

Electronic Supplementary Information

Guanidine-Based Self-Assembled Monolayers on Au Nanoparticles as Artificial Phosphodiesterases

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Experimental Section

Instruments and General Methods. NMR spectra were recorded on a 300-MHz spectrometer. Chemical shifts are reported as δ values in ppm from tetramethylsilane added as an internal standard. Mass spectra were performed by an Electrospray Ionization Time-of-Flight spectrometer.

Materials. DMSO purged 30 min with argon and mQ water were used in the preparation of 80% DMSO. Thioacetic acid was purged for 30 min with argon. Anhydrous CH_2Cl_2 was obtained by distillation over CaCl_2 . Toluene used in NP preparation was distilled over Na and purged 30 min with argon. Other solvents and reagents were commercially available and used without any further purification.

1-Nitro-4-(undec-10-en-1-yloxy)benzene (3) A solution of *p*-nitrophenol (3.89 g, 28.0 mmol) in 50 mL of acetone were refluxed in presence of 10 g of K_2CO_3 . After 30 min 11-bromoundec-1-ene (3.20 g, 13.8 mmol) was added and the mixture refluxed for 24 h. After cooling the mixture was filtrated and the acetone evaporated. The residue was dissolved in 60 mL of dichloromethane and washed with a saturated solution of Na_2CO_3 (3×60 mL). The organic phase was dried over MgSO_4 and the solvent evaporated under reduced pressure. **3** was obtained as white solid (3.45 g; 11.8 mmol 86% yield) pure enough to be used in the following step without any further purification: mp 36-38 °C. ^1H NMR (300 MHz, CDCl_3) δ : 8.19 (d, $J=9$ Hz, 2 H), 6.94 (d, $J=9$ Hz, 2H), 5.81 (m, 1H), 4.95 (m, 2H), 4.04 (t, $J=6$ Hz, 2H), 2.04 (q, $J=6$ Hz, 2 H), 1.81 (m, 2 H), 1.56-1.31 (m, 12 H). ^{13}C NMR (75 MHz, CDCl_3) δ 164.2, 141.2, 139.1, 125.8, 114.3, 114.1, 68.8, 33.7, 29.4, 29.3, 29.2, 29.0, 28.9, 28.8, 25.8. HR ES-MS m/z : $[\text{M}+\text{H}]^+$ Calcd for $\text{C}_{17}\text{H}_{26}\text{NO}_3$ 292.1913, found 292.1911; $[\text{M}+\text{Na}]^+$ Calcd for $\text{C}_{17}\text{H}_{25}\text{NO}_3\text{Na}$ 314.1732, found 314.1719.

(11-(4-Nitrophenoxy)undecyl)thioacetate (4) 1.00 g (3.43 mmol) of **3** were dissolved in 3 mL of degassed thioacetic acid and irradiated for 2 h at 300 nm in a photoreactor. After reaction the solution was cooled down, diluted with 20 ml of CH_2Cl_2 and washed with a saturated solution of Na_2CO_3 (4×20 mL). The organic phase was dried over MgSO_4 and the solvent evaporated under

reduced pressure. A pure sample of **4** was obtained by flash column chromatography (SiO₂, hexane:AcOEt from 40:1 to 30:1) as a pale yellow solid (842 mg, 2.29 mmol, 67% yield): mp 66-68 °C. ¹H NMR (300 MHz, CDCl₃) δ: 8.17 (d, J=9 Hz, 2H), 6.92 (d, J=9 Hz, 2H), 4.03 (t, J=6 Hz, 2H), 2.84 (t, J=7 Hz, 2H), 2.31 (s, 3H), 1.80 (m, 2H), 1.54 (m, 2H), 1.48-1.21 (m, 14H). ¹³C NMR (75 MHz, CDCl₃) δ 196.0, 164.2, 141.2, 125.8, 114.3, 68.8, 30.6, 29.44, 29.40, 29.38, 29.35, 29.21, 29.06, 29.02, 28.9, 28.7, 25.8. HR ES-MS *m/z*: [M+Na]⁺ Calcd for C₁₉H₂₉NO₄SNa 390.1715, found 390.1714.

(11-(4-aminophenoxy)undecyl)thioacetate (5) 402 mg of **4** (1.09 mmol) were dissolved in 10 mL of THF and 50 mg of Pd/C were added. The mixture was stirred at rt under H₂ atmosphere for 36 h. After filtration over Celite and evaporation of the solvent the crude product was purified by flash column chromatography (SiO₂, hexane:AcOEt from 15:1 to 8:1) obtaining **5** as a white solid (228 mg, 0.677 mmol, 62% yield): mp 86-88 °C. ¹H NMR (300 MHz, CDCl₃) δ: 6.72 (d, J=9 Hz, 2 H), 6.62 (d, J=9 Hz, 2H), 3.86 (t, J=6 Hz, 2H), 3.48 (br s, 2H), 2.85 (t, J= 6 Hz, 2H), 2.30 (s, 3H), 1.71 (m, 2H), 1.54 (m, 2H), 1.46-1.20 (m, 14H). ¹³C NMR (75 MHz, CDCl₃) δ 196.0, 152.3, 139.4, 116.4, 115.5, 68.5, 30.5, 29.39, 29.36, 29.35, 29.31, 29.28, 29.26, 29.01, 28.97, 28.7, 25.9. HR ES-MS *m/z*: [M+Na]⁺ Calcd for C₁₉H₃₂NO₂S 338.2154, found 338.2166.

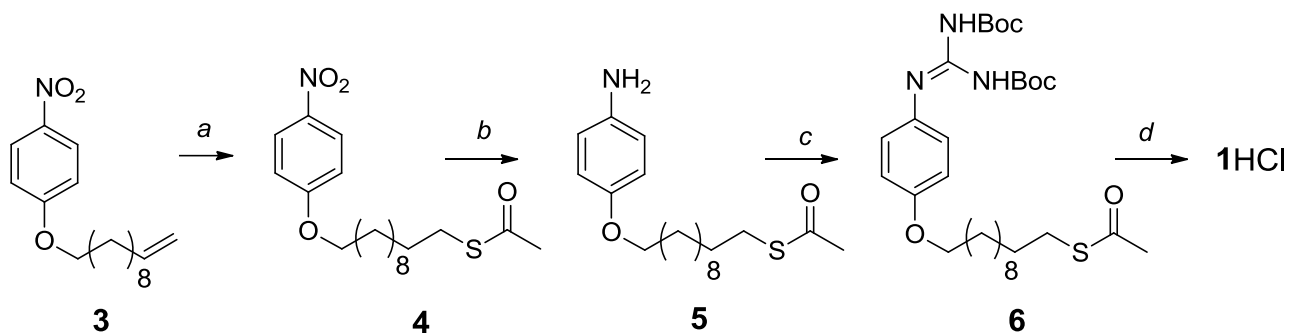
(11-(4-(*N,N'*-bis(*tert*-butoxycarbonyl)-guanidine)phenoxy)undecyl)thioacetate (6) 280 mg of **5** (0.830 mmol), 373 mg (0.953 mmol) of *N,N'*-bis(*tert*-butoxycarbonyl)-*N''*-triflylguanidine and 0.15 mL of Et₃N were dissolved in 20 mL of anhydrous CH₂Cl₂. The solution was stirred at rt for 40 h, extracted with a 2 M NaHSO₄ aqueous solution (50 mL), separated, and washed with a saturated NaHCO₃ water solution (50 mL). The aqueous phase was extracted with dichloromethane (3 × 30 mL), and the combined organic phases were washed with a saturated NaCl aqueous solution, dried over MgSO₄, and evaporated. The crude material was purified by column chromatography (SiO₂, hexane:AcOEt from 40:1) giving compound **6** as a white solid (410 mg, 0.707 mmol, 85% yield): mp 92-94 °C. ¹H NMR (300 MHz, CDCl₃) δ: 11.65 (br s, 1H), 10.18 (br s, 1H), 7.43 (d, J=9 Hz, 2H), 6.82 (d, J=9 Hz, 2H), 3.90 (t, J=6 Hz, 2H), 2.84 (t, J=6 Hz, 2H), 2.30 (s,

3H), 1.74 (m, 2H), 1.51 (s, 9H), 1.47 (s, 9H), 1.45-1.24 (m, 16H). ^{13}C NMR (75 MHz, CDCl_3) δ : 195.6, 163.3, 156.3, 153.5, 153.2, 129.4, 123.7, 114.6, 83.4, 79.4, 68.1, 30.5, 29.42, 29.40, 29.37, 29.34, 29.26, 29.13, 29.04, 29.00, 28.7, 28.1, 28.0, 25.9. HR ES-MS m/z : $[\text{M}+\text{H}]^+$ Calcd for $\text{C}_{30}\text{H}_{50}\text{N}_3\text{O}_6\text{S}$ 580.3420, found 580.3396.

11-(4-guanidinephenoxy)undecane-1-thiol Hydrochloride (1·HCl) 137 mg of **6** (0.236 mmol) were dissolved in 10 mL of a 2 M solution of hydrochloric acid in MeOH/ H_2O 10:1. After refluxing under argon for 6 h, the solvent mixture was evaporated under reduced pressure obtaining compound **1**·HCl as a pale yellow glassy solid (88 mg, 0.235 mmol, 100% yield). ^1H NMR (300 MHz, CD_3OD) δ : 7.19 (d, $J=9$ Hz, 2H), 6.99 (d, $J=9$ Hz, 2H), 3.99 (t, $J=6$ Hz, 2H), 2.49 (t, $J=9$ Hz, 2H), 1.78 (m, 2H), 1.59 (m, 2H), 1.52-1.28 (m, 14H). ^{13}C NMR (75 MHz, CD_3OD) δ : 160.2, 128.8, 128.0, 125.2, 116.7, 69.3, 35.2, 30.7, 30.6, 30.5, 30.3, 30.2, 29.4, 27.1, 25.0. HR ES-MS m/z : $[\text{M}+\text{H}]^+$ Calcd for $\text{C}_{18}\text{H}_{32}\text{N}_3\text{OS}$ 338.2266, found 338.2283.

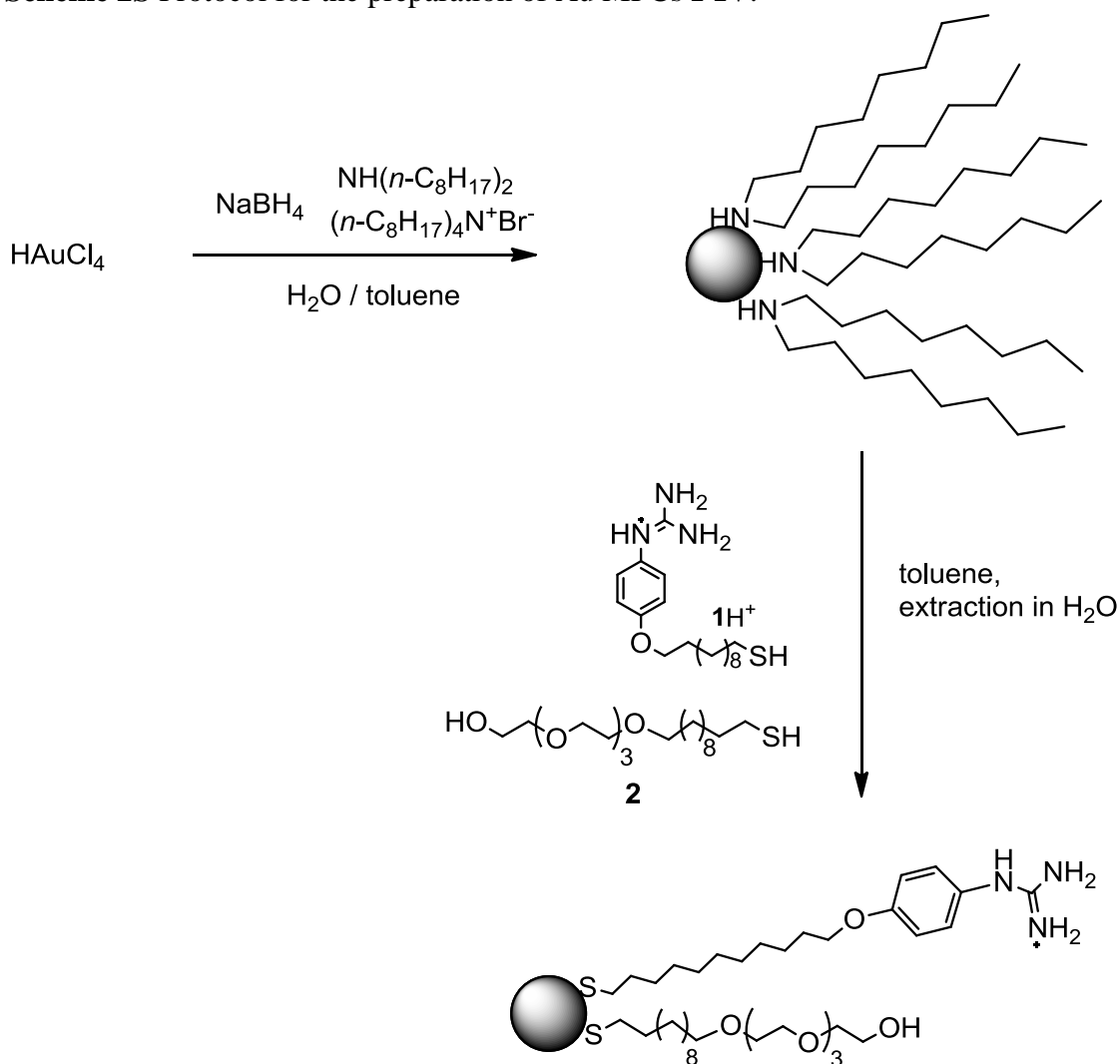
Nanoparticle preparation: 121 mg of $\text{HAuCl}_4 \cdot 3\text{H}_2\text{O}$ (0.307 mmol) were dissolved in 8.5 mL of mQ water. Separately a solution of tetraoctylammonium bromide (6.50 g, 11.9 mmol) in degassed toluene (300 mL) was prepared. The aqueous solution of chloroauric acid was extracted with the TOABr solution (3×15 mL) causing the transfer of the gold to the organic phase. Dioctylamine was added to the remaining amount of the TOABr solution and this mixture added to the collected organic phases containing Au^{III} . The resulting mixture was vigorously stirred under argon for 30 min. Then 57 mg of NaBH_4 (1.52 mmol) in 4 mL of water were added and the mixture stirred at rt under argon for 3 h. After that time the aqueous phase was removed. The nanoparticle solution was divided in four batches and thiols **1** and **2**, in different ratios and previously dissolved in 1 mL of MeOH, were added under vigorous stirring. After a few minutes the formation of a dark brown precipitate was observed and after 10 min from the formation of the precipitate 10 mL of mQ water were added and the solution stirred for 2 more hours. The aqueous phase was separated and washed with toluene (2×20 mL), diethyl ether (3×20 mL) and AcOEt (3×20 mL). Finally, the water was removed by lyophilization the Au MPCs were obtained as dark brown solid.

Scheme 1S Synthesis of **1H⁺**



^a AcSH, hv 360 nm; ^b H₂, Pd/C, THF, rt; ^c *N,N*-Bis(tert-butoxycarbonyl)-*N'*-triflylguanidine, Et₃N, DCM, rt; ^d 2M HCl, MeOH/H₂O 10:1, reflux.

Scheme 2S Protocol for the preparation of Au MPCs **I-IV**.



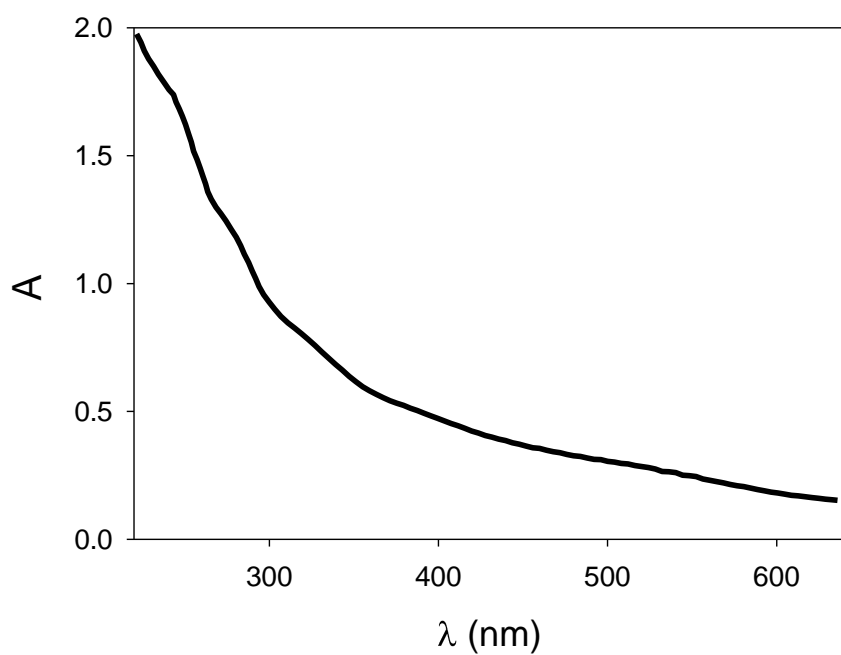


Figure 2S UV-Vis spectrum of Au MPCs I (100 $\mu\text{g}/\text{mL}$) in H_2O

TGA Experiments

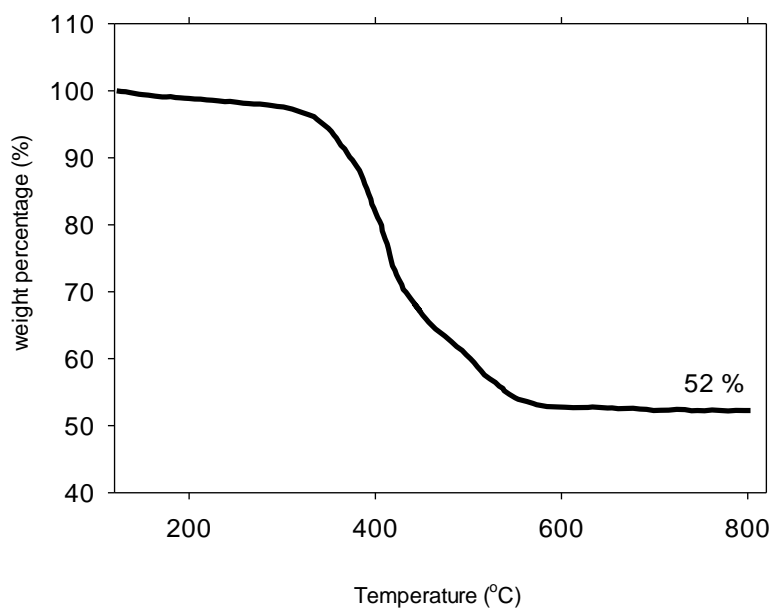


Figure 3S TGA plot of AuNPs I

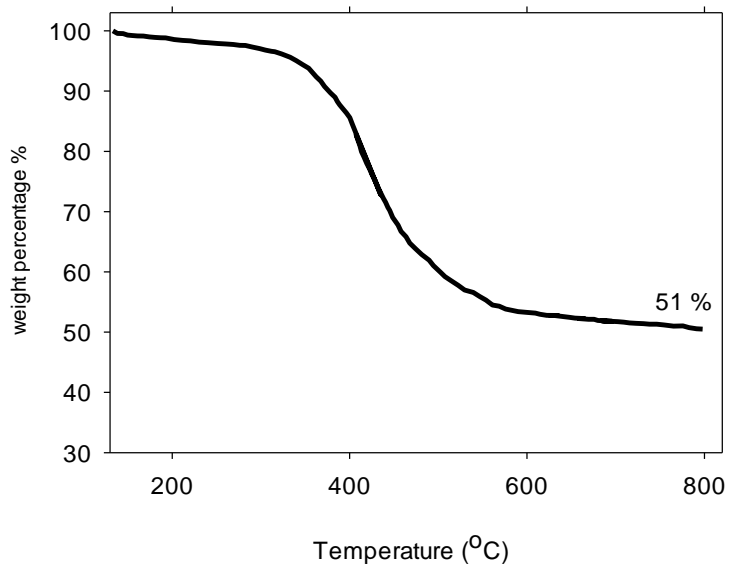


Figure 4S TGA plot of AuNPs II

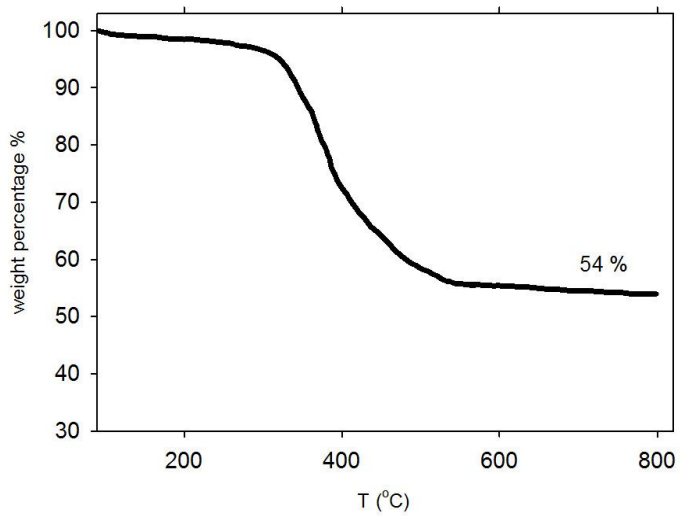


Figure 5S TGA plot of AuNPs III

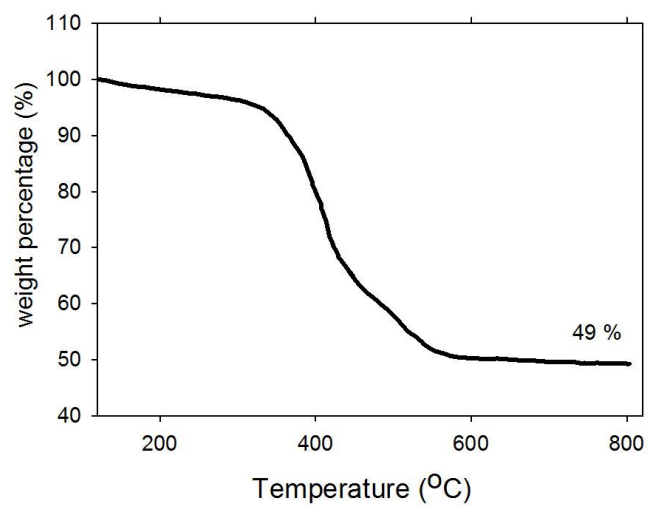


Figure 6S TGA plot of AuNP IV

Determination of the mole fractions in the monolayer

The determination of the molar fraction x_1 of compound **1** in the monolayers of Au NPs **II** and **III** (see Table 1) has been carried out using equation (1S) where M is the total mass of the monolayer, n_1 and n_2 the molar amounts of thiols **1** and **2**, and PM_1 and PM_2 the corresponding molecular weights.

$$M = n_1 \cdot PM_1 + n_2 \cdot PM_2 \quad (1S)$$

M has been determined by the TGA analysis and n_1 by potentiometric titrations (see Table 1 and Figure 1). The molecular weights are known: $PM_1 = 373.19$, $PM_2 = 380.26$. Therefore the unknown quantity n_2 is simply calculated. x_1 and x_2 are given by (2S).

$$x_1 = n_1 / (n_1 + n_2), \quad x_2 = 1 - x_1 \quad (2S)$$

TEM Analysis

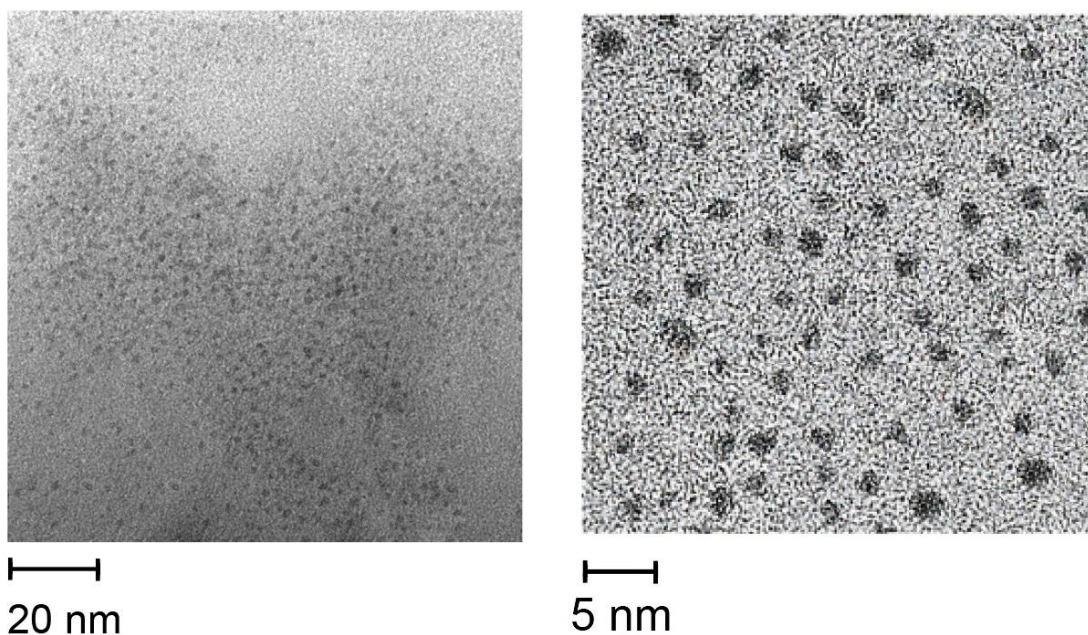
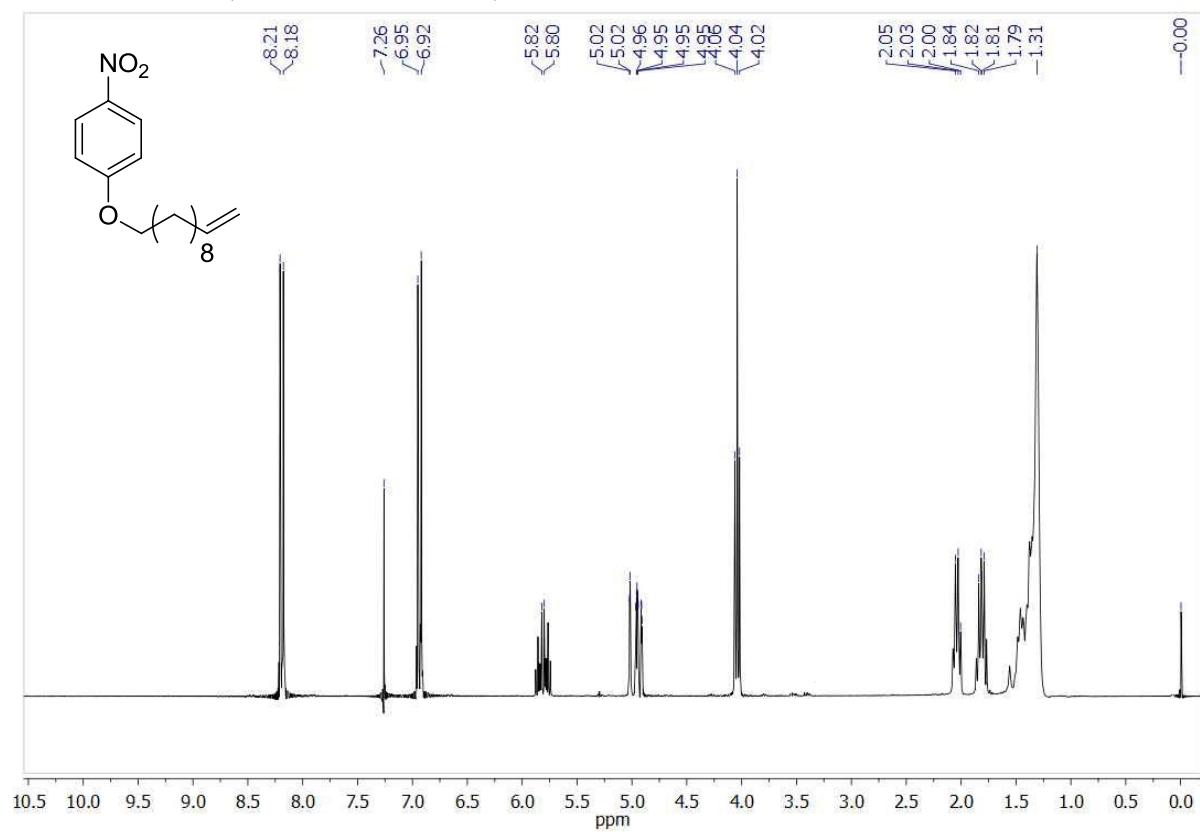
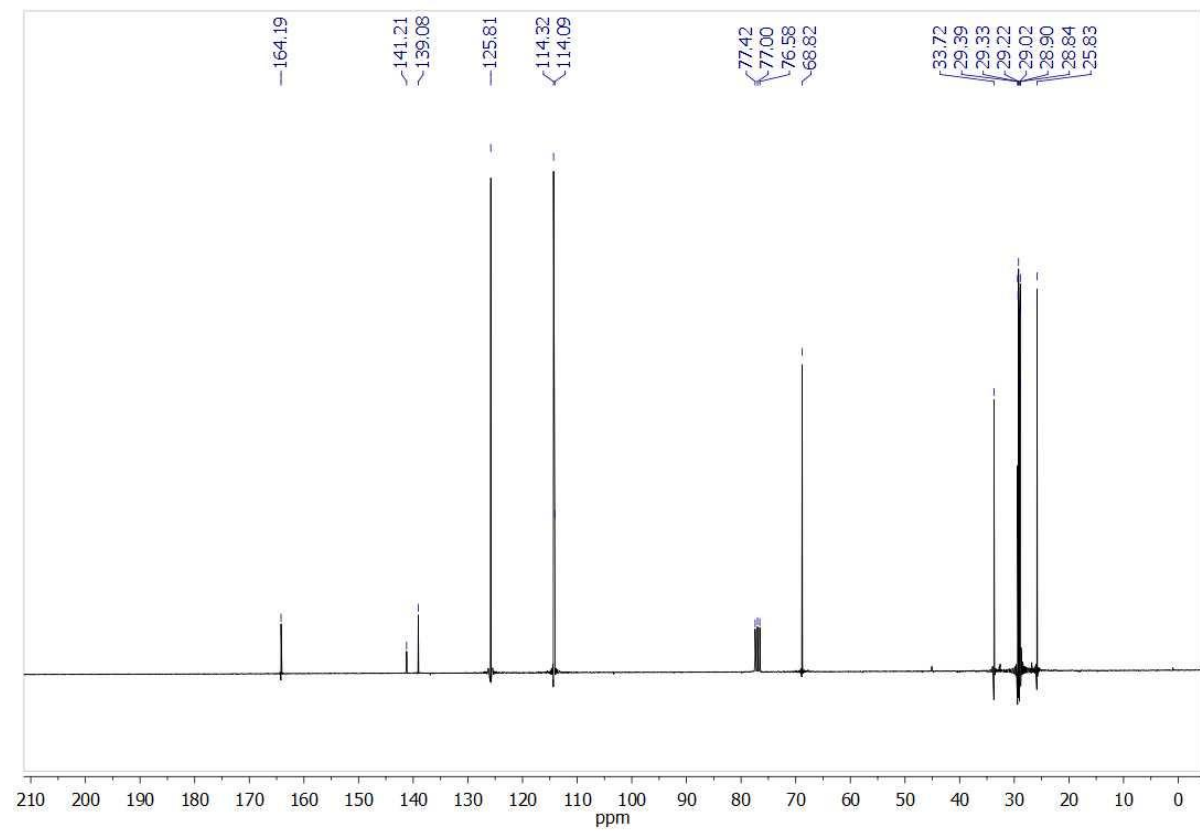


Figure 7S HR-TEM image of Au NPs **I**. Average diameter: 1.8 ± 0.2 nm

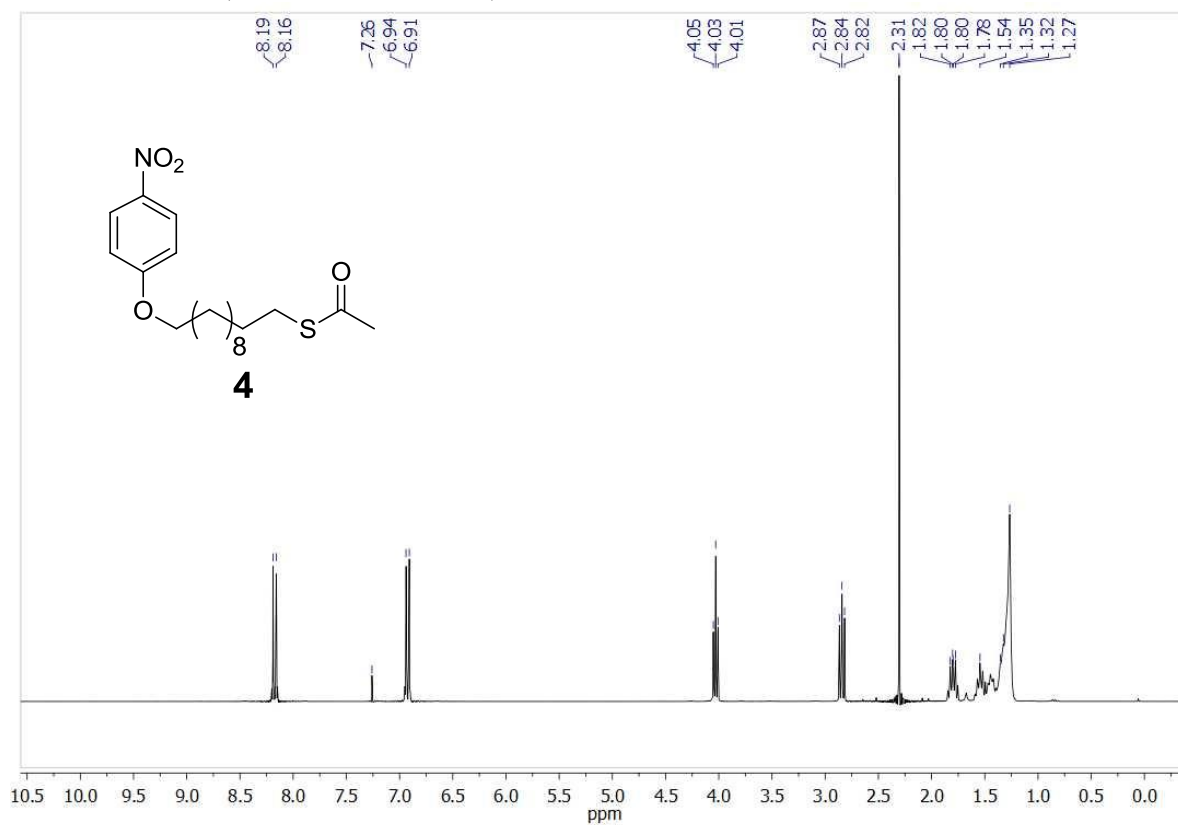
^1H NMR of **3** (CDCl_3 , 300 MHz)



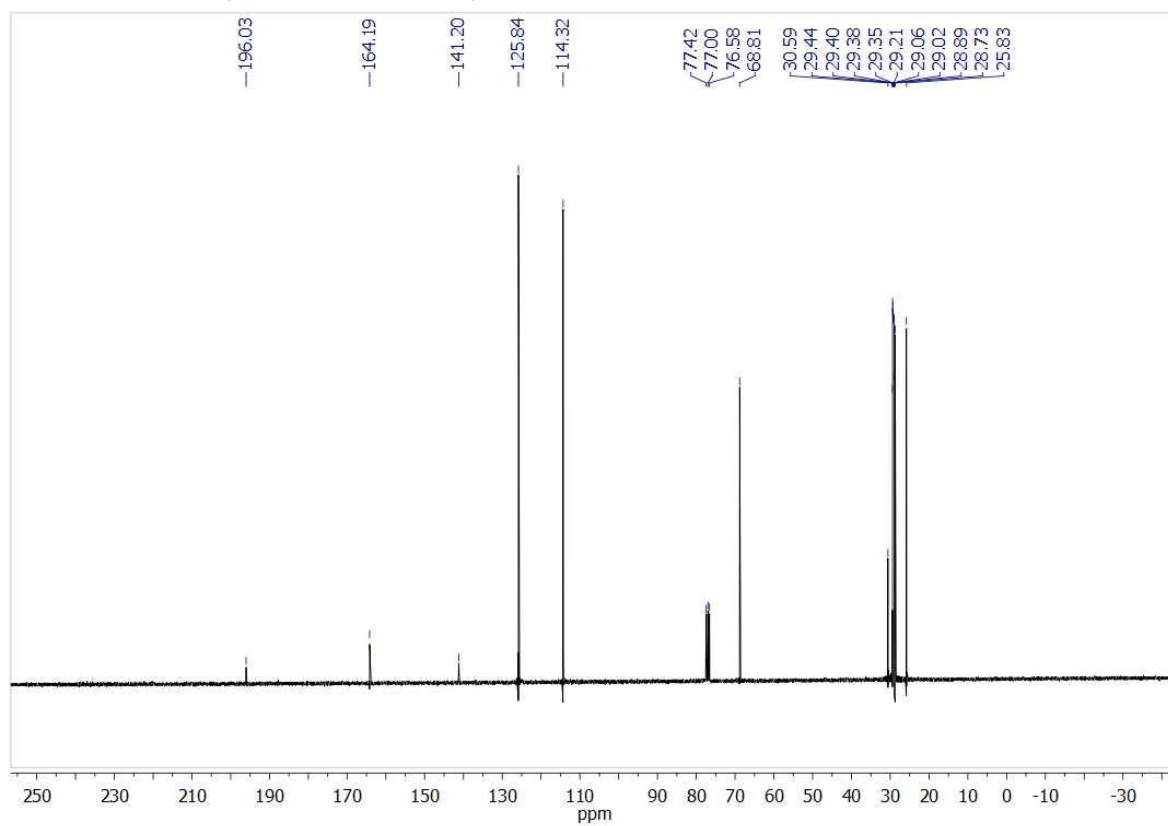
^{13}C NMR of **3** (CDCl_3 , 75 MHz)



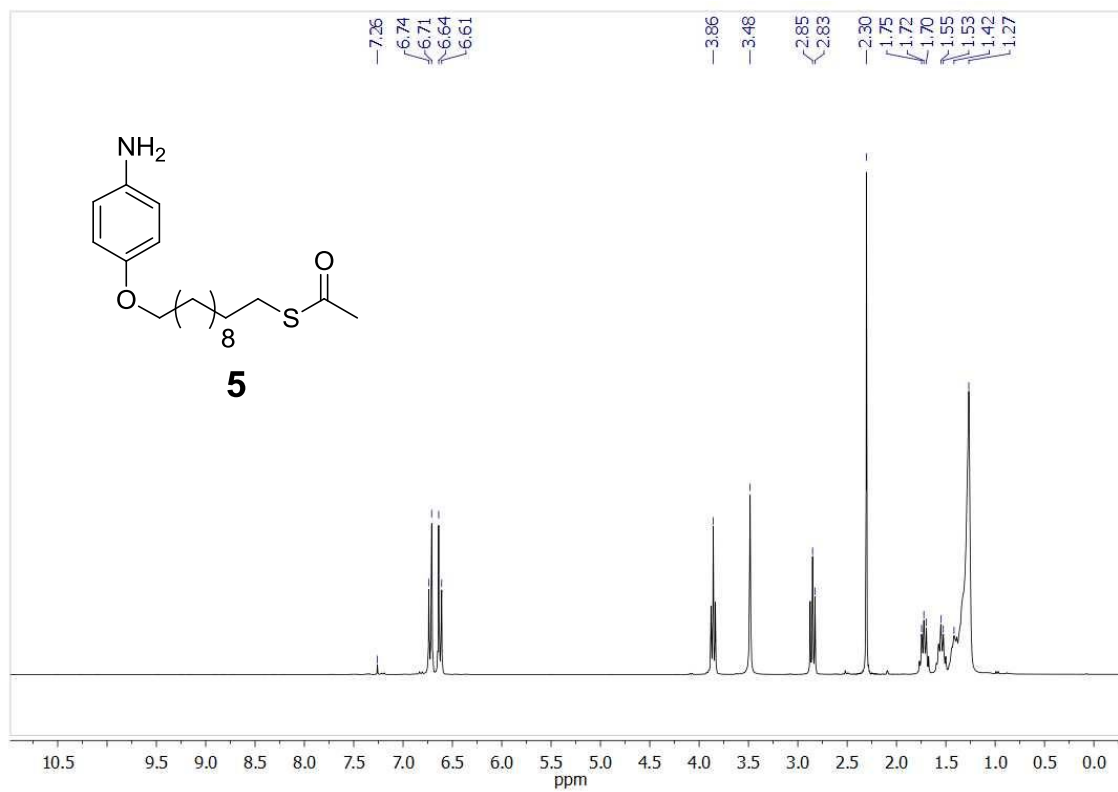
^1H NMR of **4** (CDCl_3 , 300 MHz)



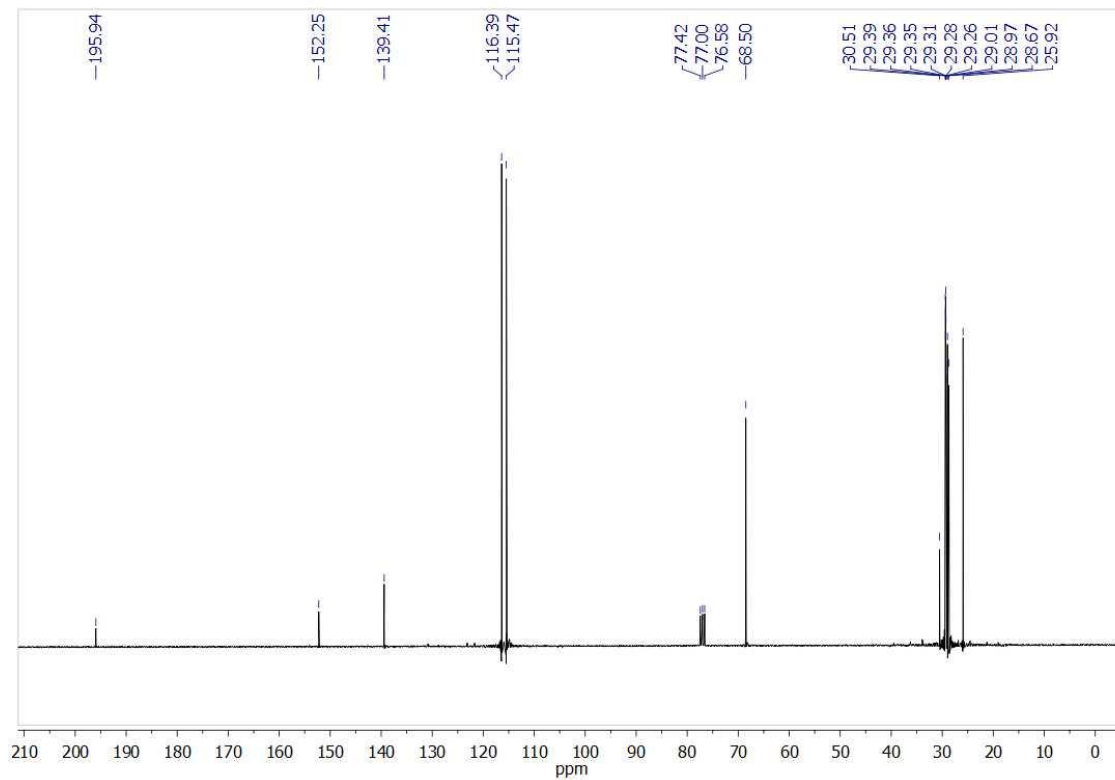
^{13}C NMR of **4** (CDCl_3 , 75 MHz)



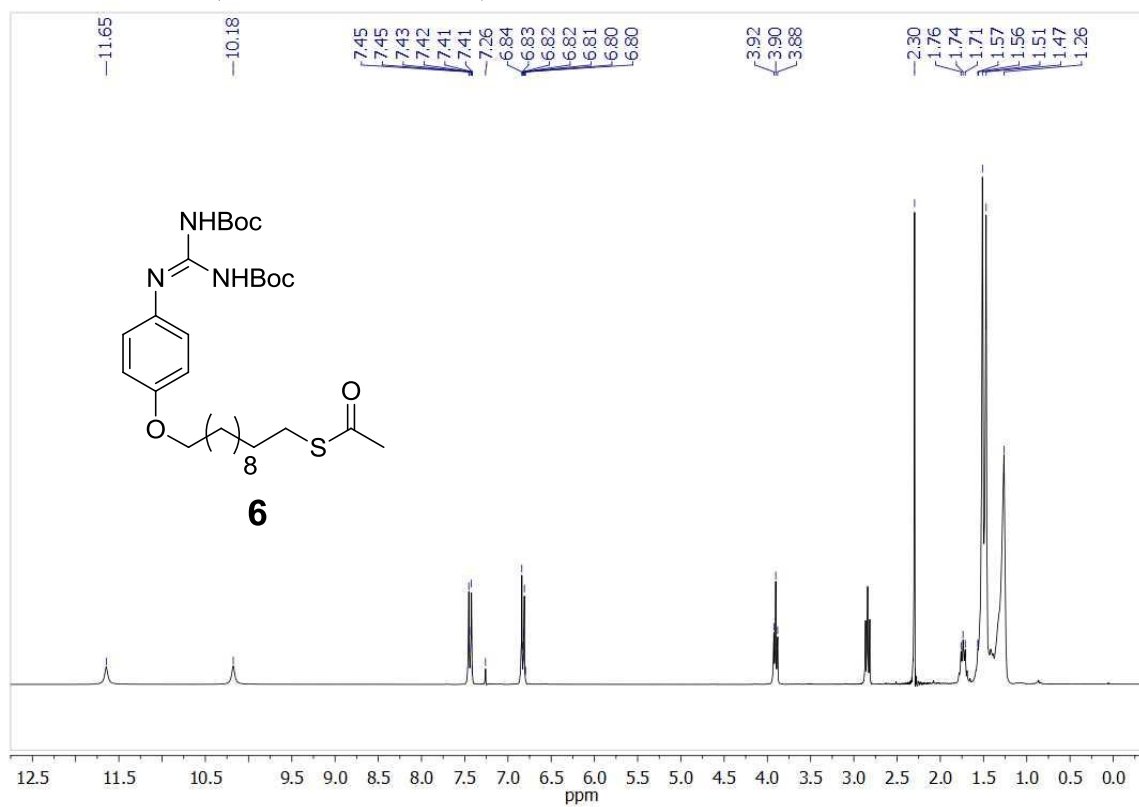
^1H NMR of **5** (CDCl_3 , 300 MHz)



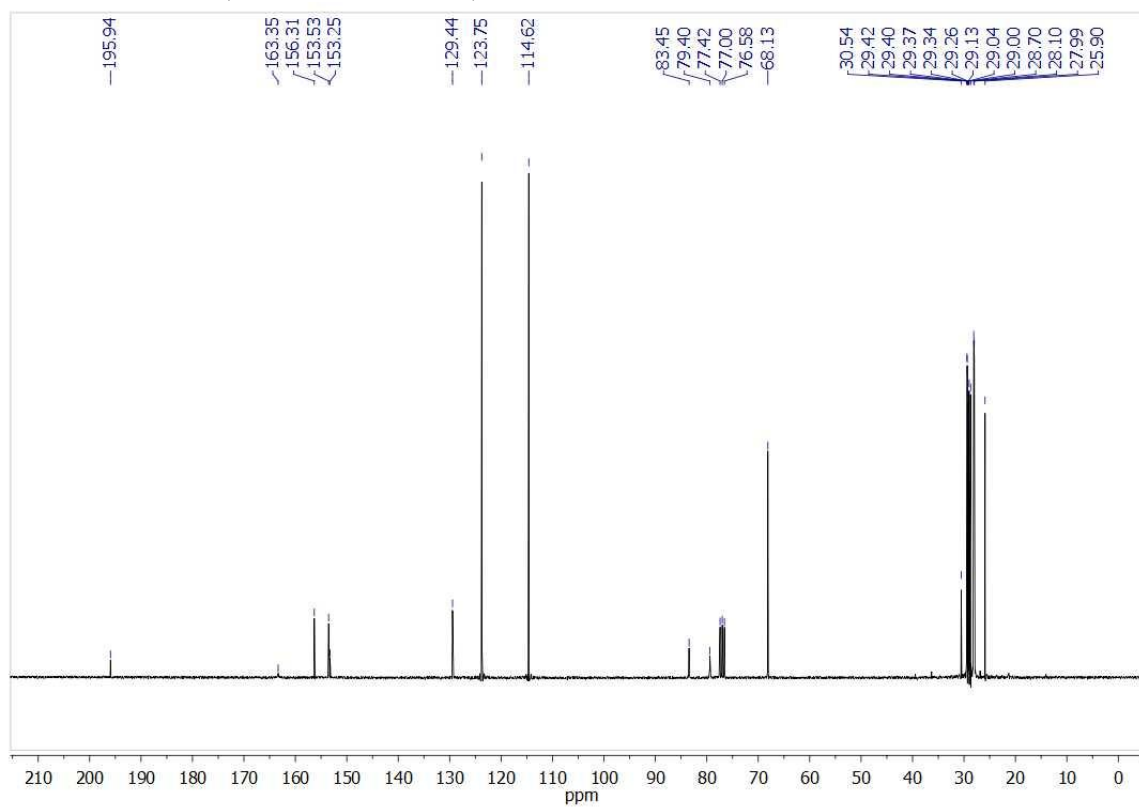
^{13}C NMR of **5** (CDCl_3 , 75 MHz)



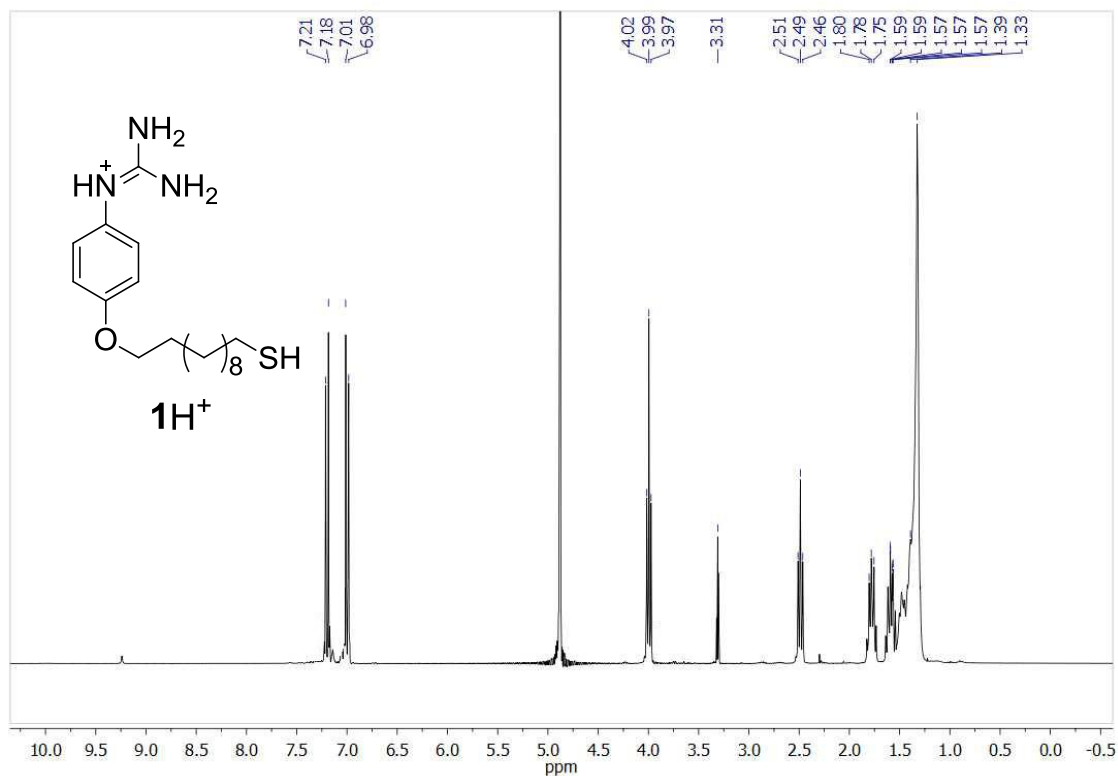
¹H NMR of **6** (CDCl₃, 300 MHz)



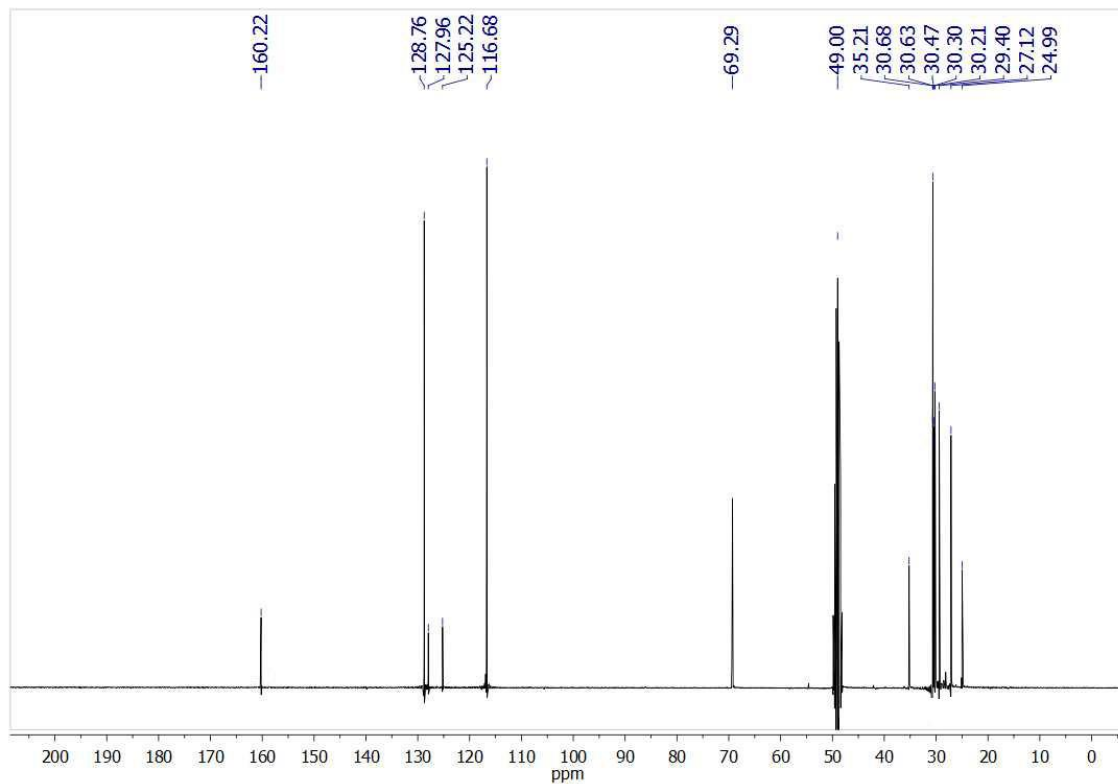
¹³C NMR of **6** (CDCl₃, 75 MHz)



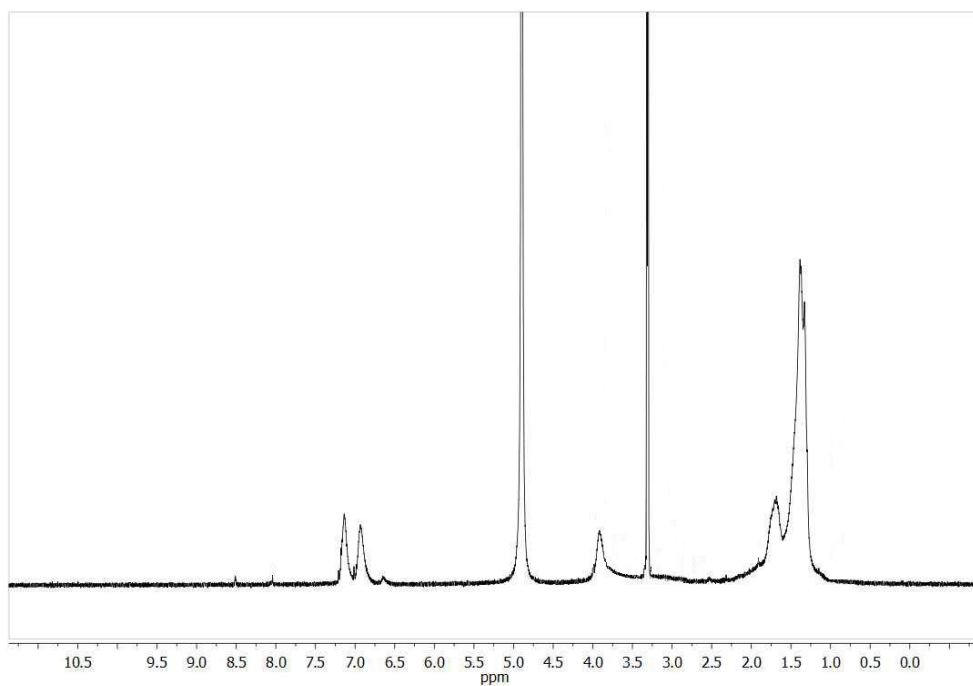
^1H NMR of $1\cdot\text{HCl}$ (CD_3OD , 300 MHz)



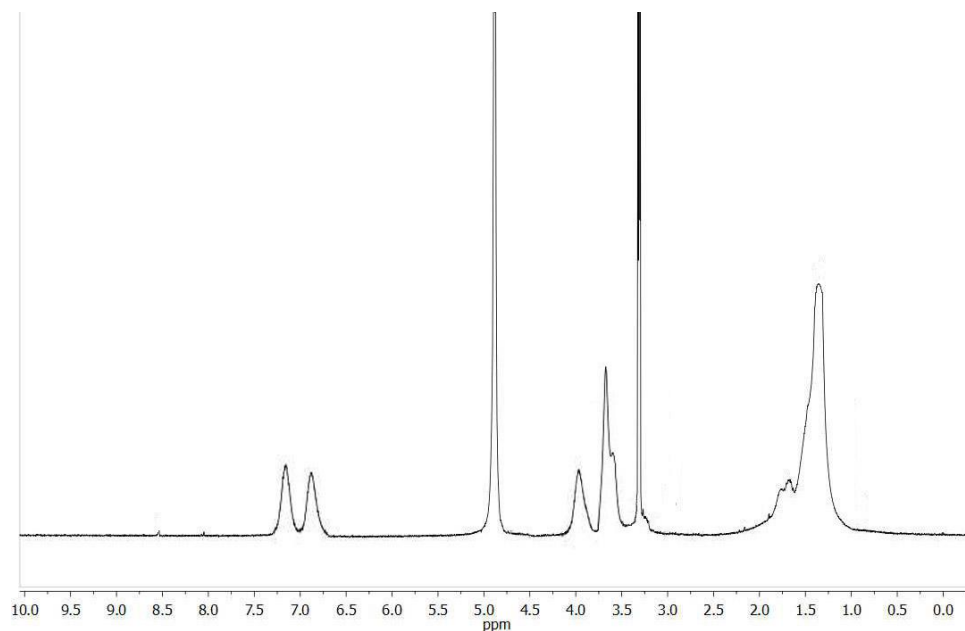
^{13}C NMR of $1\cdot\text{HCl}$ (CD_3OD , 75 MHz)



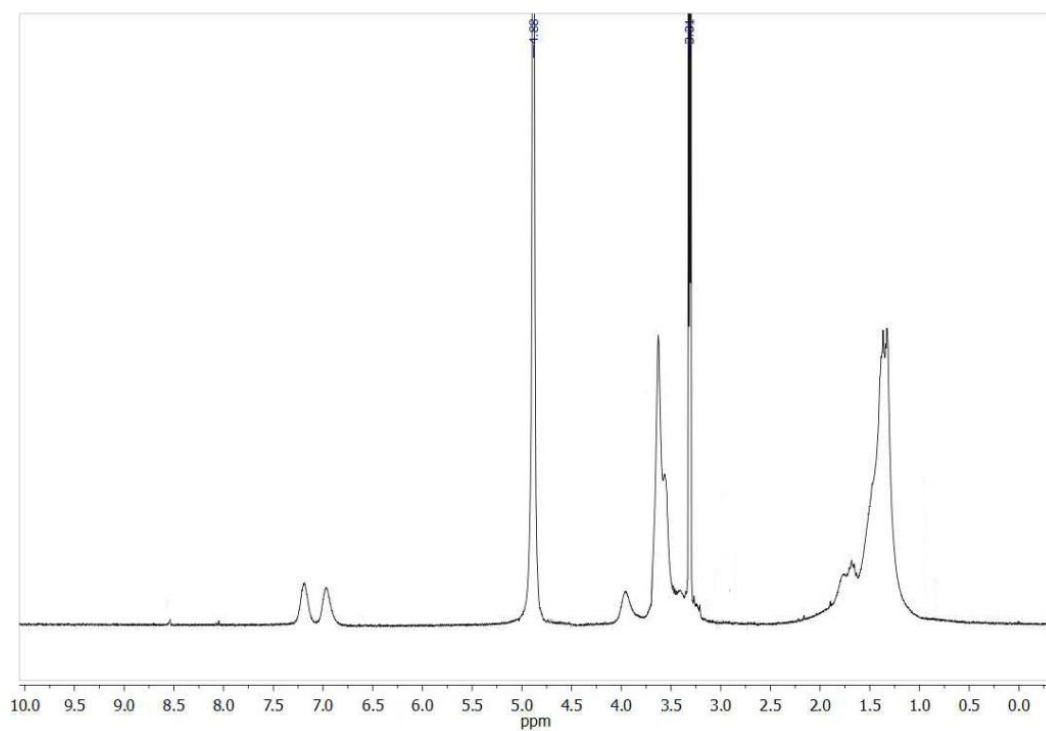
^1H NMR of Au NPs **I** (CD_3OD , 300 MHz)



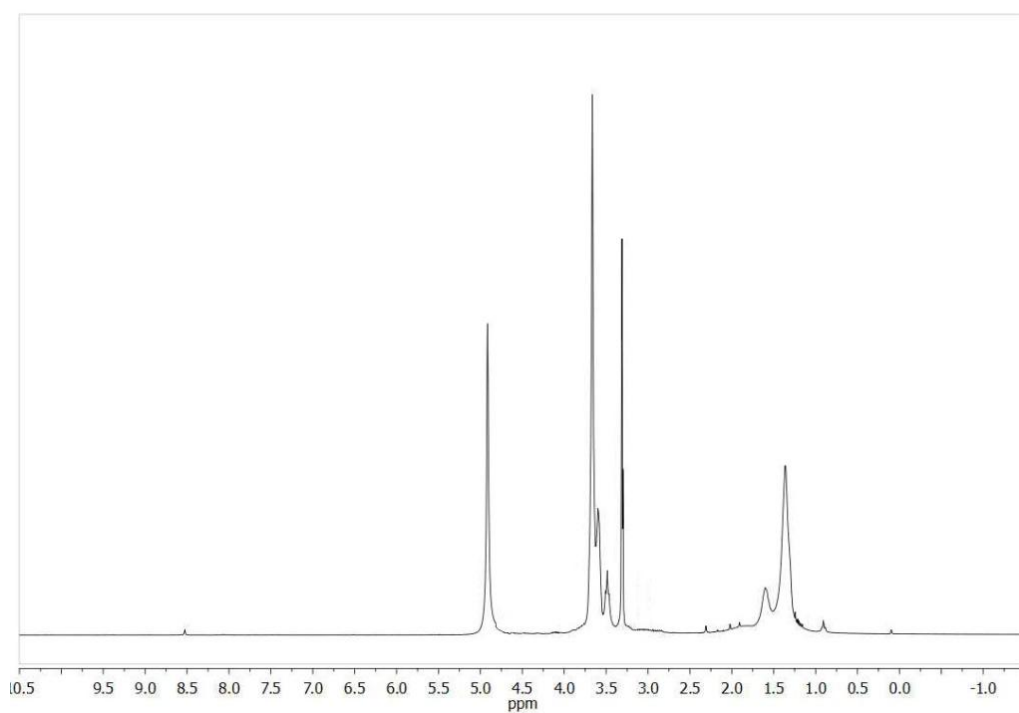
^1H NMR of Au NPs **II** (CD_3OD , 300 MHz)



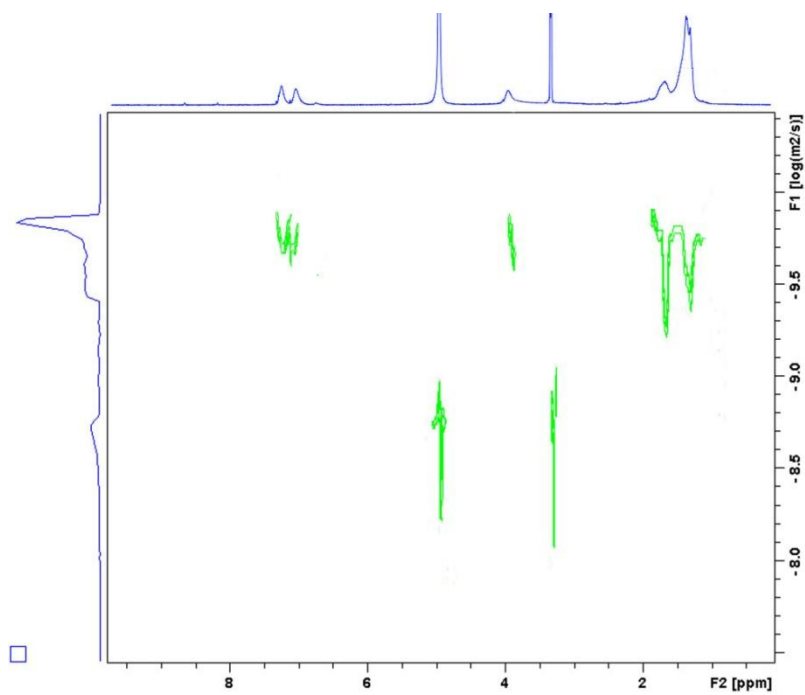
^1H NMR of Au NPs **III** (CD_3OD , 300 MHz)



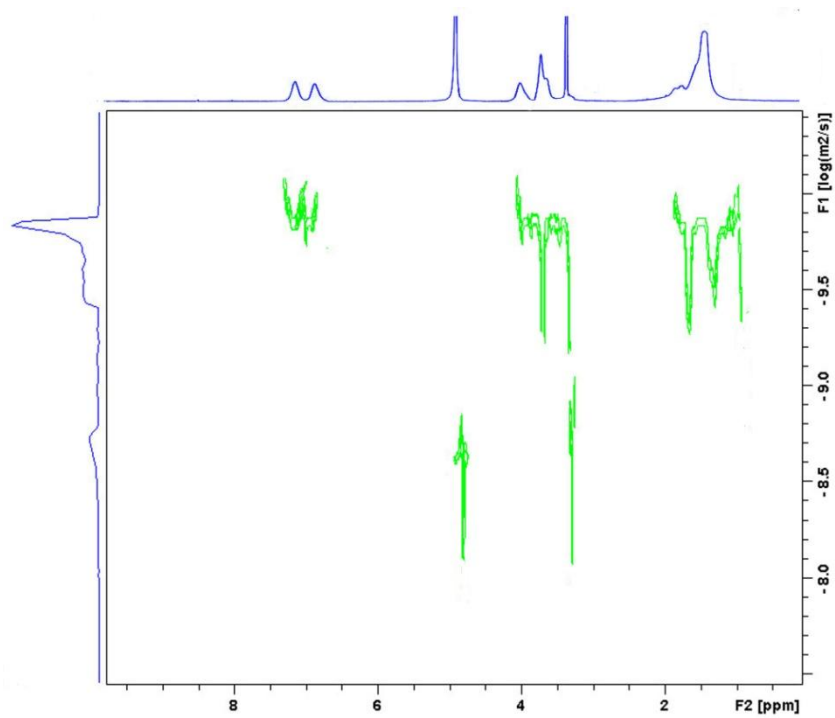
^1H NMR of Au NPs **IV** (CD_3OD , 300 MHz)



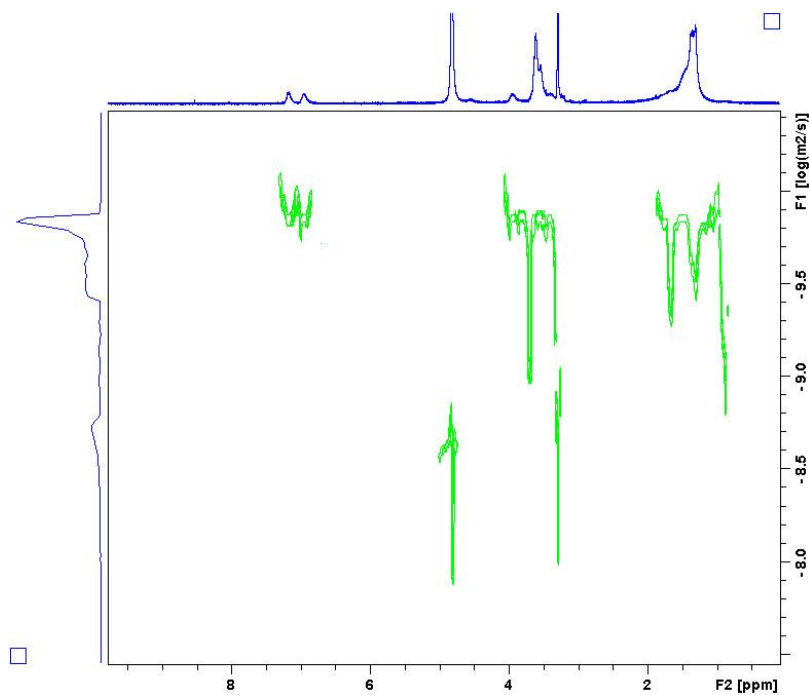
DOSY NMR of Au NPs **I** (CD₃OD, 300 MHz)



DOSY NMR of Au NPs **II** (CD₃OD, 300 MHz)



DOSY NMR of Au NPs **III** (CD₃OD, 300 MHz)



DOSY NMR of Au NPs **IV** (CD₃OD, 300 MHz)

