

Out-of-equilibrium active membrane

DIRECTEUR DE THESE : THIERRY CHARITAT

TEL : 03 88 41 40 05 ; E-MAIL : thierry.charitat@ics-cnrs.unistra.fr

Biological membranes are known to exhibit thermal fluctuations. Close to equilibrium properties have been widely investigated, both theoretically and experimentally, in living systems and on model systems like multilamellar stacks or giant unilamellar vesicles. In living systems, the transport of small molecules across the cytoplasmic or internal organelles membrane plays a crucial rôle. For example, it is essential in ensuring the integrity of organelles, such as endosomes, endoplasmic reticulum, and Golgi... In many cases, the transport process through the membrane involves specific membrane proteins, which use metabolic energy as ATP hydrolysis or photochemical reactions to process conformational change. This protein activity breaks the fluctuation-dissipation theorem leading to out-of-equilibrium fluctuations. A complete understanding of the mechanism at play in fluctuation's activation implies a fine characterization of the fluctuation spectrum of fluctuations at sub-micronic length scales. The first part of the work will involve the reconstitution of a transmembrane active protein as bacteriorhodopsin or Sarcoplasmic reticulum Ca²⁺-ATPase (SERCA1a) in supported single and double bilayer by using detergent-mediated incorporation method. This new technique was recently developed at the Institut Curie in Paris to reconstitute transmembrane proteins in Giant Unilamellar Vesicles. The first step of the project will be to extend this method to supported lipid bilayers. We will try to incorporate the bacteriorhodopsin, a light driven proton pump, in single and double supported bilayers and to characterize the protein incorporation and activity. The second main part of the PhD will be devoted to perform combined specular and off-specular neutron and x-ray reflectivity experiments on double supported bilayer decorated by active proteins. Again, we will need to develop a specific cell to perform this experiments under light illumination for protein activation.

The PhD will benefit from all the experimental techniques available in both ILL and ICS laboratories. In particular, Fluorescence Microscopy will be used to have a first check of BR incorporation in bilayer. Fluorescence Recovery After Patterned Photobleaching, an original technique available at ICS will be used to probe the ability of proteins to freely diffuse in the bilayer. Neutron reflectivity experiments will be of high importance to understand the structural properties of supported bilayers with inserted proteins and to indirectly determine the effect of proteins activation on the floating bilayer fluctuations. Reflectivity measurements will be carried out also at the European Synchrotron Radiation Facility in Grenoble.

The PhD project will be located primarily in Grenoble (France), at the ILL (under the supervision of Giovanna Fragneto). Stays at the ICS (Strasbourg) (under the supervision of Thierry Charitat) are foreseen as well as visits to the synchrotron SOLEIL and the Institut Curie (Paris). The successful candidate will be employed for a period of up to three years, with a gross salary of around 2350 €/month, together with other benefits depending on the student's social status (for more details see: <http://www.ill.eu/science-technology/phd-students/phd-recruitment/phd-work-at-the-ill/>). A team of experts, including Jean Dailant (SOLEIL) and Patricia Bassereau (Institut Curie, Paris) will participate and advice the work of the PhD student.

Supervisors: Pr. Thierry Charitat, Univ. of Strasbourg, and Giovanna Fragneto, ILL (Grenoble).

Contact information: thierry.charitat@ics-cnrs.unistra.fr, fragneto@ill.fr