NBD-based Fluorescent Chemosensor for the Selective Quantification of Copper and Sulfide in Aqueous Solution and Living Cells

Qingtao Meng\textsuperscript{a}, Yu Shi\textsuperscript{b}, Cuiping Wang\textsuperscript{a}, Hongmin Jia\textsuperscript{a}, Xue Gao\textsuperscript{a}, Run Zhang\textsuperscript{b}\textsuperscript{*}, Yongfei Wang\textsuperscript{a} and Zhiqiang Zhang\textsuperscript{a}\textsuperscript{*}

\textsuperscript{a} School of Chemical Engineering, University of Science and Technology Liaoning, Anshan, 114044, China

E-mail: zhangzhiqiang@ustl.edu.cn; Tel: +86-421-5928009

\textsuperscript{b} Department of Chemistry and Biomolecular Sciences Faculty of Science, Macquarie University, Sydney NSW, 2109, Australia

E-mail: run.zhang@mq.edu.au. Tel: +61 (2) 9850 1175
Fig. S1 $^1$H-NMR of NL (DMSO-$d_6$)

Fig. S2 $^{13}$C-NMR of NL (DMSO-$d_6$)
Fig. S3 High resolution TOF-MS of NL

Fig. S4 Time-dependent fluorescence changes of NL in HEPES aqueous buffer (THF: H₂O = 3:7, 20 mM, pH = 7.4). Excitation at 430 nm.
**Fig. S5** Variations of fluorescence intensity at 519 nm of NL (10 μM) in aqueous solution with (bottom) and without (up) Cu$^{2+}$ (0–20 μM) as a function of pH. Excitation at 430 nm.

**Fig. S6** Absorption spectra of NL (10 μM) in HEPES aqueous buffer (THF: H$_2$O = 3:7, 20 mM, pH = 7.4) upon addition of various metal ions (20 μM).
Fig. S7. The Job's plot of NL toward Cu^{2+}.

$$x_{Cu}^{2+} = \frac{[Cu^{2+}]}{[Cu^{2+} + NL]}$$

Fig. S8 Linear relationship between fluorescence intensity of NL (1 μM) at 519 nm versus the concentration of Cu^{2+} in HEPES buffer (THF: H₂O = 3:7, 20 mM, pH = 7.4). Excitation at 430 nm.

$$Y = 1.5148 + 589.0325X$$

$$R^2 = 0.9964$$
**Fig. S9** UV-vis spectra of NL (10 μM) sequential in the presence of Cu^{2+} (20 μM) and S^{2−} (20 μM) in HEPES aqueous buffer (THF: H₂O = 3:7, 20 mM, pH = 7.4).

**Figure S10** ESI-mass spectra of NL-Cu^{2+} ensemble

**Figure S11** ESI-mass spectra of NL-Cu^{2+} ensemble in the presence of S^{2−}.
**Fig. S12** Flow cytometric analysis of MDA-MB-231 cells stained with **NL** and its fluorescent response to Cu²⁺ and S²⁻. (a) Control group: MDA-MB-231 cells only, (b) cells stained with 10 μM **NL** for 20 min, (c) **NL** stained cells treated with 20 μM Cu²⁺ for 15 min, (d) then incubated with 40 μM S²⁻ for another 30 min.