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CONTENTS.

FIRST LECTURE.

THE STRUCTURE OF PROTOPLASM.[1]

EDMUND B. WILSON.

It would be superfluous to dwell in this place on the deep and enduring interest that attaches to the microscopical study of protoplasm. Since the time when the studies of Cohn and Schultze led to the general recognition of protoplasm as the material substratum of vital activity, — a conclusion so eloquently set forth by Huxley in his celebrated essay on the physical basis of life, — this interest has continually increased, as we have come to see even more clearly that all biological phenomena are directly or indirectly traceable to the effects of protoplasmic activity, for we have thus been impelled to seek for an understanding of that activity in the morphological structure of protoplasm, as revealed by the microscope. It is small wonder that to this quest some of the ablest of modern biologists have devoted their best energies. And yet, if we take account of the actual knowledge gained, we cannot repress a certain sense of disappointment, partly that microscopical research should have fallen so far short of giving the insight for which we had hoped, but still more because of the failure of the best observers to reach any unanimity in the interpretation of what is actually visible under the microscope. In any consideration of the general subject, therefore, it is well to keep clearly in view the fact that such disagreement exists, and that

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[1] A more adequately illustrated special paper on this subject, containing more specific references to the literature, is now in press. It should be borne in mind that such delicate textures as those seen in the protoplasm of living cells cannot be properly illustrated by black and white figures. The accompanying text figures, though copied as accurately as possible from the original drawings, are of necessity relatively rude and schematic.

we are not yet in a position to justify any very certain or far-reaching conclusions.

I would like, at the outset, to express the opinion that, if we except certain highly specialized structures, the hope of finding in visible protoplasmic structure any approach to an understanding of its physiological activity is growing more, instead of less, remote, and is giving way to a conviction that the way of progress lies rather in an appeal to the ultra-microscopical protoplasmic organization and to the chemical processes through which this is expressed. Nevertheless, it is of very great importance to arrive at definite conclusions regarding the visible morphology of protoplasm, not only because of its intimate connection with all the problems of cell-morphology, but also in order to find the right framework, as it were, for our physiological conceptions, and thus to gain suggestion for further physiological and chemical inquiry. And this must be my excuse for reviewing a subject which is still so largely obscured by doubt, and of which the outcome gives, after all, so little satisfaction.

It is especially important in this field of biological inquiry to distinguish clearly between theory and observed fact; for theories of protoplasmic structure have always far outrun the actual achievements of observation. From the time of Brücke (one of the first to insist that protoplasm must possess a far more complicated organization than that visible under the microscope), speculation has gone steadily forward, to reach, perhaps, its most elaborate expression in Weismann's interesting, but unconvincing, work on the germ-plasm — an elaborate speculative system built out of hypotheses which for the most part float in the air without visible means of support. We need not consider this side of the subject *in extenso*, but I will ask attention, for a moment, to what is the most characteristic and, to the morphologist, the most interesting point in these speculations, namely, the doctrine of genetic continuity as applied to the corpuscular, or micellar, theory of protoplasm. We are all familiar with the successive steps by which that doctrine gradually developed. Harvey's celebrated formula, *ex ovo omnia*, — or, as usually quoted, *omne vivum ex ovo*, — took with Redi the far more philosophical form, *omne vivum e vivo*, thus expressing a truth which forms the very foundation of all biological teaching at the present day. The development of the cell-theory, long afterwards, enabled Virchow to pronounce the more specific aphorism, *omnis cellula e cellula* (1855), — a statement involving the highly interesting conclusion that protoplasm is never formed *de novo*, but always arises from or through the activity of preexisting protoplasm differentiated into the form of a cell. Still later a like conclusion was reached with respect to at least one of the structural components of the cell, namely, the nucleus, and the work especially of Flemming and Strasburger justified the saying, *omnis nucleus e nucleo*. Not long afterwards, the researches of Schmitz, Schimper, and others showed that in plant cells some, if not all, forms of plastids (for example, the chlorophyll-bodies) likewise arise by the division of preëxisting bodies of the same kind. Thus the law of genetic continuity was gradually extended downwards from the grosser and more obvious characters of the organism into the finer details of its structural elements. Genetic continuity, the origin of like from like, may now safely be regarded as a demonstrated fact in the case of all existing organisms and of all cells; it hardly falls short of the same degree of certainty as applied to the nucleus; it is probable in the case of various forms of plastids in plant cells; while the centrosome is now being weighed in the balance with the evidence for the moment apparently accumulating on the negative side.

Up to this point we have been dealing with matters of observed fact. The next and final step was, however, taken in the region of pure speculation, which had in the mean time been at work building upwards from hypotheses regarding the basic composition of protoplasm. Brücke's suggestion, that the cell might be a congeries of bodies more elementary than itself, found a much fuller expression in Herbert Spencer's theory of physiological units; but it was Darwin's theory of pangenesis that laid the real basis for what followed in the works of De Vries, Wiesner, Weismann, and Hertwig. The common feature in all these later views is the conception of protoplasm, not as a homogeneous substance or mixture of substances, but as made up of a host of elementary ultra-microscopical corpuscles ("pangens," "biophores," etc.), specifically different, capable of assimilation, growth, and multiplication, and arising by division of preëxisting bodies of like kind. Developed as a purely theoretical hypothesis, and within somewhat narrower limits, by Darwin, this conception was expanded, and brought into more direct relation with observed fact, especially by De Vries and Wiesner, who showed how the assumption of such elementary self-propagating corpuscles at the basis of living matter enabled us to bring all the observed phenomena of genetic continuity under a common point of view. The fundamental hypothesis itself — *i.e.*, the genetic continuity of the ultimate morphological units — has, however, always remained, and still remains, a pure assumption, incapable of direct proof or disproof; for, with the exception of Altmann and a few of his followers, all are agreed that such elementary corpuscles, if they exist, must lie beyond the limits of microscopical vision. Altmann, however, has sought to identify the elementary units, or "bioblasts," with the visible protoplasmic granules; and, in his writings, the series of Latin aphorisms initiated by Redi culminates in the saying, *omne granulum e granulo* (!), but this conclusion has not been taken very seriously by most other investigators.

I have given this very brief sketch of the theoretical side of the question merely as an introduction, and shall dwell no farther on it at this point, since my main purpose is to ask attention to the visible, as opposed to the hypothetical invisible, structure of protoplasm. A subject so vast, displaying so great a conflict of opinion, must be very briefly treated within the limits of a single lecture; and I shall, therefore, confine the discussion in the main to the protoplasm of the echinoderm-egg, which is accessible to every one, has been made a classical object through the studies of such leaders of research as Flemming, Bütschli, and Hertwig, and illustrates as clearly, perhaps, as any other the various interpretations of protoplasmic structure that have been given.

In thin sections of well-preserved material, the protoplasm of a star-fish or sea-urchin egg gives the appearance, under a high power, of a fine meshwork or framework composed of innumerable minute granules, or *microsomes*, suspended in a clearer, less deeply staining, continuous substance (Figs. 1, *a*, and 4). The spaces of the meshwork, which measure from one to nearly two microns, are occupied by a third substance, clear, homogeneous, and of only slight staining capacity, which has often been called the *ground-substance*. During cell-division, the meshwork in the neighborhood of the dividing nucleus assumes a radiating appearance, giving rise to the so-called asters, or astral systems which are typically double, forming the *amphiaster* (Fig. 3, *b*). We may define the problems suggested by these appearances by a series of questions as follows:

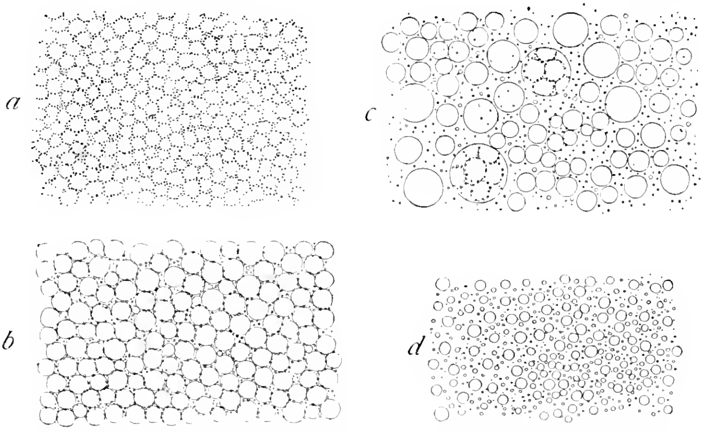

Fig. 1. — (*a*) Protoplasm of the egg of the sea-urchin (*Toxopneustes*) in section; (*b*) protoplasm from a living star-fish egg (*Asterias*); (*c*) the same in a dying condition after crushing the egg; (*d*) protoplasm from a young ovarian egg of the same. (All the figures magnified 1200 diameters.)

1. What is the actual structure that gives the appearance of a meshwork?

2. How faithfully does the preserved structure, as seen in sections, reproduce that existing in life?

3. What is the relation of the astral systems to it?

4. What is the finer structure and origin of the meshwork?

5. Can this structure be taken as typical of all protoplasm; and if not, what is its relation to other forms of protoplasmic structure?

After seeking for answers to these queries, we may finally inquire how they bear on the theoretical views briefly reviewed above. Incidentally, still another interesting question arises, namely: Is it possible to identify any one of the three elements in question — granules, continuous substance, ground-substance — as the *living* substance or *protoplasm* proper, as distinguished from a lifeless *metaplasm*, and, if so, what are its structural relations?

Could we positively answer all these questions, we should have taken a long step forwards in the study of the cell. Far from this, however, in point of fact hardly any two observers have given exactly the same answers to them. Leaving aside the earlier views, we find in the recent literature of the subject two principal general views with a number of modifications of each.

The first of these agrees with the early view of Klein and Van Beneden, that the protoplasm forms a network, *reticulum*, or thread-work, composed of branching fibres embedded in a homogeneous ground-substance which fills the interstices of the network, and with granules or microsomes lying along the course of the threads, or at the nodes of the network. Many of those who adopt this interpretation further agree with their predecessors, that the astral systems formed during cell-division arise directly through a rearrangement of the preëxisting network about active centres of attractive or other forces, somewhat as iron-filings arrange themselves along the radiating lines of force in a magnetic field, — an arrangement which bears a remarkably close though only superficial resemblance to the protoplasmic amphiaster. Boveri, and some others, however, regard the astral system as having no direct relation to the preexisting network, believing that the rays either arise from a specific substance ("archoplasm"), distinct both from the general network and from the ground-substance, or are wholly new formations which, as it were, crystallize afresh out of the protoplasmic substance.

The second view is that of Bütschli, who believes it to be applicable to all forms of protoplasm, and who has been followed by a considerable number of recent investigators. Bütschli's interpretation differs entirely from the foregoing, the meshwork being regarded not as a network, but as an appearance resulting from the optical section of "alveolar" or emulsion-structure. The spaces of the meshwork are drops of liquid occupying spherical spaces or "alveoli"; the "fibres" are optical sections of the thin layers, or lamellæ, by which the drops, or alveoli, are surrounded. Even the astral systems receive the same interpretation, the astral "rays" and "spindle-fibres" being an optical illusion resulting from the radial arrangement of the alveoli, and hence of the inter-alveolar septa by which they are separated.

The greater number of observers of protoplasm have given their adherence to one or the other of the two widely dissimilar views just outlined, though there are others to which we shall return later. Some investigators have taken a position intermediate between these two extremes. Thus Reinke has maintained that the cytoplasm of the echinoderm-egg is alveolar, as described by Bütschli (though, as will appear beyond, he ascribes to this structure a different physiological interpretation), while the astral systems are fibrillar, as held by Van Beneden, and arise as new formations at the cost of the alveolar walls. More recently, Strasburger has developed the related, but still different, view that the cytoplasm of the cell at large consists of two distinct substances, namely, the *trophoplasm*, or general nutritive plasma, which is alveolar, and the *kinoplasm*, or the substance active in division, which is fibrillar and gives rise to astral systems consisting of true rays and fibres.

It is remarkable that the best observers, working in many cases at the same object, should have reached conclusions so diverse. It is obvious, further, that in the face of such contradictions it is impossible to give any discussion of the subject that is not more or less strongly tinged with the personal views of the writer. Such views, by whomsoever expressed, can at present have no more than a provisional value; and this is the last subject on which dogmatism should be allowed. It is with full recognition of these difficulties that I venture to state some of my own conclusions, partly because they may serve to explain, in some measure, to those who have not specialized in this field, how the existing diversity of opinion has arisen, partly because they have perhaps some bearing on the more general questions that were referred to at the outset. I shall take up in order the questions raised at page 5.

The Nature of the Meshwork. — Although in earlier papers I was inclined to regard the meshwork of the echinoderm-egg as a reticulum, further studies have left no doubt whatever, in my opinion, that in the resting cell it is in reality an alveolar structure — or, as I do not hesitate to call it, an *emulsion* — such as Bütschli has described. I was first led to this conclusion through the study of sections of the eggs of sea-urchins (*Toxopneustes*) and star-fish (*Asterias*); but whatever doubt may have remained was completely dissipated by the study of the living eggs of *Asterias* (Fig. 1, *b*), *Echinarachnius*, *Arbacia*, *Ophiura* (Fig. 2, *a*), under high powers. All of these eggs give in life essentially the same appearance, though no two are exactly alike. In all, the protoplasm consists of innumerable closely crowded minute spheres suspended in a clear basis. The spheres may be called the alveolar spheres, or more briefly the *alveoli*, though strictly speaking the latter term should designate the cavities which the spheres fill. The clear basis in which they lie, and which forms the inter-alveolar walls, may, with Mrs. Andrews, be called the *continuous substance.* Scattered about in these walls are numerous granules, or *microsomes*, far smaller than the alveoli, which often give the appearance of an irregular network. If now we compare these appearances of the living protoplasm with those seen in the sections mounted in balsam, we find at first sight very considerable differences. More critical study shows, however, that the differences are almost wholly due to the effect of differential staining and to the difference of refractive index in the mounting media in the two cases. The alveoli of the living protoplasm form the spaces of the meshwork. The latter consists of the continuous substance with the granules suspended in it. In the section, what especially strikes the eye is the meshwork; for the alveolar spheres do not stain, and their contours become indistinct in the highly refracting balsam, while the continuous substance stains slightly, and the granules intensely, thus giving the appearance of a conspicuous granular meshwork. We thus arrive at a definite answer to two of the questions propounded on page 5, namely: (1) the meshwork shown in sections is not a network, but the expression of an alveolar or emulsion-structure, and (2) proper fixation does not produce a mass of coagulation-artifacts, but preserves the visible structure very nearly as it exists in life.

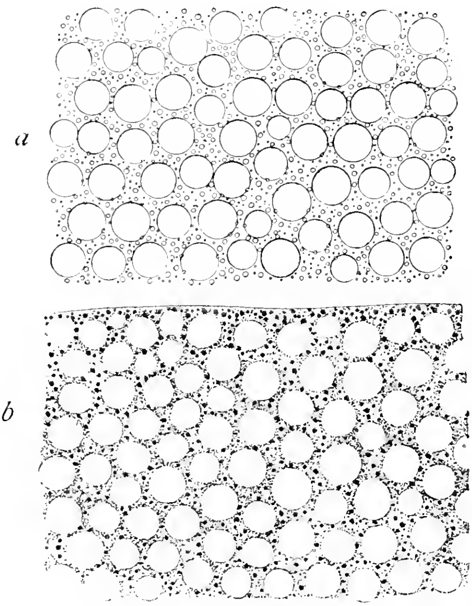

Fig. 2. — (*a*) Protoplasm from a living ophiuran egg (*Ophiura*), slightly compressed, so as to spread the yolk-spheres somewhat apart; (*b*) the same as seen in a section (sublimate-acetic, iron-hæmatoxylin; 1200 diameters).

The above conclusions are based mainly on the study of starfish eggs, but are confirmed by the facts observed in other forms. In *Arbacia* the emulsion is considerably finer, the alveoli measuring on an average no more than 1.0 micron, while the finer granules are relatively less numerous. The pigment-granules characteristic of this form appear to be nothing other than modified alveolar spheres. In *Toxopneustes* the alveoli measure approximately from 1.0 to 1.3 microns, while the granules are more numerous than in *Asterias*. In *Echinarachnius* the alveoli are less uniform in size than in *Asterias*, the largest measuring up to about 1.7 microns, while the granules are less numerous. The egg of *Ophiura*, finally, has an extremely coarse structure, the alveolar spheres measuring on an average 3.0 to 4.0 microns, while the granules or microsomes are also very large and, in the superficial layers of the protoplasm, even more numerous than in *Toxopneustes*. The protoplasm of *Ophiura* (Fig. 2) is highly favorable for study, not only on account of the great size of its elements, but also by reason of the remarkable fact that these elements are colored in life, the alveolar spheres being in most individuals distinctly of an olivaceous or pinkish-brown color, while the larger granules or microsomes are lemon yellow. This circumstance makes possible an observation of great importance, namely, that *all the elements of the protoplasm are liquid or viscid.* If the eggs of *Ophiura* be crushed by pressure on the cover-glass, the protoplasm flows out, most of the alveolar spheres going in advance, while the granules and continuous substance lag behind. Meanwhile, the alveolar spheres often run together to form larger drops of all sizes, the origin of which is placed beyond question by their color. The same is true of the yellow microsomes, though this takes place less readily, and only under somewhat rough treatment. This demonstrates the liquid, or at least viscid, nature of both the spheres and the microsomes, and no less certainly that of the continuous substance in which both lie. As far as the alveolar spheres are concerned, the same observation may readily be made in the colorless protoplasm of *Asterias* (Fig. 1, *c*), *Echinarachnius*, or *Arbacia*, but I could never satisfy myself of the liquid nature of the microsomes in these forms. The case of *Ophiura* renders it highly probable, however, that the granules are liquid in these forms also, — a conclusion which I confess was a surprising result to me; for we are so accustomed from our studies on sections to regard the granules as solid bodies that we are apt to forget that sections show us only coagulated material.

To sum up, a critical study of the living protoplasm of echinoderm-eggs shows that it is a liquid, or rather a mixture of liquids, in the form of a fine emulsion consisting of a continuous substance in which are suspended drops of two general orders of magnitude and of different chemical nature, as indicated by their staining reactions. The larger drops, forming the alveolar spheres, stain only slightly in hæmatoxylin, and constitute the so-called "ground-substance" in the spaces of the meshwork; these have an average size, ranging in the various forms studied from 1.0 micron or less (*Arbacia*) up to 4.0 microns (*Ophiura*). The smaller drops, forming the granules or microsomes, are very much more minute, and stain intensely with iron-hæmatoxylin. The presence of the larger drops determines the primary alveolar structure as described by Bütschli. The smaller drops ("granules") lying between these gives rise to the "secondary," or finer alveolar structure as described by Reinke, and subsequently by Mrs. Andrews, as I understand these authors.

Relations of the Astral Rays to the Meshwork. — We may now make a brief digression to consider the third question propounded on page 5, namely: What is the relation of the astral rays and spindle-fibres to the alveolar substance? It is easy to see, both in sections and in living material, that in a well-developed aster the alveoli are arranged in radiating lines between the astral rays (Fig. 4), precisely as Bütschli and so many others have described. The rays themselves are, however, something more than the radially arranged inter-alveolar septa, for, in the first place, they are often much thicker than these septa, and, in the second place, they stain more intensely than the continuous substance. A careful study of the rays in the echinoderms, and in many other forms (especially in *Nereis*, *Thalassema*, *Lamellidoris*, and *Ascaris*), leaves, I think, no room for doubt that, in sections at least, the rays are actual branching fibrillæ, as described by so many observers since the time of Van Beneden, that thread their way through the continuous substance between the alveoli, often in a zigzag course. The strongest evidence that they are fibrillæ is given by the appearance of the cut ends of the rays as they appear in somewhat excentric or rather thick sections of the asters. In such sections, particularly in the case of large and coarse asters like those of *Nereis* (Fig. 3, *b*), the rays may be seen in the clearest manner to terminate as they pass upwards towards the eye in well-defined cut ends, and I think no one who studies these preparations can doubt that in them the asters are true fibrillar structures.

We may now inquire in what manner the rays arise and grow, and what is the origin of their substance. In the growing aster the rays progressively extend themselves from the centre outwards, gradually losing themselves in the general meshwork. It has been maintained by some writers that the rays grow outwards from their bases like the roots of a plant, and in a certain sense this is undoubtedly true. But it is difficult to believe that all of the material of the rays comes from the base (*i.e.*, from the nucleus or the centrosome), for they often extend themselves throughout the entire cytoplasm, even in cases where, as in the sperm-aster of echinoderms, the centre of the aster remains very small, and the nucleus still consists of a compact mass of chromatin (Fig. 4). It is more probable that they grow at the tip, continually extending themselves at the cost of the material lying in the meshwork. When the rays are followed out periph-

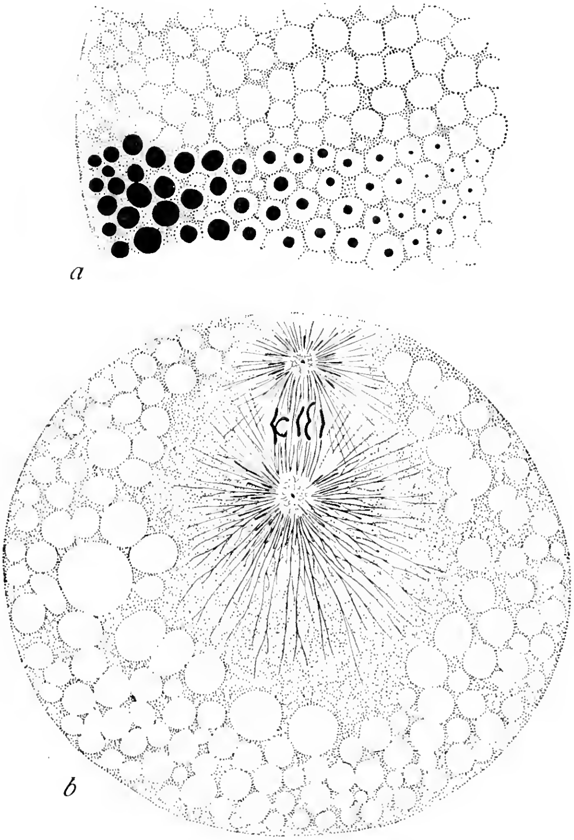

Fig. 3. — (*a*) Protoplasm and yolk-spheres from the egg of *Thalassema* in section. The upper part of the section shows the result of prolonged extraction of the dye (iron-hæmatoxylin), the lower half represents varying degrees of extraction (1200 diameters); (*b*) egg of *Nereis* in section showing yolk-spheres and the first polar amphiaster above (600 diameters).

erally they may often be seen to run out into rows of granules like beads on a string. Van Beneden, who has been followed by many later writers, was inclined to regard the rays as essentially rows of microsomes strung together by a homogeneous clear substance, — *i.e.*, by the continuous substance, — and I was led to the same conclusion in the case of sea-urchin eggs. A study of the asters in *Ophiura* throws doubt upon this conclusion, for it is here certain that the larger and deeply staining microsomes do not build up the ray, but are quite irregularly scattered along its course. The rays here mainly arise, I believe, in, and at the expense of, the continuous substance, and the linear arrangement of the microsomes is incidental to the differentiation of this substance along a definite tract which more or less involves the microsomes as it progresses. This conclusion probably also applies to other forms. The material active in the ray-formation appears to be the continuous substance,

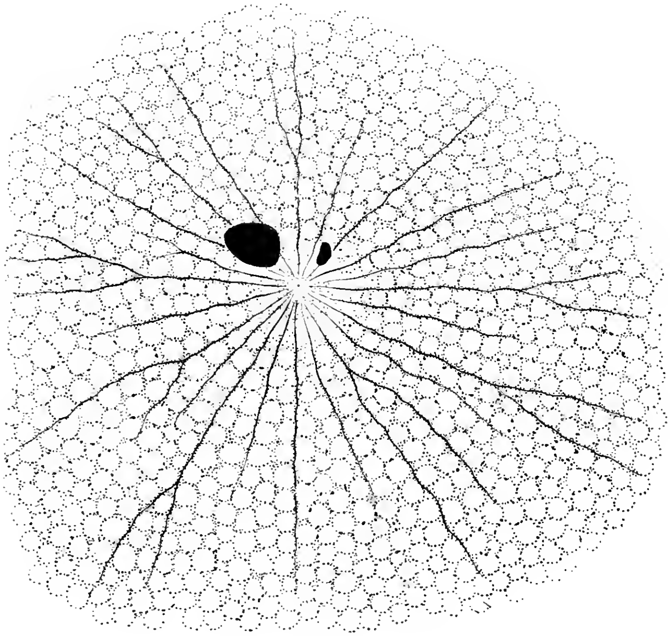

FIG. 4. — Section of sea-urchin egg (*Toxopneustes*), $1\frac{1}{2}$ minutes after entrance of the spermatozoön, showing sperm-nucleus, middle piece, and aster (about 2000 diameters).

and while the microsomes may, and probably in many cases do, contribute to the ray, they probably play the part of reserve-material rather than of active elements.[1]

To sum up, the general result indicates that the opinions regarding the aster-formation referred to on page 6 can in a measure be reconciled. In the case of echinoderm-eggs Bütschli and Erlanger correctly describe the aster as involving a radial arrangement of the alveoli, but they have failed to recognize the fibrillæ that lie between them, and Boveri is therefore thoroughly justified in the contention that the astral systems cannot be regarded as merely a radial configuration of the preëxisting meshwork. I nevertheless think that Hertwig, Reinke, and myself were right in the contention, which has been made also by many others, that the rays grow by progressive differentiation out of the general cytoplasmic meshwork, and that there is no ground, in the echinoderm-egg at least, for the recognition of a specific "archoplasm" or "kinoplasm" from which they arise.

Finer Structure and Origin of the Meshwork. — We may now consider what is, I think, the most suggestive of the questions propounded, namely, that relating to the finer structure and origin of the meshwork. We have thus far distinguished sharply between alveolar spheres, granules, or microsomes, and continuous substance. Morphologically considered, however, there is good reason for the view that all these are but different gradations of one structure. In the first place, a nearly or quite complete series of size-gradations exists between the largest alveoli and the microsomes (Fig. 1, *b*, *c*). Although most of the alveoli vary but slightly in size from the mean, a little search shows the presence of many smaller ones, and here and there they may seem almost, if not quite, as small as the larger microsomes. In the second place, careful study of the "continuous" substance in life, especially in the crushed protoplasm, shows that the larger microsomes in turn graduate down to granules so small as to lie near or at the limit of microscopical vision. The "continuous" substance is, in other words, filled

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[1] As already pointed out, we cannot assume that the ray is *merely* an accumulation of the continuous substance on account of its different staining capacity.

with granules, *i.e.*, drops, of all sizes, ranging from the smallest visible ones up to the largest alveoli. It is this fact which Mrs. Andrews, as I understand her statements, has in view in maintaining that the coarser alveolar structure "is not indeed the final structure of the living substance, but is part only of an infinitely graded series of vesiculations of the protoplasmic foam" (**'97**, p. 12), and with this statement I entirely agree. But we cannot stop here. Irresistibly the further question suggests itself: Why should we place the end of this series at the end of microscopical vision under a 1.5 mm. immersion objective — which is of course a perfectly arbitrary and artificial limit? It is impossible to doubt that powers still higher than any at our command would reveal the existence of granules still smaller, and that what appears as "continuous" or "homogeneous" substance is itself an emulsion beyond the range of vision.

We may now inquire whether the coarser visible alveolar structure is characteristic of all protoplasm. This question has in a measure already been answered, for in these very eggs we have seen the alveolar structure giving rise to a fibrillar one in the aster-formation — in other words, the protoplasm of the same cell may in different phases pass back and forth from one state into another. This fact appears in its clearest form when we study the growth of the ovarian ova, which gives us many additional suggestions of high interest. *The entire coarser alveolar structure, as described above, — i.e., the foam-structure of Bütschli, — is in these eggs of secondary origin.* The very young living ovarian eggs consist of "homogeneous" protoplasm, such as has been described by many botanists in the embryonic tissue-cells, through which are irregularly scattered a few small spheres and many excessively small granules. As growth proceeds, both the spheres and the granules increase in size, the latter enlarging to form new spheres, while new granules continually emerge from the protoplasmic background into the limits of vision. In the middle stages of growth, the protoplasm is thus converted into an emulsion, being filled with spheres of all sizes, ranging downwards from 1.0 micron to the smallest granules, but still showing no regular arrangement

(Fig. 1, *d*). As the egg approaches maturity, the spheres become differentiated into two groups, the larger ones becoming approximately of the same size (cf. p. 10) to form the alveolar spheres and crowding together, while the smaller ones remain as the microsomes and finer granules embedded in the remains of the continuous substance which forms the basis of the meshwork. In one sense, therefore, the alveolar spheres and the microsomes are only different stages in the same morphological series, — though it should be remembered that they differ chemically as well as in size, and I do not mean to imply that the one may develop into the other, — and both the alveolar and the fibrillar or reticular structures in these eggs are of secondary origin. If this be the case, neither of these types of structure can be of fundamental importance; and I fully agree with the opinion of Kölliker, which has been adopted by an increasing number of later observers, that *no universal or even general formula for protoplasmic structure can be given.* The evidence indicates that alveolar, granular, fibrillar, and reticular structures are all of secondary origin and importance, and that *the ultimate background of protoplasmic activity is the sensibly homogeneous matrix or continuous substance* in which those structures appear.

I do not mean to say that this is the only "living" element in the cell. The distinction between "living" and "lifeless," between "protoplasmic" and "metaplasmic," substances is exceedingly difficult to define, — largely on account of our vague and inconsistent use of terms, for in practice we continually use the word "living" to denote various degrees of vital activity. Protoplasm deprived of nuclear matter has lost, wholly or in part, one of the most characteristic vital properties, namely, the power of synthetic metabolism; yet we still speak of it as "living," because it may for a long time perform some of the other functions, manifesting irritability and contractility, and showing also definite coördinations of movements (as in the enucleated protozoan); and in like manner various structural elements of the cell may be termed living in a still more restricted sense. In its fullest meaning, however, the word "living" implies the existence of a group of coöperating factors more complex than those manifested by any one substance or structural element in the cell, and I am therefore thoroughly in accord with those who have insisted that life in its full sense is the property of the cell-system as a whole rather than of any one of its separate elements. Nevertheless, we are perhaps justified in maintaining that the continuous substance is the most constant and active element, and that which forms the fundamental basis of the system, transforming itself into granules, drops, fibrillæ or networks in accordance with varying physiological needs.[1]

Whether any or all of these elements are "living" or "lifeless" depends largely on the sense in which these words are used; and it is well, therefore, to follow the example of Sachs, in substituting for these words, as applied to special structural elements of the cell, the terms "active" and "passive," which properly admit of degrees of comparison. The distinction between "protoplasmic" (active) and "metaplasmic" or "paraplasmic" (passive) elements, though a real and necessary one, thus becomes, after all, one of degree only.

We are thus brought to consider another point of some interest suggested by the comparative study of the facts described above. Bütschli states that in the true or finer alveolar structure, characteristic of protoplasm in general, the alveoli do not measure more than 2.0 microns, and as a rule are considerably smaller. This, he insists, is not to be confounded with a "coarser vacuolization," characterized by larger drops or spheres, which may secondarily arise in the finer structure. Again, Reinke and Waldeyer in a somewhat similar manner characterize as "pseudo-alveolar" a structure arising secondarily through the deposit of passive metaplasmic products of metabolism, such as yolk-spheres, fat-drops, and the like, in the living protoplasmic basis. Both distinctions break down, I think, in the light of the foregoing facts. In most of the forms considered, —*Arbacia*, *Toxopneustes*, *Echinarachnius*, *Asterias*, — the alveolar spheres are considerably less than 2.0 microns (1.0 to 1.7),

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[1] It is hardly necessary to state that this view is not original, except in so far as it has been directly suggested by the observations described above; for it has been more or less definitely maintained by many others, and I am only expressing what seems to be a growing opinion among workers in this field.

and the structure is therefore a true alveolar one in Bütschli's sense; indeed, Bütschli himself describes and figures the protoplasm of the *Sphaerechinus* egg as an example of that structure. In *Ophiura*, however, the spheres measure up to 3.0 or 4.0 microns, and are undoubtedly "yolk-spheres" in the usual sense. It is, however, quite certain from the ovarian development of these eggs that they differ from the others only in degree, and that Bütschli's criterion of size gives no satisfactory ground for any real distinction. The alternative is to regard all the forms as pseudo-alveolar, irrespective of the size of the alveolar spheres, which are in all cases to be regarded as metaplasmic bodies; and this is the view which Reinke specifically applies to *Sphaerechinus.* But if this view be adopted, we seek in vain for any ground of distinction between such a fine "pseudo-alveolar" structure as that of *Arbacia*, and the "true" alveolar structure of tissue cells, and are forced to the conclusion that in the latter case also the alveolar substance consists of passive or metaplasmic material, — a view which has in fact been adopted by some writers. For my part, I am convinced that the entire distinction is without adequate basis, and that no definite boundary-line can be drawn between even the largest deutoplasm-spheres, vacuoles, or other metaplasmic deposits, the alveolar spheres of *Arbacia* or *Toxopneustes*, and those occurring in tissue-cells; and probably all are, in the sense indicated above, to be classed among the relatively passive or metaplasmic material.

How generally the alveolar, reticular, or fibrillar formations may occur is a matter still to be determined by observation. It is probable that the alveolar structure will be found to be of more general occurrence than has been supposed; and judging by the appearance observed in echinoderm and other eggs, and in coagulated albumen and other structureless proteids, I suspect that some cases of so-called "reticular" formations will be found to arise through the more or less imperfect fixation of the alveolar, leading to the coagulation, contraction, and breaking down of the alveolar walls,[1] though I do not for a moment mean to imply that such is the case with all reticula.

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[1] It may be well to point out that Rhumbler has produced true fibrillar and reticular formations in coagulated artificial gelatine-emulsions.

What light, if any, do the foregoing general conclusions throw on the theoretical views outlined at the beginning of this lecture? The answer must be: None that is clear and satisfactory, for the background of all the phenomena appears to lie in the invisible organization of a substance which seems to the eye homogeneous. Yet there is, I think, much in these conclusions to suggest, and nothing to contradict, the hypothesis that the "homogeneous" or "continuous" substance may be composed of ultra-microscopical bodies by the growth and differentiation of which the visible elements arise, and which differ among themselves chemically and otherwise, as is the case with the larger masses to which they give rise. I will not enter upon a discussion of the question whether these bodies are merely molecules, more or less complex, or groups of molecules forming protoplasmic units or micellæ, but will only make three suggestions. First, if such units exist, they cannot be identified with the visible granules or "bioblasts" of Altmann, but are bodies far smaller. Second, if there be any truth in what has been said above regarding the localization of "living" matter in the cell, such protoplasmic units, if they exist, cannot properly be called "biophores," since life is a manifestation of the system which they form and not of the individual units. The corpuscular or micellar theory of protoplasm, as an hypothesis of morphological organization, should not be confounded with the physiological theory that biophores or pangens are "elementary living units." Third, by ascribing to these hypothetical units the power of growth and division, in accordance with the pangen theory, we are enabled to get a certain amount of light upon some of the most puzzling questions of cytology, such, for example, as the ultimate nature and origin of dividing cell-organs like the nucleus or the plastids, and especially such a contradiction as that presented by the centrosome which may apparently arise either *de novo* or by division of a preëxisting body of the same kind. As De Vries and Wiesner have so suggestively urged, the power of division on which the law of genetic continuity rests and which is manifested by morphological aggregates of so many different degrees, may have its root in a like power of the primary units at the bottom of the series, out of which all the higher members are built. But while giving due weight to this suggestive hypothesis, we may question whether its acceptance does not introduce as many new special difficulties as those which it sets aside; while we must admit that it leaves untouched the fundamental problem of division. The solution of this problem may perhaps have to be sought in a quite different direction from the pangen hypothesis. Whether we shall succeed in finding it is another question.

SECOND LECTURE.

CELL-LINEAGE AND ANCESTRAL REMINISCENCE.[1]

EDMUND B. WILSON.

EVERY living being, at every period of its existence, presents us with a double problem. First, it is a complicated piece of mechanism, which so operates as to maintain, actively or passively, a moving equilibrium between its own parts and with its environment. It thus exhibits an adaptation of means to ends, to determine the nature of which, as it now exists, is the first task of the biologist. But, in the second place, the particular character of this adaptation cannot be explained by reference to existing conditions alone, since the organism is a product of the past as well as of the present, and its existing characteristics give in some manner a record of its past history. Our second task in the investigation of any problem of morphology or physiology must accordingly be to look into the historical background of the phenomena; and in the course of this inquiry we must make the attempt, by means of comparisons with related

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[1] This lecture is based on a paper entitled "Considerations on Cell-Lineage and Ancestral Reminiscence, Based on a Reëxamination of Some Points in the Early Development of Annelids and Polyclades," in *Ann. N. Y. Acad. Sci.*, 1898. In some passages the wording of that paper has been reproduced with only slight change. With the exception of Fig. 4, *the figures are entirely schematic and are designed to show only the broadest and most essential topographical features.* For this purpose the subdivisions of the micromeres have been omitted, and, except in Fig. 4, none of the figures represent the actual condition of the embryo at any given period. While, therefore, very misleading in matters of detail, they are, I think, true to the essential phenomena; and through the simplification thus effected the reader is spared a mass of confusing descriptive detail in no way essential to the broad relation on which it is desired to focus the attention.

phenomena, to sift out adaptations to existing conditions from those which can only be comprehended by reference to former conditions. Phenomena of the latter class may, for the sake of brevity, conveniently be termed "ancestral reminiscences," — though it may not be superfluous to remark that every characteristic of the organism is in a broad sense reminiscent of the past.

It is in embryological development that ancestral reminiscence is most familiar and most striking. We all know that development rarely takes the shortest and most direct path, but makes various detours and sometimes even moves backward so that the adult may actually be simpler than the embryo. Such vagaries of development are in many cases only intelligible when regarded as reminiscences of bygone conditions, either of the adult or of the embryo. Sometimes these records of the past are so consecutive and complete that the individual development, or ontogeny, may be said to repeat or recapitulate the ancestral development, or phylogeny. The development of the toad's egg, for example, probably gives in its main outlines a fairly true picture of the ancestral history of the toad race, which arose from fish-like ancestors, developed into aquatic air-breathing tailed forms, and finally in their last estate became tailless terrestrial forms. It was such facts as these that led Haeckel, building on the basis laid by Darwin and Fritz Müller, to the enunciation of the famous so-called "biogenetic" law, that the ontogeny, or history, of the individual tends to repeat in an abbreviated and more or less modified form the phylogeny, or history, of the race. The event has shown that actual recapitulation or repetition of this kind is of relatively rare occurrence. Development more often shows, not a definite record of the ancestral history, but a more or less vague and disconnected series of reminiscences, and these may relate either to the adult or to the embryonic stages of the ancestral type. Thus the embryo mammal shows in its gill-slits and aortic arches what must probably be regarded as reminiscences of a fish-like adult ancestor, while in the primitive streak it gives a reminiscence not of an adult form but of an ancestral mode of development from a heavily yolk-laden egg like that of the reptiles.

If we survey the general field of embryology, we find that ancestral reminiscence in development is most conspicuously shown and has been longest known in the later stages, and many of the most interesting and hotly contested controversies of modern embryology have been waged in the discussion of the possible ancestral significance of larval forms, such as the trochophore, the *Nauplius*, the ascidian tadpole, and many others. It is generally admitted, too, that ancestral reminiscences may occur in earlier embryonic stages. While few naturalists would to-day accept Haeckel's celebrated Gastræa theory in its original form, probably still fewer would deny that the diblastic embryo (gastrula, planula, etc.) of higher forms is in a certain sense reminiscent of the origin of these forms from diblastic ancestors having something in common with existing cœlenterates.

It is in respect to still earlier stages, namely, those including the cleavage of the egg, that the greatest doubt now exists; and there is hardly a question in embryology more interesting or more momentous than whether these stages may exhibit ancestral reminiscence, and whether they, like the later stages, exhibit definite homologies, and thus afford in some measure a guide to relationship. None of the earlier embryologists were disposed to answer this question in the affirmative. To them, and it should be added to some of our contemporaries as well, the cleavage of the ovum was "a mere vegetative repetition of parts," the details of which had no ancestral significance, and the ontogeny first acquired a definite phyletic meaning and interest with the differentiation of the embryonic tissues and organs. To these observers the cleavage of the ovum presented merely a series of problems in the mechanics of cell-division, and its accurate study was almost wholly neglected as having no interest for the historical study of descent. And yet it was long ago shown that the blastomeres of the cleaving ovum have in some cases as definite a morphological value as the organs that appear in later stages. Kowalevsky and Rabl traced the mesoblast-bands in annelids and gasteropods back to a single cell, which still later research has shown to have the same origin and fate, and hence to be homologous in the two cases by every criterion at our command. A long series of later researches, beginning with Whitman's epoch-making studies on the cleavage of *Clepsine*, has demonstrated analogous facts in the case of many other cells of the cleaving ovum, and has finally shown that in many groups of animals (though apparently not in all) the origin of the adult organs may be determined cell by cell in the cleavage stages; that the *cell-lineage* thus determined is not the vague and variable process it was once supposed to be, but is in many cases as definitely ordered a process as any other series of events in the ontogeny; and that it may accurately be compared with the cell-lineage of other groups with a view to the determination of relationships.

The study of cell-lineage has thus given us what is practically a new method of embryological research. The value and limitations of this method are, however, still under discussion, and among special workers in this field opinion as to its morphological value is still so widely divided that most of its results should be taken as suggestive rather than demonstrative. Like other embryological methods, it has already encountered contradictions and difficulties so serious as to show that it is no *open sesame*. In some cases closely related forms (*e.g.*, gasteropods and cephalopods) have been shown to differ very widely, apparently irreconcilably, in cell-lineage. In other cases (echinoderms, annelids) the normal form of cleavage has been artificially changed without altering the outcome of the development. In still other cases (*e.g.*, in teleost fishes) the form of cleavage has been shown to be variable in many of its most conspicuous features, so that apparently no definite cell-lineage exists. These and many other facts, less striking but no less puzzling, can be built into a strong case against the cell-lineage program, and I wish to acknowledge its full force. Admitting all the difficulties, I am nevertheless on the side of those who as morphologists believe that the study of cell-lineage has demonstrated its value, and that it promises to yield more valuable results in the future. In this lecture I propose to illustrate some of the more interesting results already attained, and some of the suggestions that they give for future work, by a broad consideration of the cell-lineage of three related groups of animals which on the one hand have been very carefully examined as regards their anatomical and general embryological relationships, while on the other hand their cell-lineage has been more exhaustively studied than that of any other forms. These groups are the platodes (more especially the *Turbellaria*), the mollusks, and the annelids.

That these three groups belong in the same morphological series will probably be admitted by all zoölogists, and most will no doubt further agree with the view of Lang, that in the essential features of their organization the platodes are not very far removed from the ancestral type from which the two higher groups have sprung, the former having remained non-metameric like the platodes, while the latter have acquired metamerism. Accepting this view we should expect, if there be any evidence of race-lineage in cell-lineage, to find in the annelids and mollusks a common type of cleavage, and one which in its main features may be derived from that of the platode. Recent studies in cell-lineage have, on the whole, justified this expectation, and have brought to light some cases of vestigial processes in cleavage which are, I believe, to be reckoned among the most striking and beautiful examples of reminiscence in development. It is especially to these cases that I wish to direct attention.

The cleavage of a number of *Turbellaria* and nemerteans, and of many annelids, gasteropods, and lamellibranchs, has now been shown to conform to a common type which, though complex in detail, is exceedingly simple in its essential plan. A few exceptions there certainly are; but some of these are apparent only (for example, in the acœlous *Turbellaria*), and are readily reducible to the type, while others are undoubtedly correlated with bygone changes in the mode of nutrition of the ovum (as in some of the earthworms and leeches). The most conspicuous exception is afforded by the cephalopods, which have a mode of cleavage entirely unrelated to that of the other mollusks; but the entire development of this group is of a highly modified character. Fully recognizing the real exceptions, we nevertheless cannot fail to wonder at the marvellous constancy with which the cleavage of the polyclades, nemertines, annelids, gasteropods, and lamellibranchs conforms to the typical mode of development. In all these forms the egg first divides into four quadrants. From these at least three and sometimes four or five quartets of cells — usually smaller, and hence designated as *micromeres* — are successively produced by more or less unequal cleavages towards the upper pole. The arrangement of these micromeres (Fig. 1) is constant and highly characteristic, the first quartet being more or less displaced, or, as it were, rotated in a direction corresponding with the hands of a watch (clockwise), the second in the opposite direction (anti-clockwise), the third clockwise again, and so on, the spindles of each division being at right angles to those of the preceding and following. In the later subdivisions of the micromeres, also, a most remarkable agreement has been observed; but I shall pass this over entirely in order to focus attention on the broader features of the development.

A large part of the work in cell-lineage during the past ten years has been devoted to a comparison of the morphological value of these quartets of cells in the annelids, mollusks, and platodes; and the remarkable and interesting fact is now becoming apparent that while they do not have exactly the same value in all the forms, they nevertheless show so close a correspondence both in origin and in fate that it seems impossible to explain the likeness save as a result of community of descent. The very differences, as we shall see, give some of the most interesting and convincing evidence of genetic affinity; for processes which in the lower forms play a leading *rôle* in the development are in the higher forms so reduced as to be no more than vestiges or reminiscences of what they once were, and in some cases seem to have disappeared as completely as the teeth of birds or the limbs of snakes. The processes in question relate to the formation of the mesoblast in its relation to the micromere-quartets, and on them the whole discussion may be made to turn.

The higher types — *i.e.*, the annelids, gasteropods, and lamellibranchs — have for some time been known to agree closely in the general value of the quartets. Rabl first demonstrated that in *Planorbis* the entire ectoblast is formed from the first three quartets, while the mesoblast-bands arise from the posterior cell of the fourth quartet, the other three, with the remains of the primary quadrants, giving rise to the entoblast (Fig. 1). The same general result has been reached by subsequent investigators of molluscan cell-lineage, though there are one or two apparent exceptions (*e.g.*, *Teredo*, according to Hatschek) that demand reinvestigation. The same remarkable fact holds true throughout the annelids,[1] the well-determined

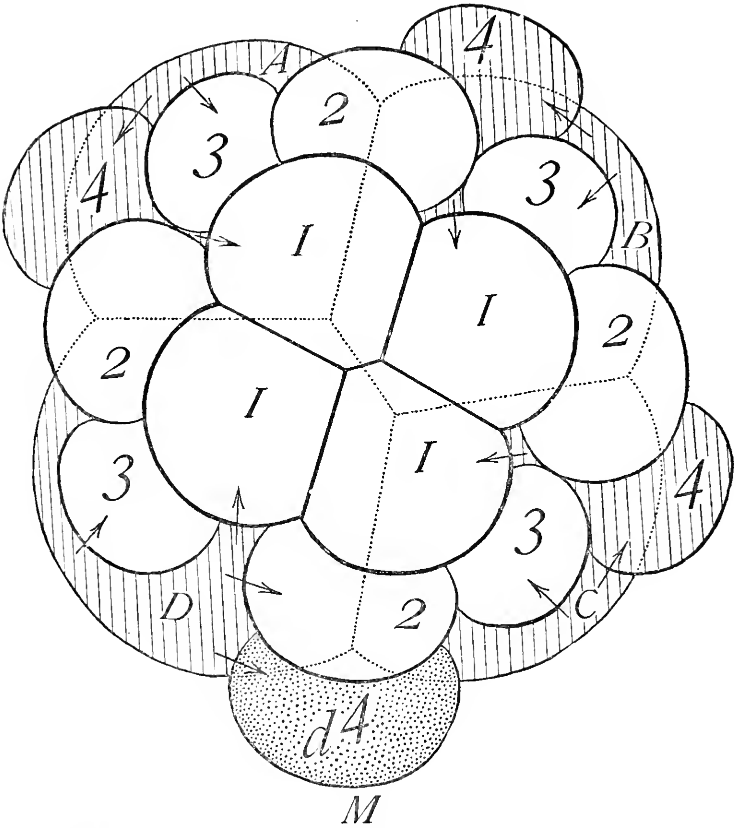

FIG. 1. — Diagram of the typical quartet-formation in an annelid or gasteropod; the quartets numbered in the order of their formation; *A*, *B*, *C*, *D*, the basal quadrants. Ectoblast unshaded, mesoblast dotted, entoblast ruled in parallel lines. In many forms (*e.g.*, *Aricia*) a fifth quartet (entoblastic) is formed; in others (*e.g.*, *Nereis*) only three complete quartets and the posterior member of the fourth quartet (d^4 or M).

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[1] See footnote at p. 36 for reference to Eisig's widely divergent account of the development of *Capitella*.

exceptions being some of the earthworms and leeches referred to above, in which the typical relations seem to have been disturbed through changes in the nutrition of the embryo. Wherever the typical quartet formation takes place — and this is the case in nearly all the forms that have been adequately examined — the general value of the quartets is the same, the first three giving rise to the entire ectoblast, the fourth giving rise, one cell to the mesoblast-bands and the other three to entoblast, while the remnants of the primary quadrants, including the fifth quartet if one is formed, give rise to the entoblast. This result seems almost too simple and produces an impression of artificiality which may probably account for the reluctance with which it has been accepted in some quarters; but I think it is not too much to say that few facts in embryology have been more patiently studied or more accurately determined. The above statement does not, however, contain the whole truth; but before completing it we may advantageously turn to the development of the *Turbellaria*.

It was long since shown by researches, beginning with Hallez and Götte and culminating in those of Lang, that the cleavage of polyclades shows an extraordinarily precise resemblance to that of the annelids and mollusks. Taking Lang's work on *Discocœlis* as a type, we find four quartets of cells successively produced from the primary or basal quadrants, following exactly the same law of displacement as in the higher types, assuming the same arrangement, and in their subsequent subdivision up to a relatively late stage following so exactly the plan of the annelid egg that even a skilled observer might easily mistake one for the other (Fig. 4, *A*). Despite this accurate agreement in the form of cleavage, Lang's observations seemed to show that the cell-quartets had a totally different value from those of the higher forms; for he believed the first quartet to produce the entire ectoblast, the second and third to give rise to the mesoblast, while the fourth quartet, with the basal cells, formed the entoblast (Fig. 2, B). Such a result was more than a stumbling-block in the way of the comparison. It was subversive of the whole cell-lineage program; for it seemed to show that the cell-lineage of derivative animals (*i.e.*, annelids and gasteropods), while exactly conforming to the ancestral *form* of cleavage (*i.e.*, that of the *Turbellaria*), differed *toto cœlo* from it in morphological significance. When, some years ago, I first called attention to this difficulty, I felt constrained to the admission that, in the face of such a contradiction, the study of cell-lineage could only be regarded as of very restricted value in morphological investigation; indeed, in a lecture delivered here four years ago on the inadequacy of the embryological criterion of homology,[1] I cited this very case as representing a climax in the contradictions of comparative embryology.

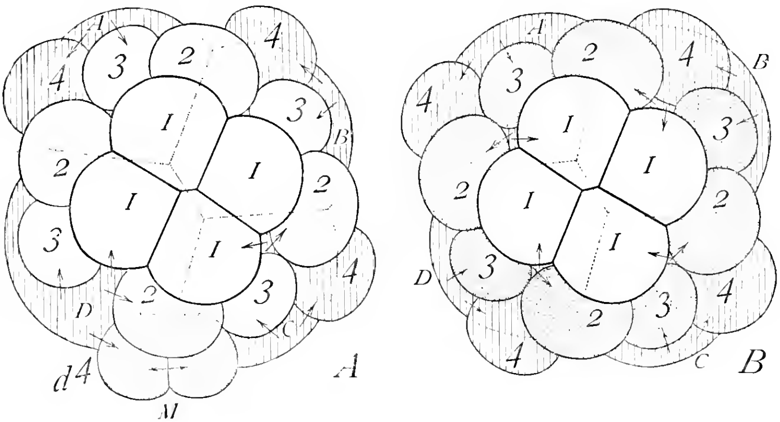

FIG. 2. — Diagrams contrasting the value of the quartets in an annelid or gasteropod (*A*) with those of a polyclade according to Lang's original account (*B*). Lettering and shading as in Fig. 1. (The true proportions of the basal quadrants and the fourth quartet, which are here misrepresented, are shown in Fig. 4. It is characteristic of the polyclades that the fourth quartet-cells are greatly enlarged at the expense of the basal quadrants.)

It is not rare in the history of science to find that fuller knowledge may so change the point of view as to transform a seeming difficulty into a pillar of support; and it seems not unlikely that such may be the case with the present one, though some new difficulties have arisen which still await solution. The new evidence relates, on the one hand, to the annelids and mollusks, on the other hand to the polyclades; and since on both sides it tends to bridge a gap which once seemed hopelessly wide, I shall consider it in some detail. In approaching

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[1] "The Embryological Criterion of Homology." Wood's Holl Biological Lectures, 1894, p. 113.

this evidence the two principal difficulties should be clearly borne in mind. The first lies in the fact that the mesoblast-bands of the annelids and mollusks arise from one cell of the *fourth* quartet, while in the polyclade the mesoblast was stated to arise from all of the eight cells of the *second* and *third* quartets. The second difficulty relates to the ectoblast, which in the annelid and mollusk arises from the twelve cells of the first, second, and third quartets; while in the polyclade it was believed to arise solely from the first quartet (Fig. 2). We may consider these two difficulties in order.

As regards the first point, a series of researches during the past three years have shown that in some of the mollusks and annelids the mesoblast has a double origin, a part — and usually the major part — arising from the posterior cell of the fourth quartet, as stated above, while a part arises from cells of the second or third quartet, as in the polyclade (Fig. 3). The major part — which, for reasons that will appear beyond, I propose to call the *entomesoblast* — gives rise to the so-called mesoblast-bands. The minor part, or *ectomesoblast* ("secondary mesoblast," "larval mesoblast," of various authors), apparently does not contribute to the formation of the mesoblast-bands, and in at least one case — namely, that of *Unio*, as described by Lillie — it gives rise to cells of a purely larval character and designated as "larval mesenchyme." The first step in this direction was that of Lillie, just referred to, who in 1895 announced the discovery that in a lamellibranch, *Unio*, one cell of the *second* quartet (a^2 on the left side) gives rise not only to ectoblast, but also to a single mesoblast-cell which passes into the interior, divides, and gives rise to some of the larval muscles ("larval mesenchyme," Fig. 3, *C*). Lillie's discovery was quickly followed by the no less interesting one of Conklin that in another mollusk, the gasteropod *Crepidula*, *three* cells of the second quartet, median anterior, right, and left (b^2, c^2, d^2), likewise give rise to mesoblastic as well as to ectoblastic elements (Fig. 3, *B*), — a process still more forcibly recalling the origin of the mesoblast in the polyclade.

Two years later mesoblastic cells were found, both in the mollusks and in the annelids, to arise from members of the *third* quartet. The first of these cases was observed by Wierzejski (1897) in the case of *Physa*, where the two anterior cells of this quartet (c^3, b^3) give rise to mesoblastic as well as to ectoblastic cells, and exactly similar facts were soon afterwards observed by Holmes in *Planorbis*. Simultaneously with these researches I independently discovered in the annelid *Aricia* two mesoblast cells arising from the two posterior cells of either the second or the third quartet (*i.e.*, from c^3 and d^3 or

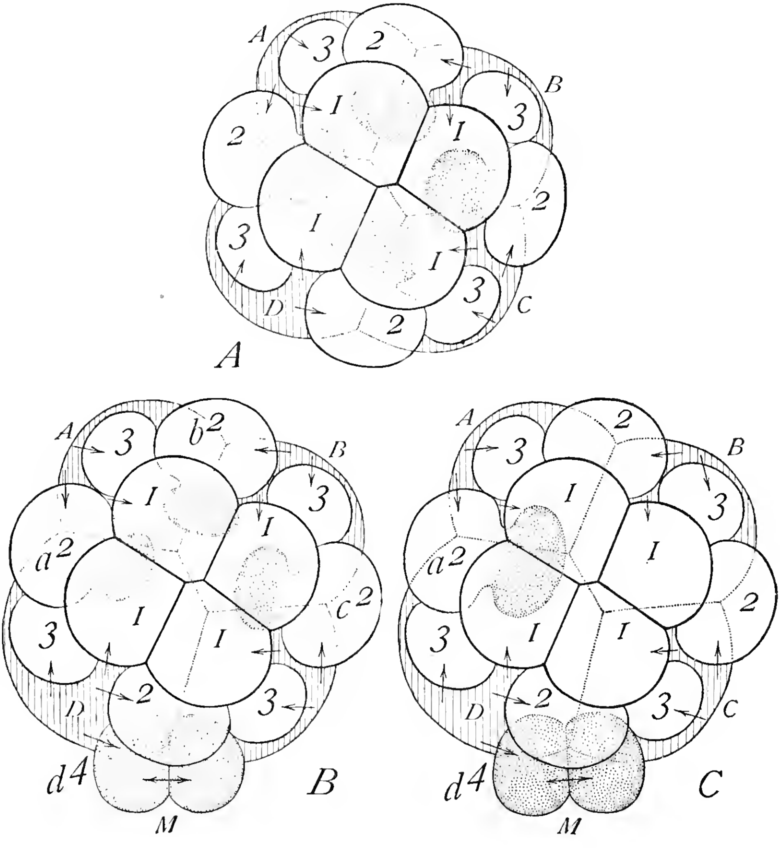

FIG. 3. — Diagrams illustrating the value of the quartets in a polyclade (*Leptoplana*), a lamellibranch (*Unio*), and a gasteropod (*Crepidula*). Lettering and shading as in Fig. 1. (For comment on these figures see footnote at first page.) *A*, *Leptoplana*, showing mesoblast-formation in the second quartet. (Cf. Fig. 4.) *B*, *Crepidula*, showing source of ectomesoblast (from a^2, b^2, c^2) and entomesoblast (from quadrant *D*). *C*, *Unio*, ectomesoblast formed only from a^2.

from d^2 and c^2), though I could not positively determine which. This was immediately followed by Treadwell's discovery that in the annelid *Podarke* mesoblast cells are formed from three cells of the third quartet, namely, the anterior median and the two lateral cells (a^3, c^3, d^3). It was thus shown that in at least four genera of mollusks and two of annelids a part of the mesoblast has an origin which recalls that of the polyclades, and the view is irresistibly suggested that the formation of this *ectomesoblast* in one, two, or three quadrants in the higher types is a vestigial process or ancestral reminiscence of what occurred in all four quadrants in the ancestral prototype and still persists in the polyclade.

The second difficulty — *i.e.*, the origin of the ectoblast — has entirely disappeared upon a reëxamination of the cell-lineage of a polyclade (*Leptoplana*) which I was enabled to make in the summer of 1897. In this form careful study shows in the clearest manner that the formation of ectoblast is not confined to the first quartet, but that all of the twelve cells of the first three quartets contribute to the ectoblast, precisely as is the case in the annelids and mollusks (Fig. 3, *A;* Fig. 4, for details). A comparison with Lang's figures gives every reason to believe that the same is true in *Discocœlis* and the other forms studied by him, and that on this point he fell into an error which was certainly very pardonable at the time. The quartet-cells from which in the polyclade the mesoblast arises are, therefore, not pure mesoblasts, as Lang supposed, but are *mesectoblasts*, precisely like the cells from which the "larval mesoblast" arises in *Crepidula* or *Unio*.[1]

The researches reviewed up to this point have cleared up the contradiction relating to the second quartet. Passing now to the third and fourth quartets, we find that the newer researches have introduced a new difficulty with respect to each of these quartets; but the new difficulties differ from the old in

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[1] In *Leptoplana* each cell of the second quartet divides off in succession three ectoblast cells before the delamination of mesoblast into the interior occurs at the fourth division (Fig. 4). In *Unio*, according to Lillie, the larval mesoblast is definitely separated at the third division of the micromere (a^2). Professor Conklin informs me that in *Crepidula* the ectomesoblast is formed at about the fourth or fifth division of the micromeres (a^2, b^2, c^2).

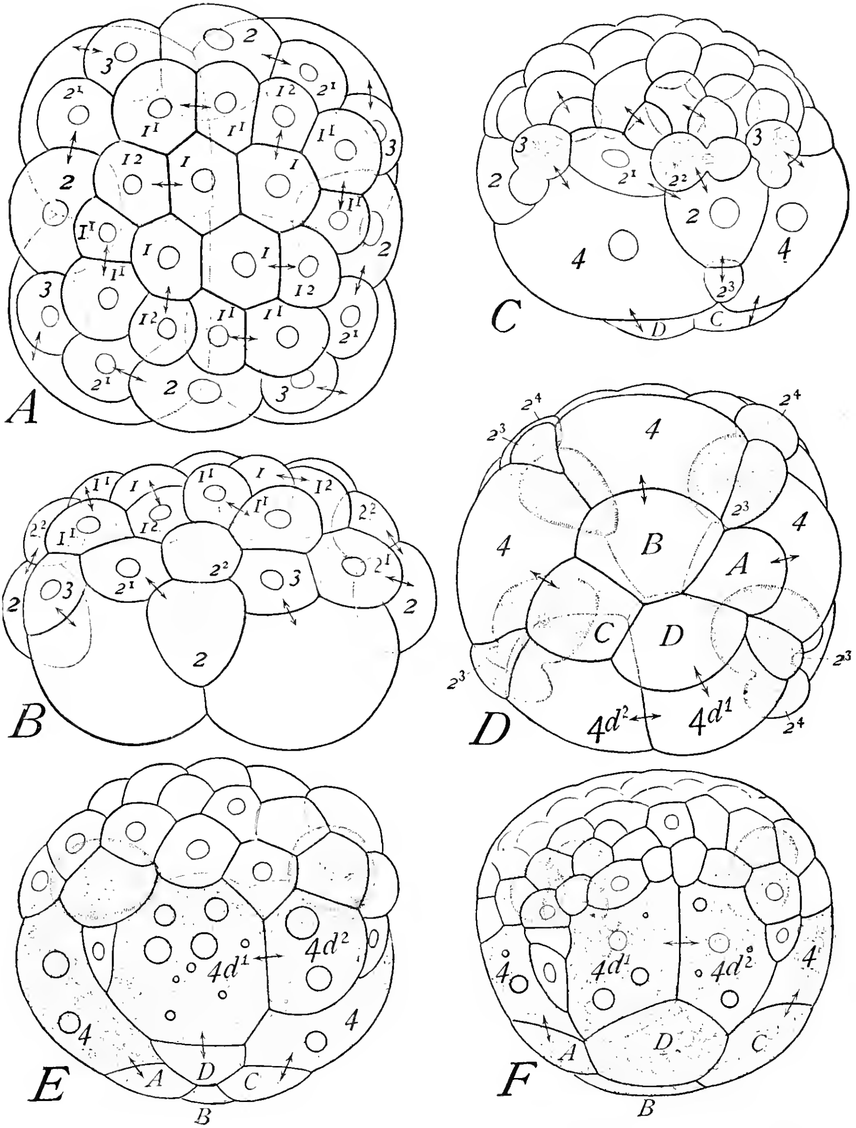

FIG. 4, LEPTOPLANA. — (Camera drawings from the transparent living embryos. In these figures the subdivisions of the micromeres are accurately shown.)

A, 32-cell stage, from the upper pole; *B*, 36-cell stage, from the side, showing second division of 2; *C*, side view, approximately 60 cells, showing the third ectoblast cell (2^3) derived from 2, the fourth quartet (4), and the basal entoblasts (*D*, *C*). *D*, delamination of mesoblast in the fourth division of 2 (shaded), from the lower pole, showing the basal quartet of entomeres (*A*–*D*), and the two somewhat unequal cells ($4d^1$, $4d^2$) formed by the vertical division of the posterior cell of the fourth quartet. *E*, posterior view of ensuing stage, showing the two posterior mesoblast cells (shaded) lying in the interior, and a marked inequality between ($4d^1$ and $4d^2$). *F*, later stage; multiplication of the mesoblast-cells (shaded), equality of $4d^1$ and $4d^2$, as in *Discocœlis*.

that they suggest a number of highly interesting problems for future research. As regards the third quartet I was unable to find in *Leptoplana* any evidence that it gives rise to mesoblastic elements such as we should expect to find in the *Turbellaria* in view of the formation of ectomesoblast from this quartet in *Physa*, *Planorbis*, *Podarke*, and probably in *Aricia*. As far as I could find, the third quartet gives rise only to ectoblast cells at the lip of the blastopore (Fig. 4), and Lang's results seem to me inconclusive on this point. Only renewed researches can determine whether this difficulty be real or only apparent. In the mean time it would be well not to lose sight of the fact that the polyclades cannot, of course, be the actual ancestors of the annelids and mollusks, and that the cleavage in the former may differ very considerably from the common ancestral type. A natural hypothesis is that in the ancestral mode of development all of the first three quartets gave rise both to ectoblast and to mesoblast, and that in all the existing forms the mesoblast formation has been lost in the first quartet and variously reduced or entirely suppressed in one or both of the two succeeding quartets. I think, therefore, that we need not hereafter be surprised to find the formation of ectomesoblast from more than one of the first three quartets, whether in the *Turbellaria* or in the higher forms.

It is when we attempt to bring the foregoing considerations into relation with the history of the fourth quartet in annelids and mollusks that we arrive at a far more serious difficulty; but we can hardly regret a difficulty that is so suggestive of further research. In the polyclade the fourth quartet is relatively very large, the basal quadrants being correspondingly reduced (Fig. 4). All of the eight cells formed give rise, as far as known, to entoblast only. In the annelids and mollusks, on the other hand, only three cells of this quartet — anterior, right, and left — are purely entoblastic, while the fourth, or posterior, cell ("d^4") divides into symmetrical halves to form the "primary mesoblasts," or pole-cells, from which arise the two mesoblast-bands characteristic of these groups (Fig. 2, *A*). Now, in comparing this mode of development with that of the polyclade, we must choose between the following alternatives. *Either* the mesoblast of the annelid or mollusk, as a whole, corresponds with that of the polyclade — in which case we must assume that in the course of the phylogeny the posterior cell of the fourth quartet has gradually taken upon itself more or less completely the mesoblast formation formerly occurring in the second or third quartet; *or* the mesoblast of the polyclade has dwindled away, perhaps has even disappeared, in the higher forms, where it is represented only by the ectomesoblast, its place having been taken, through a process of substitution, by the mesoblast-bands derived from the fourth quartet. To vary the statement we must assume that a substitution has taken place either in the cell-mechanism by which the mesoblast is formed or in the mesoblast itself, and upon our choice between these alternatives depends the entire point of view from which we regard cell-lineage.

Now, it must be admitted, forthwith, that we have not at command sufficient data to give any certain answer to this question, and we should be careful not to draw premature conclusions in a matter which involves further consequences of such importance. But there are a number of well-ascertained facts drawn from widely diverse sources that point towards the second of the above alternatives; *i.e.*, the view that the mesoblast-bands of the annelid or gasteropod are not as such represented at all in the polyclade, but, phyletically considered, are neomorphs which have more or less completely replaced the ancestral mesoblast. This evidence may be arranged in three lines: —

1. As a result of exact and thorough studies upon the histology and larval development of the annelids, Eduard Meyer was several years ago led to the conclusion that the mesoblast-bands, both in origin and in fate, differed widely from the scattered larval mesenchyme-cells, though the lineage of the latter was then unknown. Developing this idea, Meyer was led to the remarkable conclusion that the mesoblast-bands of the higher types represent the paired *gonads* of the ancestral form — a view nearly related with the earlier one of Hatschek, that the primary mesoblasts were originally eggs, which, in the course of the phylogeny, became in part transformed into peritoneal and other somatic cells, and in part remained as germ-cells. Thus the original mesoblast — *which Meyer definitely compared*

with that of the Turbellaria—was gradually replaced, though still persisting in a reduced form as the larval mesenchyme.

I would not at present urge the acceptance of this daring hypothesis; but in the light of later research it has become highly significant, and whether true or false is of great interest as giving a clear picture of how such a process of substitution may have been possible.

2. In the second line of evidence lies Lillie's discovery that the ectomesoblast of *Unio* (derived from a^2) gives rise to purely larval transitory structures; namely, to the adductor muscle and the scattered contractile myocytes of the *Glochidium* larva. In the annelids, too, the same conclusion seems probable, and my friend Professor Treadwell informs me that in *Podarke* there is every reason to believe that the ectomesoblast (derived from a^3, c^3, d^3) is entirely devoted to the formation of the ring-muscle and myocytes of the trochophore, which apparently take but an insignificant part, if any, in the building of the adult body. This result tallies with the view that the ectomesoblast formation in the higher types is a reminiscence of the ancestral process still existing in the polyclade, but in the higher forms relegated to the early stages, and even in them is more or less reduced.[1]

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[1] Eisig has very recently (*Mitth. Zool. Station*, Neapel, xiii, 1, 2, 1898) published the results of a study of the cell-lineage and later development of *Capitella*, which are totally at variance with the view here suggested, and the facts on which it is based. Broadly speaking, his results exactly reverse those of all the authors cited above, the mesoblast-bands ("Cœlomesoblast") being derived from the third quartet ($c^{3.1}$ and $d^{3.1}$), while the larval mesoblast ("Pædomesoblast") arises from a portion of M (d^4), the remaining portion giving rise to ectoblast. If well founded, this result is not only fatal to the view I suggest, but is, I believe, nothing less than a *reductio ad absurdum* of the whole cell-lineage program, regarded as a method of morphological research. No one will lightly call in question the results of so conscientious and eminent an observer; and they must be regarded as by far the most serious obstacle that the morphological study of cell-lineage has thus far encountered. I will not attempt to explain away this adverse evidence, based on so prolonged and thorough a research. It should not be forgotten, however, that, as Professor Eisig is himself careful to point out, the nature of the material has forced him to contend with great difficulties, since the eggs are normally distorted by pressure (a factor which, as I have experimentally shown in *Nereis*, may greatly alter the form of cleavage) between the membranes of the tube; and, further, the development cannot be continuously followed in life. A result, based on this material, which stands in such flat contradiction to what is known in other and more favorable forms, must await the test of further research.

3. In the third line lies the evidence, recently obtained, that the pole-cells or teloblasts of the mesoblast-bands of the annelids and mollusks are to be regarded as derivatives of the archenteron, and hence differ wholly from the ectomesoblast in their relation to the primary germ-layers. Kowalevsky, the discoverer of these teloblasts, expressed the opinion, more than twenty-five years ago (1871), that they were to be regarded as derivatives of the archenteron; and a large number of later workers from the time of Rabl (1876) have accepted his view, though only very recently has the full strength of the evidence been developed. In the first place, it was shown through the studies of Rabl, Blochmann, and later workers, that while the posterior cell of the fourth quartet gives rise to the mesoblastic pole-cells the other cells are purely entoblastic. In the second place, the recent studies of Conklin and myself have shown that even the posterior cell of the fourth quartet (d^4) may contain entoblastic as well as mesoblastic material. I showed several

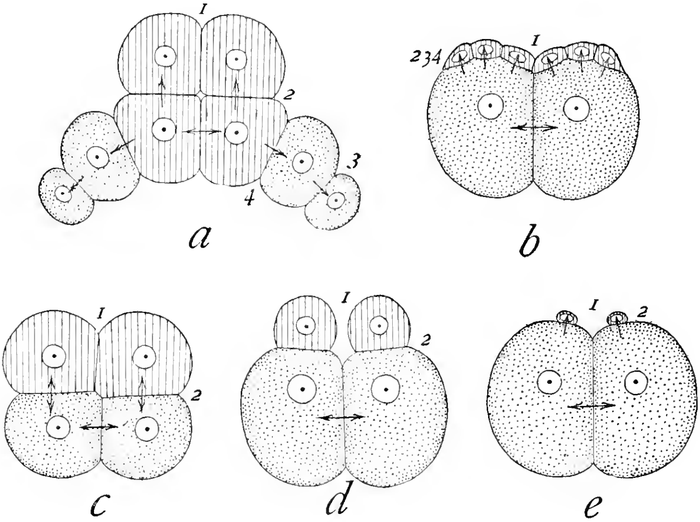

Fig. 5. — Diagrams comparing the early divisions of the posterior cell of the fourth quartet (d^4 or M) in *Crepidula* (*a*), *Nereis* (*b*), *Clymenella* (*c*), *Unio* (*d*), and *Aricia* (*e*). The numerals show the order of division. Cells destined to form entoblast (their fate as actually observed in *Crepidula* and *Nereis*, but only inferred in the other cases) ruled in parallel lines, mesoblast dotted. After the divisions here shown, the symmetrical mesoblast-bands are formed from the dotted cells.

years ago that in *Nereis* each of the cells into which d^4 divides buds forth several small cells (Fig. 5, *b*), which do not enter into the mesoblast-bands, though I did not correctly determine their fate. More recently Conklin was able to show that a similar process occurs in *Crepidula* (Fig. 5, *a*), and that the cells thus formed are *entoblast-cells* which enter into the formation of the archenteron. On reëxamining the matter in *Nereis* I found the clearest evidence that the same was true here. In both these cases, therefore, the posterior cell of the fourth quartet is of mixed character, and divides into two mesentoblasts, each of which first gives rise to a number of entoblast cells (two in *Crepidula*, four or five in *Nereis*), the residue constituting the mesoblast. In both these forms, therefore, the ectoblast (and in *Crepidula* the ectomesoblast) are first completely segregated, and the archenteron which remains gives rise to the mesoblastic pole-cells. The latter are, therefore, of entoblastic rather than ectoblastic origin, and may be designated as the *entomesoblast*.

Further examination of these phenomena brings out some highly interesting facts which seem to constitute a striking case of ancestral reminiscence in cleavage. Several years ago I found in two genera of annelids, *Aricia* and *Spio*, that the small entoblast-cells of *Nereis* and *Crepidula* (*i.e.*, those budded forth from the two mesoblasts derived from the division of d^4 or *M*) are represented by a single pair of quite rudimentary cells, scarcely larger than polar bodies (Fig. 5, *e*), which apparently take no part in the building of the archenteron, and can only be explained as vestiges or reminiscences of such a process as occurs in *Crepidula* or *Nereis*. Later researches have revealed the presence of these vestigial entoblasts in several other forms, and have shown further that they are connected by several intermediate steps with the larger functional cells found in *Crepidula*. Thus in *Amphitrite* (Mead) and *Planorbis* (Holmes) they are quite vestigial, agreeing essentially in size and origin with those of *Aricia*. In *Unio* (Lillie) they are considerably larger (Fig. 5, *d*), in *Clymenella* (Fig. 5, *c*) they are as large as the mesoblastic moiety (Mead); while in *Crepidula* (Fig. 5, *a*) their bulk surpasses that of the mesoblastic part.[1] Such a series creates a strong probability that we have before us a vanishing series like those so well known in adult organs, such as the limbs, the tail, or the teeth. Further, just as the lateral toes of the horse seem to have wholly vanished, even from the ontogeny, so the vestigial entoblasts would seem to have disappeared in some annelids and mollusks, leaving the posterior cell of the fourth quartet purely mesoblastic.

These considerations invest with a special interest the corresponding cell in the *Turbellaria* (*i.e.*, the posterior member of the fourth quartet, 4*d*); and this interest is heightened by Lang's discovery that in *Discocœlis* this cell divides earlier than the other cells of the quartet, and into equal halves which lie symmetrically at the posterior end of the embryo. These two cells thus correspond exactly in origin and position with the paired mesentoblasts of the annelids and gasteropods, and the facts naturally led to the suggestion, made by Mead, that they would perhaps be found to give rise to paired mesoblast-bands, as in the higher types. In *Leptoplana* (Fig. 4, *D*, *E*, *F*) a similar division occurs, but as far as their fate is concerned my own observations do not sustain Mead's suggestion, on the one hand giving no evidence that these cells give rise to anything other than the posterior cells of the archenteron, on the other showing that they are often unequal or asymmetrically placed (Fig. 4, *D*, *E*) and only rarely conform to Lang's scheme (Fig. 4, *F*). If, therefore, the polyclades represent the ancestral type in this respect, we must conclude that the entomesoblast was a later development. The remarkable fact is that, if such has been the case, the new mesoblast-formation has been fitted, as it were, upon an old form of cleavage occurring regularly in *Discocœlis* and occasionally in *Leptoplana*. The two symmetrical posterior entoblast-cells of the polyclade might thus be conceived as the prototypes of the primary mesoblasts or mesentoblasts of the higher forms, which in the course of the phylogeny undertook the formation of mesoblastic as well as of entoblastic elements.[2] The old building pattern was still

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[1] I am here placing my own interpretation on Mead's and Lillie's observations.

[2] Lang has pointed out a motive for this form of cleavage in the polyclade, correlating the early and symmetrical division of d^4 with the posterior bifurcation of the gut.

retained but adapted to a new use, precisely as has been the case with the evolution of larval or adult organs, such as the branchial or aortic arches and the limbs. As the change progressed the posterior cell of the fourth quartet became more and more strictly given over to the formation of mesoblast, its entoblastic elements becoming correspondingly reduced to truly rudimentary or vestigial cells (*Aricia*, etc.), or finally, perhaps, disappearing wholly.

I have endeavored to place these special conclusions in strong relief, not because they can yet be accepted as demonstrated, — and it is quite possible that some other interpretation may yet be placed upon some of the facts, — but because they seem to me highly suggestive of further research in the field of cell-lineage. There are among them two general considerations on which I would lay emphasis.

First, the study of cleavage or cell-lineage in the case of these groups raises a number of highly interesting and suggestive questions in pure morphology. If the mesoblast-bands are a new formation, what is the motive, so to speak, for their origin? Did they perhaps arise through the development of a new body-region, or a new growth-zone, or budding-region from the posterior part of the ancestral body, as has been assumed by Leuckart, Haeckel, Hatschek, and Whitman in explanation of metamerism? Is the body of the turbellarian homologous to the entire body of an annelid or mollusk, or does it represent only the head or the larval body, to which a trunk-region is afterwards added? What is the relation of the entomesoblast to the archenteric pouches of the enterocœlous types? How do the above results harmonize with the general doctrine of development by substitution? These are examples of some of the morphological questions suggested by the general inquiry. They are admittedly of a highly speculative character, and I, for one, am not prepared to give a positive answer to any of them. But the mere fact that morphological questions of such character and scope are inevitably suggested by studies in pure cell-lineage shows that such studies must not be passed over by the morphologist as having no interest or value for his own researches.

Second, the phenomena we have considered seem to leave no escape from the acceptance of ancestral reminiscence in cleavage, with all that that implies. That the rudimentary entoblasts of *Aricia* or *Spio* are such reminiscences of former conditions seems almost as clear as that the mammalian yolk-sac or the avian primitive streak are such. The formation of the ectomesoblast in annelids and mollusks is nearly if not quite as strong a case. Both these are processes that appear to be vestigial, or, at any rate, approach that character. But the evidence of genetic affinity is no less clearly shown in processes that are not vestigial, such as the formation of the ectoblast in *Turbellaria,* annelids, and gasteropods or lamellibranchs, from neither more nor less than three quartets of micromeres, or in the origin of the archenteron from the fourth quartet with the remains of the basal quadrants. Between the annelids, gasteropods, and lamellibranchs a far more precise and extended series of resemblances exists. The question has been much discussed of late whether such resemblances can be called homologies. Probably no one will deny that the ectoblast-cap, arising from twelve cells, is as a whole homologous in the annelid and the gasteropod embryo. Are the individual micromeres respectively homologous? In the present state of our knowledge this is a question of name rather than of fact; for homologies only gradually emerge during development from their unknown background in the egg. It is for this reason that, as I have urged in a preceding lecture, *the ultimate court of appeal in this question lies in the fate of the cells.* If the structures to which they give rise are homologous, I can find no logical ground for refusing the claim to the cells from which they arise. Furthermore, this homology must be irrespective of the origin of the cells, just as the ganglion of a bud-embryo of *Botryllus* is homologous with that of an egg-embryo in the same form, despite the total difference of origin in the two cases. When, however, we find that the homologous protoblasts or parent-cells have the same origin as well as the same fate, the homology becomes the more striking; and it is in the determination of common origin as well as common fate, as has been done in so many cases, that the principal significance of recent work in cell-lineage seems to me to lie. Some of the objections urged against the reality of cell-homology have, I think, arisen through a failure to recognize among cell-homologies the same distinction between complete and incomplete homology that was long ago urged by Gegenbaur in the case of organ-homologies. The posterior member of the fourth quartet in annelids, for example, is in a broad sense homologous throughout the group; but the homology is probably not an absolute or complete one, since this cell may contain functional entoblast (*Nereis*), rudimentary or vestigial entoblast (*Aricia*), or apparently in some cases no entoblast, as I have described in *Polymnia*. Again, the acceptance of cell-homology does not, I think, carry with it the necessity of finding a homologue for every individual cell throughout the ontogeny; for in the case of later structures no one demands or expects that, in the comparison of related forms, an exact equivalent shall be found for every subdivision of homologous nerves or blood-vessels or sense organs. Finally, the fact that cleavage *may* show no constant or definite relation to the adult parts — as is the case in the teleost fishes — does not alter the equally indubitable fact that cleavage often *does* show such a constant relation. The probability that the *Nauplius* larva is not a true ancestral form does not come into collision with the probability that the ascidian tadpole is such a form. How far in the course of phylogeny the ontogeny has adhered to its original type and retained the same relation to the adult parts is a question which stands, as far as I can see, both *a priori* and *a posteriori* on essentially the same basis, whether it be applied to the cleavage or to the later stages. Let us not forget the difficulties that still beset us in the application of the biogenetic law to the larval stages and to general organogeny, and let us not make a greater demand in this regard upon cell-lineage than on other lines of embryological research. The time has not yet come for a last word on this subject, and we shall probably have to await the result of much more extended research before a satisfactory point of view can be attained.

THIRD LECTURE.

ADAPTATION IN CLEAVAGE.

FRANK R. LILLIE.

I. INTRODUCTION.

IT has happened, very naturally, that writers on the subject of cell-lineage have laid special emphasis on the resemblances, which are nothing short of marvellous from the older points of view, between the cleavage of the eggs of even widely separated forms. Over and over again it has been demonstrated that in gasteropods, lamellibranchs, annelids, and turbellarians, the ectoderm is made up of three quartets of cells, formed from the first four cells; that the fourth product of the left posterior macromere contains the mesoblast, excepting in the turbellarians; that the turbellarian mesoblast is represented in the other groups; that the four basal cells, after the separation of the ectoderm and mesoblast, form endoderm; that the ectoderm of the entire trunk is derived from d^2 (speaking technically), and that there is a wide-reaching sameness in the cell-lineage of the prototroch, cross, and of other larval organs. Ancestral reminiscences even have been discovered in the cleavage (E. B. Wilson, 14). I do not in the least underestimate the immense value of these results, which form one of the most brilliant and interesting chapters in modern embryology. But the tendency to schematize has naturally arisen, and one of the most instructive aspects of cell-lineage is thus lost sight of, namely, *the special features of the cleavage in each species, which are, I believe, as definitely adapted to the needs of the future larva as is the latter to the actual conditions of its environment.*

This principle is meant to apply only to the "determinate type of cleavage" (Conklin), in which larval, and hence adult structures, can be shown to have a definite cell-lineage extending back to the unsegmented ovum. The illustrations and arguments will be drawn, first, from the cleavage of the egg of a fresh-water lamellibranch, *Unio,* which I have specially studied, and then, to secure a broader foundation in fact, some significant variations in the cleavage of the eggs of annelids will be considered. This theory was first used, I believe, for the explanation of determinate cleavage in my papers on the "Embryology of the *Unionidæ*" (6 and 7), where I pointed out that the size of the cells and the rate and direction of their cleavages "are ruled by the needs of the embryo," and concluded: "The peculiarities of the cleavage in *Unio* are but a reflection of the structure of the glochidium, the organization of which controls and moulds the nascent material." In his lecture on "Cleavage and Differentiation" (*Biological Lectures,* 1896–97), Conklin has maintained a similar view, *viz.,* "that all differential cleavages . . . are directly and causally related to the uses to which these cells are put." In the second part of this paper the retrospective significance of this principle will be considered in the light of some observations on the maturation, fertilization, and first two cleavages of the egg of *Unio.*

If we are to understand the special features of the cleavage in *Unio,* which are, I believe, purely adaptive, it will be necessary, first of all, to describe the peculiar larva. The glochidium (Fig. 1) possesses two shell-valves, shaped something like the bowls of two spoons cut across near their bases, and united by a central hinge. The curvature of the anterior margin of each valve is somewhat greater than that of the posterior margin, and each lateral angle is provided with a strong hinged hook, which is secondarily toothed in *Anodonta* on its outer face. The valves may be closed with great force by a single strong adductor muscle, the main bulk of which is somewhat anterior to the middle of the body. The interior of the spoon-shaped valves is lined by the larval mantle, composed of very large five or six sided cells, in the exact centre of which is the opening of a long tubular gland, the thread-gland, coiled several times round in the right valve of the shell. The secretion of this gland takes the form of a long thread, which can, apparently, be respun when lost, and which floats freely in the water. There are four tufts of sense-hairs on each side of the larval mantle, three arranged in the form of a right-angled triangle just within the outer angle of each valve, and one on either side of the middle line, a trifle behind the opening of the thread-gland.

Now all of the structures mentioned thus far, the shell, the larval mantle, the adductor muscle, the thread-gland, and the tufts of sense-hairs, are purely larval organs, destined to form no part of the adult, but to degenerate. They are already old when the life of the individual has hardly begun, and they form by far the greater bulk of the larval body. The embryonic material, from which the body of the adult is to develop, is all included within a small area (*E. A.*) just in front of the posterior angle of the valves of the shell. This area is only about one-third of the length of the glochidium, and less than a third of its width, when the valves are expanded; yet within it we find the rudiments of the intestine with minute liver-diverticula and end-gut well marked but no opening to the exterior, of the mesoblastic structures, and of the foot, mantle, and gills.

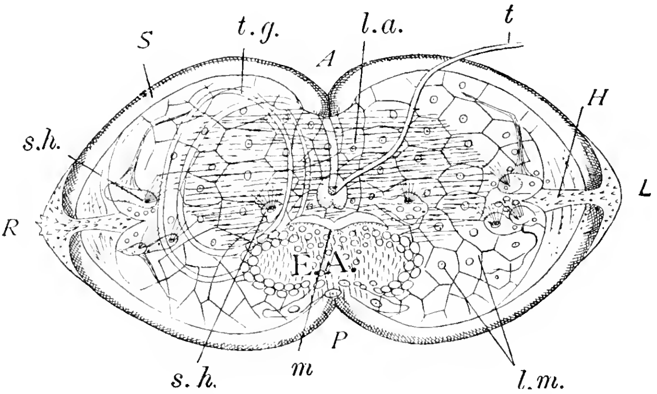

FIG. 1.—Glochidium of *Anodonta*, one of the *Unionidæ*, lying on its dorsal surface, with the shells expanded so as to show the soft parts. *A*, anterior; *P*, posterior; *R*, right; *L*, left; *S*, shell; *H*, hook; *l. m.*, larval mantle; *l. a.*, larval adductor muscle; *t.g.*, thread-gland; *t.*, thread; *s.h.*, sensory hairs; *m.*, mouth; *E. A.*, embryonic area.

All these parts are entirely functionless, and great only in potentialities.

The glochidium is a larva highly specialized for a peculiar mode of development. When the larva is extruded into the water by the mother-clam, it falls to the bottom and lies there with its thread floating in the water, violently snapping its valves together at the least disturbance. It is really a most sensitive little creature, and so readily perceives any fish swimming by. If it be so fortunate as to have its thread caught on the fish, it at once attacks it with its hooks. It is, of course, only on the unprotected parts of the fish, such as the fins or the gills, that the larva is able to make any impression; and it is stated that the larvæ of *Anodonta* usually become parasitic on the fins, and those of *Unio* on the gills. The irritation caused by the presence of the larva sets up a proliferation of the epidermis, which speedily encloses the little parasite in a sort of cyst, within which the later development takes place.

II. Cleavage of the Egg of Unio.

Adaptation in cleavage can manifest itself only in the three possible modes of cleavage variation, which are, as has been pointed out by Mead, Jennings, zur Strassen, and others, first, differences in the rate of cleavage of cells; second, differences in size; and third, differences in the direction of the cleavage or position of the resulting parts. Let us now examine the cleavage of the egg of *Unio* with these general principles and the structure of the larva in mind. I may as well state at once that I regard the *general form* of the cleavage as inherited from a long series of ancestors, extending back, probably, to the *Turbellaria.*

The general plan of the cleavage is the same as in annelids, *Turbellaria*, and most mollusks, described in other lectures contained in this volume. From the first four cells, separated by meridional planes, the ectoderm is separated by three slightly oblique cleavages horizontal in their general direction, the first being dexiotropic, the second læotropic, and the third dexiotropic again. A fourth division of the left posterior macromere (basal cell) forms the mesoblast cell. (See Figs. 2–5.)

The first cleavage is meridional and very unequal, although the distribution of the yolk throughout the cytoplasm is perfectly uniform in the form of small granules (Fig. 20). The larger cell marks the position of the posterior end of the embryo. This cell divides before the smaller one, and unequally, the larger product again being posterior in position. The smaller cell of the two-celled stage then divides somewhat unequally, so that its larger product lies on the left side of the embryo. Thus we reach a four-celled stage (Fig. 2) composed of one very large cell, *D*, posterior in position, and three smaller cells, of which the left one, *A*, is the largest. The anterior and posterior cells, *B* and *D*, meet in a broad cross-furrow at the animal pole (Fig. 3), the cross-furrow at the vegetative pole being much smaller, contrary to the general rule in eggs of this type of cleavage. Thus the first two cleavages illustrate the three possible modes of variation, *viz.*, inequality of the cleavage products, difference in the rate of cleavage, and difference in the position of the cells, indicated by the broad cross-furrow at the animal pole. What is the meaning of these differences?

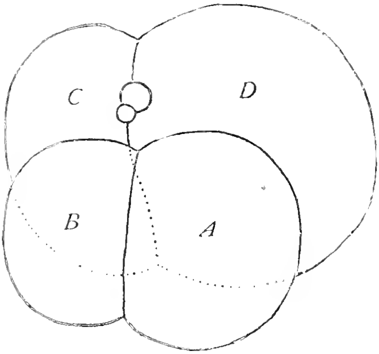

FIG. 2. — Four-celled stage of *Unio complanata* seen obliquely from the left side and above.

a. Relative Rates of Cleavage of the Constituent Cells. — Let us place side by side a table of the constitution of the thirty-two-celled stage of the egg of *Unio* and of an ideal ovum following, in direction of cleavage, the ordinary spiral type, with equal cleavage throughout, and the same rate of division in all the cells.

	IDEAL OVUM.	UNIO.
First generation of ectomeres	16	10
Second generation of ectomeres	8	13
Third generation of ectomeres	4	4
Fourth division of *D* (mesoblast cell)		1
Entomeres	4	4
	32	32

It is plain that as regards the relative rate of cleavage of the constituent cells, the egg of *Unio* departs very far from the ideal condition. Let us attempt to analyze this and see if these variations in the rate of cleavage are adaptive or not. The first thing we notice is that the number of the first generation of ectomeres is ten instead of sixteen; this means that these cells have divided more slowly than the average rate. Now in most mollusks and annelids they form the prototroch and the entire region in front of it, with the apical plate in the centre. But in *Unio* this region is degenerate, and I have never been able to find any trace of prototroch or apical sense-organ. It is plain, then, that the slow rate of cleavage is anticipatory of this, and is, in this sense, an adaptive modification. But why ten cells in place of eight? This means that two of the cells, c^1 and d^1, of this group have divided once oftener than their fellows. These cells occupy the posterior portion of the upper hemisphere, and later enter into the formation of the head-vesicle, and I believe that their more rapid cleavage is an early indication of their subsequent fate.

In the second generation of ectomeres we have a very different set of conditions as to number; the ideal number of cells of this generation in the thirty-two-celled stage is eight, but in *Unio* thirteen. This increase above the ideal number is due almost entirely to the rapidly succeeding divisions of one cell of this generation, d^2, but in part also to another, a^2. The other two cells, the anterior and the right members of the quartet, fall in with the ideal scheme, each having divided once. In this stage d^2 has broken up into six cells, a^2 into three. Is there anything adaptive in this hastening of the process of cleavage? d^2 is the cell from which the entire ectoderm of the trunk region is formed. The most characteristic thing about this region in the embryo of *Unio* is the relatively enormous size and very early formation of the shell-gland, and we have seen that the shell of the glochidium is of very special importance to it, not only as an organ of defence, but also of offence. Is it not wonderful that this important part should be represented, not only by the largest, but also by the most rapidly segmenting cell in the embryo, as though the egg knew what was required of it, and acted accordingly? Can we deny that the rate of cleavage of this cell is an adaptive feature of the cleavage? The other cell of the second generation that divides more rapidly than the average rate is $a^{2 \cdot 2}$, or *Y*, the lower product of the first division of a^2. In the thirty-two-celled stage (Fig. 5) this cell already projects some distance into the segmentation cavity; after budding off two more small cells at the surface, it withdraws entirely within the segmentation cavity, and divides equally into two cells, which arrange themselves symmetrically on either side of the middle line. From these cells of the *larval mesoblast* the adductor muscle of the glochidium and certain isolated myocytes of the cleavage cavity are derived. Now the adductor muscle is an extremely large organ, of the utmost functional importance to the larva, and the variation in the rate of cleavage of the cells which are to form it appears to be adaptive.

The third generation of ectomeres possesses the normal number of cells for this stage, but it is interesting to notice that the posterior member, d^3, is formed much before the others, owing to the tendency to more rapid division of the basal cell from which it arises. This cell, *D*, has also undergone another division, in excess of the divisions of the other basal cells, giving rise to the mesoderm proteloblast, d^4 or *M*.

To summarize: groups of cells and even single cells vary greatly in their rates of segmentation. Each acts as though animated by an independent force. Moreover, these differences possess prospective significance, looking forward to the final outcome; and this phenomenon may fairly be called *adaptation in the rate of cleavage*.

But the objection may be raised: granted that the rate of division varies in these cells, and that those segmenting more rapidly form the earliest functioning parts, is it not possible that the variation is induced from without, and that this determines the subsequent fate? So far as we know, the only external factors which influence the rate of cleavage are temperature, chemical constitution of the medium, and perhaps some other general conditions of the environment, which must act uniformly on the whole egg, and hence cannot be held accountable for differences in the rate of cleavage of constituent elements of the same egg. In addition to these factors, the presence of a larger proportion of yolk in certain cells may determine a slower rate of cleavage. But in the egg of *Unio* the yolk is distributed with perfect uniformity among the cells, so that each possesses practically the same relative quantity. Is it not, then, the absolute amount of yolk that determines the rate? No, unless one is willing to adopt the hypothesis that yolk tends to hasten the cleavage, for it is the largest cell in the egg that segments most rapidly. However, there is no constant relation between size and rate of cleavage, so even such an inverted hypothesis will not avail. There is, in fact, no escape from the conclusion that the factors determining differences in the rate of cleavage of the separate cells or definite cell-groups are of the same nature as those that determine differences between ova of different species, *viz.*, purely constitutional causes. In a very real sense, each cell in the segmenting egg of *Unio* acts, in respect to rate of cleavage, like an *ovum in ovo*.[1]

b. Variations in Size. — Although difference in size of daughter-cells in normal cleavage generally indicates difference in quality or constitution, it is not always safe to conclude that daughter-cells of the same size necessarily possess the same constitution. In other words, a division may be differential, even though the resulting cells are of the same bulk. The adaptiveness of determinate cleavage is illustrated in a manner even more striking than in differences of rate, by the inequality of certain divisions. I believe that it can be shown that the relative sizes of the cells in the early cleavages of the egg of *Unio* is adapted to the size and time of development of the larval organs. The first two cleavages are unequal with the result that two cells of the four-celled stage are larger than the two other equal ones. The largest of all contains the material of the two somatoblasts, and the next largest the material of the larval mesoblast.

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[1] I believe that the entire egg is an undivided organism, and not a heap of equivalent cells, each of which may occupy any position in the whole, as Hertwig and Driesch have maintained. The above statement must be understood in this sense.

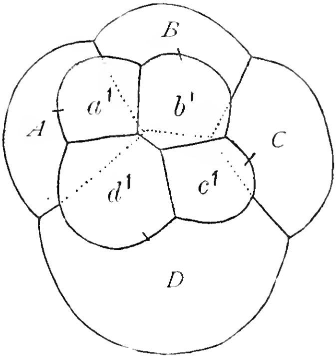

FIG. 3.

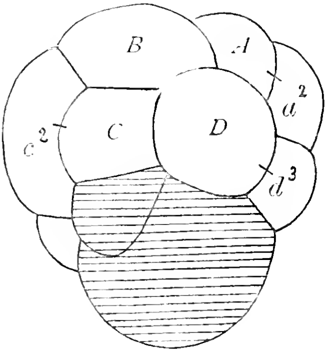

FIG. 4.

FIG. 3.—Eight-celled stage of *Unio complanata* from the animal pole.

FIG. 4.—Eighteen-celled stage of *Unio complanata* from the vegetative pole. The two cells ruled with horizontal lines are products, the first somatoblast, $d^2 = X$, which forms the trunk, including the shell-gland and foot of the larva.

Let us now look at the first four divisions of *D*, the largest cell of the four-celled stage (Fig. 2). Its first division (Fig. 3) is very unequal, and the smaller product, perhaps not one-tenth of the whole, lies nearer the animal pole; it is one of the first generation of ectomeres. The second division is likewise unequal (Fig. 4), but here the relations are reversed, for two-thirds, at least, of the substance of the cell passes into the upper product, the first somatoblast, d^2 or X; the third division is likewise unequal (*D* and d^3, Fig. 4), and this time again the smaller product is uppermost, forming one of the relatively unimportant third generation of ectomeres. The fourth division, finally, is extremely unequal (Fig. 5), only a minute portion of the cell remaining at the lower pole, while the remainder forms the second somatoblast, d^4 or *M*, the proteloblast of the mesoderm. In each of these unequal divisions there is a manifest adaptation, the

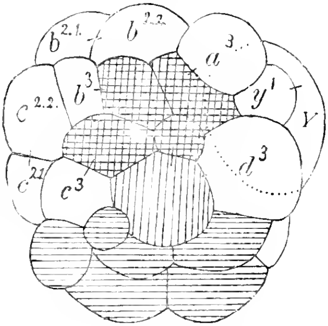

FIG. 5.—Thirty-two-celled stage of *Unio complanata* from the vegetative pole. The separation of the germ-layers is practically complete in this stage. Products of first somatoblast, $d^2 = X$, with horizontal lines: second somatoblast, $d^4 = M$ (mesoblast cell), with vertical lines: endoderm cells with both vertical and horizontal lines. $Y = a^{2.2}$ larval mesoblast.

great bulk of the material passing into the two somatoblasts. In the thirty-two-celled stage (Fig. 5) the largest cells are those of the second generation of ectomeres, and this finds a sufficient explanation in the fact that the material of the first somatoblast, of the larval mesoblast, and of the larval mantle, is found in these cells. The relatively small size of the first and third generations of ectomeres and of the endoderm-cells is correlated with the relatively small part that they play in the upbuilding of the larva. The mesoderm proteloblast d^4 is actually smaller than the larval mesoblast Y (Fig. 5) because of its deferred functional activity.

If there were any other explanation of these differences in size of the cleavage products than this seemingly teleological one, it might seem to simplify the problem of development. However, all theories that seek to explain inequalities of cleavage by the presence of yolk or the action of any known external force seem inadequate to explain the unequal cleavages in the egg of *Unio*.

c. Variations in the Direction of the Cleavage-Planes. — The early divisions follow the law of alternating spiral cleavage (Kofoid) for some time; and I shall not stop to consider these, although we are far from an adequate mechanical explanation of this law. I shall take up, instead, certain special divisions, which illustrate in a more striking manner the adaptiveness of variations in the direction of cleavage. The first somatoblast is established in the nine-celled stage by the second division of D; it is much the largest cell in the embryo, as we have seen, and it goes through with an exceedingly characteristic series of cleavages. It first buds off a small cell, x^1, on the right side of the vegetative pole, then another small cell, x^2, symmetrically placed on the left side, then a third small cell, x^3, in the middle line, but towards the animal pole, then a fourth small cell, x^4, in the middle line towards the vegetative pole (see Fig. 5); it then divides equally and bilaterally, and each half buds forth another small cell, x^5, behind x^4 on the vegetative pole. I do not know the fate of the cell x^3, budded forth towards the animal pole. However, the other small products form the rudiment of the foot, which is an exceedingly small organ in the larva; and the large cells, which henceforward divide equally, the immense shell-gland. Can one imagine a more manifest adaptation? The small cells are placed just where they are needed, and are of the proper dimensions. Is there any other reason why the somatoblast should not have divided equally from the first, and the cells lying in one position have gone into the foot, and the rest into the shell-gland? If this were to happen, there would be no indication of adaptiveness in the cleavage, for then it might seem possible that, simply because of their different positions and conditions, part of the cells formed the foot and part the shell-gland.

d. To summarize.— In the cleavage of the egg of *Unio* there are marked variations in the size of the cells and in the rate and direction of their cleavages; in every case, these possess prospective significance, and by means of them the organism is able, so to speak, to realize, in the most direct manner possible, on its available capital, the substance of the egg. *To this principle I have given the name of adaptation in cleavage.*

III. Adaptation in Cleavage in the Annelids.

In the annelids, as in the mollusks, the entire ectoblast of the trunk is segregated in a single cell, d^2 or X, known as the first somatoblast, and the mesoblastic germ-bands in the second somatoblast, d^4 or M. Now the cell-lineage of a great many annelids is known through the studies of Whitman (11), Wilson (13 and 14), Mead (9), Eisig (5), Treadwell (10), Child (1), and others. It is possible to arrange these annelids in a series according to the relative size of their somatoblasts. First there are forms like *Polygordius*, *Lepidonotus*, *Podarke*, *Hydroides*, and *Eupomatus*, with equal cleavage, in which the somatoblasts do not differ in size from the other cells of the same quartets (Fig. 6). Then in the order of increasing relative size of the somatoblasts come *Amphitrite* (Fig. 7) and *Chætopterus*, *Arenicola*, *Nereis limbata* (Fig. 8), *Clymenella* (Fig. 9), *Capitella*, *Aricia* (Fig. 10), *Scolecolepis* and *Spio*, *Nereis dumerilii*, and finally, *Clepsine* (Fig. 11). Now all of the forms with equal cleavage, although most widely separated in relationships, possess trochophores characterized by an almost equatorial prototroch, a very large exumbrellar region, and relatively extremely slow development of the trunk. I merely allude to this here, because in another lecture of this series the causes of equal cleavage in the annelids are considered in detail by Mr. Treadwell. At the other end of the series comes *Clepsine* with its huge somatoblasts, in which the parts characteristic of the trochophore are reduced to a mere rudiment, and the entire trunk develops on the surface of the egg. Between these two extremes come the other forms mentioned, and it may be said in general of them, that there is a gradual acceleration in the time of development of the trunk very nearly in proportion to the increase in relative size of the somatoblasts. Treadwell has elsewhere called attention to part of this series, and has come to the conclusion that "the extra amount of material stored in cell *D* of *Nereis*, *Amphitrite*, etc., is in some way related to the need for an extra amount of somatic and mesoblastic material in the young larva."

IV.

But in order to show that the adaptation need not run in the one direction of the trunk, let me cite one more case, which I am able to add through the kindness of Dr. E. B. Wilson, from some unpublished observations of his on the cell-lineage of a Nemertean. Here the four upper cells of the eight-celled stage are larger than the four surrounding the vegetative pole, the only case of this sort known, I believe. We should expect on *a priori* grounds that the resulting larva would possess a large helmet in front of the ciliated band, which marks the posterior boundary of the products of the first quartet of ectomeres, with a hardly developed trunk region and a rudimentary archenteron. Such is, in fact, the case. The enormous development in the pilidium of the exumbrella is predelineated in the unique third cleavage.

I do not believe that adaptation is one whit less far-reaching in the cleavage than in the larva. In fact, if the principle which I am defending be correct, the two must be coextensive; that is to say, adaptation in cleavage is no more caused by

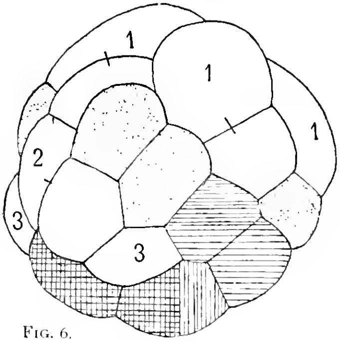

FIG. 6.

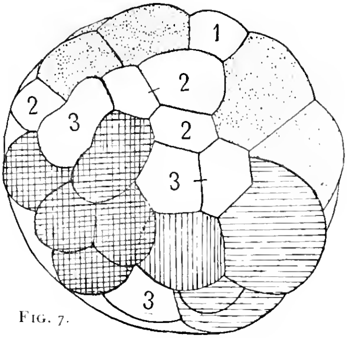

FIG. 7.

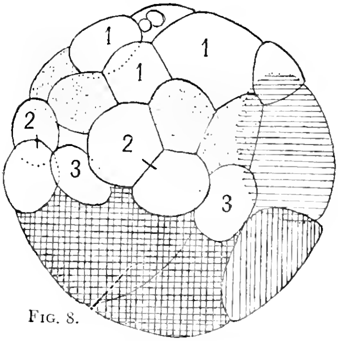

FIG. 8.

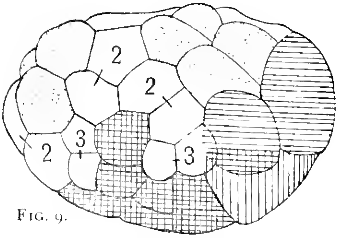

FIG. 9.

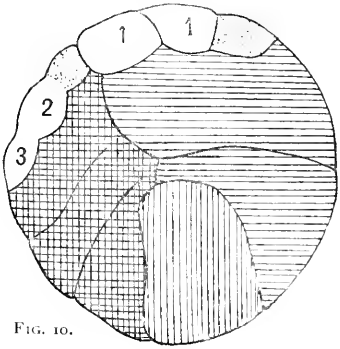

FIG. 10.

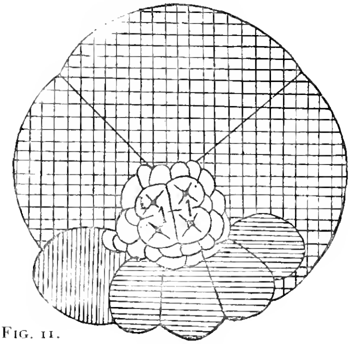

FIG. 11.

FIGS. 6–11. — Cleavage of a number of annelids, showing progressive increase in the size of the trunk-forming cells, *viz.*, the first and second somatoblasts, $d^2 = X$ and $d^4 = M$. The first somatoblast, which forms the ectoderm of the trunk, is indicated throughout by horizontal lines, and the second somatoblast, which forms the mesoderm of the trunk, by vertical lines. The endoderm-cells are ruled with lines crossing at right angles. The stippled cells are the primary trochoblasts, destined to form the prototroch. 1. First generation of ectomeres; 2. Second generation of ectomeres; 3. Third generation of ectomeres.

FIG. 6. — Thirty-two-celled stage of *Podarke* from the left side. Taken, with the kind permission of Mr. Treadwell, from one of his unpublished drawings.

FIG. 7. — *Amphitrite*, after Mead. From the left side and below.

FIG. 8. — *Nereis limbata*, after E. B. Wilson, from the left side.

FIG. 9. — *Clymenella torquata*, after Mead, from the left side.

FIG. 10. — *Aricia*, optical longitudinal section, after E. B. Wilson.

FIG. 11. — *Clepsine*, after C. O. Whitman. From the animal pole. The great size of the endoderm-cells is due to yolk. The somatoblasts far exceed in bulk all the other cells.

adaptation in the larva than *vice versa*, but the adaptation in all stages of the development is due to some common cause. It is, of course, impossible to be blind to the fact that this "explains" determinate cleavage only in a very limited sense. But if the principle serves to restrict the too universal application in the cleavage of the egg of the so-called "mechanical laws of cell-division," much will have been accomplished.

V. PROTOPLASMIC BASIS OF ADAPTATION IN CLEAVAGE.

It is possible to explain variations in size, position, and rate of cleavage of the cells, by the hypothesis of differentiation of the cytoplasm into materials of different qualities and positions (His, Whitman *et al.*).[1] A much more difficult question is immediately suggested, namely, How do these different substances arise within the cell, and how are they distributed to definite regions? Are all of the differences that exist in the thirty-two-celled stage, for instance, also to be found in different substances of the unsegmented ovum, or are they formed, wholly or in part, during the cleavage, and, if so, how?

The following observations, selected from my work on the fertilization and first two cleavages of the egg of *Unio*, seem to throw some light on this difficult problem. The spermatozoön enters always at the centre of the vegetative pole, and describes a penetration-path that carries it to a point between the centre and the periphery of the egg near the lower boundary of the upper (animal) half. The penetration-path is marked by a portion of the cytoplasm cleared of yolk-granules by the activity of the sperm-amphiaster (Fig. 12). During its penetration the sperm-head has formed some caryolymph, and hence is somewhat vesicular; but, arrived at its region of rest, it contracts into a clump of small chromatic elements. All this has taken place during the formation of the first maturation-spindle, and is completed before the cytoplasm of the animal pole begins to protrude to form the first polar globule. The sperm-amphiaster and visible penetration-path have disappeared by the time of

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[1] A theory of polarization of the ultimate morphological units would afford only a partial explanation of the differences in rate and direction of cleavage and in the size of the cleavage products.

the metaphase of the first maturation-spindle, and the sperm-nucleus remains absolutely quiescent during the remainder of

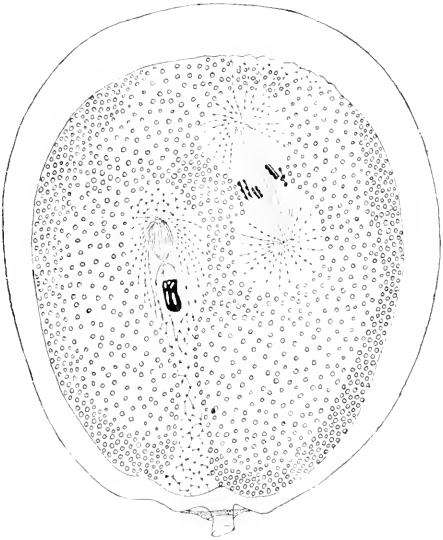

FIG. 12. — Early fertilization of the egg of *Unio complanata* in actual section. The space between the membrane and the egg proper has been reduced to economize space; the micropyle, penetration-path of the spermatozoön, sperm-amphiaster, and first maturation-spindle are shown. A large proportion of the yolk-granules is temporarily forced to the periphery.

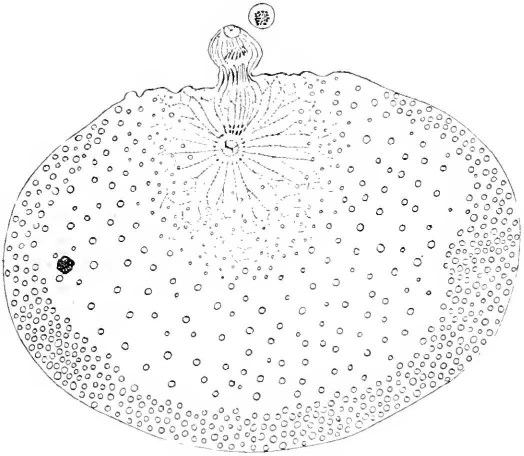

FIG. 13. — Formation of the second polar globule in *Unio*. Sperm-nucleus to the left.

the maturation processes, until the egg-nucleus begins to grow. Let us now study the egg during the final constriction off of the second polar globule. At this time (Fig. 13) the sixteen chromosomes that are to remain in the egg are closely packed together near the animal pole, and are almost in contact with the inner sphere of the aster. The inner sphere is derived from the centrosome of the first maturation-spindle (see No. 8) and has the following structure: it is a spherical body with a perfectly definite wall, containing a non-staining substance in which is excentrically placed a single centrosome united to the wall by a few strands. The cytoplasm in the immediate vicinity is arranged in the form of an aster, the fibres of which are centrally attached to the inner sphere; the first row of microsomes on the radiations of the aster bound a fairly definite outer sphere. The sperm-nucleus occupies the same position as before, and has undergone no change of structure; in Fig. 13 it is represented lying to the left. During the final stages of formation of the second polar globule, the inner sphere begins to enlarge, and the interior is occupied by a reticulum, in the nodes of which are a number of centrosome-like bodies. After the second polar globule is fully formed, the sphere enlarges with great rapidity (Fig. 14), and, as its boundary spreads out, it becomes less and less definite, until its substance merges with the general cytoplasm. During its enlargement, the interior is occupied by a vesicular substance, at the nodes of which are deeply staining granules, in no wise distinguishable from microsomes. The substance of the inner sphere is now, in fact, part of the general cytoplasm. Yolk-granules are entirely absent in it (Fig. 15), and this enables one to follow its subsequent fate for a considerable period of time.

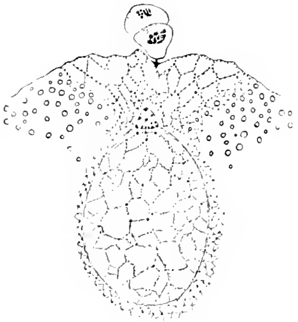

FIG. 14. — *Unio.* Small portion of the upper pole. Egg-nucleus reconstituting; sphere-substance forming.

We shall call this substance, provisionally, *sphere-substance*, following Conklin (4); it is important to remember that it is derived entirely from the inner sphere of the second maturation-spindle. Either this is the case, or else the apparent enlargement of the inner sphere at the close of the maturation is due to a wave of condensation in the cytoplasm, travelling outwards, sweeping away the yolk-granules, but leaving the cytoplasm behind. The latter theory would explain the appearance of vesicular cytoplasm within the boundary during the phenomenon, and so avoid the difficulty of explaining the extremely rapid growth of substance within the sphere; it could be compared to the wave that spreads out from a pebble dropped in the water. But it is difficult to understand the nature of such a disturbance. On the whole the *expansion* theory seems to me much more probable than the *wave* theory, and the latter is certainly untenable in the first slow stages of the process.

Another important thing to notice is that just before the expansion begins the sphere is three-quarters surrounded by the chromosomes, and I would like to hazard the conjecture that, at this time, there may be a diffusion of some chromatin substance within the sphere, the interior of which tends to stain more darkly than before. If this be actually the case, it has an extremely interesting bearing on subsequent events.

Let us now trace the further actions of the germ-nuclei and sphere-substance during the first cleavage. In general, what I propose to show is that the sphere-substance moves and elongates so as to mark out a definite horizontal plane in the egg, and that the first cleavage-spindle places itself in conformity with this predetermined arrangement.

Each of the germ-nuclei, at the close of the maturation, is a small, dense clump of chromosomes, and both begin to swell up into the vesicular form of the ordinary resting nucleus at the same time, and keep step throughout the process. This indicates a new general condition of the egg-cytoplasm; for, while we might explain the enlargement of the egg-nucleus alone as part of the usual sequence of mitosis, due to purely localized conditions of the egg-substance, we can only explain the effect simultaneously produced on both nuclei by the assumption that the entire egg-cytoplasm is entering on a new phase of its development.

The first movements of the germ-nuclei begin after each has enlarged considerably. In its early movements the egg-nucleus is preceded by the sphere-substance, which moves towards the centre of the egg, and *always towards the side opposite to that in which the sperm-nucleus lies* (Fig. 15). Thus the egg-nucleus first moves away from the sperm-nucleus, which immediately takes up its march in an amœboid manner towards the centre of the egg, and in the general direction of the egg-nucleus. I may finish at once with the movements of the germ-nuclei by saying that they ultimately come together in the centre of the egg. Whether or not the first movement of the egg-nucleus away from the sperm-nucleus is passively brought about by the sphere-substance, the behavior of the germ-nuclei shows that their meeting is not entirely due to mutual attraction, but partly, at least, to a common tendency to seek a dynamic centre of the egg.

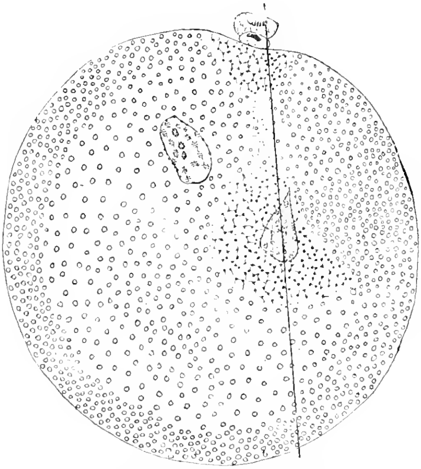

FIG. 15.—*Unio.* Early movements of the germ-nuclei and of the sphere-substance.

To return to the sphere-substance: its invariable migration towards the side of the egg opposite to that in which the sperm-nucleus lies indicates one of two things, — either that the sperm-nucleus has driven it away, or else that it is moving along lines of orientation of the egg-substance. To suppose that the minute sperm-nucleus could exercise such an effect seems impossible, and the corollary of the second alternative is that the sperm-nucleus has occupied throughout its entire resting period a definite position within the egg, which can only be explained on the assumption of a definite orientation of the egg at the time of fertilization, not only polar, but also corresponding to one of the other chief axes of the embryo. The first cleavage-plane passes very nearly through the point at which the sperm-nucleus has been resting during the maturation.

During its migration towards one side of the egg, the sphere-substance has, undergone a remarkable change of form; it has *elongated greatly in a horizontal direction, at right angles to the line uniting the germ-nuclei* (Fig. 16), *and has thus marked out the direction of the first cleavage-spindle. Now this elongation has begun, and the plane of division is indicated before the germ-nuclei have met.*

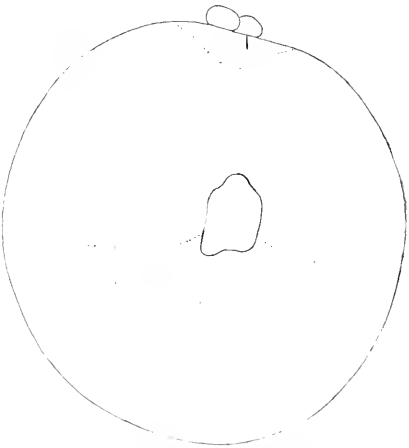
FIG. 16. — *Unio.* Outline of section in the direction of the line ruled across Fig. 15; to show the elongation of the sphere-substance.

It might very readily be assumed that the plane of the first cleavage is determined by the copulation-path of the germ-nuclei, as is stated to be the case in the ova of some other animals (frog, Roux; *Toxopneustes*, E. B. Wilson). But whoever should take this position for *Unio* would have to explain how it happens that the sphere-substance elongates in the plane of the first cleavage-spindle before the germ-nuclei come together. It would be necessary, I believe, to assume that the distant sperm-nucleus exercises an influence on the direction of migration and of elongation of the sphere-substance in the first cleavage, but that, in the next division, the sphere-substance acts independently; and this assumption is absurd on the face of it.

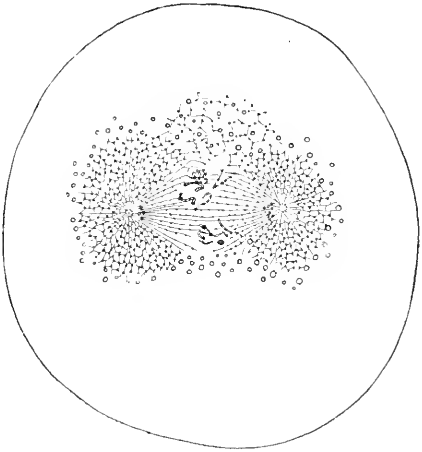
FIG. 17. — *Unio.* First cleavage-spindle forming. Basichromatin granules near the ends of the spindle; oxychromatin granules on the spindle-fibres.

The first cleavage-spindle forms in the centre of the egg (Fig. 17), in the plane already indicated by the elongation of the sphere-substance. At first there is a single very minute centrosome at each pole, almost in contact with the nuclear membrane, then a group of centrosome granules imbedded in a ground substance; then comes a clearing of the centre, accompanied by peripheral arrangement and subsequent fusion of all the centrosome granules, excepting one, which remains in the centre. Thus is established a hollow sphere ("centrosome") with an included centrosome ("centriole") (Fig. 18). During the early stages of the metamorphosis of the centrosomes, chromatin granules from the nucleus are found in their immediate neighborhood, and, as the spindle forms, they become closely pressed to the centrosomes (Fig. 17). The entire spindle then moves directly along the prolongation of its axis, thus parallel to the direction of elongation of the sphere-substance, to one side of the egg (Fig. 18), until the centrosome of one end comes almost in contact with the peripheral layer of protoplasm. Then comes the metaphase (Fig. 18), and, in a late stage of the anaphase, the sphere begins to enlarge, and a reticulum (or vesicular substance) with nodal microsomes (Fig. 19) develops in it in place of the single centrosome.

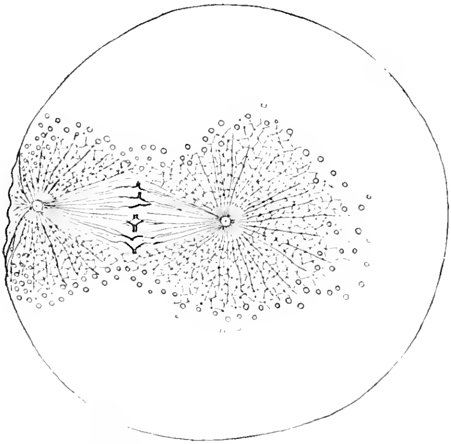

FIG. 18. — Migration of the spindle to one side of the egg; metaphase; for the sake of clearness only a few of the chromosomes were drawn in.

The egg now begins to elongate at right angles to the plane of division, and the entire spindle, including the enlarged spheres, shifts towards the centre of the egg a short distance, and finally comes to rest (Fig. 19). The spindle acts as though oscillating through a point of equilibrium, at first shifting too far in one direction (Fig. 18), then swinging back (Fig. 19), and possibly undergoing lesser shiftings before coming to rest for the first cleavage.

To what extent this centre of equilibrium is determined by the primary orientation of the cytoplasm, and how much by the secondary distribution of the sphere-substance, it is impossible to say; but the orientation of the cytoplasm is, in either event, the primary factor, inasmuch as it determines the distribution of the sphere-substance.

The first cleavage-furrow now forms rapidly, and, as it forms, the spheres undergo an enlargement, migration, and change of shape analogous to that preceding the first cleavage. We shall follow it only in the larger cell. The sphere-substance in this cell elongates during the early stages of reconstitution of the nucleus in a horizontal direction parallel to the axis later taken by the second cleavage-spindle in this cell (Fig. 20). At the same time the entire cell elongates in the same direction and becomes slightly constricted in a plane and position corresponding precisely with the next cleavage-plane; this, before the nucleus is reconstituted, or has moved away from the neighborhood of the first cleavage-wall. It is as though the cytoplasm were making an attempt at division, which is rendered abortive by the stage of development of the nucleus. Later both cells round off and then become applied together, and the second cleavage-spindle forms and moves into the position of the division already indicated.

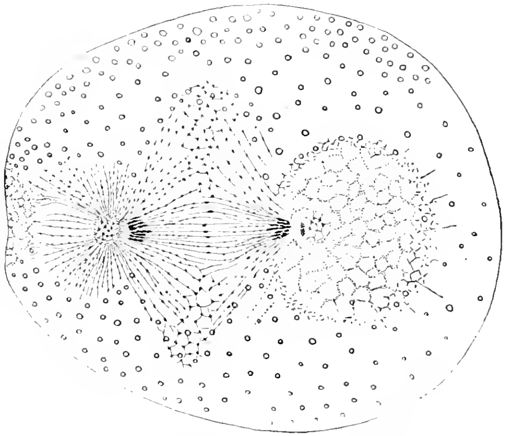

FIG. 19.—*Unio*. Secondary shifting of the first cleavage-spindle. Beginning of growth of the sphere-substance.

Conklin (4) was the first to call attention to the possible importance of the sphere-substance in the cleavage of the egg. He summarizes his results on the egg of *Crepidula* thus : "After the first two cleavages the sphere-substance is differently distributed to the different cells, the entire sphere-substance of one generation always going into those cells of the next generation which lie nearest the animal pole. This differential distribution of the spheres has been followed through every cleavage up to the twenty-four-cell stage. As the form of the cleavage is perfectly constant, it follows that the sphere-substance of any generation goes into certain definite cells which have a perfectly constant origin and destiny. This differential distribution of the spheres is not caused by their specific weight, since their movements are the same in whatever position the cell may be placed. It seems to be the result of a form of polarity which, like that of the egg itself, is not the result of gravity.

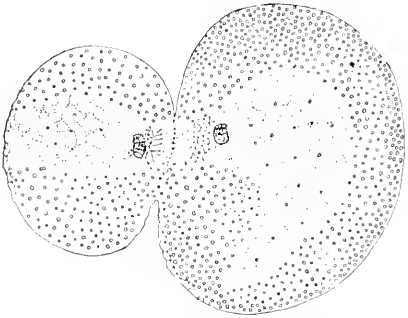

FIG. 20. — *Unio*. First cleavage; the elongation of the sphere-substance in the larger cell and of the cell itself marks the plane of the second cleavage. In a slightly later stage there is a well-marked constriction across the cell in the position of the future second cleavage-spindle in this cell.

"The centrosomes do not, apparently, arise from the sphere-substance of the previous division, but some distance from it, and the sphere-substance never divides, but each sphere ultimately grows ragged at its periphery and gradually fades out into the general cytoplasm.

"The differential distribution of the spheres and their subsequent conversion into cytoplasm suggest that they may be important factors in the differentiation of cleavage cells, and if further investigation should establish the fact that they are, in part, composed of the oxychromatin of the nucleus, it would furnish a basis in fact for certain well-known speculations of de Vries, Weismann, and Roux."

Thus, putting Conklin's results[1] on the movements of the sphere-substance and my own together, it would appear that, whereas in the first two cleavages this substance is divided between the cells in proportion to their size, in the formation of the generations of ectomeres the substance enters special cells. This would coincide very closely with the differential value of the cleavages, the first four cells possessing ectoblastic, entoblastic, and, in part, mesoblastic materials, while the three subsequent divisions of the macromeres separate ectoblastic portions. This tends, it seems to me, to strengthen Conklin's conclusion that the sphere-substance may be an important factor in the differentiation of cells.

Finally, I do not believe that the process of nuclear or cell-division is ever in itself an act of differentiation. That it is not, in certain cases at any rate, is shown beyond the possibility of any doubt by examples of non-determinate cleavage, such as that of the fish-egg, in which the cleavage-planes bear no constant relation to each other or to the embryonic parts, and, still more strikingly, in the case of ciliate *Infusoria*, where the entire process of development takes place without any cell-division. If my observations are correctly interpreted in what has preceded, the essential process in early embryo-formation proceeds on the basis of a definite orientation and organization of the egg-substance, carried forward and elaborated by certain intercellular processes, in which the production of special substances which have been acted on by the chromatin may play an important rôle. Now the distribution of these substances is not dependent on cell-division, though by this they may be isolated in separate cells; but it is conceivable that the cleavage-planes may, so to speak, ignore the lines of orientation of the egg and of distribution of specific parts of it; thus it may be that determinism in the cleavage is no measure of the degree of organization of the egg, as Whitman has so ably argued.

It is quite possible that there is no sharp distinction between

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[1] In *Crepidula*, apparently, the substance of the spheres is not divided in the second cleavage, but passes into special cells. See lecture by Dr. Conklin in this volume.

determinate and indeterminate cleavage, and that one grades into the other, the apparent difference being due to insufficient knowledge, as Conklin and Eisig (5) have suggested. But there can be no doubt in the mind of any one as to the existence of a very real difference between determinate and indeterminate types of cleavage, who has compared, for instance, the cleavage of the egg of an annelid, possessing a perfectly definite and unvarying mode of cleavage and cell-lineage of organs, with that of a fish, in which slight alterations of the external conditions cause the very greatest variations in cleavage, so that often the cells of two eggs of the same species cannot be homologized, and no definite cell-lineage of organs exists. The explanation of this difference, it seems to me, is a prospective one. It is dependent, I believe, on the actual number of cells composing the embryo at the time that the first larval or embryonic organs come into service. In other words, I would think of determinate cleavage as an adaptation to a condition in which the functional activity of organs begins with a relatively small number of cells, and in which, therefore, each cell is of special importance.

MARINE BIOLOGICAL LABORATORY,
August, 1898.

LITERATURE.

1. CHILD, C. M. A Preliminary Account of the Cleavage of Arenicola cristata, with Remarks on the Mosaic Theory. *Zoöl. Bulletin.* Vol. i. 1897.
2. CONKLIN, E. G. Cleavage and Differentiation. *Biological Lectures.* Delivered at the Marine Biological Laboratory. 1896, 1897.
3. CONKLIN, E. G. The Embryology of Crepidula. *Journ. of Morph.* Vol. xiii. 1897.
4. CONKLIN, E. G. See summary of a paper delivered before the American Society of Morphologists at the Ithaca meeting, December, 1897. *Science.* March, 1898.
5. EISIG, HUGO. Zur Entwicklungsgeschichte der Capitelliden. *Mitth. aus der Zool. Station zu Neapel.* Bd. xiii. 1898.
6. LILLIE, FRANK R. Preliminary Account of the Embryology of Unio complanata. *Journ. of Morph.* Vol. viii. 1893.
7. LILLIE, FRANK R. Embryology of the Unionidæ. *Journ. of Morph.* Vol. x. 1895.
8. LILLIE, FRANK R. Centrosome and Sphere in the Egg of Unio. *Zoöl. Bulletin.* Vol. i. 1898.
9. MEAD, A. D. The Early Development of Marine Annelids. *Journ. of Morph.* Vol. xiii. 1897.
10. TREADWELL, A. D. The Cell-Lineage of Podarke obscura. Preliminary Communication. *Zoöl. Bulletin.* Vol. i, No. 4. 1897.
11. WHITMAN, C. O. The Embryology of Clepsine. *Quar. Journ. of Micr. Science.* Vol. xviii. 1878.
12. WHITMAN, C. O. The Inadequacy of the Cell-Theory of Development. *Biological Lectures.* Delivered at the Marine Biological Laboratory. 1893.
13. WILSON, E. B. The Cell-Lineage of Nereis. *Journ. of Morph.* Vol. vi. 1892.
14. WILSON, E. B. Considerations on Cell-Lineage and Ancestral Reminiscence. *Annals of the New York Academy of Sciences.* Vol. xi, No. 1. 1898.

FOURTH LECTURE.

PROTOPLASMIC MOVEMENT AS A FACTOR OF DIFFERENTIATION.

EDWIN G. CONKLIN.

The fundamental problems of development and inheritance are in the last analysis questions of differentiation. Development is progressive differentiation coördinated as to time and place; hereditary likeness consists in the repetition by the offspring, at certain stages of its life cycle, of definite differentiations of the parent; and hereditary unlikeness, or variation, is a modification of these differentiations either as to their character or as to the time of their appearance. The phenomena of differentiation are therefore of the greatest interest, and their causes one of the most important problems of biology.

In many respects the simplest and yet most important phenomena of differentiation occur in the early stages of development, while the later differentiations of tissues and organs are more complicated and less general in character. The polarity of the egg is one of the earliest differentiations of the developing organism; it consists not only in the aggregation of yolk at one pole and of protoplasm at the other, but also in the establishment of certain structural peculiarities which in most cases determine the position and direction of the two maturation spindles and cause the first two cleavage planes to pass through the polar axis of the egg. Further, it has a definite prospective significance, since it is probable that in all animals it determines the ectodermal and endodermal poles of the embryo, while in most cases the animal and vegetal poles of the egg give rise, respectively, to the apical and oral poles of the larva or adult. This polar differentiation may appear at an early stage in the ovarian egg, or it may be delayed until after fertilization. In some cases (insects and cephalopods) not only the primary axis of the egg, but all the axes and regions of the future animal are marked out in the ovarian egg; in other cases these axes, except the primary one, are not apparent until the end of cleavage or even after gastrulation.

In the cleavage of the egg, differentiations occur in a remarkable degree in certain cases, while they appear to be absent in others. Typically, cell divisions are rhythmical, alternating, quantitatively and qualitatively equal, and consequently non-differential. The differentiations of cleavage cells are due to departures from this typical condition in one or more particulars. In certain animals these departures are very notable, the cleavages being from the first non-alternating, non-rythmical, unequal, and qualitatively dissimilar. These differentiations of cleavage have also a far-reaching prospective significance, since in certain cases (polyclades, nematodes, rotifers, annelids, mollusks) the principal axes and body regions of the future animal are marked out by the cleavage planes, and the building material of entire organs is segregated into a single cell or group of cells.

I have repeatedly observed these unequal, non-alternating, and non-rhythmical cleavages with the feeling that the causes of such differentiation were almost within sight, and with the conviction that continued study could not fail to reveal them; and yet it must be said that these causes, which seem so near at hand, generally elude one's grasp. Unequal cleavage is due to the eccentricity of the mitotic spindle, but why is the spindle eccentric? Non-alternating cleavage is due to the spindles having approximately the same direction during successive cleavages; but why do the spindles take this peculiar position? Non-rhythmical cleavage can be referred only to differences in the substance of cells, but how these differences operate cannot in most cases be explained. It is hopeless to look for an answer to the last question that may be asked concerning the cause of these or of any other phenomena; all that can reasonably be expected is that the many different phenomena and factors of development may some time be harmonized and unified by the discovery of some common causal principles.

I. MECHANICAL FACTORS OF DIFFERENTIATION.

Of the many factors of differentiation which have been proposed within recent years, the majority are in the nature of simple physical causes. Thus the polar differentiation of the egg has been attributed to differences in the specific weight of protoplasm and yolk; *e.g.*, Hertwig[1] says: "Polar differentiation consists in this, that the lighter protoplasm collects at one pole and the heavier yolk substance at the other." Experiments on the frog's egg led Pflüger, Born, and Schultze to essentially the same conclusion.

The differentiations of cleavage are commonly attributed to factors of a similar character; thus the direction of cell division is said to be due to the fact that the mitotic spindle lies in the direction of least resistance (Pflüger) or in the longest axis of the protoplasmic mass (Hertwig); the shape and position of cells, and consequently to a certain extent the direction of division, are said to be due to the rectangular intersection of cleavage planes (Sachs), or to the principle of smallest surfaces (Berthold); the rate of division and the relative size of daughter-cells are commonly attributed to the mechanical influences of inert yolk (Balfour, Hertwig). Finally, the differences in the quality of cells have been referred to intercellular reactions (Hertwig, Wilson), which, in some cases at least, are regarded as of a physical rather than of a physiological character.

It has been repeatedly shown that none of these principles are of universal application, and it seems doubtful whether in any case they are the real causes of the phenomena in question. How little gravity has to do with polar differentiation is well shown in the eggs of many gasteropods where the eggs lie in all possible positions in the egg capsules with their primary axes turned in all possible directions, and yet the polar differentiation occurs as perfectly and as rapidly in one position as in another. That gravity can have nothing directly to do

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[1] The Cell, p. 215.

with this differentiation is further shown by the fact that the protoplasm which lies at the animal pole in early stages of cleavage lies at the vegetal pole of the macromeres in later stages; while the yolk, which originally lay at the vegetal pole, comes to lie at the animal pole of the macromeres; in short, the polarity of the macromeres is reversed during cleavage, and this always happens in the same way, irrespective of the positions in which the eggs may lie.

How far these mechanical principles fall short of explaining the differentiations of cleavage has been pointed out by Mead (**'94**), Kofoid (**'94**), Lillie (**'95**), McMurrich (**'95**), Wheeler (**'95**), Zur Strassen (**'95**), Castle (**'96**), Jennings (**'96**), Conklin (**'92**, **'94**, **'97**), and many others. Much has been said, and very justly, of the difficulty of explaining by simple mechanical causes non-alternating and unequal cleavages, and yet it ought not to be forgotten that the causes of alternating and equal cleavages have not been given. To say merely that cell divisions are typically alternating and equal affords no causal explanation. If we knew why cleavages are usually alternating and equal, we should probably be able to explain why they are sometimes neither. How little alternating cleavage has to do with the mere divergence of centrosomes in planes successively at right angles to one another will be apparent further on, where we shall see that the centrosomes do not preserve their original positions in the daughter-cells, and that the direction of their divergence bears no constant relation to the direction of the cell division; and how little yolk has to do with the inequality of division is shown by the fact that in the formation of the polar bodies we have two very unequal divisions of the egg, while the first and second cleavages. are frequently equal; the first, second, and third divisions of the macromeres thus formed are usually very unequal, while the fourth and fifth divisions of these cells are again nearly equal; finally, among the micromeres, which frequently contain no yolk, the early divisions are more frequently unequal than equal. These same considerations apply in the main to the rate of division; in most cases of determinate cleavage it bears no constant relation to the presence or absence of yolk in the cells. In every case of determinate cleavage these so-called "laws" are repeatedly set at naught, and this fact has led those who have studied such cleavage to the conclusion that no simple physical explanation of these processes of differentiation is possible, and that their cause must be found in the structure of the protoplasm or in some physiological factor.

II. Physiological Factors.

The causes of differentiation are frequently referred to the structure of the germinal protoplasm, as if this were a satisfactory explanation. But to say that polarity and differentiations of cleavage are due to the constitution of the egg is merely a form of words which means little or nothing. In the same way it might be said that all the multifarious aspects of the universe are the results of the constitution of matter. To refer vital phenomena to the constitution of protoplasm and there to rest is merely to juggle with words. The phenomena in question must be analyzed and their *immediate* causes determined step by step before any "explanation" can be thought of. If, then, those who attempt to explain differentiation as the result of simple physical factors have taken too narrow a view of the problem, and have unduly simplified it, those who suggest the structure of protoplasm as an explanation propose a factor so vague and remote that it has no real value. The immediate causes of differential cleavage have been located, by different authors, in various parts of cells.

a. Nucleus. — According to the views of Weismann, Roux, De Vries, and many others, the nucleus is the prime mover in all processes of differentiation; and hence we find that they attribute the direction, size, and rate of division, and the quality of cells, to the action of the nucleus. According to Roux ('95) there is immanent in the nucleus a direction of division which may be independent of the chief dimensions of the protoplasmic body. Almost all observers agree that the direction and size of division depend upon the direction and position of the mitotic spindle, and many consider that the position of the spindle is not determined by the cell body; thus Jennings says of *Asplanchna* ('96, p. 71), "The form of the cell is greatly influenced by the direction of the contained spindle," but "the form of the cell has no effect upon the position of the spindle" (p. 72). Experiment has shown that the position of the spindle and the direction and size of division may be modified by pressure, and this again has lent support to the view that the position of the spindle is the determining feature in cell division.

b. Centrosome. — Others consider the centrosome the prime mover in cell division, "the dynamic centre of the cell." Rauber ('83) holds that the position of the spindles is the result of the mutual attractions of neighboring asters. Heidenhain ('95) refers the direction of division to a definite angle of rotation of the centrosomes.

c. Cytoplasm. — Boveri ('97) has shown that neither nucleus nor centrosome can be said to take the initiative in cell division, since the mitotic processes may go on independently in each, and both he and Driesch ('98) have shown that the rhythm of cleavage depends upon the egg cell and not upon the sperm, and therefore, in all probabilities, upon the cytoplasm and not upon the nucleus or centrosome.

III. Protoplasmic Movement as a Factor.

Almost all views as to the factors of differentiation regard the cell as in a static condition; very few consider it from the kinetic standpoint, though the very names *karyokinesis* and *cytokinesis* suggest movement as the fundamental fact of cell division.

In the maturation, fertilization, and cleavage of certain gasteropods I have observed successive stages of a complex and orderly movement of the entire cell body by which the positions of the cytoplasm, yolk, nuclei, centrosomes, and mid-bodies (Zwischenkörpern) are changed in a definite and orderly way. Unfortunately, I have been unable to actually observe these movements in the living egg, since the eggs studied contain a large amount of yolk and are therefore opaque, and since the movements described are very slow. However, the evidences of these movements are so abundant and unmistakable that one could not be more certain of them if he had seen the actual flowing of the cell substance. These observations are based in the first instance upon the eggs of *Crepidula plana*, though many other species and genera of gasteropods have been studied with the same results. The phenomena are, therefore, not isolated, and some of them are probably of general occurrence.

a. Movements in the Unsegmented Egg.

The cell movements during maturation result in the segregation of yolk and cytoplasm at opposite poles of the egg, and in the transportation of the mitotic figure to the animal pole. While the germinal vesicle is still intact it is closely surrounded by yolk spherules, and there is a very incomplete separation of yolk and cytoplasm throughout the entire egg. As soon as the mitotic spindle is formed and the nuclear membrane is broken, there is an area immediately surrounding the spindle and asters free from yolk, but elsewhere in the egg there is an intimate mingling of yolk and cytoplasm. Gradually the spindle, which at first has a position nearly at right angles to the egg axis and some distance from the surface of the egg, turns until its axis nearly coincides with that of the egg, and at the same time the whole spindle is moved out toward the surface, until finally the outer end of the spindle comes in contact with the cell membrane, and the surface of the egg is elevated into a papilla at this point. This movement is in part due to the mere lengthening of the nuclear spindle which doubles in length during the process, but in part also to a general movement of the cell body by which the spindle is turned and carried bodily toward the surface of the egg. At the same time there are movements within the egg which lead to an accumulation of cytoplasm at the animal pole and a movement of the yolk spherules toward the opposite pole. There is no evidence that this movement is due to activity on the part of the nucleus or centrosomes. The initial position of the centrosomes and the direction of the central spindle are not the same in different eggs, and yet the final position of the mitotic figure is the same in all cases; the centrosomes and asters at the two poles are identical in form, size, and staining reactions until the outer pole of the spindle comes into contact with the surface of the egg. Under these circumstances it is difficult to conceive what causes the spindle to rotate into a radial position and to move bodily toward the surface, unless it be a general movement of the cytoplasm, and the fact that throughout the egg the separation of yolk and cytoplasm is going on at the same time is additional evidence in favor of such a general movement of the cell body.

During the fertilization similar movements of the egg contents are apparently taking place; the polar segregation of yolk and cytoplasm goes on during the approach of the germ nuclei, and, as during maturation, appears to be due to movements of the cytoplasm. The spermatozoön usually enters near the vegetal pole, and is carried through almost the whole diameter of the egg to the animal pole, but it may enter at any place except the protoplasmic area immediately around the animal pole. If the sperm enters at the vegetal pole, its course toward the animal pole is nearly straight; if it enters elsewhere, its course is curved, and the nearer the point of entry to the animal pole the greater the curvature. An aster is formed in advance of the sperm nucleus and precedes it in its course through the egg. The egg nucleus and aster lie very near the animal pole and do not move from this position; they are surrounded by an area of protoplasm free from yolk. The sperm nucleus and aster in their advance through the yolk leave no path behind them; either they are carried along by a general movement of the cell contents, or the yolk is pushed out of their way, to close in again behind them immediately after they have passed. The germ nuclei and asters approach each other as I described in a former lecture at this laboratory ('**94**), and when the two are close together they lie in an area entirely free from yolk, except that a few spherules are usually found between the two nuclei or asters. These spherules, which are separated from all the rest of the yolk, appear to have been carried before the sperm elements in their advance. After the origin of the cleavage centrosomes the remnants of the asters are carried to a point above the nuclei and immediately under the polar bodies, where they disintegrate and are scattered as coarse granules, — a process which will be described more fully when we come to consider the cleavage.

What brings the germ nuclei and asters together? In a former lecture I suggested that the nuclei were passively drawn together by the formation, attachment, and contraction of astral rays. Wilson ('96) regards this view as untenable, and concludes that "the nuclei are drawn together by an actual attraction which is neutralized by union, and their movements are not improbably of a chemotactic character." Unless the nuclei have organs of locomotion it must still be true that they are brought together by something outside themselves. This something must of necessity be found in the cytoplasm (including the aster), unless the nuclei are able of themselves to move actively. I presume that amœboid movement of the nucleus would be the only kind possible, and yet I have never seen a single case in which either of the germ nuclei was amœboid. There is every evidence that the nuclei in this, as in most other cases of movement, are passive, and that their movements are brought about by the activity of the cytoplasm.

The migration of the sperm nucleus, like that of the maturation spindles, is accompanied by progressive separation of yolk and cytoplasm, and it is probable that these coincident phenomena have a common cause in general movements of the cytoplasm.

Furthermore, there are certain elements of constancy in the polar differentiation and in the plane of the first cleavage which cannot be attributed to the nuclei, and, so far as I can see, can be due only to definite characters of the cell body. It is the egg cell and not the nuclei which shows polar differentiation. The sperm nucleus and aster approach the animal pole from various positions; there is great variation in all the positions of the nuclei and asters relative to each other, and yet there is no variation in the plane of the first cleavage which always passes through the point of extrusion of the polar bodies, and in cases where the first cleavage is unequal the mitotic figure is always eccentric to the same degree. Now the first cleavage, as we shall see, is accompanied by extensive rotary movements of the cell contents, and this fact, joined to the evidences of cytoplasmic movement during maturation and fertilization, leads me to believe that definite movements of cell substance exist in the unsegmented egg. The constancy of cleavage in later stages is associated with constancy of movements in the cytoplasm, and it is probable that the same is true of the constancy which precedes cleavage.

That the movements within the cell substance of the unsegmented egg are, in certain cases at least, of a vortical character is indicated by spiral asters, first described by Mark for *Limax*, and since observed by several investigators in other animals, and also by my observation that the first cleavage in *Crepidula* is a spiral one, being oblique to the right, or dexiotropic.

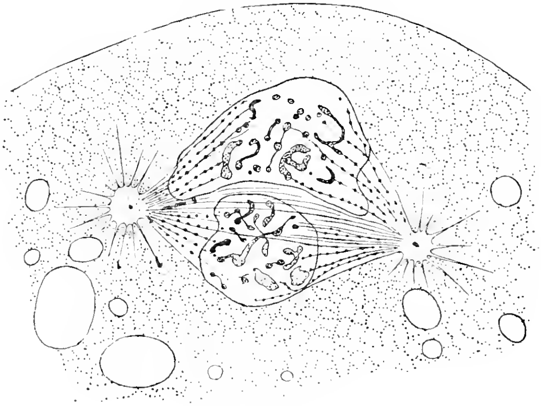

FIG. 1.

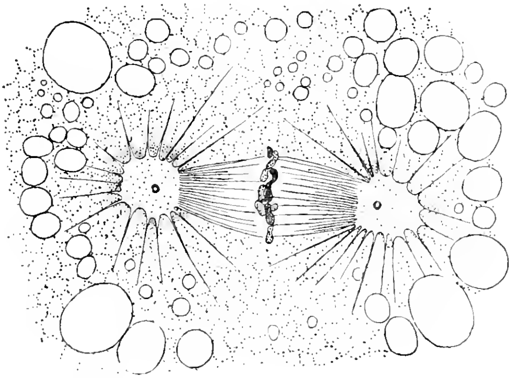

FIG. 2.

FIGS. 1 AND 2. — Sections of eggs of *Crepidula plana*, showing prophase and metaphase of first cleavage.

b. Movements during Cleavage.

It is, however, in the cleavage cells that I have found the most unmistakable evidences of the definite and orderly movements of cell contents. In the first cleavage the spindle lies at right angles to the chief axis of the egg, and the centrosomes are no nearer the surface of the egg than is the middle of the spindle (Figs. 1–3). As the daughter-nuclei pass into the resting phase, however, the centrosomes are transformed into cen-

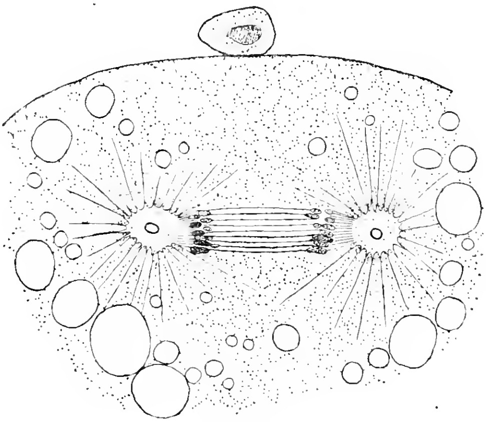

FIG. 3.

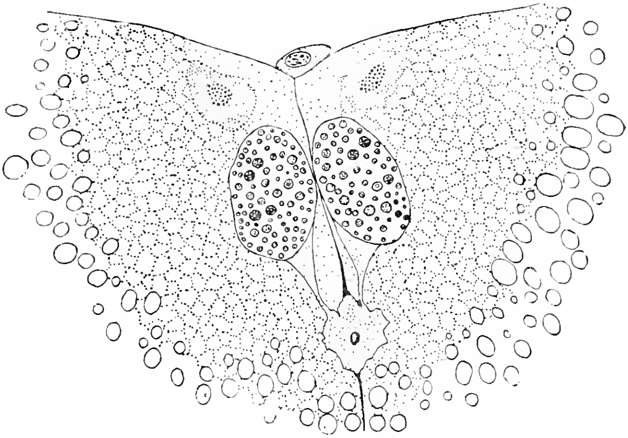

FIG. 4.

FIGS. 3 AND 4. — Sections of eggs of *C. plana*, showing anaphase and telephase of first cleavage.

trospheres, which are carried up toward the surface of the egg, and the mid-body, which marks the middle point of the spindle, is carried down into the egg, until finally the centrospheres come to lie immediately under the cell membrane and above the nuclei, while the mid-body is located near the centre of the egg (Figs. 4–6). The currents which bring about this affect the whole cell body, for not only are the centrospheres, nuclei, and mid-bodies moved, but the yolk and protoplasmic areas shift so that the protoplasm, which was spread out as a rather broad area above the yolk, becomes narrower and deeper, the yolk moving up at the periphery and down in the middle of the egg (Figs. 3–5). Such a change of position could be brought about only by a general movement of the entire cell body in the direction of the arrows in Fig. 5. The movements in the two cells are not, however, directly at right angles to the plane of the first cleavage, but viewed from the animal pole they are slightly dexiotropic, as is shown by the fact that the nuclei, spheres, and protoplasmic areas all move in a dexiotropic direction (Fig. 6). The remains of the centrospheres of the first cleavage can be seen, until the anaphase of the second cleavage, lying near the upper surface of the two blastomeres and close to the wall between them; in this position they gradually fade out into the cytoplasm, until at the close of the second cleavage no trace of them can be seen. The origin of the centrosomes of the second cleavage has not been traced in detail, but judging by analogy with other cleavages it is almost certain that they come from the inner portion of the centrospheres of the first cleavage. The plane of divergence of the centrosomes is not constant; in extreme cases the spindles may vary as much as 90° from the usual direction, and yet in all cases the spindle is ultimately turned into a læotropic position. In this case, therefore, as in the formation of the polar bodies and in many of the later cleavages, the initial position of the mitotic spindle does not determine the direction of the cell division. The flow of cell substance indicated in Fig. 6 continues until the superficial extent of the protoplasmic area is smaller and its depth greater than is indicated in that figure, and until the new centrosomes have taken their positions at the poles of the greatly inflated nuclei. During the mitosis the surface extent of the protoplasmic area becomes greater, and the blastomeres, which had become so flattened against each other that they were nearly perfect hemispheres, again become more and more spherical in shape; at the same time currents seem to move outward from the centre of the egg, and this time in such direction as to bring about a læotropic shifting of the nuclei and protoplasmic areas (Fig. 7). This movement continues until the anaphase of the second cleavage, when again the cell currents move toward the centre of the egg (Fig. 8). The direction of these currents, though nearly radial, is still slightly læotropic in the two blasto-

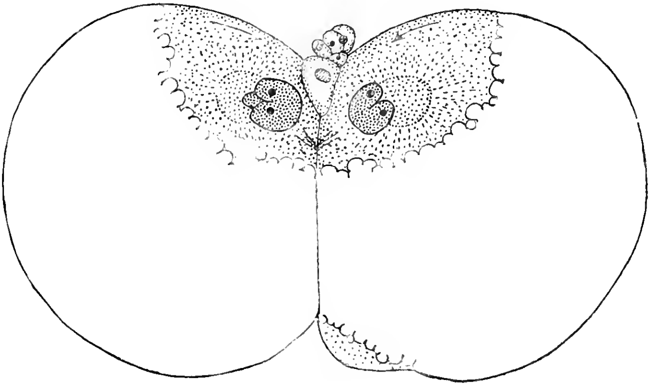

FIG. 5.

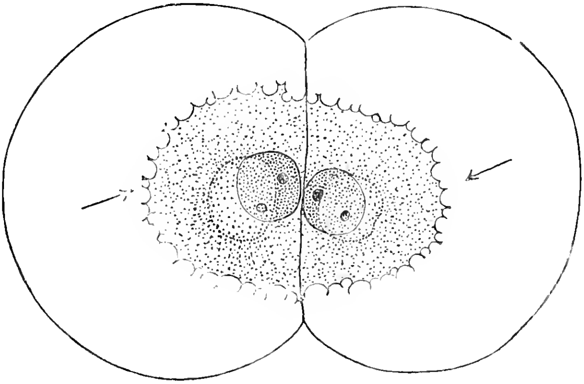

FIG. 6.

FIGS. 5 AND 6. — Entire eggs of *C. plana*, lateral and apical views. The centrospheres are shown in light shading. The arrows indicate the directions of the protoplasmic movements.

meres which lie at the higher level and dexiotropic in the two lower ones. This movement continues until the spheres are carried into the inner angles of the cells, immediately below the

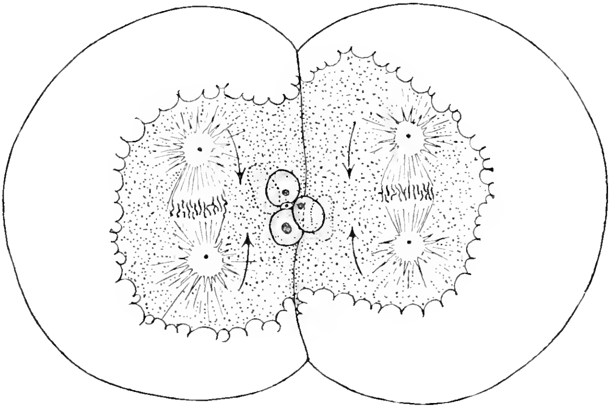

Fig. 7.

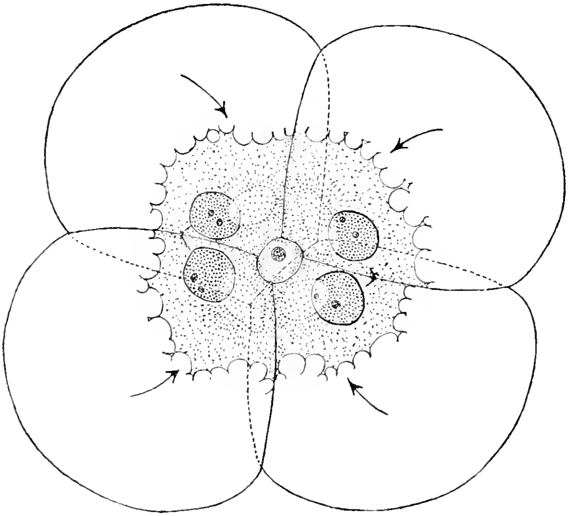

Fig. 8.

Figs. 7 and 8. — Entire eggs of *C. plana*; second cleavage.

polar bodies, and until the daughter-nuclei, which at first lie very near each other on opposite sides of the second cleavage plane (Fig. 8), are swung out into the centre of the protoplasmic area. The centrosomes and spindles then appear as in the

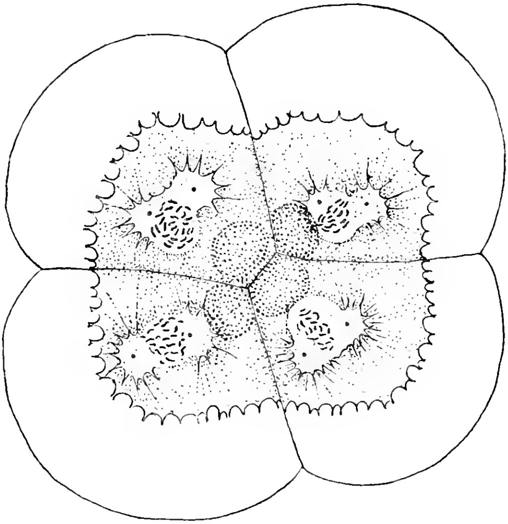

FIG. 9.

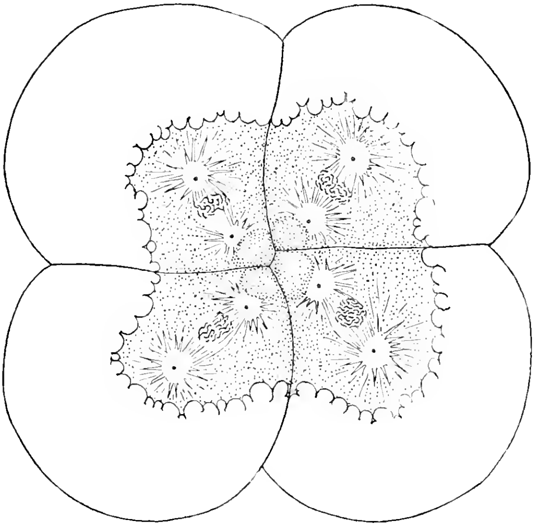

FIG. 10.

FIGS. 9 AND 10. — Entire eggs of *C. plana*; third cleavage. Sphere substance of second cleavage at central and upper angles of cells. Initial direction of the spindles not uniform.

preceding cleavage. The protoplasmic area increases with the breaking of the nuclear membrane, and the spindles do not occupy similar positions in the four cells but are often dexiotropic in the two lower blastomeres (the ones meeting in the polar furrow), and frequently læotropic in the upper ones (Fig. 9). As the cleavage advances the spindles are all turned in a dexiotropic direction. All this time the remains of the spheres of the preceding cleavage occupy the central angles of the cells, where they are plainly undergoing disintegration (Figs. 9–11). During and after this cleavage the cell currents rotate in a dexiotropic direction in the upper cells (micromeres), and in a læotropic direction in the lower cells (macromeres); this movement is manifest during cleavage in the dexiotropic lobing of the cytoplasm in each cell preparatory to the formation of the micromeres, and in the obliquity of the cell plate to the axis of the spindle (Fig. 11); after cleavage it appears in the rotation of the centrospheres and nuclei. Even after the third cleavage is completed the sphere substance of the second cleavage can be seen in the inner angles of the micromeres, where it gradually disintegrates and passes into the cytoplasm. The dexiotropic rotation of the protoplasm in the micromeres continues until the daughter-nuclei are carried from the left to the right side of the cells; at the same time the substance of the macromeres rotates to the left, until the nuclei and spheres are brought into contact with the cell membranes at the left of these cells (Fig. 12). Here the centrosomes and central spindles arise from the inner spheres of the centrospheres, and the læotropic movement of the cytoplasm continues until the spindles, which at first are equatorial, become decidedly læotropic in position, and the second quartette of micromeres is given off in a læotropic direction. As the whole of the sphere substance of the second cleavage goes into the first quartette of micromeres, so all the sphere substance of the third cleavage remaining in the macromeres goes into the second quartette, where it rapidly disintegrates and disappears. The contents of the second quartette move in a læotropic direction until the centrosphere is carried from the extreme left to the extreme right of each cell; at the same time the substance of the macromeres moves in a dexiotropic direction until the centrospheres and the nuclei are carried to the right side of each cell, and in this position the third quartette is given off in a dexiotropic direction (Figs.

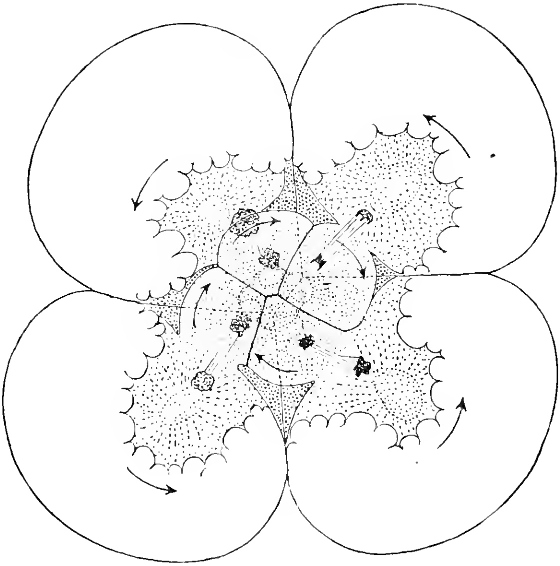

Fig. 11.

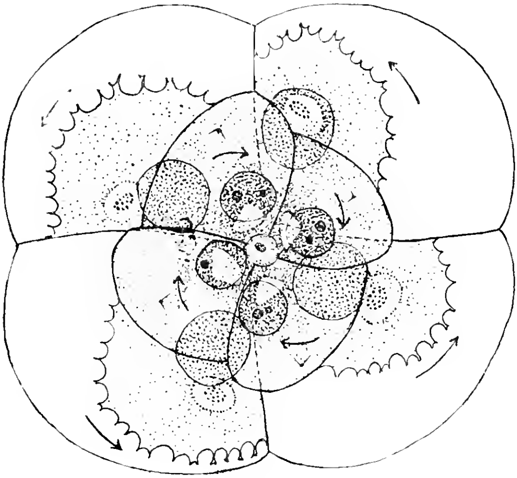

Fig. 12.

Figs. 11 and 12. — Entire eggs of *C. plana*. Anaphase and telephase of third cleavage. Sphere substance of second cleavage in inner angles of micromeres (Fig. 11). The two figures show the beginning and the end of the rotations indicated by the arrows.

13 and 14). Here, as in the preceding cleavages, the whole of the sphere substance left in the macromeres goes into the upper cell products, *i.e.*, in this case, the third quartette.

Finally, I shall describe a few subdivisions of the first and second quartettes with reference to these cell movements. During and after the formation of the first quartette, the rotations in these cells are dexiotropic; when the nuclei have been

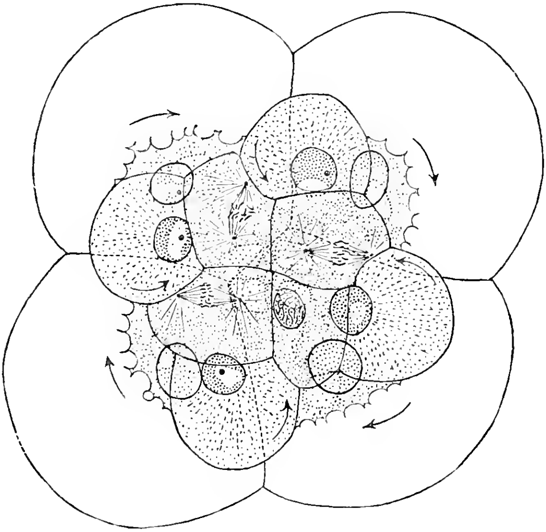

Fig. 13.

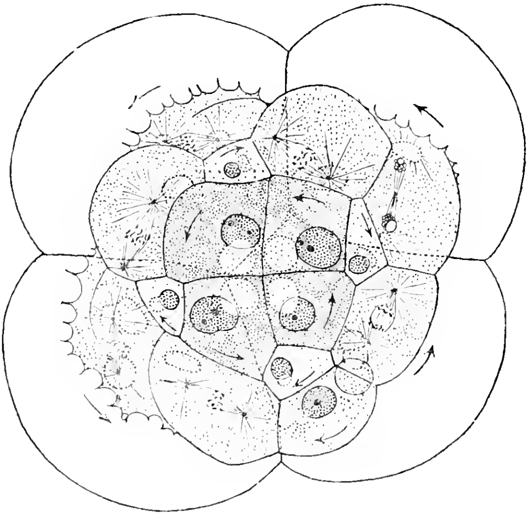

Fig. 14.

Figs. 13 and 14. — Entire eggs of *C. plana*. Fig. 13, Unequal division of first quartette cells. Fig. 14, Subdivision of second quartette cells and formation of third quartette.

carried from the left to the right side of each cell, the spindles for their first subdivision appear, and the cleavage takes place in a læotropic direction (Fig. 13). This cleavage is a very unequal one, the outer products being the small trochoblasts. The inequality is preceded by a marked eccentricity of the nuclear spindle (Fig. 13); this eccentricity is not indicated in the position of the nuclei before the spindle appears, for they lie near the inner angles of the cells (see the cell not dividing in Fig. 13), but during the prophase the nuclei and spindles move outward in the cells until the outer (lower) centrosome comes almost into contact with the cell membrane, while the inner (upper) one lies near the middle of the cell. In this position the division takes place in a læotropic direction (Fig. 13), and immediately the currents in the upper cell moiety begin to rotate in a læotropic direction, while those in the lower moiety usually rotate in a dexiotropic direction. In this case the eccentricity of the spindle is the immediate cause of the unequal cleavage, and this eccentricity is the result of vortical movements in the cell, which begin coincidently with the breaking of the nuclear membrane.

The first division of the second quartette is a nearly equal one, and is slightly dexiotropic in direction. These cells were formed by læotropic cleavage, and consequently the rotation within them is a læotropic one. When this læotropic movement has carried the centrospheres from the extreme left to the extreme right of the cell, the cleavage begins, the whole of the sphere substance going into the right product of division (Fig. 14).

Especial interest attaches to the cell movements in reversed cleavage, or cases in which two successive divisions are in the same direction. Such an instance occurs in the first division of the basal cells of the cross. (See Conklin, '97 and '98.) These cells were formed by dexiotropic division of the apical cells, and to preserve the law of alternation they should divide in a læotropic direction, but they all divide dexiotropically. Immediately after the apicals have given rise to these basal cells, the contents of the former rotate in a dexiotropic direction until the centrospheres are carried through an angle of about 90°;

at the same time the centrospheres of the basal cells remain almost exactly in their first position, though they move slightly toward the surface and sometimes a little to the left. The reversed cleavage of these cells seems not to be due to reversed currents in the cells, but rather to the absence of any currents.

Further divisions have been followed in detail up to a late stage in the cleavage, but as they illustrate merely the principles which have been already described, no account is given of them here.

IV. Summary and Comparisons.

The principal facts as to the rotation of the cell contents, or what might be called the cytokinesis of cleavage, are the following: 1. With the escape of nuclear sap into the cell body at the beginning of mitosis, vortical movements are set up in the cytoplasm, the poles of the spindles being the centres of such vortices. 2. The vortices in daughter-cells are in reverse directions, the movement in the upper cell being in the direction of the cleavage by which that cell was formed. 3. Cell walls form where vortices meet. 4. Successive divisions alternate in direction when the rotations carry the nucleus to the side of the cell opposite to that in which it first lay; non-alternation is due to the absence of such rotation. 5. When the cell movements carry the mitotic spindle out of the middle of the cell, unequal cleavage results. 6. After the first two cleavages, every cell division is qualitatively differential, since the sphere substance of the preceding cleavage is carried into one only of the two daughter-cells.

Such movements of the cell substance as are here described have long been known in certain plant cells and among protozoa. Ryder (**'94**) in particular has discussed the movements of amœboid organisms and finds the cause of such movement in a vortical flow of protoplasm. A few observations have also been made on the movements of cell substance in certain ova, but in no case have these movements been followed in detail, and in no case have they been connected with processes of differentiation. Whitman (**'87**) emphasized the importance of the cytoplasm in the movements of the germinal vesicle and of the pronuclei during maturation and fertilization. For these active changes in the cytoplasm he proposed the name *cytokinesis.* Morgan ('93) observed that the reddish pigment granules found over the surface of the eggs of *Arbacia* move entirely away from the micromere pole of the egg before the micromeres are formed. In some eggs this movement begins in the two-cell stage, and is carried on until the micromeres are formed at the sixteen-cell stage. Nussbaum ('93) observed in the division of entoderm and mesoderm cells of young embryos of *Rana temporaria* that the brown-black pigment collected in a ring around the equator of the dividing cell, and as the division advanced the ring became narrower and deeper until it formed a true cell plate between the daughter-cells. Van Bambeke ('96) has observed a similar phenomenon in the cleavage of the toad's egg. Gardiner ('95) observed in the eggs of *Polychœrus* and *Aphanostoma* a reddish-yellow pigment which, because of its form and peculiar movements, he supposed might be some form of alga. After the egg is laid it migrates from the centre toward the periphery, and forms a girdle around the ovum in the plane of the first cleavage. A similar line of pigment marks out the division plane of every succeeding cleavage up to the ten-cell stage. He also observed that these pigment granules migrated from one pole of the egg to the other, though they never passed from one cell to the other. These movements greatly impressed Gardiner with the wonderfully active and powerful forces within the egg. When the living egg is seen under an immersion lens he says "the surface fairly scintillates with the movements of the protoplasm and these pigment granules."

About the same time Loeb ('95^1) suggested that a mechanical explanation of the division of the egg or embryo was to be found in diffusion and vortex movements of the protoplasm, similar to those observed by Quincke in an emulsion of oil and soda solution. "I conceive," says he, "that on the surface of the egg, possibly in the meridian or circle whose plane separates from one another the two radiating systems of the centrosome, diffusion phenomena occur as soon as the nuclear division has physically ended. These lead to the formation of vortical movements, symmetrical in relation to this plane." If these movements are violent, they lead to the complete separation of the daughter-cells; if not, ordinary cleavage results. Later ('95²) he observed in the segmenting eggs of *Ctenolabrus*, droplets over the surface of the egg which collected in the plane of the next succeeding cleavage; this phenomenon he considered a confirmation of his theory.

The cell movements which occur during the maturation, fertilization, and cleavage of *Crepidula* not only confirm Loeb's view as to the mechanics of cleavage, but they throw light upon the mechanics of differentiation. Of the four features of differential cleavage, *viz.*, differentiations in the (1) direction, (2) size, (3) quality, and (4) rate of division, the first three are due, in part at least, to these movements of the cytoplasm. Evidently, varying rates of division must be attributed to some other cause.

If we go further and inquire what causes and directs these movements, we cannot at present find any positive answer. It seems probable that they are due to the appearance of unlike substances in different parts of the cell, the movements being of a chemotropic character. Further, these movements are correlated with the growth and collapse of the nucleus and centrosome,[1] and especially with the escape of nuclear sap into the cell. It is *a priori* improbable that any one cell constituent, as distinguished from the whole, is responsible for these movements. The centrosomes are not the sole dynamic or kinetic centres of the cell, since the movements of the cytoplasm carry the centrosomes where they will, and control the direction of division and the relative size and quality of the daughter-cells. In this, as in other phenomena, the cell acts as a whole, and in the interaction of its various parts are to be found the causes of all vital phenomena.

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[1] My complete paper on the history of these structures during maturation, fertilization, and cleavage, will appear shortly.

BIBLIOGRAPHY.

'96 BAMBEKE, VAN. Sur un groupement de granules pigmentaires dans l'œuf en segmentation d'Amphibieus. *Bull. Acad. Belg.* T. 31.

'97 BOVERI. Zur Physiologie der Kern- und Zellteilung. *Sitz.-Ber. Phys. med. Ges. z. Würzburg.*

'96 CASTLE. The Early Embryology of Cione intestinalis. *Bull. Mus. Comp. Zoöl.* (Harvard University). Vol. xxvii.

'92 CONKLIN. The Cleavage of the Ovum in Crepidula fornicata. *Zool. Anzeiger.* Bd. xv.

'94 CONKLIN. The Fertilization of the Ovum. *Wood's Holl Lectures.*

'97 CONKLIN. The Embryology of Crepidula. *Journ. of Morph.* Vol. xiii.

'98 CONKLIN. Cleavage and Differentiation. *Wood's Holl Lectures.*

'98 DRIESCH. Ueber rein-mütterliche Charactere an Bastardlarven von Echiniden. *Arch. f. Entw. d. Mech.* Bd. vii.

'95 GARDINER. Early Development of Polychœrus caudatus. *Journ. of Morph.* Vol. xi.

'95 HEIDENHAIN. Cytomechanische Studien. *Arch. f. Entw. d. Mech.* Bd. i.

'96 JENNINGS. The Early Development of Asplanchna Herickii. *Bull. Mus. Comp. Zoöl.* (Harvard University). Vol. xxx.

'94 KOFOID. On Some Laws of Cleavage in Limax. *Proc. Amer. Acad. Arts and Sci.* Vol. xxix.

'95 LILLIE. The Embryology of the Unionidæ. *Journ. of Morph.* Vol. x.

'95 LOEB. Beiträge zur Entwicklungsmechanik der aus einem Ei entstehenden Doppelbildung. *Arch. f. Entw. d. Mech.* Bd. i.

'95 LOEB. Untersuchungen über die physiologischen Wirkungen des Sauerstoffmangels. *Pflüger's Archiv.* Bd. lxii.

'95 MCMURRICH. Cell Division and Development. *Wood's Holl Lectures.*

'94 MEAD. Preliminary Account of the Cell Lineage of Amphitrite and other Annelids. *Journ. of Morph.* Vol. ix.

'93 MORGAN. Experimental Studies on Echinoderm Eggs. *Anat. Anzeiger.* Bd. ix.

'93 NUSSBAUM. Ueber die Verteilung der Pigmentkörnchen bei Karyokinese. *Anat. Anzeiger.* Jahrg. viii.

'83 RAUBER. Neue Grundlegungen zur Kentniss der Zelle. *Morph. Jahrb.* Bd. viii.

'94 ROUX. Discussion of Ziegler's paper, "Furchung unter Pressung." *Verhandl. Anat. Gesellsch., Achte Versamml.*

'94 RYDER. Dynamics in Evolution. *Wood's Holl Lectures.*
95 WHEELER. The Behavior of the Centrosomes in the Fertilized Egg of Myzostoma glabrum. *Journ. of Morph.* Vol. x.
'87 WHITMAN. Oökinesis. *Journ. of Morph.* Vol. i.
'96 WILSON. The Cell in Development and Inheritance.
'96 ZUR STRASSEN. Embryonal Entwicklung der Ascaris megalocephala. *Arch. f. Entw. d. Mech.* Bd. iii.

FIFTH LECTURE.

EQUAL AND UNEQUAL CLEAVAGE IN ANNELIDS.

A. L. TREADWELL.

INVESTIGATIONS in cell lineage, the line of work contemptuously regarded by some morphologists as a process of "cell counting," have given us in the last few years some important data concerning the early cleavage stages of animals belonging to a number of different classes, the best-known groups being the annelids, molluscs, and platodes. We are now in possession of a sufficient number of facts to draw some conclusions concerning the cleavage of the ovum and the formation of the embryo. Some of the results of these investigations, bearing upon questions of cell homology, differentiation, etc., have been discussed by other lecturers in this course, and I shall therefore limit myself to problems connected with the early cleavages.

Concerning cleavage in general, its origin and meaning, very diverse views are held by different writers. On the one hand, it is maintained that there is from the beginning a definite organization of the ovum, and that cleavage merely brings this organization to light by isolating certain areas by means of cell boundaries. On the other hand, it is argued that the ovum is at the beginning isotropic, and that differentiation only begins after cells are formed, and as a result of their mutual action upon one another. It is maintained by some that the similarities which appear, for example, between the cleavage stages of an annelid and a platode are of as great value in establishing relationships as are the resemblances which exist between the so-called homologous parts of the adult animals; in other words, that cleavage has a phylogenetic significance. Others regard these resemblances as superficial and accidental, and cleavage as having no phylogenetic significance whatever, except as a possible indication of the mode of origin of the primitive metazoön from its protozoön ancestor.

It is noteworthy in this connection that the theory which one adopts is apt to be dependent upon the animal which he has studied. In some groups, for example, in many annelids, molluscs, platodes, ascidians, and nematodes, the cleavage always follows a definite form or pattern, and, apparently, is so correlated with differentiation that Conklin has proposed to designate it as *determinate*, as opposed to the *indeterminate* type found in many vertebrates, echinoderms, and *Cnidaria*, where the cleavage seems to be very indefinite and inconstant. Investigators who have worked on the former groups are generally inclined to give greater importance to cleavage than those who have studied the latter. No one who has followed the cell lineage of an annelid or a mollusc can deny an *apparent* differentiation, manifest even in the earliest cleavages, and the notion of such differentiation seems not disproved by the results of the experimental embryologists. The argument in this line has been so ably presented in another place[1] that I will not attempt to repeat it here, but pass to the more especial discussion of equal and unequal cleavages.

It is a fact, familiar to all who have ever seen a segmenting ovum, that the first cleavage in all holoblastic eggs divides the cell into two equal or unequal parts, according to the particular animal studied, the widest possible variations appearing among closely related animals. Inasmuch as unequal cleavage is usually associated with the presence of a considerable amount of food yolk, it was formerly supposed that yolk alone is responsible for this form of division. A careful study of the developing ovum shows, however, that this explanation will not apply in all, or even in the majority of cases, and I shall offer some suggestions, based on a study of the annelids, which may, perhaps, throw some light on the subject. For this purpose

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[1] Conklin, "Embryology of Crepidula," *Biol. Lectures* (1896, 1897), and *Journ. of Morph.*, vol. xiii.

I shall make a somewhat detailed comparison of *Amphitrite*,[1] which has been studied by Dr. Mead, as an example of unequal, and *Podarke*, on which I have been working for some time, as an example of equal cleavage. In the unequal type, to which most of the annelids and all of the gasteropods and lamellibranchs which have been studied belong, frequently at the two-, and always at the eight-cell stage, differences in size appear among the segments; and, supposing the first and second cleavage planes to divide the body into four parts, the *quadrants* are never exactly symmetrical; so that, from the very beginning, orientation is comparatively a simple matter. In *Amphitrite* the first division is unequal, and this difference in size is continued into the four-cell stage (Fig. 1).[2] Here one cell is very much larger than either of the other three, and lies at what will be the posterior end of the future embryo. Notice here that the second plane of cleavage is not a straight line, but it cuts the first in two points some distance apart. The portion of the second furrow between these two points is the so-called "cross furrow," or "Brechungslinie." I shall follow Conklin in calling it the "polar furrow." It is obviously formed by the rotation of cells *A* and *C* to the left, so that they lie partly over *B* and *D*. In *Amphitrite*, as in other

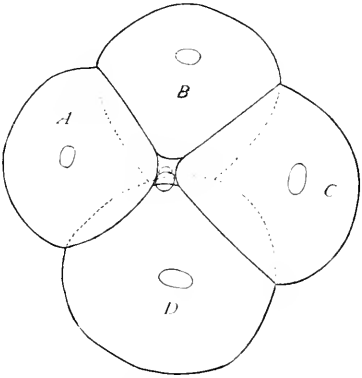

FIG. 1. — *Amphitrite*, four-cell stage.

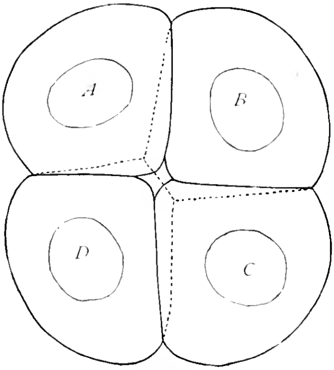

FIG. 2. — *Podarke*, four-cell stage.

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[1] Mead, "Development of Marine Annelids," *Journ. of Morph.*, vol. xiii.

[2] I have modified slightly the lettering in the figures taken from Mead to correspond with a nomenclature proposed for *Podarke*.

annelids and molluscs, this "polar furrow" retains its original direction until a late stage of the development, and is of great service in orientation.

At the next division each cell divides equatorially, so that there results an eight-cell stage, having four cells above and four below, the four upper being slightly smaller than the lower. (See Fig. 3 of an eight-cell stage from below.) Note especially the large size of *D* and the difference in size between the upper and lower quartettes. Because of this difference in size, we may speak of the upper quartette as micromeres, and

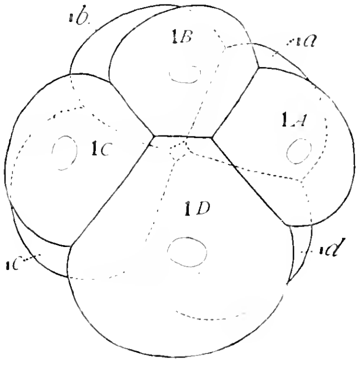

Fig. 3.—*Amphitrite*, eight cells from below.

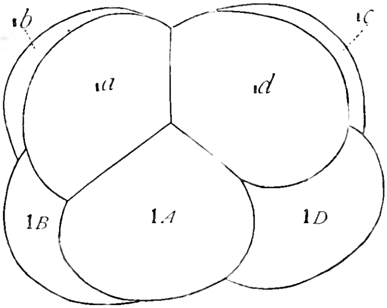

Fig. 4.—*Podarke*, eight cells from the side.

the lower quartette as macromeres, the distinction between the two quartettes being much less noticeable here than in some other annelids.

This process is repeated during the formation of two more groups of micromeres, which form the body ectoderm, and a fourth group, one member of which forms the mesoderm, while the other three unite with the remaining macromeres to form the entoderm. Meanwhile cells earlier formed have divided, so that the separation of the mesoblast cell coincides with the completion of the ideal sixty-four-cell stage, a stage which, owing to delay in the division of some cells and a hastening of the division of others, may never actually be attained.

A marked feature of this formation of micromeres is the division of each group in an alternating spiral direction. The eight-cell stage (Fig. 3) arises from the four-cell by a division of each cell and a rotation of the upper portion in a clockwise direction. The next division to the sixteen-cell stage is anti-clockwise; so that we can speak of an alternating direction of cleavage as characteristic. This alternating direction is often obscured in cases where the segmentation does not follow a regular geometrical progression; *e.g.*, *Nereis*, *Crepidula*, and *Unio*, and beyond the sixty-four-cell stage it is lost more or less rapidly in all. Why cleavages should always take such definite directions we do not know. Mechanical conditions undoubtedly play a large part in the arrangement, but mechanical explanations are not sufficient to account for the definite direction assumed by each cleavage. The fact that in molluscs with a dextral shell the first rotation is to the right, while in those with a sinistral shell it turns to the left, may indicate some relation between the form of the first divisions and the symmetry of the future body; but in the present state of our knowledge such an assumption would be premature.

The important points in the development thus far are these: 1. At the sixteen-cell stage a separation of certain cells form the first generation of ectomeres, which go to form the primary prototroch, and are, therefore, trochoblasts. These divide twice, and twice only; so that in the sixty-four-cell stage there are sixteen of these cells, which become ciliated and form the larval locomotor apparatus. 2. The second micromere, separated from the large macromere *D*, is larger than any of the other members of the same quartette, and, as further study shows, forms a large part of the ectoderm of the future trunk of the animal. According to Dr. Mead, all of the ectoderm of the body behind the first septum arises from this cell. 3. The separation of the mesoderm, in large part at least, in the fourth micromere in quadrant *D*. 4. The separation of four rosette cells in the upper pole, which later divide once, and carry the apical tuft of cilia, which is such a marked feature of the larva. 5. The segregation of the entoderm in the macromeres, which invaginate to form the alimentary canal.

Evidently, then, the egg of such an animal as *Amphitrite* possesses from the beginning a definite orientation. This is indicated by the "polar furrow," which always has a definite relation to the future body axes, and by the fact that the large cell *D* always lies at the posterior end of the animal. Comparison with other annelids shows that wherever this difference in size appears, the large cell always lies at the posterior end of the body, and from it are given off the cells *2d* and *4d*, which are larger than the corresponding members of the same quartette. In *Unio*, among the molluscs, a similar relation appears. In other molluscs apparent exceptions occur, of which I will speak later.

Let us now turn to a consideration of equal cleavage among annelids. For an explanation of this type I will take *Podarke*, as the one whose development has been most completely studied. The four-cell stage of this form is shown in Fig. 2. All the cells are equal in size, and the only orientation point which presents itself is the "polar furrow." Observation of the living egg shows that this "polar furrow" remains constant in direction up to a comparatively late stage in the cleavage, when it becomes possible to orient the egg by means of other landmarks.

In the next figure (Fig. 4) is shown the eight-cell stage, seen from the side. Instead of a set of smaller micromeres, lying on top of four larger macromeres, we have eight cells, all exactly alike in size; and only by means of the polar globules are we able to distinguish the animal from the vegetative pole of the egg. The "polar furrow" enables us to distinguish the second from the first plane of cleavage; but there is no difference in the size of the cells, as in *Amphitrite*, by means of which one end of this furrow can be distinguished from the other.

At the next division a second group of micromeres is formed, and, by a division of the first group, four smaller cells, which are the parent cells of the primary prototroch. These, exactly as in *Amphitrite*, divide twice, and twice only, and form the beginning of the larval locomotor organ, which becomes functional at the completion of the sixty-four-cell stage.

The cells of the second quartette, given off at the sixteen-cell stage, are all of the same size, in contrast to what occurs in *Amphitrite*, where *2d* is so much larger than the other members of the same quartette.

As in *Amphitrite*, a third quartette of ectomeres is given off, and these are followed by the formation of a fourth quartette, of which three of the members become mainly entoderm, while a fourth contains the mesoderm. This fourth micromere, arising from the posterior cell *3D*, is of the same size as the other members of the quartette, and at the sixty-four-cell stage is only distinguishable from them by its position. But, while the others enter into the formation of the entoderm, this divides bilaterally at the surface, and, after dividing off small entoderm cells (the "secondary mesoblast" of Wilson), each divides to form the mesoblast of the corresponding side. This cell, *4d*, is separated from *3D* at the forty-cell stage, and the sixty-four-cell stage, which is actual rather than ideal in this animal, is completed by the division of the primary trochoblasts, the division of the intermediate cells lying between the arms of the cross, and by the division of the second quartette of micromeres, now two in number in each quadrant. The latter division presents some features of importance. In each of three quadrants the ectomeres divide in such a way that, of the resulting four cells, two smaller are above, and two larger are below. In the remaining quadrant, on the other hand, while one of the cells divides like those of the other quadrants, the other divides in a different way, and there is given off a large cell above, and a very small cell below, which has a deeply staining nucleus, and is an important landmark in the further study of the embryo. This small cell, as later study shows, lies directly over the micromere *4d*, and since the descendants of *2d* are conventionally distinguished by the term *X*, this cell ($2d_{2,2}$) would be in this system of nomenclature $X_{1,2}$. Exactly this cell has been found in a number of other annelids, including those with the most unequal cleavage, and in *Unio*[1] and *Crepidula*, among the molluscs. I have followed the living egg up to the stage where this cell appears, and find that the "polar furrow" retains its position, at least, until then — a position which agrees with the position assumed in *Amphitrite* and *Arenicola*.[2] By the aid of this cell, $X_{1,2}$, and the mesoblast cell, which lies immediately under it, orientation of the embryo is complete.

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[1] Lillie, "Embryology of the Unionidae," *Journ. of Morph.*, vol. x.

[2] Child, "Cleavage of Arenicola cristata," *Zoöl. Bulletin*, vol. i, No. 2.

A comparison of the embryo of *Podarke* with that of *Amphitrite* at the sixty-four-cell stage will be instructive. Fig. 6 represents the completion of the sixty-four-cell stage in *Podarke* by the division of the descendants of the second group of micromeres. Note especially the small size of the macromeres, *4A*, *4B*, *4C*, *4D*, the equality in size between the members of the fourth quartette of micromeres, and the arrangement of spindles in the cells undergoing karyokinesis. At this time the embryo of *Podarke* is *apparently* radially symmetrical, a symmetry which is only apparent, as is indicated by the position of the "polar furrow." This apparent symmetry is destroyed by the completion of the division shown in the figure, separating the small cell $X_{1,2}$, which comes to lie just over *4d*, the mesoderm.

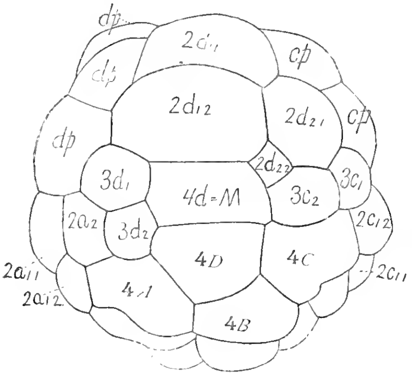

Fig. 5. — *Amphitrite*, sixty-four cells from below; *cp–dp*, primary trochoblasts.

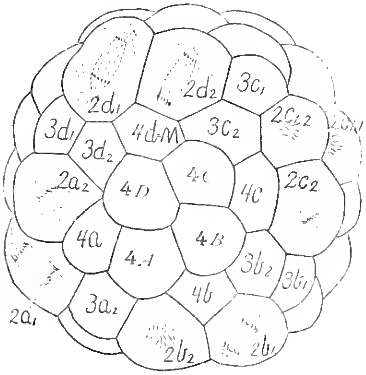

Fig. 6. — *Podarke*, fifty-six to sixty-four cells from below.

From this time on, bilateral divisions appear among the cells of both hemispheres, the most important being those which lead to the formation of the bilaterally symmetrical cross at the upper pole, and the bilateral division of *4d* with the cells of the third quartette of micromeres, which immediately adjoin it at the lower pole. The last division is of especial interest, as we shall see, as it leads to the formation of a larval mesoblast.

Let us now compare this with *Amphitrite*. Fig. 5 represents an embryo of *Amphitrite* at approximately the sixty-four-cell stage. Note the large macromeres, and the large size of $2d = X$ and $4d = M$ when compared with other members of the corresponding quartette, while in *Podarke* these cells are no larger than corresponding cells in the other quadrants.

Fig. 7, of *Amphitrite*, and Fig. 8, of *Podarke*, show the upper poles of embryos in practically the same stages as Figs. 5 and 6. Note here the comparatively large size of the cross-cells in *Podarke*, composing a larger portion of the umbrella than do the corresponding cells in *Amphitrite* (*cf.* Figs. 7 and 8).

Leaving the question of strictly cell development, let us pass to the later embryonic history and note how the trochophore arises from the embryo of sixty-four cells. The differentiation of the prototroch cells enables us to distinguish an upper umbrella from a lower subumbrella region. Later, the prototroch ring is partially completed in *Amphitrite* by the addition of three cells in each of three quadrants from the lower hemisphere, the fourth, that of *D*, remaining open for some time. Through this opening a number of cells from the upper hemisphere invaginate and form a portion of the subumbrella ectoderm. The descendants of 2*d*, or *X*, have meanwhile multiplied rapidly, and compose the greater part of the trunk ectoderm. As I said before, Mead believes that all of the trunk ectoderm behind the first segment is composed of the descendants of these cells. Meanwhile certain of them become ciliated and form the paratroch, a band of cilia incompletely surrounding the body just in front of the anus. At this time, when the

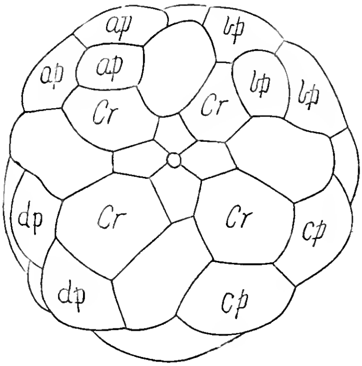

FIG. 7.—*Amphitrite*, about sixty-four cells from above; *ap–dp*, primary trochoblasts.

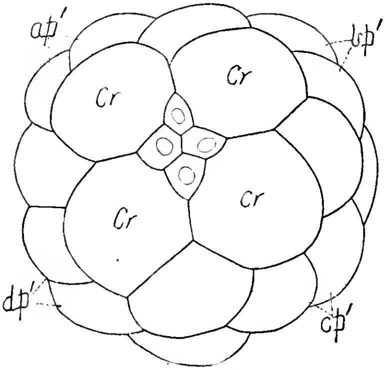

FIG. 8.—*Podarke*, forty cells from above; *ap'–dp'*, trochoblasts before division.

paratroch is differentiated, the embryo of *Amphitrite* contains about two hundred cells, and the mesoblast bands are composed of four cells each. At this stage the large invaginated entoderm cells are still solid, and have not formed the cavity of the alimentary canal, and the large prototroch cells are very prominent on the surface of the larva.

In general, the development of *Podarke* through these later stages agrees with *Amphitrite*, but from the standpoint of strictly cellular development important differences appear. The prototroch is at first completed by one "intermediate" cell (*i.e.*, one of the cells which originally lay between the arms of the cross), and later two more cells come into it from the lower hemisphere. These latter acquire their cilia very late, after the trochophore is fully formed. Cells from the upper hemisphere also migrate through the dorsal break in the prototroch, but instead of giving rise to a very small portion of the subumbrella ectoderm fully three-fourths of this latter arises from them. The descendants of *2d* form a narrow band of ectoderm around the proctodæum, and some of them pass into the wall of the latter. In Fig. 12 the portion of the trochophore behind the dotted line, *x*, *x*, is the only part whose ectoderm is formed by the *X* cells. I have been unable to discover any trace of a paratroch.

In *Podarke*, also, immediately after the formation of the fifth quartette of micromeres, which happens about ten hours after fertilization, the entoderm plate invaginates and rapidly gives rise to the alimentary canal, which is pretty definitely formed by the twentieth hour, and the trochophore must begin to feed very soon after this time. In *Amphitrite*, on the other hand, if I understand Dr. Mead correctly, the alimentary canal is formed much later, — about sixty hours, — and considerably later the embryos settle down to the bottom and begin to feed. While, too, there is a similarity in the method of formation of certain organs, there is a marked difference, becoming more and more noticeable as the development progresses, in the relative rate of development of different parts. The early stages of *Podarke* and of *Amphitrite* are passed through in about equal spaces of time, and both begin to swim immediately after reaching the sixty-four-cell stage, about five hours after fertilization; but, from this time on, important differences are to be noticed. Briefly stated, these differences may be said to be connected with the more rapid development of the descendants of *2d* and *4d* in *Amphitrite* than in *Podarke*. Figs. 5 and 6 show the relative development of these two sets of cells in early stages, and Figs. 9 and 10 show the proportional size of the descendants of these cells in a later stage. In the figure of *Amphitrite* the mesoblast consists of three cells on a side, while in *Podarke* there is only one, the third division of *4d* resulting in the separation of two small cells, which enter into the formation of the wall of the archenteron. Similar cells have been described for *Nereis* and *Aricia* by Wilson,[1] and for *Crepidula* by Conklin. Note in the figures the relative development of *2d* and its descendants (indicated by the letter *x*), when compared with one another and with corresponding cells of the other quadrants (indicated by Fig. 2). Evidently in *Amphitrite* the descendants of *2d* form a large part of the subumbrella, and the difference between the development of these cells here and in *Podarke*, a difference which was noticeable in earlier stages, has now become much more pronounced.

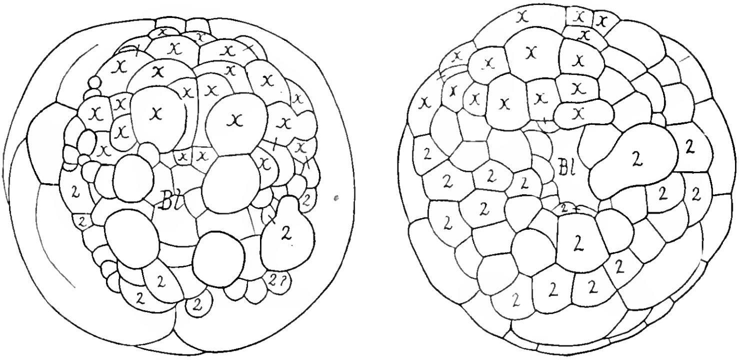

Fig. 9. Fig. 10.

Figs. 9 and 10 — *Amphitrite* and *Podarke*, respectively, in a late stage of segmentation; *Bl.*, blastopore.

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[1] Wilson, (*a*) "Cell Lineage of Nereis," *Journ. of Morph.*, vol. vi; (*b*) "Considerations on Cell Lineage," etc., *Annals of the N. Y. Academy of Sciences*, vol. xi, No. 1.

Extending the comparison into the later larval stages, we see that this difference still persists, and is even more noticeable than in the earlier. In the larva of *Amphitrite* of forty hours there is a very marked development of mesodermal and trunk ectodermal tissue, or, in other words, a very large amount of material derived from the descendants of *2d* and *4d* (Fig. 11, *Amphitrite* of forty-four hours, from Mead). *Amphitrite* has four well-marked body segments, with strong setæ on two of them (ectodermal structures which have arisen from *2d*), and, according to Dr. Mead, well-developed mesodermal tissue lining the gut and body wall. To be noted, also, is the relatively small development of the umbrella portion (the portion in front of the prototroch) and the extreme width of the prototroch itself. The large mucous glands and the "problematic bodies" of Mead occupy a large part of the surface of the umbrella, the former extending back into the subumbrella portion.

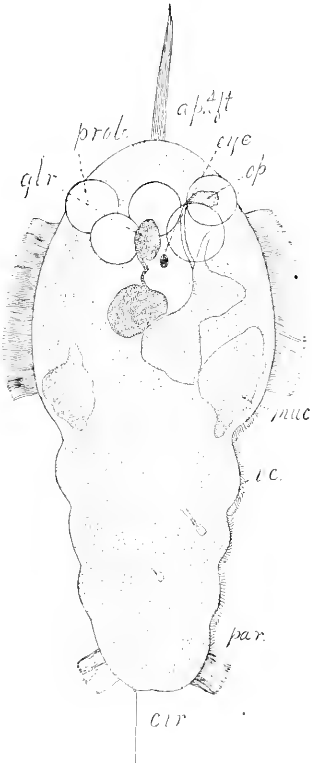

FIG. 11.—*Amphitrite* (after Mead), forty-four hours.

Turning now to *Podarke*, we find a very different condition of affairs. The most noticeable feature is the complete lack of any trace of segmentation. The subumbrella has elongated considerably, but shows no trace whatever of a division into somites. The umbrella is relatively much larger than in *Amphitrite*, a fact which is undoubtedly correlated with the unusual development of the cells comprising the cross in the former animal. I have not succeeded in finding any mucous glands, but what are probably identical with Mead's "problematic bodies" and Wilson's "frontal bodies" are found lying just in front of the small eye spot on either side (*prob.*, Fig. 12, *Podarke* of forty-two hours).

From their staining reactions I infer that in *Podarke*, as in *Nereis*, these organs have a glandular function. A ventral band of cilia is present, but I have not been able to discover any paratroch.

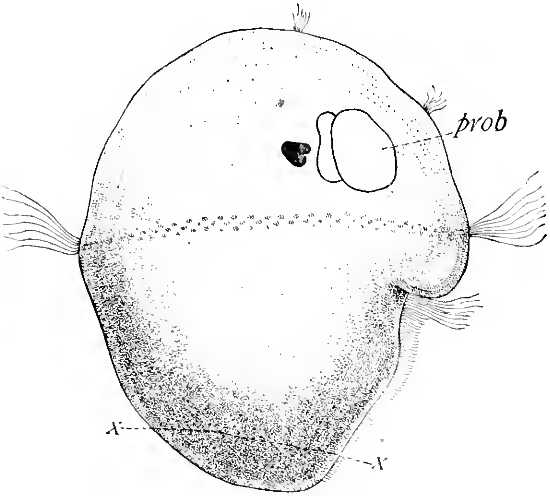

Fig. 12.— *Podarke*, forty-two hours; *prob.*, problematic bodies.

The enteron has grown and expanded so as to completely obliterate the body cavity. Owing to the slow development of muscular tissue on the dorsal surface of the larva, the body wall is composed of two very thin layers, ectoderm and endoderm, while the body wall on the ventral side is much thicker, and the amount of mesodermal tissue is evidently much greater. At present I am unable to state just what are the relations of the mesodermal portion of the larva of this age, but an investigation now in progress will undoubtedly clear them up. Certain it is that the amount of definite mesoderm is very small when compared with such a form as *Amphitrite*, for in an earlier stage of approximately twenty hours, when *Amphitrite* has already begun to elongate and the germ bands are composed of "a great many" cells (Mead does not state how many), *Podarke* has no more than two mesoblast cells on a side. The young larva is very muscular, undergoing all sorts of distortions when irritated; but this activity is produced by the *larval* mesoblast cells derived from *3d*, *3c*, and *3a*. These cells wander apart from one another, elongate, and form the larval musculature. As said before, I am not yet sure about the relations between these two sorts of mesoderm and the other body layers during the later larval stages; but I believe that it will be found that by far the larger part, if not the whole of the musculature of the trochophore, comes from this larval mesoderm, and that it is only as the larva elongates that the definitive mesoderm takes any important part in body formation.

For this comparison I have used *Amphitrite* as being the most convenient for the purpose, but exactly similar results would have been attained had I used any of the other annelids with unequal cleavage, whose cell lineage is at all accurately known. Of the annelids with equal cleavage, *Podarke* happens to be the only one which has yet been studied with any completeness; but, so far as one can judge from the figures given by Hatschek[1] for *Eupomatus*, Van Drasche[2] for *Pomatoceros*, and Mead for *Lepidonotus*, similar results would be obtained there, were the development fully studied. The trochophores of *Eupomatus* and *Pomatoceros* are thin walled, and with exactly the feeble development of ectodermal and mesodermal tissue that has been noted for *Podarke*. The same is true of *Hydroides*, as I am informed by Dr. Wilson, and of *Lepidonotus*, according to Dr. Mead. Connected with the absence of a large amount of yolk in these forms is the rapid development of the archenteron, so that the embryo begins to feed much earlier than in the other case.

How now are we to explain the difference in manner of cleavage which has been observed in these forms? Why does one egg divide with absolute equality, while others, little or no larger, divide very unequally? Within certain limits, undoubtedly, presence or absence of yolk does affect the form of cleavage. It can hardly be disputed that the peculiar form of cleavage found, for example, in the egg of the teleost is caused by an accumulation of yolk at the lower pole, or that a similar cause must be invoked to explain the form of segmentation of the egg of the bird; and earlier writers assumed almost universally that to the effects of yolk, either actually present, or exerting its influence through the action of heredity, must be ascribed the differences in the manner of cleavage. Rabl (**'79**), in his work on *Planorbis*,[3] finds a series of molluscs in which,

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[1] "Entwick. von Eupomatus uncinatus," *Arbeiten a. d. Inst. zu Wien*, 1885.

[2] Beiträge zu Entw. d. Polychaeten. Wien, 1884.

[3] "Entwick. der Tellerschnecke," *Morph. Jahrb.*, 1879.

at the four-cell stage, are found all gradations from four equal cells, through others with an accumulation of yolk in three, then in two, and finally in one cell, the yolk-laden cells being larger than the others. From these he concludes that unequal cleavage is caused first by an accumulation of yolk, and second by an unequal distribution of this yolk; *e.g.*, if there is a difference between "micromeres" above and "macromeres" below, it is because the yolk is collected at the lower pole of the egg; if a difference in size appears at the four-cell stage, it is because of an unequal distribution of the yolk among the blastomeres, etc. As far as I can discover, however, Rabl makes no attempt at explaining why the yolk should have this unequal distribution. While, as far as the first three cleavages are concerned, this explanation might suffice, the most superficial observation will show that it does not apply at all in later stages. There cells of the same size divide equally or unequally, and that, whether there is any yolk present or not. A glance at any plate of illustrations of a cell-lineage paper would show plenty of examples of this. In a discussion of this subject, Lillie (*loc. cit.*, p. 45) comes to the following conclusion: "Unequal cleavage is conditioned by the constitution of the segmenting ovum, and always means the precocious localization of an organ or set of organs in the larger cell. This organ may be the entoderm, in which case it is usually accompanied by yolk; but the inequality of the first two cells in the annelids and molluscs is the earliest visible indication of another differentiation, the larger cell containing the two somatoblasts." This explanation seems to me in perfect accord with the facts.

Turning now to equal cleavage, we might suppose it to be due to a lack of differentiation in the early stages, and, in fact, this explanation has been offered. Rabl[1] (**'76**) tried to explain the difference between the unequal cleavage of a mollusc and the equal cleavage of such a form as an ascidian by the assumption that in the former there is an early differentiation of parts manifesting itself in cells of different sizes, which does not appear in the latter. This early differentiation, being of distinct advantage in the struggle for existence, probably has developed

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[1] "Entwick. der Malermuschel," *Jenaische Zeitschrift*, 1876.

for that reason, while in the ascidian the equal cleavage indicates that differentiation begins much later.

From the facts which I have given above there can be no question that in *Podarke* there is as great a differentiation as in any annelid of the unequal type. The position of the "polar furrow," the appearance of the cell $X_{1, 2}$, and the perfectly regular formation of definite cells at definite times, all indicate this beyond the shadow of a doubt.

In a note published about a year ago[1] I ventured a suggestion concerning the meaning of this equal cleavage, and what I have seen since then confirms me in my original opinion. Accepting the principle laid down by Lillie, and further developed by Conklin (*Biol. Lectures*, 1896, 1897), that the initial size of a blastomere stands in direct relation to the size and *the time of formation* of the part to which it gives rise, I pointed out that in all probability the small size of *2d* and *4d* is connected with the slow development of the parts of the embryo arising from these cells. The facts upon which this conclusion is based will be evident from the comparison which I have made between *Podarke* and *Amphitrite*, where the slow development of ectoderm and mesoderm in the former (the descendants of *2d* and *4d*), and the rapid development of the corresponding portions of the embryo in the latter, are noticeable features of the ontogeny. In *Podarke*, also, a large part of the embryo, which is composed of definitive mesoderm in other annelids, is made up of the larval mesoderm mentioned above, — an additional reason for the slow development of *4d*. I believe further that absence of yolk is not alone responsible for the equality of the first three cleavages; but that the fact that the third division (forming the eight-cell stage) is equal is connected with the large size of the umbrella, relative to the subumbrella of *Podarke*, when compared with other annelids, and with the fact that a large part of the subumbrella ectoderm really comes from the umbrella, having migrated through the break in the prototroch.

We have already seen that at the sixteen-cell stage, and later, the largest cells in the embryo are at the upper pole.

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[1] *Zoöl. Bulletin*, No. 4.

To avoid misunderstanding, let me say here that in accepting this principle of Lillie's, and its application by Conklin, I can hardly follow the latter author in assuming as complete a cellular differentiation as he does. Inasmuch as we know that protoplasmic continuity undoubtedly exists between blastomeres, we are hardly at liberty to suppose that differentiation is complete at the time of cell formation. In using the term "differentiation" I mean that a certain amount of material destined to form a certain organ is accumulated at a definite place, even though this material at the time of its separation may be in an undifferentiated condition. For example, I do not believe that *at the time of formation 4d* is *necessarily* mesoderm, but that it is destined to develop into mesoderm; and, according as it contains a larger or a small amount of material, the mesoderm of the body will be large or small.

This evidently is but a limited application of the principle stated by Lillie, and it would be possible in the ontogeny of *Podarke* to find many illustrations of the law which might, perhaps, strengthen the position I have taken here. Inasmuch as differentiation in cleavage forms the subject of another lecture in this course, I shall not attempt to describe them, but will refer to Dr. Lillie's lecture for additional evidence in favor of the theory.

Quite recently, as said before, it has been shown that the cell *4d* is not, as was originally supposed, purely mesodermal, but that in some animals it constitutes in addition a certain amount of entoderm. It might be supposed that the large size of *4d* in these forms is due to this double function. Apart from the fact that in certain annelids, *Amphitrite*,[1] *Polymnia*, *Arenicola*, no such entodermal portion has been found, — and yet here the cleavage is very unequal, — is the further consideration that in no case is the amount of entodermal tissue there present sufficient to account for the difference in size. In *Aricia* there is but one small entodermal cell budded off from *4d* on either side; while in *Nereis*, though some three or four

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[1] I can hardly agree with Wilson that the small cell described by Mead as budded off from *4d* during its migration inward is entoderm, inasmuch as its permanent position is at the anterior end of the germ band.

are formed from each product of *4d*, they are very small, "scarcely larger than polar globules," and make up but an insignificant portion of *4d*.

Among the molluscs the same principle applies. We have already seen that it was the study of *Unio* which led Lillie to an enunciation of the law; and, from the results of his work on *Crepidula*, Conklin[1] has stated that "the initial size of the blastomere stands in direct relation to the size and time of formation of the part to which it gives rise." *Crepidula*, indeed, shows apparent exceptions, for the entodermal portion of *4d* is as large as the mesodermal. Here, however, there is a larval mesoblast, and the mesoderm derived from *4d* is, I infer from Conklin's figures, comparatively small in amount. In the earlier stages, also, *2d* is no larger than its sister cells of the same quartette; but from this cell arises the shell gland, which, according to Conklin, "appears late in the development." Other exceptions are *Umbrella*,[2] according to Heymons, and *Cymbulia*[3] (Fol), where *D* is the smallest cell of the four-cell stage; but I believe that careful study would show that here, as in the other molluscs, this small size is correlated with a slow development of organs. Certain it is that in *Umbrella*, at least, where, if we can trust analogy at all, the shell gland should arise from *2d*, this organ develops very slowly when compared with such a form as *Unio*. Unfortunately, too little is known of the later stages to enable us to draw any very certain conclusions.

Even in molluscs, where yolk appears to play an important part in determining the size of cells, the constancy in the arrangement of this yolk seems to me an indication that it is distributed according to the needs of the embryo, and is related to the greatest amount of protoplasm. If, for example, all the yolk could be removed from the egg of *Umbrella* without interfering with the normal development, I venture to suggest that, while the egg as a whole would be smaller, nearly the same size differences would appear as in the normal egg. In

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[1] *Biol. Lectures*, 1896, 1897.

[2] "Zur Entwick. von Umbrella," *Zeit. Wiss. Zool.*, Bd. lvi.

[3] "Sur le Développement des Pteropodes," *Arch. de Zoöl. Exp. et Gén.*, 1885.

other words, the greatest amount of yolk accumulates in the largest cell, and is not in itself the cause of that larger size.

To summarize: equality of cleavage is not an indication of lack of differentiation in the ovum, for definite cells appear at definite places and at definite times, just as accurately as in unequal cleavages.

Inequality of cleavage may be due to a limited extent to the accumulation of yolk; but a more important factor is the method of arrangement of the formative material. If an organ is to appear early in the ontogeny, or is very large, it will be represented in the cleavage by a larger amount of material than if it appears late, or is small when formed; and it is this accumulation of material within what we call a "cell" that leads to the "unequal," as opposed to the "equal" type. It is not merely the segregation of material in a single cell, but the segregation of a large amount of material, which produces the differences between the two types; *i.e.*, this difference is not qualitative, but quantitative.

SIXTH LECTURE.

THE CELL ORIGIN OF THE PROTOTROCH.

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Notwithstanding the wholesome reaction against the frequently reckless application of Baer's biogenetic law, larval and embryonic characters still hold an important place as criteria of the relationship of organic forms. The larval characters of the tunicates are significant indices of the relationship of these animals to the vertebrates, but perhaps no larval form has rivalled the trochophore or trochosphere in provoking phylogenetic speculation. The splendid work of Roule, *L'Anatomie Comparée*, which has appeared within the last year, bears witness to the importance which is at present attached to this larval type in establishing the relationship of such various forms as the annelids, rotifers, bryozoans, mollusks, etc. All these forms are grouped together by him into one branch of the animal kingdom, the *Trochozoa;* for, although the bodies of the different *Trochozoa* are highly variable in form and structure, they all may be derived in the normal embryonic development from the trochophore larva, which is constructed on one constant organic plan.

The features characteristic of the trochophore arise in various *Trochozoa* under very different conditions of embryonic development, and are possessed by some adult animals, for example, *Trochosphæra* (Semper). These facts give deep interest and wide bearing to the comparative study of the embryonic origin of the trochophore itself.

One of the essential structural characters of this larval form is the transverse ciliated fringe, *the prototroch* (Figs. 13–16), and it is the purpose of this paper to bring together and compare the observations of several workers who have employed the method of cell-lineage to determine the exact origin of this organ.

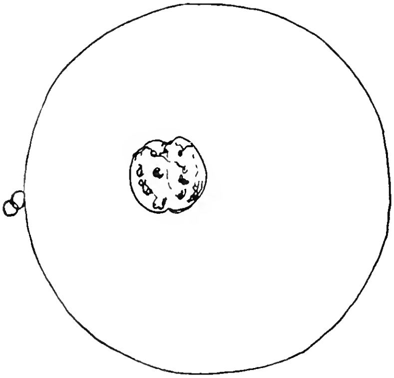

FIG. 1. — Left side of fertilized unsegmented egg of *Amphitrite*. Polar globules indicate the position of the animal pole.

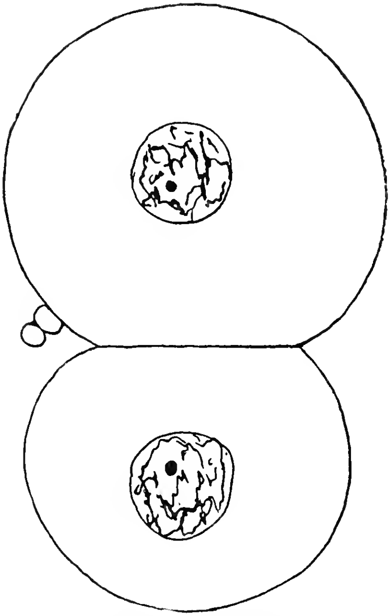

FIG. 2. — Left side of *Amphitrite* egg, 2-cell stage, oriented as in Fig. 1.

In order to establish a concrete example for comparison, let us proceed at once to the account of the origin of a simple and fairly typical prototroch, namely, that of the marine annelid *Amphitrite*.

The spherical egg is of medium size, about one-tenth of a millimeter in diameter, without much yolk, and is unprotected save by a thin wrinkled egg membrane. Fig. 1 represents the egg seen from the left side. Fertilization has taken place, and the polar globules remain attached. The succeeding figures represent the egg in the same position, unless otherwise stated. Fertilization takes place in the sea water, and the cleavage progresses so rapidly that the larva swims in a few hours. The first cleavage being unequal, the 2-cell stage is characterized by the possession of a larger and a smaller blastomere, which are represented in side view in Fig. 2. After this the cells divide with some irregularity in time and in the size of the resulting blastomeres, yet the irregularity is not so great as to obscure the rhythm of cleavage, which expresses itself in the following manner:

Each of the two cells divides, so that we have a 4-cell stage; then each of these cells divides, and so on, the number of cells increasing in geometrical progression until the egg consists of sixty-four cells. A distinct rhythm is observed also in the direction of cleavage, for the cleavage planes lie obliquely across the meridians of the egg. The cells in the 2-, 8-, and 32-cell stages are divided by cleavage furrows which take the general direction of the loxodromic curve in Fig. 3,[1] which may be rather loosely but conveniently defined as a left-handed spiral described around a sphere from pole to pole. The cells of alternate generations, namely, of the 4- and 16-cell stages, are cut by furrows which have an opposite direction, as represented in Fig. 4. These curves may be called, for convenience, left and right respectively, following the conventional nomenclature for the threads on a screw.

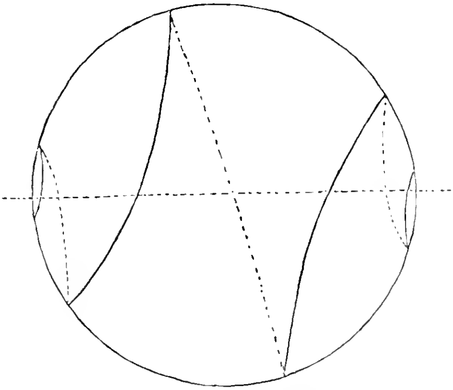

FIG. 3. — Loxodromic curve ("left-handed spiral") described about a sphere, showing the general direction of the furrows which cut the cells of the 2-, 8-, and 32-cell stages.

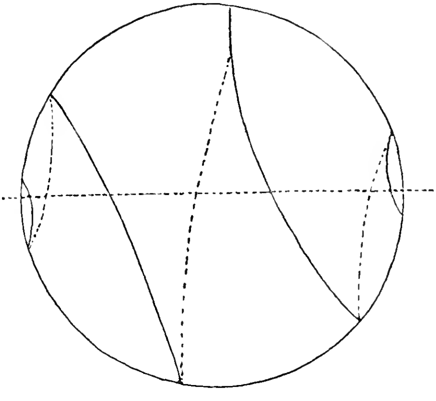

FIG. 4. — Loxodromic curve ("right-handed spiral"), having the general direction of the furrows which cut the 4- and 16-cell stages and many of those of the 64-cell stage.

The rhythm of division

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[1] In the diagram this curve cuts the equator at one point only. The curve will, of course, cut the equator at any point, if rotated.

ceases abruptly at the 64-cell stage. The cells of this stage are, of course, all of the same generation, the seventh, counting the ovum as one; yet, as a consequence of the obliquity of the cleavage furrows, the cells are not directly superimposed, but alternate with one another.

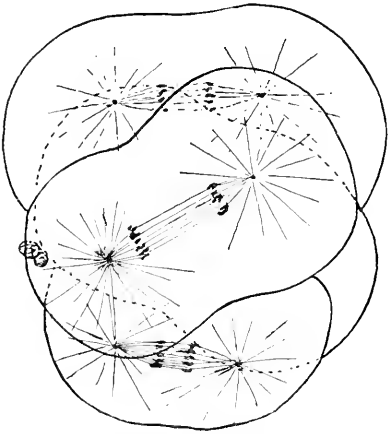

FIG. 5. — *Amphitrite*, left side, four cells getting ready to divide into eight.

For many reasons it is helpful to conceive of the egg as consisting of four quadrants, each of which is composed of the derivatives of one of the cells of the 4-cell stage. Any cell in one quadrant corresponds in origin and in position to a cell in each of the other quadrants.

Let us consider the egg in the 4-cell stage (Fig. 5) as composed of one quartette of cells about to divide. Four of the descendants of each of these cells will belong to the *primary* prototroch. In a similar way the egg in the 8-cell stage consists of two quartettes of cells, those of one quadrant meeting at the anterior pole, the position of which is indicated in Fig. 6 by the polar globules, those of the other meeting at the posterior pole. *The primary prototrochal cells are all descended from the cells of the anterior quartette*, and from that portion of the cells which is stippled in Fig. 6. The cells of this quartette, or, in the latter stages their descendants, constitute the anterior *hemisphere*, or *umbrella*, and the cells of the posterior quartette constitute the *posterior hemisphere*, or *subumbrella*. The hemispheres are separated in the drawings by a dotted line.

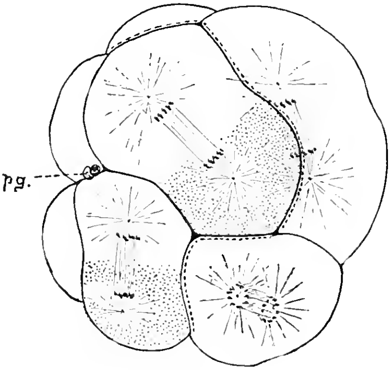

FIG. 6. — *Amphitrite*, left side, last phase of 8-cell stage.

After the next cleavage the 16-cell stage is formed, in which the cells are disposed in four quartettes. Fig. 7 represents the egg in this stage seen from the side, but slightly rolled to show all the eight cells of the anterior hemisphere. The polar globules lie in the middle of the hemisphere. The derivation of the cells from those of the 8-cell stage is indicated by the arrows, and the furrow between each pair of cells has the general direction of the loxodromic curve in Fig. 3. The dotted line indicates the boundary between the two hemispheres, and the heavy lines separate the quadrants. The four cells which are stippled in Fig. 7, one in each quadrant, are the *primary trochoblasts in the strict sense, for they contain all the material of the primary prototroch and no other material.*

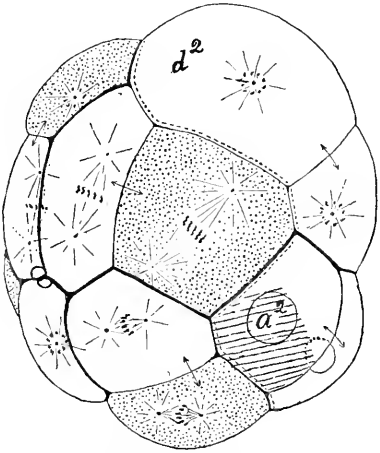

Fig. 7.—*Amphitrite*, from left side and top, 16-cell stage. Heavy solid lines divide the quadrants; dotted lines divide the anterior and posterior hemispheres.

By the division of the sixteen cells represented in Fig. 7 the 32-cell stage is attained (Fig. 8). It consists of eight quartettes (four cells in each quadrant), two of them, the third and fourth, from the animal pole, being derivatives of the primary trochoblast and stippled as in Fig. 7. (Fig. 8 really represents the beginning of the transition between the 32- and 64-cell stage. Two of the cells of the subumbrella hemisphere have divided. These cells, however, do not concern us especially.)

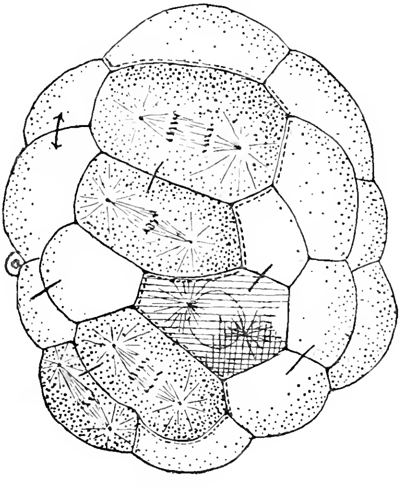

Fig. 8.—*Amphitrite*, left side, 32-cell stage.

The spindles in the trochoblast cells indicate the direction of their next division, which results in the 64-cell stage (Fig. 9). Fig. 10 is an apical view of the anterior hemisphere in the 32-cell stage, showing the four alternating quartettes of umbrella cells, and the uppermost quartette of sub-umbrella cells. The cells of the first quartette have the nuclei figured; those of the second are left clear; those of the third and fourth quartettes (primary trochoblast cells) are stippled. All of the thirty-two cells divide again, so that the egg is composed of sixty-four cells, arranged in sixteen quartettes, making sixteen cells in each quadrant. Four of these quartettes, the fifth, sixth, seventh, and eighth, stippled in Fig. 11, comprise the cells of the *primary prototroch.*

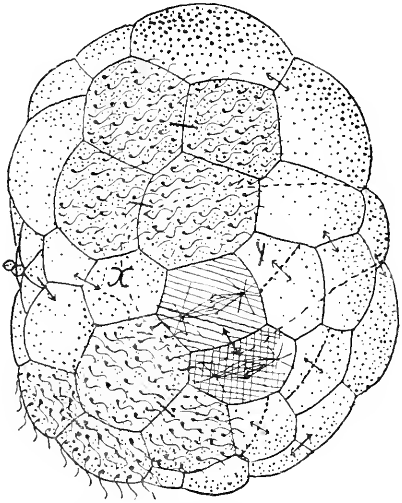

FIG. 9. — *Amphitrite*, 32-cell stage, from anterior end. View of umbrella or anterior hemisphere.

It is obvious that, in consequence of the regularity of cleav-

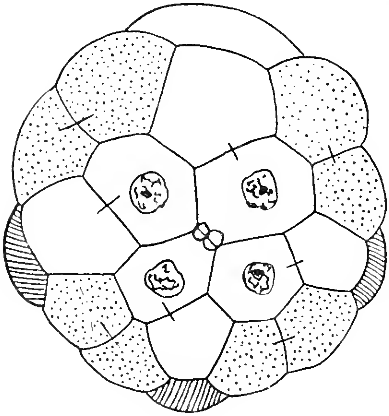

FIG. 10. — *Amphitrite*, from left side. Primary and secondary, shaded, respectively, with dots and lines.

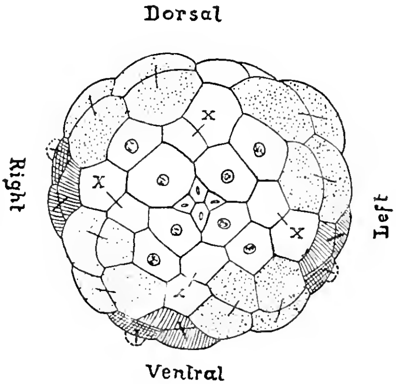

FIG. 11. — *Amphitrite*, beyond 32-cell stage, from anterior poles, slightly distorted to show all 25 prototrochal cells.

age, one-half of all the cells are descendants of the four upper cells of the 8-cell stage (the anterior hemisphere or umbrella), and that the sixteen primary prototroch cells all belong to this hemisphere.

By means of the primary prototroch, which consists of four isolated groups of cells, the egg, or trochophore, as we may now call it, swims about for a considerable time. In the course of a few hours, however, nine more cells, *secondary prototroch*, become differentiated and develop cilia, and these, together with the sixteen cells already described, constitute the *definitive prototroch*. As is shown in Fig. 9, these additional cells, which are shaded with lines, are disposed in such a manner that they fill the spaces between the isolated portions of the primary prototroch except in one quadrant. *Thus the definitive prototroch consists of twenty-five ciliated cells which lie in a nearly complete belt around the trochophore* (Fig. 13, p. 120).

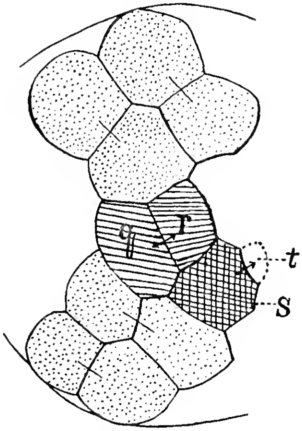

FIG. 12. — Diagram of prototrochal cells, seen from left side, in *Amphitrite*, *Clymenella*, and *Arenicola*. Two groups of primary and one of secondary prototrochal cells.

The primary and the secondary prototroch cells, although together they form a single larval organ, have different modes of origin, for the cells of the secondary prototroch are derived from the lower (posterior) quartette in the 8-cell stage, that is, from the posterior hemisphere, or subumbrella.

For convenience of description, we may assume the existence of a specific material which will become the secondary prototroch, and may locate this material in the subumbrella during the successive stages of cleavage. In the 16-cell stage the material occupies a portion of three of the cells of the third quartette, counting from the upper pole, which alternate with the primary trochoblasts. In Fig. 7, p. 117, the cell marked a^2 and partially shaded with lines is one of the three cells in question. The cell marked d^2 belongs to the quadrant which does not contribute any secondary prototroch cells. In the 32-cell stage the material is still in these cells of the highest quartette of the subumbrella. One of them is shown in the side view, Fig. 8, and all three in Fig. 9, shaded with lines. *These three cells may be called secondary trochoblasts, although they contain a small amount of material* (see unshaded portion of cell in Fig. 8) *which does not enter the prototroch.* With the next cleavage each of these cells divides so that the six cells, two in each of the three quadrants, contain the material of the

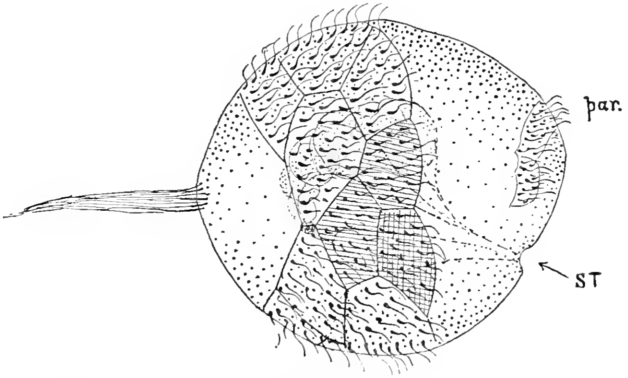

FIG. 13. — *Amphitrite* trochophore, showing apical tuft, prototroch, paratroch, stomadæma, and one arm of mesoblast consisting of six cells.

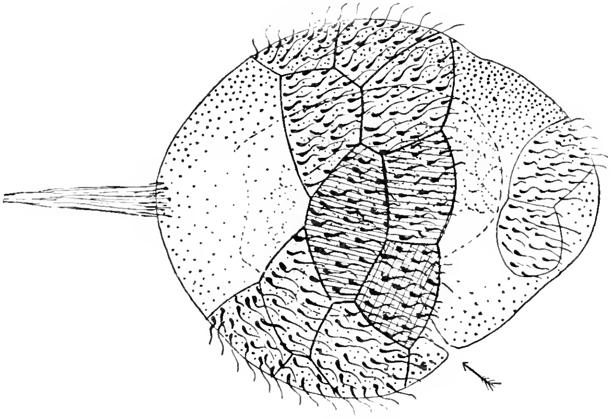

FIG. 14. — *Amphitrite*, left side; prototroch, paratroch; mouth at arrow; outline of archenteron stippled; mesoderm stippled.

secondary prototroch. These finally divide again in the direction indicated by the spindles in Fig. 10. Thus each of the three secondary trochoblasts is at last represented by a group of four cells, *q*, *r*, *s*, and *t*, in Fig. 12. Three of these, *q*, *r*, and *s*, are about equal in size, and constitute the *secondary prototroch.*

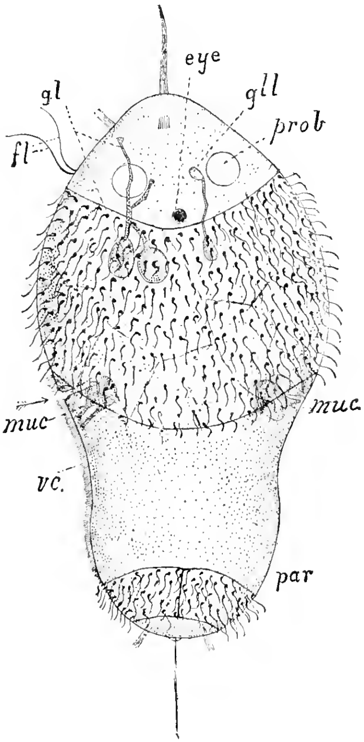

FIG. 15.—*Amphitrite.* Prototroch, paratroch closed ventrally. *muc. gl.*, mucous glands; *fl.*, flagella; *v.c.*, ventral band of cilia; *prob.*, problematic bodies which persist only as long as the prototroch.

The relative position of all the prototroch cells is shown in the apical view of the umbrella hemisphere in Fig. 11, which is slightly distorted and brings the secondary prototroch cells into sight. The shading accords with that of the previous figures. The primary prototroch cells are stippled, the secondary shaded with lines, and the minute products of the secondary trochoblasts are drawn with dotted lines. The unshaded cells in the upper margin between two groups of primary prototrochal cells interrupt the continuity of the prototroch. They correspond in origin, however, to the secondary prototroch cells of the three other quadrants. The remaining cell, *t*, Fig. 12 (dotted in Fig. 11), is small, divides again much later, and has an unknown destiny. The interruption in the continuity of the ciliated band in the mid-dorsal region is later obliterated by the approach and concrescence of the prototroch cells from either side. The completed prototroch, consisting of twenty-five cells, persists for five or six days, until the larva has developed several seta-bearing metameres.

Some of the later stages of the trochophore are represented in Figs. 13, 14, 15, and 16. For a considerable time the outlines of the component cells of the prototroch can be distinguished, but in late stages only a few of them can be made out in surface view, though tangential sections prove that they still exist. As the larva elongates, the prototroch extends over more surface, but the cells become thinner. The cilia are fine, not very long, and are thickly and evenly distributed. In the figures they are represented in a purely diagrammatic manner, being much too coarse and not nearly numerous enough to reproduce the actual appearance. When the larva begins to elongate, a second belt of cilia, the paratroch, develops around its posterior end. The paratroch consists of four ciliated cells which are derived from the cell marked d^2 in Fig. 7, p. 117, and, therefore, from that quadrant which did not contribute to the secondary prototroch. At first the four cells lie in a nearly straight row, but the end (lateral) cells gradually approach each other, and finally meet on the ventral side, as may be seen by comparing Figs. 13, 14, 15, and 16. Other cilia and flagella appear in tufts or bands over various parts of the body, while the larva is leading a free-swimming existence; but all these, including prototroch and paratroch, suddenly disappear when the larva, about five or six days old, settles to the bottom.

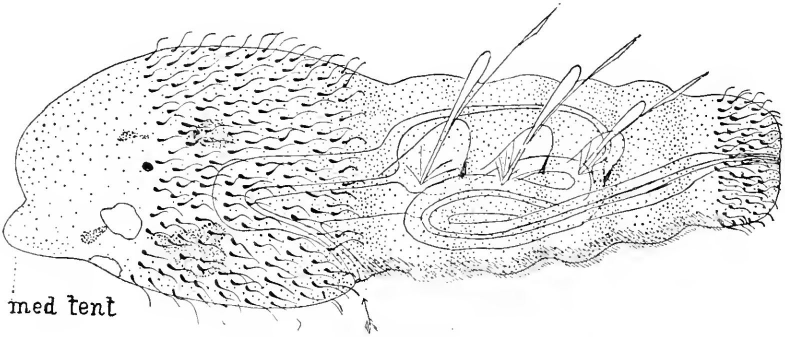

Fig. 16.— Lateral view of *Amphitrite* just before the disintegration of prototroch, paratroch, and mucous glands; *med. tent.*, median tentacle.

Figs. 13–16, when compared with the earlier drawings, illustrate the orientation of the body and the elongation of the trunk. The anterior end is at the left of the picture, the posterior end at the right; the dorsal side at top, and the ventral side at bottom. The anterior end is marked by the polar globules in the earlier drawings, and by the apical tuft of flagella in Figs. 14–16. The median tentacle grows out just ventral to the apical tuft. The posterior end is the middle of the area which becomes enclosed by the paratroch cells, and here the proctodæum is formed (Fig. 16). The "budding zone" lies just in front of the paratroch, and it is by the rapid growth in this region that the trunk elongates. The distance between the prototroch and paratroch, therefore, continually increases.

We stated at the beginning of this paper that, inasmuch as the trochophore is a highly representative larval form, the embryonic origin of one of its essential organs might prove to be of especial value, and the history of the origin of the prototroch in the one species which we have reviewed strengthens this conjecture and brings out some additional points of interest.

The prototroch is differentiated as a definite functional organ at a very early stage in development, when the whole body consists of less than 150 cells. It is composed of comparatively few cells (twenty-five), which are constant in number, in size, in relative position, and in mode of origin, in all individuals of the species. The portion of the larva differentiated as the prototroch is sharply marked off from the surrounding areas by the cell boundaries, and the cells in the immediate vicinity of the prototroch do not especially resemble the latter either in behavior during cleavage or in ultimate function.

Having thus taken our bearings, we find ourselves in this position. The trochophore is a larval form characteristic of a large group of animals, annelids, gasteropods, rotifers, etc., and the prototroch in one species, at least, arises always in the same manner from certain cells through a definite process of cleavage. Although the origin of the prototroch is comparatively simple, yet it is sufficiently complex to offer ample opportunity for variation, and by comparing its cell-origin in other animals we may hope to ascertain whether it always arises from the same cleavage cells. If it does so arise, then we may consider that its component cells are homologous, and, of course, the homology of the prototroch itself will thereby be strengthened. If it arises sometimes from one group of cells and sometimes from another, then its origin may be considered either an accident of the cleavage processes which are without morphological significance, or as indicating that the prototroch is not homologous in the forms examined. To illustrate: let us suppose that we were ignorant of the internal structure of the limb in the vertebrates. This organ, being characteristic of the higher vertebrate body, and having a general external resemblance in different forms, would seem to be homologous throughout the series. Now let us suppose that we have ascertained the origin and arrangement of the component structures of this organ in one species man. If, on comparison with other species, we find that the component structures of the limb have the same arrangement and arise in the same way, we may then consider these parts also to be homologous structures, and the homology of the entire limb is sustained thereby. If, however, we find that the origin of the component structures is essentially different in the various forms, we may then conclude either that the embryonic origin is without morphological significance, or that the limbs themselves are not homologous.

We will begin the comparison of the trochophore of *Amphitrite* with those of other annelids, assuming that the prototroch cells are homologous structures, and, in keeping with the illustration we have just given, we may expect to find:

First, that the particular features of the cell-origin of the prototroch in *Amphitrite* are not the result of the environmental conditions which are peculiar to this egg, but that the same origin obtains in other well-developed trochophores whose environment differs considerably from that of *Amphitrite*.

Second, that in those annelids in which the prototroch is more or less suppressed and develops at a late stage, the cells which we have identified as trochoblasts are small and slow in dividing.

Third, that in distantly related *Trochozoa* which possess a larval structure similar to the annelid prototroch some similarities obtain in the cell-origin of this structure.

Lastly, we might expect that annelid trochophores would be found in which the mode of origin of the prototroch is a modification of that in the assumed type, a modification analogous to that in the skeleton of the vertebrates, due either to the fusion or reduplication of parts.

Fortunately the few annelids in which the cell-origin of the prototroch has been worked out represent different families. *Clymenella*, for example, belongs to the *Maldanidæ*, a family not closely related to the *Terebelidæ* to which *Amphitrite* belongs.

The egg of *Clymenella* is nearly twice the diameter of that of *Amphitrite*, contains more yolk, and develops more slowly. The upper hemisphere is relatively smaller, and there are many minor differences in the rhythm of the cleavage and in the size of the cells. The trochophore is similar in many respects, but is less active. The definitive prototroch, however, consists of twenty-five cells, forming a nearly complete band around the larva. In the 16-cell stage we find the four primary trochoblasts which give rise, by two subsequent cell divisions, to sixteen cells, which constitute the primary prototroch. Each of the three secondary trochoblasts (32-cell stage) divides into a group of four cells, of which three are larger and subequal, while a fourth is small. The three larger cells in each group become ciliated and complete the prototroch, while the small one does not enter into it. *In brief, the origin of the definitive prototroch of Clymenella is identical, cell for cell, with that of Amphitrite.*

In *Arenicola cristata* we have a representative of another family of annelids, the *Arenicolidæ*. The eggs of this worm undergo their early development while encased in a gelatinous capsule, and the cleavage differs somewhat from that of *Amphitrite* and *Clymenella* in rhythm and in the relative size of the blastomeres; but Dr. Child has shown that the primary prototroch and, later, the definitive prototroch originate in precisely the same manner, cell for cell, as in *Amphitrite* and in *Clymenella*.

Though, I think, the cell-origin of the definitive prototroch has not been traced in other annelids, there are records of the mode of origin of the primary prototroch in several.

In *Lepidonotus* it is composed of sixteen cells, which arise exactly as in the annelids just described (see Figs. 6–11, stippled cells). These cells put out cilia very soon after they are formed, as they do in *Amphitrite* (Fig. 10). Later, three of the gaps between the separated portions of the primary prototroch are filled by other ciliated cells, while the mid-dorsal interruption persists just as in Fig. 11 of *Amphitrite*. It is not improbable, therefore, that the *definitive* prototroch of this worm has the same origin as that of other forms. *Lepidonotus* belongs to the *Aphroditidæ;* the eggs are comparatively small, and the cleavage is equal, so that up to the 64-cell stage it is not possible to distinguish one quadrant from another. This fact makes it of especial interest that the gaps in the primary prototroch are filled in only three quadrants instead of in all four, for, if the destiny of the secondary trochoblasts were dependent only upon their *position* in the cell complex, we might expect that the destiny of the corresponding cells would be the same in all four quadrants.

Podarke, which has been described by Treadwell, has a functional primary prototroch of sixteen cells of the same origin as in the other forms just described. (Mr. Treadwell has not yet published the history of the secondary prototrochal cells.) *Podarke* belongs to the family *Nereidæ*. The egg is even smaller than that of *Lepidonotus*, and the cleavage is equal.

Sthenelais, family *Aphroditidæ*, according to Treadwell, also has an equal cleavage, and a functional primary prototroch of sixteen cells is derived exactly as in the five annelids which we have already described.

We may now proceed to those annelids in which the mode of origin of the primary prototroch appears to be a modification of the type. Among these forms *Hydroides* is of special interest.

Wilson and Treadwell have shown that the functional prototroch of *Hydroides* consists of eight cells, two in each quadrant; but the origin of the primary prototroch in *Hydroides* is different from that of *Amphitrite* only in this respect, that the primary trochoblasts divide only once instead of twice. In the seven annelids which we have considered, the primary trochoblasts of the 16-cell stage, stippled in Fig. 8, p. 117, contain material for the prototroch only, and the phenomenon in *Hydroides*, as compared with that of other species, seems to indicate that a cell in which material of a specific structure only is segregated may divide once, twice, or perhaps more times. As long as *all* the descendants of the trochoblasts enter into the formation of the structure in question, it does not matter how many divisions take place. This view is further supported by what has been found in *Capitella*. According to Eisig's recent account, we find in this annelid a modification of the type in the direction of multiplication rather than reduction. Though sixteen cells are produced from the four trochoblasts, and all contribute to the prototroch, they undergo still further division before they become functional.

The first attempt to determine the exact cell-origin of the prototroch was made by E. B. Wilson on the egg of *Nereis*. He described what now seems to be a peculiar variation from the type. The four trochoblasts divide as in *Amphitrite* into sixteen cells, but four of these cells, the upper one in each quadrant, do not enter into the prototroch, but the other twelve cells do so. More cells are subsequently added, but their origin is uncertain. If this account is correct, *Nereis* is the only annelid in which the primary trochoblast has been shown to give rise to other than prototrochal cells.

We may now pass from a comparison of the prototroch and umbrella hemispheres in those annelids which represent the indirect type of development, and examine these features in annelids of more direct development, in which the trochophore is more or less suppressed.

The partially suppressed trochophore of *Scolecolepis* is developed from an egg which contains an abundance of yolk and is protected during its early development within the sand-tube dwelling of the worm. In the 8-cell stage the four anterior cells (upper quartette) are very small in comparison with the four posterior ones. All eight cells divide so nearly at the same time that a 16-cell stage results. Figs. 17 and 18 show that the four stippled cells, which correspond to the primary trochoblasts of other forms, are far smaller than any of the other cells ;

not only are they smaller, but they are the most tardy in dividing. After all the other cells have undergone another cleavage, these trochoblasts remain undivided. For this reason the next stage, which is shown in Figs. 19 and 20, consists of but twenty-eight cells instead of thirty-two. Three of these cells correspond exactly, of course, to the secondary trochoblast of *Amphitrite*, *Clymenella*, and *Arenicola*, and are therefore shaded as in Figs. 8 and 9, though they are very small in *Scolecolepis*. Thus in *Scolecolepis* the smallness of the anterior quartette of cells in the 8-cell stage, and the conspicuous minuteness of the trochoblasts, as well as their tardiness in dividing, correspond exactly with the small size of the umbrella of the trochophore and the small size and late development of the prototroch. This annelid, then, in its mode of development stands intermediate between such forms as *Amphitrite* or *Lepidonotus*, which possess a typical trochophore, and *Rhynchelmis* and *Clepsine* (Fig. 22), in which the development is direct and the trochophore undeveloped.

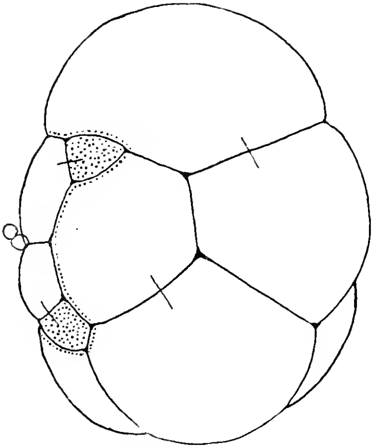

FIG. 17.—*Scolecolepis*, 16-cell stage, left side. Primary trochoblasts stippled; anterior hemisphere (umbrella) bounded by dotted line.

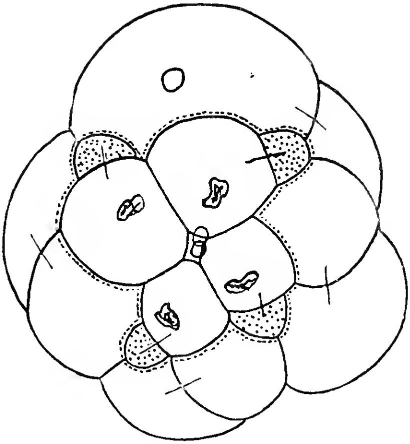

FIG. 18.—*Scolecolepis*, 16-cell stage, seen from anterior end.

Nereis Dumerillii (von Wistinghausen) approaches even nearer than *Scolecolepis* to the direct type. In the former the anterior quartette (umbrella cells) in the 8-cell stage (Fig. 21) are smaller still in proportion to the posterior quartette. With regard to the origin of the prototroch, however, it must be admitted that, according to von Wistinghausen's account, this organ is formed from cells which do not correspond with any of the trochoblasts of other annelids. His account places us in the same predicament with regard to the origin of the prototroch as with regard to the origin of the primary mesoblast cell. In respect to both, if the account is correct, *Nereis Dummerillii* is unique. In order to bring this annelid into line with the others, most writers on cell-lineage have expressed doubt, and rightly I think, as to the accuracy of the account in the case of the mesoblast. The origin of the mesoblast cell, however, is definitely described by von Wistinghausen, while he did not even pretend to work out the prototroch cell by cell. So we may, perhaps, be permitted at least to suspend judgment in regard to the discrepancy in the origin of the prototroch in this species.

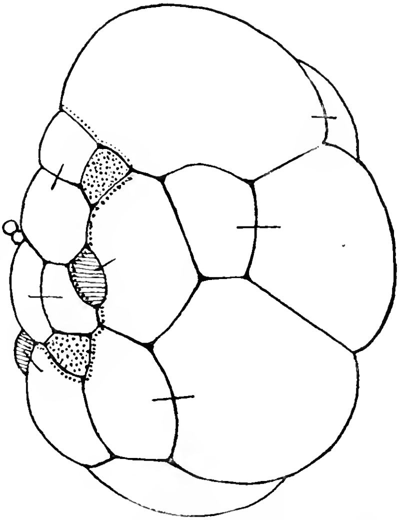

Fig. 19. — *Scolecolepis*, 28-cell stage, from left side. Shaded like figures of *Amphitrite*.

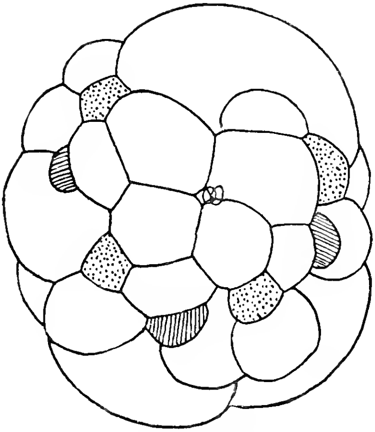

Fig. 20. — *Scolecolepis*, 28-cell stage, from anterior end.

In those annelids in which the trochophore is still farther suppressed, and which undergo a direct development, *e.g.*, in *Rhynchelmis* and *Clepsine*, the anterior quartette in the 8-cell stage is still farther reduced. Fig. 22 is from an unpublished drawing of *Clepsine*, 8-cell stage, given me by Dr. Whitman.

We may say in general of all the forms which we have reviewed, that when the cells of the anterior quartette are comparatively large and divide readily, the umbrella is large and the trochophore active. When, on the other hand, the cells of the anterior quartette are smaller and divide less rapidly, the umbrella is smaller and the trochophore less active. *Lepidonotus*, *Amphitrite*, *Nereis limbata*, *Clymenella*, *Nereis Dummerillii*, *Rhynchelmis*, and *Clepsine* form a series of annelids in which there is a gradual decrease in the relative size and karyokinetic activity of the anterior four cells and a corresponding decrease in the size of the umbrella and the activity of the trochophore.

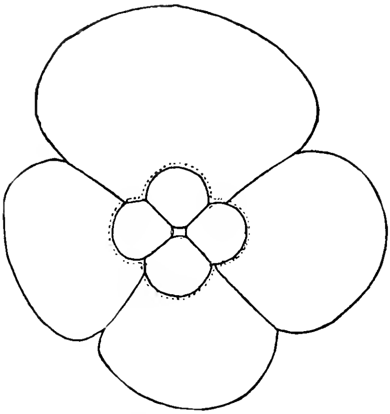

FIG. 21. — *Nereis Dummerillii* (after von Wistinghausen), 8-cell stage, from anterior end ("upper pole").

To sum up: the cell-origin of the prototroch has been completely worked out in three annelids, *viz.*, in *Amphitrite*, *Clymenella*, and *Arenicola*, which represent three distinct families. *In all three the origin of the prototroch is identical, cell for cell. In eight genera of annelids which represent seven families, the whole primary prototroch is formed from identical cleavage cells, the four so-called primary trochoblasts of the 16-cell stage* (Fig. 7), *and all the derivatives of these four cells enter into its formation.* These genera are *Lepidonotus*, *Podarke*, *Sthenelais*, *Hydroides*, *Amphitrite*, *Clymenella*, *Arenicola*, and *Capitella*. In all but two the primary prototroch consists of sixteen

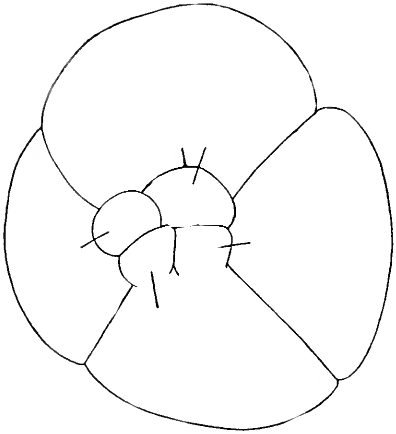

FIG. 22. — Eight-cell stage of *Clepsine* ——. Apical view from drawing furnished by Dr. Whitman.

cells — in *Hydroides* of eight, and in *Capitella* of more than sixteen. *Nereis limbata* differs from these forms in that four of the sixteen descendants of the trochoblast do not enter the prototroch, and *Nereis Dummerillii* is the only annelid, so far as I am aware, in which the primary prototroch has been thought to arise from other than the primary trochoblasts.

Passing now to other representatives of the branch *Trochozoa*, we have at present among mollusks more or less complete accounts of the cell-origin of the velum, in *Neritina* by Blochmann, in *Crepidula* by Conklin, in *Planorbis* by Holmes (preliminary), and in *Ischnochiton* by Heath (as yet unpublished). The most elaborate published account is given by Conklin in his beautiful work on the gasteropod *Crepidula*, and for this reason we may consider this form first.

Crepidula lays its eggs in capsules, which are protected under the shell of the animal. In two species "it is about four weeks from the time the ova are laid until the fully formed escape from the egg capsules," and probably much longer in other species. Hence it is not surprising that the velum becomes functional at a late period compared with the annelid prototroch, for the annelid begins to swim usually in a few hours after fertilization.

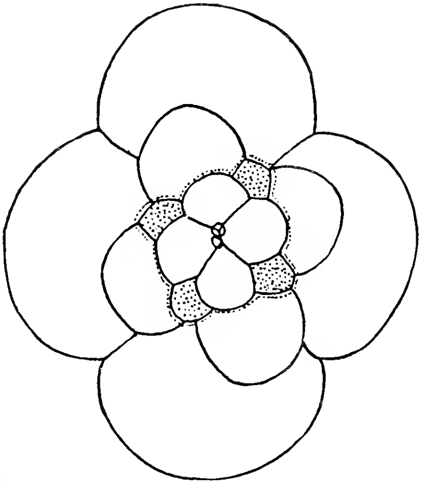

Fig. 23. — *Crepidula*, 16-cell stage, shaded like previous figures, viewed from anterior end.

The general type of cleavage in *Crepidula* is the same as that in the annelids. Fig. 23 of the 16-cell stage of *Crepidula* recalls in particular the same stage in *Scolecolepis*, whose eggs are protected like those of *Crepidula*. In both, the cells of the subumbrella, or posterior hemisphere, are very large and full of yolk, while those of the anterior hemisphere are very small and clear. The cells which correspond to the primary trochoblasts are the smallest of all, and the slowest to divide. In *Crepidula* and in other gasteropods these four cells are known as "turret-cells," but for the sake of uniformity in this paper I will call them primary trochoblasts. They remain undivided, or at most divide only once up to a very late cleavage stage. During these stages, however, they undergo a remarkable growth, so that finally, instead of being the smallest, they are the largest cells in the umbrella. It is the *destiny* of the primary trochoblast, however, which is of special interest to us. On this

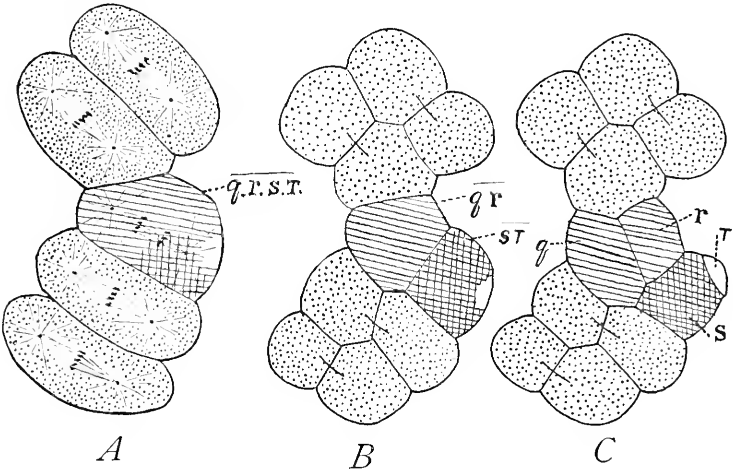

FIG. 24. — Three successive stages, representing the relative position of derivatives of the secondary trochoblast $\overline{q\ r\ s\ t}$ in several forms. Diagram approaches most nearly the condition seen in *Amphitrite*.

point Conklin says: "In four species of *Crepidula*, at least the two anterior ones form a portion of the preoral velum (prototroch), and this is probably true of the two posterior ones also." In other words, the primary trochoblasts in *Crepidula* have probably the same destiny as those of the eight genera of annelids mentioned on page 130.

We may now consider the behavior of the secondary trochoblasts in *Crepidula* — those three cells of the ideal 32-cell stage, most of whose descendants in *Amphitrite*, *Clymenella*, and *Arenicola* complete the prototroch by filling three of the gaps between its separate parts. *In Crepidula the secondary* *trochoblasts arise in exactly the same manner as in the annelids named, and most, if not all, of their descendants, enter into the prototroch.* This general statement should be qualified by an account of at least some of the details of the later cleavage, in order that it may give a fair idea of the degree of resemblance in question. For this purpose, and to avoid technical expressions, we will make free use of diagrams and arbitrary symbols.

The secondary trochoblast in each of the three quadrants lies between the primary prototroch cells, as in Diagram 24, which is from *Amphitrite* (compare Figs. 8, 10, and 12). It is represented by the compound symbol *qrst*. The phenomena are the same in three quadrants, but the corresponding cells in the fourth quadrant never enter the prototroch. In both *Amphitrite* and *Crepidula* each trochoblast *q*, *r*, *s*, and *t* divides obliquely in the same manner into two cells, *qr* and *st*, the former lying a little anterior to the latter (Fig. 24 *B*). Later, in both animals, each of these two cells divides in the reverse oblique direction, making a group of four cells, *q*, *r*, *s*, *t* (Fig. 24 *C*). In the annelid *q*, *r*, and *s*, soon after their formation, become ciliated and form a part of the prototroch in the manner already described, while *t*, a very minute cell, does not do so. In *Crepidula q* and *r* enter the prototroch ; *s* divides also, its upper product probably entering the prototroch, while the lower product may enter it or may not. The remaining cell, *t*, is very much larger than in *Amphitrite*, and at least part of it probably enters the prototroch, thus giving rise to the only probable discrepancy in the destiny of all the descendants of the primary and secondary trochoblasts in the two forms. What the significance of this discrepancy may be, I will not venture to say, but I wish to call attention again to the fact that the cell *t* in *Amphitrite* is exceedingly small, does not divide for a long time, and has the appearance of a vestigial cell.

It must be said here that Conklin thinks it possible, although by no means certain, that cells other than the trochoblasts enter into the formation of the velum, and in respect to many of the cells which we have already described there is also an element of uncertainty, since the velum of *Crepidula* does not become functional until a very late period of cleavage, so that it is almost impossible to trace its cell-origin accurately.

Blochmann, in his work on *Neritina*, was able to trace the descendants of two cells in the early cleavage into the velum. They could be traced the more readily because of a peculiar texture which distinguished them from the surrounding cells. These two cells in *Neritina* correspond precisely to the two lateral secondary trochoblasts of *Amphitrite* and the other annelids and *Crepidula* (the two lateral cells shaded with lines in Fig. 9). The primary trochoblasts of *Neritina* are also different from the surrounding cells. They are very minute, as in *Scolecolepis*, and were never observed by Blochmann to divide. As far as observations go, therefore, *Neritina* falls in line with the other forms which we have studied.

Conklin has called attention to the altogether characteristic appearance of the primary trochoblast, not only in *Neritina* and in four species of *Crepidula* which he studied, but in *Umbrella* (Heymons), *Urosalpynx*, and *Fulgar*. In all it is conspicuous for its small size, and does not divide for a comparatively long time.

It will appear from Holmes's preliminary paper that the origin of the velum in *Planorbis* bears a remarkable resemblance to that of the annelids. *Planorbis* resembles *Amphitrite* even more than it does *Crepidula* in this respect, *viz.*, the primary trochoblasts are large when formed, and the whole prototroch becomes functional at an early period. With regard to the cell-origin of the prototroch Holmes says: "The prototroch is formed from the [primary] trochoblasts previously mentioned, and the uppermost cells of the second quartette which form the tips of the arms of the cross [secondary trochoblasts, *q*, *r*, *s*, *t*, in the three quadrants]. Possibly other cells of the second quartette may take part in this formation ; it is certain that the third quartette has no share in the process."

I am greatly indebted to Mr. Harold Heath for a very complete account of the origin of the prototroch in *Ischnochiton*, and for his courtesy in allowing me to use his discoveries in this paper. The four primary trochoblasts are formed at the 16-cell stage, exactly as in *Amphitrite*, etc., etc. (Fig. 7, stippled cells). These four cells divide into sixteen, all of which enter the prototroch; so far, therefore, the resemblance is complete. The secondary trochoblast also arises in the 32-cell stage, precisely as it does in *Amphitrite*, *Clymenella*, and *Arenicola* (Fig. 8, cell shaded with lines). All of the derivatives of this cell enter the prototroch in *Ischnochiton*, and all but one very minute cell product do so in the annelids named (Figs. 10–12).

The essential features of Heath's account can be most effectively presented by means of diagrams. Fig. 25 represents a lateral view of the prototrochal cells of one side in three stages of development, and the various cells are shaded like those in Fig. 24 (*Amphitrite*). Diagram *A*, Fig. 25, shows the condition of the trochoblasts at about the 32-cell stage. It shows two of the four groups of primary trochoblasts (stippled), each group consisting at this period of two cells, as in *Amphitrite*, etc., Figs. 8 and 24, *A*. Between these cells lies the trochoblast marked *q*, *r*, *s*, *t*. The clear cell marked *x* is a trochoblast, and its two derivatives in each quadrant enter the prototroch,

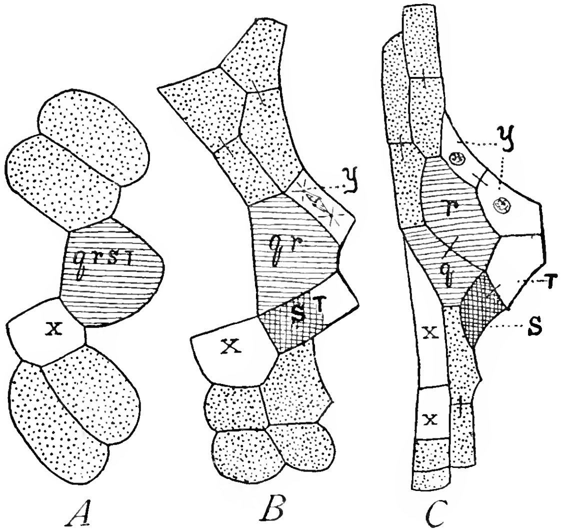

FIG. 25. — Represents trochoblast as seen on left side in *Ischnochiton* (Heath). Three stages shaded like previous drawings. Unshaded cells are not part of the prototroch in the other forms examined. See p. 132.

as may be seen from Diagram *C*. To show the position of this cell in *Amphitrite*, I have marked it *x* in Fig. 10 in one quadrant, and in each of the four quadrants in Fig. 11. If my observations on *Amphitrite* and *Clymenella* and those of Child on *Arenicola* are correct, this cell does not enter the prototroch in these annelids. However, I have never followed its career in either form, except in the posterior quadrant, and here Heath has called my attention to the fact that one of the derivatives of this cell in *Amphitrite* is excessively minute (not as large as a polar globule), and has the appearance, therefore, of a "vestigial cell." As the matter stands at present, this is, I believe, the first important well-sustained discrepancy in the origin of the prototroch, in all the *Trochozoa* which have been studied, and for this reason I would recommend the investigation of the destiny of this cell in the annelids as a fruitful problem in cell-lineage.

Diagrams *B* and *C* in Fig. 25 show that all four derivatives of the secondary trochoblast *q*, *r*, *s*, and *t* enter the prototroch. This is true in three quadrants. It will be recalled that in the three annelids in which these cells are known, *t* is not a prototrochal cell, but is minute and of a vestigial appearance. It is shown in Figs. 11, 12, and 24.

The cell *y* in Fig. 25, Diagram *B*, remains to be discussed. It divides once, and the two daughter-cells take their position in the prototroch, as seen in *C*. This cell *y*, like the cell *x*, does not enter the prototroch in *Amphitrite* (see Fig. 11), *Clymenella*, and *Arenicola*. It is small in *Amphitrite*, but not minute enough to suggest that it is a vestigial cell, though one of its products is distinctly of this appearance. In respect to the cells y, y', t, and s, however, there is an important fact to mention. They are, as far as could be observed, *not ciliated*, and probably serve, Heath suggests, as a sort of supporting layer. For this reason it may, perhaps, be a question whether they should be considered part of the prototroch. If we throw them out, however, there arises another discrepancy between this prototroch and that of the annelid, for in the latter the cell *s* is undoubtedly a part of the prototroch and is ciliated.

For the sake of convenience in comparing the cell-origin of the prototroch in the various annelids and gasteropods in which it has been investigated, we assumed the annelid *Amphitrite* as the type. In this form the prototroch arises from two sources: (*a*) Four cells, primary trochoblasts, one belonging to each quadrant of the umbrella hemisphere, give rise to a functional "primary prototroch," which is for a time the only ciliated portion of the larva. The four cells in question are formed at the 16-cell stage. (*b*) Three cells, secondary trochoblasts, belonging to the right, left, and ventral quadrants of the subumbrella hemisphere, give rise, later, to the rest of the prototroch. These three cells are formed at the 32-cell stage. The whole prototroch is at first interrupted in the mid-dorsal region, but ultimately grows together.

Almost all of the positive evidence furnished by the comparisons which we have drawn points, I believe, to the conclusion, first, that the component cells of the prototroch are homologous in the various types studied, in the same sense as are the component bones in the skeleton of the vertebrate limb. If this conclusion is correct, the homology of the whole prototroch is placed on a firmer basis, and the evidence of the relationship of the two diverse groups of the *Trochozoa* — annelids and mollusks — is likewise strengthened. As a corollary of this conclusion, it would appear that, since certain cells form the same organ under environmental conditions which are unlike, the environmental factors do not have the importance in directly shaping development which is sometimes claimed for them.

Some of the evidence, however, seems to controvert this conclusion. I refer particularly to the discrepancies between Wilson's account of *Nereis limbata* and all other accounts in regard to the destiny of four cells, and to that between Heath's account of *Ischnochiton* and all the other forms in regard to the destiny of four or more cells.

Considering the practical difficulties, we may say with regard to the value of different observations that positive evidence that a certain cell enters the prototroch should have more weight than the negative evidence that it does not, unless a different destiny for the cell is established in the latter instance.

A re-investigation of the prototroch focussed upon these discrepancies would be of great value in determining the trend of the whole mass of evidence, whether it is for or against the doctrines of the homology of cleavage cells, of the significance of cleavage as a factor in differentiation, and of the preorganization of the egg itself.

SEVENTH LECTURE.

RELATION OF THE AXIS OF THE EMBRYO TO THE FIRST CLEAVAGE PLANE.

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WHAT is the meaning of cleavage? is the constantly recurring question of the embryologist. Is cleavage a differentiating process in development, or is it a process which may run on independently of differentiation, sometimes coinciding with it, sometimes not?

Is the egg a mosaic, as Roux maintains? Does the first plane of cleavage determine the axial position of the embryo, or is the egg already definitely oriented with respect to the future embryo before cleavage begins? Is the egg an isotropic body to be converted gradually into a definite mosaic as cleavage advances, or are the blastomeres all endowed with the same potentialities, the fate of each being settled by the position it happens to hold?

Does the division of the egg by the first line of cleavage mean the separation of the parts destined to become the right and left sides of the bilaterally symmetrical animal, or may this first cleavage run in any direction, and have no fixed and necessary relation to the future embryo?

The well-known discoveries of Newport ('54) and of Pflüger and Roux in '87 and '88 seem to point toward some general law controlling the phenomena of cleavage.

In 1891, while studying the development of *Batrachus tau* at the Marine Biological Laboratory at Wood's Holl, Professor Whitman directed my attention to the fact that the egg was a favorable one for testing the conclusions of Roux. An egg provided with an adhesive disk, by means of which it is held constantly in a fixed position, seemed to offer just the condition required for testing the mosaic theory of cleavage.

The results have been reported in the *Journal of Morphology*, vol. v, p. 498. In only *three* cases out of twenty-three was there coincidence of the first line of cleavage with the median plane of the embryo.

In 1892 Born[1] called attention to my paper and pointed out what he regarded as the "sources of error" in the case. His criticism was accepted by Roux,[2] who adds: "Only slight errors of experiment are required in order to obtain almost equally incorrect figures in the frog's egg."

Born doubts the validity of the results, because of the long period of time that elapses between the appearance of the first furrow and the outline of the embryo. During this period of six days, slight changes may occur to account for the "abweichende Resultat." Born also finds difficulty in accepting the assertion that "the adhesion of the yolk to the egg-membrane prevented rotation," and says that "such adhesion would certainly hold good only for the first stages."

Weysse,[3] in 1894, believing that if there was adhesion of the yolk to the membrane it could be demonstrated in sections of the egg, made a careful examination, but found no trace of any such attachment. He concludes, with Born, that "during the six days mentioned abundant opportunity is furnished for a rotation of the yolk within the egg-membrane."

These experiments have been twice repeated since '91, with essentially the same result. In order that the possible sources of error may be fairly estimated, it seems desirable, in giving the results of my later experiments, to repeat and perhaps expand the brief description of the toadfish egg given in '91.

The egg is large, — 5 mm. in diameter, — and as the blastodisk always develops at the free pole of the egg, the early cleavage lines can be observed with a hand-lens without dis-

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[1] Merkel and Bonnet, Erg., Bd. I, p. 502.

[2] "Beiträg zur Entwickelungsmechanik des Embryo," *Anat.*, Heft 7, p. 313.

[3] Proc. of Amer. Acad., vol. xxx, p. 308.

turbing the egg. The egg is hardy, and develops well in shallow dishes holding sea-water about two inches deep.

The membrane of the egg has a peculiar adhesive disk, about 3 mm. in diameter, which has a constant position, with the centre of the disk at the vegetative pole, directly opposite the micropyle. By means of this disk the egg is firmly glued to the supporting surface, usually the underside of a rock, the inside of a broken jug, tin can, a piece of stovepipe, or even an old boot-leg. The disk consists of a transparent secretion, which becomes opaque and gluey on contact with water. It is of nearly uniform thickness, and is closely applied to the egg-membrane everywhere except for a narrow margin which projects all around as a thin rim (Fig. I). The disk is saucer-shaped, and only a little thicker than the egg-membrane itself. I have been able to separate it from the membrane in the case of eggs hardened before attachment. As the egg is generally fastened to more or less plane surfaces, it appears strongly flattened on the side of attachment, as described by Dr. Ryder and as shown in Fig. II.

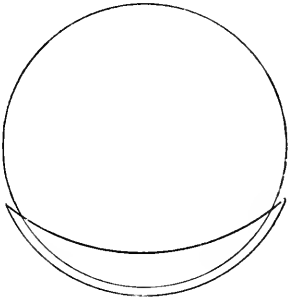

FIG. I.

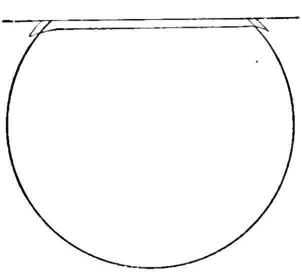

FIG. II.

That the egg-membrane, with its adhesive disk, does not change position after attachment is, then, very certain; but is the egg itself also fixed within the membrane, and does it maintain a fixed position for the time required to reach an early stage in the axial differentiation — six or seven days after fertilization? or is it liable to rotate and get displaced, and so invalidate the results obtained?

In the experiment of '91 the eggs were allowed to flow from the opened ovary of the fish into a glass dish containing just water enough to cover them. After becoming fixed to the bottom they were fertilized. The side of the egg resting on the glass was much flattened, and the fact that the blastodisk retained its position at the free pole of the egg when that pole was directed upward, led to the statement that "the adhesion of the yolk to the egg-membrane, as it rested on the disk area, prevented rotation."

In the experiments of '96 and '98 the eggs were allowed to fasten themselves on pieces of glass which were inverted and which rested on supports within a large dish of sea-water. Consequently the cleavage and development had to be observed from below, by looking up through the bottom of the glass dish.

In this case the eggs were in their natural position, that is, in the position in which they are usually deposited by the fish when making her nest under a flat stone.

It makes no difference, then, how the egg is attached, whether on the roof of the nest, or on the side or on the floor; the axis of the egg is always *perpendicular to the plane of attachment.* The fish usually fixes its eggs so that they hang from the surface above; but if they are deposited in a piece of stovepipe, so as to cover the upper half of the concave inner surface, the position of the axis of the egg will vary at all angles between a vertical and horizontal position.

If the eggs are dropped loosely into a glass dish containing very little water, some of them will fall so as to become quickly fixed to the bottom of the dish, while others may fall so that the adhesive surface fails to come into contact with the glass, and so soon loses its adhesive property, leaving the egg free to roll. The axis of the fixed eggs will be vertical, but the free pole will face upward instead of downward, as it does when the egg is placed on the roof of the nest, or on the underside of a glass plate. It is evident, therefore, that gravity has no decisive influence in determining the direction of the egg axis. In other words, the egg has a definite and constant position in its membrane, the vegetative pole always lying at the centre of the adhesive disk, while the animal pole and micropyle coincide on the opposite side.

This is not saying that the egg cannot be forced to change its position within the membrane. The eggs that fail to become attached to the bottom of the dish are liable to be rolled about, and the movement may be sufficient to cause some shifting in position. The important fact here is, that, whatever be the place of attachment, whether above, below, or on the side, the axis of the egg always maintains the constant relation of being perpendicular to that point, and there is no inherent tendency to assume a *vertical* position, as is the case in most pelagic fish eggs.

There is, then, no cause for rotation within the membrane unless it be supplied artificially from without. The egg remains in the position in which it becomes fixed, neither rotating nor becoming displaced except as the result of rough treatment.

In the experiment of '98 great care was taken to guard against any possible disturbance of the eggs. The pieces of glass with the adherent eggs were arranged conveniently for observation before they were fertilized, and the water changed from time to time by means of a siphon, so that no disturbance seemed possible.

If the egg does not rotate during the early stages of development, it may be asked, What is the *nature* of the adhesion that prevents rotation?

In regard to this matter Professor Whitman assures me that adhesion of the yolk to the membrane is a very general phenomenon which he has noticed in the egg of *Necturus*, *Amblystoma*, frog, newt, pelagic fish eggs, and even in the small eggs of many annelids. Speaking of pelagic fish eggs, he says: "It is only necessary for an egg to be left at rest for a few minutes in order to see how readily it adheres to its membrane. I have often taken advantage of this adhesion to roll over and hold them some moments by the aid of needles, with the lower pole uppermost. Others, I am sure, have done the same thing while studying the cleavage." He also adds that in the case of the toadfish "the strength of the adhesion is probably due to the relatively large weight of the egg and the large surface of contact." The perivitelline space in this egg is so slight as to be difficult of recognition, and the extent of the surface in actual contact with the membrane is correspondingly very great as compared with small eggs with ample space between the yolk and the membrane. The conditions, then, are most favorable for strong adhesion.

Born asserts that "such an adhesion cannot hold for more than the first stages." As this statement does not rest upon observation of the toadfish egg, it can have no value beyond that of conjecture suggested by experience with other eggs. It is quite true that six days gives *ample time* for any number of rotations, but it is not a question whether there is *time* for rotation, but *whether rotation actually occurs*, and that can only be determined by close study of the developing egg, not by sectioning of hardened and imbedded eggs, nor by any amount of experiments on frogs' eggs.

In one very important respect the toadfish egg is far superior to the frog's egg for the study of the question here considered, since it can be observed under perfectly normal conditions, without resort to those artificial means of fixation or marking which are necessary in a frog's egg, and which must always cast some doubt on the reliability of the results.

In the experiment of '91 the method of determining the relation of the first cleavage plane to the axis of the embryo was as follows: After the eggs had become fixed by the adhesive disk to the bottom of a glass dish they were artificially fertilized. The blastodisk appeared on the free pole of the egg, where it was easily watched by means of a lens. The eggs were plotted on paper, each egg being represented by a circle (Fig. III), and the paper and the dish containing the eggs oriented by fastening a label on each in the same relative position. When the first line of cleavage appeared the direction was indicated in the circle representing the egg, by the diameter, and when the axis of the embryo became visible, that was indicated by an arrow drawn across the same circle.

The first furrow appeared seven hours after fertilization, and on the seventh day the axis of the embryo could be distinctly seen as a light streak in the blastoderm.

The result of this experiment is seen by an examination of the circles given in Fig. III. Of the twenty-three developing embryos, *three* show coincidence of the axis of the embryo with the first cleavage plane. There is no case of exact coincidence with the second cleavage plane. Fourteen of the embryos have the head directed towards the right of the first line of cleavage, the axis of the body being at an angle with the first cleavage plane of from 30° to 70°. In the remaining six the head was to the left of the first cleavage plane, the angle varying as before.

In the experiments of '96 the main purpose in view was to settle the question of rotation of the egg within its membrane and to find out, if possible, whether gravity had anything to do with the position of the blastodisk. The eggs attached to pieces of glass were fertilized and the glass plates placed in various positions, so that the axis of the egg in different cases was vertical, horizontal, and inclined at various angles.

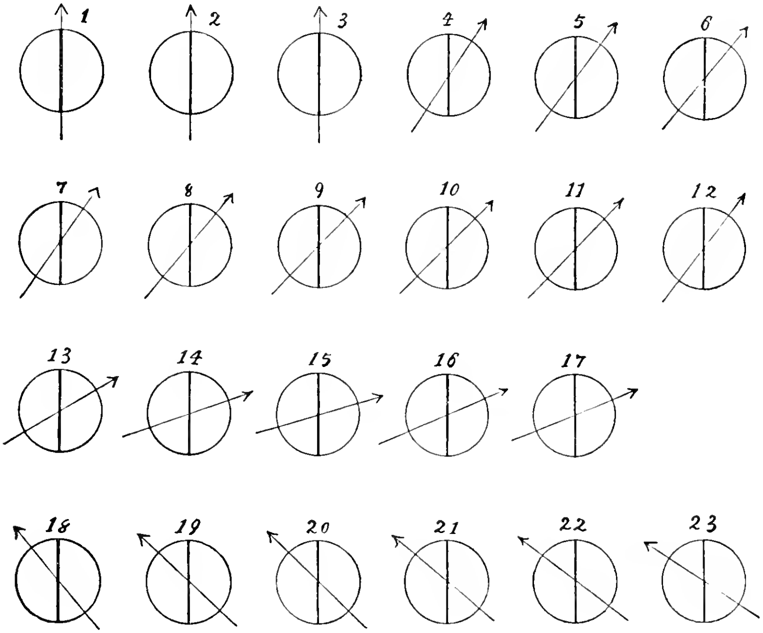

FIG. III.

It was observed that the small oil globules within the egg were always found uppermost in whatever position the egg was placed, in one case being directly under the blastodisk at the animal pole, and again directly underneath the adhesive disk at the vegetative pole, or halfway between these poles *on the side that was uppermost.*

As for the blastodisk itself, *that retained its position at the free pole of the egg, near the micropyle, in whatever position the eggs were placed.*

The position of the eggs was plotted in the case of ten eggs attached in the inverted position.

The coincidence of the first furrow with the embryonic axis was observed in three out of ten cases (Fig. IV).

The third experiment was made during the summer of '98. A broken bottle from the eel pond was brought into the laboratory containing a toadfish and a few eggs which had been already deposited by the fish.

On the morning of July 6 the fish was opened and the mature eggs allowed to flow out of the ovaries and fasten themselves on pieces of glass.

About one-third of the eggs set free from the ovaries became fixed to three glass plates. The rest failed to adhere and so were of no use in the experiment. Two minutes was sufficient time to allow for the eggs to become fixed. The plates were then inverted and placed on supports in a large shallow dish with sufficient sea-water to cover them.

The eggs were then fertilized, and soon after the water was siphoned off by means of a rubber tube and fresh sea-water quickly supplied. The dish was so placed that the eggs could be observed from below.

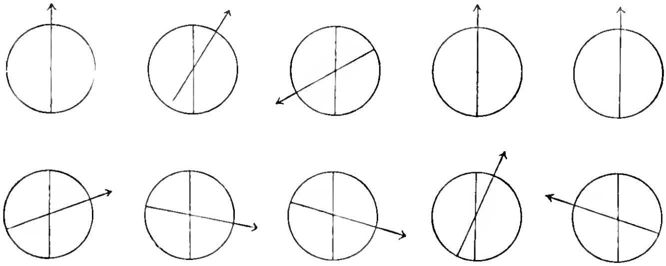

FIG. IV.

At 4.30 P.M.—eight hours after fertilization—the first line of cleavage was noted in fourteen eggs, and the direction of the furrow indicated on the paper oriented as in the earlier experiments.

On July 12, six days afterward, during which time the dish containing the eggs was not moved, the axis of the embryo was observed, and its direction indicated as before. The result is seen in Fig. V. Of the fourteen eggs only *two* show coincidence of the first cleavage furrow with the embryonic axis (Fig. V, 5, 11).

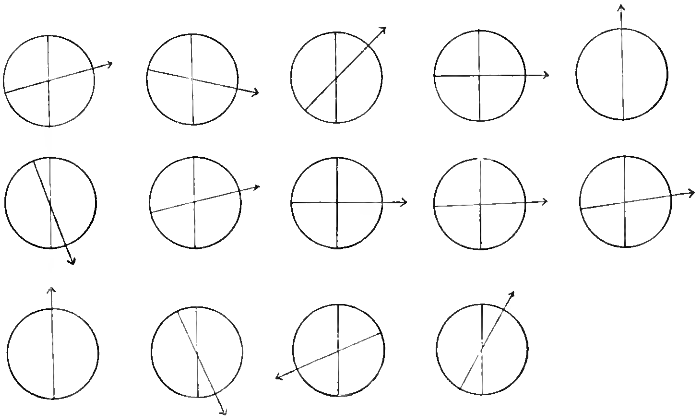

FIG V.

Similar experiments with the eggs of *Amia* have been made by Eycleshymer, '96.[1] Observations were made on three sets of eggs and the results are given in Fig. VI.

Twenty eggs developed normal embryos, and *two* showed coincidence of the first plane of cleavage with the axis of the embryo (Fig. VI, 2 and 6). The following is quoted: "The egg is oval in profile view, measuring in its longest diameter, including the membrane, 2.5 to 3 mm.; in its shortest, 2 to 2.5 mm. The freshly deposited egg is firmly fixed to the object with which it first comes in contact by means of the threads of the villous layer, which are elongated over the lower hemisphere of the egg-membrane.

The eggs still attached to blades of grass or rootlets are placed in shallow watch-glasses and held in position by weighting with small pebbles. The watch-glasses are then placed

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[1] *Journ. of Morph.*, vol. xii, p. 344.

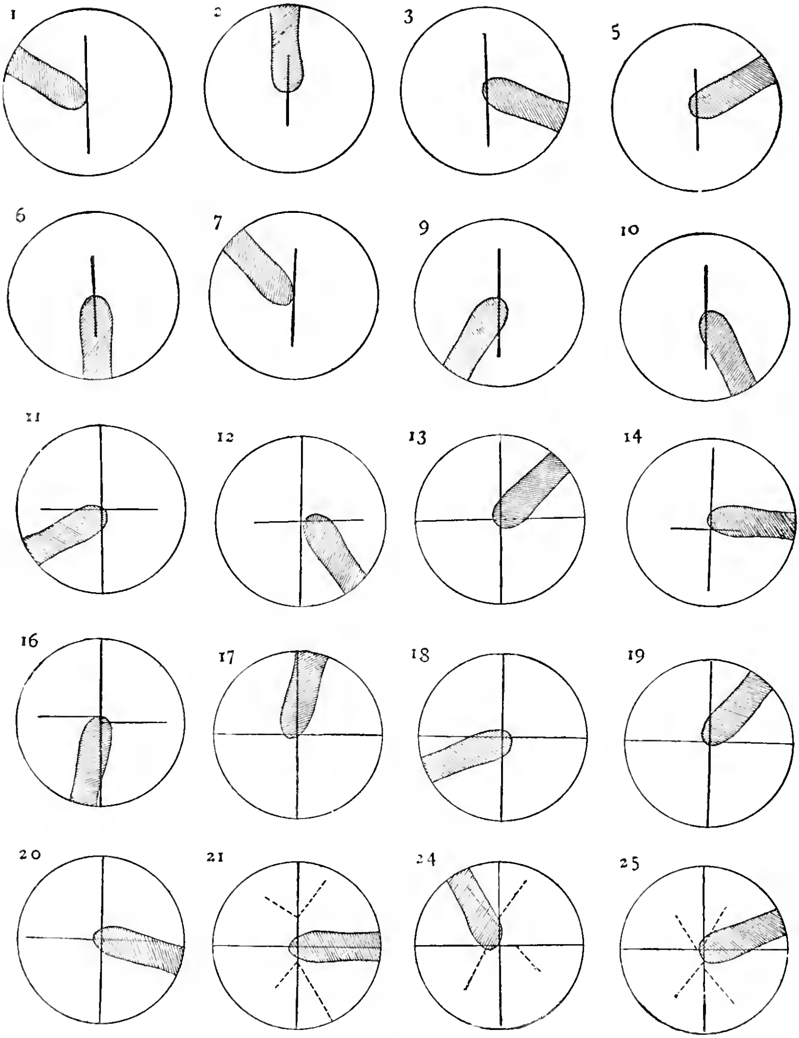

Fig. VI.

upon a mirror fastened to the stage of a dissecting microscope. We could thus observe the changes occurring on opposite sides of the egg without disturbing its position.

The elongated form of the egg of *Amia*, in a closely applied envelope, prevents rotation about its minor axes. It is, therefore, a favorable egg for ascertaining what effects, if any, gravity may have on the direction of cleavage, and for determining the relation of the early cleavage planes to the median plane of the embryo. Dean found "in the early stages of segmentation that the cleavage planes occurred in the normal way when the position of the egg was reversed." This is true not only for inverted eggs, but for eggs placed in any position whatsoever. It seems to follow that gravity can have no directing influence on the cleavage. In order to ascertain whether there is any constant relation of the embryonic axis to either of the first two cleavage planes, eggs were fixed in given position by weighting, as before mentioned, and a sketch of the early grooves was carefully made in each case. These grooves are easily identified for a long time in the lower hemisphere of the egg, even as late, in some cases, as the early stages of gastrulation. As the sketches made at successive intervals showed no movement of the egg during all this time, it seemed probable that the position of the egg remained practically unchanged up to the time when the median plane of the embryo was ascertainable. In some cases accidental markings on the surface of the egg remained in a fixed position until the embryo was well defined.

In 1893 Morgan[1] experimented with the eggs of teleosts. The pelagic eggs of *Ctenolabrus* and *Serranus* were selected. Morgan says: "My results show that there is no relation whatsoever between the cleavage planes of the egg and the median plane of the adult body. I have definite records of twenty-two eggs carefully marked. The results, expressed as nearly as possible, show for *Ctenolabrus* the plane of first cleavage agrees approximately with the median plane of the body in *five* cases, the plane of second cleavage with the median plane in *ten* cases. The median plane of the adult body lay between the first and second cleavage planes in *two* cases. For *Serranus* the first cleavage agreed in one case, the second agreed in two cases, and neither in two cases."

Two methods were employed. In one case the egg was watched continuously, and the median plane of the embryo corresponded to a plane midway between the first and second cleavage planes.

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[1] *An. Anz.*, viii. Jahrg., p. 803.

The second method consisted in marking the membrane above the blastoderm and using such marks as points of orientation. "The success of the second method depended upon the close adherence between the egg and its membrane. After a very careful test it was found that even after rough treatment the egg retained a fixed position relatively to the surface markings. And again by watching individual eggs for some hours it was seen that the egg did not rotate within the membrane in the early stages or change its orientation with respect to the marks on the egg-membrane. In order to mark the eggs they were removed from the water and partially dried. A needle covered with finely divided carmine was drawn horizontally over the eggs. Small particles of carmine stuck to the membrane in many cases. The eggs were returned to the water and the best marked chosen."

Jordan and Eycleshymer[1] published a joint paper in 1893, from which it becomes plain that in the case of two species of *Urodeles* and two *Anura* there exists no constant relation between the early cleavage planes and the adult axes. "The first and second cleavage planes undergo extensive torsion, and the cells originally on one side of the mid-line come to lie on the opposite side."

Jordan[2] states, in regard to the cleavage in the newt, that "the total absence of any regularity in the arrangement of the cells is the most conspicuous feature."

Experiments of a somewhat different nature by Driesch, Wilson, and others have also shown conclusively that the first line of cleavage cannot have the significance which the earlier experiments of Roux would indicate. Recent studies in cell-lineage, together with the work of experimental embryologists, have thrown a great deal of light on the subject here considered, but without furnishing a decisive answer as to the meaning of cleavage.

The opinion expressed by van Beneden,[3] as early as 1883, that all bilateral animals would be found to agree in having the

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[1] *Journ. of Morph.*, vol. ix, p. 407.

[2] *Journ. of Morph.*, vol. viii, p. 269.

[3] *Archives de Biologie*, IV, 1883.

axis of the body located and pre-marked by the first plane of cleavage, has steadily lost ground.

The study of cleavage has not yet given us any such fundamental law of development as the mosaic theory claims. The number of animals whose development does not conform to the supposed law of coincidence has increased so rapidly of late, that the only reasonable conclusion seems to be that while the first cleavage plane may coincide with the median axis of the embryo, as Roux and others have shown, it is not a constant rule in any single case, much less a universal law.

The opinion is gaining ground that the phenomena of cleavage are to be regarded as the expression of non-differential rather than qualitative divisions of the germinal material. The pressure experiments have proved that there can be extreme variation in cleavage produced without in any way interfering with the normal development of the embryo.

NOTE. — Batrachus tau = Opsanus tau. See *Bulletin of U. S. National Museum*. No. 47, Part 3, p. 2313. 1898.

EIGHTH LECTURE.

OBSERVATIONS ON VARIOUS NUCLEOLAR STRUCTURES OF THE CELL.

THOS. H. MONTGOMERY, JR.

(*Lecture given August 3, 1898.*)

WITHIN cells occur structures of various kinds which have been loosely termed "Nucleoli." In the strict sense of the term, however, the word "nucleolus" should be applied solely to those elements of the nucleus which, with suitable double stains, such as haematoxylin-eosin, or the Ehrlich-Biondi-Heidenhain mixture, stain differently from the chromatin, at least when the latter substance is in the chromosome stage. Thus the term "nucleolus," introduced by Valentin in 1839, corresponds to the "plasmosoma" of Ogata, 1883.

Besides the true nucleolus, as just defined, numerous other so-called "nucleolar" structures occur in various cells. Among them are the *karyosomes*, which are merely thickened nodal-points of the chromatin reticulum; further, what I shall term the "chromatin-nucleus," which is found in certain spermatocytes; and then various structures, which stain neither like the true nucleolus nor the chromatin, and to which such terms as "Paranucleoli," "Nebennucleoli," and "Pseudonucleoli" have been applied. It is one of the most difficult questions to determine the nature and correspondence of the latter structures, and very extensive comparative studies are necessary for their elucidation. But from the cases studied by me, it would appear that some of these structures in *Metazoa* probably must be placed within the category of true nucleoli, and be regarded as true nucleoli of a different chemical nature. That is to say, all segregations of formed substance within the nucleus, not including the chromatin, linin, caryolymph (and the problematical lanthanin granules?), probably should be regarded as true nucleoli. Or, in other words, our criterion of nucleoli probably should not be based as much upon chemical as upon morphological facts: the criterion should be rather its mode of formation and deposition than its chemical nature, for there is reason to believe, as I think the facts here presented will show, that the nucleolus is a substance formed under the action of nutritive metabolism, and hence that its chemical nature may vary in different cells, and at different stages of the same cell, just as the kind of nutrition varies in different cells and at different stages. In estimating the valence of a nucleolar structure, its morphology must be considered as well as its chemistry, and not its chemical nature alone.

The present lecture gives a short review of a few of the observations made by me upon different cells, while the detailed observations, accompanied by figures and literature reviews, are contained in two papers, one (not yet published) in the *Journal of Morphology*, and the other in Spengel's *Zoologische Jahrbücher* for 1898; the former paper, dealing mainly with the true nucleolus, was completed in February, 1897; and the second, on the spermatogenesis of *Euchistus* (*Pentatoma*), in April, 1898. The objects studied in the former paper were the germinal vesicles of nudibranch Mollusca (*Montagua*, *Doto*), of Nemerteans (*Tetrastemma*, *Amphiporus*, *Zygonemertes*, *Lineus*), of the polychaete annelid *Polydora*, of the leech *Piscicola*, and of the siphonophore *Rodalia;* further, nuclei of two Gregarines, and of various ganglion, gland, muscle, and blood cells of the forms mentioned above, and the hypodermis of the larva of *Carpocapsa*.

I. The True Nucleolus.

With Delafield's or Ehrlich's haematoxylin, followed by eosin, the nucleolus stains red; while with the Ehrlich-Biondi-Heidenhain mixture as usually employed it is stained red or orange.

1. *Number and Size.* — The usual number is about 1–5 to a nucleus, but in many cells a much larger number is present.

Thus, in germinal vesicles of *Amphibia*, *Selachii*, *Sagitta*, *Metanemertini*, most *Teleostei*, and many *Arthropoda*, frequently hundreds are present, and an equally large number is found in clitellar gland cells of *Piscicola* and in various glands of *Arthropoda*. The number in a germinal vesicle is not dependent upon the amount of yolk present in the cytoplasm, since it is the rule in birds and many arthropods that only one or a few occur. There are very few cells in which no nucleoli are present, but that may be the case occasionally in spermatozoa, and in certain connective-tissue elements; so it would appear that the absence of nucleoli is to be noted mainly in such cells where the nutritive metabolism has ceased or is reduced to a minimum. The number is not constant for all the cells of a given organism, as was claimed by Auerbach. The volume of a nucleolus stands, as a general rule, in inverse ratio to the number; *i.e.*, at a given stage of a given species of cell the amount of nucleolar substance remains more or less constant. The greatest relative amount of this substance is found in cells which are undergoing rapid growth, and in which, consequently, the nutritive metabolism is intense; it is particularly in germinal vesicles, during the growth period, that a large amount of nucleolar substance is found.

2. *General Structure and Position.* — The ground substance is usually homogeneous, but not infrequently granular (*e.g.*, Gregarines, ova of insects). In most cases it has no limiting membrane, the supposed membranes described by various writers being probably in most objects only optical illusions due to light refraction by the curved surface of the nucleolus, and in other cases to the presence of a chromatin reticulum around the nucleolus. Since the nucleolus lies in the meshes of the chromatin reticulum, and is rarely, if ever, penetrated by chromatin strands, it follows that when a nucleolus is increasing in size it pushes aside the chromatin so that the latter may form a latticework around it. But in a few cases a true membrane, a differentiated portion of the ground substance, is present, as in the germinal vesicle of *Polydora*.

The position of the nucleolus is usually excentric, rarely central. But the position often varies at different stages of the nuclear history, as is shown very clearly in the germinal vesicles of Nemerteans, where the nucleoli are formed at the periphery, wander towards the centre, and finally pass to the periphery again. Similar "migrations" have been described also for *Amphibia*.

Within nucleoli vacuoles are usually present, which are probably, in most cases at least, fluid globules; they are normal structures, since they may frequently be seen in the living cell. The origin of these vacuoles or globules was traced by me in the germinal vesicles of nudibranch mollusks. In these objects the nucleolus is at first homogeneous, but with its increase in size, which keeps pace with that of the nucleus, lighter staining globules make their appearance within the nucleolus. Just at this time, fluid drops of similar chemical nature are seen within the caryolymph, and these drops, on the other hand, resemble chemically and structurally the early yolk globules, which are simultaneously appearing in the cytoplasm. Consequently, we may conclude that globules which are probably closely related to the earliest yolk globules are taken up into the nucleus, probably as nourishment, and that some of them are deposited within the nucleolus in the form of intranucleolar globules or vacuoles. Thus, at least in these objects, there are facts to show that the nucleolar vacuoles are extranucleolar in origin. In these ovarial ova the vacuoles gradually fuse together until a single large, excentric vacuole is formed, bounded by a thin layer of nucleolar ground substance. Similar fusions of vacuoles to produce one large one are found also in nucleoli of many other germinal vesicles, as, *e.g.*, in *Polydora* and *Piscicola*. In the objects studied by me the nucleolar vacuoles would appear to be in all cases fluid globules and not empty spaces, which is proved by their becoming stained with suitable reagents. In some cases these vacuoles are not spherical (the usual case), but irregular in outline; and the latter form may be easily explained by the assumption that their consistency is occasionally viscid. The same assumption serves to explain likewise the irregular forms of many nucleoli. That is to say, the form of the nucleolus and of its contained vacuoles will be spherical, only provided that they have a thinly fluid consistency.

In some cases, as in the nucleolus of the egg of *Polydora*, the vacuolar substance is distributed in the nucleolar ground substance in the form of irregular canals. I believe that the study of similar nucleoli misled Carnoy in his idea of a "nucléole-noyau," *i.e.*, of the assumption of a particular form of nucleolus which contains all the chromatin of the nucleus in the form of a looped skein, and so represents a nucleus within the nucleus. That is to say, in such a nucleolus as the one just mentioned, he mistook the network of true nucleolar ground substance for chromatin, and the true vacuolar substance for the nucleolar ground substance. Certainly in no true nucleolus of any Metazoan have I seen any trace of chromatin, much less a loop of chromatin; but it is possible, though I have been unable to decide the point positively, that in the Gregarines the chromatin may be contained in the nucleoli.

A few observers have remarked contractile vacuoles in the nucleolus.

Very frequently there may be observed, within a nucleolar vacuole, one or several small granules, the *nucleolini;* but as far as my observations extend, such nucleolini appear to be inconstant in number and occurrence, and to be merely detached portions of the ground substance of the nucleolus. Accordingly there is no good evidence for believing that the nucleolinus represents the dynamic centre of the cell.

3. *Divisions, Fusions, and Amoeboid Movements in the Resting Cell.* — Divisions and fusions are normal phenomena of many nucleoli. In fact, the general mode of increase in volume of a nucleolus, apart from accessions to it of vacuolar substance, appears to be by the addition to the surface of secondary nucleolar masses. This may be observed very clearly in the germinal vesicle of *Lineus*. In the case of the other Nemerteans examined by me, the nucleoli formed at the periphery of the nucleus remain there for a while and increase in size, then pass towards the centre, where they may either fuse first to form a single large one (*Stichostemma*), or else immediately subdivide and so produce numerous smaller ones, all of which finally pass again to the nuclear membrane. In fusions of nucleoli which have been formed at successive stages, and

which accordingly may differ chemically, nucleoli are produced which are not chemically homogeneous throughout; this seems to be the case in the ova of numerous arthropods (*e.g.*, *Gryllus*, *Euchistus*, *Phalangopsis*, and other insects).

Amoeboid movements have been observed in life by numerous authors. But from the probable nature of the nucleolus, which will be discussed below, the conclusion seems probable that all movements, fusions, and divisions are passive in regard to the nucleolus; that they are not spontaneous movements of the nucleolus, but due rather to chemical dissolutions and changes. This is in agreement with Rhumbler's conclusions.

Besides the movements due to change of form, in which process divisions and fusions are frequently concerned, there remain to be considered those of change of location. The migration of nucleoli in the germinal vesicles of Nemerteans has been already considered; and such migrations are possibly of general occurrence in all cases where the number of nucleoli is large. Besides such phenomena, migrations of nucleoli into the cytoplasm in the resting cell have been frequently described. Among others may be mentioned the conclusions of Roule in Ascidian ova, to the effect that such nucleoli eventually become nuclei of new cells, and similar observations on endogenetic cell formation in molluscan ganglion cells, as described in a recent paper by Rohde. But it is hardly necessary to remark that all recent cytological studies render it very probable that no such endogenetic cell formation occurs in the *Metazoa*. Among the numerous objects studied by me, only one was found showing in the resting cell a migration of nucleoli into the cytoplasm, but that one, now to be described, proved to be of a unique character.

The clitellar gland cells of *Piscicola* are cells of enormous size, to be readily seen with the naked eye, which occur in the body cavity throughout the whole length of the worm, except in the cephalic region, and the ducts of which open in the region of the genital pores. During the process of formation of their secretion two main stages may be distinguished: a *prophase*, from the stage of the young cell until all the secretion has been formed and the cell has attained its maximum dimensions;

and a *metaphase*, from the time of the discharge of the secretion until the latter has been completely discharged. This includes the cycle of activity of the cell, and it is probable that this cycle is passed through once during each season of sexual maturity of the animal. My deductions as to the sequence of the stages were based upon an examination of a number of individuals of various sizes.

At the commencement of the *prophase* the spherical nucleus contains a single, more or less rounded nucleolus. The cell body increases gradually in size, and secretion globules, which stain much in the same way as the nucleoli, make their appearance at some distance from the nucleus. The cell steadily increases in size as do the nucleus and nucleolus, and then the latter elongates and passes to the nuclear membrane. There it continues to grow in volume, at the same time becoming so irregular in form that in no two nuclei does it present the same outlines; *U*-shaped, *S*-shaped, and all kinds of irregularly lobular forms are gradually produced. Then the nucleolus commences to break up into pieces, and these fragment further, until finally several hundred minute nucleoli are scattered through the nucleus, in the place of the original large one. During this time the cell body has become greatly dilated, owing to the formation of secretion globules which are now crowded throughout the cytoplasm. Simultaneously the nucleus has increased in volume, and to a greater relative extent than the cell body; and it has gradually become excessively amoeboid in form, sending out long, branching, and anastomosing processes, on which a nuclear membrane does not appear to be present. These nuclear processes extend nearly to the proximal end of the cell duct, and so penetrate between and come into direct contact with the secretion globules.

The *metaphase* of the gland cell commences with the discharge of the secretion corpuscles; and here it may be noted that the secretion appears to have been formed in the cytoplasm of the duct as well as of the cell body. During the process of discharge the duct becomes swollen. After a considerable portion of the secretion has been extruded, approximately a third of that previously present in the cell body, the nucleus suddenly commences to contract, and by the withdrawal of its pseudopodia gradually to assume the spherical form again. Now the point of chief interest to us is, that during the process of nuclear contraction the nucleoli are discharged into the cytoplasm. And the appearance of concentric rows of discharged nucleoli within the cytoplasm leads to the conclusion that the nuclear contraction is not gradual, but sudden and intermittent. The case is as if one holds in his hand a sponge filled with water, and by successive pressures of the hand forces out the water in jets; so by the contraction of the nucleus, which is automatic, as shown by the retraction of its pseudopodia, the nucleoli are forced out through the nuclear membrane into the cytoplasm, where the pressure is less. And the nucleolar substance in the cytoplasm is found in the form of rods, radial to the surface of the nucleus, which proves that it is forced out rapidly in streams. But a remarkable point remains to be noted: one nucleolus, and one only, appears in every case to be retained within the nucleus; for this phenomenon I can give no explanation, since this one does not appear to differ chemically from the others. Finally the nucleus becomes as small as it was at the commencement of the prophase and gradually rounds off; no longer are secretion globules present in the cytoplasm; and the extranuclear substance gradually disappears,— *how*, it is impossible to determine, though since it is not discharged through the cell duct, and there is no evidence of any migration through the cell membrane, it would seem probable that it becomes dissolved within the cytoplasm. Such is the history of a cycle of functional activity.

In these remarkable stages certain salient points are to be noted: the increase, first in size and then in number, of the nucleoli, accompanying the formation of the secretion; and the discharge of nucleolar substance following upon the extrusion of the secretion. These phenomena would point to some connection between formation of nucleolar and secretory substances, which is rendered the more probable, since by its extreme amoeboid form the nucleus appears to be in close physiological connection with the cytoplasmic processes. The fact that the secretion globules appear first in the cytoplasm at some distance from the nucleus would speak against any direct connection of nucleolar and secretion substances. The nucleus evinces great metabolic activity, and so would appear to play an important part in the formation of the secretion; but for the production of such activity an intense process of nutrition is necessary, and it would seem to me probable that the nucleoli are in some sense by-products of this nutritive metabolism. There are few other observations on nucleolar changes accompanying secretion stages; but reference may be made to those of Nussbaum on gland cells of *Argulus*, to the effect that increase in the number of nucleoli is produced by starvation.

4. *Ontogenetic Origin.* — How and where does a nucleolus make its first appearance within the nucleus? Obviously it would be incorrect to attempt to decide this point by a study of the reformation of nucleoli within a daughter-nucleus after mitosis, since the objection might well be raised that in such a case nucleolar material discharged into the cytoplasm during mitosis might be taken up again after mitosis, and accordingly that the first origin of this substance is not explained.

In studying this point I was so fortunate as to find a fitting object in the germinal vesicles of Nemerteans. In all the Metanemerteans examined, but not in *Lineus*, the germinal vesicle appears to arise by the increase in size of a nucleus of a connective tissue — a tissue of branching cells of an embryonal character, which has been termed by me "mesenchym." These nuclei of the undifferentiated tissue show no trace of nucleoli. By increase in size of a nucleus and its cell body, and by the rounding off of the latter, an ovarial ovum (first ovocyte) is formed. Now, when the latter has attained a certain size, nucleoli make their first appearance, and at first are always closely applied to the inner surface of the nuclear membrane. Simultaneously the yolk globules begin to segregate within the cytoplasm; yolk and nucleoli show nearly the same chemical reaction. But one essential point remains to be emphasized: the yolk substance, when it can first be differentiated by suitable stains, is found to be at some distance from the nucleus. These facts would seem to lead to the conclusion that in these cases the nucleolar substance is extranuclear in origin, and that it may stand in a genetic connection with that early stage of the yolk substance, when the latter cannot as yet be differentiated by staining methods. In other words, it would seem that the yolk is at first present in the cytoplasm in the form of a diffused, unstainable fluid; that a portion of it, that remaining in the cell body, later becomes segregated as, or chemically changed into, yolk globules; and that another portion of it is taken into the nucleus and, after passing the nuclear membrane, changed into nucleolar substance.

The objection might be raised by those who assume the nuclear origin of yolk, that the nucleoli are yolk globules formed within the nucleus and destined to be expelled into the cell body. But two answers may be given to such a criticism. First, we have seen that the first discernible yolk globules within the cytoplasm lie near the periphery of the cell, consequently at some distance from the nucleus; and, second, the broader reason, that the ovarial ovum is essentially characterized by its rapid growth, and that this growth is maintained necessarily by accretion of nutriment from without; the cytoplasm derives its nourishment from the body cavity or blood vessels, and the nucleus from the cytoplasm. This being the case, and the fact recalled that increase in nucleolar substance appears to always accompany rapid growth of the nucleus, leads to the conclusion that nucleolar substance stands in a pretty close relation to nutritive metabolism; while the fact that nucleoli first appear at the periphery of the nucleus speaks for their extranuclear origin.

There can be no doubt that the physiological processes of nucleus and cytoplasm stand in a most intimate relation, and possibly even that the nucleus directs and controls the cytoplasmic processes. Accordingly, there are good reasons for concluding that the formation of yolk globules may be directed by the nucleus. But the point for which I am contending is that the yolk substance, which is undoubtedly nutritive and hence in the first place extracellular in origin, is not produced but only chemically changed and segregated by the action of the nucleus. And since, in the germinal vesicles of Nemerteans and nudibranch mollusks at least, the nucleolar substance shows a close chemical resemblance to yolk globules, we have every reason for supposing that in these objects, if not generally, the nucleolar substance is nutritive and extranuclear in origin.

5. *Behavior in Nuclear Divisions.* — In cases of amitosis it is the general rule, as I conclude after a thorough review of the literature on the subject, that division of the nucleolus accompanies or precedes that of the nucleus. I have seen preparations showing this very clearly in cells of the ovarial follicle of *Gryllus;* Dr. Conklin kindly demonstrated these preparations to me, and allowed me to mention here this result of his as yet unpublished observations.

In mitosis, on the other hand, it is the general rule that the nucleolus disappears by dissolution during the prophases, dissolving either in the caryolymph or in the cytoplasm after the disappearance of the nuclear membrane. Such a dissolution may be either gradual, or the nucleolus may first fragment into pieces, and then these dissolve. In the first maturation division of the ovum of *Piscicola* the dissolved nucleolar substance is easily demonstrable by the aid of suitable stains in the caryolymph, and after the disappearance of the nuclear membrane seems to commingle with the cytoplasm.

In some few cases, relatively speaking, the nucleolus may not at all or only partially become dissolved in mitosis, and so be cast out into the cytoplasm, where it may be discerned through the metaphase. This has been observed, for instance, in the ova of *Aequorea* (Häcker), *Chaetopterus* (Mead), *Allolobophora* (Foot, whose preparations I have had the pleasure of seeing), and *Myzostoma* (Wheeler), though in plant cells this phenomenon would appear to be more usual than in animal cells.

In still fewer cases the nucleolus divides in the equatorial plate along with the chromosomes. Such cases, which could not, however, be proved with certainty, were observed by me in the ovogonia of *Lineus* and *Polydora,* and more or less similar cases have been noted by a few other writers. But such a phenomenon is very unusual, and it is very doubtful in such cases whether the nucleolus always divides regularly. It might be regarded as a case of a nucleolus extruded whole into the cytoplasm, which, becoming caught by mantle fibres of the spindle, would be passively divided. Or, to state the case differently, it would appear that in mitosis equal division of the nucleolar substance does not appear to be effected.

Judging from my observations on the reduction divisions in *Anasa* and *Euchistus* (Hemipterous Insects), and in *Piscicola*, as well as on the ovogonic divisions of *Lineus*, there appears to be no good evidence that the nucleolar substance takes part in the production of the spindle fibres. Yet such a view is held by Strasburger and others, based upon the generally simultaneous disappearance of nucleolus and appearance of spindle fibres. Thus in *Euchistus*, the object in which this question has been most thoroughly studied by me, in the spermatogonia the central spindle connecting the centrosomes is formed at some distance from the nucleus, and the pole rays, also of cytoplasmic origin, may be seen before the nuclear membrane has disappeared; while the mantle fibres are undoubtedly of nuclear origin, as are the interchromosomal threads, both being produced by a gradual change of position of the linin threads before the disappearance of the nuclear membrane. However, it may be going too far to conclude that in no objects are the spindle fibres formed of nucleolar substance. But in view of the evidence showing that nucleoli are masses of passive material, and that the spindle fibres are (temporary?) states of preexisting living matter, one would be justified in concluding that in all probability there is no genetic connection between spindle fibres and nucleoli.

Another point may be briefly referred to. The central spindle corpuscles ("Zwischenkörperchen" of Flemming, by whom they were first described) may by certain methods be caused to stain like nucleoli. The general consensus of opinion is that these corpuscles are thickenings of central spindle fibres or of connecting fibres. But might not in some cases undissolved portions of nucleolar substance come to lie in the equator of the spindle, and there be confused with true "Zwischenkörper"? This question is of interest with regard to the older view of Strasburger, that the nucleolar substance forms the cell plate in plant cells, and consequently the cell membrane.

Metzner (1894) considers that in the mitoses of the salamander testicle the nucleoli play an important mechanical part, since he considers some of them serve to attach the spindle fibres to the chromosomes.

In regard to the reappearance of nucleoli in the daughter-nuclei, after mitosis, very little has been ascertained. Some authors consider that the nucleolar substance dissolved within the nucleus may be carried over into the daughter-nuclei by the chromosomes. Others, as, *e.g.*, Zimmermann (with the conclusion "omnis nucleolus e nucleolo"), consider that in the anaphase all the nucleolar substance in the cytoplasm is taken up again by the daughter-nuclei. But the majority of observers leave the question undecided. My own standpoint inclines to the assumption that the nucleolar substance discharged at the stage of the equatorial plate into the cytoplasm is not again taken into the nuclei. This view is difficult to prove. But from a number of comparative observations it seems to me evident that the nucleolar substance is of no special morphological value, and that it is in all probability not carried over in consecutive cell generations. If this assumption be proved correct, it leads to the further conclusion that one reason for the disappearance of the nuclear membrane in mitosis may be the necessity for a discharge of those nuclear waste products which are in such a physical state that they cannot pass through the nuclear membrane. In the final stages of their existence the nucleoli may well be considered waste products, and the disappearance of the nuclear membrane offers an opportunity for their discharge from the nucleus. Thus in mitosis, while nutritive processes are arrested, free play is given to nuclear excretion, and so the amassing of waste products in the nucleus is prevented.

6. *Relation to Centrosomes.*—The following writers have assumed a genetic connection between nucleoli and centrosomes: Karsten (*Psilotum*, the observations combatted by Humphrey); Wasielevsky, Sala, Carnoy and Lebrun (*Ascaris*); Lavdowsky (various cells of animals and plants); Wilcox (spermatocytes of *Caloptenus*); Balbiani (*Spirochona*); Bremer (blood corpuscles of Reptiles); Toyama (spermato-

cytes of *Bombyx*); and Julin. Keuten finds in *Ceratium* a "Nucleolo-Centrosoma," which is supposed to unite the characteristics of nucleolus and centrosome, but the nature of this body is still problematical, and Lauterborn found a nucleolus but no centrosome.

The great mass of evidence goes to show that there is no relation between nucleoli and centrosomes, and the conclusion that such a connection exists is probably incorrect. With certain stains it is possible to color the nucleolus like the centrosome, but there are other methods which in most cases serve to differentiate the two. The centrosomes are frequently apparent at an early stage of the prophase, while the nucleolus is still present within the nucleus (as I have observed in *Piscicola* and *Euchistus*), so that in such cases no genetic relation of the two is possible; and it would be contrary to all well-founded ideas to conclude that an inert mass like the nucleolus is capable of becoming a dynamic centre of the cell.

7. *Nature and Function of the Nucleolus.* — From a careful and unbiased study of the subject, one is led to conclude that the true nucleolus is an inert mass and not an organ. Its substance appears to be extranuclear in origin, as already discussed, and at the time of its first appearance would not seem to be a derivative of the chromatin, as maintained by Häcker. Since nucleolar substance is found in the greatest abundance in cells which are undergoing a rapid process of growth, as is well shown in the growth period of ovarian ova, its amount would seem to stand in a pretty direct relation to the intensity of nutritive metabolism; and this is the reason why its substance may well be considered a product of nutritive metabolism. Exactly what kind of a product, we are not yet in position to decide; it may represent accumulated nutritive material, or waste products of such material, or both. Perhaps it is at an earlier stage a mass of mixed nutritive matter, from which the other elements of the nucleus gradually extract the nourishment, until finally only indigestible substances are left in it. Possibly chromatin débris may become mingled with the nucleolar substance, but there is good reason for supposing that the nucleolar substance is not wholly a

secretion of the chromatin, and that little, if any, chromatin substance enters into its composition.

That the chemical nature of nucleoli varies in different cells is very probable; and in such objects as germinal vesicles of insects the component granules of a nucleolus appear quite heterogeneous in nature. These cases we may explain on Rhumbler's theory, that compound nucleoli are formed by consecutive fusions of smaller nucleoli, the latter varying physically and chemically, since they are produced at different times.

Such appears to me to be the genesis of nucleolar substance. But is there any probability that a nucleolus at some stage in its history may become an organ, *i.e.*, actively fulfil a particular function? It has been considered by Häcker as an excretory organ, contractile vacuoles in it having been observed by him and Balbiani; while van Beneden has described rhythmic expansion and contraction of a nucleolus. It seems to me probable, but by no means definitely settled, that in most cases the nucleolus cannot be considered more than an inert mass; and that if it should in any case prove to become differentiated, or vitalized, into an organ, it could be regarded as little more than a receptacle for waste products of the nucleus, possibly exerting a force to attract such substances to itself. But the mass of evidence would show the nucleolus to be a mass of non-living substance. The usually globular form can well be explained upon its usual fluid consistency; and the regularity in number, which is a striking phenomenon in certain nuclei, as, *e.g.*, the pronuclei of fecundation, may well be explained by the assumption that there are particular spaces within the nucleus specially fitted for the deposition of nucleolar substance.

The hypothesis of Julin and Henneguy, that the nucleolus and Balbianian corpuscle of metazoan ova together correspond to the macronucleus of *Infusoria* is interesting, but an interesting speculation only. It is of importance to note that in this group of *Protozoa* only the vegetative macronuclei contain true nucleoli, while they are absent in the animal micronuclei.

Here may be mentioned briefly the conclusions of certain authors in regard to the nature of the nucleolus, in order to demonstrate how little unanimity exists in regard to it. Many (*e.g.*, Leydig, Klein, Retzius) regard it as merely a portion of the chromatin reticulum, confusing it with the karyosome. Macfarlane considers it the dynamic centre of the cell. Carnoy distinguishes "nucléoles nucléiniens" (*i.e.*, karyosomes), "n. plasmatiques" (*i.e.*, true nucleoli), "n. mixtes" (formed by the union of the preceding two), and "n.-noyaux," which he regards as nuclei within nuclei. Strasburger and others regard it as a reserve supply of chromatin; Häcker, as a secretion of the chromatin, with possible excretory function. Flemming considers it a nuclear organ, and as a "chemische Modification, Vorstufe oder Doppelverbindung," of chromatin. Korschelt's view is approximately the same as that reached by myself; namely, that nucleoli are formed as depositions of nutritive substance. Rhumbler considers them to represent reserve material, standing in some connection with chromatin; Watasé, that they are metabolic products of the cell. Born concludes that "die Nucleolen stehen in Beziehung zum individuellen Zell-Leben, nicht zur Fortpflanzung." Further, there are numerous modifications of these several views.

As a matter of fact, the nucleolar phenomena are so multiform and complex, on a broad comparative view, that it is extremely difficult to decide the formation and nature of nucleoli. The nucleolus is a structure upon which careful metabolic studies can be well pursued, but such investigation must be by the comparative anatomical method as well as by the minute research of a particular object. Neither method should be disregarded in morphological work, but the comparative would seem to offer safer results.

II. OTHER NUCLEOLAR STRUCTURES.

Besides the true nucleoli, which have just been discussed, and the karyosomes, which in the strict sense of the term are merely thickenings of the chromatin reticulum, there are found in some cells structures which appear to resemble the chromatin chemically, but which are not integral portions of the nuclear network. Such, for instance, are the "double nucleoli" of many spermatocytes and of some germinal vesicles (particularly of polychaete annelids), which are less frequent in somatic cells. These double nucleoli are usually formed of two apposed spheres, of which the one is a true nucleolus, while the other consists of a substance staining like chromatin. Not only may these bodies occur in pairs, but in chains of three or more (as first noted by Hermann in follicle cells of the testicle of the mouse); in other cases the true nucleolus may be separated from the other sphere, or the two may be at one time separated and at another united. Many of the older observers, who did not make use of sections, have described as double nucleoli, true nucleoli which enclose a large excentric vacuole, considering this vacuole to be a second nucleolus applied to the first; such cases should not be regarded as double nucleoli. These remarkable "nucleolar" structures, which stain like chromatin, have been observed by numerous writers, but as yet no satisfactory description has been given of their mode of origin. They have been observed by me in spermatocytes of various insects, in hypodermal and other cells of *Carpocapsa*, and in follicle cells of the testicles of *Plethodon* and *Mus*. Whether structures comparable to them occur in germinal vesicles, I have not as yet been able to determine. In two cases I have followed their mode of formation, and these cases may be described here.

One point may be emphasized: the term "chromatin nucleolus" should be applied only to those structures which can be proved to be derived from chromatin; in those cases where their mode of formation has not been followed it seems best to avoid the use of a special term, since it is by no means certain that all these structures should be classed in the same category.

1. *The Spermatocytes of Euchistus* (*Pentatoma*). — The spermatogonia contain only a true nucleolus, which disappears in the prophase of mitosis. The fourteen chromosomes of the last spermatogonic division are halved in metakinesis, so that each daughter-cell (first spermatocyte) receives fourteen daughter-chromosomes. The stain necessary for demonstrating the consequent phenomena is one which will stain the dividing chromatin a different color from the resting chromatin: the

only method known to me which does so satisfactorily is the excellent safranine-gentian violet-iodine stain of Hermann (published in 1889 in the *Arch. f. mikr. Anatomie*), by which the resting chromatin is colored violet, and the dividing chromatin a bright red. By the term "dividing chromatin" is understood the chromatin from about the commencement of the prophasic loose spirem stage until about the commencement of the anaphase. This method of Hermann cannot be too highly recommended for cytological work, and in many respects is far superior to the favorite method of the day, the iron haematoxylin stain, with which it is difficult to secure satisfactory differentiation of the various cell constituents.

To return to our first spermatocyte, at the stage of the last spermatogonic metakinesis. The fourteen short chromosomes are stained red until the nuclear membrane appears; they seem to be all alike in form and color. At the commencement of the anaphase the chromosomes become irregular in outline and elongated, and now lose the red and commence to take on the violet stain, passing first through a purple stage. But now a most remarkable phenomenon is to be noted: one of the fourteen chromosomes does not become violet but remains red, the color characteristic for the dividing chromatin. As the anaphase proceeds, thirteen chromosomes gradually become intensely violet, while the fourteenth remains red. The latter is the "chromatin nucleolus," as it has been termed by me; genetically it is a chromosome, but destined to have an entirely different metamorphosis from the others. The chromosomes and the chromatin nucleolus continue to elongate until their length nearly equals that of the diameter of the nucleus. Then the chromatin nucleolus ceases to elongate, passes to the periphery of the nucleus, and gradually shortens up into a more or less spherical form; during this process it may divide into two or three parts, in which case one is usually much larger than the others. At this time the true nucleolus, which does not stain at all, or only a faint lilac, is forming at the periphery of the nucleus; it may at every stage be sharply distinguished from the chromatin nucleolus. The thirteen true chromosomes elongate to a length of about double the diameter of the nucleus, and then form an inextricable coil at the centre of the nucleus, this being the stage of synapsis.

In *Euchistus* there is a well-marked stage following the synapsis, which may be termed the post-synapsis ; it is characterized by the loosening up of the long chromosomal loops, so that the latter may now be counted, which was impossible during the synopsis. A careful count of the loop in a number of nuclei shows that a reduction of the number of chromosomes had taken place in the synapsis ; in the post-synapsis the chromatin loops (chromosomes) are of unequal length, and in different nuclei vary in number from three to six, while four is the usual number. It is specially to be noted here that in the post-synapsis there are never exactly seven chromosomes, which would be exactly half the normal number, but less than seven, which fact must be brought into connection with the fact that one of the earlier chromosomes had been metamorphosed into the chromatin nucleolus. Interesting as the relations are from the standpoint of chromatin reduction, they are not exactly in line with the subject in hand, and so cannot be further described here.

After the post-synapsis follows the telophase, and then the rest stage of the spermatocyte. In the latter period the chromatin nucleolus, still red in color, lies against the nuclear membrane and encloses a large globule of lighter staining substance. The true nucleolus, larger than the former, is usually unstained (only browned by the action of Hermann's fixative) and lies near the centre of the nucleus. The chromatin reticulum is deep violet, and special chromosomes are not distinguishable.

In the prophase of the first reduction division no continuous spirem is formed, but at the stage of the dense spirem the chromatin reticulum segregates into from three to six long loops. The latter are of unequal lengths, and, according to the number of segments present in the dense spirem stage, one or more of these loops divide transversely, until in the loose spirem stage always exactly seven chromosomes (exactly half the normal number) are evolved. This remarkable power of self-regulation of the number of the chromosomes has been discussed more fully in another publication. In the loose spirem stage each of these seven chromosomes shortens up into a regular dumb-bell shape.

During these prophases the chromatin nucleolus decreases in size, partly by the loss of the substance of the contained vacuole, partly by a dissolution of a portion of its own substance; but a portion of it always remains, and this portion gradually becomes dumb-bell shaped before the disappearance of the nuclear membrane. The true nucleolus disappears completely during the prophases.

In the equatorial plate of the first reduction spindle lie the seven chromosomes, and by their side the chromatin nucleolus, in every case easily recognizable by its much smaller size. The seven chromosomes and the chromatin nucleolus are then divided transversely in metakinesis, so that each daughter-cell (second spermatocyte) receives seven chromosomes and one chromatin nucleolus. But in the second reduction division, unlike the first, the chromatin nucleolus does not divide as a rule, but is carried undivided into the one or the other spermatid.

Thus it is that in the spermatocytes of *Euchistus* a whole chromosome becomes metamorphosed into a chromatin nucleolus; and from observations made by me upon another genus of the same group, *Anasa*, it would appear that the chromatin nucleolus has a similar origin here also. In the spermatocytes of some other insects (*Phalangopsis*, *Gryllus*, and *Harpalus*) I have found similarly staining structures (which in *Harpalus* are attached to the true nucleolus, thus constituting a double nucleolus), but in these objects have not yet traced their origin. May it not be that chromatin nucleoli will be found to be of general occurrence in the spermatocytes of insects, if not indeed of other groups also? If this be proved to be the case, and the chromatin nucleolus in all objects be found to be a metamorphosed chromosome, then the chromatin reduction in such spermatocytes would be of a different character from that in ovocytes and the whole problem be still further complicated.

2. *Hypodermal Cells of Carpocapsa.* — The budding extremities of the apple-worm, which is the larva of the moth *Carpocapsa*, offer a beautiful object for the study of certain nucleolar structures. The hypodermis covering the body consists of

somewhat flattened epithelial cells, in the nuclei of which only true nucleoli are present. But in the hypodermal cells of the larval extremities both nucleoli and chromatin nucleoli occur within the same nucleus; and at the place where the extremity joins the body proper the hypodermal nuclei offer a perfect succession of gradations, showing all stages in the formation of the chromatin nucleolus. The same gradations of nuclear structure are also to be found where the hypodermis is invaginated to form the stomodaeum and proctodaeum.

At the proximal end of a larval extremity the nuclei contain, besides the chromatin reticulum, one or two true nucleoli. As we proceed, distad nuclei are found, which, in addition to the true nucleoli, contain each one small granule, which stains exactly like the chromatin, and for the designation of which the name "chromatin nucleolus" is proposed. At the time when it may first be clearly distinguished it is simply a small granule, a little larger than the nodal points of the chromatin network. While it is difficult to be positive of the conclusion, it would appear to be a portion of this network, separated from the rest of the chromatin. It is at the start usually in contact with a true nucleolus, so that double nucleoli are thus produced. Now, as the succession of stages is studied, it is found that the chromatin nucleolus steadily increases in size, while the true nucleolus attached to it either remains constant in volume or grows much more slowly. A vacuolar substance appears within the chromatin nucleolus and stains like the true nucleolus; the nucleus itself is increasing in size and gradually becoming amoeboid. Putting these facts together, — though unfortunately they can be mentioned here only briefly, — it appears that to the true nucleolus new substance is continually being added from without, while the chromatin nucleolus is absorbing material from the true nucleolus. In no other way can be explained the apposition of the two, accompanied by the rapid growth of the chromatin nucleolus and the deposition of nucleolar-like substance within the latter. Finally, in the large amoeboid nuclei, which are found at the distal end of the extremity, where these cells produce cuticular spines, a large chromatin nucleolus is seen, but no longer any trace of true nucleoli.

In this interesting series of changes we observe a granule which appears to be a derivative of the chromatin reticulum, applying itself to a true nucleolus, and by gradually absorbing the substance of the latter, becoming a large chromatin nucleolus. Is it a specially modified portion of the chromatin reticulum which is predestined to undergo this change? It would seem more probable that that portion of the chromatin reticulum lying closest to the true nucleolus would be the portion selected to grow.

The cells of the extremities are the producers of the cuticular structures of the latter, and their large size, amoeboid nuclei, and chromatin nucleoli are all evidently occasioned in the course of this production; and beneath each of the cuticular spines, which occur elsewhere on the body surface, lies a mass of cells much larger than those of the surrounding hypodermis, the relatively enormous amoeboid nuclei of which contain each a chromatin nucleolus. There can be no doubt that these chromatin nucleoli have been formed in the same manner as those in the cells of the extremities, though I have not seen the various stages of their formation. And in the body cavity are enormous cells, each with an amoeboid nucleus and a chromatin nucleolus, which are, undoubtedly, cells which at one time were situated within the hypodermis and functionated as spine producers, and later lost this connection, falling into the body cavity, where they increase greatly in size and probably fulfil some new function.

In cells of other tissues of the *Carpocapsa* larva chromatin nucleoli are also to be found, as in the intestine and salivary gland cells; but their genesis could not be traced by me, as it was for the hypodermal cells of the extremities and of the stomodaeum and proctodaeum; earlier ontogenetic stages might give the solution of the question.

What is the meaning of the evolution of such chromatin nucleoli — structures which have not frequently been observed in cells, and the genesis of which has not heretofore been followed? The chromatin is a living, and, from the morphological standpoint, most important substance. So that, if my observations prove the chromatin origin of the chromatin nucleoli in *Carpocapsa* and *Euchistus*, these nucleolar structures might be considered organs of the cell, *e.g.*, centres of nutrition, and so be of a nature essentially different from that of the true nucleoli.

In conclusion, the hope may be expressed that the study of nucleolar structures will receive more attention in the future than it has in the past. Whoever has had the patience to read this brief summary of observations will grant that the nucleoli are structures of manifold complexity, the study of the genesis and fate of which may throw needed light upon questions of cellular metabolism; and there are three groups of animals which bid fair to offer the best opportunity for work: the *Protozoa*, and particularly the *Mastigophora* and *Foraminifera*, the study of which must eventually give the best basis for cytological conclusions; the *Arthropoda*, which surpass all other forms in the diversity of cellular structure; and the *Mollusca*, which in this regard are close rivals of the arthropods.

NINTH LECTURE.

PROTOPLASMIC CONTRACTILITY AND PHOSPHORESCENCE.

S. WATASÉ.

I.

It is now generally admitted that the emission of light from an animal organism is due to oxidation of a certain substance produced by the metabolism of the cell. This is easily proved in the case of a glowworm or a firefly, where the photogenic tissue can be separated from the organism artificially and subjected to various physical and chemical tests. We learn from such experiments that anything that helps the oxidation in general, such as alkaline media, moisture, agitation, heat, etc., also facilitates the phosphorescence of the material; while, on the other hand, exposure to carbon dioxide stops it. The photogenic material is formed as the result of metabolism in the cell, and in the process of its formation is identical with that known as secretion.

Before we go any further into our subject a few words are needed on the use of the term "secretion," with special reference to the formation of photogenic material in phosphorescent organisms, because, as I shall point out more fully later, it was the narrow, limited use of the term that impeded the better understanding of the phenomena of phosphorescence as a whole.

Secretion, as popularly understood, suggests the existence of a gland, secreting a material which is eventually discharged from the organism, as is familiarly seen in the secretion of saliva, mucus, or poison of various kinds in different organisms. But this is a rather limited use of the term "secretion." As we know, there are several glands in the organism whose products of secretion are never emitted to the external world, but put to some definite use within the organism. Discharge of the material from the organism that produced it is not the essential characteristic of the secretion process.

The process of secretion does not necessarily imply the existence of a gland. When one examines the cytological aspect of the phenomena of secretion, it resolves itself into this: disintegration of the protoplasm and the subsequent conversion of the material thus produced into the secretion granules, or the separation of the specific substance from the more or less crude material incorporated within, or brought in contact with the body of the cell, under the influence of the protoplasm. So far as the fundamental process of secretion is concerned, it may be carried out by an isolated single cell just as well as by the hundreds and thousands of similarly constituted gland cells forming a sac-like recess, opening on the free surface of the organ or of the organism.

In the case of ordinary secretion, such as saliva, the granules formed by the metabolic activity of the protoplasm undergo still further changes, and eventually escape from the body of the cell. In other cases these granules remain as such, and subsequently become consumed by the activity of the protoplasm that produced them, as is well seen in the history of the yolk-granules in the egg-cell. The granules produced by the process of secretion at one period become utilized by the growing egg-cell in the course of its own development.[1]

Just as the liver or the thyroid is an organ of "internal

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[1] "The egg-cell of most animals, at any rate during the period of growth, is by no means an indifferent cell of the most primitive type. At such a period its cell-body has to perform quite particular and specific functions; it has to secrete nutritive substances of a certain chemical nature and physical constitution, and to store up this food material in such a manner that it may be at the disposal of the embryo during its development. In most cases the egg-cell also forms membranes which are often characteristic of particular species of animals. The growing egg-cell is, therefore, histologically differentiated, and in this respect resembles a somatic cell. It may perhaps be compared to *a gland cell, which does not expel its secretion, but deposits it within its own substance.*" Weismann, The Continuity of the Germ-plasm as the Foundation of a Theory of Heredity, 1885.

secretion" for the whole organism, so the yolk-granules in the egg-cell are the products of "internal secretion" for the unicellular stage of the same organism.

The cytological significance of secretion, then, whether in a single cell or in a multicellular gland, is essentially the same, and the method of the secretory process is wholly independent of the ultimate disposal or use of the products. Secretion, then, consists essentially in the production of specific substances, with the help of, or at the expense of, the protoplasm; and whether that material be expelled from, or consumed in, the organism is a question of secondary importance.

Bearing in mind, then, the general sense in which the term "secretion" has been defined, let us examine luminous organs in different organisms and see how the light-giving material is disposed of. According to the way in which it is used, luminous organisms may be divided into three groups. I am here speaking of the true phosphorescent organisms which produce light by the activity of their own tissues, and not of those which assume a luminous appearance by the habitual or accidental incorporation of other luminous organisms in their own body.

(*a*) The first group of luminous organisms includes those in which photogenic substance is thrown out of the body, from a special gland, in the form of liquid, as *Pholas*, Copepods, etc.; or from the external surface of the body epithelium in the form of fine refringent granules, as in *Chætopterus*, luminous earthworms, myriapods, etc. The secreted material assumes its luminous appearance when it comes in contact with the surrounding media, the air or sea-water, as the case may be.

(*b*) The second group embraces those in which photogenic material produced by the secretory activity of the cell is never thrown out to the external world. Oxidation of the material is accomplished by the oxygen taken with the air, or with the oxygen dissolved in the body fluid. In many cases elaborate mechanisms are developed in connection with the luminous tissue to facilitate the oxidation of the photogenic material. Various devices to intensify the luminous effect may be superadded, making them wonderfully like the visual organs, with which, indeed, they have often been confounded. This type of luminous organ, in which photogenic substance never leaves the body, is illustrated by the more familiar phosphorescent organisms, such as fireflies (Fig. 1), glowworms, *Pyrophorus*, *Euphausia*, *Nyctiphanes* (Fig. 2), *Phengodes*, *Scopelus* (Fig. 3), etc.[1]

The phosphorescent organism of this group, as a rule, has good control over its luminous organ, and may often be able to

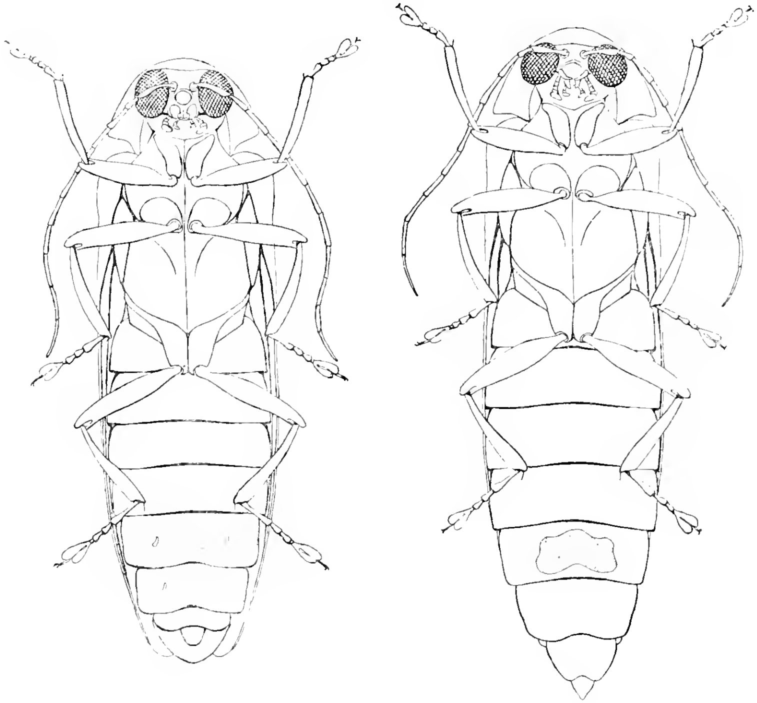

Fig. 1. — *Photinus consanguineus*, right figure, *female* ; left, *male*. Luminous organs are shaded.

inhibit the exhibition of light for a considerable length of time. The organism may be endowed with a wealth of structures suggestively like the luminous organs, yet they may never satisfy the naturalist that they are such if the exhibition of their lumi-

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[1] It is impossible to give any adequate idea of the beauty and complexity of the luminous mechanism of this type without the use of a large number of illustrations. Some ideas in regard to the size and distribution of the luminous organs that obtain in some forms may be gathered from a few figures that accompany the present paper.

nous property is left to the choice of the organism itself. The best way to deal with an organism of this nature is to expose it to the fumes of ammonia, or add a few drops of strong ammonia to the sea-water in which it lives. Even the most obstinate of phosphorescent organisms may be made to reveal their luminous property by this method. Another and perhaps a simpler method is to extract the tissue of the suspected organ, and crush it or tease it and expose the contents freely to the air, in the presence of moisture. If the tissue is really photogenic, it will shine when thus separated from the inhibitory control of the organism, and the direct access of oxygen to the tissue is thus artificially made possible. A small and slender phosphorescent earthworm which inhibited the emission of light in a most obstinate way, when gently crushed between two slides and exposed to the air or thrown into water, shone like the filament of an incandescent lamp.[1]

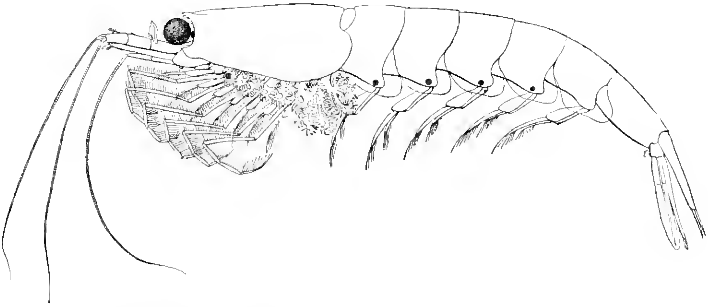

FIG. 2. — *Nyctiphanes norvegica.* Luminous organs are shown by dark dots.

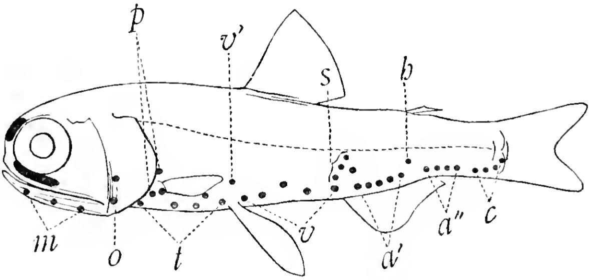

FIG. 3. — *Scopelus Rafinesquii.* (After Lütken.) Luminous organs are shown by dark dots.

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[1] The presence of water seems to exercise quite a marked influence upon the luminous oxidation of the substance, although water does not appear to take any

It is, however, possible that some tissues in a non-luminous organism, when thus treated and examined in the dark, may prove to be phosphorescent.

It is highly desirable that those who have access to various organisms whose photogenic property is suspected from anatomical grounds, but not yet definitely proved from the functional standpoint, subject such organisms or their suspected tissues to these methods of inquiry. The long-standing controversy whether the lantern-fly (*Fulgora*) is phosphorescent or not, might be settled at once by taking the tissues from the suspected region in the living insect and crushing them. The question whether the eye-like organs in the different parts of the body in bony fishes, Cephalopods, and other organisms, are phosphorescent or not, ought not to be difficult to settle for those who have access to living individuals, or even to dead ones, if the decomposition is not very far advanced; for the photogenic substance retains the power of phosphorescence long after the death of the organism.

(*c*) The third group consists of those in which the definite photogenic organ as such does not exist, but the light-giving material formed by the secretory process of the protoplasm accumulates along the course of muscle-fibres or other contractile protoplasmic material, the light manifesting itself in sparks or scintillations along the course of the fibres at the time of their contraction. This species of phosphorescence is the most interesting one, complicated as it is with the fundamental property of the protoplasm, namely, contraction. It was this kind of luminous phenomena which retarded the acceptance of the theory that phosphorescence is due primarily to the secretion of a certain material which shines in the process of oxidation. Thus, Alexander Agassiz,[1] in his interesting observations on the phosphorescence of Ctenophores, says: "Although we know now something of the nature of the phosphorescence of a few marine invertebrates from the observations of Panceri,

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part in the process itself. It is probable that water acts as a catalytic agent through which the process of oxidation is accelerated.

[1] Agassiz, Alexander, "Embryology of the Ctenophoræ," *Mem. Amer. Acad. of Arts and Sci.*, vol. x, No. 3, 1874.

who has plainly traced it to the secretion of special glands, yet when we find the same phosphorescence equally as brilliant in eggs of Ctenophoræ as in the adults, even in the stages in which the masses of segmentation can still be counted, we have evidently not yet reached the solution of the true nature of this phosphorescence. The whole embryonic mass becomes brilliantly phosphorescent when the least shock is given to the jar in which the eggs are kept."

The probable explanation of this remarkable phenomenon is that the mechanical shock given to the jar causes the protoplasm of the egg to contract, and this contraction is accompanied by the manifestation of light. The well-known case of phosphorescence of *Noctiluca*, to which I shall refer in detail later, belongs to the same category. Any stimulus that causes the contraction of the protoplasm also causes the phosphorescence of the organism.

One of the most eminent students of phosphorescence, Quatrefages,[1] while freely admitting the efficiency of the oxidation theory of phosphorescence, sharply draws a line at the last class, where phosphorescence is associated with contraction, and falls back on the vitalistic theory of luminous phenomena in vogue at the beginning and in the middle of the present century. But I think it is especially this sort of phosphorescence which is of peculiar interest to us. Intimately connected as it is with the phenomena of contraction, we may hope that the careful consideration of the subject may throw some light on the understanding of protoplasmic contraction itself.

Let us examine somewhat in detail the facts of the case, and then consider them in connection with the phenomena of muscular contraction. *Noctiluca* is a little microscopic animal, sometimes attaining the diameter of 1 mm., bearing a general resemblance to little melons, deeply indented at one end (Fig. 4). Near this depression there is an appendage by which the animal moves slowly to and fro, swaying it from right to left.

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[1] Quatrefages, A. de, "Mémoire sur la phosphorescence de quelques invertébrés marins," *Ann. des Sci. Nat.*, 1850, vol. xiv, pp. 236–287; "The Phosphorescence of the Sea," *Popular Science Review*, vol. i, 1862, pp. 275–298.

The body is so transparent as to admit of its structure being studied in minutest detail. The protoplasm of the body shows a fine ramification from the centre outward to the periphery.

The ultimate branches of the protoplasm spread themselves over the whole inner surface of the animal. The central mass of the protoplasm and its ramifications are constantly moving and changing; larger branches becoming thinner; thinner branches becoming thicker by borrowing material from their neighbors. The protoplasm is highly contractile, so much so that when the organism is stimulated energetically, the minute branches along the periphery contract so violently as to tear themselves completely from the external membrane.

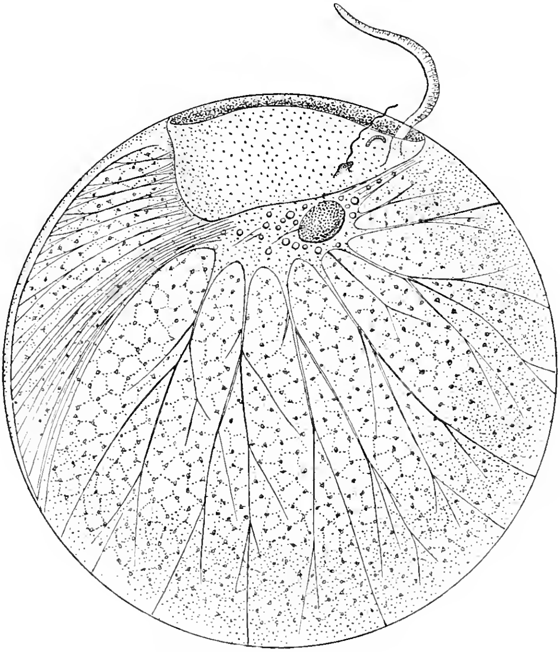

FIG. 4. — *Noctiluca miliaris.* (After Leuckart and Nitsche.)

The most interesting phenomenon in this connection is the close correlation of the phosphorescence with contraction, as I have already said. This Quatrefages proved by two sets of experiments, the one tried in the daytime, and the other at night. Thus, the action of heat, electricity, compression, and of different chemical substances — alkaline, acid, and neutral — upon *Noctiluca* have been investigated, the general result being that *any agent whatsoever capable of causing the protoplasmic substance to contract was efficacious to precisely the same degree and at the same time in producing phosphorescence.* An isolated shock caused during the daytime a violent but momentary contraction; at night it gave rise to a bright but transient exhibition of light.

Under the influence of continued or too violent irritation, the whole substance of the body is seen in daytime to contract to such an extent as to become free from the external envelope, and gradually become disorganized or lose its vital properties. At night the same operation leads to a continual phosphorescence, apparently pervading the whole animal. The steady and permanent brilliancy always denotes approaching dissolution. Thus contraction and phosphorescence are invariably met side by side, and, as it were, hand in hand. The contraction of the protoplasm may be either partial or general; it may manifest itself in different places at the same time, and so also with the luminosity. The important point to bear in mind in this connection is Quatrefages' discovery that *the luminous area of the Noctiluca is composed of an enormous number of luminous points.*

Examined with a power of twenty or thirty diameters, the illuminated portion is uniformly bright. With sixty diameters, a number of small but brilliant scintillations become visible, and these come and go with the rapidity of lightning. An enlargement of one hundred and fifty diameters reveals the true character of the phenomenon. It then becomes obvious that *the light emitted from the whole body, or any of its parts, is composed of a vast number of instantaneous scintillations, closely approximating to one another at the centre of the "phosphorescent" portions, but clearly distinguishable at the edges* (Fig. 5).

Occasionally there may be seen isolated sparks at the extreme limit of the luminous areas, or even beyond.

Quatrefages extended his observations to higher forms, where the phosphorescence is intimately associated with contractility. He succeeded in seeing the motor muscles of the luminous *Ophiura* suddenly illuminated under the microscope. The muscle alone emitted light, the other portions of the foot remaining perfectly obscure. These muscles were not perfectly luminous throughout their entire length, but there appeared disseminated over them a vast number of exceedingly minute, but at the same time remarkably brilliant points, which appeared and vanished with lightning-like rapidity. The outlines which they presented consisted not of an uninterrupted track of light, but of a line formed by a succession of scintillations.

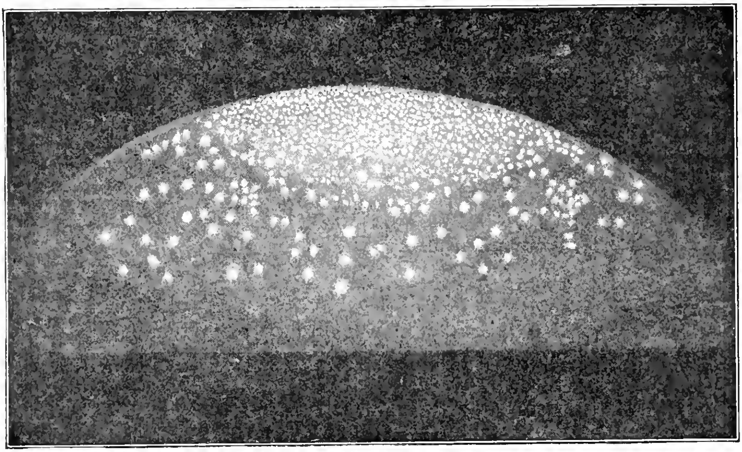

FIG. 5.—*Noctiluca miliaris*, representing a portion of the body, with a large number of scintillating points. (After Quatrefages.)

It would be easy to multiply examples, but the preceding, quoted freely from Quatrefages, will suffice to show some deep-rooted physiological connection between the production of light and the contractility of protoplasm.

I shall now pass on to the second section of my paper, concerning the nature of protoplasmic contraction; more particularly the contraction of protoplasm in one of its most differentiated aspects—the striated muscle cell.

II.

Among many theories that have been proposed by physiologists, that of Engelmann[1] is most complete. This theory Engelmann calls the "thermodynamic theory of muscular contraction," in which a muscle cell, or a bundle of muscle fibres, is compared to a thermodynamic machine, not unlike our steam engine. The theory was proposed by Julius Robert Mayer and elaborated by Engelmann, from the careful study of the anatomical, physiological, chemical aspects of the muscle cell, cilia, unicellular forms, in short, in all organisms in which truly contractile phenomena are exhibited. According to this theory, *the work the muscle accomplishes derives its energy from the heat developed in the tissue*, just as the work an engine accomplishes is developed by the combustion of the fuel. The primary form of contractile energy in the muscle is heat, which becomes transformed into the mechanical work of contraction.

The fact that the filamentous structures of the muscle are really contractile elements had been demonstrated by Engelmann by separating the fibres from the muscle cell. The isolated fibrils contract under a definite stimulus, thus refuting an idea entertained by some histologists, that the liquid portion of the muscle cell is contractile. According to Engelmann, the impulse given to the muscle cell causes the chemical change in the non-contractile substance surrounding the contractile filaments; the latter are metamorphosed to such a high degree that they do not readily undergo chemical changes. The chemical disturbance started in the neighborhood and in the space surrounding the contractile filaments takes the form of oxidation chiefly, as shown by the nature of by-products. The heat developed by the oxidation of thermogenic particles in the interstices of contractile fibrils enables the latter to absorb more water. And when an organized substance absorbs water, it becomes thicker and shorter. This thickening and shortening of the component fibres is the contraction of the entire cell.

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[1] Engelmann, Th. W. Über den Ursprung der Muskelkraft. Leipzig, 1893. "On the Nature of Muscular Contraction," Croonian Lecture before the Royal Society of London, 1895.

Engelmann illustrates this by an ingenious model (Fig. 6). A piece of an *E* string of a violin about 5 cm. long, previously soaked in water, is fastened to the rigid end of the rod *a*, of *ab*, while the upper end of the string is fixed on the shorter arm of the lever *H*. To this string different tensions may be imparted by weights d, d_1.

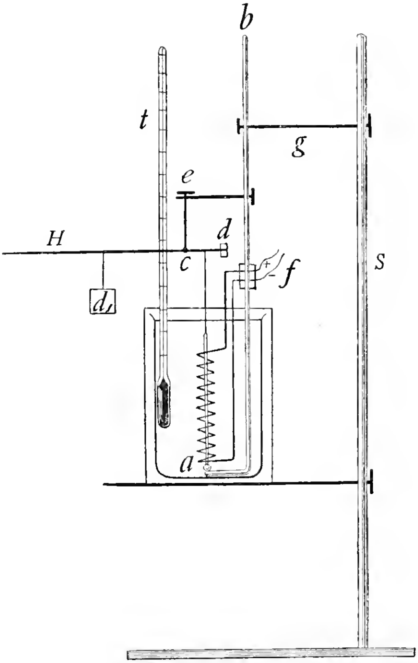

Fig. 6. — *Diagrammatic Model of the Muscle.* (After Engelmann.)

Round the string, but without touching it, runs for a length of about 20 mm. and in about 20 curves a spiral of thin platinum wire. The poles of the wire are connected with two wires coming from the poles of a Grove or Bunsen battery of three or more cells. The whole is immersed in a glass of about 50 cc. contents, and filled with water of about 55°–60° C. A thermometer is placed through the aperture of the ebonite cover.

When no change in the position of the lever occurs, *close for some seconds the circuit of the battery through the spiral; the lever rises. Upon opening the circuit it falls. The thermometer in the glass indicates scarcely any rise in temperature.*

The doubly refracting violin string corresponds to the doubly refracting particles in muscle fibrils; the water in the glass represents the watery isotropic substance round the contractile fibril, doing duty as refrigerant; the spiral wire supplies the place of the chemically active *thermogenic* molecules; the closure of the galvanic circuit corresponds with the process of the stimulation of the muscular element.

Some physiologists object that the body of an organism or a muscle "is sensibly uniform in temperature throughout, and that the more work done, the more rapid the circulation; and the more certain this uniformity of temperature, the more it seems to be impossible that such thermodynamic processes are carried on in the animal system as are familiar to us in our heat-engines."[1]

To the objection that the body of an organism, or muscle cell, does not satisfy the conditions of a thermodynamic machine because the temperature is uniform, Engelmann replies that "we must assume, on the contrary, exceedingly large differences of temperature in the stimulated muscle. What holds good of the whole body holds good of the muscle also." As Pflüger observes, the temperature measured with our instruments is but an *arithmetical average*, "*comprising an infinite number of different temperatures, pertaining to an infinite number of different points.*

"From the fact that at the contraction of the muscle an infinitesimal part only of the muscular mass is chemically active, we infer that the *temperature of their particles must, at the moment of combustion, be an uncommonly high one.* Great as the specific heat of muscular substance is, it would be impossible otherwise to account for a rise in temperature of the whole mass even of 0.001° C., only. Without any exaggeration, we must assume that the temperature of the chemically active particles may, at the moment of combination, exceed the average muscular temperature by hundreds of degrees.

"Since each thermogenic particle is surrounded by a relatively enormous cool mass, conducting heat and diathermous,

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[1] Thurston, R. H. The Animal as a Machine and a Prime Motor, and the Laws of Energetics. New York, 1894.

the principal condition for the transformation of heat into mechanical work is satisfied, and, on account of the enormous difference in temperature which we have to assume in such a high degree that even an economic coefficient of 30 per cent, nay, 50 per cent, and even more, seems to be theoretically possible."

The objection has been raised "that those high temperatures must necessarily destroy the life of the muscle, since the latter becomes rigid and dies, even at 50° C." But Engelmann considers this objection is of small value. "For it is ever an infinitesimal part only of the muscular mass that is exposed to the high temperatures. At a small distance from these furnaces of heat the temperature must have fallen so low as to be harmless. The muscle will no more be destroyed by stimulation than a steamer will be destroyed by heating the furnaces. The material of combustion only will be destroyed; the whole steamer as such remaining unharmed."

How the heat thus generated by the thermogenic particles of sarcoplasm becomes converted into mechanical energy of contraction may be stated in a few words. It acts as a stimulus (Reiz) to the contractile fibrils, which, under the influence of a higher temperature, absorb watery liquids with avidity. Production of heat, then, is the essential antecedent to the contraction of the contractile fibrils, because it is through increased temperature that contractile filaments are made to absorb more watery substance, and therefore become shorter and thicker.

There is one point which ought to be kept in mind in applying this theory to the explanation of contractility in general, and that is, we must not confound the conception of heat with that of temperature. Heat is a definite thing, something objective; temperature is a state, a mere condition of the body. Burn ten pounds of coal, and warm ten gallons of water, and we get a certain temperature of the water. Burn the same amount of coal, and warm twenty gallons of water instead, and we get a temperature of the water differing from the first. The amount of heat developed in each case is the same, but the temperatures of the water are different. We must not conclude, therefore, because a certain body is cold to our touch it is devoid of heat. The organism may live in water of freezing temperature, yet it is perfectly possible that there is enough difference of temperatures in different parts of the living cell to serve as a stimulus for the contraction of the protoplasm.

III.

Now, coming to the third and concluding part of the paper, *viz.*, the relationship of protoplasmic contractility and phosphorescence, we may note at the outset that the distinction drawn between heat and light is purely of an artificial character. They are simply variations of the same radiant energy. Their difference objectively considered is simply that of degree and not of kind. The wave-length of the one is a little longer than that of the other. If they appear to be so different to us as heat and light, the trouble lies in our organization, and not in the nature of original radiant energy. The difference is subjective and not objective.[1] The heat-producing particles and the light-producing particles, objectively considered, may not be very different from each other. *They may be variations of similar chemical substances, as the resulting energies, the products of their oxidation, are the variations of the same radiant energy.* They may be very nearly related in chemical structure as well as in anatomical origin; they may exist in one and the same cell, as the products of "secretion" or of the metabolism of the cell.

The stimuli, therefore, such as mechanical jars, shocks, agitation, nervous impulse, etc., which induce combustion of the thermogenic molecules, may also be presumed to incite combustion of the photogenic molecules as well, which exist side by side.

When there is a muscle in which these two varieties of oxidizable substance exist side by side, then we have contraction as well as phosphorescence at the same time. From one variety of granules, heat is generated, which becomes converted into the mechanical work of contraction; from another, light,

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[1] See in this connection my previous paper, "On the Physical Basis of Animal Phosphorescence," *Biological Lectures*, 1895.

which affects our eyesight. The cause which lies at the basis of muscular contraction is essentially of the same nature as that which produces the phosphorescence in several organisms.

Every time our muscles are put in a state of contraction there takes place a phenomenon which is analogous to the emission of light.[1] If we do not see the flash of light in the contraction, the explanation perhaps lies in the nature of our visual organs with a comparatively limited range of visual reaction.

It is theoretically possible that all living protoplasm is phosphorescent, and that there may exist some organisms endowed with different kinds of visual organs, to which organisms like ourselves may appear luminous in the dark, especially when our muscles are in motion. And that only when the photogenic property becomes grossly exaggerated, as in comparatively few representatives in each of different phyla, of living organisms, does our eye recognize them as such. The true physical basis of phosphorescence finds its closest analogue in the common phenomena of heat-production, and is as extensive as life itself.

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[1] Engelmann (Ursprung der Muskelkraft, 1893, p. 10), for example, stimulated the muscle in the dark, until the state of tetanus was induced, thereby hoping to see the emission of light from those heat-producing particles, which may become incandescent at the height of their combustive process. His result was negative, however; but, as he says elsewhere ("Die Purpurbacterien und ihre Beziehungen zum Licht," *Bot. Zeit.*, 46 Jahrg. 1888, Reprint, p. 17), if it were not for the fact that such thermogenic particles are so very minute and comparatively scarce, one would be able to see the scintillations emanating from the inner recesses of cell-substance, on account of the enormous heat developed in the combustion of such particles.

The position assumed in the present paper is, that when contraction is accompanied by phosphorescence, which is the emission of light without any sensible heat, there may be two kinds of oxidizable substances very nearly related to each other genetically, one of which gives rise to heat, and the other to light. The assumption that there exist particles which give rise practically to light, and to light alone, is supported by the fact that such particles form the physical or anatomical basis of phosphorescence in fireflies and glowworms.

If the facts and inferences presented in the present paper are somewhat foreign to Engelmann's theoretical conception of the relation of protoplasmic contraction and the emission of light, they nevertheless seem to me to throw a strong side-light on the correctness of the fundamental assumptions in Engelmann's theory of muscular contraction.

TENTH LECTURE.

SOME PROBLEMS OF REGENERATION.

T. H. MORGAN.

THE experiments made by Trembley, in 1740, on the regeneration of *Hydra* excited amongst his contemporaries the liveliest interest. Curiously enough, Trembley's experiments were made in order to discover whether the new creature that he had found was an animal or a plant, for at that time it was well known that pieces cut from a plant would give rise to new plants. Trembley says: "I thought once more that perhaps these organized bodies that I had observed might be plants, and I had the good fortune not to reject this idea. I say that I had the good fortune not to reject this idea because, although the less natural one, it led me to cut up some of the polyps. I judged that if the two parts of one polyp should live after being separated and become each a perfect polyp, it would be evident that this organized body was a plant. Yet, since I was much more inclined to believe that it was an animal, I did not count much on this experiment. I expected to see the polyps die."

Réaumur had some time before shown that plant-lice could, if they were isolated, multiply without sexual union, and Bonnet, in 1740, convinced himself of the same fact, and Lyonet also in the same year. These discoveries, Trembley said, sufficed to show that in regard to the property of multiplying without sexual union there is no difference between plants and animals.[1]

Trembley's results on *Hydra* led to the cutting up of count-

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[1] Je sentois vivement que la Nature étoit trop vaste et trop peu connue pour qu'on pût décider sans témerité que telle ou telle propriété ne se trouvoit pas dans telle ou telle classe de corps organisés.

less animals of all classes in the endeavor to discover whether the same power of regeneration existed in other forms. Bonnet (1745) and Spallanzani (1768) were most successful with their experiments made on fresh-water worms, earthworms, the limbs of newts, and the heads of snails (*Helix*). The results of the experiments on snails, made by Spallanzani, called forth the most vehement criticism from his contemporaries; for many of them seemed to consider the discovery that a snail could regenerate its head and all the contained organs was beyond credence. No fewer than eleven writers who had themselves repeated this experiment (in one case on 1400 snails) affirmed that when the head was *completely* cut off no regeneration took place. On the other hand, as many as ten experimenters confirmed Spallanzani's results. Bonnet championed Spallanzani's cause, and the weight of his authority did much toward the acceptation of the latter's results.

Similar discussions have arisen from time to time in regard to the power of regeneration of many other forms, and the fact is of interest as showing that the conditions of the experiment, as well as the condition of the animal, are of importance in deciding whether or not regeneration will take place.

The majority of naturalists, at the time of which I have just spoken, were content to wonder at the results; a few, however, took a philosophic interest in the phenomena, and attempted to bring them into line with the current theories of generation. The speculations of Spallanzani and of Bonnet are of great interest, and not dissimilar from certain modern attempts to explain the process of regeneration by the preformation theory.

With the development of our modern technique a new interest was awakened in the phenomenon of regeneration, particularly in connection with the cellular and germ-layer theories. Numerous attempts have been made to discover the origin of the new cells in the restored part, and to determine whether the newly formed organs arise from the same germ-layers that give rise to similar organs of the embryo. It is instructive to observe how often the facts have been interpreted so as to bring them into line with the results of embryology because of an assumed necessity of the so-called germ-layer theory that ectoderm, endoderm, and mesoderm always give rise to the same layers and organs. Recent years have witnessed the decline of the germ-layer theory, and numerous discoveries in budding and dividing animals as well as in regeneration of new parts have shown that the germ-layers do not in some cases play the part assigned to them. The overthrow of the germ-layer dogma has done much to prepare the way for a broader conception of development and cell-differentiation. If I mistake not, there is a tendency at present, that is slowly gaining ground, to give up as unprofitable the interpretation of regenerative and even embryological phenomena in terms of speculative phylogeny.

A large body of information has now been gathered in regard to the power of regeneration in the plant and animal kingdoms, and the time has come, I believe, when we can profitably cease from indiscriminate mutilations, and when, after an examination of the known facts, we can ask ourselves what are the real problems that are presented to us in the regeneration of living things. We can then hope to direct our experiments to some definite purpose — toward the solution, if solution be possible, of the main problem. It must be admitted that well-directed attempts often prove futile, while accidental discoveries are sometimes of the greatest importance. Nevertheless, the solution of one well-propounded problem may fully repay the investigator for a dozen failures. The failures are due, no doubt, not to the capriciousness of nature, but to our inability to grasp all of the conditions within and without the animal or plant, and also in no small part to our failure to appreciate what questions we can expect to be answered and what not; for lacking, as we do, all fundamental knowledge of the phenomena that we call life, it is not surprising that we find ourselves so often foiled.

My present object is to examine some of the problems of regeneration that are at present before us, and then to see if we can get any clearer idea of the main problem of regeneration. Under the following headings I have arranged as many problems.

I.

Numerous experiments on *Protozoa* and *Protophyta* have shown that a nucleated part is capable of forming a new individual. So far as we can see there is not in most cases, perhaps in none, the formation of new indifferent protoplasm in which the new parts are developed, but the entire piece is changed over into a complete animal or plant of smaller size. At first sight there seems to be here a marked difference between the regeneration of unicellular and multicellular forms, for in the latter it is usual for a knob of new tissue to appear, and out of this the new part develops. However, Trembley saw that when a *Hydra* is cut longitudinally the cut edges bend in and fuse, forming a new tube of smaller diameter. Nussbaum has also observed in *Hydra* the rolling in and fusion of the cut edges. In both cases the new form develops without the previous formation of new tissue. In a tubularian hydroid Bickford has found that when a piece is cut from the stem the new tentacles appear in the old tissue, and I shall describe more fully below the results of some experiments on planarians which show that the old part plays an important rôle in the formation of a new individual. We see then that the difference between unicellular and multicellular forms is not so great as appears at first sight. The special problem that we meet with here is this: How far are the processes of regeneration in unicellular and multicellular forms the same? Can we compare with the *Protozoa* only those multicellular forms in which regeneration takes place directly in the old parts? If so, is the relation between the cells only one of correlation, as Hertwig has recently urged for all multicellular forms; or are we still prejudiced by the cell-dogma? Do not these cases of regeneration in the multicellular form indicate that the individual is a whole in the same sense that the unicellular form is a whole?

II.

In the regeneration of the lower animals and lower plants we do not find any differences that need detain us, but a curious difference is found between the higher animals and plants; for instance, in vertebrates and flowering plants. In the former, regeneration takes place by the formation of a knob of indifferent cells and subsequent differentiation. In the higher plants, on the contrary, regeneration of this kind is extremely rare (root-tips). The new part does not appear at the cut end, but the injury acts as a stimulus to the development of buds more or less distant from the exposed surface. The buds that develop may be in some cases already formed, but in other cases new buds appear after the injury, and in places where there were no buds before the injury. Why do we find this difference in the renewal of organs between the higher animals and plants? The usual reply is that plants do not regenerate at the cut end, because they have acquired a new method of replacing lost parts. The answer is not only teleological, but a quibble, so long as it cannot be shown that there is a connection between the development of buds and regeneration at the cut ends. If, for instance, the new buds be continually destroyed as soon as they appear, will the plant then regenerate at the cut surface? Can we imagine that, were it possible to destroy or injure the buds for many generations, the plant would then "acquire" a power to regenerate at the cut end? If, on the other hand, it is assumed that the plant has lost its power of regeneration at the cut end, because it acquired a new method of renewal of lost parts, the argument may be met by the fact that certain hydroids regenerate both at the cut end of a stem, as Loeb has shown, and also along the stem itself. There is, so far as we can see, no contradiction between development from the cut surface and development by means of buds at other points. Our immediate problem is to examine the conditions of regeneration in the plant itself, to see if we can discover why regeneration takes place at some distance from the exposed surface.

III.

It is not unusual to find that development will take place in one direction and not in another. A newt that has had a leg cut off will regenerate the lost limb (Spallanzani, Bonnet, Barfurth, etc.), but the limb does not make a new newt; because, one may say, it dies before it can bring about the development. But if it were grafted on to the body of the same or of another newt by its distal end, so that its proximal end is exposed, would a new newt bud out from that end? We should certainly be surprised if this happened. The experiment has not been tried with the newt, but a not dissimilar experiment has been made with the rat by Bert. The tail was bent over and fixed beneath the skin of the back of the animal, and after it had grown to that part it was cut off at its root. The exposed end of the transplanted tail did not — so far as the experimenter has recorded — give rise to a new rat.[1]

The same problem is met with in the regeneration of the earthworm. If a few of the anterior segments (one to five) be cut off, the same number come back; if more are cut off, the process of regeneration begins only after a longer interval, and only four or five segments come back, as a rule; if the cut be posterior to the middle, the time before regeneration begins is still longer, and fewer worms succeed in regenerating at all; and still nearer to the posterior end there is no regeneration anteriorly. In the same worms we find that the posterior end will not regenerate from a small anterior piece, although the piece will regenerate anteriorly if a few of its anterior segments are also cut off. The latter result shows most strikingly that it is not due to the size of the piece that regeneration does not take place posteriorly. We see, therefore, that the posterior end of the body can regenerate in one direction only, namely, posteriorly, and conversely the anterior end can only regenerate anteriorly. There is no structural peculiarity, such as differentiation of the cells, that can account for the facts. What, then, is the explanation of this curious phenomenon? Is there not here a problem that we can hope to solve by means of further experiment?

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[1] The experiment is not, perhaps, altogether to the point, since the rat does not regenerate a new tail.

IV.

An examination of the literature of regeneration will show that there is a widespread dogma that the power of regeneration of a part is commensurate with its liability to injury. I have been able to trace this belief back to Bonnet, who says that it is possible that the number of times a worm can regenerate a new part is in proportion to the number of accidents to which it is exposed during its life. Bonnet found it convenient to propound this statement in order that the results of the experiments on regeneration might be fitted into the inelastic preformation theory.

The modern preformationists *avoid* the difficulty in other ways, as may be seen in Weismann's book on the Germ-plasm.

What is even more surprising is the fact that modern Darwinians pretend to be able to "explain" the process of regeneration as the result of natural selection. None of the writers have, so far as I know, taken the trouble to tell us how an indefinite series of injuries to a part could at last make that part "acquire" a power to regenerate in succeeding generations. Darwin does not discuss the point, and his more enthusiastic followers can only state that those animals are "fittest" that regenerate. They fail to see that even on their own assumption regeneration often takes place without relation to the survival or non-survival of the animal, and at best the only possible statement that can be made is that all forms have died that have not had the power to regenerate, and this admission would show only too clearly that they cannot pretend to offer any "explanation" of the process itself. Such a statement is, moreover, in direct contradiction to a large number of known facts, for all animals alive at the present time do not possess this power of regeneration. It would surely be as useful to man to be able to reproduce a new arm or leg as it is for a salamander to regenerate its limb or for an earthworm its tail; and I cannot but feel assured that mankind will never acquire the property of replacing his arms and legs by a minutely graduated series of injuries combined with an inbreeding of the unfortunate individuals subjected to the experiment. I need not here recall numerous facts in regard to regeneration of internal organs that can seldom or never (except in artificial operations) be exposed to injury.

Last year I undertook a series of experiments on the regeneration of the appendages of the hermit crab, in order to determine whether regeneration takes place more readily in the appendages exposed to injury than in those protected by the shell. The results show that although the first three legs are often lost by the animals, yet they do not regenerate more readily than do the other appendages protected by the shell, and some of which at least can scarcely ever be lost or injured.

The question may be asked: Do we find regeneration more common in animals and plants that are most exposed to danger? If, as several naturalists believe, this is true, what explanation may be offered? The most frequent "explanation" is that in some way this is the result of natural selection, but it may be also "explained" in another way; namely, only those animals that had from the beginning the power of complete regeneration of a lost part can exist where the danger exists. In other words, only these forms can extend their area of distribution over places dangerous to their existence. At present, however, we lack the data to show that regeneration is really more frequent in animals and plants that are most exposed.

V.

In both unicellular and multicellular forms it has been found that below a certain limit of size a part fails to regenerate, although in some cases the small piece may live to a time when larger pieces have begun or completed their regeneration. The same result has been obtained for nucleated fragments of the unsegmented egg. Do we meet here with the same problem mentioned above, where a leg does not give rise to a newt, although a newt gives rise to a leg, or are the two problems quite different? I am inclined to think that they are different, because the small pieces may come from regions of the body that are capable of regeneration, and if the small pieces are grafted on to larger ones they may then regenerate, as Joest has shown for the earthworm.

There is a curious fact in regard to the development of small pieces, namely, that they do not give rise, in some cases at least, to the complete number of organs characteristic of the form. For instance, Miss Peebles has found that a small piece of *Hydra* forms only one or two tentacles. Why does this reduction in number of organs take place?

VI.

We come now to some of the most important problems of regeneration — the external and internal conditions that determine whether or not a part will regenerate. The little that we know in regard to the effect produced by external factors is almost entirely due to Loeb's important experiments. A hydroid *Eudendrium* failed to develop new heads when kept in the dark, but when brought into the light the new heads quickly appeared. Here it seems that light, in some unknown way, acts as a stimulus necessary for the growth of the new head. In another hydroid — *Tubularia* — Loeb has shown that instead of a head a root will develop at the distal end of a piece if that end be brought into contact with some fixed object, and conversely a new head will appear at either the proximal or distal end if the end be freely surrounded by water. Here also we must look upon the external agent as a stimulus that determines the differentiation of the part. At present we can form scarcely any idea of what this relation is between the external agent and the organism. The vital question is, whether we can extend our experiment so as to determine what this relation is, or have we here a fact beyond which we cannot hope to go?

Loeb has also described some further experiments on another hydroid, *Antennularia.* A piece cut from the stem and suspended vertically in the water will develop a new stem at the upper end and roots at the lower end, regardless of whether the upper end corresponds to the distal or proximal end of the original form. Here gravity alone determines that the upper end shall grow into a new stem and the lower end into a new root. In the one case, where the upper end corresponds to the proximal end of the original hydroid, the new part (a stem) replaces the lost root end, and this change Loeb calls heteromorphosis. In the other case, where the upper end corresponds to the distal end of the original hydroid, the new part (a stem) replaces the lost part of the stem. This is what is usually meant by regeneration. It would be most instructive to find out what pieces of this hydroid would do were they fixed to a revolving wheel so arranged that the direction in which gravity acts on the piece would change at every instant.

It is of the greatest importance to note that in all these cases described by Loeb, in which regeneration is determined by external circumstances, the same influences affect the growth of the uninjured hydroids in the same way. Only those forms whose normal growth is influenced by light, or by gravity, or by contact, could be induced to respond to the same influences during the time of regeneration.

In review of the few facts that we possess in regard to the effect of external factors, we must bear in mind that in some cases the external agent, gravity for instance, does not seem in itself necessary for the growth of the animal, but it acts in such a way that it determines what sort of differentiation may take place. Other factors, temperature for example, determine only whether or not growth can take place at all, and if so, the rapidity of the growth. The salts contained in the water act in the same way, in so far as they are necessary for the metabolism of the cells. In so far as the salts affect the osmotic condition of the cells they seem to determine — as Loeb has shown for these hydroids — the amount of growth, but not its kind. It may be that other factors, light for instance, may affect not only the amount, but also the kind of growth.

In this same connection I should like to describe an ingenious experiment made by Herbst. He found that when the eyes of a prawn were cut off, sometimes an eye and sometimes an antenna regenerated at the cut surface. He then tried the experiment of placing an equal number of new individuals — after cutting off the eyes — in the light and in the dark, to see if the light would act as a stimulus on the forms in the light, and thus cause an eye to appear. It was also conceivable that in the absence of light an antenna might develop, since an eye would be of no value to an animal in the dark, while an antenna would be a useful organ. But these results did not follow, since eyes and antennæ appeared in equal numbers in the animals in the dark and in the light. In the hermit crab also I have found that sometimes eyes and sometimes antennæ appear after the eye-stalk is cut off, and I have discovered that when only the tip of the eye-stalk is cut off a new eye reappears, but when the eye-stalk is cut off near its base an antenna-like organ develops. In this case it seems that the factor that determines whether an eye or an antenna develops comes from within and not from without. We find here a process of heteromorphosis depending, so far as we can see, on internal factors. Does this kind of heteromorphosis belong to the same class of phenomena shown by the hydroids? In the latter the stimulus that determines the kind of regeneration comes from outside the animal.

VII.

It is of some theoretical interest to know whether the old cells form directly the new tissue, or whether reserve cells are present that bring about the result. From the work already done it seems probable that this may vary with different forms. In some cases, as Randolph has shown for *Lumbriculus*, reserve cells are present that assist in the regeneration of the mesoderm; in other cases, as Hescheler has shown for the earthworm, the old cells seem to give rise to the new ones. In *Tubularia*, as Bickford has shown, the old cells go directly over into the new tissue.

Not only do the old cells give rise to new ones, but in the case of *Planaria maculata* the old part may itself change its form as a whole, and in this way play an important part in the development of the new worm. There is an extensive remoulding of the old tissue, so that the new parts form a miniature copy of the adult worm. It is difficult, even impossible, to form any idea of what internal changes are taking place during this period of change, and our ideas of reorganization and of correlation of parts are entirely inadequate at present to make clear the process. More elaborate series of experiments must be undertaken before we can even attempt to make an analysis of these results.

VIII.

There are certain problems in connection with the relation between the old parts and the new that need careful study. I have already touched on some of these points in the preceding pages. The limb of a newt, for instance, is replaced by a new one precisely like the old. If only the hand is cut off, just this much is reproduced; if the forearm and the hand are both cut off, then a new forearm and hand reappear; and this is true at whatever level the cut is made. In other forms less (or even more) reappears, under certain circumstances, than was removed; but as yet our experiments have not been carried far enough to make clear how this result is related to the renewal of only as much as removed.

In the earthworm when one, two, three, four, or five anterior segments are cut off, the same number come back; but when more than five are cut off, only five are regenerated. In *Lumbriculus*,[1] also, the number of new head segments is never more than seven or eight, even when a much greater number have been cut off. In *Planaria* a new head forms on the anterior end of a posterior half of the worm, but the entire anterior half is never replaced by new tissue. In this same planarian Randolph has discovered a most important relation existing between the old and new parts. If a planarian be cut in two longitudinally in the median plane, the right half regenerates a new left half of the same size as the part removed, and the left half also develops a new right half of corresponding size. If, however, the worm be cut longitudinally into a larger and a smaller strip, the former replaces as much as contained in the smaller part that was removed, but the smaller part does not develop the lost larger part, but forms only as much new tissue at its cut side as is about equivalent to its own breadth.

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[1] An American species.

When the posterior end of an earthworm is cut off, there appears at first at the posterior end of the anterior part a small knob, out of which a few segments and a growing end develop. The latter continues to extend backward and add new segments. After a time the posterior growth comes to an end, *i.e.*, when the full complement of segments has been reached. If, however, instead of the entire anterior end we take a small piece from the middle of the worm, we find that even this piece only forms as many new posterior segments as lay originally behind that piece in the worm. In other words, the growing region ceases to give rise to new segments after those that were lost have been replaced, so that the entire number of segments in the new worm may be far below the normal number. It seems to me that we see here a factor at work that we cannot in the least grasp. To call it correlation is of no avail unless we can define what we mean by the term. The usual definition of correlation is only a restatement of the facts in different words, and not a causal explanation of the facts.

It is not difficult to show how much we must include in our definition of correlation if we use the word to cover all the known facts. According to Blumenbach (1784), the eye of *Triton* will regenerate if only a small part of the original eye be left. If the anterior end of the earthworm be cut off very obliquely, the pointed end will bend over the cut surface and fuse with it, and slowly a new anterior end will develop between the bent-over portion and the anterior cut surface. In both these cases the relation of the new part to the old must be an extraordinarily complex one, and yet a new organ develops like the one lost. Again, pieces may be cut from the egg, and it will still, in some cases, produce a whole animal of smaller size — a part developing a new whole much in the same way that a part of a protozoön may form a new animal of smaller size. Something more is included in these phenomena, I think, than can be explained by simple physical interaction or by chemical influences. The process that takes place suggests that something like an intelligent process must be at work — I mean that what we call correlation of the parts seems here to belong rather to the category of phenomena that we call intelligent, than to physical and chemical processes as known in the physical sciences. The action seems, however, to be intelligent only so far as concerns the internal relations of the part, *i.e.*, it acts rather as a "perfecting principle" than as a process of adaptation to external needs (adaptation).

IX.

Finally, I wish to touch briefly on a few points that seem to me to bring us nearer to the heart of the problem. In much of our biological work we have been guided by methods derived from the physical sciences, and most fortunately so, for perhaps only in this way can we hope to reduce living phenomena to simpler terms. But sooner or later we meet with a factor that defies further physical analysis, and this factor seems to be present in all biological phenomena. We gain nothing by calling it a vital force, unless we can define what we mean by vitality. Whether or not this factor[1] is only a complex of physical forces that we cannot unravel, or whether there exists something that cannot be expressed in terms of physics and chemistry — that is the question!

We err, I think, in going at present to either extreme, *i.e.*, either in ignoring this something that has been called a vital force and pretending that physics and chemistry will soon make everything clear, or, on the other hand, in calling the unknown a vital force and pretending to explain results as the outcome of its action.

In our studies of the development of form we meet most often with this factor. Are we at bottom trying to give a causal explanation of form itself, and, if so, is not our problem insoluble? Can we hope to do more than determine under what internal and external conditions a given form appears? If we limit our researches to this problem we can hope to succeed. But can we go back of this and explain the reaction itself? At present we have not succeeded in doing so, any more than has the mineralogist explained the form of a crystal. It may be

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[1] It is simpler to speak of it as one factor, but it may equally well be true that there are many factors.

that what we call a formative force or a vital force is the property of living things to assume a given form under certain conditions. If so, is there here legitimate ground for investigation, or rather let me ask, can we hope to extend our investigations beyond the knowledge of the internal and external conditions within which new forms arise. It is this uncertainty in regard to the problem of vitality that we need first to clear up, and it seems to me that this is the cardinal point for us to examine at present. It is possible, I think, by means of experiment alone, to determine how far and in what sense we can pursue the investigation of the causes of form. In this regard experimental studies on the regeneration of animals and plants offer a most admirable field for future work.

ELEVENTH LECTURE.

THE ELIMINATION OF THE UNFIT AS ILLUSTRATED BY THE INTRODUCED SPARROW, *PASSER DOMESTICUS.*

(*A FOURTH CONTRIBUTION TO THE STUDY OF VARIATION.*)

HERMON C. BUMPUS.

We are so in the habit of referring carelessly to the process of natural selection, and of invoking its aid whenever some pet theory seems a little feeble, that we forget we are really using a hypothesis that still remains unproved, and that specific examples of the destruction of animals of known physical disability are very infrequent. Even if the theory of natural selection were as firmly established as Newton's theory of the attraction of gravity, scientific method would still require frequent examination of its claims, and scientific honesty should welcome such examination and insist on its thoroughness.

A possible instance of the operation of natural selection, through the process of the elimination of the unfit, was brought to our notice on February 1 of the present year (1898), when, after an uncommonly severe storm of snow, rain, and sleet, a number of English sparrows were brought to the Anatomical Laboratory of Brown University. Seventy-two of these birds revived; sixty-four perished; and it is the purpose of this lecture to show that the birds which perished, perished not through accident, but because they were physically disqualified, and that the birds which survived, survived because they possessed certain physical characters. These characters enabled them to withstand the intensity of this particular phase of selective elimination, and distinguish them from their more unfortunate companions. It will be convenient for us to arrange our material in the form of tests, as follows.

Test 1: Sex. — It will be noted by reference to the tables that of the *surviving* birds the males are much more numerous than the females. Of the former there are fifty-one (thirty-five adults and sixteen young), while of the latter there are only twenty-one. Among the birds which perished, the females are absolutely and relatively more numerous than they are among the birds which survived, although more than one-half (thirty-six out of sixty-four) of the unfortunate birds are males. Of course it may be that male birds are naturally more abundant than females, but the present question is not one of distribution of sex, but rather of distribution of fitness, and the inference is that the females are less competent to resist severe winter weather than are the males, for, while only 28% of the survivors are females, they constitute 43% of those that perished.

Test 2: Length. — The first column of figures on the several tables gives, in millimeters, the lengths of the birds from the tip of the beak to the tip of the tail. An examination of the averages, printed at the bottom of each column, will prove particularly instructive. It will be noted on Tables I and I^a that the average length of the adult males which survived (159 mm.) is really less than that of the adult males which perished (**162** mm.).[1] Similar figures, 159 mm. and **162** mm. on Tables II and II^a, indicate the same relative lengths of the young males of the two groups. The average lengths of the females of the two groups, 157 mm. and **158** mm., Tables III and III^a, also indicate an excess in the average length of the birds which perished. The birds which perished, then, males or females, adult or young, are longer than those which endured, and we are justified in concluding that when nature selects, through the agency of winter storms of this particular kind of severity, those sparrows which are relatively short stand a better chance of surviving.

Test 3: Alar Extent. — Averages based on measurements from tip to tip of the extended wings fail to bring out any

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[1] The numbers printed in light type, both in the text and in the tables, refer to birds which survived; those printed in heavy type refer to birds which perished.

striking difference between the two classes of birds. Both have an indicated average of 2.45 mm., although, to be more exact, the birds which perished averaged 2.449, while those that survived averaged 2.455, a difference too slight to be of material significance. This similarity of the two groups is not to be wondered at, since it is not to be expected that one eliminative agent will express itself in all possible anatomical features. Were the eliminative agent, for example, a severe northerly wind of protracted duration, the alar extent might then enter in as a factor of considerable selective value, and survivors would then have an alar extent materially different from that of the birds eliminated.

The alar extent of the females, corresponding with their smaller size, is less than that of the males.

Test 4: Weight. — Had I been called upon to express an opinion as to whether heavy or light birds would be more successful in resisting the severity of the February storm, I should have declared unhesitatingly in favor of the heavy birds. An examination of the third column of measurements, however, will show that the birds which survived invariably average less in weight than those which perished, and that among the males this difference amounts to more than a gram; that is, to about one twenty-fifth of the weight. The surviving birds of both sexes had an average weight of 25.2 grams, and those which succumbed had an average weight of 25.8 grams.

It may not be out of place to call attention here to certain objections which may be raised to the method which I have adopted, and to the conclusions thus far derived therefrom. One may claim that the greater relative number of females in the group of birds which perished vitiates the numerical result, since the females are of less stature than the males. But it will be noted that this objection answers itself, for the birds which perished are not shorter, but longer, than those which survived; and again, that the birds which perished, though having a disproportionate number of the lighter sex, nevertheless have an average weight considerably greater than that of the birds which survived. Moreover, comparing, in the two groups, adult males with adult males, young males with young males, and females with females, we find that the differences between the two classes of birds are expressed in these three smaller divisions, and I think we are justified in concluding that the differences are really significant.

The explanation that the birds which lived were those which sought, or at least enjoyed, better shelter cannot be entertained, for the storm was of long duration, and the birds were picked up, not in one locality, but in several localities; and, moreover, it is a fact that the survivors are *structurally different* from those which perished. If to these structural characters one desires to add also the intellectual character that the birds knew enough to go in out of the storm, the difference between the two groups becomes so much the greater.

Test 5: Length of Head. — A comparison of the average lengths of head, from the tip of the beak to the occiput, shows only a similarity between the survivors and those which perished, and indicates that under the present environmental conditions this feature is not sufficiently prominent to be expressed by this method of computation.

Test 6: Length of Humerus. — An examination of the fifth column of figures will show that the length of the arm bones of the birds which perished always averages less than that of the survivors. This difference is most conspicuous in the adult males, where the surviving birds have an average length of humerus of .738 of an inch, considerably more than that of their unfortunate companions, .727.

Here again I wish to emphasize the fact that these differences cannot be merely accidental, because they so often tend in the same direction. If among the survivors it is the proper thing for adult males to have a long humerus, then the young males have a long humerus, and the females follow the prevailing fashion with characteristic servitude. If a short humerus is an index of inferiority, all three groups of eliminated birds (adult males, young males, and females) bear this same mark of inferiority. This fact is the more striking since the averages are established on a relatively small number of birds, while usually in the statistical methods of the study of variation an abundance of material is necessary.

Test 7: Length of Femur. — An examination of the general averages on Tables III and III^a shows that the survivors possess longer thigh bones than do the birds which succumbed. The average length of femur in the former is .716 inch; in the latter .709. This difference in the averages cannot be ascribed to the large number of dead females, since the difference prevails also for both the adult and young males.

Test 8: Length of Tibio-Tarsus. — Measurement of the tibio-tarsus yields practically the same comparative data as the measurement of the upper bone of the leg, although in both groups of birds this bone in the females is considerably longer than in the adult males, notwithstanding that the females are smaller. This series of measurements agrees with the sixth, in that the young males have longer legs than the adult males.

Test 9. — Measurements across the skull, from the postorbital bone of one side to the postorbital bone of the other, are given in the eighth column, and are less satisfactory, perhaps, than those of other portions of the skeleton. The breadth of the cranium, as thus indicated, is somewhat less in the females than in the males. The averages denote that the birds which survived had wider heads than those which perished, but these averages are considerably influenced by data furnished by the young males. The irregularities in the subordinate groups induce me to place less confidence in these numerical results than in the results from measurements of other structures.

Test 10: Length of Sternum. — This test differs from other tests in that it relates to measurements in the longitudinal axis of the body. In the males the sternum is long, and in the females it is short. In the birds which survived it has a general average length of .845 inch; in those which perished it has a general average length of only .834.

I think these tests prove that there are fundamental differences between the birds which survived and those which perished. While the former are shorter and weigh less (*i.e.*, are of smaller body), they have longer wing bones, longer legs, longer sternums, and greater brain capacity. These characters are in accordance with our ideas of physical fitness; their defective development is evidently a mark of inferiority, and we are justified in concluding that the birds so handicapped failed to pass one of Nature's rigorous tests and perished.

In an earlier lecture, on the "Variations and Mutations of the Introduced Sparrow," facts were adduced which, it was claimed, were sufficient to show that the English sparrow, since its introduction into this country, has found life so easy that the operation of natural selection has been practically suspended, and that the American type consequently has become degenerate. No active agent had eliminated anomalies, and certain "freaks" had increased in number, until they had become over four times as numerous as in England.

When calling attention to the occurrence of these variations, and to the fact that they were an indication of the absence of an active eliminative factor, I little thought that within a few months I might witness the action of an eliminating factor that would test the structural qualifications of *all* the birds: destroy those which had departed unduly from the ideal type, and thus raise the general standard of excellence.

It will be recalled that, after the storm of February 1, one hundred and thirty-six birds were taken, and that, of these, seventy-two revived, while sixty-four failed to recover. But the fact that the birds which perished had in the *average* longer and larger bodies, and shorter head, wing, and leg bones, does not tell all the story of selective elimination.

Reference to the tables will show, not only that the longest bird perished, but also that the shortest bird perished. The longest bird was No. **33**, the shortest No. **40**. (In these and other cases of extreme departure from the mean, the exponent 1 is placed in the table beside the number of the bird.)

Again, if we examine the columns of figures which indicate the alar extent of the different birds, we find that both the bird with greatest spread of wings, No. **32**, and the one with least spread of wings, No. **52**, perished.

The heaviest bird, No. **23**, weighed 31 grams; it perished. The honors for lightness are evenly divided; No. 53, among the survivors, and No. **60**, among the eliminated, have the same weight, *viz.*, 22.6 grams.

The bird (No. **55**) whose head was longest (measured from the tip of the beak to the occiput) suffered elimination. The extreme variant in the opposite direction (No. 9) survived.

The honors for the longest humerus, .780 mm., are divided, Nos. 6 and **44**. The bird with the shortest humerus, No. **21**, perished.

The longest femur was possessed by bird No. **55**, the shortest by No. 51.

The surviving birds represent both extremes of variation of the tibio-tarsus (Nos. 18 and 41). In respect to all other columns of measurements the survivors possess exclusively never more than one of the *extreme* forms.

Both extremes of variation in width of cranium (Nos. **55** and **52**) are found among the eliminated birds.

The longest sternum is found in one of the surviving birds (No. 15), and it will be remembered that a long sternum was considered a mark of excellence. The shortest sternum (No. **52**) is found among the eliminated birds where the standard is low.

These extremes of variation are represented on Table IV, and by counting the dark numbers we find that eleven extreme positions (maximum or minimum) are occupied exclusively by the birds which perished, whereas the light numbers show that only five extreme positions are occupied by those which survived. In two cases (the minimum weight and the maximum length of humerus) the extreme positions are occupied alike by birds of both groups, and consequently I have left the spaces blank. In three cases two birds of the same group occupy the same extreme position, but the table is designed to indicate only the *extreme positions* and not the number of birds occupying them. The *number* of birds occupying these extreme positions is represented on the previous tables by the exponent 1, and if we count up these exponents, we shall find that among the surviving birds there are nine cases of this extreme type, whereas among the birds which perished there are fourteen cases. These numbers are the more impressive when one considers that, inasmuch as there are seventy-two of the former birds and only sixty-four of the latter, the chances for the occurrence of extreme variation are not equal in the two groups.

The birds which perished are at a decided disadvantage because of their smaller representation, yet there are many more "freaks" among them than among the surviving birds.

If it is thought that the association of the larger number of *extreme* variants with the eliminated birds is merely a matter of accident, we will not stop to argue the matter, but will apply the same test to the birds that remain after these extreme examples have been removed. We find even after the removal of these twenty-three examples, that extreme examples of the second order, indicated by the exponent 2, show the same tendency to occur more frequently among the eliminated birds.

The longest birds now are 166 mm.; the shortest, 153 mm. Of the former, Nos. **22**, **24**, **28**, **32**, **35**, perished, and No. 18 survived; of the latter, Nos. **45** and **62** perished, and Nos. 35, 54, and 55 survived.

If we count the times that the exponent 2 occurs in the tables, we shall find that there are ten birds of extreme abnormality of this second grade which survived, while there are twenty of the same grade which perished.

These figures indicate that the amplitude of variation of the surviving birds is less than that of the birds which perished. Were we to attempt the arrangement of the data into curves of distribution, the curve representing the distribution before the storm would be found to have a broad base, whereas the curve representing the distribution after the storm would be found to have a narrow base, for the eliminative process concentrated its energy on the individuals which occupied extreme positions.

Lest there remain some doubt as to the importance of this eliminative process, and of its efficiency in exterminating extreme variants, let us examine our figures again and see whether the group of birds which has already contributed thirty-four of the extremes of variation has still an excess of variability.

If we count up the exponents (3) of this third order of variable individuals, we find that the birds which survive give eleven examples, whereas those which perished give twenty-one. — It appears unnecessary to carry our investigations further along this line, for our results point always in one direction.

Natural selection is most destructive of those birds which have departed most from the ideal type, and its activity raises the general standard of excellence by favoring those birds which approach the structural ideal.

Inasmuch as the variation in structure in the birds which perished tends to centre about certain individuals, as, for example, Nos. **45**, **52**, and **55**, it might be claimed that the accidental presence of a few of these extremely abnormal individuals in this group is what really makes all the difference. Let us see.

There are twenty-three birds among the seventy-two survivors whose measurements bear exponents of extreme variation, and there are twenty-four birds similarly distinguished among the sixty-four which perished. But none of the birds in the first group has more than three exceptional features, whereas several of the birds which perished have a considerably larger number of exceptional features: four, five, and in one case, No. **52**, even six.

Of the twenty-three survivors which bear exponents, nineteen have only one exceptional character, and it is not surprising, considering the high standard of excellence possessed by these birds as a whole, that a single unfavorable feature does not prove fatal. There are but ten of the eliminated birds which have only one exceptional character, and the fact that some are burdened with more than one is apparently the reason for their mortality.

In an earlier contribution to the Study of Variation I called attention to a coincidence which may have considerable significance. When specimens of *Necturus* varied in respect to any one feature, there was a tendency for such specimens to present other and not necessarily correlated variations. Stated in another way, instability in respect to any one feature is an index of general organic instability. A similar coincidence of variations occurs among the sparrows.

Of the one hundred and thirty-six birds, five (Nos. 3, 47, 70, **21**, **52**) had albino feathers. Like other abnormalities endured by the surviving birds, albinism in two out of the three cases is the only affliction. But among those that were eliminated, where albinism twice occurs, it affects in one case a bird marked by four other abnormalities (No. 21), and in the other a bird (No. 52) already cursed by six abnormalities, the most miserable individual in the entire collection.

While we have shown that the birds which perished have certain average structural peculiarities which distinguish them from the survivors, and that the intensity of selective elimination has been felt most by birds of extreme structure, it remains to be shown that a *general instability* of structure is as characteristic of the birds which perished as a *general stability* of structure is characteristic of those which survived.

If we had sufficient data, this fundamental difference in the two groups of birds might be indicated by curves of distribution, one curve narrow and elevated, showing that its components are closely crowded around an ideal mean, the other broad and low, showing that its components are relatively indifferent to any ideal. But in the absence of sufficient data to illustrate the differences in this manner, we can arrive at a numerical result equally instructive by another method.

Having determined the ideal means for the several characters in each group of birds, we can then find the distance that each individual departs from this ideal. By adding these degrees of departure in respect to the several characters, and dividing by the number of individuals, we shall have numbers which represent the *average* departures from the ideal means. These numbers will be large if the members of a group of birds show a general tendency towards *disregard* of the ideals, and they will be small if the birds tend to crowd around the ideals. If all the birds actually attain the ideals, the number will be zero. — This is simply following out the principle that one man at the end of a ten-foot lever can do as much work as ten men at the end of a one-foot lever. A bird removed ten units from the mean exerts the same divergent influence upon its group that ten birds would exercise if removed one unit.

The results of this test, numerically expressed in Table V, are most instructive. In every case but one the numbers indicating the average departure from the ideal mean are smaller for the birds which survived, and thus indicate a general tendency toward conservatism on the part of the survivors. In the single exceptional case the numbers are not very different, 32 and 31. Granting this exception to the uniformity in the figures, it is exceedingly interesting to examine the series. In respect to length, the birds which perished had an average departure from the ideal mean expressed by the number 3.48, while the average departure of the birds which survived was only 2.51, or, expressed in tabular form:

In respect to length,	3.48	is greater than	2.51.
“ “ “ alar extent,	4.60	“ “ “	4.20.
“ “ “ weight,	12.6	“ “ “	10.9.
“ “ “ length of head,	5.64	“ “ “	5.51.
“ “ “ “ “ humerus,	20.1	“ “ “	16.0.
“ “ “ “ “ femur,	20.0	“ “ “	14.0.
“ “ “ “ “ tibio-tarsus,	33.8	“ “ “	29.4.
“ “ “ width of head,	12.	“ “ “	10., but
“ “ “ length of keel,	31.	“ less “	32.

A series of eight consecutive cases like the above, all pointing in the same direction, can hardly be considered accidental.

To summarize:

(1) We have found that there are fundamental differences between the surviving birds and those eliminated, and we conclude that the birds which survived survived because they possessed certain structural characters, and that the birds which perished perished not through accident, but because they did not possess certain structural characters which would have enabled them to withstand the severity of the test imposed by nature; they were eliminated because they were unfit.

(2) The process of selective elimination is most severe with extremely variable individuals, no matter in what direction the variations may occur. It is quite as dangerous to be conspicuously above a certain standard of organic excellence as it is to be conspicuously below the standard. It is the *type* that nature favors.

(3) Disregard of structural qualifications finally produces a throng of degenerates, whose destruction will follow the arrival of adversity.

TABLE I.

Measurements of Thirty-five Males which Survived.

	TOTAL LENGTH.	ALAR EXTENT.	WEIGHT.	LENGTH OF BEAK AND HEAD.	LENGTH OF HUMERUS.	LENGTH OF FEMUR.	LENGTH OF TIBIO-TARSUS.	WIDTH OF SKULL.	LENGTH OF KEEL OF STERNUM.
1 ♂	154^3	241	24.5	31.2	.687	.668	1.022^2	.587	.830
2 ♂	160	252	26.9	30.8	.736	.709	1.180	.602	.841
3 ♂	155	243	26.9	30.6	.733	.704	1.151	.602	.846
4 ♂	154^3	245	24.3	31.7	.741	.688	1.146	.584	.839
5 ♂	156	247	24.1	31.5	.715	.706	1.129	.575	.821
6 ♂	161	253	26.5	31.8	$.780^1$	.743	1.144	.607	.893
7 ♂	157	251	24.6	31.1	.741	.736	1.153	.610	.862
8 ♂	159	247	24.2	31.4	.728	.718	1.126	.609	.793
9 ♂	158	247	23.6	29.8^1	.703	.673	1.079	.602	.820
10 ♂	158	252	26.2	32.	.749	.739	1.153	.614	.857
11 ♂	160	252	26.2	32.	.741	.723	1.129	.624	.892
12 ♂	162	253	24.8	32.3	.766	.752	1.134	.633	$.923^2$
13 ♂	161	243	25.4	31.8	.721	.722	1.126	.597	.891
14 ♂	160	250	23.7	29.8^1	.730	.703	1.103	.590	.820
15 ♂	159	247	25.7	31.4	.729	.717	1.141	.592	$.927^1$
16 ♂	158	253	25.7	31.9	.743	.699	1.150	.600	.860
17 ♂	159	247	26.5	31.6	.733	.714	1.155	.611	$.923^2$
18 ♂	166^2	253	26.7	32.5	.767	$.765^2$	1.230^1	.600	.878
19 ♂	159	247	23.9	31.4	.752	.723	1.113	.602	.825
20 ♂	160	248	24.7	31.3	.752	.737	1.176	.603	.803
21 ♂	161	252	28.	31.8	$.770^2$	.731	1.190	.590	.885
22 ♂	163	251	27.9	31.9	$.769^3$	.745	1.168	.622	.860
23 ♂	156	242	25.9	32.	.723	.711	1.116	.609	.886
24 ♂	165^3	251	25.7	32.2	.751	.742	1.161	.613	.865
25 ♂	160	247	26.6	32.4	.728	.707	1.108	.590	.836
26 ♂	158	244	23.2^3	31.6	.730	.713	1.142	.585	.888
27 ♂	160	242	25.7	31.6	.709	.705	1.124	.620	.788
28 ♂	157	245	26.3	32.2	.741	.726	1.143	.595	.850
29 ♂	159	244	24.3	31.5	.723	.698	1.107	.615	.847
30 ♂	160	253	26.7	32.1	.739	.714	1.117	.592	.864
31 ♂	158	245	24.9	31.4	.726	.703	1.119	.580	.854
32 ♂	161	247	23.8	31.4	.735	.694	1.101	.602	.789
33 ♂	160	247	25.6	32.3	.756	.745	1.135	.607	.902
34 ♂	160	247	27.	32.	.755	.736	1.174	.631	.873
35 ♂	153^2	241	24.7	32.2	.728	.680	1.092	.592	.884
Average . .	159	247	25.4	31.6	.738	.716	1.135	.602	.857

TABLE I a.

Measurements of Twenty-four Adult Males which Perished.

	TOTAL LENGTH.	ALAR EXTENT.	WEIGHT.	LENGTH OF BEAK AND HEAD.	LENGTH OF HUMERUS.	LENGTH OF FEMUR.	LENGTH OF TIBIO-TARSUS.	WIDTH OF SKULL.	LENGTH OF KEEL OF STERNUM.
1 ♂	165^{3}	249	26.5	31.	.738	.704	1.095	.606	.847
2 ♂	160	245	26.1	32.	.736	.709	1.109	.611	.842
3 ♂	161	249	25.6	32.3	.743	.718	1.128	.602	.828
4 ♂	162	246	25.9	32.3	.738	.709	1.135	.607	.869
5 ♂	163	250	25.5	32.5	.752	.731	1.197	.623	.888
6 ♂	162	247	27.6	31.8	.731	.719	1.113	.597	.869
7 ♂	163	246	25.8	31.4	.689	$.662^{3}$	1.073	.604	.836
8 ♂	161	246	24.9	30.5	.739	.726	1.138	.580	.803
9 ♂	160	242	26.	31.	.745	.713	1.105	.600	.803
10 ♂	162	246	26.5	31.5	.720	.696	1.092	.606	.809
11 ♂	160	249	26.	31.4	.726	.689	1.097	.602	.850
12 ♂	161	250	27.1	31.6	.737	.711	1.120	.631	.852
13 ♂	162	248	25.1	31.9	.744	.722	1.154	.591	.839
14 ♂	165^{3}	252	26.	32.3	.726	.710	1.145	.609	.887
15 ♂	161	243	25.6	32.5	.709	.707	1.122	.607	.832
16 ♂	161	244	25.	31.3	.702	.685	1.082	.595	.874
17 ♂	162	248	24.6	31.	.713	.700	1.086	.590	.837
18 ♂	164	244	25.	31.2	.703	.690	1.074	.608	.795
19 ♂	158	247	26.	32.	.729	.710	1.145	.607	.803
20 ♂	162	253	28.3	31.8	.752	.718	1.152	.600	.857
21 ♂	156	239	24.6	30.5	$.659^{1}$	$.658^{2}$	1.042^{3}	$.570^{3}$	.810
22 ♂	166	251	27.5	31.5	.720	.691	1.118	.612	.847
23 ♂	165^{3}	253	$31.^{1}$	32.4	.765	.750	1.183	.613	.905
24 ♂	166^{2}	250	28.3	32.4	.754	.718	1.179	.607	$.916^{3}$
Average . .	162	247	26.2	31.6	.727	.706	1.120	.603	.845

TABLE II.

Measurements of Sixteen Young Males which Survived.

	Total Length.	Alar Extent.	Weight.	Length of Beak and Head.	Length of Humerus.	Length of Femur.	Length of Tibio-tarsus.	Width of Skull.	Length of Keel of Sternum.
36 juv. ♂	156	246	24.6	32.	.741	.735	1.167	.592	.849
37 juv. ♂	156	245	25.5	32.1	.761	.717	1.147	.620	.816
38 juv. ♂	163	248	24.8	32.2	.742	.733	1.165	.606	.854
39 juv. ♂	163	248	26.3	33.[3]	.736	.704	1.148	.609	.839
40 juv. ♂	160	250	24.4	31.5	.746	.715	1.173	.604	.893
41 juv. ♂	156	237	23.3	30.6	.692	.664	1.011[1]	.588	.774
42 juv. ♂	162	253	26.7	32.	.759	.734	1.197	.630	.878
43 juv. ♂	163	254[3]	26.4	32.	.766	.750	1.165	.605	.886
44 juv. ♂	164	251	26.9	32.	.755	.742	1.171	.620	.886
45 juv. ♂	163	244	24.3	31.3	.718	.680	1.082	.610	.892
46 juv. ♂	160	247	27.	31.5	.764	.732	1.177	.617	.846
47 juv. ♂	160	250	26.8	32.5	.764	.729	1.123	.635[3]	.842
48 juv. ♂	158	247	24.9	32.4	.745	.724	1.139	.588	.865
49 juv. ♂	158	249	26.1	32.2	.742	.736	1.148	.602	.817
50 juv. ♂	158	243	26.6	32.4	.747	.711	1.163	.612	.891
51 juv. ♂	155	237	23.3	30.2[3]	.685	.653[1]	1.011[1]	.587	.794
Average . .	159	246	25.4	31.8	.741	.716	1.136	.607	.851

TABLE II^a.

Measurements of Twelve Young Males which Perished.

	TOTAL LENGTH.	ALAR EXTENT.	WEIGHT.	LENGTH OF BEAK AND HEAD.	LENGTH OF HUMERUS.	LENGTH OF FEMUR.	LENGTH OF TIBIO-TARSUS.	WIDTH OF SKULL.	LENGTH OF KEEL OF STERNUM.
25 juv. ♂	160	249	24.2	30.4	.740	.717	1.130	.620	.840
26 juv. ♂	156	236	26.8	30.^{2}	.690	.671	1.067	.563^2	.832
27 juv. ♂	158	240	23.5	31.	.715	.702	1.113	.595	.805
28 juv. ♂	166^2	245	26.9	31.7	.715	.695	1.107	.601	.847
29 juv. ♂	165^3	255^2	28.6	31.5	.766	.744	1.175	.613	.854
30 juv. ♂	157	238	24.7	31.2	.680^3	.677	1.156	.599	.769
31 juv. ♂	164	250	27.3	31.8	.764	.726	1.171	.588	.860
32 juv. ♂	166^2	256^1	25.7	31.7	.752	.751	1.187	.595	.858
33 juv. ♂	167^1	255^2	29.^{3}	32.2	.765	.745	1.197	.638^2	.855
34 juv. ♂	161	246	25.	31.5	.739	.707	1.123	.587	.850
35 juv. ♂	166^2	254^3	27.5	31.4	.760	.742	1.124	.604	.914
36 juv. ♂	161	251	26.	31.5	.731	.707	1.122	.589	.828
Average . .	162	248	26.2	31.5	.734	.715	1.141	.599	.842

TABLE III.

Measurements of Twenty-one Adult and Young Females which Survived.

	Total Length.	Alar Extent.	Weight.	Length of Beak and Head.	Length of Humerus.	Length of Femur.	Length of Tibio-tarsus.	Width of Skull.	Length of Keel of Sternum.
52 ♀	156	245	25.3	31.6	.729	.710	1.152	.620	.809
53 ♀	154[3]	240	22.6[1]	30.4	.705	.686	1.103	.584	.770
54 ♀	153[2]	240	25.1	31.	.724	.713	1.123	.585	.812
55 ♀	153[2]	236	23.2[3]	30.9	.698	.678	1.132	.596	.795
56 ♀	155	243	24.4	31.5	.734	.736	1.170	.596	.801
57 ♀	163	247	25.1	32.	.748	.734	1.166	.602	.821
58 ♀	157	238	24.6	30.9	.726	.727	1.175	.588	.797
59 ♀	155	239	24.	32.8	.732	.742	1.175	.601	.835
60 ♀	164	248	24.2	32.7	.752	.752	1.201	.604	.830
61 ♀	158	238	24.9	31.	.741	.689	1.091	.592	.866
62 ♀	158	240	24.1	31.3	.733	.706	1.107	.591	.867
63 ♀	160	244	24.	31.1	.731	.730	1.152	.589	.808
64 ♀	161	246	26.	32.3	.758	.732	1.154	.623	.859
65 ♀	157	245	24.9	32.	.752	.740	1.186	.593	.787
66 ♀	157	235	25.5	31.5	.712	.704	1.132	.611	.781
67 ♀	156	237	23.4	30.9	.708	.691	1.123	.613	.798
68 ♀	158	244	25.9	31.4	.729	.705	1.146	.597	.851
69 ♀	153[2]	238	24.2	30.5	.715	.707	1.116	.595	.821
70 ♀	155	236	24.2	30.3	.727	.705	1.120	.585	.790
71 ♀	163	246	27.4	32.5	.732	.711	1.163	.630	.862
72 ♀	159	236	24.	31.5	.709	.713	1.129	.607	.845
Average . .	157	241	24.6	31.4	.728	.714	1.143	.600	.819
General average for 72 birds . . .	158	245	25.2	31.6	.736	.716	1.138	.603	.845

TABLE III a.

Measurements of Twenty-eight Adult and Young Females which Perished.

	Total Length.	Alar Extent	Weight.	Length of Beak and Head.	Length of Humerus.	Length of Femur.	Length of Tibio-tarsus.	Width of Skull.	Length of Keel of Sternum.
37 ♀	155	240	26.3	31.4	.709	.710	1.125	.614	.815
38 ♀	156	240	25.8	31.5	.715	.678	1.127	.597	.812
39 ♀	160	242	26.	32.6	.740	.732	1.157	.597	.854
40 ♀	152^{1}	232^{3}	23.2^{3}	30.3	$.676^{2}$	.683	1.048	.590	.780
41 ♀	160	250	26.5	31.7	.741	.731	1.187	.615	.886
42 ♀	155	237	24.2	31.	.727	.723	1.118	.610	.787
43 ♀	157	245	26.9	32.2	.766	.751	1.227^{2}	.620	.841
44 ♀	165^{3}	245	27.7	33.1^{2}	$.780^{1}$	$.757^{3}$	1.195	.633	.895
45 ♀	153^{2}	231^{2}	23.9	30.1	$.680^{3}$	$.662^{3}$	1.042^{3}	.592	.781
46 ♀	162	239	26.1	30.3	.709	.685	1.092	.587	.911
47 ♀	162	243	24.6	31.6	.741	.729	1.162	.605	.840
48 ♀	159	245	23.6	31.8	.727	.700	1.129	.610	.855
49 ♀	159	247	26.	30.9	.711	.666	1.098	.580	$.749^{2}$
50 ♀	155	243	25.	30.9	.730	.711	1.127	.598	.839
51 ♀	162	252	24.8	31.9	.752	.738	1.180	.615	.875
52 ♀	152^{1}	230^{1}	22.8^{2}	30.4	.682	.664	1.042^{3}	$.551^{1}$	$.734^{1}$
53 ♀	159	242	24.8	30.8	.717	.667	1.090	.575	.809
54 ♀	155	238	24.6	31.2	.706	.702	1.102	.588	$.758^{3}$
55 ♀	163	249	30.5^{2}	33.4^{1}	.767	$.767^{1}$	1.207^{3}	$.640^{1}$	.896
56 ♀	163	242	24.8	31.	.713	.713	1.128	.607	.813
57 ♀	156	237	23.9	31.7	.718	.716	1.090	.611	.800
58 ♀	159	238	24.7	31.5	.726	.701	1.145	.600	.800
59 ♀	161	245	26.9	32.1	.751	.704	1.142	.607	.819
60 ♀	155	235	22.6^{1}	30.7	.695	.692	1.119	.584	.771
61 ♀	162	247	26.1	31.9	.751	.735	1.157	.618	.802
62 ♀	153^{2}	237	24.8	30.6	.732	.718	1.172	.594	.802
63 ♀	162	245	26.2	32.5	.728	.731	1.102	.614	.832
64 ♀	164	248	26.1	32.3	.739	.707	1.159	.592	.823
Average . .	158	241	25.3	31.4	.726	.709	1.131	.601	.820
General average for 64 birds . . .	160	245	25.8	31.5	.728	.709	1.128	.601	.834

TABLE IV.

The Maximum and Minimum Measurements.

	TOTAL LENGTH.	ALAR EXTENT.	WEIGHT.	LENGTH OF BEAK AND HEAD.	LENGTH OF HUMERUS.	LENGTH OF FEMUR.	LENGTH OF TIBIO-TARSUS.	WIDTH OF SKULL.	LENGTH OF KEEL OF STERNUM.
Maximum . .	**167**	**256**	**31**	**33.4**	(*d*)	**.767**	1.230	**.640**	.927
Minimum . .	(*a*) **152**	**230**	(*b*)	(*c*) 29.8	**.659**	.653	(*c*) 1.011	**.551**	**.734**

(*a*) The minimum length (152 mm.) occurs twice among the birds which perished : Nos. **40** and **52**.
(*b*) The minimum weight (22.6 grams) occurs in each group : Nos. 53 and **60**, and therefore is not entered.
(*c*) The minimum length of head (29.8 mm.) and of tibio-tarsus (1.011 inch) occurs twice among the surviving birds : Nos. 9 and 14, 41 and 51.
(*d*) The maximum length of humerus (.780 inch) occurs in each group : Nos. 6 and **44**, and therefore is not entered.

TABLE V.

Average Departures from Ideal Mean.

	TOTAL LENGTH.	ALAR EXTENT.	WEIGHT.	LENGTH OF BEAK AND HEAD.	LENGTH OF HUMERUS.	LENGTH OF FEMUR.	LENGTH OF TIBIO-TARSUS.	WIDTH OF SKULL.	LENGTH OF KEEL OF STERNUM.
Seventy-two which survived	2.51	4.20	10.9	5.51	16.	14.	29.4	10.	32.
Sixty four which perished	**3.48**	**4.60**	**12.6**	**5.64**	**20.1**	**20.**	**33.8**	**12.**	**31.**

TWELFTH LECTURE.

ON THE HEREDITY OF THE MARKING IN FISH EMBRYOS.

JACQUES LOEB.

I.

UNTIL recently heredity has been treated chiefly as a problem for whose solution one single theory or one single principle was considered possible and sufficient. The theories of heredity by Darwin, Weismann, Spencer, and Jaeger are proofs of this. Heredity is explained by Darwin on the assumption that the organs of the parents give off particles from which the organs of the offspring originate. According to Weismann, the egg contains determinants for every organ contained in the later organism and which have a definite arrangement. According to Jaeger, the offspring resembles the parents because both consist of the same chemical constituents. None of these theories of heredity have been generally accepted, nor do I believe they ever will be. They overlook the fact that heredity is a collective term for a series of heterogeneous circumstances which cannot possibly be explained by one principle. Thus it happens that while each of these theories is adapted to some of the facts of heredity, other facts can be better explained by some other theory.

In contradistinction to these attempts to explain heredity by one single principle and by means of one single theory, a more analytical study of the subject has been undertaken. This has led to the conception that very different circumstances determine the various details in heredity. We thus find ourselves face to face with the task of investigating in detail which circumstances determine the single facts of heredity. The change in our attitude toward this problem is similar to that which has taken place in psychology. Psychologists no longer try to give a theory of the soul or of consciousness, but try to analyze the various groups of psychical phenomena more or less independently of each other.

It may be desirable to illustrate what we mean by the analytical study of heredity. From the egg of the sea urchin only one embryo originates. If we isolate the first two cleavage cells, we get two embryos. This agrees very well with Jaeger's chemical theory, but it does not agree with Weismann's. I have made an experiment which seems to show that neither Jaeger's nor Weismann's theory is correct. If the contents of the egg be transformed into a double sphere before segmentation sets in, either a dumb-bell-shaped blastula or two blastulae are formed which are at first grown together, but as a rule are separated later. *It seems that a blastula originates by the cleavage cells being forced to migrate to the limit between sea water and egg* or *to the surface of the egg*. Probably tropisms of the cleavage cells, chemotropism, or stereotropism are involved in this. It is easy to observe that the cleavage cells of the sea urchin's egg can execute amoeboid motions. Hence we have to deal with a circumstance which neither the theory of Jaeger nor of Sachs nor of Weismann nor of any other author could anticipate; namely, the necessity of the cleavage cells creeping to the periphery of the egg.

We may select another circumstance, for instance, the heredity of the arrangement of organs in an organism. From the germ of Hydroids an organism develops which has stolons at one end of its body and polyps at the other. This definite arrangement is in many cases due to a fact which none of the general theories of heredity could have predicted. We find in many Hydroids that an excised piece of the stem forms stolons if the end comes in contact with solid bodies, while the same end forms polyps if it is surrounded by sea water. When a germ or a larva settles down at the close of the swimming stage, that end of the germ which is in contact with a solid body will produce stolons, while the other end, that is in contact with sea water, must give rise to polyps. Hence the hereditary arrangement of organs is in such cases due to the fact that while one side of the germ comes in contact with solid bodies, the other end remains in contact with sea water.

Let us next consider the hereditary power of assimilation. We know that the corresponding proteids of a horse, a dog, and a cat are different. If we feed a cat with horse flesh, it transforms it into the specific proteids of a cat, while in a dog the same horse flesh is transformed into the specific proteids of a dog. For this transformation specific enzymes are necessary which are different in both forms. As the fertilized egg is able to carry on processes of assimilation, we must assume that such enzymes are transmitted through the egg to the offspring. The presence of enzymes or zymogens, from which the ferments originate, is another circumstance in heredity which none of the general theories has considered. We see thus that what we call heredity is composed of very heterogeneous constituents which cannot be explained by one single principle or theory, unless the theory should become so indefinite as to be almost meaningless. But such a theory would cease to be of value. This paper will deal especially with the heredity of the markings in fish embryos, which will offer another example of the analytical study of heredity.

II.

Seven years ago I published a series of observations on the origin of the markings in the yolk sac of the embryo of Fundulus. The yolk sac of Fundulus possesses a tiger-like coloration. Its origin is as follows: black and red chromatophores are found on the surface of the yolk sac. They gradually creep upon the blood vessels and form a sheath around them. This creeping of the chromatophores upon the blood vessels is due to a tropism of the former. I then believed it was chemotropism and assumed that the chromatophores were oriented by a chemical constituent of the blood, especially by oxygen. But it may be that it is stereotropism, or that both tropisms coöperate. The heredity of the marking is, therefore, in this case determined by a stimulus which the blood vessels exercise upon another tissue, namely, the chromatophores. Both tissues are formed rather independently of each other, but from the fact that the chromatophores must creep upon the blood vessels, and that the latter have a hereditary arrangement, the marking becomes hereditary too. This contradicts those theories of heredity which try to derive all the peculiarities of the animal from corresponding peculiarities of the sexual cell, for instance, Weismann's theory.

The observations I made on Fundulus have since been confirmed for other classes of animals too. Dr. Arnold Graf made similar observations in leeches, and Zennek has found that the longitudinal stripes in the Ringelnatter are determined by blood vessels. But as he has overlooked my papers on the subject he did not realize that the marking is produced by the chromatophores being bound to creep upon the blood vessels. I have continued my observations on the yolk sac and the embryo of Fundulus, and will try to explain the history of the origin of the marking in connection with some drawings.

Fundulus has two kinds of pigment cells: (1) large black cells with almost no ramifications and processes, and (2) small red pigment cells with an enormous amount of ramification. Both differ somewhat in their reactions. The black cells react and move quicker than the red cells. In Fig. 3 the large black pigment cells can easily be discriminated from the star-shaped red pigment cells. The latter are characterized in the drawings by fine lines.

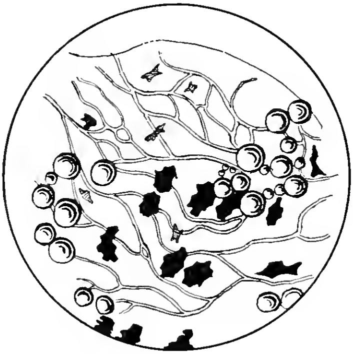

Fig. 1.

At first the yolk sac is free from pigment cells. As soon as the vessels and the blood corpuscles develop, the first pigment cells make their appearance on the yolk sac. Fig. 1 shows this stage. Some of the large black pigment cells are situated on blood vessels, others are scattered in the gaps between the capillaries, but there is no definite relation between the position of the pigment cells and the blood vessels, and there is no definite marking. The same is true in regard to the red pigment cells. (The little spheres are oil globules.)

A few days later the number of capillaries as well as that of the pigment cells has increased (Fig. 2), but a further change is also noticeable; each pigment cell has crept upon a blood vessel, and the shape of the black pigment cells has changed somewhat. Now they are all rather long; that is, they follow the direction of the blood vessels. In the small ramified red pigment cells no change of form is as yet perceptible. Fig. 3 shows a very characteristic condition, such as is usually found toward the end of the first week. The black pigment cells follow the blood vessels entirely. Where a blood vessel branches off into two parts the pigment cells show the same ramification. The black pigment cells now form a sheath around the blood vessels.

Fig. 4 shows the yolk sac when the marking is almost completed. The black and red pigment cells surround the blood vessels so completely that no one who sees the eggs for the first time at this period could realize that the black and red stripes in the yolk sac could have originated from chromatophores. That part of the blood

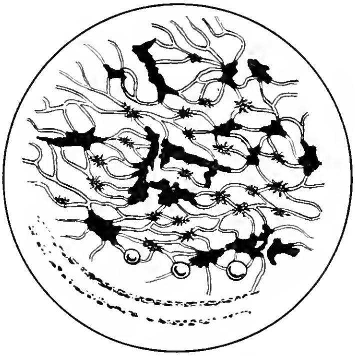

Fig. 2.

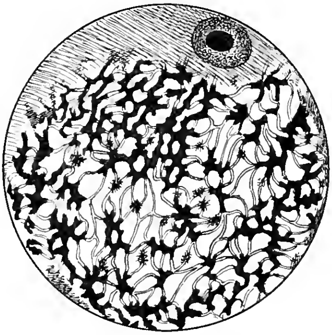

Fig. 3.

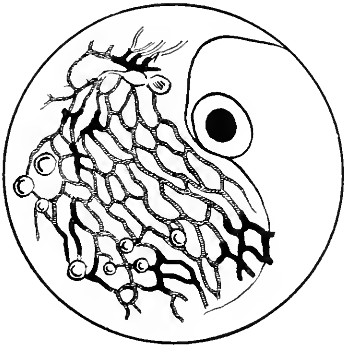

Fig. 4.

vessels which is marked by fine lines is covered by red pigment cells, the other part by black pigment cells.

In my first publication on the subject I confined myself to a description of the origin of the marking in the yolk sac. I mentioned, however, that in the embryo itself the marking seemed to originate in the same way. I have since examined the origin of the marking in the embryo more closely, and have found it the same as in the yolk sac. Fig. 5 shows a comparatively young embryo, in which the pigment cells are still scattered irregularly. Later on, however, the pigment cells here, too, creep upon the blood vessels. Fig. 6 shows the blood vessels and pigment cells in the tail of an older embryo in which the marking has taken place. In some of the blood vessels the direction of the blood flow is indicated by arrows. This drawing, like all the others, was taken from life.[1] We see that in the embryo, as in the yolk sac, all the pigment cells have crept upon blood vessels. I should like to direct the attention of the reader to one special point: in the middle of the body two large blood vessels, an artery and a vein, run side by side. They are marked by two arrows. Here all the chromatophores have crept upon the artery while the vein has remained free. It is obvious that the artery contains more oxygen than the vein, and this seems to suggest

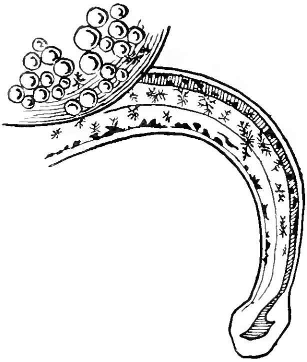

Fig. 5.

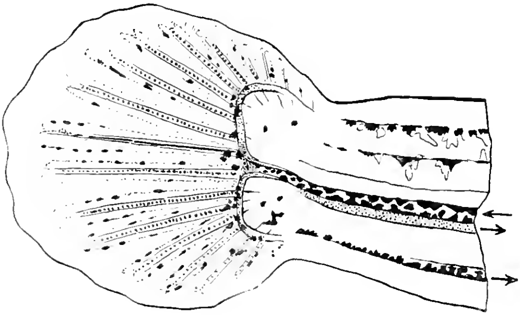

Fig. 6.

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[1] These drawings were made by Mr. Bridgham, of Providence, R. I., under my direction.

that the oxygen of the blood may be one of the causes that force the chromatophores to creep upon the blood vessels. I have expressed a similar belief in my former papers, but the oxygen in the arterial blood is possibly not the only reason why the chromatophores creep upon the blood vessels, since wherever a vein is isolated they creep upon the vein too. Thus it seems that positive chemotropism for oxygen is one stimulus, but possibly not the only one that causes the migration of the chromatophores upon the blood vessels.

III.

The further examination of the marking of the Fundulus embryo has shown that the blood vessels are not the only factors that determine the marking. A second factor is the central nervous system. The back of the embryo is colored black by pigment cells which follow the brain and the spinal column also. They follow the outline of these organs almost as closely as they follow the blood vessels. The details of the origin of this marking must be reserved for future investigation.

Everything we have said thus far about the marking of the embryo refers to chromatophores, but in addition to chromatophores two other elements aid in producing the markings in fish embryos. In certain places a diffused yellowish pigment is deposited. It is a kind of excretion. The chemical character of this pigment and its origin have yet to be determined. This pigment appears at a rather early stage in the development. The second element is the structural colors. They do not play as much of a rôle in the early stages of the embryo. I intend to continue my study of the development of the marking in these embryos, especially in regard to the influence of hybridization. I have already begun experiments on the latter topic, but they are not far enough advanced to be published.

Finally, may I be pardoned for adding a few remarks concerning the theories of Eimer. I am no believer in Weismann's theories of heredity, but it seems to me that the so-called theories of Eimer on the origin of the longitudinal striation in animals are nothing but a play on words. One of his general "laws" maintains that every marking is at first longitudinal. The truth of the matter is that most animals are not spherical, and that most organs, especially in segmented animals, run in a longitudinal direction through the animal; for instance, the spinal cord, vertebral column, blood vessels, intestine, etc. In the embryo it is especially true for the large blood vessels. The first striation in such embryos is, of course, longitudinal, simply because it is determined by the blood vessels; but wherever we have to deal with the coloration in a spherical organ, as, for instance, in the yolk sac of the Fundulus embryo, the absurdity of Eimer's law becomes evident. Here, on the surface of the sphere, there is no longitudinal direction, and yet here, too, the chromatophores follow the blood vessels. In the same way the so-called law of Eimer appears absurd if we consider the marking, which is determined by the pigment cells that cover the brain.

Although it seems to me that at present we cannot avoid dealing with the problem of heredity from an analytical point of view, it is clear that this is only a temporary necessity. Sooner or later the facts found by the analytical method must be utilized for the establishment of more general laws.

THIRTEENTH LECTURE.

DO THE REACTIONS OF LOWER ANIMALS DUE TO INJURY INDICATE PAIN-SENSATIONS?

W. W. NORMAN.

Associate Professor of Biology, University of Texas.

A brief note by me concerning pain-sensations in the lower animals appeared in *Pflüger's Archiv* for 1896.[1] It was my aim to give experimental proof that the reactions of lower animals upon injury furnish no safe evidence of pain-sensations. For these experiments chiefly the earthworm (*Allolobophora fœtida*) was used.

It had already been observed by Friedländer[2] that when an earthworm is cut in two in the middle, the two halves react differently — the anterior half crawling away and burying itself like a normal worm, the posterior end making strong winding and jerking motions. [Unmittelbar nach der Operation machen sie (die geköpften Würmer) heftige schlagende und windende Bewegungen.] Loeb[3] had observed similar motions in other worms similarly treated.

If Friedländer had carried his experiment further, namely, if he had cut each half of the already halved worm in two in the middle, the results would have become, indeed, still more striking. It is here that my experiment presents an essentially new point.

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[1] "Dürfen wir aus den Reactionen niederer Thiere auf das Vorhandensein von Schmerzempfindungen schliessen?" *Pflüger's Archiv*, Bd. lxvii, p. 137.

[2] Friedländer, "Ueber das Kriechen der Regenwürmer," *Biol. Centralblatt*, Bd. viii.

[3] Loeb, J., "Beiträge der Gehirnphysiologie der Würmer," *Pflüger's Archiv*, Bd. lvi.

For convenience let us call the anterior half of the worm *a* and the posterior half *b*. Now if *a* be cut in two in the middle, the anterior piece a_1 elongates and begins to crawl, while the posterior piece a_2 acts like *b*; *i.e.*, executes jerking and squirming motions. Thus far in the experiment each piece that began the progressive movements immediately after the injury was the anterior piece, containing the so-called brain. Now if we cut in two the posterior half *b*, we get just the same kind of reactions as in case *a* — namely, the anterior part b_1 elongates and begins progressive motion, while the posterior piece b_2 acts as a_2, carrying out a series of jerking and squirming motions. We observe, then, that each time after the halving, the anterior piece elongates without any irregular muscular contractions and begins to crawl forwards, while the posterior piece always executes the same kind of squirming motions.

The experiment may be carried still further, and each of the four pieces cut in two. In each case the posterior half (one-eighth of the whole worm) immediately begins the jerking and squirming motions, while the corresponding anterior half elongates and, if not too short, begins to crawl. Indeed, if small vigorous worms be used, the characteristic reactions of the two halves may be observed in pieces that are only about five millimeters in length.

As to the significance of the reactions of the lower animals, there are two opposite views. Those supporting the one view assume that the reactions to external stimuli are the outward expression of psychical processes resulting therefrom. The larvæ of crustaceans, for example, may prefer blue light to red, if there be opportunity for choice. "The flesh-fly, *Musca carnaria*, deposits its eggs in the flowers of the carrion plant, the smell of which resembles that of putrid meat, and so deceives (!) the fly."[1] Again, according to the same way of reasoning, the moth flies into the flame, prompted by curiosity to get acquainted with the strange object.

The supporters of this view ignore the difficulty that confronts them — namely, that the only means of determining the presence of consciousness in the lower animals are the

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[1] Romanes, Mental Evolution in Animals, p. 167.

movements themselves, which in turn are to be explained through psychical processes. Under the title "Mechanik," Verworn,[1] for example, expresses himself as follows: "The question might be raised as to the possibility of reaching a conclusion concerning the subjective conditions of another organism, since they lie outside the investigator. The following consideration, however, proves the possibility of gaining definite knowledge in this direction. We know from our own subjective experiences our own subjective states, and, on the other hand, the objective expressions which characterize them. Here we have two known quantities. A third known quantity we can get through the objective expressions of psychical processes (!) of the organisms under observation. If now we make a proportion between the processes of man and of the organism to be studied, *i.e.*, if we compare the two with each other, we can reach a conclusion as to the subjective processes of the organism under consideration according to the principle $\frac{x}{a}=\frac{c}{b}$, $x=a\cdot\frac{c}{b}$, in which a represents the subjective processes of man, c the objective expression of the same, b the objective expression of the subjective processes of the object under investigation, and x its unknown subjective processes to-be-found-out."

Man könnte bestreiten, dass es überhaupt möglich sei, irgend welchen Aufschluss über die subjectiven Zustände eines anderen Organismus zu erlangen, da dieselben ausserhalb des Untersuchers liegen. Indessen beweist die folgende Ueberlegung trotzdem die Möglichkeit, wenigstens durch Schlüsse sichere Erfahrungen in dieser Richtung zu gewinnen. Wir kennen aus unserer subjectiven Erfahrung einerseits unsere eigenen subjectiven Zustände und andererseits die objectiven Aeusserungen welche dieselben characterisiren. Hierin besitzen wir aber zwei bekannte Grössen. Eine dritte bekannte Grösse können wir uns schaffen durch die Beobachtung der objectiven Aeusserungen psychischer Vorgänge bei den zu untersuchenden Organismen. Wenn nun zwischen den betreffenden Vorgängen beim Menschen und bei den Untersuchungsobjecten eine Proportion gesetzt wird, d. h. wenn beide mit einander verglichen werden, so kann man nach dem Prinzip $\frac{x}{a}=\frac{c}{b}$, $x=a\cdot\frac{b}{c}$, wobei a

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[1] Verworn, Psycho-Physiologische Protisten-Studien, p. 18. Jena, 1889.

die subjectiven Vorgänge beim Menschen, *c* deren objective Aeusserungen, *b* die objectiven Aeusserungen der subjectiven Vorgänge bei den Versuchsobjecten und *x* die zu erforschenden subjectiven Zustände dieser selbst sind, Aufschluss über die subjectiven Vorgänge der betreffenden Organismen erlangen.

The striking feature in this method is the assumption of the fact to be proved — namely, the existence of psychical processes in the organisms to be investigated. According to the equation given, not only the whole earthworm but any isolated piece of the same would be capable of psychical processes, since upon injury it reacts in the same manner as the whole worm.

Those holding the second view seek to analyze the reactions of animals on purely mechanical grounds. The most pronounced defender of this view is Loeb.[1] He has by this method shown that the phenomena of orientation of animals towards light agree in every particular with the phenomena of orientation of plants towards the same source of stimulus. Hence the heliotropic reactions of plants must be referred to "curiosity," or to some other anthropomorphic process, or else it must be admitted that the phenomena of orientation of animals are to be explained just as mechanically as in the case of plants.

That consciousness is a function of associative memory (associatives Gedächtniss) has been emphasized by Loeb.[2] But memory has thus far been proved with certainty only in such forms as have a well-developed cerebrum. The facts of brain physiology speak decidedly against the view that phenomena of consciousness are everywhere present in the animal kingdom.[3]

It cannot, however, be denied that certain reactions of lower animals against injury, which in man cause pain, lead the inexperienced person easily to the conclusion that these animals really suffer pain. An earthworm, for example, touched with

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[1] Loeb, J., Der Heliotropismus der Thiere und seine Uebereinstimmung mit dem Heliotropismus der Pflanzen. Würzburg, 1890.

[2] Loeb, J., "Beiträge der Gehirnphysiologie der Würmer," *Pflüger's Archiv*, Bd. lvi.

[3] Loeb, J., ". . . Zur Physiologie und Psychologie der Actinien," *Pflüger's Archiv*, Bd. lix.

a needle, or otherwise slightly stimulated, may be thrown into violent jerking and squirming motions. According to Loeb we have no more right in this case to conclude that these motions are due to pain-sensations than we have to make a similar conclusion upon the contractions of an isolated frog's muscle when put into a too concentrated salt solution. My experiments upon the earthworm clearly prove that this view in contrast to that of the pain-sensations is the correct one.

It has already been observed that if we attribute feeling to the earthworm because of certain reactions, we must do likewise for any isolated piece of the worm; and, further, that the *front half* of any such piece must have a state of feeling different from that of the posterior half. But since the piece to be experimented upon may be taken from any part of the worm, the front half could as easily have been the posterior half, and *vice versa.* Hence it follows that the nature of the reaction of any given piece of the earthworm is a function of the direction of the impulse. If the impulse travels from the injured spot *anteriorly*, the piece of worm elongates; this initiates the normal progressive movements; if the impulse travels from the injured spot *posteriorly*, the piece of worm executes the characteristic jerking and squirming motions. In the first case the circular muscles are the first to contract, and this causes the elongation of the piece; in the second case the longitudinal muscles are the first to contract. This causes, of course, the jerking and winding motions. Why the impulses traveling forwards should always reach first the circular, and those traveling backwards first the longitudinal muscles, I have not yet been able to explain.

These reactions are not peculiar to this special mode of application of the stimulus, but are the consequence of locally applied stimuli. If an earthworm, for example, while crawling, be struck somewhat posterior to its middle point with the dull edge of a scalpel, the part of the worm in front of the offended spot continues to crawl, but at a more rapid gait; while the part beyond the offended spot at once begins to squirm, at the same time being dragged along by the anterior part. Or, again, if the entire normal worm be irritated at its anterior end, it is thrown into violent squirmings; while the same stimulus applied at the posterior end only hastens the normal progressive movements. *After a full study of the reactions of the earthworm against injury, including also heat and electricity, it was found that any stimulus having a definite local action would call forth the two characteristic sets of reactions.*

Are there other animals whose reactions to injury are as difficult to explain according to the pain-sensation theory as those of the earthworm? This question may be answered positively in the affirmative. Let us consider the reactions of animals chosen from four different groups. A live *starfish*, as every one knows, may be cut into pieces with a pair of scissors, or otherwise mutilated, without so much as calling forth a responsive reflex action other than a temporary withdrawal of the tube-feet. An arm thus cut off from the body and left free in a little water soon begins to move about in the same easy way that characterizes the motions of the whole animal.

The *leech* is perhaps the most highly differentiated of the worms, and it is here that we should expect definite expression of pain-sensations, if they are at all to be expected among such forms of life. Yet what do we actually find? The normal progressive movement of the leech in water is a rapid sinuous swimming motion, carried out by the entire animal, but most effectively by the flattened posterior portion. If now while the animal is gliding through the water it be cut in two in the middle with a sharp pair of scissors, both pieces without the slightest reaction to the injury (or at most the slightest momentary pause) continue to swim, with, however, a modified rate of speed, due to their modified length and shape.

The peculiar habit of *crabs*[1] of breaking off their thoracic appendages near the base, due to strong stimulation, either directly or indirectly, has been proved to be a purely reflex action, and may take place even after removal of the brain.

Among *insects*, the *honey-bee* has held an undisputed place for intelligence by all who put the lower animals in the list of

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[1] Fredericq, Léon, "Nouvelles Recherches sur L'Autotomie chez le Crabe," *Archives de Biologie*, XII, p. 169.

conscious beings. Bethe,[1] however, says: "I have cut away the entire abdomen from bees and have seen them live over an hour, during which time, from the moment of the operation on, if I placed them at some honey, they sucked at it unceasingly. Indeed, while a bee sat on my hand and sucked honey I have suddenly cut off its abdomen. It raised up for a moment and then quietly sucked on at the honey." I have cut off the abdomen, piece by piece, of *Libellula* without the animal's making any reactions whatever.

In conclusion it may be here remarked that of course the absence of reactions on the part of the lower animals to injury does not directly prove the absence of pain. It may, however, be as strongly asserted that the reactions of these animals to injury furnish no safe evidence that they are due to the presence of pain-sensations; and, further, if such reactions do indicate pain, then by the same criterion we must attribute pain sensations to pieces of an animal which likewise react to injury, — a view which to me seems entirely unbiological.

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[1] Bethe, Albrecht, "Vergleichende Untersuchungen über die Functionen des Centralnervensystems der Arthropoden," *Pflüger's Archiv*, Bd. lxviii, p. 509.

FOURTEENTH LECTURE.

NORTH AMERICAN RUMINANT-LIKE MAMMALS.

W. B. SCOTT.

In palaeontology, as in other lines of inquiry, the progress of discovery calls for frequent revision of opinion and changes of standpoint. In all kinds of investigation, the steps of which cannot be rigidly demonstrated, but depend upon the accumulation of evidence and the balancing of probabilities, it sometimes happens that obscure and difficult problems are suddenly lighted up by a new discovery, which gives a totally new and unexpected aspect to the subject. I have come here this evening to make public recantation of some of the errors which I lately upheld, having had my opinion completely changed by the force of new evidence, which has led to some very surprising results. This new evidence consists in the fossil mammals lately collected in the Uinta and White River formations, by Mr. Hatcher for the museum of Princeton University, and by Messrs. Wortman and Peterson for the American Museum of Natural History in New York.

The Uinta fauna is a most interesting transitional one, which is still very imperfectly known, but every new collection made of it increases our appreciation of its supreme importance, for by its aid one phyletic series after another is being completed. These series have a far-reaching significance for all departments of morphology, for they bring before us what we have every reason to regard as the actual steps of descent, and thus enable us to learn exactly what kinds of evolutionary changes do take place. The Uinta beds have a comparatively limited geographical extent, and have been found only in the basin south of the Uinta Mountains in northeastern Utah and northwestern Colorado. They overlie the strata of the upper Bridger stage (Washakie substage) and clearly precede the White River in time, though not separated from the latter by any great interval. Ordinarily the Uinta beds have been called Upper Eocene, but it would perhaps be better to refer them to the Lower Oligocene, for they are to be correlated with the Paris Gypsum, which the best French opinion now refers to the base of the Oligocene. It will be convenient to repeat here a part of the table of American fresh-water Tertiary formations given in a previous volume of these lectures.

Oligocene	John Day.
	White River.
	Uinta.
Eocene	Bridger.
	Wasatch.
	Torrejon.
	Puerco.

The correlation of the Uinta with the Paris Gypsum is made in spite of very marked differences in the mammals which are found in the two regions, for the differences are geographical rather than geological, and are doubtless to be explained by the existence of barriers which made migration between the two continents difficult, though not impossible. In the Wasatch the mammalian faunas of Europe and North America were remarkably similar and clearly indicate that the two continents were connected by land areas, which allowed the freest migration between one region and the other. In the Bridger there appear to have been some obstacles raised in the way of this free interchange of terrestrial mammals between the northern continents, though the correlative fauna of Europe is still too incompletely known to show the exact amount of difference. In the Uinta, however, the materials for comparison are abundant and prove that intermigration was opposed by such difficulties that only a few forms were able to overcome them. By White River times these obstacles, of whatever nature they may have been, were removed and once more the mammalian faunas of the two continents contained a large number of genera common to both. In the long interval between the Wasatch and the White River, North America had ample opportunity to develop a peculiar mammalian fauna, and one which was composed of types specially adapted to the conditions of life on this continent, and well fitted to resist the invasion of more or less similar forms from the other continents, when easy intercommunication was reëstablished.

Any fauna may, for our present purpose, be conveniently divided into two somewhat heterogeneous assemblies, one of which is composed of immigrants from other regions, and the other is indigenous. By the latter term are meant those forms which have long been established in a continent and whose ancestry may be traced through several geological horizons. Using this convenient mode of discrimination, the ruminants which from time to time have inhabited North America may be distinguished as either indigenous or immigrant types. The indigenous ruminants (it will be more accurate to call them *selenodonts*) predominated for a very long period of time and only toward the end of the Miocene did Old World types of selenodonts obtain a permanent foothold here. It is a remarkable fact that of these indigenous types which so long held sway in this continent, not one is left here at the present time, and, except the camels of the Old World and the llamas of South America, they have become altogether extinct and have left no descendants among recent mammals.

The indigenous selenodonts of North America have long puzzled the students who have attempted to work out their systematic position and their relationships to the selenodonts of other continents. Even the complete skeletons of various genera, recovered from time to time, seemed to give little help in solving the problem. The great obstacle to progress has been the absence of well-defined phylogenetic series, which would enable the observer to trace out the history of the various families and groups, step by step, through their numerous ramifications to final extinction, or to their modern representatives. An isolated genus, standing by itself, the predecessors and successors of which are unknown, offers almost insuperable difficulties to the determination of its proper taxonomic position. Such a genus always displays resemblances to a number of widely separated groups, not all of which resemblances can possibly be due to relationship, and many of which must have been independently acquired. It is easy to recognize this general principle, but it is by no means easy to apply it in a given case by estimating the taxonomic value of the various resemblances and differences, and to distinguish the characters which are due to real affinity from those which have resulted from a convergent or parallel course of development. Hence it can hardly be a matter for surprise that, even among competent observers, great differences of opinion should arise concerning the systematic position of isolated groups, one writer giving special emphasis to one set of characters, and another to another set. Only when the ancestry of the group in question has been made out does a satisfactory solution of the problem become possible.

The indigenous selenodonts of North America first began to be important in the Uinta formation, for in the strictest sense of the term the Bridger has as yet yielded none, though there are two or three genera which obviously represent the incipient stages of the group. Their culmination, so far as numbers, variety, and relative faunal importance are concerned, may be regarded as falling within the White River age, though they nearly held their own in the succeeding John Day. In the White River we find mingled with the indigenous selenodonts certain genera, like *Ancodus* and *Anthracotherium*, which had evidently migrated from the Old World, but did not secure a lasting foothold here, for no trace of them has been found in the John Day. In the Loup Fork the North American type of selenodonts underwent a very marked reduction, while migrants from Eurasia assumed a more and more important rôle, until, at the present, all our representatives of the group—deer, antelope, sheep, bison, etc.—are descendants of Old World ancestors, which, in some instances, reached North America at a very recent geological date. The only survivors of the American type, the camels and llamas, are no longer found in their original home, but are confined to two widely separated regions, the camels to the Old World and the llamas to South America. The explanation of this curious case of discontinuous distribution is that the group originated in North America, and after sending out migrants to the other regions, became extinct here.

The study of the peculiarly American selenodonts can most conveniently begin with those of White River times, because they are the best known, nearly every genus being represented in the collections by complete skeletons, and because they were then perhaps in the most characteristic stage of their development. As to the number of families represented in the formation, it is difficult to reach a conclusion, and almost as good reasons may be given for grouping them into seven families as into four, the arrangement here adopted. The position of only one of these groups has long been understood and very generally agreed upon, and this is the family represented in White River times by *Poebrotherium*, a genus which, there can be little doubt, is the ancestor of the modern Tylopoda, or, at the very least, of the llamas. The whole appearance of the skeleton, with its small tapering head, long neck, and elongate slender limbs and feet, is like that of a small llama, but of course it is very much more primitive and less specialized than the existing members of the group. The teeth are still undiminished in number; the canines are hardly larger than the incisors and only beginnings of diastemata are visible; the premolars are much elongated in the antero-posterior direction, and the molars are commencing to take on the prismatic or hypsodont shape. The skull is unmistakably tylopodan in character, with its triangular form and slender tapering muzzle, and while primitive features are retained, the enlarged tympanic bulla filled with cancellous bone has already been formed. The cervical vertebrae display the tylopodan peculiarity of a concealed vertebrarterial canal, perforating the neural arch, and only in the sixth vertebra does the canal occupy its normal position. The limbs and feet are already very elongate; the ulna and radius are coössified, and the fibula is completely reduced; the feet are didactyl, the lateral digits being reduced to mere nodules. The phalanges are slender and the unguals long and pointed, the shape of the latter showing that the hoofs were like those of the deer and antelope, and that the characteristic pad of the modern tylopodan foot was not developed till a later time.

While there has been no dispute that *Poebrotherium* is the White River representative of the main line of tylopodan descent, there has been no such agreement concerning the other selenodonts of that age, and this for the reason that nothing has been known of their ancestry, and that among existing mammals none could be selected as being a descendant of any of these genera; hence almost every conceivable opinion as to their systematic position has been held and defended. It is upon this very problem that the newly discovered Uinta forms shed such welcome light, and they render it exceedingly probable that *all the strictly indigenous North American selenodonts are branches of the great tylopodan stem.*

Paradoxical as this conclusion may appear to be, I believe that it is fully justified by the evidence, though it is, unfortunately, not possible to bring before you any adequate digest of that evidence, for it consists of a great mass of tediously minute dental and osteological comparisons of many genera and species. However, some of the more striking parts of the testimony may be made intelligible without an undue amount of detail. The Tylopoda are thus seen to be a very ancient and highly diversified group, comparable in these respects to the Pecora, or true ruminants, which in so many features they closely resemble, though the resemblances have, for the most part, been independently acquired in the two suborders. The Pecora are an Old World group, which underwent a great expansion and diversification in Eurasia, but did not reach this continent till late Miocene times, and they never attained the importance here that they have so long had in the eastern hemisphere. Even at the present time, when they have completely supplanted and driven out the Tylopoda from North America, they are far less numerous and varied here than in the Old World. Several of their American representatives, such as the bison, sheep, and musk-ox, are very recent immigrants, not occurring in beds older than the Pleistocene. In America the place of the Pecora was taken, to a very great extent, by the Tylopoda, which ran a course of development, in many respects, parallel to that followed by the Pecora and Tragulina—the latter a group which never reached America at all.

It is this very parallelism of the Tylopoda with the Pecora and Tragulina which has led astray so many students of the peculiar North American selenodonts, myself among the number. We have continually been endeavoring to detect relationships between these forms and the European ruminants and chevrotains, where no such relationships existed, but only analogies, parallelisms, or convergences. The truth appears to be that the indigenous American selenodonts make up a natural assemblage of forms which, with a remarkable degree of diversity in size and structure, are yet all quite closely related among themselves, but only distantly with the Old World types which more or less resemble them. The group formed by the association of these American families is, in fact, so diversified that a definition can hardly be framed for it; but that is, of course, no valid reason for refusing to recognize it as a natural group. Just as the Pecora are typically Old World both in origin and development, so the Tylopoda are typically North American, and did not reach the eastern hemisphere till the end of the Miocene or beginning of the Pliocene, and then in very limited numbers, *Camelus* and its immediate forerunners being the only known Eurasian representatives of the group.

The late Professor Rütimeyer of Basle, one of the greatest of palaeontologists, reached practically the same conclusion long ago, though no American agreed with him. It is an excellent example of his wonderful power of insight into a tangled problem of phylogeny that he should have discerned this fact, as I believe it to be, at a time when the fauna of the White River was but very imperfectly known and that of the Uinta had not yet been discovered. Dr. Wortman, in his latest paper, seems to have adopted Rütimeyer's views, though he does not explicitly say so, and for the same reasons that have led me to change my previous opinions, namely, the convincing power of the Uinta genera, all of which seem to be converging into a common term with the primeval member of the main tylopodan phylum.

It remains to bring forward the evidence upon which the conclusion here advocated is founded; and for that purpose we must first return to the White River selenodonts. One of the most largely represented families in that fauna is the *Leptomerycidae*, though the four genera which are associated in it — *Leptomeryx*, *Hypertragulus*, *Hypisodus*, and *Protoceras* — are so different from one another that much might be said in favor of referring each of them to a separate family. *Leptomeryx* was a very small animal, the skeleton of which is extraordinarily like that of a traguline, in which group most students of the subject have placed it; but the resemblance is almost certainly a deceptive one, and the real affinities are with the Tylopoda — a conclusion in which I am glad to find myself in complete accord with Dr. Wortman. Of the dentition only the upper incisors are unknown, and at least one of these is present, but the canine has been lost. The lower canine has become an incisor in form and function, while the first lower premolar, though minute, is caniniform, and its shape strongly suggests that in the immediate ancestors of the genus this tooth functioned as a canine. The other premolars are sharp and trenchant, and the molars, as Rütimeyer pointed out, are singularly cameloid in character, though with traguline features also. The skulls hitherto figured and described have all been broken across the very delicate and fragile muzzle, but newly collected specimens show that the skull has a very llama-like aspect and much more nearly resembles that of *Poebrotherium* than had been supposed. One important difference from the latter should, however, be noted; namely, that the auditory bulla is small and free from cancellous bone. The neck is short, and the cervical vertebrae have none of the tylopodan peculiarities. The fore-limb is much shorter than the hind, as in the tragulines, but the individual limb-bones are very like those of *Poebrotherium*, though the ulna and radius are separate. The forefoot has four digits, the lateral pair very much reduced; the trapezoid and magnum are coössified, as are the cuboid and navicular in the tarsus — both very exceptional features in the family and suborder. The hindfoot has a cannon-bone, of which the distal end is split in the characteristic tylopodan way. The hoofs are slender and pointed.

Hypertragulus is very much like *Leptomeryx*, and also much like *Poebrotherium*. It has kept the original shape and function of the canines, which are long and slender. The skull is thoroughly tylopodan in form and proportions and, except for the small and hollow tympanic bulla, greatly resembles that of *Poebrotherium*. The limbs and feet differ only in a few details from those of *Leptomeryx;* for example, the ulna and radius are coössified, but there is no cannon-bone in the hindfoot.

Hypisodus is the smallest member of the family and was a minute animal. It is remarkable as the most ancient American type with hypsodont molars growing from persistent pulps, and for having apparently ten lower incisors, the canine and first premolar having gone over to that series. So far as it is known, the skeleton is like that of *Leptomeryx*, though no cannon-bone is formed in the pes.

Protoceras, the largest genus of the group, equaling in stature the modern musk-deer (*Moschus*), is also the most curious — indeed, one of the most peculiar and bizarre looking of known mammals. The upper incisors have disappeared, but the upper canine, which in the female is small, is in the male a formidable tusk, opposed by the caniniform first lower premolar. The other teeth have resemblances partly to those of *Leptomeryx* and partly to those of *Poebrotherium*. The skull is extraordinary, especially in the male, in which sex there are horn-like protuberances on the parietals and great thickened plates arising from the upper edges of the maxillaries. In both sexes the nasals are extremely short and the narial opening exceedingly large, much as in the saiga antelope. The skeleton, limbs, and feet are those of an enlarged *Hypertragulus*. It may seem to involve a great strain upon credulity to refer *Protoceras* to the Tylopoda; yet its relationship to *Leptomeryx* and *Hypertragulus* is perfectly clear, and wherever the latter are placed, the former must accompany them. Wortman is of the same opinion, and in the paper so frequently cited he speaks of "the early Cameloids, *Protoceras* and *Leptomeryx*."[1]

One of the most characteristic of American families is the

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[1] "The Extinct Camelidae of North America," etc., *Bull. Amer. Mus. Nat. Hist.*, vol. x, p. 100.

long-lived and diversified group of the *Oreodontidae.* The type genus is *Oreodon,* by far the commonest of White River fossils, at which time great herds of *O. culbertsoni* roamed over the western plains. In this genus the dentition is closed, without diastemata; the upper canine is large, but the lower is incisiform, its place being taken by the caniniform first premolar. The other premolars are simple and trenchant, and the molars very like those of the deer. The skull has a rather short face and very long, narrow cranium; the orbits are completely encircled with bone, and a deep pit impresses the surface of the lachrymal. The neck is short, the trunk long, and the tail very long and heavy. The limbs and feet are short and thick; a rudimentary pollex is retained in the forefoot (this is the first artiodactyl in which that structure was demonstrated); and the hoofs are curiously rounded and flattened. In appearance and general proportions the skeleton of this genus recalls that of the modern peccary.

The taxonomic position of the oreodonts has been the subject of a great deal of discussion, and almost every possible opinion has been expressed; but the evidence of the Uinta fauna is very strongly in favor of the view held by Rütimeyer and Schlosser; namely, that they are aberrant members of the Tylopoda. In time they ranged from the Uinta through the Loup Fork, each successive horizon yielding peculiar genera; but with their later modifications we need not concern ourselves. Geographically they were entirely restricted to North America.

The most extraordinary and puzzling of White River mammals, not even excepting *Protoceras,* is *Agriochœrus.* Before complete skeletons of this creature had been collected, the skull and feet had been found separately, and were referred to no less than three distinct and widely separated mammalian orders. The dentition resembles that of *Oreodon* in the character of the canines, but the molars are much less distinctly selenodont and have a decided resemblance to those of the European genus *Ancodus.* The skull is oreodont except in a few details, such as the absence of the lachrymal pit, the incomplete closure of the orbit, etc. The neck is short, the backbone heavy, and the tail exceedingly long and stout, like that of the great cats. The limb-bones are so peculiar that they cannot be described in a few words; suffice it to say that they have a great many points of resemblance to those of the carnivorous groups, and the feet are provided with great claws, instead of hoofs, giving them a very sloth-like appearance.

If it seemed too great a demand upon our imagination to refer *Protoceras* to the tylopodans, it will appear obviously absurd to call *Agriochœrus* a camel; and yet that is the direction in which the evidence, as yet unfortunately incomplete, distinctly points. The Uinta forerunner of *Agriochœrus* (or what I regard as such) and that of *Oreodon* have drawn so close together as to indicate the origin of both of these families from some common Bridger ancestor.

The selenodont fauna of the Uinta, as a whole, is obviously ancestral to that of the White River, with the exception of certain forms, like the anthracotheres, which had immigrated from the Old World in the interval between the two epochs. But we may go farther than this, and may in several instances quite confidently point out the Uinta ancestor of a given White River genus. In other cases there is more uncertainty, because of less complete information, but even in these the Uinta has yielded forms which, if not directly ancestral, are yet very near to the ancestors sought for, and may be taken as representing them for all practical purposes of comparison and study.

Of these newly discovered Uinta genera one of the most interesting is the genus described by Wortman under the name of *Protylopus*, which is unmistakably the ancestor of *Poebrotherium*. It closely resembles the White River genus, but, as we should naturally expect, is smaller in size and more primitive in structure. For example, the dentition is closed and without diastemata; the premolars are not much extended antero-posteriorly; and the molars are very short-crowned. The canines, as in the descendant, are small—hardly larger than the incisors. The skull, while recalling that of *Poebrotherium* at the first glance, has a shorter muzzle, a widely open orbit, and a small, hollow tympanic bulla. The neck is, most unfortunately, not known. It would be of great interest to learn to what extent the peculiar specialization of the cervical vertebrae had proceeded. The vertebrae of the trunk and tail are like those of the White River genus on a small scale. The limbs are similar to those of the latter, but more primitive; they, and especially the feet, are less elongate; the ulna and radius are separate, except in old individuals; the fibula, though extremely slender, is still uninterrupted. In the manus are four functional digits, and in the pes two, with the lateral pair reduced to long filiform splints. *Protylopus* carries the main line of tylopodan descent one stage farther back than had previously been known, and, what is of even wider interest, it approximates this main line very distinctly to the other selenodont groups above described, and which we have already allotted to the tylopodan suborder.

Another genus recently named by Wortman—*Leptoreodon*—is very similar to *Protylopus*, but with most interesting and significant differences. It has diastemata in the dentition, large upper canines, and caniniform first lower premolar, as in the oreodonts and *Protoceras*, and also, though in rudimentary fashion, in *Leptomeryx;* the limbs are rather short and the feet tetradactyl. This genus nearly, but not quite, represents the meeting point of the main tylopodan phylum, the *Leptomerycidae*, and the oreodonts, and greatly diminishes the gaps which in White River times separated the three families. I regard *Leptoreodon* as the probable ancestor of *Protoceras*, and I do not know of any objection to such an arrangement, which would explain the oreodont characters of the descendant genus. These oreodont characters are so distinct that they led the late Professor Cope to the conclusion that *Protoceras* was merely an aberrant oreodont. While probable enough, the descent of *Protoceras* from *Leptoreodon* cannot yet be proven, for the gap between the two genera in structure and in time is still too wide. The former has been found only in the uppermost division of the White River stage (*Protoceras* beds), and we have as yet found no ancestors for it in the middle and lower divisions. Until these missing ancestors have been recovered, the relation of *Leptoreodon* and *Protoceras* must remain somewhat uncertain.

Camelomeryx is another genus which differs only in minor details from *Leptoreodon* and which seems to be either the ancestor of *Leptomeryx* or very near to that ancestor. In the dentition of the White River genus we found reason to think that it had been derived from some form in which the first lower premolar functioned as a canine, and this condition, with many others, is fulfilled in *Camelomeryx*, which is quite a small and delicately built animal.

The Uinta ancestor of *Hypertragulus* cannot yet be determined, because of the possible genera none are sufficiently well known. The genus *Leptotragulus* seems, at the present writing, to be the most likely candidate, but *Oromeryx*, or even *Bunomeryx*, may prove to be the chosen one.

The forerunner of the White River *Oreodon* has long been known and has received the suggestive name of *Protoreodon*, which expresses the ancestral relation. *Protoreodon* is, to all intents and purposes, an oreodont, but it has several most interesting resemblances to the agriochœres, such as the open orbits, the absence of the lachrymal pit, the elongate cranium, and the pattern of the lower molars. The upper molars have the fifth, or unpaired, cusp in the anterior half of the crown, as Schlosser predicted would be found — a prediction made before the present genus was known. The skeleton is not sufficiently different from that of its White River successor to require any description. This genus shows us that the oreodont family had become segregated as a distinct group in the Uinta, but the very many likenesses of *Leptoreodon* to *Protoreodon*, on the one hand, and to *Protylopus* on the other, afford the strongest confirmation to the opinion of Rütimeyer and Schlosser that the oreodonts are a branch of the Tylopoda.

What I believe to be the ancestor of *Agriochœrus* is a Uinta genus as yet undescribed, which I propose to name *Protagriochœrus*. The only known specimen of this most interesting form belongs to the American Museum of Natural History, and for the opportunity of making a study of it I am indebted to the kindness of Morris K. Jesup, Esq., President. It is, unfortunately, very fragmentary, and hence indecisive upon certain important points, but it is, nevertheless, exceedingly suggestive. The upper premolars are like those of *Protoreodon*, and, as in the latter genus, the upper molars have an unpaired cusp, but the most cursory glance at the pattern of these molars is sufficient to recognize their essential likeness to those of *Agriochœrus*. The tarsus is also strongly suggestive of the ancestral position here assigned to the genus, but it is provoking that the ungual phalanges are all missing. Whether or not the ancestral position rightfully belongs to *Protagriochœrus*, it is certain that this genus and *Protoreodon* bring the two families very close together and make it altogether probable that both groups lead back to a common ancestor in Bridger times.

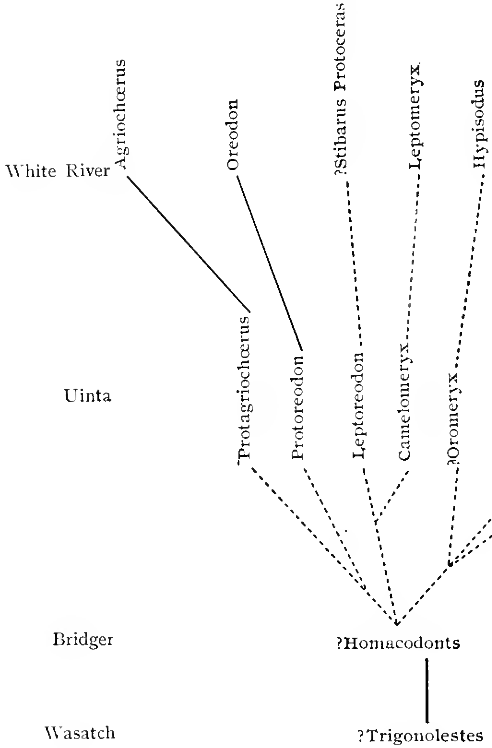
It may well prove to be the case that some of the relations between Uinta and White River genera here suggested will be shown by future discoveries to be erroneous. It matters little, however, whether we have chosen precisely the proper ancestors for the later forms; there still remains the highly important and significant fact that in this Uinta fauna all these different families are seen to be obviously converging back to some common term, and that they are much nearer together than they afterwards became in White River times. It is this fact which justifies us in maintaining the essential unity of all the indigenous American selenodonts, diversified and highly specialized as many of them eventually became. Having the great continent practically to themselves, they adopted many rôles, which naturally resulted in a greater or less likeness to the forms among the ancient Pecora and Tragulina which were playing similar parts in the eastern hemisphere. This explains the tantalizing and elusive likenesses to European genera, which have so long misled us, and shows why it was impossible to make any satisfactory arrangement of these American genera in European families or even suborders.

When we attempt to trace these various lines back of the Uinta, we find ourselves very much in the dark, because of our ignorance of the more ancient forms. The little Bridger genus *Homacodon* comes very near to filling the requirements of the common ancestor of all these groups, and it is exceedingly probable that the family of which it is the representative will eventually prove to be the fountain head whence these diverging streams were derived. *Homacodon* is of a very generalized type and seems to approximate the European genus *Dichobune*, which Schlosser regards as the remote ancestor of the Pecora. But the European genus is not known with sufficient completeness to make it plain whether it should be included in the same family as *Homacodon* or not. The latter is very primitive in structure, and has an unreduced dentition with relatively large canines, simple premolars, and sexituberculate upper molars which are just beginning to assume the selenodont pattern; the feet are probably pentadactyl. Nothing is known of *Homacodon* which can militate against the view that it represents a family whence were derived the various selenodont lines that have become distinctly segregated in the Uinta and widely diversified in the White River.

The probable ancestor of *Homacodon* is *Trigonolestes* (*Pantolestes*) of the Wasatch, a little creature which, having typically artiodactyl feet, possesses an extremely primitive type of dentition, so much so that, when only the teeth were known, the genus was supposed to be a lemuroid. In *Trigonolestes* we perhaps have the ancestor of all those selenodonts which I have described as indigenous to North America. It is of interest that one little character which persists throughout all the later genera of the group is already present in the Wasatch type; namely, the coössification of the meso- and ecto-cuneiforms in the tarsus.

The table on the following page will show conveniently the mutual relationships of the various selenodont genera as presented in what has been said.

If these results are well founded, we shall have to regard the Tylopoda as a highly important and eminently characteristic group in the history of mammalian life on this continent, a group which was very ancient, very peculiar, very long-lived, and greatly diversified, and one which in these respects may sustain a comparison with the Pecora. Admitting these facts, it becomes well-nigh impossible to define the suborder. As originally limited and defined by Flower, the demarcation is easy enough, because the definition is taken from the existing



members of the suborder, which are in many ways exceedingly peculiar. When, however, the extinct members of the group are taken into account, we find that every item of almost any definition that can be framed will be transgressed by one or other genus. This difficulty of definition is, of course, no objection to the classification proposed; on the contrary, it is the inevitable result of greater completeness in phylogenetic history. Definition is easy in exact proportion to the isolation of the group defined, but with a knowledge of the history and ancestry of a group its isolation disappears. As the boundaries between connected groups grow hazy, definition grows more and more difficult, until at last it becomes impossible.

Another necessary consequence of these conclusions is that the Tylopoda and Pecora are but very remotely connected and can have no common ancestor later than the middle or lower Eocene. Hence the many characters which these two suborders have in common must have been independently acquired by each. Startling as such a statement may seem to many, there is no escape from it, even though the position which has here been assigned to such genera as *Leptomeryx*, *Protoceras*, the oreodonts, etc., be an altogether mistaken one. We can now trace the history of the main tylopodan phylum, step by step, back to the Uinta epoch, and the Uinta representative of the series, *Protylopus*, has no pecoran features, save only the selenodont molars. As we may feel perfectly confident that *Protylopus* is not ancestral to the Pecora or any part of them, it follows that characters common to the two suborders, but not found in the Uinta genus, must have been separately developed. Rütimeyer long ago pointed out, from a comparative study of the recent forms, that the camels and llamas were but distantly connected with the true ruminants, and his masterly work on these questions is abundantly confirmed by the American fossils, which were very imperfectly or not at all known when he wrote.

The conclusions which we have reached suggest the importance of giving due weight to geographical considerations in dealing with phylogenetic and taxonomic problems. A little study and experience enable the observer to detect the foreign and migrant elements in a fauna, even though they firmly establish themselves in their new home and there give rise to new branches. When isolated genera are found, one of the most important questions which arises concerning them is, are they most like the types of this or of some other continent? This question rightly answered will serve as a most valuable clue in following out the history of the genus. Rütimeyer seems to have been guided largely by geographical considerations in the statement that I have mentioned as to the importance of the Tylopoda in North American life, and I am thoroughly convinced of the value of such criteria. Hitherto we have ignored them entirely too much.

I am perfectly conscious that I have been asking you to accept a great deal upon my *ex-cathedra* statements; but the difficulty lies, not in the absence of evidence, but in the impossibility of producing that evidence in court. It can be gained only by a minute study of the fossils themselves. Let us assume, however, that the conclusions so far reached are well established and consider some of the consequences which have an important bearing upon evolutionary philosophy in general.

(1) We see, in the first place, that parallelism and convergence of development are very real phenomena, and that they have played a highly important part in the course of evolution. Many morphologists now accept this mode of development unreservedly, but some still reject it altogether, or regard it as something unusual and exceptional. However that may be, the well-defined and established phyla of extinct mammals demonstrate the actuality of these modes of development beyond peradventure. Shuffle the cards as we may, we cannot arrange them so as to bring out any other result. One is sometimes tempted to believe that the number of possible tooth-patterns or osteological structures must be limited, so often are the same ones repeated in different phyla. In many instances we are able to follow out the history of a dental or a skeletal structure, step by step, from its point of origin to its final completion, and to show that the same structure has been independently attained over and over again. So far as single structures are concerned, this is now an old and familiar story; the spout-shaped odontoid process of the axis, which is so common among hoofed animals, the double bicipital groove on the humerus of horses, camels and giraffes, have often been pointed out as examples of this mode of development. Only of late, however, have we been in a position to prove that the entire structure may be so modified in two different groups as to keep parallel courses through long periods of time. One of the most striking instances of this is offered by the history of the cats. The family comprises two subfamilies, the *Felinæ*, or true cats, and the *Machairodontinæ*, or sabre-tooth cats; these two groups have been separate, at least since early Oligocene times, when the principal family characteristics had not yet appeared, and through the Miocene and Pliocene and into the Pleistocene they follow parallel courses, the final genus of one series, *Smilodon*, being almost identical with the terminal genus of the other, *i.e.*, *Felis*.

Another case of similar kind, and perhaps even more remarkable, has been brought to light by Ameghino. Until the junction of the two Americas, which was effected at the close of the Miocene, the southern continent was an extremely isolated region and had an altogether peculiar fauna, which differs from that of the northern hemisphere not merely in the genera and families of its mammals, but in their orders. So far as I am at present able to judge, the only mammalian order common to North and South America in the late Oligocene or early Miocene is the Rodentia. Among the peculiar Patagonian mammals is one which, upon a superficial examination, seems to be an undoubted horse. The aspect of the teeth, skull, trunk, and limb-bones is strikingly equine, and of the feet even more so ; the feet are functionally monodactyl, a large median toe bearing almost the entire weight, and two much reduced lateral toes forming dew-claws, quite as in *Protohippus* or *Hipparion*. Yet, when we come to make any careful examination of this skeleton, we soon learn that not only is it not a horse, but that it is not even a perissodactyl. It forms a most interesting and striking example of convergent development.

Between the Tylopoda and the Pecora we may observe another example of curiously complete parallelism. In this case we see how the former group firmly established themselves in North America at a time when communication with the Old World had, by some means, been rendered difficult, and how they ramified in many directions, taking here the rôles which in the eastern hemisphere were filled by the Pecora and Tragulina. In adapting themselves to these various parts they came to resemble the Eurasian groups in many important respects, but if we attempt to interpret these resemblances as having arisen from relationship and genetic affinity, we are at once landed in hopeless confusion. The conception of the indigenous American selenodont fauna as composing one diversified suborder removes these difficulties and marshals the families and genera in orderly array, but it involves such a degree of parallelism in development as may stagger the belief of those who have not made themselves familiar with the actual steps of descent in the mammalian phyla.

(2) A problem as to the modes in which evolution operates, concerning which there has been much discussion, is whether development is always by a series of direct and unswerving changes, each successive step in a given phylum coming, in every detail of structure, just so much nearer to the final result. Some years ago I had occasion to make a careful comparison of several successive genera in the equine phylum, and this study led me to the following conclusion. As a rule, development is remarkably unswerving and direct, in a large sense, yet in minor details a certain latitude is permitted, and in these evolution may pursue a more or less zigzag course, with many ups and downs. This conclusion is considerably strengthened by what we have learned concerning the main line of descent in the tylopodan phylum. Comparing the more ancient genera of this series with its modern representatives, we are at once struck by the remarkable difference in their canine teeth. In *Protylopus* and *Poebrotherium* the canines are very small and may almost be called incisiform, but in *Gomphotherium* of the John Day the canines begin to enlarge, and from that time onward these teeth become larger and larger, until the formidable lacerating apparatus of the modern type is attained. In the probable ancestors of the Uinta camel, *Homacodon* and *Trigonolestes*, the canines are relatively long and pointed, but even though we should exclude those genera from the series, the analogy of all the ungulate groups would justify the assumption that the ancestors of *Protylopus* had canines which were of fairly large size and formed effective weapons. No one can imagine that in the Uinta genus these teeth are in their primitive condition. We are forced to infer, then, that the canines first dwindled to very small proportions, only to enlarge again and become formidable.

Another instance of much the same kind is afforded by the history of the premolars. In *Protylopus* these teeth resemble in general form those of the other contemporary selenodont genera and of the White River *Leptomeryx*, but they are distinguished by an incipient elongation in the antero-posterior direction. In *Poebrotherium* the premolars are greatly elongate, carrying much farther the process which was begun in the

Uinta type. This elongation of the teeth accompanies the extension of the muzzle and, as it were, prevents the formation of diastemata, though these appear in the later species, *P. labiatum*, in which the growth of the rostrum outstrips that of the premolars. In the John Day the tendency is changed, and the premolars of *Gomphotherium* revert almost to the Uinta type in form, while *Procamelus* and the subsequent genera of the phylum are remarkable for the reduction of their premolars both in size and number.

Wortman has called attention to a third example of this fluctuation in the tylopodan phylum, affecting the form of the tympanic bulla. In *Protylopus* the bulla is small and hollow, but in *Poebrotherium* it has become greatly inflated and filled with cancellous bone, the inflation especially affecting the medial portion of the bulla. In *Gomphotherium*, once more, the direction of development is changed and the outer portion of the bulla begins to enlarge at the expense of the inner, a change which reaches its culmination in the existing genera.

So general are these minor fluctuations, that it would be difficult to point out a single genus which in every minute detail is exactly fitted to be the ancestor of a later genus, assuming that these fluctuations do not occur. Just how great they may be in degree we have at present no means of determining. It seems, *a priori*, improbable that, after a structure has been lost or reduced to a rudimentary condition, it can ever be regained, or become functional once more, and yet certain cases do suggest that even such regeneration may occasionally take place. At all events, it would be premature to deny the possibility of changes of this character.

The features of alternating, up and down, or zigzag development, to which attention has been called, are, after all, of a very trifling nature. When we survey the successive and closely connected genera of a long and crowded phylum, we cannot fail to be impressed with the steady, orderly, unswerving advance in all important structural features. This advance is not always, perhaps not even usually, uniform in all parts of the structure. One part may be accelerated and another retarded, and what was retarded in one genus may be accelerated in a succeeding one. Thus, in the reduction of digits the hindfoot is more rapidly modernized than the forefoot. In *Protylopus*, for example, the hand has still four functional digits, while the pes already has but two ; in the succeeding *Poebrotherium* both manus and pes are in the same stage of simplification, with two functional digits, and two small nodules representing the rudiments of the lateral pair. But while one structure may thus be retarded in its development and another accelerated, the differentiation of the organism, as a whole, keeps steadily advancing. It almost seems as if the animals were consciously striving for a goal, though, of course, this is only an impression given by the direct and unswerving course their evolution takes.

There are many other "morals" which might be drawn from the history of the American selenodonts, but I shall lay no further tax upon your patience. If it seems to you that I am attributing too much importance to palaeontology and ignoring other means of investigation, this is simply because to praise morphology and physiology in Wood's Holl would be carrying coals to Newcastle. The fair structure of our science must be reared upon a broad and deep foundation, not of a single department of inquiry, but of all of them combined and "fitly joined together."

FIFTEENTH LECTURE.

CASPAR FRIEDRICH WOLFF AND THE *THEORIA GENERATIONIS*.

WILLIAM MORTON WHEELER.

> Mag 's die Welt zur Seite weisen,
> Edle Schüler werden 's preisen,
> Die an deinem Sinn entbrannt,
> Wenn die Vielen dich verkannt.
>
> GOETHE, *Morphologie*. p. 256.

THE universe which we apprehend—reducible in last analysis to various sequences and coexistences in time and space—seems to have a twofold aspect to the contemplative mind. The minds of some men are vividly affected by the *succession* of phenomena, the ceaseless current of events, the changes that alter the complexion of the world, the great qualitative and quantitative differences produced by these changes in that which we call matter. These observers may note the rhythm that is forever recurring in nature, the alternate repetition of day and night, the return of the seasons, the cyclical recurrence of stages in the development of living organisms—in short, the regular emergence from time to time of typical forms and conditions from the flowing current of events. This rhythm and repetition does not, however, produce the same deep impression on these observers as the successive and multiform changes themselves.

The other class of observer, although he may note the onrushing current of events, is more vividly impressed with the similarity of the forms and conditions that recur from time to time and from place to place. The attention is fixed on these recurring objects and conditions, and gradually builds them into general concepts that ultimately acquire a stability which nothing can shake. The movement of the stream of phenomena takes a subordinate position in consciousness, and the mental activities attach themselves by preference to stable, island-like forms and principles.

Thinkers from the earliest times to the present day seem to be referable to one or the other of these two classes. The differentiation begins in early Greek philosophy with men like Heraclitus and Parmenides. To Heraclitus the world was an unceasing flux—*παντα ῥει, οὐδεν μενει, all things are flowing, nothing is standing still.* All things are forever *becoming*, nothing ever *is*. Parmenides, who fixed the trend of the Eleatic school, belonged to the other class. He is the philosopher of rest. The chaotic, multiform world of Heraclitus, forever in motion, becomes for him merely a world of nonexistent appearances, a shifting phantasmagoria, and only being *is*—the *absolute*—the *one*, forever at rest.

The contrast in these two views reappears between Aristotle and Plato. This difference is seen in the all-pervading movement as conceived by Aristotle in his Physics, in contrast with the "ideas" of Plato. Movement to Aristotle is "something very analogous to our modern biological conception of transformation in development, for he analyzes 'movement' as every change, as every realization of what is possible."[1] Plato, on the other hand, under the influence of Parmenides and the philosophy of rest, emphasizes the forms and qualities that keep recurring to our minds in time and space, generalizing them into his "ideas" and endowing them with all the attributes of reality.[2] He would say, *e.g.*, of a living animal as it stands before us: "This animal as we see it does not exist in reality, but is only an apparition, a continual becoming, a relative existence, which can as well be called nonexistent as existent. The idea alone actually exists which is represented in this animal, or the animal itself (*αὐτο το θηριον*). This idea is independent of everything; it exists by itself; it has not become; it does not decay, but exists always in the same

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[1] Osborn, H. F. From the Greeks to Darwin. New York, Macmillan & Co., 1894. p. 50.

[2] See Pater, Walter. Plato and Platonism, Chaps. I, II.

manner (*ἀει ὀν, και μηδεποτε οὐτε γιγνομενον, οὐτε ἀπολλυμενον*). If we can recognize the *idea* in this animal, it is immaterial and unimportant whether we are looking at the animal now before us or its ancestor that lived a thousand years ago, or whether the animal is here or in a distant land, or whether it appears in this or that manner, position or action, whether, finally, it be this or another individual of the same species: all this is unessential and appertains only to appearances: the *idea* of the animal alone really *is*, and really is an object of the understanding."[1]

It would not be difficult to trace the Heraclitean conception of the flux through Aristotle down to such modern philosophers as Hegel and Herbert Spencer, and to trace the Platonic idea, through the *λογος σπερματικος* of the Stoics, the *forma substantialis* and the *causae primordiales* of the scholastics, to Kant's Ding-an-sich, Schelling's Absolute, and the Platonic idea as adopted by Schopenhauer. But the tracing of these conceptions in detail would lead us far afield in metaphysics. I should beg your indulgence for mentioning these matters did they not seem to me to be, in some measure, necessary to a proper understanding of the two great views of embryonic development that have been and still are held by thinking students of nature —*preformation* and *epigenesis*.

The development of the living organism is the most striking special case of development we know. The development of what appears to be a simple egg, within a comparatively short time, and beneath our very eyes, into a complex living animal, is development *par excellence* — the very perfection of that development which is more dimly apprehended in the much slower growth of worlds, of human societies and human institutions. Hence we do not wonder that the development of the individual organism has become one of the main tests of two alternative views which, with a more general application, have from the earliest times vexed philosophic thinkers.

Under the influence of the Christian church the Platonic conception seems to have led to the notion of the special crea-

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[1] Schopenhauer, A. Die Welt als Wille und Vorstellung. Leipzig, Brockhaus, 1888. Bd. i, p. 203.

tion of fixed types or forms. It culminated in that finished theory of predelineation in embryonic development known as *emboîtement*.[1] This was, in reality, the very negation of all development, since the theory held that all the individuals of a species had been created simultaneously for all time.[2] In the forcible language of the last century, Eve's ovary contained the compressed and diminutive germs of all coming human beings incapsulated one within the other. Such a theory could arise only from overestimation of the definitive form attained through development, and an underestimation of the changes undergone by the egg during its development. The typical adult form usurped the theorist's attention, and the elaborate process whereby the type was gradually realized shrunk to a mere unshelling and subsequent growth in size of the next individual in order in the incapsulated series.

For the theory of *emboîtement* the creation not only of every species, but of every individual organism on our planet, by a single preadamite fiat, was a necessary postulate. The rival theory, epigenesis, implied in the cosmology of Heraclitus and easily traceable to Aristotle, starts with a simple form of unorganized matter, which through the agency of certain forces undergoes the complicated changes that finally result in the adult living organism. The homogeneous becomes the heterogeneous. The creation of new organisms is no longer conceived as having taken place once for all in a remote and inscrutable past, but as taking place everywhere and at all times. An exaggeration of epigenesis is spontaneous genera-

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[1] Passages which show the close genetic relationship of Neo-Platonic and Christian thought on the subject of creation are not infrequent in the writings of the Church Fathers. The following quotations from Augustine clearly express the idea of *emboîtement*: "Sicut autem in ipso grano invisibiliter erant omnia simul, quæ per tempora in arborem surgerent, ita ipse mundus cogitandus est, cum Deus simul omnia creavit, habuisse simul omnia, quæ in illo et cum illo facta sunt, quando factus est dies: non solum coelum cum sole et luna et sideribus . . . sed etiam illa quæ aqua et terra produxit, potentialiter atque causaliter priusquam per temporum moras ita exorentur, quomodo nobis jam nota sunt in eis operibus, quæ Deus usque nunc operatur." De Genesi ad lit., v, 45. "Omnium quippe rerum quæ corporaliter visibiliterque nascuntur, occulta quædam semina in istis corporis mundi hujus elementis latent." De Trinitate, iii, 8.

[2] "Qui igitur systemata praedelineationis tradunt, generationem non explicant, sed, eam non dari, affirmant." C. F. Wolff, Theoria Generationis, 1759, p. 5.

tion. Aristotle even believed that mud could become earthworms and earthworms become eels.[1]

Before the end of the past century these two views of development which I have attempted to trace back respectively to Aristotle and Plato had assumed definite and contrasting forms. Bonnet, Haller, and Leibnitz, following a Platonizing tendency in dealing with natural phenomena, had elaborated and accepted the theory of *emboîtement*, or "evolution," as the word was then understood. Bonnet's contributions to this view have been adequately presented by Professor Whitman, in his lectures to the members of the Marine Biological Laboratory during the summer of 1894.[2] Haller, justly styled by his contemporaries an "abyss of learning," though devoted to *emboîtement*, had too great a store of mental riches to give himself up year after year, like Bonnet, to exhaustive rumination on a single theory. The opinion of Leibnitz on *emboîtement* is not so generally known, and may be considered briefly. The philosopher of a preëstablished harmony could hardly overlook a theory like that of predelineation. Like many philosophers of the present day, Leibnitz was glad to accept the theories of contemporary scientists, weave them into his general scheme, and, without adding anything really new, again present them to the public, heavier with the weight of his name and authority. In his "Monadologie," he says[3]: "Philosophers have had much difficulty in dealing with the origin of forms, entelechies, and souls. Of late, however, careful investigations on plants, insects and animals, have led to the conclusion that in nature organic bodies never arise from chaos or decomposing matter, but always from germs (semences), in which, *without a doubt*, they are already preformed. Hence we may conclude that in this Anlage not only do organic bodies exist before generation, but that there is a soul in these bodies, in a word, the indi-

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[1] Aristoteles. *'Ιστοριαι περι ζωων.* Ed. Aubert u. Wimmer. Leipzig, 1868. ii, 6. 16. pp. 56 and 58. — J. Bona Meyer. Aristoteles Thierkunde. Berlin, 1855. pp. 97, 98.

[2] Whitman, C. O. (1) "Bonnet's Theory of Evolution a System of Negations." (2) "The Palingenesis and the Germ Doctrine of Bonnet," *Biological Lectures* (1894). Boston, Ginn & Co., 1895.

[3] Leibnitz, Op. Phil., p. 711.

vidual itself, and that reproduction is merely a means of enabling this individual to undergo a greater change in form, to become an individual of a different kind. Something similar to generation is seen when maggots become flies and caterpillars butterflies." At another place, in the "Theodicee," he says,[1] after referring to the microscopic observations of Leeuwenhoek: "Thus I would contend, that the souls, which are some day to become human souls, were already present in the germ like the souls of other species, that they have always existed in our fore-fathers as far back as Adam, *i.e.*, since the beginning of things, in the form of organized bodies." These remarks of Leibnitz are the *ne plus ultra* formulation of the theory of *emboîtement* — its extension to embrace not only the physical but also the psychical and spiritual aspect of living things.

It is, perhaps, easy to understand how philosophical and religious preconceptions could give this final form to the theory of *emboîtement*. Other considerations, however, of a more real and scientific character seem to have led men's minds in the same directions. The microscope, invented in the sixteenth and bequeathed to the seventeenth century, had profoundly influenced speculation. Magnification had revealed, as if by magic, the existence of a great world of structures undreamed of by the greatest intellects the race had hitherto produced. The authority of the ancients weakened perceptibly, for little value could thenceforth be attached to their opinions on the nature of the great world that stretched out beyond the confines of unaided vision. The mind, full of the great microscopic discoveries of the time, was carried away by its own inertia, and, outrunning the instrument, first dreamed of and then believed in the existence of structures too minute to be revealed by the available lenses. This speculation was, perhaps, justifiable, except when it undertook to define the precise nature of what was at that time an ultra-microscopic region. It was natural but erroneous to conceive unseen structures as diminutive duplicates of the seen. The verisimilitude of this error increased when it became apparent that the microscope was unable to resolve perfectly transparent structures even of

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[1] Op. Phil., p. 527.

considerable size. And here the theorist triumphed over the empirical observer, for he could assert, what was not easily disproved, that owing to their transparency the microscope must ever fail to reveal the germs incased one within the other.

The Siegfried destined to overcome this monstrous theory of *emboîtement*, a theory not only false in itself, but one jealously guarding the problem of development, and preventing all access to it, as the dragon guarded the treasure of the Niebelungen, was *Caspar Friedrich Wolff*. Wolff, one of the many great intellects that northern Germany has produced, was born in Berlin in 1733. You will find nearly all that is known of his life in a letter by his amanuensis Mursinna to Goethe, published by the great German poet in his *Morphologie*.[1] The scant facts of this letter, with Wolff's own writings, in which his personality is studiously kept in reserve after the manner of scientific men, leaves us with a sense of uncertainty not entirely free from sadness. We long to know more of this sweet-natured student who, at the early age of six-and-twenty, was an intellectual giant, defending an epoch-making thesis, the *theoria generationis*, simply *pro gradu doctoris medicinae*.

Before giving a brief account of this *Theoria* it may be well to try to form some idea of its author's general mental characteristics. Wolff was a disciple of Aristotle. The training of the schoolman is only too apparent in all his scientific writings, apart from Mursinna's statement[2] to the effect that when Wolff was lecturing on medicine in Berlin "he taught logic probably better than it had ever been taught before, and applied it in particular to medicine, thereby creating, so to speak, a new spirit in his hearers, so that they were enabled to understand and assimilate his other teachings more easily." His skill in deductive logic seems to have been noticed by Sachs,[3] who claims that some of Wolff's observations on plant structure "are highly inexact, and influenced by preconceived opinions, and his account of them is rendered obscure and often quite

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[1] Goethe. Morphologie, 1820, pp. 252–256.

[2] Goethe. Morphologie, p. 254.

[3] Sachs. History of Botany, p. 251.

intolerable by his eagerness to give an immediate philosophic explanation of objects which he had only imperfectly examined." The same statement may be extended to many of his zoölogical observations, but this is far from convincing us that his method of investigation was at fault. Wolff's method, which did not differ from that of the scientist of to-day, was, if anything, more admirable than his observations. The very fact that he was full of his Aristotelean hypothesis of epigenesis places him head and shoulders above the investigators both of his day and of to-day, who naïvely believe that they are starting their investigations on a solid foundation of facts divorced from all theory. Even Sachs admits that Wolff's phytotomical work, though poor from the standpoint of observation, was the most important that appeared in the period between Grew (1682) and Mirbel (1802), "because its author was able to make some use of what he saw, and to found a theory upon it."[1]

Apart from this preconceived hypothesis of epigenesis it is surprising with what perfect *naïveté* Wolff approaches the phenomena to be observed. Armed with his microscope, which it does not require a Sachs to tell us "was of insufficient power and its definition imperfect," he entered what was practically an unknown domain, peopled only with the figments of the predelineationists. The fascination of the growing plant and developing embryo soon possessed him and never afterwards left him. During his long life he returned again and again to the study of the chick. Those who teach embryology year after year cannot fail to appreciate Wolff's power and greatness when they observe the superficial impression left on nine-tenths of the students who study the developing chick in the well-equipped laboratories of to-day.

Wolff's instruments, poor as they were, enabled him, nevertheless, to traverse a considerable and very significant portion of the region that lies beyond the boundary of our unaided. vision. What he saw there at once convinced him that *emboîtement* was a myth. We should expect so young a man as Wolff was when he wrote the *Theoria* to do two things — to repeat his main thesis *ad nauseam*, and to be rather unsparing

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[1] Sachs. History of Botany, p. 251.

of his opponents. He did neither. His main contention is clear enough, although not often expressly stated. He rarely refers directly to the theory of predelineation and when he does there is no sting in his refutation.

Stripped of many details that are somewhat wearisome to the modern reader, the result of Wolff's observations may be expressed in his own words taken from the very middle of the *Theoria.*[1] I translate: "In general we cannot say that what cannot be perceived by the senses does not therefore exist. This principle is more facetious than true when applied to these observations. The particles which constitute all animal organs in their earliest inception are little globules, which may always be distinguished under a microscope of moderate magnification. How, then, can it be maintained that a body is invisible because it is too small, when the *parts* of which it is composed are easily distinguishable?" If we of to-day read in the place of "globules" the word "cells," which are what Wolff actually saw and distinguished in both plants and animals, we shall have no difficulty in understanding how his observations disproved *emboîtement,* at least for any one who would take the trouble to repeat them. Wolff had looked further than the adult form and had found not a series of similar, incapsulated embryos, but a single embryo made up of a vast number of minute particles, the cells, closely resembling one another, but placed side by side. There was no expanding of a preëxisting organism till it entered the field of vision, but a host of minute and always visible elements that assimilated food, grew and multiplied, and thus gradually in associated masses produced the stem, leaves, stamens — in short, every organ of the plant. This he shows in the first part of the *Theoria.* In the second part, carrying on the same method, he shows how in the animal body the heart, blood vessels, limbs, alimentary canal, kidneys, etc., arise in a similar manner.

The third part of the work is devoted to theoretical considerations. Wolff conceives living things to be constructed like a plant, to consist of a main stem, or trunk with roots and branches. In the embryo of the bird the umbilical duct cor-

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[1] Theoria Generationis, p. 72.

responds to the stem of the plant; the blood vessels of the vascular area that bring the nutriment from the yolk to the embryo are the roots; the organs and appendages of the embryo correspond to the branches of the plant.[1] The organism starts out on its development with a stem which is to connect it with the source of nutrition on the one hand and its branches on the other. All the substance of the embryo is originally unorganized, inorganic. Organization sets in at one point in the stem and thence gradually spreads to the tips of the branches. A branch is first formed as a little bud of unorganized substance, then the sap (in plants) or the blood (in animals) flows into it from the adjacent organized part; thus it becomes organized, and the process continues till the organism has acquired its definitive size and development. The blood or sap is propelled into the unorganized substance, consisting of globules, by a peculiar force — Wolff's *vis essentialis*, which is defined in the opening chapter of the *Theoria*, and was made the special subject of Wolff's last work, written thirty years later.[2] Organization of the unorganized substance is the combined result of this *vis essentialis* and a property which Wolff calls *solidescibilitas*, a tendency to solidify, shown most clearly in the formation of the walls of plant cells. The *vis essentialis* propels liquid nutriment into the dense unorganized matter already present. The paths along which it flows become the cavities of the blood vessels or plant vessels not before existent. The liquid nutriment solidifies to form more unorganized substance, by intussusception with that already present, and the part grows. Wolff explains the origin of the kidney which he discovered in the chick — the Wolffian body, or mesonephros, as we now call it — in a similar manner. Here it is the urine that is impelled by the *vis essentialis* into a mass of preëxist-

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[1] Wolff (Theorie von der Generation, 1764) compares four-footed animals to pinnatifid leaves and "the bat is a perfect leaf — a startling statement, but, as I have shown, the analogy is not chimerical, for the *mode of origin of the two is the same*." Quoted by Huxley ("The Cell Theory," *Brit. and Foreign Medico-Chir. Review*, vol. xii, 1853).

[2] Wolff, C. F. Von der eigenthümlichen und wesentlichen Kraft der vegetabilischen sowohl als auch der animalischen Substanz. St. Petersburg, 1789.

ing, unorganized substance, and the paths along which it flows become the lumina of the uriniferous tubules and the ureter.

We know that Wolff's main error lay in grossly underestimating the complexity of the problem he attempted to solve. This has always been a great pitfall in attempting an explanation of life. Perhaps it is well that it is so, for Wolff would hardly have had the heart to attempt it if he could have seen the problem with our eyes. And may not we, too, daily commit the same blunder when we lend a willing ear to those who regard living protoplasm as nothing more than a "complex chemical compound"?

Wolff accepted a simple substance as the basis of life because he was unable to detect structure in the embryo beyond a certain limit which happened to coincide with the limits of magnification of his lenses. We should suppose that Wolff would have longed for a better lens and have at least suspected the possible existence of some kind of structure beyond that which he could detect. Instead of doing this, however, he writes the following remarkable sentences which will draw a smile from the modern searchers after centrosomes[1]: "No one has ever yet, with the aid of a stronger lens, detected parts, which he could not perceive by means of a weaker magnification. These parts either have not been seen at all, or they have appeared of sufficient size. That parts may remain concealed on account of their infinitely small size and then gradually emerge, is a fable." There it is in cold Latin! Was Wolff merely nodding when he wrote this, or was he trying to hoodwink the predelineationists into believing that he had seen everything that was worth seeing in the embryo?

In the closing paragraph of his great work on the development of the intestinal tract, a work which appeared in 1768, some nine years after the *Theoria*, Wolff seems to rise to a clearer perception of the complexity of the problem. He appears to be far more doubtful concerning the way in which simple matter becomes organized. Referring to the development of the anterior body wall, he says[2]: "This is one of the

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[1] Theoria, Sect. 166.

[2] Wolff, C. F. Ueber die Bildung des Darmkanals im bebrüteten Hühnchen. Uebersetzt von J. F. Meckel. Halle, 1812, p. 245.

most important proofs of epigenesis. We may conclude from it that the organs of the body have not always existed, but have been formed successively: no matter how this formation has been brought about. I do not say that it has been brought about by a combination of particles, by a kind of fermentation, through mechanical causes, through the activity of the soul, but only that it has been brought about."

Remaining within the province of observation which he staked out for himself, and pursuing his excellent method, Wolff was not only able to undermine the theoretical edifice of the predelineationists, but also to lay the foundations for future structures of great promise. Thus all conscientious investigation with good methods leads to subordinate facts of value besides the main line of facts accumulated in support of the theory in hand. Wolff was a biologist in the true sense of the word. He regarded plant and animal life as but slightly different aspects of a single set of phenomena. It can be shown that he anticipated to some extent the modern theories of protoplasm and the cell.[1] According to Sachs "it was Wolff's doctrine of the formation of cellular structure in plants which was in the main adopted by Mirbel at the beginning of the present century," and "the opposition which it encountered contributed essentially to the further advance of phytotomy." [2]

The theory of the metamorphosis of plants, usually attributed to Goethe, was clearly expressed by Wolff. In fact, Wolff seems to have had clearer notions on the subject than Goethe, according to Schleiden's statement.

To embryology Wolff made many valuable contributions, not the least of which was his description of the formation of the intestinal tract of the chick. This work was styled by Carl Ernst von Baer "die grösste Meisterarbeit, die wir aus dem Felde der beobachtenden Naturwissenschaften kennen." It was published in Latin in the twelfth and thirteenth volumes of the *St. Petersburg Commentaries*, where it lay buried and forgotten till it was unearthed and translated into German by

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[1] Cf. Huxley.

[2] Sachs. Hist. of Botany, p. 250. For the relations of Wolff's views to those of Schleiden and Schwarm, see Huxley, The Cell Theory.

the younger Meckel in 1812 and used for the purpose of refuting some of Oken's erroneous views on the development of the alimentary tract.

In general it may be said that the effect of Wolff's work on his contemporaries was anything but immediate.[1] There are writers who even doubt the truth of the oft-repeated statement that Wolff refuted the theory of predelineation. Sachs, *e.g.*, speaking of Wolff's *Theoria*, says that the "weight of his arguments was not great" and that "the hybridization in plants which was discovered at about the same time by Koelreuter supplied much more convincing proof against every form of evolution."[2] We cannot lay much stress on this statement, which seems to imply, what some physiologists seem never to tire of implying, that evidence derived from experiment is *eo ipso* more convincing than evidence derived from observation. It is certain that the predelineationists had considered the case of hybrids, for did not the ever-watchful Bonnet endeavor to explain the origin of the mule on the assumption of *emboîtement?* And why should Koelreuter's plant hybrids have more value in refuting *emboîtement* than that commonest of all hybrids, the mule? If Sachs wishes to imply that at the present day we should regard the evidence from hybrids as a complete and satisfactory refutation of the theory of *emboîtement*, we may assent; but this is not tantamount to saying that in the latter half of the eighteenth century it was Koelreuter and not Wolff who refuted the theory of evolution. Perhaps it would be better to leave this question of the relative merits of Wolff and Koelreuter to the student who has the time and the opportunity to study all the relevant literature of the closing decades of the eighteenth century.

Wolff's position in the history of thought on the subject of organic development becomes somewhat clearer when we compare him with Darwin, for whose coming he helped to prepare

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[1] "Though every reader of the Theoria Generationis must see that Wolff triumphantly establishes his position, yet, seventy years afterwards, we find even Cuvier (Histoire des Sciences Naturelles) still accrediting the doctrine of his opponents." — Huxley, The Cell Theory.

[2] Sachs. History of Botany, p. 405.

men's minds. Wolff's *Theoria* was published in 1759; Darwin's *Origin of Species* in 1859. Wolff had been preceded by Harvey in much the same way as Darwin was preceded by Lamarck. Both Wolff and Darwin were ideal investigators, patterns for all time. Darwin's love of truth, his perfect fairness and modesty withal, seem to have been Wolff's possession also. This is shown in a letter to Haller,[1] thanking the great champion of *emboîtement* for his kindly notice of the *Theoria* in his "*Elementa*": "I thank you for wishing me well, for loving me, sublime man, although you have never seen me, and know me and my character only from my letters. May God reward you for this, since I can never hope in all my life to attain to such distinction, that I may show you worthy acknowledgment of your goodness, if you will not receive in lieu of it my everlasting veneration of your intellect. And as to the matter of contention between us, I think thus: For me, no more than for you, glorious man, is truth of the very greatest concern. Whether it chance that organic bodies emerge from an invisible into a visible condition, or form themselves out of the air, there is no reason why I should wish that the one were truer thân the other, or wish the one and not the other. And this is your view, also, glorious man. We are investigating for truth only; *we seek that which is true.* Why, then, should I contend with you? Why should I withstand you, when you are pressing towards the same goal as myself? I would rather confide my epigenesis to your protection, for you to defend and elaborate, if it is true; but if it is false, it shall be a detestable monster to me also. I will admire evolution, if it is true, and worship the adorable Author of Nature as a divinity past human comprehension; but if it is false, you, too, even if I remain silent, will cast it from you without hesitation."

Both Wolff and Darwin devoted their lives to the investigation of the same great• problem — the development of life on

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[1] Epistolae ad Hallerum, October, 1766. Quoted from Alf. Kirchhoff, "Caspar Friedrich Wolff. Sein Leben und seine Bedeutung für die Lehre von der organischen Entwickelung," *Jen. Zeit. f. Med. u. Naturwiss.*, Bd. iv, Heft 1, 1868, pp. 193–220. This valuable essay has been of great assistance in the preparation of my lecture.

our planet. Both found answers to their respective parts of this problem. Wolff published his answer when he was very young; Darwin waited till he was well along in years. Each was confronted by a formidable, clearly formulated theory of special creation. The theory that confronted Wolff was special creation of all *individual* organisms by a preadamite fiat. Darwin was confronted by a theory of the special creation of all the *species* at the same inscrutable time. This view had found favor with such eminent men as Linné, Cuvier, L. Agassiz, Owen, and the numerous systematists who followed in their footsteps, making species Platonic ideas, just as individual organisms had been made Platonic ideas in Wolff's day. During the closing half of the eighteenth century it became clear to thinking men that individual organisms always have an epigenetic origin from preëxisting individuals. The closing half of the present century has been consumed in demonstrating that species always arise from preëxisting species.

Both Wolff and Darwin collided with prevailing theological views. Darwin's experience in this matter is well known, and perhaps the less said about it the better. It is not so generally known that Wolff's failure to establish himself as a professor in Germany, and his departure in 1769 for St. Petersburg, where he spent the remainder of his life, was probably due not only to professional jealousy, but also to a certain antagonism on the part of religious contemporaries. In his letters Haller often warned Wolff of the dangers of his views to religious dogma, and endeavored to persuade him to abandon them "on grounds of utility."[1] Just before leaving for St. Petersburg Wolff wrote the following to Haller: "There is, of course, no reason why a divine being should not exist, even if organic bodies are formed by natural forces and through natural causes; for these very forces and causes, yes, Nature herself, has as much need of an Originator as organic bodies; still the evidence would be far more cogent and apparent, if we should find from contemplation of Nature that her individual products, the organic bodies, required a Creator, and that nothing organic could be produced through natural causes." Does not this

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[1] Kirchhoff, A., *l.c.*

remind us of the following passage in the last chapter of Darwin's *Descent of Man?* "I am aware that the conclusions arrived at in this work will be denounced by some as highly irreligious; but he who denounces them is bound to shew why it is more irreligious to explain the origin of man as a distinct species by descent from some lower form, through the laws of variation and natural selection, than to explain the birth of the individual through the laws of ordinary reproduction. The birth both of the species and of the individual are equally parts of that grand sequence of events, which our minds refuse to accept as the result of blind chance. The understanding revolts at such a conclusion, whether or not we are able to believe that every slight variation of structure, — the union of each pair in marriage, — the dissemination of each seed, — and other such events, have all been ordained for some special purpose."

Both Wolff and Darwin left their theories unfinished. They maintained a transformation of simpler into more complex matter, but they did not succeed in demonstrating how this transformation is accomplished. I have already quoted Wolff's confession of ignorance of the way in which epigenetic development is brought about. The doubts entertained by Darwin and his successors concerning the adequacy of natural selection as a complete explanation of descent are familiar to us all. The absolute completeness of the old *emboîtement* and special creation hypotheses doomed them to a speedy death. Wolff's and Darwin's hypotheses have lived because they represented only parts of a great truth. On this account, also, they have supplied and will continue to supply powerful incentives to investigation.

Both the theory of epigenesis and the modern theory of descent are manifestly imbued with the old Heraclitean and Aristotelean conception of heterogeneity arising from homogeneity in a continual flux of events. We have come to regard this as the essence of evolution. We still repeat Herbert Spencer's definition[1]: "Evolution is an integration of

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[1] Spencer, Herbert. First Principles. New York, Appleton & Co., 1886. p. 396.

matter and a concomitant dissipation of motion; during which the matter passes from an indefinite, incoherent homogeneity to a definite, coherent heterogeneity; and during which the retained motion undergoes a parallel transformation." If Wolff could have read this sonorous definition he would probably have accepted it as the expression of a general truth, as it is accepted to-day. Still when we compare our views on the development of living organisms with those of Wolff, we observe a vast difference, to which Professor Whitman calls attention when he says[1]: "The indubitable fact on which we now build is no bit of inorganic homogeneity, into which organization is to be sprung by a coagulating principle, or cooked in by a *calidum innatum*, or wrought out by a spinning archaeus, but the *ready-formed, living germ, with an organization cut directly from a preëxisting, parental organization of the same kind.*

"The essential thing here is, not simply continuity of germ-substance of the same chemico-physical constitution, but *actual identity of germ-organization with stirp-organization.*"

To-day we recognize three conditions of matter: dead matter, undifferentiated living protoplasm, and differentiated living protoplasm. The germ is cut from its parent as organized but undifferentiated living protoplasm; during ontogeny it is converted into differentiated living protoplasm. Wolff saw no difficulty in leaping with a bound from dead matter to highly differentiated living protoplasm. There are scientific acrobats still living who are not at all afraid of taking a like hop, skip, and jump. The majority of biologists, however, are too heavy with the past century's observations on living matter to be able to leap so fast and so far. They find the distance between dead matter and undifferentiated protoplasm enormous — a chasm so wide and deep that they prefer making a long detour to attempting a perilous leap across it. They are more intrepid in passing over the gap, great as it undoubtedly is, which separates the undifferentiated from the differentiated phase of organized matter, the germ from the adult. Current discussion is, therefore, mainly limited to the valuation of the

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[1] Whitman, C. O., "Evolution and Epigenesis," *Biological Lectures* (1894). Boston, Ginn & Co., 1895. p. 212.

extent of complexity in the germ as compared with the complexity of the fully developed organism.

He who finds little difficulty in passing from the simple to the complex, from the homogeneous to the heterogeneous, will take an epigenetic view of development. The physiologist, who deals with processes, who is ever mindful of the Heraclitean flux, inclines naturally to this view. On the other hand, he who readily idealizes and schematizes, whose mind is endowed with a certain artistic keenness, an appetite for forms and structures, and a tendency to make these forms final patterns, eternal molds, more permanent than the substance that is poured into them — such a one will find more difficulty in understanding *how* the homogeneous can become the heterogeneous. Of this type is the modern morphologist who is continually diagrammatizing, who has his eye fixed on complex static structures and conceives the continually changing form of the developing egg as a series of kinematograph pictures in three dimensions of space. He is as much inclined to Platonize as is the modern physiologist to reason along lines suggested by Aristotle. He is by nature a preformationist.

Just as Wolff's followers have split into two schools — one believing in little, the other in much preformation in the germ — so Darwin's followers have split into two schools, the Neo-Lamarckians and the Neo-Darwinians, in obedience to the two psychological tendencies to which I have called your attention. The Neo-Darwinians, in laying great stress on the segregation, stable and complex *intrinsic* structure of the germ plasma and its importance as a vehicle of hereditary characters, and in attributing less value to the *extrinsic* factors, like food and environment, are allying themselves with theorists of the type of Parmenides and Plato. On the other hand, the Neo-Lamarckians who believe in the permanent change-producing effects of the *extrinsic* factors (environment, etc.) on structure, and attribute less value to the architecture of an *intrinsic* vehicle of heredity (germ plasma), range themselves with Heraclitus and Aristotle.

The amount of differentiation displayed during the ontogeny of an organism or during its phylogenetic history will be differently estimated by different workers, till we are in possession of some means of mathematically measuring differentiation and variation. The demand for mathematical measurement is already being made in certain quarters, and this demand progressing science will undoubtedly supply. At present we are quite adrift in our discussions, so long as we ignore the more general and philosophical aspect of the question and thereby overestimate its simplicity. Even if we accept differentiation and the interaction of differentiated products as the root ideas of the evolution of the individual and of the race, we are still at a loss to understand how the initial differentiation arose — how the homogeneous first became the heterogeneous. Lloyd Morgan has recently expressed this hopeless and baffling search for initial differentiation as follows[1]: —

> "Assuming, with the nebular hypothesis, a primitive fire-mist, we must assume also an environment from which it is already differentiated and to which its heat energy is communicated by radiation. Or if we accept the meteoric hypothesis, we must grant the existence of already differentiated cosmic dust and the interaction of its constituent meteors. If we give yet freer rein to the speculative tendency, which, chastened or running riot, is man's blessing or curse, and, straining our mental vision, search deeper still into the beginnings of our universe, to find in the homogeneous substance that Sir William Crookes calls *protyle*, the stuff from which the chemical elements were differentiated; even in this dim and wholly hypothetical region we are forced to assume, as the antecedent conditions of differentiation, transformations and redistributions of energy, implying a prior differentiation to render such interaction conceivable. Or if, once more, we conceive the elemental atoms as vortex rings, differentiated from the ether and thenceforth interacting, even here at the very threshold of differentiation, we seek for an answer to the question: Under what physical conditions did such vortex motion originate?"

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[1] Morgan, Lloyd, "The Philosophy of Evolution," *Monist*, July, 1898, p. 489. Weismann, too, expresses this difficulty in his "Germinal Selection": "Die sog. 'epigenetische' Theorie mit *gleichen* Keimeseinheiten ist deshalb eigentlich nichts Anderes, als eine Evolutionstheorie mit unbewusster Zurückverlegung der Anlagen in die Moleküle und Atome, eine, wie mir scheint, unstatthafte Vorstellung. Eine *wirkliche* Epigenese aus völlig *gleichartigen*, nicht bloss aus untereinander *gleichen* Einheiten ist nicht denkbar."

The pronounced "epigenesist" of to-day who postulates little or no predetermination in the germ must gird himself to perform Herculean labors in explaining how the complex heterogeneity of the adult organism can arise from chemical enzymes,[1] while the pronounced "preformationist" of to-day is bound to elucidate the elaborate morphological structure which he insists must be present in the germ. Both tendencies will find their correctives in investigation.

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[1] See, *e.g.*, Loeb, J., "Assimilation and Heredity," *Monist*, July, 1898, p. 555.

SIXTEENTH LECTURE.

ANIMAL BEHAVIOR.

C. O. WHITMAN.

"*Natura non facit saltum*, is applicable to instincts as well as to corporeal structure." — DARWIN, *Origin of Species*, p. 231.

CONTENTS.

ANIMAL behavior, long an attractive theme with students of natural history, has in recent times become the centre of interest to investigators in the field of pyschogenesis. The study of habits, instincts, and intelligence in the lower animals was not for a long time considered to have any fundamental relation to the study of man's mental development. Biologists were left to cultivate the field alone, and psychologists only recently discovered how vast and essential were the interests to which their science could lay claim.

The contribution which I have to offer aims at no extensive exposition of the subject, but rather to call attention to some phenomena which I have observed, and to connect therewith such interpretations and theoretical considerations as may come within the sphere of general biology.

In animal life there are many interesting modes of keeping quiet, which are instinctive and adapted to special purposes. It is a very general means of concealment and escape from enemies. For illustration, we may take first the leeches, animals relatively low in the scale. One of the lower and least active forms, occurring everywhere in ponds, lakes, and streams, is known under the generic name *Clepsine*. There are many species, varying from one-quarter inch to one inch or more in length. They are found on their regular hosts, turtles, frogs, fishes, molluscs, etc., or on the under side of stones, boards, branches, or other submerged objects near the shore. One of the larger species, found often in large numbers on turtles, will be favorable for observation.

BEHAVIOR OF CLEPSINE.

a. *Deceptive Quiet.*

Place the animal in a shallow, flat-bottomed dish and leave it for a few hours or a day, in order to give it time to get accustomed to the place and come to rest on the bottom. Then, taking the utmost care not to jar the dish or breathe upon the surface of the water, look at the *Clepsine* through a low magnifying lens and see what happens when the surface of the water is touched with the point of a needle held vertically above the animal's back. If the experiment is properly carried out, it will be seen that the respiratory undulations (if such movements happen to be going on) suddenly cease and that the animal *slightly* expands its body and hugs the glass. Wait a few moments until the animal, recovering its normal composure, again resumes its respiratory movements. Then let the needle descend through the water until the point rests on the bottom of the dish at a little distance from the edge of the body. Again the movements will cease and the animal will hug the glass with itş body somewhat expanded. Now push the needle slowly along towards the leech, and notice, as the needle comes almost in contact with the thin margin of the body, that the part nearest the needle begins to retreat slowly before it. This behavior shows a surprising keenness of tactile sensibility, the least touch of the water with a needle-point being felt at once. This delicate sensitiveness is manifested in such a quiet way that it would be generally overlooked, and an observer unfamiliar with the habits of *Clepsine*, and not realizing the necessity of extreme quiet in his own movements, would almost certainly draw false conclusions. If the dish were moved or the water carelessly disturbed in any way, the *Clepsine* would assume its motionless attitude and appear to be wholly indifferent to the disturbance. If its back were rubbed with a brush or the handle of a dissecting needle, in order to test its sensitiveness to touch, the appearance would probably still be that of insensibility and indifference to the treatment. Closer examination, however, would show that the flesh of the animal was more rigid than usual, and that the surface was covered with numerous small, stiff, conical elevations, the dermal papillæ or warts, which are so low and blunt in the normal state of rest as to be scarcely visible. It would be seen that the animal, although motionless, was in a state of active resistance to attack. Every muscle would be strained; the whole skin would be tense and rough with the stiff, pointed papillæ; and at the same time the body would be found excessively slippery and difficult to lay hold of, owing to the mucous secretion poured forth from the dermal glands. To guard still further against dislodgment, the body would be flattened out as much as possible and tightly applied to the glass. The activity of the resistance offered by this passive-looking creature would be very forcibly realized if the observer attempted to circumvent it by slipping a thin blade or spatula beneath it with a view to forcing its hold. If overcome in one part it would stick by another, and skillful manipulation would be necessary to get both ends free at the same time. With one end detached, the other will often hold against a pull strong enough to snap the body in two.

b. *Rolling into a Ball.*

Clepsine has another and entirely different method of keeping quiet. The behavior bears striking analogy to that which has been described as "feigning death" in some insects. The animal rolls itself up (head first and ventral side innermost) into a hard ball, outwardly passive, and free to roll or fall whithersoever gravity and currents in the water may direct it. The ball will bear considerable pressure and rough handling without unfolding or exhibiting any marked movements. Left in quiet for a few seconds, the animal slowly unrolls itself and creeps off. This instinct has many advantages for a slow-moving creature like *Clepsine*, as will presently be seen.

1. *Provoked by Exposure.* — The ball-like attitude is assumed under various circumstances. If a stone or board with *Clepsine* attached to its under surface be quietly turned upside down, thus bringing the leech from shade or darkness into light and exposure, it may sometimes maintain its position of rest unchanged, only hugging the stone a little more closely and not moving until all is quiet. More generally, however, it rolls itself up, and by the time the stone is turned, or before, it falls to the bottom, where it can unfold and escape without danger of discovery. If, by chance, the animal has eggs, it will not desert them to escape in this way. As soon as the eggs hatch and the young become attached to the ventral side of the parent, the latter may roll itself up with its brood inside, fall to the bottom as before, and thus escape with all its progeny.

This species, then, has two quite distinct and peculiar ways of keeping quiet and thus avoiding its enemies. If the animal has no eggs, or if it has young, it may adopt either mode of escape, while if it has eggs it has no choice but to remain quiet over them. In the species here considered, the eggs are held together in a thin, gelatinous sheet, secreted at the time of ovipositing, and of a size and form to be entirely covered by the expanded body of the parent. In some species of *Clepsine* the eggs are laid in thin membrane-like sacs, which are fastened to the under side of the parent, and in this case the rolling up into the form of a ball is the safest course of behavior and the one generally adopted.

2. *Forced by Attack.* — The same behavior will almost invariably follow when any species of *Clepsine* is closely pursued and finds itself unable to fix itself by either end, as when a spatula is repeatedly thrust under it in such a way as to break its hold and defeat its efforts to regain a footing.

3. *Induced by Gorging Blood.* — The provocations to such behavior thus far considered have all been such as might, and probably do, cause more or less alarm. It is important to note, however, that the instinct may manifest itself frequently under conditions that seem to exclude the influence of fear. I have often seen these leeches fold themselves into balls at the end of a good meal, and so roll to the edge of the shell of their host, and fall to the bottom. This mode of concluding a quiet repast, with no assignable cause for alarm, and with every reason for satisfaction and contentment, except for the desire to get out of light into darkness, under cover of a stone or some other object, will hardly pass as feigning; and cataplexy and the tropisms are equally out of question. We could not assume, for example, that *Clepsine* is positively heliotropic when hungry, and negatively heliotropic after feeding. *Shade is preferred at all times in both conditions.* If hungry, *Clepsine* leaves the shade, not because it prefers light, but because it prefers its host more than it prefers shade. If the host is not found it will again return to a shady retreat, if one is to be found, however hungry it may be. The rolling up cannot be attributed to light, as the animal takes the extended position when at rest, even if compelled to remain in the light. What, then, shall we conclude?

4. *Origin and Utility.* — Observation and inference may be stated as follows:

1. The act of rolling up into a passive ball may be performed (*a*) *under compulsion*, as when it is the last resort in self-defence; (*b*) *under a milder provocation*, as *one* of three courses of behavior, as when the resting place is turned up to light, and the choice is offered between remaining quiet in place, creeping away at leisure, or rolling into a ball and dropping to

the bottom; (*e*) or, finally, *under no special external stimulus*, but rather *from internal motive*, the normal demand for rest and shady seclusion, presumably very strong in *Clepsine* after gorging itself with the blood of its turtle host.

2. This mode of taking leave of the turtle, after a full meal, is the easiest, the quickest, and the safest way available. To drop off fully extended, as *Clepsine* sometimes does, would retard descent and increase the chances of capture by fish. To creep about on the back of the host, waiting for an opportunity to grasp a plant or stone, would be decidedly hazardous, for if it came within the stretch of its host's neck, annihilation would be almost certain, while if lucky enough to keep out of reach of its host, it would still be in danger of the same fate from other turtles.

3. This behavior is instinctive, since it is performed by the young after the first meal as perfectly as by the adult.

4. Looking more closely at the nature and origin of this instinct, it will be seen to be quite a natural performance, in keeping with the most fundamental features of the animal's organization, and only a special application of a more general act that is primary and organic as much as tasting, seeing, or sleeping.

The more general act consists simply in tucking or rolling the head under, as often happens when the animal is resting. The habit may be observed in the young as soon as they are sufficiently developed to be capable of bending the tip of the head under. The same act, carried a little further, gives the half-rolled condition, in which only the anterior of the animal is folded, while the posterior portion remains unrolled and attached by the sucker. This attitude is often assumed if the leech is sick or has been injured. It is only a step further to release the sucker and fold it over the part already rolled up, thus completing the part ball to a whole ball, which can move passively more rapidly and safely than is possible by active creeping. From beginning to end we have only one act, in different stages of completion, simply different degrees of one and the same process.

5. Having the general act to start with, it is easy to see

how it might be made of use for particular purposes; in other words, how special adaptations of a useful kind might arise. If the act is a natural concomitant of the resting condition, and is associated with a feeling of ease and security, we see how sickness, injury, fear, a heavy meal, etc., might prompt it, and in higher or lower degree, according to the nature and intensity of the inciting cause. Full and prompt action under exposure, pressure, injury, and in the event of a good meal, would carry decisive advantages, so that individuals reacting in the more favorable degrees would stand the best chance of escape and survival. Natural selection would steadily improve upon the results, and the special adaptation, in different stages of development in different species, as we find it to-day in different *Clepsines*, would lie in the direct line of progress. This view does not of course presuppose intelligence as a guiding factor, and therefore lends no support to the theory of instinct as "lapsed intelligence," or "inherited habit."

6. An instinct of the kind here considered does not depend for its development upon effort and the transmission of functionally acquired improvements in organization, but upon *the natural selection of the best qualified germs*, for that is what the survival of the fittest individuals always means. Many species of *Clepsine* require but one full meal a year, and as they seldom live more than two or three years, the number of meals is very limited. There is little room, then, for repeating the experiment often enough to affect the organization. Indeed, such a supposition would here appear to be absurd in the last degree. On the other hand, the selection of the fittest germs, provided for in the survival of the best-adapted individuals, would inevitably advance the species along the line leading to the special instinct.

If the view here taken be correct, the instinct of rolling into a ball is not a matter of deliberation at all, but merely the action of an organization more or less nicely adjusted to special conditions and stimuli. Intelligence does not precede and direct, but the indifferent organic foundation with its general activities is primary; the special behavior or instinct is built up by slowly modifying the organic basis.

c. *Sensitiveness to Light.*

The question as to how much intelligence, if any, *Clepsine* may have, I do not here undertake to settle or discuss. That the animal is endowed with keen sensibilities is evident from the behavior before described. The following simple experiment affords a striking demonstration: Pass the hand over a dish in which a number of *Clepsines* are resting quietly on the bottom, at a distance of a few inches above the animals, taking care not to make the least jar or other disturbance. If the animals are quite hungry, the slight shadow of the hand, imperceptible though it be to our eyes, will be instantly recognized by them, and a lively scene will follow, every leech rising up, supported on its posterior sucker, and swinging at full length back and forth, from side to side, round and round, as if intensely eager to reach something. Put a turtle in the dish and see what a scramble there will be for a bloody feast. The shadow of the hand was to these creatures like the shadow of a turtle swimming or floating over them in their natural haunts, and hence their quick and characteristic response. A piece of board floating over them would have the same effect. Although so sensitive to a small difference in light, the *Clepsine* eyes can give no pictures, and hence there is no power of visual discrimination between objects. They probably recognize their right host by the aid of organs of taste; at any rate they are often able to distinguish their host from closely allied species.

INSTINCT OF ROLLING INTO A BALL AMONG INSECTS.

The following examples of the instinct of rolling into a ball among insects are from Kirby and Spence.[1]

"I possess a diminutive rove-beetle (*Aleochara complicans* K. Ms.), to which my attention was attracted as a very minute, shining, round, black pebble. This successful imitation was produced by folding its head under its breast, and turning up its abdomen over its elytra, so that the most piercing and discriminating eye would never have discovered it to be an insect. I

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[1] Entomology, pp. 411, 412.

have observed that a carrion beetle (*Silpha thoracica*) when alarmed has recourse to a similar manœuvre. Its orange-colored thorax, the rest of the body being black, renders it particularly conspicuous. To obviate this inconvenience, it turns it head and tail inwards till they are parallel with the trunk and abdomen, and gives its thorax a vertical direction, when it resembles a rough stone. The species of another genus of beetles (*Agathidium*) will also bend both head and thorax under the elytra, and so assume the appearance of shining, globular pebbles.

"Related to the defensive attitude of the two last-mentioned insects, and precisely the same with that of the Armadillo (*Dasypus*) amongst quadrupeds, is that of one of the species of wood-louse (*Armadillo vulgaris*). The insect, when alarmed, rolls itself up into a little ball. In this attitude its legs and the underside of the body, which are soft, are entirely covered and defended by the hard crust that forms the upper surface of the animal. These balls are perfectly spherical, black, and shining, and belted with narrow white bands, so as to resemble beautiful beads; and could they be preserved in this form and strung, would make very ornamental necklaces and bracelets. At least so thought Swammerdam's maid, who, finding a number of these insects thus rolled up in her master's garden, mistaking them for beads, employed herself in stringing them on a thread; when, to her great surprise, the poor animals beginning to move and struggle for their liberty, crying out and running away in the utmost alarm, she threw down her prize. The golden wasp tribe also (*Chrysididæ*), all of which I suspect to be parasitic insects, roll themselves up, as I have often observed, into a little ball when alarmed, and can thus secure themselves — the upper surface of the body being remarkably hard, and impenetrable to their weapons — from the stings of those *Hymenoptera* whose nests they enter with the view of depositing their eggs in their offspring. Latreille noticed this attitude in *Parnopes carnea*, which, he tells us, *Bembex rostrata* pursues, though it attacks no other similar insect, with great fury; and, seizing it with its feet, attempts to dispatch it with its sting, from which it thus secures itself. M. Lepelletier de Saint-

Fargeau, to whom entomology is indebted for so many new facts relative to the manners of hymenopterous insects, has given us a striking account of a contest between the art of one of these parasites (*Hedychrum regium*) and the courage of one of the mason-bees in endeavoring to defend its nest from its attack. The mason-bee had partly finished one of her cells, and flown away to collect a store of pollen and honey. During her absence the female parasitic *Hedychrum*, after having examined this cell by entering it head foremost, came out again, and walking backwards, had begun to introduce the posterior part of her body into it, preparatory to depositing an egg, when the mason-bee arriving laden with her pollen-paste threw herself upon her enemy, which, availing herself of the means of defence above adverted to, rolled herself up into a compact ball, with nothing but the wings exposed, and equally invulnerable to the stings or the mandibles of her assailant. In one point, however, our little defender of her domicile saw that her insidious foe was accessible; and, accordingly, with her mandibles cut off her four wings, and let her fall to the ground, and then entering her cell with a sort of inquietude, deposited her store of food, and flew to the fields for a fresh supply; but scarcely was she gone before the *Hedychrum*, unrolling herself, and, faithful to her instinct and her object, though deprived of her wings, crept up the wall directly to the cell from whence she had been precipitated, and quietly placed her egg in it *against the side* below the level of the pollen-paste, so as to prevent the mason-bee from seeing it on her return."

BEHAVIOR OF NECTURUS.

a. *Refusal of Food from Fear.*

Our large fresh-water salamander, popularly called mud-puppy, water-dog, hellbender, etc., is another animal that may be profitably studied with reference to its modes of quiet. The first adults which I kept in captivity in a large aquarium refused to eat pieces of raw beef or small fish, whether dead or alive. For months they went on, seemingly entirely indifferent to any proffered food, not paying the least attention, so far as I noticed, to tempting morsels dropped quietly in front of them or held in suspension before them. Living earthworms and insect larvæ were presented to them, all of which were known to be palatable to the creature in its natural habitat; but nothing availed to draw attention or elicit any evidence of hunger. Quiet and wholly indifferent in outward behavior, yet the animals were actually starving and wasting away. Were the creatures *feigning* quiet and indifference? Or was the behavior merely the expression of timidity, the animal not having the courage to perform the acts necessary to secure the food which it must have craved? I confess that I did not for a long time understand the cause of this refusal of food.

Further acquaintance with the adults, supplemented by an experience of two seasons in rearing the young, opened my eyes to the extreme timidity of these animals, which is so deep-seated and persistent that one can form only a poor idea of it without considerable actual contact with it. The outward behavior is very quiet and mild and gives little indication of fear. The animal will often submit to gentle handling without making any violent effort to escape. In short, the behavior is misleading, and one stands no chance of understanding it until he learns to keep quiet himself while observing, and discovers how to get into confidential relations with the creature. This can be done with the adults, but to better advantage with the young.

b. *Behavior of Young in Taking Food.*

The eggs may be readily hatched in a shallow dish and young thus obtained which have never learned anything from the parents. I had about fifty young hatched in this way towards the end of July. When first hatched they were loaded with food-yolk sufficient to meet their needs for about two months. By the end of September I began to get intimations of a desire for food. The method of feeding was as follows: The dish containing the young was kept on a table, where, without being moved, food could be offered in perfect quiet. I used the tiniest bits of raw beef and offered only one piece at a time, which I held in small forceps or on the point of a needle a little in front of the animal to be tested. If the meat is held closely enough to touch the head, the animal is frightened and may retreat with such haste as to alarm all its companions. If the bait is held a little to one side, an inch or so away, and very quietly for a minute or more, a slight turning of the head in that direction may be noticed, in case the animal is ready to eat and feels confidence enough to try to reach it. The turning of the head is done very cautiously and almost as slowly as the minute-hand of a clock moves, so that one may become aware of it, not by seeing the movement, but by noticing the inclination of the head to the axis of the body. If there be a decided turn of the head of this kind, the case is hopeful, as it shows an interest which may be encouraged to action by bringing the bait a little nearer, but very slowly and without any jerky movement. Halting about half an inch away, wait for further movement on the part of the animal, if you are fortunate enough not to have frightened it away. If the animal's courage holds out — in most cases it does not in the first trials — it will soon begin to move, but with a slowness that tries the observer's patience. The head at length comes up to a point a quarter of an inch away, more or less, and after making sure of the position of the bait, which seems to be done less by the aid of the eyes than by the sense of touch, the animal tries to seize it by a quick side movement of the head and a snap of the jaws. *The first attempt to take the bait corresponds in all essential points with the behavior of the adult when trying to capture a fish, a worm, or an insect larva, although the aim may not be quite so sure.*

If one is successful in getting one or more to feed, the more timid ones may be brought forward in the same way by patiently alluring them from day to day, until they are tempted to an effort. Once made, the effort becomes easier at the next trial, and in the course of a month or six weeks the bolder ones will respond fairly promptly. A few of my specimens became very familiar with me, and would come towards me when I approached the dish, looking up at me wistfully, as if knowing well the meaning of my visits.

c. *Influence of Innate Timidity.*

In the behavior above described, we see an instinctive mode of capturing prey held in check, and probably directed to some extent, by innate timidity. Fear seems to be the main factor in control at the start, holding the animal in a trance-like quiet, undecided as to what to do, waiting for confidence to attack, or for a stronger motive to flee. As fear subsides a little, the preparatory movement of attack begins, but the sly behavior is due to fear rather than the slyness of stratagem. The slow and cautious method of approach is certainly not all finesse, for the deportment bears still the stamp of hesitating timidity, and this part of the act may become much freer as the animals become tamer and more fearless. The final part of the act, that of snapping the bait, was always performed in the same characteristic way. The piece of meat seemed always to be regarded as a *living* prey, which was to be seized quickly, held firmly for a moment or two, and then swallowed. Unfortunately I did not experiment to see what could be done in modifying this part of the act.

Instinctive fear is evidently a very important element in the conduct of the lower as well as the higher animals. In *Necturus* we see how it may be just as effective as intelligence in securing a sly mode of attack. So strong is its influence that I doubt whether there is any finesse in the movement. The adaptation of acts to purposeful ends must not be accepted too quickly as proof of intelligence in the doer. Such acts are common enough in plants, and there we are under the necessity of finding some other explanation.

d. *Organization Shapes Behavior.*

Necturus appears to understand well the act of capturing its prey, and the nice adaptation of each act to the end in view naturally enough suggests forethought and refined experience.

But we see the performance executed by the young, which have never had any experience of that kind, nor any opportunity to copy from others. We cannot therefore suppose that they perform these acts understandingly. The young *Necturus*, hatched in a dish where it has never met any living thing except its companions, has nothing to guide its first effort to capture food except its organization and its simple experience in walking and swimming, which acts are again like those of the adult, not because directed by intelligence or example, but because they are performed with the same organs under similar conditions. The young has the same sensory and motor apparatus as the adult, but it has never before known the feeling of hunger, it has never experienced pain from contact with an enemy, it has never learned that a prey may escape if not approached slyly. Its movements in approaching and snatching a piece of meat, as if it were a living object, are, then, those characteristic of the species, not because they are measured and adapted to a definite end by intelligent experience, but because they are organically determined; in other words, depend essentially upon a specific organization.

The timidity of young hatched in a dish is the same as that of specimens hatched in the lake, and therefore it cannot be charged to individual experience or to parental influence. It, too, inheres in the special brand of organization, and has nothing to do with memory of pain sensations.

e. *Origin and Meaning of Behavior.*

We have taken a very important step in our study when we have ascertained that behavior, which at first sight appeared to owe its purposive character to intelligence, cannot possibly be so explained, but must depend largely, at least, upon the mechanism of organization. The origin and meaning of the behavior antedate all individual acquisitions and form part of the problem of the origin and history of the organization itself. It is the first and indispensable step, without which it would be impossible to reach sound views, either as regards the particular behavior or the difficult question of the relation of instinct to habit and intelligence. If the problem is not simplified, its nature is better defined and its perspective is relieved of many a myth that might otherwise obscure our vision. We see at once that the behavior does not stand for a simple and primary adaptation of a preëxisting mechanism to a special need. As the necessity for food did not arise for the first time in *Necturus*, the organization adapted to securing it must be traced back to foundations evolved long in advance of the species. The retrospect stretches back to the origin of the vertebrate phylum, and, indeed, to the very beginning of genealogical lines in protozoan forms. The point of special emphasis here is that instincts are evolved, not improvised, and that their genealogy may be as complex and far-reaching as the history of their organic bases.

f. *Sensibility — Sources of Error.*

Another important factor in animal behavior, namely, sensibility, is very generally underestimated and often sadly misunderstood. We are apt to gauge sensibility according to the intensity of the overt response to stimulus, forgetting that the animal has the power to inhibit such manifestations or to moderate them in a way to mislead the observer. In the struggle for existence a high premium has been placed on this power, with the result that it is well-nigh a universal attribute. The best proof of its high value to the possessor is our own readiness to accept the disguise it affords as an evidence of lack of sensibility. We are so prone to think that the exercise of such power depends upon considerable intelligence that we are incredulous of its existence in forms that give only doubtful signs of intelligence. The power is possessed to a very marked degree by *Clepsine*, and it is only when we become aware of this fact and take all necessary precautions that we can get any reliable tests of the animal's keenness of sensibility. *Necturus* is even more difficult to manage, for not until after we have won its confidence by slow degrees can we expect free responses.

Besides the great danger of being deceived by the response, or the lack of response, to stimulus, there are two other insidious sources of error to be guarded against. We habitually assume that intelligence and sensibility rise and fall together. This idea may lead to false conclusions in two directions — to overestimating sensibility at the upper end of the scale and underestimating it at the lower. That high sensibility does not imply high intelligence is clear in the case of *Clepsine* and equally so in almost any other case that might be selected among the lowest segmented animals. That high intelligence does not necessarily imply correspondingly high sensibility is shown by the well-known fact that many animals greatly surpass man in their sense powers. It can be shown, I believe, that the difference in sensibility between higher and lower animals is very much less than is generally supposed.

The second source of error is the common assumption that the grade of sensibility rises with the structural complexity of the sense organs. This view is likewise untenable. It is true that the sense organs as a rule become more complex in structure as we go up the scale, but this advance in structure is mainly confined to accessory and non-sensory parts, which are either of a protective nature or else concerned in some subsidiary function, such as muscular adjustments and regulation of the stimuli. Such improvements in the non-sensory parts may be carried to a high state of perfection and greatly raise the general efficiency of the organ (*e.g.*, the vertebrate eye), without adding much, if anything, to the sensitiveness of the individual sense cells. The sense cells may be multiplied in number and placed in a position of safety and advantage for receiving stimuli, and the stimuli may be strengthened, directed, and otherwise regulated so as to secure the best results; but all that may obviously not affect the functional power of the cells themselves. We do not know the range of variability in this power, but we do know that the sense cells often vary relatively little in structure, sometimes retaining in the higher forms the same typical features that characterize them in the lower forms. There is no known difference of structure that would warrant the assumption that the dermal sensillæ in annelids are less sensitive than those in aquatic vertebrates. We have, then, no reliable test of sensibility either in the structure of the sense organ, in the rank of the organism, or in its intelligence. We have to depend upon the response to stimuli, and, remembering that this may be deceptive, observe and experiment under conditions that insure *free behavior.*

No one who has never come into close communion with the lower animals can begin to appreciate the delicacy and efficiency of their sensory apparatus. We take up the earthworm and, as we see no eyes, we conclude that it cannot see. A little experiment shows that it is extremely sensitive to light, and further study of its structure reveals unpigmented eyes lying beneath the skin, and the whole surface thickly set with minute delicate tactile sensillæ. Even *Amphioxus*, so long reputed to have no visual organs, turns out to have such organs from end to end, imbedded in its spinal cord. I have before called attention to the highly sensitive organization of *Clepsine* and its allies. In the very lowest organisms, plant and animal alike, where special visual organs do not exist, the living protoplasm has, as has been demonstrated in many ways, a keen sensibility to light, so that one might look upon the whole organism as fulfilling the light-perceiving function.

g. *Orientation through the Dermal Sensillæ.*

Necturus, as before remarked, has a very keenly sensitive organization. The skin is richly provided with sense organs, which terminate at the surface in very short, fine hairs, invisible to the naked eye. These organs, which are of the same nature and function as the dermal sensillæ in *Clepsine* and in so many other aquatic animals, are sensitive to slight vibrations in the water that are far beyond the reach of any of our sense organs, and they are the main reliance, both in avoiding enemies and seeking prey.

It is interesting to see how little the eyes are depended upon in finding a piece of meat. A bit dropped in front of a young *Necturus* receives no attention after it reaches the bottom.

An object must be in *motion* in order to excite attention, and it is not generally the moving form that is directly perceived, but the movements of the water, travelling from the object to the sensory hairs, are felt, and in such a way as to give the direction of the disturbing centre with most surprising accuracy. If a bit of beef is taken up adhering to the point of a needle, and held in the water, the vibrations imparted to the needle by the most steady hand will be sufficient to give the animal the direction. If the meat falls to the bottom, and the needle is held in place, the animal approaches the needle and tries to capture it, without paying the slightest attention to the meat lying directly below. If, after the meat has fallen, the needle is withdrawn and touched to the surface of the water behind or at one side of *Necturus*, it turns instantly in the direction of the needle, not because it sees, but because it *feels* wave motions coming from that direction. Long experience with *Necturus* and with many of its nearer allies enables me to speak very positively on this point. When it is remembered that in the higher animals the direction of sound waves is given by the auditory sense organs, which are primarily surface sensillæ homologous with those in the skin of *Necturus*, it may not seem so strange that the animal directs its movements in the way described. *Necturus* can see, but it can feel (perhaps we should say hear) so much more efficiently that its small eyes seem almost superfluous.[1]

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[1] Professor Eigenmann has kindly written the following note on the use of the tactile organs in the blind fishes:

Chologaster papilliiferus, a relative of the blind fishes living in springs, detects its prey by its tactile organs, not by its eyes. A crustacean may be crawling in plain view without exciting any interest unless it comes in close proximity to the head of the fish, when it is located with precision and secured. The action is in very strong contrast to that of a sunfish, which depends on its eyes to locate its prey. A *Gammarus* seen swimming rapidly through the water and approaching a *Chologaster* from behind and below was captured by an instantaneous movement of the *Chologaster*, when it came in contact with its head. The motion brought the head of the *Chologaster* in contact with the stem of a leaf, and instantly it tried to capture this also. Since the aquarium was well lighted, the leaf in plain sight, it must have been seen and avoided if the sense of sight and not that of touch were depended upon.

In *Amblyopsis*, the largest of the blind fishes of the American caves, the batteries of tactile organs form ridges projecting beyond the general surface of the

h. *Origin and Nature of the Behavior in Taking Food.*

1. *Some Intelligence Implied.* — Let us now return to the question of the origin and nature of the behavior of *Necturus* in capturing its food; not, however, with the expectation of reaching a complete solution, but rather in the hope of coming nearer to the problem and to the guiding principles in dealing with it. It is obvious, first of all, that automatism will not suffice to account for the whole behavior. That there is organic coördination of movements no one will dispute. But these movements must be steered in the direction of the object, and this orientation does not seem to be a purely automatic arrangement. The dermal sensillæ ("lateral-line" organs) give the impressions which enable the animal to steer its course; but action and sense impression are evidently not linked in a way to be independent of inhibitory influences. I assume that the creature is conscious, and that it has a certain intelligent appreciation of the sense impressions received. This is not saying that the young *Necturus* is a born philosopher; I assume nothing more than that it has already learned by experience how to *direct* its movements. That does not imply much, but certainly some, intelligence. I cannot otherwise understand why the same stimulus should not always evoke the same response under the same conditions. But we see that there is hesitation about starting, and this hesitation may be prolonged to any length, showing conclusively that sensation and response are not so connected as to exclude

skin. Its prey, since it lives in the dark and its eyes are mere vestiges, is located entirely by its tactile organs. This is done with as great accuracy as could be done with the best of eyes in the light, but only when the prey is in close proximity to the head. Coarser vibrations in the water are not perceived or are ignored, and apparently stationary objects are not perceived when the fish approaches them. If a rod is held in the hand, the fish always perceives it when within about half an inch of it, and backs water with its pectorals. If the head of a fish is approached with a rod, the direction from whence it comes is always perceived and the correct motion made to avoid it. This reaction is much more intense in the more active young than in the adult. One young, about 10 mm. long, determined with as great precision the direction from which a needle was coming as any fish with perfect eyes could possibly have done. It reacted properly to avoid the needle, and this without getting excited about it.

inhibitory influences. There is unmistakably a power of inhibition strong enough to counteract the strongest motive to act — the hunger of a starving animal in the presence of food.

2. *Orientation Learned by Experience.* — In assuming that the young *Necturus*, at the time of its first attempt to capture a piece of beef, has already learned to orient itself with reference to external objects, I have not gone beyond the possibilities. The animal has been out of the egg envelopes for about two months. It has been confined in a glass dish about ten inches in diameter, holding water about one inch in depth. Its life has been about as exclusively vegetative as if it had been all the while within the egg membrane, the only difference being that it has had room to straighten itself and to move about to some extent among its fellows. It has been heavily laden with food-yolk and has maintained a quiet attitude except when disturbed by change of water. Simple as the life has been, the animal has had some experience in swimming and walking, and opportunity to use to some extent its organs of orientation. The bait offered to it is something totally new in its experience, but we cannot, of course, claim that its behavior towards the bait is wholly uninfluenced by its previous experience.

3. *Deferred Instinct.* — We have to do with what Mr. Lloyd Morgan has termed a "deferred" instinct, *i.e.*, an action performed for the first time, but not until some time after birth. Mr. Morgan's remarks on the first dive of a young moor hen[1] bring out very clearly the possible influence of experience in the case of such deferred instincts.

Mr. Morgan says:

> In the case of such an instinctive procedure of the deferred type as that presented by the diving of a young moor hen, though, on the first occasion of its performance, the congenital automatism predominates, *yet it is difficult to believe and is in itself improbable that the individual experience of the young bird does not, even on the first occasion, exercise some influence on the way in which the dive is performed.* If we desire to reach a true interpretation of the facts, we must realize the fact that an activity may be of mixed origin. And if we distinguish

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[1] Habit and Instinct, pp. 136, 137.

— as we have endeavored clearly to distinguish — between instinct as congenital and habit as acquired, we must not lose sight of the fact that there is much interaction between instinct and habit, so that the first exhibition of a deferred instinct may well be carried out in close and inextricable association with the habits which, at the period of life in question, have already been acquired.

Although Mr. Morgan's young moor hen had undoubtedly learned far more by experience before its first dive than my young *Necturus* could have learned before its first effort to capture food, we are nevertheless well admonished to keep in mind the fact that the activity here considered may not be pure instinct. Allowing for the small though important part played by intelligence, there remains a purposive sequence of coördinated acts, which are always performed in essentially the same manner by young and old, and by the young without instruction or example or previous experience of like motive and stimulus. In so far, then, as intelligence cannot possibly be a regulating factor we must refer the activity to organization.

4. *Pause before the Bait.* — In order to exclude as much as possible the influence of experience it will be well to confine attention to the least variable part of the behavior. The concluding phase of the performance is so typical and characteristic, and so far removed from anything previously experienced, that it may be regarded as a very near approach to pure instinct. I have in mind *the pause before the bait and then the quick side-movement of the head as the jaws are opened to seize.*

If this series of acts represents an organic sequence, and if the behavior as a whole takes the form determined by the organization, as seems to me beyond reasonable doubt, we have an instinct the history of which may be coextensive with the evolution of the animal. We stand at the end of an interminable vista. The specific peculiarities of organization in *Necturus* form but an infinitesimal element of the problem. Scarcely a feature of the instinct belongs exclusively to *Necturus.* It is at least widely diffused among vertebrates, especially among fishes. The differences in the manner of execution among different forms, so far as I have observed, are of quite a superficial nature. The instinct evidently has its root in the general instinct of preying, which is doubtless coeval with animal organization. The cannibalism of our protozoan ancestors was the starting-point, and their carnal propensities were not acquired by the aid of intelligence, but given in the fundamental properties of protoplasm. The stronger ate the weaker, and made themselves stronger and more prolific by so doing. The promise of the whole animal world was contained in the act. The constitutional disposition to feed, with variable foods available, would give occasion for different appetites and various modes of getting outside of palatable victims. In primitive organisms multiplying by simple fission, structural modifications acquired during the lifetime of the individual would be carried right on from generation to generation, and hence the structural foundations for a whole animal world such as we now see could be laid in a relatively short period as compared with the time necessary to advance organization in forms limited to reproduction by germs. In fact, these fundamentals could all be established within the realm of the unicellular protozoa. Nucleus and cell-body, inner and outer layers, nerve-muscle elements, sensory and locomotor organs, mouth and stomach, respiratory and excretory mechanisms, reproductive elements, anticipating embryological development from germs — all these essentials of higher organization are presented in the protozoan.

The organic bases furnished in the protozoan world might be passed directly on to the first metazoa, or they might be reacquired in essentially the same manner as before, and in a not much longer period, as reproduction by fission would still be a condition favoring rapid organo-genesis.

To try to fill up the gaps between the protozoan and *Necturus* would lead us too far into the field of speculation, and would not contribute much to a grasp of the problem. We have to content ourselves with general facts and principles and probabilities drawn therefrom. It is enough for present purposes to know that the roots of the instinct organization we are considering run clear back to the beginnings of organo-genesis, and that they are natural products of the properties of living protoplasm. We start with known properties and get to known rudiments of organization without invoking the aid of intelligence, or finding any way in which it could be supposed to have been a guiding factor in development.

The organic basis of the preying instinct may have grown and multiplied in different phyla a long time before receiving much aid from intelligence. The rapidity and freedom of modification would be very much limited when fission ceased and reproduction by germs became the sole mode of generation. Very early in the vertebrate phylum, possibly at its dawn, the chief characters of the instinct, as we now find it, were probably fixed in structural elements differing from those in *Necturus* only in superficial details. The strikingly fish-like character of the behavior certainly suggests as much.

5. *Meaning and General Occurrence.* — If now we look more closely at the purposive character of the behavior, it will become clearer that the instinct is shared, not only by animals below *Necturus*, but also by some far above it. The pause before the final act of seizing is a well-marked feature, which means *locating the prey and fixing the aim.* The same action with the same meaning runs all through the different branches of the vertebrate phylum. It is, as I have already said, especially characteristic of the fishes and amphibia, and it is not rare among the higher branches, the reptiles, birds, and mammals. It may be seen to good advantage in the turtles, and even the common fowl halts on coming up to the insect it is pursuing in order to make sure its aim. I believe the same instinct underlies the act of *pointing* in the dog. The origin of this behavior in pointers cannot be referred to training, as was clearly seen by Darwin.[1]

> It may be doubted [says Darwin] whether any one would have thought of training a dog to point had not some one dog naturally shown a tendency in this line, and this is known occasionally to happen, as I once saw in a pure terrier; the act of pointing is probably, as many have thought, *only the exaggerated pause of an animal preparing to spring on its prey.* When the first tendency to point was once displayed, methodical selection and the inherited effects of compulsory training in each successive generation would soon complete the work.

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[1] Origin of Species, p. 207.

The "tendency" manifested in some one dog was regarded by Darwin as an accidental variation, the cause being unknown. May not many of the variations appearing in domestic animals, which we call "accidental," be manifestations of instinct roots of more or less remote origin?

6. *Part Played by Fear.* — We may now glance once more at the behavior as a whole, for the purpose of pointing out the part played by instinctive timidity. Gentle movements in the water, kept up with steadiness, such as are imparted by a needle in feeding as before described, may induce an attack, while less gentle or unsteady movements may lead the animal to remain quiet or to take flight. The same stimulus, according to amplitude and evenness, may then be followed either by advance, by quiet, or by retreat. In retreat, fear is manifest; in quiet it is concealed; in advance it is less concealed. There can be no doubt that fear predominates in flight and in quiet, while it certainly tempers the advance, giving the appearance of slyness deliberately acted in order to take the prey by surprise. This sly manner of advance, whatever it be due to, has a double advantage, for it is concealment against a possible foe and prevents alarming a harmless prey. If I could suppose that fear did not strongly influence the advance, I should certainly incline to think that the animal really appreciated the great advantages in quietly surprising its prey; but for reasons before given I believe the animal is quite blind to any such bearing of its action. The advantages of this manner of action, however, are just the same as if it were deliberately assumed, and the *Necturus* conducting itself in this way would certainly fare better than one reacting in a contrary way. The instincts of *Necturus* in this case coöperate to secure its welfare, while if the creature depended upon its intelligence it is difficult to see how it could escape immediate extinction.

GENERAL CONSIDERATIONS.

a. *Instinct Precedes Intelligence.*

The view here taken places the primary roots of instinct in the constitutional activities of protoplasm[1] and regards instinct in every stage of its evolution as action depending essentially upon organization. It places instinct before intelligence in order of development, and is thus in accord with the broad facts of the present distribution and relations of instinct and intelligence, instinct becoming more general as we descend the scale, while intelligence emerges to view more and more as we ascend to the higher orders of animal life. It relieves us of the great inconsistencies involved in the theory of instinct as "lapsed intelligence." Instincts are universal among animals, and that cannot be said of intelligence. It ill accords with any theory of evolution, or with known facts, to make instinct depend upon intelligence for its origin; for if that were so, we should expect to find the lowest animals free from instinct and possessed of pure intelligence. In the higher forms we should expect to see intelligence lapsing more and more into pure instinct. As a matter of fact, we see nothing of the kind. The lowest forms act by instinct so exclusively that we fail to get decided evidence of intelligence. In higher forms not a single case of intelligence lapsing into instinct is known. In forms that give indubitable evidence of intelligence we do not see conscious reflection crystallizing into instinct, but we do find instinct coming more and more under the sway of intelli-

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[1] Professor Loeb* refers instinct back to "(1) polar differences in the chemical constitution in the egg substance, and (2) the presence of such substances in the egg as determine heliotropic, chemotropic, stereotropic, and similar phenomena of irritability." According to this view, the power to respond to stimuli lies in unorganized chemical substances, and the same powers exist in the adult as in the egg, because the same chemical substances are present. Organization serves at all stages merely as a mechanical means of giving definite directions to responses.

The view I have taken regards instinctive action as *organic* action, whatever be the stage of manifestation. The egg differs from the adult in having an organization of a very simple primary order, and correspondingly simple powers of response. Instinct and organization are, to me, two aspects of one and the same thing, hence both have ontogenetic and phylogenetic development.

* "Egg Structure and the Heredity of Instincts," *The Monist*, vol. vii, July, 1897.

gence. In the human race instinctive actions characterize the life of the savage, while they fall more and more into the background in the more intellectual races.

Every hypothesis that would derive instinctive action from teleological reflection is open to the same objections. In many cases it would be necessary to postulate an amount of prevision on the part of the animals in which the instincts arose that would simply be psychologically impossible. Conscious prevision without a possible basis in the experience of the individual, or any means of learning from others, is simply a self-contradiction. The frequently cited instance of the emperor moth puts this point in strong light. The caterpillar of this moth so constructs the upper part of its cocoon that it will resist strong pressure from without and yield to slight pressure from within. Easy egress for the imago and security against attacks from outside enemies are thus provided for. As the spinning of the cocoon happens but once in a lifetime, the caterpillar could not anticipate such needs from its own experience, nor could it learn from its parents, which were dead long before it hatched. The possibility of imitation is also excluded, as the species is not a social one.

b. *Theories of Instinct.*

1. *Pure Instinct the Point of Departure.* — The first criterion of instinct is, that it can be performed by the animal without learning by experience, instruction, or imitation. The first performance is therefore the crucial one. It is of the utmost importance in all discussion of the origin of instinct to make sure of this point, and keep clear of all ambiguous activities such as have been designated "instinct habits" (Lloyd Morgan), "acquired instincts" (Wundt), "secondary instincts" (Romanes), etc. We must not allow the question as to the relation of instinct to habit and intelligence to be obscured by confusing terminology. There may be "mixtures" and all sorts of "interactions" between habit and instinct, and these may have a far-reaching theoretical import, but they lack definiteness, and are therefore dangerous foundations for theories. A

theory of instinct must obviously make pure instinct its first concern, and keep the general course of evolution always in view.

It is not my purpose to engage in a critical examination of theories, but to indicate briefly which of the two rival theories now most in favor accords best with facts and general principles as I understand them. These two theories are the *habit theory* of Lamarck and the *selection theory* of Darwin, Wallace, and Weismann.

2. *Embryology and the Lamarckian Theory.* — The habit theory is a part of the more general theory of the transmission of acquired characters. This doctrine has never been reconciled with the teachings of embryology, the science which deals directly with the phenomena of heredity, and which is, therefore, the touchstone of every theory of inheritance. It is a fundamental tenet in embryology that all organisms reproducing exclusively by germs owe their inherited characters to the germs from which they arose, and that germs carry the primordials of adult structure, not by virtue of any mysterious transference of parental features, but by virtue of the constitution they bring with them when they arise by division of preëxisting germs. That is, I believe, a fair statement of the embryological doctrine of inheritance, which must be the final test of our theories.

The selection theory propounded by Darwin and Wallace, and further developed by Weismann, starts from the embryological law of germ continuity (Weismann), or, otherwise expressed, germ lineage, and interprets the phenomena of variation, heredity, and development, in harmony with this law and the principle of selection. This theory is incompatible with the idea that instinct is inherited habit. We could not, for example, say with Professor Wundt[1]:

> "We have supposed that father can transmit to son the physiological dispositions that he has acquired by practice during his own life, and that in the course of generations these inherited dispositions are strengthened and definitized by summation."
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> "The occurrence [p. 405] of connate instincts renders a subsidiary hypothesis necessary. We must suppose that the physical

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[1] Lectures on Human and Animal Psychology, p. 408.

changes which the nervous elements undergo can be transmitted from father to son. . . . The assumption of the inheritance of acquired dispositions or tendencies is inevitable if there is to be any continuity of evolution at all."

3. *Darwin's Refutation of Lamarck's Theory.* — Although Darwin dwelt at some length on the points of resemblance between habits and instincts, and although he thought it possible that habits could sometimes be inherited, it should be remembered that he was the first to show conclusively that "the most wonderful instincts with which we are acquainted, namely, those of the bee hive and of many ants, could not possibly have been acquired by habit" (*Origin of Species*, p. 202). Indeed, it was he who first found in the case of neuter insects a refutation of Lamarck's doctrine of inherited habit, and at the same time a demonstration of the high efficiency of the principle of natural selection. Darwin concludes his chapter on instinct with these memorable words:

> "The case of neuter insects, also, is very interesting, as it proves that with animals, as with plants, any amount of modification may be effected by the accumulation of numerous slight, spontaneous variations, which are in any way profitable, *without exercise or habit having been brought into play. For peculiar habits confined to the workers or sterile females, however long they might be followed, could not possibly affect the males and fertile female, which alone leave descendants. I am surprised that no one has hitherto advanced this demonstrative case of neuter insects against the well-known doctrines of inherited habit, as advanced by Lamarck.*"

What could more forcibly illustrate the importance of crucial cases than just this work of Darwin's on the instincts of neuter insects? Here a conclusive test is reached, and no theory of the origin of instinct can stand that disregards it. If habit cannot possibly have had anything to do with the origin of such typical instincts, then we should at least be very cautious in appealing to it in any case. We certainly do not want two theories to account for the same phenomenon, if one will suffice. If the theory of inherited habit is certainly false in a single case, it must be deemed false in every case, until at least it has been shown that some cases cannot be explained without it. Is there any case where it can be clearly shown that an undoubted instinct arose from inherited habit, or any case in which it can be made clear that the theory adopted for neuter insects cannot possibly hold? Both questions, it seems to me, must be answered in the negative.

4. *Weak Points in the Habit Theory.* — The habit theory has many adherents still, and Darwin himself often found it a convenient hypothesis. But neither Darwin nor anybody else has given us a crucial test that would stand beside that furnished in neuter insects. The failure to find such a test is certainly not due to any lack of zeal or effort on the part of the advocates of the theory. The tests claimed are numerous enough, but they always fall short of the requirement. The weak points in the theory are:

1. It starts on a disputed, if not refuted assumption; namely, that habits wholly new to the individual and the species, having no hereditary basis predisposing to them, may, as the result of exercise frequently repeated, and continued in successive generations, eventually become hereditary instincts.

2. It appeals to the less typical rather than to the more typical cases — to cases in which the critical points are undetermined or doubtful, or open to a different interpretation.

3. Its definition of habit and instinct verges towards a *petitio principii.* Two or more classes of instincts are set up so as to facilitate a nearer approach to habit, *e.g.*, acquired and connate (Wundt); primary and secondary (Romanes). Habit is used indiscriminately for an action originating in some congenital variation and an action forced upon the individual by special circumstances. A fundamental distinction, on which the validity of the theory must be tested, is thus ignored.

c. *Two Demonstrations of the Habit Theory Claimed by Romanes.*

The evidence adduced to show that habit may pass into instinct cannot here be examined in detail. Romanes brings forward two cases — the instincts of *tumbling* and *pouting* in pigeons — which he declares are alone sufficient to demonstrate the theory. We may, therefore, take these as fair samples of the arguments generally appealed to.

After quoting Darwin's remarks on this subject, Romanes adds:

> "This case of the tumblers and pouters is singularly interesting and very apposite to the proposition before us; for not only are the actions utterly useless to the animals themselves, but they have now become so ingrained into their psychology as to have become severally distinctive of different breeds, and so not distinguishable from true instincts. This extension of an hereditary and useless habit into a distinction of race or type is most important in the present connection. *If these cases stood alone, they would be enough to show that useless habits may become hereditary*, and this to an extent which renders them indistinguishable from true instincts."[1]

Granting that we have here true instincts, — and I do not doubt that, — what proof have we that they originated in habits? Did there preëxist in the ancestors of these breeds organized instinct bases, which, through the fancier's art of selective breeding, were gradually strengthened until they attained the development which now characterizes the tumblers and pouters? Or was there no such basis to start with, but only a new mode of behavior, accidentally acquired by some one or more individuals, and then perpetuated by transmission to their offspring, and further developed by artificial selection? The original action in either species is called a "habit," and this so-called habit must have been inherited; ergo, habit can become instinct. Obviously, argument of that kind can have weight only with those who overlook the test-point, namely, the real nature and origin of the initial action.

If the instinct had its inception in a true habit, *i.e.*, in an action reduced to habit by repetition in the individual, and not determined in any already existing hereditary activity, is it at all credible that it could have been transmitted from parent to progeny? Does not our general experience contradict such an assumption in the most positive manner? But may not the habit have originated a great many times, and by repetition in successive generations, gradually have become "stereotyped

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[1] Mental Evolution in Animals, p. 189.

into a permanent instinct"? To suppose that such *utterly useless* action originated a great many times without compelling conditions or any organic predisposition is not at all admissible.

Darwin saw at once from the nature of the actions that they could not have been taught, but "*must have appeared naturally*, though probably afterwards vastly improved by the continued selection of those birds which showed *the strongest propensity*." Darwin, then, postulates as the foundation of each instinct a "propensity" — something given in the constitution. That view of the matter is in entire accord with the theory adopted in the case of neuter insects and quite incompatible with the habit theory.

1. *The Instinct of Pouting.* — I believe the case is much stronger than Darwin suspected, and that it shows, not the genesis of instinct from habit, but from a prëexisting congenital basis. Such a basis of the pouting instinct exists in every dovecot pigeon, and is already an organized instinct, differing from the instinct displayed in the typical pouter only in degree. I could show that the same instinct is widely spread, if not universal, among pigeons. It will suffice here to call attention to the instinct as exhibited in the common pigeon. Observe a male pigeon while cooing to his mate or his neighbors. Notice that he inflates his throat and crop, and that this feature is an invariable feature in the act, often continued for some moments after the cooing ceases. Compare the pouter and notice how he increases the inflation whenever he begins cooing. The pouter's behavior is nothing but the universal instinct enormously exaggerated, as any attentive observer may readily see under favorable circumstances.

2. *The Instinct of Tumbling.* — The origin of the tumbling instinct cannot be fixed by the same direct mode of identification; but I believe that here also it is possible to point to a more general action, instinctively performed by the dovecot pigeon, as the probable source of origin. I have noticed a great many times that common pigeons, when on the point of being overtaken and seized by a hawk, suddenly flirt themselves directly downward in a manner suggestive of tumbling, and thus elude the hawk's swoop. The hawk is carried on by

its momentum, and often gives up the chase on the first failure. In one case I saw the chase renewed three times, and eluded with success each time. The pigeon was a white dovecot pigeon with a trace of fantail blood. I saw this same pigeon repeatedly pursued by a swift hawk during one winter, and invariably escaping in the same way. I have seen the same performance in other dovecot pigeons under similar circumstances.

But this is not all. It is well known that dovecot pigeons delight in quite extended daily flights, circling about their home. I once raised two pairs of these birds by hand, in a place several miles from any other pigeons. Soon after they were able to fly about they began these flights, usually in the morning. I frequently saw one or more of the flock, while in the middle of a high flight, and sweeping along swiftly, suddenly plunge downwards, often zigzagging with a quick, helter-skelter flirting of the wings. The behavior often looked like play, and probably it was that in most cases. I incline to think, however, that it was sometimes prompted by some degree of alarm. In such flights the birds would frequently get separated, and one thus falling behind would hasten its flight to the utmost speed in order to overtake its companions. Under such circumstances the stray bird coming from the rear might be mistaken for the moment for a hawk in pursuit, and one or more of the birds about to be overtaken be thus induced to resort to this method of throwing themselves out of reach of danger.

The same act is often performed at the very start, as the pigeon leaves its stand. The movement is so quick and crazy in its aimlessness that the bird often seems to be in danger of dashing against the ground, but it always clears every object.

As this act is performed by young and old alike, and by young that have never learned it by example, it must be regarded as instinctive, and I venture to suggest that it probably represents the foundation of the more highly developed tumbling instinct.

The behavior of the Abyssinian pigeon, which, when "fired at, plunges downwards so as to almost touch the sportsman, and then mounts to an immoderate height," may well be due to the same instinct. The noise of the gun, even if the bird were not hit, would surprise and alarm it, and the impulse to save itself from danger would naturally take the form determined by the instinct, if the instinct existed. This seems to me more probable than Darwin's suggestion of a mere trick or play.

d. *The Habit Theory Losing Ground.*

The two instincts of pouting and tumbling, claimed as demonstrations of the habit theory, thus turn out to be explicable only on the selection theory. It is significant that this theory is fast losing ground even among the psychologists. A. Forel's conversion illustrates the trend of opinion. "I, too," he says, "used to believe that instincts were hereditary habits, but I am now convinced that this is an error, and have adopted Weismann's view. It is really impossible to suppose that acquired habits, like piano playing and bicycle riding, for instance (these are certainly acquired), could hand over their mechanism to the germ-plasm of the offspring."[1]

In his latest work, *Habit and Instinct,* Lloyd Morgan has also abandoned the theory. On the same side stand James, Baldwin, Ziehn, Flügel, and others. The following, from Karl Groos, pp. 60, 61, will show how the difficulties with the theory are multiplying.

"As regards instinct," says Groos, "there is, further, the *a priori* argument that it is inconceivable how acquired connections among the brain cells could so affect the inner structure of the reproductive substance as to produce inherited brain tracts in later generations. And, finally, there is this consideration mentioned by Ziegler as a suggestion of Meynert's: 'It is well known that in the higher vertebrates acquired associations are located in the cortex of the hemispheres. As an acquired act becomes habitual, it may be assumed that the corresponding combination of nervous elements will become more dense and strong, and the tract proportionally more fixed. This being the case, it follows that the tracts of acquired and habitual association, as well as those of acquired movement, pass

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[1] Gehirn und Seele, 1894, p. 21. Taken from The Play of Animals, by Groos, p. 56. (Translated by Elizabeth L. Baldwin.)

through the cerebrum. Instincts and reflexes, however, have their seat for the most part elsewhere. The tracts of very few of them are found in the cortex of the hemispheres. It is chiefly in the lower parts of the brain and spinal cord that the associations and coördinations corresponding to instincts and reflexes have their seat. When the comparative anatomist investigates the relative size of the hemispheres in vertebrates (especially in amphibians, reptiles, birds, and mammals), a very evident increase in size is observed which apparently goes hand in hand with the gradual gain in intelligence. In the course of long phylogenetic development, during which the hemispheres have gradually attained their greatest dimensions, they have constantly been the organ of reason and the seat of acquired association. If, then, habit could become instinct through heredity, it is probable that the cerebrum would, in much greater degree than is the fact, be the seat of instinct.' "

The stronghold of the Lamarckian view is Paleontology. It is here that the doctrine of acquired characters, or ctetology as Professor Hyatt calls it, has been nearly as unyielding as the fossils to which it adheres. But a new light seems to be penetrating even here under the name of "organic selection." This idea, first formulated by Professor Baldwin, but almost simultaneously and independently reached by Lloyd Morgan and Professor Osborn, is, that adaptive modifications are not transmissible, but that they have, nevertheless, acted as *the fostering nurses of congenital variations*, since organisms surviving through them would carry forward to the next generation such congenital variations as happened to be coincident with them. It may be, perhaps, a fine question to determine whether so-called "adaptative modifications," which really have selection value, are not themselves the coincidents of congenital bases. Be that as it may, the conversion of so eminent a paleontologist as Professor Osborn to the selection theory is all the more significant on account of the prominent part he has taken in defending the Lamarckian idea.

e. *Hyatt on Acquired Characters.*

Professor Hyatt was the first to demonstrate a wonderfully complete parallelism between the ontogenetic and the phylogenetic series, and he has presented the paleontological argument in terms that seem, to many at least, to be beyond controversy. With all respect to Professor Hyatt's monumental work, I must say that I find nothing in the evidence that compels one to take his view of acquired characters.

> "We have been unable" [says Professor Hyatt] "to find any characters which were not inheritable in some series. The behavior of all characteristics which have been introduced into any series of species shows them to be subject to the law of acceleration, in whatever way they have originated, whether primarily as adaptive characters, according to our hypothesis, or by natural selection and through the combination of the sexual variations, as supposed by Weismann."[1]

This is a very sweeping statement, at least in implication. I can hardly believe that the author would have us understand that acquired characters are just as readily and invariably transmitted as congenital characters; and yet, if that is not the argument, there is no argument there. Nothing is more certain than that, in living forms accessible to direct experimental test, acquired characters are not invariably, if at all, transmissible. Demonstrations have been sought for, but so far without avail. Unless the *Arictidæ* are a wholly exceptional group, we must conclude from the above statement that all the characters found were of congenital origin, and that no acquired characters were recognized. It is easier to believe that such characters were overlooked than to believe a miracle.

The *law of acceleration* established by Professor Hyatt is complemental to *the biogenetic law* formulated by Fr. Müller and Haeckel, and both laws rest on the theory of germ continuity, as formulated by Weismann. Logically, neither of these laws implies the transmission of acquired characters. That is an assumption which has never been reconciled with the fundamental law of the genetic continuity of germs. The

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[1] "Genesis of the Arietidæ," p. 43, *Mem. Mus. Comp. Zoöl.*, Cambridge, 1889.

pangenesis theory of Darwin was an attempt in this direction, but that theory has no scientific basis and it stands as a theoretical failure, rejected because it could not possibly be reconciled with what we know about the genesis of germs. That is the inevitable fate of every view which fails to adjust itself to the primary law of germ continuity.

Sense impressions and physical impressions or modifications stand on the same footing. Repetition may become habit and produce marked effects on the nervous mechanism or other organs; but the individual structure so affected is not continued from generation to generation, so that the effects are cancelled with each term of life, and there is no conceivable way by which they could be stamped upon the germs and so carried on cumulatively. If they reappear in the offspring, it cannot be because they were inherited, but because they are reproduced in the same way as they were acquired in the parent.

f. *Preformation the Essence of the Doctrine.*

This doctrine of the transmission of acquired characters is a species of preformation that eclipses the old creation hypothesis, for the miracle of stamping the germ with the form it is to present in the adult has to be repeated at each generation.

It may be objected that "stamping" is not the method by which parental characters are given to the germ. They are commonly said to be inherited. But it is too late to juggle with the term "heredity." That term either means something or nothing. If it means that characters acquired by the parent can be transmitted to the offspring, then the transmitted characters, which *ex hypothesi* are not originally determined in the germ, must in some way be determined for it by the parent. What better term than "stamp" or "impress" can be suggested? Whatever the *modus operandi*, the determining influence or impress must be imparted, at least in the great majority of cases, before development begins. Is it conceivable that perfectly definite form features can be in any way reflected back upon the ovum? Can we think of the germ as vibrating sympathetically with each acquired peculiarity of the parent organism? What vibration could there be between germ and passive structures, such as shell configurations? Could an indentation, groove, ridge, or protuberance forced upon a shell by environmental action be at the same time wrought into the germs in such a definite way as to reappear in the offspring without the aid of the same environmental causes? Or could the repetition of the same environmental action on a long line of parents gradually modify the germs in the same direction? In whatever way we turn the question, we are confronted with the same miracle of preformation. *The character arises in the parent organism by epigenesis, but it is thrown back on the germ, nobody knows or can conceive how, in such a way that it becomes a preformation capable of unfolding without the aid of its epigenetic causes.*

On the other hand, the hypothesis that all hereditary characters in organisms exclusively gamogenetic originate in spontaneous or induced (by *direct* action of environment) germ variations, appeals only to known facts and principles, and provides for the same amount of preformation as before without any miraculous transfer of characters from one organism to another. We know that germ variations are transmissible; we do not know that individually acquired modifications can be transferred to germs; we know the principle of selection to be rational and verifiable; we know of no substitute for it.

THE GENETIC STANDPOINT IN THE STUDY OF INSTINCT.

a. *The Genealogical History Neglected.*

The problem of psychogenesis requires a more definite genetic standpoint than that of general evolution. It is not enough to recognize that instincts have had a natural origin; for the fact of their connected genealogical history is of paramount importance. From the standpoint of evolution as held by Romanes and others, instincts are too often viewed as disconnected phenomena of independent origin. The special and more superficial characteristics have been emphasized to the exclusion of the more fundamental phylogenetic characters.

Biologists and psychologists alike have very generally clung tenaciously to the idea that instincts, in part at least, have been derived from habits and intelligence; and the main effort has been to discover how an instinct could become gradually stamped into organization by long-continued uniform reactions to environmental influences. The central question has been: How can intelligence and natural selection, or natural selection alone, initiate action and convert it successively into habit, automatism, and congenital instinct? In other words, the genealogical history of the structural basis being completely ignored, how can the instinct be mechanically rubbed into the ready-made organism? Involution instead of evolution; mechanization instead of organization; improvisation rather than organic growth; specific *versus* phyletic origin.

This inversion, or rather perversion, of the genealogical order leads to a very short-focussed vision. The pouting instinct is supposed to have arisen *de novo*, as an anomalous behavior, and with it a new race of pigeons. The tumbling instinct was a sort of *lusus naturæ*, with which came the fancier's opportunity for another race. The pointing instinct was another accident that had no meaning except as an individual idiosyncrasy. The incubation instinct was supposed to have arisen *after* the birds had arrived and laid their eggs, which would have been left to rot had not some birds just blundered into "cuddling" over them and thus rescued the line from sudden extinction. How long this blunder-miracle had to be repeated before it happened all the time does not matter. Purely imaginary things can happen on demand.

b. *The Incubation Instinct.*

1. *Meaning to be Sought in Phyletic Roots.* — It seems quite natural to think of incubation merely as a means of providing the heat needed for the development of the egg, and to assume that the need was felt before the means was found to meet it. Birds and eggs are thus presupposed, and as the birds could not have foreseen the need, they could not have hit upon the means except by accident. Then, what an

infinite amount of chancing must have followed before the first "cuddling" became a habit, and the habit a perfect instinct! We are driven to such preposterous extremities as the result of taking a purely casual feature to start with. Incubation supplies the needed heat, but that is an incidental utility that has nothing to do with the nature and origin of the instinct. It enables us to see how natural selection has added some minor adjustments, but explains nothing more. For the real meaning of the instinct we must look to its phyletic roots.

If we go back to animals standing near the remote ancestors of birds, to the amphibia and fishes, we find the same instinct stripped of its later disguises. Here one or both parents simply remain over or near the eggs and keep a watchful guard against enemies. Sometimes the movements of the parent serve to keep the eggs supplied with fresh water, but aëration is not the purpose for which the instinct exists.

2. *Means Rest and Incidental Protection to Offspring.* — The instinct is a part of the reproductive cycle of activities, and always holds the same relation in all forms that exhibit it, whether high or low. It follows the production of eggs or young, and means primarily, as I believe, *rest* with incidental *protection to offspring.* That meaning is always manifest, no less in worms, molluscs, crustacea, spiders, and insects, than in fishes, amphibia, reptiles, and birds. The instinct makes no distinction between eggs and young, and that is true all along the line up to birds which extend the same blind interest to one as to the other.

3. *Essential Elements of the Instinct.* — Every essential element in the instinct of incubation was present long before the birds and eggs arrived. These elements are: (1) the disposition to remain with or over the eggs; (2) the disposition to resist and to drive away enemies; and (3) periodicity. The birds brought all these elements along in their congenital equipment, and added a few minor adaptations, such as cutting the period of incubation to the need of normal development, and thus avoiding indefinite waste of time in case of sterile or abortive eggs.

(1) **Disposition to Remain over the Eggs.** — The disposition to remain over the eggs is certainly very old, and is probably bound up with the physiological necessity for rest after a series of activities tending to exhaust the whole system. If this suggestion seems far-fetched, when thinking of birds, it will seem less so as we go back to simpler conditions, as we find them among some of the lower invertebrate forms, which are relatively very inactive and predisposed to remain quiet until impelled by hunger to move. Here we find animals remaining over their eggs, and thus shielding them from harm, from sheer inability or indisposition to move. That is the case with certain molluscs (*Crepidula*), the habits and development of which have been recently studied by Professor Conklin.[1] Here full protection to offspring is afforded without any exertion on the part of the parent, in a strictly passive way that excludes even any instinctive care. In *Clepsine* there is a manifest unwillingness to leave the eggs, showing that the disposition to remain over them is instinctive. If we start with forms of similar sedentary mode of life, it is easy to see that remaining over the eggs would be the most likely thing to happen, even if no instinctive regard for them existed. The protection afforded would, however, be quite sufficient to insure the development of the instinct, natural selection favoring those individuals which kept their position unchanged long enough for the eggs to hatch.

(2) **Disposition to Resist Enemies.** — The disposition to keep intruders from the vicinity of the nest I have spoken of as an element of the instinct of incubation. At first sight it seems to be inseparably connected with the act of covering the eggs, but there are good reasons for regarding it as a distinct element of behavior. In birds this element manifests itself before the eggs are laid, and even before the nest is built; and in the lower animals the disposition to cover the eggs is not always accompanied by an aggressive attitude. This attitude is one of many forms and degrees. A mild self-defensive state, in which the animal merely strives to hold its position without trying to rout intruders, would perhaps

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[1] *Journ. of Morph.*, vol. xiii, No. 1, 1897.

be the first stage of development. In some of the lower vertebrates the attitude remains defensive and is aggressive only in a very low degree, while in others pugnacity is more or less strongly manifested. Among fishes the little Stickleback is especially noted for its fiery pugnacity, which seems to develop suddenly and simultaneously with the appearance of the dark color of the male at the spawning season.

In pigeons, as in many other birds, this disposition shows itself as soon as a place for a nest is found. While showing a passionate fondness for each other, both male and female become very quarrelsome towards their neighbors. The white-winged pigeon (*Melopelia leucoptera*) of the West Indies and the southern border of the United States is one of the most interesting pigeons I have observed in this respect. At the approach of an intruder the birds show their displeasure in both tone and behavior. The tail is jerked up and down spitefully, the feathers of the back are raised as a threatening dog "bristles up," the neck is shortened, drawing the head somewhat below the level of the raised feathers, and the whole figure and action are as fierce as the bird can make them. To the fierce look, the erect feathers, the ill-tempered jerks of the tail, is added a decidedly spiteful note of warning. If these manifestations are not sufficient, the birds jump toward the offender, and if that fails to cause retreat, wings are raised and the matter settled by vigorous blows.

This pugnacious mood is periodical, recurring with each reproductive cycle, and subsiding like a fever when its course is run. The birds behave as if from intelligent motive, but every need is anticipated blindly; for the young pair, without experience, example, or tradition, behave like the parents.

It seems to me that this mood or disposition, although in some ways appearing to be independent of the disposition to cover the eggs, can best be understood as having developed in connection with the latter. It has primarily the same meaning, — protection to the eggs, — but the safety of the eggs and young depends upon the safety of the nest, and this accounts for the extension of its period to cover all three stages — building, sitting, and rearing.

(3) **Periodicity.** — The periodicity of the disposition to sit coincides in the main with that of the recuperative stage. Its length, however, at least in birds, is nicely correlated with, though not exactly coinciding with, the time required for hatching. It may exceed or fall short of the time between laying and hatching. The wild passenger pigeon (*Ectopistes*) begins to incubate a day or two in advance of laying, and the male takes his turn on the nest just as if the eggs were already there. In the common pigeon the sitting usually begins with the first egg, but the birds do not sit steadily or closely until the second egg is laid. The birds do not, in fact, really sit on the first egg, but merely stand over it, stooping just enough to touch the egg with the feathers. This peculiarity has an advantage in that the development of the first egg is delayed so that both eggs may hatch more nearly together. The bird acts just as blindly to this advantage as *Ectopistes* does to the mistake of sitting before an egg is laid. *Ectopistes* is very accurate in closing the period, for if the egg fails to hatch within twelve to twenty hours of its normal time, it is deserted, and that too if, as may sometimes happen, the egg contains a perfect young, about ready to hatch. Pigeons, like fowls, will often sit on empty nests, filling up the period prescribed in instinct, leaving the nest only as the impulse to sit runs down. It happens not infrequently that pigeons will go right on with the regular sequence of activities, even though nature fails in the most important stage. Mating is followed by nest-making, and at the appointed time the bird goes to the nest to lay, and after going through the usual preliminaries, brings forth no egg. But the impulse to sit comes on as if everything in the normal course had been fulfilled, and the bird incubates the empty nest, and exchanges with her mate as punctiliously as if she actually expected to hatch something out of nothing. This may happen in any species under the most favorable conditions. It is possible by giving an abundance of rich food to wind up the instinctive machinery more rapidly than would normally happen, so that recuperation may end in about a week's time, when incubation will stop and a new cycle begin, leading to the production of a second set of eggs in the same nest.

This has happened several times with the crested pigeon of Australia (*Ocyphaps lophotes*).

Schneider[1] says: "The impulse to sit arises, as a rule, when a bird sees a certain number of eggs in her nest." Although recognizing a *bodily disposition* as present in some cases, sitting is regarded as a *pure perception impulse*. I hold, on the contrary, that the *bodily disposition* is the universal and essential element, and that sight of the eggs has nothing to do primarily with sitting. It comes in only secondarily and as an adaptation in correlation with the inability in some species to rear more than one or two broods in a season. In such species the advantage would lie with birds beginning to incubate with a full nest.

The suggestions here offered on the origin of the incubation instinct, incomplete and doubtful as they may appear, may suffice to indicate roughly the general direction in which we are to look for light on the genesis of instincts. The incubation instinct, as we now find it perfected in birds, is a nicely timed and adjusted part of a periodical sequence of acts. If we try to explain it without reference to its physiological connections in the individual, and independently of its developmental phases in animals below birds, we miss the more interesting relations, and build on a purely conjectural chance act that calls for a further and incredible concatenation of the right acts at the right time and place, and is not even then completed until its perpetuation is secured by a miracle of transmission.

A FEW GENERAL STATEMENTS.

1. Instinct and structure are to be studied from the common standpoint of phyletic descent, and that not the less because we may seldom, if ever, be able to trace the whole development of an instinct. Instincts are evolved rather than involved (stereotyped by repetition and transmission), and the key to their genetic history is to be sought in their more general rather than in their later and incidental uses.

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[1] Der Thierische Wille, pp. 282, 283. As cited in Professor James's Psychology, p. 388.

2. The primary roots of instincts reach back to the constitutional properties of protoplasm, and their evolution runs, in general, parallel with organogeny. As the genesis of organs takes its departure from the elementary structure of protoplasm, so does the genesis of instincts proceed from the fundamental functions of protoplasm. Primordial organs and instincts are alike few in number and generally persistent. As an instinct may sometimes run through a whole group of organisms with little or no modification, so may an organ sometimes be carried on through one or more phyla without undergoing much change. The dermal sensillæ of annelids and aquatic vertebrates are an example.

3. Remembering that structural bases are relatively few and permanent as compared with external morphological characters, we can readily understand why, for example, five hundred different species of wild pigeons should all have a few common undifferentiated instincts, such as, drinking without raising the head, the cock's time of incubating from about 10 A.M. to about 4 P.M., etc.

4. Although instincts, like corporeal structures, may be said to have a phylogeny, their manifestation depends upon differentiated organs. We could not, therefore, expect to see phyletic stages repeated in direct ontogenetic development, as are the more fundamental morphological features, according to the biogenetic law. The main reliance in getting at the phyletic history must be comparative study.

5. Instinct precedes intelligence both in ontogeny and phylogeny, and it has furnished all the structural foundations employed by intelligence. In social development also instinct predominates in the earlier, intelligence in the later stages.

6. Since instinct supplied at least the earlier rudiments of brain and nerve, since instinct and mind work with the same mechanisms and in the same channels, and since instinctive action is *gradually* superseded by intelligent action, we are compelled to regard instinct as the actual germ of mind.

7. The automatism, into which habit and intelligence may lapse, seems explicable, in a general way, as due more to the preorganization of instinct than to mechanical repetition. The

habit that becomes automatic, from this point of view, is not an action on the way to becoming an instinct, but action preceded and rendered possible by instinct. Habits appear as the uses of instinct organization which have been learned by experience.

8. The suggestion that intelligence emerges from blind instinct, although nothing new, will appear to some as a complete *reductio ad absurdum*. But evolution points unmistakably to instinct as nascent mind, and we discover no other source of psychogenetic continuity. As far back as we can go in the history of organisms, in the simplest forms of living protoplasm, we find the sensory element along with the other fundamental properties, and this element is the central factor in the evolution of instinct, and it remains the central factor in all higher psychic development. It would be strange if, with this factor remaining one and the same throughout, organizing itself in sense organs of the keenest powers and in the most complex nerve mechanisms known in the animal world — it would be strange, I say, if, with such continuity on the side of structure, there should be discontinuity in the psychic activities. Such discontinuity would be nothing less than the negation of evolution.

9. We are apt to contrast the extremes of instinct and intelligence — to emphasize the blindness and inflexibility of the one and the consciousness and freedom of the other. It is like contrasting the extremes of light and dark and forgetting all the transitional degrees of twilight. In so doing we make the hiatus so wide that derivation of one extreme from the other seems about as hopeless as the evolution of something from nothing. That is the last pit of self-confounding philosophy.

Instinct is blind; so is the highest human wisdom blind. The distinction is one of degree. There is no absolute blindness on the one side, and no absolute wisdom on the other. Instinct is a dim sphere of light, but its dimness and outer boundary are certainly variable; intelligence is only the same dimness improved in various degrees.

When we say instinct is blind, we really mean nothing more than that *it is blind to certain utilities* which we can see. But we ourselves are born blind to these utilities, and only discover them after a period of experience and education. The discovery may seem to be instantaneous, but really it is a matter of growth and development, the earlier stages of which consciousness does not reveal.

Blindness to the utilities of action no more implies unconsciousness in animals than in man. It is the worst form of anthropomorphism to claim that animal automatism is devoid of consciousness, for the claim rests on nothing but the assumption that there are no degrees of consciousness below the human. If human organization is of animal origin, then the presumption would be in favor of the same origin for consciousness and intelligence. Automatism could not exclude every degree of consciousness without excluding every form of organic adaptation.

10. The clock-like regularity and inflexibility of instinct, like the once popular notion of the "fixity" of species, have been greatly exaggerated. They imply nothing more than a low degree of variability under normal conditions. Discrimination and choice cannot be wholly excluded in every degree, even in the most rigid uniformity of instinctive action. Close study and experiment with the most machine-like instincts always reveal some degree of adaptability to new conditions. This was made clear by Darwin's studies on instincts, and it has been demonstrated over and over again by later investigators, and by none more thoroughly than by the Peckhams in the case of spiders and wasps.[1] Intelligence implies varying degrees of freedom of choice, but never complete emancipation from automatism. The fundamental identity of instincts and intelligence is shown in *their dependence upon the same structural mechanisms and in their responsive adaptability.*

INSTINCT AND INTELLIGENCE.

In order to see how instinctive action may graduate into intelligent action it is well to study closely animals in which the instincts have attained a high degree of complexity, and

[1] Wisconsin Geological and Natural History Survey, Bulletin No. 2, 1898.

in which there can be no doubt about the automatic character of the activities. These conditions are perfectly fulfilled in the pigeons, a group in which we have the further advantage that wild and domestic species can be studied comparatively.

It is quite certain that pigeons are totally blind to the meanings which we discover in incubation. They follow the impulse to sit without a thought of consequences; and no matter how many times the act has been performed, no idea of young pigeons ever enters into the act.[1] They sit because they feel like it, begin when they feel impelled to do so, and stop when the feeling is satisfied. Their time is generally correct, but they measure it as blindly as a child measures its hours of sleep. A bird that sits after failing to lay an egg, or after its eggs have been removed, is not acting from "expectation," but because she finds it agreeable to do so and disagreeable not to do so. The same holds true of the feeding instinct. The young are not fed from any desire to do them any good, but solely for the relief of the parent. The evidence on this point cannot be given here, but I believe it is conclusive.

But if all this be true, where does the graduation towards intelligence manifest itself. Certainly not in a comprehension of utilities which are discoverable only by human intelligence. Whatever the pigeon instinct-mind contains, it is safe to say that the intelligence is hardly more than a grain hidden in bushels of instinct, and one may search more than a day and not find it.

a. *Experiment with Pigeons.*

Among many tests, take the simple one of removing the eggs to one side of the nest, leaving them in full sight and within a few inches of the bird on the nest. The bird sees the uncovered eggs, but shows no interest in them; she keeps

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[1] Professor James, Psychology, II, p. 390, thinks such an idea may arise and that it may encourage the bird to sit. "*Every instinctive act in an animal with memory*," says James, "*must cease to be 'blind' after being once repeated.*" That must depend on the kind of memory the animal has. It is possible to have memory of a certain kind in some things, while having absolutely none of any kind in other things. That is the case in pigeons, as I feel very sure.

her position, if she is a tame bird, and after some moments begins to act as if the current of her feelings had been slightly disturbed. At the most she only acts as if a little puzzled, as if she realized dimly a change in feeling. She is accustomed to the eggs, and now misses them, or, rather, misses something, she knows not what. Although she does not know or show any care for the eggs out of the nest, she does appear to sense a difference between having and not having.

There is, then, something akin to memory and discrimination, and little as this implies it cannot mean less than some faint adumbration of intelligence. Now this inkling of intelligence, or, if you prefer, this nadir of stupidity, so remote from the zenith of intelligence, is not something independent of and foreign to instinct. It is instinct itself just moved by a ripple of change in the environment. The usual adjustment is slightly disturbed, and a little confusion in the currents of feeling arises, which manifests itself in quasi-mental perplexity. That is about as near as I can get to the contents of the pigeon mind without being able, by a sort of metempsychosis suggested by Bonnet, to live some time in the head of the bird.

In this feeble perplexity of the pigeon's instinct-mind, in this "nethermost abyss" of stupidity, there is a glimmer of light, and nature's least is always suggestive of more. The pigeon has no hope of graduating into a *homo sapiens*, but her little light may flicker a little higher, and all we need to know is, how instinct behavior can take one step toward mind behavior. This is the dark point on which I have nothing really new to offer, although I hope not to make it darker.

b. *The Step from Instinct to Intelligence.*

Some notion of what is involved in the step may be gathered by comparing wild with semi-domesticated and fully domesticated species. These grades differ from each other in respects that are highly suggestive. In the wild species the instincts are kept up to the higher degrees of rigid invariability, while in species under domestication they are reduced to various degrees of flexibility, and there is a correspondingly greater freedom of action, with, of course, greater liability to irregularities and so-called "faults." These faults of instinct, so far from indicating psychical retrogression, are, I believe, the first signs of greater plasticity in the congenital coördinations and, consequently, of greater facility in forming those new combinations implied in choice of action.

If we place the three grades of pigeons under the same conditions and test each in turn in precisely the same way, we can best see how domestication lets down the bars to choice and at the same time gives more opportunities for free action. The simplest experiment is always the best. Let us take three species at the time of incubation and repeat with each the experiment of removing the eggs to a distance of two inches outside the edge of the nest. The three grades are well represented in the wild passenger pigeon (*Ectopistes*), the little ring-neck (*Turtur risorius*), and the common dovecot pigeon (*Columba livia domestica*). The results will not, of course, always be the same, but the average will be about as follows:

1. *The Passenger Pigeon.* — The passenger pigeon leaves the nest when approached, but returns soon after you leave. On returning she looks at the nest, steps into it, and sits down as if nothing had happened. She soon finds out, not by sight, but by feeling, that something is missing. Her instinct is keenly attuned and she acts quite promptly, leaving the nest after a few minutes without heeding the egg. The conduct varies relatively little in different individuals.

2. *The Ring-neck Pigeon.* — The ring-neck is tame and sits on while you remove the eggs. After a few moments she moves a little and perhaps puts her head down, as if to feel the missing eggs with her beak. Then she may glance at the eggs and appear as if half consciously recognizing them, but make no move to replace them, and after ten to twenty minutes or more leave the nest with a contented air, as if her duty were done; or, she may stretch her neck toward the eggs and try to roll one back into the nest. If she succeeds in recovering *one*, she is satisfied and again sinks into her

usual restful state, with no further concern for the second egg. The conduct varies considerably with different individuals.

3. *The Dovecot Pigeon.* — The dovecot pigeon behaves in a similar way, but will generally try to get *both* eggs back; and, failing in this, she resigns the nest with more hesitation than does the ring-neck.

4. *Results Considered.* — The passenger pigeon's instinct is wound up to a high point of uniformity and promptness, and her conduct is almost too blindly regular to be credited even with that stupidity which implies a grain of intelligence. The ring-neck's stupidity is satisfied with one egg. The dovecot pigeon's stupidity may claim both eggs, but it is not always up to that mark.

In these three grades the advance is from extreme blind uniformity of action, with little or no choice, to a stage of less rigid uniformity, with the least bit of perplexity and a very feeble, uncertain, dreamy sense of sameness between eggs *in* and eggs *out* of the nest, which prompts the action of rolling the eggs back into the nest. That is the instinctive way of placing the eggs when in the nest, and the neck is only a little further extended in drawing the eggs in from the outside. How very narrow is the difference between the ordinary and the extraordinary act! How little does the pendulum of normal action have to swing beyond its usual limit![1]

But this little is in a forward direction, and we are in no doubt as to the general character of the changes and the modifying influences through which it has been made possible. Under conditions of domestication the action of natural selection has been relaxed, with the result that the rigor of instinctive coördinations which bars alternative action is more or less

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[1] We come to equally surprising results in many different ways. Change the position of the nest-box of the ring-neck, without otherwise disturbing bird, nest, or contents, and the birds will have great difficulty in recognizing their nest, for they know it only as something in a definite position in a fixed environment. If a pair of these birds have a nest in a cage, and the cage be moved from one room to another, or even a few feet from its original position in the same room, the nest ceases to be the same thing to them, and they walk over the eggs or young as if completely devoid of any acquaintance with or interest in them. Return the cage to its original place and the birds know the nest and return to it at once.

reduced. Not only is the door to choice thus unlocked, but more varied opportunities and provocations arise, and thus the internal mechanism and the external conditions and stimuli work both in the same direction to favor greater freedom of action.

When choice thus enters, no new factor is introduced. There is greater plasticity within and more provocation without, and hence the same bird, without the addition or loss of a single nerve-cell, becomes capable of higher action and is encouraged and even constrained by circumstances to *learn* to use its privilege of choice.

Choice, as I conceive, is not introduced as a little deity, encapsuled in the brain. Instinct has supplied the teleological mechanism, and stimulus must continue to set it in motion. But increased plasticity invites greater interaction of stimuli and gives more even chances for conflicting impulses. Choice runs on blindly at first, and ceases to be blind only in proportion as the animal learns through nature's system of compulsory education. The teleological alternatives are organically provided; one is taken and fails to give satisfaction; another is tried and gives contentment. This little freedom is the dawning grace of a new dispensation, in which education by experience comes in as an amelioration of the law of elimination. This slight amenability to natural educational influences cannot, of course, work any great miracles of transformation in a pigeon's brain; but it shows the way to the open door of a freer commerce with the eternal world, through which a brain with richer instinctive endowments might rise to higher achievement.

The conditions of amelioration under domestication do not differ in kind from those presented in nature. Domestication merely bunches nature's opportunities and thus concentrates results in forms accessible to observation. Natural conditions are certainly working in the same direction, only more slowly. The direction and the method of progress must, in the nature of things, remain essentially the same.

Nature works to the same ends as intelligence, and to the natural course of events I should look for just such results as

Lloyd Morgan[1] so clearly pictures and ascribes to intelligence. "Suppose," says Mr. Morgan, "the modifications are of various kinds and in various directions, and that, associated with the instinctive activity, a tendency to modify it *indefinitely* be inherited. Under such circumstances *intelligence would have a tendency to break up and render plastic a previously stereotyped instinct.* For the instinctive character of the activities is maintained through the constancy and uniformity of their performance. But if the normal activities were thus caused to vary in different directions in different individuals, the offspring arising from the union of these differing individuals would not inherit the instinct in the same purity. The instincts would be imperfect, and there would be an inherited tendency to vary. *And this, if continued, would tend to convert what had been a stereotyped instinct into innate capacity; that is, a general tendency to certain activities (mental or bodily), the exact form and direction of which are not fixed, until by training, from imitation or through the guidance of individual intelligence, it became habitual. Thus it may be that it has come about that man, with his enormous store of innate capacity, has so small a number of stereotyped instincts.*"

The following from Professor James[2] is suggestive:

"Nature implants contrary impulses to act on many classes of things, and leaves it to slight alterations in the conditions of the individual case to decide which impulse shall carry the day. Thus, greediness and suspicion, curiosity and timidity, coyness and desire, bashfulness and vanity, sociability and pugnacity seem to shoot over into each other as quickly, and to remain in as unstable equilibrium, in the higher birds and mammals as in man. They are all impulses, congenital, blind at first, and productive of motor reactions of a rigorously determinate sort. Each one of them, then, is an instinct, as instinct is commonly defined. *But they contradict each other; experience, in each particular opportunity of application, usually deciding the issue. The animal that exhibits them loses the 'instinctive' demeanor and appears to lead a life of hesitation and choice, an intellec-*

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[1] Animal Life and Intelligence, pp. 452, 453.

[2] Psychology, II, pp. 392, 393.

tual life; not, however, because he has no instincts — rather because he has so many that they block each other's path."

Looking only to the more salient points of direction and method in nature's advance towards intelligence, the general course of events may be briefly adumbrated. Organic mechanisms capable of doing teleological work through blindly determined adjustments, reproduced congenitally, and carried to various degrees of complexity and inflexibility of action, were first evolved. With the organization of instinctive propensities, liable to antagonistic stimulation, came both the possibility and the provocation to choice. In the absence of intelligent motive, choice would stand for the outcome of conflicting impulses. The power of blind choice could be transmitted, and that is what man himself begins with.

Superiority in instinct endowments and concurring advantages of environment would tend to liberate the possessors from the severities of natural selection; and thus nature, like domestication, would furnish conditions inviting to greater freedom of action, and with the same result, namely, that the instincts would become more plastic and tractable. Plasticity of instinct is not intelligence, but it is the open door through which the great educator, experience, comes in and works every wonder of intelligence.

Spencer[1] has shown clearly that this plasticity must inevitably result from the progressive complication of the instincts. "*That progressive complication of the instincts,*" he says, "*which, as we have found, involves a progressive diminution of their purely automatic character, likewise involves a simultaneous commencement of memory and reason.*"

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[1] Psychology, I, pp. 443 and 454, 455.

IN MEMORIAM.

JAMES INGRAHAM PECK.

Professor Peck died of pneumonia on Friday, Nov. 4, 1898, after a brief illness.

He had lived thirty-five years, and had accomplished so much in making his character fine and strong, in serving others as a rarely successful teacher, in winning results as an investigator, and in brightening and encouraging the lives of many others by his faith and his loyal friendship, that the sense of loss is deeply and widely felt.

He is remembered in his college days as one of the earnest men who was seeking knowledge, not prizes nor marks. He was an untiring and eager worker, but withal so interested in all the doings of his class and the various general interests of the student, that he was never looked upon as a "dig." He was too genial, too good a comrade, too sympathetic with all that was bright and wholesome, for that.

While I recall long evenings of work with him in the laboratory, frequently lasting until midnight and after, others remember him for his work in athletics, as artist of his class, and as generally one of the best and truest fellows in 1887.

He was graduated from Williams College with high rank and spent the following year there as Assistant in Biology. He began his original work that year by a study of the "Variation of the Spinal Nerves in the Caudal Region of the Domestic Pigeon." He showed at once the true scientific spirit. He was so eager and determined to know the truth that no perplexing details could overcome his patience, nothing could discourage his perseverance and enthusiasm. He must have the truth, and no price was too great to give for it.

In the following year, 1888–89, at the Johns Hopkins University as a graduate student, he came under the influence and special guidance of Professor Brooks. The opportunities for development that were there provided him were met in the same eager way so characteristic of him. He appreciated this training keenly, and often said that nothing could have been more enjoyable and more successful in developing his powers than the wisely ordered work so sympathetically directed, which he carried on at the Johns Hopkins in the two years of his life there.

Among the results of his university work are two published papers, one a report on Pteropods and Heteropods, and one on the "Anatomy and Histology of Cymbuliopsis calceola."

In 1889 he was appointed Biologist to the Boston Water Works and had begun a careful system of investigation of the water supplied to the city of Boston, when failing health made it necessary for him to give up his work for a long rest. With health recovered after two years of farm life, he accepted a call to become Assistant in Biology at Williams College. He received the degree of Ph.D. there in 1893, and in the following year he was made Assistant Professor.

For several years Professor Peck was connected with the United States Fish Commission, at first as Assistant and later in charge of the laboratory of the Commission at Wood's Holl. He developed a line of work there concerning the food of the Menhaden, and the distribution of the food of certain marine fishes, which led him to results quite unexpected and of far-reaching importance. Further opportunity for work of this nature was offered him by observations and collections made by Mr. N. R. Harrington in Puget's Sound. The results of this work were studied and a report upon them by J. I. Peck and N. R. Harrington was published in the *Transactions* of the New York Academy of Science in 1898.

It was Professor Peck's hope that he might sometime have a chance to extend his observations on the Atlantic coast outward to and across the Gulf Stream and also into greater depth.

In 1896 the position of Assistant Director of the Marine Biological Laboratory at Wood's Holl was offered him and he accepted it. During the three sessions of 1896, 1897, and 1898, Professor Peck devoted himself to the interests and the development of the laboratory, taking the supervision of the class in Animal Morphology. The course of study had to be revised and a new staff of teachers brought in and trained to the work. Professor Peck met all the difficulties of the new situation with remarkable energy, perseverance, and tact. Teachers and students alike caught the spirit of his enthusiasm and zeal, and every day's work was carefully planned in a teachers' conference the evening before. The example of such whole-souled devotion to the work will not be soon forgotten.

I have already referred to the scientific character of his mind, and there was no lack of what that should include — the unprejudiced, reasonable position, at all times, toward all views. He was a genial, loyal friend, a man of high ideals, of great courage and strong faith, and especially a man of full and ready sympathy. I never knew a teacher who gave himself so generously, without measure, at all times to all his students, as he did. They honored and loved him in an unusual degree, and gladly did for him their best. It is said in the college that very few have ever obtained so deep a hold upon the students and have been such a power for good here as Professor Peck.

We shall sadly miss him in the world of science, but the memory of his true, brave, eager spirit, with his great love for truth and his ready sympathy toward all, will be an uplifting and an abiding force to all who knew him.

SAMUEL F. CLARKE.

List of Dr. Peck's publications:

1888: A Report on the Pteropods and Heteropods Collected by the Steamer *Albatross* on a Voyage from New York to San Francisco.
1889: Variation in the Nerves of the Caudal Region of the Domestic Pigeon.
1890: Anatomy and Histology of Cymbuliopsis calceola.
1893: On the Food of the Menhaden.
1895: Sources of Marine Food.

J. I. PECK AND N. R. HARRINGTON.

1898: Observations on the Plankton of Puget Sound.

JAMES ELLIS HUMPHREY

A SKETCH of Dr. Humphrey's life and the minutes of a memorial meeting are given in the Johns Hopkins University Circular of November, 1897. From this source the following extract embodies the sentiments of Professor Humphrey's colleagues at the Marine Biological Laboratory:

"Professor Humphrey's value in the department of instruction under his charge in this university increased rapidly and steadily during his three years of service, until he became the essential factor in its success. Pupils and colleagues soon learned to draw upon his well-ordered store of knowledge and to trust his judgment, which ever proved sound and accurate. His boundless patience and care as teacher gave assurance and encouragement to his pupils, who soon learned to seek his advice in well-founded confidence of ready and willing response. Nothing which promised to be helpful to others was a trouble. Few have exemplified better than he the beauty of orderliness. Books, papers, instruments, and his store of knowledge were all at his ready command, and nothing with which one had to do, either of facts or things tangible, was ever out of place.

"He was a ready writer and a well-known contributor to botanical literature, and all his work is marked by minute care, as well as by broad culture and great power for original research and reflection, for he was broad and liberal in all his thoughts and in all his dealings, and nothing that he undertook was left without new additions with permanent scientific value.

"To overwork, and a consequent lowered state of health, may probably be attributed the lamentable catastrophe of his death. In spite of warning and against the wishes of one whose pleasure and comfort were always of supremest moment, he went because the duty of going seemed too plain to resist, and then he fell ; — a loss to the university which the memory of his character and example lightens but little ; a loss to his more intimate friends which nothing but the hand of unfeeling time will remove ; a bereavement to those yet nearer to him upon which we must not intrude, except in sympathy."

The following is a list of Dr. Humphrey's published works :

1886 : On the Anatomy and Development of Agarum Turneri. *Proc. Amer. Acad. A. & S.*, June, 1886, p. 195, 1 pl.

1887 : The Preparation of Agarics for the Herbarium. *Bot. Gaz.*, vol. xii, 1887, p. 271.

1888 : Potato Scab. *Rep. Mass. Ag. Expt. Sta.*, 1888, p. 131, 1 pl.

1889 : Mildews. *Mass. Hort. Soc.*, 1889, pp. 1–11.

—— Fungous Diseases of Plants. *Bull. No. 6, Mass. Ag. Expt. Sta.*, Oct. 1889. p. 9.

—— A General Account of the Fungi. *Mass. Ag. Expt. Sta. Rep.*, 1889, pp. 195–230.

1890 : Notes on Technique. *Bot. Gaz.*, vol. xv, 1890, p. 168.

—— Black Knot of Plum, Mildew of Cucumber, etc. *Rep. Mass. Ag. Expt. Sta.*, 1890, p. 200, 2 pl.

1891 : Treatment of Fungous Diseases. *Bull. No. 39, Mass. Ag. Expt. Sta.*, April, 1891, pp. 1–12, 5 figs.

—— Protoplasmic Physics. *Amer. Nat.*, vol. xxv, 1891, p. 376.

—— Comparative Morphology of the Fungi. *Amer. Nat.*, vol. xxv, 1891, p. 1055.

—— Notes on Technique. *Bot. Gaz.*, vol. xvi, 1891, pp. 71–73.

—— Some Diseases of Lettuce, etc. *Bull. No. 40, Mass. Ag. Expt. Sta.*, 1891.

—— The Rotting of Lettuce. *Rep. Mass. Ag. Expt. Sta.*, 1891, pp. 218–248, 1 pl.

1892 : Amherst Trees. *Amherst, Mass.*, 1892, pp. 1–78.

—— Fungous Diseases and Their Remedies. *Mass. Hort. Soc.*, Jan. 1892, p. 1.

—— The Saprolegniaceae of the United States. *Amer. Phil. Soc.*, Nov. 1892, p. 63, 7 pl.

—— Rep. of Mycologist. *Mass. Ag. Expt. Sta.*, 1892, p. 1, 5 pl.

1893: On Monilia Fructigena. *Bot. Gaz.*, vol. xviii, 1893, p. 85, 1 pl.
—— Botanical Microtechnique (from the German by Zimmermann). New York, 1893, pp. 1–296.
1894: Where Bananas Grow. *Pop. Sci. Month.*, vol. xliv, 1894, p. 486.
—— Neucleolen und Centrosomen. *Berich. der Deutschen Bot. Gesell., Bd.* xii, 1894, p. 108, 1 pl.
—— Nucleoli and Centrosomes. *Ann. of Bot.*, vol. viii, 1894, p. 373.
—— Eduard Strasburger. *Bot. Gaz.*, vol. xix, 1894, p. 401, 1 pl.
1895: Some Recent Cell Literature. *Bot. Gaz.*, vol. xx, 1895, p. 222.
—— Some Constituents of the Cell. *Ann. of Bot.*, vol. ix, 1895, p. 561, 1 pl.
1896: Botany and Botanists in New England. *New England Mag.*, vol. xiv, 1896, p. 27, 2 pl.
—— On the Development of the Seed in the Scitamineae. *Ann. of Bot.*, vol. x, 1896, p. 1, 4 pl.
—— Some Modern Views of the Cell. *Pop. Sci. Month.*, vol. xlix, 1896, p. 603.
—— A University Celebration in Germany. *Amer. Univ. Mag.*, Nov. 1896.

Dr. Humphrey was an associate editor of the *Zeitschrift für Pflanzenkrankheiten*, a frequent contributor of notes on American botany to the *Botanisches Centralblatt*, and of specimens to the *Phycotheca Boreali Americani* of Collins, Holden, and Setchell.

WESLEY WALKER NORMAN.

As this volume of Biological Lectures was going to print, Dr. W. W. Norman, one of the contributors, died of typhoid fever. He had been for a long time a firm friend of, and zealous laborer in, the Marine Biological Laboratory. Although already suffering, he had come from Texas to take up his work as an instructor in the laboratory during the summer of 1899. He died at the Massachusetts General Hospital, July 2, in his thirty-fourth year. A biographical notice will appear in the next volume of these Lectures.

Notices of Drs. Arnold Graf and Nathaniel Russel Harrington must also be deferred.