Electronic Supplementary Material (ESI) for RSC Advances.

Highly Promising Discrimination of Various Catecholamine by Ratiometric Fluorescence under Intermolecular Self-Association of Two Sensing Elements

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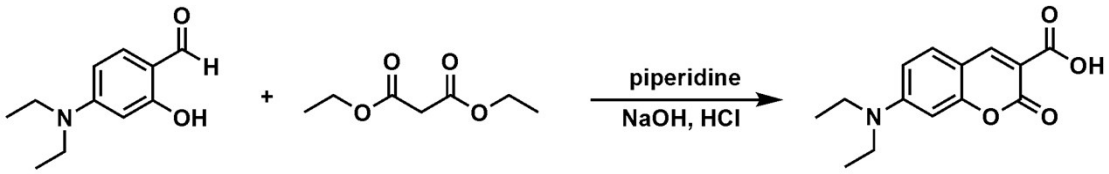
Supplementary data

(16 pages)

1. Synthesis of crown ether/coumarin based sensor

*1.1 Preparation of 7-diethylamino-2-oxo-2H-chromen-3-carboxylic acid (**1**)*



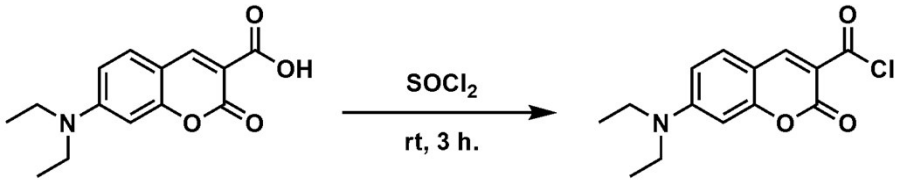
1

Scheme S1 Synthesis of compound **1**

Into a two-neck round bottom flask equipped with a magnetic bar, 4-diethylaminosalicylaldehyde (0.386 g, 2 mmol), diethylmalonate (0.61 mL, 4 mmol), and piperidine (0.2 mL) were dissolved in absolute ethanol (6 mL). The reaction mixture was stirred and refluxed for 6 hours under nitrogen atmosphere. Then 10% NaOH (6 mL) solution was added to the reaction and refluxed for 15 min. The solution was cooled to room temperature and acidified to pH 2 using concentrated hydrochloric acid at 0ºC affording a precipitate deposit which was filtered out, washed with water, then recrystallized with ethanol to give an orange crystal (**1**, 74%). ^{1}H-NMR (400 MHz, $CDCl_3$): δ (in ppm) = 12.37 (s, 1H, COO*H*), 8.67 (s, 1H, Ar*H*), 7.46 (d, *J* = 9.2 Hz, 1H, Ar*H*), 6.71 (dd, *J* = 4.27 Hz, 1H, Ar*H*), 6.53 (d, *J* = 2.0 Hz, 1H, Ar*H*), 3.49 (q, *J* = 7.07 Hz, 4*H*, CH_2CH_3), 1.26 (t, *J* = 7.2 Hz, 6H, CH_2CH_3). ^{13}C-NMR (400 MHz, $CDCl_3$): δ (in ppm) = 165.52, 164.42, 158.07, 153.78, 150.27, 131.94, 110.91, 108.59, 105.70, 96.89, 45.34, 12.39.

MALDI-TOF mass: Anal. Calcd for $[C_{14}H_{15}NO_4]^+$ m/z = 261.10
Found m/z = 261.32

*1.2 Preparation of 7-diethylamino-2-oxo-2H-chromen-3-carboxylic chloride (**2**)*



1 **2**

Scheme S2 Synthesis of compound **2**

Into a two-neck round bottom flask equipped with a magnetic bar**, 1** (0.187 g, 0.7 mmol) was added to dry thionyl chloride (3 mL) and the suspension was stirred at room temperature for 3 hours under nitrogen atmosphere. The precipitate was washed with dichloromethane under vacuum to give a yellow solid **2** in a quantitative yield.

^{1}H-NMR (400 MHz, $CDCl_3$): δ (in ppm) = 8.68 (s, 1H, Ar*H*), 7.49 (d, *J* = 4.4 Hz, 1H, Ar*H*), 6.76 (m, 1H, Ar*H*), 6.59 (s, 1H, CC*H*), 3.50 (q, *J* = 6.9 Hz, 4H, C*H*$_2$CH$_3$), 1.28 (t, *J* = 3.4 Hz, 6H, CH$_2$C*H*$_3$)

2. Characterization of the compounds

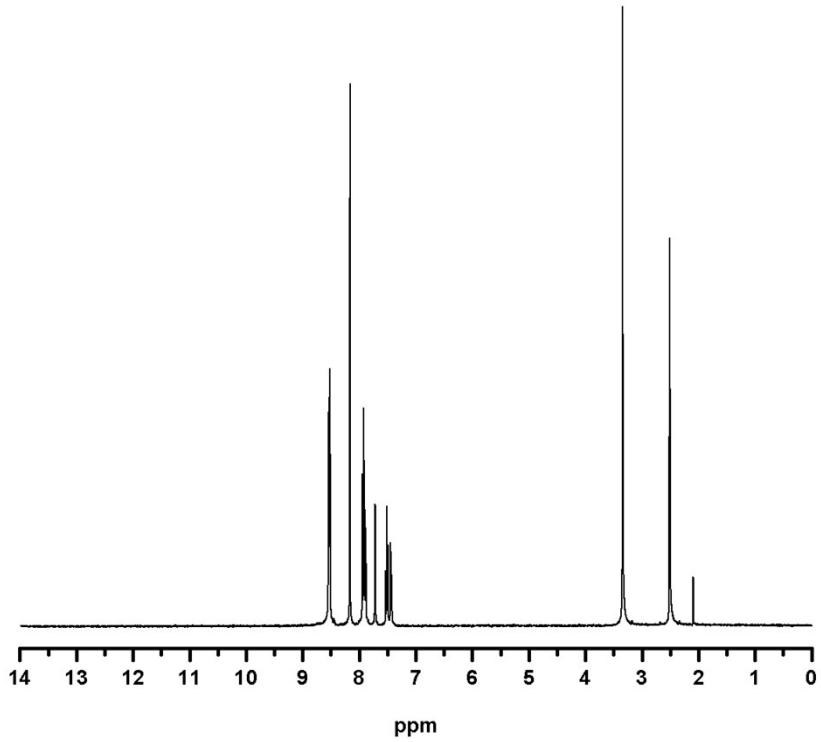


Figure S1 The ^{1}H-NMR spectrum of sensor **NB** in DMSO-d_6 at 400 MHz

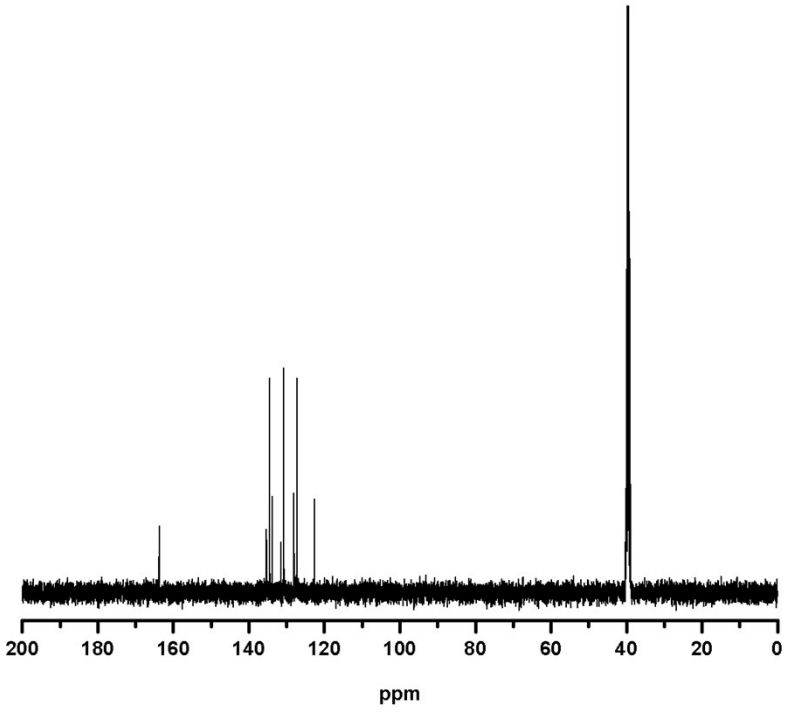


Figure S2 The ^{13}C-NMR spectrum of sensor **NB** in DMSO-d_6 at 400 MHz

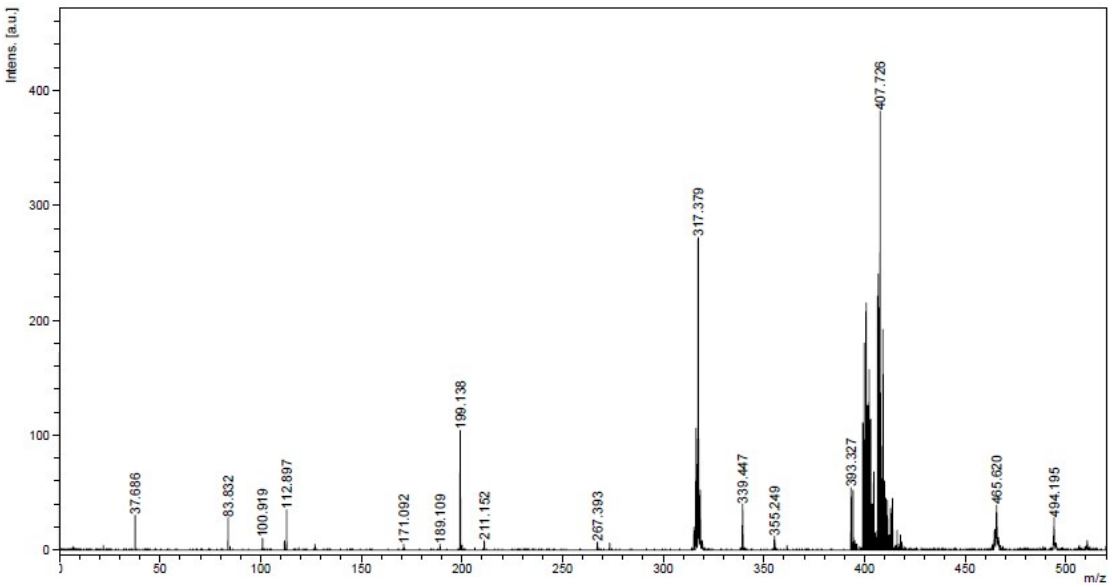


Figure S3 MALDI-TOF mass spectrum of sensor **NB** shown at 317.379 m/z

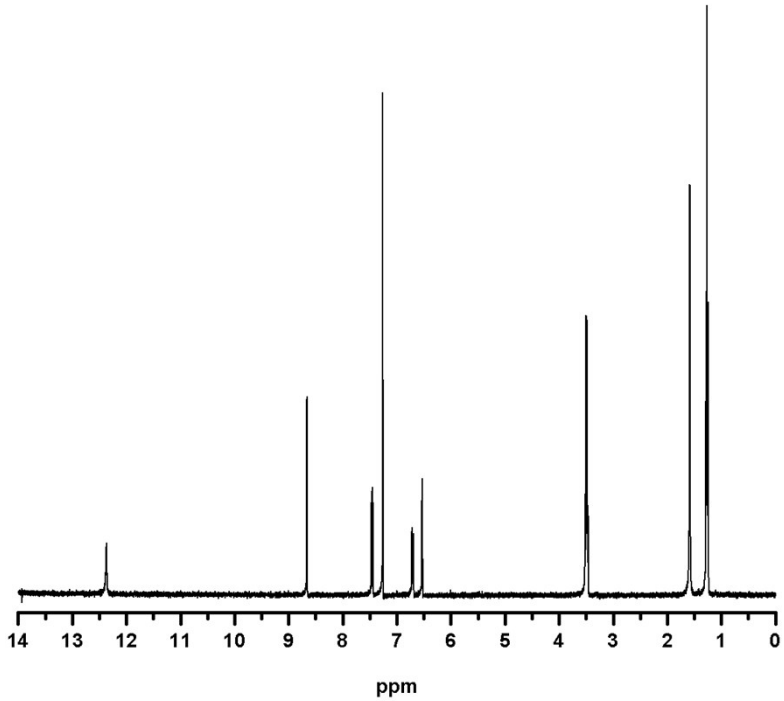


Figure S4 The ^{1}H-NMR spectrum of **1** in $CDCl_3$ at 400 MHz

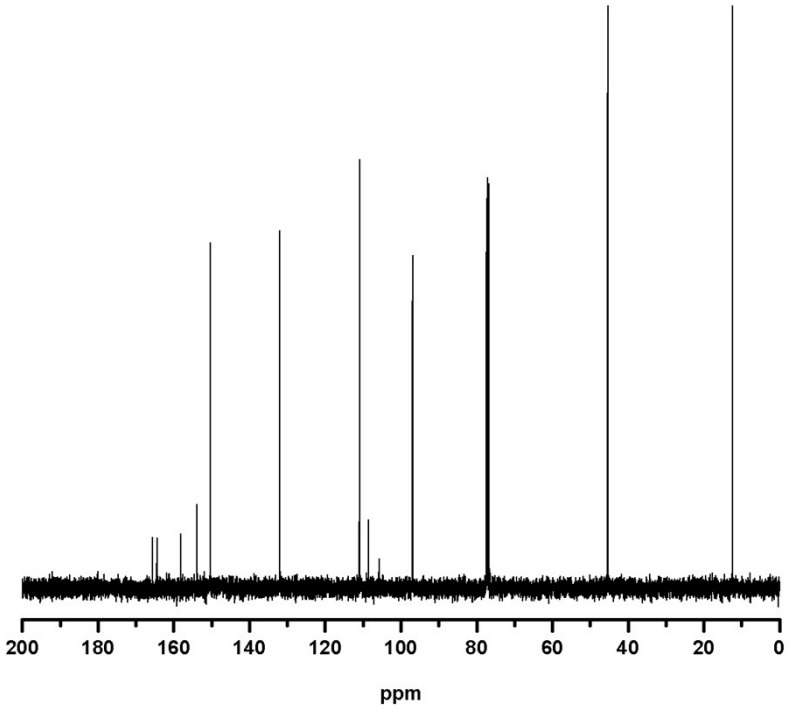


Figure S5 The ^{13}C-NMR spectrum of **1** in $CDCl_3$ at 400 MHz

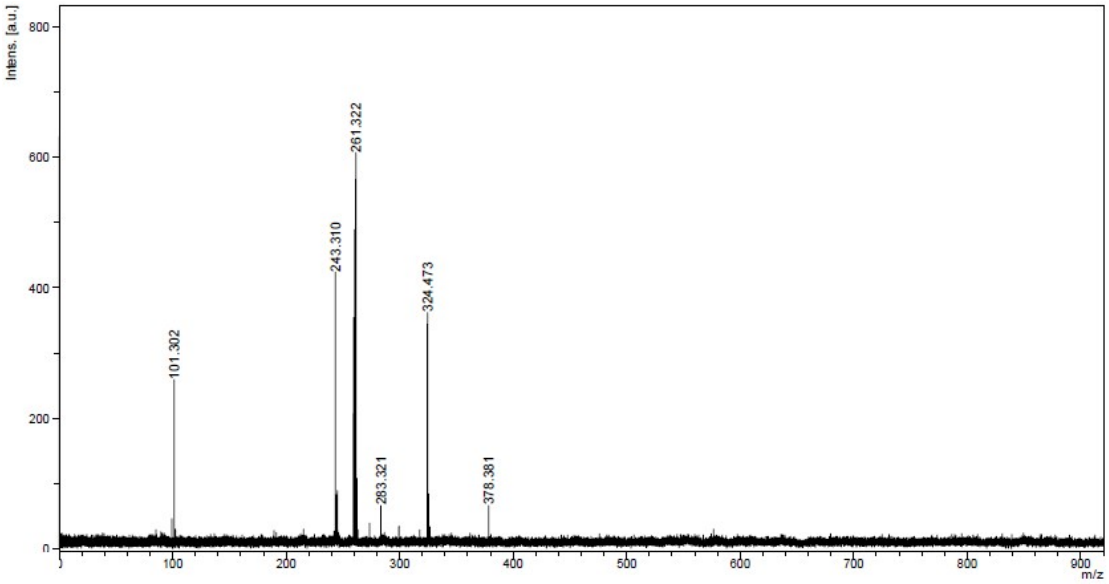


Figure S6 MALDI-TOF mass spectrum of **1** shown at 261.322m/z

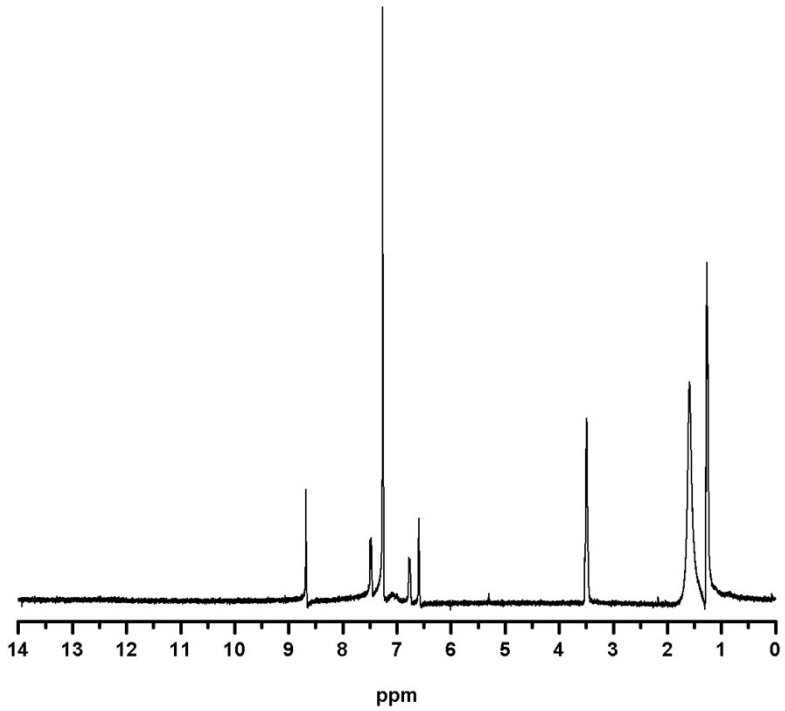


Figure S7 The ^{1}H-NMR spectrum of **2** in $CDCl_3$ at 400 MHz

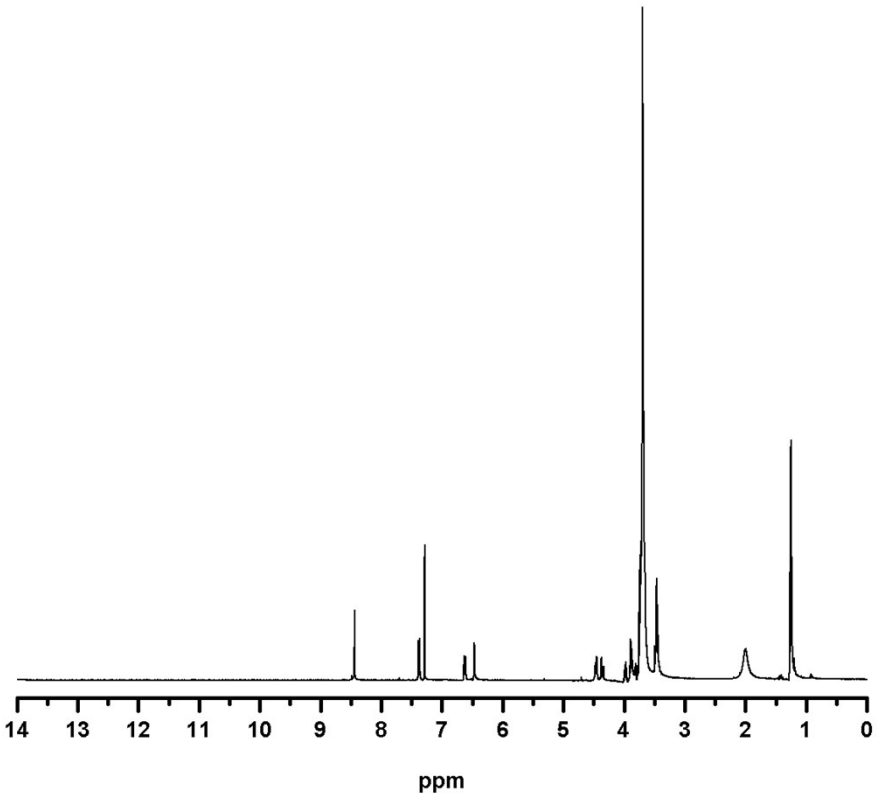


Figure S8 The ^{1}H-NMR spectrum of sensor **CC** in $CDCl_3$ at 400 MHz

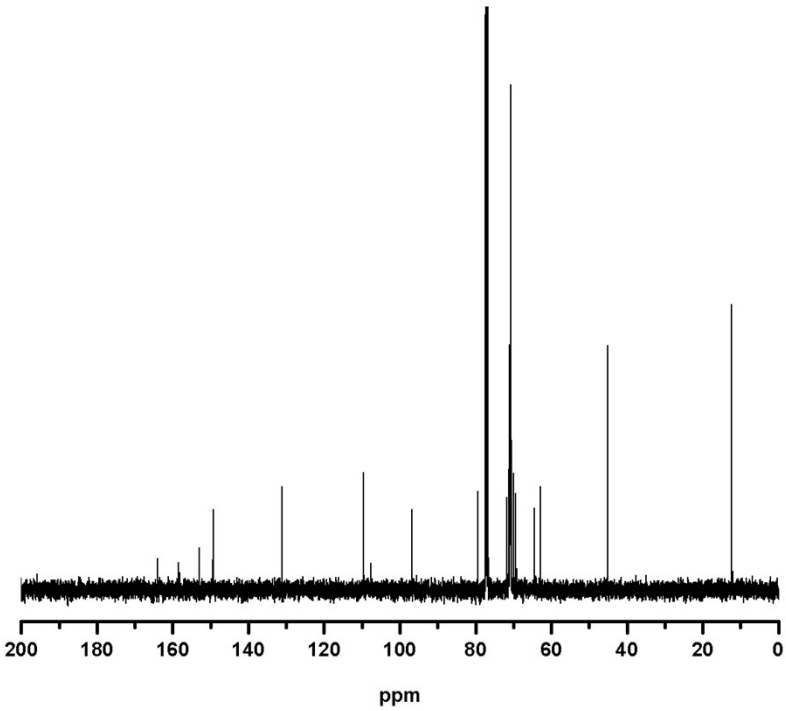


Figure S9 The ^{13}C-NMR spectrum of sensor **CC** in $CDCl_3$ at 400 MHz

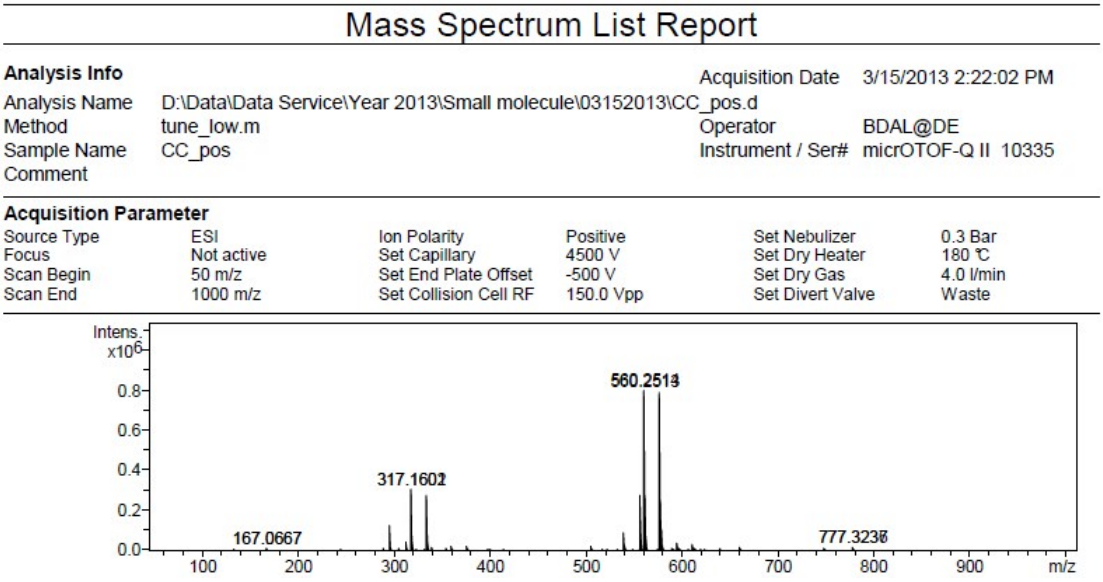


Figure S10 The ESI-High Resolution Mass spectrum of sensor **CC** at 560.25 m/z

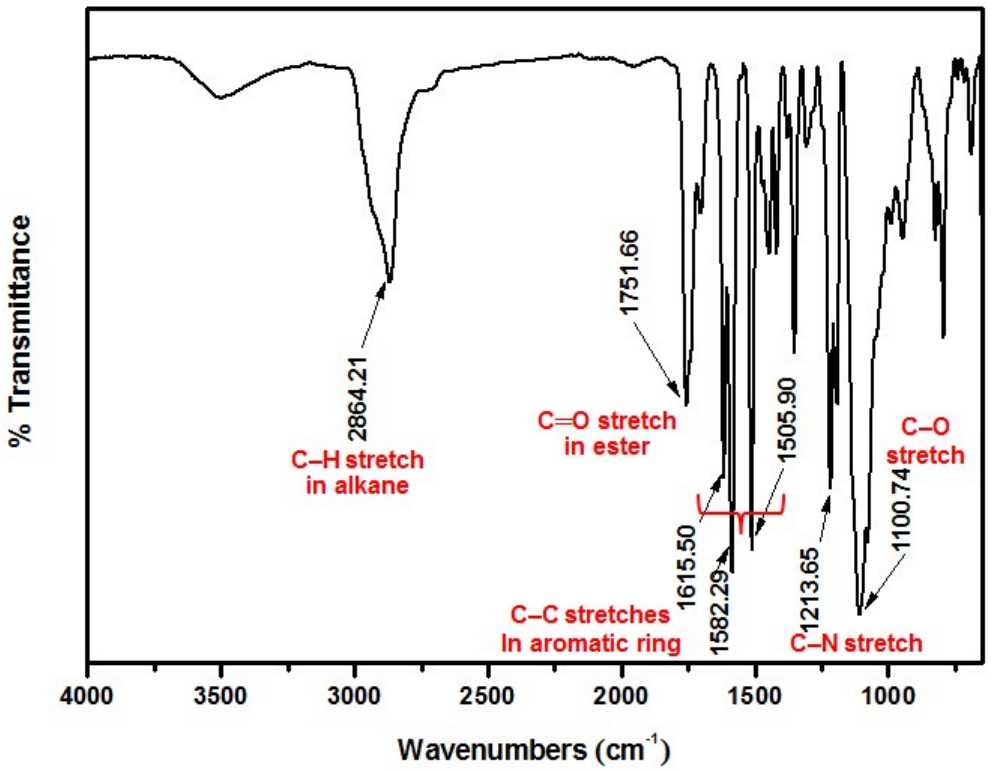
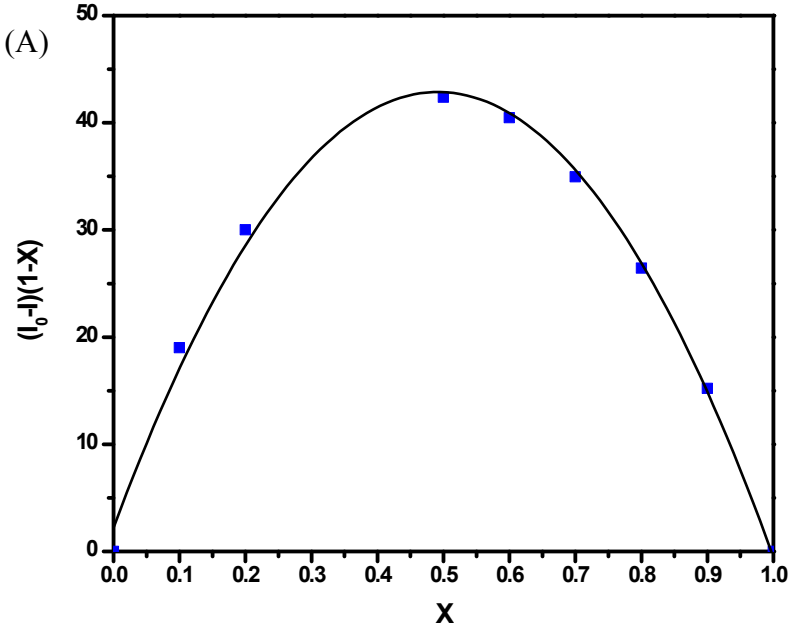


Figure S11 IR spectrum of sensor **CC**

3. Job's plots studies



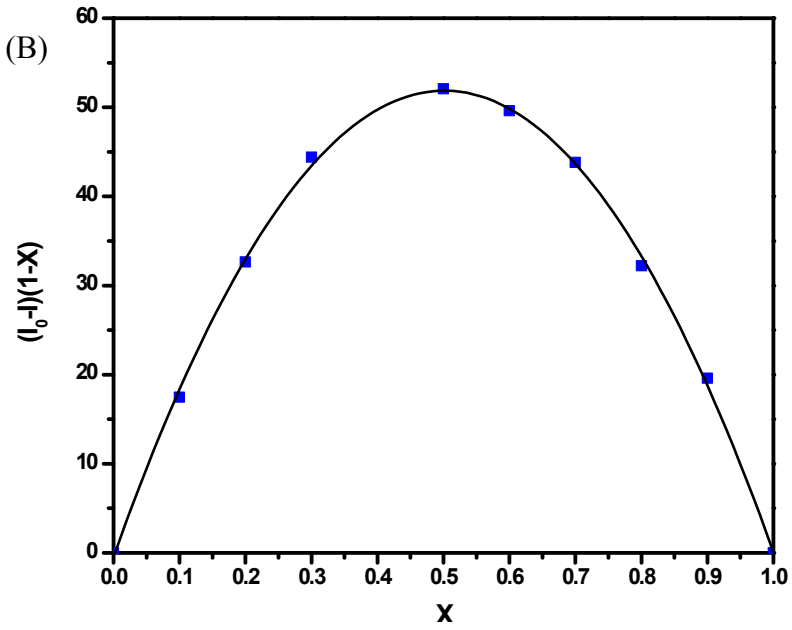
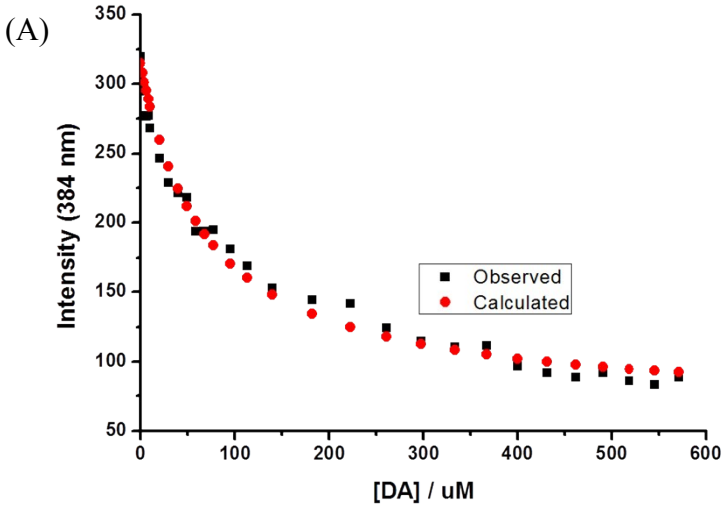


Figure S12 Job's plots for 1:1 complex of sensor **NB** with (A) DA (B) NE at 1 x 10^{-5} M in DMSO:phosphate buffer (0.01 M, pH 7.4, 1:9, v/v)

4. Non-linear regression plots



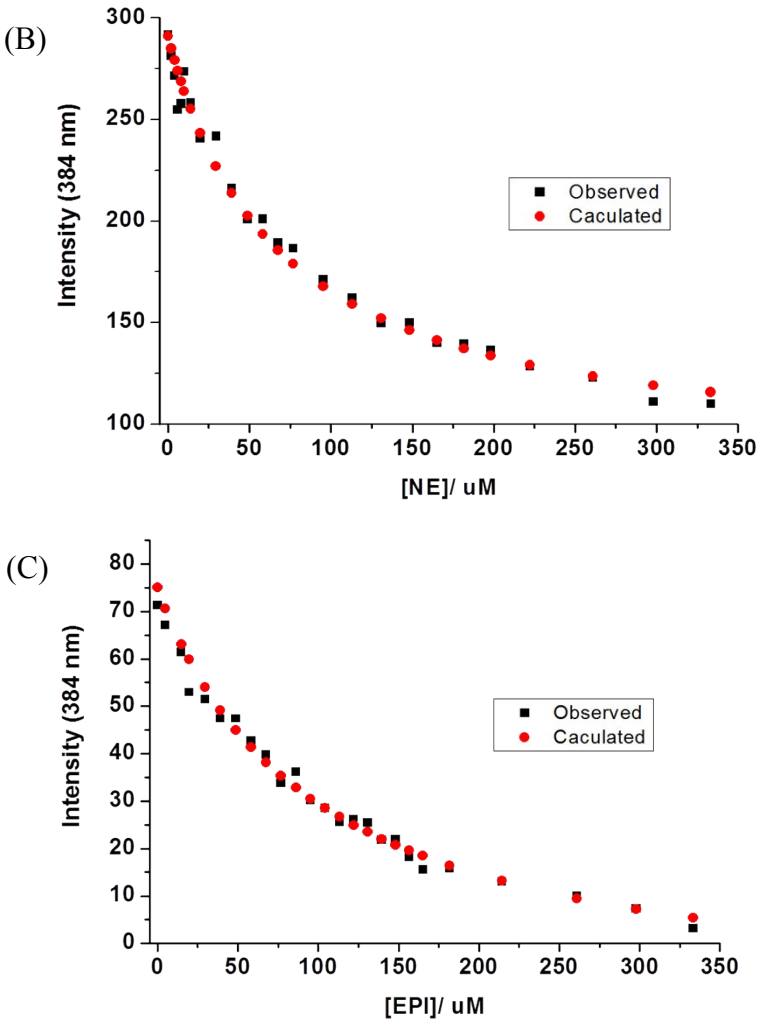


Figure S13. Non-linear regression plots of sensor **NB** (1 x 10^{-5} M) toward 0-80 equiv. of (A) DA (B) NE and (C) EPI in DMSO:phosphate buffer (0.01 M, pH 7.4, 1:9, v/v) for calculation of binding constant (K_s)

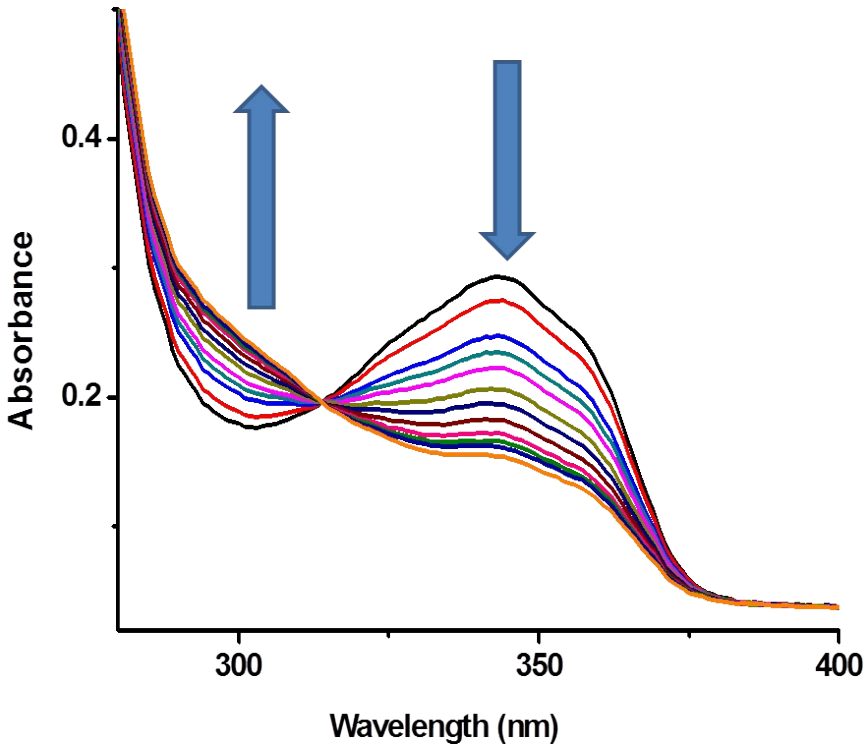


Figure S14. UV-vis spectra titration **NB** with EPI

5. Calibration curves of detection limit for DA, NE and EPI

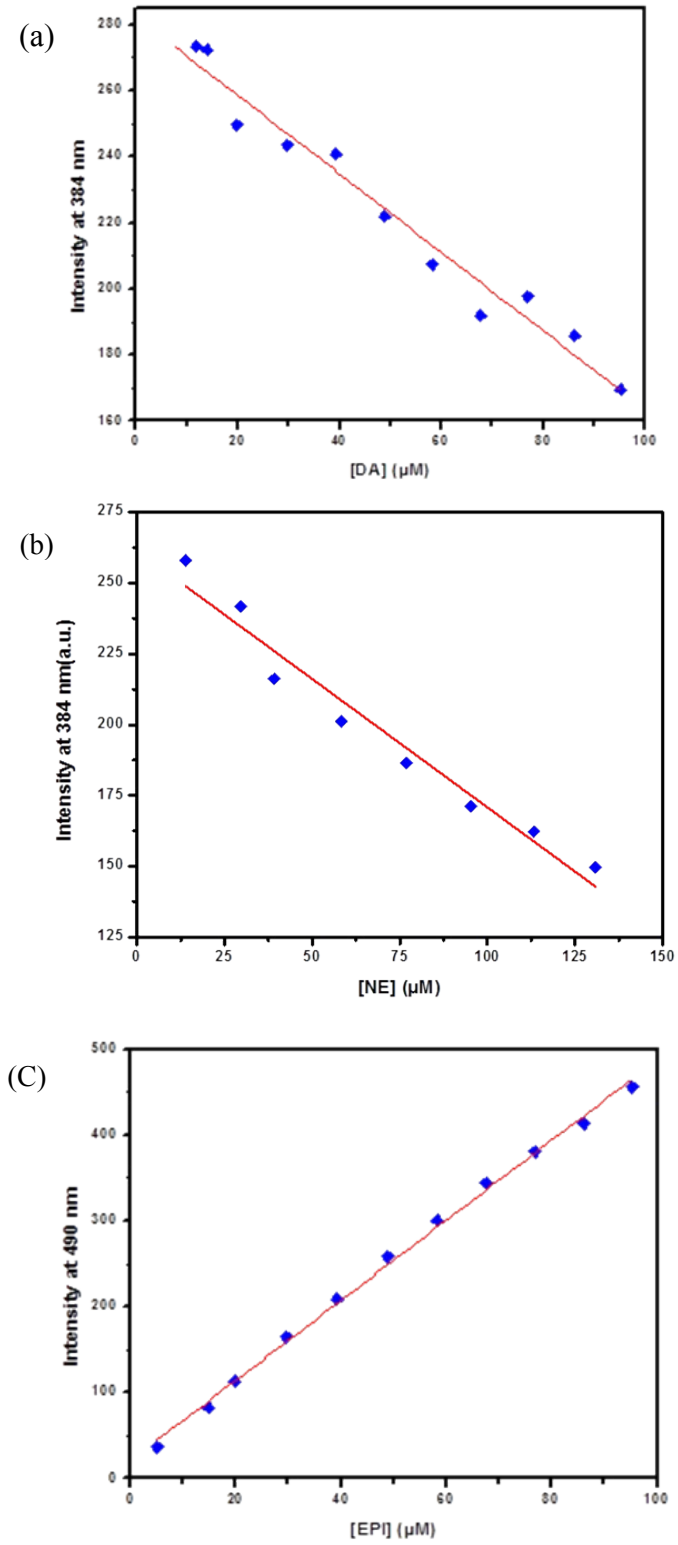


Figure S15. Linear plot of fluorescence intensity between sensor with a) DA b) NE c) EPI complexes and concentration of catecholamine guests

*6. Complexation studies of sensor **CC** with various guests by fluorescence spectrophotometry technique*

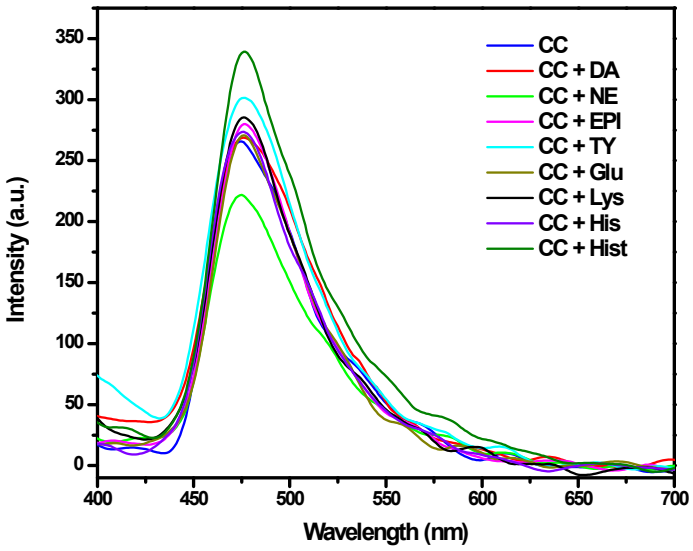


Figure S16 Fluorescence spectra of sensor **CC** (5 x 10^{-5} M) in DMSO:phosphate buffer (0.1 M, pH 7.4, 1:9, v/v) in the present of various guest 100 equiv. (λ_{ex} = 340 nm)

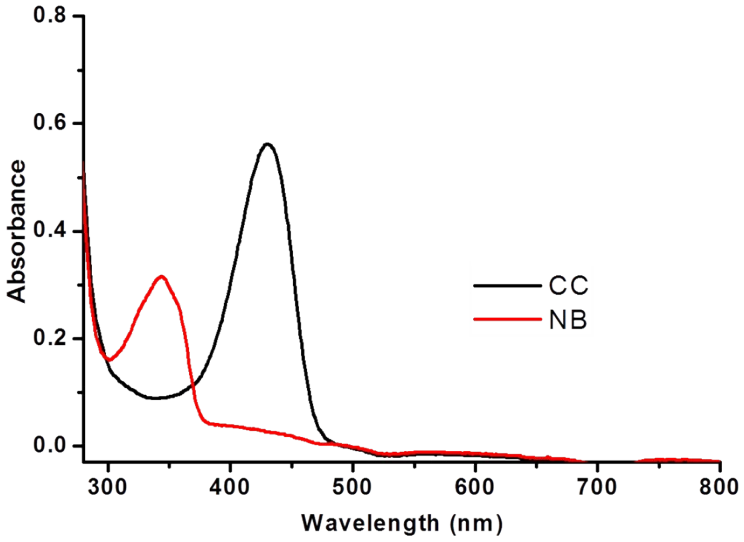


Figure 17. UV-vis spectra of **NB** and **CC** 1x10^{-4} M

7. Principle component analysis (PCA) method for analysis of complexation

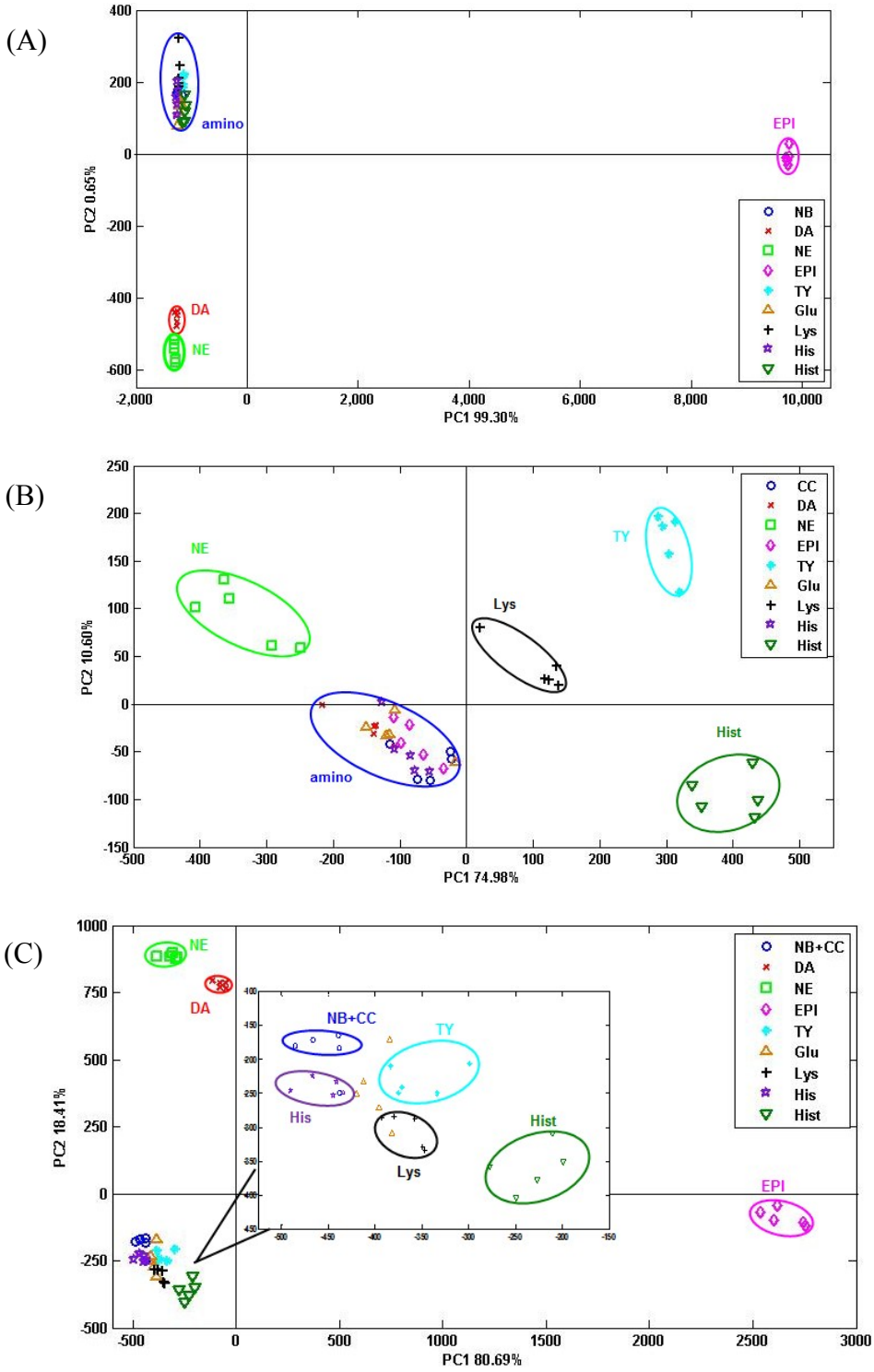


Figure S18. PCA score plot of (A) sensor **NB** (B) sensor **CC** and (C) the mixture sensors **NB** and **CC** upon addition of various guests (100 equiv.) in 1:9, v/v DMSO:phosphate buffer (pH 7.4). PCA score plot shows clustering for all 9 samples.

*8. Complexation study of sensor **NB** with EPI in human urine sample*

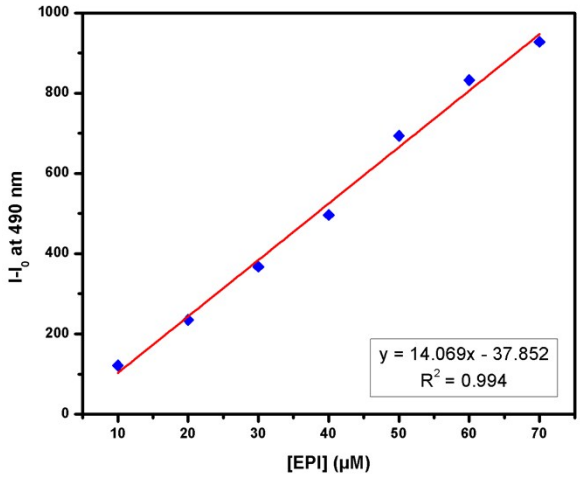


Figure S19. Calibration curve of sensor **NB** with the spiked EPI in the synthetic urine

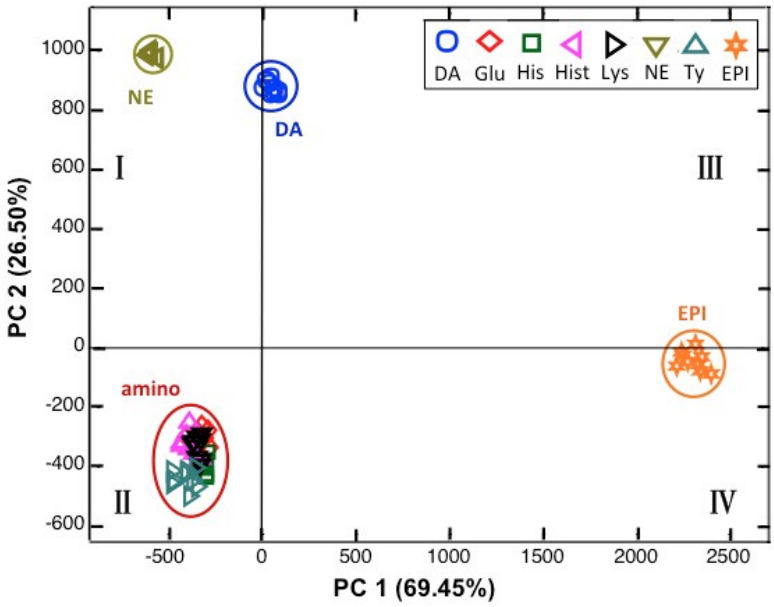


Figure S20. PCA score plots of mixed NB and CC upon addition of various guests (100 equiv.) in urine samples and including fluorescence data of EPI for determining of PCA analysis

Table S1 Composition of synthetic urine [1]

Species	Concentration (g l^{-1})	Concentration (mmol l^{-1})
$CaCl_2.H_2O$	0.65	Ca: 4.3
$MgCl_2.6H_2O$	0.651	Mg:3.2
NaCl	4.6	SO_4: 16
Na_2SO_4	2.3	Citrate: 2.3
$Na_3C_6H_8O_7.2H_2O$	0.65	Oxalate: 0.149
$Na_2C_2O_4$	0.020	
KH_2PO_4	2.8	PO_4: 20.5
KCl	1.6	
NH_4Cl	1	NH_4: 19
$CO(NH_2)_2$	25	
$C_4H_7N_3O$	1.1	
Total Na = 118 mEq Total K = 42 mEq pH = 5.8		

Table S2 Comparison of the sensitivity of sensory system for dopamine

Sensing materials	Detection	Linear range	LOD	Ref.
MIP-Si-ITO[a] electrode	Electrochemistry	$2x10^{-6}$– $8x10^{-4}$ M.	$2x10^{-6}$ M.	2
Au at SiO_2-MIPs-GCE	Electrochemistry	$4.8x10^{-8}$-$5x10^{-5}$ M.	$2x10^{-8}$ M	3
C60-CNT/IL/GC electrode	Electrochemistry	0.06-25 μM	15 nM	4
Citrate-caped AuNPs	Colorimetric	2.5-20 μM	2500 nM	5
MBA-and DSP-modified AuNPs[b]	Colorimetric	5-180 nM	0.5 nM	6
Fe_3O_4 NPs	Fluorescence	0.01-0.4 μM	3 μM	7
Self-assembled **PBA** and **CC**[c]	Fluorescence	16.7-47.4 μM	1.47 μM	8
Phosephate-modified TiO_2 NPs	Fluorescence	0.5-100 μM	30 nM	9
Napthalimide-boronic acid	Fluorescence	11.9-95.2 μM	7.71 μM	This work

[a]Si-ITO: silanizedindiumtinoxideelectrode.

[b]**MBA**, 4-mercaptopheylboronic acid; **DSP**, dithiobis (succinimidyl propionate)

[c]**PBA**, Pyrene boronic acid; **CC**, crown-coumarin

[1] E. Tilley, J. Atwater, D. Mavinic, Effects of storage on phosphorus recovery from urine, Environ. Technol. 29 (2007) 807-816.

[2] N. Gao, Z. Xu, F. Wang, S. Dong, Electroanalysis, 2007, **19**, 1655–1660.

[3] D. Yu, Y. Zeng, Y. Qi, T. Zhou, G. Shi, *Biosen. Bioelectron.,* 2012, **38**, 270–277.
[4] M. Mazloum-Ardakani, A. Khoshroo, *Electrochem. Commun.* 2014, **42**, 9–12.
[5] R. Baron, M. Zayats, I. Willner, *Anal. Chem*., 2005, **77**, 1566-1571.
[6] B.A. Kong, A.W. Zhu, Y.P. Luo, Y. Tain, Y.Y.Yu, G.Y. Shi, *Angew, Chem, Int. Ed*., 2011, **50**, 1837-1840.
[7] C.-H. Liu, C. J. Yu, W.-L. Tseng, *Anal. Chim. Acta*, 2012, **745**, 143-148.
[8] A.Chaicham, S. Sahasithiwat, T. Tuntulani, B. Tomapatanaget, *Chem. Commun.,* 2013, **49**, 9287-9289
[9] H.P. Wu, T.L. Chang, W.L. Tseng, *Langmuir*, 2007, **23** 7880-7885.