SUPPORTING INFORMATION

Facile Solid Phase Synthesis and Assessment of Nucleoside Analogs as Inhibitors of Bacterial UDP-Sugar Processing Enzymes

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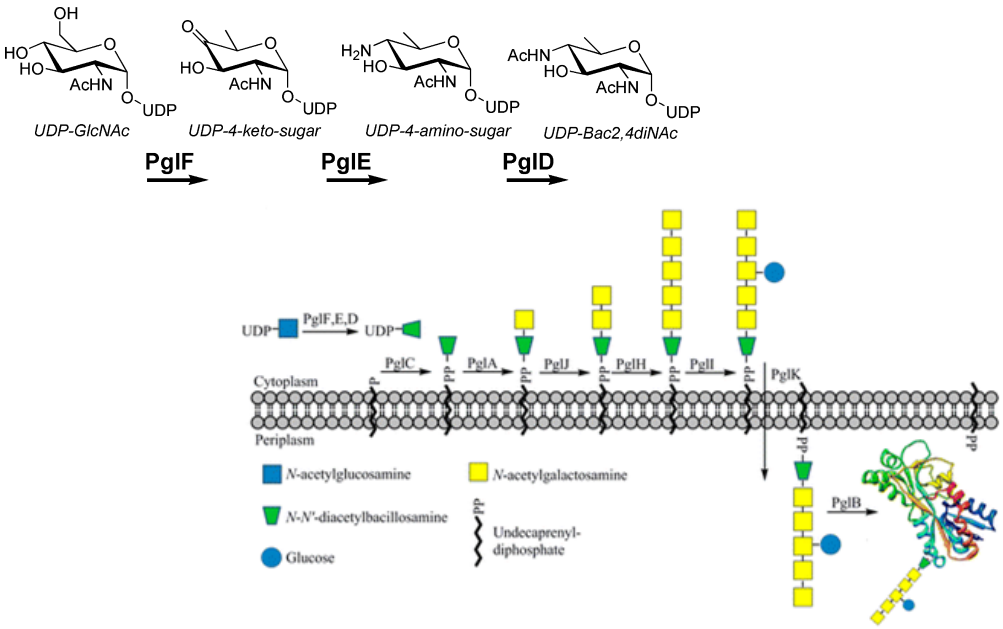
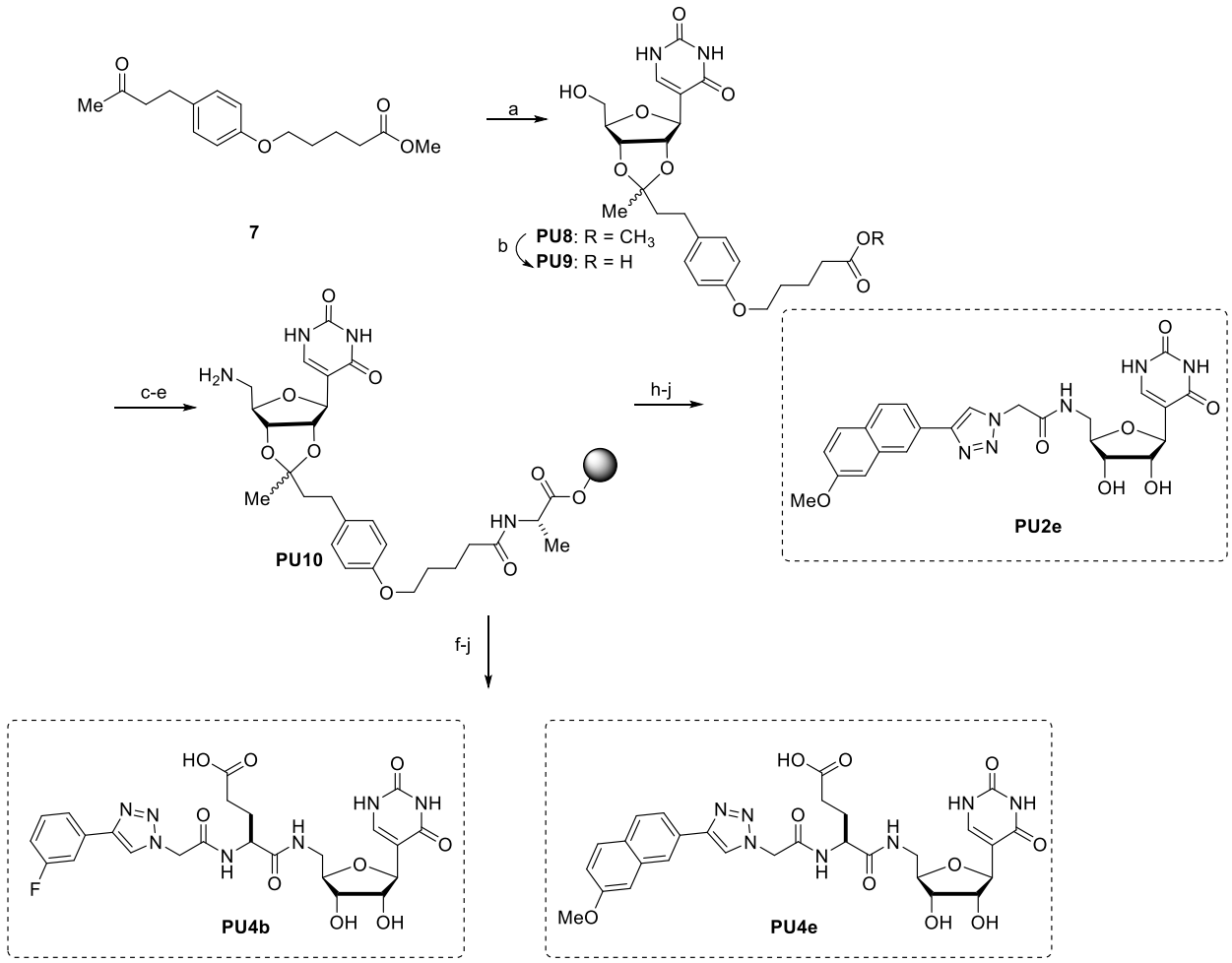
Supplementary Figure S1. N-linked protein glycosylation pathway in *C. jejuni.*

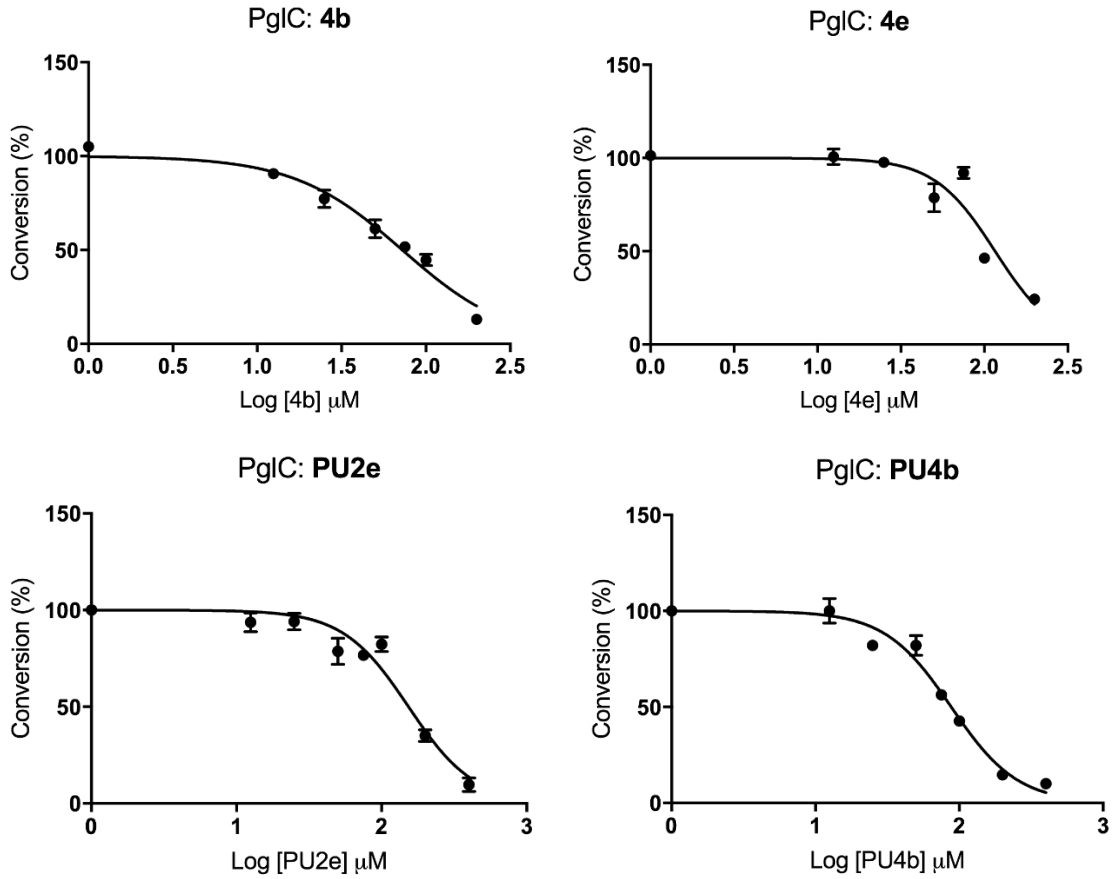


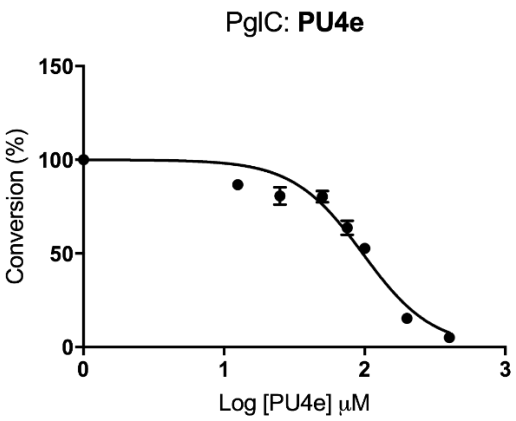
Supplemental Scheme 1. Synthesis of common pseudouridinyl intermediate PU10 and pseudouridine compounds PU2e, PU4b and PU4e.[a]

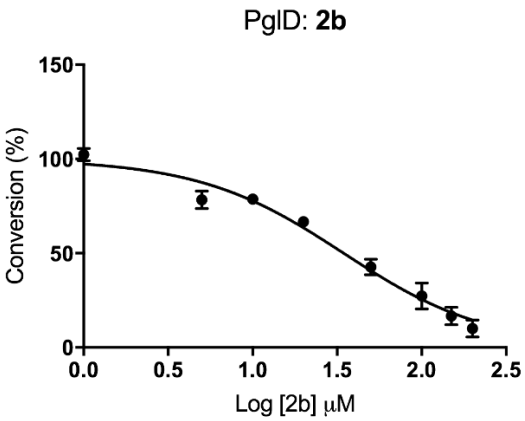


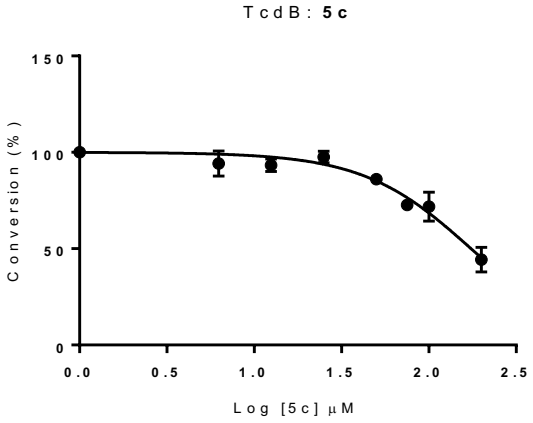
[a]Reagents and conditions: (a) i: trimethylorthoformate, *p*-TsOH, MeOH; ii: Pseudouridine, 2M HCl in dioxane, DMF; (b) LiOH, MeOH/H_2O, 61%, 2 steps; (c) H-Ala-Wang resin, HBTU, DIPEA, DMF; (d) DIAD, PPh_3, tetrachlorophthalimide, DMF/THF; (e) EDA, DMF. (f) Fmoc-Glu(OtBu)-OH, HBTU, DIPEA, DMF; (g) 20% piperidine in DMF; (h) 2-Azido-acetic acid, HBTU, DIPEA, DMF; (i) THPTA, $CuSO_4$, sodium ascorbate, alkyne-R; (j) 95/2.5/2.5 TFA:TIS:H_2O;

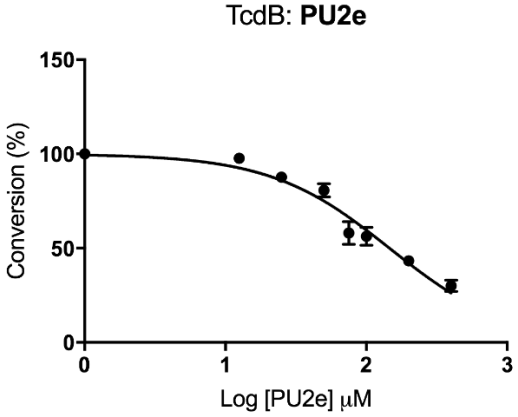
Supplementary Figure S2. IC_{50} curves of select inhibitors, measured by UMP-Glo monitoring luminescence (PglC), CoASH release using DTNB (PglD), or UDP-Glo monitoring luminescence (TcdB). Percent inhibition was determined after pre-incubation with inhibitor as described in the main text, in reference to control with no inhibitor. Error bars indicate mean ± SD; n=3. Generated using GraphPad Prism.



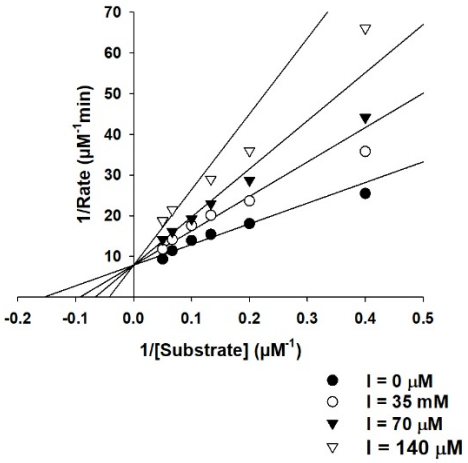

PglC: **PU4e**
Conversion (%)
150
100
50
0
0
1
2
3
Log [PU4e] μM


PglD: **2b**
Conversion (%)
150
100
50
0
0.0
0.5
1.0
1.5
2.0
2.5
Log [2b] μM

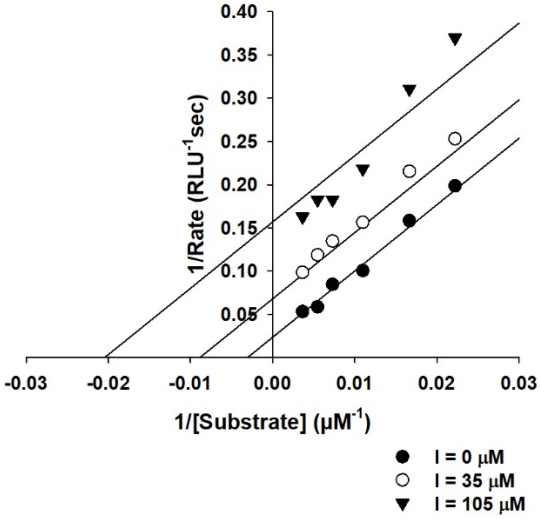

TcdB: **5c**
Conversion (%)
150
100
50
0
0.0
0.5
1.0
1.5
2.0
2.5
Log [5c] μM


TcdB: **PU2e**
Conversion (%)
150
100
50
0
0
1
2
3
Log [PU2e] μM

Supplementary Figure S3. Lineweaver-Burk plots of PglC with compound **4b** and UDP-diNAcBac, The substrate conversion was monitored by UMP-Glo luminescence assay. Generated using SigmaPlot.



Supplementary Figure S4. Lineweaver-Burk plots of PglD with compound **2b** and UDP-4-amino-NAcBac. The substrate conversion was monitored by measure of CoASH release using DTNB. Generated using SigmaPlot.



Supplementary Table 1. Kinetic inhibition data of PglD with 100 μM of **2b** and various concentrations of AcCoA.

Conditions	Measured PglD inhibitions
300 μM of AcCoA, 274 uM of 4-aminosugar (standard)	**61 ± 5.9%**
600 μM of AcCoA, 274 uM of 4-aminosugar	**58 ± 3.5%**
1500 μM of AcCoA, 274 uM of 4-aminosugar	**63 ± 3.4%**

Supplementary Figure S5. Lineweaver-Burk plot of TcdB with compound **5c** and UDP-glucose, The substrate conversion was monitored by UDP-Glo luminescence assay. Generated using SigmaPlot.

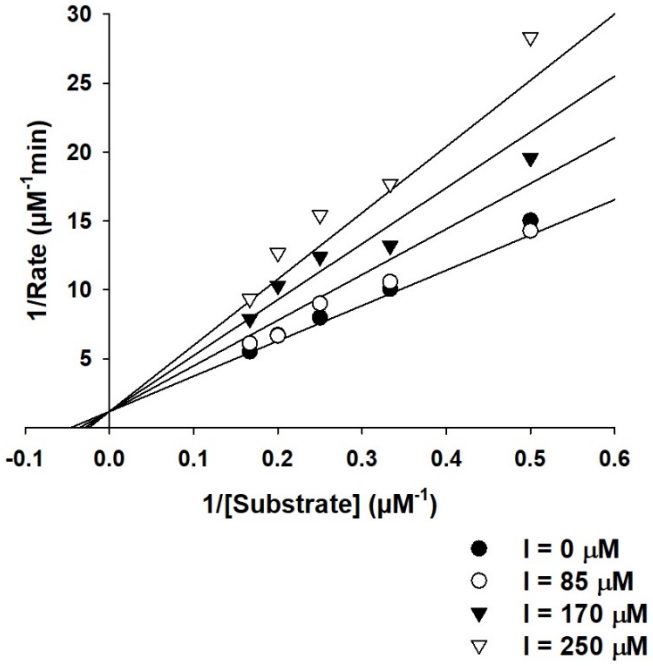
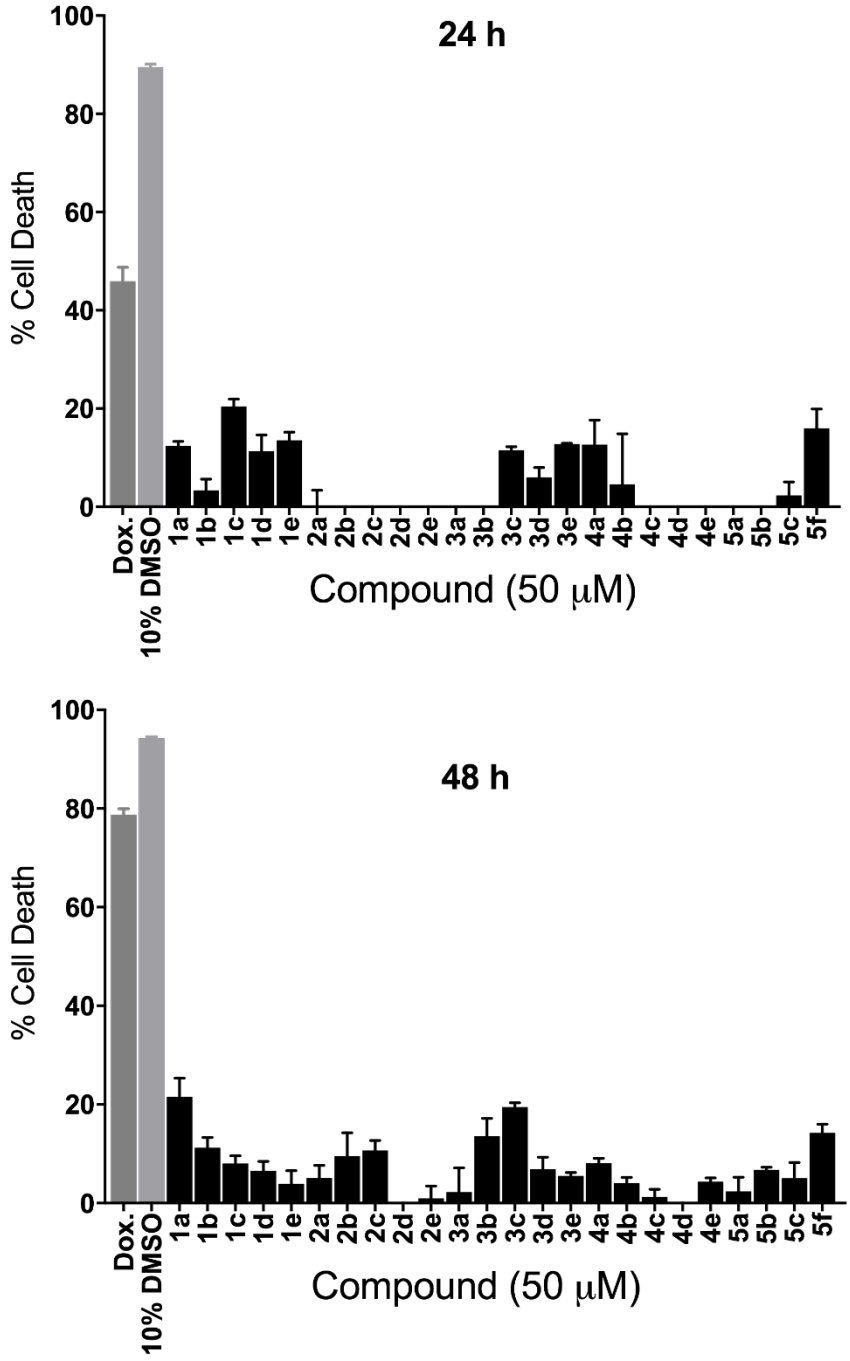


Figure S6. Luminescence values of 20,000 cells/well of IMR-90 lung fibroblasts using Cell-Titer Glo Cell Viability Assay at 24 h and 48 h, after incubation at 37°C in 5% CO_2 incubator with 50 μM of nucleoside analogs **1-5** in growth media with 0.5% DMSO. Controls for cell toxicity were either 1.7 μM doxorubicin hydrochloride or 10% DMSO in complete media. Error bars indicate mean ± SD; n=3.



Cloning, expression, and purification of TcdB-GTD.

The gene for the glycosyl transferase domain of TcdB (TcdB-GTD) from *Clostridium diffficile* in a pUC57 vector was purchased from Genscript. using AAAAAAGGATCCATGAGTTTAGTTA ATAGAAAACAGTTAGAAAAAATG and AAAAAACTCGAGTCAAAGAGAA CCTTCAAAATAATTCC as primers, the gene was amplified and cloned into a modified pET30b(+) vector containing an N-terminal His_7 tag and a TEV protease cleavage site. TcdB-GTD was expressed in *E. coli* grown to an OD_{600} of 0.6 with Isopropyl β-D-1-thiogalactopyranoside (IPTG) induction, and cells were harvested by centrifugation (3200 x g for 30 min) and stored at -80 °C until needed. Cells were resuspended in buffer containing 50 mM sodium phosphate pH 7.3, 400 mM NaCl, 5 mM β-mercaptoethanol, and 10 mM imidazole; and tumbled with 4 µL protease inhibitor cocktail and 20 mg lysozyme for 15 min. 10 µg/mL DNaseI was added, followed by lysis: sonication by pulsing 2 x for 90 s each at 40% amplitude. Lysate was cleared by centrifugation (100,000 x g for 50 min) and added to 3 mL of Ni-NTA resin in a chromatography column at a rate of 1 mL/min. Resin was washed with a gradient of 10-100 mM imidazole buffer, followed by elution with 300 mM imidazole lysis buffer, and fractions containing the purified protein were pooled after analysis by SDS-PAGE. Overnight dialysis was conducted in 20 mM HEPES pH 7.3, 150 mM NaCl, 5 mM DTT, with addition of a 500 µL aliquot of 100 µM His-tagged TEV protease. Cleaved protein was purified away from the tag by Ni-NTA chromatography by collecting the column flow through, and purity determined by SDS-PAGE gel. Purified enzyme was aliquoted, flash frozen, and stored at -80 °C.

Experimental Details.

All standard chemicals and reagents were purchased from Sigma-Aldrich and VWR unless otherwise noted. The following is a list of sources of other key reagents and consumable materials used in these studies. Fmoc-Ala-Wang resin (100-200 mesh, 0.6 mmol/g) was purchased from Bachem (4027342.0005); 3,4,5,6-tetrachlorophthalimide (TCP) was purchased from TCI-America (1571-13-7); Pseudouridine was purchased from Berry & Associates, Inc.(PYA 11080); 50 mL Torvic solid phase plastic reaction vessels (Torvic).

The solid-phase reactor was shaken using an IKA VXR basic vibrax. Compounds **1a-e**, **2a-e**, **3a-e**, **4a-e**, **5a-c**, **5f** were fully characterized by ^{1}H and ^{13}C NMR and HRMS. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker 401 MHz NMR spectrometer. Chemical shifts (δ) are reported in ppm relative to the residual proton in deuterated DMSO ($(CD_3)_2SO$) at 2.50 ppm for ^{1}H and 39.52 ppm for ^{13}C. HPLC purification was performed on a Waters 1525 binary HPLC mounted with a 00G-4252-PO-AX (LUNA Co.) preparative column, using a MeCN:H_2O with 0.1 % TFA gradient. Resin-bound intermediates were characterized by cleavage of a sample of the resin followed by analytical reverse-phase LC-MS using an Agilent Series 1100 HPLC equipped with a YMC AQ12S03-1003 wt C18 column and a Finnigan LCQ Deca electrospray ionization mass spectrometer, using a gradient of 5–95% acetonitrile in water with 0.1% TFA over 15 min. High-resolution MS spectra were collected using direct analysis in real time (DART) ionization on a Bruker Daltonics APEXIV 4.7 T Fourier transform ion cyclotron resonance mass spectrometer (FT-ICR-MS).

General Procedure A for analysis of resin-bound intermediates

To cleave an analytical sample of resin-bound intermediate, a sample of resin was transferred from the main reactor to a 1.5-mL Eppendorf containing 0.5 mL of TFA:TIPS (95:5), and shaken for 30 min at room temperature. The solvent was removed under a flow of nitrogen. The resulting residue was dissolved in CH_3CN and the polystyrene beads of the resin were filtered using a syringe filter. The crude mixture was then submitted to LC-MS analysis.

General Procedure B: Synthesis of 1a-e

To a round-bottom flask was added *N*-α-azido-Lys(Boc)-OH (735 mg, 2.70 mmol) and HBTU (1.03 g, 2.70 mmol) and 30 mL of DMF. DIPEA (704 µL, 4.05 mmol) was slowly added to the reaction mixture, which was reacted for 5 min at room temperature before being added to the solid-phase reactor containing **10** (1.35 mmol). The reaction was carried out for 3 h at room temperature. The solvent was removed and the resin was washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{38}H_{55}N_8O_{12}$ $[M+H]^+$: 815.39, found: 815.42. The resin was equally divided in five solid phase reactors each containing 0.27 mmol of resin. In a scintillation vial was added 135 µL of a 0.2 M solution of $CuSO_4$ in H_2O, and 270 µL of a 0.2 M THPTA solution in H_2O. 2 mL of DMF was added and the solution was transferred to the solid phase reactor. Then 216 µL of 0.2 M sodium ascorbate in H_2O was diluted in 2 mL of DMF and added to the resin. Finally, the corresponding alkyne (1.35 mmol) was dissolved in 2 mL of DMF and added on the resin. The resin was shaken overnight, solvent was removed, and the resin was washed three times with 5 mL of DMF and three times with 5 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. To cleave the compounds from the resin, 3.0 mL of a TFA:TIPS:H_2O (95:2.5:2.5) were added and the reactor was shaken for 1 h. The flow through was collected and the resin washed 3 times with 5 mL of CH_2Cl_2. The same cleavage reaction was repeated once. Both washes were combined and concentrated *in vacuo*. The crude residue was precipitated in cold diethyl ether, centrifuged the supernatant removed and the solid residue was resuspended and purified by reverse phase HPLC.

General Procedure C: Synthesis of 2a-e

To a round-bottom flask was added 2-azidoacetic acid (303 µL, 4.05 mmol) and HBTU (1.53 g, 4.05 mmol) in 30 mL of DMF. DIPEA (673 µL, 3.78 mmol) was slowly added to the mixture, and reacted 5 min at room temperature before being added to the solid-phase reactor containing **10** (1.35 mmol). The reaction was carried out for 3 h at room temperature. The solvent was removed and the resin was washed three times

with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{29}H_{38}N_7O_{10}$ $[M+H]^+$: 644.26, found: 644.75. The resin was equally divided in five solid phase reactors each containing 0.27 mmol of resin. In a scintillation vial was added 135 µL of a 0.2 M solution of $CuSO_4$ in H_2O, and 270 µL of a 0.2 M THPTA solution in H_2O. 2 mL of DMF was added and the solution was transferred to the solid phase reactor. Then 216 µL of 0.2 M sodium ascorbate in H_2O was diluted in 2 mL of DMF and added to the resin. Finally, the corresponding alkyne (1.35 mmol) was dissolved in 2 mL of DMF and added on the resin. The resin was shaken overnight, solvent was removed, and the resin was then washed three times with 5 mL of DMF and three times with 5 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. To cleave the compounds from the resin, 3.0 mL of a TFA:TIPS:H_2O (95:2.5:2.5) were added and the reactor was shaken for 1 h. The flow-through was collected and the resin washed 3 times with 5 mL of CH_2Cl_2. The same cleavage reaction was repeated once. Both washes were combined and concentrated *in vacuo*. The crude residue was precipitated in cold diethyl ether, centrifuged the supernatant removed and the solid residue was resuspended and purified by reverse phase HPLC.

General Procedure D: Synthesis of 3a-e

To a round-bottom flask was added *N*-α-Fmoc-*N*-ε-Boc-L-lysine (3.18 g, 6.8 mmol) and HBTU (2.57 g, 6.8 mmol) in 30 mL of DMF. DIPEA (2.35 mL, 13.5 mmol) was slowly added to the reaction**,** and reacted 5 min at room temperature before being added to the solid-phase reactor containing **10** (1.35 mmol). The reaction was carried out for 3 h at room temperature. The solvent was removed and the resin was washed three times with 20 mL of DMF and three times with 20 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{53}H_{67}N_6O_{14}$ $[M+H]^+$: 1011.47, found: 1011.05 The solid-phase reactor was filled with 20 mL of 20% solution of 4-methylpiperidine in DMF. The reactor was shaken for 20 min at room temperature. The resin was subsequently washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. To a round

bottom flask was added 2-azidoacetic acid (303 µL, 4.05 mmol) and HBTU (1.53 g, 4.05 mmol) in 30 mL of DMF. DIPEA (673 µL, 3.78 mmol) was slowly added to the mixture, and reacted for 5 min at room temperature before being added to the solid-phase reactor. The reaction was carried out for 3 h at room temperature. The solvent was removed and the resin was washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{40}H_{58}N_9O_{13}$ $[M+H]^+$: 872.41, found: 872.58. The resin was equally divided in five solid phase reactors each containing 0.27 mmol of resin. In a scintillation vial was added 135 µL of a 0.2 M solution of $CuSO_4$ in H_2O, and 270 µL of a 0.2 M THPTA solution in H_2O. 2 mL of DMF was added and the solution was transferred to the solid phase reactor. Then 216 µL of 0.2 M sodium ascorbate in H_2O was diluted in 2 mL of DMF and added to the resin. Finally, the corresponding alkyne (1.35 mmol) was dissolved in 2 mL of DMF and added on the resin. The resin was shaken overnight, solvent was removed, and the resin was washed three times with 5 mL of DMF and three times with 5 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. To cleave the compounds from the resin, 3.0 mL of a TFA:TIPS:H_2O (95:2.5:2.5) were added and the reactor was shaken for 1 h. The flow-through was collected and the resin washed 3 times with 5 mL of CH_2Cl_2. The same cleavage reaction was repeated once. Both washes were combined and concentrated *in vacuo*. The crude residue was precipitated in cold diethyl ether, centrifuged the supernatant removed and the solid residue was resuspended and purified by reverse phase HPLC.

General Procedure E: Synthesis of 4a-e:

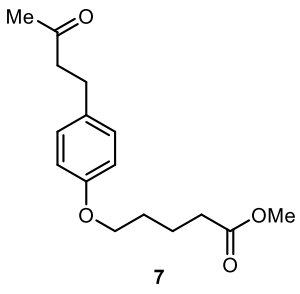
To a round-bottom flask was added *N*-α-Fmoc-Glu(OtBu)-OH (2.87 g, 6.8 mmol) and HBTU (2.57 g, 6.8 mmol) in 30 mL of DMF. DIPEA (2.35 mL, 13.5 mmol) was slowly added to the reaction**,** and reacted 5 min at room temperature before being added to the solid-phase reactor containing **10** (1.35 mmol). The reaction was carried out for 3 h at room temperature. The solvent was removed and the resin was washed three times with 20 mL of DMF and three times with 20 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{51}H_{62}N_5O_{14}$ $[M+H]^+$:

968.42, found: 968.97. The solid-phase reactor was filled with 20 mL of 20% solution of 4-methylpiperidine in DMF. The reactor was shaken for 20 min at room temperature. The resin was subsequently washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. To a round bottom flask was added 2-azidoacetic acid (303 µL, 4.05 mmol) and HBTU (1.53 g, 4.05 mmol) in 30 mL of DMF. DIPEA (673 µL, 3.78 mmol) was slowly added to the mixture, and reacted 5 min at room temperature before being added to the solid-phase reactor. The reaction was carried out for 3 h at room temperature. The solvent was removed and the resin was washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A $C_{38}H_{53}N_5O_{13}$ $[M+H]^+$: 829.37, found: 829.55. The resin was equally divided in five solid phase reactors each containing 0.27 mmol of resin. In a scintillation vial was added 135 µL of a 0.2 M solution of $CuSO_4$ in H_2O, and 270 µL of a 0.2 M THPTA solution in H_2O. 2 mL of DMF was added and the solution was transferred to the solid phase reactor. Then 216 µL of 0.2 M sodium ascorbate in H_2O was diluted in 2 mL of DMF and added to the resin. Finally, the corresponding alkyne (1.35 mmol) was dissolved in 2 mL of DMF and added on the resin. The resin was shaken overnight, solvent was removed, and the resin was washed three times with 5 mL of DMF and three times with 5 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. To cleave the compounds from the resin, 3.0 mL of a TFA:TIPS:H_2O (95:2.5:2.5) were added and the reactor was shaken for 1 h. The flow-through was collected and the resin washed 3 times with 5 mL of CH_2Cl_2. The same cleavage reaction was repeated once. Both washes were combined and concentrated *in vacuo*. The crude residue was precipitated in cold diethyl ether, centrifuged the supernatant removed and the solid residue was resuspended and purified by reverse phase HPLC.

General Procedure F: Synthesis of 5a-c and 5f:

To the solid-phase reactor containing **10** (1.08 mmol) was added a mixture of diethyl squarate (479 µL, 3.24 mmol) and Et_3N (757 µL, 5.4 mmol) in 10 mL of CH_2Cl_2. The reaction was carried out for 1 h at room temperature. The solvent was removed and the resin was washed three times with 5 mL of DMF and three

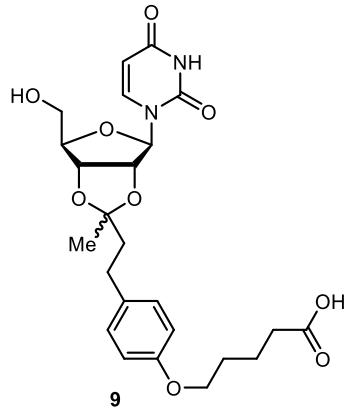
times with 5 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{33}H_{41}N_4O_{12}$ $[M+H]^+$: 685.27, found: 685.79. The resin was equally divided in four solid phase reactors, each charged with 0.27 mmol of resin. To each solid phase reactor was added the corresponding primary amine (3.0 equiv.) and Et_3N (189 µL, 1.35 mmol) in 5.0 mL of CH_2Cl_2. The resin was shaken for 2 h, then the solvent was removed and the resin was washed three times with 5 mL of DMF and three times with 5 mL of CH_2Cl_2 The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. To cleave the compounds from the resin, 3.0 mL of TFA:TIPS:H_2O (95:2.5:2.5) was added and the reactor, and was shaken for 1 h. The flow-through was collected and the resin washed 3 times with 5 mL of CH_2Cl_2. The same cleavage reaction was repeated once. Both flow-through were combined and concentrated *in vacuo*. Both washes were concentrated *in vacuo*. The crude residue was precipitated in cold diethyl ether, centrifuged the supernatant removed and the solid residue was resuspended and purified by reverse phase HPLC.



7

Methyl 5-(4-(3-oxobutyl)phenoxy)pentanoate (7).

To a solution of 4-(4-hydroxyphenyl)butan-2-one (5.0 g, 30.4 mmol), K_2CO_3 (8.4 g, 60.8 mmol) in 150 mL of DMF was added bromovaleric-methyl-ester (4.6 mL, 32.0 mmol). The reaction mixture was heated at 50 ℃ for 48 h. The reaction was then diluted in 300 mL of EtOAc and extracted successively with 200 mL $NaHCO_3$, water and brine. The organic layer was dried over Na_2SO_4 and concentrated *in vacuo*. The crude residue was purified by flash chromatography (20% EtOAc/Hex). The final product was obtained as a yellow oil in (8.1g, 96%). ^{1}H NMR (400 MHz, $CDCl_3$-d_6) [1] 7.09 (2H, d, J = 8.8 Hz, 2 ×Ar**H**), 6.81 (2H, d, J = 8.8 Hz, 2 ×Ar**H**), 4.01-3.85 (2H, m, O-C**H**$_2$-), 3.68 (3H, s, O-C**H**$_3$), 2.90-2.75 (2H, m, CH-C-C**H**$_2$), 2.74–2.65 (2H, m, C(O)-C**H**$_2$), 2.46-2.32 (2H, m, C**H**$_2$-C(O)-OCH_3), 2.13 (3H, s, C**H**$_3$-C(O)), 1.88-1.73 (4H, m, C**H**$_2$-C**H**$_2$-CH_2-C(O)-OCH_3); ^{13}C NMR (101 MHz, $CDCl_3$) δ 208.2, 173.9, 157.3, 132.9, 129.2, 114.4, 67.3, 51.5, 45.4, 33.7, 30.1, 28.9, 28.7, 21.6. HRMS (ESI) Exact mass calculated for: 279.1591, found: 279.1598

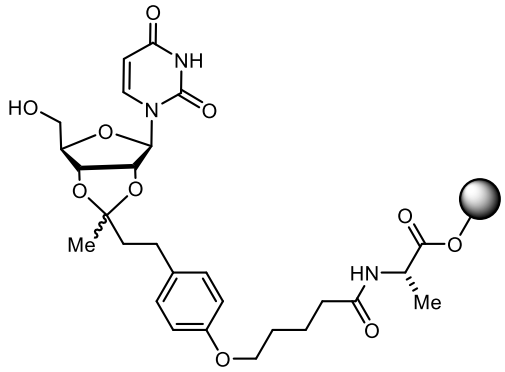


9

5-(4-(2-((3aR,4R,6R,6aR)-4-(2,4-Dioxo-3,4-dihydropyrimidin-1(2H)-yl)-6-(hydroxymethyl)-2-methyltetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoic acid (9)

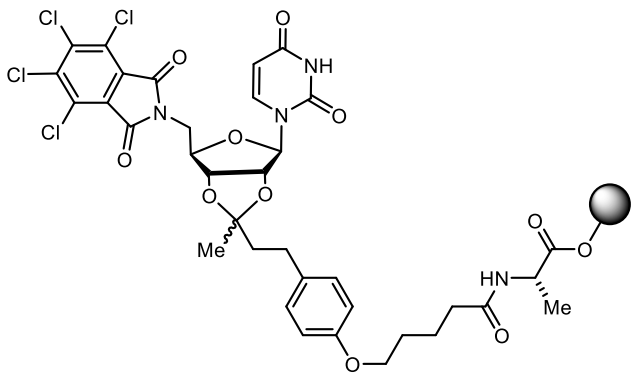
In an oven dried round-bottom flask, was added **7** (5.12 g, 18.4 mmol) along with *p*-TsOH (317 mg, 1.84 mmol). To the flask was added 100 mL of MeOH and trimethyl orthoformate (TMOF) (10.6 mL, 92 mmol). The reaction was flushed with N_2 and stirred at 50 ℃ for 14 h. The reaction was cooled to rt and quenched with 566 µL of Et_3N. The crude was concentrated under vacuum, resuspended in 200 mL of EtOAc and extracted twice with 200 mL of H_2O and once with 200 mL of brine. The organic layer was dried over Na_2SO_4 and concentrated *in vacuo*. The crude intermediate was obtained as a colorless oil and was used without other purification. The crude was dissolved in 10.0 mL of DMF and added to a round bottom flask previously filled with uridine (3.2 g, 13.10 mmol) and purged under N_2. This was followed by the addition of 25 mL of dioxane and 25 ml of 4.0 M HCl in dioxane. The reaction was stirred for 14 h at rt. After what it was cooled down to 0 ℃and quenched with 13.9 mL of Et_3N. The reaction was diluted in 200 mL of EtOAc and extracted three times with 200 mL of H_2O. The organic layer was dried over Na_2SO_4, filtered and concentrated *in vacuo* to yield compound **8** as an off-white foam. The aforementioned compound **8** was dissolved in 100 mL MeOH and 15 mL of H_2O to which was added LiOH (1.0 g, 41.70 mmol). The reaction was stirred at rt for 14 h and subsequently quenched with a 0.5 M HCl solution to pH 5. The solution was concentrated *in vacuo* to remove the methanol, diluted in 200 mL of H_2O and extracted three times with 200 mL of EtOAc. The organic layers were combined dried over Na_2SO_4 filtered and concentrated *in vacuo* to afford the crude residue. Purification of the residue by column chromatography (5% to 10% MeOH in $CHCl_3$) gave the title compound **9** as an off-white foam (6.26 g, 97%). The final product was obtained in a 1:1.07 inseparable mixture of diastereoisomers. The NMR spectrum is reported as the mixture without discrimination between diastereoisomers. ^{1}H NMR (400 MHz, DMSO-d_6) δ = 12.00 (1H, s, C(O)O**H**), 11.38 (1H, s, N**H**-C(O)-N), 7.79 (1H dd, *J* = 8.1, 7.2 Hz, C**H**-N-

C(O)), 7.11 (1H, dd, *J* = 10.3, 8.6 Hz, Ar**H**), 6.90-6.68 (1H, dd, *J* = 9.4, 8.2 Hz, Ar**H**), 5.88 (1H, dd, *J* = 6.3, 2.6 Hz, CH-O-C**H**-N), 5.64 (1H, dd, *J* = 8.1, 2.0 Hz, C**H**-C(O)-NH), 4.95 (1H, td, *J* = 5.9, 2.6, C**H**-CH-N), 4.80 (1H, app dt, *J* = 7.1, 3.8 Hz, CH_2-CH-C**H**), 4.11 (1H, app dq, *J* = 12.9, 4.3 Hz, 1H), 3.91 (2H, app q, *J* = 5.8 Hz, O-C**H**$_2$), 3.67-3.49 (2H, m, HO-C**H**$_2$), 2.72-2.61 (1H, m, C**H**$_A$CH$_B$-C-CH), 2.55-2.50 (1H, m, CH$_A$C**H**$_B$-C-CH), 2.27 (2H, td, *J* = 7.2, 2.4 Hz, -C**H**$_2$-C(O)OH), 2.02-1.94 (1H, m, C-C**H**$_A$CH$_B$-CH$_2$), 1.85-1.75 (1H, m, C-CH$_A$C**H**$_B$-CH$_2$), 1.74-1.58 (4H, m, -CH$_2$-C**H**$_2$-C**H**$_2$), 1.50 (1.45 H, s, -C**H**$_3$), 1.30 (1.55 H, s, -C**H**$_3$); Note: the signal at 2.55-2.50 is partially obscured by the residual water signal; ^{13}C NMR (101 MHz, DMSO) δ 174.6, 163.4, 157.0, 157.0, 150.6, 150.6, 142.3, 142.2, 133.6, 129.3, 114.8, 114.6, 114.5, 114.4, 102.0, 91.5, 91.4, 87.0, 86.7, 84.3, 83.8, 81.1, 80.8, 67.2, 61.6, 41.3, 40.7, 33.5, 29.4, 28.7, 28.4, 25.3, 23.9, 21.5, 21.3. HRMS (ESI) Exact mass calculated for 491.2056, found: 491.1285



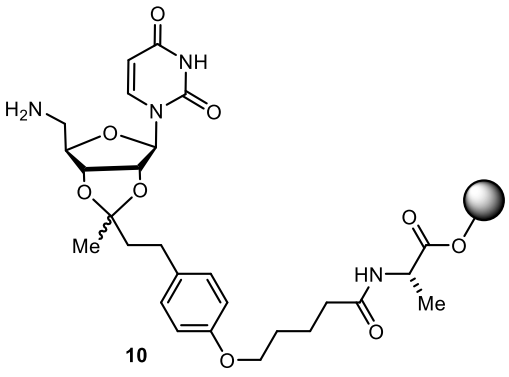
Resin bound (5-(4-(2-((3a*R*,4*R*,6*R*,6a*R*)-4-(2,4-Dioxo-3,4-dihydropyrimidin-1(2H)-yl)-6-(hydroxymethyl)-2-methyltetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine

The Fmoc-Ala-Wang resin (5.0 g, 3.0 mmol) was introduced in a solid phase synthesis glass reactor. The resin was swelled in 50 mL of CH_2Cl_2 for 20 min. The solvent was removed. The solid-phase reactor was filled with 40 mL of 20% solution of 4-methylpiperidine in DMF. The reactor was shaken for 20 min at room temperature. The resin was subsequently washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. To a scintillation vial were introduced uridine compound **9** (1.12 g, 3.73 mmol) and HBTU (1.41 g, 3.73 mmol) which were dissolved in 20 mL of DMF. DIPEA (1.56 mL, 9.0 mmol) was slowly added to the reaction mixture containing **9** and HBTU, which was allowed to react 5 min at room temperature before being added to the solid-phase reactor. The reaction was carried out for 45 min at room temperature. The solvent was removed and the resin was subsequently washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2.



Resin bound (5-(4-(2-((3a*R*,4*R*,6*R*,6a*R*)-4-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-2-methyl-6-((4,5,6,7-tetrachloro-1,3-dioxoisoindolin-2-yl)methyl)tetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine

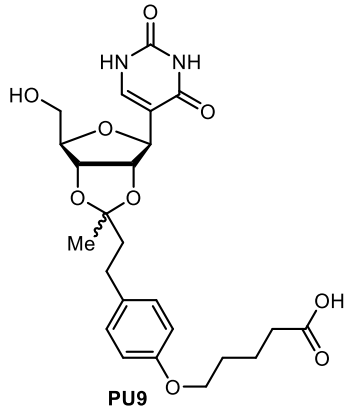
PPh_3 (2.10 g, 8.0 mmol) was added to a round bottom flask and dissolved in 30 mL of a 2:1 mixture of THF:DMF. To the solution was added DIAD (1.57 mL, 8.0 mmol). The reaction was stirred for 5 min upon which was added TCP (2.28 g, 8.0 mmol). The reaction was further stirred for an additional 5 min before being added to the solid phase reactor containing 2.0 mmol of the resin bound (measured by mass % of the total mass resin) **(5-(4-(2-((3a*R*,4*R*,6*R*,6a*R*)-4-(2,4-Dioxo-3,4-dihydropyrimidin-1(2H)-yl)-6-(hydroxymethyl)-2-methyltetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine.** The reaction was shaken for 14 h, after which the solvent was removed and the resin was washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction was resubmitted for 3 h with fresh reagents. The solvent was removed and the resin was subsequently washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{35}H_{35}Cl_4N_4O_{11}$ $[M+H]^+$: 828.10, found: 828.87



10

Resin bound (5-(4-(2-((3a*R*,4*R*,6*R*,6a*R*)-4-(aminomethyl)-6-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-2-methyltetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine (10)

To the solid phase reactor containing **resin bound (5-(4-(2-((3a*R*,4*R*,6*R*,6a*R*)-4-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-2-methyl-6-((4,5,6,7-tetrachloro-1,3-dioxoisoindolin-2-yl)methyl)tetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine** (2.0 mmol) was added ethylenediamine (334 µL, 5.0 mmol) in 30 mL of DMF. The reaction was shaken for 2 h. The solvent was removed and the resin was resubmitted to ethylenediamine (334 µL, 5.0 mmol) for another 2 h. The solvent was removed and the resin was washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{27}H_{37}N_4O_9$ $[M+H]^+$: 561.25, found: 561.20

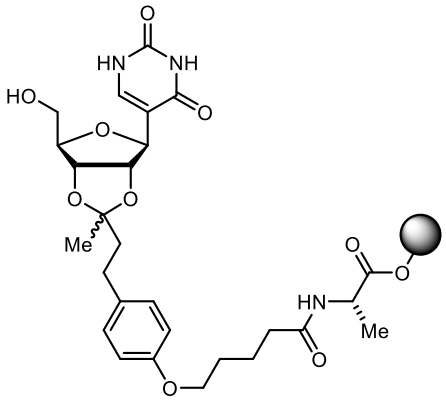


PU9

5-(4-(2-((3a*S*,4*S*,6*R*,6a*R*)-4-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-6-(hydroxymethyl)-2-methyltetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoic acid (PU9)

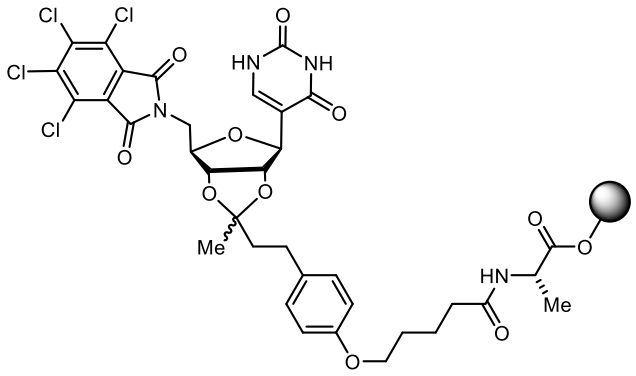
In an oven dried round-bottom flask, under N_2 was added **7** (2.12 g, 7.63 mmol) along with *p*-TsOH (263 mg, 1.52 mmol). To the flask was added 50 mL of MeOH and TMOF (4.17 mL, 38.2 mmol). The reaction was stirred at 50 ℃ for 14 h. The reaction was cooled to RT and neutralized with 300 µL of Et_3N. Then the solution was concentrated *in vacuo*, the crude residue was resuspended in 100 mL of EtOAc and extracted twice with 100 mL of H_2O and once with 100 mL of brine. The organic layer was dried over Na_2SO_4 and concentrated *in vacuo*. The crude product was obtained as a colorless oil and was used without other purification. The crude oil was dissolved in 10 mL of DMF and added to a

round bottom flask containing, previously filled with pseudouridine (800 mg, 3.27 mmol) and purged under N_2. This was followed by the addition of 25 mL of dioxane and 25 ml of 4.0 M HCl in dioxane. The reaction was stirred for 14 h at RT, then cooled to 0 ℃ and quenched with 5.0 mL of Et_3N. The crude reaction was diluted in 200 mL of EtOAc and extracted twice with 100 mL of H_2O and once with 100 mL of brine. The organic layer was dried over Na_2SO_4 and concentrated *in vacuo* to yield compound **PU8** as an off-white foam. The aforementioned compound **PU8** was dissolved in 80 mL MeOH and 20 mL of H_2O to which was added LiOH (2.2 g, 96 mmol). The reaction was stirred at RT for 14 h and subsequently quenched with a 0.5 M HCl solution to pH 5. The solution was concentrated *in vacuo* to remove the methanol, diluted in 50 mL of H_2O and extracted three times with 100 mL of EtOAc. The organic layers were combined dried over Na_2SO_4 filtered and concentrated *in vacuo* to afford the crude residue. Purification of the residue by column chromatography (6% to 15% MeOH in $CHCl_2$) gave the title compound **PU9** as an off-white foam (976 g, 61%). The final product was obtained in a 5:1 inseparable mixture of diastereoisomers. The NMR spectrum is reported as the mixture without discrimination between diastereoisomers. δ = 12.17 (1H, br s, C(O)O**H**), 11.16 (1H, d, *J* = 2.0 Hz, C(O)-N**H**-C(O)), 10.91 (1H, dd, *J* = 5.9, 2.0 Hz, CH-N**H**-C(O)), 8.64 (1H, d, *J* = 8.2 Hz, C(O)-N**H**-CH), 8.55 (1H, s, C**H**-N-N)), 8.34 (1H, d, *J* = 1.5 Hz, Ar**H**), 8.19 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH_2), 7.95 (1H, dd, *J* = 8.5, 1.7 Hz, Ar**H**), 7.88 (2H, app d, *J* = 7.8 Hz, Ar**H**), 7.43 (1H, d, *J* = 5.9 Hz, C**H**-C-C(O)), 7.34 (1H, d, *J* = 2.5 Hz, Ar**H**), 7.19 (1H, dd, *J* = 8.9, 2.5 Hz, 1H), 5.26 (2H, s, N-N-C**H**$_2$), 4.41 (1H, d, *J* = 5.1 Hz, CH-O-C**H**-C), 4.34 (1H, td, *J* = 8.1, 5.4 Hz, NH-C**H**-CH_2), 4.00 (1H, app t, *J* = 4.7 Hz, C**H**-CH-C), 3.89 (3H, s, OC**H**$_3$), 3.80-3.67 (2H, m, CH_2-C**H** & CH_2-CH-C**H**), 2.27 (2H, ddd, *J* = 9.0, 6.6, 2.4 Hz, C**H**$_2$-C(O)OH), 2.01 – 1.88 (1H, m, C**H**$_A$CH$_B$-CH_2-COOH), 1.87 – 1.72 (1H, m, C**H**$_A$CH$_B$-CH_2-COOH). 2.27 (1H, ddd, *J* = 9.4, 6.6, 3.1 Hz, C**H**$_2$-C(O)OH), 1.99-1.89 (1H, m, C**H**$_A$CH$_B$-CH_2-COOH), Proton 5_A' and 5_B' are partially obscured by the residual water signal; ^{13}C NMR (101 MHz, DMSO) δ 174.1, 171.0, 165.7, 163.8, 157.7, 151.4, 146.5, 140.5, 134.1, 129.8, 128.8, 127.6, 126.2, 124.3, 123.6, 123.1, 119.3, 110.6, 106.2, 81.5, 79.9, 73.5, 72.4, 55.5, 52.4, 51.9, 41.6, 30.3, 28.0. HRMS (ESI) Exact mass calculated for: 491.2024, found: 491.0587



Resin bound (5-(4-(2-((3a*S*,4*S*,6*R*,6a*R*)-4-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-6-(hydroxymethyl)-2-methyltetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine

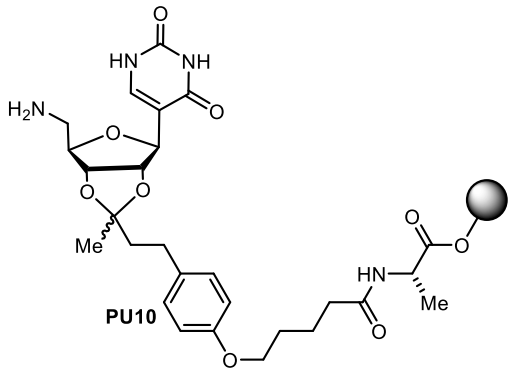
The Fmoc-Ala-Wang resin (3.0 g, 1.80 mmol) was introduced in a solid phase synthesis glass reactor. The resin was swelled in CH_2Cl_2 by shaking at room temperature for 20 min. The solvent was then removed. The solid-phase reactor was filled with 40 mL of 20% solution of 4-methylpiperidine in DMF. The reactor was shaken for 20 min at room temperature, the solvent was removed and the resin was subsequently washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. Then the pseudouridine compound **PU9** (0.971 g, 1.98 mmol) and HBTU (0.751 g, 1.98 mmol) were dissolved in 20 mL of DMF. DIPEA (0.523 mL, 3 mmol) was added to the reaction mixture containing **PU9** and HBTU, which was allowed to react 5 min at room temperature before being added to the solid-phase reactor. The reaction vessel was shaken for 3 h at room temperature. The solvent was removed and the resin was washed three times with 20 mL of DMF and three times with 20 mL of CH_2Cl_2.



Resin bound (5-(4-(2-((3aS,4S,6R,6aR)-4-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-2-methyl-6-((4,5,6,7-tetrachloro-1,3-dioxoisoindolin-2-yl)methyl)tetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine

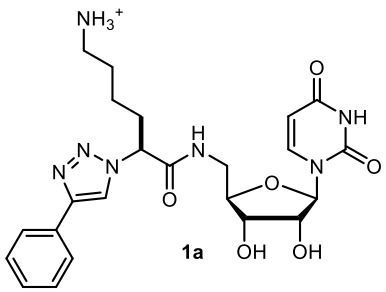
A quantity of resin representing 1.80 mmol (by mass % of the total mass resin) was swelled in DMF. PPh_3 (2.36 g, 9.0 mmol) was added to a round bottom flask and dissolved in 30 mL of a 2:1 mixture of THF:DMF. To the solution was added DIAD (1.77 mL, 9.0 mmol). The reaction was stirred for 2 min upon which was added TCP (2.56 g, 9.0 mmol). The reaction was further stirred for an additional 5 min before being added to the solid phase reactor containing **resin**

bound (5-(4-(2-((3a*S*,4*S*,6*R*,6a*R*)-4-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-6-(hydroxymethyl)-2-methyltetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine. The reaction vessel was shaken for 14 h, and the solvent was removed. The reaction was resubmitted for 3 h with fresh reagents. The solvent was removed and the resin was washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{35}H_{35}Cl_4N_4O_{11}$ $[M+H]^+$: 827.10, found: 827.87.



Resin bound (5-(4-(2-((3aR,4R,6S,6aS)-4-(aminomethyl)-6-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-2-methyltetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine (PU10)

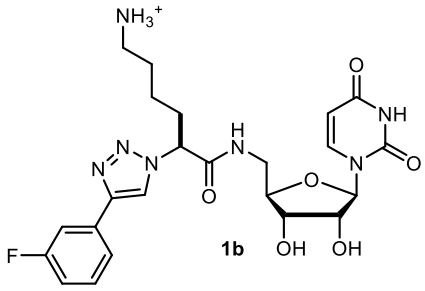
To the solid phase reactor containing **the resin bound (5-(4-(2-((3aS,4S,6R,6aR)-4-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-2-methyl-6-((4,5,6,7-tetrachloro-1,3-dioxoisoindolin-2-yl)methyl)tetrahydrofuro[3,4-d][1,3]dioxol-2-yl)ethyl)phenoxy)pentanoyl)-L-alanine** (1.80 mmol) was added ethylenediamine (0.301 mL, 4.5 mmol) in 30 mL of DMF. The reaction vessel was shaken for 2 h, and the solvent was removed. The reaction was resubmitted with fresh ethylenediamine for another 2 h until completion. The solvent was removed and the resin was washed three times with 40 mL of DMF and three times with 40 mL of CH_2Cl_2. The reaction progress was followed by LC-MS analysis of a cleaved sample of the resin, following general procedure A. $C_{27}H_{37}N_4O_9$ $[M+H]^+$: 561.25, found: 561.67



(S)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-6-oxo-5-(4-phenyl-1H-1,2,3-triazol-1-yl)hexan-1-aminium (1a)

Compound **1a** was prepared using general method B, using phenylacetylene (148 μL, 1.35 mmol) and was purified by reverse phase

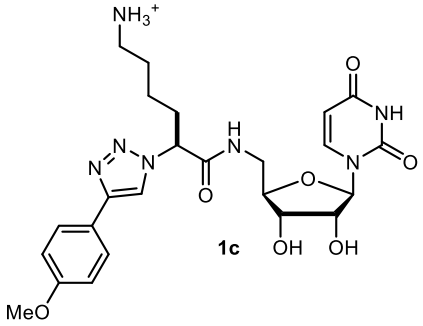
HPLC (15 to 30% MeCN/H_2O; 0.1% TFA). The title compound **1a** was obtained as a white fluffy solid (14 mg, 10%). ^{1}H NMR (400 MHz, DMSO-d_6) δ =11.34 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 8.76-8.70 (2H, m, C(O)-N**H**-CH_2 & C**H**-N-N), 7.89 (2H, dd, *J* = 7.2, 1.5 Hz, 2 ×Ar**H**), 7.69 (3H, br s, N$\mathbf{H}_3^+$), 7.62 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.45 (2H, t, *J* = 7.7 Hz, 2 ×Ar**H**), 7.34 (1H, dt, *J* = 7.5, 1.6 Hz, Ar**H**), 5.73 (1H, d, *J* = 5.5 Hz, CH-O-C**H**-N), 5.64 (1H, dd, *J* = 8.1, 2.2 Hz, C**H**-C(O)-NH-CH_2), 5.45 (1H, br s, O**H**), 5.37 (1H, app dd, *J* = 9.1, 6.2 Hz, C**H**-C(O)-NH-CH_2), 5.21 (1H, br s, O**H**), 4.08 (1H, t, *J* = 5.5 Hz, C**H**-CH-N), 3.87 (1H, t, *J* = 4.7 Hz, NH-CH_2-C**H**), 3.80 (1H, app dt, *J* = 7.2, 4.4 Hz, CH_2-CH-C**H**), 3.50 (1H, ddd, *J* = 13.9, 6.1, 4.4 Hz, NH-C$\mathbf{H}_A$$CH_B$), 2.79-2.70 (2H, m, C$\mathbf{H}_2$-$NH_3^+$), 2.19-2.01 (2H, m, C$\mathbf{H}_2$-$CH_2$-$NH_3^+$), 1.61-1.47 (2H, m, CH-C$\mathbf{H}_2$-CH_2), 1.31-1.11 (2H, m, CH-CH_2-C$\mathbf{H}_2$); NH-CH_AC$\mathbf{H}_B$ is hidden beneath the water peak; ^{13}C NMR (101 MHz, DMSO) δ 168.2, 163.1, 150.7, 146.3, 141.3, 130.8, 128.9, 127.9, 125.1, 120.7, 102.0, 88.5, 82.2, 72.4, 70.8, 62.7, 41.2, 38.6, 31.4, 26.4, 22.3. HRMS (ESI) Exact mass calculated for $C_{23}H_{30}N_7O_6$ $[M+H]^+$: 500.2252, found: 500.2233



(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-5-(4-(3-fluorophenyl)-1H-1,2,3-triazol-1-yl)-6-oxohexan-1-aminium (1b)

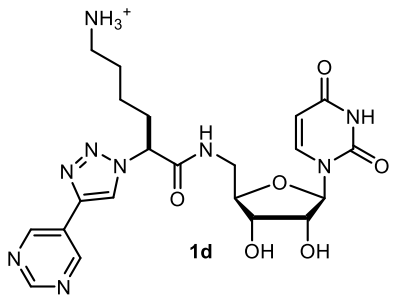
Compound **1b** was prepared using general method B, using 1-ethynyl-3-fluorobenzene (155 µL, 1.35 mmol) and was purified by reverse phase HPLC (17 to 30% MeCN/H_2O; 0.1% TFA). The title compound **1b** was obtained as a white fluffy solid (24 mg, 17%). ^{1}H NMR (400 MHz, DMSO-d_6) δ =11.36 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 8.83 (1H, s, C**H**-N-N), 8.75 (1H, t, *J* = 5.8 Hz, C(O)-N**H**-CH_2), 7.75 (1H, d, *J* = 7.9 Hz, 2 ×Ar**H**), 7.73-7.66 (4H, br s, Ar**H** & N$\mathbf{H}_3^+$), 7.62 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.50 (1H, td, *J* = 8.0, 6.1 Hz, Ar**H**), 7.17 (1H, td, *J* = 8.6, 2.6 Hz, Ar**H**), 5.73 (1H, d, *J* = 5.5 Hz, CH-O-C**H**-N), 5.63 (1H, dd, *J* = 8.1, 2.1 Hz, C**H**-C(O)-NH), 5.45 (1H, br s, O**H**), 5.37 (1H, app dd, *J* = 8.9, 6.4 Hz, C**H**-C(O)-NH-CH_2), 5.21 (1H, br s, O**H**), 4.08 (1H, t, *J* = 5.5 Hz, C**H**-

CH-N), 3.87 (1H, t, *J* = 5.0 Hz, NH-CH_2-C**H**), 3.80 (1H, app dt, *J* = 8.5, 4.5 Hz, CH_2-CH-C**H**), 3.55-3.46 (1H, m, NH-C$\mathbf{H}_A$$CH_B$), 3.36-3.25 (1H, m, NH-$CH_AC\mathbf{H}_B$), 2.75 (2H, app q, *J* = 6.8 Hz, C$\mathbf{H}_2$-NH_3^+), 2.22-2.05 (2H, m, C$\mathbf{H}_2$-CH_2-NH_3^+), 1.66-1.48 (2H, m, CH-C$\mathbf{H}_2$-CH_2), 1.33-1.10 (2H, m, CH-CH_2-C$\mathbf{H}_2$); ^{13}C NMR (101 MHz, DMSO) δ 168.1, 163.0, 162.6 (d, *J* = 249.6 Hz) , 150.7, 145.2 (d, *J* = 4.8 Hz), 141.3, 133.1 (d, *J* = 9.0 Hz), 131.1 (d, *J* = 12.5 Hz), 121.5, 121.1 (d, *J* = 3.0 Hz), 114.6 (d, 26.3 Hz), 111.7 (d, 24.2 Hz), 102.0, 88.5, 82.1, 72.4, 70.8, 62.7, 41.2, 38.5, 31.4, 26.4, 22.3. HRMS (ESI) Exact mass calculated for $C_{23}H_{29}FN_7O_7$ $[M+H]^+$: 518.2158, found: 518.2135



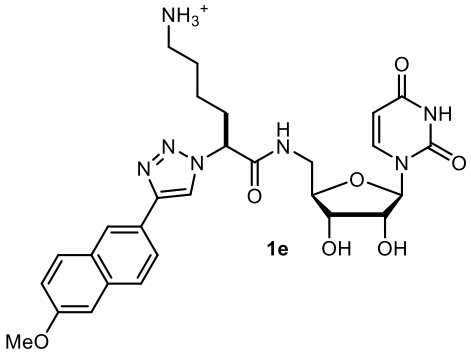
(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-5-(4-(4-methoxyphenyl)-1H-1,2,3-triazol-1-yl)-6-oxohexan-1-aminium (1c)

Compound **1c** was prepared using general method B, using 4-ethynylanisole (175 µL, 1.35 mmol) and was purified by reverse phase HPLC (15 to 25% MeCN/H_2O; 0.1% TFA). The title compound **1c** was obtained as a white fluffy solid (18 mg, 13%). ^{1}H NMR (400 MHz, DMSO-d_6) δ =11.36 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 8.74 (1H, t, *J* = 5.8 Hz, C(O)-N**H**-CH_2), 8.60 (1H, s, C**H**-N-N), 7.81 (2H, d, *J* = 8.7 Hz, 2 ×Ar**H**), 7.72 (3H, br s, N$\mathbf{H}_3^+$), 7.62 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.01 (2H, d, *J* = 8.8 Hz, 2 ×Ar**H**), 5.73 (1H, d, *J* = 5.6 Hz, CH-O-C**H**-N), 5.63 (1H, dd, *J* = 8.0, 2.1 Hz, C**H**-C(O)-NH), 5.45 (1H, br s, O**H**), 5.37 (1H, app dd, *J* = 9.0, 6.3 Hz, C**H**-C(O)-NH-CH_2), 5.22 (1H, br s, O**H**), 4.07 (1H, t, *J* = 5.6 Hz, C**H**-CH-N), 3.87 (1H, t, *J* = 4.7 Hz, NH-CH_2-C**H**), 3.83-3.74 (4H, m, CH_2-CH-C**H** & O$\mathbf{CH}_3$), 3.54-3.42 (1H, m, NH-C$\mathbf{H}_A$$CH_B$), 2.81-2.67 (2H, m, C$\mathbf{H}_2$-$NH_3^+$), 2.18-2.01 (2H, m, C$\mathbf{H}_2$-$CH_2$-$NH_3^+$), 1.61-1.48 (2H, m, CH-C$\mathbf{H}_2$-CH_2), 1.30-1.11 (2H, m, CH-CH_2-C$\mathbf{H}_2$); NH-CH_AC$\mathbf{H}_B$ is hidden beneath the water peak; ^{13}C NMR (101 MHz, DMSO) δ 168.2, 163.1, 159.0, 150.7, 146.2, 141.3, 126.5, 123.4, 119.7, 114.3, 102.0, 88.4, 82.2, 72.4, 70.8, 62.6, 55.2, 41.1, 38.5, 31.4, 26.4, 22.3.HRMS (ESI) Exact mass calculated for $C_{24}H_{32}N_7O_7$ $[M+H]^+$: 530.2358, found:530.2329



(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-6-oxo-5-(4-(pyrimidin-5-yl)-1H-1,2,3-triazol-1-yl)hexan-1-aminium (1d)

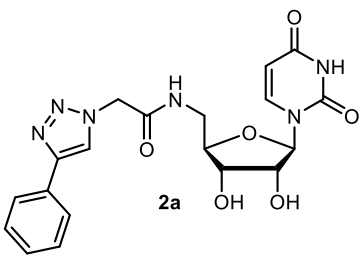
Compound **1d** was prepared using general method B, using 5-ethynylpyrimidine (140 mg, 1.35 mmol) and purified by reverse phase HPLC (2 to 17% MeCN/H_2O; 0.1% TFA). The title compound **1d** was obtained as a white fluffy solid (22.5 mg, 17%). ^{1}H NMR (400 MHz, DMSO-d_6) δ =11.36 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 9.28 (2H, s, 2 × Ar**H**), 9.17 (1H, s, Ar**H**), 9.05 (1H, s, C**H**-N-N), 8.81 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH_2), 7.73 (3H, br s, N**H**$_3^+$), 7.63 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 5.73 (1H, d, *J* = 5.5 Hz, CH-O-C**H**-N), 5.63 (1H, dd, *J* = 8.0, 2.1 Hz, C**H**-C(O)-NH), 5.44 (1H, app t, *J* = 7.7 Hz, C**H**-C(O)-NH-CH_2), 4.08 (1H, t, *J* = 5.5 Hz, C**H**-CH-N), 3.80 (1H, app dt, *J* = 8.8, 4.5 Hz, NH-CH_2-C**H**), 3.80 (1H, app dt, *J* = 8.5, 4.5 Hz, CH_2-CH-C**H**), 3.57-3.46 (1H, m, NH-C**H**$_A$CH$_B$), 3.36-3.26 (1H, m, NH-CH$_A$C**H**$_B$), 2.75 (2H, app q, *J* = 6.7 Hz, C**H**$_2$-NH_3^+), 2.17-2.07 (2H, m, C**H**$_2$-CH_2-NH_3^+), 1.62-1.50 (2H, m, CH-C**H**$_2$-CH_2), 1.28-1.15 (2H, m, CH-CH_2-C**H**$_2$); ^{13}C NMR (101 MHz, DMSO) δ 167.9, 163.0, 157.6, 153.2, 150.7, 141.3, 140.6, 125.0, 122.2, 102.0, 88.5, 82.1, 72.4, 70.8, 62.8, 41.2, 38.5, 31.6, 26.4, 22.2. HRMS (ESI) Exact mass calculated for $C_{21}H_{28}N_9O_6$ $[M+H]^+$: 502.2157, found 502.2142



(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-5-(4-(6-methoxylen-2-yl)-1H-1,2,3-triazol-1-yl)-6-oxohexan-1-aminium (1e)

Compound **1e** was prepared using general method B, using 2-ethynyl-6-methoxynaphthalene (246 mg, 1.35 mmol) and was purified by reverse phase HPLC (20 to 32% MeCN/H_2O; 0.1% TFA). The title compound **1e** was obtained as a white fluffy solid (14 mg, 9%). ^{1}H NMR (400 MHz, DMSO-d_6) δ =11.36 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-

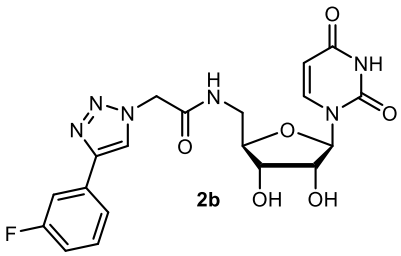
N), 8.81-8.64 (2H, m, C(O)-N**H**-CH_2 & C**H**-N-N)), 8.36 (1H, s, Ar**H**), 7.99 (1H, dd, *J* = 8.5, 1.6 Hz, Ar**H**), 7.89 (2H, app dd *J* = 8.8, 4.9 Hz, 2 × Ar**H**), 7.72 (3H, br s, N$\mathbf{H}_3^+$), 7.63 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.35 (1H, d, *J* = 2.4 Hz, Ar**H**), 7.20 (1H, dd, *J* = 8.9, 2.4 Hz, Ar**H**), 5.74 (1H, d, *J* = 5.6 Hz, CH-O-C**H**-N), 5.65 (1H, dd, *J* = 8.1, 2.1 Hz, C**H**-C(O)-NH), 5.44 (1H, app dd, *J* = 9.1, 6.3 Hz, C**H**-C(O)-NH-CH_2), 4.08 (1H, t, *J* = 5.5 Hz, C**H**-CH-N), 3.93-3.86 (4H, m, NH-CH_2-C**H** & O$\mathbf{CH}_3$), 3.82 (1H, app dt, *J* = 8.8, 4.6 Hz, CH_2-CH-C**H**), 3.56-3.48 (1H, m, NH-C$\mathbf{H}_A$$CH_B$), 3.36-3.21 (1H, m, NH-$CH_AC\mathbf{H}_B$), 2.76 (2H, app q, *J* = 6.7 Hz, C$\mathbf{H}_2$-NH_3^+), 2.22-2.07 (2H, m, C$\mathbf{H}_2$-CH_2-NH_3^+), 1.62-1.50 (2H, m, CH-C$\mathbf{H}_2$-CH_2), 1.28-1.15 (2H, m, CH-CH_2-C$\mathbf{H}_2$); ^{13}C NMR (101 MHz, DMSO) δ 168.2, 163.0, 157.5, 150.7, 146.5, 141.3, 133.9, 129.5, 128.6, 127.3, 126.0, 124.1, 123.4, 120.6, 119.2, 106.1, 102.0, 88.5, 82.1, 72.4, 70.8, 62.7, 55.3, 41.7, 38.5, 31.5, 26.4, 22.4; HRMS (ESI) Exact mass calculated for $C_{28}H_{34}N_7O_7$ $[M+H]^+$: 580.2514, found 580.2481



2a

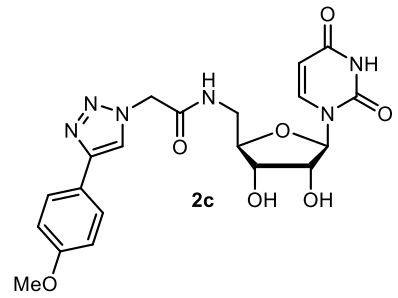
N-(((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)-2-(4-phenyl-1H-1,2,3-triazol-1-yl)acetamide (2a).

Compound **2a** was prepared using general method C using phenylacetylene (148 μL, 1.35 mmol) and was purified by reverse phase HPLC (10 to 40% MeCN/H_2O; 0.1% TFA). The title compound **2a** was obtained as a white fluffy solid (9 mg, 8 %).^{1}H NMR (400 MHz, DMSO-d_6) δ = 11.37 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 8.57 (1H, t, *J* = 5.8 Hz, C(O)-N**H**-CH_2), 8.51 (1H, s, C**H**-N-N), 7.89-7.83 (2H, m, 2 × Ar**H**), 7.67 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.47 (2H, app t, *J* = 7.0 Hz, 2 × Ar**H**), 7.37-7.30 (1H, m, Ar**H**), 5.75 (1H, d, *J* = 5.4 Hz, CH-O-C**H**-N), 5.66 (1H, dd, *J* = 8.0, 2.2 Hz, C**H**-C(O)-NH), 5.17 (2H, s, N-N-C$\mathbf{H}_2$), 4.10 (1H, app t, *J* = 5.5 Hz, C**H**-CH-N), 3.89 (1H, app t, *J* = 5.0 Hz, CH_2-CH-C**H**), 3.83 (1H, app dt, *J* = 7.1, 4.4 Hz, CH_2-C**H**), Proton 5_A' and 5_B' are hidden beneath the water peak; ^{13}C NMR (101 MHz, DMSO) δ 165.7, 163.1, 150.7, 146.1, 141.4, 130.7, 128.9, 127.8, 125.1, 123.0, 102.1, 88.5, 82.2, 72.4, 70.7, 51.7, 41.1. HRMS (ESI) Exact mass calculated for $C_{19}H_{21}O_6N_6$ $[M+H]^+$: 429.1523, found: 429.1495



N-(((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)-2-(4-(3-fluorophenyl)-1H-1,2,3-triazol-1-yl)acetamide (2b)

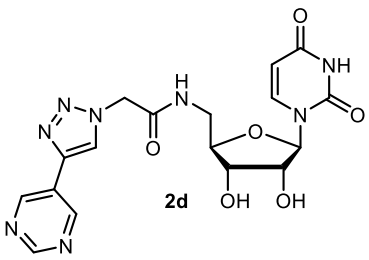
Compound **2b** was prepared using general method C, using 1-ethynyl-3-fluorobenzene (155 μL, 1.35 mmol) and was purified by reverse phase HPLC (10 to 40% MeCN/H_2O; 0.1% TFA). The title compound **2b** was obtained as a white fluffy solid (11 mg, 9 %).^{1}H NMR (400 MHz, DMSO-d_6) δ = 11.37 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 8.62-8.57 (2H, m, , C(O)-N**H**-CH_2 & C**H**-N-N), 7.76-7.64 (3H, 2 ×Ar**H** & C**H**-N-C(O)), 7.50 (1H, app td, *J* = 8.0, 6.1 Hz, Ar**H**), 7.17 (1H, app td, *J* = 8.6, 2.7 Hz, Ar**H**), 5.75 (1H, d, *J* = 5.4 Hz, CH-O-C**H**-N), 5.66 (1H, dd, *J* = 8.1, 2.2 Hz, C**H**-C(O)-NH), 5.19 (2H, s, N-N-C**H**$_2$), 4.10 (1H, app t, *J* = 5.5 Hz, C**H**-CH-N), 3.88 (1H, app t, *J* = 5.0 Hz, CH_2-CH-C**H**), 3.87-3.78 (1H, m, CH_2-C**H**), 3.58-3.47 (1H, m, NH-C**H**$_A$$CH_B$), 3.37-3.27 (1H, m, CH_AC**H**$_B$); ^{13}C NMR (101 MHz, DMSO) δ 165.8, 163.2, 162.6 (d, *J* = 249.2 Hz) 159.9, 145.2 (d, *J* = 4.1 Hz), 141.6, 133.3 (d, *J* = 9.8 Hz), 131.3 (d, *J* = 8.7 Hz), 123.4, 121.4 (d, *J* = 2.9 Hz), 114.7 (d, *J* = 21.2 Hz), 111.9 (d, *J* = 23.0 Hz), 102.2, 88.8, 82.4, 72.6, 71.0, 52.0, 43.3. HRMS (ESI) Exact mass calculated for $C_{19}H_{20}O_6N_6F$ $[M+H]^+$: 447.1428, found: 447.1411



N-(((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)-2-(4-(4-methoxyphenyl)-1H-1,2,3-triazol-1-yl)acetamide (2c).

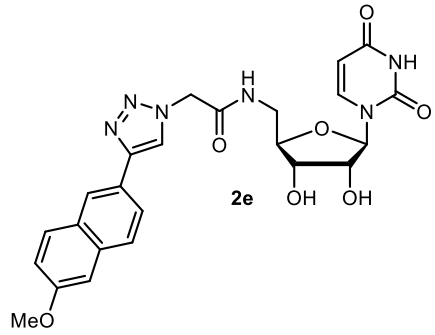
Compound **2c** was prepared using general method C, using 4-ethynylanisole (175 μL, 1.35 mmol) and was purified by reverse phase HPLC (10 to 40% MeCN/H_2O; 0.1% TFA). The title compound **2c** was obtained as a white fluffy solid (18 mg, 15 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 11.37 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 8.56 (1H, t, *J* = 5.8 Hz, C(O)-N**H**-CH_2), 8.39 (1H, s, C**H**-N-N), 7.78 (2H, d, *J* = 8.8 Hz, Ar**H**), 7.67 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.01 (2H, d, *J* = 8.8 Hz, Ar**H**), 5.75 (1H, d, *J* = 5.5 Hz, CH-O-C**H**-N), 5.66 (1H, dd, *J* = 8.0, 2.2 Hz, C**H**-C(O)-NH), 5.15 (2H, s, N-N-C**H**$_2$), 4.10 (1H, app t, *J* = 5.5 Hz, C**H**-CH-N), 3.89 (1H, app t, *J* = 5.5

Hz, CH_2-CH-C**H**), 3.86-3.81 (1H, m, CH_2-C**H**), 3.79 (3H, s, -C**H**3), 3.57-3.47 (1H, m, NH-C$\mathbf{H}_A$CH_B), 3.27-3.26 (1H, m, CH_AC$\mathbf{H}_B$); ^{13}C NMR (101 MHz, DMSO) δ 165.8, 163.0, 159.0, 150.7, 146.0, 141.4, 126.5, 123.3, 122.0, 114.3, 102.0, 89.0, 82.2, 72.4, 71.0, 55.1, 51.7, 41.1. HRMS (ESI) Exact mass calculated for $C_{20}H_{23}N_6O_7$ $[M+H]^+$: 459.1628, found: 459.1617



N-(((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)-2-(4-(pyrimidin-5-yl)-1H-1,2,3-triazol-1-yl)acetamide (2d).

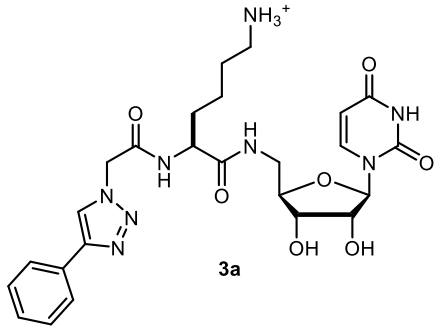
Compound **2d** was prepared using general method C using 5-ethynylpyrimidine (140 mg, 1.35 mmol) and was purified by reverse phase HPLC (2 to 20% MeCN/H_2O; 0.1% TFA). The title compound **2d** was obtained as a white fluffy solid (12 mg, 10 %).^{1}H NMR (400 MHz, DMSO-d_6) δ = 11.30 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 9.20 (2H, s, 2 × Ar**H**), 9.10 (1H, s, Ar**H**), 8.70 (1H, s, C**H**-N-N), 8.55 (1H, t, *J* = 5.8 Hz, C(O)-N**H**-CH_2), 7.61 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 5.68 (1H, d, *J* = 5.4 Hz, CH-O-C**H**-N), 5.59 (1H, dd, *J* = 8.1, 2.2 Hz, C**H**-C(O)-NH), 5.18 (2H, s, N-N-C$\mathbf{H}_2$), 4.03 (1H, app t, *J* = 5.4 Hz, C**H**-CH-N), 3.82 (1H, app t, *J* = 5.0 Hz, CH_2-CH-C**H**), 3.79-.74 (1H, m, CH_2-C**H**), 3.52-3.42 (1H, m, NH-C$\mathbf{H}_A$CH_B), 3.31-3.19 (1H, m, CH_AC$\mathbf{H}_B$); ^{13}C NMR (101 MHz, DMSO) δ 165.5, 163.1, 157.6, 153.3.2, 150.7, 141.3, 140.4, 125.0, 124.4, 102.4, 88.6, 82.2, 724, 70.7, 51.9, 41.1. HRMS (ESI) Exact mass calculated for $C_{17}H_{19}N_8O_6$ $[M+H]^+$: 431.1428, found: 431.1411



N-(((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)-2-(4-(6-methoxylen-2-yl)-1H-1,2,3-triazol-1-yl)acetamide (2e).

Compound **2e** was prepared using general method C, using 2-ethynyl-6-methoxynaphthalene (246 mg, 1.35 mmol) and was purified by reverse phase HPLC (20 to 50% MeCN/H_2O; 0.1% TFA). The title compound **2e** was obtained as a white fluffy solid (9 mg, 7 %). 11.37 (1H, d, *J* = 2.3 Hz,

N**H**-C(O)-N), 8.63-8.55 (2H, m, C(O)-N**H**-CH_2 & C**H**-N-N), 8.34 (1H, s, Ar**H**), 7.95 (1H, dd, *J* = 8.5, 1.6 Hz, Ar**H**), 7.89 (2H, app d, *J* = 8.5 Hz, Ar**H**), 7.68 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.34 (1H, d, *J* = 2.4 Hz, Ar**H**), 7.19 (1H, dd, *J* = 9.0, 2.5 Hz, Ar**H**), 5.76 (1H, d, *J* = 5.4 Hz, CH-O-C**H**-N), 5.67 (1H, dd, *J* = 8.1, 2.1 Hz, C**H**-C(O)-NH), 5.20 (2H, s, N-N-C$\mathbf{H}_2$), 4.10 (1H, app t, *J* = 5.5 Hz, C**H**-CH-N), 3.91-3.80 (5H, m, OC$\mathbf{H}_3$ & CH_2-C**H** and CH_2-CH-C**H**), the two protons are C$\mathbf{H}_2$-CH are obscured by the residual water signal; ^{13}C NMR (101 MHz, DMSO) δ 165.7, 163.0, 157.4, 150.7, 146.3, 141.4, 133.9, 129.5, 128.6, 127.4, 126.0, 124.1, 123.4, 122.8, 119.1, 106.0, 102.0, 88.5, 82.2, 72.4, 70.7, 55.2, two carbons are obscured by the residual solvent signal; HRMS (ESI) Exact mass calculated for $C_{24}H_{25}N_6O_7$ $[M+H]^+$: 509.1785, found: 509.1772

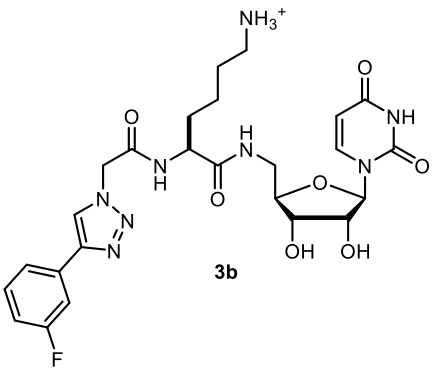


3a

(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-6-oxo-5-(2-(4-phenyl-1H-1,2,3-triazol-1-yl)acetamido)hexan-1-aminium (3a)

Compound **3a** was prepared using general method D using phenylacetylene (148 μL, 1.35 mmol) and was purified by reverse phase HPLC (15 to 30% MeCN/H_2O; 0.1% TFA). The title compound **3a** was obtained as a white fluffy solid (11 mg, 7 %).^{1}H NMR (400 MHz, DMSO-d_6) δ 11.35 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 8.64 (1H, d, *J* = 8.2 Hz, C(O)-N**H**-CH), 8.50 (1H, s, C**H**-N-N), 8.30 (1H, t, *J* = 5.9 Hz, N**H**-CH_2), 7.85 (2H, app d, *J* =7.46 Hz, 2 × Ar**H**), 7.77-7.63 (4H, m , N$\mathbf{H}_3^+$ & C**H**-N-C(O)), 7.45 (2H, app t, *J* = 7.7 Hz, 2 2 × Ar**H**), 7.34 (1H, app td, *J* = 7.6, 1.6 Hz, Ar**H**), 5.72 (1H, d, *J* = 5.8 Hz, CH-O-C**H**-N), 5.64 (1H, dd, *J* = 7.9, 2.2 Hz, C**H**-C(O)-NH), 5.22 (2H, s, N-N-C$\mathbf{H}_2$), 4.37-4.25 (1H, m, C**H**-CH_2-CH_2), 4.07 (1H, t, *J* = 5.5 Hz, C**H**-CH-N), 3.86 (1H, app t, *J* = 4.8 Hz, CH_2-CH-C**H**), 3.83-3.76 (1H, m, NH-CH_2-C**H**), 3-48-3.37 (1H, m, NH-C$\mathbf{H}_A$$CH_B$), 3.35-3-24 (1H, m, NH-$CH_AC\mathbf{H}_B$), 2.75 (2H, app q, *J* = 5.9 Hz, C$\mathbf{H}_2$-NH_3^+), 1.76-1.63 (1H, m, CH-C$\mathbf{H}_A$$CH_B$$CH_2$), 1.61-.1.43 (3H, m, CH-CH_AC$\mathbf{H}_B$$CH_2$ & C$\mathbf{H}_2$-CH_2-NH_3^+) 1.39-1.26 (2H, m, CH-CH_2-C$\mathbf{H}_2$); ^{13}C NMR (101 MHz, DMSO) δ 13C NMR (101 MHz, DMSO) δ 171.4, 165.3, 163.0, 150.7,

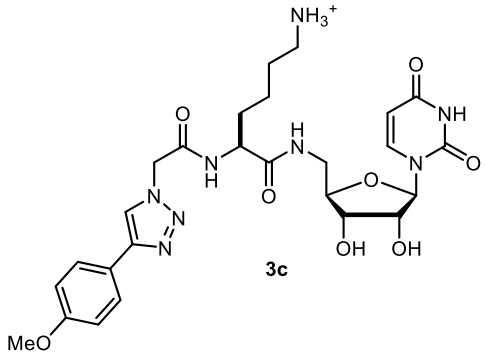
146.1, 141.4, 130.7, 128.9, 127.8, 125.1, 123.0, 101.9, 88.3, 82.4, 72.4, 70.8, 52.4, 51.6, 41.0, 38.7, 31.9, 26.6, 22.1; HRMS (ESI) Exact mass calculated for $C_{25}H_{33}N_8O_7$ $[M+H]^+$: 557.2467, found: 557.2434



3b

(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-5-(2-(4-(3-fluorophenyl)-1H-1,2,3-triazol-1-yl)acetamido)-6-oxohexan-1-aminium (3b)

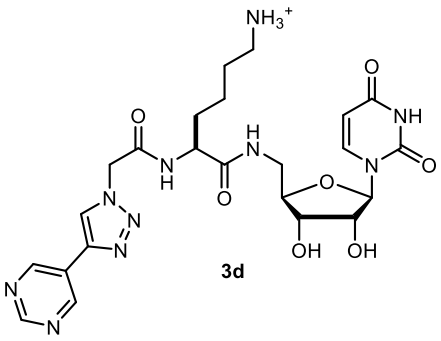
Compound **3b** was prepared using general method D using 1-ethynyl-3-fluorobenzene (155 μL, 1.35 mmol) and was purified by reverse phase HPLC (15 to 30% MeCN/H_2O; 0.1% TFA). The title compound **3b** was obtained as a white fluffy solid (25 mg, 17 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.35 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 8.66 (1H, d, *J* = 8.1 Hz, C(O)-N**H**-CH), 8.59 (1H, s, C**H**-N-N), 8.30 (1H, t, *J* = 5.9 Hz, N**H**-CH_2), 7.77-7-62 (6H, m, 2 × Ar**H**, N**H**$_3$ & C**H**-N-C(O)), 7.50 (1H, app q, *J* = 7.6 Hz, Ar**H**), 7.17 (1H, td, *J* = 8.6, 2.5 Hz, Ar**H**), 5.72 (1H, d, *J* = 5.8 Hz, CH-O-C**H**-N), 5.63 (1H, dd, *J* = 8.1, 2.1 Hz, C**H**-C(O)-NH), 5.23 (2H, s, N-N-C**H**$_2$), 4.38-4.25 (1H, m, C**H**-CH_2-CH_2), 4.07 (1H, t, *J* = 5.6 Hz, C**H**-CH-N), 3.87 (1H, app t, *J* = 4.6 Hz, CH_2-CH-C**H**), 3.81 (1H, app dd, *J* = 6.8, 4.5 Hz, NH-CH_2-C**H**), 3-47-3.40 (1H, m, NH-C**H**$_A$$CH_B$), 3.35-3-24 (1H, m, NH-$CH_A$C**H**$_B$), 2.75 (2H, app q, *J* = 6.8 Hz, C**H**$_2$-NH_3^+), 1.73-1.62 (1H, m, CH-C**H**$_A$$CH_B$$CH_2$), 1.61-.1.46 (3H, m, CH-CH_AC**H**$_B$$CH_2$ & C**H**$_2$-CH_2-NH_3^+) 1.41-1.24 (2H, m, CH-CH_2-C**H**$_2$); ^{13}C NMR (101 MHz, DMSO) δ 171.4, 165.2, 163.1, 162.6 (d, J = 242.8 Hz), 150.7, 145.0 (d, *J* = 3.5 Hz), 141.4, 133.1 (d, *J* = 10.7 Hz), 131.1 (d, *J* = 11.5 Hz), 123.7, 121.2 (d, *J* = 3.0 Hz), 114.5 (d, *J* = 24.1 Hz), 111.7 (d, *J* = 27.3 Hz), 102.0, 88.3, 82.4, 72.4, 70.8, 52.5, 51.7, 41.0, 38.7, 31.9, 26.7, 22.2. HRMS (ESI) Exact mass calculated for $C_{25}H_{32}FN_8O_7$ $[M+H]^+$: 575.2373, found: 575.2350



3c

(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-5-(2-(4-(4-methoxyphenyl)-1H-1,2,3-triazol-1-yl)acetamido)-6-oxohexan-1-aminium (3c) .

Compound **3c** was prepared using general method D, using general method D, using 4-ethynylanisole (175 μL, 1.35 mmol) and was purified by reverse phase HPLC (15 to 30% MeCN/H_2O; 0.1% TFA). The title compound **3c** was obtained as a white fluffy solid (16 mg, 10 %).^{1}H NMR (400 MHz, DMSO-d_6) δ 11.35 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 8.62 (1H, d, *J* = 8.2 Hz, C(O)-N**H**-CH), 8.38 (1H, s, C**H**-N-N), 8.29 (1H, t, *J* = 5.9 Hz, N**H**-CH_2), 7.77 (2H, d, *J* = 8.5 Hz, 2 × Ar**H**), 7.77-7.73 (3H, s, N**H**$_3^+$), 7.67 (1H, d, *J* = 8.5 Hz, C**H**-N-C(O)), 7.01 (2H, d, *J* = 8.5 Hz, 2 × Ar**H**), 5.72 (1H, d, *J* = 5.9 Hz, CH-O-C**H**-N), 5.64 (1H, dd, *J* = 7.9, 2.1 Hz, C**H**-C(O)-NH), 5.19 (2H, s, N-N-C**H**$_2$), 4.37-4.25 (1H, m, C**H**-CH_2-CH_2), 4.07 (1H, t, *J* = 5.6 Hz, C**H**-CH-N), 3.86 (1H, app t, *J* = 4.8 Hz, CH_2-CH-C**H**), 3.83-3.75 (4H, m, OC**H**$_3$ & NH-CH_2-C**H**), 3-47-3.38 (1H, m, NH-C**H**$_A$$CH_B$), 3.33-3-19 (1H, m, NH-$CH_A$C**H**$_B$), 2.75 (2H, app q, *J* = 6.7 Hz, C**H**$_2$-NH_3^+), 1.73-1.62 (1H, m, CH-C**H**$_A$$CH_B$$CH_2$), 1.61-.1.41 (3H, m, CH-CH_AC**H**$_B$$CH_2$ & C**H**$_2$-CH_2-NH_3^+) 1.39-1.25 (2H, m, CH-CH_2-C**H**$_2$); ^{13}C NMR (101 MHz, DMSO) δ 171.4, 165.4, 163.1, 159.0, 150.7, 146.0, 141.4, 126.5, 123.3, 122.0, 114.3, 102.0, 88.3, 82.4, 72.4, 70.8, 55.2, 52.4, 51.6, 41.0, 38.7, 31.9, 26.7, 22.2. HRMS (ESI) Exact mass calculated for $C_{26}H_{36}N_8O_8$ $[M+H]^+$: 587.2572, found: 587.2547

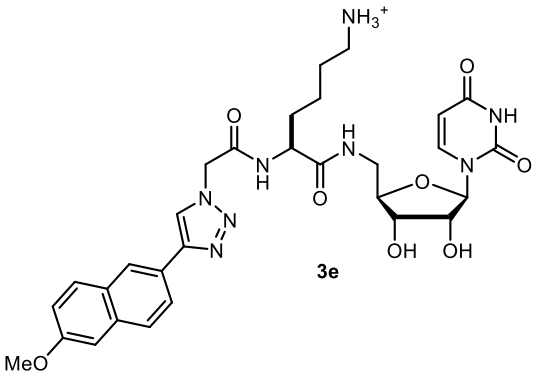


3d

(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-6-oxo-5-(2-(4-(pyrimidin-5-yl)-1H-1,2,3-triazol-1-yl)acetamido)hexan-1-aminium (3d)

Compound **3d** was prepared using general method D, using 5-ethynylpyrimidine (140 mg, 1.35 mmol) and was purified by reverse phase HPLC (4 to 19% MeCN/H_2O; 0.1% TFA). The title compound **3d** was obtained as a white

fluffy solid (5 mg, 4 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 11.35 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 9.27 (2H, s, Ar**H**), 9.17 (1H, s, Ar**H**), 8.75 (1H, s, C**H**-N-N), 8.67 (1H, t, *J* = 8.5 Hz, C(O)-N**H**-CH_2), 8.29 (1H, t, *J* = 5.9 Hz, N**H**-CH_2), 7.77-7.53 (4H, m, N**H**$_3$ & C**H**-N-C(O)), 5.73 (1H, d, *J* = 6.0 Hz, CH-O-C**H**-N), 5.62 (1H, dd, *J* = 8.2, 2.4 Hz, C**H**-C(O)-NH), 5.29 (2H, s, N-N-C**H**$_2$), 4.36-4.23 (1H, m, C**H**-CH_2-CH_2), 4.06 (1H, t, *J* = 5.5 Hz, C**H**-CH-N), 3.86 (1H, app t, *J* = 4.8 Hz, CH_2-CH-C**H**), 3.83-3.72 (1H, app dt, *J* = 8.6, 4.7 Hz, NH-CH_2-C**H**), 2.79-2.67 (2H, m, C**H**$_2$-NH_3^+), 1.77-1.62 (1H, m, CH-C**H**$_A$$CH_B$$CH_2$), 1.59-.1.43 (3H, m, CH-CH_AC**H**$_B$$CH_2$ & C**H**$_2$-CH_2-NH_3^+), 1.38-1.26 (2H, m, CH-CH_2-C**H**$_2$); Proton -NH-C**H**$_A$$CH_B$ & -NH-CH_AC**H**$_B$ are hidden beneath the residual water signal; ^{13}C NMR (101 MHz, DMSO) δ 165.1, 163.0, 157.6, 153.3, 150.7, 150.2, 140.4, 125.0, 124.4, 101.9, 88.3, 85.1, 79.5, 72.4, 70.8, 52.4, 51.8, 41.0, 38.68, 31.9, 26.6, 22.2. HRMS (ESI) Exact mass calculated for $C_{23}H_{31}N_{10}O_7$ $[M+H]^+$: 559.2372, found: 559.2351

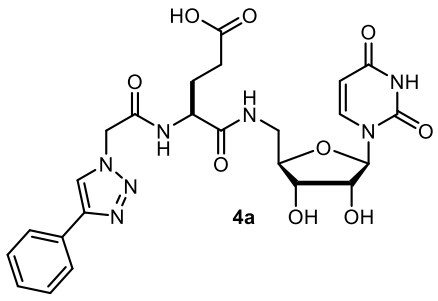


3e

(*S*)-6-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-5-(2-(4-(6-methoxylen-2-yl)-1H-1,2,3-triazol-1-yl)acetamido)-6-oxohexan-1-aminium (3e).

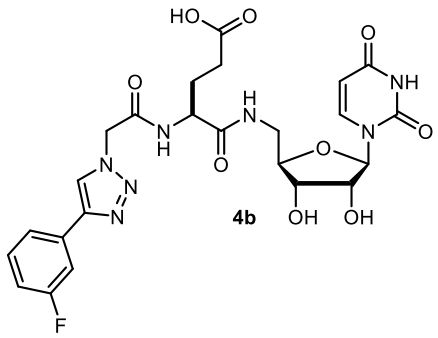
Compound **3e** was prepared using general method D, using 2-ethynyl-6-methoxynaphthalene (246 mg, 1.35 mmol) and was purified by reverse phase HPLC (20 to 50% MeCN/H_2O; 0.1% TFA). The title compound **3e** was obtained as a white fluffy solid (18 mg, 11%). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 11.35 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 8.66 (1H, d, *J* = 8.2 Hz, C(O)-N**H**-CH), 8.56 (1H, s, C**H**-N-N), 8.33 (1H, d, *J* = 1.5 Hz, Ar**H**), 8.30 (1H, t, *J* = 5.8 Hz, C(O)-N**H**-CH_2), 7.95 (1H, dd, *J* = 8.5, 1.7 Hz, Ar**H**), 7.89 (2H, app d, *J* = 8.8 Hz, Ar**H**), 7.69-7.76 (3H, m, N**H**$_3^+$), 7.67 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.34 (1H, d, *J* = 2.5 Hz, Ar**H**), 7.19 (1H, dd, *J* = 8.9, 2.5 Hz, Ar**H**), 5.73 (1H, d, *J* = 5.5 Hz, CH-O-C**H**-N), 5.64 (1H, dd, *J* = 8.1, 2.2 Hz, C**H**-C(O)-NH), 5.25 (2H, s, N-N-C**H**$_2$), 4.33 (1H, td, *J* = 8.5, 5.2 Hz, NH-C**H**), 4.07 (1H, app t, *J* = 5.6 Hz, C**H**-CH-N), 3.90-3.85 (4H, m, OC**H**3 & CH_2-CH-C**H**), 3.84-3.79 (1H, m, NH-CH_2-C**H**), 3.48-3.39 (1H, m, NH-C**H**$_A$$CH_B$), 3.30-3.23 (1H, m, NH-

$CH_AC\mathbf{H}_B$), 2.80-2.69 (2H, m, C$\mathbf{H}_2$-NH_3^+), 1.74-1.62 (1H, m, C$\mathbf{H}_A$CH_B-CH_2-NH_3^+), 1.61-1.45 (3H, m, $CH_AC\mathbf{H}_B$-CH_2-NH_3^+ & CH-C$\mathbf{H}_2$-CH_2), 1.41-1.26 (2H, m, CH-CH_2-C$\mathbf{H}_2$); ^{13}C NMR (101 MHz, DMSO) δ 171.4, 165.4, 163.1, 157.5, 150.7, 146.3, 141.4, 133.9, 129.5, 128.6, 127.4, 126.0, 124.1, 123.4, 122.8, 119.1, 106.0, 102.0, 88.3, 82.4, 72.4, 70.8, 55.3, 52.5, 51.7, 41.0, 38.7, 31.9, 26.7, 22.2. HRMS (ESI) Exact mass calculated for $C_{30}H_{37}N_8O_8$ $[M+H]^+$: 637.2729, found: 637.2711



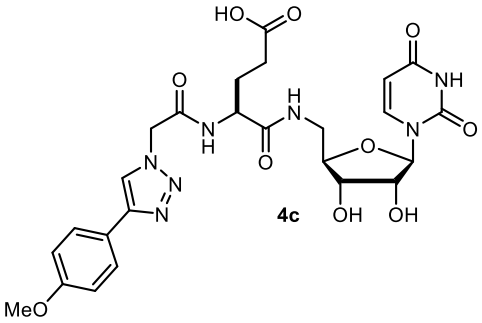
(*S*)-5-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-5-oxo-4-(2-(4-phenyl-1H-1,2,3-triazol-1-yl)acetamido)pentanoic acid (4a)

Compound **4a** was prepared using general method E and using phenylacetylene (148 μL, 1.35 mmol) and was purified by reverse phase HPLC (13 to 33% MeCN/H_2O; 0.1% TFA). The title compound **4a** was obtained as a white fluffy solid (17 mg, 12 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 12.11 (1H, br s, C(O)O**H**), 11.33 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 8.63 (1H, d, *J* = 8.1 Hz, C(O)-N**H**-CH), 8.50 (1H, s, C**H**-N-N), 8.26 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH_2), 7.85 (2H, dd, *J* = 8.2, 1.3 Hz, 2×Ar**H**), 7.65 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.45 (2H, dd, *J* = 8.2, 7.0 Hz, 2×Ar**H**), 7.36-7.32 (1H, m, Ar**H**), 5.73 (1H, d, *J* = 5.8 Hz, CH-O-C**H**-N), 5.63 (1H, dd, *J* = 8.1, 2.2 Hz, C**H**-C(O)-NH), 5.40 (1H, d, *J* = 5.7 Hz, O**H**), 5.22 (2H, s, N-N-C$\mathbf{H}_2$), 5.17 (1H, d, *J* = 5.1 Hz, O**H**), 4.34 (1H, td, *J* = 8.3, 5.3 Hz, NH-C**H**), 4.05 (1H, app q, *J* = 5.6 Hz, C**H**-CH-N), 3.88-3.76 (2H, m, NH-CH_2-C**H** & CH_2-CH-C**H**), 3.46-3.38 (1H, m, NH-C$\mathbf{H}_A$CH_B), 3.30-3.23 (1H, m, NH-CH_AC$\mathbf{H}_B$), 2.25 (2H, ddd, *J* = 9.2, 6.6, 2.9 Hz, C$\mathbf{H}_2$-C(O)OH), 1.99-1.85 (1H, m, C$\mathbf{H}_A$CH_B-CH_2-COOH), 1.84-1.70 (1H, m, C$\mathbf{H}_A$CH_B-CH_2-COOH); ^{13}C NMR (101 MHz, DMSO) δ 173.8, 171.0, 165.4, 163.02, 150.7, 146.1, 130.7, 128.9, 127.8, 125.1, 122.9, 102.0, 99.5, 88.2, 82.4, 72.5, 70.7, 52.1, 51.6, 40.9, 30.0, 27.8. HRMS (ESI) Exact mass calculated for $C_{24}H_{28}N_7O_9$ $[M+H]^+$: 558.1949, found: 558.1935



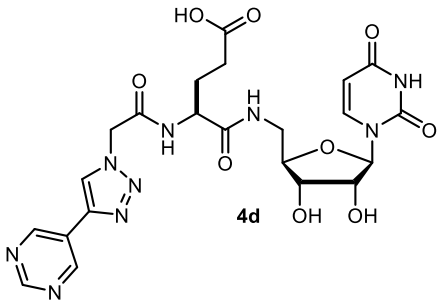
(*S*)-5-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-4-(2-(4-(3-fluorophenyl)-1H-1,2,3-triazol-1-yl)acetamido)-5-oxopentanoic acid (4b)

Compound **4b** was prepared using general method E using 1-ethynyl-3-fluorobenzene (155 µL, 1.35 mmol) and was purified by reverse phase HPLC (13 to 33% MeCN/H_2O; 0.1% TFA). The title compound **4b** was obtained as a white fluffy solid (19 mg, 13%). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 12.13 (1H, br s, C(O)O**H**), 11.33 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 8.64 (1H, d, *J* = 8.1 Hz, C(O)-N**H**-CH), 8.59 (1H, s, C**H**-N-N), 8.30 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH_2), 7.69 (3H, m, 2×Ar**H** & C**H**-N-C(O)), 7.50 (1H, app td, *J* = 8.0, 6.1 Hz, Ar**H**), 7.17 (1H, m, Ar**H**), 5.73 (1H, d, *J* = 5.7 Hz, CH-O-C**H**-N), 5.62 (1H, dd, *J* = 8.1, 2.2 Hz, C**H**-C(O)-NH), 5.40 (1H, d, *J* = 5.7 Hz, O**H**), 5.24 (2H, s, N-N-C**H**$_2$), 5.17 (1H, d, *J* = 5.1 Hz, O**H**), 4.34 (1H, td, *J* = 8.2, 5.2 Hz, NH-C**H**), 4.05 (1H, app q, *J* = 5.6 Hz, C**H**-CH-N), 3.88-3.78 (2H, m, NH-CH_2-C**H** & CH_2-CH-C**H**), 3.48-3.34 (1H, m, NH-C**H**$_A$$CH_B$), 3.30-3.23 (1H, m, NH-$CH_A$C**H**$_B$), 2.25 (2H, ddd, *J* = 9.2, 6.5, 3.0 Hz, C**H**$_2$-C(O)OH), 1.98-1.87 (1H, m, C**H**$_A$$CH_B$-$CH_2$-COOH), 1.83-1.71 (1H, m, C**H**$_A$$CH_B$-$CH_2$-COOH); ^{13}C NMR (101 MHz, DMSO) δ 173.8, 171.0, 165.3, 163.0, 162.6 (d, *J* = 240.6Hz), 150.7, 145.0 (d, *J* = 3.5 Hz), 141.2, 133.1 (d, *J* = 11.3 Hz), 131.0 (d, *J* = 10.9 Hz), 123.7, 121.2 (d, *J* = 3.2 Hz), 114.5 (d, *J* = 26.6 HZ), 111.7 (d, *J*= 21.7 Hz), 102.0, 88.2, 82.4, 72.5, 70.7, 52.1, 51.7, 41.0, 30.1, 27.8. HRMS (ESI) Exact mass calculated for $C_{24}H_{27}FN_7O_9$ $[M+H]^+$: 576.1849, found:576.1827



(*S*)-5-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-4-(2-(4-(4-methoxyphenyl)-1H-1,2,3-triazol-1-yl)acetamido)-5-oxopentanoic acid (4c)

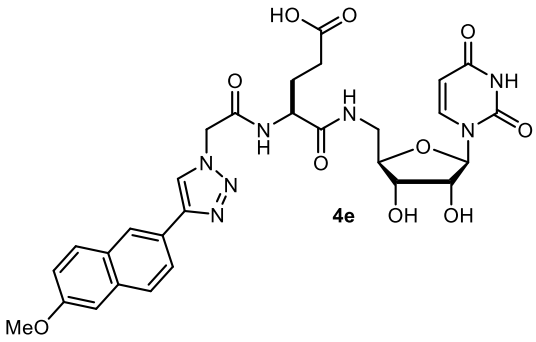
Compound **4c** was prepared using general method E, using 4-ethynylanisole (175 µL, 1.35 mmol) and was purified by reverse phase HPLC (13 to 33% MeCN/H_2O; 0.1% TFA). The title compound **4c** was obtained as a white fluffy solid (15 mg, 9%). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 12.13 (1H, br s, C(O)O**H**), 11.33 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 8.61 (1H, d, *J* = 8.2 Hz, C(O)-N**H**-CH), 8.38 (1H, s, C**H**-N-N), 8.26 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH_2), 7.77 (2H, d, *J* = 8.7 Hz, Ar**H**), 7.65 (1H, d, *J* = 8.1, C**H**-N-C(O)), 7.01 (2H, d, *J* = 8.8 Hz, Ar**H**), 5.73 (1H, d, *J* = 5.7 Hz, CH-O-C**H**-N), 5.63 (1H, dd, *J* = 8.1, 2.2 Hz, C**H**-C(O)-NH), 5.39 (1H, br s, O**H**), 5.19 (2H, s, N-N-C**H**$_2$), 5.17 (1H, d, br s, O**H**), 4.34 (1H, td, *J* = 8.3, 5.3 Hz, NH-C**H**), 4.05 (1H, app t, *J* = 5.5 Hz, C**H**-CH-N), 3.88-3.80 (2H, m, NH-CH_2-C**H** & CH_2-CH-C**H**), 3.79 (3H, s, OC**H**$_3$), 2.25 (2H, ddd, *J* = 9.2, 6.6, 2.9 Hz, C**H**$_2$-C(O)OH), 1.98-1.87 (1H, m, C**H**$_A$$CH_B$-$CH_2$-COOH), 1.83-1.70 (1H, m, C**H**$_A$$CH_B$-$CH_2$-COOH); Two protons NH-C**H**$_A$$CH_B$ & NH-CH_AC**H**$_B$ are hidden beneath the residual water signal. ^{13}C NMR (101 MHz, DMSO) δ 173.8, 171.1, 165.5, 163.0, 159.0, 150.7, 146.0, 141.2, 126.5, 123.3, 122.0, 114.3, 102.0, 88.2, 82.4, 72.5, 70.7, 55.1, 52.1, 51.6, 40.9, 30.1, 27.8. HRMS (ESI) Exact mass calculated for $C_{25}H_{30}N_7O_{10}$ $[M+H]^+$: 588.2042, found: 588.2022



(*S*)-5-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-5-oxo-4-(2-(4-(pyrimidin-5-yl)-1H-1,2,3-triazol-1-yl)acetamido)pentanoic acid (4d).

Compound **4d** was prepared using general method E, using 5-ethynylpyrimidine (140 mg, 1.35 mmol) and was purified by reverse phase HPLC (10 to 30% MeCN/H_2O; 0.1% TFA). The title compound **4d** was obtained as a white fluffy solid (9 mg, 6 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 12.14 (1H, br s, C(O)O**H**), 11.33 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 9.27 (2H, s, 2 × Ar**H**),

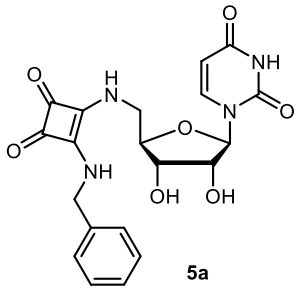
9.16 (1H, s, Ar**H**), 8.75 (1H, s, C**H**-N-N), 8.67 (1H, d, *J* =8.2 Hz, C(O)-N**H**-C(O)), 8.27 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH_2), 7.64 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 5.72 (1H, d, *J* = 5.7 Hz, CH-O-C**H**-N), 5.62 (1H, dd, *J* = 8.0, 2.2 Hz, C**H**-C(O)-NH-C(O)), 5.30 (2H, s, N-N-C$\mathbf{H}_2$), 4.40-4.26 (1H, m, NH-C**H**), 4.04 (1H, app q, *J* = 5.5 Hz, C**H**-CH-N) 3.88-3.78 (1H, m, CH_2-CH-C**H** & CH_2-C**H**), 2.26 (2H, ddd, *J* = 9.4, 6.4, 3.3 Hz, C$\mathbf{H}_2$-C(O)OH), 1.99-1.87 (1H, m, C$\mathbf{H}_A$$CH_B$-$CH_2$-COOH), 1.84-1.71 (1H, m, C$\mathbf{H}_A$$CH_B$-$CH_2$-COOH); the two missing protons NH-C$\mathbf{H}_A$$CH_B$ and NH-CH_AC$\mathbf{H}_B$ are under the water peak; ^{13}C NMR (101 MHz, DMSO) δ 173.8, 171.0, 165.2, 163.0, 157.5, 153.3, 150.7, 141.2, 140.4, 125.0, 124.4, 102.0, 88.2, 82.4, 72.5, 70.7, 52.1, 51.8, 40.9, 30.0, 27.8. HRMS (ESI) Exact mass calculated for $C_{22}H_{26}N_9O_9$ $[M+H]^+$: 560.1848, found: 560.1831



(*S*)-5-((((2*R*,3*S*,4*R*,5*R*)-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-4-(2-(4-(6-methoxylen-2-yl)-1H-1,2,3-triazol-1-yl)acetamido)-5-oxopentanoic acid (4e).

Compound **4e** was prepared using general method E, using 2-ethynyl-6-methoxynaphthalene (246 mg, 1.35 mmol) and was purified by reverse phase HPLC (13 to 33% MeCN/H_2O; 0.1% TFA). The title compound **4e** was obtained as a white fluffy solid (18 mg, 10%). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 12.17 (1H, br s, C(O)O**H**), 11.34 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 8.65 (1H, d, *J* = 8.2 Hz, C(O)-N**H**-CH), 8.55 (1H, s, C**H**-N-N)), 8.34 (1H, d, *J* = 1.5 Hz, Ar**H**), 8.27 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH_2), 7.95 (1H, dd, *J* = 8.6, 1.7 Hz, Ar**H**), 7.89 (2H, app d, *J* = 7.9 Hz, Ar**H**), 7.66 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.34 (1H, d, *J* = 2.5 Hz, Ar**H**), 7.19 (1H, dd, *J* = 8.9, 2.5 Hz, Ar**H**), 5.73 (1H, d, *J* = 5.7 Hz, CH-O-C**H**-N), 5.64 (1H, dd, *J* = 8.1, 2.2 Hz, C**H**-C(O)-NH), 5.25 (2H, s, N-N-C$\mathbf{H}_2$), 4.35 (1H, td, *J* = 8.3, 5.3 Hz, NH-C**H**), 4.05 (1H, t, *J* = 5.5 Hz, C**H**-CH-N), 3.89 (3H, s, OC$\mathbf{H}_3$), 3.88-3.77 (2H, m, NH-CH_2-C**H** & CH_2-CH-C**H**), 3.48-3.38 (1H, m, NH-C$\mathbf{H}_A$$CH_B$), 3.36-3.25 (1H, m, NH-$CH_AC\mathbf{H}_B$), 2.27 (2H, ddd, *J* = 9.4, 6.6, 3.1 Hz, C$\mathbf{H}_2$-C(O)OH), 1.99-1.89 (1H, m, C$\mathbf{H}_A$$CH_B$-$CH_2$-COOH), 1.85-1.73

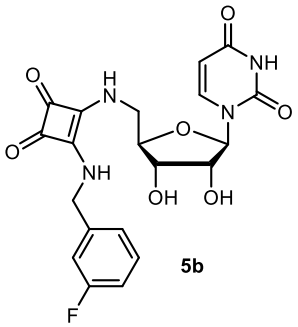
(1H, m, C**H**$_A$CH$_B$-CH$_2$-COOH); ^{13}C NMR (101 MHz, DMSO) δ 173.8, 171.1, 165.5, 163.0, 157.4, 150.7, 146.3, 141.2, 133.9, 129.6, 128.6, 127.4, 126.0, 124.1, 123.4, 122.8, 119.1, 106.0, 102.0, 88.2, 82.4, 72.5, 70.7, 55.2, 52.1, 51.7, 41.0, 30.1, 27.8. HRMS (ESI) Exact mass calculated for $C_{29}H_{32}N_7O_{10}$ $[M+H]^+$: 638.2205, found: 638.2182



5a

1-((2R,3R,4S,5R)-5-(((2-(benzylamino)-3,4-dioxocyclobut-1-en-1-yl)amino)methyl)-3,4-dihydroxytetrahydrofuran-2-yl)pyrimidine-2,4(1H,3H)-dione (5a)

Compound **5a** was prepared using general method F, using benzylamine (88 μL, 0.81 mmol) and was purified by reverse phase HPLC (12 to 35% MeCN/H_2O; 0.1% TFA. The title compound **5a** was obtained as a white fluffy solid (8 mg, 7 %); ^{1}H NMR (400 MHz, DMSO-d_6) δ = 11.37 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 7.75-7.60 (3H, m, C**H**-N-C(O) & 2 × N**H**), 7.51 (1H, td, *J* = 8.0, 6.1 Hz, Ar**H**), 7.35-7.12 (4H, m, 4 × Ar**H**), 5.76 (1H, d, *J* = 5.6 Hz, CH-O-C**H**-N), 5.62-5.55 (1H , m, C**H**-C(O)-NH), 5.48 (1H, br s, O**H**), 5.28 (1H, br s, O**H**), 4.72-4.67 (2H, m, C-C**H**$_2$-NH-C), 4.12-4.02 (1H, m, C**H**-CH-N), 3.97-3.80 (3H, m, C-NH-C**H**$_2$-CH & CH$_2$-CH-C**H**), 3.69-3.55 (1H, m, CH$_2$-C**H**-CH); ^{13}C NMR (400 MHz, DMSO-d_6) δ 178.1, 163.4, 156.9, 151.3, 145.3, 141.6, 136.4, 129.1, 128.0, 127.9, 102.5, 890, 83.3, 77.5, 72.7, 70.0, 47.2, 45.8. HRMS (ESI) Exact mass calculated for $C_{20}H_{21}N_4O_7$ $[M+H]^+$: 429.1405, found: 429.1430;

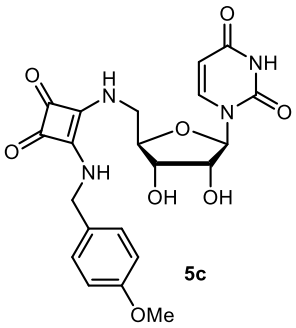


5b

1-((2R,3R,4S,5R)-5-(((2-((3-fluorobenzyl)amino)-3,4-dioxocyclobut1-en-1-yl)amino)methyl)-3,4-dihydroxytetrahydrofuran-2-yl)pyrimidine-2,4(1H,3H)-dione (5b)

Compound **5b** was prepared using general method F, using 3-fluorobenzylamine (88 μL, 0.81 mmol) and was purified by reverse phase HPLC (12 to 35% MeCN/H_2O; 0.1% TFA). The title compound **5b** was obtained as a white fluffy solid (8 mg, 7 %); (400 MHz, DMSO-d_6) δ = 11.36 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 7.82 (1H, br s, N**H**), 7.61 (1H, d, *J* = 7.9 Hz,

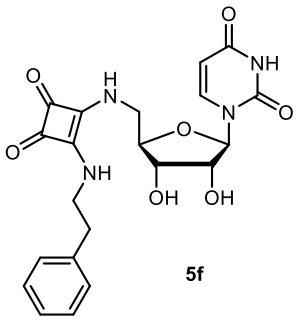
C**H**-N-C(O)), 7.51 (1H, br s, N**H**), 7.45-7.32 (1H, m, Ar**H**), 7.22-7.04 (3H, m, 3 × Ar**H**), 5.77 (1H, d, *J* = 5.6 Hz, CH-O-C**H**-N), 5.57 (1H , dd, *J* = 8.3, 2.1 Hz, C**H**-C(O)-NH), 4.73 (2H, s, *J* = 6.4 Hz, C-C**H**$_2$-NH-C), 4.08 (1H, t, *J* = 5.3 Hz, C**H**-CH-N), 3.95-3.78 (3H, m, C-NH-C**H**$_2$-CH & CH$_2$-CH-C**H**), 3.73-3.61 (1H, m, CH$_2$-C**H**-CH), ^{113}C NMR (101 MHz, DMSO) δ 182.6, 162.9, 162.2 (d, *J* = 233.8 Hz), 150.7, 148.0, 145.4, 145.0, 141.0 (d, *J* = 7.8 Hz), 130.7 (d, *J* = 20.5 Hz), 123.5 (d, *J* = 9.3 Hz), 114.3 (d, *J* = 24.2 Hz), 111.5 (d, *J* = 20.6 Hz), 101.9, 89.2 82.8, 72.3, 70.5, 46.1, 45.42. HRMS (ESI) Exact mass calculated for $C_{20}H_{20}FN_4O_7$ $[M+H]^+$: 447.1311, found: 447.1292.



5c

((2*R*,3*R*,4*S*,5*R*)-3,4-dihydroxy-5-(((2-((4-methoxybenzyl)amino)-3,4-dioxocyclobut-1-en-1-yl)amino)methyl)tetrahydrofuran-2-yl)pyrimidine-2,4(1H,3H)-dione (5c)

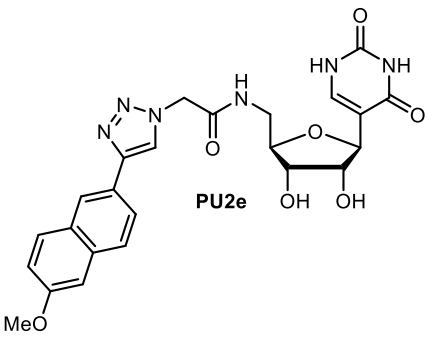
Compound **5c** was prepared using general method F, using 4-methoxybenzylamine (106 µL, 0.81 mmol) and purified by reverse phase HPLC (12 to 35% MeCN/H_2O; 0.1% TFA). The title compound **5c** was obtained as a white fluffy solid (7 mg, 6 %); ^{1}H NMR (400 MHz, DMSO-d_6) δ = 11.4 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 7.88-7.74 (1H, m, N**H**), 7.68 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O), 7.52 (1H, br s, N**H**), 7.32 (2H, d, *J* = 8.3 Hz, Ar**H**), 6.99 (2H, d, *J* = 8.3 Hz, Ar**H**), 5.84 (1H, d, *J* = 5.6 Hz, CH-O-C**H**-N), 5.65 (1H, dd, *J*=8.1, 2.1, 1H), 5.54 (1H, br s, O**H),** 5.34 (1H, br s, O**H**), 4.84-4.58 (2H, m, NH-C**H**$_2$-C=C), 4.20-4-12 (1H, m, C**H**-CH-N), 4.07-3.88 (3H, m, C-NH-C**H**$_2$-CH & C**H**-CH-N), 3.81 (3H, s, OC**H**$_3$), 3.80-3.69 (1H, s, C**H**-CH-N); ^{13}C NMR (101 MHz, DMSO) δ 182.3, 162.3, 157.5, 152.8, 147.1, 145.7, 142.9, 131.3, 128.7 114.8, 101.6, 88.5, 82.5, 72.4, 70.5, 55.3, 49.1, 42.8; HRMS (ESI) Exact mass calculated for $C_{21}H_{23}N_4O_8$ $[M+H]^+$: 459.1510, found 459.1473



5f

1-((2R,3R,4S,5R)-5-(((3,4-dioxo-2-(phenethylamino)cyclobut-1-en-1-yl)amino)methyl)-3,4-dihydroxytetrahydrofuran-2-yl)pyrimidine-2,4(1H,3H)-dione (5f)

Compound **5f** was prepared using general method F, using phenylethylamine (102 μL, 0.81 mmol) and purified by reverse phase HPLC (12 to 35% MeCN/H_2O; 0.1% TFA). The title compound **5f** was obtained as a white fluffy solid (11 mg, 9 %); (400 MHz, DMSO-d_6) δ = 11.38 (1H, d, *J* = 2.3 Hz, N**H**-C(O)-N), 7.63 (1H, d, *J* = 8.1 Hz, C**H**-N-C(O)), 7.44 (2H, br s, 2 ×N**H**), 7.31 (2H, m, 2 × Ar**H**), 7.27-7.16 (3H, m, 3 ×Ar**H**), 5.78 (1H, d, *J* = 5.5 Hz, CH-O-C**H**-N), 5.62 (1H , dd, *J* = 8.0, 2.2 Hz, C**H**-C(O)-NH), 5.47 (1H, br s, O**H),** 5.28 (1H, br s, O**H**), 4.10 (1H, t, *J* = 5.2 Hz, C**H**-CH-N), 3.88 (3H, m, CH_2-C**H**$_2$-NH & CH_2-CH-C**H**), 3.80-3.71 (2H, m, C-NH-C**H**$_2$-CH), 3.70-3.60 (1H, s, CH_2-C**H**-CH-), 2.84 (2H, t, *J* = 7.1 Hz, CC**H**$_2$CH$_2$); ^{13}C NMR (101 MHz, DMSO) δ 182.6, 167.8, 167.5, 162.9, 150.7, 141.2, 138.5, 128.8, 128.4, 126.3, 102.0, 88.4, 82.9, 72.3, 70.4, 45.4, 44.6, 37.0; HRMS (ESI) Exact mass calculated for $C_{21}H_{23}N_4O_7$ $[M+H]^+$: 443.1561, found 443.1565

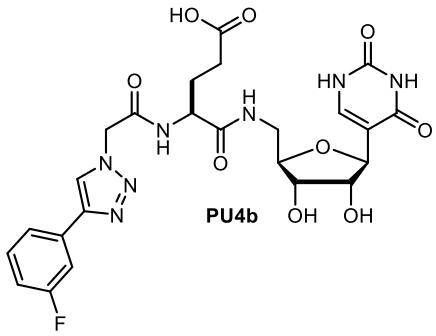


PU2e

N-(((2R,3S,4R,5S)-5-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)-2-(4-(6-methoxylen-2-yl)-1H-1,2,3-triazol-1-yl)acetamide (PU2e)

Compound **PU2e** was prepared using a modified general method C, starting from 0.27 mmol of PU**10** instead of **10** and using 2-ethynyl-6-methoxynaphthalene (246 mg, 1.35 mmol). The compound was purified by reverse phase HPLC (24 to 40% MeCN/H_2O; 0.1% TFA). The title compound **PU2e** was obtained as a white fluffy solid (8 mg, 5%). ^{1}H NMR 400 MHz, DMSO-d_6) δ = 11.19 (1H, d, *J* = 2.0 Hz, C(O)-N**H**-C(O)), 10.97 (1H, dd, *J* = 5.8, 2.0 Hz, CH-N**H**-C(O)), 8.70-8.45 (2H, m, C**H**-N-N & C(O)-N**H**-CH_2), 8.34 (1H, d, *J* = 1.6 Hz, Ar**H**), 7.95 (1H, dd, *J* = 8.6, 1.6 Hz, Ar**H**), 7.89 (2H, app d, *J* = 8.6 Hz, Ar**H**), 7.44 (1H, d, *J* = 5.9 Hz, C**H**-C-C(O)), 7.34 (1H, d, *J* = 2.5 Hz, Ar**H**), 7.19 (1H, dd, *J* = 8.9, 2.5 Hz, Ar**H**), 5.20 (2H, s, N-N-C**H**$_2$), 4.46 (1H, d, *J* = 4.8 Hz, CH-O-C**H**-C), 4.04 (1H, app t, *J* = 4.5 Hz, C**H**-CH-C),

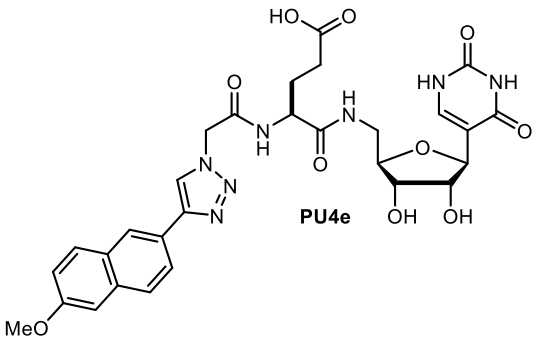
3.89 (3H, s, OC**H**$_3$), 3.84-3.68 (2H, m, CH$_2$-C**H** & CH$_2$-CH-C**H**), Proton 5$_A$' and 5$_B$' are obscured by the residual water signal; ^{13}C NMR (101 MHz, DMSO) δ 165.7, 163.9, 157.7, 151.4, 146.5, 140.5, 134.1, 129.8, 128.8, 127.6, 126.2, 124.3, 123.6, 123.1, 119.3, 110.6, 106.2, 81.3, 80.1, 73.4, 72.5, 55.5, 52.0, 41.7. HRMS (ESI) Exact mass calculated for $C_{24}H_{25}N_6O_7$ $[M+H]^+$: 509.1779, found 509.1809



PU4b

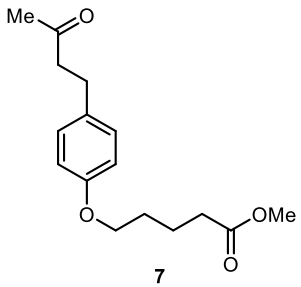
(S)-5-((((2R,3S,4R,5S)-5-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-4-(2-(4-(3-fluorophenyl)-1H-1,2,3-triazol-1-yl)acetamido)-5-oxopentanoic acid (PU4b).

Compound **PU4b** was prepared using the modified general method E, starting from 0.30 mmol of PU**10** instead of **10** and using 1-ethynyl-3-fluorobenzene (155 µL, 1.35 mmol). The compound was purified by reverse phase HPLC (12 to 36% MeCN/H$_2$O; 0.1% TFA). The title compound **PU4b** was obtained as a white fluffy solid (12 mg, 7%). ^{1}H NMR (400 MHz, DMSO-d$_6$) δ = 12.13 (1H, br s, C(O)O**H**), 11.33 (1H, d, *J* = 2.2 Hz, N**H**-C(O)-N), 8.64 (1H, d, *J* = 8.1 Hz, C(O)-N**H**-CH), 8.59 (1H, s, C**H**-N-N), 8.30 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH$_2$), 7.69 (3H, m, 2×Ar**H** & C**H**-N-C(O)), 7.50 (1H, app td, *J* = 8.0, 6.1 Hz, Ar**H**), 7.17 (1H, m, Ar**H**), 5.24 (2H, s, N-N-C**H**$_2$), 5.17 (1H, d, *J* = 5.1 Hz, O**H**), 4.48 (1H, d, *J* = 5.2 Hz, CH-O-C**H**-C), 4.37-4.29 (1H, m, NH-C**H**-CH$_2$), 4.05 (1H, app q, *J* = 5.6 Hz, C**H**-CH-N), 3.88-3.78 (2H, m, NH-CH$_2$-C**H** & CH$_2$-CH-C**H**), 3.48-3.34 (1H, m, NH-C**H**$_A$CH$_B$), 3.30-3.23 (1H, m, NH-CH$_A$C**H**$_B$), 2.25 (2H, ddd, *J* = 9.2, 6.5, 3.0 Hz, C**H**$_2$-C(O)OH), 1.98-1.87 (1H, m, C**H**$_A$CH$_B$-CH$_2$-COOH), 1.83-1.71 (1H, m, C**H**$_A$CH$_B$-CH$_2$-COOH); ^{13}C NMR (101 MHz, DMSO) δ 173.8, 170.8, 165.3, 163.0, 162.6 (d, *J* = 241.2 Hz), 151.1, 145.0 (d, *J* = 3.0 Hz), 140.2, 133.1 (d, *J* = 9.5 Hz), 131.1 (d, *J* = 8.8 Hz), 123.7, 121.2 (d, *J* = 3.2 Hz), 114.5 (d, *J* = 24.6 Hz), 111.7 (d, *J*= 23.4 Hz) 110.4, 81.2, 79.7, 73.3, 72.2, 52.2, 51.7, 41.4, 30.1, 27.8. HRMS (ESI) Exact mass calculated for $C_{24}H_{27}FN_7O_9$ $[M+H]^+$: 576.1849, found: 576.1795



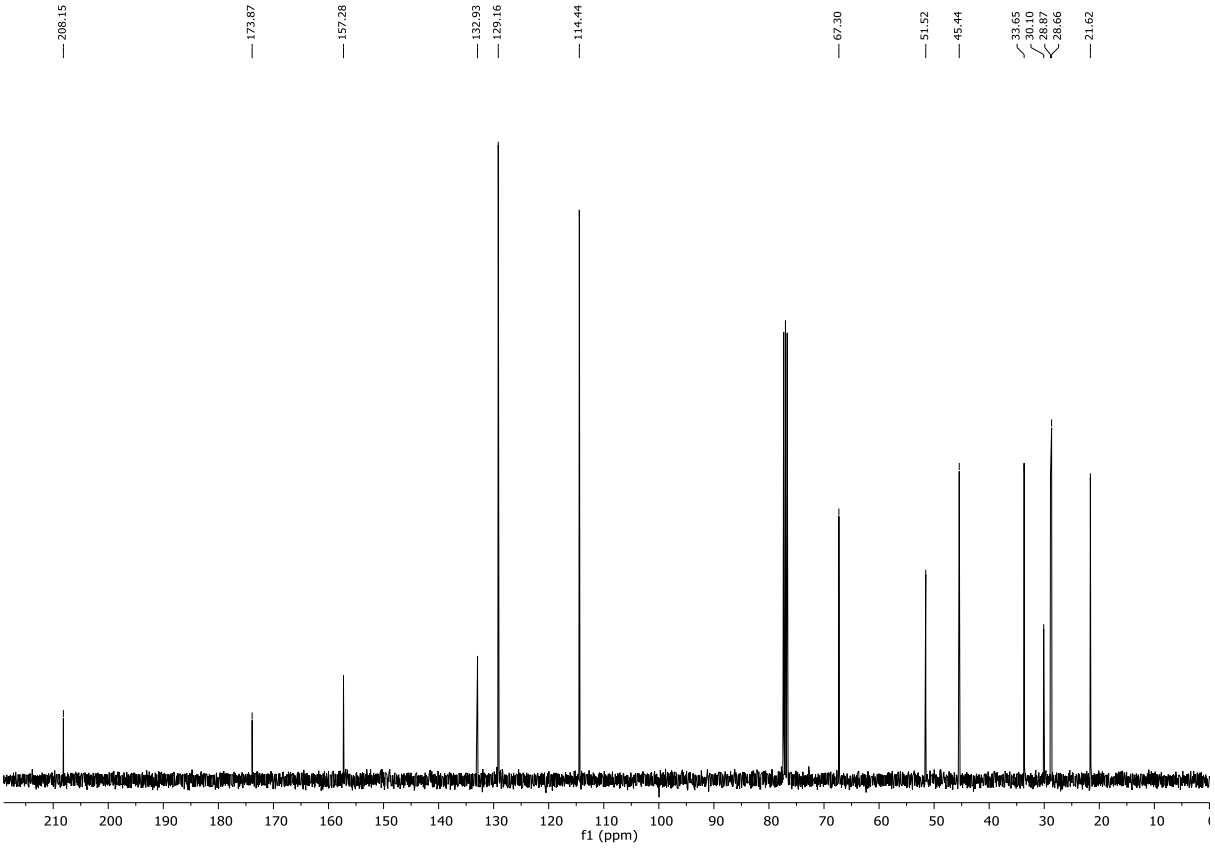
(S)-5-((((2R,3S,4R,5S)-5-(2,4-dioxo-1,2,3,4-tetrahydropyrimidin-5-yl)-3,4-dihydroxytetrahydrofuran-2-yl)methyl)amino)-4-(2-(4-(6-methoxylen-2-yl)-1H-1,2,3-triazol-1-yl)acetamido)-5-oxopentanoic acid (PU4e)

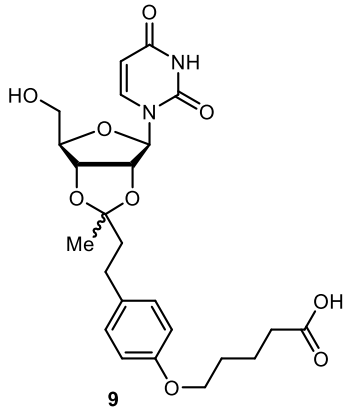
Compound **PU4e** was prepared wusing the modified general method **E**, starting from 0.30 mmol of **PU10** instead of **10** and using 2-ethynyl-6-methoxynaphthalene (246 mg, 1.35 mmol). The compound was purified by reverse phase HPLC (20 to 36% MeCN/H_2O; 0.1% TFA). The title compound **PU4e** was obtained as a white fluffy solid (11 mg, 6%). ^{1}H NMR (400 MHz, DMSO-d_6) δ = 12.17 (1H, br s, C(O)O**H**), 11.16 (1H, d, *J* = 2.0 Hz, C(O)-N**H**-C(O)), 10.91 (1H, dd, *J* = 5.9, 2.0 Hz, CH-N**H**-C(O)), 8.64 (1H, d, *J* = 8.2 Hz, C(O)-N**H**-CH), 8.55 (1H, s, C**H**-N-N)), 8.34 (1H, d, *J* = 1.5 Hz, Ar**H**), 8.19 (1H, t, *J* = 5.9 Hz, C(O)-N**H**-CH_2), 7.95 (1H, dd, *J* = 8.5, 1.7 Hz, Ar**H**), 7.88 (2H, app d, *J* = 7.8 Hz, Ar**H**), 7.43 (1H, d, *J* = 5.9 Hz, C**H**-C-C(O)), 7.34 (1H, d, *J* = 2.5 Hz, Ar**H**), 7.19 (1H, dd, *J* = 8.9, 2.5 Hz, Ar**H**), 5.26 (2H, s, N-N-C**H**$_2$), 4.41 (1H, d, *J* = 5.1 Hz, CH-O-C**H**-C), 4.34 (1H, td, *J* = 8.1, 5.4 Hz, NH-C**H**-CH_2), 4.00 (1H, app t, *J* = 4.7 Hz, C**H**-CH-C), 3.89 (3H, s, OC**H**$_3$), 3.80-3.67 (2H, m, CH_2-C**H** & CH_2-CH-C**H**), 2.27 (2H, ddd, *J* = 9.0, 6.6, 2.4 Hz, C**H**$_2$-C(O)OH), 2.01=1.88 (1H, m, C**H**$_A$C**H**$_B$-CH_2-COOH), 1.87-1.72 (1H, m, C**H**$_A$C**H**$_B$-CH_2-COOH). 2.27 (1H, ddd, *J* = 9.4, 6.6, 3.1 Hz, C**H**$_2$-C(O)OH), 1.99-1.89 (1H, m, C**H**$_A$C**H**$_B$-CH_2-COOH), Proton 5_A' and 5_B' are obscured by the residual water signal; ^{13}C NMR (101 MHz, DMSO) δ 174.1, 171.0, 165.7, 163.8, 157.7, 151.4, 146.5, 140.5, 134.1, 129.8, 128.8, 127.6, 126.2, 124.3, 123.6, 123.1, 119.3, 110.6, 106.2, 81.5, 79.9, 73.5, 72.4, 55.5, 52.4, 51.9, 41.6, 30.3, 28.0. HRMS (ESI) Exact mass calculated for $C_{29}H_{32}N_7O_{10}$ $[M+H]^+$:638.2205, found: 638.2226.


Me
O
O
O
OMe
O

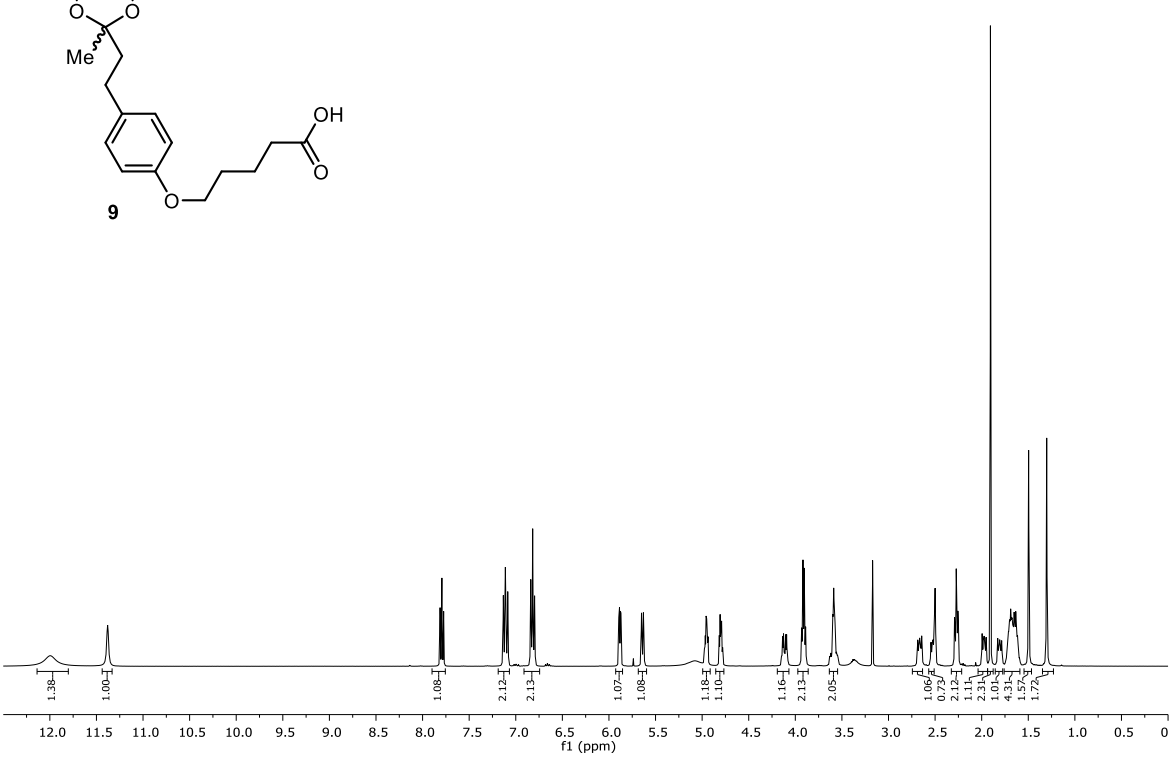
7

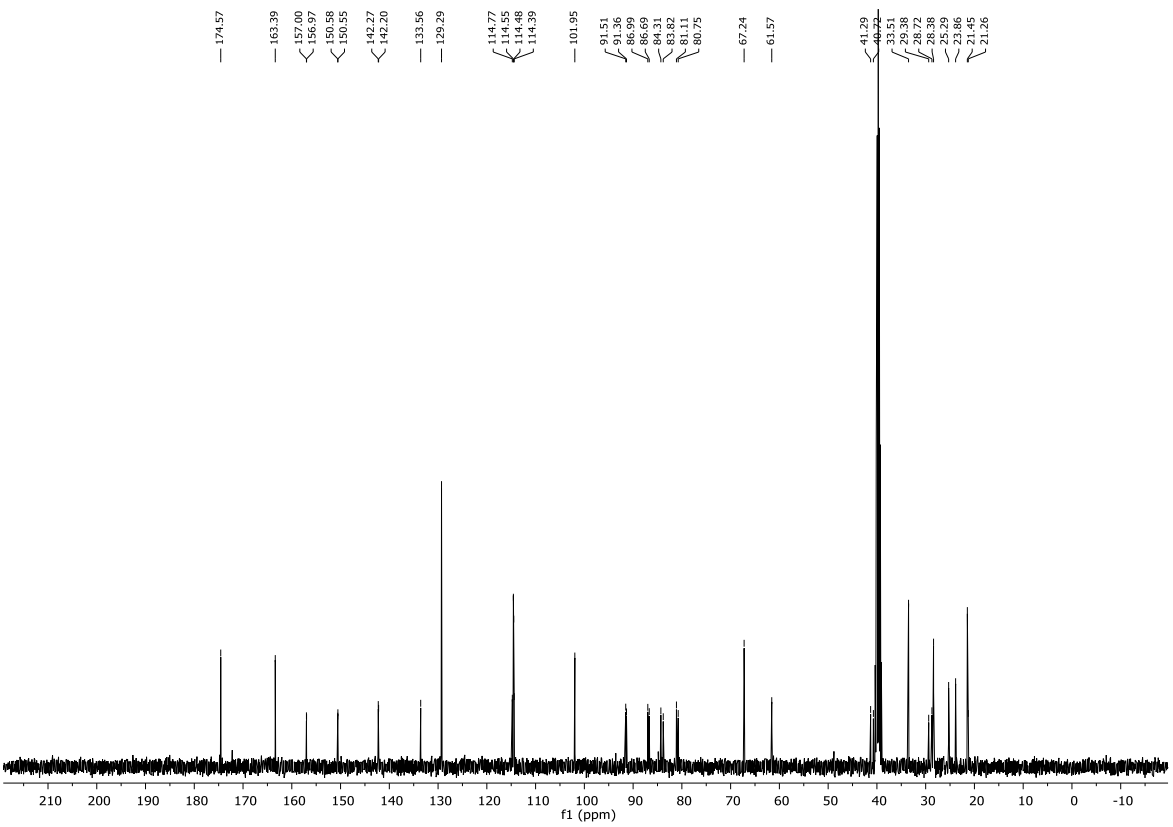
1.98
2.00
2.02
2.94
2.00
2.03
2.00
2.95
4.02
9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

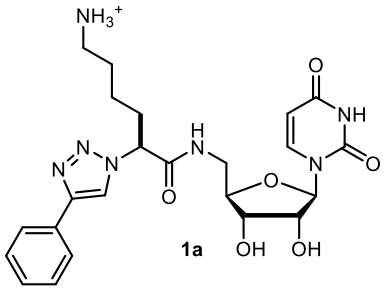

208.15
173.87
157.28
132.93
129.16
114.44
67.30
51.52
45.44
33.65
30.10
28.87
28.66
21.62
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


O
NH
HO
N
O
O
O
O
Me
OH
O
O

9


1.38
1.00
1.08
2.12
2.13
1.07
1.08
1.18
1.10
1.16
2.13
2.05
1.06
0.73
2.12
1.11
2.31
1.01
4.31
1.57
1.72
12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)


174.57
163.39
157.00
156.97
150.58
150.55
142.27
142.20
133.56
129.29
114.77
114.55
114.48
114.39
101.95
91.51
91.36
86.99
86.69
84.31
83.82
81.11
80.75
67.24
61.57
41.29
40.72
33.51
29.38
28.72
28.38
25.29
23.86
21.45
21.26
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)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NH
H
N
N
N
N
O
O
O
1a
OH
OH

1.00
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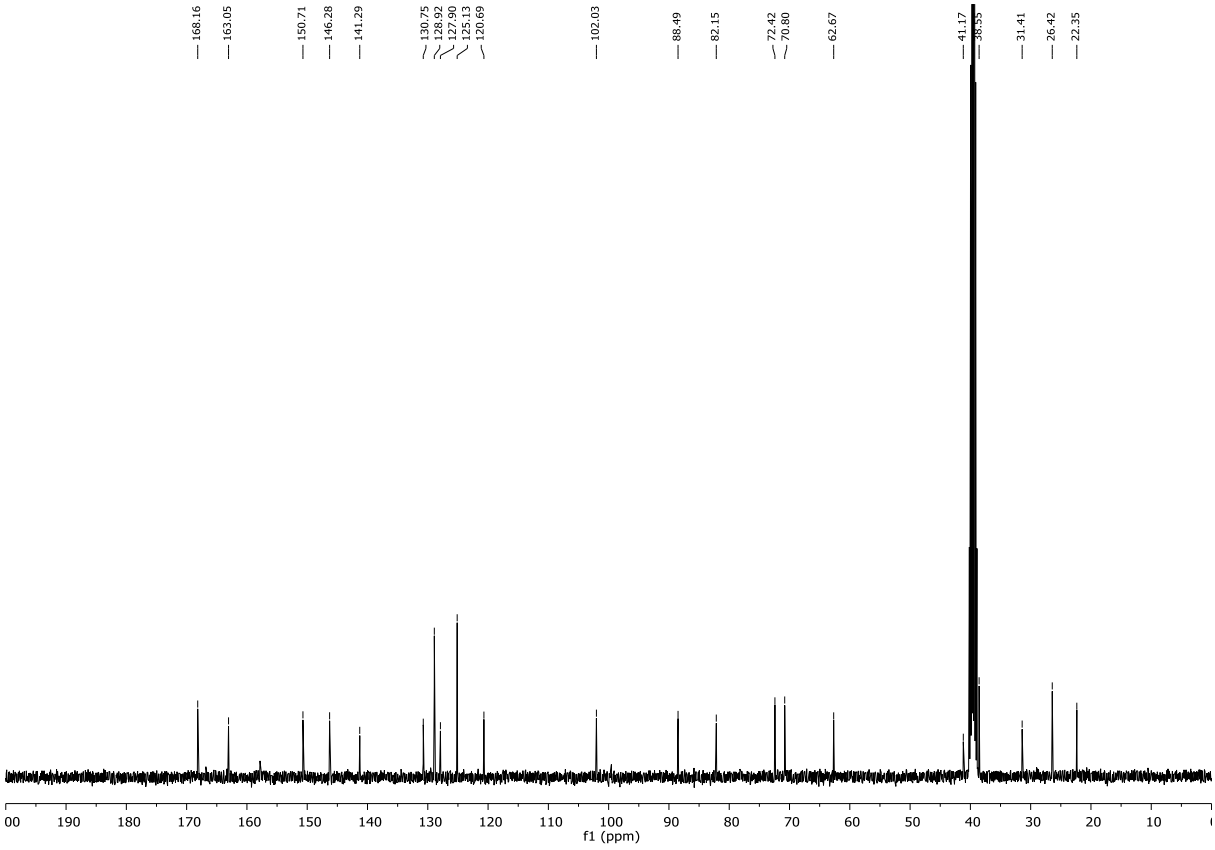
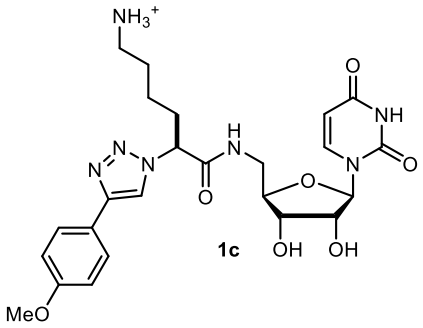
2.14
2.86
1.10
2.07
1.10
1.13
1.07
0.66
1.03
0.74
1.05
1.05
1.14
1.52
2.16
2.20
2.20
2.34
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)


168.16
163.05
150.71
146.28
141.29
130.75
128.92
127.90
125.13
120.69
102.03
88.49
82.15
72.42
70.80
62.67
41.17
38.55
31.41
26.42
22.35
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)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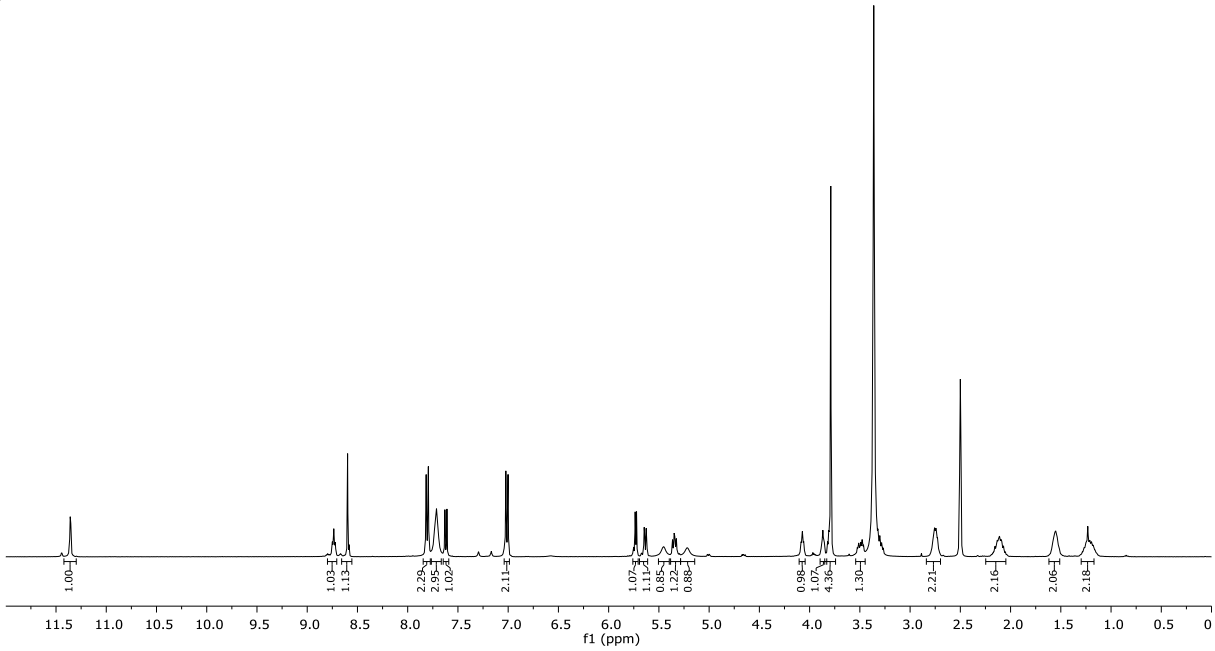
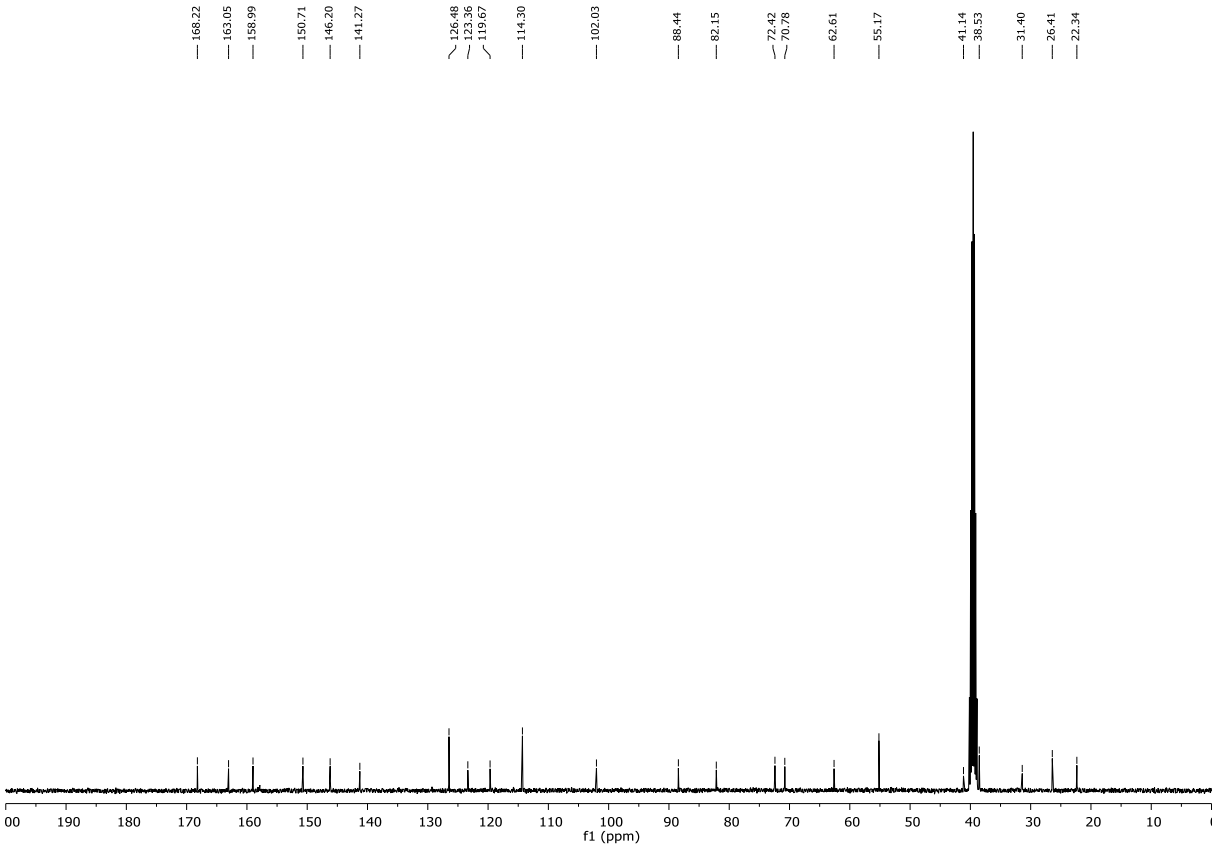
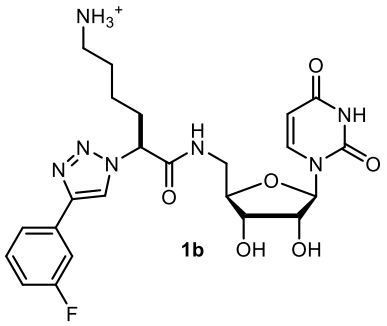
O
NH
H
N
N
N
O
O
N
O
O
1c
OH
OH
MeO


1.00
1.03
1.13
2.29
2.95
1.02
2.11
1.07
1.11
0.95
1.22
0.88
0.98
1.07
4.36
1.30
2.21
2.16
2.06
2.18
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)


168.22
163.05
158.99
150.71
146.20
141.27
126.48
123.36
119.67
114.30
102.03
88.44
82.15
72.42
70.78
62.61
55.17
41.14
38.53
31.40
26.41
22.34
00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)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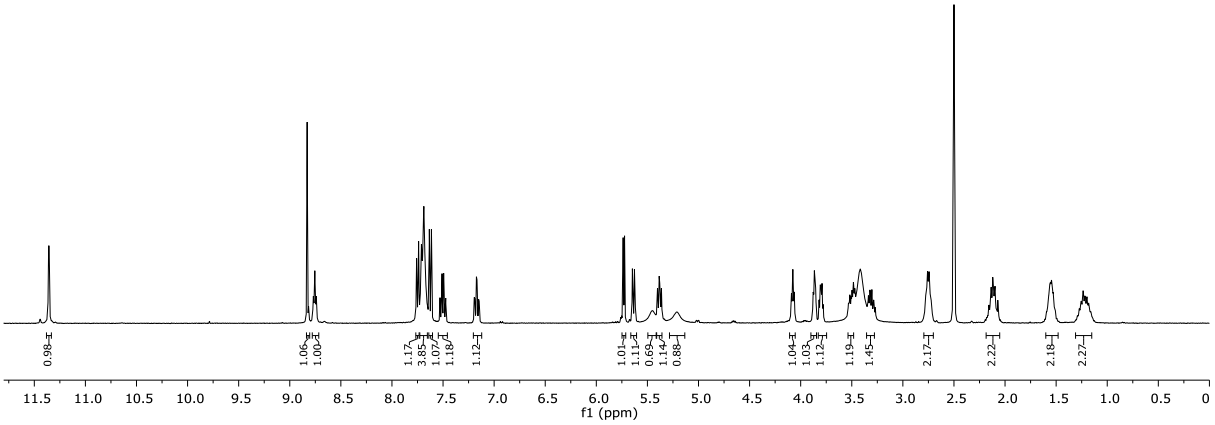
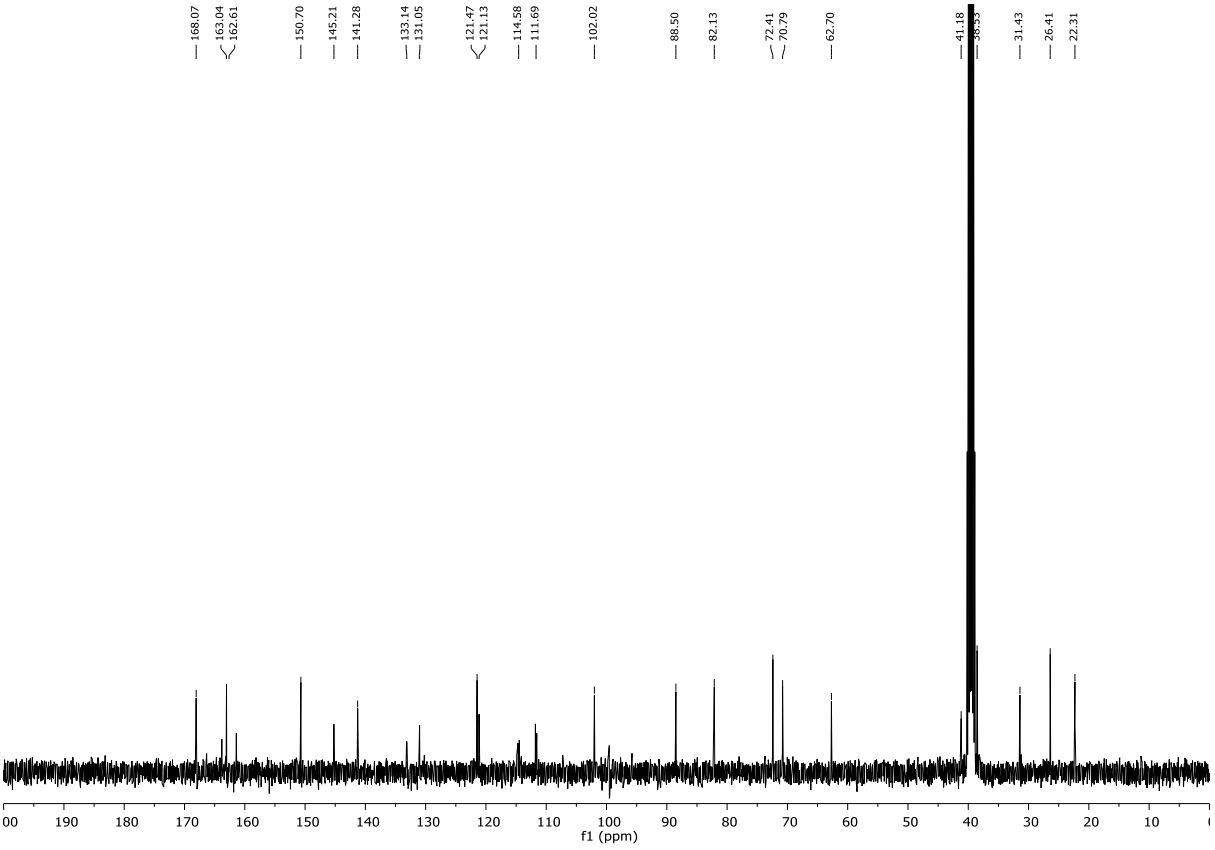
O
NH
H
N
N
N
N
O
O
O
O
OH
OH
F

1b


0.98
1.06
1.00
1.17
3.85
1.07
1.18
1.12
1.01
1.11
0.69
1.14
0.88
1.04
1.03
1.12
1.19
1.45
2.17
2.22
2.18
2.27
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)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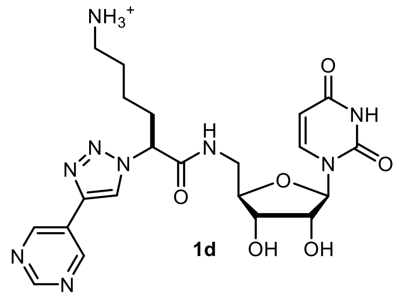
163.04
162.61
150.70
145.21
141.28
133.14
131.05
121.47
121.13
114.58
111.69
102.02
88.50
82.13
72.41
70.79
62.70
41.18
38.53
31.43
26.41
22.31
00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)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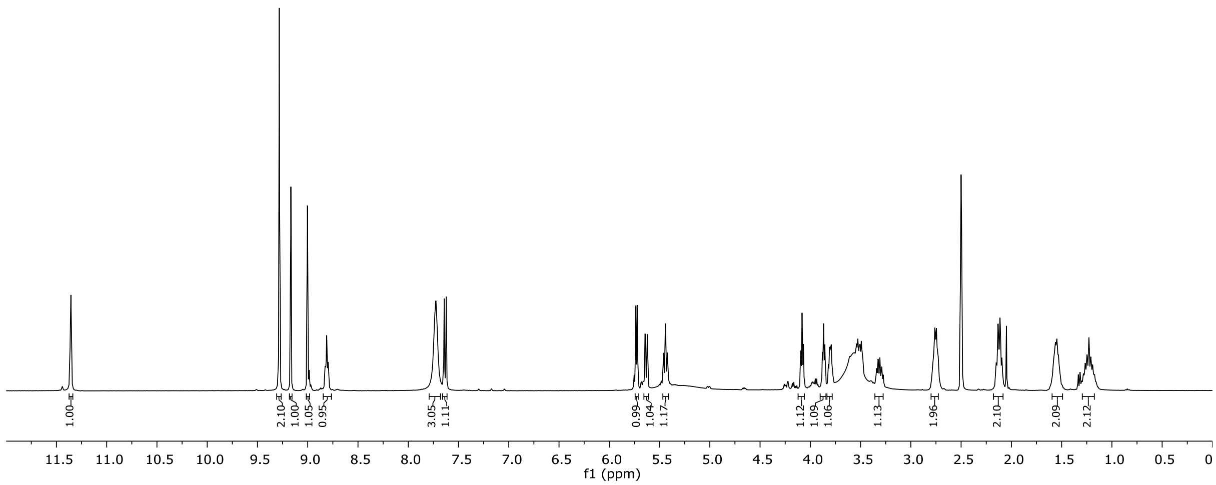
N
N
N
N
H
N
O
O
O
NH
O
N
OH
OH
N

1d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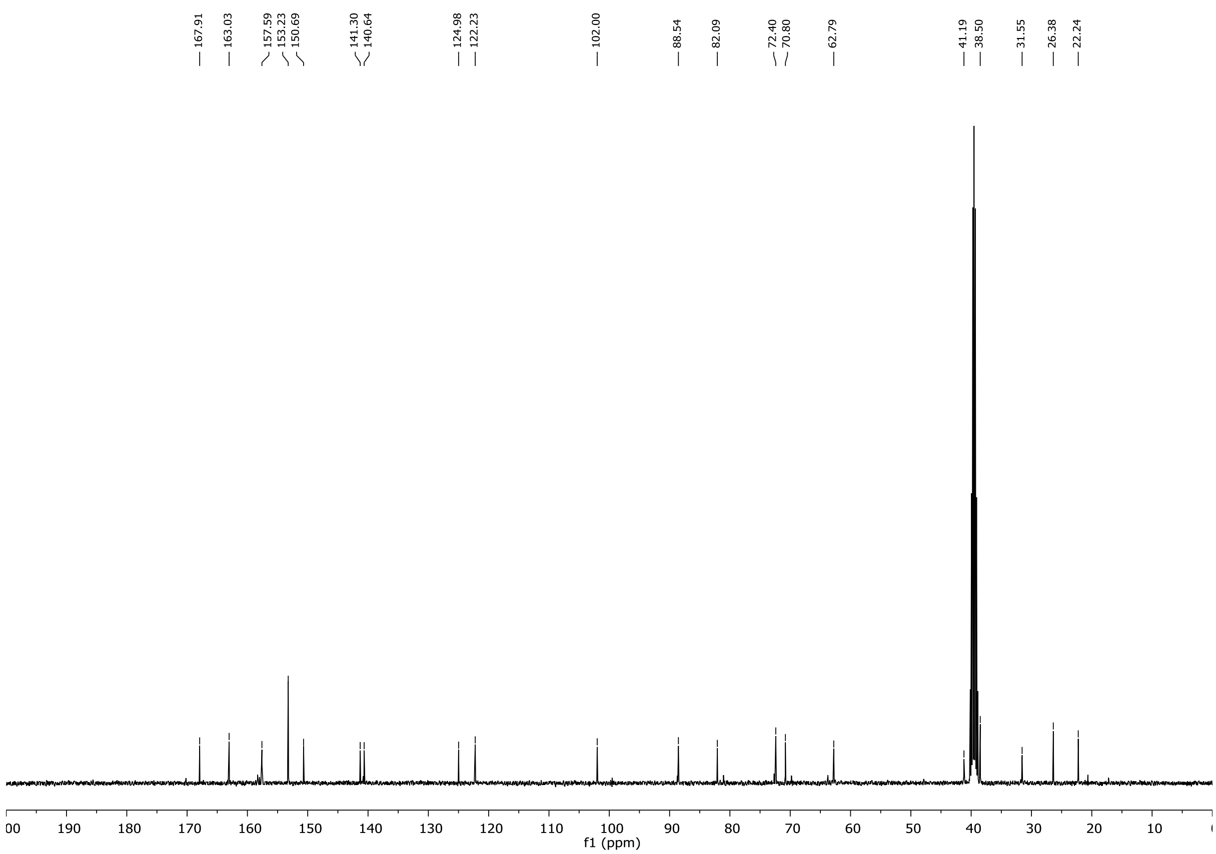
2.10
1.00
1.05
0.95
3.05
1.11
0.99
1.04
1.17
1.12
1.09
1.06
1.13
1.96
2.10
2.09
2.12
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)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153.23
150.69
141.30
140.64
124.98
122.23
102.00
88.54
82.09
72.40
70.80
62.79
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38.50
31.55
26.38
22.24
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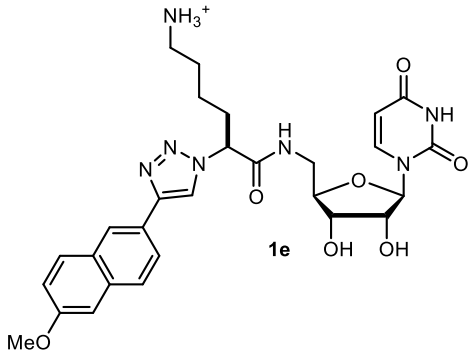
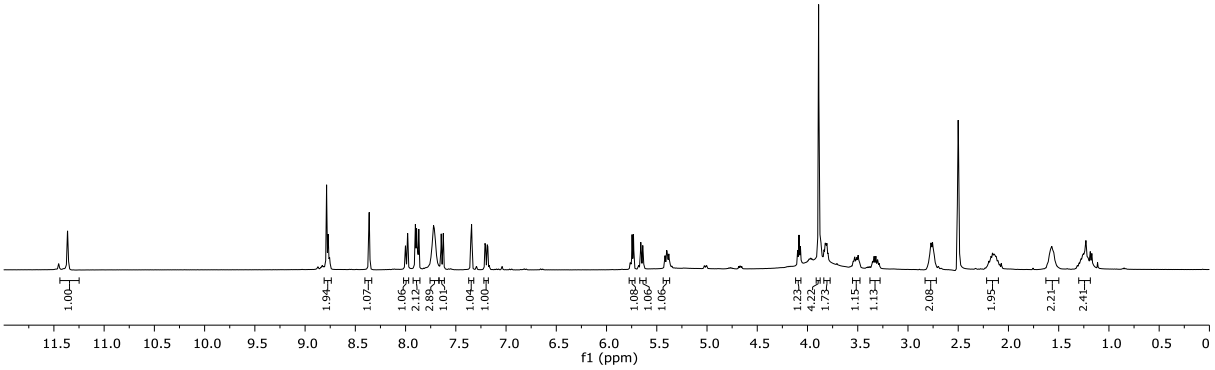
f1 (ppm)


NH3+
O
NH
H
N
N
N
N
O
O
O
N
O
1e
OH
OH
MeO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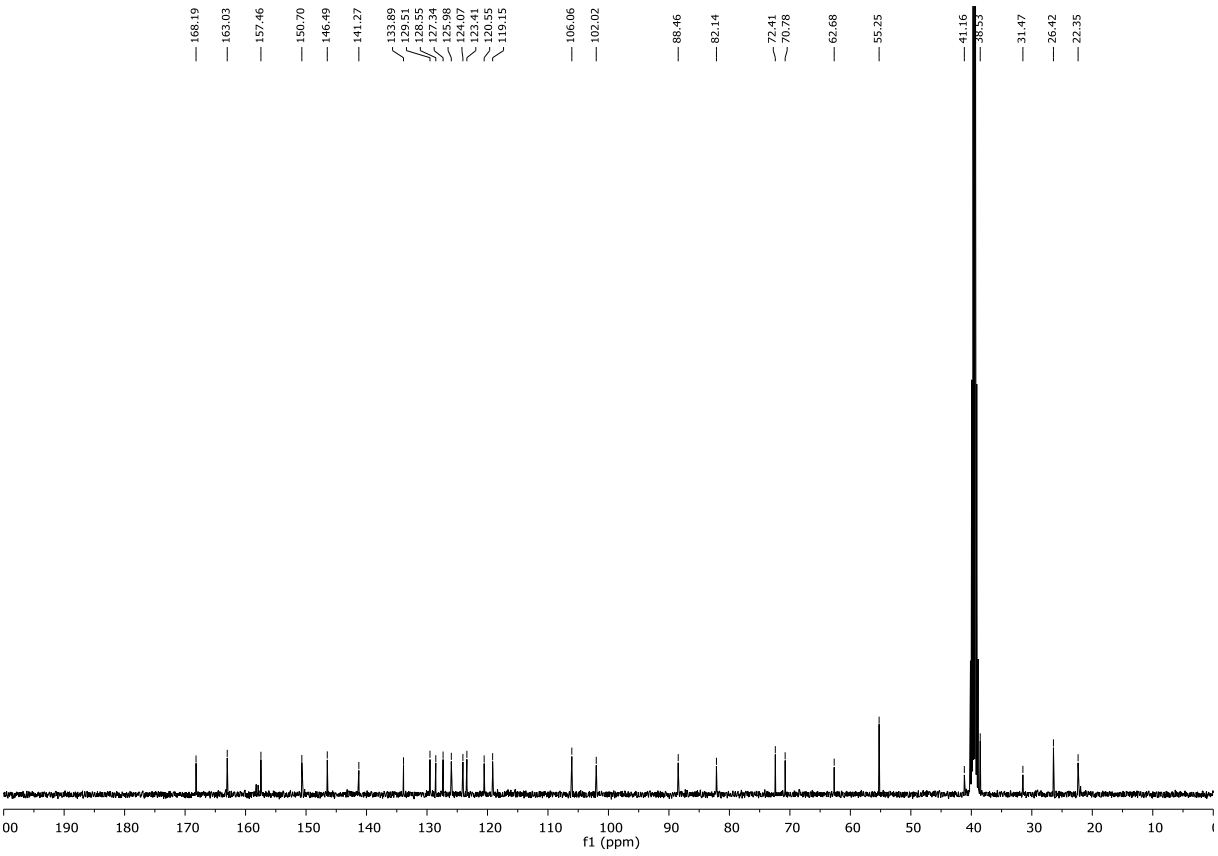
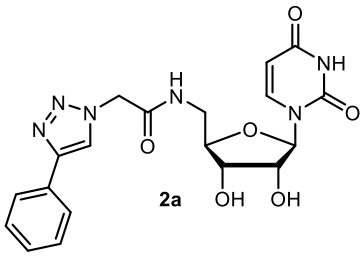
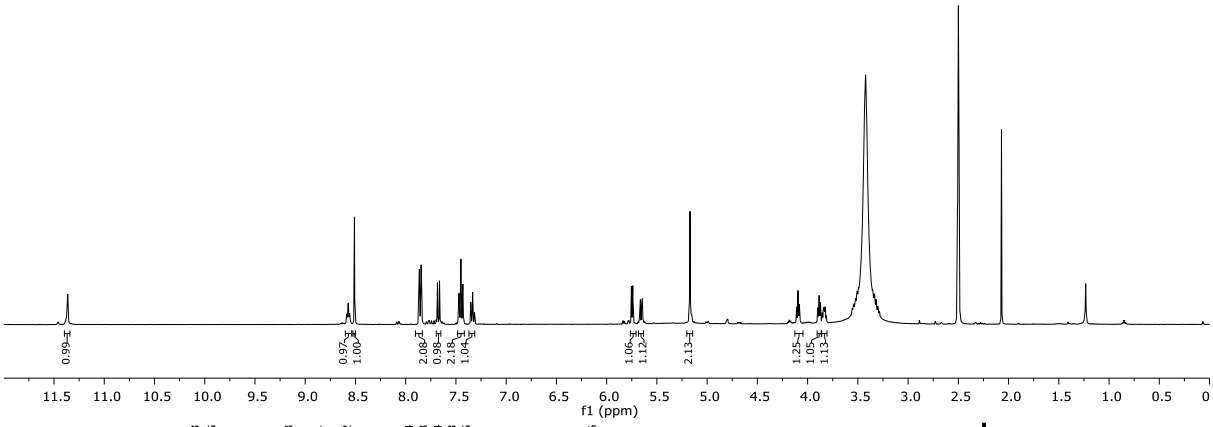
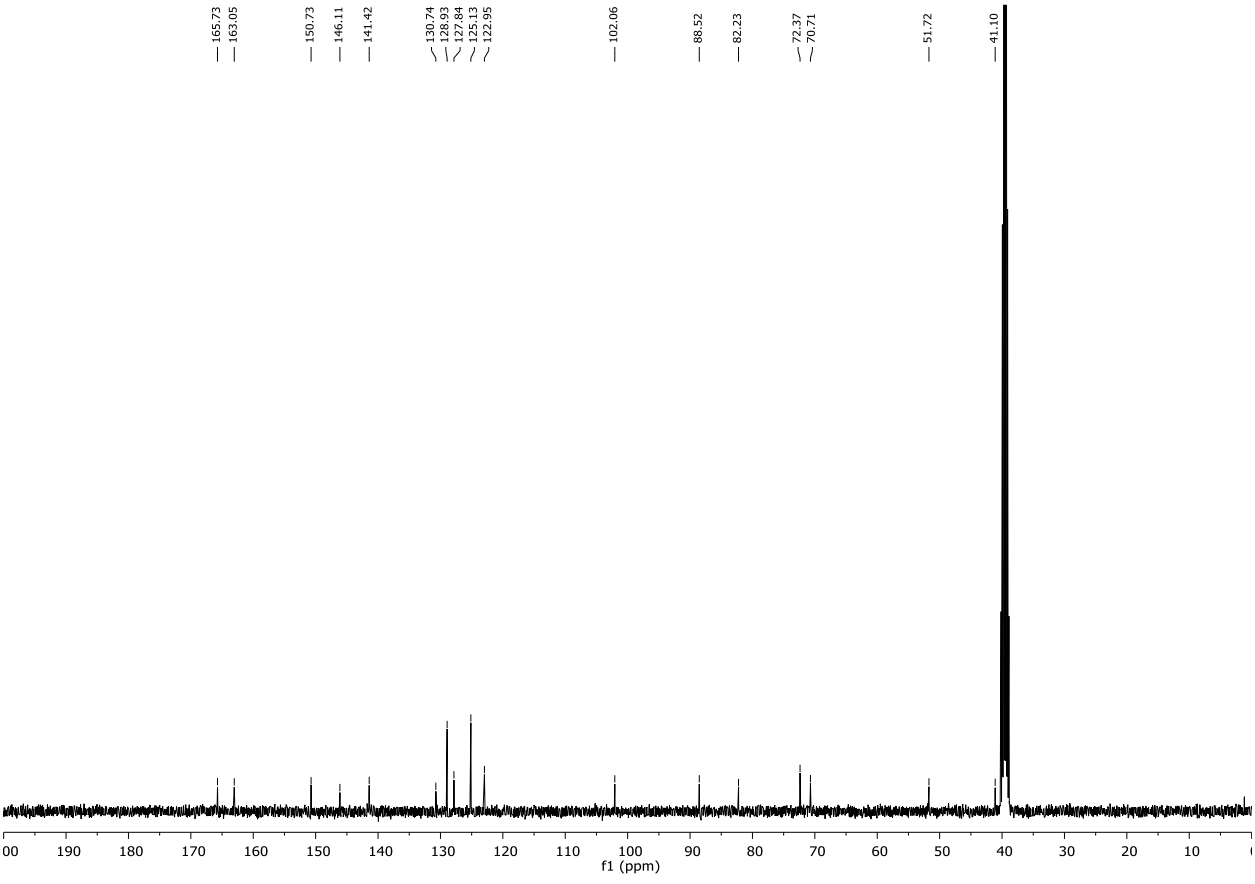
1.06
2.12
2.89
1.01
1.04
1.00
1.08
1.06
1.06
1.23
4.22
1.73
1.15
1.13
2.08
1.95
2.21
2.41
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

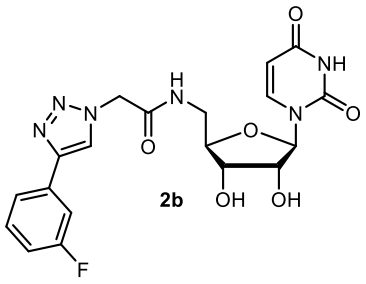

168.19
163.03
157.46
150.70
146.49
141.27
133.89
129.51
128.55
127.34
125.98
124.07
123.41
120.55
119.15
106.06
102.02
88.46
82.14
72.41
70.78
62.68
55.25
41.16
38.53
31.47
26.42
22.35
00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


O
NH
H
N
N
N
N
O
O
O
OH
OH

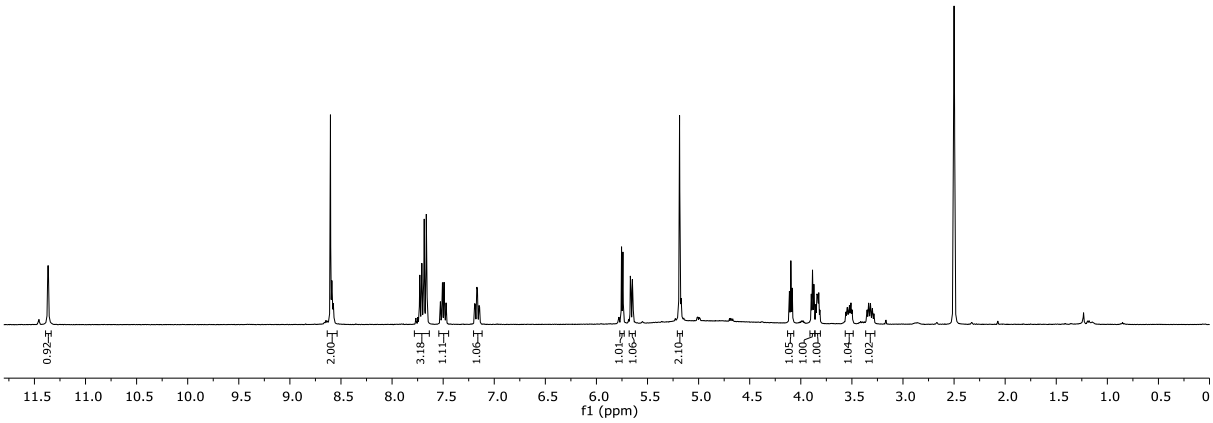
2a

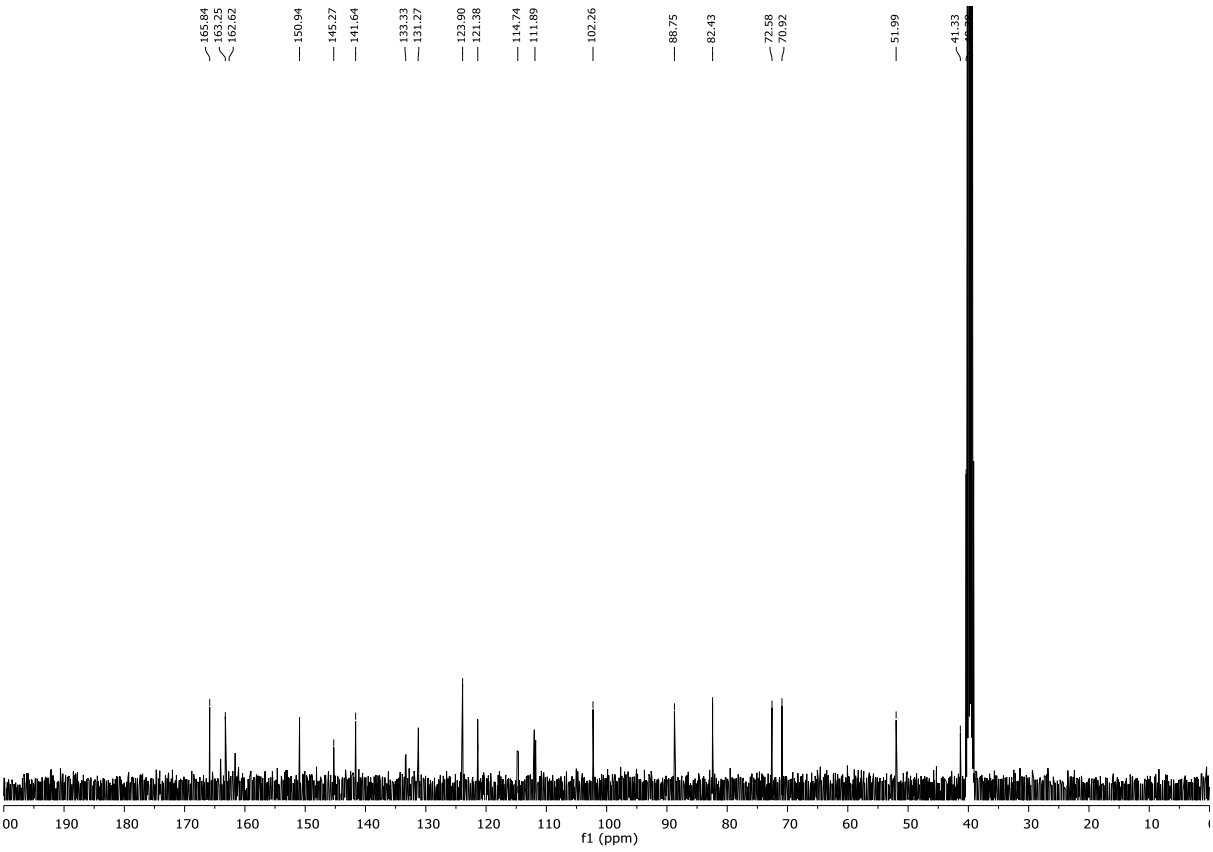

0.99
0.97
1.00
2.08
0.98
2.18
1.04
1.06
1.12
2.13
1.25
1.05
1.13
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

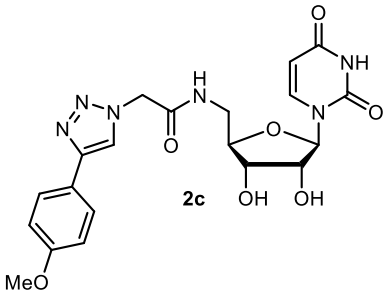

165.73
163.05
150.73
146.11
141.42
130.74
128.93
127.84
125.13
122.95
102.06
88.52
82.23
72.37
70.71
51.72
41.10
00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


O
NH
H
N
N
N
N
O
O
O
N
O
OH
OH
F

2b

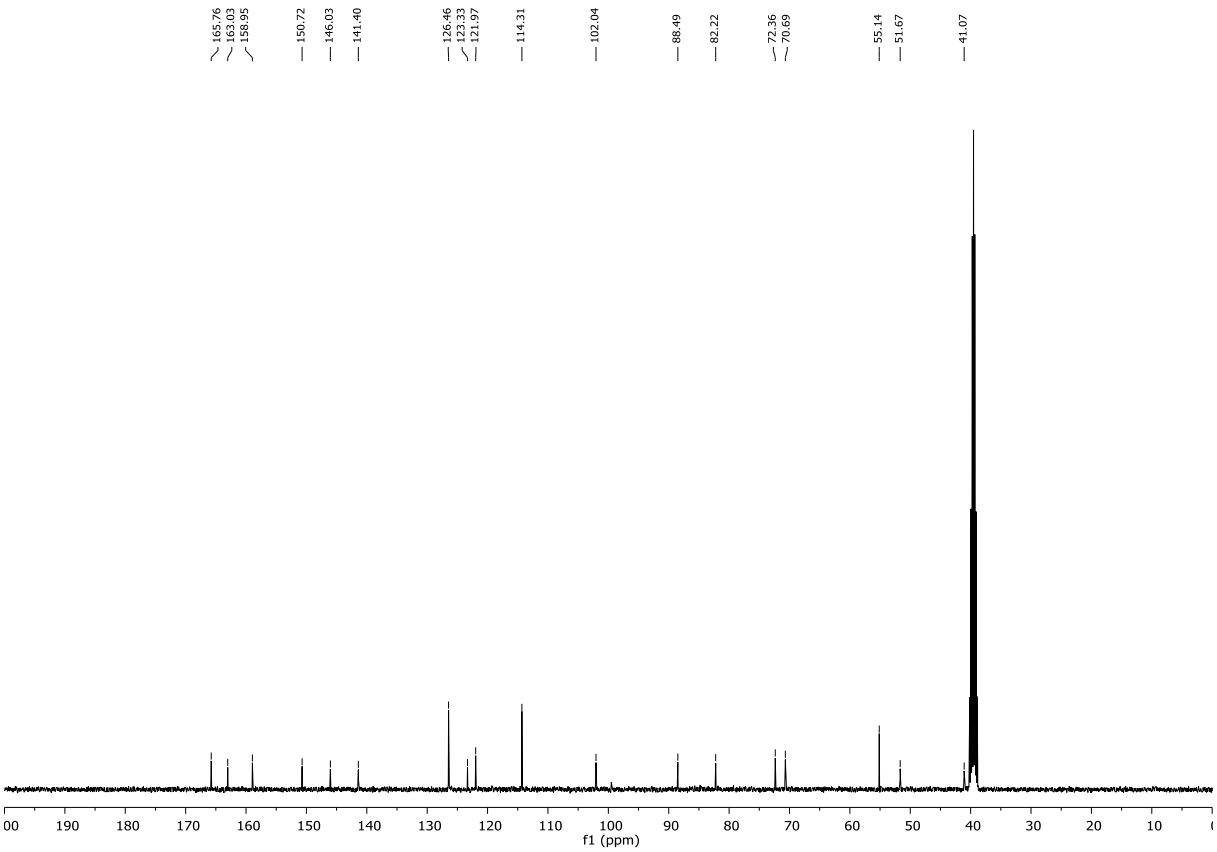

0.92
2.00
3.18
1.11
1.06
1.01
1.06
2.10
1.05
1.00
1.00
1.04
1.02
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

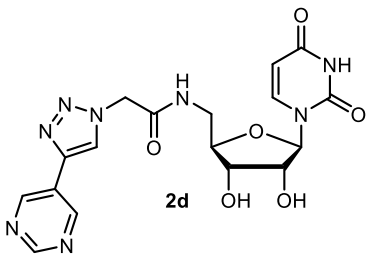

165.84
163.25
162.62
150.94
145.27
141.64
133.33
131.27
123.90
121.38
114.74
111.89
102.26
88.75
82.43
72.58
70.92
51.99
41.33
00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

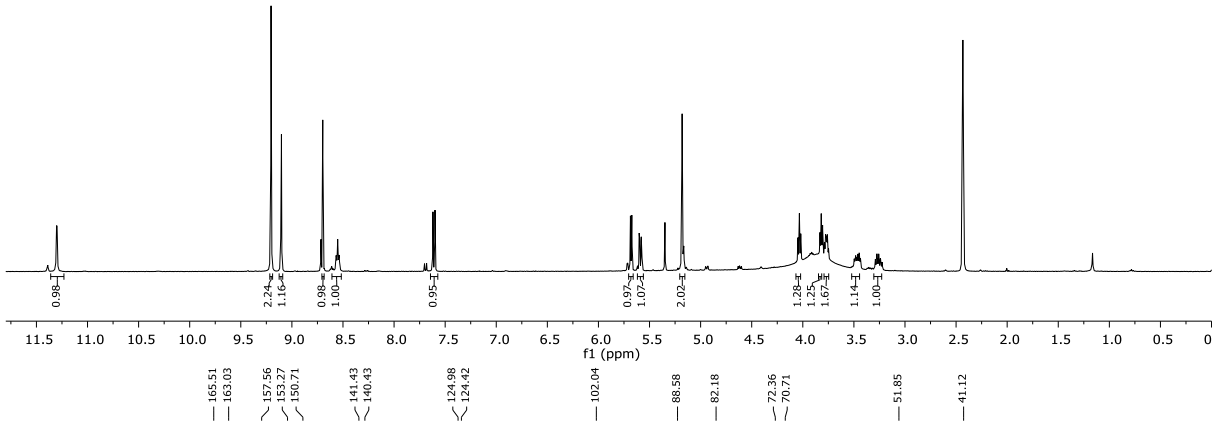

O
NH
H
N
N
N
N
O
O
O
O
OH
OH
MeO

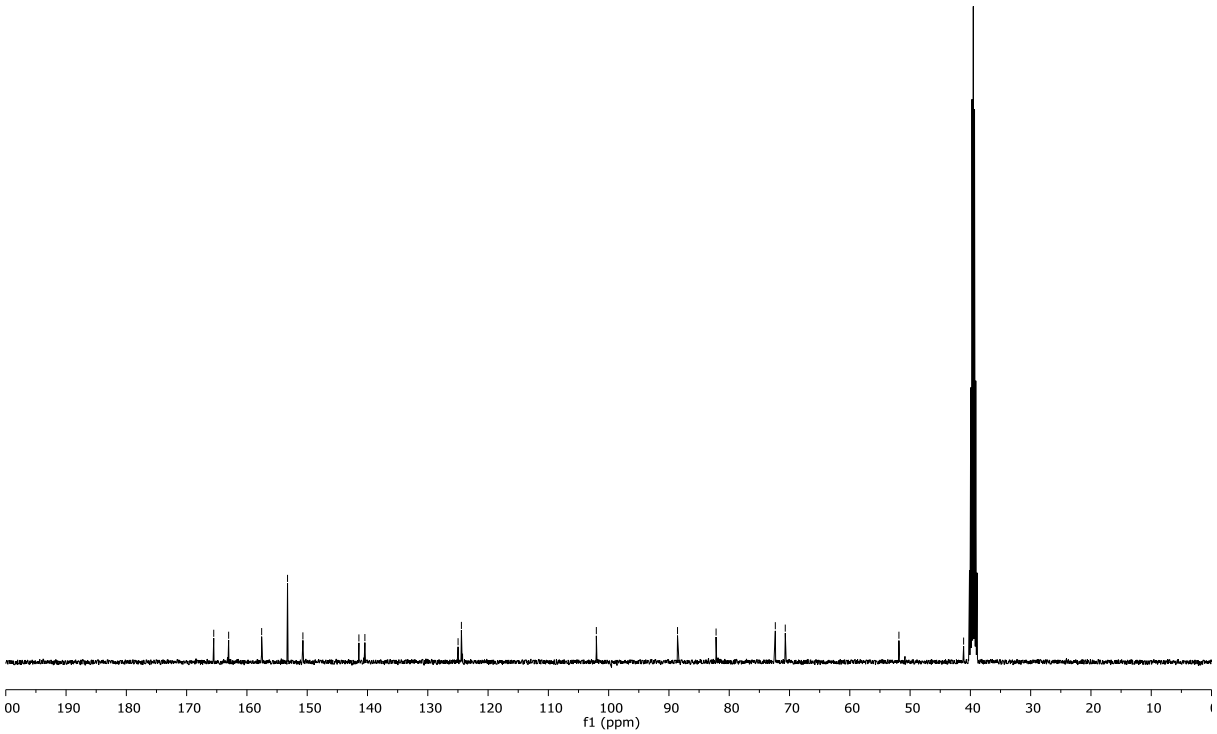
2c

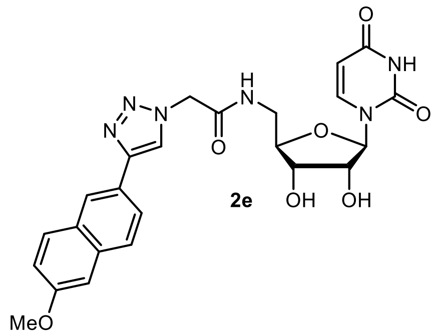
1.00
1.03
1.09
2.10
1.00
2.10
1.02
1.08
2.13
1.06
0.98
1.02
3.05
1.03
1.03
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)


165.76
163.03
158.95
150.72
146.03
141.40
126.46
123.33
121.97
114.31
102.04
88.49
82.22
72.36
70.69
55.14
51.67
41.07
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

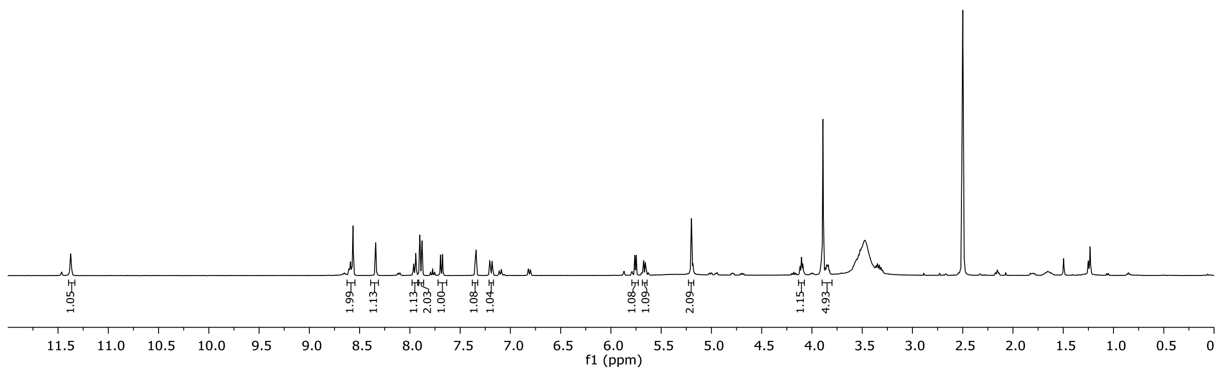

O
NH
H
N
N
N
N
O
O
N
O
O
2d
OH
OH
N
N

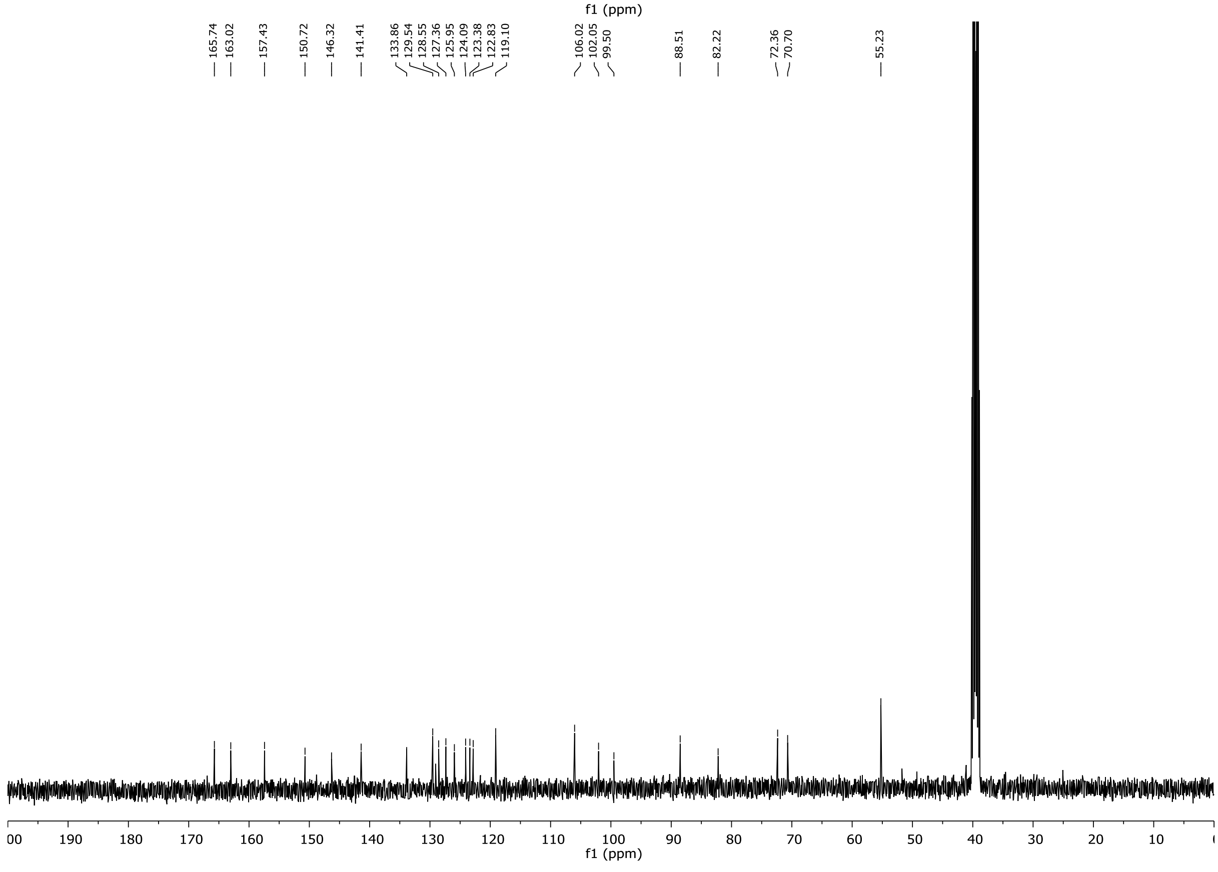

0.98
2.24
1.16
0.98
1.00
0.95
0.97
1.07
2.02
1.28
1.25
1.67
1.14
1.00
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)
165.51
163.03
157.56
153.27
150.71
141.43
140.43
124.98
124.42
102.04
88.58
82.18
72.36
70.71
51.85
41.12

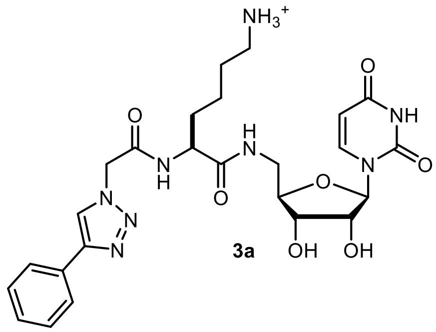

00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


MeO
N
N
N
O
H
N
O
OH
OH
O
NH
O
N
O

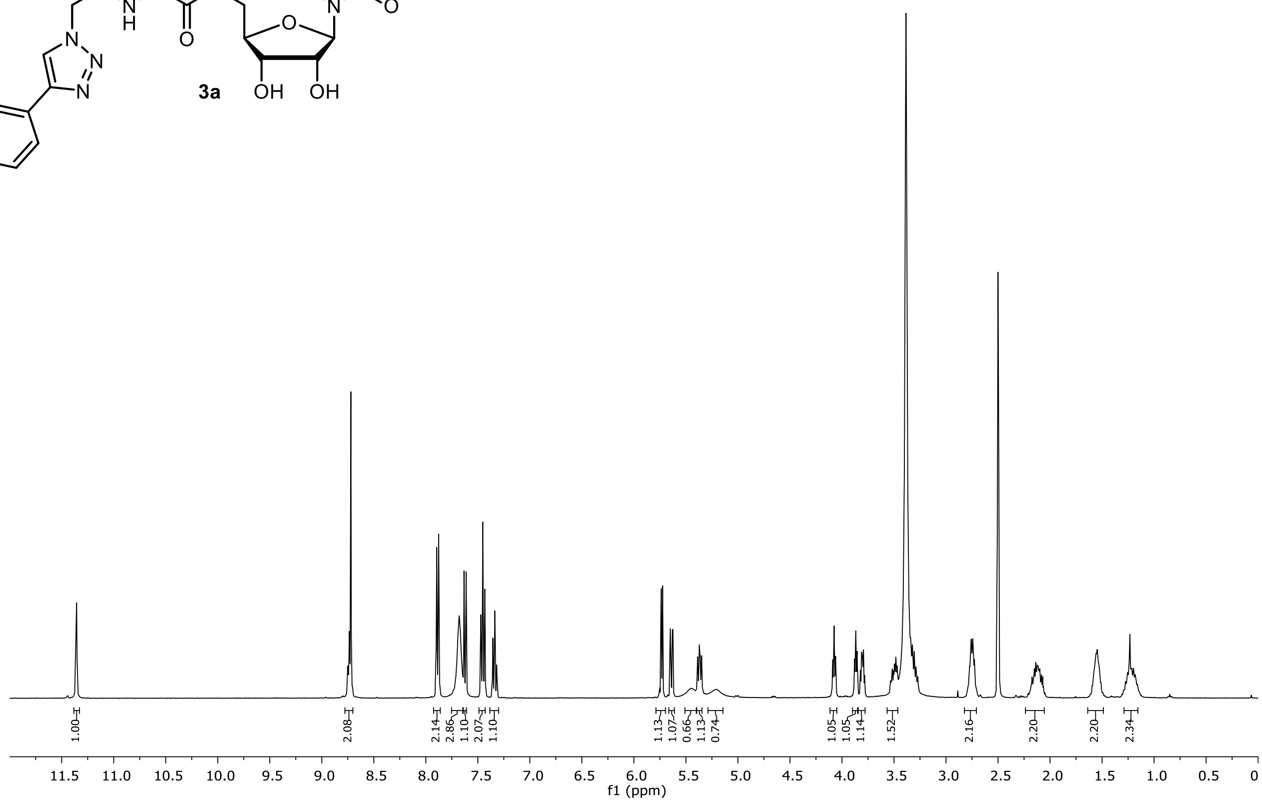
2e

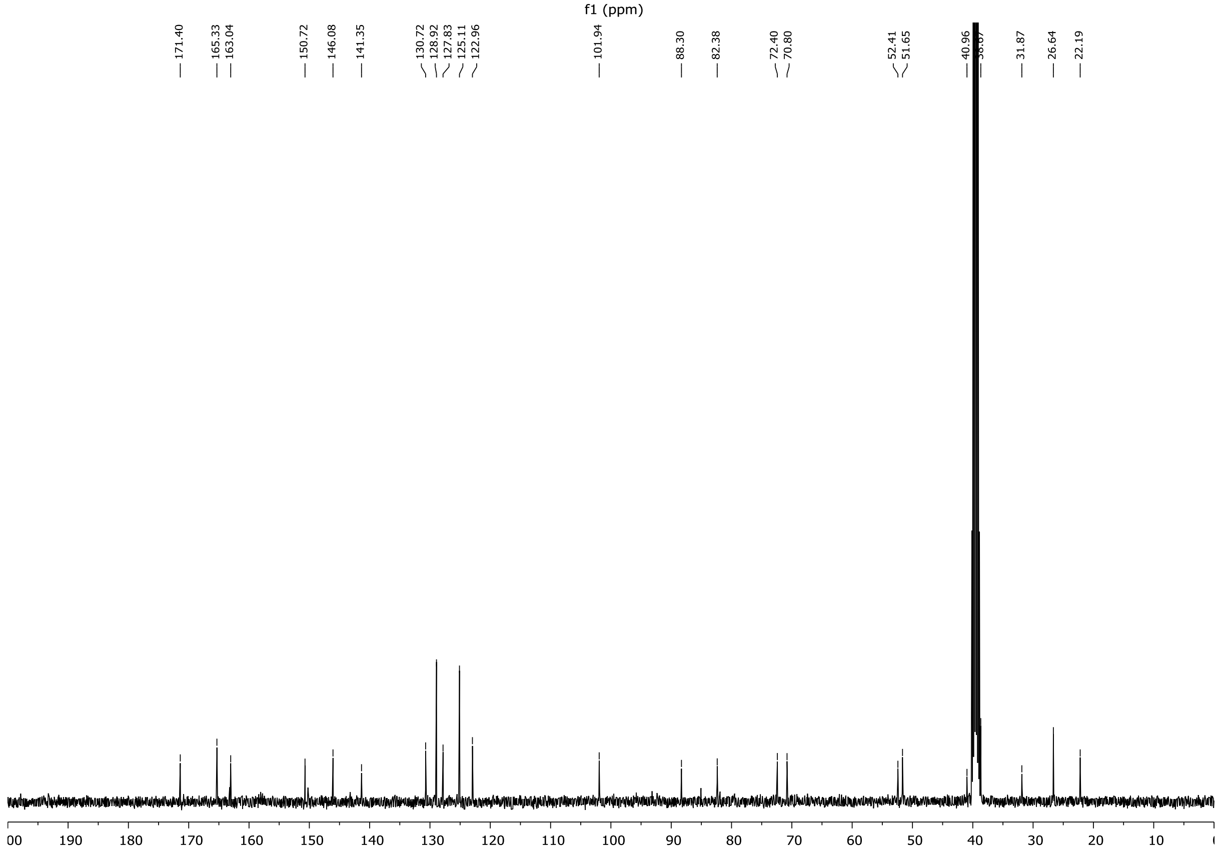

1.05
1.99
1.13
1.13
2.03
1.00
1.08
1.04
1.08
1.09
2.09
1.15
4.93
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

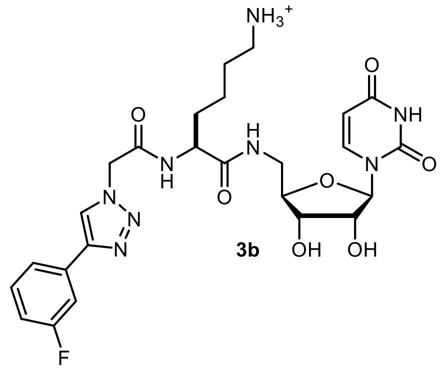

165.74
163.02
157.43
150.72
146.32
141.41
133.86
129.54
128.55
127.36
125.95
124.09
123.38
122.83
119.10
106.02
102.05
99.50
88.51
82.22
72.36
70.70
55.23
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


NH3+
O
NH
O
N
H
O
H
N
O
N
N
N
N
O
OH
OH

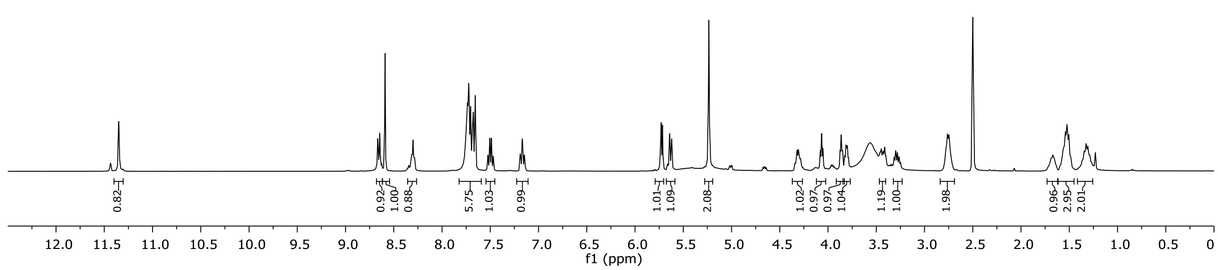
3a

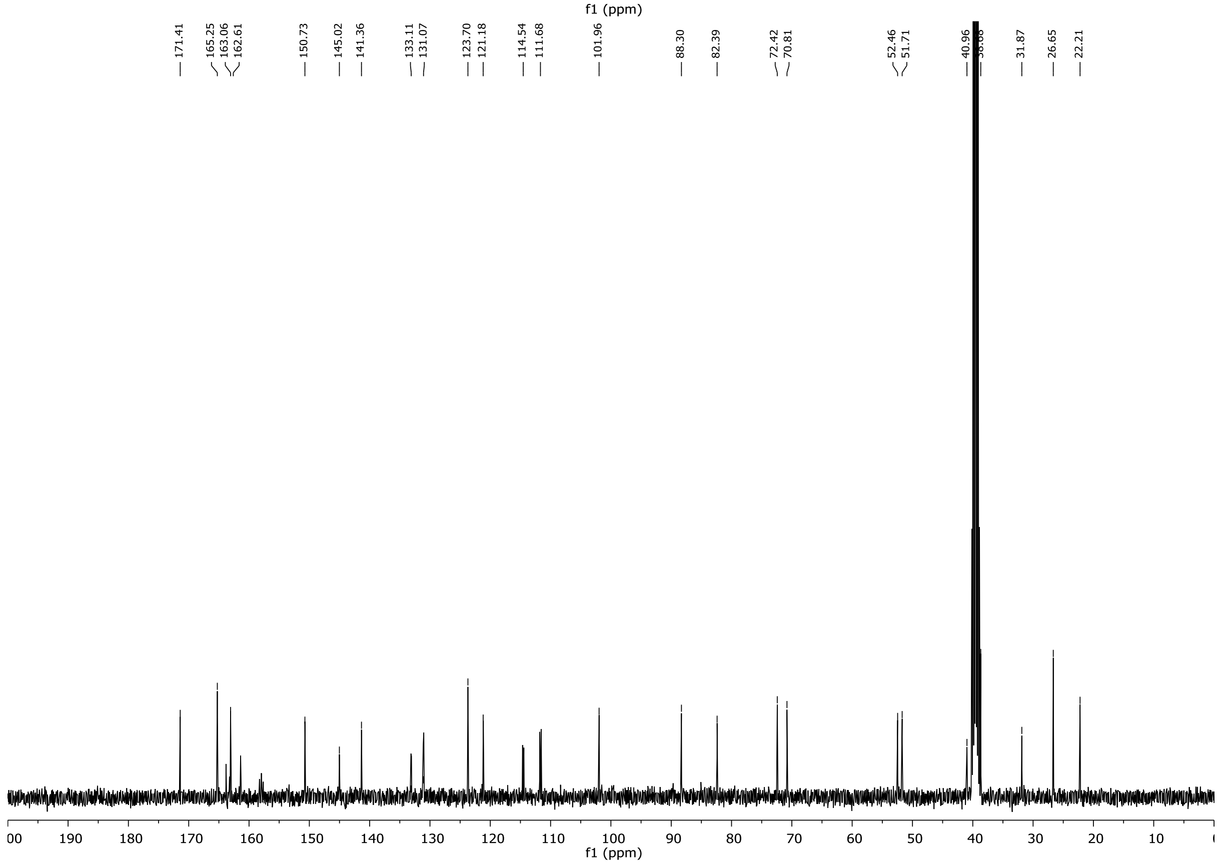

1.00
2.08
2.14
2.86
1.10
2.07
1.10
1.13
1.07
0.66
1.13
0.74
1.05
1.05
1.14
1.52
2.16
2.20
2.20
2.34
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

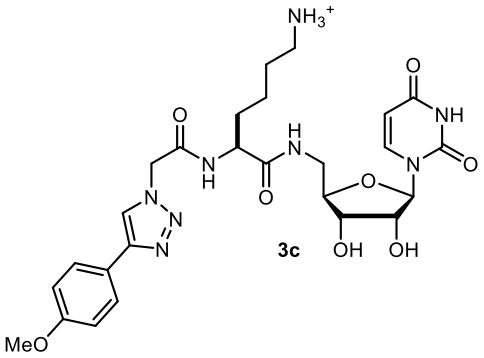

171.40
165.33
163.04
150.72
146.08
141.35
130.72
128.92
127.83
125.11
122.96
101.94
88.30
82.38
72.40
70.80
52.41
51.65
40.96
31.87
26.64
22.19
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


NH3+
O
NH
O
N
H
N
H
O
O
N
N
N
N
O
OH
OH
F

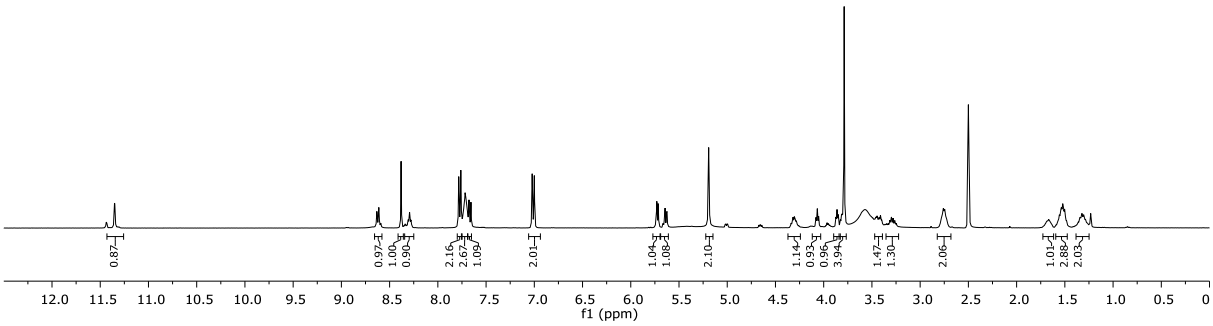
3b

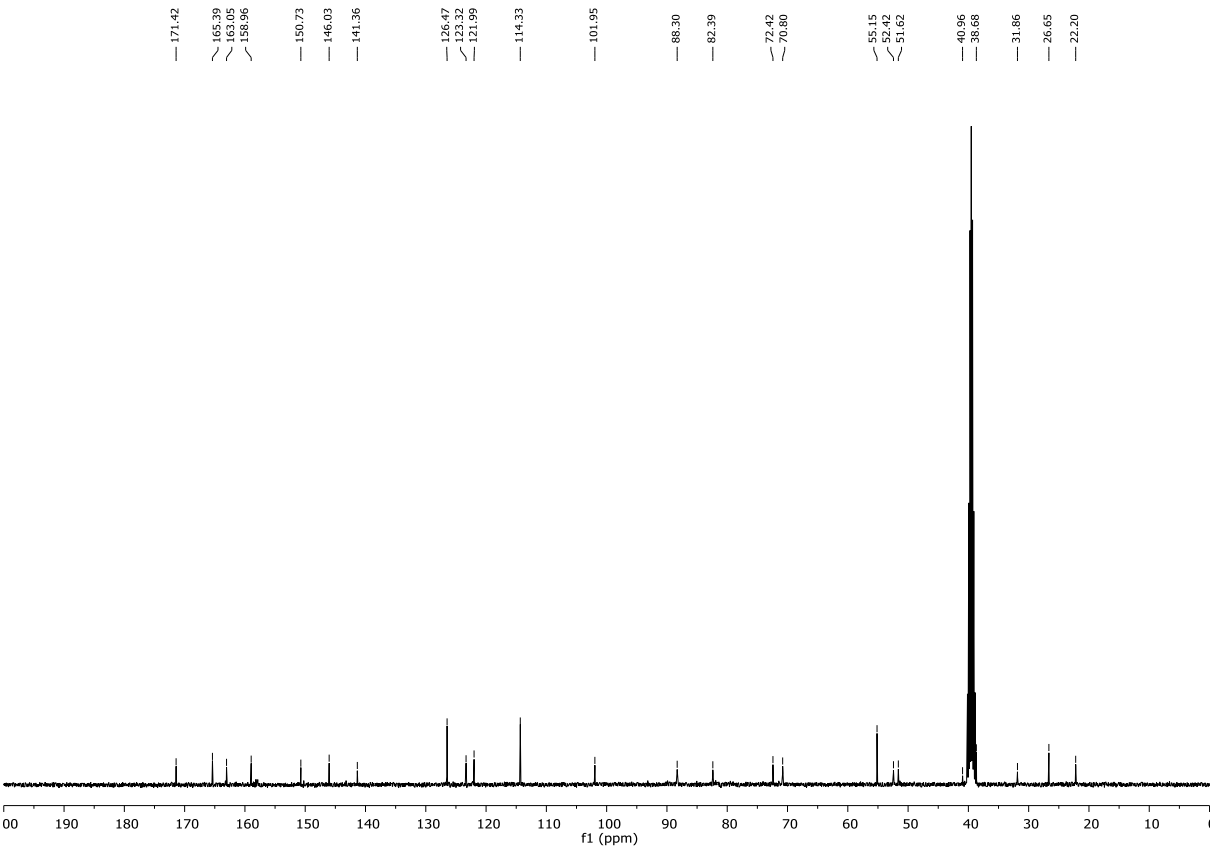

0.82
0.92
1.00
0.88
5.75
1.03
0.99
1.01
1.09
2.08
1.02
0.97
0.97
1.04
1.19
1.00
1.98
0.96
2.95
2.01
12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

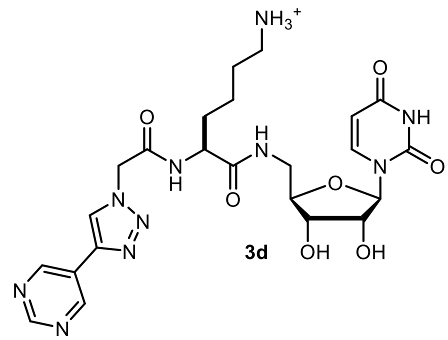

171.41
165.25
163.06
162.61
150.73
145.02
141.36
133.11
131.07
123.70
121.18
114.54
111.68
101.96
88.30
82.39
72.42
70.81
52.46
51.71
40.96
31.87
26.65
22.21
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


NH3+
O
NH
H
N
O
N
O
O
N
N
N
O
OH
OH
MeO

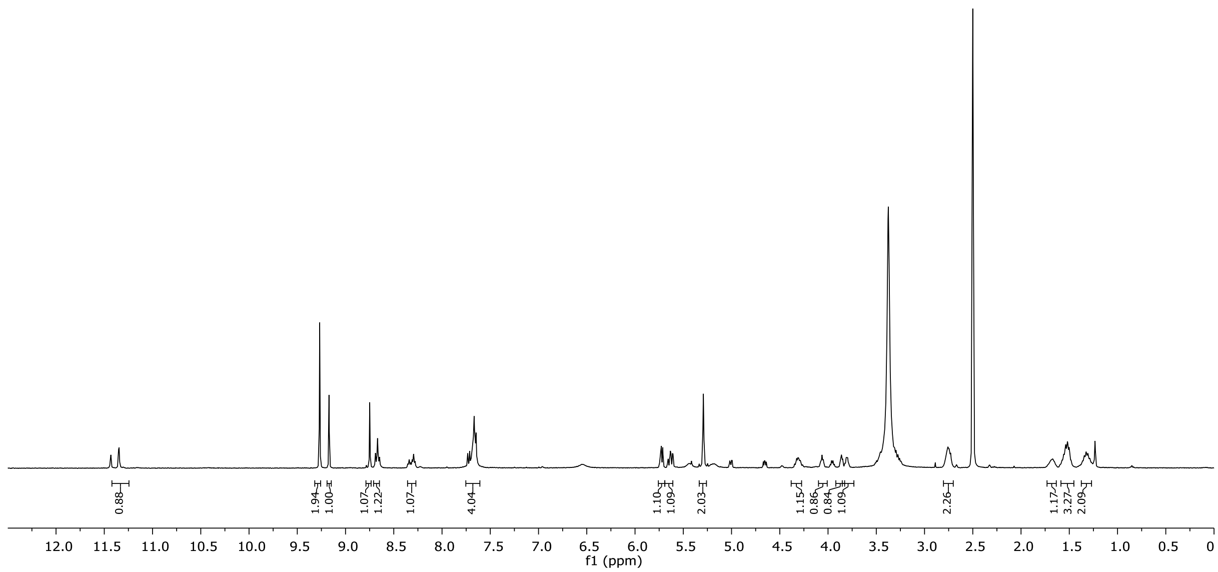
3c

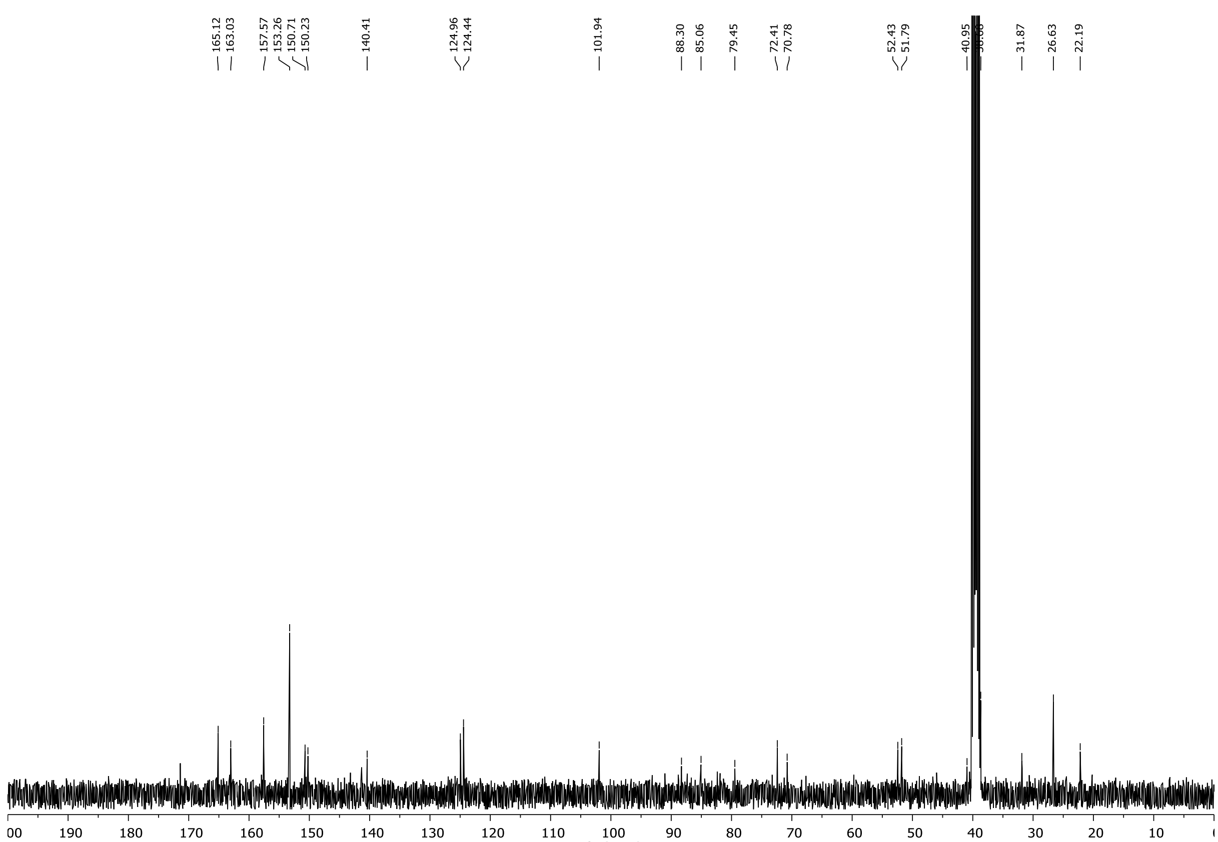

0.87
0.97
1.00
0.90
2.16
2.67
1.09
2.01
1.04
1.08
2.10
1.14
0.93
0.96
3.94
1.47
1.30
2.06
1.01
2.08
2.03
12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

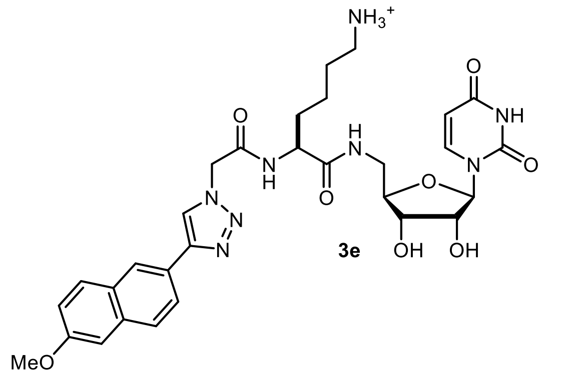

171.42
165.39
163.05
158.96
150.73
146.03
141.36
126.47
123.32
121.99
114.33
101.95
88.30
82.39
72.42
70.80
55.15
52.42
51.62
40.96
38.68
31.86
26.65
22.20
00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


NH3+
O
NH
N
H
O
H
N
O
N
O
N
N
N
OH
OH
N
N

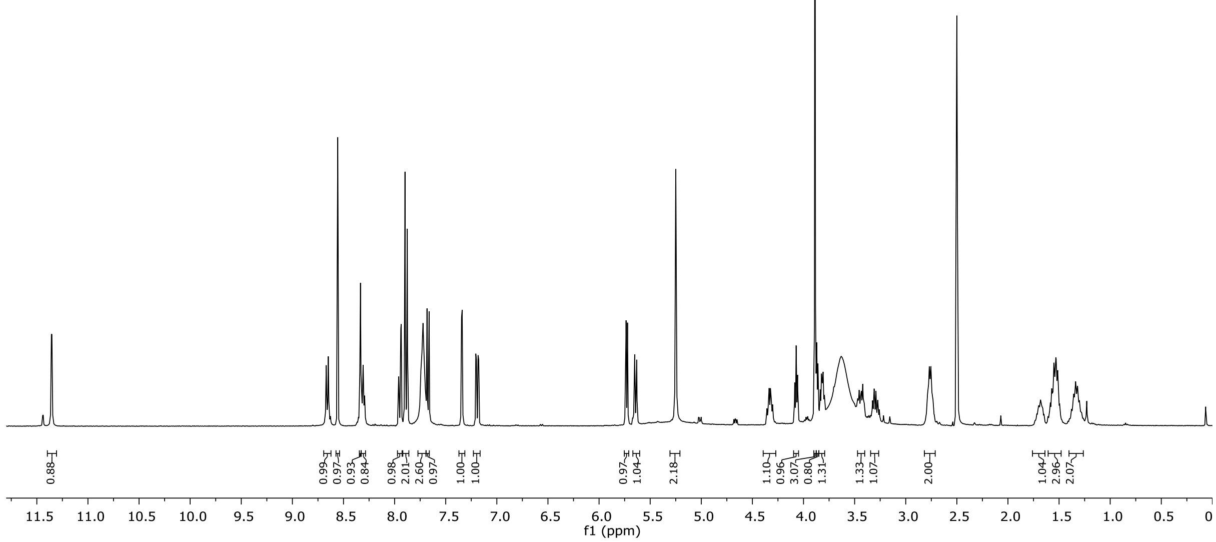
3d

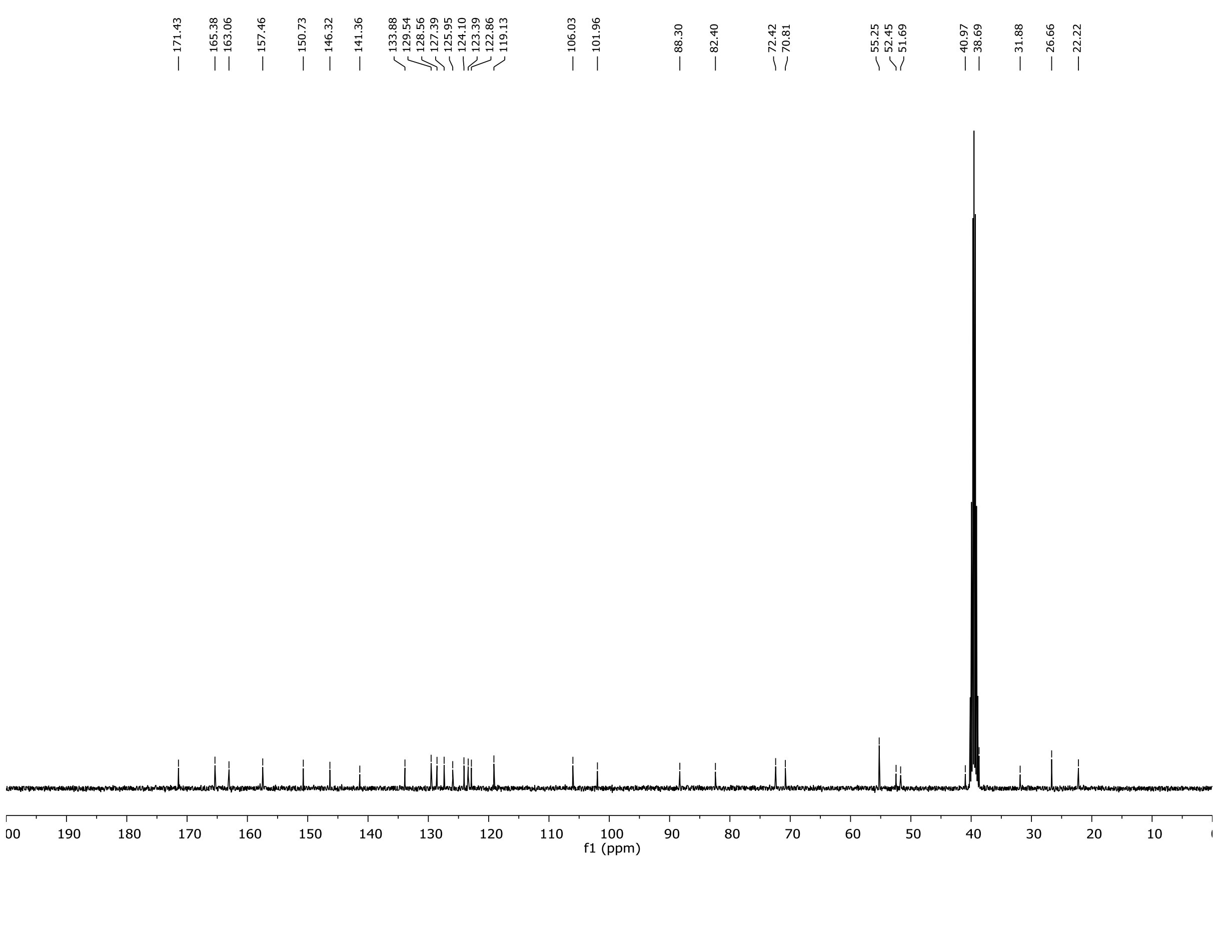

0.88
1.94
1.00
1.07
1.22
1.07
4.04
1.10
1.09
2.03
1.15
0.86
0.84
1.09
2.26
1.17
3.27
2.09
12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

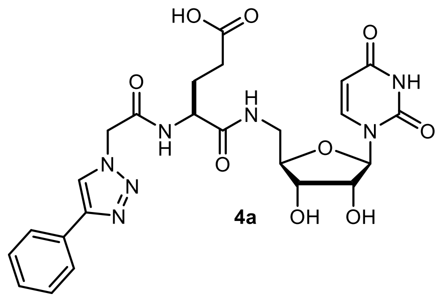

165.12
163.03
157.57
153.26
150.71
150.23
140.41
124.96
124.44
101.94
88.30
85.06
79.45
72.41
70.78
52.43
51.79
40.95
31.87
26.63
22.19
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


NH3+
O
NH
O
H
N
O
N
H
N
O
O
N
N
N
N
MeO
OH
OH

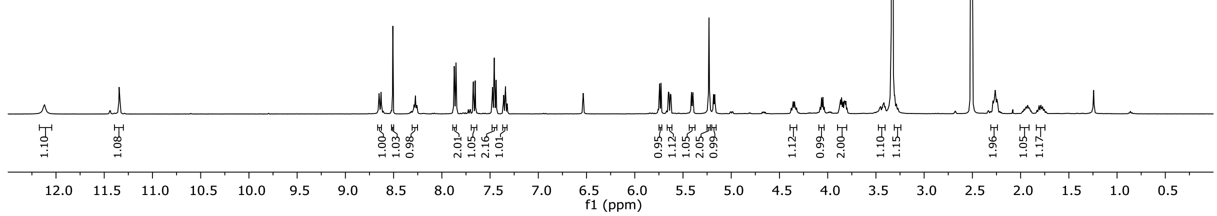
3e

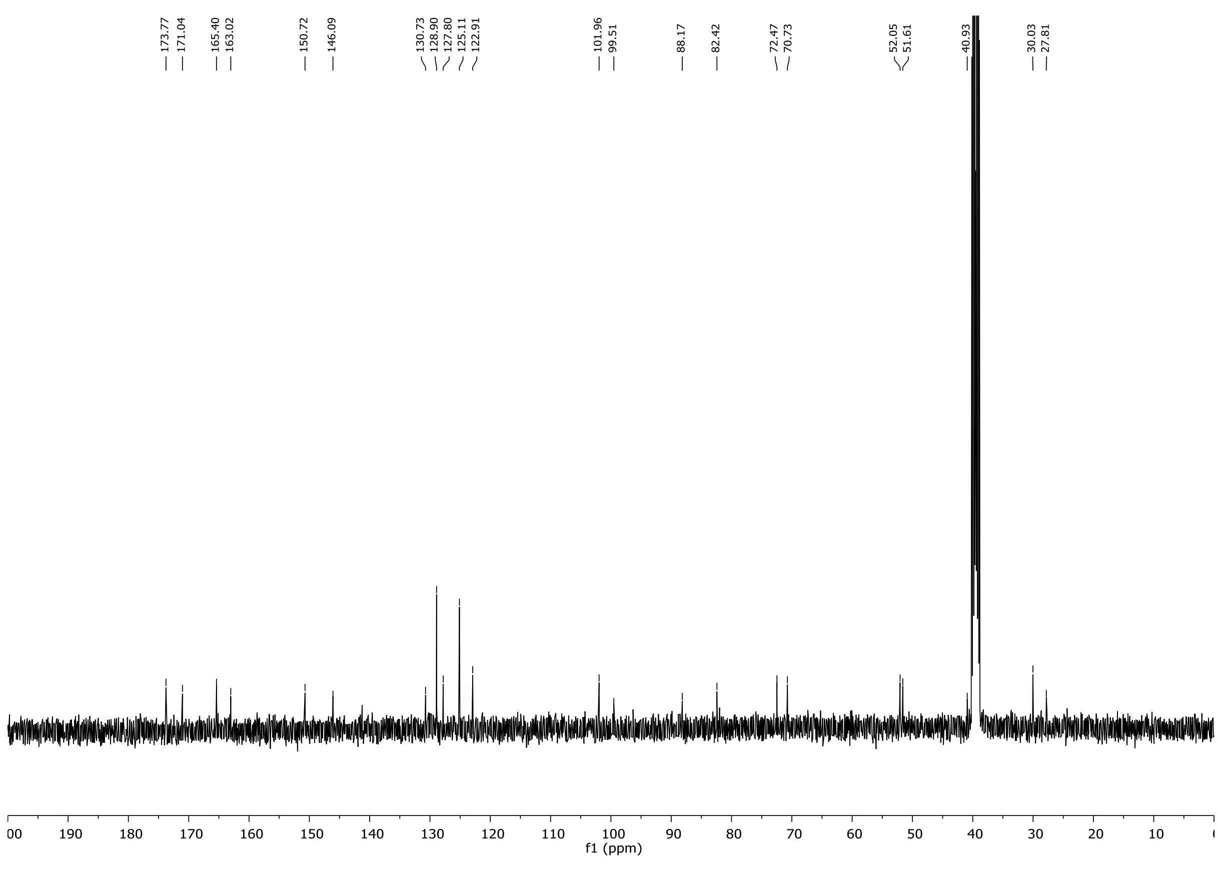

0.88
0.99
0.97
0.93
0.84
0.98
2.01
2.60
0.97
1.00
1.00
0.97
1.04
2.18
1.10
0.96
3.07
0.80
1.31
1.33
1.07
2.00
1.04
2.96
2.07
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

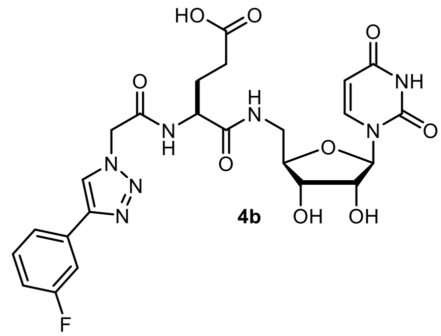

171.43
165.38
163.06
157.46
150.73
146.32
141.36
133.88
129.54
128.56
127.39
125.95
124.10
123.39
122.86
119.13
106.03
101.96
88.30
82.40
72.42
70.81
55.25
52.45
51.69
40.97
38.69
31.88
26.66
22.22
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


HO
O
O
NH
O
N
H
H
N
O
O
N
N
N
N
O
OH
OH

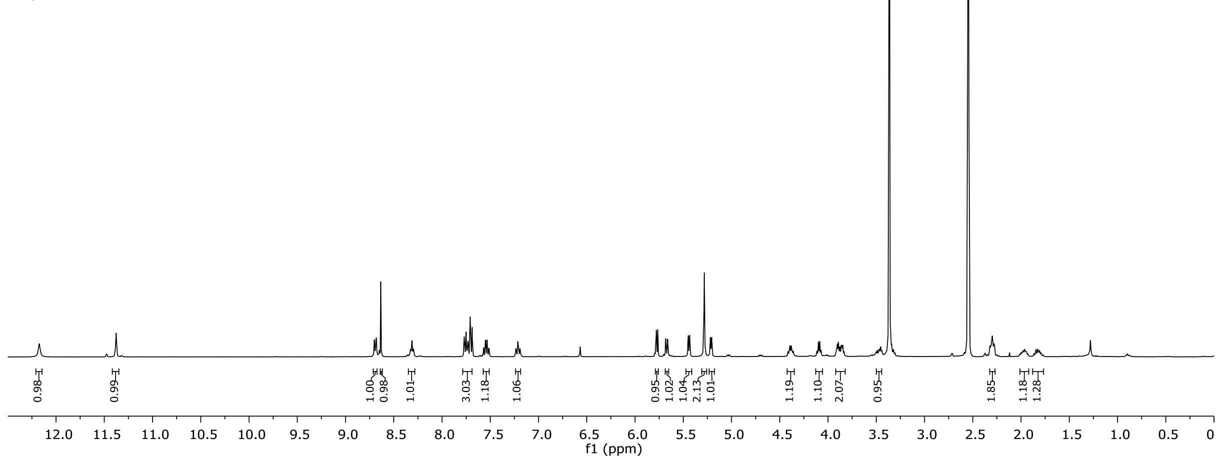
4a

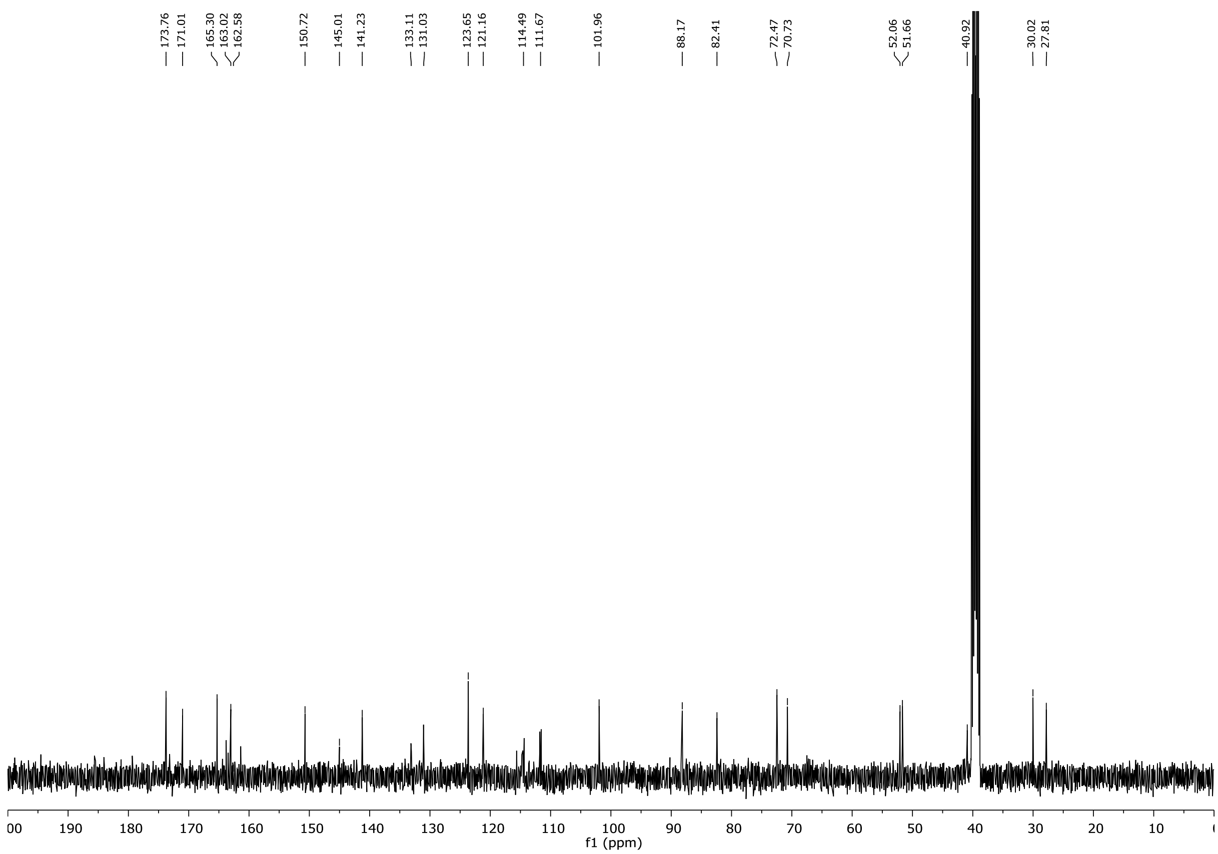

1.10
1.08
1.00
1.03
0.98
2.01
1.05
2.16
1.01
0.95
1.12
1.05
2.05
0.99
1.12
0.99
2.00
1.10
1.15
1.96
1.05
1.17
12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5
f1 (ppm)

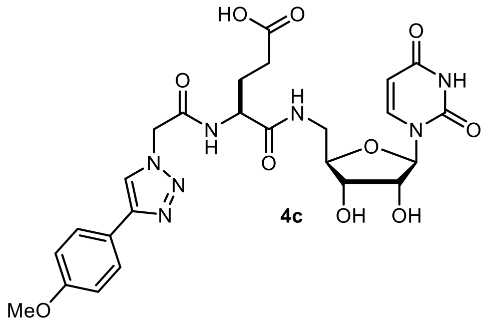

173.77
171.04
165.40
163.02
150.72
146.09
130.73
128.90
127.80
125.11
122.91
101.96
99.51
88.17
82.42
72.47
70.73
52.05
51.61
40.93
30.03
27.81
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)



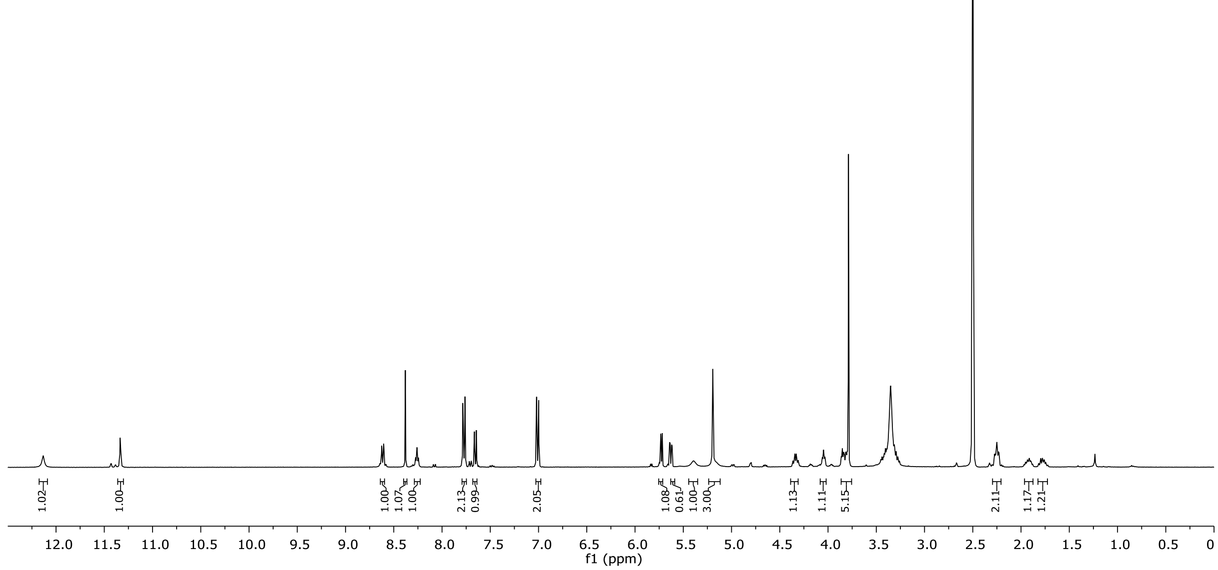
4b

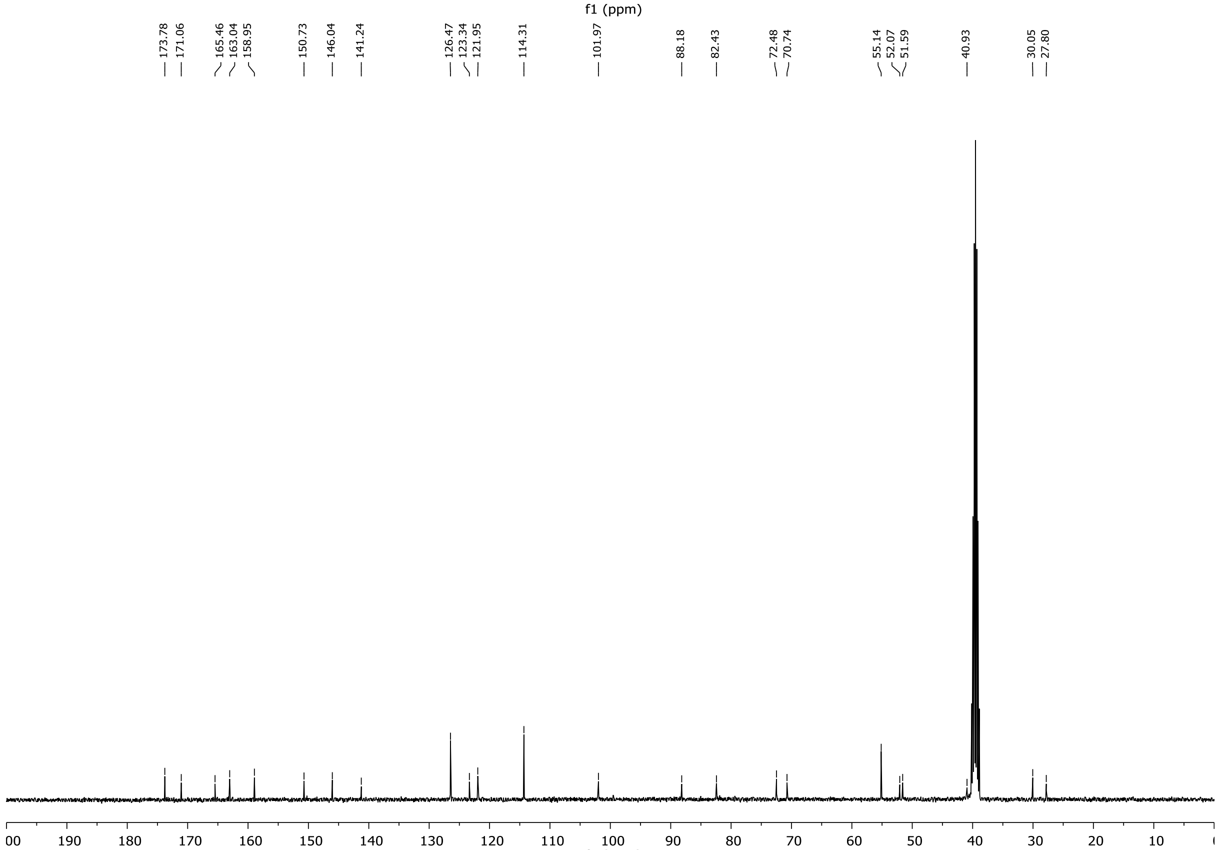

0.98
0.99
1.00
0.98
1.01
3.03
1.18
1.06
0.95
1.02
1.04
2.13
1.01
1.19
1.10
2.07
0.95
1.85
1.18
1.28
12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

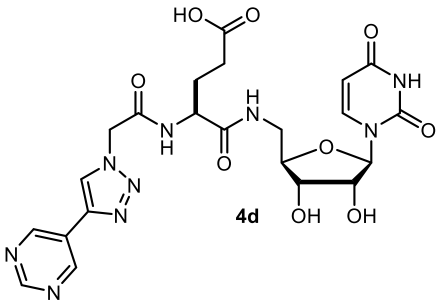

173.76
171.01
165.30
163.02
162.58
150.72
145.01
141.23
133.11
131.03
123.65
121.16
114.49
111.67
101.96
88.17
82.41
72.47
70.73
52.06
51.66
40.92
30.02
27.81
00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


HO
O
O
NH
O
O
N
H
N
H
N
O
O
N
N
N
N
OH
OH
MeO

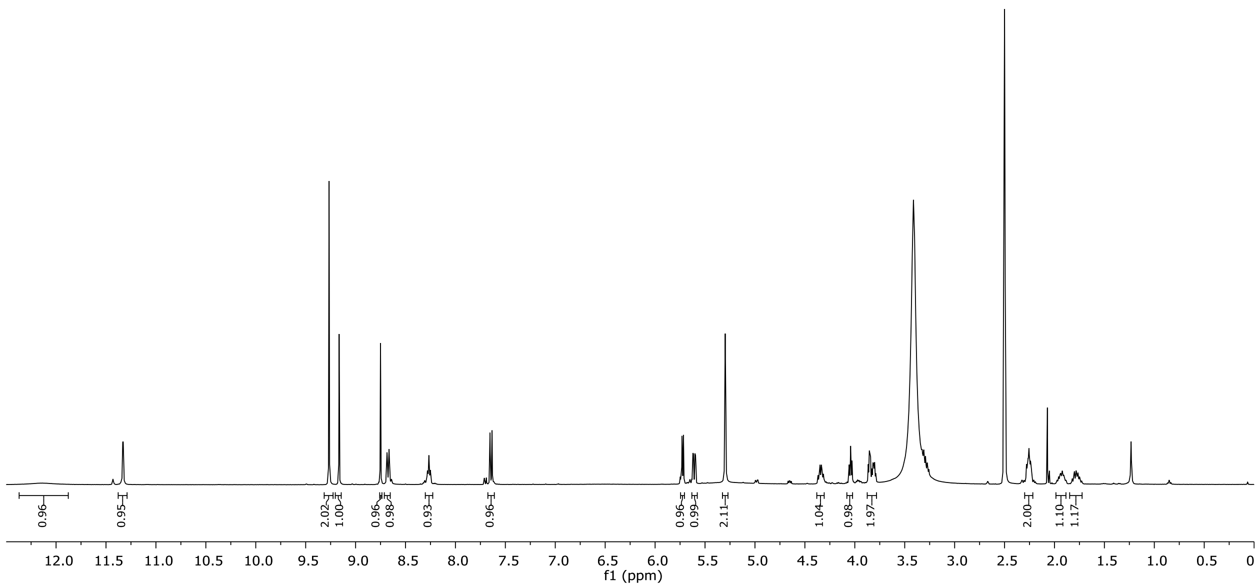
4c

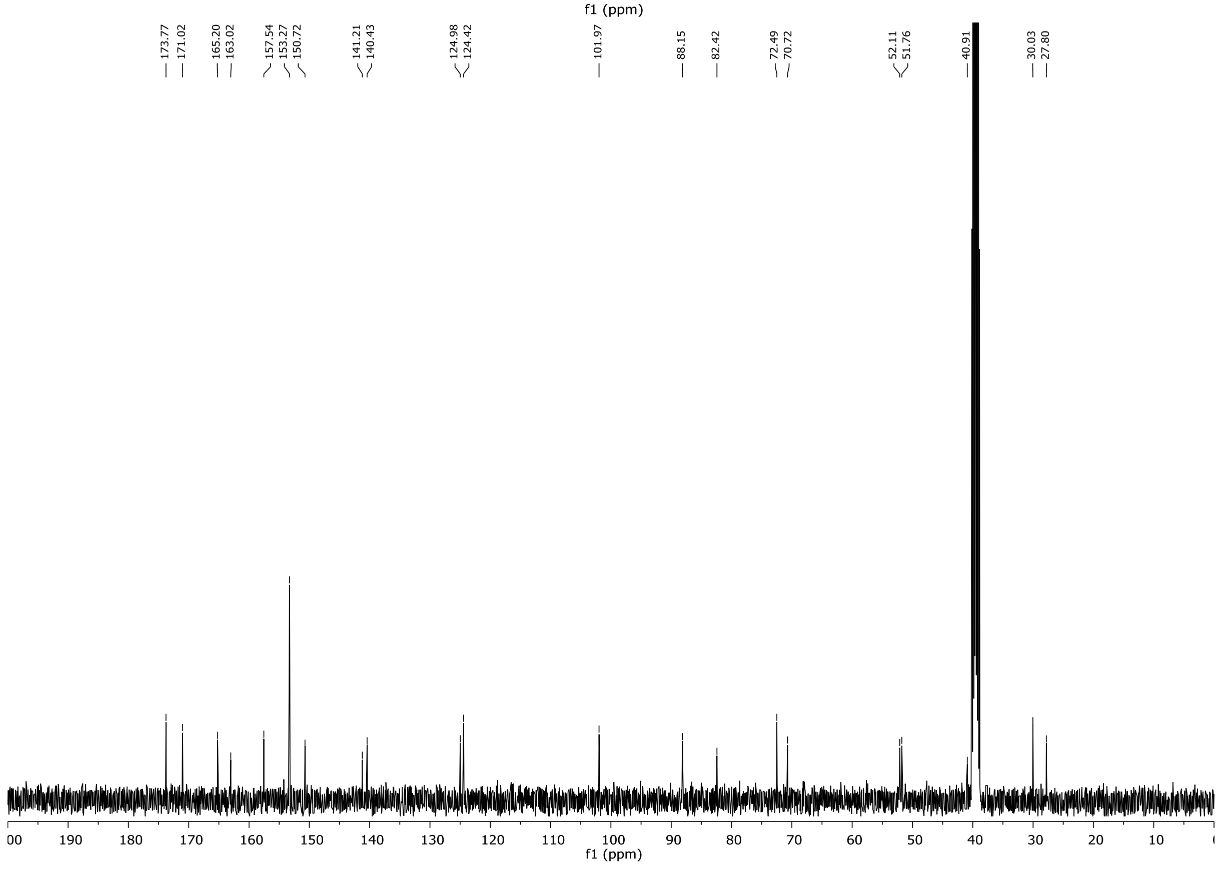

1.02
1.00
1.00
1.07
1.00
2.13
0.99
2.05
1.08
0.61
1.00
3.00
1.13
1.11
5.15
2.11
1.17
1.21
12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

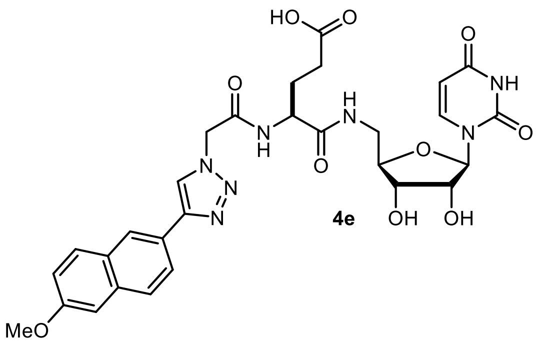

173.78
171.06
165.46
163.04
158.95
150.73
146.04
141.24
126.47
123.34
121.95
114.31
101.97
88.18
82.43
72.48
70.74
55.14
52.07
51.59
40.93
30.05
27.80
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


HO
O
O
NH
O
N
H
N
H
O
H
N
O
N
O
N
N
N
OH
OH
N
N

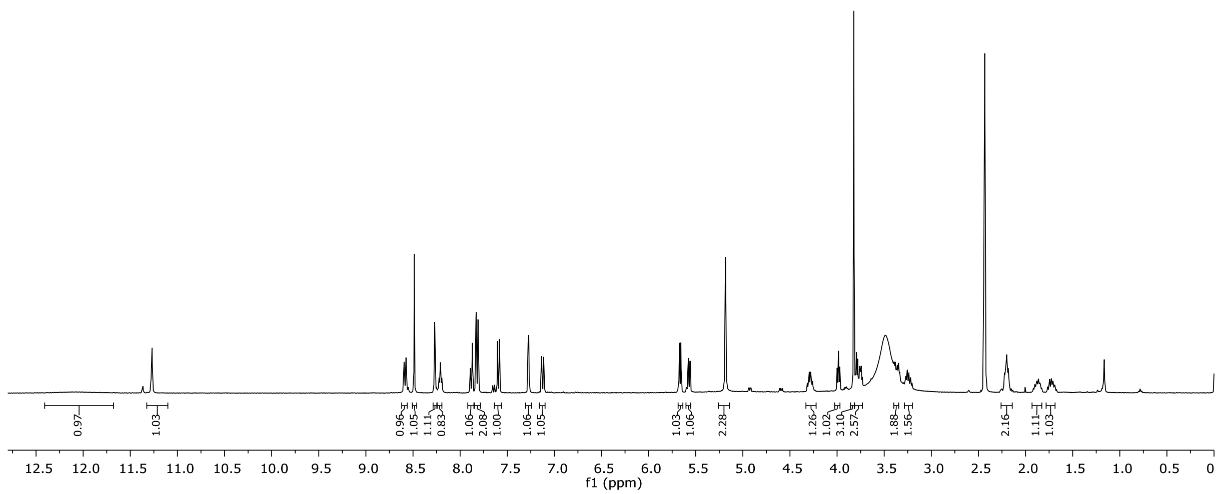
4d

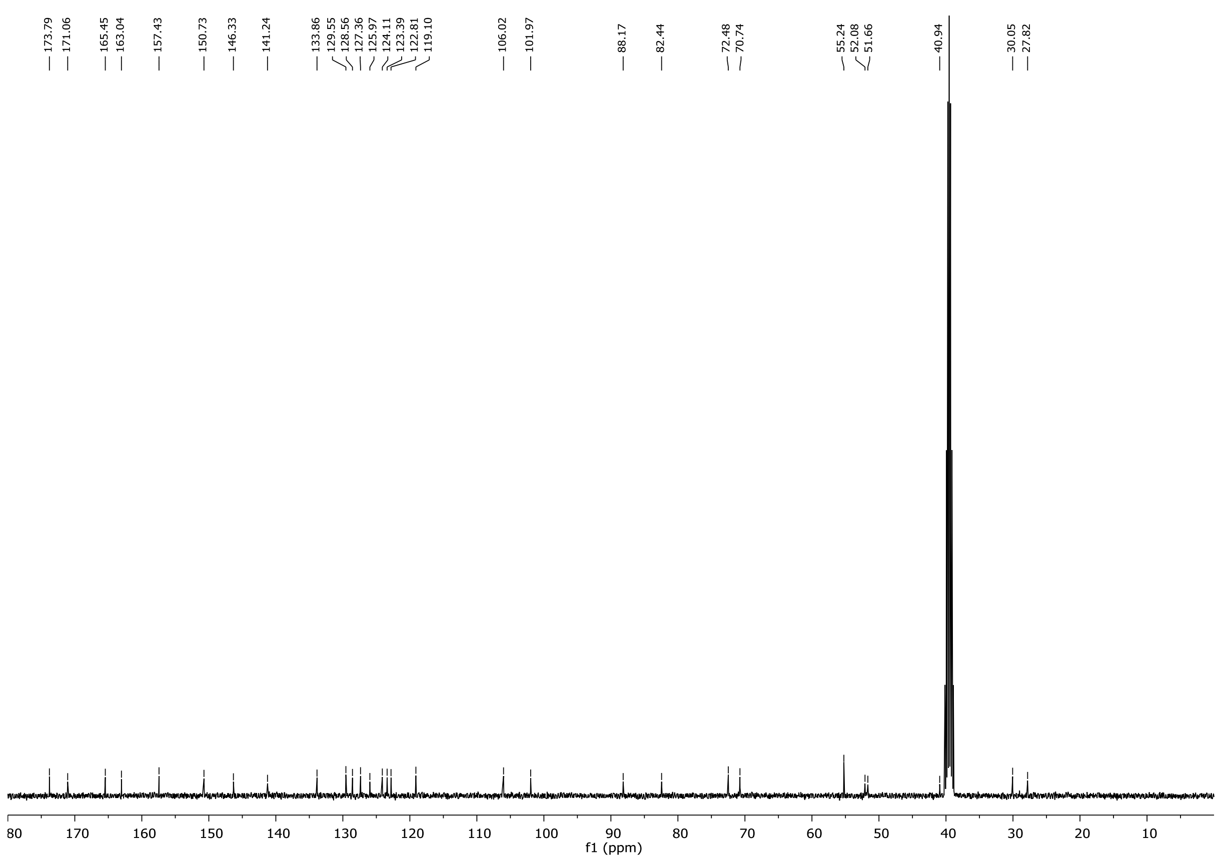

0.96
0.95
2.02
1.00
0.96
0.98
0.93
0.96
0.96
0.99
2.11
1.04
0.98
1.97
2.00
1.10
1.17
f1 (ppm)

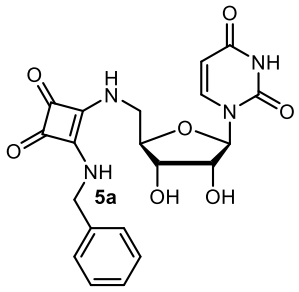

173.77
171.02
165.20
163.02
157.54
153.27
150.72
141.21
140.43
124.98
124.42
101.97
88.15
82.42
72.49
70.72
52.11
51.76
40.91
30.03
27.80
f1 (ppm)

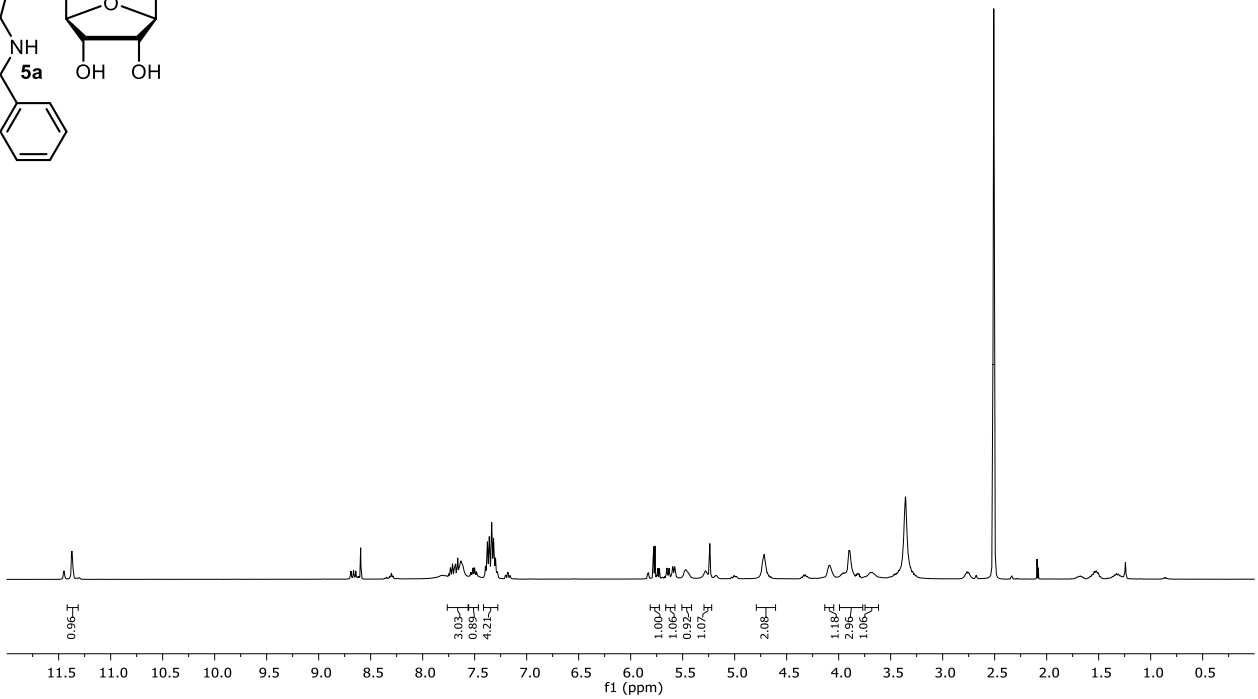

HO
O
O
NH
N
O
O
H
N
N
H
O
N
N
N
N
O
OH
OH
MeO

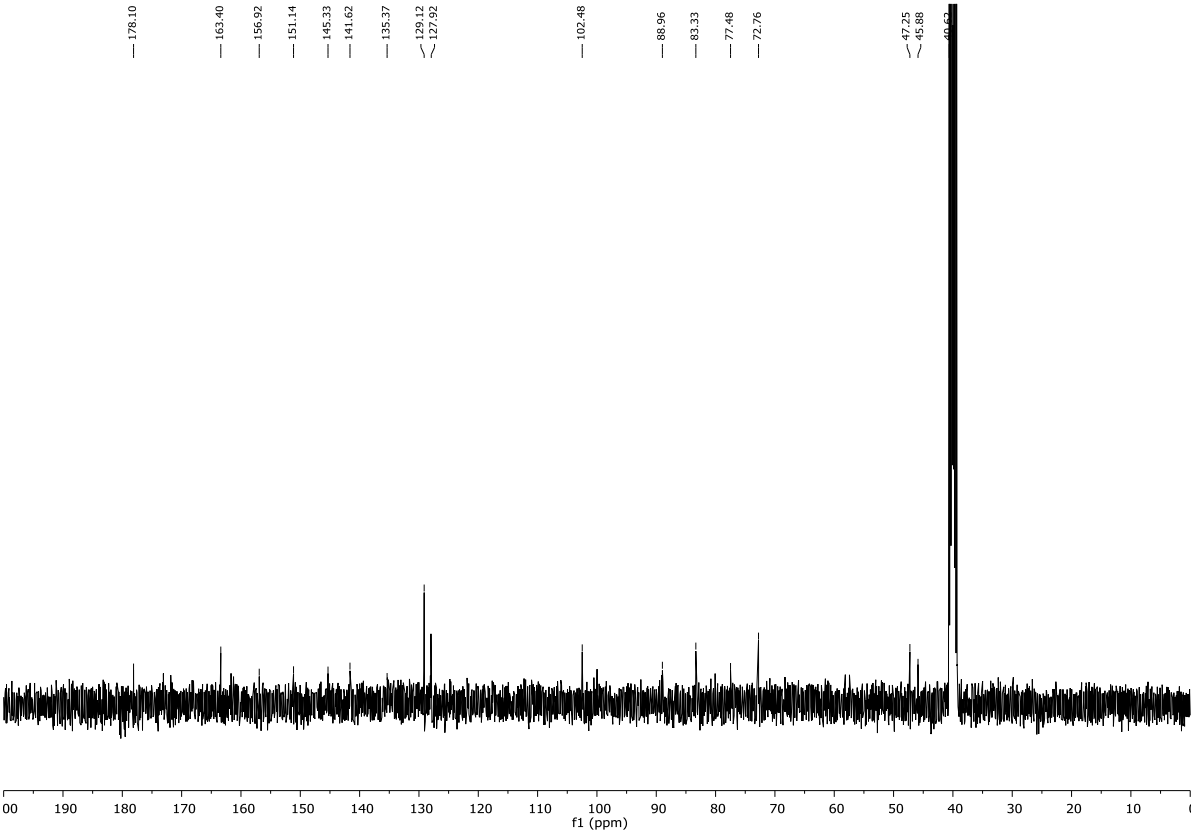
4e

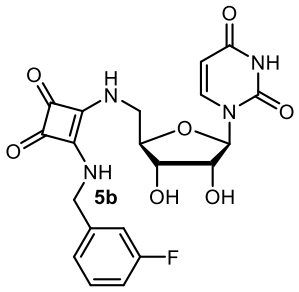

f1 (ppm)


173.79
171.06
165.45
163.04
157.43
150.73
146.33
141.24
133.86
129.55
128.56
127.36
125.97
124.11
123.39
122.81
119.10
106.02
101.97
88.17
82.44
72.48
70.74
55.24
52.08
51.66
40.94
30.05
27.82
f1 (ppm)

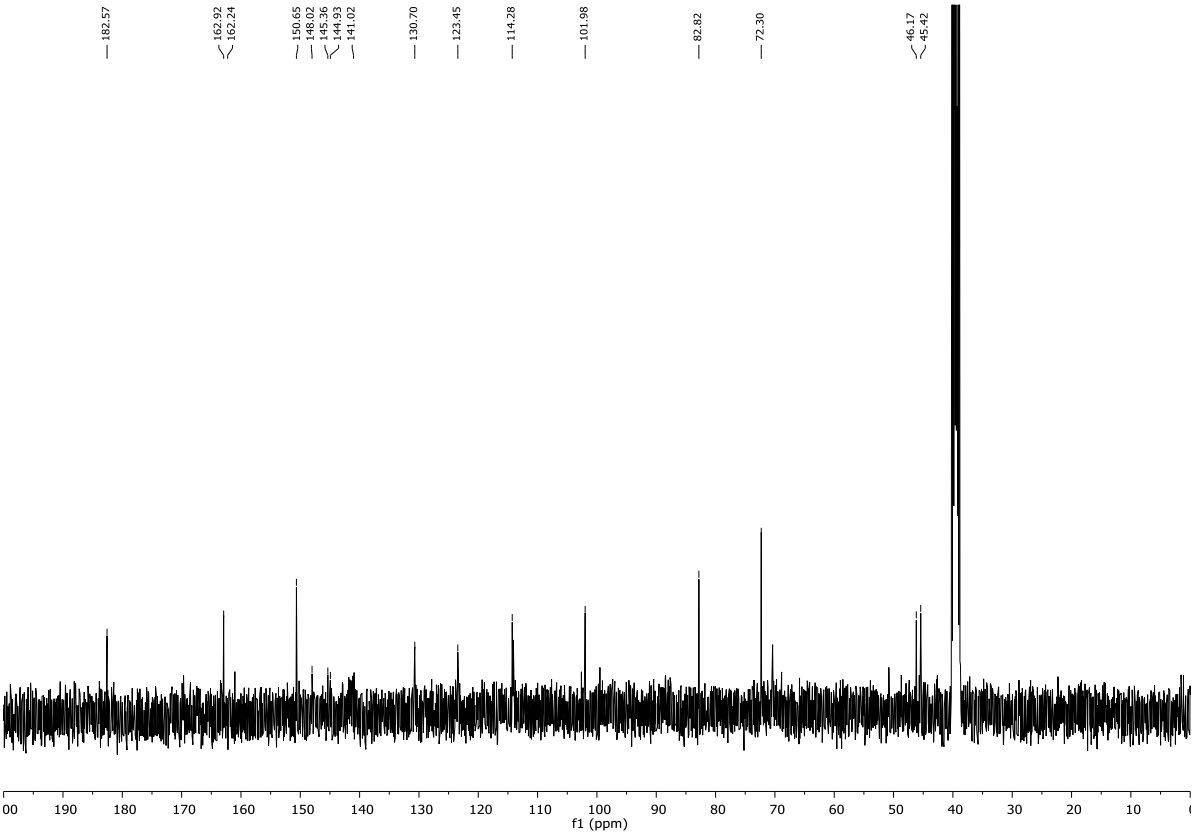

O
NH
H
N
O
O
O
N
O
NH
5a
OH
OH

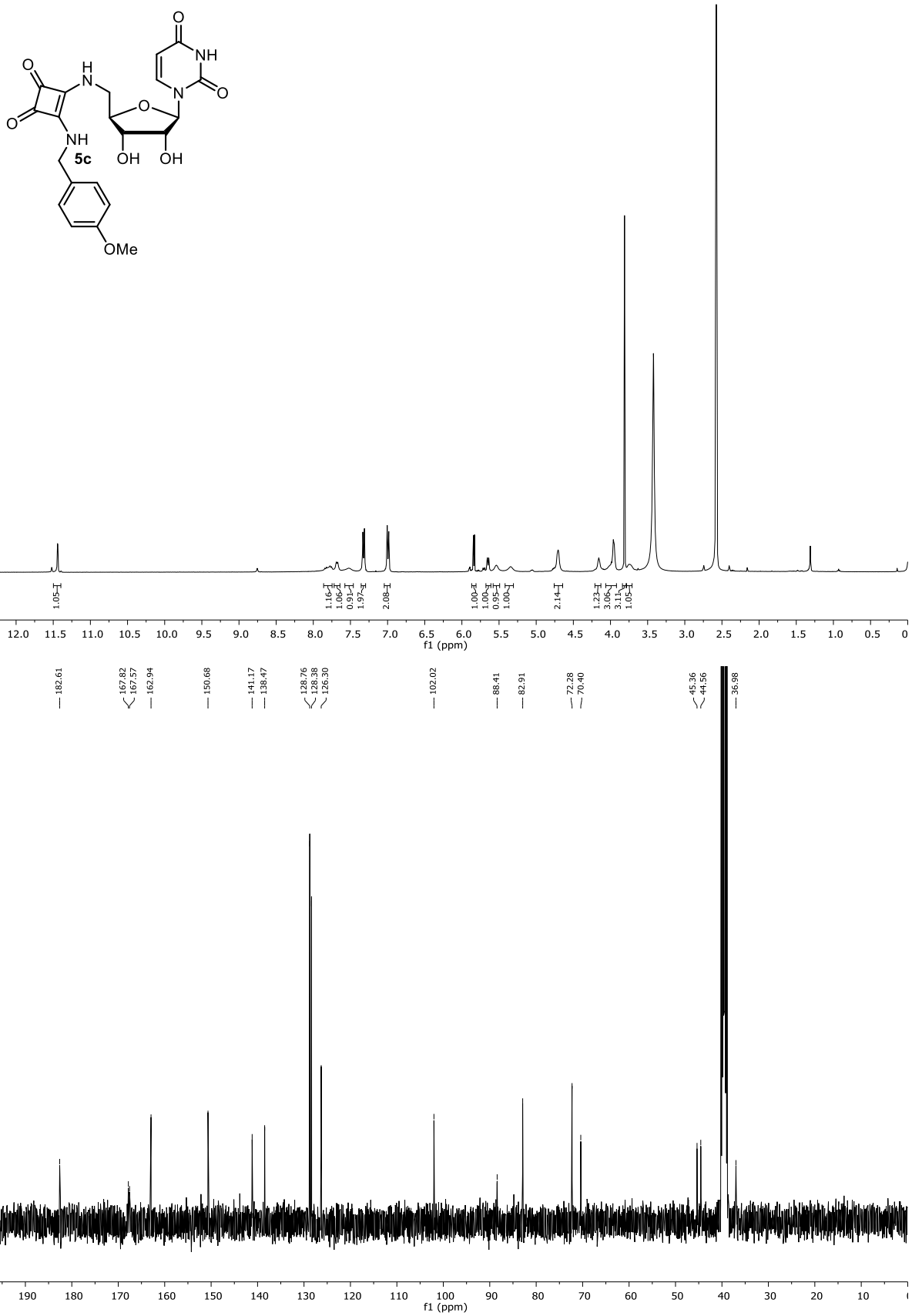

0.96
3.03
0.89
4.21
1.00
1.06
0.92
1.07
2.08
1.18
2.96
1.06
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5
f1 (ppm)

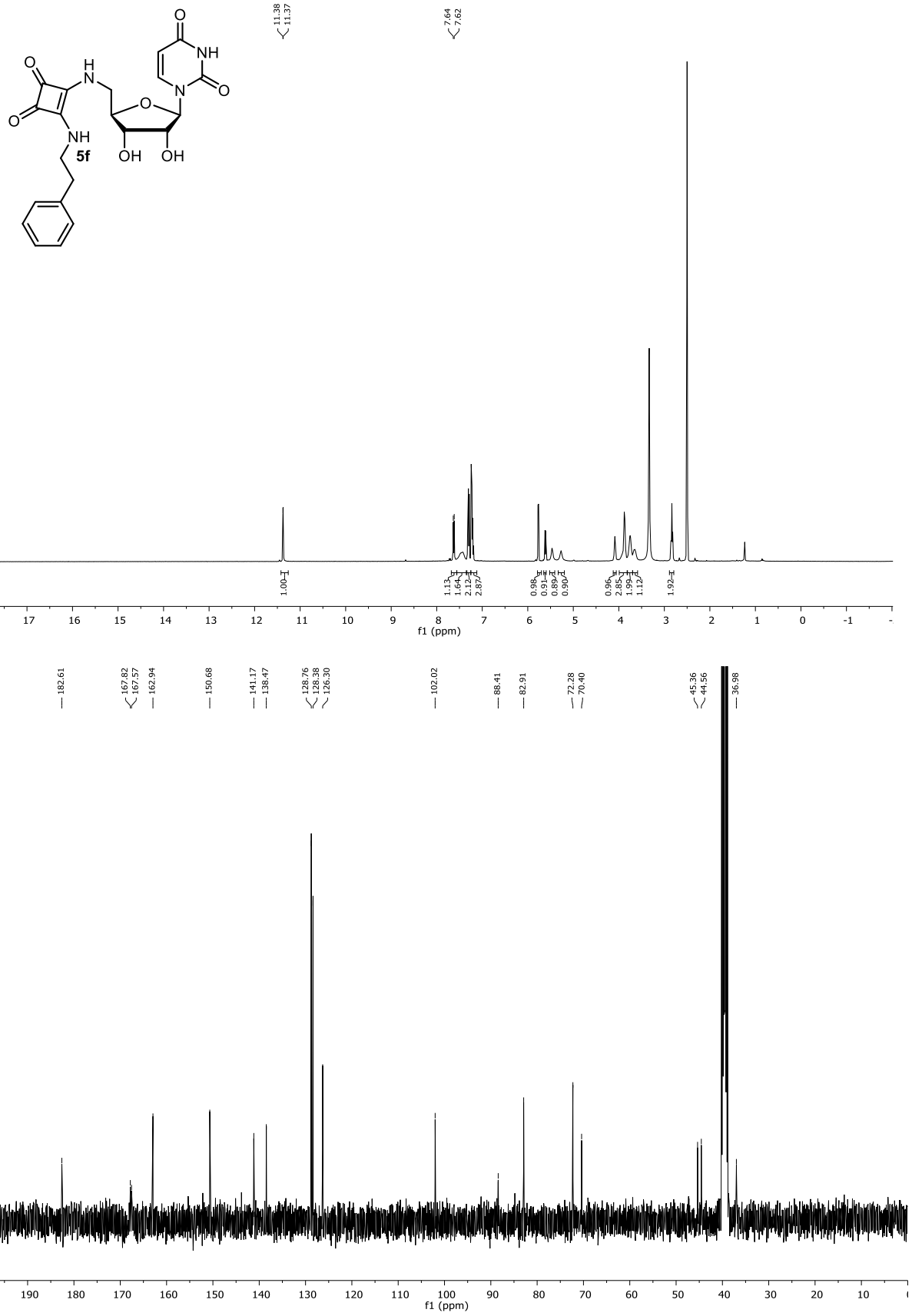

178.10
163.40
156.92
151.14
145.33
141.62
135.37
129.12
127.92
102.48
88.96
83.33
77.48
72.76
47.25
45.88
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

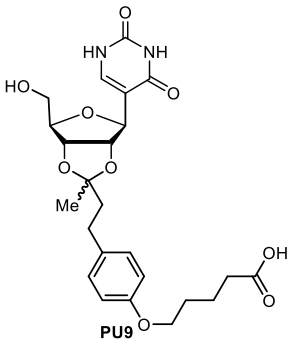

O
NH
O
H
N
O
N
O
O
NH
5b
OH
OH
F

0.95
0.93
1.01
0.70
1.15
3.05
1.00
1.06
2.14
0.97
3.06
1.37
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)


182.57
162.92
162.24
150.65
148.02
145.36
144.93
141.02
130.70
123.45
114.28
101.98
82.82
72.30
46.17
45.42
00 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

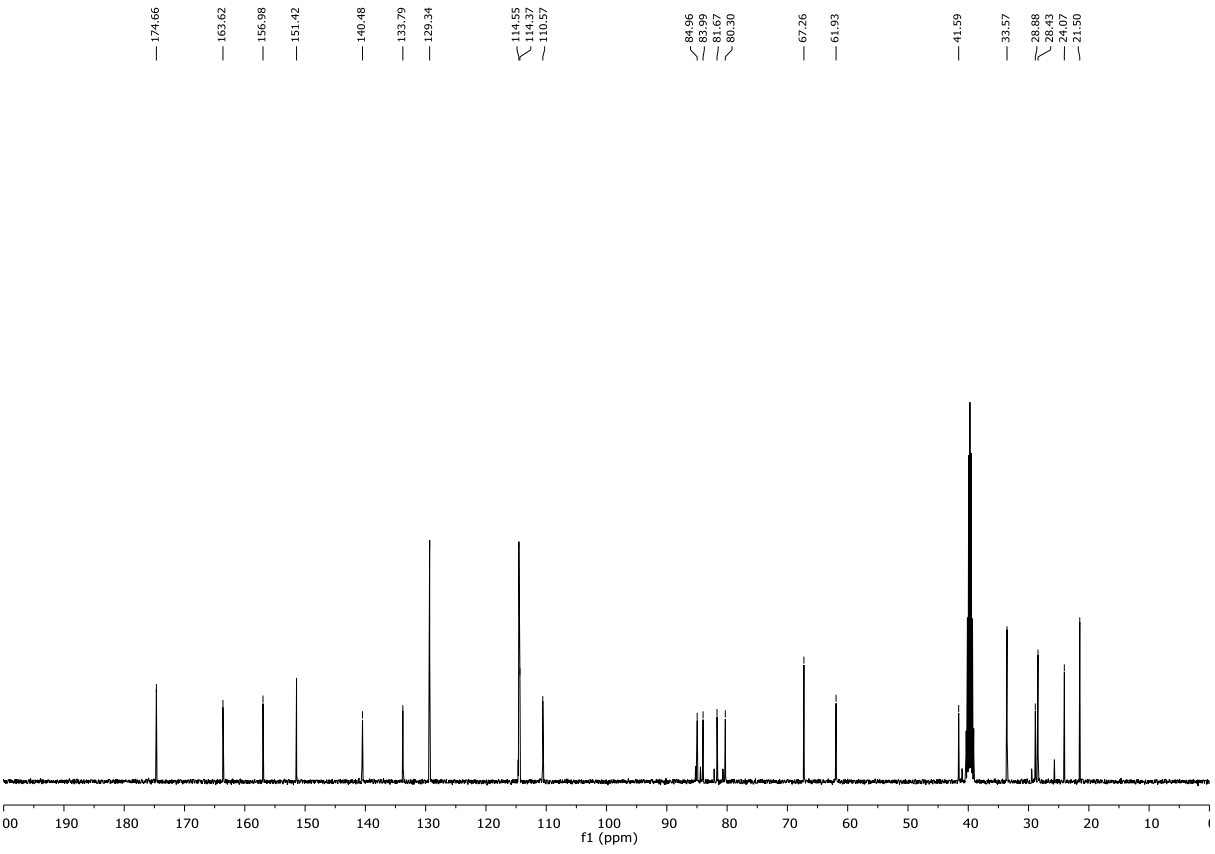

5c
OMe
f1 (ppm)
f1 (ppm)

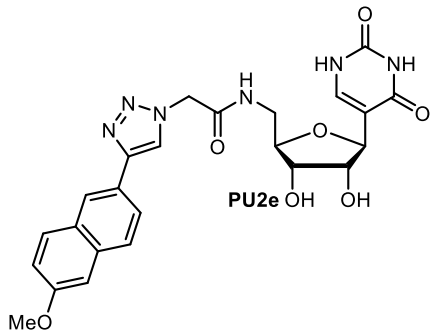

5f
11.38
11.37
7.64
7.62
1.00
1.13
1.64
2.12
2.87
0.98
0.91
0.89
0.90
0.96
2.85
1.93
1.12
1.92
f1 (ppm)
182.61
167.82
167.57
162.94
150.68
141.17
138.47
128.76
128.38
126.30
102.02
88.41
82.91
72.28
70.40
45.36
44.56
36.98
f1 (ppm)

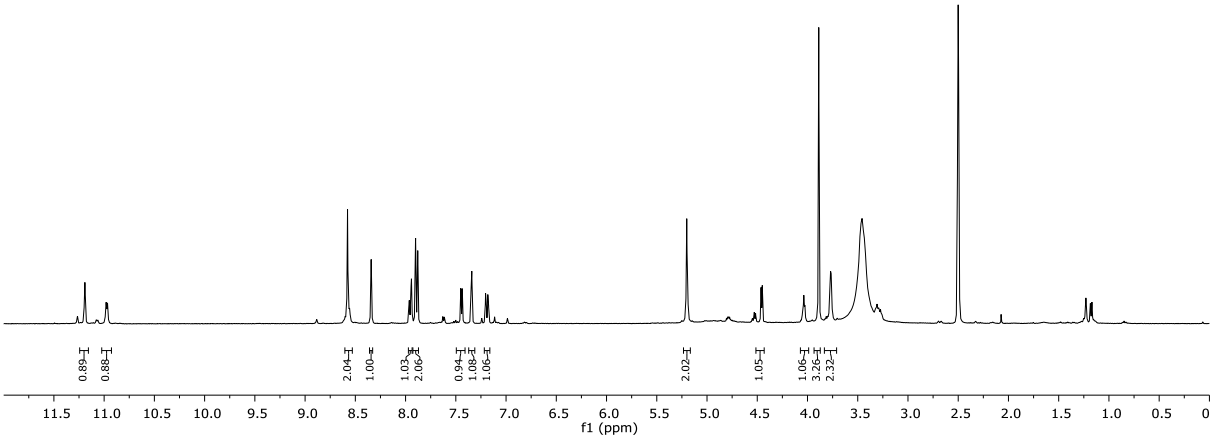

O
HN
NH
O
HO
O
O
O
Me
OH
O
O

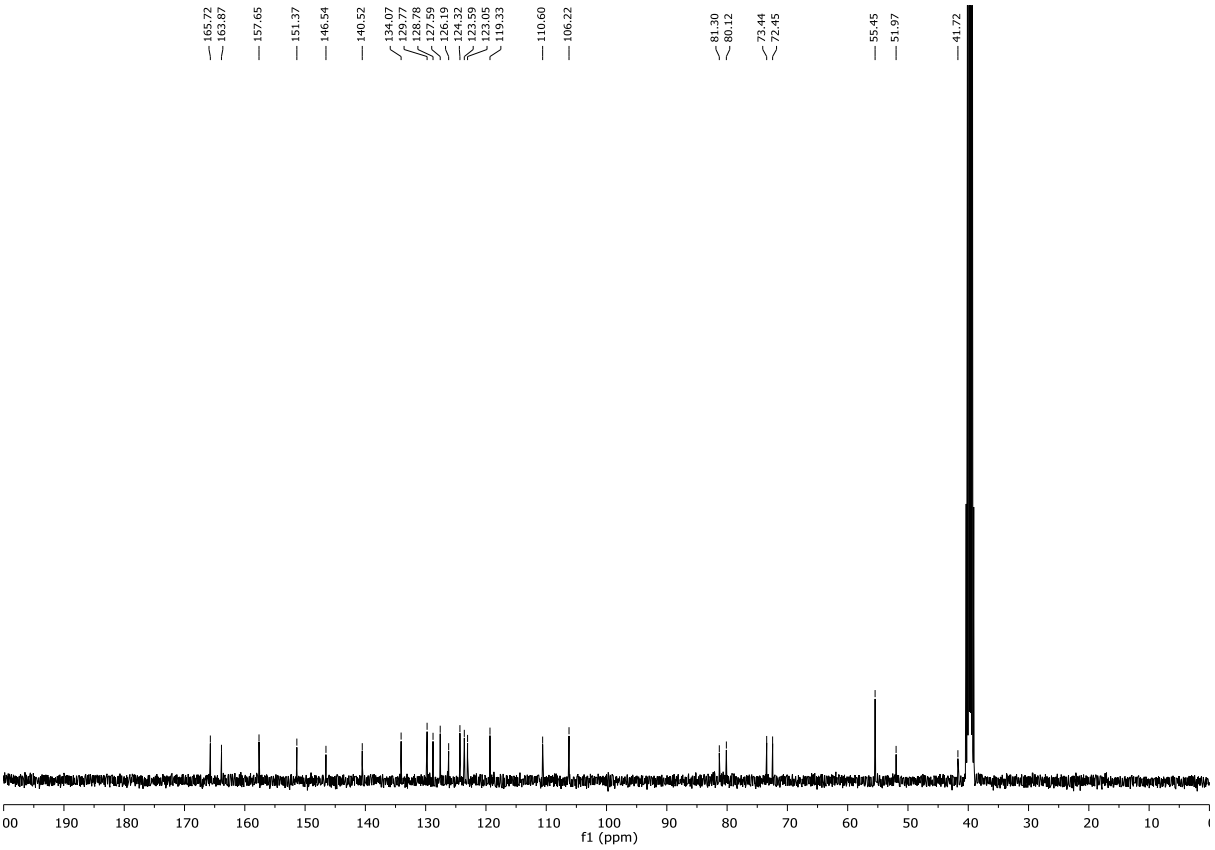
PU9

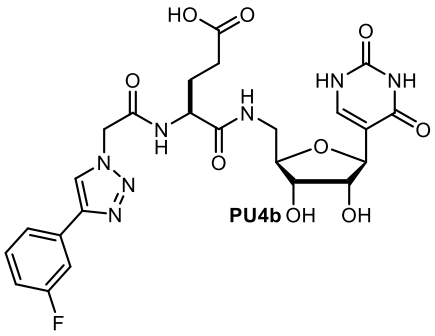
0.66
1.12
0.86
1.00
2.06
2.08
3.31
3.12
2.20
1.69
0.42
2.09
1.71
0.36
4.19
0.51
2.62
f1 (ppm)


174.66
163.62
156.98
151.42
140.48
133.79
129.34
114.55
114.37
110.57
84.96
83.99
81.67
80.30
67.26
61.93
41.59
33.57
28.88
28.43
24.07
21.50
f1 (ppm)

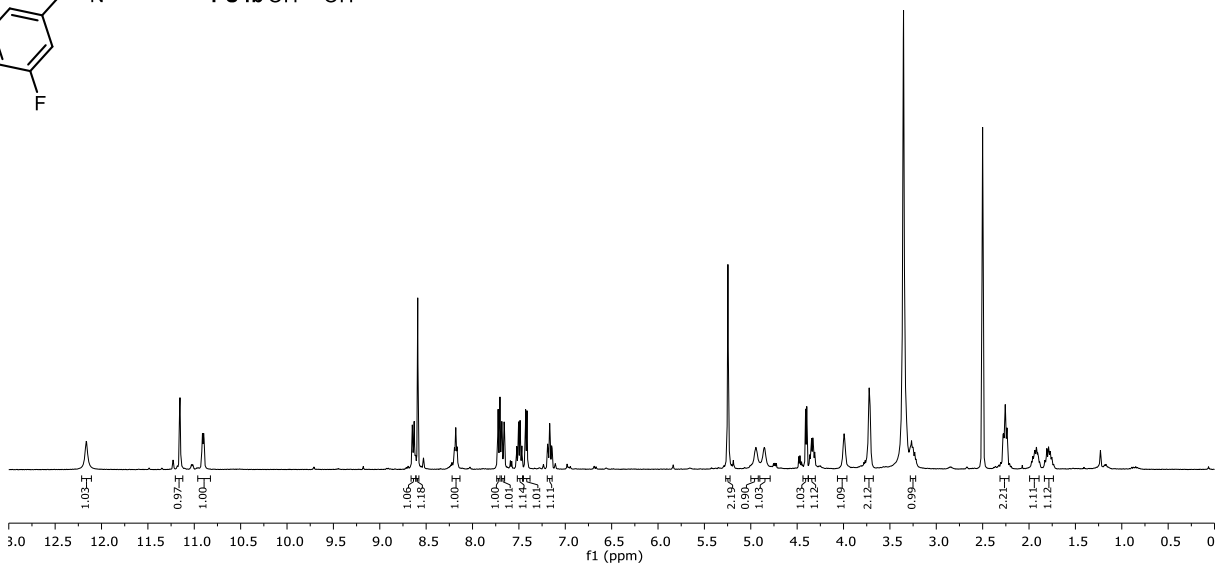

O
HN
NH
O
H
N
N
N
N
O
O
PU2e
OH
OH
MeO

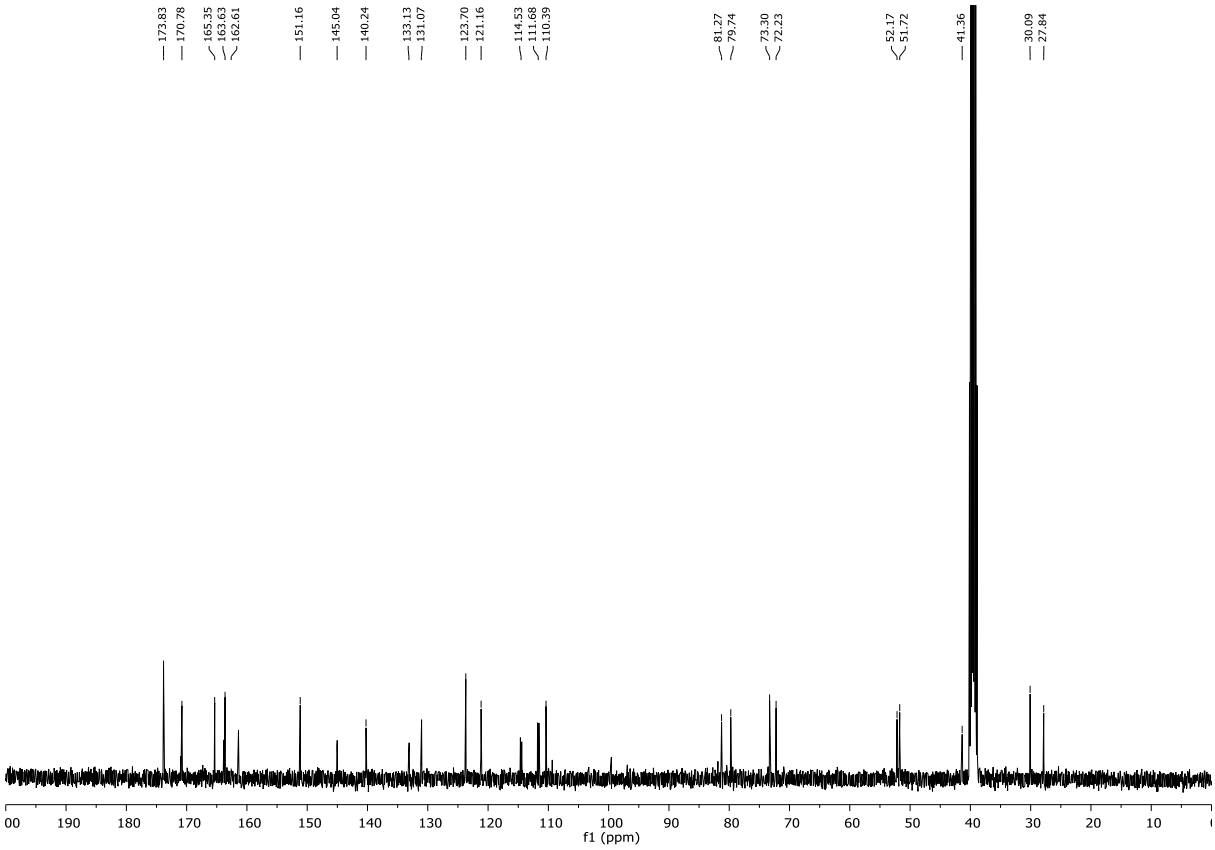

0.89
0.88
2.04
1.00
1.03
2.06
0.94
1.08
1.06
2.02
1.05
1.06
3.26
2.32
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
f1 (ppm)

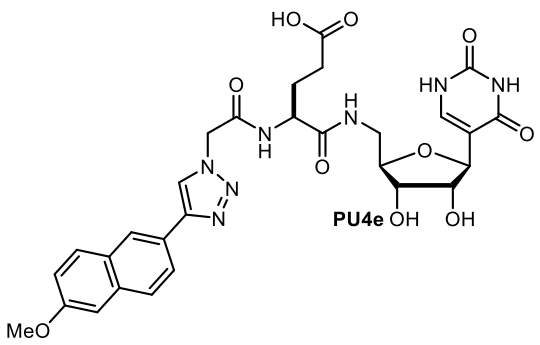

165.72
163.87
157.65
151.37
146.54
140.52
134.07
129.77
128.78
127.59
126.19
124.32
123.59
123.05
119.33
110.60
106.22
81.30
80.12
73.44
72.45
55.45
51.97
41.72
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

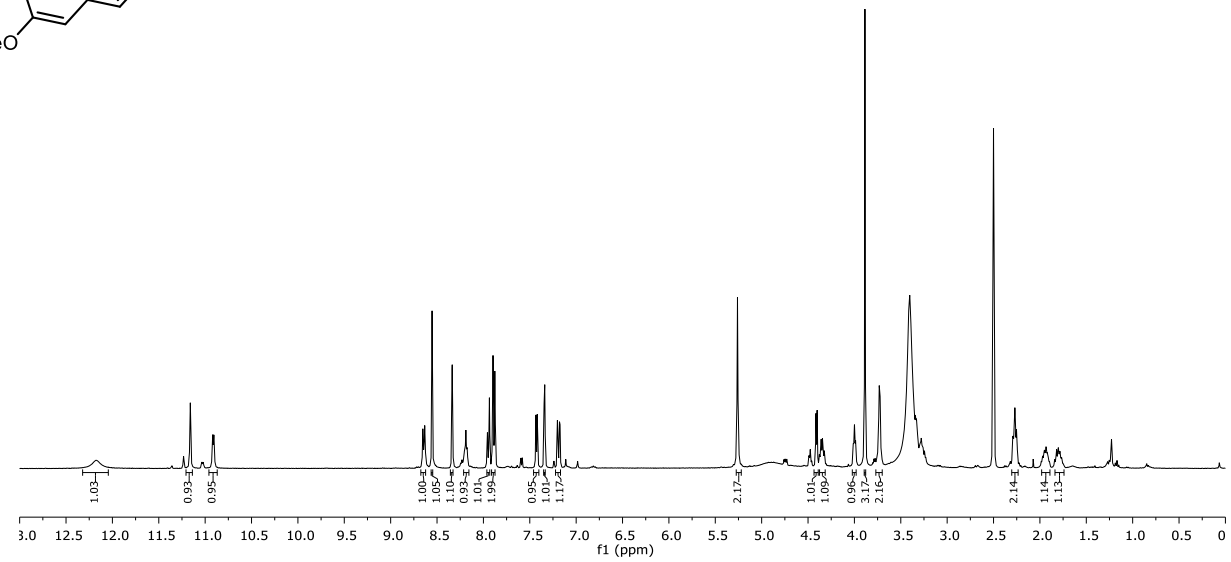

HO
O
O
N
H
N
H
O
HN
NH
O
O
N
N
N
O
OH
OH
F

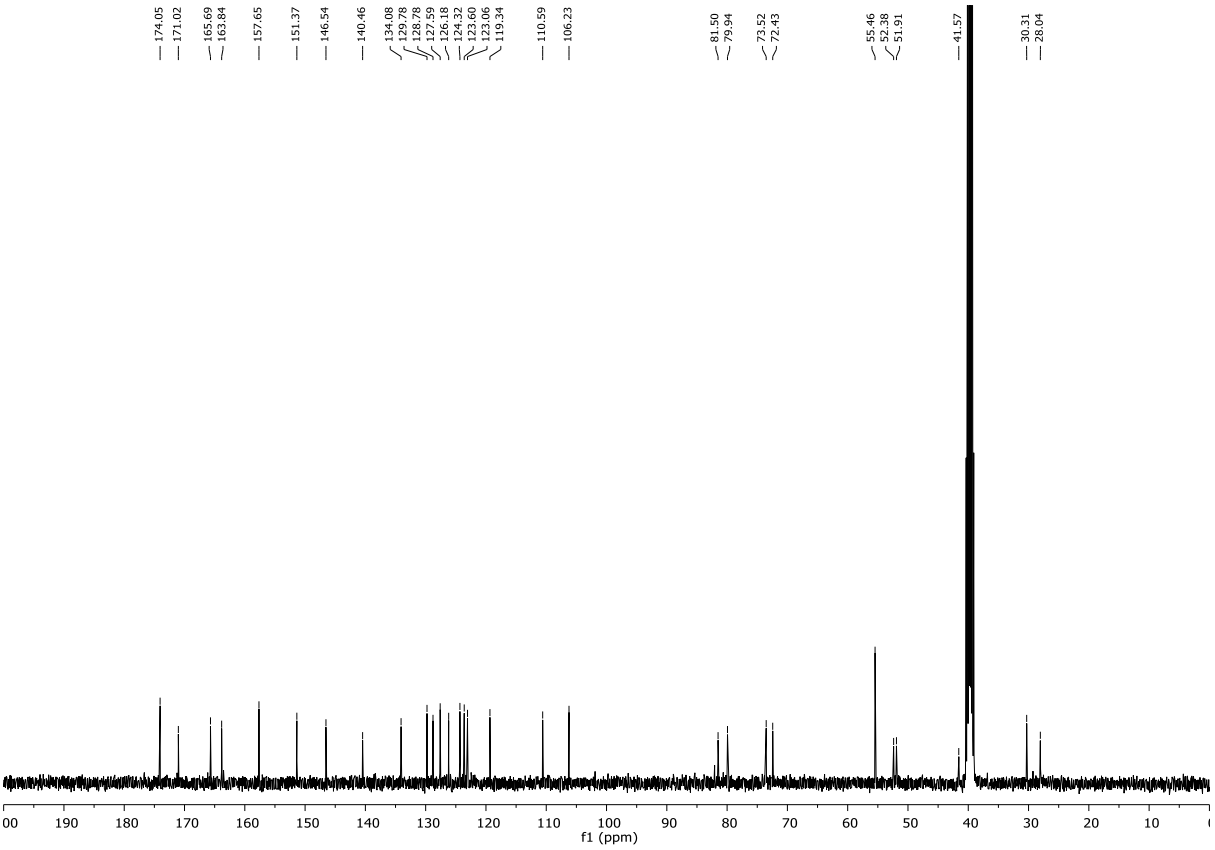
PU4b


1.03
0.97
1.00
1.06
1.18
1.00
1.00
1.07
1.04
1.01
1.11
2.19
0.90
1.03
1.03
1.12
1.09
2.12
0.99
2.21
1.11
1.12
f1 (ppm)


173.83
170.78
165.35
163.63
162.61
151.16
145.04
140.24
133.13
131.07
123.70
121.16
114.53
111.68
110.39
81.27
79.74
73.30
72.23
52.17
51.72
41.36
30.09
27.84
f1 (ppm)


HO
O
O
N
H
N
H
O
HN
NH
O
O
N
N
N
O
OH
OH
PU4e
MeO


1.03
0.93
0.95
1.00
1.05
1.10
0.93
1.01
1.99
0.95
1.01
1.17
2.17
1.01
1.09
0.96
3.17
2.16
2.14
1.14
1.13
f1 (ppm)


174.05
171.02
165.69
163.84
157.65
151.37
146.54
140.46
134.08
129.78
128.78
127.59
126.18
124.32
123.60
123.06
119.34
110.59
106.23
81.50
79.94
73.52
72.43
55.46
52.38
51.91
41.57
30.31
28.04
f1 (ppm)