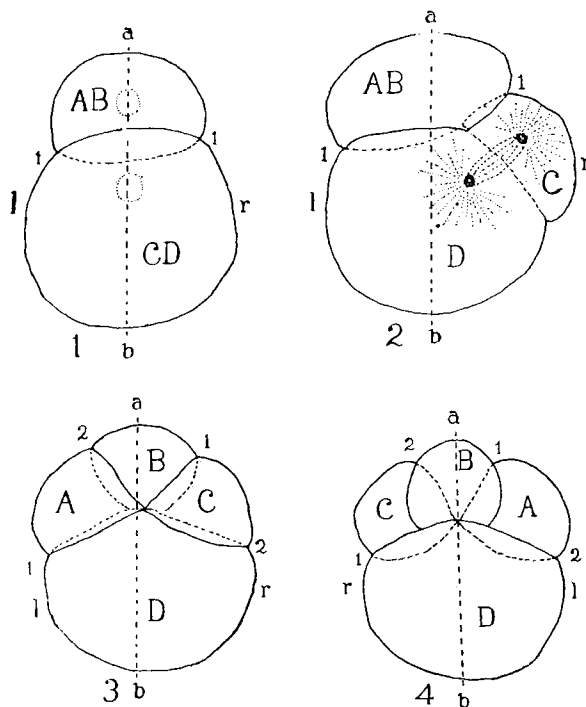


Fig. 1 Two-cell stage from apical pole view.

Fig. 2 Early three-cell stage from apical pole

Fig. 3 Four-cell stage from apical pole view.

Fig. 4 Four-cell stage from vegetal pole.

a-b, median longitudinal plane of future adult; *1-1*, first cleavage plane; *2-2* second cleavage plane; *a*, anterior; *b*, posterior; *r*, right side; *l*, left side.

the larger cell CD is posterior. The cleavage at first is very deep and the cells are rounded, but soon they begin to press against each other and flatten at their point of contact. Before the second cleavage begins the egg assumes its original elliptical shape and the point of contact externally, between the two cells is represented by a mere line or shallow groove. No actual fusion of the two cells ever takes place; sections always show a distinct line of separation between them.

The deutoplasm is equally distributed in both cells. The cytoplasm surrounding the nucleus contains very little yolk

material. This makes it possible, not only to recognize the position of the nucleus, but to be able to make out the exact position of the cleavage spindle in the living egg.

Second cleavage: The second cleavage is meridional and takes place at an angle of forty-five degrees to the median plane of the future adult. The two cells divide at different times (occasionally both cells divide simultaneously). These two cleavages taken together represent the second cleavage in other annelids. CD divides first into two very unequal parts (text fig. 2 and figs. 3, 78). The division of AB is nearly equal (figs. 5 and 6). The largest cell, D, is posterior. B is anterior, inclined a little to the right. C is right (text figs. 3-4) and A is left with reference to the median axis of the future worm. The large cell D has a tendency always to divide first. The exact formation of the four macromeres must be carefully worked out, and correctly understood, since their position largely determines the orientation of the future organs.

For descriptive conveniences the region of the first generation of ectomeres will be considered as the upper or apical pole and the point directly opposite, as the lower or vegetal pole. The centers of the upper and lower poles of the dividing ovum, coincide with the median longitudinal plane of the adult worm. The poles however may be shifted somewhat anteriorly or posteriorly, with reference to the macromeres, more especially to D in the formation of d^2 . When viewed from the upper pole A and C are in contact, while B and D are separated. But when viewed from the ventral pole A and C are separated and B and D are in contact (figs. 6-7). This extensive cross furrow found at the vegetal pole is also present in forms like *Nereis*, *Clepsine* and *Crepidula*; while in those forms like *Unio*, in which the greatest mass of the four macromeres is concerned in the formation of ectoderm instead of endoderm, the cross furrow is greatest at the animal pole. These cross furrows ('Breckungslinie' of Rauber) have no special significance in the development of *Bdellodrilus*, as the cleavage of the macromeres is carried to the end, immediately after the three generations of ectomeres are formed. In those forms like *Nereis*, where the cross furrow

DESCRIPTION OF PLATES

All drawings were made with a camera lucida under a magnification of about 125 diameters. All whole amount drawings, with one or two exceptions, were made from the living egg. The variation in size of the surface views is due to a difference in the size of the eggs. The sections were not uniformly magnified. Stippling has been adopted for the sake of clearness.

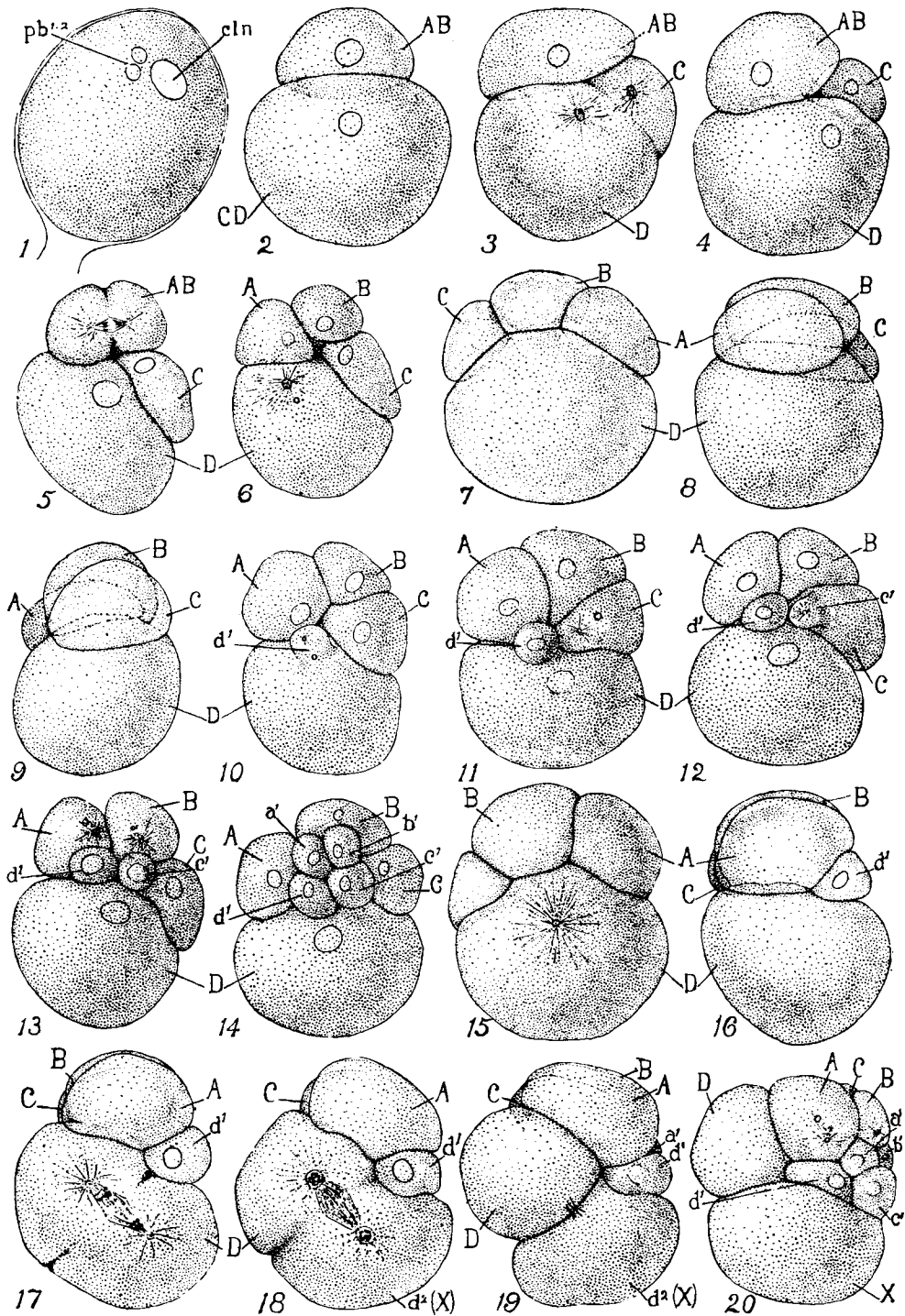
REFERENCE LETTERS

<i>a.</i> , anterior	<i>mi.a.</i> , minor annulus
<i>bl.</i> , blastopore	<i>mj.a.</i> , major annulus
<i>c.</i> , ciliated cells	<i>mo.</i> , mouth
<i>c.c.</i> , cleavage cavity	<i>p.</i> , posterior
<i>c.l.</i> , cerebral lobes	<i>pb¹⁻²</i> , polar bodies
<i>coe.</i> , coelom	<i>ph.</i> , pharynx
<i>ecp.</i> , egg capsule	<i>pr.</i> , proctodaeum
<i>ec.</i> , ectoderm	<i>so.mes.</i> , somatic mesoderm
<i>en.</i> , entoderm	<i>sp.mes.</i> , splanchnic mesoderm
<i>gn.</i> , ganglia	<i>st.</i> , stomodaeum
<i>mes.</i> , mesoderm	<i>v.</i> , ventral
<i>A</i> , left macromere	
<i>B</i> , anterior macromere	
<i>C</i> , right macromere	
<i>D</i> , posterior macromere	
<i>a¹</i> , <i>b¹</i> , <i>c¹</i> , <i>d¹</i> , <i>a¹⁻¹</i> , etc., first generation of ectomeres	
<i>a²</i> , <i>b²</i> , <i>c²</i> , <i>d²</i> , <i>a²⁻¹⁻¹</i> , etc., second generation of ectomeres	
<i>a³</i> , <i>b³</i> , <i>c³</i> , <i>d³</i> , <i>a³⁻¹</i> , etc., third generation of ectomeres	
<i>a⁴</i> , <i>b⁴</i> , <i>c⁴</i> , <i>d⁴</i> , etc., fourth generation of micromeres	
<i>X</i> = <i>d²</i> first somatoblast	
<i>X</i> , <i>X</i> , right and left proteloblasts	
<i>X⁽¹⁾</i> , neuroblast	
<i>X⁽²⁾</i> , <i>X⁽³⁾</i> , <i>X⁽⁴⁾</i> , nephroblasts	
<i>M</i> = <i>d⁴</i> second somatoblast	
<i>m</i> , secondary mesoblast	
<i>x^{1-x⁷}</i> , small derivatives from <i>X</i>	
<i>N</i> , posterior end of nephric rows	
<i>Nc</i> , posterior end of neural rows	
<i>nc</i> , neural rows	
<i>np</i> , nephric rows	

PLATE 1

EXPLANATION OF FIGURES

- 1 Surface view of an unsegmented ovum, to show the polar bodies and the cleavage nucleus.
- 2 Two-cell stage from the upper pole.
- 3 Two-cell stage; cell CD dividing.
- 4 Three-cell stage from the upper pole; division of CD is complete.
- 5 Same stage as preceding; cell AB dividing.
- 6 Four-cell stage from the upper pole; the cleavage spindle for the first ectomere forming.
- 7 Four-cell stage, ventral view.
- 8 Four-cell stage viewed from the left side.
- 9 Same as the preceding, viewed from the right side.
- 10 Four-cell stage from the upper pole, showing the formation of the first ectomere.
- 11 Five-cell stage from the upper pole, d^1 formed.
- 12 Stage showing the cleavage spindle of the second ectomere.
- 13 Six-cell stage from the upper pole; cleavage spindles for third and fourth ectomere forming.
- 14 Eight-cell stage from the upper pole; the macromeres are considerably flattened.
- 15 Same as the preceding, from the ventral pole; similar to the same view of the four-cell stage.
- 16 Eight-cell stage from the left side.
- 17 Same as the last showing the behavior of the macromere D in the formation of the first somatoblast.
- 18 Stage a little later than the preceding.
- 19 Nine-cell stage from the left side after the formation of the first somatoblast.
- 20 Nine-cell stage turned a little to the left, so that all the cells are visible.



Kline, del.

persists until late development, it serves as an unmistakable point of orientation.

Figures 8 and 9 show the four-cell stage from the left and right sides. The dorsal ventral axis of A, B and C is about the same as that of D, but immediately after the formation of the first generation of ectomeres, the cells A, B and C shorten and become more rounded (fig. 16). In later stages of development these cells often become very much flattened and cause the developing embryo to appear unusually large, when viewed from the upper or lower poles.

Third cleavage (eight-cell stage): In the formation of the first generation of ectomeres (d^1 , c^1 , b^1 and a^1), each of the four macromeres divide obliquely. The ectomere end of the cleavage spindle is uppermost. The macromere D divides first; d^1 is budded off from D towards the upper pole, in the direction of the hands of a watch (dexiotropic), (figs. 10–11). We have thus a five-cell stage. Each of the macromeres C, B and A next bud off a small cell towards the upper pole. These are not formed simultaneously, but in the invariable order c^1 , b^1 and a^1 . Thus there occurs successively, a six, a seven and an eight-cell stage (figs. 11–14). In figure 13, an upper pole view, D and C have divided and A and B are preparing to divide. In both A and B the asters of the ectomere end of the spindles are visible. The position of the opposite end of the spindles are indicated by circles. This figure shows the oblique nature of the cleavage spindles. The spindle in A points to the space between A and B. The spindle in B points to the space between B and C. In figure 14, an eight-cell stage, the exact relation of the ectomeres and macromeres are shown as they normally appear from the apical pole. The position of the first generation of ectomeres is obvious. They suggest a possible rotation, after their formation, through an angle of about forty-five degrees in the direction of the hands of a watch. If actual rotation did occur there would be no difficulty in explaining their final position. But the fact that the cleavage spindles are oblique and the position of the completely divided nucleus can be definitely determined, before there is any indication of the cytoplasmic division of the

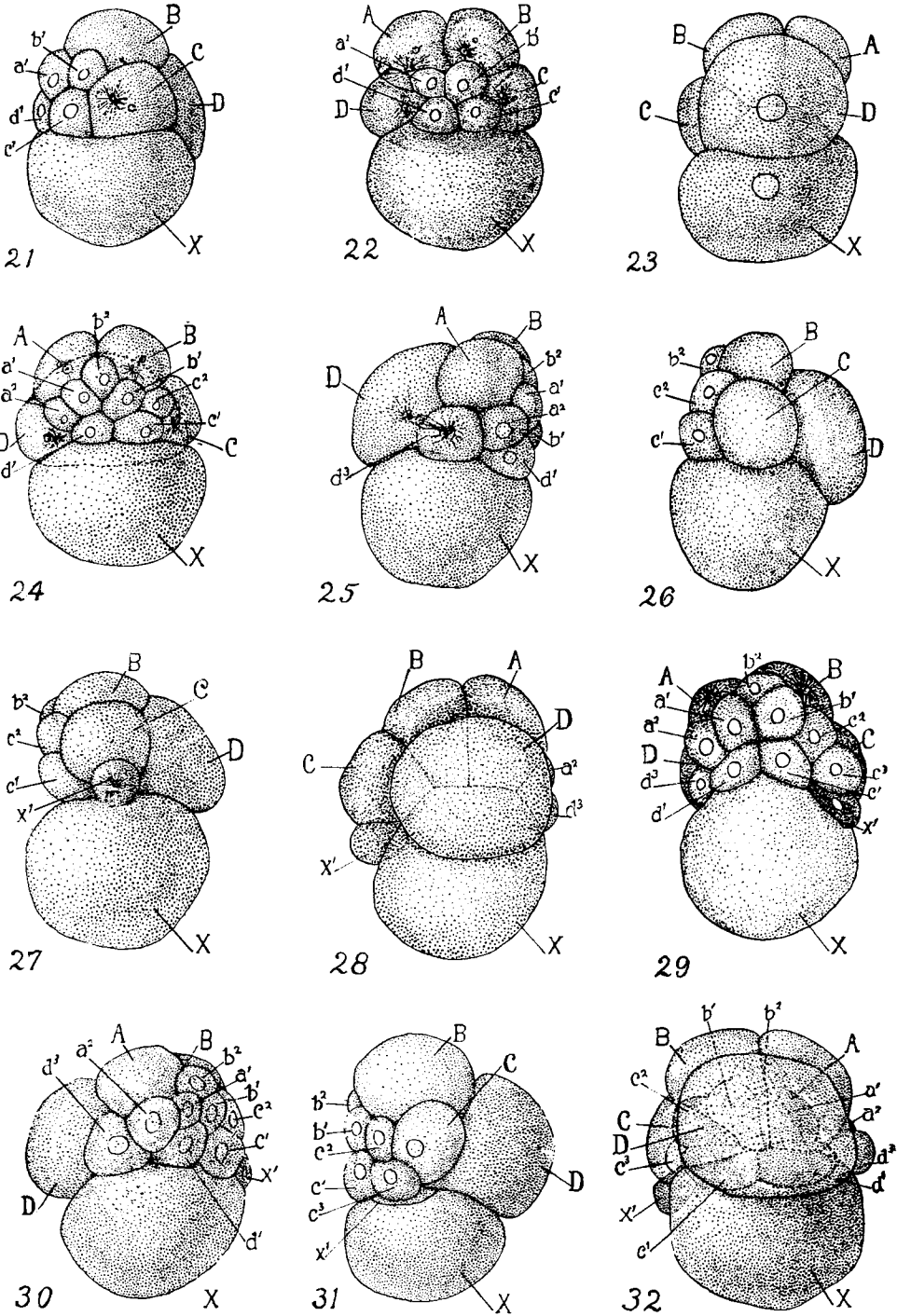
parent cell in the formation of the daughter cells, suggest that the apparent rotation process is not mechanical or even produced by pressure of the macromeres. The formation of d^1 in figure 10 shows how the process takes place; d^1 is budded off obliquely from the macromere D over the inner posterior edge of A and becomes partly imbedded in A. Its final position is determined by the direction of the cleavage spindle. This characteristic method in the formation of the ectomeres is quite a prevalent one. It occurs not only in the eggs of annelids, but in those of the molluscs and polyclads as well.

Fourth cleavage: A nine-cell stage is reached in *Bdellodrilus* by the division of the macromere D in an oblique direction. Figure 16, an eight-cell stage viewed from the left side, shows the position of the macromeres A, B and C with reference to the macromere D, before the formation of the ectomere d^2 . The large macromere D contains about two-thirds of the volume of the dividing ovum. In preparation for the formation of d^2 , D elongates in an oblique direction at an angle of about forty-five degrees to the horizontal plane of the developing embryo. The ventral anterior portion of D shifts forward beneath A, B and C (fig. 17). After the formation of d^2 , D takes a position directly beneath the first generation of ectomeres, and completely covers the inner ends of A, B and C (figs. 19–20). In some instances D is shifted more anterior and completely covers the ventral surface of the other macromeres (fig. 20); but in most cases, as in the nine-cell stage, D occupies the region of the ventral pole, directly beneath the first generation of ectomeres (figs. 22–23). The formation of d^2 is shown in figures 16 to 19. The division is equal in most cases. When unequal, d^2 is the larger cell. In figure 20, a nine-cell stage turned to the observers left so that all the cells are visible, A and B are preparing for the formation of a^2 and b^2 . In most the succeeding stages d^2 , the 'first somatoblast' will be designated by the capital letter X. It is also the first cell of the second generation of ectomeres. The formation of a^2 , b^2 and c^2 is shown in figures 20, 21, 22 and 24 in side and top views respectively. The second generation of ectomeres, with the exception of d^2 (X), is about the same size as those of the

PLATE 2

EXPLANATION OF FIGURES

- 21 Nine-cell stage, viewed from the right side.
- 22 Same stage as the preceding, from the upper pole; spindles for the second generation of ectomere are forming.
- 23 The same stage from the ventral pole.
- 24 Twelve-cell stage from the upper pole; spindles for the third generation of ectomeres are forming.
- 25 Thirteen-cell stage from the left side; the cell d^3 nearly formed.
- 26 Same stage as the preceding from the right side; the embryo is considerably elongated.
- 27 Fourteen-cell stage from the right side; X dividing to form x^1 .
- 28 Same stage as the preceding, ventral view.
- 29 Fifteen-cell stage from the upper pole; c^3 budded off symmetrical with d^2 .
- 30 Fifteen-cell stage from the left side; upper pole turned considerably to the left.
- 31 Same stage as the preceding from the right side; the small cell x^1 is drawn out between c^3 and X.
- 32 Fifteen-cell stage, ventral view, as a transparent object, with the position of all the cells indicated; drawn from a prepared specimen, cleared in xylol.



Kline and Tannreuther, del.

first. The order of their formation is d^2 (X), c^2 , b^2 , a^2 , the same as the first generation. Figure 24 illustrates the twelve-cell stage from the upper pole. The cells are somewhat flattened. The macromere D is located a little to the left of the median longitudinal plane, while X is symmetrical with reference to the median axis of the future worm.

The cleavage spindles of the third generation of ectomeres form in an oblique direction. The thirteen-cell stage is reached by the formation of d^3 . The manner in which d^3 is formed, is rather unique when we take into consideration the size and position of D with reference to the other macromeres A, B and C (figs. 23-25). It is budded off from the outer surface of D and takes up a position symmetrical with c^3 . The fourteen-cell stage is reached by the formation of x^1 (fig. 27), it is budded off from the median right side of X. Its final position is between X and C. Figure 28 shows the same stage as the preceding in ventral view, turned a little to the observer's right.

Immediately after the formation of x^1 the macromere C buds off c^3 , thus making a distinct fifteen-cell stage (fig. 29). The order of formation of the third generation of ectomeres is the same as the first and second. Figures 29, 30, 31 and 32 show the fifteen-cell stage in dorsal, left, right and ventral views respectively. In figure 33, a^3 and b^3 are formed and the first generation of ectomeres are preparing to divide; d^1 and c^1 divide first; at the same time x^2 is budded off from X, symmetrical with x^1 , between D and d^3 , thus making a twenty-cell stage (fig. 34). Figure 35 is a side view of a twenty-two-cell stage after a^1 and b^1 have divided. This figure represents an anterior posterior elongation of the embryo, which is a very common occurrence. The cells, taken as a mass, are very plastic and may assume different shapes. This peculiarity is only secondary and has no special significance. The cells become more spherical before division and flatten out somewhat after the division is complete.

The twenty-three-cell stage is reached by the formation of x^3 from the upper posterior side of X between c^1 and d^1 (fig. 36).

The division of the first generation of ectomeres is unequal and radial rather than oblique. From the twenty- to the thirty-

cell stage several types of cleavage are present; oblique, radial and bilateral. This period of variable cleavage will be designated as the transitional period.

b. The transitional period of cleavage: twenty- to thirty-cell stage

After the formation of x^3 there is a short inactive period and in many of the developing embryos, the cleavage furrows become very indistinct. Cleavage is again initiated by the formation of d^4 from the large macromere D. The cleavage is oblique and very unequal (text fig. 5 and fig. 85).

The smaller cell is almost completely hidden when first formed. It is budded off directly between A and B, near the ventral anterior surface (fig. 37). The smaller cell persists as D (entomere) and the larger cell d^4 or M becomes the 'second somatoblast.' After the formation of M the entire endoderm is contained within the entomeres A, B, C and D (figs. 37-38).

The germ layers are now distinctly separated and the embryo at this stage of development is composed of twenty-four cells (text figs. 6-7). Nereis at the same period of differentiation, shows thirty-eight cells. Unio (Lillie) at the time of the separation of the germ layers contains thirty-two cells. This difference is due, in case of *Bdellodrilus* to the lagging of division in the cells of the upper pole.

The composition of the embryo at the twenty-four-cell stage is as follows:

Entomeres.....A, B, C, D.....	4
Ectomeres.....of first generation.....	8
Ectomeres.....of second generation.....	4
Ectomeres.....of third generation.....	4
Mesoblast.....M.....	1
First somatoblast derivatives.....	3

24

Many of the cells during the transitional period have a definite shape and if separated from the cell complex, they could be readily recognized. The embryo at this stage of development is somewhat spherical (figs. 36-38). Immediately after

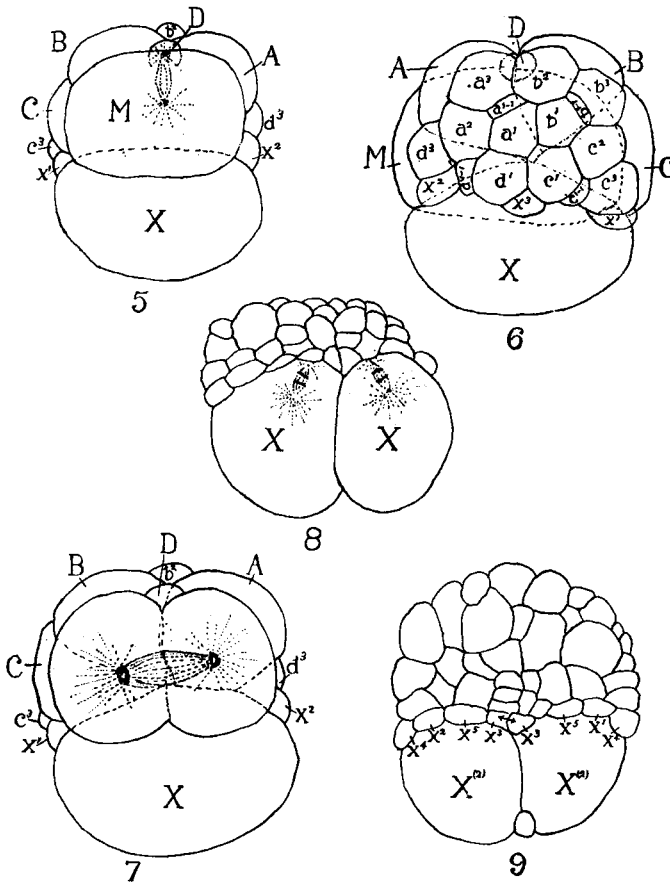


Fig. 5 Twenty-four-cell stage in ventral view, to show the division of the large macromere D. The larger of the two cells d⁴ (M) becomes the 'second somatoblast.' The smaller cell D, becomes one of the four entomeres; D is scarcely visible from the exterior.

Fig. 6 Same as the preceding, in an apical pole view.

Fig. 7 Twenty-four-cell stage, ventral view, shows the bilateral division of M. D after the bilateral division of M becomes more distinct from surface view.

Fig. 8 Horizontal section of an early embryo to show spindles in the formation of x⁵ from either proteloblast X, X.

Fig. 9 Horizontal section little later than preceding, to show the small cell x¹-x⁵; apical pole view.

the establishment of the germ layers, the bilateral division of the 'first and second somatoblasts' occurs. The bilateral division of the 'second somatoblast' usually precedes that of the first; occasionally they divide simultaneously.

c. The bilateral period of cleavage: twenty-five-cell stage

The first bilateral cleavages occur in the first and second somatoblasts (text fig. 7 and figs. 37-43). The small superficial cells of the lower pole are derived from the second and third generation of ectomeres and from the derivatives of X. The arrangement of these cells with reference to the blastopore is shown in figure 42. The entomeres A, B, C and D are partly grown over by the other cells and the open space becomes the blastopore. It is bounded anteriorly and laterally by small cells from the second and third quartettes, and posteriorly by the primary mesoblasts M, M. Its hinder lip, which is formed by the primary mesoblasts, lies anterior to the center of the lower pole. The closure of the blastopore takes place by a convergence of the cells from all sides. The principal growth of cells is from in front backwards, formed by the derivatives of the second and third generation of ectomeres (figs. 42, 43, 51). The entomeres now divide very rapidly and the cells soon become smaller than those of the ectomeres, which grow over them (figs. 43, 51).

3. THE FIRST SOMATOBLAST

The history of the 'first somatoblast' in *Bdellodrilus* is of considerable interest when considered from the standpoint of its origin and its derivatives. When first formed from the posterior macromere D, it contains one-third of the entire bulk of the developing embryo. As already described, it first buds off the small cell x^1 on the right, x^2 symmetrically on the left and a third cell, x^3 , on the median posterior upper side. These three small cells are symmetrical with reference to the median longitudinal axis. The fourth cleavage divides the somato-

PLATE 3

EXPLANATION OF FIGURES

33 Seventeen-cell stage from the upper pole; spindles in the first generation of ectomeres forming.

34 Twenty-cell stage from the upper pole; cells c^{1-1} , d^{1-1} and x^2 just formed.

35 Twenty-one-cell stage from the right side; b^{1-1} , new cell; this embryo is unusually elongated.

36 Twenty-three-cell stage from the upper pole; d^{1-1} and x^2 , two new cells formed; embryo considerably flattened.

37 Twenty-four-cell stage from the ventral side; the unequal division of D has just occurred; D partially visible; the cleavage spindle of M forming.

38 Twenty-five-cell stage, ventral view; division of M complete and the spindle for the first bilateral division of X is forming.

39 Twenty-five-cell stage from the left side.

40 Twenty-five-cell stage from the upper pole; embryo is nearly spherical.

41 Twenty-six-cell stage, ventral view.

42 Same as the preceding, ventro-anterior view.

43 Twenty-nine-cell stage, same aspect as preceding; a^4 , b^4 and c^4 are the three new cells formed.

44 Forty-two-cell stage from the upper pole; increase in number of cells due to the rapid division of the ectomeres.

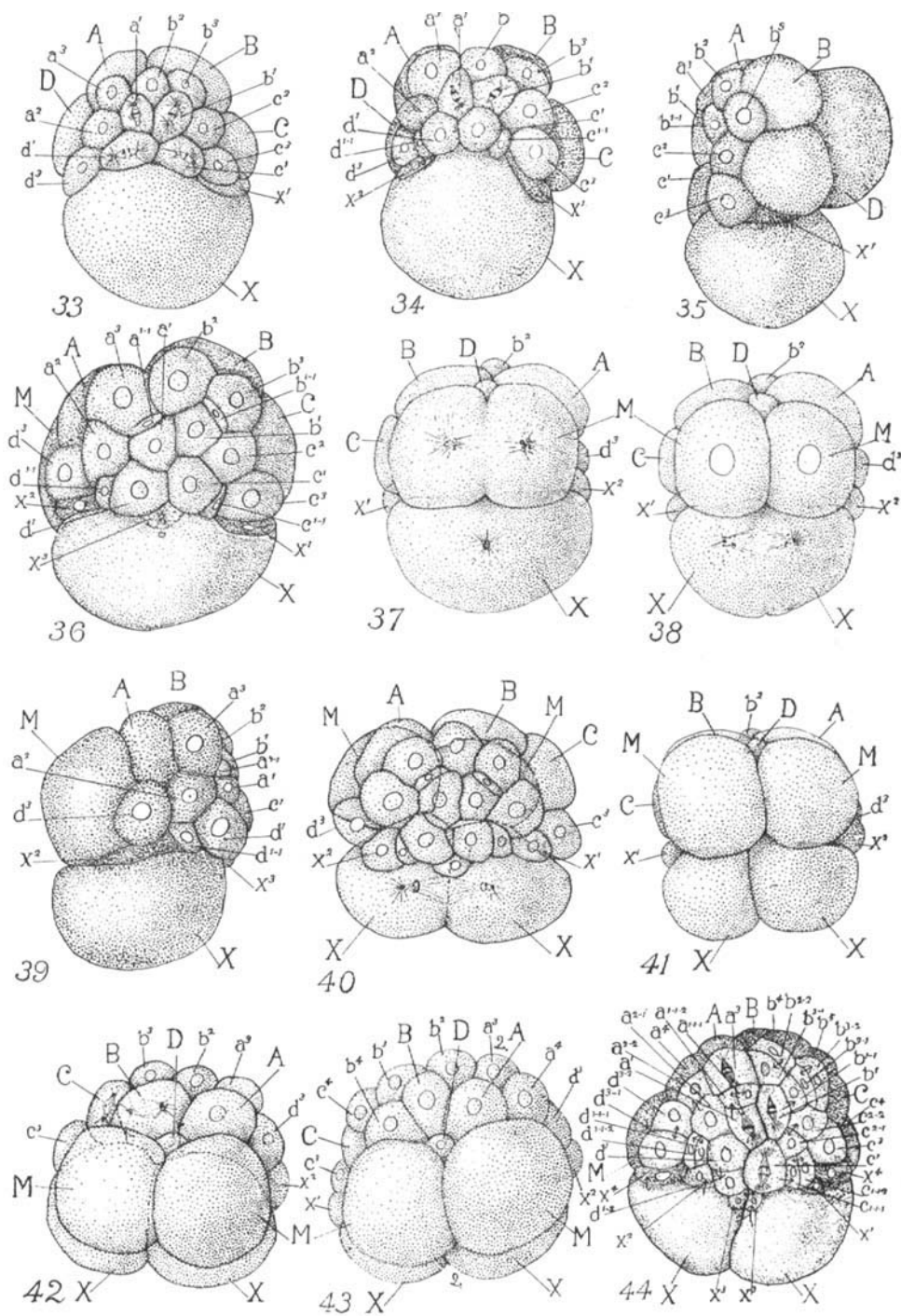


PLATE 4

EXPLANATION OF FIGURES

- 45 Sixty-three-cell stage from the upper pole.
- 46 Same stage as the preceding, from the ventral pole; very few of the ectomeres are visible.
- 47 Seventy-cell-stage, ventral view. Increase in number due to division of small cells. Consult table of cleavage. First nearly equal division of the protoblasts, X, X. This division separates the neural and nephridial elements.
- 48 Seventy-two-cell stage, ventral view.
- 49 Little later than the preceding stage, to show x^6 and x^7 .
- 50 Embryo, ventral view, to show the first division of the nephroblasts; transverse axis greater than the longitudinal.
- 51 Embryo, ventral view, turned anteriorly to show the blastopore.
- 52 Embryo to show the lengthening of the antero-posterior axis. The small cells, x^6 , x^6 , are good points to mark the orientation of the different figures. All figures on plate are similarly orientated with reference to the right and left sides of the embryo.
- 53 Stage a little later than the preceding.
- 54 Embryo from upper pole, to show derivatives of x^6 between the nephroblasts.
- 55 Embryo with upper surface turned posteriorly, to show the rapid division of the ectoblast cells.
- 56 and 57 Show that either nephroblast $X^{(2)}$ or $X^{(3)}$ may divide, to form the three nephroblasts on either side.

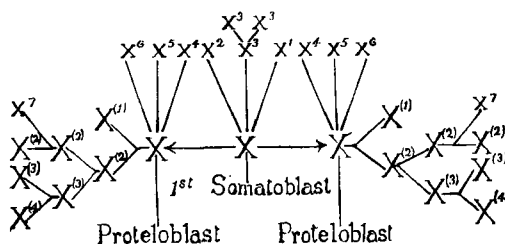


blast into two equal parts, right and left (figs. 40–41). These two cells, for convenience in description, will be called the posterior right and left proteloblasts. At the fifth division each proteloblast buds off a small cell, x^4 , external to x^1 and x^2 respectively (fig. 44). At the sixth division each of the proteloblasts buds forth a small cell, x^5 on either side of x^3 , beneath the derivatives of c^1 and d^1 (text fig. 8). At the seventh division each of the proteloblasts buds off a small cell, x^6 on the ventro-anterior edge at the junction of the two cells (fig. 46).

At the eighth division each proteloblast, on either side of the median plane of the embryo, divided into two equal parts (figs. 47, 48, 93). The four cells formed become the posterior teloblasts, $X^{(1)}$ and $X^{(2)}$, right and left of the median axis (fig. 47). $X^{(1)}$ without any further division, becomes the neuroblast on either side, and $X^{(2)}$ becomes the nephroblast on either side of the median axis (figs. 47–48). Next $X^{(2)}$, right and left, divides equally, giving $X^{(2)}$ and $X^{(3)}$ (figs. 50, 52). Next either $X^{(2)}$ or $X^{(3)}$ divides equally. If $X^{(3)}$ divide, which is the common occurrence, we get $X^{(3)}$ and $X^{(4)}$. But if $X^{(2)}$ divide instead of $X^{(3)}$, the final result is the same. In either case, we get four teloblasts on each side (one neuroblast and three nephroblasts) as shown in figures 56 and 57. Next $X^{(2)}$ on either side divides very unequally and gives rise to x^7 on the anterior ventral outer surface (fig. 49).

The progeny of the 'first somatoblast,' when the teloblasts are completely formed, is twenty cells. Table 2 shows the derivatives of the 'first somatoblast.'

TABLE 2



4. THE SECOND SOMATOBLAST

Immediately after the formation of X from the posterior macromere D, d^3 is budded off from D (figs. 24-25). Next D divides very unequally in an oblique direction and gives rise to d^4 (M) the 'second somatoblast,' as previously described. Figures 37 and 85 show the position of D and M after the cleavage of the macromere D. The twenty-five-cell stage is reached by the bilateral division of M (figs. 37-38). The cells M, M at first are a little to the left of the median plane, but later in course of development they become symmetrical to the longitudinal axis of the adult worm.

Soon after the bilateral division of M, the 'second somatoblast,' each cell M, M right and left buds off five or more small cells directly beneath the first generation of ectomeres (figs. 86-87). It is impossible to detect these cells except by means of sections, hence the uncertainty as to their exact number. They are characterized by their large nuclei with homogeneous staining chromatin and they contain but little yolk material. These small cells divide once or twice soon after their formation from the primary mesoblasts and then remain inactive until late embryological development, at least until after the germ bands are completely formed and the embryo has undergone considerable differentiation (as the formation of the lumen of the digestive system, etc.).

These undifferentiated mesodermal cells occupy the region which becomes the central dorsal side of the embryo, at the point where the developing worm is completely turned on itself (figs. 92, 98-99). The history of these cells can be readily followed through their different stages of development, so that there can be no question as to their exact origin and history. When the embryo begins to straighten, the progeny of these small cells extend toward either end and form the splanchnic and somatic mesoderm on the dorsal side of the worm. This secondary mesoderm later becomes continuous with the primary mesoderm, which forms directly from the mesoblasts M, M.

5. THE ENTOBLAST

The formation of the entoblast in *Bdellodrilus* represents an unusual type of development among the annelid worms. The macromeres A, B, C and D, after the formation of d^4 , give rise to the entire entoderm. D is greatly reduced after the formation of d^4 . The position of the entomeres is shown in figures 40 and 42. In figure 40 A, B and C appear rather large, because of the flattened condition of the cells. In figure 42 (from the ventral pole) the cells are rounded and appear more normal. The position of the entomeres and their boundary cells are distinctly shown. This figure shows more clearly the bulk of the entoderm, when compared with the mass of the entire egg. In figure 43 (a twenty-nine-cell stage) A, B and C have divided nearly equally. This division is considered by some investigators as the formation of the fourth generation of micromeres; d^4 of the D quadrant has formed earlier. Figures 44 and 45 (apical pole views) show the upper outer edge of the entodermal cells. In figure 46 (the same stage as preceding from the ventral pole) a very small part of the entodermal and ectodermal cells are visible. This figure shows the prominence of the four large cells, which later form the ten teloblasts. These four large cells, from their position, resemble the four large entomeres, which are so prominent in many other forms. These cells (X, X, M, M), according to Selensky, share equally in the formation of the germ layers, i.e., ectoderm, endoderm and mesoderm are produced by each of them.

In forms like *Clepsine*, *Crepidula* and others, at a similar or later stage of development, the entomeres are very prominent and the ectomeres with the first and second somatoblasts, form a cap of cells on their upper surface. In *Bdellodrilus* the conditions are different. The ectomeres and the entodermal cells form a cap of cells on the upper anterior surface of X, X, M and M. This difference is due to the prominence of the first and second somatoblasts, which constitute the greatest bulk of the embryo. At about the seventy-cell stage the ectodermal and endodermal cells are nearly uniform in size (figs. 47-49). In

figure 51 (a little later stage) the blastopore is nearly closed. This early closure of the blastopore in *Bdellodrilus*, is due largely, to the ventro-anterior shifting of the macromere D over A, B and C in the formation of the somatoblasts (figs. 18, 85).

The closure of the blastopore, in some of the annelids, occurs at a very late stage of development. In Clepsine the teloblasts give rise to rows of cells, which pass anteriorly around the entomeres A, B and C beneath the edge of the blastodisc or cap of cells. The blastodisc with these rows of cell cover about half of the entomeres. By the downward growth of the blastodisc and the concrescence of the germ bands, the closure of the blastopore is completed. The closure of the blastopore in Clepsine occurs on the ventral side, nearer the anterior end. In *Bdellodrilus*, the germ bands are not formed until later and take no part in the closure of the blastopore. Text figures 10 to 13 show the position of the ectoderm, entoderm, and the first and second somatoblasts, at different stages in the closure of the blastopore. The region of closure is similar to that of Clepsine.

At the time of the formation of the secondary mesoblast just beneath the first generation of ectomeres, the entire entoderm is situated in the anterior half of the embryo. But soon after the formation of the m cells (text fig. 12 and fig. 86), the entodermal cells by a rapid proliferation extend posteriorly between the m cells and the primary mesoblast. During the formation of the primary mesoblast, the meso-teloblasts themselves are carried posteriorly, ahead of the entoderm. The entoderm, thus becomes situated between the m cells or secondary mesoderm above and the mesoblast bands or primary mesoblast below. The entoderm in reality never reaches the posterior limit of the meso-teloblasts, as shown in figures 98 and 99.

The interior of the developing embryo, now consists of a solid mass of small entodermal cleavage cells (figs. 95-99), heavily laden with yolk. These cells are readily distinguished from the surrounding mesodermal cells, by their deeper cytoplasmic stain. Figure 99 (a vertical longitudinal section near the median plane) shows the position of the entodermal cells in the embryo.

As the embryo elongates, the entodermal cells increase in number. This process of growth is continued until the digestive tract is completely formed. Figure 99 shows the anterior and posterior limits of the digestive tract, which is formed from the four entomeres. The anterior end shows a distinct lumen, while the posterior end is yet a solid mass of cells. The entire digestive tract, except the insignificant stomodaeum and proctodaeum, is entodermal in origin. The proctodaeum is not formed until the time of hatching. It occurs on the dorsal side of the tenth segment. The embryo is completely turned on itself (fig. 99) and brings the anterior and posterior ends of the digestive tract in close proximity. The differentiation of the digestive tract begins anteriorly and progresses posteriorly. As growth continues, the outer cells of the entodermal mass form an epithelial layer. At first the cells are somewhat flattened, but soon take a columnar position, and form the columnar epithelium of the digestive tract. The cells within soon lose their staining properties, break up and serve as food for the developing embryo.

The digestive tract in its course of development, passes through the following stages: the first stage is represented by the four entomeres A, B, C and D; the second stage by a solid mass of cleavage cells (the cell boundaries are often very indistinct) within the center of the embryo; the third stage by an elongation of the entodermal mass as the larva lengthens, and by the establishment of a lumen; the fourth stage by a thin layer of flattened epithelium and later a columnar epithelium; the fifth stage, the cells within the epithelial layer serve as food; and sixth the formation of the stomodaeum and proctodaeum.

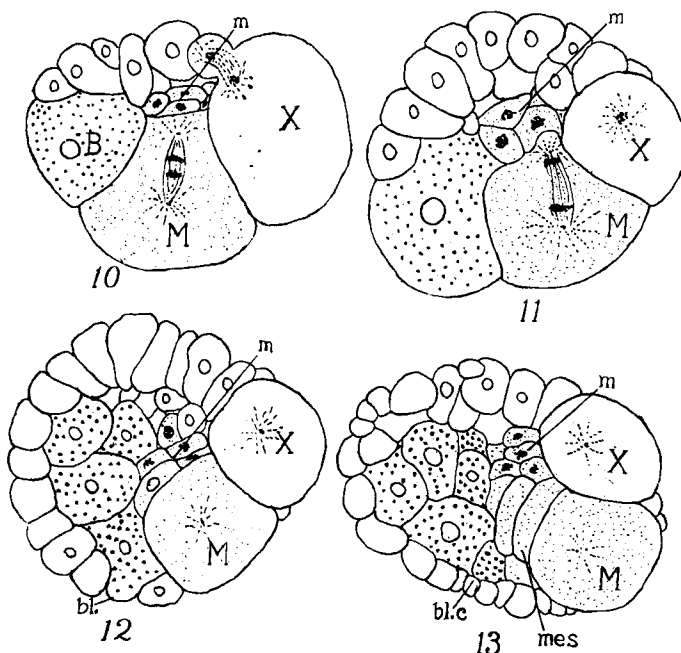
GENERAL HISTORY OF THE GERM BANDS

The term 'germ bands' has been variously interpreted by different investigators on cell lineage. The term germ bands, or the German equivalent 'Keim Steifen,' is usually restricted to the strata derived from the teloblasts, the ectoblastic layer being excluded. It is held by others that the germ bands of annelids are purely mesoblastic.

Balfour, Hatschek, Goette, Kowalevsky and many others made use of the term 'mesoblastic bands' as the equivalent of the germ bands. In Hirudinea, according to Whitman, the germ bands are composed of three distinct layers; the ectoblastic, mesoblastic and the neuroblastic elements. Wilson gave the same interpretation in his studies on "The embryology of *Lumbricus*." In *Bdelodrilus* the term 'germ bands' includes the three strata of cells as in Hirudinea and *Lumbricus*.

1. INNER STRATUM OF THE GERM BANDS

After the formation of the teloblasts, five on either side of the median axis (one neuroblast, one mesoblast and three nephroblasts), the mesoblasts or meso-teloblasts are the first to begin the formation of the germ bands by a forward proliferation of cells near the posterior lip of the blastopore (text figs. 12, 13, 17). The plane of division is nearly at right angles to the formation of cells in the secondary mesoblast (text figs. 10, 13). The cells of the mesoblast bands are considerably smaller than the teloblasts from which they originate. They grow forward between the entoderm and the ectoderm and finally meet at the anterior end of the larva. As these bands grow forward they become several cells broad, but seldom more than two cells deep. Their differentiation begins anteriorly and progresses backward. The first cells of the mesoblast bands, when formed, are on the surface, but soon become covered by the ectodermal cells. As the mesoblast band extends forward below and around the entoderm, it forces its way to the extreme anterior end of the embryo beneath the ectoderm. It finally encloses the digestive tract on the ventral and lateral sides and becomes continuous with the secondary mesoblast on the dorsal side. The two mesoblast bands fuse first at the anterior end along the median ventral side and subsequently with the dorsal secondary mesoderm. In figure 99 (a longitudinal section) the mesoblast is differentiated into splanchnic and somatic layers, with the coelom between. The longitudinal muscles become differentiated before the circular. At the extreme posterior end the



Figs. 10-13 Diagrammatic figures to show the ventro-posterior extension of the ectomeres, in the closure of the blastopore.

Fig. 10 Thirty-three cell stage, taken a little to the right of the median plane.

Fig. 11 Vertical section to the left of the median plane.

Fig. 12 Vertical section of a ninety-cell stage.

Fig. 13 Vertical section of an embryo at the time of closing of the blastopore. The mesoblast bands have just begun.

The heavy stippling represent endoderm; the light stippling mesoderm, and the unstippled the ectoderm or ectomeres. *m*, secondary mesoblast; *M*, mesoblasts; *bl*, blastopore; *bl.c*, point where the blastopore closes; *X*, derivatives of the 'first somatoblast;' *mes*, mesoblast bands.

meso-teloblasts are represented by an undifferentiated mass of cells, which later give rise to the musculature of the last three segments of the worm, and are directly concerned in the formation and movements of the posterior sucker.

2. MIDDLE STRATUM OF THE GERM BANDS

The middle stratum of the germ bands can readily be distinguished while the embryo is still nearly spherical. Upon close examination it is seen that the ectoblast cells are arranged into four distinct rows, on either side of the median ventral axis (figs. 65-66). Each row terminates posteriorly in a large cell or teloblast.

Text figures 15 and 17 and figure 58 show the early formation of these rows of cells. Sections of these various stages show that

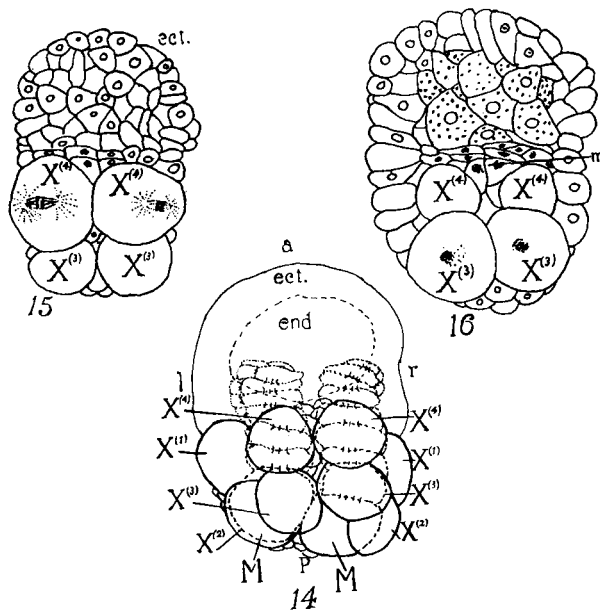


Fig. 14 Surface view from upper pole, to show the position of the ten teloblasts. The meso-teloblasts or mesoblasts have budded off eight or ten cells in the formation of the mesoblast bands. Their position is indicated by dotted outline. The broken outline represents the region of the entoderm. The position of the ectoderm is indicated by a continuous line.

Fig. 15 Third horizontal section from the top, passing through four of the large nephroblasts. The spindles represent the beginning of the first division in the formation of the middle germ band. The anterior end and the right side of the section are a little below the horizontal plane.

Fig. 16 Seventh horizontal section from the top. It passes through the upper portion of the entoderm and the secondary mesoblast (m).

these rows of cells form a part of the general ectoderm, being partly covered here and there by adjoining cells. In later stages of development, these rows of cells become completely covered as they gradually sink beneath the surface, and thus come to lie between the mesoblast and the ectoderm or ectoblast.

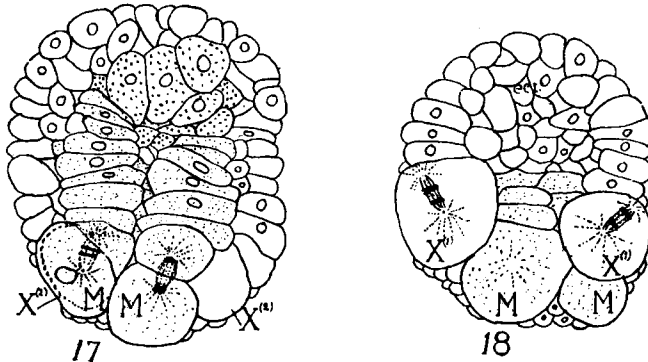


Fig. 17 Twelfth section from the top, to show the anterior extension of the mesoblast bands below and around the entoderm.

Fig. 18 Nineteenth section from the top, to show the lower side of the mesoblast. The section passes below the entoderm. The spindles represent the beginning of the first division of the neuroblasts to form the neural rows.

Heavy stippling represents entoderm; light stippling mesoderm and the unstippled portion the ectoderm. The sections of figures 15-18 were eight micra thick. There were 21 sections in all. *a*, anterior, and *p*, posterior, represent the respective ends of the cleavage cells, but not the future ends of the embryo; *r*, right; *l*, left; *ent*, entoderm and *ect*, ectoderm.

3. OUTER STRATUM OF THE GERM BANDS

This stratum forms the definitive ectoderm and needs no further description at this point of development.

The embryo now elongates very rapidly, and the general shape of the adult worm becomes recognizable. The teloblasts become less and less distinct, until finally the cell rows terminate posteriorly in a group of small cells. The mesoteloblasts extend farther posteriorly than the neuroteloblasts. New cells are always formed from the anterior surface of the teloblasts. There can be no question as to the origin of the germ bands from corresponding teloblasts, as their formation

can be followed step by step. The mesoblastic and neuroblastic portions of the germ bands can be traced to the anterior end of the embryo. The meso-teloblasts are the last to disappear. They are distinct until after the formation of the stomodaeum and its connection with the pharynx. The concrescence of the germ bands begins anteriorly and progresses posteriorly.

THE ECTODERM AND ITS PRODUCTS

The three generations of ectomeres are given entirely to the formation of the ectoderm, which later becomes differentiated into the definitive hypodermis, with its glands, the cuticle and the anterior and posterior ends of the digestive tract. The ectoderm includes, in addition to the above, all of the teloblasts, except the two larger and deeper ones which represent the mesoblasts. The reason for regarding the eight teloblasts and their derivatives as a part of the general ectoderm, is on account of their origin and position. In position, they are superficial at first and can not be distinguished from the general ectoderm, except by their arrangement in rows. Small cells are budded off from the teloblasts, which form the trunk ectoderm. In *Clepsine* these teloblasts are at first superficial at the posterior end of the embryo. In *Lumbricus* they are found directly in the general ectoderm, and beyond question form a part of it.

1. THE NERVOUS SYSTEM

The nerve chain in *Bdellodrilus* first appears as a double row of cells, nearly uniform in size. Each row of cells originates from a single cell, the neuroblast. The neuroblasts, when first formed, are widely separated, but symmetrical to the median axis of the body. Figures 47 and 48 (in ventral view) show their position when first formed by an equal division of the proteloblasts X and X. They take up their position on either side of the mesoblasts (figs. 47, 48, 93). When first formed the neuroblasts are turned somewhat anteriorly as shown in the horizontal section of figure 93. This movement of the cells to their final position, independent of the former position of the cleavage

spindle, is a common occurrence in *Bdellodrilus*. In some instances it is necessary to employ sections, in order to determine the origin of cells. The transverse axis of the embryo at this stage is often greater than the longitudinal (figs. 49–50). This condition persists for a brief period only, during the formation of the teloblasts. As the embryo increases in length the neuroblasts are carried more and more posteriorly (figs. 56–57).

In order to get a better understanding of the origin and orientation of the neuroblasts— $X^{(1)}$ right and $X^{(1)}$ left—with reference to the other teloblasts, the figures of plate 4 are so arranged that the left side of the developing embryo corresponds to the reader's left. In figure 45 the upper pole is turned a little posteriorly, to show the upper outer edges of the entodermal cells. Figure 47 (from ventral pole) shows some of the ectodermal cells. The remaining figures are either turned forward or backward on their transverse axes. The ectomeres x^6 and x^6 right and left serve as good points for orientation (figs. 46–53). After the formation of the teloblasts, bilateral symmetry is fully established. The meso-teloblasts, however in some instances, are still a little to the left of the median axis. This variation in the symmetry of the mesoblast does not in any way change the end result. In the early history of the germ band formation the teloblasts $X^{(3)}$ and $X^{(4)}$ are slightly separated, while $X^{(1)}$ and $X^{(2)}$ are widely separated from the corresponding teloblast on the opposite side (figs. 56, 58). The neuroblasts and the nephroblasts begin their proliferation of cells to form the germ bands, about the same time (fig. 58). At this stage of development, the exact orientation of the embryo is distinct. Since the embryo is completely turned on itself, the further use of the terms, apical and ventral poles, is significant only as being convenient in description. The mouth, as stated above, is formed in the center of the apical pole and the anus in close proximity on the dorsal side of the tenth segment. Figure 59 (upper pole view) shows the complete curvature of the embryo. The heavily shaded portion represents approximately, the boundary between the anterior and posterior ends. This figure shows that the teloblasts are coming more and more in a straight line. Since the two ends of

the embryo are in immediate contact, it is impossible, except by longitudinal sections, to determine the exact point of separation. The ectoderm of the anterior end of the embryo, which is derived from the three generations of ectomeres is continuous with the ectoderm derived from the 'first somatoblast.'

The separation of the two ends of the embryo becomes recognizable in the early formation of the germ bands, as shown in figures 59 and 60. The posterior and ventral shifting of the neuroblasts (figs. 58-60) continues until all of the teloblasts are in a direct line. The small cells between the teloblasts are derived from the first somatoblast. In viewing the embryo from the upper pole (which now corresponds more to the anterior and posterior ends of the future animal) the germ bands extend laterally, downward and forward, being curved somewhat posteriorly as they pass from the upper to the lower pole (fig. 59). The meso-teloblasts in figure 58 are still visible from the exterior. In figure 59 they are almost grown over, while in figure 60 they are completely covered. This is due to the posterior shifting of the neuroblasts and the growth of the ectomeres from above and below. In an embryo viewed from the right side (fig. 61, a little older than fig. 60), the position of the neural and nephric rows of the germ band are shown. As the rows extend anteriorly they are more difficult to distinguish from the ectoderm. The neural rows alone can be followed to the extreme anterior end. The posterior end of the embryo is widely blunt, while the anterior end is more rounded. The heavily shaded portion represents the point of separation between the two ends.

Figure 62 represents the same embryo from the upper pole, with the ends of the embryo rotated or turned a little posteriorly. In figures 63 and 64 (from right and left sides respectively) the embryo is more elongated and the point of separation between the two ends is more distinct. The neuroblasts are lagging in their posterior extension. Their position is median ventro-posterior, as shown in figure 65. Their concrescence is not yet complete at the posterior end. In the following stages of development the cells of the neural and nephric rows divide

PLATE 5

EXPLANATION OF FIGURES

58 Embryo from upper pole, tilted a little to the right. The position of the ten teloblasts are shown; the small cells between the teloblasts on the surface are derived from x^6 and x^7 on either side.

59 Same view as the preceding; the neuroblasts have migrated a little posteriorly and are approaching each other.

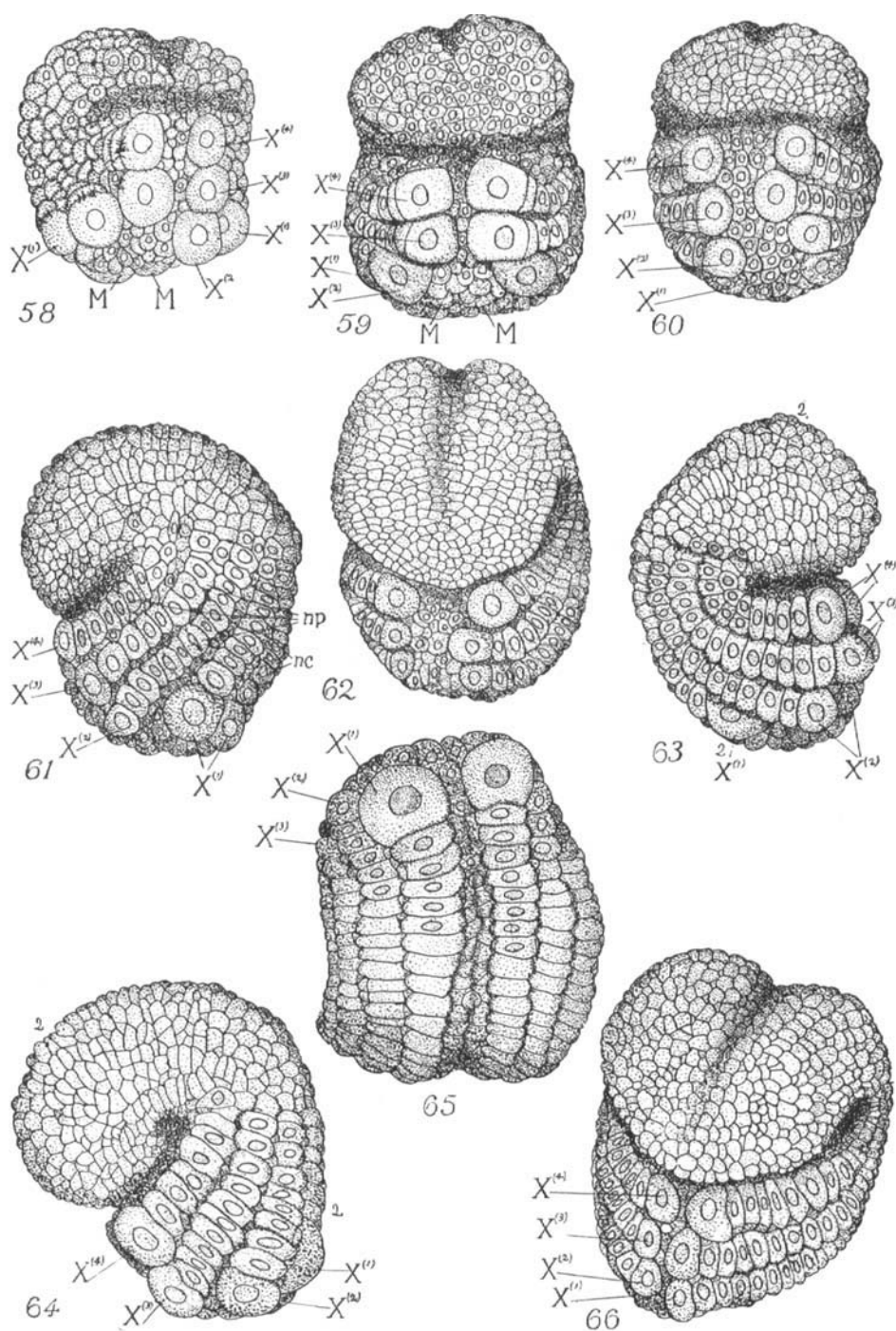
60 The ecto-teloblasts are nearly in a direct line; the germ bands have begun to form; the two primary mesoblasts M, M are no longer visible from the exterior; the transverse heavily shaded portion shows the approximate point of separation between the two ends.

61 Embryo viewed from the left side; the posterior end is extremely blunt.

62 Same embryo as preceding, from the upper pole (upper pole corresponds to the anterior and posterior ends). Shows very strikingly the close proximity of the two ends.

63-65 Represent the same embryo from the right, left and ventral sides respectively. The ectoderm which partially covers the germ bands is not shown.

66 Embryo from upper pole; bilateral symmetry is well marked; the teloblasts are considerably reduced by the time they come in contact with their fellows on the opposite side.



Tanneuther del

PLATE 6

EXPLANATION OF FIGURES

67 Embryo turned slightly to the left to show the anterior and the posterior ends; the embryo at this stage begins to rotate within the egg membrane.

68 The same as the preceding from the ventral pole, turned a little to the left.

69 Embryo viewed from the right side; condition before the posterior end becomes drawn out or pointed.

70 Embryo from the upper pole; shows compressed condition of the two ends; at this stage the embryo rotates very rapidly.

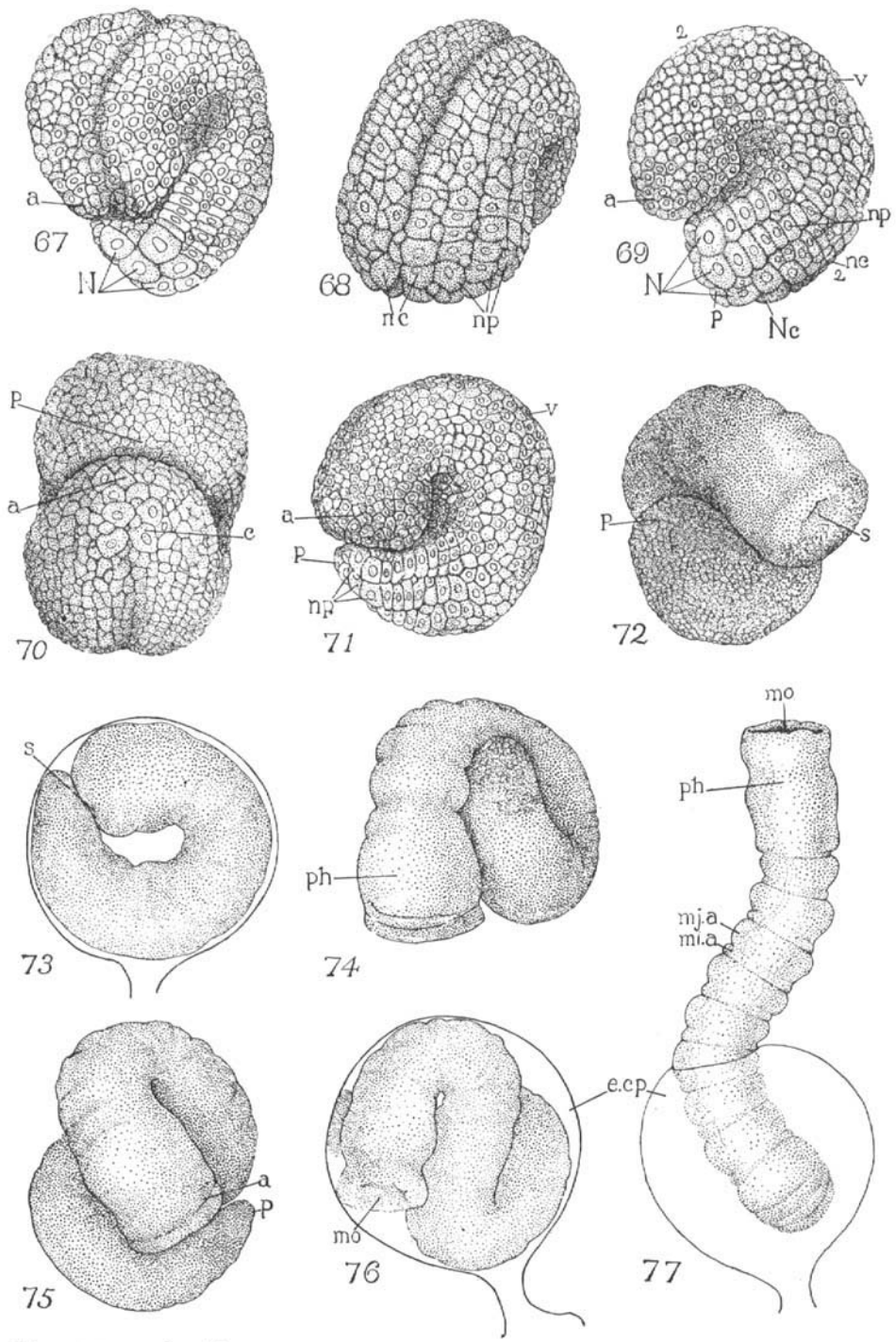
71 Embryo viewed from the right side; the teloblasts are partially visible at the posterior end; the tapering of the posterior end is well marked.

72 Embryo to show the overlapping of the ends; indications of segments are visible anteriorly; the stomadaeum is distinct.

73 Unusual condition, where the two ends remain in immediate contact until after the form of the worm is distinct; this occurs in eggs with an unusually large cocoon.

74-6 Different stages in the final growth of the embryo.

77 Condition of embryo at the time of emergence from the egg.



Kline and Tannreuther, del.

very rapidly and gradually become covered by the ectodermal cells as they sink beneath the surface (figs. 66-72), and form the middle stratum of the germ bands. In a nearly median longitudinal section (fig. 99), the relation of the parts are shown. The neural plate at different points shows the formation of ganglia. The anterior end of the section passes through the exact median plane and does not show any ganglia. The neuroblasts still persist at the posterior end.

The entire nervous system arises as two simple longitudinal rows of cells, and each row is produced by the continued proliferation of cells from a single cell, 'the neuroblast.' This proof is established by the study of surface preparations in connection with sections taken in different planes through the developing neural elements. The neural rows can be followed to the extreme anterior end, where they pass up around the pharynx and give rise to the cerebral ganglia on either side (fig. 96) by a thickening of the anterior extremity of the corresponding neural rows. There are exactly four rows of cells in the middle stratum of each germ band. The outline of the rows can be easily seen in surface views of the living embryo (figs. 63-66). They are more distinctly marked at the posterior ends, and become less distinct anteriorly, which is due to the more advanced development; that is, each row becomes double, then triple, etc. (figs. 68-69) and at the same time, its boundary lines become less distinct.

2. THE EXCRETORY SYSTEM

After the bilateral division of the 'first somatoblast,' each proteloblast contains the neural and nephric elements of their respective sides. According to Whitman, these two cells are called the neuro-nephroblasts. But when each proteloblast X , X divides equally the neural and nephric elements become separated, $X^{(1)}$ neural and $X^{(2)}$ nephridial (figs. 47-48). The nephroblast $X^{(2)}$ on either side next buds off a very small cell x^7 , which becomes ectodermal (fig. 49). Immediately after the formation of this small cell, $X^{(2)}$ divides nearly equally, and forms

$X^{(2)}$ and $X^{(3)}$ on either side (figs. 50-54). Both cells are nephridial. This fact perhaps is made more suggestive by the behavior of $X^{(2)}$ and $X^{(3)}$. Either of these cells may divide equally, but never both in the same embryo. In either case we have three teloblasts derived from the nephroblast $X^{(2)}$ on either side. The cells of the nephridial rows are somewhat smaller and narrower than those of the neural rows. In some cases the outer nephridial row of cells is very short. In other embryos it is composed of but one or two cells and its presence is hard to verify, suggesting a possible disappearance in the group. As stated above, the nephridia arise in connection with a continuous nephric cord of ectoblastic origin, which forms a part of the middle stratum of the germ band and lies along side of the neural row. Each nephric cord terminates at the posterior end in three teloblasts. Thus the entire nephric cord of cells is formed by the continued division of the 'nephroblasts,' which agree precisely with the neuroblasts in structure, action and mode of origin. The nephric cord at first is composed of three rows of cells posteriorly, but passing forward the rows are no longer definitely separated and the nephric cord or plate consists of an irregular series of cells which extend anteriorly to the posterior end of the pharynx. The formation of the nephridia progresses from in front backwards and keeps pace with the formation of new segments in the embryo. The beginnings of a pair of nephridia are found in each of the main segments. Only two pairs of nephridia are retained in the adult worm. The details of the formation of these segmental organs have not been worked out.

Berg considers the entire nephridia in *Criodrilus* as mesodermal in origin; Whitman held the extreme opposite view, that the entire nephridium was ectodermal in origin; while Wilson regarded the nephridia as being part mesodermal and part ectodermal in origin. In *Bdellodrilus* the nephridia are ectodermal. The anterior pair occurs in the first, second, third and fourth body segments. The left nephridia of the anterior pair, extends from the first to the third segments inclusive, while the right extends from the second to the fourth segments inclusive. Both

have a common opening on the dorsal side of the third segment. The funnel of the left occurs in the second and that of the right in the third segment. The posterior pair is found in the eighth segment. Each nephridium has a separate opening to the exterior on the dorsal side of the eighth segment.

GROWTH

The developing embryo does not increase appreciably in bulk until after the teloblasts are formed. Up to this period it is merely a division of the egg content into the various cell complexes. Even at this stage the increase in the long axis of the embryo is brought about by a decrease in the transverse diameter. Figures 50 and 55 show the transverse axis greater than the longitudinal, while in figure 56 and 57 the longitudinal axis is greater, due more to a change in shape than to growth. The egg content is very plastic and when removed from the cocoon the egg membrane, in most cases, is not of sufficient tenacity to retain the embryo intact. The ten teloblasts are shown in figures 56 and 57.

The first increase in length is due to the formation of the mesoblastic portion of the germ bands (text figs. 17-18). The neuroblastic and nephroblastic portions of the germ bands begin simultaneously after the meso-teloblasts have formed eight or ten cells (text figs. 15, 18 and fig. 58). Figures 58-71 show the various stages in the formation of the germ bands. Figure 71 is about the last stage when the germ bands can be detected externally. A longitudinal section of figure 71 near the median axis shows a differentiation of the germ bands into their incipient organs (fig. 99). From this point of development, growth is very rapid, and the embryo begins to rotate on its transverse axis. The movement is produced by the action of cilia, which occur on the large ectodermal cells in the median ventral half of the anterior end of the embryo (figs. 96-99). These cilia disappear before hatching, but the cells from which they are produced persist as a part of the ectoderm. The anterior and posterior ends are no longer in immediate contact, as in figure 71, but begin to overlap. The ends of the embryo

now take the position within the egg membrane of the least resistance to their further growth. Figure 74 shows the overlapping of the ends. The stomodaeum is completely formed and the annuli of the pharynx are visible. Figure 73 shows an unusual condition in the position of the ends. At this stage of development the animal often turns on its longitudinal axis, largely on account of the action of the muscles, and, instead of the convex side being ventral, it now becomes dorsal. This rotation on its longitudinal axis has no significance, as has been thought by previous investigators, in the later stages of development. The animal is extremely plastic and may assume any position or shape, as shown in figures 74 and 76. Figure 77 shows the completely developed animal at the time of emergence from the cocoon. The number of the segments is distinct. This peculiar form of growth within the cocoon is merely adaptive. Occasionally, when the cocoon is of an unusual size, the developing worm is less bent on itself.

A COMPARATIVE STUDY OF DIFFERENT FORMS

In following the cleavage cells of annelids, molluscs and polyclades, one is impressed with the striking resemblances in their different stages of development. If this marked similarity alone were a sufficient criterion for a basis of classification, some of the most widely separated forms, when considered from the standpoint of their early development, would be grouped as closely related species. How can such resemblances in development be explained in animals which are so unlike in their late stages of growth? Are they merely the result of such mechanical principles as surface tension, alternation of cleavage, and pressure, or is the nature and structure of the protoplasm the common cause? According to Driesch, 'the striking similarity' between the types of cleavage in annelids, molluscs and polyclades does not appear startling and is easy to understand, since cleavage is of no systematic worth. However, the more recent investigators on cell lineage, according to Heath, look upon the early cleavage stages as something more than a mere

manifestation of simple mechanical forces. Rather are the blastomeres the expression of the active intrinsic forces, which control development from the earliest stages unto the end. Gravity, surface tension, cohesion and pressure no doubt are effective, but not to the extent that they become the controlling or coördinating agents in development. The early cleavages are as important as those occurring in later life, and may even be considered more so. "Also the long continued resemblances which exist in the development of these different forms from the earliest segmentation of the eggs are as fundamental and deep seated as are the homologies existing in the adults."

The number of these resemblances in the annelids and molluscs is surprisingly great. In all forms accurately studied, the first three generations of ectomeres give rise to the entire ectoderm. The mesoblast arises at the fourth division of the posterior macromere D. The remaining members of this quartette and the macromeres produce the entoderm. The division and position of the cells up to the twenty-four or thirty-cell stage are identical in many different species. Beyond this point Wilson believes a divergence between the two classes ensues, and that development proceeds upon two entirely different lines. However, subsequent investigators have shown that the supposed differences are more superficial, and that the points of resemblances become more numerous and extend throughout longer periods of development. Lillie ('95) showed that points of resemblance existed in the lamellibranchs and the annelids, and that in both classes there is an essential similarity between the development of the 'first somatoblast.' In annelids this structure develops to a greater extent than in *Unio*, but the two have many points in common.

Mead ('97) and Conklin ('97) showed that the rosette series had the same origin and position in annelids and molluscs, and that in both it probably gave rise to the apical sense organ. According to Conklin, it also gave rise to the cerebral ganglia, while Mead considered this particular point doubtful. Furthermore, Mead in his annelid studies demonstrated that the same cells in five different annelids gave rise to the entoderm; that

the head kidney in *Amphritrite* and *Nereis* developed from the same cells. Conklin further states that the axial relation of all the blastomeres, with the possible exception of the macromeres, are the same in both the annelids and molluscs, and that the larval mesoblast in *Crepidula* and *Unio* arises from the same group of ectodermal cells.

Heath ('99) found that the prototroch in annelids and molluscs was homologous, and that the twenty-two to twenty-five cells concerned have exactly the same origin, direction of cleavage, and destiny. Also that the remainder of the first quartette, forming the head vesicle with its rosette series and molluscan cross cells or intermediate girdle cells, has in all probability, the same fate in both. He found many other resemblances and concludes:

Thus it is seen that not only in the origin and position of the various quartettes do resemblances appear, but that the early cleavage of these are in many cases cell for cell the same. In later stages close cell homologies cease, but the relation of the cell groups and their development in giving rise to larval or adult structures follow along much the same path. After passing these facts in review and considering the various structures in detail and modifications which they undergo, one fact presents itself with greatest clearness, that between *Ischnochiton* and the annelids the resemblances are more fundamental and closer than are the differences.

For a more direct comparative study of *Bdellodrilus* with the annelids and molluscs, special references will be made to *Clepsine* (Hirudinea), *Dinophilus* (Polychaete), and *Unio* (Lamelli-branch). In all these forms the first and second cleavages are meridional and divide the eggs into four unequal macromeres (text figs. 19-22). In *Dinophilus* C and D are approximately posterior and A and B are anterior. In the other three forms B is anterior, D posterior, C right and A left. In each case D is the largest cell; A, B and C are nearly equal; B is usually the smallest when variation occurs. The eight-cell stage has the same structure, and in all probability arises in the same manner in the four forms, the only apparent difference being the much greater relative size of the ectomeres in *Dinophilus* than in the three remaining forms. The first cleavage plane in *Bdellodrilus*

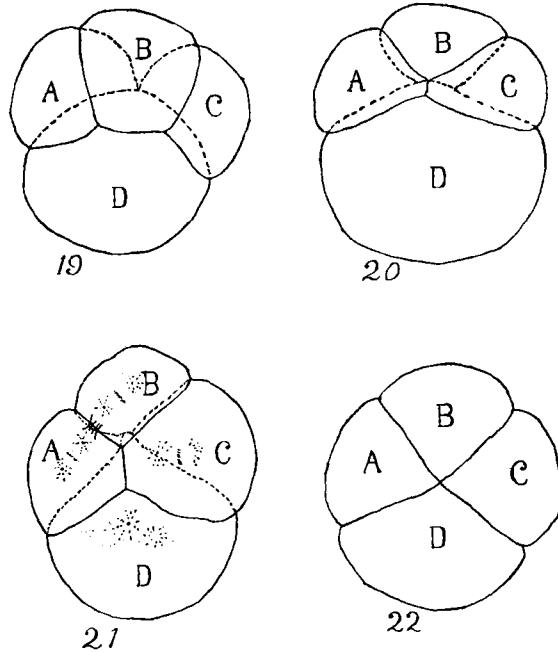


Fig. 19 Four-cell stage of *Unio*, upper pole (after Lillie).
 Fig. 20 Four-cell stage of *Bdellodrilus*, upper pole.
 Fig. 21 Four-cell stage of *Dinophilus*, upper pole (after Nelson).
 Fig. 22 Four-cell stage of *Clepsine*, upper pole (after Whitman).

occurs at nearly right angles, while in *Unio* and *Clepsine* it is inclined at an angle of about forty-five degrees to the sagittal plane of the future adult. In *Dinophilus* the direction of the first cleavage is in doubt. The second cleavage plane in *Unio*, *Clepsine* and *Bdellodrilus* occurs at an angle of about forty-five degrees to the sagittal axis. The origin of the ectoderm, the entoderm and the mesoderm is approximately the same in each form.

1. THE FIRST SOMATOBLAST

The first somatoblast in each instance is derived from the large posterior macromere D (text figs. 23-26). The cell d² (X) is extremely large and occupies a median posterior position. In *Clepsine* (Whitman) d² (X) is called the 'neuro-nephroblast.'

It divides into two, four and finally eight large cells called the teloblasts; the middle stratum of the germ bands is derived from them. These eight teloblasts are arranged into two groups of four cells each. Each group, which later is composed of four rows of cells, produces the middle stratum of the germ band on the corresponding side. The inner row of each band lies ultimately near the median ventral plane and gives rise to the corresponding half of the nervous system. The adjoining rows—'nephroblasts'—give rise to the nephridia. The derivatives of the outer row are still in doubt, but probably take part in the formation of the ectoderm.

In *Dinophilus* (Nelson) d^2 (X) is formed by a laeotropic division of the macromere D (text fig. 25); D is much smaller than X. Immediately after the formation of X, x^1 is budded off to the right at a low level. Next x^2 is budded off to the left at a higher level than x^1 ; x^3 is next formed by a dextrotropic division from the dorsal side, a little to the left. Next X divides equally and produces X and X, right and left. These two large cells correspond to the proteloblasts in *Bdellodrilus*. Finally X on either side divides equally, and produces the two teloblasts on each side of the median plane. These four cells, according to Nelson, correspond to the posterior teloblasts of *Nereis*. They also correspond to the neuroblasts and nephroblasts of *Bdellodrilus*. The division of X in *Dinophilus* and *Nereis* differs no more than do the corresponding divisions in *Nereis* and other annelids (*Amphitrite*, etc.). At the time of the closure of the blastopore in *Dinophilus*, the descendants of X are distributed dorsally and laterally to the posterior stem cells. In *Nereis* the main bulk of the descendants of X lay on the vegetal side of the stem cells.

In *Unio* (Lillie) the 'first somatoblast' X is formed by an unequal division of D (text fig. 24) in a median posterior position; x^1 is budded off from X, just behind C on the vegetal pole; next x^2 is budded off from X symmetrically with x^1 on the right side, just posterior to d^3 ; next x^3 is formed from X towards the apical pole, posterior to $d^{1,2}$; then x^4 is budded off from X anteriorly, towards the vegetal pole. This division of X does

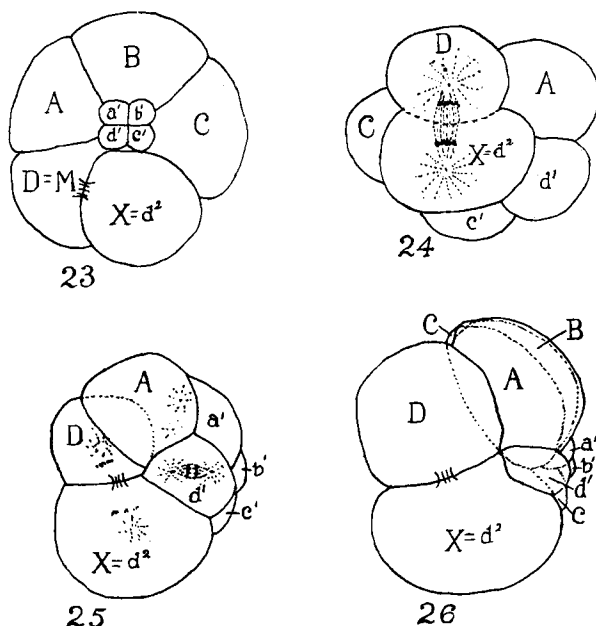


Fig. 23 Nine-cell stage of Clepsine, upper pole (after Whitman).

Fig. 24 Nine-cell stage of Unio from behind (after Lillie).

Fig. 25 Nine-cell stage of Dinophilus, left side (after Nelson).

Fig. 26 Nine-cell stage of Bdellodrilus, left side.

not occur in this manner in *Dinophilus*, *Bdellodrilus* or even in *Nereis*. The fourth division in the above three forms is equal and bilateral, while in *Unio* the fifth cleavage of X is the first bilateral division and forms X, X right and left. Next X, X on either side divides nearly equally and gives rise to the shell gland. These four cells might be regarded as the posterior teloblasts, which occur in other forms, as in *Nereis* and *Dinophilus*.

In *Bdellodrilus* X is formed by an equal division of the macromere D (text fig. 26), and takes a median posterior position. First x^1 is budded off from X to the right, posterior to C. Then x^2 is budded off to the left, symmetrical with x^1 and posterior to d^3 . Next x^3 is formed from the median dorsal anterior edge of X, between d^1 and c^1 . Now the first bilateral division of X takes place and forms the proteloblasts X, X, right and left. Each of the proteloblasts bud off x^4 , x^5 and x^6 respectively. At the

next division each proteloblast divides nearly equally, and gives rise to $X^{(1)}$, neuroblast, and $X^{(2)}$, nephroblast, on each side of the median axis of the embryo. Next each nephroblast divides nearly equally and produces $X^{(2)}$ and $X^{(3)}$. Now a very interesting thing happens; either $X^{(2)}$ or $X^{(3)}$ divides (but never both in the same egg) and produces the three nephroblasts on each side, which are designated as $X^{(2)}$, $X^{(3)}$ and $X^{(4)}$. In Clepsine only two of these teloblasts are concerned in the formation of the nephridia. The lateral teloblasts, as stated above, are probably ectodermal.

These four forms unquestionably show that there is a marked similarity in the cleavage of the 'first somatoblast,' not only in widely different individuals in the same group, but in individuals of widely separated groups. This comparison could be extended to other groups or forms, but the above will suffice for our purpose.

2. THE SECOND SOMATOBLAST

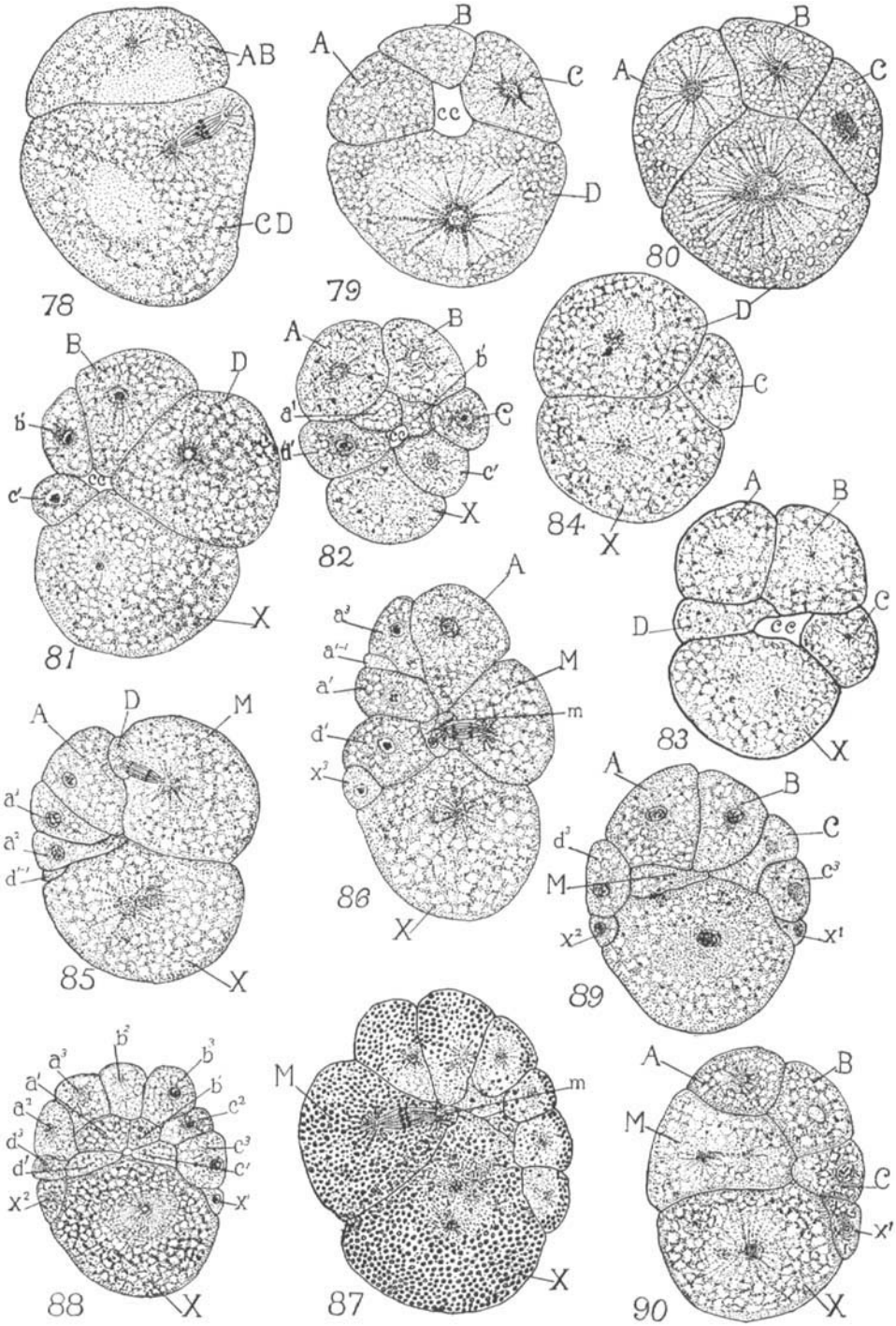
In Clepsine, the 'second somatoblast' has rather a unique origin. It is formed at about the twelve-cell stage. D, after the formation of X, becomes directly the 'second somatoblast' or M. (These cells are differently designated by Whitman; D is represented by x, X by x' and the mesoblasts by x and xy.) M divides nearly equally and produces the right and left mesoblasts, from which the inner stratum of the germ bands is formed.

In Bdellodrilus, M is formed by a very unequal division of the macromere D, at the twenty-four-cell stage (fig. 85). The larger cell or M is formed in front of X. It is inclined a little to the left of the median axis. The first division of M is equal, producing the mesoblasts, one on either side. These primary mesoblasts now bud off a number of small cells, directly beneath d^1 and c^1 (figs. 86-87). It is very difficult to make out the exact number of these small cells, since they are not visible externally. There are at least twelve formed, six on either side from each mesoblast. After this small group of secondary mesodermal cells are formed, the mesoblasts M, M, give rise to

PLATE 7

EXPLANATION OF FIGURES

- 78 Two-cell stage, horizontal section; CD dividing.
- 79 Four-cell stage; horizontal section taken above the center of the egg.
- 80 Same as the preceding, with plane of section below center.
- 81 Nine-cell stage, parasagittal section, to right of median plane.
- 82 Horizontal section of a nine-cell stage, taken four sections from the top.
Taken from embryo composed of 21 sections, each eight micra in thickness.
- 83 Same as the preceding; sixth section from top.
- 84 Same as figures 82 and 83; fifteenth section from top.
- 85 Parasagittal section of a twenty-four-cell stage; plane of section little to left of median axis. This figure shows the unequal division of the macromere D in the formation of the second somatoblast.
- 86 Thirty-three-cell stage. Parasagittal section to left of the median plane. Shows the formation of the secondary mesodermal cells (m cells) from the primary mesoblasts.
- 87 About the same stages as the preceding, to show the distribution of the yolk in different cells.
- 88 Horizontal section of an eighteen-cell stage, fourth section from top. Series composed of 20 sections, each eight micra in thickness.
- 89 Same as the preceding; seventh section from top.
- 90 Taken from series same as figure 88; eighth section from top.

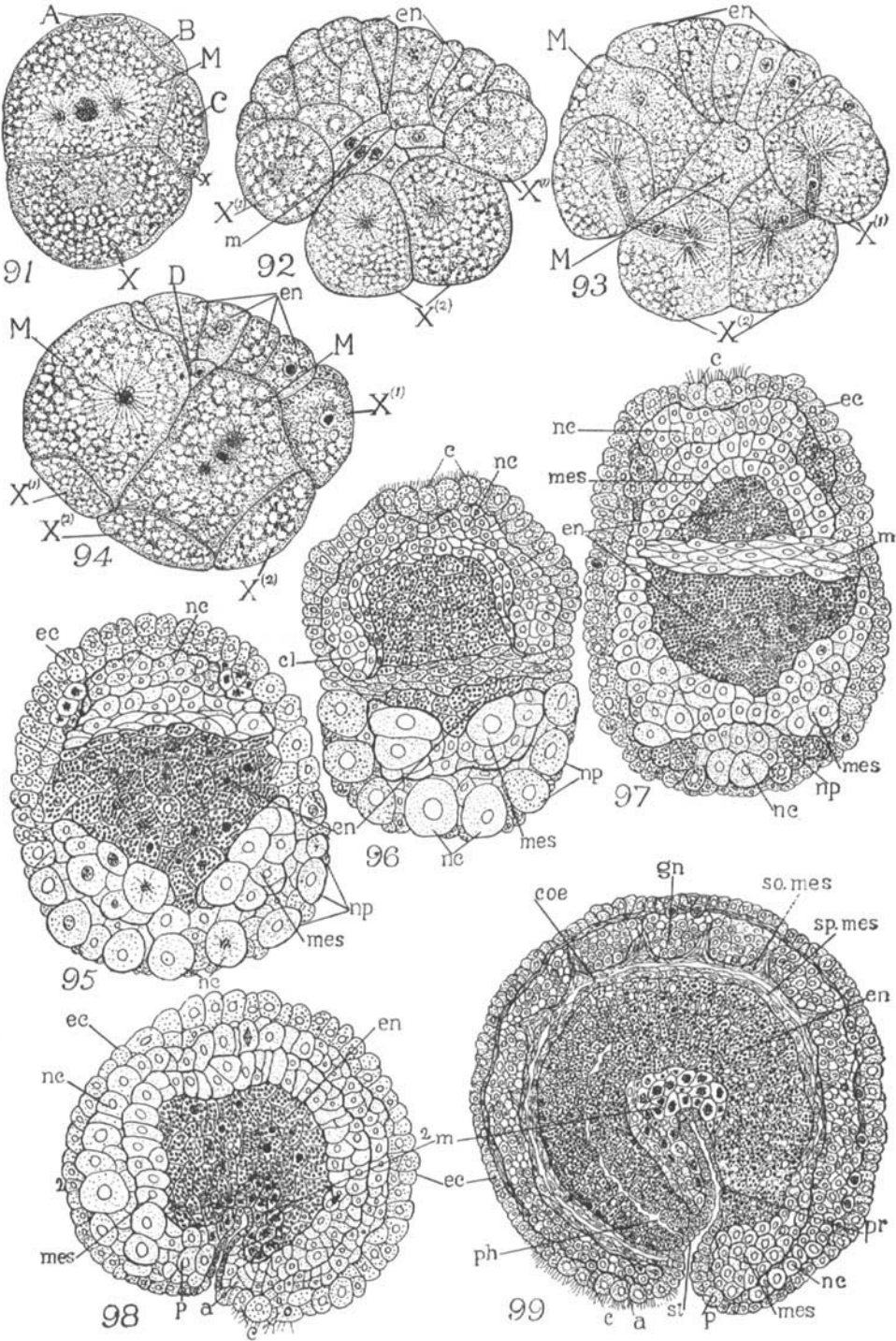


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PLATE 8

EXPLANATION OF FIGURES

- 91 Taken from the same series as figures 88 to 90; eighth section from the top.
- 92 Horizontal section of an eighteen-cell stage; sixth section from the top; shows the neuroblasts and nephroblasts.
- 93 Same as the preceding; tenth section from top; this figure shows the persistence of the cleavage spindles after the cell membranes are distinct.
- 94 Same as 92 and 93; fourteenth section from top; taken from a series of 20 section each eight micra thick. This figure shows the upper side of the macromere D wedged in between the other cells.
- 95 Transverse section of an embryo represented by figure 64; section taken at plane 2 — 2, or at a region corresponding to plane 2 — 2, of figure 98.
- 96 Transverse section of stage corresponding to figure 63; section taken at plane 2 — 2; figures 95 and 96 shows germ bands only partially covered by the ectoderm.
- 97 Transverse section of embryo represented in figure 69; section taken at level marked by line 2 — 2; here the germ bands are completely covered by the ectoderm.
- 98 Longitudinal section, near median line, of stage represented by figure 69.
- 99 Longitudinal section of an embryo represented by figure 71; plane of section near median line.



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the inner stratum of the germ bands. The plane of division in the formation of the primary mesoblast is at right angles to that of the secondary.

In *Dinophilus*, M is formed at the twenty-nine-cell stage, by an unequal division of D. M is much larger than D, as in *Bdellodrilus*, and is in front and below X, slightly to the left of the median plane. The division of M is now delayed until the seventy-two-cell stage, when two small cells are budded off anteriorly towards the vegetal pole, close to the line of junction of the two mesoblasts. At the next division two small cells are budded off, one on either side of the first pair. The following cleavages are teloblastic and produce the mesoblastic bands. The mesoblasts do not move into the cleavage cavity as in many other annelids, but remain on the surface until covered by the ectoderm.

In *Unio*, at the thirty-two-cell stage, M is formed by a very unequal division of the macromere D. The first division of M is equal and bilateral. Their position is immediately behind the entomeres. The next division of the two mesoblasts is very unequal, two small cells, m, m, are budded off at the posterior lip of the blastopore. Later the mesoblasts are included within the segmentation cavity, where they take up their final position behind the archenteron and give rise to the definitive mesoblastic germ bands with lateral teloblasts.

From the forms compared above it is very evident that there is a remarkable similarity with *Bdellodrilus*, not only in the early cleavage stages, but in the establishment of the germ bands as well. Thus cells having the same origin and lineage have the same final result in a wide series of forms (d⁴ the mesoblasts). Again, cells of unlike origin have a different fate (first and second somatoblasts); or cells of a different origin may have the same fate (d⁴ of annelids and the second and third generation of ectomeres in polyclades). Then cells of the same origin may have a different fate (a^{2.2} in *Unio* and *Bdellodrilus*). These contradictions, however, are far less striking than the resemblances. The 'first somatoblast' in each of the above four forms gives rise to the ventral plate and all or nearly all of the trunk ectoderm, while

the 'second somatoblast' produces the definitive mesodermal elements of the adult animal.

3. VARIATIONS IN THE METHOD OF MESODERMAL FORMATION

All annelids and molluscs which have been carefully studied show that the ectoderm arises from the three generations of ectomeres, the mesoderm from M and the entoderm from the remaining cells. There are, however, a few minor variations in forms like *Clepsine*, *Crepidula* and *Nereis*. In polyclades the mesoderm is directly associated with the ectomeres. The second and third generations of ectomeres, as in *Discocoelis*, produce the mesoblast, and the macromeres the entoblast. In molluscs and annelids the mesoderm is more closely associated with the macromere D. There is considerable variation in the cleavage of D in the formation of the 'second somatoblast;' in some forms D is given over entirely to the mesoderm; in other forms it shares equally, or in still others it contributes but little to the mesoderm.

Forms in which the mesoblast has two sources

(a) *Ecto-mesoblast* and (b) *coelo-mesoblast*. In *Thalassema*, Torrey distinguished between ecto-mesoblast, from the ectoderm and coelo-mesoblast from M. He states that the coelomesoblast is present in two bands, each consisting of five sub-equal cells. These are closely applied to the body wall and lie in the usual position on each side of the neural rows, but are more widely separated than in most annelids. The ecto-mesoblast on the other hand, derived from the first and second quartette of ectomeres, is present in great abundance and many of the cells have already undergone considerable differentiation in the formation of the muscles. He further states that the two mesoblast cells, M, M, are the last to sink in at gastrulation, instead of the first, as in the case where development is more direct (*Nereis* and *Amphitrite*). The two coelo-mesoblast bands of five cells each are shown to have the same origin and early history as the mesoblast bands in other annelids. The coelo-mesoblast, which is meagerly developed in the trochophore, is clearly correlated with

the long duration of its free swimming and almost stationary larval existence. In all forms where there is a trophophore stage of long duration, as is the case in all annelids with equal cleavage, the two coelo-mesoblast cells do not, in the early stages at least, bud like teloblasts. This is true in Hydroides, some species of Polygordius, in Thalassema and Podarke.

Many of the annelids and molluscs show that the so-called ecto-mesoblast (designated as larval mesoblast or as mesenchyme by some authors) arises from certain ectodermal cleavage cells of the second or third quartette of ectomeres and is entirely independent of the coelo-mesoblast. In Thalassema (Torrey) ten large ectodermal cells sink in from the ectoderm and give rise to all the mesenchyme. Three of these cells are from the a, c and d quadrants of the third quartette and seven from the first quartette of ectomeres. The most important source of mesenchyme in Thalassema is from the three cells of the third quartette ($3d^{2.2.2.1}$, $3c^{2.1.2.1}$, and $3a^{2.2.2}$). The first two sink into the cleavage cavity, just before gastrulation and at first lie close to the two coelo-mesoblast cells. They soon migrate laterally and bud off simultaneously small cells toward the M cells. They divide like teloblasts, but in the reverse order to the ordinary direction. So close is the connection of these cells with the coelo-mesoblast that one would be certainly led to think that they formed a part of these bands, unless their cytogeny had been carefully followed. Similar conditions are described by Treadwell in Podarke obscura. The progeny of these two cells form almost the entire mesenchyme of the post throchal region and become differentiated for the most part into muscles of the digestive tract. The progeny of the other ectodermal cell migrates to the mid-ventral line. The ecto-mesoblast cells of the first quartette sink into the primary body cavity later than those of the third; their exact cell lineage has not been traced, but probably give rise to gut musculature. This mesoblast has commonly been considered as purely larval and transitory. In some instances it is possible to determine its exact origin, but in many others merely the general region from which it arises.

During the last thirty years embryologists have differed in their conception of the origin of the mesoderm and of its phylogenetic significance. Hatschek ('78) was among the first to distinguish between mesenchyme and mesoderm, but held, after studying the embryology of *Polygordius*, *Echiurus* and *Eupomatus*, that these two morphologically different mesoblasts, arise from a common foundation. This same view was later put forth by the Hertwigs ('81) in their 'Coelomtheorie,' which, according to Meyer, has formed the foundation of all later work on mesoderm. Roule ('89 and '94), Burger ('91 and '94), Fraipont ('88), Häcker ('95), and others, have described the mesoblast as having a single origin. On the other hand, those who have studied the embryology of annelids and molluscs, consider the origin of the mesenchyme distinct from that of the mesoblast or coelo-mesoblast. This later view was first described by Kleinenberg ('78 and '86), and later by Whitman ('87), by Berg ('90), by Schimkewitsch ('94), by Meyer ('01), by Torrey ('03), and others.

A larval mesoblast was first described by Lillie ('95) in *Unio*. It arises asymmetrically from the derivatives of $a^{2.2}$ and later migrates into the segmentation cavity, where it divides equally and becomes symmetrically arranged on either side of the mid-line. The derivatives of these two cells become metamorphosed into 'myocytes' and larval adductor muscles, which are functional only during larval life.

Treadwell ('97) regards both mesenchyme and mesoderm as morphologically the same tissue, the apparent difference in their mode of origin being of no significance. And, further, Wilson regards the larval mesoblast (ecto-mesoblast, because of its origin from the ectoderm) as a distinct tissue from that of the definitive mesoblast or ento-mesoblast, and states that it is homologous with the mesenchyme of the turbellarian ancestors of the annelids, while the mesoblast from which the adult structures arise is phylogenetically younger and is represented prophetically in the ontogeny of such a form as *Discocoelis* (polyclade) by the peculiar lateral division of M, and states that the ecto-mesoblast and endo-mesoblast are phylogenetically of

different origin. This same point was previously urged by Meyer.

The condition, however, found by Wilson in *Nereis* and *Lumbricus* does not indicate a hard and fast distinction between the two kinds of mesoblast. In *Nereis*, cells from the anterior end of the germ bands separate early and pass forward into the segmentation cavity where they give rise to the larval musculature. This corresponds exactly in structure and function with the larval mesoblast of *Unio* (Lillie) and *Podarke* (Treadwell). In *Lumbricus* the origin of the mesenchyme is similar to that in *Nereis*. These two kinds of larval mesenchyme have also been described by Eisig ('98) as occurring in the same individual (*Capitella*, a polychaete annelid).

In *Thalassema* and *Podarke* the larval mesenchyme arises directly from the ectoderm, while in *Nereis* and *Lumbricus* it arises from the anterior ends of the mesoblast bands. According to Treadwell, no one has yet proven that no 'mesenchyme' arises from the germ bands in cases where a larval mesenchyme exists. If we accept Wilson's view that mesenchyme and mesoderm are different phylogenetically, we must regard the two sets of larval mesenchyme which have the same structure and function, as non-homologous, or we must regard the mesenchyme and mesoderm as morphologically the same tissue and the difference in their modes of origin as of no significance. Furthermore, Wilson has pointed out that the trochophore, as it occurs at present, is more than a mere ancestral stage, for it contains in a concentrated form the anlage of the whole future body. According to Mead, the ectoderm behind the first septum in *Amphitrite* arises from a group of cells which surround the procotodaeum of the young trochophore and are descended from a single cell, the 'first somatoblast.' The same is true of other trochophore forms. There is no need to assume phylogenetically a new formation of ectoderm for the body as distinct from that of the head. Neither is there any necessity to assume a distinct phylogenetic origin of the larval mesoblast from that of the mesoderm.

It is evident that in *Nereis* and *Lumbricus*, both kinds of mesoblast have the same origin, and simply shows a more complete concentration of the mesoderm than in *Thalassema* and *Podarke*, where the mesenchyme is formed direct from the ectoblast. The mesoblast cells collected at the posterior end of the trochophore, which are derived from M, represent the mesoderm of the body. It is morphologically continuous with that of the head, as in *Nereis*, and is concentrated at this point to provide for the elongation as new segments are formed. The difference in the concentration of the mesodermal elements, as to whether they have a single or double origin in no way interferes, as already pointed out, with the morphological unity of the tissue, and as to the source of its origin, whether from the ectoderm or from the endoderm phylogenetically, we are not able to say (Treadwell).

Meyer ('01) in his study of the phylogenetic significance of the two kinds of mesoblast, gave a view directly opposed to that expressed by Treadwell. After an exhaustive review of the whole mesodermal question, he concludes that the great mass of evidence, both embryological and anatomical, points to the conclusion that in annelids, at least, there are two entirely distinct forms of mesoblast, the ecto-mesoblast (primary mesoblast) and the coelo-mesoblast (secondary mesoblast). Of these two he considers the primary mesoblast to be phylogenetically the older, and as a rule, to be derived from the ectoderm. The coelo-mesoblast, on the other hand, is regarded as a later formation, which has originated from the gonad cells.

The formation of the ecto-mesoblast in annelids and molluscs from certain cells of the first, second, third and fourth generation of micromeres, can well be regarded as vestiges or survivals of the process which occurs in all four cells of the second and third quartettes of certain polyclads. The origin of the ecto-mesoblast from the ectoderm in annelids and molluscs, partially bridges the gap between them and the polyclads. In order to have a complete homology of the mesoderm in the polyclads, annelids and molluscs, it is necessary to find a polyclad in which there is a double origin of the mesoderm. The development of the polyclad *Leptoplana* (Wilson) is the nearest representative to complete the homology. In *Leptoplana* only a part of the four quadrants of the second quartette contributes to the entire mesoderm, the typical condition in polyclads being that all of

the second and third quartette is mesodermal. The behavior of d^4 in the polyclad *Discocoelis*, is also very suggestive. Here the division of d^4 is equal and gives rise to two symmetrically placed cells at the posterior end of the embryo, comparable to the primary mesoblasts found in annelids and molluscs. Some investigators have even suggested that these two posterior cells in the polyclads may give rise to the mesoblast bands in this particular group. This latter point, however, has never been verified.

Table 3 shows that the first, second, third and fourth generation of micromeres, in a series of widely separated forms, may contribute to the formation of the mesoderm.

TABLE 3

	1ST GEN.	2D GEN.	3D GEN.	4TH GEN.
Annelids:				
Thalassema	part of a, b and c quad's	none	1 cell each of a, b and c quadrants	d^4 part mes.
Bdellodrilus	none	none	none	d^4 all mes.
Molluscs:				
Unio	none	a^{2-2} (larval)	none	d^4 all mes.
Crepidula	none	a^2, b^2, c^2	none	d^4 part mes.
Physa	none	none	b^3, c^3	d^4 part mes.
Podarke	none	none	$a^{3-2-2-2}$ $c^{3-2-1-2}$ $d^{3-2-2-2}$	d^4 part mes.
Polyclads:				
Discocoelis	none	all mes.	all mes.	none
Leptoplana	none	part of each quadrant	none	none

In case of the ecto-mesoblast a complete series could be arranged, in which all of the cells of certain quartettes contribute to the mesoblast, to those forms in which only a small part of certain quartettes is mesoblastic. Again in case of the coelo-mesoblast we have a wide range of variation, in which all of d^4 is mesodermal, to those in which only a small part of d^4 is mesodermal. As far as records show, *Capitella* is the only annelid in which

d^4 does not contribute to the coelo-mesoblast. Here then we have quite a unique series ranging from those forms where the entire mesoderm is ectodermal in origin, or where it is both ectodermal and entodermal, to those where it is entirely entodermal. From the above it is evident that the entire mesoblast of polyclads is derived from the ectomeres, and, if homologies be any significance, it would be fair to conclude that this mesoblast is represented by the ecto-mesoblast in the annelids and the molluscs.

The origin and development of the mesoblast in *Bdellodrilus* contributes but little to the phylogenetic significance of the primary and secondary mesoblast. Here, beyond question, when considered from the standpoint of their origin, they are one and the same tissue. Both are formed directly from the primary mesoblasts. The secondary mesoblast cells are budded off from the two primary mesoblasts before the germ bands begin their development. Similar conditions are found in other forms, as in *Lumbricus*; here, however, the secondary mesoblast is formed later directly from the anterior ends of the mesoblastic germ bands. The difference is only in the point of time in their formation. In *Bdellodrilus* there can be no hard and fast distinction made between the two kinds of mesoblast. Both must be considered as the same tissue phylogenetically.

4. VARIATIONS IN THE SOURCE OF THE ENTODERM

In general, as stated above, the ectoderm originates from the three generations of ectomeres, the mesoderm from d^4 , and the entoderm from the remaining cells. The origin of the three germ layers, however, depart somewhat from the above rule in some of the annelids and molluscs. In some species cells from the first, second and third quartettes contribute to the mesoderm; in others d^4 gives rise to entoderm as well as mesoderm. In all annelids and molluscs, A, B and C, after the formation of the first three sets of ectomeres, are distinctly entodermal. The macromere D, after the formation of d^4 , is likewise entodermal. In some forms D is the same size as its fellows, in others

it is reduced until it is little more than a mere nucleus, while in others it has completely disappeared as an entomere, and is given over entirely to the formation of mesoderm.

In annelids, in a gradually decreasing series, D (*Nereis*) is the same size as the entoblast cells A, B and C. In *Dinophilus* it is about half the size of these cells. In *Bdellodrilus* D is little more than a mere nucleus; while in *Clepsine* D is given over entirely to the formation of the mesoderm. In molluscs it is a fairly common condition to find the entoblast cell D smaller than A, B and C, or even greatly reduced. In *Crepidula* it is very little reduced; in *Unio* it is more than half reduced, while in *Ischnochiton*, D is often little more than a mere nucleus. The second somatoblast, M, may contribute to the formation of entoderm as well as mesoderm. In forms like *Crepidula* M is mostly entodermal. In *Fiona* (Casteel) the division of M in the formation of the entoderm is very similar to that in *Crepidula*. In *Unio* two small cells are budded off from M, which lie near the entoderm, and are probably concerned in the formation of that layer.

In some of the annelids the primary mesoblasts bud off small cells directly posterior to the macromeres. This number varies; in *Nereis* there are six to ten, and in *Aricia* there are but two. In many of the other annelids and also in some of the molluscs, where their cell lineage has been traced, it is found that these small cells give rise to entoderm. There are at least sixteen to twenty species of annelids and molluscs in which similar cells have been found (small cells from the primary mesoblasts.) Diverse accounts of their behavior and fate have been given by different investigators. Table 4 shows the fate of these small cells in a few of the annelids and molluscs.

In the mollusc *Alpysia*, according to Carazzi, each primary mesoblast buds off four small cells. Three of these are mesoblastic and one is entoblastic. This interesting condition might be considered as a transitional form or as a connecting link between those forms in which these small cells are entirely mesodermal and those in which they are entodermal. Again we could arrange a series of annelids and molluscs in which at one extreme

TABLE 4

ENTODERM	MESODERM	NOT CERTAIN
Crepidula	Amphitrite	Dreissensia
Nereis	Arenicola	Patella
Podarke	Umbrella	Spio
Thalassema	Planorbis	Serpulorbis
Fiona	Unio?	Cyclas
Ischnochiton	Limax	Aricia
Physa fontanalis		
Physa hyponurum		
Aplysia		

the entoblast derived from M is greater in amount than the mesoderm, as found in *Crepidula*, and at the other extreme, where but two rudimentary cells of M are entoblastic, as in *Aricia*.

According to Wilson, a series of this nature may indicate a gradual elimination of the entodermal element from the macromere D of the fourth quartette, and finally its complete transformation into the mesoblast. Kovalevksy ('71) suggested that this transformation shows quite forcibly that the mesoblast pole cells are to be regarded, phylogenetically, as derivatives of the archenteron, because of their close association with the posterior entoblast cell, D.

The primary entoblasts, A, B, C and D, undergo but little change until late development in those forms which possess a larval stage, and may remain in this condition until after the trochophore is developed, or until after the blastopore is closed. In those individuals with a fetal type of development, they often remain distinct until after the germ bands are completely formed, as in *Clepsine*.

GENERAL ADAPTATION AND INTERPRETATION OF CLEAVAGE

The cleavage of eggs of widely separated forms exhibit unique resemblances. At certain stages of development these resemblances exceed their differences. Is the persistence of these features due to the influence of ancestral inheritance, or are they due more to the adaptive conditions of their environment, to

meet the highest need of the developing animal? It has been demonstrated, again and again, in annelids, molluscs and even in polyclads, that homologous cells of like generations give rise to like parts in the developing embryo and the adult. The occurrence of these conditions in such widely separated forms furnishes a very interesting and important phase in the study of cell lineage. The tendency has been rather to emphasize these resemblances, than to give special stress to the exact conditions which occur in any one species in its different stages of development. It is true, however, that the general form of cleavage may be inherited from a long series of ancestors, probably from some of the Turbellarian worms. But the problem of more direct importance in any one group is, why such variation in the size, form, direction and rate of cleavage?

1. IN THE CLEAVAGE OF BDELLODRILUS

In *Bdellodrilus* we have a determinate type of cleavage, i.e., the fetal as well as the adult structures can be shown to have a definite or direct cell lineage, and can be traced back to the unsegmented egg. The structure of the ovum is quite homogeneous, and at the time of maturation, the egg can be definitely oriented as to the future axis of the body. Before the first cleavage is complete, the parts of the ovum which give rise to the different germ layers can be traced or ascertained with a fair degree of accuracy, i.e., definitely localized parts which give rise to definite organs or structures.

"Adaptation in cleavage can manifest itself only in three possible ways or modes of cleavage variation, which are, as has been pointed out by Lillie, Mead, Conklin and others, the following: first differences in the rate of cleavage; second differences in the size; and third, differences in the direction of cleavage."

The general plan of cleavage in *Bdellodrilus*, is similar to that of other forms. The ectoderm is derived from the four basal cells, by three successive horizontally formed cleavages. The mesoderm from a fourth cleavage of the posterior macromere D and the entoderm from the remaining cells. The first cleavage

in *Bdellodrilus* is meridional and very unequal. In the two-cell stage the larger cell is posterior and the smaller cell anterior. The larger cell divides first and very unequally, while the smaller cell divides nearly equal (text figs. 1-3 and fig. 5). In the four-cell stage D is posterior, C right, A left and B anterior, inclined a little to the right. Thus it is very evident that the four-cell stage illustrates a difference in the rate of cleavage, a difference in the size of the cells, and a difference in the direction of the cleavage. The significance of these variations may be emphasized as follows:

a. Difference in the rate of cleavage of cells

If we compare a thirty-two-cell stage of *Bdellodrilus* with other forms or perhaps, better, with an ideal ovum, in which there is a uniform rate of cleavage in the formation of the cleavage cells, a uniform size and a uniform direction of cleavage, a distinct variation occurs as shown in table 5.

TABLE 5

	CREPIDULA	IDEAL OVUM	NEREIS	BDELLO- DRILUS
First generation of ectomeres.....	12	16	16	8
Second generation of ectomeres.....	9	8	8	11
Third generation of ectomeres.....	5	4	4	4
Mesoblast.....	2	0	0	2
Entoblast.....	4	4	4	7
	—	—	—	—
	32	32	32	32

It is evident that in the first generation of ectomeres *Bdellodrilus* departs very far from the ideal condition. The first generation contains eight cells instead of sixteen. This means that the cells have divided more slowly than in the ideal ovum. In an ideal ovum these cells form the prototroch and the entire region in front of it, with the apical plate in the center. In *Bdellodrilus*, this region is degenerate and no trace of the apical plate appears. This indicates an adaptive modification—eight cells instead of sixteen—due to a degenerate frontal region.

In the second generation of ectomeres, the ideal number is

eight, while in *Bdellodrilus* it is eleven. This increase above the ideal is due entirely to the rapid succeeding divisions of one cell— d^2 , the first somatoblast. The other cells of the quartette have not divided, while d^2 has given rise to three new cells. Does the behavior of d^2 suggest any significance, or is it adaptive? From d^2 the ectoderm of the trunk region, the nephridia and the entire nervous system is derived; d^2 is not only the largest but the most actively dividing cell of the entire embryo; hence its rate of cleavage is well adapted to its resulting formations.

The number of ectomeres in the third generation is the same as in an ideal ovum; d^3 however is often formed before a^2 or b^2 of the second generation. This interesting phenomenon is due to the tendency of the basal cell, D, and its derivatives to divide more rapidly than those of A, B or C. The differences in the rate of cleavage in the first, second and third generation of ectomeres, no doubt possess prospective significance, looking forward to the definitive parts. This may fairly be called adaptation in the rate of cleavage.

In *Bdellodrilus* the more rapidly dividing cells do not form the first functioning parts. The cells of the first quartette are the first to function, in the production of cilia for the movement of the embryo. The variation in the rate of cleavage is not due to the varying conditions of the media, or the dividing ovum would be uniformly affected, as a whole. Nor is it due to the size of the individual cells, as the largest cells divide more rapidly. At the thirty-two-cell stage the ideal ovum contains four entodermal cells, while in *Bdellodrilus* there are seven. Here the cleavage is carried to the end without any resting stage of the four basal cells. This is due to the fact that the larva develops very rapidly and the entodermal cells must keep pace with the rapid development in order to reach their final position, just where they are needed.

b. Variation in the size of cells

The relative sizes of the cells in the early cleavage of the eggs of *Bdellodrilus* are adapted to the later developing parts.

The largest cell at the four-cell stage is D. Its first division is very unequal, and the smaller cell is less than one-tenth the size of the larger. It is the first ectomere of the first generation formed. The second division, in most instances, is equal; when unequal, the largest cell passes into the upper product, and forms d^2 , the first somatoblast. The third division is unequal, and d^3 , the smaller product, is again uppermost; and finally, the fourth division is very unequal and only a small portion remains as the macromere D. The greater bulk, d^4 , becomes the second somatoblast. In each of the above instances the larger cells form a large part of the embryo and the adult, while the smaller cells, in every instance, form a very insignificant portion. The unequal division in each instance is evidently adaptive, for the great bulk of the material passes into the two somatoblasts and gives rise to the muscular, nervous and excretory systems.

c. Variation in the direction of cleavage

Here only some of the special cleavages will be emphasized. The first division of the second somatoblast is equal, and each part forms equal parts of the mesoderm. Next, each primary mesoblast buds off five or six small cells beneath the first quartette of ectomeres. These small cells remain quiescent for a considerable period and later give rise to the dorsal mesoderm. Immediately after these small cells are formed, the mesoblast bands are begun by a forward proliferation of cells from the anterior face of the primary mesoblasts. The plane of division is at right angles to that of the small cells. These bands extend forward between the ectoderm and the entoderm, and at the same time the entodermal cells extend posteriorly between the mesoblast bands and the group of small cells, thus separating the primary and secondary mesoderm. Here the direction of the cleavages place the cells where they are later used in the formation of some special part, adapted for that particular region.

The first somatoblast buds off x^1 to the right, x^2 to the left, and x^3 median dorsal anterior. X now divides equally and

each proteloblast buds off a small cell, x^4 , one to the right of x^1 and the other to the left of x^2 . Again each proteloblast buds off a small cell, x^5 , on either side of x^3 . At the next division each buds off a small cell, x^6 , on the ventral anterior edge. Later, x^7 is budded off from each neuroblast on the ventral side. These small cells give rise to the trunk ectoderm and the larger cells to the nephridia and nervous system. Here again the cells are formed just where they are needed; the smaller on the exterior or outer surface while the larger remain within.

2. ADAPTATION IN THE CLEAVAGE OF OTHER FORMS

In following the variation of cleavage cells in annelids and molluscs, special cells can be arranged in a complete series, from those of an almost insignificant size to an extremely large cell. In following these variations, step by step, we can not fail to be convinced that these variations are adaptive to the future needs and habits of the larva and of the adult animal.

In forms with equal cleavage, the first somatoblast gives rise to the ectoderm of the trunk region. In *Polydorus*, *Podarke*, *Hydroides*, *Eupomatus* and others with equal cleavage, d^2 is the same in size as the cells of the other quadrants. Equal cleavage has been offered by some as due to a lack of differentiation in the early stages but in such forms as *Podarke* with equally cleavage, very early differentiation occurs, and the prominence of these early functioning parts varies according to the size of the initial cell from which they are formed.

In forms with unequal cleavage, the first somatoblast differs in size from the remaining members of the same quartette. Beginning with *Amphitrite*, the relative size of d^2 increases successively in *Chaetopterus*, *Arenicola*, *Nereis*, *Capitella*, *Aricia*, *Spio*, *Clepsine* and *Bdellodrilus*. Those forms with equal cleavage pass through a distinct trochophore stage and are characterized by an almost equatorial prototroch, a very large exumbrella, and with a very slow trunk development. In those with unequal cleavage, especially in the second generation of ectomeres, there is a gradual decrease in the prominence of the

trochophore to its approximate or complete disappearance; on the other hand, there is a gradual acceleration in the time of the trunk development, varying according to the increase in the relative size of the first somatoblast or X. Treadwell states that the extra amount of material stored in the macromere D is in some way related to the amount of somatic and mesoblastic material needed in the future organism. This statement is true of the condition that occurs in such annelids as *Bdellodrilus* and *Clepsine*.

GENERAL SUMMARY

The undivided egg of *Bdellodrilus philadelphicus* is nearly oval. Its median longitudinal axis through the region of the polar bodies corresponds to the median axis of the future adult. The polar bodies occupy the region which later becomes the anterior end of the embryo.

The first cleavage plane is nearly at right angles to the median axis of the resulting individual, and divides the egg into two very unequal parts. The second cleavage occurs at an angle of about forty-five degrees to the first. It divides the smaller cell nearly equally and the larger cell very unequally; the larger cell divides first. In a four-celled embryo the large cell D is posterior, B is anterior, inclined a little to the right; A left and, C right.

The ectoderm is separated from the four macromeres by a series of three oblique cleavages. The first generation of ectomeres is formed in a dextrotropic direction. The second generation laeotropically and the third in a dextrotropic fashion.

In the fourth generation of micromeres, d^4 is mesoblastic. The other cells of the fourth quartette, together with the four macromeres, form the entoderm. The cleavage of the entodermal cells is carried to the end without delay, in the formation of the digestive tract, and the interior of the embryo becomes a solid mass of entodermal cleavage cells, which later become differentiated into the epithelial portion of the alimentary canal. As the core of entodermal cells grows posteriorly,

it separates the primary and secondary mesoderm. The extreme ends of the digestive tract are ectodermal. Among other annelids and also in molluscs, so far as is known, the entodermal cells are not broken up into cells but enter directly into the formation of the digestive tract.

The posterior cell, d^2 (X), of the second generation of ectomeres is the largest cell of the segmenting ovum. The derivatives of X are symmetrically placed with reference to the median plane of the future individual. The large cell, X, gives rise to the trunk ectoderm, the nervous and the excretory systems. The nervous system is derived from the two neuroblasts. The brain is formed from the extreme anterior end of the neural rows.

The largest cell, d^4 (M), of the fourth generation of micromeres, gives rise to the entire mesoderm. It is the first cell to divide in a bilaterally symmetrical manner. The primary mesoblast cells, M, M, bud off five or six small cells each, beneath the first quartette of ectomeres, which give rise to the secondary mesoderm on the dorsal side of the embryo. Immediately after these small cells are budded off, the primary mesoblasts, by a teloblastic proliferation of cells, produce the mesoblast bands.

The embryo increases but little in bulk before the germ bands are formed. The embryo as a whole, during its early stages of development, is extremely plastic and may vary considerably in its transverse and longitudinal axes. The developing embryo is completely turned on itself, and the anterior and posterior ends are in immediate contact. The outer surface is ventral and the turned in portion is dorsal. This peculiarity of development is foreshadowed in the position taken by the early cleavage cells.

At the beginning of the germ-band formation, the embryo begins to rotate on its transverse axis. This movement is due to the action of cilia, which are produced by the ectodermal cells on the median ventro-anterior end of the embryo; the rotation alternates.

As growth continues within the cocoon, the ends of the embryo soon begin to overlap. The embryo may assume almost

any position in the cocoon during its later stages of development. The embryo is completely developed before emergence; the trochophore stage is completely suppressed; the gastrulation is of the epibolic type.

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