

# Nano antenna - FCS: Visualizing membrane heterogeneity in living cells with single molecule resolution

Raju Regmi<sup>1,2</sup>, Pamina M. Winkler<sup>2</sup>, Valentin Flauraud<sup>3</sup>, Kyra J. E. Borgman<sup>2</sup>, Carlo Manzo<sup>2</sup>, Jürgen Brugger<sup>3</sup>, Hervé Rigneault<sup>1</sup>, Jérôme Wenger<sup>1</sup> and María F. García-Parajo<sup>2,4</sup>

<sup>1</sup> Institut Fresnel, CNRS, Aix-Marseille Université, Marseille, France

<sup>2</sup> ICFO-Institut de Ciències Fòniques, Barcelona, Spain

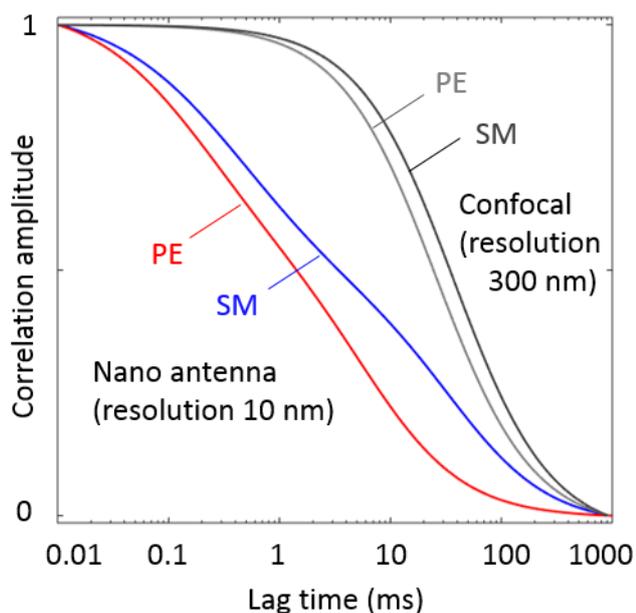
<sup>3</sup> Microsystems Laboratory, École Polytechnique Fédérale de Lausanne, Switzerland

<sup>4</sup> ICREA, Pg. Lluís Companys, Barcelona, Spain

E-mail : raju.regmi@fresnel.fr

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The spatiotemporal architecture of the cell membrane and diffusion dynamics of its constituents (lipids and proteins) are primary for cellular processes such as signaling and trafficking. In this work, we propose in-plane plasmonic nano antennas and study diffusion characteristics of phosphoethanolamine (PE) and sphingomyelin (SM) in Chinese hamster ovary cells by incorporating fluorescent lipid analogs. The enhanced electric fields in such nanoscale probe areas ( $\sim 300 \text{ nm}^2$ ) yield a high signal to noise ratio ultimately making it possible to follow single molecule events at physiological expression levels. Fluorescence correlation spectroscopy and burst analysis of these events show free 2D Brownian diffusion for PE (with both confocal and nano antennas). However the diffusion dynamics of SM at the nanoscale indicates extremely heterogeneous behavior which was otherwise hidden in the confocal ensemble (Fig.1). Further measurements on cholesterol depleted cells with fluorescent SM show diffusion behavior similar to that of PE-like scenario. This is attributed to the fact that SM is preferentially associated to cholesterol assisted small sub compartments (lipid rafts).



**Figure 1:** Normalized correlation curves of Atto647N labelled with PE (red), and SM (blue) probed with nano antennas.

Apart from the findings presented here which are crucial to understand the spatiotemporal and heterogeneous organization of lipid compartments in living cells at the nanoscale; the proposed technique is fully biocompatible and thus provides various opportunities in biophysics and live cell research.