Nanoscale organization of focal adhesions, vimentin filaments and cell stiffness

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Abstract

Focal adhesions, subcellular regions in the micrometer scale that link the cell to its surrounding and regulate cell function have been extensively investigated. However, the spatial architecture of focal adhesions at the nanometer scale remains unclear. We have used three- and two-color super-resolution STED microscopy, and computational image analysis, to determine the spatial organization of components of focal adhesions at nanoscale resolution. Our data are consistent with the concept that adhesion proteins within, but not outside of, focal adhesions are organized into distinct nanoscale protein assemblies. This knowledge of the organization of focal adhesions at the nanometer scale has furthered our understanding of the basis of cell adhesion in health and disease.

We could in addition detect that oncogenes induce a nanoscale reorganization of both the nanoscale adhesions and vimentin intermediate filament fibers, as quantified by image analysis. This reorganization was linked to changes in the stiffness of the cells. Taken together, our data are consistent with the hypothesis that oncogenes change the organization of nanoscale adhesions and cellular stiffness via a reorganization of the vimentin intermediate filament network.

References:

1. Xu. L, Braun L.J., Rönnlund D., Widengren J., Aspenström P., Gad A.K.B. Nanoscale localization of proteins within focal adhesions suggests discrete molecular assemblies. Manuscript.

2. Rathje L.S., Nordgren N., Pettersson T., Rönnlund D., Widengren J., Aspenström P., Gad A.K. (2014)

Oncogenes induce a vimentin filament collapse mediated by HDAC6 that is linked to cell stiffness. PNAS 111, Jan 13.

3. Rönnlund D., Gad A.K., Xu L., Blom H., Aspenström P., Widengren J. (2013). Spatial organization of proteins in metastasizing cells. Cytometry A, 2013.