Supporting Information

for

Carbazole-thiosemicarbazone-Hg(II) ensemble-based colorimetric and fluorescence turn-on toward iodide in aqueous media and its application in live cell imaging

Ajit Kumar Mahapatra*, Jagannath Roy, Prithidipa Sahoo† Subhra Kanti Mukhopadhyay‡, Amarnath Chattopadhyay ‡

Department of Chemistry, Bengal Engineering and Science University, Shibpur, Howrah – 711103, India.
† Present address: Chemistry and Chemical Biology Department, Rutgers University, 610 Taylor Road, Piscataway, NJ 08854, USA
‡ Department of Microbiology, The University of Burdwan, Burdwan, West Bengal, India

E-mail: mahapatra574@gmail.com

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Preparation of cells

*Candida albicans* cells (IMTECH No. 3018) from exponentially growing culture in yeast extract glucose broth medium (pH 6.0, incubation temperature, 37°C) were centrifuged at 3000 rpm for 10 minutes, washed twice with 0.1 M HEPES buffer at pH 7.4. Then, it was treated with 3 (10^-5M) ligand for 45 minutes in 0.1 M HEPES buffer (pH 7.4) containing 0.01 % Triton X100 as permeability enhancing agent. After incubation the cells were washed again with HEPES buffer at pH 7.4. 3 (10^-5M) treated cells were mounted on a grease free glass slide and observed under a Leica DM 1000 fluorescence microscope equipped with UV filter. Cells without 3 treatment were used as control. Then 10^-5M Hg^2+ (aqueous solution) was added to the specimen at the point of observation with the help of a micropipette. The change in fluorescence intensity was observed and recorded. After that 10^-5M KI was added in similar way to detect the change.
Figure S1: UV-vis spectra recorded for compound 3 (5μM) in the absence or presence of various heavy and transition metal species in pH 7.0 HEPES buffer (25 mM, pH 7.4, containing 1.0% DMSO).

**Binding constant curve for receptor 3 with Hg(II) from UV-vis in pH 7.4 HEPES buffer (25 mM, pH 7.4, containing 1.0% DMSO).**

**Figure S2:** Binding constant curve for 3 (c = 1.00 x 10^{-5} M) with Hg(II) (c = 5.00 x 10^{-4} M) [Determined using non-linear curve fitting y= (A₀+A*K*x)/(1+K*x). x = [G], y = absorbance in pH 7.4 HEPES buffer (25 mM, pH 7.4, containing 1.0% DMSO).]
Binding constant curve for receptor 3 with Hg(II) from fluorescence in pH 7.4 HEPES buffer (25 mM, pH 7.4, containing 1.0% DMSO).

Figure S3. (a) Fluorescence spectra of compound 3 (5 μM) in the absence or presence of various heavy and transition -metal species in pH 7.4 HEPES buffer (30 mM, pH 7.4, containing 1.0% DMSO), (b) Job’s plot of compound 3 with Hg$^{+2}$ according to the method of continuous variations. The total concentrations of compound 4 and Hg$^{+2}$ were kept constant at 20 μM. (c) Binding constant curve of 3 (c = 1.00 x 10$^{-5}$ M) with Hg(II) (c = 2.0 x 10$^{-4}$ M) from fluorescence titration in pH 7.4 HEPES buffer (25 mM, pH 7.4, containing 1.0% DMSO). Working formula $y=I_0+((I-I_0)/(2*x_2))*(x_1+x_2+1/K-((x_1+x_2+1/K)^2-4*x_1*x_2)^.5)$, $x_1=[G]$, $x_2=[H]$, $y=$ intensity.
Figure S4. ESI-MS spectrum of Compound 3 +Hg²⁺
Figure S5. The absorption spectra of the 3- Hg$^{2+}$ ensemble (10μM) in pH 7.4 HEPES buffer (30 mM, pH 7.4, containing 1.0% DMSO) in the presence of Iodide anions.
Figure S6. Plot of the fluorescence intensity (at 425 nm) as a function of the concentrations of Iodide anions. The concentration of the 3-Hg ensemble was 5 μM.
Figure S7. Fluorescence intensity response of 3–Hg ensemble toward various anions. 1. 3–Hg\(^{2+}\), 2. F\(^{-}\), 3. Cl\(^{-}\), 4. Br\(^{-}\), 5. NO\(_3\)\(^{-}\), 6. I\(^{-}\), 7. NO\(_2\)\(^{-}\), 8. SO\(_4\)\(^{2-}\), 9. SO\(_3\)\(^{-2}\), 10. CO\(_3\)\(^{2-}\), 11. PO\(_4\)\(^{3-}\), 12. CH\(_3\)COO\(^{-}\), 13. N\(_3\)\(^{-}\), 14. CN\(^{-}\).
Figure S8 The fluorescence intensity changes of the 3–Hg ensemble to Iodide anions in the presence of various test anions. 1. I, 2. I+ F, 3. I+ Cl, 4. I+ Br. 5. I+ NO₃, 6. I+ NO₂, 7. I+ SO₄²⁻, 8. I+ SO₃²⁻, 9. I+ CO₃²⁻, 10. I+ PO₄³⁻, 11. I+ CH₃COO⁻, 12. I+ N₃⁻, 13. I+ CN⁻.
Figure S9. The pH effects on the fluorescence intensity at 425nm of the 3-Hg$^{+2}$ ensemble (5 μM) (▸), and the ensemble (5 μM) toward iodide anions (10 μM) (●).

Figure S10. 1H NMR (400 MHz) spectra of Compound 3 (a), 3 + 0.5 equivalent Hg$^{+2}$ (b), 3 + 1 equivalent Hg$^{+2}$ (c), and 3 + 1.5 equivalent Hg$^{+2}$ in d$_6$ DMSO-D$_2$O (d).
Figure S11. $^1$H NMR spectrum of compound 2.
Figure S12. $^{13}$C NMR spectrum of compound 2.
Figure S13. $^1$H NMR spectrum of compound 3
Figure S14. $^{13}$C NMR spectrum of compound 3.
Figure S15. ESI-MS spectrum of Compound 3.