Electronic Supplementary Material (ESI) for Organic & Biomolecular Chemistry.

Supporting Information

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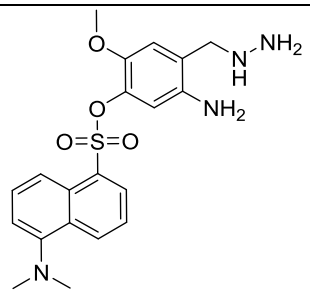
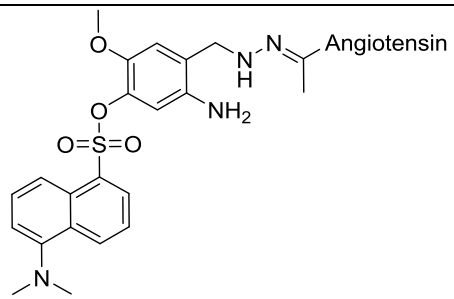
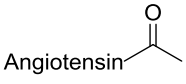
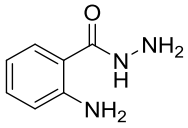
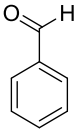
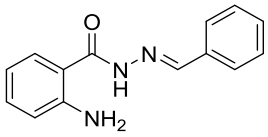
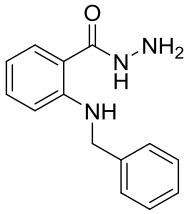
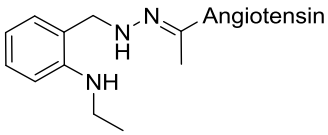
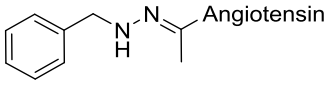
1. General information:

All the reactions were performed in oven-dried glassware. Chemical reagents were purchased either from Sigma-Aldrich, Alfa Aesar or Avra Synthesis and were used without any further purification. Angiotensin I and horse heart myoglobin were purchased from Sigma-Aldrich. Dry DMF was purchased from Acros Organics. Distilled water was used for reaction work ups and ultrapure Type 1 water was used for buffer preparations. The progress of reactions was monitored by thin layer chromatography (TLC) using 0.25 mm Merck precoated (60 F254) silica gel plates. For visualization of TLC spots, UV light of wavelength 254 nm (for UV active compounds) and 365 nm (for fluorescent compounds) was used. In some cases TLC spots were visualized using vanillin, ninhydrin and phosphomolybdic acid (PMA) staining solutions.

Purifications were performed using column chromatography on silica gel (100-200 mesh). ^{1}H and ^{13}C NMR spectra were recorded on Bruker-400 or Jeol-400 spectrometers. Chemical shifts are reported as parts per million (δ) relative to tetramethylsilane (TMS) as internal standard and coupling constants (*J* values) in Hertz (Hz). Multiplicities are indicated as follows: s (singlet), d (doublet), t (triplet), q (quartet), dd (doublet of doublet), bs (broad singlet) and m (multiplet). High resolution mass spectra (HRMS) were recorded on HRMS-ESI-Q-Time of Flight LC/MS (Synapt G2, Waters). UV analyses were performed on Cary 300 UV-Vis spectrophotometer using quartz cuvettes. MALDI-TOF mass spectrometry was performed on 4800 Plus MALDI TOF/TOF instrument from AB Sciex by using a mixture of 2,6-dihydroxyacetophenone (65.7 mM) and diammonium hydrogen citrate (56.9 mM) in 43% ethanol in water as matrix for myoglobin, and α-cyano-4-hydroxycinnamic acid (52.9 mM) in 50:50 water/acetonitrile with 0.1% TFA as a matrix for angiotensin. HPLC analyses were performed on an Agilent 1260 infinity LC instrument on a C18 analytical column (Zorbax ODS 4.6 × 250 mm, 5 µm). The flow rate was 1 mL/min and UV absorbance was monitored at 215 nm. Lipidomics was performed on a triple quadrupole mass spectrometer (QTRAP® 4500 system from AB SCIEX).

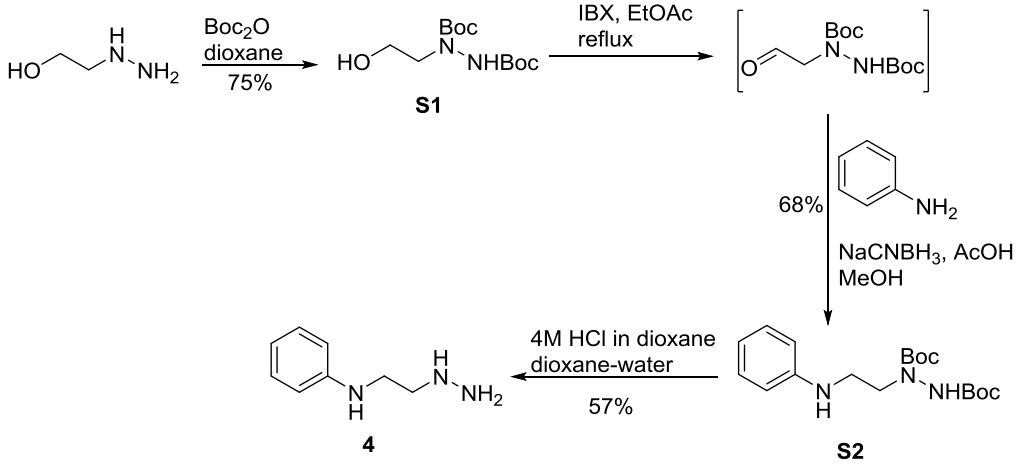
2. Table S1. Masses (MALDI) of peaks isolated from HPLC

S. No.	Structure	Mass obtained	Expected mass	HPLC Chromatogram
1		455.1	455.1 $[M+K]^+$	Fig. 3a (top panel)
		439.2	439.1 $[M+Na]^+$	Fig. 3b (top panel)
2		1650.5	1650.9 $[M+H]^+$	Fig. 3a (top panel)
		1649.8	1650.9 $[M+H]^+$	Fig. 3b (bottom panel)
3		1251.7	1252.5 $[M+H]^+$	Fig. 3b (top panel)
		1251.8	1252.5 $[M+H]^+$	Fig. S5 (2nd panel)
		1251.7	1252.5 $[M+H]^+$	Fig. S6-a (first panel)
		1251.7	1252.5 $[M+H]^+$	Fig. S6-b (first panel)

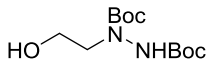
		1252.1	1252.5 $[M+H]^+$	Fig. S7-b (top panel)
4		174.0	174.1 $[M+Na]^+$	Fig. S3 (4th panel)
		174.0	174.1 $[M+Na]^+$	Fig. S3 (bottom panel)
5		129.8	129.0 $[M+Na]^+$	Fig. S3 (4th panel)
6		262.1	262.1 $[M+Na]^+$	Fig. S3 (4th panel)
7		264.1	264.1 $[M+Na]^+$	Fig. S3 (bottom panel)
8	Angiotensin	1296.8	1297.5 $[M+H]^+$	Fig. S5 (2nd panel)
9		1399.1	1399.5 $[M+H]^+$	Fig. S6-a (first panel)
		1398.9	1399.5 $[M+H]^+$	Fig. S7-a (bottom panel)
		1398.9	1399.5 $[M+H]^+$	Fig. S7-a (top panel)
10		1356.1	1356.5 $[M+H]^+$	Fig. S6-b (first panel)
		1355.8	1356.5 $[M+H]^+$	Fig. S7-b (bottom panel)
		1355.8	1356.5 $[M+H]^+$	Fig. S7-b (top panel)

3. Synthetic schemes and procedures:

Scheme S1 Synthesis of *N*-(2-hydrazinylethyl)aniline (**4**)



di-*t*-butyl 1-(2-hydroxyethyl)hydrazine-1,2-dicarboxylate



S1

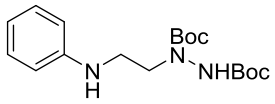
To a stirred solution of 2-hydrazinoethanol (1.0 g, 13.14 mmol) in dioxane (23 mL), di-*t*-butyl dicarbonate (6.64 mL, 28.91 mmol) was carefully added at 0°C. The mixture was warmed to room temperature and stirred for 18 h. The reaction mixture was concentrated on a rotary evaporator to obtain crude product which was purified by silica gel column chromatography (40% ethyl acetate in hexane) to give **S1** as a white solid (2.71 g, 75%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 6.48 (s, 1H), 4.21 – 3.77 (m, 1H), 3.69 (bs, 2H), 3.56 (bs, 2H), 1.53 – 1.34 (m, 18H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 157.4, 155.3, 82.3, 81.7, 59.6, 52.9, 28.3, 28.3.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{12}H_{24}N_2O_5Na$, 299.1583; found, 299.1584.

di-*t*-butyl 1-(2-(phenylamino)ethyl)hydrazine-1,2-dicarboxylate



S2

To a solution of **S1** (1.0 g, 3.62 mmol) in ethyl acetate, IBX (3.04 g, 10.86 mmol) was added. After refluxing the mixture for 3 h, it was filtered through a celite pad and the filtrate was concentrated on a rotary evaporator to obtain crude aldehyde which was used in the next step without further purification. A solution of this crude aldehyde (0.2 g, 0.73 mmol) in dry methanol (4 mL) was added to a suspension of aniline (0.067 mL, 0.73 mmol), acetic acid (0.042 mL, 0.73 mmol) and 4 Å molecular sieves (0.2 g) in dry methanol (4 mL) under N_2 atmosphere. After stirring the reaction mixture at room temperature for 1 h, sodium cyanoborohydride (0.046 g, 0.73 mmol) was added and the resulting mixture was stirred at room temperature overnight. The reaction mixture was filtered through a celite pad and the filtrate was concentrated on a rotary evaporator. The resulting residue was dissolved in ethyl acetate (30 mL) and water (20 mL) was added. After separating the organic and aqueous layers, the aqueous layer was

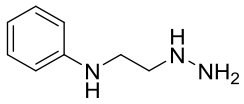
washed with ethyl acetate (2 × 30 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated under reduced pressure and the residue was purified by silica gel column chromatography (8% ethyl acetate in hexane) to obtain **S2** as pale yellow oil (0.17 g, 68%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.19 – 7.12 (m, 2H), 6.68 (t, *J* = 7.3 Hz, 1H), 6.62 (d, *J* = 7.9 Hz, 2H), 6.47 – 6.08 (m, 1H), 4.84 – 3.89 (m, 1H), 3.66 (bs, 2H), 3.34 (t, *J* = 5.6 Hz, 2H), 1.56 – 1.25 (m, 18H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 155.5, 148.1, 129.4, 117.3, 113.0, 81.7, 49.5, 41.6, 28.3, 28.2.

HRMS (ESI) $[M+H]^+$ calculated for $C_{18}H_{30}N_3O_4$, 352.2236; found, 352.2238.

***N*-(2-hydrazinylethyl)aniline (4)**



4

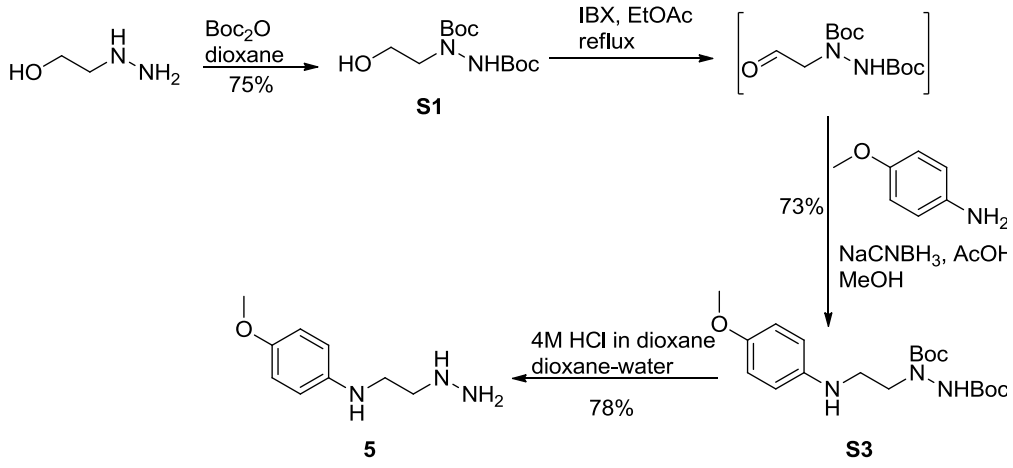
To solution of **S2** (0.059 g, 0.17 mmol) in 1,4-dioxane/water (0.50 mL/0.05 mL), 4 M HCl in dioxane (1.00 mL) was added. After stirring this mixture for 5 h, diethyl ether was added to it and the resulting solution was filtered to obtain **1** as a pale yellow solid (0.025 g, 57%).

^{1}H NMR (400 MHz, D_2O) δ 7.62 – 7.55 (m, 3H), 7.53 – 7.47 (m, 2H), 3.67 (t, *J* = 5.6 Hz, 2H), 3.30 (t, *J* = 5.6 Hz, 2H).

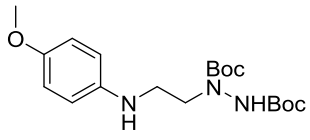
^{13}C NMR (100 MHz, DMSO-D_6) δ 137.1, 130.0, 128.1, 122.3, 47.2, 45.2.

HRMS (ESI) $[M+H]^+$ calculated for $C_8H_{14}N_3$, 152.1188; found, 152.1194.

Scheme S2 Synthesis of N-(2-hydrazinylethyl)-4-methoxyaniline (**5**)



di-*t*-butyl 1-(2-((4-methoxyphenyl)amino)ethyl)hydrazine-1,2-dicarboxylate



S3

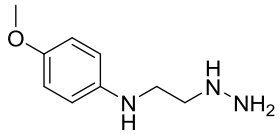
To a solution of **S1** (1.0 g, 3.62 mmol) in ethyl acetate, IBX (3.04 g, 10.86 mmol) was added. After refluxing the mixture for 3 h, it was filtered through a celite pad and the filtrate was concentrated on a rotary evaporator to obtain crude aldehyde which was used in the next step without further purification. A solution of this crude aldehyde (0.2 g, 0.73 mmol) in dry methanol (4 mL) was added to a suspension of 4-methoxy aniline (0.09 g, 0.73 mmol), acetic acid (0.042 mL, 0.73 mmol) and 4 Å molecular sieves (0.2 g) in dry methanol (4 mL) under N_2 atmosphere. After stirring the reaction mixture at room temperature for 1 h, sodium cyanoborohydride (0.046 g, 0.73 mmol) was added to it and stirred at room temperature overnight. The reaction mixture was filtered through a celite pad and the filtrate was concentrated on a rotary evaporator. The resulting residue was dissolved in ethyl acetate (30 mL) and water (20 mL) was added. After separating the organic and aqueous layers, the aqueous layer was washed with ethyl acetate (2 × 30 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated under reduced pressure and the residue was purified by silica gel column chromatography (8% ethyl acetate in hexane) to obtain **S3** as yellow oil (0.2 g, 73%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 6.77 (d, *J* = 9.2 Hz, 2H), 6.60 (d, *J* = 8.4 Hz, 2H), 6.48 – 6.09 (m, 1H), 3.74 (s, 3H), 3.64 (s, 2H), 3.29 (t, *J* = 5.6 Hz, 2H), 1.55 – 1.32 (m, 18H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 155.5, 152.3, 142.2, 115.1, 114.6, 81.7, 56.0, 49.5, 42.7, 28.3, 28.2.

HRMS (ESI) $[M+H]^+$ calculated for $C_{19}H_{32}N_3O_5$, 382.2342; found, 382.2340.

***N*-(2-hydrazinylethyl)-4-methoxyaniline (5)**



5

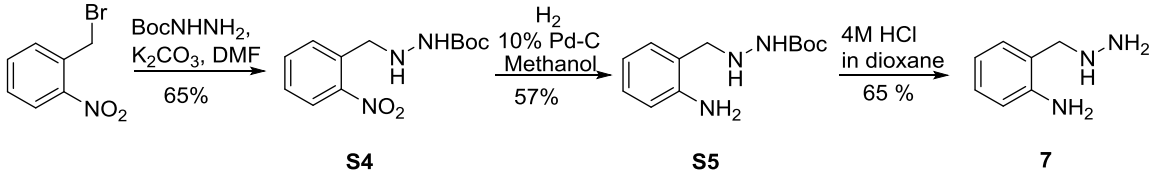
To solution of **S3** (0.15 g, 0.39 mmol) in 1,4-dioxane/water (2.00 mL/0.2 mL), 4 M HCl in dioxane (4.00 mL) was added. After stirring this mixture for 5 h, diethyl ether was added to it and the resulting solution was filtered to obtain **5** as a pale brown solid (0.025 g, 57%).

^{1}H NMR (400 MHz, D_2O) δ 7.40 (d, *J* = 9.1 Hz, 2H), 7.09 (d, *J* = 9.1 Hz, 2H), 3.82 (s, 3H), 3.58 (t, *J* = 5.6, 2H), 3.24 (t, *J* = 5.6 Hz, 2H).

^{13}C NMR (100 MHz, DMSO-D_6) δ 159.0, 128.8, 124.0, 115.0, 55.6, 47.8, 44.9.

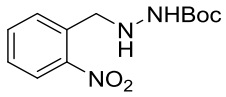
HRMS (ESI) $[M+H]^+$ calculated for $C_9H_{16}N_3O$, 182.1293; found, 182.1295.

Scheme S3. Synthesis of 2-(hydrazinylmethyl) aniline **(7)**



S4 **S5** **7**

***t*-butyl 2-(2-nitrobenzyl)hydrazine-1-carboxylate**



S4

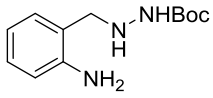
According to the procedure of Bare *et al.*,[1] to a solution of 2-nitrobenzyl bromide (1.0 g, 4.62 mmol) in DMF (10 mL) were added *t*-butyl carbazate (4.7 g, 35.50 mmol) and K_2CO_3 (0.7 g, 5.03 mmol). The mixture was heated to 90°C and stirred for 2 h under N_2 atmosphere. Subsequently, the mixture was cooled, diluted with diethyl ether (40 mL), and washed with H_2O (5 × 20 mL) and brine (1 × 10 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to obtain crude product which was purified by silica gel column chromatography (10% ethyl acetate in hexane) to give **S4** as a yellow oil (0.81 g, 65%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.94 (d, *J* = 8.1 Hz, 1H), 7.60 – 7.48 (m, 2H), 7.48 – 7.37 (m, 1H), 6.12 (bs, 1H), 4.53 (bs, 1H), 4.26 (s, 2H), 1.38 (s, 9H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 156.7, 149.6, 133.3, 133.1, 131.9, 128.5, 124.9, 80.8, 53.4, 28.4.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{12}H_{17}N_3O_4Na$, 290.1117; found, 290.1119.

***t*-butyl 2-(2-aminobenzyl)hydrazine-1-carboxylate**



S5

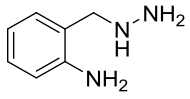
To solution of **S4** (0.75 g, 2.81 mmol) in methanol (10 mL) was added 10% Pd-C (0.15 g). The resulting heterogeneous mixture was then stirred for 2 h under hydrogen atmosphere. The reaction mixture was then filtered through a celite pad and the filtrate was concentrated on a rotary evaporator to obtain a yellowish oil. The residual yellow oil was then purified by silica gel column chromatography (20% ethyl acetate in hexane) to give **S5** as a colorless oil (0.38 g, 57 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.11 (t, *J* = 7.6 Hz, 1H), 7.06 (d, *J* = 7.4 Hz, 1H), 6.72 – 6.64 (m, 2H), 6.06 (bs, 1H), 4.48 (bs, 3H), 4.00 (s, 2H), 1.47 (s, 9H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 156.9, 146.7, 131.3, 129.2, 121.0, 118.0, 115.8, 80.7, 54.1, 28.4.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{12}H_{19}N_3O_2Na$, 260.1375; found, 260.1376.

2-(hydrazinylmethyl) aniline



7

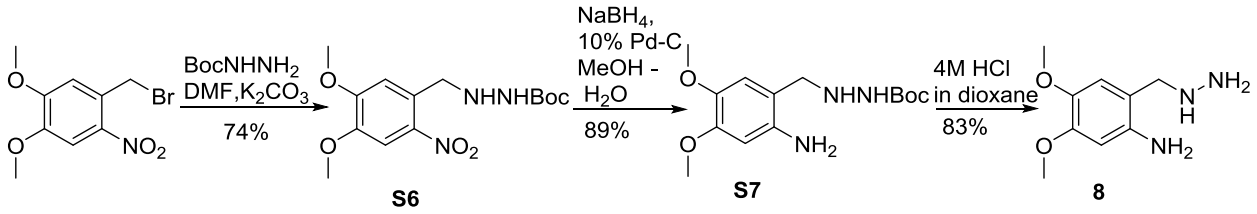
To solution of **S5** (0.33 g, 1.40 mmol) in 1,4-dioxane/water (4.3 mL/0.4 mL), 4 M HCl in dioxane (4.3 mL) was added. After stirring this mixture for 5 h, diethyl ether was added to it and the resulting solution was filtered to obtain **7** as a pale yellow solid (0.35 g, 65%).

^{1}H NMR (400 MHz, CD_3OD) δ 7.62 – 7.46 (m, 4H), 4.35 (s, 2H).

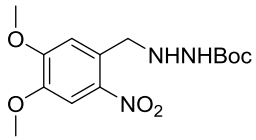
^{13}C NMR (100 MHz, CD_3OD) δ 133.2, 131.7, 131.5, 130.8, 130.1, 125.4, 51.3.

HRMS (ESI) $[M+H]^+$ calculated for $C_7H_{12}N_3$, 138.1031; found, 138.1029.

Scheme S4. Synthesis of 2-(hydrazinylmethyl)-4,5-dimethoxyaniline **(8)**



***t*-butyl 2-(4,5-dimethoxy-2-nitrobenzyl)hydrazine-1-carboxylate**



S6

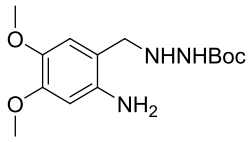
According to the procedure of Bare *et al.*,[1] to a solution of 1-(bromomethyl)-4, 5-dimethoxy-2-nitrobenzene (0.45 g, 1.63 mmol) in DMF (3 mL) were added *t*-butyl carbazate (1.65 g, 12.50 mmol) and K_2CO_3 (0.25 g, 1.79 mmol). The mixture was heated to 90°C and stirred for 2 h under N_2 atmosphere. Subsequently, the mixture was cooled, diluted with diethyl ether (20 mL), and washed with H_2O (5 × 10 mL) and brine (1 × 5 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to get crude product which was then purified by silica gel column chromatography (5% ethyl acetate in dichloromethane) to give **S6** as a yellow solid (0.53 g, 74%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.62 (s, 1H), 7.09 (s, 1H), 6.02 (bs, 1H), 4.54 (bs, 1H), 4.27 (s, 2H), 3.98 (s, 3H), 3.93 (s, 3H), 1.40 (s, 9H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 156.6, 153.2, 148.1, 141.5, 128.6, 113.0, 108.4, 80.8, 56.6, 56.5, 53.8, 28.4.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{14}H_{21}N_3O_6Na$, 350.1328; found, 350.1334.

***t*-butyl 2-(2-amino-4,5-dimethoxybenzyl)hydrazine-1-carboxylate**



S7

According to the procedure of Basu *et al.*,[2] to a 10 mL round-bottomed flask charged with 10% Pd-C (0.005 g) was added a solution of $NaBH_4$ (0.023 g, 0.61 mmol in 2 mL water). Gas evolution was observed. The reaction mixture was slowly heated to 50°C. To this solution, a methanolic solution (1 mL) of **S6** (0.1 g, 0.31 mmol) was then added dropwise with constant stirring. Cessation of gas evolution indicated completion of reduction within 20 minutes. After completion of the reaction, the reaction mixture was filtered through a celite pad and the filtrate

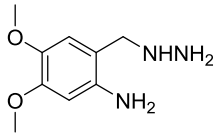
was concentrated on a rotary evaporator. The resulting residue was dissolved in ethyl acetate (20 mL) and water (10 mL) was added. After separating the organic and aqueous layers, the aqueous layer was washed with ethyl acetate (2 × 10 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered and concentrated on a rotary evaporator to obtain **S7** as brown solid (0.081 g, 89%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 6.63 (s, 1H), 6.27 (s, 1H), 6.12 (bs, 1H), 4.12 (bs, 3H), 3.92 (s, 2H), 3.82 (s, 3H), 3.78 (s, 3H), 1.46 (s, 9H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 157.0, 149.9, 141.4, 140.7, 115.7, 112.3, 101.0, 80.8, 56.8, 56.0, 53.7, 28.5.

HRMS (ESI) $[M+H]^+$ calculated for $C_{14}H_{24}N_3O_4$, 298.1767; found, 298.1767.

2-(hydrazinylmethyl)-4,5-dimethoxyaniline



8

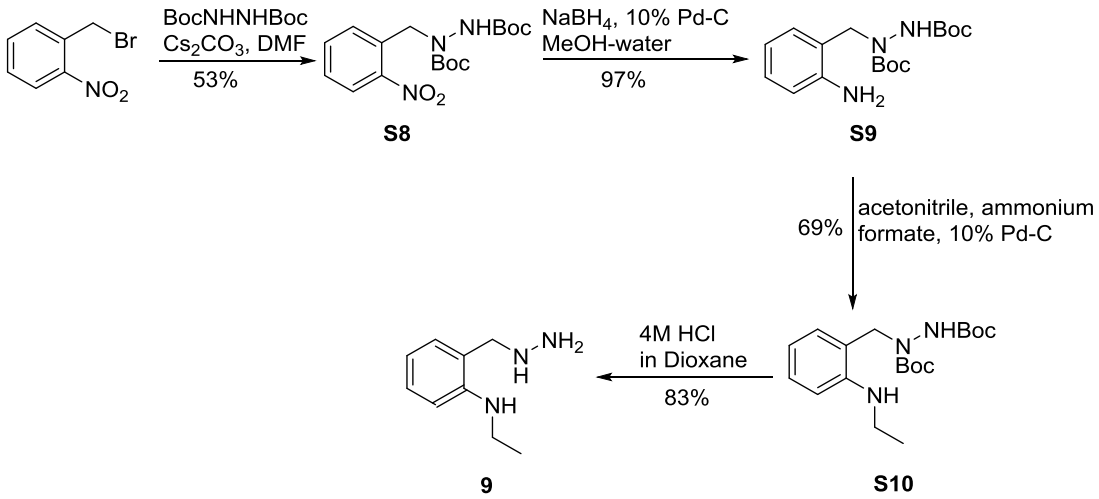
To solution of **S7** (0.05 g, 0.17 mmol) in 1,4-dioxane/water (0.5 mL/0.05 mL), 4 M HCl in dioxane (0.5 mL) was added. After stirring this mixture for 5 h, diethyl ether was added to it and the resulting solution was filtered to obtain **8** as a dark brown solid (0.05 g, 83%).

^{1}H NMR (400 MHz, CD_3OD) δ 7.13 (s, 1H), 7.02 (s, 1H), 4.25 (s, 2H), 3.89 (s, 3H), 3.88 (s, 3H).

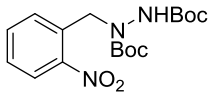
^{13}C NMR (100 MHz, CD_3OD) δ 151.6, 150.9, 123.9, 121.6, 115.8, 108.9, 56.8, 50.9.

HRMS (ESI) $[M+H]^+$ calculated for $C_9H_{16}N_3O_2$, 198.1243; found, 198.1232.

Scheme S5. Synthesis of *N*-ethyl-2-(hydrazinylmethyl)aniline **(9)**



di-*t*-butyl 1-(2-nitrobenzyl)hydrazine-1,2-dicarboxylate



S8

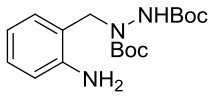
To a stirred solution of di-*t*-butyl hydrazodiformate (BocNHNHBoc) *(*2.15 g, 9.26 mmol) and Cs_2CO_3 (1.66 g, 5.09 mmol) in dry DMF (5 mL) at 80°C was added a solution of 2-nitrobenzyl bromide (1.0 g, 4.63 mmol) in dry DMF (5 mL). After stirring for 45 min at 80°C under N_2 atmosphere, the mixture was cooled, diluted with diethyl ether (30 mL), and washed with H_2O (5 × 20 mL) and brine (1 × 10 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to obtain crude product which was purified by silica gel column chromatography (4% ethyl acetate in DCM) to give **S8** as a sticky pale yellow solid (0.9 g, 53%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.97 (d, *J* = 8.0 Hz, 1H), 7.71 – 7.33 (m, 3H), 6.58 – 6.16 (m, 1H), 5.00 (s, 2H), 1.43 (s, 18H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 155.5, 154.9, 148.7, 133.4, 132.9, 130.5, 128.3, 124.9, 82.0, 81.5, 51.6, 28.2, 28.2.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{17}H_{25}N_3O_6Na$, 390.1641; found, 390.1645.

di-*t*-butyl 1-(2-aminobenzyl)hydrazine-1,2-dicarboxylate



S9

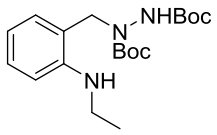
To a 10 mL round-bottomed flask charged with 10% Pd-C (0.067 g) was added a solution of $NaBH_4$ (0.18 g, 4.85 mmol in 18 mL water). Gas evolution was observed. Reaction mixture was slowly heated to 50°C and to this solution, a methanolic solution (9 mL) of **S8** (0.89 g, 2.42 mmol) was then added dropwise with constant stirring. Cessation of gas evolution indicated completion of the reduction within 20 minutes. After completion of the reaction, the reaction mixture was filtered through a celite pad and the filtrate was concentrated on a rotary evaporator. The resulting residue was dissolved in ethyl acetate (30 mL) and water (20 mL) was added. After separating the organic and aqueous layers, the aqueous layer was washed with ethyl acetate (2 × 30 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to obtain **S9** as pale yellow sticky oil (0.79 g, 97%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.10 (td, *J* = 7.6, 1.6 Hz, 1H), 7.04 (dd, *J* = 7.6, 1.6 Hz, 1H), 6.71 – 6.59 (m, 2H), 6.32 – 5.93 (m, 1H), 4.81 – 4.14 (m, 4H), 1.58 – 1.25 (m,18H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 155.8, 154.8, 146.0, 132.0, 129.4, 120.3, 117.6, 115.6, 81.8, 81.3, 50.5, 28.3, 28.1.

HRMS (ESI) $[M+H]^+$ calculated for $C_{17}H_{28}N_3O_4$, 338.2080; found, 338.2084.

di-*t*-butyl 1-(2-(ethylamino)benzyl)hydrazine-1,2-dicarboxylate



S10

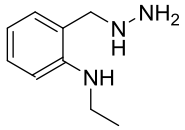
To a stirred solution of **S9** (0.01 g, 0.30 mmol) in acetonitrile (1.1 mL) was added a suspension of 10% Pd-C (0.005 g) and ammonium formate (0.17 g, 2.67 mmol) in water (0.28 mL). After stirring at room temperature for 18 h, the reaction mixture was filtered through a celite pad and the filtrate was concentrated on a rotary evaporator to obtain crude product which was purified by silica gel column chromatography (12% ethyl acetate in hexane) to give **S10** as colorless oil.

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.20 (t, *J* = 7.2 Hz, 1H), 7.03 (d, *J* = 6.8 Hz, 1H), 6.71 – 6.49 (m, 2H), 6.23 – 5.83 (m, 1H), 5.23 – 4.15 (m, 3H), 3.13 (q, *J* = 6.8 Hz, 2H), 1.59 – 1.20 (m, 21H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 156.0, 154.7, 147.3, 131.9, 129.7, 119.7, 115.6, 110.0, 81.8, 81.3, 50.5, 38.3, 28.3, 28.0, 14.6.

HRMS (ESI) $[M+H]^+$ calculated for $C_{19}H_{32}N_3O_4$, 366.2393; found, 366.2397.

***N*-ethyl-2-(hydrazinylmethyl)aniline (9)**



9

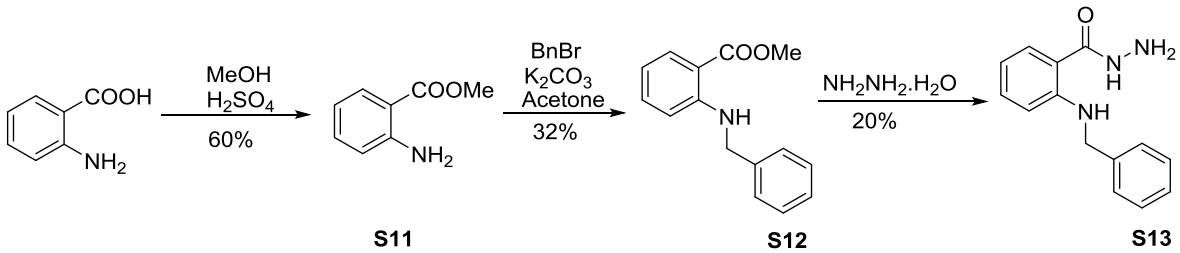
To a solution of **S10** (0.064 g, 0.18 mmol) in 1,4-dioxane/water (0.98 mL/0.098 mL) 4 M HCl in dioxane (1.96 mL) was added. After stirring this mixture for 5 h, diethyl ether was added to it and the resulting solution was filtered to obtain **9** as a pale yellow solid (0.048 g, 83%).

^{1}H NMR (400 MHz, D_2O) δ 7.66 – 7.44 (m, 4H), 4.40 (s, 2H), 3.56 (q, *J* = 7.3 Hz, 2H), 1.35 (t, *J* = 7.3 Hz, 3H).

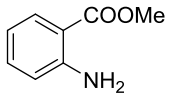
^{13}C NMR (100 MHz, DMSO-D_6) δ 136.0, 132.4, 129.8, 128.5, 128.2, 123.2, 48.9, 46.3, 11.4.

HRMS (ESI) $[M+H]^+$ calculated for $C_9H_{16}N_3$, 166.1344; found, 166.1346.

Scheme S6. Synthesis of 2-(benzylamino)benzohydrazide **(S13)**



methyl 2-aminobenzoate



S11

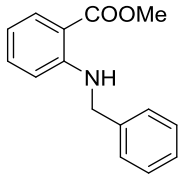
To a solution of 2-aminobenzoic acid (3.0 g, 21.87 mmol) in MeOH (50 mL) was added conc. H_2SO_4 (1.5 mL). The reaction mixture was refluxed for 36 h. Subsequently, the reaction mixture was cooled, neutralized by adding saturated $NaHCO_3$ solution (20 mL) and extracted with ethyl acetate (3 × 30 mL). The combined organic layer was dried over anhydrous Na_2SO_4 and concentrated under reduced pressure to afford **S11** as slightly orange oil (1.96 g, 60 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.85 (dd, *J* = 8.0, 1.2 Hz, 1H), 7.30 – 7.22 (m, 1H), 6.74 – 6.55 (m, 2H), 5.71 (bs, 2H), 3.87 (s, 3H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 168.7, 150.6, 134.2, 131.4, 116.8, 116.4, 110.9, 51.7.

HRMS (ESI) $[M+H]^+$ calculated for $C_8H_{10}NO_2$, 152.0712; found, 152.0722.

methyl 2-(benzylamino)benzoate



S12

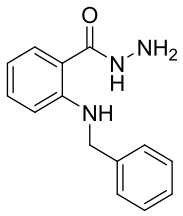
To a stirred solution of **S11** (0.8 g, 5.29 mmol) in acetone (16 mL) was added K_2CO_3 (1.83 g, 13.23 mmol) and benzyl bromide (0.91 g, 5.29 mmol) and the reaction mixture was refluxed for 24 h. Subsequently, the solvent was removed under reduced pressure, water (10 mL) was added and the resulting solution was extracted with ethyl acetate (3 × 20 mL). The combined organic layer was dried over anhydrous Na_2SO_4 and concentrated under reduced pressure and the residue was purified by silica gel column chromatography (100% hexane) to give **S12** as a white solid (0.41 g, 32%)

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.17 (bs, 1H), 7.93 (dd, *J* = 8.0, 1.6 Hz, 1H), 7.39 – 7.22 (m, 6H), 6.70 – 6.51 (m, 2H), 4.46 (d, *J* = 6.0 Hz, 2H), 3.87 (s, 3H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 169.2, 151.1, 151.0, 139.0, 134.7, 131.7, 128.8, 127.3, 127.2, 115.0, 111.8, 110.3, 51.6, 47.1.

HRMS (ESI) $[M+H]^+$ calculated for $C_{15}H_{16}NO_2$, 242.1181; found, 242.1193.

2-(benzylamino)benzohydrazide



S13

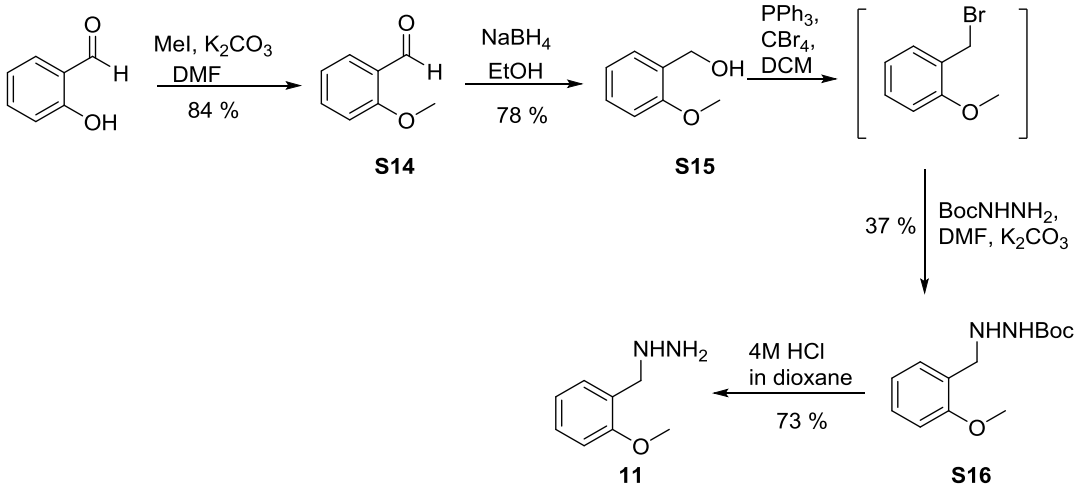
To a solution of **S12** (0.25 g, 1.03 mmol) in ethanol (4 mL) was added hydrazine hydrate (0.11 mL of 78% aqueous solution) and the resulting mixture was refluxed for 16 h. Subsequently, the reaction mixture was concentrated under reduced pressure, water (6 mL) was added and the resulting solution was extracted with ethyl acetate (3 × 20 mL). The combined organic layer was dried over anhydrous Na_2SO_4 and concentrated under reduced pressure and the resulting residue was purified by silica gel column chromatography (10% ethyl acetate in dichloromethane) to yield a mixture of compounds. The resulting crude product was further purified by trituration with ethanol to afford **S13** as a white solid (0.05 g, 20%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.84 (bs, 1H), 7.42 (bs, 1H), 7.39 – 7.21 (m, 7H), 6.64 (d, *J* = 8.4 Hz, 1H), 6.61 – 6.56 (m, 1H), 4.41 (d, *J* = 4.8 Hz, 2H), 4.04 (bs, 2H).

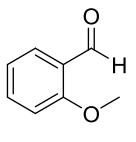
^{13}C NMR (100 MHz, $CDCl_3$) δ 170.9, 149.5, 139.1, 133.4, 128.8, 127.21, 127.2, 127.1, 115.3, 113.5, 112.3, 47.3.

HRMS (ESI) $[M+H]^+$ calculated for $C_{14}H_{16}N_3O$, 242.1293; found, 242.1299.

Scheme S7. Synthesis of (2-methoxybenzyl)hydrazine (**11**)



2-methoxybenzaldehyde



S14

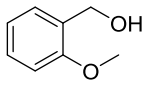
A solution of salicylaldehyde (2.0 g, 16.37 mmol) in DMF (10 mL) was cooled to 0°C and then K_2CO_3 (6.79 g, 49.12 mmol) was added to it in small portions. After addition of K_2CO_3, methyl iodide (3.06 mL, 49.12 mmol) was added to the reaction mixture at 0°C in a dropwise manner. The reaction mixture was slowly warmed to room temperature and stirred at room temperature for 12 h. Subsequently, the mixture was diluted with diethyl ether (30 mL), and washed with H_2O (5 × 15 mL) and brine (1 × 10 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered and concentrated on a rotary evaporator to obtain the crude product which was then purified by silica gel column chromatography (16% ethyl acetate in hexane) to give **S14** as a pale yellow liquid (1.86 g, 84%)

^{1}H NMR (400 MHz, $CDCl_3$) δ 10.48 (s, 1H), 7.83 (dd, *J* = 7.8, 1.8 Hz, 1H), 7.58 – 7.52 (m, 1H), 7.14 – 6.75 (m, 2H), 3.93 (s, 3H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 190.0, 162.0, 136.1, 128.7, 124.9, 120.8, 111.7, 55.7.

HRMS (ESI) $[M+H]^+$ calculated for $C_8H_9O_2$, 137.0603; found, 137.0602

(2-methoxyphenyl)methanol



S15

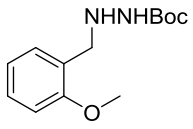
Sodium borohydride (0.42 g, 11.02 mmol) was added to a solution of **S14** (1.0 g, 7.35 mmol) in ethanol (20 mL) at 0°C and stirred for 2 h at room temperature. Subsequently, the solvent was removed in vacuo and a saturated solution of NH_4Cl (10 mL) was carefully added. The mixture was extracted with dichloromethane (3 × 20 mL) and the combined organic layer was washed with brine (1 × 10 mL), dried over anhydrous Na_2SO_4 and concentrated in vacuo to obtain a crude product which was purified by silica gel column chromatography (16% ethyl acetate in hexane) to yield **S15** as pale brown liquid (0.79 g, 78%)

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.34 – 7.26 (m, 2H), 6.98 – 6.92 (m, 1H), 6.89 (d, *J* = 8.8, 1H), 4.69 (s, 2H), 3.87 (s, 3H), 2.42 (bs, 1H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 157.6, 129.2, 129.1, 128.9, 120.8, 110.3, 62.2, 55.4.

HRMS (ESI) $[M+H]^+$ calculated for $C_8H_{11}O_2$, 139.0759; found, 139.0762.

***t*-butyl 2-(2-methoxybenzyl)hydrazine-1-carboxylate**



S16

To a solution of the **S15** (0.2 g, 1.45 mmol) in dichloromethane (6 mL) at 0°C was added CBr_4 (0.72 g, 2.17 mmol) and PPh_3 (0.57 g, 2.17 mmol). The reaction mixture was stirred for 3 h at the room temperature. Subsequently, the solvent was evaporated on a rotary evaporator to obtain the crude bromide intermediate which was used without purification. To a solution of crude bromide (0.145 g, 0.72 mmol) in DMF (4 mL) were added *t*-butyl carbazate (0.73 g, 5.55 mmol) and K_2CO_3 (0.11 g, 0.80 mmol) and the mixture was heated to 90°C and stirred for 2 h under N_2 atmosphere. Subsequently, the mixture was cooled, diluted with diethyl ether (20 mL), and

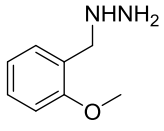
washed with H_2O (5 × 10 mL) and brine (1 × 5 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to obtain crude product which was purified by silica gel column chromatography (16% ethyl acetate in hexane) to give **S16** as a colorless oil (0.08 g, 37%)

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.33 – 7.21 (m, 2H), 6.95 – 6.90 (m, 1H), 6.88 (d, *J* = 8.0 Hz 1H), 5.98 (bs, 1H), 4.33 (bs, 1H), 4.01 (s, 2H), 3.85 (s, 3H), 1.45 (s, 9H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 158.0, 156.6, 130.9, 129.0, 125.9, 120.5, 110.5, 80.4, 55.5, 51.5, 28.5.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{13}H_{20}N_2NaO_3$, 275.1372; found, 275.1371.

(2-methoxybenzyl)hydrazine



11

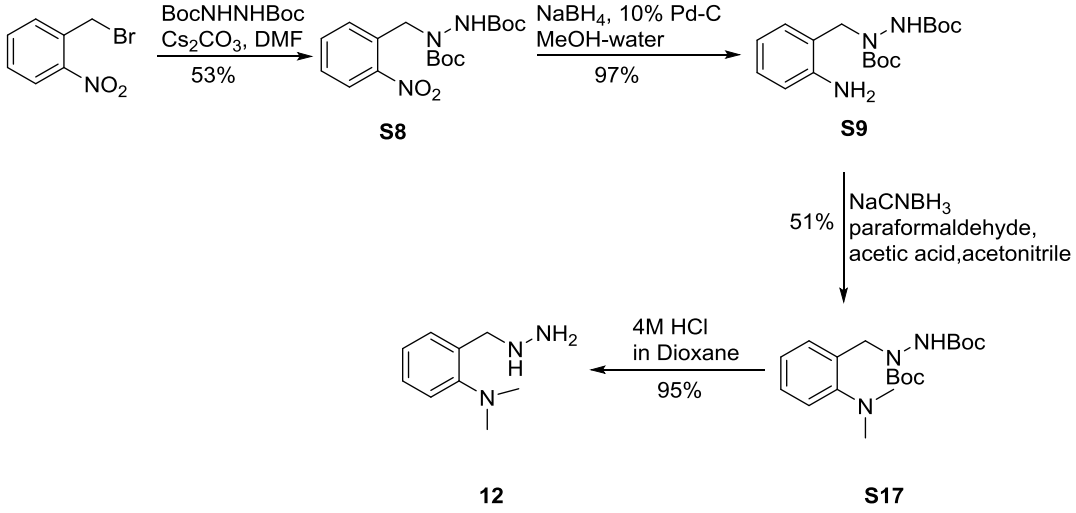
To solution of **S16** (0.06 g, 0.24 mmol) in 1,4-dioxane/water (1 mL/0.1 mL), 4 M HCl in dioxane (1.5 mL) was added. After stirring this mixture for 5 h, diethyl ether was added to it and the resulting suspension was filtered to obtain **11** as a pale yellow solid (0.04 g, 73%).

^{1}H NMR (400 MHz, CD_3OD) δ 7.43 (ddd, *J* = 8.3, 7.6, 1.7 Hz, 1H), 7.36 (dd, *J* = 7.5, 1.6 Hz, 1H), 7.08 (d, *J* = 8.3 Hz, 1H), 7.04 – 6.97 (m, 1H), 4.24 (s, 2H), 3.91 (s, 3H).

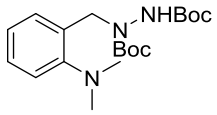
^{13}C NMR (100 MHz, CD_3OD) δ 159.5, 132.8, 132.5, 121.9, 120.4, 112.0, 56.1, 52.5.

HRMS (ESI) $[M+H]^+$ calculated for $C_8H_{13}N_2O$, 153.1028; found, 153.1034.

Scheme S8 Synthesis of 2-(hydrazinylmethyl)-*N*,*N*-dimethylaniline (**13**)



di-*t*-butyl 1-(2-(dimethylamino)benzyl)hydrazine-1,2-dicarboxylate



S17

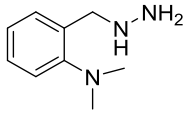
To a solution of **S9** (0.19 g, 0.57 mmol) in acetonitrile (3.84 mL) was added paraformaldehyde (0.17 g, 5.58 mmol) followed by acetic acid (0.56 mL) and sodium cyanoborohydride (0.17 g, 2.68 mmol) under N_2 atmosphere. After stirring for 18 h at room temperature, another portion of sodium cyanoborohydride (0.17 g, 2.68 mmol) was added to the reaction mixture and it was stirred for further 18 h at room temperature. The reaction mixture was diluted with water (10 mL), basified to pH 10 with 5 M NaOH and extracted with ethyl acetate (3 × 30 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered, concentrated under reduced pressure and the residue was purified by silica gel column chromatography (12% ethyl acetate in hexane) to obtain **S17** as white solid (0.11 g, 51%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.25 – 7.16 (m, 2H), 7.11 (d, *J* = 8.0 Hz, 1H), 7.04 (t, *J* = 7.6 Hz, 1H), 6.68 – 5.82 (m, 1H), 4.78 (s, 2H), 2.65 (s, 6H), 1.60 – 1.34 (m, 18H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 155.8, 153.1, 131.4, 128.7, 128.1, 123.3, 119.6, 81.2, 48.8, 45.1, 28.3, 28.3.

HRMS (ESI) $[M+H]^+$ calculated for $C_{19}H_{32}N_3O_4$, 366.2393; found, 366.2397.

2-(hydrazinylmethyl)-*N*,*N*-dimethylaniline (12)



12

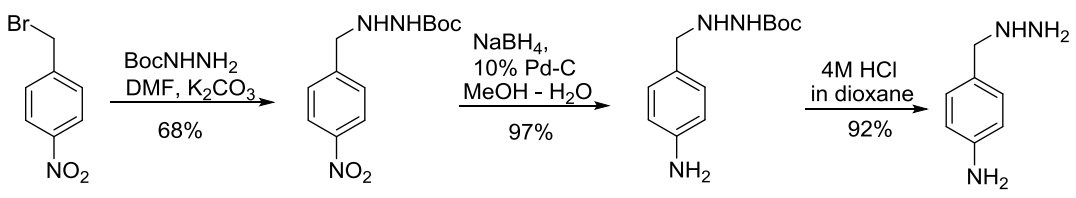
To solution of **S17** (0.087 g, 0.24 mmol) in 1,4-dioxane/water (0.6 mL/0.006 mL), 4 M HCl in dioxane (1.20 mL) was added. After stirring this mixture for 5 h, diethyl ether was added to it and the resulting solution was filtered to obtain **12** as a pale yellow solid (0.062 g, 57%).

^{1}H NMR (400 MHz, D_2O) δ 7.81 (d, *J* = 8.4 Hz, 1H), 7.71 – 7.65 (m, 1H), 7.56 (t, *J* = 7.6 Hz, 1H), 7.53 – 7.49 (m, 1H), 4.44 (s, 2H), 3.35 (s, 6H).

^{13}C NMR (100 MHz, DMSO-D_6) δ 143.0, 132.3, 130.5, 129.7, 128.0, 121.7, 49.2, 47.4.

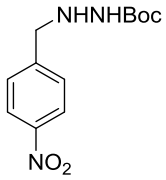
HRMS (ESI) [M+H] + calculated for $C_9H_{16}N_3$, 166.1344; found, 166.1350.

Scheme S9. Synthesis of 4-(hydrazinylmethyl)aniline (**13**)



S18 **S19** **13**

***t*-butyl 2-(4-nitrobenzyl)hydrazine-1-carboxylate**



S18

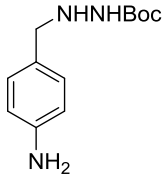
According to the procedure of Bare *et al.*,[1] to a solution of 4-nitrobenzyl bromide (0.2 g, 0.93 mmol) in DMF (1.5 mL) were added *t*-butyl carbazate (0.94 g, 7.10 mmol) and K_2CO_3 (0.14 g, 1.02 mmol). The mixture was heated to 90°C and stirred for 2 h under N_2 atmosphere. Subsequently, the mixture was cooled, diluted with diethyl ether (20 mL), and washed with H_2O (5 × 10 mL) and brine (1 × 5 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to obtain the crude product which was purified by silica gel column chromatography (4% ethyl acetate in dichloromethane) to give **S18** as a yellow solid (0.17 g, 68%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.19 (d, *J* = 8.8 Hz, 2H), 7.52 (d, *J* = 8.4 Hz, 2H), 6.05 (bs, 1H), 4.33 (bs, 1H), 4.11 (d, *J* = 3.2 Hz, 2H), 1.45 (s, 9H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 156.9, 147.5, 145.7, 129.6, 123.7, 81.1, 55.1, 28.4.

HRMS (ESI) $[M+H]^+$ calculated for $C_{12}H_{18}N_3O_4$, 268.1297; found, 268.1295.

***t*-butyl 2-(4-aminobenzyl) hydrazine-1-carboxylate**



S19

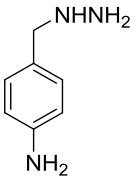
According to the procedure of Basu *et al.*,[2] to a 10 mL round-bottomed flask charged with 10% Pd-C (0.01 g) was added a solution of $NaBH_4$ (0.03. g, 0.75 mmol in 2 mL water). Gas evolution was observed. Reaction mixture was slowly heated to 50°C and to this solution, a methanolic solution (1 mL) of **S18** (0.1 g, 0.37 mmol) was then added dropwise with constant stirring. Cessation of gas evolution indicated completion of the reduction within 20 minutes. After completion of the reaction, the reaction mixture was filtered through a celite pad and the filtrate was concentrated on a rotary evaporator. The resulting residue was dissolved in ethyl acetate (20 mL) and water (10 mL) was added. After separating the organic and aqueous layers, the aqueous layer was washed with ethyl acetate (2 × 10 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to obtain **S19** as white solid (0.09 g, 97%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.12 (d, *J* = 8.0 Hz, 2H), 6.64 (d, *J* = 8.0 Hz, 2H), 6.13 (bs, 1H), 4.00 – 3.41 (m, 5H), 1.46 (s, 9H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 156.8, 145.9, 130.3, 127.6, 115.2, 80.5, 55.6, 28.5.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{12}H_{19}N_3NaO_2$, 260.1375; found, 260.1377.

4-(hydrazinylmethyl)aniline



13

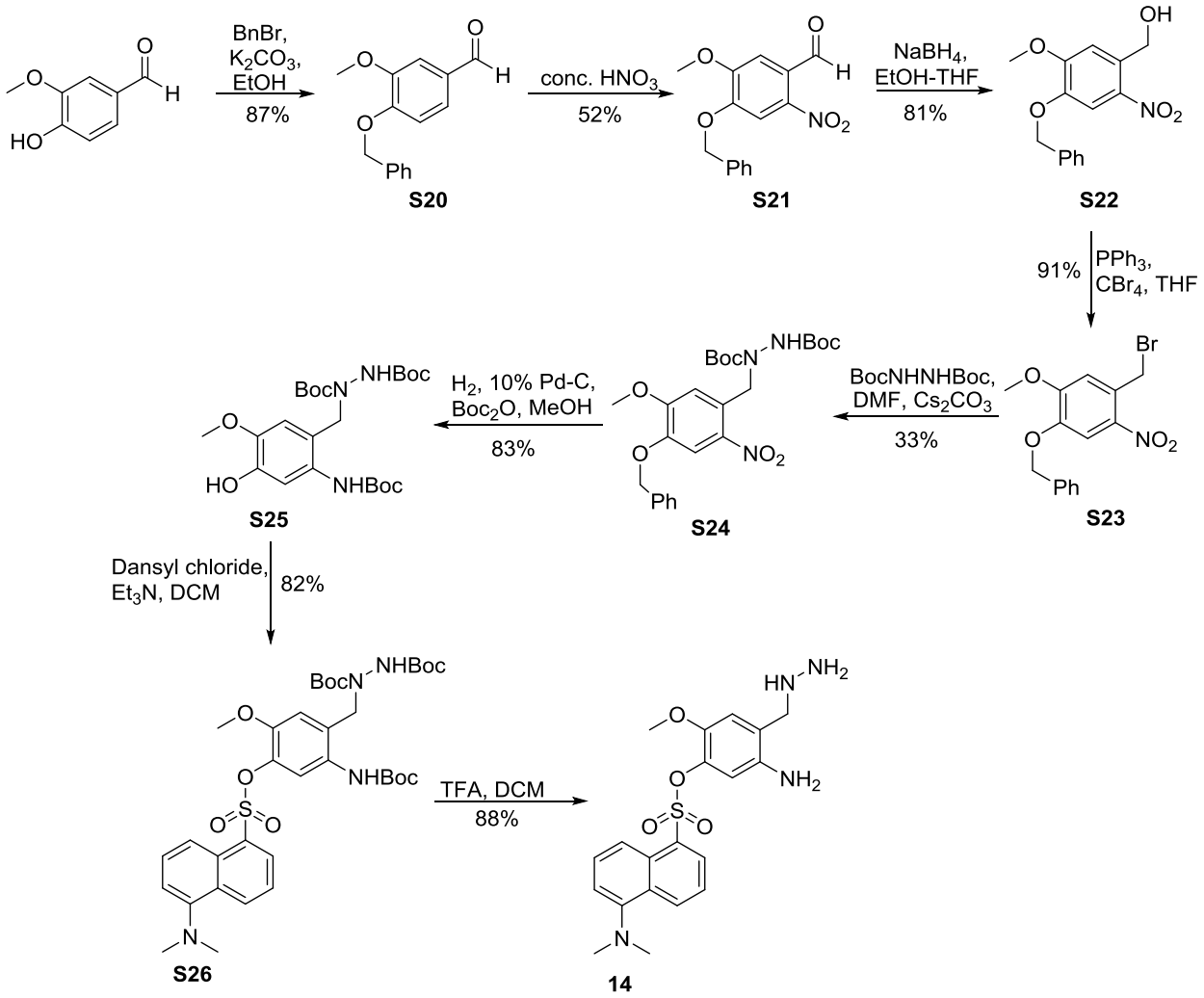
To solution of **S19** (0.08 g, 0.34 mmol) in 1, 4-dioxane/water (1.3 mL/0.13 mL), 4 M HCl in dioxane (2 mL) was added. After stirring this mixture for 5 h, diethyl ether was added to it and the resulting solution was filtered to obtain **13** as a pale yellow solid (0.08 g, 92%).

^{1}H NMR (400 MHz, CD_3OD) δ 7.65 (d, *J* = 8.5 Hz, 2H), 7.48 (d, *J* = 8.5 Hz, 2H), 4.21 (s, 2H).

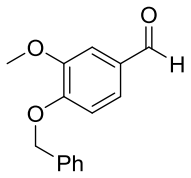
^{13}C NMR (100 MHz, CD_3OD) δ 136.3, 132.4, 132.3, 124.7, 54.6.

HRMS (ESI) $[M+Na]^+$ calculated for $C_7H_{11}N_3Na$, 160.0851; found, 160.0847.

Scheme S10. Synthesis of 5-amino-4-(hydrazinylmethyl)-2-methoxyphenyl 5-(dimethylamino)naphthalene-1-sulfonate **(14)**



4-(benzyloxy)-3-methoxybenzaldehyde



S20

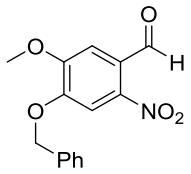
To a stirred solution of vanillin (12.0 g, 78.87 mmol) in ethanol (60 mL) were added K_2CO_3 (14.99 g, 108.45 mmol) and benzyl bromide (10.52 mL, 88.57 mmol), and the mixture stirred under a nitrogen atmosphere overnight. Subsequently, the mixture was filtered through a celite plug, washed with DCM (3 × 50 mL), and the combined organic solvents were removed in vacuo. The resulting oily residue was dissolved in DCM (100 mL), and washed with 5% NaOH solution (70 mL), dried over K_2CO_3, concentrated, and crystallized from ethanol to afford **S20** as a white solid (16.92 g, 87%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 9.83 (s, 1H), 7.45 – 7.29 (m, 7H), 6.99 (d, *J* = 8.4 Hz, 1H), 5.24 (s, 2H), 3.94 (s, 3H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 191.0, 153.7, 150.2, 136.1, 130.4, 128.8, 128.3, 127.3, 126.7, 112.5, 109.5, 71.0, 56.2.

HRMS (ESI) $[M+H]^+$ calculated for $C_{15}H_{15}O_3$, 243.1021; found, 243.1023.

4-(benzyloxy)-5-methoxy-2-nitrobenzaldehyde



S21

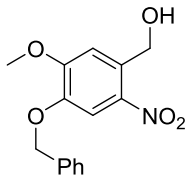
In a 50 mL round-bottomed flask, conc. HNO_3 (16 mL) was added and the flask was cooled to 0°C. To this mixture, **S20** (3.30 g, 13.61 mmol) was added in small portions at 0°C over a period of 15 min. After 60 min of stirring at 0°C, the reaction mixture was poured into an aluminum foil-covered beaker containing ice cold water (100 mL). After stirring this mixture for 15 min in the dark, the solid was filtered and dried to get crude **S21**, which was purified by recrystallization from ethyl acetate (2.04 g, 52%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 10.44 (s, 1H), 7.67 (s, 1H), 7.53 – 7.32 (m, 6H), 5.27 (s, 2H), 4.02 (s, 3H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 187.9, 153.8, 151.5, 143.7, 135.0, 129.0, 128.8, 127.7, 125.8, 110.1, 109.0, 71.7, 56.9.

HRMS (ESI) $[M+H]^+$ calculated for $C_{15}H_{14}NO_5$, 288.0872; found, 288.0876.

(4-(benzyloxy)-5-methoxy-2-nitrophenyl)methanol



S22

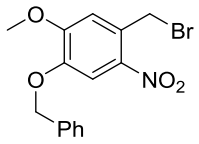
Sodium borohydride (0.27 g, 7.03 mmol) was added to a solution of **S21** (1.35 g, 4.69 mmol) in 1:1 ethanol-THF (20 mL) at 0°C and stirred for 2 h at room temperature. The solvent was removed in vacuo and a saturated solution of NH_4Cl (10 mL) was carefully added. The mixture was extracted with dichloromethane (3 × 20 mL) and the combined organic layers were washed with brine (1 × 20 mL) , dried over anhydrous Na_2SO_4 and concentrated in vacuo to obtain the crude product which was purified by silica gel column chromatography (30% ethyl acetate in hexane) to obtain **S22** as yellow solid (1.10 g, 81%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.76 (s, 1H), 7.48 – 7.30 (m, 5H), 7.18 (s, 1H), 5.19 (s, 2H), 4.94 (d, *J* = 6.4 Hz, 2H), 3.99 (s, 3H), 2.73 (t, *J* = 6.4 Hz, 1H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 154.6, 147.0, 139.7, 135.8, 132.7, 128.9, 128.5, 127.7, 111.3, 110.4, 71.4, 62.9, 56.6.

HRMS (ESI) $[M+H]^+$ calculated for $C_{15}H_{16}NO_5$, 290.1028; found, 290.1030.

1-(benzyloxy)-4-(bromomethyl)-2-methoxy-5-nitrobenzene



S23

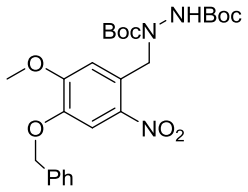
To a solution of the **S22** (1.10 g, 3.80 mmol) in dichloromethane (16 mL) at 0°C was added CBr_4 (2.21 g, 6.66 mmol) and PPh_3 (1.75 g, 6.66 mmol). The reaction mixture was stirred for 3 h at the room temperature. Subsequently, the solvent was removed on a rotary evaporator to obtain the crude product, which was then purified by silica gel column chromatography (20% ethyl acetate in hexane) to give **S23** as orange solid (1.22 g, 91%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.73 (s, 1H), 7.56 – 7.30 (m, 5H), 6.95 (s, 1H), 5.20 (s, 2H), 4.86 (s, 2H), 3.99 (s, 3H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 153.9, 148.1, 140.2, 135.5, 128.9, 128.6, 127.8, 127.7, 114.1, 110.6, 71.4, 56.7, 30.3.

HRMS (ESI) $[M+H]^+$ calculated for $C_{15}H_{15}BrNO_4$, 352.0184; found, 352.0187.

di-*t*-butyl 1-(4-(benzyloxy)-5-methoxy-2-nitrobenzyl)hydrazine-1,2-dicarboxylate



S24

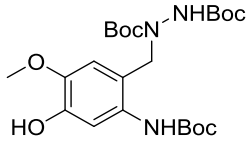
To a stirred solution of di-*t*-butyl hydrazodiformate (BocNHNHBoc) *(*2.64 g, 11.36 mmol) and Cs_2CO_3 (2.03 g, 6.25 mmol) in dry DMF (5 mL) at 80°C was added a solution of **S23** (2.0 g, 5.68 mmol) in dry DMF (5 mL). After stirring for 45 min at 80°C under N_2 atmosphere, the mixture was cooled, diluted with diethyl ether (40 mL), and washed with H_2O (5 × 20 mL) and brine (1 × 10 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to obtain crude product which was purified by silica gel column chromatography (4% ethyl acetate in DCM) to give **S24** as a yellow solid (0.94 g, 33%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.70 (s, 1H), 7.57 – 7.29 (m, 6H), 6.42 (bs, 1H), 5.18 (s, 2H), 5.00 (s, 2H), 3.99 (s, 3H), 1.44 (m, 18H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 155.4, 154.2, 146.8, 140.2, 135.9, 128.9, 128.5, 127.7, 112.2, 111.7, 110.3, 82.1, 81.5, 71.3, 56.8, 52.7, 28.3, 28.2.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{25}H_{33}N_3NaO_8$, 526.2165; found, 526.2167.

di-*t*-butyl 1-(2-((*t*-butoxycarbonyl)amino)-4-hydroxy-5-methoxybenzyl)hydrazine-1,2-dicarboxylate



S25

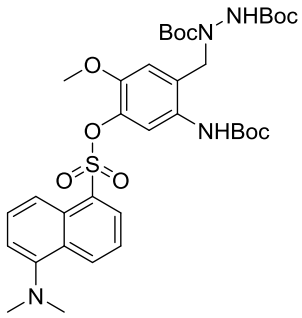
To solution of **S24** (0.9 g, 1.79 mmol) in methanol (15 mL) were added di-*t*-butyl dicarbonate (0.43 g, 1.97 mmoles) and 10% Pd-C (0.10 g). After stirring the resulting heterogeneous mixture for 5 h under H_2 atmosphere, the H_2 balloon was replaced with a N_2 balloon and stirring was continued overnight. The reaction mixture was subsequently filtered through a celite pad and the filtrate was concentrated on a rotary evaporator to obtain the crude product. The crude product was purified by silica gel column chromatography (30% ethyl acetate in hexane) to give **S25** as white solid (0.72 g, 83 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.80 – 7.42 (m, 1H), 6.66 (s, 1H), 6.37 – 6.12 (m, 1H), 5.69 (s, 1H), 4.80 – 4.19 (m, 2H), 3.82 (s, 3H), 1.58 – 1.19 (m, 27H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 155.7, 155.1, 153.7, 146.1, 142.6, 131.5, 117.8, 114.0, 109.3, 82.3, 81.5, 80.1, 56.5, 50.3, 28.5, 28.3, 27.8.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{23}H_{37}N_3NaO_8$, 506.2478; found, 526.2471.

di-*t*-butyl 1-(2-((*t*-butoxycarbonyl)amino)-4-(((5-(dimethylamino)naphthalen-1-yl)sulfonyl)oxy)-5-methoxybenzyl)hydrazine-1,2-dicarboxylate



S26

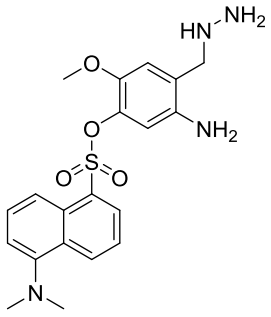
To a stirred solution of **S25** (0.1 g, 0.21 mmoles) in dry dichloromethane (5 mL) was added dry triethylamine (0.07 mL, 0.51 mmol). The resulting solution was cooled to 0°C and a solution of dansyl chloride (0.05 g, 0.17 mmol) in dry dichloromethane (10 mL) was added to it at 0°C. The reaction mixture was warmed to room temperature and stirred for 4 h. The reaction mixture was extracted with water and the aqueous layer was washed with dichloromethane (3 × 15 mL). The combined organic layers were washed with brine (1 × 15 mL), dried over anhydrous Na_2SO_4 and concentrated in vacuo to obtain the crude product which was purified by silica gel column chromatography (4% ethyl acetate in dichloromethane) to obtain **S26** as yellow solid (0.1 g, 82%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.61 (d, *J* = 8.8 Hz, 1H), 8.54 (d, *J* = 8.8 Hz, 1H), 8.17 (d, *J* = 7.2 Hz, 1H), 7.94 – 7.61 (m, 2H), 7.53 – 7.44 (m, 1H), 7.23 (d, *J* = 7.6 Hz, 1H), 6.64 (s, 1H), 6.23 – 5.97 (m, 1H), 4.67 – 4.23 (m, 2H), 3.38 (s, 3H), 2.90 (s, 6H), 1.78 – 0.97 (m, 27H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 155.7, 155.0, 153.3, 151.4, 138.5, 132.7, 131.6, 131.1, 130.7, 130.5, 129.9, 128.6, 127.2, 123.1, 120.7, 116.0, 115.6, 105.3, 82.7, 81.8, 80.3, 56.1, 50.5, 45.7, 28.4, 28.3, 28.0.

HRMS (ESI) $[M+H]^+$ calculated for $C_{35}H_{49}N_4O_{10}S$, 717.3169; found, 717.3179.

5-amino-4-(hydrazinylmethyl)-2-methoxyphenyl 5-(dimethylamino)naphthalene-1-sulfonate



14

To a 25 mL round-bottomed flask charged with **S26** (0.02 g, 0.03 mmol), was added a 1:1 mixture of trifluoroacetic acid and dichloromethane (6 mL) at room temperature. This mixture was allowed to stir for 30 min. The solvent was then evaporated in vacuo and the crude residue was dissolved in 3 mL of dichloromethane and concentrated in vacuo. The residue was again dissolved in 3 mL of dichloromethane and the solvent was evaporated. This process was

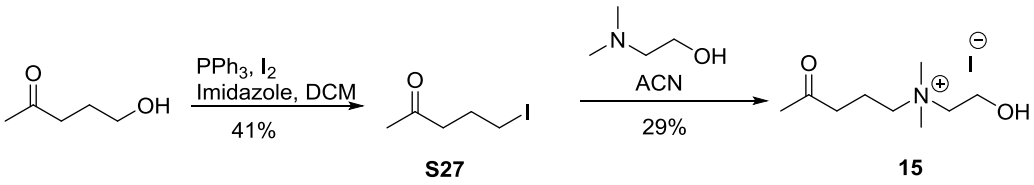
repeated one more time to ensure that trifluoroacetic acid was completely removed. Finally, the residue was dissolved in 1:1 water-acetonitrile mixture (1 mL) and lyophilized to yield **14** as pale yellow powder (0.02 g, 88%).

^{1}H NMR (400 MHz, D_2O) δ 8.94 (d, *J* = 8.8 Hz, 1H), 8.57 (d, *J* = 8.4 Hz, 1H), 8.29 (d, *J* = 7.6 Hz, 1H), 8.19 (d, *J* = 7.6 Hz, 1H), 8.04 – 7.96 (m, 1H), 7.90 – 7.82 (m, 1H), 7.38 (s, 1H), 6.98 (s, 1H), 4.21 (s, 2H), 3.54 (s, 6H), 3.04 (s, 3H).

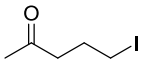
^{13}C NMR (100 MHz, CD_3OD) δ 151.4, 139.9, 133.5, 132.2, 131.6, 130.3, 129.6, 129.3, 128.6, 127.5, 124.9, 124.8, 123.0, 119.4, 117.8, 117.0, 56.3, 51.3, 46.3.

HRMS (ESI) $[M+Na]^+$ calculated for $C_{20}H_{24}N_4NaO_4S$, 439.1416; found, 439.1416.

Scheme S11 Synthesis of *N*-(2-hydroxyethyl)-*N*,*N*-dimethyl-4-oxopentan-1-aminium iodide (pentanoyl choline) (**15**)



5-iodopentan-2-one



S27

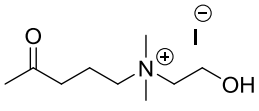
To a solution of PPh_3 (7.71 g, 29.37 mmol) and imidazole (4.47 g, 29.37 mmol) in dry DCM (40 mL) was added I_2 (7.46 g, 39.37 mmol) at 0°C under N_2 atmosphere. After stirring the solution for 30 min at 0°C, a solution of 5-hydroxypentanone (1.99 mL, 19.58 mmol) in dry DCM (20 mL) was added to it at 0°C. The reaction mixture was warmed to room temperature and stirred at room temperature for overnight. The reaction was quenched by adding methanol (3.3 mL), diluted with water (50 mL) and extracted with 1:3 Et_2O-hexane (5 × 100 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated under reduced pressure and the residue was purified by silica gel column chromatography (20% ethyl acetate in hexane) to obtain **S27** as pale yellow liquid (1.70 g, 41%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 3.22 (t, *J* = 6.4 Hz, 2H), 2.59 (t, *J* = 7.2 Hz, 2H), 2.17 (s, 3H), 2.07 (p, *J* = 6.8 Hz, 2H).

^{13}C NMR (100 MHz, $CDCl_3$) δ 207.4, 44.0, 30.3, 27.1, 6.6.

HRMS (ESI) [M+H] + calculated for $C_5H_{10}IO$, 212.9776; found, 212.9778.

***N*-(2-hydroxyethyl)-*N*,*N*-dimethyl-4-oxopentan-1-aminium iodide (pentanoyl choline)**



15

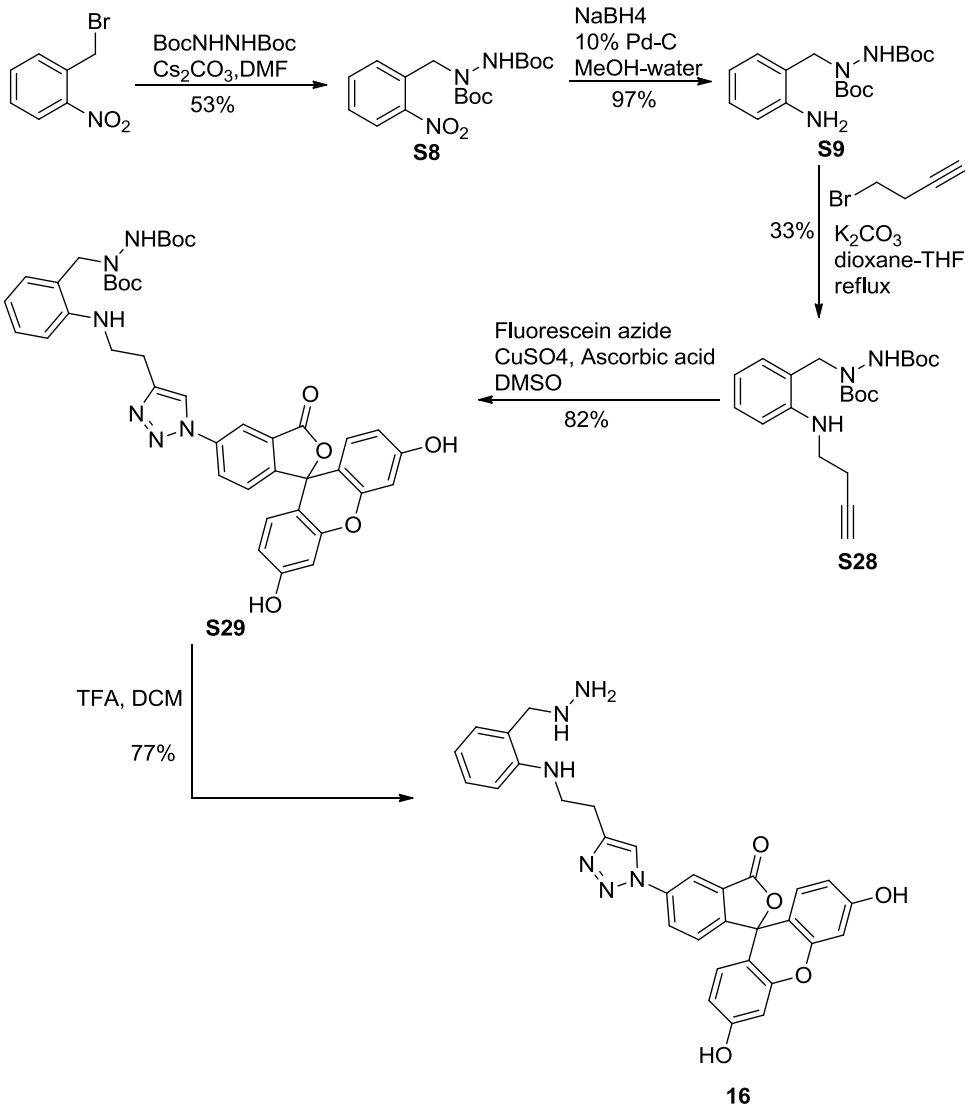
To a solution of **S27** (0.15 g, 0.71 mmol) in acetonitrile (5 mL) was added *N*,*N*-dimethylaminoethanol (0.071 mL, 0.71 mmol). The reaction mixture was heated to 60°C and stirred for 48 h. Subsequently, the solvent was removed under reduced pressure and the residual oil was triturated with ethyl acetate (6 × 2 mL). The residue was purified by reverse phase flash chromatography (Redisep Rf 43 g C18 column) using water-acetonitrile as mobile phase (Gradient: 0-20 min (100 % water), 20-30 min (100-0 % water), 30-35 min (0 % water), 35-40 min (0-100 % water); flow rate = 5 mL/min) to afford **15** as yellow oil (0.062 g, 29%).

^{1}H NMR (400 MHz, D_2O) δ 4.09 – 4.02 (m, 2H), 3.53 – 3.48 (m, 2H), 3.39 – 3.31 (m, 2H), 3.15 (s, 6H), 2.71 (t, *J* = 6.9 Hz, 2H), 2.21 (s, 3H), 2.07 – 1.97 (m, 2H).

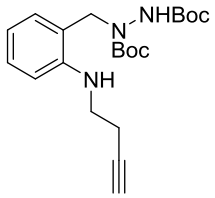
^{13}C NMR (100 MHz, CD_3CN) δ 207.8, 66.1, 65.1, 56.2, 52.5, 39.7, 30.0, 17.5.

HRMS (ESI) $[M]^+$ calculated for $C_9H_{20}NO_2$, 174.1489; found, 174.1500.

Scheme S12 Synthesis of 5-(4-(2-((2-(hydrazinylmethyl)phenyl)amino)ethyl)-1H-1,2,3-triazol-1-yl)-3',6'-dihydroxy-3H-spiro[isobenzofuran-1,9'-xanthen]-3-one (**16**)



di-*t*-butyl 1-(2-(but-3-yn-1-ylamino)benzyl)hydrazine-1,2-dicarboxylate



S28

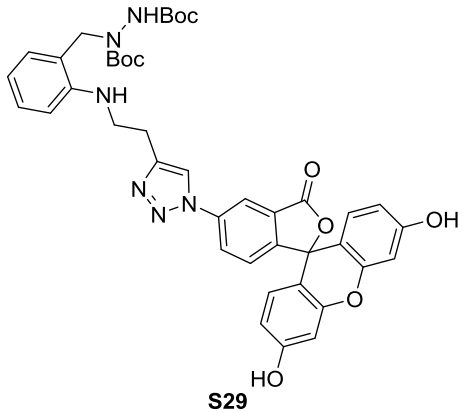
To a solution of **S9** (0.2 g, 0.59 mmol) in 1:1 THF-dioxane (4 mL) was added 4-bromo-1-butyne (0.084 mL, 0.89 mmol) and DIPEA (0.12 mL, 0.71 mmol). After refluxing the mixture for 18 h, 4-bromo-1-butyne (0.084 mL, 0.89 mmol) and DIPEA (0.12 mL, 0.71 mmol) were added again and refluxing was continued for further 24 h. The reaction mixture was concentrated on a rotary evaporator. The resulting residue was dissolved in ethyl acetate (20 mL) and water (10 mL) was added. After separating the organic and aqueous layers, the aqueous layer was washed with ethyl acetate (2 × 20 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated under reduced pressure and the residue was purified by silica gel column chromatography (16% ethyl acetate in hexane) to obtain **S28** as yellow oil (0.076 g, 33%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.21 (td, *J* = 7.6, 1.6 Hz, 1H), 7.05 (dd, *J* = 7.2, 1.6 Hz, 1H), 6.70 – 6.57 (m, 2H), 6.29 – 5.75 (m, 1H), 5.56 – 4.17 (m, 3H), 3.60 – 3.10 (m, 2H), 2.50 (td, *J* = 7.2, 2.8 Hz, 2H), 2.02 (t, *J* = 2.8 Hz, 1H), 1.53 – 1.21 (m, 18H).

^{13}C NMR (101 MHz, $CDCl_3$) δ 156.0, 154.8, 146.4, 132.1, 129.6, 120.4, 116.5, 110.2, 82.1, 81.9, 81.3, 69.8, 50.8, 42.6, 28.4, 28.1, 19.1.

HRMS (ESI) $[M + H]^+$ calculated for $C_{21}H_{32}N_3O_4$, 390.2393; found, 390.2391.

di-*t*-butyl 1-(2-((2-(1-(3',6'-dihydroxy-3-oxo-3H-spiro[isobenzofuran-1,9'-xanthen]-5-yl)-1H-1,2,3-triazol-4-yl)ethyl)amino)benzyl)hydrazine-1,2-dicarboxylate (S29)



S29

To a solution of 5-azidofluorscein (0.040 g, 0.11 mmol) in DMSO (2.5 mL) was added a solution of **S28** (0.064 g, 0.16 mmol) in DMSO. Subsequently, $CuSO_4$ (0.053 g, 0.21 mmol) and L-ascorbic acid (0.056 g, 0.32 mmol) were added to the reaction mixture. The reaction mixture was heated to 60°C and stirred for 24 h. The mixture was diluted with ethyl acetate (25 mL) and extracted it with water (5 × 30 mL) and brine (1 × 5 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered, and concentrated on a rotary evaporator to get crude product which

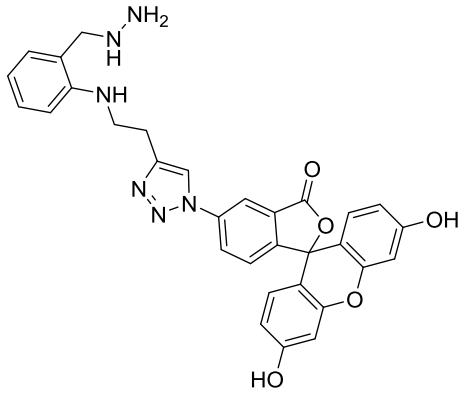
was then purified by silica gel column chromatography (4/5.5/0.5 ethyl acetate/hexane/methanol) to give **S29** as a yellow solid (0.063 g, 82%).

^{1}H NMR (400 MHz, DMSO-d_6) δ 10.17 (s, 2H), 8.99 – 8.82 (m, 1H), 8.49 – 8.39 (m, 1H), 8.39 – 8.29 (m, 1H), 7.51 (dd, *J* = 8.4, 2.0 Hz, 1H), 7.33 – 7.10 (m, 2H), 7.07 – 6.95 (m, 1H), 6.76 – 6.47 (m, 7H), 3.51 – 3.41 (m, 2H), 3.05 (t, *J* = 7.2 Hz, 1H), 2.99 (s, 1H), 1.53 – 1.29 (m, 18H).

^{13}C NMR (100MHz, DMSO-d_6) δ 167.7, 159.7, 154.8, 151.5, 146.3, 146.1, 138.1, 130.3, 129.2, 128.4, 127.9, 127.0, 125.8, 121.2, 120.9, 115.8, 115.2, 112.7, 109.7, 109.0, 102.3, 83.5, 80.2, 79.4, 42.6, 42.1, 28.0, 27.9, 27.8.

HRMS (ESI) [M+H] + calculated for $C_{41}H_{43}N_6O_9$, 763.3092; found, 763.3091.

5-(4-(2-((2-(hydrazinylmethyl)phenyl)amino)ethyl)-1H-1,2,3-triazol-1-yl)-3',6'-dihydroxy-3H-spiro[isobenzofuran-1,9'-xanthen]-3-one (16)



16

To a 10 mL round-bottomed flask charged with **S29** (0.008 g, 0.011 mmol), was added a 1:1 mixture of trifluoroacetic acid and dichloromethane (1.6 mL) at room temperature. This mixture was allowed to stir for 30 min. The solvent was then evaporated in vacuo and the crude residue was dissolved in 2 mL of dichloromethane and concentrated in vacuo. The residue was again dissolved in 2 mL of dichloromethane and the solvent was evaporated. This process was repeated one more time to ensure that trifluoroacetic acid was completely removed. Finally, the residue was dissolved in 1:1 water-acetonitrile mixture (1 mL) and lyophilized to yield **16** as yellow powder (0.072 g, 77%).

^{1}H NMR (400 MHz, DMSO-D_6) δ 8.92 (d, *J* = 6 Hz, 1H), 8.43 (d, *J* = 1.6 Hz, 1H), 8.33 (dd, *J* = 8.4, 2.0 Hz, 1H), 7.51 (d, *J* = 8.4 Hz, 1H), 7.22 (t, *J* = 7.2 Hz, 1H), 7.12 (d, *J* = 7.6 Hz, 1H), 6.81 – 6.61 (m, 6H), 6.57 (dd, *J* = 8.4, 2.4 Hz, 2H), 3.98 (s, 2H), 3.49 (d, *J* = 7.2 Hz, 3H), 3.06 (t, *J* = 7.2 Hz, 2H).

^{13}C NMR (100 MHz, DMSO-D_6) δ 167.8, 159.6, 160.0, 151.6, 146.9, 146.3, 138.1, 129.3, 128.0, 127.1, 126.0, 121.6, 121.3, 116.5, 116.2, 115.4, 112.7, 110.7, 109.1, 102.3, 96.8, 51.0, 42.6, 42.2, 25.1.

HRMS (ESI) [M+H] + calculated for $C_{31}H_{27}N_6O_5$, 563.2043; found, 563.2053.

4. Procedures employed for kinetic studies:

a) General procedure for kinetic experiments:

Hydrazines and 2-formylpyridine were dissolved in Type I water to a concentration of 25 mM and 5 mM respectively and used immediately. To begin with, baseline scans were obtained with the sample cuvette containing the hydrazine solutions (25 mM, 2 µL) and sodium phosphate buffer (250 mM) at pH 7.0 (196 µL), and the reference cuvette containing sodium phosphate buffer (250 mM) at pH 7.0 (200 µL). Subsequently, a solution of 2-formylpyridine (5 mM, 2 µL) was added to the sample cuvette, and UV scans were recorded at 5 min intervals over 1 h. These UV scanning experiments were performed with each hydrazine compound and yielded the wavelength at which maximal increase in absorbance occurred due to hydrazone formation, which were chosen for performing the kinetics experiments. To obtain kinetic traces for the formation of a each hydrazone, the respective hydrazine solution (25 mM, 2 µL) was added to the sample cuvette containing sodium phosphate buffer (250 mM) at pH 7.0 (196 µL), and absorbance at the chosen wavelength (as described above) was set to zero. The reference cuvette contained sodium phosphate buffer (250 mM) at pH 7.0 (200 µL). Subsequently, a solution of 2-formylpyridine (5 mM, 2 µL) was added to the sample cuvette and the absorbance was monitored until saturation was reached (except for slow-reacting hydrazine compounds, benzhydrazide (**1**) and 2-aminobenzhydrazide (**2**), for which hydrazone formation was monitored for 12 h). Second order rate constants (k_2) and maximum absorbance (A_{max}) were determined by fitting the equation below to the absorbance vs time plot.[3]

$A_t = (A_{max} \times (1\text{-}\exp(0.0002 \times k_2 \times t)) / (0.2\text{-}\exp(0.0002 \times k_2 \times t))$, where A_t is the absorbance at a particular time (t).

To obtain normalized absorbance at different time points, the following equation was employed:

$$\text{Normalized absorbance} = A_t/A_{max}$$

b) Procedure for kinetic studies on intermolecular catalysis:

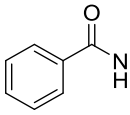
Acetylhydrazide (**10**) and 2-formylpyridine were dissolved in Type I water to a concentration of 25 mM and 5 mM respectively and used immediately. Aniline was dissolved in acetonitrile to a concentration of 250 mM and used immediately. *p*-methoxy anthranilic acid was dissolved in DMSO to a concentration of 250 mM and used immediately. Acetylhydrazide (**10**) solution (25 mM, 2 µL) and catalyst solution (250 mM, 2 µL) were added to a cuvette containing sodium phosphate buffer (250 mM) at pH 7.0 (194 µL), and the absorbance was set to zero at 300 nm in case of aniline and 290 nm for *p*-methoxy anthranilic acid. The reference cuvette contained sodium phosphate buffer (250 mM) at pH 7.0 (196 µL), the acetylhydrazide (**10**) solution (25 mM, 2 µL) and catalyst solution (250 mM, 2 µL). Subsequently, a solution of 2-formylpyridine (5 mM, 2 µL) was added to the sample cuvette and absorbance was monitored until saturation was reached. Each reaction was performed in triplicate. Second order rate constants (k_2) and maximum absorbance (A_{max}) were determined by fitting the equation below to the absorbance vs time plot.[3]

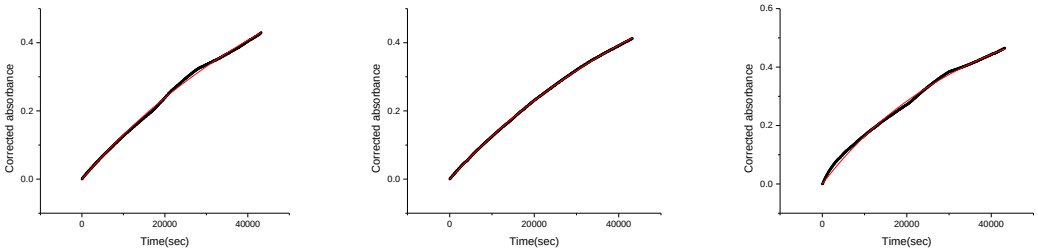
$A_t = (A_{max} \times (1\text{-}\exp(0.0002 \times k_2 \times t)) / (0.2\text{-}\exp(0.0002 \times k_2 \times t))$, where A_t is the absorbance at a particular time (t).

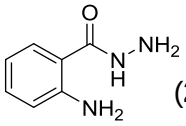
Figure S1. Kinetic traces:

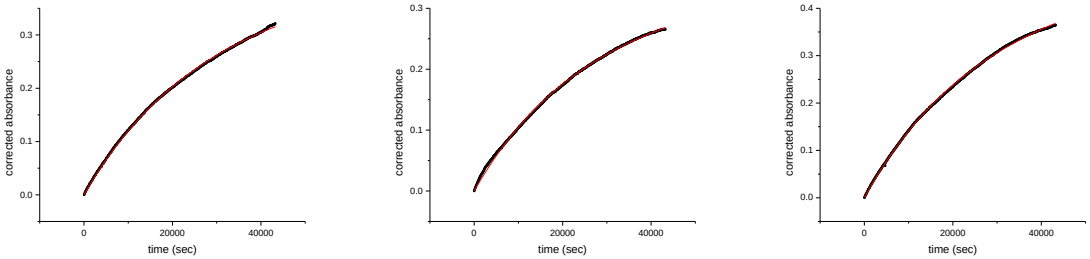
Final concentration used: [hydrazine] = 250 μM, [2-formylpyridine] = 50 μM, [aniline or *p*-methoxy anthranilic acid] = 2.5 mM except for S. No. 16 where 250 μM aniline was used, Buffer = 250 mM sodium phosphate (pH = 7).

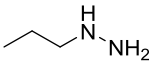
The wavelength at which hydrazone formation was monitored for each hydrazine is mentioned adjacent to the compound structure below.

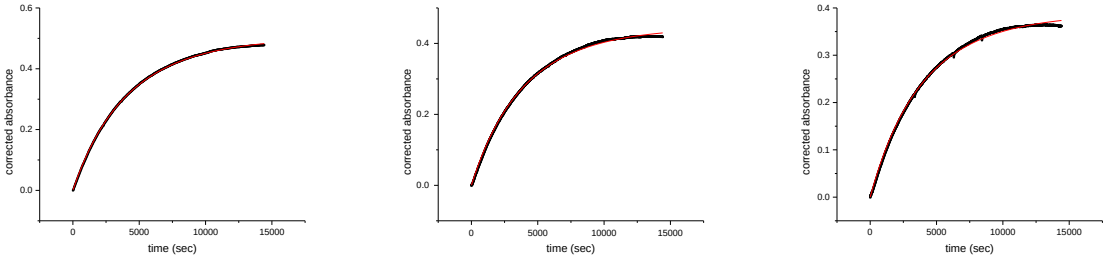
1.  (**1,** 303 nm)

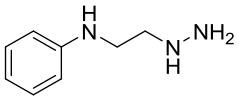


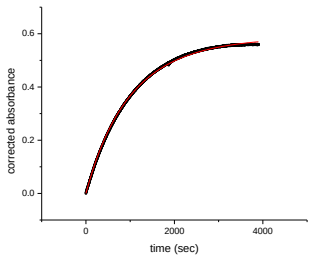
2.  (**2,** 294 nm)

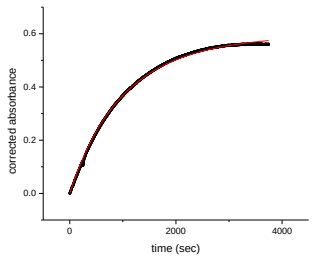


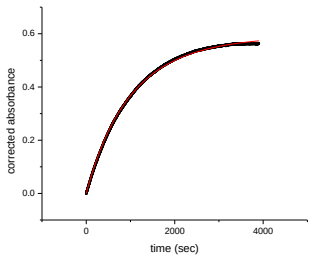
3.  (**3,** 301 nm)

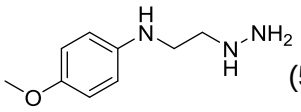


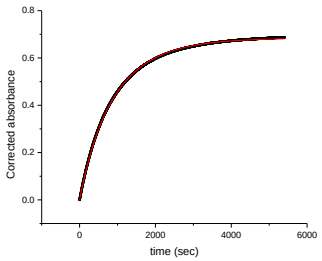
4.  (**4,** 305 nm)

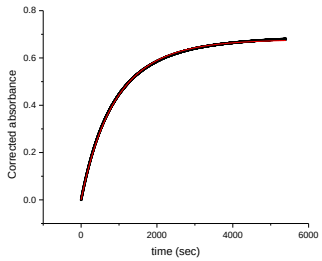


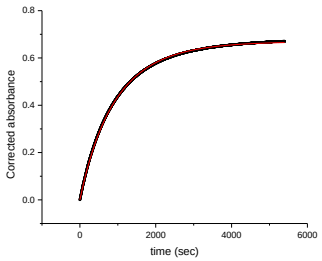


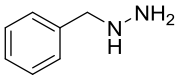


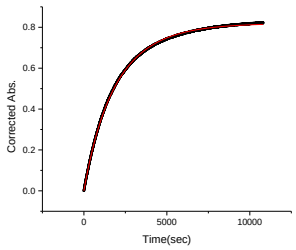
5.  (**5,** 302 nm)

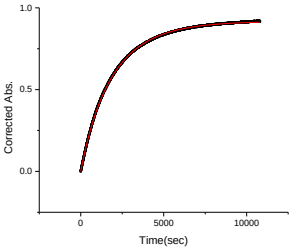


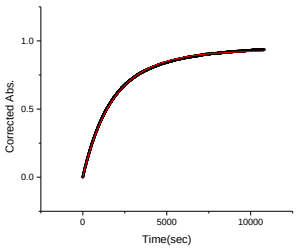


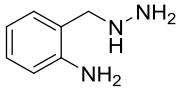


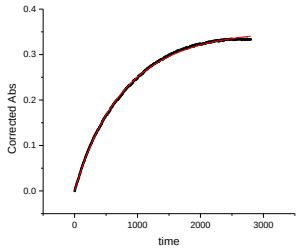
6.  (**6,** 300 nm)

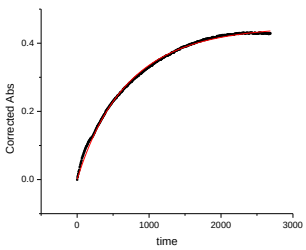


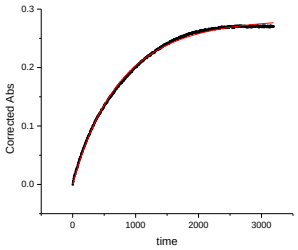


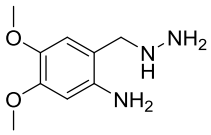


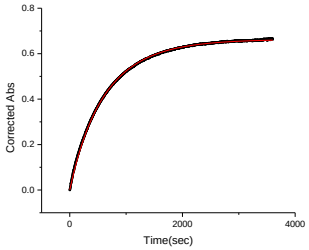
7.  (**7,** 313 nm)

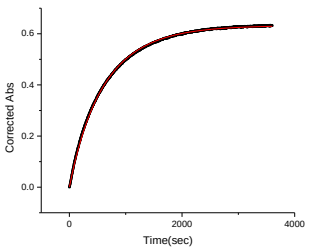


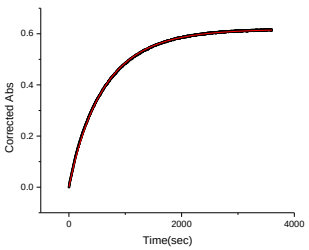


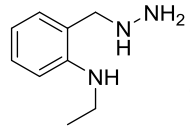


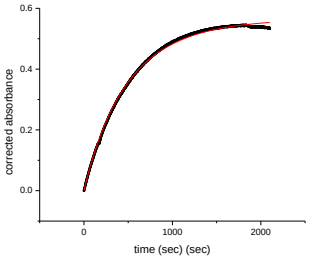
8.  (**8,** 306 nm)

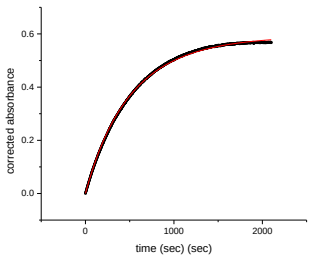


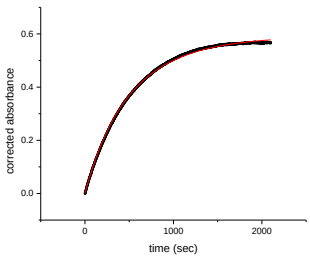


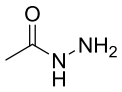


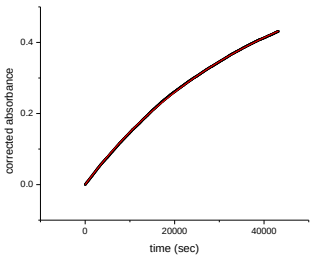
9.  (**9,** 304 nm)

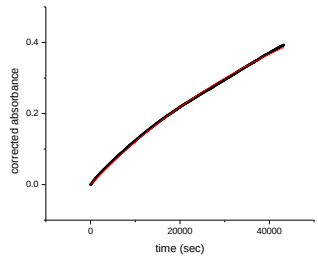


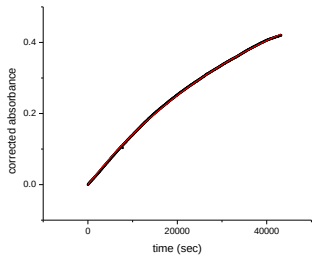


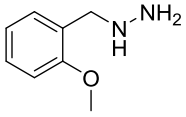


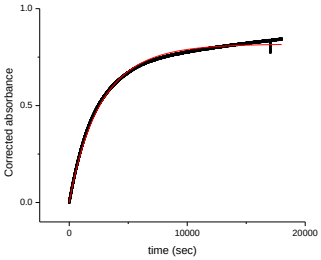
10.  (**10,** 290 nm)

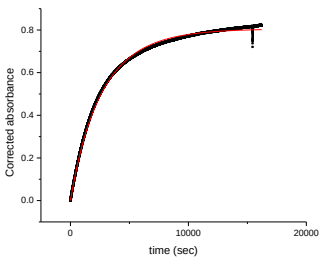


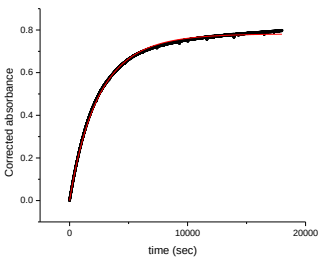




11.  (**11,** 300 nm)







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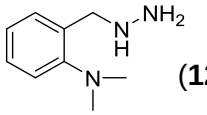
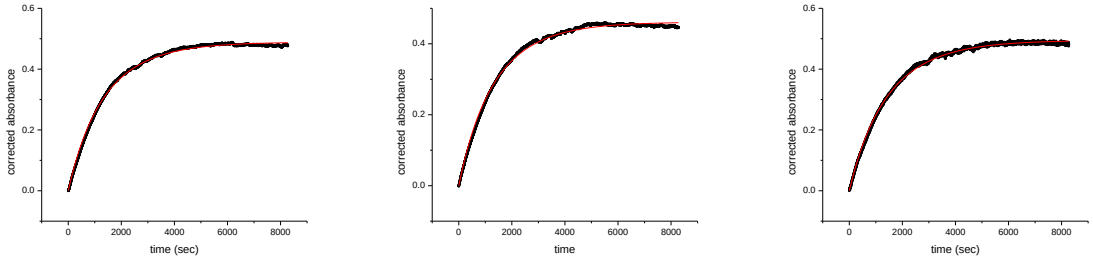


(**12,** 301 nm)



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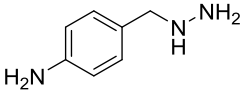
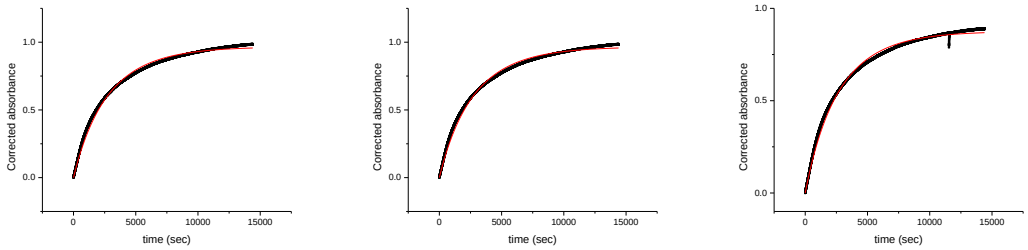


(**13,** 305 nm)



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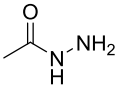
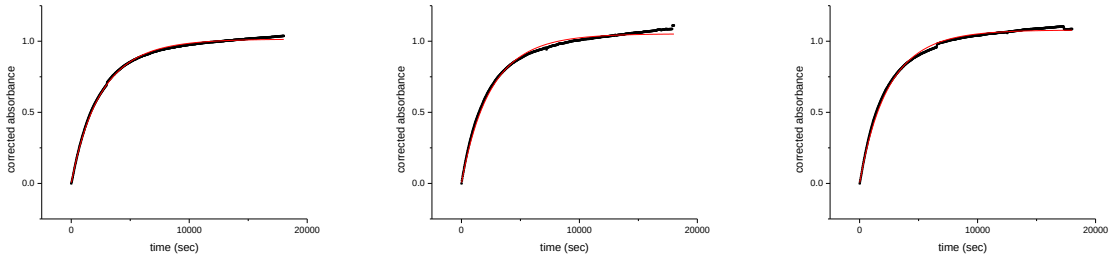


(**10,** 300 nm) + Aniline



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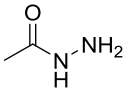
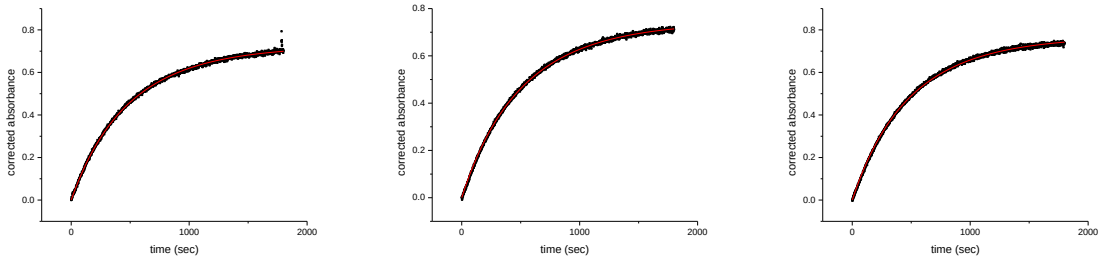


(**10,** 290 nm) + *p*-methoxy anthranilic acid



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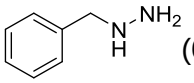
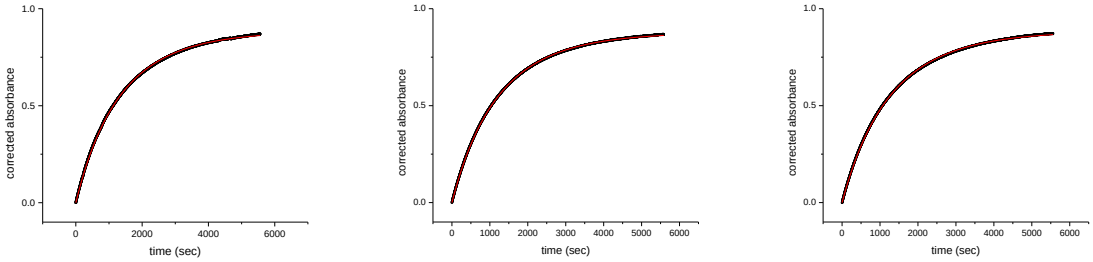
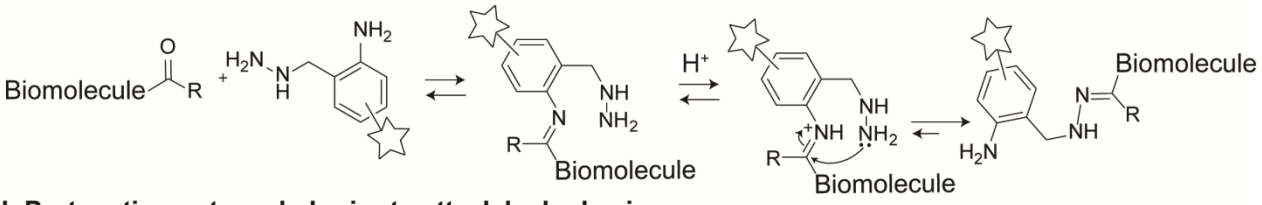


(**6,** 300 nm) + 250 µM Aniline



5. Attack of hydrazine on imine formed by compound 7 as compared to its attack on the imine formed by compound 9:

a Protonation needed prior to attack by hydrazine



b Protonation not needed prior to attack by hydrazine

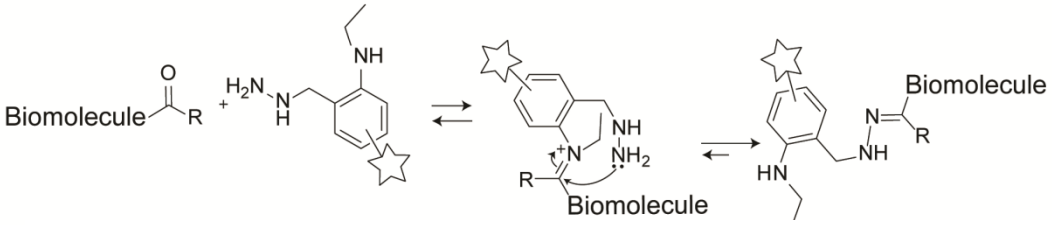


Figure S2. Intramolecular attack of hydrazine to protonated imine vs positively charged imine

6. Procedure for HPLC-based mechanistic studies:

A solution of 2-aminobenzhydrazide (200 µL, 120 mM in 5% acetonitrile/water) was added to a microfuge tube containing phosphate buffer (500 µL, 250 mM sodium phosphate, pH 4) and acetonitrile (100 µL). Subsequently, to this mixture was added a solution of benzaldehyde (200 µL, 12 mM in 5% acetonitrile in water). The resulting mixture was incubated at 37°C for 12 h and the reaction mixture was subjected to HPLC analysis by employing a linear gradient of water in acetonitrile containing 0.1% TFA. The details of the gradient are as follows: 100% water (0-5 min), 100-0% water (5-20 min), 0% water (20-25 min), 0-100% water (25-30 min). To a 500 µL aliquot of this reaction mixture was added a solution of $NaCNBH_3$ (100 µL, 600 mM in water). After incubating the mixture at 37°C for 15 h, it was subjected to HPLC analysis by employing the same gradient protocol mentioned above. The results of these experiments are depicted in Figure S3. The peaks obtained from the HPLC runs were collected and characterized by MALDI-TOF mass spectrometry (Table S1).

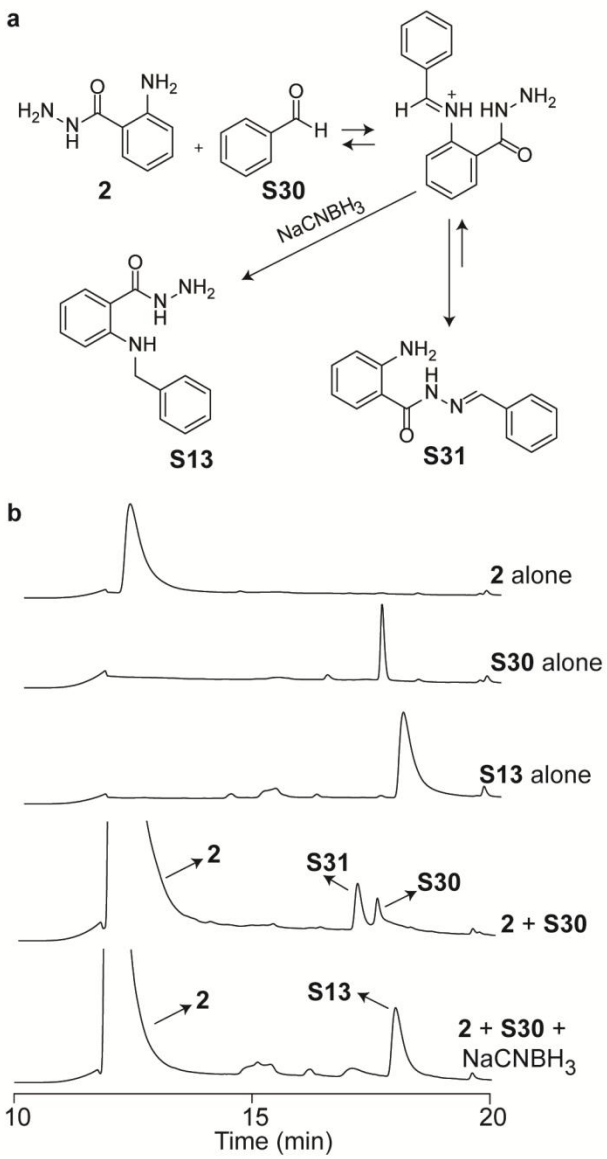


Figure S3. Mechanistic studies on hydrazone formation. a) Reaction scheme, and b) HPLC traces for the imine trapping experiment. Synthesis of pure standard **S13** is described in Scheme S6.

7. Intra vs intermolecular attack on imines formed by *o*-amino benzyl hydrazines:

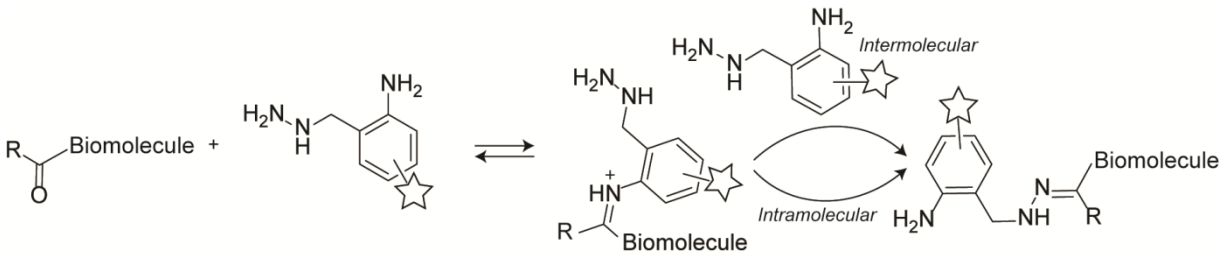
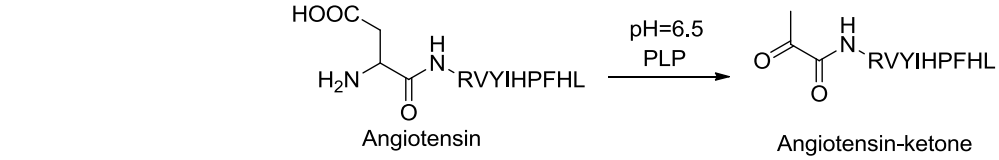


Figure S4. Two possible mechanisms for hydrazone formation by *o*-amino benzyl hydrazines

8. Bioconjugation studies on angiotensin:



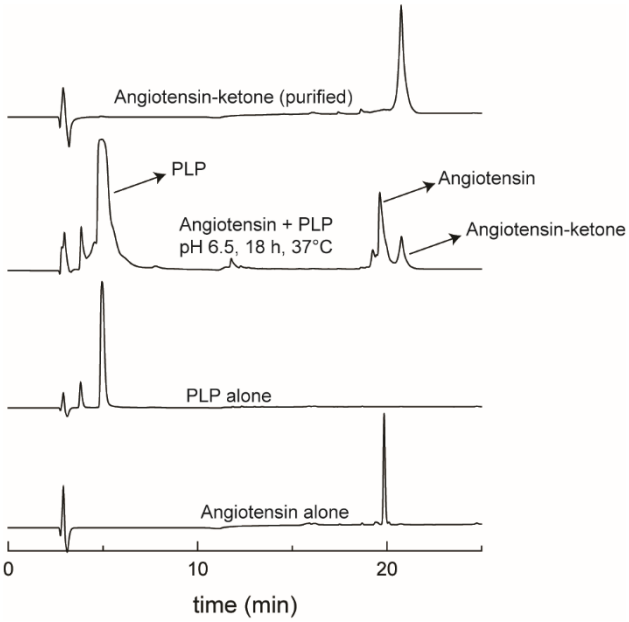


Figure S5. Synthesis of angiotensin-ketone

To a solution of angiotensin I (140 µL, 5 mM) in sodium phosphate buffer (50 mM at pH 6.5), was added sodium phosphate buffer (210 µL, 50 mM at pH 6.5) and pyridoxal 5′ phosphate (350 µL of a 20 mM solution in 50 mM sodium phosphate buffer at pH 6.5). After incubating the resulting mixture at 37°C for 18 h, it was subjected to HPLC by employing a linear gradient of water in acetonitrile containing 0.1% TFA. The details of the gradient are as follows: 100% water (0-5 min), 100-0% water (5-35 min), 0% water (35-40 min), 0-100% water (40-45 min). The peaks from the HPLC runs were collected and subjected to MALDI-TOF analysis resulting in the identification and purification of the desired angiotensin-ketone (Table S1). The relevant HPLC traces are shown in Figure S5.

Synthesis of fluorescent angiotensin conjugate:

To a solution of purified angiotensin-ketone (200 µL, 730 µM solution in water) were added sodium phosphate buffer (100 µL, 160 mM at pH 5) and compound **12** (100 µL, 14.6 mM solution in 20% acetonitrile-water). After incubating the resulting mixture at 37°C for 12 h, it was subjected to HPLC analysis by employing a linear gradient of water in acetonitrile containing 0.1% TFA. The details of the gradient are as follows: 80-20% water (0-30 min), 20-0% water (30-33 min), 0% water (33-37 min), 0-80% water (37-40 min) followed by MALDI-TOF analysis on collected fractions (Table S1). The HPLC trace thus obtained is depicted in Figure 3a (top panel).

Hydrolysis studies on fluorescent angiotensin conjugate:

The HPLC-purified fluorescent angiotensin conjugate (synthesis and purification described above) was dissolved in 20% acetonitrile in buffer (110 µL of acetonitrile dissolved in 440 µL of 160 mM sodium phosphate at pH 5) and 200 µL of this solution was immediately subjected to HPLC analysis to obtain a trace corresponding to the t = 0 h time point (Figure 3b, bottom panel). The solvent system employed was a linear gradient of water in acetonitrile containing 0.1% TFA. The details of the gradient are as follows: 80-20% water (0-30 min), 20-0% water (30-33 min), 0% water (33-37 min), 0-80% water (37-40 min). The remaining mixture was incubated at 37°C for 18 h and subsequently, 200 µL of the mixture was subjected to HPLC (gradient protocol was identical to the one used for the 0 h time point above) to obtain a trace corresponding to the t = 18 h time point (Figure 3b, top panel). The peaks from each of these runs were collected and characterized by MALDI-TOF (Table S1).

Comparison of reactivity of 6 and 9 with Angiotensin-ketone:

To a solution of purified angiotensin-ketone (100 µL, 300 µM solution in water) were added water (91.2 µL), acetonitrile (60 µL), sodium phosphate buffer (18.8 µL, 160 mM at pH 5) and compound **6** or **9** (30 µL, 10 mM solution in water). The resulting mixture was incubated at 37°C and 70 µL-aliquots were withdrawn periodically and subjected to HPLC analysis by employing a linear gradient of water in acetonitrile containing 0.1% TFA. The details of the gradient are as follows: 80-20% water (0-30 min), 20-0% water (30-33 min), 0% water (33-37 min), 0-80% water (37-40 min). The resulting peaks were characterized by MALDI-TOF mass spectrometric analysis (Table S1). The HPLC trace thus obtained is depicted in Figure S6a and S6b. The fraction of angiotensin-ketone consumed at each time-point was calculated by dividing the area under the peak for angiotensin-ketone obtained at that time-point with area under the peak of angiotensin-ketone obtained at time zero (before addition of the hydrazine).

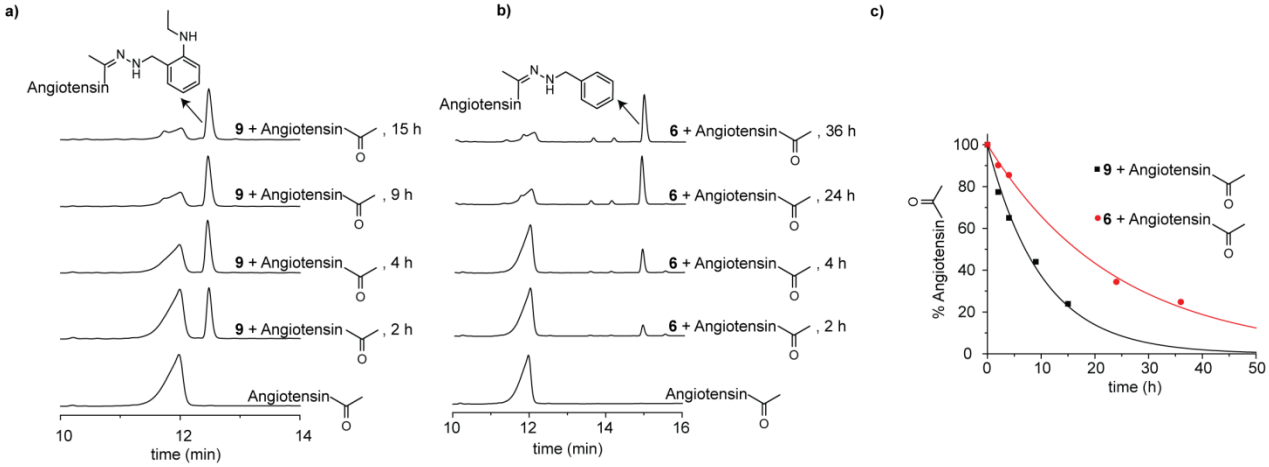


Figure S6: Comparison of reactivity of compounds **6** and **9** with angiotensin-ketone: a) HPLC traces for the reaction of compound **9** with angiotensin-ketone, b) HPLC traces for the reaction of compound **6** with angiotensin-ketone, c) Exponential decay curves for angiotensin-ketone representing reaction kinetics for both the reactions.

Comparison of Hydrolytic stability of hydrazone conjugates of 6 and 9 with angiotensin-ketone:

The HPLC-purified angiotensin conjugates of **6** and **9** (synthesis and purification described above) were dissolved in 20% acetonitrile in buffer (200 µL of acetonitrile dissolved in 800 µL of 160 mM sodium phosphate at pH 5) and 250 µL each of these solutions were immediately subjected to HPLC analysis to obtain a trace corresponding to the t = 0 h time point (Figure S7a and S7b, bottom panel). The solvent system employed was a linear gradient of water in acetonitrile containing 0.1% TFA. The details of the gradient are as follows: 80-20% water (0-30 min), 20-0% water (30-33 min), and 0 % water 36 (33-37

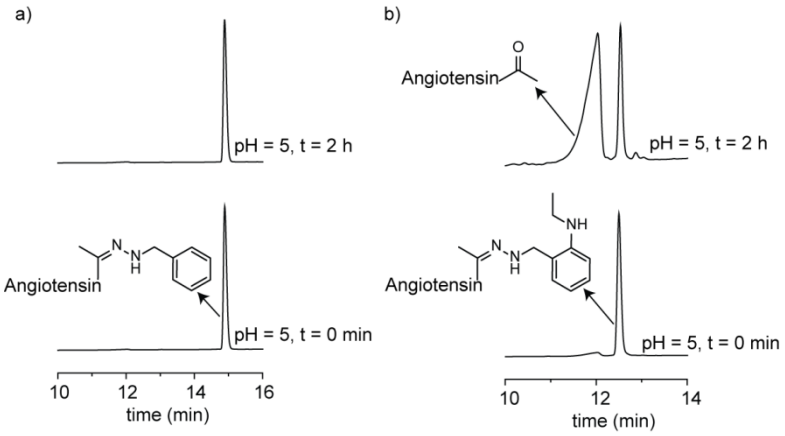
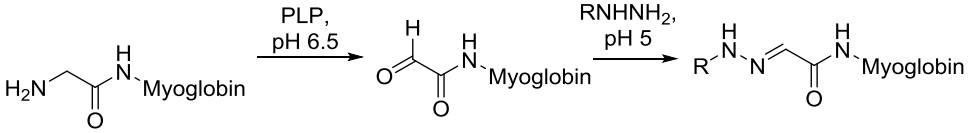


Figure S7. Hydrolytic stability of hydrazone bioconjugates: a) Stability of angiotensin hydrazone of **6**, b) Stability of angiotensin hydrazone of **9**

min), 0-80% water (37-40 min). The remaining mixtures were incubated at 37°C for 2 h and subsequently, 250 µL of the mixtures were subjected to HPLC (gradient protocol was identical to the one used for the 0 h time- point above) to obtain a trace corresponding to the t = 2 h time-point (Figure S7a and S7b, top panel). The peaks from each of these runs were collected and characterized by MALDI-TOF (Table S1).

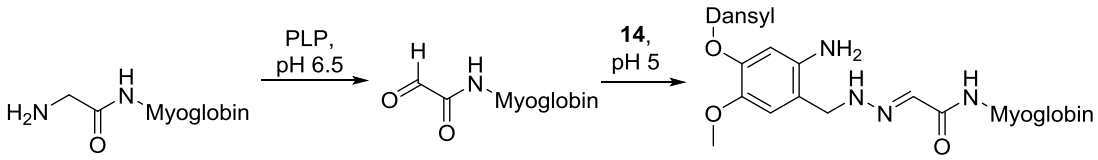
Procedure for bioconjugation of myoglobin:

a) With 8 and 9:



To a solution of myoglobin (144.3 µL of a 415.9 µM solution in 50 mM sodium phosphate buffer at pH 6.5), was added buffer (155.7 µL of 50 mM sodium phosphate at pH 6.5) and a solution of pyridoxal 5′- phosphate (300 µL of 40 mM solution in 50 mM sodium phosphate buffer at pH 6.5). The mixture was incubated at 37°C without agitation for 18 h. Subsequently the resulting myoglobin-CHO was buffer exchanged in to 27 mM Na_3PO_4 at pH 5 using 7K MWCO zeba spin column. To an aliquot of this buffer-exchanged myoglobin-CHO (37.5 µL) was added a solution of hydrazines (**8** or **9)** (12.5 µL, 6 mM solution in 80% acetonitrile-water). The resulting mixtures were incubated at 37°C for 24 h and subjected to MALDI-TOF analysis.

b) With 14:



To a solution of myoglobin (178.4 µL of a 280.2 µM solution in 50 mM sodium phosphate buffer at pH 6.5), was added buffer (321.6 µL of 50 mM sodium phosphate at pH 6.5) and a solution of pyridoxal 5′- phosphate (500 µL, 20 mM solution in 50 mM sodium phosphate buffer at pH 6.5). Simultaneously, a control reaction was started wherein instead of 500 µL of pyridoxal 5′-phosphate solution, buffer (500 µL of 50 mM sodium phosphate at pH 6.5) was added. Both the mixtures were incubated at 37°C without agitation for 18 h. Subsequently, both reaction mixtures were dialyzed against water at 4°C using snake skin dialysis tubing (3.5 KDa M.W.C.O, Thermo Fisher Scientific). To an aliquot of each of these dialyzed reaction mixtures (300 µL) was added buffer (60 µL of 160 mM sodium phosphate at pH 5) and a solution of **12** (40 µL, 18.75 mM solution in DMSO). Both of the resulting mixtures were incubated at 37°C for 24 h and then diluted by adding water (600 µL each) before being dialyzed against water at 4°C by using snake skin dialysis tubing (3.5 KDa M.W.C.O, Thermo Fisher Scientific). The dialyzed solutions were concentrated down to 140 µL by using a vacuum concentrator and subjected to SDS-PAGE on a 15% polyacrylamide gel. The gel was imaged with a fluorimager and then stained with coomassie (Figure 3d).

9. Cell culture:

HEK293 cells were cultured in DMEM medium supplemented with 10% FBS,100 units/ml penicillin, and 100µg/ml streptomycin in a humidified CO_2 incubator with 5% CO_2 at 37°C.

10. Metabolic labeling of choline lipids of HEK293 cells with keto-choline (15) and characterization by lipidomics:

HEK293 cells (0.2 million cells per mL) were grown in 10 cm dishes overnight and incubated with **15** (5 mM) for 24 h under cell culture conditions for metabolic labeling. After the incubation, cells were washed with PBS (2 × 10 mL) and scraped into cold PBS (5 mL) for lipid extraction. Cells were pelleted by centrifugation (1000 g for 5 min at 4°C), and the total cellular lipids were extracted by methanol-chloroform extraction.[4] Extracted lipids were then dried under reduced pressure for 1 h at room temperature. After drying, lipids were suspended in chloroform (750 µL) and stored at -80°C. For mass spectrometric analysis, the lipid solution (40 µL) was mixed with chloroform (320 µL), methanol (798 µL), and ammonium acetate (42 µL of a 300 mM solution in water) and directly infused (10 µL/min) into a triple quadrupole mass spectrometer (QTRAP® 4500 system from AB SCIEX). Precursor Ion Scan (PIS) in the positive scan mode was employed to identify the precursor of 254.1 corresponding to the phosphocholine derivative of **15**. Scanning speed was set at 200 Da/S, and the electron spray voltage was maintained at +5500 V. Declustering potential, entrance potential, collision energy, and exit potential were 163.0 V, 8.0 V, 42.10 V, and 31.15 V respectively. Mass spectrum obtained from the 'Analyst' software was fed into 'Lipidview' software to identify choline lipids labeled with **15**.

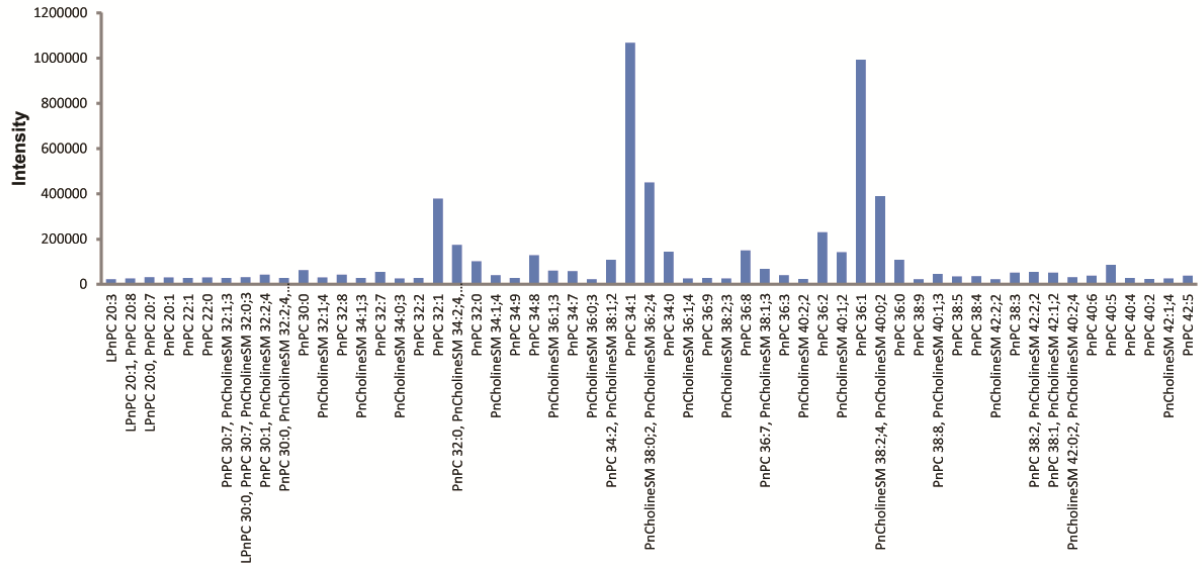


Figure S8: Choline lipids containing phosphocholine derivative of **15**. LPnPC is an abbreviation for lysophosphatidylcholine lipids labeled with **15**, PnPC is an abbreviation for phosphatidylcholine lipids labeled with **15**, and PnCholineSM is an abbreviation for sphingomyelin lipids labeled with **15**.

11. Imaging of choline lipids in HEK293 cells metabolically labeled with 15 via hydrazone bioconjugation:

HEK293 cells (0.05 million cells per mL) grown on cover slips were incubated with **15** (5 mM) for 24 h in cell culture conditions. Cells were washed twice with PBS (1 mL) and incubated with **16** (100 µM) in sodium acetate buffer of pH 4.7 (1 mL) for 60 min at 4°C. Cells were then washed with PBS (2 X 1 mL) to remove unreacted **16** and this was followed by fixation at -20°C in 70% ethanol for 10 min. Cells were washed again in PBS (1 mL) and counterstained with Hoechst stain (1 µg/mL) for 5 min at room temperature. Cells were then washed serially each with 1 mL of TBS (tris buffered saline), 0.5 M NaCl, TBS and imaged by confocal laser scanning microscope.

12. Catalyst-free reversible fluorescent labelling of sialylated glycoproteins on HEK293 cells via hydrazone bioconjugation:

HEK293 cells (0.1 million cells per mL) grown on coverslips were incubated with $NaIO_4$ (1 mM in PBS) for 5 min at 4°C to perform the oxidation of sialic acids on the cell surface. This periodate oxidation reaction was quenched with glycerol (1 mM in PBS) for 5 min at room temperature. Subsequently cells were washed with PBS (3 X 1 mL) and incubated with **16** in PBS (100 µM in PBS of pH 6.4 for 90 min at 4°C. PBS wash, ethanol fixation, Hoechst counterstaining, and confocal imaging were carried out as described in section 12. The same protocol was followed for the preparation of control samples, except they were not incubated with $NaIO_4$ (1 mM) prior to treatment with **16.** For hydrolysis and transimination experiments, **16** treated cells were incubated in hydroxylamine (10 mm in PBS) for 60 min at 4°C or in sodium acetate buffer (100 mM, pH 4.0) for 60 min at 4°C (buffer replaced at every 15 min time intervals). After this incubation, cells were fixed by ethanol, counter stained with Hoechst stain, and imaged by confocal microscopy.

13. Cell viability assay:

HEK293 cells (5000 cells/well in 200 µL DMEM media) were seeded in a 96 well plate and incubated overnight under cell culture conditions for cell attachment. Subsequently, cells were incubated separately with 10 mM and 50 mM of p-methoxy anthranilic acid (PMA), and 100 µM and 500 µM of compound **7**, **8**, **9** and **16** for 90 min at room temperature. Solutions of varying concentrations of **7**, **8** and **9** were made in DMEM media, and that of **16** and p-methoxy anthranilic acid (PMA) were prepared in DMEM media containing 0.2% DMSO. Cells incubated with only DMEM media and DMEM media containing 0.2% DMSO were taken as controls. After the incubation, the culture media was replaced with 180 µL of fresh DMEM media + 20 µL of MTT (3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyl tetrazolium bromide) solution (3 mg/mL in PBS, filtered through 0.22-micron syringe filter) and the cells were incubated for 3 h under cell culture conditions. After the incubation, the culture media was removed completely and DMSO (200 µL) was added to solubilize the formazan crystals formed by the reduction of MTT in live cells. The intensity of the resulting purple color was recorded at a wavelength of 570 nm by using a plate reader (Varioskan Flash, Thermo Scientific). Cell viability graph was plotted from the relative optical density of treated samples to the corresponding control data (which is taken as 100%). Each value on the graph corresponds to the average of 4 technical replicates.[5]

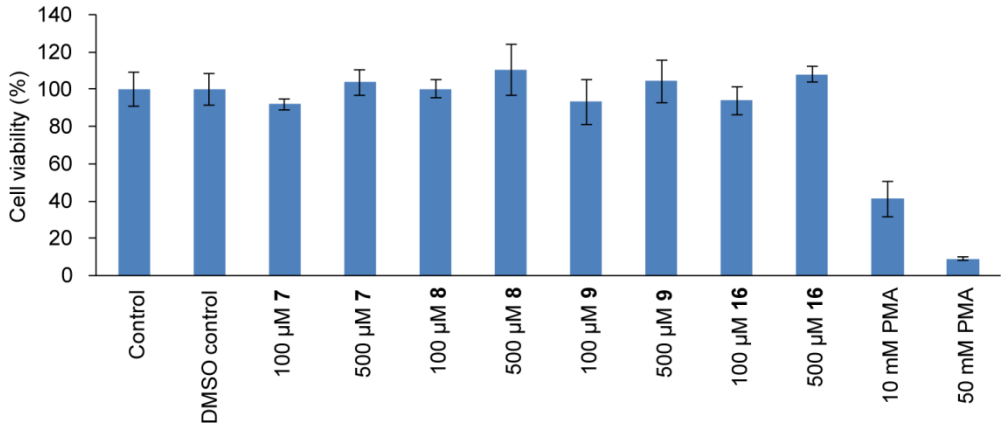
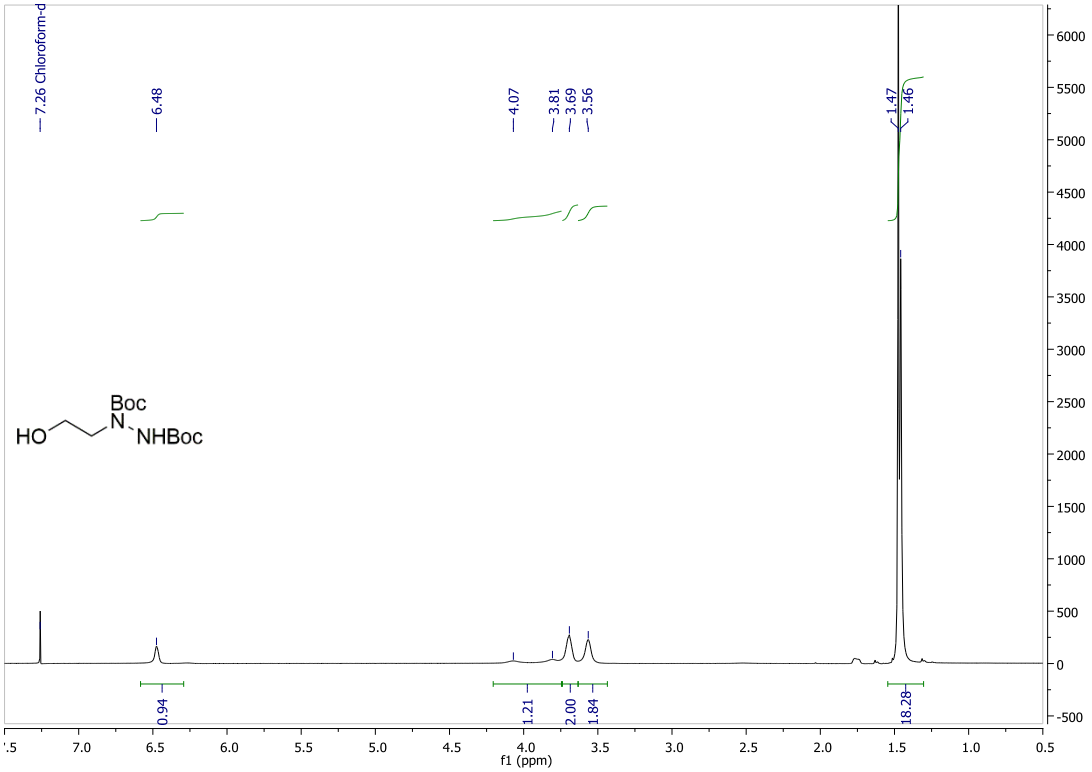
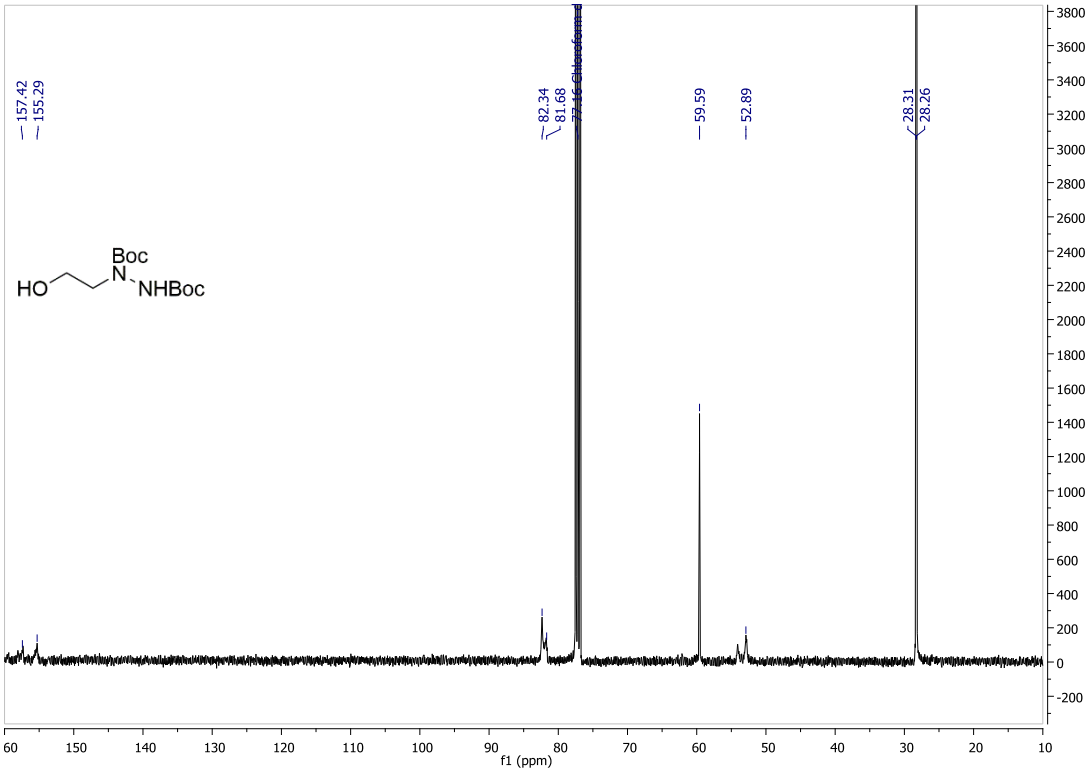
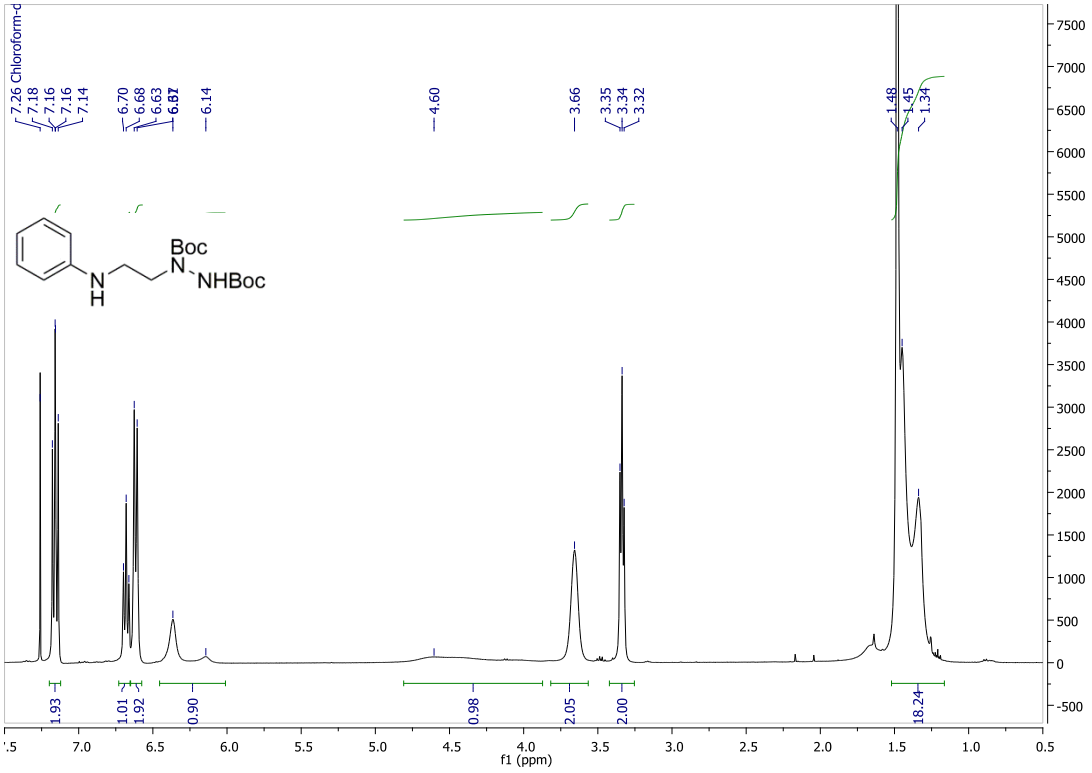


Figure S9: Cell viability assay of various *o*-amino benzyl hydrazines and *p*-methoxy anthranilic acid (PMA). HEK293 cell viability estimated after 90 min incubation with *p*-methoxy anthranilic acid (PMA) at 10 mM and 50 mM and **7**, **8**, **9**, and **16** at 100 µM and 500 µM.

14. NMR spectra



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4.60
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3.32
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1.34
Boc
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1.92
0.90
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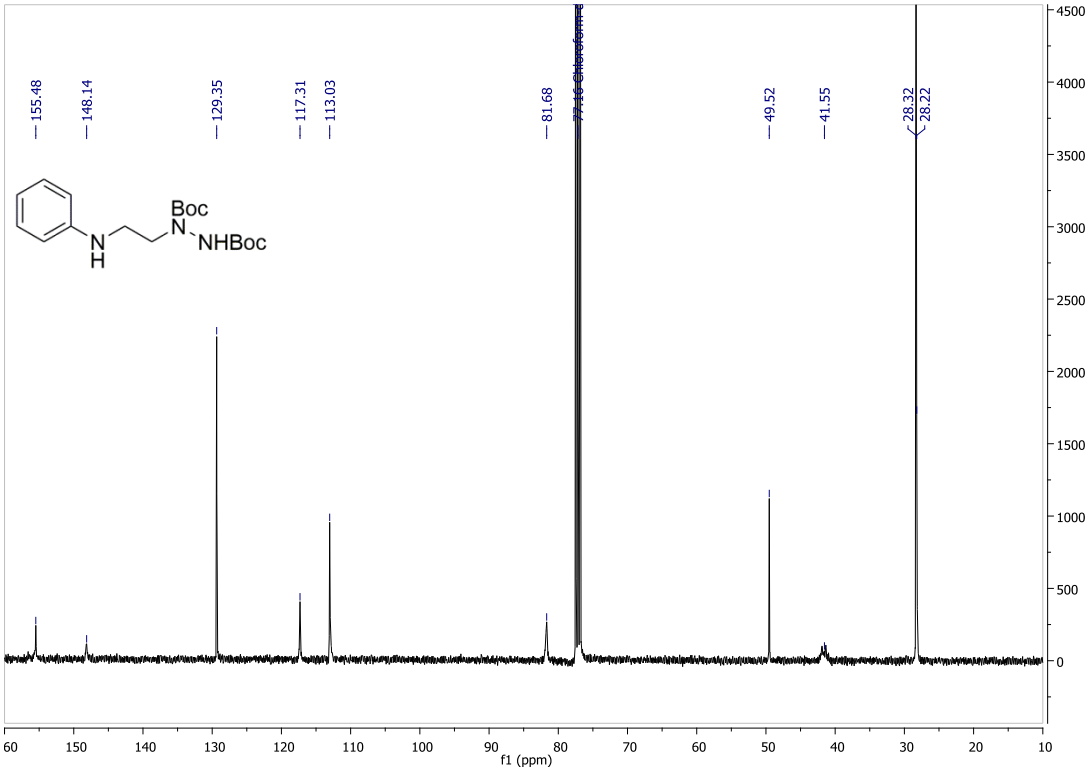
f1 (ppm)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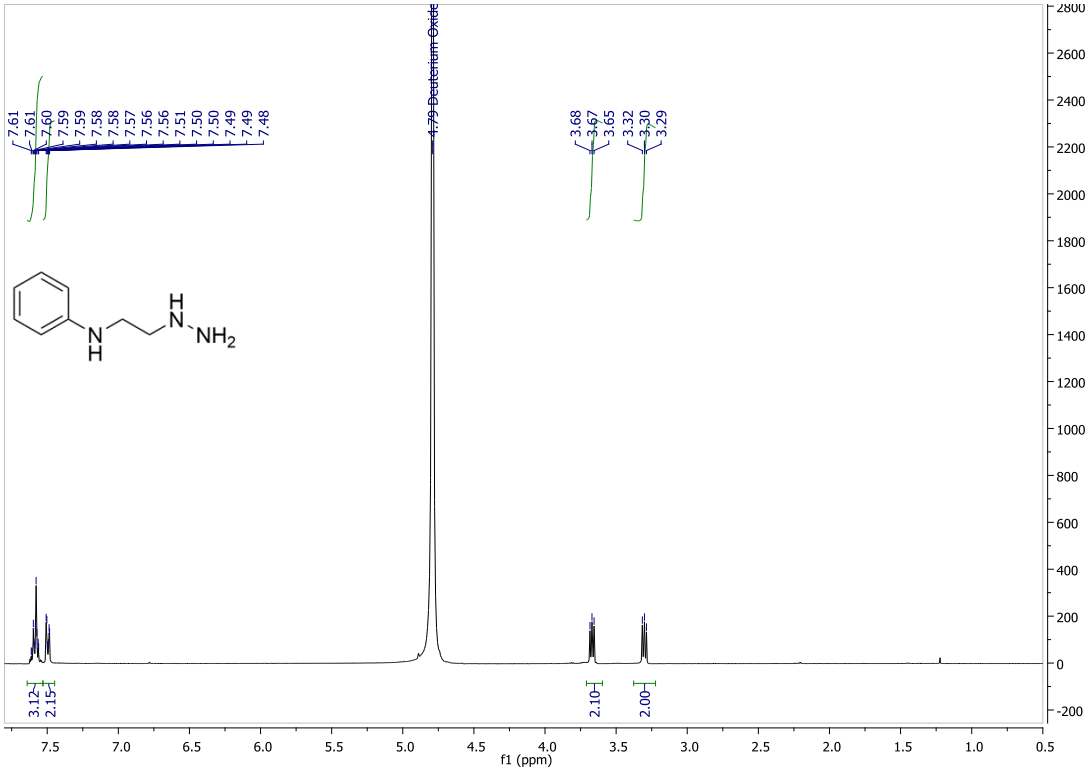
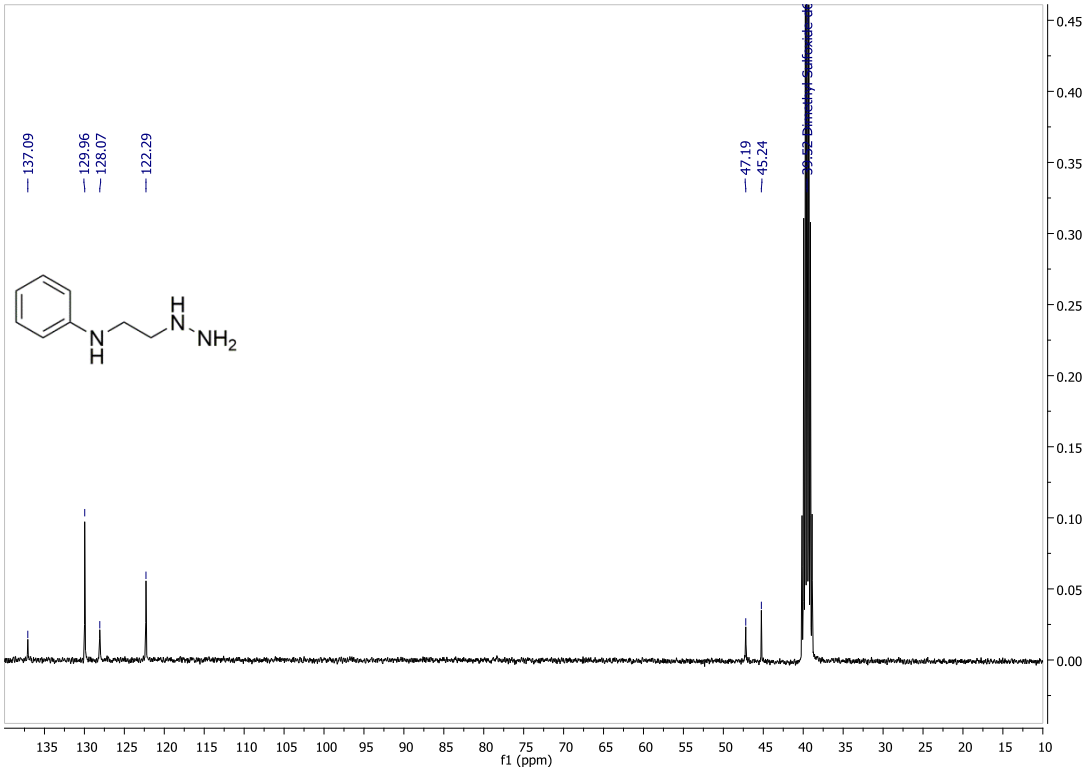
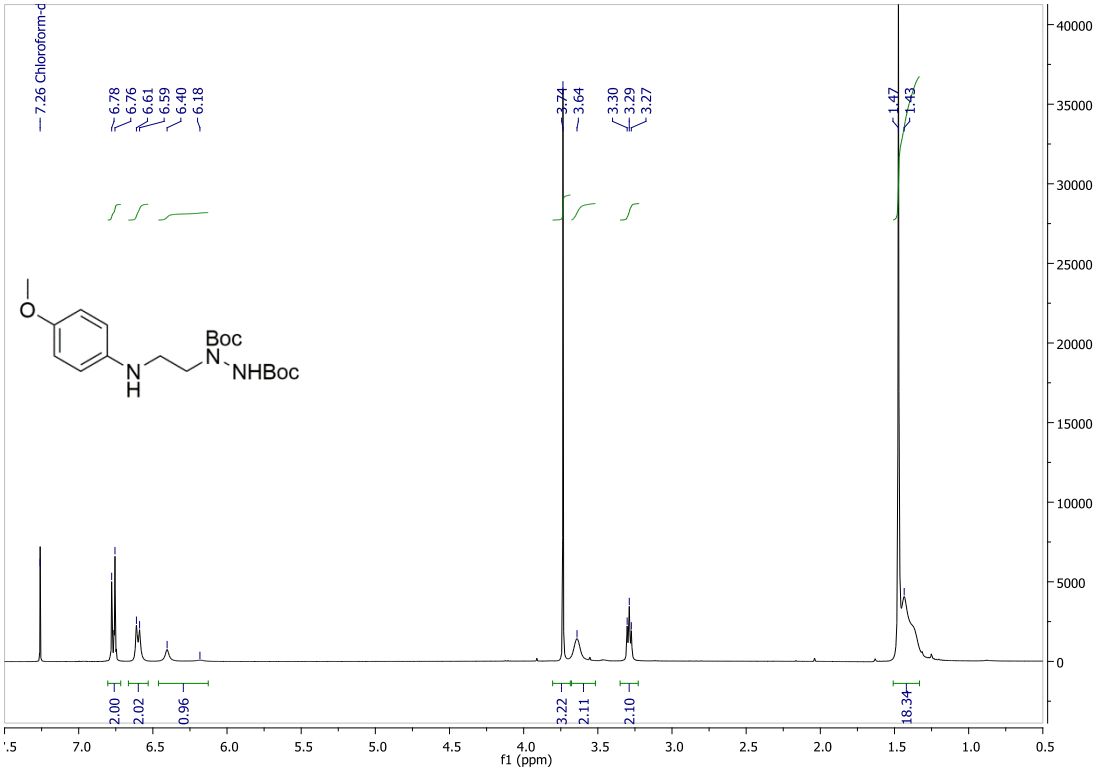
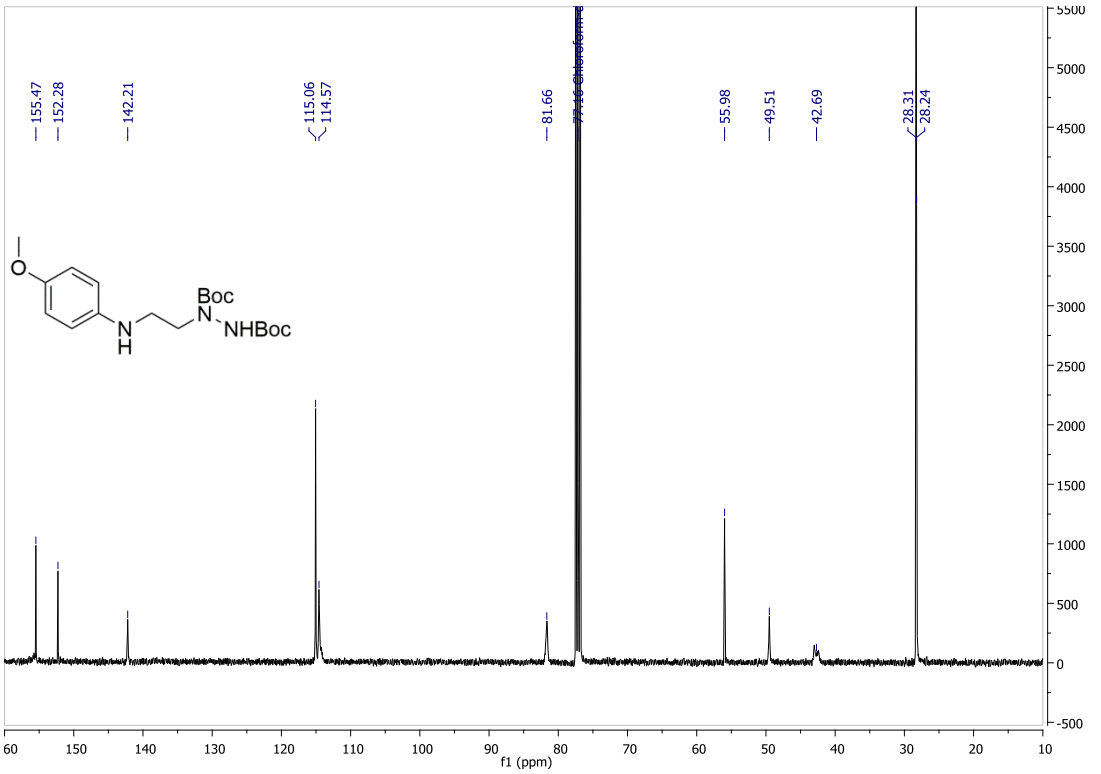
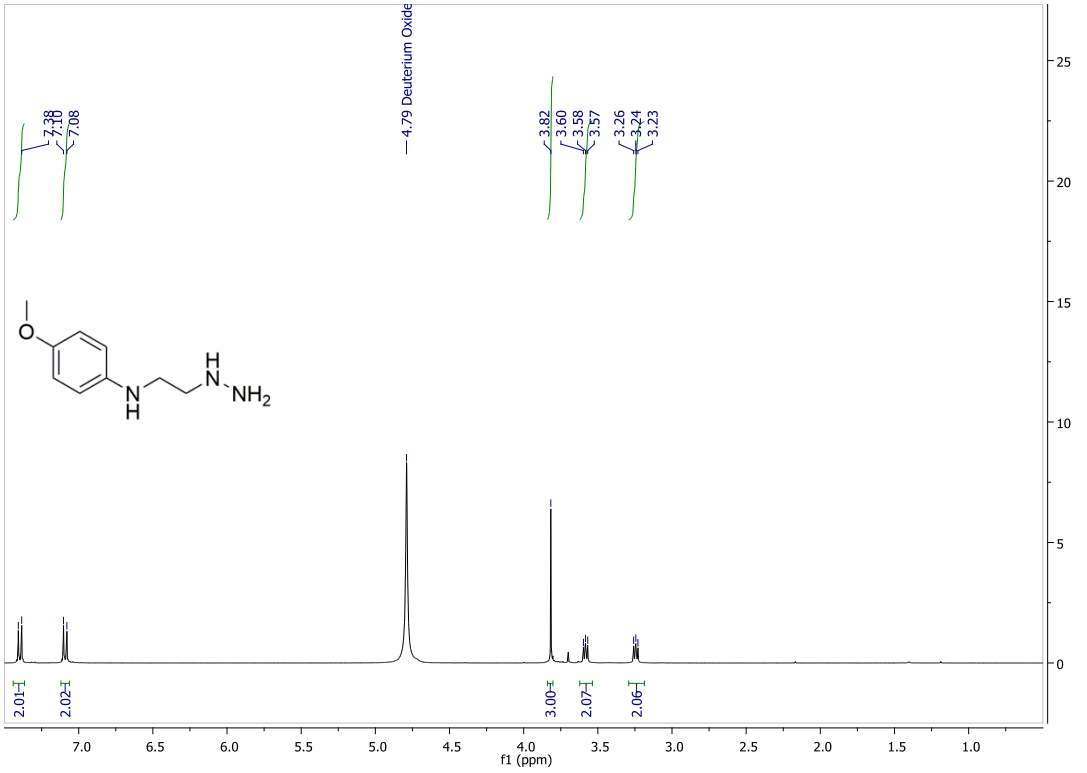
49.52
41.55
28.32
28.22
Boc
N
NHBoc
N
H
f1 (ppm)

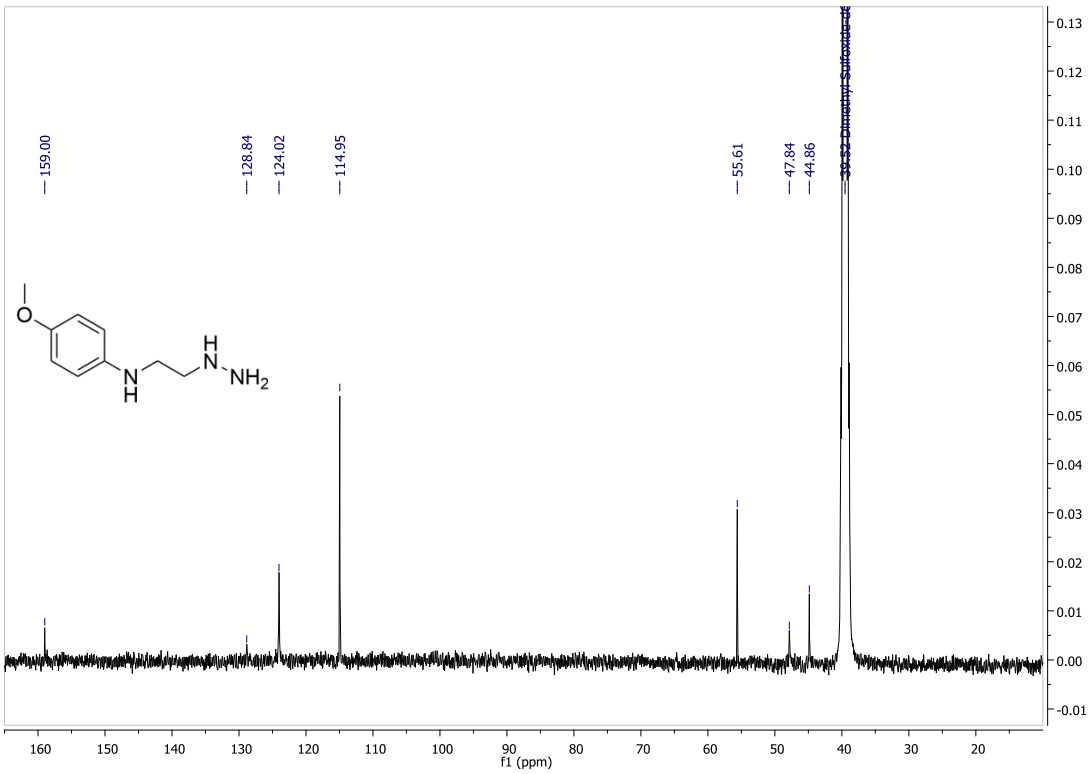

7.61
7.61
7.60
7.59
7.59
7.58
7.58
7.57
7.56
7.56
7.51
7.50
7.50
7.49
7.49
7.48
4.79 Deuterium Oxide
3.68
3.67
3.65
3.32
3.30
3.29
3.12
2.15
2.10
2.00
f1 (ppm)


137.09
129.96
128.07
122.29
47.19
45.24
39.52 Dimethyl Sulfoxide-d6
f1 (ppm)


7.26 Chloroform-d
6.78
6.76
6.61
6.59
6.40
6.18
3.74
3.64
3.30
3.29
3.27
1.47
1.43
Boc
N
NHBoc
N
H
O
2.00
2.02
0.96
3.22
2.11
2.10
18.34
f1 (ppm)


155.47
152.28
142.21
115.06
114.57
81.66
77.16 Chloroform-d
55.98
49.51
42.69
28.31
28.24
Boc
N
NHBoc
N
H
O
f1 (ppm)


4.79 Deuterium Oxide
7.38
7.10
7.08
3.82
3.60
3.58
3.57
3.26
3.24
3.23
2.01
2.02
3.00
2.07
2.06
f1 (ppm)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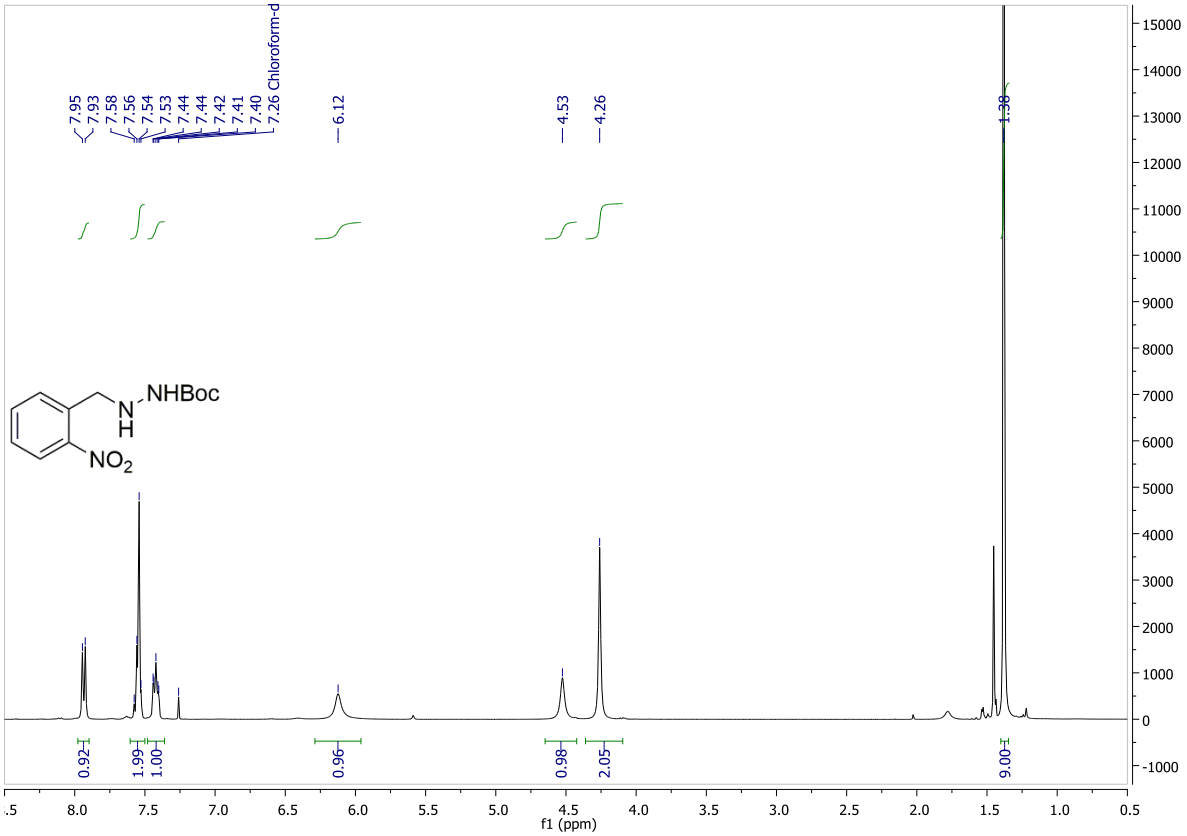
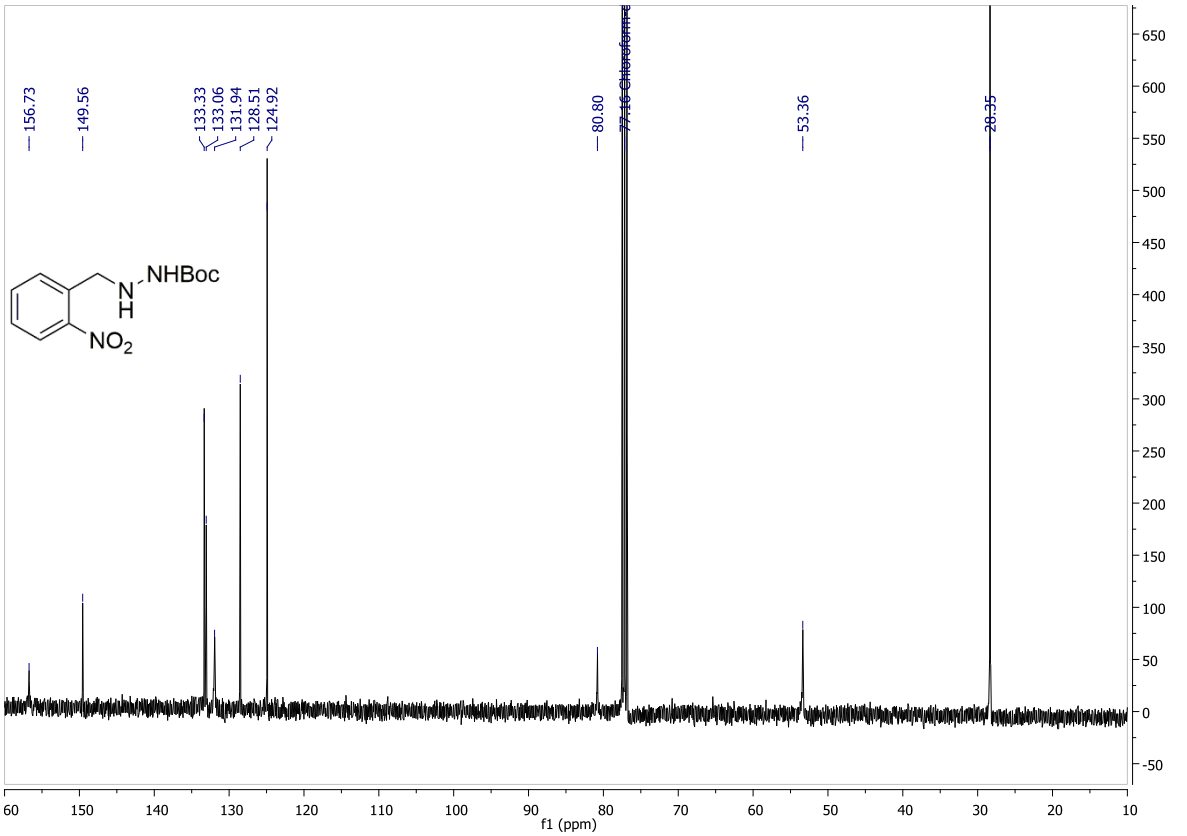
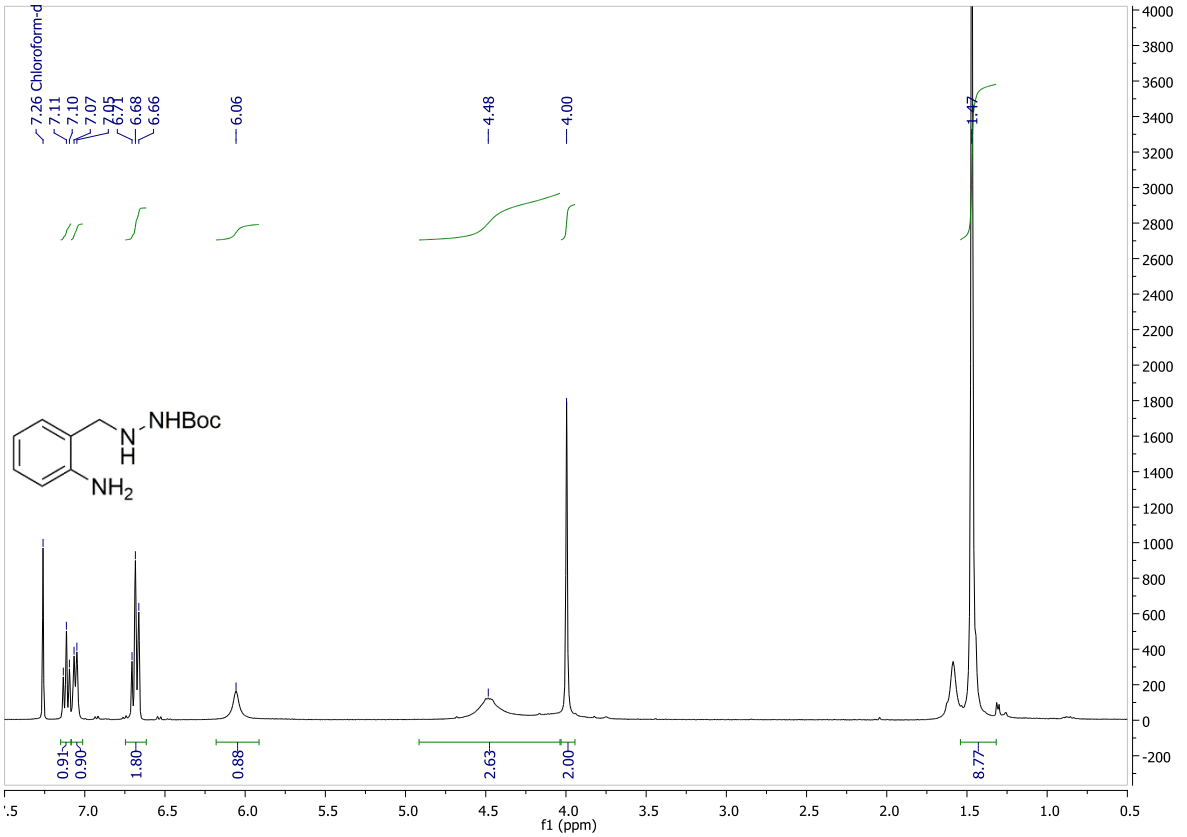
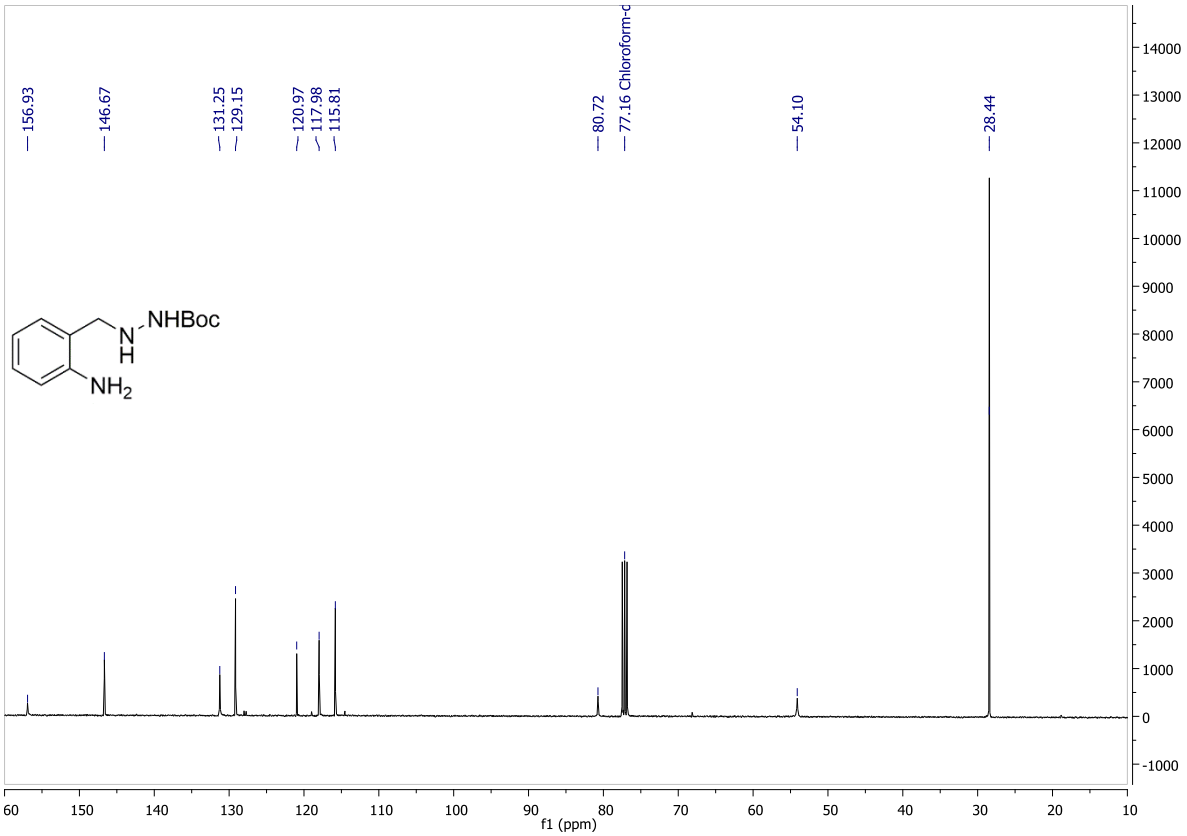
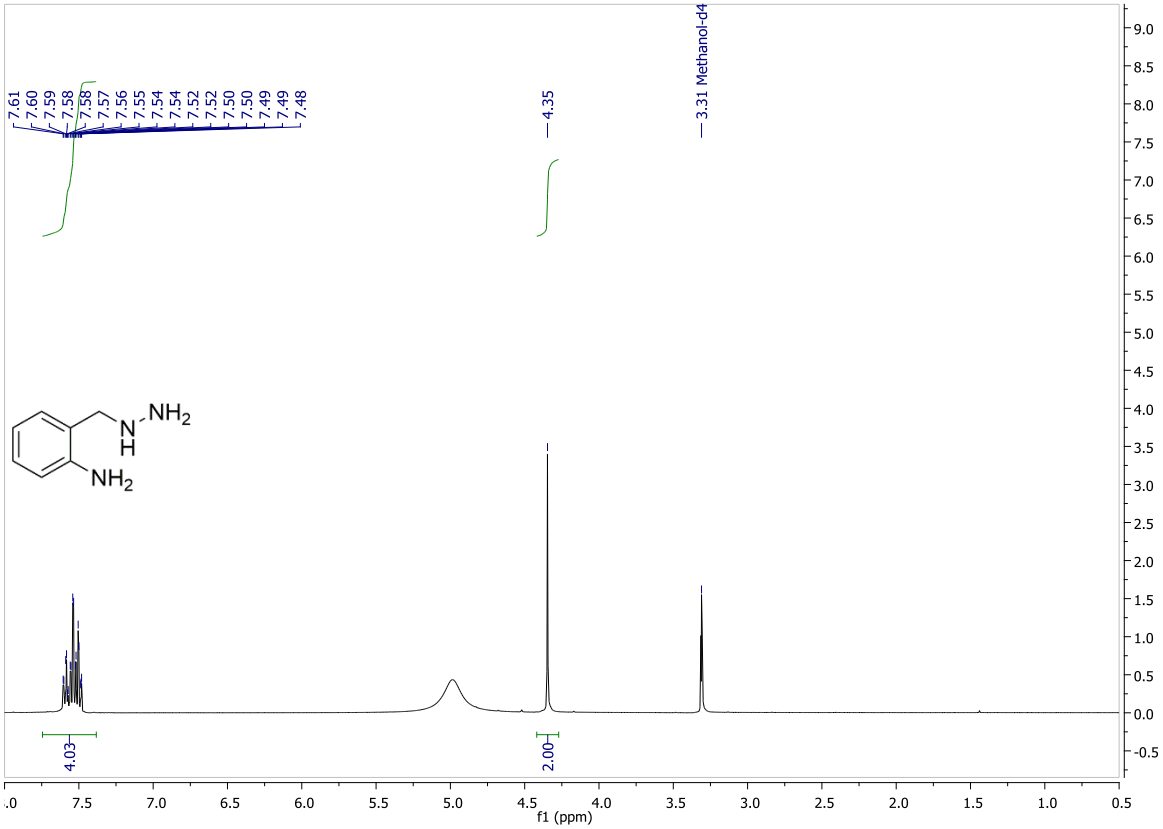
128.84
124.02
114.95
55.61
47.84
44.86
f1 (ppm)


7.95
7.93
7.58
7.56
7.54
7.53
7.44
7.44
7.42
7.41
7.40
7.26 Chloroform-d
6.12
4.53
4.26
1.38
NHBoc
N
H
NO2
0.92
1.99
1.00
0.96
0.98
2.05
9.00
f1 (ppm)


156.73
149.56
133.33
133.06
131.94
128.51
124.92
80.80
77.16 Chloroform-d
53.36
28.35
NHBoc
N
H
NO2
f1 (ppm)


7.26 Chloroform-d
7.11
7.10
7.07
6.95
6.71
6.68
6.66
6.06
4.48
4.00
1.47
NHBoc
NH2
0.91
0.90
1.80
0.88
2.63
2.00
8.77
f1 (ppm)


156.93
146.67
131.25
129.15
120.97
117.98
115.81
80.72
77.16 Chloroform-d
54.10
28.44
NHBoc
NH2
f1 (ppm)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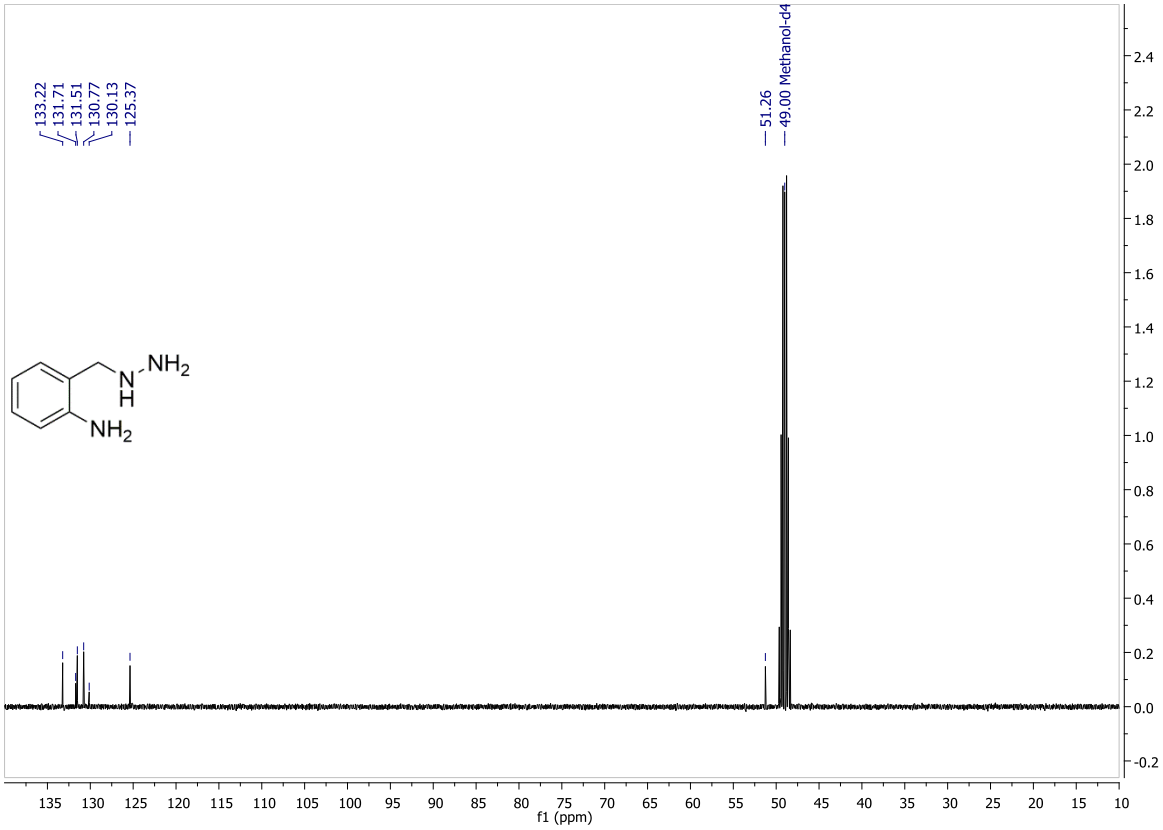
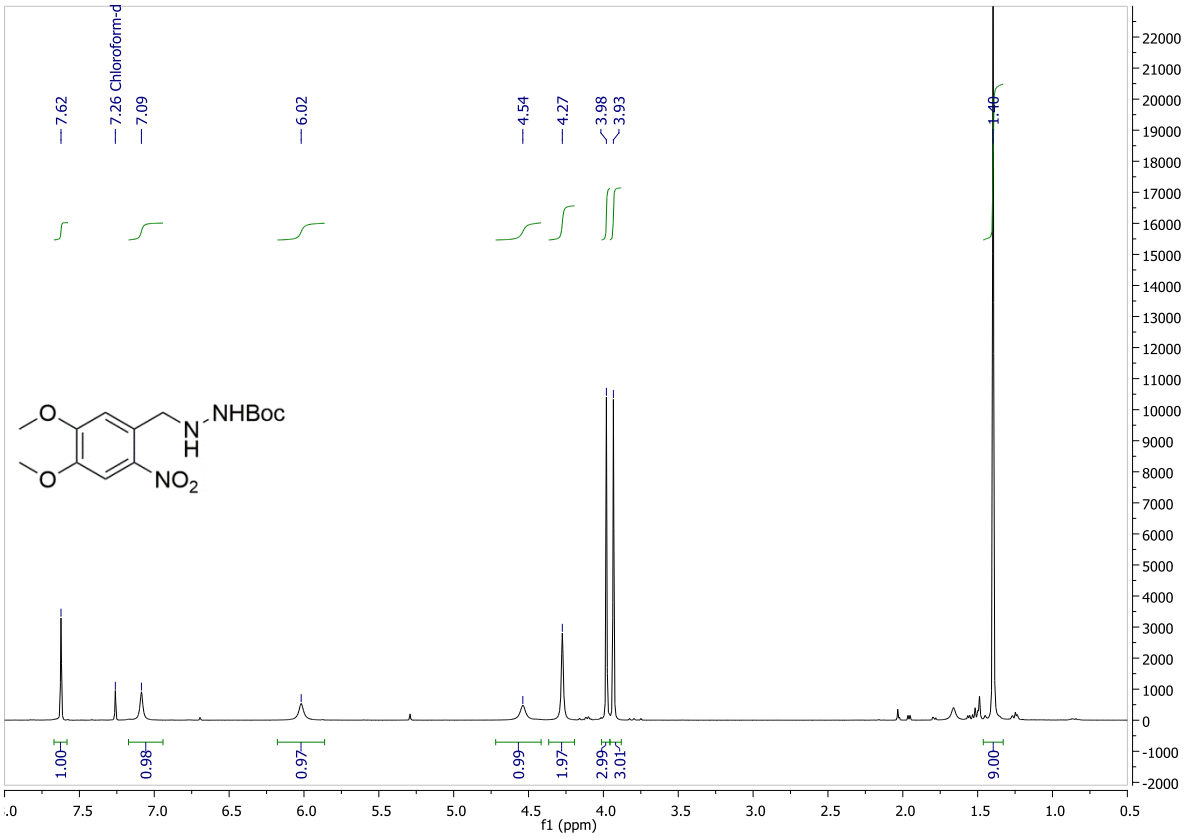
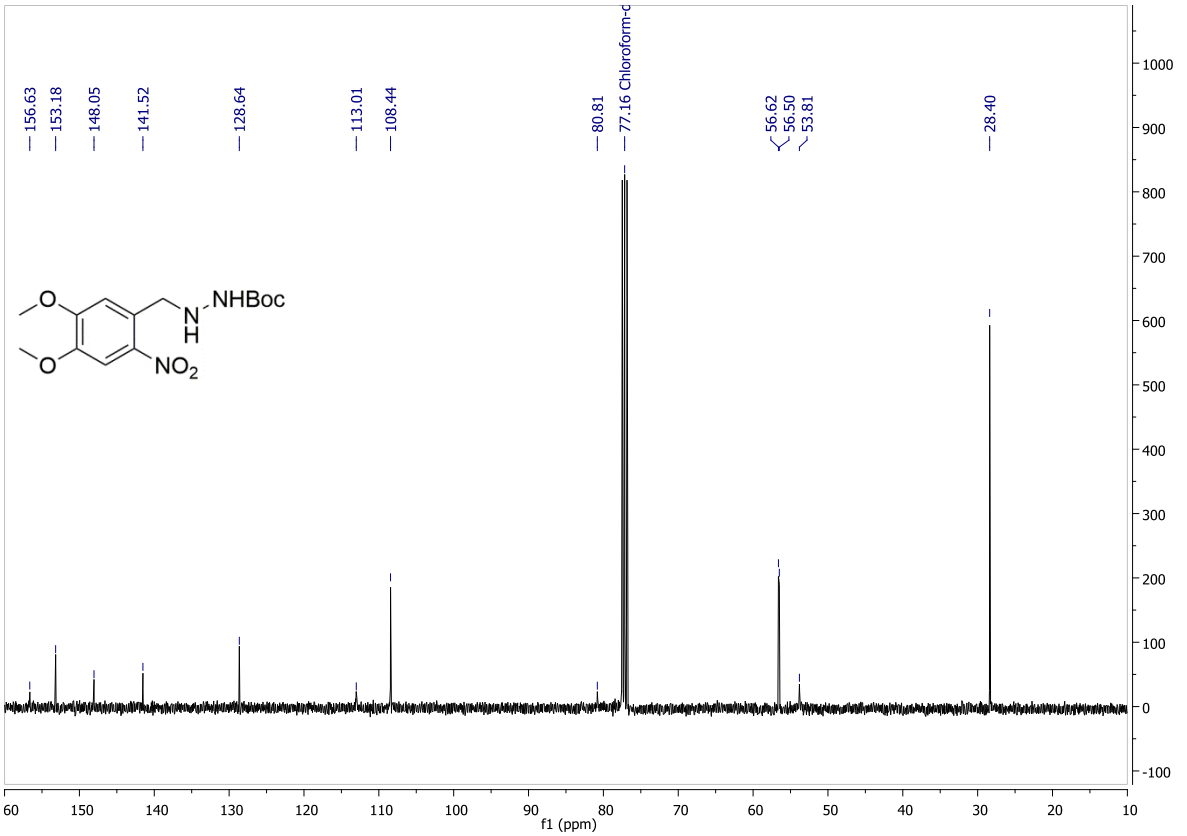
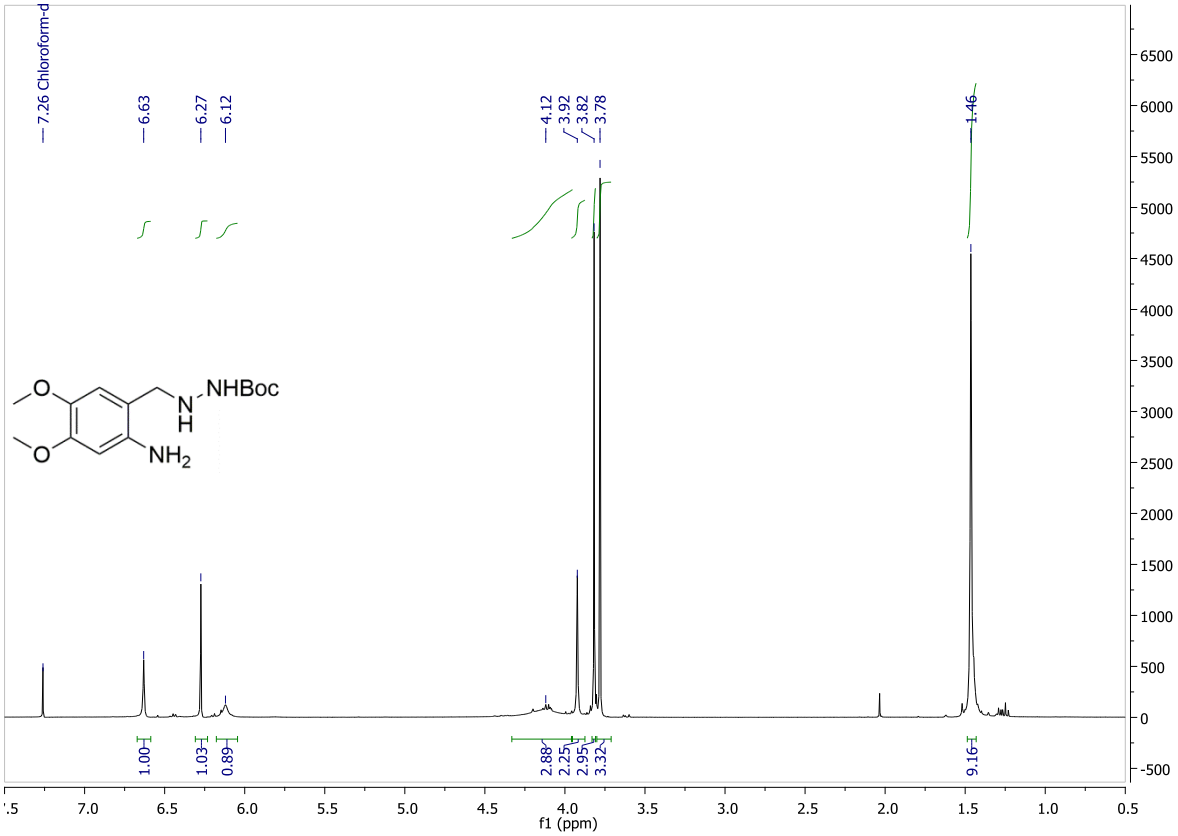
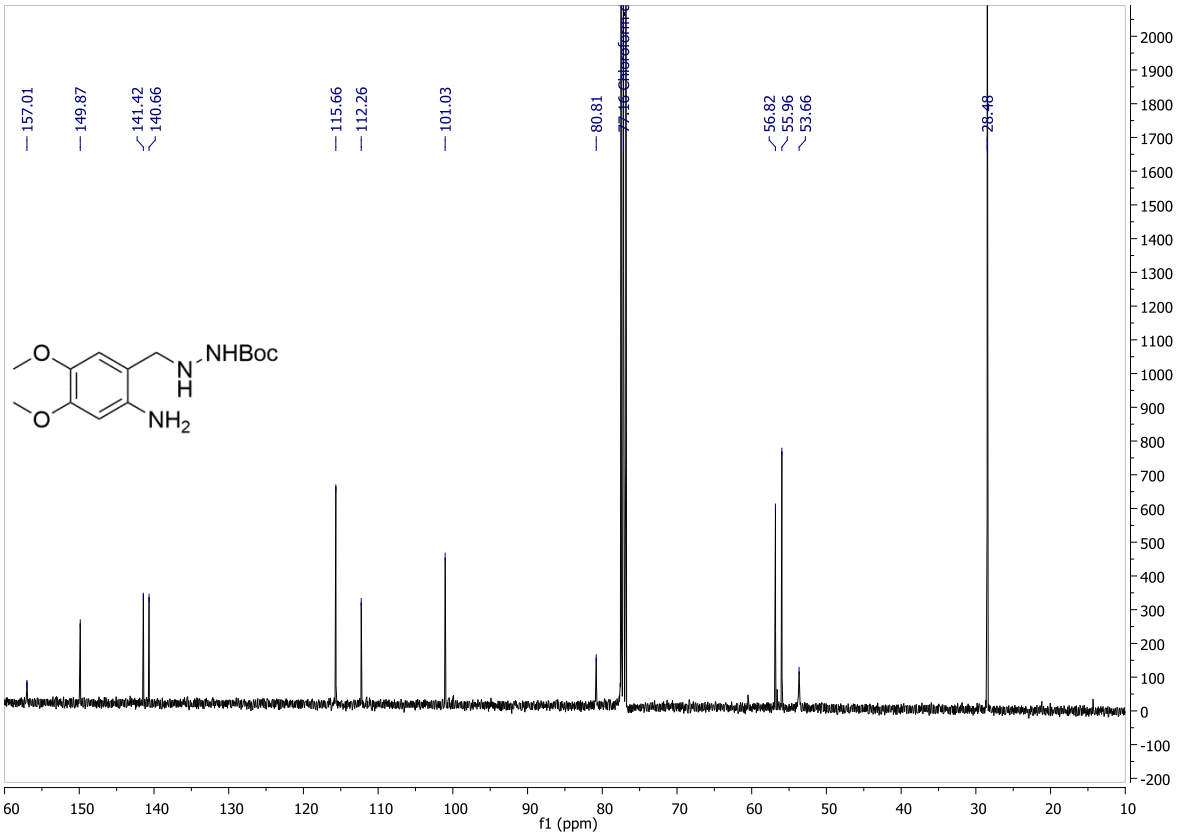
7.60
7.59
7.58
7.58
7.57
7.56
7.55
7.54
7.54
7.52
7.52
7.50
7.50
7.49
7.49
7.48
4.35
3.31 Methanol-d4
NH2
N
H
NH2
4.03
2.00
f1 (ppm)

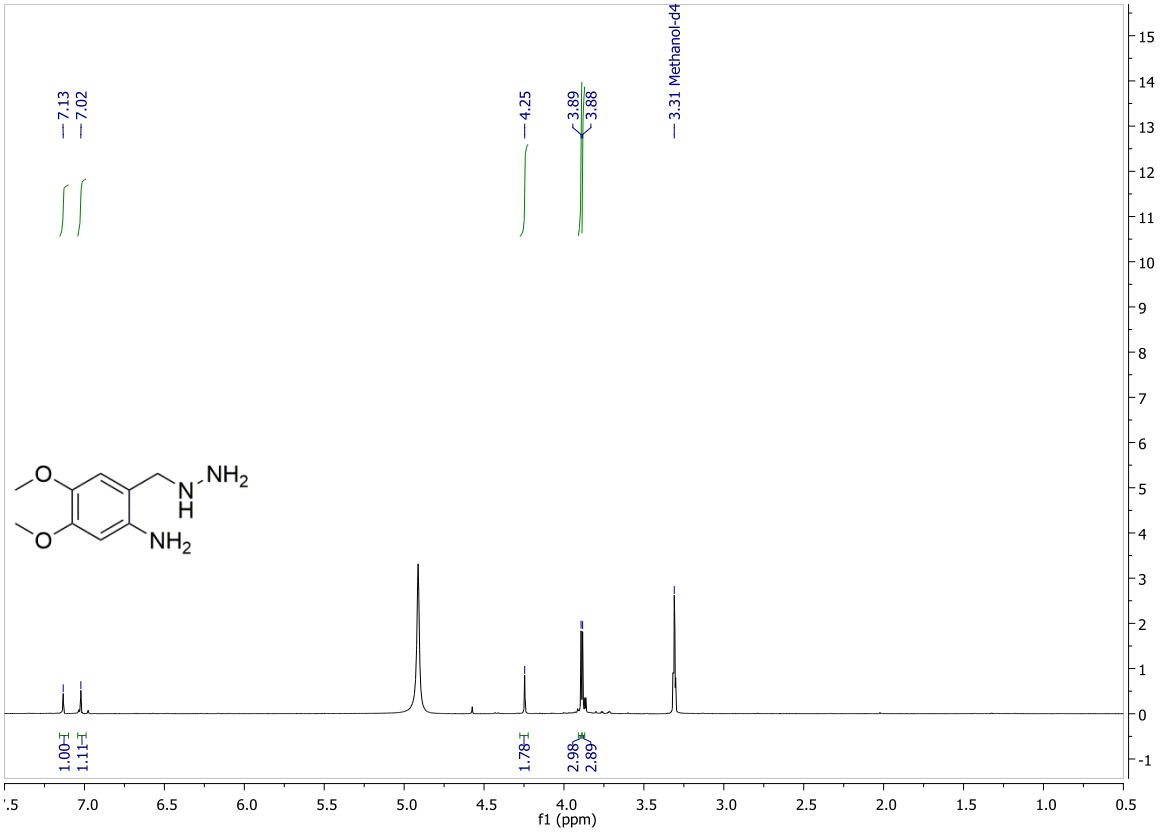

133.22
131.71
131.51
130.77
130.13
125.37
51.26
49.00 Methanol-d4
NH2
N
H
NH2
f1 (ppm)

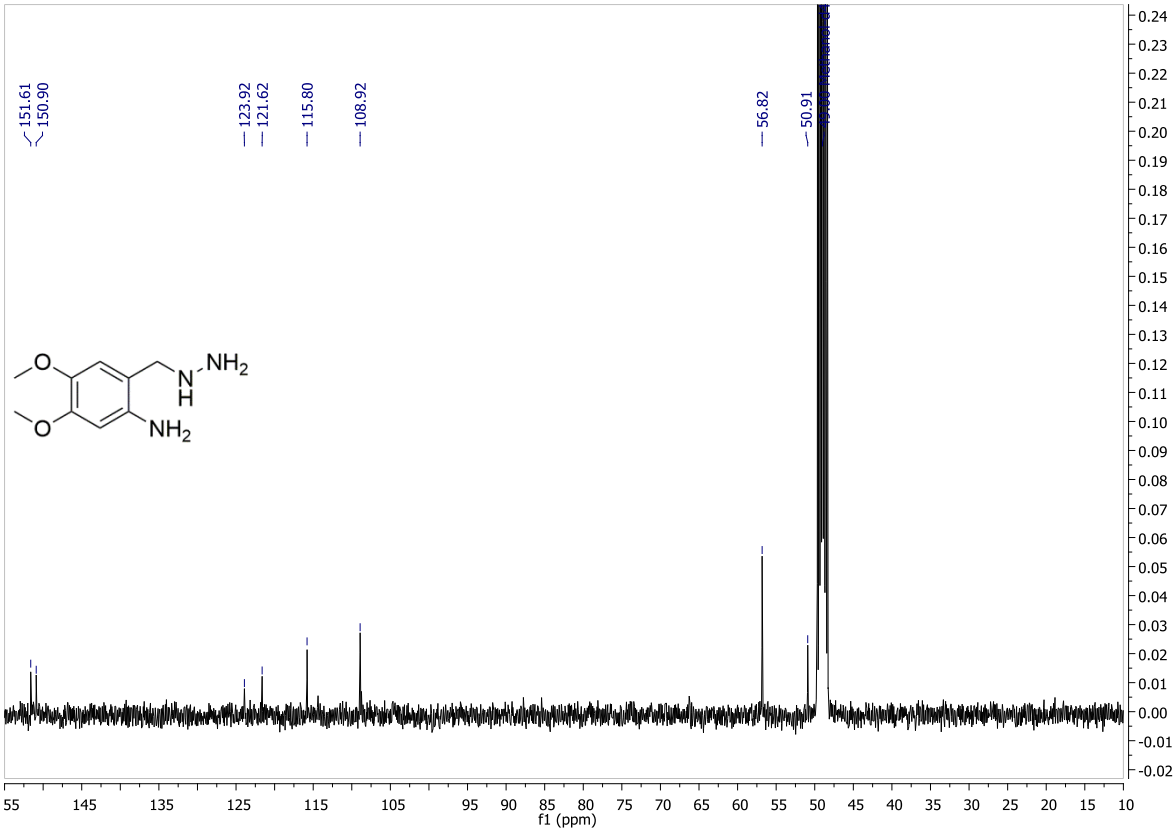

7.62
7.26 Chloroform-d
7.09
6.02
4.54
4.27
3.98
3.93
1.40
NHBoc
NO2
1.00
0.98
0.97
0.99
1.97
2.99
3.01
9.00
f1 (ppm)

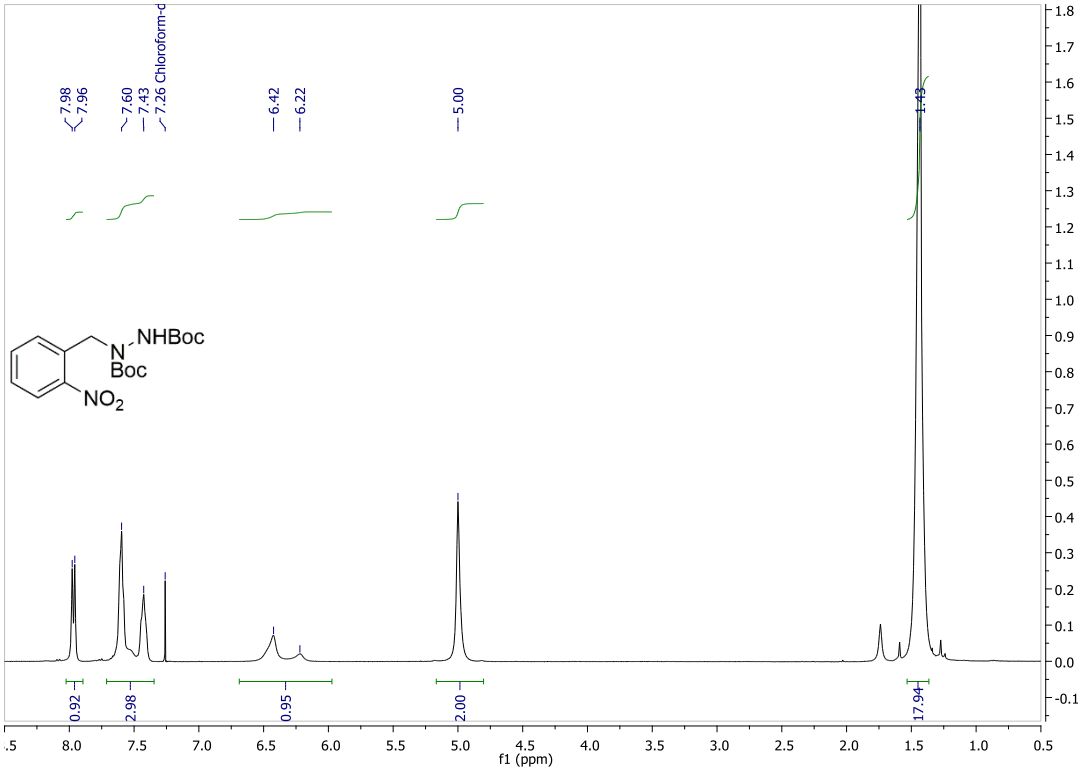

156.63
153.18
148.05
141.52
128.64
113.01
108.44
80.81
77.16 Chloroform-d
56.62
56.50
53.81
28.40
NHBoc
NO2
f1 (ppm)

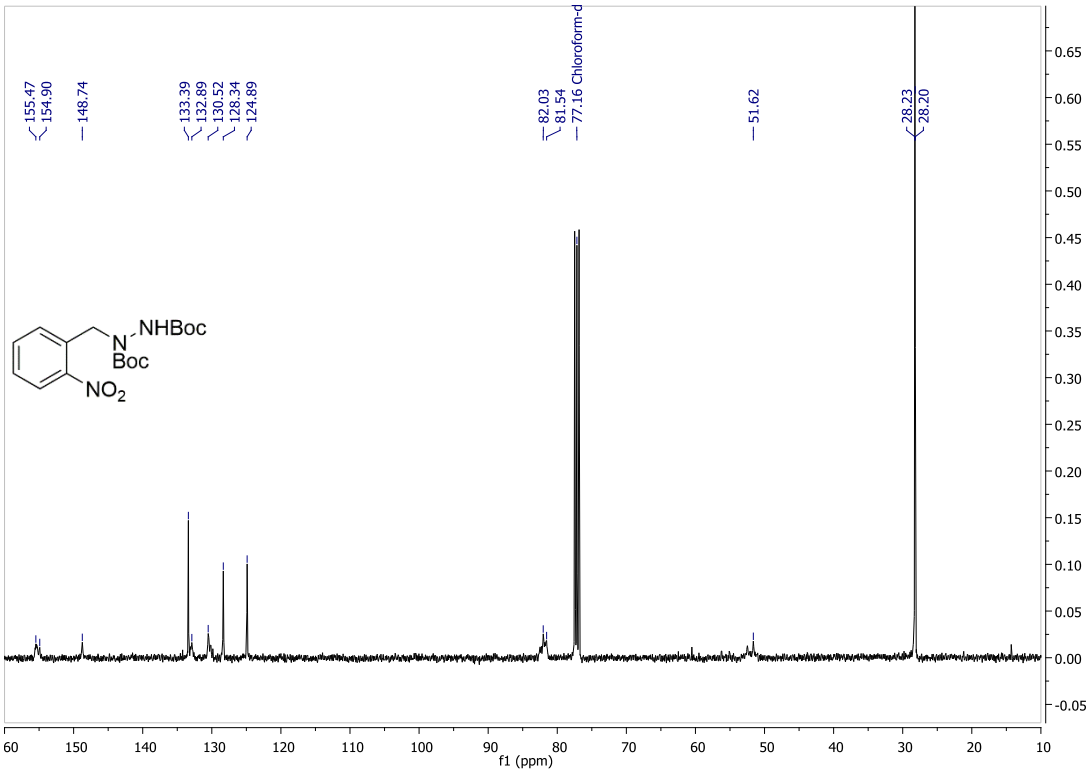

7.26 Chloroform-d
6.63
6.27
6.12
4.12
3.92
3.82
3.78
1.46
NHBoc
NH2
1.00
1.03
0.89
2.88
2.25
2.95
3.32
9.16
f1 (ppm)

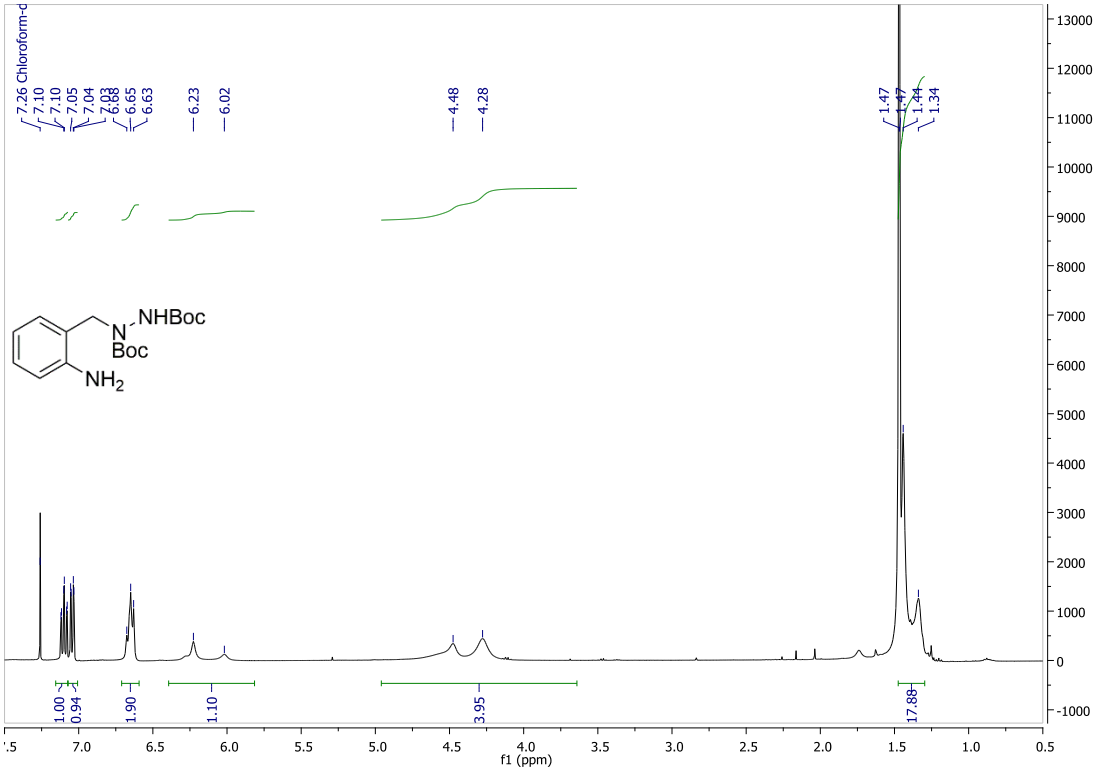

157.01
149.87
141.42
140.66
115.66
112.26
101.03
80.81
77.16 Chloroform
56.82
55.96
53.66
28.40
NHBoc
NH2
f1 (ppm)

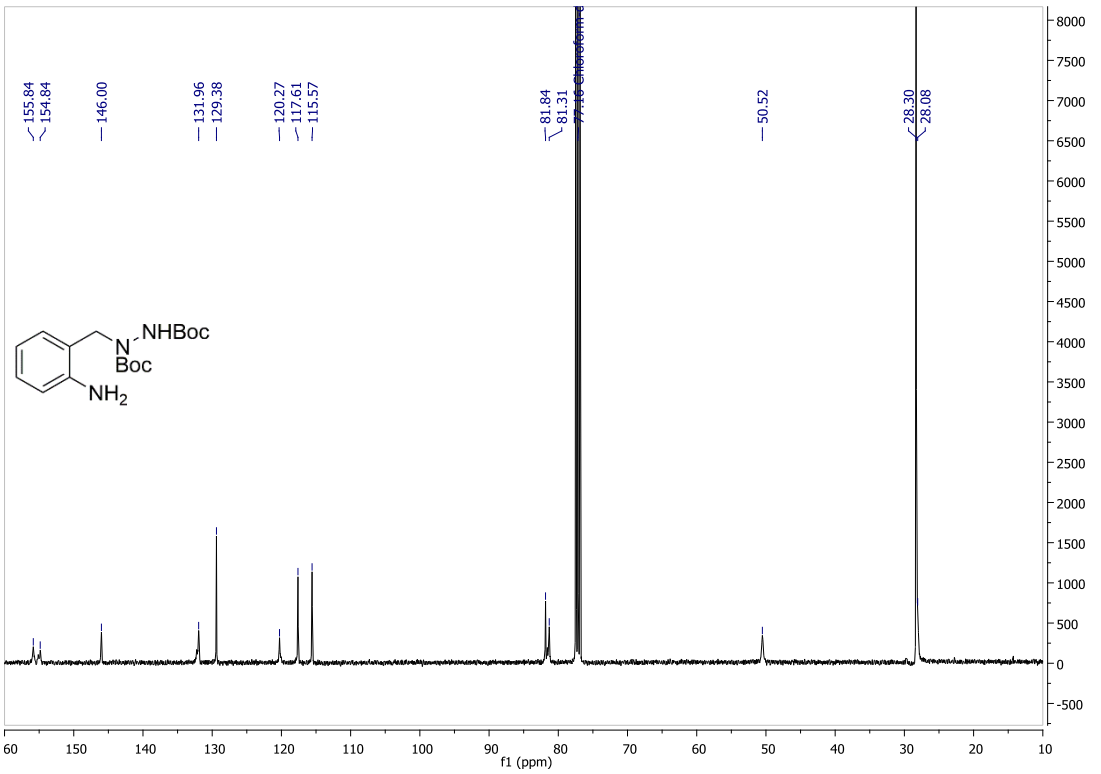

7.13
7.02
4.25
3.89
3.88
3.31 Methanol-d4
1.00
1.11
1.78
2.98
2.89
f1 (ppm)


151.61
150.90
123.92
121.62
115.80
108.92
56.82
50.91
f1 (ppm)


7.98
7.96
7.60
7.43
7.26 Chloroform-d
6.42
6.22
5.00
1.43
NHBoc
N
Boc
NO2
0.92
2.98
0.95
2.00
17.94
f1 (ppm)


155.47
154.90
148.74
133.39
132.89
130.52
128.34
124.89
82.03
81.54
77.16 Chloroform-d
51.62
28.23
28.20
NHBoc
N
Boc
NO2
f1 (ppm)


7.26 Chloroform-d
7.10
7.10
7.05
7.04
7.03
6.65
6.63
6.23
6.02
4.48
4.28
1.47
1.47
1.44
1.34
NHBoc
N
Boc
NH2
1.00
0.94
1.90
1.10
3.95
17.88
f1 (ppm)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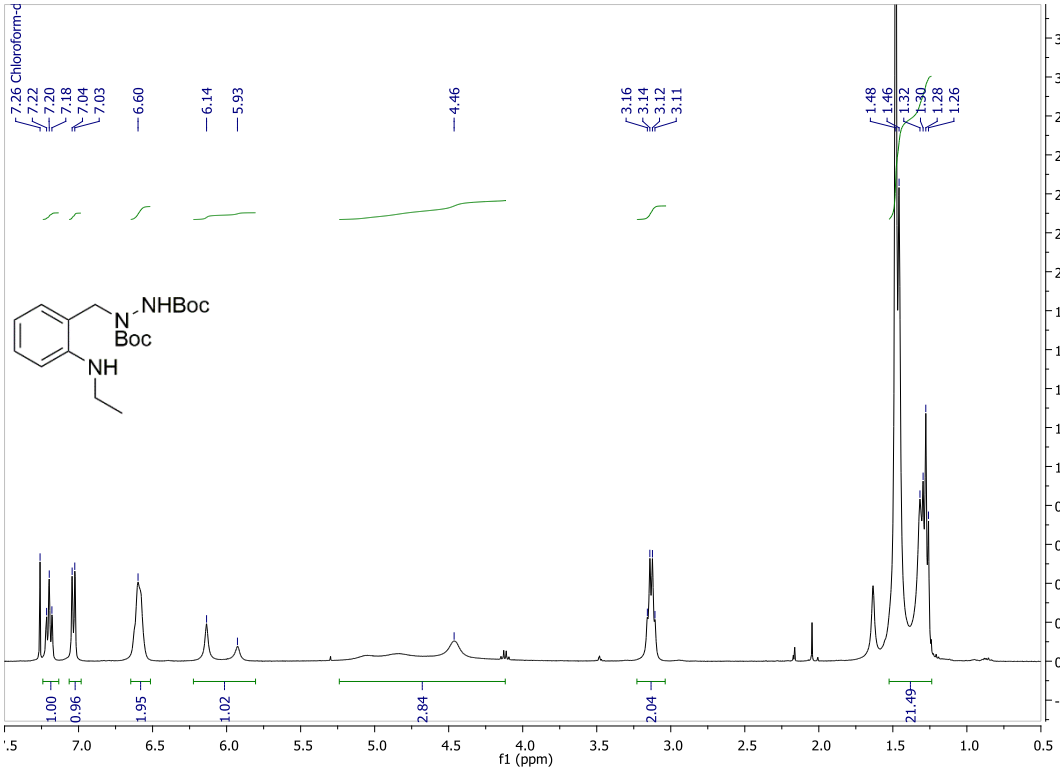
131.96
129.38
120.27
117.61
115.57
81.84
81.31
77.16 Chloroform-d
50.52
28.30
28.08
NHBoc
N
Boc
NH2
f1 (ppm)


7.26 Chloroform-d
7.22
7.20
7.18
7.04
7.03
6.60
6.14
5.93
4.46
3.16
3.14
3.12
3.11
1.48
1.46
1.32
1.30
1.28
1.26
NHBoc
N
Boc
NH
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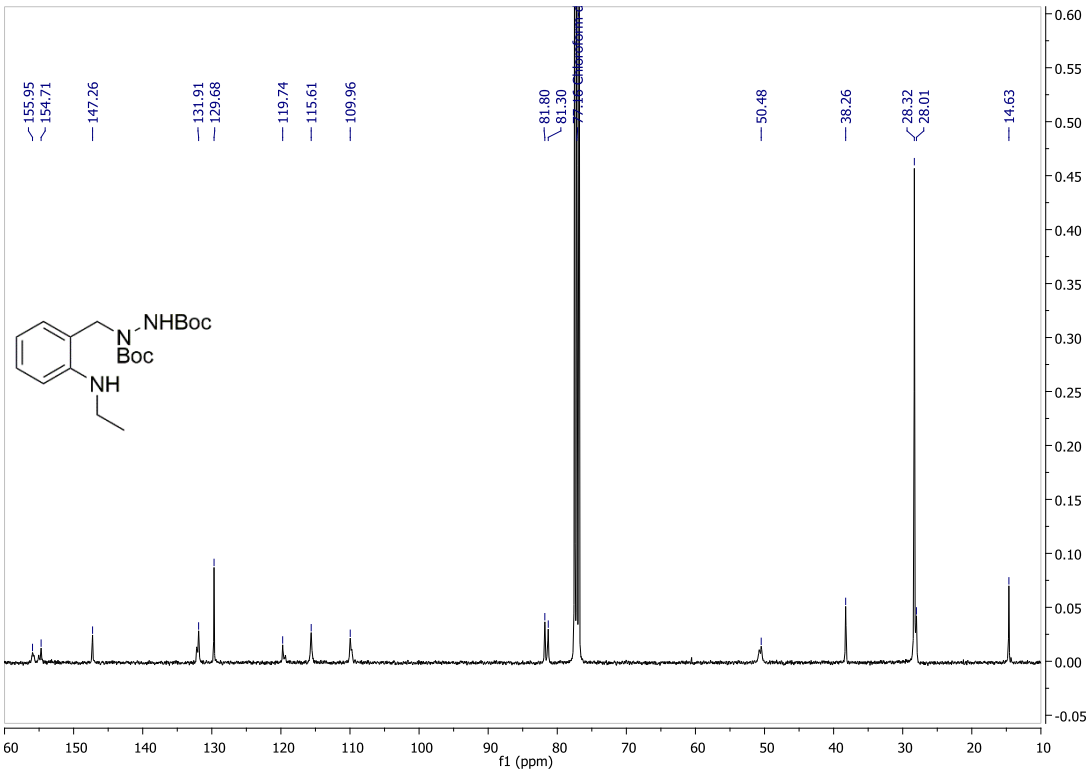
1.95
1.02
2.84
2.04
21.49
f1 (ppm)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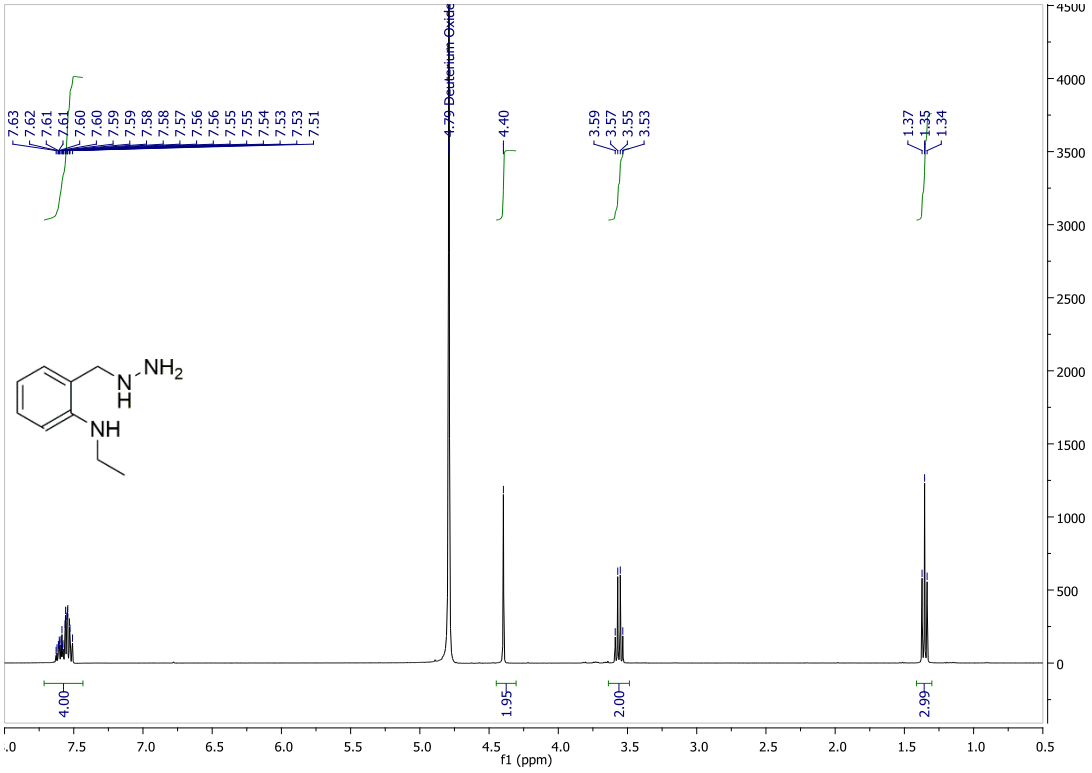
131.91
129.68
119.74
115.61
109.96
81.80
81.30
77.16 Chloroform-d
50.48
38.26
28.32
28.01
14.63
NHBoc
N
Boc
NH
f1 (ppm)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7.59
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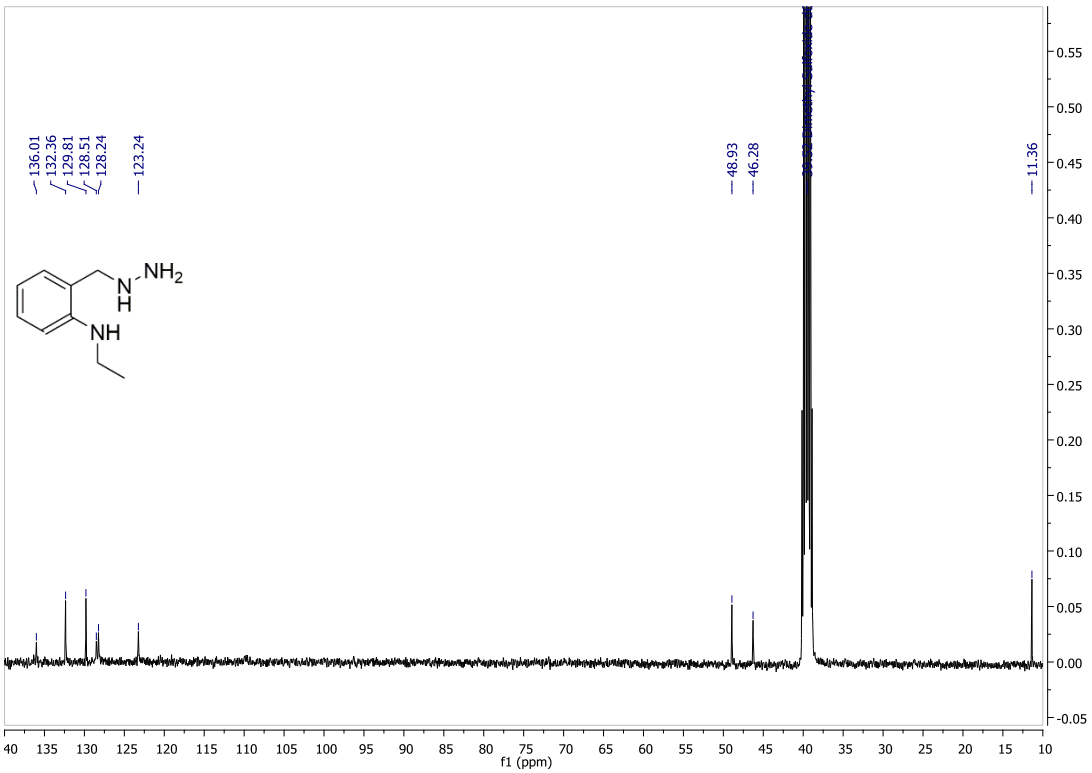
7.56
7.56
7.55
7.55
7.54
7.53
7.53
7.51
4.79 Deuterium Oxide
4.40
3.59
3.57
3.55
3.53
1.37
1.35
1.34
NH2
N
H
NH
4.00
1.95
2.00
2.99
f1 (ppm)


136.01
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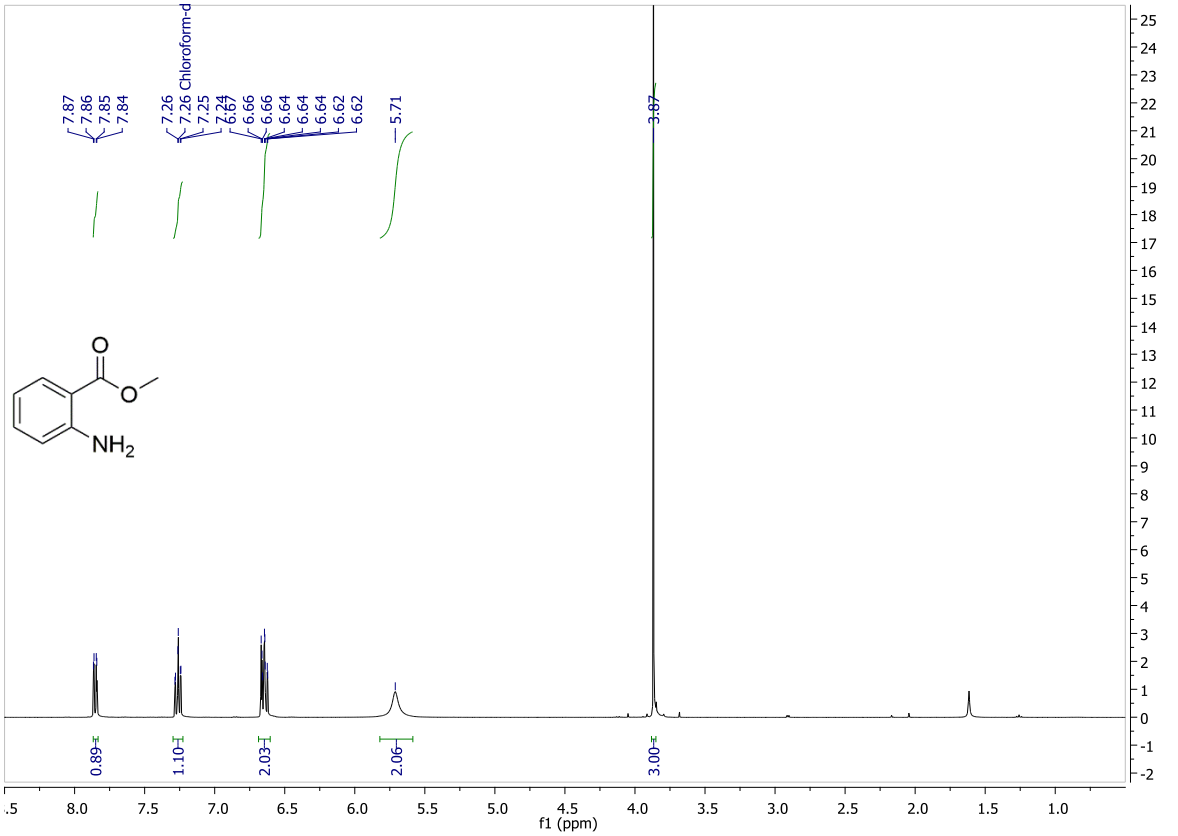
128.51
128.24
123.24
48.93
46.28
11.36
NH2
N
H
NH
f1 (ppm)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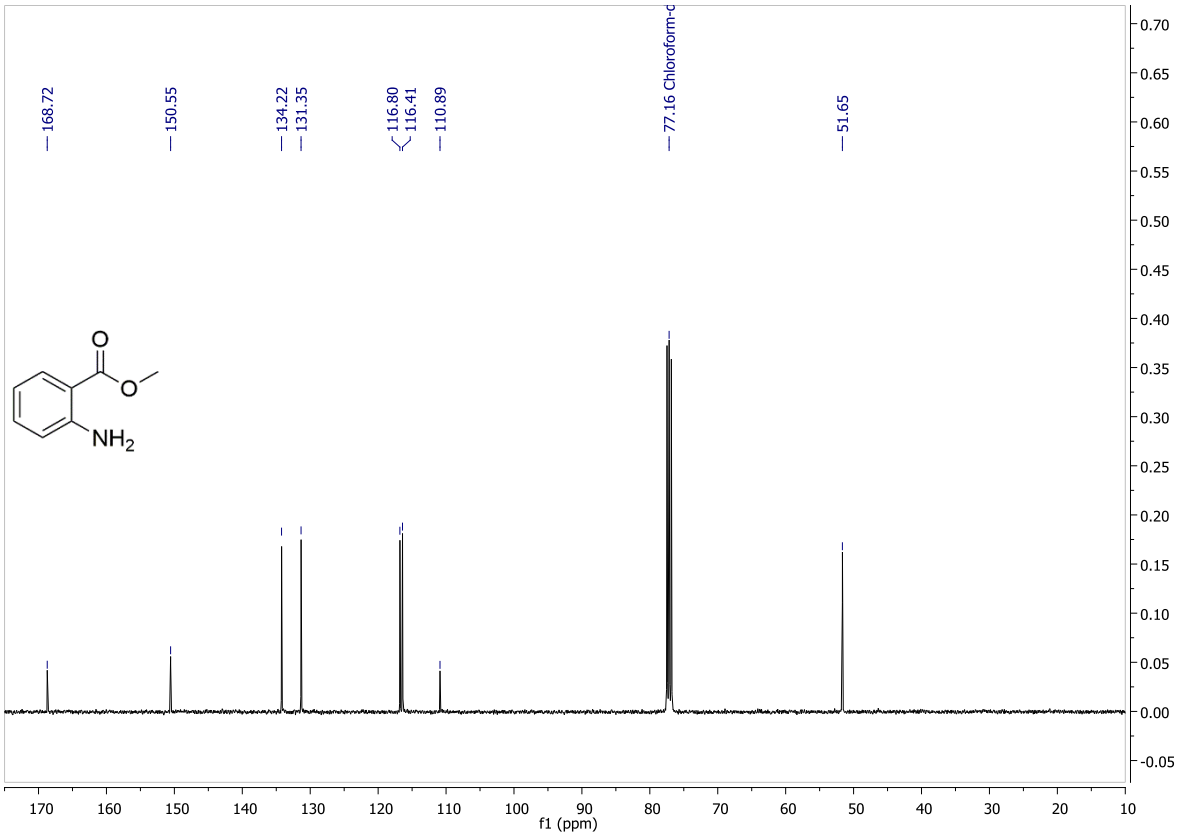
7.26 Chloroform-d
7.25
6.67
6.66
6.66
6.64
6.64
6.64
6.62
6.62
5.71
3.87
O
O
NH2
0.89
1.10
2.03
2.06
3.00
f1 (ppm)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116.80
116.41
110.89
77.16 Chloroform-d
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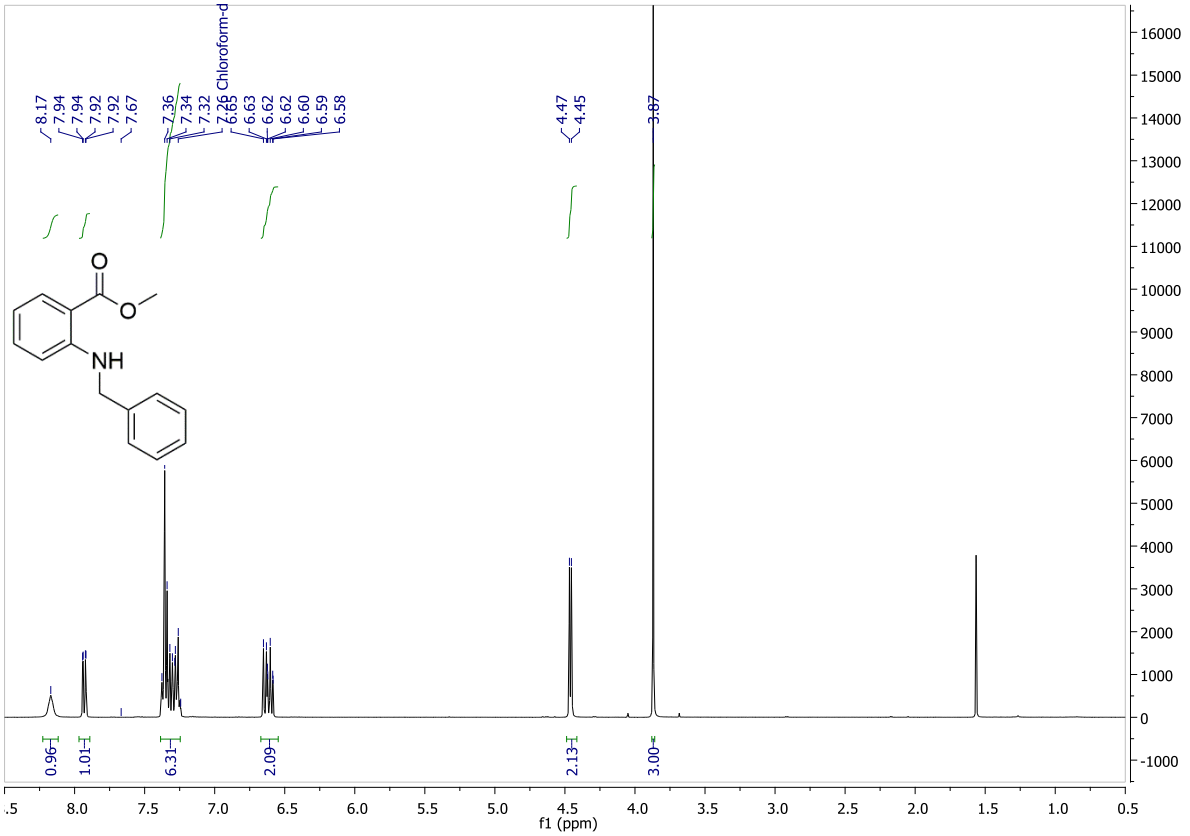
O
O
NH2
f1 (ppm)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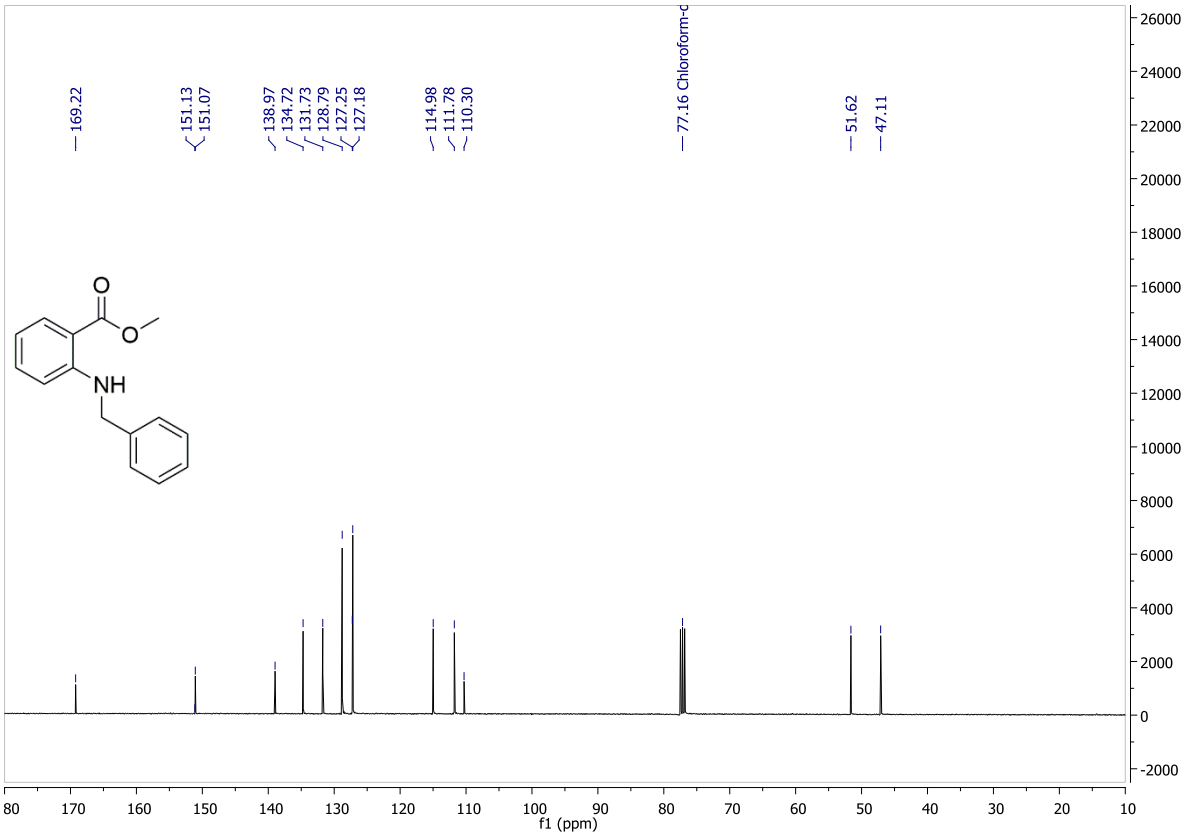
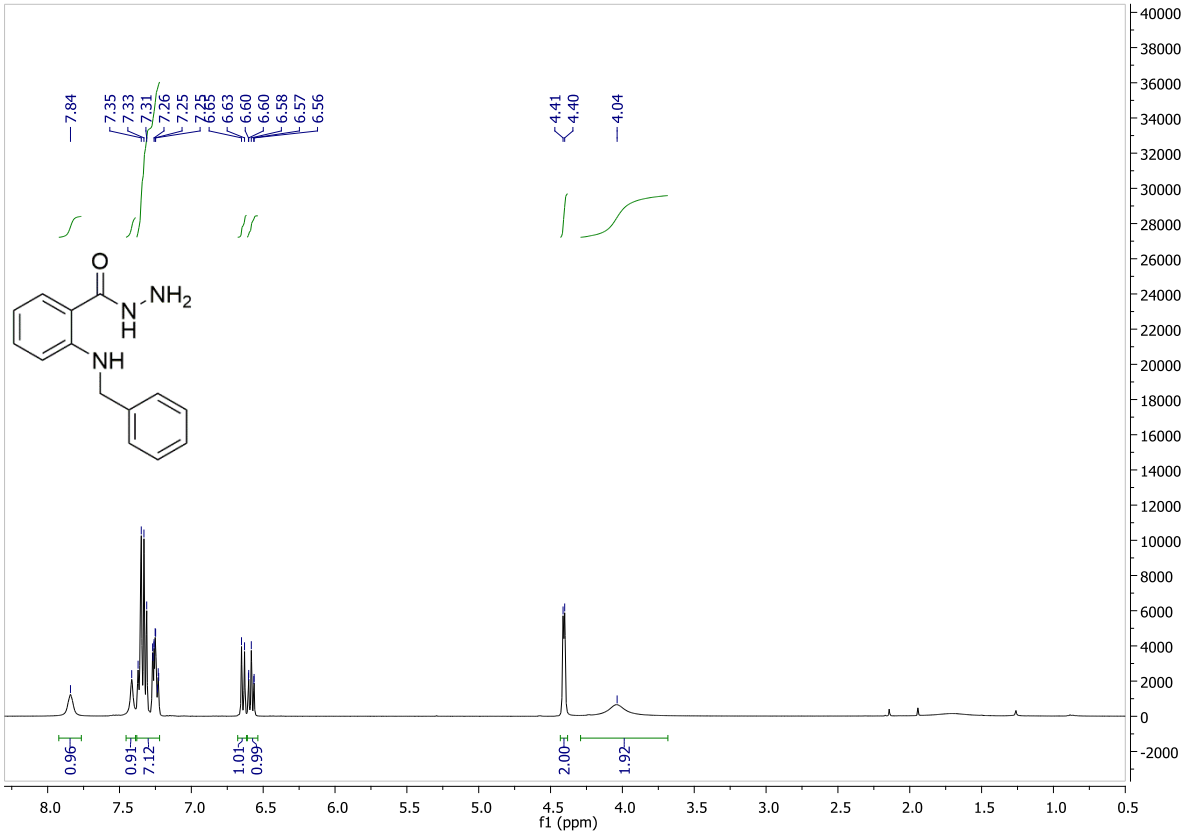
7.94
7.94
7.92
7.92
7.67
7.36
7.34
7.32
7.26 Chloroform-d
6.63
6.62
6.62
6.60
6.59
6.58
4.47
4.45
3.87
O
O
NH
0.96
1.01
6.31
2.09
2.13
3.00
f1 (ppm)


169.22
151.13
151.07
138.97
134.72
131.73
128.79
127.25
127.18
114.98
111.78
110.30
77.16 Chloroform-d
51.62
47.11
O
O
NH
f1 (ppm)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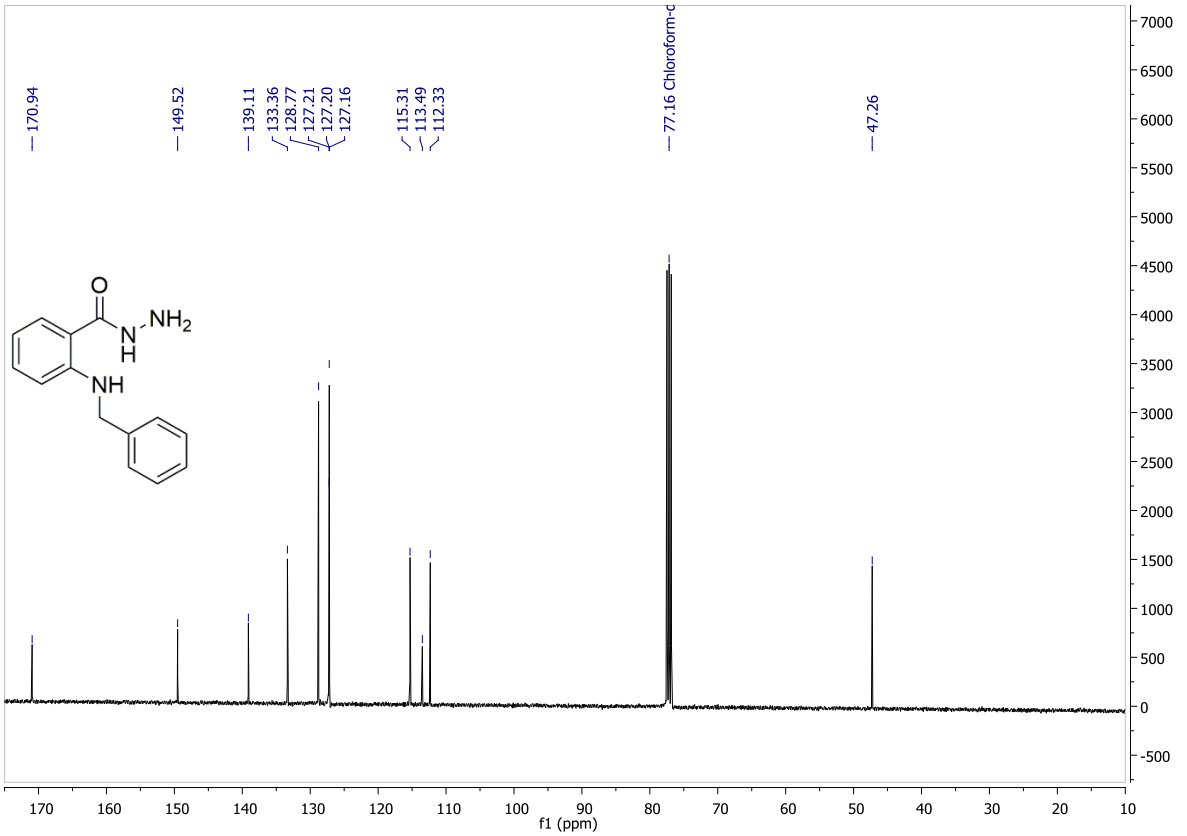
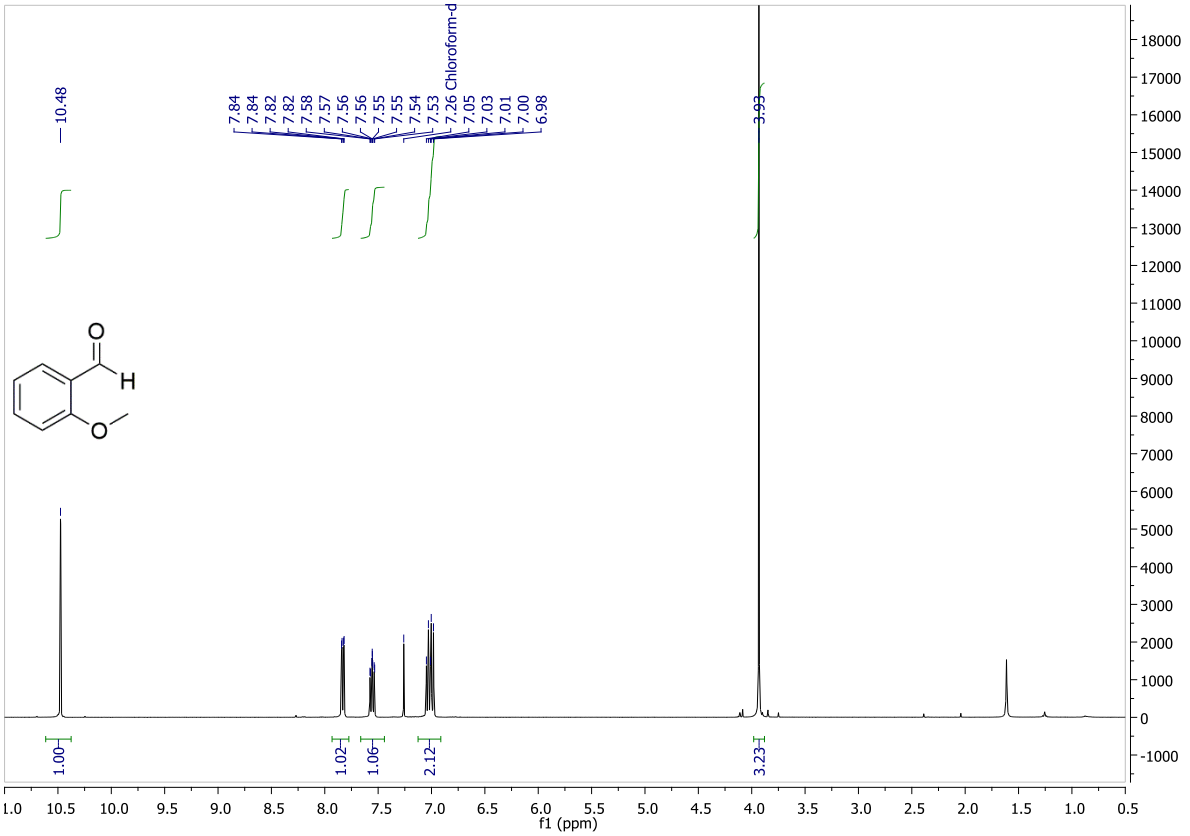
7.25
6.65
6.63
6.60
6.60
6.58
6.57
6.56
4.41
4.40
4.04
O
N
NH2
H
NH
0.96
0.91
7.12
1.01
0.99
2.00
1.92
f1 (ppm)


170.94
149.52
139.11
133.36
128.77
127.21
127.20
127.16
115.31
113.49
112.33
77.16 Chloroform-d
47.26
O
N
NH2
H
NH
f1 (ppm)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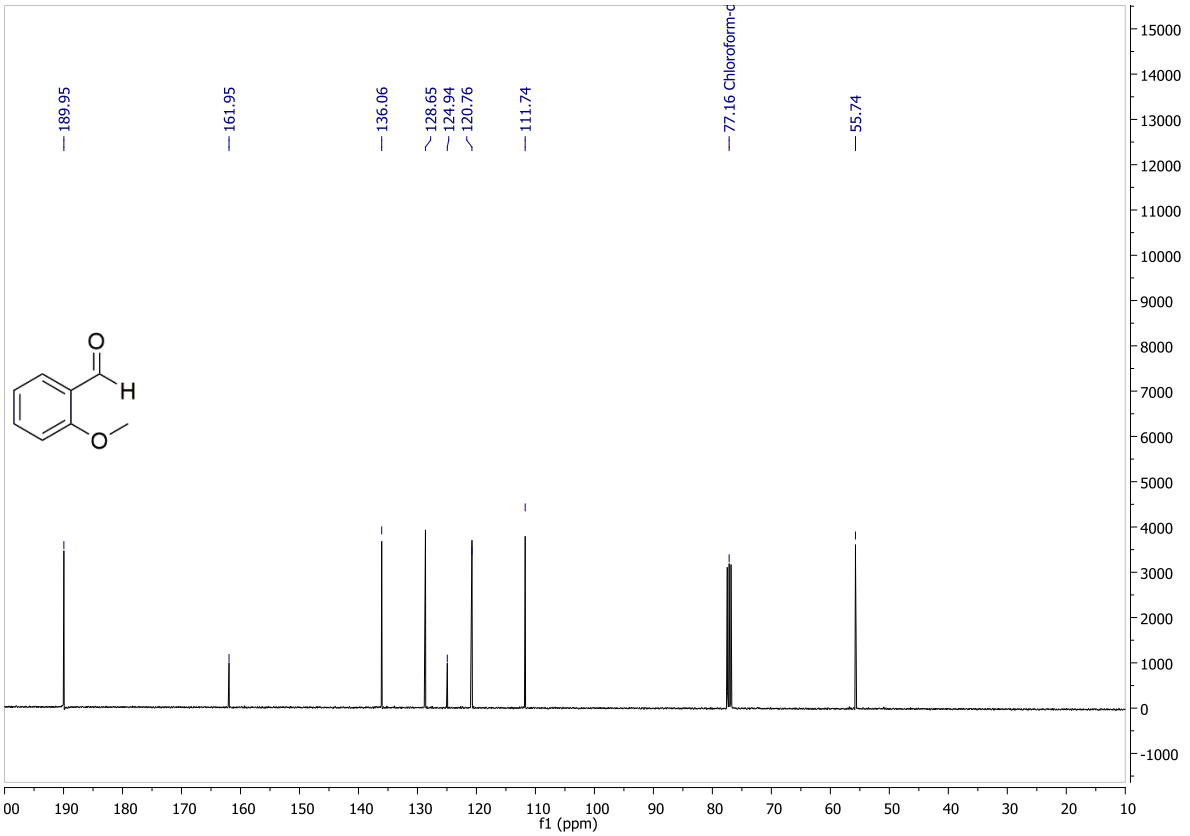
7.84
7.84
7.82
7.82
7.58
7.57
7.56
7.56
7.55
7.55
7.54
7.53
7.26 Chloroform-d
7.05
7.03
7.01
7.00
6.98
3.93
1.00
1.02
1.06
2.12
3.23
f1 (ppm)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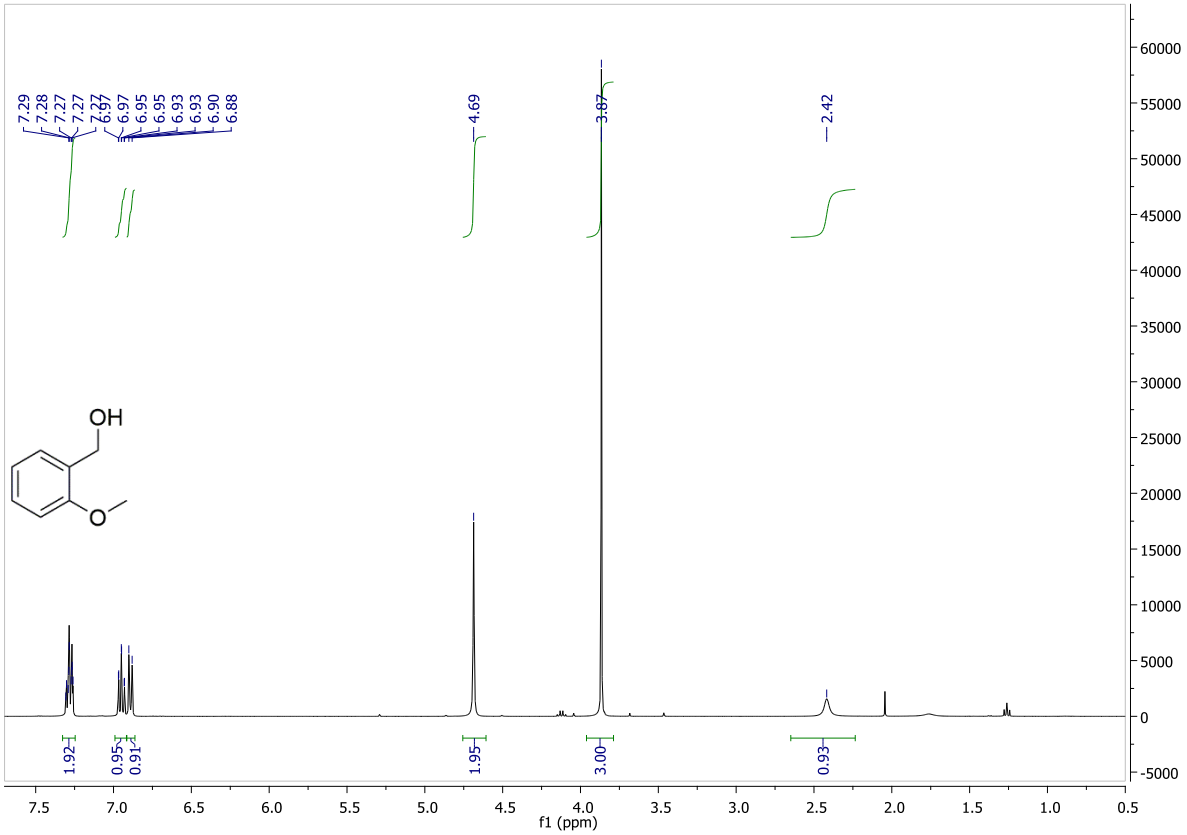
120.76
111.74
77.16 Chloroform-d
55.74
f1 (ppm)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7.28
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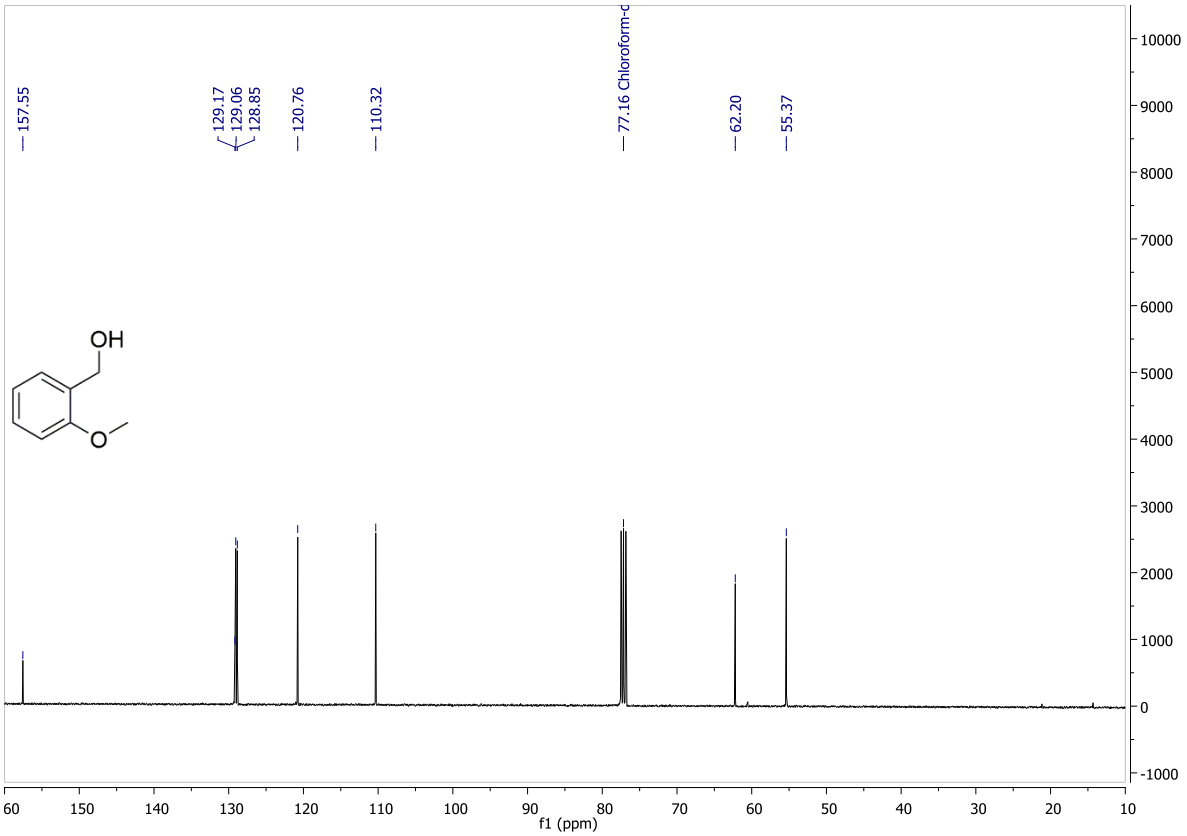
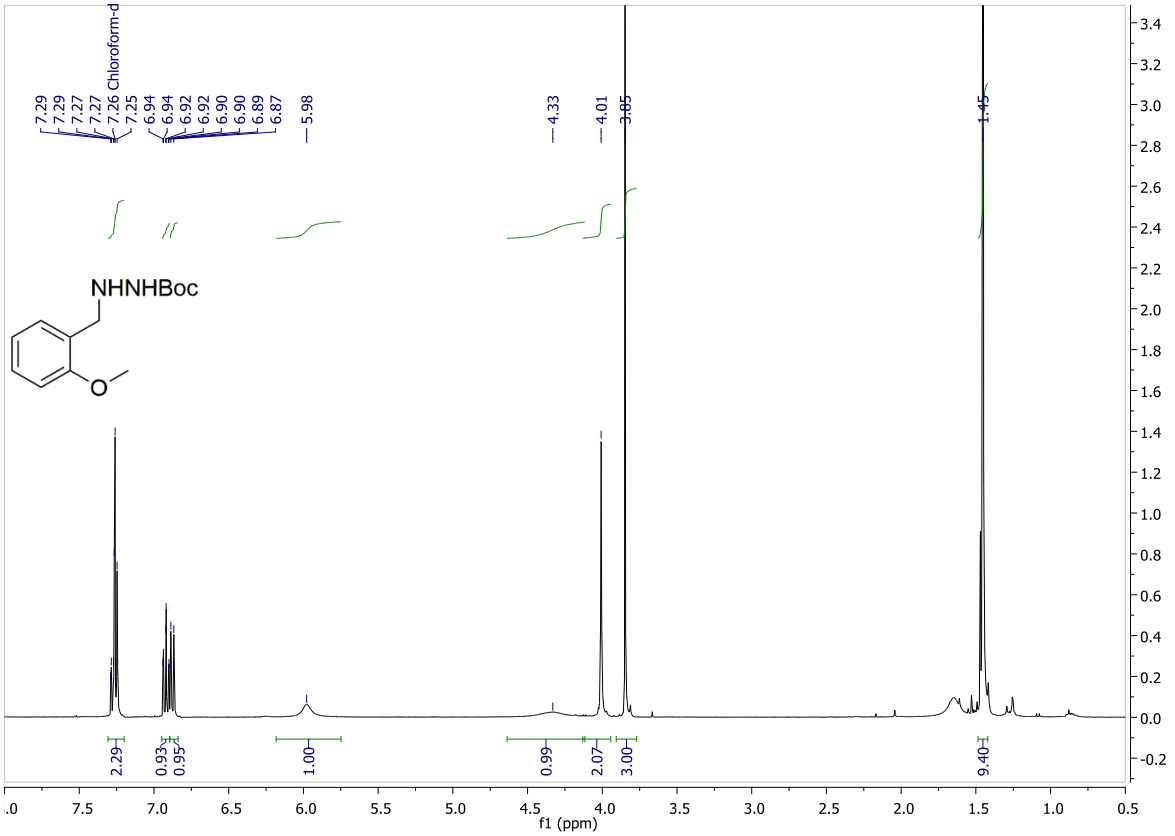
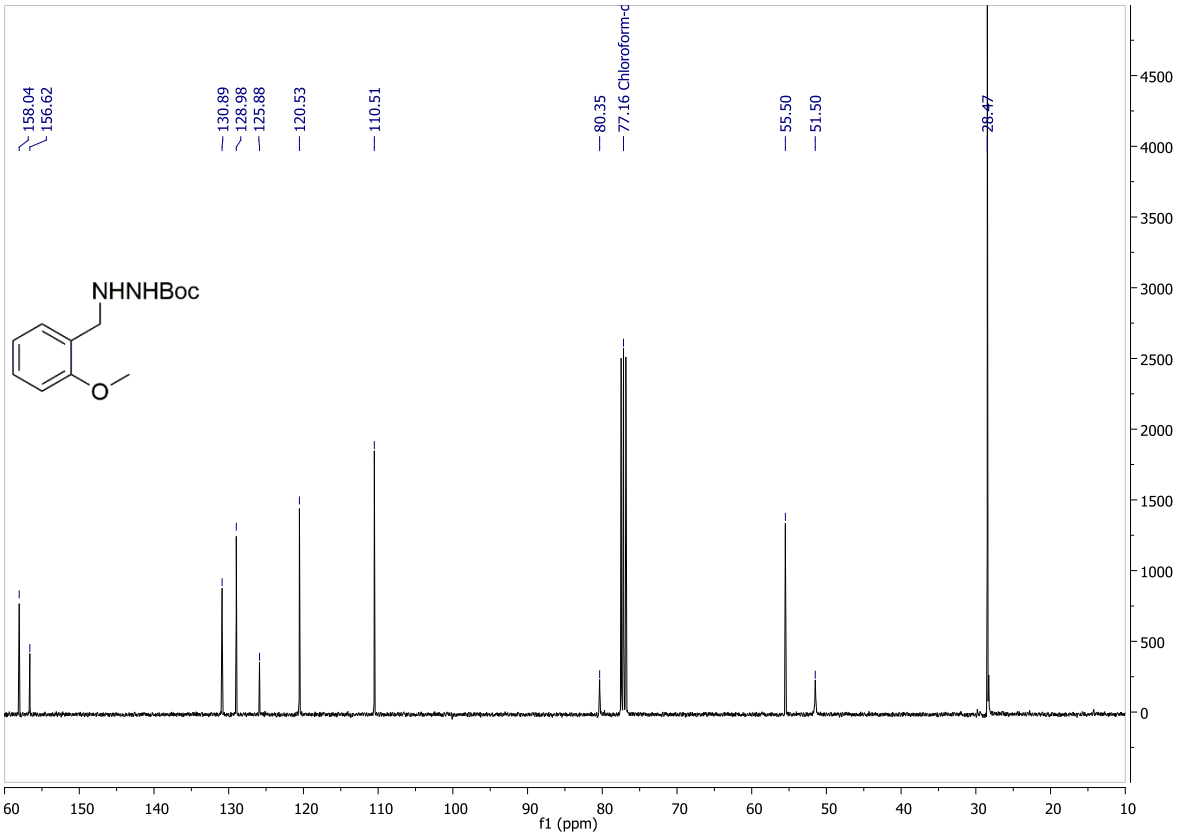
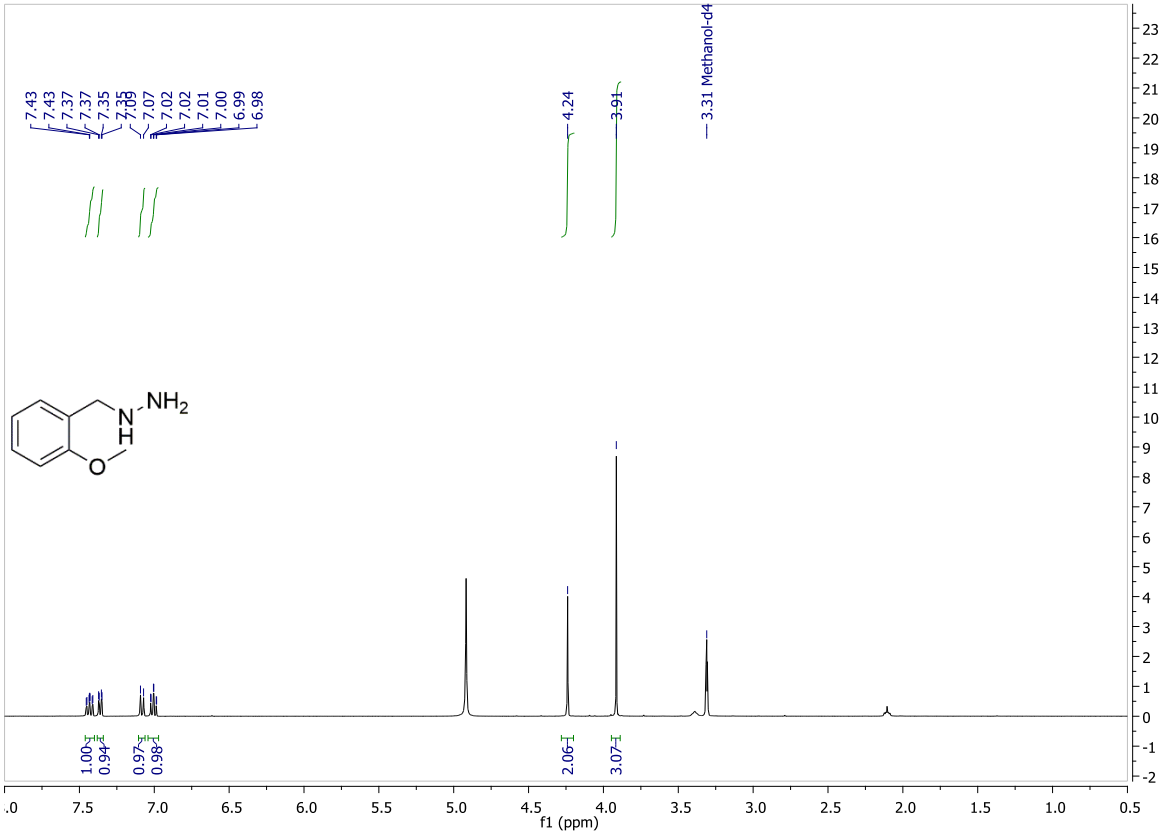
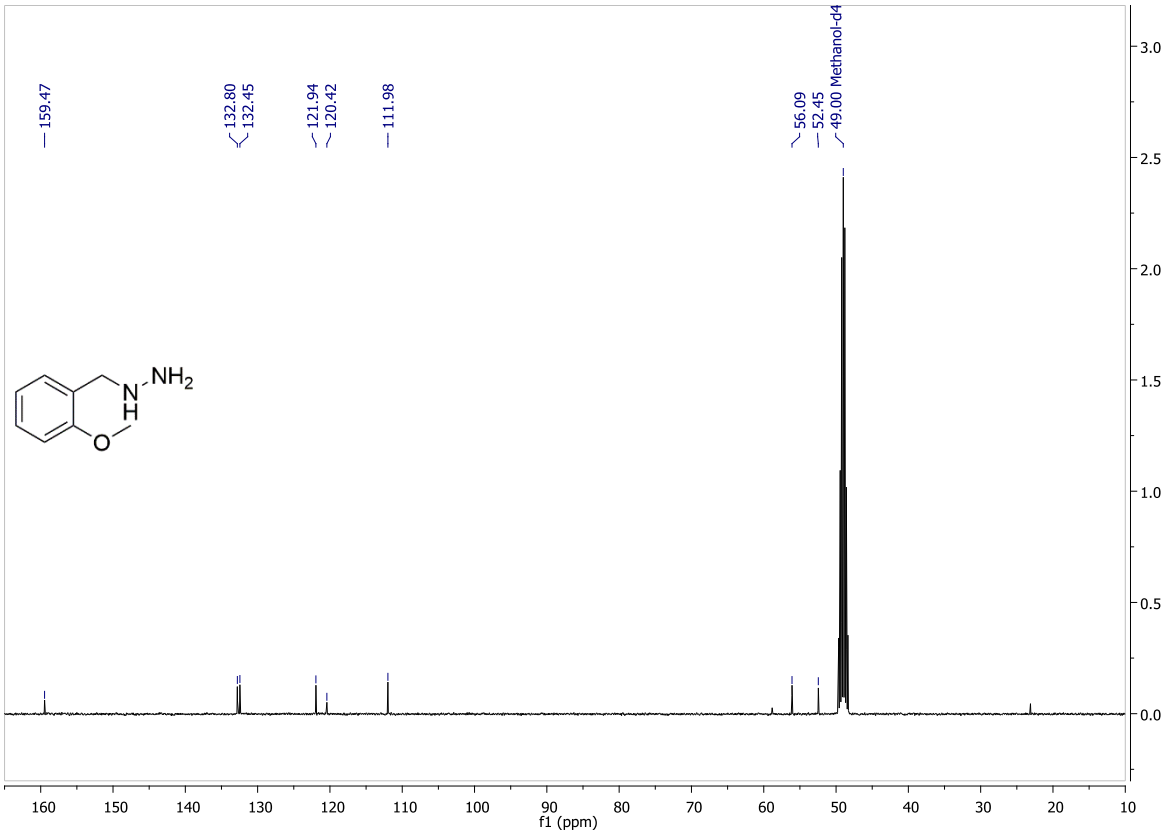
7.27
6.97
6.97
6.95
6.95
6.93
6.93
6.90
6.88
4.69
3.87
2.42
OH
O
1.92
0.95
0.91
1.95
3.00
0.93
f1 (ppm)

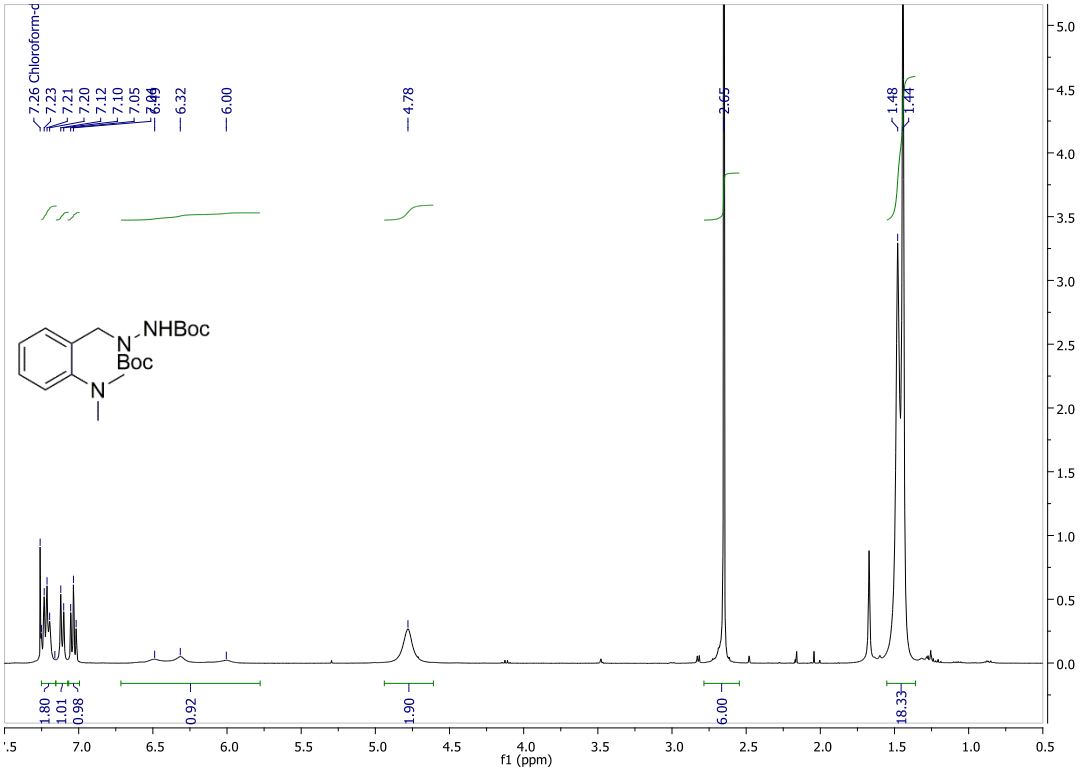

157.55
129.17
129.06
128.85
120.76
110.32
77.16 Chloroform-d
62.20
55.37
OH
O
f1 (ppm)

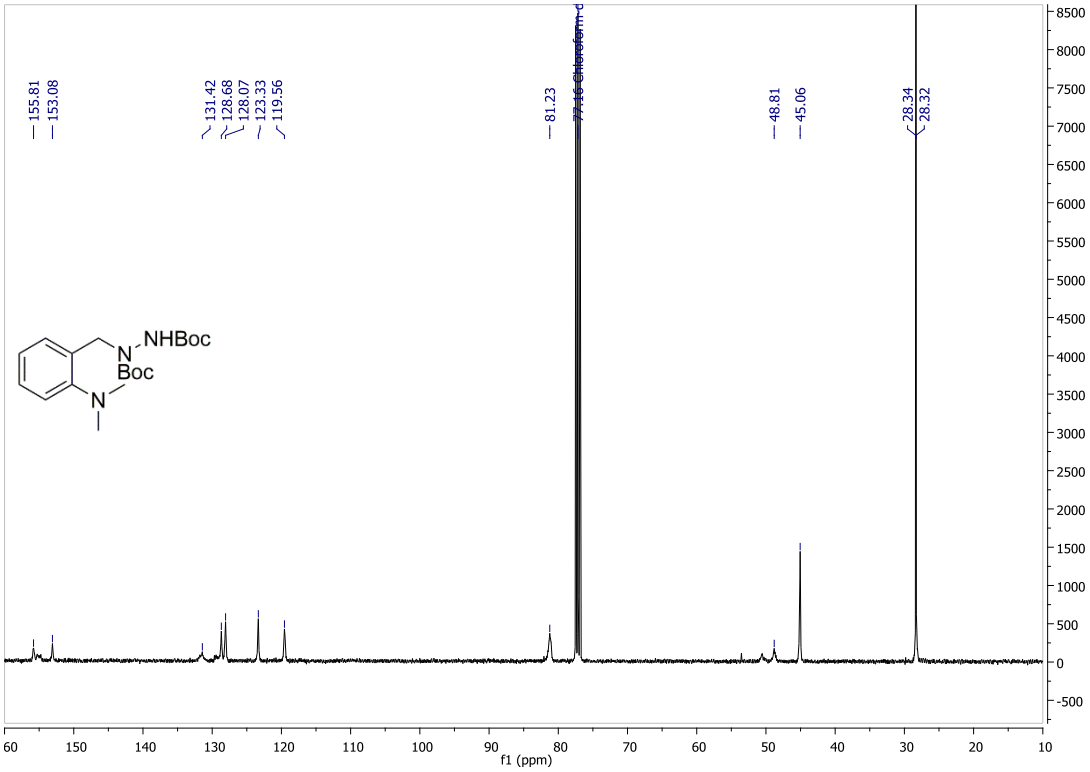

7.29
7.29
7.27
7.27
7.26 Chloroform-d
7.25
6.94
6.94
6.92
6.92
6.90
6.90
6.89
6.87
5.98
4.33
4.01
3.85
1.45
NHNHBoc
O
2.29
0.93
0.95
1.00
0.99
2.07
3.00
9.40
f1 (ppm)

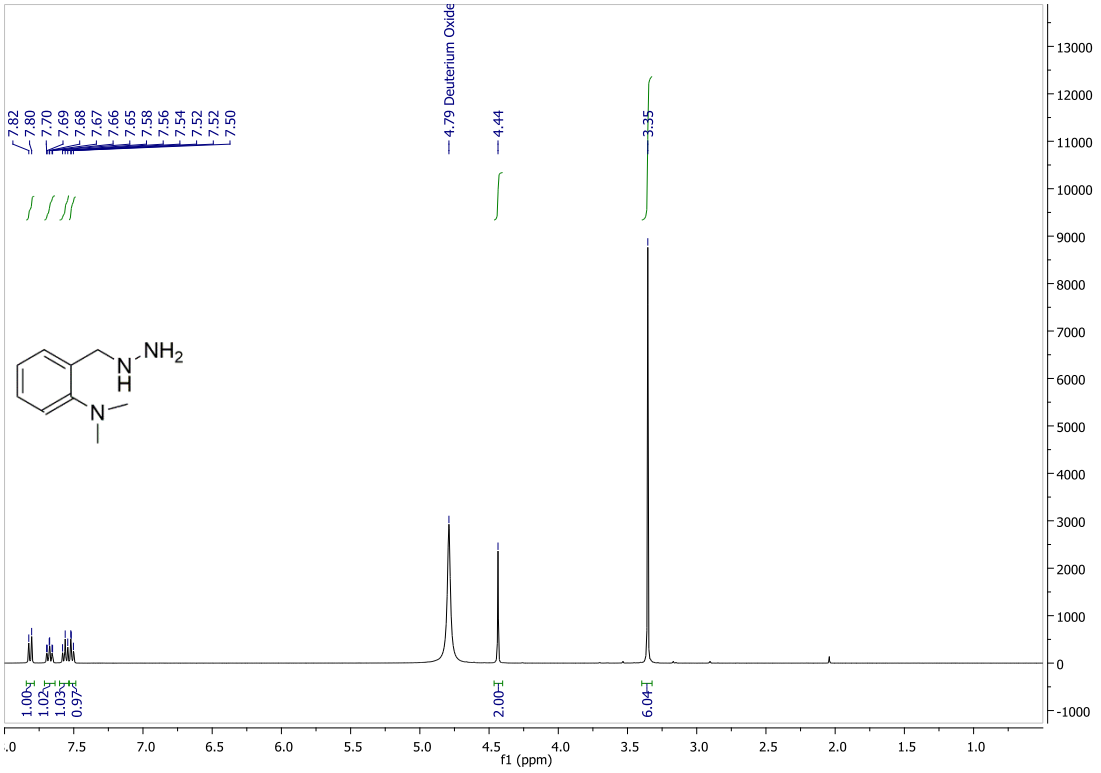

158.04
156.62
130.89
128.98
125.88
120.53
110.51
80.35
77.16 Chloroform-d
55.50
51.50
28.47
NHNHBoc
O
f1 (ppm)

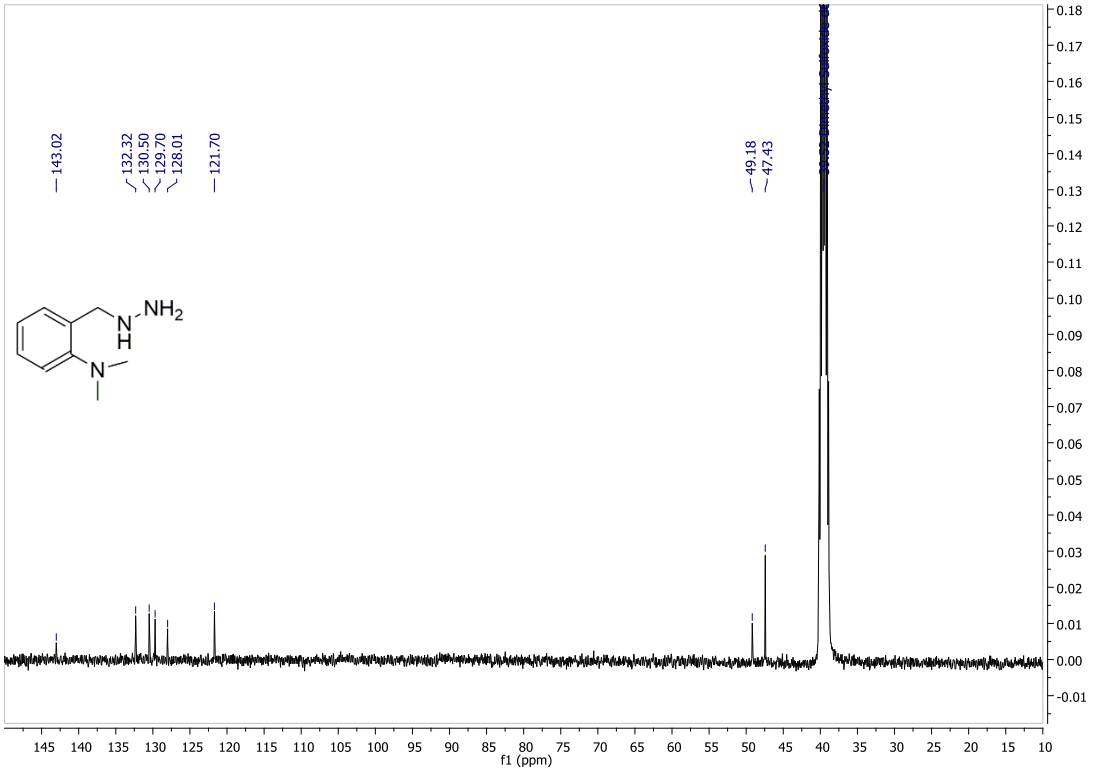

7.43
7.43
7.37
7.37
7.35
7.35
7.08
7.07
7.02
7.02
7.01
7.00
6.99
6.98
4.24
3.91
3.31 Methanol-d4
NH2
N
H
O
1.00
0.94
0.97
0.98
2.06
3.07
f1 (ppm)

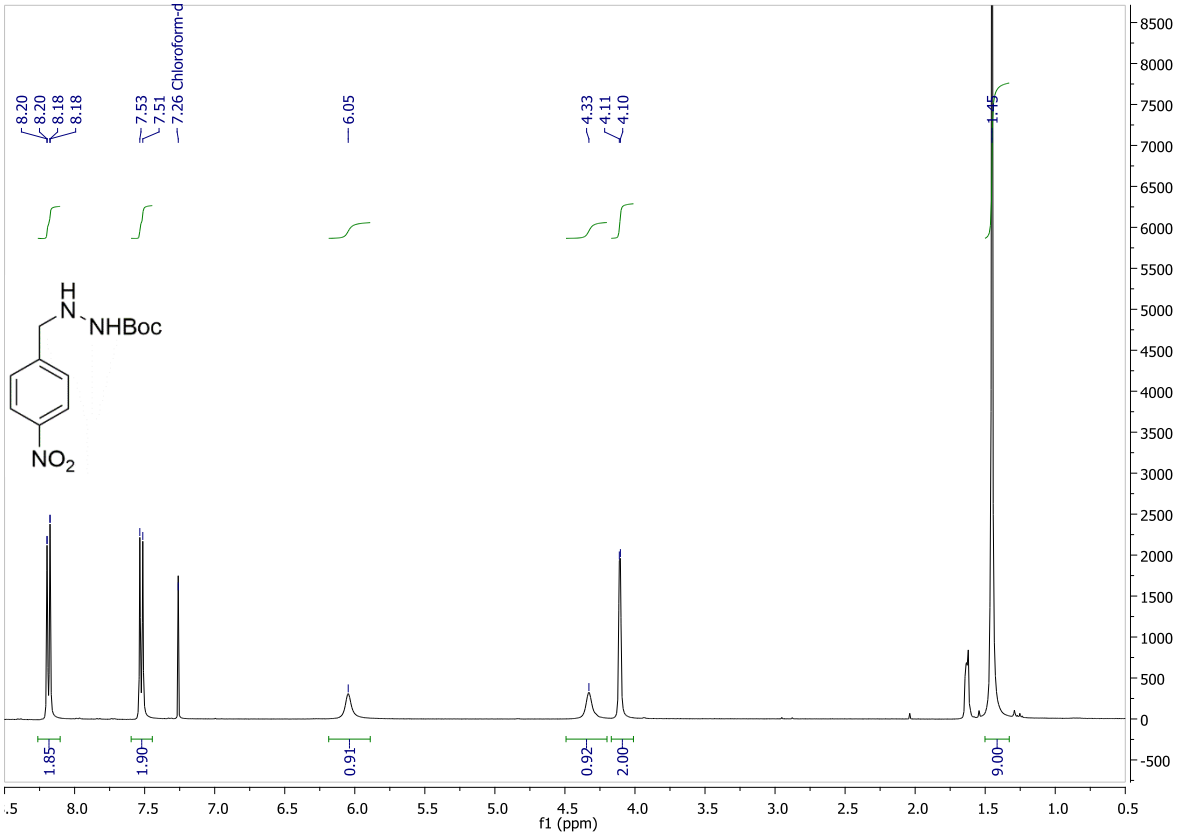

159.47
132.80
132.45
121.94
120.42
111.98
56.09
52.45
49.00 Methanol-d4
NH2
N
H
O
f1 (ppm)

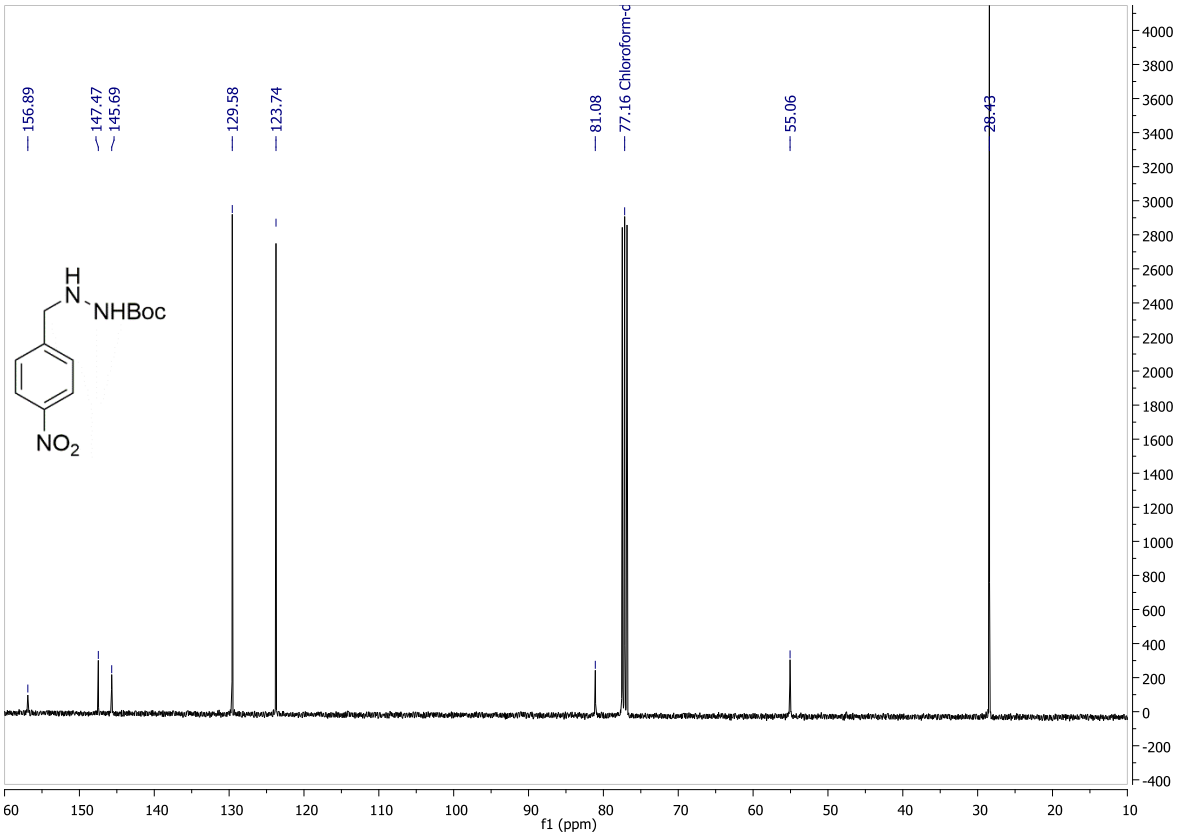

7.26 Chloroform-d
7.23
7.21
7.20
7.12
7.10
7.05
6.49
6.32
6.00
4.78
2.65
1.48
1.44
NHBoc
Boc
1.80
1.01
0.98
0.92
1.90
6.00
18.33
f1 (ppm)

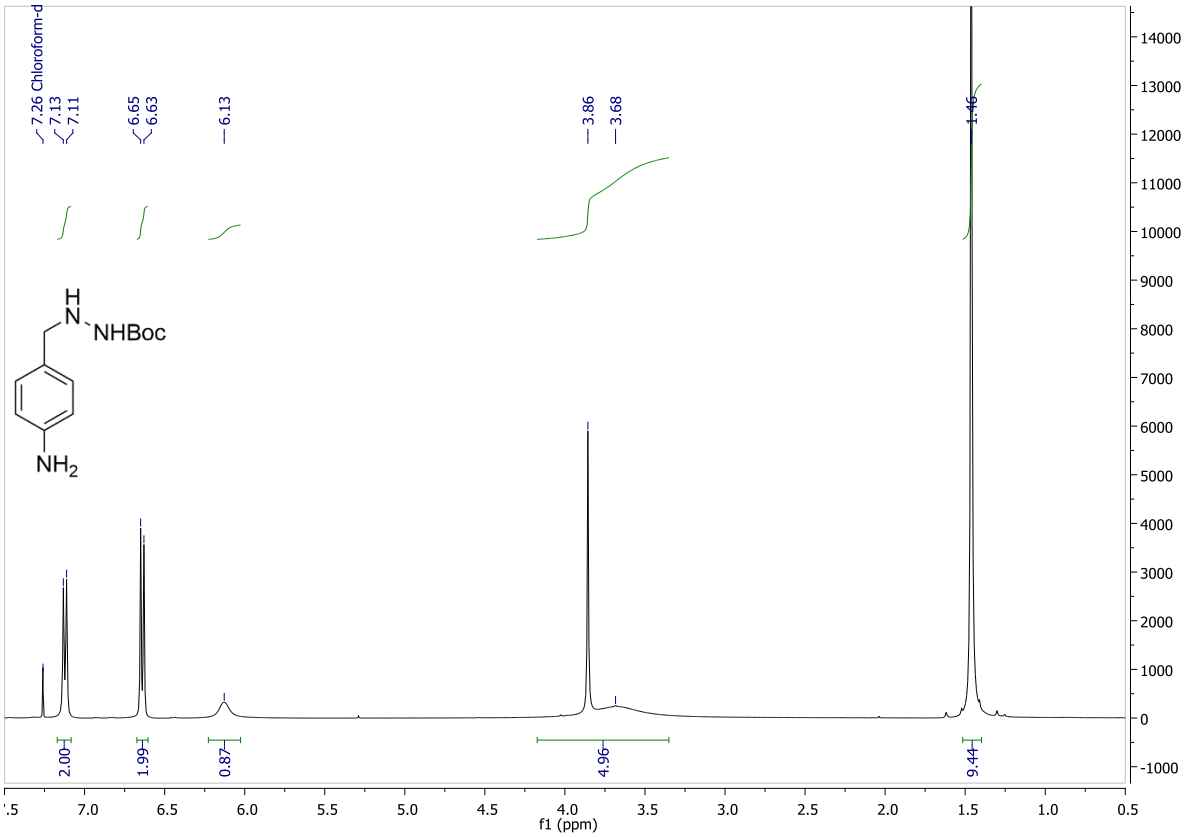

155.81
153.08
131.42
128.68
128.07
123.33
119.56
81.23
77.16 Chloroform-d
48.81
45.06
28.34
28.32
NHBoc
Boc
f1 (ppm)

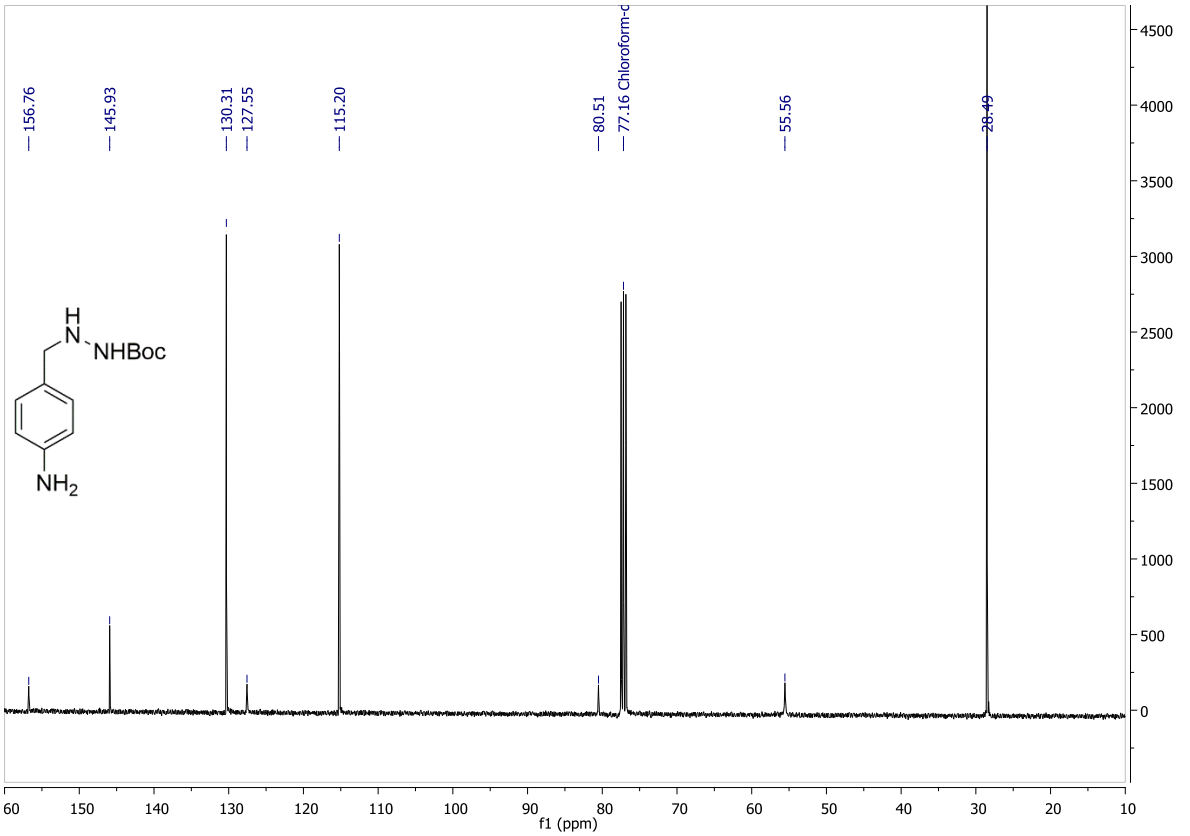

7.82
7.80
7.70
7.69
7.68
7.67
7.66
7.65
7.58
7.56
7.54
7.52
7.52
7.50
4.79 Deuterium Oxide
4.44
3.35
NH2
N
H
N
1.00
1.02
1.03
0.97
2.00
6.04
f1 (ppm)

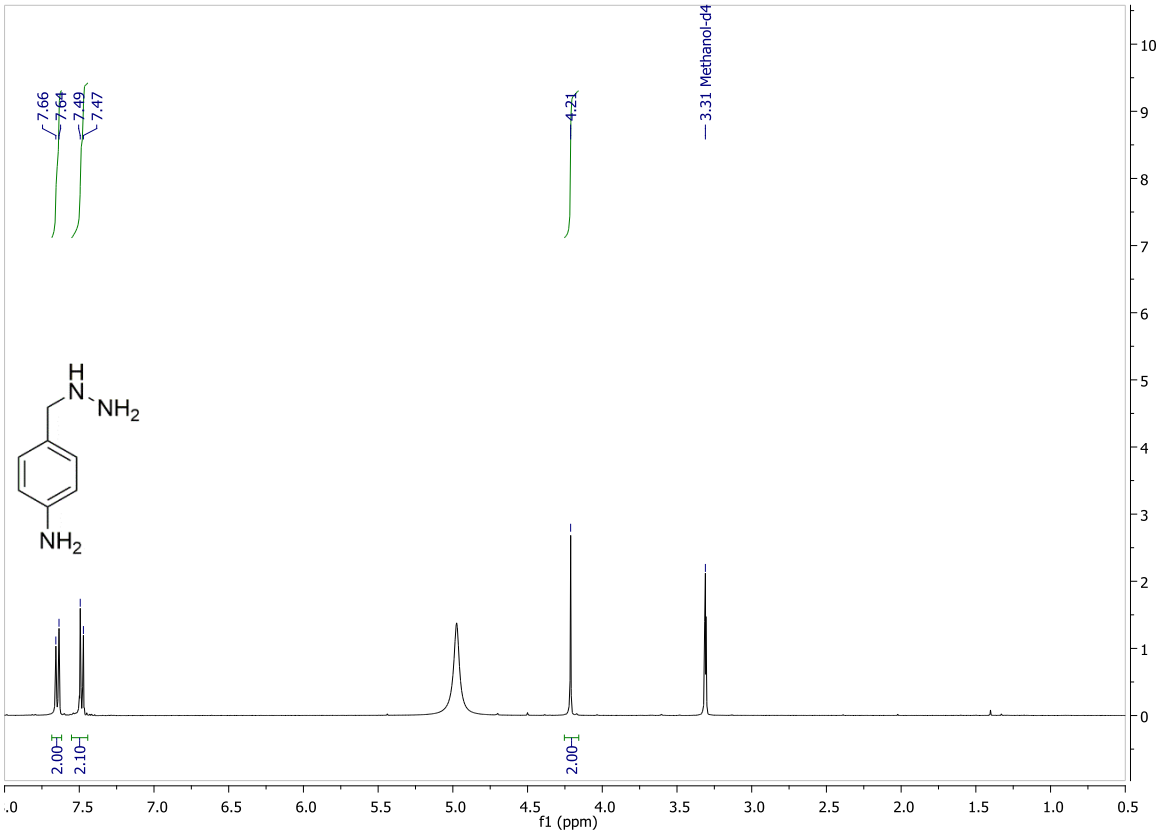

143.02
132.32
130.50
129.70
128.01
121.70
49.18
47.43
NH2
N
H
N
f1 (ppm)

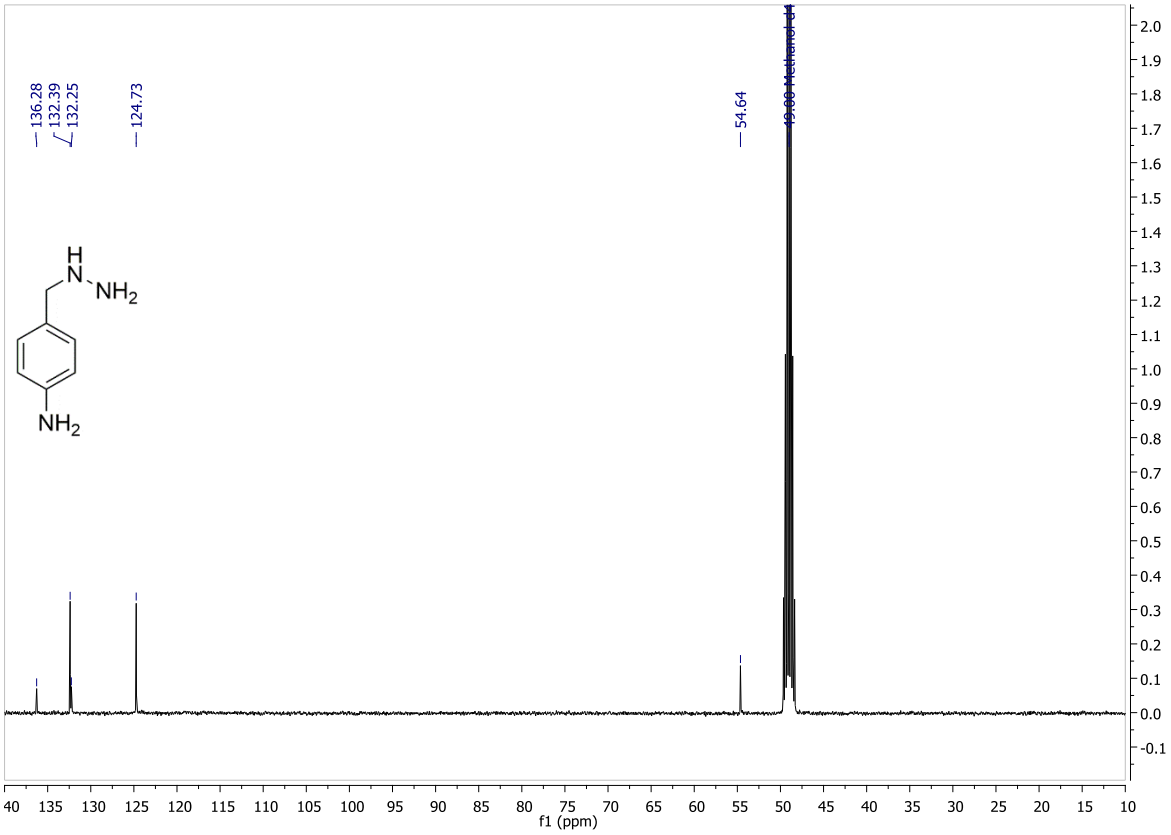

8.20
8.20
8.18
8.18
7.53
7.51
7.26 Chloroform-d
6.05
4.33
4.11
4.10
1.45
NHBoc
NO2
1.85
1.90
0.91
0.92
2.00
9.00
f1 (ppm)

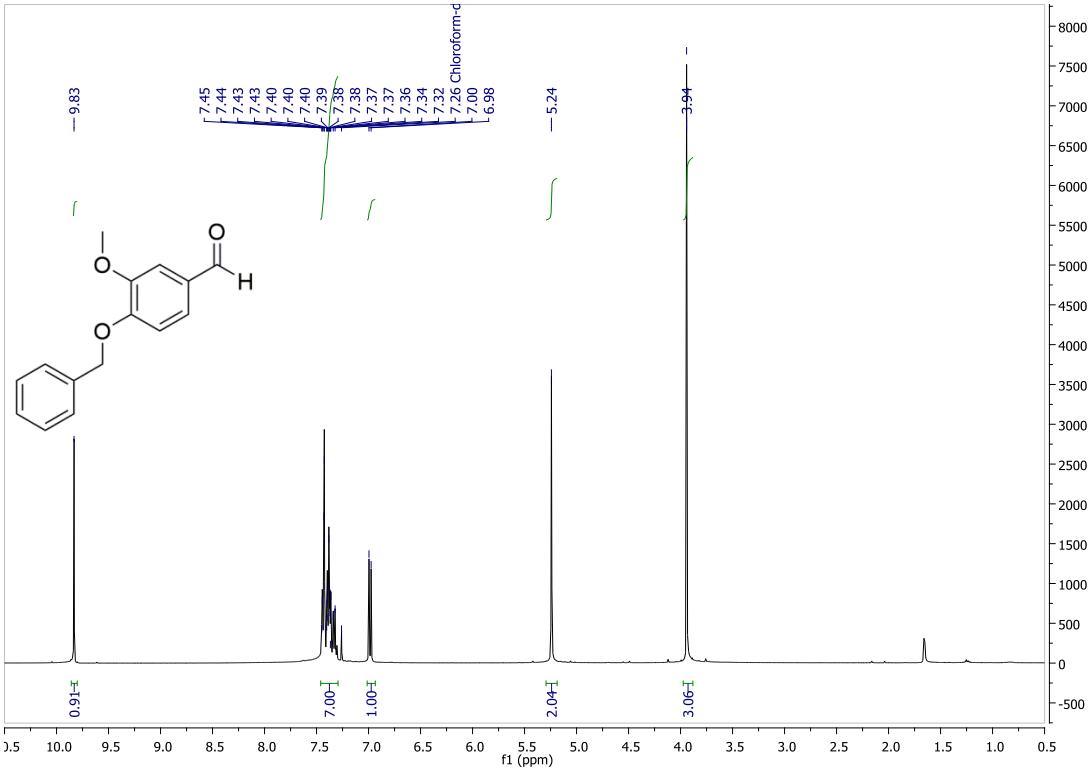

156.89
147.47
145.69
129.58
123.74
81.08
77.16 Chloroform-d
55.06
28.43
NHBoc
NO2
f1 (ppm)

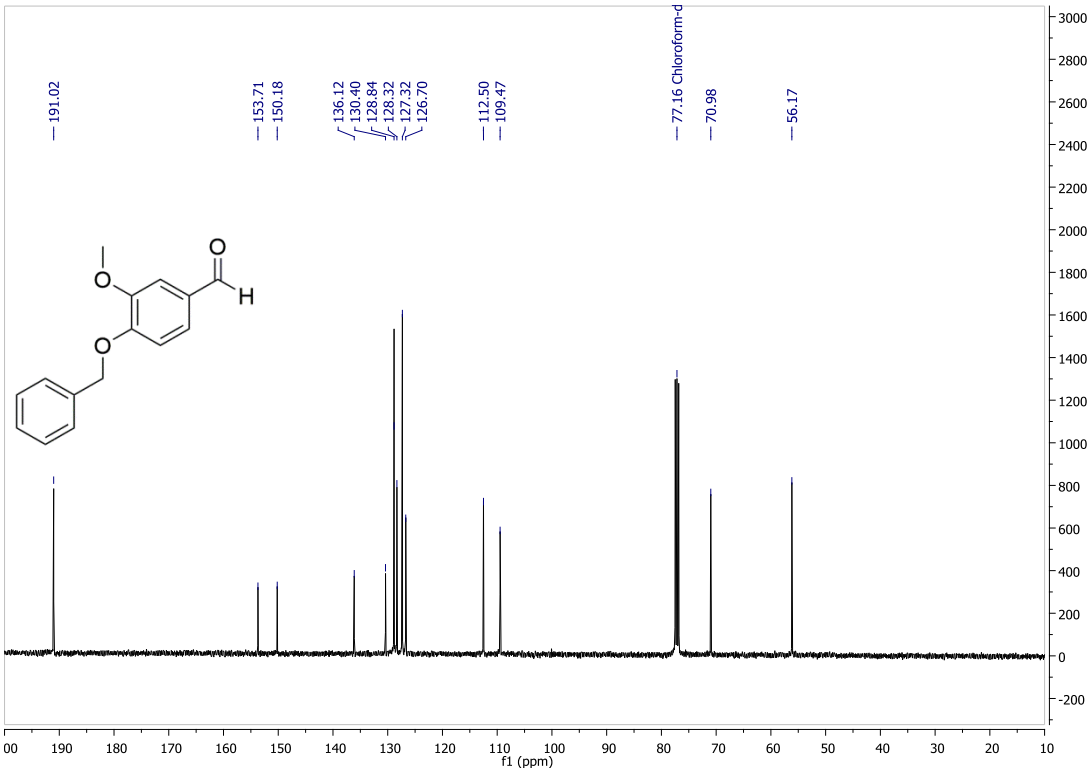

7.26 Chloroform-d
7.13
7.11
6.65
6.63
6.13
3.86
3.68
1.46
NHBoc
NH2
2.00
1.99
0.87
4.96
9.44
f1 (ppm)

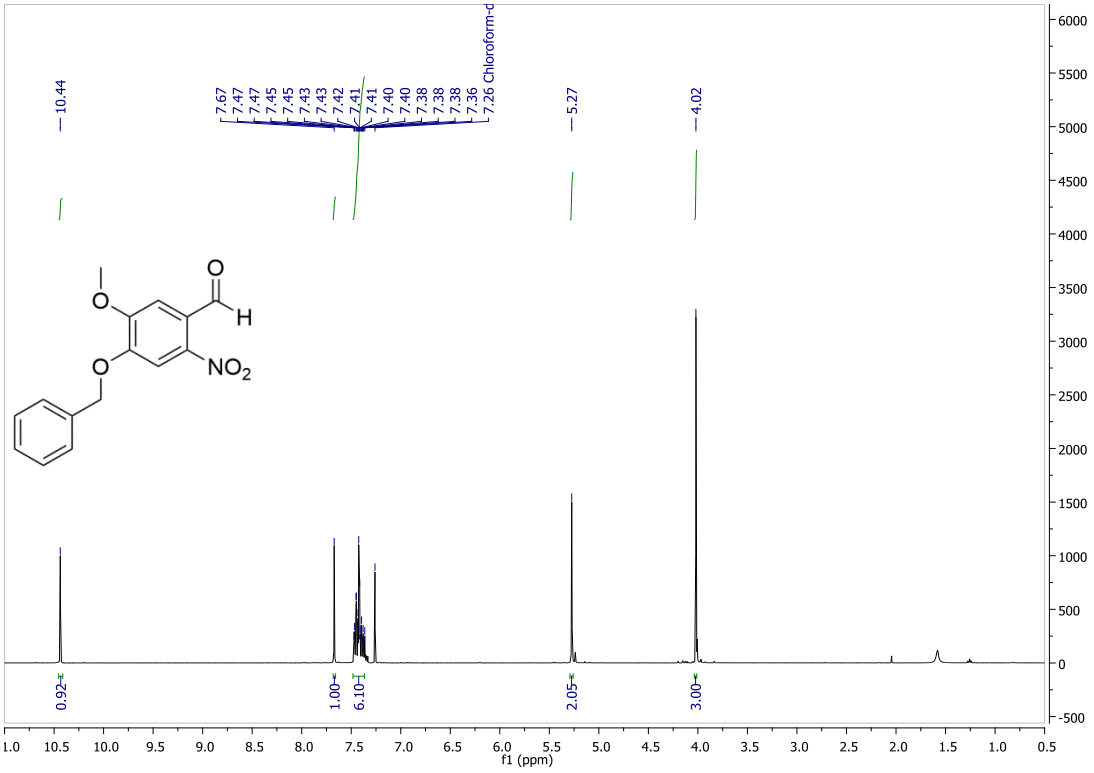

156.76
145.93
130.31
127.55
115.20
80.51
77.16 Chloroform-d
55.56
28.49
NHBoc
NH2
f1 (ppm)

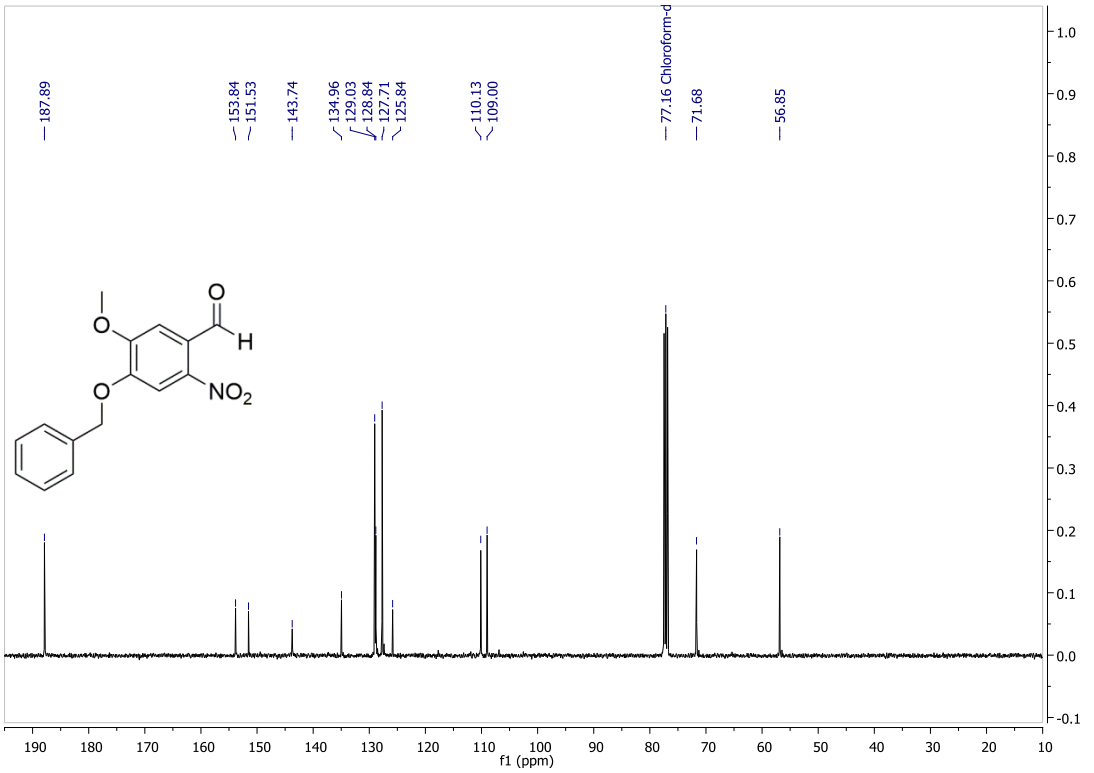

7.66
7.64
7.49
7.47
4.21
3.31 Methanol-d4
2.00
2.10
2.00
f1 (ppm)

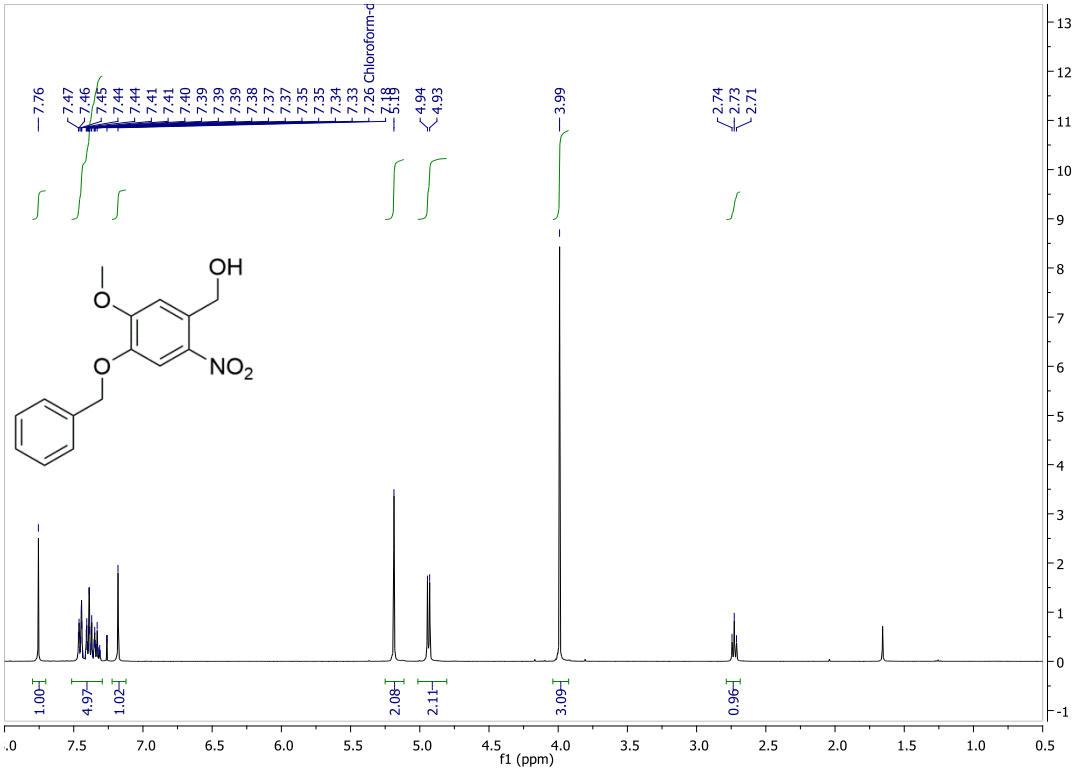

136.28
132.39
132.25
124.73
54.64
49.00 Methanol-d4
f1 (ppm)

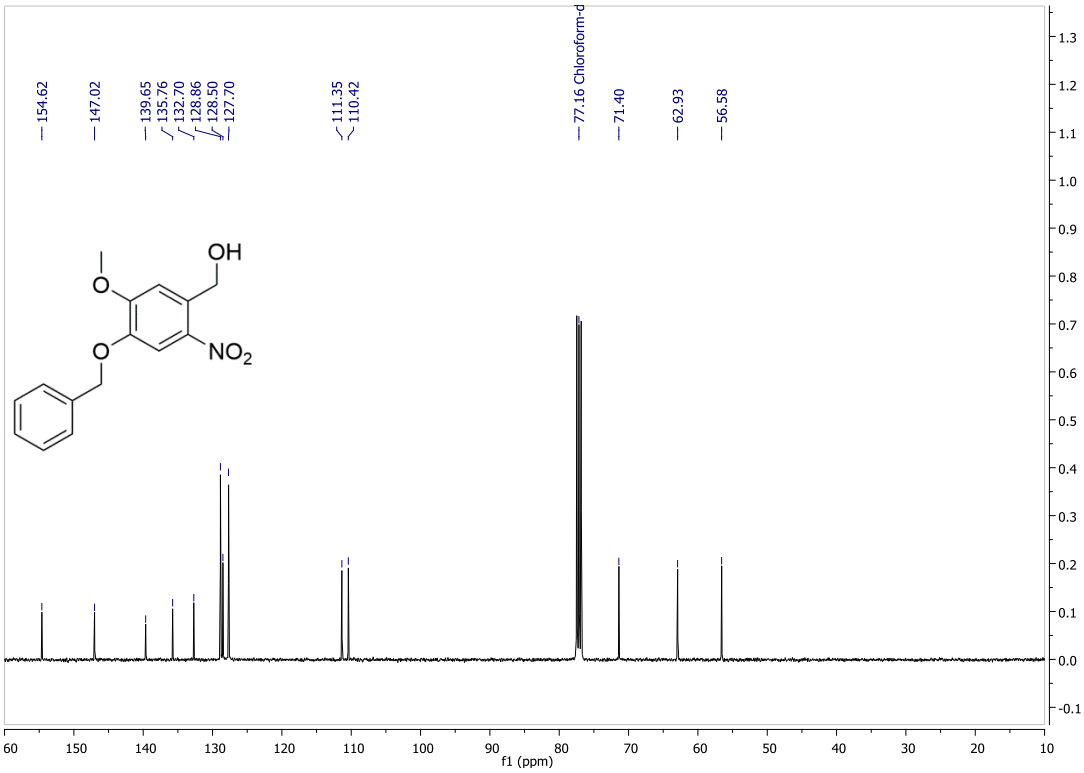

9.83
7.45
7.44
7.43
7.43
7.40
7.40
7.40
7.39
7.38
7.38
7.37
7.37
7.36
7.34
7.32
7.26 Chloroform-d
7.00
6.98
5.24
3.94
0.91
7.00
1.00
2.04
3.06
f1 (ppm)

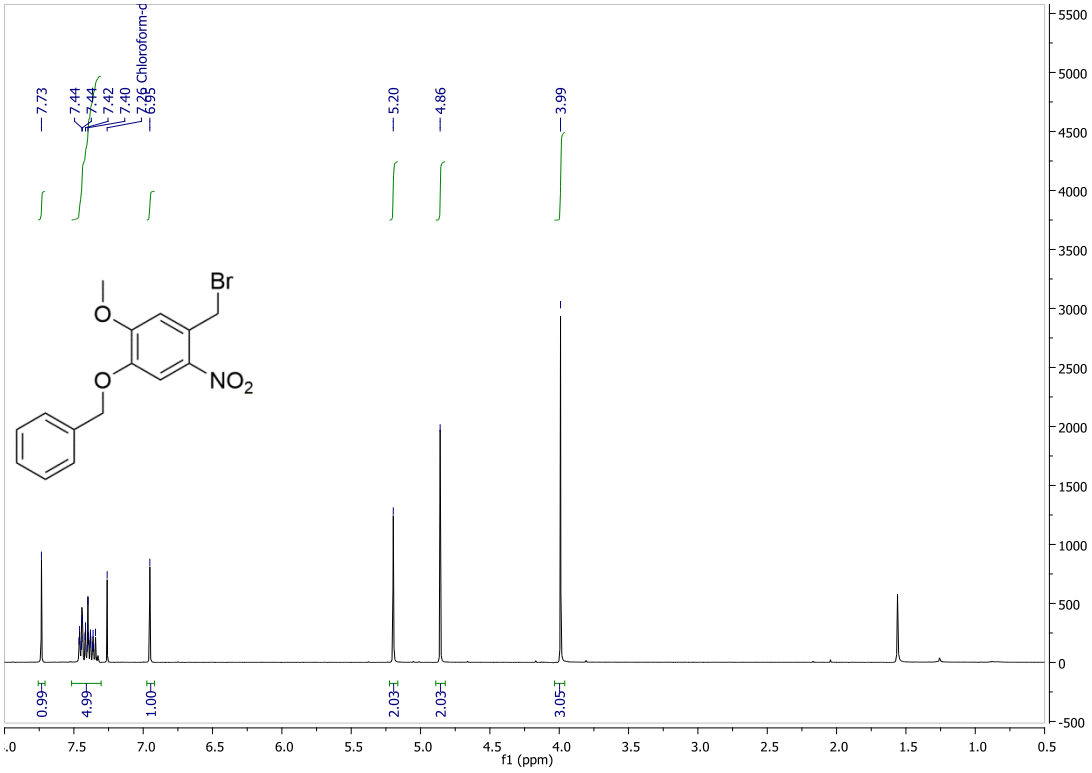

191.02
153.71
150.18
136.12
130.40
128.84
128.32
127.32
126.70
112.50
109.47
77.16 Chloroform-d
70.98
56.17
f1 (ppm)

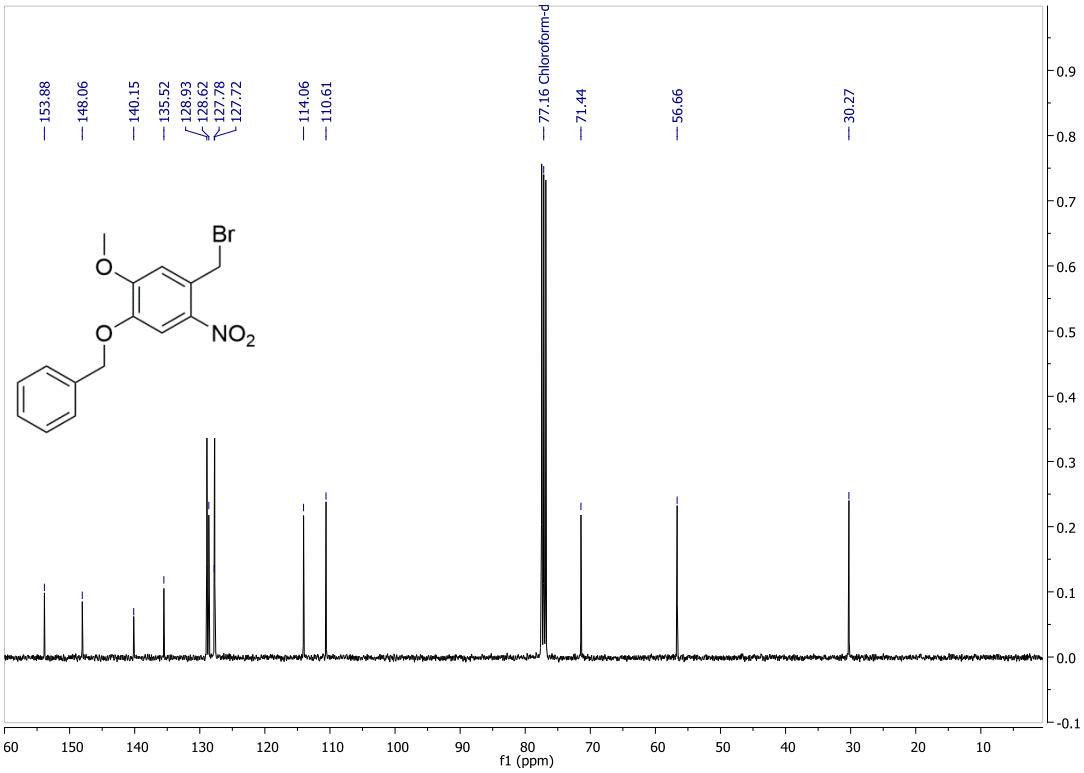

10.44
7.67
7.47
7.47
7.45
7.45
7.43
7.43
7.42
7.41
7.41
7.40
7.40
7.38
7.38
7.38
7.36
7.26 Chloroform-d
5.27
4.02
O
H
O
O
NO2
0.92
1.00
6.10
2.05
3.00
f1 (ppm)

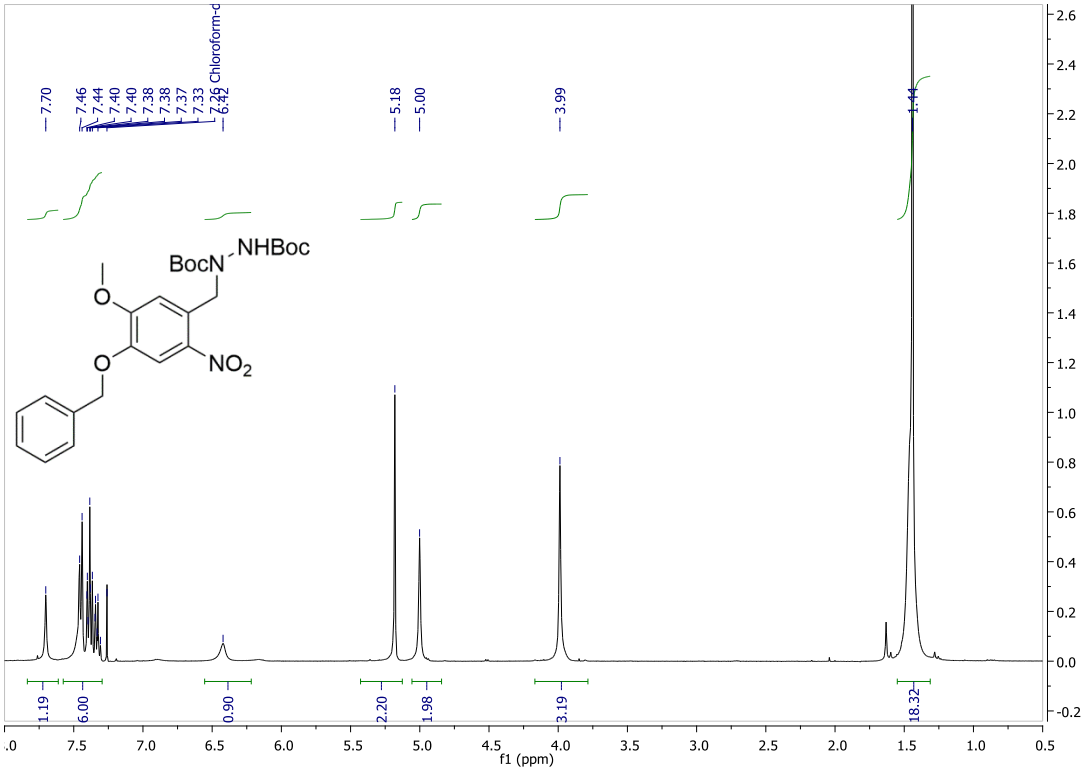

187.89
153.84
151.53
143.74
134.96
129.03
128.84
127.71
125.84
110.13
109.00
77.16 Chloroform-d
71.68
56.85
O
H
O
O
NO2
f1 (ppm)

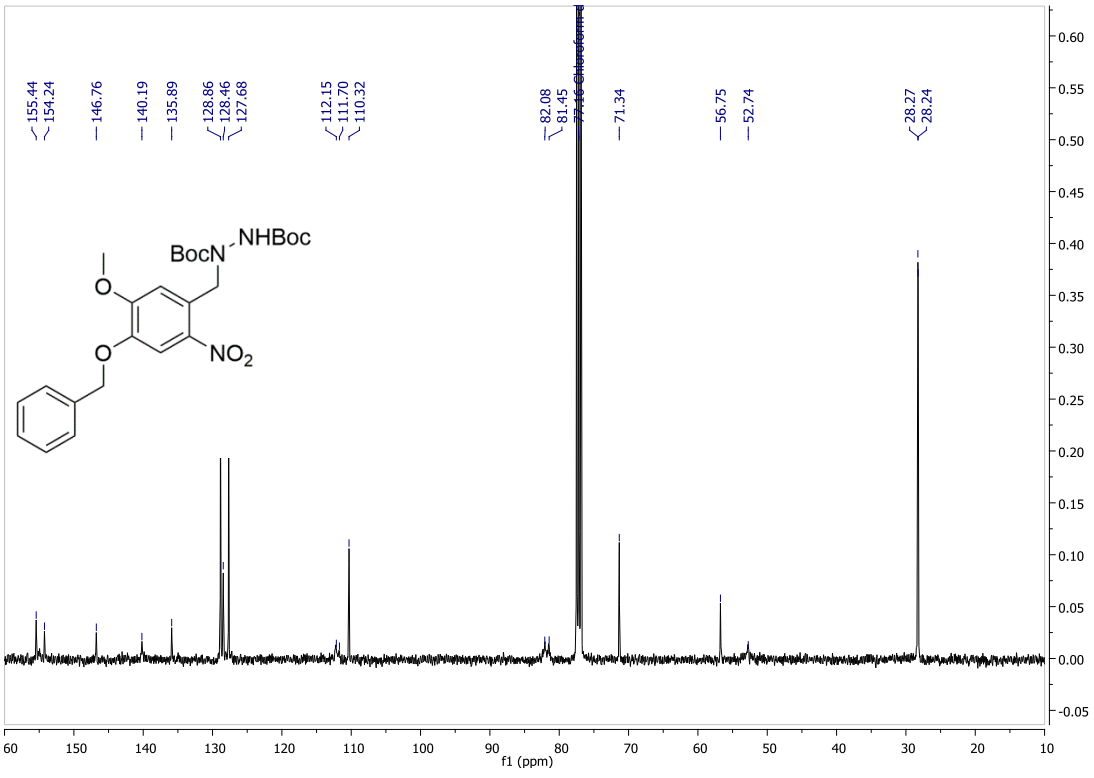

7.76
7.47
7.46
7.45
7.44
7.44
7.41
7.41
7.40
7.39
7.39
7.39
7.38
7.37
7.37
7.35
7.35
7.34
7.33
7.26 Chloroform-d
5.18
4.94
4.93
3.99
2.74
2.73
2.71
OH
O
O
NO2
1.00
4.97
1.02
2.08
2.11
3.09
0.96
f1 (ppm)

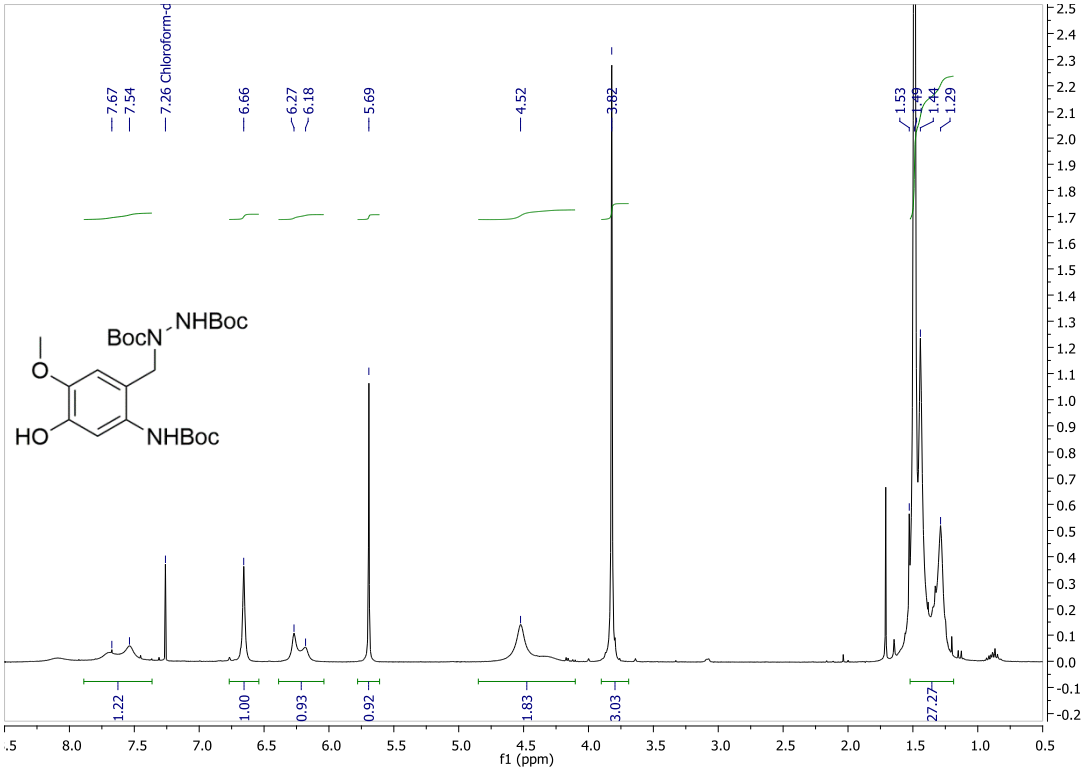

154.62
147.02
139.65
135.76
132.70
128.86
128.50
127.70
111.35
110.42
77.16 Chloroform-d
71.40
62.93
56.58
OH
O
O
NO2
f1 (ppm)


7.73
7.44
7.44
7.42
7.40
7.26 Chloroform-d
6.95
5.20
4.86
3.99
Br
O
O
NO2
0.99
4.99
1.00
2.03
2.03
3.05
f1 (ppm)


153.88
148.06
140.15
135.52
128.93
128.62
127.78
127.72
114.06
110.61
77.16 Chloroform-d
71.44
56.66
30.27
Br
O
O
NO2
f1 (ppm)


7.70
7.46
7.44
7.40
7.40
7.38
7.38
7.37
7.33
7.26 Chloroform-d
6.42
5.18
5.00
3.99
1.44
BocN
NHBoc
O
O
NO2
1.19
6.00
0.90
2.20
1.98
3.19
18.32
f1 (ppm)


155.44
154.24
146.76
140.19
135.89
128.86
128.46
127.68
112.15
111.70
110.32
82.08
81.45
77.16 Chloroform-d
71.34
56.75
52.74
28.27
28.24
BocN
NHBoc
O
O
NO2
f1 (ppm)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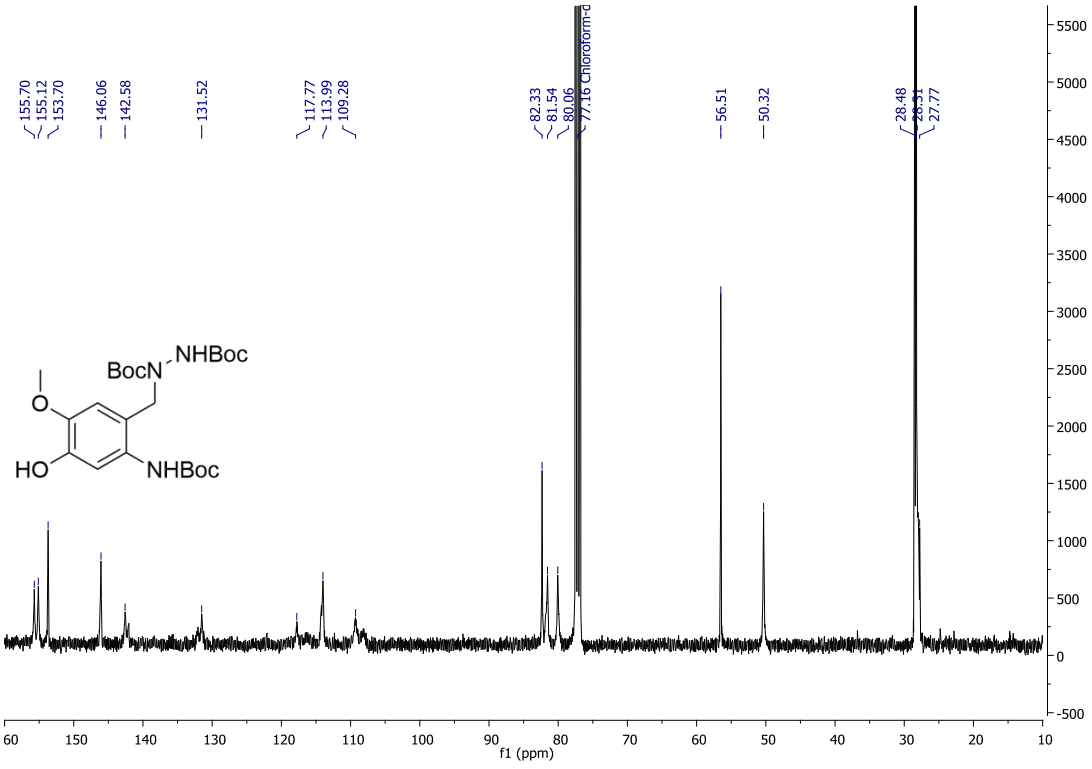
7.54
7.26 Chloroform-d
6.66
6.27
6.18
5.69
4.52
3.82
1.53
1.49
1.44
1.29
BocN
NHBoc
O
HO
NHBoc
1.22
1.00
0.93
0.92
1.83
3.03
27.27
f1 (ppm)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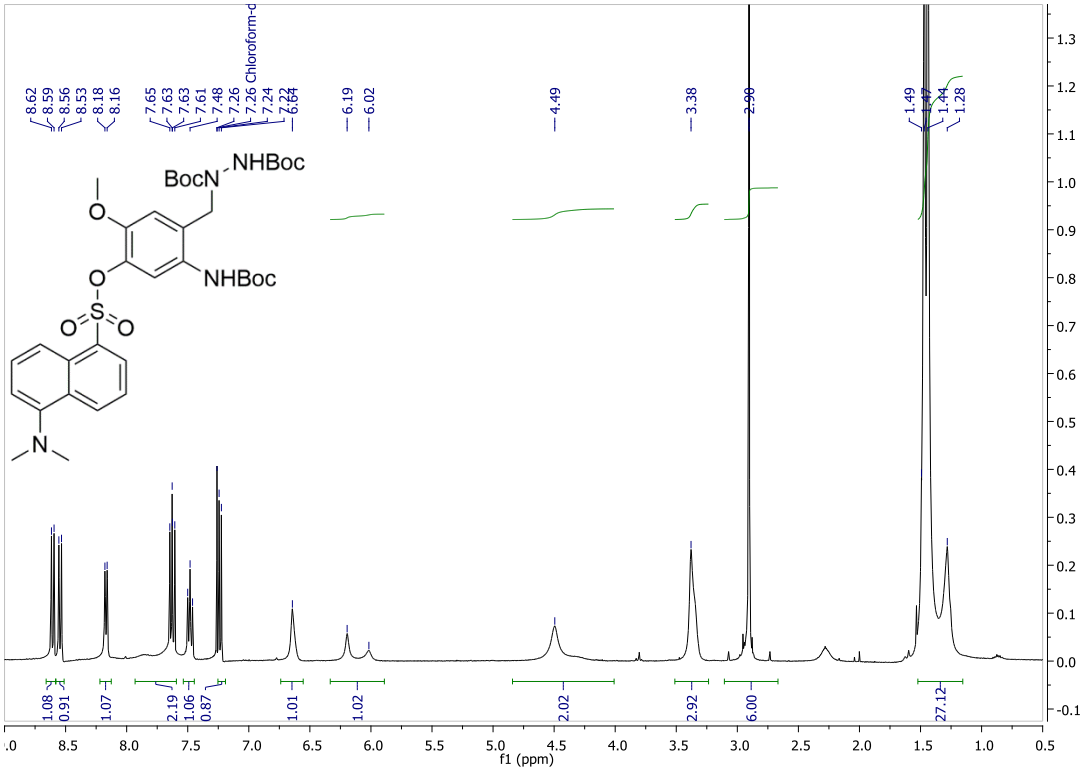
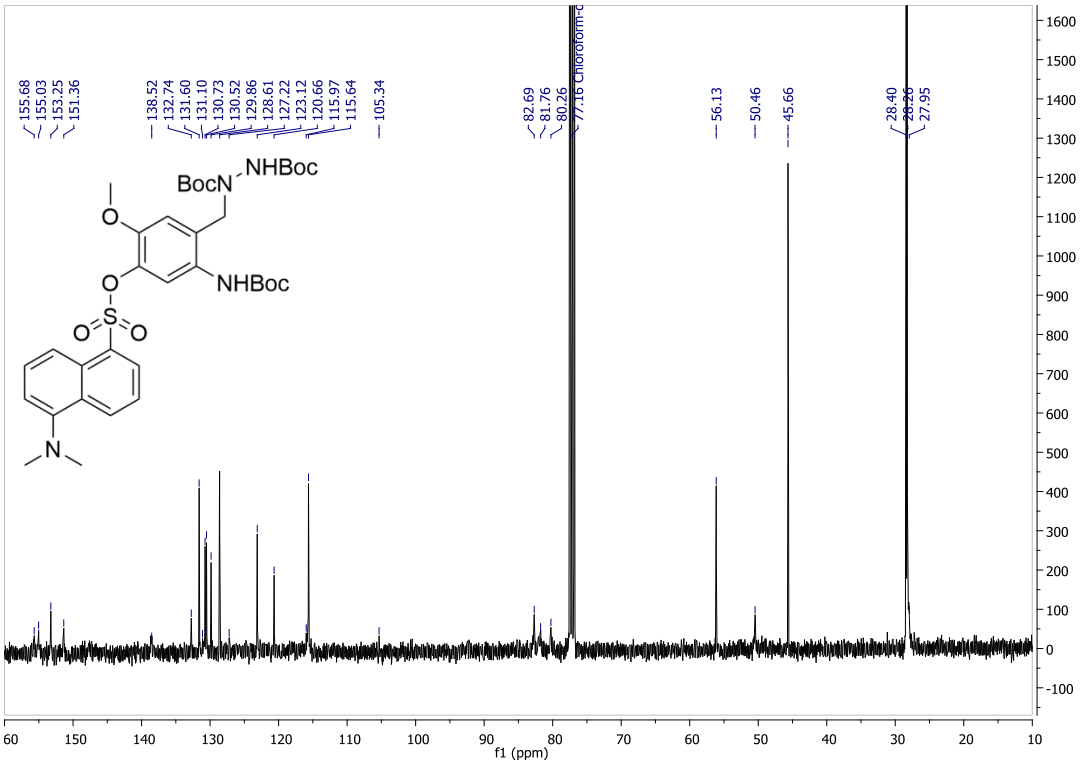
155.12
153.70
146.06
142.58
131.52
117.77
113.99
109.28
82.33
81.54
80.06
77.16 Chloroform-d
56.51
50.32
28.48
28.31
27.77
BocN
NHBoc
O
HO
NHBoc
f1 (ppm)


8.62
8.59
8.56
8.53
8.18
8.16
7.65
7.63
7.63
7.61
7.48
7.26 Chloroform-d
7.24
7.22
6.64
6.19
6.02
4.49
3.38
2.90
1.49
1.47
1.44
1.28
BocN
NHBoc
NHBoc
O
O
S
O
O
N
1.08
0.91
1.07
2.19
1.06
0.87
1.01
1.02
2.02
2.92
6.00
27.12
f1 (ppm)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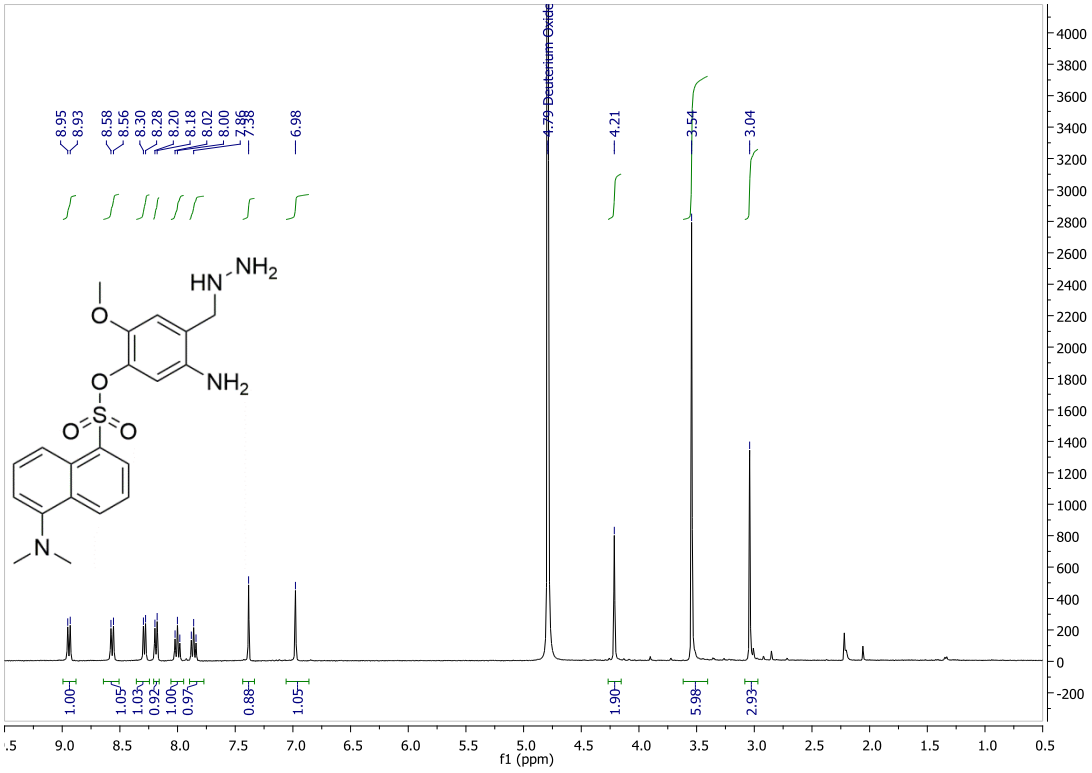
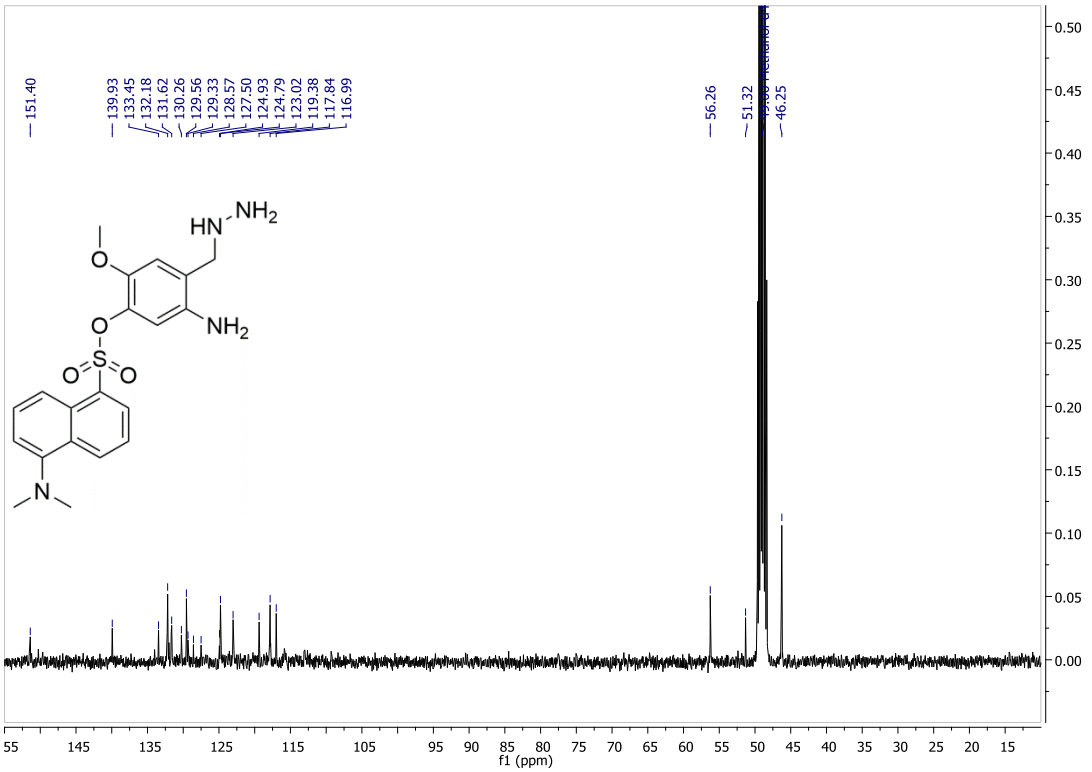
132.74
131.60
131.10
130.73
130.52
129.86
128.61
127.22
123.12
120.66
115.97
115.64
105.34
82.69
81.76
80.26
77.16 Chloroform-d
56.13
50.46
45.66
28.40
28.26
27.95
BocN
NHBoc
NHBoc
O
O
S
O
O
N
f1 (ppm)


8.95
8.93
8.58
8.56
8.30
8.28
8.20
8.18
8.02
8.00
7.86
7.38
6.98
4.79 Deuterium Oxide
4.21
3.54
3.04
HN
NH2
NH2
O
O
S
O
O
N
1.00
1.05
1.03
0.92
1.00
0.97
0.88
1.05
1.90
5.98
2.93
f1 (ppm)


151.40
139.93
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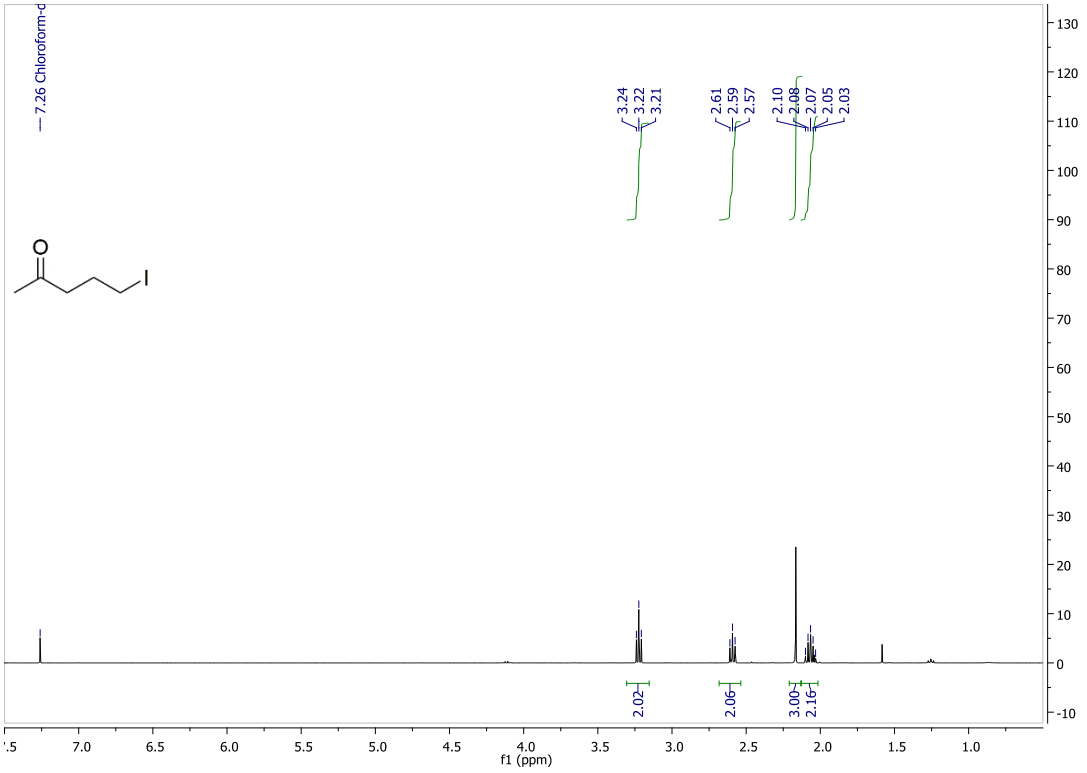
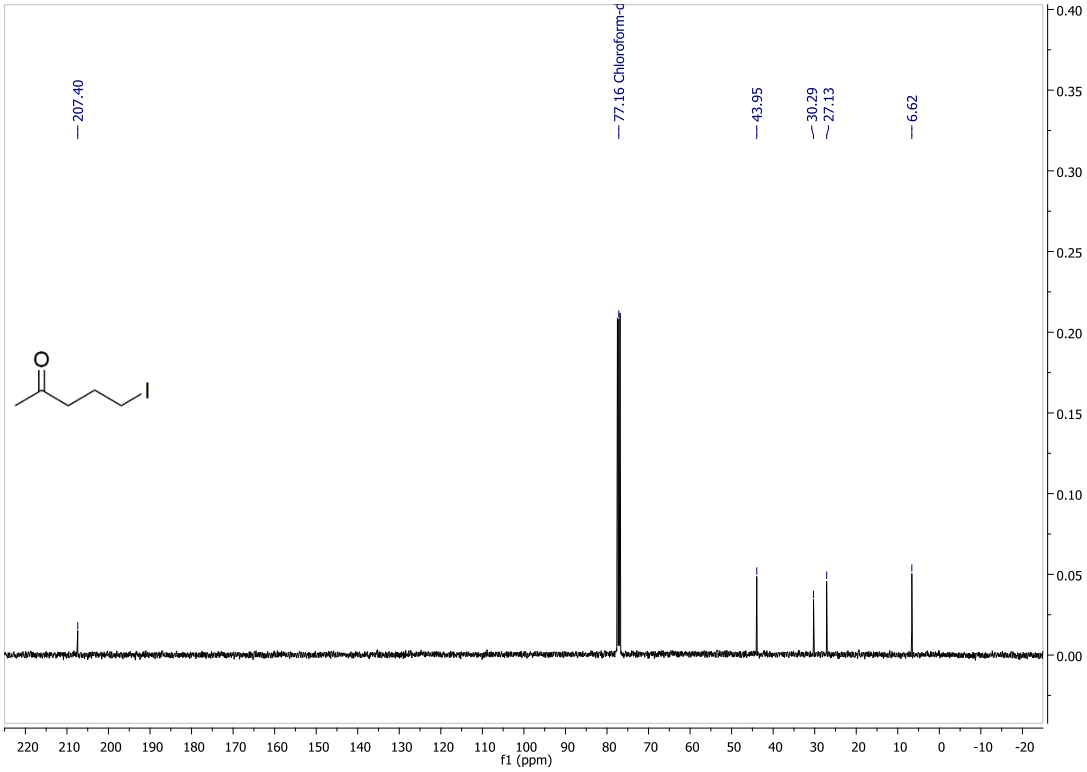
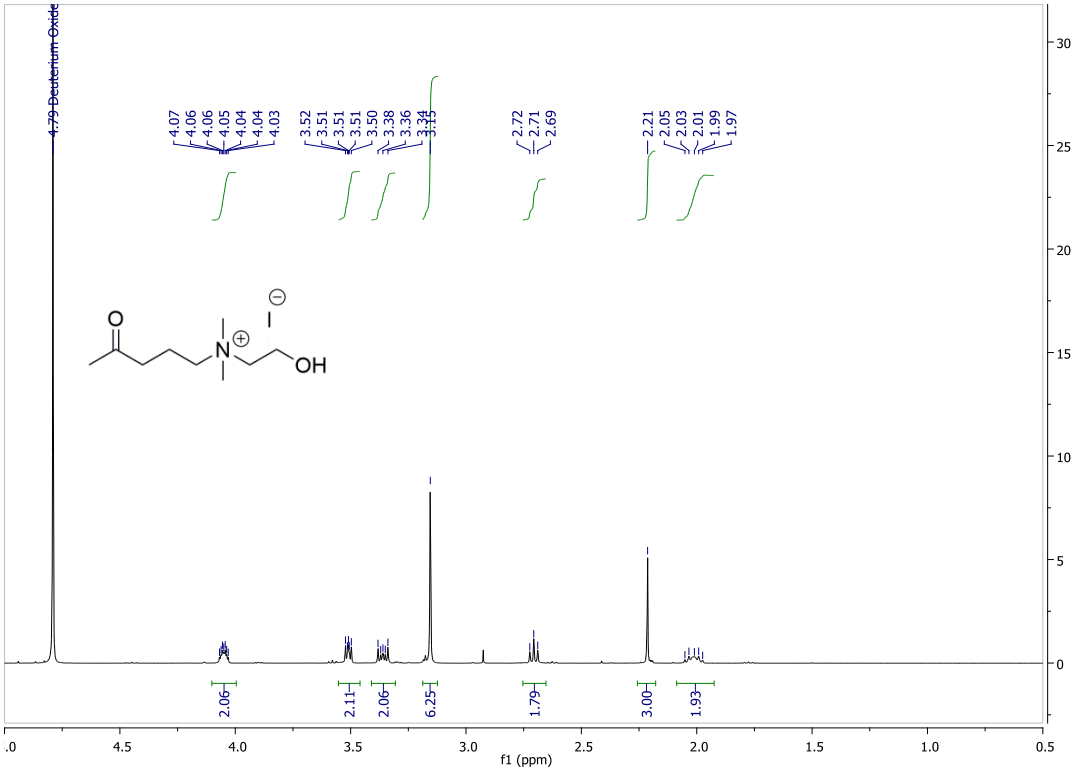
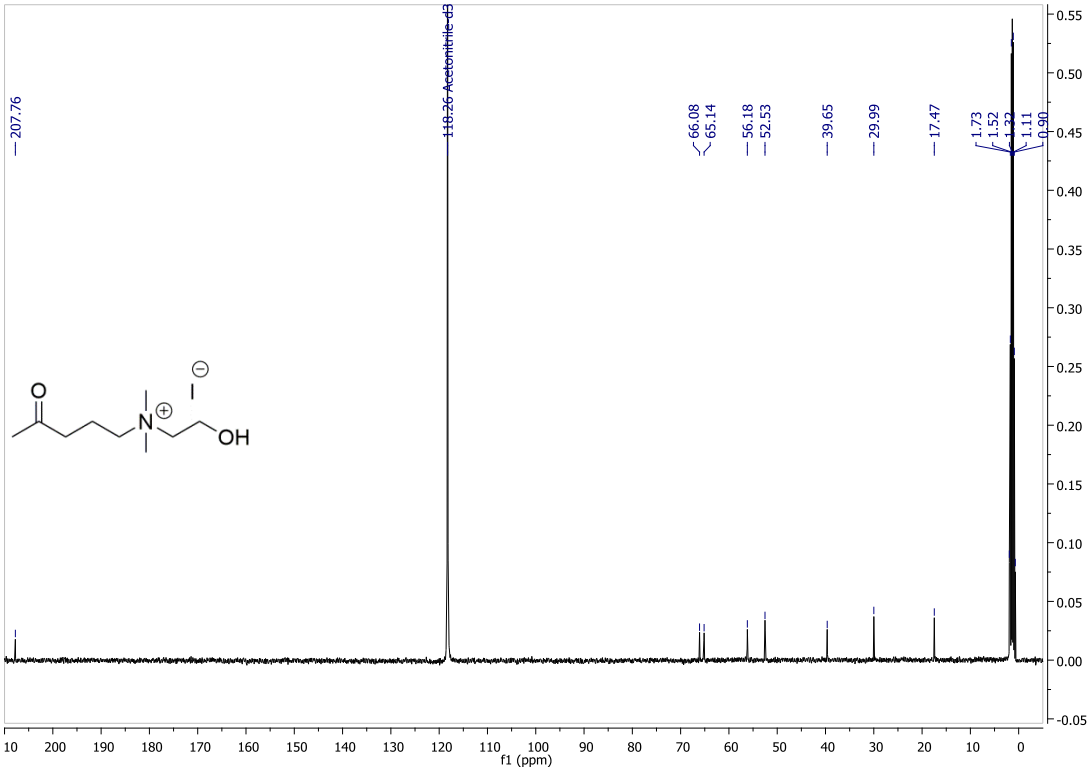
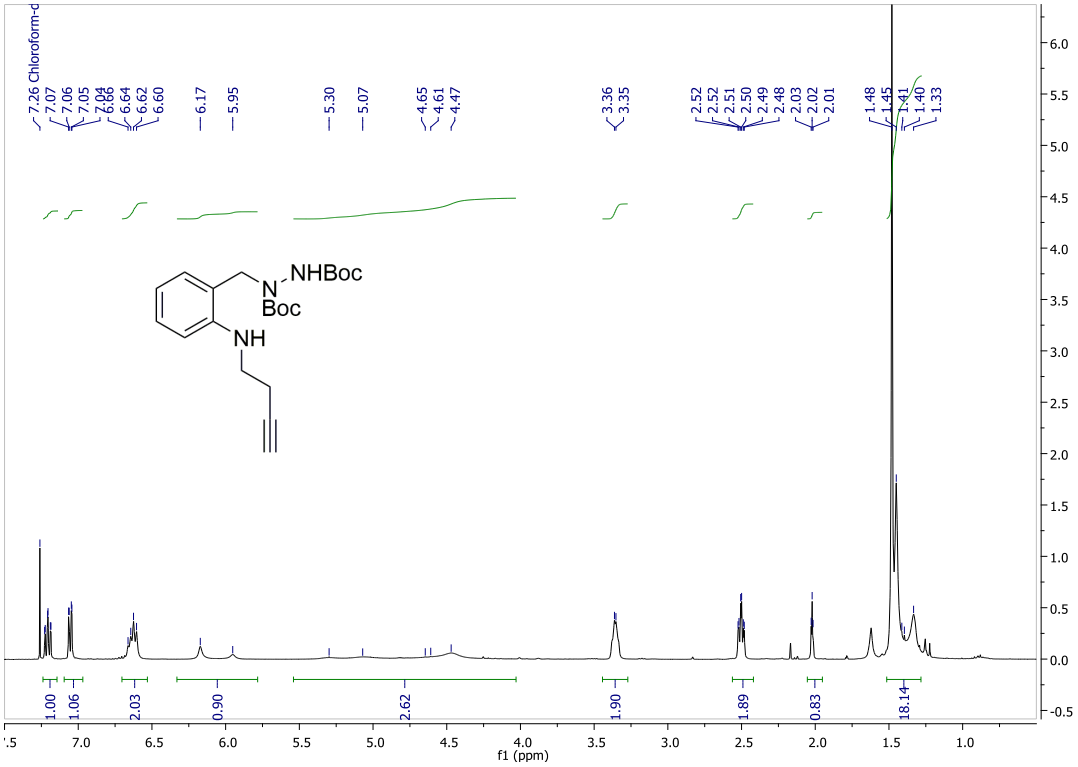
132.18
131.62
130.26
129.56
129.33
128.57
127.50
124.93
124.79
123.02
119.38
117.84
116.99
56.26
51.32
46.25
HN
NH2
NH2
O
O
S
O
O
N
f1 (ppm)

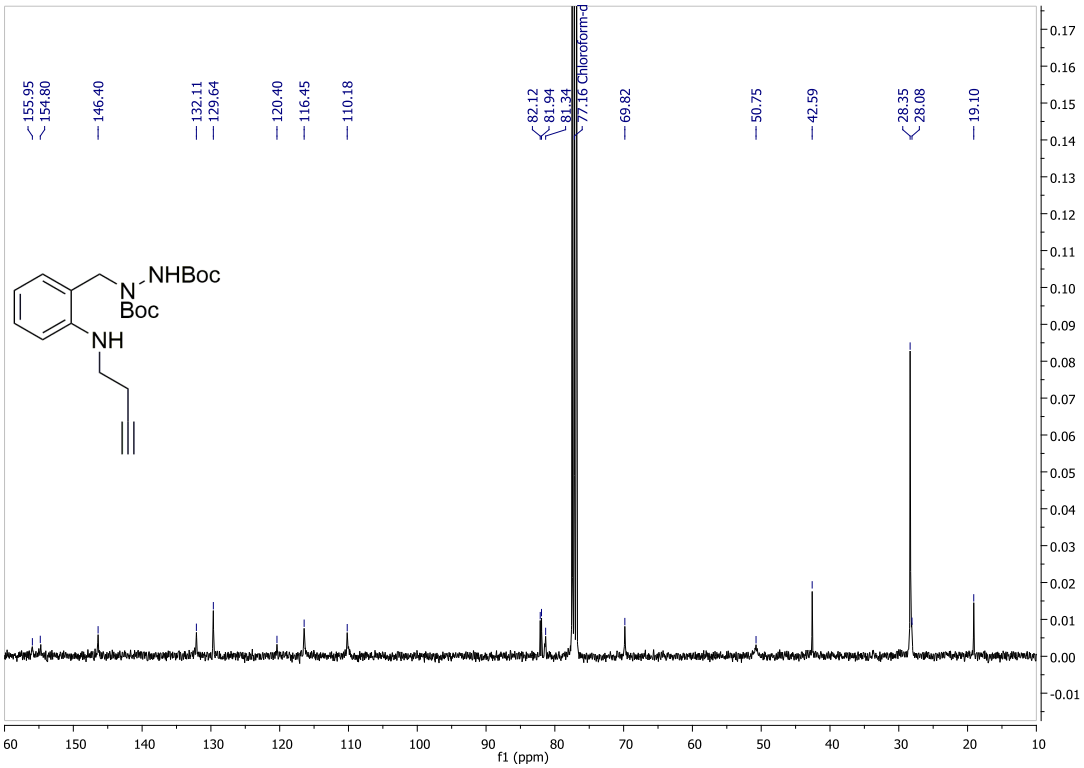

7.26 Chloroform-d
3.24
3.22
3.21
2.61
2.59
2.57
2.10
2.08
2.07
2.05
2.03
2.02
2.06
3.00
2.16
f1 (ppm)

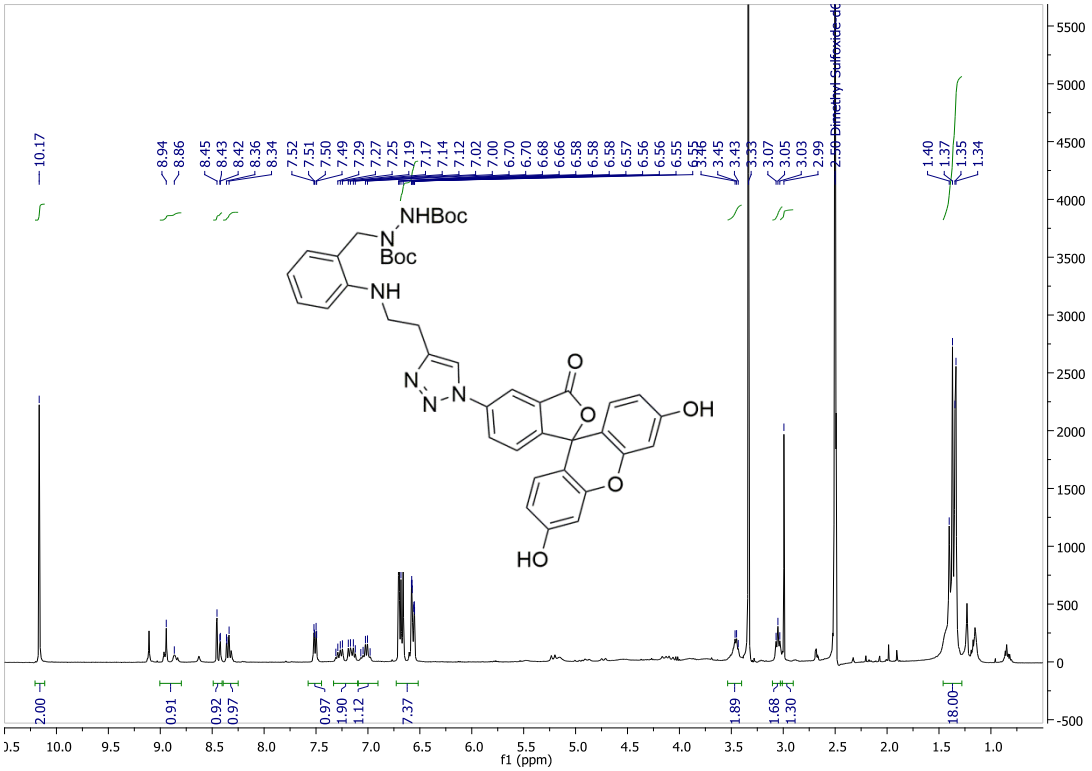

207.40
77.16 Chloroform-d
43.95
30.29
27.13
6.62
f1 (ppm)

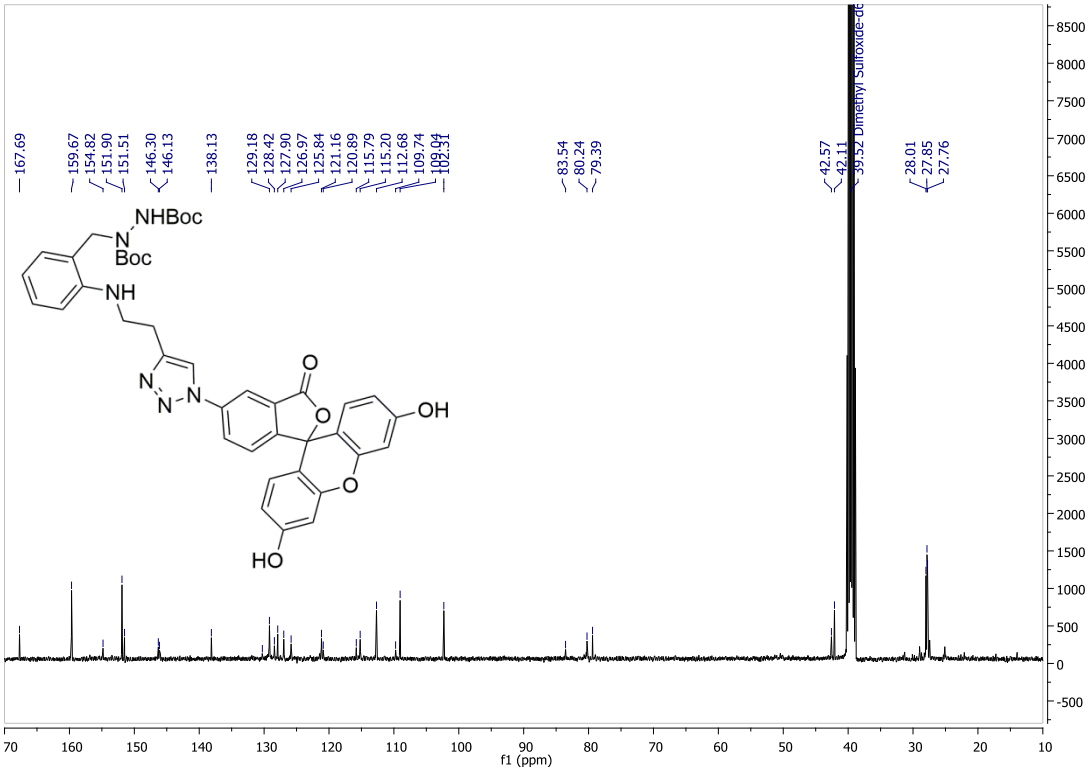

4.79 Deuterium Oxide
4.07
4.06
4.06
4.05
4.04
4.04
4.03
3.52
3.51
3.51
3.51
3.50
3.38
3.36
3.34
3.15
2.72
2.71
2.69
2.21
2.05
2.03
2.01
1.99
1.97
O
N
I
OH
2.06
2.11
2.06
6.25
1.79
3.00
1.93
f1 (ppm)

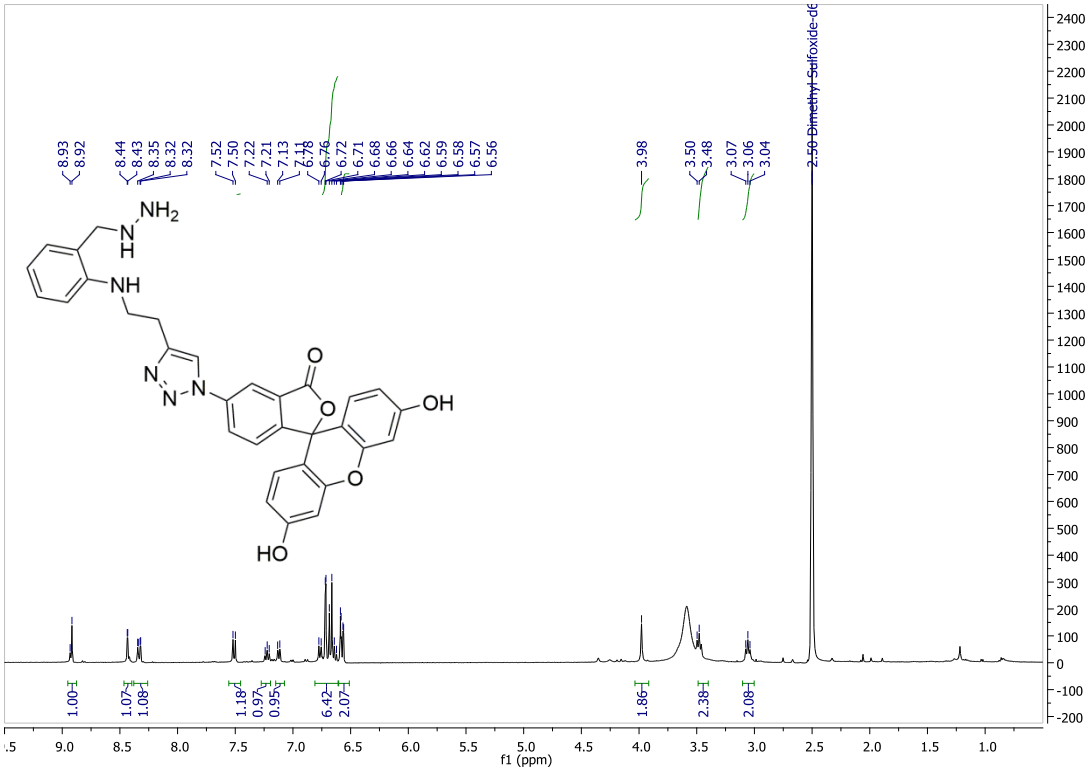

207.76
118.26 Acetonitrile-d3
66.08
65.14
56.18
52.53
39.65
29.99
17.47
1.73
1.52
1.33
1.11
0.90
O
N
I
OH
f1 (ppm)

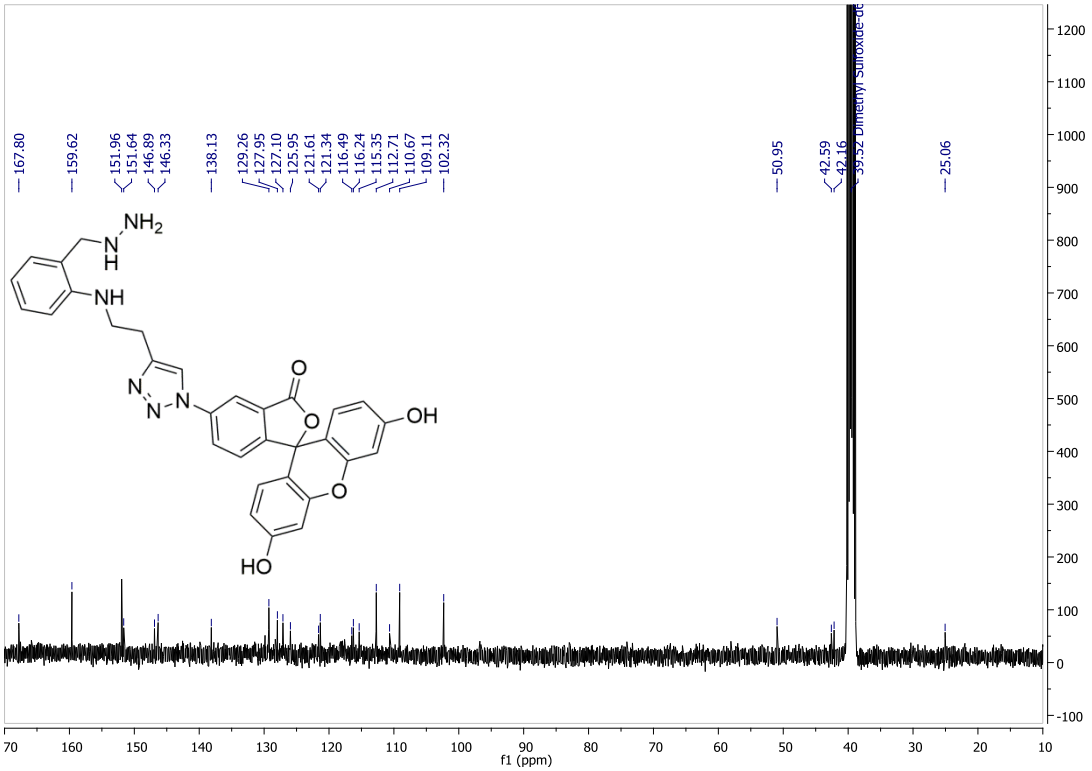

7.26 Chloroform-d
7.07
7.06
7.05
7.04
6.64
6.62
6.60
6.17
5.95
5.30
5.07
4.65
4.61
4.47
3.36
3.35
2.52
2.51
2.50
2.49
2.48
2.03
2.02
2.01
1.48
1.45
1.41
1.40
1.33
NHBoc
N
Boc
NH
1.00
1.06
2.03
0.90
2.62
1.90
1.89
0.83
18.14
f1 (ppm)


155.95
154.80
146.40
132.11
129.64
120.40
116.45
110.18
82.12
81.94
81.34
77.16 Chloroform-d
69.82
50.75
42.59
28.35
28.08
19.10
NHBoc
N
Boc
NH
f1 (ppm)


10.17
8.94
8.86
8.45
8.43
8.42
8.36
8.34
7.52
7.51
7.50
7.49
7.29
7.27
7.25
7.19
7.17
7.14
7.12
7.02
7.00
6.70
6.70
6.68
6.66
6.58
6.58
6.58
6.57
6.56
6.56
6.55
3.45
3.43
3.33
3.07
3.05
3.03
2.99
2.50 Dimethyl Sulfoxide-d6
1.40
1.37
1.35
1.34
NHBoc
N
Boc
NH
N
N
N
O
O
OH
O
HO
2.00
0.91
0.92
0.97
0.97
1.90
1.12
7.37
1.89
1.68
1.30
18.00
f1 (ppm)


167.69
159.67
154.82
151.90
151.51
146.30
146.13
138.13
129.18
128.42
127.90
126.97
125.84
121.16
120.89
115.79
115.20
112.68
109.74
109.04
102.31
83.54
80.24
79.39
42.57
42.11
39.52 Dimethyl Sulfoxide-d6
28.01
27.85
27.76
NHBoc
N
Boc
NH
N
N
N
O
O
OH
O
HO
f1 (ppm)


8.93
8.92
8.44
8.43
8.35
8.32
8.32
7.52
7.50
7.22
7.21
7.13
7.11
6.76
6.72
6.71
6.68
6.66
6.64
6.62
6.59
6.58
6.57
6.56
3.98
3.50
3.48
3.07
3.06
3.04
2.50 Dimethyl Sulfoxide-d6
NH2
NH
OH
HO
1.00
1.07
1.08
1.18
0.97
0.95
6.42
2.07
1.86
2.38
2.08
f1 (ppm)


167.80
159.62
151.96
151.64
146.89
146.33
138.13
129.26
127.95
127.10
125.95
121.61
121.34
116.49
116.24
115.35
112.71
110.67
109.11
102.32
50.95
42.59
42.16
39.52 Dimethyl Sulfoxide-d6
25.06
NH2
NH
OH
HO
f1 (ppm)

15. References

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