Electronic Supplementary Material (ESI) for New Journal of Chemistry.

Electronic Supplimentary Information

Rhodamine based Turn-On Chemosensor for Fe^{3+} in Aqueous Medium and Interactions of Its Fe^{3+} Complex with DNA.

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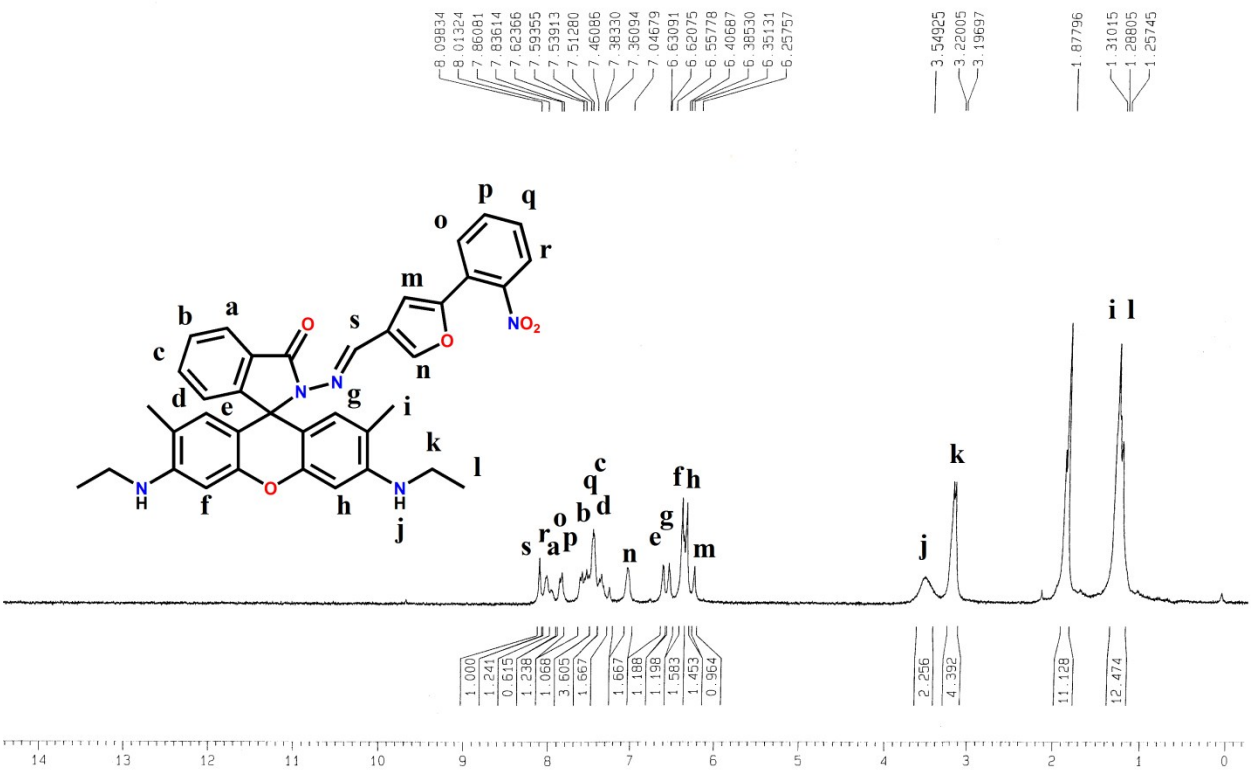
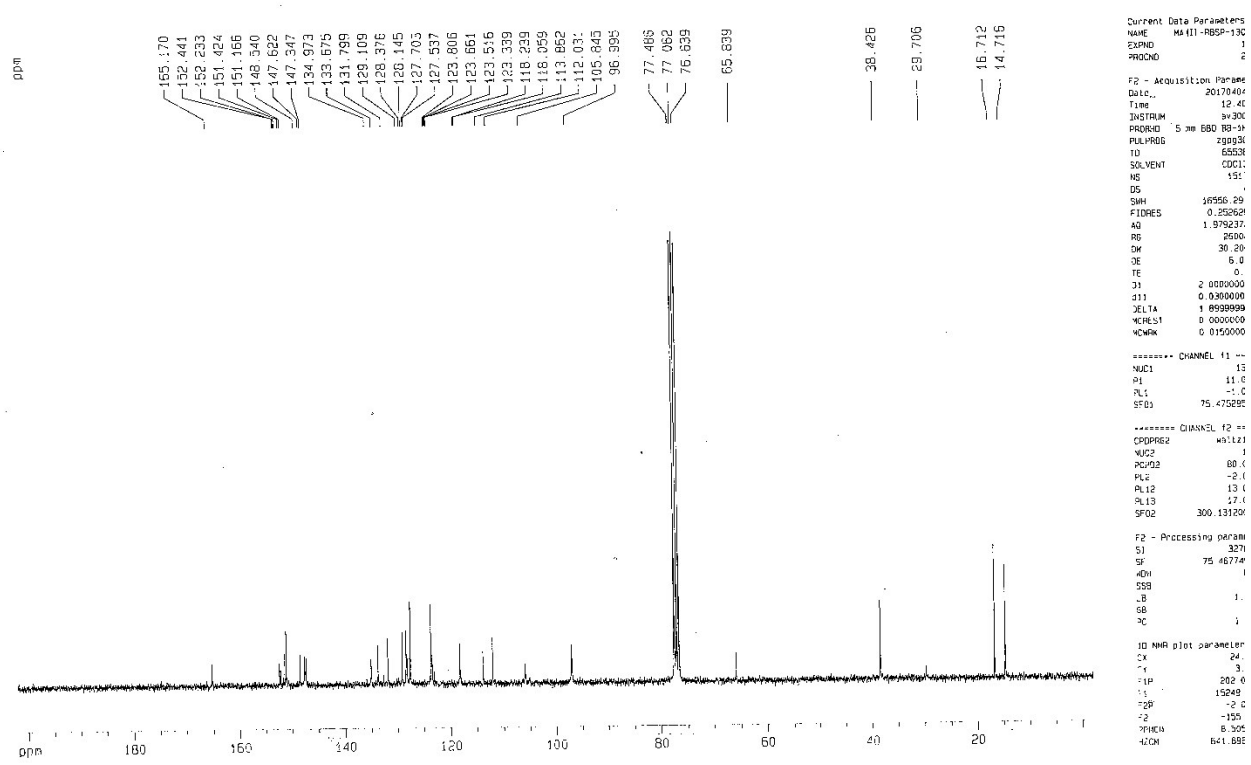


FigureS1. ^1H NMR of L in $CDCl_3$.



FigureS2. ^{13}C NMR of L in $CDCl_3$.

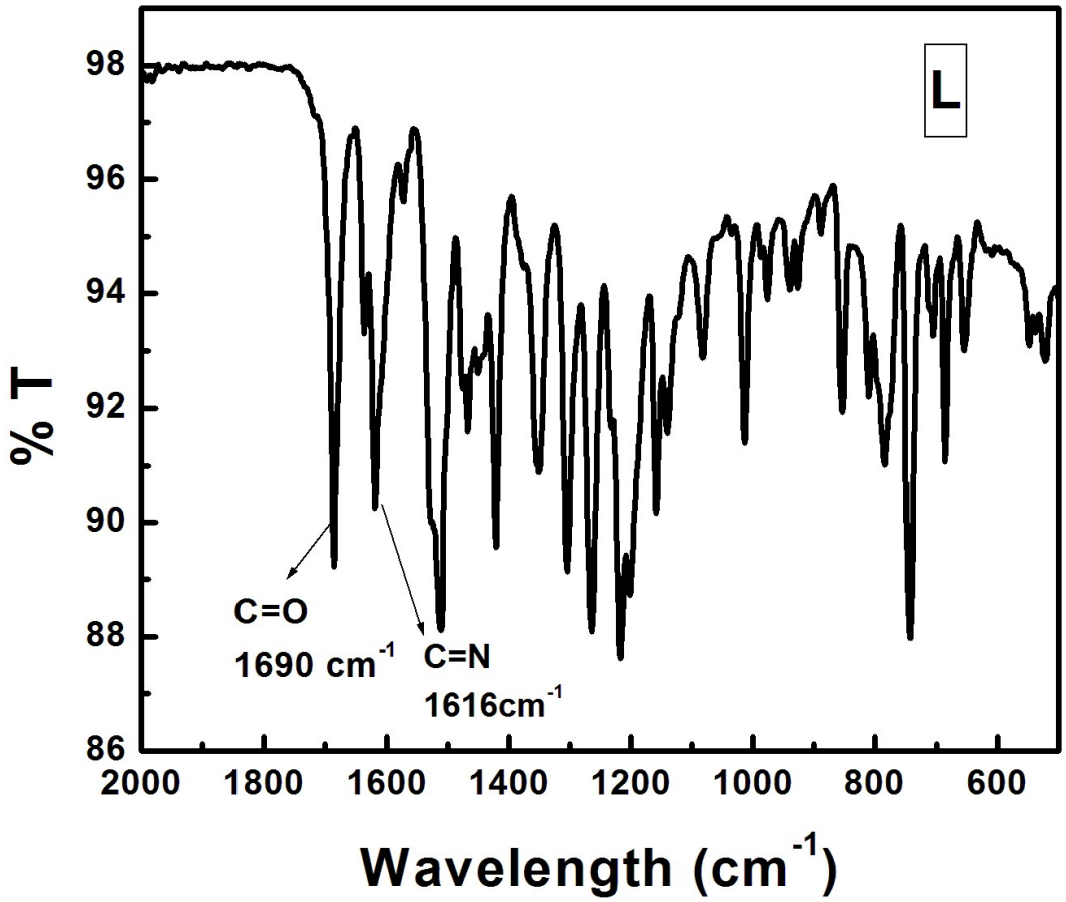


Figure S3. IR spectra of **L.**

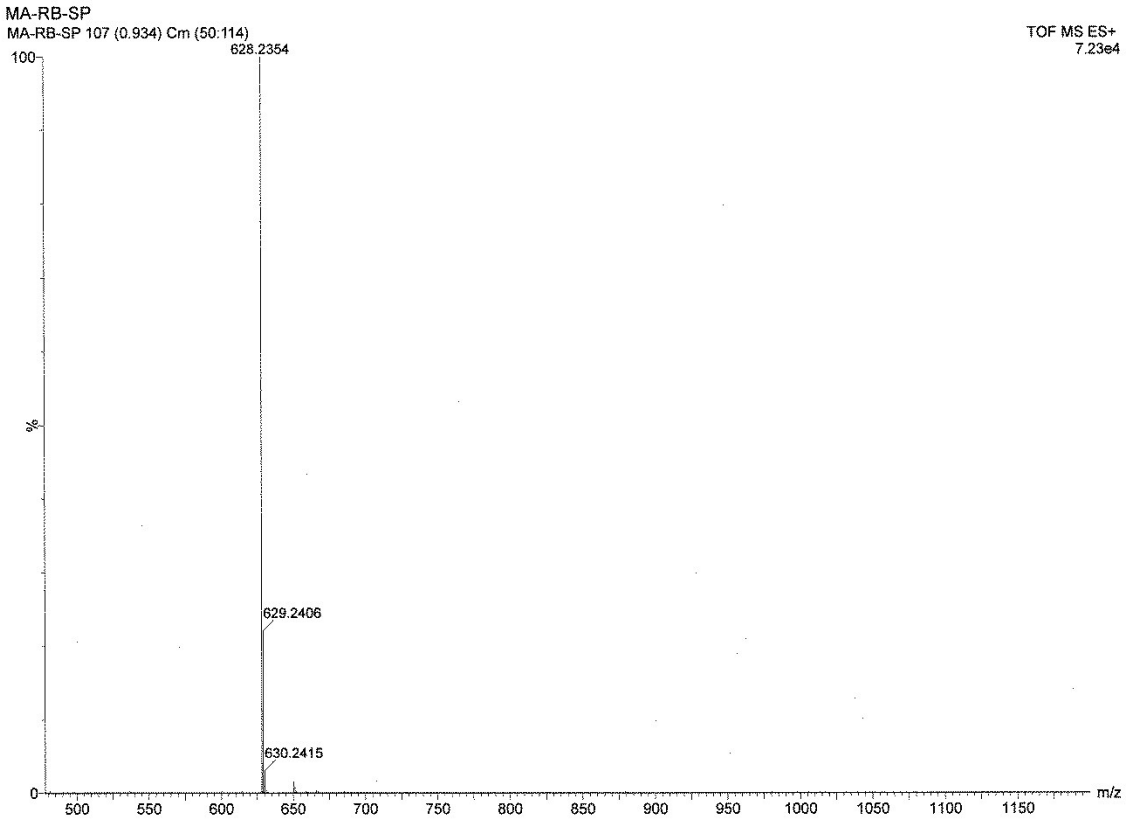


Figure S4. Mass Spectrum of L in MeCN.

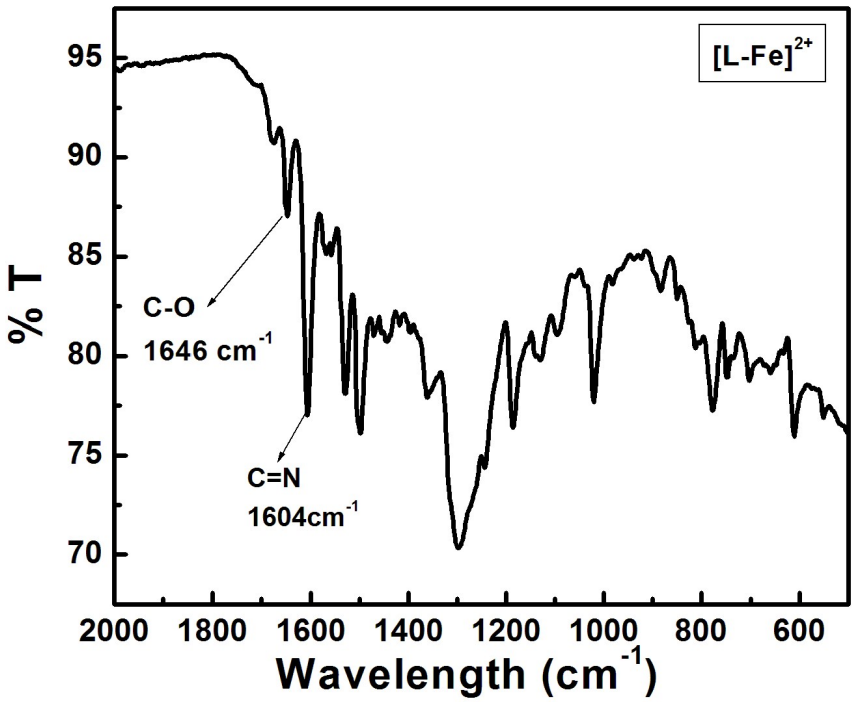


Figure S5. IR Spectrum of $[\mathbf{LFe(NO_3)_2}]^+$ complex.

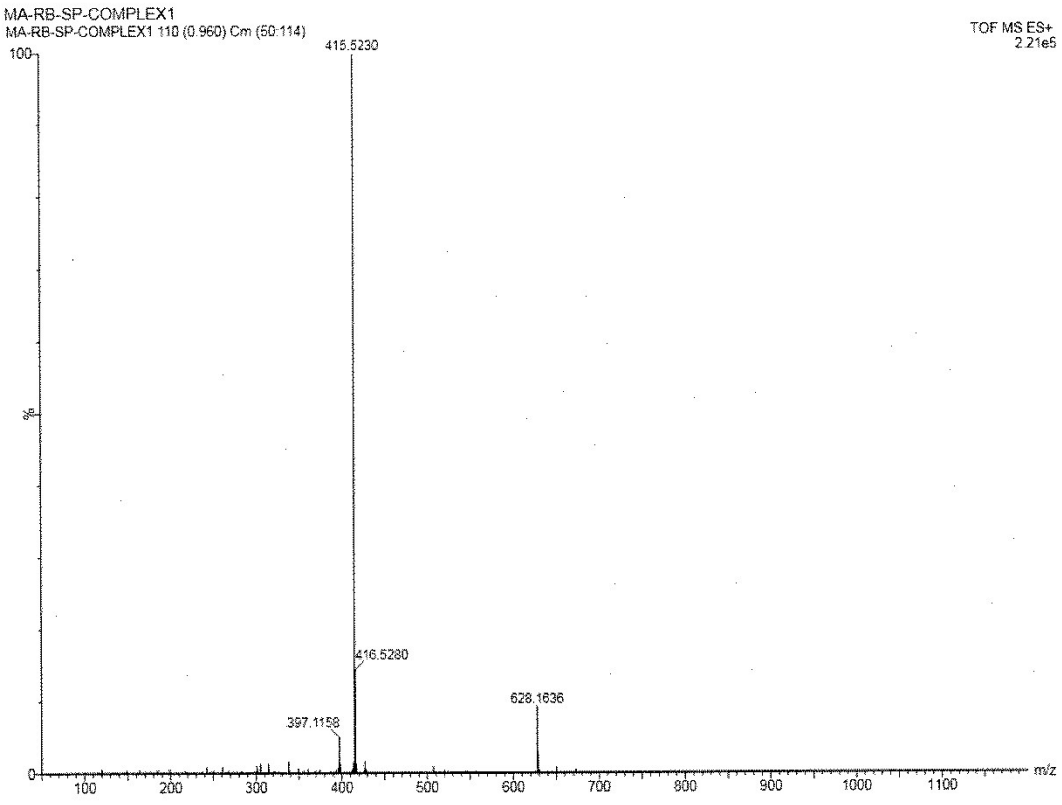


Figure S6. Mass spectra of **[NaL**Fe$(NO_3)_2]^{2+}$ complex

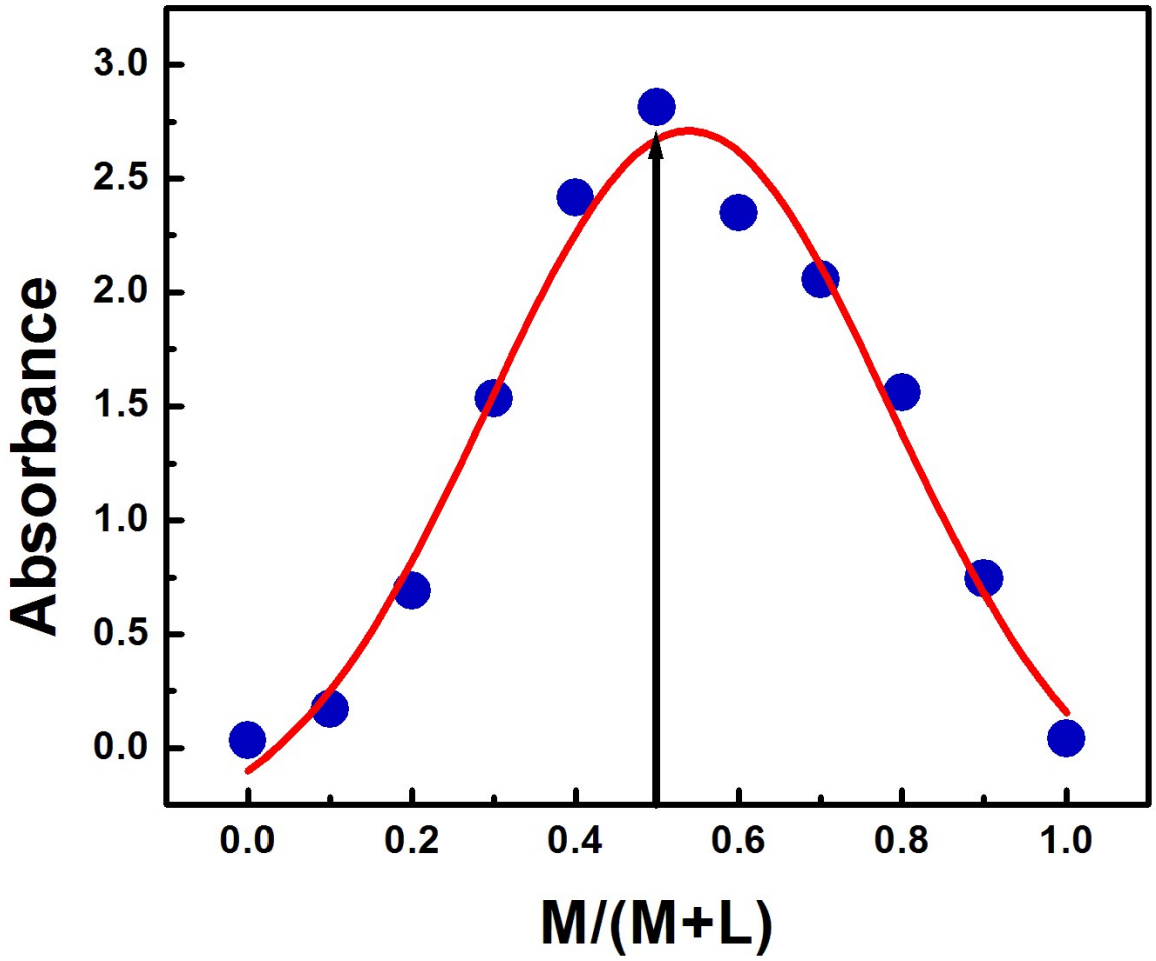


Figure S7. Job's plot for the determination of the composition of the L–Fe^{3+} complex

Absorbance and Fluorescence Spectral Titrations

The absorption spectral titrations were performed at 25 ± 0.5 °C on a Jasco V660 unit (Jasco International Co. Ltd., Hachioji, Japan) equipped with a thermoelectrically controlled cell holder and temperature controller in matched quartz cuvettes of 1 cm path length, following generally the methods standardized in our laboratory and reported earlier.[2,3] The electronic spectra of the $[\mathbf{LFe(NO_3)_2}]^+$ complexes were monitored as a function of the concentration of DNA. In each case a fixed concentration of the $[\mathbf{LFe(NO_3)_2}]^+$ (10 µM) in 10 mM CP buffer was titrated with increasing concentration of DNA over a range of 1– 30 µM. After each addition of the DNA, the solution was incubated at room temperature for 10 min at 25 ± 0.5 °C and the absorbance values were recorded under an equilibrium condition. During the measurement equal amounts of DNA were added to the control solution to eliminate the absorbance of DNA itself. Steady state fluorescence measurements were performed on a Shimadzu RF-5301PC fluorometer in fluorescence free quartz cuvettes of 1 cm path length as described previously.[2,3]

The excitation wavelength for complex was 490 nm. All of the measurements were carried out under conditions of stirring and keeping excitation and emission band passes of 3 nm. The sample temperature of the fluorometer was maintained at 25 ± 1.0 °C using an Eylea Uni Cool U55 water bath (Tokyo Rikakikai Co. Ltd., Tokyo, Japan).

Circular dichroism spectral study

Circular dichroism (CD) spectra were acquired on a Jasco J815 model unit (Jasco International Co. Ltd.) equipped with a Jasco temperature controller (PFD 425L/15) at 25 ± 0.5 ^{o}C.[1] A rectangular strain free quartz cuvette of 1 cm path length was used. Each spectrum was averaged from four successive accumulations at a scan rate of 50 nm/min keeping a band width of 1.0 nm at a sensitivity of 100 milli degree. The base line correction was performed and each spectrum smoothed within allowed limits using the Jasco software of the unit. Titrations were performed by the addition of increasing concentrations of [**LFe(NO$_3$)$_2$]$^+$** to a fixed concentration of DNA (60 µM). The molar ellipticity values [θ] were calculated from the equation [θ]=100×θ/(C×l), where θ is the observed ellipticity in milli degrees, C is the concentration in moles/lit, and l is the cell path length of the cuvette in cm. The molar ellipticity [θ](deg.cm^2/dmol) values are expressed in terms of base pairs in the region 200–400 nm.[1]

DAPI displacement assay

The well-known minor groove binders, DAPI was used to perform the minor groove displacement assays as reported earlier.[2,3] In a typical experiment, the concentration of DNA and DAPI was 20 µM and 15 µM, respectively. Fluorescence spectra were recorded in 400–650 nm using an excitation wavelength of 338 nm keeping excitation and emission band passes of 5 nm. The changes in the emission spectra of DAPI complexed with the DNA were monitored upon addition of increasing concentrations of [**LFe(NO$_3$)$_2$]$^+$** at 25±0.5 °C.

Reference

1 A. Basu, P. Jaisankar and G. Suresh Kumar, *Bioorg. Med. Chem.,* 2012, 20, 2498.

2 A. Basu and G. Suresh Kumar, *J. Agric. Food Chem.*, 2014, 62, 317.

3 J. Zhou, A. Chang, L. Wang, Y. Liu, X. Liu and D. Shangguan, *Org. Biomol. Chem.*, 2014, 12, 9207.

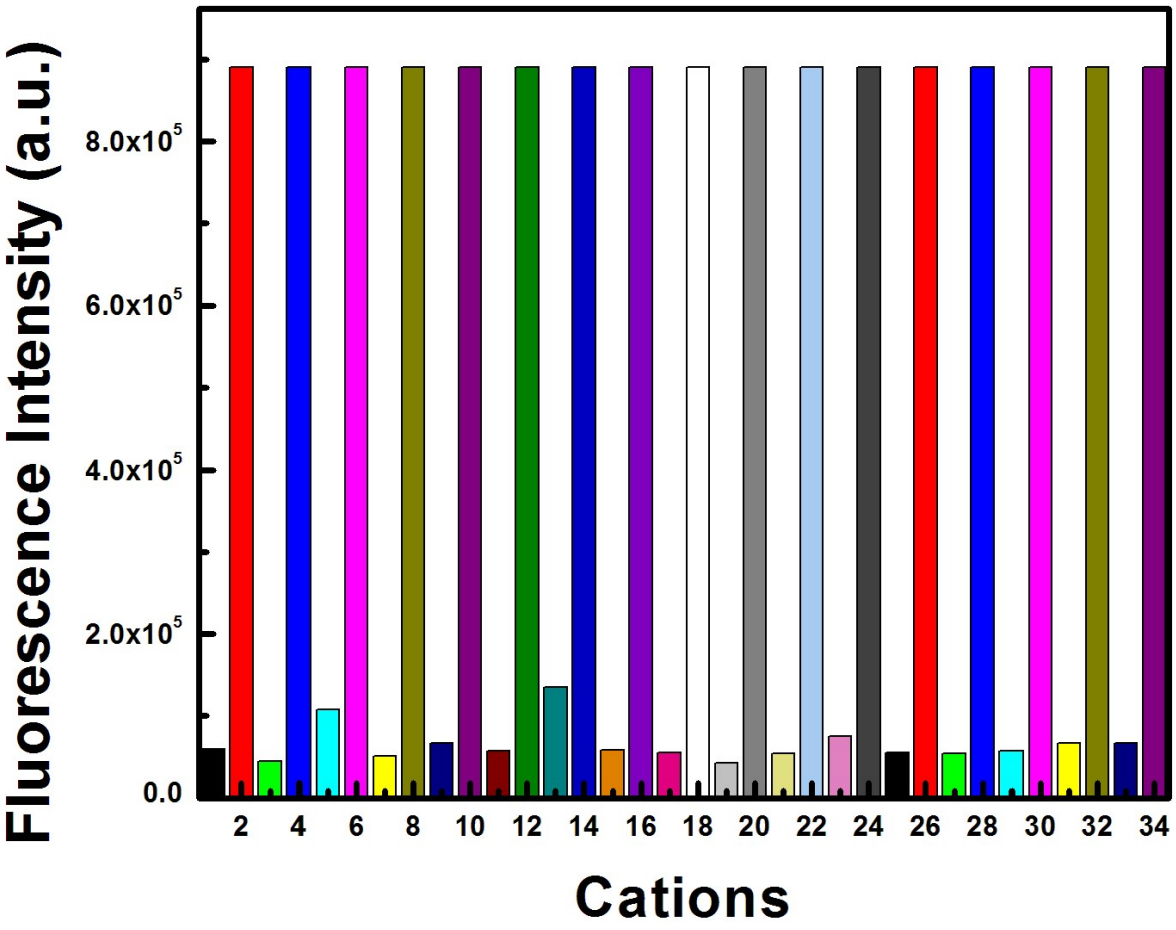


Figure S8. Fluorescence emission of L (50 μM) induced by different cations (250 μM) (1-34 are: L, L+Fe^{3+}, L+Na^{+}, L+Na^{+}+Fe^{3+}, L+Cr^{3+}, L+Cr^{3+}+Fe^{3+}, L+Ca^{2+}, L+Ca^{2+}+Fe^{3+}, L+Mg^{2+}, L+Mg^{2+}+Fe^{3+}, L+K^{+}, L+K^{+}+Fe^{3+}, L+Al^{3+}, L+Al^{3+}+Fe^{3+}, L+Mn^{2+}, L+Mn^{2+}+Fe^{3+}, L+ Fe^{2+}, L+ Fe^{2+}+Fe^{3+}, L+ Co^{2+}, L+ Co^{2+}+Fe^{3+}, L+Cu^{2+}, L+Cu^{2+}+Fe^{3+}, L+Ni^{2+}, L+Ni^{2+}+Fe^{3+}, L+Zn^{2+}, L+Zn^{2+}+Fe^{3+}, L+Cd^{2+}, L+Cd^{2+}+Fe^{3+}, L+Hg^{2+}, L+Hg^{2+}+Fe^{3+}, L+Pb^{2+}, L+Pb^{2+}+Fe^{3+}, L+ Ag^{+}, L+ Ag^{+}+Fe^{3+}).

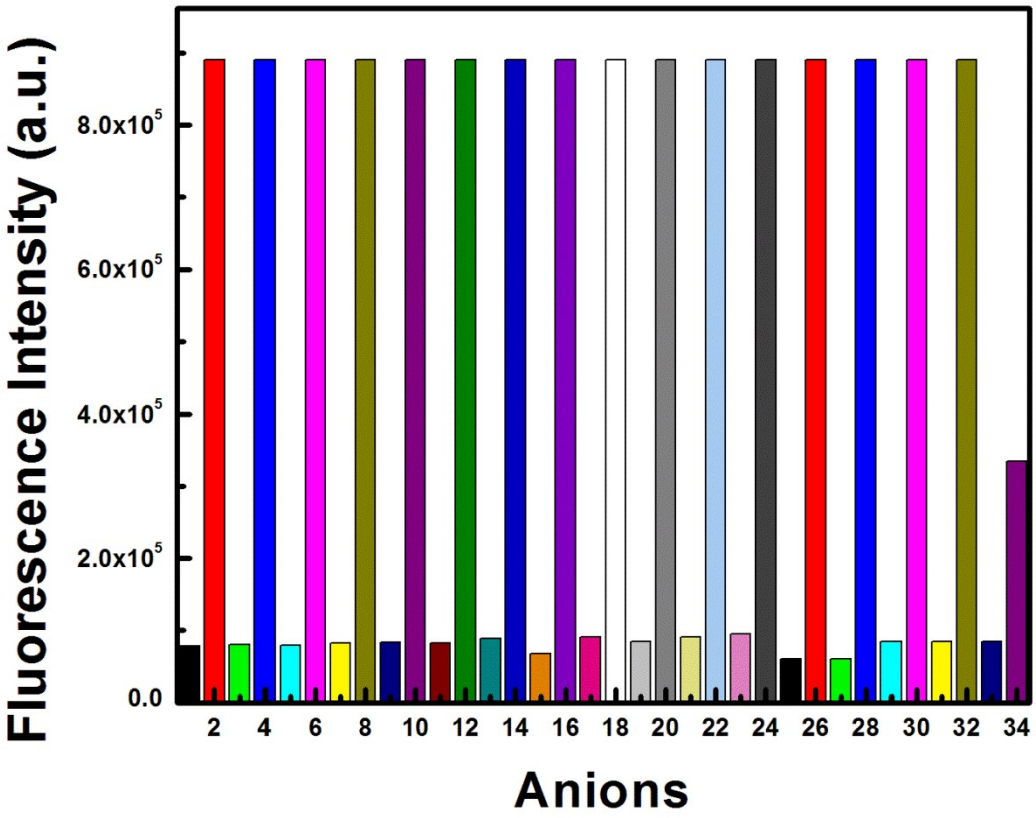


Figure S9. Fluorescence emission of L (50 μM) induced by different anions (250 μM). (1-34 are L, L+Fe^{3+}, L+ SO_4^{2-}, L+ SO_4^{2-}+Fe^{3+}, L+ NO_3^-, L+ NO_3^-+Fe^{3+}, L+ PO_4^{3-}, L+ PO_4^{3-}+Fe^{3+}, L+ S^{2-}, L+ S^{2-}+Fe^{3+}, L+ Cl^-, L+ Cl^- +Fe^{3+}, L+ Br^-, L+ Br^-+Fe^{3+}, L+ I^-, L+ I^-+Fe^3 , L+ OAc^-, L+ OAc^-+Fe^{3+}, L+ $H_2AsO_4^-$, L+ $H_2AsO_4^-$+Fe^{3+}, L+ ClO_4^-, L+ ClO_4^-+Fe^{3+}, L+ $S_2O_4^{2-}$, L+ $S_2O_4^{2-}$+Fe^{3+}, L+ HCO_3^-, L+ HCO_3^-+Fe^{3+}, L+ SCN^-, L+ SCN^-+Fe^{3+}, L+ $P_2O_7^{4-}$, L+ $P_2O_7^{4-}$ +Fe^{3+}, L+CO_3^{2-}, L+CO_3^{2-}+Fe^{3+}, L+ F^{-+} ,L+ F^-+Fe^{3+}).

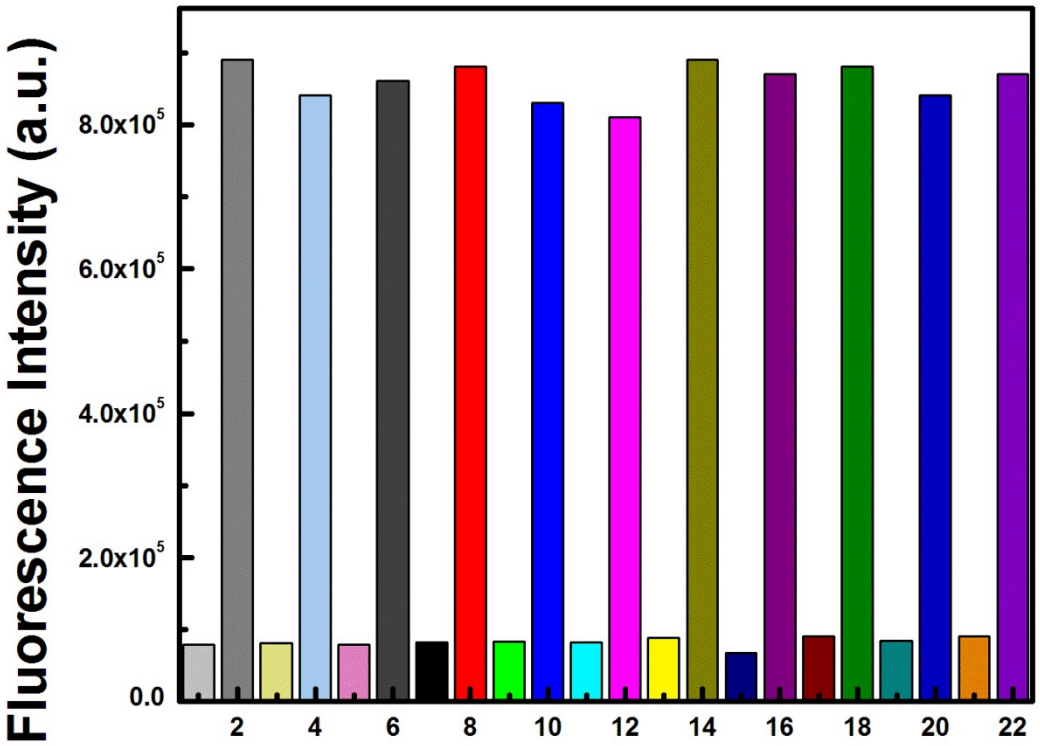


Figure S10. Fluorescence emission of L (50 μM) induced by different ROS, RNS, reducing ion and biological molecules (50 μM). (1-34 are L, L+Fe^{3+}, L+ HS^-, L+ HS^-+Fe^{3+}, L+HSO_3^-, L+ HSO_3^-+Fe^{3+}, L+ Cu^+, L+ Cu^++Fe^{3+}, L+ $^\bullet OH$, L+ $^\bullet OH$ +Fe^{3+}, L+ H_2O_2, L+ H_2O_2+Fe^{3+}, L+ cysteine, L+ cysteine +Fe^{3+}, L+ ascorbic acid, L+ ascorbic acid +Fe^3 , L+ dehydro ascorbic acid, L+ dehydro ascorbic acid +Fe^{3+}, L+ $O_2^{\cdot-}$, L+ $O_2^{\cdot-}$+Fe^{3+}, L+ ClO^-, L+ ClO^-+Fe^{3+}, .

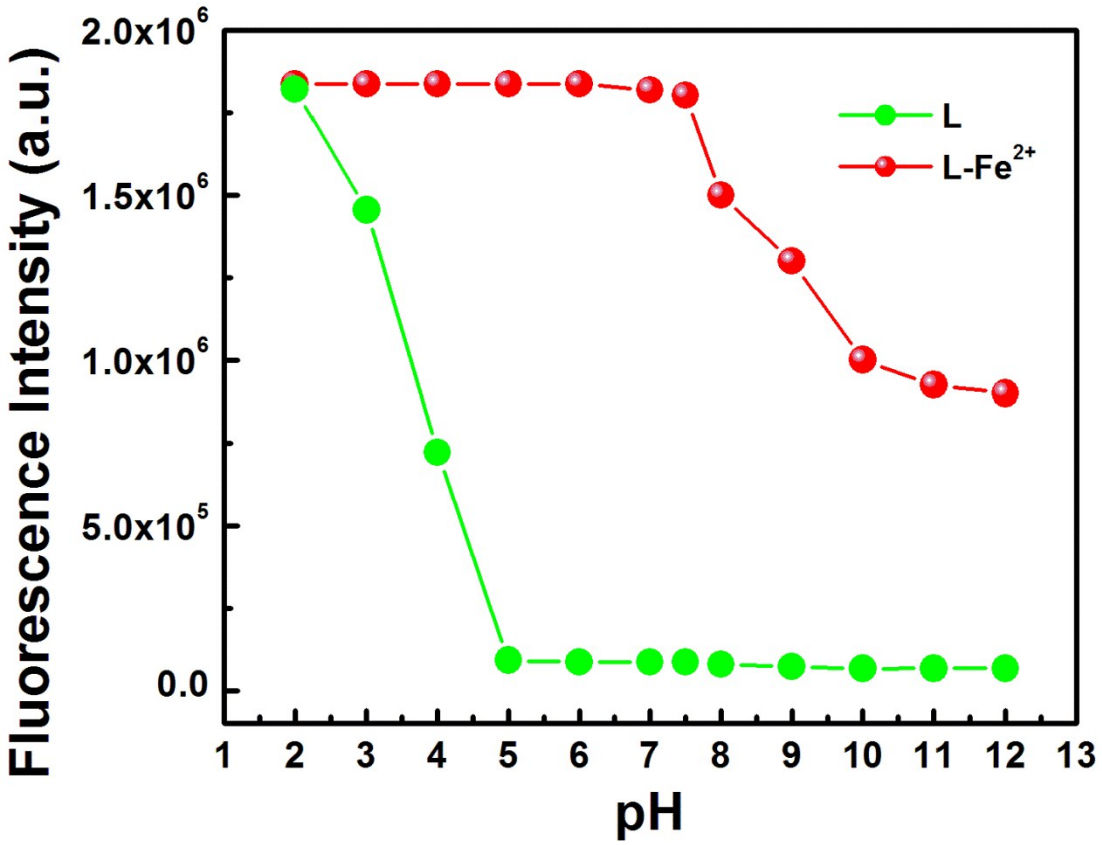


Figure S11. pH dependence of the FIs of the free ligand L (green) and the $[\mathbf{LFe(NO_3)_2}]^+$ complex with $L:Fe^{3+}$= 1:1.05 (red) in the HEPES buffer medium with λ_{ex} = 510 nm.

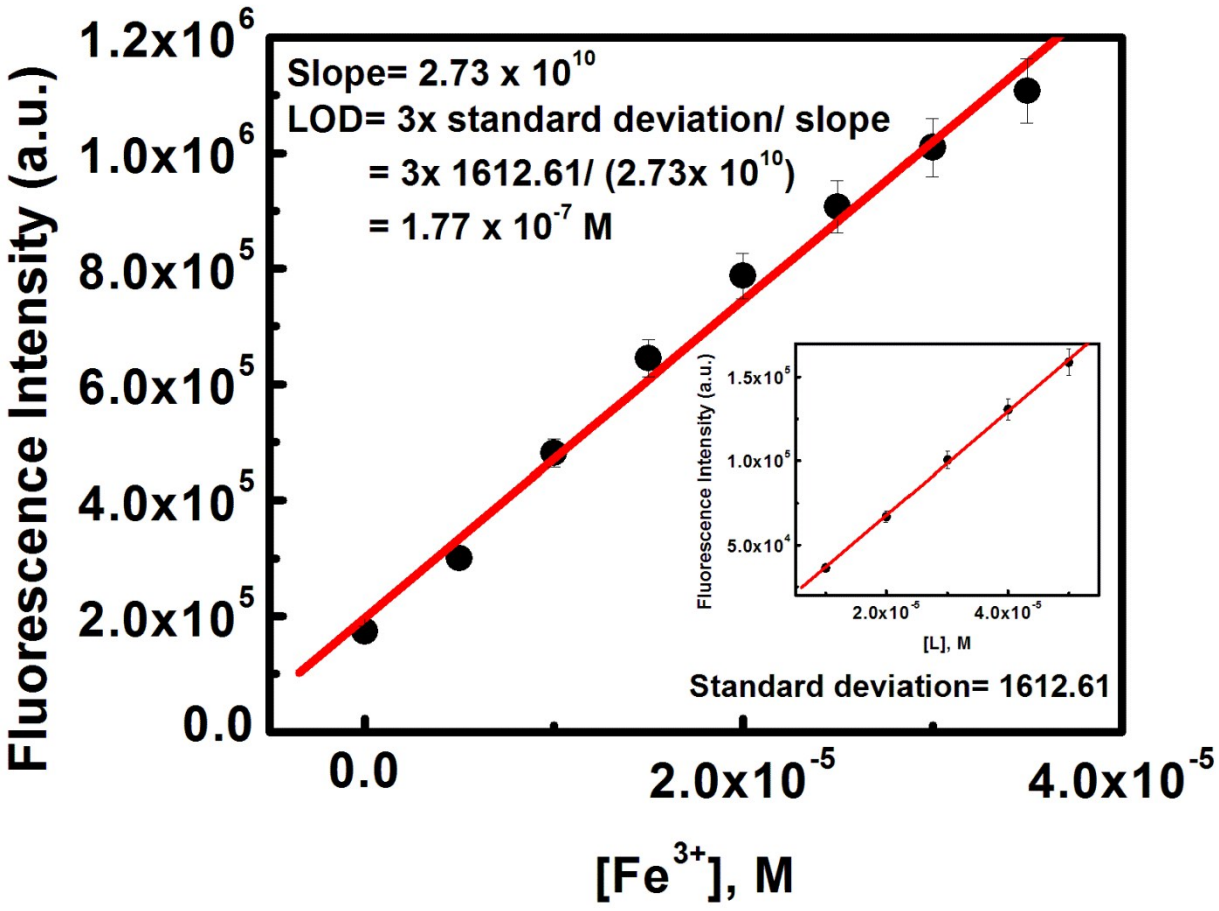


Figure S12. Limit of detection of Fe^{3+}.

Quantum Yield Calculation:

Fluorescence quantum yields (**Φ**) were estimated by integrating the area under the fluorescence curves with the equation: $\Phi_{sample} = \frac{OD_{std}}{OD_{sample}} \times \frac{A_{sample}}{A_{std}} \times \Phi_{std}$

where, A is the area under the fluorescence spectral curve and OD is optical density of the compound at the excitation wavelength. The standard used for the measurement of fluorescence quantum yield was Rhodamine 6G Φ_{std}=0.92 in Water).

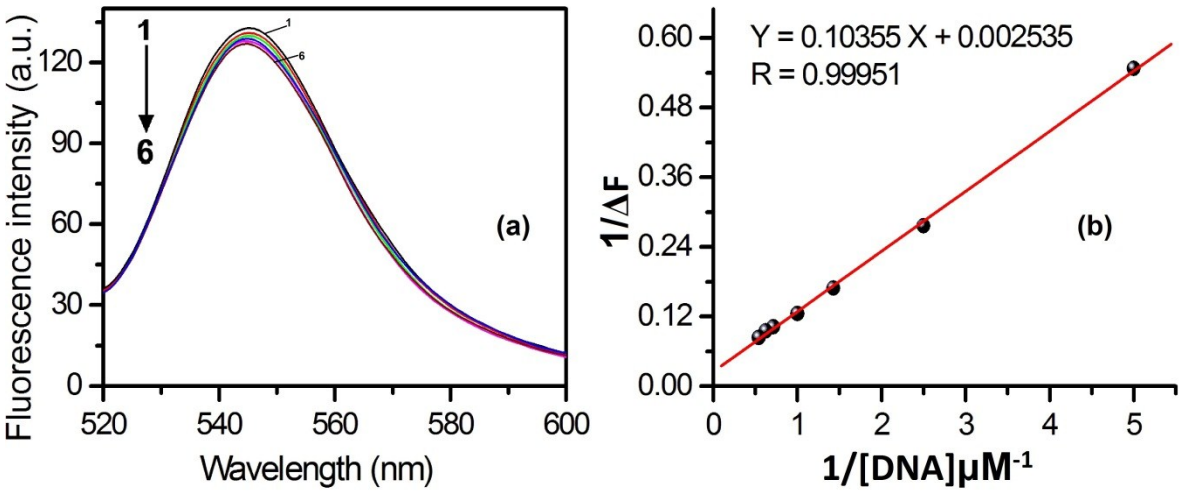


Figure S13 (a) Fluorescence titration profile of **[L]** (10 μM) in the presence of increasing concentration of CT DNA [0-30 μM]; (b) Modified Benesi–Hildebrand plots from fluorescence titration data are shown in inset. Excitation wavelength was 500 nm.

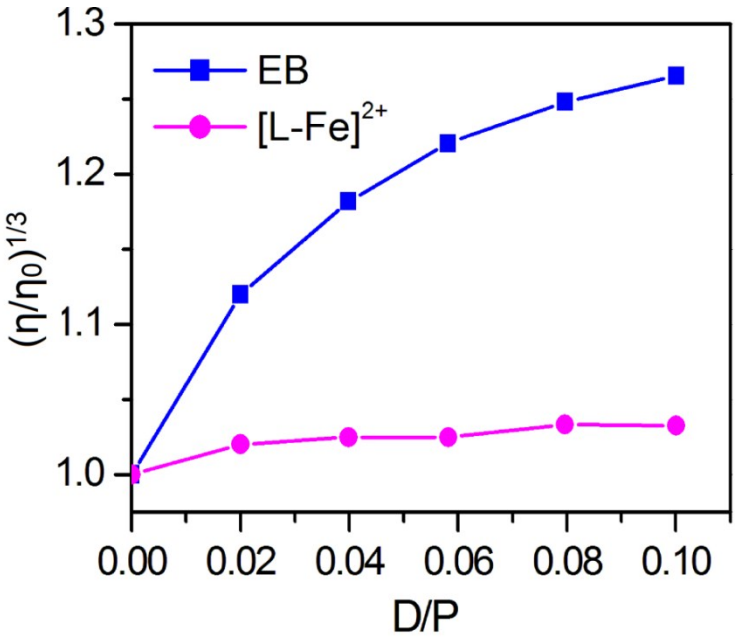


Figure S14. A plot of relative specific viscosity of CT DNA with increasing concentration of EB and $[\mathbf{LFe(NO_3)_2}]^+$. Each experimental point is average of three determinations. The DNA concentration was 100 μM. D/P= 0.6.

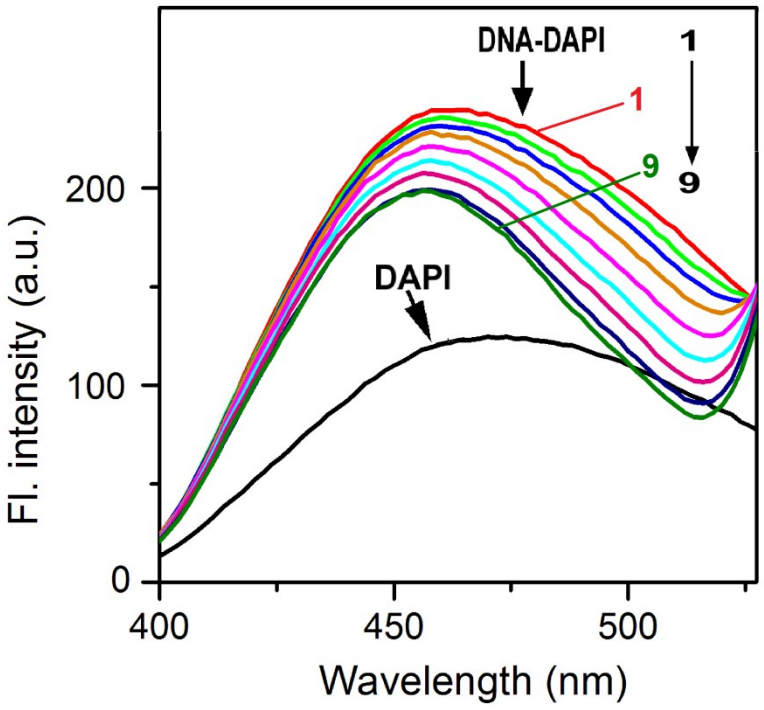


Figure S15 Fluorescence emission spectra of the competition between the DAPI–DNA complex (λ_{exc}: 338 nm) and $[\mathbf{LFe(NO_3)_2}]^+$ treated with: 0.0-20 μM (curves 1–9) of $[\mathbf{LFe(NO_3)_2}]^+$. C_{DAPI}= 15 μM and C_{DNA}: 20 μM.

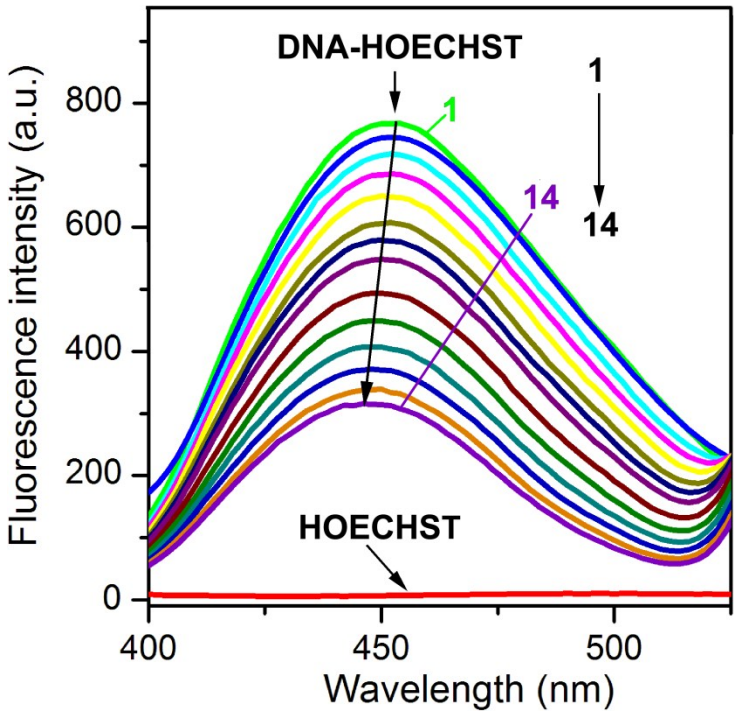


Figure S16. Fluorescence emission spectra of the competition between the Hoechst–DNA complex (λ_{exc}: 350 nm) and **[LFe(NO$_3$)$_2$]$^+$** treated with: 0.0-25 μM (curves 1–14) of **[LFe(NO$_3$)$_2$]$^+$**. $C_{Hoechst}$= 1.91 μM and C_{DNA}: 20 μM.

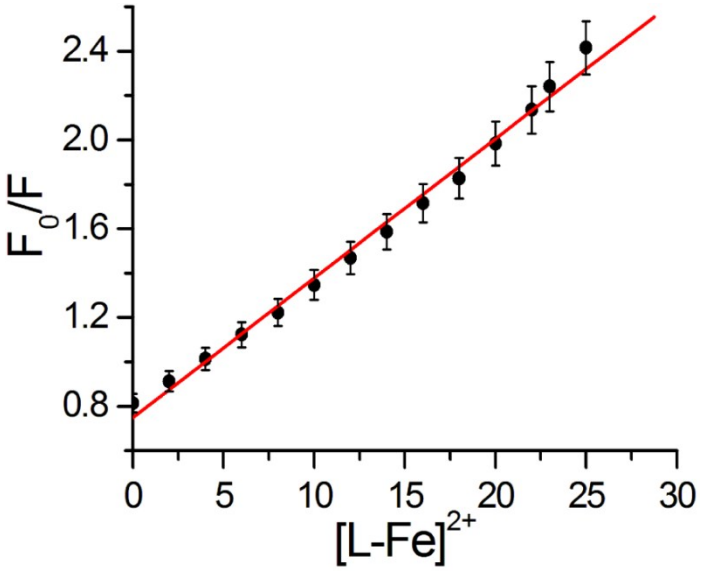
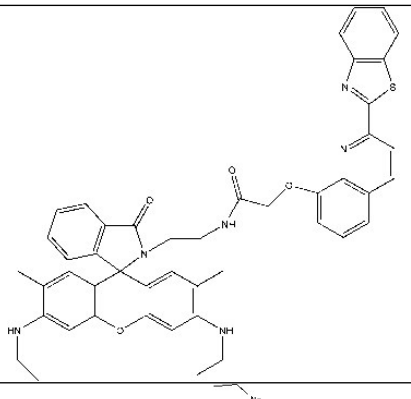
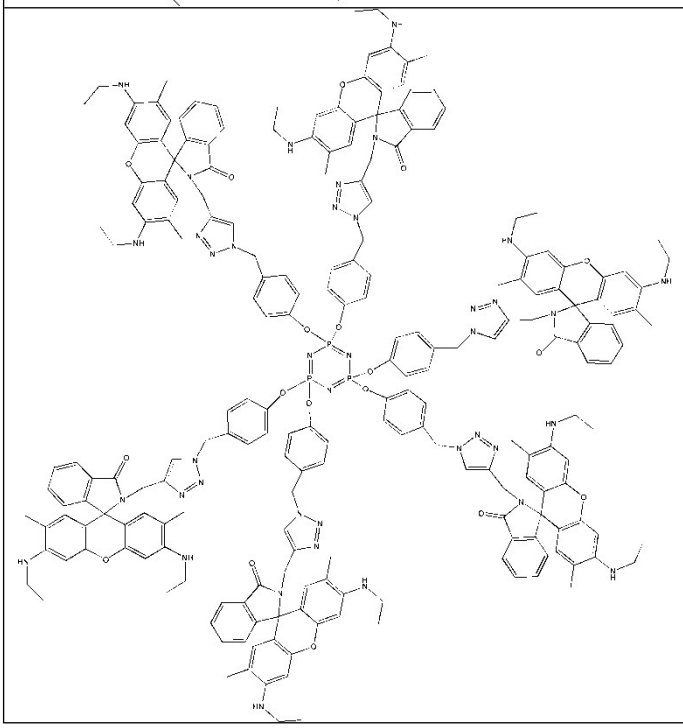
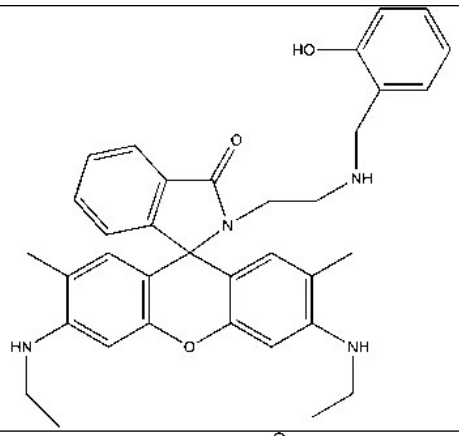
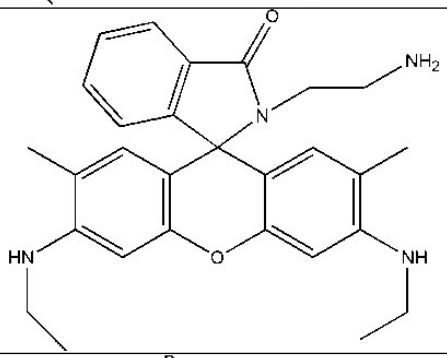
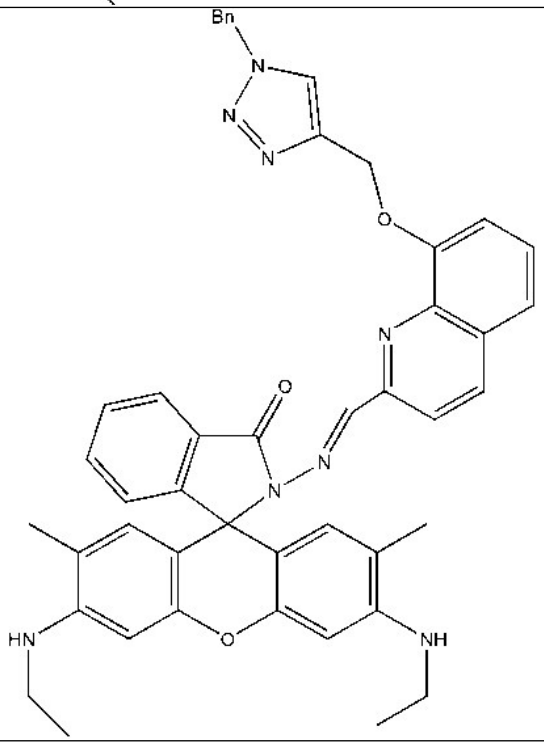
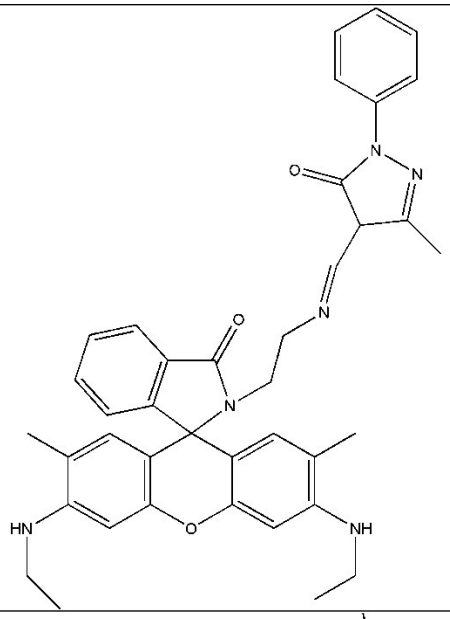
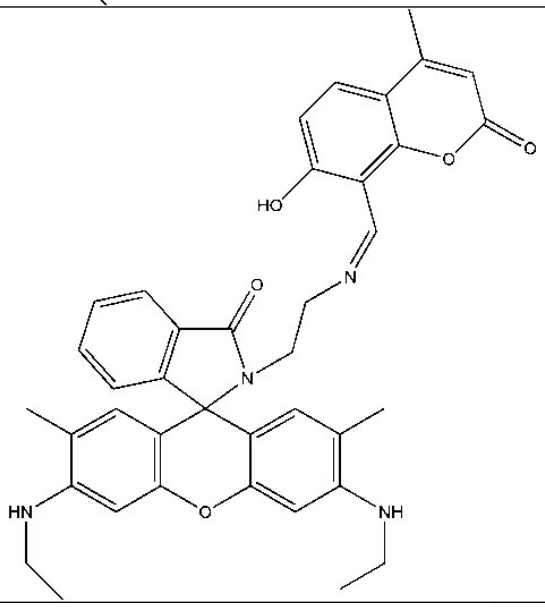
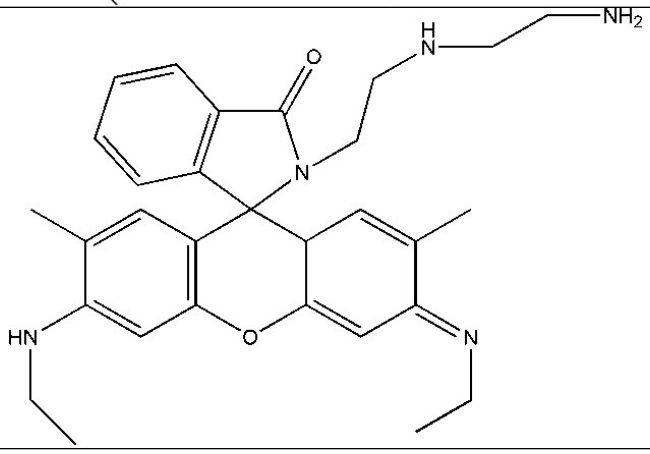


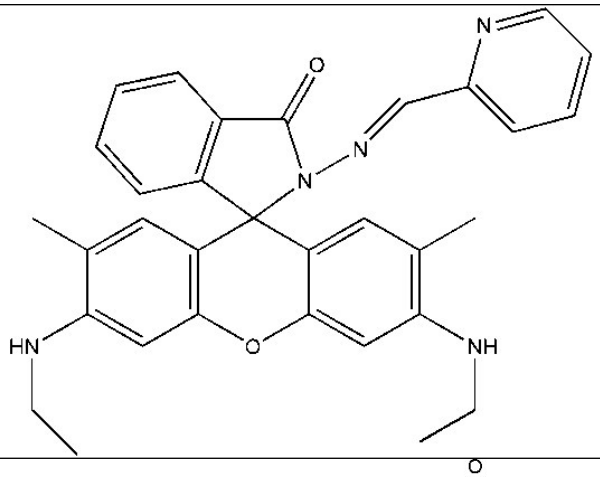
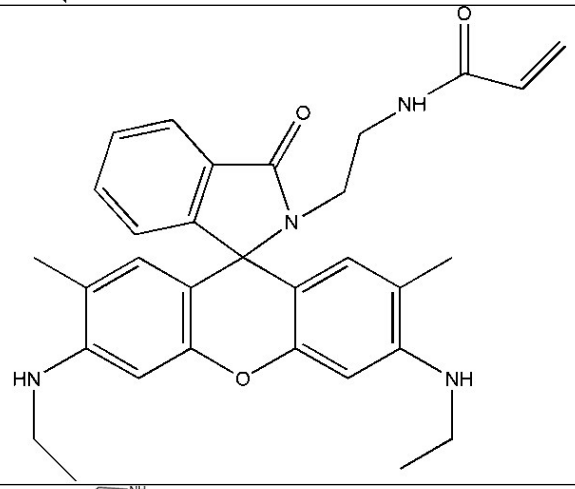
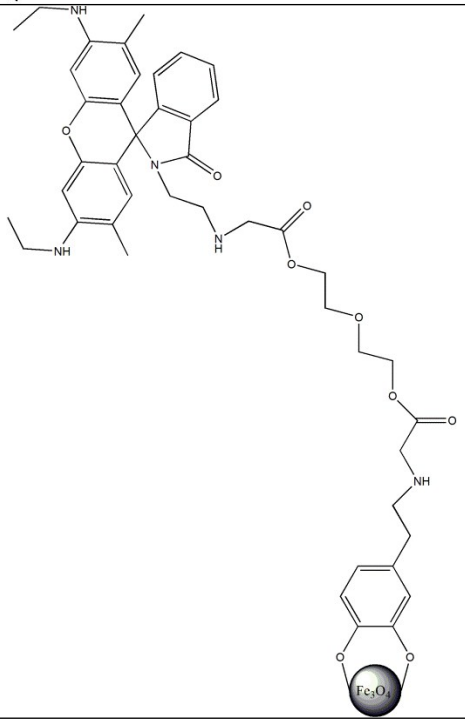
Figure S17. Stern–Volmer plot for the fluorescence quenching of the Hoechst–DNA system by **[LFe(NO$_3$)$_2$]$^+$**.

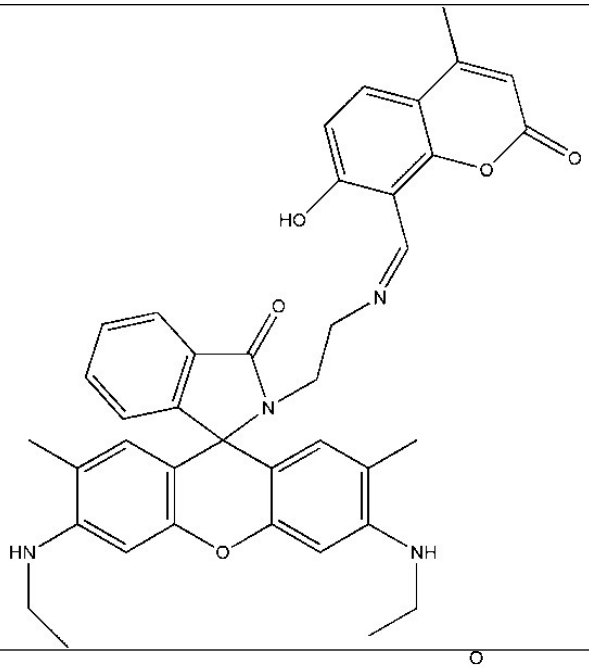
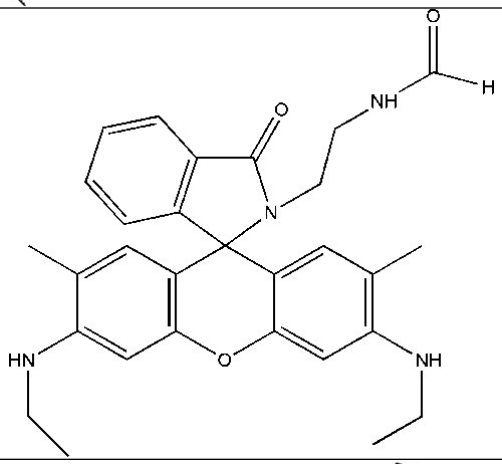
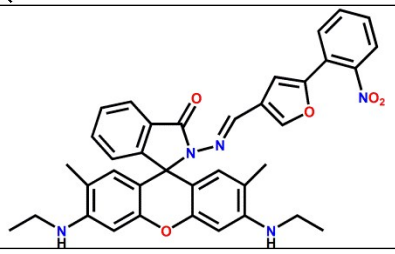
Table S1. Comparison of results of the probe L with other reported Fe^{3+} sensors.

Ligand	Solvent(s)	Limit of Detection	Ref
	CH_3OH/H_2O (2/3)	0.01μM	53
	THF/H_2O (99:1)	4.8μM	54

	EtOH	-	55
	10% Aqueous ethanol	-	56
	Tris-HCl: CH_3CN(1:1)	0.05µM	57

	H_2O: CH_3CN(1:1)	6.1µM	58
	Ethanol	5.6ppb	59
	DMF/H_2O	-	60

	H_2O: CH_3CN(1:1)	0.09 µM	61
	Hydrogel formation insluble in H_2O	-	62
	Aqueous	2ppb	63

	Aqueous	0.72µM	64
	Aqueous	0.01µM	65
	Aqueous	0.17 µM	This work