Quantification of Receptor Internalization via Q-dot labeling and Multi-Scale Image Segmentation

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The activation of epidermal growth factor receptor (EGF-r) by ligand or antibody can be of significance in understanding the kinetics of its internalization in various cell lines and hence serve as a discovery tool in cell biology research in general and in developing cancer therapies in particular. The ability to dynamically monitor EGFr internalization has been improved recently by the use of appropriately functionalized quantum dots (Q-dots), due to the long lasting fluorescence and brightness of these nanoparticles. However, quantitative information on EGFr kinetics is derived based on average properties of cell cultures, rather than tracking individual events within one cell, and relative positional information is gained from traditional staining procedures prone to noise, fading and low contrast. We apply confocal microscopy to image cell cultures followed by multiscale image segmentation to define regional sectors in cells prior to determination of concentration of Q-dots. This allows quantitative description of the internalization of EGFr activated by a biotinylated EGF/streptavidin quantum dot complex with A431 cells at various time periods. The capabilities realizable by the simultaneous application of specialized high resolution imaging techniques and functionalized quantum dots in conjunction with advanced image analysis can enhance our understanding of receptor dynamics. Targeting multiple species in the EGF pathway enhances our ability to develop spatio-temporal models of cellular dynamical processes.