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OBSERVATIONS ON THE GEMMULE AND EGG DEVELOPMENT OF MARINE SPONGES.

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TABLE OF CONTENTS.

PAGE

INTRODUCTION 278

I. ADULT STRUCTURE AND GEMMULE DEVELOPMENT OF ESPERELLA FIBREXILIS, N. SP.

1. ADULT 279

Embryological Methods 285

2. FORMATION OF GEMMULES 287

3. DEVELOPMENT OF GEMMULE INTO SWIMMING LARVA 294

4. METAMORPHOSIS 298

Ectoderm 301

Subdermal cavities and canals 305

Dermal membrane 307

Efferent canals and oscula 308

Afferent canals and pores 309

Flagellated chambers 310

Spicules 314

Summary of the leading facts in the gemmule development of Esperella 315

Previous knowledge of the development of Esperia (Esperella) .. 316

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PAGE

II. ADULT STRUCTURE AND GEMMULE DEVELOPMENT OF TEDANIA BRUCEI, N. SP.
1. ADULT ... 320
Collecting and Embryological Methods ... 325
2. DEVELOPMENT OF THE SWIMMING LARVA ... 327
Formation of gemmules ... 327
Development of gemmule into swimming larva ... 329
Structure of the swimming larva ... 331
3. METAMORPHOSIS ... 332
Attachment ... 332
Ectodermal membrane and peripheral mesodermic zone ... 335
Canal system ... 337

III. ADULT STRUCTURE AND EGG DEVELOPMENT OF TEDANIONE FOETIDA, N. G.
1. ADULT ... 338
2. DEVELOPMENT ... 341

IV. EARLY STAGES IN EGG DEVELOPMENT OF HIRCINIA ACUTA ... 346

V. REMARKS ON THE MORPHOLOGY OF SPONGES ... 348
Evidence from comparative anatomy as to sponge phylogeny ... 350
Embryological evidence as to sponge phylogeny ... 357
Origin of the Olynthus ... 362

VI. REMARKS ON THE GEMMULE DEVELOPMENT OF SPONGES.
1. ASEXUAL DEVELOPMENT IN GENERAL OF THE SPONGES ... 364
2. COMPARISON BETWEEN THE EGG LARVA AND GEMMULE LARVA OF SILICIOUS SPONGES ... 368
Resemblance between the two kinds of larvae ... 368
Cause of the resemblance ... 370

THE observations described in the present paper were begun more than four years ago, when as Bruce Fellow of the Johns Hopkins University I was permitted by the directors of the fellowship to spend a considerable part of the academic year in the Bahama islands. For their kindness in complying with my wishes in this matter I thank them heartily, and trust they may find in the following pages some justification of that pleasant excursion.

The investigation has been subject to many interruptions. On returning from the Bahamas it was prosecuted for a time in Professor Brooks's laboratory, but other duties incident to my connection with the U.S. Fish Commission interfering, it was laid aside until midsummer, 1890. The Commissioner, Hon. Marshall McDonald, feeling that any enlightened attempts

to be made in the future in the direction of the cultivation of useful sponges would be greatly aided by a knowledge of the life-histories of sponges in general, approved of my wish to continue the investigation, which was made the more attractive by the discovery in a sponge,[1] common about Woods Holl, Mass., of gemmules essentially like those I had already found in a Bahama form. The work was accordingly carried on in the Fish Commission Laboratory at Woods Holl until the fall of 1891, by which time my observations were finished. Realizing that the completed paper would be slow in appearing, I published in the JOURNAL OF MORPHOLOGY (Vol. V, No. 3, 1891) a brief account of the more important results.

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I. ADULT STRUCTURE AND GEMMULE DEVELOPMENT OF ESPERELLA FIBREXILIS, N. SP.

1. ADULT.

Esperella fibrexilis is small, the masses usually having a greatest diameter of 4 or 5 inches. It may assume any shape, sometimes appearing as a flat incrustation, and again as a spheroidal mass. Quite commonly its upper surface forms conical processes, often acute and very ragged. And with these there may be combined irregular ridges with sharply cut edges, as in the sponge shown in Fig. 1. It is rare to find this sponge moderately clean, it being nearly always covered with hydroids, polyzoa, and especially a cylindrical alga, all of which are firmly rooted in the body. The feeble development of the skeleton more than anything else marks it off from the known members of this genus.

Diagnosis. Esperella fibrexilis, n. sp. — Sponge amorphous, yellowish-brown, and of slight consistency. Dermal membrane containing no spicules or almost none, everywhere separated from subjacent tissues by subdermal cavities, and everywhere

[1] For calling my attention to this interesting sponge (Esperella fibrexilis) I am indebted to Prof. T. H. Morgan.

perforated by closely set pores. Surface appears porous to the eye, owing to abundance of comparatively deep subdermal cavities, which when magnified appear as pore-riddled areas. Between such areas the pores are inconspicuous. Oscula fairly abundant and small, often leading into wide shallow spaces, covered with imperforate membrane. Sponge body consists of network of trabeculae, containing single rows of closely set flagellated chambers. Spicules include smooth oxytylotes of about $\frac{2}{1}\frac{5}{0}\frac{}{0}$ mm. long, toxaspires, sigmaspires, and sigmas $\frac{1}{1}\frac{0}{0}\frac{}{0}$ mm. long, large shovels $\frac{6}{1}\frac{}{0}\frac{}{0}$ mm. and small shovels $\frac{3}{1}\frac{}{0}\frac{}{0}$ mm. long. The small shovels abundant, but large ones rare; sigmas and sigmaspires abundant, toxaspires less so. Oxytylotes scattered irregularly through the mesoderm, not united into a meshwork. In peripheral region the oxytylotes form radial bundles, which divide into brushes supporting the dermal membrane. In the body of sponge, spicular bundles are few in number and without order in their arrangement. *Wharf piles, Woods Holl, Mass.*

The body of the sponge consists of a network of narrow trabeculae, separated by a system of canals, Pl. XIV, Fig. 2. Esperella is one of those sponges in which there is no symmetrical arrangement of parts, and which offer the greatest difficulty to the solution of the question as to what constitutes the typical structure of a sponge individual. For in such a sponge neither the oscula, pores, nor canals, are arranged in such a way as to indicate the division of the body into regions which could be compared with one another, and so be taken as the representatives of individuals.

The canal system belongs to Vosmaer's third type. The distinguishing features of the canal system are that all the canals, afferent as well as efferent, are so wide and spacious. Each chamber has not a special afferent canal of its own, but many chambers are grouped round a single comparatively wide afferent canal into which they open directly. Similarly there is no special efferent canal for each chamber, but many chambers open directly into a wide canal.

The development of numerous wide and for the most part comparatively shallow, afferent canals directly beneath the skin

(the so-called subdermal cavities, *s.d.c.*, Pl. XIV, Fig. 2), separates more or less completely the superficial layer of the sponge from the rest of the body. This superficial layer (ectoderm + thin layer of mesoderm) is known as the dermal membrane, *d. mem.*, Fig. 2. The dermal membrane is bound to the subjacent part of the body by the mesoderm lying between the subdermal cavities. The cavities are so numerous that this mesoderm takes the form of irregular beams or trabeculae, often being nothing more than slender cords, as in Fig. 4, *mes. b.* The dermal membrane may easily be pulled off, and is then found to be everywhere perforated by closely set pores, Fig. 4 (surface view of dermal membrane, from below). The subdermal cavities into which the pores open are of two kinds, of which the larger and deeper appear as pore areas (often rounded) on the surface. These pore areas are conspicuous when the surface of the sponge is examined with a lens, and the dermal membrane between them seems at first sight not to possess any pores (Fig. 5, view of a small area of the surface). This is owing to the fact that the second kind of subdermal cavity, underlying the apparently aporous portion of the dermal membrane, is very small and shallow. That the dermal membrane does possess pores between the pore areas can easily be shown by scraping the membrane free of the subjacent tissue, when the pores at once come into view (region *a* in Fig. 5). Owing to the great number of pore areas the surface of the sponge acquires a characteristically porous appearance. When examined under a low objective or magnifying glass the surface is further diversified by an irregular meshwork of dark bands, which represent the coarser mesodermic trabeculae connecting the dermal membrane with the body of the sponge (Fig. 5).

The water passing through the pores enters the subdermal cavities, *s. d. c.*, Fig. 2, whence it may pass by afferent canals, *a.f. c.*, to the flagellated chambers. The flagellated chambers are arranged along the sides of the afferent canals, and open directly into them. At their opposite pole the chambers open in the same way into efferent canals (*ef. c.*). Both afferent and efferent canals are relatively large. There is no great difference in size between the afferent canals

and the smaller efferent, and in the body of the sponge it is not practicable to distinguish them. This arrangement of the canal system brings it about that the sponge body is cut up into narrow trabeculae, in which flagellated chambers are arranged in a single layer.

The larger efferent canals are distinguishable without any trouble (*ef. c.*, Fig. 2). They unite with one another and very often open into spacious cavities, just underneath the dermal membrane (here aporous), which are precisely like the subdermal cavities, only much larger. The membrane covering these oscular cavities is perforated by the oscular opening itself, which thus differs from a pore only in size. In Pl. XIV, Fig. 6, a portion of the surface is represented showing an oscular cavity, *os. c.*, with an osculum, *os.* Looking through the osculum, two efferent canals are shown, *ef. c.* Surrounding the oscular cavity are numbers of the conspicuous subdermal cavities, *s. d. c.* Other and much smaller oscula are found, two of which are shown in Fig. 7, *os.* Such oscula seem to be nothing in the world but the openings of certain subdermal cavities from which the covering pore-membrane has disappeared. The canals into which such oscula open, branch very quickly, as is shown in Fig. 7. The oscula taken together are few in number, and are distributed over the surface with entire irregularity. They are not seated on elevations, and are inconspicuous. Nothing in their nature or surroundings could of itself warrant one in regarding them as a different type of structure from the pores.

The flagellated chambers, *f. c.*, Fig. 2, are spheroidal, and in the normal regions (those unaffected by the formation of gemmules) are closely set. The collared cells have small cell bodies, which stain but slightly, but have very characteristic deeply staining nuclei. The mesoderm in the normal trabeculae is rather scanty. It consists of cells of many shapes and sizes, which however pass one into the other by slight gradations. As common a type as any is the rounded or amoeboid cell with a well-staining body, Pls. XIV and XV, Figs. 8 and 9. The body varies greatly in size, and these cells pass by insensible gradations into delicate spindle-shaped cells in which the body

scarcely stains at all and consists of a mere coating for the nucleus, prolonged at opposite ends into slender protoplasmic processes. Such cells are especially abundant in the mesoderm of the dermal membrane, Fig. 4. The edges of pores and oscula are always provided with such cells, which here are especially long and fibre-like, and serve as a support for the free edge. The mesodermic bands which support the dermal membrane (Fig. 4), and the partition walls found in branching canals (*p. w.* Fig. 7) contain great numbers of these fibre-like cells. The ectoderm and the epithelioid lining of the canals are formed of flat cells.

The disposition of the peripheral skeletal bundles is shown in Fig. 2. The bundles are composed of spicules, such as that shown in Fig. 3 *a* (oxytylotes, Sollas), which project slightly from the surface of the dermal membrane, Fig. 2. Besides forming bundles, the oxytylotes are scattered in abundance through the mesoderm, but are not united into a meshwork. A variation from the ordinary type of spicule is occasionally found, with a head like that shown in Fig. 3 *b*. After boiling in caustic potash, some spicules are always found with the pointed end split as in Fig. 3 *c*, doubtless an effect due to the action of caustic potash. The bow-shaped spicules, Fig. 3 *d* (toxaspires, Sollas), the s-shaped spicules, Fig. 3 *f* (sigma-spires), and the sigmas, Fig. 3 *e*, are all of about the same size, the first form being less abundant than the other two. They are found scattered about in the mesoderm in all parts of the sponge.

The large shovel-shaped spicules, of which a face view is given in Fig. 3″ *a*, and a side view in Fig. 3″ *b*, are rare. Shovels of about half the size, of which Fig. 3′ *b* gives a face view and Fig. 3′ *c* a side view, are comparatively abundant. When the spicule is viewed more or less from the end, Fig. 3′ *a*, it is seen that the shovel shape is an illusion, that the blade of the shovel is not flat, but is a figure of three dimensions. If an oval body should be divided by a transverse plane, passing through a point on the equator and a point on the opposite surface somewhat nearer one of the poles, two parts would be obtained, of which the smaller would roughly correspond in

shape with the blade of the shovel. That this is the shape of the shovel blade is gathered from a comparison of the figures *a*, *b*, *c*, Fig. 3′, the difference lying in the fact that the transverse diameter of the blade *ll* is greater than the dorso-ventral diameter, *d. v.* The wall of the oval is thickened all round (compare *b* and *c*), the thickening being greatest near the apex and gradually decreasing towards the equator. A special thickening produces a tooth *t* on the ventral surface of the blade. The handle of the shovel is directly continuous with the dorsal surface of the blade, and at its apex is divided into three small sharp-pointed lobes, one ventral, two lateral. Now a shovel-shaped spicule of this sort is developed from a small sigma such as *d*, Fig. 3′. Small sigmas like this and bow-shaped spicules are the only microscleres (smaller spicules) found in the ciliated larva and in the recently attached sponge. The small sigmas are also present in the mesoderm of the adult, and transitional forms between them and the shovels are found. The sigma appears to develop into the shovel in this way. It increases in length and one of the ventral arms, *v′* in *d*, becomes relatively long while the other *v″* grows shorter. The longer arm and the axis back of it then flatten out, and grow in such a way as to become connected at the sides forming the blade of the shovel. The other half of the axis does not flatten, but remains as the handle, the shorter arm of the original sigma persisting as the ventral lobe of the handle apex, the other two lobes being formed as outgrowths. The larger shovels are no doubt derived from the smaller by the production of lateral notches (*l. n.*, Fig. 3″), which divide the continuous blade of the small shovel into a dorsal (*d. l.*, Fig. 3″) and a ventral lobe (*v. l.*, Fig. 3″), the tooth *t* remaining as a thickening of the ventral lobe. The only other observations I am aware of, on the development of this class of spicules, are contained in Ridley & Dendy's Challenger Report on the Monoxonida (21, p. xx). Their account anticipates mine in the chief point, viz. that the chelae are produced by the gradual alteration of sigmas.

The study of the anatomy of such a sponge as Esperella would of itself lead one to homologize pores with oscula, and

efferent with afferent canals. One would also be inclined to believe that the position of the oscula is not determined by any deep-lying (though veiled) division of the sponge body into individuals. The homology between pores and oscula would rest on the absence of any structural difference between them (they differ in size, but the variation in the size of the osculum weakens this argument), and on their similarity in the matter of local surroundings (comp. Figs. 6 and 7 — both pores and oscula open into comparatively shallow, spacious cavities strikingly alike). The homology between the two sorts of canals would rest on their entire similarity — there is no discoverable difference between the subdermal cavities into which the pores open, Fig. 7, and the oscular cavities shown in this figure and in Fig. 6. (The development shows also that in this sponge they are formed in precisely the same way.) As to the basis of the third conclusion, the oscula are distributed with entire irregularity, and the oscular cavities cannot be regarded as so many centers round which the canal system of the sponge groups itself. Rather, it would seem from an examination of such portions of the surface as that shown in Fig. 7, that circumstances may determine the transformation of a pore area into an osculum almost anywhere. The comparative anatomy of sponges in general, however, forces upon us the conviction that forms like this are phylogenetically colonies, even though it be true that new oscula may be formed in an individual independently of any process of budding. And, further, we are driven to believe that phylogenetically, afferent and efferent canals are radically different things, the latter being lined with endoderm, while the former are invaginations from the exterior.

Embryological Methods. — If an Esperella be examined during the summer months, it is found to contain great numbers of embryos imbedded in the mesoderm. When these embryos are studied they are found not to be egg embryos, but gemmules (*i.e.* internal buds). Nevertheless, the gemmules in sponges kept in aquaria escape through the oscula as ciliated larvae, essentially identical in structure with the typical egg larva of silicious sponges. After swimming about for a day or

two the ciliated larvae attach themselves to the wall of the aquarium, and undergo a metamorphosis.

Esperella, being of small size, is easily kept in aquaria, but the larvae, as a rule, do not escape in great numbers. A confinement over night in a simple aquarium of good size, say three gallons, through which no water is passing, will nearly always result in the liberation of some larvae. Instead of trying to aërate the water, it was found more convenient to transfer the larvae with the help of a pipette to fresh dishes of water. As most of them attach in a day or so, it is only necessary to transfer a few times. Once attached to the wall of the dish, the dish itself may be placed in a running aquarium and the little sponges thus kept without further trouble. I could not, however, succeed in getting them to increase much in size, in spite of the aquarium facilities in the Fish Commission laboratory. They seemed willing to live indefinitely, but grow they would not — for lack of proper food, I suppose.

The young sponge after the metamorphosis is so thin (scarcely more than an incrustation) that it cannot be scraped off the dish without injury to it. I, therefore, coated my dishes with a thin layer of paraffine. Collodion was also used. A little piece of the paraffine, or collodion, could then be cut out with the sponge sticking to it, and the whole thing placed in the killing fluid, and subsequently kept in alcohol until ready for use. In many cases the little sponge separates at once from the paraffine in the killing fluid. I satisfied myself, by comparison with larvae scraped from the dish, that the paraffine or collodion did not affect the character of the tissues. To be sure such larvae as remained stuck to the paraffine were only kept in alcohol for a few weeks. If the action of the alcohol on the paraffine be kept up for months, I am not sure but the effect on the tissues of the sponge is injurious.

For fixing purposes I found very much the best fluid was the mixture of acetic acid, alcohol, and osmic acid, recommended by Zacharias (glacial acetic 1 part, absolute alcohol 4 parts, osmic acid few drops). I allowed this to act 10–20 mins. It is excellent for all stages of the development.

Kleinenberg's picric proved itself of use for special points, often preserving the individual cells in a more natural and uncontracted condition than the Zacharias. But in general it dissociated the elements too much. Borax carmine and haematoxylin stained in a very satisfactory way. For macerating purposes Bela Haller's mixture was chiefly used.

2. Formation of Gemmules.

Any portion of the sponge body may develop gemmules. They may be found in the extreme peripheral region, visible under the surface of the uninjured sponge, or may be present in the center of the body. In whatever region they are found they are usually so abundant as to greatly change the structure of the sponge body in that district. In many Esperellas, during the summer, the whole body seems given over to the formation of gemmules. In such individuals gemmules are thickly scattered through every part, and the organization of the entire sponge is seriously interfered with. (This interference, as will be shown later, consists largely in the reduction in number of the flagellated chambers, in the obliteration of many canals, and the rupturing of trabeculae.) In other individuals the gemmules may be extremely abundant in certain portions of the body, while the normal sponge structure is retained elsewhere. The older gemmules and the larvae are easily seen with the naked eye. All gradations of size are found down to minute gemmules consisting of but a few cells. The older gemmules and larvae project into the larger canals, the younger gemmules lie in the trabeculae imbedded in the mesoderm. In the section Pl. XV, Fig. 12, are shown a young larva, *l*, a full sized gemmule, *g*, medium sized gemmule, g', and several little gemmules, g''. In the section Pl. XIV, Fig. 8, four young gemmules of different sizes are shown lying in the mesoderm.

The formation of gemmules in large numbers is associated with a certain degeneration of the normal sponge structure. This is evident when sections through a region in which gemmules are numerous (Pl. XIV, Fig. 8) are compared with sections

through a region in which few or no gemmules appear (Pl. XIV, Fig. 2). In the latter the trabeculae are made up chiefly of rows of flagellated chambers, with but a scanty amount of mesoderm between the chambers. But in the former the flagellated chambers are either absent or are very few in number. The trabeculae in such a region are composed of mesoderm, with gemmules, and a flagellated chamber here and there. What I take to be the remains of degenerated flagellated chambers are scattered about through the mesoderm. Such are the groups of cells, *deg. f. c.*, in Pl. XIV, Fig. 8, and Pl. XV, Fig. 15. The cells composing such groups are quite like the lining cells of the chambers in general appearance, that is, they have a small clear body which stains scarcely at all and the peculiar nucleus of the collared cell. The inference from these data is that where gemmules begin to develop in large numbers, the flagellated chambers of the region degenerate. What becomes of the collared cells I cannot say, but Metschnikoff's observations on the disappearance and reappearance of the flagellated chambers in young spongillas (12) make it probable that these cells are transformed into amoeboid mesoderm cells. Where gemmules are very numerous, the trabeculae themselves are ruptured and broken down in many places. This is the natural result of the compression of the tissues due to the growth of the gemmules, in the course of which many of the neighboring smaller canals are obliterated, and of the liberation of the gemmules. In such spots the sponge body consists of scarcely more than an amorphous aggregate of cells and gemmules, and affords a noticeable appearance of degeneration when compared with the smoothly outlined trabeculae of a non-gemmular district.

Very young gemmules, such as g', Pl. XIV, Fig. 8, and g', Pl. XV, Fig. 9, are composed of a small number of polygonal cells surrounded by a follicle of flattened cells (*g. f.*). I have never found a gemmule surrounded by a follicle to have less than five cells in cross section. The bodies of the gemmule cells are filled with a finely granular yolk, and take the stain well (haematoxylin or carmine). The nuclei are always conspicuous but differ much in appearance, the difference being due, as I think,

to a difference in the stage of division. In young gemmules such as these, and in considerably larger ones as well, the cell outlines are exceedingly plain.

Young gemmules like those just described are formed from groups of mesoderm cells, such as are shown in Pl. XIV, Fig. 8, and Pl. XV, Figs. 13, 14, 15 (*mes. gr.*). The cells composing the mesoderm group are essentially like the gemmule cells. Like the latter they have plump bodies filled with the same finely granular yolk, in consequence of which they stain well, and have conspicuous nuclei. Such groups of mesoderm cells occur in abundance. They have no definite shape and may contain few cells or many, and the component cells may lie together very loosely or be packed pretty closely. They are formed by the migration towards a common point of certain mesoderm cells in which a considerable amount of yolk has been deposited. Such cells are found in abundance lying singly, or in twos and threes through the mesoderm. In Pl. XV, Fig 13, there are several (*g. m. c.*). They do not form a class by themselves, but are merely ordinary mesoderm cells containing a maximum amount of yolk, and are connected by transitional stages, containing less and less of yolk, with the delicate spindle-shaped mesoderm cells, the body of which contains no yolk and scarcely stains at all. The congregation of such cells to form groups may be inferred from such preparations as those shown in Pl. XV, Figs. 13 and 14.

In the transformation of such masses of mesoderm cells as are shown in Fig. 15 (*mes. gr.*) into gemmules, the outer cells must flatten and become the follicle. But I have not succeeded in getting preparations actually showing this. I do not believe the gemmule, when first formed, is of any particular size, for groups of mesoderm cells are met with, differing greatly in this respect. The great number of very small gemmules such as g^1, Fig. 8, and g^1, Fig. 9, make it evident that very frequently gemmules are formed from masses consisting of but a few mesoderm cells, for instance *mes. gr.* in Fig. 8. On the other hand, it seems likely that a mass of cells so rounded as the larger group in Fig. 15 was about to form a single gemmule, which would have been of considerable size.

Bearing in mind the theoretical possibility of a gemmule originating from a single cell, I went to considerable pains in looking for any such indication. I could not convince myself with certainty that a gemmule ever was so formed, though I found cell groups such as *a*, Pl. XV, Figs. 10, 11, 13, looking as though they had been derived from single cells. Such groups though were very rare.

Gemmules increase in size by cell-division. This is inferred at once from the large size of the cells forming the youngest gemmules as compared with the cells of older ones, Fig. 9. No karyokinetic figures were found, but the nucleus appears in several conditions, representing, no doubt, different phases of nuclear division. These different conditions of the nucleus are shown in Fig. 8′ (1, 2, 3, 4, 5), the arrangement of the figures indicating what I take to be the order in which the several phases follow one another. In stage 1, in which are all of the nuclei in the gemmules of Fig. 8, the chromatin forms a solid, usually angular, mass, and the nucleus is small. The mass of chromatin is relatively so large that very often it is difficult to make out the surrounding nuclear membrane, and the nucleus appears to be simply an angular mass of chromatin, as in the larger gemmule of Fig. 9. In what I take to be the second stage the nucleus is larger, and the chromatin forms a tangled skein lying in the center of the nuclear cavity. The smaller gemmule in Fig. 11 has its nuclei in this phase. In the third stage the nucleus is large, and the nucleoplasm very conspicuous, the chromatin being distributed all round the periphery. In the fourth stage there is no increase in size, but the chromatin is here collected at opposite poles. Examples of both these stages may be found in Figs. 9 and 11 — it is here seen that the several cells of a gemmule may be in very different stages of division. The remaining stage, which I take to be the one resulting from the act of division, is shown in Fig. 8′ (5). It is considerably smaller than 3 and 4, and the chromatin is confined to one side of the nucleus where it forms a thin but dense layer. Nuclei in this condition are shown in Pl. XIV, Figs. 13 and 14. These several conditions of the nucleus of the gemmule cell are all abundant and easily

found. Whether or not they follow one another in precisely the sequence I have indicated, it is plain that the nuclear division, though it perhaps cannot be ranked as a karyokinetic one, is something more complex than a simple constriction of nuclear matter into two parts.

Gemmules increase in size not only by means of ordinary growth, but by fusion with one another. I think this is evident from the following facts. It is extremely common for small gemmules to occur in groups. In such groups, Pl. XIV, Fig. 8, and Pl. XV, Fig. 17, the separate follicles are often so closely pressed together as to be indistinguishable one from the other. Instances are met with not infrequently, where the shape of the gemmule gives strong indication that it has been formed by the fusion of separate parts. This is true of the gemmule x in Fig. 17, and still more so of x in Fig. 16. In Fig. 16 the dual origin of the gemmule is further indicated by the fact that one half the gemmule has nearly all its nuclei in one phase, while the other half has its nuclei in a different phase. Fusion is, I think, confined to the smaller gemmules such as those just referred to. I have not met evidence of it in the case of larger gemmules such as that shown in Fig. 19.

The gemmule, increasing in size in these two ways, grows steadily larger. An idea of the amount of increase may be got from a series of figures representing gemmules of successively larger size from quite small ones up to the mature gemmule. Such a series is given in Pl. XV, Figs. 17, 20, 20′, 19, 18. It is remarkable that while the small and large gemmules are both very abundant, medium sized ones such as that shown in Fig. 19 are hard to find. As the gemmule increases in size, it undergoes certain other changes as well. The fine yolk contained in the cells becomes more abundant, and the cells in consequence take a somewhat deeper stain. The cells become gradually much more tightly packed together than they were in the younger gemmules, and the cell outlines grow less distinct. The nuclei grow smaller. In the mature gemmule, Pl. XV, Fig. 18, the cells are so full of yolk and so tightly packed that it is very difficult to make out the cell outlines. They appear as cracks in a uniformly granular and deeply staining substance.

The nuclei are so small that one cannot make much out of them. The central chromatin mass is conspicuous and relatively so large that it is only in exceptional cases that the nuclear membrane can be made out. As a rule, all one can see in a section of the mature gemmule, is a number of small chromatin masses scattered through a finely granular and deeply staining matrix. This veiling of the cellular nature of the mature gemmule in Esperella is of importance, it will be seen, as explaining the nature of the gemmule in Tedania.

The young gemmule, as has been said, lies in the mesoderm of a trabecula. It does not project into the canals, and it is surrounded by a follicle composed of a single layer of flattened cells. Such young gemmules are shown in Pl. XV, Fig. 12, g''. As the gemmule grows it compresses the surrounding tissue, and begins to project into one of the adjacent canals. The gemmule g' in Fig. 12 may be taken as illustrating this stage. With the increase in growth the gemmule comes eventually to lie in the cavity of a canal, the surrounding tissue having been gradually compressed into the form of a sheath, which is suspended from the wall of the canal by strands of tissue. The larva *l*, and the mature gemmule *g*, of Fig. 12, illustrate this stage. The sheath, *sh.*, Pl. XV, Fig. 12, and Pl. XVI, Figs. 21, 22, consists of several layers of flattened cells and is indistinguishably fused with the original follicle, except in rare places such as that shown in Fig. 22, where inside the sheath is seen, at one end of the gemmule, the original follicle, *g.f.* In the case of this gemmule, Fig. 22, the compression of the surrounding tissue has not involved the mesoderm at one end of the gemmule, and in this region flagellated chambers are still to be seen.

Though the sponge during the summer is filled with gemmules, the asexual breeding season being apparently at its height, small egg-cells are met with here and there. They are not common but can be found after a little search. The egg-cells are always quite small and in the midst of a large collection of mesoderm cells closely packed, Pl. XV, Fig. 20'', *o. ov.* As a rule they have not a follicle, and in this condition are probably amoeboid — witness the process of the ovum in Fig.

20″. The egg-cell has always a large nucleus with a very large nucleolus. In the cytoplasm there are usually several deeply staining bodies, each surrounded by a clear space. One of these bodies in Fig. 20″ is quite large. The bodies stain as deeply as chromatin, and I suspect them to be the remnants of engulfed cells. The occurrence of such bodies in the cytoplasm coupled with the fact that the outlines of the egg-cell are often indistinct in spots, suggests that the ovum is feeding on the surrounding mesoderm cells. In a very few cases I have met an egg-cell in the peculiar situation illustrated by Pl. XV, Fig. 20‴. A gemmule, *g*, of about full size, is only partially surrounded by its follicle. The bare portion is continuous with a thickly packed mass of mesoderm cells, in which lies the egg-cell, *o. ov.* Pl. XVI, Fig. 20″, is a more highly magnified view of the bare end of the gemmule. The gemmule cells, *g*, fade away into the less densely packed mesoderm cells, in the midst of which is the ovum, *o. ov.*, surrounded by a follicle, *ov. f.*, which was not present in the egg shown in Fig. 20″. It seems pretty clear that the gemmule, *g*, after reaching its full size, burst or absorbed its follicle and became continuous with the surrounding mesoderm. Only these very small egg-cells are met with, but they serve to indicate that a sexual breeding season follows the gemmular season.

In this connection I may speak of certain gemmule-like bodies, which I am unable to explain, but which resemble a stage in spermatogenesis more than anything I know of. (See Fiedler's figures for Spongilla, 5, and those of Vosmaer for Leucosolenia, 33, Taf. xxix.) Two of these problematical bodies are shown in Pl. XV, Fig. 17, *pr. g.*, and another in Pl. XV, Fig. 14. They are comparatively common. They consist of a follicle inside which are small spherical cells entirely free from one another. The substance of these cells, if cells they are, stains feebly and appears homogeneous, and to the outer surface of each clings a crescentic band of chromatin. These bodies are always of small size, like those shown in Figs. 14 and 17. Their size and follicle suggest that they are derived from gemmules. At first I thought they were degenerating gemmules, but their uniform appearance scarcely admits of this

idea. I could find no further stage in their development, and so am unable to explain their nature, but their resemblance to certain stages in spermatogenesis is obvious.

3. DEVELOPMENT OF GEMMULE INTO SWIMMING LARVA.

After the gemmule reaches its full size (see figure of mature gemmule, Pl. XV, Fig. 18), it next undergoes a process in a measure analogous to the segmentation of an egg. It breaks up into masses, which are at first large, but which themselves break up into smaller and smaller masses, and ultimately into the individual cells, Pl. XVI, Figs. 21, 22, 23. There is no regularity at all in this splitting up of the gemmule. The masses of cells into which the gemmule first splits are of all shapes, and this continues to be true of the further division of the separate masses. Almost from the very start a certain number of individual cells separate themselves from the gemmule masses. This is shown in Fig. 21, the splitting up of this gemmule having got well under way. It is here seen that cells are set free both between the masses and on the surface of the gemmule, but that there is a special tendency for the superficial cells to become free at an early period. In this gemmule the superficial cells already form a nearly complete investment of the gemmule masses. It is evident from a study of masses such as *a* and *b* in Fig. 21, from the surface of which cells are being set free, that the outer cells act as independently motile units. They change their shape and creep away from the tightly compressed mass of cells. In the course of the separation of the individual cells, their nuclei undergo a change. The nucleus enlarges to such an extent that the membrane, nucleoplasm and nucleolus all become easily distinguishable. As the gemmule splits up it increases very considerably in size, fluid being absorbed which fills the spaces between the separating masses.

During the gradual breaking up of the gemmule masses the superficial cells come to form a connected layer. The formation of this layer was begun in the gemmule shown in Fig. 21, and in Fig. 22 the layer is established (*ect.*). The cells of

which it is composed are flat and of considerable size. Since this layer forms the ectoderm of the adult, and is a perfectly well marked embryonic layer, it seems justifiable to speak of it as the ectoderm. In the gemmule shown in this figure (Fig. 22) the separation of the gemmule masses into their constituent cells has nearly reached its end. There are still left some small multinucleate masses (*m. n. m.*), in which the cell boundaries are difficult, in many cases impossible, to make out. But the bulk of the gemmule is now composed of distinct cells. The mass of cells inside the ectoderm will be spoken of as the parenchyma or mes-entoderm, for from it are formed all the tissues of the adult except the epidermis. The cells of the parenchyma are abundantly provided with processes which connect with one another, and so establish an intercellular network. The multinucleate masses also have delicate processes, like those of the separate cells, and form part of this network.

The gemmule shown in Fig. 23 represents a slightly older stage than Fig. 22. The ectoderm cells have increased in number and have become smaller. The multinucleate masses of the earlier stage have broken up into separate cells. There are found here and there karyokinetic figures in the cells of the parenchyma. Hitherto the gemmule has been spheroidal, but now it begins to assume an oval shape.

With the assumption of a definitely oval shape there goes hand in hand a differentiation of the poles, Pl. XVI, Fig. 24. The ectoderm cells steadily increase in number, becoming all the time narrower and gradually elongating. In this way the cells become very long and slender, and the nuclei come to be arranged in several layers. In Fig. 24 the nuclei are just beginning to abandon their original arrangement in a single layer, and in Fig. 25 the transformation of the ectoderm cells is nearly complete. When the ectoderm cells have taken on the columnar shape, an orange pigment is deposited in their peripheral ends, and each cell develops a flagellum. Over one pole, however, the pole (*p.p.* in Fig. 25) which is to be the hinder one in swimming, the ectoderm cells do not become columnar. At this pole the parenchyma cells accumulate in such numbers as to form a dense mass, *p.p.*, Fig. 24. This

mass becomes more and more compact until the cells of which it is composed acquire an irregularly polygonal shape owing to mutual pressure (Fig. 25). The ectoderm cells covering the pole become more or less cubical, and at this time do not differ in appearance from the subjacent parenchyma. They neither develop pigment nor cilia, and this end of the embryo is therefore sharply marked off from the rest of the body. The remaining part of the parenchyma is made up of amoeboid cells provided with slender processes connected together into a network. The bodies of all these cells are plump and stain well. When the embryo has reached this stage of development (Fig. 25) spicules make their first appearance. They are few in number and mostly the long, slender oxytylotes. Besides the oxytylotes some curved spicules appear, the embryonic representatives of the bow-shaped spicules shown in Fig. 3*d*.

The development proceeds a little farther than the stage shown in Fig. 25, and the embryo is then set free as a ciliated larva which escapes from the body of the parent through an osculum. In Pl. XVI, Fig. 26, a surface figure of this larva is given, and in Pl. XVI, Fig. 29, a longitudinal section. The greater part of the body is of a deep orange color but the posterior pole (*p.p.*) is unpigmented. The line of separation between the two regions is a perfectly sharp one. The posterior pole ends in a pointed protuberance (Fig. 26) which appears to be a specific characteristic. A bundle of straight spicules (oxytylotes) is conspicuous in this end of the larva. In its general appearance and motion the larva is very like a coelenterate planula. Like the latter it may swim freely through the water, or may creep worm-like over the bottom and sides of the dish, the pigmented pole being posterior.

The cells of the ciliated ectoderm are very long and slender and the nuclei are packed closely in several tiers, so as to form a very conspicuous zone in sections (Fig. 29). The arrangement of several ectoderm cells is shown in the maceration preparation, Pl. XVI, *b*, Fig. 31, and one of the cells more highly magnified in *a* of same figure. There is a single flagellum to each cell. In the peripheral end of the cell is deposited the orange pigment, in the shape of small rounded masses (*p.a.*).

There then follows a clear area (*c.a.*) in which no large granules are found. A coarsely granular region (*g.a.*) comes next. The nucleus is always at the lower end of the cell, which terminates in a delicate process. The ectoderm cells over the posterior pole are of the sort shown in Pl. XVII, Fig. 33, which represents a maceration preparation of this region. The bodies of the cells extend down in an irregular fashion into the mass of parenchyma, and they take a deep stain with haematoxylin. The transition from them to the ciliated and columnar ectoderm is an abrupt one, as may be seen in the section, Fig. 29.

The parenchyma of the swimming larva is considerably more differentiated than in Fig. 25. The cells in the posterior part of the body, Fig. 29, are closely packed and polygonal. They stain feebly and their cell outlines are indistinct. In front of these cells and about in the middle of the body, is a region containing a large number of cells with plump, finely granular bodies, taking the stain well. These cells are of special importance in building up the internal tissues of the sponge and may be spoken of as formative cells. The formative cells are rounded or amoeboid in shape, with slender processes which connect the cells together. In Pl. XVI, Fig. 32, a group of such cells, as seen in a maceration preparation, is shown. The anterior part of the larva is largely occupied by fusiform cells with small bodies, taking the stain very feebly, and terminating at each end in a slender process. Scattered here and there amongst the fusiform cells are a few well-staining granular cells. It is probable from the structure of the earlier larva as well as the later, that all the cells in this stage are connected together by processes. But in macerations this was only clearly brought out in the case of the formative cells. The direction of the fusiform cells round the periphery (Fig. 29) probably indicates a connection between them and the slender terminal processes of the ectoderm cells, Fig. 31.

In the swimming larva there are three kinds of spicules present. Imbedded in the mass of pale polygonal cells of the posterior end are a number of straight spicules (oxytylotes) arranged in a loose bundle with their sharp ends pointing towards the posterior pole. These spicules very often are

found with a little mass of protoplasm and a nucleus sticking to one side, but I could come to no conclusion as to their mode of formation. The bow-shaped spicules, mentioned as present in the earlier stage, are now found in greater number, but still there are only a few of them. In the hollow of the bow there is an accumulation of protoplasm with a nucleus, and the indications are that the spicule is formed as a superficial secretion of this mass of protoplasm. The bow-shaped spicules are almost all found in the posterior half of the larva. The same is true of the third kind of spicule, the rosettes of embryonic shovels. No rosettes are shown in Fig. 29, but in Fig. 30 there are three shown, and Pl. XVII, Fig. 34, represents such a rosette (seen in section) more highly magnified. The spicules are very thin and delicate as well as small, and are not (at this time) found separately, but always united in rosettes. The rosettes are few in number and are usually found close under the ectoderm at the posterior pole.

4. METAMORPHOSIS.

The ciliated larva swims freely for a day or two. As a rule, some time during the second day after birth, it sinks to the bottom and begins to attach. The first step in the metamorphosis takes place while the larva is still swimming freely about. This consists in the flattening of the ectoderm. Pl. XVI, Fig. 27, represents a surface view of a larva 36 hours after birth, and Fig. 30 a longitudinal section of the same stage. On comparing these figures with the corresponding figures made from a larva just hatched (Figs. 26 and 29), it will be seen that the posterior unpigmented area, or region of flat ectoderm cells, has increased in extent at the expense of the pigmented area or region of columnar cells. By keeping the same larva under observation, it can be seen that the unpigmented area gradually extends forwards. As I have said, the process begins while the larva is swimming freely about. It continues after the larva has sunk to the bottom. Pl. XVI, Fig. 28, shows a surface view of a larva in course of attachment. In this larva the pigmented region is reduced to a small area at

the non-spicular pole, and this area will gradually disappear, the disappearance taking place from the spicular pole forwards. Though I have not actually witnessed the transformation of columnar cells into flat cells, this is undoubtedly what takes place. Close observation fails to reveal the casting off of any portion of the larval ectoderm, and sections give every indication that the columnar ectoderm is gradually transformed into a covering of flat unciliated cells. The replacement of columnar cells by flat ones never fails to take place in the manner described, *i.e.*, gradually from the spicular pole forwards. Now the surface area of one of the flat cells is considerably greater than that of a columnar cell and since the entire area to be covered remains approximately the same, it is obvious that all the columnar cells cannot be transformed into flat cells. What becomes of those that are not so transformed? A partial answer to this question is suggested by the very characteristic appearance of the anterior pole in the older swimming larvae (Fig. 30). As may be seen in this figure, the nuclei of the ectoderm cells are arranged in a dense zone, except at the anterior end, where they are much less densely packed, and where they form a columella-like mass projecting some distance into the interior of the larva. The cells composing this mass are so small that I cannot speak of their outlines with certainty, but they appear to be spindle-shaped. The mesoderm cells at this end of the larva are nearly all spindle-shaped, as may be seen in the figure, and the general appearance of the region suggests that the ectoderm cells are migrating at this pole into the interior of the larva. With my small store of facts this must remain a mere conjecture, and yet the point seems worth mentioning.

After the larva reaches the stage shown in Fig. 27, it sinks to the bottom and attaches in the following manner. Keeping its spicular pole applied to the bottom of the dish and its long axis more or less vertical, it begins to rotate. The rotation lasts for several hours, and may be interrupted by the larva moving to a new quarter of the dish, there to begin again its monotonous rotation. All this time the transformation of the ectoderm is taking place. After the ciliated ectoderm has

become confined to the anterior (or upper in rotation) pole, the larva ceases to rotate and applies itself to the dish obliquely, that is in the plane $x-y$ of Fig. 27. It then flattens out at its spicular pole, and in this stage is shown in Fig. 28. The flattening out continues and the patch of columnar ectoderm grows smaller, until the young sponge has assumed a flat cake-like shape. In this condition it is approximately circular in outline (see Pl. XVIII, Fig. 55, surface view of recently attached sponge), and is entirely covered with a flat epithelium, and is practically solid. The straight spicules, which in the swimming larva formed a loose bundle at the posterior end, become distributed during the flattening of the sponge, through all quarters of the body. After the flattening is completed, as is shown in Fig. 55, the spicules project slightly all over the upper surface. The outline of the sponge soon becomes irregular, and the body undergoes many changes of shape, which, however, are so slow and gradual as to escape notice, unless drawings of the outline are made at intervals. In the solid body of the sponge the canals and flagellated chambers appear as separate cavities, which subsequently unite with one another; and the pores and oscula make their appearance as simple perforations of the outer skin. All the essential features of the sponge body are established two or three days after attachment. At this time the area of the body is considerably greater than that of the swimming larva, but its actual bulk cannot much exceed that of the latter. After this stage, practically no growth occurred in the sponges I kept. They lived for weeks, but whether from lack of proper food or for some other cause, they did not continue to develop.

It sometimes happens that a larva attaches to the surface film of the water. In this case fixation takes place at the non-spicular pole, which flattens out to form a wide surface of attachment (see Pl. XVII, Fig. 37, vertical section through a larva so attached). The columnar ectoderm in such larvae metamorphoses in a different fashion from that ordinarily followed, in that the ectoderm over the surface of attachment becomes flat, while that on the sides is still columnar, as may be seen in Fig. 37. Of the larvae that attached in this way,

those I watched did not develop any further than the stage shown in Fig. 37.

In his memoir on Spongilla, Götte (6) claimed that the entire ectoderm of the larva was lost, the inner mass of cells giving rise to all the layers of the adult. This account was opposed to the earlier one of Ganin (7) who described the larval ectoderm as retained and becoming the ectoderm of the adult. In his preliminary paper on the development of Spongilla, Maas (15) stated that the larval ectoderm was not thrown off, but after loss of cilia and gradual flattening became the thin membrane-like ectoderm of the adult; and the excellent series of figures given in his later paper (14) retrace the process step by step. Some of the older writers, Metschnikoff (11) and Schmidt (22) described a partial or complete loss of the larval ectoderm in several silicious sponges during the metamorphosis; Barrois (1) believed that in his Desmacidon and Isodyctia larvae, the ectoderm was partially lost; and among the more recent investigators, Marshall (18) describes a partial loss of the ectoderm in Reniera filigrana. On the other hand, the flattening of the larval ectoderm and its transformation into the adult covering, has been observed not only in the case of Spongilla, but in other carefully studied silicious sponges : in Chalinula, Keller (10), and Myxilla, Vosmaer (34). For the views of Yves Delage and Maas on the relation of the larval ectoderm to that of the adult in Esperia, reference may be made to pp. 317–319. It seems to me that the alleged cases of total or partial loss of the larval ectoderm (ectodermic hernia) so completely lack the requisite detailed proof, that none of them can be accepted. In all such cases it is probable that the ectoderm is not lost, but is flattened into an extremely thin membrane.

Ectoderm. — In the flat epithelium into which the columnar ectoderm changes, the separate cells are at first easily made out (see Pl. XVII, Fig. 36, longitudinal section of a larva like that shown in Fig. 28). When the metamorphosis is complete, however (see Pl. XVII, Fig. 38, entire vertical section through recently attached sponge, and Pl. XVII, Fig. 44, ditto through an older sponge), the ectoderm on both upper and lower surfaces forms a very thin membrane, in which nuclei are discernible

here and there, but in which I could not make out the cell boundaries. The ectoderm covering the surface of attachment is noticeable for the deeply-stained thickenings found in it in comparative abundance (*pr. th.* in Fig. 38, and in Fig. 39, part of vertical section through recently attached sponge). These thickenings are shaped and distributed as if they might be nuclei, surrounded by an accumulation of protoplasm, but the stain reveals nothing but a homogeneous mass. These bodies are found, often of large size (Fig. 44), in the ectodermal membrane surrounding the sponge, of which I shall speak presently. In both situations they give the impression of degeneration products.

The little sponge, when the flattening out is completed, has a smooth and nearly circular outline, the mes-entoderm extending quite to the edge of the body. Pl. XVIII, Fig. 55, shows a surface view of such a stage. The ectoderm at the edge of the sponge soon begins, however, to grow out in the shape of a thin membrane which completely surrounds the sponge, extending outwards to a considerable distance. This membrane, *ec. m.*, is shown in Pl. XVII, Figs. 38, 39, 44 (vertical sections), and in Pl. XVIII, Figs. 56, 58 (surface views). Nuclei can be made out here and there in it, but the cell outlines are indistinguishable. Before continuing the description of this membrane, it will be necessary to say a word or two in regard to the mes-entoderm of the recently attached sponge.

The first change which the mes-entoderm of the larva undergoes during metamorphosis may be gathered from a comparison of Pl. XVI, Figs. 29, 30, and Pl. XVII, Figs. 36, 37. It will be seen that the formative cells increase greatly in numbers, and become distributed uniformly through the body. The pale, densely packed polygonal cells which occupy the posterior end of the swimming larva, gradually disappear, probably becoming transformed into the more independent and consequently rounded formative cells (see periphery of Fig. 36). The slender spindle cells which occupy the anterior end of the swimming larva, are in their turn distributed through all parts of the body. The next change in the development of the mes-entoderm can best be studied in surface views (see Pl. XVIII, Figs. 55,

56, 57). After the sponge takes on the round cake-like shape, the mes-entoderm becomes divided into two regions (Fig. 55), a main central region in which the formative cells are more or less rounded and pretty densely packed, and a peripheral zone in which the formative cells become branched and amoeboid, and in which they are loosely packed. The distinction between the two regions is conspicuous in Fig. 55, drawn under a low power; and the manner in which the peripheral zone is formed is clearly seen in Fig. 56 (surface view of a part of the peripheral region of a young sponge). Further development bestows on the peripheral zone the character of an exquisite intercellular network (see *p. z.*, Fig. 57, surface view of a small part of the peripheral region of a young sponge). In the sponge represented in Fig. 57, the cells of the network are of about the same size as the formative cells in the rest of the body; but as the sponge grows older, the cells of the peripheral zone grow smaller, many of them becoming delicate spindle-shaped cells. The peripheral zone after it has assumed this character, is shown more or less well in all the sections of older sponges figured (see Pl. XVII, Fig. 44, and especially Pl. XVIII, Fig. 49, the peripheral part of a section such as Fig. 44). The processes of many of the cells run directly into the ectodermal membrane, and strongly suggest an intercellular connection between the ectoderm and mesoderm in this region.

When the ectodermal membrane grows out round the periphery of the sponge, the peripheral zone of mes-entodermic cells, which is already differentiated from the central mass, begins to push out lobes and processes within the membrane which is at its inner edge obviously composed of two layers (Fig. 49). This brings it about that the inner mass of cells loses the smooth contour of younger stages (Fig. 55), its edge becoming, instead, jagged and irregular (Fig. 58). The changes of shape which the sponge undergoes at this time are due to the fluctuations of the edge of the mes-entoderm mass, not to amoeboid movements of the ectoderm cells. Sponges are sometimes found in which the mes-entoderm has pushed out processes of very considerable length between the layers of the membrane. Such a sponge is shown in Pl. XVIII, Fig. 54.

The two mesodermic processes, *m. p.*, do not come to an abrupt end, but die out imperceptibly. The ectodermal membrane, from the very beginning of its appearance, becomes covered with *débris*, much of which is organic. In Fig. 54, round the main body of the sponge are strewn little masses of degenerating cells, dead protozoa, and homogeneous, rounded masses which betray their organic nature only by their affinity for the stain, and which are evidently degeneration products. A good part of this *débris* looks as if it came from the sponge itself, as if it were composed of cells which had lost their connection with the sponge in some way, and then degenerated. Before completing the description of the ectodermal membrane, Pl. XVIII, Fig. 57, needs a word of explanation. The sponge was surrounded by an ectodermal membrane of average width, which in the region drawn was thrown, perhaps artificially, into a fold, *x*, close to the edge of the mes-entoderm.

My description of the peripheral region of the young sponge differs from that given by Maas for Spongilla (14), which he finds can also be applied to the case of Esperia (16). In the accuracy of my own observations as far as they go, I am confident; and those of Maas seem to have been made with such care that I am inclined to believe farther study will reconcile the two descriptions.

In the young Spongilla, Maas observed that the whole periphery becomes amoeboid. The formation of processes was followed in the living sponge, and it could be seen that a hyaline prolongation was thrown out far beyond the inner tissues, into which the inner cells slowly flowed, the outline then becoming more even. To this outer region, which is in constant motion, Maas gives the name of "der amoeboide Hof." The amoeboid "Hof" forms the peripheral zone of the sponge, and is composed of ectoderm. Internal to it is the body of the sponge, *i.e.*, the mass of mes-entoderm. The movements of the amoeboid "Hof" are due to the amoeboid movements of its constituent (ectoderm) cells. Silver nitrate preparations show that the cells at the extreme edge of the Hof are in active motion, throwing out pseudopodia, and combining to form lobes (the hyaline prolongations, the formation

of which was observed in the living sponge). It is possible that the ectodermal amoeboid "Hof" of Maas corresponds to my "ectodermal membrane," but I have never observed the peripheral cells to be amoeboid. Even if they were amoeboid, their movements could exert no influence on the shape of the mes-entodermic mass, merely for the reason that the edge of the ectodermal membrane is too far away from this mass. Maas does not describe the peripheral zone of amoeboid mes-entodermic elements, which is so conspicuous in the sponge I studied. The extensive ectodermal membrane I have described, which surrounds the body of the sponge, is perhaps confined to the silicious sponges, and may not, of course, be universal in them. I regard it as an excessive development of a simple layer of amoeboid ectoderm, such as clothes the attaching Sycandra, Schulze (25).

NOTE. — I find that owing to the extreme awkwardness of the term mes-entoderm, I have frequently used mesoderm as synonymous with it. No confusion will arise from this, if it is remembered that until the canal system is formed, the body of the sponge consists solely of two layers, — an outer covering (ectoderm) and an inner mass of cells (parenchyma, mes-entoderm, or mesoderm). After the canals are formed the term mesoderm is applied exclusively to the tissues lying between the ectoderm and the canal system.

Subdermal Cavities and Canals. — Both the subdermal cavities and canals arise as intercellular spaces in exactly the same manner. Intercellular spaces appear in the larva while it is attaching, *in. sp.*, Pl. XVII, Fig. 36. There are not many of them, and they are small and round. After attachment, Pl. XVII, Fig. 38, extensive cavities appear in the body, which are entirely independent of one another. The cavities formed directly beneath the upper surface are especially large, though shallow. These, *s. d. c.*, Fig. 38, are the subdermal cavities; the deeper lying spaces, *can.*, are the canals. At this time the mass of cells lying inside the ectoderm, the mes-entoderm, is, as has been said, largely composed of formative cells, with smaller slender cells scattered about here and there. The cells of the mes-entoderm are all connected together by delicate processes, and there are many indications that the ectoderm

cells too take part in this intercellular network. In Pl. XVII, Figs. 39 and 42, are shown parts of vertical sections through two sponges in the same stage as Fig. 38. Owing to the quantity of water in the sponge at this age, the tissues are extremely delicate and gelatinous, and the intercellular network in the best preparations is naturally more or less broken, with many of the cells fallen out of their proper places. That the canals and subdermal cavities arise as great intercellular spaces or lacunae in the mes-entoderm, can easily be seen in these figures. The lacunae when first established have no definite walls, but are merely surrounded by ordinary undifferentiated mes-entoderm cells, *can.*, Fig. 39. The cells immediately surrounding the cavity then begin to flatten, throwing out lateral processes in such a way as to form a more or less complete wall, in which, however, the component cells are of very irregular and diverse shapes, Fig. 42, and *can.*, Pl. XVIII, Fig. 47 (small part of a section, such as Pl. XVII, Fig. 44). The lining cells continue to flatten, ultimately forming a continuous investment of epithelioid cells, so thin indeed that they constitute nothing more than a nucleated membrane, *can. w.*, Fig. 47. In Fig. 44 there is shown a small canal, *can.'*, in which a part of the wall has reached the condition of a nucleated membrane, while the other part is still composed of cells which have not yet flattened out to any great degree. Cavities are developed everywhere directly beneath the upper surface, and there constitute, as has been said, the system of subdermal cavities, Pl. XVII, Figs. 38, 44, and 50; Pl. XVIII, Figs. 48, 51, 52, and 53. The spaces formed deeper in the tissue of the sponge become the canals. The number of subdermal cavities and canals is at first relatively small, so that the space occupied by the mesoderm is comparatively great, Pl. XVII, Figs. 38 and 44. But as new canals are formed, and as the cavities and canals gradually connect with one another, the mesoderm becomes reduced in quantity, and before long assumes the adult condition, in which it consists of uniformly thin trabeculae separating the various canals. The increase in the extent of the series of cavities may be seen in a comparison of Pl. XVII, Fig. 44, with sections through older sponges, Pls. XVII and XVIII,

Figs. 48, 50, and 51. In the latter two figures the adult condition of the mesoderm has practically been reached (compare section of adult, Pl. XIV, Fig. 2). Communication between the various cavities is established by simple perforation of the intervening tissue, the cavities in question growing towards one another, and finally meeting. In Pl. XVII, Fig. 45, it would seem that the two canals, *can.'* and *can.''*, have but lately met; and in Fig. 44 the canal, *can.''*, has made connection with the subdermal cavity, *s. d. c.*

A more comprehensive idea of the formation of subdermal cavities and canals may be obtained from a study of surface views. In Pl. XVIII, Fig. 55, the earliest cavities are shown, as yet surrounded only by undifferentiated mes-entoderm cells. Two cavities in the same early stage of development are likewise shown in Fig. 56. In the sponge drawn in Pl. XVIII, Fig. 58, in about the same stage as Pl. XVII, Fig. 44, the cavities are numerous and a higher power would show they were lined by an epithelioid membrane. The cavities shown in this figure, as those in Pl. XVIII, Fig. 54, all lie directly beneath the surface. Other deeper lying cavities are present, but these naturally are not obvious. The smooth rounded outlines of the cavities coupled with the extreme transparency of the overlying sponge tissue (dermal membrane) at first sight makes many of the cavities appear as oscula (Figs. 54 and 58), but examination soon reveals the membrane covering them.

Dermal Membrane.— The portion of the sponge body which directly covers the subdermal cavities, develops into what is known as the dermal membrane. In its adult condition, *d. mem.*, Pl. XIV, Fig. 2, and Pl. XVIII, Figs. 48, etc., it consists of three layers: on the outside the ectoderm, on the inside the epithelioid lining of the subdermal cavities, and between the two a layer of mesoderm consisting for the most part of slender spindle-shaped or fibre-like cells (comp. Pl. XIV, Fig. 4). The first stages in the formation of the membrane are shown in Pl. XVII, Figs. 38, 39, 42. As the lining cells of the cavities flatten out, the superjacent mesoderm cells grow smaller and become transformed into spindle-shaped or branched cells, most of which lie in planes parallel to the surface. In the somewhat older stages,

Pl. XVII, Figs. 44 and 45, the mesoderm cells of the dermal membrane still retain some trace of their former plump protoplasmic body, but in stages yet older, Pl. XVIII, Figs. 48, 51, the cell body consists of a mere covering for the nucleus, continued into, in the majority of cases, two long slender processes. The large number of such fibre-like cells converts the dermal membrane of the oldest stages reared into a tough, strong covering. In surface views the dermal membrane can best be studied over the subdermal cavities, where the slender bipolar cells of the middle layer of the membrane are very conspicuous (Pl. XVIII, Figs. 58 and 59, the latter representing a part of the peripheral region of a sponge like Fig. 58). The basal portion of the sponge undergoes a development somewhat similar to that of the upper crust. Many of the mesoderm cells in this region become transformed into bipolar or branched cells with very small bodies and slender long processes; compare the successive stages shown in Pl. XVII, Figs. 42, 44 and 50. Scattered amongst the bipolar cells are quite a number of larger rounded or branched (formative) cells, Pl. XVII, Fig. 50 and Pl. XVIII, Fig. 51. In this part of the sponge flagellated chambers are not developed. To be sure while the canals are still few and wide apart, a few flagellated chambers may be found close to the basal ectoderm, Pl. XVII, Fig. 44, but after the system of canals becomes more extensive the basal portion of the sponge is no longer found to contain any chambers. The same is true of the dermal membrane, in which during the earlier stages there is occasionally (very rarely) found a chamber, Fig. 44, but which in later life is entirely devoid of such structures.

Efferent Canals and Oscula. — Efferent canals are formed in this way. Some canal which usually extends deep into the tissue of the sponge (*cf. c.*, in Pl. XVII, Fig. 50, is very probably going to develop into an efferent canal) breaks through to the exterior by a large opening, the canal becoming the efferent canal, the opening the osculum, Fig. 57. The osculum is pro duced by simple perforation of the dermal membrane, the ectoderm becoming continuous round the edge of the aperture with the lining of the canal. Oscula may be formed anywhere on the surface of the sponge, in the central region of the upper

surface, *os.*, Pl. XVIII, Figs. 48, 51, 52, at the extreme periphery, Pl. XVII, Fig. 45, and Pl. XVIII, Fig. 52, and even on the under surface, Pl. XVIII, Figs. 52, 53. The appearance of oscula on the under surface is not of frequent occurrence, still I have found several in this position. The number and distribution of the oscula in these young sponges, as in the adult, is quite without regularity. There may be one or several, and they may be anywhere on the surface of the sponge.

Afferent Canals and Pores. — The subdermal cavities in the young sponges act directly as afferent canals. The membrane above a cavity becomes perforated by pores, and in the floor develop flagellated chambers, Pl. XVII, Fig. 50, and Pl. XVIII, Fig. 51. In Fig. 51, for instance, it is plain that the water entering into the subdermal cavity, *s. d. c.*, must pass directly through the flagellated chambers in order to get into the efferent canal, *ef. c.* In the adult, many of the subdermal cavities have a floor made up of a layer of flagellated chambers (*s. d. c'*, Pl. XIV, Fig. 2), and in such cases it would look as if the water passed directly from the cavity into the chambers. Though the subdermal cavities undoubtedly act directly as afferent canals in the young sponge, other afferent canals are also developed. Some of the deeper lying cavities establish connection with the subdermal spaces, for instance *can.''* in Pl. XVII, Fig. 44, and these, it would seem, become afferent canals.

In the sponges I reared, the number of pores that developed was not very great. In the oldest individuals each subdermal cavity had as a rule one or a few pores, Pl. XVIII, Figs. 58 and 59. The pores developed as perforations, the edge being strengthened, as is true also of the edge of the oscula, by the presence of fibre-like mesoderm cells. In many cases a very fair number of pores had developed before an osculum made its appearance. This is true of the sponge shown in Fig. 58. The distribution of the pores in the young sponge may be gathered without further description from the two Figs. 58 and 59. A curious phenomenon analogous to the formation of a pore sometimes takes place in the peripheral zone (*p. z.*, Fig. 57), an instance of which is shown in Fig. 59, *p. for.* The peripheral zone is perforated from one surface to the other by an aper-

ture of about the size and of the same character as a pore. Such peripheral foramina (*p. for.*) are not common in Esperella, and seem to have no function.

Flagellated Chambers. — After the larva attaches, Pl. XVII, Figs. 36, 37, 38, it is, as has been said, largely composed of formative cells. Other smaller cells, bipolar or otherwise branched, are scattered amongst them. Moreover, all the mesentoderm cells are united into a network, Pl. XVII, Figs. 39, 42. The formative cells, many of them at any rate, are multinucleate. In Pl. XVII, Fig. 46, a group of formative cells is shown, some of which are multinucleate. These possess, besides the central larger nucleus with its nuclear membrane and chromatin mass, one or more smaller peripheral nuclei, each having its chromatin mass with nucleoplasm and surrounding membrane. The peripheral nuclei at first sight look like mere chromatin spots, but more careful study satisfied me they were surrounded by nucleoplasm and a membrane. There are, however, scattered about in the cell protoplasm other bodies which stain like chromatin balls, but which are in all probability yolk granules. Two of these are shown in the lowermost cell of Pl. XVII, Fig. 46.

In Spongilla, Götte (6) has described multinucleate cells, which break up by a process analogous to budding, and form cell-groups which give rise to the flagellated chambers. The multinucleate cells are derived from mesoderm cells containing a nucleus and large yolk granules, the yolk granules becoming transformed into nuclei! Maas (14) has studied the same cells ("Dotterzellen") in Spongilla, using a differential stain (Lyons blue and carmine, or malachite green and carmine). He thinks that the cells in question contain only a single nucleus, together with a number of yolk granules of varying size. The nucleus stains red, the yolk granules blue. Maas does not believe that these cells are concerned in the formation of the chambers, but describes the latter arising as diverticula from a main entodermic cavity. Maas has also studied (16) what I have called "formative cells" in Esperia, and does not believe they are multinucleate. The bodies which I regard as small nuclei peripherally placed, he thinks

are not nuclei, but are products of cell metabolism. He further believes that these cells (formative cells) do not give rise to the flagellated chambers. For a statement of his views on this head, see p. 319.

Now, the precise way in which the flagellated chambers are formed in Esperella depends on the behavior of the formative cells. The simplest way in which a chamber is ever formed is for several formative cells to group themselves together in a hollow sphere, *f. c.'* and *f. c.''* in Pl. XVII, Fig. 42, and *f. c.* in Pl. XVII, Fig. 39. They then divide up into smaller cells, which gradually acquire the characteristic features of collared cells. In the chamber *f. c.''*, Fig. 42, the division into smaller cells has already progressed to some extent, but the cells still retain their rounded independent shape, one of them remaining much larger than the others. In the surface view, Pl. XVIII, Fig. 57, a number of chambers are shown which, I take it, are being formed in the manner described. In *f. c.'*, *f. c.''*, *f. c.'''* the formative cells are as yet only loosely combined, especially loosely in *f. c.'* In *f. c.''* the connection between the separate cells is a closer one, and some of the cells have divided, as is evidenced by the difference in size. In most of the other chambers, of which *f. c.*^{IV} may be taken as an example, the division of the cells has been carried so far that they are tightly compressed and more or less columnar. Early in the development of the chamber the nuclei acquire the characteristic appearance of the nuclei of collared cells, becoming small, and staining very deeply. The development of the collar I was not able to follow. As formed in this way, a flagellated chamber is nothing more than an intercellular space or lacuna, and in its first stages is essentially similar to a canal (compare in Fig. 42 the canal *can.'* and the chambers *f. c.'* and *f. c.''*), the lacuna becoming the cavity of the canal or chamber respectively. This means of producing flagellated chambers is only employed in the early stages, directly after fixation. In a few sponges, at this time, it seems almost the only means employed, but in most individuals it is made use of side by side with another method, which I shall now describe.

The formative cells in the recently attached sponge tend to break up into solid masses of much smaller cells. Such masses are common, several of them being shown in Fig. 39 and one in Fig. 42 (*c. m.*). The multinucleate condition of so many of the formative cells must be regarded as preliminary to division, the division resulting in some cases in the production of the masses just spoken of. In other cases, where the formative cells, as such, have grouped themselves in the shape of a chamber, cell division merely leads to increase in the number of the enclosing cells. In other cases again, no doubt, the formative cells break up into finer cells, which separate and become scattered about. In whichever way they are used up, the number of formative cells, which is very large in the just attached sponge, grows steadily smaller during the production of the canals and chambers. The solid masses of small cells to which certain formative cells give rise, are irregular in shape, and retain their connection with the cell network, Pl. XVII, Figs. 39, 40, and 41. The small cells of which such solid masses are composed quickly acquire the characteristic nucleus of the collared cell, and the mass itself constitutes the anlage of a flagellated chamber. I do not, however, believe that a single formative cell, unaided, gives rise to a flagellated chamber. On the contrary, I believe that several of the smaller solid masses of cells, each of which has been derived from a single formative cell, unites to form one of the larger masses, and this develops into a flagellated chamber. In looking over Figs. 39, 40, and 41 it is seen that the masses of cells are of various sizes; and while it is permissible to assume that one of the smaller masses can reach by simple growth to the size of one of the larger masses, the close connection which exists between many of the smaller masses (*c. m.'* in Fig. 39), coupled with the shape of some of the larger masses (*an. f. c.* in Fig. 39; this mass has already acquired its cavity), creates the impression that the latter have been formed by the fusion of the former. The solid mass of cells so formed acquires a central cavity, which at first is extremely small, *an. f. c.*, Figs 40 and 41. While the cavity is quite small, the surrounding cells are packed in several layers, but as the cavity increases

in size they become arranged in a single layer, *an. f. c.*^{II}, Fig. 40. The surface of the flagellated chamber so formed gradually becomes smooth, and its shape, which may be of almost any character (*an. f. c.*^{II}, *an. f. c.*^{III}, Fig. 41, and *an. f. c.*, Fig. 42), becomes spheroidal.

The two methods of forming the flagellated chambers just described are distinct methods, though, as will be pointed out, one may be regarded as a modification of the other. That the two methods are distinct, that one is not a mere stage of the other, must be evident from the description. On the one hand we have solid masses of quite small cells, of a characteristic appearance, giving rise to a chamber; on the other, formative cells are found grouped in a hollow sphere (only in rare instances do these large cells form solid groups, *for. c. g.*, Pl. XVIII, Fig. 57), giving rise directly to a chamber. After the system of cavities has got well started, Pl. XVII, Fig. 44, and Pl. XVIII, additional chambers are formed, I think, exclusively after the second method; at least no hollow groups of formative cells are found, but, on the other hand, solid masses of small cells are comparatively abundant.

In some few individuals the chambers are formed in yet another fashion. The cells of the just attached sponge may nearly all split up into fine cells, so that the mes-entoderm is transformed into a nearly uniform mass of fine cells, with a few larger (formative) cells scattered about here and there. In Pl. XVII, Fig. 43, is shown a part of a vertical section through such a sponge. In it one flagellated chamber, *f. c.*, is marked out. In a sponge which happens to develop in this way it seems that the flagellated chambers must be produced simply by the appearance of cavities or lacunae in the tissue, round which the cells arrange themselves in a regular wall. This manner of forming the flagellated chambers is obviously only an extension of the second method, in that very many of the formative cells early break up into masses of fine cells.

In the larva during the course of attachment one or two flagellated chambers sometimes make their appearance, as in Pl. XVII, Fig. 36, *f. c.*; but the details of the formation of such chambers were not worked out.

The three methods employed for the production of flagellated chambers in the recently attached sponge may be regarded as fundamentally the same, the second and third being modifications of the first. The third method I have already reduced to the second. The second method comes into existence because of the precocious division of the formative cells. A group of formative cells, instead of first arranging themselves round a central space and then dividing, divide and the masses resulting from their division become aggregated together and subsequently acquire a central cavity. Of the three methods the first is probably the most primitive, the other two having been derived from it. Viewed in this light, the flagellated chamber is formed in a manner essentially identical to that in which a canal is produced, and is, like the canal in its origin, an intercellular space. As may be seen in a later section of this paper, I am forced to believe that the formation of chambers and canals in the young Esperella as intercellular spaces, is best regarded as an instance of coenogeny.

The chambers increase in number with the increase in extent of the canal system ; and the number of formative cells and indeed the quantity of mesoderm in general, decreases at a corresponding rate. The gradual manner in which the distribution of the chambers, characteristic of the adult, is acquired, may be gathered from a comparison of the successive stages shown in Pls. XVII and XVIII, Figs. 44, 48, 50. In the later stages reared, the chambers are found to open into the canals, as is shown in Pl. XVII, Fig. 50, and Pl. XVIII, Fig. 48.

Spicules. — The long spicules found at the posterior end of the swimming larva become distributed through the body of the sponge during the course of attachment, Pl. XVII, Fig. 36. In the young sponge, after attachment, they are found with their sharp ends projecting for a short distance all over the upper surface, Pl. XVIII, Figs. 55, 58. Sections show that the projecting spicules do not perforate the ectoderm, but that they lift the ectoderm up, supporting it like so many tent poles, Pl. XVII, Figs. 43, 45. The first indications of the spicular bundles which support the dermal membrane in the adult, are to be seen in such stages as Fig. 44, where a few spicules are

shown lying near one another in the pillars of tissue separating the subdermal cavities. Other long straight spicules are scattered freely about in the deeper parts of the body. The bow-shaped spicules present in the swimming larva are found in small numbers distributed irregularly through the mesoderm of the attached sponge, Fig. 44. The embryonic shovels which in the swimming larva are united in rosettes, Pl. XVI, Fig. 30, and Pl. XVII, Fig. 34, are always found free in the attached larva. In the young sponges there are not many of these spicules, and the few to be seen are usually found in the dermal membrane, Pl. XVII, Figs. 44, 50.

Summary of the Leading Facts in the Gemmule Development of Esperella.

1. Gemmules appear in any part of the sponge mesoderm, and when present in large numbers, cause degeneration in the sponge tissue.

2. A number of mesoderm cells well supplied with yolk collect together and the mass so formed rounds itself off into a gemmule, the outer cells becoming the follicle.

3. The gemmule grows not only by cell division, but by the fusion with it of other small gemmules. It becomes a large mass of closely packed cells, full of fine yolk.

4. The gemmule, when mature, breaks up into irregular masses of cells, and these separate into the constituent individual cells.

5. The outer cells become ectoderm. Those at the posterior pole flatten, and develop neither flagella nor pigment. The other ectoderm cells become columnar, and develop both flagella and pigment.

6. The inner mass of cells forms an intercellular network. It is a parenchyma in which there is no distinction between an ectoderm and a mesoderm. The parenchyma cells at the posterior pole become closely compressed.

7. In the swimming larva there is a bundle of long straight spicules in the posterior end. Bow-shaped spicules and embryonic shovels (in rosettes) are scattered through the parenchyma.

8. The ectoderm begins to flatten from the posterior pole forward during swimming life.

9. The swimming larva attaches itself by the posterior pole, but obliquely, so that it lies on its side.

10. During attachment the entire ectoderm grows flat, and afterwards spreads out round the sponge, as a membrane containing no mesoderm.

11. A peripheral mesodermic zone is formed, consisting of a network of cells. To the fluctuations in the edge of this zone are due the changes in contour of the young sponge.

12. The canals and subdermal spaces arise as lacunae or intercellular spaces in the parenchyma, which are at first independent of one another, and only subsequently become connected by the perforation of the intervening tissue. The parenchyma cells immediately surrounding the lacuna develop into a lining membrane of epithelioid cells. In their origin and method of formation, there is no difference between subdermal cavities, afferent canals, and efferent canals.

13. Pores and oscula arise in the same way, as perforations of the dermal membrane overlying the subdermal cavities and efferent canals respectively.

14. Flagellated chambers arise independently of each other and of the canals, only later acquiring connection with the canal system. A chamber may be formed from a group of formative cells which arrange themselves in a hollow sphere, the intercellular space becoming the cavity of the chamber. Or else the chamber may be produced by the appearance of a central cavity in a solid mass of fine cells, derived from the division of formative cells.

Previous Knowledge of the Development of Esperia (= *Esperella*). — Besides the older observations of Metschnikoff (11), Carter (2), and Oscar Schmidt (22) — see p. 370, section VI — on the development of this genus, there are papers dealing with the subject by Yves Delage (36, 1890), Maas (16, 1892), and myself (35, 1891).

Yves Delage describes an incomplete layer of rounded cells at the surface of the larva, between which protrude the peripheral ends of the ciliated cells. The ciliated cells migrate into the interior of the larva, subsequently forming the lining epithelium of the canals, and are regarded by the author as constituting the endoderm. The superficial rounded cells then form a continuous layer and constitute the ectoderm. These facts enable the author to make a detailed comparison between the amphiblastula of calcareous sponges, and the solid larva of the silicious sponges. The former is hollow, but the cavity of the latter is filled with a mass of mesoderm (precociously formed, as compared with the amphiblastula development). And instead of the endodermic and ectodermic elements being confined to opposite halves of the larva, as in the amphiblastula, they are, in the solid larva, intermingled over the whole surface. Consequently, in the latter, the endoderm (ciliated cells) cannot invaginate as a continuous layer, but the component cells have to migrate into the interior separately. The flagellated chambers are formed from special mesoderm cells.

The description of the larvae of Esperella Lorenzi and E. lingua, given by Maas, agrees in essential points with my account of the larva of Esperella fibrex. The resemblance holds good even for many details. On the other hand, Maas describes a cavity or lacuna in the anterior end of the larva, traversed by branching cells, of which I saw no sign in the form I studied. Again, the chelae, which, in Maas's larva as in mine, are united in spherical groups, are in the former differentiated shovels, while in the latter they remain in a more embryonic condition. A more important difference is exhibited in a layer of cells which Maas describes at the anterior end of the larva, with nuclei very close to the periphery, and which he believes to be "intermediary cells," *i.e.*, cells lying between the ectoderm elements proper. I have frequently seen nuclei here and there very close to the periphery, but saw no reason for regarding them as belonging to a set of cells distinct from the ectoderm. Maas suggests that the arrangement of the spicules in the swimming larva is such that the weight is evenly distributed round the long axis — an

arrangement evidently adapted to the rotating swimming habit of the larva. This seems to me an excellent suggestion, and is very probably the true explanation of the arrangement of the spicules in such larvae.

Maas's views on the relation of the layers of the sponge body after metamorphosis to the layers of the larva may be read in the following extract (*l.c.* p. 426): "Die festgeheftete Larva besteht auf diesem Stadium wie die freischwärmende hauptsächlich aus zwei verschiedenen Gewebsschichten, die im Aussehen ganz dieselben wie die der Larve sind ; es bleibt daher nichts anderes übrig als anzunehmen, dass die innern und untern Zellen mit kleinern Kernen eines solchen Stadiums, dieselben sind, wie die äussern kleinkernigen Elemente der Larve, und dass die obern und äussern Zellen des gerade angehefteten Stadiums den innern und hintern Zellen der Larven entsprechen, dass also beide Zellschichten in der Metamorphose (wie bei Sycandra raphanus) ihre Lage zu einander verändert haben. Dieser Wechsel scheint mit dem Ansetzen in directer Verbindung zu stehen, indem die Masse der kleinkernigen Elemente am Vorderpol zusammengedrängt wird, und indem zuerst am Hinterende, dann von allen Seiten die Zellen der innern Schicht um sie herum wachsen." As may be seen from this passage (see also Maas's recent paper in the Biolog. Centralblatt, 17, p. 570), the author's views on the morphology of the larva are fundamentally different from mine. He regards the larva as composed of two layers — a layer of ciliated columnar cells covering the body except at the posterior pole, and a layer of irregularly-shaped cells filling the interior and covering the surface at the posterior pole. (Now that I have shown that the covering cells of the posterior pole and the ciliated columnar cells belong to the same early layer, Maas's views on this point seem to me untenable.) Further, the author believes that during the metamorphosis the columnar ciliated cells migrate into the interior of the attached sponge, forming the flagellated chambers and, in part at least, the efferent canals ; while the inner cells of the larva take up a position on the surface, forming the adult ectoderm and the rest of the sponge body. The author's views on this

point approach those of Delage. That the solid larva turns itself inside out in this fashion is certainly a remarkable phenomenon, and one that calls for abundant evidence. Maas's argument, as far as I can make it out, is that the attached sponge consists of two layers which exactly resemble those of the swimming larva, but that the inner cells in the attached sponge are like the outer cells of the larva; and, conversely, the outer cells of the attached sponge are like the inner cells of the larva. It seems to me that deductions made from histological similarities of this sort can never be relied on with much confidence. And especially must this be true in a case like the one in hand, where so many of the cells are undergoing histological change. I cannot see that either Delage or Maas proves his case. I have, as has been mentioned, found indications that some of the ectoderm cells of the larva migrate into the interior during metamorphosis, but I found no evidence that the ectoderm as a whole does not continue on the surface.

Maas finds that the subdermal spaces, canals, and chambers arise separately, the spaces and canals as large lacunae in the parenchyma of the sponge. Maas does not believe that the cells which I have called "formative cells" have any share in producing the flagellated chambers. He thinks the chambers are formed from aggregations of the small cells with small nuclei "which in the larva constituted the ciliated epithelium, and during the metamorphosis migrated into the interior" (16, p. 432). Maas believes that the efferent canals (in part, at least) are formed by similar cells having the same origin. I must confess that all this seems to me highly theoretical, the whole belief resting on a partial histological resemblance between the ciliated cells of the larva and the cells of which the finished or nearly finished chambers are made. As for the canals, I have always seen them formed by cells bearing no resemblance at all to the small slender cells which Maas supposes to be migrated ectoderm cells. In regard to the chambers, I am disposed to believe that the aggregates of small cells described by Maas are not different from those I have described as resulting from the division of larger cells.

II. ADULT STRUCTURE AND GEMMULE DEVELOPMENT OF TEDANIA BRUCEI, N. SP.

1. ADULT.

Diagnosis. — Tedania Brucei, n. sp. Sponge body large, usually massive, though sometimes forming flat incrustations. Color red, and body firm and fleshy. Surface marked with shallow meandering furrows and low intervening ridges, the tissue of the ridges being firm, that of the furrows gelatinous and containing large canals. The dermal membrane contains microscleres (oxeas), and is perforated by numerous large rounded pores, both over the furrows and ridges, opening into subdermal cavities. Oscula usually large and conspicuous and often at the end of oscular papillae. The main efferent canals surrounded by a large amount of gelatinous tissue; the body of the sponge is thereby broken up into an interlacing network of gelatinous and dense tracts. Flagellated chambers found only in the latter. Flagellated chambers open directly into afferent and efferent canals. The spicules include slightly curved strongyloxeas (Sollas) $\frac{20}{100}$ to $\frac{25}{100}$ mm. in length, tylotes with sharply nicked heads of same length, and very slender oxeas varying in length from $\frac{20}{100}$ mm. to microscleres of $\frac{4}{100}$ mm. and less. Skeleton of dense regions of sponge consists of a close and confused meshwork of strongyloxeas; a few bundles composed of tylotes and oxeas, crossing the meshwork; and brushes of tylotes supporting the dermal membrane, which in gelatinous regions give place to bundles of tylotes following curves of subdermal cavities. Gelatinous tracts contain scattered tylotes and oxeas, the latter from $\frac{20}{100}$ mm. to microscleres. Microscleres especially abundant round larger canals, often radially arranged. — *Green Turtle Cay, Bahamas.*

This very handsome sponge is abundant in the quiet waters of the "sounds" or deep bays which run into the island. It is especially found on the roots of the mangrove, which grows luxuriantly round the borders of the sounds, though it may also be found on the bottom, where it is apt to become incrusting. Its large size, often ten inches in diameter, and bright

red color make it exceedingly conspicuous. The specimens found on the mangroves are generally more or less ovoid in shape, with two or three oscular papillae, an inch or two long, projecting from the upper surface. The surface is furrowed in the most intricate and irregular fashion (Pl. XIX, Fig. 60, shows a small portion of the surface). The furrows are generally very shallow, but are conspicuous both in fresh and alcoholic specimens, because the tissue here is gelatinous. In the depigmented alcoholic specimens, the furrows look much darker than the ridges. The ridges, as will be seen in the figure, exhibit numerous rounded and slight elevations, which in places may appear as well-marked papillae. On the oscular papillae the furrows often become regularly arranged, when they pursue a comparatively straight course towards the end of the papilla, meeting one another at acute angles. At the end of each papilla there is usually a single osculum leading into a shallow cavity, into which open several large efferent canals. There may, however, be two or three oscula set close together on the end of the same papilla.

The division of the sponge body into the interlacing network of dense and gelatinous tracts mentioned above is shown in any section. Pl. XIX, Fig. 61, is a section vertical to the sponge surface, including two furrows, *f.*, with the intervening ridges, *r.* Both the superficial gelatinous tracts directly beneath the furrows, as well as the deeper ones, show in their centre one or more large canals (efferent canals). The dense tracts alone have the true sponge tissue, the gelatinous tracts containing no flagellated chambers. The difference between the two is made the more striking in that the dense or spongy tracts contain a close meshwork of spicules, which is absent in the gelatinous tracts. Pl. XIX, Fig. 64, is a small portion of such a section as Fig. 61 with skeleton omitted, and shows a part of a superficial gelatinous tract, *g.*, together with a part of the adjoining spongy tract, *sp.*, in which the higher magnification permits the flagellated chambers to be shown. Pl. XIX, Fig. 62, is a section across the base of an oscular papilla with skeleton omitted, and shows the connection between a superficial gelatinous tract and a deep lying one. Like the network

of sponge tissue, the gelatinous tracts form a connected system in which run the main efferent canals.

The dermal membrane, Pl. XIX, Fig. 66, contains numerous microscleres (oxeas), and the supporting brushes of tylotes are usually torn away with it. The pores are thickly distributed over most of the surface, but there are aporous or nearly aporous tracts found here and there. The pores lead directly into subdermal cavities, *s. d. c.*, Figs. 61 and 64, which are in general smaller in the spongy regions than in the gelatinous (Fig. 61). Even in the spongy regions, Fig. 64, the subdermal cavities are surrounded by a certain amount of gelatinous tissue, there being very few flagellated chambers in their immediate neighborhood. The subdermal cavities, both those under the ridges and the furrows, communicate in an irregular fashion with one another and open into main afferent canals, *af. c.*, Fig. 64, which, it will be seen from Figs. 61 and 64, enter the dense mass of flagellated chambers directly from above, and from the superficial portions of the gelatinous tracts as well. The main afferent canals subdivide, their terminal divisions opening laterally into the flagellated chambers, as is shown in Pl. XIX, Fig. 65 (*af. c.*), this figure representing a small portion of the mesoderm of Tedania, showing flagellated chambers and both afferent and efferent canals. The flagellated chambers open in the same manner into the efferent canals. The water passes out of the spongy tracts by numerous efferent canals distributed along the margin of the spongy and gelatinous tracts. These canals are well shown round the edge of several of the gelatinous tracts of Fig. 62. They open into the one or more larger vessels (main efferent canals) lying in the central part of the gelatinous tracts (Figs. 61 and 64, *ef. c.*). The main efferent canals have a denser wall than the rest, which is usually well provided with microscleres, arranged in a radial fashion. These canals communicate with one another and open at the oscula, as described above. The communication of a superficial efferent canal with a deeper lying one is partly shown in Fig. 62, *com. ef. c.*, the plane of the section cutting the connecting canal into two portions. The superficial efferent canals are especially interesting in the upper part of

the oscular papillae, or in the upper part of young sponges of a conical shape, a few inches high. In both the oscular papilla and the upper part of the young sponge, the furrows or superficial gelatinous tracts run with comparative directness straight towards the upper end. An indistinct radial symmetry is thus given the papilla, which appears more pronounced after the efferent canals are studied. For it nearly always happens that the papilla has in its axis some one canal larger than the rest, and each superficial gelatinous tract has likewise, as a rule, one large efferent canal, so that near its upper end the efferent system of the papilla consists of a central canal, round which are disposed several superficial canals, all either opening by a single osculum or by two or three separate but closely adjoining oscula. Pl. XIX, Fig. 63, represents a section cut from a young sponge of a conical shape and some five inches high. At the apex of the sponge was a single osculum, and the section drawn was cut a short distance below it. In this region the superficial gelatinous tracts contain no subdermal cavities. All that each contains is a single efferent canal running parallel to the surface and towards the apex, *sup. cf. c.* in Fig. 63. Of these, three appear in the section. A fourth unites at this level with the central canal, *c. cf. c.* Lower down the number of superficial canals increases, but they no longer run in the same direction, pursuing, on the contrary, the irregular meandering course characteristic of the general surface. The canals shown in Fig. 63 all open by a common orifice, which I have spoken of as the osculum, at the apex of the sponge.

The flagellated chambers are spheroidal, and the mesoderm of the spongy tracts is comparatively abundant, consisting of rounded or branched amoeboid and spindle-shaped cells. The gelatinous tissue is composed of a network of cells with an abundance of watery jelly in the interstices. The cells as a rule have small bodies and several long slender processes, Fig. 64.

The tylotes found in Tedania B. all have nicked heads as in Pl. XX, Fig. 67 *c*. This seems to be a common variation in the species of Tedania. Oscar Schmidt in speaking of the Atlantic Tedanias, says: "Die meisten dieser Tedanien besitzen

neben den Doppelkeulchen mit glatten Köpfen solche, wo auf dem Scheitel des Kopfes einige kleine Knötchen entstehen. Diese letztere Form nimmt in einzelnen Individuen überhand, und ist in noch anderen ausschliesslich vornanden." All the individuals of Tedania B. examined agreed in this respect, and it seems proper here to regard the nicked head as a specific characteristic. Tylotes are found free and scattered about in the gelatinous tissue. Round the subdermal cavities of this tissue they form loose bundles which follow the curves of the cavities, Pl. XIX, Fig. 61. The brushes of tylotes which support the dermal membrane have mixed with them a number of microscleres (oxeas). Crossing the meshwork of spicules and pursuing an entirely independent course, are found here and there a few long and slender skeletal bundles consisting largely of tylotes intermingled with oxeas. The oxea, Pl. XX, Fig. 67*b*, aside from the situations just spoken of, is found in abundance in the gelatinous tracts where it varies in size, as has been mentioned, from the dimensions figured to those of a microsclere, the latter form being especially common round the walls of the larger efferent canals. The skeletal meshwork is made up exclusively of strongyloxeas, which are all as is shown in Fig. 67*a*, slightly bent. I have spoken of this meshwork as confused. It is confused in the first place because the spicules are so closely packed, that the meshes are not bounded by single spicules but by little bundles. And in many places it so happens that the spicules are arranged in such a way that they both bound the meshes and help to form a continuous skeletal bundle. Two such bundles are shown in Fig. 61, which fairly well represents the meshwork of spicules.

The homology between pores and oscula upheld by Barrois (I) and others, receives perhaps some additional support from the occurrence in Tedania of such openings as those shown in Pl. XX, Fig. 68 (a small portion of the surface). These openings lie in the gelatinous furrows, and in their immediate neighborhood there are but few pores. They are larger than the pores, but very much smaller than the ordinary oscula, and may therefore be classified as structures intermediate between the two.

Collecting and Embryological Methods.— During August, September, and October, the Tedanias at Green Turtle Cay were found to contain large numbers of embryos which turned out to be gemmules, the development of which is essentially like those of Esperella. The embryos were imbedded in the mesoderm, but though they were abundant, they were not sufficiently so to cause any breaking down of the sponge tissue. The mesoderm of Tedania B. is a bright red and the gemmule embryos which were present in many stages were of the same color. If a Tedania is put in an aquarium, almost at once ciliated larvae begin to be cast out of the oscula. For the purpose of obtaining embryos, I found it was useless to keep the adult sponge more than two or three hours. During that time they throw out a good many embryos, but afterwards scarcely any. By changing the water frequently, large as the adults were, I could keep them alive for many hours, but after the first couple of hours they, like so many other marine animals, lose their irritability, and eject no more embryos. In order to get a great number of embryos it was therefore necessary to collect many adults, keeping each of them but a short while. In bringing a large sponge like Tedania B. from the collecting grounds to the laboratory, care should be taken to supply it with an abundance of water, and if it must be lifted out of the water, let the exposure to the air be as short as possible. It will be found well to support the sponge with one hand just below the surface of the water, and with the other dip a bucket beneath it. In this way a sponge may be brought into an aquarium without having been out of the water for a moment. It being a matter of considerable time and labor to bring so many sponges from the collecting grounds to the laboratory in a small sail boat, I tried on a few occasions getting my larvae directly on the grounds. Paddling along the mangroves at the head of "Black Sound," whenever we saw a good sponge, my negro boy or I would fish it up and carefully bring it into one of the two large tubs I kept full of water in the bottom of the boat. When we had pretty well filled one of the tubs, I would wait fifteen minutes and then transfer all the sponges to the other tub, and begin examining the water

of the first. This was done by dipping it out with a two-gallon glass aquarium jar, in which the bright red sponge larvae could easily be seen swimming about. These were sucked out with a pipette and put into an aquarium jar well protected from the sun. In observing and dipping at the larvae, the negro boy soon became expert, and proved himself of very considerable use for this purpose. By the time we had finished examining the water of the first tub, the water in the second would contain enough larvae to be worth looking through in this manner. The sponges were then thrown away and another lot collected.

Having obtained a stock of the swimming larvae (for several reasons I had need of a large number) they were then put in flat shallow dishes in which I wished them to attach. Many of these dishes I coated with paraffine, allowing the larvae to attach to the paraffine, as already described for Esperella. In other cases the larvae attached to the walls of the dish, or to cover-glasses placed on the bottom. As in the case of Esperella, the whole process of fixation could be observed with perfect ease by placing one of the small paraffine-coated dishes on the stage of the microscope, using reflected light and a low power. The larvae swim about for a day, as a rule, and then attach, undergoing a metamorphosis essentially like that of Esperella. The just attached sponge is a thin incrustation-like mass, in which the canals, flagellated chambers, *etc.*, appear in the course of a couple of days. The sponges I reared lived indefinitely in the aquaria, but did not increase in size after the first two or three days (and that increase was probably one of area alone, not of bulk), except in a single case where the little sponge, when killed, had reached a diameter of nearly a quarter of an inch, but had not gone beyond his brothers in morphological differentiation. I attempted to get older stages in a way which it certainly seems should have been successful, but which was not. Having allowed the larvae to attach to pieces of wood or glass, I tied these pieces to the mangrove roots, in the very home of the sponge, but even there the little sponges did not increase in size. The pieces of wood hung to the mangroves were in some cases protected by wire cages, and in

others not. The protection made no difference. The protected and unprotected sponges lived for two or three weeks (and no doubt would have lived much longer if I had not grown tired of the experiment), undergoing practically no change, and apparently exempt from attacks on the part of the little fish and crustacea which swarmed round the mangroves.

In preserving the Tedania material I was not as fortunate as I was later in the case of Esperella. For the Tedania embryos (larvae and attached stages also) I used Perenyi's fluid and also osmic acid. Neither is nearly so good as the Zacharias mixture already spoken of, though of course they give fairly good results.

2. Development of the Swimming Larva.

Formation of Gemmules. — The gemmules are not confined to any particular part of the body, but are distributed more or less uniformly through the mesoderm. The very young gemmules are simply imbedded in the mesoderm, Pl. XX, Fig. 69 *g.*, but the ripe gemmules, *r. g.*, Pl. XX, Fig. 71, are provided with a definite sheath, *g. sh.*, and are immediately surrounded by good sized canals. The gemmule of Tedania is puzzling, and I cannot claim to have actually disclosed its true structure. Still, the facts I have discovered, when compared with the development of Esperella, make it very probable that the gemmule of Tedania has essentially the same structure as that of Esperella.

The ripe or full-sized gemmule of Tedania is a large spheroidal mass, in which neither cell boundaries nor nuclei can be made out. It is densely and uniformly granular, the granules being fine yolk granules (like the yolk in the cells of the Esperella gemmule) which stain well with any stain I tried (haematoxylin, carmine, cochineal, and other aniline stains). Repeated attempts with many stains on the thinnest of sections have failed to reveal nuclei, but it is possible that the employment of a different killing fluid would lead to better results. Some idea of the puzzling appearance of the deeply staining, uniformly granular gemmules may be gathered from Pl. XX, Figs. 71 and 70, the latter showing only a small part of a gemmule with the neighboring tissues. Strange to say, in the Tedanias

which I preserved, in spite of the great abundance of full-sized gemmules, it was a very difficult matter to find medium sized and quite small ones. The only explanation I can think of is, that after a certain period no more gemmules are produced in the sponge, but those already formed are allowed to mature and develop into the swimming larva. In this way it might come about that in the latter part of the season a sponge should contain only mature gemmules, and I suppose it was towards the end of the season when my material was preserved. A few stages in the formation of the gemmule were, however, observed. A small gemmule is shown in Fig. 70 *g*. Its shape and the fact that it lies free in one of the canals indicate that it is amoeboid and was creeping about when killed. In structure it is precisely like the ripe gemmule, consisting of a finely and uniformly granular mass, the granules taking a deep stain. No cell boundaries nor nuclei were visible. Some very small masses, consisting of the same finely granular material and in which again no nuclei nor cell boundaries could be made out, were occasionally found imbedded in the mesoderm, Pl. XX, Fig. 69 *g*. These, from their histological similarity to the older stages, were construed as very young stages in the formation of gemmules. Now it will be remembered that in the mature gemmule of Esperella the cells are so closely packed and are so full of fine yolk granules, that the cell boundaries are very indistinct and the appearance is given to the gemmule of a uniformly granular mass with nuclei scattered through it, the nuclei being so small as to look like mere chromatin masses. This is what I suppose to be the true structure of the Tedania gemmule. I take it to be a mass of mesoderm cells in which the cell boundaries, owing to the compression of the cells and abundance of yolk, and the nuclei, owing to their small size, are obscured.

In the mature gemmule a few spicules or pieces of spicule are usually found, Fig. 70, *sp.*, which undoubtedly have not been formed in the gemmule itself, but have got in from the maternal tissue. The sheath or capsule which surrounds the gemmule is made up of closely packed fibre-like cells, *g. sh.*, Figs. 70 and 73. It is probably formed much as the corre-

sponding structure in Esperella, by the compression of the surrounding tissue owing to the growth of the gemmule.

Development of Gemmule into Swimming Larva. — The mature gemmule breaks up into masses, and these into smaller masses, and so on until the entire gemmule has been resolved into distinct cells, as in the development of Esperella. The process is odder and more striking looking in Tedania, owing to the absence of any indication of the individual cells and owing to the extreme irregularity of the first fissures, Fig. 72 (section through a gemmule just beginning to break up). In Fig. 73 is shown a portion of a section through another gemmule which has already broken up into small masses of varying size, even in the smallest of which nuclei are as yet invisible. In a stage a little later, Fig. 74 (the entire embryo was spheroidal), nuclei make their first appearance. In this embryo the division of the gemmule masses has been carried so far that the individual cells are easily recognisable. The superficial cells are packed tightly enough to make a continuous layer, which will become the ectoderm, inside which are scattered cells and rounded masses, separated by a clear fluid and more or less united by delicate protoplasmic processes. The bodies of the cells and rounded masses are just as full of the finely granular yolk as was the mature gemmule, and nuclei are only visible in some of the cells and a few of the masses. The masses are usually divided into rounded lobes and are obviously about to split up into individual cells.

The hitherto spherical embryo begins to assume an oval shape. In Pl. XXI, Fig. 75, is shown part of a section through a roughly oval embryo, in which the differentiation of the layers is noticeably more advanced than in Pl. XX, Fig. 74. Nuclei are apparent in all the cells, and the ectoderm is more distinctly marked off from the inner mass of cells (mes-entoderm). In some of the ectoderm cells two nuclei can be seen, indicating that cell division is taking place. The mes-entoderm in this stage consists of a very loose network of cells connected together by long slender protoplasmic processes.

Many of the mes-entoderm cells are closely packed in dense groups, and there are a number of multilobed and often multi-

nucleate masses, as in the previous stage. Most of the mes-entoderm cells have, like the ectoderm cells, plump bodies full of fine yolk granules, but there are some with smaller slender bodies, in which there is but little yolk and which begin to assume the appearance of the spindle-shaped cells, so abundant in the older embryo (compare Pl. XXI, Fig. 81). The pieces of spicules found here and there in embryos of this stage were already present in the gemmule before it began to break up. It will be noticed in this stage, Fig. 75, that the ectoderm cells are, in many instances at any rate, connected with the mes-entoderm cells by fine terminal processes. This connection probably continues to exist in the later stages, but I did not satisfactorily demonstrate it.

The ectoderm cells which already form a distinct layer in Fig. 75 divide in planes vertical to the surface, and become long slender columnar cells. These slender columnar cells form for a time a uniform investment for the whole embryo, Pl. XX, Fig. 76, though later they flatten out over one pole. In Pl. XXI, Fig. 76, is shown a small part of Fig. 76, more highly magnified. The rounded multilobed masses of the earlier stage are no longer found in the mes-entoderm, which now consists only of separate cells.

While the embryo is still in the body of the mother and surrounded by its capsule, the ectoderm cells over one of the poles flatten out, while elsewhere they develop cilia and become deeply pigmented. In Pl. XX, Fig. 77, is shown a section through this pole of the embryo at a stage just before the flattening has begun. The ectoderm cells over the general surface have flagella, and nuclei near their lower ends, the nuclei forming a zone several layers thick. The ectoderm cells at the pole, however (*ec. un-p. p.*), are not quite as slender and have no flagella. Their connection with the cells of the mes-entoderm is still obvious. These cells gradually flatten until, by the time the embryo leaves the body of the mother, they have assumed the character shown in Pl. XXI, Fig. 78 (*ec. un-p. p.*). The mes-entoderm in the stage shown in Fig. 77 is much as in the earlier stage, with the exception that a number of spicules are now scattered through it, all of them

very short and slender, and pointed at both ends (oxeate microscleres).

Structure of the Swimming Larva. — The larva, when it escapes from the body of the mother, is solid, of an oval shape, with one unpigmented unciliated pole, the rest of the body being covered with cilia and of a bright red color. It moves rapidly about in the water, occasionally creeping, but usually swimming, and it seems especially fond of making series of long shallow dives, coming up to or near the surface between the dives. The swimming larva can also change its shape to a slight extent.

The general ectoderm of the larva is composed of very long and slender cells, *ec.*, Pl. XXI, Fig. 78 (section through the unpigmented pole of a larva just born), which contain the bright red pigment. Each of these cells has a single flagellum, and the nuclei contained in their inner ends make a broad, deeply staining zone. The extreme peripheral ends of the cells are modified to form a cuticle, *cu.* in Fig. 78. The columnar ectoderm cells become shorter towards the unpigmented pole, as is shown in the figure, and yet pass with considerable abruptness into the flat cells covering this pole. The latter cells do not contain pigment, but are granular and stain deeply. The ectoderm remains unchanged during the free larval life (comp. Pl. XXI, Fig. 81, longitudinal section through a swimming larva a day old).

Like the ectoderm, the parenchyma of the swimming larva remains essentially the same throughout larval life — compare the two sections, Fig. 78 (through larva just born) and Fig. 81 (larva a day old). The parenchyma of the larva is much more differentiated than it was in the stage shown in Fig. 77. The parenchyma cells of the latter stage were essentially alike and were pretty evenly distributed, but during the last period of embryonic life, they become variously modified. Some of them crowd into the unpigmented end of the larva, becoming more and more tightly packed, and forming ultimately a dense mass of closely appressed polygonal cells, which stain faintly and the cell outlines of which are distinguished with difficulty, *p. c.*, Fig. 78. (In regard to this mass of cells, as in so many other

points, the larvae of Tedania and Esperella are essentially identical.) Next after the mass of pale cells comes an aggregation of large granular well-staining cells, *gr. c.*, Figs. 78, 81. Following upon the granular cells, the axial part of the larva, *ax. p.*, Fig. 81, is occupied by a mass of slenderer and less granular cells, provided with delicate processes, many of the cells being bipolar. This part of the larva is considerably denser than the peripheral part, *per. p.*, in which the cells are relatively less numerous, many of them lying in a more or less radial direction, parallel with the short spicules, which in the swimming larva are confined to this region. Besides the cells mentioned, there are found scattered here and there through the body of the larva a small number of *very* coarsely granular, deeply staining cells. A few of them are shown in Fig. 78, lying amidst the pale cells at the end of the larva; others are shown in Fig. 81, some of them in the ectoderm, others in the axial part of the larva. As in the case of Esperella, so in this larva, there is a loose bundle of long spicules in the unpigmented end of the body, Figs. 78 and 81. The spicules are the tylotes with nicked heads. The strongyloxeas, the spicules which in the adult form the skeletal meshwork, do not appear in the swimming larva.

There is only one noticeable change which occurs in the larva during its short swimming life, and which concerns the unpigmented pole. When the larva is just born, this pole does not protrude to any great extent, Fig. 78. Indeed, quite often this end of the body is pulled in, Fig. 79 (surface view of larva just escaped from parent sponge). But after fifteen or twenty hours of larval life, it is found that the unpigmented end protrudes to such an extent, that it is a very conspicuous feature of the living larva, Fig. 80. (Surface view of a larva a day old. In this figure the peripheral zone of short spicules is shown.) In sections, too, this difference between larvae just born and older ones, is noticeable — compare Figs. 78 and 81.

3. METAMORPHOSIS.

Attachment.—With most individuals the swimming life lasts about a day, with some two and three days. Towards the end

of the period, whatever it may be, the larvae become sluggish and lie about on their sides on the bottom of the dish. They then attach, the columnar ectoderm cells are transformed into flat cells, and the body of the larva flattens out into a round cake-like mass. The distinction between the pigmented and unpigmented portions of the body is entirely lost, the whole surface becoming red. In Pl. XXI, Fig. 84, is shown a surface view of recently attached sponge, and in Pl. XXII, Fig. 92, one-half of a vertical section through the same. The long spicules, which form a bundle in the unpigmented end of the larva, become scattered irregularly through the body. In attaching, almost all the larvae I have watched have stuck fast by their sides and not by one end. Such a larva just attached is shown in Pl. XXI, Fig. 85. Its outline still recalls the outline of the swimming larva (Fig. 80), it not yet having assumed the circular shape of Fig. 84. The body is solid; the ectoderm is entirely composed of flat and very thin cells; and the unpigmented or spicular pole of the larva (*sp. p.*) can still be identified both by the absence of pigment and the presence of the long spicules. The line of demarcation between the pigmented and unpigmented regions is not a sharp one, as it was in the swimming larva, and the spicules are no longer arranged in a bundle, but have begun to scatter about, though as yet they are still confined to one end of the sponge.

The attachment may take place obliquely, so as to bring the spicular pole on the upper surface of the metamorphosed larva, though near the periphery. In a few cases I have seen the attachment take place by the non-spicular pole, a little obliquely, to be sure, as is shown in the surface view, Fig. 82. In this larva the spicular pole was pulled in to such an extent, that at first sight it looked like an opening leading into the interior, though as a matter of fact it was nothing of the kind. When the attachment takes place by the end, as in Fig. 82, the spicular pole comes to occupy a more or less central position on the upper surface of the metamorphosed larva. In Pl. XXI, Fig. 83, is given a vertical section through a little sponge, which must have attached by the non-spicular pole, for on the upper surface and more or less in the centre is found the

ectodermal area (*ec. un-p. p.*) which covers the spicular pole in the swimming larva. These cells of the swimming larva, as has been said, stain very intensely, and have too characteristic an appearance for their identity to be doubted. The rest of the ectoderm in Fig. 83 is composed of flat, thin cells, but the area in question has precisely the same appearance as in the swimming larva. The long spicules too have remained about in the same position which they occupy in the swimming larva, pieces of them being shown in the figure directly beneath the deeply stained patch of ectoderm. The variation which the Tedania larvae exhibit in their manner of attachment, is shown in other silicious sponges (see section on Morphology of Sponges, p. 364).

That the columnar ectoderm of the larva flattens and is not cast off, is evidenced by the fact that during the metamorphosis the sponge retains its smooth surface, and that no membrane or bit of membrane is seen to be sloughed off. The flattening of the ectoderm takes place quickly, being completed a very short time after the fixation of the larva. In Fig. 83 the flattening of the columnar cells has taken place. After the polar ectoderm (*ec. un-p. p.*) has in like manner flattened, the entire investing layer of cells is so thin that it is best described as a nucleated membrane, *ec.*, Pl. XXI, Fig. 89.

During the flattening of the ectoderm the parenchyma of the larva also undergoes changes, as may be seen on comparing Figs. 81 and 83. In the attached sponge there are two kinds of cells which have no definite arrangement. There are first, great numbers of very small cells, so small that only the nuclei are seen with distinctness, the cell outlines being practicably indistinguishable; and there are also numbers of deeply staining, plump-bodied, granular cells, such as were found in the spicular end of the swimming larva. The sponge shown in Fig. 83 (vertical section) flattens out considerably, especially at its periphery, and assumes the shape indicated by the vertical section, Fig. 92, and the surface view, Fig. 84, the parenchyma remaining practically unchanged. With regard to the rearrangement of the spicules of the swimming larva, something has already been said of the long (tylote) spicules.

They become irregularly distributed through the body of the sponge. The short spicules (microscleres) likewise become distributed through the sponge body (Fig. 83), though the majority of them retain their peripheral location, many projecting from the surface of the sponge, as shown in Figs. 84 and 85. Sometimes a sponge is found with the peripheral microscleres arranged in as noticeably radial a fashion as in the swimming larva. Such an instance is shown in Pl. XXI, Fig. 86. As a rule the radial arrangement of the microscleres is not nearly so conspicuous as in this figure. At this period of its existence (Fig. 84), the sponge is a much simpler organism than during its swimming life, consisting as it does of a solid mass of parenchyma cells, in which there is no nice arrangement as in the free larva, and of an ectoderm which is nothing more than a nucleated membrane.

Ectodermal Membrane and Peripheral Mesodermic Zone. — After attachment the edge of the sponge is for a time more or less circular and smooth, and the mesoderm extends quite to the periphery, Pl. XXI, Fig. 84, and Pl. XXII, Fig. 92. The contour of the sponge then begins to change, and the periphery becomes more or less lobed and irregular, Pl. XXI, Fig. 86 (compare also the ectodermal outline, *ec.*, of the sponge given in Fig. 88). An accumulation of fluid then takes place in the extreme peripheral part of the sponge, by which means the ectodermal edge is pushed out some little distance from the edge of the mesoderm. In Fig. 88 this separation has taken place on opposite sides of the sponge, and between the edge of the mesoderm (*mes.*) and that of the ectoderm (*ec.*) is seen a clear space occupied by fluid alone. In Fig. 89 is represented a small part of the periphery of a sponge in which this process is going on — the parenchyma cells are separated from the ectoderm much farther in the middle than at the sides of the figure. In Pl. XXI, Fig. 87, a portion of the periphery of another sponge, fluid separates ectoderm from mesoderm in the regions *a*, *b*, *c*. The ectoderm continues to grow peripherally, the distance between its edge and the edge of the mesoderm continually increasing. In this way the sponge body comes to be surrounded by a purely ectodermal membrane. In the

immediate neighborhood of the mesoderm, as is shown in the section, Pl. XXII, Fig. 94, the membrane, *ec. mem.*, consists of two layers, the upper and lower ectoderm respectively, but farther out it is one-layered. The ectodermal membrane extends for some distance beyond the body of the sponge, and is more or less covered with debris. It is essentially like the corresponding structure in Esperella. The membrane is shown in sections in Fig. 93, and in the surface view, Fig. 90, its outline, *ec. mem.*, is partly indicated. The sponge shown in Fig. 90 is only partially surrounded by the ectodermal membrane, retaining its earlier character in the region *a*, where the ectoderm has as yet taken no step towards forming a membrane. Nuclei could be made out here and there in the membrane and in the ectoderm proper, but the cell outlines I could not distinguish. The same deeply staining thickenings which were found in Esperella, are again found in the basal ectoderm and membrane of this sponge, *pr. th.*, Pl. XXII, Figs. 93 and 94. The only construction to be put upon them seems to be that they are nuclei surrounded by protoplasm.

As the ectoderm grows out to form the membrane, the peripheral mesoderm throws out lobes and processes, its outline becoming jagged and irregular, as in Fig. 88, *mes.* The cells of this part of the mesoderm gradually form a peripheral zone, distinguishable from the rest of the body by the fact that they are much less closely packed than the cells elsewhere (*p. z.* in Pl. XXII, Figs. 90, 91, 93). The cells of this peripheral mesodermic zone develop slender processes, and form a net-work (sections, Figs. 93 and 94, *p. z.*), which, however, is not nearly so open and exquisite as in Esperella.

During the formation of the ectodermal membrane, and afterwards during all the time I kept the young sponges, they underwent an incessant change of shape, which was more conspicuous during the first three or four days than it was later. This change of shape, though gradual, was greater and more rapid than in Esperella, and the little sponges were much disposed to assume peculiarly irregular shapes, such as that of the sponge shown in Fig. 91 (*mes.* indicates outline of the parenchyma — the whole sponge is supposed to be surrounded

by an ectodermal membrane). The change of contour concerns especially the parenchyma, which pushes out lobes and processes inside the ectodermal membrane, thus acquiring from time to time entirely different outlines, while the surrounding ectodermal membrane remains practically unchanged. If the parenchyma, however, continues to change its contour in such a way that the shape of the whole sponge is altered, as, for instance, in passing from a circular outline to a shape such as that in Fig. 91, then the ectodermal membrane is involved and its edge gradually altered so as to remain more or less parallel with the general contour of the parenchyma.

In sponges which have assumed elongated irregular shapes, like that of Fig. 91, the change of contour sometimes leads to the complete division of the body into two independent sponges. This phenomenon I have twice observed. I thought at one time that I had witnessed the converse phenomenon, *i.e.* the fusion of two attached sponges into one. I observed two sponges, a couple of days after attachment, which lay near each other, grow nearer and nearer until after fifteen to twenty hours they met and seemed to fuse into one body of an irregularly oval shape. Across this body, however, could be seen the seam or line of fusion, and the union must have been one of close juxtaposition only, for after a few hours the sponges again separated along this line and afterwards remained independent. Fusion of the swimming larvae, into a single large one, as occasionally happens in the Coelenterates (Manicina) I have never observed.

Canal System. — The canals and subdermal cavities appear as separate lacunae in the parenchyma, the surrounding cells becoming modified into epithelioid membranes, Figs. 93 and 94, *s. d. c.*, *can.* The separate lacunae subsequently become united into a canal system, as in Esperella. The flagellated chambers likewise originate as independent structures, which later acquire connection with the canals, Fig. 93. The subdermal cavities, which in some cases are very extensive, as in Fig. 91, *s. d. c.*, are roofed over by a dermal membrane (*d. mem.* in Fig. 93), quite like the same structure in the young Esperella. As in Esperella, no system is followed in regard to

the order of formation of the canals. In some individuals the first cavities formed are narrow rounded canals, *can.*, Pl. XXII, Fig. 88, covered in only by the dermal membrane, and which from the surface look like oscula. Occasionally a stage is found where but one of these canals exists, and that in the centre of the body, Fig. 90. Such a stage is interesting, because of its essential resemblance to the young Reniera (Marshall), or Chalinula (Keller), etc., in which sponges the first canal to form is regularly a main central cavity which is ordinarily homologised with the central cavity of calcareous sponges and regarded as the gastrula cavity.

Pores and oscula were developed in only a few of the sponges I reared, and were themselves few in number. They made their appearance without order, scattered about as in Esperella, and in other respects too their formation agreed with that of the same structures in the latter sponge.

III. Adult Structure and Egg Development of Tedanione Foetida, n. g.

1. Adult.

It is necessary to create a new genus for this form, which, it would seem, however, is closely related to Tedania. The spiculation of the two genera separates them, though the occasional presence of tylotes in Tedanione coupled with the great similarity in the canal system and histological structure makes a close kinship between the two very probable.

Diagnosis of Genus. — Spicules mostly oxeas, with microscleres of same pattern, and a very few tylotes. Flagellated chambers open directly into afferent and efferent canals.

Tedanione foetida, n. sp. — Sponge amorphous with two or three cylindrical oscular papillae one inch high. Size, rarely over three inches from osculum to base. Sponge is slatebrown, has a fetid odor in life, and the surface is irregularly and inconspicuously furrowed. Main efferent canals surrounded by large amount of gelatinous tissue which only occasionally comes to the surface. Pores rare and scattered. Subdermal cavities

everywhere beneath a dermal membrane. Spicules, stout skeletogenous oxeas, $\frac{3.5}{1\,0\,0}$ mm. long and often slightly bent; microscleres (oxeas) of varying length; also a few tylotes. Dermal membrane strewn with oxeas of full size, amongst which are scattered microscleres, with here and there a tylote. Membrane supported by brushes of oxeas containing a very few tylotes. Spongy tissue contains radial skeletal bundles, composed of oxeas, running in from the brushes; bundles composed of same spicules crossing the former at right angles some little distance below the surface; and numbers of oxeas scattered freely through the tissue in such a way that they cross one another in every direction, but are not cemented together to form a network. Gelatinous tissue contains both ordinary oxeas and microscleres scattered freely about, microscleres most abundant immediately round main efferent canals. *Green Turtle Cay, Bahamas.*

Tedanione foetida is found in the "sounds" on the roots of the mangrove. The surface of the sponge is furrowed in a manner recalling the surface of Tedania, but the furrows are not nearly so abundant nor conspicuous as in the latter genus. On cutting the sponge open it is seen that the body is divided as in Tedania into spongy and gelatinous tracts, the gelatinous tissue lying around the main efferent canals. But it is only occasionally that the gelatinous tissue comes to the surface. In most places it lies in the interior completely covered by spongy tissue. In Pl. XXII, Fig. 96, a vertical section through the base of the sponge is shown, and it is seen that the gelatinous tissue is wholly in the interior of the body. In Pl. XXII, Fig. 95, a transverse section through an oscular papilla is shown, and here the resemblance to Tedania is greater, for the gelatinous tissue comes to the surface in several places.

The dermal membrane is strengthened by numerous oxeas of full size, Pl. XXIII, Fig. 100, and the pores are few and scattered. Pl. XXII, Fig. 97, represents a section vertical to the surface, and shows the gross features of the canal system. The subdermal cavities are numerous and open into larger or smaller afferent canals, by which the water is introduced into the spongy regions. The flagellated chambers communicate

directly with the afferent canals on the one side and the efferent canals on the other, as may be gathered from Pl. XXII, Fig. 98, representing a small portion of the mesoderm of the sponge. Efferent canals are abundant round the edge of the spongy regions (Figs. 95 and 97), and communicate with the larger efferent canals lying in the heart of the gelatinous tissue. In the body of the sponge the main efferent canals pursue an irregular course, but in the oscular papillae they run longitudinally, there being at the base of the papilla several which gradually run into one another as they near the summit of the papilla. There is usually one canal in the axis of the papilla, which is larger than the rest and may be considered the main canal of the papilla.

The gelatinous tissue is much like that of Tedania, consisting of a network of cells with a watery jelly in the meshes. As in Tedania, there is an abundance of delicate bipolar cells, the processes of which are long, slender, and branching. There are also numerous large granular cells, not present in Tedania. Pl. XXIII, Fig. 99, is a small portion of a section showing the gelatinous tissue lying between two canals (*c. w.* = canal wall).

The general arrangement of the skeleton is shown in Pl. XXII, Fig. 97. The brushes of spicules supporting the dermal membrane, the radial and tangential bundles, and the distribution of the free spicules, need no further description. Where the gelatinous tissue comes to the surface, the brushes of spicules supporting the dermal membrane are either absent, or feebly developed. The skeletogenous oxea is very often found with its two ends modified after the fashion shown in Pl. XXII, Fig. 101. The length of the process x varies considerably. What few tylotes occur are found either in the dermal membrane or in the brushes supporting it. I have seen three or four tylotes with nicked heads like those of Tedania. As in Tedania, the microscleres are most abundant round the walls of the efferent canals, but while they are larger than the microscleres of Tedania, they are much less numerous. It may be mentioned that after hunting persistently through many sections and caustic potash preparations of this sponge, I have found four anchors, varying in size but otherwise alike. Being unable to

find any more, my conclusion is that these anchors are foreign particles, and that bits of the sponge to which they originally belonged entered Tedanione and were used as food.

2. Development.

My observations on the development of Tedanione and Hircinia deal only with the egg development, going in the former sponge as far as the formation of the swimming larva, but in the latter no farther than the segmentation.

Tedanione was with eggs in September and October and possibly for a much longer time at Green Turtle Cay, Bahamas. Adults were kept in aquaria, and after an hour or two, as a rule, a few ciliated larvae were thrown out of the oscula.

The very young ovarian egg is of an irregular shape and lies in the mesoderm surrounded by a follicle of flattened cells, Pl. XXIII, Fig. 102, *ov. o.* It has a large nucleus and single nucleolus. As the egg increases in size it becomes rounded, its protoplasm becomes filled with yolk, and the nucleus undergoes certain changes, which are not completed until the egg has attained its full size and is ready for segmentation. A general idea of the change in size and character of the egg during its growth may be gathered from a comparison of Pl. XXIII, Figs. 102, 103, 104, 105, 106, drawn to the same scale and representing successive stages in the life of the egg.

During the increase in the size of the egg, its follicle is constantly surrounded by a dense mass of mesoderm cells, as may be seen in the section, Fig. 103 (*mes.* = the cells in question; the egg is one-half the full size). These cells have large, plump bodies which stain well, being full of a finely granular yolk. The shape and direction of the cells on the outskirts of the mass indicate a migration of mesoderm cells from all quarters to the egg. By the time the egg has reached its full size the surrounding mass of cells has dwindled away to a small number, Fig. 104, and during the remaining period of its life in the parent sponge the embryo is surrounded by ordinary mesoderm, in which the cells are not more thickly crowded than elsewhere in the body. It is only while the egg

is growing larger and becoming stored with yolk that it is surrounded by the cells in question, the purpose of which it would seem is to bring food to the young egg. Since none of the surrounding mesoderm cells are ever seen to break through the follicle, it must be that the food is passed through the follicular membrane in a liquid shape and is then absorbed by the egg. Fiedler's description (5) of the manner in which nutrition is brought to the growing egg of Spongilla, differs from the above account in some respects. In Spongilla special "Nährzellen" congregate round the egg during its growth, and penetrate between the follicular cells, supplying the egg with food. The "Nährzellen" do not fuse with the egg, and it would seem that the food must be transferred by osmosis. As in Tedanione, the nourishing cells disappear when segmentation begins.

The very small ovarian egg is filled with the extremely fine granular yolk which is found in the body of any mesoderm cell at all noticeable for its size. But as the egg increases in size, yolk of a different character makes its appearance, consisting of small spheres thickly packed. In eggs of about one half the adult size, Fig. 103, these small spheres may be found filling the entire peripheral region, but leaving round the nucleus an area containing only fine granules. With continued increase in size the whole egg becomes filled with yolk spheres, which themselves increase considerably in size, as may be seen on comparing Fig. 103 with Fig. 104, the egg in the latter figure being of full size.

The nucleus of the young egg cell contains a single nucleolus which occupies a more or less central position, Fig. 102. By the time the egg has reached a size equal to one half that of the ripe egg, the single nucleolus has given place to two, which are invariably placed on opposite sides of the nucleus and adhere to the inner surface of the nuclear membrane, Fig. 103. In eggs which have reached the adult size it is the rule to find either one nucleolus peripherally placed, as in Fig. 104, or the nucleus contains no nucleolus at all, as in Fig. 105. It sometimes happens that an egg of full size is found with two nucleoli, but this is rare. From this evidence it would seem that

the two nucleoli present in the developing egg are lost, one after the other, at the time when the egg reaches its full size. As to how the first of the two is lost, I have no evidence, but the second nucleolus may often be seen lying just outside the nucleus in the yolk, Fig. 105 *n''*, showing that it has been extruded from the nucleus. The nucleus differs in size so little from the yolk balls, and the latter stain so deeply, that I was at first in doubt whether to claim the object seen just outside the nucleus as an extruded nucleolus, or to regard it as merely a yolk ball. But so many eggs showed this one very deeply staining sphere in about the same position, that I was finally convinced it could be nothing less than the extruded nucleolus. Very rarely an egg much less than the full size is found with but a single peripherally placed nucleolus, indicating that the first nucleolus has already been lost. But this is a rare exception, the rule being that the nucleoli disappear only after the egg attains its full size. The nucleus which remains after the extension of the nucleoli, Fig. 105, has a membrane and finely granular contents which stain feebly.

My observations on the formation and loss of the nucleoli were made in the Bahamas in the fall of 1888. On my return I found that Fiedler (5) had just described the same phenomena in Spongilla, and regarded the two small nucleoli as polar bodies. Fiedler finds that the two small nucleoli are constricted off as buds from a larger central one, the latter remaining after the extrusion of the former. Further, at the time when the nucleoli are extruded, the nuclear membrane disappears. There are thus some differences of detail between our accounts. In the interpretation of these bodies as polar globules I cannot agree with Fiedler, because they are formed (though not discharged) long before the egg reaches its full size. Moreover, polar bodies of the ordinary metazoon type exist within the group of sponges, as is shown by Magdeburg's discovery of them in Plakina trilopha (see the notice of Magdeburg's unpublished observations in Korschelt & Heider, p. 1).

The segmentation of the egg of Tedanione is total, and regular, at any rate as regards the first two planes. In Pl. XXIII, Fig. 106, is shown the stage of two segments, and in Fig. 107

the stage of four. In these early stages the nuclear membrane could not be made out, the clear space round the nucleolus being only vaguely outlined. In a later stage, Fig. 108, probably sixteen segments, the membrane first makes its appearance, though it has no doubt been there the whole time. An advanced stage of segmentation is shown in Fig. 109, a still later stage in Fig. 110, an embryo as yet unciliated in Fig. 111, and the ciliated free swimming larva in Fig. 112.

In Fig. 110 is shown a curious phenomenon. What appear to be cell membranes are distinctly seen round many of the segments, the body of the cell in some cases having fallen out of its surrounding membrane. These membranes are protoplasmic and are undoubtedly artefacts caused by the fixing fluid. They are of interest only as indicating how sharp the demarcation is between the cortical layer of pure protoplasm which invests each segment, and the yolk-containing protoplasm which makes up the mass of the segment. In the sudden contraction due to the stimulus of the fixing fluid, it would appear that the central and cortical protoplasms of one segment part company more easily than the cortical layers of adjacent segments, which in life must be closely appressed. Though I regard these "membranes" as artefacts, I am aware that Schulze has described somewhat similar structures in the larva of Euspongia (37), which he considers to be of a normal nature. Between the parenchyma cells of the solid larva of Euspongia, Schulze describes partitions, which he is in doubt whether to regard as secretions or as the modified cortical layers of the cells. He thinks it probable that they are later transformed into the uniform watery jelly in which the cells lie, and points out the analogy to cartilage, comparing the intercellular partitions with the capsules which go to form the cartilage matrix.

The cells of the embryo, Fig. 111, are full of fine yolk, and, being very closely appressed, the outlines are indistinct as compared with earlier stages. The metamorphosis of the large yolk spheres of the ripe egg into fine yolk goes on gradually during the segmentation (compare Figs. 104, 106, 108, 109, 110, 111), retracing the path which was followed in the development of the small egg cell into the ripe ovum.

In the transformation of the simple embryo, Fig. 111, into the ciliated larva, the outer cells become the ectoderm, the inner cells forming an undifferentiated mass, the parenchyma or mes-entoderm. When the embryo escapes from the parent and begins its free swimming life, it is in the condition shown in Fig. 112 (longitudinal section). The ciliated larva is of an oval shape, one end being considerably broader than the other, and of a uniform brown color. It is ciliated all over, there being no differentiation of an unciliated, unpigmented pole as in the gemmule larva of Tedania and Esperella. I did not follow the further development of the swimming larva, but it is quite possible that an unciliated pole may later make its appearance, as in the *Desmacidon* described by Barrois.

The ectoderm of the larva is uniformly composed of very long slender cells, the peripheral ends of which contain the nuclei and being free from yolk form a zone clearly marked off from the rest of the larva, Fig. 112. In this zone the outlines of the ectoderm cells are plain enough. The ectoderm cells, however, extend a long distance internally from this zone, and their inner portions containing fine yolk, similar to that with which the mes-entoderm cells are filled, the cell outlines are here not very distinct. The parenchyma at first sight appears to be a uniformly granular matrix containing nuclei. But in very thin sections its cellular nature can be made out. It is composed of irregularly polygmal cells, which are so closely appressed and so full of fine yolk granules, that the cell boundaries are obscured.

In sectioning the parent sponge for embryos, I came across the curious case of attachment illustrated in Fig. 113. A ciliated larva of an irregular shape, and containing two or three flagellated chambers, is present in one of the larger canals, and appears to have attached to the wall of the canal instead of passing out of the body of the mother. There is, as can be seen in the figure, a perfect continuity between the mesoderm of the parent and the parenchyma of the larva. The columnar ectoderm on the other hand does not seem to be continuous with the epithelioid lining of the canal, but rather to pass into the mesoderm of the adult through a break in the canal wall.

There are two or three flagellated chambers present in this sponge, one of which is shown in the figure, *f. c.*, but I can communicate nothing as to the details of their formation, except that they are independent of one another and surrounded by a solid mass of cells.

IV. EARLY STAGES IN EGG DEVELOPMENT OF HIRCINIA ACUTA.

The "Loggerhead" sponge, Hircinia acuta, is very abundant in the shallow water round Green Turtle Cay, forming circular masses often of very large size, which contain great numbers of annelids, Alpheus, and other semi-parasitic forms. It is with eggs in this locality during September, and probably for a much longer period. My observations on this form are very few, dealing only with the development of the ovarian egg and the segmentation.

The mesoderm of Hircinia has in many regions a cartilaginous appearance, consisting of a clear non-stainable matrix containing cavities in which lie the cells. When the cell shrinks away from the wall of the cavity, the latter comes plainly into view, Pl. XXIII, Fig. 114 (bit of the mesoderm). Tracts of this sort are often found in which flagellated chambers are absent, and in such places egg cells frequently occur. In Fig. 114 is shown an egg cell (*o. ov.*) about twice the size of the surrounding mesoderm cells, and containing a large nucleus with a single nucleolus. The egg is enclosed by an incomplete follicle, composed of neighboring mesoderm cells which apply themselves closely to the wall of the egg cavity. During its increase in size the egg becomes stored with yolk, and its nucleus undergoes changes similar to those described for Tedanione.

To the few mesoderm cells enclosing the young egg, others are gradually added, and in this way a complete follicle is formed, Pl. XXIII, Figs. 115, 115[1], consisting of a single layer of cells. In Tedanione the follicular cells very early flatten out into a thin membrane, but in Hircinia this change does not take place until after the beginning of segmentation. In

Hircinia there is the same indication as in Tedanione, that the mesoderm cells bring food to the growing egg. Until segmentation begins the egg is thickly surrounded by mesoderm cells, Pl. XXIV, Figs. 116 and 119, which have large bodies full of fine yolk granules. Amongst these cells are scattered a few with very coarsely granular bodies, Fig. 119. The mesoderm cells are applied so closely to the follicle that the latter cannot be distinguished as a definite layer. All that can be said is that at this time the egg is surrounded by closely packed cells, arranged irregularly in two or three strata, the inner stratum doubtless representing the follicle of the very young egg, while the outer strata consist of mesoderm cells which have applied themselves to the follicle. The inner stratum of cells is, as may be seen in Fig. 116, often very irregular, and there are certain indications that the follicular cells are sometimes pinched off and engulfed by the egg — notice the very protuberant rounded cells projecting into the substance of the egg. Whatever be the precise manner in which food is conveyed from the surrounding cells to the egg, it seems pretty certain that food is so conveyed, and that this is the object of the migration of so many mesoderm cells to the immediate neighborhood of the growing egg. The egg at the time when it reaches full size is still surrounded by several strata of cells, Fig. 119. The inner stratum, however, soon becomes transformed into a follicular membrane, consisting of flattened but still rounded and protuberant elements, Pl. XXIV, Fig. 120. The cells of the outer strata gradually wander away, leaving the mesoderm round the segmenting egg not more abundantly supplied with cellular elements than is the mesoderm in most parts of the sponge.

The ripe egg of Hircinia is closely packed with yolk spheres of a large size, Fig. 119, which make their appearance in the developing egg-cell after a fashion essentially similar to that already described for Tedanione. In the finely granular body of the very young egg-cell, yolk spheres appear which are at first small, Figs. 115, 115[1], but which gradually increase in size, becoming at the same time more closely packed. In the egg shown in Fig. 116, the bulk of the yolk is still composed

of quite small spheres, but scattered about in the fine yolk are certain large rounded bodies which I take to be the first large spheres formed.

While the egg is still small, long before it reaches the full size, the single nucleolus gives place to two nucleoli peripherally placed, as in Pl. XXIII, Figs. 115 and 115[1]. The egg of Pl. XXIV, Fig. 116, has likewise two nucleoli, situated in the same way, but the section passed through only one of them. Between the two nucleoli there is a sphere of granular material, which stains much less deeply than the nucleoli themselves, and which is separated from the nuclear membrane by a clear space. When the egg attains its full size, one of the nucleoli is lost, leaving no perceptible trace behind. In the nucleus thus left with a single nucleolus, Fig. 117, the nuclear membrane could not be made out in my preparations, though the sphere of faintly staining granular material was obvious, Fig. 118 (portion of the periphery of such an egg as that of Fig. 117). The second nucleolus is then extruded, and may be seen lying in the yolk in the immediate neighborhood of the nucleus, which now consists exclusively of the sphere of granular material, separated from the yolk by a narrow clear space, Fig. 118.

Segmentation results in the formation of a solid morula. An early stage in the segmentation is shown in the section, Fig. 120, and two morulas are shown in Figs. 121, 122. In the latter morula one of the segmentation spheres has been retarded in its division, and is consequently of a much larger size than the rest. Scattered between the segmentation spheres and forming a layer round them, there will be noticed a peculiar granular stuff which stains feebly but which is very conspicuous in the sections. It is probably a precipitate from the fluid bathing the segmentation spheres, caused by the fixing fluid (Perenyi).

V. REMARKS ON THE MORPHOLOGY OF SPONGES.[1]

I have shown that in Esperella and Tedania the subdermal cavities, canals, and chambers develop as separate lacunae in

[1] The figures which illustrate this article, excepting Fig. 5, are borrowed. For their sources see description of the plates.

the parenchyma or mes-entoderm of the attached sponge, subsequently becoming connected into a continuous system. As regards the development of the canal system, such varying accounts are given by different authors that, were it not for the help lent by comparative anatomy, it would be quite impossible to form any idea of the fundamental morphology of sponges. Fortunately for the student entering this puzzling domain, comparative anatomy has in the hands of Haeckel, Schulze, and Polejaeff provided a standpoint from which the varying phenomena of development and structure may be viewed with at least a partially understanding eye. It may be that an increasing accumulation of facts will show that Haeckel's conception of the relation of the simple calcareous sponges to the complex horny and silicious forms is not well founded, and that Schulze's view of the parts played by the embryonic layers in producing the adult anatomy is not the true one. But at present it is only with the aid of these theories that one can form any clear conception of sponges in general, and so for the present at least we are bound to accept them.

Comparative anatomy points in no undecided manner to the phylogenetic path along which sponges have developed, and so permits us to construct a standard of ontogeny, with which we may compare the actual development of each species as we witness it to-day, and so be enabled to note the amount and kind of divergence (coenogeny) exhibited. That coenogeny is exhibited to a great degree in the embryology of sponges is evident from the various types of development described, and in the future much may be hoped from the study of a group like this for the understanding of the laws of development. For the present all we can do is to accept what seems the most probable phylogeny, recording the instances of supposed coenogeny as they are observed. Adopting this method, I have to regard the development of Esperella and Tedania (*i.e.*, the later development or metamorphosis) as far removed from the phylogenetic path. Before pointing out the pronounced coenogeny exhibited in the development of these sponges, it will be worth while to review briefly the evidence on which rests the current view of sponge morphology.

Evidence from Comparative Anatomy as to Sponge Phylogeny. — The strongest evidence offered by comparative anatomy lies in the series of forms, passing by gradations from very simple to complex types, found in the calcareous sponges (Haeckel 8, Polejaeff 20), and in the little group of silicious sponges, the Plakinidae, described by Schulze (26). A comparison of these forms goes to show that the simple Ascon sponge (Olynthus) must be regarded as the ancestral type of the group, and that by the continued folding of the wall of this simple form were produced the more complicated sponges. Further, the exceedingly complex silicious and horny sponges must be interpreted as colonies in which the limits of the individual can in many cases no longer be recognized.

The calcareous sponges offer a series of increasingly complex forms, which Haeckel divided into Ascons, Sycons, and Leucons. Haeckel's views on the relationship of these forms must be in great measure accepted to-day, though in certain respects, especially as regards the anatomy of the Leucons, later researches (Polejaeff, *l.c.*) have shown that he was not always in possession of the real facts of the case.

The simplest calcareous sponges, or Ascons, which serve as the basis for Haeckel's hypothetical sponge ancestor, the Olynthus, are too familiar to call for any description. The interesting form *Homoderma sycandra* (von Lendenfeld) may, however, be mentioned, in which the body is surrounded by radial tubes, after the fashion of a Sycandra, but with the difference that the central cavity as well as the radial tubes is lined with collared cells. A figure of this interesting sponge is accessible, in Sollas's article on Sponges in the Encyc. Brit. (or Zoölogical Articles by Lankester, etc., p. 40).

Homoderma bridges the way from the Ascon type to the simplest Sycons, in which the radial tubes are distinct from one another. A surface figure of such a Sycon (*Sycetta primitiva*) is given in Vosmaer(33), Taf. IX, taken from Haeckel's monograph. In the majority of Sycons, however, the radial tubes are not distinct, but are connected together more or less by strands of mesoderm covered with ectoderm (Pl. XXV, Fig. 1, transverse section of *Anamixilla torresi*). In this

sponge all the tubes are connected together, and the canals lying between them (Intercanals, *In. can.* in the figure) are complicated. Water enters the intercanals through the openings on the surface (surface pores, *s. p.*), and passes into the radial canals through the numerous chamber pores (*c. p.*).

The embryology of the Sycons as far as known confirms the belief that they are derived from the Ascons. Thus Sycandra raphanus passes through a distinctly Ascon phase (Schulze 25), the radial tubes appearing later as outgrowths. The actual development of complicated intercanals such as those of Anamixilla has never been witnessed, but a comparison of a large number of forms in which the connection between the radial canals varies within wide limits, makes it pretty certain that the intercanals of a form like Anamixilla are homologous with the simple ectodermic spaces between the radial tubes of Sycetta or Sycandra ciliata. It is exceedingly probable that the actual development of the complicated Sycons will show that the radial tubes are in young stages distinct from one another, and only later become connected together by bridges of tissue in such a way as to form complex intercanals. And so, we must at present regard the intercanals of a form like Anamixilla as lined with ectoderm.

Coming now to the Leucons, we find that Polejaeff's description of the anatomy of this family accords with their derivation from the Sycons quite as well as did Haeckel's more imaginative conception of the structure of these forms. Taking one of the simplest of Polejaeff's types, let us compare it with a Sycon. In Pl. XXV, Fig. 2, is shown part of a transverse section of *Leucilla connexiva.* Such a form is obviously derived from a Sycon by the evagination of the wall of the paragastric cavity at certain points (*x, x*). These evaginations give rise to numerous diverticula of the central cavity, which constitute efferent canals, *ef. c.* The radial chambers are at the same time thrown into groups, each group opening into one of the new diverticula. The intercanals (*In. can.*) penetrate as before between the several radial chambers, bringing water to the chamber pores (*c. p.*), the complexity of their

arrangement naturally having been increased by the folding of the wall of the paragastric cavity.

The increasing complexity in the Leucon family is brought about by the ramification of the primitively simple efferent canals, the radial tubes growing shorter and becoming in the most complicated types spheroidal chambers quite like the flagellated chambers of the non-calcareous sponges. In *Leucilla uter*, for instance, of which part of a transverse section is given in Pl. XXV, Fig. 3, the efferent canals exhibit branching of a simple character. But in such a form as *Leuconia multiformis* (transverse section, Pl. XXV, Fig. 4), the ramification of the efferent canals becomes exceedingly complex, and the radial tubes here appear as spheroidal flagellated chambers. The intercanals (or afferent canals, as they are called in all sponges but the Sycons) follow the efferent canals in all their windings, bringing water from the surface pores to the pores in the walls of the flagellated chambers.

The chief conclusions to be drawn from this anatomical comparison of the various forms of Sycons and Leucons are, that the afferent canals of Leucons are homologous with the intercanals of Sycons and are lined with ectoderm; that the flagellated chambers are homologous with the radial tubes; that increasing complexity is brought about by the ramification (or folding of the wall) of the efferent canals.

The canal system of a complicated Leucon, like *Leuconia*, is essentially like that of a common silicious or horny sponge (having flagellated chambers, afferent and efferent canals), except in the one respect that in the Leucon there is a single central cavity opening by a terminal osculum, while in most silicious and horny sponges there are several oscula leading into as many spacious efferent cavities. But here the disposition of the calcareous sponge to form indubitable colonies helps us out, for if we compare the silicious or horny sponge with a colony of Leucons instead of with a single one, we find that its derivation from such simple symmetrical forms is made easy. Robbed of its details, a silicious sponge of the character of Esperella, Tedania, or Tedanione, exhibits a structure illustrated by the diagram of an hypothetical silicious

sponge shown in Pl. XXV, Fig. 5. In the section drawn, three main efferent canals (*ef. c.*) are shown, each with its osculum (*os.*) and its very irregular set of branches (*ef. c'.*, *ef. c''.*, *etc.*), on the walls of which open the flagellated chambers (*f. c.*). The pores on the surface of this sponge (*s. p.*) lead into wide chambers (*s. d. c.*), the so-called subdermal cavities, from which run the afferent canals (*af. c.*), carrying water to the pores in the walls of the flagellated chambers. The distinction between subdermal cavities and afferent canals is more or less artificial, for the sharpness with which they are marked off from one another varies within wide limits. They are both parts of the same system, the subdermal cavity being merely a main afferent canal, which is especially enlarged in a tangential direction. The water may enter the chambers in some cases directly from the subdermal cavities, but for the most part it is carried to the chambers by the afferent canals, which branch and twist about, following the irregular course of the efferent canals. The mesoderm between the two sets of canals is reduced to comparatively narrow trabeculae, in which lie the flagellated chambers, arranged in a much folded but still single layer. The spicules which are not shown in the figure are in the mesoderm, either scattered about or united into a meshwork or a series of bundles. The genital products are also to be found in the mesoderm, scattered about, as a rule, in any part of the body.

The structure of a horny sponge, such as the sponge of commerce (Euspongia), is essentially similar to that of the hypothetical silicious sponge I have just described. The differences concern especially the skeleton and the precise manner in which the flagellated chambers are connected with the canals. In the horny sponges the silicious spicules give place to a meshwork of horny fibres, which lie in the mesoderm between the canals to which they lend support, and to the course of which their arrangement is adapted. The flagellated chambers in Euspongia, as in many other horny and silicious sponges, do not open directly into spacious efferent canals (as in Fig. 5), but indirectly by means of special canals, one of which runs from each chamber. And so it is with the afferent canals,

which in these sponges send a special little canal to each chamber (see Schulze's figure of Euspongia. *Zeit. f. Wiss. Zool.*, Bd. XXXII, Taf. XXXVI, Fig. 2). The difference between the two canal systems is easily explained, that of Euspongia being derived from the type shown in Fig. 5, by the pushing out of minute diverticula from both afferent and efferent canals.

Having now obtained a generalised idea of a complex non-calcareous sponge, it will be found a simple matter to compare such a form with a Leucon colony, of which I give a surface figure, taken from Vosmaer, in Pl. XXV, Fig. 6. The structure of the silicious sponge is readily understood if we suppose it to be a colony, in which the limits of the individuals have been lost or obscured by the increasing thickness of the walls of adjacent individuals. This increasing thickness would finally result in a more or less complete fusion of the members of a colony into an undivided mass with oscula scattered over the surface. Each of the main efferent canals of the silicious sponge is homologous with the paragastric cavity of a single Leucon. Both the canal and its set of branches, though, are extremely irregular, having completely lost the symmetry of the ancestral type. The flagellated chambers, however, still bear the same relation to the efferent canals as they did in the Leucon, *i.e.* they are simple diverticula of the canal wall. The system of afferent canals is obviously homologous with the same system in the Leucons, bearing identically the same relation as in the latter group, both to the flagellated chambers and the efferent canals. The subdermal chambers, communicating with the exterior by numerous pores, though a late acquisition, are found in certain Leucons, *e.g.* Eilhardia Schulzei (Polejaeff, Pl. IX).

In many of the non-calcarea the colonial nature of the sponge is indicated by the presence of elevations (oscular tubes or papillae) bearing oscula on their summits. But the number of oscula is not always to be taken as indicating the number of individuals of which the sphere is composed, for the colonies of calcareous sponges show plainly that the budding individuals do not always develop oscula. And on the other hand there

are certain indications in the silicious sponges (p. 8) that in the adult, oscula may be developed almost anywhere. Such facts make it impossible to fix upon the number of component individuals in any sponge. Perhaps the nearest approach made in other groups to the formation of colonies, in which the personality of the component individual is so nearly lost, is found in corals like Maeandrina, in which the united gastric cavities of the polyps form continuous canals, perforated at intervals by mouths.

We therefore reach the conclusion that the higher sponges (Non-Calcarea) have been derived from colony producing, symmetrical forms, in which the evaginations of the primitively simple paragastric cavity had already taken the form of efferent canals and flagellated chambers, that is from forms allied to the existing Leucons. And further we come to the conclusion that the subdermal cavities and afferent canals are homologous with the intercanals of Sycons, and hence, phylogenetically at least, are infoldings of the ectoderm. The whole efferent system, canals and flagellated chambers both, on the contrary is homologous with the same system in the calcareous sponges, and is endodermic.

This conclusion as to the parts played by the germ layers in producing the adult non-calcareous sponge, is the one enunciated by Schulze in his classical paper on the Plakinidae (p. 438). In this little family of silicious sponges Schulze finds a genus, Plakina, the three species of which form links in a chain of increasing complexity, showing quite as plainly as do the calcareous sponges that the afferent system is derived from ectodermal infoldings, and the efferent from endodermal outfoldings.

The Plakinidae are Tetractinellids. The three species of Plakina are small encrusting sponges found in the Mediterranean. They all adhere to the under side of stones, shells, *etc.* A vertical section of the simplest species, Plakina monolopha, is given in Pl. XXV, Fig. 7. There is a continuous basal cavity crossed by strands of tissue, in which lie developing eggs. From the cavity run vertical efferent canals (*cf. c.*), which are simple or very slightly branched, and into which open the

flagellated chambers. The afferent canals (*af. c.*) are spacious cavities opening on the surface by wide mouths. The periphery of the sponge forms a continuous rounded rim (the "ringwall," *r. w.*), and the oscula, one or several, are situated here. The surface of the sponge inside the ringwall is divided up into low rounded elevations, caused by the upper ends of the efferent canals, between which lie the wide apertures leading into the afferent canals. Schulze was fortunately able to observe the main features in the development of this interesting form. There is a solid swimming larva which settles down, forming a flat circular mass. A central cavity appears in the mass, the lining cells becoming columnar, and the sponge is thus transformed into a flat three-layered sac, Pl. XXV, Fig. 8, the three layers being respectively ectoderm, mesoderm, and entoderm. The flagellated chambers appear in a single layer round the central cavity, into which they open. They are very probably formed as diverticula of this cavity. Schulze did not follow the development further, but a comparison of the adult with the sac-like young form makes it pretty certain that the young form undergoes a process of folding which gives rise to the efferent and afferent canals of the adult; or in other words the efferent canals arise as vertical evaginations of the sac-like stage. The afferent canals are consequently to be regarded as lined with ectoderm.

A vertical section of the second species, Plakina dilopha, is shown in Pl. XXV, Fig. 9, and of the third species, Plakina trilopha, in Pl. XXV, Fig. 10. The oscula in these species are not situated at the periphery as in Plakina monolopha, but at some distance internal to it; and in them the efferent canals do not form projections on the surface as in the first species. On comparing the canal systems in Figs. 7 and 9 it is seen that Plakina dilopha has probably been derived from Plakina monolopha by an increase in the thickness of the mesoderm lying beneath the surface of the sponge. The wide afferent canals of Plakina monolopha become transformed into the narrow efferent canals of Plakina dilopha. In other respects there has been no great change. (Schulze, pp. 438 and 439.)

Plakina trilopha goes a step farther in the direction of complexity than does Plakina dilopha. It has probably been derived from the latter species (compare Figs. 9 and 10) by the appearance of secondary folds in the radial efferent tubes; by a transformation of the basal cavity into a system of lacunae, owing to the increase in number of the connecting strands of tissue between the basal layer and the part of the sponge containing the flagellated chambers; and by a complication in the afferent canals in consequence of which they do not open each by a single aperture but by a number of small apertures, the surface pores (*s. p.*).

Schulze's conclusion that these species all lie in one line of descent, that is that the second has been derived from the first, and the third from the second, receives as much support from a study of the spicules, as of the canal system. But on this head, reference will have to be made to the original paper.

From comparative anatomy we conclude that the phylogeny of the sponges is something as follows: The Olynthus is the common ancestor of the group. The outgrowth of radial tubes gave rise to the Sycon type. The growth of the mesoderm and development of new endodermic diverticula, coupled with the metamorphosis of radial tubes into flagellated chambers, produced the Leucons. The non-calcareous sponges have been derived from types with a canal system more or less like that of the Leucons. And the conclusion with regard to the germ layers is that the efferent system is entirely endodermic, and the afferent system entirely ectodermic.

Embryological Evidence. — Let us see now how far the known facts of development support the above conclusions. The evidence from the calcareous sponges (Sycandra passes through Olynthus stage) has already been given. Several of the non-calcareous sponges (Oscarella lobularis, Reniera filigrana, Chalinula fertilis, Plakina monolopha) run through a stage known as the Rhagon (Sollas), which it is permissible to regard as the ontogenetic representative of the Sycon type. The rhagon of Oscarella (Heider 9) is shown in Pl. XXV, Fig. 11. Regarding it, as seems best, as equivalent to the Sycon type, it will be noticed that the radial tubes of the

Sycon are coenogenetically replaced by flagellated chambers. The rhagon of Oscarella is formed as an invaginate gastrula, which attaches mouth down. The gastrula mouth closes, and the osculum is a new formation. The flagellated chambers arise as true diverticula of the central cavity. The adult Oscarella, the canal system of which is not far removed from that of Plakina monolopha, is very probably formed from the rhagon, by the development in the latter of a number of simple diverticula from the central cavity. These diverticula are the efferent canals into which open the flagellated chambers. The ectodermic spaces between the efferent diverticula become the afferent canals. The adult Oscarella, like Plakina monolopha, is directly comparable with a simple Leucon. The development of Oscarella in large measure confirms the conclusions drawn from comparative anatomy, and may therefore be considered as phylogenetic.

The development of Plakina monolopha (Schulze) has already been described. The sac with its single layer of flagellated chambers opening into it, is a rhagon, and may be taken as representing the Sycon stage. The adult Plakina itself is the Leucon stage.

In Reniera filigrana (Marshall 18) there is a solid swimming larva, which after attaching acquires a central cavity with an apical osculum. The flagellated chambers arise as diverticula from this cavity. Thus in this sponge also there is a rhagon stage. But in one matter we strike upon a coenogenetic modification. The afferent canals, instead of being ontogenetically formed from the ectoderm, as they seem to have been phylogenetically, are really formed from endodermic diverticula, which grow outwards, meeting the surface epithelium.

In Chalinula fertilis (Keller 10) there is also a solid larva in which a central cavity is hollowed out. But in this sponge the flagellated chambers of the rhagon stage do not arise as endodermic diverticula, but are formed independently from solid groups of mesoderm cells. This origin of the flagellated chambers must be regarded as coenogenetic. The fact that the mesoderm may take upon itself the function of forming organs ordinarily formed by the entoderm would seem to

indicate that the two layers are of much the same nature. This essential similarity between the two layers has always been maintained by Metschnikoff, not only on the ground of development, but for physiological reasons as well. Thus in young Spongillas when the water became bad, he has witnessed the entire disappearance of the flagellated chambers, the sponge then consisting of ectoderm and mesoderm alone. With a fresh supply of water the chambers reappeared (12, p. 375). Again, after feeding carmine in an excessive amount to Halisarca pontica, he found that the canals and chambers entirely disappeared, the whole body of the sponge inside the ectoderm consisting merely of a mass of amoeboid cells full of carmine (*ibid.*, p. 372). The development of the afferent system in Chalinula was not worked out with certainty.

The embryology of the preceding sponges, in which a rhagon type is developed, agrees pretty well with our general notions of sponge phylogeny. But there are other sponges, the development of which has been so excessively modified as no longer to be of any use as finger-posts to phylogeny, but which afford an excellent field for the study of what may be called the methods of coenogeny. In Halisarca Dujardinii (Metschnikoff 12), for instance, there is a solid larva in which the canals appear as so many separate lacunae surrounded by parenchyma (mes-entoderm) cells. The canals only subsequently acquire a connection with each other.

In Esperia (Maas 16), the subdermal spaces, canals, and chambers arise separately as lacunae in the parenchyma. The chambers are formed from aggregations of small cells (which Maas believes, on what seems to me insufficient evidence, to be ectoderm cells of the larva that have migrated into the interior). The efferent canals, Maas thinks, are formed from similar cells.

In Esperia, according to Yves Delage (36), the chambers arise by division of special mesoderm cells. The epithelium of the canals comes from the larval ectoderm, which migrates into the interior. In Spongilla, according to the same author, the ectoderm cells of the larva are *engulfed by mesoderm cells*, and then become the lining cells of the flagellated chambers!

In young Stellettas (Sollas 28, pp. xvi–xvii) the subdermal cavities seem to arise as lacunae in the parenchyma. And in the external buds of Tethya maza, Selenka (29) believes the subdermal cavities have a similar origin.

In Spongilla, according to Götte (6), the subdermal cavities and canals are formed as independent lacunae in the parenchyma, and the flagellated chambers are formed from groups of cells, each group (and chamber) being produced by the budding of a single large mesoderm cell. This account of the development of Spongilla is contradicted by Maas (14) who brings Spongilla in line with those forms having a rhagon. Maas describes in the larva a single central cavity from which the chambers arise as diverticula, the central cavity persisting in a modified shape as the efferent system of canals. The subdermal spaces arise as ectodermal invaginations, from which the afferent canals are formed as ingrowths. Thus according to Maas in the ontogeny of Spongilla, the whole afferent system is formed from the ectoderm and the whole efferent system from the endoderm. Ganin's earlier account (7) likewise makes the chambers originate as diverticula from a main endodermic cavity.

In the metamorphosis of a larva, which probably belongs to Myxilla, Vosmaer (34) finds the subdermal cavities begin as fissures which gradually become wider, and the canals and chambers likewise appear as intercellular spaces.

Finally, in the gemmule development of Esperella and Tedania I find that subdermal cavities, both sorts of canals, and the flagellated chambers, all arise as independent lacunae in the parenchyma.

Accepting as ancestral the development (*i.e.* later development or metamorphosis) of Oscarella and Plakina monolopha, the various coenogenetic modifications which appear in other sponges may be classified as follows :—

1. The efferent canal system, instead of arising as a single cavity which throws out diverticula, may be formed as so many distinct cavities which subsequently unite (Esperella, Tedania, Esperia [Maas], Halisarca Dujardinii, Myxilla).

2. The flagellated chambers, instead of arising as endodermic

diverticula, may be formed from groups of mesoderm cells (Esperella, Tedania, Chalinula fertilis, Myxilla).

3. The afferent canals, including the subdermal cavities, instead of being formed as invaginations from the ectoderm, arise as lacunae in the mes-entoderm (Esperella, Tedania, Esperia, Stelletta, Myxilla). In Reniera filigrana (Marshall) they are formed as entodermic diverticula.

The coenogenetic development of the flagellated chambers and efferent canals suggests, as I have said, an essential similarity of nature in the so-called entoderm and mesoderm of sponges. This belief, so long upheld by Metschnikoff, derives some of its strongest support from this author's physiological investigations (see *ante*, p. 359), as well as from the fact, first emphasized by Metschnikoff and Barrois, that in the most common sponge larva (the solid larva) mesoderm and entoderm form a single indivisible layer.

And likewise the development of the afferent system of canals, in some sponges from the ectoderm, in others from the mes-entoderm, may possibly be taken as meaning that even these two primary layers (the outer and the inner) are not distinctly differentiated from each other in the sponges ; or, in other words, that the mes-entoderm is still enough like the ectoderm to form organs ordinarily produced by the latter layer.

There is another (hypothetical) way of explaining these phenomena, which consists in supposing that ectoderm cells of the larva migrate into the interior, and, although indistinguishable from the surrounding mes-entoderm cells, alone take part in forming the afferent canals. Similarly we may suppose that in the solid mass which constitutes the parenchyma of Esperella there are two radically distinct classes of cells, one of which is potentially gifted with the power of forming efferent canals and flagellated chambers, while the other has not this power and must remain as amoeboid mesoderm. But this is pure hypothesis.

The result of this critical examination seems to be that the Olynthus must be regarded as the ancestor of sponges (Haeckel, Kalk-spongien), and that the entoderm and mesoderm are not

sharply differentiated from each other as they are in the higher animals (Metschnikoff, *Spong. Studien*, p. 378).

Origin of the Olynthus. — The prevalence of the solid larva in sponges and hydromedusae, coupled with the widespread presence of intracellular digestion in the lowest metazoa, led Metschnikoff years ago to the belief that the solid larva represents the ancestral form of the metazoa, while the gastrula is a coenogenetic modification (12, 13). To my own mind, all the facts that we know indicate this conclusion to be well founded. This hypothetical ancestral form was named Parenchymella (changed later to Phagocytella). I may be permitted to recall its leading features as deduced by Metschnikoff. The animal consisted of an outer layer of flagellated cells and an inner mass of amoeboid cells. The digestion was intracellular, the food being taken in through openings scattered over the surface. A central cavity having a special opening to the exterior was a later acquisition, the opening being in all probability one of the small apertures especially enlarged. This solid ancestor of the metazoa, Metschnikoff derives from colonial forms like Protospongia. Barrois (1), as early as 1876, stated his belief that the ancestor of sponges was a solid animal composed of two layers, the outer representing the ectoderm, the inner mass representing a parenchyma, from which have developed the ectoderm and mesoderm of higher animals (p. 78).

According to this view, the early development of Plakina (or Reniera, Chalinula, etc.) gives the first chapters in the history of the group of sponges more faithfully than does a form like Oscarella or Sycandra. In the former sponges, it will be remembered, there is a solid larva hollowed out to form a three-layered sac, which then breaks open to the exterior, forming the osculum. In the latter there is an invaginate gastrula which settles mouth downwards, the gastrula mouth subsequently closing and the osculum appearing as a perforation at the upper end of the sac. In these forms, Oscarella and Sycandra, we have to suppose that the Parenchymella stage is skipped, the central cavity (which properly belongs to the Olynthus stage) being precociously developed coincidently with

the immigration of the entoderm. The blastopore of the sponge gastrula on this view does not represent a primitive organ (Urmund), but merely comes into existence owing to the highly modified method of forming the entoderm. We do not, therefore, have to construe the Oscarella development (with Heider and Sollas) as meaning that a gastraea ancestor settled mouth downwards, and that the mouth gradually became functionless, finally closing up, while a new series of openings, pores and oscula, was established.

The only remaining point I wish to speak of is the relation of the sponges to the coelenterates. That the two groups have had a common ancestor in the Parenchymella is highly probable, but the similarity between the Olynthus and the simplest coelenterates inclines one to go further and, at any rate, homologize the paragastric cavity of the former with the gastric cavity of the latter. This, of course, is done by authors like Sollas, who derive both groups from a gastrula-like ancestor. Whether the osculum of the Olynthus is also homologous with the coelenterate mouth, as Haeckel originally held, is a question which needs for its answer more facts relating to the actual use to which the osculum is put in the simplest sponges. Sollas and Heider urge against the homology, the fact that the coelenterate larva attaches by the pole opposite the blastopore, while in the sponge larva the blastopore is at the pole of attachment. But this I cannot regard as a very strong argument, for I do not believe that the opening into the gastrula cavity represents a primitive organ (mouth of an ancestor). And if it does not, but is merely an incidental product of a particular mode of endoderm-formation, it becomes evident that the position of the blastopore at opposite poles in sponge and coelenterate larvae has no bearing on the question of homology between mouth and osculum.

It is, moreover, doubtful if any such sweeping distinction can be drawn between the larvae of the two groups, for it is a question whether any sponge larva has a particular pole by which it must attach. Even in Sycandra, Schulze records (25, p. 274) that exceptional cases occur which cannot be regarded as pathological, in which fixation takes place not by

the gastrula mouth, but on the side. Fixation may also be delayed until the gastrula mouth has closed and spicules have begun to appear, in which case it is not stated by what part the larva attaches. In the solid larvae of silicious sponges the variation is much greater. Such larvae attach in some cases by the posterior pole, in others by the anterior pole, and yet in others on the side. All these variations may occur in larvae of the same species, for instance Maas records (16) that in Esperia he observed fifteen individuals attach by the posterior pole, seventy by the anterior pole, and five or six on the side. It thus appears that in the larvae of silicious sponges at any rate there is no constant point of attachment.

VI. REMARKS ON THE GEMMULE DEVELOPMENT OF SPONGES.

1. *Asexual Development in General of the Sponges.*

The asexual method of development exhibits itself in sponges in a variety of ways. Besides the simple coelenterate-like process of budding which leads either to the production of new individuals or to the formation of more or less clearly marked colonies, and which is seen at its simplest in the calcareous sponges, the following instances of non-sexual reproduction may be called to the mind of the reader.

In Oscarella, Schulze (27) found that hollow outgrowths were constricted off from the surface of the sponge, which led a free-swimming life for several days, ultimately sinking to the bottom and developing each into a new sponge. The outgrowth contained a diverticulum from the canal system of the mother, and the wall of the outgrowth agreed in structure with the wall of the parent sponge, *i.e.* it contained flagellated chambers with the short afferent and efferent canals. The method of bud formation here employed seems to be fundamentally the same as that exhibited in the calcareous sponges and the coelenterates.

The propagation of sponges by cuttings may be mentioned in this connection. The experiments of Oscar Schmidt and those of the U. S. Fish Commission (made on the Florida

coast) have demonstrated that small pieces or cuttings of the commercial sponge have the ability to reproduce the entire organism. Cuttings from Tedania, which were suspended from mangrove roots in one of the "sounds" of Green Turtle Cay, grew very perceptibly in a month.

In Tethya and Tetilla (Selenka 29, Deszö 3, 4) external buds are produced in a curious way. The buds consist of a solid mass of cells, and are formed in the peripheral region of the mother beneath the skin. As they mature they are gradually pushed out of the body along the spicules of the mother, until their only connection with the parent is through a slender stalk made up chiefly of these spicules. The bud then drops off. Deszö's account of the early formation of these structures is extremely interesting, although his recorded facts scarcely seem to warrant his inferences. According to Deszö, the bud or gemmule is derived from a single cell, which undergoes a segmentation, and growing all the while gives rise to a solid morula. By the time the original cell has divided into four, a differentiation of "germ layers" takes place: one of the four cells constitutes the entoderm, the remaining three the ectoderm. The ectoderm then grows entirely round the entoderm cell. Cell multiplication continues, the primary entoderm cell producing a solid mass of entoderm, surrounded by a single layer of ectoderm cells. The latter layer then splits off from its inner surface a layer of mesoderm, and itself gives rise to the external epithelium of the mature bud and to a stratum of tissue just beneath the epithelium, in which small asters (spicules) are developed. Deszö's interpretation of certain cells as constituting distinct germ layers, is not very strongly supported by his figures. Vosmaer's criticism in regard to this point may be given: "Es ist wohl klar dass für die Deutungen der Zellen, wie sie Deszö vornimmt, kein Grund vorliegt" (Bronn's Class. und Ordnung, p. 427). Deszö points out the importance, from a biological standpoint, of the discovery of germ layers in a non-sexually produced embryo, and calls to mind a similar discovery by Oscar Schmidt in the developing buds of Loxosoma. Schmidt's account of the development of the Loxosoma buds (23, 24),

however, has not been confirmed by later investigators. On the contrary Seeliger finds that the bud is not derived from a single cell, but is formed in a very different way. According to Seeliger (30, 31) the bud, both in Loxosoma and Pedicellina, is formed as a papilla of the body wall (or stolon, in the case of Pedicellina), an invagination at the end of the papilla giving rise to the atrium and alimentary tract. The mesoderm of the bud is derived from the mesoderm of the parent, the ectoderm of the bud is derived from the ectoderm of the parent; and the only new formation is the entoderm, which is produced by an *invagination of the adult ectoderm.* Seeliger's account destroys the possibility of drawing a parallel between sponge gemmules, which develop germ layers, and the buds of Loxosoma.

The internal buds or gemmules of the fresh-water sponges have been known since the time of Linnæus, but their precise origin is still open to discussion.

The ripe gemmule consists of a solid mass of polygonal cells, full of yolk, surrounded by a complex capsule. The capsule is perforated by an opening (hilum), through which in the spring the cellular mass creeps out, developing into a new sponge. The capsule is composed of an inner and outer cuticular layer, between which is a layer containing skeletal elements (amphidisks or other spicules). According to Götte (6), all the cells in a particular region of the body of the parent sponge, not only those of the mesoderm, but those of the flagellated chambers and canals as well, become transformed into a mass of yolk-containing cells, which constitutes the gemmule. According to Marshall (19), however, the gemmule is formed exclusively from an aggregation of mesoderm cells. In whichever way formed, the young gemmule becomes differentiated into two layers, an inner mass of larger cells full of yolk, and a peripheral layer of cells (Götte). According to Götte, the peripheral layer of cells secretes the inner and outer cuticle, and gives rise to the amphidisks. According to Wierzejski (cited from Vosmaer 33, p. 429), the peripheral layer assumes the character of a columnar epithelium. Between it and the central mass appears the inner cuticular layer. The spicules and amphidisks are formed entirely outside the gem-

mule, in the parenchyma of the mother sponge, and only after formation do they get into the peripheral layer of the gemmule. The cells of the latter layer, however, secrete the outer cuticle, and subsequently entirely disappear.

Gemmules fundamentally like those of Spongilla have been found in certain marine sponges by E. Topsent (32). The sponges in which the gemmules were observed are Chalina oculata, Chalina gracilenta (I have seen them myself in Chalina arbuscula, Verrill, during the summer at Woods Holl), Cliona vastifica, Suberites ficus. The gemmules consist of a mass of cells surrounded by an envelope of horny matter (keratode), the protoplasm of the cells being full of highly refractive granules (presumably yolk). An earlier notice (1880) of the existence of such gemmules in marine sponges is contained in Claus's Grundzüge, Bd. I, p. 214: "Auch bei den Meerenschwämmen ist die Vermehrung durch Gemmulae verbreitet. Dieselben entstehen unter gewissen Bedingungen als kleine von einer Haut umschlossene Kügelchen, deren Inhalt im Wesentlichen aus Schwammzellen und Nadeln gebildet ist und nach längerer oder kürzerer Zeit der Ruhe nach Zerreissen der Haut austritt."

In Craniella are found embryos which Vosmaer interprets as gemmules (Bronn's Class. und Ord., p. 428). Sollas has, however, seen the same structures and regards them as egg embryos (28, pp. 33–39).

Oscar Schmidt (22) stated it as his opinion that there was no true segmentation in the eggs of horny and silicious sponges, but that the egg very early lost its cellular character. It seems probable that Schmidt had seen cases of gemmule development, more or less like the development of Esperella and Tedania, as described by myself.

The ciliated larvae of species of Esperia (Esperella) have been repeatedly seen and studied (Metschnikoff 11, Carter 2, Schmidt 22, Maas 16, Yves Delage 36). It has been assumed in all cases that the larva observed was an egg larva, and of course this may have been true. The close resemblance, between the larvae observed by Maas and myself, suggests, however, that the former larvae were, like mine, gemmule larvae.

2. *Comparison between the Egg Larva and Gemmule Larva of Silicious Sponges.*

Resemblance of the two kinds of larvae. — A comparison of the gemmule larvae I have described, with the egg larvae of silicious sponges, reveals the fact that the two are similar in essential respects. These essential points are the presence and character of the germ layers, and the peculiar differentiation of one pole. The similarity between the two sorts of larvae will be seen after a brief survey of what is known concerning the egg larvae of silicious sponges.

The larva of Isodyctia (Barrois 1) is a solid oval larva (parenchymella). Except at the posterior pole it is everywhere covered by a layer of columnar ciliated cells, the ectoderm. At the posterior pole, according to Barrois, the ectoderm is absent and the inner mass (mes-entoderm) is laid bare to the exterior. In this larva as in others in which it has been claimed that the entoderm is exposed to the exterior through a break in the ectoderm, recent investigations (especially Maas's study of the flattening of the columnar epithelium in Spongilla, and the facts recorded in the descriptive part of the present paper) make it probable that the mes-entoderm is really not laid bare but is covered by a layer of flat ectoderm cells. The unciliated posterior pole of the Isodyctia larva is made further noticeable by a deposition of red pigment in its cells, and the cilia immediately surrounding it are unusually long, forming a conspicuous circle. In this larva the posterior pole ("calotte") is at no time ciliated nor, as I understand the author, is it ever covered (not even before birth) with columnar ectoderm. The resemblance between this larva and the gemmule larva of Esperella is, to say the least, striking.

The larva of Desmacidon (Barrois 1) is very like that of Isodyctia. As in the latter sponge, the columnar ciliated ectoderm is lacking at the posterior pole which is further distinguished by its pigment and by a circle of long cilia immediately surrounding it. Unlike Isodyctia, the larva of Desmacidon when set free is ciliated all over. But pigment

accumulates at its posterior pole, and the cilia and "son revêtement cellulaire" disappears (this, as I have said, is probably to be interpreted as meaning that the columnar ciliated ectoderm of this pole becomes transformed into flat unciliated cells). In this last detail the Desmacidon larva is more like Tedania than Esperella. It will be remembered that in the Tedania embryo all the ectoderm cells become columnar, those at the posterior pole subsequently flattening out. In Desmacidon and Isodyctia both, the ectoderm is subsequently "broken through" at the anterior pole also. But this, it would seem, is only the first step in the general flattening of the ectoderm.

The larva of Reniera filigrana (Marshall 18) is a solid oval larva with a pigmented pole, and covered at first uniformly with columnar ciliated ectoderm. Unlike the two preceding sponges, the pigmented pole is the anterior. The ectoderm "bursts" at the pigmented pole, and the mes-entoderm is laid bare. Subsequently the ectoderm "bursts" at the opposite pole, and at about the time of fixation the whole ectoderm flattens.

In Chalinula fertilis (Keller 10) there is a solid larva essentially like the gemmule larva of Esperella and Tedania, in that the columnar ciliated ectoderm is absent at the posterior pole. According to Keller, this larva is derived from an epibolic gastrula, and the cells occupying the posterior pole are a part of the mes-entoderm, which is here from the start exposed to the exterior. My observations on the way in which this pole is formed in Esperella and Tedania, make it probable, I think, that the surface cells in this region of the Chalinula larva are ectodermic. Indeed, for a short time after birth the posterior pole is ciliated, but the cilia are, however, soon lost. Believing, as I have said, that the inner mass of cells is from the beginning exposed to the exterior at the posterior pole of the embryo, Keller naturally regards this region as a blastopore.

Vosmaer (34) describes the development of a larva which probably belongs to the genus Myxilla. The larva is solid and is covered with a cylindrical epithelium. A portion of the surface loses its cilia, the cells becoming cubical. Attachment

takes place in the region of the cubical cells, and the larval epithelium is not lost, but is modified cell by cell.

The larva of Amorphina (Schmidt, 22) is a solid larva ciliated all over. The cilia are lost at the posterior end. In the same paper, Schmidt describes the larva of a species of Esperia. The larva is solid and is ciliated all over. The cilia are lost at one pole, the spicules collecting at this pole. The larva of Reniera (Schmidt, *l. r.*) is a solid ciliated form with a deeply pigmented pole. The cilia on the pigmented pole are lost. There are other observations by Metschnikoff (11) and Carter (2) to the effect that the ectoderm is absent at the posterior pole of the larvae of silicious sponges. In these cases, as in the case of the Myxilla larva described by Vosmaer, it remains doubtful until the early development is known, whether the larva is really an egg-larva.

It will be seen that the larvae of the above-mentioned silicious sponges agree in fundamental respects. They all consist of two germ layers: an inner parenchymatous mass (mes-entoderm) and an outer layer of columnar ciliated cells (ectoderm). At one pole, usually the posterior, the ectoderm is apparently absent, the appearance being probably due to the fact that at this pole it is composed of flat unciliated cells. Like the egg larvae, the gemmule larvae (of Esperella and Tedania) consist of two germ layers, an inner parenchymatous mass (mes-entoderm) and an outer layer of columnar ciliated cells (ectoderm), the columnar ciliated cells giving place to flat unciliated cells at the posterior pole which is thus differentiated. It is plain that the two sorts of larvae agree in essential structure.

There are other silicious sponges, Spongilla (6, 14), the tetractinellid form Plakina monolopha (26), and Tedanione (see *ante*, p. 345), in which the larva has not the peculiar differentiation of one of the poles which is seen in the above-mentioned forms. But this differentiation is so common that it may fairly be considered as typical of a large though ill-defined group of sponges.

Cause of the Resemblance. — Accepting as a fact the resemblance between the egg and gemmule larvae in the possession

of germ layers and the differentiation of one of the poles, we must now put the question as to the cause of the resembance.

We have for long been accustomed to regard the two primary germ layers of an embryo as representing the primitive metazoan organs, *i.e.* the outer (nervous) and inner (digestive) layers of a simple two-layered form. It is possible that this view is not an entirely correct one, and that many so-called germ layers are not the ontogenetic representatives of the layers of the metazoan ancestor. And the occurrence of germ layers in an asexually produced embryo may possibly be interpreted as favoring the latter belief. It seems to me, however, that, while it is perhaps permissible to *suspect* the doctrine that the primary germ layers are homologous (I refer of course to the general homology maintained by Balfour in his *Comp. Embr.*, Vol. 2, p. 286) throughout the metazoa, we are not at present in a position which would warrant our giving up the doctrine. Certain it is that some form of two-layered embryo is found in every group, and that the various forms may be considered as modifications of a type ; and, to my mind, the best explanation of these facts is still the old one, that the germ layers are inheritances from a far distant two-layered ancestor.

Accepting the premise that germ layers are not independently acquired, but are inheritances from a common stock, we reach the conclusion that an asexually developed embryo (sponge gemmule) can reproduce features of a far distant ancestor (germ layers).

Coming now to the second point of resemblance (differentiation of a pole) between the egg and gemmule larvae of silicious sponges, we have first to ask ourselves, what is the meaning of this curious differentiation of one of the poles in the egg larva itself. This question I am quite unable to answer. Barrois (1) and more recently Keller (10) have regarded the unciliated pole as a blastopore, thus making it possible to compare the larva of silicious sponges with the amphiblastula of calcareous sponges. The basis on which their view rests is, that the endoderm at the pole in question is exposed to the exterior, and this, it is pretty certain, is not the case. The differentiation of the pole can have no such deep-seated morphological

significance as is advocated in this theory. It is probably an adaptive feature acquired within the group of silicious sponges. Now whether the gemmule larva has independently acquired this adaptation, is open to discussion. I am inclined to believe that it exhibits the feature in question, for the same reason that it develops germ layers: both features come to it as inheritances, the latter from a far distant ancestor, the former from a comparatively near one, both features being of actual physiological use to the larva.

To repeat, the conclusion I reach in regard to the marked resemblance between the egg larva and gemmule larva of silicious sponges is, that it is one not due to independent adaptation to similar circumstances, but to inheritance from a common source. What I believe I have found is, a bud embryo exhibiting ancestral traits. To illustrate by means of an imaginary example: suppose the bud of a simple ascidian, instead of developing directly into a new ascidian, first developed into an ascidian tadpole, with its notochord, nervous system, *etc.*, we should then have, I take it, a parallel case to the gemmule development of sponges. Only, in the imaginary case there could be no doubt of the larval features being inheritances, while in the case at hand I am free to admit that this view could be disputed.

The exhibition of ancestral traits in a bud embryo is perhaps a very rare phenomenon. The supposed occurrence of this phenomenon in Loxosoma has been shown to be without foundation, and Deszö's claim that it does occur in the development of Tethya buds cannot, in view of the insufficient evidence, be admitted. (Moreover, if Deszö's statement that the Tethya bud is derived from a single cell, be a fact, such a cell could properly be regarded as an undeveloped germ cell, and the "budding" of Tethya would then be a process analogous to the "sporogonie" discovered by Metschnikoff in Cunina proboscidea (38), or to the paedogenesis of the Cecidomyia larva, and therefore not a case of asexual reproduction.) In fact but a single instance of this phenomenon, as far as I know, has been recorded for the animal kingdom, previously to the appearance of these observations. The case referred to

is the remarkable development of the hydromedusa, Epenthesis McCradyi, described by Brooks (39). The novel development of this jelly-fish is thus sketched in the opening paragraph of Professor Brooks's paper: "In June, 1889, I found at Nassau, N. P., in the Bahama Islands, a few specimens of a hydromedusa belonging to the family Eucopidae (Haeckel), bearing upon each one of its four reproductive organs a number of hydroid blastostyles from which young medusae are produced by budding; a method of reproduction which has no exact parallel among the hydroids nor, as far as I am aware, anywhere else in the animal kingdom; for the reproduction, by a medusa, of blastostyles which are morphologically equivalent to hydras, is a reversion, through asexual reproduction, to a past larval stage; a phenomenon which is thoroughly anomalous and exceptional."

While Brooks regards the production of blastostyles on the medusa as a case of asexual reproduction, he finds they are not produced as simple buds. The ectoderm of the blastostyle is continuous with the ectoderm of the medusa, and arises as a bud-like outgrowth from the latter. The endoderm of the blastostyle has, however, no connection with the endoderm of the medusa, but is rooted in the mass of germ cells composing the reproductive organ of the latter. "These germ cells give rise to the endoderm of the blastostyle by a process of specialization which is very similar to what Metschnikoff has described in Cunina and has termed *sporogenesis*." The formation of blastostyles in Epenthesis is thus a composite method of reproduction, a part of the blastostyle being formed by budding, and a part by the development of rudimentary germ cells. Professor Brooks's opinion of this interesting development had best be given in his own words: "It is probable that Epenthesis is also an example of sporogenesis, and that the endodermal tube is derived from a single cell by segmentation, but this is certainly not true of the ectoderm of the blastostyle, and if we have sporogenesis at all in Epenthesis, we have it in combination with budding."

At the root of Weismann's theory of inheritance lies the supposed essential difference between somatic and germ cells.

In a little paper (35) embodying the main results of the present one, I endeavored to ascertain in terms of Weismannism the precise nature of the cells which combine to form the sponge gemmule, and arrived at the conclusion "that the gemmule cell, according to this view (Weismann's) must be regarded as a true germ cell, in which all the germ plasm remains undifferentiated, *viz.* in which none of it is transformed into ovogenetic plasm. Further, the gemmule cell pursues the parthenogenetic course of development — it keeps all its germ plasm" (p. 579). But at bottom it does not seem to me that a case of this kind, in which there is essential similarity between the products of a developing bud and a developing egg, tends to strengthen Weismann's fundamental proposition that germ cells and somatic cells are radically different.

Appendix. — I am fortunately able, some months after the completion of the present paper, to notice the remarkable memoir on the development of sponges, which M. Yves Delage has recently published.[1] In this memoir Delage describes in detail the post-larval development of Spongilla, Reniera, Aplysilla, and Esperella sordida. The essential features of development were found to be the same in all. I will briefly review his account of the Esperella development, and will then comment on certain points in which the account agrees or differs with mine.

In the ciliated larva Delage distinguishes four classes of cells each of which is destined to form a particular part of the adult body. There is a covering layer of ciliated cells, wanting at the posterior pole. Scattered about between the basal portions of these cells is a discontinuous layer of cells called by the author *epidermic.* At the posterior pole these lie at the surface, forming a nearly complete layer (in similar larva of Reniera they form, according to Delage, a complete layer). The remaining inner mass is composed of amoeboid and *intermediary* cells, the latter immobile and of a rather negative character.

The ciliated cells absorb their flagella and migrate into the interior, ultimately becoming the lining cells of the flagellated

[1] Embryogénie des Éponges. Archives de Zoologie Expérimentale et Générale. Année 1892. No. 3.

chambers. Simultaneously the epidermic elements come to the surface and fuse with one another to form a complete membrane, the definitive epidermis. The amoeboid cells become the wandering cells of the adult mesoderm, while a part of the intermediary cells form the epithelium of the canals, the rest becoming the stationary elements of the mesoderm. These conclusions differ, it will be seen, in some important respects from those presented by the author in his preliminary notes (Comptes Rendus 1890, 1891), cited *ante*, p. 317, 359.

Formation of epidermis. — In believing that the cells which cover the posterior pole form a different part of the adult body from the rest of the covering cells of the larva, I think Delage is wrong. That no such distinction exists between these two sets of the superficial cells of the larva, is made probable at the very beginning where it is seen that the young embryo is covered with a continuous layer of similar cells (columnar in Tedania, 35, p. 576, and *ante*, p. 330), which subsequently differentiate into the ciliated cells and the flattened ectoderm of the posterior pole. Delage like myself is unable to offer a satisfactory explanation of the peculiar character of this pole. He does put forth the suggestion that it is due to a rupture in the covering of ciliated cells, produced at a point of weakness by the growth of the inner mass. But the observation I have just cited upsets such an explanation.

The immigration into the interior of a part of the ciliated cells, I am prepared to believe in, some of my own observations suggesting, though by no means proving, the occurrence of such a phenomenon (*ante*, p. 299). On the other hand I am sceptical as to the existence of Delage's layer of epidermic cells, not having found any such layer in the larvae I have studied. I regret that my observations on the actual transformation of the ciliated cells of the larva into the flattened epidermis of the adult are so meagre, but such as they are they are in harmony with the views of those writers (*ante*, p. 301) who claim to have seen such a transformation, and not with the views of Delage. It may be mentioned that Delage finds the formation of the definitive epidermis to begin at the anterior pole and gradually progress towards the posterior pole

of the larva. In Esperella fibrexilis I have found the process, interpreted so differently, to take place in the opposite direction.

Marginal Membrane. — A marginal membrane, essentially like the ectodermal membrane I have described surrounding the young Esperella and Tedania, is formed in all the sponges studied by Delage. The author's account of the manner in which the membrane is formed differs, however, from mine. In Spongilla and Esperella sordida Delage describes the marginal ectodermic cells of the just attached sponge as creeping outwards in an amoeboid fashion and so forming a considerable membrane, at the edge of which the cells remain amoeboid (Pls. XIV, XV). As my figures show I have never found the marginal ectodermic cells amoeboid. On the contrary I have found the ectoderm (epidermis), as it extends out to form the membrane in question, retaining a continuous edge, which could not be the case if the individual cells of the margin threw out processes (*ante*, pp. 303 and 335, and especially Pls. XXI and XXII). The condition, at least the later condition, of the membrane in Aplysilla, as described by Delage, accords better with my observations than does his account of the membrane in Spongilla and Esperella. In Aplysilla the marginal epidermic cells at first throw out amoeboid processes, but later assume regular shapes, and arrange themselves alongside one another in such a way as to give to the membrane an even continuous edge.

Flagellated Chambers. — Several of the stages in the formation of the chambers that M. Delage has found, are quite like such as I have seen, but the whole process is construed very differently. Delage's account is as follows: "The ciliated cells after their migration into the interior are seized upon and engulfed, amoeba-fashion, by the amoeboid cells. Complete fusion takes place between the bodies of the absorbed cells and that of the amoeboid, but the nuclei of the former remain distinct and arrange themselves round the much larger nucleus of the latter. In this way are formed the multinucleate cells which have been interpreted so differently by previous observers. In Spongilla all the ciliated cells are absorbed by

the amoeboids. In Esperella and the other sponges only a portion are so absorbed, while the remainder throw out processes and unite with one another and the now multinucleate amoeboids, to form a syncitial net-work. In the development of a chamber several of the multinucleate masses approach one another and form a continuous wall round a central space. The space becomes the cavity of the chamber, round which the nuclei of the absorbed ciliated cells arrange themselves in a regular fashion, while the nucleus of the amoeboid surrounded by protoplasm escapes from the periphery of the chamber anlage, and becomes a wandering cell of the mesoderm. The ciliated cells not associated with the amoeboids, but which are merely part of the syncitium, unite in the same manner and form chambers."

The multinucleate formative cells I have described evidently correspond to Delage's multinucleate amoeboids. But while Delage agrees with Götte and myself (35, and *ante*) in regarding the smaller peripheral bodies as nuclei, he differs completely in his explanation of their origin — I regard them as derived from the central larger nucleus of the cell.

Delage's observation that chambers arise from the fusion of several multinucleate groups, corroborates the account I have given of one of the methods of chamber formation (35, and *ante*, p. 312), though, as before said, we differ greatly in our views of the ultimate origin of such groups. But on the other hand I have repeatedly observed that chambers may also be formed by formative (amoeboid) cells which group themselves in hollow spheres. Some of these cells may contain but a single nucleus, while others contain more. Observations such as this would seem to disprove Delage's thesis that the collared cells are the immigrated ciliated cells of the larva.

Canal Epithelium. — Delage, like myself, finds that the canals arise independently of one another, as irregular spaces in the inner mass, that they gradually become lined with a definite epithelium and unite with one another and with the chambers to form a connected system. Regarding the origin of the canal epithelium, however, Delage entertains widely different views from my own. His account is as follows:

"The (once) ciliated cells surrounding the irregular spaces arrange themselves so as to form a nearly continuous wall, in which, here and there, an intermediary cell is found. This is, however, not the permanent epithelium, for intermediary cells lying outside it gradually take the place of the ciliated cells, which in their turn come to lie outside the definitive epithelial wall. Such ciliated cells, which have temporarily been occupied in lining the canals, now follow the example of their brethren and unite to form flagellated chambers."

Observations such as are embodied in my Pl. XVIII, Fig. 47, and Pl. XVII, Figs. 39 and 42, seem to me to contradict the above account. The canals shown in these figures are evidently just forming, and yet their walls are made up of elements which, to judge from Delage's figures, I must conclude he would regard as amoeboid and intermediary, certainly not as immigrated ciliated cells.

Finally, the distinction which Delage makes between intermediary and amoeboid cells, is to my own mind an artificial one. His amoeboid cells evidently correspond to my formative cells, but I find no special place for his "intermediary" group, because the plump formative cells are being constantly changed into elements which Delage would class as intermediary. An instance of this is found in the development of the dermal membrane, where formative cells are gradually transformed into the slender elongated cells forming the mesoderm of this membrane (*ante*, p. 307).

NOTE. — While this paper is passing through the press, a new contribution to the subject by Otto Maas appears.[1] The author has studied a large number of marine cornacuspongiae and has worked over the development of Spongilla. His account of the metamorphosis for all these forms differs but little from his previous account of the metamorphosis of the Esperia larva. In some points Maas differs from Delage's recent conclusions. Thus Maas does not find that the ciliated cells of the larva are engulfed by the amoeboids and subsequently liberated. No such peculiar association of the two kinds of cells occurs. With this I thoroughly agree, although differing entirely with Maas in the general view of the metamorphosis. Again Maas states that in those larvae with a "bare" posterior pole, like

[1] Die Embryonal-Entwicklung und Metamorphose der Cornacuspongien. *Zoolog. Jahrbücher*, Abth. für Anat. und Ontogenie. Bd. VII., 2. H.

that of Esperella, the immigration of the ciliated cells does not take place in spots anywhere over the surface, but the inner layer actually *overgrows* the layer of ciliated cells from the posterior pole forward. In a sectional figure of a metamorphosing larva (Pl. XX, Fig. 19) he represents the layer of ciliated cells as *overlapped* by the inner mass for a considerable distance. I have cut many sections through similar stages, but have never seen a trace of such overlapping. But I can understand how a section through a larva, whose surface had been pitted in on opposite sides, could give rise to such a figure. Such pitting in of the surface may occur when the fixing fluid permits the larva to contract in the moment of death. And I am inclined to believe that the section figured was made through such a larva.

It is, however, always venturesome to suggest a new interpretation of another's figures. The suggestion is not often happy. This is certainly true of Maas's intimation that my figures of "segmenting" gemmules (Notes on the Development of some Sponges, JOURNAL OF MORPHOLOGY, 1891,) indicate that the bodies in question are not gemmules but eggs, and the "segmentation" is a real segmentation. A glance at the series of figures given in this paper, illustrating the development of the gemmule of Esperella, will show that such an interpretation is impossible.

The author's renewed study of Spongilla has led him to abandon his former views (for a statement of which see p. 360) on the development of this sponge. He finds that the flagellated chambers and exhalent canals do not arise as diverticula from a central cavity, and that the inhalent canals are not formed as ectodermic invaginations, but that the whole development agrees substantially with that of the marine cornacuspongiae, as described by himself and Delage. My statement, therefore (p. 360), that Maas "brings Spongilla in line with those forms having a rhagon" is interesting only historically.

On pp. 301 and 368 I cite Maas's observations on the flattening of the ectoderm of the Spongilla larva and its transformation into that of the adult, as a strong argument for the universality of this phenomenon in the sponges. But the author's recent paper makes these citations antiquated, since he now believes that no such transformation takes place.

CHAPEL HILL, N.C., June 15, 1894.

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EXPLANATION OF THE PLATES.

Most of the drawings were made with the camera. The lenses referred to in the description of the figures, are those of Zeiss, except when the focal distance of the objective is given in inches. The drawings have been reduced to four-sevenths original size.

Common Reference Letters used in the Figures.

af. c. Afferent canal.
an. f. c. Anlage of flagellated chamber.
can. Canal.
c. m. Solid anlage of flagellated chamber.
cna. w. / *(c. w.)* Canal wall.
c. ef. c. Central efferent canal.
cu. Cuticle.
d. mem. Dermal membrane.
deg. f. c. Degenerated flagellated chamber.
ef. c. Efferent canal.
ect. (ec.) Ectoderm.
ec. m. / *(ec. mem.)* Ectodermal membrane.
ec. un. p. p. Ectoderm of unpigmented pole.
f. c. Flagellated chamber.
for. c. g. Solid group of formative cells.
f. Furrow.
g. Gemmule.
g. f. Follicle of gemmule.
g. sh. Sheath of gemmule.
gr. c. Granular cell.
In. sp. Intercellular space.
l. Larva.
mes. b. Mesoderm band.
mes. gr. Group of mesoderm cells.
mn. m. *Multinucleate mass.*
m. p. Peripheral mesodermic process.
mes. Mesoderm (in places, mesentoderm).
os. Osculum.
os. c. Oscular cavity.
o. ov. Ovarian egg.
ov. f. Follicle of ovum.
p. w. Partition wall.
pr. g. Problematical gemmule-like bodies.
p. p. Posterior pole of larva.
pr. th. Problematical thickenings.
p. for. Perforation through peripheral mesodermic zone.
p. z. Peripheral mesodermic zone.
p. c. Pale cell.
r. Ridge.
r. g. Mature gemmule.
s. d. c. Subdermal cavity.
sh. Gemmule sheath.
sup. ef. c. Superficial efferent canal.
sp. Spicule.
sp. p. Spicular pole.
s. p. Surface pore.

EXPLANATION OF PLATE XIV.

(*Esperella fibrexilis.*)

FIG. 1. Esperella fibrexilis. × 1½.

FIG. 2. Vertical section of Esperella, showing canal system and skeleton. A.4.

FIG. 3. Spicules. *a*, oxytylote; *b*, modification of the same; *c*, after treatment with caustic potash; *d*, toxaspire; *e*, sigma; *f*, sigmaspire. D.4.

FIG. 3′. *a*, *b*, *c*, views of small shovels: *a*, somewhat from the end; *b*, face view; *c*, side view. *d*, a small *sigma* which will develop into a shovel. × 900.

FIG. 3″. Large shovels—*a*, face view; *b*, side vi· ·; *d.l.* dorsal lobe; *v.l.*, ventral lobe; *l.n.* lateral notch; *t.*, tooth. × 900.

FIG. 4. Dermal membrane. A.4.

FIG. 5. Surface of adult. 4-inch objective.

FIG. 6. Ditto, showing osculum and oscular cavity.

FIG. 7. Surface of adult. 4-inch objective.

FIG. 8. Section: gemmules in mesoderm of parent. × 800.

FIG. 8′. Different conditions of the nucleus in the gemmule cells.

EXPLANATION OF PLATE XV.

(*Esperella fibrexilis.*)

FIG. 9. Section: gemmules in mesoderm of parent. × 800.

FIG. 10. Section, showing possible origin of a gemmule from a single cell. × 800.

FIG. 11. Section: gemmules in mesoderm of parent.

FIG. 12. Section, showing situation of young and ripe gemmules in body of parent. A.4.

FIG. 13. Section: adult tissue with gemmules and cell groups. The group *a* probably derived from simple cell. × 800.

FIG. 14. Section: adult tissue with cell group and problematical gemmule-like body. × 800.

FIG. 15. Section: adult tissue with cell groups (gemmule anlages). D.4.

FIG. 16. Section, indicating the fusion of gemmules. × 800.

FIG. 17. Section: adult tissue with gemmules. Gemmule *x* has probably been formed by fusion. D.4.

FIG. 18. Section through mature gemmule. D.4.

FIG. 19. Section through gemmule of about half the full size. D.4.

FIG. 20, 20′. Sections through immature gemmules. D.4.

FIG. 20″. Ovarian egg surrounded by mesoderm of parent. F.4.

FIG. 20‴. Adult tissue with two full-sized gemmules, one of which, *g.*, is continuous with the external mesoderm, in which is an ovarian egg, *o.ov.* A.4.

EXPLANATION OF PLATE XVI.

(*Esperella fibrexilis.*)

FIG. 20^{iv}. The ovarian egg of Fig. 20‴, with neighboring tissue. D.4.

FIG. 21. Gemmule breaking up or "segmenting." Section. D.4.

FIG. 22. Section through a gemmule near the close of "segmentation." D.4.

FIG. 23. Part of a section through a gemmule, entirely broken up into separate cells. D.4.

FIG. 24. Longitudinal section through gemmule, in which the posterior pole is differentiating. C.4.

FIG. 25. Longitudinal section through gemmule larva, still in body of parent. C.4.

FIG. 26. Surface view of swimming larva shortly after birth. A.4.

FIG. 27. Surface view of swimming larva 36 hours after birth. A.4.

FIG. 28. Surface view of incompletely metamorphosed larva. A.4.

FIG. 29. Longitudinal section of larva shortly after birth. D.4.

FIG. 30. Longitudinal section of older larva. D.4.

FIG. 31. Ectoderm cells of swimming larva — maceration products.

FIG. 32. Parenchyma cells of swimming larva — maceration products.

EXPLANATION OF PLATE XVII.

(*Esperella fibrexilis.*)

FIG. 33. Ectoderm of posterior pole and adjoining parenchyma of swimming larva — maceration product.

FIG. 34. Rosette group of sigma spicules from swimming larva — seen in optical section.

FIG. 36. Longitudinal section through incompletely metamorphosed larva. D.4.

FIG. 37. Vertical section of sponge attached to surface film of water. D.4.

FIG. 38. Vertical section of recently attached sponge. A.4, tube out.

FIG. 39. Peripheral part of section through recently attached sponge. D.4.

FIGS. 40, 41. Parts of sections similar to the preceding, showing anlages of flagellated chambers. D.4, tube out.

FIG. 42. Part of section through recently attached sponge, showing formation of canals and flagellated chambers. D.4.

FIG. 43. Part of section through young sponge, the mesoderm of which consists almost entirely of fine cells. D.4.

FIG. 44. Vertical section through young sponge — subdermal cavities, canals and chambers completely differentiated. D.4.

FIG. 45. Vertical section through a young sponge. D.4.

FIG. 46. Group of multinucleate mes-entoderm cells.

FIG. 50. Section of a young sponge — for canal system. C.4.

EXPLANATION OF PLATE XVIII.

(*Esperella fibrexilis.*)

FIG. 47. Part of section through young sponge, showing formation of canal wall. $\frac{1}{12}$ Immersion 4.

FIG. 48. Section of young sponge, showing osculum. D.4.

FIG. 49. Section through peripheral part of young sponge. D.4.

FIGS. 51, 52, 53. Sections of young sponges, for canal system. C.4.

FIG. 54. Surface view of young sponge with surrounding ectodermal membrane covered with débris. A.4.

FIG. 55. Combined surface view and horizontal optical section of young sponge —note abundance of formative cells and absence of chambers. A.4.

FIG. 56. Horizontal optical section of peripheral part of young sponge—formation of peripheral mesodermic zone and canals. D.4.

FIG. 57. Horizontal optical section of older sponge in which flagellated chambers have formed. D.4.

FIG. 58. Surface view of sponge with openings into subdermal spaces. A.4.

FIG. 59. Part of periphery of preceding figure, to show pores. D.4.

EXPLANATION OF PLATE XIX.

(*Tedania Brucei.*)

FIG. 60. Portion of surface of adult, showing the meandering ridges and fu rows. × 8.

FIG. 61. Section of adult, vertical to surface — arrangement of gelatinous an spongy tissue, and skeleton. × 10.

FIG. 62. Transverse section through base of oscular papilla. × 5.

FIG. 63. Transverse section near oscular end of young sponge, 6 inches hig × 10.

FIG. 64. Vertical section of adult — arrangement of canals in gelatinous a spongy regions. A.4.

FIG. 65. Section of adult — disposition of the ultimate branches of t canals. D.4.

FIG. 66. Dermal membrane. D.4.

EXPLANATION OF PLATE XX.

(*Tedania Brucei.*)

FIG. 67. Spicules: *a*, slightly bent strongyloxea; *b*, oxea; *c*, tylote. (*a*, $\frac{20}{100}$ mm.)

FIG. 68. Surface view, showing orifices intermediate in size between pores and oscula. × 8.

FIG. 69. Section showing bodies *g*, probably young gemmules, imbedded in mesoderm. × 900.

FIG. 70. Section: adult tissue with part of mature gemmule and a young gemmule. D.4.

FIG. 71. Section: mature gemmule with surrounding tissue. A.4.

FIG. 72. Section through gemmule beginning to "segment." C.4.

FIG. 73. Part of section through gemmule, completely broken up into masses. D.4.

FIG. 74. Part of section through gemmule near the close of "segmentation." C.4.

FIG. 76. Section through embryo — columnar ectoderm entirely covers embryo. A.4.

FIG. 77. Longitudinal section through unpigmented pole of embryo — cells at this pole still columnar. D.4.

EXPLANATION OF PLATE XXI.

(*Tedania Brucei.*)

FIG. 75. Section of gemmule, in which the ectoderm layer is clearly differentiated. D.4.

FIG. 76'. Part of Fig. 76. D.4.

FIG. 78. Longitudinal section through unpigmented pole of swimming larva, shortly after birth. D.4.

FIG. 79. Surface view of larva just born. A.4.

FIG. 80. Surface view of older larva — unpigmented "plug" now conspicuous. A.4.

FIG. 81. Longitudinal section through swimming larva, a day old. C.4.

FIG. 82. Attached larva with spicular pole pulled in. A.4.

FIG. 83. Vertical section through sponge just attached. Ciliated ectoderm entirely metamorphosed, unpigmented pole still recognizable. C.4.

FIG. 84. Surface view of recently attached sponge — spicular pole lost. A.4.

FIG. 85. Surface view of sponge just attached — shows how spicular pole loses its identity — spicules are being distributed to various parts of sponge. A.4.

FIG. 86. Surface view. Sponge is solid, and margin is thrown into irregular lobes (first stage in formation of ectodermal membrane). A.4.

FIG. 87. Part of periphery of preceding figure. D.4.

FIG. 89. Part of periphery of Fig. 88. D.4.

EXPLANATION OF PLATE XXII.

(*Tedania Brucei.*)

FIG. 88. Surface view. Cavities have formed in sponge, and mesoderm no longer extends to edge of body. A.4.

FIG. 90. Surface view — ectodermal membrane well developed except in one region — central canal. A.4.

FIG. 91. Surface view — sponge of very irregular shape, which is, however, common. 1-inch objective.

FIG. 92. Vertical section of recently attached sponge — no ectodermal membrane. C.4.

FIG. 93. Vertical section of sponge with developed canal system. C.4.

FIG. 94. Peripheral region of same sponge — double nature of ectodermal membrane. D.4.

(*Tedanione foetida.*)

FIG. 95. Section through oscular papilla. × 4.

FIG. 96. Vertical section through body of sponge. × 3.

FIG. 97. Vertical section of adult — canal system and arrangement of skeleton. 3-inch objective.

FIG. 98. Section of adult — disposition of ultimate branches of canals. D.4.

FIG. 101. Modified oxea — very common form of spicule.

EXPLANATION OF PLATE XXIII.

(*Tedanione foetida.*)

FIG. 99. Section of adult — histology of gelatinous tissue. D.4.
FIG. 100. Dermal membrane. A.4.
FIG. 102. Section showing very young egg-cell. D.4.
FIG. 103. Section showing immature egg surrounded by numerous mesoderm cells. D.4.
FIG. 104. Section of adult tissue with egg of full size — only one nucleolus. D.4.
FIG. 105. Section of mature egg — extruded nucleolus. D.4.
FIGS. 106–110. Sections of segmenting eggs. D.4.
FIG. 111. Section of planula — coarse yolk has disappeared. D.4.
FIG. 112. Longitudinal section of swimming larva. D.4.
FIG. 113. Section showing a larva attached to canal-wall of mother. D.4.

(*Hircinia acuta.*)

FIG. 114. Section of adult tissue with young ovum. D.4.
FIGS. 115, 115′. Sections showing young ova and follicles. D.4.

EXPLANATION OF PLATE XXIV.

(*Hircinia acuta.*)

FIG. 116. Section of immature ovum, surrounded by numerous mesoderm cells. D.4.

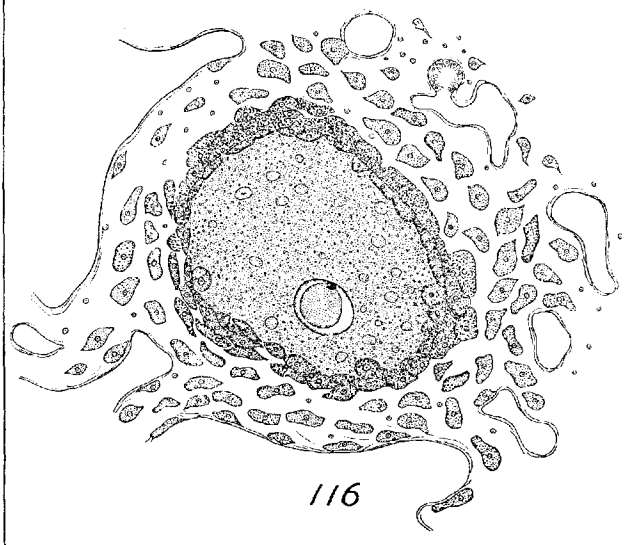
FIG. 117. Section showing mature (as to size) ovum in situ. A.4.

FIGS. 118, 118′. Two sections of mature ova showing stages in the extrusion of the second nucleolus. D.4.

FIG. 119. Section through part of ripe egg with surrounding follicle. D.4.

FIG. 120. Section through segmenting egg. C.4.

FIGS. 121, 122. Sections of morulas. C.4.



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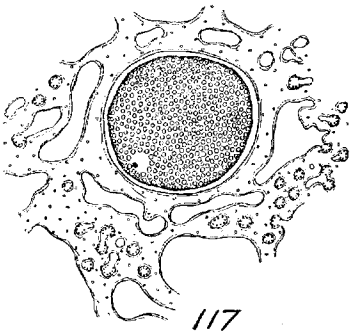

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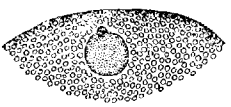

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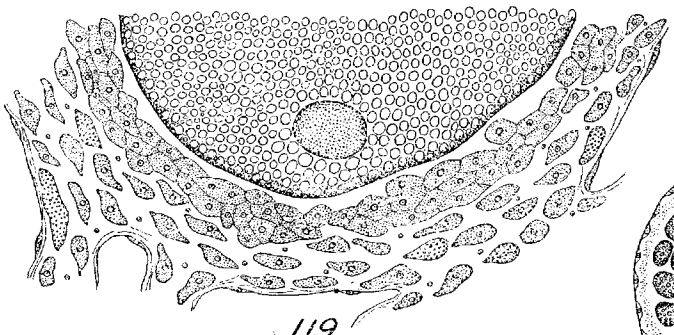
118'



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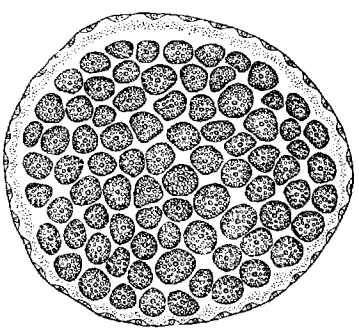

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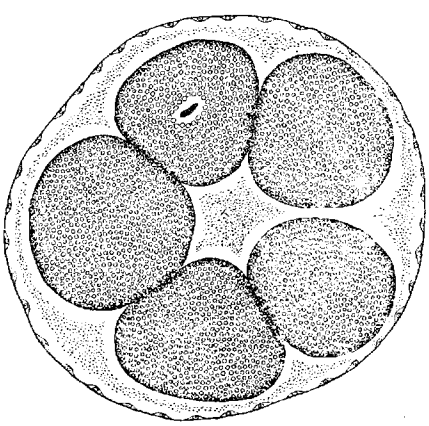

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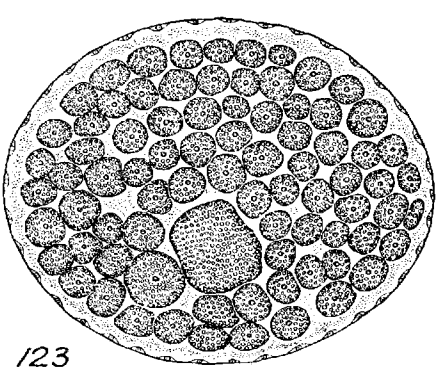

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HVW del

EXPLANATION OF PLATE XXV.

(Fig. 5 original — the other figures borrowed to illustrate the section "Remarks on the Morphology of Sponges.")

FIG. 1. Anamixilla torresi. Trans. section, from Polejaeff, Challenger Report on Calcarea, Pl. IV, Fig. 2a.

FIG. 2. Leucilla connexiva. Trans. section, from Polejaeff, Pl. VI, Fig. 1a.

FIG. 3. Leucilla uter. Trans. section, from Polejaeff, Pl. VI, Fig. 2a.

FIG. 4. Leuconia multiformis. Trans. section, from Polejaeff, Pl. VI, Fig. 3a.

FIG. 5. Diagrammatic section of an hypothetical silicious sponge.

FIG. 6. Leucandra caminus. From Vosmaer's Spongien, Taf. I, Fig. 18, after Haeckel.

FIG. 7. Plakina monolopha. Vertical section, from Schulze. Zeit. für Wiss. Zool., 34. Bd., Taf. XX, Fig. 4.

FIG. 8. Young Plakina monolopha, just attached. Vertical section, from Schulze, l. c.

FIG. 9. Plakina dilopha. Vertical section, from Schulze, l. c. Taf. XX, Fig. 11.

FIG. 10. Plakina trilopha. Vertical section, from Schulze, l. c. Taf. XXI, Fig. 12.

FIG. 11. Rhagon stage of Oscarella lobularis, after Heider, from Korschelt und Heider's Lehrbuch, p. 6.

REFERENCE LETTERS TO THIS PLATE.

af. c.	Afferent canal.	*mes.*	Mesoderm.
c. p.	Chamber pore.	*os.*	Osculum.
ec.	Ectoderm.	*Par. cav.*	Paragastric cavity.
en.	Entoderm.	*r. w.*	"Ringwall."
ef. c.	Efferent canal.	*R. t.*	Radial tube.
f. c.	Flagellated chamber.	*s. p.*	Surface pore.
In. can.	Intercanal.		