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Hydroxide-Ion Binding to Nonionic Interfaces in Aqueous Solution

Supporting information

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S1. Experimental

Materials. Gemini surfactants were synthesised as will described in a forthcoming paper.¹ Hepes, Mes (4-morpholinoethanesulfonic acid), APS (3-amino-1-propanesulfonic acid) and taurine (2-aminoethanesulfonic acid) were purchased from Sigma and used as received.

Sample Preparation. Solutions of gemini surfactant in chloroform were dried under a stream of nitrogen. Traces of residual solvent were removed under high vacuum. To obtain small unilamellar vesicles (SUVs) with a narrow and reproducible size distribution, the lipid films were hydrated at room temperature in bidistilled water containing 5 mM each of the buffer substances Hepes, Mes and taurine and 10 mM of APS at a pH close to the pH of phase separation of the gemini surfactant (unless stated otherwise), vortexed for several minutes, briefly tip sonicated (<30 sec), freeze-thawed [$N_2(l) \leftrightarrow$ water bath (40 °C)] 5 times, and extruded 11 times through a 200-nm-pore-size polycarbonate filter.

The samples for light scattering measurements were prepared by diluting the 5 mM (total lipid concentration) vesicular stock solutions to a final concentration of 0.25 mM and adding the appropriate amount of HCl (aq) or NaOH (aq) to the desired pH. The dispersions were allowed to equilibrate overnight before measuring.

Static and Dynamic Light Scattering. Static and dynamic light scattering measurements were performed at 25 °C on a Zetasizer 5000 instrument (Malvern Instruments, U.K.) at $\lambda=633$ nm. To obtain the hydrodynamic radii, the intensity autocorrelation functions were analyzed using CONTIN.

Cryo-Transmission Electron Microscopy. A drop of the lipid suspension was deposited on a glow discharged holey carbon-coated grid. After blotting away the excess of lipid, the grids were rapidly plunged into liquid ethane. The frozen specimen were mounted in a Gatan (model 626) cryo-stage and examined in a Philips CM 120 cryo-electron microscope operating at 120 kV. Micrographs were recorded under low-dose conditions. Cryo-electron microscopy pictures of **2** were taken from grids vitrified in a controlled environmental chamber at 100% humidity.

Turbidity. Turbidity was measured by taking the absorbance at 450 nm of a 0.25 mM solution of **2**. The samples were equilibrated overnight at 35°C. Then the temperature was lowered in steps of a few degrees and the turbidity was taken when the absorbance reached a stable level. The turbidity data was plotted against the

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temperature and fitted to a sigmoidal equation leading to the phase transition temperature and the width of the transition.

S2. Static and Dynamic Light Scattering Results

Figure 1. Relative scattered intensity (A) and size distributions (B) of solutions containing aggregates formed from compound **2** (x), **3** (□) and **4** (●) as a function of pH. Error bars denote the width of the size distribution.² In A the data for **3** and **4** has been normalised with respect to each other at pH 6.6 and pH 7.3, respectively.

For the dynamic and static light scattering experiments the 5 mM stock solution of **3** was prepared at pH 3.4 and therefore not freeze-thawed and extruded.

S3. Cryo-electron microscopy

Figure 2. Cryo-electron microscopy pictures **2** equilibrated over night at pH 7.4. Samples were vitrified at 10°C (A) and 25°C (B). Bar represents 100 nm.

Figure 3. Cryo-electron microscopy pictures of **4** equilibrated over night at pH 11.1. Bar represents 100 nm.

S4. Turbidity

Figure 4. Transition temperatures of **2** of the transition from vesicles to wormlike micelles as a function of pH. Error bars denote the width of the transition.

References

- (1) Wagenaar, A.; Engberts, J. B. F. N. *manuscript in preparation*
- (2) Size distributions were obtained by fitting a Gaussian function to intensity as function of the logarithm of the particle diameter. The width of the size distribution is then the width at half height.