The Effect of α-Tocopherol on the Membrane Dipole Potential

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Abstract

α-Tocopherol has a well known antioxidant action but is also considered likely to exert significant non-antioxidant effects in cell membranes. Due to its lipophilic nature α-tocopherol inserts into biological membranes where it influences the organisation of the component lipids and may therefore influence biophysical parameters including the membrane dipole potential. The dipole potential has been demonstrated to modulate the function of several membrane associated proteins and perturbation of this physical parameter by α-tocopherol may prove to be a significant non-antioxidant mechanism underlying several of its cellular effects. This study investigates the influence of α-tocopherol, and the non-antioxidant structural analogue α-tocopherol succinate, on the membrane dipole potential employing fluorescence spectroscopy techniques with the dipole potential sensitive probe Di-8-ANEPPS. Similar techniques are utilised with the surface potential sensitive probe FPE to investigate the interaction of the charged α-tocopherol succinate molecule with membranes. α-Tocopherol and α-tocopherol succinate are shown to decrease the dipole potential of egg-phosphatidylcholine vesicles and Jurkat T-lymphocyte cell membranes. This effect is placed in the context of the significant influence of membrane cholesterol oxidation on the dipole potential. 7-ketocholesterol, an oxidised form of cholesterol, significantly influences several cellular processes and is thought to mediate these effects, in part, through its physical effects on the cell membrane. These include altering the composition, and therefore biophysical properties, of rafts; structures which are considered to support the function of a host of membrane proteins. This study attempts to correlate the effect of 7-ketocholesterol on the dipole potential of microdomains with the influence of the oxysterol on the function of two microdomains associated receptors: P-glycoprotein and the insulin receptor, assessed by determining the extent of ligand binding using flow fluorocytometry. α-Tocopherol has been suggested to inhibit the raft-mediated effects of 7-ketocholesterol and the influence of this molecule on the effect of 7-ketocholesterol on the dipole potential are investigated as a potential mechanism for this inhibition. It is hypothesized that α-tocopherols may protect against the deleterious effects of cholesterol oxidation in cell membranes by excluding 7-ketocholesterol from specific microdomains, of which rafts are a subset, acting to preserve their dipole potential and maintain the function of the proteins they support. However, where significant cholesterol oxidation has previously occured the concurrent changes in the microdomain landscape of the membrane is suggested to prevent α-tocopherol succinate from eliciting this protective effect.

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