Residue Specific Partitioning Of KL_4 into Phospholipid Bilayers

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Introduction

KL_4, which has demonstrated success in treatment of respiratory distress, is a synthetic helical, amphipathic peptide mimetic of lung surfactant protein B. The unusual periodicity of charged residues within KL_4 and its relatively high hydrophobicity distinguish it from canonical amphipathic helical peptides.

Experimental

Here we utilized site specific spin labeling of both lipids and the peptides coupled with EPR spectroscopy to discern the effects of KL_4 on lipid dynamics, the residue specific dynamics of hydrophobic regions within KL_4, and the partitioning depths of specific KL_4 residues into the DPPC/POPG and POPC/POPG lipid bilayers under physiologically relevant conditions.

Results and Discussion

KL_4 induces alterations in acyl chain dynamics in a lipid-dependent manner, with the peptide partitioning more deeply into DPPC-rich bilayers. Combined with an earlier NMR study of changes in lipid dynamics on addition of KL_4 [1], we are able to distinguish how KL_4 affects both collective bilayer motions and intramolecular acyl chain dynamics in a lipid-dependent manner. EPR power saturation results for spin labeled lipids demonstrate that KL_4 also alters the accessibility profiles of paramagnetic colliders in a lipid-dependent manner.

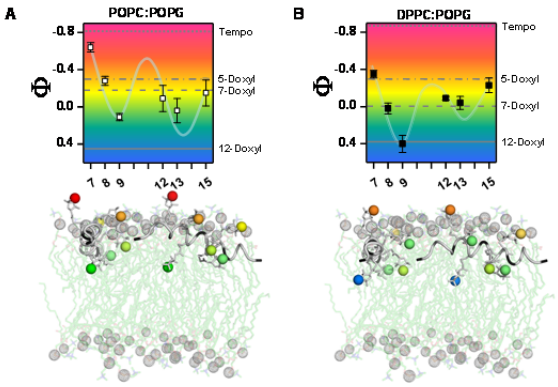

Figure 1. (Top) Φ values for KL_4-P1 variants partitioning into (A) POPC/POPG and (B) DPPC/POPC. Data were collected for a 50:1 L/P ratio. Lines indicate Φ values obtained for spin-labeled lipids (indicated by tags) at equivalent L/P ratios. (Bottom) Models of KL_4 partitioning into (C) POPC-rich and (D) DPPC-rich regions. Partitioning depths are based on Φ values obtained for the KL_4-P1 constructs. The oxygens of the IAP N-O moieties used in this study are color-coded with respect to relative partitioning depth. The structural models for KL_4 are based on NMR measurements in POPC/POPG [2] and lipid coordinates are from model DPPC simulations [3]. Error bars represent the standard deviations determined from samples measured in triplicate on differing days. Errors arise primarily from temperature fluctuations.

Conclusions

Measurements of dynamics and depth parameters for individual spin-labeled residues within KL_4 are consistent with a model where the peptide partitions deeply into the lipid bilayers but lies parallel to the bilayer interface in both lipid environments; the depth of partitioning is dependent on the degree of lipid acyl chain saturation within the bilayer.

Acknowledgements

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