PP-50, pH sensitive polymer interaction with DOPC membrane using Fluorescence Lifetime Correlation Spectroscopy (FLCS) and Electrochemical Impedance spectroscopy (EIS)

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Abstract

Amphiphilic polymers can strongly modulate membrane permeability, alter the curvature energy of the membrane and haemolyse red blood cells. They have been widely used in drug delivery/targeting systems, stabilization of membrane proteins, gene therapy and to drive uptake of cryoprotection agents. PP-50, the focus of this study, is an amphipathic membrane permeabilizing polymer designed to enhance trehalose loading into human erythrocytes to promote their cryopreservation[i] . Though the promoted loading of a variety of hydrophilic molecules into human erythrocytes by PP-50 has been demonstrated its influence on the structural and functional dynamics of the biological membrane has yet to be explored. Dynamic biophysical methods can provide important insights into interactions of these polymers with the cell membrane in real time leading to improvements on their design. Here, the interaction of PP-50 with model biological membranes was elucidated using optical and electrochemical techniques. The 23 kDa PP-50 polymers are made from biodegradable polyamide [poly(L-lysine iso-phthalamide)] grafted with L-phenylalanine, they possess ionisable protons which renders their behaviour pH responsive. The effect of introduction of PP-50 to 1,2-Dioleoyl-*sn*-glycero-3-phosphocholine (DOPC) lipid bilayers assembled at an aqueous filled microcavities on the fluidity of the bilayers was evaluated using fluorescence lifetime correlation spectroscopy (FLCS)[ii] where β-BODIPY-HPC(530/550nm) was used as fluorescent lipid marker along with Atto655 labelled-PP-50 to study the changes in the mobility of lipids and PP-50 polymers as a function of pH and time. The changes in the resistivity of lipid bilayers in the presence of PP-50 polymers on gold modified microcavities were studied by electrochemical impedance spectroscopy (EIS). We observe that both electrochemical impedance of the lipid layer and the diffusion coefficients of the lipid are significantly modulated by the interaction of PP50 and rationalise this behaviour on the basis of adsorption and penetration of the polymer through the bilayer.

[i] Lynch L.Andrew, Chen Rongjun, Slater K.H.Nigel, pH-responsive polymers for trehalose loading and desiccation protection of human red blood cells (2011). Biomaterials,32:4443-4449

[ii] Basit Hajra, Lopez G.Sergio, Keyes E.Tia, Fluorescence correlation and lifetime correlation spectroscopy applied to the study of supported lipid bilayer models of the cell membrane (2014). Methods, 68(2):286-299.