Supporting Information for

Selective detection of zwitterionic arginine with a new Zn (II)-terpyridine complex: potential application in protein labeling and determination

*Xin Zhou, Xuejun Jin, Donghao Li, Xue Wu**

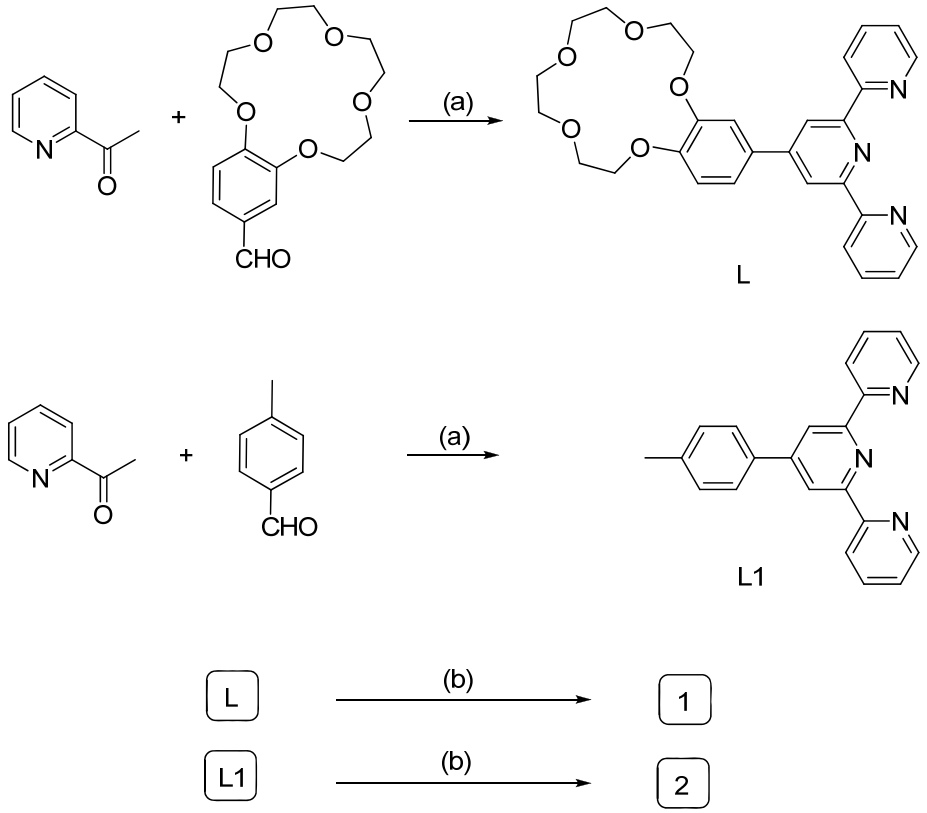
Key Laboratory of Nature Resource and Functional Molecular of the Changbai Mountain, Ministry of Education, Yanbian University, Yanji, Jilin, China, 133002

E-mail: wuxue@ybu.edu.cn

Experimental

General Methods

Commercially available chemicals were used without further purification unless stated otherwise. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker AV-300 (300 MHz) and 75 (75 MHz) spectrometer in $CDCl_3$ with TMS as the reference. UV-vis spectra were recorded on Perkin Elmer Lambda 35 UV-vis spectra and PL spectra were conducted on Fluorescence Spectrophotometer (RF-540). MALDI-TOF mass spectra were recorded on a Shimadzu MALDI AXIMA-CFR+ Spectrometer.



Scheme. S1 Synthesis routes of receptors; conditions: (a) $NH_3{\cdot}H_2O$; KOH; EtOH; rt; (b) Zn $(ClO_4)_2$; ACN; rf

4'-B15C5-2, 2':6', 2"-terpyridine (Ligand L). 2-Acetylpyridine (240 mg, 2 m mol) was added into a solution of 4-formylbenzo-15-crown-5 (200 mg, 0.7 m mol) in EtOH (30 mL). KOH pellets (300 mg, 4 m mol) and NH_3-H_2O (10 mL, 29.3%) was added to the solution. The solution was stirred at 34 °C for 3 days. The mixture was cold to 20 °C, and then the off-white precipitate was collected by filtration and washed with ice-cold EtOH (10 mL). Recrystallization from EtOH obtained white needle solid, yield: 41%. ^{1}H NMR (300 MHz, $CDCl_3$), δ (ppm): 8.74 (d, *J* = 4.8 Hz, 2H), 8.67 (s, 2H), 8.66 (d, *J* = 7.8Hz, 2H), 7.88 (t, *J* = 7.8Hz, 4H), 7.49 (d, *J* = 8.1Hz, 1H), 7.42 (s, 1H), 7.36 (dd, *J* = 5.4 ,1.8Hz, 2H), 6.98 (d, *J* = 8.1 Hz,1H), 4.29 (t, 2H), 4.22 (t, 2H), 3.96 (t, 4H), 3.79 (s, 8H). ^{13}C NMR (75 MHz,$CDCl_3$) δ: 156.21, 155.75, 150.24, 149.84, 149.32, 149.02, 136.84, 136.80, 131.47, 123.80, 123.76, 121.33, 120.71, 118.33, 113.67, 113.38, 113.12, 77.62, 77.19, 76.77, 71.07, 70.48, 70.38, 69.57, 69.45, 68.79. IR (cm^{-1}): 2927, 2868, 1604, 1583, 1516, 1469, 1392, 1267, 1247, 1143, 1076, 1053, 987, 792. MALDI-TOF-MS: found m/z: 500.10 (100%), 501.11 (14%), 502.11 (4%). 4'-tolyl-2,2':6',2"-terpyridine (Ligand L1), yield 47%. ^{1}H NMR (300 MHz, $CDCl_3$) δ(ppm): 8.73 (s, 4H), 8.68 (d, *J* = 8.1Hz, 2H), 7.8 (t, *J* = 8.4Hz, 2H),7.84 (t, *J* = 8.4Hz, 2H), 7.37-7.31 (m, 4H), 2.43 (s, 3H).

General procedure for the receptor 1. An equivalent ligand **L** and Zn (II) perchlorate salts was refluxed in EtOH solution for 3 h. After the reaction solution was cooled to room temperature, precipitation and subsequent recrystallization from acetonitrile/ethanol mixture afforded the desired receptor **1** in 70% yields. The receptor 2 was prepared for comparison study in the same procedure.

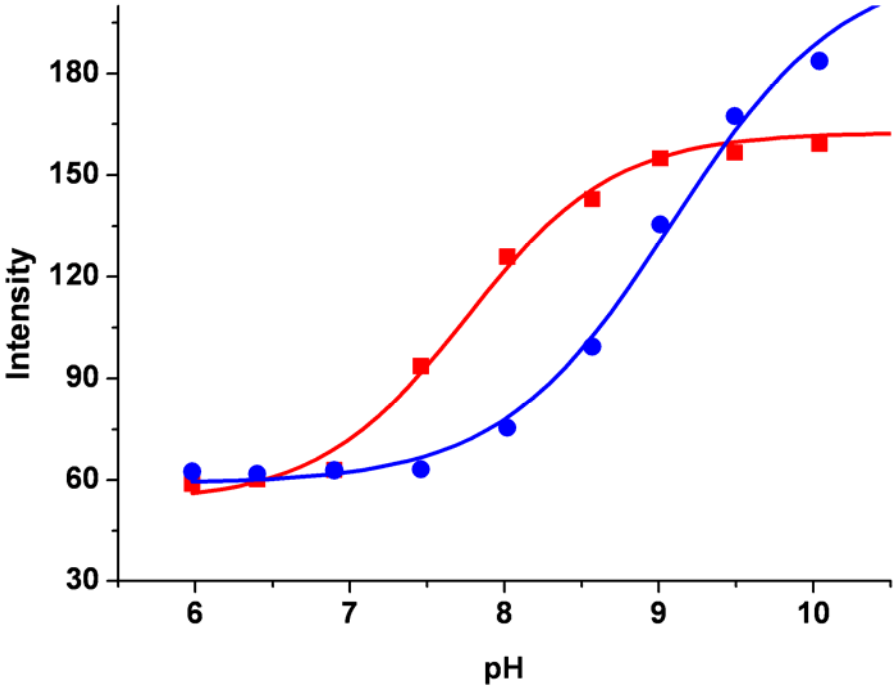


Fig. S1 Fluorescence spectra of receptor **1** (10 μM) (red) and complex **1-Arg** (10 μM) (blue) at barious pH values in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃.

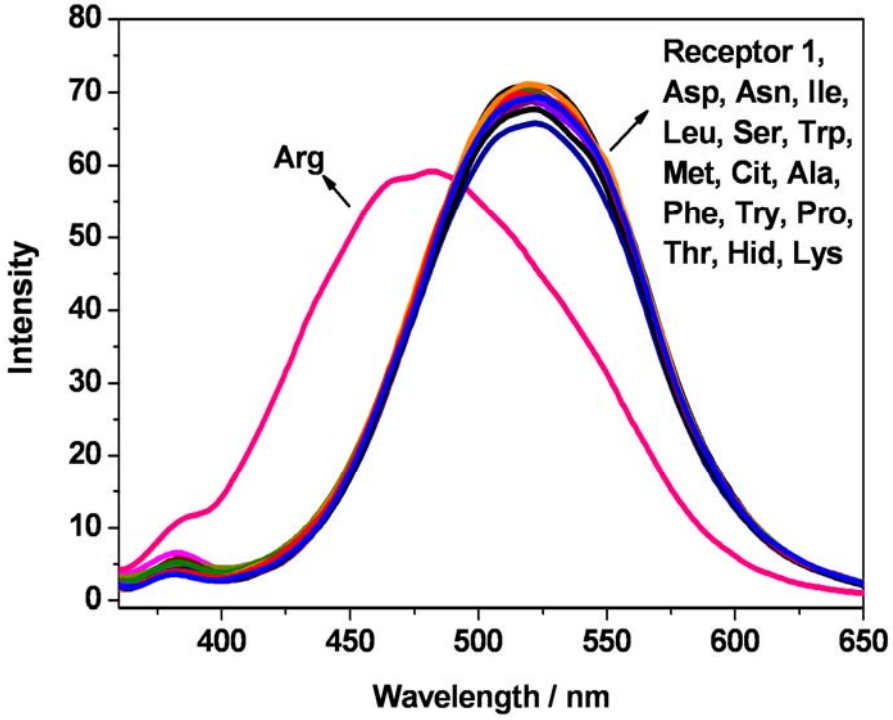


Fig. S2 Emission spectrum (λex = 320 nm, with slit width = 5nm) of receptor **1** (10 μM) upon addition of different L-amino acids (1 mM) in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃.

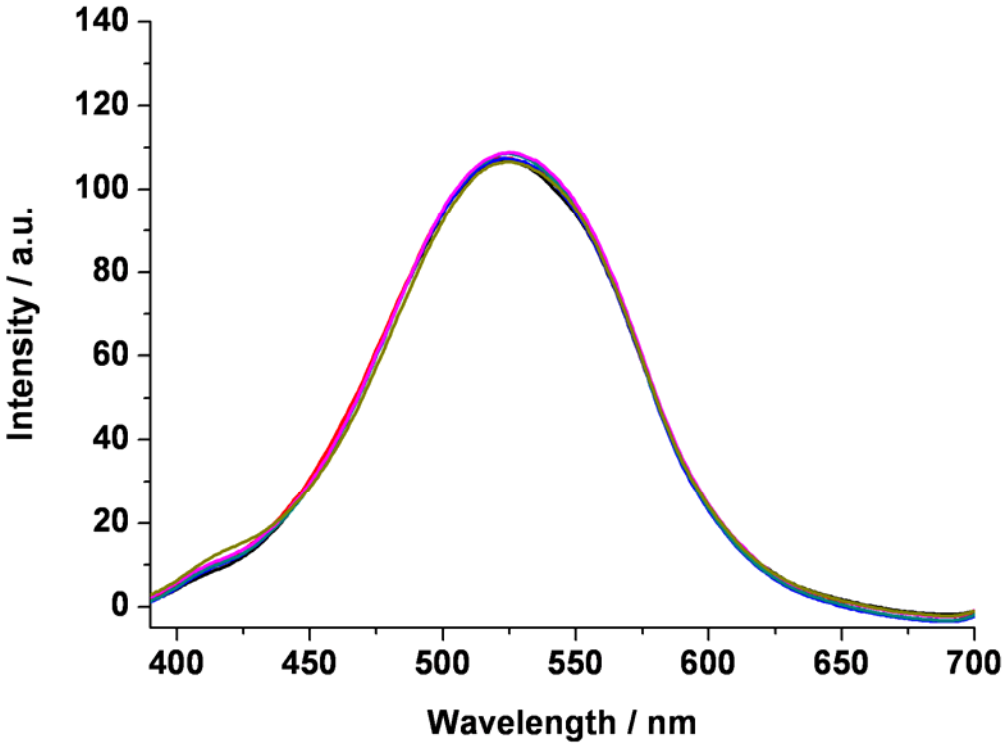


Fig. S3 Emission spectrum (λex = 320 nm, with slit width = 5nm) of receptor **1** (10 μM) upon addition of Cit (0-1 mM) in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃.

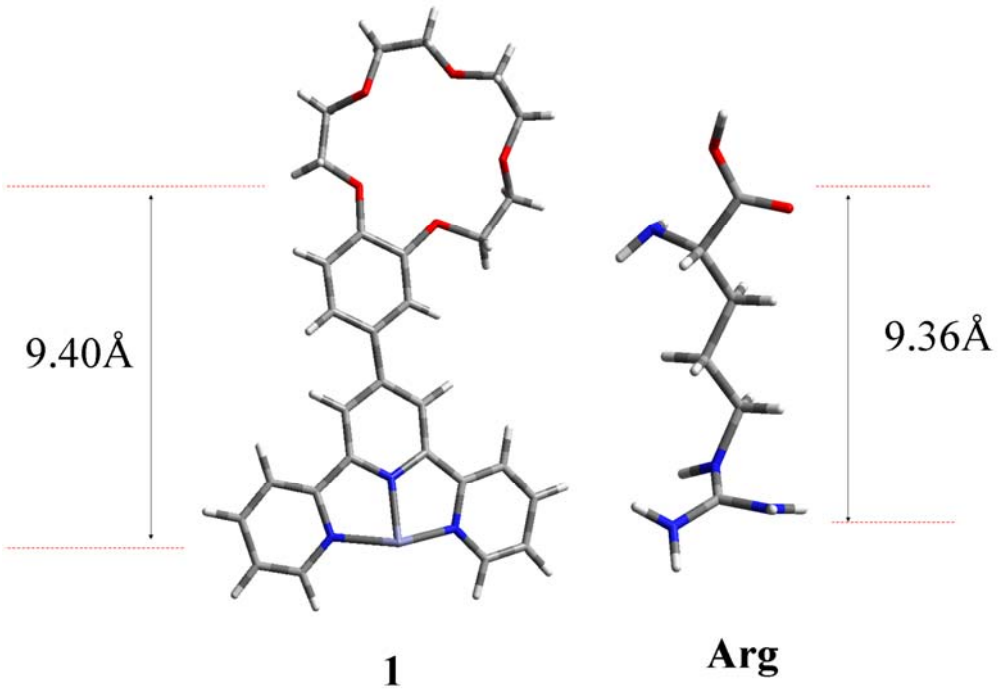


Fig. S4 Molecular size of receptor **1** and Arg obtained through the quantum chemical DFT calculations.

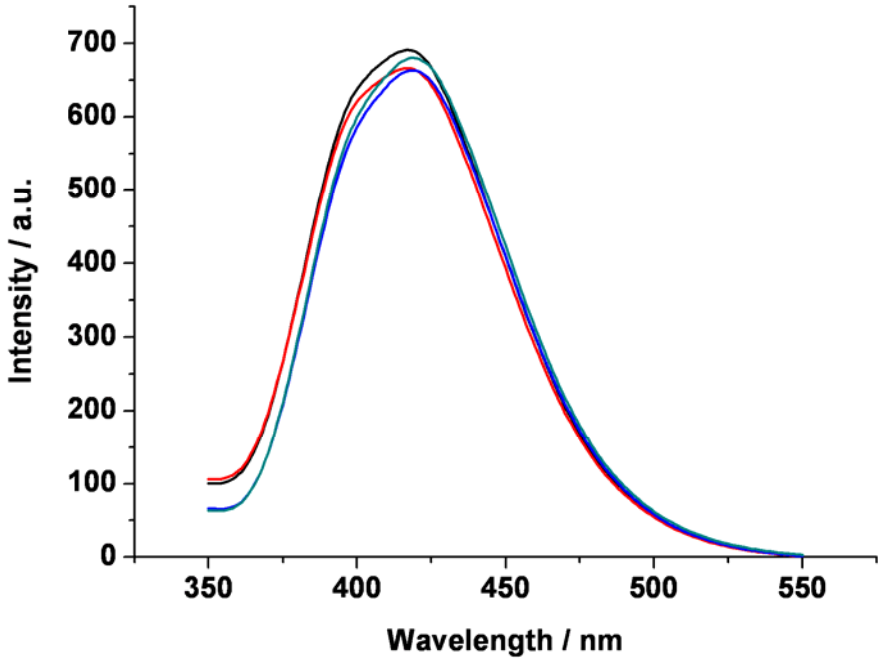


Fig. S5 Emission spectrum (λex = 320 nm, with slit width = 5nm) of receptor **2** (10 μM) upon addition of Arg (0-1 mM) in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃.

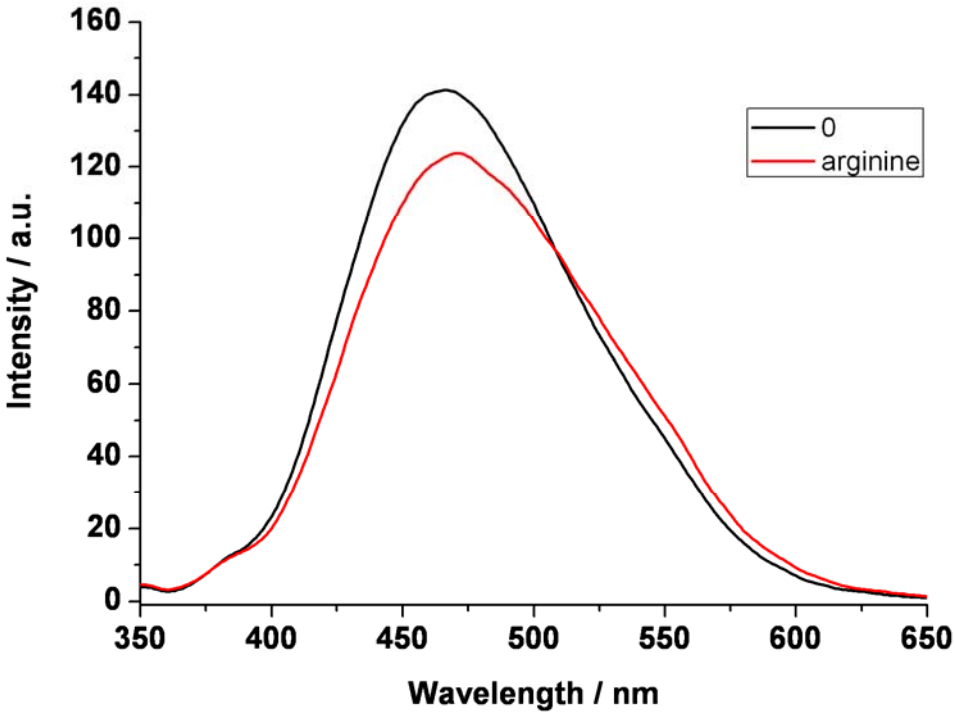


Fig. S6 Emission spectrum (λex = 320 nm, with slit width = 5nm) of ligand **L** (10 μM) upon addition of Arg (1 mM) in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃.

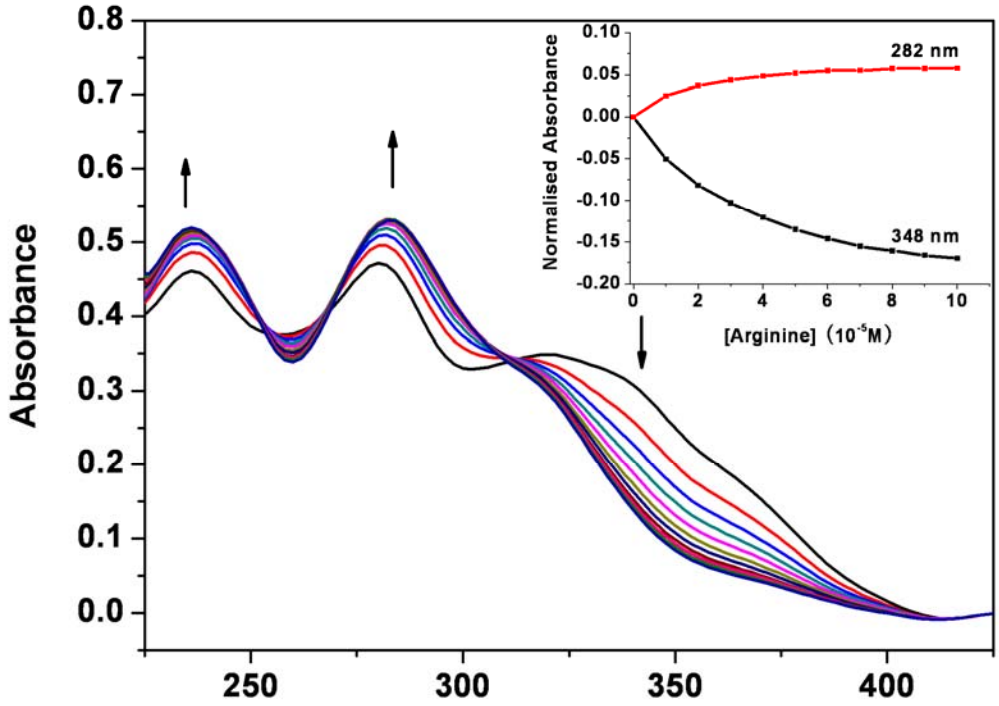


Fig. S7 The UV titrations of of receptor **1** (10 μM) upon addition of Arg (0-1mM) in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃. Inset: titration of the change in the absorbance measured at 282 nm and 348 nm versus the concentration of Arg.

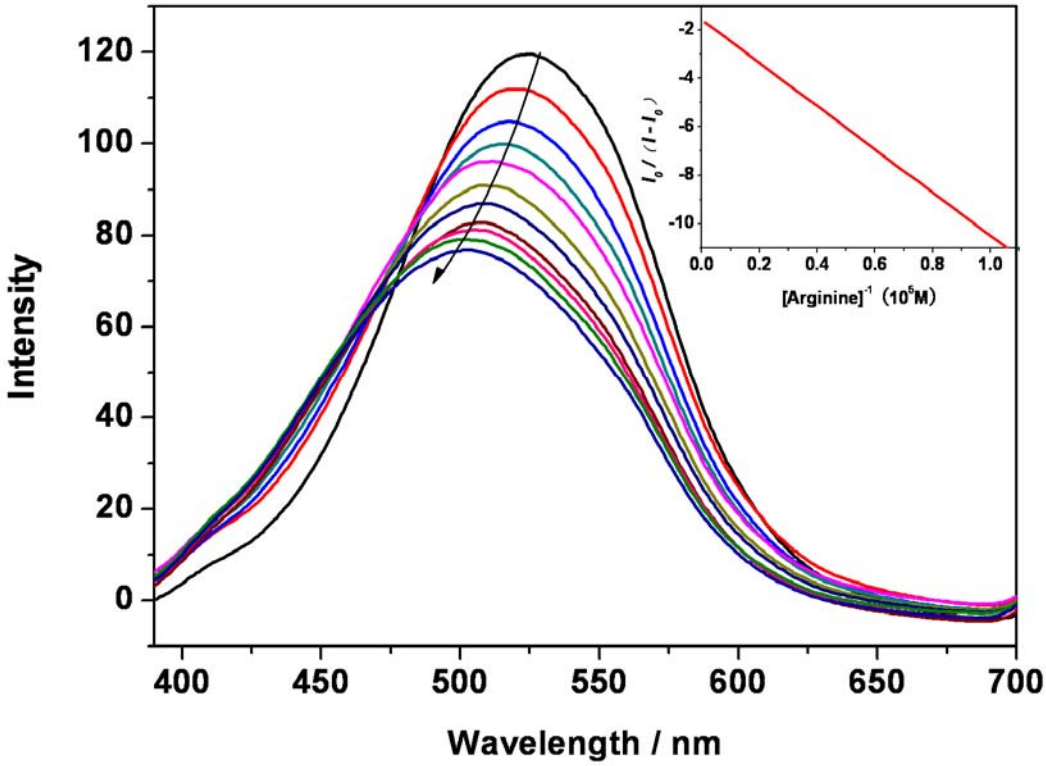


Fig. S8 The fluorescence spectra (λex = 320 nm, slit width = 5 nm) of receptor 1 (10 μM) upon addition with Arg (0-1 mM) in a buffered solution with 10 mM of HEPES (pH 7.4) at 25℃. Inset: the Hildebrand-Benesi plot of F_0 / (F - F_0) versus [Arg]$^{-1}$ at 525 nm.

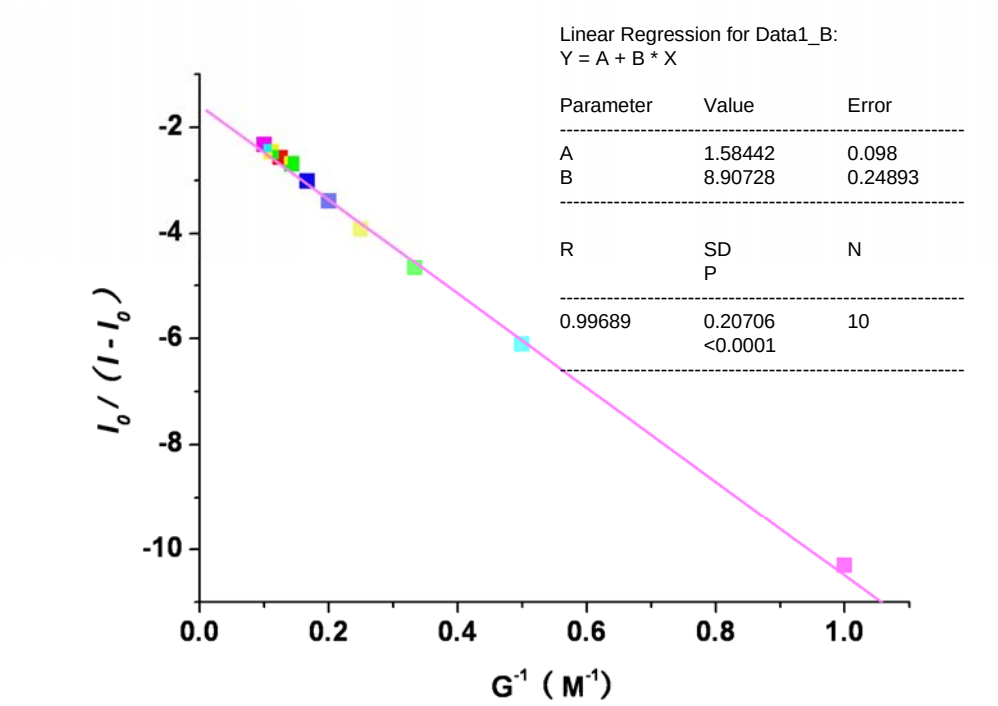


Fig. S9 Plots of $\boldsymbol{I_0}$ / ($\boldsymbol{I}$-$\boldsymbol{I_0}$) vs $[G]^{-1}$ and the linear function

The association constant (K) for 1:1 complexation can be expressed by Eq.

$$\frac{1}{I - I_0} = \frac{1}{(I_1 - I_0)Kass[G]} + \frac{1}{I_1 - I_0}$$

where I, I_1, I_0, and [G] denote the observed fluorescence intensity, fluorescence intensity of receptor **1** when totally bonded to arginine, initial fluorescence intensity of receptor **1** in the absence of amino acid, and total arginine concentration, respectively.

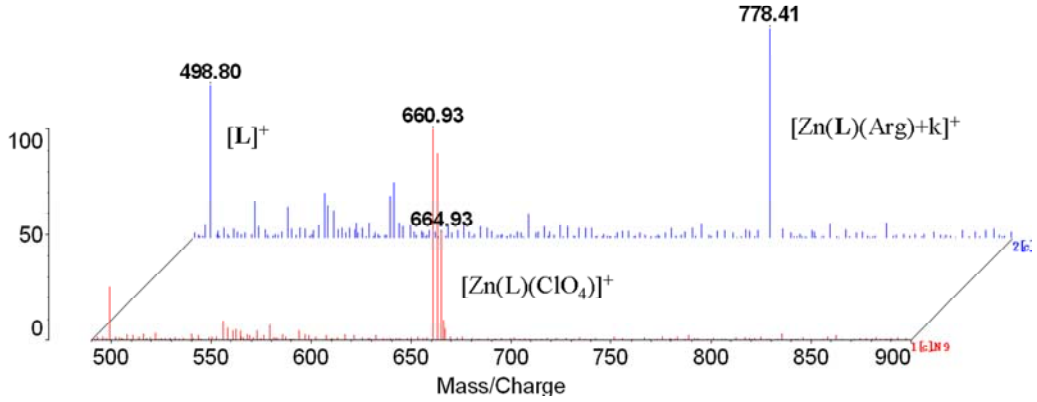


Fig. S10 MALDI-TOF-MS spectra of receptor **1** (red) and after addition of Arg (blue)

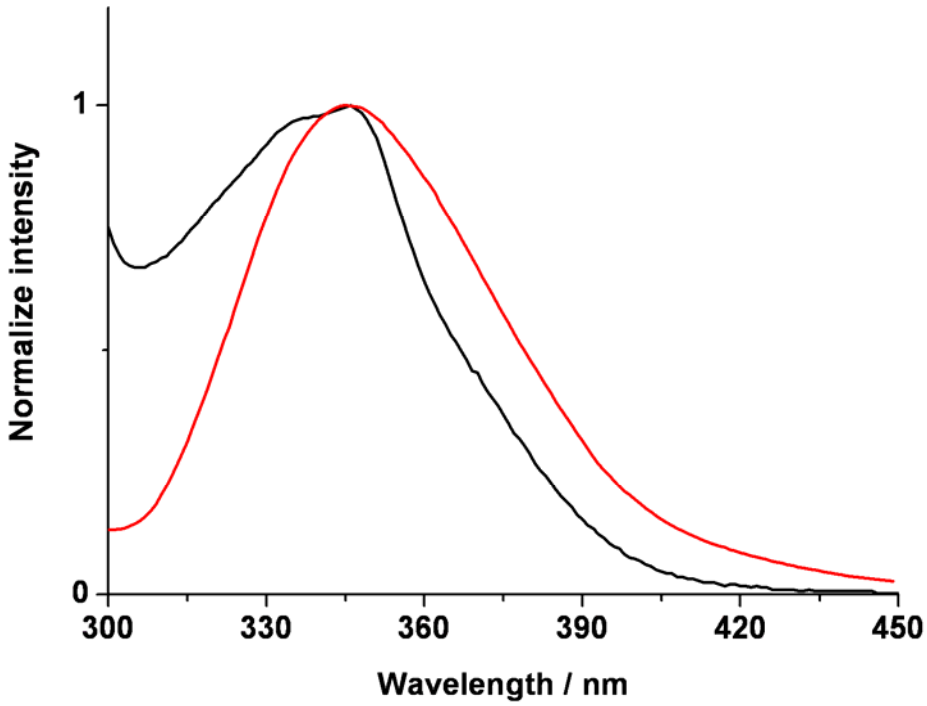


Fig. S11 The spectrum overlaps the between emission spectrum of BSA (red)

and the absorption spectrum of the receptor **1** (drak)

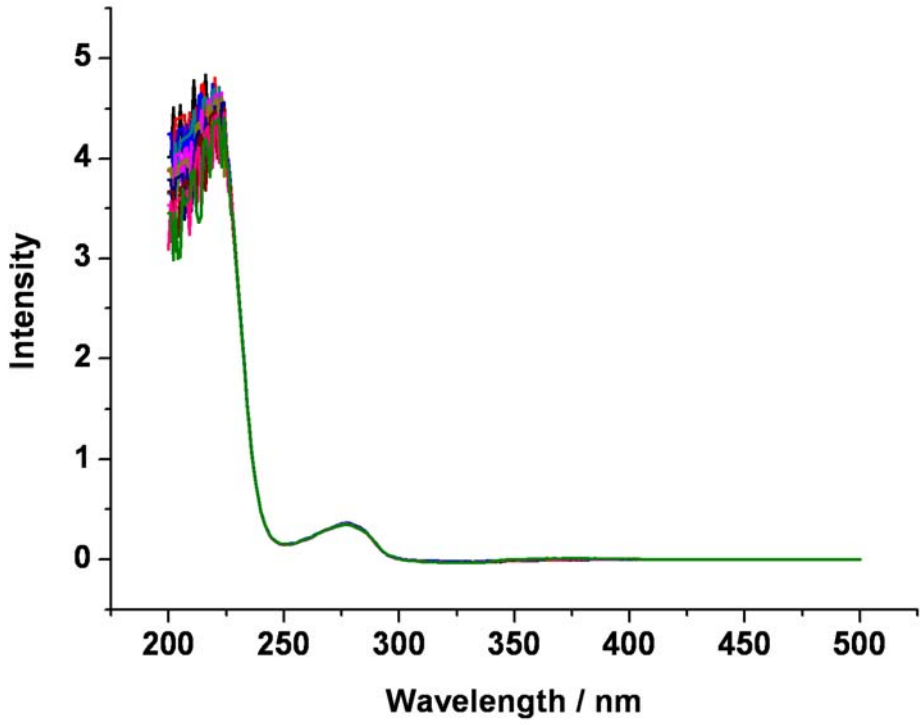


Fig. S12 The UV spectra of of BSA (10 μM) upon addition of receptor **1** in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃.

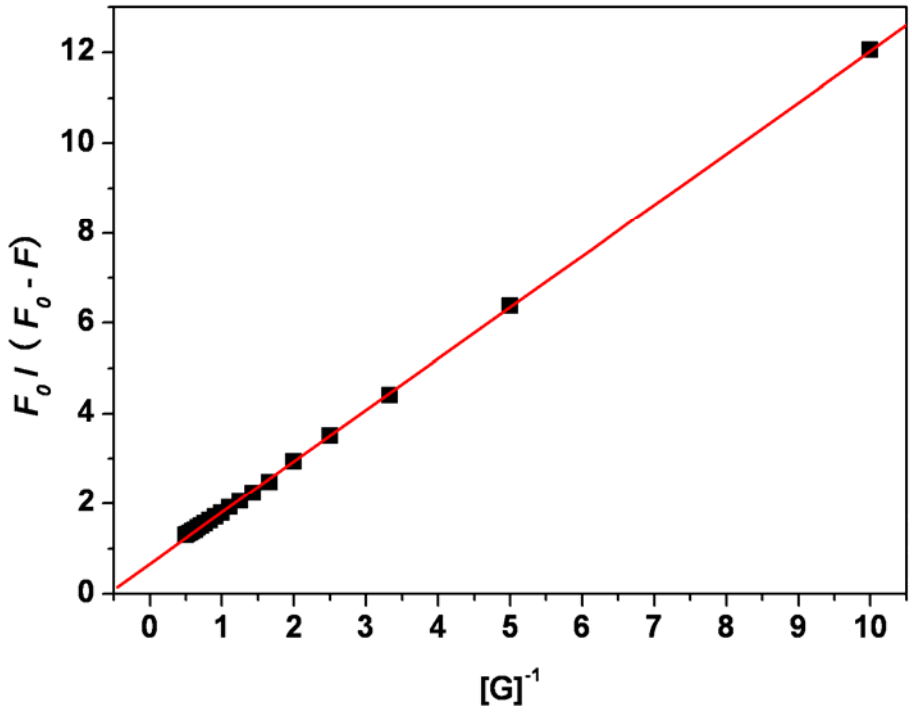


Fig. S13 Plots of $F_0 / (F_0 - F)$ vs $[G]^{-1}$ and the linear function

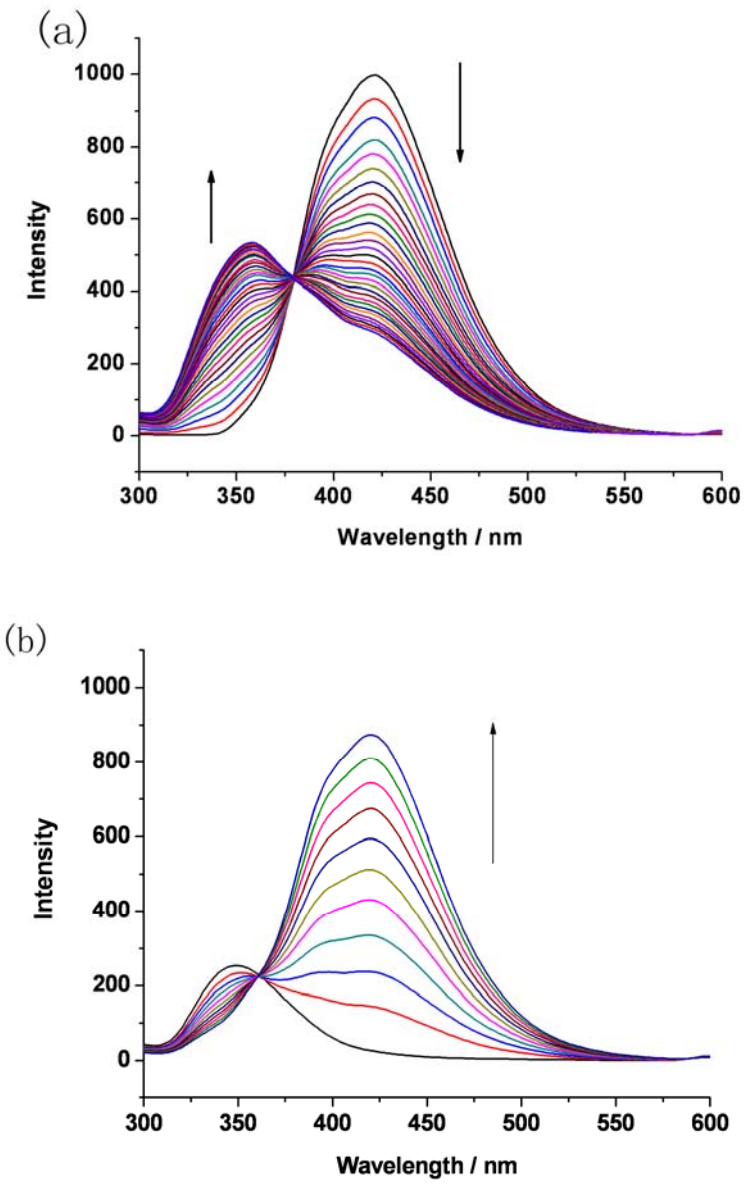


Fig. S14 (a) Emission spectrum of receptor **2** (10 μM) upon addition of BSA (0-0.1 mM) upon excited with 290 nm in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃. (b) Emission spectrum of BSA (10 μM) upon addition of receptor **2** (0-0.1 mM) upon excited with 290 nm in buffered solution with 10 mM of HEPES (pH 7.4) at 25℃

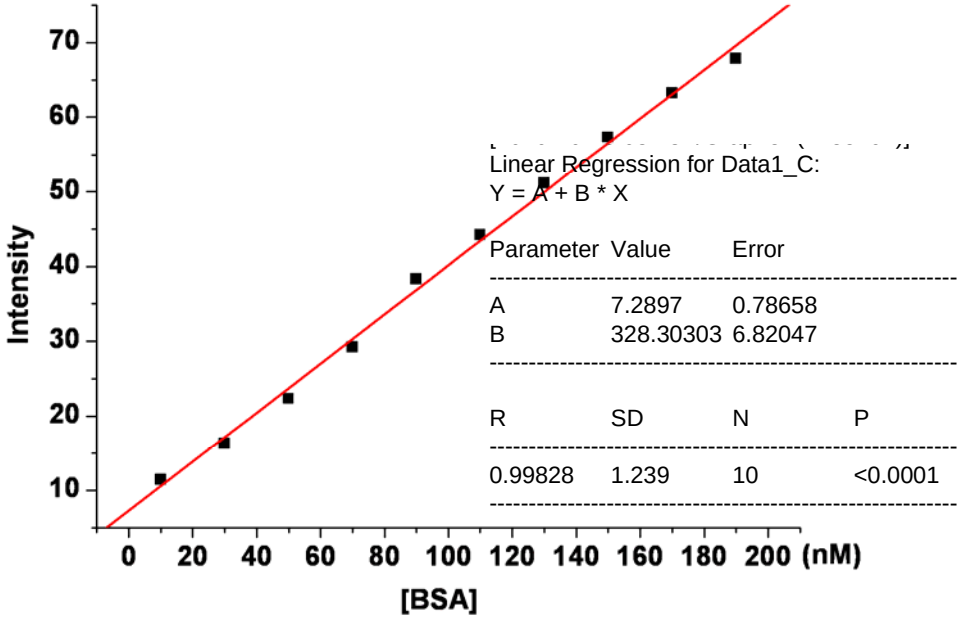


Fig. S15 Plots of F-F_0 vs $[BSA]^{-1}$ and the linear function

Table S1 Binding parameters and thermodynamic parameters

Entry	K_q [b] ($M^{-1}\cdot S^{-1}$)	K_{sv} [b] (M^{-1})	K_{ass} [b] (M^{-1})	ΔG ($kcal\cdot M^{-1}$)	R	LOD (nM)
1. Arg	8.41×10^{11}	8.41×10^{3}	1.87×10^{4}	-24.26	0.9968	205
2. BSA	7.84×10^{12}	7.84×10^{4}	5.91×10^{5}	-34.95	0.9998	15

[a] All data were acquired in buffered HEPES solution (pH 7.4) at 25□. [b] Determined by fluorescence titration. [c] ($\Delta G = -RT\ln Kass$)

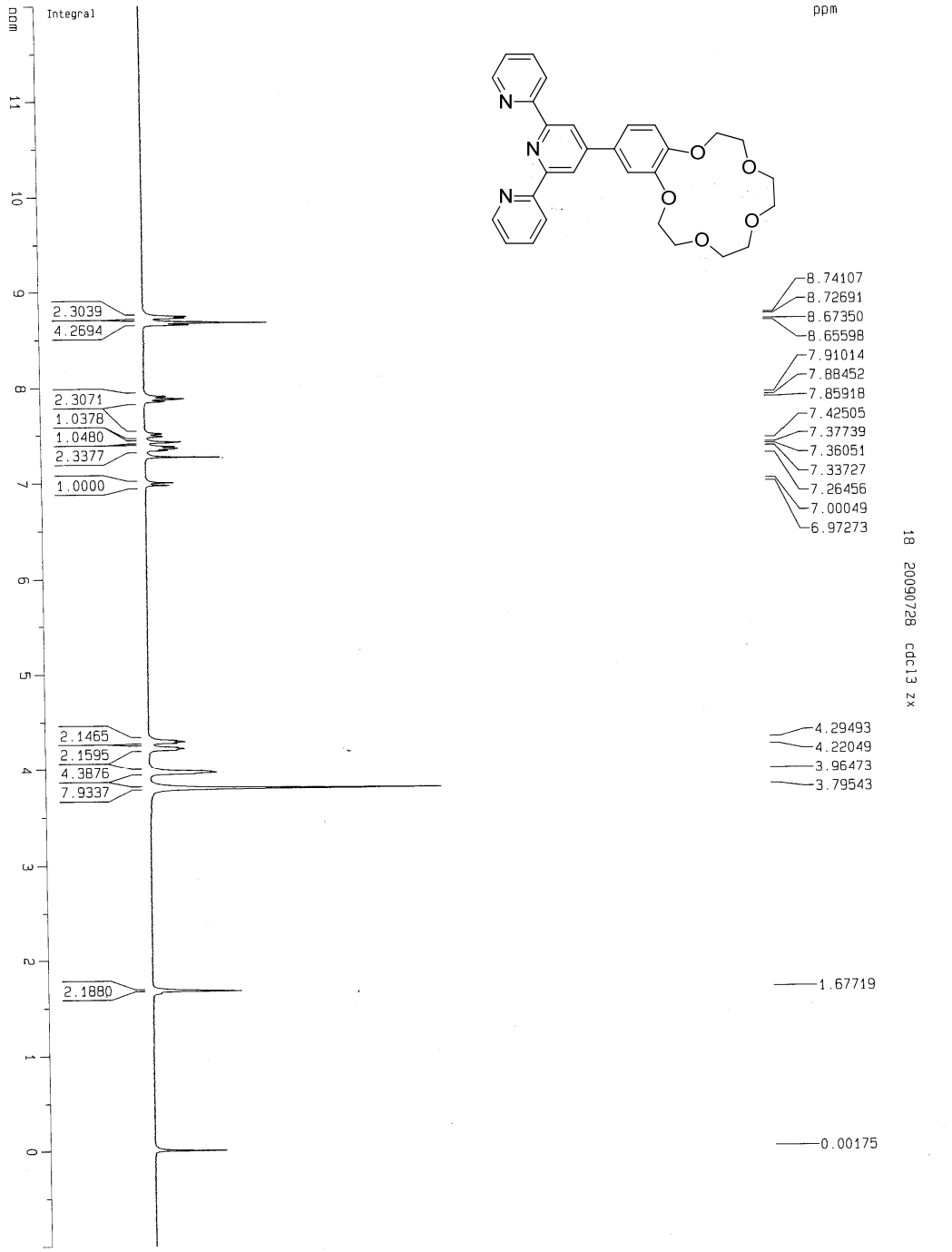


Fig. S16 ^{1}H-NMR of ligand **L**

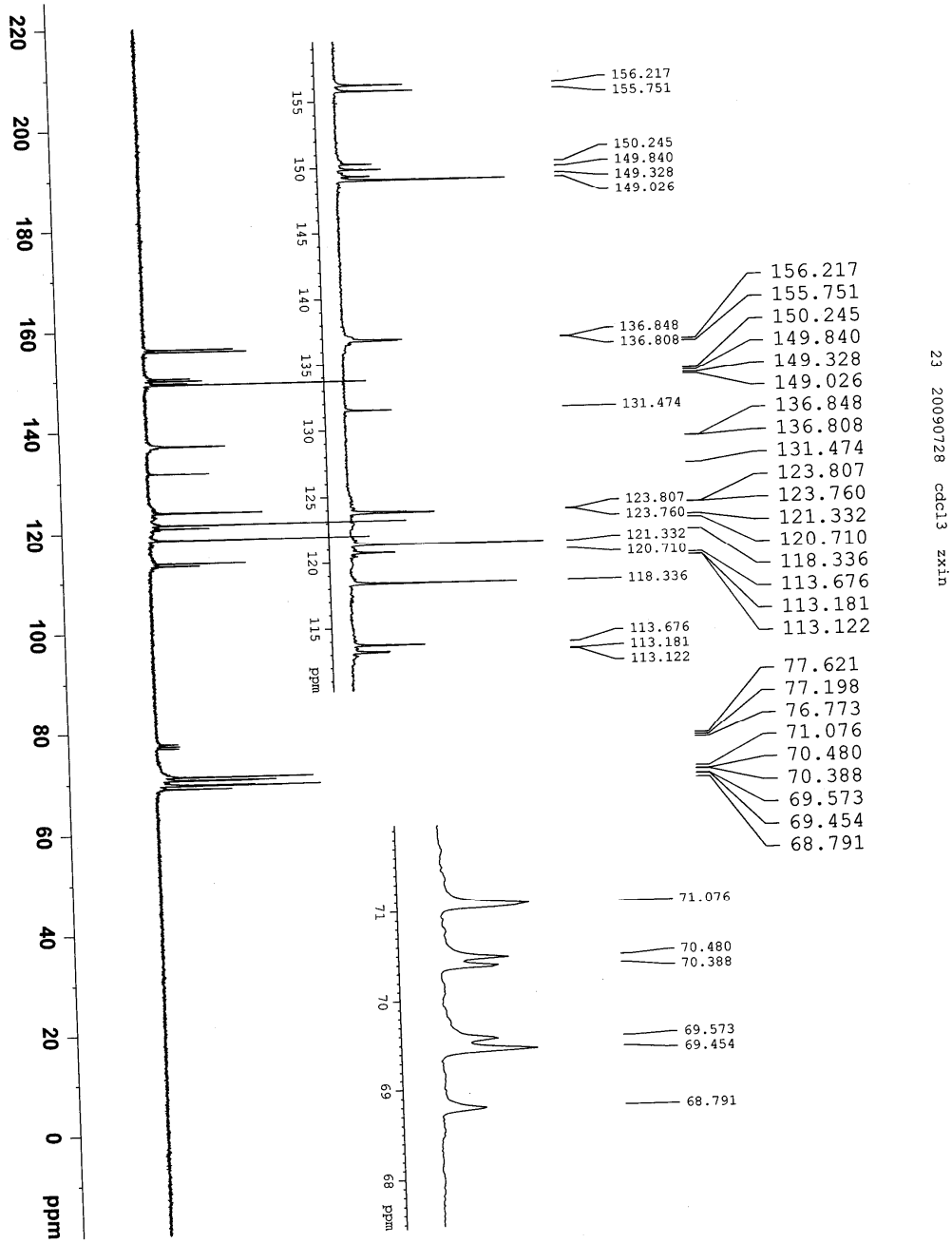


Fig. S17 ^{13}C-NMR of ligand **L**

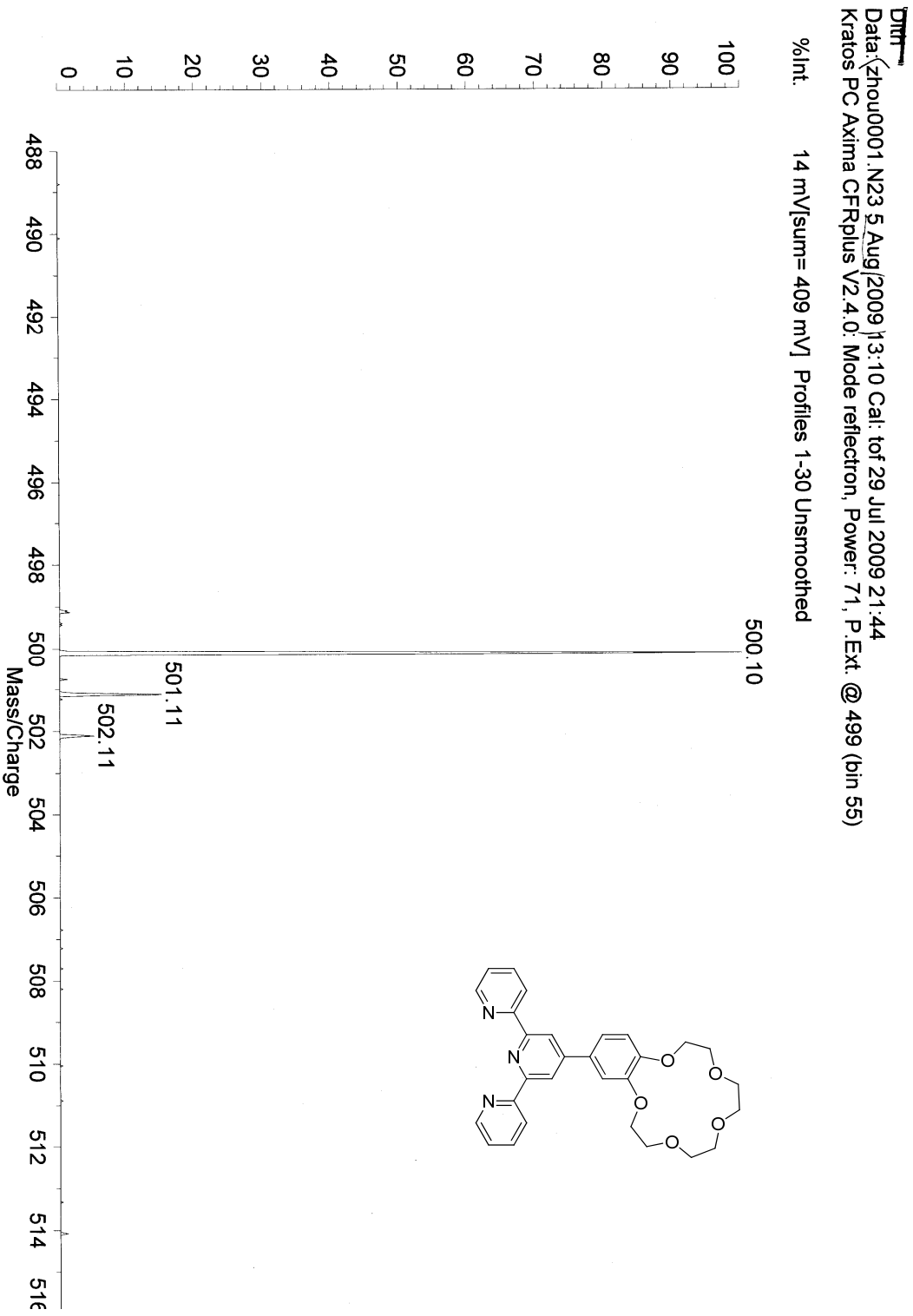


Fig. S18 MALDI-TOF-MS of ligand **L**

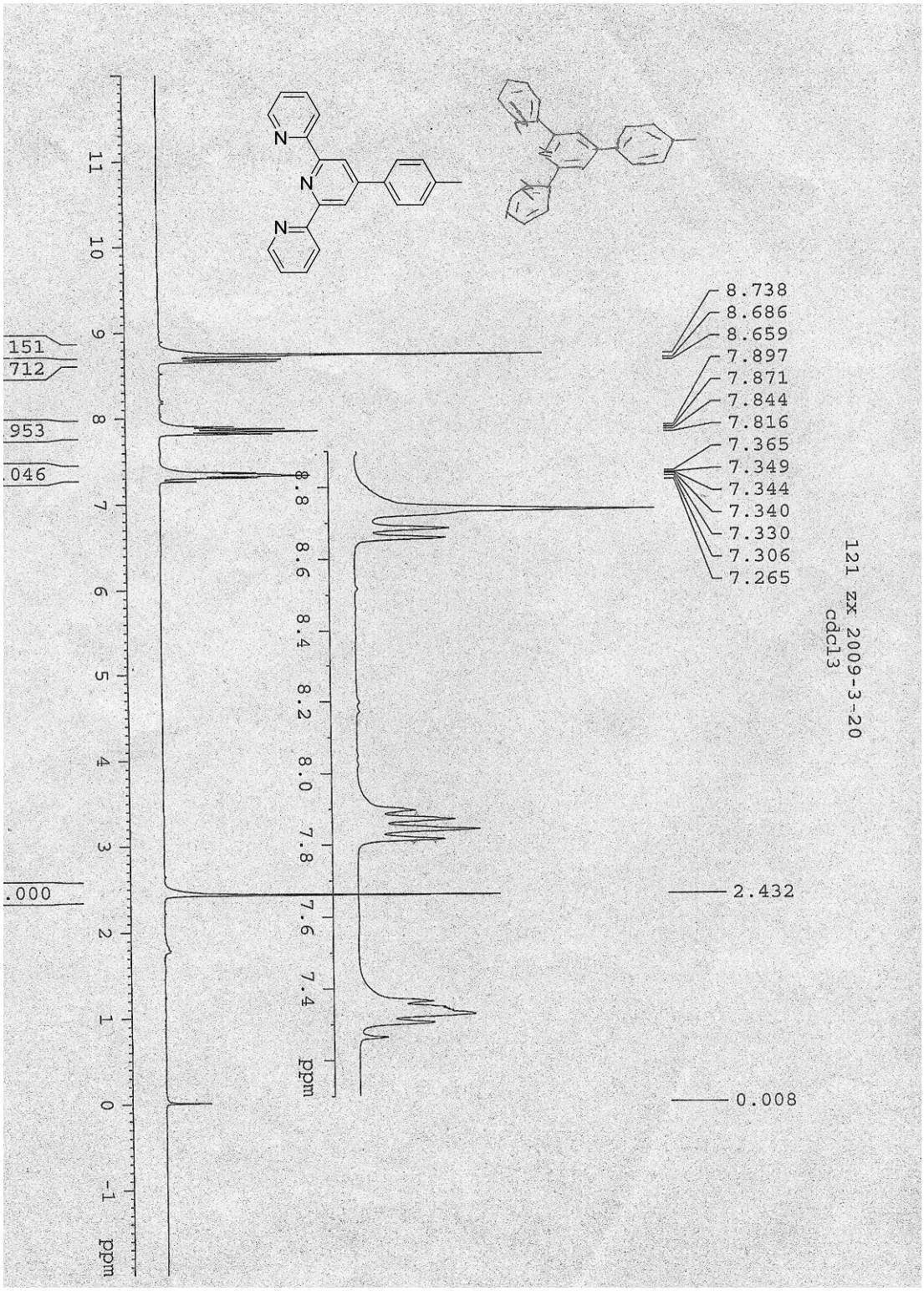


Fig. S19 ^{1}H NMR of ligand **L1**

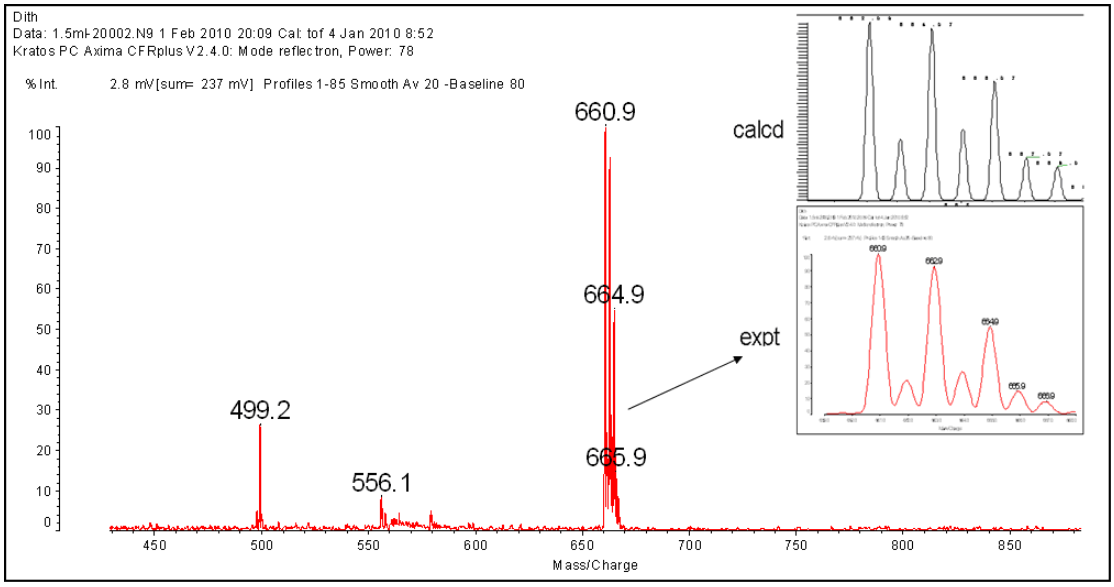


Fig. S20 MALDI-TOF-MS of receptor **1** and isotopic distributions (red) along with caculated one(drak)

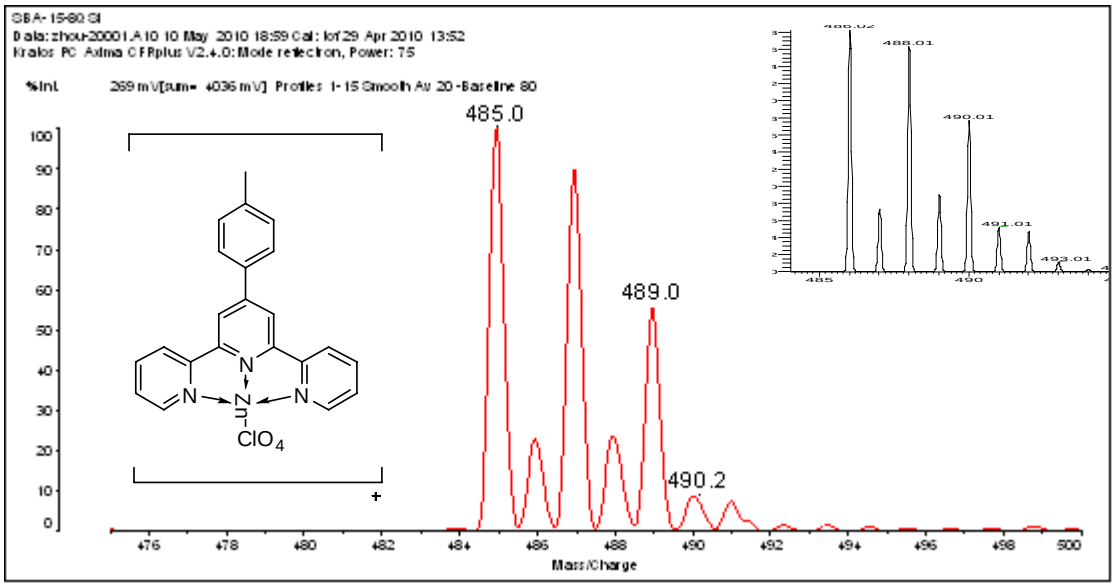


Fig. S21 MALDI-TOF-MS of receptor **2** and **3** with isotopic distributions (red) and caculated one(drak)