Supporting Information

Detection and identification of designer drugs by nanoparticle-based NMR chemosensing

Luca Gabrielli,[‡] Daniele Rosa-Gastaldo,[‡] Marie-Virginie Salvia,[†] Sara Springhetti, Federico Rastrelli, Fabrizio Mancin*

Department of Chemical Sciences Università degli Studi di Padova via Marzolo 1, 35131 Padova (Italy).

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1. Experimental Procedures

General: Solvents were purified by standard methods. All commercially available reagents and substrates were used as received. Compounds **S1-4**, were prepared as detailed in Section 7 of this SI.

TLC analyses were performed using Merck 60 F_{254} precoated silica gel glass plates. Column chromatography was carried out on Macherey-Nagel silica gel 60 (70-230 mesh).

NMR spectra were recorded using a Bruker AV III 500 spectrometer operating at 500 MHz for ^{1}H, 125.8 MHz for ^{13}C. Chemical shifts are reported relative to internal Me_4Si. Multiplicity is given as follow: s = singlet, d = doublet, t = triplet, q = quartet, qn = quintet, m = multiplet, br = broad peak.

ESI-MS mass spectra were obtained with an Agilent Technologies LC/MSD Trap SL mass spectrometer.

HRMS mass spectra were obtained with a Mariner Applied Biosystem (API-TOF) mass spectrometer (MeOH, 0.5% formic acid).

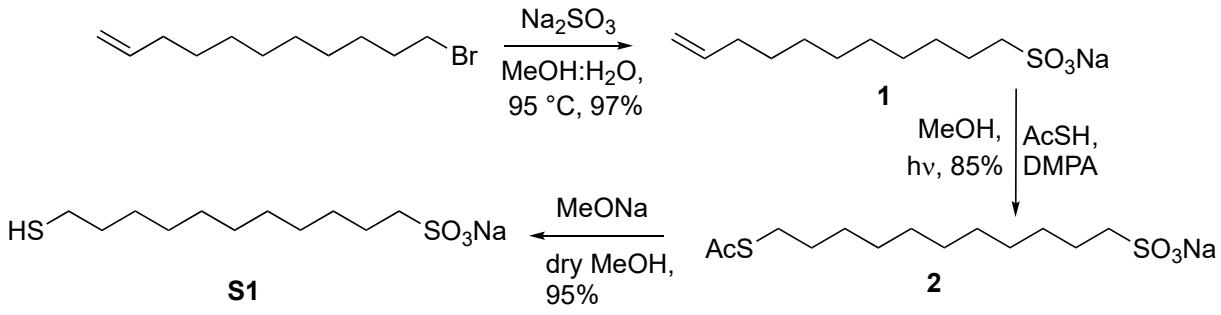
TEM images were recorded on a Jeol 300 PX electron microscope. One drop of sample was placed on the sample grid and the solvent was allowed to evaporate. TEM images were analysed with ImageJ software to measure the diameters distribution and average values.

TGA were run on 1 mg nanoparticle samples using a Q5000 IR model TA instrument from 30 to 1000 °C under a continuous air flow.

Fluorescence spectra were recorded on a Perkin Elmer LS50B fluorimeter.

2. Synthesis of Sulfonate thiols (S1, S2, S3, S4)

Thiol **S1** was prepared as reported in the following scheme:



Sodium undec-10-ene-1-sulfonate (1).

11-bromoundec-1-ene (510 mg, 2.190 mmol, 1 equiv) was dissolved in 9 mL of H_2O and 4 ml of MeOH. Na_2SO_3 (554 mg, 4.400 mmol, 2 equiv) was added and the mixture was stirred under reflux overnight. Then the solvent was evaporated under reduced pressure and the residue was purified by flash column chromatography (DCM:MeOH from 9:1 to 1:1), affording the desired sulfonate compound **1** (533 mg, 97%).

^{1}H-NMR (D_2O, 200 MHz): δ 1.26 (br s, 12H) 1.68 (m, 2H), 1.99 (m, 2H), 2.82 (m, 2H), 4.92 (m, 2H), 5.80 (m, 1H).
^{13}C-NMR (D_2O, 500 MHz): δ 23.95, 27.68, 27.70, 28.12 28.24 28.40 28.46, 33.11, 51.08, 113.89, 140.46.
MS (ESI) *m/z*: 233.1 ($[M]^-$)

Sodium 11-(acetylthio)undecane-1-sulfonate (2).

Sulfonate **1** (200 mg, 0.78 mmol, 1 equiv) was dissolved in 2 mL of MeOH and the solution was degased under N_2 bubbling for 30 min. Then thioacetic acid (220 µl, 3.120 mmol, 4 equiv) and 2,2-dimethoxy-2-phenylacetophenone (DMPA, 20 mg, 0.078 mmol, 0.1 equiv) were added and the mixture was stirred under UV irradiation (365 nm) for 2.5 h. The solvent was evaporated under vacuum and the crude was purified by flash column chromatography (DCM:MeOH from 9:1 to 1:1), affording the desired sulfonate compound **2** (220 mg, 85%).

1H-NMR (D_2O, 200 MHz): δ 1.25 (br s, 14H), 1.39 (m, 2H), 1.64 (m, 2H), 2.31 (s, 3H, C*H*3-), 2.69 (m, 4H).
MS (ESI) *m/z*: 309.1($[M]^-$)

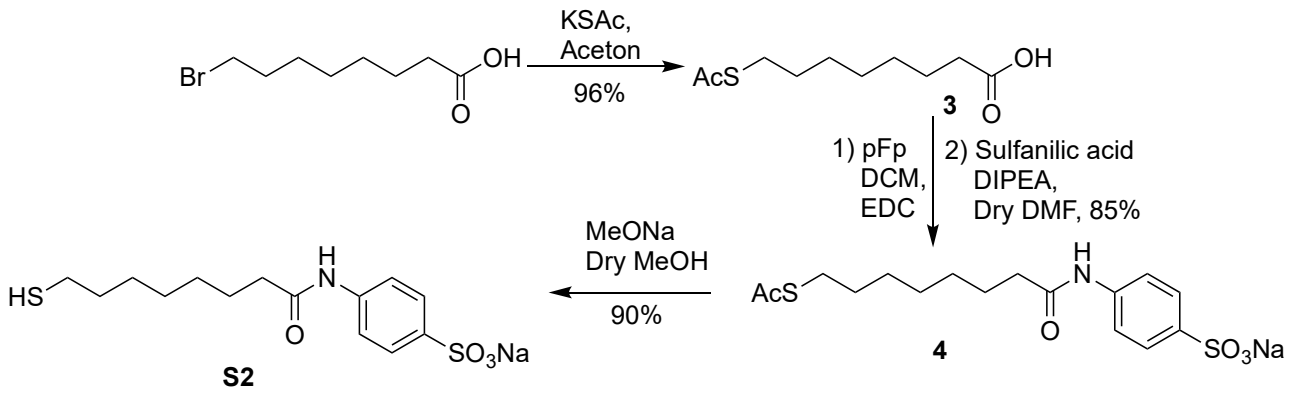
Sodium 11-mercaptoundecane-1-sulfonate (S1).

The acetylated sulfonate **2** (84 mg, 0.250 mmol, 1 equiv) was dissolved in 2 mL of dry MeOH and then sodium methoxide was added (32 mg, 0.590 mmol, 2.4 equiv). After stirring for 2.5 hours under N_2 atmosphere, the reaction was quenched adding IR 120H$^+$ resin until pH neutralization, then the resin was filtered off, the solvent was evaporated, giving the deprotected Sodium 11-mercaptoundecane-1-sulfonate **S1** (70 mg, yield 95%), that was freshly used for the nanoparticles' synthesis.

^{1}H-NMR (MeOD, 300 MHz): δ 1.32 (br s, 14H), 1.57 (m, 2H), 1.78 (m, 2H), 2.49 (m, 2H), 2.79 (m, 2H).
^{13}C-NMR (MeOD, 500 MHz): δ 23.78, 23.96, 27.71, 28.10, 28.24, 28.40, 28.46, 28.55, 33.01, 51.08.
TOF ES$^-$ HRMS: [M$^-$] calcd. for $C_{11}H_{23}O_3S_2$=267.109. Found =267.113

Spectroscopic data are in agreement with those reported in literature.[1]

Thiol **S2** was prepared as reported in the following scheme:



8-(Acetylthio)octanoic acid (3).

8-bromooctanoic acid (4 g, 17.928 mmol, 1 eq) was dissolved in 100 mL of aceton, then KSAc (2.46 g, 21.514 mmol, 1.2 eq) was added and the mixture was stirred at room temperature overnight. Then the solvent was evaporated under reduced pressure and the residue was purified by flash column chromatography (PE:EtOAc from 8:2 to 7:3), affording the desired compound **3** (3.76 g, 96%).

^{1}H-NMR ($CDCl_3$, 500 MHz): δ 1.35 (m, 6H), 1.58 (m, 2H), 1.64 (m, 2H), 2.30 (s, 3H), 2.36 (t, J=7.5Hz, 2H), 2,89 (t, , J=7.3Hz 2H).

^{13}C-NMR ($CDCl_3$, 300 MHz): δ 24.54, 28.54, 28,70, 28.85, 29.07, 29.41, 30.64, 33.98, 179.98, 196.12.

4-(8-(acetylthio)octanamido)benzenesulfonate · DIPEA (4).

Compound **3** (1 g, 4.581 mmol, 1 equiv) was dissolved in 10 mL of dry DCM and then pentafluoro phenol (pFp 626 µl, 5.955 mmol, 1.3 equiv) and EDC*HCl (1.14 g, 5.955 mmol, 1.3 equiv) were added. The mixture was stirred for 20 h, then the solvent was evaporated under reduced pressure. The crude was dissolved in dry DMF (15 mL), then sulfanilic acid (937 mg, 5.406 mmol, 1.18 equiv) and DIPEA (3 mL, 17.408 mmol, 3.8 equiv) were added; the reaction was stirred at 68°C and monitored by TLC (7:3 Tol:MeOH). After completion the solvent was evaporated and the crude was purified by flash column chromatography (Tol:MeOH 7:3), affording the desired sulfonate compound **4** (1.54 g, 85%).

^{1}H-NMR (MeOD, 500): δ 1.39 (m br, 27H), 1.57 (m br, 2H), 1.70 (m br, 2H), 2.32 (s, 3H), 2.42 (t, J=7.5 Hz, 2H), 2.88 (t, J=7.3 Hz, 2H), 3.23 (q, J=7.4 Hz, 2H), 3.72 (hept, J=6.6 Hz, 2H), 7.38, 7.76 (m, 4H)

^{13}C- NMR (MeOD, 500): 11.93, 16.03, 17.47, 25.36, 28.27, 28.47, 28.55, 28.77, 29.35, 36.63, 42.51, 54.47, 118.82, 126.31, 140.27, 140.57, 173.33, 196.06

TOF ES$^-$ HRMS: [M$^-$] calcd. for $C_{16}H_{22}O_5NS_2$=372.094 Found =372.102.

Sodium 4-(8-mercaptooctanamido)benzenesulfonate (S2).

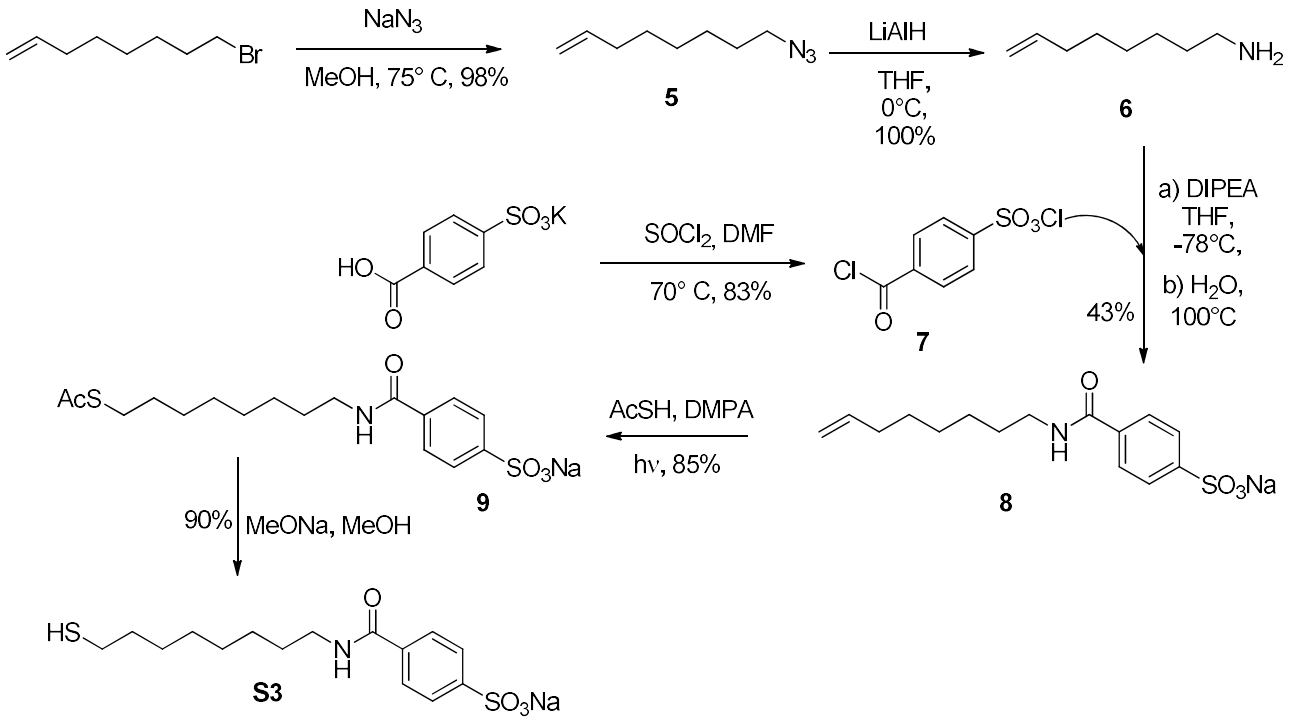
The acetylated sulfonate **4** (223 mg, 0.445 mmol, 1 equiv) was dissolved in 5 mL of dry MeOH and then sodium methoxide was added (72 mg, 1.334 mmol, 3 equiv); after stirring at for 3 hours under N_2 atmosphere, the reaction was quenched adding IR 120H$^+$ resin until pH neutralization, then the resin was filtered off, the solvent was evaporated, giving the deprotected Sodium 4-(8-mercaptooctanamido)benzenesulfonate **S2** (145 mg, yield 90%), that was freshly used for the nanoparticles' synthesis.

^{1}H-NMR (MeOD, 300): δ 1.36 (m, 8H), 1.56 (m br, 1H), 1.69 (m br, 2H), 2.41 (t, J=7.5 Hz, 2H), 2.88 (t, J=7.3 Hz, 2H), 7.38, 7.76 (m, 4H)

^{13}C-NMR (MeOD, 500): δ 25.29, 28.46, 28.58, 28.63, 28.97, 29.39, 33.22, 136.17, 137.76, 139.54, 141.07.

TOF ES$^-$ HRMS: [M$^-$] calcd. for $C_{14}H_{20}O_4NS_2$=330.083 Found =330.085

Thiol **S3** was prepared as reported in the following scheme:



8-azidooct-1-ene (5).

8-bromooct-1-ene (1 g, 5,23 mmol, 1 equiv) and NaN_3 (605 mg, 9,29 mmol, 1,77 equiv) were refluxed overnight in MeOH (7 mL). Then the solvent was removed under reduced pressure and the mixture was carefully extracted in DCM/H_2O. The organic layers were collected, dried over $NaSO_4$ anhydrous concentrated, obtaining 791 mg (98% yield) of a colorless liquid.

^{1}H-NMR ($CDCl_3$, 500 MHz): δ 1.41 (br s, 6H) 1.62 (m, 2H), 2.08 (m, 2H), 3.28 (t, J=7.0 Hz, 2H), 5.00 (m, 2H), 5.83 (m, 1H).

^{13}C-NMR ($CDCl_3$, 500 MHz): δ 26.57, 28.60, 28.72, 28,80, 33.64, 51.46, 114.38, 138.88

Oct-7-en-1-amine (6).

In a rbf, under N_2 atmosphere, 1M THF solution of $LiAlH_4$ (12 mL, 12 mmol, 2,3 equiv) was diluted with dry THF (30 mL) and the obtained solution was cooled at 0°C. Compound 5 (0,791 g, 5,13 mmol, 1 equiv) was dissolved in dry THF (20 mL) and this solution was slowly added (over 20

min) to the stirring $LiAlH_4$ solution. The ice bath was removed and the mixture was stirred under rt for 2 hours. After reaction completion, NaOH 1% solution (40 mL) was added and the mixture was extracted three times with Et_2O (40 mL). The organic layers were collected and dried over Na_2SO_4 anhydrous. No further purifications were required and due to highly volatility of compound 6, the solution was concentrated and used for the next step as a 0,3 M Et_2O solution (17 ml, 99%).

^{1}H-NMR ($CDCl_3$, 500 MHz): δ 1.35 (m, 6H) 1.42 (m, 2H), 2.09 (m, 2H) 2.66 (t, J=6.9 Hz, 2H), 4.97 (m, 2H), 5.85 (m, 1H).
^{13}C-NMR ($CDCl_3$, 500 MHz): δ 26.72, 28.94, 29.00, 33.74, 33.96, 42.25, 113.82, 139.24.
MS (ESI) *m/z*: 128.2 ($[M+H]^+$)

4-(chlorosulfonyl)benzoylchloride (7).

Benzensulphonic acid (1,5 g, 6,25 mmol, 1 equiv), thionyl chloride (15 mL, 76,88 mmol, 12 equiv) and dry DMF (0,1 mL) were stirred at 70°C for 4 hours. Then the mixture was concentrated under vacuum and Toluene was added. The solution was filtered and the solvent was evaporated under vacuum, obtaining 1,31 g (87%) of compound **7**.

^{1}H-NMR ($CDCl_3$, 500 MHz): δ 8.23 (m, 2H) 8.39 (m, 2H)
^{13}C-NMR ($CDCl_3$, 500 MHz): δ 127.75, 132.42, 138.54, 148.99, 167.03

Sodium 4-(oct-7-en-1-ylcarbamoyl)benzenesulfonate (8).

Compound **7** (165 mg, 0,69 mmol, 1 equiv) and DIPEA (300 µl, 1,72 mmol, 2,5 equiv) were dissolved in dry THF and the resulting solution was stirred for 10 minutes under N_2 atmosphere at -78°C. Then compound **6** (1,3 mL of Et_2O 0,3 M solution, 0,39 mmol, 0,58 equiv) was added in two portions over 30 minutes and the mixture was stirred for 2 hours at -78°C. Then the solution was brought to rt and filtered. The solvent was removed under reduced pressure and the obtained crude was dissolved in 40 mL of $CHCl_3$ and subsequently extracted with $NaHCO_3$ saturated solution, HCl 1M and finally brine. The organic layer was dried with Na_2SO_4, the solvent was evaporated under reduced pressure and the mixture was purified *via* flash chromatography (PE:EtOAc from 9:1 to 7:3). The obtained product was dissolved in H_2O and refluxed for 3 hours. Then water was evaporated under reduced pressure giving, without need of further purifications, the desired sulfonate **8** (57 mg, 43%).

^{1}H-NMR (MeOD, 200 MHz): δ 1.44 (br s, 6H, -CH2-) 1.67 (m, 2H), 2.11 (m, 2H), 3.40 (t, 2H), 5.06 (m, 2H), 5.83 (m, 1H) 7.91 (m, 4H).
MS (ESI) *m/z*: 333.2 ([M+Na]$^-$)

Sodium 4-((7-(acetylthio)heptyl)carbamoyl)benzenesulfonate (9).

Compound **8** (127 mg, 0.38 mmol, 1 equiv) was dissolved in 2 mL of MeOH and the solution was degased under N_2 bubbling for 30 min. Then thioacetic acid (115 µl, 1.63 mmol, 4 equiv) and 2,2-dimethoxy-2-phenylacetophenone (DMPA, 6 mg, 0.02 mmol, 0.05 equiv) were added and the mixture was transferred in UV cuvettes and stirred under UV irradiation (365 nm) for 2.5 h. The solvent was evaporated under vacuum and the crude was purified by flash column chromatography (DCM:MeOH from 9:1 to 8:2), affording the desired sulfonate compound **9** (125 mg, 85%).

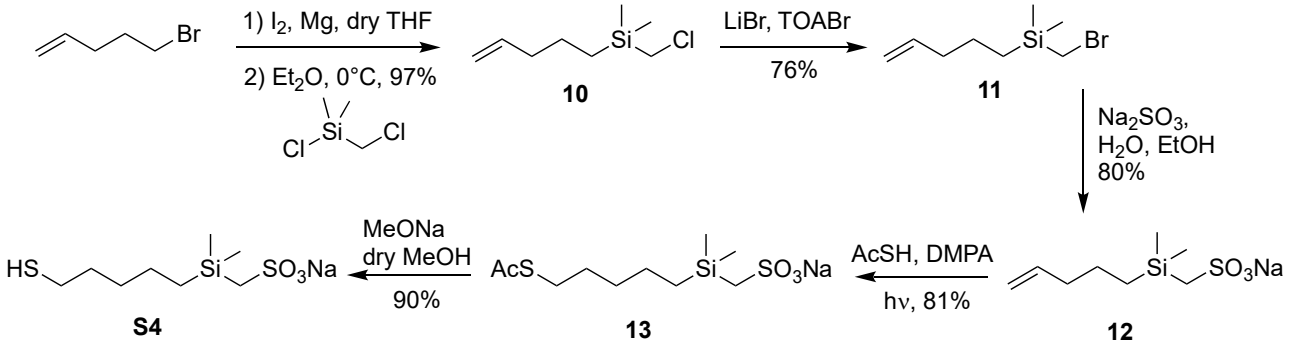
^{1}H-NMR (MeOD, 200 MHz): δ 1.37 (br s), 1.59 (m, 4H), 2.29 (s, 3H), 3.38 (m, 4H), 7.89 (m, 4H).
^{13}C-NMR (MeOD, 200 MHz): δ 26.8 29.5 29.7 30.0, 30.6, 31.9, 40.2, 128.1, 129.9, 138.8, 157.9, 167.5, 195.2.

Sodium 4-((7-mercaptoheptyl)carbamoyl)benzenesulfonate (S3).

The protected thiol **9** (45 mg, 0.12 mmol, 1 equiv) was dissolved in 2.5 mL of dry MeOH and then sodium methoxide was added (20 mg, 0.35 mmol, 3 equiv); after stirring at for 3 hours under N_2 atmosphere, the reaction was quenched adding IR 120H$^+$ resin until pH neutralization, then the resin was filtered off and the solvent was evaporated. Flash chromatography purification (8:2 DCM:MeOH) gave the deprotected **S3** (38 mg, yield 90%), that was freshly used for the nanoparticles' synthesis.

^{1}H-NMR (MeOD, 200 MHz): δ 1.37 (br s, 6H), 1.59 (m, 4H), 3.38 (m, 4H), 7.89 (m, 4H).
^{1}H-NMR (MeOD, 500 MHz): δ 23.55, 26.61, 27.93, 28.28, 28.29, 28.99, 33.80, 39.70, 125.73, 126.89, 136.02, 147.73, 167.94
TOF ES$^-$ HRMS: [M$^-$] calcd. for $C_{15}H_{22}NO_4S_2$=344.099. Found =344.106

Thiol **S4** was prepared as reported in the following scheme:



(Chloromethyl)dimethyl(pent-4-enyl)silane (10).

Mg turnings (693 mg, 29.713 mmol, 1.1 equiv) were vigorously stirred overnight in a two neck flask under N_2. Then 3 mL of dry THF and a catalytic amount of I_2 are added and the solution is heated in order to activate the Mg. Then 5-bromopent-1-ene (3.2 mL, 27.012 mmol, 1 equiv) and dry THF (57 mL) were added drop by drop. The solution was stirred under N_2 at 40°C for 6 hours. Then the solution was cooled to 0°C and slowly added *via* canula to a 0°C solution of chloro(chloromethyl)dimethylsilane (4.3 mL, 32.414 mmol, 1.2 equiv) in dry EtO_2. The obtained solution was stirred overnight at room temperature and then it was quenched by adding a saturated solution of NH_4Cl. Then the mixture was extracted with Et_2O for five times, the organic layers were collected, dried over Na_2SO_4 and then concentrated under reduced pressure. The obtained crude was purified was purified by flash column chromatography (100% PE), affording the desired compound **10** (4.7 g, 97%).

^{1}H NMR (500 MHz, $CDCl_3$) δ 6.03 – 5.75 (m, 1H), 5.21 – 4.98 (m, 2H), 2.89 (s, 2H), 2.25 – 2.13 (m, 2H), 1.59 – 1.49 (m, 2H), 0.81 – 0.72 (m, 2H), 0.22 (s, 6H).

^{13}C NMR (126 MHz, $CDCl_3$) δ 138.60, 114.74, 37.44, 30.34, 23.03, 13.23, -4.62.

^{29}Si NMR (99 MHz, $CDCl_3$) δ 3.71.

(Bromomethyl)dimethyl(pent-4-enyl)silane (11).

In a Schlenk tube compound **10** (2.5 g, 14.143 mmol, 1 equiv), LiBr (previously dried at 150°C under N_2, 3.69 g, 42.430 mmol, 3 equiv) and TOABr (1.50 g, 2.829 mmol, 0.2 equiv) were added and the mixture was stirred for 24 h at 60°C. The obtained crude was purified by flash column chromatography (100% PE), affording the desired compound **11** (2.37 g, 76%).

^{1}H NMR (500 MHz, $CDCl_3$) δ 6.02 – 5.79 (m, 1H), 5.21 – 4.98 (m, 2H), 2.58 (s, 2H), 2.24 – 2.11 (m, 2H), 1.59 – 1.49 (m, 2H), 0.82 – 0.72 (m, 2H), 0.23 (s, 6H).
^{13}C NMR (126 MHz, $CDCl_3$) δ 138.61, 114.74, 37.43, 23.07, 17.09, 13.69, -4.04.
^{29}Si NMR (99 MHz, $CDCl_3$) δ 3.69.

Sodium (dimethyl(pent-4-enyl)silyl)methanesulfonate (12).

In a Schlenk tube compound **11** (400 mg, 1.808 mmol, 1 equiv) and Na_2SO_3 (456 mg, 3.616 mmol, 2 equiv) were dissolved in 13 mL of H_2O:EtOH 1:1 and the reaction was stirred for 24 h at 60°C. The obtained crude was purified by reverse phase chromatography (C18 resin, from 100% H_2O to 100% MeOH), affording the desired compound **12** (352 mg, 80%).

^{1}H NMR (500 MHz, MeOD) δ 5.96 – 5.70 (m, 1H), 5.09 – 4.90 (m, 2H), 2.58 (s, 2H), 2.16 – 2.01 (m, 2H), 1.54 – 1.37 (m, 2H), 0.81 – 0.63 (m, 2H), 0.18 (s, 6H).
^{13}C NMR (126 MHz, MeOD) δ 138.5, 113.6, 41.9, 37.3, 23.0, 14.4, -4.2.
^{29}Si NMR (99 MHz, $CDCl_3$) δ 0.85.
MS (ESI) *m/z*: 221.1 ($[M]^-$)

Sodium ((5-(acetylthio)pentyl)dimethylsilyl)methanesulfonate (13).

Sulfonate **12** (221.3 mg, 1.471 mmol, 1 equiv) was dissolved in 3 mL of MeOH and the solution was degased by N_2 bubbling for 30 min. Then thioacetic acid (415 µl, 5.890 mmol, 4 equiv) and 2,2-dimethoxy-2-phenylacetophenone (DMPA, 39 mg, 0.147 mmol, 0.1 equiv) were added and the mixture was stirred under UV irradiation (365 nm) for 2.5 h. The solvent was evaporated under vacuum and the crude was purified by flash column chromatography (DCM:MeOH from 9:1 to 8:2), affording the desired sulfonate compound **13** (381 mg, 81%).

^{1}H NMR (500 MHz, MeOD) δ 2.88 (t, J = 7.3 Hz, 2H), 2.59 (s, 2H), 2.32 (s, 2H), 1.64 – 1.53 (m, 2H), 1.46 – 1.35 (m, 2H), 0.78 – 0.65 (m, 2H), 0.18 (s, 6H).
^{13}C NMR (126 MHz, MeOD) δ 196.23, 41.87, 32.24, 29.11, 28.99, 28.41, 22.85, 14.67, -4.16.
^{29}Si NMR (99 MHz, MeOD) δ -0.84.
MS (ESI) *m/z*: 297.0 ($[M]^-$)

Sodium ((5-mercaptopentyl)dimethylsilyl)methanesulfonate (S4).

The acetylated sulfonate **13** (30 mg, 0.0987 mmol, 1 equiv) was dissolved in 1 mL of dry MeOH and then sodium methoxide was added (16 mg, 0.296 mmol, 3 equiv). After stirring for 2.5 hours under N_2 atmosphere, the reaction was quenched adding IR 120H$^+$ resin until pH neutralization,

then the resin was filtered off, the solvent was evaporated, giving the deprotected Sodium 11-mercaptoundecane-1-sulfonate **S4** (25 mg, yield 90%), that was freshly used for the nanoparticles' synthesis.

^{1}H NMR (500 MHz, MeOD) δ 2.57 (s, 2H), 2.52 – 2.45 (m, 2H), 1.66 – 1.56 (m, 2H), 1.49 – 1.32 (m, 4H), 0.76 – 0.66 (m, 2H), 0.18 (s, 6H).

^{13}C NMR (126 MHz, MeOD) δ 41.90, 37.31, 33.56, 24.70, 23.30, 15.06, -4.16.

^{29}Si NMR (99 MHz, MeOD) δ -0.87.

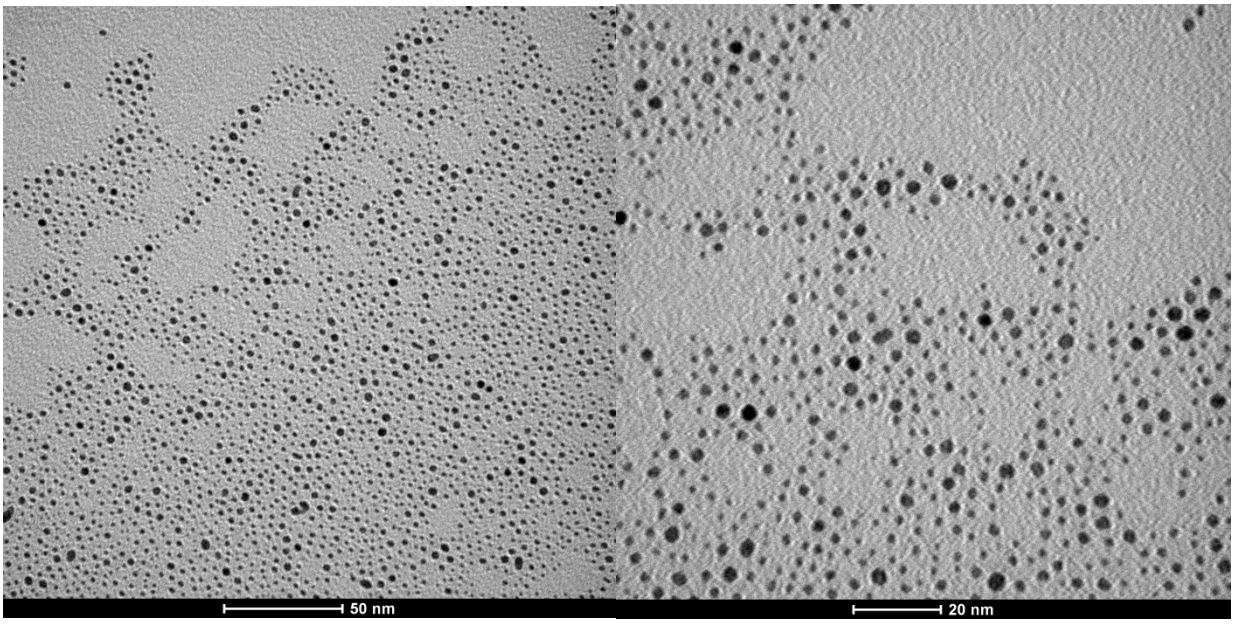
TOF ES^{-} HRMS: [M^{-}] calcd. for $C_8H_{19}O_3S_2Si$=255.055. Found = 255.049.

3. Synthesis and characterization of monolayer protected gold nanoparticles

Monolayer protected gold nanoparticles (**S1**, **S2**, **S3**, **S4-AuNPs**) were prepared following a previously reported two-step procedure.[2] A solution of $HAuCl_4 \cdot 3H_2O$ (50 mg, 0.127 mmol, 1 equiv) in water (2 mL) was extracted with a solution of tetraoctylammonium bromide (0.175 g, 0.318 mmol, 2.5 equiv) in N_2 purged toluene (125 mL). To the resulting reddish-orange organic solution dioctylamine (0.613 g, 2.539 mmol, 20 equiv) was added (the amount of dioctylamine was calculated in order to obtain 2 nm nanoparticles). The mixture is vigorously stirred under N_2 for 1,5 hours. During this period of time the color of the mixture fades. Then the solution is cooled at 0°C and a $NaBH_4$ solution (48.0 mg, 1.269 mmol, 10 equiv) in H_2O (1 mL) is then rapidly added. The color of the solution turns rapidly to black and after 1.5 hours of stirring at 0°C, the aqueous layer is removed. To the obtained nanoparticle solution, the desired thiol **S1-4** (0.254 mmol, 2 equiv) dissolved in 3 mL of MeOH was rapidly added. The reaction mixture was stirred for 3 hours at 0°C. All the formed AuNPs were unsoluble in toluene, hence the mixtures were centrifuged and the collected AuNPs were washed under sonication 7 times, with EtOAc and MeOH. The resulting NPs were finally purified by gel permeation chromatography with Sephadex G-25.

3.1. Characterization of S1-, S2-, S3-, S4-AuNPs

TEM analysis of the different samples of small nanoparticles (Figures S1, S2, S3, S4) yields an average diameter for **S1-AuNPs** of 1.8 ± 0.2nm, for **S2-, S3-, S4-AuNPs** of 1.6 ± 0.4 nm. This data, together with the loss of organic weigh obtained by TGA analysis (Figures S5,S6, S7, S8), indicate that the formula for AuNP is $\mathbf{Au_{127}SR_{39}}$ for **S1-AuNPs**, $\mathbf{Au_{180}SR_{54}}$ for **S2-AuNPs**, $\mathbf{Au_{127}SR_{55}}$ for **S3-AuNPs**, $\mathbf{Au_{127}SR_{44}}$ for **S4-AuNPs**. The gold core of AuNP was approximated as a sphere, and the weight loss considered is related to the thiol minus NaHSO3 that stays as inorganic residue. NMR analysis (Figure S9, S10, S11, S12) indicates monolayer formation (broadening of all signals and missing of the SCH_2CH_2 protons' signals). UV-vis spectra (Figure S13, S14, S15, S16) were recorded.



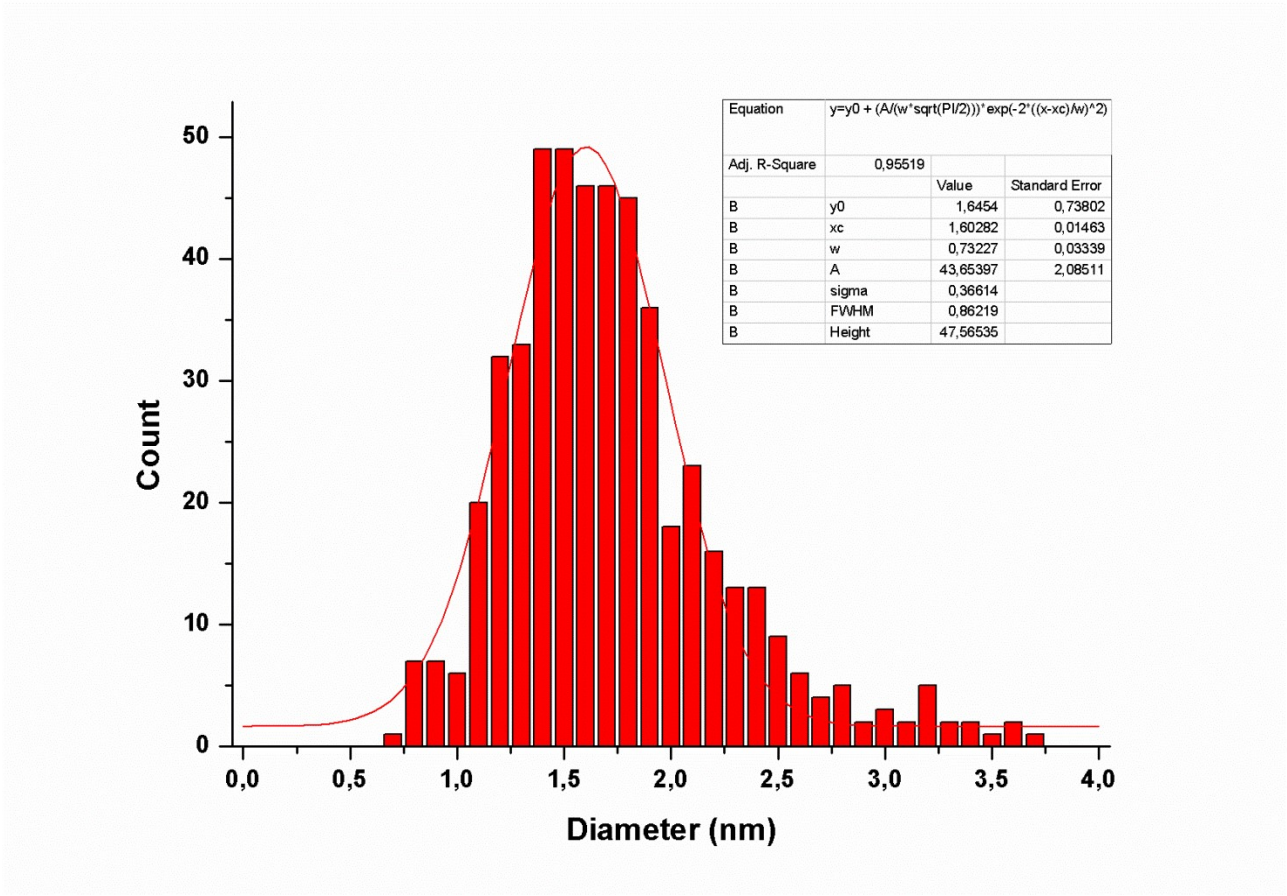
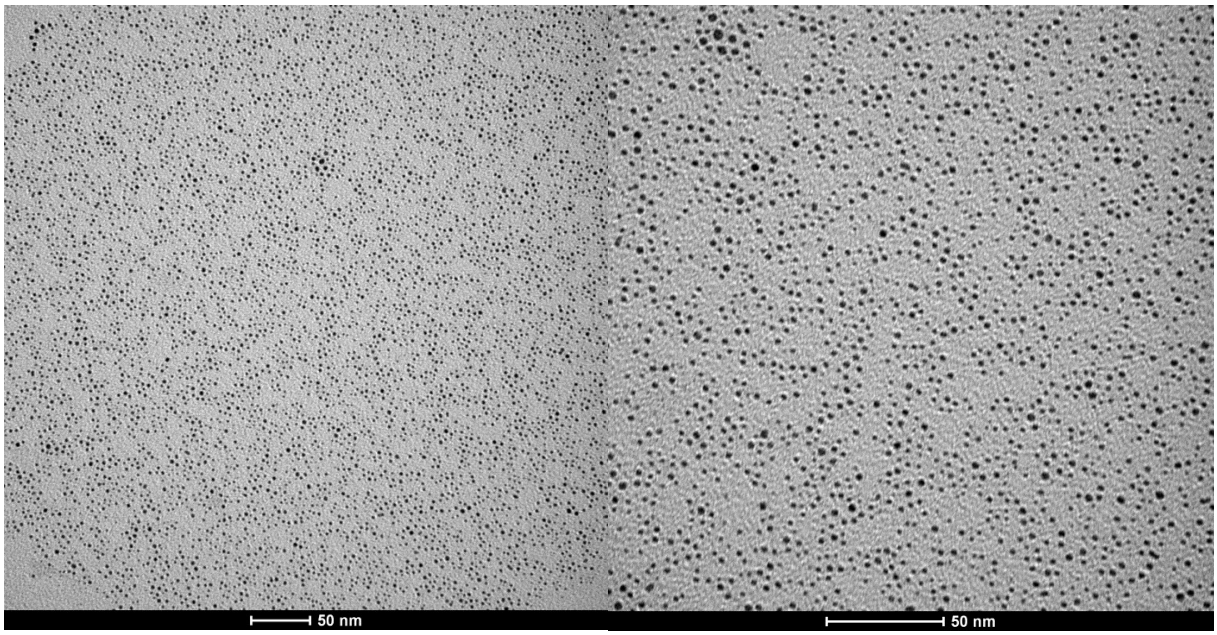


Figure S1. Sample TEM images **S1-AuNP** and size distribution: average diameter = 1.6 nm (σ= 0.4 nm).



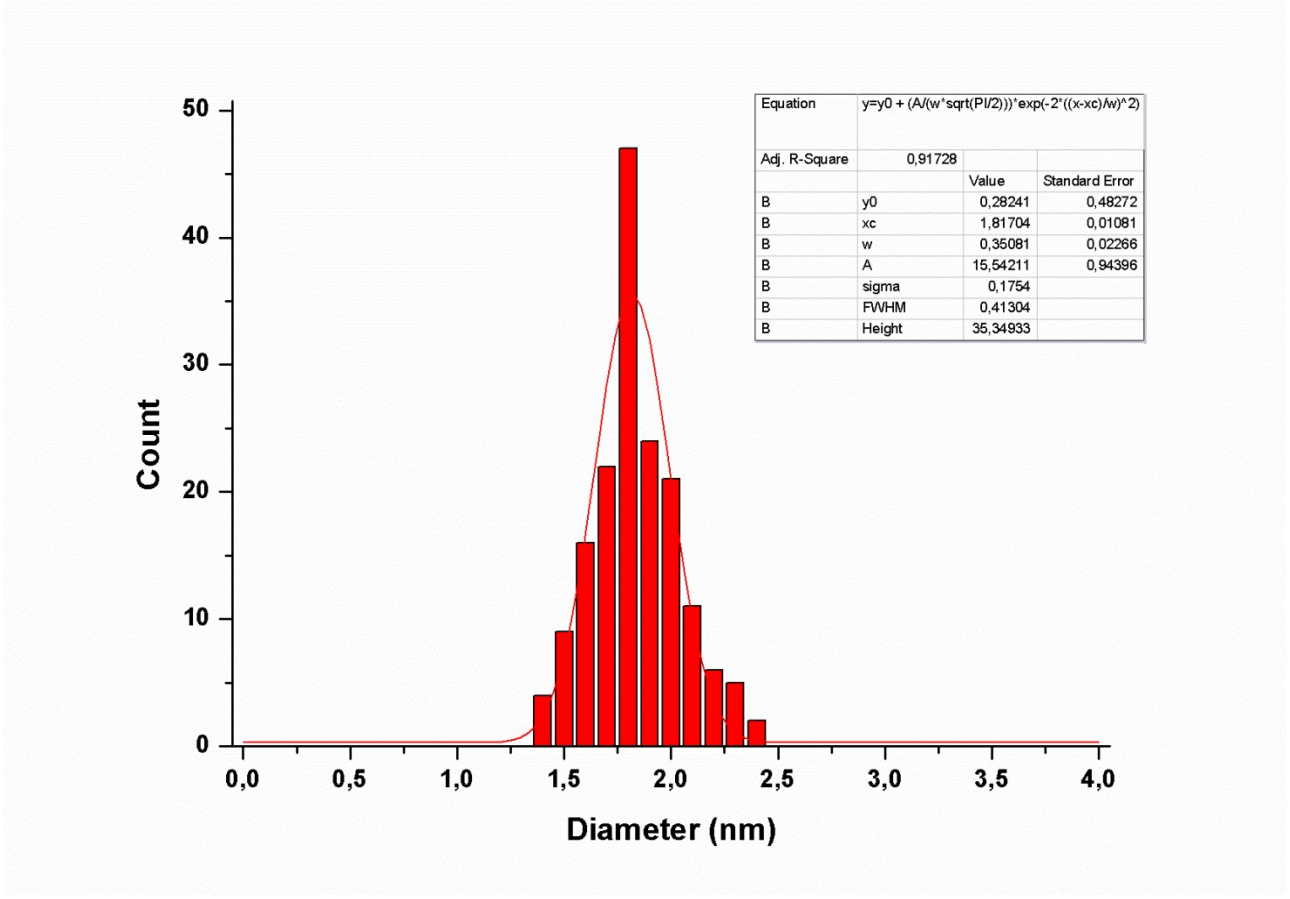
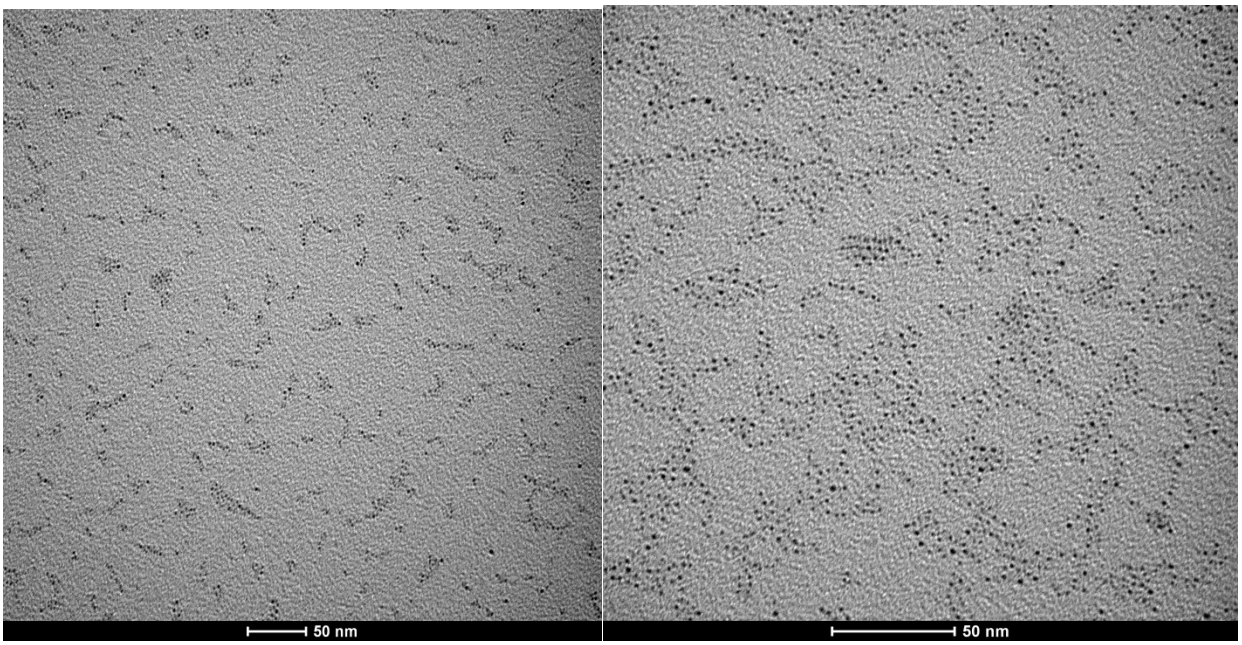


Figure S2. Sample TEM images **S2-AuNP** and size distribution: average diameter = 1.8 nm (σ= 0.2 nm).



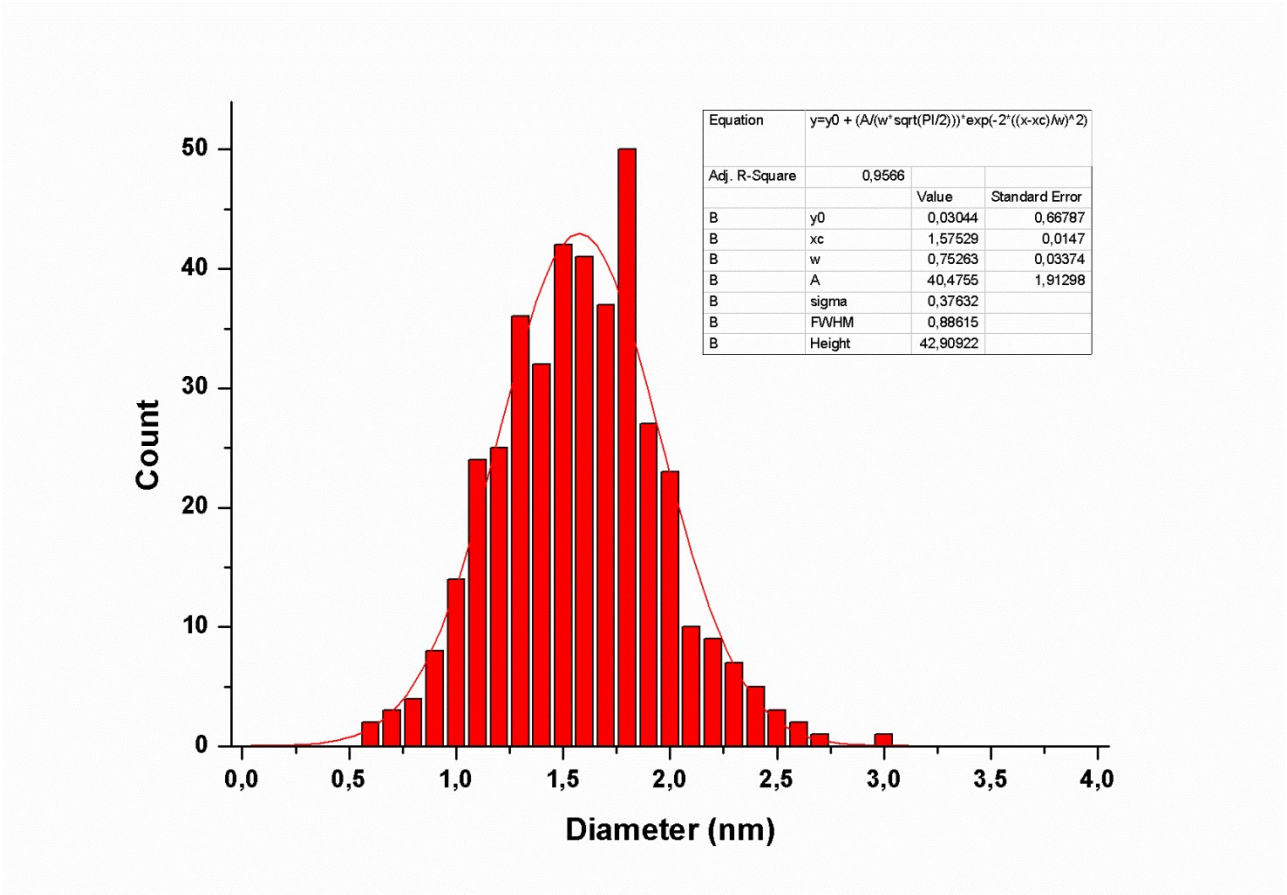
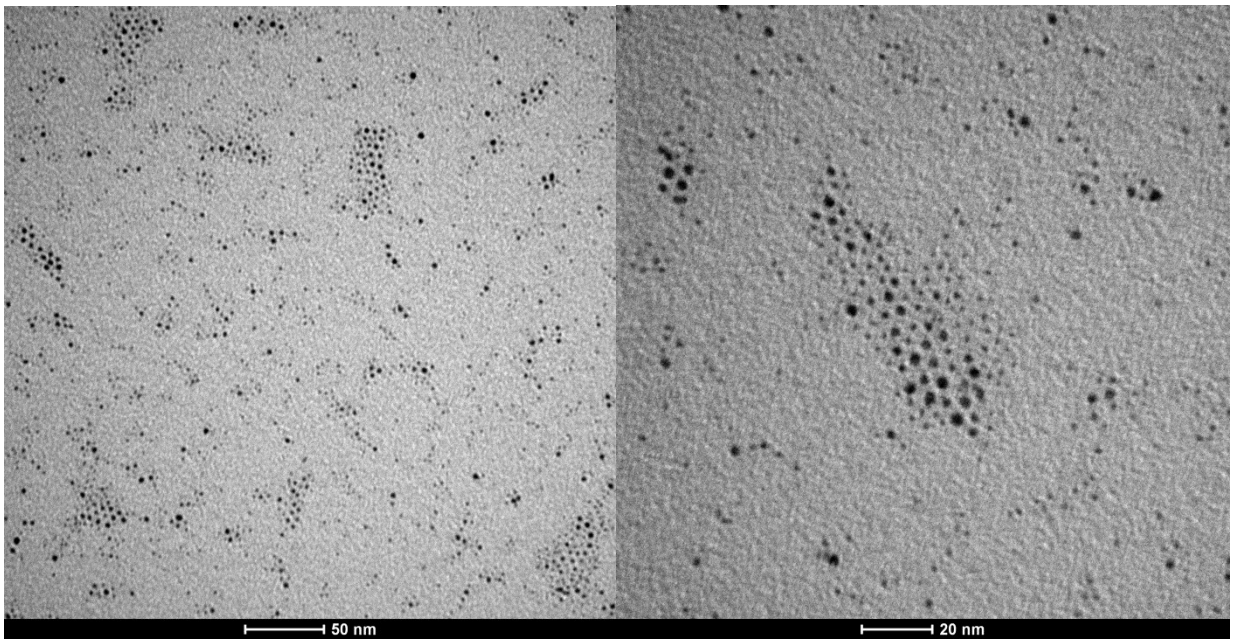


Figure S3. Sample TEM images of **S3-AuNP** and size distribution: average diameter = 1.6 nm (σ= 0.4 nm).



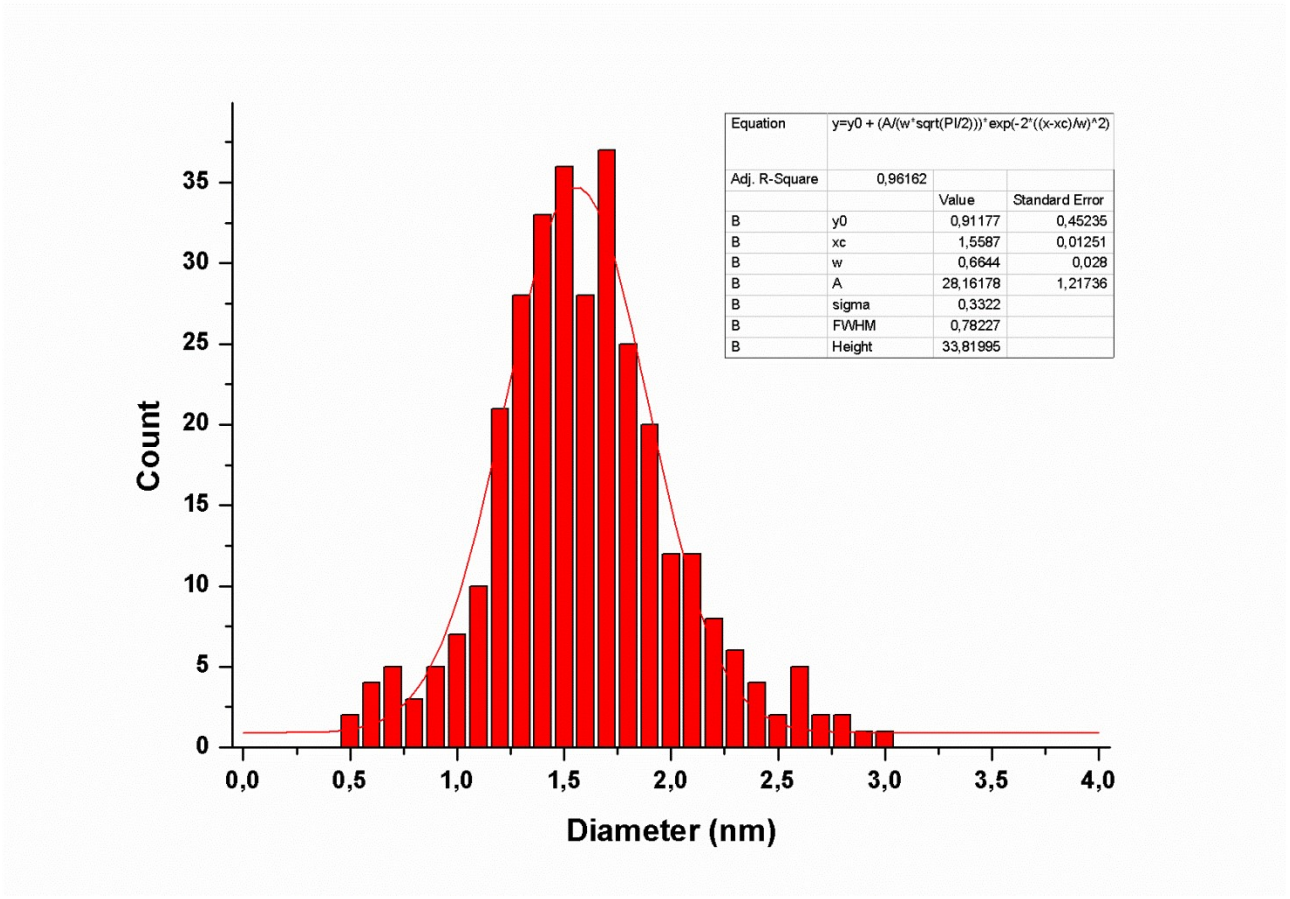


Figure S4. Sample TEM images **S4-AuNP** and size distribution: average diameter = 1.6 nm (σ= 0.3nm).

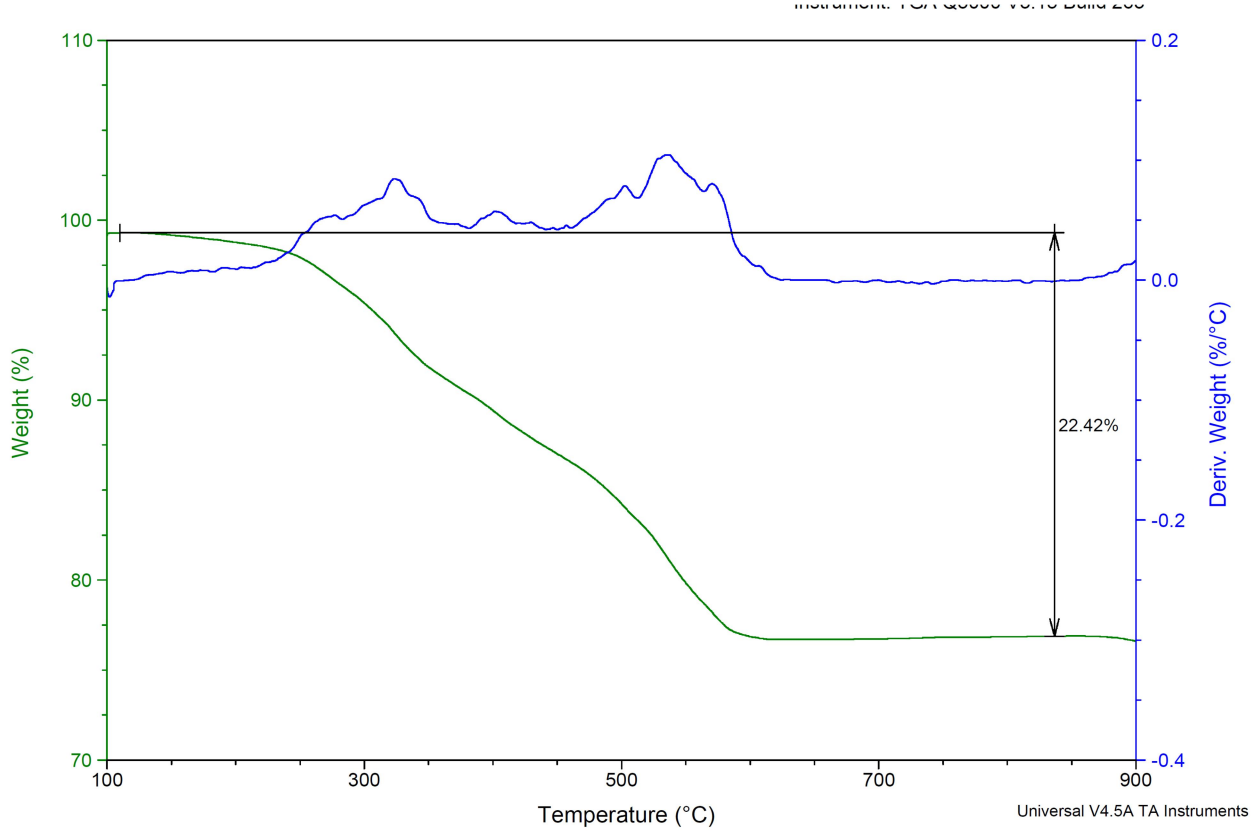


Figure S5. TGA analysis of **S1-AuNPs** sample, under air atmosphere.

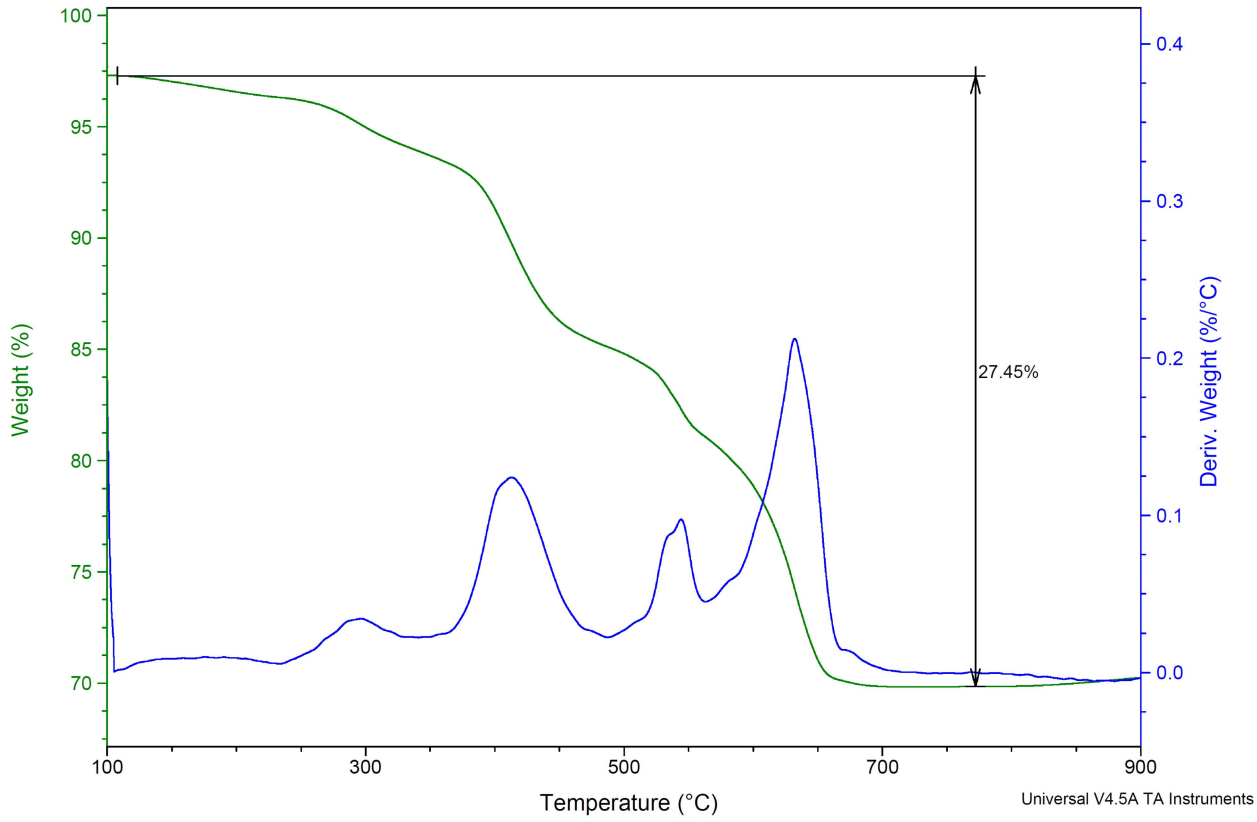


Figure S6. TGA analysis of **S2-AuNPs** sample, under air atmosphere.

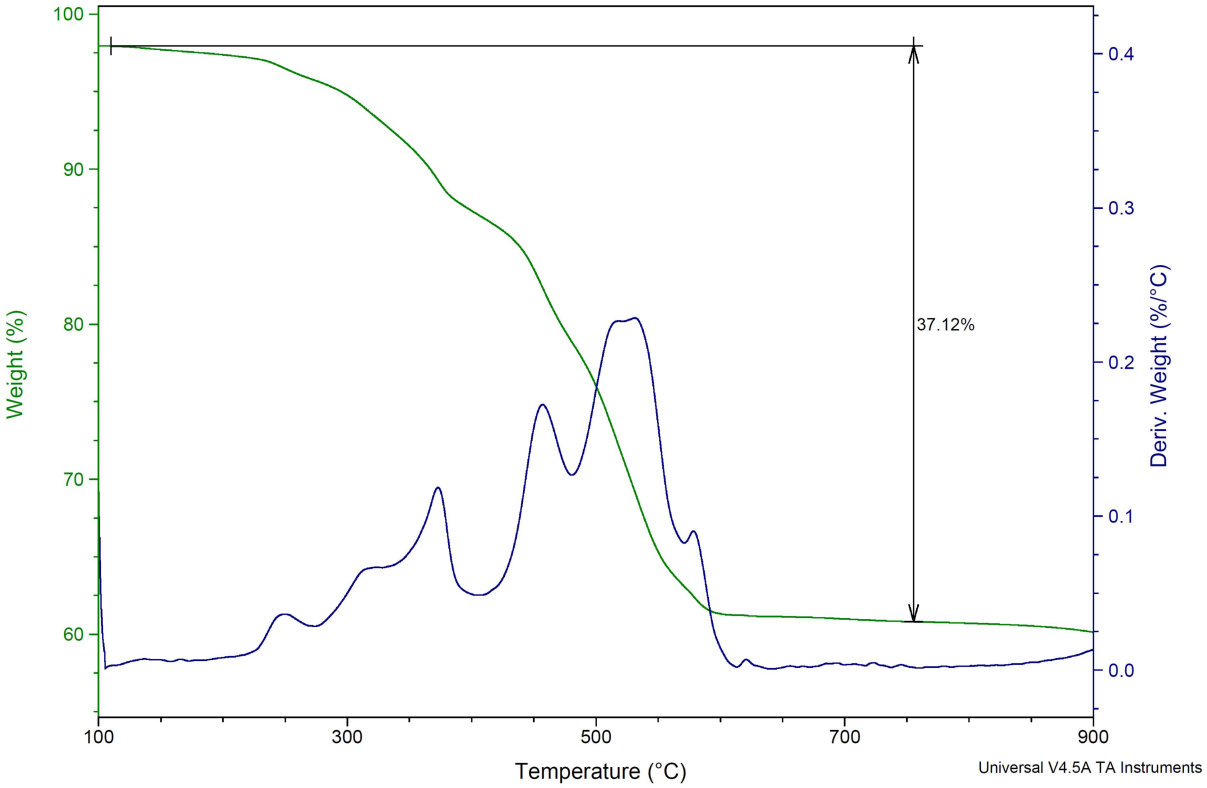


Figure S7. TGA analysis of **S3-AuNPs** sample, under air atmosphere.

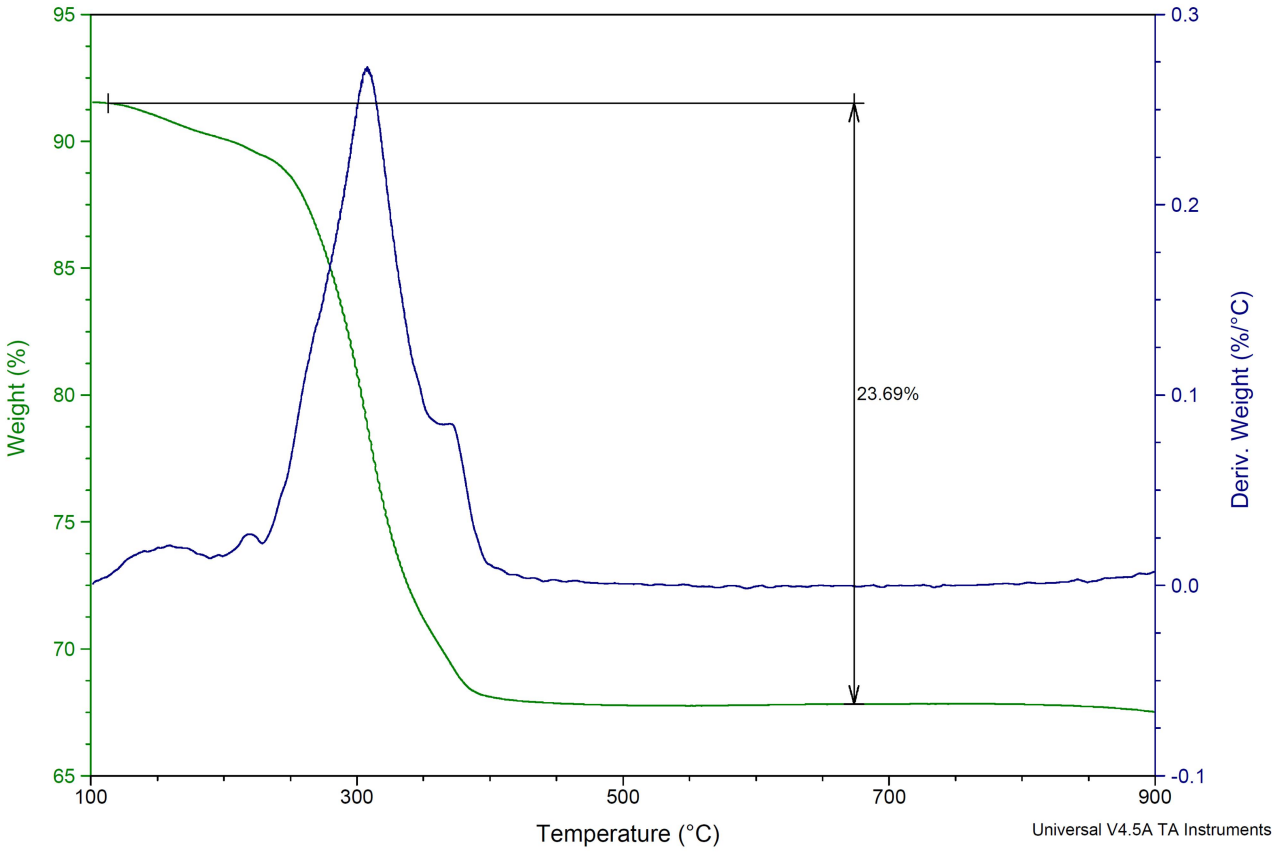


Figure S8. TGA analysis of **S4-AuNPs** sample, under air atmosphere.

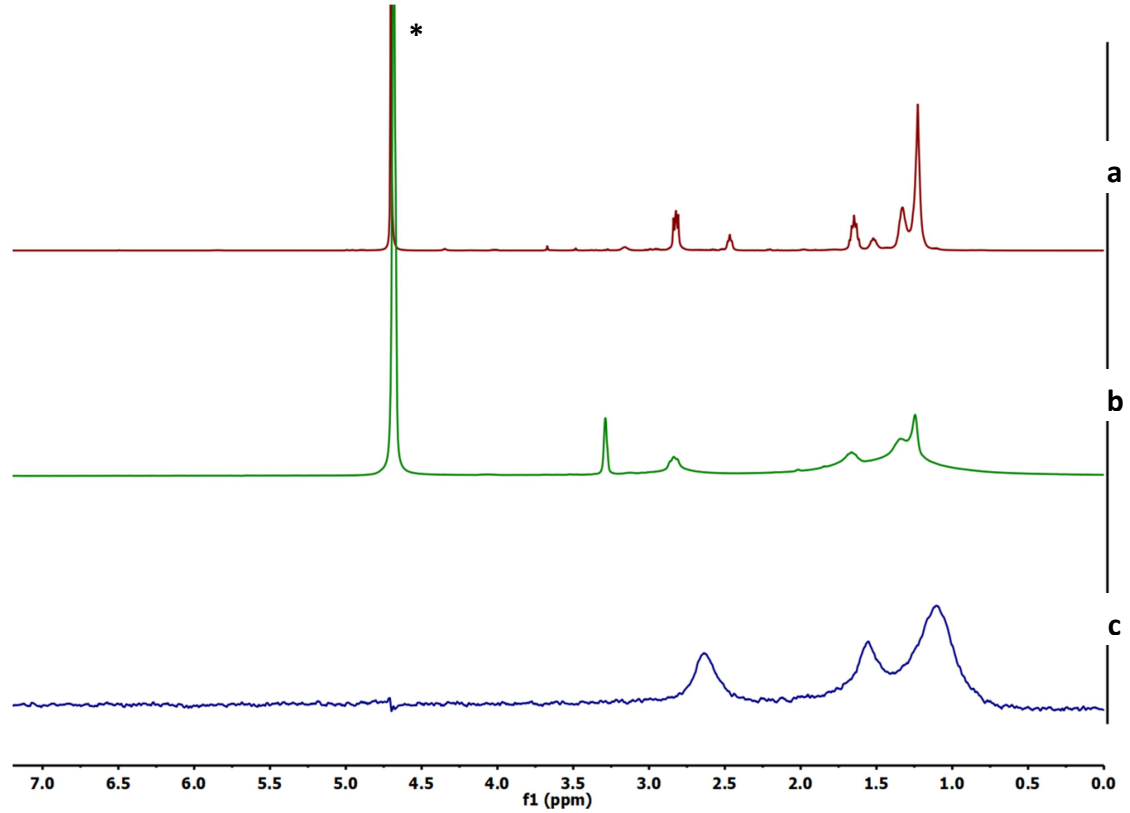


Figure S9. ^{1}H-NMR (500 MHz) spectrum in D_2O of: a) deprotected thiol **S1**; b) **S1-AuNPs**; c) diffusion filtered spectrum of **S1-AuNPs**. (* indicates the residual solvents).

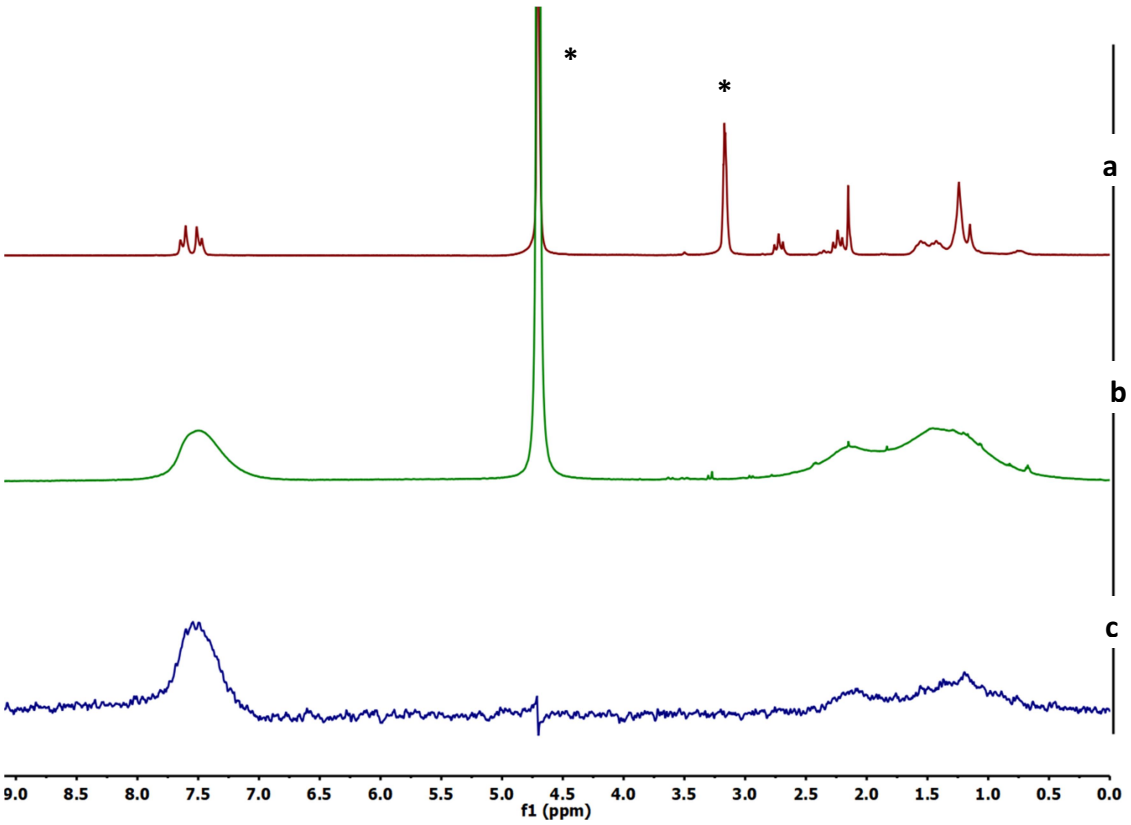


Figure S10. ^{1}H-NMR (500 MHz) spectrum of: a) deprotected thiol **S2** in MeOD; b) **S2-AuNPs** in D_2O; c) diffusion filtered spectrum of **S2-AuNPs** in D_2O. (* indicates the residual solvents).

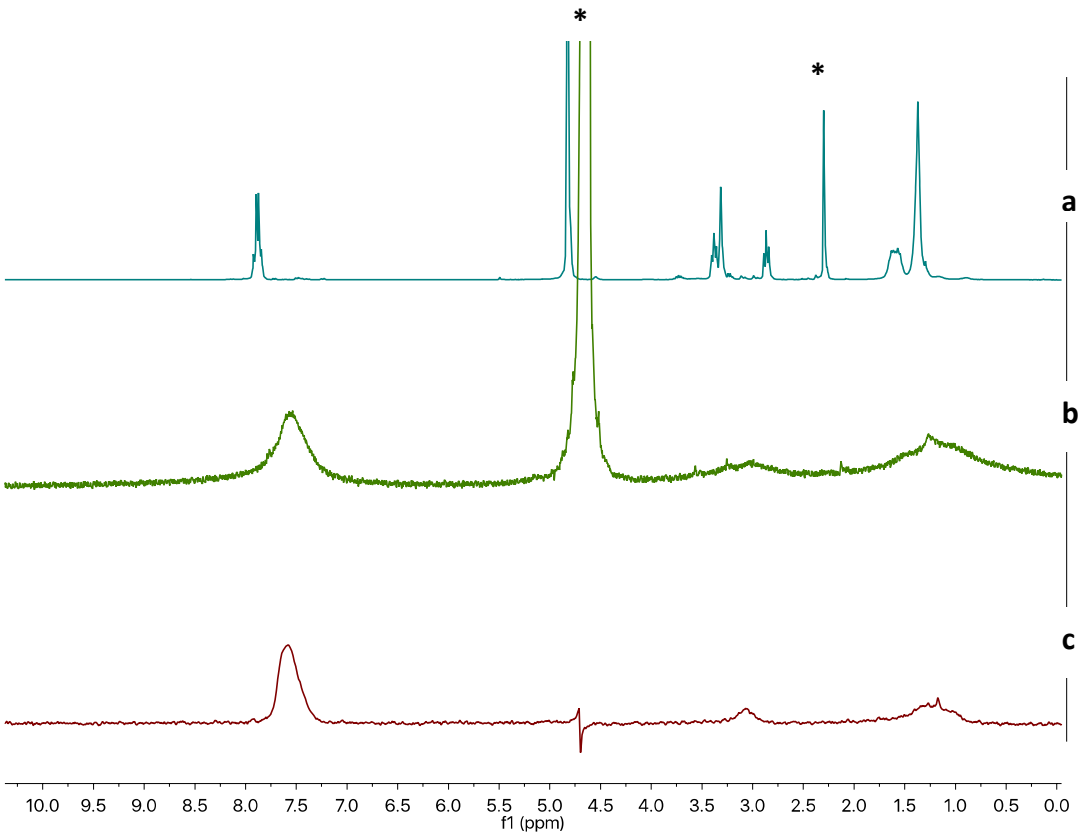


Figure S11. [1]H-NMR (500 MHz) spectrum of: a) protected thiol **9**; b) Diffusion filter of **S3-AuNPs** and c) [1]HNMR of the **S3-AuNPs** (* indicates the residual solvents).

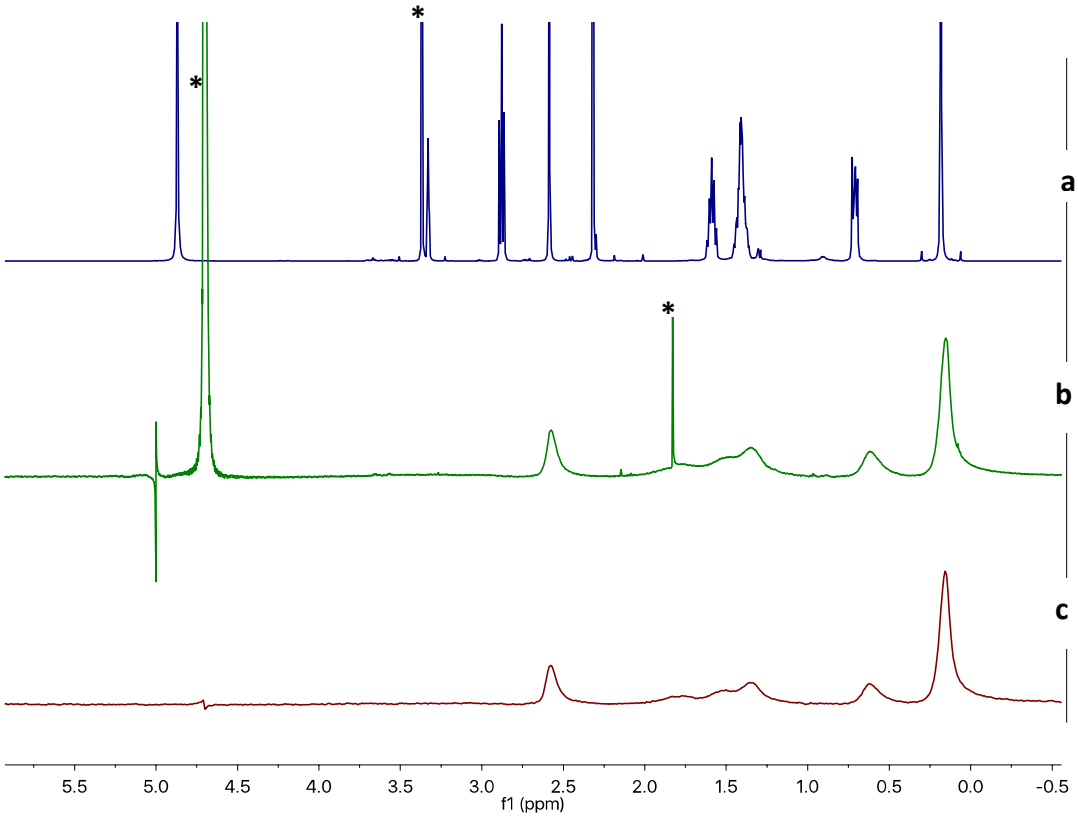


Figure S12. [1]H-NMR (500 MHz) spectra of: a) Protected thiol (MeOD) **13**; b) [1]H-NMR of the **S4-AuNPs** and c) Diffusion filter of **S4-AuNPs** (D_2O) (* indicates the residual solvents).

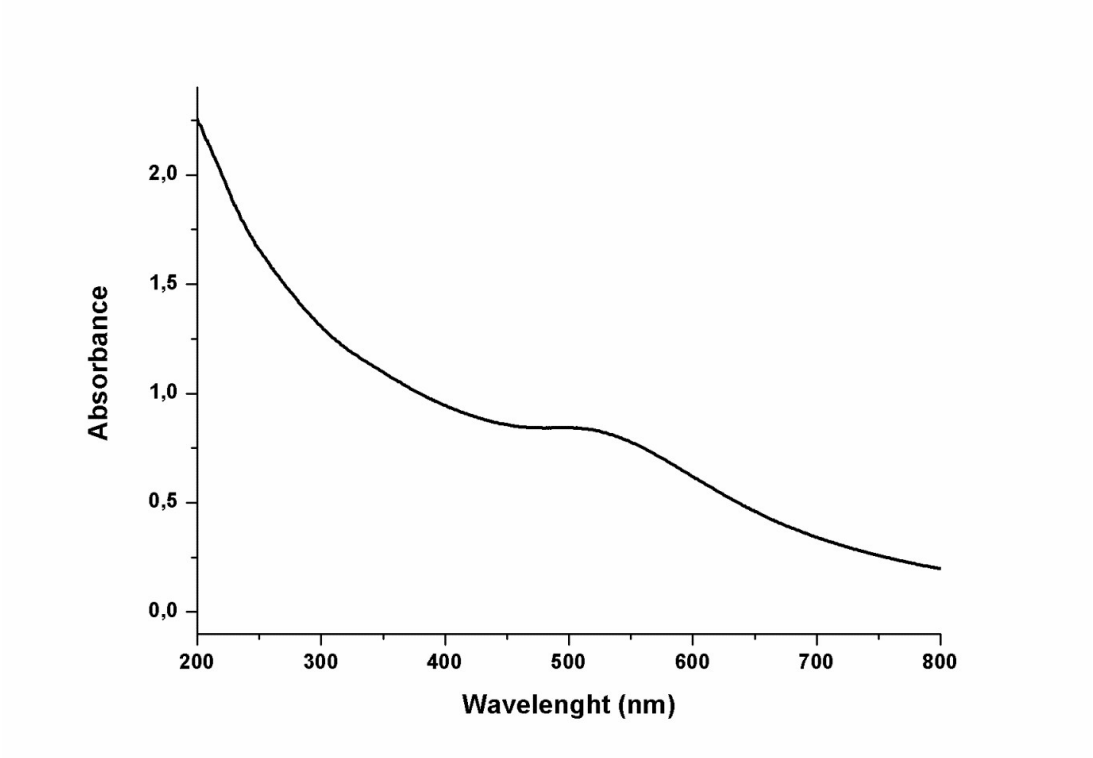


Figure S13. UV-Vis spectrum of **S1-AuNPs** (0.1 mg/mL) at 25°C in water.

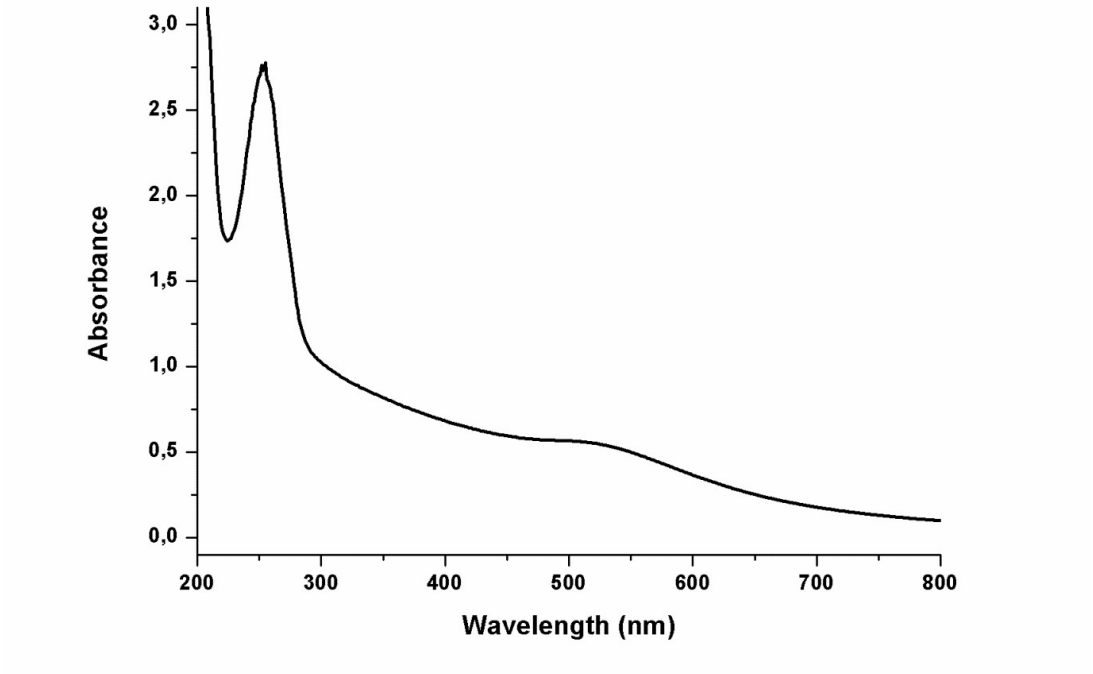


Figure S14. UV-Vis spectrum of **S2-AuNPs** (0.1 mg/mL) at 25°C in water. Absorbance peak of thiol aromatic moiety at 270nm.

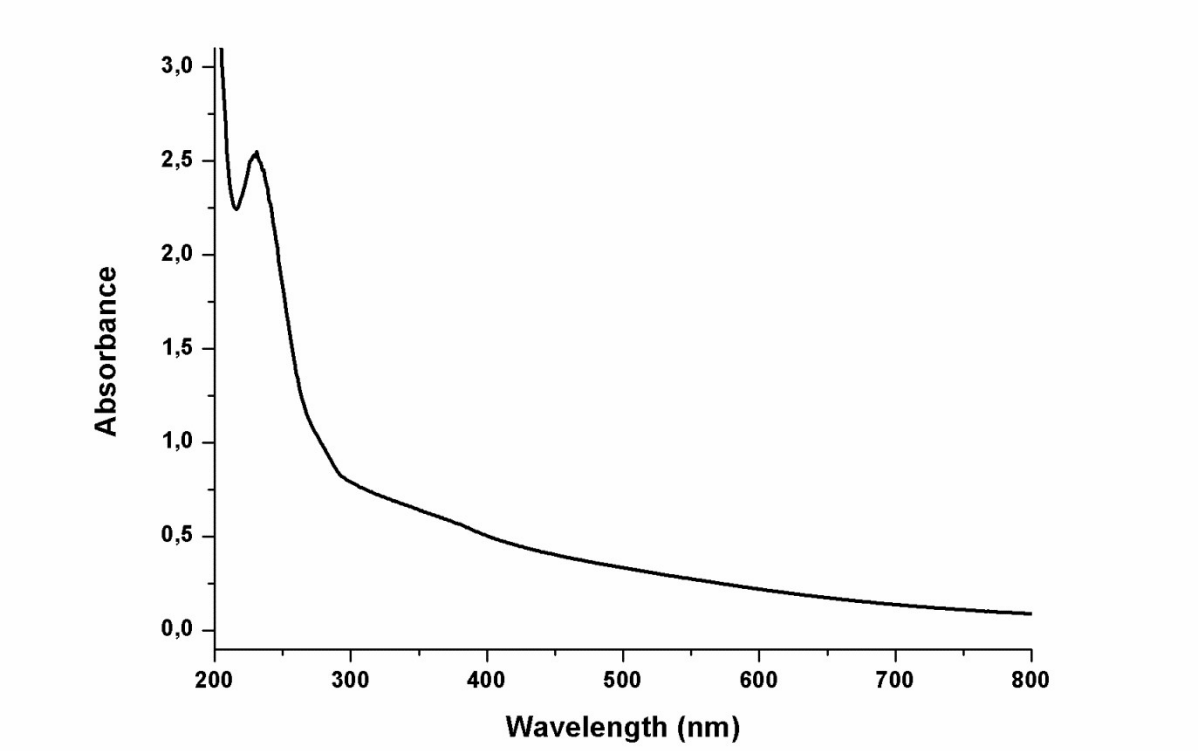


Figure S15. UV-Vis spectrum of **S3-AuNPs** (0.1 mg/mL) at 25°C in water. Absorbance peak of thiol aromatic moiety at 270nm.

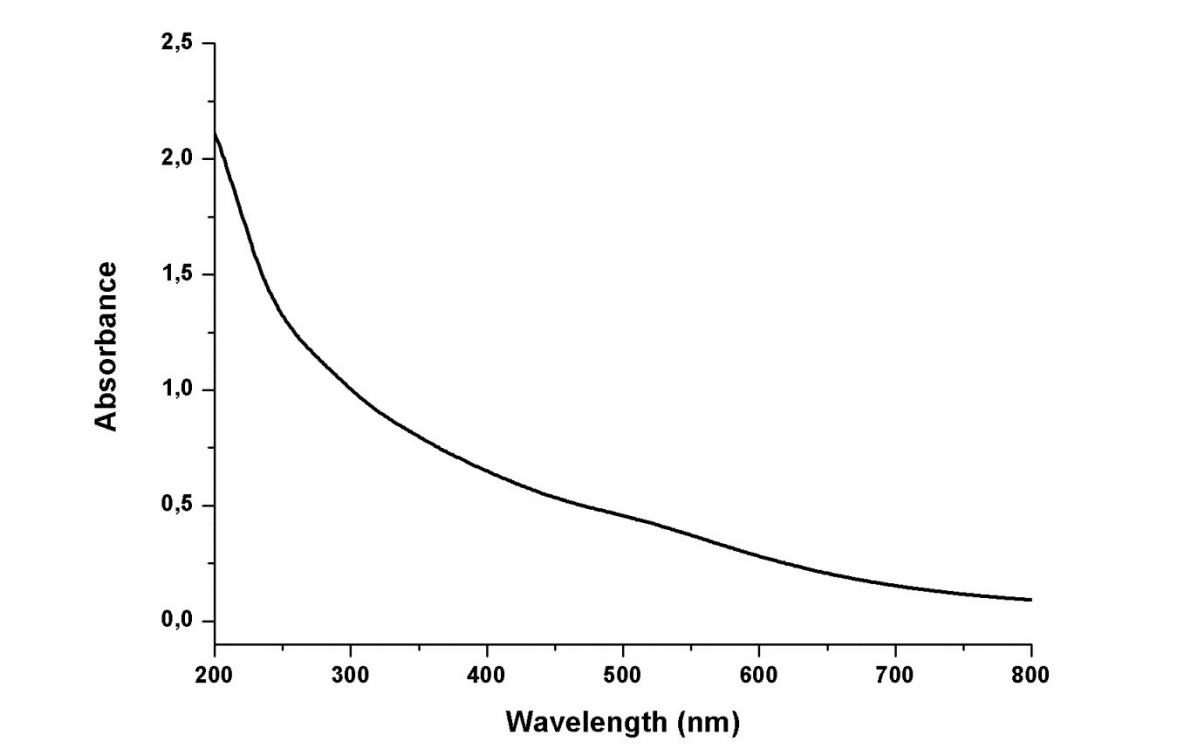


Figure S16. UV-Vis spectrum of **S4-AuNPs** (0.1 mg/mL) at 25°C in water.

4. Fluorescence experiments

4.1 Direct titrations

The fluorescence titrations were performed using a Perkin Elmer LS50B instrument. Intensities generated upon subsequent additions of a 3mM (2µl) solution of the analytes to a 1.4µM solution of the AuNP (100µM in thiol) in buffered H_2O (HEPES 2 mM, pH=7) were recorded after the signal had stabilized (2-3 min). Each point is the average of three measurements. The fluorescence intensities were plotted vs the concentration of analyte added. The titration were fitted to the following 1:1 binding model using DynaFit for Windows.

```
 [task]
   data = equilibria
   task = fit
[mechanism]
   P + L <==> P.L     :    Kd    dissoc
[constants]
   Kd = 0.00000 ?
[concentrations]
   P = 0.000000 ?
[data]
   variable L
   directory NAME
   sheet NAME.csv
   column 2 | response L = 2e4 ?     | label I, au
[output]
   directory NAME
[end]
```

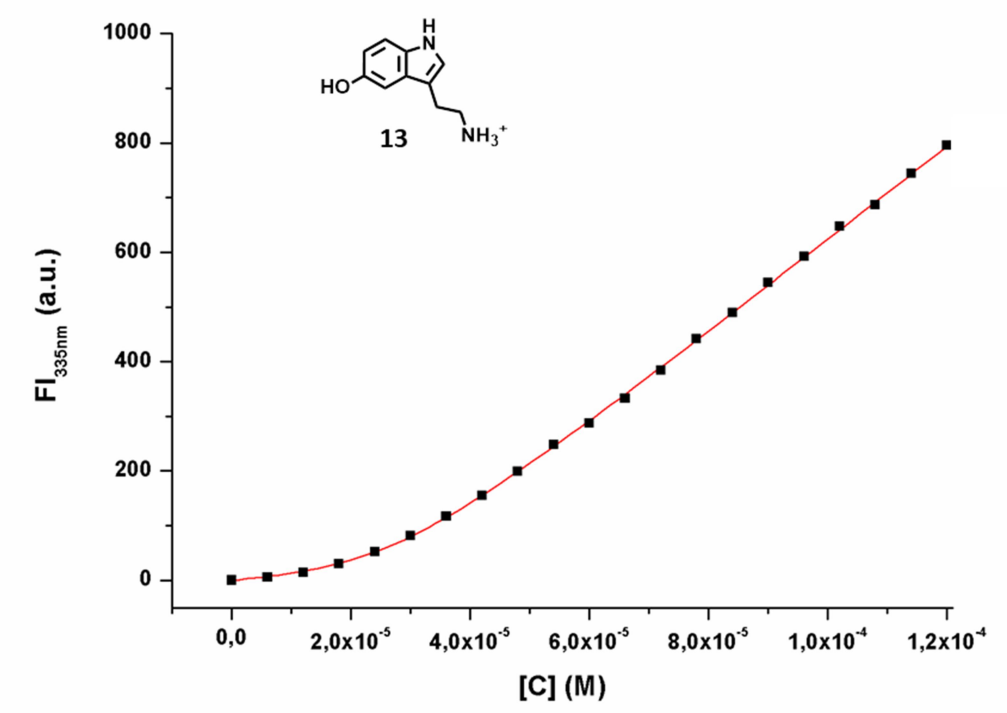


Figure S17. Plot of the fluorescence intensities vs the concentration of serotonine (**13**) added. In red the fitting curve obtained from DynaFit. Conditions **S1-AuNP**s 0.1 mM , HEPES 2mM.

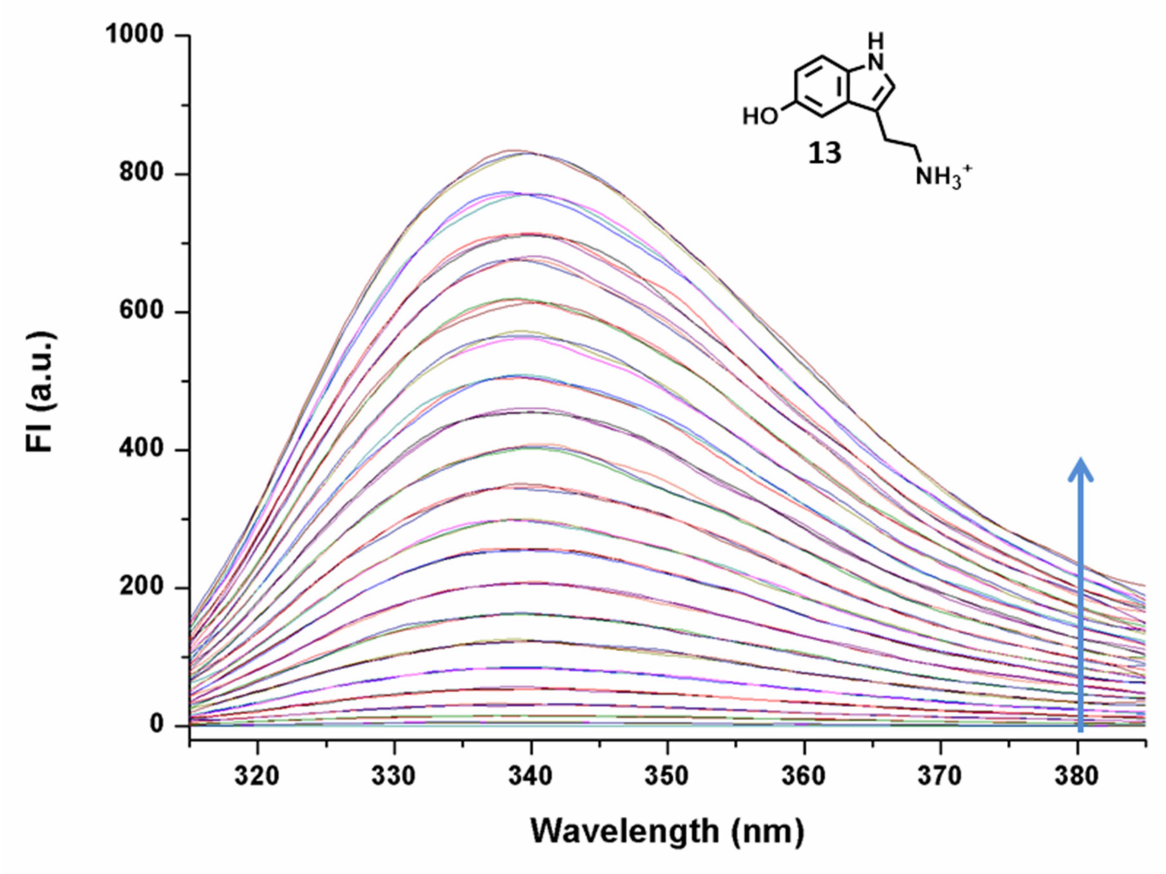


Figure S18. Fluorescence spectra obtained from the titration of **S1-AuNPs** (0.1mM) with serotonine (solution 3mM, 2µL additions) at 25°C in water. Every spectrum is recorded three times.

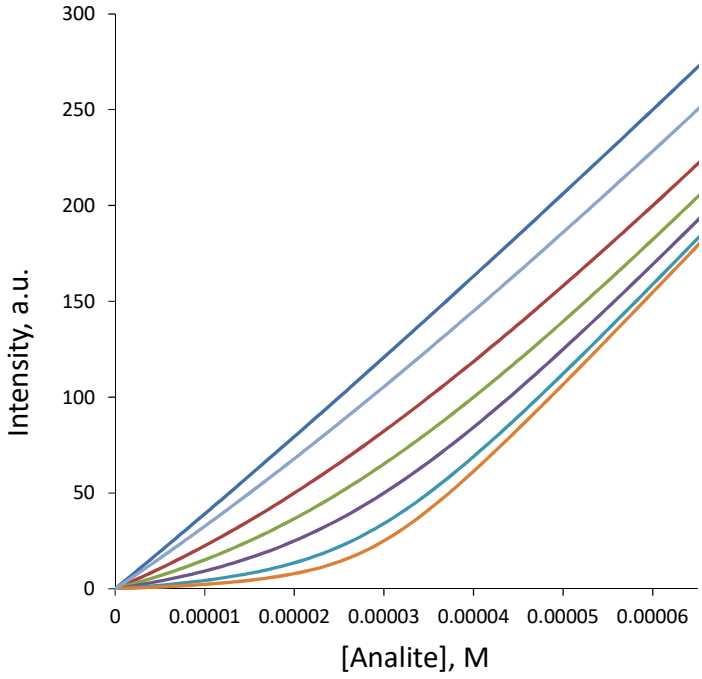


Figure S19. Simulated titration experiments for different binding constants ([binding sites] = 3×10^{-5} M). From the bottom: K = 1×10^{6} M^{-1}, 5×10^{5} M^{-1}, 2×10^{5} M^{-1}, 1×10^{5} M^{-1}, 5×10^{4} M^{-1}, 2×10^{4} M^{-1}, 1×10^{4} M^{-1}. Inspection of the plot clearly indicates that binding constants smaller than 5×10^{4} M^{-1} (red) are difficult to be measured in these experimental conditions.

4.2 Displacement titrations

The fluorescence displacement titrations were performed using the same procedure reported in the previous section **4.1**. Conditions: additions of a 3mM (2µl) solution of the analytes to a 1.4 µM solution of the AuNP (100 µM in thiol) in buffered H_2O (HEPES 2 mM, pH=7) containing dopamine 72 µM. Fitting model as follows.

```
 [task]
   data = equilibria
   task = fit
[mechanism]
   P + L <==> P.L     :    Kd    dissoc
   P + D <==> P.D     :    Kc    dissoc
[constants]
   Kd = 0.000000
   Kc = 0.000000 ?
[concentrations]
   P = 0.000040 ?
   L = 0.000029
[data]
   variable D
   directory NAME
   sheet NAME.csv
   column 2 | response L = 2e7 ?   | label I, au
[output]
   directory NAME/fit_tit
[end]
```

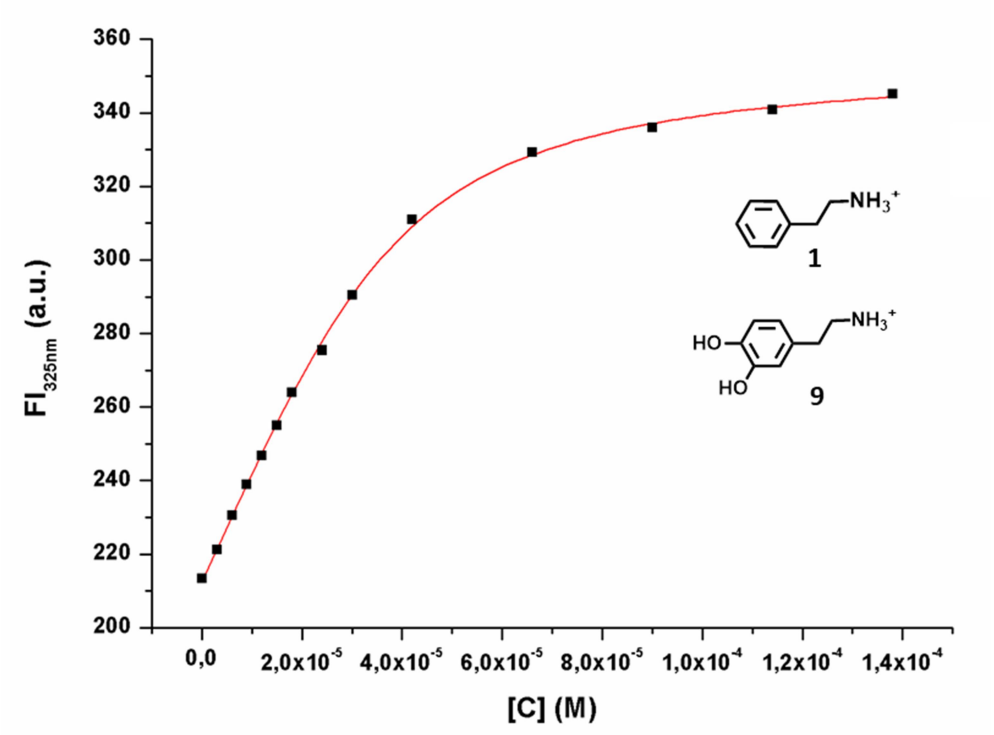


Figure S20. Plot of the fluorescence intensities vs the concentration of phenethylamine (**9**) added. In red the fitting curve obtained from DynaFit. Conditions **S1-AuNP**s 1.4 µM , Dopamine 72 µM ,HEPES 2mM $K_{(dopamine)}=1.2\times10^5$

4.3 Comparison of S1, S2, S3, S4-AuNPs

Table S1. Data of Figure 7 of the paper. Binding parameters of analytes **10**, **11**, **18**, **19** to **S1**, **S2**, **S3**, **S4**-AuNPs in water. The errors reported are derived from fitting errors from estimation of K.

Analyte	*AuNp*	*K ,M^{-1}*	*[binding sites], M*
10	**S1**	**(4.1±0.4)×10^5**	**(4.2±0.1)×10^5**
	S2	**(9.3±1.2)×10^4**	**(4.0±0.3)×10^5**
	S3	**-**[b]	**-**[b]
	S4	**(1.1±0.2)×10^5**	**(2.4±0.2)×10^5**
11	**S1**	**(4.8±0.5)×10^5**	**(3.9±0.1)×10^5**
	S2	**(8.5±1.5)×10^4**	**(4.7±0.4)×10^5**
	S3	**-**[b]	**-**[b]
	S4	**(1.0±0.2)×10^5**	**(2.3±0.3)×10^5**
18	**S1**	**(6.1±1.4)×10^5**	**(3.3±0.1)×10^5**
	S2	**(5.1±0.3)×10^5**	**(4.1±0.1)×10^5**
	S3	**-**[b]	**-**[b]
	S4	**(4.3±0.2)×10^5**	**(3.1±0.1)×10^5**
19	**S1**	**(2.2±0.1)×10^6**	**(5.5±0.1)×10^5**
	S2	**(1.3±0.1)×10^6**	**(5.7±0.1)×10^5**
	S3	**(3.6±0.1)×10^5**	**(4.7±0.1)×10^5**
	S4	**(8.5±0.1)×10^5**	**(5.4±0.1)×10^5**

a) [AuNp] = 10×10^{-5} M, pH 7.0 (HEPES buffer 10 mM); b) no binding observed.

5. Affinity tuning and NMR sensitivity

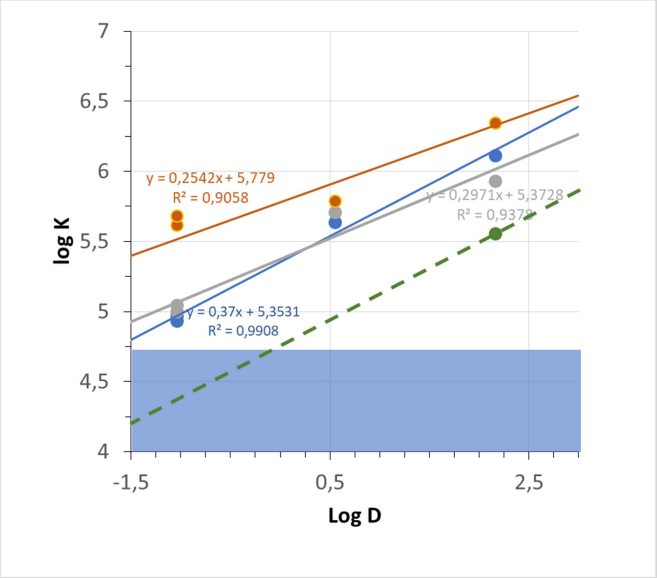


Figure S21. Plot of the log K *vs* log D (pH=7.4) values relative to the binding of the luminescent analytes **10**, **11, 18** and **19** to **S1**/**S4**-AuNp (orange: **S1**-AuNp, blue: **S2**-AuNp, green: **S3**-AuNp, grey: **S4**-AuNp). The lines represent the linear fit of the data for **S1**-AuNp, **S2**-AuNp and **S4**-AuNp, in the case of **S3**-AuNp the green dotted line is not the result of a fit and it has been drawn with the same slope of the one relative to **S2**-AuNp. The blue area represents the binding constant values which cannot be measured by fluorescence titrations.

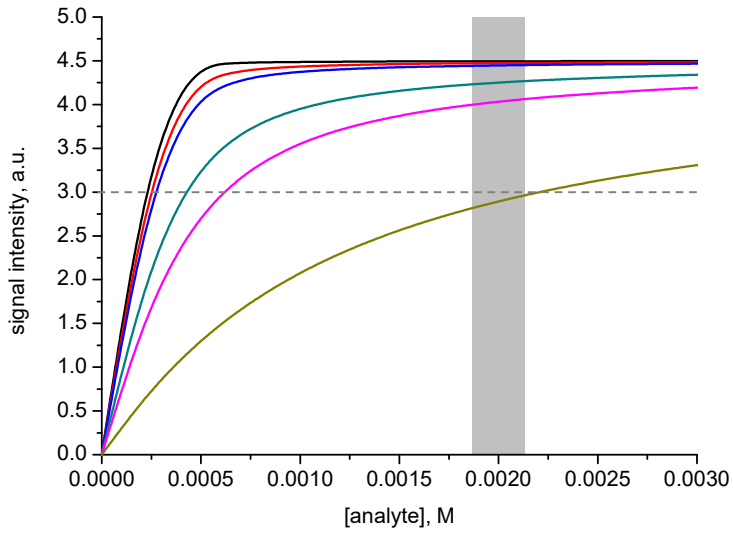


Figure S22. Simulated plots of the NOE pumping signal intensities for different binding constants ([binding sites] = 5×10^{-5} M). From the top: K = 5×10^{5} M^{-1}, 1×10^{5} M^{-1}, 5×10^{4} M^{-1}, 1×10^{4} M^{-1}, 5×10^{3} M^{-1}, 1×10^{3} M^{-1}.

The grey area evidences the typical analyte concentration used in this work (2 mM): signal intensity is not significantly affected by the binding constant when above 5×10^{3} M^{-1}.

The grey dotted line represents the limit of detection with the typical signal intensities found in this work: expected limit of detection for binding constant larger than 5×10^{4} M^{-1} is about 0.25 mM. Larger values are expected in experiments because of the partition of the analytes in the monolayer, which increases the relaxation times.

6. Additional NMR experiments

NOE-pumping spectra were acquired as reported in previous works[3,4]. Main parameters used: spectral width = 6 KHz, acquired points = 16 k, number of scans = 3072, recycle delay = 2 s, mixing time = 1.2 s, diffusion delay Δ = 50 ms.

STD spectra were performed using the Bruker *stddiff.2* sequence with a saturation time D20=2 s saturating at 10000 Hz for the off resonance experiment.

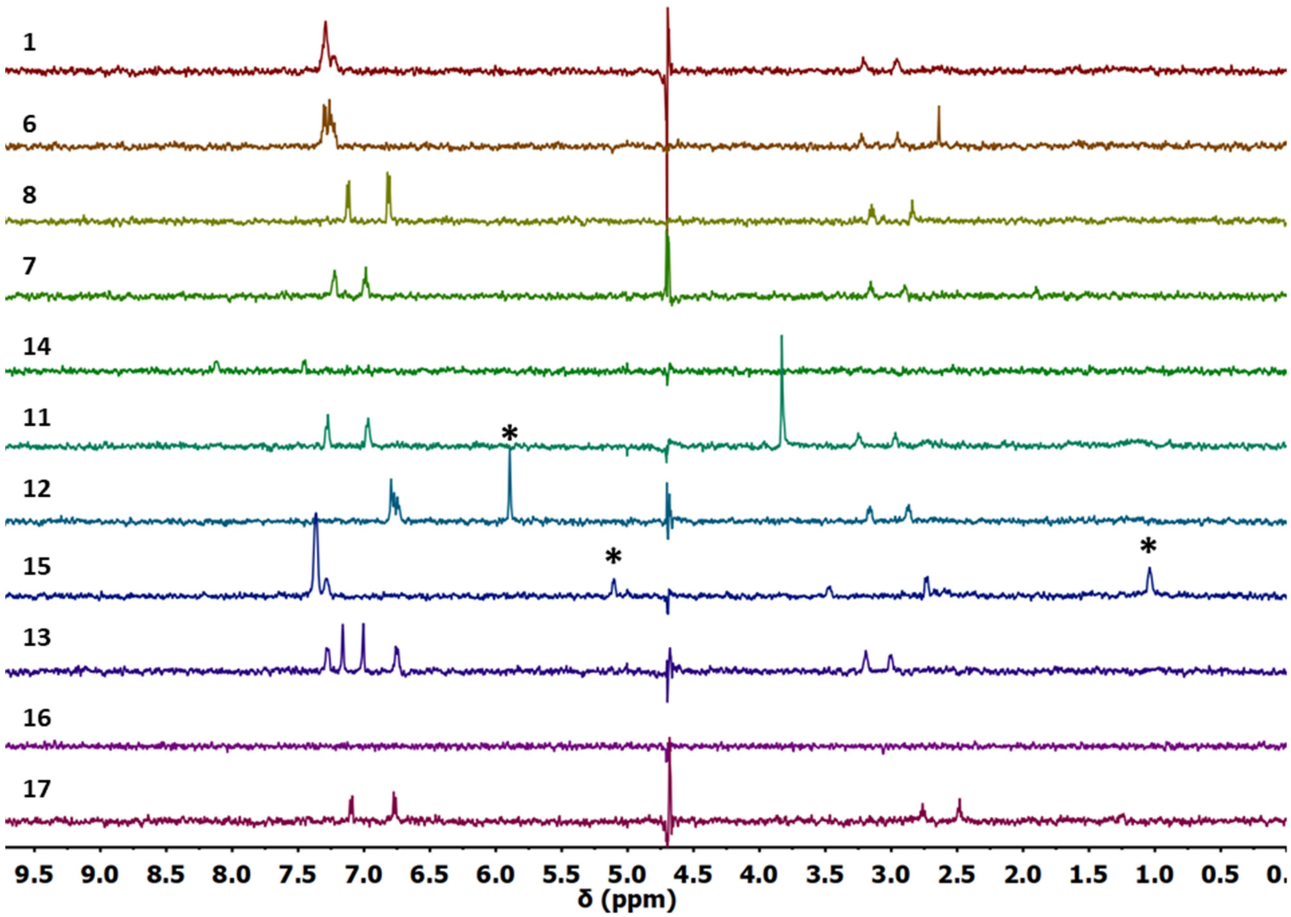


Figure S23. ^{1}H-NMR NOE pumping-CPMGz full spectra (3072 scan, 4 h) of AuNP-S1 (14 μM in D2O), HEPES buffer (10.0 mM) and different analytes (2 mM): (a) – (k). For 4-nitrophenethylamine (e) the NOE pumping spectrum is shown (same acquisition parameters).

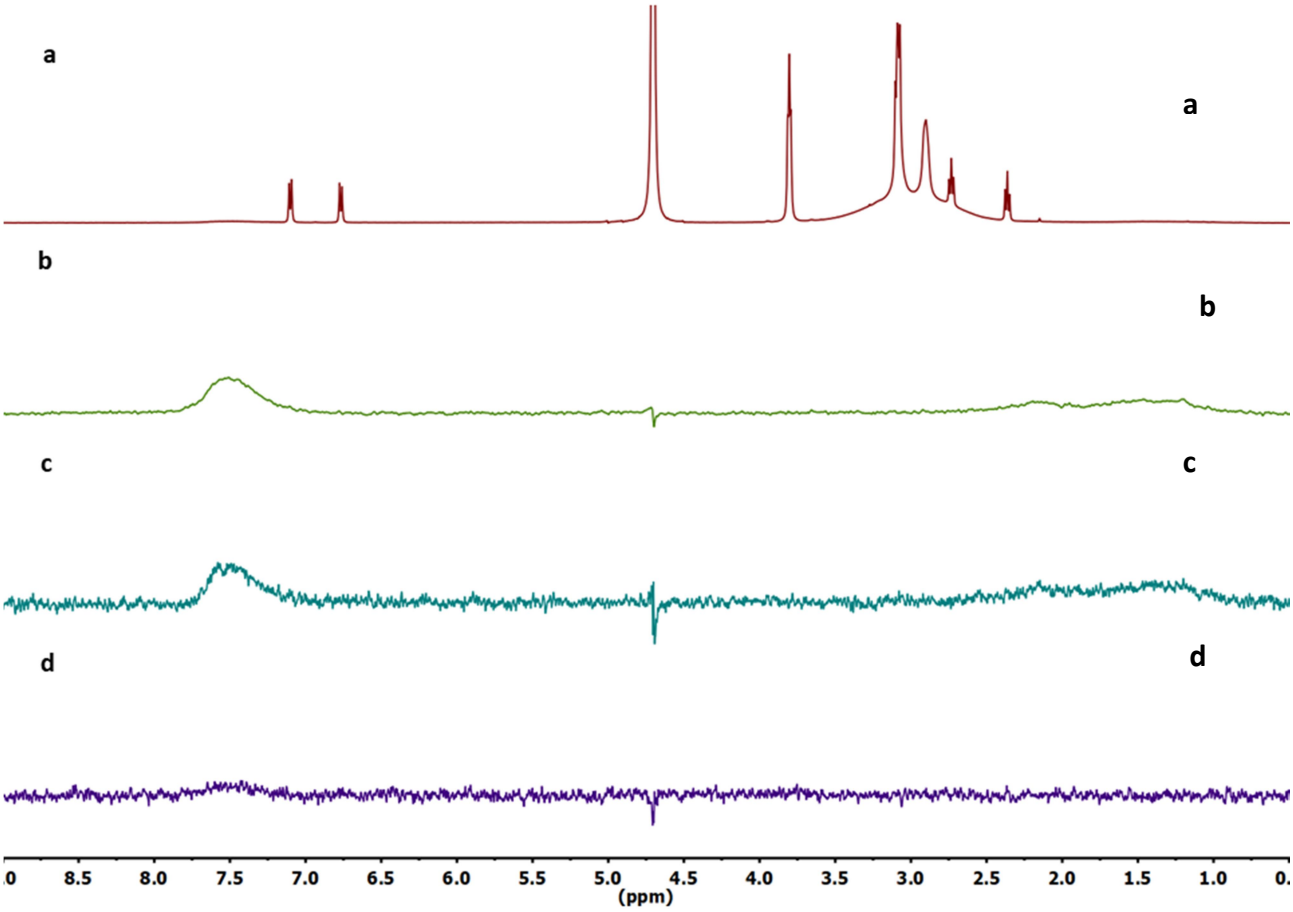


Figure S24. (a) ^{1}H-NMR; (b) diffusion filter spectrum (640 scan, 40 min); (c) NOE-Pumping experiment (3072 scan, 4h); (d) NOE-Pumping CPMGz experiment (3072 scan, 4h). Conditions: AuNP-S2 (15 μM), phloretic acid (2 mM), HEPES (10 mM, pD 7.0), D_2O. Phloretic acid is not detected in these conditions.

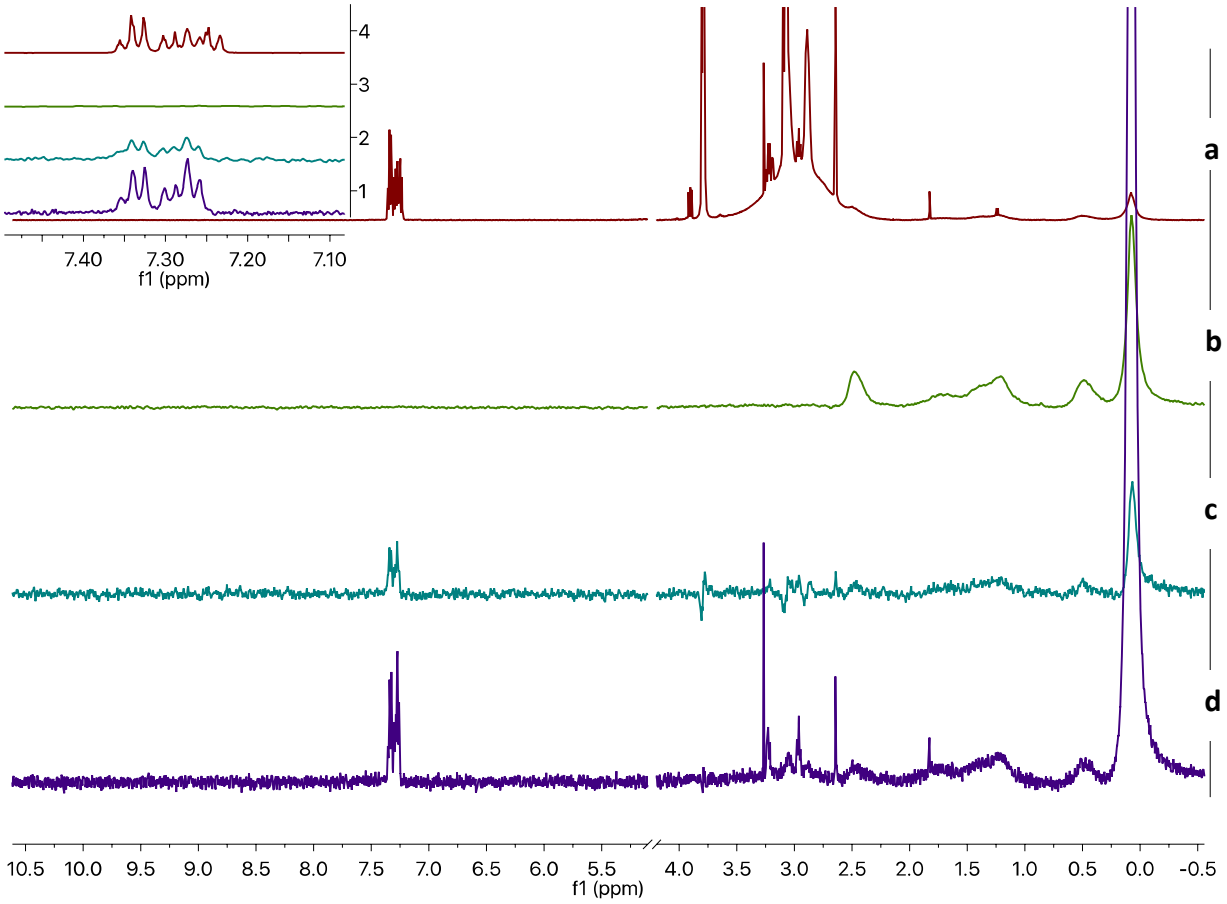


Figure S25. Comparison between NOE pumping experiment and STD experiment (a) ^{1}H-NMR of the mixture; (b) diffusion filter spectrum; (c) NOE-Pumping experiment; (d) STD-NMR experiment (on res. 40 Hz). Conditions: (15 μM), Phenylalanine (1 mM), N-Methylphenetylamine (1 mM), HEPES (10 mM, pD 7.0), D_2O. In the inset is highlighted the aromatic region.

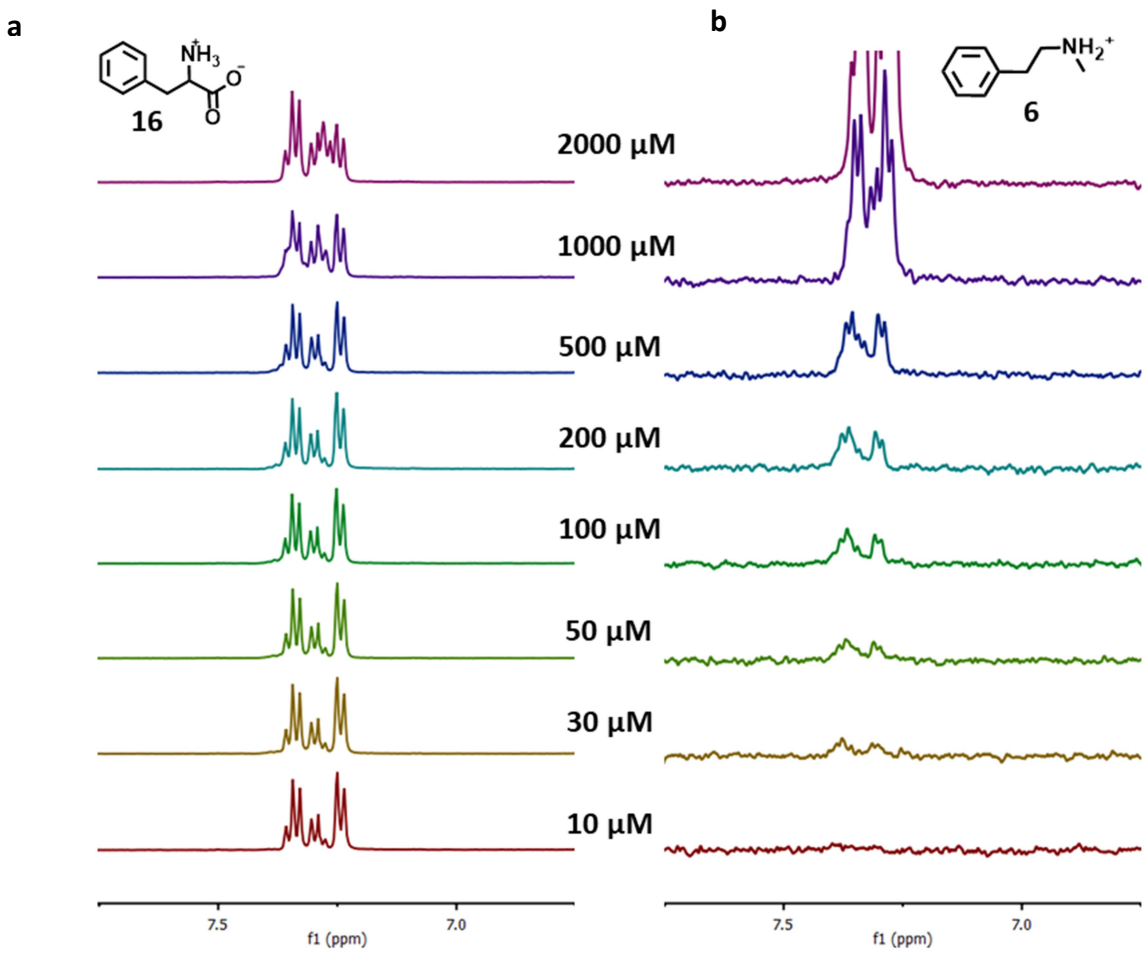


Figure S26. N-Methylphenetylamine detection: **S4**-AuNP (15 µM), Phenylalanine (1 mM), N-Methylphenetylamine (from bottom to the top: 10, 30, 50, 100, 200, 500, 1000, 2000 µM), HEPES (10 mM, pD 7.0), D_2O: (**a**) ^{1}H-NMR and (**b**) STD-NMR experiment (on res. 40 Hz) of the same mixtures.

a

7. ^{1}H, ^{13}C and ^{29}Si NMR spectra of the synthesized compounds

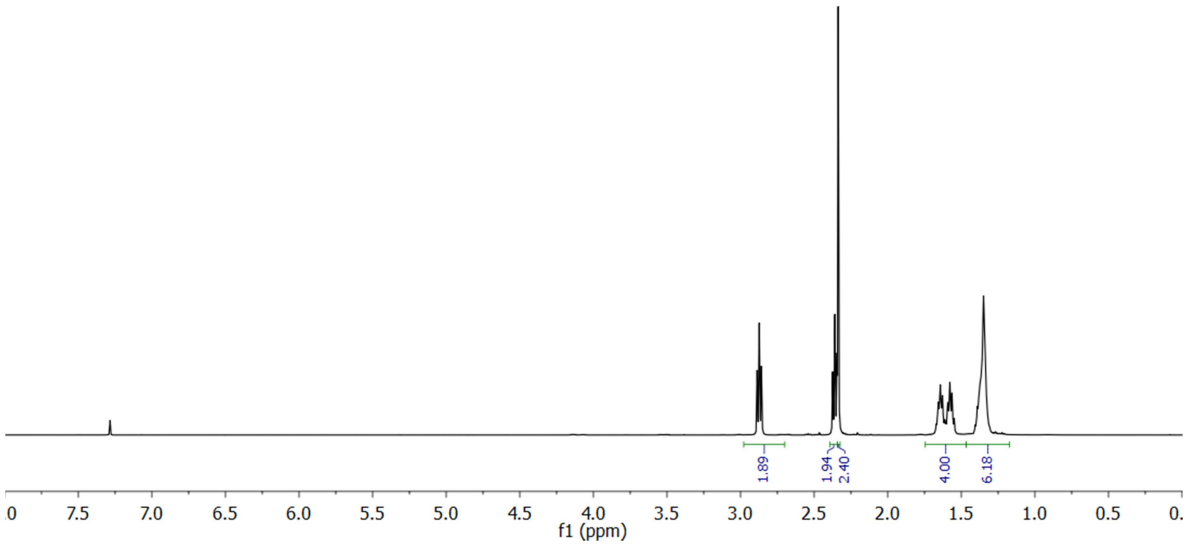


Figure S27. ^{1}H-NMR of compound **3** in $CDCl_3$.

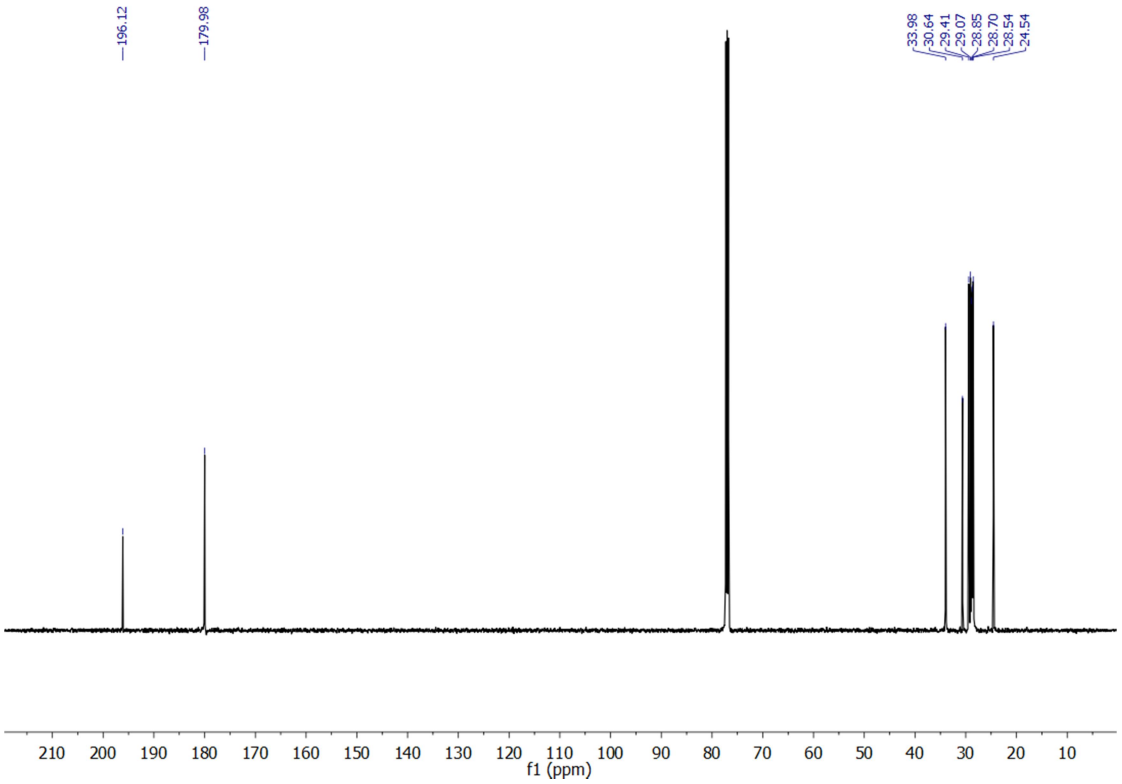


Figure S28. ^{13}C-NMR of compound **3** in $CDCl_3$.

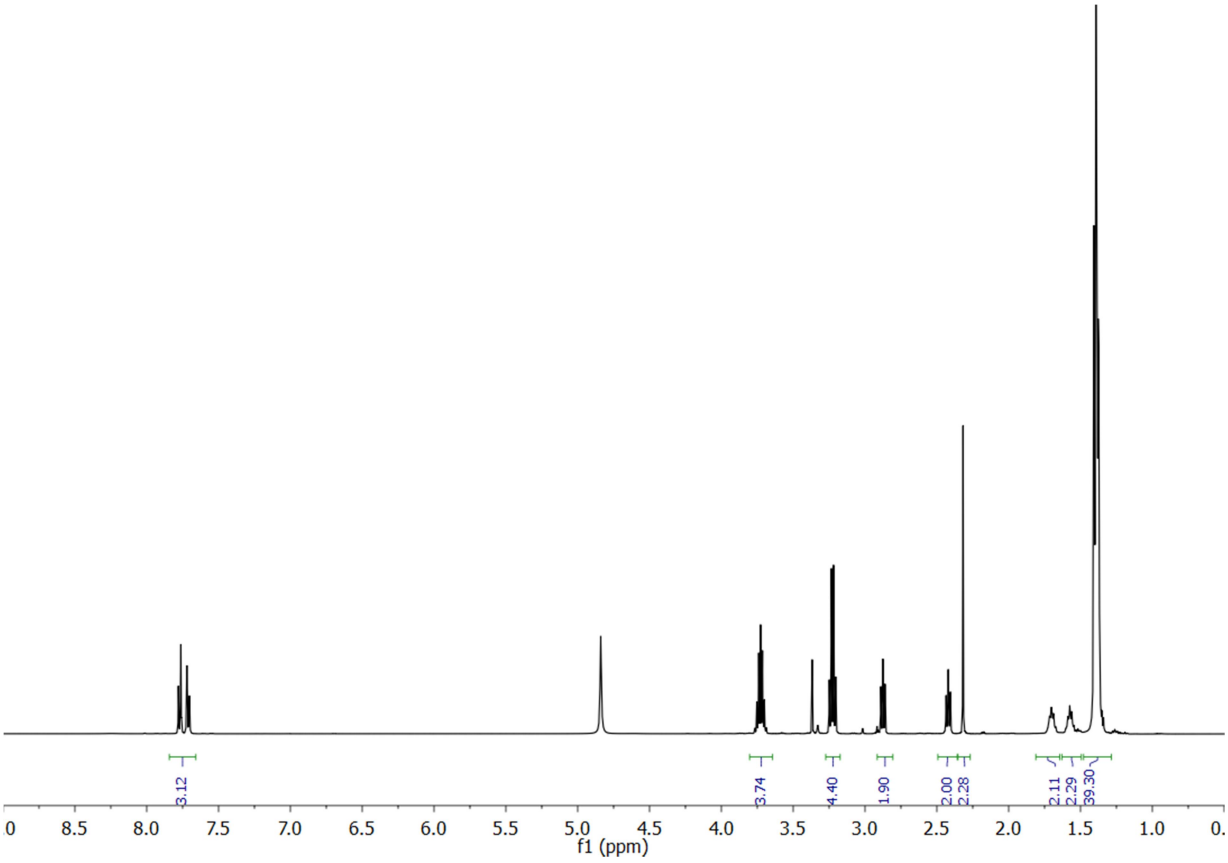


Figure S29. [1]H-NMR of compound **4** in MeOD.

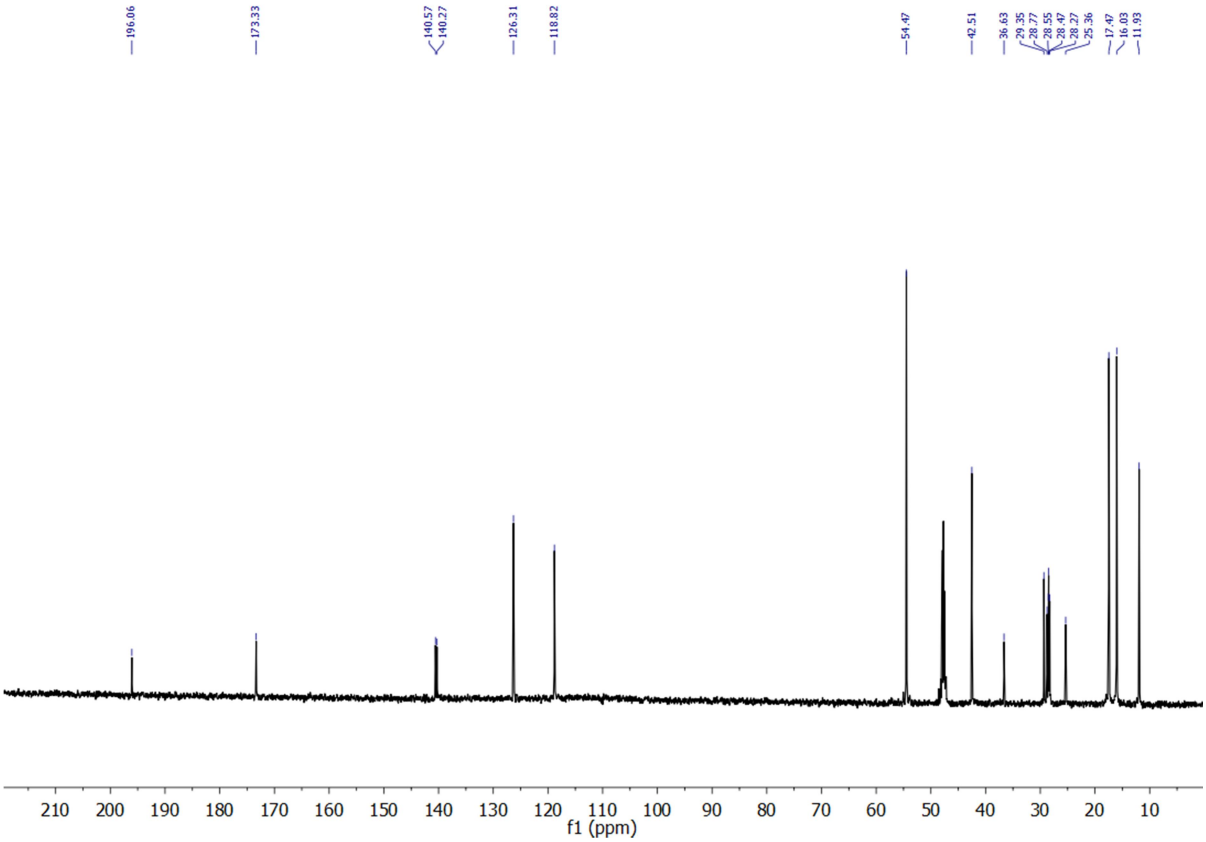


Figure S30. [13]C-NMR of compound **4** in MeOD.

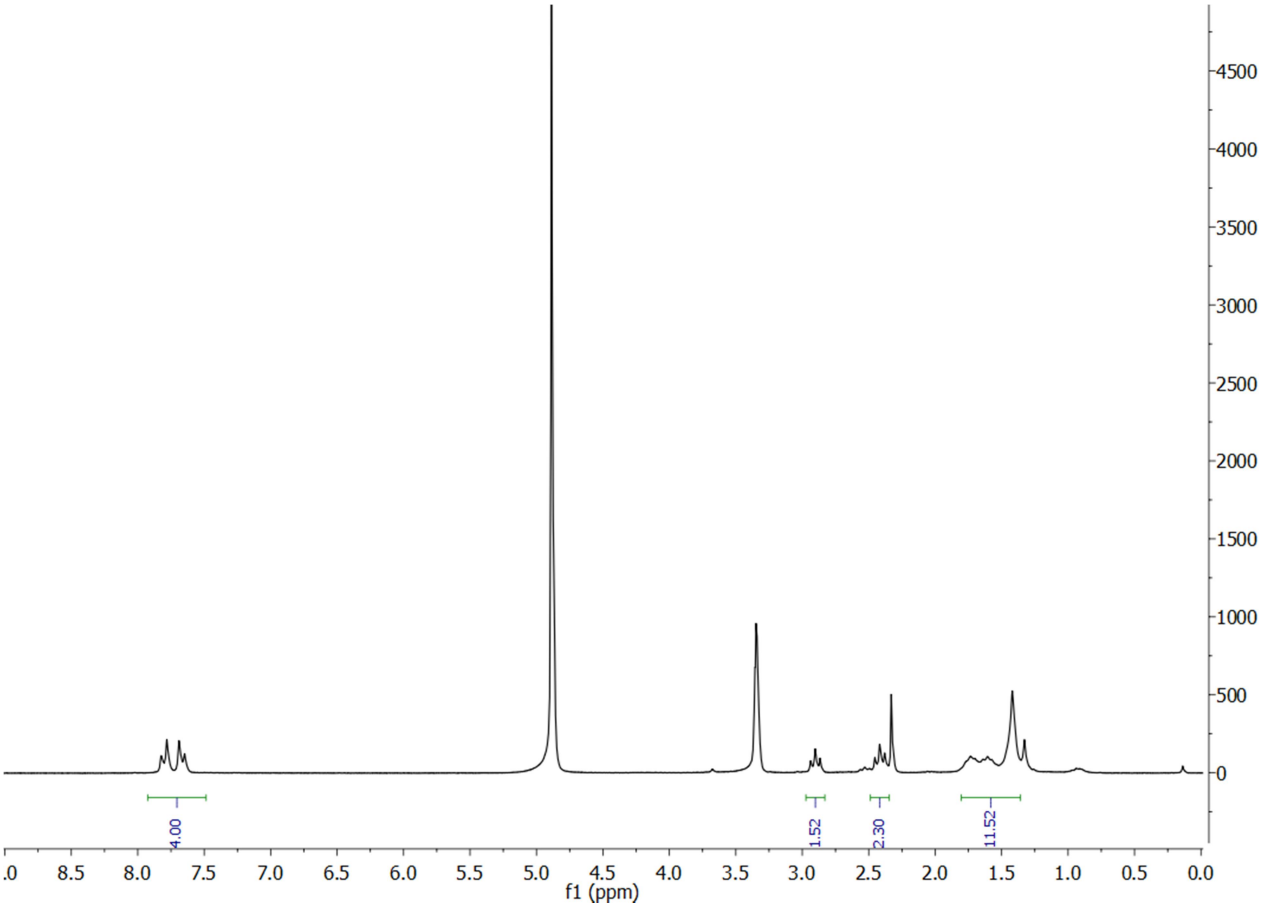


Figure S31. [1]H-NMR of compound **S2** in MeOD.

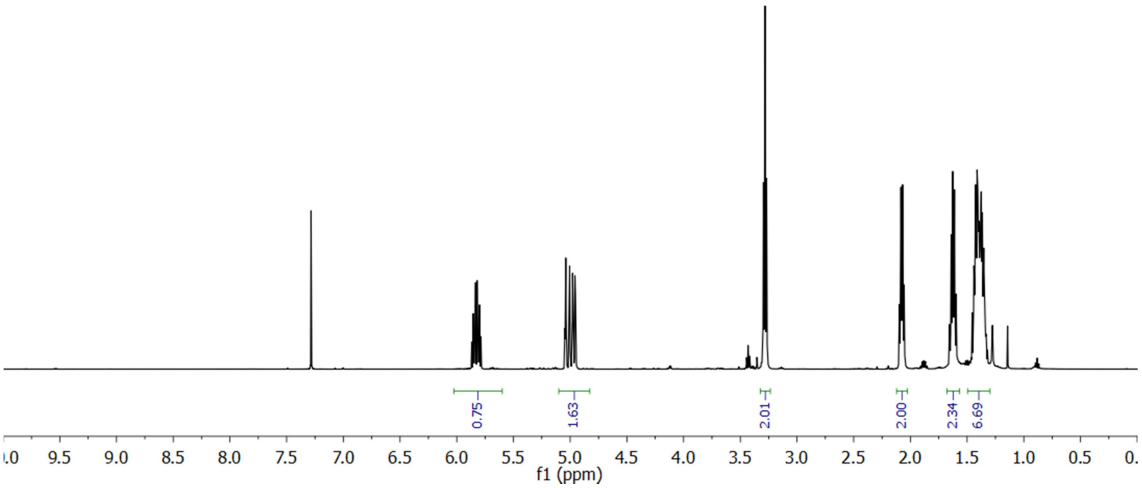


Figure S32. [1]H-NMR of compound **5** in $CDCl_3$.

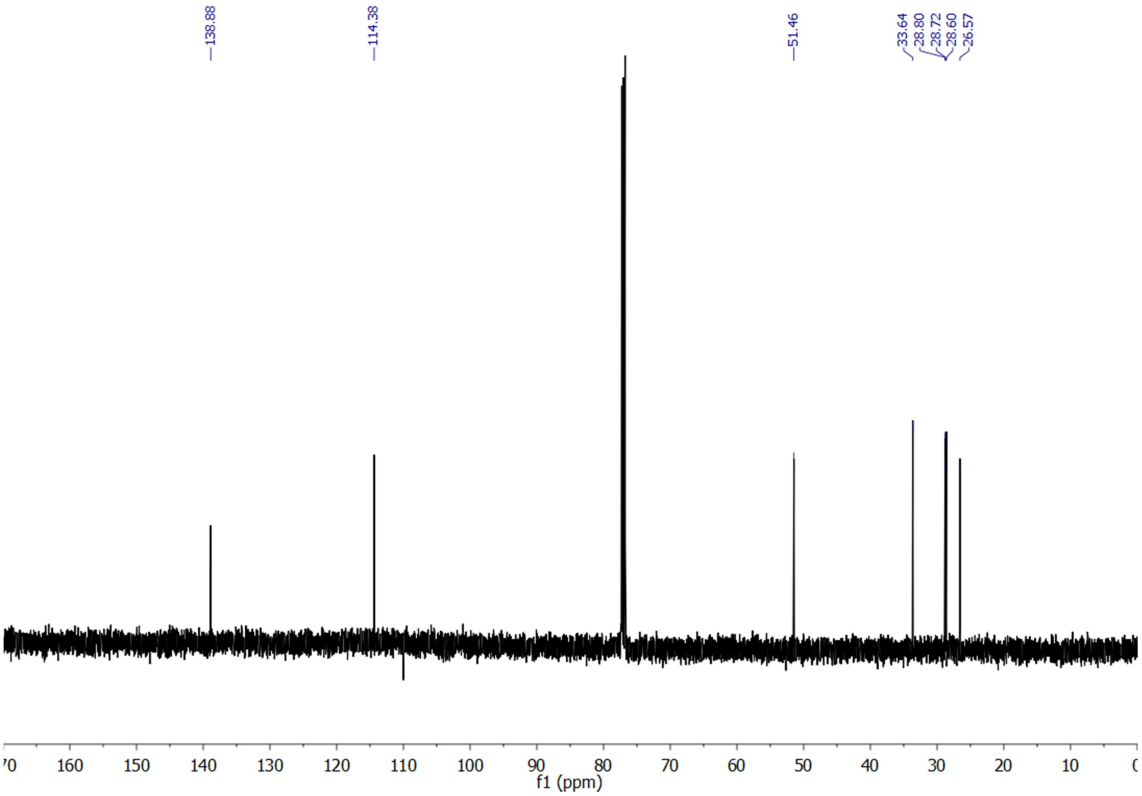


Figure S33. ^{13}C-NMR of compound **5** in $CDCl_3$.

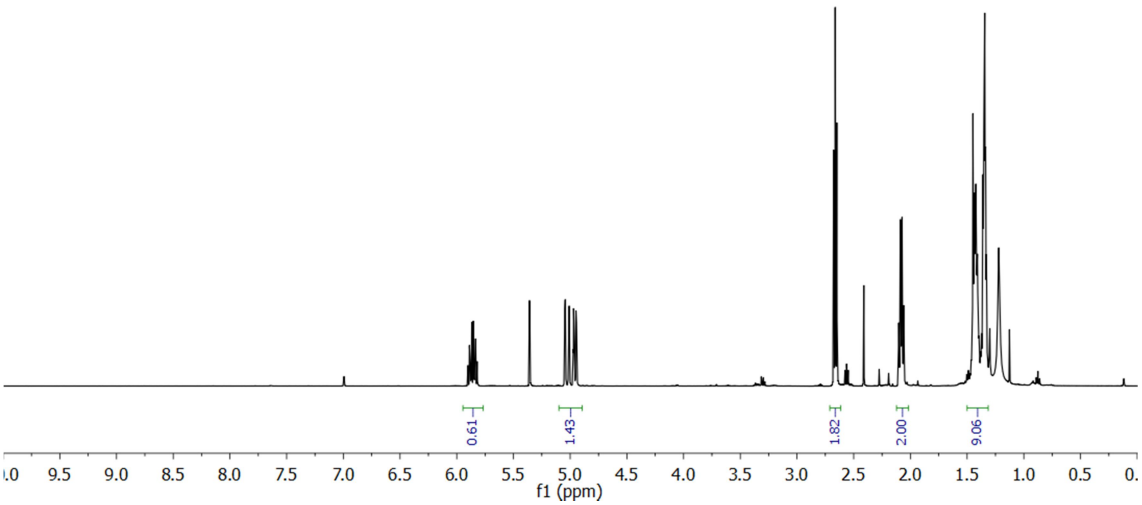


Figure S34. ^{1}H-NMR of compound **6** in $CDCl_3$.

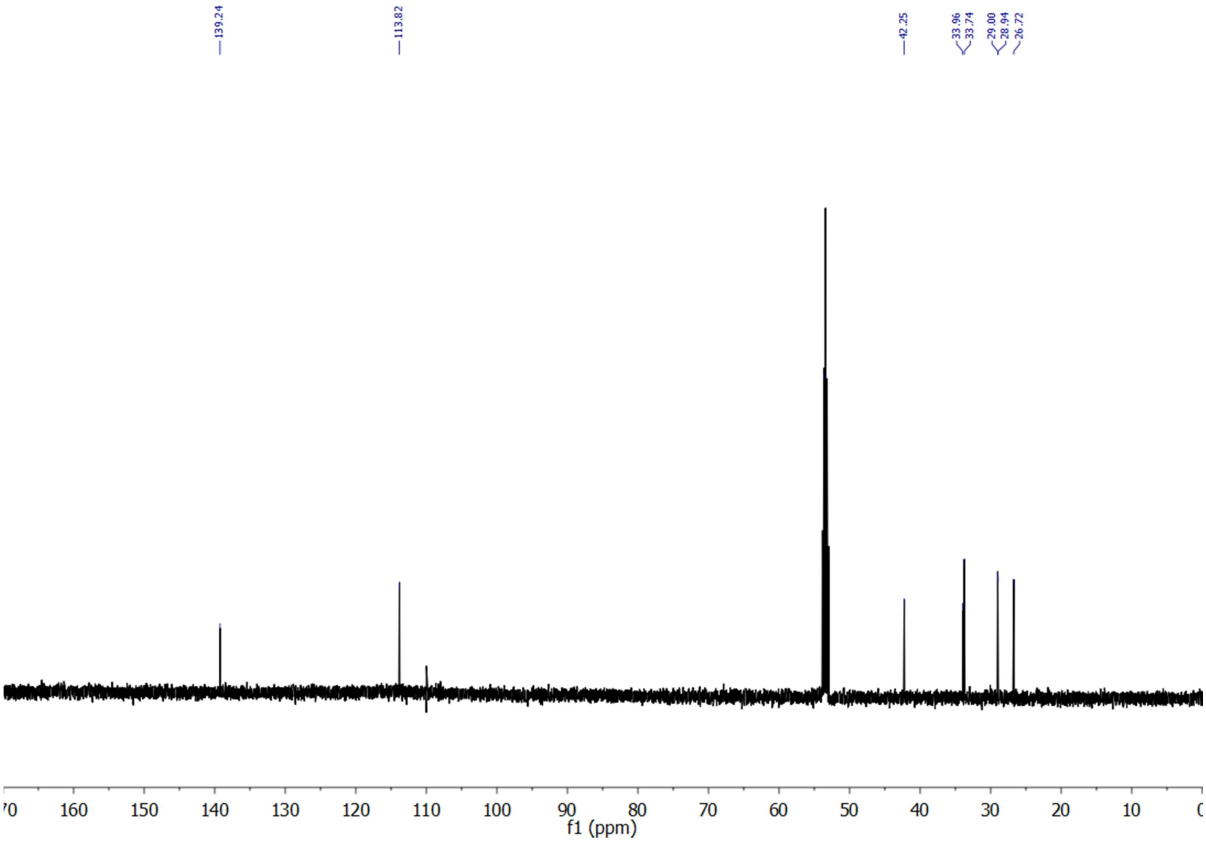


Figure S35. ^{13}C-NMR of compound **6** in $CDCl_3$.

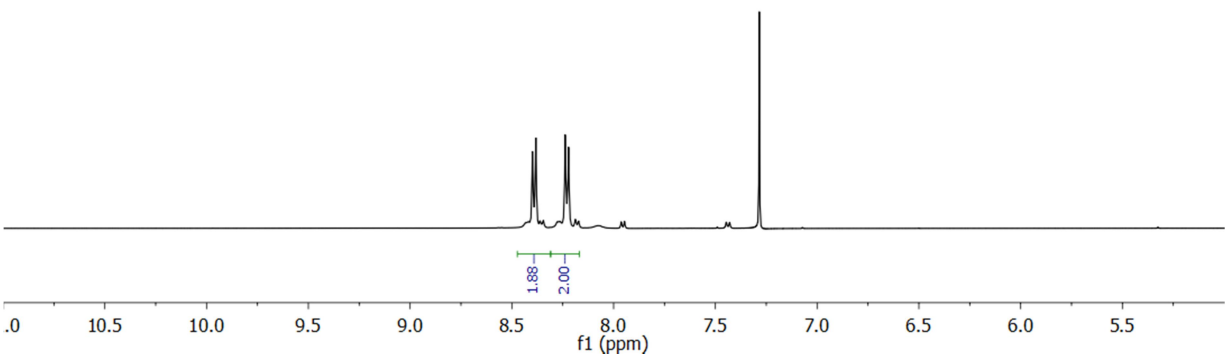


Figure S36. ^{1}H-NMR of compound **7** in $CDCl_3$.

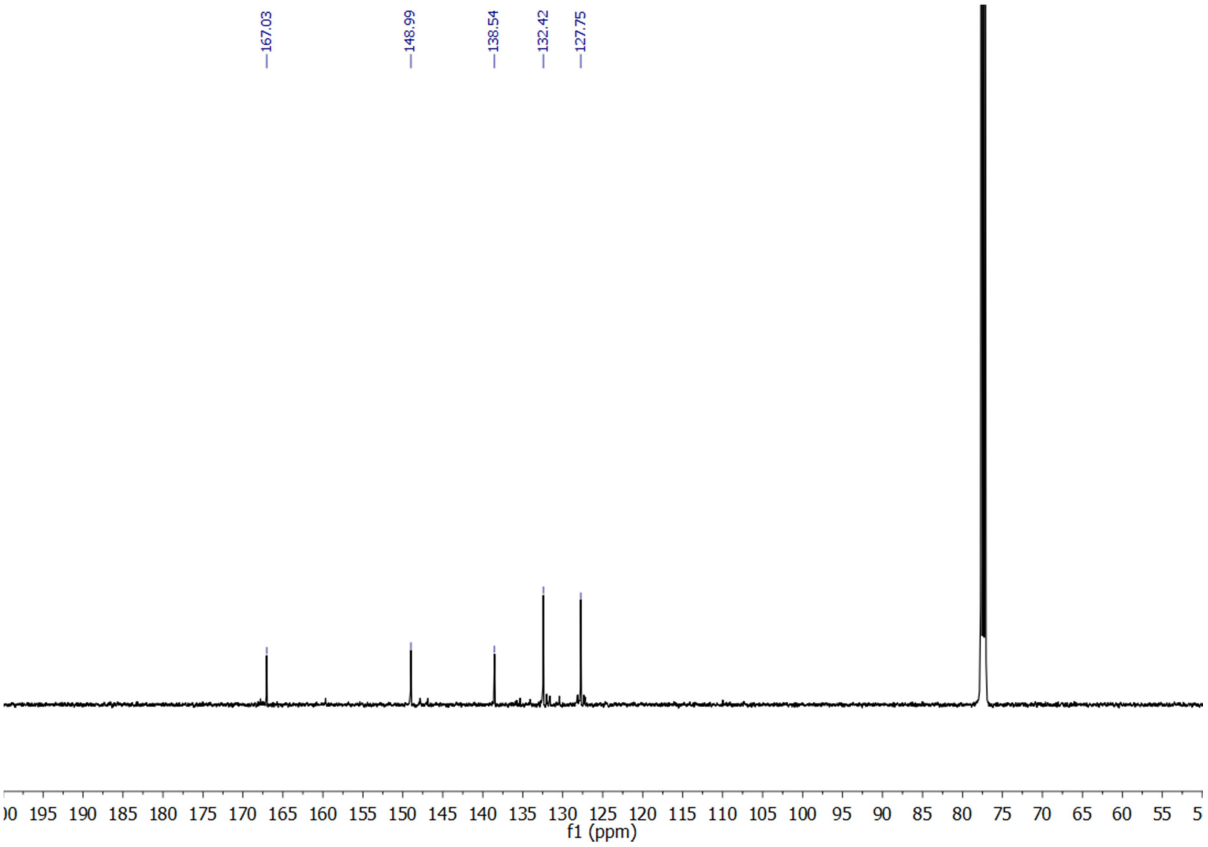


Figure S37. ^{13}C-NMR of compound **7** in $CDCl_3$.

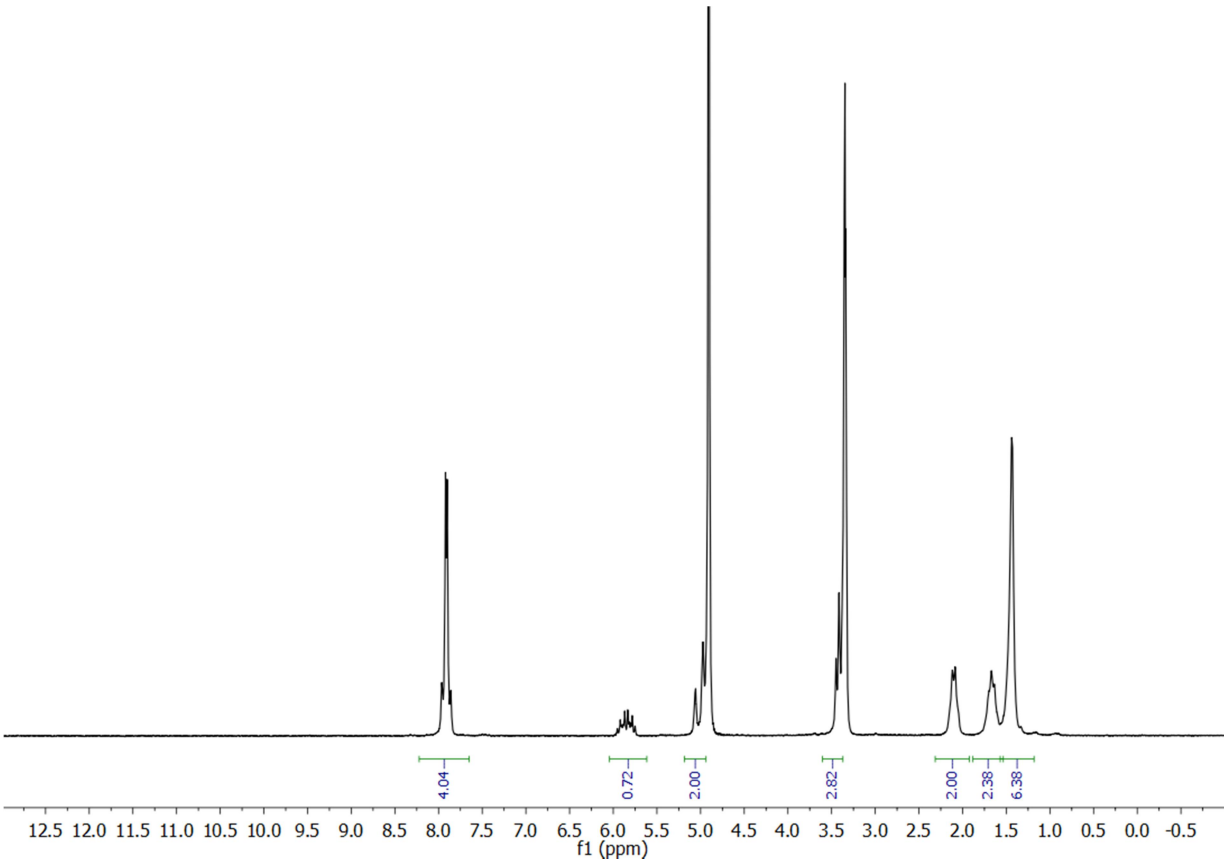


Figure S38. ^{1}H-NMR of compound **8** in MeOD.

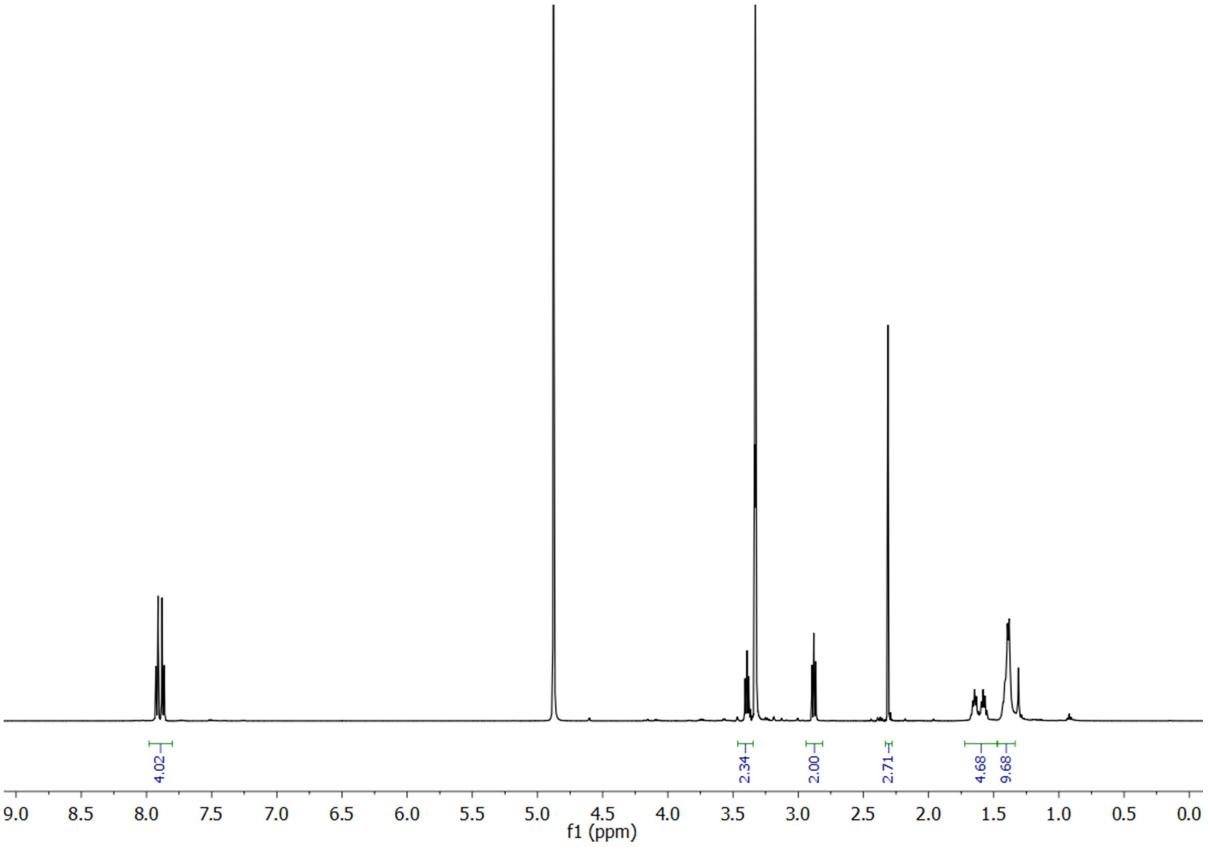


Figure S39. ^{1}H-NMR of compound **9** in $MeOD_3$.

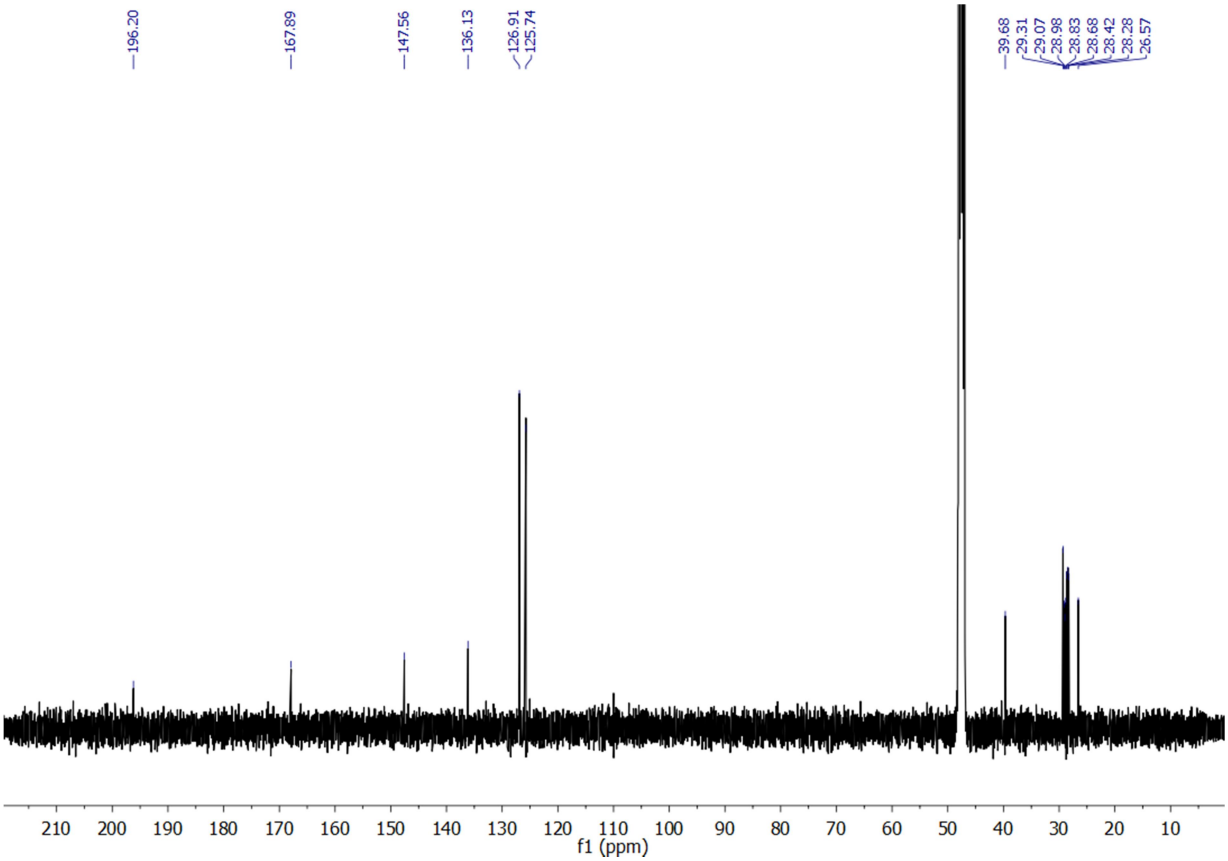


Figure S40. ^{13}C-NMR of compound **9** in MeOD.

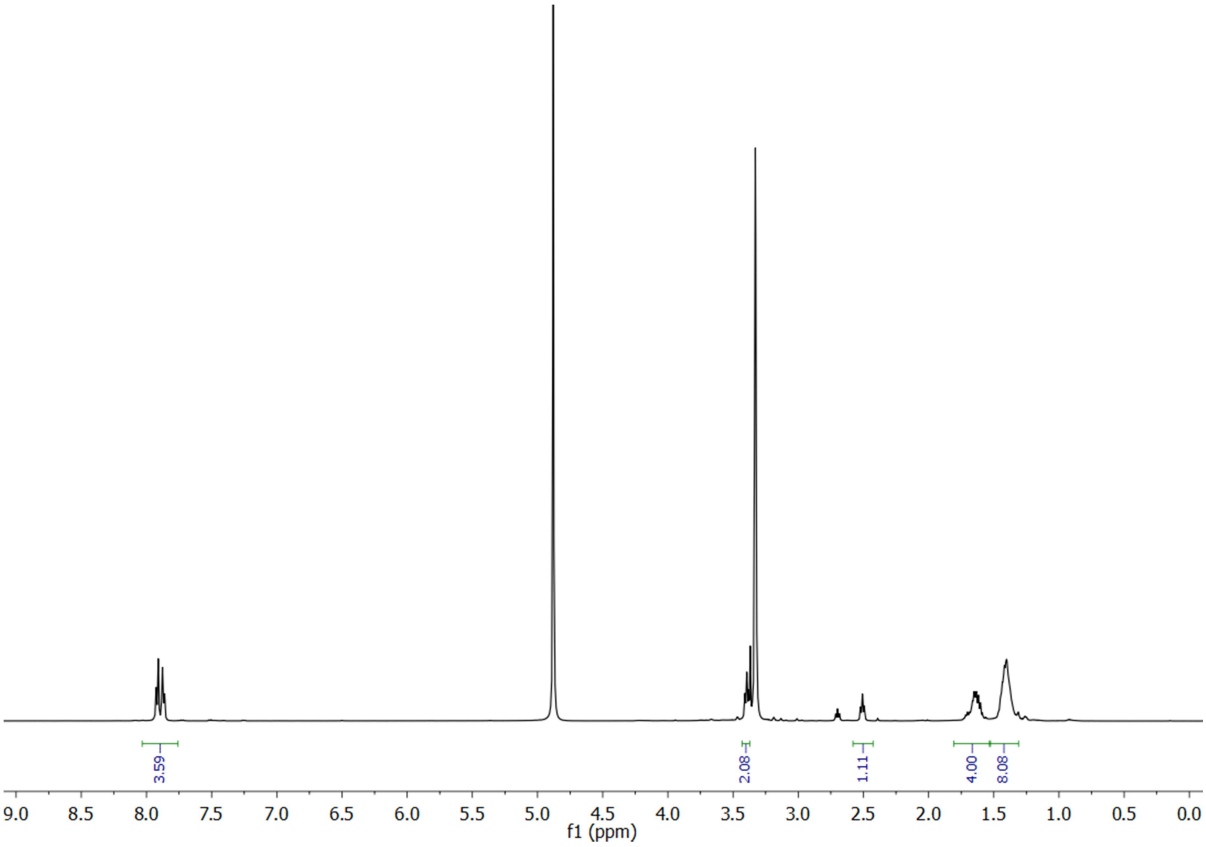


Figure S41. ^{1}H-NMR of compound **S3** in $MeOD_3$.

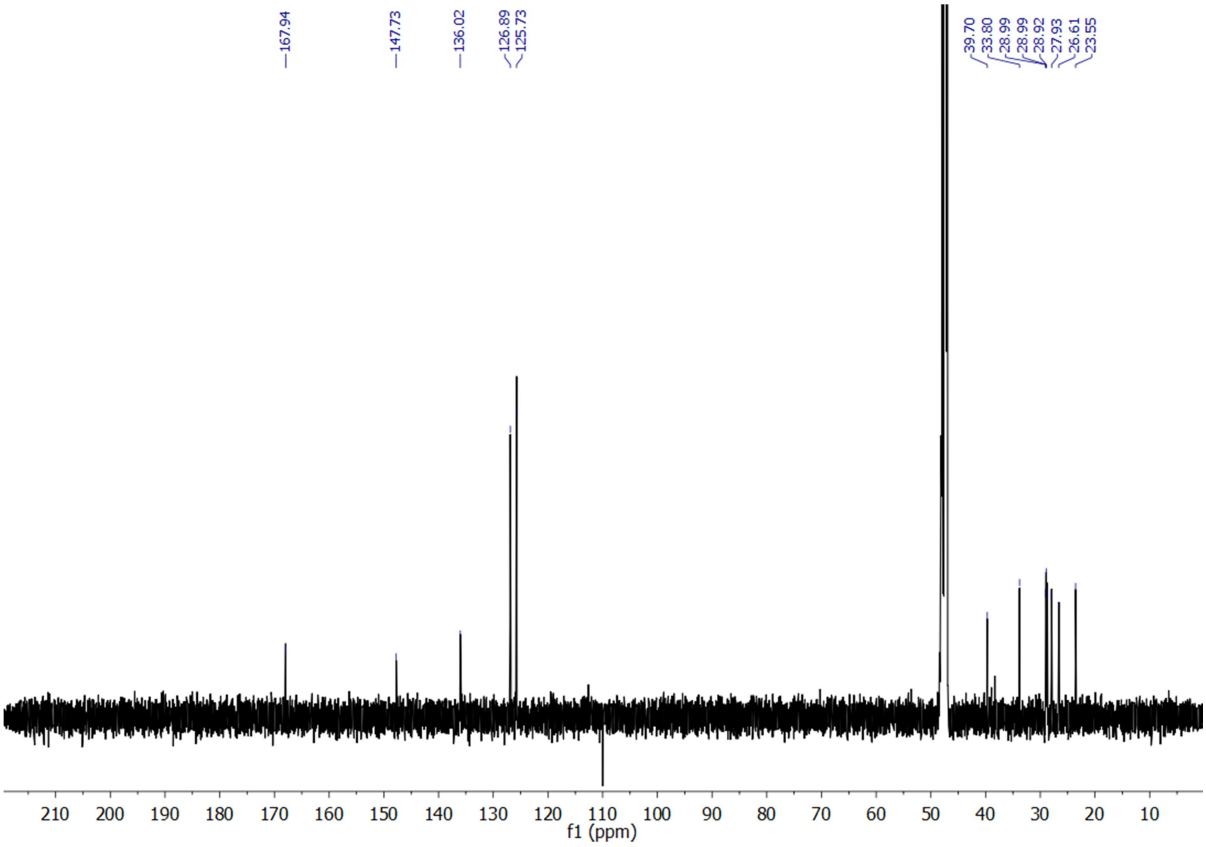


Figure S42. ^{13}C-NMR of compound **S3** in MeOD.

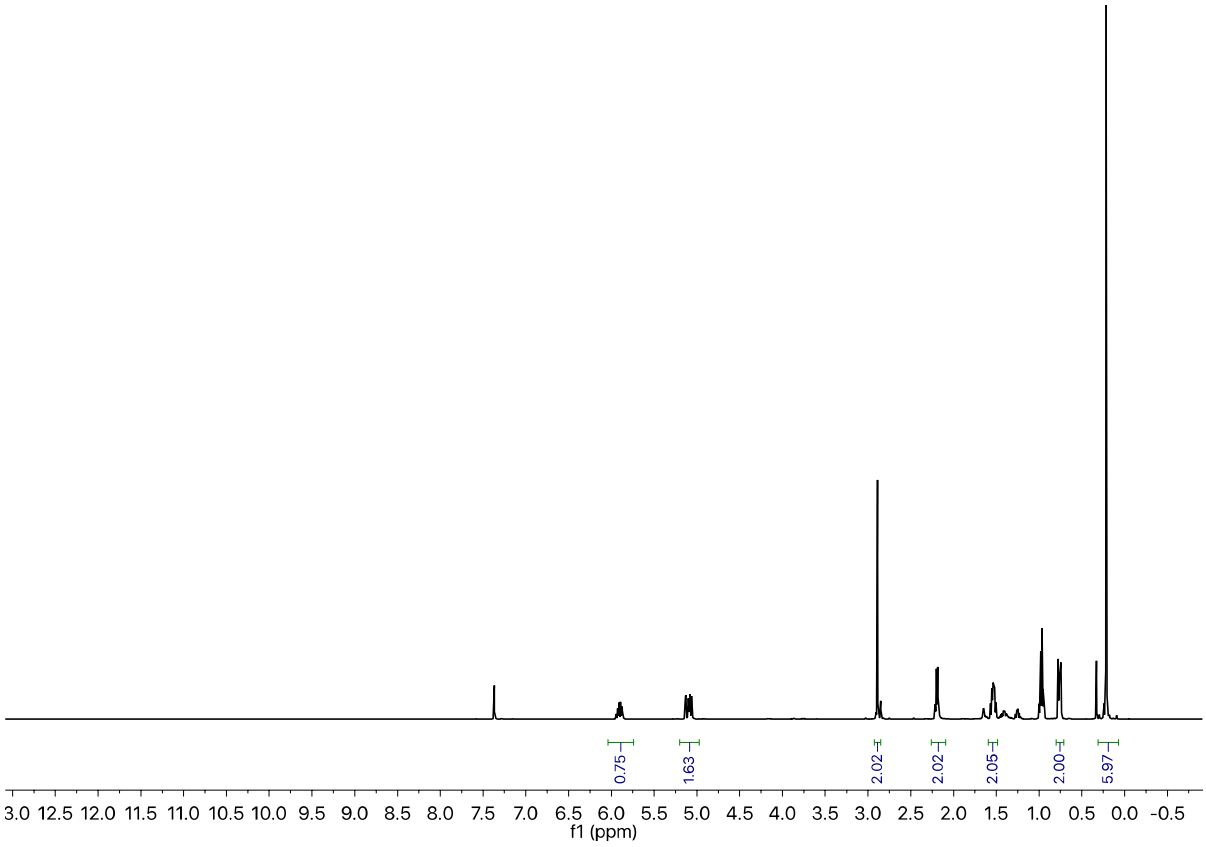


Figure S43. ^{1}H-NMR of compound **10** in $CDCl_3$.

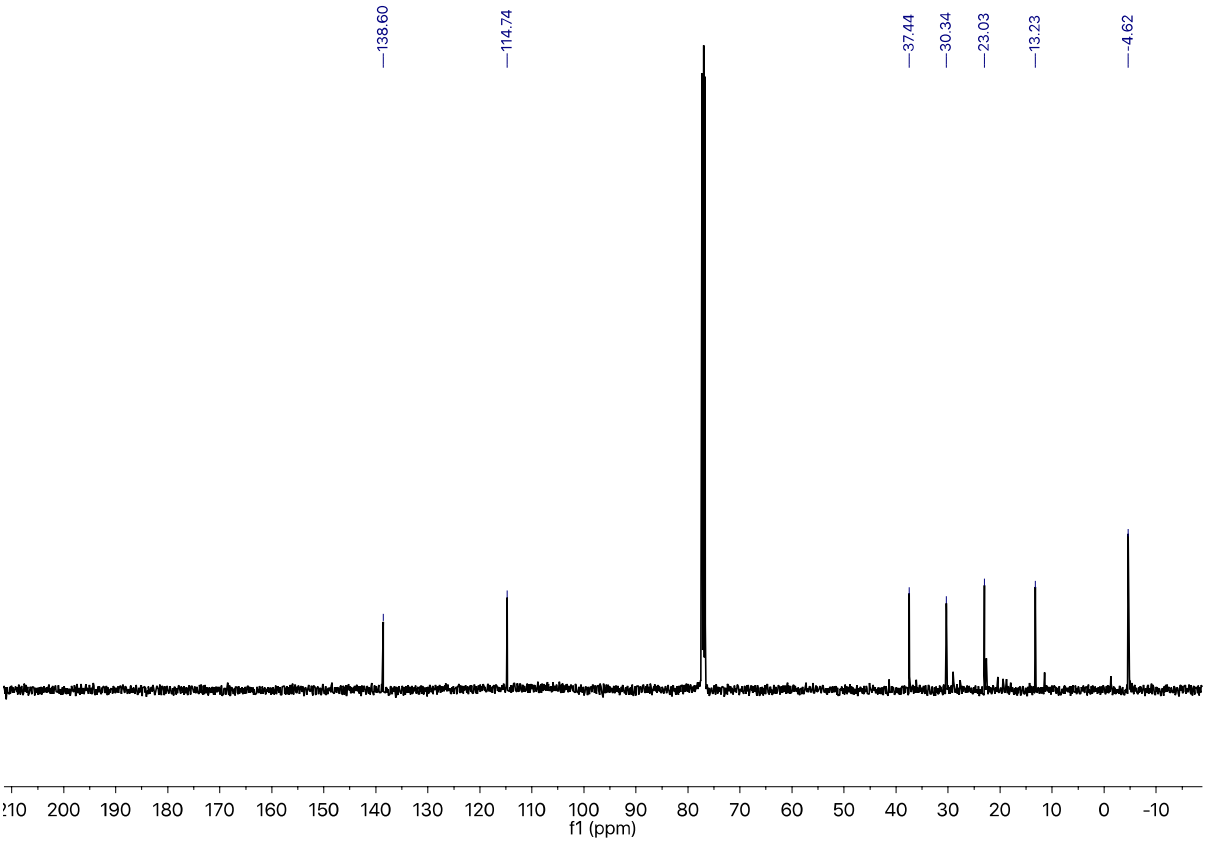


Figure S44. ^{13}C-NMR of compound **10** in $CDCl_3$.

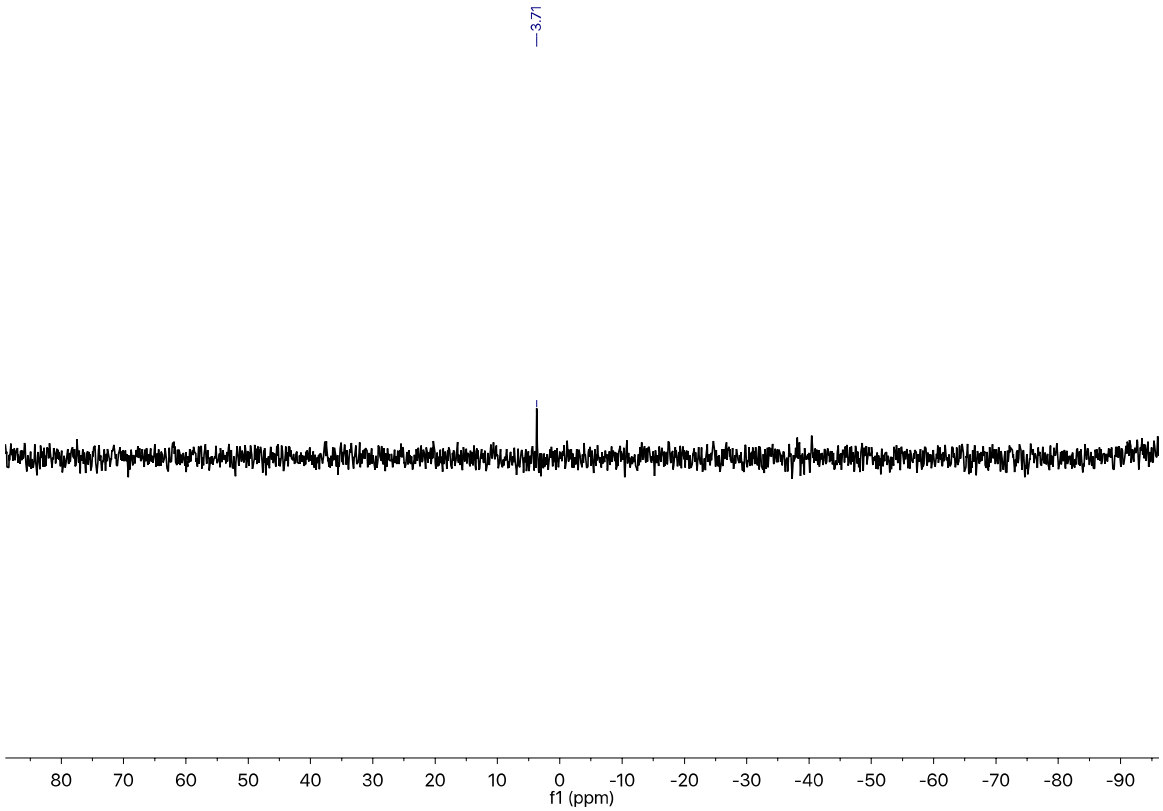


Figure S45. ^{29}Si-NMR of compound **10** in $CDCl_3$.

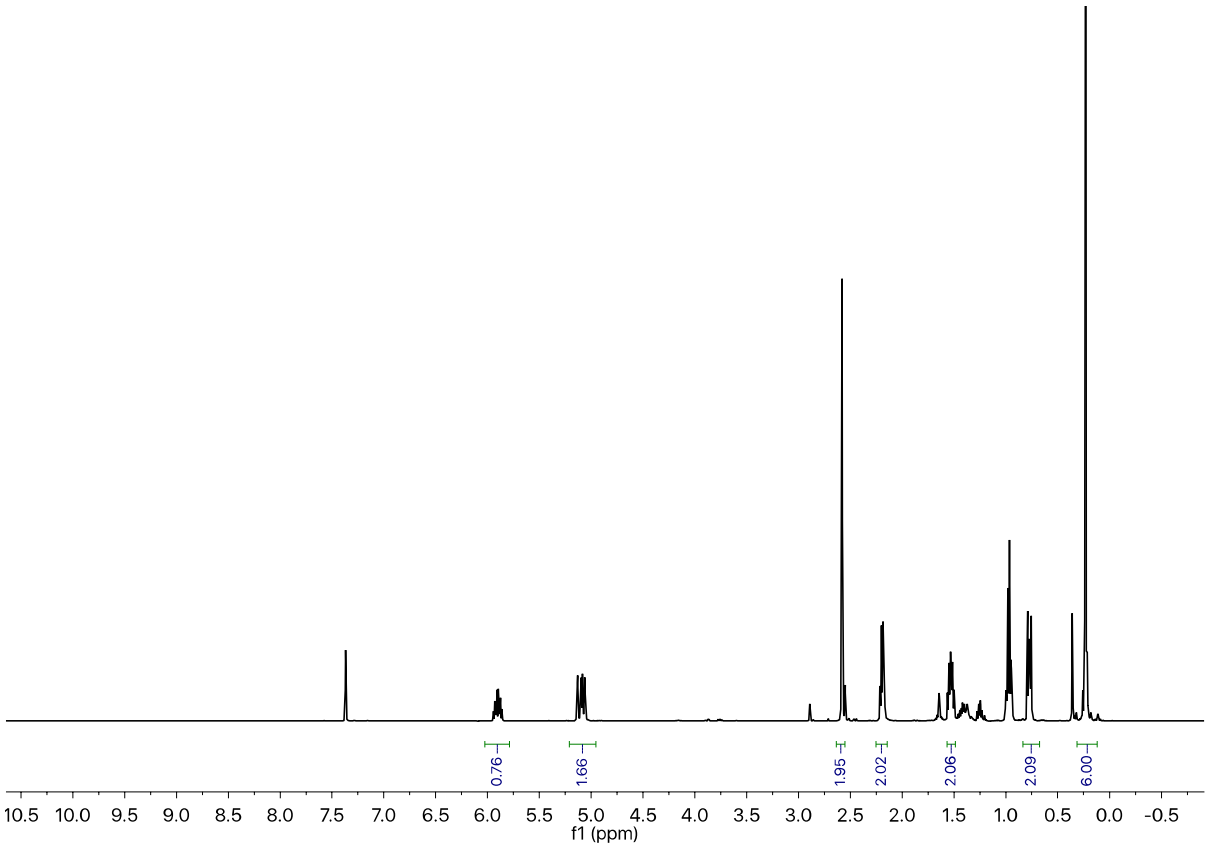


Figure S46. ^{1}H-NMR of compound **11** in $CDCl_3$.

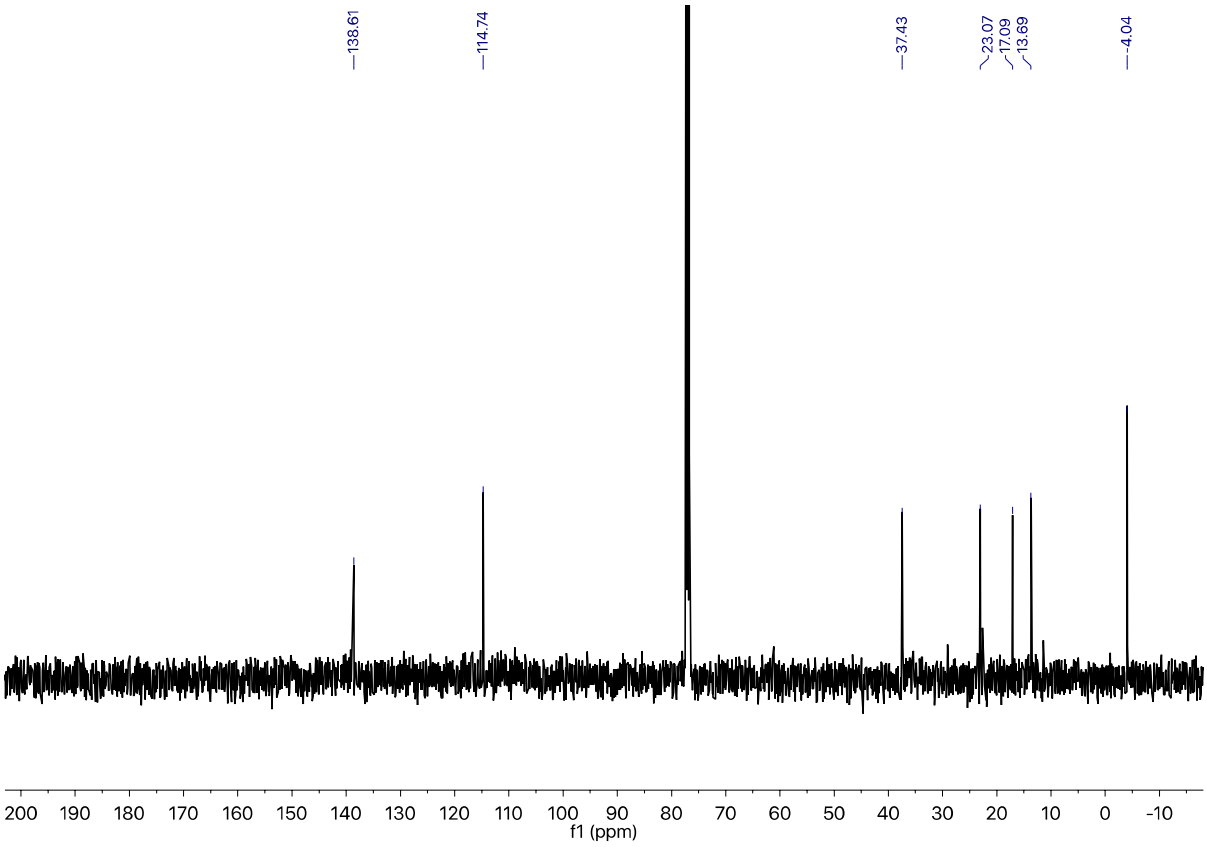


Figure S47. ^{13}C-NMR of compound **11** in $CDCl_3$.

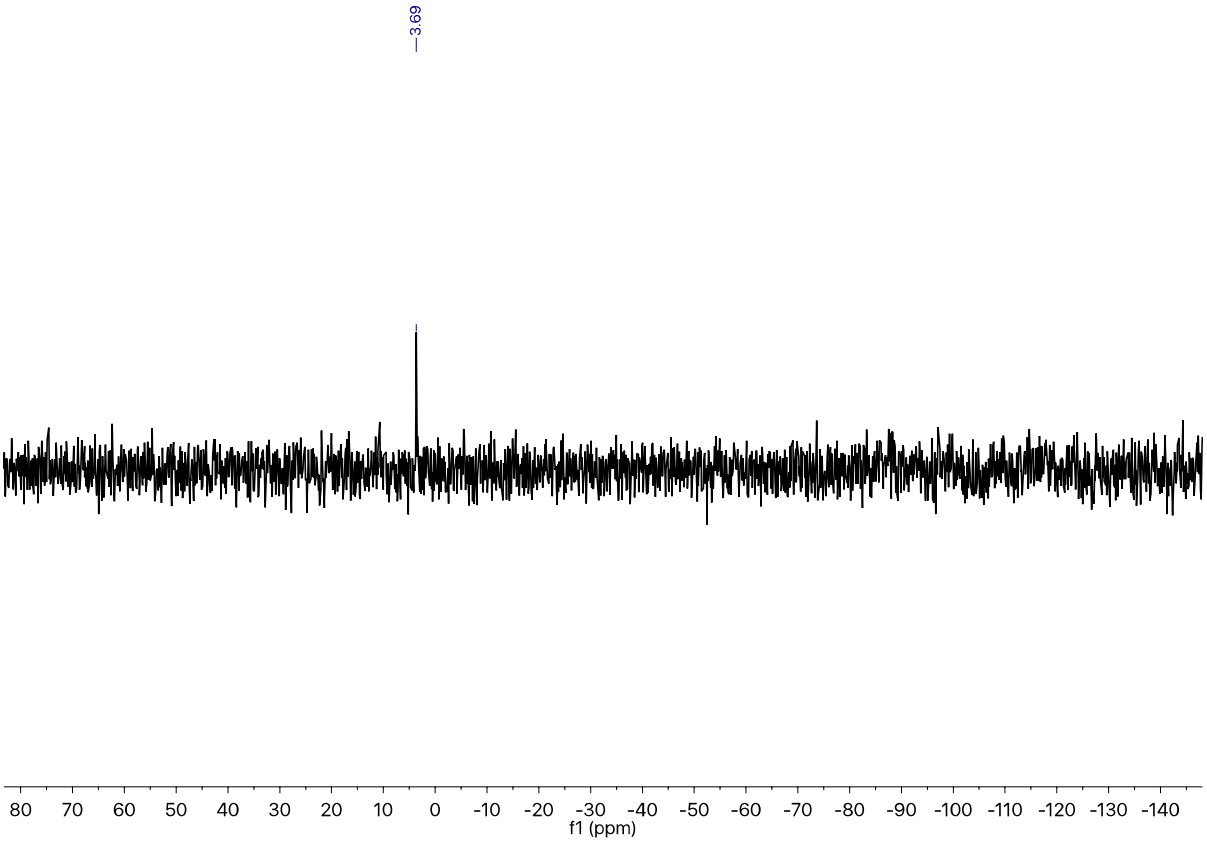


Figure S48. ^{29}Si-NMR of compound **11** in $CDCl_3$.

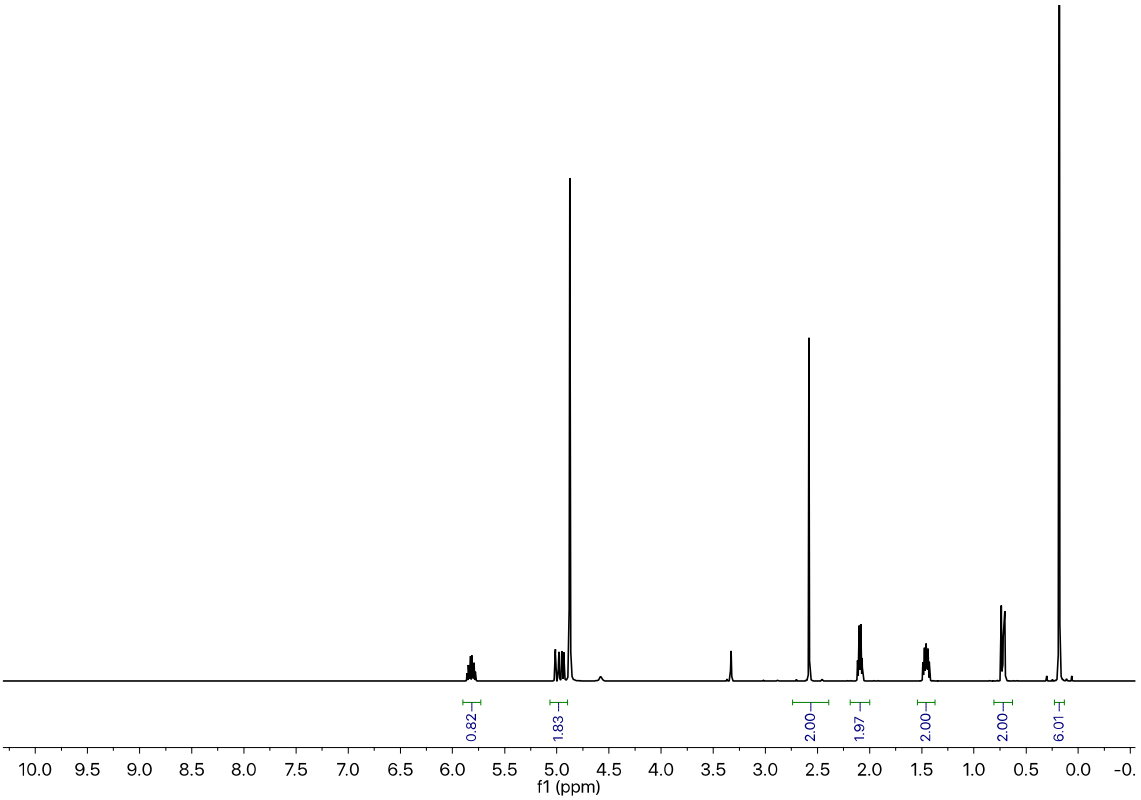


Figure S49. [1]H-NMR of compound **12** in MeOD.

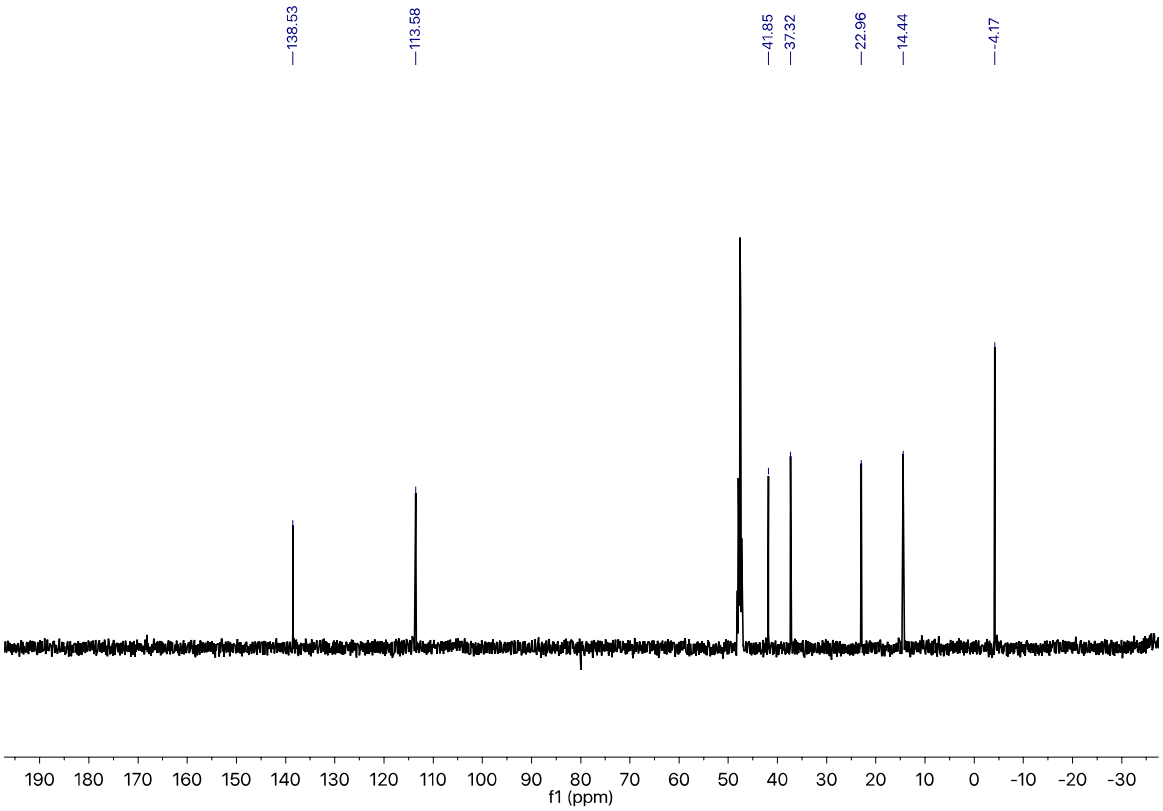


Figure S50. [13]C-NMR of compound **12** in MeOD.

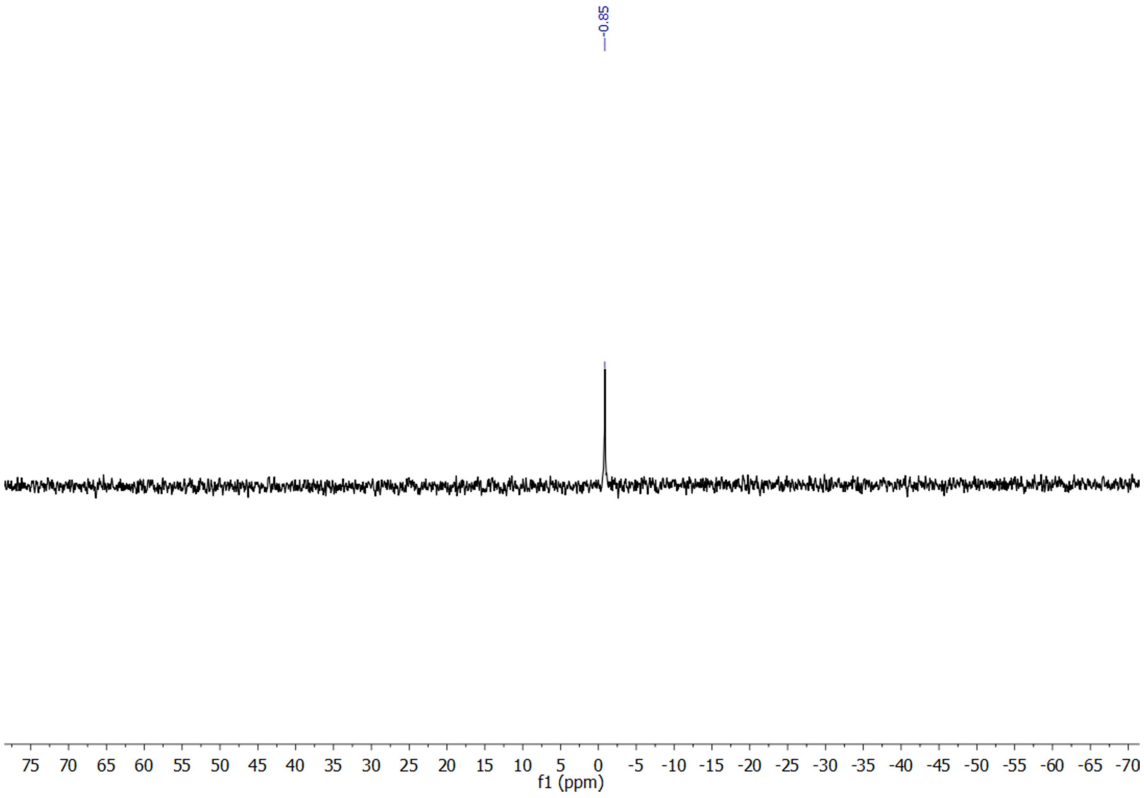


Figure S51. ^{29}Si-NMR of compound **12** in MeOD.

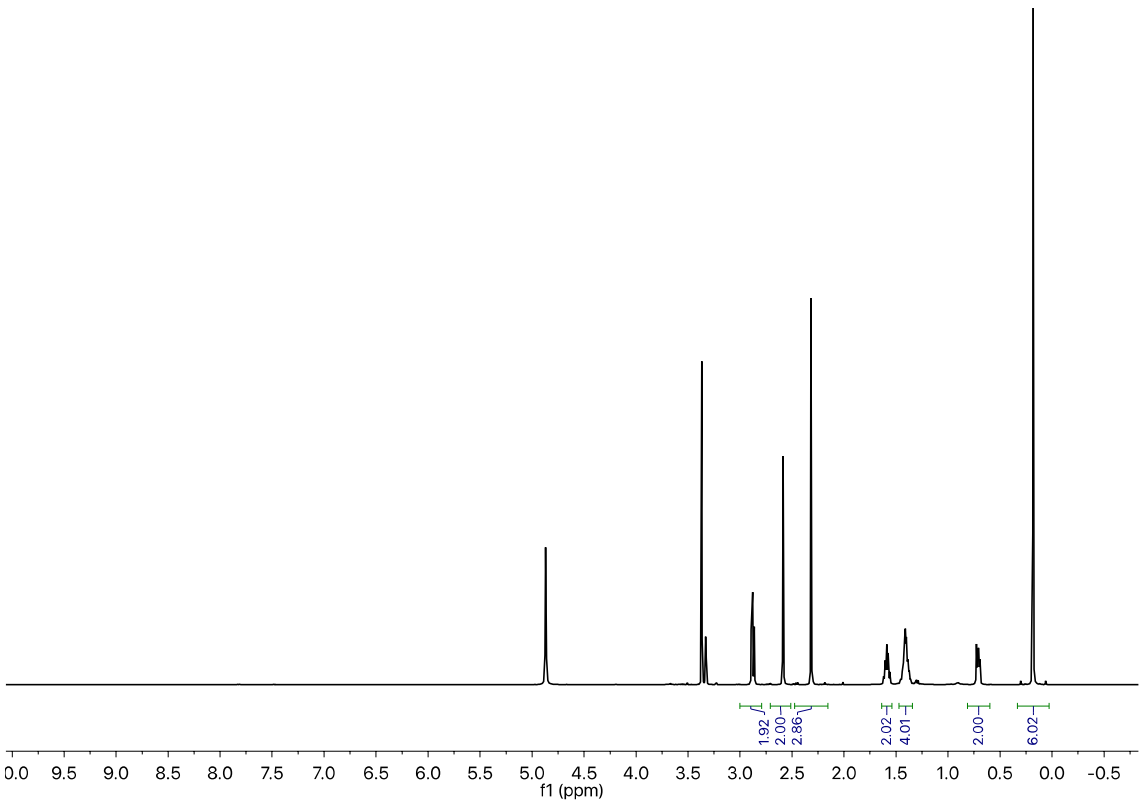


Figure S52. ^{1}H-NMR of compound **13** in MeOD.

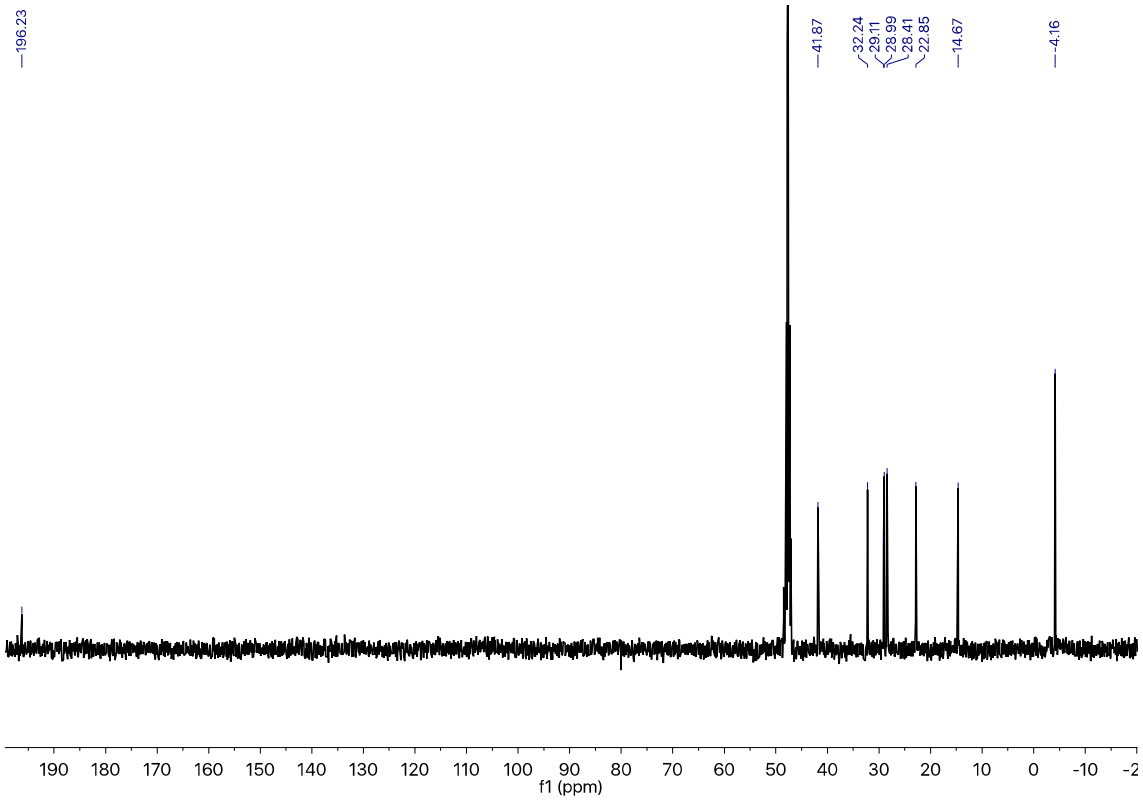


Figure S53. ^{13}C-NMR of compound **13** in MeOD.

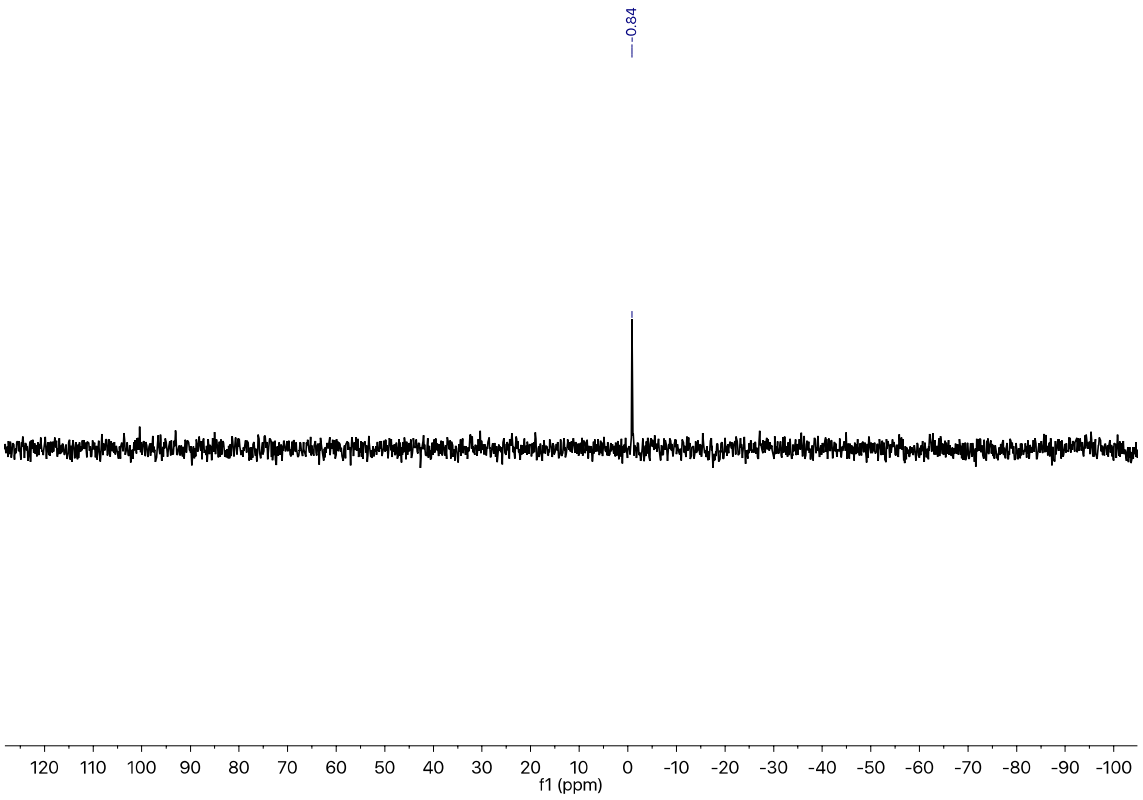


Figure S54. ^{29}Si-NMR of compound **13** in MeOD.

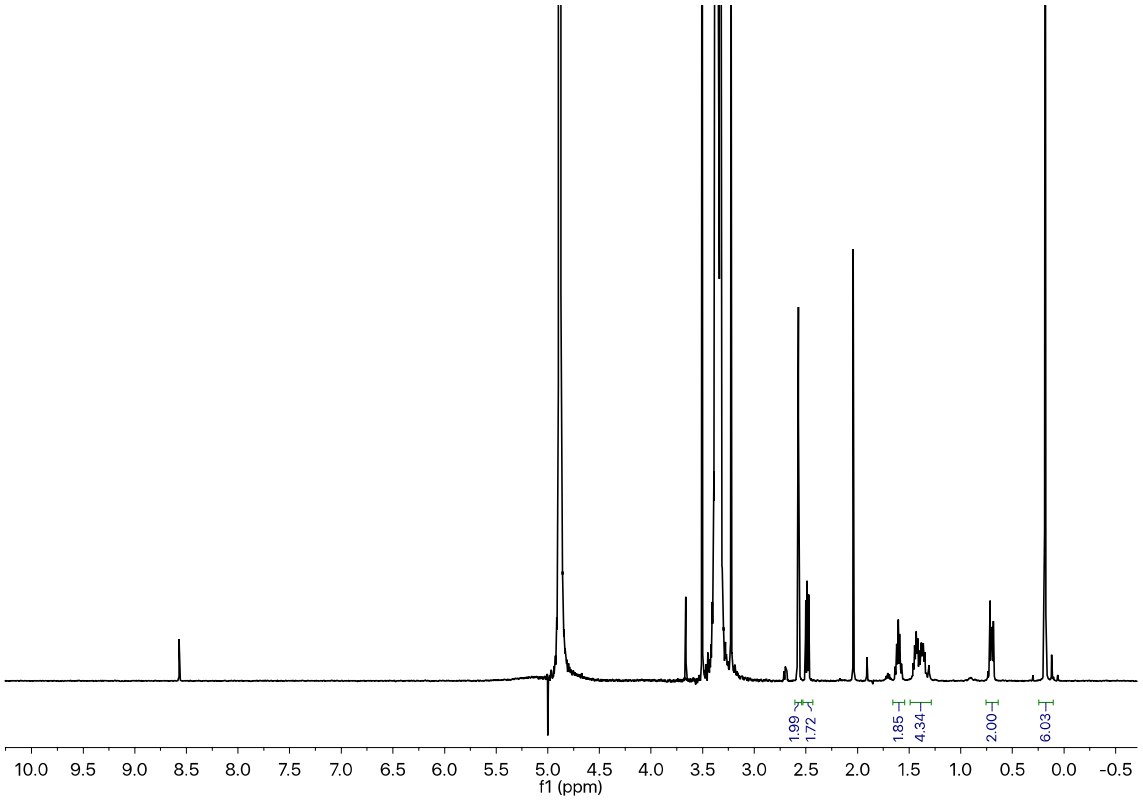


Figure S55. [1]H-NMR of compound **S4** in MeOD.

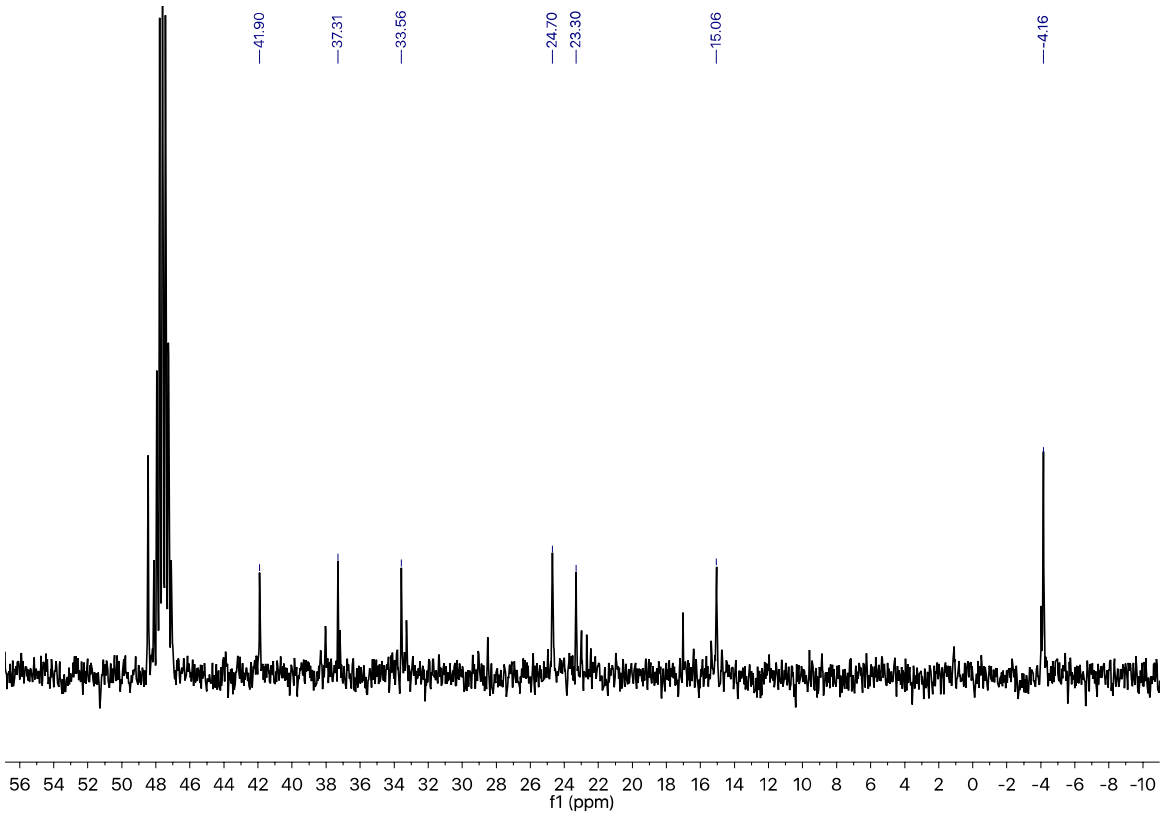


Figure S56. [13]C-NMR of compound **S4** in MeOD.

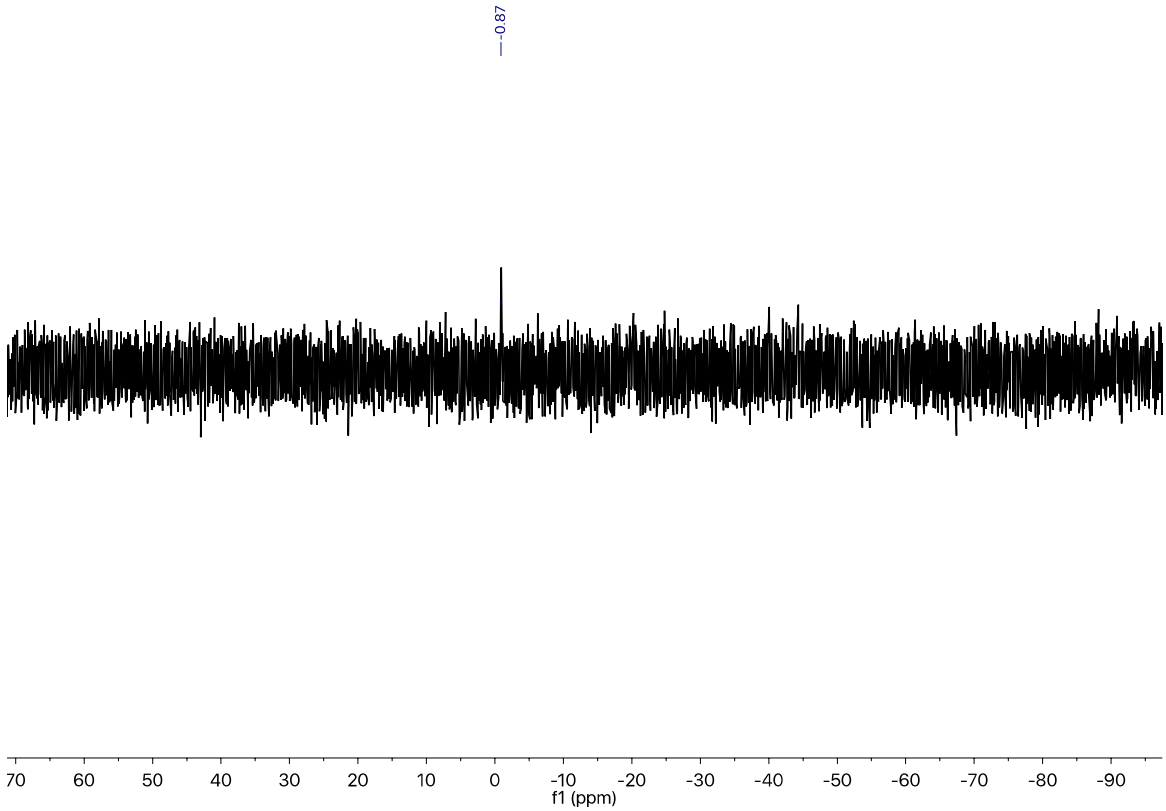


Figure S57. ^{29}Si-NMR of compound **S4** in MeOD.

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3.Salvia, M.-V.; Salassa, G.; Rastrelli, F.; Mancin, F.; *J. Am. Chem. Soc.,* **2015**, 137, 11399-11406.
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