

### Delay of Normal Development of Sea Urchin Embryos by Cytosine Arabinoside

When demembranated frog gastrulae (*Rana pipiens*) are cultured in 0.05% cytosine arabinoside for 24 h, there is a pronounced inhibition of DNA synthesis with little effect upon the synthesis of RNA and protein during this period of development<sup>1</sup>. The rate of development during gastrulation is retarded by this treatment, but after removal from the solution of cytosine arabinoside, development proceeds in a normal manner. Presumably this delay is due to the inhibition of DNA synthesis and cell division during the period of gastrulation. A developmental delay also was obtained when isolated neural plates were treated with 0.05% cytosine arabinoside for 24 h, but after removal from the inhibitor, development often was abnormal.

Since most chemical compounds that retard the rate of development also cause abnormalities, it was of interest to learn if cytosine arabinoside would cause a delay of normal development of sea-urchin embryos. Eggs of 6 *Sphaerochinus granularis* females were fertilized and, after 2½ h of development, cytosine arabinoside at concentrations of 0.005, 0.025, 0.05 and 0.1% was added to separate batches of these eggs cultured in sea water with stirring at 18°C. After 4 and 8 h of exposure the developing embryos in each concentration of cytosine arabinoside were washed with sea water to remove them from the cytosine arabinoside. They were allowed to continue development and were examined microscopically each day for 5 days. After 19 h of development, both the control eggs and the eggs treated with the various concentrations of cytosine arabinoside had hatched and were starting to gastrulate. After 44 h of development, the controls and the eggs treated with the various concentrations of cytosine arabinoside for 4 h, as well as those treated with 0.005% cytosine arabinoside for 8 h, were late gastrulae. Gastrulation was retarded in eggs treated with 0.025, 0.05 and 0.1% cytosine arabinoside for 8 h. Most of the embryos that had been exposed to 0.1% cytosine arabinoside for 8 h had not yet gastrulated. By the fourth day the controls and the embryos in 0.005% cytosine arabinoside were early plutei. The eggs treated with 0.025, 0.05 and 0.1% cytosine arabinoside for 4 h were early plutei, but with an increasing number of late prism stages at the higher concentrations. The eggs exposed to 0.025 and

0.05% cytosine arabinoside for 8 h were mostly prism stages, while those eggs exposed to 0.1% cytosine arabinoside for 8 h were a mixture of early prism and late gastrula stages. On the fifth day of development the controls, 0.005 and 0.025% cytosine arabinoside (4 and 8 h) treated embryos were well developed plutei (Figure 1); however, there were more prism stages in the embryos exposed to 0.25% cytosine arabinoside for 8 h. The embryos exposed to 0.05 and 0.1% cytosine arabinoside for 4 and 8 h were at a late prism stage of development.

The sea-urchin embryos exposed during cleavage to certain concentrations of cytosine arabinoside for 4 and 8 h underwent a delay in development. These eggs resumed development after removal from cytosine arabinoside and appeared normal, although they were at earlier stages of development than the untreated embryos<sup>2</sup>.

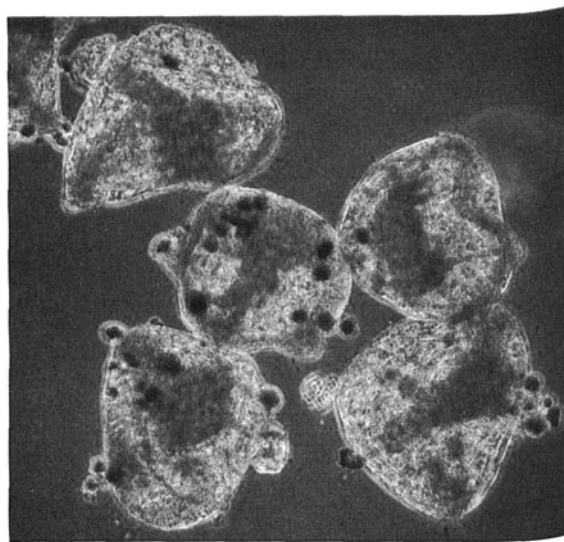


Fig. 2. These embryos had been treated with 0.05% cytosine arabinoside for 8 h starting 2½ h after fertilization. They have developed for the same length of time as the controls (Figure 1), but have only reached a prism stage. × 160.

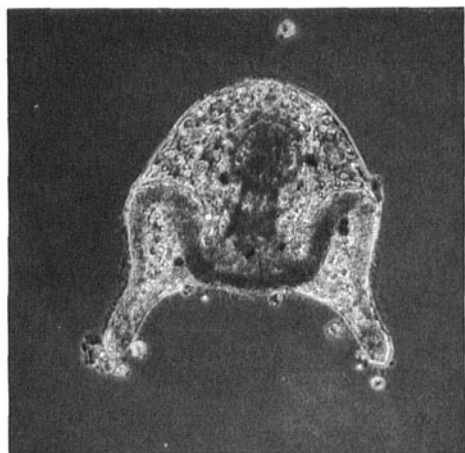


Fig. 1. A normal pluteus larva of *Sphaerochinus granularis* on the fifth day of development. × 160.

**Résumé.** Le développement des œufs d'oursins est ralenti par la cytosine arabinoside à laquelle on les expose pendant 4 à 8 h, mais il reprend son cours après le traitement.

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