Lasing in blood: supplementary material

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1. Absorption/Emission spectra of ICG

Fig. S1. Normalized absorption spectrum (blue curve) and emission spectrum (red curve) of purified ICG in DI water from 660 nm to 880 nm.

2. Fluorescence spectra of ICG in ethanol, ICG in water, ICG+BSA, and ICG+HSA

Fig. S2. Emission spectra of ICG in water (green curve), ICG in ethanol (blue curve), ICG with BSA (0.6 mM) in PBS (red curve), ICG with HSA (0.6 mM) in PBS (purple curve), and background signal (black curve) under the same pump energy density. Excitation wavelength=660 nm. The ICG concentration was fixed at 0.2 mM for all measurements. To estimate the quantum yield (QY), we used the well-known QY of ICG in ethanol (13.2%) [1,2] as the reference. The measured QY is 0.48% for ICG alone in water, 4.1 % for ICG-HSA, and 3.9 % for ICG-BSA.

3. Fluorescence spectra of different BSA/ICG molar ratios

Fig. S3. **a**, Fluorescence emission spectra of ICG in different concentrations of BSA. The concentration of ICG was fixed at 0.2 mM. The BSA/ICG ratio varied from 0.75 to 3.8. **b**, Normalized fluorescence intensity as a function of the BSA/ICG molar ratio extracted from the peak value in **a**. The red dashed curve shows the quadratic fitting of experimental data to guide an eye ($R^2 > 0.95$). Excitation wavelength=660 nm.

4. Lasing profile of ICG + Human Serum Albumin (HSA)

Fig. S4. **a**, Lasing spectra of ICG (0.2 mM) with Human Serum Albumin (HSA) under various pump energy densities. Curves are vertically shifted for clarity. **b**, Spectrally integrated (900 nm - 920 nm) laser output as a function of pump energy density extracted from **a**. Solid line is the linear fit above the threshold, showing a lasing threshold of approximately 1.68 $\mu\text{J}/\text{mm}^2$.

5. Fluorescence spectra of ICG + Globulins

Fig. S5. Emission spectra of ICG with γ -globulins (0.1 mM) (blue curve) and ICG with γ -globulins (0.3 mM) (red curve), compared to ICG with BSA (0.6 mM) under the same pump energy density. The ICG concentration was fixed at 0.2 mM for all experiments. Excitation wavelength=660 nm.

6. Emission/absorption spectra of ICG+LDL and ICG+BSA

Fig. S6. **a**, Comparison of the emission spectra of ICG with LDL (0.01 mM) and ICG with BSA (0.6 mM) shows that the QY for ICG with LDL is approximately 12.8%. **b**, Normalized absorption spectra of ICG with LDL (0.01 mM) and ICG with BSA (0.6 mM). The absorption peak at 750 nm is due to ICG molecules, whereas the strong absorption at UV wavelengths is due to the intrinsic absorption of proteins and lipoproteins. The ICG concentration was fixed at 0.2 mM. Excitation wavelength=660 nm.

7. Lasing profile of human serum

Fig. S7. Comparison of the lasing spectra of serum alone and serum mixed with ICG (0.04 mM) under the same pump energy density of 20 $\mu\text{J}/\text{mm}^2$. Excitation wavelength=660 nm. Curves are vertically shifted for clarity.

8. Lasing profile of human whole blood

Fig S8. Comparison of the lasing spectra of human whole blood alone and whole blood mixed with ICG (0.04 mM) under the same pump energy density of 20 $\mu\text{J}/\text{mm}^2$. Excitation wavelength=660 nm. Curves are vertically shifted for clarity.

9. Optical system setup

Fig. S9. **a**, Illustration of the experimental setup. **b**, Picture of the experimental setup. The red line indicates the position of the OFRR, as the actual OFRR is too thin to be visible.

10. Lasing thresholds for different components

Table S1. Lasing threshold ($\mu\text{J}/\text{mm}^2$) for ICG with different components

	0.04 mM ICG	0.2 mM ICG	0.4 mM ICG
BSA	5.3	2.30	0.38-1.21
HSA	4.3	1.68	N/A
LDL	N/A	0.17	N/A
Serum	3.4	0.45	2.7
Blood	9.9	N/A	N/A

11. Discussion on lasing threshold

The lasing threshold, I_{th} , is determined by the fraction of dyes in the excited state, which is given by [3,4]

$$I_{th} \propto \frac{\gamma}{1-\gamma} \quad \text{and} \quad \gamma = \frac{n_1}{n_T} \propto \frac{\sigma_a}{\sigma_e},$$

where n_1 and n_T are the dye concentration in the excited state and total dye concentration, respectively. σ_a and σ_e are the dye absorption and emission cross section at the lasing wavelength, respectively. From Fig. S6, we can see that the fluorescence spectrum and hence $\sigma_e(\lambda)$ of ICG in LDL are stronger than and red-shifted with respect to those of ICG in BSA, whereas the corresponding absorption spectra and hence $\sigma_a(\lambda)$ do not change. Although it is difficult to accurately measure the actual emission cross section of ICG-LDL and ICG-BSA at their lasing wavelength (~ 920 nm), the emission cross section of ICG-LDL is about 10 times higher than that of ICG-BSA at 860 nm. Therefore, the γ value is about 10 times lower for ICG+LDL than for ICG-BSA at 860 nm. We expect that this difference in the γ value still holds for both samples at 920 nm, which explains the ~ 10 times difference in the lasing threshold between the two samples.

Supplementary References

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