Supplementary Information

A simple yet effective AIE-based fluorescent nano-thermometer for temperature mapping in living cells using fluorescence lifetime imaging microscopy

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Sample preparation for Figures S2B and S3

HPS was dissolved in THF first at a final concentration of 0.2 mg/mL. 0.5 mL of HPS in THF solution was then poured into 5 mL Milli-Q water under sonication expeditiously for 4 min using an ultrasonic probe sonicator at 15 W output. After sonication, the solution was stirred for several hours to remove THF by evaporation.

Experimental details for Figure S3

A thermostat Water Bath (TENLIN, DC-0506, China) and a digital thermocouple (LINI-T UT325 Thermometer) were used to control the solution temperature during the measurement of fluorescence emission by FLS-55 spectrometer. Generally, 2.5 mL of solution was introduced into a quartz cuvette, and the corresponding fluorescence spectra of the solution were recorded at different temperatures.



Figure S1. (A) Size distribution and (B) zeta potential of the HPS/Butter/DSPE-PEG-Biotin nanorod examined by DLS in water. Average hydrodynamic diameter (<d>) was around 160 nm at room temperature. Average zeta potential was around -50 mV at room temperature.



Figure S2. UV-vis absorption spectra of (A) the Butter/DSPE-PEG-Biotin, HPS/Butter/DSPE-PEG-Biotin nanorod and (B) HPS aggregates at room temperature.



Figure S3. Fluorescence spectra of HPS aggregates in different temperatures. (A) Increased from 20 to 60 °C. (B) Decreased from 60 to 20 °C. Sample solutions were excited at 375 nm. Insets show the fluorescence intensity at 490 nm at different temperatures normalized to the corresponding peak fluorescence intensity at 20 °C. The sample concentration depended on HPS is same with the sample shown in Figure 2.



Figure S4. Fluorescence spectra of the Butter/DSPE-PEG-Biotin nanorod upon a temperature change between 20 and 60 °C. Sample solutions were excited at 375 nm. The sample concentration of butter and DSPE-PEG-Biotin is same with the sample shown in Figure 2.



Figure S5. Thermal reversibility of the peak fluorescence ratio of HPS/Butter/DSPE-PEG-Biotin nanorod in a single warming and cooling cycle. Individual fluorescence intensity at 490 nm at different temperatures is normalized to that at 20 °C of corresponding warming or cooling cycle.



Figure S6. Representative reversibility of the normalized peak fluorescence intensity of the HPS/Butter/DSPE-PEG-Biotin nanorod to temperature variations. Sample solutions were excited at 375 nm. Fluorescence intensity in different conditions was normalized to the first measurement at 20 °C.



Figure S7. Fluorescence lifetime images of the HPS/Butter/DSPE-PEG-Biotin nanorod dispersed in ultrapure water. Fluorescence lifetimes are displayed in a pseudocolor format. The excitation wavelength was 375 nm.



Figure S8. Representative reversibility of the fluorescence lifetime of the HPS/Butter/DSPE-PEG-Biotin nanorod to temperature variations. The excitation wavelength was 375 nm.



Figure S9. Cell viability of the HPS/Butter/DSPE-PEG-Biotin nanorod determined by the CCK-8 assay.