Electronic Supplementary Material (ESI) for Analyst

Supporting Information

Real-time Fluorometric Turn-On Assay for Protease Activity and Inhibitor Screening with a Benzoperylene Probe

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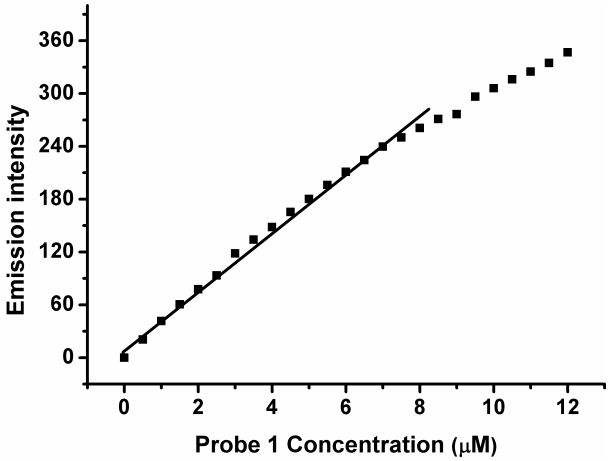

Figure S1. Emission intensity changes at 419 nm with probe **1** concentration.

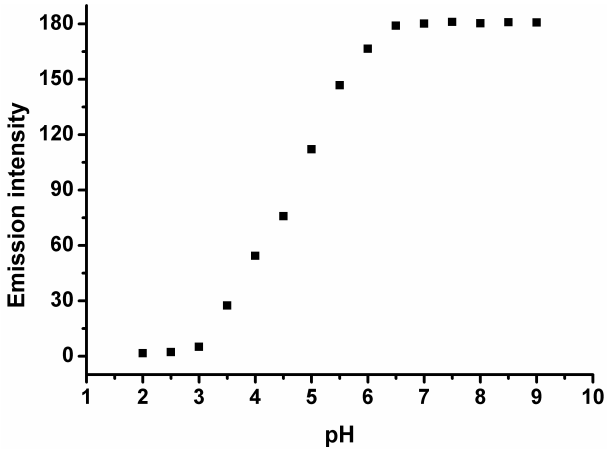

Figure S2. Plot of the changes in emission intensity of probe **1** (5 μM) at 419 nm at different buffer pH values. Conditions: 5 mM Glycine-HCl, pH 2.0 – 3.5; 5 mM NaAc-HAc, pH 4.0 – 5.5; 5 mM Na_2HPO_4-NaH_2PO_4, pH 6.0 – 8.0; 5 mM Glycine-NaOH, pH 8.5 – 9.0.

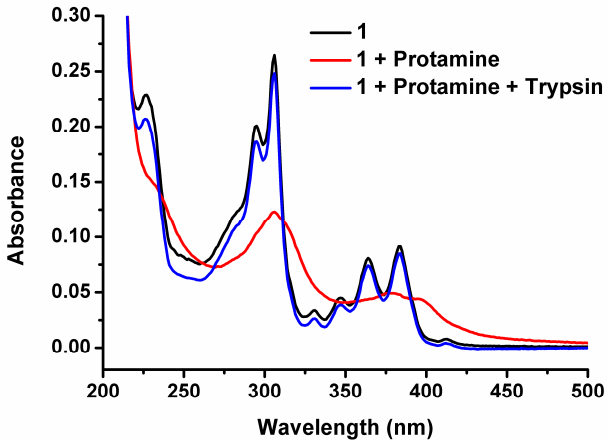

Figure S3. The absorption spectra of 5 μM probe **1** (black), probe **1** mixed with 0.5 μM protamine (red), and the probe **1**/protamine mixture incubated with 240 mU/mL trypsin for 30 min (blue).

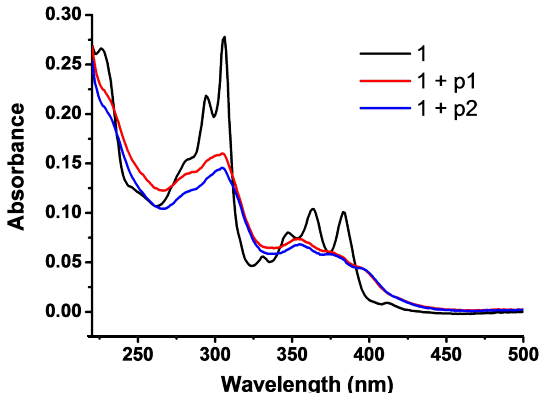

Figure S4. The absorption spectra of 5 μM probe **1** (black), probe **1** mixed with 10 μM poly(diallyldimethylammonium chloride) (**p1**, red) and 10 μM poly(allylamine) (**p2**, blue).

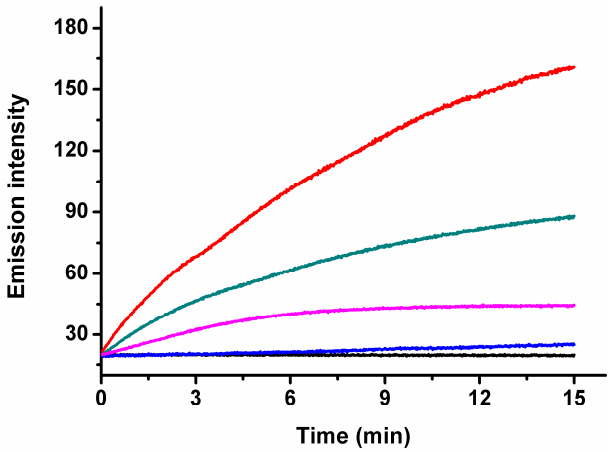

Figure S5. Real-time emission intensity changes of probe **1** (5 μM) at 419 nm upon the addition of other proteases (from up to bottom: 240 mU/mL trypsin, 10 mU/mL chymotrypsin, 2 mU/mL carboxypeptidase B, 1 U/mL protease K, and no protease). Protamine concentration: 0.5 μM.

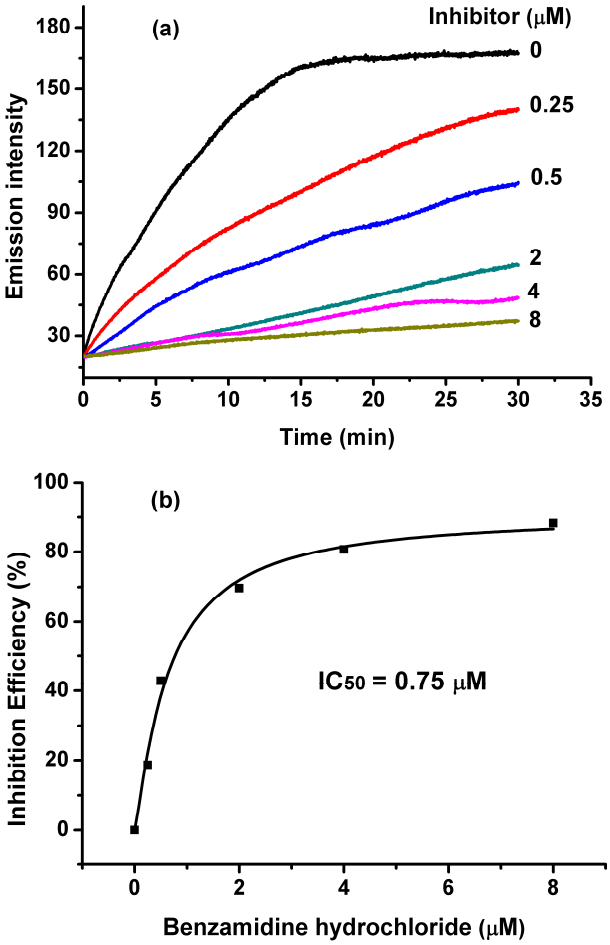

Figure S6. (a) Real-time emission intensity changes of probe **1** (5 μM) at 419 nm at different inhibitor concentrations. Assay solution contained 0.5 μM protamine, 240 mU/mL trypsin, and different concentrations of benzamidine hydrochloride. (b) Plot of the inhibition efficiency versus benzamidine hydrochloride concentration. Assay solutions contained 5 μM probe **1**, 0.5 μM protamine, 240 mU/mL trypsin, and different amounts of benzamidine hydrochloride (0, 0.25, 0.5, 2 and 4 μM, respectively).