Imaging HIV Transfer between T Cells with Optical Superresolution Microscopy

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Live cell imaging with molecularly specific contrast is a particular strength of optical microscopy, permitting the dynamic observation of events at the single cell level. This provides us with highly quantitative biophysical data e.g. to unravel the systems biology of HIV-1 transfer between cells. Most live cell imaging experiments make use of the high specificity provided by labeling cellular components with fluorescent dyes and fluorescent proteins. Here, I will present our latest data on tracking HIV-1 transfer between cells by conducting 4D live cell fluorescence microscopy with a replication-competent clone of the virus. By combining 4D live cell microscopy with holographic optical tweezers we are able to actively manipulate infected and uninfected cells within the field of view of the microscope and test the parameters affecting the formation of the virological synapse between cells. To further dissect the spatiotemporal architecture of these adhesive structures between T cells we utilize 3D structured illumination super-resolution microscopy which allows us to resolve individual virus particles. This permits us to exercise great control - temporally and spatially - over the events preceding and following the transfer of virus from infected to uninfected cells and determine the nature of the transfer mechanism.

References:

[1] B.M. Dale et al, "Endosomal HIV-1 particle maturation activates viral membrane fusion following transfer across virological synapses", *Cell Host and Microbe* **10**, 551-562 (2011)

[2] B. Dale et al, "Tracking and quantitation of fluorescent HIV during cell-cell transmission", *Methods* **53**(1), 20 - 26 (2011)

[3] B. Dale et al, "Visualizing cell-to-cell transfer of HIV using fluorescent clones of HIV and live confocal microscopy", *J. Vis. Exp.* **44** (2010), DOI: 10.3791/2061

[4] G.P. McNerney et al, "Manipulating CD4+ T cells by optical tweezers for the initiation of cell-cell transfer of HIV-1", *J. Biophoton.* **3**(4), 216 – 223 (2010)

[5] W. Hübner et al, "Quantitative 3D Video Microscopy of HIV Transfer Across T cell Virological Synapses", *Science* **323**, 1743-1747 (2009)