Experimental Report

Council: 4/2014 Proposal: 9-13-544

Title: Porphyrin adsorption and penetration in a polymer-cushioned bilayer

This proposal is a new proposal

Researh Area: Soft condensed matter

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Samples: Porphyrin

poly(ethylene oxide) - H-(CH4O)n-OH

POPC - C42H82NO8P

(d31)POPC - C42H51NO8PD31

Instrument	Req. Days	All. Days	From	То
D17	2	2	14/11/2014	16/11/2014

Abstract:

Photodynamic therapy is based on the interaction, in the presence of oxygen, of light with a photosensitive drug that is inactive and nontoxic in the absence of light. Drug molecules bind more or less specifically to the cell membrane and penetrate into the cytoplasm. Subsequent illumination with a laser of an appropriate wavelength induces a photochemical reaction, which produces phototoxic substances such as singlet oxygen or free radicals, leading to tissue necrosis. Photodynamic therapy has many applications, and is considered a promising treatment against retinoblastoma, a malignant childhood intraocular tumour. Porphyrin derivatives are a class of drug molecules that can be used in this therapy. However, these molecules must be chemically modified to tailor specific interaction with cell membranes. In this context the study of the interactions between a model lipid bilayer and novel synthetic porphyrin derivatives is crucial. Understanding the type of interaction and the modifications induced by porphyrin derivatives on the lipid bilayers will help to develop better drug molecules and to understand the recognition mechanism.

Experiment 9-13-544 Porphyrin adsorption and penetration in a polymer-cushioned bilayer

The aim of the experiment was to assess the effect of a glycodendrimeric phenylporphyrin on a POPC bilayer formed on top of a poly(ethylene) oxide (PEO) cushion. Previous experiments using a quartz crystal microbalance with dissipation measurement, coupled to electrochemical impedance spectroscopy, have shown that although this porphyrin was unable to penetrate deeply into the lipid bilayer, it provoked changes in molecules organisation leading to higher electrical resistance. We used NR to get a better insight into the organisation of the tethered lipid bilayer and the mechanism of interaction between the porphyrin and the lipid molecules.

Liposomes were prepared by the Bangham's method, followed by extrusion through 100 nm and 50 nm in diameter polycarbonate membranes.

Three bilayer formation protocols were tested:

a) POPC liposomes rupture onto the SiO₂ surface *in situ* on D17: formation of a symmetric *h*- or *d*-POPC bilayer; Analysis before and after *h*- or *d*-TPP injection (Figure 1);

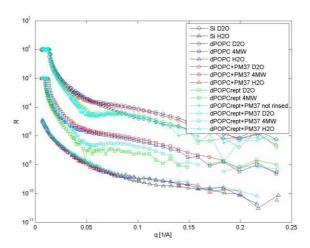


Figure 1: d-POPC symmetric bilayer prepared from liposomes, before and after contact with d-TPP

Supported POPC bilayers were easily obtained by liposome rupture. Preliminary analysis showed that the porphyrin did not affect the bilayer.

b) LB transfer of a h-POPC-DSPE-PEG₂₀₀₀ (97:3) monolayer onto the SiO₂ surface, and rupture of d-POPC liposomes $in\ situ$ on D17: formation of an asymmetric bilayer. After analysis of the bilayer, the h-TPP was injected into the aqueous medium (Figure 2);

The preliminary fitting indicated the successful formation of the asymmetric bilayer.

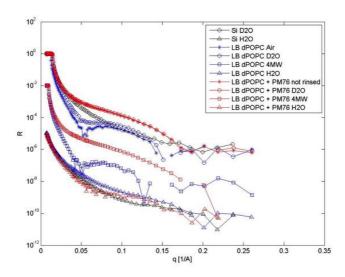


Figure 2: h-POPC-DSPE-PEG₂₀₀₀ (97:3)/d-POPC asymmetric bilayer prepared from LB transfer and rupture of d-POPC liposomes, before and after contact with h-TPP.

c) LB of a h-POPC-DSPE-PEG₂₀₀₀ (97:3) monolayer onto a SiO₂ surface, and then LS transfer of a d-POPC monolayer onto the first monolayer: formation of an asymmetric bilayer and transfer to D17. After analysis of the bilayer, the d-TPP was injected into the aqueous medium (Figure 3).

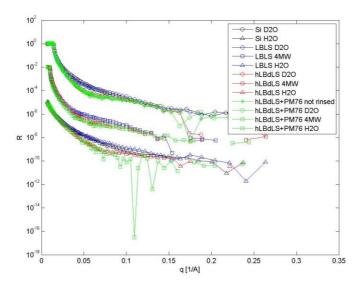


Figure 3: *h*-POPC-DSPE-PEG₂₀₀₀ (97:3)/*d*-POPC asymmetric bilayer prepared by LB-LS transfer, and analysed before and after contact with *d*-TPP

The first trend given by the fitting indicates that the bilayer was probably incomplete. The interaction with the porphyrin did not produce any change in the bilayer.

Analysis of the data is still under progress. A final report will be uploaded in the next few weeks.