Functional characterisation of membrane proteins stably incorporated in tethered lipid bilayers

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Systems composed of membrane proteins (MP) embedded in lipid bilayers have relevant potential application in biomedical science. They have the potentiality of becoming the core of novel biomimetic implantable devices with a wide range of applications, such as the monitoring of chemical messengers and biological markers inside the body for improved prognosis, energy scavenging devices to produce energy directly within the body of a patient or animal, or new drug delivery systems. We investigated the functional incorporation of different proteins into tethered lipid bilayer membranes (tBLMs). tBLM consists of a planar lipid bilayer supported by an array of tethered lipids anchored to a molecularly smooth substrate [1]. tBLMs are more stable due to the chemical attachment to the substrate necessary condition for the design of practical devices.

We will present a nanostructural and functional characterisation of two important MPs performed by a combination of techniques including Neutron Reflectometry and Electrical Impedance Spectroscopy [2]. *OprF*, the main porin of the P. Aeruginosa bacterium outer membrane, is thought to have an important role in the antibiotic resistance of the bacterium that is responsible for 10% of all hospital-acquired infections. OprF is a very novel target for new drugs that will kill bacteria resistant to standard antibiotics. In our laboratories a cell-free method to synthesis the protein has been developed to provide controlled studies of the structure and function of OprF [3]. The *NhaA* protein is a pH-dependent sodium-proton antiporter found in prokaryotes that was used by our laboratory to develop a prototype of energy device that transforms a gradient of salt to an electrical potential [4]

These studies showed that MPs purified using optimised protocols and then incorporated into tethered lipid bilayers yield controllable (and measurable at the nanoscale) biomimetic systems constructed in vitro. These systems provides the means to identify ways to control membrane proteins, and hence to foster the development of novel biomimetic devices.

[1] M. Maccarini, B. Stidder, J-P. Alcaraz, E. B. Watkins, B. A. Cornell, D. K. Martin '*Nanostructure determination of lipid bilayers tethered to gold substrates*' Eur. Phys. J. E (2016) 39: 123
[2] M. Maccarini, L. Gayet, B. Stidder, J-P. Alcaraz, E. Watkins, L. Liguori, J-L. Lenormann, D. Martin '*Functional nanostructure of the Pseudomonas aeruginosa porin OprF incorporated into a tethered lipid bilayer membrane'* Langmuir (2017) 33(38), 9988
[3] L. Liguori, B. Marques, A. Villegas-Mendez, R. Rothe, J-L. Lenormand '*Liposomes-mediated delivery of proapoptotic therapeutic membrane proteins'* Journal of Controlled Release (2008), 126: 217-227
[4] Cinquin P, Martin DK (2007). "Biomimetic artificial membrane device" PCT/EP2008/058253, WO/2009/003936