# MICROTUBULES AND ACTIN FILAMENTS ARE INVOLVED IN FACILITATING OF FUSIONS AND FISSIONS OF VESICLES OF EGF-RECEPTOR COMPLEXES' ENDOCYTIC PATHWAY

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Effect of microtubule depolymerization by nocodazole and actin cytoskeleton disassembly by cytochalazine D alone and in combination, on the dynamics of endocytosis of epidermal growth factor receptor complexes with EGF (EGF-R) in HeLa cells was studied. The use of confocal microscopy on the cells fixed at different time intervals after stimulation of endocytosis showed that at the early stages there is a high degree of the receptor colocalization with early endosome marker EEA1, which correlates with enlargement of the endosomes as a result of fusions, and the movement of endosomes at later stages to the perinuclear region (PR). Translocation to PR depends on microtubules. Nocodazole does not prevent endosomes' formation, but blocks their fusions and movement. At the same time, the destruction of actin microfilaments, without disturbing the process of EEA1-dependent fusions and the transport of endosomes in the PR, leads to enlargement of endosomes, preventing the separation of early endosomes and mature multivesicular endosomes. With the destruction of both types of cytoskeleton, the newly formed receptor-containing endocytic vesicles do not enlarge, do not mature and do not move to the PR. Live imaging of endosomes using EGF-quantum dots complexes confirmed the data obtained on fixed cells and allowed us to analyze the nature of the movements of endosomes. It turned out that rare periods of fast linear runs alternates with more frequent periods of chaotic oscillations within a small area. The directions of linear paths can be different, and not just in the direction of MTOC. Our data suggest that the role of MT is not only to facilitate endosomes' movements, but also to provide a platform for their fusion and fissions. This assumption can also explain the seemingly chaotic movements of endosomes, which increase the probability of their contacts and, therefore, fusions.

***Keywords:*** EGF receptor, endocytosis, endosomes, fusion, EEA1, microtubules, microfilaments, nocodazole, cytochalazine D