Electronic Supplementary Information

for

Supramolecular color-tunable photoluminescent materials based on a chromophore cascade as security inks with dual encryption

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1. General methods and materials

All the reagents and solvents were commercially available and used as received unless otherwise specified purification. Thioflavin T (**ThT**) and nile red (**NiR**) were purchased from Sigma-Aldrich. 5,11,17,23,29-pentacarboxylic acid-31,32,33,34,35-n-dodecyloxy calix[5]arene (**CA**), amphiphilic *β*-cyclodextrin (**CD**), 4-(dodecyloxy)benzamido-terminated methoxy poly(ethylene glycol) (**PEG-C12**) and 2-(4-phenylboronic acid)-1-pyrenemethamide (**PB**) were synthesized and purified according to the literature procedures.[1,2,3,4] The 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES) buffer solution (10 mM) of pH 8.0 was prepared by dissolving 2.383 g of HEPES in approx. 900 ml of double-distilled water. Titrate to pH 8.0 at the lab temperature of 25 °C with NaOH and make up volume to 1000 ml with double-distilled water. The pH value of the buffer solution was then verified on a pH-meter calibrated with two standard buffer solutions.

Preparation of the **CD/CA** co-assembly: **CD** and **CA** were dissolved in chloroform at a concentration of 1.0 mM, respectively. A mixed chloroform solution of **CD** and **CA** (1:1 molar ratio) was dried by slow rotary evaporation to yield a thin film in a glass vial. Residual organic solvent was removed under high vacuum. HEPES buffer (10 mM, pH 8.0) was added and the solution was sonicated at 50 °C for 2 h to make the **CD/CA** co-assembly.

UV-Vis spectra were recorded in 1 mL low-volume disposable PMMA cuvettes (Brand GmbH & CO KG, Wertheim) on a JASCO V-650 double-beam spectrophotometer (JASCO Labor- and Datentechnik GmbH, Gross-Umstadt) at 25 °C. Steady-state fluorescence spectra were recorded in 1 mL low-volume disposable PMMA cuvettes (Brand GmbH & CO KG, Wertheim) on a JASCO FP 6500 and a conventional quartz cell (light path 10 mm) on a Varian Cary Eclipse equipped with a Varian Cary single-cell peltier accessory to control temperature, all the peaks caused by second-order transmission have been eliminated by derivation of original peaks. Fluorescence lifetimes were recorded on a FluoTime300 spectrometer from PicoQuant equipped with two emission monochromators (Czerny-Turner, selectable gratings blazed at 500 nm with 2.7 nm/mm dispersion and 1200 grooves/mm, or blazed at 1250 nm with 5.4 nm/mm dispersion and 600 grooves/mm), Glan-Thompson polarizers for emissions, PMA Hybrid 40 (transit time spread FWHM<120 ps, 300-720 nm) as detector and used in TCSPC mode by a PicoHarp 300 (minimum base resolution of 4 ps). Lifetime analysis was performed using the commercial FluoFit software. The quality of the fit was assessed by minimizing the reduced chi-squared function (χ^2) and visual inspection of the weighted residuals and their autocorrelation. Dynamic light scattering (DLS) measurements were examined on a laser light scattering spectrometer (BI-200SM) equipped with a digital correlator (Turbo Corr.) at 636 nm at a

scattering angle of 90°. Scanning electron microscopy (SEM) images were recorded on a Hitachi S-3500N scanning electron microscope. The samples for SEM measurements were prepared by dropping the solution onto a coverslip, followed by evaporating the liquid in air. Differential scanning calorimetry (DSC) measurements were carried out on a DSC Q100 (TA Instruments). The co-assembly solutions (50 μL) were sealed in pressure-resistant DSC cups and scanned against a reference cup of HEPES buffer (pH = 8.0, 10 mM). The samples were equilibrated at 5 °C, then heated to 80 °C at 1 °C/min. After cooling the sample to room temperature, the measurements were repeated at least five times. The molecular packing model was performed by molecular dynamic simulation using MMFF94 force field.

Ink-writing tests were performed as follows: The aqueous solutions of the **CD**/**CA** co-assembly with three chromophores as fluorescent inks were taken by pipette (Thermo, Scientific) and written on the ordinary filter paper. After drying in air, the written papers were photographed or cut into appropriate size and inserted into a conventional quartz cell (light path 10 mm) for solid-state fluorescence measurement.

2. Chemical structures of the employed compounds

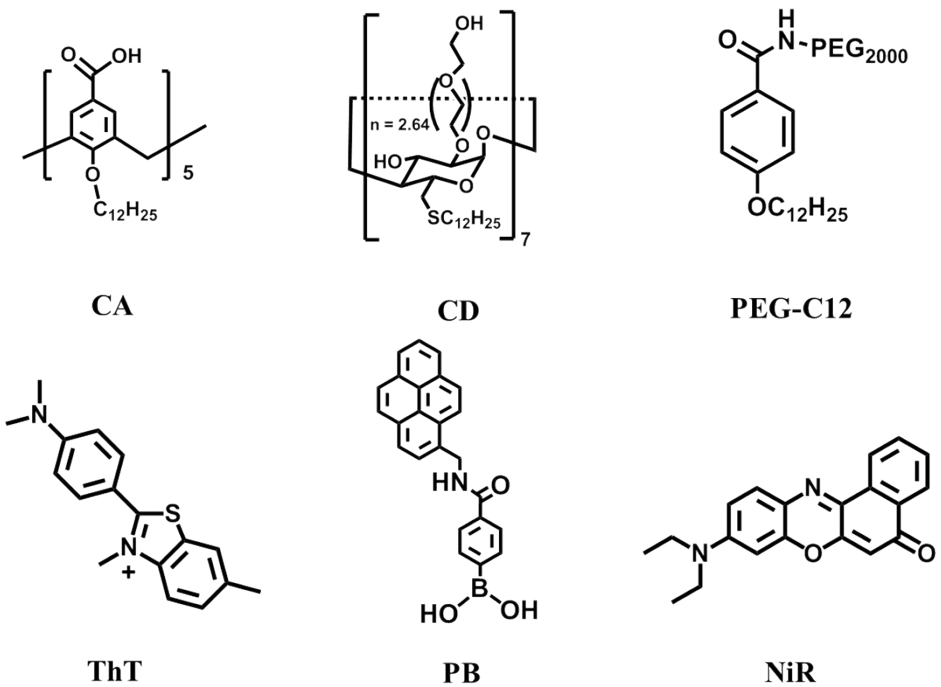

Figure S1. Chemical structures of **CA**, **CD**, **PEG-C12**, **ThT**, **PB** and **NiR**.

3. Characterization of the CD/CA co-assembly

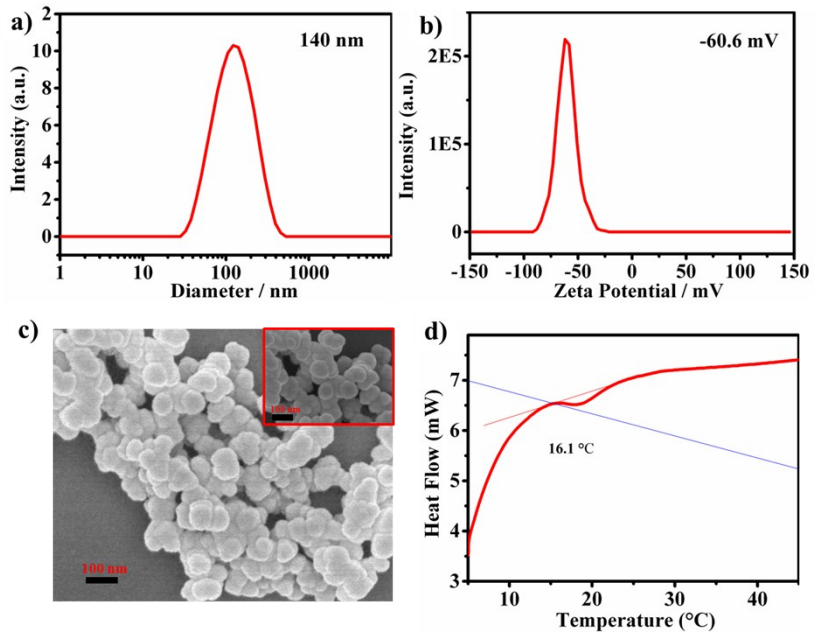

Figure S2. (a) DLS data, (b) Zeta potential, and (c) SEM image of the **CD/CA** co-assembly in 10 mM HEPES buffer (pH = 8.0, [**CA**] = 100 μM, [**CD**] = 100 μM). (d) DSC heating curve for phase transition temperature measurement in 10 mM HEPES buffer (pH = 8.0, [**CA**] = 2 mM, [**CD**] = 2 mM).

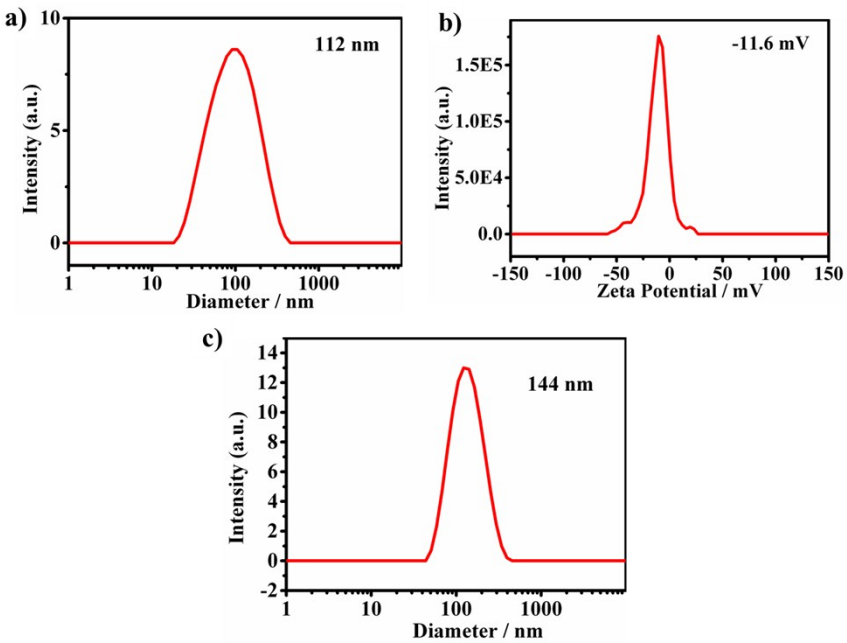

Figure S3. (a) DLS data and (b) Zeta potential of the **CD** assembly in 10 mM HEPES buffer (pH = 8.0, [**CD**] = 100 μM); (c) DLS data of the **CD/CA** co-assembly in 10 mM HEPES buffer (pH = 8.0, [**CA**] = 100 μM, [**CD**] = 100 μM) standing for 13 days.

Due to their characteristic amphiphilic features, **CA** and **CD** form co-assemblies in aqueous solution. It should be mentioned that the alone **CA** cannot be hydrated well in HEPES buffer (10 mM pH = 8.0), but can be done when mixing with **CD**, indirectly proving the formation of co-assemblies. Direct evidences came from the DLS and Zeta potential measurements. The averaged diameter of the **CD**/**CA** co-assembly is about 140 nm (Figure S2a), bigger than that of the **CD** assembly (about 112 nm) (Figure S3a). The **CD**/**CA** co-assembly gives a Zeta potential of about −60.6 mV (Figure S2b), much more negative than free **CD** (−11.6 mV) (Figure S3b), since **CA** possesses negatively charged carboxyl groups at its upper rim. SEM image of the **CD**/**CA** co-assembly shows the spherical-like morphology with comparable size to DLS results (Figure S2c). DSC results show the phase transition temperature (only exited for vesicles rather than micelles) of 16.1 °C for the **CD**/**CA** co-assembly (Figure S2d), much higher than the previously reported **CD** vesicle whose phase transition temperature is too low to be measured.[5] These combined results indicate that **CA** and **CD** co-assembled into the vesicular aggregation.

4. Host-guest complexation and complexation-induced fluorescence enhancement

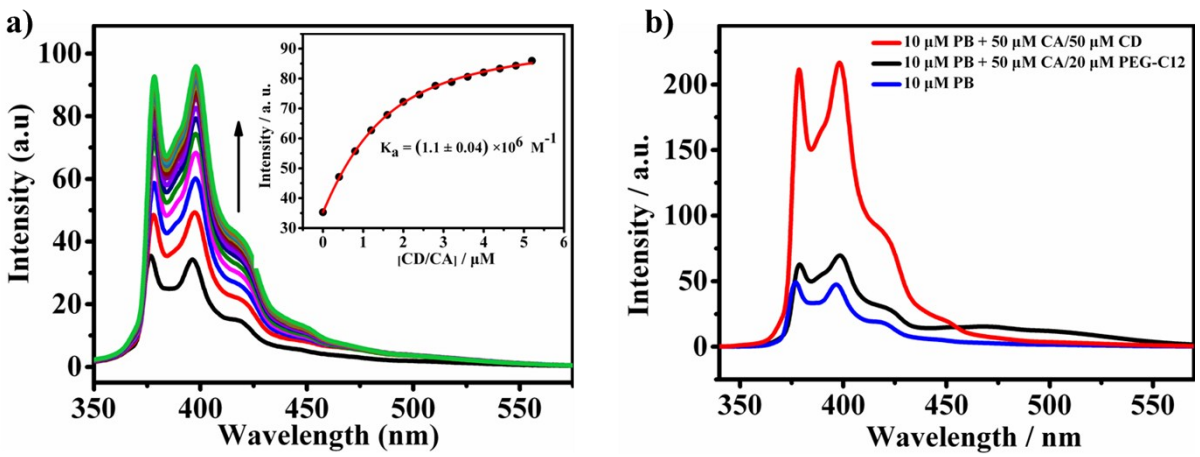

Figure S4. a) Host-guest fluorescence titration of **PB** (1 μM) with the **CD**/**CA** co-assembly in HEPES buffer (10 mM, pH = 8.0), λ_{ex} = 326 nm, inset showing the curve fitting data at λ_{em} = 377 nm; b) Fluorescence spectra of **PB** (10 μM), **PB** (10 μM) with **PEG-C12** (20 μM) and **CA** (50 μM), **PB** (10 μM) with **CD** (50 μM) and **CA** (50 μM) in HEPES buffer (10 mM, pH = 8.0) , λ_{ex} = 326 nm.

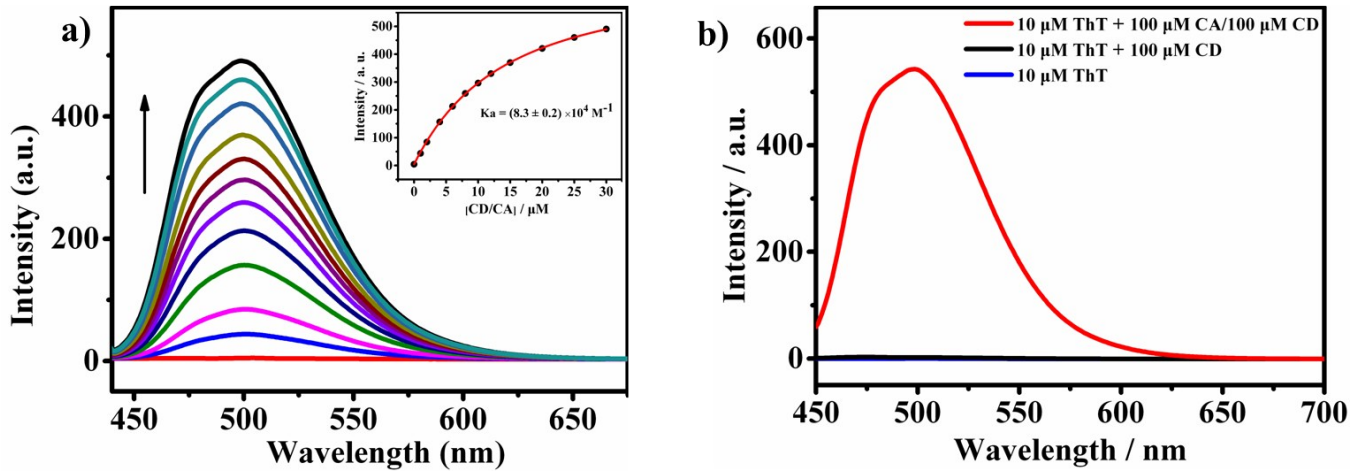

Figure S5. a) Host-guest fluorescence titration of **ThT** (5 μM) with the **CD**/**CA** co-assembly in HEPES buffer (10 mM, pH = 8.0), λ_{ex} = 430 nm, inset showing the curve fitting data at λ_{em} = 500 nm; b) Fluorescence spectra of **ThT** (10 μM) with **CD** (100 μM), **CD** (100 μM) and **CA** (100 μM) in HEPES buffer (10 mM, pH = 8.0), λ_{ex} = 430 nm.

Upon complexation with the **CD/CA** co-assembly, the fluorescence of **PB** was enhanced for 5 times, that of **ThT** was enhanced for 285 times. However, no appreciable enhancement was observed when adding **CA** to **PB**, and adding **CD** to **ThT** (Figures S4b and S5b). These results imply that **PB** was selectively encapsulated into the cavity of **CD**, while **ThT** was selectively encapsulated into the cavity of **CA**. Herein, we should mention that **PEG-C12** was used to solubilize the **CA** assembly (free **CA** cannot be hydrated well as mentioned above), to perform the fluorescence measurement between **PB** and **CA** (Figure S4b).

5. Energy transfer (ET) between dyes

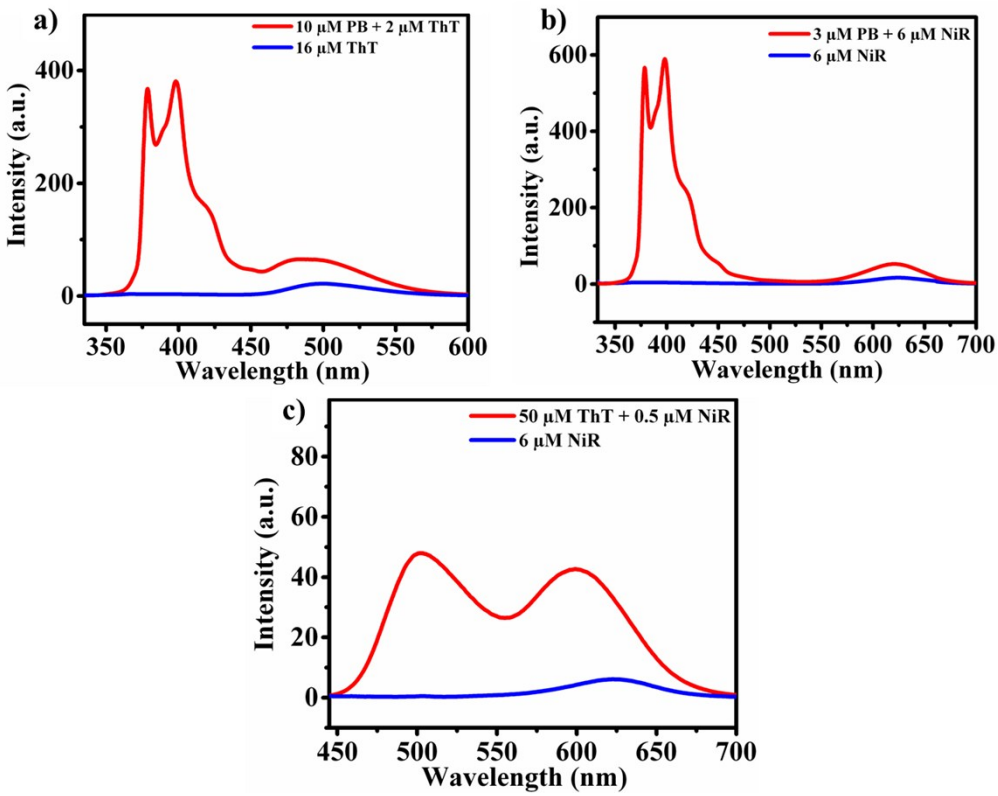

Figure S6. Fluorescence spectra of a) **PB**/**ThT** (10/2 μM) and **ThT** (16 μM), λ_{ex} = 326 nm; b) **PB**/**NiR** (3/6 μM) and **NiR** (6 μM), λ_{ex} = 326 nm; c) **ThT**/**NiR** (50/0.5 μM) and **NiR** (6 μM), λ_{ex} = 430 nm in the **CD**/**CA** co-assembly ([**CD**] = [**CA**] = 50 μM) in HEPES buffer (pH = 8.0, 10 mM).

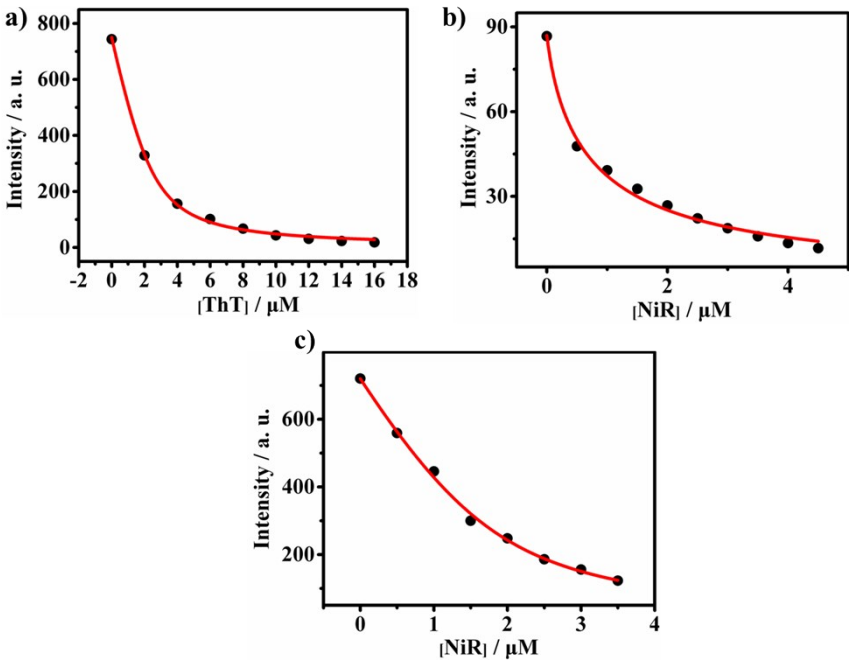

Figure S7. The nonlinear fluorescence changes of a) 10 μM **PB** upon gradual addition of **ThT** (0, 2, 4, 6, 8, 10, 12, 14, 16 μM), λ_{ex} = 326 nm, λ_{em} = 377 nm; b) 50 μM **ThT** upon gradual addition of **NiR** (0.0, 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 4.0, 4.5 μM), λ_{ex} = 430 nm, λ_{em} = 500 nm; c) 10 μM **PB** upon gradual addition of **NiR** (0.0, 0.5, 1.0, 2.0, 3.0, 4.0, 5.0, 6.0 μM), λ_{ex} = 326 nm, λ_{em} = 377 nm. All experiments were performed in the presence of the **CD**/**CA** co-assembly ([**CD**] = [**CA**] = 50 μM) in HEPES buffer (10 mM, pH = 8.0).

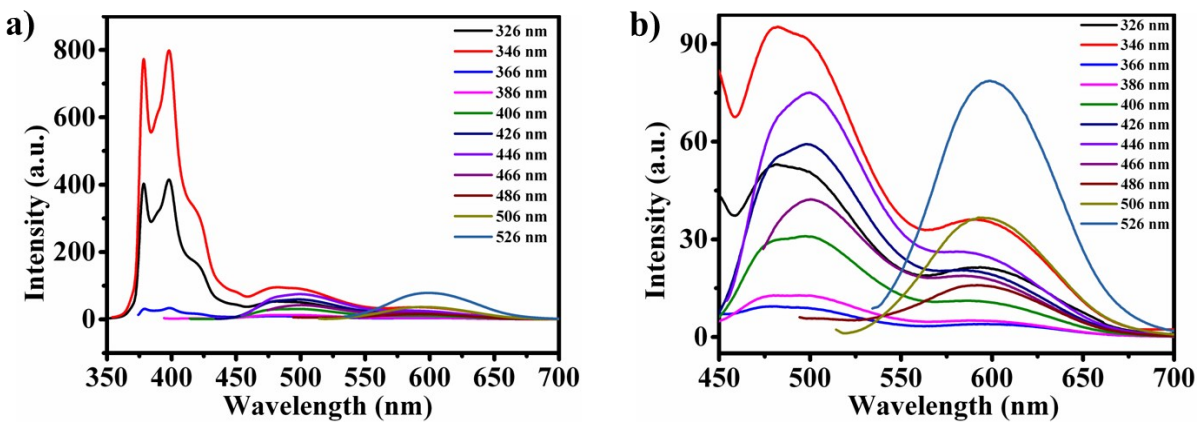

Figure S8. (a) Fluorescence spectra of 10 μM **PB**, 2 μM **ThT** and 0.5 μM **NiR** in the **CD** /**CA** co-assembly ([**CD**] = [**CA**] = 50 μM) in HEPES buffer (10 mM, pH = 8.0) upon excitation with different wavelengths (326, 346, 366, 386, 406, 426, 446, 466, 486, 506, 526 nm). (b) showing the enlarged fluorescence spectra from 450 to 700 nm.

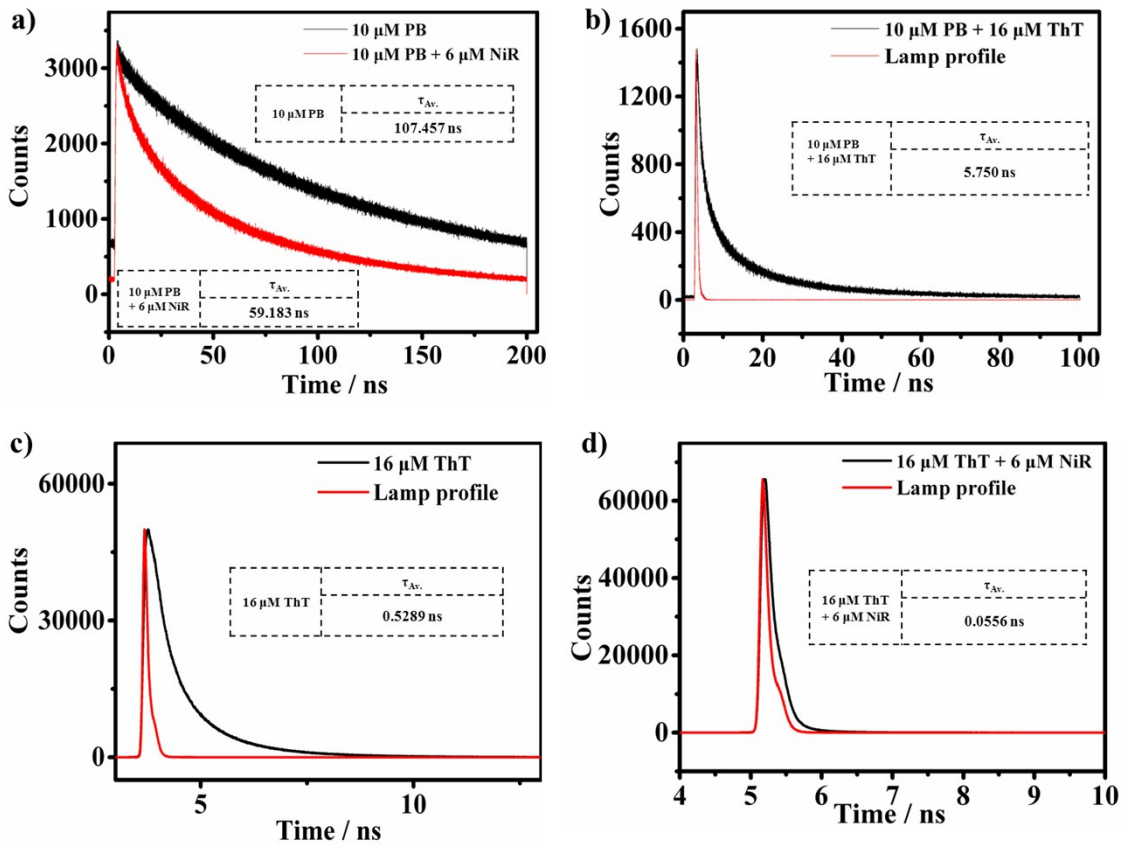

Figure S9. Fluorescence decay profiles of a) 10 μM **PB** and 10 μM **PB** with 6 μM **NiR**, λ_{ex} = 310 nm, $\lambda_{detection}$= 378 nm; b) 10 μM **PB** with 16 μM **ThT**, λ_{ex} = 310 nm, $\lambda_{detection}$= 378 nm; c) 16 μM **ThT**, λ_{ex} = 410 nm, $\lambda_{detection}$= 500 nm; d) 16 μM **ThT** with 6 μM **NiR**, λ_{ex} = 410 nm, $\lambda_{detection}$= 500 nm in the **CD /CA** co-assembly ([**CD**] = [**CA**] = 50 μM) in HEPES buffer (10 mM, pH = 8.0). Note: The value of $\tau_{Av.}$ is an amplitude-weighted average lifetime.[6]

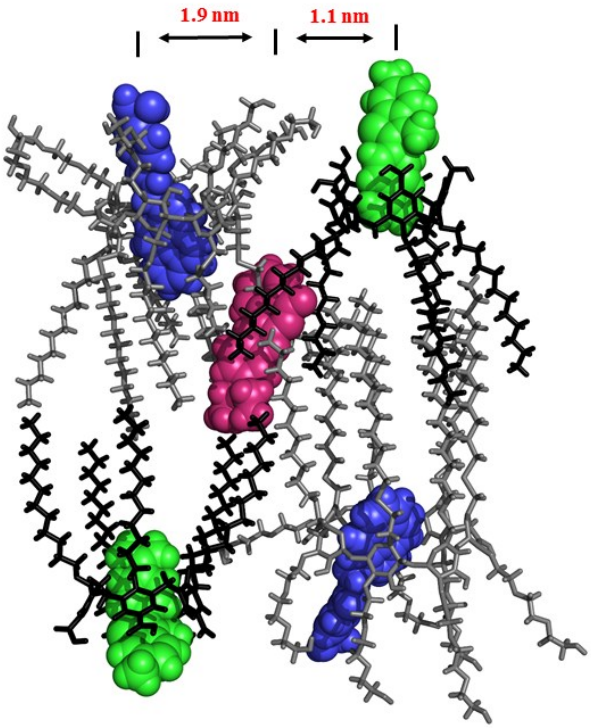

Figure S10. Distance and orientation estimates for donors and acceptors in the **CD**/**CA** co-assembly. **CD** is shown in grey, **CA** in black, **ThT** in green, **PB** in blue and **NiR** in red.

6. Ink-writing tests

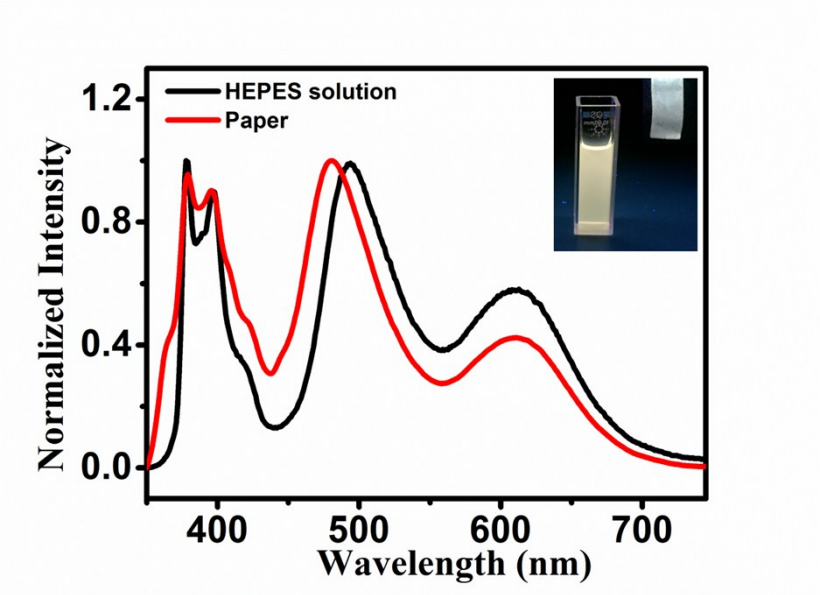

Figure S11. Fluorescence spectra of 20 μM **PB**, 32 μM **ThT** and 1 μM **NiR** in the **CD**/**CA** co-assembly ([**CD**] = [**CA**] = 100 μM) in HEPES buffer (10 mM, pH = 8.0, black line) and in paper (red line), λ_{ex} = 326 nm, inset showing the photographs under 365 nm UV light.

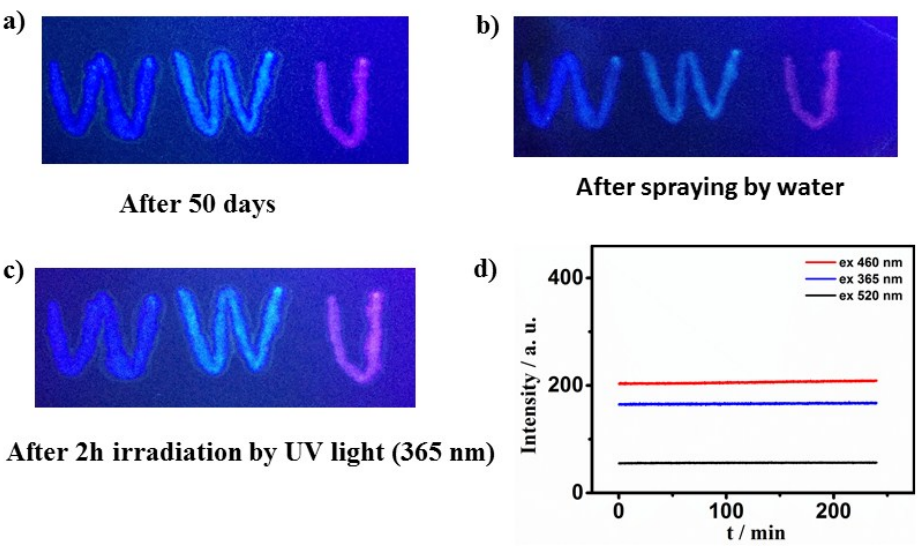

Figure S12. Stability test of the fluorescent inks: photographs of the written paper a) kept in dark for 50 days; b) sprayed by water; c) irradiated for 2 h under UV light (365 nm, 14.6 mJ/cm^2); d) photobleaching measurements of the fluorescent inks in HEPES solution, irradiated by 365 nm (blue line), 460 nm (red line), 520 nm (black line), λ_{em} = 615 nm.

7. References

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