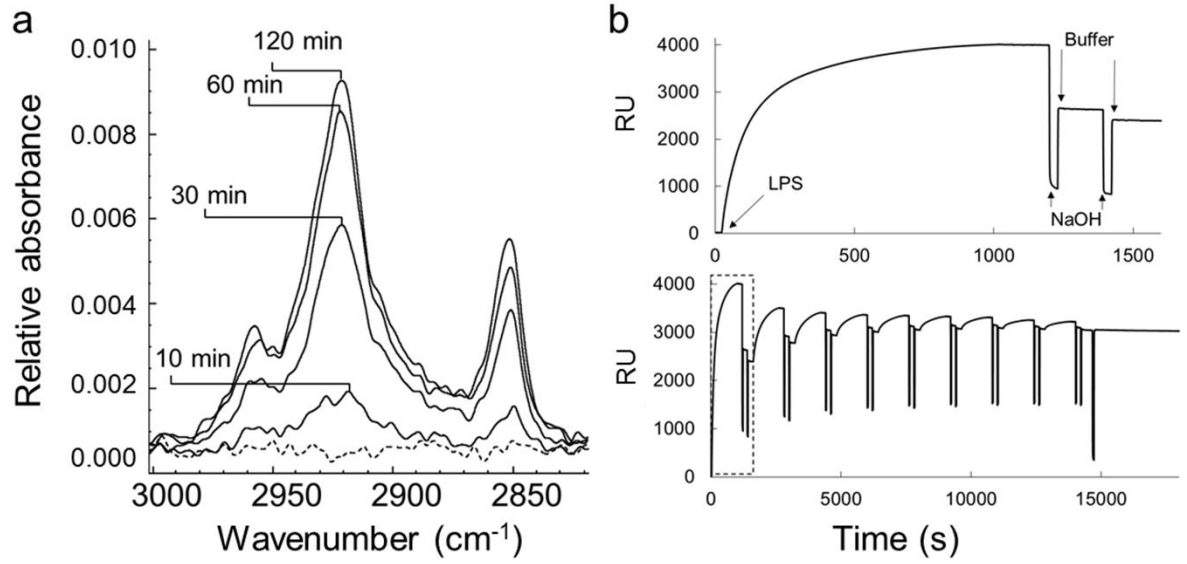
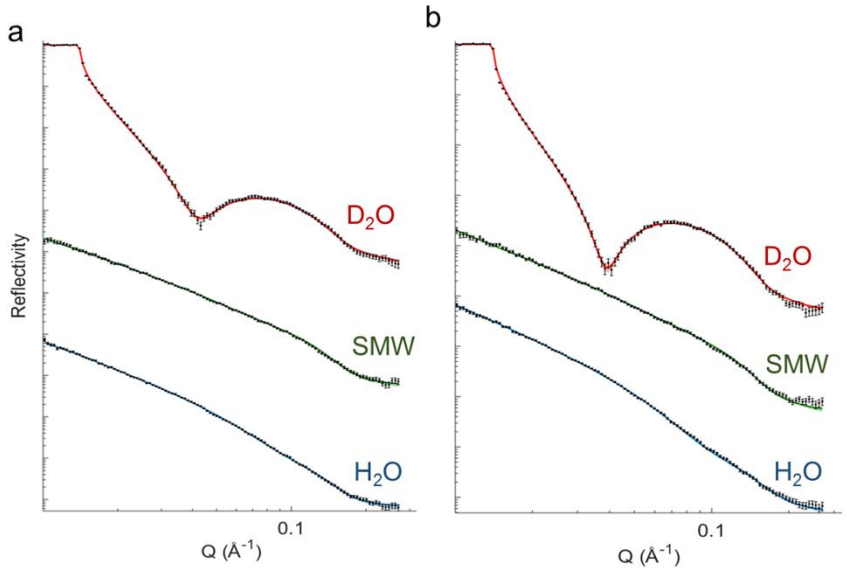
SUPPLEMENTARY FIGURES:



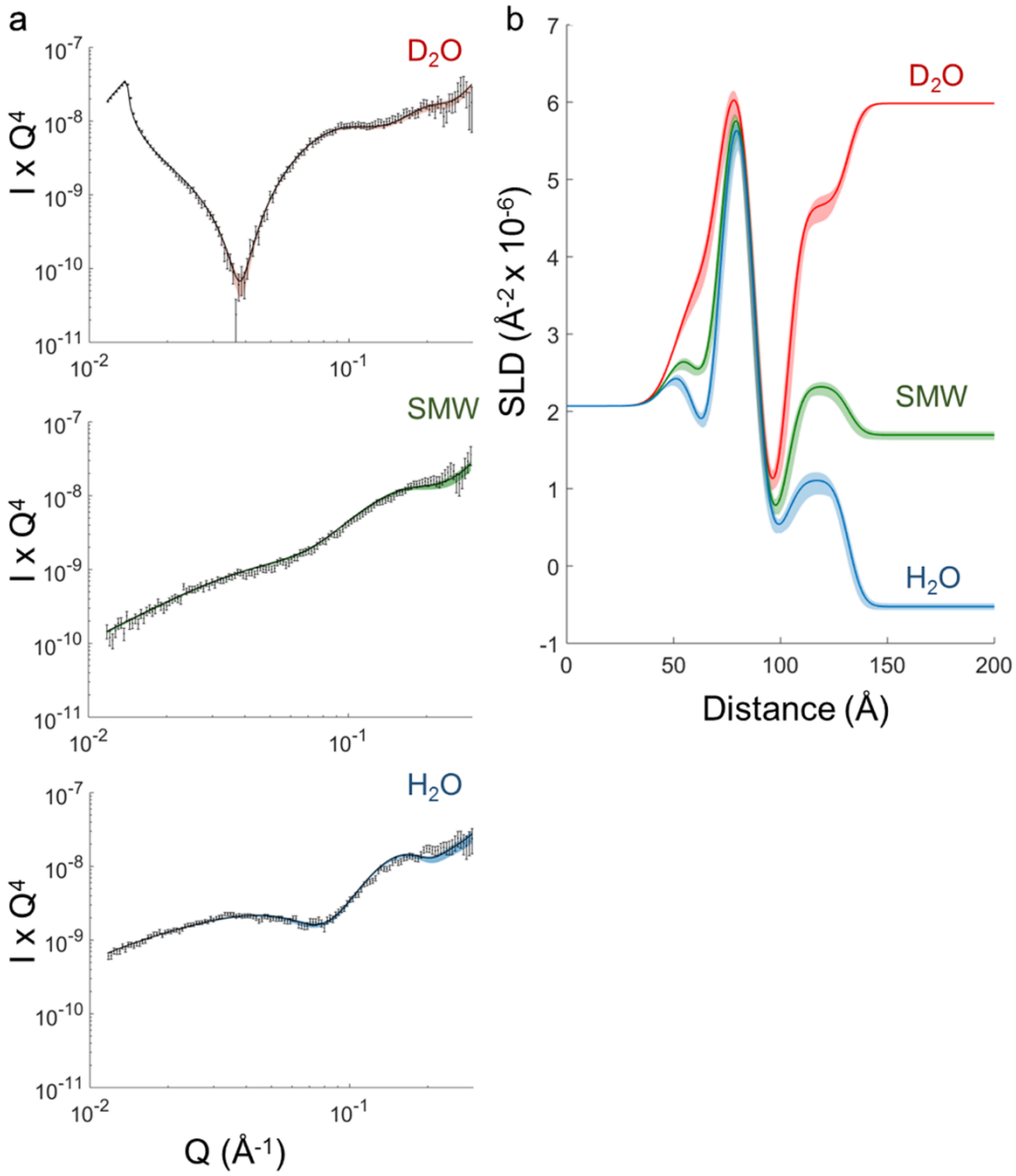
Supplementary Figure 1 | Formation of $_{LPS}$HBM monitored by ATR-FTIR and SPR

(**a**) ATR-FTIR absorption spectra of LPS deposited on an OTS-coated silicon crystal. The three main peaks above the background signal (dashed line, T=0) record the time dependent adsorption of LPS on the hydrophobic surface and arise from the CH bond stretching vibrations. From the left: asymmetric CH_3 (~2957 cm^{-1}), asymmetric CH_2 (~2920 cm^{-1}) and symmetric CH_2 (~2850 cm^{-1}) vibrations. (**b**) SPR trace of LPS deposition on a thioalkane coated gold surface. The relative response units (RU) record the amount of material adsorbed on the surface. Arrows show the injection starting points of different components by the microfluidic system. Due to instrumental limitations, a single prolonged LPS incubation was not possible and sequential lipid injections were performed to achieve surface saturation by the monolayer (bottom).



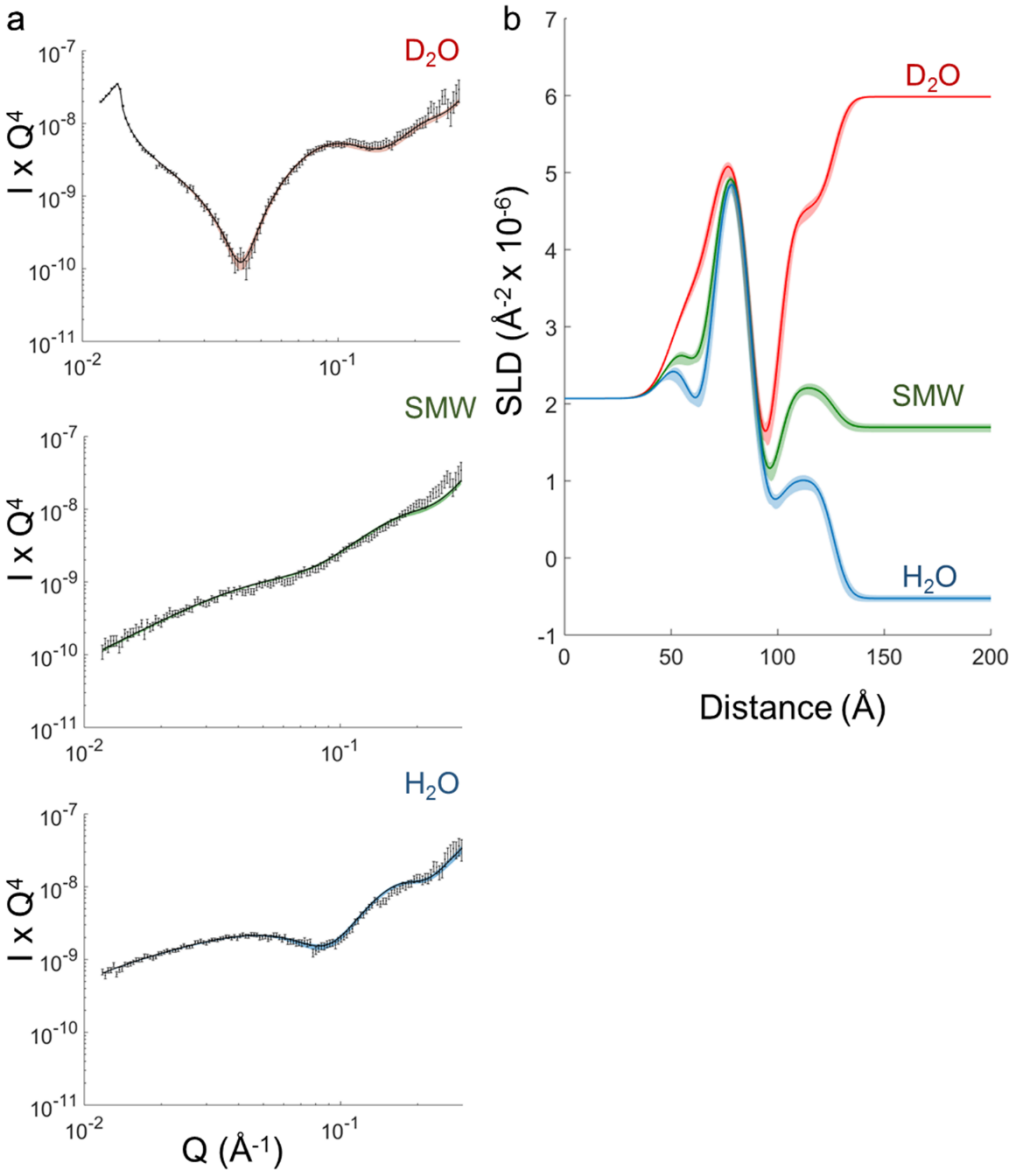
Supplementary Figure 2 | NR characterisation of $_{LPS}$HBM before and after PmB addition

Neutron reflectivity data points and simultaneous best fit lines for a $_{LPS}$HMB formed on deuterated OTS before (**a**) and after (**b**) the addition of PmB measured at three solution contrasts: D_2O (red line), silicon matched water (SMW, green line) and H_2O (blue line), data sets are offset vertically for clarity. The datasets collected in D_2O are shown in **Fig 2 c**. Here the reflected intensity is displayed unmodified to show its intrinsic decay proportional to Q^4. All the rest of reflectivity data are displayed in the form I x Q^4 to highlight the features in the curves. The reduced chi-squared values relative to the fits are 2.73 (D_2O), 0.96 (SMW), 1.26 (H_2O) in **a** and 3.81 (D_2O), 1.14 (SMW) 1.15 (H_2O) in **b**



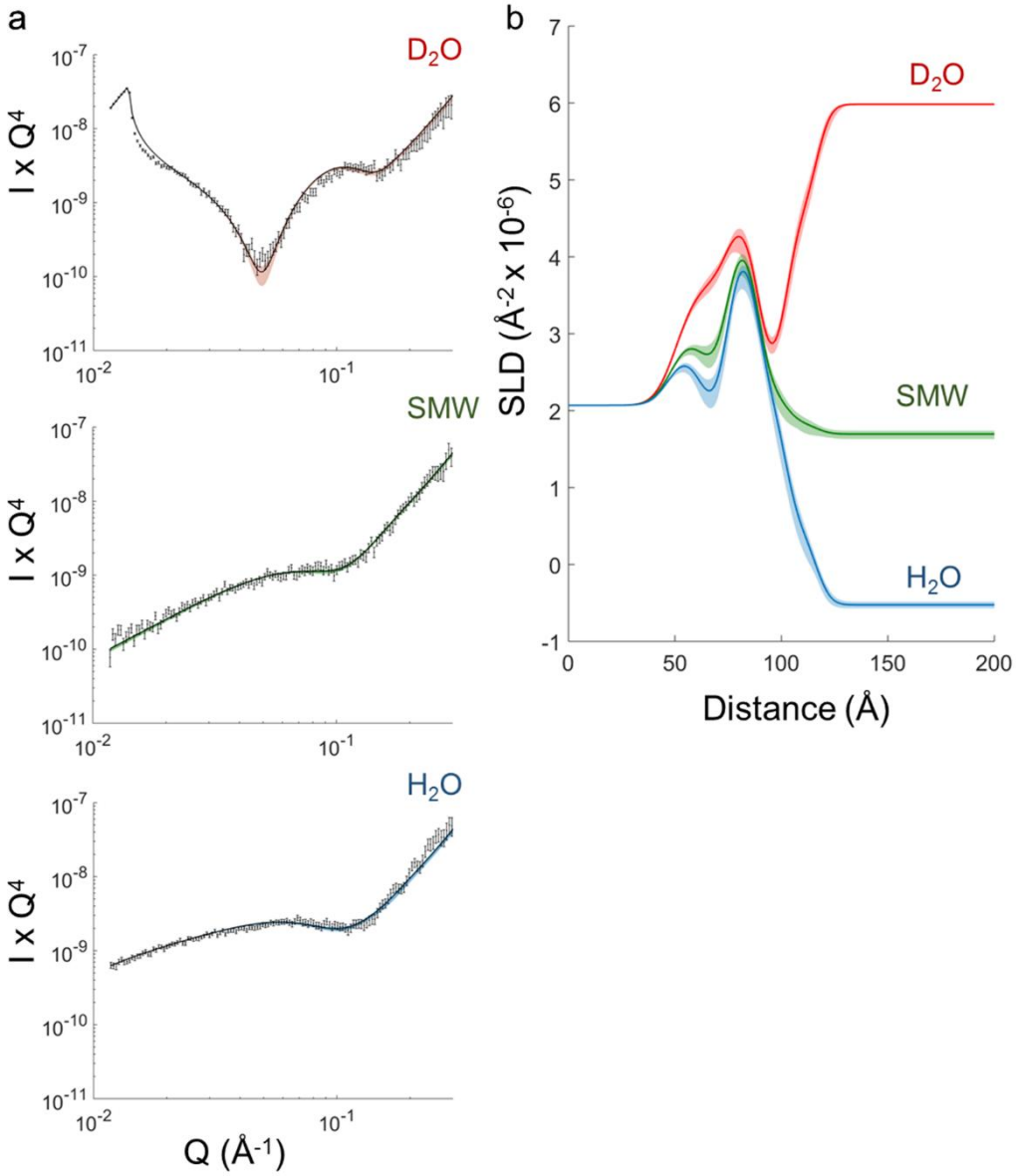
Supplementary Figure 3 | NR characterisation of $_{dPL/LPS}$OMM at room temperature before heating to 37°C

(**a**) Neutron reflectivity data points and simultaneous best fit lines for a $_{dPL/LPS}$OMM measured at room temperature at three solution contrasts: D_2O (red line), silicon matched water (SMW, green line) and H_2O (blue line). The dataset collected in D_2O is shown in **Fig. 3d** in black (**b**) SLD profile derived from the simultaneous fit of the data shown in **a.** The SLD profiles are also shown in **Fig. 3e** in black. Shading around fit and SLD lines represent 95% confidence intervals. The reduced chi-squared values relative to the fits are 3.42 (D_2O), 1.22 (SMW) and 1.71 (H_2O)



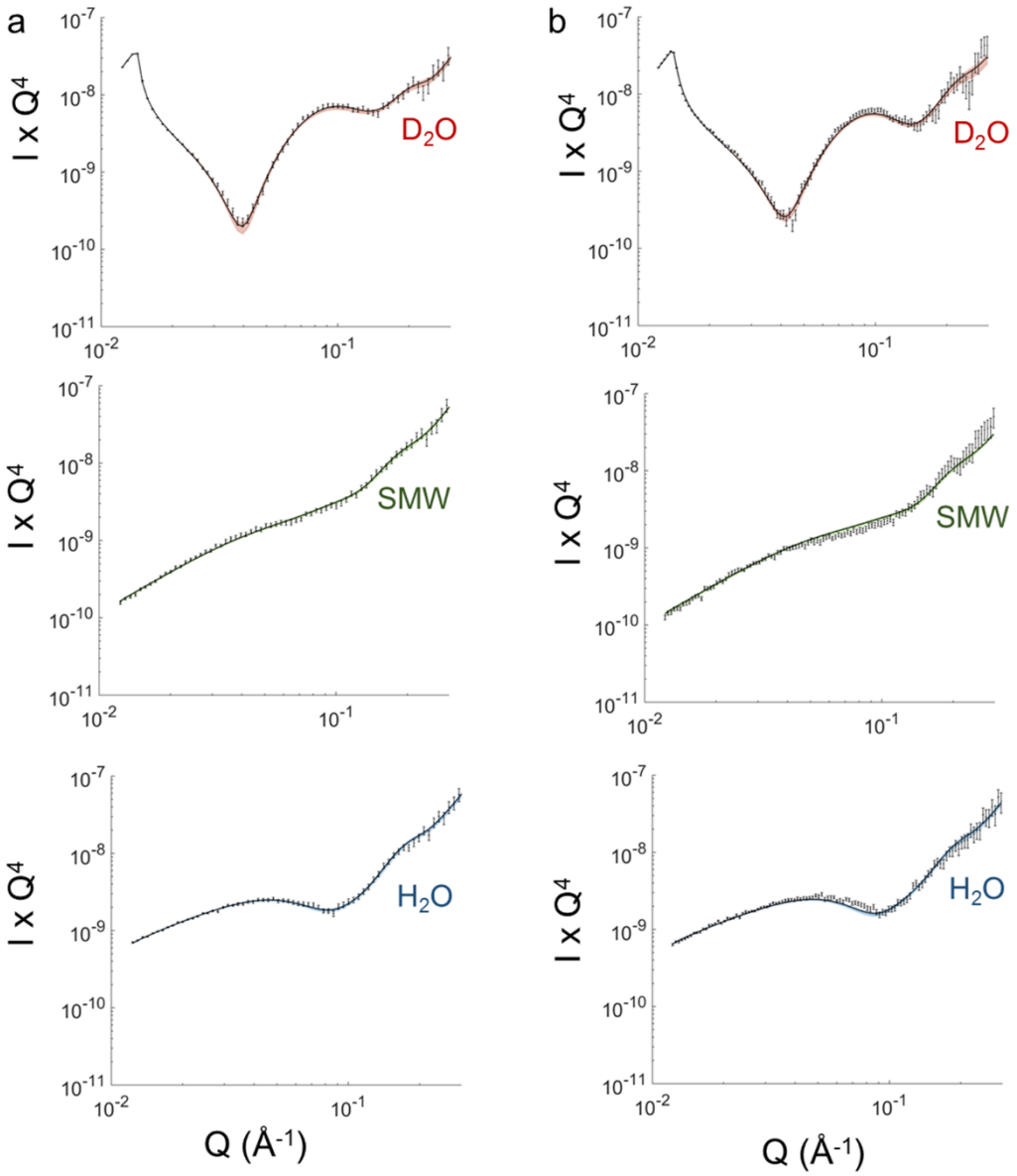
Supplementary Figure 4 | NR characterisation of $_{dPL/LPS}$OMM at 37°C before PmB addition. This is the same model shown in supplementary Figure 3 and Table 3.

(**a**) Neutron reflectivity data points and simultaneous best fit lines for a $_{dPL/LPS}$OMM measured at 37°C at three solution contrasts: D_2O (red line), silicon matched water (SMW, green line) and H_2O (blue line). SLD profile derived from the simultaneous fit of the data shown in **a.** The SLD profiles are also shown in **Fig. 3e** and **f** in red. Shading around fit and SLD lines represent 95% confidence intervals. The reduced chi-squared values relative to the fits are 1.31 (D_2O), 1.63 (SMW) and 1.25 (H_2O)



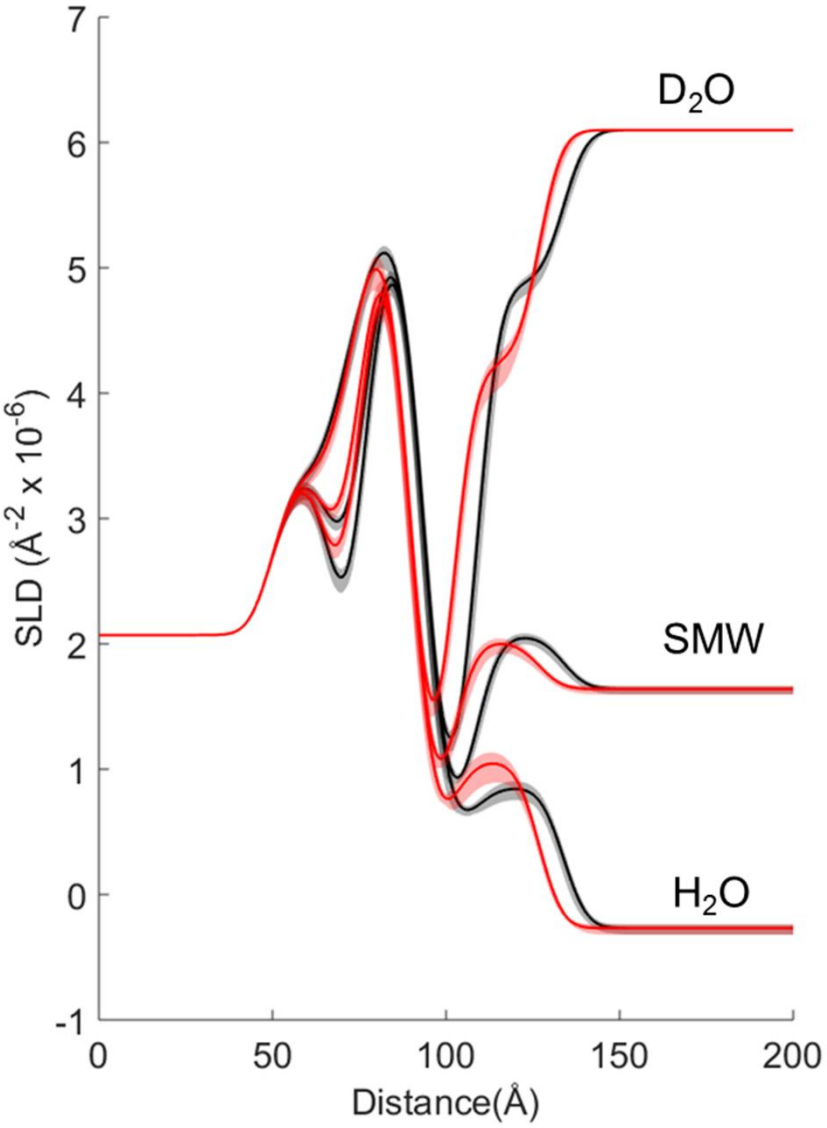
Supplementary Figure 5 | NR characterisation of $_{dPL/LPS}$OMM at 37°C after PmB addition. This is the same model shown in supplementary Figures 3&4 and Tables 3&4.

(**a**) Neutron reflectivity data points and simultaneous best fit lines for a $_{dPL/LPS}$OMM measured at 37°C in the presence of 100µg/ml PmB at three solution contrasts: D_2O (red line), silicon matched water (SMW, green line) and H_2O (blue line). SLD profile derived from the simultaneous fit of the data shown in **a.** The SLD profiles are also shown in **Fig. 3f** in blue. Shading around fit and SLD lines represent 95% confidence intervals. The reduced chi-squared values relative to the fits are 11.39 (D_2O), 1.19 (SMW) and 1.95 (H_2O)



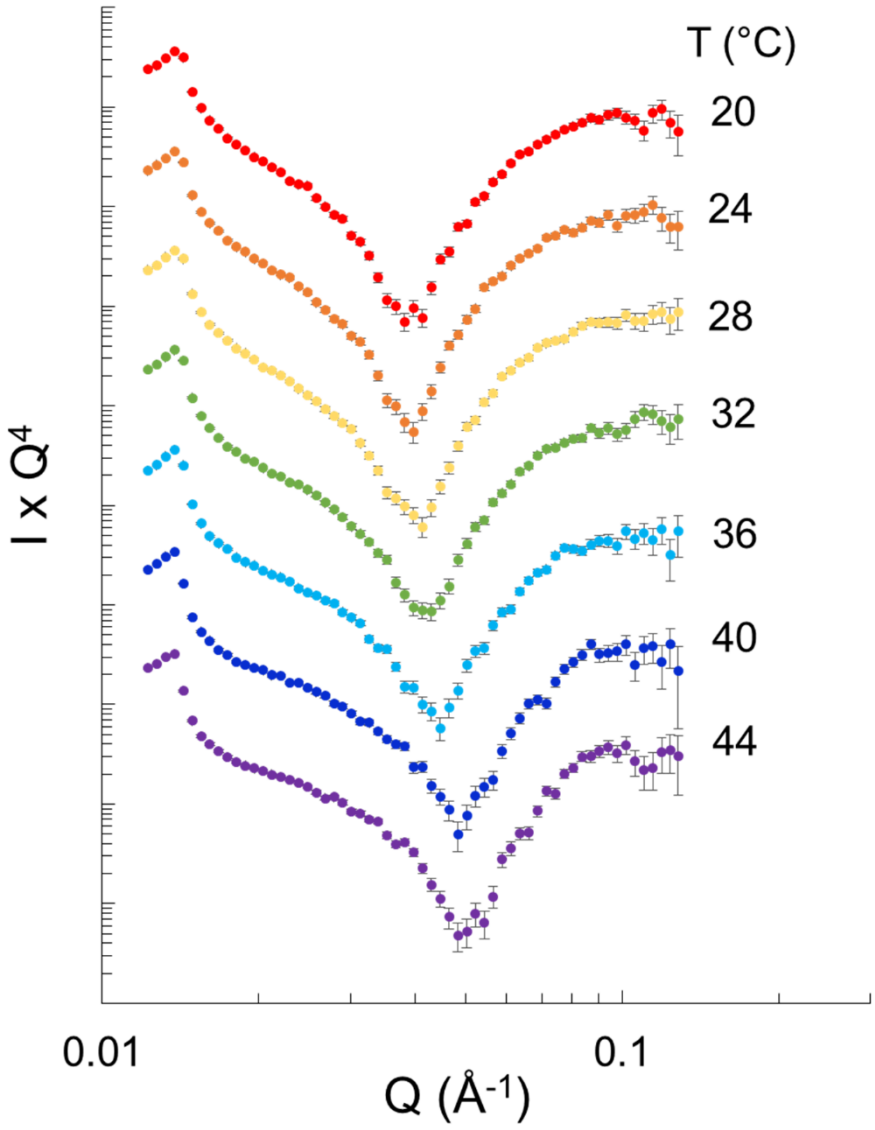
Supplementary Figure 6 | NR characterisation of $_{dPL/LPS}$OMM at room temperature and at 37°C in the presence of Polymyxin B nonapeptide

Neutron reflectivity data points and simultaneous best fit lines for a $_{dPL/LPS}$OMM measured at (**a**) room temperature and (**b**) at 37°C in the presence of polymyxin B nonapeptide (PmBN) at three solution contrasts: D_2O (red line), silicon matched water (SMW, green line) and H_2O (blue line). The corresponding SLD profiles derived from the fits are shown in Supplementary Fig. 7. The reduced chi-squared values relative to the fits are 3.17 (D_2O), 1.25 (SMW) 0.94 (H_2O) in **a** and 6.31 (D_2O), 2.56 (SMW) and 2.18 (H_2O) in **b**



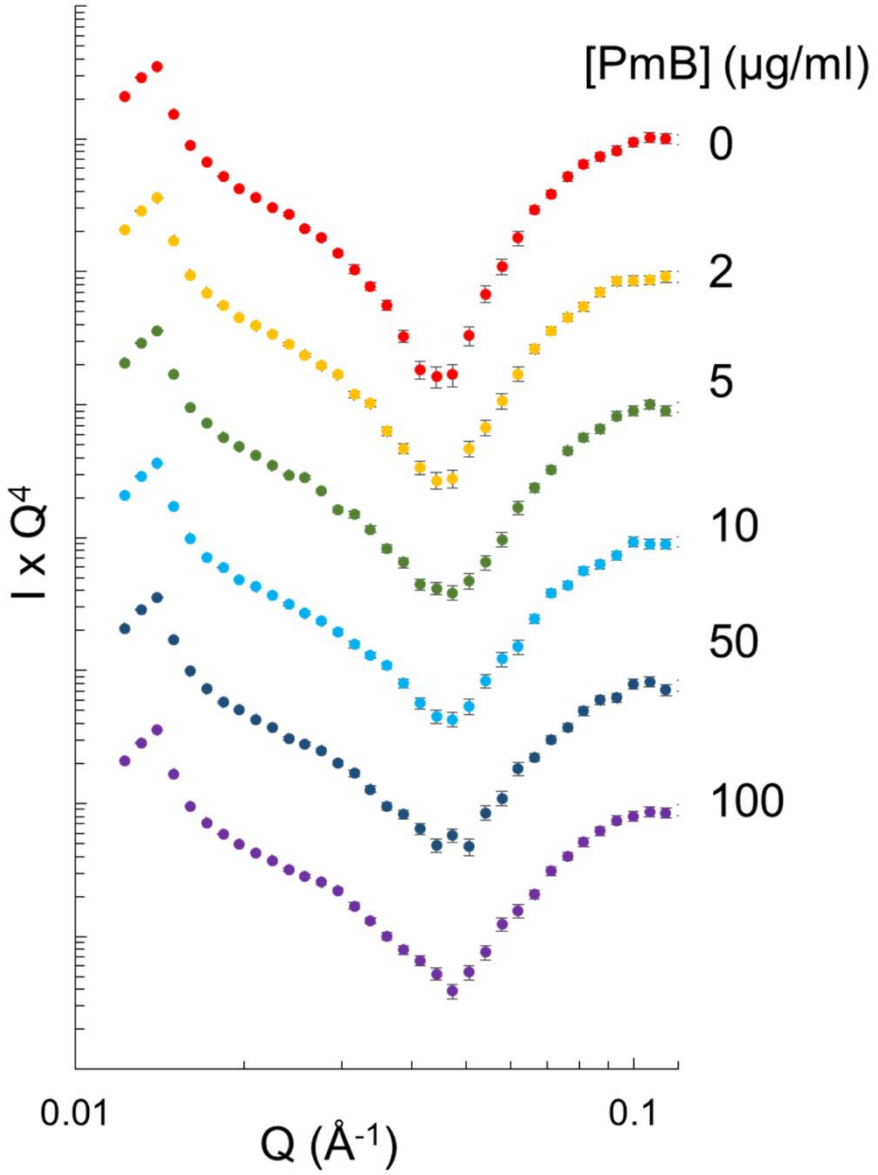
Supplementary Figure 7 | NR characterisation of $_{dPL/LPS}$OMM in the presence of PmBN

SLD profile (lines) and corresponding 95% confidence intervals (shading) derived from the fits shown in Supplementary Fig 6 of a $_{dPL/LPS}$OMM at room temperature (black) and at 37°C in the presence of Polymyxin B nonapeptide (PmBN) (red). Contrary to what was observed for PmB, PmBN did not cause any evident mixing between the hydrogenous and deuterated leaflets of the membrane above the phase transition temperature of the bilayer. At 37°C the SLD profile of the bilayer shows the thinning caused by the lipid phase transition also observed in **Fig. 3e**



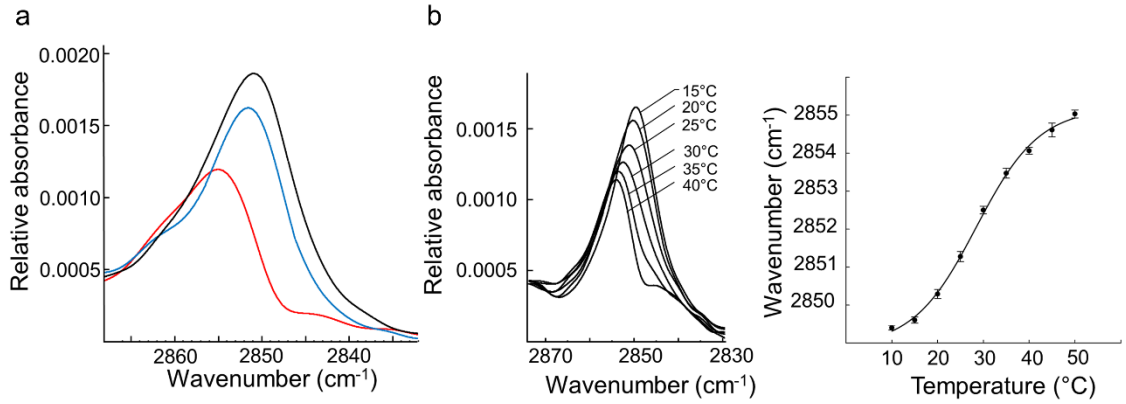
Supplementary Figure 8 | Effect of temperature on $_{dPL/LPS}$OMM disruption by PmB

Full neutron reflectivity data sets collected in D_2O during the temperature ramp in the presence of PmB shown in **Fig. 3a.** Datasets are offset vertically for clarity



Supplementary Figure 9 | PmB titration at 37°C

Neutron reflectivity data sets collected in D_2O during the addition of increasing amounts of PmB at 37°C. Datasets are offset vertically for clarity. The barycentric mean of the Kiessig fringe of each data set is plotted in **Fig. 4c.**



Supplementary Figure 10 | Reversibility of the phase transition and phase behaviour of $_{LPS}$HBM

(**a**) Absorption of the LPS H-C-H symmetric stretching vibration of the $_{dPL/LPS}$OMM at room temperature before the heating ramp (black line) at 45C (red line) and after cooling at room temperature (blue line). (**b**) Peak absorption of the LPS H-C-H symmetric stretching vibration measured between 15°C and 40°C in the $_{LPS}$HBM (left) and plot of the peak absorption values as a function of temperature (right).

SUPPLEMENTARY TABLES:

Supplementary Table 1 | Fitting parameters obtained before and after PmB addition to the $_{LPS}$HBM. Parameters obtained from the fit of the data shown in Supplementary Fig. 2. In brackets the 95% confidence intervals obtained from the error analysis.

LPS Parameters	**Fitted value**	**Priors**
LPS tails thickness (Å)	14.2 (13.5, 14.8)	Uniform (min=8, max=20)
LPS tails SLD ($Å^{-2}$ x 10^{-6})	0.19 (0.07 0.33)	Uniform (min=-0.4x10^{-6}, max=3x10^{-6})
LPS tails coverage (%)	92.3 (87.8, 94.8)	Uniform (min=0, max=100)
LPS core thickness (Å)	20.9 (19.1, 22.6)	Uniform (min=10, max=40)
LPS core SLD in D2O ($Å^{-2}$ x 10^{-6})	5.4 (5.3, 5.5)	Uniform (min=2x10^{-6}, max=7x10^{-6})
LPS core hydration (%)	68.5 (64.8, 72.3)	Uniform (min=0, max=100)
LPS + PmB Parameters		
LPS tails thickness (Å)	18.0 (16.8, 19.3)	Uniform (min=8, max=20)
LPS tails SLD ($Å^{-2}$ x 10^{-6})	0.30 (0.20 0.47)	Uniform (min=-0.4x10^{-6}, max=3x10^{-6})
LPS tails coverage (%)	90.0 (86.3, 93.7)	Uniform (min=0, max=100)
LPS core thickness (Å)	12.9 (10.9, 14.6)	Uniform (min=10, max=40)
LPS core SLD in D2O ($Å^{-2}$ x 10^{-6})	5.6 (5.2, 5.9)	Uniform (min=2x10^{-6}, max=7x10^{-6})
LPS core hydration (%)	40.9 (34.1, 49.1)	Uniform (min=0, max=100)
Substrate Parameters		
Substrate roughness (Å)	5.4 (4.8, 6.2)	Uniform (min=0, max=12)
Silicon oxide thickness (Å)	14.2 (13.2, 15.1)	Uniform (min=0, max=40)
Silicon oxide SLD ($Å^{-2}$ x 10^{-6})	3.46 (3.38, 3.5)	Uniform (min=3.3x10^{-6}, max=3.5x10^{-6})
Silicon oxide hydration (%)	9.7 (6.5, 13.0)	Uniform (min=0, max=100)
OTS thickness (Å)	26.4 (25.0, 27.0)	Uniform (min=15, max=40)
OTS SLD ($Å^{-2}$ x 10^{-6})	6.6 (6.4, 6.9)	Uniform (min=4x10^{-6}, max=8x10^{-6})
OTS coverage	95.1 (94.2, 96.3)	Uniform (min=0, max=100)

Supplementary Table 2 | Fitting parameters obtained for $_{dPL/LPS}$OMM at room temperature. Parameters obtained from the fit to the data shown in Fig. 3a. In brackets the 95% confidence intervals obtained from the error analysis. This model was used for the addition of PmB at room temperature see Fig 3c.

Bilayer Parameters	**Fitted value**	**Priors**
dDPPC head groups thickness (Å)	8.8 (7.5, 10.1)	Uniform (min=0, max=15)
dDPPC head groups SLD ($Å^{-2}$ x 10^{-6})	1.7 (1.3, 2.3)	Uniform (min=0.5x10^{-6}, max=2.5x10^{-6})
dDPPC head groups hydration (%)	50.6 (44.7, 55.7)	Uniform (min=0, max=100)
dDPPC tails thickness (Å)	17.6 (16.5, 18.6)	Uniform (min=12, max=27)
dDPPC tails SLD ($Å^{-2}$*10^{-6})	6.6 (6.3, 6.9)	Uniform (min=4x10^{-6}, max=7.6x10^{-6})
LPS tails thickness (Å)	16.6 (16.3, 17.0)	Uniform (min=12, max=20)
LPS tails SLD ($Å^{-2}$ x 10^{-6})	0.01 (-0.01 0.02)	Uniform (min=-0.4x10^{-6}, max=3x10^{-6})
LPS core thickness (Å)	29.0 (28.4, 29.6)	Uniform (min=20, max=40)
LPS core SLD in D2O ($Å^{-2}$ x 10^{-6})	4.5 (4.3, 4.8)	Uniform (min=2x10^{-6}, max=7x10^{-6})
Core hydration (%)	42.5 (35.9, 50.1)	Uniform (min=0, max=100)
Bilayer coverage (%)	99.2 (97.8, 100)	Uniform (min=0, max=100)
Substrate Parameters	**Fitted value**	**Priors**
Substrate roughness (Å)	3.2 (2.6, 3.8)	Uniform (min=0, max=12)
Silicon oxide thickness (Å)	16.4 (14.7, 18.3)	Uniform (min=0, max=40)
Silicon oxide SLD ($Å^{-2}$ x 10^{-6})	3.35 (3.30, 3.43)	Uniform (min=3.3x10^{-6}, max=3.5x10^{-6})
Silicon oxide hydration (%)	6.6 (3.8, 9.2)	Uniform (min=0, max=100)

Supplementary Table 3 | Fitting parameters obtained for $_{dPL/LPS}$OMM at room temperature. Parameters obtained from the fit of the data shown in Supplementary Fig. 3. In brackets the 95% confidence intervals obtained from the error analysis.

Bilayer Parameters	**Fitted value**	**Priors**
dDPPC head groups thickness (Å)	10.2 (9.7, 10.6)	Uniform (min=0, max=15)
dDPPC head groups SLD ($Å^{-2}$ x 10^{-6})	1.8 (1.7, 2.0)	Uniform (min=0.5x10^{-6}, max=2.5x10^{-6})
dDPPC head groups hydration (%)	52.7 (47.8, 57.3)	Uniform (min=0, max=100)
dDPPC tails thickness (Å)	16.3 (15.4, 17.1)	Uniform (min=12, max=27)
dDPPC tails SLD ($Å^{-2}$*10^{-6})	6.5 (6.3, 6.8)	Uniform (min=4x10^{-6}, max=7.6x10^{-6})
LPS tails thickness (Å)	15.7 (15.2, 16.2)	Uniform (min=12, max=20)
LPS tails SLD ($Å^{-2}$ x 10^{-6})	0.28 (0.25 0.30)	Uniform (min=-0.4x10^{-6}, max=3x10^{-6})
LPS core thickness (Å)	28.3 (27.1, 29.4)	Uniform (min=20, max=40)
LPS core SLD in D2O ($Å^{-2}$ x 10^{-6})	4.0 (3.9, 4.3)	Uniform (min=2x10^{-6}, max=7x10^{-6})
Core hydration (%)	35.4 (33.0, 37.6)	Uniform (min=0, max=100)
Bilayer coverage (%)	97.6 (94.8, 99.7)	Uniform (min=0, max=100)
Bilayer roughness (Å)	5.3 (5.0, 5.6)	Uniform (min=0, max=10)
Substrate Parameters	**Fitted value**	**Priors**
Substrate roughness (Å)	7.7 (7.1, 8.3)	Uniform (min=0, max=12)
Silicon oxide thickness (Å)	11.3 (10.5, 11.1)	Uniform (min=0, max=40)
Silicon oxide SLD ($Å^{-2}$ x 10^{-6})	3.33 (3.30, 3.38)	Uniform (min=3.3x10^{-6}, max=3.5x10^{-6})
Silicon oxide hydration (%)	5.3 (4.7, 5.9)	Uniform (min=0, max=100)

Supplementary Table 4 | Fitting parameters obtained for $_{dPL/LPS}$OMM at 37°C before PmB addition. Parameters obtained from the fit of the data shown in Supplementary Fig. 4. In brackets the 95% confidence intervals obtained from the error analysis. This is the same model shown in supplementary Figure 3 and Table 3.

Bilayer Parameters	Fitted value	Priors
dDPPC head groups thickness (Å)	8.1 (7.5, 8.7)	Uniform (min=0, max=15)
dDPPC head groups SLD ($Å^{-2}*10^{-6}$)	1.6 (1.4, 1.8)	Uniform (min=$0.5x10^{-6}$, max=$2.5x10^{-6}$)
dDPPC head groups hydration (%)	50.2 (47.0, 53.8)	Uniform (min=0, max=100)
dDPPC tails thickness (Å)	18.2 (17.4, 19.1)	Uniform (min=12, max=27)
dDPPC tails SLD ($Å^{-2}*10^{-6}$)	5.3 (5.2, 5.5)	Uniform (min=$4x10^{-6}$, max=$8x10^{-6}$)
LPS tails thickness (Å)	12.2 (11.8, 12.6)	Uniform (min=8, max=20)
LPS tails SLD ($Å^{-2}*10^{-6}$)	0.4 (0.37, 0.43)	Uniform (min=$-0.4x10^{-6}$, max=$3x10^{-6}$)
LPS core thickness (Å)	26.4 (25.4, 27.2)	Uniform (min=20, max=40)
LPS core SLD in D2O ($Å^{-2}*10^{-6}$)	3.8 (3.6, 3.9)	Uniform (min=$2x10^{-6}$, max=$7x10^{-6}$)
LPS core hydration (%)	36.9 (34.1, 39.4)	Uniform (min=0, max=100)
Bilayer coverage (%)	99.0 (97.2, 100)	Uniform (min=0, max=100)
Bilayer roughness (Å)	5.5 (5.2, 5.8)	Uniform (min=0, max=10)

Supplementary Table 5 | Fitting parameters obtained for $_{dPL/LPS}$OMM at 37°C after PmB addition. Parameters obtained from the fit of the data shown in Supplementary Fig. 5. In brackets the 95% confidence intervals obtained from the error analysis. This is the same model shown in supplementary Figures 3&4 and Tables 3&4. The layers have been labelled inner and outer leaflets as the components are now mixed and contain a fraction of PmB

Bilayer Parameters	**Fitted value**	**Priors**
Inner leaflet head groups thickness (Å)	13.4 (12.3, 13.8)	Uniform (min=0, max=30)
Inner leaflet head groups SLD ($Å^{-2}*10^{-6}$)	2.8 (2.6, 3)	Uniform (min=$0.5x10^{-6}$, max=$4x10^{-6}$)
Inner leaflet head groups hydration (%)	29.6 (27.3, 32.1)	Uniform (min=0, max=100)
Inner leaflet tails thickness (Å)	14.7 (13.5, 16.1)	Uniform (min=8, max=27)
Inner leaflet tails SLD ($Å^{-2}*10^{-6}$)	4.4 (4.2, 4.6)	Uniform (min=$3x10^{-6}$, max=$8x10^{-6}$)
Outer leaflet tails thickness (Å)	13.1 (12.0, 14.0)	Uniform (min=8, max=25)
Outer leaflet tails SLD ($Å^{-2}*10^{-6}$)	2.3 (2.0, 2.4)	Uniform (min=$-0.4x10^{-6}$, max=$7x10^{-6}$)
Outer leaflet head groups thickness (Å)	14.3 (13.1, 15.5)	Uniform (min=10, max=50)
Outer leaflet head groups SLD in D2O ($Å^{-2}*10^{-6}$)	2.3 (2.2, 2.5)	Uniform (min=$2x10^{-6}$, max=$7x10^{-6}$)
Outer leaflet head groups hydration (%)	60.9 (54.4, 70.1)	Uniform (min=0, max=100)
Bilayer coverage (%)	95.5 (93.6, 97.5)	Uniform (min=0, max=100)
Bilayer roughness (Å)	5.3 (4.8, 5.7)	Uniform (min=0, max=10)