Electronic Supplementary Material (ESI) for Organic & Biomolecular Chemistry

Supporting Information for

Thiol-inducible Direct Fluorescence Monitoring of Drug Delivery

Jun Wu,[a‡] Rong Huang,[a‡] Changcheng Wang,[a] Wenting Liu,[a] Jiaqi Wang,[a] Xiaocheng Weng,[a] Tian Tian,[a] Xiang Zhou [ab]*

College of Chemistry and Molecular Sciences, Key Laboratory of Biomedical Polymers of Ministry of Education, State Key Laboratory of Virology, Wuhan University, Hubei, Wuhan, 430072, P. R. of China, Corresponding Author Email Address: xzhou@whu.edu.cn

Apparatus

Fluorescent emission spectra were collected from 460-650 nm on PerkinElmer LS 55 with an excitation wavelength of 420 nm, the excitation and emission slit widths were 15 nm and 9 nm, respectively. Quartz cuvettes with 2 mL volume used for emission measurements. UV-Vis absorption spectra were collected on SHIMADZU UV-2550 from 350-600nm with 600 μL quartz cuvettes. Unless otherwise specified, all spectra were taken at 37 ℃ in 10mM sodium phosphate buffers. All pH measurements were performed with a pB-10 pH-meter (Sartorius, Shanghai, China) with a combined glass-calomel electrode. ^{1}H and ^{13}C NMR spectra were recorded on Varian Mercury 300 spectrometers, respectively. HRMS were recorded on a Brucker APEX IV (7.0 T). Florescent images were acquired on Nikon Confocal Laser Scanning Microscope (TE2000, Japan) with an oil objective lens (×60). Images and merges were obtained with EZ-C1 software.

Materials

All solvents and reagents were commercially available and used without further purification unless for special needs: MEM (HyClone, Thermo Scientific), fetal bovine serum (FBS, HyClone), penicillin and streptomycin (Invitrogen), MTT (Sigma), γ-H2AX (phosphor S139) rabbit polyclonal (ab11174, Abcam), Hoechst-33258 (Calbiochem) and propidium iodide (Sigma-Aldrich). HeLa cells were purchased from China Center for Type Culture Collection.

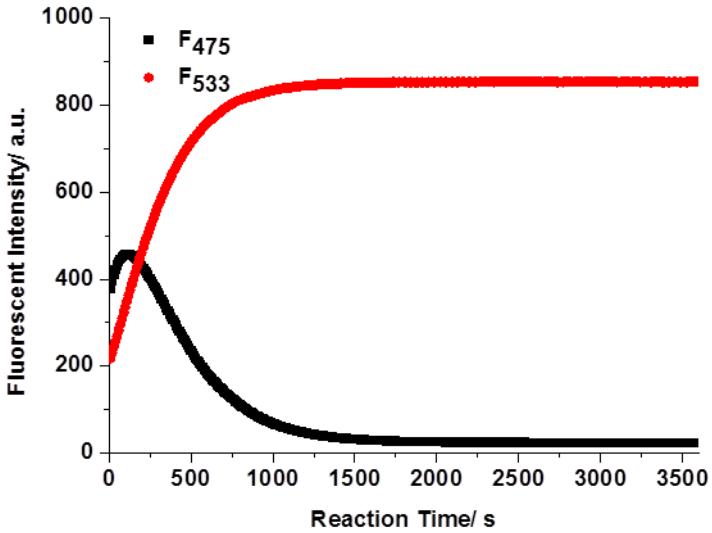


Figure S1. Fluorescence intensity changes at 475 nm (F475) and 533 nm (F533) recorded as a function of time. Monitored time is 1 h. The concentrations of NADH-CLB and DTT were 10 μM and 5.0 mM, respectively. Excitation wavelength= 420 nm.

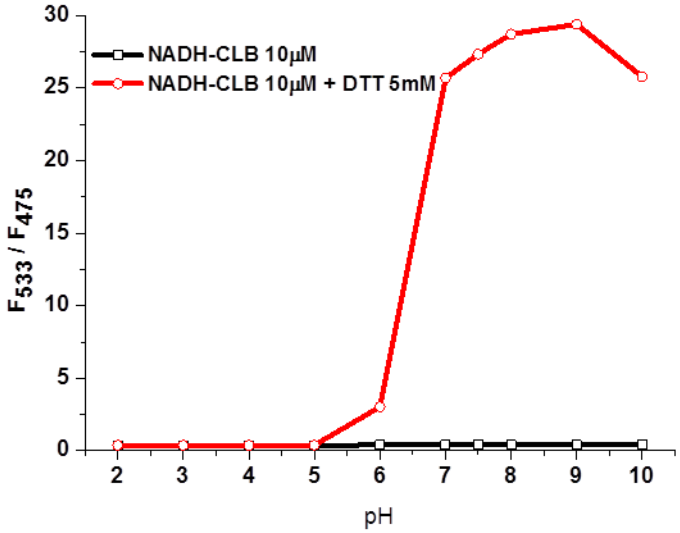


Figure S2. The ratio of fluorescence intensity at 533 nm to that at 475 nm (F_{533}/F_{475}) of NADH-CLB (10 μM) with and without DTT (5.0 mM) as a function of pH. Sodium phosphate buffers with different pH value ranging from 2 to 10 were used at the concentration of 10 mM.

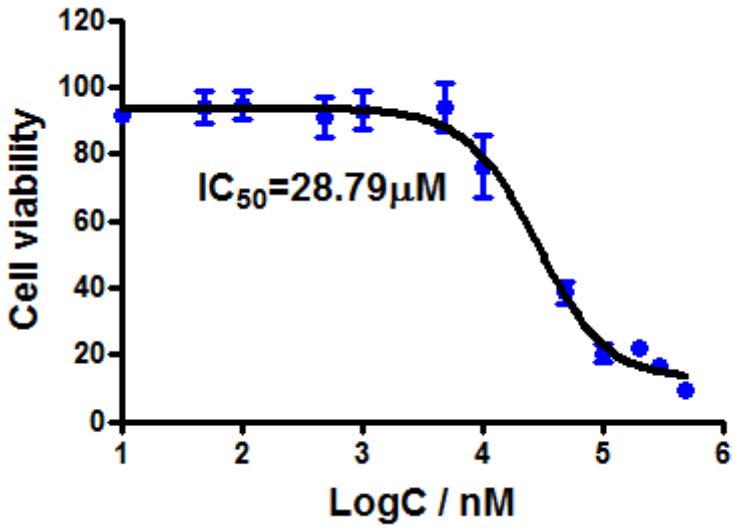
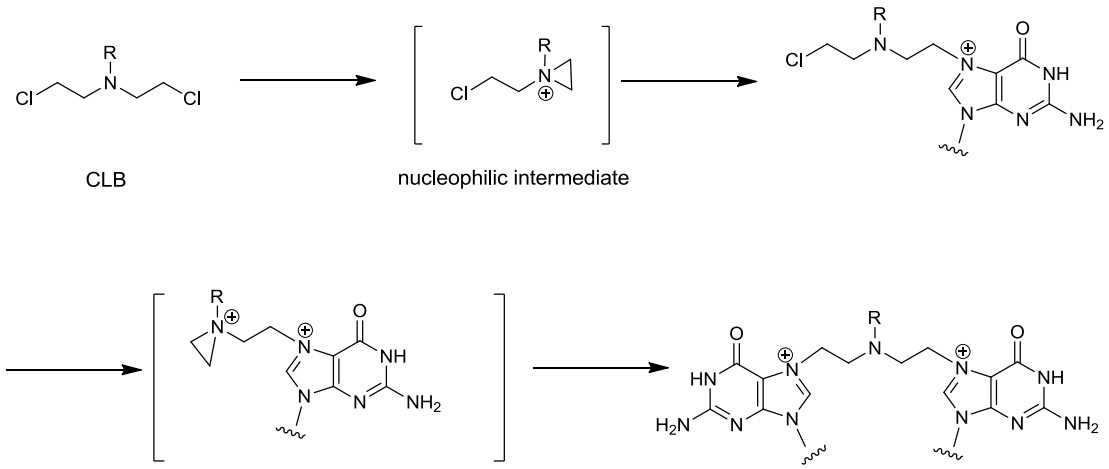


Figure S3. Cell viability in the presence of NADH-CLB at different concentrations (10 nM- 500 μM). The data were obtained through MTT assay and presented as mean± SD (n= 3).



Scheme S1. Mechanism of DNA damage by alkylating agent

[1]A. Gilman, F. S. Philips, The biological actions and theraputic applications of β-chloroethyl am ines and sulfides, *Science*, 1946, 103-409.

[2] M. T. McClure, I. Stupans, Investigation of the Mechanism by which Cyclophosphamide Alters Cytochrome P450 in Male Rats, *Biochem. Pharmacol*, 1992, **43**, 2655-2658.