zufassen. In diesem Sinne das Beispiel betrachtend, wie ich selbst es am betreffenden Ort gethan habe, wird Kölliker mir gewiß beistimmen.

Fasse ich zusammen, so glaube ich im Vorausgehenden gezeigt zu haben, daß sowohl Hæckel als ich beanspruchen können, in der Erklärung der Vererbung am frühesten den richtigen Weg eingeschlagen zu haben. Ich gedenke auf diesen Punct an anderer Stelle ausführlicher zurückzukommen. Was aber die Verwendung desselben Gesichtspunctes zur Aufstellung eines histologischen Systems betrifft, so erlaube ich mir auf eine frühere Abhandlung zu verweisen[1].

5. The Germ-layers of Clepsine.

By C. O. Whitman, Cambridge, Mass.

eingeg. 15. Januar 1886.

Since the publication of my memoir on »The Embryology of Clepsine« (Quart. Journ. Micr. Sc. 1878), several communications have been made by different observers on the same subject. First of all came Hoffmann's second memoir (Untersuch. ü. d. Bau u. d. Entwicklungsgesch. d. Hirudineen, Haarlem, 1880), in which the author repeatedly declares that he is unable to distinguish germ-layers in the Hirudinea. Then two preliminary notices have been published by Joseph Nusbaum (Zool. Anz. Nos. 181 u. 191), the first dealing more particularly with the germ-layers, the origin of the nerve-system, etc., and the second with the development of the sexual organs. Finally, R. S. Bergh has published extended observations on the Metamorphoses of *Aulostomum* and *Nephelis* (Zool. Anz. No. 160; Semper's Arbeiten, VII[1]; Zeitschr. f. w. Zool., 41. Bd.), in which he has dealt with *Clepsine* by way of comparison; and I have myself given briefly some new facts on the development of *Clepsine* (American Naturalist, p. 1134, Nov. 1885). I have, further, replied to a criticsim of Balfour's on my interpretation of the elements composing the germ-bands (Quart. Journ. Micr. Sc., XXIII, p. 391—392, July, 1883), and have compared the formation of the embryo with that of vertebrates.

In the present communication, I propose to give the more important results of a study on the germ-layers of *Clepsine parasitica* (?) Say. The methods employed have been given in detail in the American Naturalist, Nov. 1885, p. 1134—1135. The use of osmic acid fol-

[1] Die Entwicklung der Gewebe des Säugethierkörpers und die histologischen Systeme. in: Sitzgsber. d. naturforsch. Ges. zu Leipzig 1883.

[1] »Undersogelser over Metamorphosen hos *Aulostoma gulo*«. Kjobenhavn. 1885.

lowed by Merkel's chrom-platinum solution, has enabled me to trace the history of the entoderm, and the precise origin of the nerve-chord, nephridia, salivary glands, larval glands etc.

Each germ-band, as was made clear in my first paper, and as had already been stated by Metschnikoff[2], is composed of three distinct layers, namely: (1) An epidermal layer, (2) a layer of four longitudinal rows of cells, and (3) a deeper layer, next to the yolk, composed of larger cells. The precise origin of all these layers was very carefully worked out in my paper, and none of my successors has called in question the accuracy of those results, so far as this point is concerned. It may therefore be considered a settled fact, that the second and third layers of each germ-band are derived from five cells. But the question as to the interpretation of these two layers is one on which opinions are greatly at variance. Metschnikoff held the second layer to be the basis of the nerve-chord, and the third layer to be mesoblastic. Quite independently of this author's paper, which I had quite overlooked, I came to the same conclusion, and accordingly named the four cells from which the second layer is derived »neuroblasts«, and the single large cell from which the third layer arises, »mesoblast«. Hoffmann, in his second paper, attaches the same meaning to the second layer, while maintaining that the third layer gives rise to both the mesoderm and entoderm. Bergh, while conceding the fact that the nerve-chain has its origin in a part of the second layer, remarks with rather more assurance than the case appears to call for, that my statement on this point is a pure assertion. Nusbaum follows Balfour in regarding both the second and third layers as mesoderm, and derives the nerve-chain from a median thickening of the epidermal layer, similar to what has been described by Salensky in *Branchiobdella* (Biol. Centralblatt, 2. Bd., No. 7).

As to the origin of the entoderm, we learn nothing definite from Nusbaum. This author regards the eight »neuroblasts« as entoderm cells, and their products as mesoblastic (sexual cells). Hoffmann denies the cell nature of my »entoplasts«, and holds that the entoderm arises from the germ-bands. Bergh also rejects my view of the origin of the permanent entoderm, and suspects that it has an early origin from cells at the anterior end of the embryo, similarly as in *Nephelis*.

Nusbaum's statements on the origin of the nerve-chord are, as I am able to show, utterly at variance with fact; and, in this connection, I would also say that this author appears to have a very confused idea

[2] »Beiträge zur Entwicklungsgeschichte einiger niederen Thiere.« Vorläufige Mittheilung im Bull. de l'Acad. imp. des Sci. de St. Pétersbourg, t. XV, 1871; and in Mélanges Biologiques, t. VII. p. 671—673.

in respect to the composition of the germ-bands, and especially in respect to the precise origin of the cells. Strangely enough, he has left the two large »mesoblasts« entirely out of consideration, and derives the sexual cells (my »segment-cells«) from the »neuroblasts«! At the hind end of the embryo, are found, according to his account, four rows of these sexual cells on the ventral side of each germ-band; farther forward only two rows are present; and at the anterior end, only one row. This remarkable reduction of four rows to one is passed over without explanation or comment. My recent study confirms my former statements, in regard to the origin of these cells from the »mesoblasts«. The »neuroblasts« do give rise to four rows of cells in each band, but the number of these cells seen in transverse section does not diminish, but increases from behind forward. The origin of the sexual cells may be seen to best advantage in a series of sagittal sections.

The Nerve-chord and Nephridia.

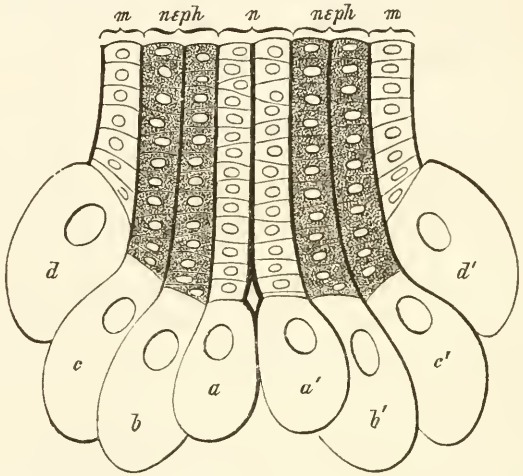
In my first paper, I gave an accurate account of the composition of the germ-bands, and traced the origin of the cells to ten primitive blastomeres, thus furnishing the needed foundation for a more detailed study. I satisfied myself that the nerve-chain was formed from the products of the so-called »neuroblasts«, but I was mistaken in supposing that all of these products entered into its composition.

More favorable material and better methods have shown that, of the eight rows of these cells (four in each band) only the two median ones give rise to the nerve-chain (*n*). The lateral row (*m*) of each band probably gives rise to muscular elements, while the two rows (*neph.*) lying between the median and lateral rows furnish the basis of the nephridial organs. The two median rows of nerve-cells are faintly browned with osmic acid, while the four rows of nephridial cells are more granular and deeply browned, forming thus a sharp contrast in color and general appearance. The two lateral rows are slightly stained, differing but little from the nerve-cells. The development of the ganglionic chain progresses from the head backwards, so that in surface views it is not difficult to trace the two simple rows of nerve-cells forward into the fully outlined ganglia. The same has been done on sections, leaving no doubt as to the origin of the nerve-system of the trunk. I am not able to say whether the two rows of nerve-cells extend into the cephalic lobe; but I am certain that the nerve-collar, including the supra-œsophageal ganglia, is formed from cells that lie beneath the epidermis, and not from a thickening of the epidermis itself.

The epidermis overlying the nerve-cells destined to form the four subœsophageal ganglia, thickens up at an early date and eventually

becomes from two to three or more cells deep. This thickened portion of the epidermis has nothing whatever to do with the formation of any part of the nervous system. The deeper cells of this thickening form provisional gland-cells, which serve to attach the embryo, after its escape from the egg-membrane, to the ventral side of the parent. In this manner the young are carried about until the posterior sucker is developed sufficiently to serve as an organ of attachment. These gland-cells are stained dark brown, and are thus very easily distinguished from the lighter-colored nerve-cells lying beneath them.

In *Aulostomum* and *Nephelis*, according to Bergh, the primitive epidermal layer is lost, and the second layer of the germ-bands gives



A diagrammatic surface view of the second layer at the posterior end of the nearly completed germ-bands.

n = nerve-chain.
neph = nephridia.
m = mesoblast (?)
a b c d a' b' c' d' = the terminal proliferating blastomeres.

origin to the nerve-chord and the definitive epidermis. Ten proliferating cells are found at the posterior ends of the germ-bands, as in *Clepsine*; and in *Nephelis* five distinct lines of cells are traced to the five proliferating cells of each band. These lines of cells are present in *Aulostomum*, but are not so regularly arranged as in *Clepsine* and *Nephelis*. It is interesting to note that the two median rows in *Aulostomum* are perfectly distinct from the other rows, a fact which makes it all the more probable that they have the same destination as in *Clepsine*. It would seem from one of Bergh's figures (Semper's Arbeiten, VII, Pl. XII, Fig. 5) that the larval nephridia may arise from the same

rows of cells as the permanent nephridia in *Clepsine*. Nusbaum's statement that the nephridia arise in connection with the somatopleura, is in plain contradiction with my observations.

The permanent Entoderm.

The epithelium of the whole alimentary tract, excluding the stomodæum (pharynx) and proctodæum, which are derived from the epidermal layer, arises from free nuclei belonging to the three large blastomeres (*a*, *b* and *c* in my figures). The cells which form the œsophagus are the first in order of development, making their appearance just beneath the stomodæal thickening, in the very earliest stage of the germ-bands. From the mass of cells formed at this point arise not only the œsophageal epithelium, but also the salivary glands. The method employed gives preparations in which all the embryonic tissues of the head and anterior portion of the trunk (epidermis, larval gland, salivary glands, nerve-cells, muscle-cells, and œsophageal epithelium) are distinguishable.

The cells destined to form the epithelial lining of the stomach arise later than those of the œsophagus. They appear first as distinct cells, on the ventral side, at the anterior end of the future stomach, at about the time of hatching. Their development is progressive from this point backward and upward towards the dorsal side. In an embryo just hatched, I can trace these cells along nearly the anterior half of the median ventral line, and farther back I find free nuclei in the surface of the yolk. In the median dorsal line, I find no fully formed entoderm cells (except salivary gland-cells), but do find free nuclei in the anterior half.

Sense-organs.

The sense-organs of the lip arise as bulb-like thickenings of the epidermis. At the time of hatching, long before the eyes and segmental sense-organs appear, two pairs of these sense-bulbs are found, symmetrically placed on the surface that is to form the margin of the lip. The symmetrical arrangement in pairs, the second pair being a little behind the first and farther apart, suggests that these organs were primarily strictly segmental.

Larval Gland-cells.

The origin of these provisional gland-cells has been given above. It appears to me not improbable that the epidermal thickening, from which these glands arise, has misled Nusbaum into his view of the origin of the nervous system. This author has fallen into the error of supposing that the function of these gland-cells is to bind the embryos to each other, while they attach themselves to the ventral side of the

parent leech by their anterior suckers. It is quite true that the newly hatched young, if prevented from attaching themselves to the parent, become fixed to each other in pairs by means of these glands; but their real purpose is to fix the young to the mother at a time when neither sucker is sufficiently well developed to serve this end. Later they attach themselves by the posterior sucker.

Mus. Comp. Zool., Cambridge, Mass., Jan. 1. 1886.

III. Mittheilungen aus Museen, Instituten etc.

1. Linnean Society of London.

4th March 1886. — A paper was read Description of *Strongylus Arnfieldi*, with observations on *Strongylus tetracanthus* by Prof. Spencer J. Cobbold. Of Arnfield's Strongyle he drew attention to the morphology of the hood and its rays, to the position of the vulva, and to the structure of the embryo. He afterwards contrasted these peculiarities with those of allied forms. Regarding his observations on the Four-spined Strongyle, the following are his conclusions: — (1) The eggs are expelled from their parent in a state of fine yolk-cleavage. (2) The embryos are formed after egg expulsion, and in a few days escape from their envelopes, undergoing a primary change of skin in moist earth during warm weather. (3) As rhabditiform nematoids they enjoy a more or les prolonged existence, probably living many weeks in this state. (4) In all likelihood an intermediary host is unnecessary. (5) The rhabditiform larvae are passively transferred to the intestinal canal, thence they enter the walls of the cœcum and colon, encyst themselves, and according to Leuckart, undergo another change of skin. (6) Their presence in the intestinal walls is associated with pathological conditions which frequently prove fatal to the bearer, sometimes creating severe epizooty. (7) Ordinarily the young worms perforate their cysts and migrate to the lumen of the bowel, where they already afford external indications of sex (Trichonema stage of growth). (8) They next form Cocoons by the agglutination of Vegetable debris within the gut and undergo a third ecdysis, attended with intestinal metamorphosis. (9) The formation of the internal sexual organs and the completion of the definitive form is acquired within the colon of the host. — J. Murie.

Bemerkung.

Um Irrthümer bei Herstellung von Sonderabdrücken zu vermeiden, werden die Herren Mitarbeiter ersucht, die Zahl der etwa gewünschten Separate mir gleich auf dem Manuscript selbst anzugeben und auch thunlichst gleichzeitig der Verlagshandlung von Wilhelm Engelmann mitzutheilen. Nachträgliche, d. h. nach Ausgabe einer Nummer, eingehende Bestellungen können nicht mehr berücksichtigt werden.

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