

REINVESTIGATION OF REGIONAL DIFFERENCES IN BURSTING STRENGTH OF CLEAVING SAND
DOLLAR (ECHINARACHNIUS PARMA) EGGS

R. Rappaport and Barbara N. Rappaport
Mount Desert Island Biological Laboratory
Salsbury Cove, ME 04672

Cytokinesis in animal cells is caused by physical activity of the cortical cytoskeletal region located immediately beneath the plasma membrane. A better understanding of possible regional differences in the physical properties of the cortex could reveal some aspects of the formation of the division mechanism. In 1922, E. E. Just (Am. J. Physiol. 61: 505-15) concluded that the polar surfaces of spherical sand dollar eggs were mechanically weaker than the future furrow region immediately before cleavage, because eggs immersed in dilute sea water at that time "invariably" ruptured over the poles. These observations have been accorded considerable theoretical significance. Just did not remove the extracellular hyaline layer from the eggs and, because it could have contributed to the apparent strength of the surface, we repeated his work and compared the cell's behavior in the layer's presence and absence.

A conical chamber containing 40% sea water, 60% fresh water was mounted on the stage of an inverted microscope. Ten to twelve min before anticipated time of cleavage, a suspension of eggs in a few μ l of normal sea water was pipetted to the surface of the water in the observation chamber. The eggs fell through 17 mm of dilute sea water to the chamber bottom where their subsequent behavior was videotaped. Only those eggs in which the orientation of the mitotic apparatus (MA) and the point of rupture could be clearly seen were included in the data. The point of rupture was marked on a previously drawn cell profile that was divided into polar, subpolar, subfurrow and furrow regions. Fertilized eggs with intact jelly, fertilization membranes and hyaline layer, and fertilized eggs from which the jelly, membrane and hyaline layer were removed by treatment with 1.0 M glycine were studied.

Contrary to Just's findings, we observed that ruptures were not localized in any particular region. When the eggs with or without jelly and membranes were immersed in dilute sea water, the percentage of ruptures in the surface regions roughly approximated the percentage of the total surface that the regions comprised. The furrow, subfurrow, subpolar and polar surfaces comprise, respectively, 18, 33, 26 and 23% of the total egg surface. In 131 eggs with intact extracellular layers, the regional distribution of rupture points was, in percentages, furrow: 27%, subfurrow: 40%, subpolar: 18%, and polar: 16%. In 79 eggs lacking extracellular material following glycine treatment and immersion in 50% calcium free sea water, the distribution of rupture points listed in the same order was 22%, 26%, 22% and 29%.

These data indicate no localized region of cortical weakness immediately before division. We speculate that Just's results may have resulted from his method and timing of observations (which were not clearly described). We found that the outflow of cytoplasm that followed rupture caused internal cytoplasmic rearrangement that resulted in reorientation of the MA so that its axis was parallel to the direction of outflow. Shortly after rupture, the pole of the MA was repositioned close to the point of outflow. To someone who did not observe the instant of rupture, the outflow would appear to have originated at the pole of one of the asters.

This work was supported by National Science Foundation Grant DCB-8903341.