

# Perfluorinated HDAC inhibitors as Selective Anticancer Agents

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**EXPERIMENTAL METHODS:** Commercially available reagents were used as received from suppliers. Solvents were laboratory grade and dried using an appropriate drying agent when required. Reactions requiring anhydrous conditions were carried out under an atmosphere of dry argon using Schlenk-line techniques. Where appropriate, solvents were degassed using the freeze-thaw cycle method. Thin-layer chromatography was carried out on silica plates (Merck 5554) or neutral alumina plates (Merck Art 5550) and visualised under UV (254/365 nm) irradiation or by staining with iodine. Preparative column chromatography was carried out using silica (Merck Silica Gel 60, 230400 mesh) or neutral alumina (Merck Aluminium Oxide 90, activity II-III, 70230 mesh), pre-soaked in ethyl acetate. pH measurements were carried out at 295 K using a Thermo Scientific Orion Star A111 pH meter with a Sigma-Aldrich micro-pH combination electrode. Calibration was performed using commercially

available buffer solutions at pH = 4.0  $\pm$ 0.02, pH = 7.00  $\pm$ 0.02 and pH = 10.00  $\pm$ 0.02. NMR spectra ( $^1\text{H}$ ,  $^{13}\text{C}$ ,  $^{31}\text{P}$ ) were recorded on a Varian VXR-400 spectrometer ( $^1\text{H}$  at 399.97 MHz,  $^{13}\text{C}$  at 100.57 MHz,  $^{19}\text{F}$  at 376.5 MHz) or a Varian VNMR-700 spectrometer ( $^1\text{H}$  at 699.73 MHz,  $^{13}\text{C}$  at 175.95 MHz). Spectra were recorded at 295 K in commercially available deuterated solvents and referenced internally to the residual solvent proton resonances ( $\text{CDCl}_3$  at 7.26 ppm;  $\text{DMSO-d}_6$  at 2.50 ppm;  $\text{CD}_3\text{OD}$  at 3.31 ppm), carbon resonances ( $\text{CDCl}_3$  at 77.2 ppm;  $\text{DMSO-d}_6$  at 29.8 ppm;  $\text{CD}_3\text{OD}$  at 49.0).  $^{19}\text{F}$  NMR spectra were calibrated to  $\text{CF}_3\text{CO}_2\text{H}$  (−76.6 ppm) prior to running experiments. Electrospray ionisation high resolution mass spectrometry was performed on a Thermo-Finnigan LTQ FT system using methanol as the carrier solvent.

**Cell culture.** Human A2780 ovarian carcinoma cells were obtained from the European Centre of Cell Cultures (ECACC, UK) and noncancerous HEK-293 cells were obtained from ATCC (Sigma, Switzerland). A2780 were routinely grown in RPMI 1640 GlutaMAX (Lifetechnologies, Switzerland), while HEK-293 were grown in DMEM medium, both containing 10% heat-inactivated fetal bovine serum (FBS, Pan Biotech, Germany) and 1% antibiotics (penicillin/streptomycin), at 37 °C and  $\text{CO}_2$  (5%).

**Determination of antiproliferative activity.** Cytotoxicity was determined using the MTT assay<sup>1</sup> (MTT = 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2H-tetrazolium bromide). Cells were seeded in 96-well plates as monolayers with 100  $\mu\text{L}$  of cell solution per well and pre-incubated for 24 h in the cell culture medium. Compounds were prepared as DMSO stock solutions that were dissolved in the culture medium and twofold serially diluted to the appropriate concentration to give a final DMSO concentration of maximum 0.5%. 100  $\mu\text{L}$  of the drug solution were added to each well and the plates were incubated for another 72 h. Subsequently, MTT (5 mg/mL solution, 20  $\mu\text{L}$  per well) was added to the cells and the plates were incubated for further 4 h. The culture medium was aspirated, and the purple formazan crystals formed by the mitochondrial dehydrogenase activity of vital cells were dissolved in DMSO. The optical density, directly proportional to the number of surviving cells, was quantified at 590

nm using a multiwell plate reader (Molecular Devices). The fraction of surviving cells was calculated from the absorbance of untreated control cells. The IC<sub>50</sub> values for the inhibition of cell growth were determined by fitting the plot of the logarithmic percentage of surviving cells against the logarithm of the drug concentration using a linear regression function. Evaluation is based on means from three independent experiments, each comprising four tests per concentration level.

**Enzyme Inhibition Assay.** HDAC enzyme inhibition assays were carried out using a commercially available assay kit (EnzoLifeSciences). HeLa nuclear extract, potential inhibitor (final concentration 1  $\mu$ M or 100 nM) and assay buffer (50 mM Tris/Cl, pH 8.0, 137 mM NaCl, 2.7 mM KCl, 1 mM MgCl<sub>2</sub>) were incubated in a 96 well plate at 37 °C for 15 min. Acetylated substrate (100  $\mu$ M) was added and the mixture incubated at 37 °C for a further 20 min. Developer was added and the mixture incubated at 37 °C for a further 15 min. Fluorescence was measured using a microplate reader ( $\lambda_{\text{ex}}$  = 360 nm,  $\lambda_{\text{em}}$  = 460 nm). Percentage HDAC activity is determined by quantification of fluorescence relative to control, with no added inhibitor.

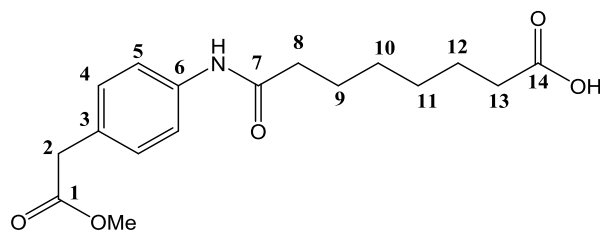
**Docking Study.** Docking was performed using GOLD Suite. The protein structure of HDAC 8 with SAHA bound substrate (PDB ID: 1t69) was prepared using the software's protein preparation wizard. Docking grids were defined as within 10 Å of the SAHA bound substrate. The docked molecules SAHA and **3b** were prepared using the software wizard. Each hydroxamic acid group is deprotonated, based on pK<sub>a</sub> values. SAHA and **3b** were then minimised in the software's docking program with CHEM-PLP used to rank poses. The top 10 poses were kept for visual inspection. Clustering of poses were used to determine most likely docking mode and poses shown in Fig 2 were chosen as an example of most popular cluster.

## Synthetic Details

**Methyl 2-(4-aminophenyl)acetate.** Thionyl chloride (2.4 mL, 33 mmol) was added dropwise to a solution of 4-aminophenylacetic acid (5.0 g, 33 mmol) in anhydrous methanol (25 mL). The mixture was stirred at 60 °C for 16 h. The resulting solution was cooled and the solvent was removed under reduced pressure. The brown solid hydrochloride salt of the title compound was triturated with Et<sub>2</sub>O (2 × 20 mL) to remove impurities. The free amine was liberated from its hydrochloride salt by addition of aqueous NaHCO<sub>3</sub> (2M, 50 mL), followed by extraction into CHCl<sub>3</sub> (3 × 30 mL). The organic layers were combined, dried over MgSO<sub>4</sub> and the solvent removed to leave the title compound as a light brown oil (3.2 g, 59%),  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 7.08 (2H, d, *J* 8.4 Hz, H<sup>4</sup>), 6.65 (2H, d, *J* 8.4 Hz, H<sup>5</sup>), 3.69 (3H, s, OCH<sub>3</sub>), 3.53 (2H, s, H<sup>2</sup>), 3.46 (2H, br s, NH<sub>2</sub>);  $\delta_{\text{C}}$  (CDCl<sub>3</sub>) 172.7 (C<sup>1</sup>), 145.4 (C<sup>6</sup>), 130.1 (C<sup>4</sup>), 123.8 (C<sup>3</sup>), 115.3 (C<sup>5</sup>), 51.9 (OCH<sub>3</sub>), 40.3 (C<sup>2</sup>); *m/z* (ESI ESI HRMS<sup>+</sup>) 166.0870 [M + H]<sup>+</sup> ([C<sub>9</sub>H<sub>12</sub>NO<sub>2</sub>]<sup>+</sup> requires 166.0868).

**Suberoyl anhydride.** Following a literature procedure,<sup>2</sup> a solution of suberic acid (7.0 g, 40 mmol in acetic anhydride (14.1 mL) was heated at 140 °C for 3 h. Excess anhydride was removed by vacuum distillation to leave the *title compound* as a white solid (6.1 g, 98%),  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 2.56–2.45 (4H, m, H<sup>2</sup>), 1.62–1.43 (4H, m, H<sup>3</sup>), 1.36–1.24 (4H, m, H<sup>4</sup>);  $\delta_{\text{C}}$  (CDCl<sub>3</sub>) 170.1 (C<sup>1</sup>), 34.8 (C<sup>2</sup>), 28.2 (C<sup>3</sup>), 23.9 (C<sup>4</sup>).

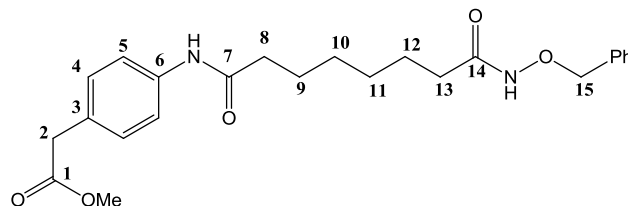
## 8-((4-(2-Methoxy-2-oxoethyl)phenyl)amino)-8-oxooctanoic acid, 2



Methyl 2-(4-aminophenyl)acetate (700 mg, 4.69 mmol), suberoyl anhydride (560 mg, 3.59 mmol) and NaHCO<sub>3</sub> (200 mg, 2.38 mmol) were dissolved in anhydrous THF (15 mL) and stirred at 22 °C for 16

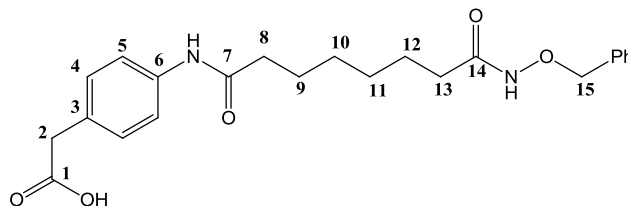
h. Solid impurities were removed by filtration and the solvent removed under reduced pressure to give the *title compound* as a white solid that was used without further purification (1.02 g, 88%),  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 7.40 (2H, d,  $J$  8.4 Hz,  $\text{H}^5$ ), 7.10 (2H, d,  $J$  8.4 Hz,  $\text{H}^4$ ), 3.62 (3H, s,  $\text{OCH}_3$ ), 3.49 (2H, s,  $\text{H}^2$ ), 2.32–2.22 (4H, m,  $\text{H}^8$  and  $\text{H}^{13}$ ), 1.71–1.51 (4H, m,  $\text{H}^9$  and  $\text{H}^{12}$ ), 1.36–1.24 (4H, m,  $\text{H}^{10}$  and  $\text{H}^{11}$ );  $\delta_{\text{C}}$  ( $\text{CDCl}_3$ ) 179.6 ( $\text{C}^{14}$ ), 172.3 ( $\text{C}^1$ ), 171.6 ( $\text{C}^7$ ), 136.9 ( $\text{C}^6$ ), 129.8 ( $\text{C}^4$ ), 129.7 ( $\text{C}^3$ ), 120.1 ( $\text{C}^5$ ), 52.1 ( $\text{OCH}_3$ ), 40.6 ( $\text{C}^2$ ), 37.5 ( $\text{C}^8$ ), 33.9 ( $\text{C}^{13}$ ), 28.8 ( $\text{C}^{10/11}$ ), 28.7 ( $\text{C}^{10/11}$ ), 25.3 ( $\text{C}^{9/12}$ ), 24.4 ( $\text{C}^{9/12}$ );  $m/z$  (ESI ESI HRMS<sup>+</sup>) 322.1656 [ $\text{M} + \text{H}$ ]<sup>+</sup> ( $[\text{C}_{17}\text{H}_{24}\text{NO}_5]^+$  requires 322.1649).

### Methyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate



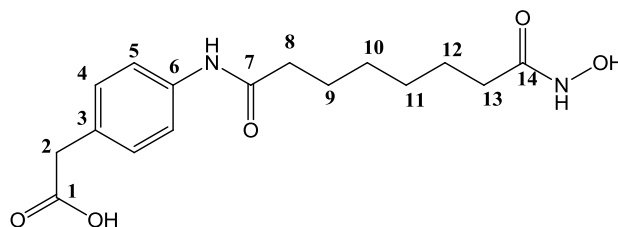
To a solution of 8-((4-(2-methoxy-2-oxoethyl)phenyl)amino)-8-oxooctanoic acid, **2**, (770 mg, 2.40 mmol) in anhydrous DMF (15 mL) was added HATU (865 mg, 3.60 mmol). The mixture was stirred for 10 min before O-benzylhydroxylamine hydrochloride (575 mg, 3.60 mmol) and *N,N*-diisopropylethylamine (1.25 mL, 7.20 mmol) were added. The solution was stirred at 22 °C for 16 h before the resultant solution was dropped into aqueous NaCl (2M, 60 mL). The suspension was cooled to 5 °C for 1 h and the precipitate collected by filtration. Recrystallisation from hot EtOAc afforded the *title compound* as an off-white solid (740 mg, 72%),  $\delta_{\text{H}}$  ( $\text{d}_6$ -DMSO) 10.94 (1H, br s,  $\text{CONHOBn}$ ), 9.86 (1H, br s,  $\text{ArNHCO}$ ), 7.53 (2H, d,  $J$  8.4 Hz,  $\text{H}^5$ ), 7.40–7.32 (5H, m, Ph), 7.17 (2H, d,  $J$  8.4 Hz,  $\text{H}^4$ ), 4.79 (2H, s,  $\text{H}^{15}$ ), 3.61 (3H, s,  $\text{OCH}_3$ ), 3.59 (2H, s,  $\text{H}^2$ ), 2.31 (2H, t,  $J$  7.6 Hz,  $\text{H}^8$ ), 1.95 (2H, t,  $J$  7.6 Hz,  $\text{H}^{13}$ ), 1.65–1.40 (4H, m,  $\text{H}^9$  and  $\text{H}^{12}$ ), 1.37–1.20 (4H, m,  $\text{H}^{10}$  and  $\text{H}^{11}$ );  $\delta_{\text{C}}$  ( $\text{d}_6$ -DMSO) 172.2 ( $\text{C}^1$ ), 171.6 ( $\text{C}^7$ ), 169.8 ( $\text{C}^{14}$ ), 138.6 ( $\text{C}^6$ ), 136.6 ( $\text{C}^{\text{Ph}}$ ), 130.0 ( $\text{C}^4$ ), 129.3 ( $\text{C}^{\text{Ph}}$ ), 129.2 ( $\text{C}^3$ ), 128.7 ( $\text{C}^{\text{Ph}}$ ), 128.6 ( $\text{C}^{\text{Ph}}$ ), 119.6 ( $\text{C}^5$ ), 77.2 ( $\text{C}^{15}$ ), 52.1 ( $\text{OCH}_3$ ), 39.9 ( $\text{C}^2$ ), 36.9 ( $\text{C}^8$ ), 32.7 ( $\text{C}^{13}$ ), 28.9 ( $\text{C}^{10/11}$ ), 28.8 ( $\text{C}^{10/11}$ ), 25.5 ( $\text{C}^{9/12}$ ), 25.3 ( $\text{C}^{9/12}$ );  $m/z$  (ESI ESI HRMS<sup>+</sup>) 449.2059 [ $\text{M} + \text{Na}$ ]<sup>+</sup> ( $[\text{C}_{24}\text{H}_{30}\text{N}_2\text{O}_5\text{Na}]^+$  requires 449.2047).

### Methyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetic acid, **3a**



Methyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate (185 mg, 0.43 mmol) was dissolved in 1:1 THF:H<sub>2</sub>O (4 mL). LiOH.H<sub>2</sub>O (36 mg, 0.87 mmol) in H<sub>2</sub>O (0.2 mL) was added and the homogenous solution was stirred at 22 °C for 1 h. The solvent was removed under reduced pressure until a precipitate began to form. Acidification by addition of HCl (2M) resulted in further precipitation. The mixture was placed in the fridge for 1 h before the precipitate was collected by filtration and dried under high vacuum to yield the *title compound* as a white solid (130 mg, 73%),  $\delta_{\text{H}}$  (d<sub>6</sub>-DMSO) 12.27 (1H, br s, CO<sub>2</sub>H), 10.95 (1H, br s, CONHOBn), 9.84 (1H, br s, ArNHCO), 7.52 (2H, d, *J* 8.4 Hz, H<sup>5</sup>), 7.45–7.30 (5H, m, Ph), 7.17 (2H, d, *J* 8.4 Hz, H<sup>4</sup>), 4.79 (2H, s, H<sup>15</sup>), 3.50 (2H, s, H<sup>2</sup>), 2.27 (2H, t, *J* 7.6 Hz, H<sup>8</sup>), 1.96 (2H, t, *J* 7.6 Hz, H<sup>13</sup>), 1.70–1.41 (4H, m, H<sup>9</sup> and H<sup>12</sup>), 1.35–1.15 (4H, m, H<sup>10</sup> and H<sup>11</sup>);  $\delta_{\text{C}}$  (d<sub>6</sub>-DMSO) 173.2 (C<sup>1</sup>), 171.5 (C<sup>7</sup>), 169.8 (C<sup>14</sup>), 138.4 (C<sup>6</sup>), 136.5 (C<sup>Ph</sup>), 130.0 (C<sup>4</sup>), 129.3 (C<sup>Ph</sup>), 129.2 (C<sup>3</sup>), 128.7 (C<sup>Ph</sup>), 128.6 (C<sup>Ph</sup>), 119.4 (C<sup>5</sup>), 77.2 (C<sup>15</sup>), 40.6 (C<sup>2</sup>), 36.8 (C<sup>8</sup>), 32.7 (C<sup>13</sup>), 28.9 (C<sup>10/11</sup>), 28.8 (C<sup>10/11</sup>), 25.5 (C<sup>9/12</sup>), 25.3 (C<sup>9/12</sup>); *m/z* (ESI HRMS<sup>+</sup>) 435.1896 [M + Na]<sup>+</sup> ([C<sub>23</sub>H<sub>28</sub>N<sub>2</sub>O<sub>5</sub>Na]<sup>+</sup> requires 435.1890).

### 2-(4-(8-(Hydroxyamino)-8-oxooctanamido)phenyl)acetic acid, **3b**



Methyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetic acid, **3a**, (30 mg, 0.073 mmol) and Pd/C (26 mg) in MeOH (5 mL) were reacted according to General Procedure 2 to give the *title*

*compound* as an off-white solid (18 mg, 77%),  $\delta_{\text{H}}$  ( $\text{d}^4\text{-MeOH}$ ) 7.48 (2H, d,  $J$  8.4 Hz,  $\text{H}^5$ ), 7.21 (2H, d,  $J$  8.4 Hz,  $\text{H}^4$ ), 3.54 (2H, s,  $\text{H}^2$ ), 2.34 (2H, t,  $J$  7.6 Hz,  $\text{H}^8$ ), 2.08 (2H, t,  $J$  7.6 Hz,  $\text{H}^{13}$ ), 1.72–1.58 (4H, m,  $\text{H}^9$  and  $\text{H}^{12}$ ), 1.45–1.32 (4H, m,  $\text{H}^{10}$  and  $\text{H}^{11}$ );  $\delta_{\text{C}}$  ( $\text{d}^4\text{-MeOH}$ ) 173.1 ( $\text{C}^1$ ), 171.5 ( $\text{C}^7$ ), 171.3 ( $\text{C}^{14}$ ), 137.2 ( $\text{C}^6$ ), 130.5 ( $\text{C}^3$ ), 129.3 ( $\text{C}^4$ ), 119.8 ( $\text{C}^5$ ), 40.1 ( $\text{C}^2$ ), 36.4 ( $\text{C}^8$ ), 32.3 ( $\text{C}^{13}$ ), 28.5 ( $\text{C}^{10/11}$ ), 28.4 ( $\text{C}^{10/11}$ ), 25.3 ( $\text{C}^{9/12}$ ), 25.1 ( $\text{C}^{9/12}$ );  $m/z$  (ESI HRMS $^-$ ) 321.1443 [ $\text{M} - \text{H}$ ] $^-$  ( $[\text{C}_{16}\text{H}_{21}\text{N}_2\text{O}_5]^-$  requires 321.1456); Anal. Calcd. for  $\text{C}_{16}\text{H}_{22}\text{N}_2\text{O}_5(\text{LiCl})_{0.4}$ : C, 56.64; H, 6.54; N, 8.25. Found: C, 56.68; H, 6.65; N, 8.23

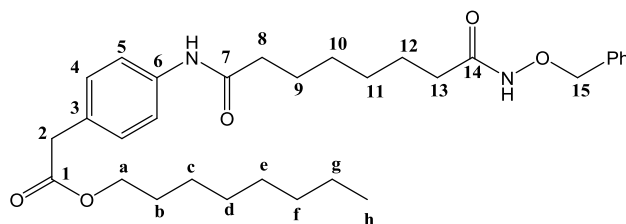
### General Procedure 1

Methyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetic acid, **3a**, (500 mg, 1.21 mmol) was dissolved in DMF (6 mL) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide (233 mg, 1.21 mmol). After stirring for 10 min, the *alcohol* (1 equiv., 1.21 mmol) and 4-dimethylaminopyridine (30 mg, 0.24 mmol) were added and the solution stirred at 22 °C for 16 h. The solution was dropped into  $\text{H}_2\text{O}$  (25 mL), leading to the formation of a white precipitate. The mixture was cooled in the fridge for 1 h before the precipitate was collected by filtration. The crude product was recrystallised from hot EtOAc (2 mL).

### General Procedure 2

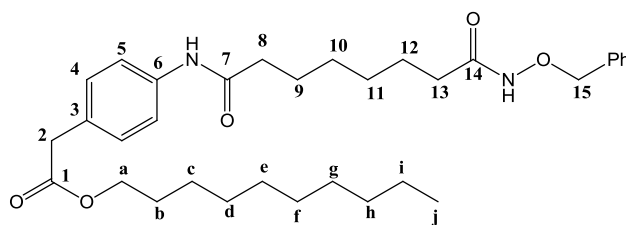
The *ester compound*, **4a-f**, (1 equiv.) was dissolved in MeOH and 5 mol% Pd/C (0.2 equiv.) was added. The mixture was purged with  $\text{H}_2$  and stirred under an atmosphere of  $\text{H}_2$  at 22 °C for 1 h. The mixture was filtered through Celite, which was rinsed with MeOH (5 mL). The MeOH fractions were combined and dried under reduced pressure.

**Octyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, 4a**



1-Octanol (190  $\mu$ L, 1.21 mmol) was reacted according to General Procedure 1, to form the *title compound* (360 mg, 57%),  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 9.07 (1H, br s, CONHOBn), 8.13 (1H, br s, ArNHCO), 7.43 (2H, d,  $J$  8.0 Hz,  $\text{H}^5$ ), 7.33–7.19 (5H, m, Ph), 7.10 (2H, d,  $J$  8.0 Hz,  $\text{H}^4$ ), 4.80 (2H, s,  $\text{H}^{15}$ ), 3.98 (2H, t,  $J$  6.8 Hz,  $\text{H}^a$ ), 3.48 (2H, s,  $\text{H}^2$ ), 2.21 (2H, t,  $J$  7.6 Hz,  $\text{H}^8$ ), 2.00–1.87 (2H, m,  $\text{H}^{13}$ ), 1.64–1.44 (6H, m,  $\text{H}^9$ ,  $\text{H}^{12}$  and  $\text{H}^b$ ), 1.34–1.08 (14H, m,  $\text{H}^{10}$ ,  $\text{H}^{11}$ ,  $\text{H}^c$ ,  $\text{H}^d$ ,  $\text{H}^e$ ,  $\text{H}^f$  and  $\text{H}^g$ ), 0.80 (3H, t,  $J$  6.8 Hz,  $\text{H}^h$ );  $\delta_{\text{C}}$  ( $\text{CDCl}_3$ ) 172.0 ( $\text{C}^1$ ), 171.8 ( $\text{C}^7$ ), 171.1 ( $\text{C}^{14}$ ), 137.3 ( $\text{C}^6$ ), 135.4 ( $\text{C}^{\text{Ph}}$ ), 129.7 ( $\text{C}^4$ ), 129.6 ( $\text{C}^3$ ), 129.1 ( $\text{C}^{\text{Ph}}$ ), 128.7 ( $\text{C}^{\text{Ph}}$ ), 128.5 ( $\text{C}^{\text{Ph}}$ ), 120.0 ( $\text{C}^5$ ), 78.1 ( $\text{C}^{15}$ ), 65.2 ( $\text{C}^a$ ), 40.8 ( $\text{C}^2$ ), 37.2 ( $\text{C}^8$ ), 32.9 ( $\text{C}^{13}$ ), 31.8 ( $\text{C}^b$ ), 29.1 ( $\text{C}^c$ ), 29.0 ( $\text{C}^d$ ), 28.6 ( $\text{C}^e$ ), 28.5 ( $\text{C}^{10/11}$ ), 28.4 ( $\text{C}^{10/11}$ ), 25.9 ( $\text{C}^f$ ), 25.3 ( $\text{C}^{9/12}$ ), 25.1 ( $\text{C}^{9/12}$ ), 22.6 ( $\text{C}^g$ ), 14.2 ( $\text{C}^h$ );  $m/z$  (ESI HRMS $^+$ ) 547.3148 [ $\text{M} + \text{Na}$ ] $^+$  [ $\text{C}_{31}\text{H}_{44}\text{N}_2\text{O}_5\text{Na}$ ] $^+$  requires 547.3142;  $t_R$  (Method A) = 10.6 min, purity: 98.1% (see below for HPLC method and traces).

**Decyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, 4b**

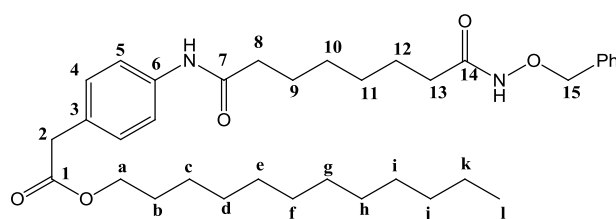


1-Decanol (231  $\mu$ L, 1.21 mmol) was reacted according to General Procedure 1, to form the *title compound* (173 mg, 26%),  $\delta_{\text{H}}$  ( $\text{d}_4\text{-MeOH}$ ) 7.41 (2H, d,  $J$  8.0 Hz,  $\text{H}^5$ ), 7.33–7.20 (5H, m, Ph), 7.10 (2H, d,  $J$  8.0 Hz,  $\text{H}^4$ ), 4.72 (2H, s,  $\text{H}^{15}$ ), 3.97 (2H, t,  $J$  6.8 Hz,  $\text{H}^a$ ), 3.48 (2H, s,  $\text{H}^2$ ), 2.24 (2H, t,  $J$  7.2 Hz,  $\text{H}^8$ ),



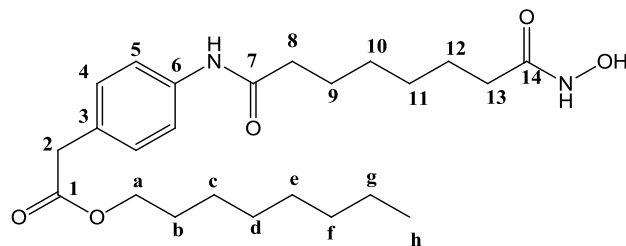
1.95 (2H, t,  $J$  7.6 Hz,  $H^{13}$ ), 1.63–1.44 (6H, m,  $H^9$ ,  $H^{12}$  and  $H^b$ ), 1.30–1.13 (18H, m,  $H^{10}$ ,  $H^{11}$ ,  $H^c$ ,  $H^d$ ,  $H^e$ ,  $H^f$ ,  $H^g$ ,  $H^h$  and  $H^i$ ), 0.80 (3H, t,  $J$  6.4 Hz,  $H^j$ );  $\delta_C$  ( $d_4$ -MeOH) 173.1 ( $C^1$ ), 172.2 ( $C^7$ ), 171.4 ( $C^{14}$ ), 137.5 ( $C^6$ ), 135.6 ( $C^{Ph}$ ), 130.0 ( $C^3$ ), 129.3 ( $C^4$ ), 128.9 ( $C^{Ph}$ ), 128.2 ( $C^{Ph}$ ), 128.1 ( $C^{Ph}$ ), 119.8 ( $C^5$ ), 77.5 ( $C^{15}$ ), 64.7 ( $C^a$ ), 40.2 ( $C^2$ ), 36.5 ( $C^8$ ), 32.2 ( $C^{13}$ ), 31.6 ( $C^b$ ), 29.2 ( $C^c$ ), 29.0 ( $C^d$ ), 28.9 ( $C^e$ ), 28.6 ( $C^{10/11}$ ), 28.5 ( $C^f$ ), 28.4 ( $C^g$ ), 28.4 ( $C^{10/11}$ ), 25.5 ( $C^h$ ), 25.3 ( $C^{9/12}$ ), 25.1 ( $C^{9/12}$ ), 22.3 ( $C^i$ ), 13.1 ( $C^j$ );  $m/z$  (ESI HRMS $^+$ ) 575.3464  $[M + Na]^+$  ( $[C_{33}H_{48}N_2O_5Na]^+$  requires 575.3455).

#### Dodecyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, 4c



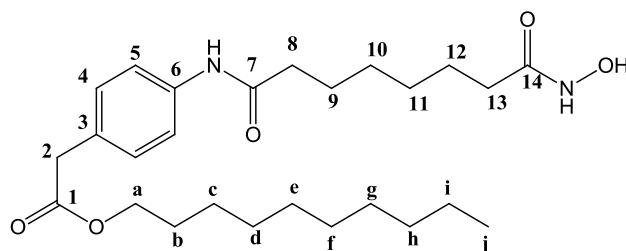
1-Dodecanol (225 mg, 1.21 mmol) was reacted according to General Procedure 1, to form the *title compound* (320 mg, 46%),  $\delta_H$  ( $CDCl_3$ ) 7.50 (2H, d,  $J$  8.0 Hz,  $H^5$ ), 7.42–7.36 (5H, m, Ph), 7.23 (2H, d,  $J$  8.0 Hz,  $H^4$ ), 4.93 (2H, s,  $H^{15}$ ), 4.09 (2H, t,  $J$  6.8 Hz,  $H^a$ ), 3.58 (2H, s,  $H^2$ ), 2.23 (2H, t,  $J$  7.2 Hz,  $H^8$ ), 2.10–1.99 (2H, m,  $H^{13}$ ), 1.77–1.53 (6H, m,  $H^9$ ,  $H^{12}$  and  $H^b$ ), 1.42–1.26 (22H, m,  $H^{10}$ ,  $H^{11}$ ,  $H^c$ ,  $H^d$ ,  $H^e$ ,  $H^f$ ,  $H^g$ ,  $H^h$ ,  $H^i$ ,  $H^j$  and  $H^k$ ), 0.91 (3H, t,  $J$  6.4 Hz,  $H^j$ );  $\delta_C$  ( $CDCl_3$ ) 171.9 ( $C^1$ ), 171.7 ( $C^7$ ), 171.0 ( $C^{14}$ ), 137.2 ( $C^6$ ), 135.3 ( $C^{Ph}$ ), 130.0 ( $C^3$ ), 129.7 ( $C^4$ ), 129.2 ( $C^{Ph}$ ), 128.7 ( $C^{Ph}$ ), 128.6 ( $C^{Ph}$ ), 119.9 ( $C^5$ ), 78.1 ( $C^{15}$ ), 65.2 ( $C^a$ ), 40.9 ( $C^2$ ), 37.2 ( $C^8$ ), 32.9 ( $C^{13}$ ), 31.9 ( $C^b$ ), 29.8–28.2 (9C,  $C^c$ ,  $C^d$ ,  $C^e$ ,  $C^f$ ,  $C^g$ ,  $C^h$ ,  $C^i$ ,  $C^{10}$ , and  $C^{11}$ ), 25.9 ( $C^j$ ), 25.2 ( $C^{9/12}$ ), 25.0 ( $C^{9/12}$ ), 20.6 ( $C^k$ ), 14.1 ( $C^l$ );  $m/z$  (ESI HRMS $^+$ ) 603.3773  $[M + Na]^+$  ( $[C_{35}H_{52}N_2O_5Na]^+$  requires 603.3768).

**Octyl 2-(4-(8-(hydroxyamino)-8-oxooctanamido)phenyl)acetate, 5a**



Octyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, **4a**, (150 mg, 0.29 mmol) and Pd/C (64 mg) in MeOH (10 mL) were reacted according to General Procedure 2 to give the *title compound* as an off-white solid (100 mg, 79%),  $\delta_{\text{H}}$  ( $d^4$ -MeOH) 7.41 (2H, d,  $J$  8.4 Hz,  $\text{H}^5$ ), 7.11 (2H, d,  $J$  8.4 Hz,  $\text{H}^4$ ), 3.98 (2H, t,  $J$  6.4 Hz,  $\text{H}^a$ ), 3.49 (2H, s,  $\text{H}^2$ ), 2.26 (2H, t,  $J$  7.2 Hz,  $\text{H}^8$ ), 1.99 (2H, t,  $J$  7.2 Hz,  $\text{H}^{13}$ ), 1.63–1.44 (6H, m,  $\text{H}^9$ ,  $\text{H}^{12}$  and  $\text{H}^b$ ), 1.35–1.11 (14H, m,  $\text{H}^{10}$ ,  $\text{H}^{11}$ ,  $\text{H}^c$ ,  $\text{H}^d$ ,  $\text{H}^e$ ,  $\text{H}^f$  and  $\text{H}^g$ ), 0.79 (3H, t,  $J$  7.2 Hz,  $\text{H}^h$ );  $\delta_{\text{C}}$  ( $d^4$ -MeOH) 173.2 ( $\text{C}^1$ ), 172.3 ( $\text{C}^7$ ), 171.6 ( $\text{C}^{14}$ ), 137.5 ( $\text{C}^6$ ), 130.0 ( $\text{C}^3$ ), 129.3 ( $\text{C}^4$ ), 119.8 ( $\text{C}^5$ ), 64.6 ( $\text{C}^a$ ), 40.2 ( $\text{C}^2$ ), 36.5 ( $\text{C}^8$ ), 32.3 ( $\text{C}^{13}$ ), 31.5 ( $\text{C}^b$ ), 28.9 ( $\text{C}^c$ ), 28.8 ( $\text{C}^d$ ), 28.5 ( $\text{C}^{10/11}$ ), 28.4 ( $\text{C}^{10/11}$ ), 28.3 ( $\text{C}^e$ ), 25.5 ( $\text{C}^f$ ), 25.3 ( $\text{C}^{9/12}$ ), 25.2 ( $\text{C}^{9/12}$ ), 22.3 ( $\text{C}^g$ ), 13.0 ( $\text{C}^h$ );  $m/z$  (ESI HRMS<sup>+</sup>) 457.2683 [ $\text{M} + \text{Na}$ ]<sup>+</sup> ( $[\text{C}_{24}\text{H}_{38}\text{N}_2\text{O}_5\text{Na}]^+$  requires 457.2673); Anal. Calcd. for  $\text{C}_{24}\text{H}_{38}\text{N}_2\text{O}_5$ : C, 66.87; H, 8.91; N, 6.04. Found: C, 66.76; H, 8.98; N, 6.02.

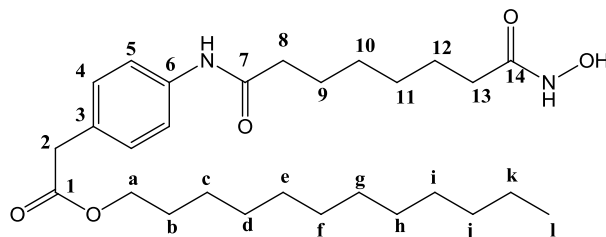
**Decyl 2-(4-(8-(hydroxyamino)-8-oxooctanamido)phenyl)acetate, 5b**



Decyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, **4b**, (173 mg, 0.31 mmol) and Pd/C (70 mg) in MeOH (10 mL) were reacted according to General Procedure 2 to give the *title compound* as an off-white solid (87 mg, 61%),  $\delta_{\text{H}}$  ( $d_6$ -DMSO) 10.34 (1H, br s,  $\text{NHOH}$ ), 9.85 (1H, br s,  $\text{ArNHCO}$ ), 8.66 (1H, br s,  $\text{NHOH}$ ), 7.52 (2H, d,  $J$  8.0 Hz,  $\text{H}^5$ ), 7.17 (2H, d,  $J$  8.0 Hz,  $\text{H}^4$ ), 4.02 (2H, t,  $J$  6.4 Hz,  $\text{H}^a$ ),

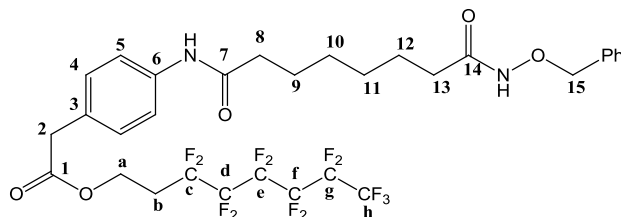
3.59 (2H, s, H<sup>2</sup>), 2.29 (2H, t, *J* 7.2 Hz, H<sup>8</sup>), 1.94 (2H, t, *J* 7.4 Hz, H<sup>13</sup>), 1.62–1.45 (6H, m, H<sup>9</sup>, H<sup>12</sup> and H<sup>b</sup>), 1.34–1.20 (18H, m, H<sup>10</sup>, H<sup>11</sup>, H<sup>c</sup>, H<sup>d</sup>, H<sup>e</sup>, H<sup>f</sup>, H<sup>g</sup>, H<sup>h</sup> and H<sup>i</sup>), 0.86 (3H, t, *J* 6.4 Hz, H<sup>j</sup>);  $\delta_{\text{C}}$  (d<sub>6</sub>-DMSO) 171.8 (C<sup>1</sup>), 171.6 (C<sup>7</sup>), 169.5 (C<sup>14</sup>), 138.6 (C<sup>6</sup>), 129.9 (C<sup>3</sup>), 129.3 (C<sup>4</sup>), 119.4 (C<sup>5</sup>), 64.6 (C<sup>a</sup>), 40.6 (C<sup>2</sup>), 36.8 (C<sup>8</sup>), 32.7 (C<sup>13</sup>), 31.8 (C<sup>b</sup>), 29.4 (C<sup>c</sup>), 29.4 (C<sup>d</sup>), 29.2 (C<sup>e</sup>), 29.1 (C<sup>10/11</sup>), 28.9 (C<sup>f</sup>), 28.8 (C<sup>g</sup>), 28.5 (C<sup>10/11</sup>), 25.8 (C<sup>h</sup>), 25.5 (C<sup>9/12</sup>), 25.4 (C<sup>9/12</sup>), 22.6 (C<sup>i</sup>), 14.4 (C<sup>j</sup>); *m/z* (ESI HRMS<sup>+</sup>) 485.2987 [M + Na]<sup>+</sup> ([C<sub>26</sub>H<sub>42</sub>N<sub>2</sub>O<sub>5</sub>Na]<sup>+</sup> requires 485.2985); Anal. Calcd. for C<sub>26</sub>H<sub>42</sub>N<sub>2</sub>O<sub>5</sub>: C, 67.50; H, 9.15; N, 6.06. Found: C, 67.16; H, 9.21; N, 5.67.

### Dodecyl 2-(4-(8-(hydroxyamino)-8-oxooctanamido)phenyl)acetate, **5c**



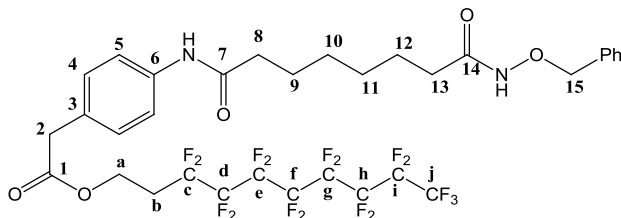
Dodecyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, **4c**, (250 mg, 0.43 mmol) and Pd/C (100 mg) in MeOH (20 mL) were reacted according to General Procedure 2. The crude material was triturated with H<sub>2</sub>O (2 × 2 mL) to remove residual MeOH to give the *title compound* as an off-white solid (168 mg, 80%),  $\delta_{\text{H}}$  (d<sub>6</sub>-DMSO) 10.34 (1H, br s, NHOH), 9.85 (1H, br s, ArNHCO), 8.67 (1H, br s, NHOH), 7.53 (2H, d, *J* 8.0 Hz, H<sup>5</sup>), 7.17 (2H, d, *J* 8.0 Hz, H<sup>4</sup>), 4.02 (2H, t, *J* 6.8 Hz, H<sup>a</sup>), 3.59 (2H, s, H<sup>2</sup>), 2.29 (2H, t, *J* 7.6 Hz, H<sup>8</sup>), 1.95 (2H, t, *J* 7.2 Hz, H<sup>13</sup>), 1.62–1.45 (6H, m, H<sup>9</sup>, H<sup>12</sup> and H<sup>b</sup>), 1.34–1.18 (22H, m, H<sup>10</sup>, H<sup>11</sup>, H<sup>c</sup>, H<sup>d</sup>, H<sup>e</sup>, H<sup>f</sup>, H<sup>g</sup>, H<sup>h</sup>, H<sup>i</sup>, H<sup>j</sup> and H<sup>k</sup>), 0.87 (3H, t, *J* 6.4 Hz, H<sup>j</sup>);  $\delta_{\text{C}}$  (d<sub>6</sub>-DMSO) 171.8 (C<sup>1</sup>), 171.6 (C<sup>7</sup>), 169.5 (C<sup>14</sup>), 138.6 (C<sup>6</sup>), 129.9 (C<sup>4</sup>), 129.3 (C<sup>3</sup>), 119.4 (C<sup>5</sup>), 64.6 (C<sup>a</sup>), 40.4 (C<sup>2</sup>), 36.8 (C<sup>8</sup>), 32.7 (C<sup>13</sup>), 31.8 (C<sup>b</sup>), 29.7–28.2 (9C, C<sup>c</sup>, C<sup>d</sup>, C<sup>e</sup>, C<sup>f</sup>, C<sup>g</sup>, C<sup>h</sup>, C<sup>i</sup>, C<sup>10</sup>, and C<sup>11</sup>), 25.7 (C<sup>j</sup>), 25.6 (C<sup>9/12</sup>), 25.5 (C<sup>9/12</sup>), 22.6 (C<sup>k</sup>), 14.4 (C<sup>l</sup>); *m/z* (ESI HRMS<sup>+</sup>) 513.3299 [M + Na]<sup>+</sup> ([C<sub>28</sub>H<sub>46</sub>N<sub>2</sub>O<sub>5</sub>Na]<sup>+</sup> requires 513.3298); Anal. Calcd. for C<sub>28</sub>H<sub>46</sub>N<sub>2</sub>O<sub>5</sub>·0.5(H<sub>2</sub>O): C, 67.30; H, 9.48; N, 5.61. Found: C, 66.93; H, 9.09; N, 5.75.

**3,3,4,4,5,5,6,6,7,7,8,8,8-Tridecafluorooctyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, 4d**



1*H*,1*H*,2*H*,2*H*-Perfluoro-1-octanol (267  $\mu$ L, 1.21 mmol) was reacted according to General Procedure 1, to form the *title compound* (369 mg, 40%),  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 7.43 (2H, d,  $J$  8.0 Hz,  $\text{H}^5$ ), 7.33–7.23 (5H, m, Ph), 7.13 (2H, d,  $J$  8.0 Hz,  $\text{H}^4$ ), 4.83 (2H, s,  $\text{H}^{15}$ ), 4.31 (2H, t,  $J$  6.4 Hz,  $\text{H}^a$ ), 3.52 (2H, s,  $\text{H}^2$ ), 2.37 (2H, tt,  $^3J_{\text{H-F}}$  18.4 Hz,  $^3J_{\text{H-H}}$  6.4 Hz,  $\text{H}^b$ ), 2.25 (2H, t,  $J$  7.2 Hz,  $\text{H}^8$ ), 2.02–1.92 (2H, m,  $\text{H}^{13}$ ), 1.68–1.49 (4H, m,  $\text{H}^9$  and  $\text{H}^{12}$ ), 1.36–1.19 (4H, m,  $\text{H}^{10}$  and  $\text{H}^{11}$ );  $\delta_{\text{C}}$  ( $\text{CDCl}_3$ ) 171.9 ( $\text{C}^1$ ), 171.6 ( $\text{C}^7$ ), 171.3 ( $\text{C}^{14}$ ), 137.3 ( $\text{C}^6$ ), 135.3 ( $\text{C}^{\text{Ph}}$ ), 129.8 ( $\text{C}^4$ ), 129.2 ( $\text{C}^{\text{Ph}}$ ), 129.1 ( $\text{C}^3$ ), 129.0 ( $\text{C}^{\text{Ph}}$ ), 128.6 ( $\text{C}^{\text{Ph}}$ ), 120.0 ( $\text{C}^5$ ), 78.2 ( $\text{C}^{15}$ ), 56.8 ( $\text{C}^a$ ), 40.4 ( $\text{C}^2$ ), 37.2 ( $\text{C}^8$ ), 32.8 ( $\text{C}^{13}$ ), 30.4 (t,  $^2J_{\text{C-F}}$  22 Hz,  $\text{C}^b$ ), 28.3 ( $\text{C}^{10/11}$ ), 28.0 ( $\text{C}^{10/11}$ ), 25.2 ( $\text{C}^{9/12}$ ), 24.9 ( $\text{C}^{9/12}$ ),  $\text{C}^c$ – $\text{C}^h$  not observed due to C-F coupling;  $\delta_{\text{F}}$  ( $\text{CDCl}_3$ ) -80.8, -113.6, -121.9, -122.9, -123.6, -126.1;  $m/z$  (ESI HRMS $^+$ ) 781.1923 [ $\text{M} + \text{Na}$ ] $^+$  [ $\text{C}_{31}\text{H}_{31}\text{F}_{13}\text{N}_2\text{O}_5\text{Na}$ ] $^+$  requires 781.1917).

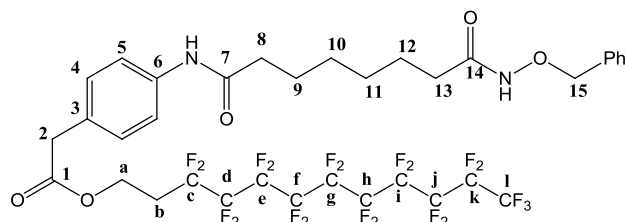
**3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptadecafluorodecyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, 4e**



1*H*,1*H*,2*H*,2*H*-Perfluoro-1-decanol (563 mg, 1.21 mmol) was reacted according to General Procedure 1, to form the *title compound* (230 mg, 22%),  $\delta_{\text{H}}$  ( $d_6$ -DMSO) 10.95 (1H, br s,  $\text{CONHOBn}$ ), 9.85 (1H, br s,  $\text{ArNHCO}$ ), 7.54 (2H, d,  $J$  8.4 Hz,  $\text{H}^5$ ), 7.44–7.32 (5H, m, Ph), 7.17 (2H, d,  $J$  8.4 Hz,  $\text{H}^4$ ), 4.79 (2H, s,  $\text{H}^{15}$ ), 4.35 (2H, t,  $J$  6.4 Hz,  $\text{H}^a$ ), 3.62 (2H, s,  $\text{H}^2$ ), 2.67 (2H, tt,  $^3J_{\text{H-F}}$  19.6 Hz,  $^3J_{\text{H-H}}$  5.6 Hz,  $\text{H}^b$ ), 2.29 (2H,

t,  $J$  7.2 Hz,  $H^8$ ), 1.96 (2H, t,  $J$  7.2 Hz,  $H^{13}$ ), 1.62–1.45 (4H, m,  $H^9$  and  $H^{12}$ ), 1.32–1.21 (4H, m,  $H^{10}$  and  $H^{11}$ );  $\delta_C$  ( $d_6$ -DMSO) 171.6 ( $C^1$ ), 171.5 ( $C^7$ ), 169.8 ( $C^{14}$ ), 138.7 ( $C^6$ ), 136.6 ( $C^{Ph}$ ), 130.0 ( $C^4$ ), 129.2 ( $C^{Ph}$ ), 129.1 ( $C^3$ ), 128.7 ( $C^{Ph}$ ), 128.7 ( $C^{Ph}$ ), 119.4 ( $C^5$ ), 77.2 ( $C^{15}$ ), 56.8 ( $C^a$ ), 39.9 ( $C^2$ ), 36.8 ( $C^8$ ), 32.7 ( $C^{13}$ ), 29.9 (t,  $^2J_{C-F}$  21 Hz,  $C^b$ ), 28.9 ( $C^{10/11}$ ), 28.8 ( $C^{10/11}$ ), 25.5 ( $C^{9/12}$ ), 25.3 ( $C^{9/12}$ ),  $C^c$ – $C^j$  not observed due to C-F coupling;  $\delta_F$  ( $d_6$ -DMSO) -80.2, -112.7, -121.5, -121.7 (2F), -122.5, -123.2, -125.8;  $m/z$  (ESI HRMS $^+$ ) 881.1861 [ $M + Na$ ] $^+$  [ $C_{33}H_{31}F_{17}N_2O_5Na$ ] $^+$  requires 881.1853).

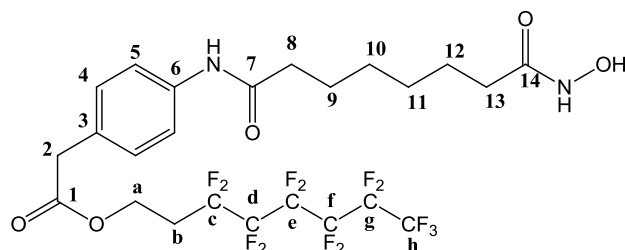
**3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,12-Henicosafuorododecyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, 4f**



1*H*,1*H*,2*H*,2*H*-Perfluoro-1-dodecanol (274 mg, 0.48 mmol) was dissolved in DMF (65 mL) and 2-(4-{7-[(benzyloxy)carbamoyl]heptanamido}phenyl)acetic acid, **3a**, (200 mg, 0.48 mmol), 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide (93 mg, 0.48 mmol) and DMAP (12 mg, 0.096 mmol) were added and the solution stirred at 22 °C for 16 h. The solution was dropped into H<sub>2</sub>O (300 mL), leading to the formation of an off-white precipitate. The mixture was cooled in the fridge for 1 h before the precipitate was collected by filtration. The crude product was recrystallised from hot EtOAc (2 mL) to give the *title compound* as an off-white solid (66 mg, 14%),  $\delta_H$  (CDCl<sub>3</sub>) 7.42 (2H, d,  $J$  8.0 Hz,  $H^5$ ), 7.34–7.22 (5H, m, Ph), 7.14 (2H, d,  $J$  8.0 Hz,  $H^4$ ), 4.84 (2H, s,  $H^{15}$ ), 4.31 (2H, t,  $J$  6.8 Hz,  $H^a$ ), 3.53 (2H, s,  $H^2$ ), 2.39 (2H, tt,  $^3J_{H-F}$  18.4 Hz,  $^3J_{H-H}$  5.6 Hz,  $H^b$ ), 2.26 (2H, t,  $J$  6.4 Hz,  $H^8$ ), 2.00–1.92 (2H, m,  $H^{13}$ ), 1.67–1.45 (4H, m,  $H^9$  and  $H^{12}$ ), 1.35–1.21 (4H, m,  $H^{10}$  and  $H^{11}$ );  $\delta_C$  (CDCl<sub>3</sub>) 171.7 ( $C^1$ ), 171.4 ( $C^7$ ), 170.1 ( $C^{14}$ ), 138.5 ( $C^6$ ), 136.9 ( $C^{Ph}$ ), 130.4 ( $C^4$ ), 129.9 ( $C^{Ph}$ ), 129.4 ( $C^3$ ), 129.1 ( $C^{Ph}$ ), 128.9 ( $C^{Ph}$ ), 119.8 ( $C^5$ ), 78.6 ( $C^{15}$ ), 56.7 ( $C^a$ ), 40.4 ( $C^2$ ), 37.2 ( $C^8$ ), 32.6 ( $C^{13}$ ), 28.2 ( $C^{10/11}$ ), 28.1 ( $C^{10/11}$ ), 25.2 ( $C^{9/12}$ ), 25.1 ( $C^{9/12}$ ),  $C^b$ – $C^l$  not

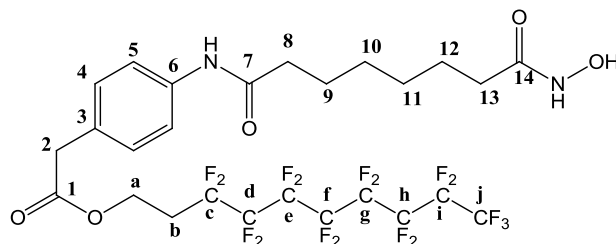
observed due to C-F coupling;  $\delta_F$  (CDCl<sub>3</sub>) -80.1, -112.6, -121.5 (4F), -121.6, -122.4, -123.1, -125.7;  $m/z$  (ESI HRMS<sup>+</sup>) 981.1796 [M + Na]<sup>+</sup> ([C<sub>35</sub>H<sub>31</sub>F<sub>21</sub>N<sub>2</sub>O<sub>5</sub>Na]<sup>+</sup> requires 981.1789).

**3,3,4,4,5,5,6,6,7,7,8,8,8-Tridecafluorooctyl 2-(4-(8-(hydroxyamino)-8-oxooctanamido)phenyl)acetate, 5d**



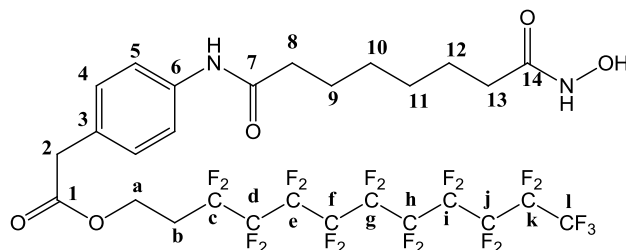
3,3,4,4,5,5,6,6,7,7,8,8,8-Tridecafluorooctyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, **4d**, (324 mg, 0.43 mmol) and Pd/C (95 mg) in MeOH (18 mL) were reacted according to General Procedure 2 to give the *title compound* as an off-white solid (228 mg, 79%),  $\delta_H$  (d<sub>6</sub>-DMSO) 10.34 (1H, br s, NHOH), 9.86 (1H, br s, ArNHCO), 8.67 (1H, br s, NHOH), 7.54 (2H, d,  $J$  8.0 Hz, H<sup>5</sup>), 7.18 (2H, d,  $J$  8.0 Hz, H<sup>4</sup>), 4.35 (2H, t,  $J$  6.0 Hz, H<sup>a</sup>), 3.63 (2H, s, H<sup>2</sup>), 2.68 (2H, tt,  $^3J_{H-F}$  20.4 Hz,  $^3J_{H-H}$  5.2 Hz, H<sup>b</sup>), 2.29 (2H, t,  $J$  7.2 Hz, H<sup>8</sup>), 1.95 (2H, t,  $J$  7.2 Hz, H<sup>13</sup>), 1.64–1.44 (4H, m, H<sup>9</sup> and H<sup>12</sup>), 1.35–1.22 (4H, m, H<sup>10</sup> and H<sup>11</sup>);  $\delta_C$  (d<sub>6</sub>-DMSO) 171.6 (C<sup>1</sup>), 171.5 (C<sup>7</sup>), 169.5 (C<sup>14</sup>), 138.7 (C<sup>6</sup>), 130.0 (C<sup>4</sup>), 128.8 (C<sup>3</sup>), 119.4 (C<sup>5</sup>), 56.8 (C<sup>a</sup>), 40.3 (C<sup>2</sup>), 36.8 (C<sup>8</sup>), 32.7 (C<sup>13</sup>), 29.9 (t,  $^2J_{C-F}$  22 Hz, C<sup>b</sup>), 28.9 (2C, C<sup>10</sup> and C<sup>11</sup>), 25.5 (2C, C<sup>9</sup> and C<sup>12</sup>), C<sup>c</sup>–C<sup>h</sup> not observed due to C-F coupling;  $\delta_F$  (d<sub>6</sub>-DMSO) -80.3, -112.7, -121.7, -122.7, -123.2, -125.7;  $m/z$  (ESI HRMS<sup>+</sup>) 691.1444 [M + Na]<sup>+</sup> ([C<sub>24</sub>H<sub>25</sub>F<sub>13</sub>N<sub>2</sub>O<sub>5</sub>Na]<sup>+</sup> requires 691.1448); Anal. Calcd. for C<sub>24</sub>H<sub>25</sub>F<sub>13</sub>N<sub>2</sub>O<sub>5</sub>: C, 43.12; H, 3.77; N, 4.19. Found: C, 43.07; H, 3.76; N, 4.18.

**3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptafluorodecyl 2-(4-(8-(hydroxyamino)-8-oxooctanamido)phenyl)acetate, 5e**



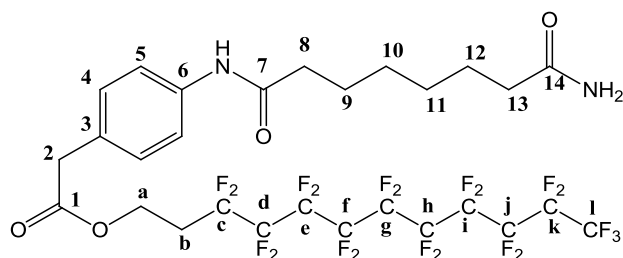
3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptafluorodecyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, **4e**, (230 mg, 0.27 mmol) and Pd/C (64 mg) in MeOH (12 mL) were reacted according to General Procedure 2 to give the *title compound* as an off-white solid (185 mg, 89%),  $\delta_{\text{H}}$  ( $d_6$ -DMSO) 10.34 (1H, br s, *NHOH*), 9.85 (1H, br s, *ArNHCO*), 8.66 (1H, br s, *NHOH*), 7.53 (2H, d, *J* 8.4 Hz,  $\text{H}^5$ ), 7.17 (2H, d, *J* 8.4 Hz,  $\text{H}^4$ ), 4.35 (2H, t, *J* 6.0 Hz,  $\text{H}^a$ ), 3.63 (2H, s,  $\text{H}^2$ ), 2.69 (2H, tt,  $^3J_{\text{H-F}}$  19.6 Hz,  $^3J_{\text{H-H}}$  5.6 Hz,  $\text{H}^b$ ), 2.29 (2H, t, *J* 6.4 Hz,  $\text{H}^8$ ), 1.95 (2H, t, *J* 7.2 Hz,  $\text{H}^{13}$ ), 1.62–1.46 (4H, m,  $\text{H}^9$  and  $\text{H}^{12}$ ), 1.33–1.21 (4H, m,  $\text{H}^{10}$  and  $\text{H}^{11}$ );  $\delta_{\text{C}}$  ( $d_6$ -DMSO) 171.6 ( $\text{C}^1$ ), 171.5 ( $\text{C}^7$ ), 169.6 ( $\text{C}^{14}$ ), 138.7 ( $\text{C}^6$ ), 130.0 ( $\text{C}^4$ ), 128.7 ( $\text{C}^3$ ), 119.4 ( $\text{C}^5$ ), 56.7 ( $\text{C}^a$ ), 40.0 ( $\text{C}^2$ ), 36.8 ( $\text{C}^8$ ), 32.7 ( $\text{C}^{13}$ ), 29.9 (t,  $^2J_{\text{C-F}}$  20 Hz,  $\text{C}^b$ ), 28.8 ( $\text{C}^{10}$  and  $\text{C}^{11}$ ), 25.5 ( $\text{C}^9$  and  $\text{C}^{12}$ ),  $\text{C}^c$ – $\text{C}^j$  not observed due to C-F coupling;  $\delta_{\text{F}}$  ( $d_6$ -DMSO) -80.8, -113.1, -121.8, -122.1 (2F), -122.9, -123.4, -126.2; *m/z* (ESI HRMS<sup>+</sup>) 791.1396 [*M* + Na]<sup>+</sup> ([*C*<sub>26</sub>*H*<sub>25</sub>*F*<sub>17</sub>*N*<sub>2</sub>*O*<sub>5</sub>Na]<sup>+</sup> requires 791.1384); Anal. Calcd. for *C*<sub>26</sub>*H*<sub>25</sub>*F*<sub>17</sub>*N*<sub>2</sub>*O*<sub>5</sub>: C, 40.64; H, 3.28; N, 3.65. Found: C, 40.50; H, 3.28; N, 3.70.

**3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,12-Henicosafuorododecyl 2-(4-(8-(hydroxyamino)-8-oxooctanamido)phenyl)acetate, 5f**



3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,12-Henicosafuorododecyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, **4f**, (60 mg, 0.063 mmol) and Pd/C (50 mg) in MeOH (100 mL) were reacted according to General Procedure 2 to give the *title compound* as an off-white solid (13 mg, 24%),  $\delta_{\text{H}}$  ( $d_4$ -MeOD) 7.51 (2H, d,  $J$  8.4 Hz,  $\text{H}^5$ ), 7.16 (2H, d,  $J$  8.4 Hz,  $\text{H}^4$ ), 4.34 (2H, t,  $J$  6.4 Hz,  $\text{H}^a$ ), 3.61 (2H, s,  $\text{H}^2$ ), 2.73–2.58 (2H, m,  $\text{H}^b$ ), 2.28 (2H, t,  $J$  7.2 Hz,  $\text{H}^8$ ), 1.94 (2H, t,  $J$  7.2 Hz,  $\text{H}^{13}$ ), 1.61–1.44 (4H, m,  $\text{H}^9$  and  $\text{H}^{12}$ ), 1.34–1.21 (4H, m,  $\text{H}^{10}$  and  $\text{H}^{11}$ ;  $m/z$  (ESI HRMS<sup>+</sup>) 891.1328 [ $\text{M} + \text{Na}$ ]<sup>+</sup> ( $[\text{C}_{28}\text{H}_{25}\text{F}_{21}\text{N}_2\text{O}_5\text{Na}]^+$  requires 891.1320);  $t_{\text{R}}$  (*Method A*) = 11.6 min, purity: 97.3% (see below for HPLC method and traces).

**3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,12-henicosafuorododecyl 2-(4-(8-amino-8-oxooctanamido)phenyl)acetate, 6f**



3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,12-Henicosafuorododecyl 2-(4-(8-((benzyloxy)amino)-8-oxooctanamido)phenyl)acetate, **4f**, (30 mg, 0.032 mmol) was dissolved in MeOH (30 mL) and 5 mol% Pd/C (80 mg) was added. The mixture was purged with  $\text{H}_2$  and stirred under an atmosphere of  $\text{H}_2$  at 60 °C for 3 h. The mixture was filtered through Celite, which was rinsed with MeOH (5 mL). The MeOH



fractions were combined and dried under reduced pressure to give the *title compound* as an off-white solid (8 mg, 29%),  $\delta_{\text{H}}$  ( $d_6$ -DMSO) 9.62 (1H, br s, ArNHCO), 7.52 (2H, d,  $J$  7.2 Hz, H<sup>5</sup>), 7.17 (2H, d,  $J$  7.2 Hz, H<sup>4</sup>), 4.36 (2H, t,  $J$  6.0 Hz, H<sup>a</sup>), 4.09 (2H, br s, NH<sub>2</sub>), 3.61 (2H, s, H<sup>2</sup>), 2.72–2.56 (2H, m, H<sup>b</sup>), 2.29 (2H, t,  $J$  7.2 Hz, H<sup>8</sup>), 2.06 (2H, t,  $J$  7.2 Hz, H<sup>13</sup>), 1.65–1.48 (4H, m, H<sup>9</sup> and H<sup>12</sup>), 1.39–1.24 (4H, m, H<sup>10</sup> and H<sup>11</sup>;  $m/z$  (ESI<sup>+</sup>) 853.25 [M + H]<sup>+</sup> ([C<sub>28</sub>H<sub>26</sub>F<sub>21</sub>N<sub>2</sub>O<sub>4</sub>]<sup>+</sup> requires 853.16);  $t_R$  (*Method A*) = 12.8 min, purity: 97.0% (see below for HPLC method and traces).

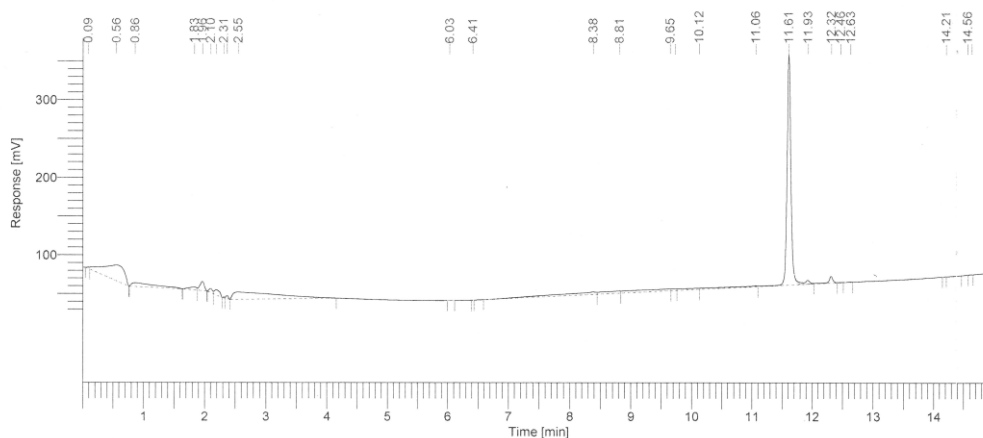
## HPLC Method and Traces

Reverse phase HPLC analysis was performed at 298 K on a Perkin Elmer system, comprising a Perkin Elmer Series 200 pump, Perkin Elmer Series 200 autosampler, Perkin Elmer Series 200 UV/Vis detector and Perkin Elmer Series 200 fluorescence detector. An XBridge C18 100 × 5.00 mm 3.5 μm particle size column was used with a flow rate of 1 mL / min and a run time of 15 min. A solvent system of H<sub>2</sub>O (0.1 % HCOOH) / CH<sub>3</sub>CN (0.1 % HCOOH) (gradient elution) was used, according to Table 1. The UV/Vis and fluorescence detectors were set at 225 nm . Purity determined by integration of all peaks above threshold of 0.1% of major peak.

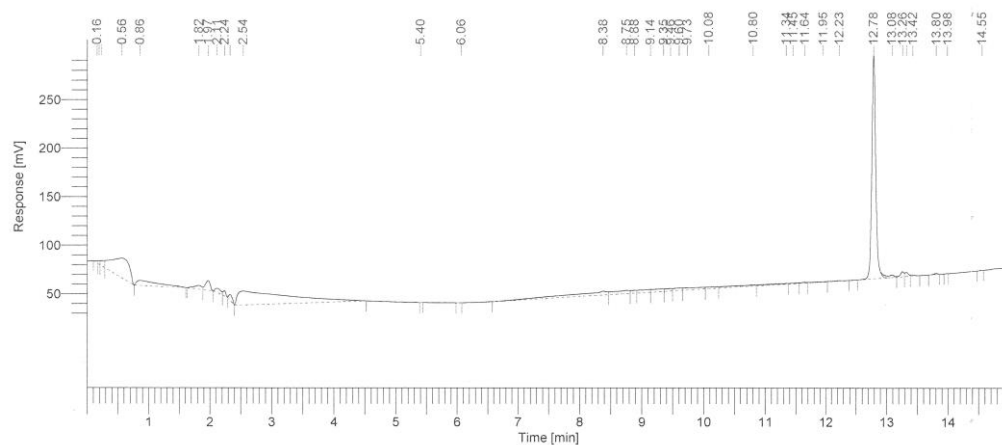
### Method A:

Time (min)	H <sub>2</sub> O + 0.1 % HCO <sub>2</sub> H	CH <sub>3</sub> CN + 0.1 % HCO <sub>2</sub> H
0	95	5
2	95	5
10	0	100
13	0	100
15	95	5

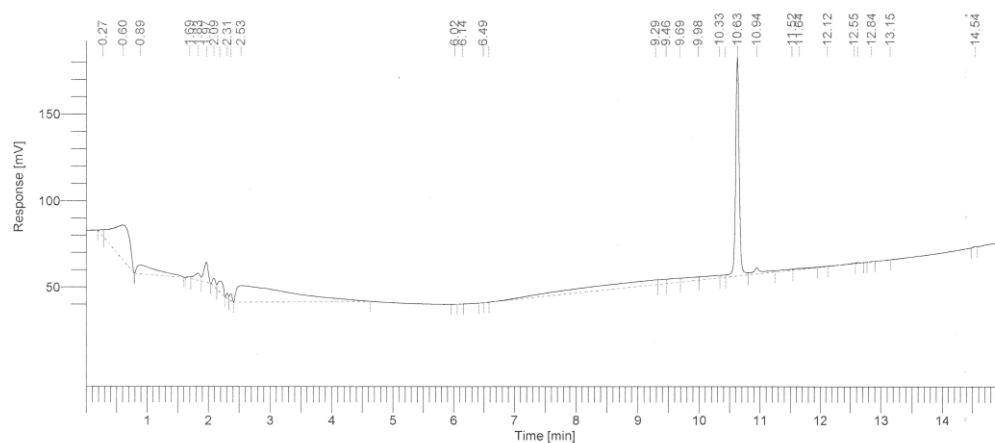
1. Analytical HPLC trace for compound **5f**.  $t_R$  (Method A) = 11.6 min. Purity: 97.3%



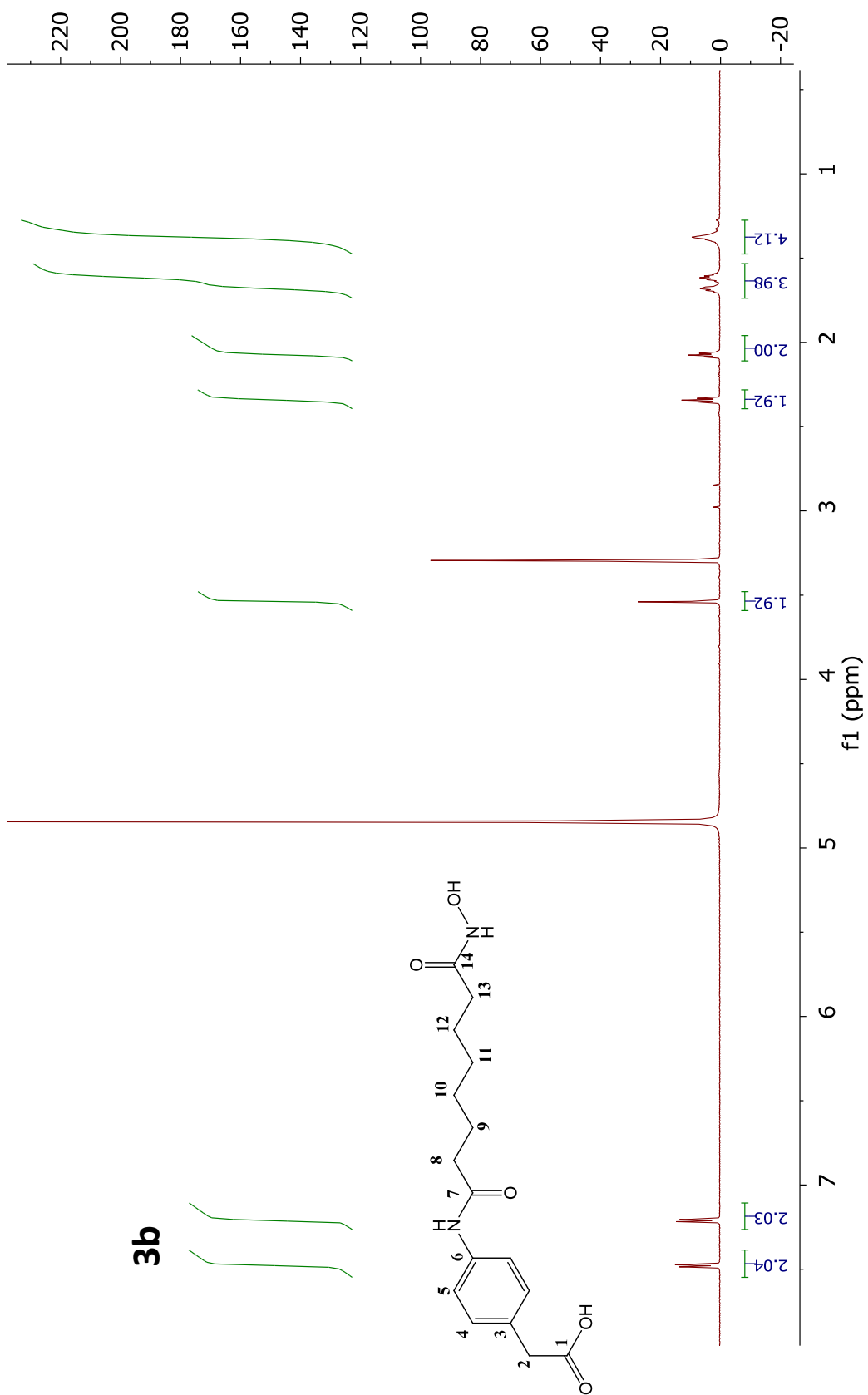
2. Analytical HPLC trace for compound **6f**.  $t_R$  (Method A) = 12.8 min. Purity: 97.0%

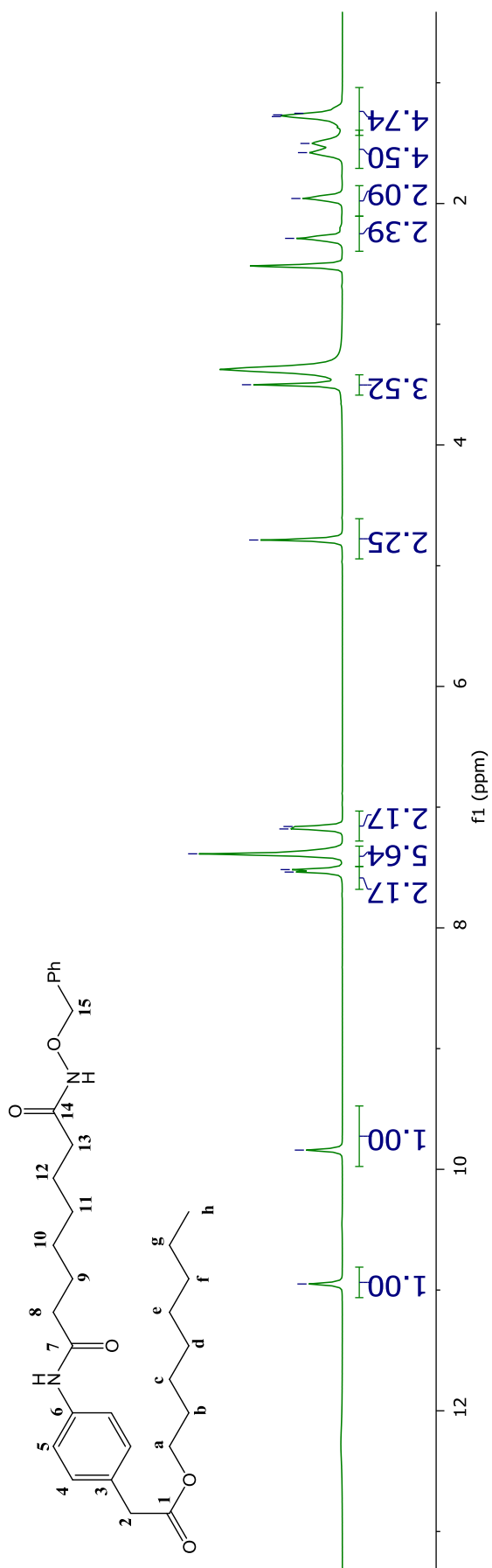


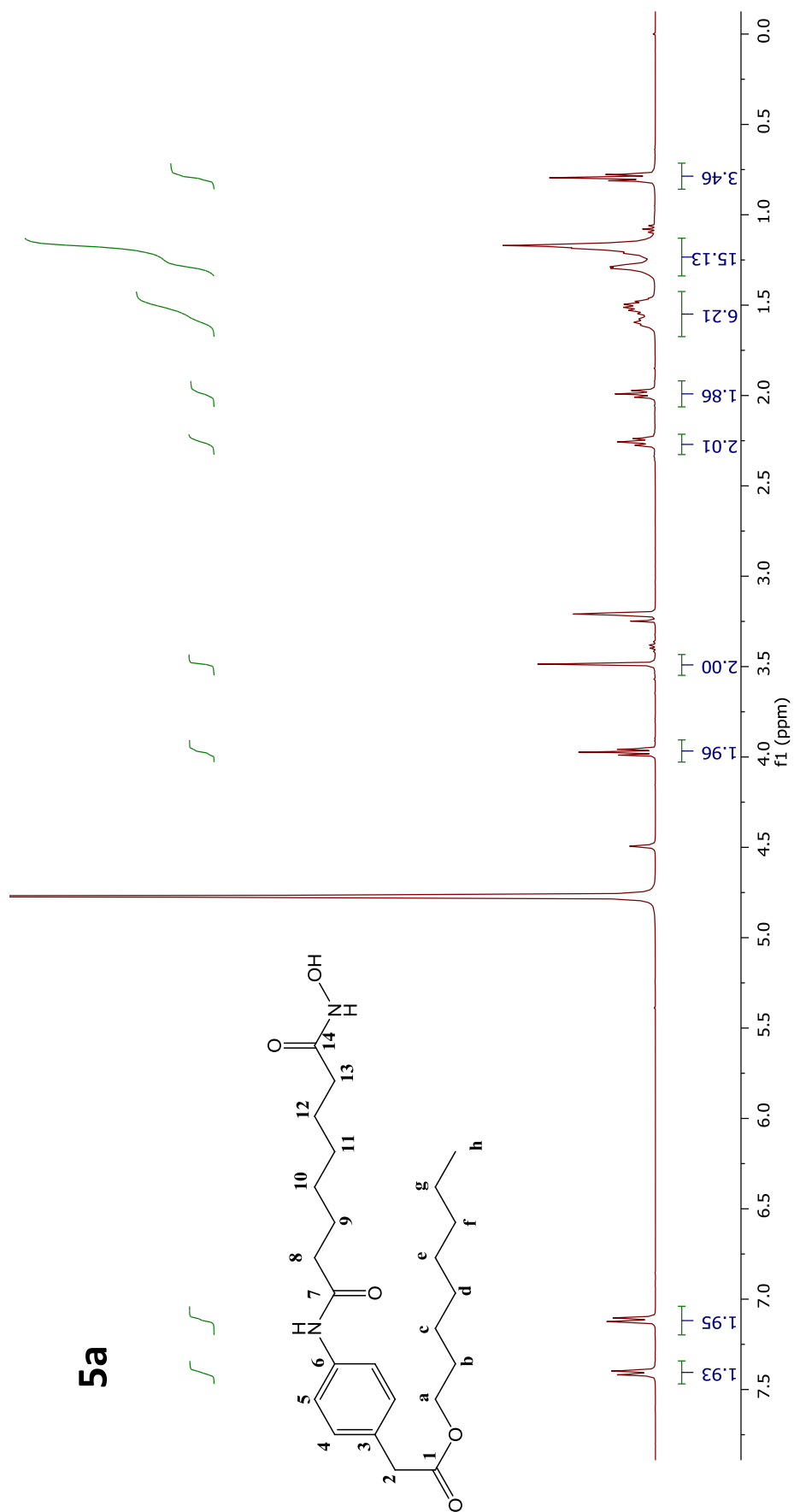
3. Analytical HPLC trace for compound **4a**.  $t_R$  (Method A) = 10.6 min. Purity: 98.1%



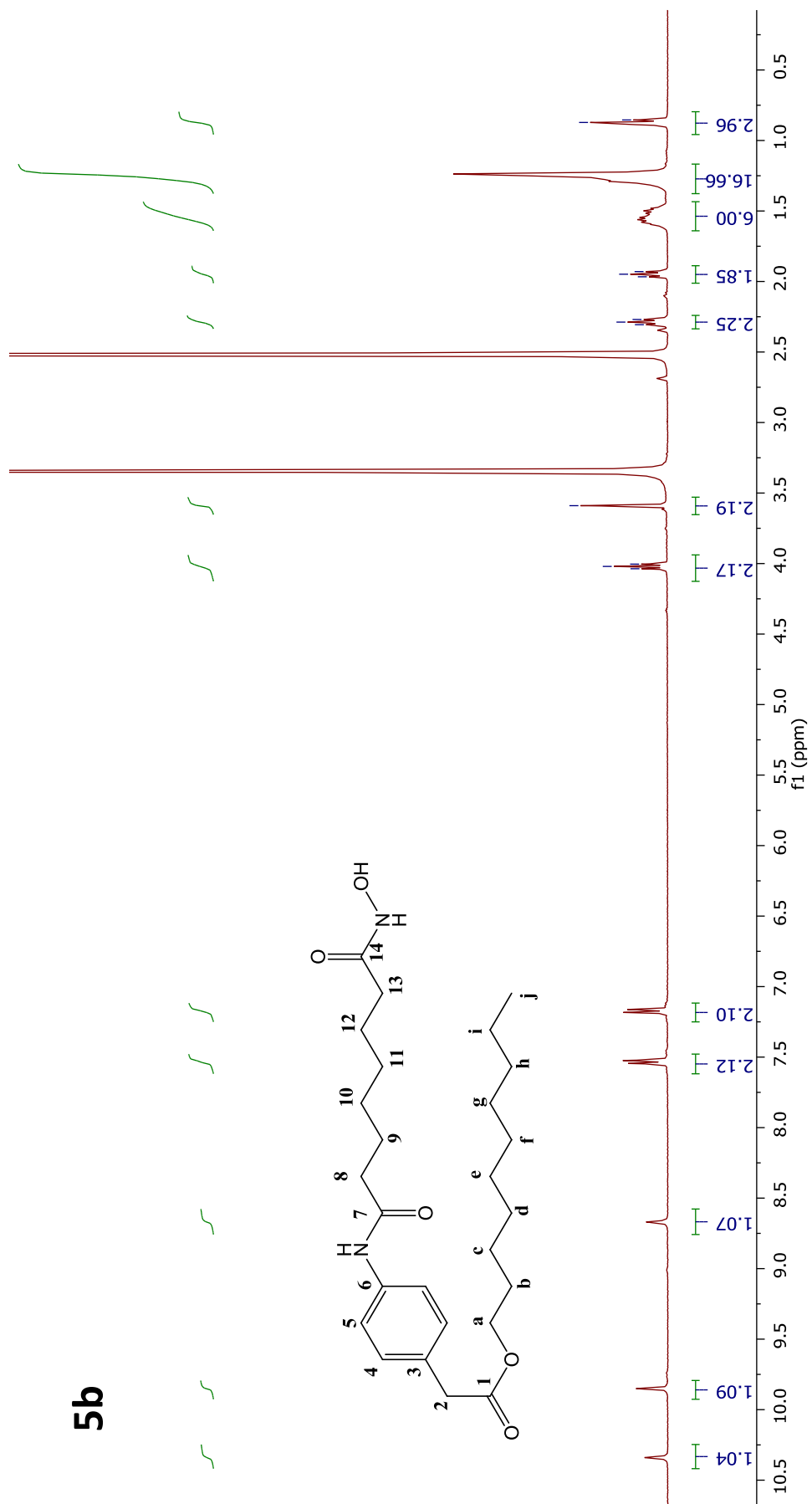
## NMR Spectra



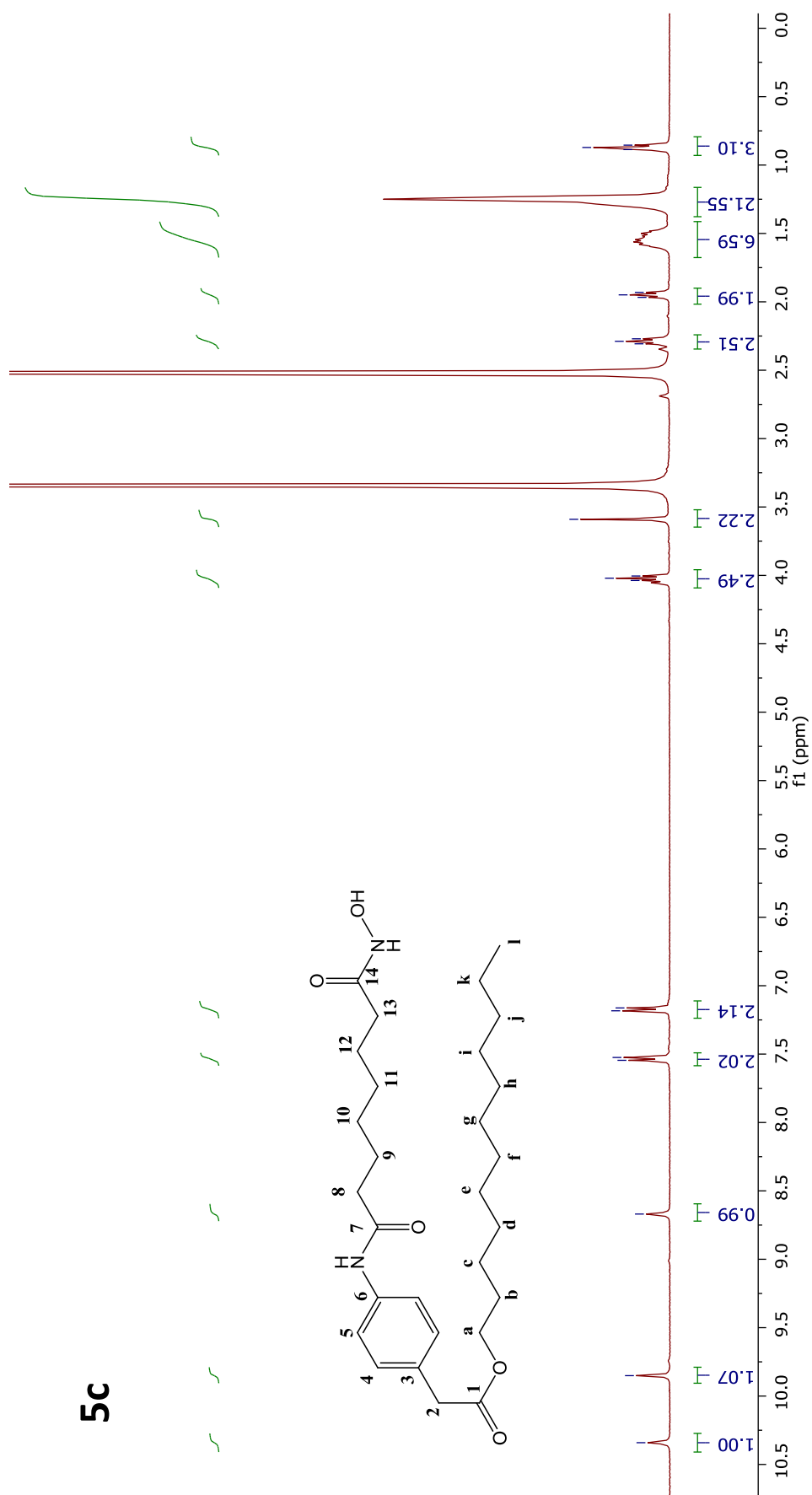
**4a**



5b

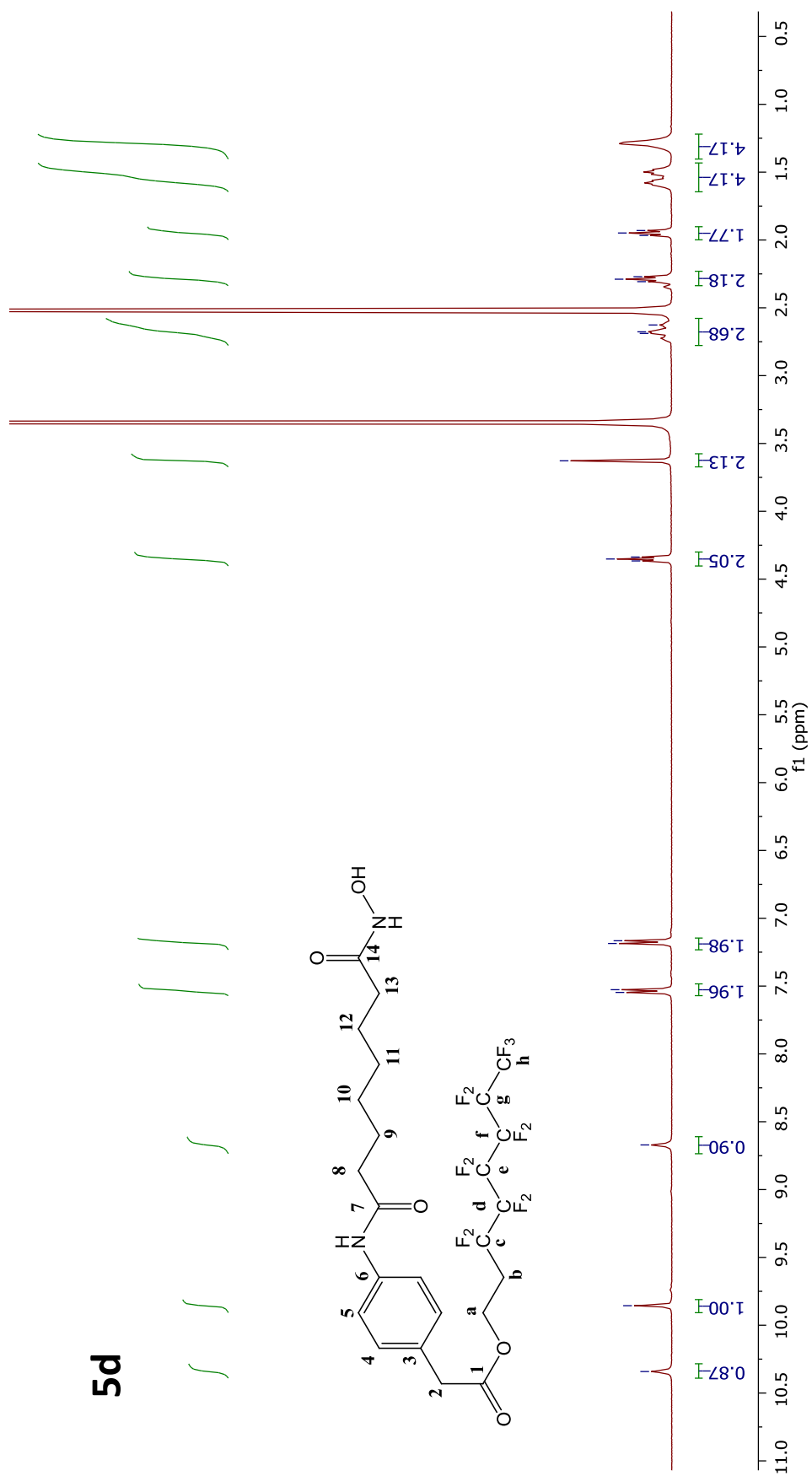


5c

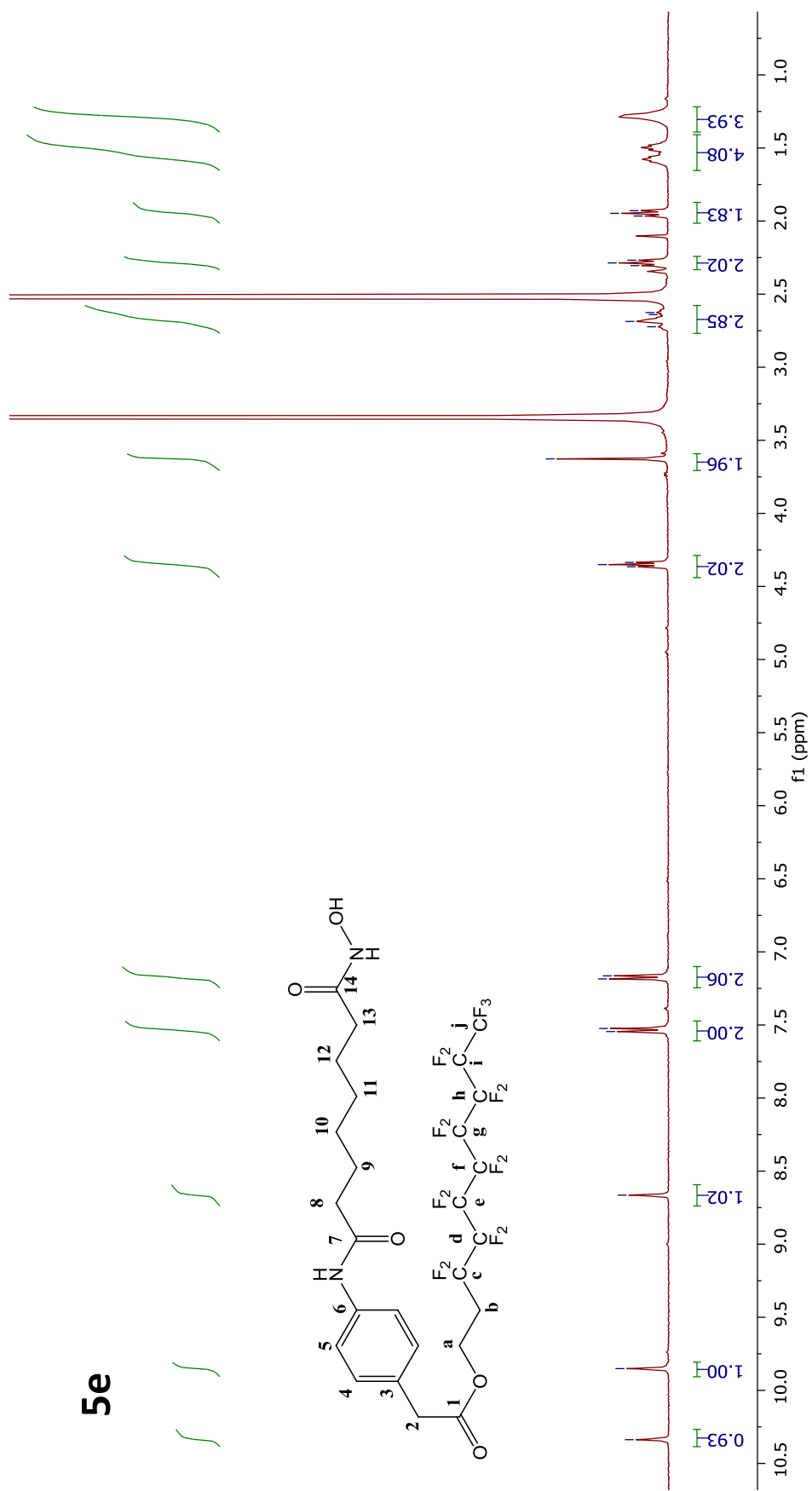




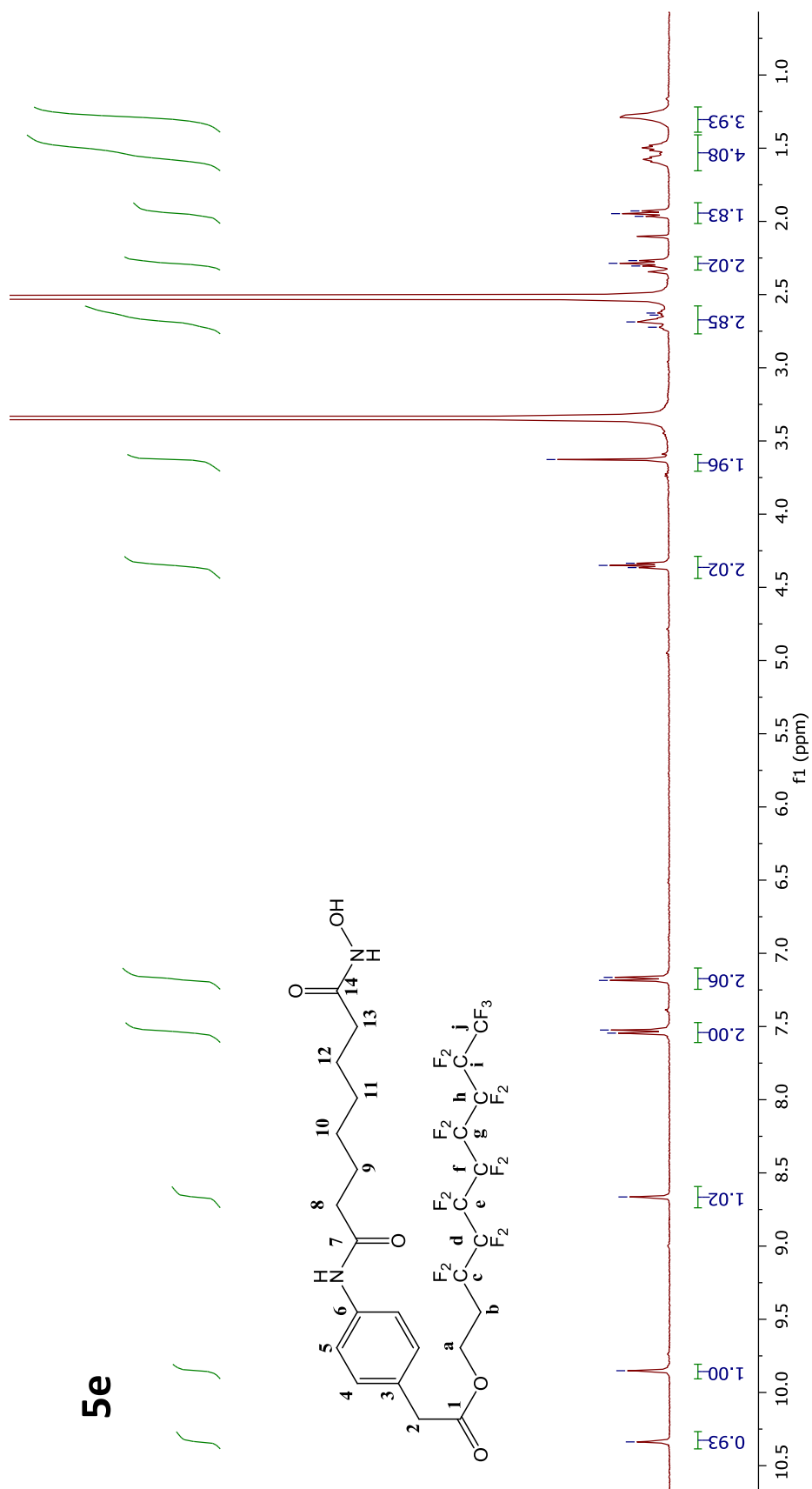
5d

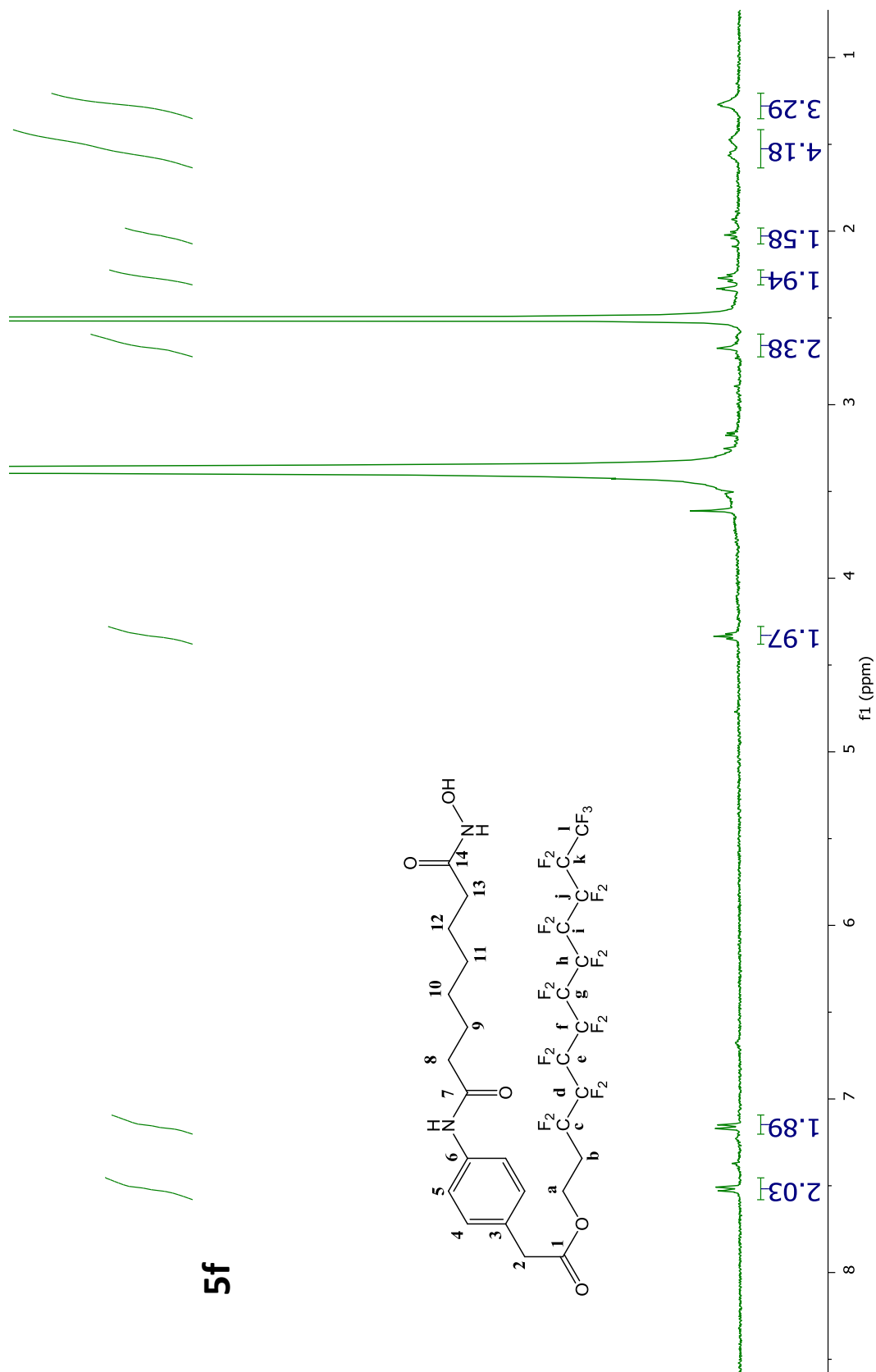


5e



5e



**5f**

## References

- 1 T. Mosmann, *J. Immunol. Methods*, 1983, **65**, 55–63.
- 2 F. Oger, A. Lecorgne, E. Sala, V. Nardese, F. Demay, S. Chevance, D. C. Desravines, N. Aleksandrova, R. Le Guével, S. Lorenzi, A. R. Beccari, P. Barath, D. J. Hart, A. Bondon, D. Carettoni, G. Simonneaux and G. Salbert, *J. Med. Chem.*, 2010, **53**, 1937–1950.