Electronic Supplementary Information for

Colorimetric and fluorescent dual probe based on a polythiophene derivative for the detection of cysteine and homocysteine

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Materials. All chemicals were purchased from Aldrich and Beijing Chem. Reagents Co. (Beijing, China) and used as received. Water-soluble polythiophene derivative, PMTPA, was synthesized and purified as reported previously (C. Li, M. Numata, A.-H. Bae, K. Sakurai and S. Shinkai, *J. Am. Chem. Soc*., 2005, **127**, 4548).

Sample Preparation. The *in situ* derived reaction of Cys as well as Hcy and CBT was performed as the following procedures: stock solutions of Cys (or Hcy) and CBT were mixed together to give a mixture with the given concentrations in PBS buffer containing 5% (v/v) DMSO. The formation of CBT-Cys conjugate, a luciferin derivative, can be monitored by CD spectra at 300 nm.

Control experiments for addressing the specificity of PMTPA toward Cys and Hcy were carried out under the identical conditions. The 20 natural amino acids, Hcy, Cyt, GC and GSH were pre-reacted with CBT for 30 min and then the PMTPA was added into CBT/analyte mixture to give a solution containing 0.1 mM PMTPA, 0.25 mM CBT and 0.25 mM analyte. After 5 min, the sample was measured by UV-visible spectrometer.

For ^{1}H-NMR measurements, 8.0 mM CBT and 5.0 mM Cys were pre-reacted for 30 min in DMSO-d_6/D_2O (4/1; v/v), and then the reacted mixture was applied to NMR measurement.

Measurements. Absorption, emission and CD spectra were collected by using a Hitachi 3010 UV-visible spectrometer, a LS55 fluorescence spectrometer (PerkinElmer) and a Jasco J-815 spectropolarimeter, respectively. ^{1}H-NMR spectra were carried out on a JNM-ECA300 spectrometer (JEOL).

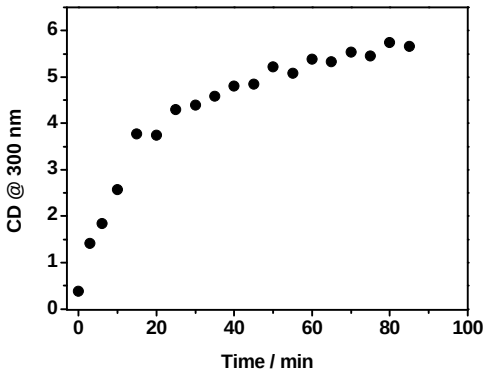


Fig. S1 Time course of the ICD intensity of a mixture of CBT-Cys at 300 nm in PBS buffer containing 5% DMSO. [CBT] = [Cys] =0.25 mM.

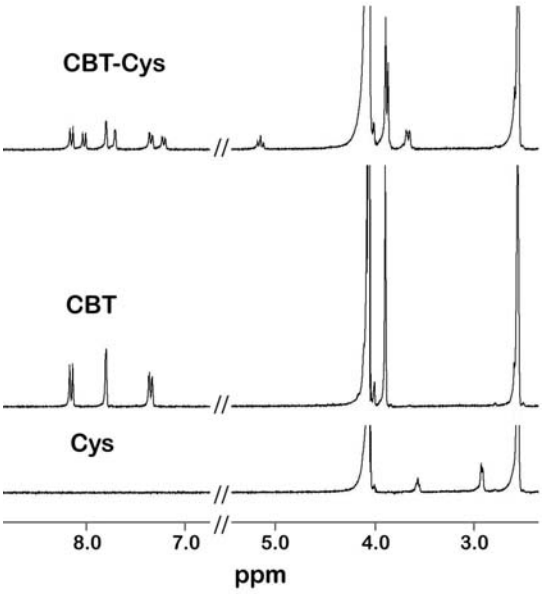
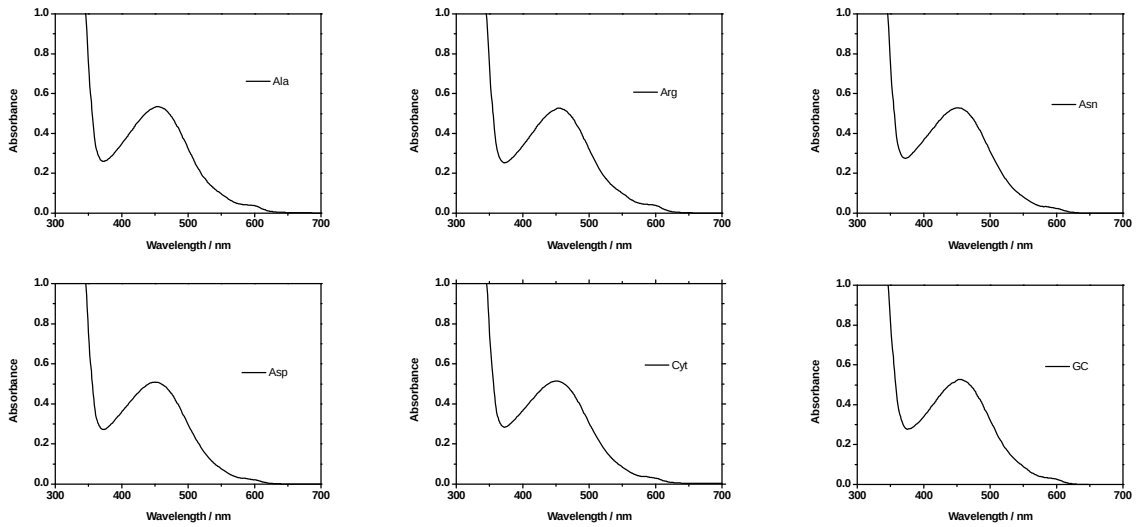


Fig. S2 ^{1}H-NMR spectra of Cys, CBT and a mixture of CBT and Cys pre-reacted for 30 min in DMSO-d_6/D_2O (4/1; v/v). [CBT] = 8.0 mM; [Cys] = 5.0 mM.



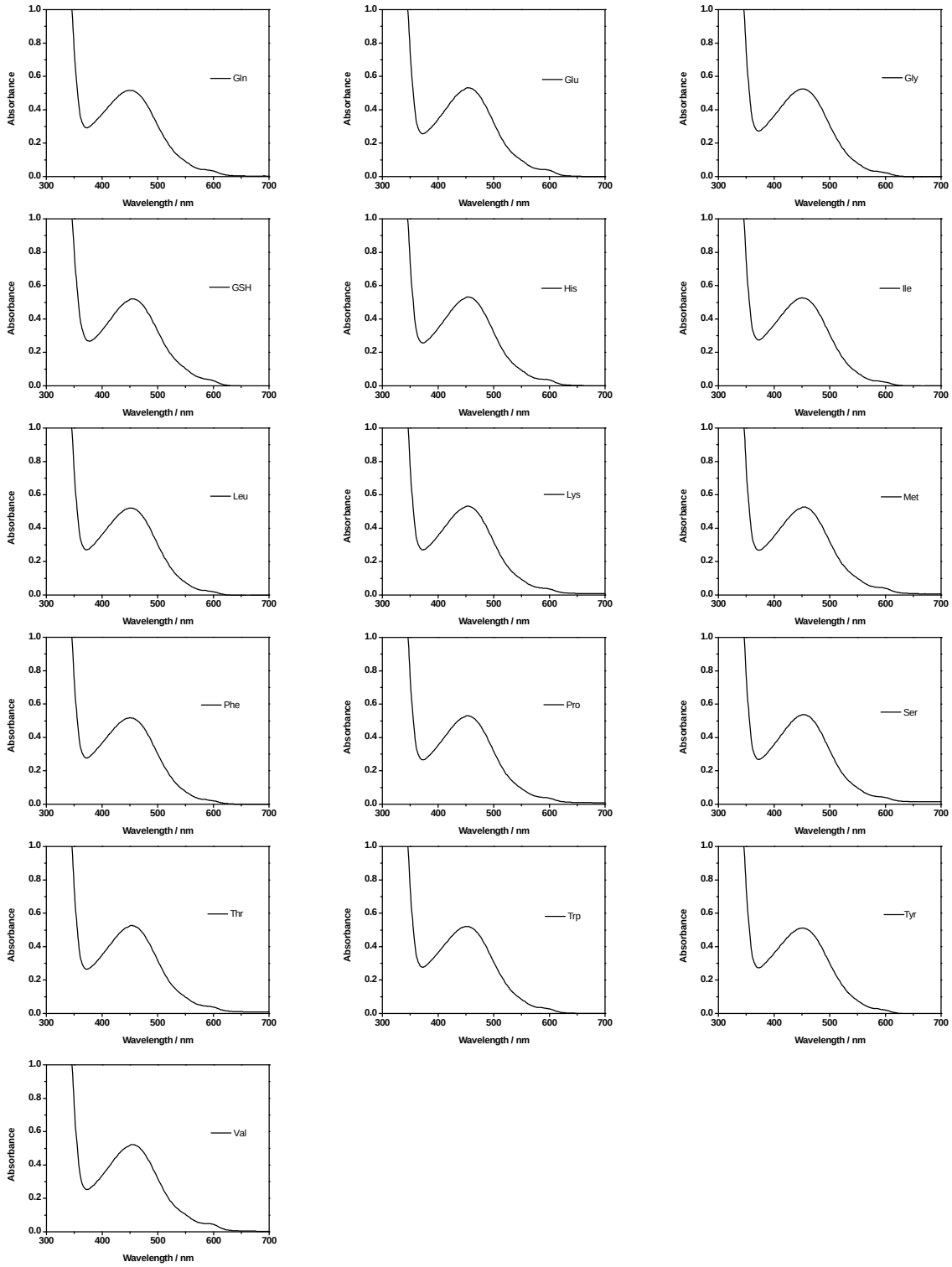


Fig. S3 Absorption spectra of PMTPA (0.1 mM) in the presence of CBT and 19 natural amino acids, Cyt, GC and GSH in 10 mM PBS buffer (pH 7.4) containing 5% DMSO. [CBT] = 0.25 mM, [Analyte] = 0.50 mM

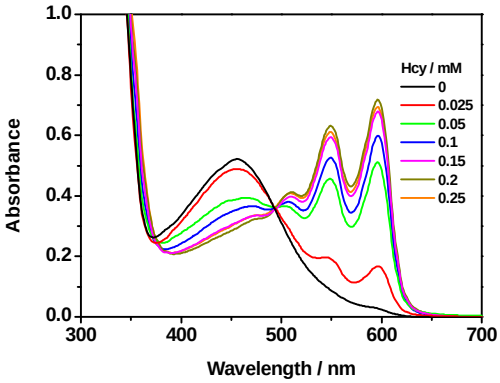


Fig. S4 Variation in the absorption spectra of PMTPA (0.1 mM) in PBS buffer containing 5% DMSO with increasing concentrations of Hcy as indicated. [CBT] = 0.25 mM.

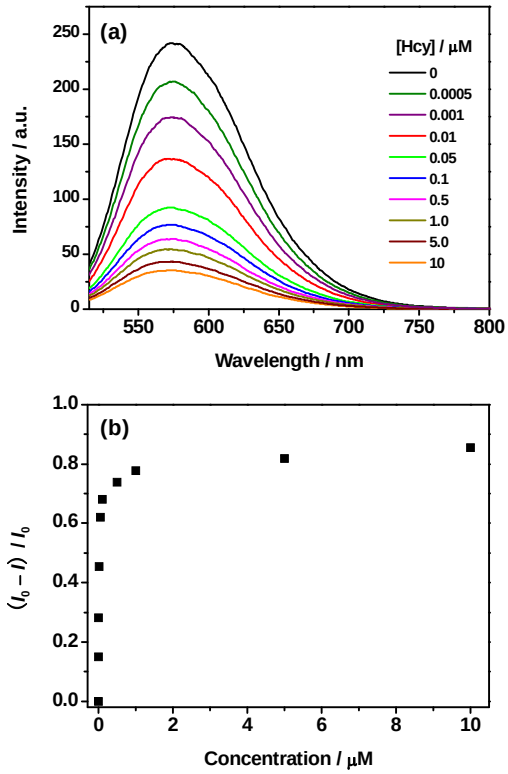


Fig. S5 (a) Variation in the emission spectra of PMTPA (0.01 mM) in PBS buffer containing 5% DMSO with increasing concentrations of CBT-Hcy as indicated. [CBT] = 2[Hcy]. (b) Fluorescence quenching of PMTPA by CBT-Hcy at various concentrations. I_0 is the fluorescence intensity at 575 nm of a solution of PMTPA; I is the fluorescence intensity at 575 nm of a solution of PMTPA in the presence of different amounts of CBT-Hcy. Excitation wavelength: 495 nm.