----------Supporting Information----------

Highly selective fluorometric sensing of Cu^{2+} and Hg^{2+} by using a benzothiazole based receptor in semi-aqueous medium and molecular docking studies

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General information

Melting points were measured in open capillary tubes and uncorrected. FT-IR spectra were obtained on Shimadzu IR-Affinity spectrometer (KBr Pellets). The ^{1}H NMR, ^{13}C NMR were recorded on FT NMR spectrometer model Avance - II (Bruker). The instrument is equipped with a cryomagnet of field strength 9.4 T. Its ^{1}H frequency is 400 and ^{13}C frequency is 100 MHz. NMR instrument uses a TMS as an internal standard and $CDCl_3$ as a solvent. Chemical shifts are given in parts per million (δ-scale) and the coupling constants are given in Hertz. Mass spectra were performed on an Ultima Global spectrometer with an ESI source. Silica gel (60–120 mesh size) was used for the column chromatography and silica gel-G plates (Merck) were used for TLC analysis with a mixture of acetone in n-hexane (10%) as the eluent. 2-Amino benzothiazole and DEAD were obtained from Sigma-Aldrich chemical company and were used without further purification.

Synthesis of *(Z)-ethyl 2-((Z)-2-(benzo[d]thiazol-2-ylimino)-4-oxo-3-phenylthiazolidin-5-ylidene) acetate*) (1)

To a magnetically stirred solution of 1-(benzo[d]thiazol-2-yl)-3-phenylthiourea (2 mmol) and ethanol (5 mL) in 10 mL round bottom flask, diethyl acetylenedicarboxylates (2 mmol) was added drop wise at room temperature. The progress of reaction was monitored by TLC (hexane: acetone, 9:1 ratio), during which solid appears into the solution. After completion of reaction, the excess solvent was removed under reduced pressure and the residue was recrystallized from ethanol and few drops of water affording desired product as pale yellow crystals. Yield- 91.0%, m.p**.** 216-218 ^{o}C.

Spectral Data

IR (KBr, cm^{-1}): 3063, 2981, 1723, 1693, 1598, 1569, 1493, 1429, 1376, 1318, 1198, 1141, 1018, 879, 759, 736, 690, 566;

^{1}H NMR (400 MHz, $CDCl_3$): δ(ppm); 1.39 (t, 3H, *J = 7.12 Hz*), 4.36 (q, 2H, *J = 7.12 Hz*), 7.07 (s, 1H, C=C<u>H</u>), 7.29-7.33 (m, 1H, ArH), 7.37-7.39 (m, 2H, ArH) 7.43-7.57 (m, 4H, ArH), 7.76 (d, 1H, ArH, *J = 8.04 Hz*), 7.98 (d, 1H, ArH, *J = 8.12 Hz*);

^{13}C NMR (100 MHz, $CDCl_3$): δ(ppm); 14.20, 62.0, 119.61, 121.41, 122.81, 124.67, 126.33, 127.97, 129.38, 133.83, 134.71, 141.67, 151.06, 158.28, 164.7, 165.6, 167.5;

LC-MS (*m/z*) = 432.1 $[M + Na]^+$ 100%.

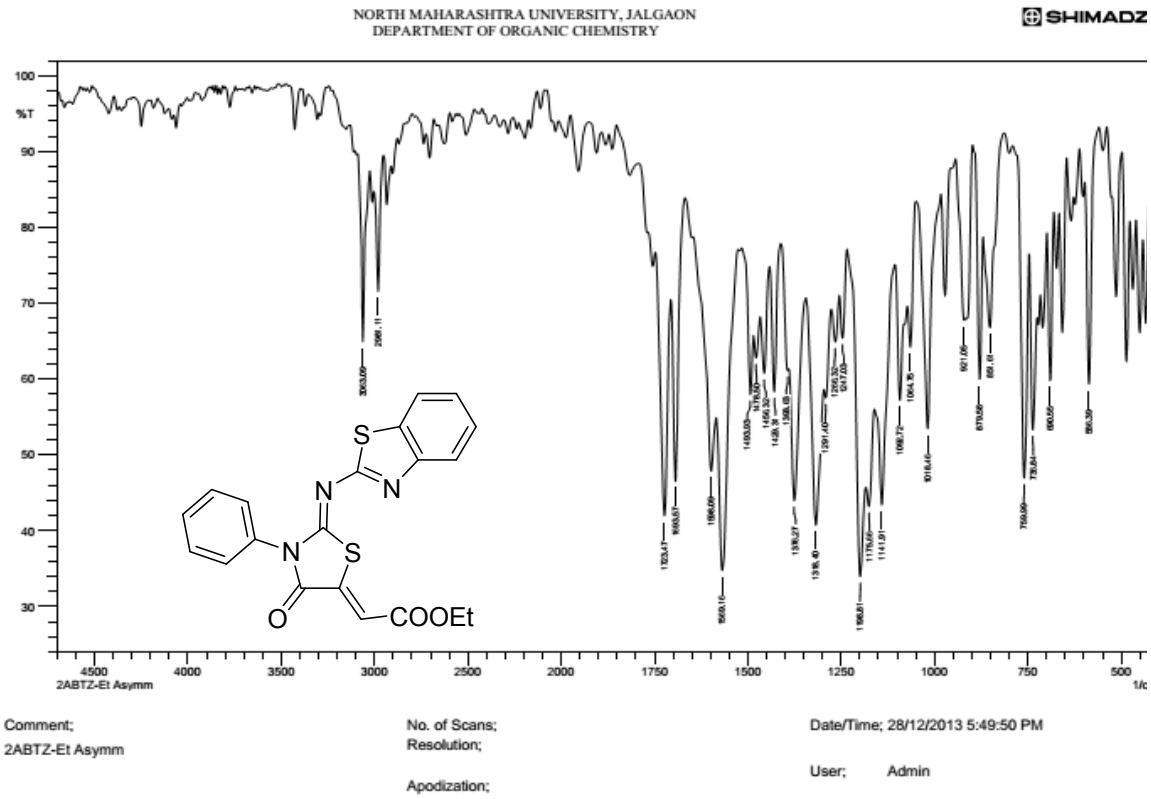


Figure S1. IR spectra of receptor **1**

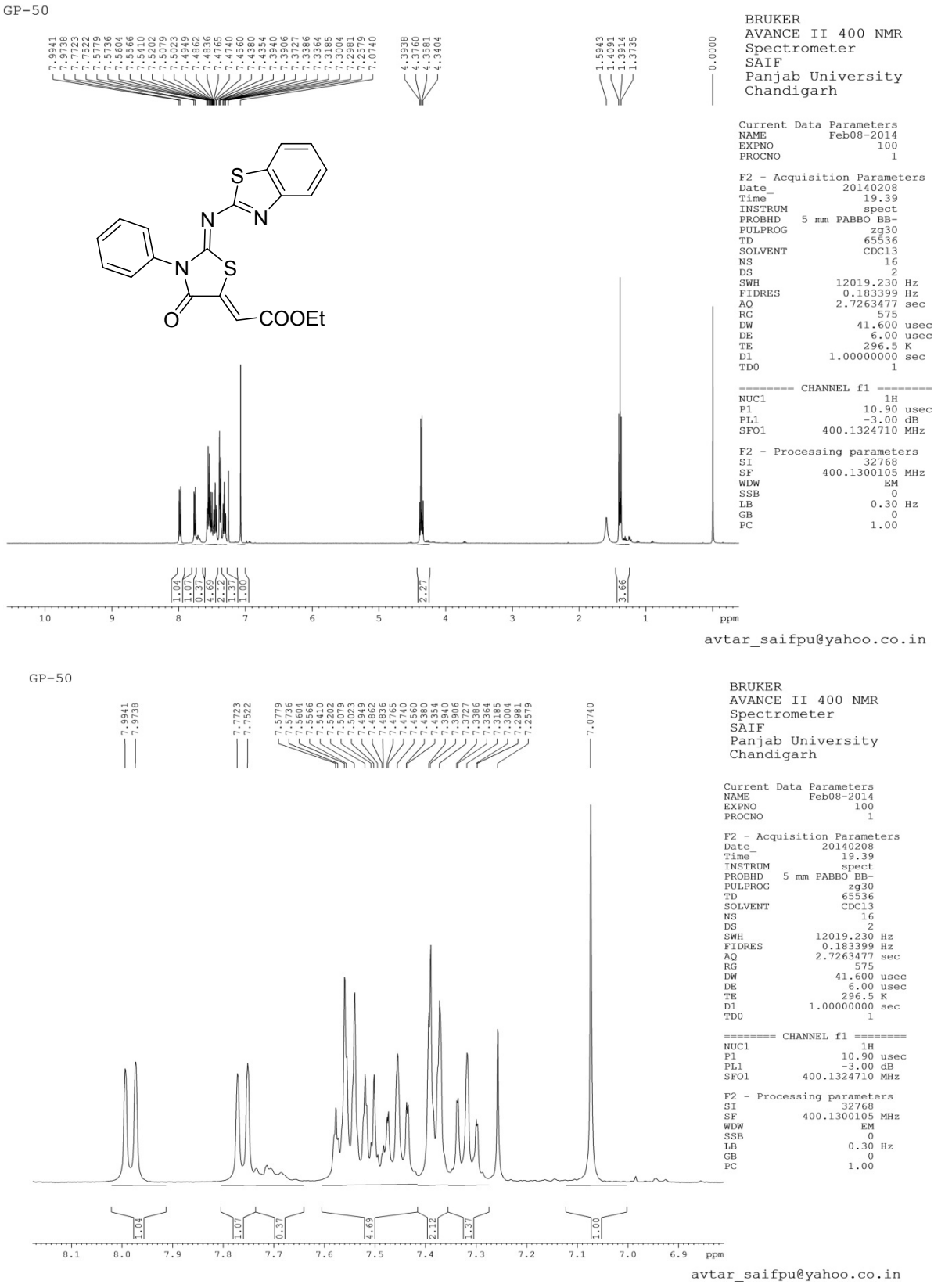


Figure S2. ^{1}H-NMR spectra of receptor **1**

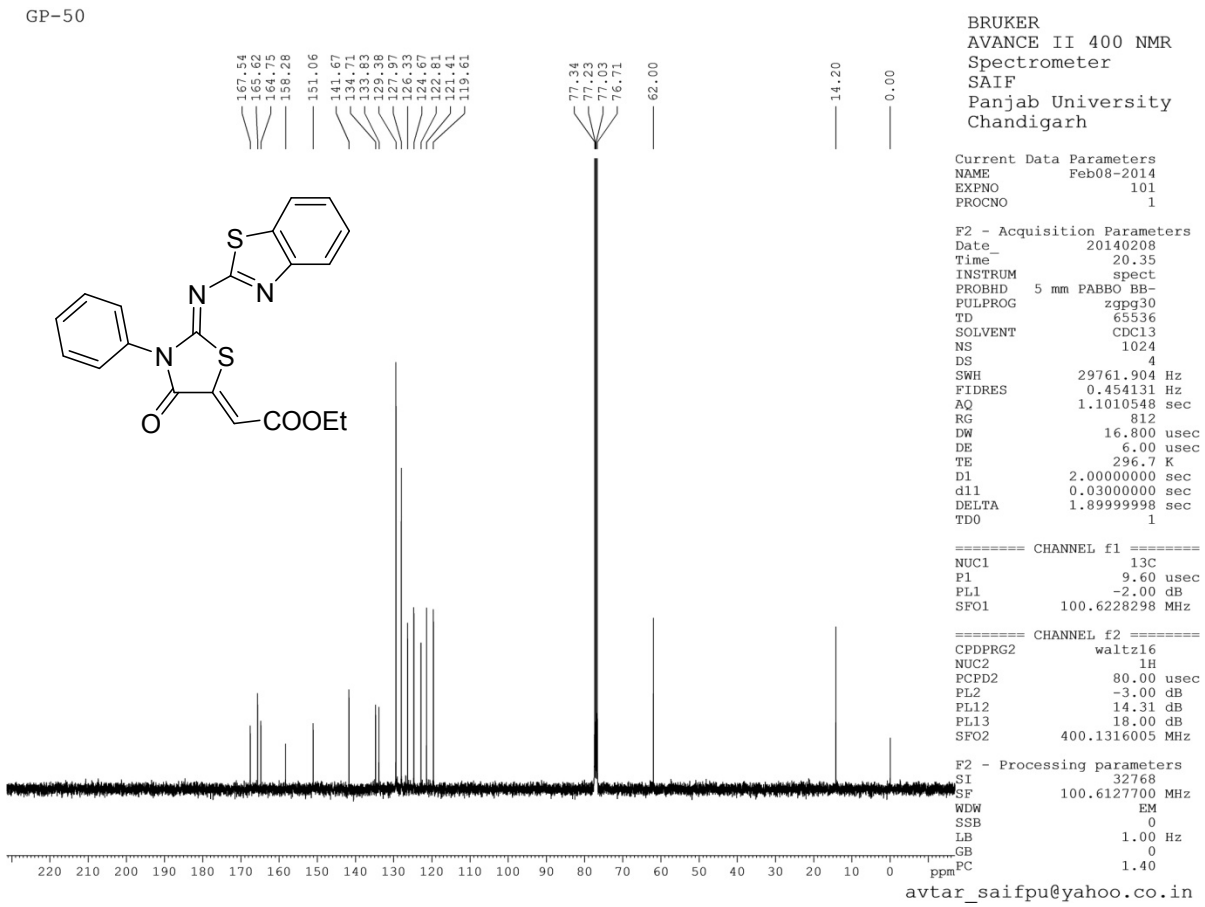


Figure S3. ^{13}C-NMR spectra of receptor **1**

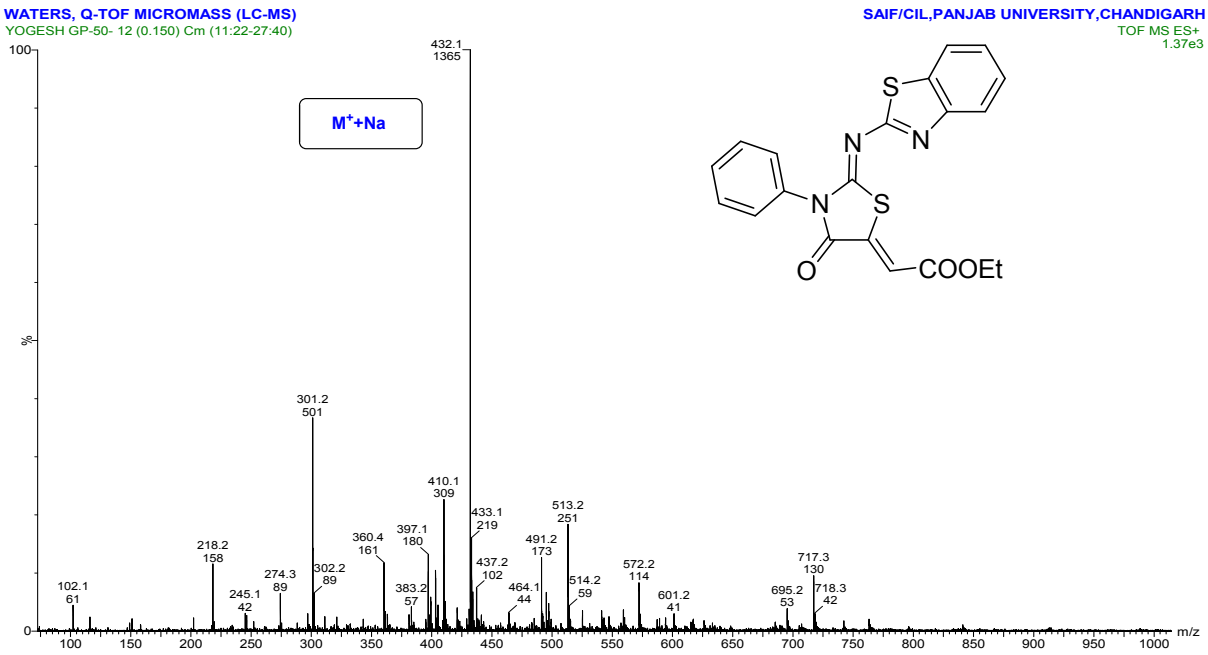


Figure S4. LC-MS spectra of receptor **1 (M+Na$^+$)**

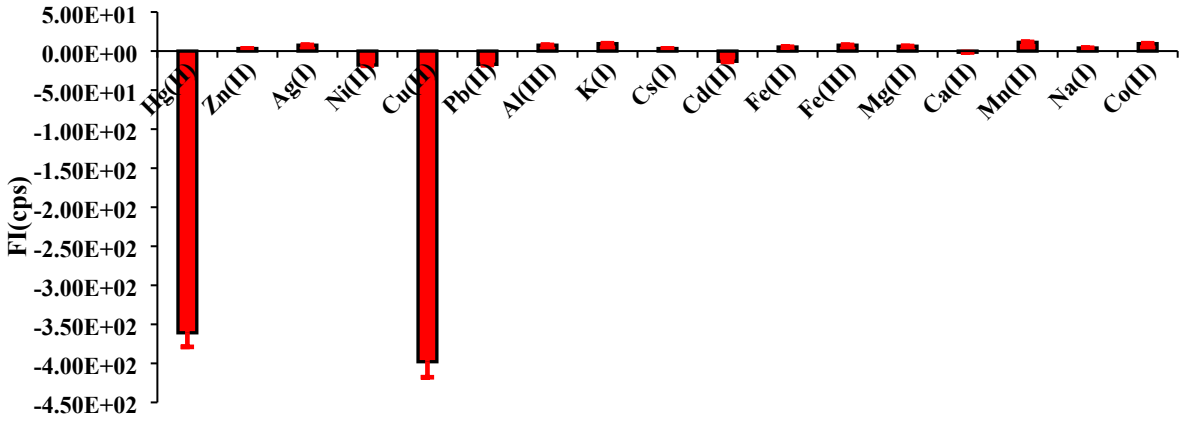


Figure S5 Fluorescence ratio metric response of receptor **1** ($c = 1 \times 10^{-5}$ M) upon the addition of a particular cations ($c = 1 \times 10^{-4}$ M) in CH_3OH /H_2O (50:50, *v/v*).

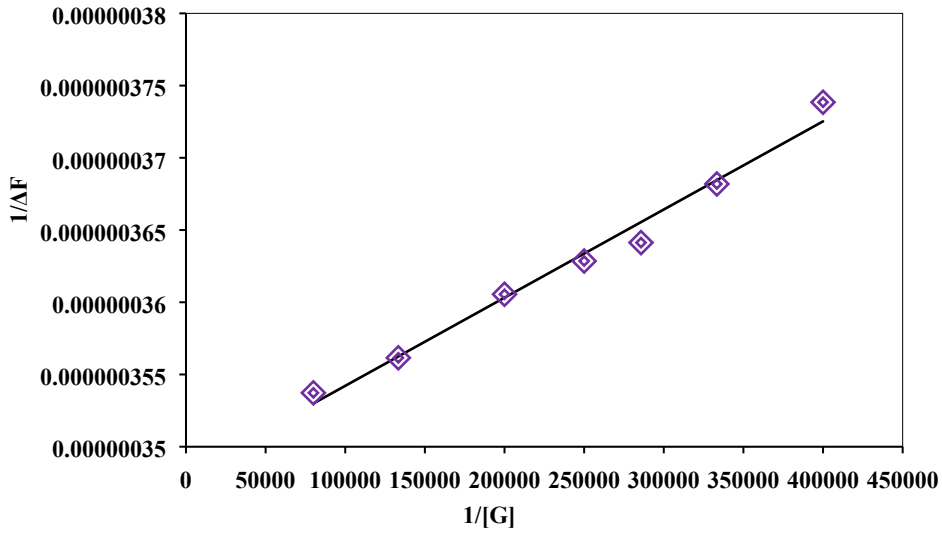


Figure S6 Benesi-Hildebrand Plot receptor **1** (adjusted equation: $1/F\text{-}F_0$ = -2E-13x-3E-07 1/[G], R=0.944) at the K value $5.70 \times 10^6\ M^{-1}$ for Cu^{2+}.

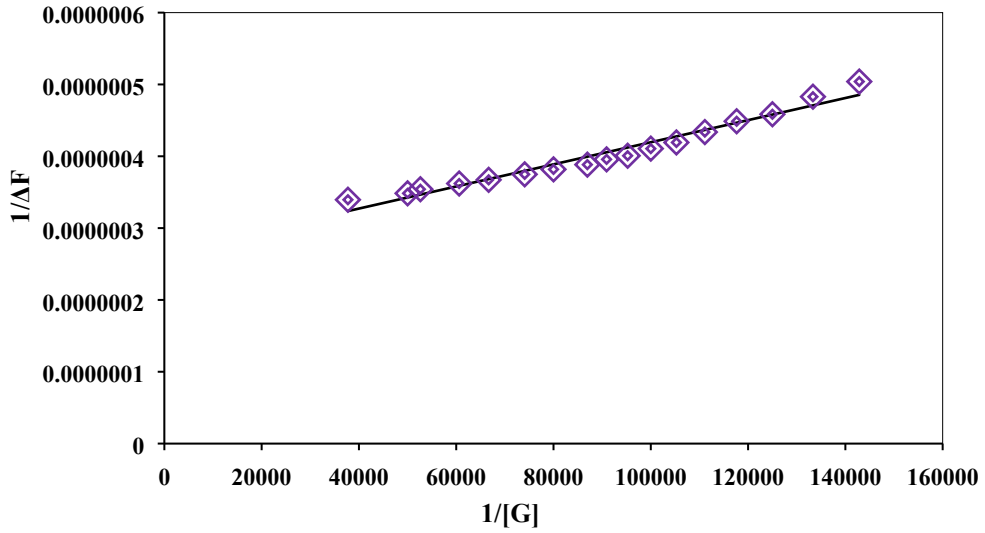


Figure S7 Benesi-Hildebrand Plot receptor **1** (adjusted equation: $1/F\text{-}F_0$ = -8E-12x+7E-07 1/[G], R=0.977) at the K value 1.73 $\times 10^5 M^{-1}$ for Hg^{2+}.

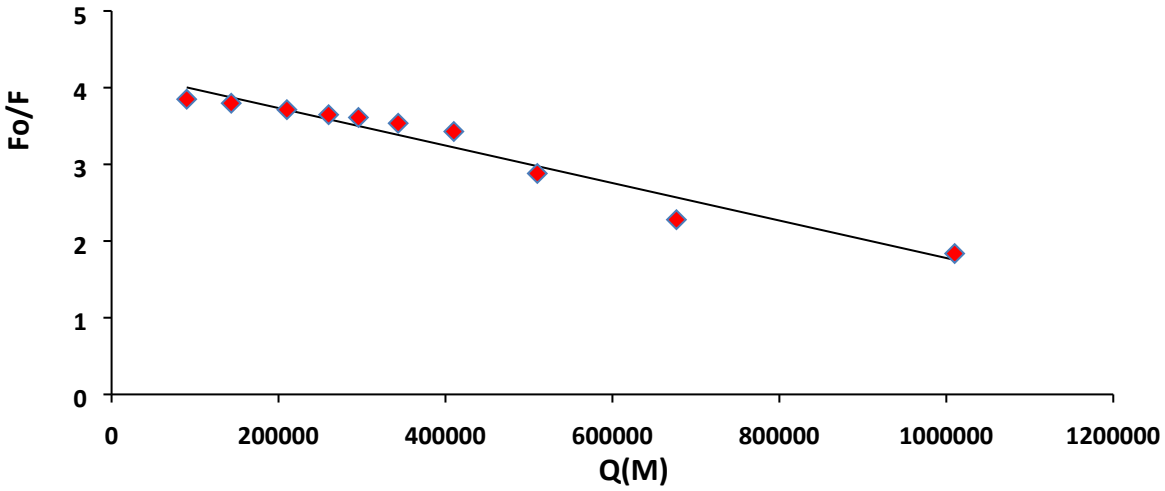


Figure S8 Stern–Volmer plots for titrations of receptor **1** with different concentrations of Cu^{2+} metal salt.

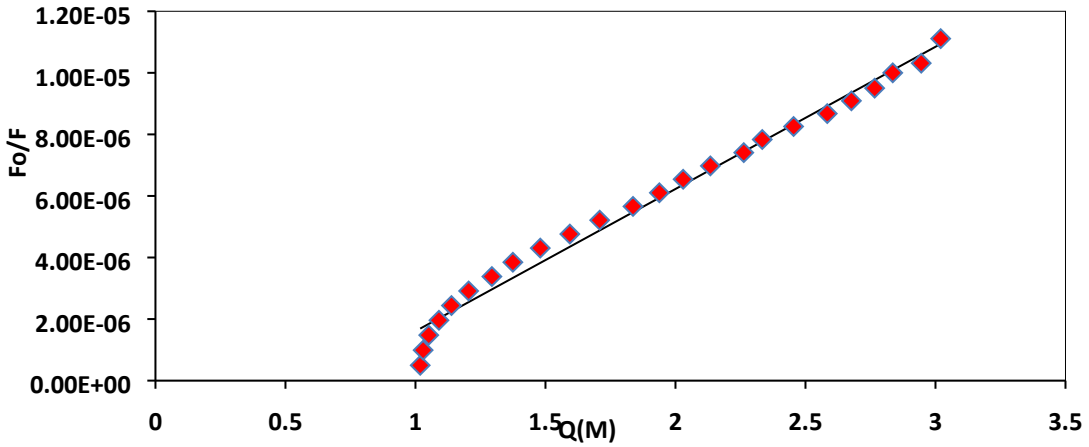


Figure S9 Stern–Volmer plots for titrations of receptor **1** with different concentrations of Hg^{2+} metal salt.

JOB PLOT

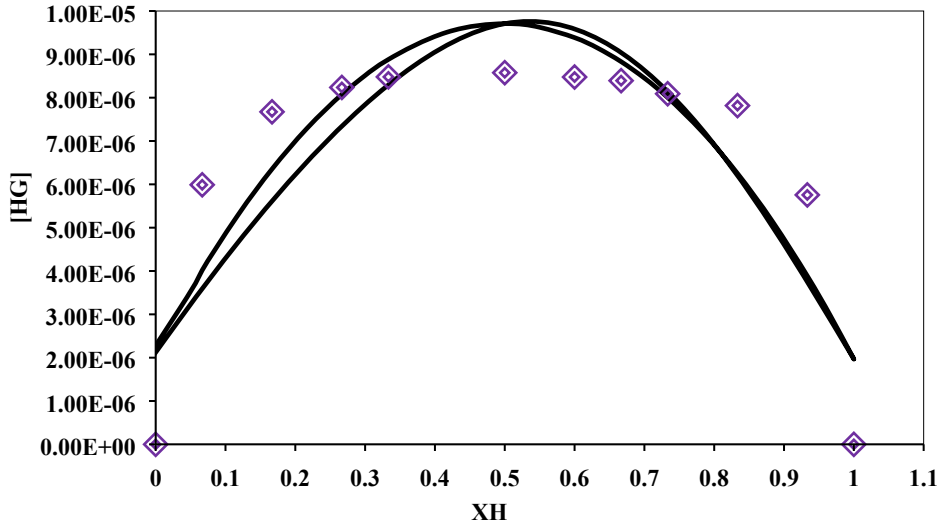


Figure S10 Job plots for titrations of receptor **1** with different concentrations of Cu^{2+} metal salt

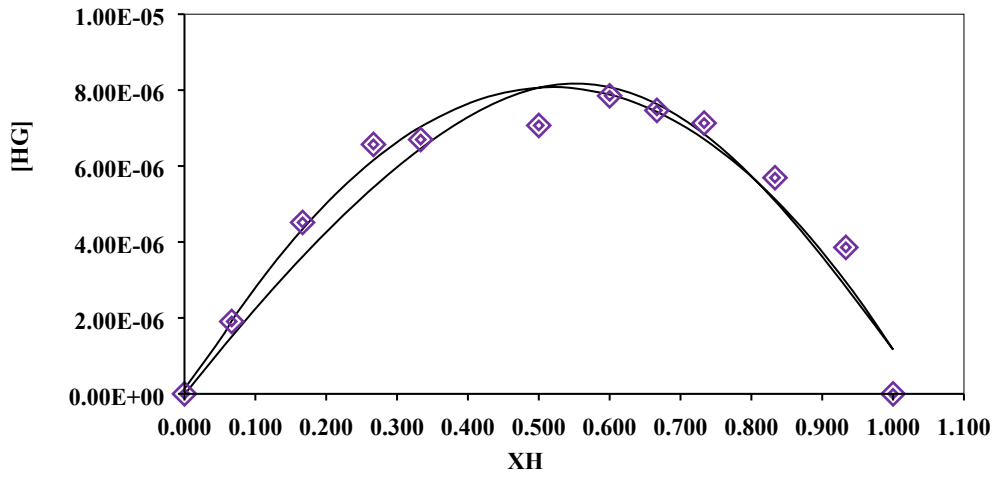


Figure S11 Job plots for titrations of receptor **1** with different concentrations of Hg^{2+} metal salt

Figure S12 Docking Procedure

Protein structure preparation

The X-ray crystallographic structure of Aldose Reductase Inhibitors (ARIs) protein (PDB code 1US0) was obtained from Brookhaven Protein Data Bank (RCSB) (http://www.rcsb.org/pdb). For docking studies, all water molecules and IDD594 were removed from the complex.

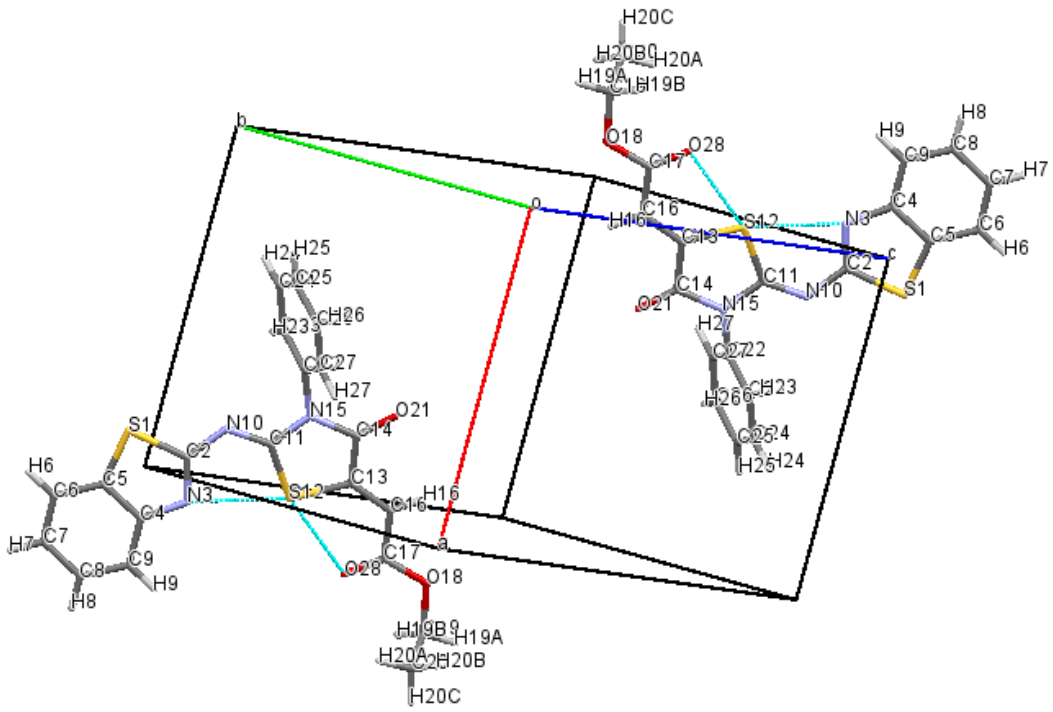
Ligand Preparation

Receptor **1** structure was drawn using Chem Office 8.0. Chem3D Ultra 8.0 was used to convert 2D structure into 3D and energy minimization is carried out using MM2 force field. All ligand structures were saved as pdb file format for input to docking.

3.3 Docking Parameters

Docking Control		
1	Correlation type	Shape + Electrostatics
2	FFT mode	3DFast Lite
3	Post processing	MM Minimization
4	Grid Dimension	0.6
5	Solution	500
6	Ligand range	180
7	Step size	7.5
8	Twist Range	360
9	Step size	5.5
10	Distance Range	40
11	Scan step	0.75
12	Sub Step	2.0
13	Steric Scan	25
14	Final Search	25
15	Solution	0

Figure S13 Crystal Data



The molecular packing diagram of the **1**, viewed along the b*-axis. Dashed lines indicate the hydrogen-bonding interactions

Table 1. Crystallographic details for **1**

Formula	C20 H15 N3 O3 S2	
Formula weight	409.47	
Temperature	150(2) K	
Wavelength	0.71073 Å	
Crystal system	Triclinic	
Space group	$P\bar{1}$	
Unit cell dimensions	a = 10.2244(2) Å	α= 117.291(1)°
	b = 10.6450(2) Å	β= 104.012(1)°
	c = 10.7022(2) Å	γ = 102.864(1)°
Volume	926.11(3) $Å^3$	
Z	2	
Density (calculated)	1.468 Mg/m^3	

Absorption coefficient	0.315 mm^{-1}
F(000)	424
Crystal size	0.15 x 0.19 x 0.27 mm^3
Theta range for data collection	2.226 to 27.459°
Index ranges	-13<=h<=13, -13<=k<=13, -13<=l<=13
Reflections collected	28487
Independent reflections	4233 [R(int) = 0.039]
Completeness to theta = 25.242°	100.0 %
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	4233 / 0 / 254
Goodness-of-fit on F^2	1.038
Final R indices [I>2sigma(I)]	R1 = 0.0352, wR2 = 0.0801
R indices (all data)	R1 = 0.0558, wR2 = 0.0892
Largest diff. peak and hole	0.302 and -0.260 e/$Å^3$

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters ($Å^2$x 10^3) for **1**. U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)
C(2)	9250(2)	7223(2)	8702(2)	22(1)
C(4)	11364(2)	8979(2)	9407(2)	23(1)
C(5)	10550(2)	8985(2)	8145(2)	25(1)
C(6)	11181(2)	9960(2)	7739(2)	29(1)
C(7)	12637(2)	10930(2)	8605(2)	30(1)
C(8)	13459(2)	10909(2)	9834(2)	32(1)
C(9)	12839(2)	9944(2)	10242(2)	31(1)
C(11)	8388(2)	5845(2)	9678(2)	21(1)
C(13)	9145(2)	5601(2)	11941(2)	22(1)
C(14)	7652(2)	4471(2)	10773(2)	22(1)
C(16)	9761(2)	5611(2)	13203(2)	24(1)
C(17)	11261(2)	6693(2)	14298(2)	24(1)
C(19)	13286(2)	7350(2)	16454(2)	30(1)
C(20)	14248(2)	6632(2)	15779(2)	37(1)

C(22)	5935(2)	3707(2)	8250(2)	22(1)
C(23)	4950(2)	4329(2)	7930(2)	26(1)
C(24)	3621(2)	3367(2)	6676(2)	33(1)
C(25)	3292(2)	1810(2)	5784(2)	34(1)
C(26)	4287(2)	1204(2)	6125(2)	33(1)
C(27)	5629(2)	2157(2)	7357(2)	27(1)
N(3)	10592(1)	7975(2)	9700(2)	25(1)
N(10)	8161(1)	6095(2)	8601(2)	23(1)
N(15)	7308(1)	4684(2)	9566(1)	21(1)
O(18)	11749(1)	6431(1)	15395(1)	28(1)
O(21)	6843(1)	3515(1)	10852(1)	27(1)
O(28)	11967(1)	7690(1)	14185(1)	32(1)
S(1)	8775(1)	7648(1)	7301(1)	30(1)
S(12)	9953(1)	6802(1)	11411(1)	23(1)

Table 3. Bond lengths [Å] and angles [°] for **1**.

C(2)-N(3)	1.297(2)
C(2)-N(10)	1.385(2)
C(2)-S(1)	1.7483(16)
C(4)-N(3)	1.383(2)
C(4)-C(9)	1.394(2)
C(4)-C(5)	1.409(2)
C(5)-C(6)	1.391(2)
C(5)-S(1)	1.7358(17)
C(6)-C(7)	1.383(2)
C(6)-H(6)	0.9500
C(7)-C(8)	1.391(3)
C(7)-H(7)	0.9500
C(8)-C(9)	1.379(2)
C(8)-H(8)	0.9500
C(9)-H(9)	0.9500
C(11)-N(10)	1.278(2)
C(11)-N(15)	1.398(2)
C(11)-S(12)	1.7721(16)

C(13)-C(16)	1.339(2)
C(13)-C(14)	1.490(2)
C(13)-S(12)	1.7499(16)
C(14)-O(21)	1.2105(19)
C(14)-N(15)	1.388(2)
C(16)-C(17)	1.469(2)
C(16)-H(16)	0.9500
C(17)-O(28)	1.2166(19)
C(17)-O(18)	1.3351(19)
C(19)-O(18)	1.4652(19)
C(19)-C(20)	1.499(3)
C(19)-H(19A)	0.9900
C(19)-H(19B)	0.9900
C(20)-H(20A)	0.9800
C(20)-H(20B)	0.9800
C(20)-H(20C)	0.9800
C(22)-C(23)	1.382(2)
C(22)-C(27)	1.384(2)
C(22)-N(15)	1.4444(19)
C(23)-C(24)	1.391(2)
C(23)-H(23)	0.9500
C(24)-C(25)	1.385(3)
C(24)-H(24)	0.9500
C(25)-C(26)	1.383(3)
C(25)-H(25)	0.9500
C(26)-C(27)	1.388(2)
C(26)-H(26)	0.9500
C(27)-H(27)	0.9500

N(3)-C(2)-N(10)	127.27(14)
N(3)-C(2)-S(1)	116.28(12)
N(10)-C(2)-S(1)	116.43(11)
N(3)-C(4)-C(9)	125.61(15)
N(3)-C(4)-C(5)	115.13(14)
C(9)-C(4)-C(5)	119.26(15)
C(6)-C(5)-C(4)	121.24(15)

C(6)-C(5)-S(1)	129.51(13)
C(4)-C(5)-S(1)	109.25(12)
C(7)-C(6)-C(5)	118.35(16)
C(7)-C(6)-H(6)	120.8
C(5)-C(6)-H(6)	120.8
C(6)-C(7)-C(8)	120.83(16)
C(6)-C(7)-H(7)	119.6
C(8)-C(7)-H(7)	119.6
C(9)-C(8)-C(7)	121.07(16)
C(9)-C(8)-H(8)	119.5
C(7)-C(8)-H(8)	119.5
C(8)-C(9)-C(4)	119.22(16)
C(8)-C(9)-H(9)	120.4
C(4)-C(9)-H(9)	120.4
N(10)-C(11)-N(15)	119.77(14)
N(10)-C(11)-S(12)	128.70(12)
N(15)-C(11)-S(12)	111.53(11)
C(16)-C(13)-C(14)	121.73(14)
C(16)-C(13)-S(12)	126.30(12)
C(14)-C(13)-S(12)	111.93(11)
O(21)-C(14)-N(15)	124.45(14)
O(21)-C(14)-C(13)	125.93(15)
N(15)-C(14)-C(13)	109.62(13)
C(13)-C(16)-C(17)	120.44(15)
C(13)-C(16)-H(16)	119.8
C(17)-C(16)-H(16)	119.8
O(28)-C(17)-O(18)	124.86(15)
O(28)-C(17)-C(16)	122.99(15)
O(18)-C(17)-C(16)	112.14(14)
O(18)-C(19)-C(20)	110.17(14)
O(18)-C(19)-H(19A)	109.6
C(20)-C(19)-H(19A)	109.6
O(18)-C(19)-H(19B)	109.6
C(20)-C(19)-H(19B)	109.6
H(19A)-C(19)-H(19B)	108.1
C(19)-C(20)-H(20A)	109.5

C(19)-C(20)-H(20B)	109.5
H(20A)-C(20)-H(20B)	109.5
C(19)-C(20)-H(20C)	109.5
H(20A)-C(20)-H(20C)	109.5
H(20B)-C(20)-H(20C)	109.5
C(23)-C(22)-C(27)	121.66(15)
C(23)-C(22)-N(15)	119.64(14)
C(27)-C(22)-N(15)	118.69(14)
C(22)-C(23)-C(24)	118.87(16)
C(22)-C(23)-H(23)	120.6
C(24)-C(23)-H(23)	120.6
C(25)-C(24)-C(23)	120.09(17)
C(25)-C(24)-H(24)	120.0
C(23)-C(24)-H(24)	120.0
C(26)-C(25)-C(24)	120.30(16)
C(26)-C(25)-H(25)	119.9
C(24)-C(25)-H(25)	119.9
C(25)-C(26)-C(27)	120.18(17)
C(25)-C(26)-H(26)	119.9
C(27)-C(26)-H(26)	119.9
C(22)-C(27)-C(26)	118.89(16)
C(22)-C(27)-H(27)	120.6
C(26)-C(27)-H(27)	120.6
C(2)-N(3)-C(4)	110.54(13)
C(11)-N(10)-C(2)	119.47(14)
C(14)-N(15)-C(11)	116.10(13)
C(14)-N(15)-C(22)	121.15(13)
C(11)-N(15)-C(22)	122.70(13)
C(17)-O(18)-C(19)	116.24(13)
C(5)-S(1)-C(2)	88.78(8)
C(13)-S(12)-C(11)	90.80(7)

Table 4. Anisotropic displacement parameters ($Å^2 x\ 10^3$) for **1**. The anisotropic displacement factor exponent takes the form: $-2\pi^2[\ h^2 a^{*2} U^{11} + ... + 2\ h\ k\ a^*\ b^*\ U^{12}\ ]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
C(2)	22(1)	19(1)	23(1)	11(1)	8(1)	5(1)
C(4)	24(1)	20(1)	24(1)	11(1)	9(1)	6(1)
C(5)	25(1)	23(1)	25(1)	12(1)	10(1)	9(1)
C(6)	32(1)	30(1)	32(1)	21(1)	13(1)	11(1)
C(7)	35(1)	25(1)	35(1)	19(1)	20(1)	10(1)
C(8)	26(1)	26(1)	35(1)	15(1)	11(1)	0(1)
C(9)	26(1)	30(1)	27(1)	16(1)	4(1)	1(1)
C(11)	16(1)	16(1)	26(1)	9(1)	8(1)	3(1)
C(13)	17(1)	18(1)	26(1)	10(1)	8(1)	5(1)
C(14)	17(1)	21(1)	27(1)	13(1)	9(1)	7(1)
C(16)	21(1)	24(1)	29(1)	15(1)	11(1)	6(1)
C(17)	23(1)	24(1)	24(1)	14(1)	9(1)	7(1)
C(19)	22(1)	33(1)	23(1)	14(1)	2(1)	1(1)
C(20)	25(1)	42(1)	36(1)	20(1)	7(1)	7(1)
C(22)	16(1)	24(1)	21(1)	12(1)	7(1)	2(1)
C(23)	23(1)	27(1)	26(1)	14(1)	9(1)	8(1)
C(24)	21(1)	47(1)	31(1)	23(1)	9(1)	11(1)
C(25)	20(1)	42(1)	23(1)	14(1)	4(1)	-3(1)
C(26)	31(1)	26(1)	28(1)	10(1)	11(1)	-2(1)
C(27)	23(1)	26(1)	31(1)	16(1)	12(1)	5(1)
N(3)	23(1)	24(1)	25(1)	15(1)	7(1)	2(1)
N(10)	20(1)	22(1)	25(1)	14(1)	8(1)	4(1)
N(15)	15(1)	20(1)	24(1)	12(1)	5(1)	2(1)
O(18)	20(1)	32(1)	26(1)	17(1)	4(1)	2(1)
O(21)	20(1)	28(1)	33(1)	20(1)	8(1)	2(1)
O(28)	26(1)	28(1)	31(1)	18(1)	2(1)	0(1)
S(1)	22(1)	33(1)	31(1)	22(1)	5(1)	4(1)
S(12)	18(1)	20(1)	24(1)	12(1)	5(1)	2(1)

Table 5. Calculated hydrogen coordinates (x 10^4) and isotropic displacement parameters (Å^{2}x 10^3) for **1**

	x	y	z	U(eq)
H(6)	10625	9960	6887	35
H(7)	13082	11618	8357	35
H(8)	14463	11571	10401	38
H(9)	13411	9937	11081	37
H(16)	9225	4913	13391	29
H(19A)	13429	7424	17438	36
H(19B)	13555	8400	16662	36
H(20A)	14151	6621	14840	56
H(20B)	13954	5579	15538	56
H(20C)	15267	7227	16518	56
H(23)	5176	5396	8557	32
H(24)	2937	3779	6430	39
H(25)	2379	1155	4934	41
H(26)	4052	134	5514	40
H(27)	6326	1750	7583	32

Table 6. Torsion angles [°] for **1**.

N(3)-C(4)-C(5)-C(6)	178.77(15)
C(9)-C(4)-C(5)-C(6)	-1.5(2)
N(3)-C(4)-C(5)-S(1)	-1.27(18)
C(9)-C(4)-C(5)-S(1)	178.45(13)
C(4)-C(5)-C(6)-C(7)	0.1(3)
S(1)-C(5)-C(6)-C(7)	-179.83(13)
C(5)-C(6)-C(7)-C(8)	1.2(3)
C(6)-C(7)-C(8)-C(9)	-1.2(3)
C(7)-C(8)-C(9)-C(4)	-0.2(3)
N(3)-C(4)-C(9)-C(8)	-178.78(16)
C(5)-C(4)-C(9)-C(8)	1.5(3)
C(16)-C(13)-C(14)-O(21)	2.8(3)
S(12)-C(13)-C(14)-O(21)	-179.29(13)

C(16)-C(13)-C(14)-N(15)	-177.86(14)
S(12)-C(13)-C(14)-N(15)	0.06(16)
C(14)-C(13)-C(16)-C(17)	177.13(14)
S(12)-C(13)-C(16)-C(17)	-0.5(2)
C(13)-C(16)-C(17)-O(28)	5.7(3)
C(13)-C(16)-C(17)-O(18)	-173.27(14)
C(27)-C(22)-C(23)-C(24)	-0.1(2)
N(15)-C(22)-C(23)-C(24)	-178.81(14)
C(22)-C(23)-C(24)-C(25)	1.0(2)
C(23)-C(24)-C(25)-C(26)	-0.7(3)
C(24)-C(25)-C(26)-C(27)	-0.6(3)
C(23)-C(22)-C(27)-C(26)	-1.1(2)
N(15)-C(22)-C(27)-C(26)	177.62(14)
C(25)-C(26)-C(27)-C(22)	1.4(2)
N(10)-C(2)-N(3)-C(4)	178.32(15)
S(1)-C(2)-N(3)-C(4)	0.03(18)
C(9)-C(4)-N(3)-C(2)	-178.88(16)
C(5)-C(4)-N(3)-C(2)	0.8(2)
N(15)-C(11)-N(10)-C(2)	-178.45(13)
S(12)-C(11)-N(10)-C(2)	1.9(2)
N(3)-C(2)-N(10)-C(11)	9.2(2)
S(1)-C(2)-N(10)-C(11)	-172.55(12)
O(21)-C(14)-N(15)-C(11)	-179.83(15)
C(13)-C(14)-N(15)-C(11)	0.80(19)
O(21)-C(14)-N(15)-C(22)	-2.4(2)
C(13)-C(14)-N(15)-C(22)	178.22(13)
N(10)-C(11)-N(15)-C(14)	178.96(14)
S(12)-C(11)-N(15)-C(14)	-1.31(17)
N(10)-C(11)-N(15)-C(22)	1.6(2)
S(12)-C(11)-N(15)-C(22)	-178.68(11)
C(23)-C(22)-N(15)-C(14)	116.76(17)
C(27)-C(22)-N(15)-C(14)	-62.0(2)
C(23)-C(22)-N(15)-C(11)	-66.0(2)
C(27)-C(22)-N(15)-C(11)	115.25(17)
O(28)-C(17)-O(18)-C(19)	-4.9(2)
C(16)-C(17)-O(18)-C(19)	174.04(13)

C(20)-C(19)-O(18)-C(17)	-84.01(18)
C(6)-C(5)-S(1)-C(2)	-179.02(17)
C(4)-C(5)-S(1)-C(2)	1.01(12)
N(3)-C(2)-S(1)-C(5)	-0.64(13)
N(10)-C(2)-S(1)-C(5)	-179.12(13)
C(16)-C(13)-S(12)-C(11)	177.16(15)
C(14)-C(13)-S(12)-C(11)	-0.65(12)
N(10)-C(11)-S(12)-C(13)	-179.21(16)
N(15)-C(11)-S(12)-C(13)	1.09(12)

Table 7. Hydrogen bonds for **1** [Å and °]

D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
C(16)-H(16)...O(18)#1	0.95	2.53	3.461(2)	165.1
C(19)-H(19A)...O(21)#1	0.99	2.49	3.426(2)	156.6
C(23)-H(23)...O(21)#2	0.95	2.65	3.257(2)	122.1

Symmetry transformations used to generate equivalent atoms:

#1 -x+2,-y+1,-z+3 #2 -x+1,-y+1,-z+2

Table 8. A comparison of literature reported solvent with present methodology

Group	Synthetic strategy	Detection limit	Fluorescence response	Solvent	Ref.
Kaur *et al.*	Bis(N-methylindolyl)methane-based chemical probes	Cu =0.12 μM Hg=9.96 μM	quenching	Acetonitrile	24a
Kaur *et al.*	Indole-Based Chemosensor	Cu =1.67 μM, Hg=1.21 μM,	quenching	Acetonitrile	24b
Present work	Regioselective benzothiazole based receptor	Cu = 0.36 μM Hg= 2.49 μM	quenching	Aq. Methanol	This work