SUPPLEMENTARY INFORMATION FOR "Transmembrane oligomeric intermediates of pore forming toxin Cytolysin A determine leakage kinetics"

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Figure 1: Typical SUV vesicle distribution obtained from dynamic light scattering experiments.



Figure 2: Determination of lipid concentration using the ammonium ferrothiocyanate method¹. A linear fit is used for the absorbance data vs lipid concentration. The absorbance of a suitably diluted sample at a lipid concentration of 0.05 mg/ml is illustrated by the blue data point corresponding to a lipid concentration of 1.3 mg/ml. Lipid concentrations in our experiments were found to lie in the range of 1.0 - 1.3 mg/ml.



Figure 3: The influence of oligomer fractions on the temporal evolution of the calcein leakage data shows that it necessary to include higher order oligomers or arcs to capture the leakage over the entire duration of the experiment. The smaller oligomers ranging from 5-7 mers accurately capture the initial leakage transient indicating that these fractions play a dominant role at this stage. Contribution from higher order oligomers are required to capture the evolution of the leakage curve at later times.

Oligomers	α	Tf	Proteins per vesicle (γ)
5-12	5.2	5.2	23
7-12	9.1	9.6	26
9-12	10.3	12.1	35
12	13.5	19.5	87

Table 1: The optimized parameters for the irreversible non-sequential oligomerization (IRNS) mechanism for different oligomer fractions that contribute to leakage.

Oligomers	ab	α	T _f	Proteins per vesicle (γ)
5-12	0.286	6.7	3.5	50
7-12	0.087	5.5	5.7	54
9-12	0.065	8.9	7.9	90
12	0.021	7.3	11.4	227

Table 2: The optimized parameters for the reversible non-sequential oligomerization (RNS) mechanism for different oligomer fractions that contribute to leakage.

Oligomers	α _c	T _f	Proteins per vesicle (γ)
5-12	7.0	5.5	65
7-12	0.38	14.2	266
9-12	0.075	14.6	379
12	0.04	13.7	416

Table 3: The optimized parameters for the irreversible sequential oligomerization (IRS) mechanism for different oligomer fractions that contribute to leakage.

References

1. Stewart, J. C. M. Colorimetric determination of phospholipids with ammonium ferrothiocyanate. Analytical biochemistry 104, 10–14 (1980).