THE ROLE OF MICROTUBULES IN THE FORMATION AND RELAXATION OF CONTRACTILE RINGS IN ILYANASSA OBSOLETA

Abigail H. Conrad and Gary W. Conrad
Division of Biology, Kansas State University, Manhattan, KS 66506

At the time of first cleavage, the Ilyanassa obsoleta Stimpson (=Nassarius obsoletus Say) zygote forms two contractile ring structures at right angles to each other within the same cell, the cleavage furrow and the third polar lobe constriction. The cleavage ring contracts to a tight neck and pinches the two resulting daughter cells apart, while the third polar lobe ring contracts to a tight neck and then relaxes again, allowing the polar lobe cytoplasm to be merged with one of the two mitotic daughter cells. Immunocytochemical localization of alpha-tubulin in permeabilized extracted cells reveals significant alpha-tubulin presence in the animal poles of the daughter cells and in the cleavage furrow midbody, but not in the polar lobe or the polar lobe constriction of the normal zygote. Electron micrographs of the third polar lobe neck at the time of maximum contraction reveal a few microtubules at the animal hemisphere end of the neck, but there are no midbody condensations associated with these polar lobe neck microtubules, such as are found in association with the many microtubules running through the tight neck cleavage furrow constriction. When 10 uM taxol, a drug which stabilizes microtubules, is added to eggs prior to the onset of third polar lobe constriction and mitotic cleavage, the site of initiation and the number of cleavage furrows formed can be markedly altered, while the number of and site of initiation of the third polar lobe constriction remains unchanged. The polar lobe ring contracts to a tight neck, whether or not a cleavage ring forms, and this polar lobe constriction ring does not relax. Furthermore, the lobe itself often becomes crenulated. If taxol is added to eggs at the time of third polar lobe initiation, but before cleavage furrow initiation, subsequent cleavage ring location, initiation, and contraction and polar lobe contraction occur normally, but again the polar lobe constriction does not relax, the polar lobe cytoplasm remains sequestered in the lobe, and the polar lobe often separates from the animal hemisphere daughter cells. Thus, the polar lobe constriction is converted into a cleavage furrow. Immunocytochemical localization of alpha-tubulin in taxol-treated, permeabilized, extracted eggs shows greatly increased alpha-tubulin localization throughout the daughter cells, in the polar lobe neck, and in the polar lobe itself. Electron micrographs reveal the presence of greatly increased numbers of microtubules in both the polar lobe neck and polar lobe cytoplasm of taxol-treated eggs.

These results suggest, first, that when microtubules are stabilized in areas of cells enclosed by contractile rings, cytoplasmic contractile rings constrict, become stabilized in their contracted state, and do not relax. Under these conditions, polar lobe constrictions often become cleavage furrows. Second, the position of the polar lobe contractile ring is determined independently of mitotic spindle microtubule configuration, whereas the location of the cleavage furrow contractile ring is determined by spindle astral microtubules. Third, once a constriction ring is positioned, the stimulus to contract is, or can be, delivered to the cell cortex by a microtubule-independent mechanism. Fourth, the recruitment of contractile elements appears to be dependent upon microtubule induction in both cleavage furrow and polar lobe contractile rings.

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