REVERSING CYTOPLASMIC FLOW IN CONSTRICTED EGGS OF THE SAND DOLLAR (ECHINARACHNIUS PARMA)

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It is generally agreed that cytokinesis in animal cells results from the contractile activity of a short-lived band of equatorial surface that exerts measurable force and is characterized by specific ultrastructure. Presently, some controversy exists concerning the general mechanism by which the asters can create this specialized region in a surface that shortly before was uniformly capable of response. The tension difference between the equator and the rest of the surface could arise from two alternative hypothetical mechanisms, either reduction in tension at the poles or increase in tension at the equator. The uncertainty is in part related to present inability to demonstrate regional differences in surface physical properties before division begins.

In normal spherical sand dollar eggs, the size of the mitotic apparatus (MA) and the egg shape are such that all parts of the surface appear likely to be affected by the MA shortly before division. This relationship was changed by snaring a fertilized egg between opposed, overlapping hooks and constricting the cell so that it was reshaped from a single sphere with a 68μm radius into two partial spheres with a 55 μm radius joined by a constricted neck 27 μm in diameter. When both asters of the MA lay on the same side of the constriction, cytoplasm flowed out of the nucleated part across the constriction, beginning about 30 min before anticipated cleavage time. After the asters moved across the constriction with the cytoplasm, the direction of flow was reversed, and reversal usually occurred several times before cleavage (n=18). Cells were bisected by hand and constricted as described in order to determine the role of the MA. Bisected cells with two asters behaved like constricted normal cells (n=15). Cells with no MA material developed no cytoplasmic flow (n=11). In cells with one aster, flow occurred only at anticipated cleavage time (n=14). When whole constricted eggs were treated with Cytochalasin B at the minimum concentration that blocked cleavage in otherwise normal eggs (3 μg/ml), neither cytoplasmic flow nor cleavage occurred, although the MA developed normally (n=13). Cytochalasin B interferes with actin polymerization.

These results indicate that the cytoplasmic flow of constricted eggs in cleavage stages requires asters and functional actin-associated cytoskeleton. They are consistent with a hypothetical mechanism in which asters cause tension increase in nearby surface. They are not predicted by a hypothetical mechanism in which the asters cause surface relaxation. Their relation to changes in surface stiffness that can be demonstrated during the cell cycle remains to be elucidated.

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