A FUNCTION OF MESSENGER RNA AS A CYTOPLASMIC FACTOR IS INVOLVED IN POLE CELL FORMATION OF <u>DROSOPHILA</u> EMBRYOS

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Cytoplasmic factors have been postulated to perform important roles in pole cell formation and determination of the pole cells to differentiate into germ cells in <u>Drosophila</u> embryogenesis. One of the factors active in pole cell formation has been found precipitating with a postmitochondrial subcellular fraction prepared from homogenate of whole embryos. The factor lost its activity when the subcellular fraction was digested with RNase.

To find the molecule species responsible for pole cell formation, RNA was extracted from the postmitochondrial subcellular fraction from homogenate of whole embryos and assayed for its pole cell inducing activity. For assaying the activity, RNA was injected into the posterior pole of cleavage embryos that had been sterilized with radiation of ultra violet light (u.v.) with the wavelength of 280 nm at a dose of 200 J/m2, and the recipient embryos were allowed to develop up to blastoderm stage and examined for presence or absence of pole cells. Crude RNA fraction showed pole cell inducing activity. RNA was divided into two fractions, one with and the other without polyadenylate sequences $(poly(A)^+$, and $poly(A)^-$ RNA fraction). Only $poly(A)^+$ RNA fraction retained apparent pole cell inducing activity. The activity of a $poly(A)^+$ RNA fraction was depending on stages of embryos from which RNA was extracted. A high activity was retained in RNA fraction extracted from embryos at 20 min after egg laying (min AEL), but not in one from 150-min-AEL embryos. A cDNA library was prepared from a poly(A)⁺ RNA fraction extracted from 20-min-AEL embryos, and screened for cDNA clones that hybridize with RNA sequences speci-

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fically present in 20-min-AEL embryos with a competitive colony hybridization method. Hundred colonies have been selected and being under further screening for the sequences responsible for pole cell formation.

Treatment of cleavage with cycloheximide at the posterior pole prevented the embryos from pole cell formation without affecting somatic cells. This suggests that translation from messages is required in the posterior pole cytoplasm for accomplishment of pole cell formation.

Morphology of pole cells formed in u.v.-irradiated embryos injected with poly(A)⁺ RNA was described. These pole cells showed similar features to normal pole cells as far as structures TEM and SEM revealing were concerned. In addition, the RNA-induced pole cells were normal also for their behavior, migration to penetrate gonads. These pole cells were different from normal ones only in their developmental fate: for the present it has not been shown that the RNA-induced pole cells are able to differentiate into germ cells.

References

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