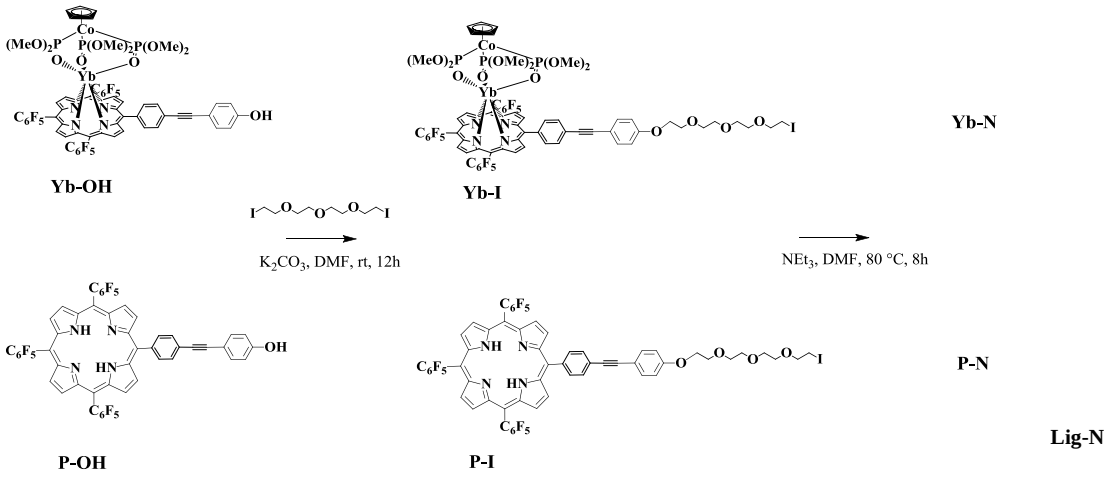
Supporting Information

Experimental

Preparation of Yb-N, Yb-Rh and Lig-N

All chemicals used were of reagent-grade and were purchased from Sigma–Aldrich and used without further purification. Preparations of intermediates Yb-OH and P-OH were performed according to our previous procedures. All analytical-grade solvents were dried by standard procedures, distilled and deaerated before use. NMR spectra were recorded on a Bruker Ultrashield 400 Plus NMR spectrometer. The ^{1}H NMR chemical shifts were referenced to tetramethylsilane, TMS (d = 0.00). High-resolution mass spectra, reported as m/z, were obtained on a Bruker Autoflex MALDI-TOF mass spectrometer. The synthetic route of intermediates and Yb complexes **Yb-N** and **Lig-N** were shown in the figures S1.



Figures S1. The synthetic routes of **Yb-N** and **Lig-N**

Yb-N: Yield: 74%; ^{1}HNMR ($CDCl_3$): δ -5.15 (s, 5H), -4.11 (s, 4H), -2.69 (s, 8H), 0.45 (s, 2H), 0.64 (t, *J* = 7.20 Hz, 2H), 1.19 (t, *J* = 7.0 Hz, 1H), 2.58 (s, 2H), 2.96 (s, 2H), 3.28 (t, *J* = 4.0 Hz, 2H), 3.47 (m, 2H), 3.82 (s, 2H), 4.25 (s, 2H), 6.31 (s, 18H), 8.24 (d, *J* = 8.0 Hz, 2H), 8.64 (s, 1H), 9.10 (s,1H), 14.63 (s, 2H), 14.98 (s, 2H), 15.30 (s, 2H), 15.65 (s, 2H), 17.40 (s, 1H); ^{31}PNMR ($CDCl_3$): δ 64.38; MALDI-TOF MS: calcd. for [M^+] 1883.2664, found 1883.2688;

Lig-N: Yield: 82%; ^{1}HNMR ($CDCl_3$): δ -2.85 (s, 2H), 1.33 (t, *J* = 7.20 Hz, 9H), 3.45 (q, *J* = 7.20 Hz, 6H), 3.66 (m, 10H), 3.88 (t, *J* = 4.40 Hz, 2H), 3.98 (s, 2H), 4.20 (t, *J* = 4.4 Hz, 2H), 6.98 (d, *J* = 8.80 Hz,2H), 7.62 (d, *J* = 8.80 Hz, 2H), 7.93 (d, *J* = 8.4 Hz, 2H), 8.19 (d, *J* = 8.4 Hz, 2H), 8.83 (m, 8H); MALDI-TOF MS: calcd. for [M^+] 1260.3545, found 1260.3547;

Photo-physical Measurement

Linear induced photophysical properties: UV-Visible absorption spectra in the spectral range 200 to 1100 nm were recorded by an HP Agilent UV-8453 spectrophotometer. Single-photon luminescence spectra were recorded using an Edinburgh Instrument FLS920 Combined Fluorescence Lifetime and Steady state spectrophotometer that was equipped with a visible to near-infrared -sensitive photomultiplier by in nitrogen flow cooled housing and PTI QM40 fluorometer. The spectra were corrected for detector response and stray background light phosphorescence. The quantum yields of the **Yb-N** was measured by comparative method using the Yb emission in (Yb-TFPP-acac), Yb(tta)Phen, $Yb(tta)_3$ and **Yb-Rh** as reference. [1c] Sample quantum yield was evaluated by the following equation:

$$\Phi_s = \left(\frac{G_s}{G_r}\right)\left(\frac{\eta_s}{\eta_r}\right)^2 \Phi_r$$

Where the subscripts r and s denote reference and sample respectively, Φ is the quantum yield, G is the slope from the plot of integrated emission intensity vs absorbance, and η is the refractive index of the solvent.

Stability test via titration: Titration experiments were conducted to investigate the effect of several common biological anions and human serum albumin (HSA) on the two ytterbium complexes. Liquid concentrated stock solutions of each anion, as well as HSA, were added individually and gradually to a solution of the complex concerned. Addition was ceased either when the volume of added anion totaled 5% of the complex solution or the influence on complex luminescence was saturated. Single-photon luminescence spectra were determined via the aforementioned procedures. [1c]

Two/three photon induced emission measurement: For multi-photon experiments, the 750 nm pump source was from the fundamental of a femtosecond mode-locked Ti:Sapphire laser system (output beam ~ 150 fs duration and 1 kHz repetition rate). The lasers were focused to spot size ~ 50 μm via an f = 10 cm lens onto the sample. The emitting light was collected with a backscattering configuration into a 0.5 m spectrograph and detected by a liquid nitrogen-cooled CCD detector. A power meter was used to monitor the uniform excitation.

Cell culture

Human HeLa (cervical carcinoma) and WPMY-1 (normal prostate stroma immortalized cell) cells were grown in DMEM medium; A549 (lung adenoma) were maintained in Ham's F12K medium with L-glutamine (N3520, Sigma, St. Louis, MO, USA); QSG-7701 (normal liver cell), HK-1, HONE1 (nasopharyngeal carcinoma) were grown in RMPI-1640 medium; MRC-5 (normal lung fibroblasts) and SK-N-SH (neuroblastoma) cells were grown in MEM medium. All the mediums were supplemented with 10% (v/v) fetal bovine serum (FBS), 100 units/ml penicillin and 100 μg/ml streptomycin.

Cytotoxicity tests by MTT cell viability assay

cancer/normal cells ($5x10^3$) were seeded and exponentially grown in 96-well plates and were dosed with various concentrations of chemicals **Yb-N**, **Lig-N** and **Yb-Rh** for 24 h, respectively. Cell viability were analyzed using the MTT assay by addition of equal amounts of 3-(4, 5-dimethylthiazol-2-yl)-2 and 5-diphenyltetrazolium bromide (0.5 mg/ml) and incubation at 37 ºC and 5% CO_2 for 4 hours to produce formazan in living cells. The reaction products were dissolved by dimethyl sulfoxide and the absorbance of solutions were measured in Bio-Rad iMark microplate reader. Quadruplicates were performed for each dose of the chemicals. Data collection, procession and graphing were managed with the GraphPad Prism 5 software.

Isothermal Titration Calorimetry

Phospholipids 1,2-dioleoyl-sn-glycero-3-phosphocholine (DOPC) (#850375P) and 1,2-dioleoyl-sn-glycero-3-phospho-L-serine (sodium salt) (DOPS) (#840035P) were obtained from Avanti Polar Lipids (Alabster, AL, USA). Weighted lipids (DOPC or DOPS) were dissolved in 70% methanol plus 30% water to make a final concentration of 0.2 mM. ITC experiments were performed with the MicroCal iTC200 titration calorimeter (Northampton, MA). Chemicals **Yb-N**, **Lig-N** and **Yb-Rh** were dissolved in the identical buffer for lipids as mentioned, to a final concentration of 2 mM. Titrations were carried out at 25 ^oC with a stirring speed of 650 r.p.m. and a 120 second spacing between each 2 μL injection. Total 20 injections of chemicals were performed into a cell of 200 μL volume loaded with the phospholipids, respectively. Parallel experiments were performed by injecting chemicals into the buffer to determine heats of dilution, which were subtracted from their respective titrations in

the final figure. Experimental data was fitted by the *one set of sites* model with MicroCal Origin.

Flow cytometry

The density of the trypsinized cancer/normal cell in DMEM medium was set to be 100,000 cells/mL. The cell was transferred to the 6-well plate incubated for 24 hours at 37 ^{o}C and 5% CO_2. After that, the medium was removed and added the medium/DMSO (99:1, v/v) sample at 5μM and incubated for 24 hours. Then, the medium was removed and washed with PBS for three times. The cell was trypsinized and the PBS was added up to 1mL. The test tube containing sample-loaded cell was transferred to the Flow Cytometer (BD FACS Calibur) and undergo the analysis.

Two-Photon induced emission and cell imaging

Cells were seeded on coverslip in 35-mm culture dishes for overnight. The cells were initially incubated with compounds **Yb-N**, **Lig-N** and **Yb-Rh** (1 μM and 3 hours incubation). Then the unabsorbed chemicals were washed out with PBS and the cells were subjected to two-photon induced confocal microscopic imaging. Briefly, images were captured using the Leica SP5 (upright configuration) confocal microscope equipped with a femtosecond-pulsed Ti:Sapphire laser (Libra II, Coherent) inside the tissue culture chamber (5% CO_2, 37 ^{o}C). The excitation beam produced by the femto-second laser, which was tunable from 680-1050 nm, was focused on the adherent cells through a 40x oil immersion objective.

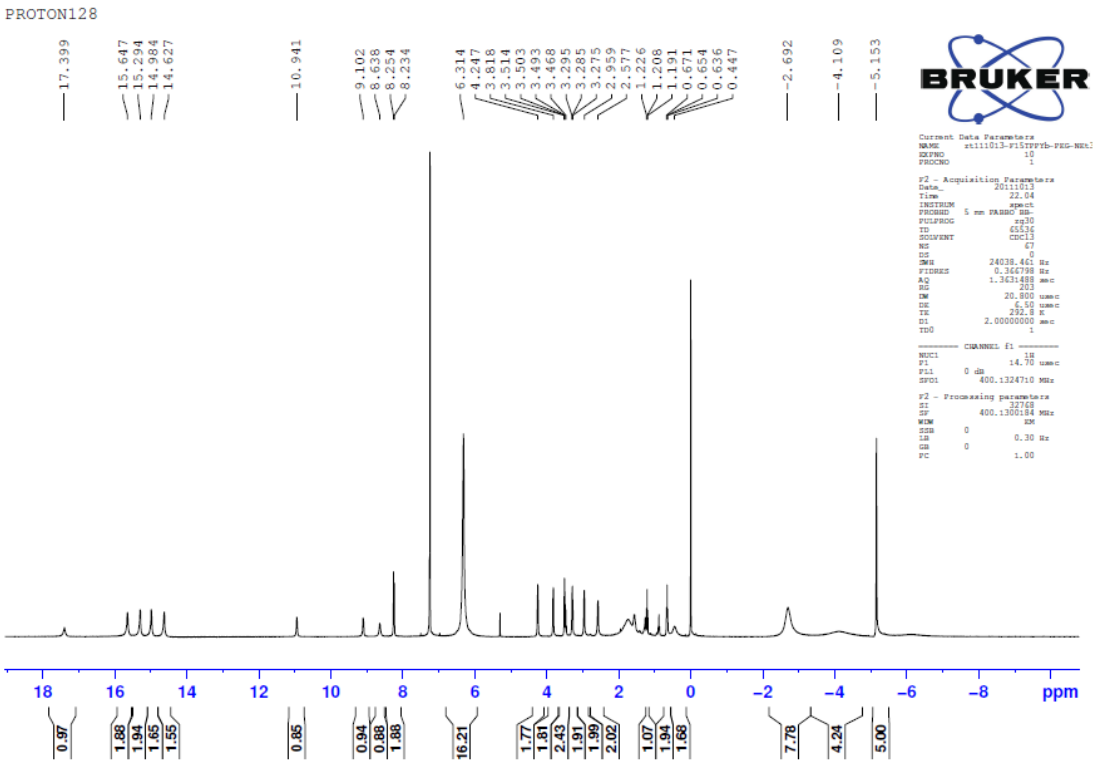


Figure S2. 400 MHz-^{1}H-NMR ($CDCl_3$) spectra of **Yb-N**

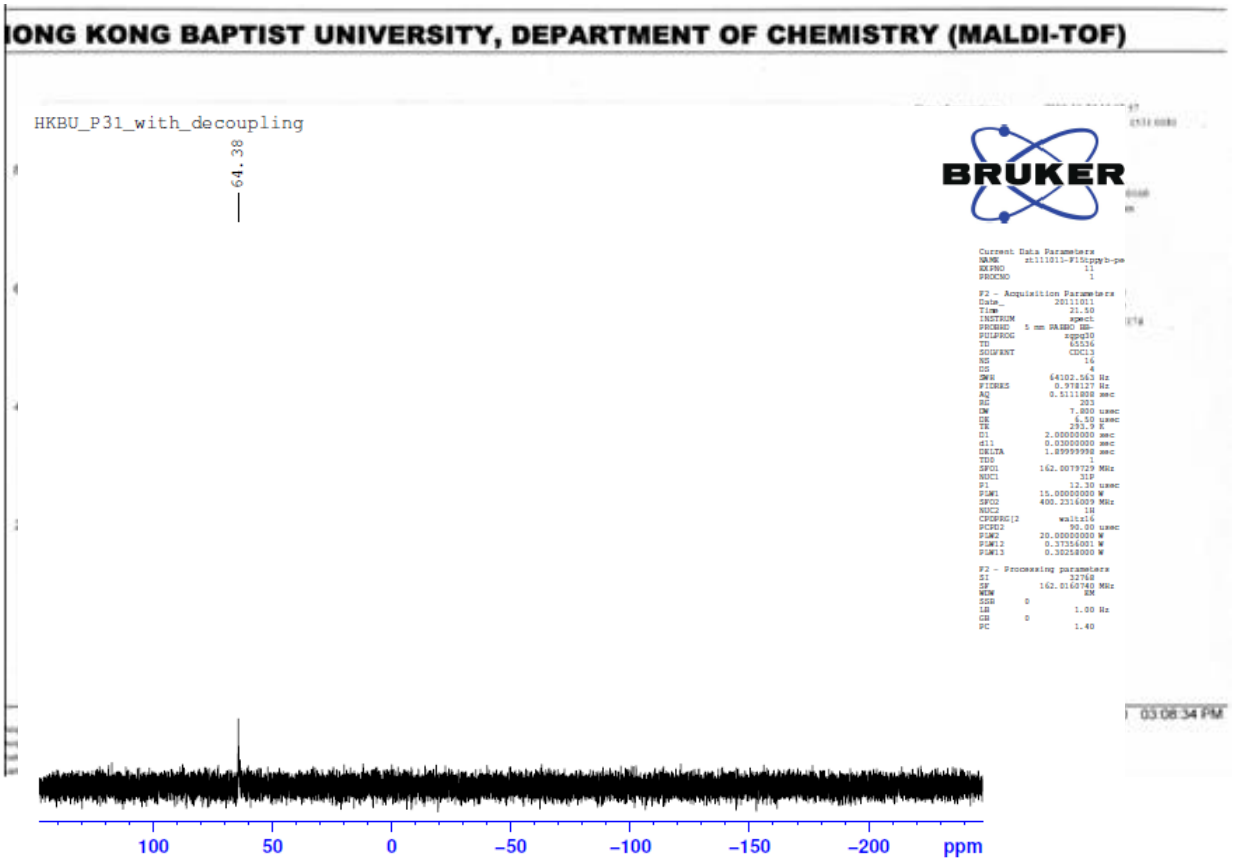


Figure S3. 400 MHz-^{31}P-NMR ($CDCl_3$) spectra of **Yb-N**

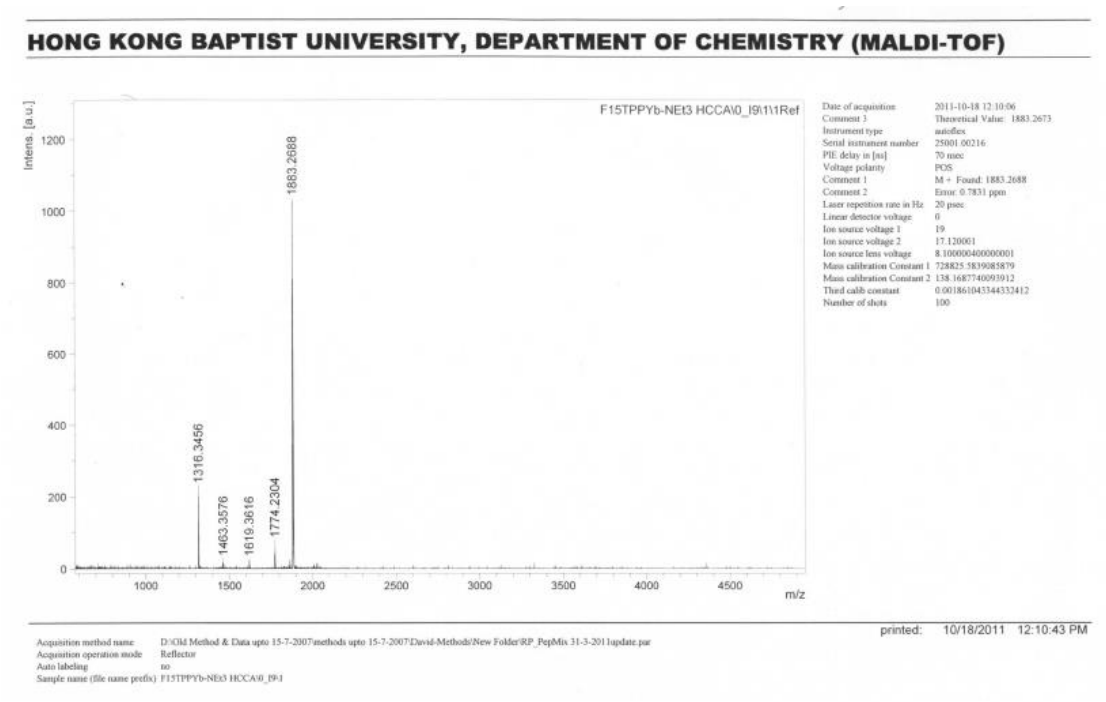


Figure S4. MALDI-TOF spectra of **Yb-N**

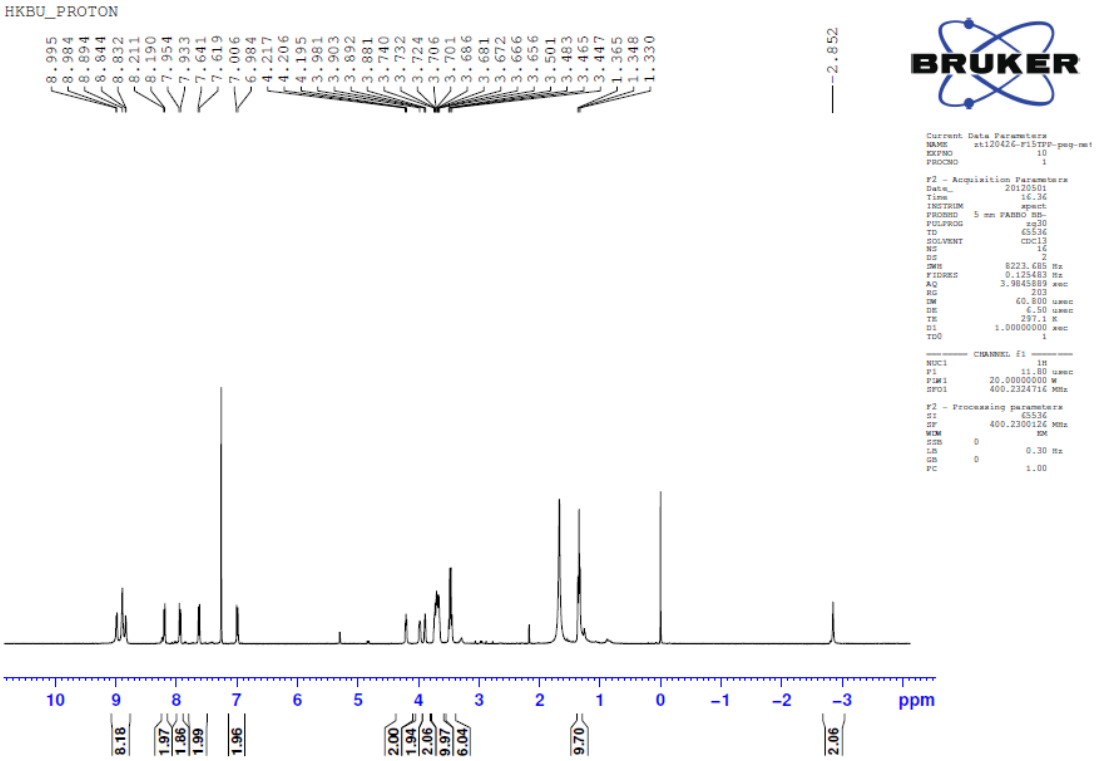


Figure S5. 400 MHz-^{1}H-NMR ($CDCl_3$) spectra of **Lig-N**

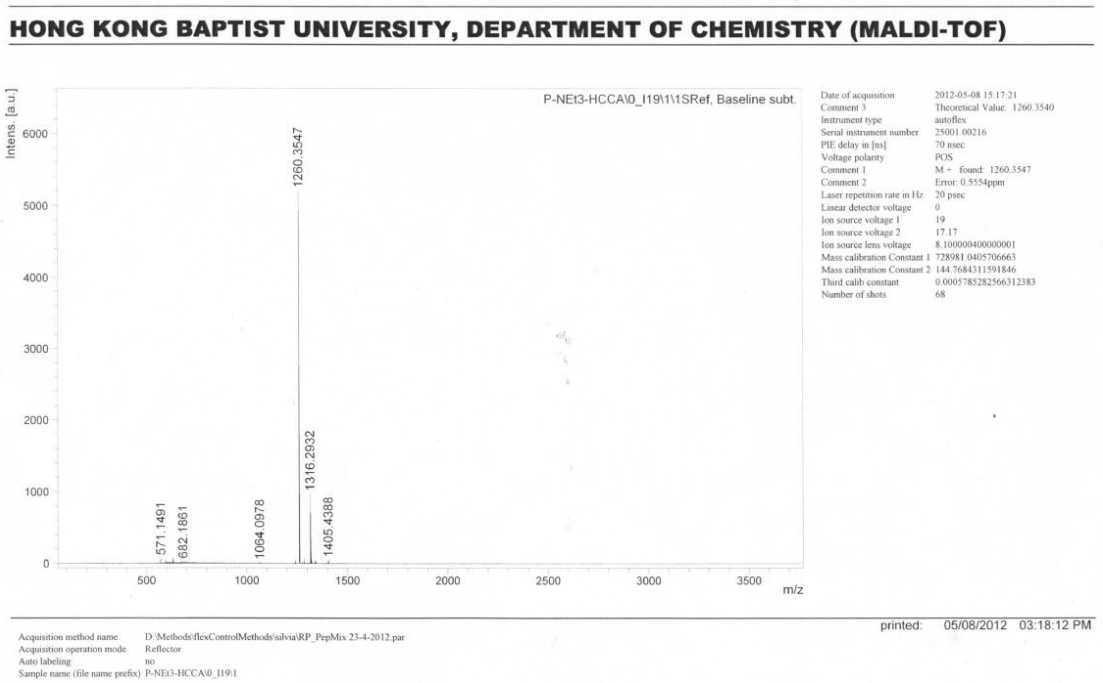


Figure S6. MALDI-TOF spectra of **Lig-N**

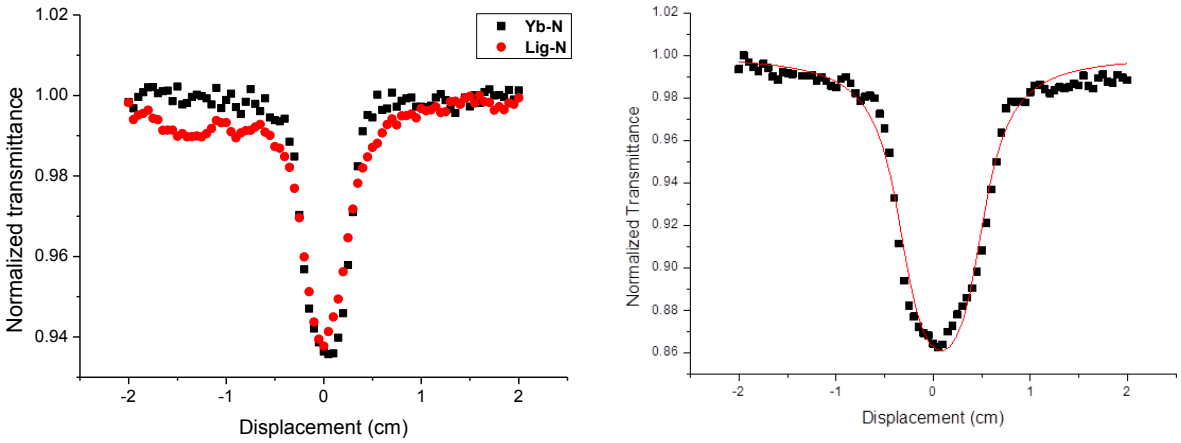


Figure S7. Two-Photon absorption cross section measurements of (left) **Yb-N, Lig-N** and (right) **Yb-Rh** in aqueous solution. (λ_{ex} = 860 nm)

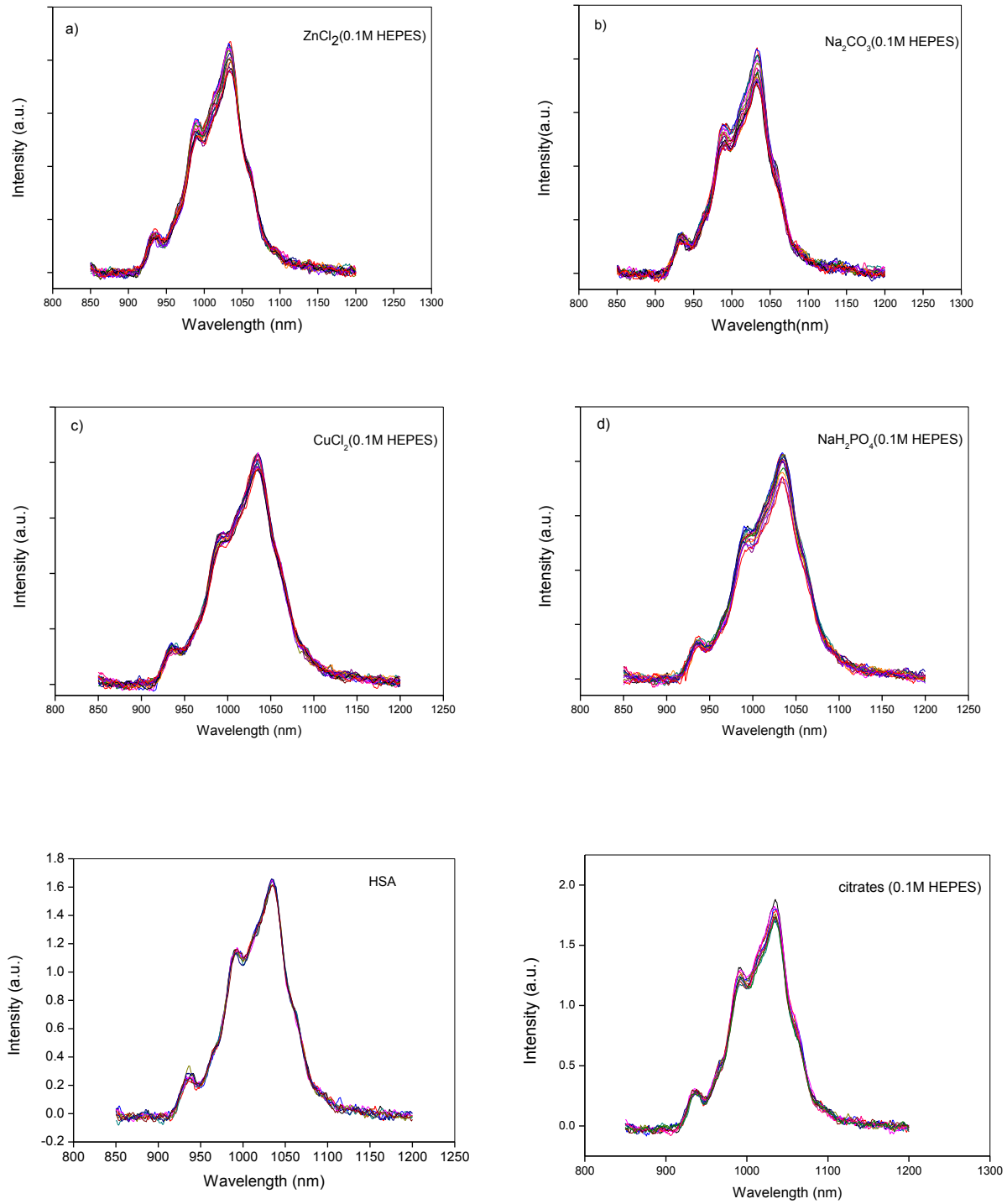


Figure S8 Changes in ytterbium emission spectra for complex **Yb-N** (10 μM in HEPES, pH = 7.4), following international addition of (a) $ZnCl_2$, b) bicarbonates (1 mM to 50 mM), (c) $CuCl_2$, d) biphosphates (1 mM to 50 mM), e) HSA (0.1 mM to 1 mM) and (f) citrates (0.1 mM to 2.0mM). (λ_{ex} = 430 nm)

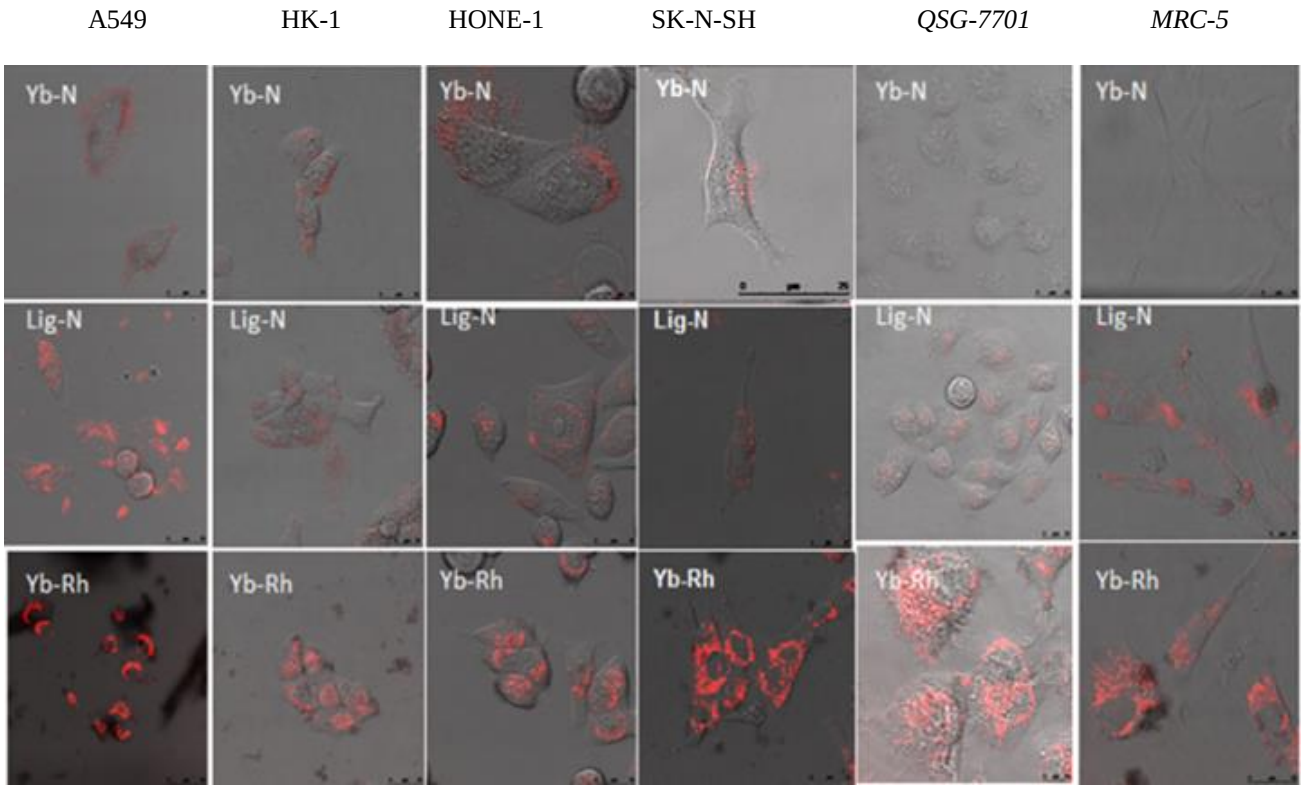


Figure S9. In vitro imaging of (upper) Yb-N, (middle) Lig-N and (lower) Yb-Rh in four human cancer cell lines- A549, HONE-1, HK-1, SK-N-SH and two normal human cell lines - QSG-7701 and MRC-5. (Incubation time = 3 hours, 1 μM, λ_{ex} = 430 nm)

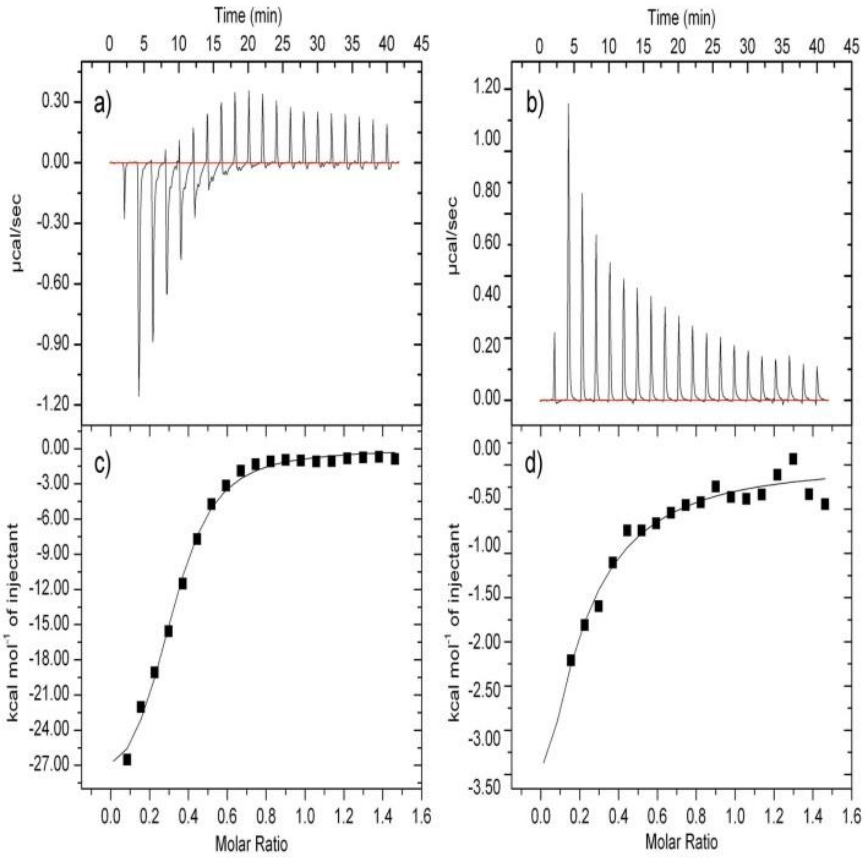


Figure S10. Isothermal calorimetric titrations of Yb-N titrated with DOPS or DOPC at 25 °C. Upper panels (a and b) are the raw data for titrations of DOPS or DOPC with Yb-N, and the lower panels (c and d) show the integrated heats of the binding corresponding to the raw data after subtracting the heat of dilution. Full lines represent the best fit using *one set of sites* model by MicroCal Origin 6.0.

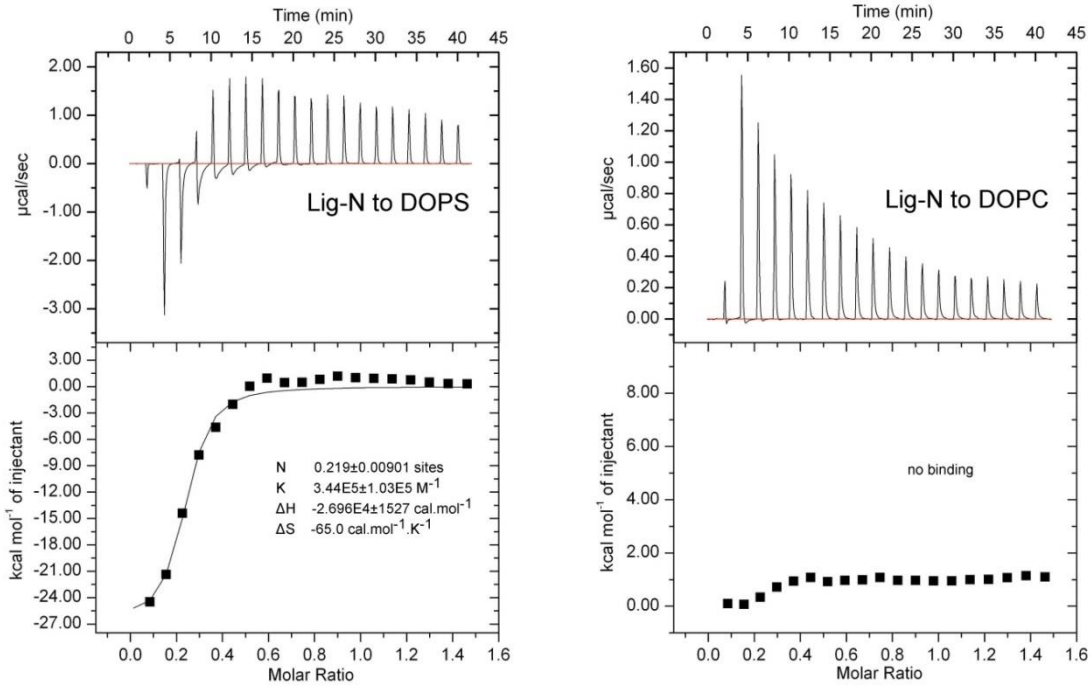


Figure S11 Isothermal titration calorimetry of **Lig-N** to DOPS or DOPC. **Lig-N** showed interaction with DOPS but not DOPC.

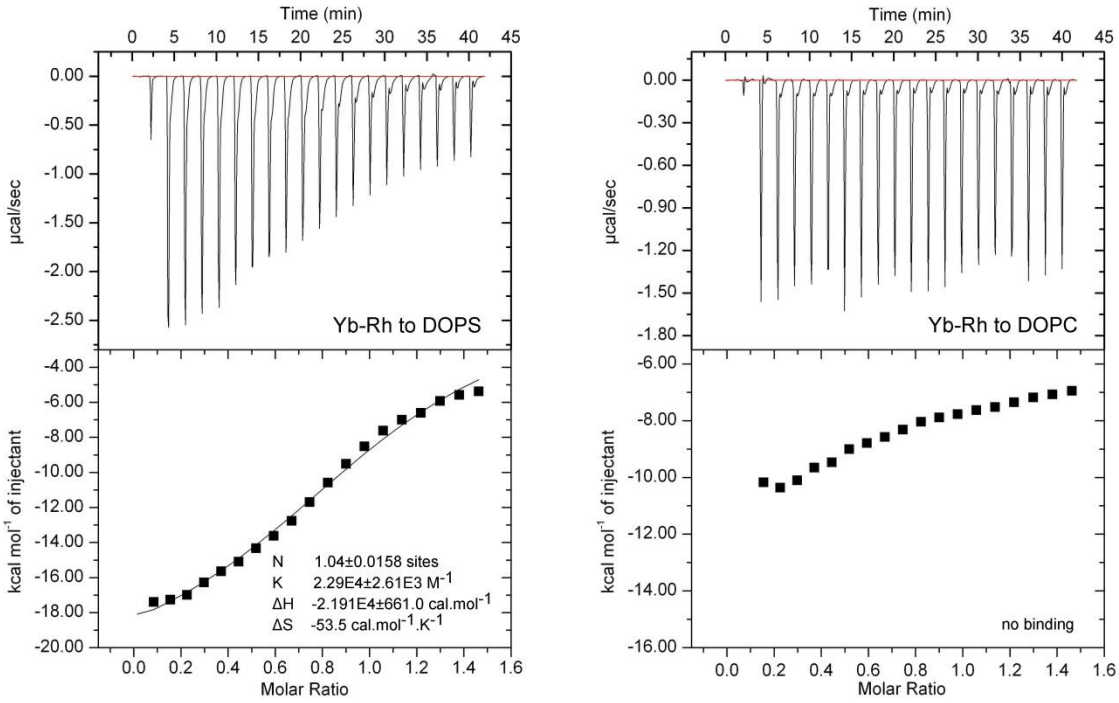


Figure S12 Isothermal titration calorimetry of **Yb-Rh** to DOPS or DOPC. **Yb-Rh** showed a slight interaction with DOPS but not DOPC. **Yb-Rh** (which specifically binds to mitochondria) is serving as control molecule in the experiments.

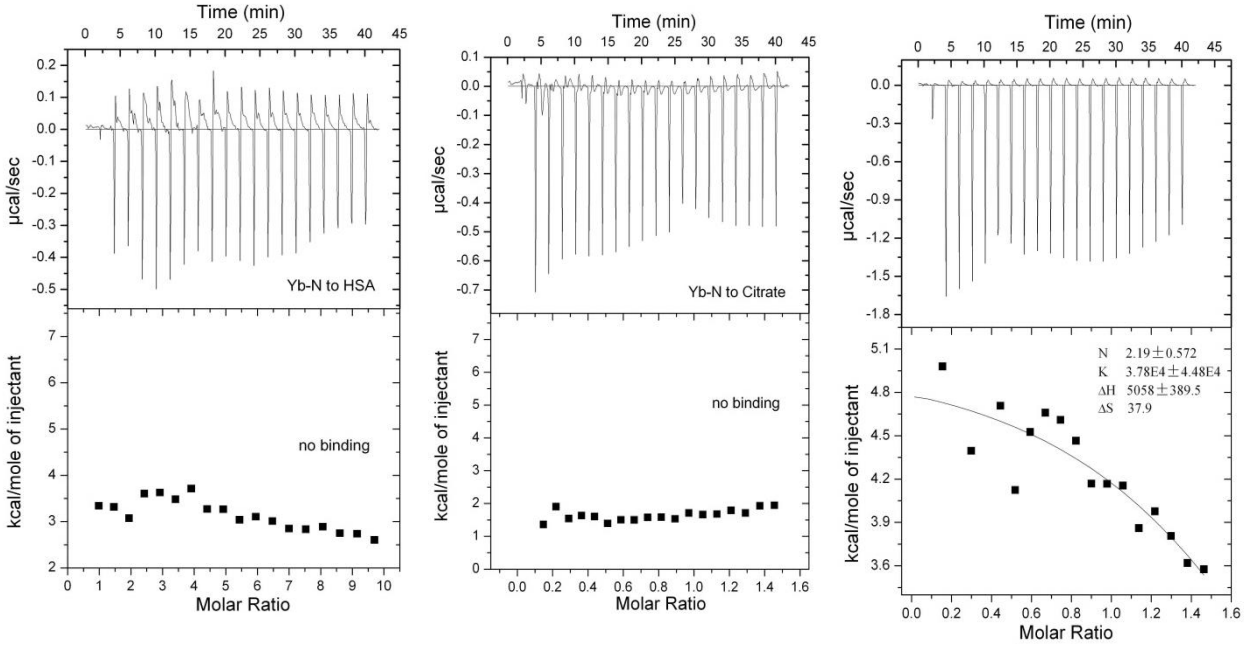


Figure S13 Selectivity assays: Isothermal titration calorimetry of **Yb-N** to HSA (left), Citrate (middle) and bicarbonate (right). **Yb-N** showed a slight interaction with bicarbonate but not citrate and HSA.

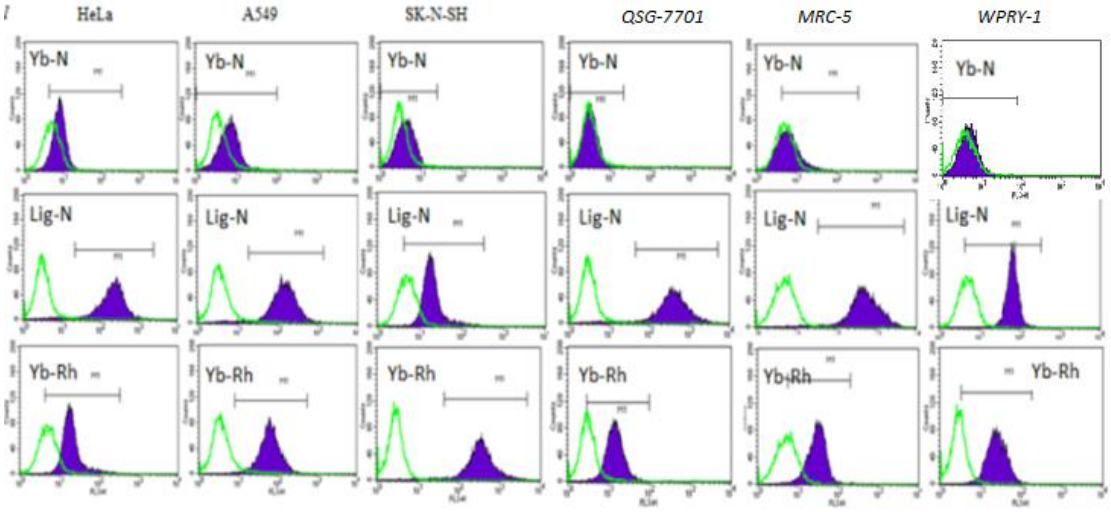


Figure S14. Cell permeability of (upper) **Yb-N**, (middle) **Lig-N**, (lower) **Yb-Rh** by flow cytometry in HeLa, A549, SK-M-SH, QSG-7702, MRC-5 and WPMY-1 with 6 hours incubation. (10 □M) – Part of data for figure 5

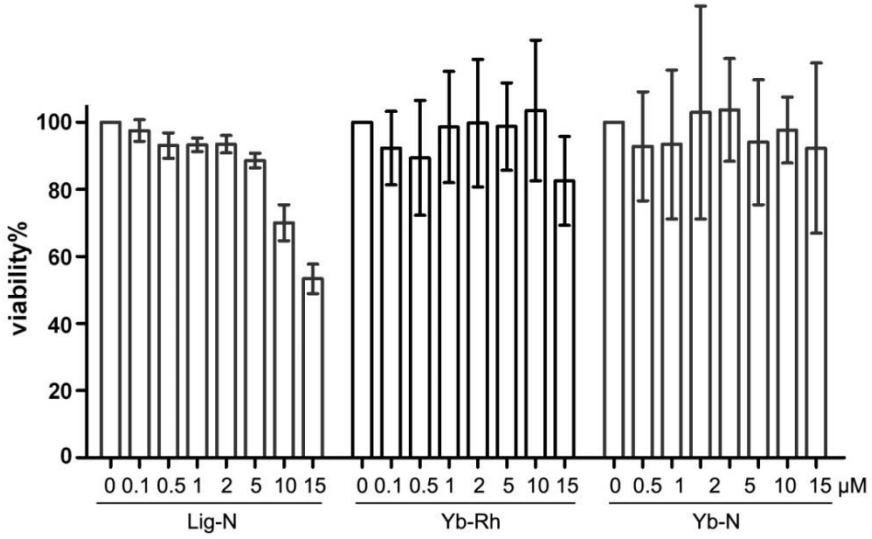


Figure S15. Cytotoxicity tests of **Yb-N**/**Lig-N**/**Yb-Rh** by MTT viability assay.

Reference:

1. (a) J. N. Demas, G. A. Crosby, *J. Phys. Chem.*, 1971, 75, 991; (b) G. E. Khalil, E. K. Thompson, M. Gouterman, J. B. Callis, L. R. Dalton, N. J. Turro, S. Jockusch, *Chem. Phys. Lett*., 2007, 435, 45; (c) T. Zhang, X. Zhu, C. C. W. Cheng, W. M. Kwok, H.-L. Tam, J. Hao, D. W. J. Kwong, W. K. Wong, K.-L. Wong, *J. Am. Chem. Soc*., 2011, 133, 20120.
2. R. Pal, L. C. Costello, D. Parker, *Org. Biomol. Chem*., 2009, 7, 1525.