Spontaneous vesicles formation of a catanionic association involving a head and tail functionalized amino-calix[6]arene

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1. Characterization Methods

$^1$H and $^{13}$C NMR spectra of the amino-calix[6]arene were recorded on Brucker ARX 250 spectrometer at nominal frequencies of, respectively, 250 MHz for $^1$H and 62.5 MHz for $^{13}$C. $^1$H and $^{13}$C NMR spectra of the sugar-based surfactant and of the catanionic association were recorded on Brucker Avance 300 spectrometer at nominal frequencies of, respectively, 300.18 MHz for $^1$H and 75.48 MHz for $^{13}$C. Chemical shifts ($\delta$) are denoted in ppm using residual solvent peaks as internal standard.

A Waters Q-Tof Ultima API mass spectrometer with an electrospray ionisation source (ESI) in positive mode was used for HRMS measurement. The capillary, cone and Rf lens potentials were set to 3 kV, 35 V and 40 V respectively. The source and desolvation temperature were set to 100 and 120°C and the collision energy was 10 eV in TOF-MS mode. Analyses were monitored with MassLynx 4.0 software. To measure the exact mass, the spectrometer was calibrated with H$_3$PO$_4$ clusters.

2. Synthesis


The tris-azido calixarene derivative$^1$ (100mg, 80μmol) was dissolved in THF (5.3mL). N,N-dimethylpropargylamine (52μL, 480μmol), sodium ascorbate (19mg, 96μmol) and CuSO$_4$.5H$_2$O (650μL) were added to the solution. The reaction mixture was refluxed under argon for 96 h and the concentrated. The crude product was dissolved in CH$_2$Cl$_2$ (20mL) and washed with water (20mL) containing 10 drops of 28% aq. ammonia. The organic layer was separated, washed with brine (20mL), dried over Na$_2$SO$_4$, filtered and concentrated. The brown solid was triturated with Et$_2$O (2x20mL). After centrifugation, 100mg (83%) of amino-calix[6]arene were obtained as a tan solid.

ESI-MS (MeOH) m/z: 1501.9 [M+H]^+, 752.1 [M+2H]^2+. RMN £H (250 MHz, CD3CN, in the presence of 1 equiv. of Zn(H2O)6(ClO4)2, 300 K) δ ppm: 1.36 (s, 27H, tBu); 2.35 (br s, 18H, NMe2); 3.56-3.73 (m, 30H, NCH3, OCH3, CH2NMe2 and ArCH2); 4.10 (d, J = 16 Hz, 6H, ArCH2); 5.12 (br s, 6H, CH2im); 6.50 (s, 6H, HAr tria); 6.94 (s, 3H, HIm); 7.42 (s, 6H, HAr tBu); 7.46 (s, 3H, HIm); 7.58 (br s, 3H, Htriazole). RMN £C (62.5 MHz, CD3CN, in the presence of 1 equiv. of Zn(H2O)6(ClO4)2, 300K) δ ppm: 26.3, 31.2, 31.7, 35.3, 45.2, 53.1, 54.4, 61.2, 64.9, 68.4, 118.3,119.5, 120.9, 123.4, 125.0, 128.2, 130.2, 132.4, 133.7, 137.2, 144.0, 148.1, 149.2, 155.3, 156.1. IR (ATR) ν cm⁻¹: 2953.7, 1601.6, 1363.1, 1285.1, 1229.8, 1045.2, 1006.8. Anal. Calcd C 87H108N18O6 -5/3CH2Cl2: C, 64.80; H, 6.83; N, 15.34. Found: C, 65.15; H, 7.09; N, 14.71.

Note: in the absence of Zn(II), the £H NMR spectrum presents extremely broad and ill-defined resonances, which makes its interpretation impossible. This is due to conformational moves that occur at the same rate as the NMR time scale analysis. Upon the addition of just one equiv. of Zn(II), the whole structure is rigidified and the spectrum displays sharp resonances characteristic of the mononuclear complex, the metal ion being bound at the tris-imidazole core. This Zn(II) complex was further characterized by ESI-MS (MeOH) m/z: 603 [M+3H+Zn+2(ClO4)+CH3CN]3+, 570 [M+2H+Zn+ClO4+CH3CN]3+, 536 [M+H+Zn+CH3CN]3+.

![Figure S1](image-url): £H NMR spectrum (250 MHz, CD3CN, 300 K) of the amino-calix[6]arene in the presence of 1 equiv. of Zn(H2O)6(ClO4)2.

2.2. Synthesis of the sugar-based surfactant

1,7-lactobioamidohexanoate de sodium

A mixture of sodium hydroxide (224mg, 5.59mmol), 7-aminoheptanoic acid (810mg, 5.59mmol) and lactobionic acid (2,00g, 5.59mmol) in 50mL of methanol was heated to 50°C for 24h. Then the resulting mixture was concentrated to dryness. Flash chromatography of the residue (CHCl3/CH3OH/H2O: 2.5/7/0.5, Rf=0.6) was performed to give a white powder (64%).
ESI-MS (H₂O): 508 [M+H]⁺, 530 [M+Na]⁺, 484 [M-Na]⁻. RMN ¹H (300 MHz, D₂O, 298 K) δ ppm : 1.25 (m, 4H, CH₂); 1.47 (m, 4H, CH₃CH₂COO⁻ and CH₃CH₂CONH); 2.18 (t, J=7.5 Hz, 2H, CH₂COO⁻); 3.14 (m, 2H, CH₂CONH); 3.46-3.30 (m, 12H, CH and CH₂ of the sugar moiety); 4.28 (d, J=9 Hz, 1H, anomeric H). RMN ¹³C (75 MHz, D₂O, 298 K) δ ppm : 25.05, 25.67, 28.06, 28.22, 35.85, 39.06, 61.06, 61.93, 68.59, 70.36, 71.03, 71.40, 72.33, 72.44, 75.35, 80.96, 103.47, 173.88, 182.02. IR (KBr) ν cm⁻¹: 1635 (s, HNC=O); 1571 (s, C=O asym); 1406 (s, C=O sym).

1,7-lactobioamidoheptanoic acid

To a suspension of 1,7-lactobioamidoheptanoate de sodium (0.88 mmol, 450 mg) in 50 mL of methanol was added about 2 g of proton-exchange resin (Amberlite IR 120-H⁺). The resulting mixture was stirred at room temperature for 3 h and filtered to eliminate the resin. The organic layer was concentrated under reduced pressure. The residue was purified by flash chromatography (acetone/H₂O: 9/1, Rf=0.4) and then freeze-dried to give a white powder (90%).

ESI-MS (H₂O): 508 [M+Na]⁺, 484 [M-H]⁻. RMN ¹H (300 MHz, D₂O, 298 K) δ ppm : 1.27 (m, 4H, CH₂); 1.49 (m, 4H, CH₃CH₂COO⁻ and CH₃CH₂CONH); 2.31 (t, J=6 Hz, 2H, CH₂COO⁻); 3.17 (t, J=6 Hz, 2H, CH₂CONH); 3.46-4.31 (m, 12H, CH and CH₂ of the sugar moiety); 4.48 (d, J=9 Hz, 1H, anomeric H). RMN ¹³C (75 MHz, D₂O, 298 K) δ ppm : 24.16, 25.56, 27.76, 28.18, 33.71, 39.00, 61.06, 61.92, 68.59, 70.37, 71.02, 71.40, 72.33, 72.44, 75.35, 80.96, 103.47, 173.89, 179.26. IR (KBr) ν cm⁻¹: 1644 (s, HNC=O); 1724 (s, COOH). Anal. Calcd C₁₉H₃₅NO₁₃: C, 47.01; H, 7.22; N, 2.89; Found : C, 46.88; H, 7.11; N, 2.60.

Figure S2: ¹H NMR spectrum (300 MHz, D₂O, 298 K) of the 1,7-lactobioamidoheptanoic acid
2.3. Association 3:1 between the polyamino-calix[6]arene and the sugar-derived surfactant

To 0.10mmol of 1,7-lactobioamidoheptanoic acid (48.4mg) in 30mL of distilled water was added 0.22mmol of calixarene (50mg). After 24h of stirring at room temperature, a homogeneous solution was obtained giving quantitatively a tan powder after water removal by freeze-drying.

ESI-HRMS (H2O): 986. 5796 [M+3H]+. RMN 1H (300 MHz, DMSO, 298 K) δ ppm : 0.74 (m, 4H, CH₂); 1.22 (s, 27H, tBu); 1.40 (m, 4H, CH₂CH₂COO⁻ and CH₂CH₂CONH); 2.20 (m, 20H, NMe₂ and CH₂COO⁻); 3.04-4.07 (m, 50H, NCH₃, OCH₃, CH₂NMe₂, ArCH₂, CH₂CONH, CH and CH₂ of the sugar moiety); 4.26 (d, 3JHH=6Hz, 1H, anomeric H); 4.98 (br s, 6H, CH₂Im); 6.63 (s, 6H, HAr₃tn); 6.88 (s, 3H, HIm); 7.21 (s, 3H, HIm); 7.56 (s, 6H, HAr₃tn); 7.90 (br s, 3H, H₃tn); 8.63 (br s, 1H, NHCO). RMN 13C (75 MHz, DMSO, 298 K) δ ppm : 183.70 (COO⁻). IR (KBr) ν cm⁻¹: 1647 (s, HNC=O); 1560 (s, C=Oasym); 1413 (s, C=Osym).

Note: ¹H NMR spectra of the 3:1 catanionic association showed a loss of resolution comparatively to the sugar-based surfactant under the same conditions of analysis. This modification in the relaxation time of the protons was attributed to a self-assembling phenomenon where protons were enclosed in a less dynamic and more structured environment. In this regard, it is also very difficult to obtain ¹³C NMR spectra that can be fully interpreted.

Figure S3: ¹H NMR spectrum (300 MHz, DMSO, 298 K) of the catanionic association
Figure S4: Plot of the surface tension versus logarithm of the concentration for the catanionic assemblies at 25°C

Figure S5: Size distribution of the aggregates formed by the catanionic association at pH=6.8

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Table S1: Mean hydrodynamic diameter and polydispersity index (into the brackets) for aggregates spontaneously form in aqueous solution, determined by DLS at 25°C