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Supporting Information

Versatile Iron-Catechol based Nanoscale Coordination Polymers. Antiretroviral Ligand Functionalization and their use as Efficient Carriers in HIV/AIDS Therapy

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Materials and Methods

Materials. Solvents and starting materials were purchased from Sigma–Aldrich and used as received, without further purification, unless otherwise stated. 1,4-bis(imidazol-1-ylmethyl)benzene (bix) was synthesized according to previously reported methodology.[1]

Synthesis of 6-(3,4-bis(benzyloxy)phenyl)hex-5-enoic acid, **2**. To a 2 h stirred suspension of (4-carboxybutyl)triphenylphosphonium bromide (9.08 g, 20.5 mmol) and NaH (suspension 60% wt) (3.27 g, 81.7 mmol) in dry toluene (50 mL) under N_2 atmosphere, a solution of 3,4-bis(benzyloxy)benzaldehyde, **1**, (6.50 g, 20.4 mmol) in dry toluene (70 mL) was added dropwise. The mixture was then heated to reflux and stirred overnight. TLC analysis (EtOAc 100%) revealed the entire consumption of 3,4-bis(benzyloxy)benzaldehyde. The reaction crude was washed with water (70 mL and 2x30 mL), acidified with HCl (5%) to pH 3 and then extracted with EtOAc (4x60 mL). The combined organic phases were dried over anhydrous Na_2SO_4 and concentrated under vacuum. The resulting brownish wax was purified by column chromatography (CH_2Cl_2/EtOAc, 90:10 → 80:20 → 50:50) to afford a yellow solid identified as a mixture of isomers (5:1 *E*/*Z*) of carboxylic acids **2** (7.21 g, 17.93 mmol, 88% yield); R_f (EtOAc 100%) = 0.40. HRMS (EI) calcd for $[C_{26}H_{26}O_4]^+$ 402.1831, found 402.1829. mp. 70 – 72 °C. ^{1}H NMR of the major isomer E (400 MHz, $CDCl_3$) δ 7.49 – 7.41 (m, 4H, Ph), 7.39 – 7.29 (m, 6H, Ph), 6.98 (d, $^4J_{2',6'}$ = 1.7 Hz, 1H, H-2'), 6.86 – 6.84 (m, 2H, H-5', H-6'), 6.29 (d, $^3J_{6,5}$ = 15.8 Hz, 1H, H-6), 5.98 (dt, $^3J_{5,6}$ = 15.7 Hz, $^3J_{5,4}$ = 7.0 Hz, 1H, H-5), 5.16 (s, 2H, OCH_2-Ph), 5.14 (s, 2H, OCH_2-Ph), 2.40 (t, $^3J_{2,3}$ = 7.4 Hz, 2H, H-2), 2.24 (qd, $^3J_{4,3}$ = 6.7 Hz, $^3J_{4,5}$ = 6.7 Hz, 2H, H-4), 1.81 (quint, $^3J_{3,2}$ = 7.5 Hz, $^3J_{3,4}$ = 7.5 Hz, 2H, H-3). ^{13}C NMR of the major isomer *E* (101 MHz, $CDCl_3$) δ 178.20 (C-1), 148.47 (C-3'/C-4'), 147.71 (C-4'/C-3'), 136.70 (C-1'), 130.85 (Ph), 129.79 (C-6), 127.82 (C-5), 127.80 (Ph), 127.77 (Ph), 127.14 (Ph), 127.10 (Ph), 127.07 (Ph), 126.73 (Ph), 126.64 (Ph), 118.99 (C-5'/C-6'), 114.59 (C-6'/C-5'), 112.02 (C-2'), 70.78 (Ph-CH2-O), 32.51 (C-2), 31.49 (C-4), 23.68 (C-3).

Synthesis of 6-(3,4-dihydroxyphenyl)hexanoic acid, **3**. Pd/C (10% wt.) (0.03 g) was added to a solution of the (5:1 *E*/*Z*) mixture of olefins **2** (0.31 g, 0.76 mmol) in 15 mL EtOAc. H_2 (2 atm) was then introduced into the reaction vessel and the mixture was stirred at rt for 2 d, refilling H_2 pressure every day. After no presence of the benzyl signal was observed at ^{1}H NMR spectrum, the mixture was filtered through Celite® and the resulting brownish solid was purified by column chromatography (EtOAc 100%) to afford a yellow solid identified as the saturated carboxylic acid **3** (0.17 g, 0.74 mmol, 97% yield). R_f (EtOAc) = 0.41. HRMS (EI) calcd for $[C_{12}H_{16}O_4]^+$ 224.1049, found 224.1046. IR (ATR) ν 3424, 3180 (broad), 2923, 1707, 1290, 1245, 1196, 1171 cm^{-1}. mp. 86 – 88 °C. ^{1}H NMR (400 MHz, acetone-d6) δ 5.45 (d, $^3J_{5',6'}$ = 8.0 Hz, 1H, H-5'), 5.39 (s, 1H, H-2'), 5.25 (dd, $^3J_{6',5'}$ = 8.0 Hz, $^4J_{6',2'}$ = 2.2 Hz, 1H, H-6'), 1.21 (t, $^3J_{6,5}$ = 7.6 Hz, 2H, H-6), 1.03 (t, $^3J_{2,3}$ = 7.5 Hz, 2H, H-2), 0.43 – 0.25 (m,

4H, 2H-3, 2H-5), 0.15 – 0.00 (m, 2H, H-4). ^{13}C NMR (101 MHz, acetone-d6) δ 175.3 (C-1) 143.20 (C-3'), 141.28 (C-4'), 132.86 (C-1'), 118.04 (C-6'), 113.85 (C-2'), 113.56 (C-5'), 33.36 (C-6), 32.10 (C-2), 29.82 (C-3/C-5), 27.04 (C-4), 23.26 (C-5/C-3).

Synthesis of 6-(3,4-bis((*tert*-butyldiphenylsilyl)oxy)phenyl)hexanoic acid, **4**. DBU (0.11 mL, 0.74 mmol) was added dropwise to a stirred solution of catechol **3** (0.039 g, 0.17 mmol) and TBDPSCl (0.13 mL, 0.52 mmol) in dry ACN (1 mL) under N_2 atmosphere at rt. After 1 h, the reaction was heated to 40 °C and stirred overnight. Then, the solvent was evaporated, the crude was dissolved in CH_2Cl_2 and washed with NH_4Cl (~ 0.1 M) (3x5mL). The organic layers were combined and dried over anhydrous Na_2SO_4. Evaporation of the solvent furnished a brown waxy crude, which was purified by column chromatography (hexane/EtOAc 90:10 → 60:40) to afford **4** (0.079 g, 0.11 mmol, 64% yield) as a white solid. R_f (Hexane/EtOAc 60:40) = 0.31. HRMS (EI) calcd for $[C_{44}H_{52}O_4Si_2]^+$ 700.3404, found 700.3401. IR (ATR) ν 3071, 2930, 1705, 1513, 1129 cm^{-1}. mp. 50 – 53 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.85 – 7.78 (m, 8H, Ph), 7.48 – 7.35 (m, 12H, Ph), 6.34 (d, $^3J_{5',6'}$ = 8.2 Hz, 1H, H-5'), 6.21 (d, $^4J_{2',6'}$ = 2.2 Hz, 1H, H-2'), 6.15 (dd, $^3J_{6',5'}$ = 8.2 Hz, $^4J_{6',2'}$ = 2.2 Hz, 1H, H-6'), 2.17 (t, $^3J_{2,3}$ = 7.6 Hz, 2H, H-2), 2.04 (t, $^3J_{6,5}$ = 7.2 Hz, 2H, H-6), 1.41 (quint, $^3J_{4,3}$ = 7.4 Hz, $^3J_{4,5}$ = 7.4 Hz, 2H, H-4), 1.16 (s, 9H, *t*Bu), 1.15 (s, 9H, *t*Bu), 1.07 – 0.93 (m, 4H, 2H-3, 2H-5). ^{13}C NMR (101 MHz, $CDCl_3$) δ 179.56 (C-1), 146.01 (C-3'), 144.22 (C-4'), 135.99 (Ph), 135.97 (Ph), 134.88 (C-1'), 133.78 (Ph), 133.70 (Ph), 130.11 (Ph), 130.08 (Ph), 128.07 (Ph), 128.05 (Ph), 120.84 (C-2'), 120.73 (C-6'), 120.23 (C-5'), 34.76 (C-6), 34.06 (C-2), 30.58 (C-3), 28.48 (C-5), 27.13 ($C(CH_3)_3$), 27.09 ($C(CH_3)_3$), 24.71 (C-4), 19.86 ($C(CH_3)_3$).

Synthesis of ((2*S*,3*S*,5*R*)-3-azido-5-(5-methyl-2,4-dioxo-3,4-dihydropyrimidin-1(2*H*)-yl)tetrahydrofuran-2-yl)methyl 6-(3,4-bis((*tert*-butyldiphenylsilyl)oxy)phenyl)hexanoate, **5**. Carboxylic acid **4** (5.77 g, 8.23 mmol), HATU (4.27 g, 11.23 mmol) and DIPEA (5.2 mL, 29.94 mmol) were dissolved in dry THF (70 mL) under N_2 atmosphere and stirred at rt. After 45 min, a solution of **3** (2.00 g, 7.48 mmol) in dry THF (45 mL) was added dropwise. The mixture was stirred overnight at rt. Then, the mixture was filtered, the solvent was removed under vacuum and the crude was purified by column chromatography (CH_2Cl_2/EtOAc 80:20 to EtOAc 100%) to furnish the title compound **5** (6.09 g, 6.4 mmol, 86%) as a white solid. R_f (EtOAc) = 0.67. HRMS (EI) calcd for $[C_{54}H_{63}N_5O_7Si_2]^+$ 949.4266, found 949.4262. IR (ATR) ν 3049, 2932, 2859, 2101, 1703, 1512, 1128 cm^{-1}. $[\boldsymbol{\alpha}]_{\boldsymbol{D}}^{\boldsymbol{20}}$ = + 11.6 (*c* 1, $CHCl_3$). mp. 55 – 58 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.66 (s, 1H, H-3), 7.84 – 7.77 (m, 8H, Ph (TBDPS)), 7.47 – 7.34 (m, 12H, Ph(TBDPS)), 7.18 (q, $^4J_{6,CH3-C5}$ = 1.2 Hz, 1H, H-6), 6.33 (d, $^3J_{5''',6'''}$ = 8.1 Hz, 1H, H-5'''), 6.20 (d, $^3J_{2''',6'''}$ = 2.1 Hz, 1H, H-2'''), 6.14 (dd, $^3J_{6''',5'''}$ = 8.2 Hz, $^3J_{6''',2'''}$ = 2.2 Hz, 1H, H-6'''), 6.10 (t, $^3J_{1',2'}$ = 6.4 Hz, 1H, H-1'), 4.35 (dd, J_{gem} = 12.2 Hz, $^3J_{5',4'}$ = 4.7 Hz, 1H, H-5'), 4.27 (dd, J_{gem} = 12.2 Hz, $^3J_{5',4'}$ = 4.0 Hz, 1H, H-5'), 4.15 (dt, $^3J_{3',2'}$ = 7.6 Hz, $^3J_{3',4'}$ = 5.2 Hz, 1H, H-3'), 4.08 – 4.04 (m, 1H, H-4'), 2.47 (ddd, J_{gem} = 13.9 Hz, $^3J_{2',1'}$ = 6.4 Hz, $^3J_{2',3'}$ = 5.2 Hz, 1H, H-

2'), 2.32 (ddd, J_{gem} = 13.9 Hz, $^{3}J_{2',3'}$ = 7.6 Hz, $^{3}J_{2',1'}$ = 6.4 Hz, 1H, H-2'), 2.21 (t, $^{3}J_{6'',5''}$ = 7.7 Hz, 2H, H-6''), 2.04 (t, $^{3}J_{2'',3''}$= 7.2 Hz, 2H, H-2''), 1.90 (d, $^{4}J_{CH3\text{-}C5,6}$ = 1.2 Hz, 3H), 1.48 – 1.38 (m, 2H, H-3''), 1.15 (s, 9H, *t*Bu), 1.14 (s, 9H, *t*Bu), 1.09 – 0.94 (m, 4H, 2H-4'', 2H-5''). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.34 (C-1''), 163.67 (C-4), 150.20 (C-2), 146.02 (C-3'''), 144.27 (C-4'''), 135.97 (Ph (TBDPS)), 135.96 (Ph (TBDPS)), 135.51 (C-6), 134.72 (Ph (TBDPS)), 133.74 (C-1'''), 133.71(Ph (TBDPS)), 130.08 (Ph (TBDPS)), 130.06 (Ph (TBDPS)), 128.07 (Ph (TBDPS)), 128.06 (Ph (TBDPS)), 120.83 (C-2'''), 120.66 (C-6'''), 120.23 (C-5'''), 111.60 (C-5), 85.86 (C-1'), 82.13 (C-4'), 63.53 (C-5'), 61.04 (C-3'), 37.91 (C-2'), 34.75 (C-2''), 34.25 (C-6''), 30.60 (C-5''), 28.60 (C-4''), 27.11($(CH_3)_3$C (TBDPS), 24.88 (C-3''), 19.84 ($(CH_3)_3$C (TBDPS)), 12.96 (CH_3-C5).

Synthesis of ((2*S*,3*S*,5*R*)-3-azido-5-(5-methyl-2,4-dioxo-3,4-dihydropyrimidin-1(2*H*)-yl)tetrahydrofuran-2-yl)methyl 6-(3,4-dihydroxyphenyl)hexanoate, **CatAZT**. Triethylamine trihydrofluoride (2.1 mL, 12.9 mmol) was added to a stirred ice-cooled solution of **5** (1.97 g, 2.07 mmol) in dry THF (40 mL). The mixture was allowed to warm to rt and stirred overnight. The reaction was quenched with 0.3 mL of brine, diluted with 20 mL diethyl ether and filtered. The resulting crude was purified by column chromatography ($CHCl_3$/MeOH 97:3 → 94:6) to furnish **CatAZT** as a white solid (0.669 g, 1.41 mmol, 68%). R_f (EtOAc) = 0.55. HRMS (EI) calcd for $[C_{22}H_{27}N_5O_7]^+$ 473.1910, found 473.1915. $[\boldsymbol{\alpha}]_{\boldsymbol{D}}^{\boldsymbol{20}}$ = + 28.8 (*c* 1, $CHCl_3$). mp. 50 – 51 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.29 (d, $^{4}J_{6,CH3\text{-}C5}$ = 1.2 Hz, 1H, H-6), 6.76 (d, $^{3}J_{5''',6'''}$ = 8.1 Hz, 1H, H-5'''), 6.63 (d, $^{4}J_{2''',6'''}$ = 2.0 Hz, 1H, H-2'''), 6.55 (dd, $^{3}J_{6''',5'''}$ = 8.1 Hz, $^{3}J_{6''',2'''}$ = 2.0 Hz, 1H, H-6'''), 6.06 (t, $^{3}J_{1',2'}$ = 6.2 Hz, 1H, H-1'), 4.40 (dd, J_{gem} = 12.4 Hz, $^{3}J_{5',4'}$ = 3.6 Hz, 1H, H-5'), 4.31 (dd, J_{gem} = 12.4 Hz, $^{3}J_{5',4'}$ = 3.6 Hz, 1H, H-5'), 4.13 (dt, $^{3}J_{3',2'}$ = 7.4 Hz, $^{3}J_{3',4'}$ = 5.4 Hz, 1H, H-3'), 4.07 (dt, $^{3}J_{4',3'}$ = 5.4 Hz, $^{3}J_{4',5'}$ = 3.6 Hz, 1H, H-4'), 2.51 – 2.44 (m, 3H, 2H-6'', H-2'), 2.38 – 2.30 (m, 3H, 2H-2'', H-2'), 1.89 (d, $^{4}J_{CH3\text{-}C5,6}$ = 1.3 Hz, 3H, CH3-C5), 1.70 – 1.61 (m, 2H, H-3''), 1.59 – 1.51 (m, 2H, H-5''), 1.35 – 1.27 (m, 2H, H-4''). ^{13}C NMR (101 MHz, $CDCl_3$) δ 172.39 (C-1''), 163.31 (C-4), 149.25 (C-2), 142.97 (C-3'''), 141.20 (C-4'''), 135.12 (C-6), 134.35 (C-1'''), 119.99 (C-6'''), 114.68 (C-5'''), 114.66 (C-2'''), 110.26 (C-5), 85.25 (C-1'), 81.49 (C-4'), 62.18 (C-5'), 59.66 (C-3'), 37.18 (C-2'), 34.08 (C-6''), 33.38 (C-2''), 30.17 (C-5''), 27.54 (C-4''), 24.04 (C-3''), 11.96 (CH3-C5).

Synthesis of ((2*R*,3*S*,5*R*)-3-hydroxy-5-(5-methyl-2,4-dioxo-3,4-dihydropyrimidin-1(2*H*)-yl)tetrahydrofuran-2-yl)methyl 6-(3,4-bis((tert-butyldiphenylsilyl)oxy)phenyl)hexanoate, **6**. Diisopropyl azodicarboxylate (DIAD) (0.15 mL, 0.57 mmol) was added dropwise to an ice-cooled solution of carboxylic acid **4** (0.200 g, 0.29 mmol), triphenylphosphine (0.112 g, 0.43 mmol) and thymidine (0.090 g, 0.37 mmol) in a mixture of anhydrous THF/DMF (3 and 0.8 mL, respectively). The mixture was allowed to warm to rt and stirred overnight. The solvent was evaporated under vacuum and the crude was purified by column chromatography (EtOAc 100%) to afford **6** as a white solid (0.193 g, 0.21 mmol,

56%). R_f (EtOAc) = 0.39. HRMS (ESI+) calcd for $[C_{54}H_{65}N_2O_8Si_2]^+$ 925.4274, found 925.4263. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.41 (s, 1H, H-3), 7.84 – 7.77 (m, 8H, Ph (TBDPS)), 7.47 – 7.34 (m, 12H, Ph(TBDPS)), 7.23 (q, $^4J_{6,CH3\text{-}C5}$ = 0.9 Hz, 1H, H-6), 6.33 (d, $^3J_{5''',6'''}$ = 8.2 Hz, 1H, H-5'''), 6.27 (t, $^3J_{1',2'}$ = 6.7 Hz, 1H, H-1'), 6.20 (d, $^3J_{2''',6'''}$ = 2.1 Hz, 1H, H-2'''), 6.13 (dd, $^3J_{6''',5'''}$ = 8.2 Hz, $^3J_{6''',2'''}$ = 2.2 Hz, 1H, H-6'''), 4.36 (dd, J_{gem} = 12.2 Hz, $^3J_{5',4'}$ = 4.5 Hz, 1H, H-5'), 4.31 (dt, $^3J_{3',2'}$ = 6.8 Hz, $^3J_{3',4'}$ = 3.8 Hz, 1H, H-3'), 4.23 (dd, J_{gem} = 12.2 Hz, $^3J_{5',4'}$ = 3.8 Hz, 1H, H-5'), 4.11 (dt, $^3J_{4',5'}$ = 4.5 Hz, $^3J_{4',3'}$ = 3.8 Hz, 1H, H-4'), 2.38 (ddd, J_{gem} = 13.8 Hz, $^3J_{2',1'}$ = 6.7 Hz, $^3J_{2',3'}$ = 3.8 Hz, 1H, H-2'), 2.21 (t, $^3J_{6'',5''}$ = 7.5 Hz, 2H, H-6''), 2.09 (ddd, J_{gem} = 13.8 Hz, $^3J_{2',1'}$ = $^3J_{2',3'}$ = 6.8 Hz, 1H, H-2'), 2.03 (t, $^3J_{2'',3''}$= 7.5 Hz, 2H, H-2''), 1.89 (d, $^4J_{CH3\text{-}C5,6}$ = 1.2 Hz, 3H), 1.43 (quint, $^3J_{3'',2''}$ = $^3J_{3'',4''}$ = 7.5 Hz, 2H, H-3''), 1.15 (s, 9H, *t*Bu), 1.14 (s, 9H, *t*Bu), 1.05 – 0.93 (m, 4H, 2H-4'', 2H-5''). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.52 (C-1''), 163.40 (C-4), 150.11 (C-2), 145.81 (C-3'''), 144.06 (C-4'''), 135.77 (Ph (TBDPS)), 135.66 (Ph (TBDPS)), 135.51 (C-6), 135.02 (Ph (TBDPS)), 133.48 (C-1'''), 129.90 (Ph (TBDPS)), 129.71 (Ph (TBDPS)), 129.51 (Ph (TBDPS)), 127.86 (Ph (TBDPS)), 127.76 (Ph (TBDPS)), 120.64 (C-2'''), 120.50 (C-6'''), 120.02 (C-5'''), 111.26 (C-5), 85.22 (C-1'), 84.34 (C-4'), 71.76 (C-5'), 63.69 (C-3'), 40.52 (C-2'), 34.54 (C-2''), 34.13 (C-6''), 30.37 (C-5''), 28.36 (C-4''), 26.88($(CH_3)_3$C (TBDPS), 24.72 (C-3''), 19.65 ($(CH_3)_3$C (TBDPS)), 12.80 (CH_3-C5).

Synthesis of ((2*S*,3*S*,5*R*)-3-hydroxy-5-(5-methyl-2,4-dioxo-3,4-dihydropyrimidin-1(2*H*)-yl)tetrahydrofuran-2-yl)methyl 6-(3,4-dihydroxyphenyl)hexanoate, CatTHY. Triethylamine trihydrofluoride (0.45 mL, 2.75 mmol) was added to a stirred ice-cooled solution of **6** (0.490 g, 0.53 mmol) in dry THF (15 mL). The mixture was allowed to warm to rt and stirred overnight. The reaction was quenched with 0.15 mL of brine, diluted with 10 mL diethyl ether and filtered. The resulting crude was purified by column chromatography ($CHCl_3$/MeOH 95:5) to furnish **CatTHY** as a white solid (0.162 g, 0.36 mmol, 68%). R_f (EtOAc) = 0.09. HRMS (EI) calcd for $[C_{22}H_{29}N_2O_8]^+$ 449.1919; found 449.1918. $[\alpha]_D^{20}$ = + 5.1 (*c* 1, DMSO). mp. 72 – 75 °C. ^{1}H NMR (400 MHz, CD_3OD) δ 7.48 (d, $^4J_{6,CH3\text{-}C5}$ = 1.3 Hz, 1H, H-6), 6.64 (d, $^3J_{5''',6'''}$ = 8.0 Hz, 1H, H-5'''), 6.59 (d, $^4J_{2''',6'''}$ = 2.1 Hz, 1H, H-2'''), 6.46 (dd, $^3J_{6''',5'''}$ = 8.0 Hz, $^3J_{6''',2'''}$ = 2.1 Hz, 1H, H-6'''), 6.25 (t, $^3J_{1',2'}$ = 7.0 Hz, 1H, H-1'), 4.35 (dd, J_{gem} = 12.1 Hz, $^3J_{5',4'}$ = 4.4 Hz, 1H, H-5'), 4.33 (m, 1H, H-4'), 4.24 (dd, J_{gem} = 12.1 Hz, $^3J_{5',4'}$ = 3.5 Hz, 1H, H-5'), 4.06 (dt, $^3J_{3',2'}$ = 5.4 Hz, $^3J_{3',4'}$ = $^3J_{3',2'}$ = 3.5 Hz, 1H, H-3'), 2.44 (t, $^3J_{2'',3''}$ = 7.5 Hz, 2H, H-2''), 2.36 (td, $^3J_{6'',3''}$ = 7.3 Hz, $^3J_{6'',6'''}$ = 1.2 Hz, 2H, H-6''), 2.30 (ddd, J_{gem}= 13.8 Hz, $^3J_{2',1'}$ = 7.0 Hz, $^3J_{2',3'}$ = 3.5 Hz, 1H, H-2'), 2.21 (ddd, J_{gem}= 13.8 Hz, $^3J_{2',1'}$ = 7.0 Hz, $^3J_{2',3'}$ = 5.4 Hz, 1H, H-2') 1.88 (d, $^4J_{CH3\text{-}C5,6}$ = 1.2 Hz, 3H, CH_3-C5), 1.64 (quint, $^3J_{3'',2''}$ = $^3J_{3'',4''}$ = 7.2 Hz, 2H, H-3''), 1.56 (quint, $^3J_{5'',4''}$ = $^3J_{5'',6''}$ = 7.2 Hz, 2H, H-5''), 1.38 – 1.28 (m, 2H, H-4''). ^{13}C NMR (101 MHz, CD_3OD) δ 174.92 (C-1''), 166.24 (C-4), 152.15 (C-2), 145.91 (C-3'''), 144.00 (C-4'''), 137.49 (C-6), 135.33 (C-1'''), 120.59 (C-6'''), 116.45 (C-5'''), 114.18 (C-2'''), 111.66 (C-5), 86.52 (C-1'), 85.72 (C-4'), 72.26 (C-5'), 64.89 (C-3'), 40.61 (C-2'), 35.96 (C-6''), 34.90 (C-2''), 32.38 (C-5''), 29.58 (C-4''), 25.81 (C-3''), 12.62 (CH_3-C5).

Characterization Methods. 250 MHz ^{1}H NMR spectra were recorded on a Bruker DPX 250 MHz spectrometer; 400 MHz ^{1}H NMR, ^{1}H-^{1}H COSY, ^{1}H-^{13}C HSQC, ^{1}H-^{13}C HMBC, DEPT135 and 100 MHz ^{13}C NMR spectra were recorded on a Bruker DPX 400 MHz spectrometer. High-resolution mass spectra were obtained by direct injection of the sample with electrospray techniques in a Bruker microTOF-Q instrument. SEM images were performed on a scanning electron microscope (FEI Quanta 650 FEG) at acceleration voltages of 5–20 kV. The samples were prepared by drop casting of the corresponding dispersion on aluminum tape followed by evaporation of the solvent under room conditions. Before analysis, the samples were metalized with a thin layer of gold by using a sputter coater (Emitech K550). IR spectra were recorded by using a Tensor 27 (Bruker) spectrophotometer equipped with a single-reflection diamond window ATR accessory (MKII Golden Gate, Specac). Size distribution and surface charge of the nanoparticles were measured by DLS, using a ZetasizerNano 3600 instrument (Malvern Instruments, UK), the size range limit of which is 0.6 nm to 6 mm. Note: the diameter measured by DLS is the hydrodynamic diameter. The samples were comprised of aqueous dispersions of the nanoparticles in distilled water or in buffer. All samples were diluted to obtain an adequate nanoparticle concentration. Powder XRD spectra were recorded at room temperature on a high-resolution texture diffractometer (PANalyticalX'Pert PRO MRD) equipped with a CoKα radiation source (λ = 1.7903 Å) and operating in reflection mode. The solid samples were placed in an amorphous silicon oxide flat plate and measured directly.

MRI experiments. The longitudinal r1 and transverse r2 relaxation rates for different concentrations of catAZT-NCPs were measured in solution under an external magnetic field of 7 Teslas (Bruker Biospec7T) in two phantom sequences. The nanoprobes were dispersed in PBS/agarose 1% solutions to ensure a good colloidal stability, resulting in a series with different metal concentrations ranging from 1 mg/mL to 25 mg/mL. The obtained relaxation rate values were plotted versus the concentrations of iron.

Synthesis of catAZT-NCPs. To prepare a material suitable for biological experiments, synthesis of the nanoparticles was performed inside a biosafety cabinet (Telstar BioVanguard B Green) and all the material and solvents used were sterilized. 1,4-Bis(imidazole-1-ylmethyl)-benzene (Bix) (7.8 mg, 0.032 mmol), CatAZT prodrug (30.5 mg, 0.064 mmol) and polyvinylpyrrolidone (PVP) (average MW 40000) (19.5 mg) were dissolved in ethanol (17.2 mL). Under stirring (700 rpm), a solution of $Fe(CH_3COO)_2$ (5.45 mg, 0.032 mmol in 2.3 mL ethanol) was added dropwise. Instantaneously, a dark-purple precipitate appeared. After the reaction mixture was stirred at rt for 5 min, the precipitate was collected by centrifugation and then washed with ethanol four times. Finally, the solid was irradiated with a UV lamp for 15 min, and the nanoparticles stored as a solid or in the fridge at a concentration of 44 mg/ml in ethanol. SEM images of the resulting material showed spherical nanoparticles with a size distribution of 147 ± 33 nm measured in ethanol. Elemental analysis: found (%)

C 51.76, H 5.38, N 13.79. Calculated empirical formula: $FeC_{43.9}H_{54.3}O_{15.0}N_{10.0}$. ICP-MS: 5.46% Fe.

HPLC methodology for ARV-NCPs analysis. Chromatographic conditions: Analyses were performed using a HPLC Waters 2695 separation module coupled to a Waters 2487 UV-Vis detector (suitable for dual detection). The column used was a Chromolith® Performance RP-18e (100 mm x 4.6 mm). Eluent A was a 0.1% (v/v) H_3PO_4 aqueous solution containing 262 mg/L sodium 1-octanesulfonate and eluent B was methanol absolute (HPLC grade). Before the analysis, the RP column was pre-equilibrated using the starting conditions of the method (99 % A (v/v)) for 6 min. The elution began with an isocratic elution of 99% A (v/v) for 5 min, followed by a gradual increase of A from 1% to 40% (v/v) until 25 min. Then, the mobile phase was raised to 98% B (v/v) (between minutes 25 and 30) to elute tight bound compounds and kept at 98% B (v/v) for additional 5 min. Finally, mobile phase was reset to the initial conditions (A:B) 99:1 (v/v) and stayed for 6 min to equilibrate for the next injection. The flow rate was set at 1.0 mL/min and the column temperature was kept at 25 °C. The detection wavelengths were 214 and 280 nm. Sample preparation: NCPs samples were prepared dissolving 1.5 mg NCPs/mL in 0.15 mL of a methanol/HCl mixture (50 µL concentrated HCl/mL methanol). The initial samples were diluted with 1.35 mL of deionized water to have a final water/methanol ratio of 90:10 and sonicated for 5 min. Then, samples were further diluted 3 and 5 times in buffer A before their injection into the HPLC system. All samples were prepared in duplicate. Calibration curves: A calibration curve using 1,4-bis(imidazol-1ylmethyl)benzene (Bix) and ((2*S*,3*S*,5*R*)-3-azido-5-(5-methyl-2,4-dioxo-3,4-dihydropyrimidin-1(2*H*)-yl)tetrahydrofuran-2-yl)methyl 6-(3,4-dihydroxyphenyl)hexanoate (catAZT) as external standards was prepared. Standards were prepared by duplicate, diluting a stock solution containing Bix and catAZT (960 and 980 µg/mL, respectively) dissolved in a methanol/HCl mixture (50 µL concentrated HCl/mL methanol) and diluted with distilled water to a final water/methanol ratio of 90:10. In both cases, results were adjusted to linear regression models with $R^2 > 0.999$ between the ranges of 9-261, 8-220 µg/mL for Bix and catAZT, respectively.

In vitro drug release studies at different pH and in presence/absence of esterases. CatAZT (5 mg) or catAZT-NCPs (6 mg) were added to a PBS/BSA 0.5 mM buffer solution at pH 5.1 or 7.4 (20 mL) with and without pig liver esterases (PLE) (180 U/L). All the samples were maintained at 37 ˚C under constant stirring. Aliquots (400 µL) were taken after different periods of time, and the volume extracted was replaced with additional 400 µL of the PBS/BSA 0.5 mM solution. Then, all the aliquots were filtered through a 10 kDa membrane (Amicon® Ultra 0.5 mL) (15 min x 14.5k RFC) before their injection in the HPLC system. When the last aliquot was taken, three additional aliquots (390 µL) were extracted and treated with HCl 2 M in methanol (10 µL) to measure the remaining amount of non-released drug.

HPLC method for release kinetics quantification. Chromatographic conditions: Analyses were performed using a HPLC Waters 2695 separation module coupled to a Waters 2487 UV-Vis detector (suitable for dual detection). The column used was a Restek Ultra C18 (250 mm x 4.6 mm). Eluent A was a 0.1% (v/v) H_3PO_4 aqueous solution and eluent B was acetonitrile (HPLC grade). Injection volume was 20 µL. Before the analysis, the RP column was pre-equilibrated using the starting conditions of the method (80 % A (v/v)) for 30 min. Initial flow rate was set at 0.8 mL/min. The elution began with a gradual increase of B from 20% to 60% (v/v) until 8 min. Then, an isocratic elution was maintained for 5.5 min. A second gradient was then performed, raising eluent B from 60% to 80% in 1 min and flow rate to 1.2 mL/min and maintained for an additional 5.5 min. Finally, mobile phase composition and flow rate was reset to the initial conditions (A:B) 80:20 (v/v) and 0.8 mL/min and stayed for 4.5 min to equilibrate for the next injection. Column temperature was kept at 25˚C and the detection wavelengths were 214 and 280 nm. Calibration curves: A calibration curve using zidovudine (AZT) and ((2*S*,3*S*,5*R*)-3-azido-5-(5-methyl-2,4-dioxo-3,4-dihydropyrimidin-1(2*H*)-yl)tetrahydrofuran-2-yl)methyl 6-(3,4-dihydroxyphenyl)hexanoate (catAZT) as external standards was prepared. Standards were prepared by duplicate, diluting a stock solution containing AZT and catAZT (3 mg/mL) dissolved in methanol and diluted with distilled water to a final water/methanol ratio of 80:20. In both cases, results were adjusted to linear regression models with $R^2 > 0.99$ between the ranges of 1-480 and 4-208 µg/mL for AZT and catAZT, respectively.

Cell culture. $CD4^+$ T cells were isolated from the PBMCs of healthy human donors by FACS-sorting using the rapid expansion method (REM). This cell line was gently donated by Prof. Dr. Mercè Martí (Molecular Immunology Research Group; Department of Cell Biology, Physiology and Immunology (IBB/UAB)). IL-2 was added at day 1 and cells remained untouched during the first 5 days of culture, and then cells were split every 3–4 days. Human T cell leukemia virus carrier MT-2 cell line was obtained from the American Type Culture Collection (ATCC, Manassas, VA, USA) provided by NIH AIDS Reagent Program. Cells were routinely cultured in cultured in RPMI 1640 medium with L-Glutamine (Lonza, Verviers, Belgium, #12-702 F), further supplemented with 10% heat-inactivated fetal bovine serum (FBS, Gibco, #10270-106), 100 U/ml penicillin, 100 mg/ml streptomycin (Penicillin-Streptomycin, S/P; Lonza, Verviers, Belgium). Cells were routinely maintained in a humidified atmosphere at 37 ºC with 5% CO2 in the biohazard P3 laboratory. Cells were maintained at $0.3x10^6$ cells/ml.

Generation of virus stock. NL4-3 HIV-1 virus was produced by transient transfection in HEK293-T cells. Briefly, the previous day $1.5x10^6$ HEK293-T cells were seeded in 75 cm2 tissue culture flasks in DMEM medium with L-Glutamine, supplemented with 10% heat-inactivated fetal bovine serum, 100 U/ml penicillin, and 100 mg/ml streptomycin (DMEM-10). Cells were replaced with fresh DMEM-10 medium three hours before transfection by the calcium-phosphate method (ProFectionH Mammalian

Transfection System; Promega, Madison, WI) according to the manufacturer's instructions, using 5 mg of pNL4-3 DNA previously purified (Qiagen, Valencia, CA).[2] Growth medium was replaced with fresh DMEM-10 medium 16–18 h post-transfection. The supernatants were harvested approximately two days after transfection, clarified by centrifugation at 800 g/4ºC for 10 minutes, aliquoted and stored at -80ºC. Viruses were quantified by determining the concentration of p24 in the supernatant with an antigen capture assay (ELISA; INNOTEST® HIV Antigen mAb test, Fugirebio, Gent, Belgium). P24 ELISA should be sufficient to determine the level of viral production (at a protein level). Viral DNA would be a step further but notice that the viral HIV genome gets integrated into the cells genome and our delivery system does not avoid infection. The level of replication-competent infectious viruses was evaluated through the tissue culture infective dose (TCID50) in TZM-bI indicator cells. The viruses were titred in vitro for their cytopathic effect on MT-2 cells with the MTT assay.

Cytotoxicity assays. Cytotoxicity of catAZT-NPs was tested against primary human CD4+ T lymphocytes by using the PrestoBlue cell viability assay (Invitrogen). Primary CD4+T cells previously isolated by the rapid expansion method (REM) were seeded in a 96-well plate at 1×10^5 cells/well, then 100 µL medium per well containing various concentrations of catAZT-NPs and AZT were added (500, 250, 125, 50 and 10 µg/mL) and incubated for 24h. Cytotoxicity was evaluated adding 20 µL of prestoBlue reagent per well. After incubation at 37°C for 4 h, fluorescence intensities were measured at a 532-nm excitation wavelength and a 571-nm emission wavelength in a microplate reader (Multilabel Processor Victor™X3 Perkin Elmer, USA). Cell cytotoxicity of the antiviral effect of the free compounds (AZT and catAZT) and catAZT-NCPs nanoparticles against MT-2 cells was evaluated by the MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay. To perform the experiments, exponentially growing MT-2 cell were plated in 96-well plates at a density of $0.4x10^5$ cells per well. The cells were treated with the vehicle (saccharose), a range of concentrations of each compound (10 different concentrations from 0 to 100 µg/mL), or with the nanoparticles at equivalent concentrations to those assayed for AZT/catAZT. After incubation at the indicated time points at 37 ºC under a humidified atmosphere with 5% CO_2, 100 µl of the cell media were carefully discarded and 10 µl of MTT solution were added to each well and incubated for 4h at 37 ºC. Then, 100 µl of solubilization solution was added to each well containing the MTT, and then plates were incubated at 37 ºC under 5% CO_2 for ON. After incubation, the colour formed on each well was measured using a spectrophotometric plate reader (BioTek, Synergy HT) at 620 nm wavelength. All the cytotoxicity experiments were performed in triplicate, and at least two independent assays. Cell cytotoxicity was evaluated in terms of cell grown inhibition in treated cultures, and expressed as % of the control condition.

HIV-1 in vitro antiviral assay. The antiviral effects of AZT, catAZT and catAZT-NCPs nanoparticles on MT-2 cells infected with HIV-1 were evaluated by the soluble-

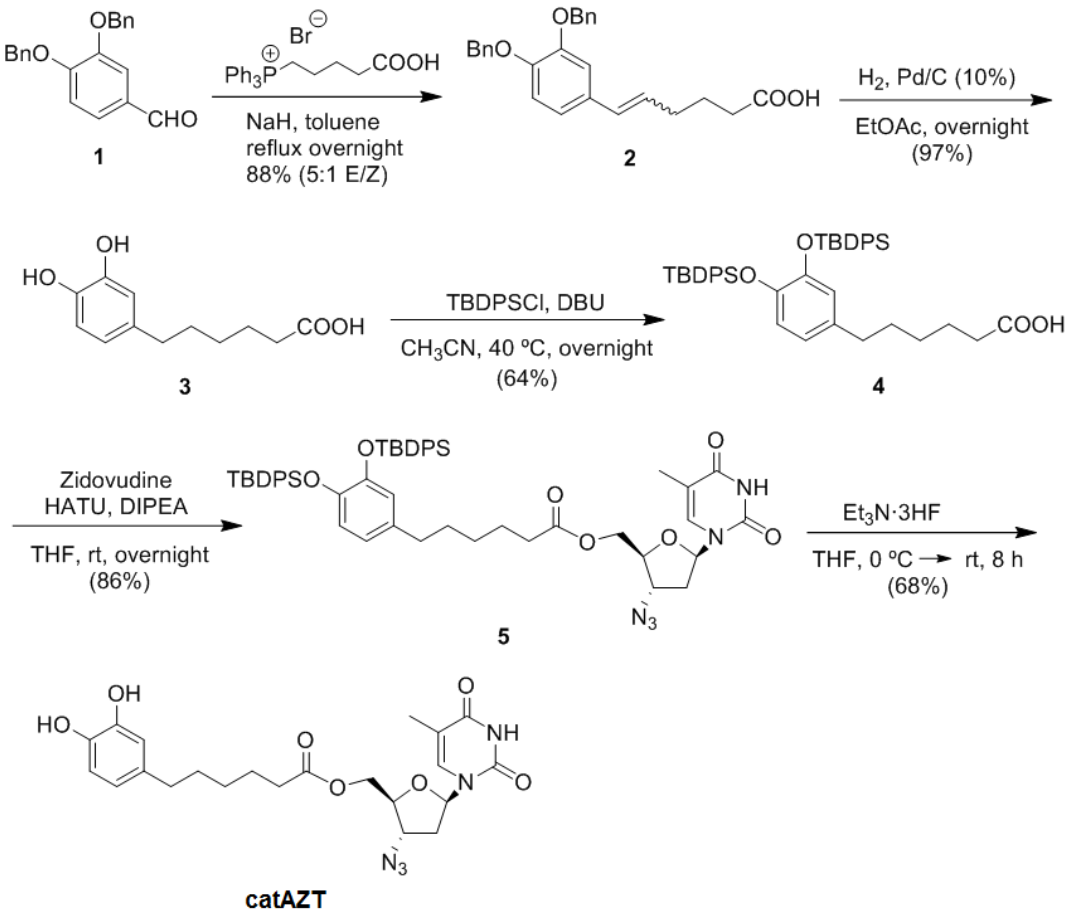
formazan method in order to follow the cytopathic effect of the infection, similarly as previously described.[3,4] Briefly, MT-2 cells were plated at a concentration of $0.4x10^5$ cells/ml in 96-well plates. Cells were then treated with different concentrations of vehicle (saccharose), AZT, catAZT or catAZT-NCPs and control nanoparticles containing thymidine (catTHY-NCPs). Following the addition of these compounds, cells were infected with CXCR4 tropic NL4.3 HIV-1 viruses (TCID50/ml=129000; p24=1.2 ug/ml). For the 7-day incubation experiments the infection was performed at MOI=0.002. For the 3-day assay, the pure stock of virus was used (100 ul) and diluted 1:2 in the well. Infected cells were incubated for 3 and 7 days in a humidified atmosphere at 37ºC with 5% CO2 in the biohazard P3 laboratory. After incubation, the HIV-1 cytopathic effect in the cells was determined by the MTT method under identical conditions as described above for the cytotoxicity experiments.

Uptake experiments. MT-2 cells were seeded at a concentration of $1.0x10^6$ cells/ml. The cells were incubated with AZT, catAZT or catAZT-NCPs at the indicated concentrations (referred to the AZT concentration) for 4 h at 37 ºC under a humidified atmosphere with 5% CO_2. Saccharose was used as vehicle control. Immediately after incubation, cells were washed with PBS and lysed in 1000 µl of a water/methanol (80%/20%) solution containing 0.1% phosphoric acid (pH=2.5) to determined intracellular free AZT. In the case of NCPs, the samples were lysed in 100 µl of water/methanol solution containing 1.85 % (v/v) HCl. The concentration of AZT in all the samples was determined by HPLC-UV as following described. The intracellular concentration of AZT present in the cells (expressed as nmols/10^6 cells) was calculated dividing the concentration of AZT by the number of cells present in each sample. The experiment was performed per duplicate and at least two independent assays.

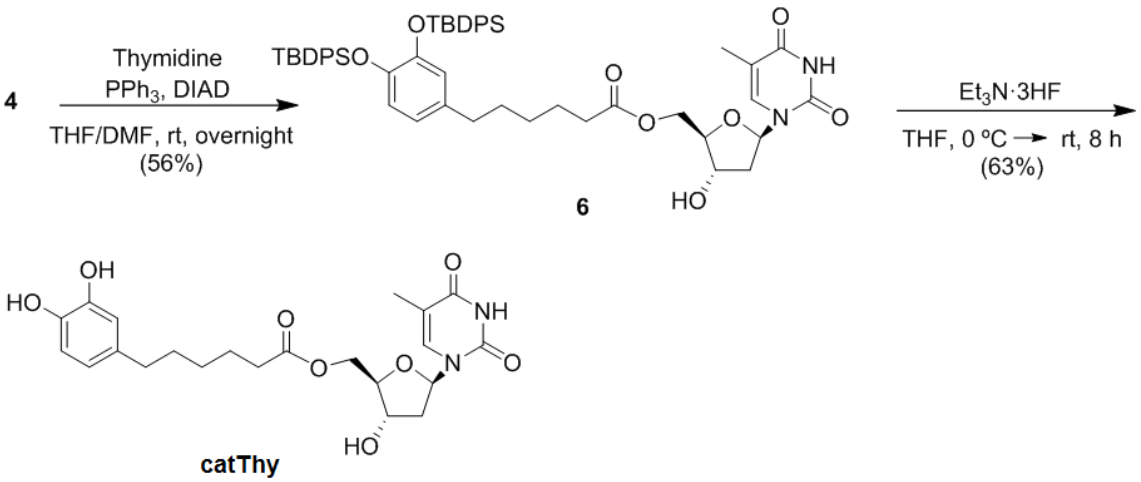
HPLC methodology for AZT uptake quantification. Chromatographic conditions: Analyses were performed using a HPLC Waters 2695 separation module coupled to a Waters 2487 UV-Vis detector (suitable for dual detection). The column used was a Restek® C-18 (250 mm x 4.6 mm). Eluent A was a 0.1% (v/v) H_3PO_4 aqueous solution and eluent B was methanol absolute (HPLC grade). Injection volume was 20 µL. Before the analysis, the RP column was pre-equilibrated using the starting conditions of the method (80 % A (v/v)) for 30 min. The elution began with a gradual increase of B from 20% to 60% (v/v) for 8 min, followed by isocratic elution (60% B) for 7 min. Then, the mobile phase was raised to 80% B (v/v) for 1 min to elute tight bound compounds. Finally, mobile phase was reset to the initial conditions (A:B) 80:20 (v/v) for 4 min and stayed for 5 min to equilibrate for the next injection. The flow rate was set at 0.4 mL/min and the column temperature was kept at 25 °C. The detection wavelengths were 214 and 280 nm. Calibration curves: A calibration curve using AZT and CatAZT as external standards was prepared. Standards were prepared diluting a stock solution containing AZT and CatAZT (2 mM) dissolved in methanol and diluted with distilled water to a final water/methanol ratio of 50:50. In both cases,

results were adjusted to linear regression models with $R2 > 0.999$ between the ranges of 10-100 and 40-250 μM for AZT and CatAZT, respectively.

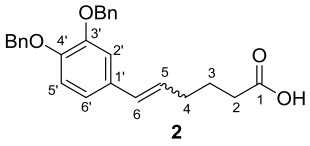
S1. Synthesis and characterization or CatAZT and CatTHY



Scheme 1



Scheme 2



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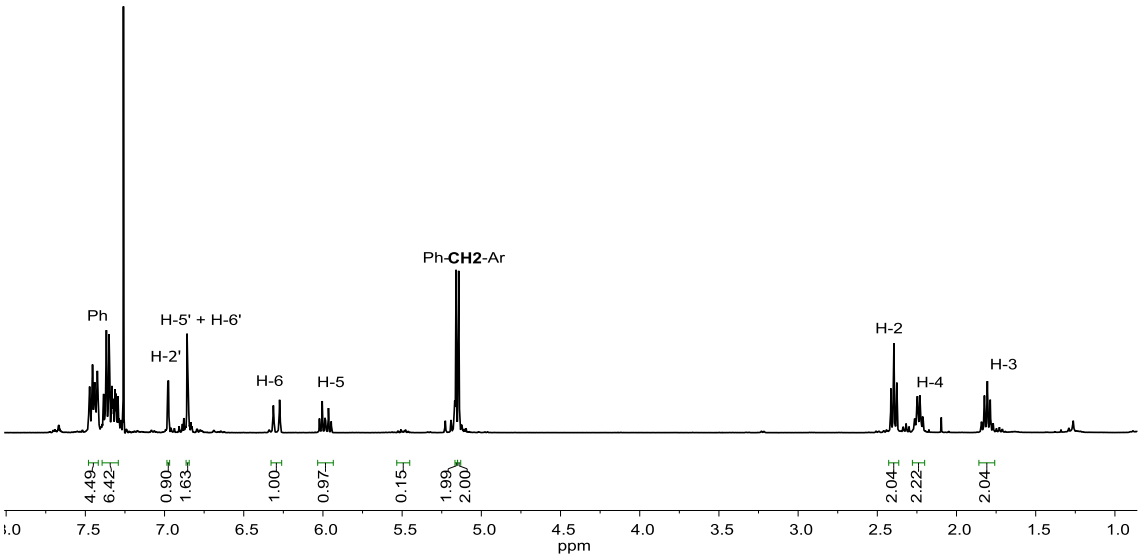
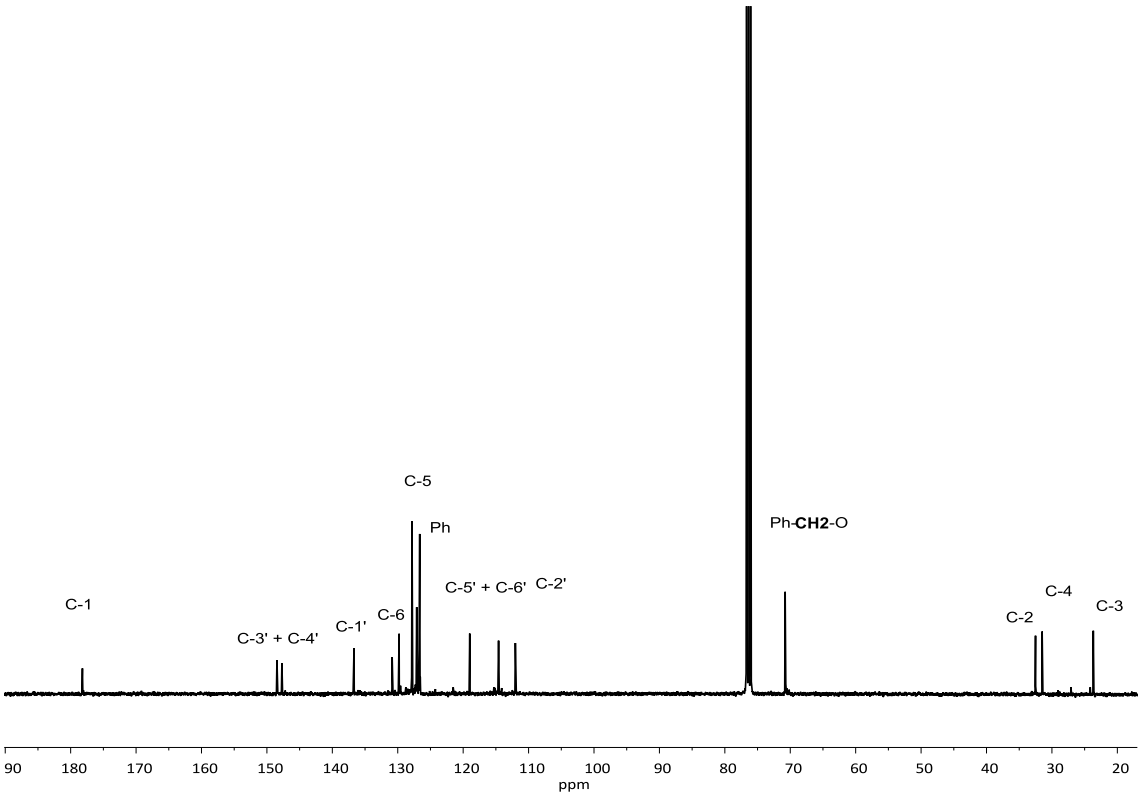
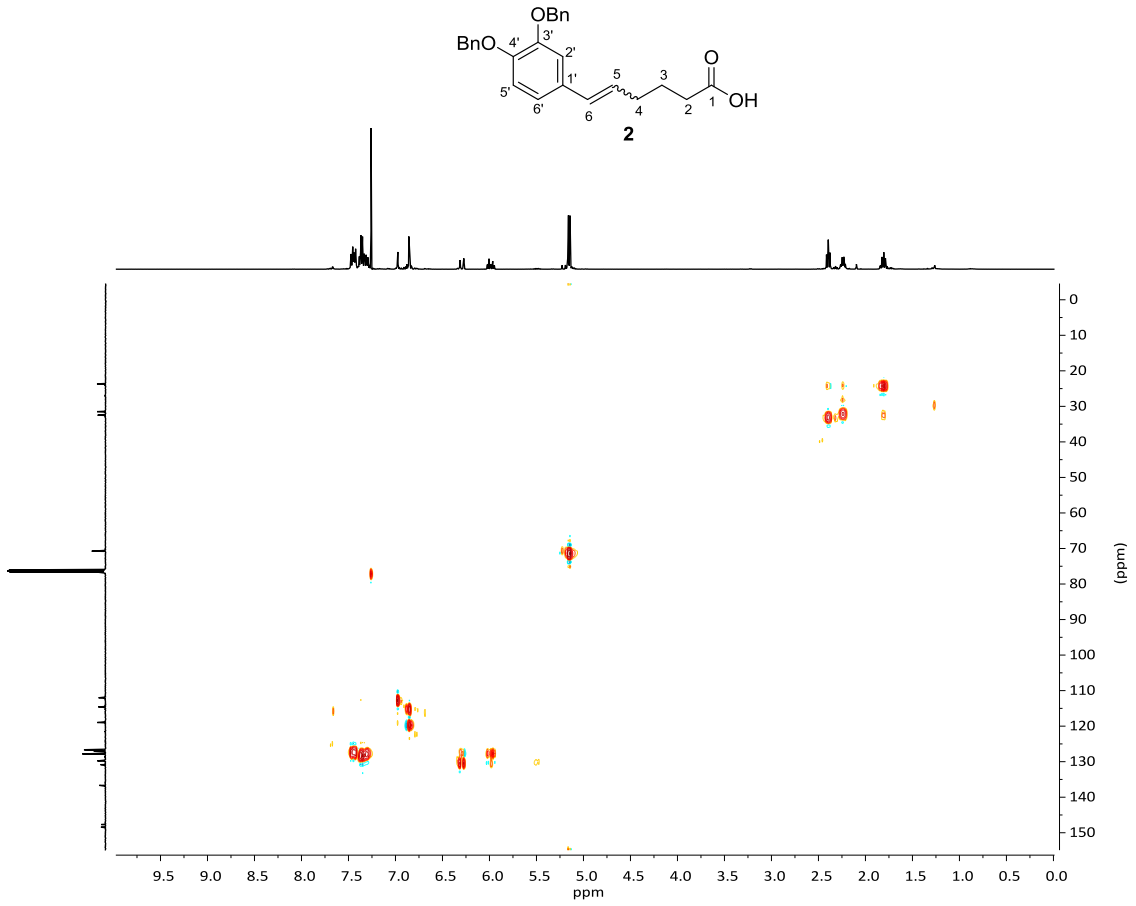


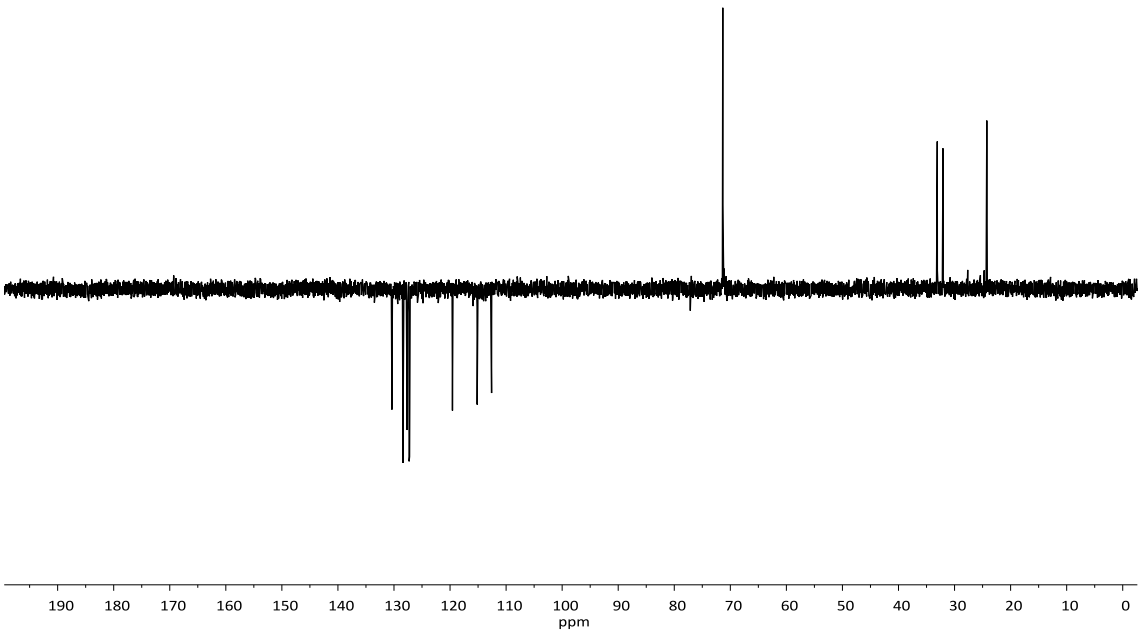
^{1}H NMR (400 MHz, $CDCl_3$)



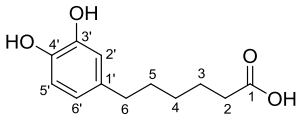
^{13}C NMR (101 MHz, $CDCl_3$)



^{1}H – ^{13}C HSQC



DEPT135 (101 MHz, $CDCl_3$)



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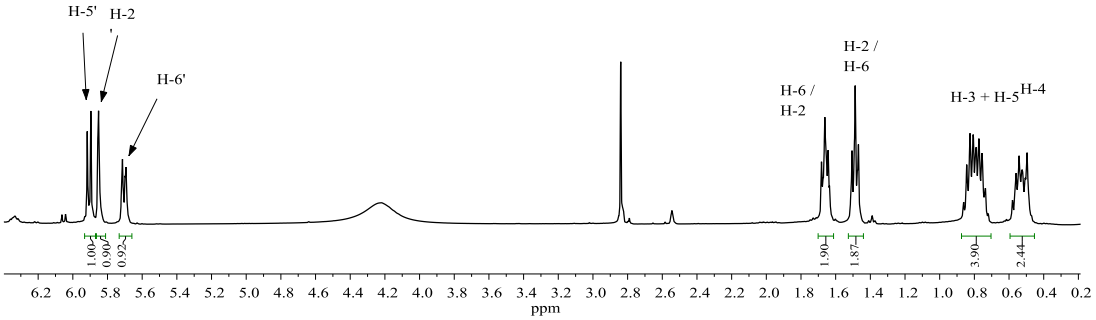

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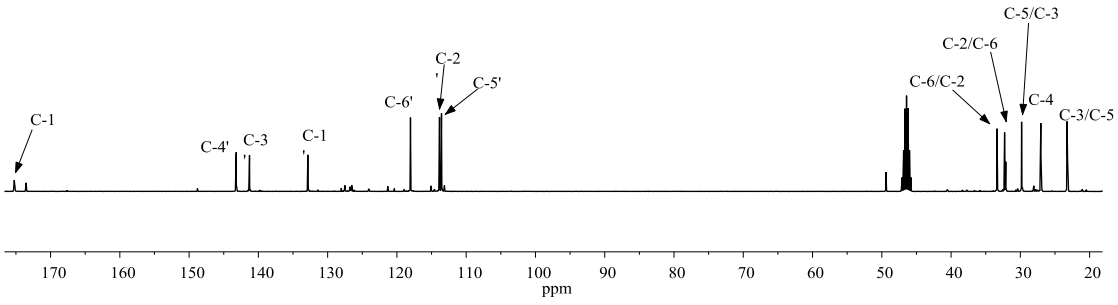

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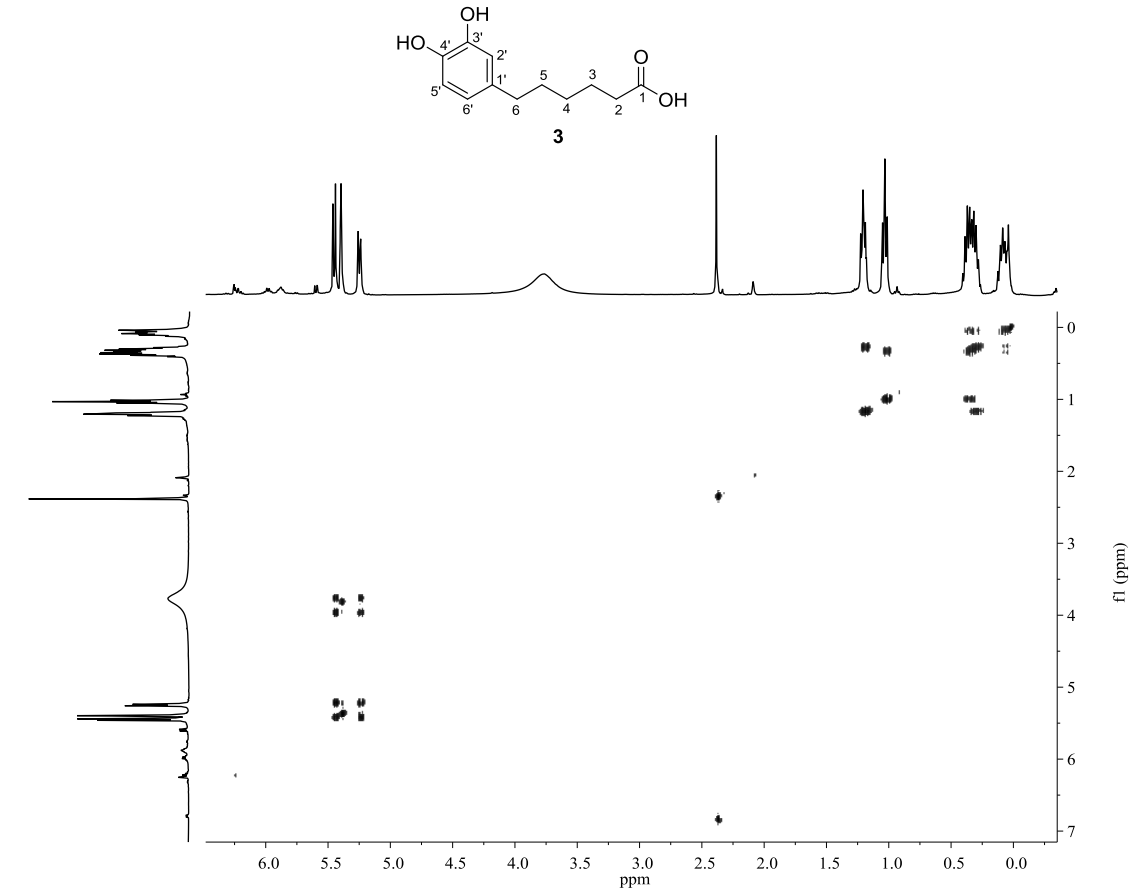
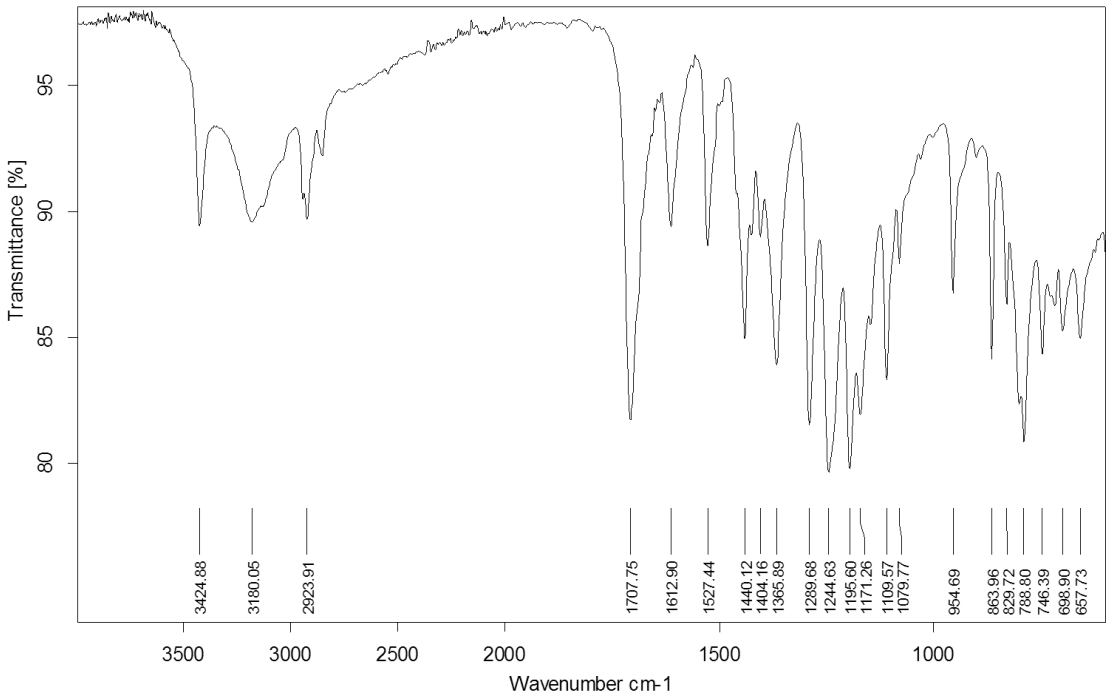


^{1}H – ^{1}H COSY (400 MHz, acetone-d_6)



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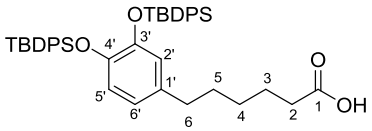

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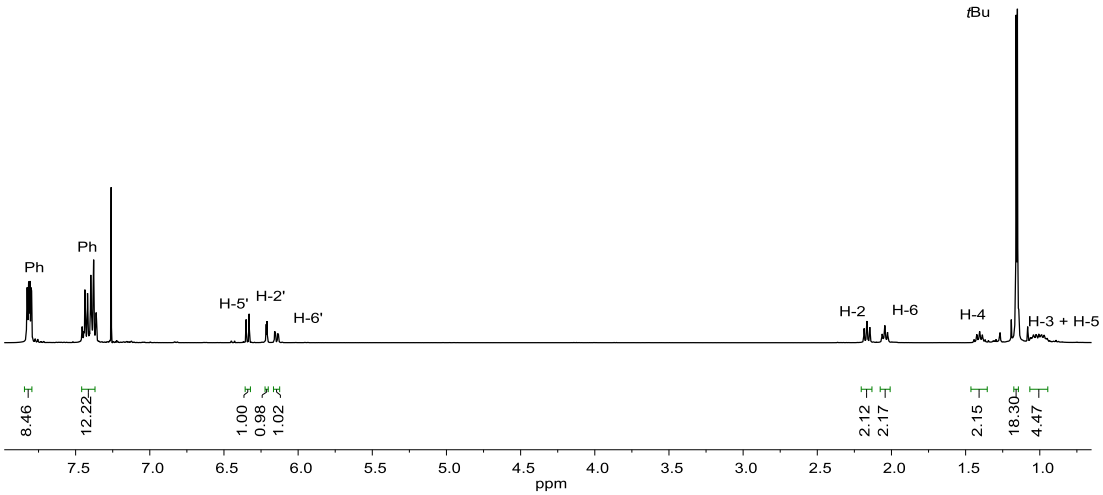

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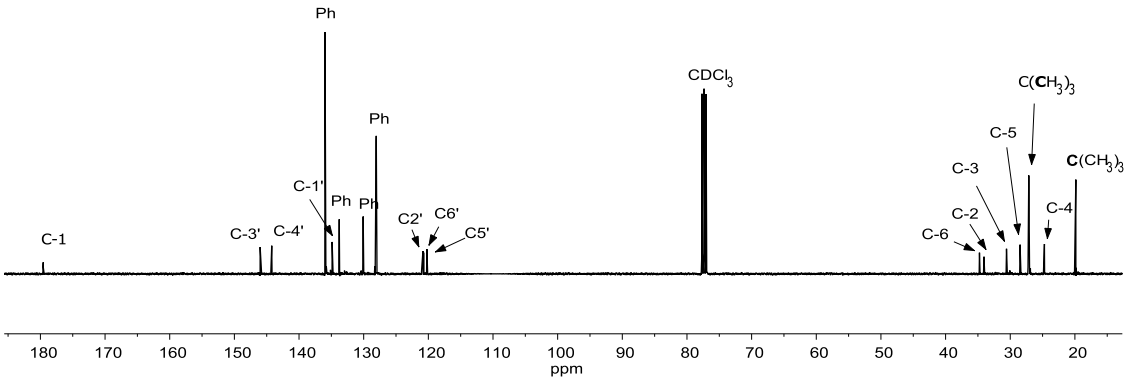

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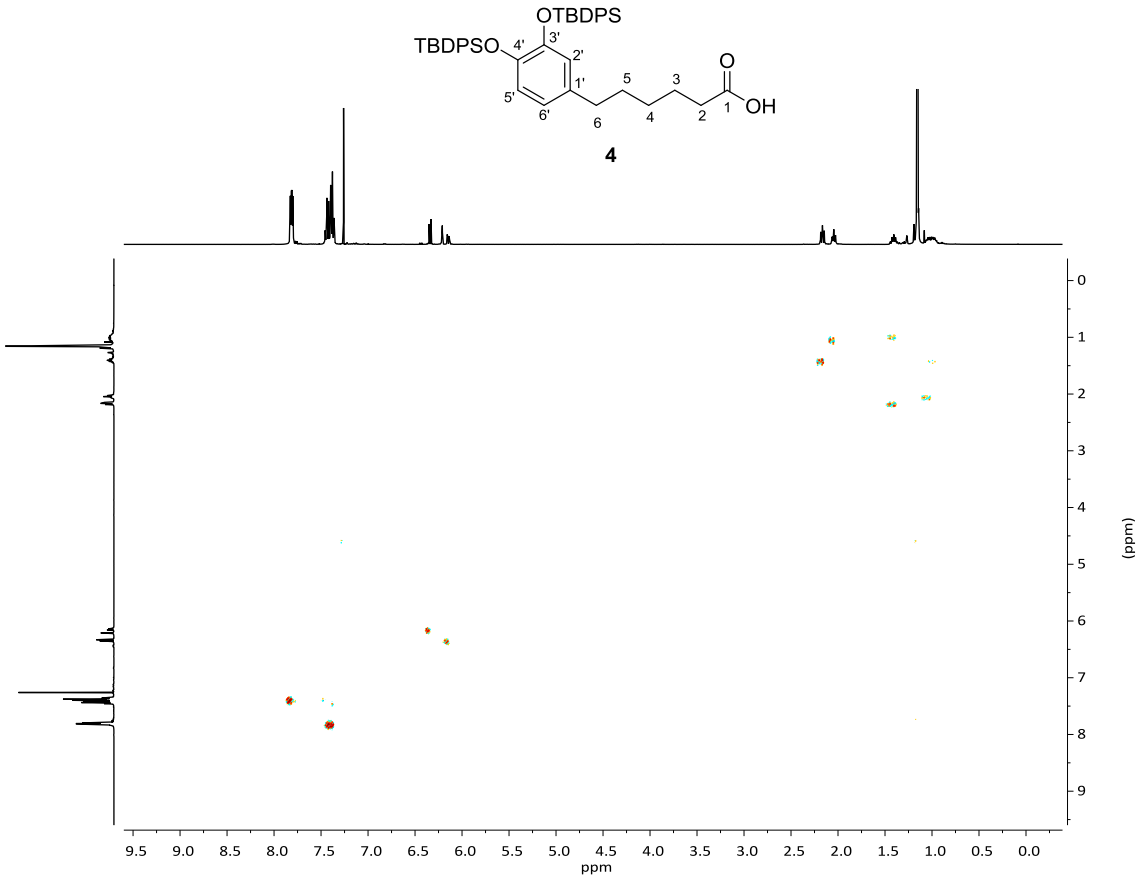
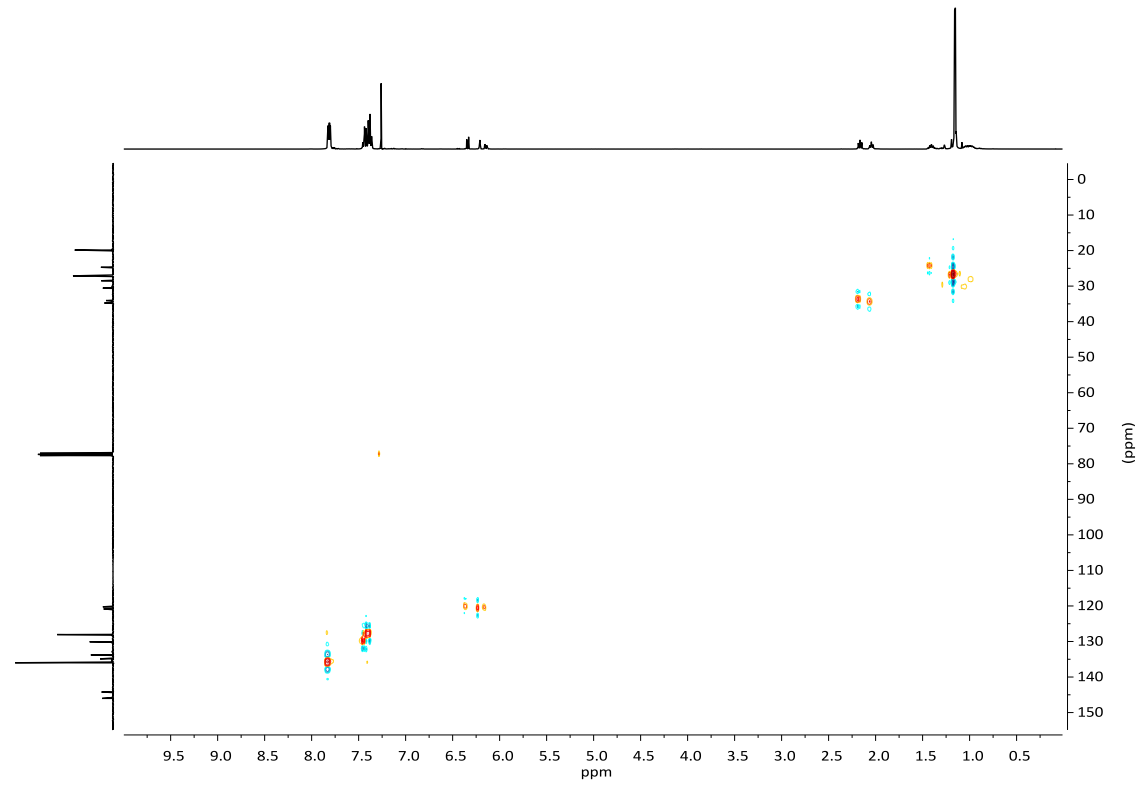


^{1}H – ^{1}H COSY (400 MHz, $CDCl_3$)



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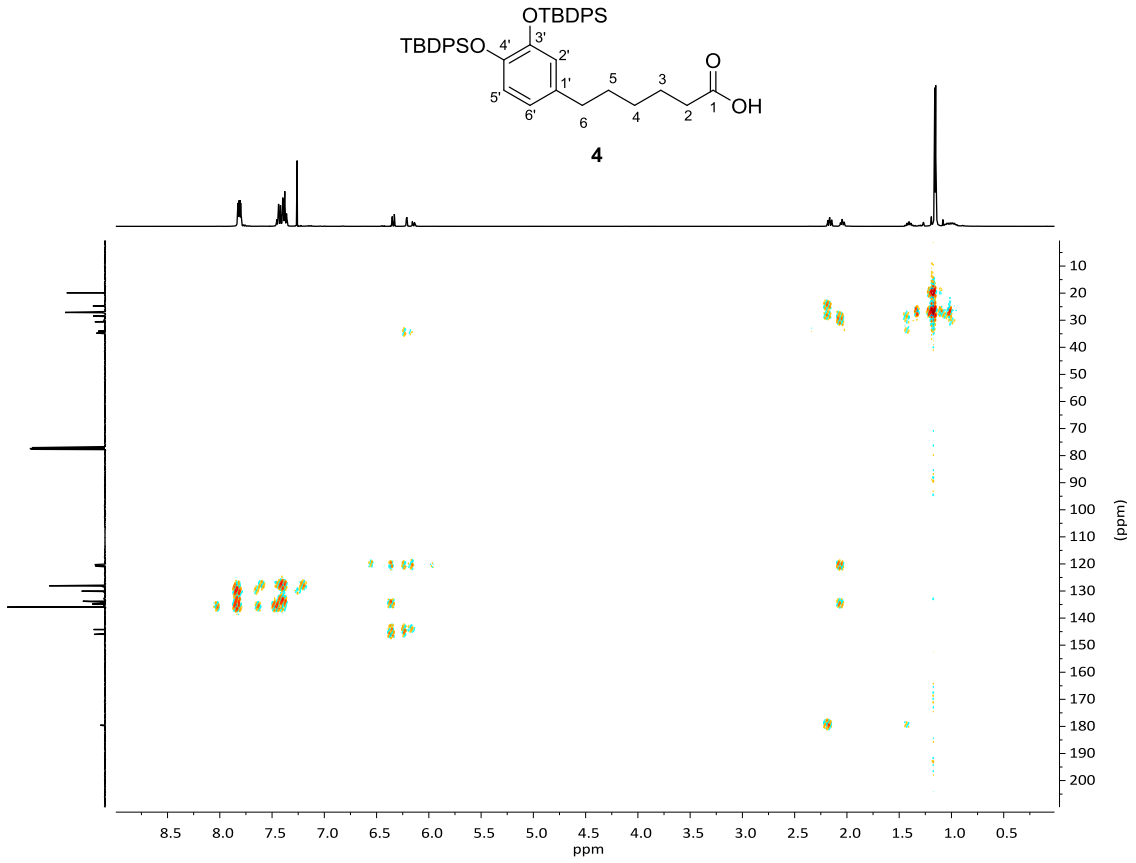

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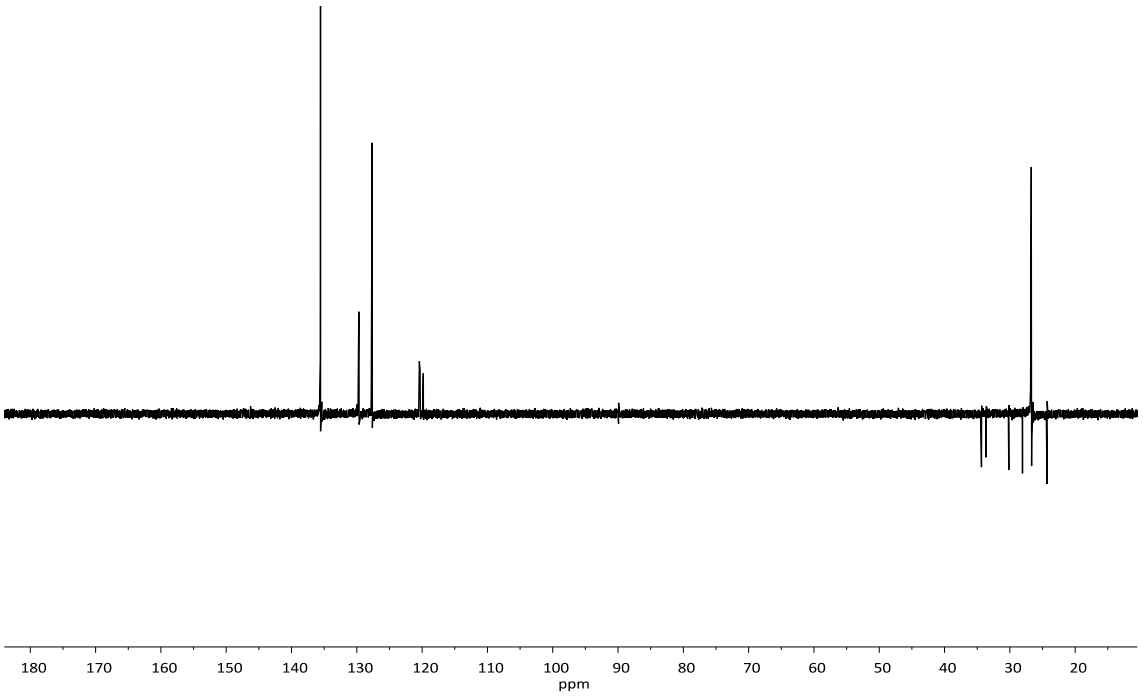
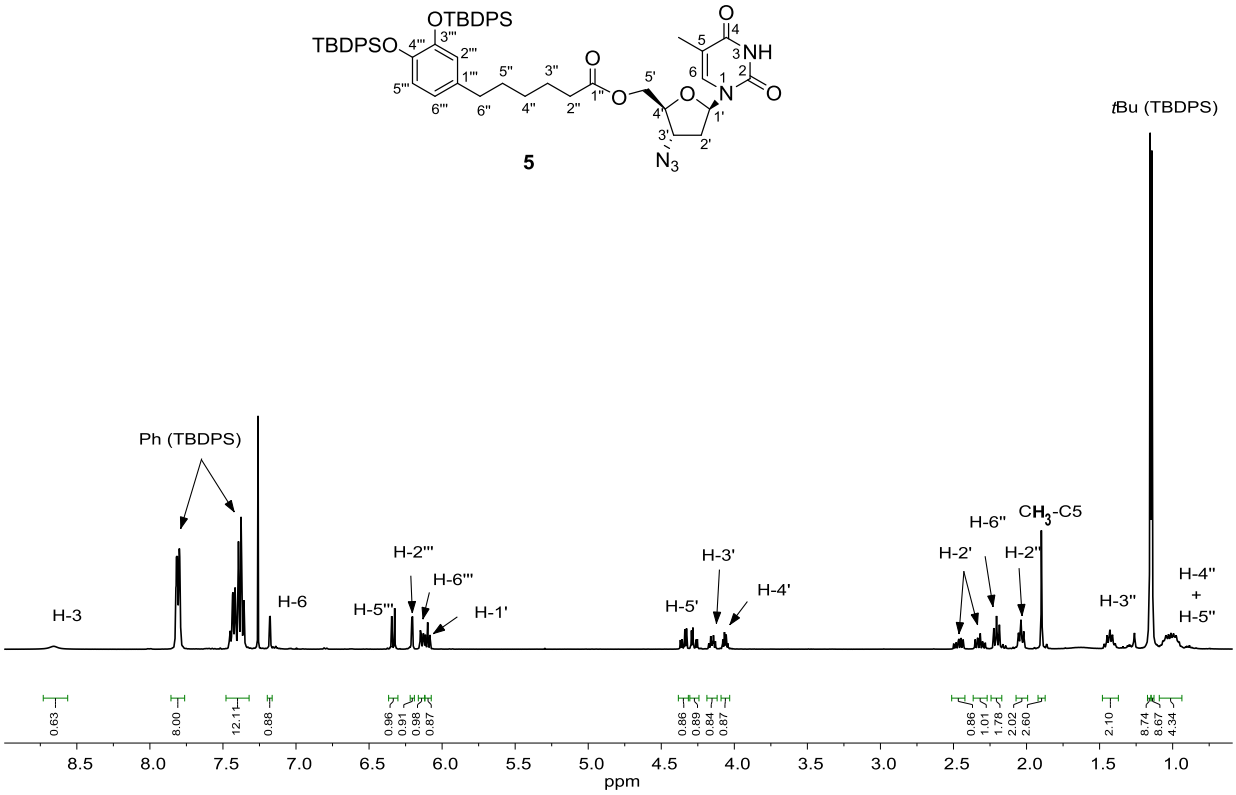


DEPT135 (101 MHz, $CDCl_3$)



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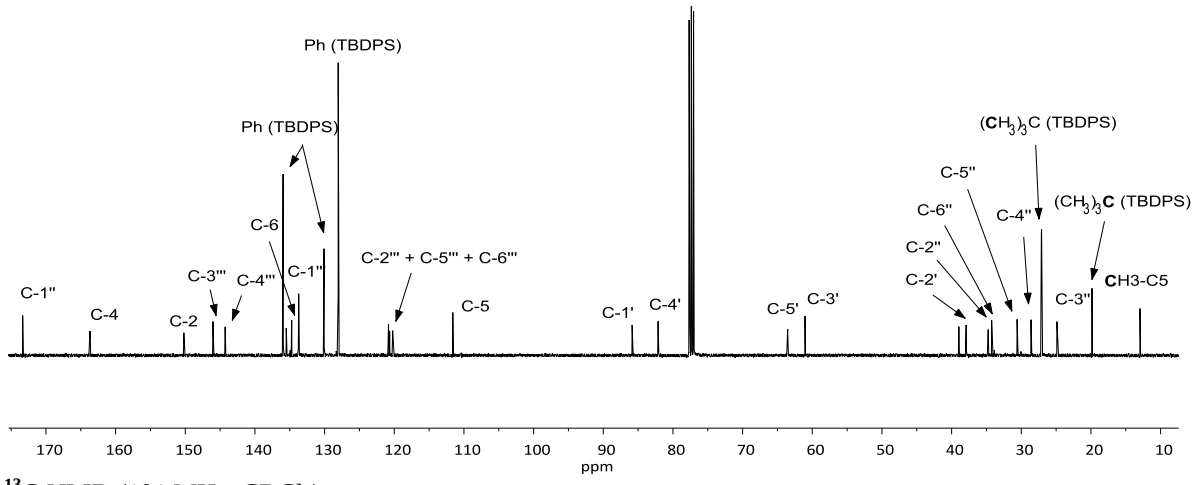

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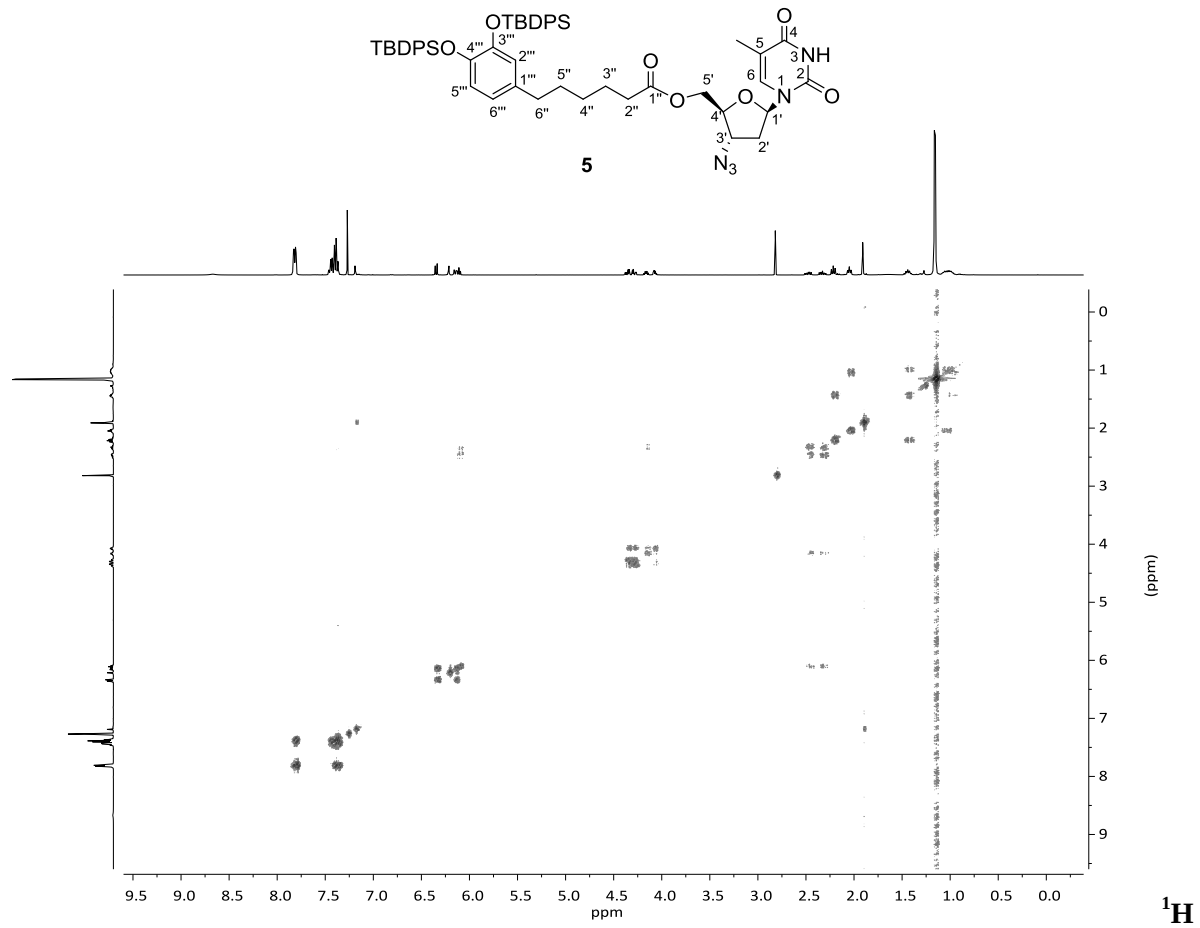

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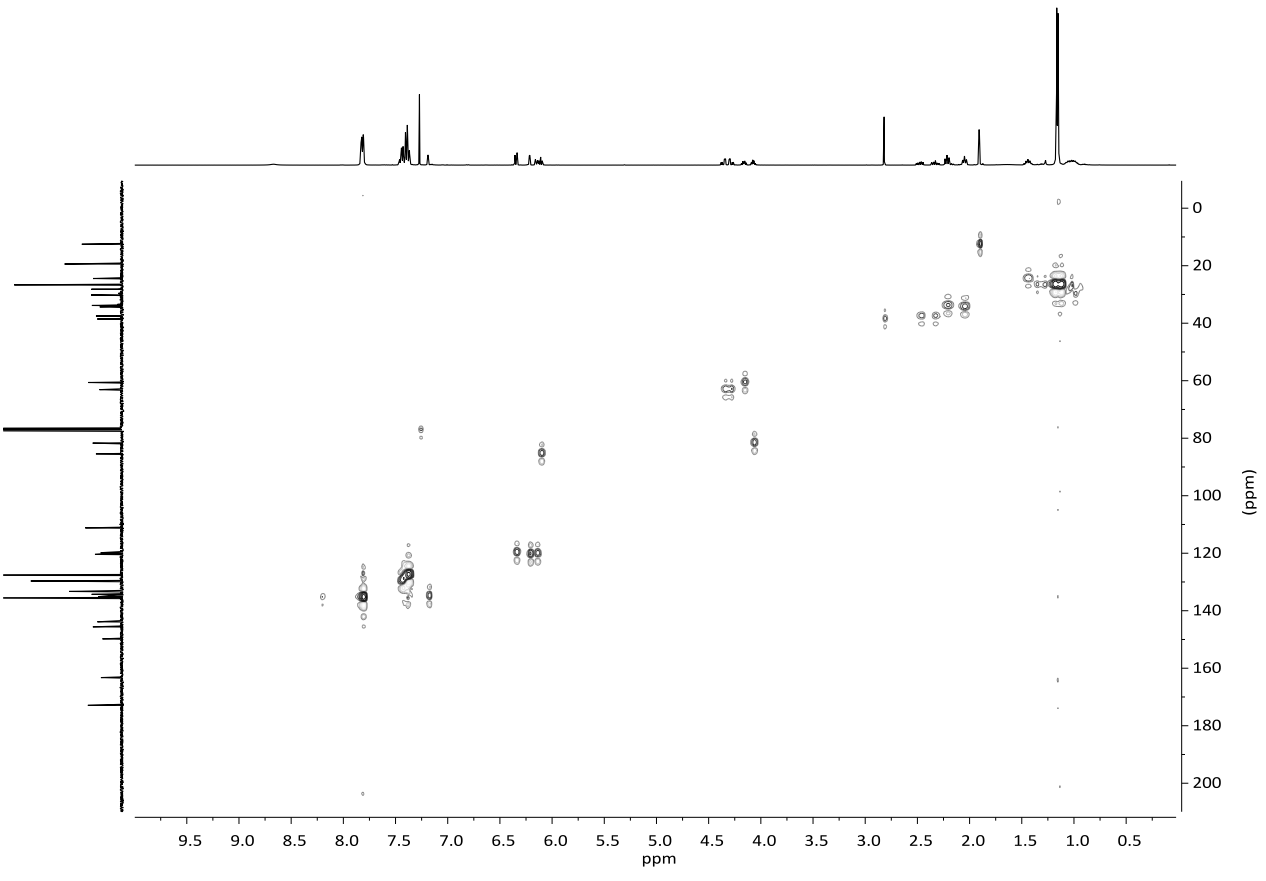

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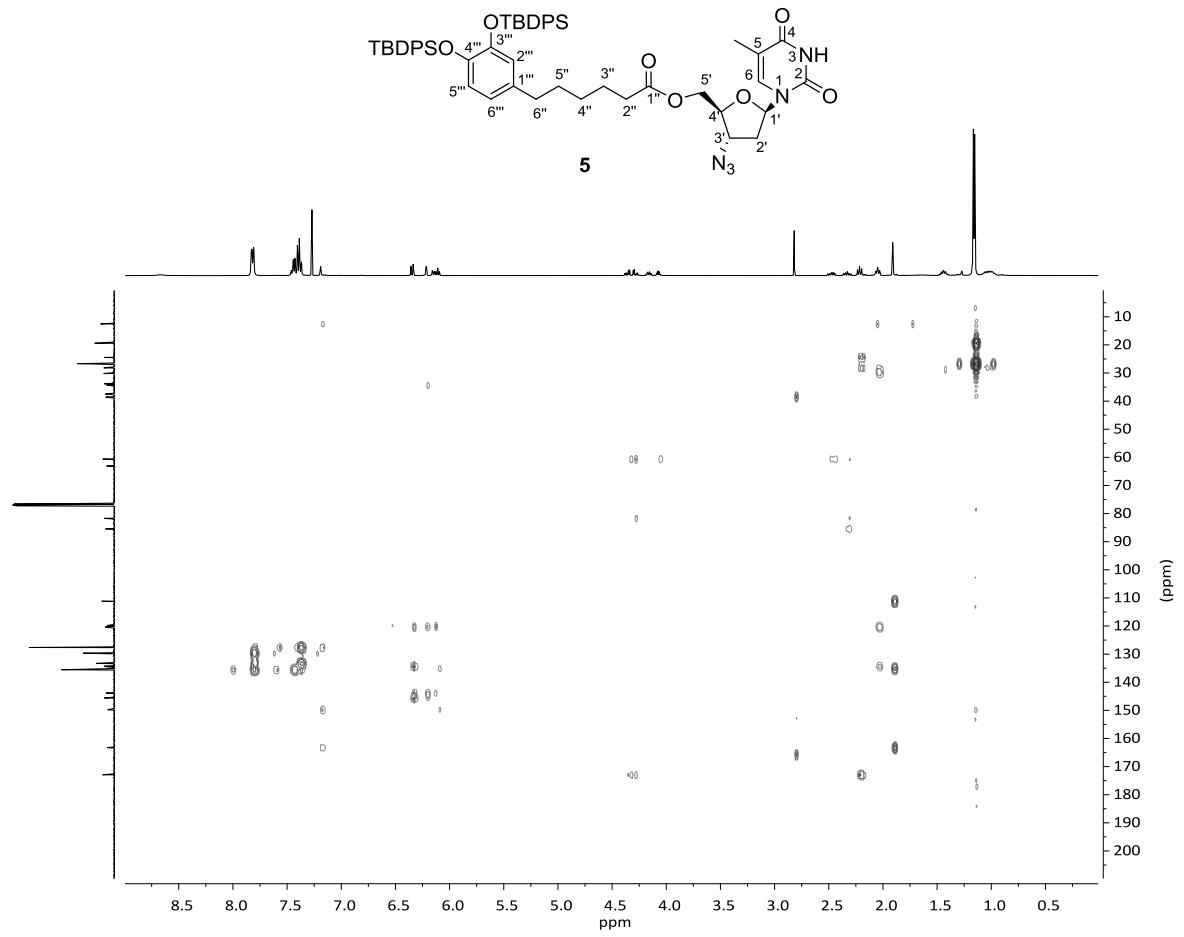

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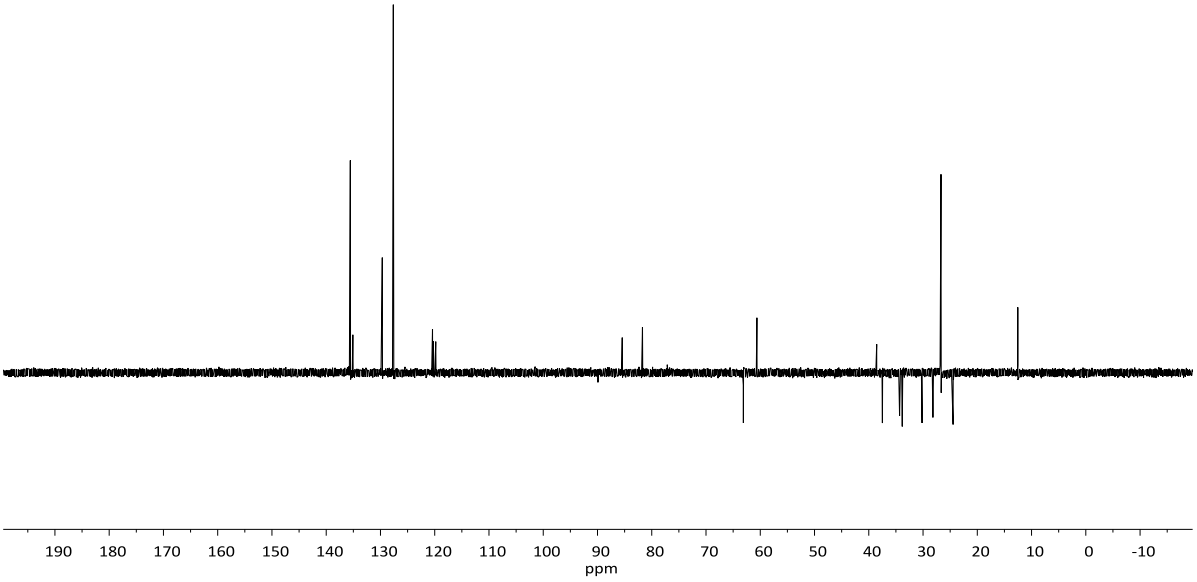
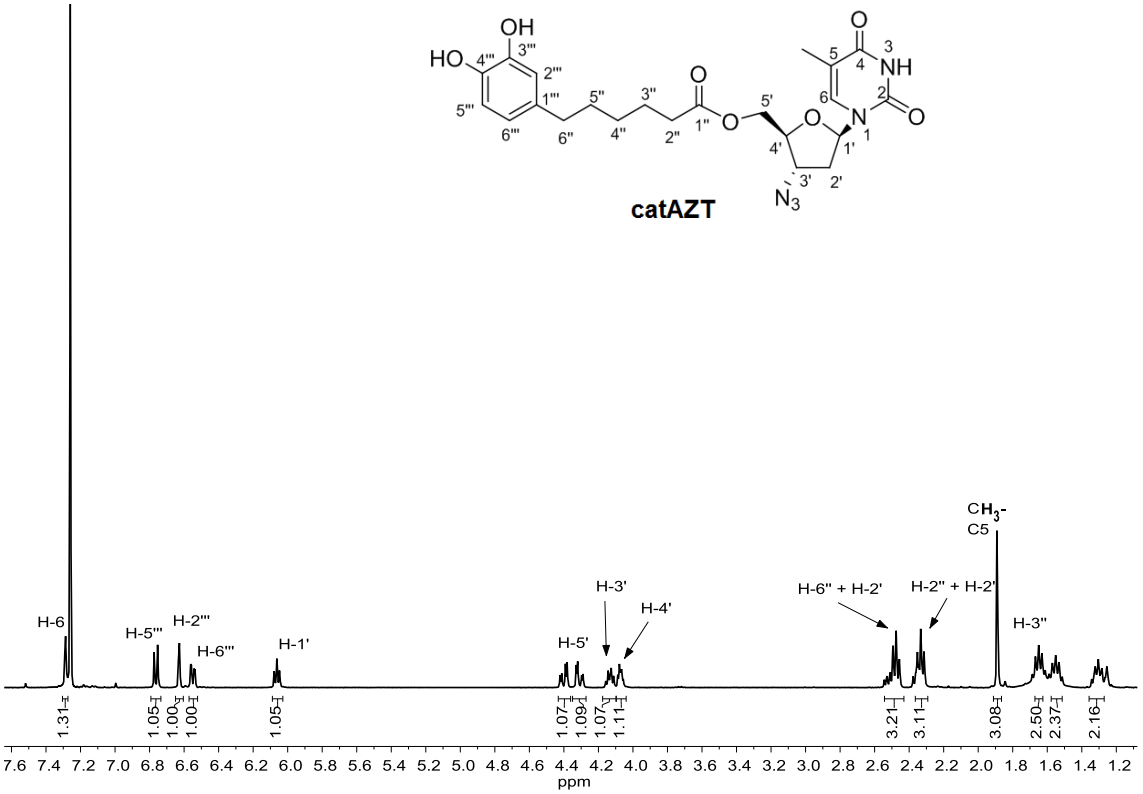
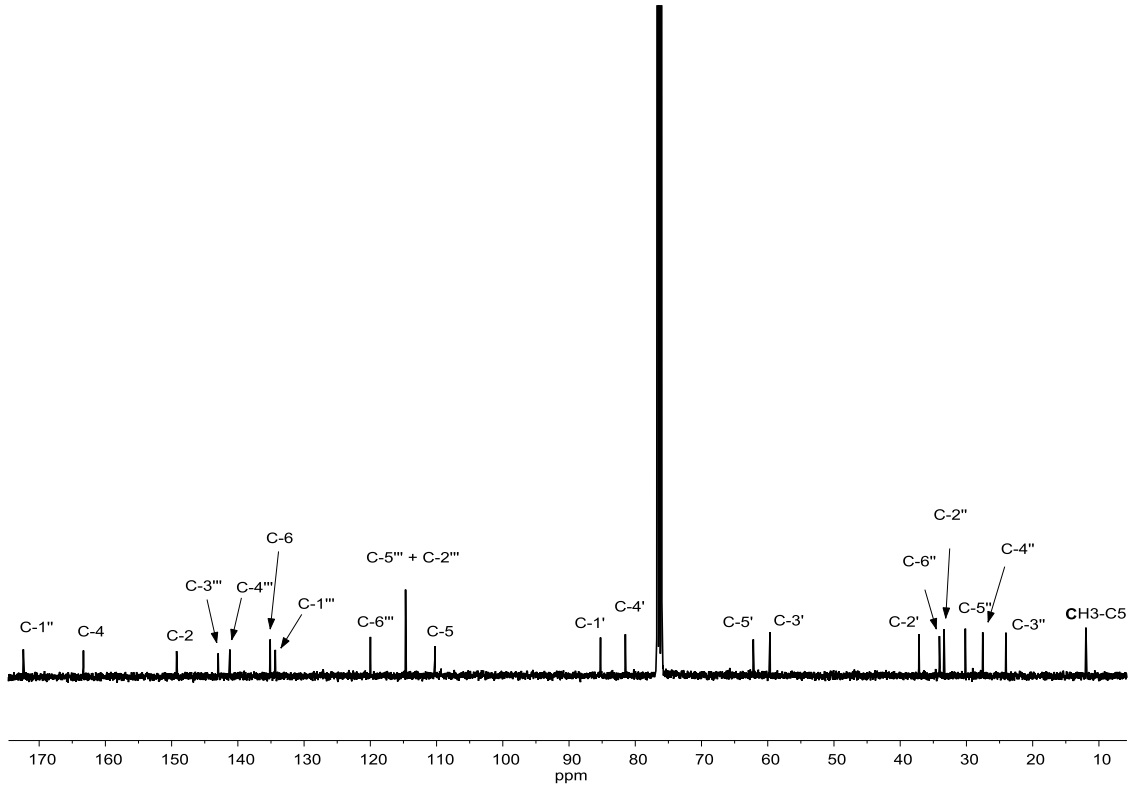


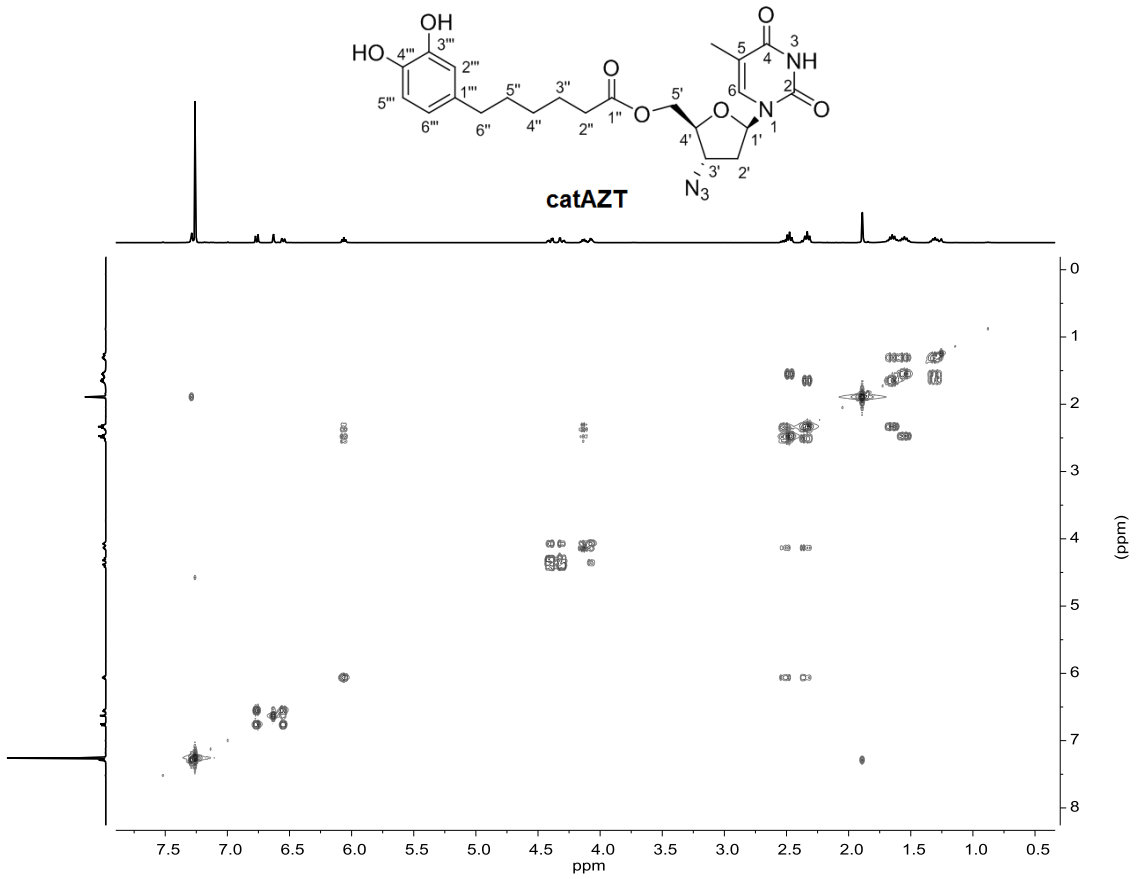
DEPT135 (101 MHz, $CDCl_3$)



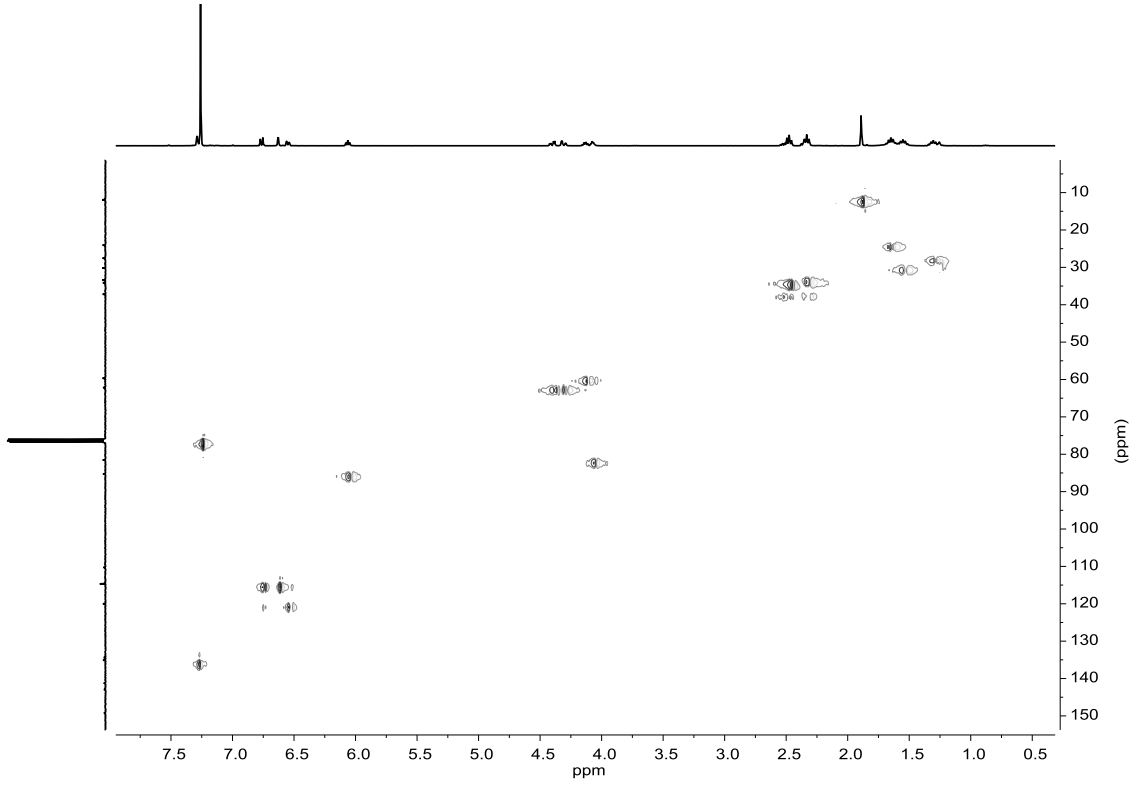
^{1}H NMR (400 MHz, $CDCl_3$)



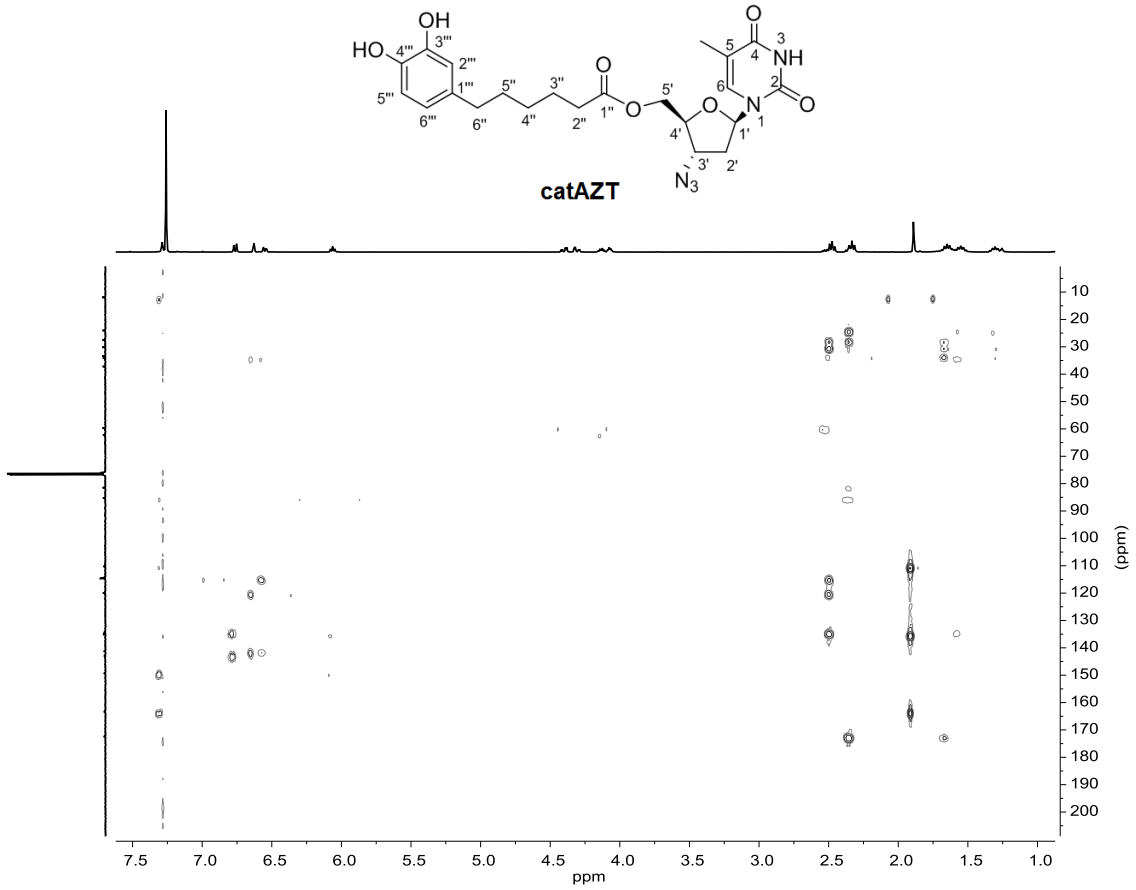
^{13}C NMR (101 MHz, $CDCl_3$)



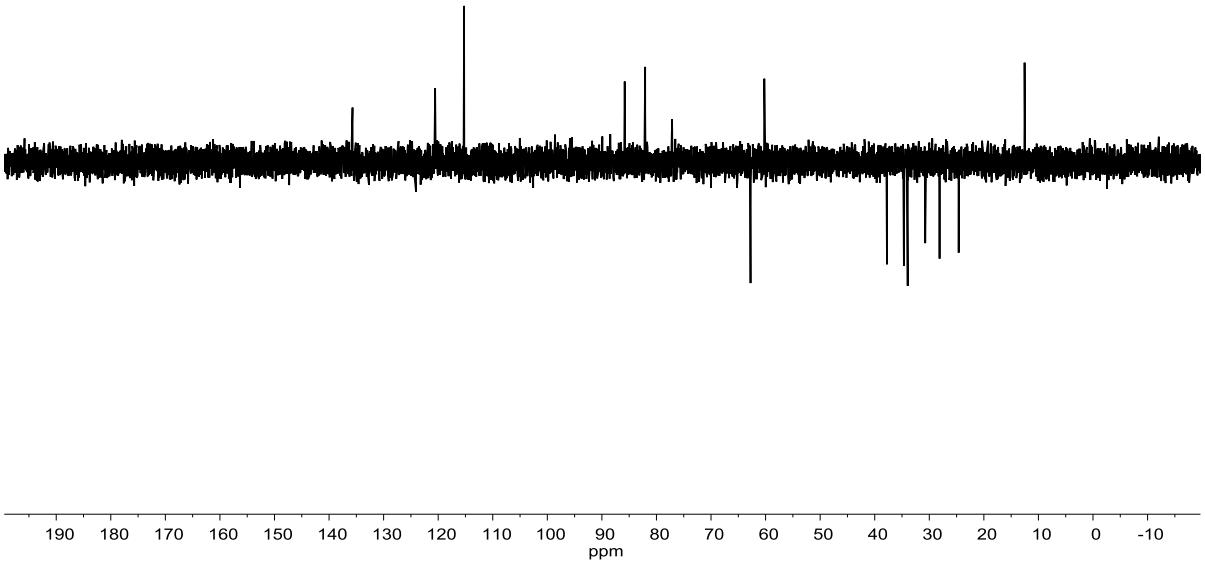
^{1}H – ^{1}H COSY (400 MHz, $CDCl_3$)



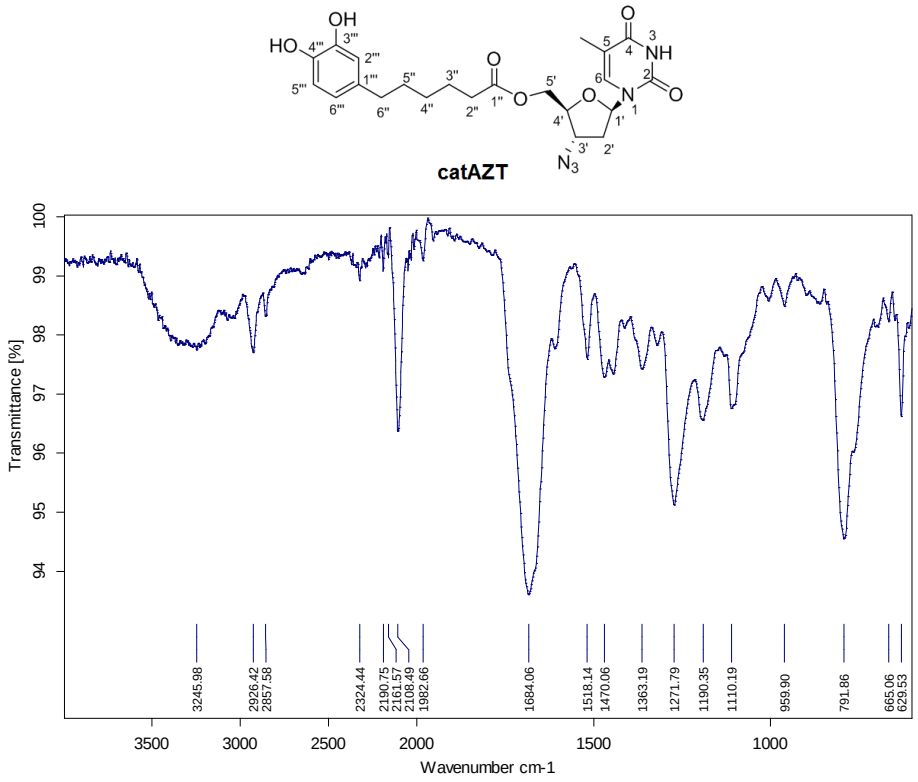
^{1}H – ^{13}C HSQC ($CDCl_3$)



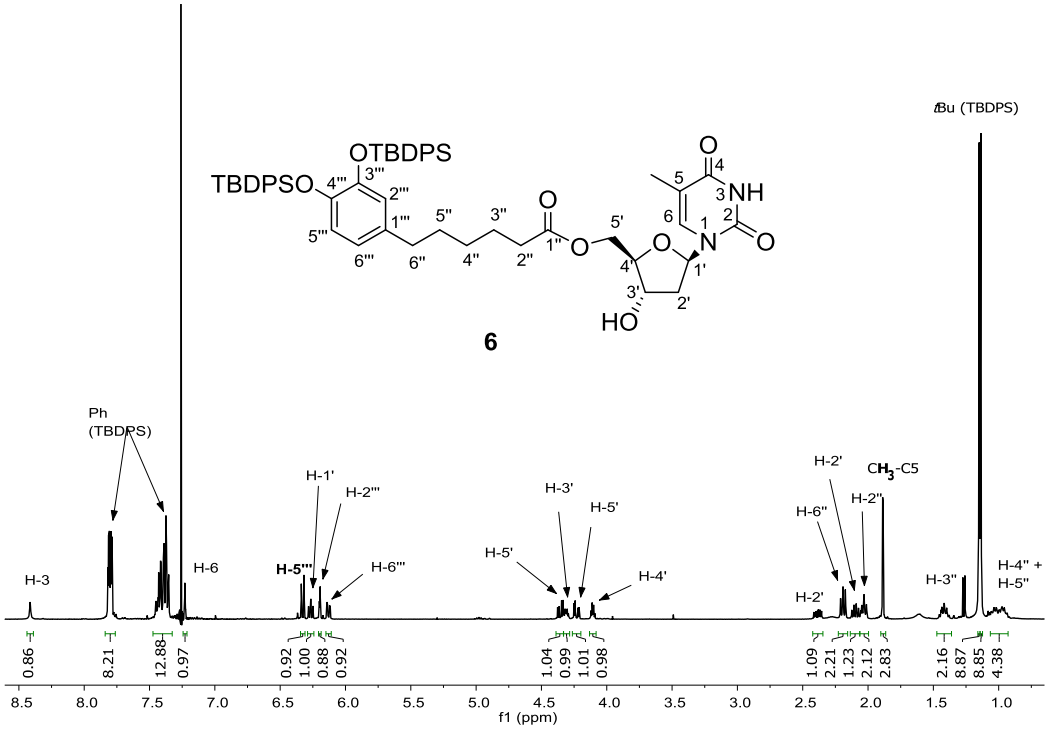
^{1}H – ^{13}C HMBC ($CDCl_3$)



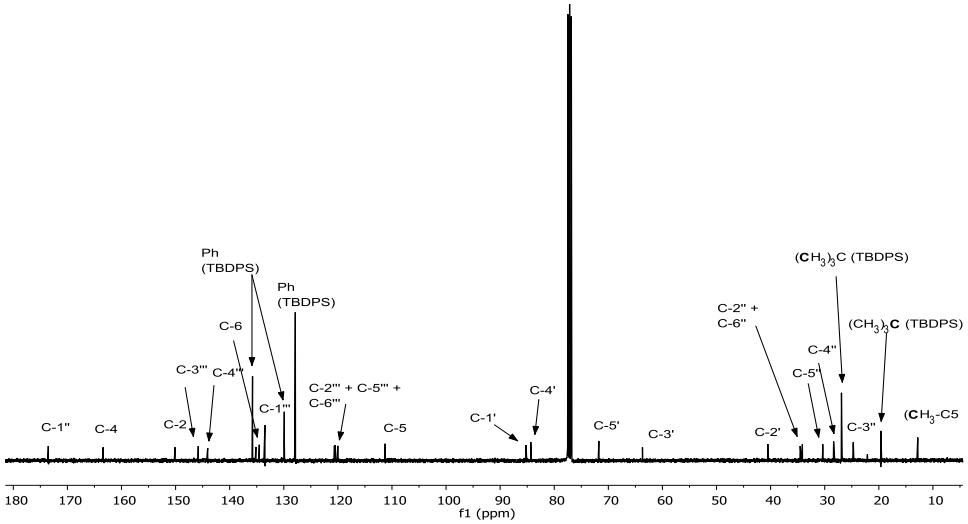
DEPT135 (101 MHz, $CDCl_3$)



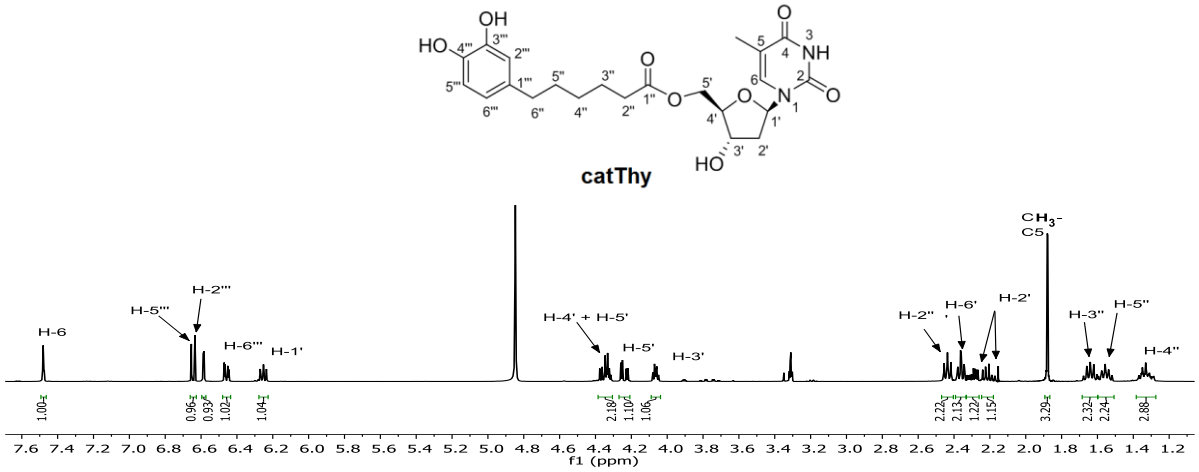
FTIR



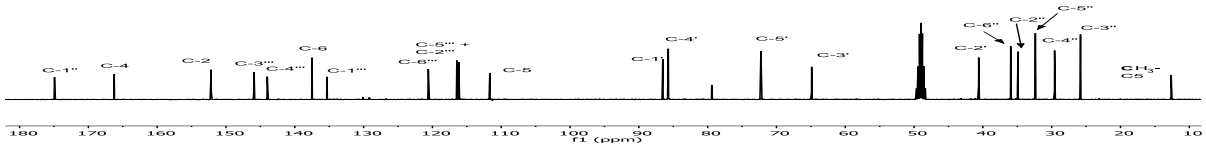
^{1}H NMR (400 MHz, $CDCl_3$)



^{13}C NMR (101 MHz, $CDCl_3$)



^{1}H NMR (400 MHz, CD_3OD)



^{13}C NMR (101 MHz, CD_3OD)

S2. Characterization of catAZT-NCPs nanoparticles

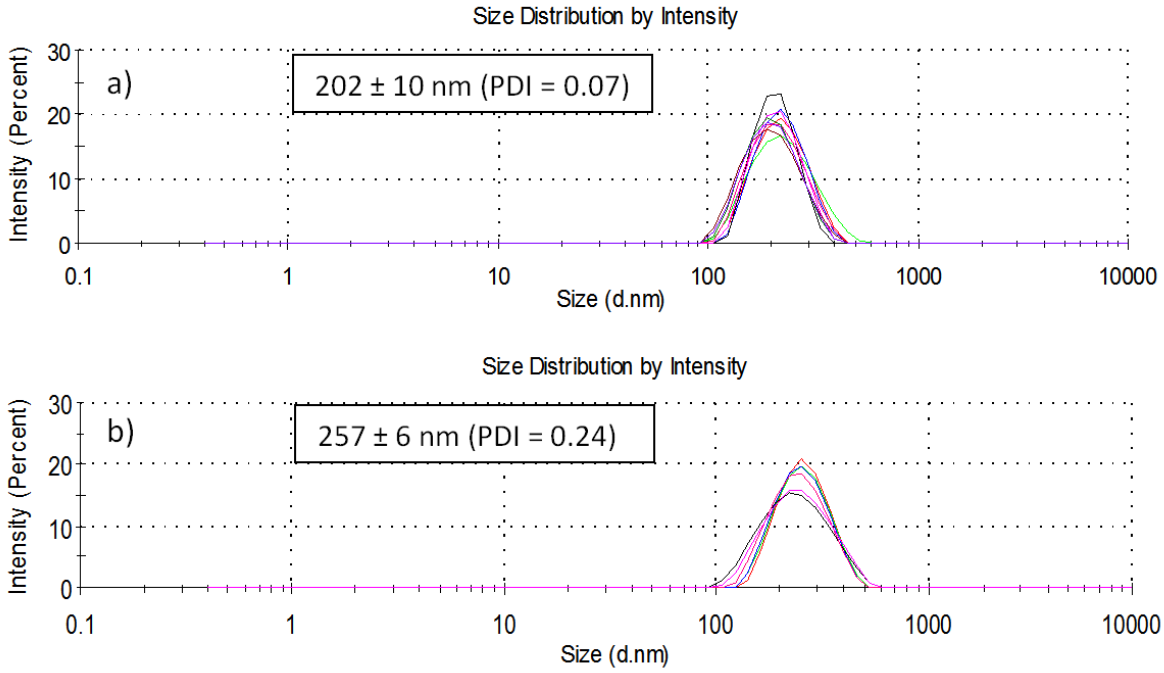


Figure S1. DLS measured of catAZT-NCPs particle size in a) ethanol, and b) PBS/BSA 0.5 mM.

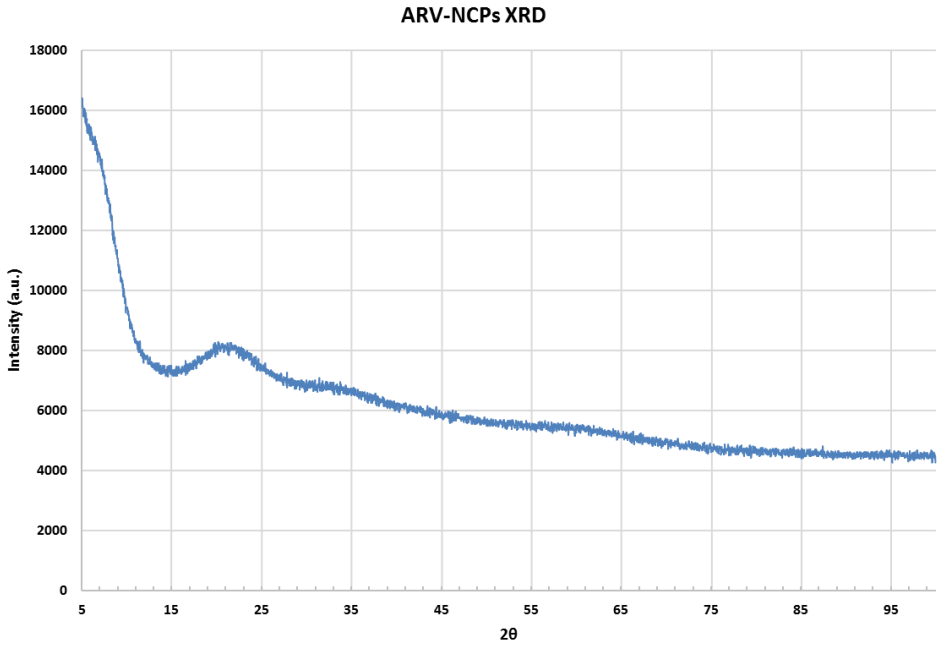


Figure S2. XRD pattern of catAZT-NCPs.

Table S1. Table of Elemental Analysis results for three different synthesis and comparison between the average results and the proposed chemical formula. The results indicate a good reproducibility of the synthetic method.

Batch	%C	%H	%N
1	51.93	5.08	13.83
2	51.96	5.00	13.77
3	51.29	5.03	13.72
Average	51.73	5.04	13.77
$[Fe(catAZT)_{1.51}(bix)_{0.62}(AcO)(H_2O)_{2.45}]$	51.76	5.38	13.79

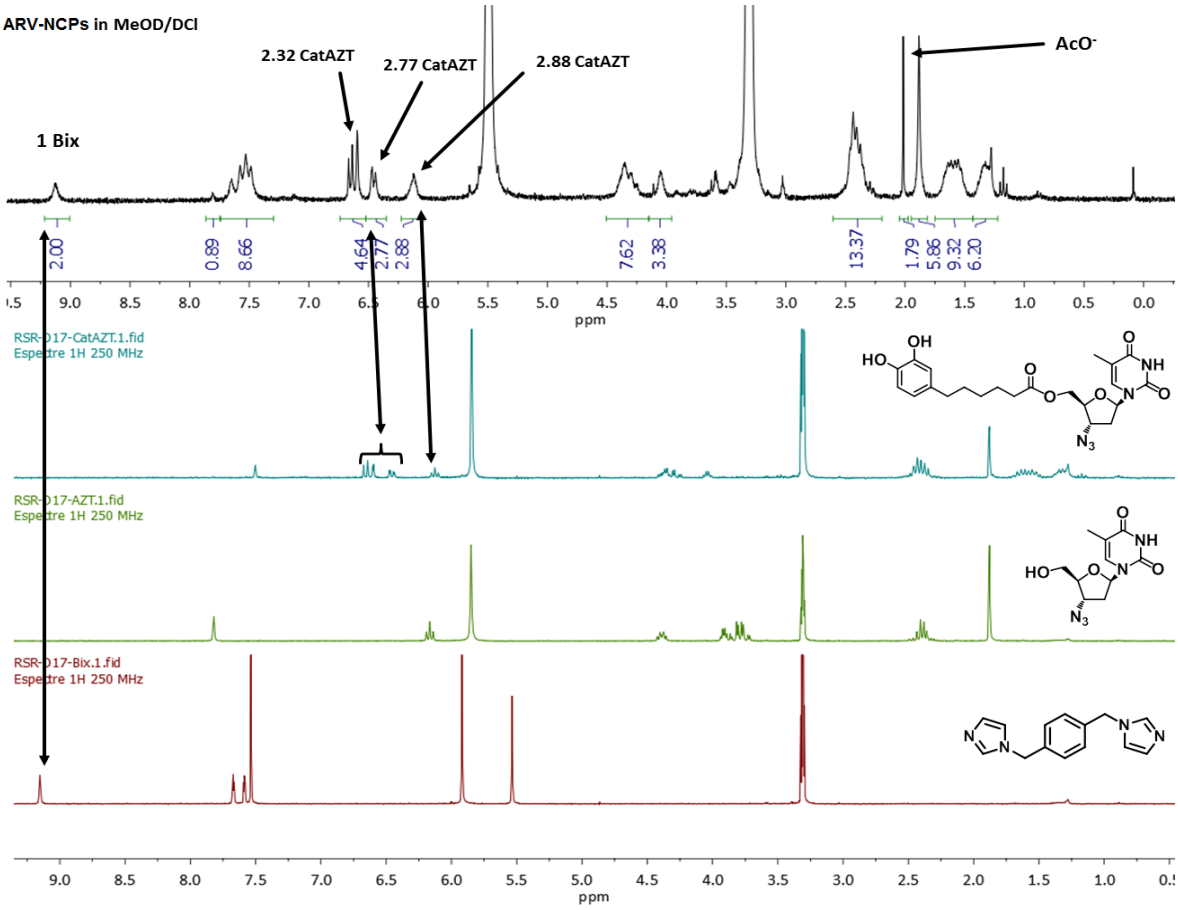


Figure S3. ^{1}H NMR spectra of (top to bottom) catAZT-NCPs, catAZT, AZT and Bix. catAZT-NCPs spectrum was recorded in a DCl/CD_3OD acidic solution (50 µL DCl / mL CD_3OD. Peaks corresponding to catAZT, Bix and acetate are observed in the ^{1}H NMR spectrum of catAZT-NCPs.

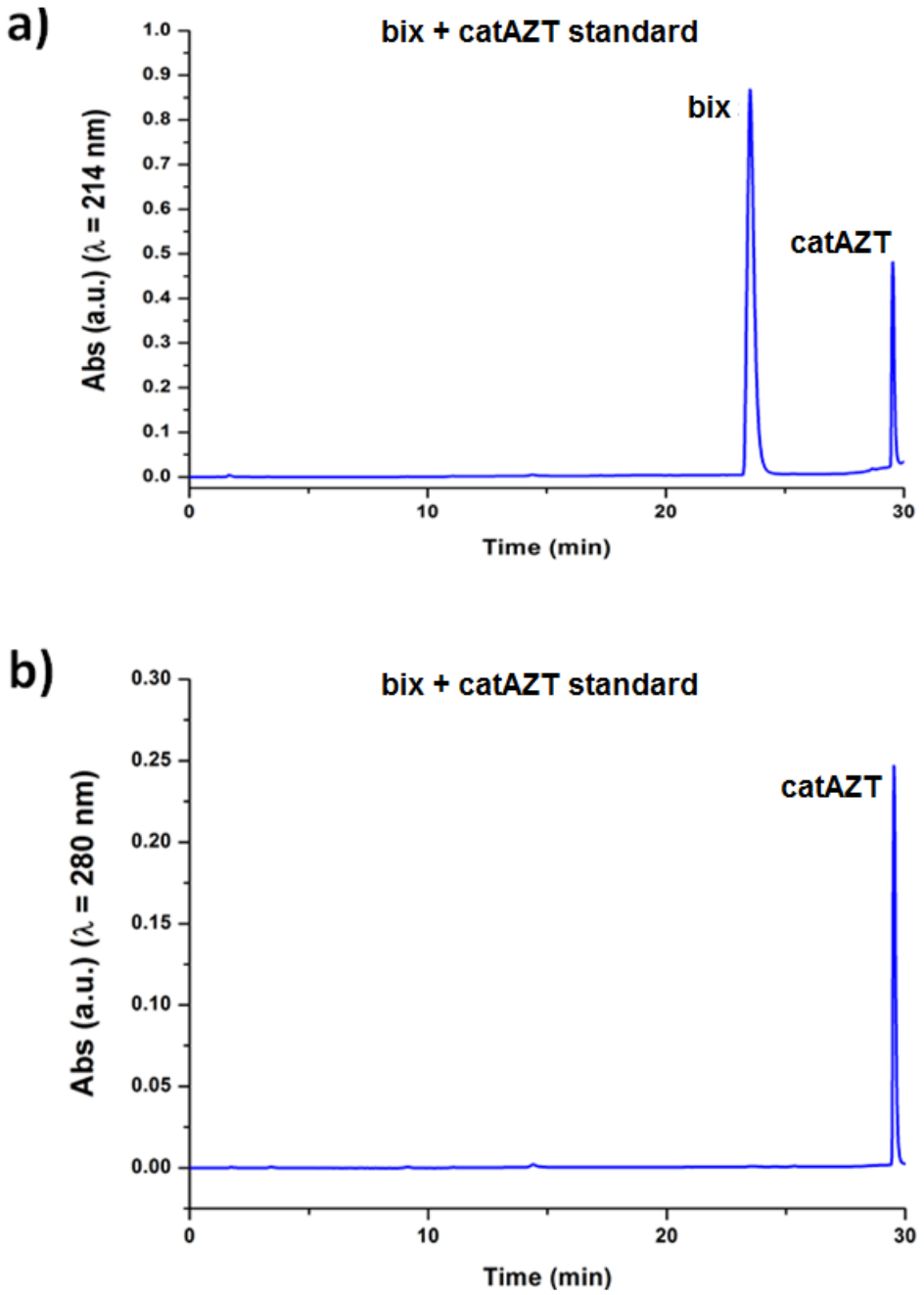


Figure S4. HPLC chromatogram of standards containing Bix (885 µM) and catAZT (456 µM). Detection was made by UV detector at a) 214 and b) 280 nm.

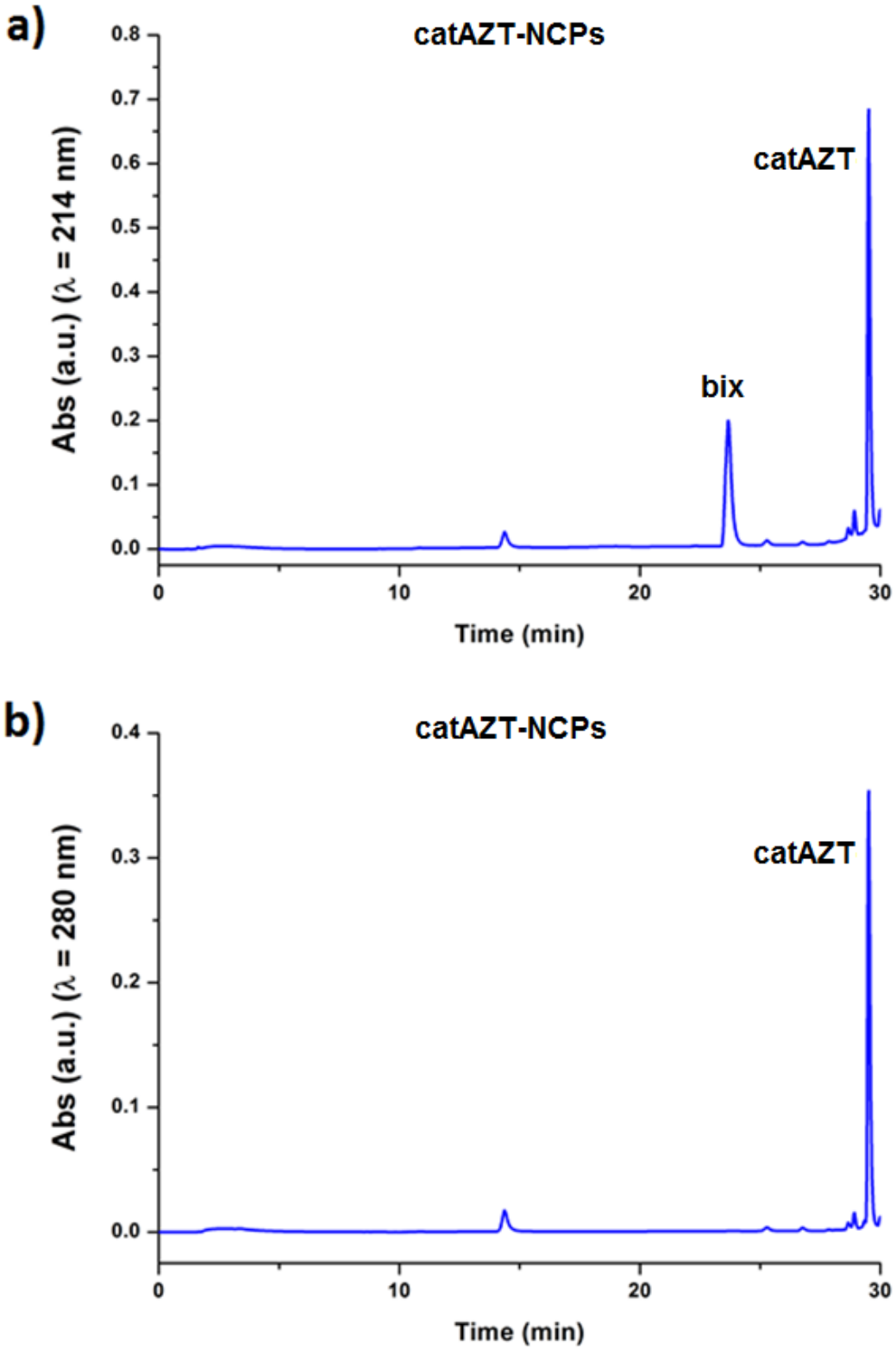


Figure S5. HPLC chromatograms of catAZT-NCPs dissolved in a mixture of HCl/methanol (50 µL concentrated HCl / mL methanol). Detection was made by UV detector at a) 214 and b) 280 nm.

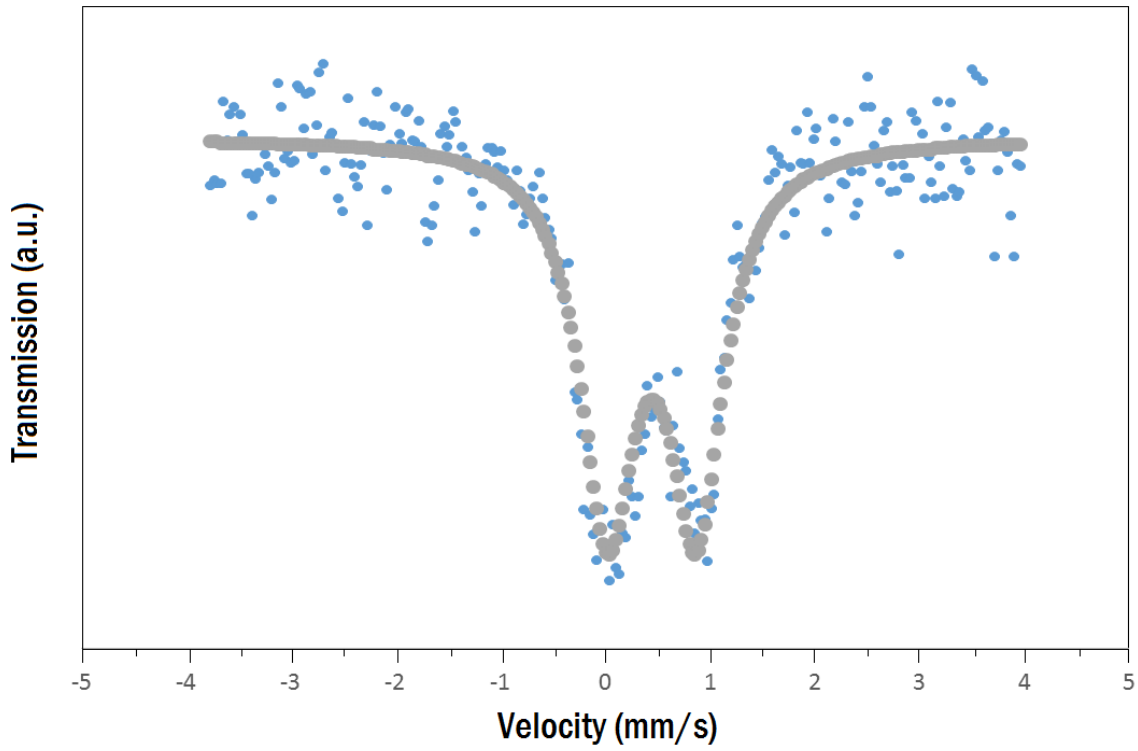


Figure S6. Mössbauer spectra for catAZT-NCPs at 293 K. Experimental data (small blue dots), and computer fitted spectrum (big grey dots) for high-spin Fe(III). Hyperfine parameters of the fitting of the Mössbauer spectra at 293 K showed the isomer shift relative to the metallic iron (δFe), quadrupolar splitting (ΔEq) and the full width at half maximum (Γ). The spectrum was fitted to a single doublet with a ΔEq= 0.85 ± 0.02 mm/s and Γ= 0.31 mm/s. The fitting was centered at an isomeric shift δ= 0.44±0.01 mm/s attributed to high-spin Fe(III) ions.

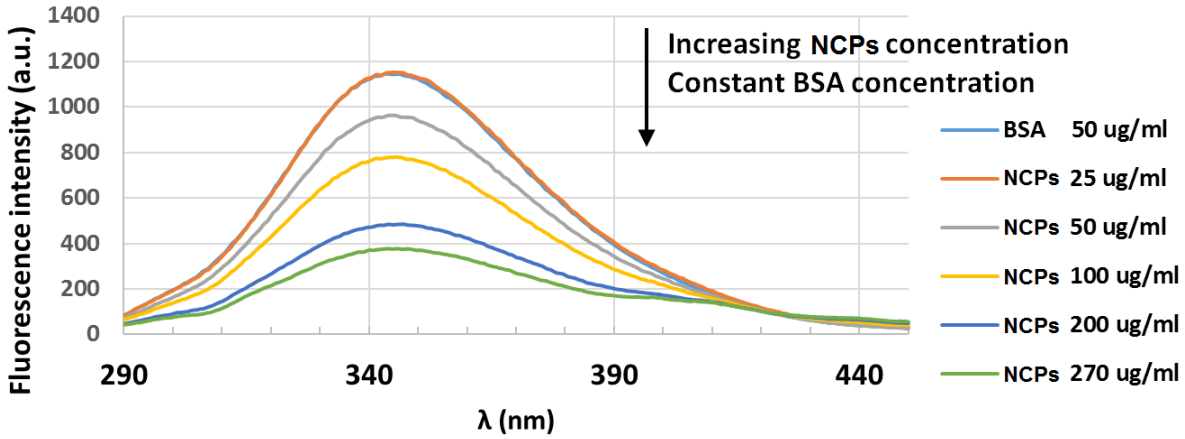


Figure S7. Study of catAZT-NCPs interaction with bovine serum albumin (BSA) by the measurement of the fluorescence quenching of BSA after the addition of different amount of catAZT-NCPs.

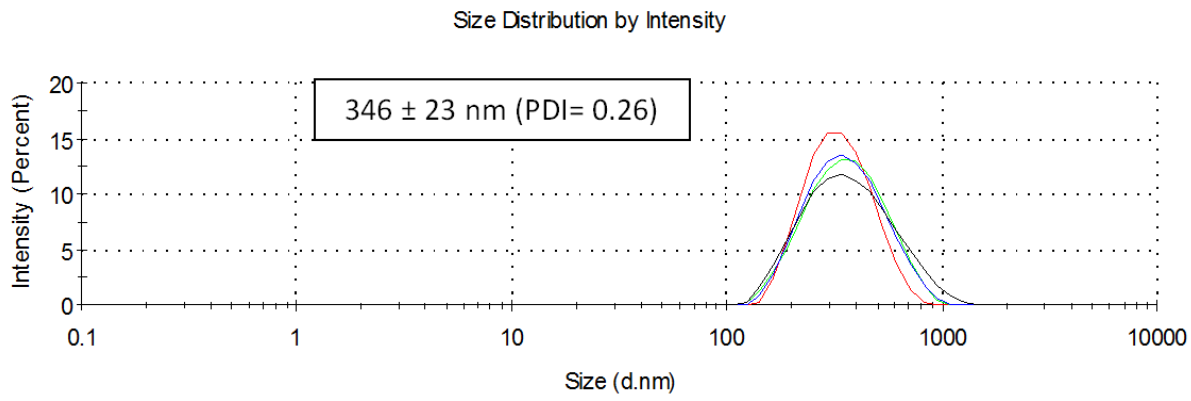


Figure S8. The highest concentrations of nanoparticles used for the *in vitro* studies resulted in unstable colloidal dispersions. After several different experiments using a broad variety of stabilizers, for example polyvinylpyrrolidone (PVP), polyethylene glycol 400 (PEG) or polysorbates, stable colloidal suspensions were obtained by adding a 50% (w/w) of sucrose to the PBS/BSA. In the figure, DLS measured of catAZT-NCPs particle size in a water/sucrose solution (1:1 ratio).

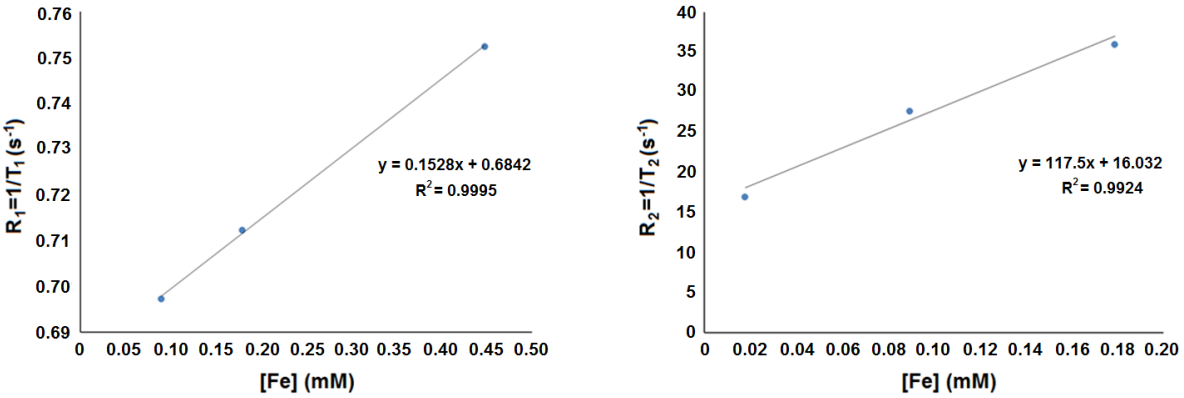


Figure S9. Results of the MR relaxivity experiments with catAZT-NCPs. The relaxation rates were measured related to [Fe]. Plot of R1 (1/*T*1) and R2 (1/*T*2) in front of total [Fe]. MR studies were carried out at the joint NMR facility of UAB and CIBER-BBN, Unit 25 of NANBIOSIS, with a 7T horizontal magnet.

S3. Characterization of catTHY-NCPs

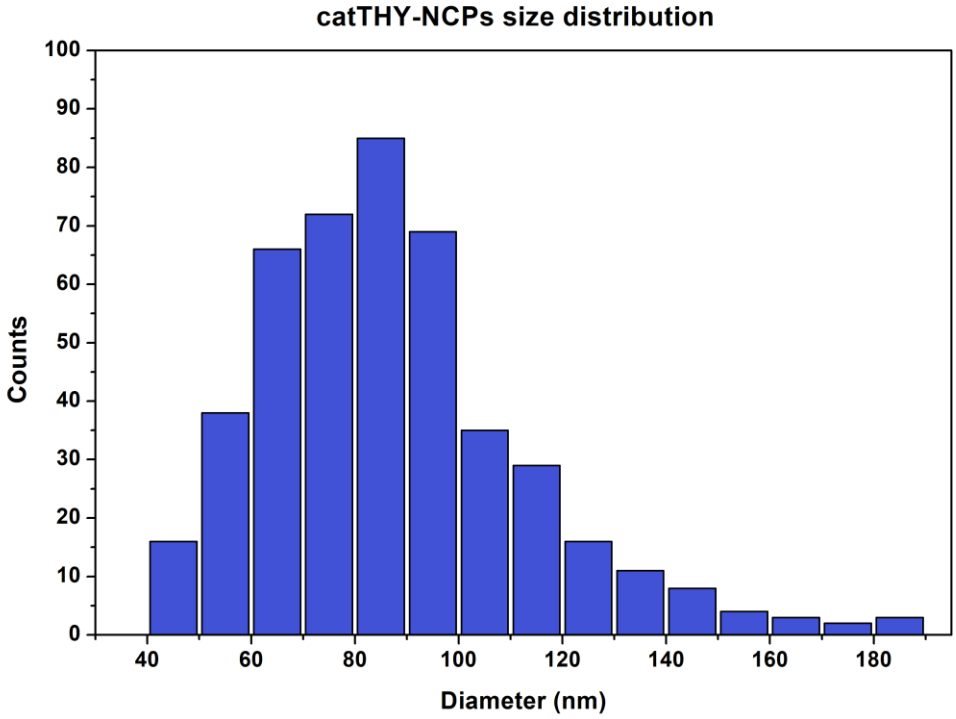


Figure S10. Histogram of catTHY-NCPs particle size extracted from SEM micrographs (457 particles, mean size 87 ± 26 nm).

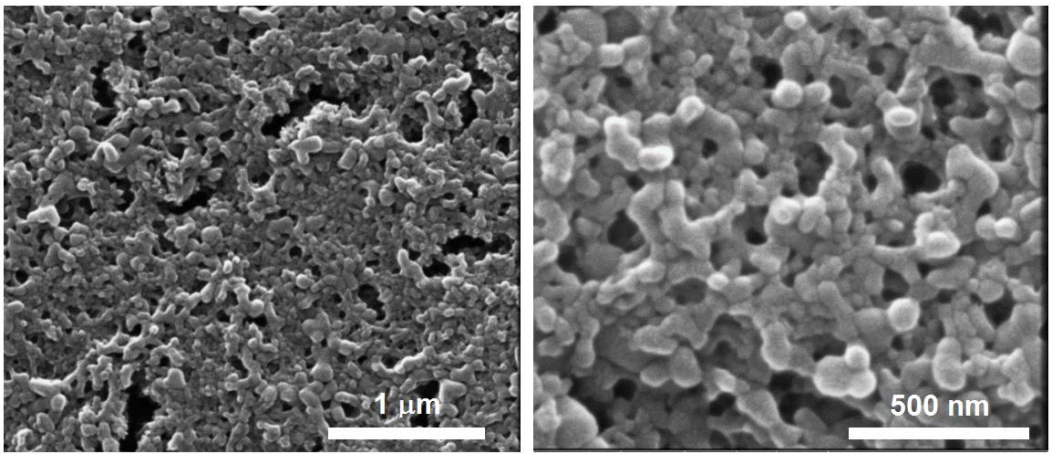


Figure S11. SEM images of catTHY-NCPs at different magnifications.

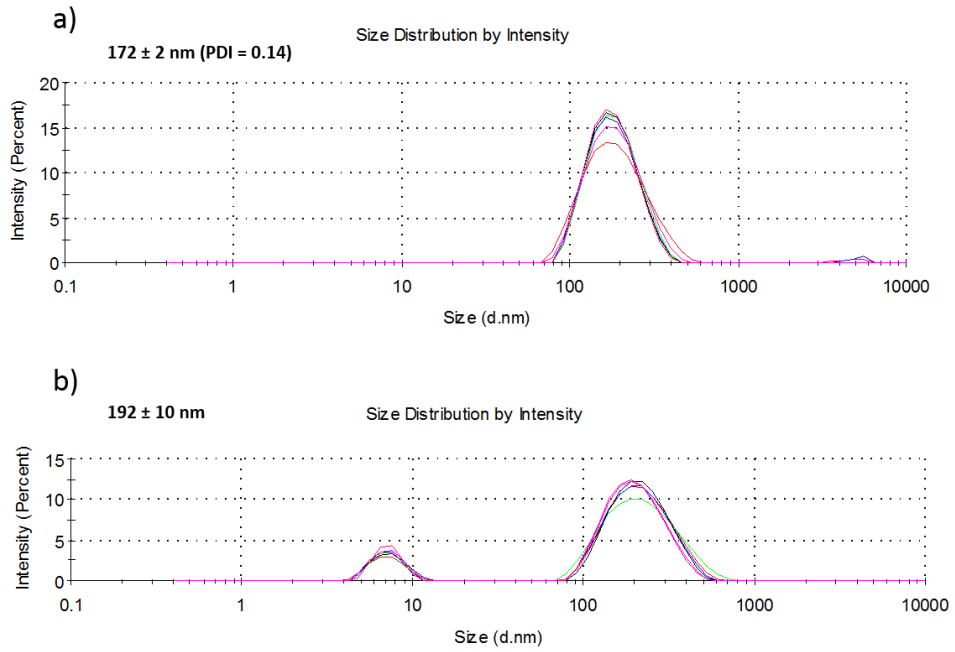


Figure S12. DLS measures of catTHY-NCPs particle size in a) ethanol and b) PBS/BSA 0.5 mM.

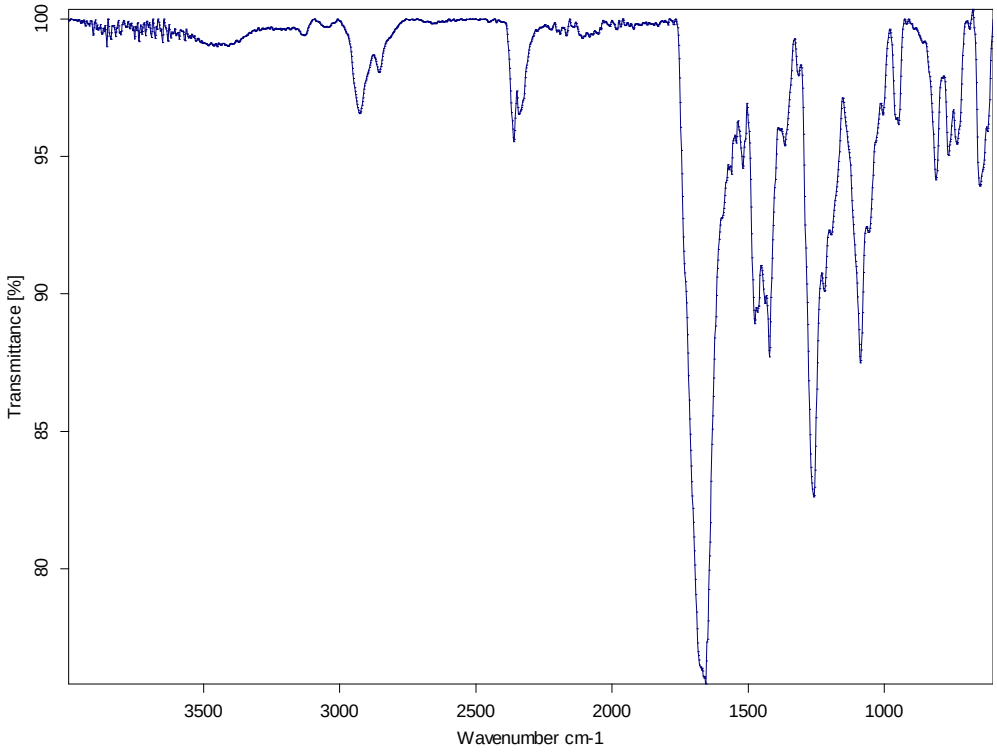


Figure S13. FT-IR spectrum of catTHY-NCPs.

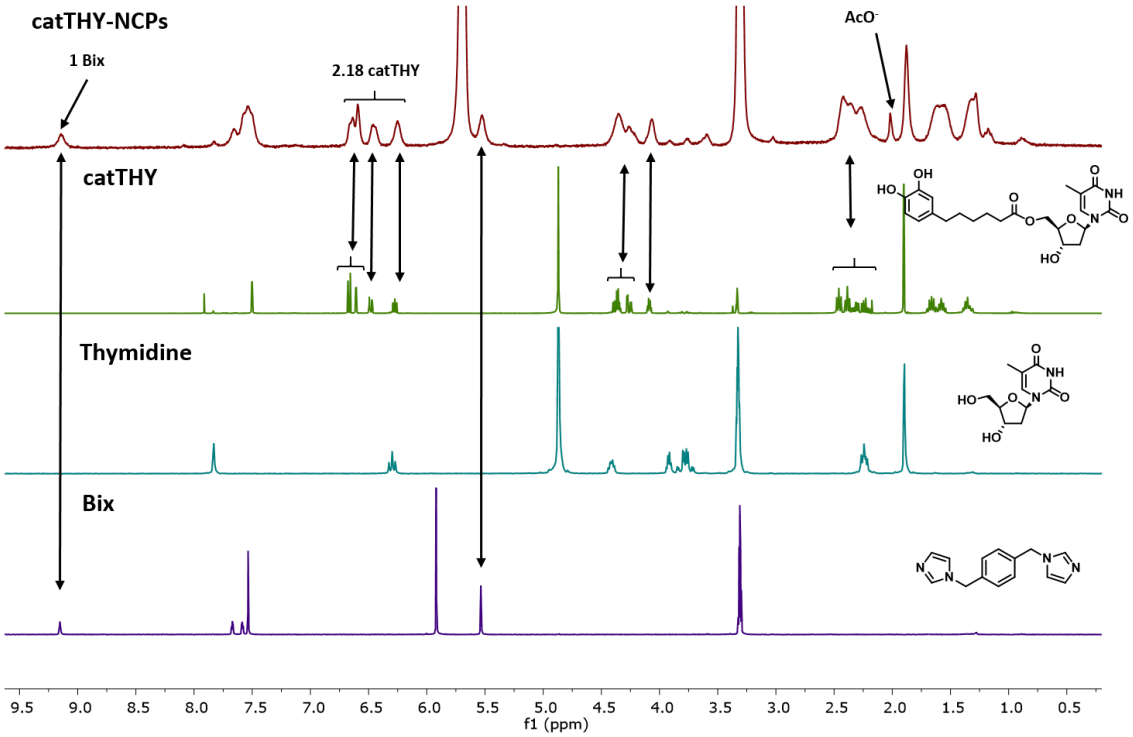


Figure S14. ^{1}H NMR spectra of (top to bottom) catThy-NCPs, catThy, thymidine and Bix. All spectra were recorded in DCl/CD_3OD (50 µL DCl / mL CD_3OD). Peaks corresponding to catThy, Bix and acetate are observed in the ^{1}H NMR spectrum of catThy-NCPs.

Table S2. Table of elemental analysis results for three different batches of catTHY-NCPs and comparison between the average and the proposed formula. The results indicate a good reproducibility of the synthetic method.

Batch	%C	%H	%N
1	45.22	4.92	6.64
2	45.18	4.85	6.91
3	45.59	4.96	6.86
Average	45.33	4.91	6.80
$Fe(catTHY)_{2.18}bix(AcO)_{0.7}(H_2O)_{20.2}$	45.48	6.82	7.0

S4. Representative chromatograms of MT-2 cell lysates incubated with AZT, catAZT and catAZT-NCPs

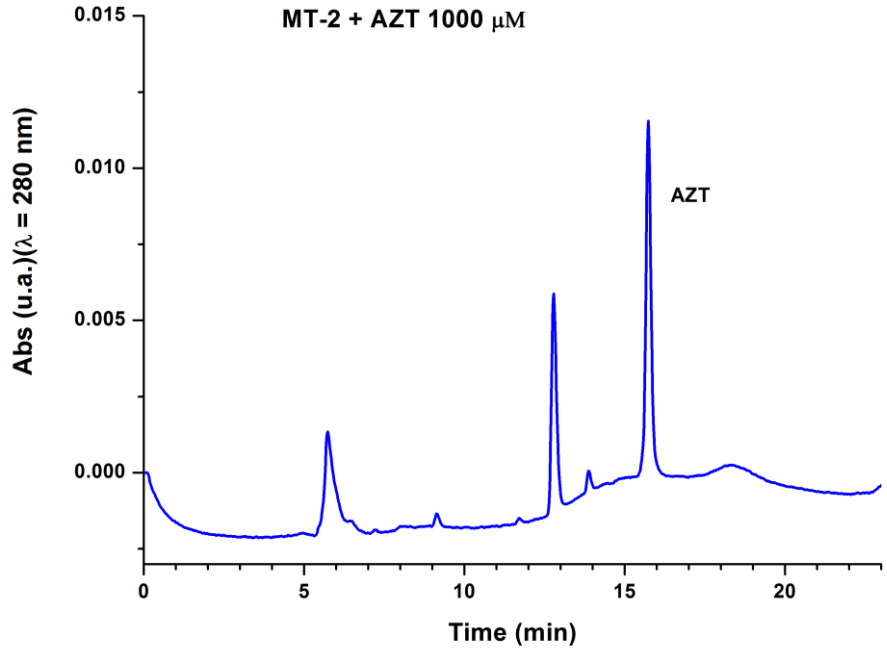


Figure S15. Chromatograms of MT-2 cell lysates incubated with AZT 1000 μM for 4 hours at 37 °C.

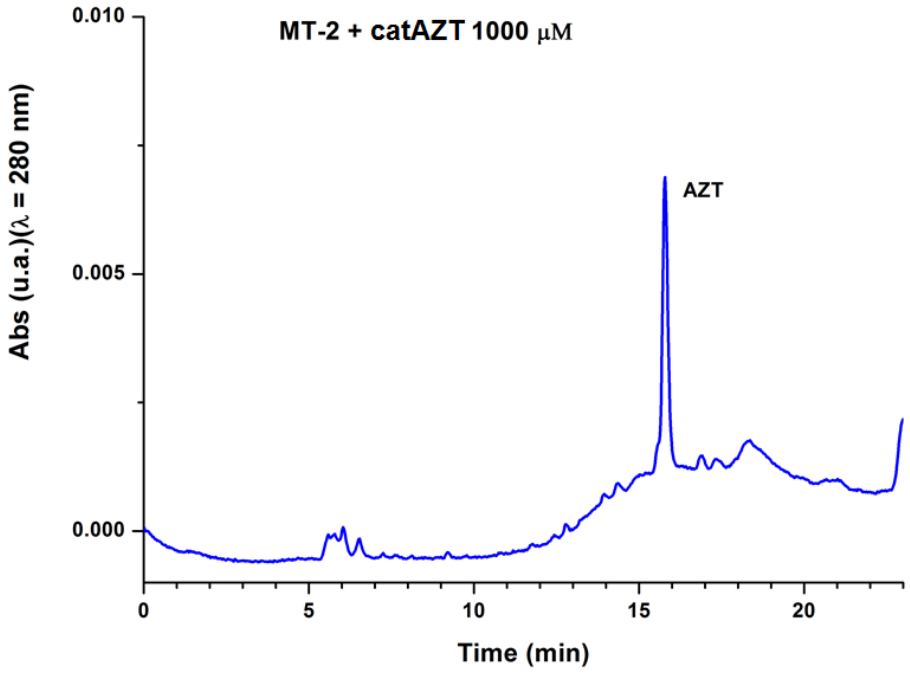


Figure S16. Chromatograms of MT-2 cell lysates incubated with catAZT 1000 μM for 4 hours at 37 °C.

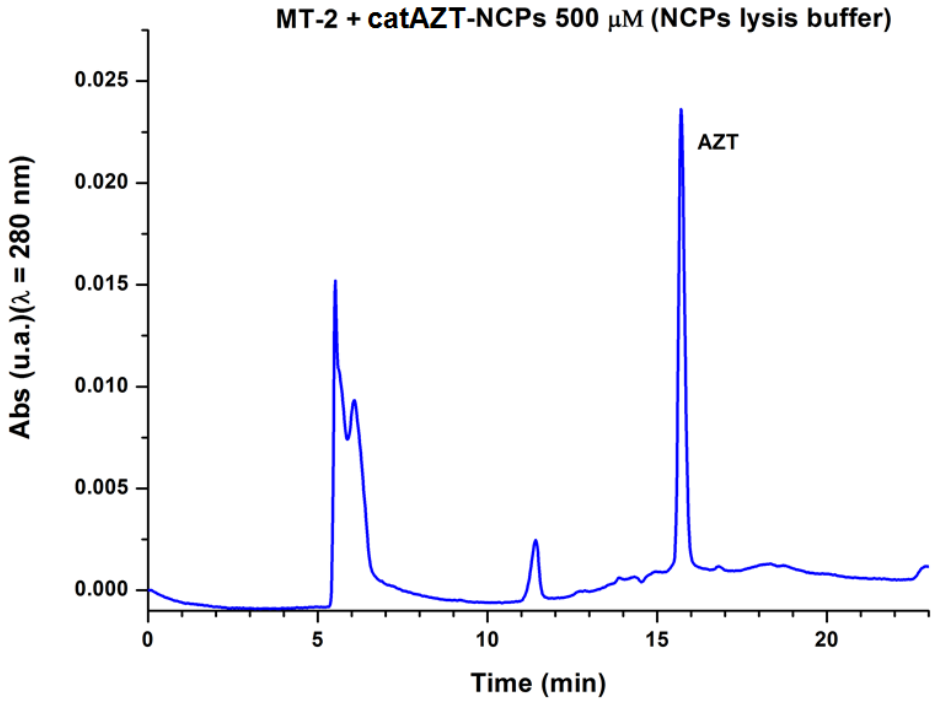


Figure S17. Chromatograms of MT-2 cell lysates incubated with catAZT-NCPs 500 μM for 4 hours at 37 °C.

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S5. Citotoxicity of AZT, catAZT and catAZT-NCPs against MT-2 cells

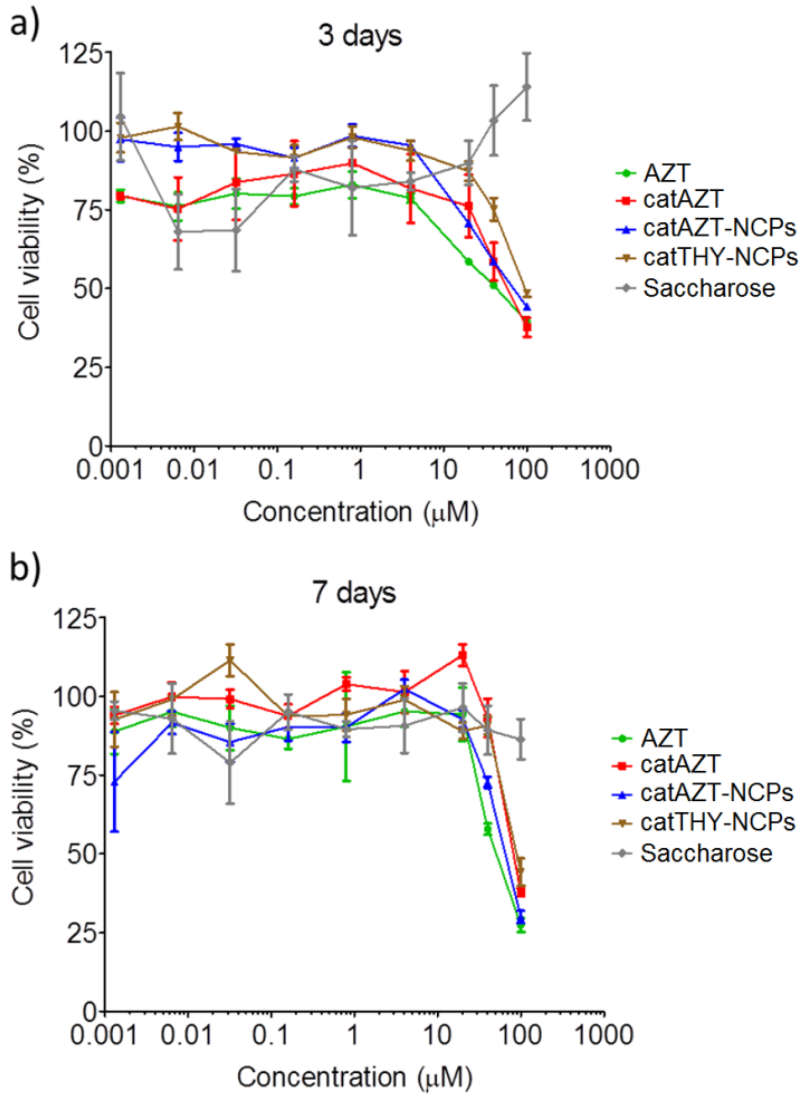

Figure S18. Cytotoxic effects of catAZT-NCPs against MT-2 lymphocytes. Cytotoxicity of catAZT-NCPs and equivalent concentrations of AZT, catAZT, catTHY-NCPs, and saccharose (vehicle) on MT-2 cell viability assessed by the MTT assay. Cell viability is expressed as percentage compared to an untreated control at a) 3 days or b) 7 days after applying the compounds. Values are shown as mean ± standard error of the mean (SEM) of two independent experiments performed in triplicate

Notes and references

1. P. K. Dhal and F. H. Arnold, *Macromolecules*, 1992, **25**, 7051-7059.
2. A. Adachi, H. E. Gendelman, S. Koenig, T. Folks, R. Willey, A. Rabson and M. A. Martin, *J. Virol.*, 1986, **59**, 284-291.
3. B. A. Larder, S. D. Kemp and D. J. M. Purifoy, *Proc. Natl. Acad. Sci. USA*, 1989, **86**, 4803-4807.
4. B. A. Larder, B. Chesebro and D. D. Richman, *Antimicrob. Agents Chemother.*, 1990, **34**, 436-441.