Electronic Supplementary Information

Temperature responsive phosphorescent small unilamellar vesicles

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General methods and materials

All reagent-grade chemicals were used without purification unless otherwise specified. Diethylenetriaminepentaacetic dianhydride (DTPAA), tryptamine, $Tb(CF_3SO_3)_3$ were obtained from Aldrich and used as received.

NMR Spectra

NMR spectra were measured with Bruker Avance 600 (1H: 600.1 MHz, 13C: 150.1 MHz, T = 300 K), Bruker Avance 400 (1H: 400.1 MHz, 13C: 100.6 MHz, T = 300 K), Bruker Avance 300 (1H: 300.1 MHz, 13C: 75.5 MHz, T = 300 K). The chemical shifts are reported in δ [ppm] relative to external standards (solvent residual peak). The spectra were analysed by first order, the coupling **S-1** constants are given in Hertz [Hz]. Characterisation of the signals: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, br = broad, dd = double doublet. Integration is determined as the relative number of atoms. The solvent used is reported for each spectrum.

Mass Spectra

Mass spectra were obtained with Varian CH-5 (EI), Finnigan MAT 95 (CI; FAB and FD), Finnigan MAT TSQ 7000 (ESI). Xenon serves as the ionisation gas for FAB.

IR Spectra

IR spectra were recorded with a Bio-Rad FTS 2000 MX FT-IR and Bio-Rad FT-IR FTS 155.

Absorption Spectroscopy

Absorption were recorded on a Varian Cary BIO 50 UV/VIS/NIR Spectrometer with temperature control by use of a 1 cm quartz cuvettes (Hellma) and aqueous buffered solution (HEPES 25 mmol, pH = 7.4).

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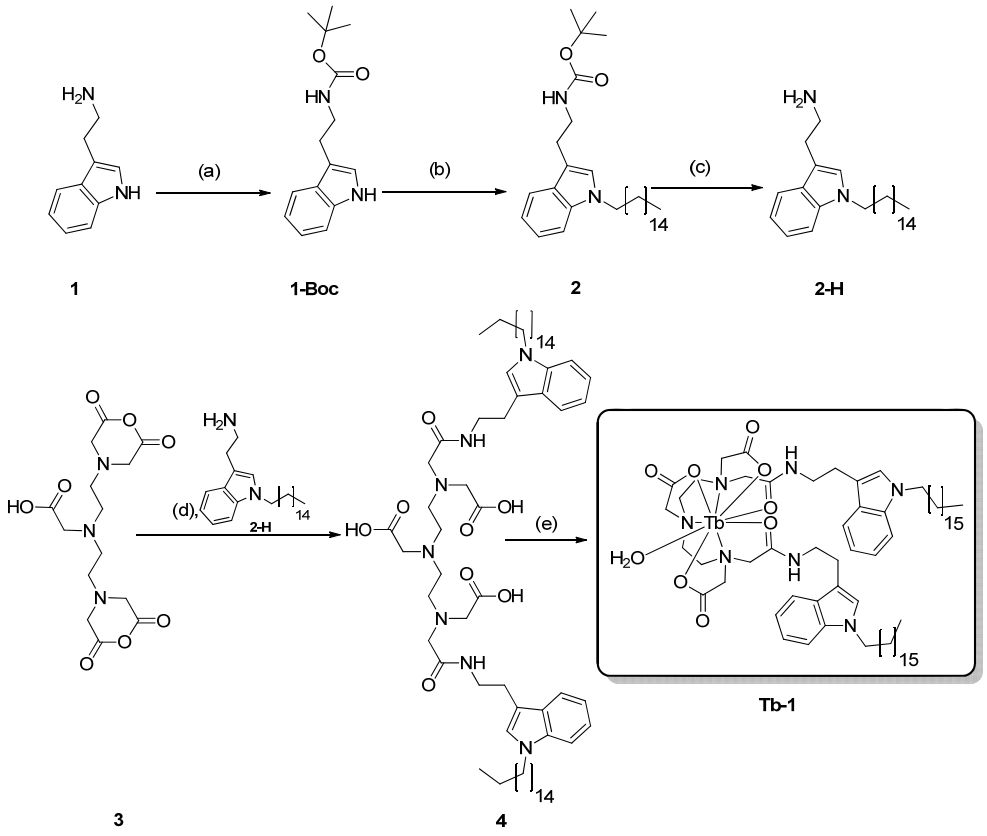
Emission Spectroscopy

Luminescence intensity and lifetime measurements were performed with aqueous buffered solution (HEPES 25 mmol, pH = 7.4) in 1 cm quartz cuvettes (Hellma) and recorded on a Varian 'Cary Eclipse' fluorescence spectrophotometer with temperature control.

Dynamic light scattering

Photon correlation spectroscopy (PCS) measurements were performed on a Malvern Zetasizer 3000 at 25°C using 1cm disposable polystyrene fluorescence cuvettes (VWR). Three subsequent measurements of 60 s each were performed for each sample. Data analysis was performed using the Malvern PCS software.

Synthetic Scheme for amphiphilic Tb(III) complex



Scheme S1: Synthesis of **Tb-1**. (a) DCM, $(Boc)_2O$, NEt_3 [quantitative yield]; (b) DMF, 1-bromooctadecane, NaH [Yield: 93%]; (c) DCM, trifluoroacetic acid [Yield: 95%]; (d) DMF, $CHCl_3$, 40°C, 24h [Yield: 55%]; (e) CH_3OH, $Tb(CF_3SO_3)_3$, 24h [Yield: 98%]

Experimental Section

Synthesis of compound 1-Boc: To a yellow suspension of tryptamine (1.00 g, 6.24 mmol) in 1, 4-dioxane (5 mL) was added Et_3N (1.80 mL, 12.9 mmol). A solution of $(Boc)_2O$ (1.50 g, 6.87 mmol) in 1,4-dioxane (5 mL) then was added to the reaction mixture. This mixture was stirred for 1 h and the resulting yellow solution was concentrated to dryness under reduced pressure. The crude residue was purified by coloumn chromatography on silica gel (30% ethylaceate in petrolether) to give the desired 1-Boc amine as an amorphous white solid (yield: quantitative).

^{1}H NMR (300 MHz, $CDCl_3$) = 1.54 (s, 9H), 3.00 (t, 2H), 3.53 (d, 2H), 4.85 (br, s, 1H), 6.97 (s, 1H), 7.15-7.28 (m, 1H), 7.37 (d, 1H), 7.65 (d, 1H), 8.68 (br, 1H) ppm **^{13}C NMR** (75 MHz, CDCl3) =14.29, 21.16, 25.86, 28.57, 41.10, 60.60, 67.12, 111.45, 112.73, 118.80, 119.28, 121.99, 122.37, 127.44, 136.56, 156.33, 171.49 ppm **MS (EI MS)** m/z = 260.31 [M^+] (Calc. =260.33)

Synthesis of compound 2: To a stirred suspension of NaH (60% dispersion in mineral oil, 0.05g, 2.37mmol) in THF (5mL), was added a solution of **1-Boc** (0.50 g, 1.97mmol) in THF (5mL). After stirring at room temperature for 10min, 1-bromooctadecane (0.76g, 2.37mmol) was added, and stirring was continued for 3h. Water was then added, and the mixture was extracted with ethylacetate. The organic layer was washed with brine, dried over $MgSO_4$, and concentrated under vacuum. The residue was chromatographed on silica gel, eluting with 10% ethylacetate in petrolether to give **2-Boc** as an amorphous solid (yield: 93%).

^{1}H NMR (300 MHz, $CDCl_3$) = 0.93 (t, 3H), 1.30 (s, 28H), 1.48 (s, 9H), 1.86 (t, 2H), 2.98 (t, 2H), 3.50 (t, 2H), 4.08 (2H), 6.96 (s, 1H), 7.10-7.16(m, 1H), 7.21-7.27 (m, 1H), 7.33 (d, 1H), 7.62(d, 1H) ppm. **^{13}C NMR** (75 MHz, CDCl3) =14.20, 22.77, 25.81, 27.12, 28.25, 28.85, 29.53, 29.57, 32.00, 44.96, 65.84, 109.34, 111.50, 118.73, 121.47, 125.69, 127.93, 136.42, 155.93, 161.00, 177.52 ppm. **MS (EI MS)** m/z = 512.7 [M^+] (Calc. =512.81)

Synthesis of compound 2-H: Compound **2** (0.54g, 1.78mmol) was dissolved in dichloromethane (5ml). Trifluoroacetic acid (2ml 24.9mmol) was added and the solution was stirred for 6h. The solvent was then evaporated of under vacuum pressure. The colourless oil obtained, which is a trifluoroacetate salt was then dissolved in methanol: water mixture (1:1) and passed through basic ion exchange resin. The solvent was removed and the compound **2** was recovered as an amorphous white solid (yield: 95%).

^{1}H NMR (300 MHz, $CDCl_3$) = 0.82 (t, 3H), 1.20 (s, 28H), 1.68 (t, 2H), 2.86-2.96 (m, 4H), 3.96(t, 2H, 6.95-7.05 (m, 3H), 7.20 (d, 1H), 7.48(d, 1H) ppm. **^{13}C NMR** (75 MHz, CDCl3) =14.59, 23.81, 27.59,

28.01, 30.66, 31.34, 33.23, 42.16, 47.19, 110.61, 111.34, 119.87, 122.77, 127.34, 129.21, 138.20 ppm. **MS (EI MS)** m/z = 412.7 [M^+] (Calc. =412.6)

Synthesis of compound 4: Diethylenetriaminepentaacetic dianhydride (**1**) (0.12g, 0.35 mmol) was dissolved in 5 mL of dry DMF. **Compound 2** (0.29 g, 0.69 mmol) was dissolved in 5 mL of dry chloroform; added drop wise to the solution and the mixture was left to stir at 40 °C for 24 h. On cooling to room temperature, a precipitate was formed, which was collected. The white solid was stirred in water at ~ 80 °C for one hour, isolated, stirred in diethyl ether for one hour and isolated. The crude product was recrystallised from 50:50 chloroform:methanol. On cooling a white solid was formed, which was isolated by filtration, washed with diethyl ether and dried under vacuum (yield: 55%).

^{1}H NMR (400 MHz, CD_3OD:$CDCl_3$= 1:1) = 0.84 (t, 6H), 1.22 (s, 60H), 1.69 (t, 4H), 2.89-2.99 (m, 12H), 3.33-3.47 (m, 18H), 3.95 (t, 4H), 6.88(s, 2H), 6.99(m, 2H), 7.09(t, 2H), 7.22(d, 2H), 7.52(d, 2H) ppm **^{13}C NMR** (100 MHz, CD_3OD:$CDCl_3$= 1:1) =14.49, 23.21, 27.29, 28.01, 30.34, 31.12, 32.98, 42.02, 47.04, 50.10, 53.33, 55.08, 56.72, 58.59, 110.59, 111.34, 119.87, 122.35, 127.04, 129.01, 138.24 ppm **MS (EI MS)** m/z = 1183.1 [M^+] (Calc. = 1182.7) ν_{max} (KBr disc): 3303*b* (OH), 2919m, 2848*m* (CH); 1625*w* (C=O); 1468*w*, 1373*m*, 1236*m*, 1098*w*

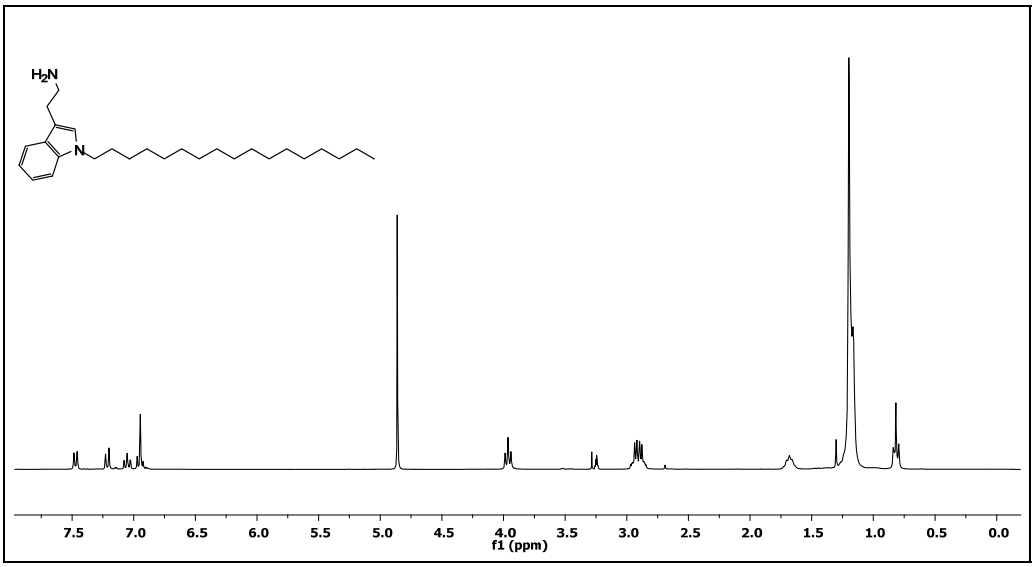
Synthesis of compound Tb-1: Compound **4** (0.1 g, 0.08 mmol) was dissolved in approximately 10 mL of a hot 1:1 $CHCl_3$/CH_3OH mixture. $Tb(CF_3SO_3)_3$ (0.05 g, 0.09 mmol,), dissolved in 2 mL CH_3OH, was added and the mixture was stirred at 50 °C for 8 hours. The solvent was removed under vacuum and the white solid obtained was washed with water and dried (yield: 98%). **MS (EI MS)** m/z = 1339.1 [M^+] (Calc. = 1340.8)

Preparation of LNTs

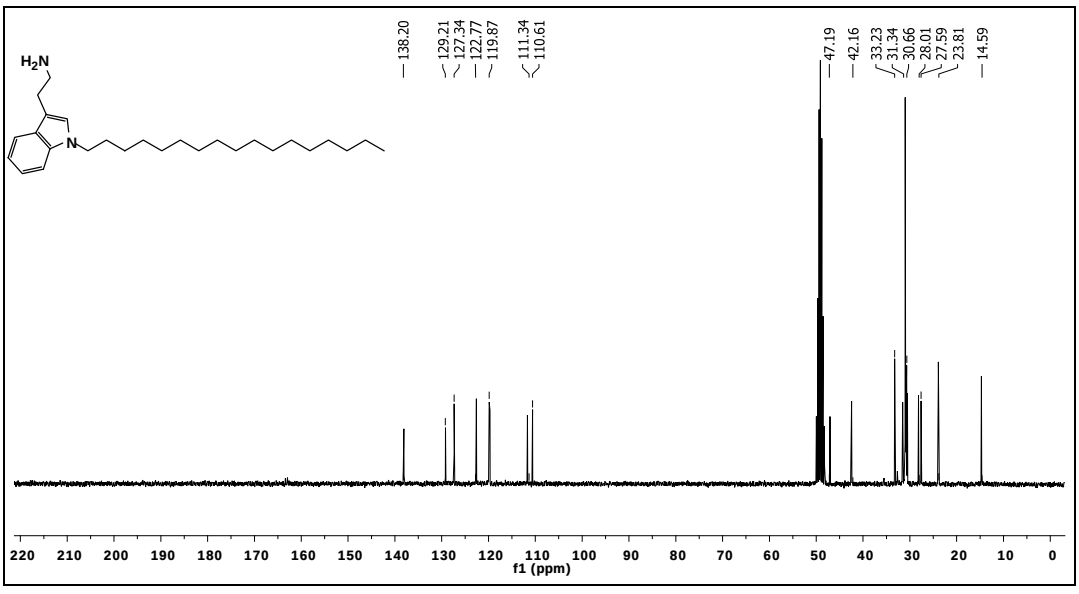
LNT, vesicular solutions were prepared by using the previously reported film hydration method. According to that a 2 mM solution of phospholipid with 10% Tb complex in dichloromethane was evaporated by a stream of air. An appropriate amount of buffered aqueous solution was added to get a 2mM solution. This solution was then heated around the phase transition temperature of the constituent phospholipid to yield a cloudy self-assembled vesicular solution. This was then extruded through definite filters (100nm) while hot to yield a unilamellar vesicular solution. The average sizes of the vesicular solutions were measured by dynamic light scattering (DLS). All the intensity and lifetime based measurements were carried out by using a diluted vesicular solution with 5 μM conc. of Tb complex.

Supporting Data

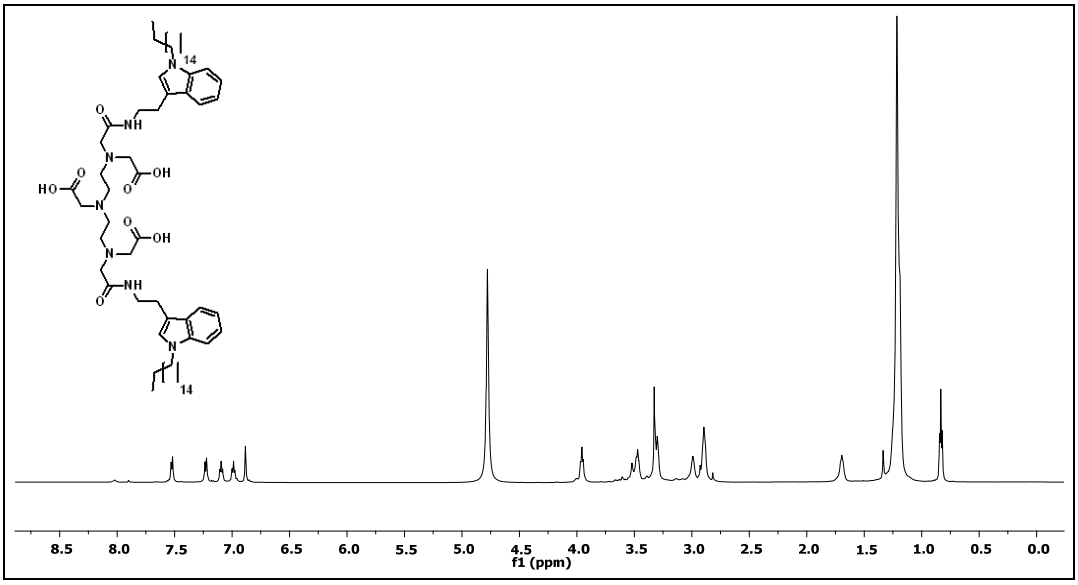
^{1}H and ^{13}C NMR of the synthesised compounds:



^{1}H NMR of compound **2** (300MHz, $CDCl_3$)



^{13}C NMR of compound **2** (75MHz, $CDCl_3$)



^{1}H NMR of compound **4** (400MHz, CD_3OD:$CDCl_3$= 1:1)

Excitation and Emission Spectra:

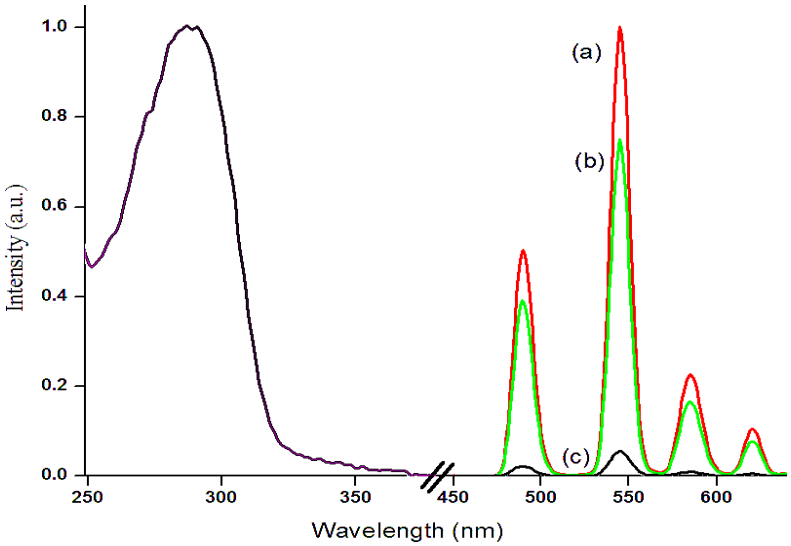


Fig. S-1 Excitation and emission spectra of **LNT** [excitation spectra: Tb(III) complex (5%)+DSPC) vesicle at 25°C, conc. of Tb(III) = 5 x 10^{-6} M; emission spectra: (a) [Tb(III) complex (5%)+DSPC] vesicle at 25°C , conc. of Tb(III) = 5 x 10^{-6} M, (b) [Tb(III) complex (5%)+DOPC] vesicle at 25°C, conc. of Tb(III) = 5 x 10^{-6} M, (c) Tb(III) complex at 25°C, conc. of Tb(III) = 1 x 10^{-4} M]

Temperature dependent emission intensity and lifetime measurements:

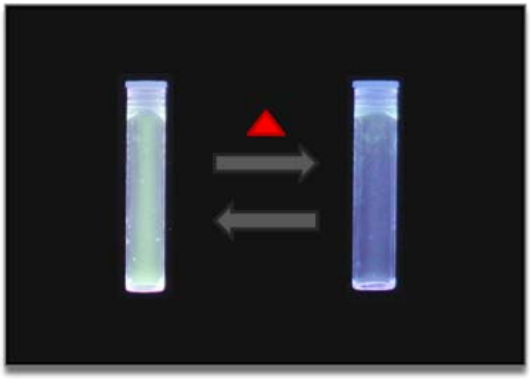


Fig. S-2 Photograph of change in emission intensity of **LNT 2** (conc. of vesicular solution = 2mM, conc. of Tb(III) = 1x 10^{-4} M) with temperature. Phosphorescence was recorded at different temperature by using a UV table (λ_{ex} = 315 nm)

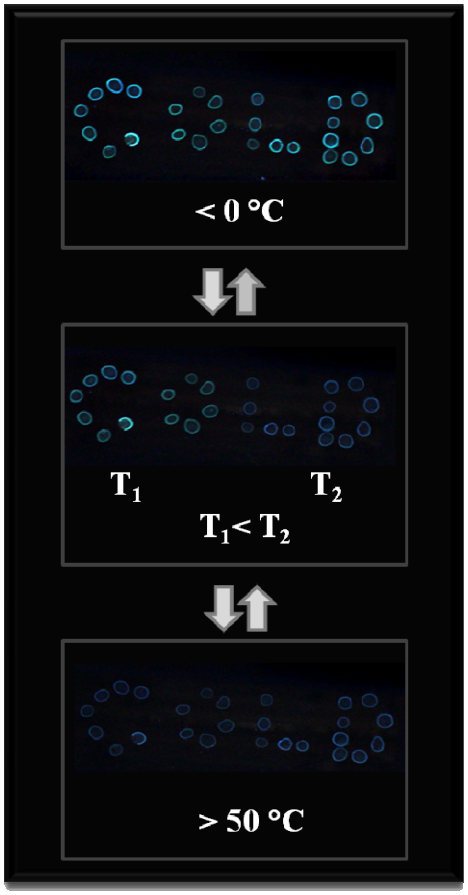


Fig. S-3. Photographs of luminescent membranes at different temperature spread on a glass surface. The letters were written on a glass surface by using a solution of **LNT 1** (2mM). After drying with blowing air the phosphorescence of the surface was recorded at different temperatures by using a UV table (λ_{ex} = 315 nm)

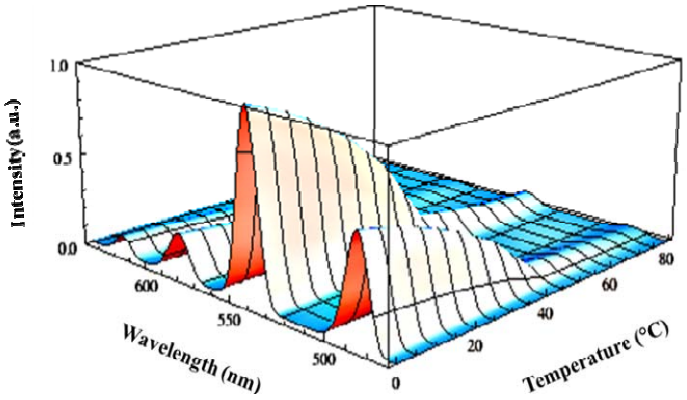


Fig. S-4: [Left] Temperature dependence of emission intensity measurements for **LNT2** (conc. of Tb(III) = 5 x 10^{-6} M)

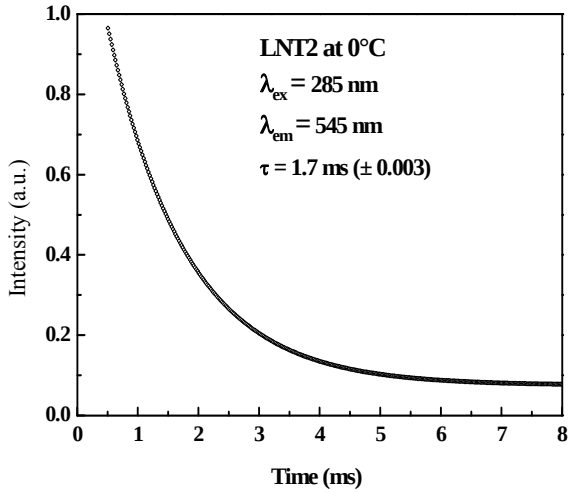


Fig. S-5: Luminescence decay vs. Time plot for **LNT2** at 0°C

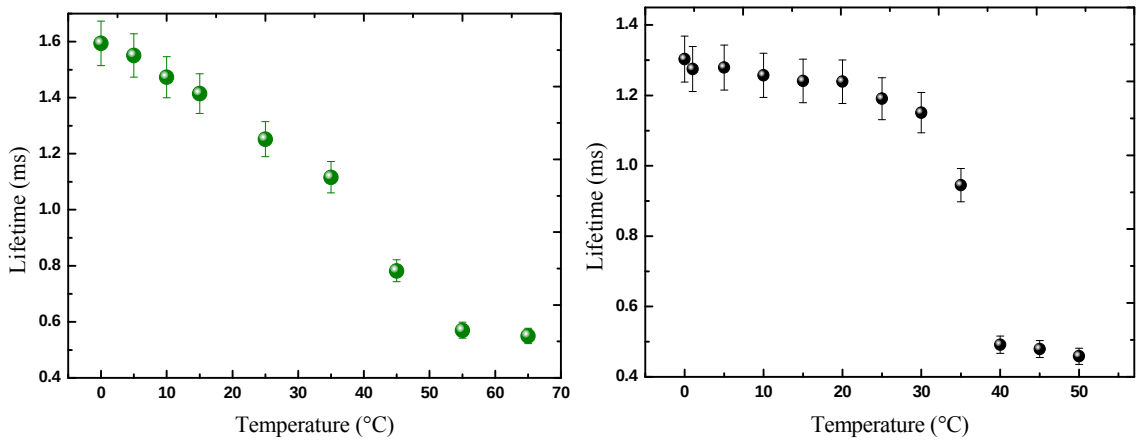


Fig. S-6: Change in lifetime (± s.d.) (ms) with change in temperature [left: **LNT4**, right: **LNT2**]

Types of buffer, pH and salt concentration dependent studies:

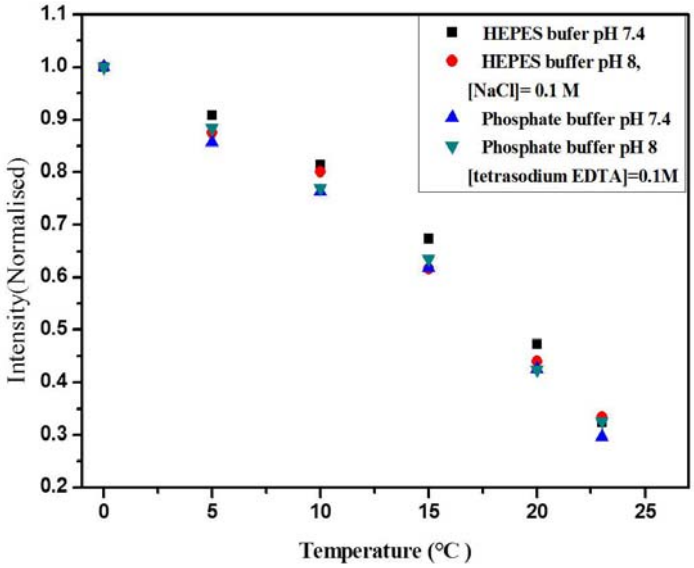


Fig. S-7: Change in intensity of LNT3 with change in temperature in different buffer, at different pH and salt concentrations (changes at 545 nm wavelength are shown)

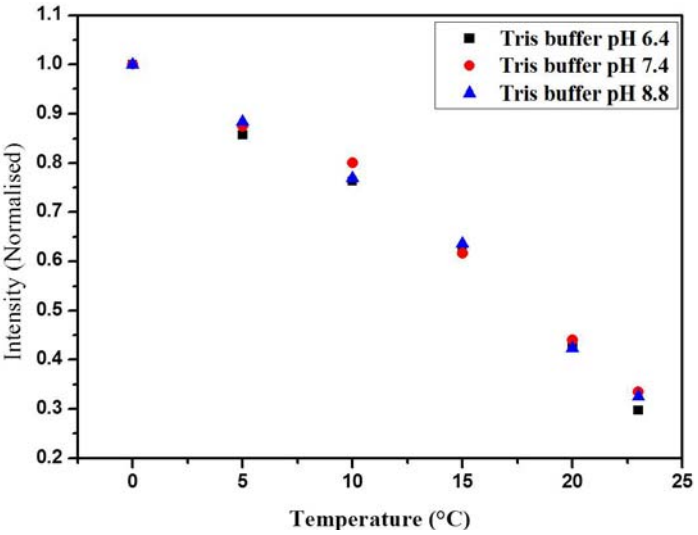


Fig S-8: Effect of pH on change in intensity of LNT3 with change in temperature (changes at 545 nm wavelength are shown)

Emission Intensity based measurements in cell culture medium

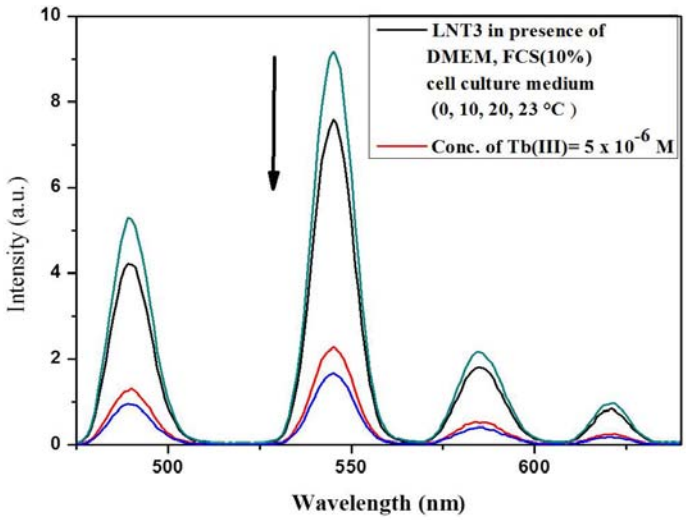


Fig S-9: Change in luminescence with change in temperature for LNT3 in cell culture medium (DMEM, 10% FCS)

Change in intensity in temperature range 34-43°C

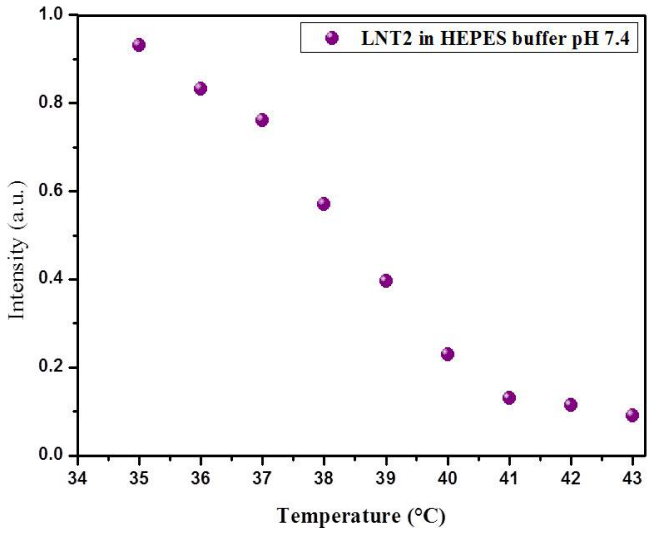


Fig S-10: Applicability of LNT2 in 37-40°C, the most important range for human body (**changes at 545 nm emission wavelength are shown)**

Dynamic light scattering measurements:

			Diam. (nm)	% Intensity	Width (nm)
Z-Average (d.nm):	95,42	**Peak 1:**	99,04	98,9	32,94
PdI:	0,212	**Peak 2:**	5384	1,1	320,2
Intercept:	0,919	**Peak 3:**	0,000	0,0	0,000
Result quality	**Good**				

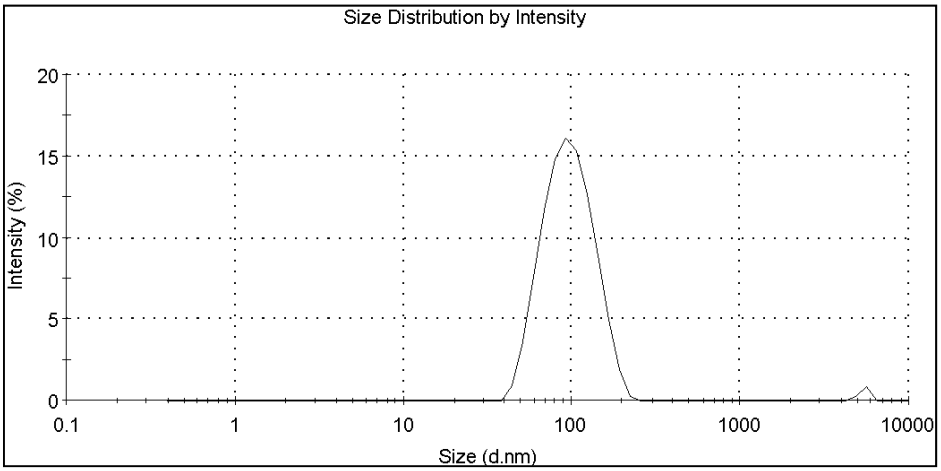


Fig. S-11: Dynamic light scattering measurements for **LNT3**

			Diam. (nm)	% Intensity	Width (nm)
Z-Average (d.nm):	89,86	**Peak 1:**	100,6	100,0	35,07
PdI:	0,096	**Peak 2:**	0,000	0,0	0,000
Intercept:	0,912	**Peak 3:**	0,000	0,0	0,000
Result quality	**Good**				

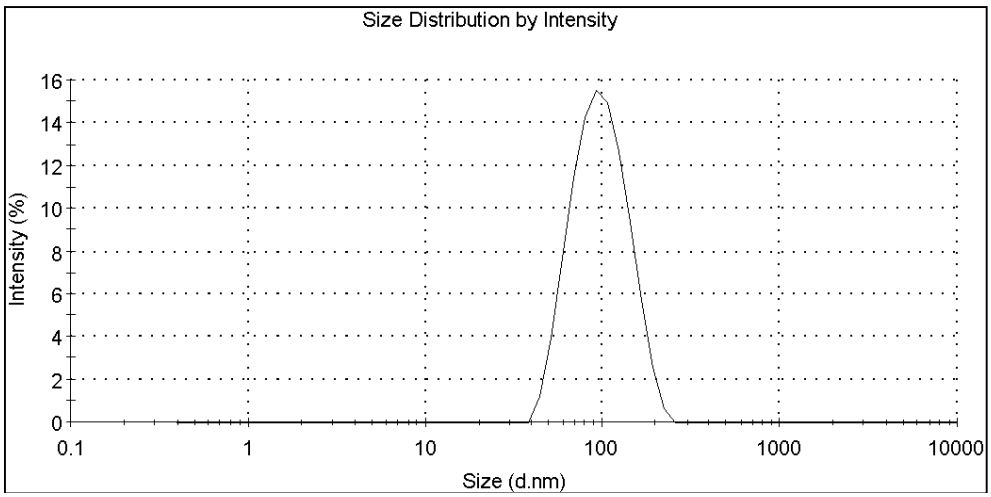


Fig. S-12: Dynamic light scattering measurements for **LNT4**