Electronic Supplementary Information

Strong plasmonic fluorescence enhancement of individual plant light-harvesting complexes

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This document comprises of the following information:

Data analysis:

Only brightness transients showing single step blinking and photobleaching dynamics were considered. Analysis of intensity brightness was performed using a home-written MATLAB (MathWorks) script based on an intensity change-point algorithm that uses the time-tagged photon data as input[1]. The fluorescence brightness of each complex was obtained from the first resolved intensity level, which was in all cases an ON state. For PFE, a minimum threshold for enhanced brightness was established. For this purpose, the intensity traces of dim complexes in the LHCII@AuNRs sample and their corresponding lifetimes were measured. Unenhanced complexes exhibited average brightness levels of 7.5 counts/10 ms, and typical lifetimes of 3.5 ns. The fluorescence lifetimes were obtained by fitting the decay traces with either a single exponential function (LHCII) or a multi-exponential function (LHCII@AuNRs), convoluted with the instrument response function (at λ_{em} = 680 nm) using a home-written Python algorithm. It uses the least-square minimization strategy to find the best-fit parameters for the given data[2]. The fluorescence peak position and FWHM distributions were obtained by fitting a skewed Gaussian to all the single-molecule fluorescence spectra as previously reported[3].

Supplementary Equations: Quantum yield and fluorescence lifetime in the presence of metallic NPs

The plasmonic excitation and emission enhancements were analyzed using a semi-empirical model proposed in ref 4. In the absence of metallic NPs or any other quenching interactions, the intrinsic quantum yield of an isolated pigment is given by:

$$Q_0 = \frac{\gamma_r}{\gamma_r + \gamma_{nr}},$$

where γ_r and γ_{nr} are the intrinsic radiative and non-radiative decay rates, respectively. The fluorescence lifetime is given by the inverse of the total decay rate:

$$\tau_0 = \frac{1}{\gamma_r + \gamma_{nr}}.$$

The coupling of pigments to metallic NPs modulates both the radiative and non-radiative decay rates. The modulated quantum yield can be expressed as follows[5]

$$Q_m = \frac{\gamma_m}{\gamma_m + \gamma_{nr,m}},$$

where γ_m and $\gamma_{nr,m}$ are the modified radiative and non-radiative rates in the proximity of metallic NPs. $\gamma_{nr,m}$ includes ohmic losses into the metal[6]. The modification of the total decay rates leads to shortening of the fluorescence lifetime, which is:

$$\tau_m = \frac{1}{\gamma_m + \gamma_{nr,m}}.$$

For simplicity, we can assume that $\gamma_{nr,m} \approx \gamma_{nr}$ when the pigment lies within an appropriate distance from the metal surface to prevent quenching of the excited state fluorescence to the metal. In our work, the minimum distance between the AuNR surface and LHCII was estimated to be ~4.8 nm, meaning that ohmic losses into the metal were minimized. Using the measured fluorescence brightness and lifetime of LHCII@AuNRs and LHCII, intrinsic fluorescence quantum yield of 0.26 of LHCII in solution and assuming that the modified non-radiative rate in the presence of AuNRs is constant and equivalent to the intrinsic non-radiative rate, we can estimate the emission enhancement using the equation $E_{em} = \frac{Q_m}{Q_0}$. Then, the excitation enhancement is determined from the overall plasmonic fluorescence enhancement as follows: $E_{exc} = \frac{PFE}{E_{em}}$.

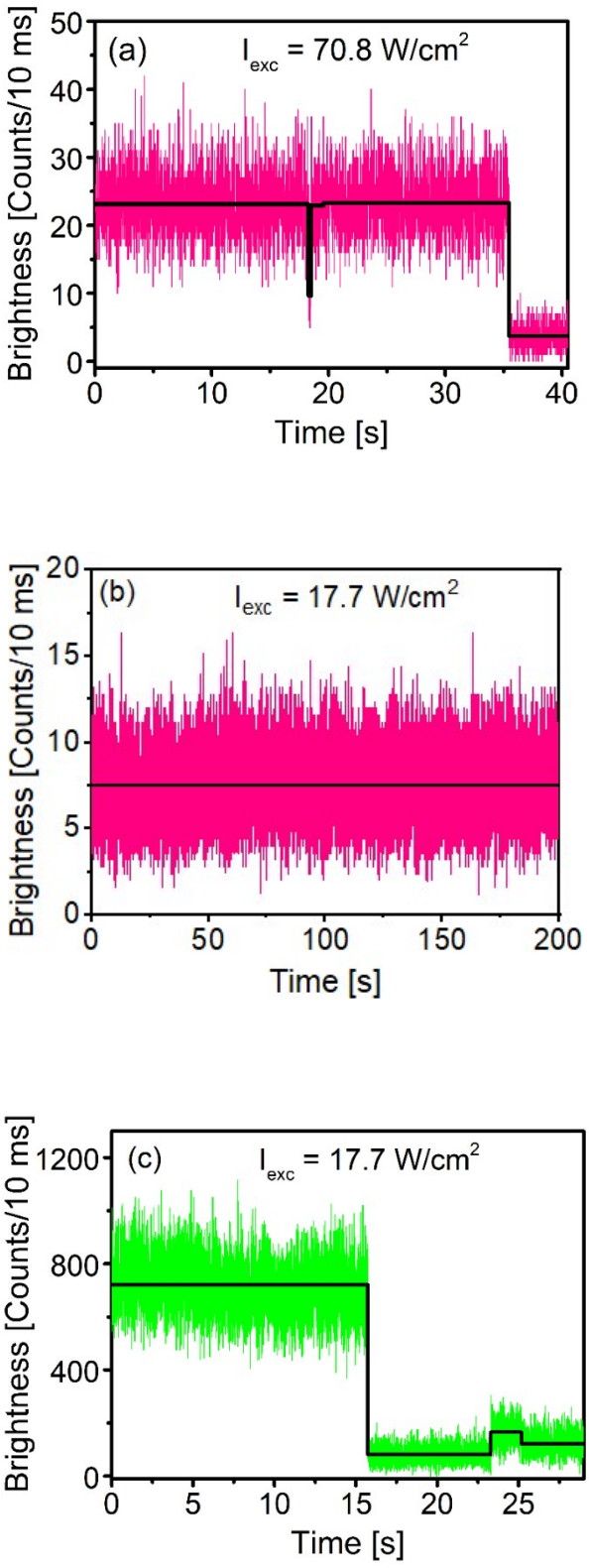

Fig. S1 Fluorescence brightness time traces of single LHCII complexes excited at 646 nm without AuNRs (a-b), and in the presence of a AuNR (c). The intensity levels (black) were obtained by a change-point algorithm after binning the time-tagged photons into consecutive 10 ms bins.

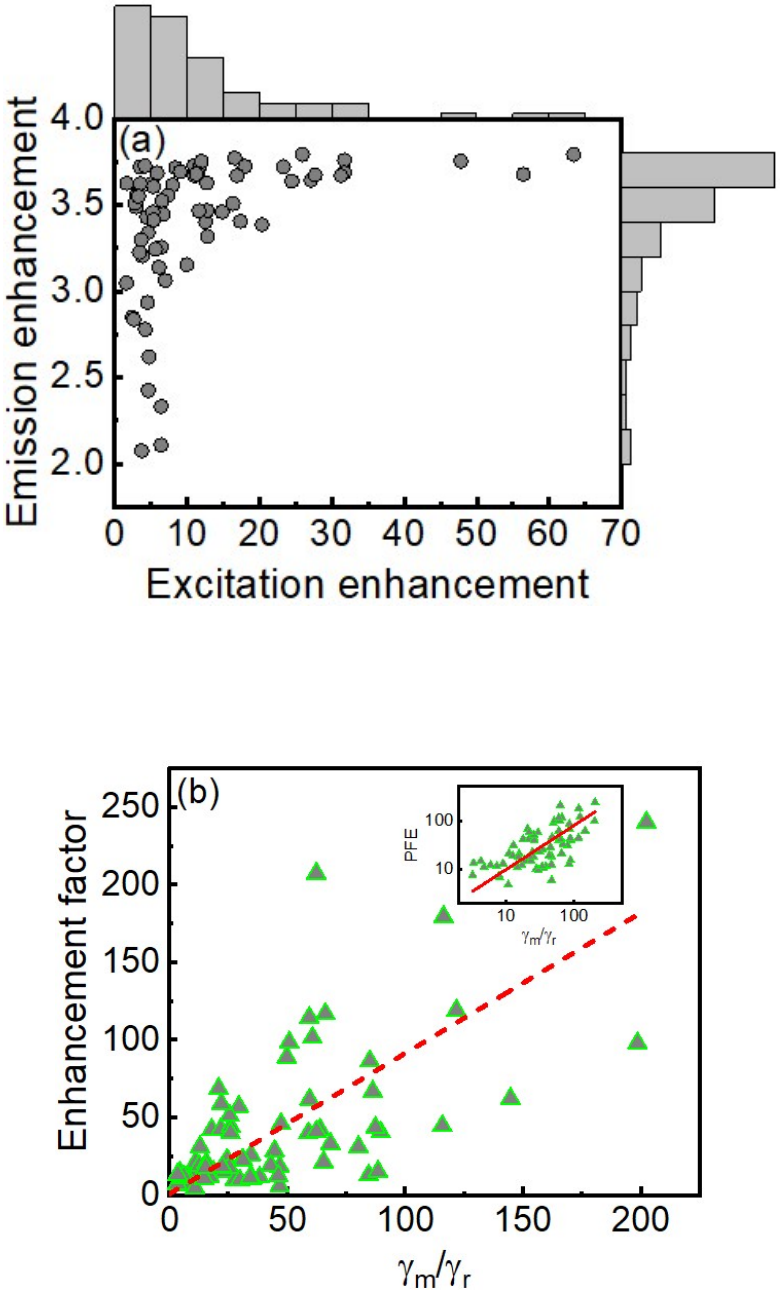

Fig. S2 (a) Correlation between excitation enhancement and emission enhancement factors. Data was distributed into bins of 5 and 0.2 for x and y-axes, respectively. (b) Correlation of plasmonic fluorescence enhancement factor and the ratio of the modified radiative rate to the intrinsic radiative rate. Insert is the same graph on a logarithmic scale, fitted with a linear function.

References

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