Séminaire ibs

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Conférencier invité

Vendredi 28 Sept.

A 11h - Salle des séminaires de l'IBS

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Novel probes and strategies for high resolution fluorescence imaging in living cells

High-resolution optical imaging techniques make now accessible the detection of biological nanofeatures in living cells by non-ionizing visible radiation. However, high-resolution imaging is critically dependent by the selected fluorescent probes- Here we shall review our recent work on the development of new fluorescent molecular systems.

Photochromic fluorescent proteins. The introduction of the E222Q mutation in the primary sequence confers good photochromic properties to several otherwise photostable *Aequorea Victoria* fluorescent proteins. We demonstrated that at physiological pH and above the photoswitching of the anionic *cis* protein chromophore yields a neutral non-fluorescent *trans* state, providing the optical basis for *on*<=>*off* cycling of fluorescence.The significance of these mutants for high-resolution cell imaging was shown by means of photochromic FRET experiments and OLID imaging.

Environmental probes. Fluorescent sensors of nanoenvironmental physicochemical properties such as polarity and viscosity are particularly interesting in view of understanding subtle cellular processes. Ideally, environmental probes should fulfill these requirements: a) optical responses (intensity, wavelength-shift, lifetime) strongly and predictably related to the environmental polarity or viscosity changes, b) brightness allowing for single-molecule detection, c) easily conjugable to biomolecules. Following these guidelines, at first we have developed a toolbox of push-pull solvatochromic coumarins virtually non emissive in water, but intensely fluorescent in less polar media.Next, we developed fluorescent protein chromophore analogs capable to report efficiently on the local dielectric constant in biological specimens. By confocal microscopy, we applied these probes to obtain spatially resolved ε maps of cultured cells.

Hôte : D. Bourgeois (IBS/DYNAMOP)