Electronic Supplementary Material (ESI) for ChemComm.

Supporting information

Fluorimetric sensing of ATP in water by Imidazolium hydrazone based sensor

Sepideh Farshbaf [a], Pavel Anzenbacher, Jr[*a].

[a] Department of Chemistry, Bowling Green State University, Bowling Green, OH 43403, USA
E-mail: pavel@bgsu.edu

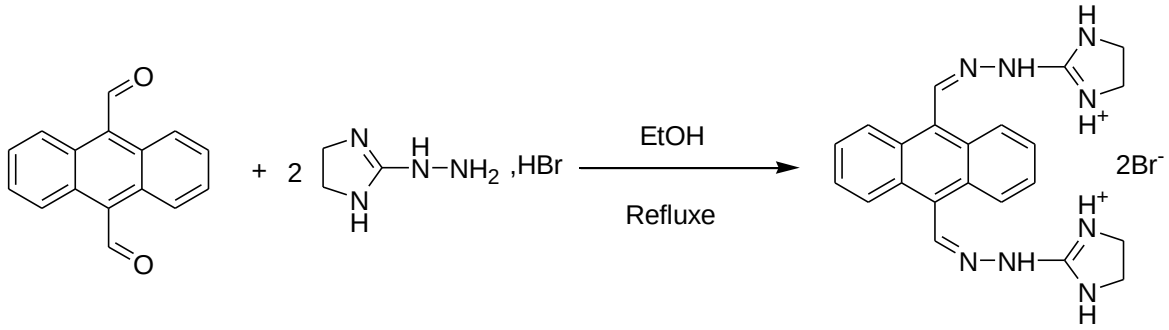
Table of Contents

1. General

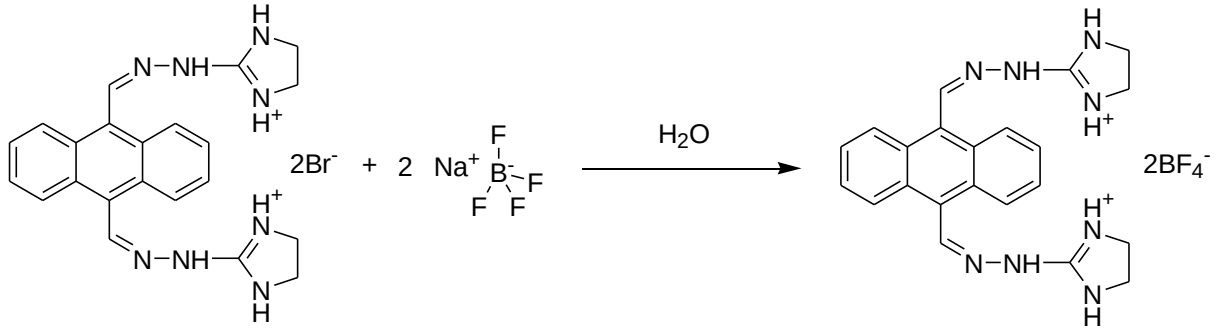
Starting materials were purchased from commercial suppliers were used without further purification. Bisantrene, 9,10-Anthracenedicarboxaldehyde, bis[(4,5-dihydro-1H-imidazol-2-yl) hydrazone], was synthesized according to the literature procedure.[1] Standard laboratory techniques were performed in the synthesis. All chemicals were analytical grade and they were used without purification. All NMR spectra were measured on a Bruker Avance 500 spectrometer operating at 500 MHz. Mass spectrometry study was performed using Shimadzu AXIMA Performance MALDI TOF mass spectrometer and Bruker Daltonics OmniFlex Maldi TOF mass spectrometer. Absorption spectra were recorded using a Hitachi U-3010 spectrophotometer. Fluorescence emission spectra were acquired using an Edinburgh single photon counting spectrofluorimeter (FLSP 920). The absolute quantum yields were measured using a Hamamatsu Quantaurus absolute quantum yield spectrometer QY-C11347. All the optical measurements were performed using a quartz cuvette with a path length of 1 cm at room temperature.

2. Synthesis of bisantrene

The synthesis of the bisantrene was reported. In order to synthesize bisantrene, a mixture of 25 mg (0.107 mmol) of 9, 10-anthracenedicarboxaldehyde, 48.30 mg (0.267 mmol) of 2-hydrazino-2-imidazoline hydrobromide and 20 mL of ethanol was heated under reflux. Then, 8 µL of concentrated HCl was added and the mixture was stirred during 2 hours.



After 2 hours the temperature was cooled down until orange crystals were formed. The crystals were filtered and washed with cold ethanol. The mobile phase used for analytical TLC (alumina plate) was $CHCl_3$: MeOH: 95:5. The orange product was recrystallized with EtOH and 30 mg, (50%) of product was obtained. The pure compound was dissolved in water and added into saturated solution of $NaBF_4$ in water. The precipitate was filtered and dried in vacuum.



3. Characterization (NMR and MS Spectra)

^{1}H NMR (500 MHz, DMSO-d_6) δ = 12.52 (s, 1H), 9.32 (s, 1H), 8.73 (s, 2H), 8.48 (dd, J=3.28, 6.52, 2H), 7.71 (dd, J= 3.23, 6.89, 2H), 3.75 (s, 4H) ppm.

^{13}C NMR (500 MHz, DMSO-d_6) δ = 158.2, 148.4, 129.5, 128.2, 127.6, 125.9, 56.3, 43.3 ppm.

MS (ESI) m/z: 399.30 $[M + H]^+$.

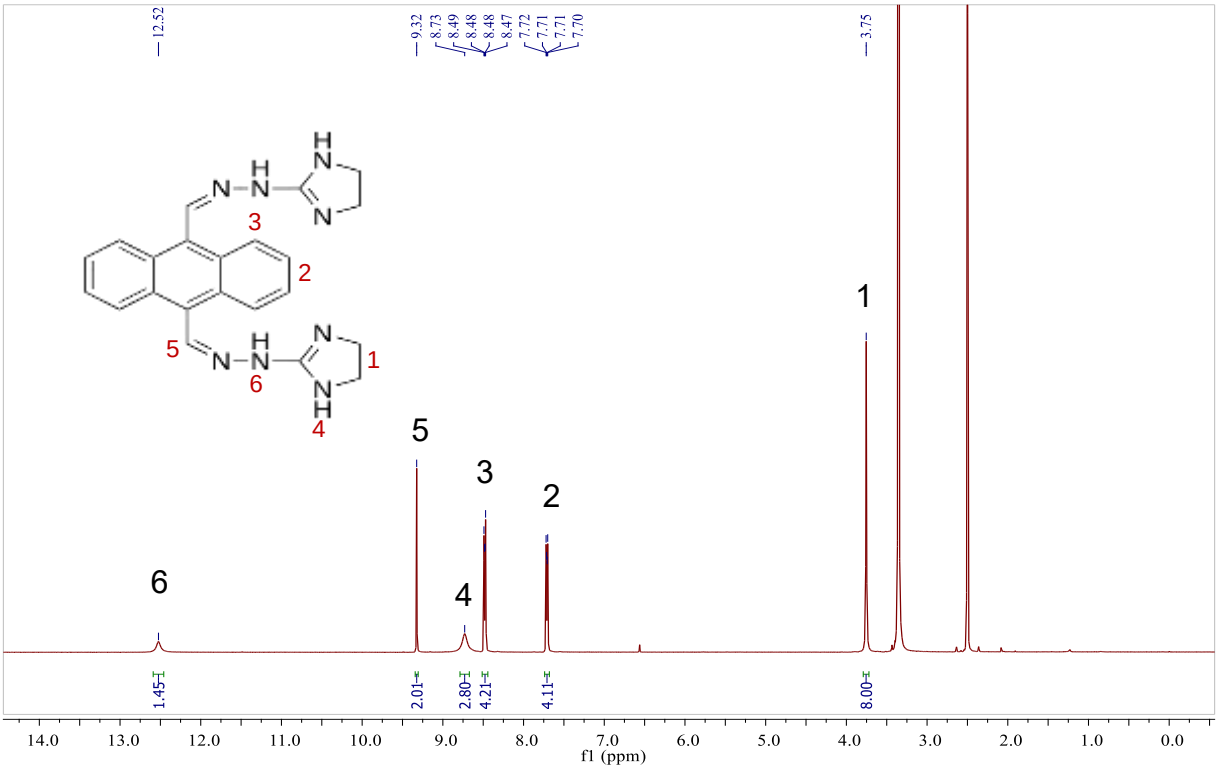


Figure 1: ^{1}H NMR spectra of bisantrene in DMSO-d_6 at 500MHz.

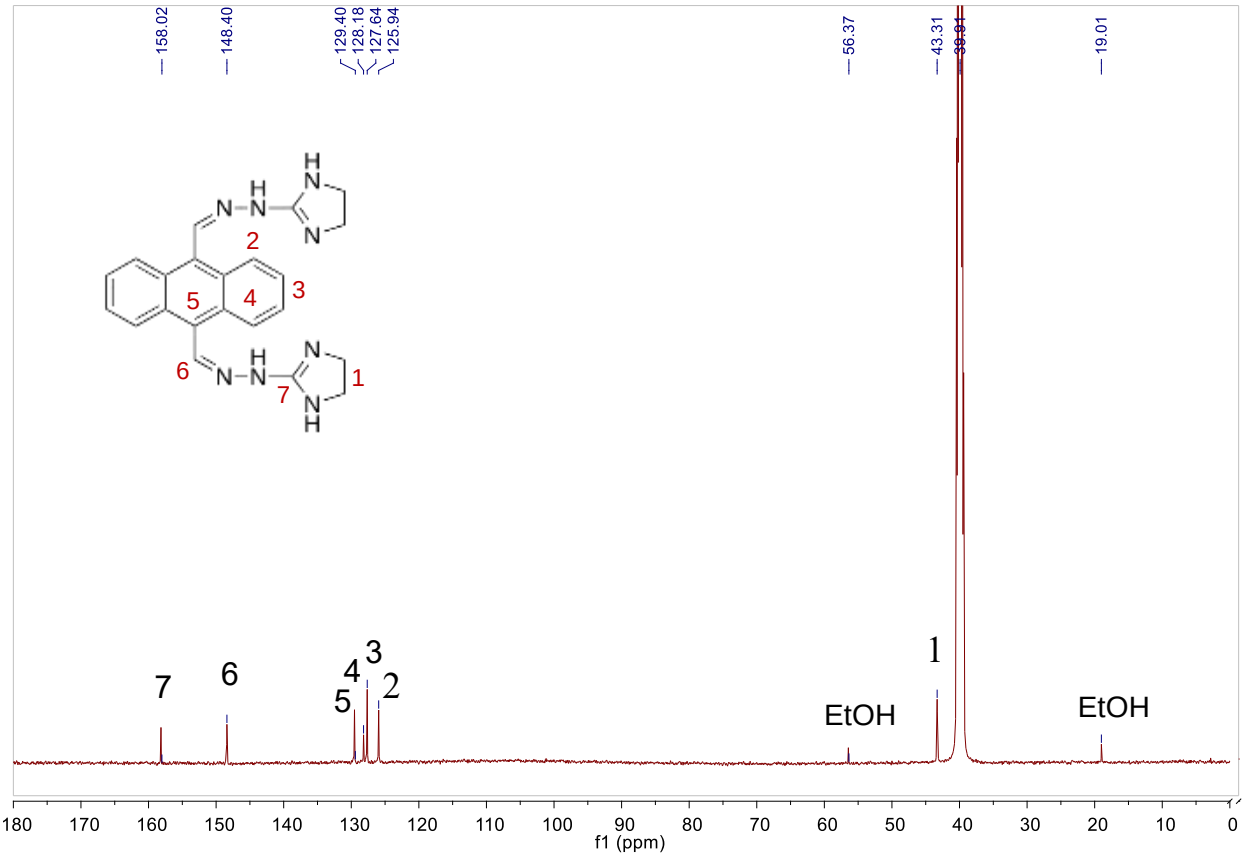


Figure 2: ^{13}C NMR, CPD, spectra of bisantrene in DMSO-d_6 at 500MHz.

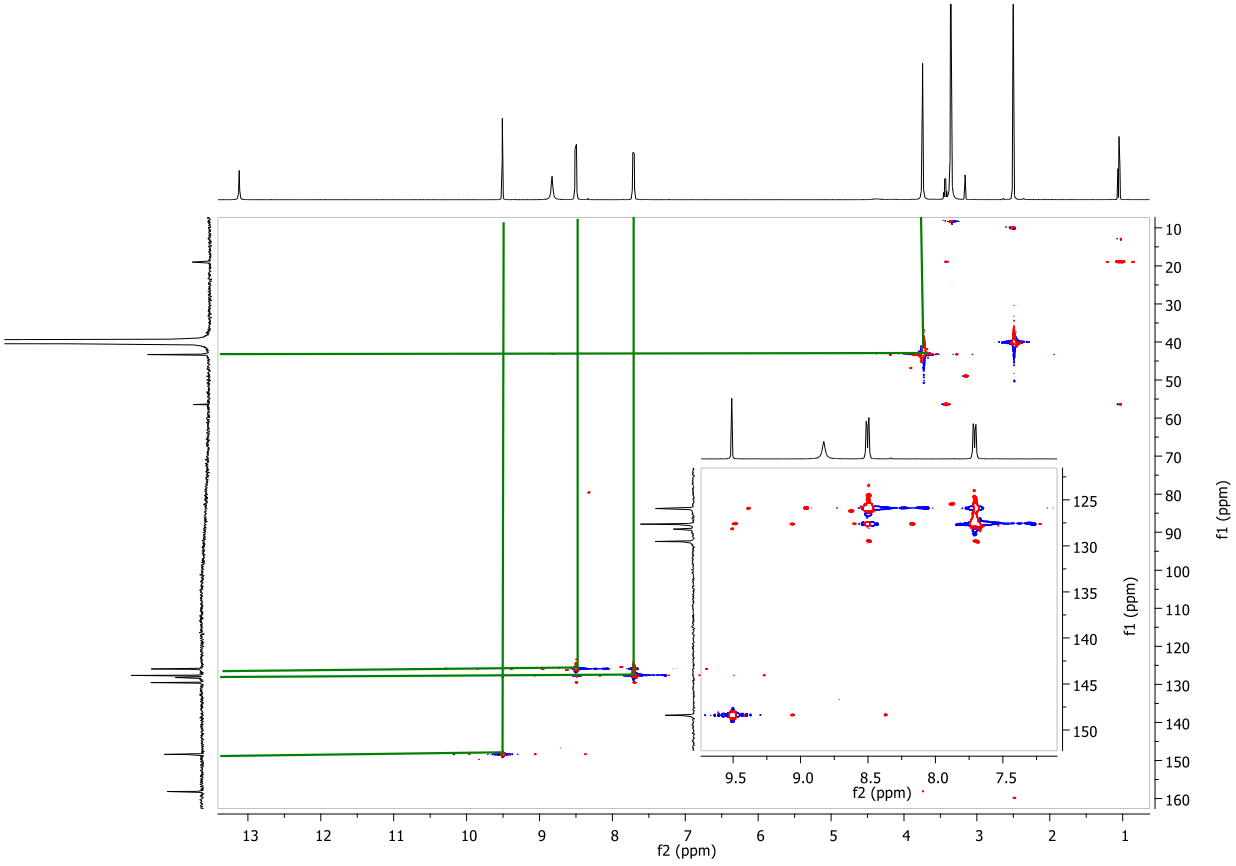


Figure 3: ^{2}D NMR,HSQC, spectra of bisantrene in DMSO-d_6 at 500MHz.

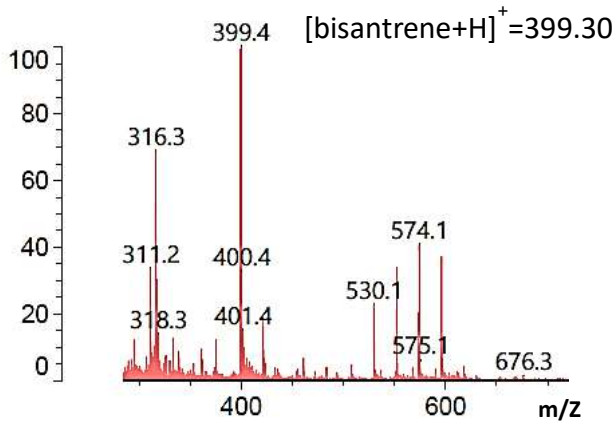


Figure 4: Mass spectra of bisantrene in MeOH.

4. Photophysical properties

Solvent	Absolute quantum yield (%)	Life time (ns)
10mM Tris : DMSO 90:10 (pH:7)	2.26	τ1 : 2.26 τ2 : 13.29
CH_3CN	4.16	τ1 : 1.95 τ2 : 10.45

5. UV-Vis titrations in Tris: DMSO 90:10, pH: 7

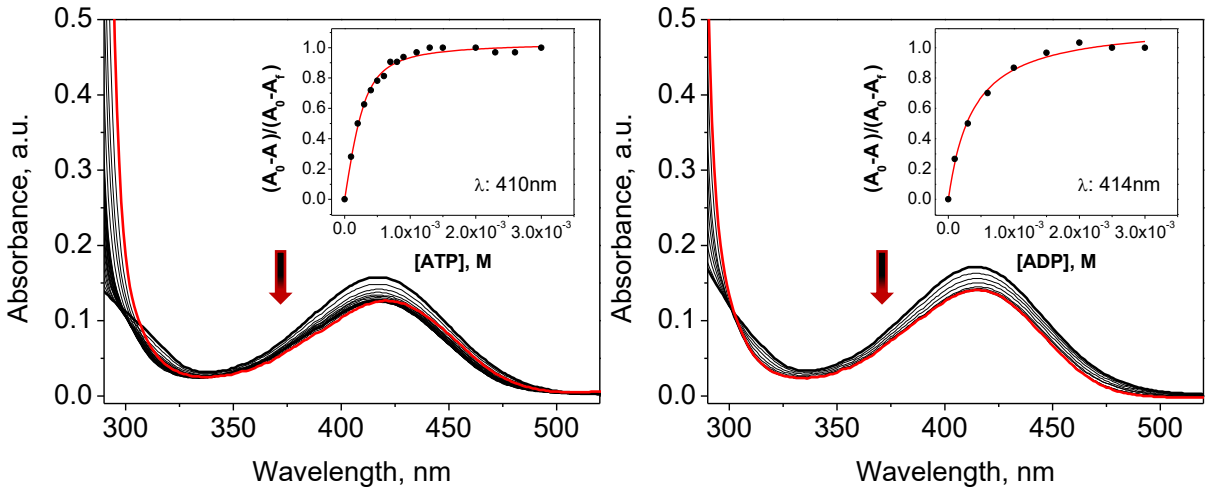


Figure 5: UV-Vis titrations of sensor (20 µM) upon the addition of disodium salts of ATP (left) and ADP (right) in Tris: DMSO 90:10, pH: 7. [ATP]: 0-3mM, [ADP]: 0-3 mM.

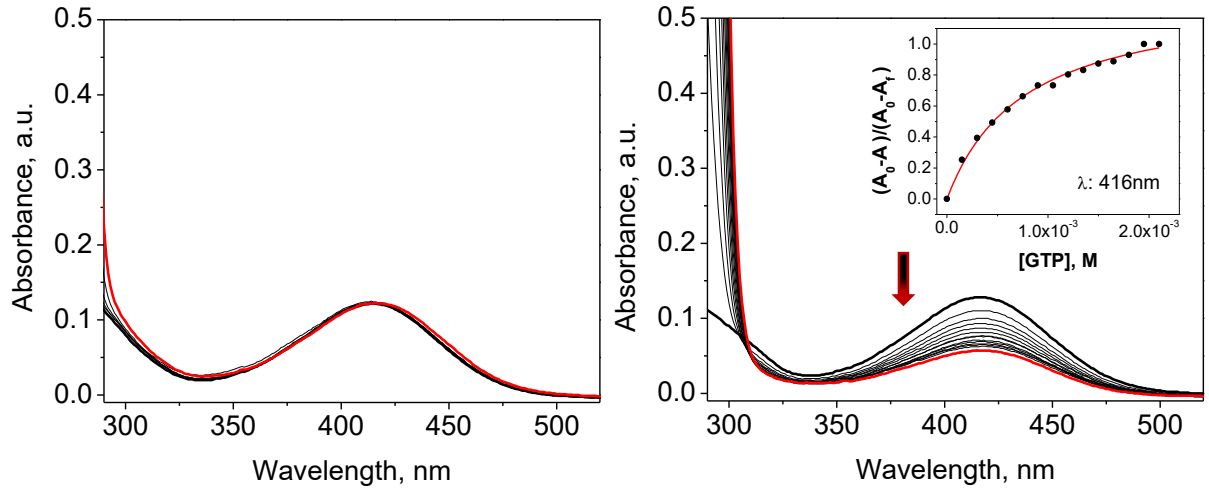


Figure 6: UV-Vis titrations of sensor (20 µM) upon the addition of disodium salts of AMP (left) and GTP (right) in Tris: DMSO 90:10, pH: 7. [AMP]: 0-1.5 mM. [GTP]: 0-2.1 mM.

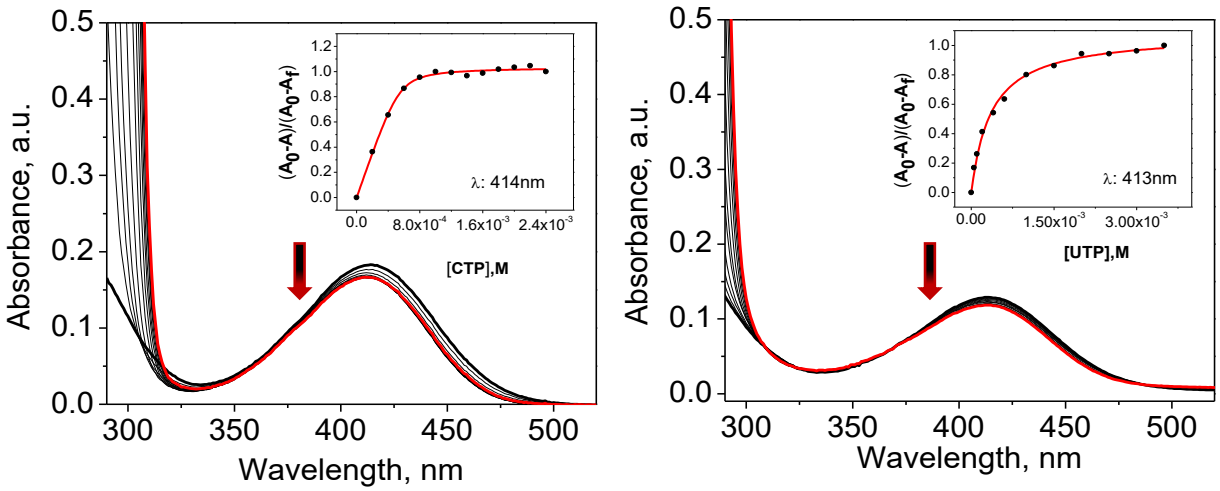


Figure 7: UV-Vis titrations of sensor (20 µM) upon the addition of disodium salts of CTP (left) and trisodium salts of UTP (right) in Tris: DMSO 90:10, pH: 7. [CTP]: 0-2.4 mM. [UTP]: 0-3.5 mM.

6. UV-Vis titrations in acetonitrile

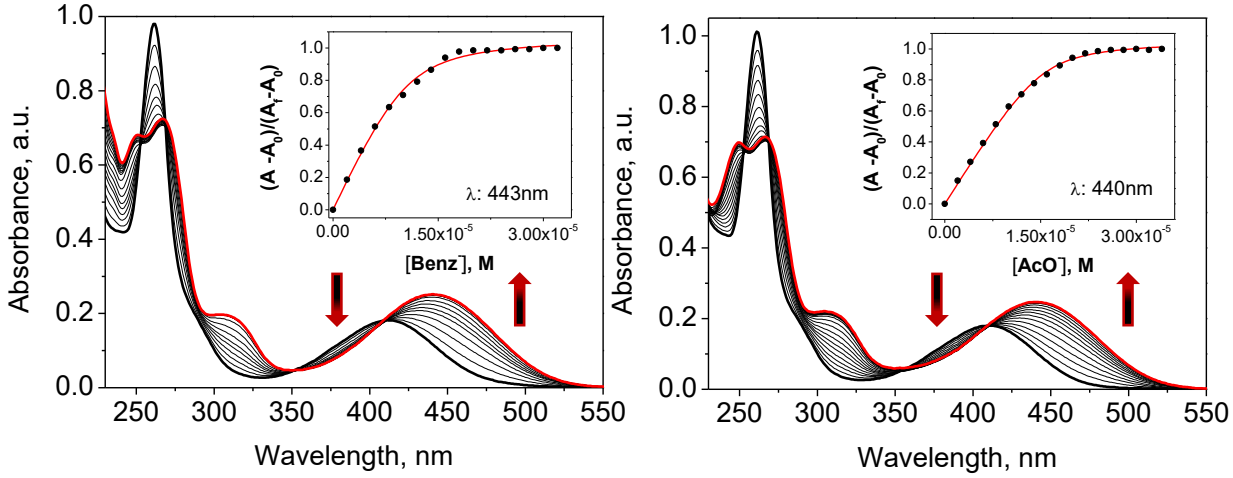


Figure 8: UV-Vis titrations of sensor (20 µM) upon the addition of tetrabutylammonium salts of benzoate (left) and acetate (right) in acetonitrile. [$Benz^-$]: 0-32 µM, [AcO^-]: 0-34 µM.

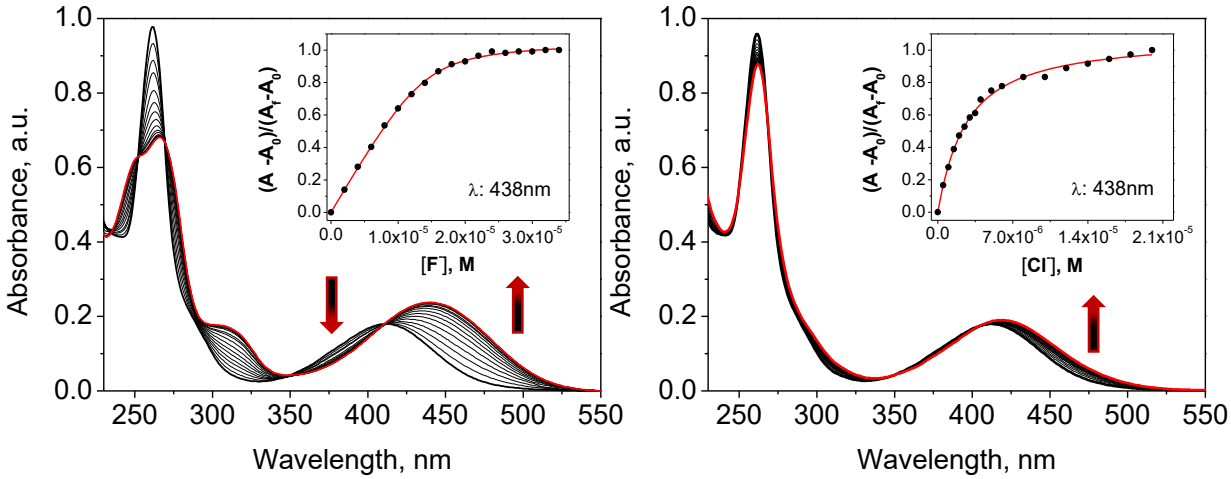


Figure 9: UV-Vis titrations of sensor (20 µM) upon the addition of tetrabutylammonium salts of fluoride (left) and Chloride (right) in acetonitrile. [F^-]: 0-34 µM, [Cl^-]: 0-20 µM.

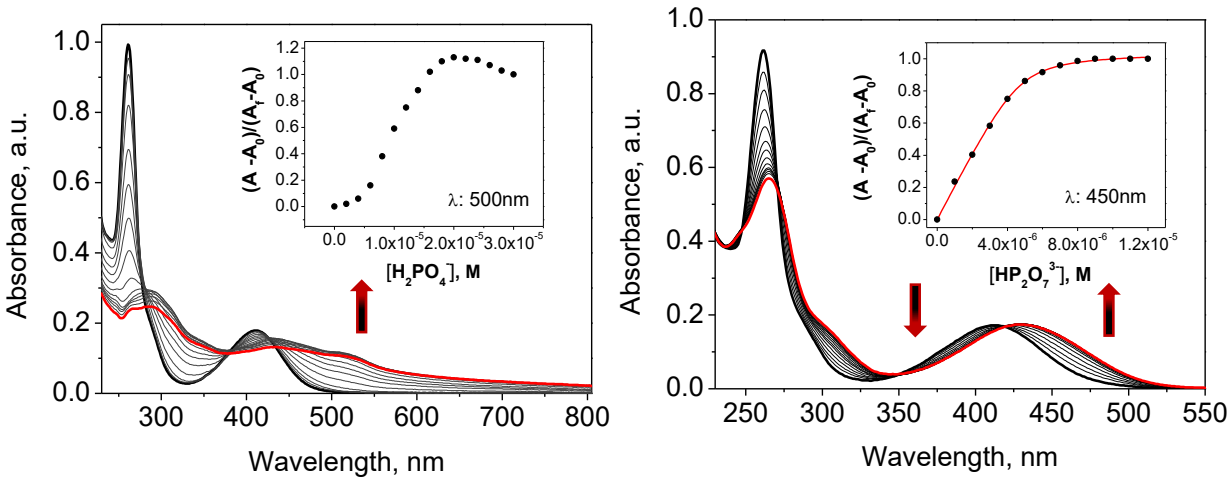


Figure 10: UV-Vis titrations of sensor (20 µM) upon the addition of tetrabutylammonium salts of phosphate (left) and pyrophosphate (right) in acetonitrile. [$H_2PO_4^-$]: 0-28 µM, [PPi]: 0-12 µM.

7. Fluorescence Titrations in Tris: DMSO 90:10, pH: 7

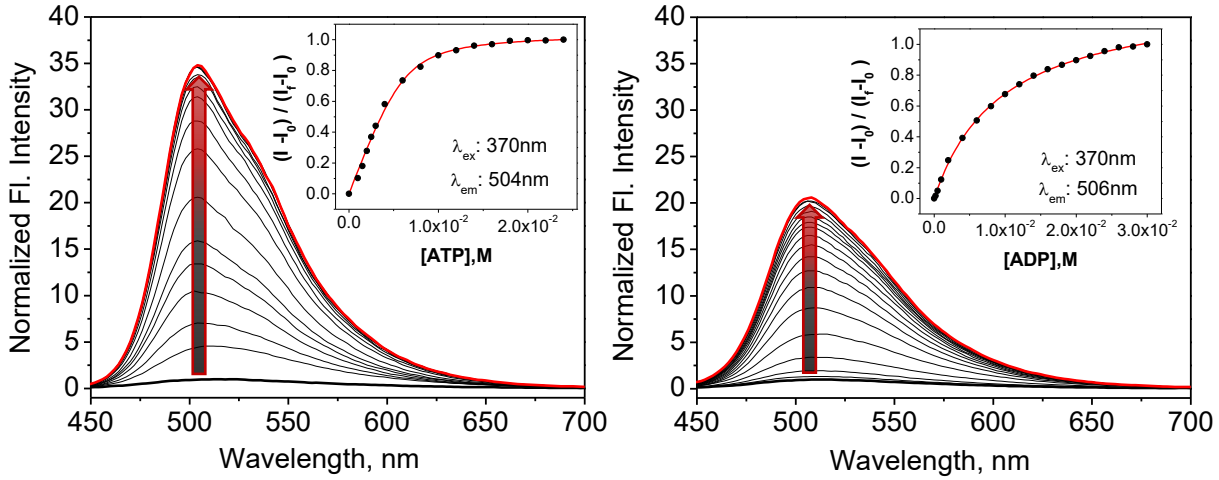


Figure 11: Fluorescence titrations of sensor (20 µM) upon the addition of disodium salts of ATP (left) and ADP (right) in Tris: DMSO 90:10, pH: 7. [ATP]: 0-24 mM, [ADP]: 0-30 mM.

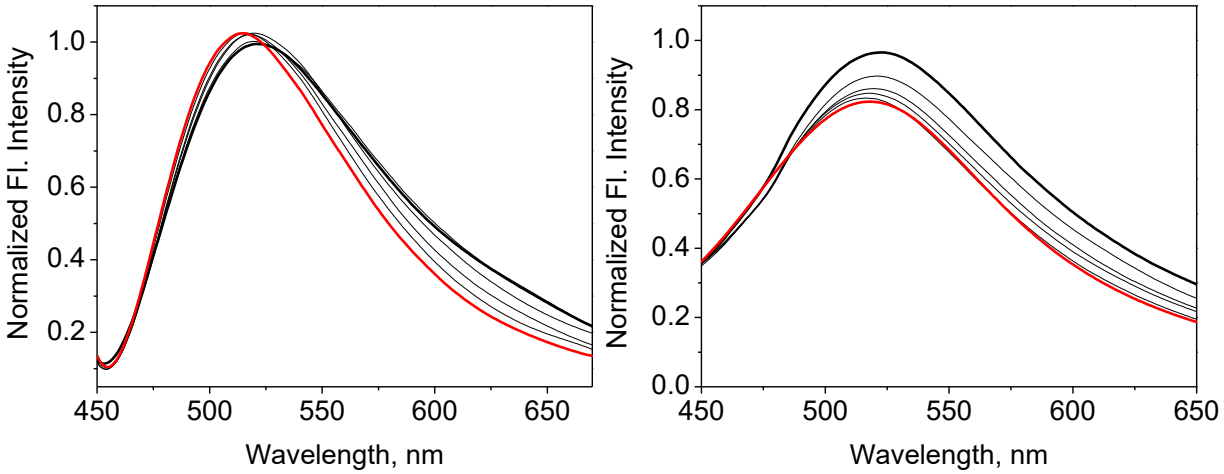


Figure 12: Fluorescence titrations of sensor (10 µM) upon the addition of disodium salts of AMP (left) and GTP (right) in Tris: DMSO 90:10, pH: 7. [AMP]: 0-7 mM, λ_{ex}: 417 nm, [GTP]: 0-3 mM, λ_{ex}: 370 nm.

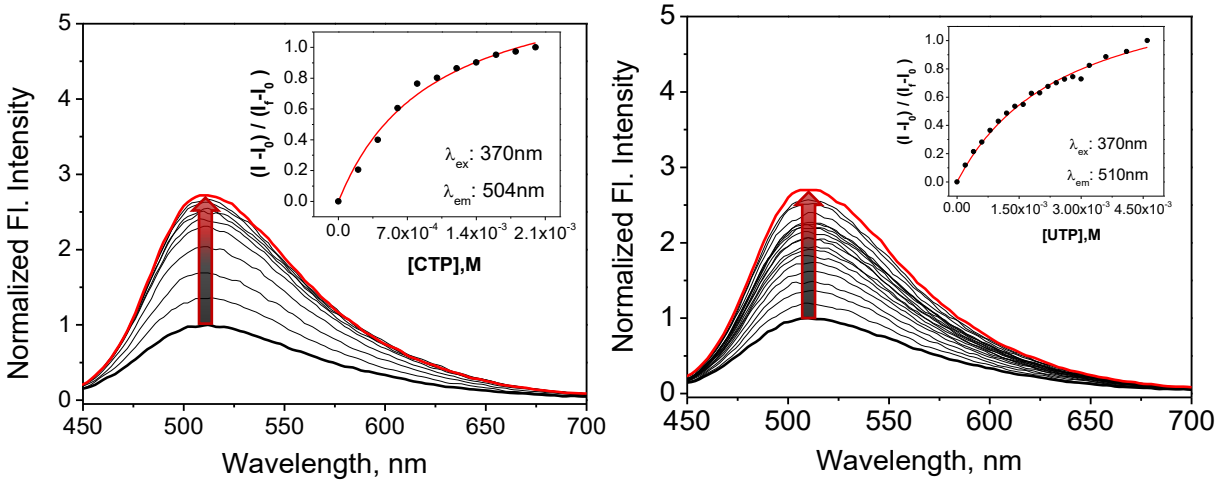


Figure 13: Fluorescence titrations of sensor (20 µM) upon the addition of disodium salts of CTP (left) and UTP (right) in Tris: DMSO 90:10, pH: 7. [CTP]: 0-2 mM, [UTP]: 0-4.6 mM.

8. Fluorescence Titrations in acetonitrile

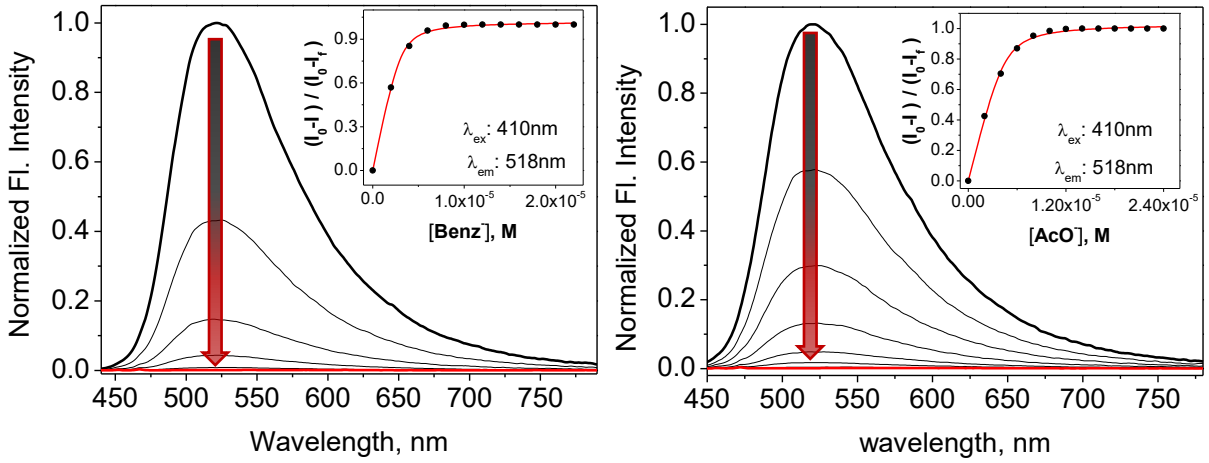


Figure 14: Fluorescence titrations of sensor (20 µM) upon the addition of tetrabutylammonium salts of benzoate (left) and acetate (right) in acetonitrile. [Benzoate]: 0-22 µM, [Acetate] 0-24 µM.

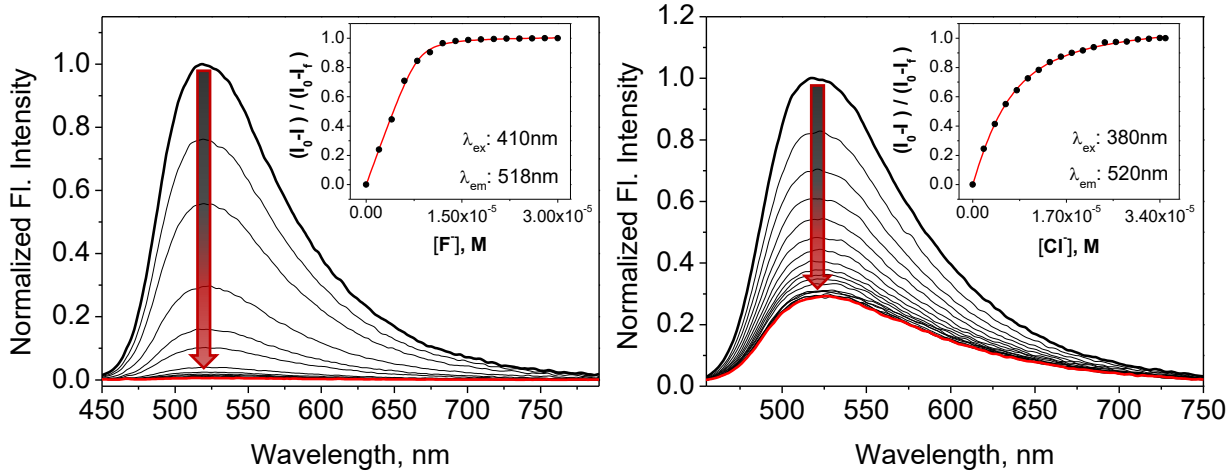


Figure 15: Fluorescence titrations of sensor (20 µM) upon the addition of tetrabutylammonium salts of fluoride (left) and chloride (right) in acetonitrile. [F^-]: 0-30 µM, [Cl^-]: 0-35 µM.

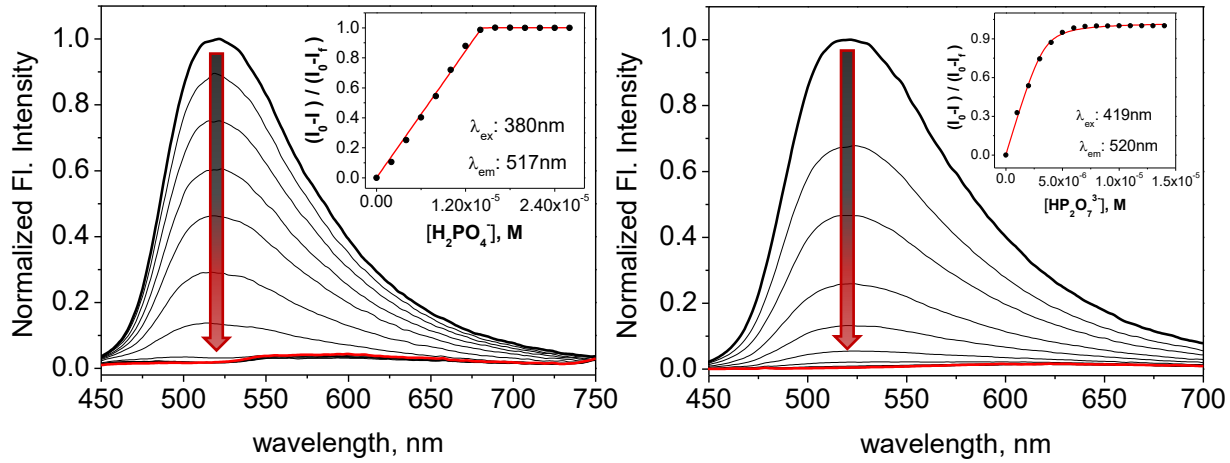


Figure 16: Fluorescence titrations of sensor (20 µM) upon the addition of tetrabutylammonium salts of phosphate (left) and pyrophosphate (right) in acetonitrile. [$H_2PO_4^-$]: 0-26 µM, [PPi]: 0-14 µM.

9. Sensor-ATP complex study

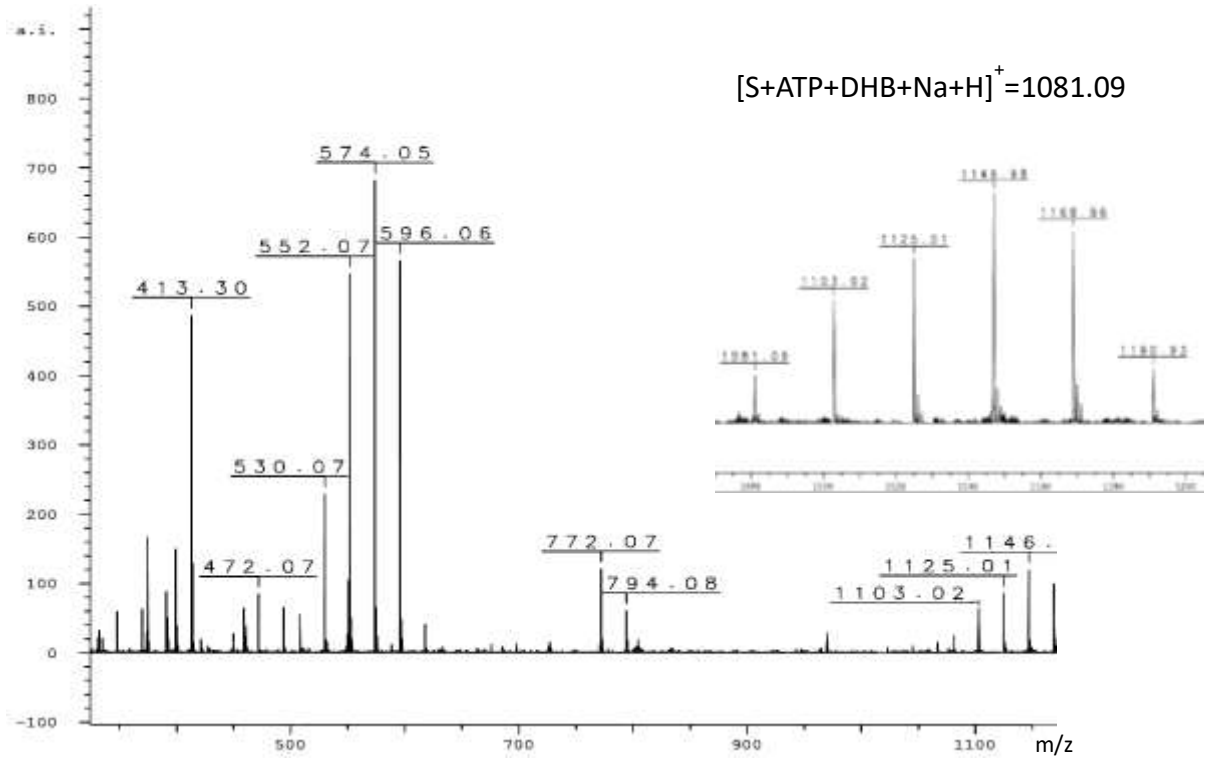


Figure 17: MALDI-TOF mass spectrum of the complex of bisantrene-ATP in Tris: DMSO 90:10, pH: 7 (DHB=dihyroxybenzoic acid).

10. Job Plot experiments

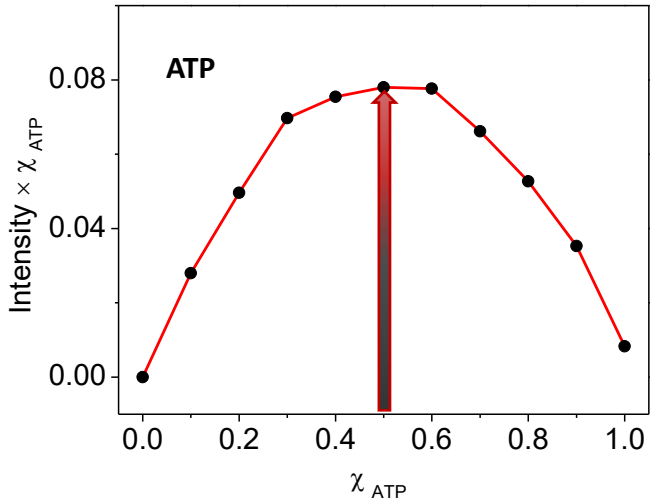


Figure 18: The Job plot of UV-Vis titration of the sensor upon the addition of ATP in Tris: DMSO 90:10, pH: 7. (Result: 1:1 binding)

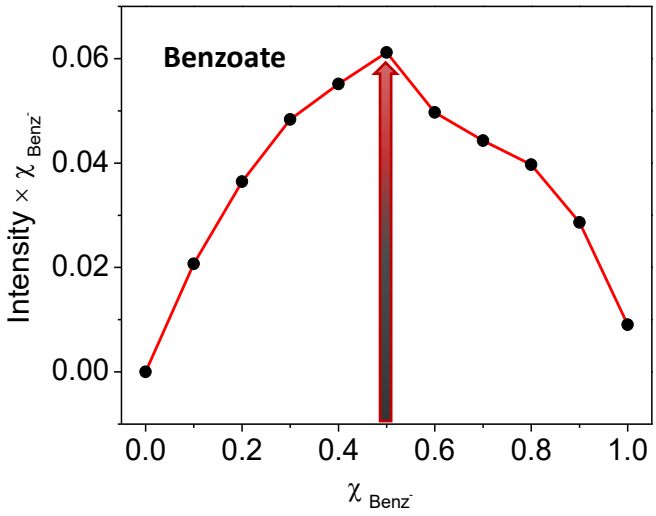


Figure 19: The Job plot of UV-Vis titration of the sensor upon the addition of benzoate in acetonitrile. (Result: 1:1 binding)

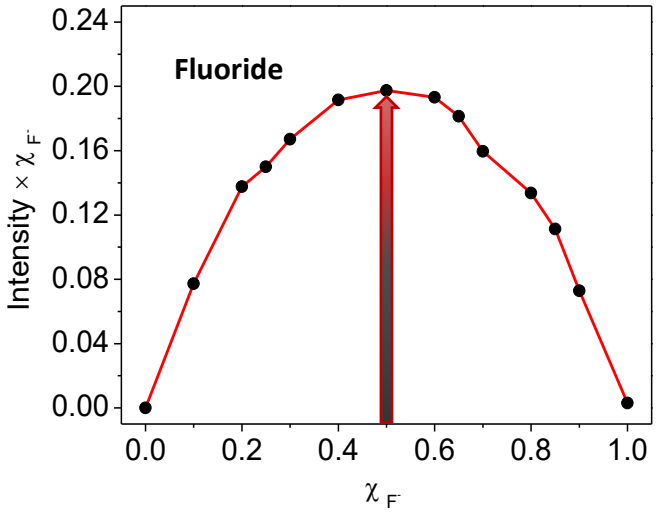


Figure 20: The Job plot of UV-Vis titration of the sensor upon the addition of fluoride in acetonitrile. (Result: 1:1 binding)

11. References

1. K. C. Murdock, R. G. Child, Y. I. Lin, J. D. Warren, P. F. Fabio, V. J. Lee, P. T. Izzo, S. A. Lang and R. B. Angier, *J. Med. Chem.*, 1982, **25**, 505–518.