A tryptophan-containing fluoroionophore sensor with high sensitivity to and selectivity for lead ion in water

(Supporting Information)

Li-Jun Ma, Yi-Fu Liu , Yuqing Wu *

Experimental

1) Synthesis and Characterization

Synthesis and Characterization of N-[4(1-pyrene) butyroyl]-L-tryptophan (PLT)[1]

L-tryptophan methyl ester (0.075 g, 0.345 mmol) was dissolved in anhydrous tetrahydrofuran (20 ml) and mixed with 4-(1-pyreny) butyric acid (0.1 g, 0.345 mmol) and dicyclohexylcarbodiimide (0.079 g, 0.383 mmol). The mixture was stirred 12h at room temperature and monitored by TLC (20:1, chloroform to methanol). After excess DCC was decomposed, the solvent was removed under reduced pressure. Purification of **PLT**-ester was performed by using column chromatography on silica gel (elution with 20:1, chloroform to methanol; R_f = 0.3). Mild hydrolysis of the **PLT**-ester (1 M HCI, 12 h, room temperature) followed by column chromatography on silica gel (elution with 5:1, chloroform to methanol; R_f = 0.5) resulted in a **PLT** in 40 % yield. The product was an aqua viscous solid. ^{1}H NMR (500 MHz, d_6-DMSO): δ = 8.344-7.844 (m, 9 H, Py (**H**)), δ = 7.519-6.848 (m, 5 H, Indole (**H**)), δ = 4.257 (m, 1 H, α-C**H**), δ = 3.231, 3.012 (m, 4 H, PyC**H**$_2$+C**H**$_2$Ar), δ = 2.215 (m, 2 H, C**H**$_2$CONH), δ = 1.930 ppm (m, 2 H, $CH_2C\mathbf{H}_2CH_2$). Mass spectral data (MALDI-TOF-MS): for $C_{31}H_{26}N_2O_3$ calcd, 474.6; found 475.0. Specific optical rotation of **PLT**: $[\alpha]_{589}^{22}$ -21.3º (c 0.01, THF).

[1] C. V. Kumar, A. Buranaprapuk and H. C. Sze, *Chem.Commum.* 2001, 297.

Synthesis and Characterization of N-[4(1-pyrene) butyroyl]-L-phenylalanine (PLP)

The **PLP** was synthesized by the reaction of the methyl ester of L-phenylalanine with 4(1-pyrene)butyric acid in tetrahydrofuran using DCC as the coupling agent in a procedure analogous to the synthesis of the **PLP** in ref. 1. ^{1}H NMR (500 MHz, d_6-DMSO): δ = 8.32-7.86 (m, 9 H, Py (**H**)), δ = 7.21-7.07 (m, 5 H, phenyl (**H**)), δ = 4.27 (m, 1 H, α-C**H**), δ = 3.21-2.86 (m, 4 H, PyC**H**$_2$+C**H**$_2$Ar), δ = 2.22 (m, 2 H, C**H**$_2$CONH), δ = 1.93 ppm (m, 2 H, $CH_2C\mathbf{H}_2CH_2$). Mass spectral data (MALDI-TOF-MS): for $C_{29}H_{25}NO_3$ calcd, 435.5; found 435.1. Specific optical rotation of **PLP**: $[\alpha]_{589}^{22}$ -32.9º (c 0.003, THF).

2) UV-vis Absorption Spectra

The measurements of UV-vis absorption spectra were carried out with a Lambda 800 Spectrophotometer.

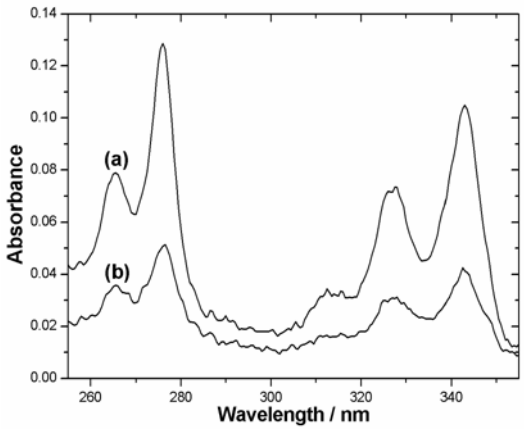


Fig. 1 UV-vis absorption spectra of **PL**T (5×10^{-6} mol L^{-1} in 98% water/2% DMSO (v/v)) in the (a) absence and (b) presence of Pb^{2+} (8.2×10^{-6} mol L^{-1}).

3) Fluorescence titration experiments

Fluorescence emission spectra were recorded on a computer-controlled SHIMADZU (Japan) RF-25301PC fluorescence spectrophotometer. A fixed maximum excitation wavelength at 340 nm was used. Fluorescence titration was performed in aqueous solution using respective chloride salt of metal ion. The concentration of **PLT** in all the fluorescent experiment is 5×10^{-6} mol L^{-1} in 98% water/2% DMSO (v/v).

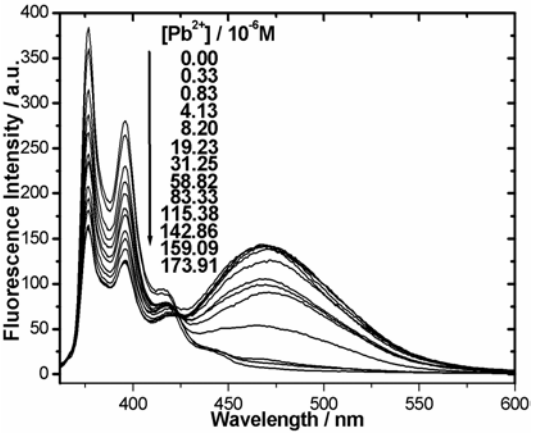


Fig. 2a Fluorescence spectra of **PLT** with the addition of Pb^{2+}.

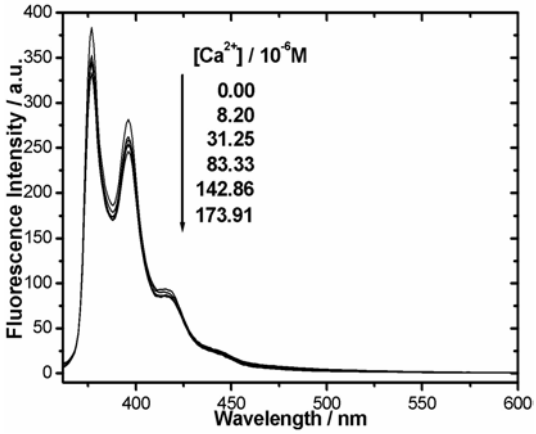


Fig. 2b Fluorescence spectra of **PLT** with the addition of Ca^{2+}.

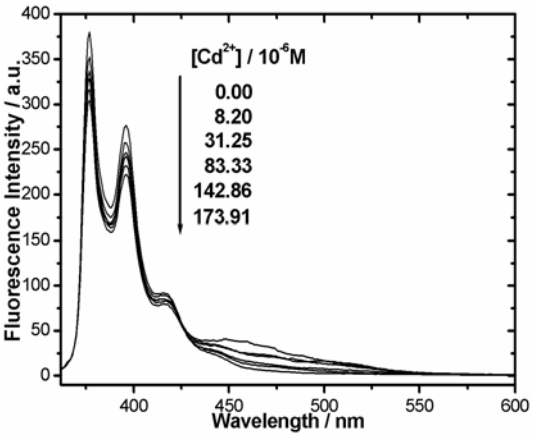


Fig. 2c Fluorescence spectra of **PLT** with the addition of Cd^{2+}.

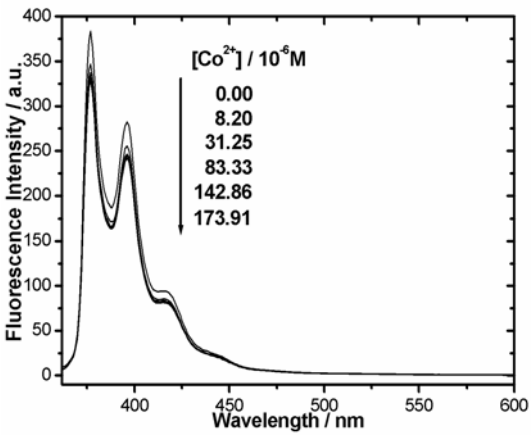


Fig. 2d Fluorescence spectra of **PLT** with the addition of Co^{2+}.

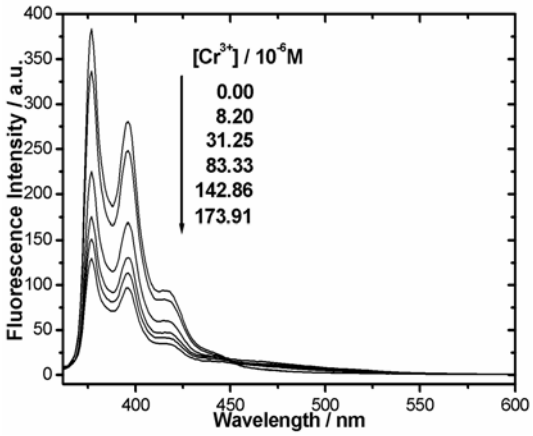


Fig. 2e Fluorescence spectra of **PLT** with the addition of Cr^{3+}.

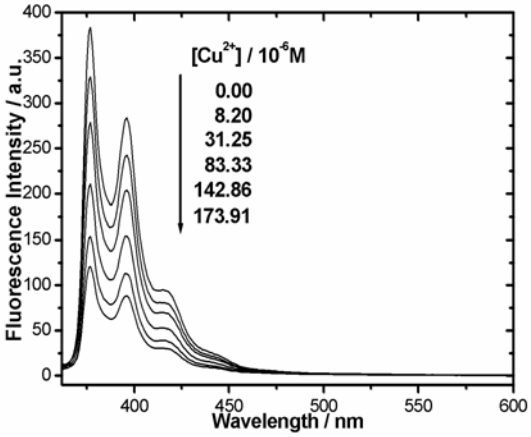


Fig. 2f Fluorescence spectra of **PLT** with the addition of Cu^{2+}.

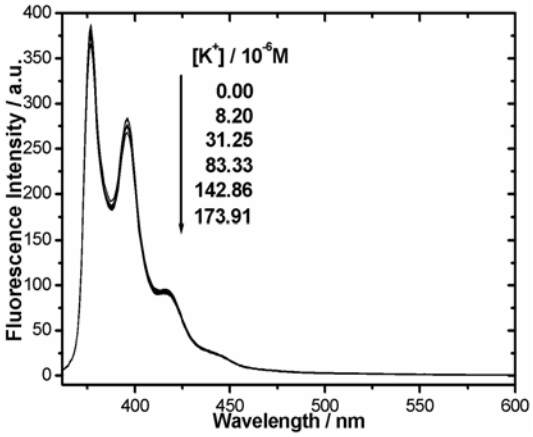


Fig. 2g Fluorescence spectra of **PLT** with the addition of K^{+}.

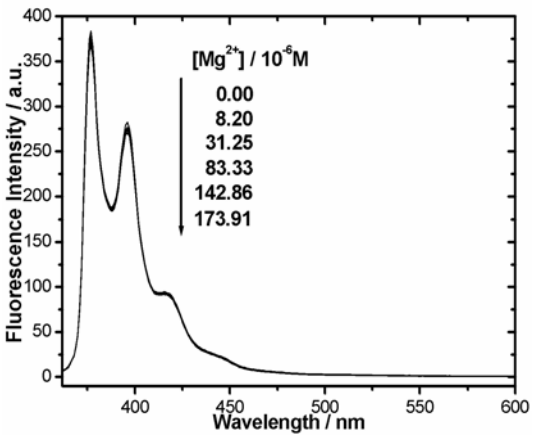


Fig. 2h Fluorescence spectra of **PLT** with the addition of Mg^{2+}.

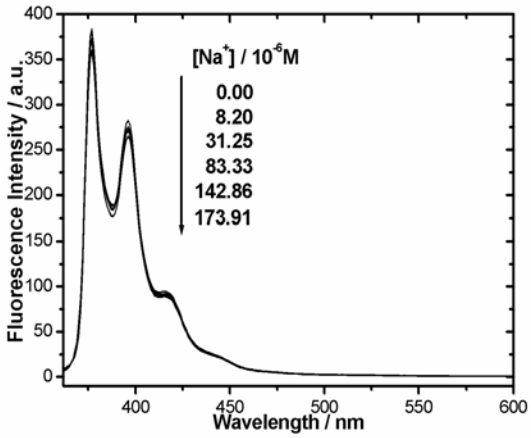


Fig. 2i Fluorescence spectra of **PLT** with the addition of Na^{+}.

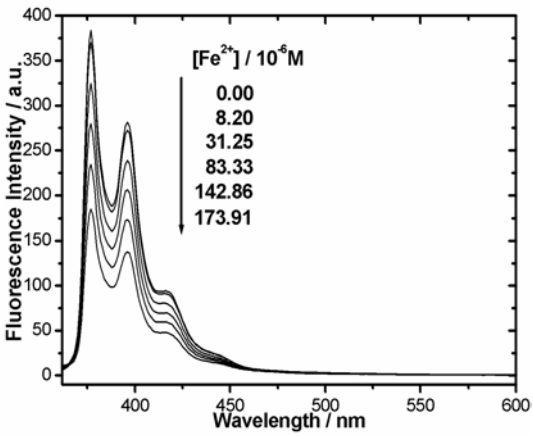


Fig. 2j Fluorescence spectra of **PLT** with the addition of Fe^{2+}.

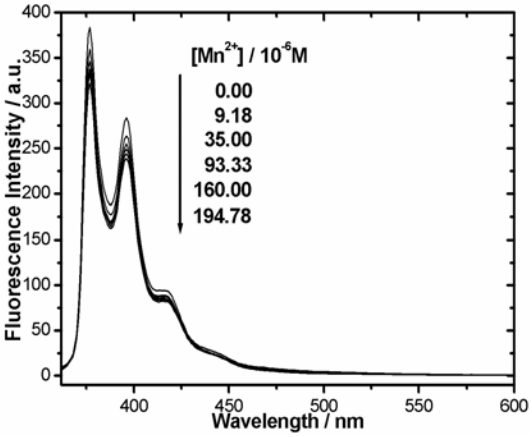


Fig. 2k Fluorescence spectra of **PLT** with the addition of Mn^{2+}.

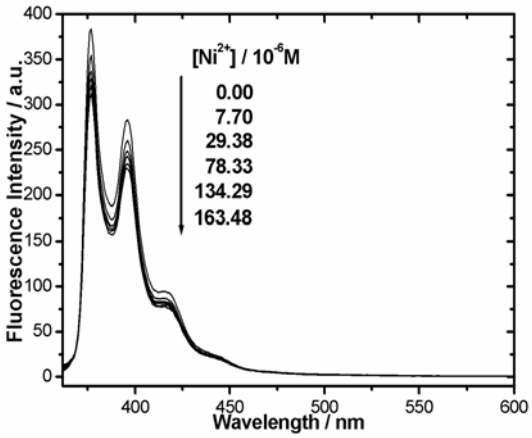


Fig. 2l Fluorescence spectra of **PLT** with the addition of Ni^{2+}.

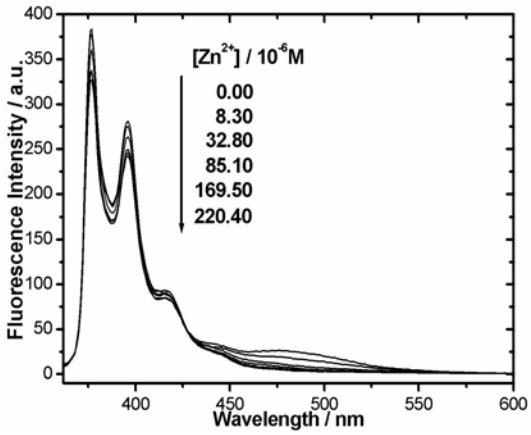


Fig. 2m Fluorescence spectra of **PLT** with the addition of Zn^{2+}.

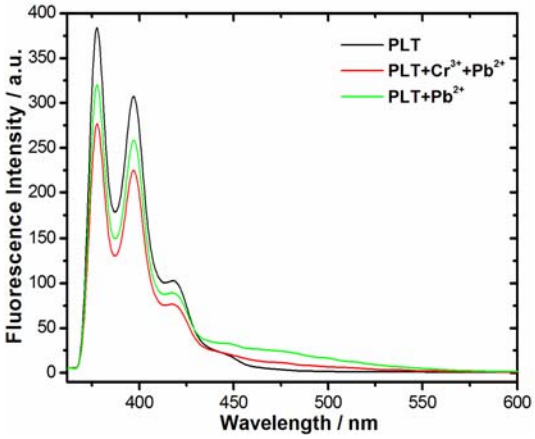


Fig. 2n Fluorescence spectra of **PLT**, in the presence of Pb^{2+} (1.3 μM), and the mixture of Pb^{2+} (1.3 μM) and Cr^{3+} (10 μM).

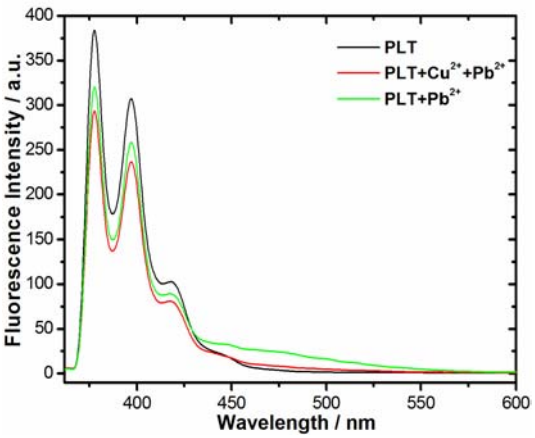


Fig. 2o Fluorescence spectra of **PLT**, in the presence of Pb^{2+} (1.3 μM), and the mixture of Pb^{2+} (1.3 μM) and Cu^{2+} (10 μM).

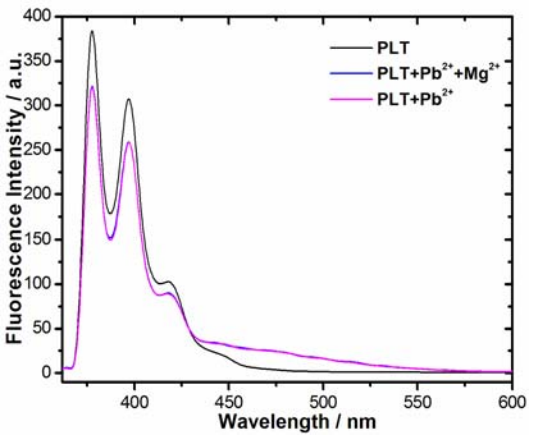


Fig. 2p Fluorescence spectra of **PLT,** in the presence of Pb^{2+} (1.3 μM), and the mixture of Pb^{2+} (1.3 μM) and Mg^{2+} (10 μM). The interference of other kinds of metal ions as Ca^{2+}, Cd^{2+}, Co^{2+}, K^{+}, Na^{+}, Fe^{2+}, Mn^{2+}, Ni^{2+}, and Zn^{2+} on Pb^{2+} are similar with Mg^{2+} (results are not shown).

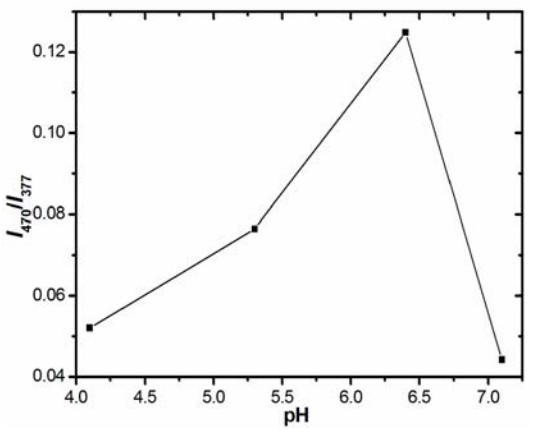


Fig. 2q pH-dependence of the fluorescent response of the PLT/Pb^{2+} (the concentration of Pb^{2+} is 1.6 μM).

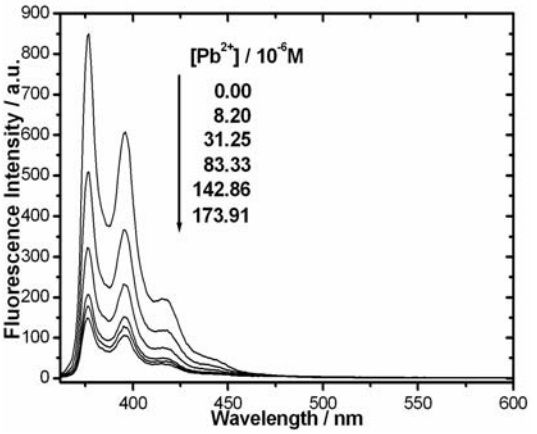
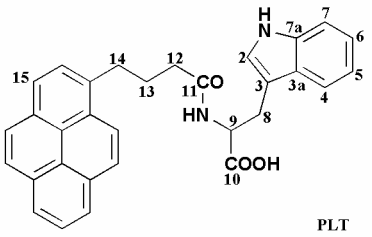


Fig. 2r Fluorescence spectra of **PLP** (5 μM) with the addition of Pb^{2+}.

4) NMR Spectra of PLT with Pb^{2+}

Ten equivalent of $PbCl_2$ was added to a solution of 5.5×10^{-3} mol L^{-1} **PLT**, which was prepared by dissolving **PLT** in D_2O/DMSO-d_6 (1:2, v/v). ^1H NMR and ^{13}C NMR were measured on a Bruker AM-500 spectrometer. All the spectra were recorded at room temperature.

Scheme 1. The chemical structure and atom numbering of **PLT**



PLT

Table 1. Proton chemical shifts[a] (in ppm) of the **PLT** and its associated complex with Pb^{2+}; atomic numbering is shown in **Scheme 1.**

	H(4)	**H**(7)	**H**(2)	**H**(5)	**H**(6)	α-C**H**
PLT	7.551 d,	7.271 d,	7.156 s	6.950 t	6.855 t	4.448 m
PLT+Pb^{2+}	7.585 d,	7.310 d,	7.219 s	7.014 t	6.939 t	4.514 f

	PyCH_2	CH_2Ar	CH_2CONH	$CH_2CH_2CH_2$	Py(**H**)*
PLT	3.261 f	3.032 m	2.239 m	1.890 m	8.295 d, 8.250 d, 8.183 t 8.144 m, 8.095 t, 7.810 d
PLT+Pb^{2+}	3.296 f	3.069 m	2.264 m	1.905 m	8.294 d, 8.247 d, 8.185 t 8.143 m, 8.094 t, 7.818 d

* Of note is that there is no much chemical shifts were observed for pyrene protons upon the addition of Pb^{2+}, which may due to the high-concentration-induced random aggregation of PLT before the Pb^{2+} addition.

Table 2. Carbon-13 chemical shifts[a] (in ppm) for the sensor **PLT** and its complex with Pb^{2+}; Atomic numbering is shown in **Scheme 1.**

	C(7a)	C(3a)[b]	C(2)[b]	C(5)	C(6)	C(4)	C(7)	C(3)
PLT	138.369	130.213	125.816	123.557	121.040	120.997	113.866	113.351
PLT+Pb^{2+}	138.467	129.885	125.987	123.760	121.234	120.835	114.020	113.033

	C(10)	C(11)	C(9)	C(12)	C(8)	C(14)	C(13)	C(15)
PLT	183.901	175.457	58.672	37.723	34.463	31.897	30.007	139.115, 133.480, 133.015, 131.877, 130.654, 130.122 t 129.171d,127.593d, 126.765d
PLT+Pb^{2+}	c	175.873	57.326	37.923	34.433	29.755	30.058	139.063, 133.457, 132.979 132.875, 130.636, 130.102 t 129.185d,127.590d, 126.758d

a. The ^{1}H and ^{13}C NMR assignment was made on the basis of HMQC (Heteronuclear Multiple Quantum Correlation) experiments and literature data (refs 7 in text); b. the peaks existed a part of overlap; c. no noted (it is due to intensely broadening upon Pb^{2+} binding of the sensor).