Supplementary Information for:

Coumarin 545: an emission reference with a record-low temperature coefficient for ratiometric fluorescence based temperature measurements

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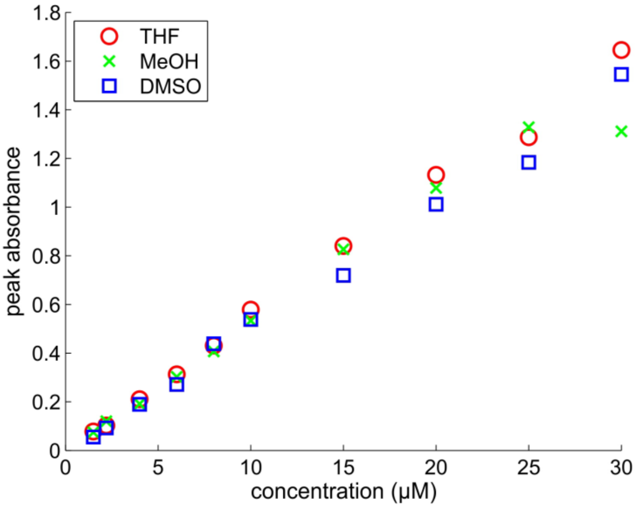

Figure S1. The peak absorbance of **2** as a function of solution concentration in THF, methanol and DMSO, respectively. The molar extinction coefficients have been calculated to be 5.8×10^4, 5.3×10^4 and 5.7×10^4 L mol^{-1} cm^{-1}, in THF, methanol and DMSO, respectively, based on the absorbance data in dilute samples (≤ 10 μM).

Table S1. Summary of Fitting Parameters to the Single-Exponential Fluorescence Decay Dynamics of **2** in methanol.[a]

λ_{em} (nm)	τ_1 (ns)	χ^2
450	2.71	1.19
460	2.72	1.116
470	2.73	1.152
480	2.76	1.166
490	2.83	1.204
500	2.92	1.148
510	3.03	1.201
520	3.07	1.155
530	3.10	1.213
540	3.09	1.044
550	3.11	1.191
560	3.11	1.122
570	3.13	1.167
580	3.12	1.15
590	3.14	1.161
600	3.13	1.176

[a] The fluorescent decay dynamics of **2** can be well modeled by a single-exponential decay function, i.e., $A_1 \exp\left(-\frac{t}{\tau_1}\right) + B$.

Table S2. Summary of Revised Fitting Parameters to the Fluorescence Decay Dynamics of **2** in methanol.[a]

λ_{em} (nm)	A_1	τ_1 (ns)	A_2	τ_2 (ns)	χ^2	$\frac{A_1\tau_1}{A_1\tau_1 + A_2\tau_2}$	$\frac{A_2\tau_2}{A_1\tau_1 + A_2\tau_2}$
450	-	2.71	-	-	1.19	100%	0
460	2975	2.71	51.287	3.13	1.117	98.0%	2.0%
470	2853.862	2.71	158.19	3.13	1.152	94.0%	6.0%
480	2744.669	2.71	360.31	3.13	1.165	86.8%	13.2%
490	2128.417	2.71	752.248	3.13	1.202	71.0%	29.0%
500	1524.769	2.71	1422.504	3.13	1.147	48.1%	51.9%
510	787.691	2.71	2390.385	3.13	1.208	22.2%	77.8%
520	468.873	2.71	2600.898	3.13	1.159	13.5%	86.5%
530	262.115	2.71	2779.625	3.13	1.214	7.5%	92.5%
540	293.258	2.71	2579.187	3.13	1.044	9.0%	91.0%
550	182.338	2.71	2897.854	3.13	1.192	5.2%	94.8%
560	155.043	2.71	2882.276	3.13	1.123	4.5%	95.5%
570	-	-	-	3.13	1.167	0	100%
580	-	-	-	3.12	1.15	0	100%
590	-	-	-	3.14	1.161	0	100%
600	-	-	-	3.13	1.176	0	100%

[a] The fluorescent decay dynamics of **2** in the transitional region is modelled by a double-exponential decay function, i.e., $A_1 \exp\left(-\frac{t}{\tau_1}\right) + A_2 \exp\left(-\frac{t}{\tau_2}\right) + B$, owing to both the H-aggregate and monomer emissions. The steady-state contributions of these two emission components are calculated using $\frac{A_1\tau_1}{A_1\tau_1+A_2\tau_2}$ and $\frac{A_2\tau_2}{A_1\tau_1+A_2\tau_2}$, respectively.

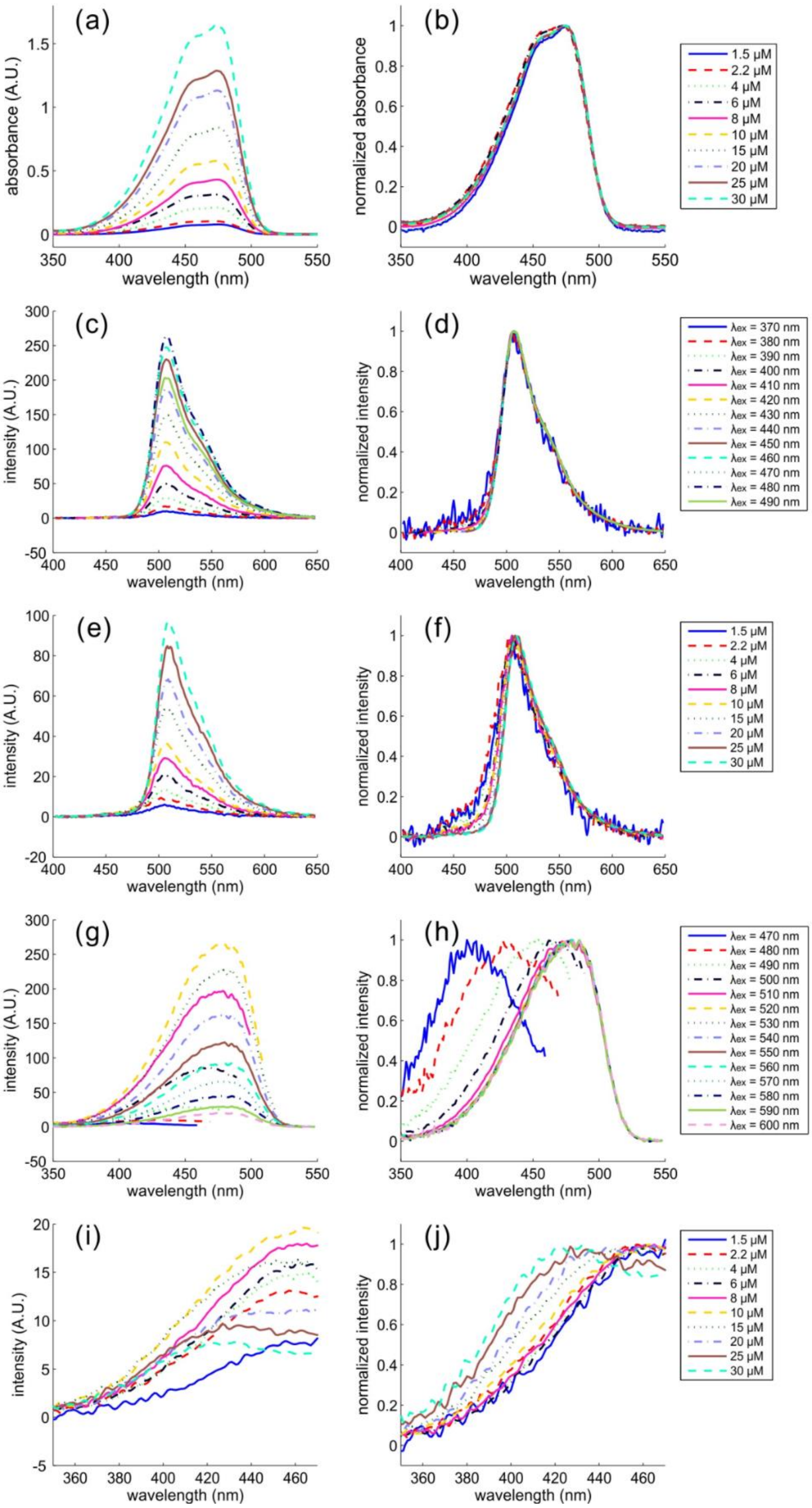

Figure S2. (a) UV—vis absorption spectra of **2** in THF at various concentrations from 1.5 to 30 μM; (c) fluorescence spectra of **2** ([**2**] = 8 μM) in THF, excited at various wavelengths from 370 to 490 nm; (e) fluorescence spectra of **2** in THF at various concentrations from 1.5 to 30 μM, excited at 390 nm; (g) fluorescence excitation spectra of **2** ([**2**] = 8 μM) in THF, monitored at various emission wavelengths from 470 to 600 nm; (i) fluorescence excitation spectra of **2** in THF at various concentrations from 1.5 to 30 μM, monitored at the emission wavelength of 480 nm. (b), (d), (f), (h) and (j) correspond to the normalized spectra of (a), (c), (e), (g) and (i), respectively.

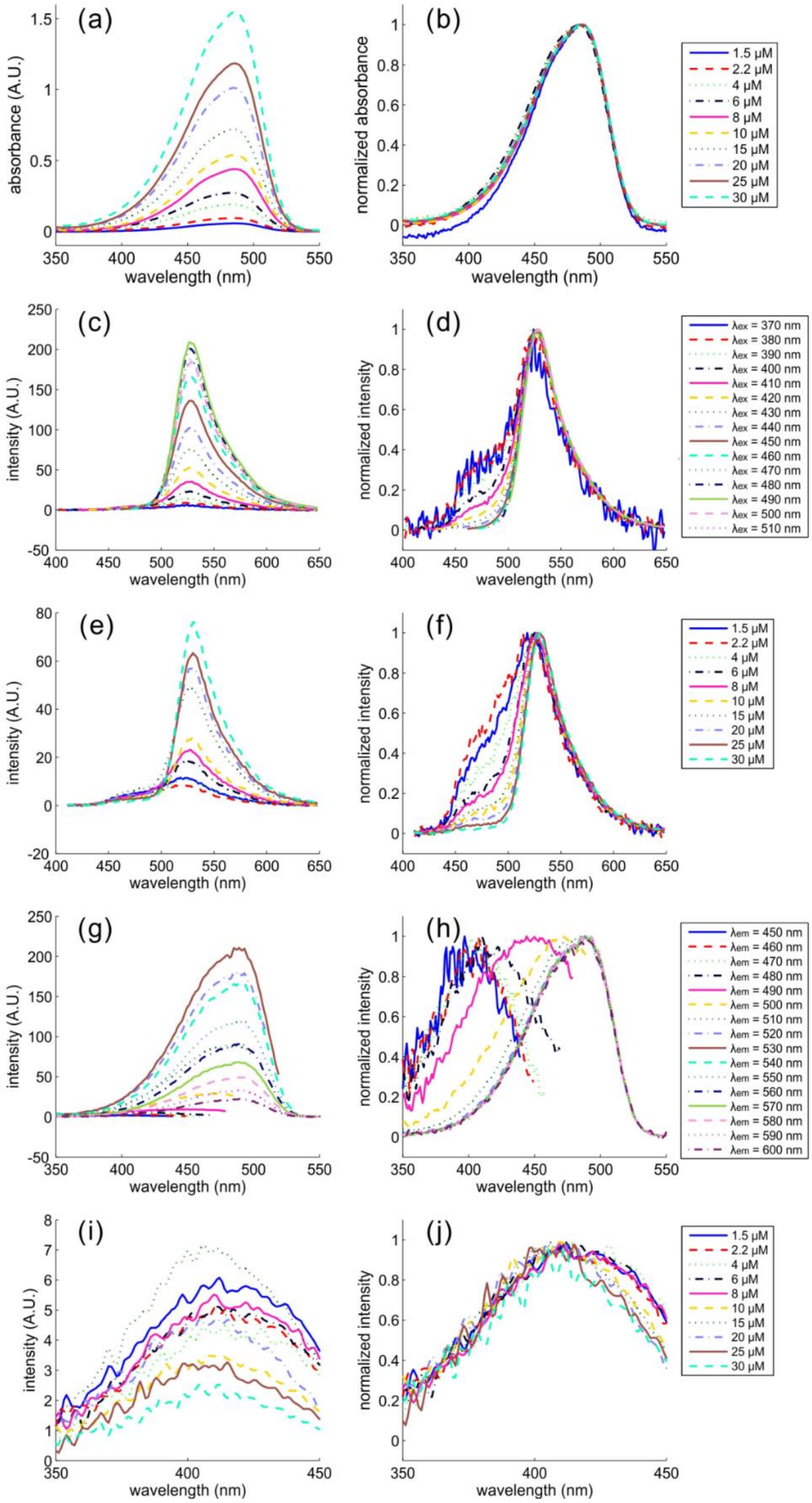

Figure S3. (a) UV—vis absorption spectra of **2** in DMSO at various concentrations from 1.5 to 30 μM; (c) fluorescence spectra of **2** ([**2**] = 8 μM) in DMSO, excited at various wavelengths from 370 to 510 nm; (e) fluorescence spectra of **2** in DMSO at various concentrations from 1.5 to 30 μM, excited at 400 nm; (g) fluorescence excitation spectra of **2** ([**2**] = 8 μM) in DMSO, monitored at various emission wavelengths from 450 to 600 nm; (i) fluorescence excitation spectra of **2** in DMSO at various concentrations from 1.5 to 30 μM, monitored at the emission wavelength of 480 nm. (b), (d), (f), (h) and (j) correspond to the normalized spectra in (a), (c), (e), (g) and (i), respectively.

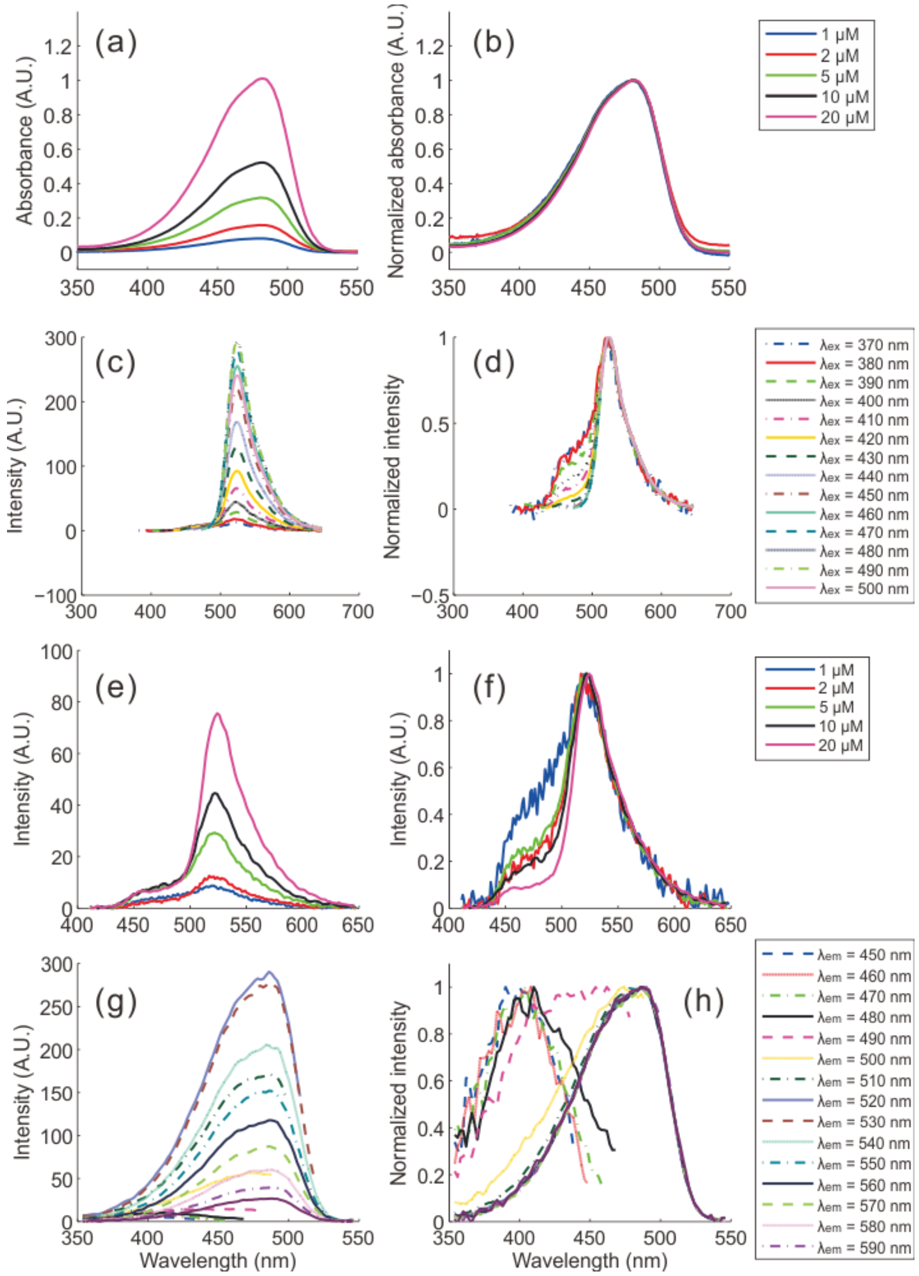

Figure S4. (a) UV—vis absorption spectra of **2** in DMF at various concentrations from 1 to 20 μM; (c) fluorescence spectra of **2** ([**2**] = 10 μM) in DMF, excited at various wavelengths from 370 to 500 nm; (e) fluorescence spectra of **2** in DMF at various concentrations from 1 to 20 μM, excited at 400 nm; (g) fluorescence excitation spectra of **2** ([**2**] = 10 μM) in DMF, monitored at various emission wavelengths from 450 to 590 nm;. (b), (d), (f), and (h) correspond to the normalized spectra in (a), (c), (e), and (g), respectively.

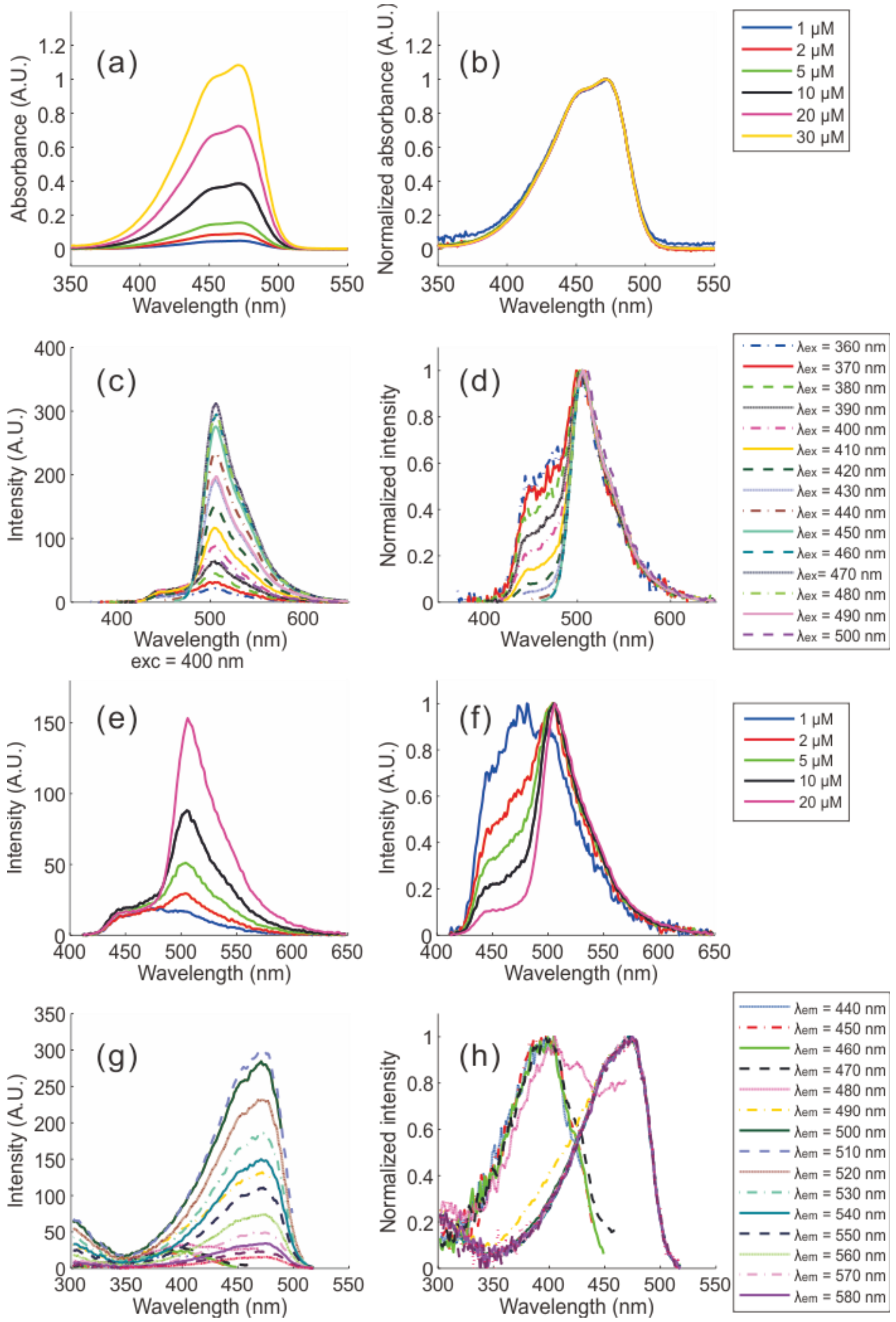

Figure S5. (a) UV—vis absorption spectra of **2** in EA at various concentrations from 1 to 30 μM; (c) fluorescence spectra of **2** ([**2**] = 10 μM) in EA, excited at various wavelengths from 360 to 500 nm; (e) fluorescence spectra of **2** in EA at various concentrations from 1 to 20 μM, excited at 400 nm; (g) fluorescence excitation spectra of **2** ([**2**] = 10 μM) in EA, monitored at various emission wavelengths from 440 to 580 nm;. (b), (d), (f), and (h) correspond to the normalized spectra in (a), (c), (e), and (g), respectively.

Table S3. The fluorescence intensity variations over different temperatures (ΔI) of **2** and the corresponding effective temperature coefficients (C) as a function of excitation wavelength (λ_{ex}) in methanol, THF, DMSO, EA and DMF. The “optimal” excitation wavelengths affording the lowest C values have been highlighted in bold. [**2**] = 8 µM in methanol, THF and DMSO; [**2**] = 10 µM in EA and DMF.

Methanol			THF			DMSO			EA			DMF		
λ_{ex} (nm)	ΔI (10—50 °C)	C (per °C)	λ_{ex} (nm)	ΔI (10—50 °C)	C (per °C)	λ_{ex} (nm)	ΔI (15—55 °C)	C (per °C)	λ_{ex} (nm)	ΔI (15—55 °C)	C (per °C)	λ_{ex} (nm)	ΔI (15—55 °C)	C (per °C)
350	13.928%	0.348%	-	-	-	-	-	-	350	8.510%	0.213%	350	16.486%	0.412%
360	13.194%	0.330%	-	-	-	-	-	-	360	2.650%	0.066%	360	15.541%	0.389%
370	9.629%	0.241%	-	-	-	-	-	-	370	4.828%	0.121%	370	7.560%	0.189%
380	10.076%	0.252%	380	9.751%	0.244%	380	18.454%	0.461%	380	3.869%	0.097%	380	7.614%	0.190%
390	8.567%	0.214%	390	11.407%	0.285%	390	10.329%	0.258%	390	4.526%	0.113%	390	6.583%	0.165%
400	9.633%	0.241%	400	9.933%	0.248%	400	12.411%	0.310%	400	3.389%	0.085%	400	5.170%	0.129%
410	2.534%	0.063%	410	6.141%	0.154%	410	4.633%	0.116%	410	2.605%	0.065%	410	4.134%	0.103%
420	2.264%	0.057%	420	3.517%	0.088%	420	5.060%	0.126%	**420**	**1.159%**	**0.029%**	**420**	**-1.724%**	**-0.043%**
430	**-1.003%**	**-0.025%**	**430**	**1.007%**	**0.025%**	430	2.092%	0.052%	430	-1.948%	-0.049%	430	-2.991%	-0.075%
440	-3.119%	-0.078%	440	-2.574%	-0.064%	**440**	**-1.749%**	**-0.044%**	440	-2.449%	-0.061%	440	-3.103%	-0.078%
450	-7.180%	-0.180%	450	-4.282%	-0.107%	450	-4.368%	-0.109%	450	-6.813%	-0.170%	450	-4.906%	-0.123%
460	-7.853%	-0.196%	460	-6.098%	-0.152%	460	-7.286%	-0.182%	460	-5.852%	-0.146%	460	-7.362%	-0.184%
470	-8.593%	-0.215%	470	-6.880%	-0.172%	470	-7.825%	-0.196%	470	-9.272%	-0.232%	470	-8.285%	-0.207%
480	-13.387%	-0.335%	480	-13.508%	-0.338%	480	-8.233%	-0.206%	480	-15.655%	-0.391%	480	-10.906%	-0.273%
490	-17.596%	-0.440%	490	-24.325%	-0.608%	490	-12.691%	-0.317%	490	-26.168%	-0.654%	490	-13.846%	-0.346%
500	-22.388%	-0.560%	-	-	-	500	-17.747%	-0.444%	-	-	-	-	-	-
510	-22.676%	-0.567%	-	-	-	510	-21.846%	-0.546%	-	-	-	-	-	-

Table S4. The fluorescence intensity variations over different temperatures (ΔI) of **2** and the corresponding effective temperature coefficients (*C*) as a function of excitation wavelength (λ_{ex}) and [**2**] in DMF. The “optimal” excitation wavelengths affording the lowest *C* values have been highlighted in bold.

1 μM			5 μM			20 μM		
λ_{ex} (nm)	ΔI (15—55 °C)	*C* (per °C)	λ_{ex} (nm)	ΔI (15—55 °C)	*C* (per °C)	λ_{ex} (nm)	ΔI (15—55 °C)	*C* (per °C)
350	23.899%	0.597%	350	8.561%	0.214%	350	4.207%	0.105%
360	25.983%	0.650%	360	22.010%	0.550%	360	9.989%	0.250%
370	14.072%	0.352%	370	5.782%	0.145%	370	9.632%	0.241%
380	16.760%	0.419%	380	8.743%	0.219%	380	7.340%	0.183%
390	12.432%	0.311%	390	4.685%	0.117%	390	7.581%	0.190%
400	9.194%	0.230%	400	6.757%	0.169%	400	5.591%	0.140%
410	10.897%	0.272%	410	4.334%	0.108%	410	2.731%	0.068%
420	2.220%	0.055%	**420**	**2.667%**	**0.067%**	**420**	**-2.327%**	**-0.058%**
430	**-1.938%**	**-0.048%**	430	-3.051%	-0.076%	430	-2.871%	-0.072%
440	-3.961%	-0.099%	440	-3.095%	-0.077%	440	-3.933%	-0.098%
450	-4.512%	-0.113%	450	-4.742%	-0.119%	450	-5.321%	-0.133%
460	-7.706%	-0.193%	460	-6.783%	-0.170%	460	-6.631%	-0.166%
470	-10.636%	-0.266%	470	-7.178%	-0.179%	470	-7.402%	-0.185%
480	-13.830%	-0.346%	480	-9.569%	-0.239%	480	-7.660%	-0.191%
490	-19.496%	-0.487%	490	-14.619%	-0.365%	490	-9.137%	-0.228%

Table S5. The fluorescence intensity variations over different temperatures (ΔI) of **2** and the corresponding effective temperature coefficients (*C*) as a function of excitation wavelength (λ_{ex}) and [**2**] in DMSO. The “optimal” excitation wavelengths affording the lowest *C* values have been highlighted in bold.

2 µM			5 µM			10 µM		
λ_{ex} (nm)	ΔI (20—65 °C)	*C* (per °C)	λ_{ex} (nm)	ΔI (20—65 °C)	*C* (per °C)	λ_{ex} (nm)	ΔI (20—65 °C)	*C* (per °C)
380	10.841%	0.241%	380	10.038%	0.223%	380	10.190%	0.226%
390	12.967%	0.288%	390	9.021%	0.200%	390	7.310%	0.162%
400	7.959%	0.177%	400	9.133%	0.203%	400	3.626%	0.081%
410	12.343%	0.274%	410	5.774%	0.128%	410	6.625%	0.147%
420	6.530%	0.145%	420	7.066%	0.157%	420	3.982%	0.088%
430	4.810%	0.107%	430	3.315%	0.074%	430	2.642%	0.059%
440	3.698%	0.082%	440	**2.324%**	**0.052%**	**440**	**1.419%**	**0.032%**
450	**2.624%**	**0.058%**	450	2.334%	0.052%	450	3.049%	0.068%
460	4.479%	0.100%	460	5.107%	0.113%	460	5.343%	0.119%
470	6.137%	0.136%	470	5.801%	0.129%	470	6.459%	0.144%
480	9.289%	0.206%	480	8.503%	0.189%	480	8.130%	0.181%
490	14.072%	0.313%	490	13.900%	0.309%	490	11.926%	0.265%
500	18.417%	0.409%	500	18.269%	0.406%	500	16.114%	0.358%
510	17.740%	0.394%	510	18.843%	0.419%	510	19.004%	0.422%

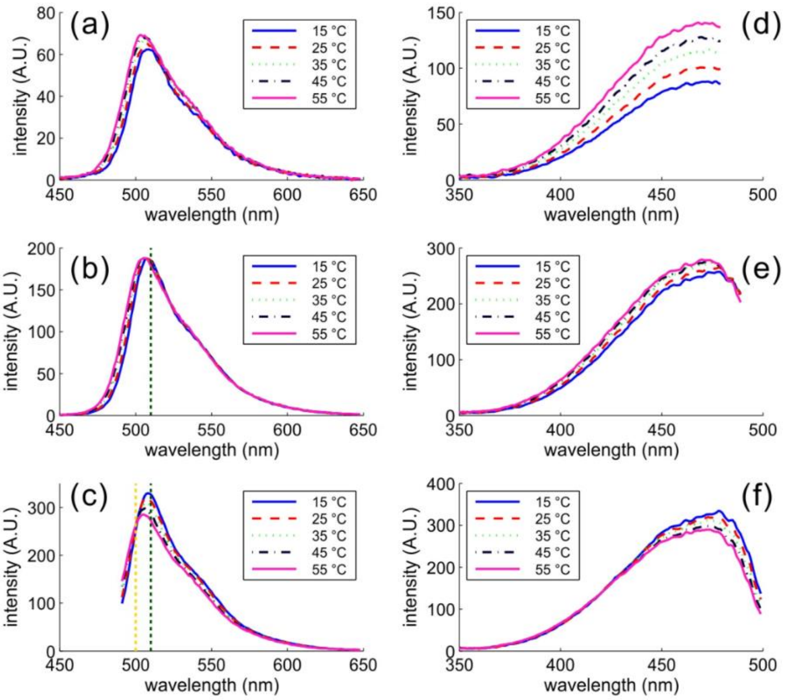

Figure S6. Fluorescence spectra of **2** in THF excited at (a) 400 nm, (b) 430 nm and (c) 480 nm; fluorescence excitation spectra of **2** in THF monitored at the emission wavelengths (d) 490 nm, (e) 500 nm and (f) 510 nm. [**2**] = 8 μM. Three vertical lines at 500 and 510 nm are drawn in (c) and (d) to illustrate the different temperature sensitivities of the corresponding emission intensities.

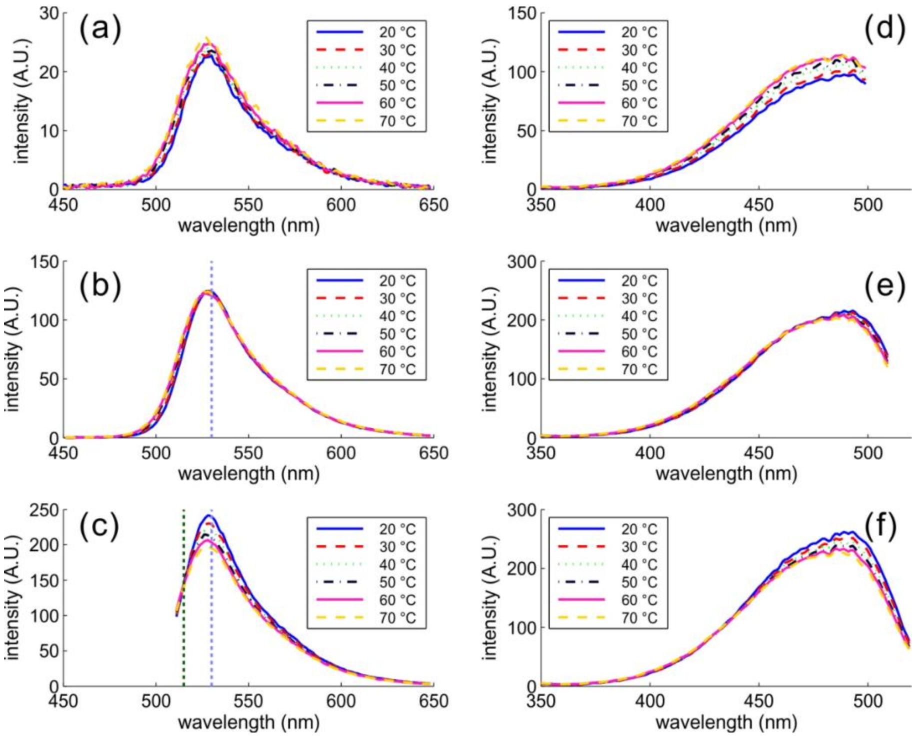

Figure S7. Fluorescence spectra of **2** in DMSO excited at (a) 400 nm, (b) 440 nm and (c) 500 nm; fluorescence excitation spectra of **2** in DMSO monitored at the emission wavelengths (d) 510 nm, (e) 520 nm and (f) 530 nm. [**2**] = 8 μM. Three vertical lines at 515 and 530 nm are drawn in (c) and (d) to illustrate the different temperature sensitivities of the corresponding emission intensities.

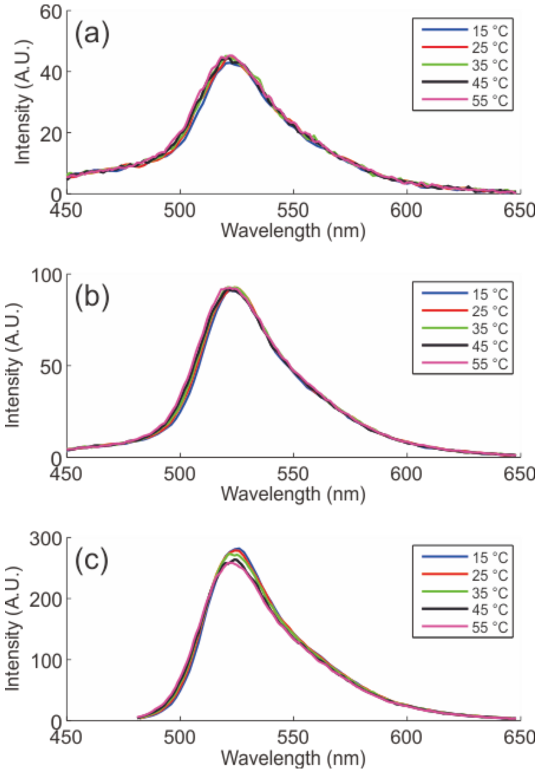

Figure S8. Fluorescence spectra of **2** in DMF excited at (a) 400 nm, (b) 420 nm and (c) 470 nm. [**2**] = 10 μM.

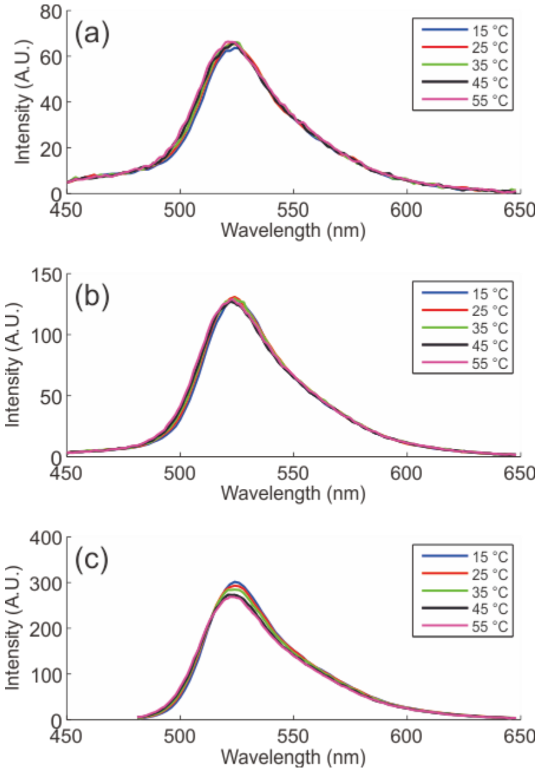

Figure S9. Fluorescence spectra of **2** in EA excited at (a) 400 nm, (b) 420 nm and (c) 470 nm. [**2**] = 10 μM.

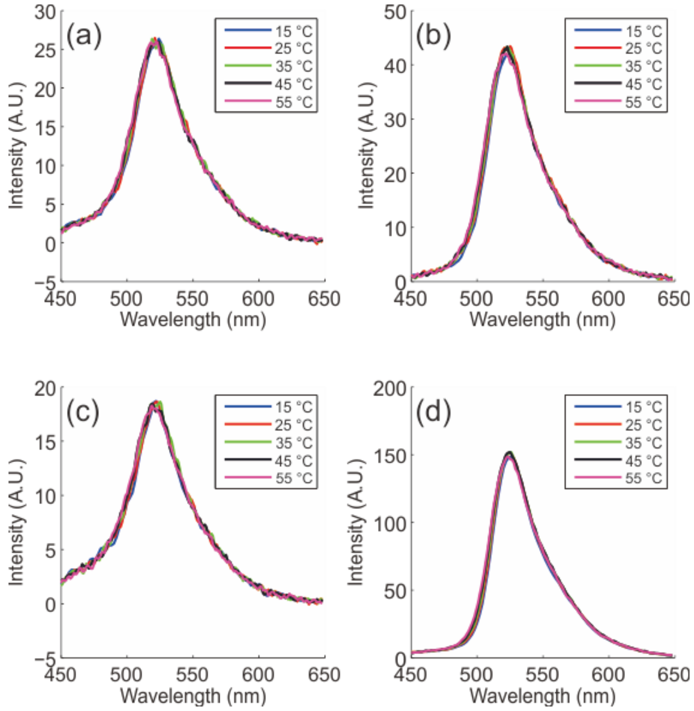

Figure S10. Fluorescence spectra of **2** in DMF excited at (a) 430 nm ([**2**] = 1 μM); (b) 430 nm ([**2**] = 1 μM); (c) 420 nm ([**2**] = 5 μM); (d) 420 nm ([**2**] = 20 μM). The excitation wavelengths are set to the “optimal” value, in order to achieve the lowest temperature coefficients of emission intensity.

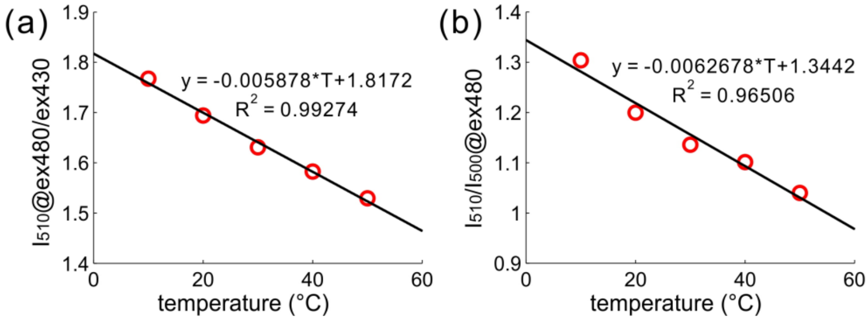

Figure S11. Temperature dependence of (a) the ratios of fluorescence intensities of **2** in THF at 510 nm, excited at 480 and 430 nm, respectively, and the associated best-fit equation; (b) the ratios of fluorescence intensities of **2** in THF at 510 and 500 nm, excited at 480 nm, and the associated best-fit equation. [**2**] = 8 μM.

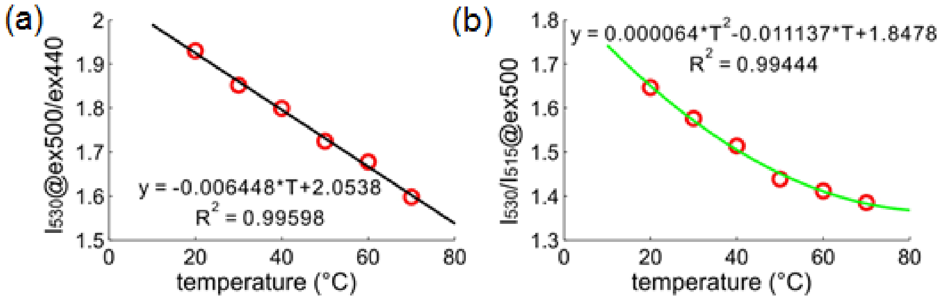

Figure S12. Temperature dependence of (a) the ratios of fluorescence intensities of **2** in DMSO at 530 nm, excited at 500 and 440 nm, respectively, and the associated best-fit equation; (b) the ratios of fluorescence intensities of **2** in DMSO at 530 and 515 nm, excited at 500 nm, and the associated best-fit equation. [**2**] = 8 μM.

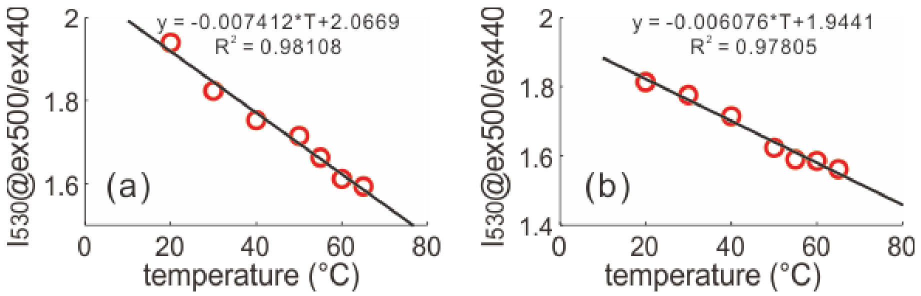

Figure S13. Temperature dependence of the ratios of fluorescence intensities of **2** in DMSO at 530 nm, excited at 500 and 440 nm, respectively, and the associated best-fit equation. (a) [**2**] = 5 μM; (b) [**2**] = 10 μM.