Supporting Information

Fluorescent-Based Thermal Sensing in Lipid Membranes

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This section contains supplemental information for the main manuscript, and it includes:

- **Characterization of the Prepared DPPC Liposomes**
- **Förster Resonance Energy Transfer Pair:**
- **Thermal Response of DPPC-DiD**
- **Thermal Response of DOPC-DiD/MPS-PPV**
- **pH stability of MPS-PPV**

I- Characterization of the Prepared DPPC Liposomes:

The average diameter of the prepared liposomes was estimated using dynamic light scattering. The hydrodynamic radius was measured for both DPPC and DPPC upon addition of MPS-PPV and DiD.

Table S1: The cumulant diameters and polydispersity indexes of DPPC and DPPC-DiD/MPS-PPV

Sample	Cumulant Diameter (nm)	Polydispersity index
DPPC	136.5	0.303
DID:DPPC/MPS-PPV	225	0.308

II- Förster Resonance Energy Transfer Pair:

DiD, a lyophilic fluorescent dye, was chosen as the acceptor given the overlap between its absorbance and the MPS-PPV emission (Figure S1).

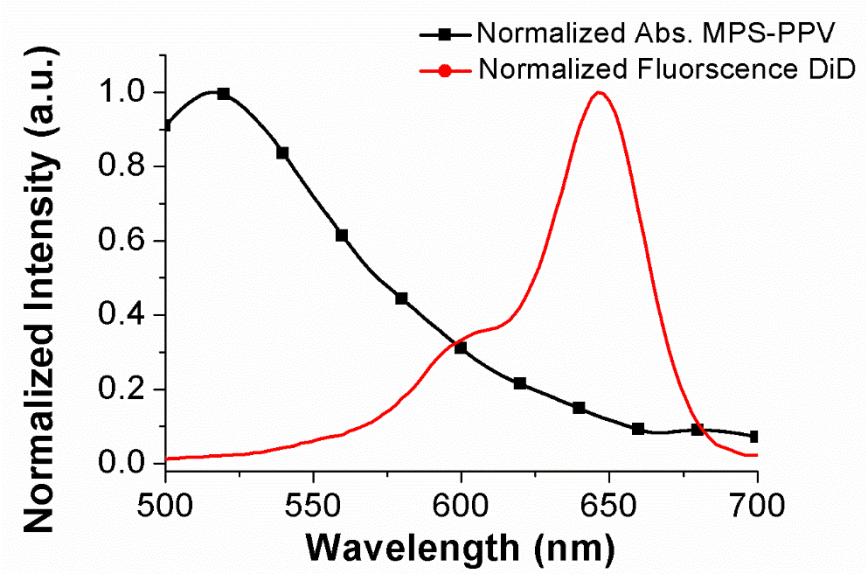

Figure S1: Normalized absorbance of MPS-PPV and normalized fluorescent emission of DiD when embedded in DPPC liposomes. Measurements were done in 10 mM HEPES buffer pH=7.3 and 150 mM NaCl and emission spectra were acquired upon excitation at 457 nm.

III- Thermal Response of DPPC-DiD:

In the presence of MPS-PPV, the fluorescent signal of DiD decreased with the increase of temperature but it was concurrent with the increase in MPS-PPV emission signal. To probe the effect of temperature on the DiD emission, a stained DPPC solution was subjected to increase in the solution temperature between 20 and 50 °C in a 5 °C increments (Figure S2). The

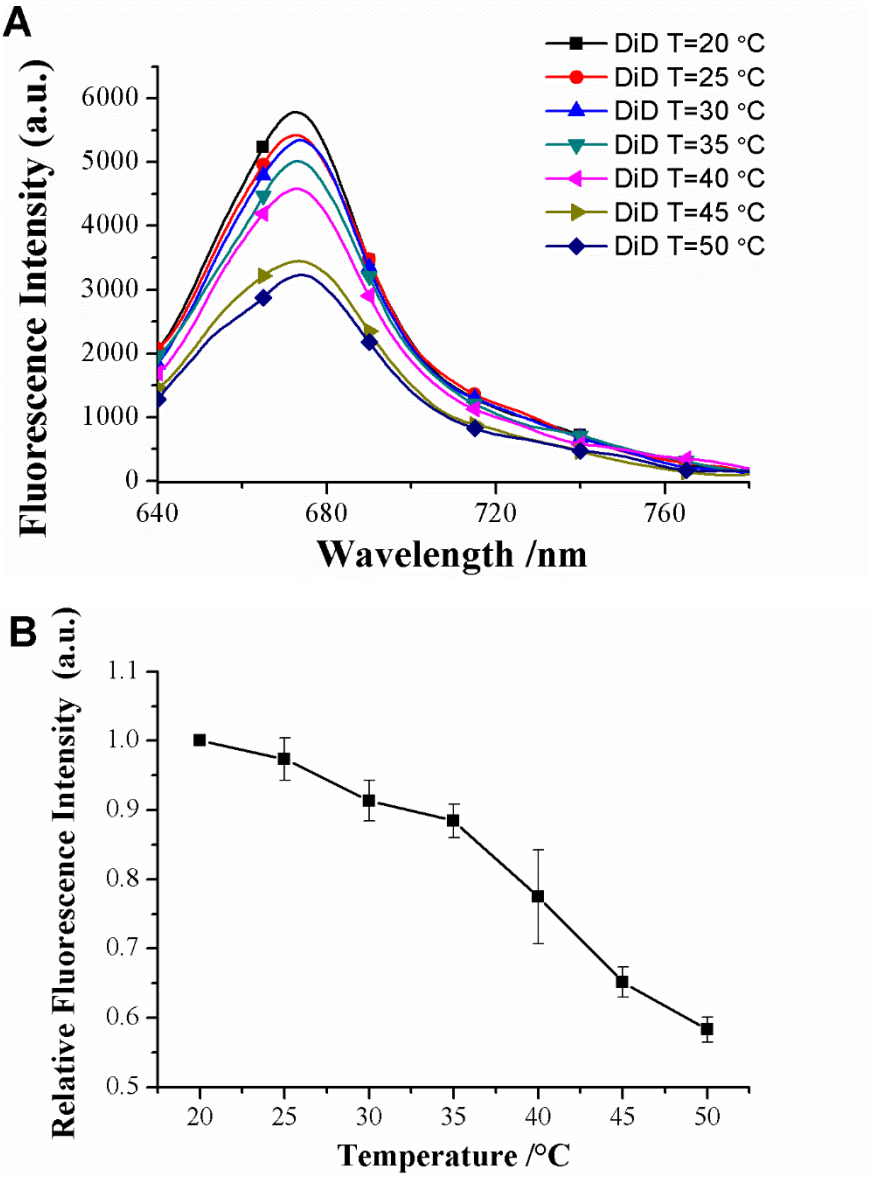

Figure S2: (**A**) Thermal response of DiD embedded in DPPC between 20 and 50 ºC. (**B**) Relative change in fluorescent emission for three trials. Error bars are calculated from the standard deviations of three independent measurements. Lines connecting the experimental points are for visual aid only. Experiment was performed in 10 mM HEPES with 150 mM NaCl (pH= 7.3) buffer solution.

IV- Thermal Response of DOPC-DiD/MPS-PPV, 10% Chol-DOPC-DiD/MPS-PPV, POPC-DiD/MPS-PPV,:

To understand the thermal response of MPS-PPV/DiD in a lipid membrane already above its transition temperature, DOPC liposomes (Tm = -17 °C) was prepared and stained with the FRET pair. The thermal response was tested between 20 and 50 °C and is summarized in **Figure S3**.

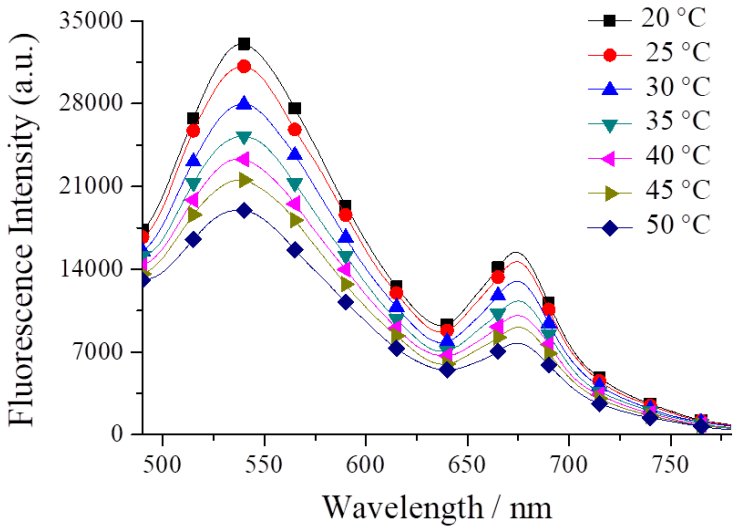

Figure S3: Fluorescence emissions of DOPC-DID/MPS-PPV between 20 and 50 °C in a 5 °C increments. Excitated at 457 nm and collected between 490 and 780 nm. Lines connecting the experimental points are for visual aid only. Experiment was performed in 10 mM HEPES with 150 mM NaCl (pH= 7.3) buffer solution.

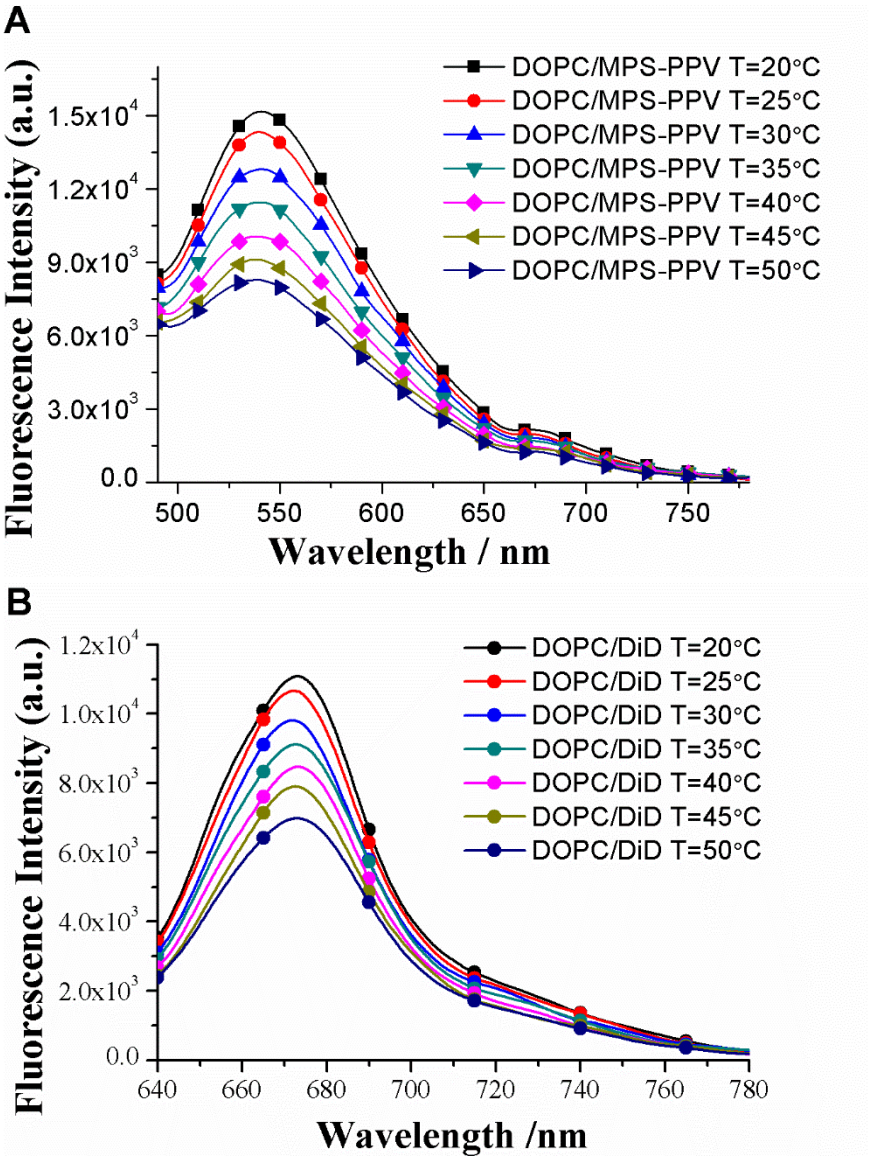

Figure S4: Fluorescence emission of (A) DOPC/MPS-PPV and (B) DOPC/DiD between 20 and 50 ºC. Measurements were done in 10 mM HEPES buffer pH= 7.3 and 150 mM NaCl.

POPC and DOPC with 10 % cholesterol are very close in molecular packing and organization. Comparing the FRET-pair thermal response in these two liposomes would allow us to better decouple the change in signal that results from thermal response versus structural.

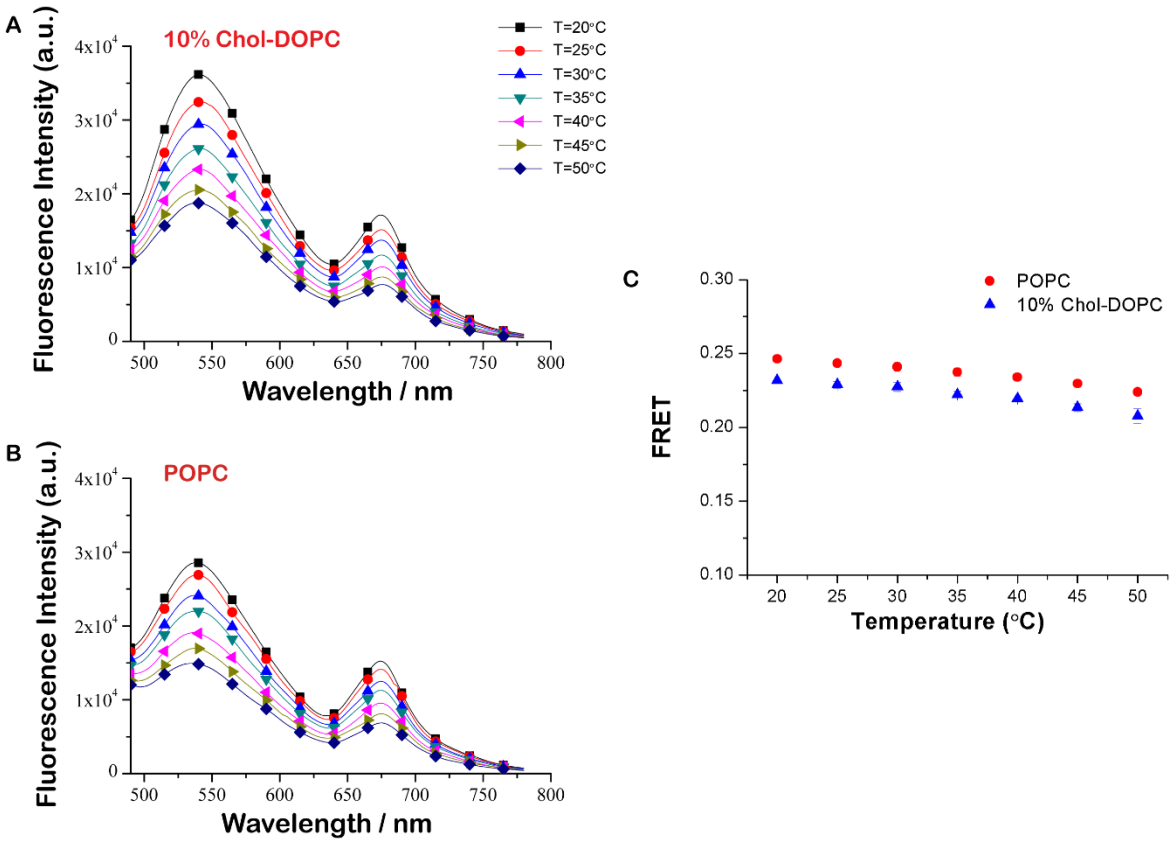

Figure S5: Fluorescence emission of (A) 10 % Chol-DOPC and (B) POPC between 20 and 50 ºC. (C) Calculated FRET at different temperatures. Error bars are calculated from the standard deviations of three independent measurements. Measurements were done in 10 mM HEPES buffer pH= 7.3 and 150 mM NaCl

V- pH stability of MPS-PPV

The stability of pristine MPS-PPV was tested under different pH conditions. The fluorescent signal was observed to drop (13 %) under acidic conditions which might be the result of the electrostatic interaction between the protonated HEPES and the sulfonate group of the side chain.

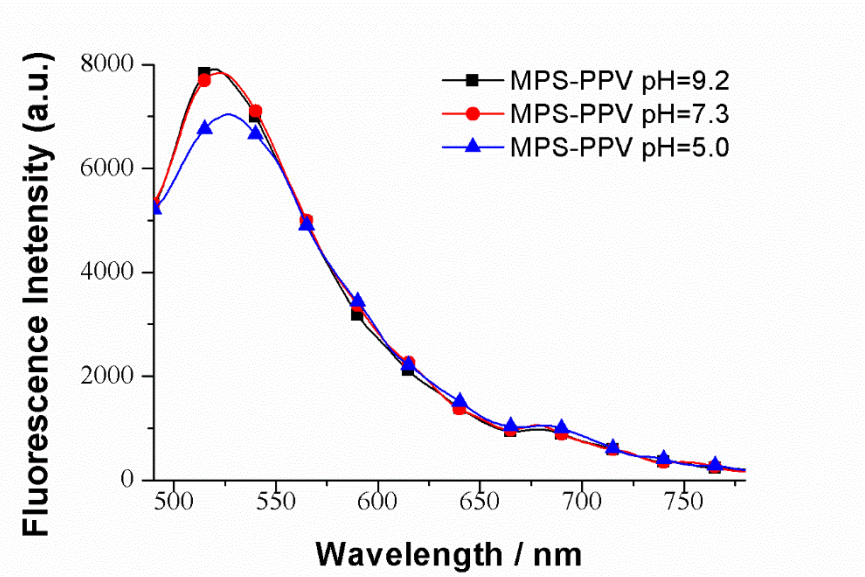

Figure S6: Emissions of MPS-PPV in basic (9.2), neutral (7.3) and acidic (5.0) mediums. Prepared in 10 mM HEPES with 150 mM NaCl buffer solution, upon exciting at 457 nm and acquired between 490 and 780 nm.

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