The Pentration Depth of Surfactant Peptide KL4 into Membranes

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Lung surfactant protein B (SP-B) is an essential protein for lowering surface tension in the alveoli. The native form of SP-B is highly hydrophobic and contains 7 disulfide bridges which make pharmaceutical expression of the protein in large quantities problematic. Thus, synthetic replacements of SP-B have been highly sought after. KL4 (Sequence: $(KLLLL)_4K$) is a 21-residue peptide mimic of SP-B which has proven to clinically successful in relieving respiratory distress syndrome (RDS) when administered in a phospholipid dispersion. The focus of this research is on the biophysical properties of KL4 and their relation to alleviating RDS. Notably, KL4 retains many of the macroscopic properties of SP-B despite bearing little sequence similarity. Understanding how the comparatively simple KL4 is so successful will lead to a better understanding of SP-B and better synthetic analogues. [1]

Neutron diffraction experiments were conducted to determine the location of selectively deuterated amino acid residues on KL4 in a phospholipid membrane. Based on the depth of the residues a 3-D orientation of KL4 could be determined based on physical properties of the peptide (Fig. 1). Two lipid compositions, 4:1 DPPC:POPG and 4:1 POPC:POPG were studied. The first composition was chosen to mimic the formulation of lung surfactant studies. The second is a common composition for the study of membrane active peptides.

The L3 label in DPPC/POPG was unambiguously high in the bilayer, close to the choline headgroups at 24 Å from the bilayer center. L3 in POPC/POPG was more ambiguous. There was clearly label mass at the 24 Å level, as in DPPC/POPG, there was also significant mass at 14 Å from the bilayer center. The L12 label sits at ~17 Å from the bilayer center in both phospholipid mixtures. This is likely at the hydrophobic/hydrophilic interface, just below the phosphates and close to the glycerol esters. The significant difference in the L3 and L12 depths for DPPC/POPG fits the model of the labels being on the opposite sides of the helix, i.e. a classical α-helix, and argues against a tighter helix with greater hydrophobic moment. This is because in a much more tightly wound helix, to fit the data, it would abnormally tilt the peptide to a large oblique angle, whereas an α-helix lies parallel to the surface, such as shown Figure 1, constructed from the measured location of the L3 and L12 labels.

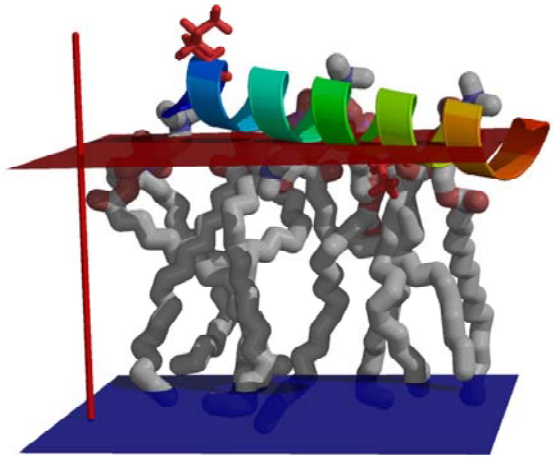

Fig. 1 Illustration of the location and orientation of the KL4 α-helix in the membrane, constructed from the measured location of the L3 and L12 labels.

Reference

[1] Antharam, V. C., Elliott, D. W., Mills, F. D., Farver, R. S., Sternin, E., and Long, J. R. (2008) The penetration depth of surfactant peptide KL4 into membranes in determined by fatty acid saturation. Submitted for publication.