

Supplementary Information – Experimental Data

Title: Robust thin-film fluorescence thermometry for prolonged measurements in microfluidic devices

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S.1 Experimental Apparatus

The heating stage was placed face down on an inverted microscope equipped with a motorized filter turret and camera port as shown in Figure S1. Two filter cubes with different emission filters and identical excitation and dichroic filters were placed in the turret. The filter transmission spectra are shown in the next section. For the initial data collection a filter cube was used without the emission or excitation filter and a spectrophotometer was placed in the camera port. This data was used to determine the optimum bands for the filters.

After the filters were chosen and placed in the system, the spectrometer was removed after characterizing each filter's transmission spectrum. The camera was reinstalled and the image acquisition software was programmed to alternate the filter cubes between images. The temperature information was obtained by post processing the data. A system using two cameras to take images simultaneously could easily generate temperature data in real time.

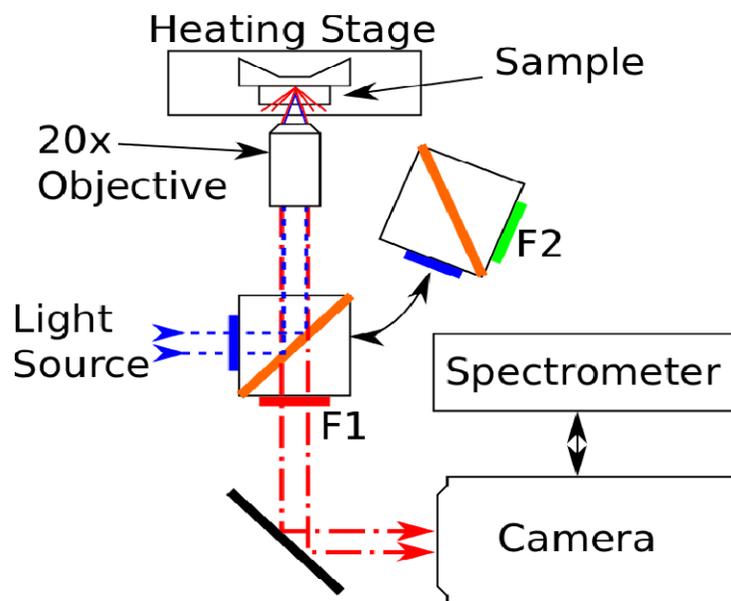


Figure S1. The excitation light passes through the excitation filter (blue) and is reflected off the dichroic mirror (orange). The fluorescent emission light passes through the dichroic mirror, then the emission filter F1 (red) or F2 (green) and is collected at the camera or spectrometer.

S.2 Filter Details

The spectrum for each filter was taken individually, and then compared to the raw (unfiltered emission) spectrum to determine the amount of light passing through the filters. The spectrum data for the BTBP-doped

polystyrene (PS) film was taken integrating over 0.6 seconds and averaged over 10 samples.

Figures S2 - S5 show the emission spectrum as well as the transmission spectra for each filter and polymer. A small amount of laser light makes its way to the spectrometer when the excitation filter is removed. This is a useful indicator of the relative efficiency of the polymer-dye combination. The laser peak is only slightly higher than the highest emission peak for the PS film. With the polymethylmethacrylate (PMMA) film the laser peak is approximately 3 times the highest emission peak. In the polydimethylsiloxane (PDMS) film the laser peak is approximately 6 times the highest emission peak, indicating a less efficient combination.

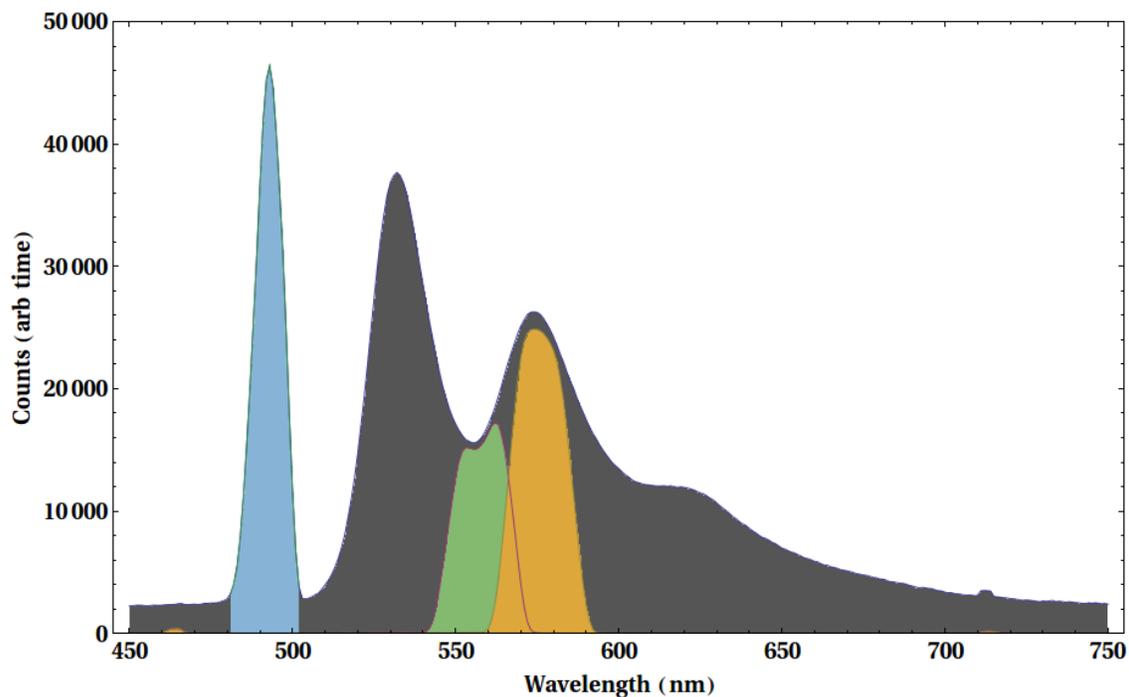


Figure S2. BTBP-doped PS film emission spectrum (gray), transmission spectra of each filter (green, orange) and offset peak of the excitation laser light (blue).

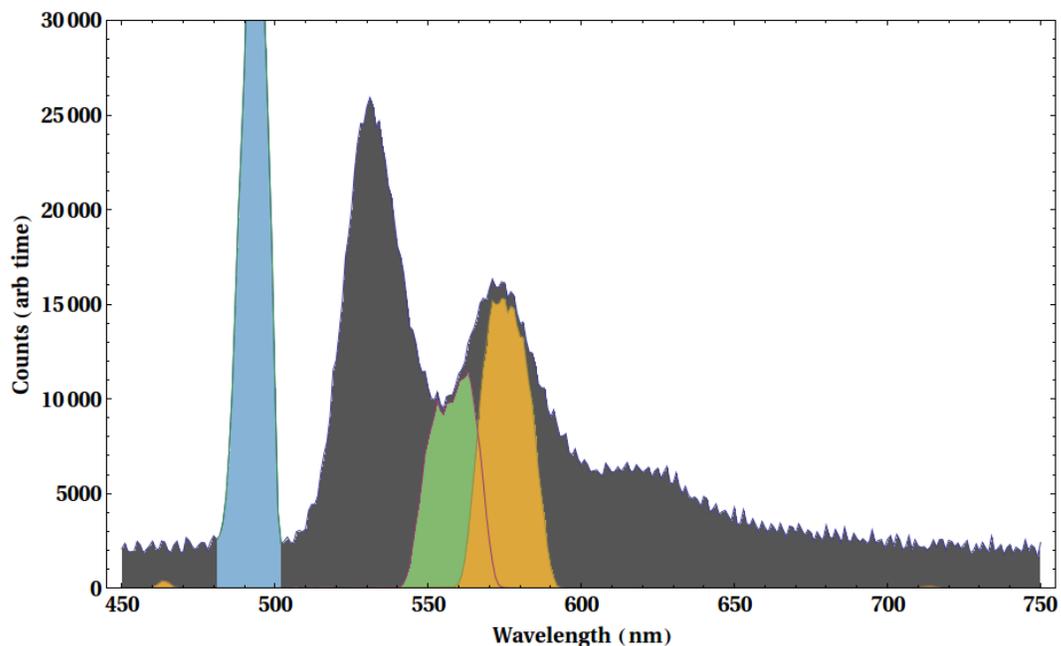


Figure S3. BTBP-doped PMMA film emission spectrum (gray), transmission spectra of each filter (green, orange) and offset peak of the excitation laser light (blue).

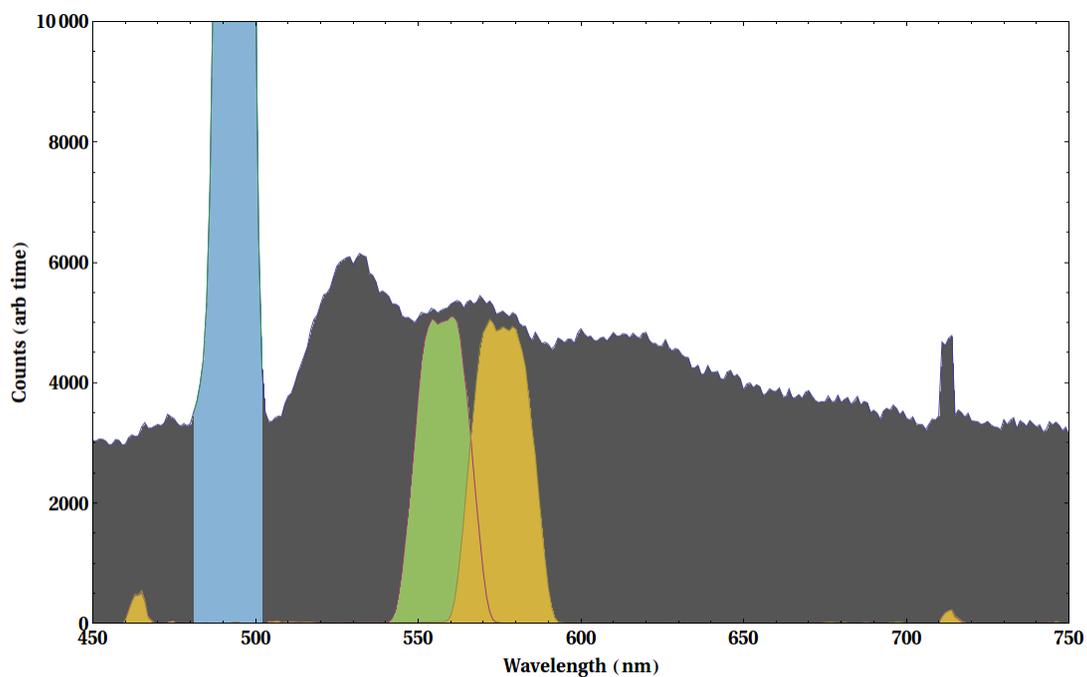


Figure S4. BTBP-doped PDMS film emission spectrum (gray), transmission spectra of each filter (green, orange) and offset peak of the excitation laser light (blue).

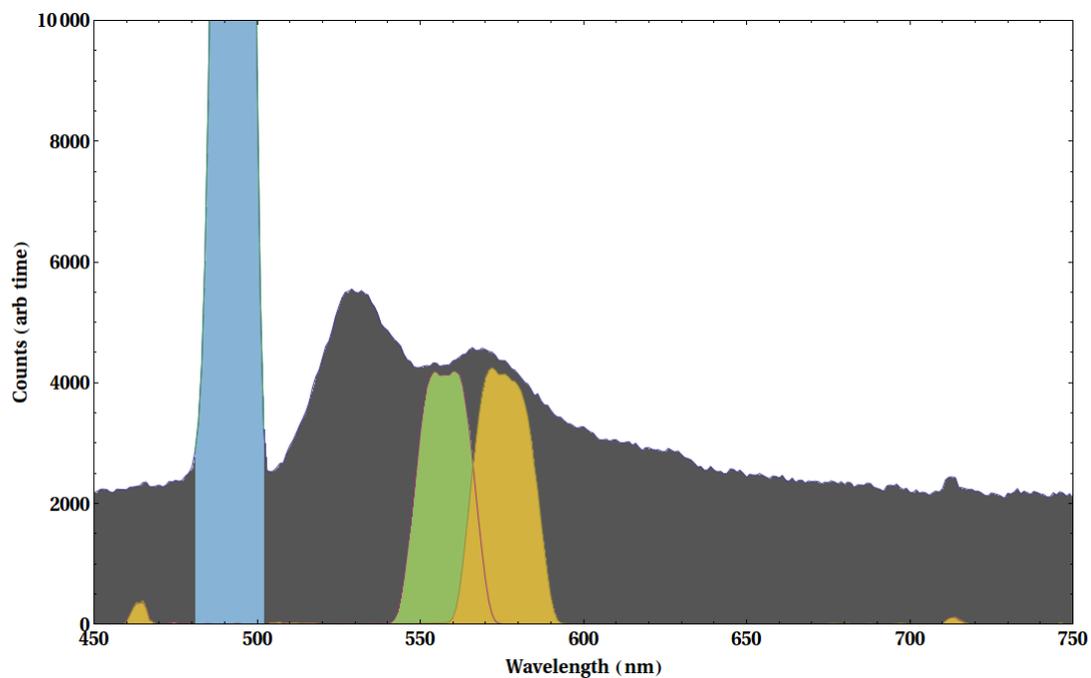


Figure S5. BTBP-doped HPDMS film emission spectrum (gray), transmission spectra of each filter (green, orange) and offset peak of the excitation laser light (blue).

S.3 PDMS Data

The spectrum data for the PDMS film was taken integrating over 2 seconds and averaged over 15 samples. The spectrum is similar to that of the PS film but with increased intensity in the red end of the spectrum. The PDMS film also exhibited a much larger blue-shift in the spectrum than the PS film, with little change in the overall intensity. This area also showed the most significant reduction in intensity due to photobleaching. The suitability analysis indicates multiple large regions of ratios with the maximum score. However the only area that has overlap with the other polymer films is around (555nm, 575nm), as used in the experiment.

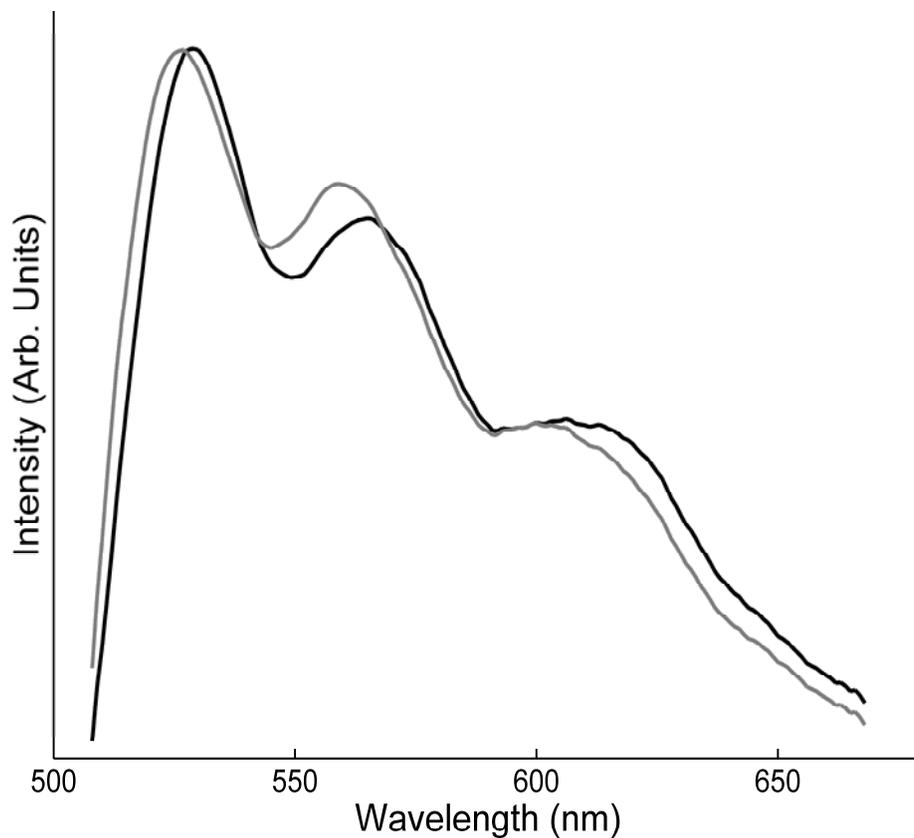


Figure S6. Emission spectra of the BTBP-doped PDMS thin film at 40°C (black line) and 80°C (gray line).

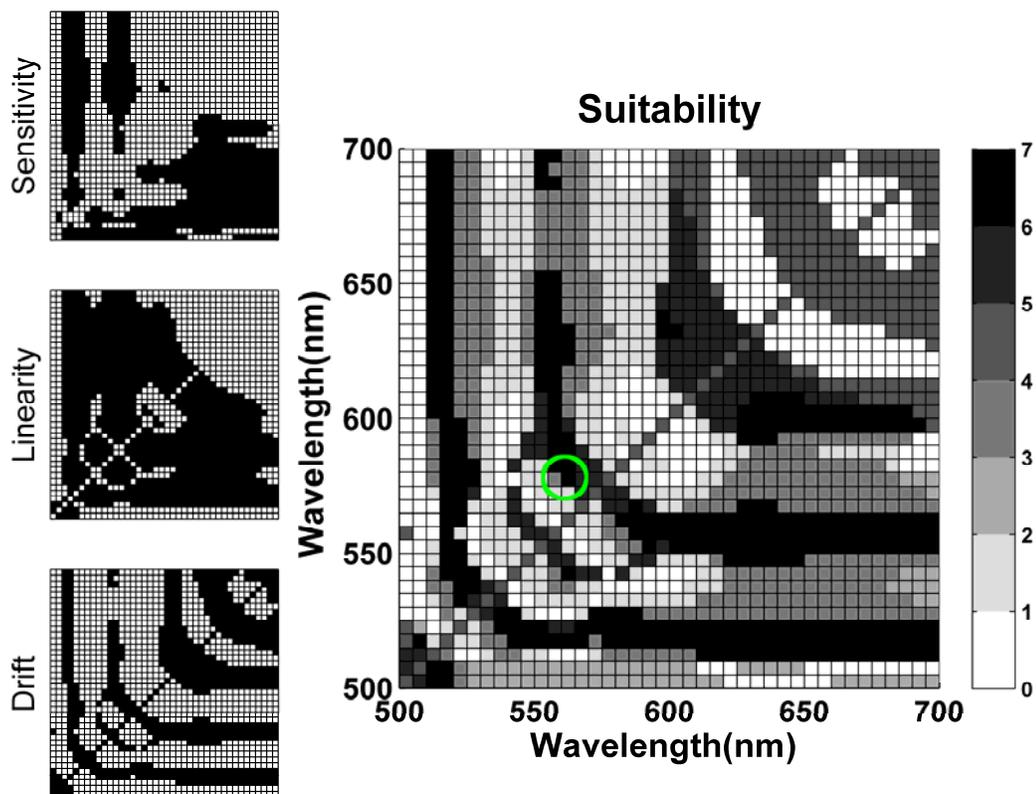


Figure S7. Suitability matrix for the BTBP-doped PDMS film (right) and component matrices (left). The region used in the experiment is highlight by the green circle.

For the BTBP-doped PDMS thin film, the binary progression criteria were as follows:

- Slope (%/C°) in the 40th percentile = 1
- Linear correlation coefficient greater than 0.97 = 2
- Drift parameter less than 10% = 4

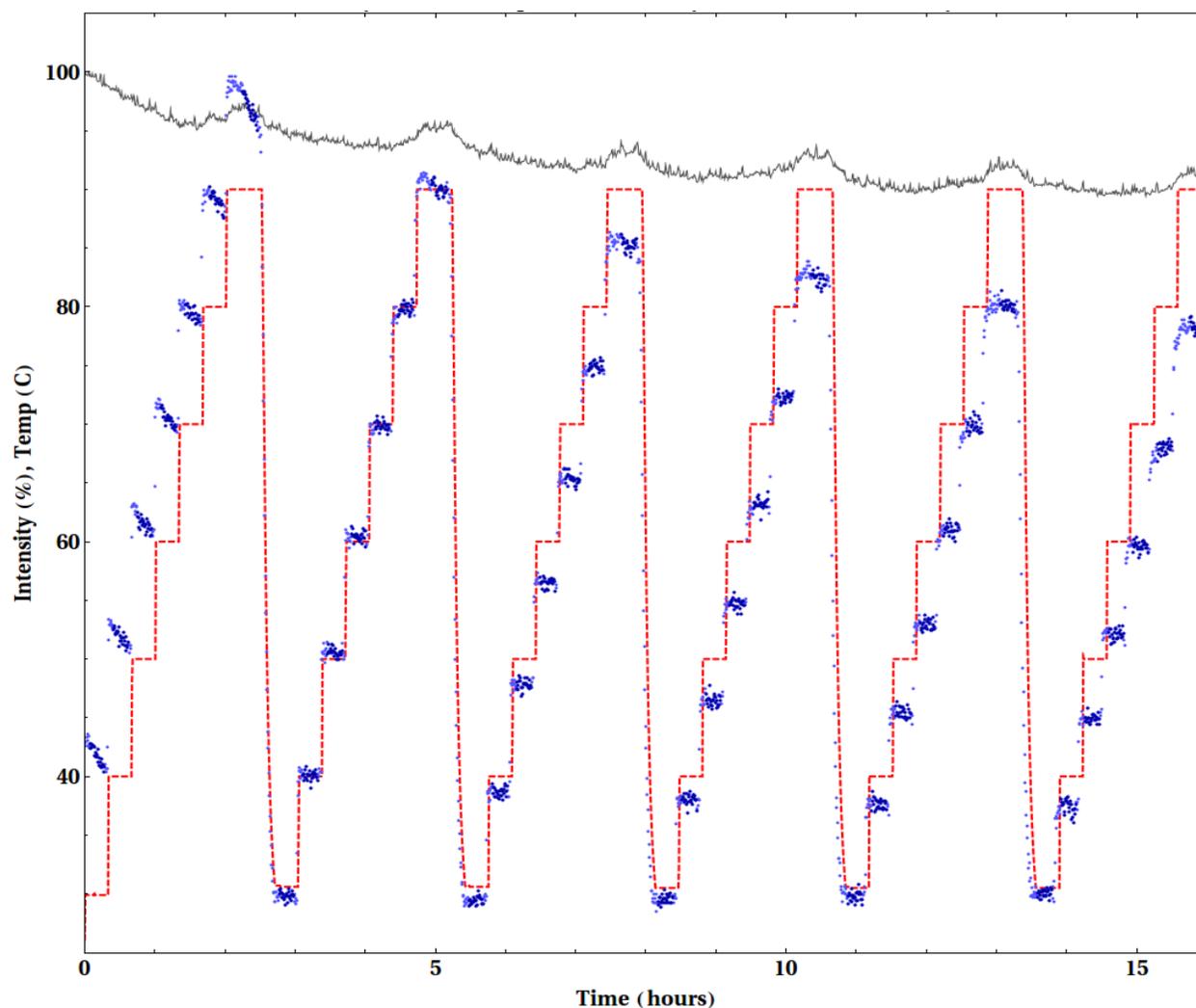


Figure S8. Real (red, dashed) and inferred (blue) temperature of the BTBP-doped PDMS film. The dark blue circles denote the data used for the fitting and suitability analysis. The gray line represents the total fluorescence intensity, scaled to 100%.

The slope and drift parameters for the BTBP-doped PDMS thin film were significantly reduced compared to the other materials due to the significantly increased bleaching. The total intensity of the PDMS film emission was reduced by ~7% over 15 hours of continuous exposure. The total emission also varied by almost 2.5% over the temperature range. For the BTBP-doped PDMS film, the Drift 1 was $\sim 1\text{C}^\circ$ while the Drift 2 was $\sim -10\text{C}^\circ$.

S.4 HPDMS

The spectrum data for the BTBP-doped HPDMS film was taken integrating over 0.6 seconds and averaged over 50 samples. The spectrum is similar to that of the PDMS film but with decreased intensity in the peak near 635nm. HPDMS film has less reduction in intensity due to photobleaching. The suitability matrix has only a small number of the maximum candidates. A local maximum in the area around (555nm, 575nm) was preferred because it has a higher overall intensity.

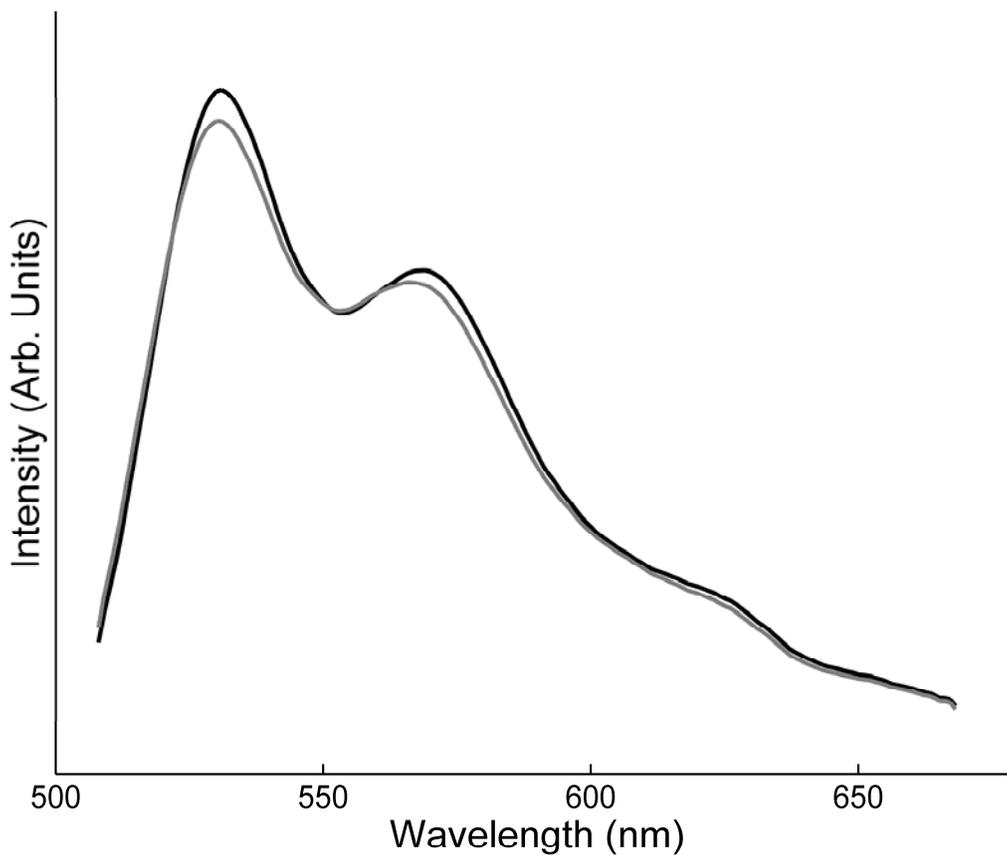


Figure S9. Emission spectra of the BTBP-doped HPDMS thin film at 40°C (black line) and 80°C (gray line).

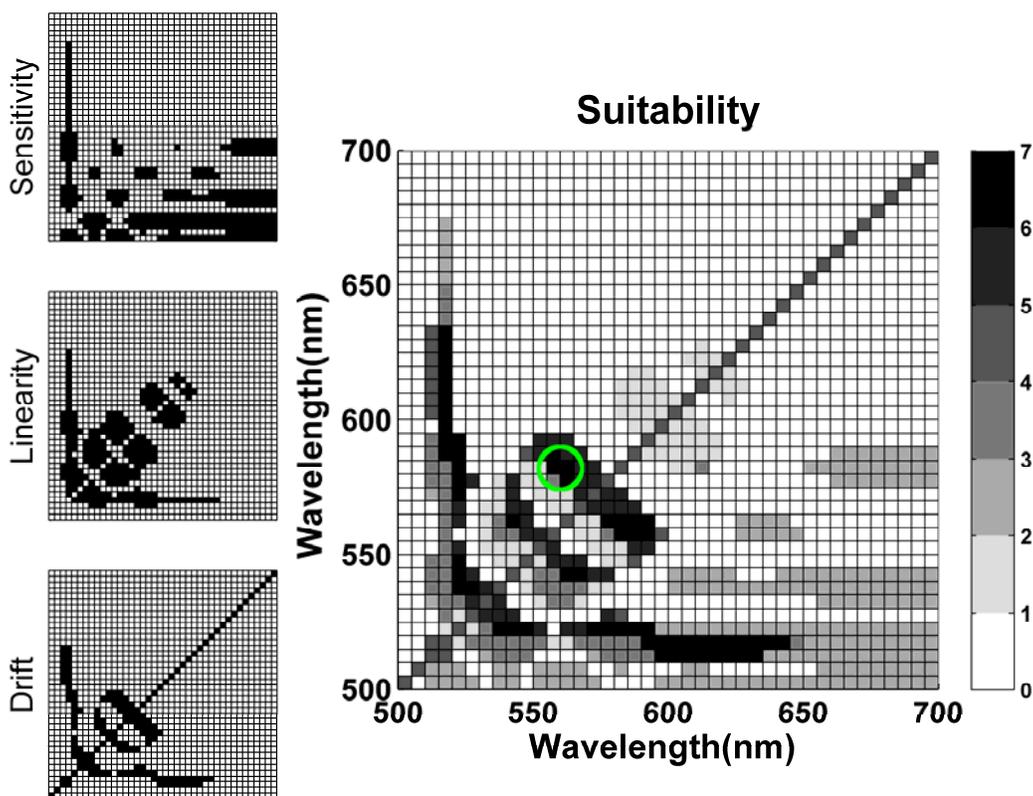


Figure S10. Suitability matrix for the BTBP-dope HPDMS film (right) and component matrices (left). The region used in the experiment is highlight by the green circle.

The binary progression criteria for the BTBP-doped HPDMS thin film were as follows:

- Slope (%/C°) in the 70th percentile =1
- Linear correlation coefficient greater than 0.97=2
- Drift parameter less than 5%=4

These values were identical to the PS film and PMMA film.

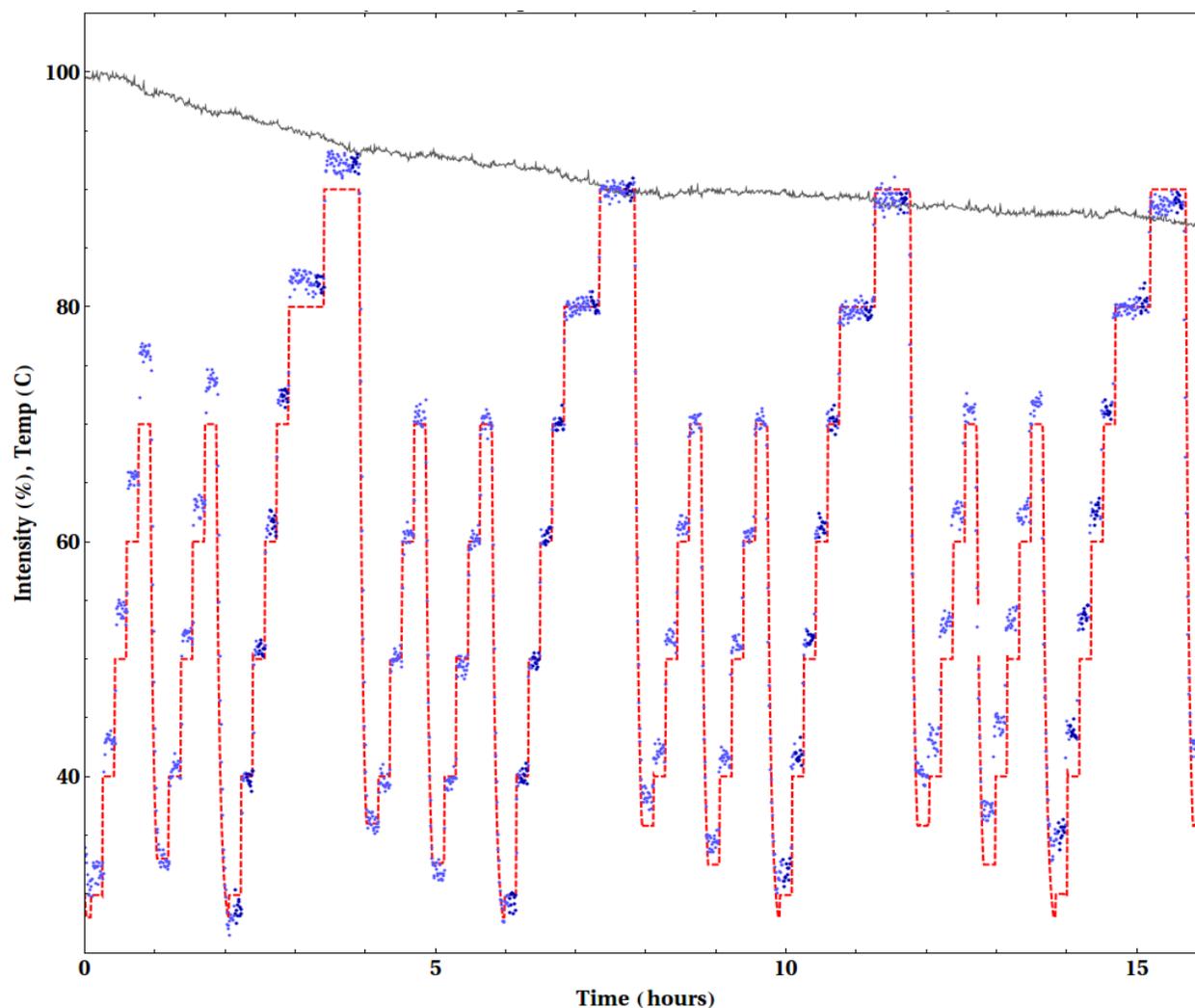


Figure S11. Real (red, dashed) and inferred (blue) temperature of the BTBP-doped HPDMS film. The dark blue circles denote the data used for the fitting and suitability analysis. The gray line represents the total fluorescence intensity, scaled to 100%.

The total intensity of the HPDMS film emission was reduced by ~10% over 15 hours of continuous exposure. The temperature dependence of the total emission was much weaker than that of the PDMS film. For the BTBP-doped HPDMS film, the Drift 1 was $\sim 4^{\circ}\text{C}$ over 8 hours, starting with the second cycle while the Drift 2 was $\sim 2^{\circ}\text{C}$ over the same period.

S.5 PMMA

The spectrum data for the BTBP-doped PMMA thin film was taken integrating over 0.5 seconds and averaged over 10 samples. The spectrum is similar to that of the BTBP-doped PDMS film but with increased intensity in the red end of the spectrum. This area also showed the most significant reduction in intensity due to photobleaching. The suitability matrix has a local maximum in the area around (555nm, 575nm), as used in the experiment.

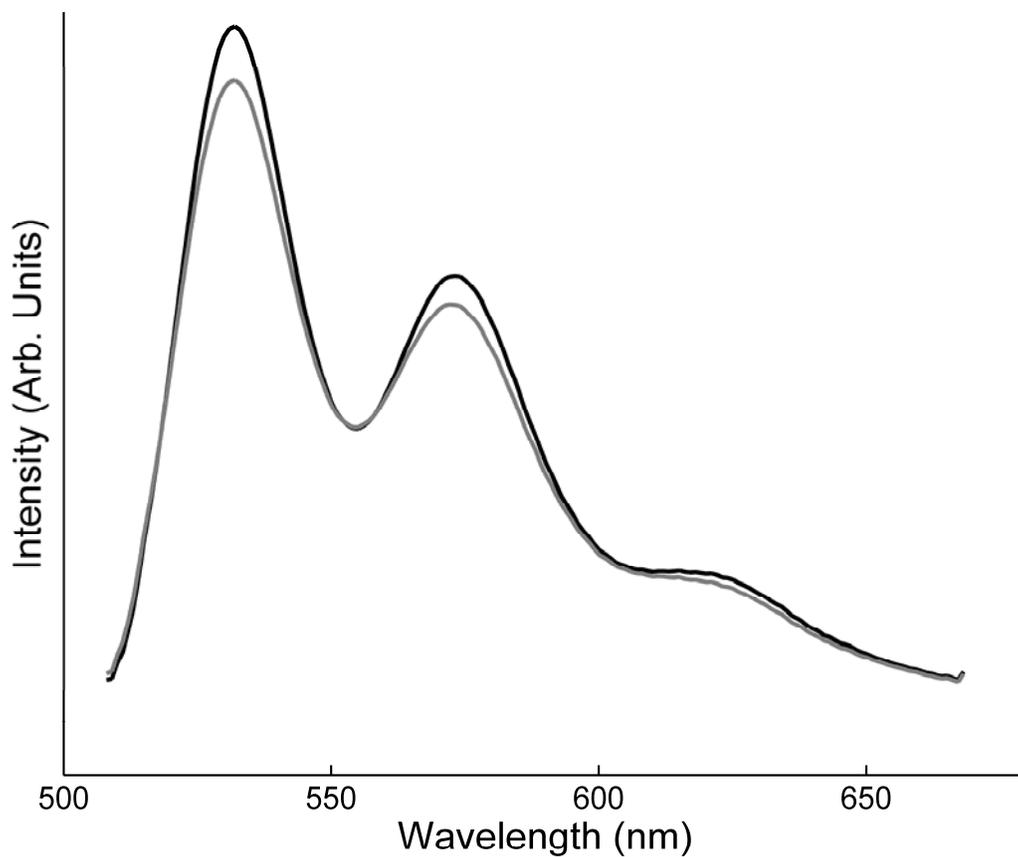


Figure S12. Emission spectra of the BTBP-doped PMMA thin film at 40°C (black line) and 80°C (gray line).

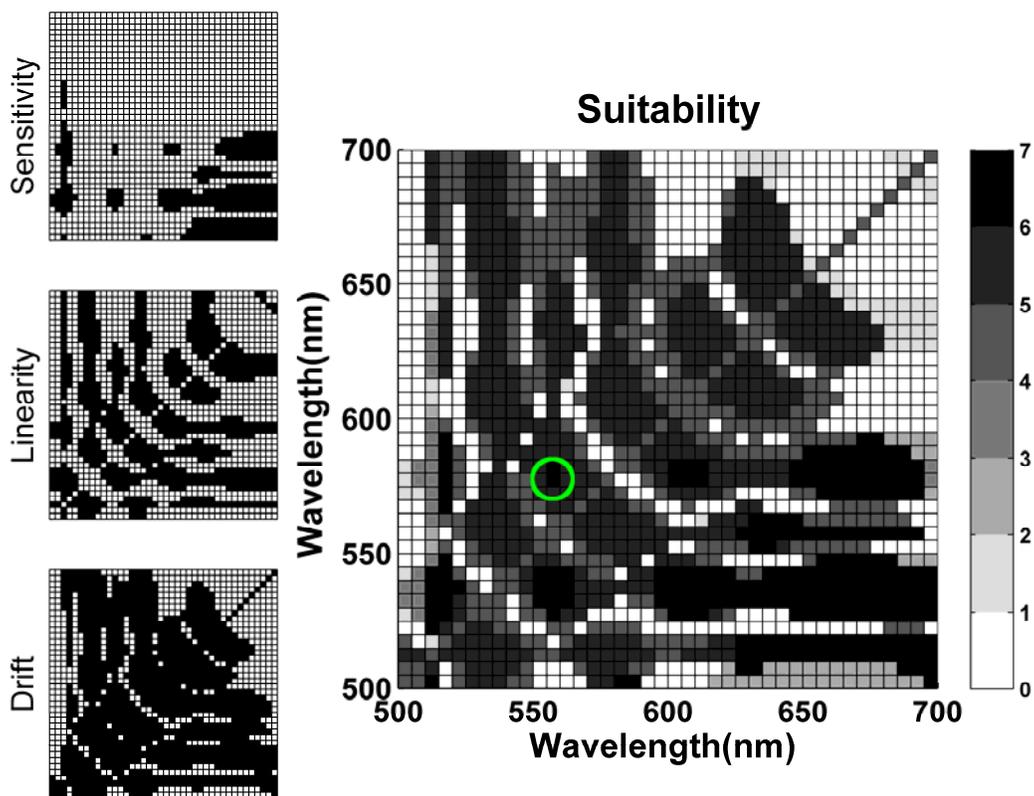


Figure S13. Suitability matrix for the BTBP-doped PMMA film (right) and component matrices (left). The region used in the experiment is highlight by the green circle.

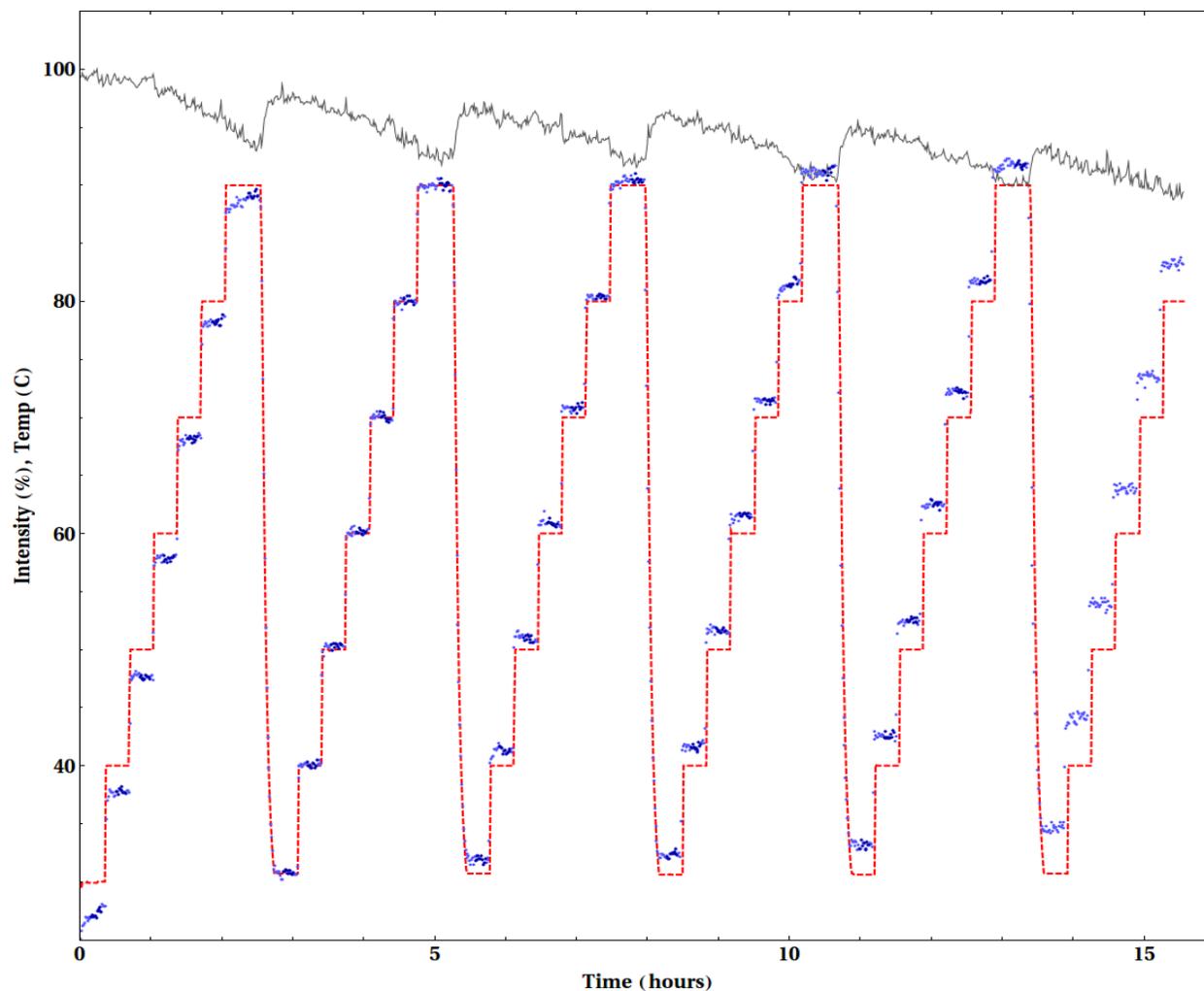


Figure S14. Real (red, dashed) and inferred (blue) temperature of the BTBP-doped PMMA film. The dark blue circles denote the data used for the fitting and suitability analysis. The gray line represents the total fluorescence intensity, scaled to 100%.

The total intensity of the BTBP-doped PMMA film emission was reduced by ~10% over 15 hours of continuous exposure. The temperature dependence of the total emission was ~ -8% and roughly linear. For the BTBP-doped HPDMS thin film, the Drift 1 was ~ 4C° over 8 hours, starting with the second cycle while the Drift 2 was ~ -2C° over the same period.