

with reports of a somewhat similar nature by other investigators, carry the definite implication that the bacteriophage is generated *de novo* by the bacteria themselves, under the "liberating" influence of a specific environment. We already have evidence that this "liberating" influence is closely associated with the phenomenon of microbic dissociation. What the specific environment may involve, remains for further study.

Finally, and most important, it must be added that the strains of *B. coli* and *B. dysenteriae* Shiga employed for the preceding experiments were "normal", sensitive, type S cultures, maintained uninterruptedly for many years in the laboratory collection. They are non-lysogenic, in the common meaning of this term, and are not open to the imputation of "contamination" with the bacteriophage in the d'Herelle sense:

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<sup>1</sup> Hoder, F., and Suzuki, K., *Centralbl. f. Bakt.*, Abt. I, Orig., 1926, xcviii, 433.

### 3683

#### Advanced Development of Some Echinoid Plutei.

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Employing a modification of the method of Allen and Nelson<sup>1</sup> for rearing echinoderm larvae, I have been able to observe the development of three Pacific coast forms. These were the sea urchins, *Strongylocentrotus purpuratus* and *S. franciscanus*, which developed to advanced pluteus stage, and the sand urchin, *Dendraster eccentricus*, which readily underwent metamorphosis.

About 20 early plutei were put into a sterilized finger bowl containing 75 cc. of sterilized San Miquel sea water, to which a drop of the culture of *Nitschia closterium* was added. The dish was then covered with a glass plate and set in a shaded place, where the temperature during the entire summer did not vary beyond the limits 15° to 17° C. Every 5 or 6 days the larvae had to be transferred to a fresh dish of modified sea water, in order to prevent their being overwhelmed with the growth of diatoms.

After 3 to 4 weeks the plutei of *S. purpuratus* and *S. franciscanus* developed a third pair of arms, into each of which a supporting skeletal spicule grew. All the arms became very long, and then deteriorated, without the animals giving indication of any steps toward metamorphosis. These larvae always rested on the bottom.

With the larvae of *Dendraster eccentricus* development was much more rapid and vigorous. Segmentation was approximately twice as rapid as in the sea urchins. In 48 hours the 4 armed plutei were formed in 10 to 12 days, and in 10 days the third pair of arms appeared. This last seemed to be a critical step because if the third pair of arms did not appear within two weeks, the larva remained juvenile. Almost immediately after the appearance of the third pair of arms, the fourth pair began to develop from the oral plate, and were complete within the following week. Subsequently, growth in size and development of the echinus element were striking phenomena. There appeared from this time on ever increasing differences in rate of development between individuals in the same culture. Until metamorphosis, the 8-armed plutei swam freely near the top or fed on the bottom. Differentiation in the sense of shortening the arms did not normally occur. The pluteus exhibited no orientation to contact, but the newly metamorphosed animal showed the stereotropic reactions of the tube feet characteristic of the adult. The earliest metamorphosis occurred 35 days after fertilization. Dimensions of metamorphosed sand urchin were: diameter, .367 mm.; depth, .334 mm. Length of pluteus before metamorphosis, .785 mm. Shortest time required for metamorphosis, 35 days.

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<sup>1</sup> Allen, E. J., and Nelson, E. W., *J. Marine Biol. Assn.*, 1910, viii, 421.

### 3684

#### Nature of Hyaline Membrane in Fertilized Egg of Sea Urchin.

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The inner or hyaline membrane of the fertilized egg of the sea urchin, *Strongylocentrotus purpuratus*, which serves to hold the blastomeres together, is rendered permanent by keeping the developing eggs in a solution composed of 35 cc. sea water + 15 cc. 3/8 M Ca Cl<sub>2</sub>. Such cultures show the following facts: (1) The outer or fertilization membrane around the inert blastula disappears normally, *i. e.*, within 24 hrs. It is therefore not broken by the hatching blastula, as is generally supposed. (2) So long as the larvae remain within the Ca-membrane they retain the form of blastulae, but segmentation goes on, and eventually the blastocoel is filled with mesenchyme cells; invagination does not occur. (3) After 3 to 5 days the