DB 78

CHANGE IN INTRACELLULAR DISTRIBUTION OF CALMODULIN DURIMG EARY DEVEROPMENT OF SEA URCHIN.

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Relative amount of calmodulin,estimated by its stimulation effect on bovine cAMP dependent phosphodiesterase,was high in cytosolic fraction than in nucleous and microsomal fraction up to 16hr after fertilization. The amount in nucleous fraction increased exponentially up to 24hr afterfertilization and then decreased markedly. The amount per DNA in nucleous fraction seemed essentially the same in deveropmental period up to 16hr after fertilization and decreased gradually. The amount in microsome fraction become high at 24hr and further at 32hr after fertilization Then,it decreased to a quite low level. At later period than 24hr,the amount in cytosolic fraction was lower than in microsome fraction.Whole amount of calmodulin become slightly low at 12 and 16hr,and then become high at 24 and 32hr after fertilization. It is likely that calmodulin plays a role in controling nuclear functions at the period up to 16hr after fertilization and thereafter contibute to morphogenesis in embryo.

DB 79

DISINTEGRATION OF PROTEINS IN YOLK GRANULES ISOLATED FROM SEA URCHIN EGGS.

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Proteins in the yolk granules isolated from eggs of Hemicentrotus pulcherrimus and Anthocidaris crassispina were analyzed by SDS-polyacrylamide gel electrophoresis. Proteins of molecular weights, 180K dalton in H. pulcherrimus and 178K in A. crassispina were the most abundant in yolk granules from unfertilized and fertilized eggs. After the incubation of yolk granules in acidic artificial sea water (pH4.2-6.0) for 24 hr, these proteins decreased, while proteins (61K,72K,94K,114K in H. pulcherrimus and 56K,70K,92K,112K in A. crassispina) predominant in the late stage of development appeared. Incubation of yolk granules in the alkaline or neutral medium did not result in the changes of electrophoretic patterns of proteins. It is suggested that acidic conditions are essential to the disintegration of yolk proteins. After the incubation in the acidic medium, yolk granules showed morphological characteristics similar to those observed in yolk granules of developed embryos. Since digestion of yolk proteins and morphological changes within yolk granules were inhibited by leupeptin, antipain, TLCK, iodoacetamide and $HgCl_2$, cathepsin B seemed the most important enzyme in the proteolytic processing of yolk proteins during embryogenesis.

DB 80

THE CHEMICAL CONDITIONS OF BLASTOCOELIC FLUID OF NEWT EMBRYOS

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Several reports hitherto have suggested that the action of the substance causing primary embryonic induction may be affected by environmental conditions such as salt concentration, pH or small molecular organic substances. Experiments to analyze the chemical conditions of intra- and extracellular fluid of newt gastrula were performed . Na^+ & K^+ ions, Ca^{++} ion, Cl^- ion and high molecular organic substances were analyzed by atomic absorption spectrochemical analysis, ICP emission spectrochemical analysis , ionic chromatography and SDS polyacrylamide gel electrophoresis, respectively. High pH of blastocoelic fluid (pH 9.2) seemed to be due to the sodium pumping action of embryonic cells towards blastocoel, because, first, blastocoelic fluid contained 1.7 mg/ml Na^+ ion, but intracellular fluid contained only 0.24 mg/ml, second, blastocoelic fluid showed no Nessler's reagent reaction. As expected, intracellular fluid contained more K^+ ion (0.77 mg/ml) than Na^+ ion, and little Ca^{++} ion. A scanty amount of two slow-moving bands were detected in the SDS PAGE of blastocoelic fluid. By exchange of blastocoelic fluid with equal osmotic pressure and different pH solution, apparently normal development was not disturbed.

DB 81

45,000 MW PROTEIN FROM THE SEA URCHIN EGG ACCELERATES THE POLYMERIZATION OF ACTIN IN THE EGG CORTEX.

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A one-to-one complex of a 45,000-mol-wt protein and actin has been purified from unfertilized eggs of the sea urchin, Hemicentrotus pulcherrimus. The complex (45K·A) is an actin filament capping-protein(barbed end)(J. Cell Biol.(1984) 99, 994-1001). By using the affinity purified monospecific antibody against the 45K protein, we have shown the protein localizes in the egg cortex and its content decreases after fertilization. It has been reported by many workers that numerous actin filaments appear in the sea urchin egg cortex after fertilization. Here, we studied the role of the 45K protein (or 45K·A) in the cortex by fluorescence microscopy using an F-actin-specific stain, NBD-phallacidin in order to elucidate the mechanism of the appearance of the actin filaments. The antibody blocked the assembly of actin filaments in the isolated cortex on a glass slide and the removal of 45K protein from the cortex by 1M KCl did not cause the polymerization of actin.