

Electronic Supplementary Information

Ratiometric detection of mercury ion in water: accelerated response kinetics of azo chromophores having ethynyl ligand tethers

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Experimental Section

General Considerations. All reagents were obtained from commercial suppliers and used as received unless otherwise noted. The compound 2,6-bis(3-methyl-3-hydroxyl-1-butynyl)-4-*tert*-butylaniline (**5**)¹ and 4-(*tert*-butyl)-2-iodoaniline² were prepared according to literature procedures. All air-sensitive manipulations were carried out by standard Schlenk-line techniques.

Physical Measurements. ¹H-NMR and ¹³C-NMR spectra were recorded on a 400 MHz Varian Inova NMR Spectrometer. Chemical shifts were reported versus tetramethylsilane and referenced to the residual solvent peaks. High resolution electrospray ionization (ESI) mass spectra were obtained on a Thermo Electron Corporation MAT 95XP-Trap. FT-IR spectra were recorded on a Nicolet 510P FT-IR spectrometer with EZ OMNIC ESP software. UV-vis spectra were recorded on a Agilent 8453 UV-vis spectrophotometer with ChemStation. Reactivity screening and competition studies with metal ions in water were conducted using Hg(ClO₄)₂, MnCl₂, CoCl₂, NiCl₂, Cu(NO₃)₂, Zn(ClO₄)₂, Cd(ClO₄)₂, MgCl₂, and CaCl₂. Additional metal salts, including Hg(OAc)₂, FeCl₃, and Pb(ClO₄)₂, were used in UV-vis measurements in MeOH.

4-(2-Amino-5-(*tert*-butyl)phenyl)-2-methylbut-3-yn-2-ol (6**).** A round-bottom flask charged with 4-*tert*-butyl-2-iodoaniline (1.61 g, 5.85 mmol), 2-methylbut-3-yn-2-ol (601 mg, 7.14 mmol), *i*-Pr₂NH (20 mL), PdCl₂(PPh₃)₂ (51.2 mg, 72.9 μmol), CuI (35.1 mg, 184 μmol), and THF (20 mL) was purged with N₂, stirred at r.t. for 24 h, and concentrated under reduced pressure. Flash column chromatography on SiO₂ (hexanes:EtOAc = 2:1, v/v) furnished **6** as a pale yellow solid (1.21 g, 5.23 mmol, 89%). ¹H NMR (400 MHz, CDCl₃, 298 K): δ 7.25 (s, 1H), 7.16 (d, *J* = 8.8 Hz, 1H), 6.64 (d, *J* = 8.8 Hz), 4.03 (s, 2H), 2.11 (s, 1H), 1.64 (s, 6H), 1.25 (s, 9H); ¹³C NMR (100 MHz, CDCl₃, 298 K) δ 145.2, 140.8, 128.7, 126.9, 114.2, 106.8, 98.8, 79.2, 65.7, 33.8, 31.7, 31.3. FT-IR (thin film on NaCl, cm⁻¹): 3360, 2963, 2866, 2210, 1620, 1501, 1463, 1409, 1362, 1303, 1252, 1161, 970, 935, 890, 872, 820, 740. HR-MS (ESI): calcd for C₁₅H₂₁NONa [M + Na]⁺ 254.1521, found 254.1510.

(*E*)-4-((4-(*tert*-Butyl)-2,6-bis(3-hydroxy-3-methylbut-1-yn-1-yl)phenyl)diazenyl)-benzene-1,3-diol (3**).** A 20 mL vial was charged with **5** (0.301 g, 0.960 mmol), MeOH (5 mL), aq HCl (2 M, 3 mL), and a magnetic stir bar. The reaction mixture was cooled to 0 °C using an ice bath and stirred. A pre-cooled (0 °C) aq solution (3 mL) of NaNO₂ (82.1 mg, 1.18 mmol) was added using a pipette over a period of 10 min, and the reaction mixture was stirred for additional 10 min at 0 °C. A 250-mL round-bottom flask was charged with resorcinol (101 mg, 0.917 mmol), aq NaOH (2 M, 3.10 mL), water (5 mL), and a magnetic stir bar, and cooled to 0 °C. Using a pipette, the diazonium reaction mixture of **5** was delivered over a period of 20 min. The resulting

¹H. Y. Lee, X. Song, H. Park, M.-H. Baik, and D. Lee, *J. Am. Chem. Soc.* **2010**, *132*, 12133–12144.

²J. Iskra, S. Stavber and M. Zupan, *Synthesis* **2004**, 1869–1873.

mixture was stirred for additional 0.5 h at 0 °C and then neutralized by addition of aq HCl (2 M). The mixture was diluted with water (50 mL), and the precipitates were isolated by filtration, washed several times with water, and dried in air to furnish **3** as a dark red solid (0.321 g, 0.738 mmol, 80%). ¹H NMR (400 MHz, DMSO-*d*₆, 298 K): δ 13.0 (s, 1H), 10.7 (s, 1H), 7.77 (d, *J* = 12.0 Hz, 1H), 7.47 (s, 2H), 6.58 (d, *J* = 12.0 Hz, 1H), 6.35 (s, 1H), 5.51 (s, 2H), 1.42 (s, 12H), 1.31 (s, 9H); ¹³C NMR (100 MHz, DMSO-*d*₆, 298 K) δ 163.6, 155.5, 151.0, 149.1, 134.0, 132.0, 130.4, 116.2, 109.4, 102.9, 100.3, 78.0, 63.7, 34.4, 31.3, 30.6. FT-IR (thin film on NaCl, cm⁻¹): 3138, 2968, 2928, 1623, 1585, 1559, 1544, 1472, 1436, 1404, 1376, 1362, 1325, 1249, 1216, 1196, 1161, 1120, 950, 881, 850, 816, 737. HR-MS (ESI): calcd for C₂₆H₃₁N₂O₄ [M + H]⁺ 435.2284, found 435.2265.

(*E*)-4-((4-(*tert*-Butyl)-2-(3-hydroxy-3-methylbut-1-yn-1-yl)phenyl)diazanyl)benzene-1,3-diol (2). This compound was prepared using **6** (0.316 g, 1.36 mmol) and resorcinol (0.150 g, 1.36 mmol) in a manner similar to that described for **3**. The neutralized solution was extracted into EtOAc (100 mL × 2) and washed with water (100 mL × 2). Volatile fractions were removed under reduced pressure, and the residual material was purified by flash column chromatography on SiO₂ (hexanes:EtOAc = 4:1 to 2:1, v/v) to furnish **2** as a red solid (0.322 g, 0.913 mmol, 67%). ¹H NMR (400 MHz, CD₃OD, 298 K): δ 7.82 (d, *J* = 8.4 Hz, 1H), 7.88 (d, *J* = 8.8 Hz, 1H), 7.58 (s, 1H), 7.53 (d, *J* = 8.8 Hz, 1H), 6.54 (d, *J* = 8.8 Hz, 1H), 6.33 (s, 1H), 1.63 (s, 6H), 1.35 (s, 9H); ¹³C NMR (100 MHz, CD₃OD, 298 K) δ 163.3, 157.0, 152.9, 148.2, 134.8, 132.7, 129.2, 126.3, 120.7, 114.2, 109.1, 102.8, 100.2, 78.0, 64.8, 34.2, 30.3, 30.0. FT-IR (thin film on NaCl, cm⁻¹): 3198, 2978, 2963, 2868, 1622, 1496, 1472, 1414, 1376, 1363, 1311, 1241, 1202, 1157, 1115, 972, 933, 840, 831, 818. HR-MS (ESI): calcd for C₂₁H₂₄N₂O₃Na [M + Na]⁺ 375.1685, found 375.1684.

(*E*)-4-((4-(*tert*-Butyl)phenyl)diazanyl)benzene-1,3-diol (1). This compound was prepared using 4-*tert*-butylaniline (0.209 g, 1.40 mmol) and resorcinol (0.167 g, 1.51 mmol) in a manner similar to that described for **3**. The neutralized solution was extracted into EtOAc (100 mL × 2) and washed with water (100 mL × 2). Volatile fractions were removed under reduced pressure, and the residual material was purified by flash column chromatography on SiO₂ (hexanes:EtOAc = 10:1, v/v) to furnish **1** as an orange solid (0.312 g, 1.15 mmol, 82%). ¹H NMR (400 MHz, CDCl₃, 298 K): δ 13.7 (s, 1H), 7.78 (m, 3H), 7.52 (m, 2H), 6.54 (d, *J* = 8.8 Hz, 1H), 6.42 (s, 1H), 5.19 (s, 1H), 1.36 (s, 9H); ¹³C NMR (100 MHz, CDCl₃, 298 K) δ 161.8, 158.2, 153.7, 147.3, 135.3, 133.1, 126.4, 121.2, 109.5, 104.0, 35.1, 31.4. FT-IR (thin film on NaCl, cm⁻¹): 3287, 2963, 2904, 2869, 1616, 1506, 1472, 1419, 1377, 1316, 1249, 1199, 1160, 1116, 976, 836, 810, 738. HR-MS (ESI): calcd for C₁₆H₁₉N₂O₂ [M + H]⁺ 271.1447, found 271.1435.

Reaction between 2 and Hg²⁺. To a stirred solution of Hg(OAc)₂ (12.3 mg, 38.5 μmol) in CD₃OD (3 mL) was added dropwise a solution of **2** (6.2 mg, 17.5 μmol) in CD₃OD (5 mL) over a period of 30 min. During the course of the addition, a white solid material precipitated out, which was separated by filtration. The filtrate was analyzed by ¹H/¹³C NMR (Figs. 3 and S5) and MALDI-TOF mass spectrometry (Fig. S6) without further purification.

X-ray Crystallographic Studies on 3. A red block (0.15 × 0.15 × 0.12 mm³) was placed onto the tip of a 0.05-mm diameter glass fiber and mounted on a Bruker APEX II Kappa Duo diffractometer equipped with an APEX II detector at 150(2) K. The data collection was carried out using MoKα radiation (graphite monochromator) with a frame time of 30 seconds and a detector distance of 5.00 cm. A collection strategy was calculated and complete data to a resolution of 0.70 Å with a redundancy of 4 were collected. Four sections of frames were collected with 0.50° ω and φ scans. Data to a resolution of 0.75 Å were considered in the reduction. Final cell constants were calculated from the xyz centroids of 1933 strong reflections from the actual data collection after

integration. The intensity data were corrected for absorption.

The crystal was pseudo-merohedrally twinned. The unit cell appeared to be *C*-centered monoclinic due to the presence of pseudo-symmetry element (2-fold axis), but the structure could be solved only in triclinic $P\bar{1}$ setting. The twin law was determined to be $(-100/00-1/0-10)$. A direct methods solution was calculated, which provided most atomic positions from the E-map. Full-matrix least squares / difference Fourier cycles were performed, which located the remaining non-hydrogen atoms. All non-hydrogen atoms were refined with anisotropic displacement parameters. The hydrogen atoms were placed in ideal positions and refined as riding atoms with relative isotropic displacement parameters. The ratio of the twin components was refined to be 0.4503. The final full matrix least squares refinement converged to $R1 = 0.1111$ and $wR2 = 0.1669$ (F^2 , all data). The remaining electron density is minuscule and located on bonds. The asymmetric unit consisted of four molecules. One molecule contained the disorder of the *tert*-butyl group which showed two possible orientations. Hydrogen bonding interactions between molecules were observed.

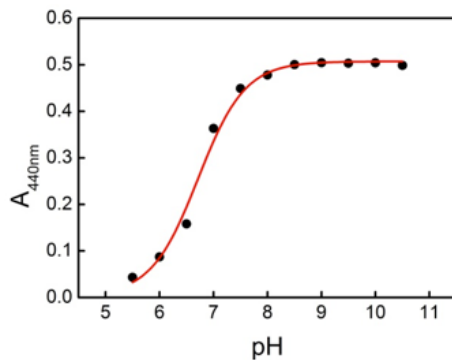


Fig. S1. pH-Dependent changes in absorbance of **3** at $\lambda = 440$ nm, $T = 298$ K. The red curve overlaid on the experimental data points is a theoretical fit with $pK_a = 6.7$.

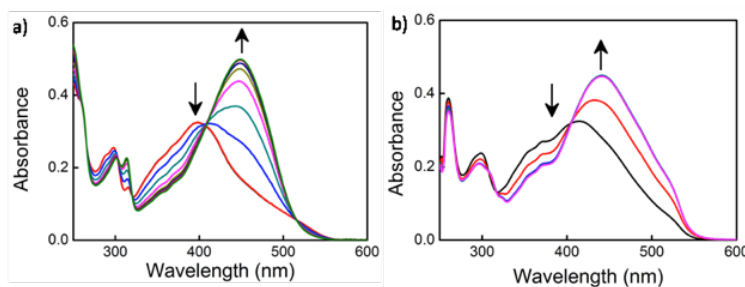


Fig. S2. UV-vis titration of **3** with DBU (a) in CHCl_3 ($[\text{DBU}] = 5, 50, 90, 130, 170, 210, 250,$ and 290 equiv), and (b) in DMSO ($[\text{DBU}] = 40, 80, 120,$ and 160 equiv).

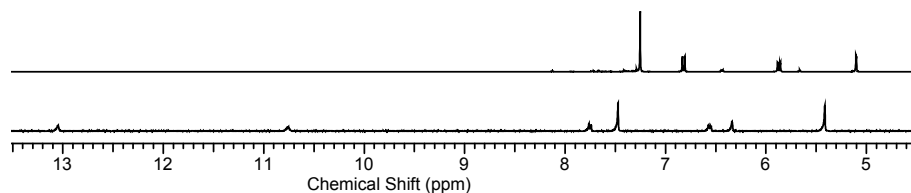


Fig. S3. Partial ^1H NMR spectra of **3** in $\text{DMSO-}d_6$ prior to (bottom) and after (top) addition of DBU (1.2 equiv) at $T = 298$ K.

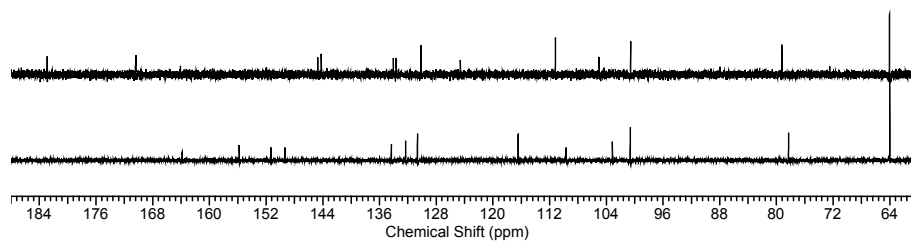


Fig. S4. Partial ^{13}C NMR spectra of **3** in $\text{DMSO-}d_6$ prior to (bottom) and after (top) addition of DBU (1.2 equiv) at $T = 298$ K.

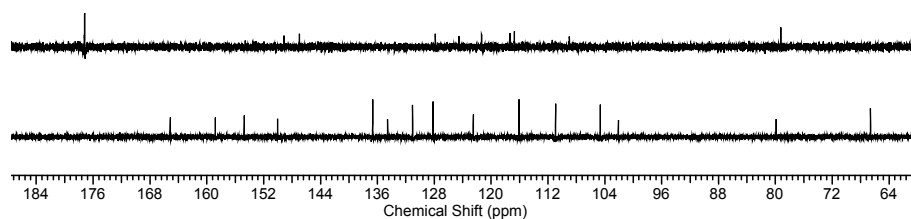


Fig. S5. Partial ^{13}C NMR spectra of **3** (bottom) and **4** (top) in CD_3OD at $T = 298$ K.

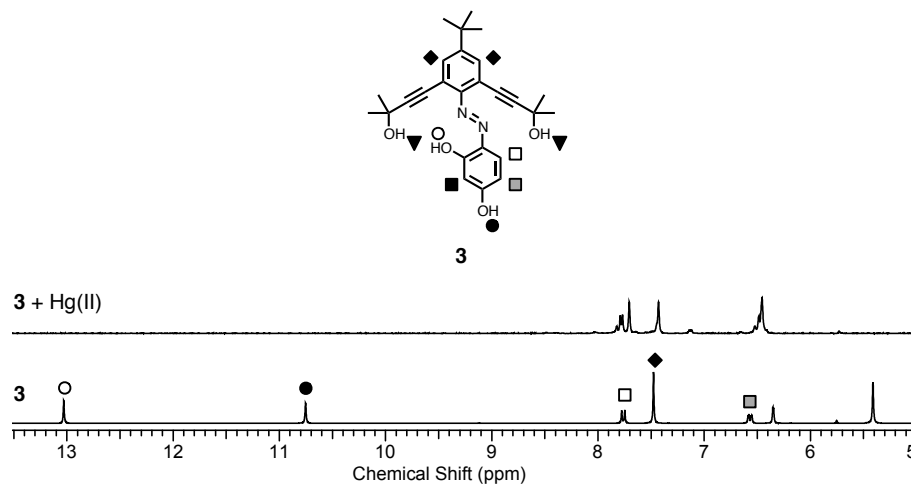


Fig. S6. Partial ^1H NMR spectra of **3** prior to (bottom) and after (top) exposure to Hg(II) in $\text{DMSO-}d_6$ at $T = 298$ K.

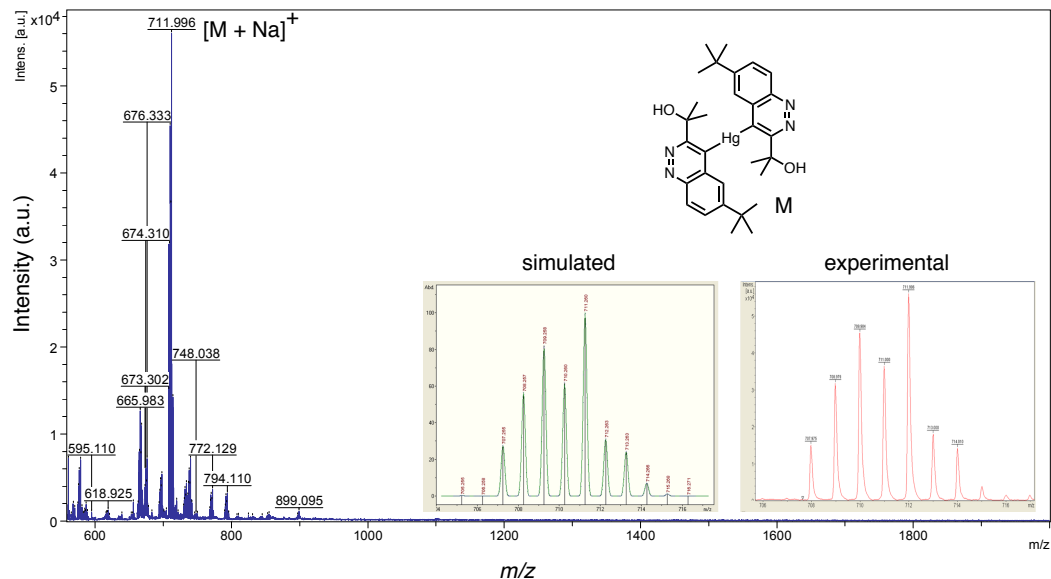


Fig. S7. MALDI-TOF mass spectrum of the intramolecular cyclization product (calcd for $C_{30}H_{38}HgN_4NaO_2$ $[M + Na]^+$, 711.260, found, 711.996). Chemical structure (top), along with simulated (bottom left) and experimental (bottom right) isotope patterns, are also provided.

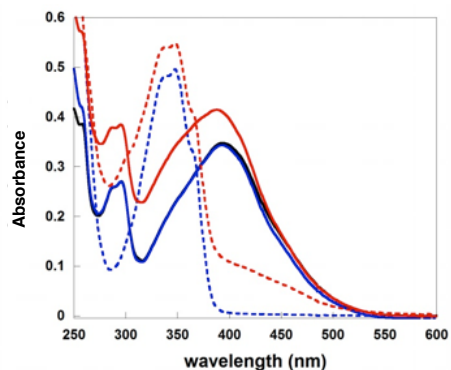


Fig. S8. UV-vis spectra of **3** ($20 \mu M$, black solid line), **3** + Pb(II) (5 equiv, blue solid line), and **3** + Fe(III) (5 equiv, red solid line) in MeOH. Spectral changes after addition of Hg(II) (5 equiv) are shown for **3** + Pb(II) + Hg(II) (blue dotted line) and **3** + Fe(III) + Hg(II) (red dotted line). $T = 298$ K.

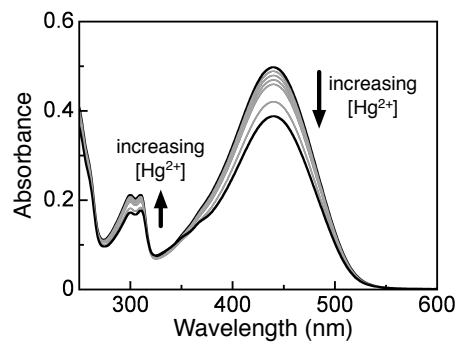


Fig. S9. Changes in the UV-vis spectra of **3** ($XX \mu\text{M}$) upon addition of $[\text{Hg}^{2+}] = 0, 50, 100, 150, 200, 400,$ and 600 nM at $\text{pH} = 7.4$ ($[\text{HEPES}] = 10 \text{ mM}; 100 \text{ mM NaCl}$), $T = 298 \text{ K}$.