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Optoporation to deliver impermeable molecules and genes for visualization and activation of cells KAMAL DHAKAL, SUBRATA BATBYAL, YOUNG-TAE KIM, SAMARENDRA MOHANTY, Univ of Texas, Arlington — Visualization, activation, and detection of the cell(s) and their electrical activity require delivery of exogenous impermeable molecules and targeted expression of genes encoding labeling proteins, ion-channels and voltage indicators. While genes can be delivered by viral vector to cells, delivery of other impermeable molecules into the cytoplasm of targeted cells requires microinjection by mechanical needle or microelectrodes, which pose significant challenge to the viability of the cells. Further, it will be useful to localize the expression of the targeted molecules not only in specific cell types, but to specific cells in restricted spatial regions. Here, we report use of focused near-infrared (NIR) femtosecond laser beam to transiently perforate targeted cell membrane to insert genes encoding blue light activatable channelrhodopsin-2 (ChR2) and red-shifted opsin (ReachR). Optoporation of nanomolar concentrations of rhodamine phalloidin (an impermeable dye molecule for staining filamentous actin) into targeted living mammalian cells (both HEK and primary cortical neurons) is also achieved allowing imaging of dynamics and intact morphology of cellular structures without requiring fixation.

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