

On the EMBRYOLOGY of LIMULUS POLYPHEMUS. By A. S. PACKARD, JR., M.D.

(Read before the American Association for the Advancement of Science, August, 1870.¹)

THE eggs on which the following observations were made were kindly sent me from New Jersey, by Rev. Samuel Lockwood, who has given an account of the mode of spawning, and other habits, in the 'American Naturalist.' They were laid on the 16th of May, but it was not until June 3rd that I was able to study them. The eggs measure .07 of an inch in diameter, and are green. In the ovary they are of various hues of pink and green just previous to being laid, the smaller ones being, as usual, white. The yolk is dense, homogeneous, and the yolk granules, or cells, are very small, and only in certain specimens, owing to the thickness and opacity of the egg-shell, could they be detected.

Not only in the eggs already laid, but in unfertilised ones taken from the ovary the yolk had shrunken slightly, leaving a clear space between it and the shell. Only one or two eggs were observed in process of segmentation. In one the yolk was subdivided into three masses of unequal size. In another the process of subdivision had become nearly completed.

In the next stage observed, the first indications of the embryo consisted of three minute, flattened, rounded tubercles, the two anterior placed side by side, with the third immediately behind them. The pair of tubercles probably represent the first pair of limbs, and the third, single tubercle the abdomen. Seen in outline the whole embryo is raised above the surface of the yolk, being quite distinct from it, and of a paler hue. In more advanced eggs three pairs of rudimentary limbs were observed, the most anterior pair representing the first pair of limbs (false mandibles of Savigny), being much smaller than the others. The mouth opening is situated just behind them. In a succeeding stage the embryo forms an oval area, surrounded by a paler coloured areola, which is raised into a slight ridge. This areola is destined to be the edge of the body, or line between the ventral and dorsal sides of the animal. There are six pairs of appendages, forming elongated tubercles, increasing in size from the head back-

¹ Dr. Packard complains that he has been misrepresented in our paper on the subject of his present paper, and we gladly avail ourselves of his permission to publish the paper at length.—Eds.

wards; the mouth is situated between the anterior pair. The whole embryo covers but about a third of that portion of the yolk in sight. At this time the inner egg membrane (blastoderm-skin?) was first detected.

The outer membrane, or chorion, is structureless; when ruptured the torn edges show that it is composed of five or six layers of a structureless membrane, varying in thickness. The inner egg membrane is free from the chorion, though it is in contact with it. Seen in profile it consists of minute cells which project out, so that the surface appears to be finely granulated. But on a vertical view it is composed of irregularly hexagonal cells, sometimes 5-sided, and rarely 4-sided, hardly two cells being alike. The walls of the cells appear double, and are either strongly waved, or have from three to five long slender projections, with the ends sometimes knobbed, directed inwards. These cells are either packed closely together, or separated by quite a wide interspace.

In a subsequent stage the oval body of the embryo has increased in size. The segments of the cephalothorax are indicated, and the legs have grown in length, and are doubled on themselves. But the most important change is in the small size of the rudiments of the mandibles, compared with the remaining five pairs of limbs; and the origin of two pairs of gills, forming pale oblique bands between the sixth pair of legs and the end of the abdomen, which forms a narrow semicircular area.

A later stage is signalised by the more highly developed dorsal portion of the embryo, and the increase in size of the abdomen and the appearance of nine distinct abdominal segments. The segments of the cephalothorax are now very clearly defined, as also the division between the cephalothorax and abdomen, the latter being now nearly as broad as the cephalothorax, the sides of which are not spread out as in a later stage. At this stage the egg-shell has burst, and the "amnion" increased in size several times exceeding its original bulk, and has admitted a corresponding amount of sea water, in which the embryo revolves. At a little later period the embryo throws off an embryonal skin, the thin pellicle floating about in the egg.

Still later in the life of the embryo the claws are developed, an additional rudimentary gill appears, and the abdomen grows broader and larger, with the segments more distinct; the heart also appears, being a pale streak along the middle of the back extending from the front edge of the cephalothorax to the base of the abdomen.

Just before hatching the cephalothorax spreads out, the

whole animal becomes broad and flat, the abdomen being a little more than half as wide as the cephalothorax. The two eyes and the pair of ocelli on the front edge of the cephalothorax are distinct; the appendages to the gills appear on the two anterior pairs; the legs have increased in length, though only a rudimentary spine has appeared on the coxal joint, corresponding to the numerous teeth in after life. The trilobitic appearance of the embryo is most remarkable. It also now closely resembles the Xiphosurian genus *Bellinurus*. The cardiac or median region is convex and prominent. The lateral regions are more distinctly marked on the abdomen than on the cephalothorax. The six segments of the cephalothorax can, with care, be distinguished, but the nine abdominal segments are most clearly demarked, and in fact the whole embryo bears a very near resemblance to certain genera of Trilobites, as *Trinuclaus*, *Asaphus* and others.

In about six weeks from the time the eggs are laid the embryo hatches. It differs chiefly from the previous stage in the abdomen being much larger, scarcely less in size than the cephalothorax; in the obliteration of the segments, except where they are faintly indicated on the cardiac region of the abdomen; and the gills are much larger than before. The abdominal spine is very rudimentary, forming a lobe varying in length, but scarcely projecting beyond the edge of the abdomen. It forms the ninth segment. The young swim briskly up and down the jar, skimming about on their backs, by flapping their gills, not bending their bodies. In a succeeding moult, which occurs between three and four weeks after hatching, the abdomen becomes smaller in proportion to the cephalothorax, and the abdominal spine is prominent, being ensiform, and about three times as long as broad. At this and also in the second or succeeding moult, which occurs about four weeks after the first moult, the young *Limulus* doubles in size.

Conclusions.—The eggs are laid in great numbers loose in the sand, the male fertilising them after they are dropped. This is an exception to the usual mode of oviposition in Crustacea; *Squilla* and a species of *Gecarcinus* being the only exception known to me to the law that the Crustacea bear their eggs about with them. Besides the structureless, dense, irregularly laminated chorion, there is an inner egg membrane composed of rudely hexagonal cells; this membrane increases in size with the growth of the embryo, the chorion splitting and being thrown off during the latter part of embryonic life. Unlike the Crustacea generally the primitive band is confined to a minute area, and rests on top of the

yolk, as in the spiders and scorpions, and certain Crustacea, *i.e.*, *Eriphia spinifrons*, *Astacus fluviatilis*, *Palæmon adspersus*, and *Crangon maculosus*, in which there is no metamorphosis.

The embryo is a Nauplius; it sheds a Nauplius skin about the middle of embryonic life.

This Nauplius skin corresponds in some respects to the "larval skin" of German embryologists.

The recently hatched young of *Limulus* can scarcely be considered a Nauplius, like the larvæ of the Phyllopoda, Apus and Branchipus, but is to be compared with those of the trilobites, as described and figured by Barrande, which are in *Trinucleus* and *Agnostus* born with only the head and pygidium, the thoracic segments being added during after life. The circular larva of *Sao hirsuta*, which has no thorax, or at least a very rudimentary thoracic region, and no pygidium, approaches nearer to the Nauplius form of the Phyllopods, though we would contend that it is not a Nauplius.

The larva passes through a slightly marked metamorphosis. It differs from the adult simply in possessing a less number of abdominal feet (gills), and in having only a very rudimentary spine. Previous to hatching it strikingly resembles *Trinucleus* and other trilobites, suggesting that the two groups should, on embryonic and structural grounds, be included in the same order, especially now that Mr. E. Billings¹ has demonstrated that *Asaphus* possessed eight pairs of five-jointed legs of uniform size. The trilobite character of the body, as shown in the prominent cardiac and lateral regions of the body, and the well-marked abdominal segments of the embryo, the broad sternal groove, and the position and character of the eyes and ocelli, confirm this view. The organization and the habits of *Limulus* throw much light on the probable anatomy and habits of the trilobites. The correspondence in the cardiac region of the two groups shows that their heart and circulation was similar. The position of the eyes shows that the trilobites probably had long and slender optic nerves, and indicates a general similarity in the nervous system. The genital organs of the trilobites were probably very similar to those of *Limulus*, as they could not have united sexually, and the eggs were probably laid in the sand or mud, and impregnated by the sperm cells of the male, floating free in the water.

¹ "Proceedings of the Geological Society of London," reported in 'Nature,' June 2nd, 1870. In this communication Mr. E. Billings announces the important discovery of a specimen of *Asaphus platycephalus*, showing that the animal possessed eight pairs of five-jointed feet, widely separated at their insertions by a broad sternal groove.

The muscular system of the trilobites must have been highly organized as in *Limulus*, as like the latter they probably lived by burrowing in the mud and sand, using the shovel-like expanse of the cephalic shield in digging in the shallow palæozoic waters after worms and stationary soft-bodied invertebrates, so that we may be warranted in supposing that the alimentary canal was constructed on the type of that of *Limulus*, with its large, powerful gizzard and immense liver.

NOTES on APPENDICULARIA and the LARVAL CONDITION of an ACANTHOCEPHALOID SCOLECID from the COAST of PORTUGAL. By W. SAVILLE KENT, F.Z.S., F.R.M.S., &c., of the Geological Department, British Museum.

The figures accompanying this communication illustrate two floating forms encountered last summer during my dredging trip with Mr. Marshall Hall and Mr. Edw. Fielding to the coast of Spain and Portugal, in the former gentleman's commodious yacht the "Norna."

The two were observed on one occasion only, early one calm morning in June, when the surface of the sea was like a millpond. Both occurred in large patches, at the surface of the water, and as they floated past the vessel received from our crew the very comprehensive term of "spawn."

The microscope, however, or the unassisted eye even at close quarters, speedily revealed to us that they were bodies of a far higher type, in the literal sense of the term, than had been accredited to them, and at the same time, that the two were essentially distinct from one another, in both histological structure and general form.

A perfect individual of the first of these (Pl. XIV, figs. 1 and 2) might be described as a minute hyaline body, roughly resembling, in configuration, a transparent tadpole, having an inflated anterior portion divided into two separate chambers, with a dependent tail-like appendage attached to it, whose rapid vibrations served to propel the organism through the water. The two chambers of the body proper differed considerably in size, as also in the nature of their respective contents and colorisation, the anterior and larger one being completely filled with pale amber-coloured spherical bodies, varying in number from 80 to upwards of 100, which must undoubtedly be identified as ova, while the posterior, and by