Supporting Information

Visual and Near IR (NIR) Fluorescence detection of Cr^{3+} in Aqueous Media via Spirobenzopyran ring opening with Application in Logic gate and Bio-imaging

Shyamaprosad Goswami[a], Avijit Kumar Das[a], Anup Kumar Maity[b], Abhishek Manna[a], Krishnendu Aich[a], Sibaprasad Maity[a,c], Partha Saha[b],Tarun Kanti Mandal[c]*

[a]*Department of Chemistry, Bengal Engineering and Science University, Shibpur, Howrah 711103, West Bengal, India E-mail: spgoswamical@yahoo.com; Fax: +91-3326682916.*

[b]*Crystallography and Molecular Biology Division, Saha Institute of Nuclaer Physics, Kolkata 700064, West Bengal, India E–mail: partha.saha@saha.ac.in*

[c] *Haldia Institute of Technology, Hatiberia, Haldia, West Bengal-721657*

CONTENTS

1. General procedure for drawing Job plot by UV–vis method:

Stock solution of same concentration of **SPNH** and Cr^{3+} were prepared in the order of ≈ 2.0 x 10^{-5} M in CH_3CN-HEPES buffer (7/3,v/v, 25°C) at pH 7.4. The absorbance in each case with different *host–guest* ratio but equal in volume was recorded. Job plots were drawn by plotting $\Delta I.X_{host}$ *vs* X_{host} (ΔI = change of intensity of the absorbance spectrum during titration and X_{host} is the mole fraction of the host in each case, respectively).

2. Association constant determination by Uv-vis methods:

Binding constant was calculated according to the Benesi-Hildebrand equation[1]. *Ka* was calculated following the equation stated below.

$$1/(A\text{-}Ao) = 1/\{K(Amax–Ao)\,[M]_n\} + 1/[Amax\text{-}Ao]$$

Here Ao is the absorbance of receptor in the absence of guest, A is the absorbance recorded in the presence of added guest, Amax is absorbance in presence of added [M]max and K is the association constant (M^{-1}). The association constant (K) could be determined from the slope of the straight line of the plot of 1/(A-Ao) against $1/[M]_n$. The association constant (K_a) as determined by UV-vis titration method for sensor with Cr^{3+} is found to be 2.2 x 10^4 M^{-1}.

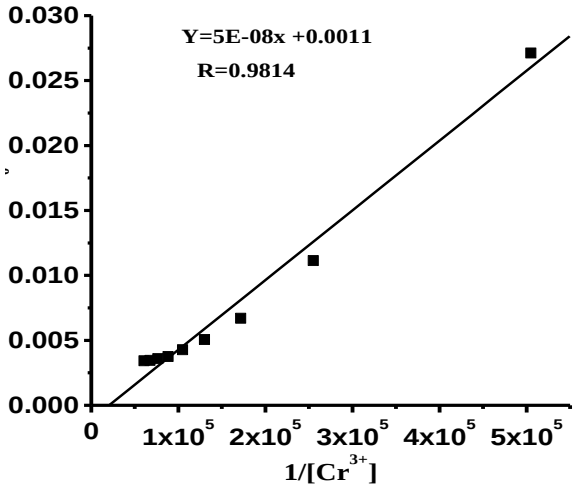


Figure S1: Benesi–Hildebrand plot from UV-vis titration data of **SPNH** (c = 1x10^{-5}M) with Cr^{3+}(c = 2x10^{-4}M).

3. Association constant determination by Fluorescence methods:

The binding constant value of Cr^{3+} with **SPNH** has been determined from the emission intensity data following the modified Benesi–Hildebrand equation, $1/\Delta I = 1/\Delta I\ max + (1/K[C])\ (1/\Delta I\ max)$. Here $\Delta I = I\text{-}Imin$ and $\Delta I\ max = Imax\text{-}Imin$, where Imin, I, and Imax are the emission intensities of sensor considered in the absence of Cr^{3+}, at an intermediate Cr^{3+} concentration, and at a concentration of complete saturation where K is the binding constant and [C] is the Cr^{3+} concentration, respectively. From the plot of $1/(I\text{-}I_{min})$ against $[C]^{-1}$ for sensor, the value of K has been determined from the slope. The association constant (K_a) as determined by fluorescence titration method for sensor with Cr^{3+} is found to be $7.7 \times 10^4\ M^{-1}$ (error < 10%).

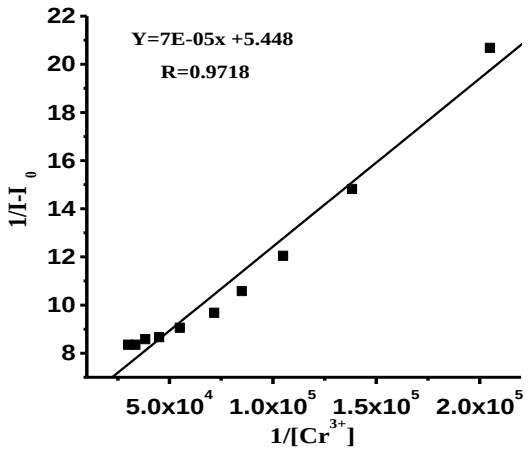


Figure S2: Benesi–Hildebrand plot from fluorescence titration data of **SPNH** (c =1x10^{-5}M) with Cr^{3+}(c = 2x10^{-4}M).

4. Determination of fluorescence quantum yield:

Here, the quantum yield φ was measured by using the following equation,

$$\varphi_x = \varphi_s\ (F_x / F_s)(A_s / A_x)(n_x^2 / n_s^2)$$

Where,

X & S indicate the unknown and standard solution respectively, φ = quantum yield,

F = area under the emission curve, A = absorbance at the excitation wave length,

n = index of refraction of the solvent. Here φ measurements were performed using rhodamine-B in ethanol as standard [φ = 0.65] (error ~ 10%).

5. Calculation of the detection limit:

The detection limit (DL) of **SPNH** in absorption and emission spectra for Cr^{3+} was determined from the following equation:

DL = K* Sb1/S

Where K = 2 or 3 (we take 3 in this case); Sb1 is the standard deviation of the blank solution; S is the slope of the calibration curve.

From the graph Fig.S3(a), we get slope = 0.0059, and Sb1 value is 0.011985.

Thus using the formula we get the Detection Limit for Cr^{3+} = 6.09 µM in Uv-vis absorption spectra.

From the graph Fig.S3(b), we get slope = 18.034, and Sb1 value is 27.6788.

Thus using the formula we get the Detection Limit for Cr^{3+} is 4.6 µM in Fluorescence spectra.

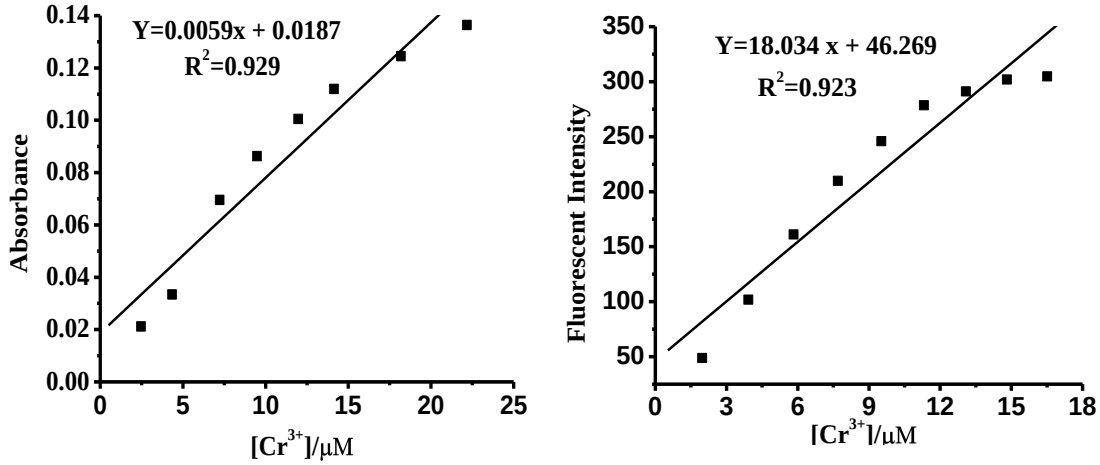
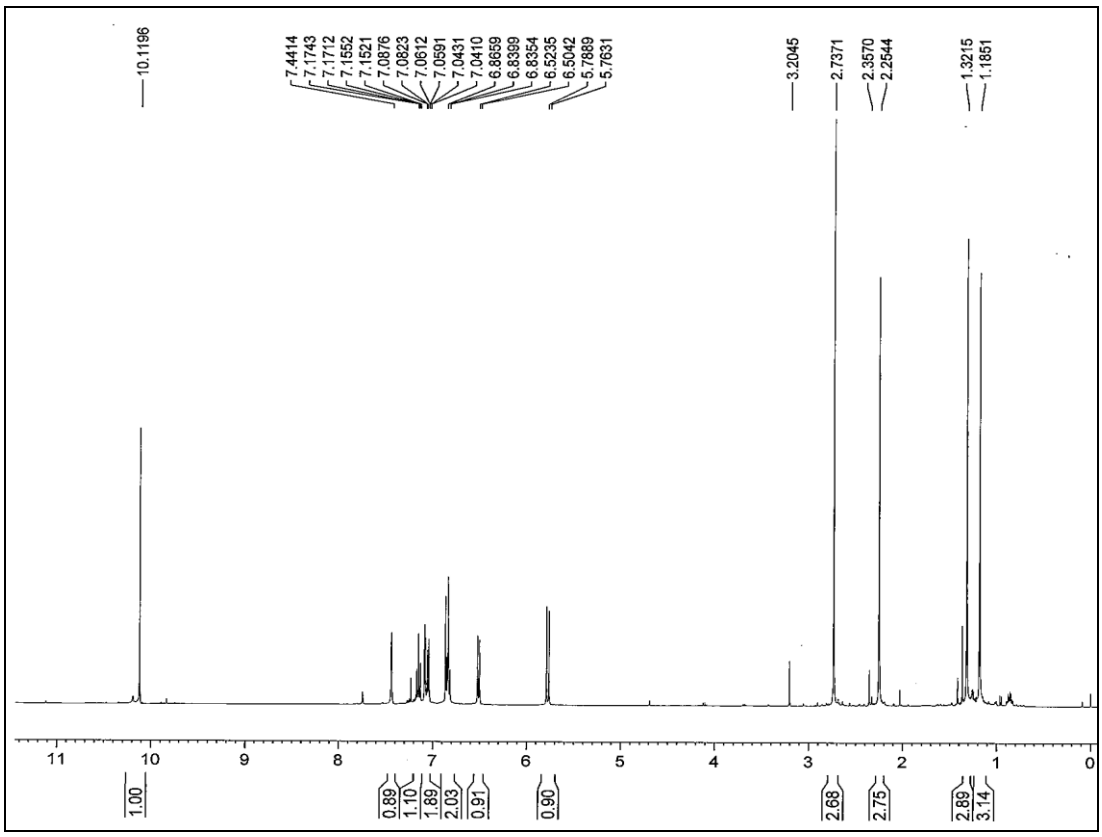
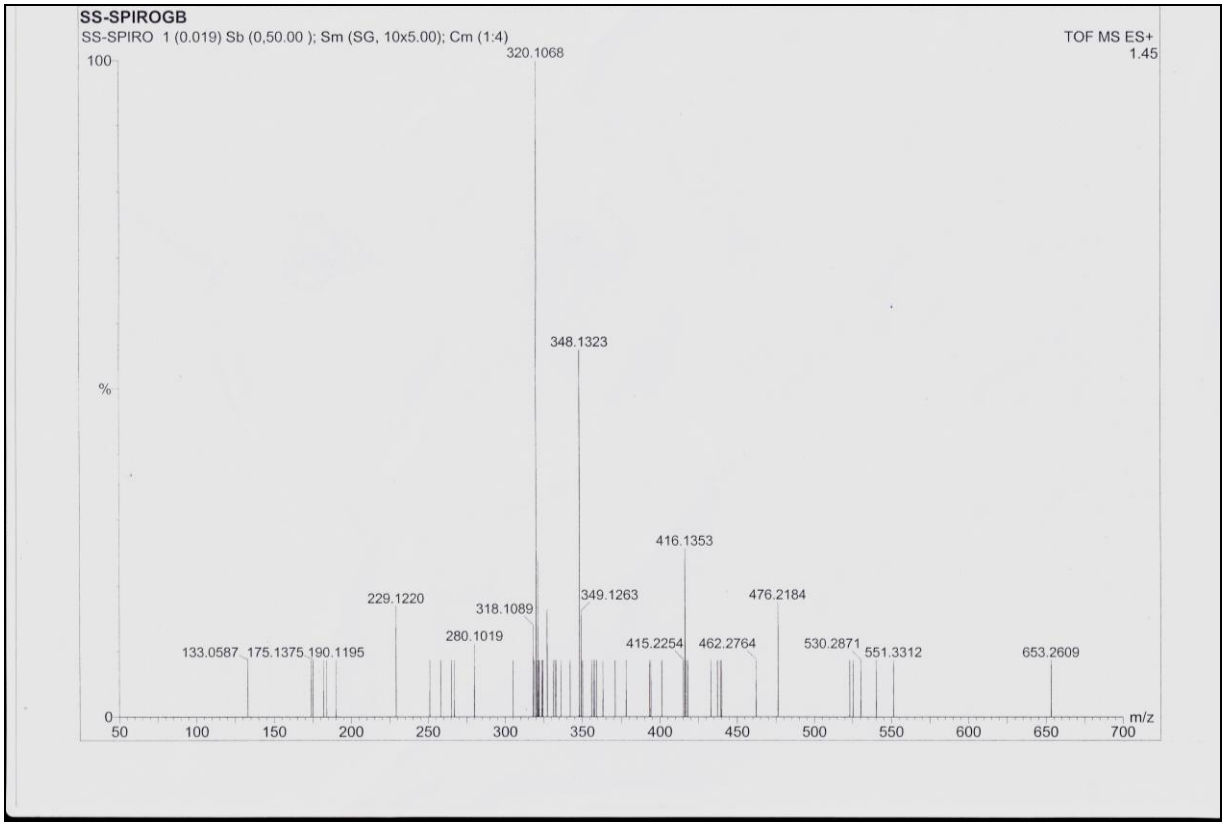


Figure S3 (a) Changes of absorbance of **SPNH** (c = $1x10^{-5}$M) as a function of Cr^{3+}(c = $2x10^{-4}$M) at 440 nm. **(b)** Changes of Fluorescence Intensity of **SPNH** (c =$1x10^{-5}$M) as a function of Cr^{3+}(*c* = $2x10^{-4}$M) at 675 nm.

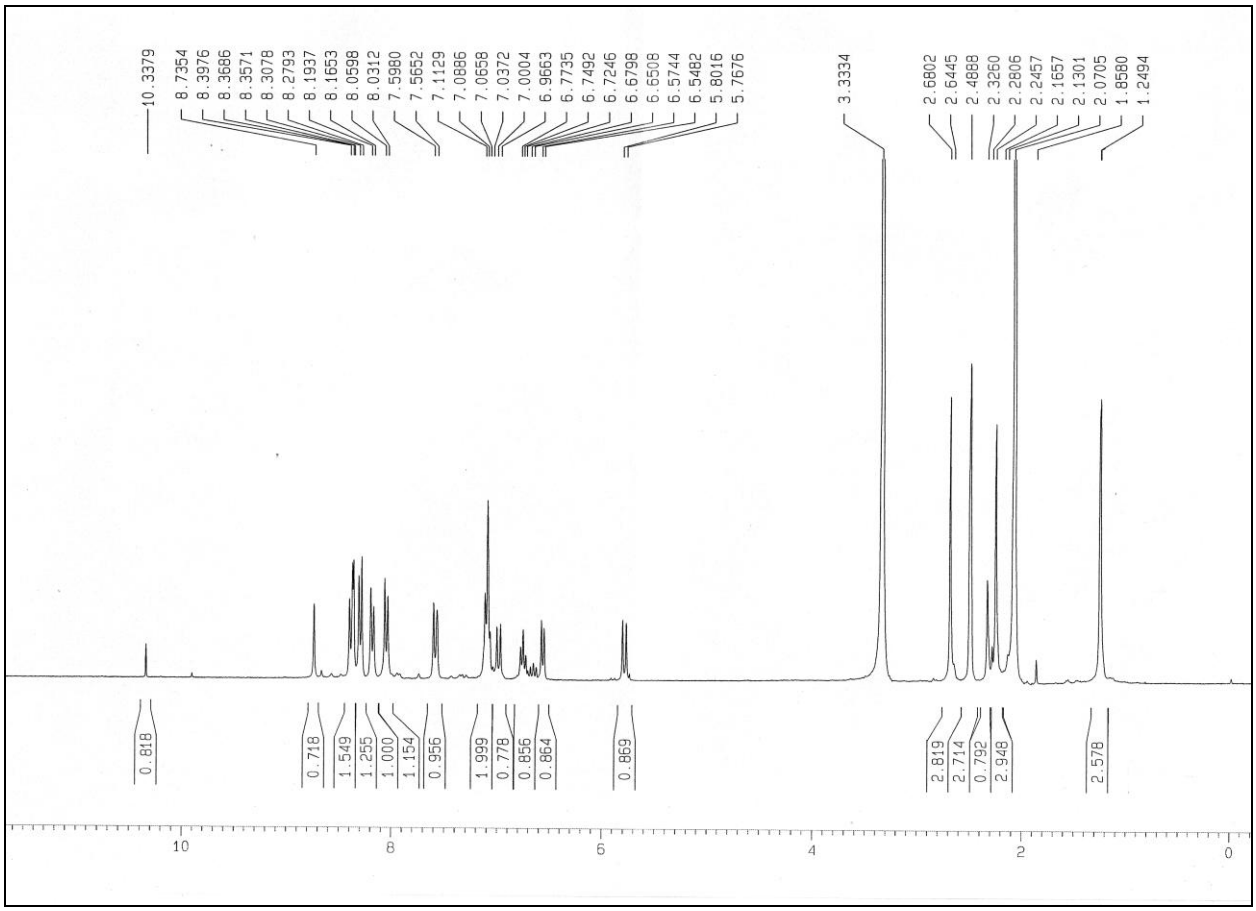
6. ^{1}H NMR spectrum (S4) of compound A:



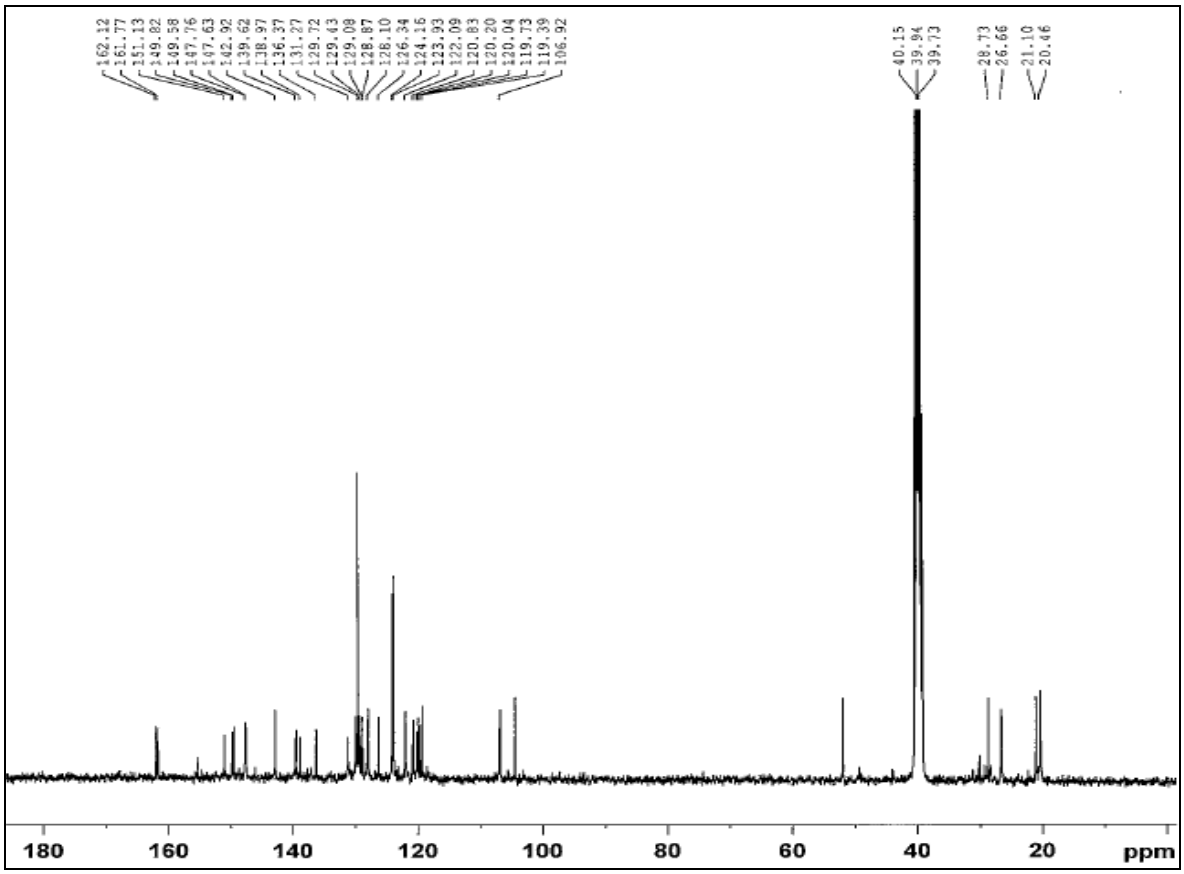
7. Mass spectrum (S5) of compound A:



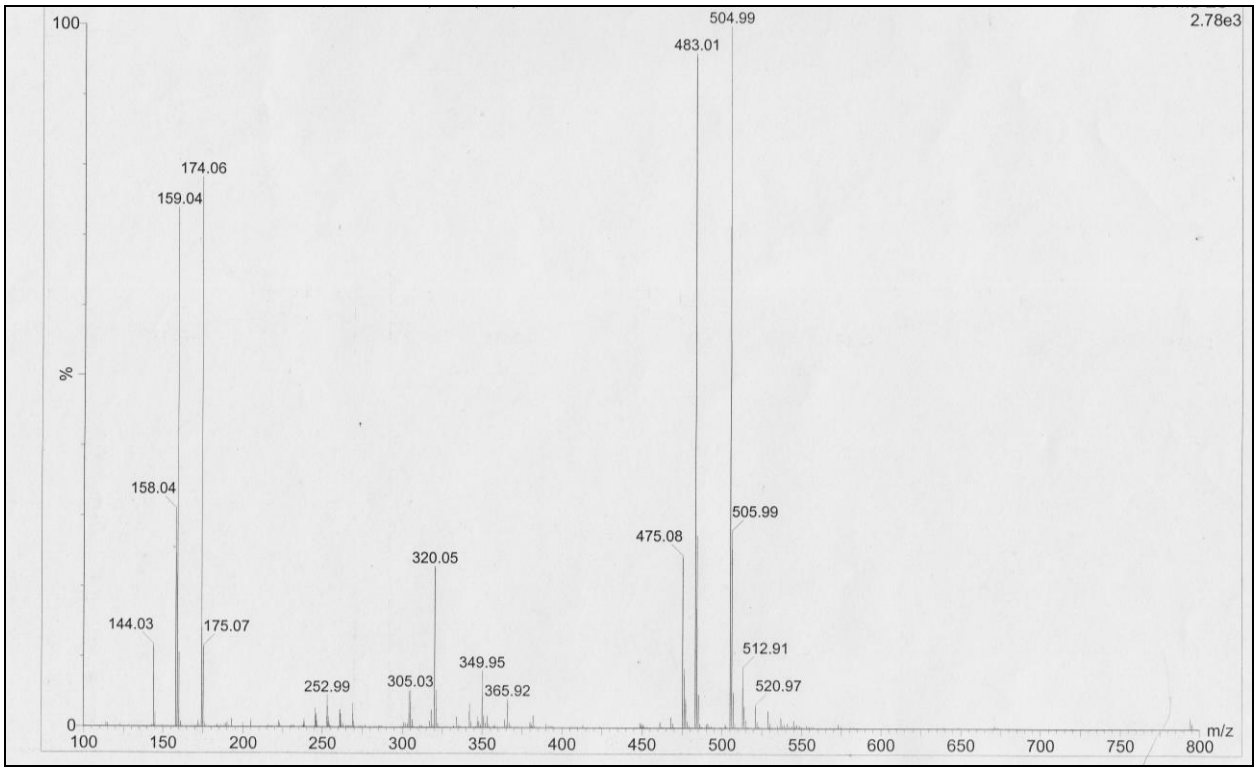
8. ^{1}H NMR spectrum (S6) of SPNH:



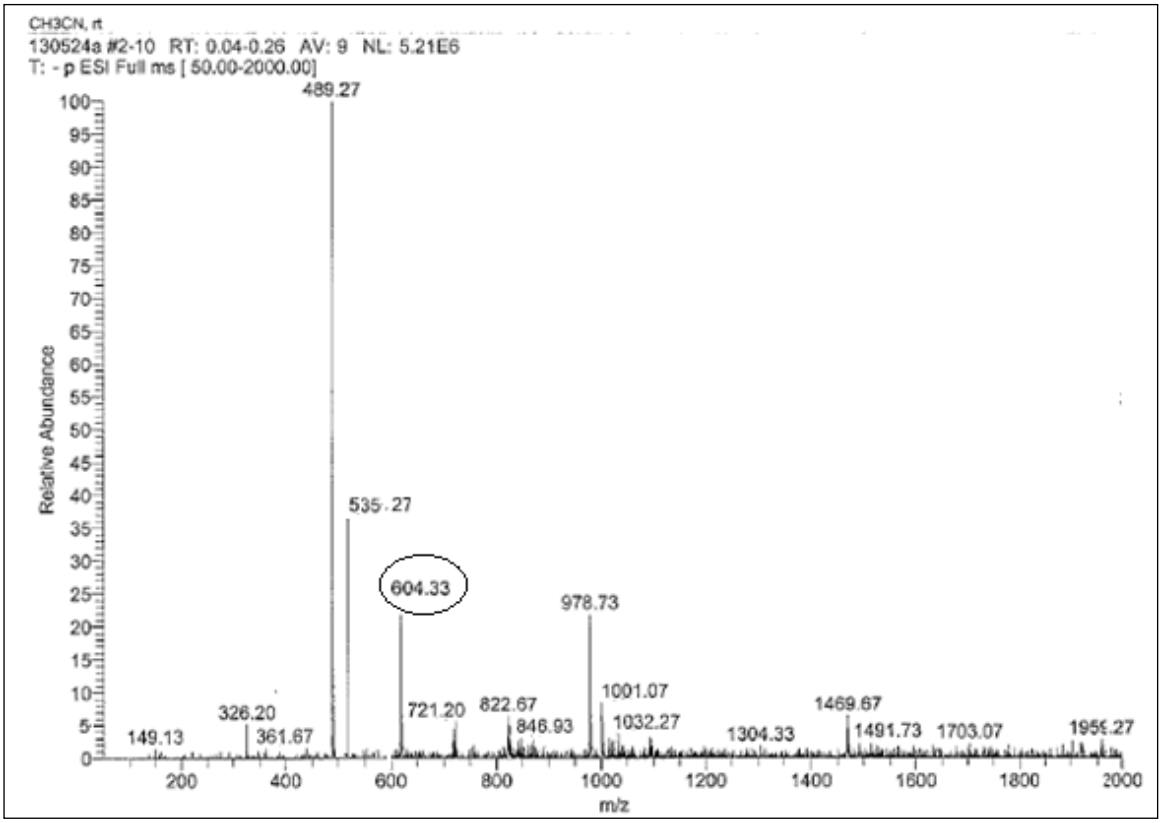
9. ^{13}C NMR spectrum(S7) of SPNH:



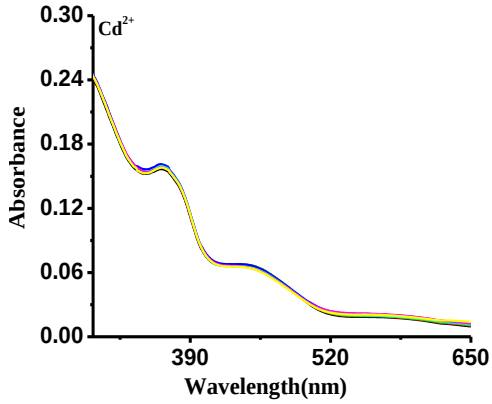
10. Mass spectrum (S8) of Sensor SPNH:

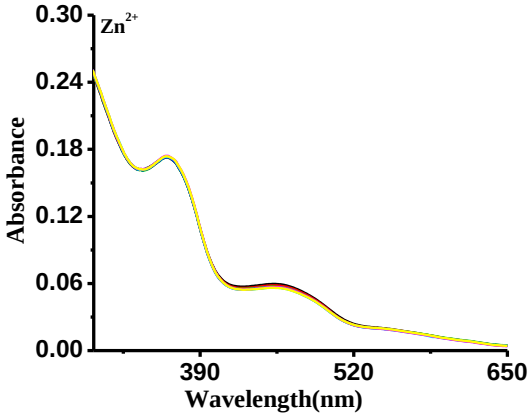


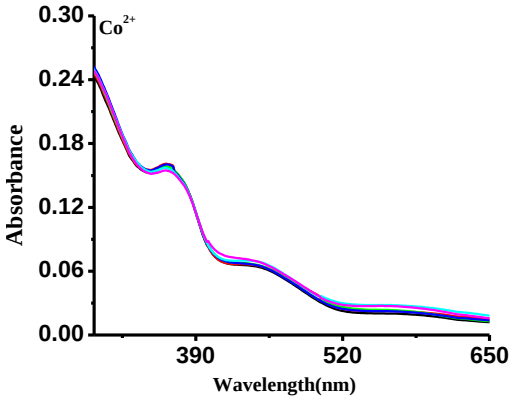
11. Mass spectrum(S9) of SPNH-Cr^{3+} complex:

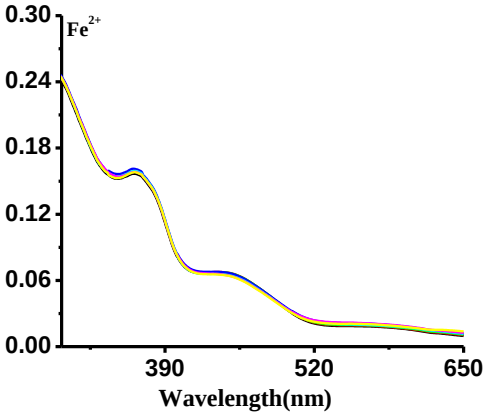


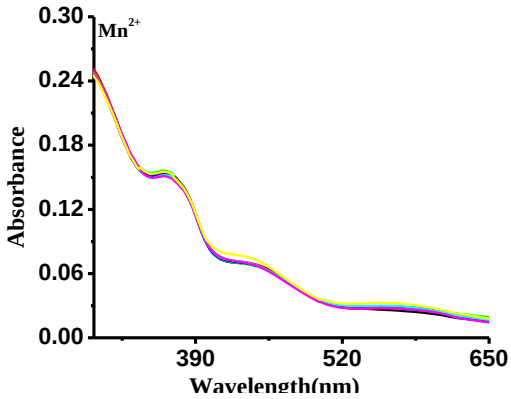
12. UV-vis absorption spectra of SPNH (S10)(c = 1x10^{-5}M) with different cations (c = 2x10^{-4}M) in CH_3CN-HEPES buffer solution (7/3, v/v, 25°C) at pH-7.4 :

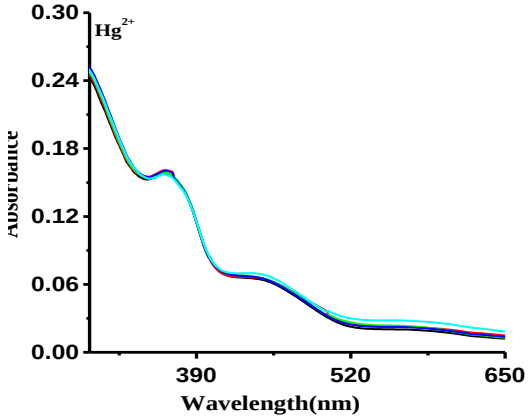


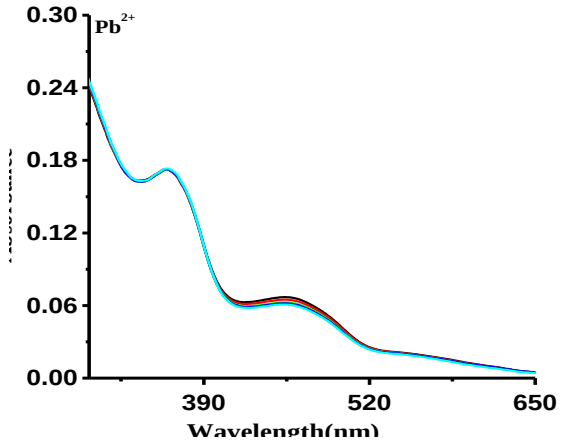


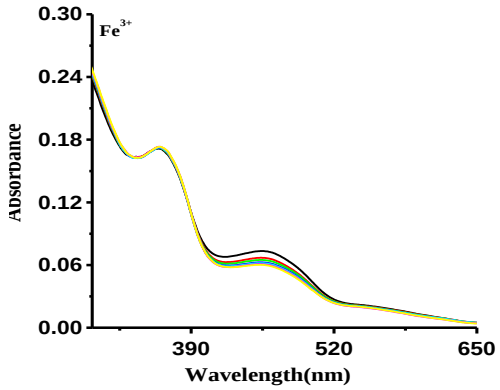


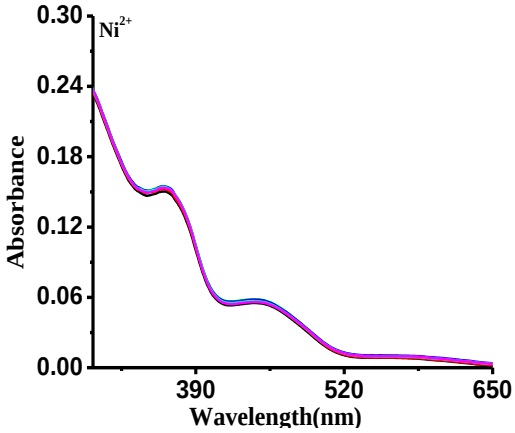


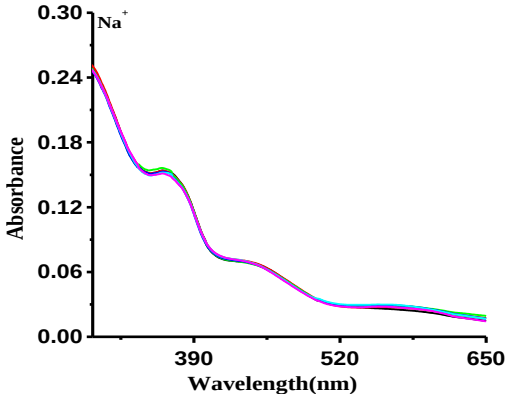


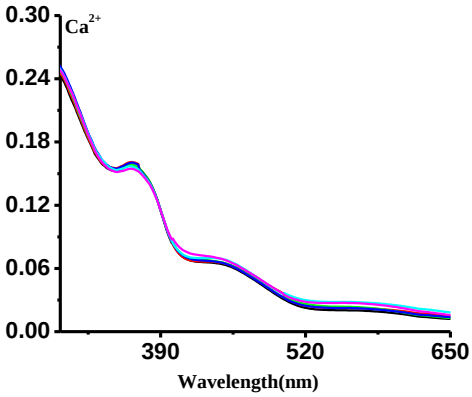


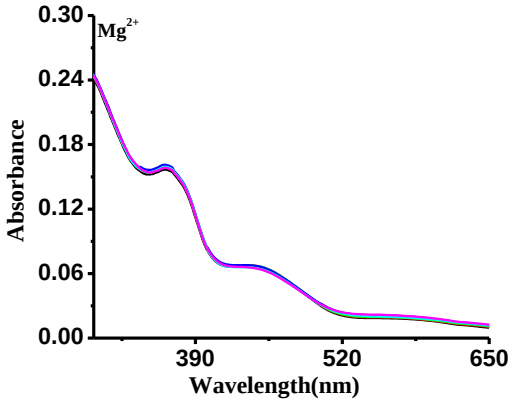


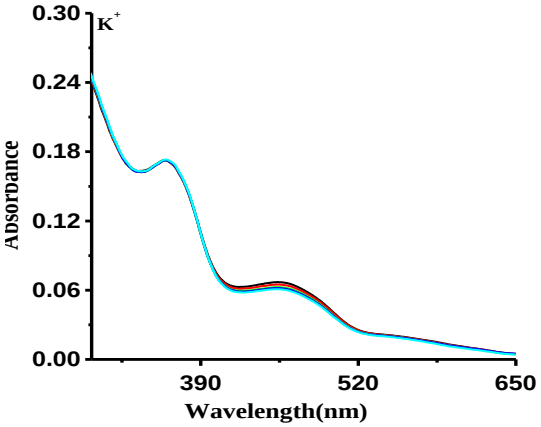




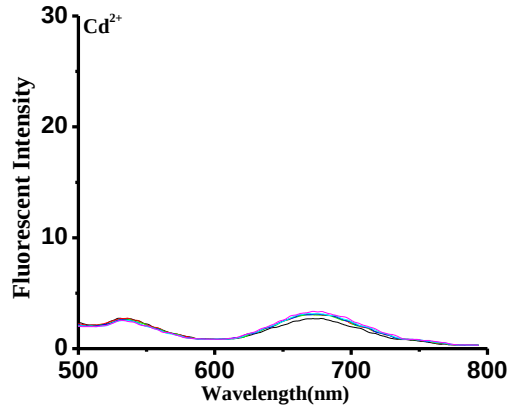


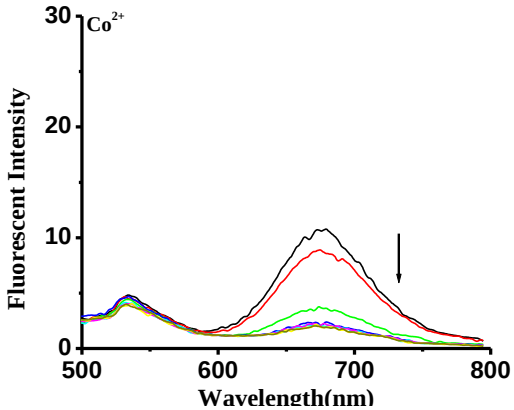


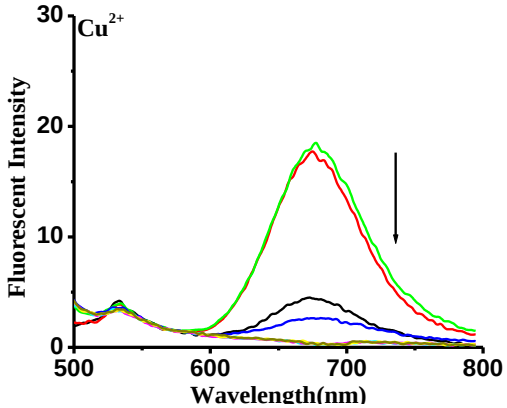


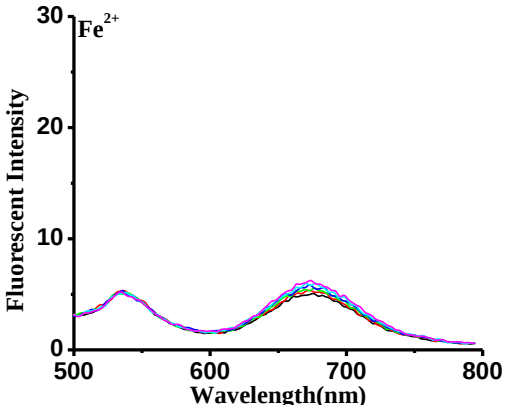


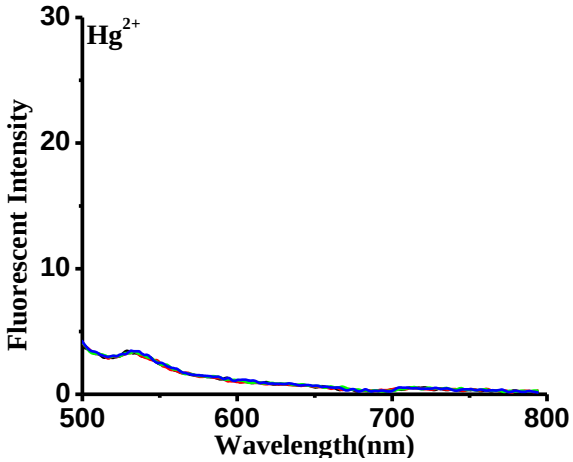
13. Fluorescence titration spectra (S11) of receptor (c = 1x10^{-5} M) with different guest cations (c = 2x10^{-4}M) in CH_3CN-HEPES buffer solution (7/3, v/v, 25°C) at pH-7.4 :

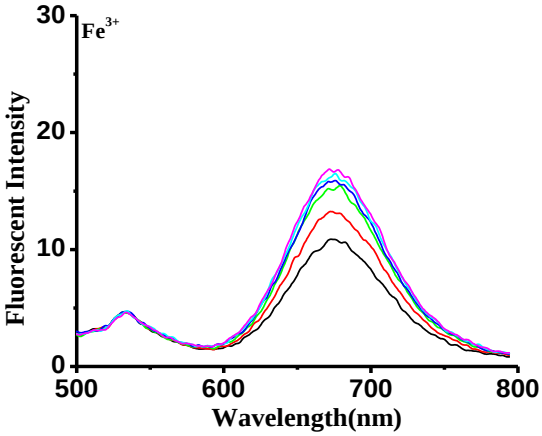


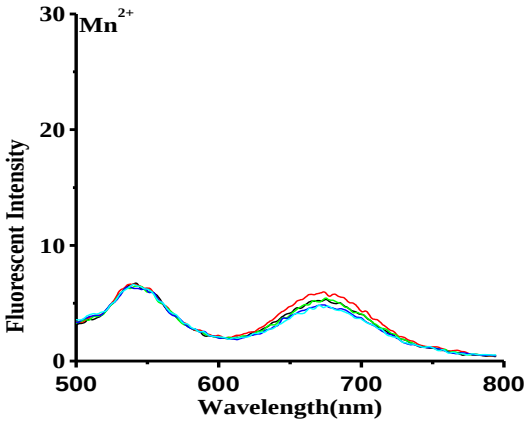


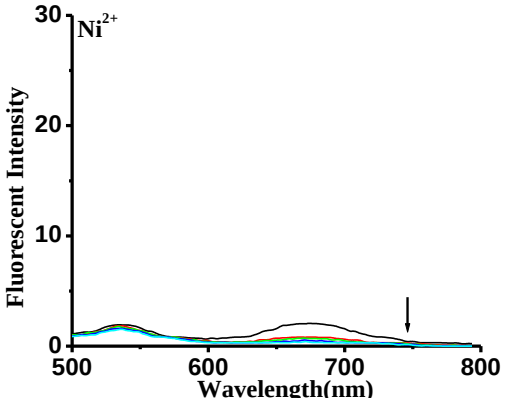


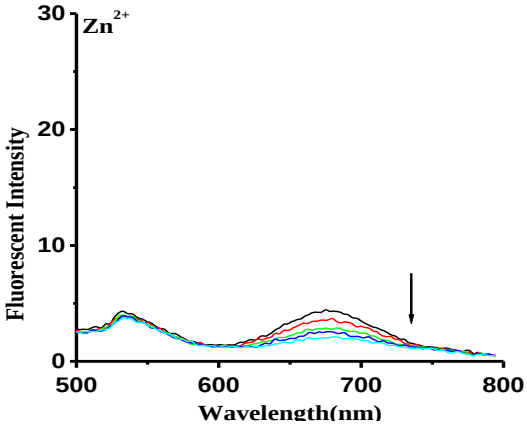


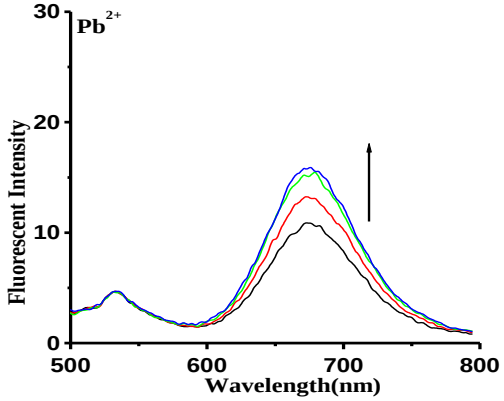


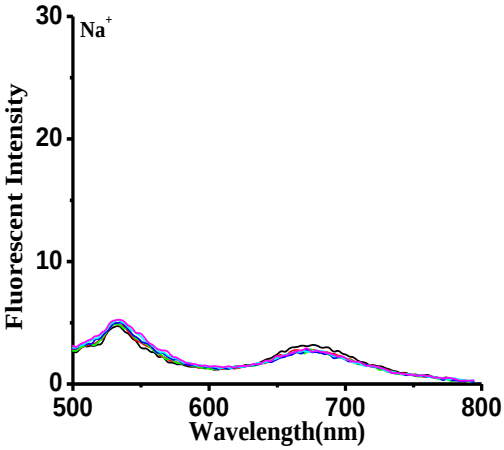


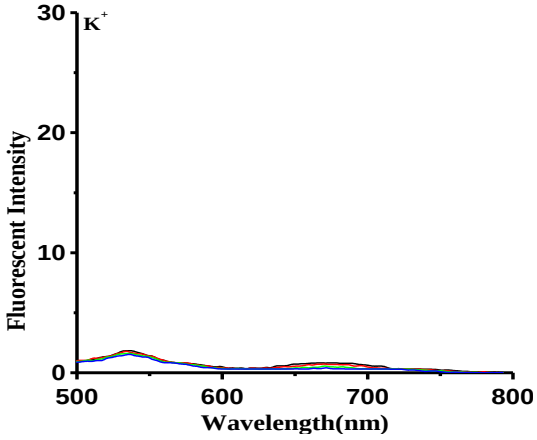


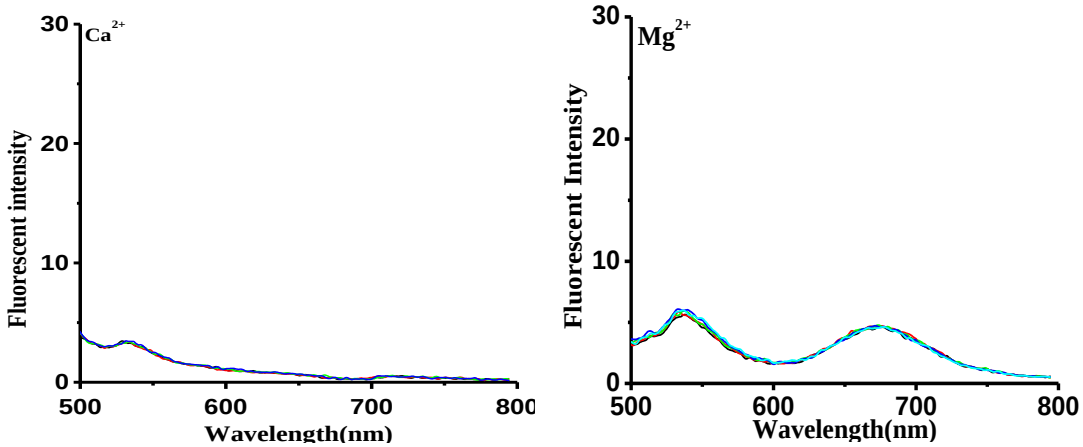












14. Theoretical calculation.

Computational Study:

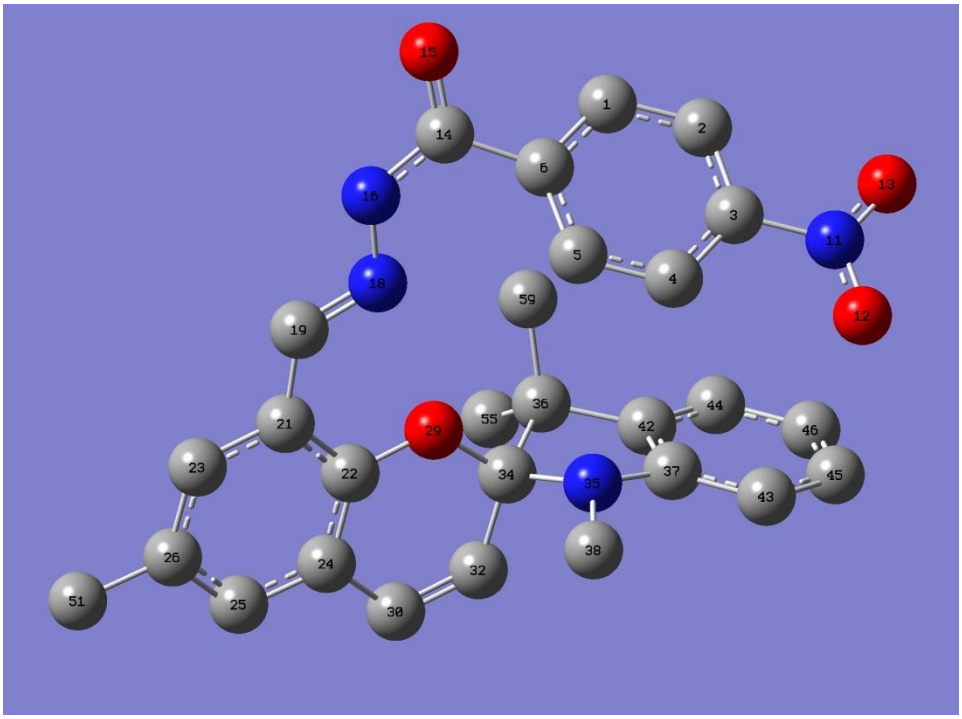
The ground state geometry of **SPNH** and its Cr-complex were optimized using Density Functional Theory (DFT). The B3LYP functional and 6-31G basis set used for all atoms. The calculation of absorption and emission were performed using Time Dependent Density Functional Theory (TD-DFT). The energy difference between the optimized geometry at the excited state which gives the λ_{max} and the ground state was used in computing emission. The first excited state of the **SPNH** and the Cr-complex did not the match the observed λ_{max}, so other excited state was used to calculate the emission. All the computation in acetonitrile solvent was carried out using Polarizable Continuum Model (PCM). All these computational work were carried out using Gaussian 09 program.

The experimentally observed absorption peak (λ_{max}) at 373 nm for **SPNH** is in good agreement with the theoretically calculated value of 400 nm in gas phase and 351 nm in acetonitrile medium (table -2). Whereas for **SPNH-Cr** complex the calculated absorption at 444 nm and 448 nm in gas phase and acetonitrile medium respectively are more excitingly supports our experimentally observed absorption of 440 nm. In emission spectrum, **SPNH** shows the emission (λ_{max}) at 675 nm which is in accord with the theoretically calculated 694 nm in gas phase and 657 in acetonitrile medium. For **SPNH-Cr** complex also we studied emission nature but we got a characteristic peak at 640 nm with low intensity. Using 6-31+G (d,p) we studied, too

which results the absorption peak at 349 nm in acetonitrile which is very close to our calculated data using 6-31G.

Cartesian Coordinates 1. SPNH (Solvent Phase).

B3LYP optimized structure (Hydrogen atom is removed for clarity)



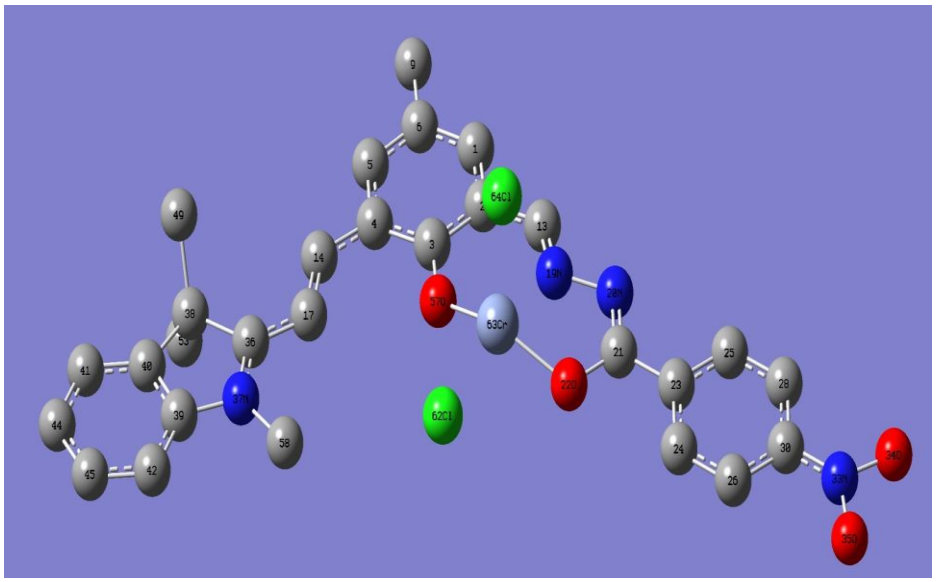
Energy(B3LYP)= -1602.5317702 a.u

Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-2.705363	3.017094	1.012498
2	6	0	-3.763924	2.214381	0.599554
3	6	0	-3.650900	1.540227	-0.617980
4	6	0	-2.513721	1.638256	-1.420822
5	6	0	-1.450230	2.422169	-0.980703
6	6	0	-1.540939	3.116169	0.235035
7	1	0	-2.776742	3.570886	1.941667
8	1	0	-4.660921	2.116948	1.197262
9	1	0	-2.464939	1.109915	-2.364059
10	1	0	-0.558694	2.499932	-1.590207
11	7	0	-4.769310	0.704950	-1.071889
12	8	0	-4.674975	0.147434	-2.169296
13	8	0	-5.754602	0.597971	-0.335310
14	6	0	-0.483250	4.077938	0.692814

15	8	0	-0.795611	5.184816	1.150302
16	7	0	0.834828	3.745002	0.571477
17	1	0	1.492416	4.478114	0.841294
18	7	0	1.297892	2.485178	0.281049
19	6	0	2.581722	2.386843	0.219325
20	1	0	3.201180	3.279975	0.367718
21	6	0	3.348443	1.173828	-0.062001
22	6	0	2.853596	-0.152514	-0.075291
23	6	0	4.721475	1.368651	-0.324284
24	6	0	3.720365	-1.221647	-0.367073
25	6	0	5.072229	-0.970569	-0.642212
26	6	0	5.599640	0.323588	-0.622992
27	1	0	5.103562	2.386496	-0.297466
28	1	0	5.717875	-1.815823	-0.868993
29	8	0	1.541421	-0.355563	0.223810
30	6	0	3.159997	-2.564231	-0.370277
31	1	0	3.815712	-3.397679	-0.608662
32	6	0	1.866613	-2.767377	-0.087361
33	1	0	1.437426	-3.764512	-0.099721
34	6	0	0.900727	-1.648434	0.200117
35	7	0	-0.157399	-1.609915	-0.832551
36	6	0	0.071996	-1.867469	1.539929
37	6	0	-1.217521	-2.404297	-0.395058
38	6	0	0.203151	-1.545965	-2.240954
39	1	0	0.495173	-2.521667	-2.656623
40	1	0	-0.645006	-1.163078	-2.814846
41	1	0	1.031363	-0.846159	-2.366562
42	6	0	-1.131696	-2.622603	0.993935
43	6	0	-2.263199	-2.948399	-1.146713
44	6	0	-2.102622	-3.368370	1.645869
45	6	0	-3.238982	-3.702239	-0.474103
46	6	0	-3.166862	-3.916992	0.904790
47	1	0	-2.328125	-2.797643	-2.219274
48	1	0	-4.058858	-4.131118	-1.043809
49	1	0	-3.926193	-4.510943	1.404363
50	1	0	-2.043007	-3.536874	2.718096
51	6	0	7.060531	0.589803	-0.907550

Cartesian Coordinates 2. SPNH –Cr complex (Solvent Phase).

B3LYP optimized structure(Hydrogen atom is removed for clarity)



Energy(B3LYP)= -3566.370858 a.u

Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.075190	4.472122	-0.226522
2	6	0	0.292225	3.114182	-0.149763
3	6	0	-0.715108	2.107266	-0.150340
4	6	0	-2.086577	2.495706	-0.211406
5	6	0	-2.389842	3.879798	-0.269790
6	6	0	-1.413756	4.878801	-0.280246
7	1	0	0.712835	5.219566	-0.242627
8	1	0	-3.435663	4.169173	-0.313309
9	6	0	-1.781813	6.343684	-0.342636
10	1	0	-2.854712	6.475698	-0.508404
11	1	0	-1.521738	6.858670	0.590387
12	1	0	-1.248885	6.853699	-1.153207
13	6	0	1.695866	2.787941	-0.122267
14	6	0	-3.217091	1.593711	-0.219720
15	1	0	2.417072	3.588113	-0.269342
16	1	0	-4.162254	2.121850	-0.258848
17	6	0	-3.250297	0.222696	-0.189644
18	1	0	-2.312244	-0.313254	-0.156611
19	7	0	2.151314	1.572880	0.037342
20	7	0	3.552644	1.415254	-0.075592
21	6	0	3.829358	0.130526	-0.118874

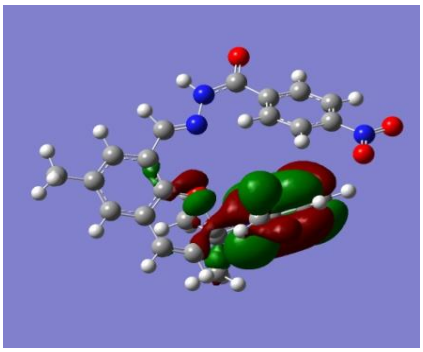
22	8	0	2.835723	-0.777744	-0.088174
23	6	0	5.216594	-0.344382	-0.224728
24	6	0	5.476306	-1.726499	-0.280592
25	6	0	6.286168	0.573066	-0.266013
26	6	0	6.785356	-2.192154	-0.375545
27	1	0	4.650064	-2.424359	-0.248611
28	6	0	7.596816	0.118186	-0.361300
29	1	0	6.079208	1.634347	-0.222528
30	6	0	7.828832	-1.261922	-0.414654
31	1	0	7.002287	-3.250523	-0.418580
32	1	0	8.428876	0.807908	-0.393582
33	7	0	9.200430	-1.743978	-0.513782
34	8	0	10.141004	-0.894768	-0.547036
35	8	0	9.402377	-2.994438	-0.563144
36	6	0	-4.463671	-0.527546	-0.201629
37	7	0	-4.486309	-1.872631	-0.174719
38	6	0	-5.918716	-0.002597	-0.244574
39	6	0	-5.818626	-2.383927	-0.199803
40	6	0	-6.707536	-1.306028	-0.238523
41	6	0	-8.078230	-1.544238	-0.264980
42	6	0	-6.233870	-3.713583	-0.188702
43	1	0	-5.533430	-4.538835	-0.161400
44	6	0	-8.526991	-2.875778	-0.253108
45	6	0	-7.616794	-3.943737	-0.216012
46	1	0	-8.790997	-0.727203	-0.293966
47	1	0	-9.591530	-3.081046	-0.273119
48	1	0	-7.984693	-4.963485	-0.208308
49	6	0	-6.269466	0.835580	1.019198
50	1	0	-6.040562	0.279735	1.933134
51	1	0	-5.727908	1.783531	1.044949
52	1	0	-7.340922	1.057127	1.010575
53	6	0	-6.212705	0.786340	-1.553736
54	1	0	-5.669192	1.732478	-1.592758
55	1	0	-5.945704	0.195686	-2.434844
56	1	0	-7.283048	1.008269	-1.599836
57	8	0	-0.384825	0.796203	-0.116587
58	6	0	-3.344236	-2.797752	-0.133597
59	1	0	-3.305543	-3.363550	-1.068294
60	1	0	-2.405752	-2.269966	0.004452
61	1	0	-3.481567	-3.488485	0.701270
62	17	0	0.204339	-2.333628	0.384259
63	24	0	1.121767	-0.138330	0.406372
64	17	0	1.220412	0.274997	2.701878

Table-1: Structural parameters of the SPNH –Cr complex (solvent ACN)

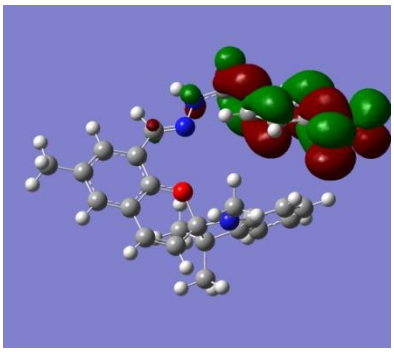
Bond	Distance (A^0)	Angle	Angle (^{0})	Dihedral Angle	Dihedral Angle
$Cr_{63} – O_{22}$	1.89501	$Cl_{64} –Cr_{63} –O_{22}$	106.149	O_{22}-Cl_{62}-O_{57}-N_{19}	-6.368
Cr63 – O57	1.84842	Cl64 –Cr63 –O57	102.892		
Cr63 – N19	2.03086	Cl64 –Cr63 –Cl62	100.882		
Cr63 – Cl62	2.37939	Cl64 –Cr63 –N19	90.462		
Cr63 – Cl64	2.33451	N19 – Cr63 –O22	77.195		
		O22 – Cr63 –Cl62	92.005		
		Cl62 –Cr63 –O57	98.602		
		O57-Cr63 –N19	86.318		

Table-2: Observed UV-Visible absorption and computed absorption of SPNH and Cr-SPNH complex in acetonitrile and gas phase.

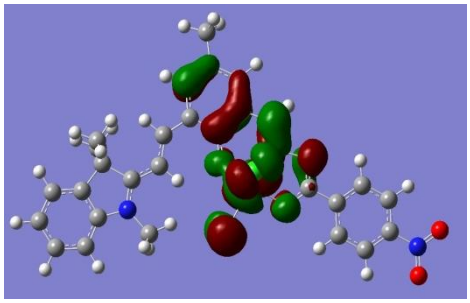
	Medium	Experimental Absorption λ_{abs} (nm) in acetonitrile/ water	TD-DFT vertical Excitation (nm)	f	Experimental Emission (nm)	Emission (nm)
SPNH	Gas Phase	373	400	0.0241	675	694
	ACN		351	0.1003		657
SPNH –Cr complex	Gas Phase	440	444	0.2129	675	640
	ACN		448	0.4548		****



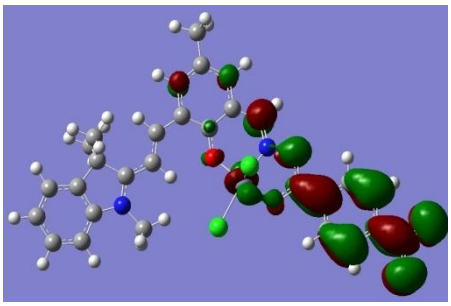
HOMO_**SPNH**



LUMO_**SPNH**



HOMO of **SPNH**-Cr_complex



LUMO of **SPNH**-Cr_Complex