

Supporting Information

for

EPR Study of Ampullosporin A, a Medium-Length Peptaibiotic, in Bicelles and Vesicles

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1. Analysis of the spectral dependence of the fitting of the EPR spectra of aligned bicelles on the angle Ψ

In Figure S1, we report the fitting of the spectra of **AmpT3** and **AmpT13** at different values of the angle Ψ from 0° to 30° (and from 90° to 100°): no significant difference can be seen between 0° and 10° , while the fitting is definitely worse at 30° .

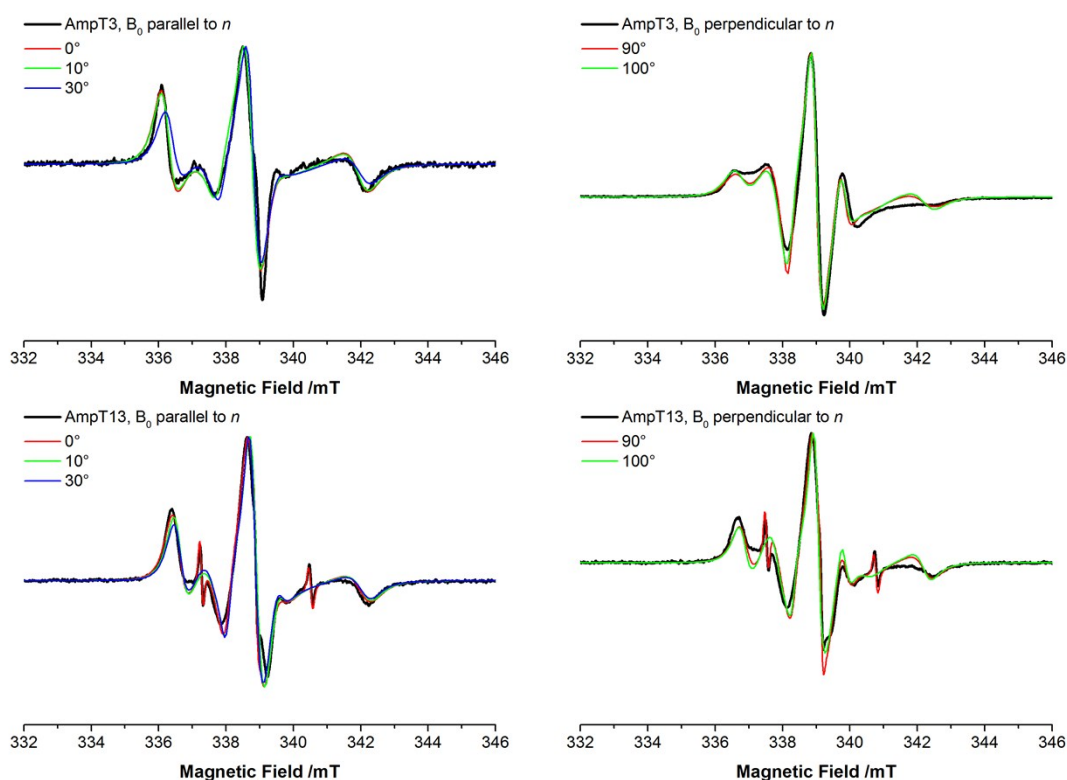


Figure S1

2. Detailed values of the P1/2 parameters from the saturation experiments

In Table S1 we report the values of the P1/2 obtained from the fitting of the individual saturation curves in separate experiments on **AmpT3** and **AmpT13**.

Table S1

Peptide	P1/2 _{O2}	P1/2 _{NiEdda}	P1/2 _{N2}	Φ P:L 1:100	P1/2 _{O2}	P1/2 _{NiEdda}	P1/2 _{N2}	Φ P:L 1:25
AmpT3	34.3	117.6	12.0	-1.6	19.4	14.9	13.0	1.2
	36.6	97.7	17.3	-1.4	38.7	19.5	9.3	1.1
AmpT13	67.0	36.8	12.6	0.8	83.5	18.5	6.5	1.9
	52.2	39.7	22.0	0.5	42.0	15.5	7.9	1.5

3. Calibration curve for the absolute membrane depth

The Φ parameter can be used to assess the distance from the membrane surface of a solute (R, in nm), provided a calibration curve is calculated. Below, in Figure S2, we show the linear regression plot: the curve has been calculated starting from the known depths of spin labeled phospholipids, considering the X-ray/NMR column from Table 1 in ref.[Dalton, L. A., McIntyre, J. O., and Fleischer, S. (1987), *Biochemistry* 26, 2117-2130].

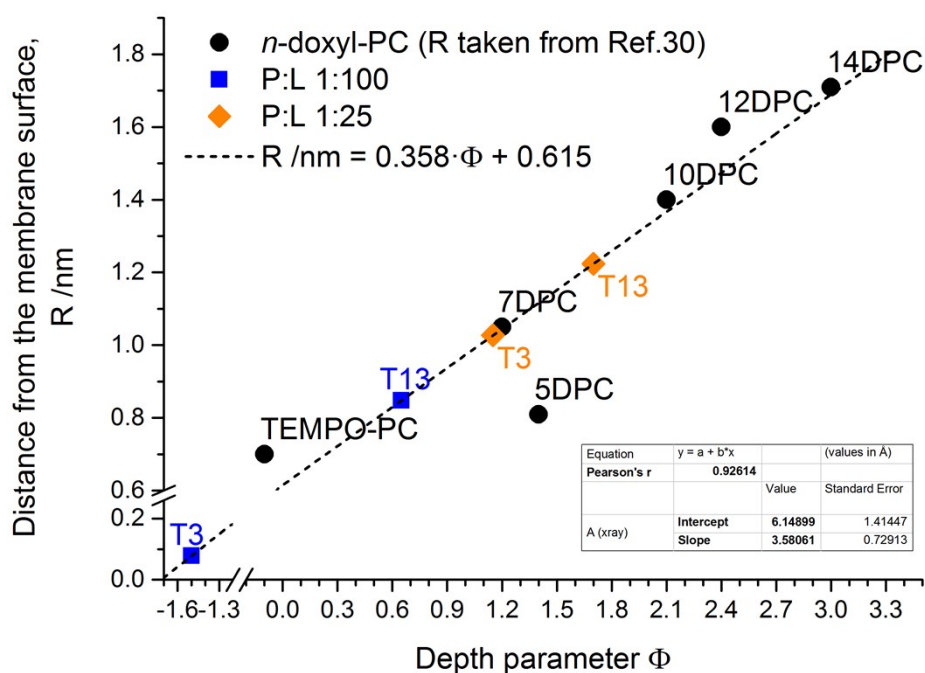


Figure S2

4. Spectra of bicelles labeled with 5DPC interacting with Amp

In Figure S3, we report the EPR spectra of oriented bicelles labeled with 5DPC and interacting with **Amp** (unlabeled) at a 1:100 P:L ratio. The spectra have been prepared and have been aligned in the same way as those of unlabelled bicelles interacting with **AmpT3** and **AmpT13**. As can be seen from the Figure, the spectra taken with the magnetic field parallel or perpendicular to the membrane normal n are very different from each other, confirming the very good alignment of the samples.

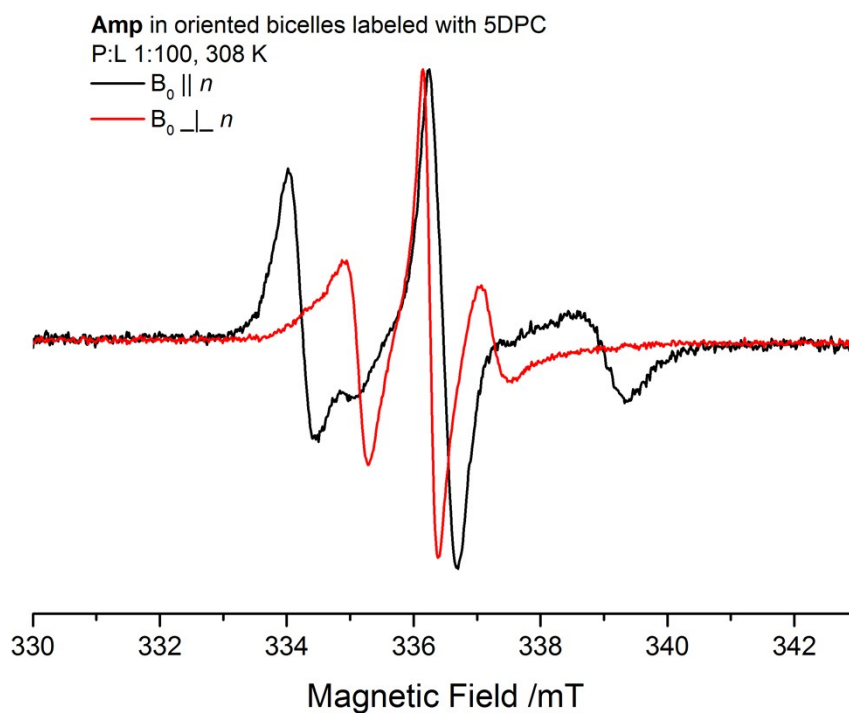


Figure S3