Electronic Supplementary Information:

BODIPY-functionalized bimetallic probe for sensitive and selective color-fluorometric chemosensing of Hg^{2+}

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Experimental Section

Preparation of compound 5: Compound **5** was prepared by the previouly reported method.[1]

Preparation of compound 4. A solution of NaOH (0.17 g, 4.27 mmol) in water (5 mL) was added dropwise to a solution of **5** (0.22g, 0.43 mmol) and p-toluenesulfonyl chloride (0.25 g, 1.38 mmol) in THF (20 mL) over 3 h under nitrogen atmosphere at 0 °C then stirred for 30 min to ensure a homogeneous solution. The solution was warmed to room temperature and stirred for an additional 24 h. After removing the solvent (THF), the reaction mixture was extracted with CH_2Cl_2 (3 x 50 mL), washed twice with water, and dried over anhyd. $MgSO_4$. The crude product was purified by flash column chromatography on silica gel, by elution with (ethyl acetate/*n*-hexane, 2:1 v/v) to afford a blue solid 0.1 g, yield 28.5%).^{1}H NMR (300 MHz ,$CDCl_3$): 7.8(m, 8H), 7.4(m, 9H), 6.8(s, 1H) 6.5(d, 2H, *J* = 9.0Hz), 6.1(S, 1H), 4.2(t, 4H, *J* = 5.7Hz), 3.7(t, 4H, *J* = 5.4Hz) 2.5(s, 3H), 2.4(s, 6H), 1.45(s, 3H), 1.4(s, 3H) ppm. ^{13}C NMR (300 MHz , $CDCl_3$): 165.6, 150.8, 145.4, 143.3, 140.8, 139.3, 138.1, 135.8, 133.5, 132.2, 130.8, 129.7, 128.6, 128.3, 127.9, 126.1,124.6,123.5, 116.8, 112.1, 109.5, 60.7, 55.8, 22.5, 15.9, 14.3ppm. MS (FAB): $[M + H]^+$ *m/z* = 823.3576 (calcd*M*w = 823.2733); Anal. Calcd for $C_{44}H_{44}BF_2N_3O_6S_6$: C, 64.15.; H, 5.38; N, 5.10. Found: C, 64.51; H, 5.26; N, 5.07.

Preparation of BODIPY-conjugate (2). A solution of **4** (0.1g, 0.12 mmol) in anhydrous DMF (10 mL) was added dropwise over 1 h to a mixture of compound **3** (0.05g, 2.4 mmol) and Cs_2CO_3 (0.7g, 2.4 mmol) in anhydrous THF(10 mL) at 60 °C. The reaction mixture continued to stir for 2 days. The reaction was filtered, and the salts were washed with DMF (20 mL). Water (50 mL) and brine (50 mL) were added to the filtrate, and the reaction was extracted with EtOAc (1 x 50 mL). The organic phase was washed with water (2 x 50 mL) and brine (2 x 50 mL), dried over Na_2SO_4, filtered, and concentrated. The crude product was purified by flash column chromatography on silica gel, by elution with (ethyl acetate/*n*-hexane, 1:3 v/v) to afford a blue solid 0.2 g, yield 20.5%). ^{1}H NMR (300 MHz ,$CDCl_3$): 7.7(d, 2H, *J* = 8.4Hz), 7.5(m, 6H), 6.6(s, 1H) 6.5(d, 2H, *J* = 8.7Hz), 6.0(S, 1H), 4.2(t, 4H, *J* = 4.5Hz), 3.6(m, 6H) 3.7(t, 4H, *J* = 6.6Hz), 3.1(m, 2H), 2.7(m, 4H) , 2.6(s, 3H), 2.5(m,

2H),2.2(m, 3H), 2.0(m, 2H), 1.9(m, 4H), 1.7(m, 7H) 1.66(s, 3H), 1.60(s, 3H) ppm. ^{13}C NMR (300 MHz, $CDCl_3$): 165.8, 149.8, 143.3, 138.8, 138.3, 135.8,132.5, 132.2, 129.7, 128.6, 128.3, 127.9, 126.1,124.6,123.5, 116.8, 111.8, 109.6, 57.7, 56.8, 40.3, 38.6, 35.1, 32.9, 30.5, 29.8, 28.1, 27.8, 22.5, 14.3ppm.δ, 170.8. MS (FAB): $[M + H]^+$ *m/z* = 895.3576 (calcd*M*w = 895.3173); Anal. Calcd for $C_{46}H_{60}BF_2N_5S_6$: C, 61.65.; H, 6.75; N, 4.69. Found: C, 61.78; H, 6.54; N, 6.68.

Preparation of Au-Fe_3O_4 nanoparticles: The bi-functional Au−Fe_3O_4 nanoparticles were prepared by decomposition of $Fe(CO)_5$ over prepared Au nanoparticle surfaces following a previously described methodology of mixing the gold nanoparticles with $Fe(CO)_5$ in 1-octadecene solvent in the presence of oleic acid and oleylamine and then heating to reflux the mixture at approximately 300 °C followed by the room-temperature oxidation under air as is outlined in Scheme S1.[2]

Immobilization of BODIPY-derivative 2 onto Au-Fe_3O_4 nanoparticles (1): Compound **2** (30 mg, 0.054 mmol) was dissolved in CH_3CN (5mL) to which Au-Fe_3O_4 colloid solution (20 mL) was added, and it was stirred at room temperature for 24 h. The immobilized nanoparticle solutions were centrifuged for 30 min and re-dispersed in aqueous solution after the supernatant was removed. The particles were washed three times more and finally resuspended in the corresponding detection buffers.

Preparation of 2-modified electrode and its fluorescence images: Platinum electrodes were polished with 0.3 μm alumina powder (Buehler, Lake Bluff, MN) and rinsed with deionized water. Then, the electrodes were immersed in a chloroform solution containing 1.0 mM **2** for 12 h. After the electrodes were removed from the solution, they were thoroughly rinsed with acetone and chloroform to remove excess **2** and dried using compressed nitrogen gas. The substrate was immersed in each solution containing metal ions for 20 min. This optimum reaction time was estimated from the saturation of the emission decrease after Hg^{2+} addition (0.01 M) in water at room temperature. After washing three times with methanol and water, the BODIPY modified platinum electrode was dried in nitrogen. To investigate the properties of the molecular switches, the BODIPY modified platinum electrode was immersed in 0.5 M NaOH for 30 min. Fluorescence images were taken with a Bio-Rad Radiance2000

confocal and multi-photon system (confocal excitation: an Ar ion laser at 488 nm). The scan area was 60-60 mm.

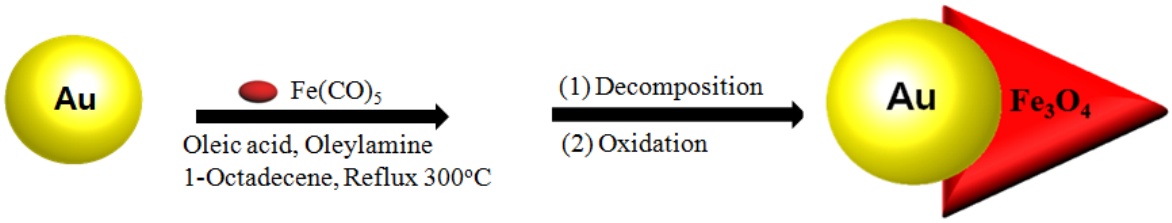
Characterization: IR spectra were obtained for KBr pellets, in the range 400–4000 cm^{-1}, with a Shimadzu FTIR 8400S instrument, and transmission electron microscopy (TEM) images were taken with a JEOL JEM-2100 F instrument operated at 50 keV. Images were recorded on 2k CCD (Gatan Inc. USC 1000).

Photospectroscopy: Fluorescence emission spectra were recorded with a Shimadzu RF-5301-PC instrument. Stock solutions (0.01 M) of the hydrated metal nitrate salts or $Hg(NO_3)_2$ were prepared in H_2O at pH 7.4. Stock solutions of **1** were prepared in pure H_2O at pH 7.4. For all measurements, excitation was at 591 nm, with excitation and emission slit widths of 1.5 nm. The pH value was adjusted by using 20 mM HEPES (pH 7.4).

Reference

1) H. Son, H. Y. Lee, J. M. Lim, D. Kang, W. S. Han, S. S. Lee and J. H. Jung, *Chem. Eur. J.*, 2010, **16**, 11549-11553.

2) H. Yu, M. Chen, P. M. Rice, S. X. Wang, R. L. White and S. Sun, *Nano Lett.,* 2005, **5**, 379-382.



Scheme S1. Preparation Method of Au-Fe_3O_4 Nanoparticles

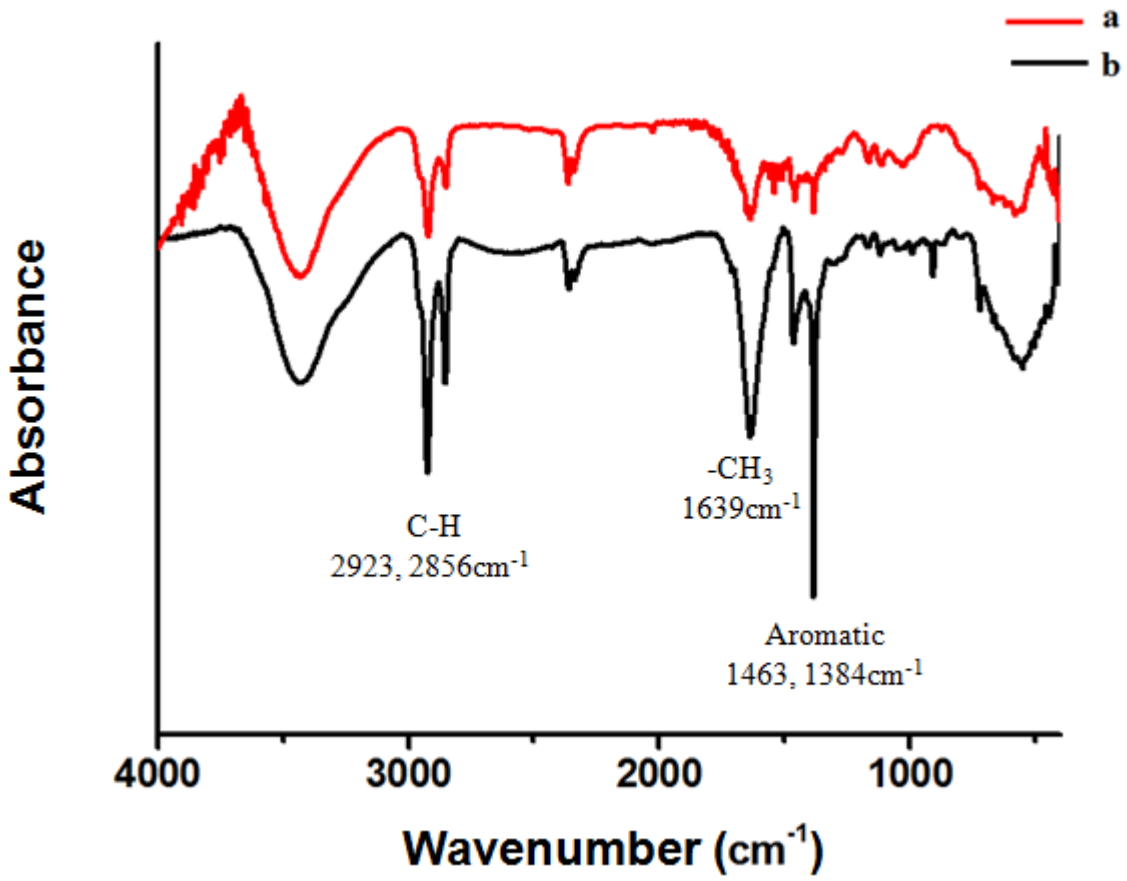


Fig. S1 FT-Infrared spectra of (a) Au-Fe_3O_4 nanoparticle and (b) **2**-immobilized Au-Fe_3O_4 nanoparticle (**1**).

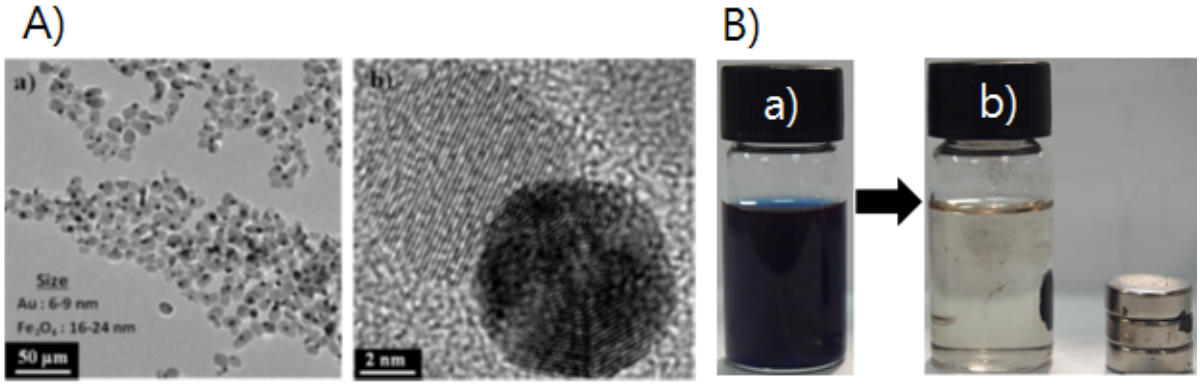


Fig. S2 (A) (a) TEM image of **2**–immobilized Au-Fe_3O_4 nanoparticles (**1**). (b) HRTEM image of one Au-Fe_3O_4 particle. Dark area: Au; Light Area: Fe_3O_4. (B) Photograph of **1** (a) without and (b) with magnet in water.

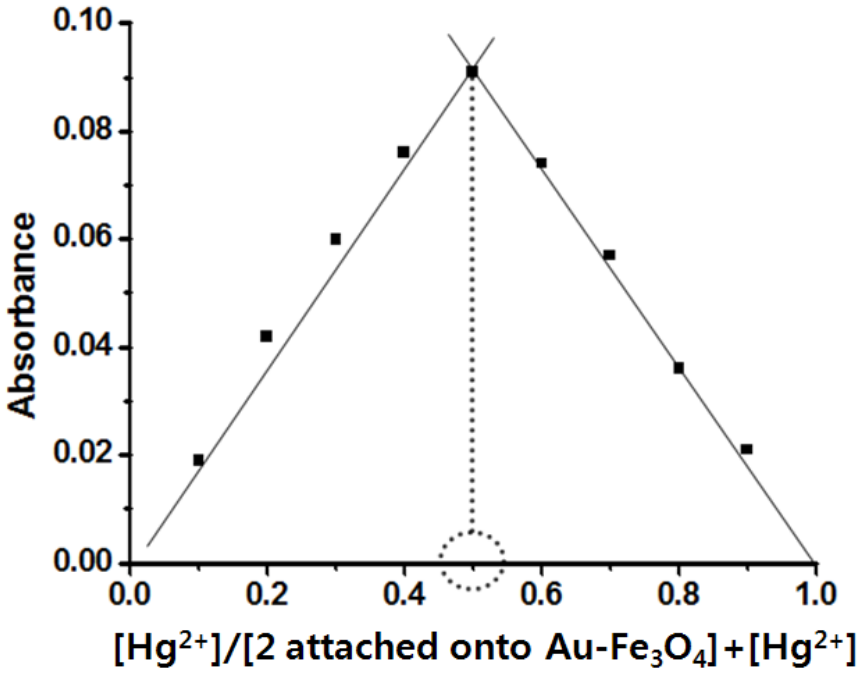


Fig. S3 Job′s plot of 1:1 complex of **1** (10 μM) and Hg^{2+}. The pH value was adjusted by using 20 mM HEPES in aqueous solution at pH 7.4.

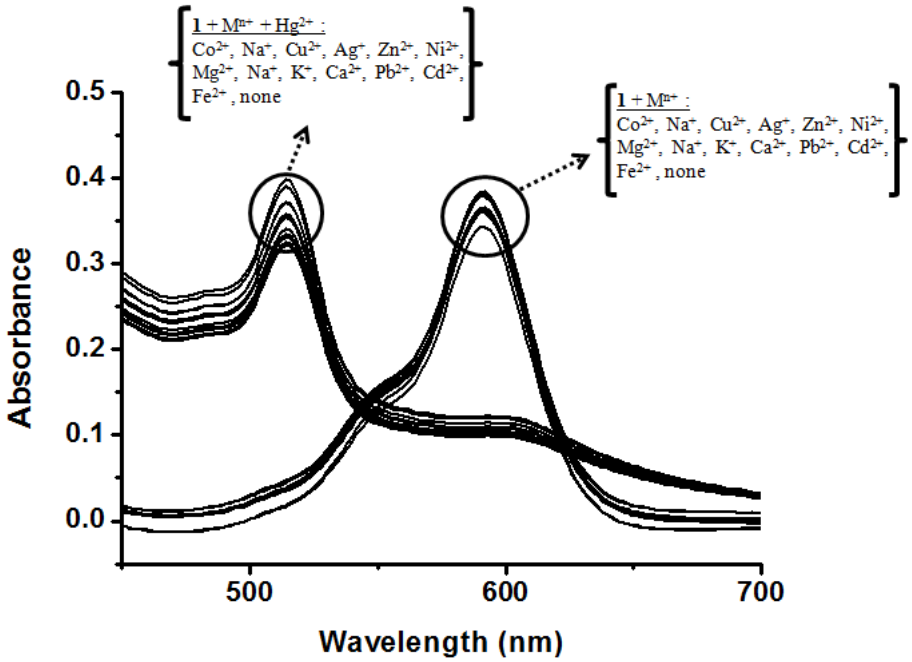


Fig. S4 Absorption spectra of **1** (10 μM) upon addition of Co^{2+}, Na^{+}, Cu^{2+}, Ag^{+}, Zn^{2+}, Ni^{2+} ,Mg^{2+} Na^{+}, K^{+}, Ca^{2+}, Pb^{2+}, Cd^{2+} or Fe^{2+} (100 equiv) and subsequent addition of Hg^{2+} (100 equiv) in aqueous solution. For all measurements, the pH value was adjusted by using 20 mM HEPES in aqueous solution at pH 7.4.

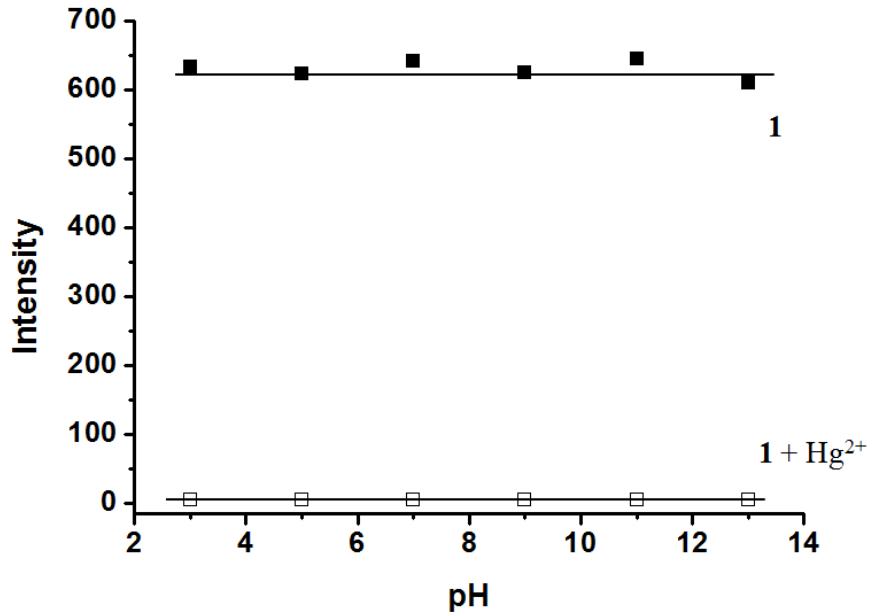


Fig. S5 Plot of pH exposure and the resulting fluorescence intensity at 657nm of **1** (10 μM) without and with $Hg(NO_3)_2$ (100 equiv) in pure aqueous solution represented by shaded and unshaded squares respectively (λ_{ex}=591 nm).

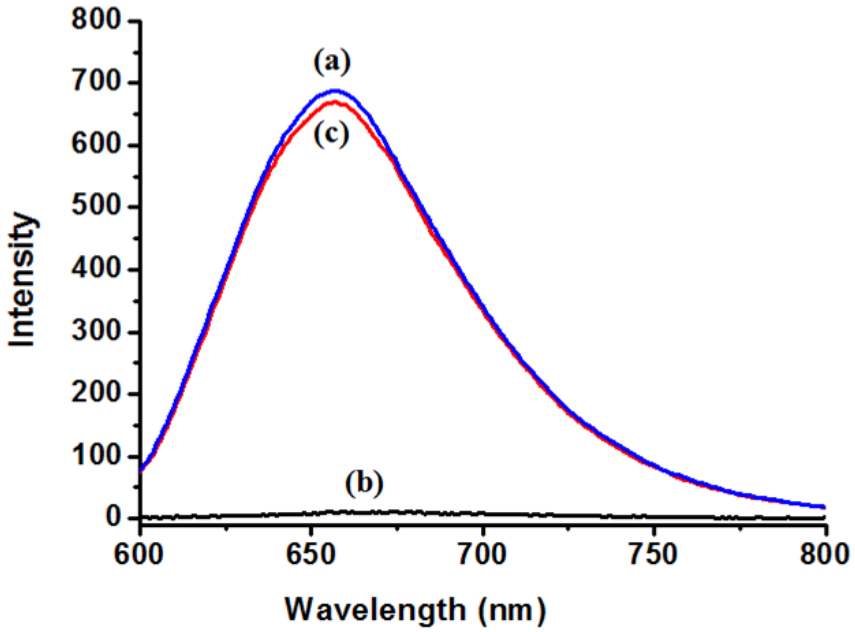


Fig. S6 Fluorescence spectra of **1** (10 μM) (a) without then (b) with Hg^{2+} ion (100 equiv), followed by (c) treatment with EDTA (10 μM) (λ_{ex}=591 nm).

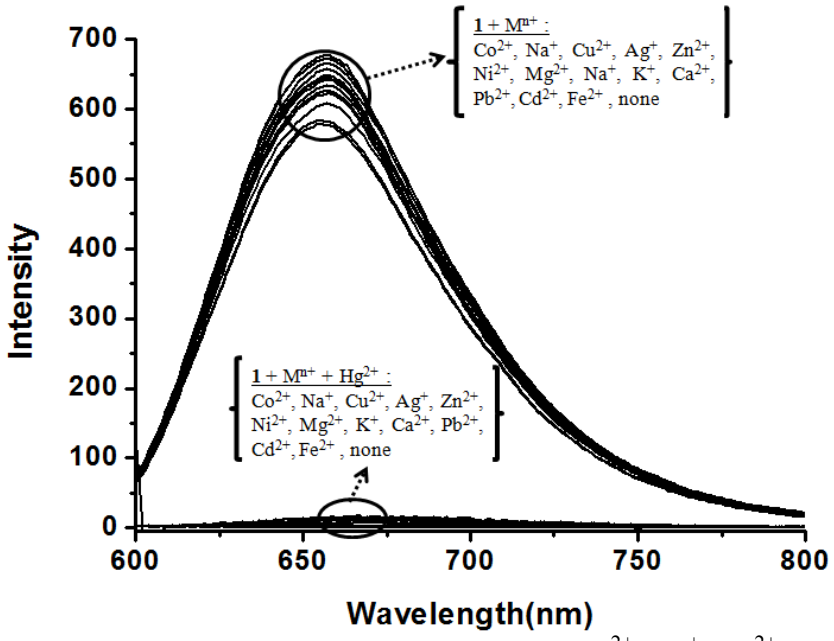


Fig. S7 Fluorescence responses of **1** (10 μM) upon addition of Co^{2+}, Na^{+}, Cu^{2+}, Ag^{+}, Zn^{2+}, Ni^{2+}, Mg^{2+}, K^{+}, Ca^{2+}, Pb^{2+}, Cd^{2+} or Fe^{2+} (100 equiv) and subsequent addition of Hg^{2+} (100 equiv) in aqueous solution. For all measurements, the pH value was adjusted by using 20 mM HEPES in aqueous solution at pH 7.4. (λ_{ex}=591 nm).

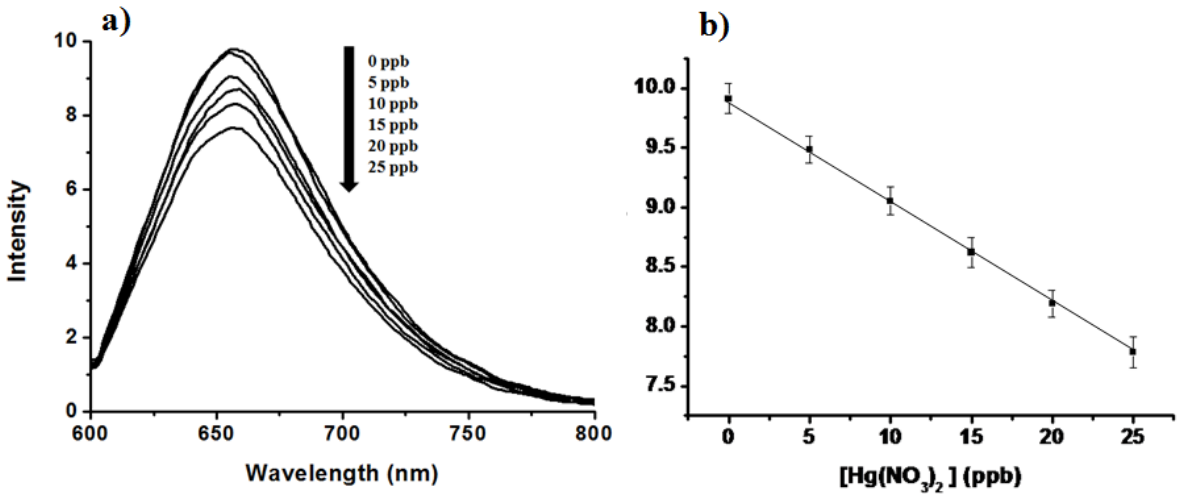


Fig. S8 Fluorescence spectra of **1** (0.1 μM) upon addition of increasing Hg^{2+} concentrations in aqueous solution at pH 7.4 (as indicated by the direction of the black arrow). (B) Calibration curve of mercury concentration against fluorescence intensity of **1**.