Polymersome Poration and Rupture Mediated by Plasmonic Nanoparticles in Response to Single-Pulse Irradiation

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Supporting Information

Figure S1. Gel-assisted rehydration method for polymersome preparation.

Schematic representation of polymersome preparation via the gel-assisted rehydration method. Upon addition of rehydration buffer and heat to the sample, the copolymer film begins to swell and forms vesicles atop of the agarose film. The micron-sized polymersomes remain partially attached to the surface facilitating single-vesicle studies.
**Figure S2.** Ultrafast (fs) laser-microscope setup.

A Ti:Sapphire laser system (shown) or Nd-YAG ns laser (not shown) is steered and focused into the Zeiss Axio Examiner upright microscope for irradiation and imaging studies. The beam path is depicted by the green line.

**Figure S3.** Spot size determination.

Spot size is determined using a glass bottom petri dish covered with a series of ink blots. Water is added to the dish to mimic the aqueous experimental conditions. The ink is ablated at several spots, each by a single pulse, and the resulting damage spots are measured which serves as a gauge of the irradiated area. Note, damage spot is reported instead of beam waist, due to difficulties in measuring beam waists associated with the short focal lengths and aqueous environments for which these studies take place. Hence, irradiations are reported in terms of energy rather than fluence. Damage spot sizes were kept constant ranging from 8-9 µm.
**Figure S4.** Representative HR-TEM image of nano-polymersomes containing AuNPs after (a) nanosecond irradiation, (b) femtosecond irradiation, and (c) no irradiation. All scale bars are 25 nm.

Images were taken at 69,000x magnification and scale bars represent 25 nm. The nanoparticles, on the TEM grids, were often not found in circular groupings likely due to the drying process leading to vesicle rupture. Multiple images were taken for each sample and particle sizes were analyzed for a minimum of 5,000 particles per sample.

![Figure S4](image)

**Figure S5.** Release profile of encapsulant from polymersomes without AuNPs.

Normalized fluorescence intensity of FITC-dextran inside the core of polymersomes without AuNPs in the membrane. Vesicles were individually irradiated with a single, 532 nm fs pulse at the indicated pulse energies. Data is shown as the mean of at least three vesicles and error is reported as SEM.

![Figure S5](image)

**Figure S6.** Rupture threshold for vesicles without FITC-dextran in the core.

Each bar represents the percentage of polymersomes, either with or without AuNP loading, that ruptured completely in response to a single, 532 nm, fs pulse at the specified energy.

![Figure S6](image)
Table S1: Example data for normalized intensity as shown in Figures 5, 6, and S5.

Release curve data for polymersomes with AuNP loading irradiated at 212 nJ. The fluorescence intensity was determined in the selected ROI and was corrected by subtracting the intensity of an ROI set in the background. The ROIs were placed in the same position for every image within a time series.

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| TIME (SEC) | INTENSITY | BACKGROUND | CORRECTED INTENSITY | NORMALIZED INTENSITY |
|------------|-----------|------------|---------------------|--------------------|
| 0          | 246.012   | 99.138     | 146.874             | 1.000              |
| 3          | 217.200   | 98.719     | 118.481             | 0.807              |
| 13         | 178.771   | 100.386    | 78.385              | 0.534              |
| 23         | 156.263   | 99.510     | 56.753              | 0.386              |
| 33         | 143.353   | 99.279     | 44.074              | 0.300              |
| 43         | 137.145   | 98.605     | 38.540              | 0.262              |
| 53         | 133.314   | 98.826     | 34.488              | 0.235              |
| 63         | 132.009   | 99.534     | 32.475              | 0.221              |
| 73         | 129.789   | 98.084     | 31.705              | 0.216              |
| 83         | 127.915   | 98.950     | 28.965              | 0.197              |
| 93         | 126.216   | 98.873     | 27.343              | 0.186              |
| 103        | 123.239   | 97.254     | 25.985              | 0.177              |
| 113        | 123.043   | 97.137     | 25.906              | 0.176              |