Designed small-molecule inhibitors of the anthranilyl-CoA synthetase PqsA block quinolone biosynthesis in *Pseudomonas aeruginosa*

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Supporting Information

A. SUPPLEMENTARY FIGURES S1 AND S2

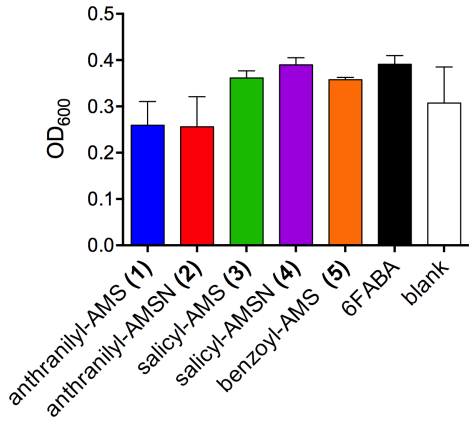


Figure S1. *P. aeruginosa* PA14 growth is not affected by PqsA inhibitors (1.5 mM) at 20 h.

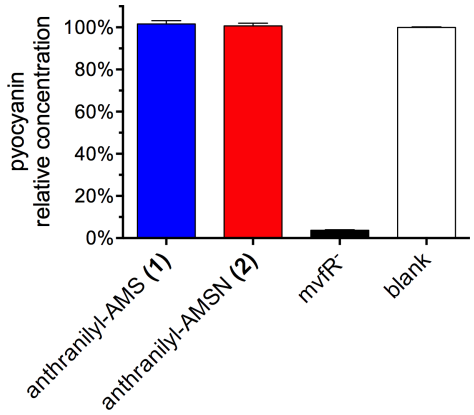


Figure S2. *P. aeruginosa* PA14 production of pyocyanin is not inhibited by PqsA inhibitors (1.0 mM) at 24 h.

B. MATERIALS AND METHODS

Chemical Synthesis

Reagents were obtained from Aldrich Chemical (www.sigma-aldrich.com) or Acros Organics (www.fishersci.com) and used without further purification. Optima or HPLC grade solvents were obtained from Fisher Scientific (www.fishersci.com), degassed with Ar, and purified on a solvent drying system. Reactions were performed in flame-dried glassware under positive Ar pressure with magnetic stirring.

TLC was performed on 0.25 mm E. Merck silica gel 60 F254 plates and visualized under UV light (254 nm) or by staining with potassium permanganate (KMnO4), cerium ammonium molybdenate (CAM), or iodine (I_2). Silica flash chromatography was performed on E. Merck 230–400 mesh silica gel 60. Samples were lyophilized using a Labconco Freezone 2.5 instrument.

NMR spectra were recorded on a Bruker Avance III 500 instrument or Bruker Avance III 600 instrument at 24 °C in $CDCl_3$ unless otherwise indicated. Spectra were processed using Bruker TopSpin or nucleomatica iNMR (www.inmr.net) software, and chemical shifts are expressed in ppm relative to TMS (^{1}H, 0 ppm) or residual solvent signals: $CDCl_3$ (^{1}H, 7.24 ppm; ^{13}C, 77.23 ppm), CD_3OD (^{1}H, 3.31 ppm; ^{13}C, 49.15 ppm), D_2O (^{1}H, 4.80 ppm); coupling constants are expressed in Hz. Mass spectra were obtained at the MSKCC Analytical Core Facility on a Waters Acuity SQD LC-MS by electrospray (ESI) ionization or atmospheric pressure chemical ionization (AP-CI).

Bacterial Strains

Pseudomonas aeruginosa strain PA14 (UCBPP-PA14) is a RifR human clinical isolate.[1] The mvfR$^-$ mutant is isogenic to UCBPP-PA14.[2]

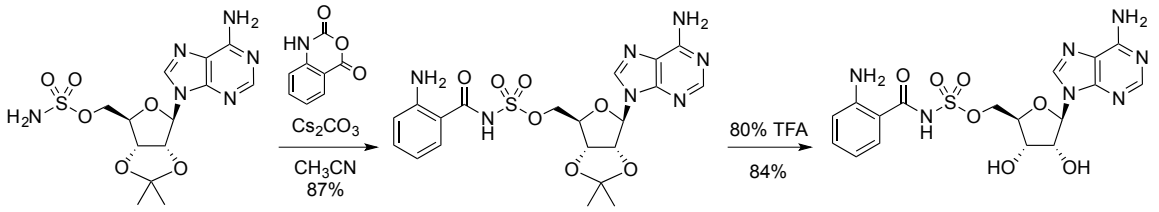
(1) Rahme, L. G.; Stevens, E. J.; Wolfort, S. F.; Shao, J.; Tompkins, R. G.; Ausubel, F. M. *Science* **1995**, *268*, 1899–1902.

(2) Cao, H.; Krishnan, G.; Goumnerov, B.; Tsongalis, J.; Tompkins, R.; Rahme, L. G. *Proc. Natl. Acad. Sci. USA* **2001**, *98*, 14613–14618

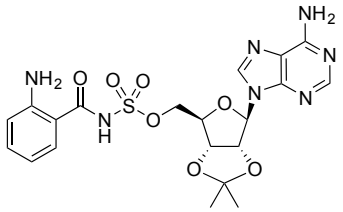
C. SYNTHESIS OF ANTHRANILYL-AMS ANALOGUES

General Procedure for Ion Exchange of Acyl-AMS Final Products

A solution of the acylsulfamate/sulfamide, in protonated form or as a triethylammonium salt, was dissolved in minimum amount of H_2O. The solution was added to a short column of Dowex 50WX8-100-Na^+ and incubated for 10 min, then eluted with H_2O. The fractions containing the product were combined and lyophilized to afford the corresponding sodium salt as a white solid. The Dowex cation exchange resin was converted to the sodium form by sequentially washing the column with MeOH, 1 N aq NaOH, and H_2O until the column washes reached neutral pH.



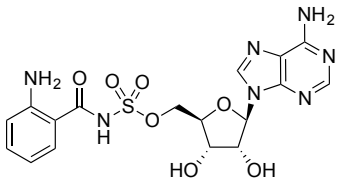
Scheme S1. Synthesis of anthranilyl-AMS (1).



5'-*O*-[*N*-(2-Aminobenzoyl)sulfamoyl]-2',3'-*O*-isopropylideneadenosine (S1). In a 25-mL roundbottom flask, isatoic anhydride (359 mg, 2.2 mmol, 1.1 equiv) was dissolved in DMF (6.0 mL) at room temperature. Isopropylidene sulfamoyl adenosine, prepared as previously described[3] (773 mg, 2.0 mmol, 1.0 equiv) and Cs_2CO_3 (1.3 g, 4.0 mmol, 2.0 equiv) were added. The reaction was stirred for 12 h at room temperature, then concentrated by rotary evaporation. Purification by silica flash chromatography (10–20% MeOH in EtOAc with 1% Et_3N) afforded protected anthranilyl-AMS **S1**, Et_3N salt (879 mg, 87%) as a white solid.

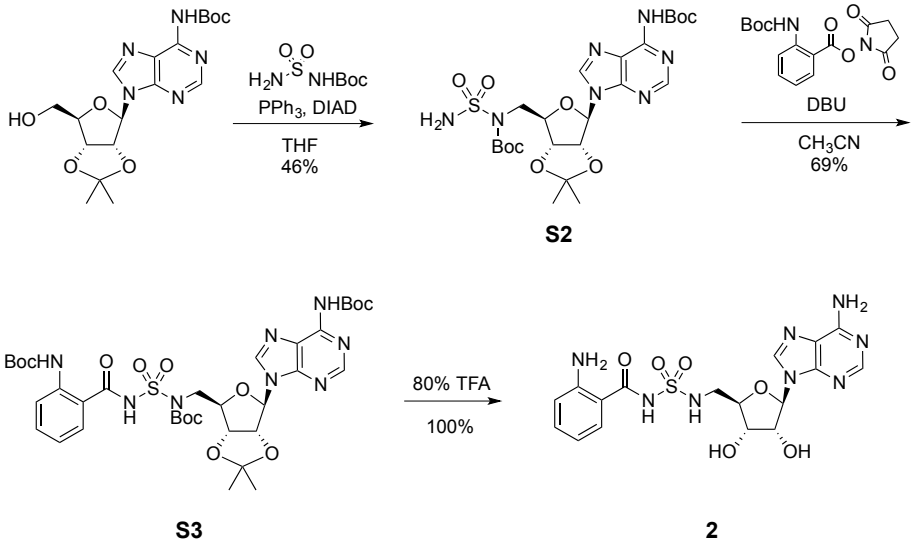
^{1}H NMR (600 MHz, CD_3OD): δ 8.49 (s, 1H), 8.16 (s, 1H), 7.90 (dd, J = 8.0, 1.7 Hz, 1H), 7.09-7.12 (m, 1H), 6.66 (dd, J = 8.2, 1.1 Hz, 1H), 6.52-6.55 (m, 1H), 6.23 (d, J = 3.2 Hz, 1H), 5.39 (dd, J = 6.0, 3.2 Hz, 1H), 5.15 (dd, J = 6.0, 2.2 Hz, 1H), 4.56 (d, J = 2.2 Hz, 1H), 4.20-4.38 (m, 2H), 3.14 (q, J = 7.3 Hz, 4H), 1.59 (s, 3H), 1.34 (s, 3H), 1.24 (t, J = 7.3 Hz, 6H); **^{13}C NMR** (151 MHz, CD_3OD): δ 176.92, 157.26, 153.94, 151.28, 150.43, 141.48, 133.13, 132.58, 120.10, 119.93, 117.94, 116.75, 115.21, 91.91, 85.69, 83.42, 69.61, 47.80, 27.46, 25.46, 9.19; **HRMS (ESI)** *m/z*: calcd. for $C_{20}H_{24}N_7O_7S$ $[M+H]^+$ 506.1458, found 506.1470.

(3) Castro-Pichel, J.; Garcia-Lopez, M. T.; De las Heras, F. G. *Tetrahedron* **1987**, *43*, 383.

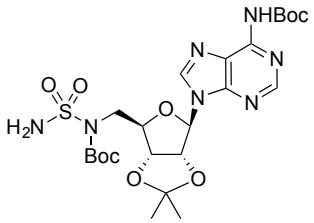


Anthranilyl-AMS (1). In a 25-mL roundbottom flask, protected anthranilyl-AMS **S1** (750 mg, 1.48 mmol) was dissolved in 80% aq TFA (10 mL) and stirred for 2 h. The reaction mixture was concentrated by rotary evaporation. Purification by silica flash chromatography (10–20% MeOH in EtOAc with 1% Et_3N) afforded anthranilyl-AMS as its Et_3N salt (580 mg, 84%). The Et_3N salt was then converted to the corresponding sodium salt using the general procedure for ion-exchange (above) to afford anthranilyl-AMS, sodium salt (500 mg, 100%) as a white solid.

^{1}H NMR (500 MHz, CD_3OD): δ 8.57 (s, 1H), 8.18 (s, 1H), 7.92 (dd, J = 8.0, 1.6 Hz, 1H), 7.11 (ddt, J = 8.5, 7.1, 1.2 Hz, 2H), 6.67 (dd, J = 8.1, 1.1 Hz, 1H), 6.55 (ddd, J = 8.1, 7.0, 1.1 Hz, 1H), 6.10 (d, J = 6.0 Hz, 1H), 4.73 (t, J = 5.5 Hz, 1H), 4.43 (dd, J = 5.1, 3.0 Hz, 1H), 4.29-4.41 (m, 3H); **^{13}C NMR** (151 MHz, D_2O): δ 175.98, 155.36, 152.55, 148.81, 147.73, 139.65, 132.52, 130.45, 119.39, 118.55, 117.46, 117.41, 86.96, 82.40, 73.44, 70.11, 68.48; **HRMS (ESI)** *m/z*: calcd. for $C_{17}H_{18}N_7O_7S$ $[M–H]^-$ 464.0988, found 464.1006.



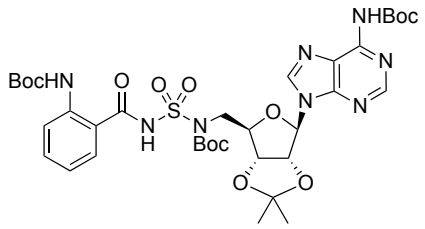
Scheme S2. Synthesis of anthranilyl-AMSN (2).



***tert*-Butyl (((3a*R*,4*R*,6*R*,6a*R*)-6-(6-((*tert*-butoxycarbonyl)amino)-9*H*-purin-9-yl)-2,2-dimethyltetrahydrofuro[3,4-*d*][1,3]dioxol-4-yl)methyl)(sulfamoyl)carbamate (S2)**.

In a 100-mL roundbottom flask, N^6-Boc-2′,3′-*O*-isopropylideneadenosine, prepared as previously described[4] (1.2 g, 2.95 mmol), *N*-(*tert*-butoxycarbonyl)sulfamide (0.75 g, 3.83 mmol, 1.3 equiv) and PPh_3 (1.00 g, 3.83 mmol, 1.3 equiv) were dissolved in anhydrous THF (30 mL) and cooled to 0 ºC. DIAD (0.75 mL, 3.83 mmol, 1.3 equiv) was added dropwise and the resulting mixture was warmed to room temperature and stirred overnight. The reaction mixture was concentrated by rotary evaporation. Purification by silica flash chromatography (0–50% EtOAc in hexanes) yielded protected AMSN **S2** (800 mg, 46%) as a white foam.

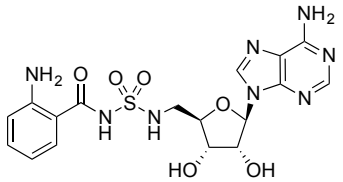
^{1}H NMR (500 MHz, $CDCl_3$): δ 8.75 (s, 1H), 8.02 (s, 1H), 7.92 (s, 1H), 6.14 (d, J = 1.8 Hz, 1H), 5.37-5.46 (m, 1H), 5.32 (s, 2H), 5.14-5.24 (m, 1H), 4.37 (d, J = 5.7 Hz, 1H), 4.06 (dd, J = 15.0, 6.0 Hz, 1H), 3.98 (dd, J = 15.0, 7.3 Hz, 1H), 1.62 (s, 3H), 1.58 (s, 9H), 1.52 (s, 9H), 1.41 (s, 3H); **^{13}C NMR** (151 MHz, $CDCl_3$): δ 153.37, 151.77, 150.47, 150.34, 149.67, 141.73, 122.48, 115.37, 89.93, 85.40, 85.18, 84.50, 82.63, 82.14, 77.44, 77.23, 77.02, 48.38, 28.34, 28.24, 27.45, 25.67; **HRMS (ESI)** *m/z*: calcd. for $C_{23}H_{34}N_7O_9S$ $[M–H]^-$ 584.2139, found 584.2144.



Protected anthranilyl-AMSN (S3). In a 50-mL roundbottom flask, protected AMSN **S2** (800 mg, 1.37 mmol, 1 equiv) and *N*-Boc-anthranilic acid NHS ester, prepared as previously described[5] (502 mg, 1.50 mmol, 1.1 equiv), were dissolved in 20 mL CH_3CN. DBU (420 mg, 2.74 mmol, 2.0 equiv) was added dropwise and the mixture was stirred at room temperature for 3 h. The reaction mixture was concentrated by rotary evaporation and the residue was partitioned between 0.2 N HCl (20 mL) and EtOAc (20 mL). The aqueous layer was extracted with EtOAc (3×20 mL), dried (Na_2SO_4), and concentrated by rotary evaporation. Purification by silica flash chromatography (50–100% EtOAc in hexanes) yielded protected anthranilyl-AMSN **S3** (760 mg, 69%) as a white solid.

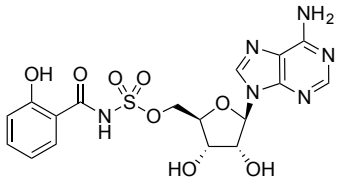
^{1}H NMR (500 MHz, CD_3OD): δ 8.61 (s, 1H), 8.40 (s, 1H), 8.13 (dd, J = 8.4, 1.1 Hz, 1H), 8.06 (dd, J = 8.0, 1.7 Hz, 1H), 7.34 (ddd, J = 8.6, 7.1, 1.7 Hz, 1H), 6.90 (ddd, J = 8.3, 7.2, 1.2 Hz, 1H), 6.20 (d, J = 2.3 Hz, 1H), 5.49 (dd, J = 6.3, 2.3 Hz, 1H), 5.29 (dd, J = 6.3, 3.5 Hz, 1H), 4.56 (td, J = 6.5, 3.5 Hz, 1H), 4.18 (dd, J = 14.9, 6.8 Hz, 1H), 3.92 (dd, J = 14.9, 6.2 Hz, 1H), 1.58 (s, 9H), 1.55 (s, 3H), 1.38 (s, 9H), 1.37 (s, 9H), 1.33 (s, 3H); **^{13}C NMR** (126 MHz, CD_3OD): δ 155.25, 154.61, 153.37, 152.43, 152.04, 151.45, 144.34, 141.67, 133.19, 132.68, 124.09, 123.55, 122.10, 119.73, 115.50, 91.62, 87.16, 85.78, 85.40, 83.94, 82.89, 80.98, 28.66, 28.47, 28.36, 27.54, 25.58. **HRMS (ESI)** *m/z*: calcd. for $C_{35}H_{47}N_8O_{12}S$ $[M–H]^-$ 803.3034, found 803.3000.

(4) Lu, X.; Zhang, H.; Tonge, P. J.; Tan, D. S. *Bioorg. Med. Chem. Lett*. **2008**, *18*, 5963–5966.
(5) Tanaka F.; Kinoshita, K.; Tanimura, R.; Fujii, I. *J. Am. Chem. Soc.* **1996**, *118*, 2332–2339.



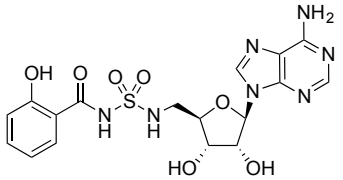
Anthranilyl-AMSN (2). In a 25-mL roundbottom flask, protected anthranilyl-AMSN **S3** (700 mg, 0.87 mmol) was dissolved in 80% aq TFA (5 mL). After 2 h, the reaction mixture was diluted with water (20 mL) and extracted with ether (5×25 mL). The aqueous layer was lyophilized to afford anthranilyl-AMSN **2** (400 mg, 100%) as a white solid.

^{1}H NMR (500 MHz, CD_3OD): δ 8.51 (s, 1H), 8.39 (s, 1H), 7.51 (dd, J = 8.2, 1.5 Hz, 1H), 7.15-7.32 (m, 1H), 6.74 (d, J = 8.4 Hz, 1H), 6.51-6.66 (m, 1H), 5.95 (d, J = 6.8 Hz, 1H), 4.80-4.83 (m, 1H), 4.34 (dd, J = 5.4, 2.6 Hz, 1H), 4.27-4.32 (m, 1H), 3.42-3.51 (m, 1H), 3.35 (d, J = 3.2 Hz, 1H); **^{13}C NMR** (151 MHz, CD_3OD): δ 169.88, 153.01, 151.97, 149.47, 147.39, 144.68, 135.03, 129.99, 121.39, 118.29, 116.95, 113.24, 91.79, 85.98, 74.96, 73.03, 46.24; **HRMS (ESI)** *m/z*: calcd. for $C_{17}H_{21}N_8O_6S$ $[M+H]^+$ 465.1305, found 465.1300.



Salicyl-AMS (3). This compound was prepared as previously described[6] and converted to sodium salt using the general procedure for ion exchange.

^{1}H NMR (600 MHz, D_2O): δ 8.18 (s, 1H), 8.06 (s, 1H), 7.55 (dd, J = 7.9, 1.7 Hz, 1H), 7.27 (ddd, J = 8.3, 7.2, 1.8 Hz, 1H), 6.71 (ddd, J = 8.2, 7.2, 1.1 Hz, 1H), 6.64 (dd, J = 8.3, 1.2 Hz, 1H), 5.91 (d, J = 5.2 Hz, 1H), 4.76 (t, J = 5.4 Hz, 1H), 4.45-4.51 (m, 3H), 4.35 (td, J = 4.5, 2.9 Hz, 1H); **^{13}C NMR** (151 MHz, D_2O): δ 174.47, 158.57, 155.24, 152.40, 148.63, 139.75, 134.07, 129.38, 119.13, 118.56, 117.88, 116.39, 87.33, 82.15, 73.16, 70.01, 69.32; **HRMS (ESI)** *m/z*: calcd. for $C_{17}H_{17}N_6O_8S$ $[M–H]^–$ 465.0829, found 465.0812.



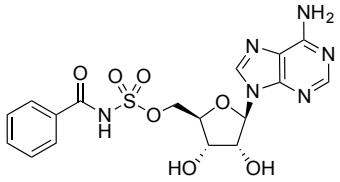
Salicyl-AMSN (4). This compound was prepared as previously described[7] and converted to sodium salt using the general procedure for ion exchange.

^{1}H NMR (500 MHz, D_2O): δ 8.22 (s, 1H), 8.19 (s, 1H), 7.63-7.74 (m, 1H), 7.35 (t,J = 7.8 Hz, 1H), 6.84 (t, J = 7.6 Hz, 1H), 6.78 (d, J = 8.3 Hz, 1H), 5.91 (d, J = 6.1 Hz, 1H), 4.84 (t, J = 5.8 Hz, 1H), 4.36-4.46 (m, 1H), 4.30 (q, J = 4.4 Hz, 1H), 3.35-3.50 (m, 2H); **^{13}C NMR** (151 MHz,

(6) Ferreras, J. A.; Ryu, J. S.; Di Lello, F.; Tan, D. S.; Quadri, L. E. *Nat. Chem. Biol*. **2005**, *1*, 29–32

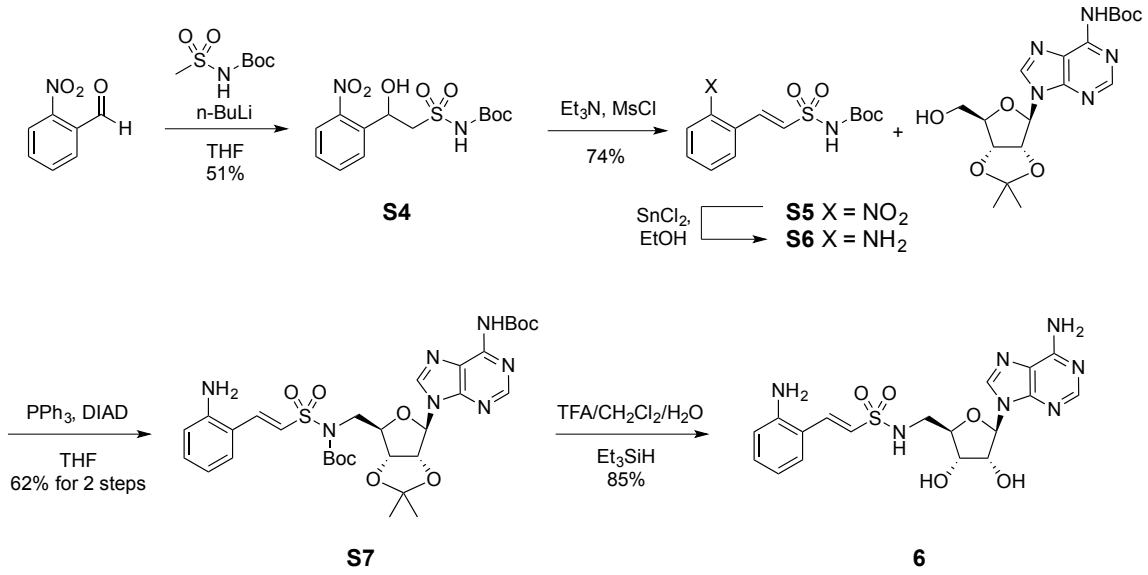
(7) Somu, R. V.; Boshoff, H.; Qiao, C.; Bennett, E. M.; Barry III, C. E.; Aldrich, C. C. *J. Med. Chem*. **2006**, *49*, 31–34.

D_2O): δ 173.98, 158.55, 155.41, 152.53, 148.46, 140.60, 133.79, 129.18, 119.18, 118.41, 116.47, 87.96, 83.39, 72.92, 71.15, 45.14. **HRMS (ESI)** *m/z*: calcd. for $C_{17}H_{20}N_7O_7S$ $[M+H]^+$ 466.1145, found 466.1130.

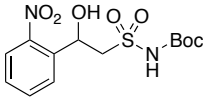


Benzoyl-AMS (5). This compound was prepared as previously described[8] and converted to sodium salt using the general procedure for ion exchange.

^{1}H NMR (600 MHz, CD_3OD): δ 8.56 (s, 1H), 8.16 (s, 1H), 8.04 (t, J = 1.2 Hz, 1H), 8.02 (d, J = 1.5 Hz, 1H), 7.41-7.45 (m, 1H), 7.32-7.37 (m, 2H), 6.10 (d, J = 5.9 Hz, 1H), 4.73 (dd, J = 5.9, 5.0 Hz, 1H), 4.44 (dd, J = 5.0, 3.2 Hz, 1H), 4.41 (dd, J = 11.2, 3.3 Hz, 1H), 4.37 (dd, J = 11.1, 3.1 Hz, 1H), 4.33 (q, J = 3.2 Hz, 1H); **^{13}C NMR** (151 MHz, CD_3OD): δ 175.35, 157.22, 153.82, 150.81, 141.16, 138.90, 132.08, 129.88, 128.79, 120.11, 89.16, 84.72, 76.14, 72.43, 69.25; **HRMS (ESI)** *m/z*: calcd. for $C_{17}H_{17}N_6O_7S$ $[M-H]^-$ 449.0879, found 449.0859.



Scheme S3. Synthesis of anthranilyl-AVSN (6).

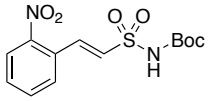


***tert*-Butyl ((2-hydroxy-2-(2-nitrophenyl)ethyl)sulfonyl)carbamate (S4).** In a 100-mL roundbottom flask, *tert*-butyl (methylsulfonyl)carbamate **1** (600 mg, 3.07 mmol, 1.0 equiv) was dissolved in 15 mL of THF and the solution was cooled to –78 °C. *n*-BuLi (3.84 mL, 6.14

(8) Qiao, C.; Gupte, A.; Boshoff, H. I.; Wilson, D. J.; Bennett, E. M.; Somu, R. V.; Barry III, C. E.; Aldrich, C. C. *J. Med. Chem*. **2007**, *50*, 6080–6094.

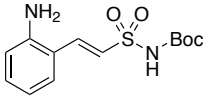
mmol, 2.0 equiv, 1.6 M in hexanes) was added dropwise. The resulting solution was allowed to warm to 0 ºC over 1 h, then cooled again to –78 ºC. A precooled (–78 °C) solution of 2-nitrobenzaldehyde (464 mg, 3.07 mmol, 1.0 equiv) in 6 mL of THF was added dropwise and the reaction mixture was allowed to warm to –40 ºC over 1 h. The reaction mixture was quenched with 1 N HCl and the aqueous layer was separated and extracted with EtOAc (2×100 mL). The combined organic extracts were washed with brine, dried (Na_2SO_4), filtered, and concentrated by rotary evaporation. Purification by silica flash chromatography (0–50% EtOAc in hexanes) afforded the benzylic alcohol **S4** (542 mg, 51%) as a light yellow oil.

TLC: R_f 0.4 (1:1 hexanes/EtOAc); **^{1}H NMR** (600 MHz): δ 8.04 (d, J = 7.8 Hz, 1H), 7.98 (d, J = 7.8 Hz, 1H), 7.73 (t, J = 7.4 Hz, 1H), 7.51 (t, J = 7.4 Hz, 1H), 5.85 (d, J = 9.5 Hz, 1H), 4.11 (dd, J = 9.6, 14.8 Hz, 1H), 3.54 (dd, J = 9.6, 14.8 Hz, 1H), 1.52 (s, 9H); **^{13}C-NMR** (151 MHz): δ 149.6, 146.8, 138.6, 136.0, 134.4, 129.6, 128.4, 124.5, 85.0, 65.0, 59.8, 41.2, 28.2; **ESI-MS** *m/z* calcd. for $C_{13}H_{18}N_2O_7SNa$ $[M+Na]^+$ 369.1, found 369.1.

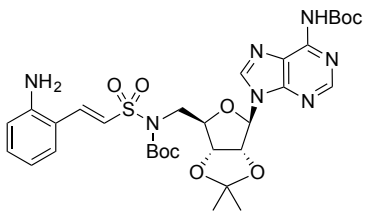


***tert*-Butyl (*E*)-((2-nitrostyryl)sulfonyl)carbamate (S5).** In a 50-mL roundbottom flask, benzyl alcohol **S4** (500 mg, 1.45 mmol, 1.0 equiv) was dissolved in 14.5 mL of CH_2Cl_2 and the solution was cooled to 0 °C. Triethylamine (0.6 mL, 4.35 mmol, 3.0 equiv) was added followed by methanesulfonyl chloride (0.168 mL, 2.18 mmol, 1.5 equiv). The resulting solution was stirred at 0 ºC for 2 h, then heated to reflux for 6 h. The mixture was quenched with saturated sodium bicarbonate solution and the aqueous layer was separated and extracted with CH_2Cl_2 (2×50 mL). The combined organic extracts were washed with brine, dried (Na_2SO_4), filtered, and concentrated by rotary evaporation. Purification by silica flash chromatography (0–40% EtOAc in hexanes) afforded 2-nitrophenylvinyl sulfonamide **S5** (351 mg, 74%) as a light yellow oil.

TLC: R_f 0.42 (2:1 hexanes/EtOAc); **^{1}H NMR** (600 MHz): δ 8.18 (d, J = 15.4 Hz, 1H), 8.14 (d, J = 8.2 Hz, 1H), 7.72 (t, J = 7.5 Hz, 1H), 7.65–7.62 (m, 2H), 7.36 (s, 1H), 7.05 (d, J = 15.4 Hz, 1H), 1.52 (s, 9H); **^{13}C-NMR** (151 MHz): δ 149.3, 148.0, 139.7, 134.0, 131.3, 129.6, 129.0, 128.5, 125.3, 84.7, 31.6, 28.0; **ESI-MS** *m/z* calcd. for $C_{13}H_{16}N_2O_6SNa$ $[M+Na]^+$ 351.1, found 351.2.

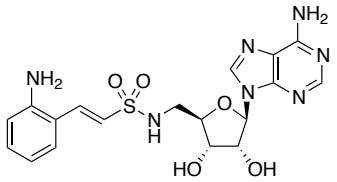


***tert*-Butyl (*E*)-((2-aminostyryl)sulfonyl)carbamate (S6).** In a 5-mL conical vial, 2-nitrophenylvinyl sulfonamide **S5** (300 mg, 0.92 mmol, 1.0 equiv) and $SnCl_2 \cdot 2H_2O$ (1.04 g, 4.6 mmol, 5.0 equiv) were dissolved in 2 mL of EtOH and heated at 40 °C for 1 h. The solution was allowed to cool to room temperature, poured onto ice (10 g), and basified with 5% aqueous sodium bicarbonate. The resulting mixture was extracted with EtOAc (3×50 mL), dried (Na_2SO_4), and concentrated by rotary evaporation. The crude 2-aminophenylvinyl sulfonamide **S6** was used for the next step without purification.



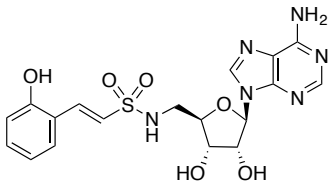
Protected anthranilyl-AVSN (S7). To solution of the crude vinyl sulfonamide **S6** (0.92 mmol, 1.0 equiv), N^6-Boc-2′,3′-*O*-isopropylideneadenosine, prepared as previously described[4] (1.2 g, 2.95 mmol (374 mg, 0.92 mmol, 1.0 equiv) and PPh_3 (362 mg, 1.38 mmol, 1.5 equiv) in 3.0 mL of THF at 0 °C was added a solution of DIAD (0.27 mL, 1.38 mmol, 1.5 equiv). The solution was gradually warmed to room temperature and stirred for 16 h. The mixture was then concentrated and purified by flash chromatography (20–100% EtOAc/hexanes) to afford the title compound (392 mg, 62%) as a foam.

TLC: R_f 0.2 (2:8 hexanes/EtOAc); **^{1}H NMR** (500 MHz): δ 8.77 (s, 1H), 8.05 (s, 1H), 7.91 (s, 1H), 7.52 (d, J = 15.0 Hz, 1H), 7.22–7.19 (m, 2H), 6.86 (d, J = 15.0 Hz, 1H), 6.76 (t, J = 7.5 Hz, 1H), 6.70 (d, J = 8.0 Hz, 1H), 6.15 (s, 1H), 5.51 (dd, J = 6.0, 1.5 Hz, 1H), 5.17 (dd, J = 6.5, 3.5 Hz, 1H), 4.52 (s, 1H), 4.01–3.97 (m, 4H), 1.60 (s, 3H), 1.56 (s, 9H), 1.47 (s, 9H), 1.37 (s, 3H); **^{13}C NMR** (151 MHz): δ 153.1, 150.9, 150.4, 150.0, 149.5, 146.0, 141.9, 139.5, 132.2, 128.5, 124.3, 122.3, 119.0, 117.6, 117.0, 114.6, 90.5, 85.7, 84.8, 84.3, 82.5, 82.3, 47.5, 28.1, 28.0, 27.9, 27.1, 25.3; **ESI-MS** *m/z* calcd. for $C_{31}H_{41}N_7O_9SNa$ $[M+Na]^+$ 710.3, found 710.3.



Anthranilyl-AVSN (6). In a 10-mL roundbottom flask, 2-aminophenylvinyl sulfonamide **S7** (70 mg, 0.1 mmol) was dissolved in CH_2Cl_2 (1 mL). 80% Aqueous TFA (1 mL) was added and the solution was stirred at 0 °C for 1 h, then at room temperature for 4 h. The resulting solution was azeotroped with toluene (2×10 mL) and concentrated by rotary evaporation. Purification by silica flash chromatography (10:1 EtOAc/MeOH) afforded anthranilyl-AVSN **6** (39 mg, 85%) as a waxy solid.

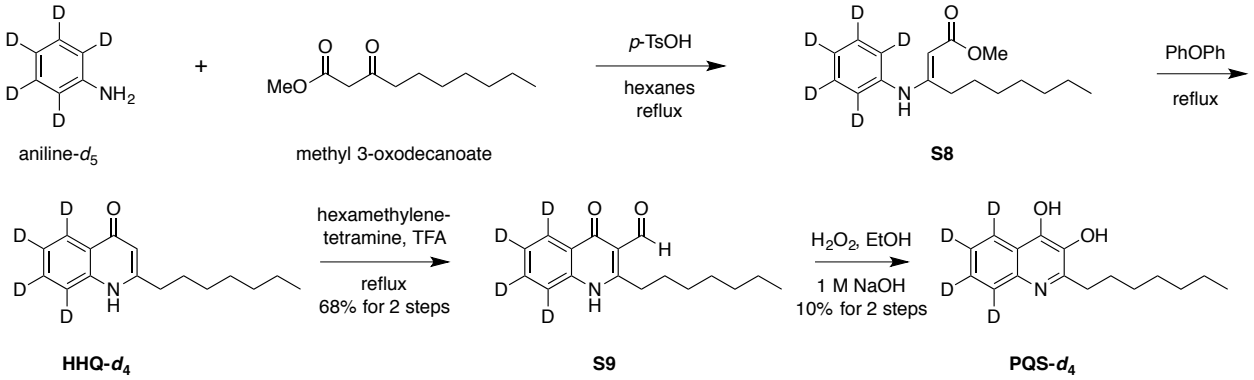
TLC: R_f 0.20 (9:1 EtOAc/MeOH); **^{1}H NMR** (500 MHz, CD_3OD): 7 exchangable proton δ 8.29 (s, 2H), 7.65 (d, J = 15.0 Hz, 1H), 7.34 (d, J = 8.0 Hz, 1H), 7.15 (t, J = 8.0 Hz, 1H), 6.84–6.78 (m, 2H), 6.68 (t, J = 7.5 Hz, 1H), 5.96 (d, J = 6.5 Hz, 1H), 4.39 (dd, J = 5.0, 3.0 Hz, 1H), 4.28 (d, J = 3.0 Hz, 1H), 3.43–3.37 (m, 3H). **^{13}C NMR** (151 MHz): δ 156.6, 152.3, 150.0, 148.4, 142.7, 138.3, 132.7, 128.8, 125.0, 121.1, 119.4, 119.0, 118.3, 91.3, 85.9, 74.6, 72.9, 45.6.; **ESI-MS** *m/z* calcd. for $C_{18}H_{21}N_7O_5SNa$ $[M+Na]^+$ 470.1, found 470.2.



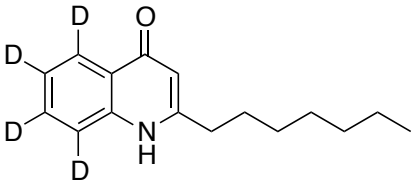
Salicyl-AVSN (7). This compound was prepared as previously described.[9]

(9) Sundlov, J. A.; Shi, C.; Wilson, D. J.; Aldrich, C. C.; Gulick, A. M. *Chem. Biol.* **2012**, *19*, 188–198

D. SYNTHESIS OF HHQ-d_4 AND PQS-d_4 STANDARDS FOR LC-MS/MS ANALYSIS



Scheme S4. Synthesis of HHQ-d_4 and PQS-d_4.



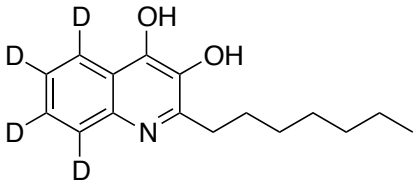
2-Heptylquinolin-4(1*H*)-one-5,6,7,8-d_4 (HHQ-d_4).[10] In a 100-mL roundbottom flask, aniline-d_5 (1.0 g, 10.2 mmol, 1.0 equiv) and methyl 3-oxodecanoate[11] (2.5 g, 12.50 mmol, 1.2 equiv) were dissolved in dry hexanes (35 mL). *p*-Toluenesulfonic acid (100 mg) was added and the mixture heated to reflux for 12 h under a Dean-Stark trap to remove water. The mixture was allowed to cool to room temperature then concentrated by rotary evaporation. The crude enamine **S8** was used without further purification.

In a 25-mL roundbottom flask equipped with a distillation head and collecting flask, diphenyl ether (8 mL) was heated to reflux (280 °C). The crude enamine **S8** was added dropwise over 10 min, then the mixture was stirred at reflux (280 °C) for an additional 30 min as the methanol byproduct was removed by distillation. The mixture was then cooled to room temperature, whereupon it solidified. Purification by silica flash chromatography (1:1 EtOAc/hexanes → EtOAc → 1:19 MeOH/EtOAc) afforded **HHQ-d_4** (1.4 g, 68% over 2 steps) as a white solid.

TLC: R_f 0.4 (19:1 EtOAc/CH_3OH). **^{1}H-NMR** (600 MHz, CD_3OD): 1 exchangable proton δ 6.22 (s, 1H), 2.71 (t, J = 7.8 Hz, 2H), 1.75 (q, J = 7.6 Hz, 2H), 1.45–1.25 (m, 8H), 0.89 (t, J = 6.8 Hz, 3H). **^{13}C-NMR** (151 MHz) δ 180.65, 157.17, 141.55, 125.41, 108.81, 35.02, 32.89, 30.22, 30.20, 30.19, 30.15, 30.13, 23.69, 14.41. **ESI-MS** for $C_{16}H_{17}D_4NO$ *m/z* (rel int): (pos) 270.2 ($[M+Na]^+$, 100).

(10) Lépine, F.; Déziel, E.; Milot, S.; Rahme, L. G. *Biochim. Biophys. Acta* **2003**, *1622*, 36–41.

(11) Reen, F. J.; Clarke, S. L.; Legendre, C.; McSweeney, C. M.; Eccles, K. S.; Lawrence, S. E.; O'Gara, F.; McGlacken, G. P. *Org. Biomol. Chem.* **2012**, *10*, 8903–8910.



2-Heptylquinoline-5,6,7,8-*d*$_4$-3,4-diol (PQS-d$_4$).[10] In a 50-mL roundbottom flask, HHQ-*d*$_4$ (1.14 g, 4.6 mmol, 1.0 equiv), hexamethylenetetramine (323 mg, 2.3 mmol, 0.5 equiv) were dissolved in trifluoroacetic acid (7.0 ml) and heated to reflux for 27 h. Methanol (11 ml) and water (11 ml) were added, and heating was continued for 50 min. Hydrochloric acid (2.5 M, 5 ml) was added, and heating was continued for 30 min. The mixture was allowed to cool to room temperature, then the precipitate was removed by filtration and washed with water to provide the crude aldehyde **S9** (1.0 g), which was used without further purification.

In a 25-mL roundbottom flask, aldehyde **S9** was dissolved in ethanol (3 mL). Aqueous 30% hydrogen peroxide (0.453 g, 3.99 mmol) was added followed by aqueous sodium hydroxide (1.08 M, 0.75 ml). The mixture was stirred at room temperature for 6 h, then diluted with water and filtered. The resulting solid was washed with water. Purification by silica flash chromatography (1:1 EtOAc/hexanes → EtOAc) afforded **PQS-*d*$_4$** (121 mg, 10% over 2 steps) as a white solid.

TLC: R_f 0.2 (EtOAc only). **^{1}H-NMR** (600 MHz, CD_3OD): 2 exchangable protons δ 2.91 (t, J = 7.8 Hz, 2H), 1.80 (p, J = 7.6 Hz, 2H), 1.51–1.30 (m, 8H), 0.92 (t, J = 6.7 Hz, 3H). **^{13}C-NMR** (151 MHz) δ 139.61, 139.60, 138.67, 123.56, 32.92, 30.44, 30.17, 29.82, 29.37, 23.70, 14.42. **ESI-MS** for $C_{16}H_{17}D_4NO_2$ *m/z* (rel int): (pos) 286.2 ($[M+Na]^+$, 100).

E. BIOCHEMICAL EVALUATION FOR PQSA INHIBITION

PqsA spectrophotometric assay

Reactions were performed at 37 °C in a 0.5 mL volume in a Varian Cary 100 UV-visible spectrophotometer with Cary WinUV software. Reaction mixtures contained 100 mM HEPES, pH 8.0, 0.2 mM dithiothreitol, 2 mM $MgCl_2$, ~3.3 μg PqsA protein (~60 nM final), and depending on which substrate varied, 1 mM ATP (disodium salt), 0.5 mM coenzyme A (tri-lithium salt), 0.5 mM sodium anthranilate, and inhibitor (usually as a DMSO stock solution). Reactions were blanked and preincubated with all components except anthranilate at 37 °C for 1 min, then initiated by addition of anthranilate. Formation of anthraniloyl-CoA was monitored by absorbance at 365 nm (ε = 5.5 $mM^{-1}cm^{-1}$) for 1 min. Data were analyzed using GraphPad Prism 6 software.

F. PQS/HHQ AND PYOCYANIN QUANTIFICATION IN *P. AERUGINOSA*

Bacterial culture and sample preparation

Production of PQS and HHQ by *P. aeruginosa* strain PA14 was performed in triplicate in 14-mL roundbottom Falcon tubes containing 3-mL cultures in LB medium. Tubes tilted at ~60º angle were incubated at 37 ºC in an orbital shaker at 225 rpm. Cultures were inoculated with 15-h culture in LB to obtain a starting OD_{600} = 0.05, with or without inhibitors added. For PQS and HHQ analysis, a 20 μL aliquot of the culture was taken at each time points as indicated. Each sample was diluted with 180 μL MeOH containing 100 μM HHQ-d_4 or PQS-d_4 internal standard and 2% AcOH (10-fold dilution of culture to 200 μL total volume with 90 μM internal standard final concentration). The solution was vortexed and centrifuged at 13,000*g* for 5 min and the supernatant was transferred to a 96-well plate for LC-MS/MS analysis. Bacterial growth was measured by recording OD_{600} at 20 h to provide the data in Figure S1.

PQS and HHQ quantification by LC-MS/MS analysis

Quantification of PQS and HHQ in bacterial culture supernatants was performed as previously described[12,13] with a slightly modified protocol. LC-MS/MS analysis was carried out on an Agilent Technologies 6410 triple quad LC-MS/MS system with autosampler in electrospray ionization (ESI) mode, with an Agilent Zorbax Eclipse XDB-C18 reverse phase column (50 × 4.6 mm, 5 μm) using a flow rate of 0.5 mL/min and an isocratic mobile phase of 60% CH_3CN in 0.1% aq formic acid over 5 min. Positive electrospray in MRM mode was employed to quantify PQS and HHQ, using the ion transitions indicated.

quinolone	ion transitions (H_4)	ion transitions (d_4)	linear range of detection (injected)	linear range of detection (culture)
HHQ	244 → 159	248 → 163	0.01 – 10 μM	0.1 – 100 μM
PQS	260 → 175	264 → 179	0.01 – 10 μM	0.1 – 100 μM

Pyocyanin quantification

Production of pyocyanin by *P. aeruginosa* strain PA14 was performed in triplicate in roundbottom glass tubes containing 5-mL cultures in LB medium. Tubes tilted at ~60º angle were incubated at 37 ºC in an orbital shaker at 200 rpm. Cultures were inoculated with 15-h preculture in LB in order to obtain a starting OD_{600nm} = 0.05, with or without inhibitors added. At 24 h, 1 mL of each culture was thoroughly vortexed and centrifuged (12,000 g, 5 min). Next, 0.8 mL of the supernatant was removed and the OD_{690nm} was measured. The ratio of the absorbance between samples from compound-treated or MvfR$^-$ cultures and untreated controls was calculated to provide the data in Figure S2.

(12) Lépine, F.; Déziel, E.; Milot, S.; Rahme, L. G. *Biochim. Biophys. Acta.* **2003**, *1622*, 36-41

(13) Lesic, B.; Lepine, F.; Deziel, E.; Zhang, J.; Zhang, Q.; Padfield, K.; Castonguay, M.-H.; Milot, S.; Stachel, S.; Tzika, A. A.; Tompkins, R. G.; Rahme, L. G. *PLoS Pathog.* **2007**, *3*, e126.

G. COMPOUND ACCUMULATION IN *P. AERUGINOSA*

Incubation, sample preparation, and LC-MS/MS analysis

P. aeruginosa PA14 (OD_{600} = 0.5, 1,000 uL, LB media, 37 °C, n = 8 samples) were incubated with the appropriate inhibitor [anthranilyl-AMS (**1**), anthranilyl-AMSN (**2**), or 6FABA] (1000 μM, 30 min), centrifuged (15,000 rpm, 4 °C, 5 min) and the supernatant removed. The pellet was washed with cold PBS (resuspend, centrifuge, decant 4 × 200 μL), and lysed by freeze-thaw cycles (200 μL PBS, 20 cycles). For each 200 μL sample (supernatant, 4 washes, lysate), 200 μL MeOH containing 2 μM benzoyl-AMS (**5**) internal standard was added (2-fold dilution of sample to 400 μL total volume with 1 μM internal standard final concentration). LC-MS/MS analysis was carried out as described in the preceding section, using an isocratic mobile phase of 30% CH_3CN in 0.1% aq formic acid over 5 min. Positive electrospray in MRM mode was employed to quantify the inhibitor, using the ion transitions indicated (benzoyl-AMS 451 → 136). The intracellular concentration of inhibitor was calculated based on CFU determination of the culture prior to centrifugation as described below.

inhibitor	ion transitions (inhibitor)	linear range of detection (injected)	avg # cells	avg intracellular volume	linear range of detection (intracellular)
1	466 → 136	0.006 – 100 μM	7.913×10^8	0.791 μL	3.036 – 50,600 μM
2	465 → 136	0.006 – 100 μM	1.203×10^9	1.203 μL	1.992 – 33,200 μM
6FABA	156 → 138	0.049 – 100 μM	2.122×10^9	2.122 μL	9.212 – 18,800 μM

Calculation of intracellular concentration

The total number of cells was determined via viable cell counts and plating of colony forming units (CFUs). After incubation with analyte, serial dilutions in fresh media were plated on agar. Colonies were grown for 24 h and plates containing 25–250 colonies were used to calculate the total number of cells. Volume of a bacterial cell was estimated as 1×10^{-15} L and total cell volume and intracellular analyte concentration were calculated based on this value. After intracellular concentration was calculated for each individual sample, outliers were removed by applying Grubbs' test to the dataset (n = 8) repeatedly until no more outliers were detected.

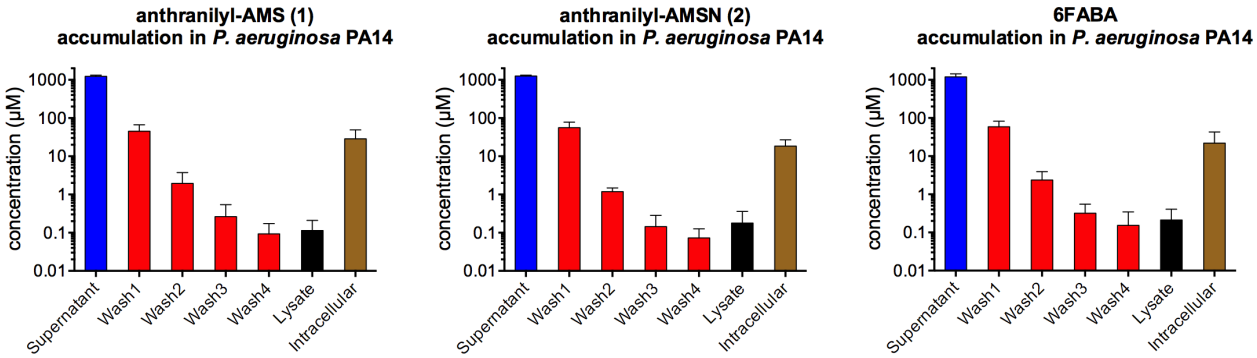
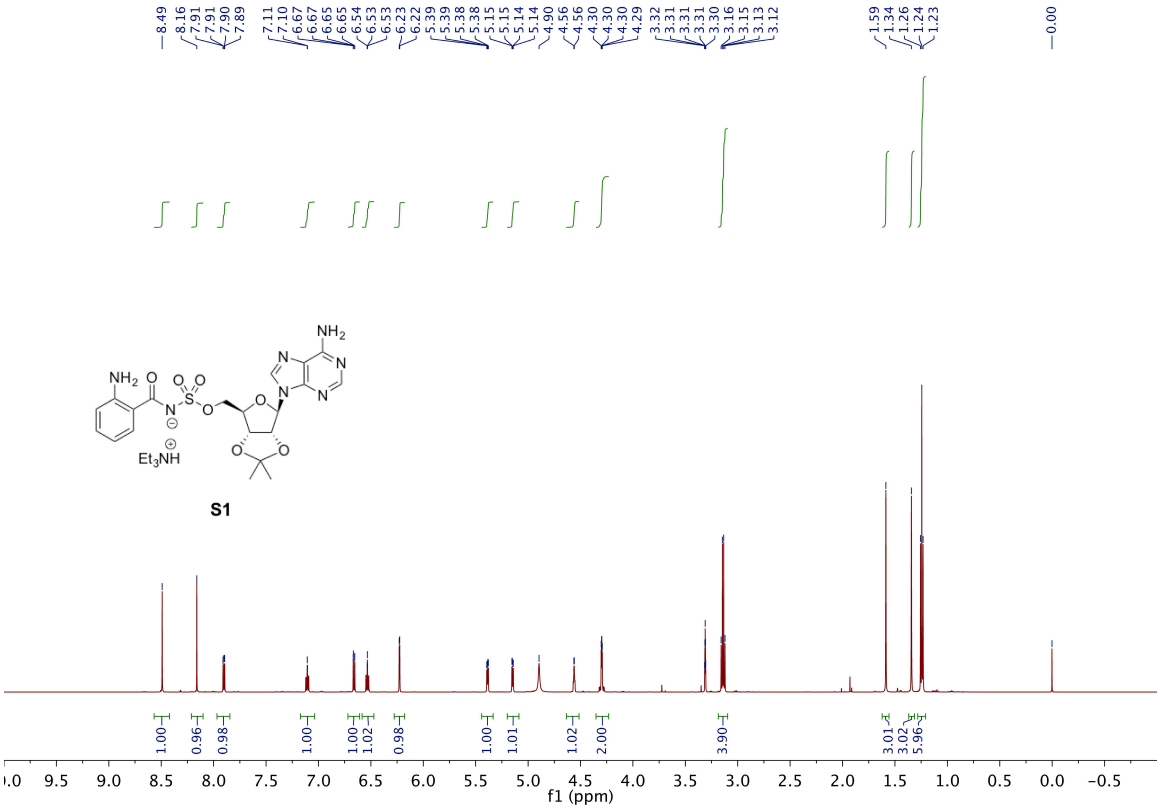


Figure S3. Compound concentrations detected in supernatant, washes, and lysates from incubations with *P. aeruginosa* strain PA14, with estimated intracellular concentrations calculated based on CFUs.

H. ^{1}H-NMR AND ^{13}C-NMR SPECTRA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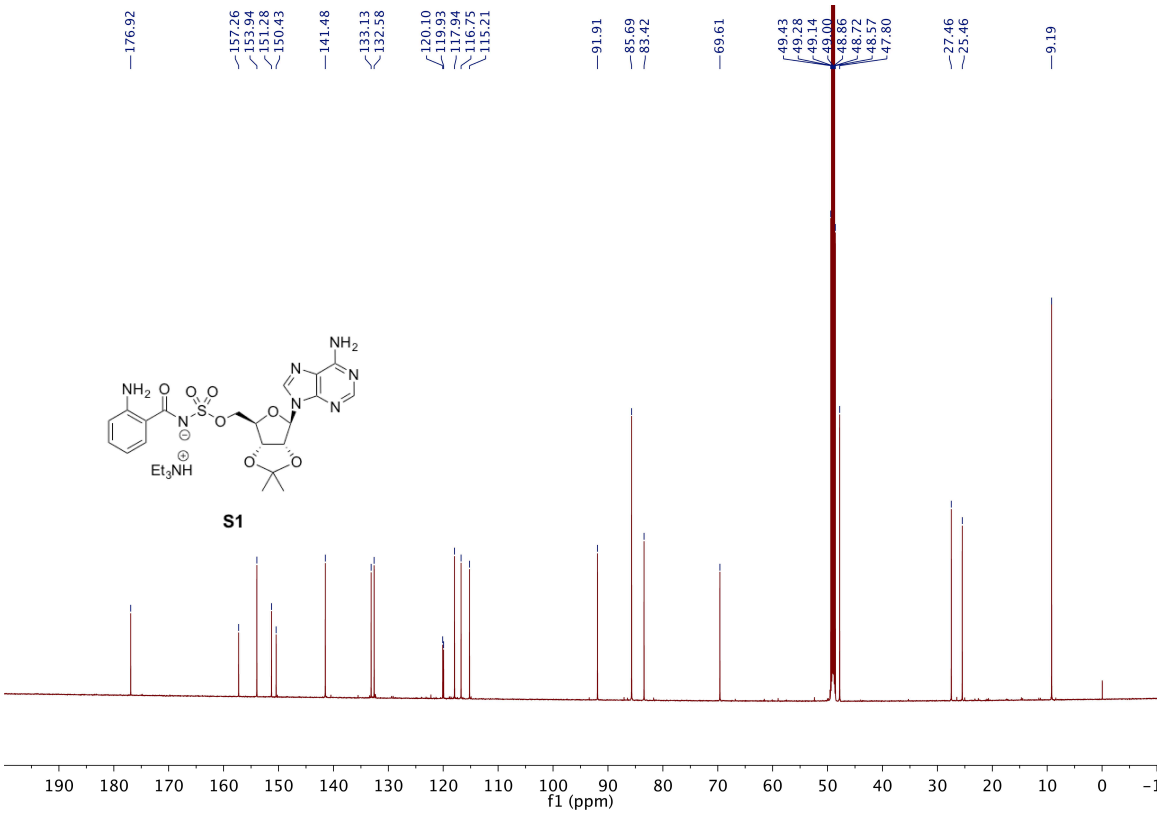
8.16
7.91
7.91
7.90
7.89
7.11
7.10
6.67
6.67
6.65
6.65
6.54
6.53
6.53
6.23
6.22
5.39
5.39
5.38
5.38
5.15
5.15
5.14
5.14
4.90
4.56
4.56
4.30
4.30
4.30
4.29
3.32
3.31
3.31
3.31
3.30
3.16
3.15
3.13
3.12
1.59
1.34
1.26
1.24
1.23
0.00
NH2
NH2
Et3NH
S1
1.00
0.96
0.98
1.00
1.00
1.02
0.98
1.00
1.01
1.02
2.00
3.90
3.01
3.02
5.96
f1 (ppm)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157.26
153.94
151.28
150.43
141.48
133.13
132.58
120.10
119.93
117.94
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91.91
85.69
83.42
69.61
49.43
49.28
49.14
49.00
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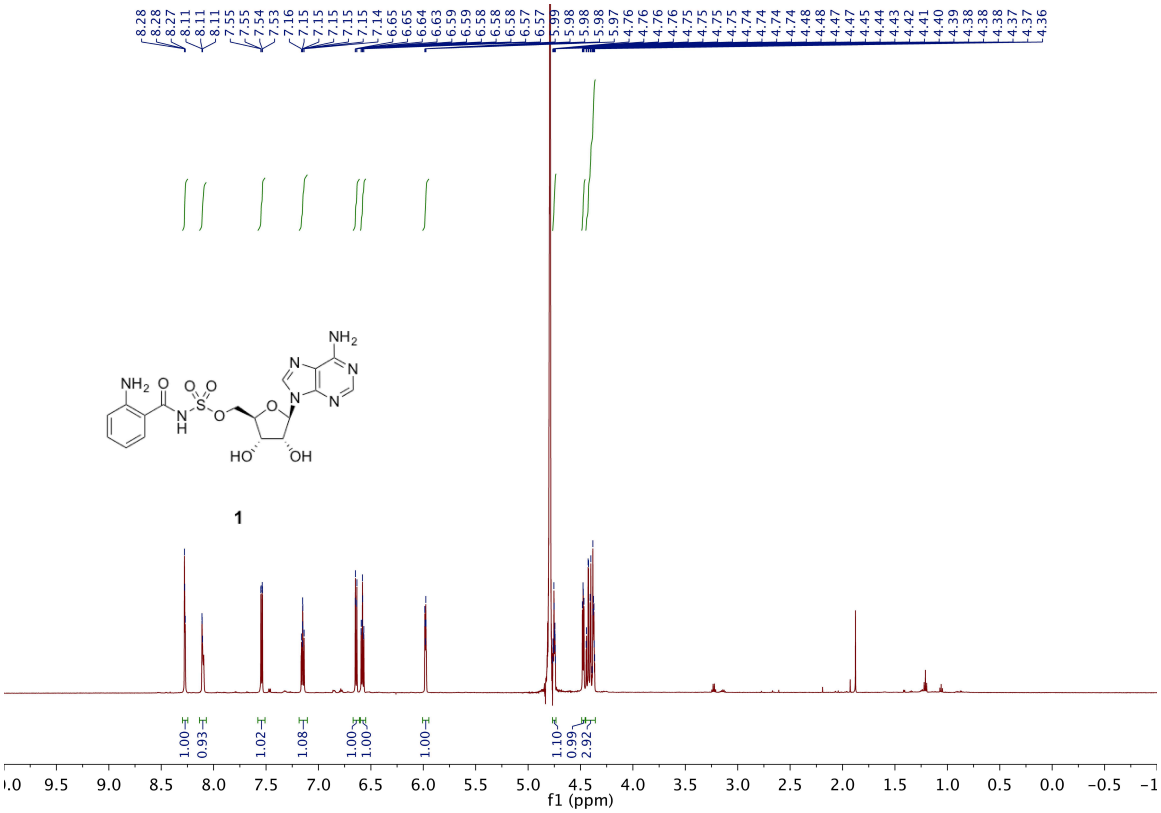
9.19
NH2
NH2
Et3NH
S1
f1 (ppm)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6.65
6.64
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6.58
6.57
5.99
5.98
5.97
4.76
4.75
4.74
4.48
4.47
4.45
4.44
4.43
4.42
4.41
4.40
4.39
4.38
4.37
4.36
NH2
O
S
N
H
HO
OH
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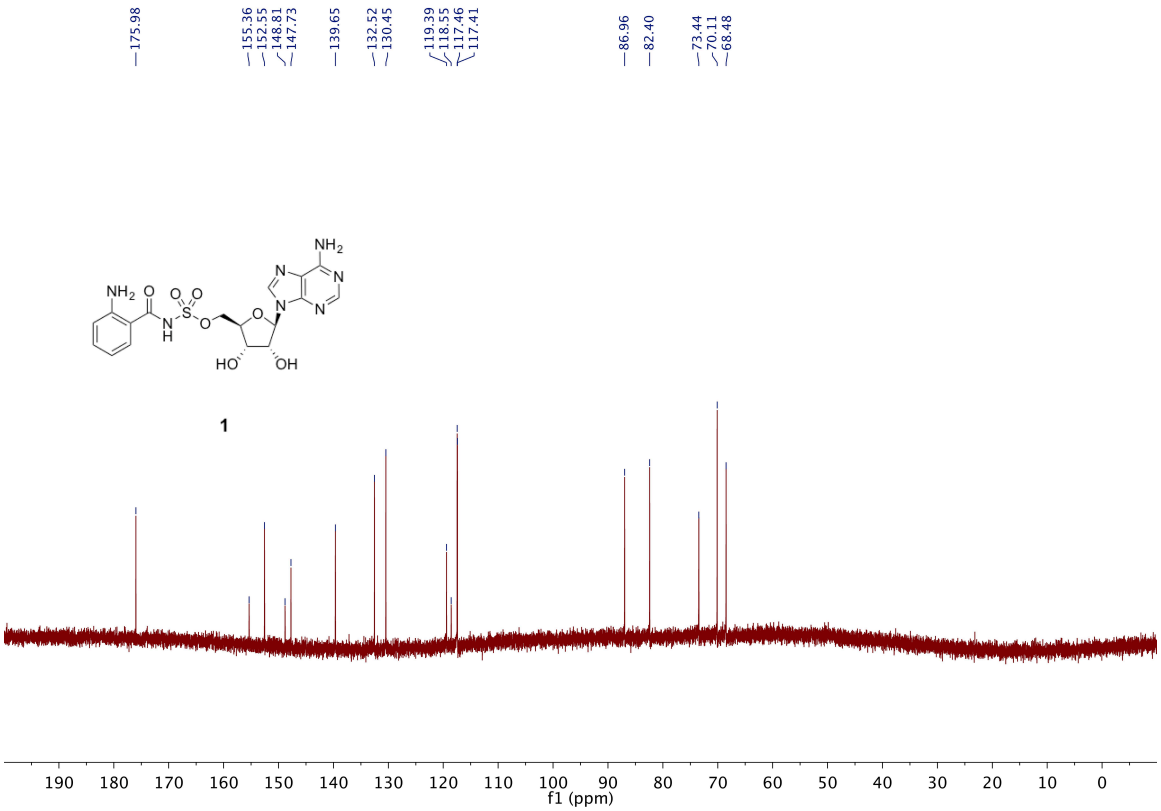
1.00
0.93
1.02
1.08
1.00
1.00
1.00
1.10
0.99
2.92
f1 (ppm)


175.98
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139.65
132.52
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118.55
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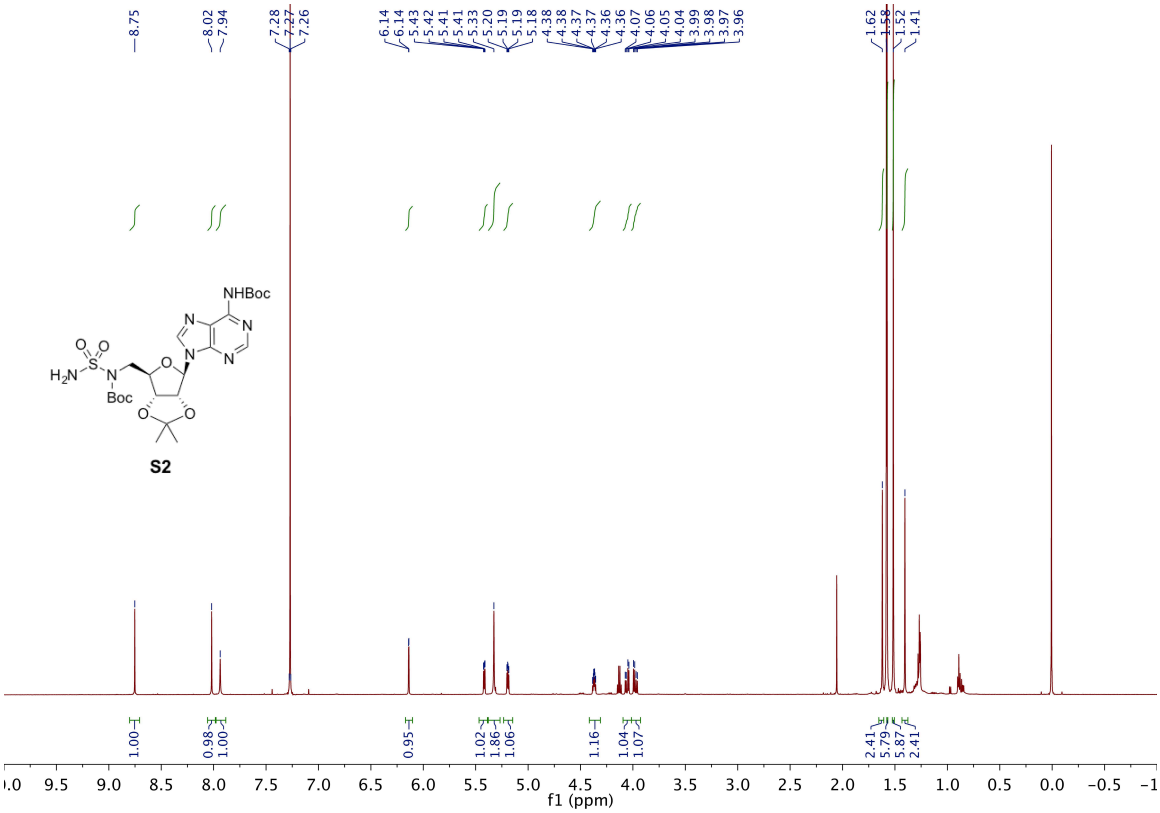
NH2
O
S
N
H
HO
OH
1
f1 (ppm)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6.14
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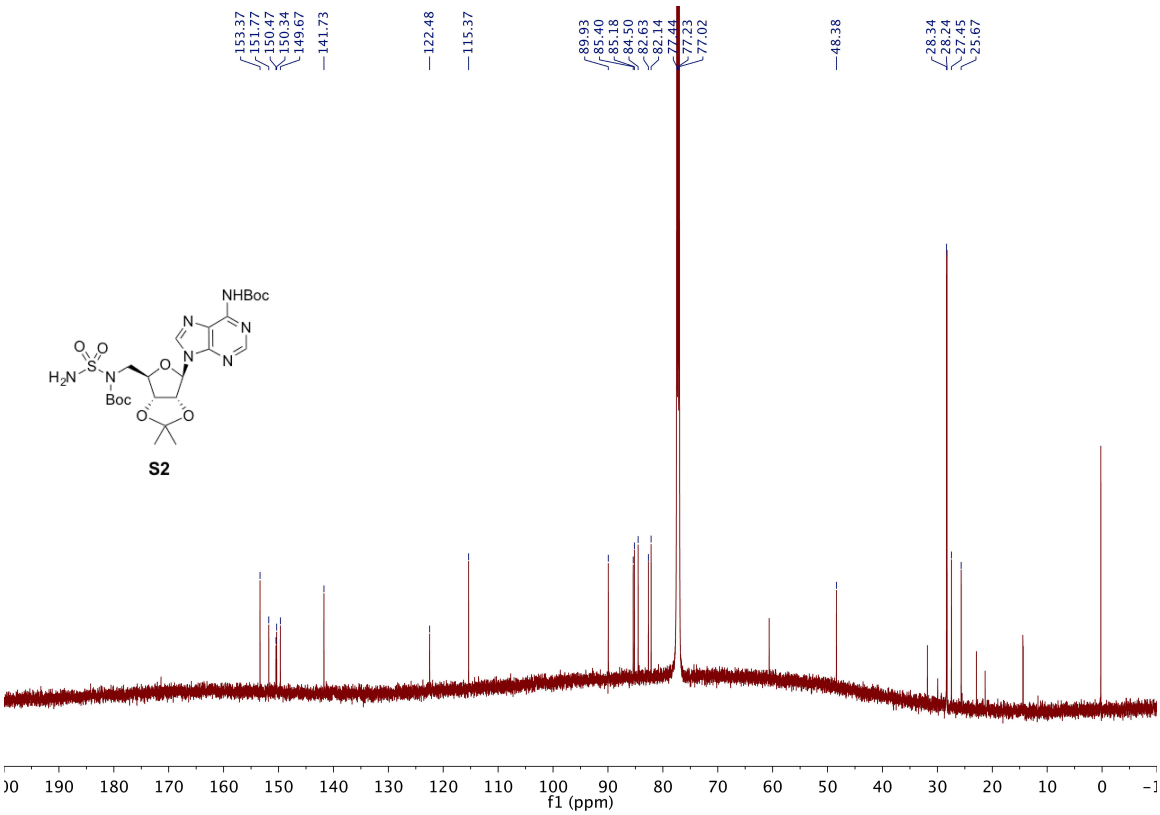
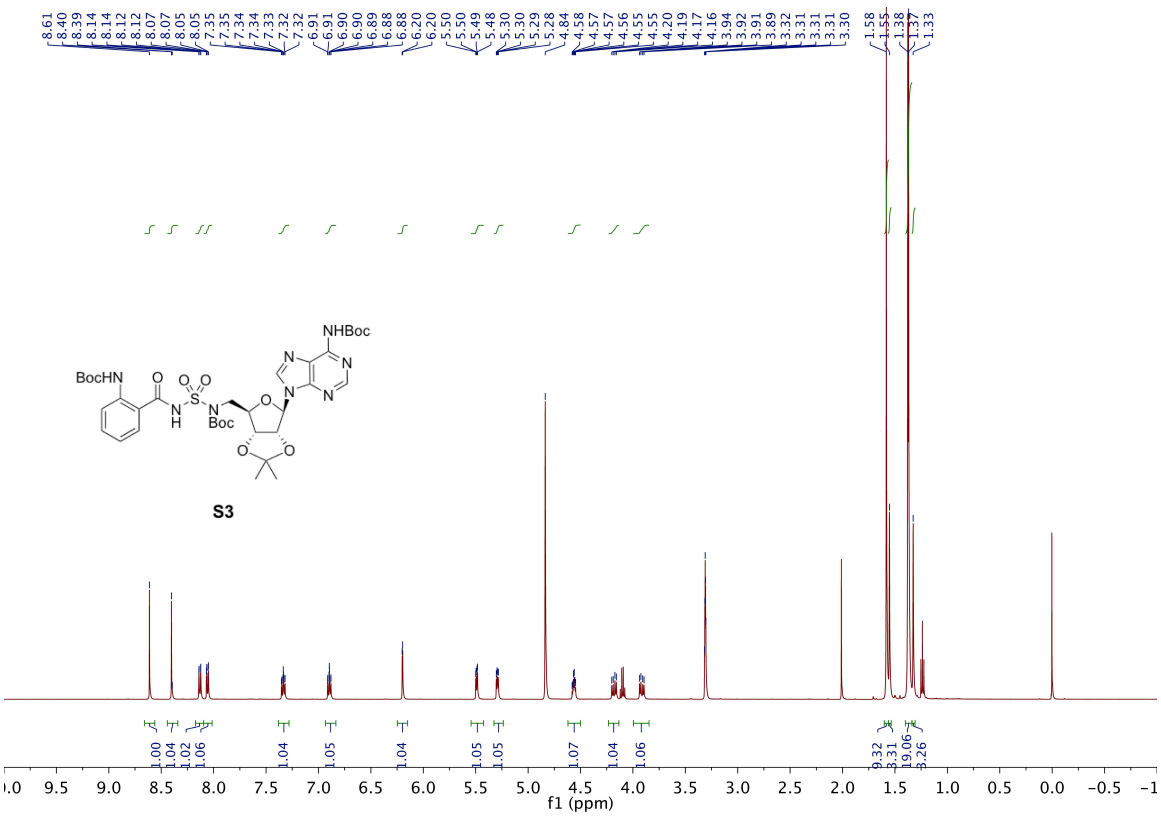
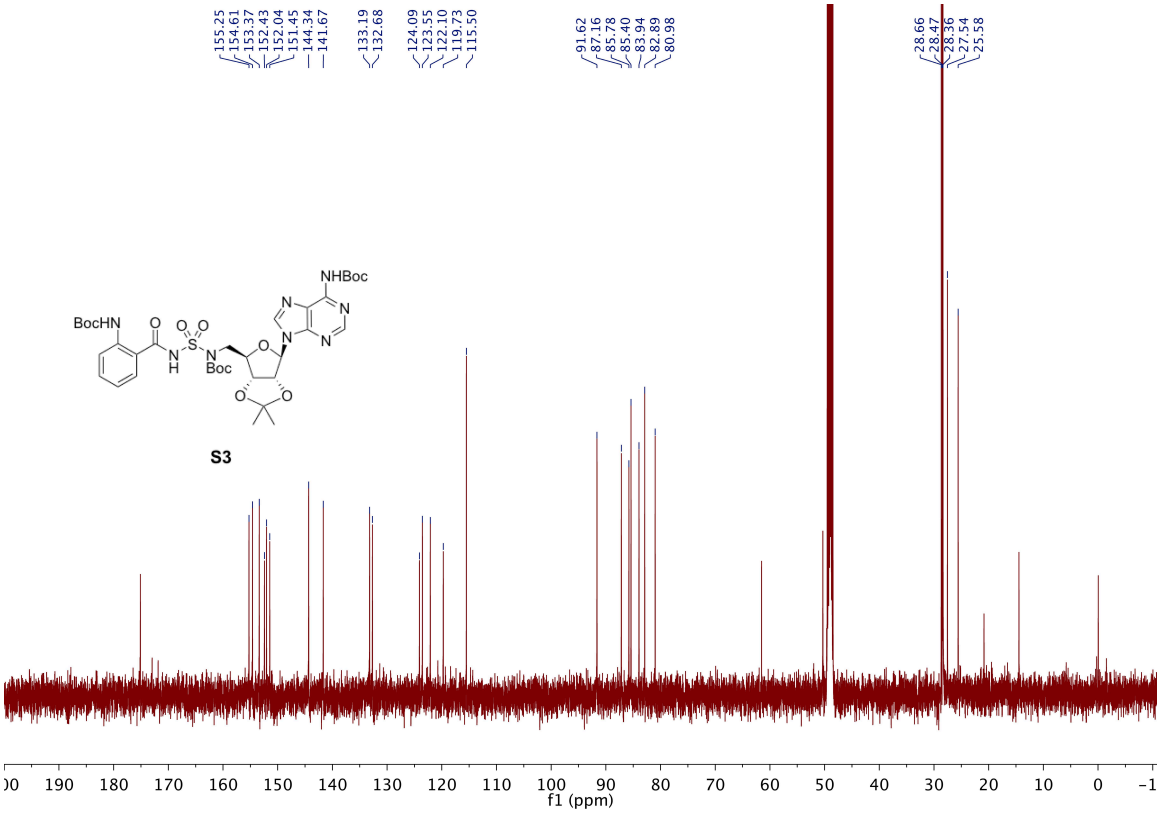
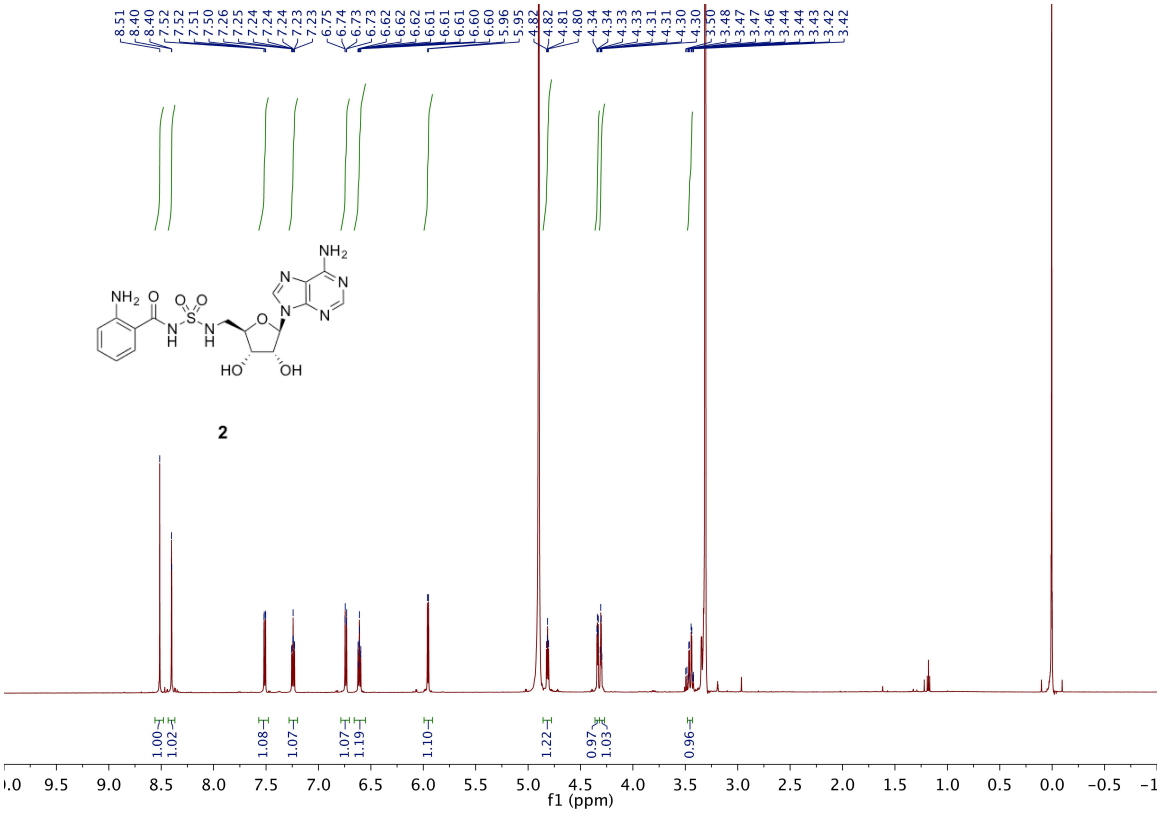
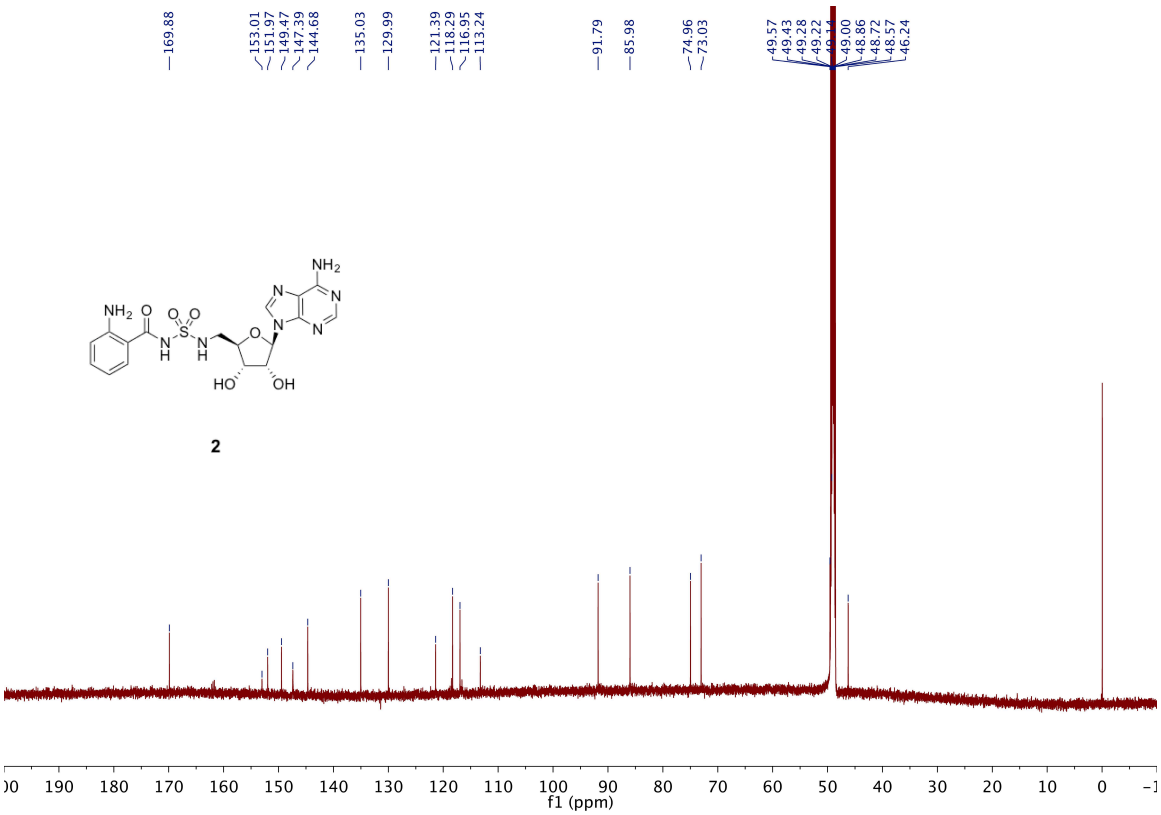
5.42
5.41
5.41
5.33
5.20
5.19
5.19
5.18
4.38
4.38
4.37
4.37
4.36
4.36
4.07
4.06
4.05
4.04
3.99
3.98
3.97
3.96
1.62
1.58
1.52
1.41
NHBoc
Boc
S2
1.00
0.98
1.00
0.95
1.02
1.86
1.06
1.16
1.04
1.07
2.41
5.79
5.87
2.41
f1 (ppm)

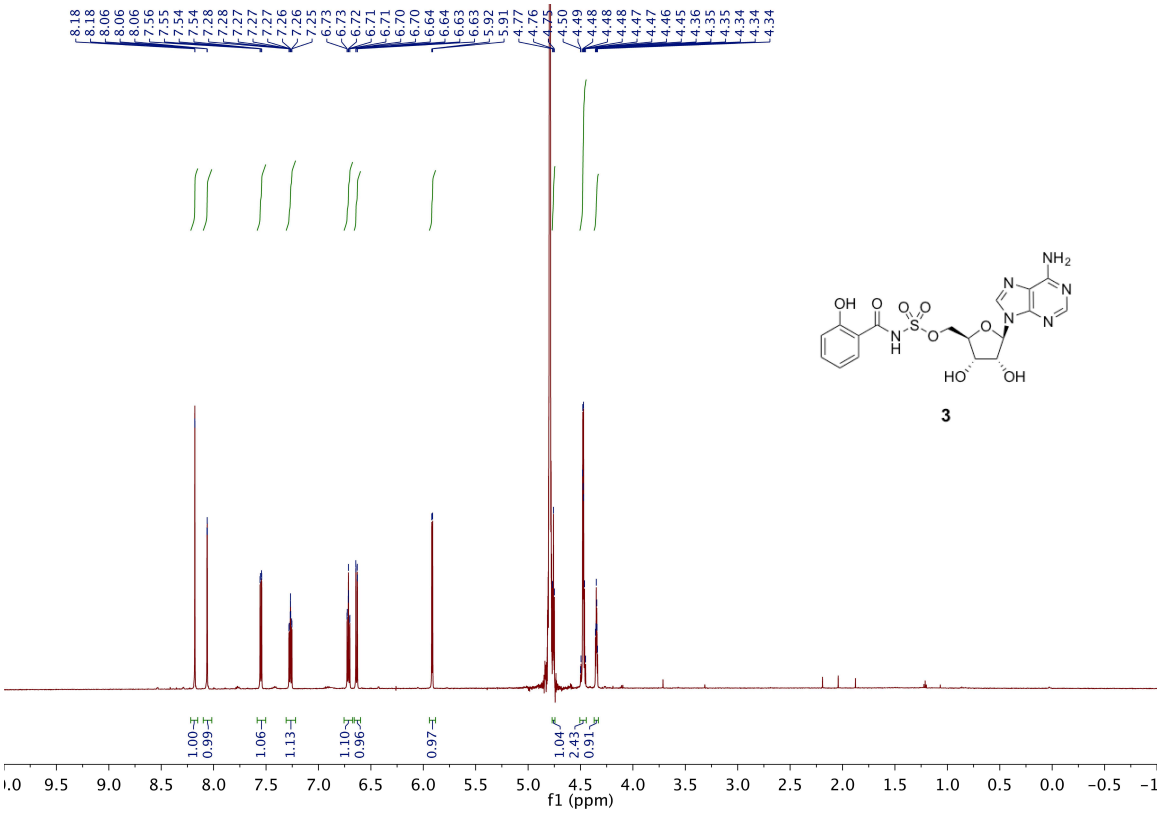

153.37
151.77
150.47
150.34
149.67
141.73
122.48
115.37
89.93
85.40
85.18
84.50
82.63
82.14
77.44
77.23
77.02
48.38
28.34
28.24
27.45
25.67
NHBoc
Boc
S2
f1 (ppm)

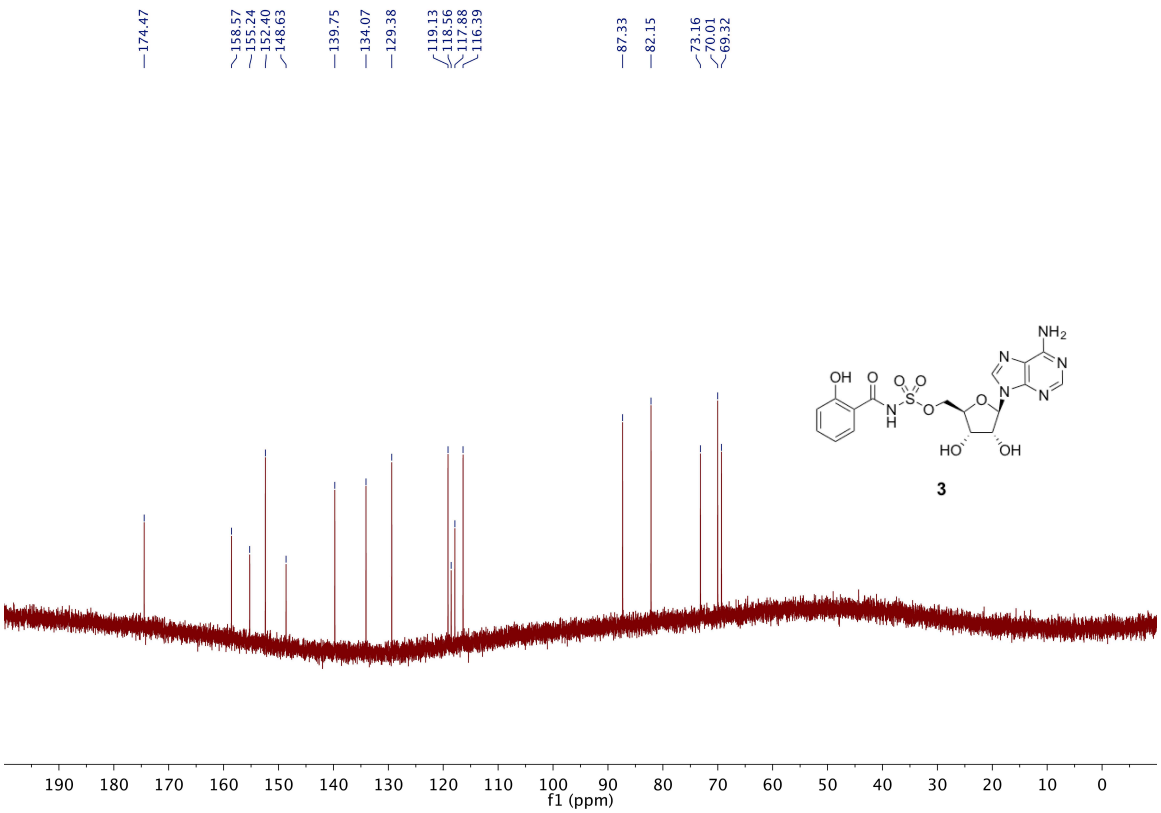

NHBoc
BocHN
Boc
S3
f1 (ppm)

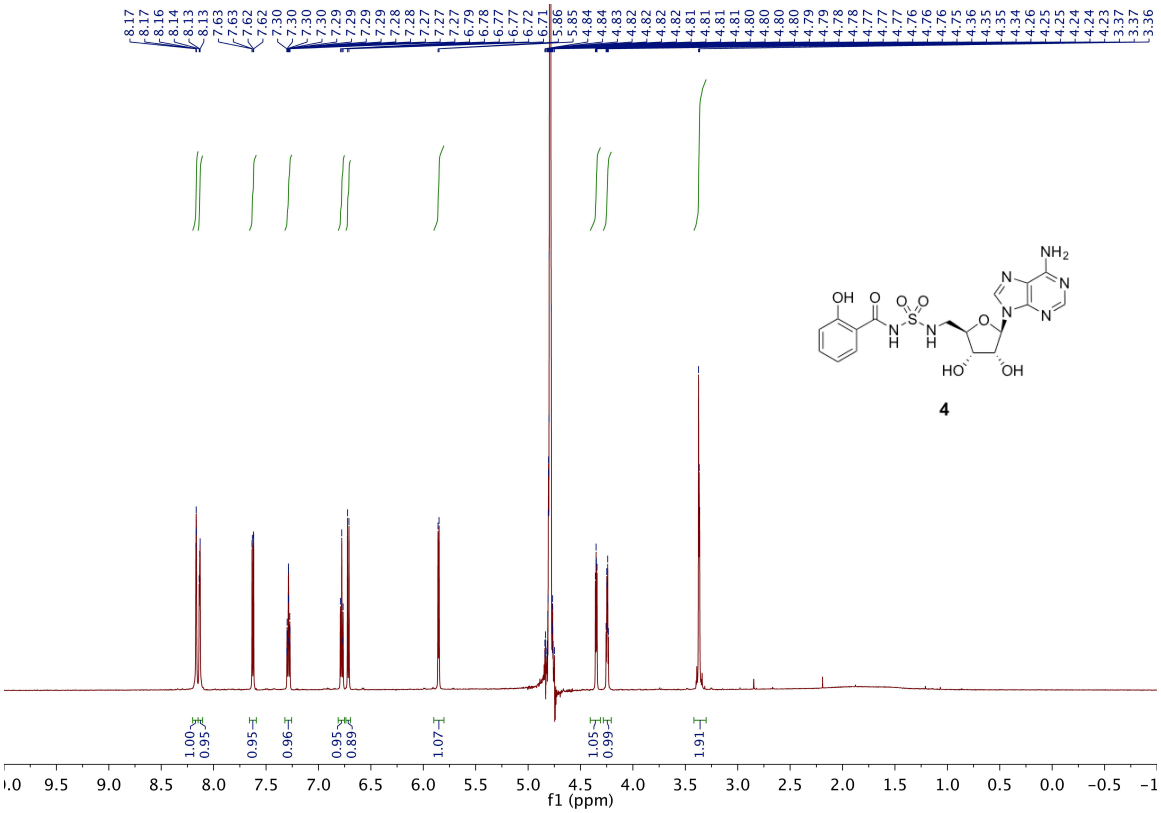

NHBoc
BocHN
Boc
S3
f1 (ppm)

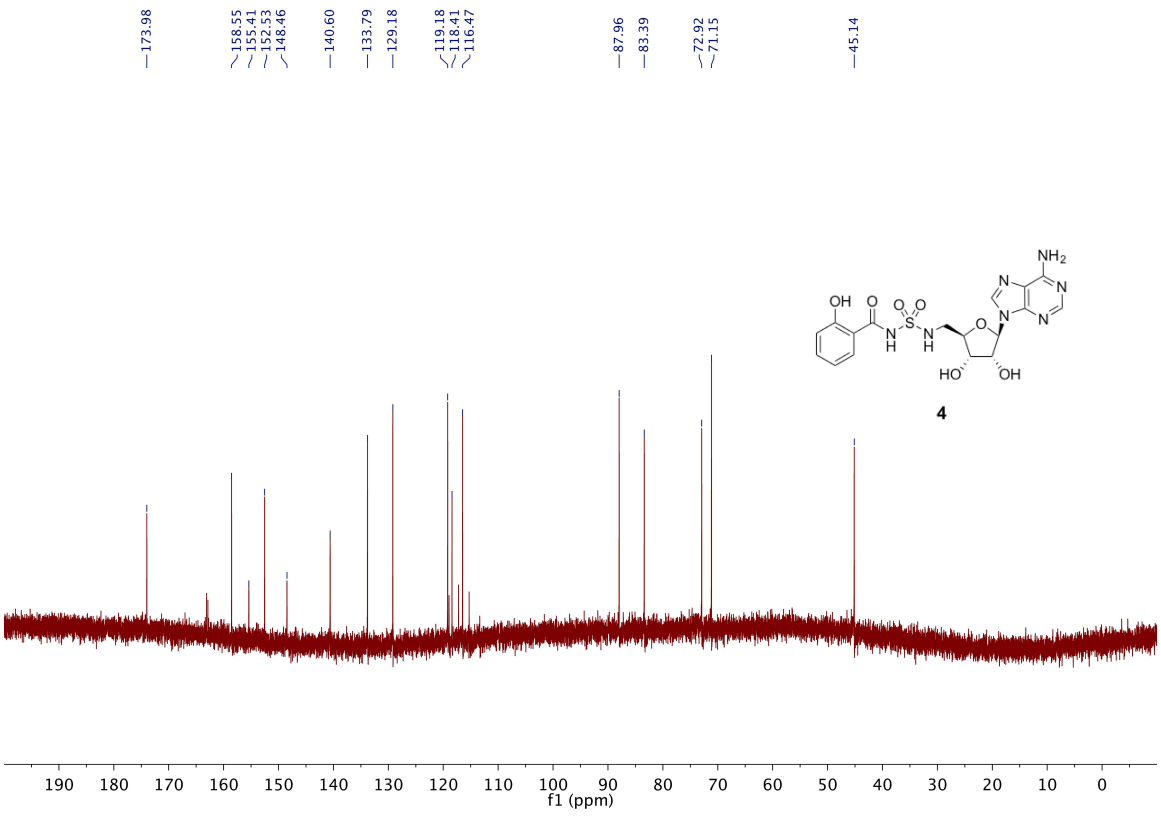

8.51
8.40
8.40
7.52
7.52
7.51
7.50
7.26
7.25
7.24
7.24
7.24
7.23
7.23
6.75
6.74
6.73
6.73
6.62
6.62
6.62
6.61
6.61
6.61
6.60
6.60
5.96
5.95
4.82
4.82
4.81
4.80
4.34
4.34
4.33
4.33
4.31
4.31
4.30
4.30
3.50
3.48
3.47
3.47
3.46
3.44
3.43
3.42
3.42
NH2
NH2
O
O
O
S
N
H
N
H
N
N
N
N
O
HO
OH
2
1.00
1.02
1.08
1.07
1.07
1.19
1.10
1.22
0.97
1.03
0.96
f1 (ppm)

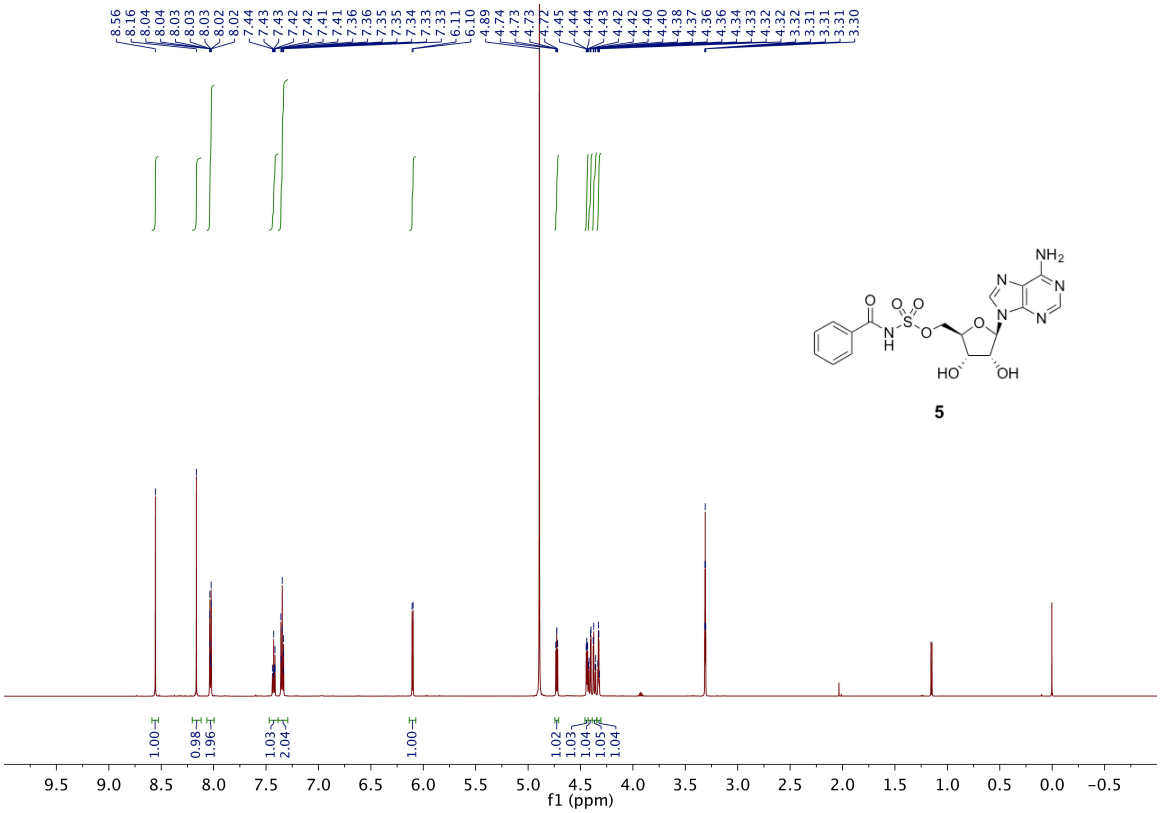

169.88
153.01
151.97
149.47
147.39
144.68
135.03
129.99
121.39
118.29
116.95
113.24
91.79
85.98
74.96
73.03
49.57
49.43
49.28
49.22
49.14
49.00
48.86
48.72
48.57
46.24
NH2
NH2
O
O
O
S
N
H
N
H
N
N
N
N
O
HO
OH
2
f1 (ppm)

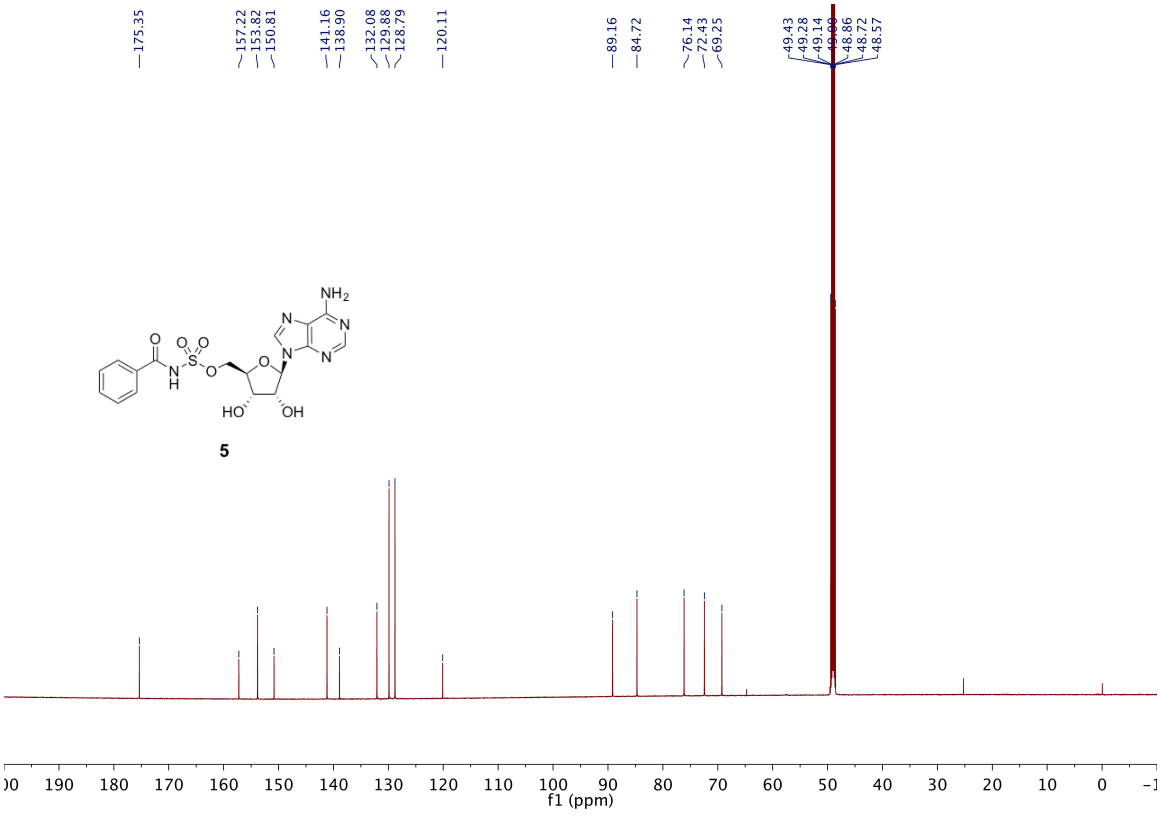

8.18
8.18
8.06
8.06
8.06
8.06
7.56
7.55
7.54
7.54
7.28
7.28
7.27
7.27
7.27
7.27
7.26
7.26
7.25
6.73
6.73
6.72
6.71
6.71
6.70
6.70
6.64
6.64
6.63
6.63
5.92
5.91
4.77
4.76
4.75
4.50
4.49
4.48
4.48
4.48
4.47
4.47
4.46
4.45
4.36
4.35
4.35
4.34
4.34
4.34
1.00
0.99
1.06
1.13
1.10
0.96
0.97
1.04
2.43
0.91
f1 (ppm)
3

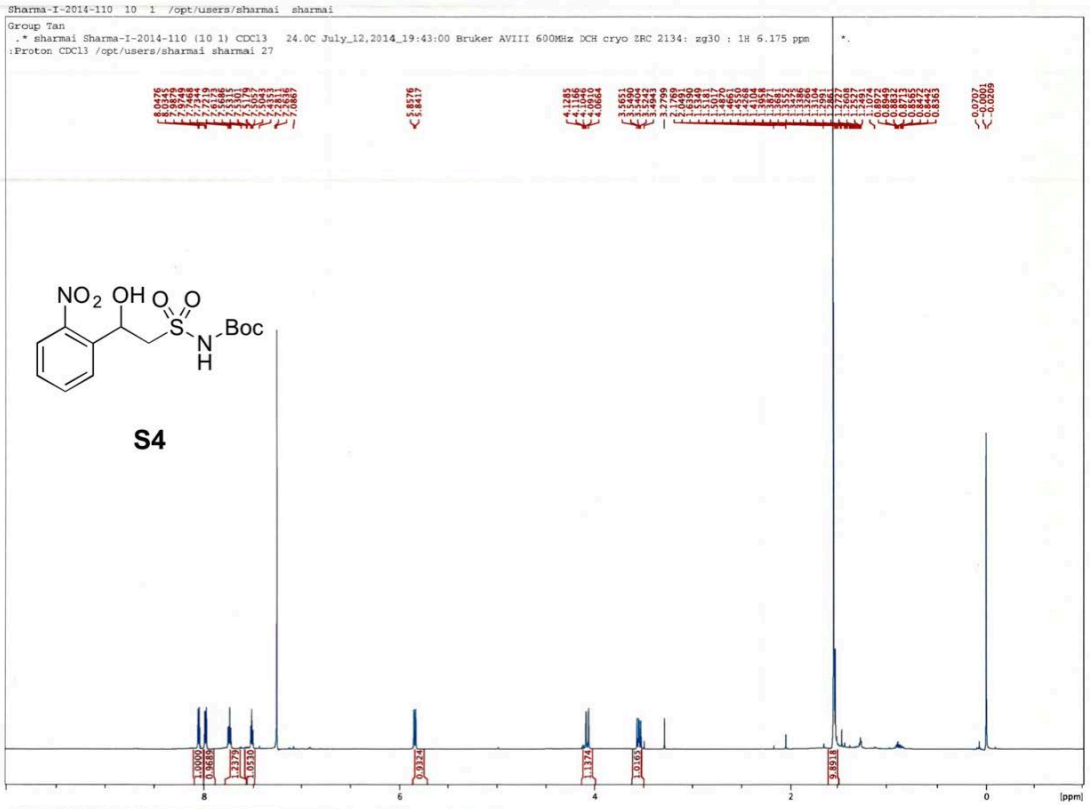

174.47
158.57
155.24
152.40
148.63
139.75
134.07
129.38
119.13
118.56
117.88
116.39
87.33
82.15
73.16
70.01
69.32
f1 (ppm)
3

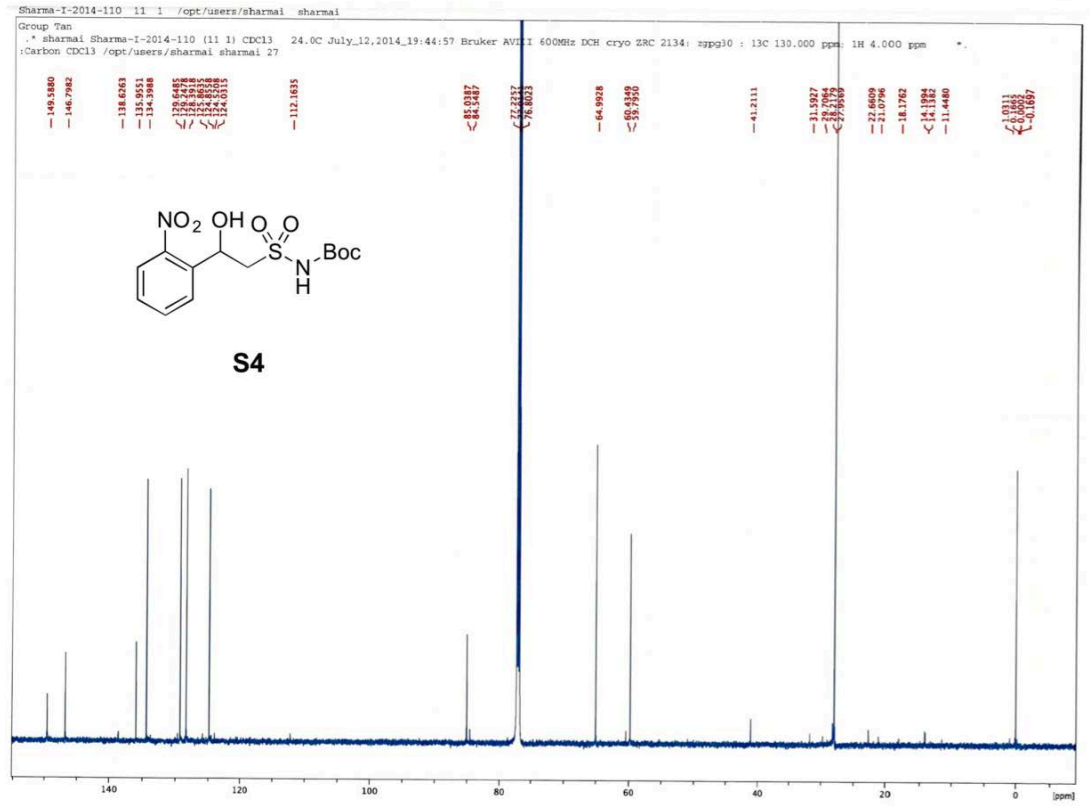

8.17
8.17
8.16
8.14
8.13
8.13
7.63
7.63
7.62
7.62
7.62
7.30
7.30
7.30
7.30
7.29
7.29
7.29
7.29
7.28
7.28
7.27
7.27
7.27
7.27
6.79
6.78
6.77
6.77
6.72
6.71
5.86
5.85
4.84
4.84
4.83
4.82
4.82
4.82
4.82
4.81
4.81
4.81
4.81
4.80
4.80
4.80
4.80
4.79
4.79
4.78
4.78
4.77
4.77
4.77
4.76
4.76
4.76
4.75
4.36
4.35
4.35
4.34
4.26
4.25
4.25
4.24
4.24
4.23
3.37
3.37
3.36
1.00
0.95
0.95
0.96
0.95
0.89
1.07
1.05
0.99
1.91
f1 (ppm)
4

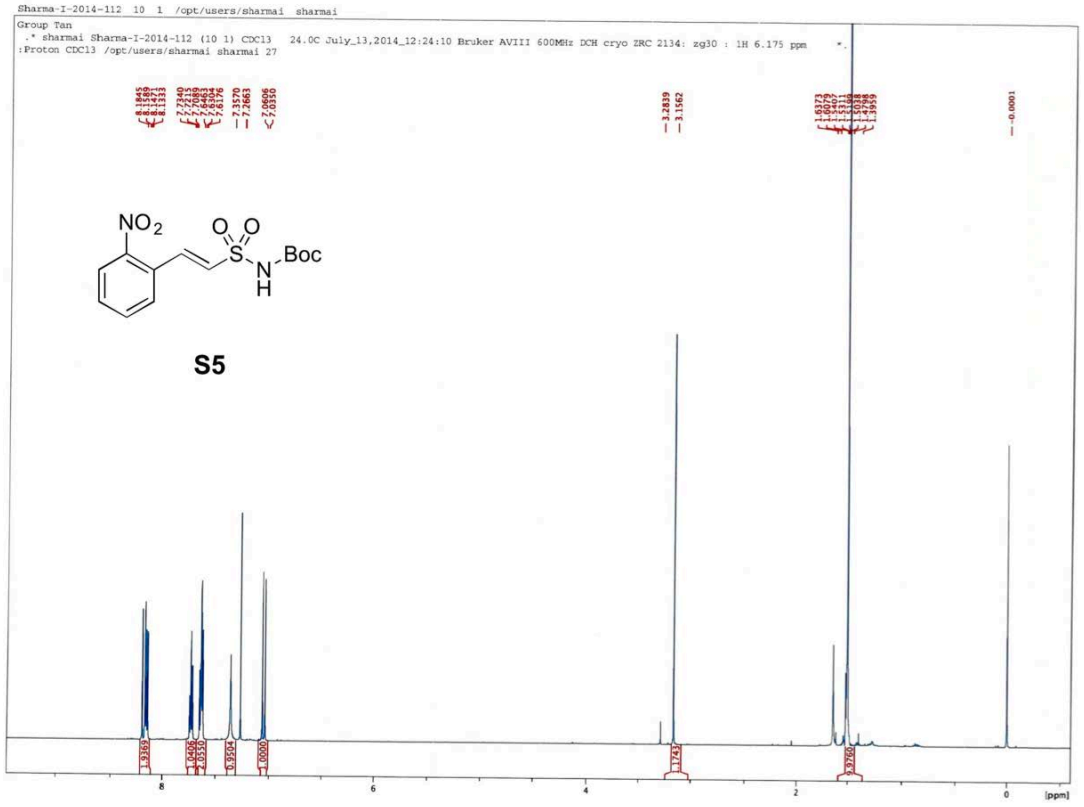

173.98
158.55
155.41
152.53
148.46
140.60
133.79
129.18
119.18
118.41
116.47
87.96
83.39
72.92
71.15
45.14
f1 (ppm)
4

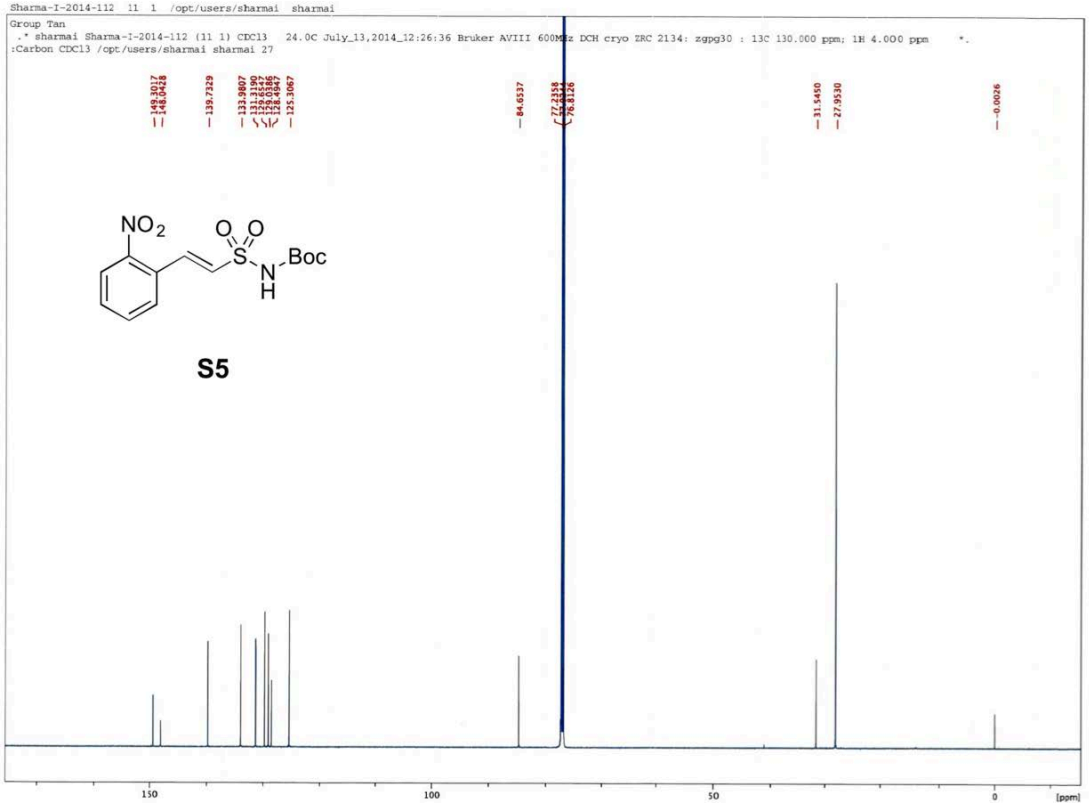

8.56
8.16
8.04
8.04
8.03
8.03
8.03
8.02
8.02
7.44
7.43
7.43
7.42
7.42
7.41
7.41
7.36
7.36
7.35
7.35
7.34
7.33
7.33
6.11
6.10
4.89
4.74
4.73
4.73
4.72
4.45
4.44
4.44
4.43
4.42
4.42
4.40
4.40
4.38
4.37
4.36
4.36
4.34
4.33
4.32
4.32
3.32
3.31
3.31
3.31
3.30
NH2
N
N
N
N
O
O
O
S
O
N
H
O
HO
OH
5
1.00
0.98
1.96
1.03
2.04
1.00
1.02
1.03
1.04
1.05
1.04
9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 −0.5
f1 (ppm)

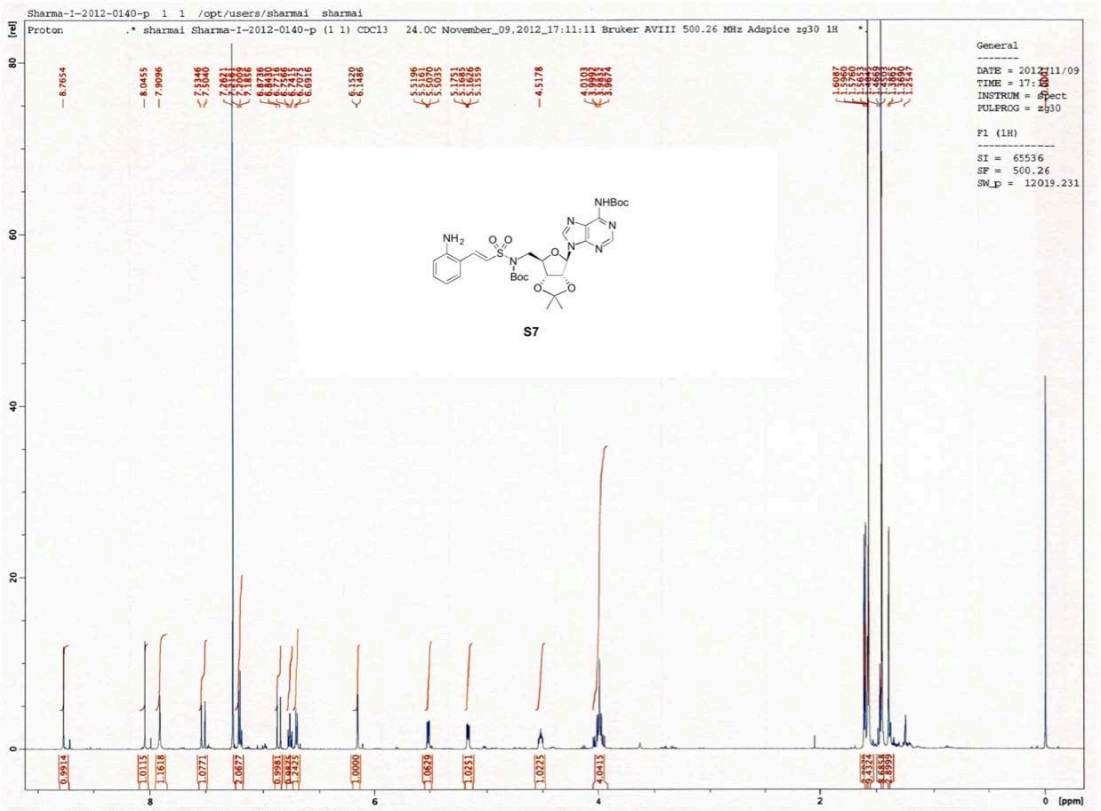

175.35
157.22
153.82
150.81
141.16
138.90
132.08
129.88
128.79
120.11
89.16
84.72
76.14
72.43
69.25
49.43
49.28
49.14
49.00
48.86
48.72
48.57
NH2
N
N
N
N
O
O
O
S
O
N
H
O
HO
OH
5
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
f1 (ppm)

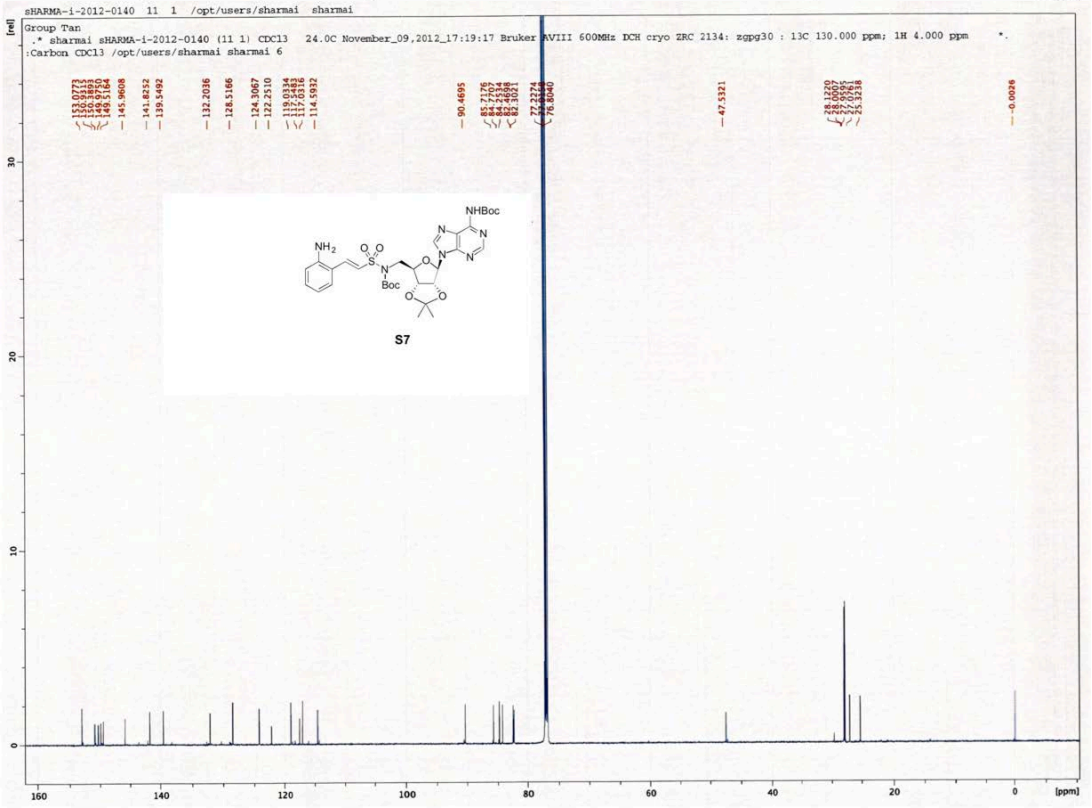

Sharma-I-2014-110 10 1 /opt/users/sharmai sharmai
Group Tan
.* sharmai Sharma-I-2014-110 (10 1) CDCl3 24.0C July_12,2014_19:43:00 Bruker AVIII 600MHz DCH cryo ZRC 2134: zg30 : 1H 6.175 ppm *.
:Proton CDCl3 /opt/users/sharmai sharmai 27
NO2 OH O O S N H Boc
S4
[ppm]

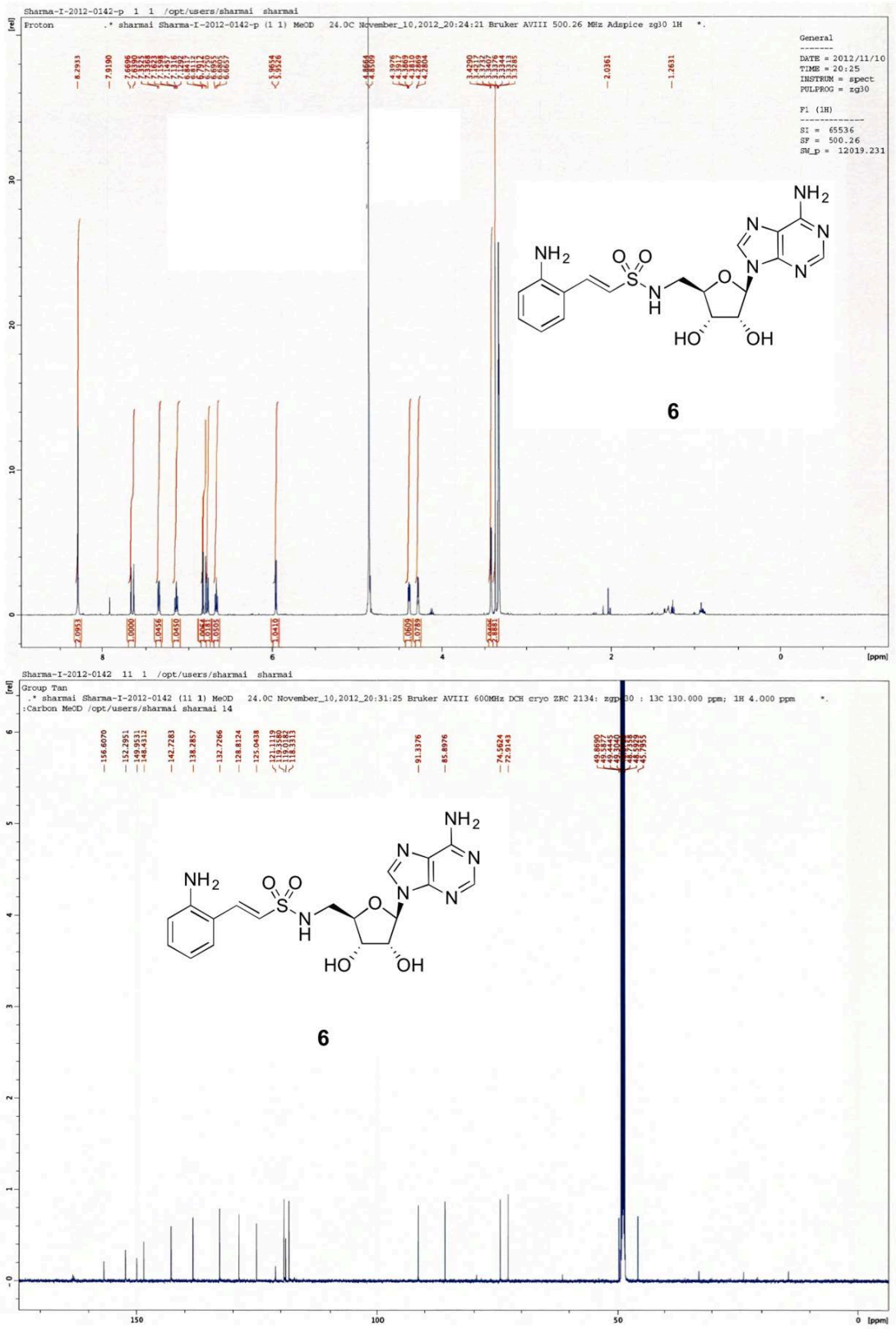

Sharma-I-2014-110 11 1 /opt/users/sharmai sharmai
Group Tan
.* sharmai Sharma-I-2014-110 (11 1) CDCl3 24.0C July_12,2014_19:44:57 Bruker AVIII 600MHz DCH cryo ZRC 2134: zgpg30 : 13C 130.000 ppm; 1H 4.000 ppm *.
:Carbon CDCl3 /opt/users/sharmai sharmai 27
NO2 OH O O S N H Boc
S4
[ppm]

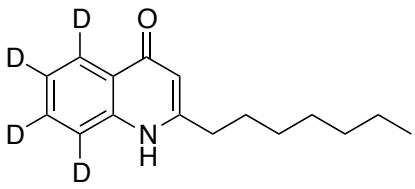

Sharma-I-2014-112 10 1 /opt/users/sharmai sharmai
Group Tan
.* sharmai Sharma-I-2014-112 (10 1) CDCl3 24.0C July_13,2014_12:24:10 Bruker AVIII 600MHz DCH cryo ZRC 2134: zg30 : 1H 6.175 ppm *.
:Proton CDCl3 /opt/users/sharmai sharmai 27
NO2
O
O
S
N
H
Boc
S5
ppm


Sharma-I-2014-112 11 1 /opt/users/sharmai sharmai
Group Tan
.* sharmai Sharma-I-2014-112 (11 1) CDCl3 24.0C July_13,2014_12:26:36 Bruker AVIII 600MHz DCH cryo ZRC 2134: zgpg30 : 13C 130.000 ppm; 1H 4.000 ppm *.
:Carbon CDCl3 /opt/users/sharmai sharmai 27
NO2
O
O
S
N
H
Boc
S5
ppm


Sharma-I-2012-0140-p 1 1 /opt/users/sharmai sharmai
Proton .* sharmai Sharma-I-2012-0140-p (1 1) CDCl3 24.0C November_09,2012_17:11:11 Bruker AVIII 500.26 MHz Adspice zg30 1H
General
DATE = 2012/11/09
INSTRUM = spect
PULPROG = zg30
F1 (1H)
SI = 65536
SF = 500.26
SW_p = 12019.231
NHBoc
NH2
Boc
S7
[ppm]

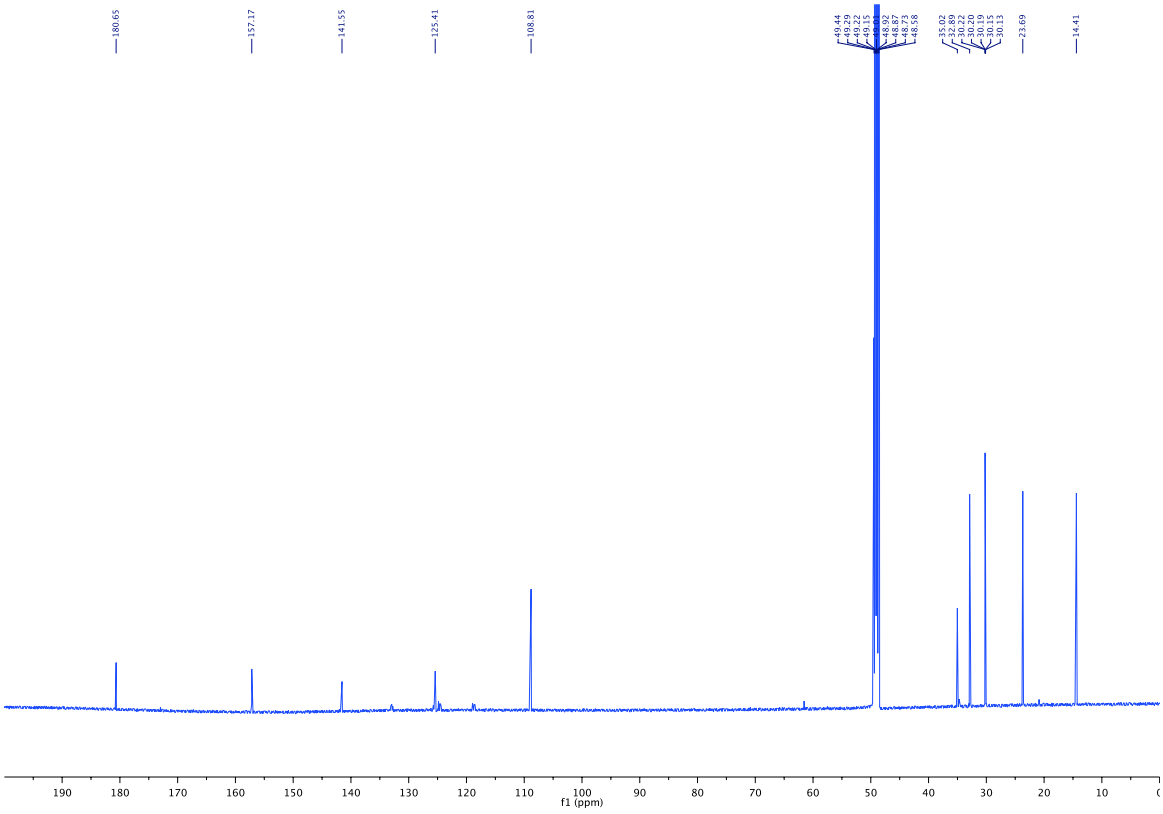

sHARMA-i-2012-0140 11 1 /opt/users/sharmai sharmai
Group Tan
.* sharmai sHARMA-i-2012-0140 (11 1) CDCl3 24.0C November_09,2012_17:19:17 Bruker AVIII 600MHz DCH cryo ZRC 2134: zgpg30 : 13C 130.000 ppm; 1H 4.000 ppm
:Carbon CDCl3 /opt/users/sharmai sharmai 6
NHBoc
NH2
Boc
S7
[ppm]

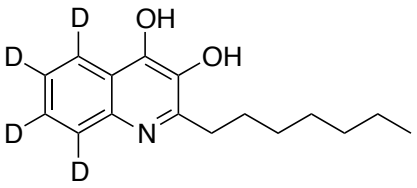

Sharma-I-2012-0142-p 1 1 /opt/users/sharmai sharmai
Proton .* sharmai Sharma-I-2012-0142-p (1 1) MeOD 24.0C November_10,2012_20:24:21 Bruker AVIII 500.26 MHz Adspice zg30 1H *.
General
DATE = 2012/11/10
TIME = 20:25
INSTRUM = spect
PULPROG = zg30
F1 (1H)
SI = 65536
SF = 500.26
SW_p = 12019.231
6
Sharma-I-2012-0142 11 1 /opt/users/sharmai sharmai
Group Tan
.* sharmai Sharma-I-2012-0142 (11 1) MeOD 24.0C November_10,2012_20:31:25 Bruker AVIII 600MHz DCH cryo ZRC 2134: zgpg30 : 13C 130.000 ppm; 1H 4.000 ppm *.
:Carbon MeOD /opt/users/sharmai sharmai 14
6


D
O
D
D
N
H
D

HHQ-d_4

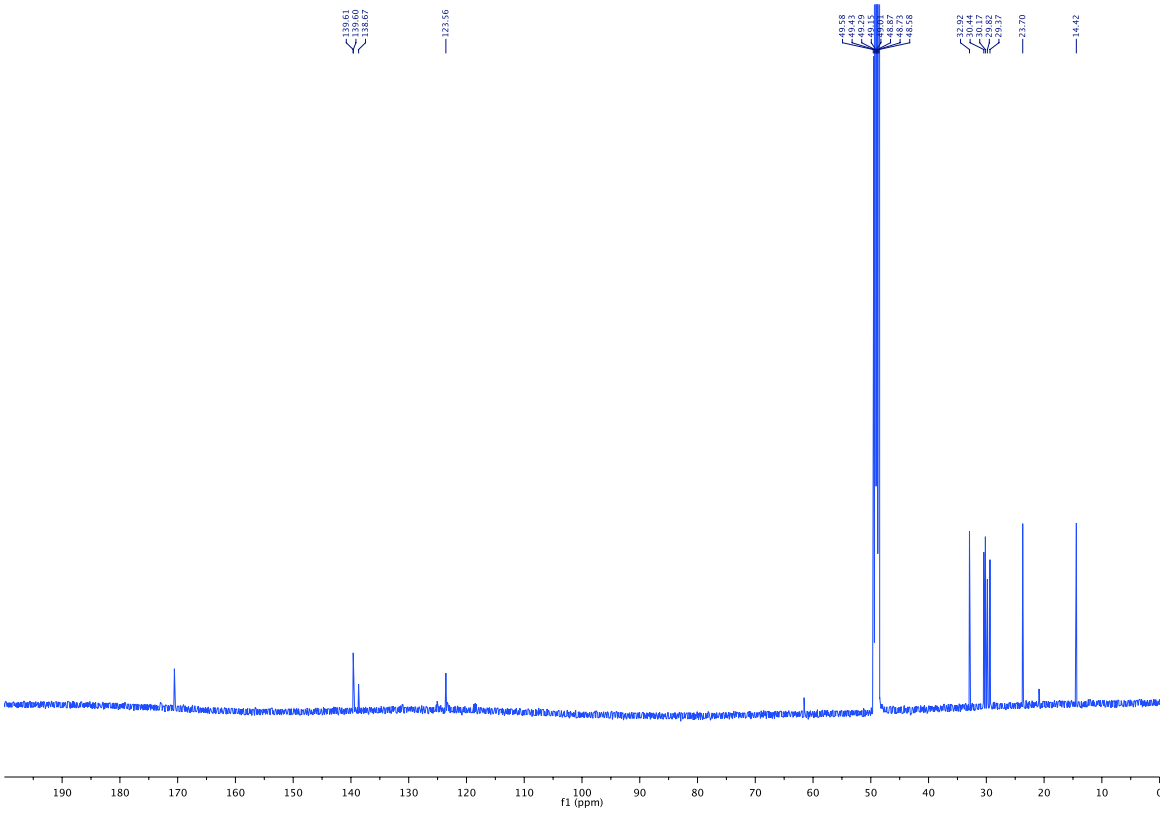
6.22
4.88
3.30
3.30
3.29
3.29
2.72
2.71
2.70
1.77
1.76
1.74
1.74
1.42
1.41
1.41
1.40
1.39
1.39
1.38
1.38
1.37
1.35
1.32
1.31
1.30
1.30
1.29
1.29
0.90
0.89
0.88
1.00
1.93
2.09
8.17
3.09
f1 (ppm)


180.65
157.17
141.55
125.41
108.81
49.44
49.29
49.12
48.97
48.82
48.73
48.58
35.02
32.80
30.22
30.20
30.19
30.15
30.13
23.69
14.41
f1 (ppm)


D
OH
D
OH
D
N
D

PQS-*d*$_4$

4.92
3.34
3.33
2.92
2.91
2.89
1.83
1.82
1.80
1.79
1.78
1.47
1.46
1.45
1.44
1.43
1.42
1.41
1.39
1.38
1.37
1.36
1.35
1.34
1.33
1.32
1.31
0.93
0.92
0.91
1.00
1.11
3.76
1.50
f1 (ppm)


139.61
139.60
138.67
123.56
49.58
49.43
49.29
49.15
48.87
48.73
48.58
32.92
30.44
30.17
29.82
29.37
23.70
14.42
f1 (ppm)