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ON THE RHYTHMIC FORMATION OF CLEAR CYTOPLASMIC
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OF THE SEA URCHIN EGG

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The writer^{1), 2)} has already reported on some rhythmic changes in the cytoplasm which occur in accordance with the mitotic cycles of fertilized and artificially activated eggs. In this work, another rhythmic variation of the cytoplasm is studied, treating eggs with hypertonic sea water.

Eggs of *Pseudocentrotus depressus* and *Hemicentrotus pulcherrimus* are exposed to hypertonic sea water (2.5 N NaCl 8 cc+sea water 50 cc) for 10 minutes at various stages after fertilization or activation. When the hypertonic treatment is applied to fertilized eggs at a stage between the streak stage and prometaphase, clear spots, about 10 μ in diameter, appear in the cytoplasm near the egg periphery within 20 minutes after they have been returned to normal sea water. These spots gradually move towards the early mitotic apparatus, increasing in size and decreasing in number. Finally, they disappear at about 10 minutes later, and, in turn, the amphiaster fully develops and cleavage follows.

On the other hand, in artificially activated eggs, the spot formation is induced only when the eggs are exposed to the hypertonic sea water at a stage between breakdown of the nuclear membrane and formation of the monaster. Later fate of these spots is quite similar to those of fertilized eggs. The spot formation has been observed until the third mitotic cycle both in fertilized and in activated eggs.

If a similar hypertonic treatment is applied to eggs stratified by centrifuging, the spots tend to appear accumulated in a definite zone. Even in activated centrifugal egg-halves, which lack nuclei, the spot formation can be induced almost at the same time after activation as in activated uncentrifuged eggs.

According to Kawamura and Dan³⁾, the cyclic appearance of -SH rich spots is found in ehterized eggs. To compare these spots with those presented by the writer, some experiments have been performed. Fertilized or artificially activated

1) KUNO, M. 1954. Zool. Mag. (Tokyo), 63 : 423. (In Japanese).

2) KOJIMA, M. K. 1958. *Ibid.*, 67 : 30. (In Japanese).

3) KAWAMURA and DAN, J. C. Biophys. Biochem. Cytol. (In press).

eggs are exposed to sea water solutions of ether, urethane and several other reagents, respectively. Cyclic spot formation is observed in a solution of ether (0.6%) or urethane (0.06M-0.08M), and it is found that the spots have a nature quite similar to those obtained by the hypertonic treatment, although the former behave rhythmically in the experimental solution. Therefore, there is a possibility that spots found by Kawamura and Dan and the present ones are of the same origin.

From the described results, it may be concluded: (1) A period in which the clear cytoplasmic spots can be easily produced by hypertonic treatment, appears rhythmically prior to the aster formation both in fertilized and in activated eggs; (2) The spot formation takes place even when there is no influence of the nucleus; (3) These spots might be formed by aggregation of -SH rich substance which might, according to Kawamura and Dan, contribute to the aster formation.