Release mechanism of octadecyl rhodamine B chloride from Au nanorods by ultrafast laser pulses

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SUPPORTING INFORMATION

R_{18} loading yield

To calculate the yield of R_{18} loading, we used the change in fluorescence intensity of the R_{18} loaded NR solution's solvent. The fluorescence intensity of 5 μM R_{18}, the concentration used during NR-R_{18} conjugation, was measured (Figure S1, red line). We saved the supernatant of the first wash from the NR-R_{18} conjugation. The fluorescence intensity of this sample was also measured (Figure S1, black line). We used the difference in fluorescence intensity of these two samples to calculate the concentration in the supernatant of the wash to be 2.88 μM. Based on the assumption that changes in fluorescence intensity are due to conjugated R_{18} being retained in the precipitate after the centrifugation, we subtracted 2.88 μM from the 5 μM added. We

calculated 2.12 μM R_{18} were attached to the NRs. Since we added NRs to make a 10 nM solution, there are an average of 212 R_{18} per NR.

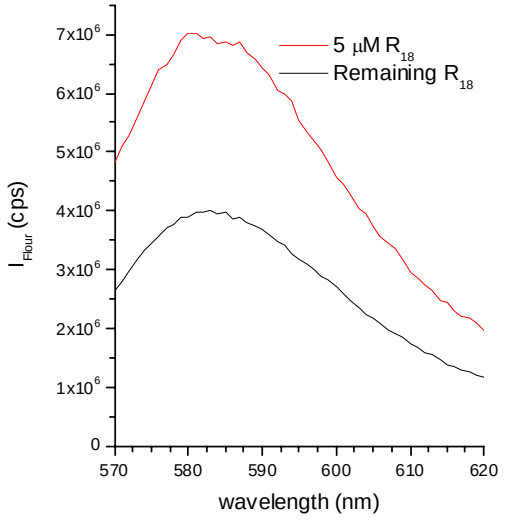


Figure S1. Fluorescence intensity of R_{18}-NR conjugation wash supernatant (black line) and a 5 μM R_{18} solution (red line), both in 10 mM CTAB.

Control for bulk heating due to the laser

Pulsed laser irradiation of NRs in solution is intended to heat the NR and its immediate vicinity only. As a function of NR concentration and laser fluence, the bulk solution temperature can rise with time. Because bulk temperature rise of the samples accelerate release, this unwanted effect could explain the acceleration of release as a function of laser fluence.

We inserted a K-type thermocouple into the cuvette containing samples for laser irradiation. We measured the temperature of 1 nM NR in 10 mM CTAB and 10 mM CTAB alone as a function of time. Because 10 mM CTAB does not absorb strongly at 792 nm, this solution's temperature rise is solely because of absorption by the thermocouple, an artifact of the

measurement technique. We subtracted the temperature of CTAB alone from the temperature measured in NR solution. The temperature rise of NR solution subjected to a 792 nm fs pulsed laser with fluence 330 $\mu J/cm^2$ was ΔT = ~0.7 °C (Figure 3a in the main paper). To determine if this temperature rise can explain the observed R_{18} release, we heated a sample of R_{18} loaded NRs in a 30 °C (ΔT = 7 °C) water bath and measured the difference between the fluorescence intensity of the solvent of heated and control samples. The fluorescence intensity of the heated sample (dots, Figure 3b in main paper) was the same as the unheated aliquot (line). These results suggest that the increased release rate is due to spatially confined temperature effects at the NR are specific to ultrafast laser irradiation, not bulk solvent heating by the laser.

Small AR NR control - Release only happens when the laser excites a SPR

We synthesized Au NRs intended to focus the heat generated by the laser onto target dye molecules. This focusing occurs because the absorption peak of the Au NRs, as determined by the aspect ratio of the NRs, corresponds to the laser's spectral output. To ensure the aspect ratio is a critical parameter for laser focusing and therefore release, we repeated the experiment with small aspect ratio NRs.

Small aspect ratio Au NRs were synthesized using the same non-seeding method previously described. Briefly, CTAB was added to a 7.5 mM NaCl solution to a final concentration of 100 mM. $HAuCl_4 \cdot 3H_2O$ and $AgNO_3$ were added to final concentrations of 2.5 mM and 0.5 mM, respectively, which turned the solution yellow-brown. After light agitation, L-ascorbic acid was added to a final concentration of 5 mM, which turned the solution clear after ~30 s of inversion agitation. $NaBH_4$ was added to a final concentration of 1 μM, and the solution was agitated by inversion for ~30 s. The solution was left at room temperature for >3 hs while the solution turned

deep purple/brown. R_{18} was loaded onto the NRs by incubating a 10 nM solution of NRs in 1 mM CTAB with a 5 μM solution of R_{18} overnight.

The small aspect ratio NRs were cylindrical with hemispherical caps and monodisperse (Figure S3a). They were 48.0 ± 7.7 nm long by 21.5 ± 3.9 nm in diameter, with a mean AR = 2.3 ± 0.4 (Figure S3b). The optical absorption spectrum (Figure S3c, solid line) confirms the longitudinal SPR, 639 nm, does not coincide with the 792 nm spectral output of the femtosecond pulsed laser (Figure S3c, dashed line).

13 μL of 10 nM NR-R_{18} conjugate stock were added to 104 μL of water. 13 μL of 100 mM CTAB were added to raised the CTAB concentration to 10 mM. A 65 μL aliquot of sample was set aside at room temperature. The remaining 65 μL aliquot was exposed to 60 – 1200 s of pulsed laser irradiation. Immediately after exposure, both aliquots were centrifuged to separate the NRs from the released R_{18}. The supernatant and precipitate from both aliquots were analyzed on the fluorometer. The fluorescence of each sample was normalized to the peak intensity of the sample that was set aside.

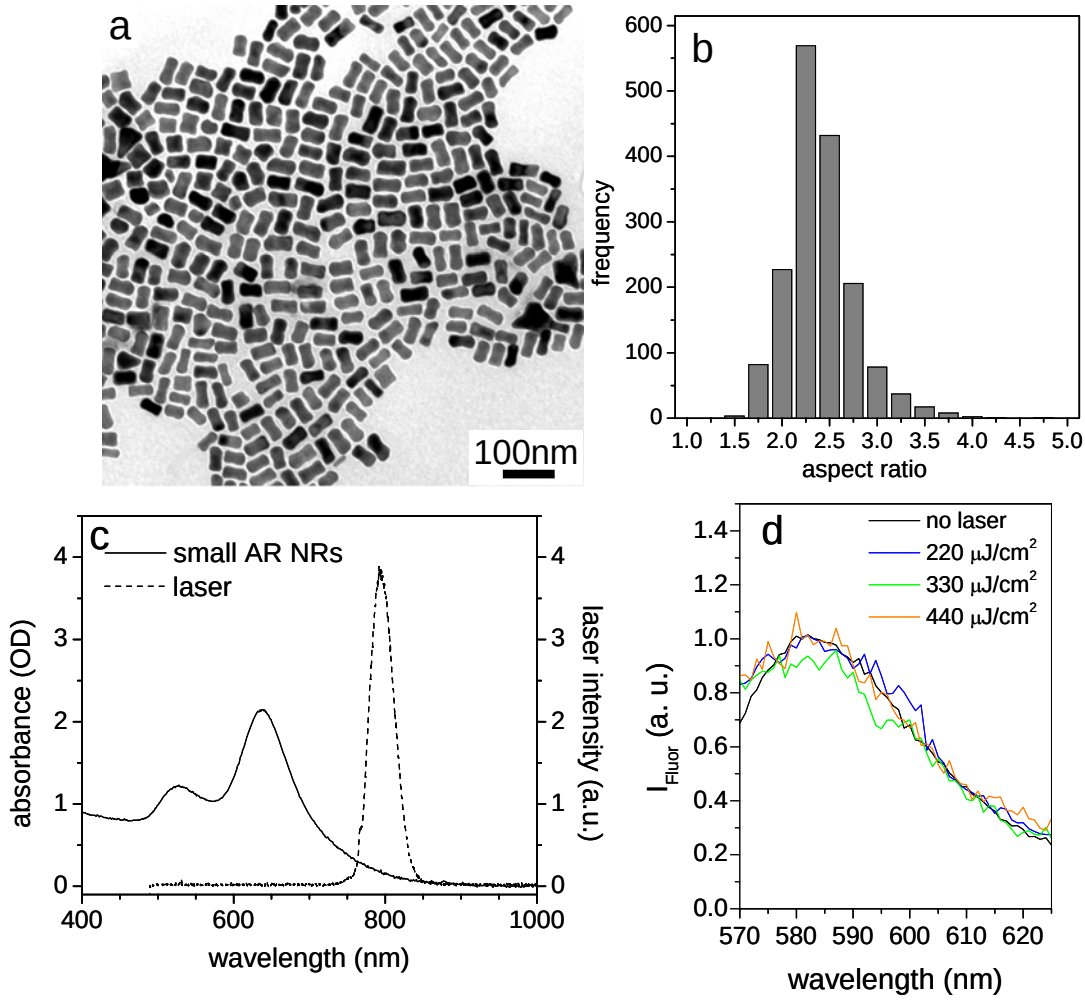


Figure S3. Small aspect ratio Au NR control. a) Typical TEM image of the small aspect ratio Au NRs. b) Histogram of Au NR aspect ratio, average AR = 2.3. c) Optical absorption spectrum of small aspect ratio Au NRs (solid black line) and spectral output of the laser excitation (black dashed line). d) Release of R_{18} from small aspect ratio NRs. Laser fluence: 220 μJ/cm^2 (blue), 330 μJ/cm^2 (green), 440 μJ/cm^2 (orange). The fluorescence intensity of the supernatant of laser irradiated samples normalized to the peak fluorescence intensity of the supernatant of an unexposed aliquot of the same sample. Supernatant fluorescence after irradiated NRs were separated from released R_{18} by centrifugation. Data for 20 minutes of laser exposure are shown.

After 20 min of irradiation, there is no observed change in fluorescence intensity of the supernatant as a function of laser fluence (Figure S3d). This is in contrast to the longer aspect NRs, which showed a strong dependence on the laser fluence (Figure 2 in the main paper). Based on this contrast, we find that laser irradiation does not affect the release rate of R_{18} from smaller aspect ratio NRs, whose longitudinal SPR absorption peak of 639 nm does not overlap the laser excitation. Thus, the laser release results are specific to NR-R_{18} conjugates where the NRs have their longitudinal SPR tuned to the spectral output of the laser.

Free R_{18} control – Observed release only happens R_{18} is conjugated to Au NRs

We selected R_{18} as a model for a biological molecule to study laser induced release because it, like most biological material, does not absorb light in the near infra red (Figure S4a). 50 nM R_{18} in 10 mM CTAB was exposed to the laser for 10 minutes. The fluorescence intensity of each sample was measured (Figure S4b). We find that laser irradiation does not affect the fluorescence intensity free R_{18}. This result indicates that the changes in fluorescence intensity of the solvent cannot be explained by direct absorption of laser irradiation by R_{18}.

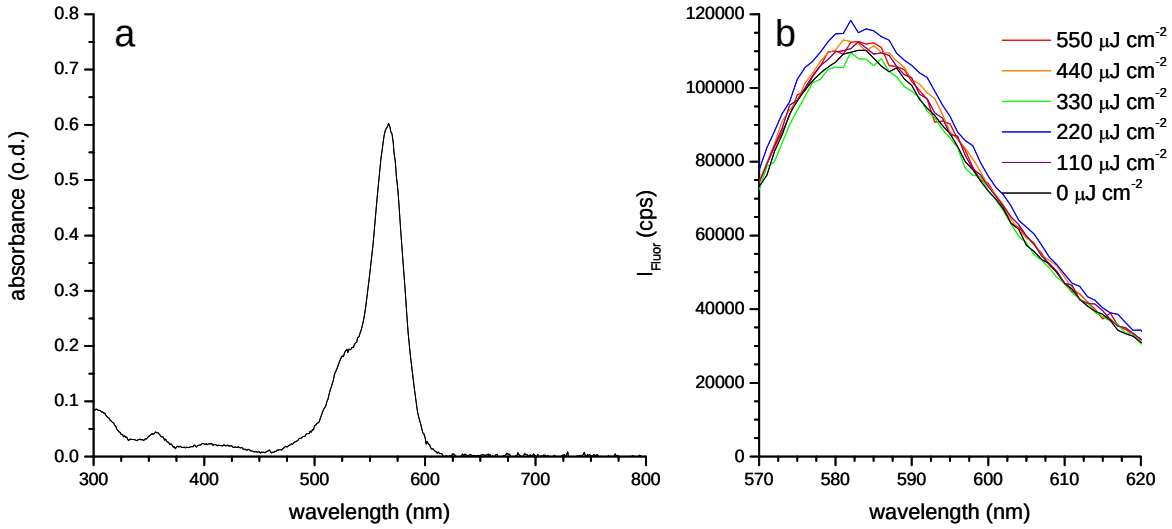
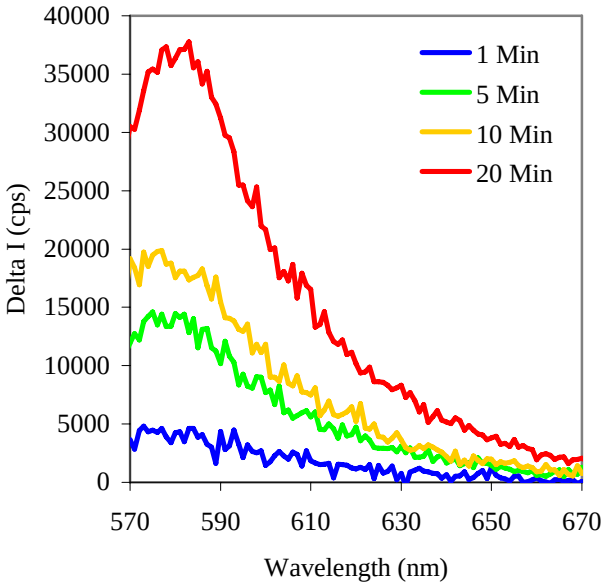
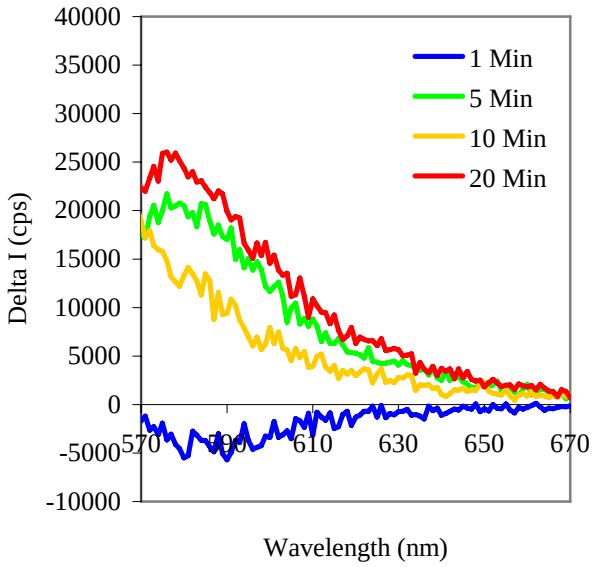


Figure S4. Characterization of R_{18} upon laser irradiation. a) Absorption spectrum of R_{18}. b) Fluorescence intensity of R_{18} in 10 mM CTAB after 10 minutes of laser irradiation. Laser fluence: 0 $\mu J/cm^2$ (black),110 $\mu J/cm^2$ (violet), 220 $\mu J/cm^2$ (blue), 330 $\mu J/cm^2$ (green), 440 $\mu J/cm^2$ (orange), 550 $\mu J/cm^2$ (red).

Additional data from laser induced release experiments





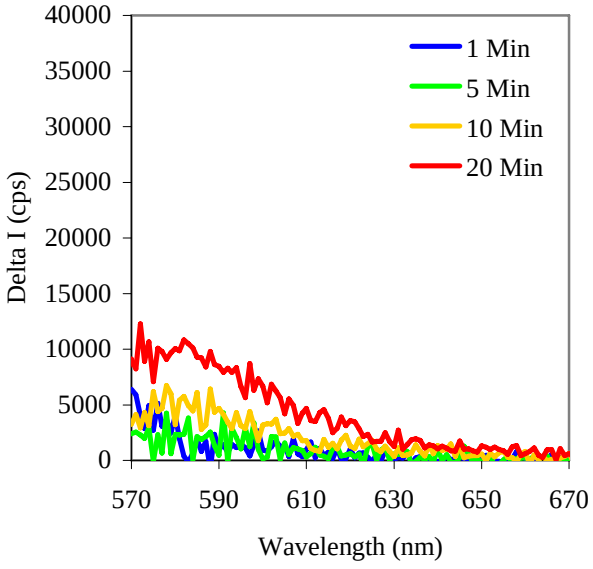


Figure S5. Additional example data (see Figure 2a in main text) from release of R_{18} from NRs under laser irradiation experiments. Plotted here is the difference between the supernatant fluorescence intensity of the laser irradiated and unexposed samples after NRs were separated from released R_{18} by centrifugation. In all plots, time length of laser irradiation: 1 min (blue), 5 min (green), 10 min (orange), 20 min (red). a) and b) Data from 2 separate trials at a laser fluence of 220 $\mu J/cm^2$. c) Data taken at a laser fluence of 110 $\mu J/cm^2$.