Supporting Information

Caging metal ions with light-responsive nano-polymersomes

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Table of Contents

Figure S1: Dynamic light scattering (DLS) of nano-polymersomes self-assembled with different concentrations of OB29 in DMSO S3

Figure S2: DLS of nano-polymersomes self-assembled with different percentages of DMSO in buffer S4

Figure S3: Cryo-TEM of nano-polymersomes self-assembled with different percentages of DMSO in buffer S5

Figure S4: DLS of nano-polymersomes with varying vortex time for self-assembly S6

Figure S5: Cryo-TEM of nano-polymersomes with varying vortex time for self-assembly S7

Figure S6: Reproducibility of nano-polymersome size S8

Figure S7: Structure of meso-to-meso ethyne-bridged (porphinato)zinc(II) dimer (PZn_2) S9

Equation S1: Equation used to calculate fractional release S9

Figure S8: FITC release from nano-polymersomes without PZn_2 or dextran S10

Methods S1: Cryo-TEM S11

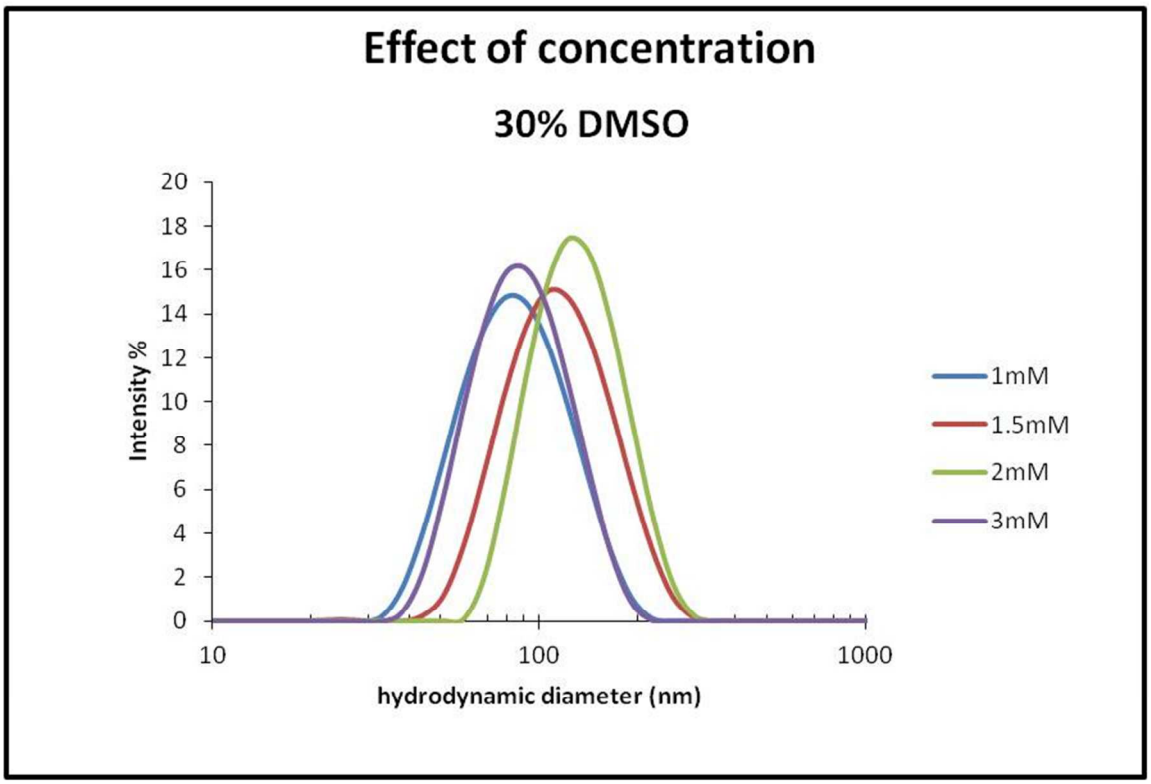
Figure S9: Nano-polymersome distribution *in vivo* S12

Figure S10: Hydrodynamic diameter of metal ion-loaded nano-polymersomes determined by DLS S13

Equation S2: Ion release approximation S14

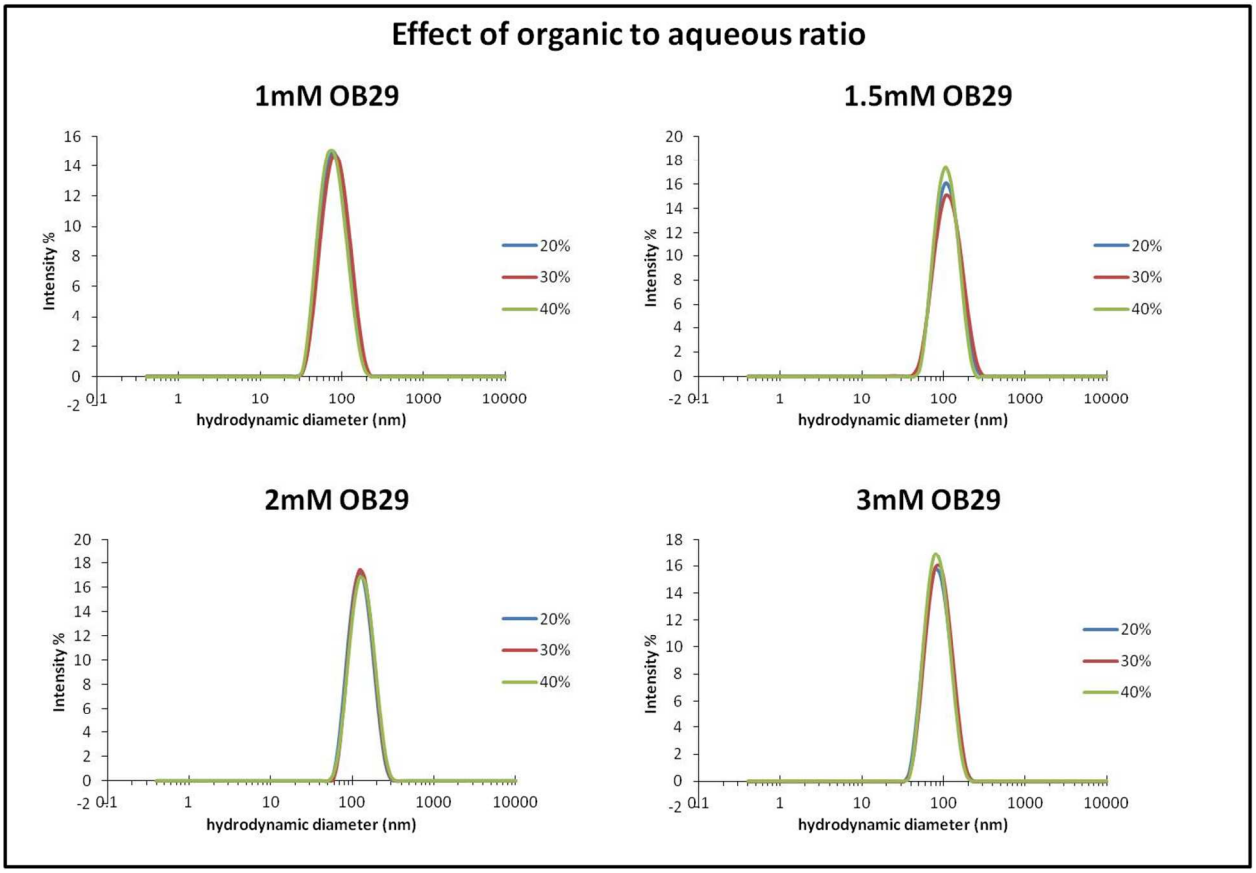
Reference S15

Figure S1: Dynamic light scattering (DLS) of nano-polymersomes self-assembled with different concentrations of OB29 in DMSO



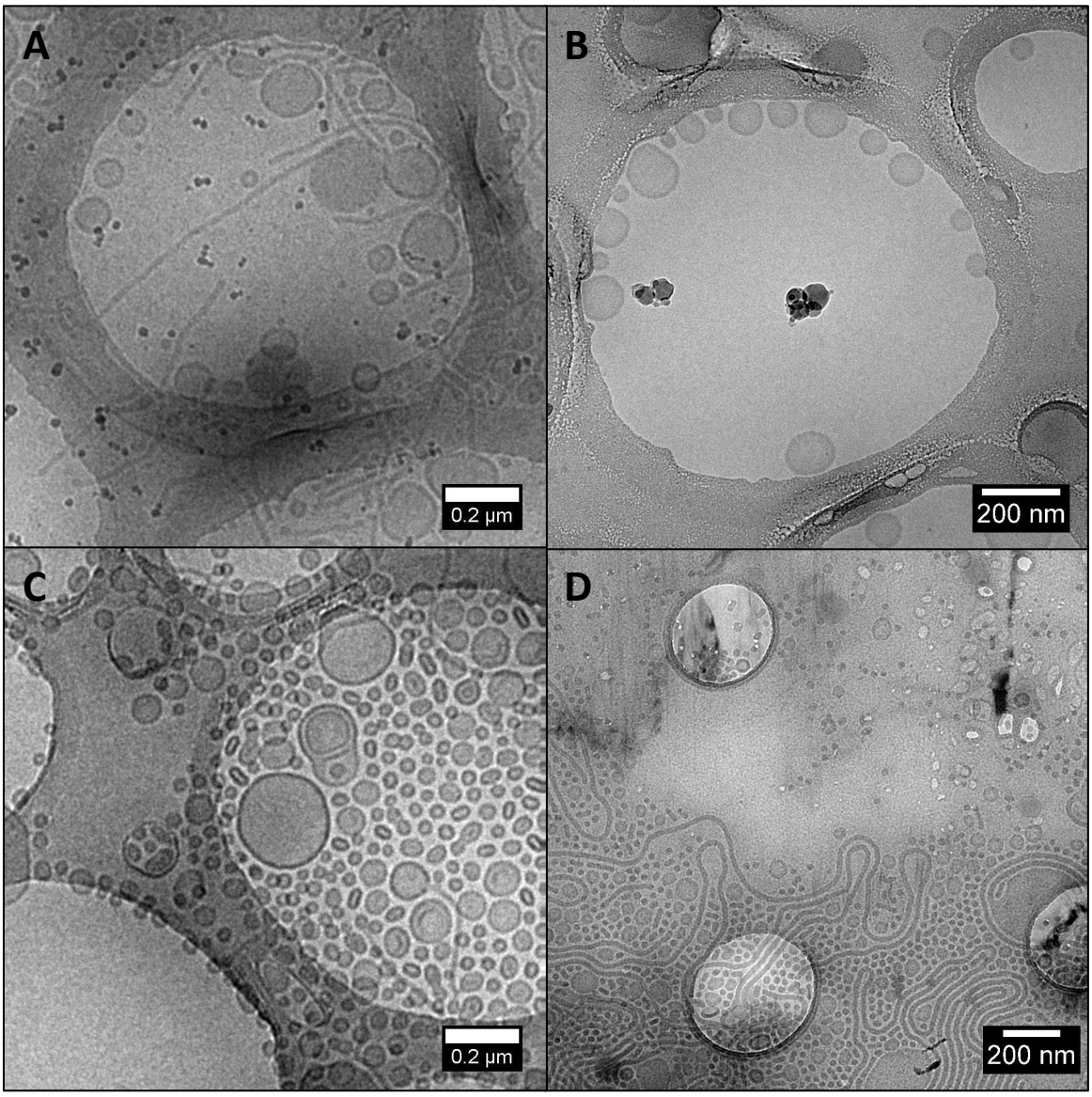
DLS measurements showing differences in vesicle size with varying OB29 concentration in 30% DMSO/70% PBS.

Figure S2: DLS of nano-polymersomes self-assembled with different percentages of DMSO in buffer



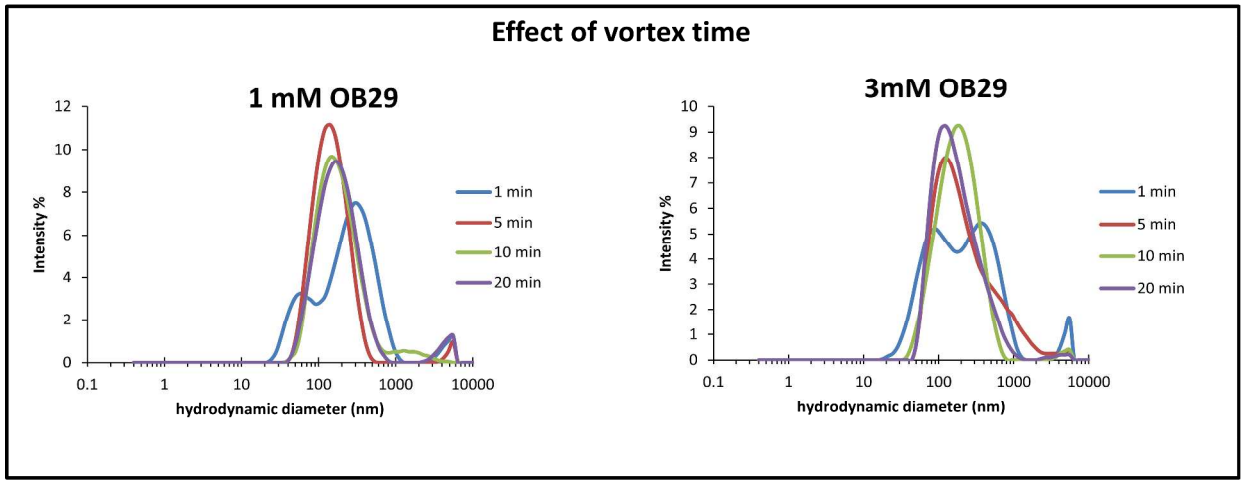
DLS measurements showing no significant effect on organic (OB29 in DMSO) to aqueous (0.1 M PBS) ratio. Percentage represents percent DMSO in final solution.

Figure S3: Cryo-TEM of nano-polymersomes self-assembled with different percentages of DMSO in buffer



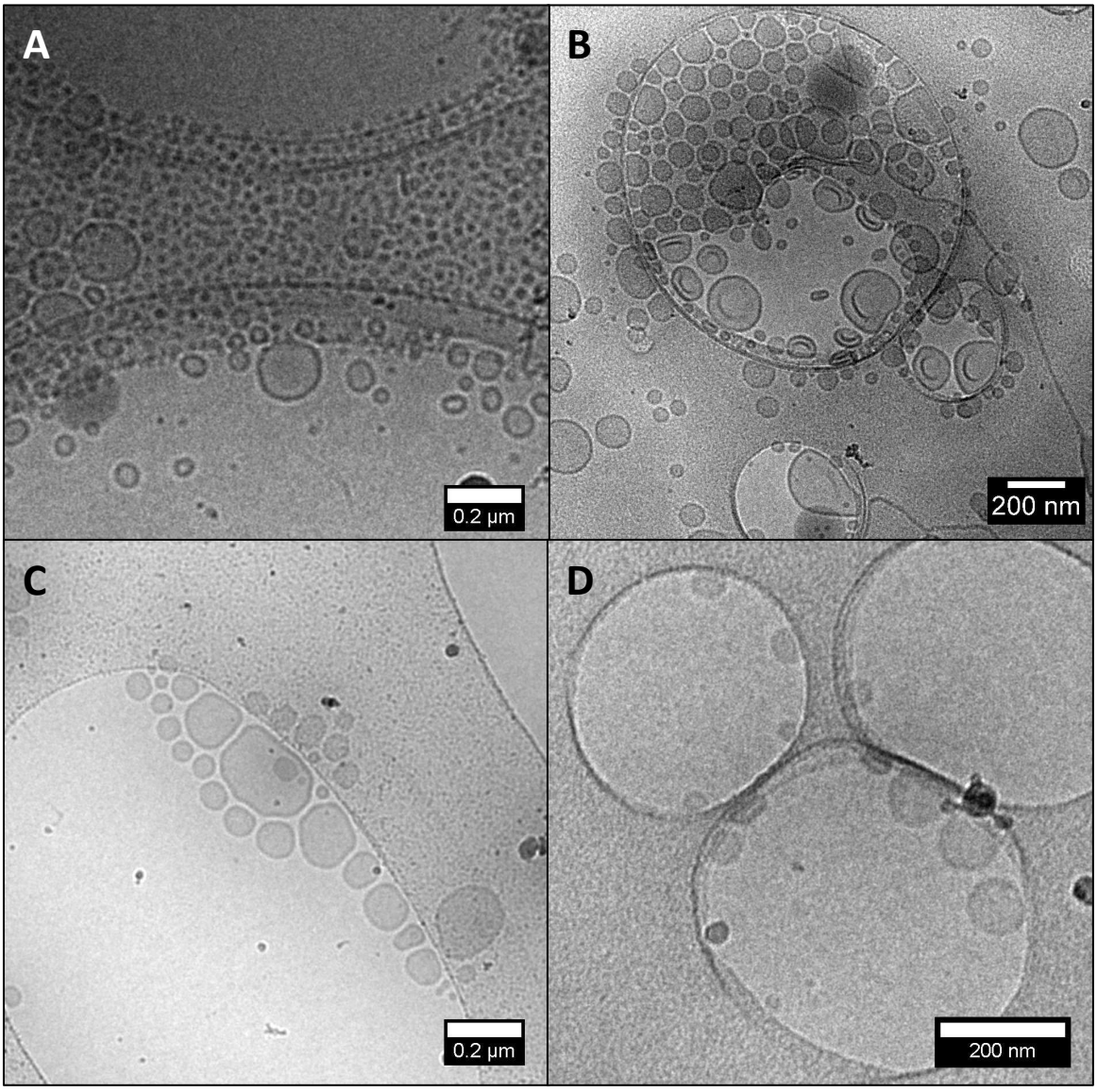
Cryo-TEM images showing the effect of organic-to-aqueous ratio on vesicle morphology. A) 10% DMSO resulted in vesicles and worm-like structures. B, C) 30% and 50% DMSO resulted in uniform, unilamellar vesicles. D) 70% DMSO resulted in vesicles and worm-like structures.

Figure S4: DLS of nano-polymersomes with varying vortex time for self-assembly



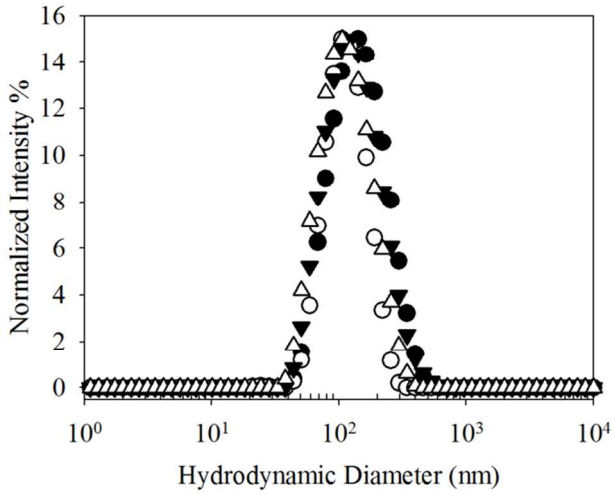
DLS showing the effect of vortex time on vesicle size. Vesicles were prepared by directly injecting OB29 in 30% DMSO into 0.1 M PBS followed by immediate vortexing. Vesicle size converged by 5 min for both 1 mM and 3 mM OB29 concentrations.

Figure S5: Cryo-TEM of nano-polymersomes with varying vortex time for self-assembly



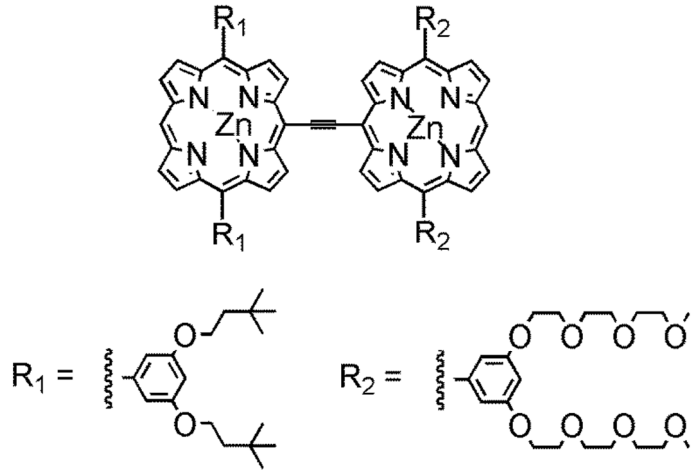
Cryo-TEM images showing the effect of vortex time on vesicle morphology. Vesicles were prepared by directly injecting OB29 in 30% DMSO into 0.1 M PBS followed by immediate vortexing. A) 1-min vortexing resulted in a large number of micelles. B) 5-min vortexing resulted in uniform, unilamellar vesicles. C, D) 10-min and 20-min vortexing showed no significant change from 5 min.

Figure S6: Reproducibility of nano-polymersome size



Dynamic light scattering (DLS) measurements for four independent nano-polymersome samples. Vesicles were prepared by directly injecting OB29 in 30% DMSO into 0.1 M PBS followed by immediate vortexing (final OB29 concentration of 1.5 mM). The average hydrodynamic radius was determined to be 120 ± 20 nm.

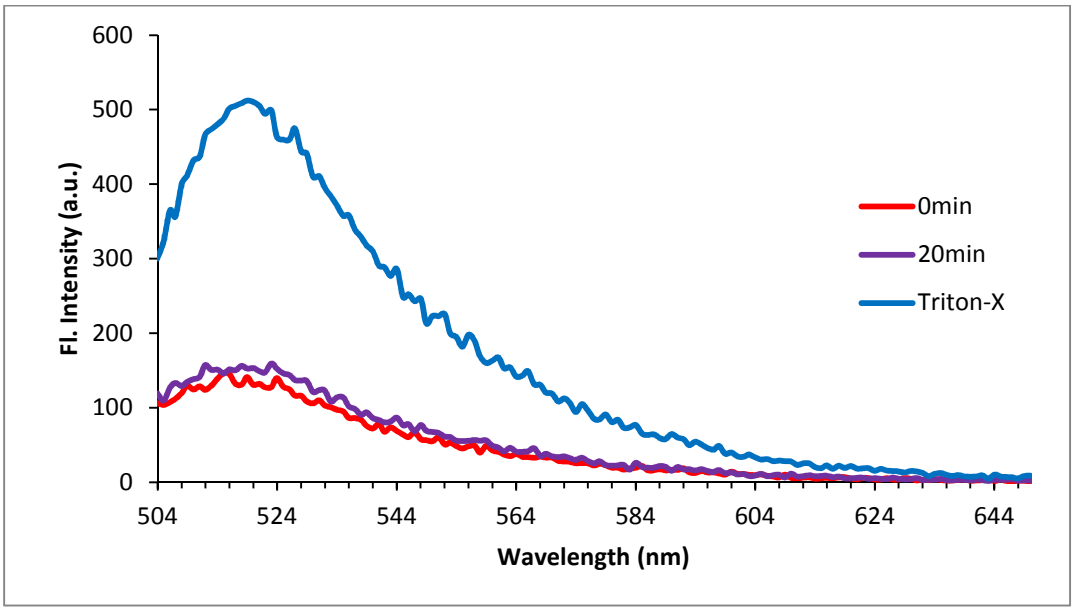
Figure S7: Structure of meso-to-meso ethyne-bridged (porphinato)zinc(II) dimer (PZn_2)



Equation S1: Equation used to calculate fractional release

$$\% \text{ Release} = \frac{Em_{\,irradiated} - Em_{\,background}}{Em_{\,triton} - Em_{\,background}} \times 100\%$$

Figure S8: FITC release from nano-polymersomes without PZn_2 or dextran



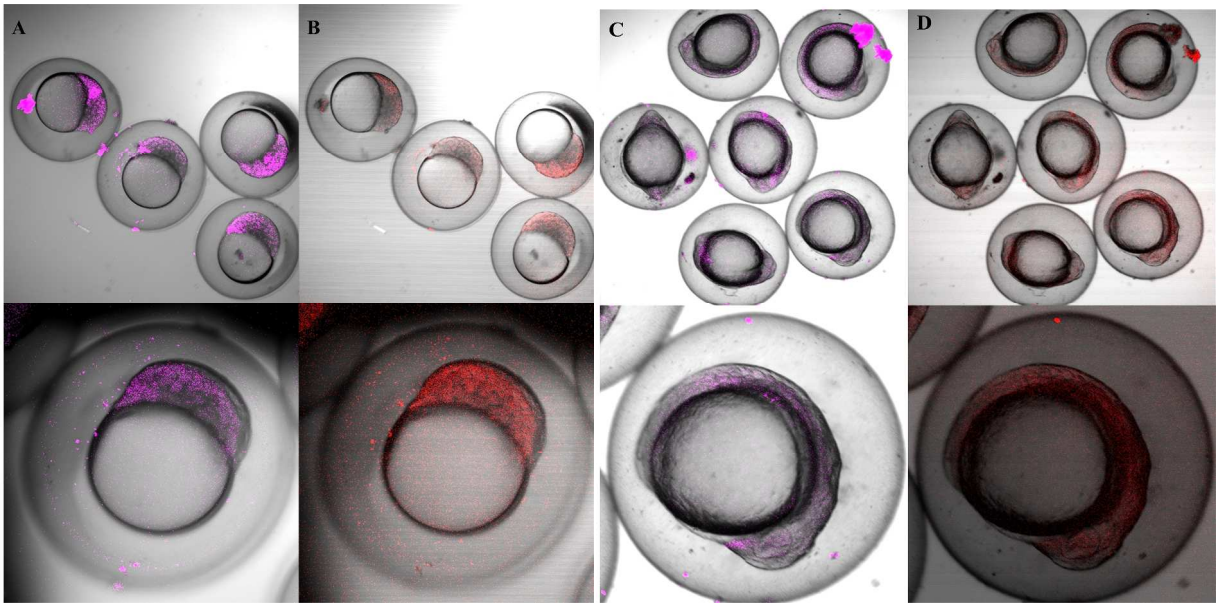
Nano-polymersomes were made with no PZn_2 in the membrane and without dextran in the aqueous core. These vesicles were subjected to 20-min irradiation (488, 515, 543, 633 nm lasers), and fluorescence intensity was compared to a positive control sample (Triton-X added to 0.1 vol%). Release from these vesicles was negligible.

Methods S1: Cryo-TEM

Krishna P. Singh Center for Nantechnology: Lacey formvar/carbon grids (Ted Pella) were cleaned with chloroform to remove the formvar coating, carbon coated with a Quorum Q150 ES carbon coater (Quorum Technologies, UK), and cleaned with hydrogen/oxygen plasma for 15 seconds using a Solarus Advanced Plasma System 950 (Gatan, Pleasanton, CA). The polymersome sample (2 μL) was deposited onto the grid and inserted into a cryoplunger (Cp3, Gatan). The sample was blotted by hand with filter paper and plunged into liquid ethane. The samples were subsequently transferred to a Gatan CT3500TR cryoholder and inserted quickly into a JEOL 2100 HRTEM (JEOL, Tokyo, Japan) operating at 200 keV. Images were captured using an Orius SC200 digital camera.

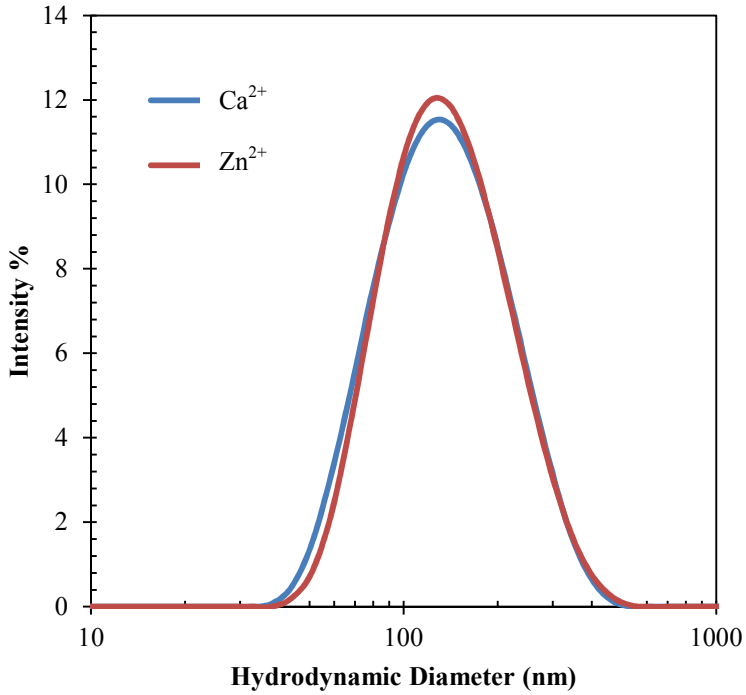
Electron Microscopy Resource Laboratory: Lacey carbon grids were glow discharged for 20 seconds at 25 mA to create a hydrophilic surface. Polymersome sample (3 μL) was applied and a thin film was formed by blotting the grid with filter paper. Vitreous ice was formed by rapid plunging into liquid ethane cooled to -180 °C by liquid nitrogen. Frozen hydrated samples were observed at -178 °C in a FEI (Hillsboro, OR.) Tecnai-12 microscope operated at 120 keV at magnifications between 6,500 and 15,000. Images were recorded on a Gatan (Warrendale, PA) US 1000 2048² CCD camera. All vesicle measurements were performed in Digital Micrograph.

Figure S9: Nano-polymersome distribution *in vivo*



Nano-vesicles were microinjected into zebrafish embryos at the 1-cell stage and monitored for A) PZn_2 emission and B) Texas Red emission. Embryos were imaged again at 20 hpf and monitored for C) PZn_2 emission and D) Texas Red emission. Collections of multiple embryos (top) were imaged with 4x objective. Single embryos (bottom) were imaged with 10x objective.

Figure S10: Hydrodynamic diameter of metal ion-loaded nano-polymersomes determined by DLS



The average hydrodynamic diameter of vesicles containing Ca^{2+} was determined to be 120 nm (blue). The average hydrodynamic diameter of vesicles containing Zn^{2+} was also determined to be 120 nm (red).

Equation S2: Ion release approximation

The number of vesicles can be approximated as follows:

$$\#\ \text{of vesicles(n)} = \frac{(\text{total polymer})(\text{vesicle surface area})}{\text{polymer chain density}}$$

The chain density is approximated to be 1 chain/nm^2, which has previously been used to estimate vesicle number.[1] The number of released ions can be determined as follows:

$$\#\ ions\ (c) = (release\ concentration)(total\ volume)(Avogadro's\#)$$

The following calculation is done for Ca^{2+} release (760 μM). Here we assume a polymersome diameter of 120 nm and a membrane thickness of 10 nm.

$$n = \frac{\left(\frac{1.5mmol}{1000mL} \times \frac{0.3mL}{1} \times \frac{mol}{10^3 mmol} \times \frac{6.022 \times 10^{23} chains}{mol}\right)\left(\frac{vesicle}{4\pi(50^2 + 60^2)nm^2}\right)}{\frac{1\ chain}{nm^2}}$$

$$c = \frac{760 \times 10^{-6} mol}{1000mL} \times \frac{1mL}{1} \times \frac{6.022 \times 10^{23} ions}{mol}$$

$$\frac{c}{n} = \frac{4.58 \times 10^{17} ions}{3.54 \times 10^{12} vesicles} = 130{,}000\ ions/vesicle$$

A similar calculation can be done for Zn^{2+} release (35 μM), where $c/n = 6{,}000$ ions/vesicle.

Reference

(1) Christian, N. A.; Milone, M. C.; Ranka, S. S.; Li, G.; Frail, P. R.; Davis, K. P.; Bates, F. S.; Therien, M. J.; Ghoroghchian, P. P.; June, C. H.; Hammer, D. A. Tat-functionalized near-infrared emissive polymersomes for dendritic cell labeling. *Bioconjugate Chem.* **2007,** *18*, 31-40.