NON-RADIATIVE EXCITATION FLUORESCENCE MICROSCOPY FOR STUDYING MEMBRANE ADHESION AT THE NANOSCALE

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Non-Radiative Excitation Fluorescence Microscopy (NEFM) [1] constitutes a new way to observe biological samples beyond the diffraction limit. By coating a substrate with an homogeneous monolayer of quantum dots (QDs), Förster Resonance Energy Transfer (FRET) could be achieved between the QDs layer (which play the role of the donor) and Giant Unilamellar Vesicles (GUVs) labelled with DiD (which play the role of the acceptor). The dyes were not directly excited by the laser source but through a non-radiative energy transfer. GUVs were added on a QD layer coated with poly-L-lysine (electrostatic attraction occurred between the positively charged surface and negatively charged GUVs). On this kind of sample, we were able to observe at the same time the emission of the DiD and the quenching of the QDs (as shown in figure 1). It clearly indicates that non-radiative energy transfer occurs from the QDs to DiD. From these two pictures, we also calculated the distance between the lipid membrane and the surface for each pixel with a nanometric resolution (figure 1C).

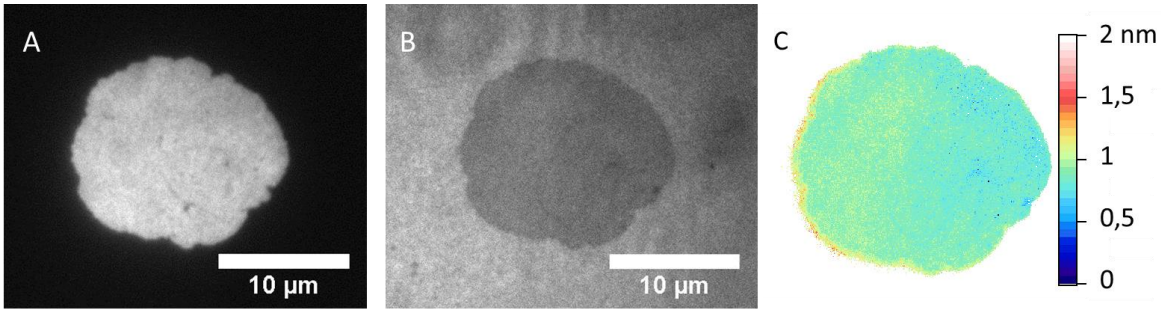

Figure 1: Giant Unilamellar Vesicle spread on a poly-L-lysine surface. The vesicle was labeled with DiD. (A): Emission of the DiD, (B): quenching of the QD layer coated with poly-L-lysine (bar = 10 µm), (C) corresponding membrane topography.

[1] P. Winckler, R. Jaffiol, J. Plain, and P. Royer. "Nanoradiative Excitation Fluorescence: Probing Volumes Down to the Attoliter Range." The Journal of Physical Chemistry Letters 1, 2451-2454 (2010).