**Supporting Information for**

**Dynamic covalent assembly of stimuli responsive vesicle gels**

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**General**

Diamines $D_3$ (1,3-diamino-propane), $D_4$ (1,4-diamino-butane), $D_5$ (1,5-diamino-pentane) $D_6$ (1,6-diamino-hexane), $D_7$ (1,7-diamino-heptane), $D_8$ (1,8-diamino-octane), $D_9$ (1,9-diamino-nonane), and $D_{10}$ (1,10-diamino-decane) are commercially available from Sigma Aldrich and Acros Organics and were used without further purification. Aldehyde $A$ (N,N-dimethyl-N,N-di(3-(4-formylphenyl)propyl)ammonium bromide) was synthesized according to a previously published procedure. LC-MS was performed on a Shimadzu LCMS-2010A instrument using a Xbridge shield RP 18 5µm 4.6*150mm column using a H$_2$O/ethanol gradient acidified with 0.1% TFA with an injection volume of 40µl and a flow of 1.0 ml/min.

**Critical Gelation Concentration (CGC) determination**

After mixing aqueous solution of $A$ and $D$ in 1 cm vials, the samples were vortexed and stabilized overnight. A material was considered to be a gel when no flow was observed in an inverted vial over the course of 8 hours.

**Fluorescence spectroscopy**

Fluorescence spectroscopy was performed on a Jasco J-815 CD-spectrometer equipped with emission accessory at 25°C. The temperature was controlled using a Jasco PFD 4252/15 Peltier temperature unit. All samples contained Nile Red probe in 2 µM concentrations and were excited at 550 nm. It should be noted that already at very low concentrations of $A\bullet D_n$ ($[A]_0 < 0.5mM$) a small but significant blue shift in Nile Red emission from 660 nm (pure water) to 650 nm was observed, most likely due to the formation of premicellar aggregates.

![Figure S1: a) CAC determination for $A\bullet D_7$ (blue) and $A\bullet D_9$ (red), using Nile Red fluorescence spectroscopy.](image-url)
Rheology

Experiments were performed on the ARES Rheometric Scientific rheometer, with a plate-plate cell and a temperature control unit.

![Figure S2: Frequency dependence of the elastic (G', blue) and loss (G'', red) moduli of an equilibrated A●D₉ gel ([A]₀ = [D₉]₀ = 40mM).]

The sol-gel transition was determined with a temperature sweep. The sample was heated from 25°C to 70°C and back to 25°C, with steps of 5°C. After each step, the samples were equilibrated for 5 minutes at the elevated temperature. All measurements were measured at an angular frequency of 6.28 rad/s within the linear viscoelastic regime.

Dynamic Light Scattering

Dynamic Light Scattering samples were measured in MilliQ water at 25°C on a ZetaSizer Nano series Nano-ZS (Malvern Instruments). Building blocks A and D₉ were mixed in pure water ([A]₀ = 40mM, [D₉]₀ = 40mM, pH 11.6 ± 0.2). Directly after mixing, the scattered intensity as a function of mixing time was measured (Figure S3).
**Figure S3:** Scattering over time measured by DLS of A●D₉ ([A]₀ = 40mM, [D₉]₀ = 40mM).

**Confocal Laser Scanning Microscopy**

Confocal Laser Scanning Microscopy (CLSM) pictures were obtained in the fluorescence mode on a Zeiss LSM 700 confocal laser scanning microscope. Nile Red was used as fluorescent probe in 10μM concentrations. The vesicle solutions were deposited on an 8 well chamber. The laser beam was focused on a 40X oil immersion objective and the sensitivity of detectors and filters were adjusted accordingly in order to obtain maximum signal to noise ratio.

**Figure S4:** a) CLSM micrograph of an A●D₇ gel ([A]₀ = 30mM, [D₇]₀ = 30mM, pH 11.6 ± 0.2).

Above the gel-sol transition temperature (T=65°C, [A]₀ = 25mM, [D₉]₀ = 25mM) no vesicles or networks could be observed by CLSM. Upon cooling back to 25°C vesicles were reformed within 8 minutes after heating (Figure S5a), while the first aggregated vesicles were observed around 15 minutes (Figure S5b). After 50 minutes, a vesicle network had formed again (Figure S5c).

**Figure S5:** Vesicle gel morphology as function of temperature studied with CLSM. A stable A●D₉ vesicle gel was heated (>T_{G-S}) and cooled back to room temperature upon standing. a) After 8 minutes cooling, scale bar = 20 μm; b) after 15 minutes cooling, scale bar = 5 μm; c) after 50 minutes cooling, scale bar = 5 μm.

**LC-MS analysis of A●D₉**

To estimate the product constitution of a A●D₉ vesicle gel ([A]₀ = 40mM, [D₉]₀ = 40mM, pH 11.6 ± 0.2), the mixture was analysed by LC-MS. The imine exchange of the equilibrated mixture was quenched by adding NaBH₄ to the vesicle gel, as such reducing A and possible (A-D)ₙ polymers to their corresponding non-dynamic alcohols and amines. As a result of adding NaBH₄, the gel was destroyed and partly precipitated from solution. Therefore ethanol
was added to the quenched slurry to re-dissolve the precipitate. The sample was analysed by LC-MS using a H₂O / EtOH gradient containing 0.1% TFA with a UV detection wavelength of 207 nm (Figure S6). Using an internal standard, it was noticed that only 30% of the expected product eluted from the LC column, indicating the formation of insufficiently mobile long oligomers and polymers. Attempts to analyse these polymers by GPC stranded due to an apparent strong binding of the polycationic material to the GPC stationary phase. The peaks in the chromatogram were identified by mass spectrometry (ESI-MS, positive ion mode) as alcohol, amino alcohol and amine. The number of repeating units for D(AD)_m is approximately between m = 6 (calculated m/z = 488.43) and m =11 (calculated m/z = 477.51). Various macrocyclic (AD)_m type constituents were also detected (calculated m/z = 464.40) together with some short oligomeric compounds (Figure S6).
Figure S6: a) Chromatogram of a A●D₉ vesicle gel ([A]₀ = 40mM, [D₉]₀ = 40mM) b) H₂O / EtOH gradient with 0.1% TFA. c) ESI-MS spectra and compound identification of imine oligomers. Repeating unit of the growing chain is displayed in brackets and estimated polymer length is displayed as subscript. Subscript m indicates a macroring compound. Constituents were detected with UV at λ = 207 nm. The corresponding m/z values for each constituent are displayed on the horizontal axis.