Electronic Supplementary Information for

Mesoporous silica particle for selective detection of dopamine with beta-cyclodextrin as the selective barricade

Changmin Yu, Ming Luo, Fang Zeng,* Fangyuan Zheng, and Shuizhu Wu*

College of Materials Science & Engineering, South China University of Technology, Guangzhou, P. R. China;

State Key Laboratory of Physics & Chemistry of Luminescence, South China University of Technology, Guangzhou, 510640, P. R. China.

E-mail: mcfzeng@scut.edu.cn, shzhwu@scut.edu.cn

Contents

1. Experimental Details

Reagents and materials. Sodium azide (N_3Na) and p-toluenesulfonyl chloride (Ts-Cl) were purchased from Acros. β-cyclodextrin, L-serine (Ser), tetraethoxysilane (TEOS), 2-adamantanamine and tetrasodium ethylenediaminetetraacetic acid ($EDTANa_4$) were obtained from Alfa. (3-Isocyanatopropyl)triethoxysilane, 3-mercaptopropyltrimethoxysilane (MPS), o-phthalaldehyde, n-cetyltrimethylammonium bromide (CTAB), sodium ascorbate, L-tryptophan (Trp), L-phenylalanine (Phe), L-tyrosine (Tyr), L-glutamic acid (Glu), L-arginine (Arg), L-lysine (Lys), L-aspartic acid (Asp), L-threonine (Thr), L-asparagine (Asn), L-glutamine (Gln), cysteine (Cys), glycine (Gly), L-proline (Pro), L-alanine (Ala), L-isoleucine (IIe), L-leucine (Leu), L-methionine (Met), L-valine (Val), L-histidine (His), ascorbic acid, dopamine hydrochloride, propargylamine, norepinephrine and epinephrine were obtained from Sigma. Fetal bovine serum was supplied by Hangzhou Sijiqing Biological Engineering Materials Co. Ltd. Human urine was from a healthy female. N,N-dimethyl-formamide (DMF) was dried with CaH_2 and vacuum distilled. Tetrahydrofuran (THF) was dried over molecular sieves and vacuum distilled. Methanol, ethanol and dichloroethane were analytically pure solvents and distilled before use.

Synthesis of the 1-(3-(triethoxysilyl)propyl)-3-(prop-2-ynyl)urea (the propynyl-containing silane precursor). (3-Isocyanatopropyl)triethoxysilane (1.0 g, 4.04 mmol) was dissolved in 10 mL of anhydrous THF. The solution was cooled to 0 - 5 °C in an ice-water bath. Then the solution of propargylamine (0.22 g, 4.04 mmol) in 6 mL of anhydrous THF was added dropwise to the reaction mixture over 10 min with stirring vigorously under N_2

protection. After 3 h of stirring at 0 - 5 °C, the reaction mixture was heated to 65 °C and stirred over night. The solvent was then evaporated, and the residue was purified by silica gel column chromatography using 3:1 (v/v) ethyl acetate/petroleum ether as the eluent to give pale solid. Yield: 0.6 g, 50%. ^{1}H NMR(400MHz, $CDCl_3$), δ: 0.62-0.66 (m, 2H), 1.20-1.26 (m, 9H), 1.58-1.66 (m, 2H), 2.22 (m, 1H), 3.16-3.20 (m, 2H), 3.69-3.85 (m, 6H), 3.98 (m, 2H). ESI MS *m/z* $[M+Na]^+$ 324.6.

Synthesis of the mercaptopropyl-functionalized mesoporous silica nanoparticle (Thiol-MSN). First, n-cetyltrimethylammonium bromide (CTAB, 1.0 g, 2.74 mmol) was dissolved in 480 mL of deionized water. NaOH (aq) (2.0 M, 3.50 mL) was added to the solution, and then the solution was heated to 80 °C. Tetraethoxysilane (TEOS, 5.0 mL, 22.4 mmol) was first introduced dropwise to the surfactant solution, followed by the dropwise addition of 3-mercaptopropyltrimethoxysilane (MPS) (1.28 ml, 6.76 mmol). After 2 h of stirring, silica nanoparticles were formed. The solid product was filtered, washed with deionized water and methanol, and dried in a vacuum.

Synthesis of the *β*-CD-covered Thiol-MSN (Thiol-MSN-CD). The mono-6-azido-6-deoxy-*β*-cyclodextrin (*β*-CD-N_3) was synthesized as previous reported (M. Xu, S. Wu, F. Zeng, C. Yu, *Langmuir* **2010**, *26*, 4529–4534). The synthesis procedures were briefly described as follows: first mono-6-O-(p-tolylsulfonyl)-*β*-cyclodextrin (6-TSO-*β*-CD) was synthesized through the reaction of *β*-cyclodextrin with p-toluenesulfonyl chloride. Then mono-6-(p-tolylsulfonyl)-*β*-CD was reacted with sodium azide to offer the *β*-CD-N_3.

The propynyl-containing silica particles (Thio-MSN-propyne with template) were prepared as follows: the as-made Thiol-MSN (with template CTAB inside mesoporous channels, 1.5 g) was dispersed in 150 mL of anhydrous toluene. This suspension was sonicated for 30 min and then placed into an oil bath heated to 120 °C. Afterwards the solution of the propynyl-containing silane precursor (0.3 g, 0.96 mmol) in 6 mL of anhydrous toluene was added dropwise to the suspension with stirring vigorously under nitrogen. The suspension was refluxed for 6 h and cooled to room temperature. These functionalized silica nanoparticles were filtered, and washed extensively with anhydrous toluene and methanol. The resulting propynyl-containing silica nanoparticles (Thio-MSN-propyne with template) were dried under high vacuum to remove the remaining solvent.

To remove the surfactant template, silica nanoparticles (Thio-MSN-propyne with template, 1.4 g) was refluxed for 24 h in a solution of 9.00 mL of HCl (37.4%) and 140.00 mL of methanol. The particles were recovered by centrifugation at 10000 rpm for 20 min. These particles were redispersed in deionized water for several times and in methanol for several times and centrifuged. The resulting surfactant-removed Thiol-MSN-propyne material was dried under high vacuum to remove the remaining solvent in the mesopores.

The "click chemistry" reaction of the propynyl-containing silica particles: the dried propynyl-containing silica particles (Thio-MSN-propyne with CTAB removed, 0.5 g) were dispersed in 30 mL of methanol and the mixture was sonicated for 30 min, and then the solution of mono-6-azido-6-deoxy-β-cyclodextrin (0.87 g, 0.75 mmol, excessive amount) in 28 mL deionized water, sodium ascorbate (0.03 g, 0.15 mmol, dissolved in 1 mL water), $CuSO_4.5H_2O$ (0.019 g, 0.075 mmol, dissolved in 1 mL water) were added. After stirring for

40 h at 40 °C, the obtained materials were recovered by centrifugation at 10000 rpm for 20 min, washed with deionized water for several times, and 10% $EDTANa_4$ for three times. The resulting nanoparticles (Thio-MSN-CD) were dried under high vacuum.

Conversion of the thio moieties of Thiol-MSN-CD into OPTA groups (OPTA-MSN-CD, the *β*-CD/MSN sensor) and those of Thio-MSN-propyne into OPTA groups (OPTA-MSN-propyne, the control sample). The mercaptopropyl functionality was then converted to the amine-sensitive OPTA group by reacting the Thiol-MSN-CD (0.5 g) or the Thio-MSN-propyne (0.5 g) with o-phthalaldehyde (OPA, 0.277 g, 2.06 mmol) in 40 mL of methanol for 12 h. After centrifugation, the resulting material was thoroughly washed with methanol and dried under vacuum, finally the *β*-CD/MSN Sensor --- OPTA-MSN-CD and the control sample --- OPTA-MSN-propyne were obtained.

Measurements. ^{1}H NMR spectra were recorded on a Bruker Avance 400 MHz NMR spectrometer. UV-vis spectra were recorded on a Hitachi U-3010 UV-vis spectrophotometer. Fluorescence spectra were recorded on a Hitachi F-4600 fluorescence spectrophotometer. Mass spectra were obtained through a Bruker Esquire HCT Plus mass spectrometer. Powder XRD diffraction data were collected on a X'Pert PRO X-ray diffractometer using Cu Kα radiation. Nitrogen adsorption and desorption isotherm, surface area and median pore diameter were measured using a Micromeritics ASAP2020 sorptometer. Transmission electron microscopy (TEM) images were gained using a JEM-2010HR transmission electron microscopy (Japan). FT-IR spectra were measured using a MAGNA 760 (USA, Nicolet

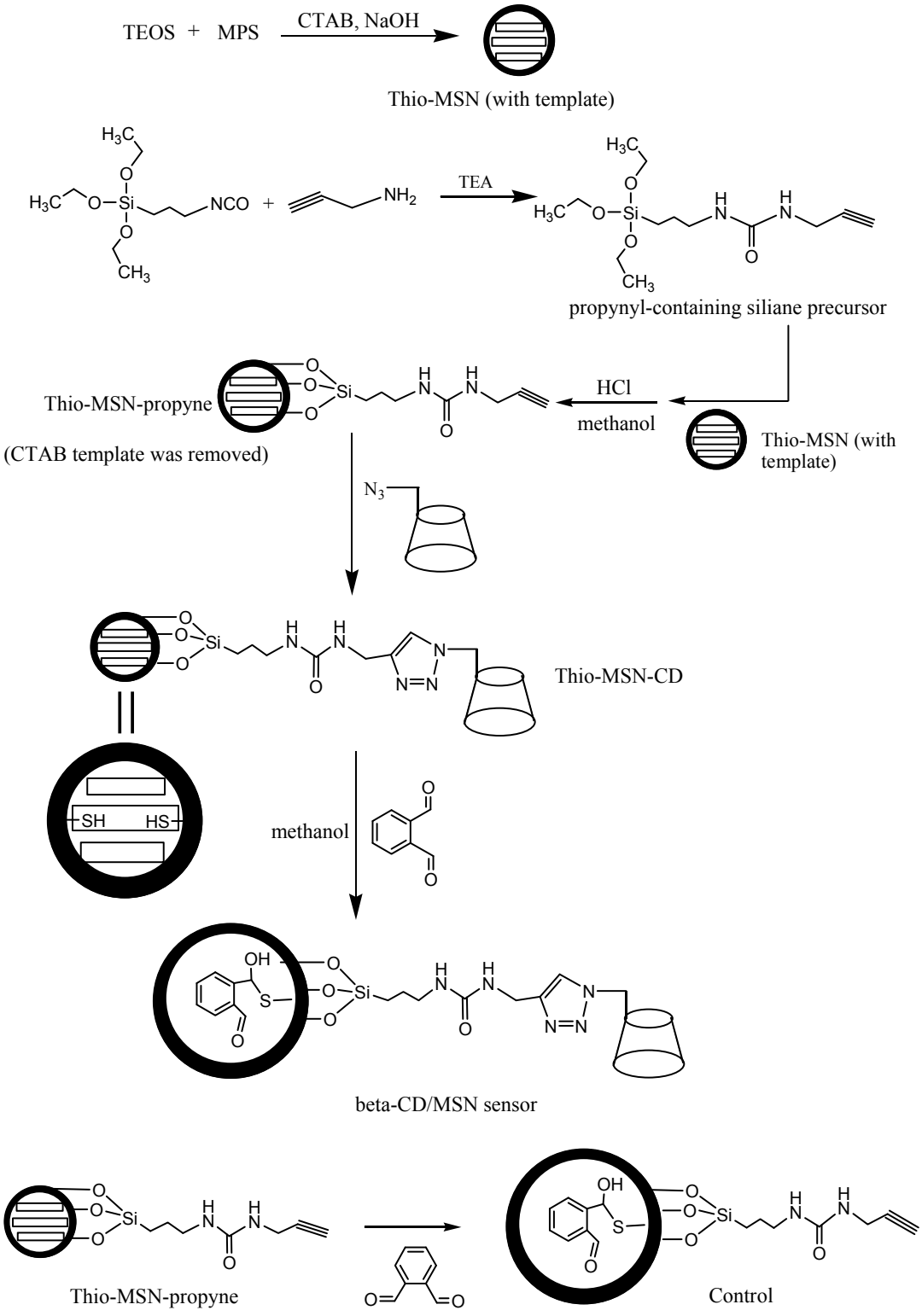
Instrument). Thermogravimetic analysis (TGA) were conducted by using TG209F1 thermal analyzer, the samples were heated from 25 °C to 900 °C with a heating rate of 10 °C per min under nitrogen. The particle size and distribution was determined by dynamic light scattering (DLS) on a Malvern Nano-ZS90 particle size analyzer.

The detection measurements were conducted as follows:

The pH 7.4 PBS buffer (10 mM) solution was bubbled with nitrogen for 15 min. The nanoparticles (dry particles, the β-CD/MSN sensor --- OPTA-MSN-CD and the control sample --- OPTA-MSN-propyne) were introduced into a pH 7.4 PBS buffer (10 mM) solution, and sonicated for 10 min, then dopamine (various amounts, or other substances) was added at 25 °C, the particles' final concentration was 1 mg/mL. After 20 min of mixing under nitrogen, the fluorescence of the particle suspension was recorded.

For the time-scan measurement of the fluorescence intensity, the measurement was conducted immediately after the amines were added into the sensor suspension.

2. Supporting scheme



Scheme S1. Synthesis route for the silica particle-based sensor (*β*-CD/MSN) and the control (OPTA-MSN-propyne).

3. Supporting Figures

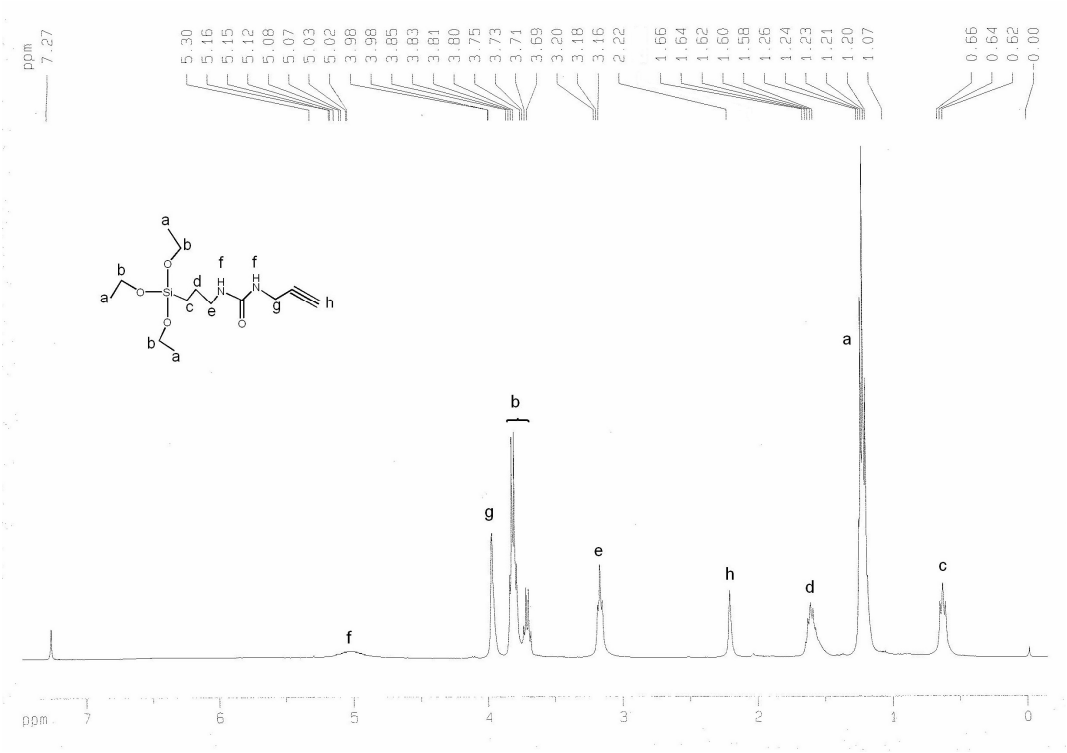
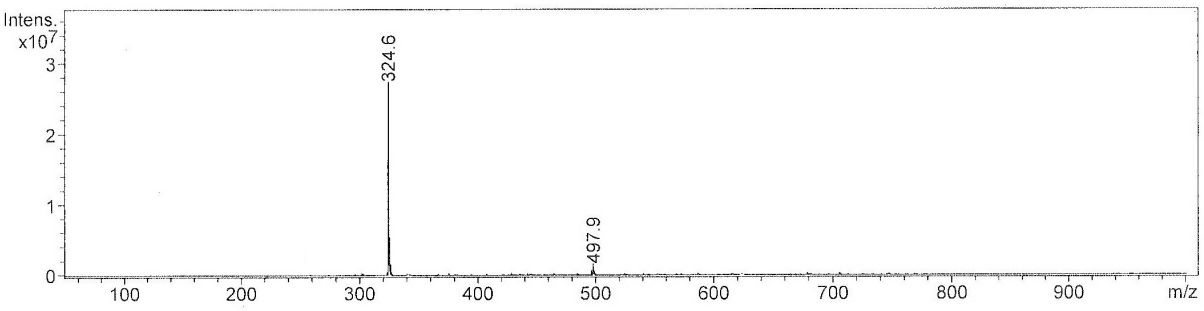


Figure S1. ^{1}H NMR spectrum for the propynyl-containing silane precursor (in $CDCl_3$, measured with NMR BBO BB probe).



#	m/z	I	I %
1	324.6	27319204	100.0
2	325.5	5354525	19.6
3	326.5	1514656	5.5
4	497.9	1568584	5.7

Figure S2. MS spectrum for the propynyl-containing silane precursor.
ESI MS m/z $[M+Na]^+$ 324.6

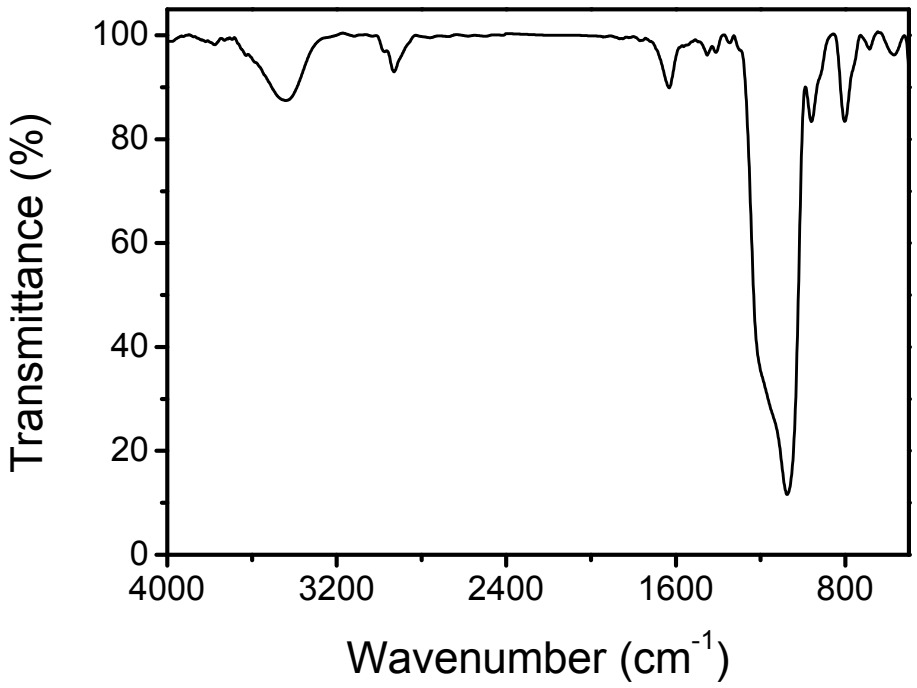


Figure S3. FTIR spectrum of the *β*-CD/MSN sensor.

3440 cm^{-1} (OH ν_{st}), 2980 cm^{-1} (aromatic C-H ν_{st}), 2936 cm^{-1} (aliphatic C-H ν_{st}), 1635 cm^{-1} (ureido C=O ν_{st}), 1450 cm^{-1} (aliphatic CH_2 ν_{δ}), 1410 (C-N ν_{st}), 1080 cm^{-1} (Si-O ν_{st}, C-O-C ν_{st}).

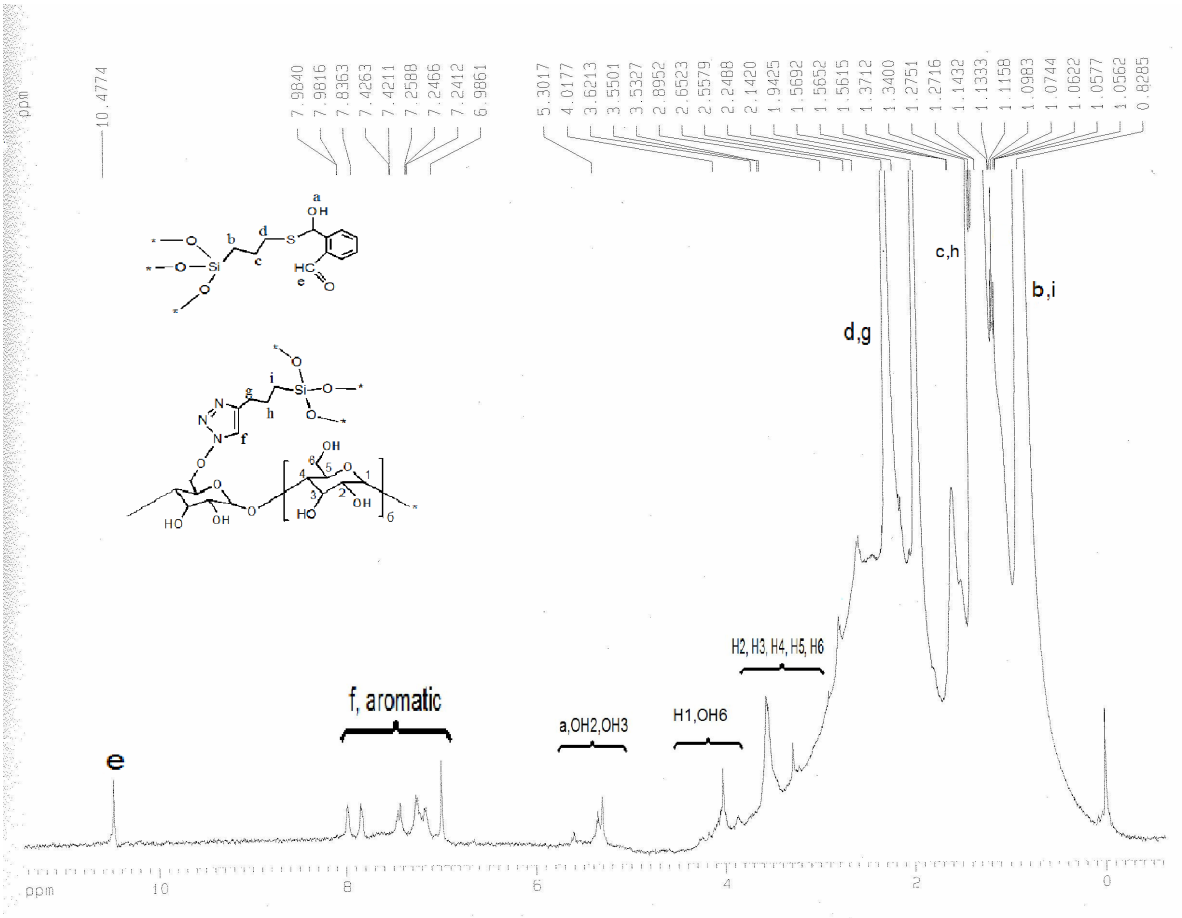


Figure S4. ^{1}H NMR spectrum for the β-CD/MSN sensor (in CD_3CN, measured with NMR HR/MAS probe).

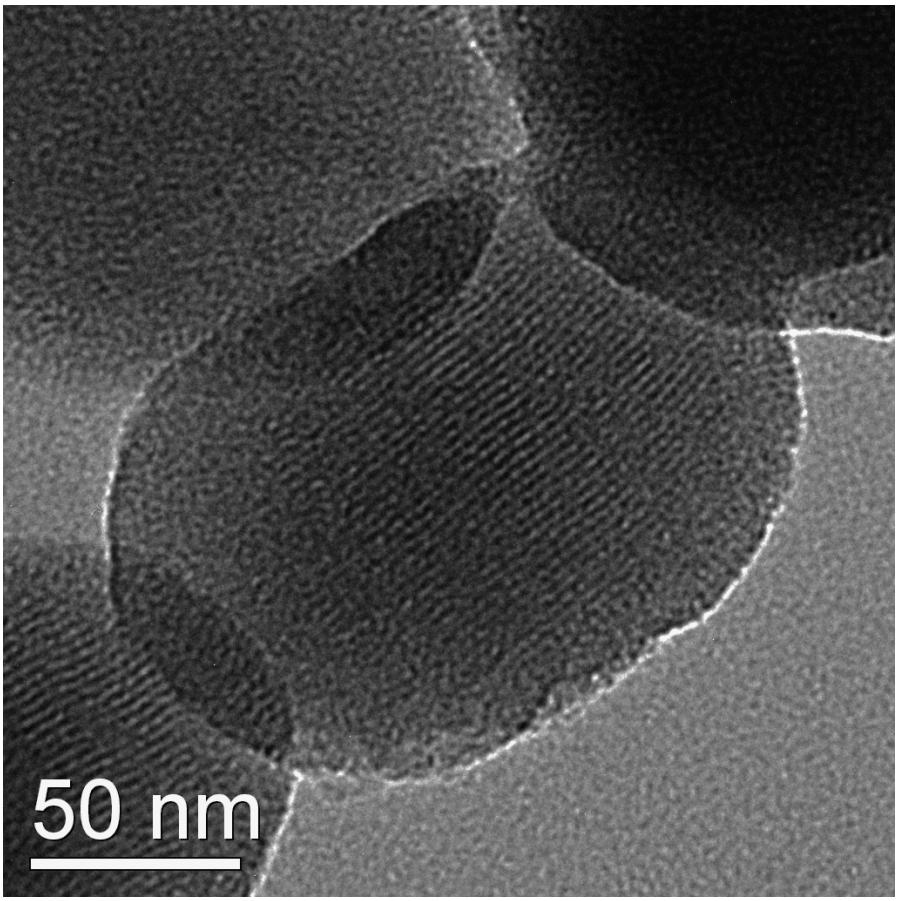


Figure S5. High resolution TEM image for the mesoporous silica nanoparticles (Thio-MSN-propyne).

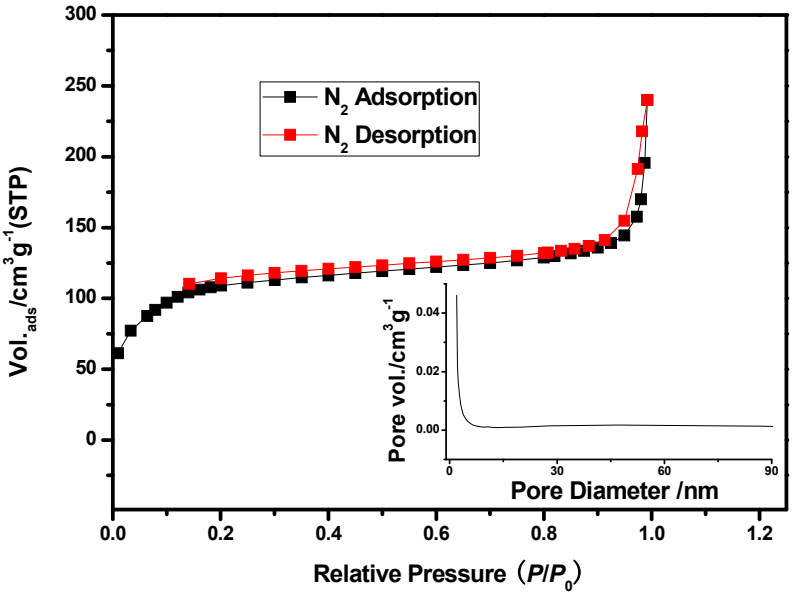


Figure S6. Nitrogen adsorption-desorption isotherms for Thio-MSN-CD.

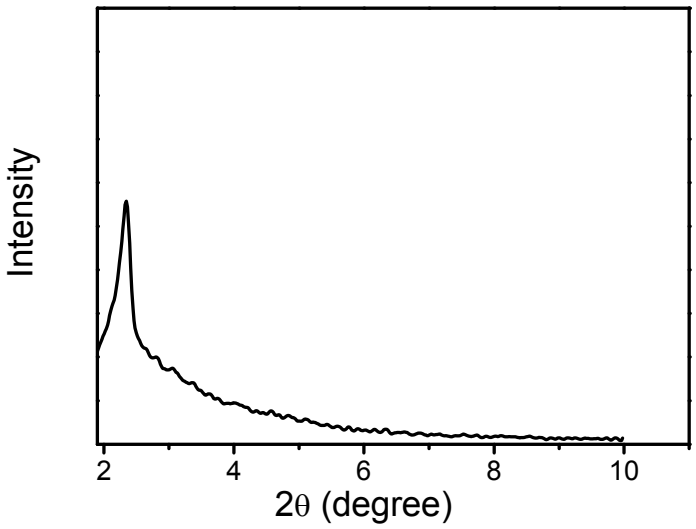


Figure S7. X-ray pattern of the particles (Thio-MSN-CD).

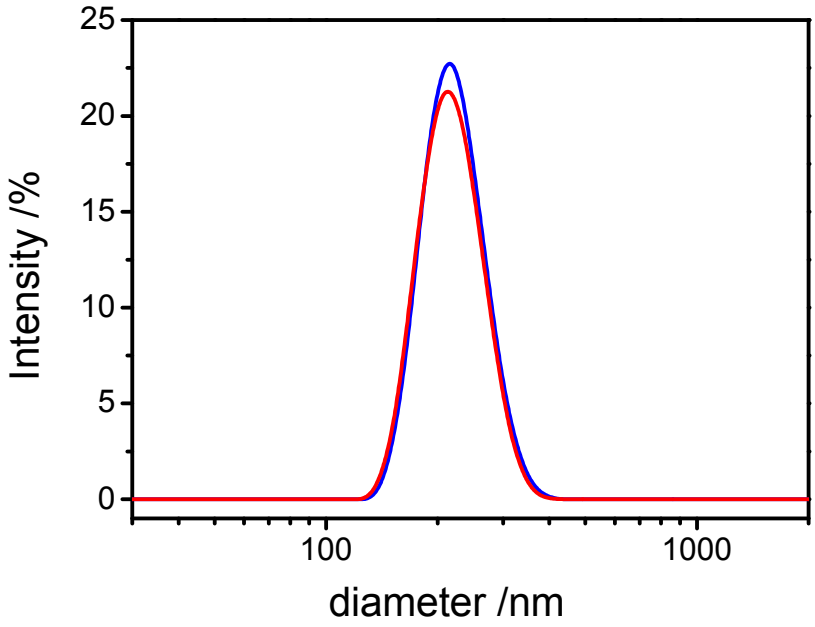


Figure S8. Particle size distribution for Thio-MSN-propyne (red curve) and Thio-MSN-CD (blue curve) respectively determined by DLS. The average diameter for the Thio-MSN-CD is 215 nm.

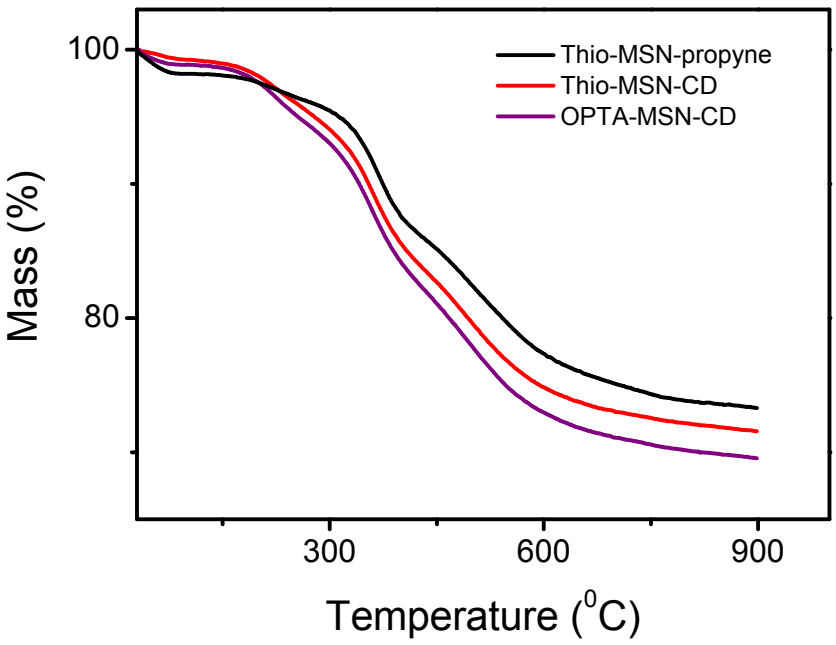


Figure S9. Thermogrametric graphs.

Mass loss at 900°C:

Thio-MSN-propyne 26.4%

Thio-MSN-CD 28.4% OPTA-MSN-CD 30.4%

It is estimated that the amount of β-CD on the particle surface is 1.7×10^{-5} mol/g, and the amount of OPTA in the silica particle was 0.15 mmol/g.

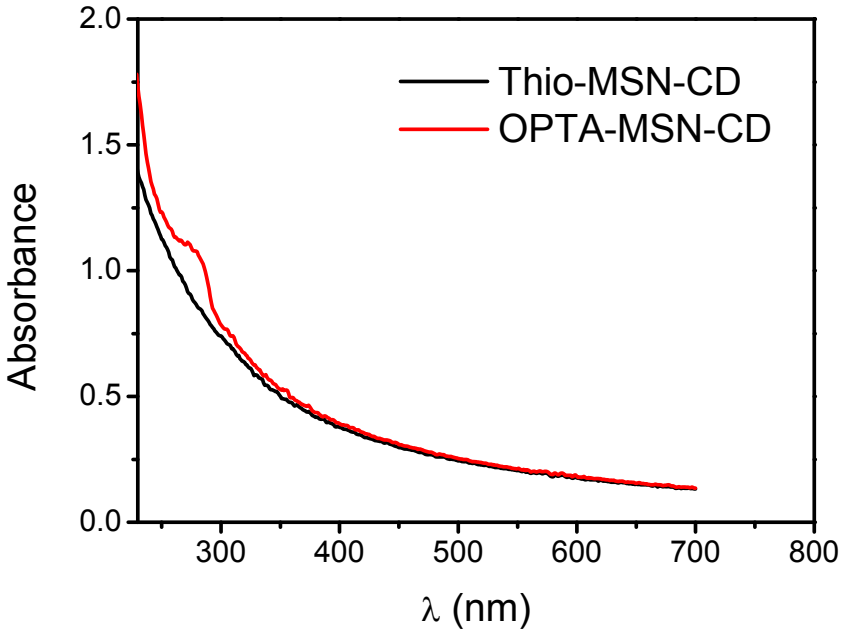


Figure S10. Absorption spectra (0.1 mg/mL).

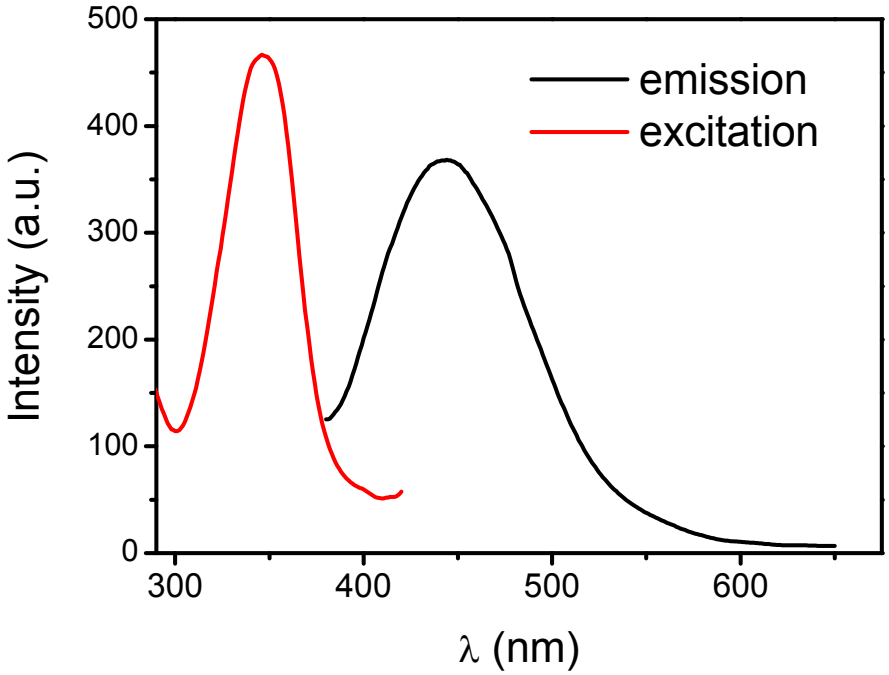


Figure S11. Excitation and emission spectra of OPTA-MSN-CD in the presence of dopamine.

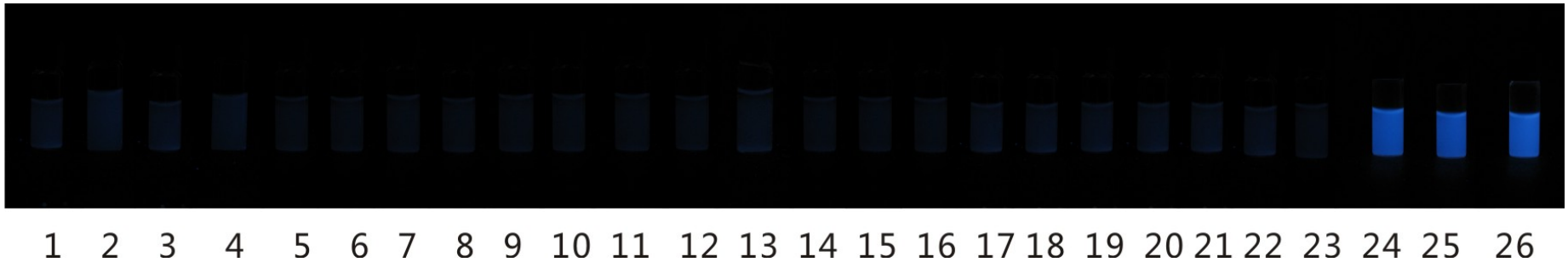


Figure S12. Photographs of the β-CD/MSN sensor (1 mg/mL, in pH 7.4 PBS buffer) in the presence of dopamine, various amino acids, adamantaneamine, epinephrine and norepinephrine respectively (5×10^{-5} M), and in the presence of ascorbic acid (1×10^{-3} M) under a 365-nm UV light (fluorescence change). 1: blank; 2: Phe; 3: Glu; 4: Tyr; 5: Trp; 6: Ser; 7: Arg; 8: Lys; 9: Asp; 10: Thr; 11: Asn; 12: Gln; 13: Cys; 14: Gly; 15: Pro; 16: Ala; 17: IIe; 18: Leu; 19: Met; 20: Val; 21: 2-adamantanamine; 22: ascorbic acid; 23: epinephrine; 24: His; 25: norepinephrine; 26: dopamine.

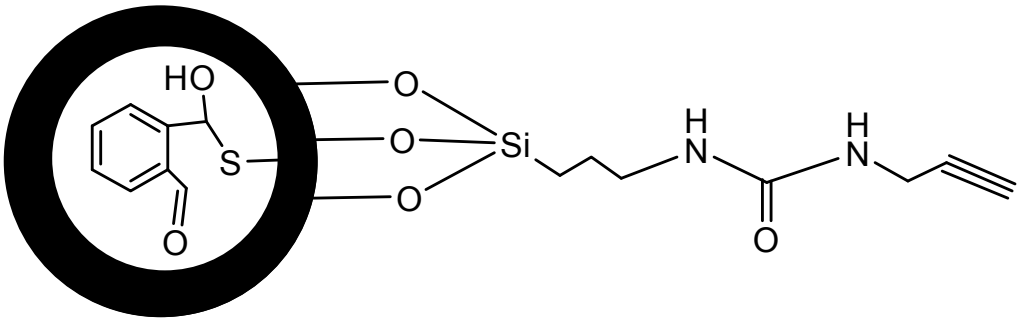


Figure S13. Structure of the control sample (OPTA-MSN-propyne).

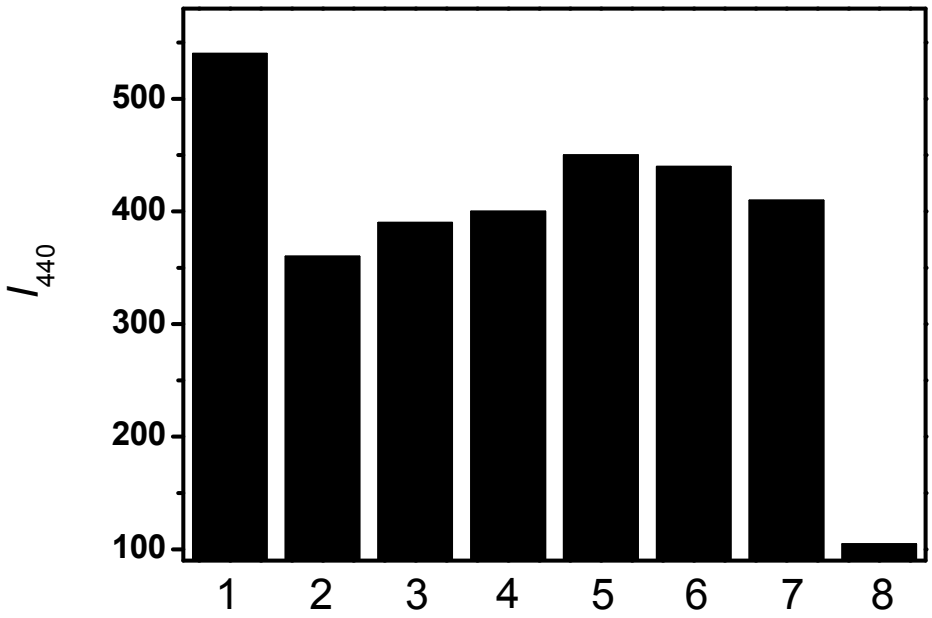


Figure S14. Fluorescence intensity (I_{440}) of the control sample (in pH 7.4 PBS buffer) upon addition of dopamine, amino acids and 2-adamantanamine (5×10^{-5} M) respectively; and in the absence of amines. (λexc = 360 nm). 1: dopamine; 2: Glu; 3: Ser; 4: Tyr; 5: Trp; 6: Phe; 7: 2-adamantanamine; 8: blank.

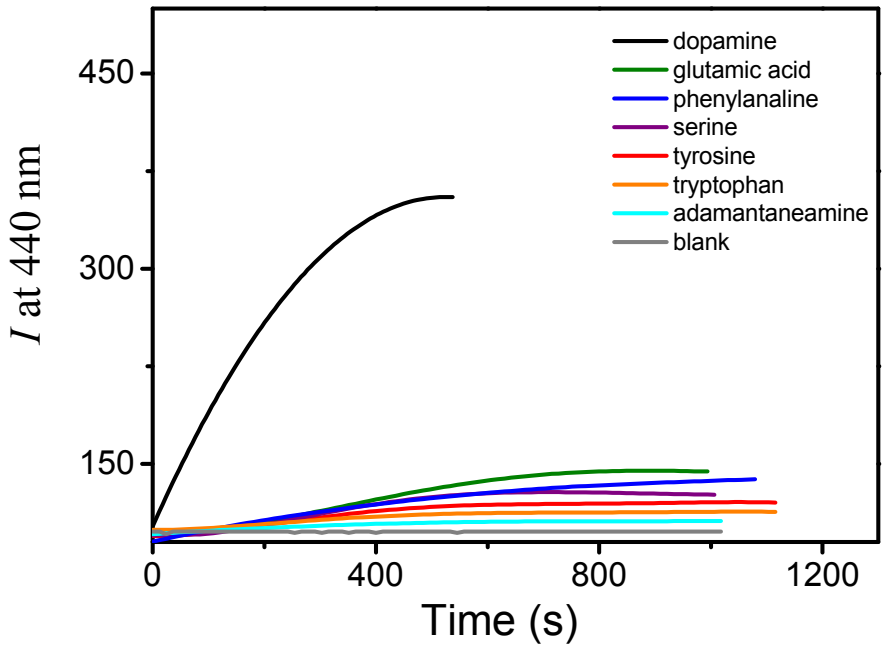


Figure S15. Fluorescence intensity (I_{440}) as a function of time for the blank β-CD/MSN sensor (1 mg/mL, in pH 7.4 PBS buffer); the β-CD/MSN sensor (1 mg/mL, in pH 7.4 PBS buffer) in the presence of dopamine, several amino acids and 2-adamantanamine (5×10^{-5} M) respectively. (The time-scan measurement of the fluorescence intensity was conducted immediately after the amines were added into the sensor suspension)

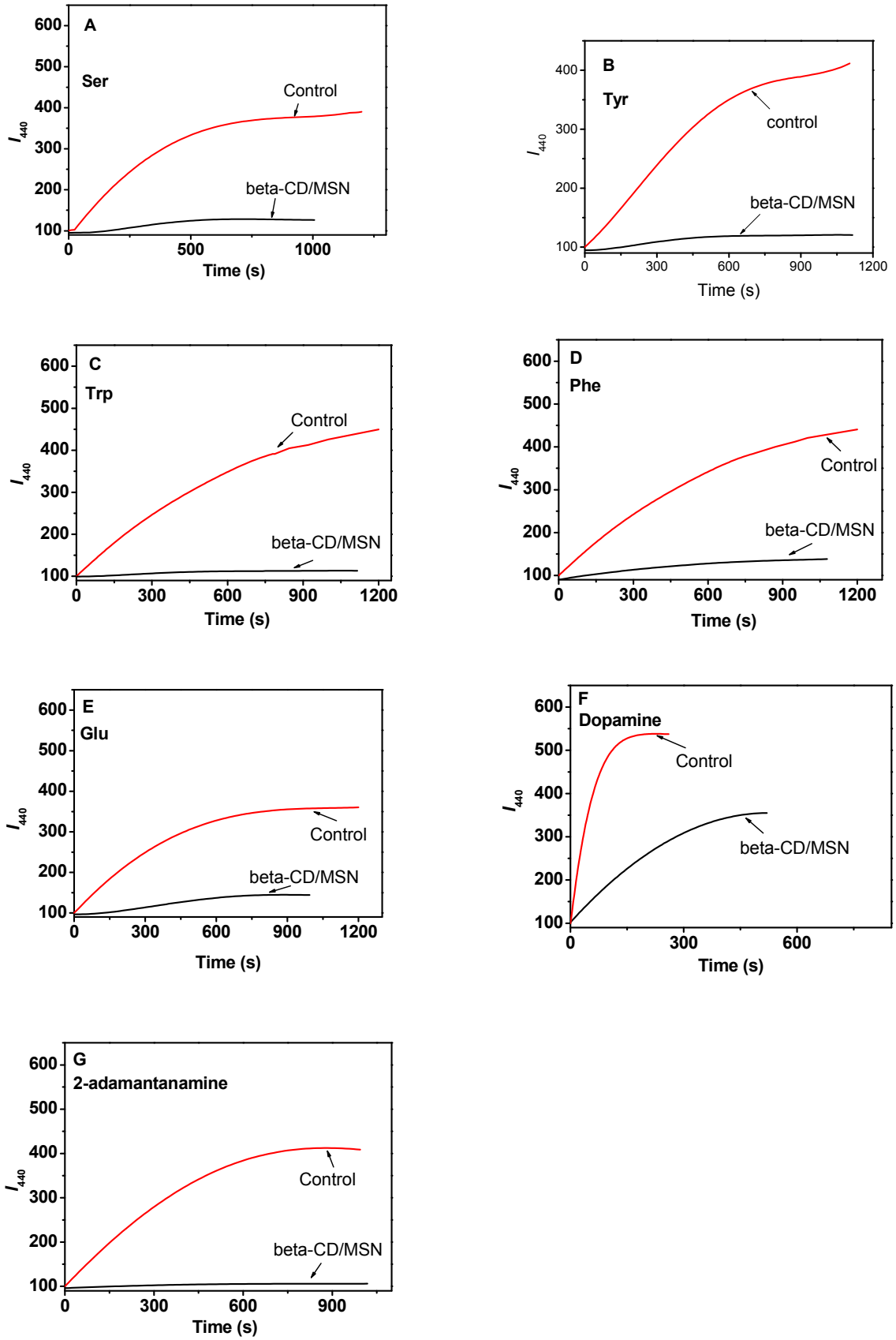


Figure S16. Fluorescence intensity (I_{440}) as a function of time for the control

(OPTA-MSN-propyne, red curves) and the β-CD/MSN sensor (OPTA-MSN-CD, black curves) in the presence of dopamine, several amino acids and 2-adamantanamine (5×10^{-5} M) respectively. A: Ser; B: Tyr; C: Trp; D: Phe; E: Glu; F: dopamine; G: 2-adamantanamine. The time-scan measurement of the fluorescence intensity was conducted immediately after the amines were added into the sensor suspension.

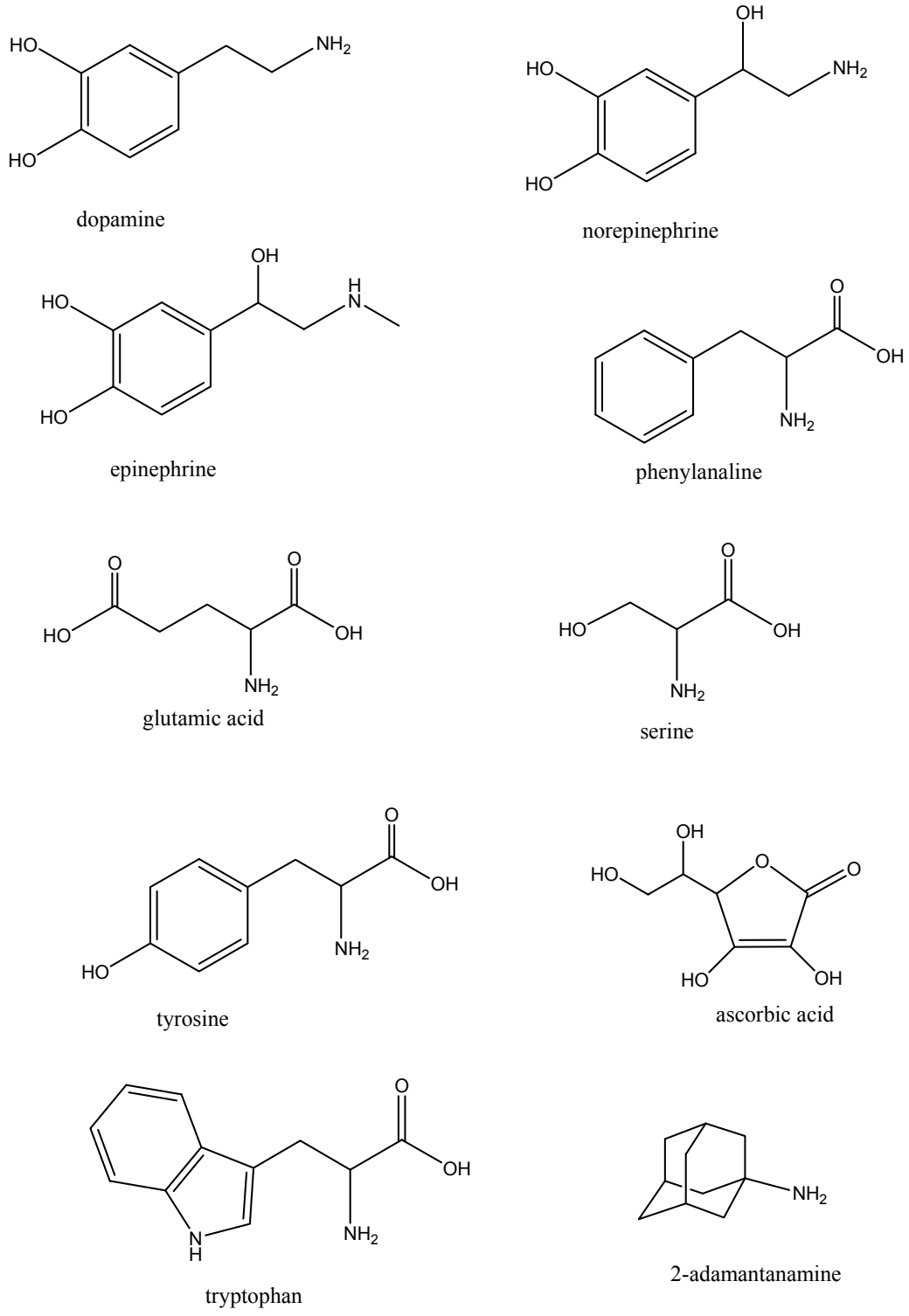


Figure S17. Structure of dopamine, some amino acids, norepinephrine, epinephrine, ascorbic acid and adamantanamine.

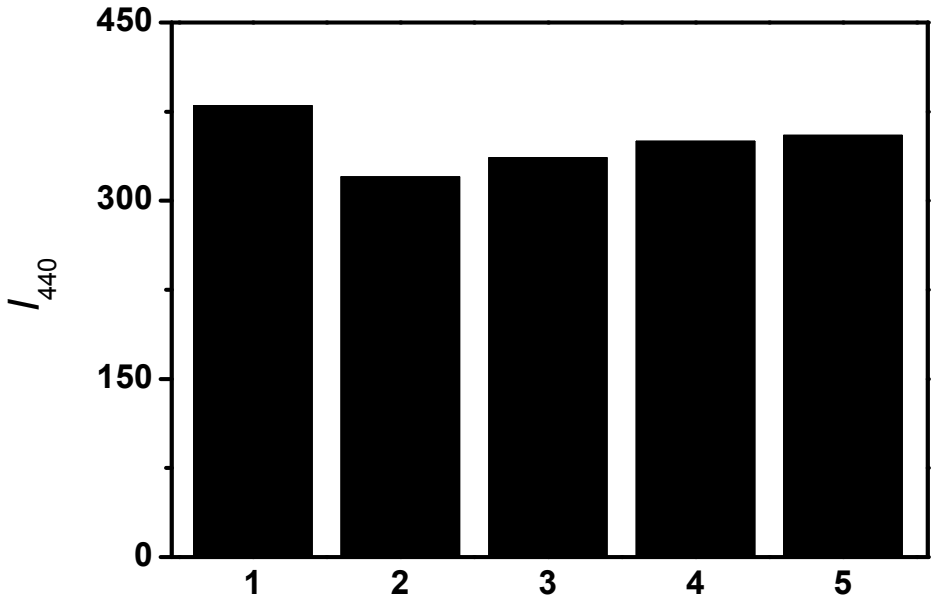


Figure S18. Fluorescence intensity (I_{440}) of the β-CD/MSN sensor (1 mg/mL, in pH 7.4 PBS buffer) upon addition of dopamine only and the suspensions containing dopamine in the presence of biological abundant anions, cations, serum and urine. (λexc = 360 nm). 1: dopamine (5×10^{-5} M), and $Cl^{-1,}$ SO_4^{2-}, HCO_3^- and PO_4^{3-} (5×10^{-5} M respectively); 2: dopamine (5×10^{-5} M), and Na^+, K^+, Ca^{2+}, Mg^{2+}, Fe^{2+} and Fe^{3+} (5×10^{-5} M respectively); 3: dopamine (5×10^{-5} M) and fetal bovine serum (final concentration: 10-fold diluted); 4. dopamine (5×10^{-5} M) and urine (final concentration: 10-fold diluted); 5: dopamine only (5×10^{-5} M).

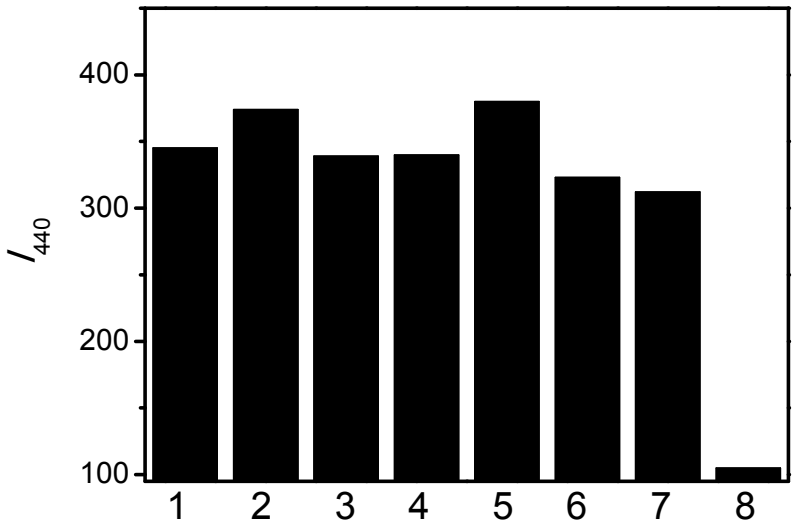


Figure S19. Fluorescence intensity (I_{440}) of the β-CD/MSN sensor (1 mg/mL, in pH 7.4 PBS buffer) upon addition of dopamine only and the suspensions containing dopamine in the presence of other amines. (λexc = 360 nm). 1: dopamine only (5×10^{-5} M); 2: dopamine (5×10^{-5} M), and Glu (5×10^{-5} M); 3: dopamine (5×10^{-5} M) and Tyr (5×10^{-5} M); 4. dopamine (5×10^{-5} M) and Trp (5×10^{-5} M); 5: dopamine (5×10^{-5} M) and Ser (5×10^{-5} M); 6. dopamine (5×10^{-5} M) and Phe (5×10^{-5} M); 7. dopamine (5×10^{-5} M) and 2-adamantanamine (5×10^{-5} M); 8. blank.

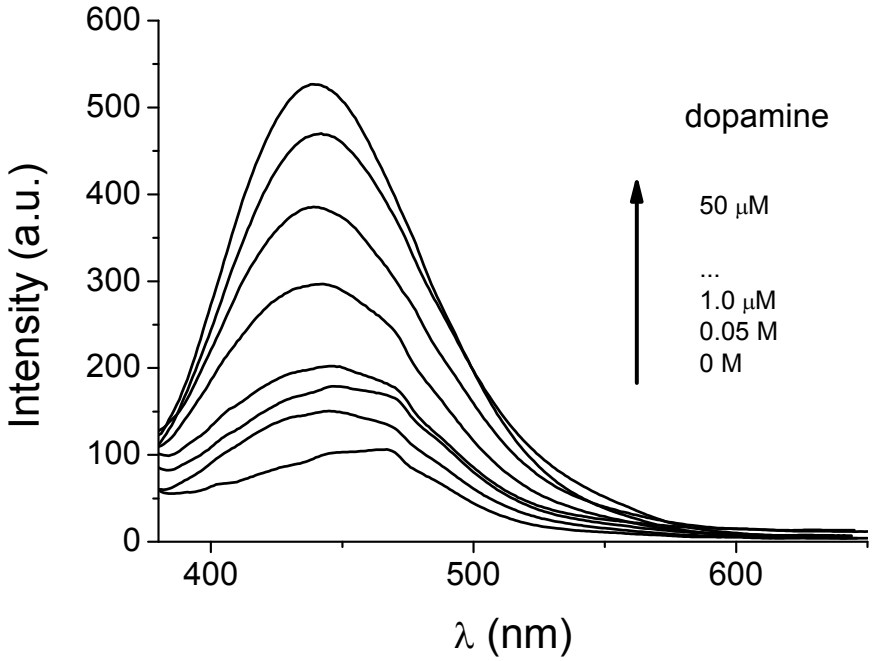


Figure S20. Fluorescence spectra of the control (1 mg/mL, in pH 7.4 PBS buffer) upon titration of dopamine.

4. Supporting table

Table S1. Binding constants of dopamine and some amino acids with β-CD.

Amine	K_a (M^{-1}) [A]	K_a (M^{-1}) [B]
Dopamine	300	240 - 2000 [C] [D]
2-adamantanamine	--	~ 10^7 [E]
L-phenylalanine	750.0	~ 800 [F]
L-serine	--	113 [G]

[A]: The association constant measured according to the Benesi-Hildebrand method by using the absorption spectra (H. A. Benesi, J. H. Hildebrand, *J. Am. Chem. Soc.* **1949**, *71*, 2703-2707; M. V. Rekharsky, Y. Inoue, *Chem. Rev.*, **1998**, *98*, 1875-1880).

[B]: The association constant reported in literature.

[C]: Cao, R.; Villalonga R.; Fragoso, A. *IEE Proc.-Nanobiotechnol.*, **2005**, *152*, 159-164.

[D]: The association constant of dopamine with sulfonated beta-cyclodextrin: "The formation of an electrochemical sensor for the selective detection of dopamine", Claire Harley, Ph.D thesis, 2009, National University of Ireland.

[E]: Wang, Y. H.; Zhu, M. Z.; Ding, X. Y.; Ye, J. P.; Liu, L.; Guo, Q. X.; *J. Phys. Chem. B* **2003**, *107*, 14087-14093.

[F]: Hammitzsch-Wiedemann, M.; Scriba, G. K. E.; *Electrophoresis* **2007**, *28*, 2619-2628.

[G]: K. B. O'Brien, M. Esguerra, C. T. Klug, R. F. Miller, M. T. Bowser, *Electrophoresis* **2003**, *24*, 1227–1235.