Supporting Information

Caged metabolic precursor for DT-diaphorase responsive cell labeling

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Materials. DBCO–Cy5 were purchased from KeraFAST, Inc (Boston, MA, USA). EZ-Link phosphine–PEG3–biotin, streptavidin–horseradish peroxidase (HRP), and Pierce ECL western blotting substrate were purchased from Thermo Fisher Scientific (Waltham, MA, USA). Anhydrous dichloromethane (DCM), hexane, tetrahydrofuran (THF) and dimethylformamide (DMF) were purified by passing them through alumina columns. Phosphate-buffered saline (PBS), RPMI-1640 Medium and Dulbecco's Modified Eagle's Medium (DMEM) were obtained from Invitrogen (Carlsbad, CA, USA). F-12K medium and Eagle's minimum essential medium (EMEM) were purchased from American Type Culture Collection (Manassas, VA, USA). Fetal Bovine Serum (FBS) was obtained from Lonza Walkersville Inc (Walkersville, MD, USA). BD Falcon culture plates were purchased from Fisher Scientific (Hampton, NH, USA). ProLong Gold antifade reagent was purchased from Life Technologies (Carlsbad, CA, USA). All other chemicals were purchased from Sigma-Aldrich (St. Louis, MO, USA).

Instrumentation: NMR spectra were recorded on Varian U500 (500 MHz), VXR-500 (500 MHz), or Bruker CB500 (500 MHz) spectrometers. HPLC analyses were performed on a Shimadzu CBM-20A system (Shimadzu, Kyoto, Japan) equipped with a SPD20A PDA detector (190–800 nm), an RF10Axl fluorescence detector, and an analytical C18 column (Shimadzu, 3 μm, 50*4.6 mm, Kyoto, Japan). Lyophilization was conducted in a Labconco FreeZone lyophilizer (Kansas City, MO, USA). Confocal laser scanning microscopy (CLSM) images were taken on a Zeiss LSM 700 Confocal Microscope (Carl Zeiss, Thornwood, NY, USA). Flow cytometry analyses of cells were conducted with a BD FACS Canto 6-color flow cytometry analyzer (BD, Franklin Lakes, NJ, USA). Western blotting membrane was imaged in ImageQuant LAS 4000 gel imaging system (GE, Pittsburgh, PA, USA).

Cell culture. Bxpc-3 pancreatic cancer cell, MIA PaCa-2 pancreatic cancer cell, A549 lung cancer cell, MDA-MB-231 triple-negative breast cancer cell, HEK293 embryonic kidney cells, and IMR-90 human fibroblast cell lines were purchased from American Type Culture Collection (Manassas, VA, USA). Cells were cultured in RPMI-1640 containing 10% FBS, 100 units/ml Penicillin G and 100 μg/ml streptomycin (Invitrogen, Carlsbad, CA, USA) at 37 °C in 5% CO2 humidified air unless otherwise noted. MIA Paca-2 cells were cultured in DMEM containing 10% FBS. A549 cells were cultured in F-12K medium containing 10% FBS. HEK293 and IMR-90 cells were cultured in EMEM containing 10% FBS. HBEC-5i were cultured in EMEM with 10% FBS and 40 μg/ml endothelial growth supplement (ECGS). MCF-10A cells were cultured in MEGM BulletKit.

General method for flow cytometry quantification of surface azido sugar

Cells were seeded in 6-well plate one day before azido sugar treatment for fully attachment. 2 mL of full growth media were replenished for each well before adding azido sugar. Azido sugars were added in stock solution of DMSO (100 mM) into cell culture wells to reach final designed concentration. The cells were cultured at 37 °C in 5% CO2 humidified air for designed time length. Then the culture media were removed and cells were washed with PBS for three times to remove residua free azido sugar. Fresh culture media with 50 μM DBCO-sulfo-Cy5 were treated to cells for 45 min. Free dye were washed away with PBS for

three times. The cells labeled with Cy5 were detached by trypsin and then fixed in 4% polyformaldehyde for flow cytometry analysis.

HPLC quantification of enzymatic degradation of HQ-NN-AAM

NADPH stock solution (10mM in PBS, 100 μL), human DT-diaphorase stock solution (200 ug/mL in PBS, 20 μL) and HQ-NN-AAM stock solution (10 mg/mL in DMSO, 40 μL) were mixed and diluted in 840 μL of PBS. The mixture was incubated at 37 °C. At each assigned time data point, 20 μL of reaction mixture were quenched with 80 μL of cold HPLC water containing 0.1% TFA (n = 3). The HPLC trace were collected at 204 nm (for quantification of non-aromatic molecule) and 262 nm (for quantification of molecule with aromatic domain). HPLC elution gradient were set from 20 % Acetonitrile to 80 % in 0.1 % TFA water. Each starting material, intermediate and final compound were quantified to standard curve generated from pure compounds.

General method for Western blotting analysis of total azido glycoprotein

Cells lysates were obtained by adding ice cold RIPA buffer to cells with azido sugar treatment. The lysate was incubated at 4 °C for 1 h and sonicated for 5 min for fully decomposition of cells. The lysate debris were removed by centrifuge. Protein concentration of cell lysate were quantified using BCA assay. Cell lysate were added with iodoacetamide stock solution (1.0 M in DMSO) to a final concentration of 15 mM and incubated at 37 °C for 30 min for fully blocking of thiol groups. DBCO-$(PEG)_4$-biotin were added to the mixture to a final concentration of 50 μM and incubated 37 °C for 2 hours. The protein mixture labeled with biotin were mixed with sample buffer subjected to electrophoresis. After standard membrane transfer, protein bands with biotin modification were detected by streptavidin-HRP and ECL western Blotting Substrate.

General procedures for confocal imaging of azido sugar labeled cells.

Cells were seeded onto coverslips in a 6-well plate at a density of 4×10^4 cells per well and allowed to attach for 12 h. Azido sugar (50 μM) was added and the cells were incubated at 37 °C for 72 h. After washing with PBS, cells were incubated with DBCO–Cy5 (50 μM) for 1 h followed by staining of cell nuclei and membrane with Hoechst 33342 (10 μM) and CellMask orange plasma membrane stain (1 μg/ml) for 10 min, respectively, and fixed with 4% paraformaldehyde (PFA) solution. The coverslips were mounted onto microscope slides and imaged under a confocal laser scanning microscope.

Inhibition assay of azido sugar cell labeling.[1]

Cells were seeded in 6-well plate one day before azido sugar treatment for fully attachment. DMXAA stock solution (200 mM in DMSO) were added to freshly replenished cell culture media to final concentration designed (e.g. 100 μM). The cells were cultured for 2 h for inhibition of DTD activity before azido sugar stock solution were added. The cells were further cultured for 12 h and subjected to either flow cytometry analysis or Western blotting analysis of azido glycoprotein.

Supporting figures

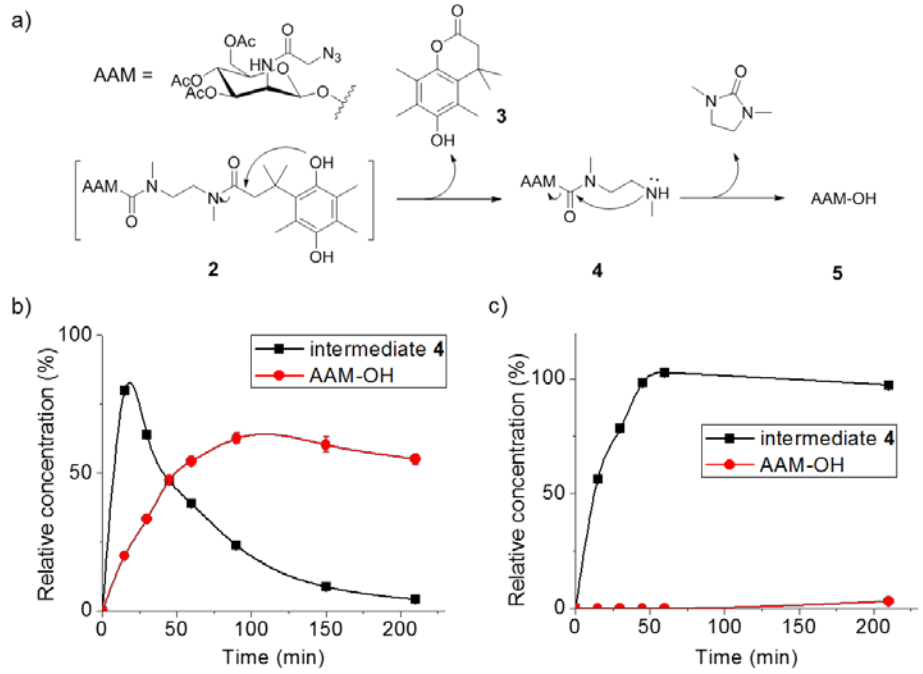


Figure S1. a) Degradation scheme of HQ-NN-AAM. b,c) Relative concentration as compared to starting HQ-NN-AAM *vs* time plot of intermediate **4** and active metabolite AAM-OH in b) pH 7.0 buffer and c) pH 5.0 buffer. Intermediate **4** has faster degradation kinetic in neutral condition as compared to acidic condition.

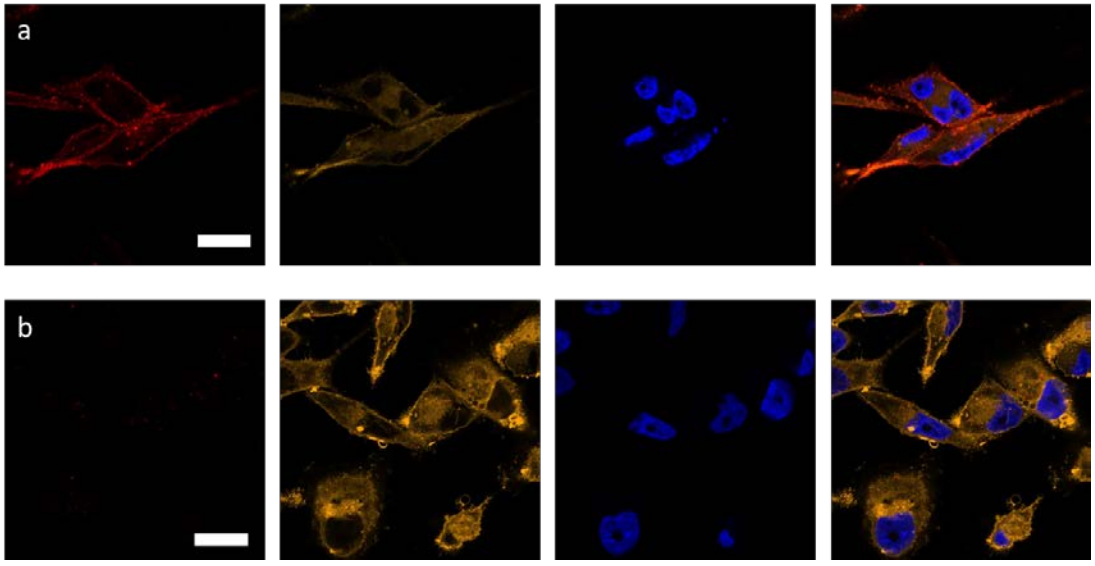


Figure S2. Confocal image of MDA-MB-231 cells treated with a) 50 μM of HQ-NN-AAM or b) 50 μM of Boc-NN-AAM. Red channel represents Cy5 signal from DBCO-Cy5 reacting with surface azide. Yellow channel represents CellMask orange membrane stain. Blue channel represents Hoechst cell nucleus stain. Last picture is the merge of three channels. Scale bar = 20 μm.

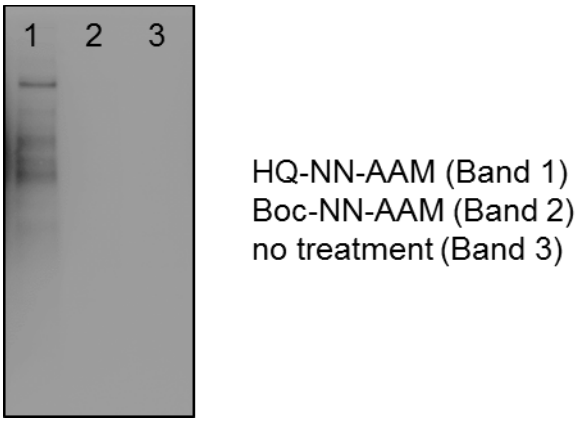


Figure S3. Western blotting of azido labeled glycoproteins in MDA-MB-231 cell treated with 50 μM of HQ-NN-AAM (Band 1), 50 μM of Boc-NN-AAM (Band 2) and no treatment (Band 3).

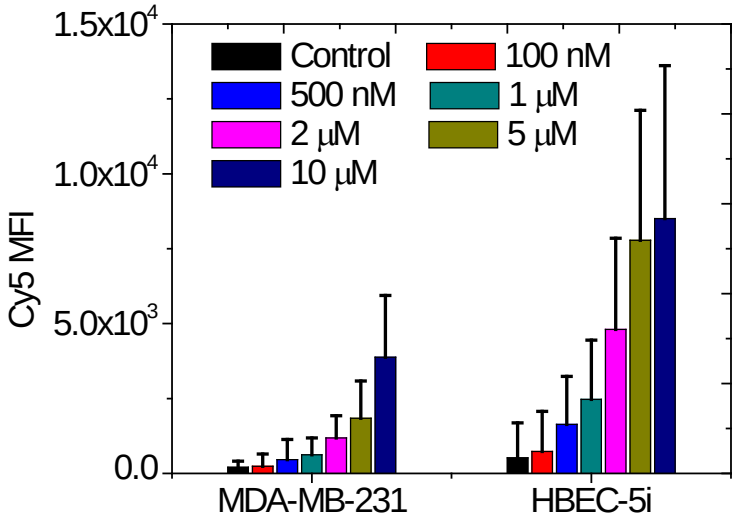


Figure S4. Concentration dependent labeling in MDA-MB-231 and HBEC-5i cell line by HQ-NN-AAM.

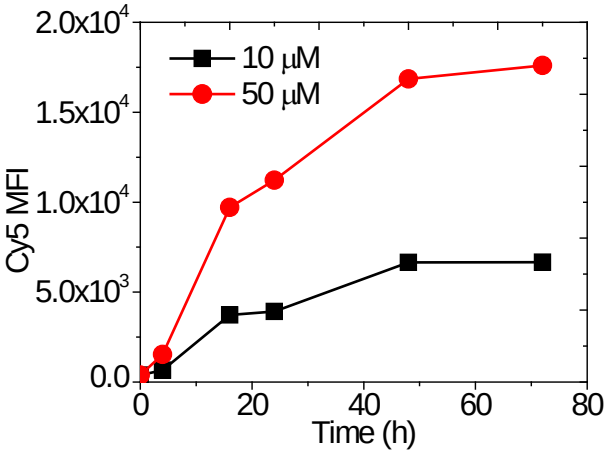


Figure S5. Labeling kinetics of different concentrations of HQ-NN-AAM in MDA-MB-231 cells analyzed by flow cytometry method

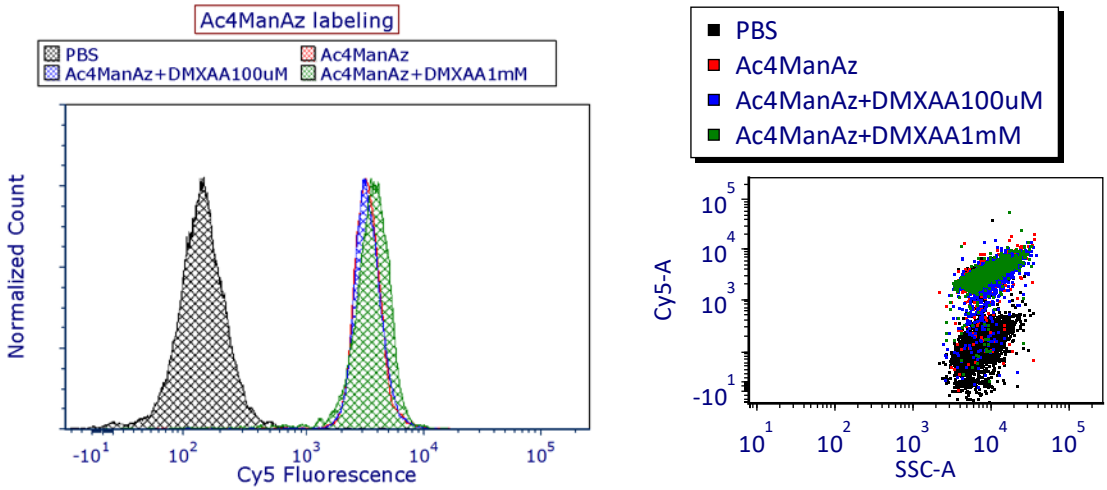


Figure S6. Flow cytometry histogram and dot plot of cell surface azido concentration of MDA-MB-231 cell treated with AAM and different concentration of DMXAA, DMXAA showed no effect on AAM labeling.

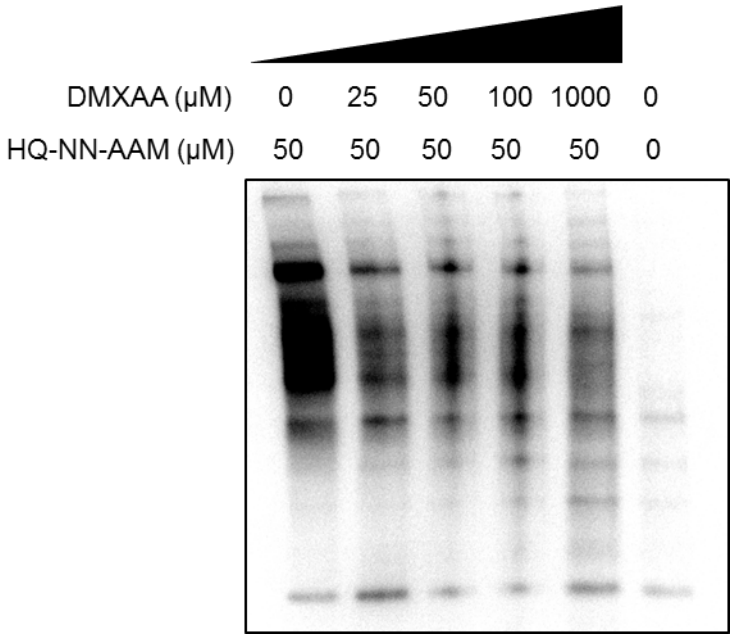
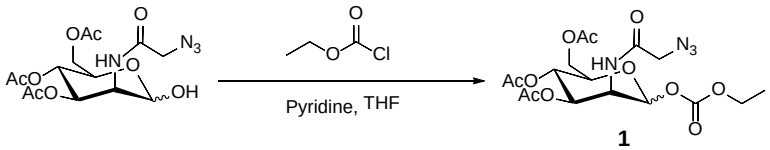


Figure S7. Western blotting of azido labeled glycoproteins in MDA-MB-231 cell treated with HQ-NN-AAM and different concentration of DMXAA (DT-diaphorase inhibitor).

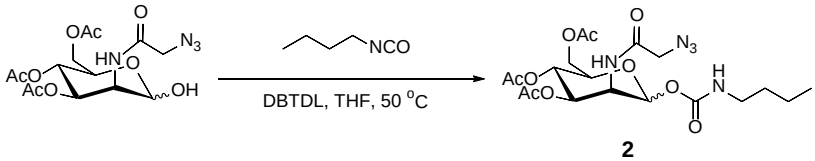
Synthetic procedures

$Ac_4ManNAz$ (Ac-E-AAM) and $Ac_3ManNAzOH$ (AAM) were synthesized according to literature report.[2]

Supplementary Scheme 1. Synthetic route of Et-O-AAM (**1**) and Bu-N-AAM (**2**)



1



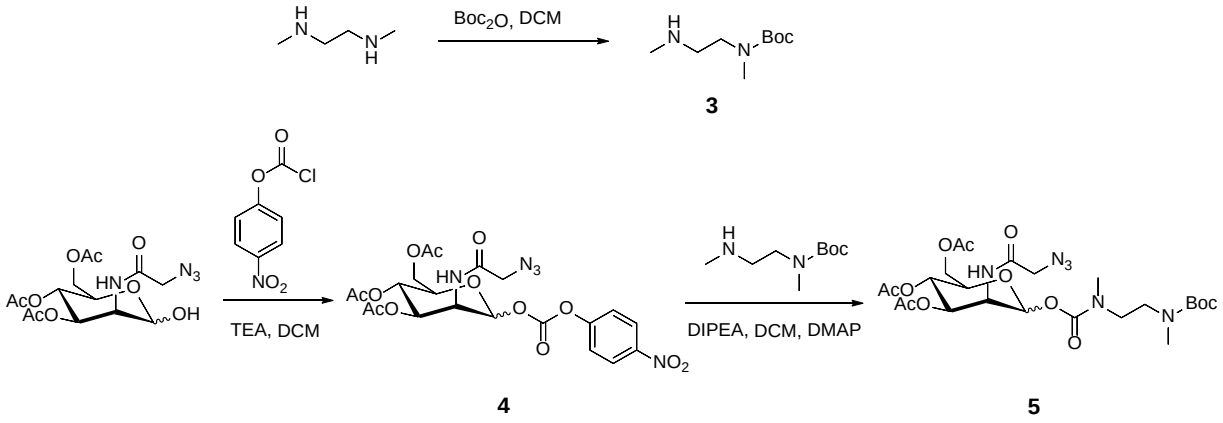
2

Synthesis of Et-O-AAM (1). $Ac_3ManNAzOH$ (78 mg, 0.2 mmol) were dissolved in 500 μL of anhydrous THF. Ethyl chloroformate (42 mg, 0.4 mmol) and pyridine (80 μL, 1.0 mmol) were added to the reaction mixture. The mixture were stirred at room temperature until starting material disappeared on TLC plate. The reaction were quenched by adding methanol and the solvent were removed in vacuum. The crude product was purified by silica gel column chromatography using ethyl acetate/hexane (1/1, v/v) to yield a colorless oil (65% yield, > 90% α isomer). ^{1}H NMR (500 MHz, $CDCl_3$): δ 6.55 (d, 1H, *J* = 9.2 Hz), 5.94 (d, 1H, *J* = 1.9 Hz), 5.37 (dd, 1H, *J* = 10.2, 4.3 Hz), 5.23 (t, 1H, *J* = 10.1 Hz), 4.69 (ddd, 1H, *J* = 9.2, 4.3, 2.0 Hz), 4.26 (m, 3H), 4.10 (m, 4H), 2.11 (s, 3H), 2.05 (s, 3H), 2.00 (s, 3H), 1.35 (t, 3H, *J* = 7.1 Hz). ^{13}C NMR (125 MHz, $CDCl_3$): δ 170.44, 169.85, 169.55, 166.65, 152.53, 94.41, 68.62, 65.15, 64.97, 61.65, 52.44, 49.25, 41.01, 20.72, 20.62, 20.58, 14.11. HRMS-ESI (*m/z*): $[M+Na]^+$ calculated for $C_{17}H_{24}N_4O_{11}Na$, 483.1339; observed, 483.1332.

Synthesis of Bu-N-AAM (2). $Ac_3ManNAzOH$ (78 mg, 0.2 mmol) were dissolved in 500 μL of anhydrous THF. *n*-butyl isocyanate (30 mg, 0.3 mmol) and dibutyltin dilaurate (5 μL) were added to the reaction mixture. The mixture were stirred at 50 °C until starting material disappeared on TLC plate. The solvent were removed in vacuum. The crude product was purified by silica gel column chromatography using ethyl acetate/hexane (1/1, v/v) to yield a colorless oil (72 % yield, mixture of α and β isomer, α:β = 1:4). ^{1}H NMR (500 MHz, $CDCl_3$), β isomer: δ 6.63 (d, 1H, *J* = 9.0 Hz), 5.87 (d, 1H, *J* = 1.6 Hz), 5.16 (t, 1H, *J* = 9.9 Hz), 5.06 (dd, 1H, *J* = 9.9, 3.9 Hz), 4.84 (t, 1H, *J* = 5.9 Hz), 4.72 (ddd, 1H, *J* = 9.0, 3.8, 1.6 Hz), 4.01-4.28 (m, 4H), 3.81 (ddd, 1H, *J* = 9.8, 4.3, 2.4 Hz), 3.19 (m, 2H), 2.11 (s, 3H), 2.05 (s, 3H), 2.00 (s, 3H), 1.49 (m, 2H),

1.34 (m, 2H), 0.92 (t, 3H); α isomer: δ 6.55 (d, 1H, *J* = 9.3 Hz), 5.97 (d, 1H, *J* = 1.9 Hz), 5.32 (dd, 1H, *J* = 10.0, 4.2 Hz), 5.21 (t, 1H, *J* = 10.1 Hz), 4.91 (t, 1H, *J* = 5.9 Hz), 4.61 (ddd, 1H, *J* = 9.4, 4.2, 2.0 Hz), 4.01-4.28 (m, 5H), 3.19 (m, 2H), 2.11 (s, 3H), 2.05 (s, 3H), 2.00 (s, 3H), 1.49 (m, 2H), 1.34 (m, 2H), 0.92 (t, 3H),; ^{13}C NMR (125 MHz, $CDCl_3$), β isomer: δ 170.51, 170.11, 169.57, 167.16, 153.08, 91.01, 73.08, 71.67, 64.98, 61.70, 52.67, 50.09, 41.03, 31.67, 20.72, 20.68, 20.65, 19.84, 13.65; α isomer: δ 170.52, 170.24, 169.49, 166.59, 152.93, 91.93, 69.85, 69.07, 65.23, 61.81, 52.46, 49.46, 40.95, 31.72, 20.79, 20.60, 20.04, 19.86, 13.68; HRMS-ESI (*m/z*): $[M+Na]^+$ calculated for $C_{19}H_{29}N_5O_{10}Na$, 510.1812; observed, 510.1826.

Supplementary Scheme 2. Synthetic route of Boc-NN-AAM (**5**)



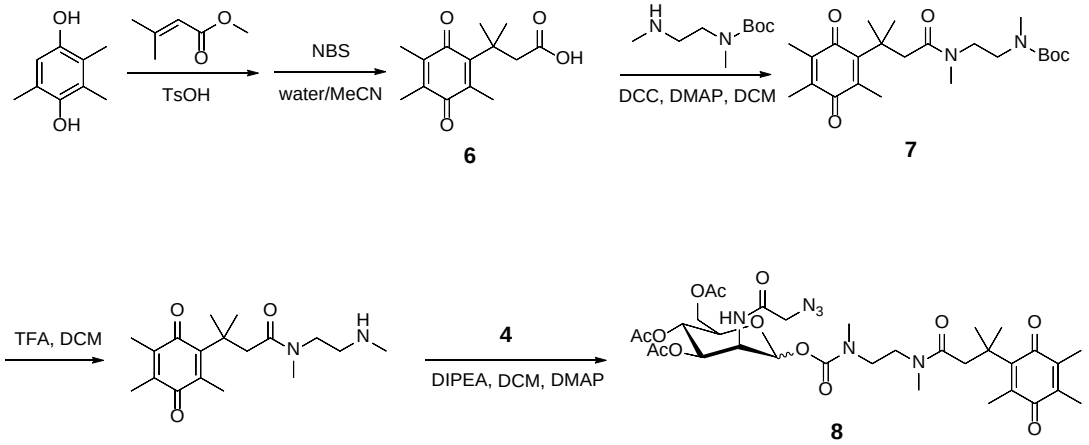
Synthesis of tert-butyl methyl(2-(methylamino)ethyl)carbamate (Boc-NN, 3). *N,N'*-dimethylethylenediamine (4.4 g, 50 mmol) were dissolved in 200 mL of DCM and cooled to 0 °C. Di-tert-butyl dicarbonate (2.7 g, 12.5 mmol) were dissolved in 40 mL of DCM and added to the diamine solution dropwise over 1 h. The mixture was slowly warmed up to room temperature and stirred for 16 h. After removing solvent in vacuum, the crude product was purified by silica gel column chromatography using ethyl acetate/ methanol/ trimethylamine (87/10/3, v/v) to yield colorless oil (80 %). ^{1}H NMR (500 MHz, $CDCl_3$): δ 3.32 (t, 2H), 2.97 (s, 3H), 2.72 (t, 2H), 2.44 (s, 3H), 1.45 (s, 9H); ^{13}C NMR (125 MHz, $CDCl_3$): δ 155.94, 79.44, 49.73, 48.43, 36.32, 34.71, 28.45; HRMS-ESI (*m/z*): $[M+H]^+$ calculated for $C_9H_{21}N_2O_2$, 189.1603; observed, 189.1606.

Synthesis of NB-O-AAM (4). Ac_3ManNAzOH (78 mg, 0.2 mmol) were dissolved in 3 mL of anhydrous DCM. 4-Nitrobenzyl chloroformate (48 mg, 0.24 mmol) and triethylamine (50 µL, 0.4 mmol) were added to the reaction mixture. The mixture was stirred at room temperature until starting material disappeared on TLC plate. The reaction was quenched by adding 2 mL of 1 M dilute HCl in water. The product was extracted with DCM (10 mL x 3) and 4-nitrophenol was washed with saturated $NaHCO_3$ (50 mL x 4). The organic phase were dried and evaporated under vacuum to give colorless product (85 %, mixture of α and β isomer,

α:β = 2:1). ^{1}H NMR (500 MHz, $CDCl_3$) β isomer: δ 8.32 (d, 2H, *J* = 9.2 Hz), 7.45 (d, 2H, *J* = 9.2 Hz), 6.58 (d, 1H, *J* = 9.1 Hz), 6.07 (d, 1H, *J* = 1.9 Hz), 5.43 (dd, 1H, *J* = 10.1, 4.4 Hz), 5.29 (t, 1H, *J* = 10.1 Hz), 4.80 (ddd, 1H, *J* = 9.1, 4.4, 2.0 Hz), 4.07-4.34 (m, 5H), 2.13 (s, 3H), 2.09 (s, 3H), 2.04 (s, 3H); α isomer: δ 8.34 (d, 2H, *J* = 9.3 Hz), 7.50 (d, 2H, *J* = 9.3 Hz), 5.93 (d, 1H, *J* = 6.7 Hz), 5.45 (dd, 1H, *J* = 10.1, 4.4 Hz), 5.20 (dd, 1H, *J* = 6.6, 5.6 Hz), 4.92 (m, 1H), 4.47 (d, 1H, *J* = 2.4 Hz), 4.07-4.34 (m, 5H), 2.12 (s, 3H), 2.10 (s, 3H), 2.07 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$): δ 170.38, 169.89, 169.48, 166.79, 154.86, 150.17, 145.78, 125.48, 121.64, 95.72, 61.56, 52.43, 49.16, 20.71, 20.62, 20.59; HRMS-ESI (*m/z*): $[M+Na]^+$ calculated for $C_{21}H_{23}N_5O_{13}Na$, 576.1190; observed, 576.1180.

Synthesis of Boc-NN-AAM (5). NB-O-AAM (**4**, 55 mg, 0.1 mmol), Boc-NN (**3**, 22 mg, 0.12 mmol), *N,N*-diisopropylethylamine (DIPEA, 25 µL, 0.15 mmol) and 4-dimethylaminopyridine (DMAP, 1 mg, 0.01 mmol) were dissolved in 0.5 mL anhydrous DCM. The solution mixture was stirred at room temperature for 16 h. The solution was then diluted with 10 mL DCM and washed with 1 M HCl (50 mL x 2) and saturated $NaHCO_3$ (50 mL x 3). The organic phase was dried using anhydrous Na_2SO_4 and concentrate in vacuum to give crude product. The crude product was further purified by silica gel column chromatography using ethyl acetate/ hexane (1/1, v/v) to give white wax like powder (70 %). The product was mixture of cis- and trans-isomers on carbamate bonds. ^{1}H NMR (500 MHz, $CDCl_3$): δ 6.56 (m, 1H), 6.02 (s, 1H), 5.28 (m, 2H), 4.66 (m, 1H), 4.12 (m, 5H), 3.38 (s, 4H), 3.01 (d, 3H), 2.88 (m, 3H), 2.12 (s, 3H), 2.06 (s, 3H), 1.99 (s, 3H), 1.45 (s, 9H); ^{13}C NMR (125 MHz, $CDCl_3$): δ 170.48, 170.17, 170.12, 169.47, 166.58, 166.57, 92.56, 70.25, 69.19, 65.05, 61.76, 54.00, 53.43, 52.46, 49.46, 35.69, 29.70, 28.39, 20.76, 20.74, 20.65, 20.59; HRMS-ESI (*m/z*): $[M+Na]^+$ calculated for $C_{24}H_{38}N_6O_{12}Na$, 625.2445; observed, 625.2433.

Supplementary Scheme 3. Synthetic route of HQ-NN-AAM (**8**)



Synthesis of HQ-COOH (6). HQ-COOH was synthesized using similar route as reported before.[3] Methyl sulfonic acid (40 mL) were heated to 70 °C. Methyl 3-methyl-2-butenoate (3.6 g, 31.5 mmol) and trimethylhydroquinone (4.0 g, 26.3 mmol) were added under stirring. The mixture was stirred for another

90 min at 70 °C. The reaction was quenched by adding 500 mL of DI water and extracted with ethyl acetate three times. The organic phase was washed with water, saturated sodium bicarbonate, and brine. The crude product was recrystallized from chloroform and hexane to yield pure intermediate. The intermediate (3.0 g, 12.8 mmol) was then suspended in 150 mL of 10 % acetonitrile in water. *N*-bromosuccinimide (2.4 g, 13.4 mmol) dissolved in 40 mL of acetonitrile was added to the suspension slowly. The mixture was stirred at room temperature for 1 hour. The reaction mixture was extracted with ether (200 mL x 3) and organic phase was combined and dried. The solvent was evaporated to give yellow powder crude product. Pure HQ-COOH was obtained by recrystallizing in DCM/hexane. ^{1}H NMR (500 MHz, $CDCl_3$): δ 3.02 (s, 2H), 2.14 (s, 3H), 1.95 (q, 3H), 1.92 (q, 3H), 1.43 (s, 6H); ^{13}C NMR (125 MHz, $CDCl_3$): δ 190.83, 187.42, 177.94, 151.97, 142.95, 139.06, 138.41, 47.14, 37.95, 28.81, 14.32, 12.51, 12.11; HRMS-ESI (*m/z*): $[M-H]^-$ calculated for $C_{14}H_{17}O_4$, 249.1132; observed, 249.1125.

Synthesis of HQ-NN-Boc (7). HQ-COOH (**6**, 1.5 g, 6.0 mmol), Boc-NN (**3**, 0.94 g, 5.0 mmol), dicyclocabodiimide (DCC, 1.55 g, 7.5 mmol), and DMAP (61 mg, 0.5 mmol) were dissolved in 50 mL of anhydrous DCM. The mixture was stirred at room temperature for 16 h. The mixture was filtered and filtrate was concentrated in vacuum. Pure product was obtained by silica gel column chromatography using ethyl acetate/ hexane (1/2, v/v) as eluent to get yellow oil. The product was mixture of cis- and trans- isomers on carbamate bonds. ^{1}H NMR (500 MHz, $CDCl_3$): δ 3.27-3.41 (m, 4H), 2.82-2.98 (m, 8H), 2.11 (m, 3H), 1.89-1.93 (m, 6H), 1.41-1.48 (m, 15H); ^{13}C NMR (125 MHz, $CDCl_3$): δ 191.22, 187.66, 172.12, 155.64, 154.57, 143.23, 137.93, 136.08, 79.49, 47.97, 46.72, 37.42, 35.03, 28.61, 28.47, 14.18, 14.12, 12.66, 12.59, 12.09; HRMS-ESI (*m/z*): $[M+H]^+$ calculated for $C_{23}H_{37}N_2O_5$, 421.2702; observed, 421.2710.

Synthesis of HQ-NN-AAM (8). HQ-NN-Boc (**7**, 400 mg, 0.95 mmol) were dissolved in 5 mL of DCM and added 5 mL of trifluoroacetic acid (TFA). The solution was stirred at room temperature for 2 h and the solvent was evaporated in vacuum. The crude product was re-dissolved in 20 mL of anhydrous DCM. NB-O-AAM (**4**, 300 mg, 0.54 mmol), DIPEA (1.0 mL, excess amount), DMAP (10 mg, 0.08 mmol) were added to the reaction mixture. The reaction mixture was stirred at room temperature for 16 h. 10 mL of 1 M HCl was added to quench the reaction. The reaction mixture was extracted using DCM (100 mL x 3) and the organic phase was combined. The organic phase was washed with saturated $NaHCO_3$ (150 mL x 3) and dried using anhydrous Na_2SO_4. The solvent was evaporated in vacuum to get crude product. Pure product was obtained after separation by silica gel column chromatography using ethyl acetate/hexane (1/1, v/v) as the eluent to get yellow powder. The product was a mixture of cis- and trans- isomers on carbamate bonds (75 % yield). ^{1}H NMR (500 MHz, $CDCl_3$): δ 6.56 (m, 1H), 6.01 (m, 1H), 5.27 (m, 2H), 4.67 (m, 1H), 4.25 (m, 1H), 4.10 (m, 4H), 3.44 (m, 4H), 2.99 (m, 8H), 2.02 (m, 18H), 1.42 (s, 6H); ^{13}C NMR (125 MHz, $CDCl_3$): δ 191.19, 187.59, 172.47, 170.44, 170.10, 169.47, 166.58, 154.42, 153.09, 143.00, 138.09, 136.60, 92.56, 70.14, 69.12, 65.02, 61.79, 60.39, 52.47, 49.51, 48.16, 45.12, 37.65, 36.49, 33.63, 28.55, 21.06, 20.74, 20.59, 14.22, 12.69, 12.12; HRMS-ESI (*m/z*): $[M+H]^+$ calculated for $C_{33}H_{47}N_6O_{13}$, 735.3201; observed, 735.3188.

NMR spectra

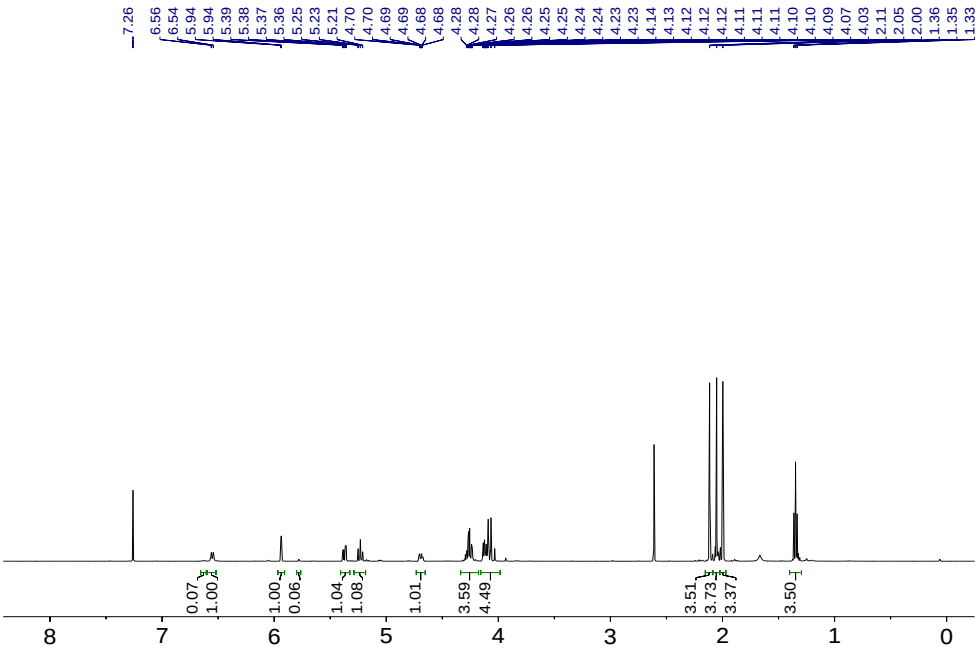


Figure S8. ^{1}H NMR spectrum of Et-O-AAM (**1**) in $CDCl_3$

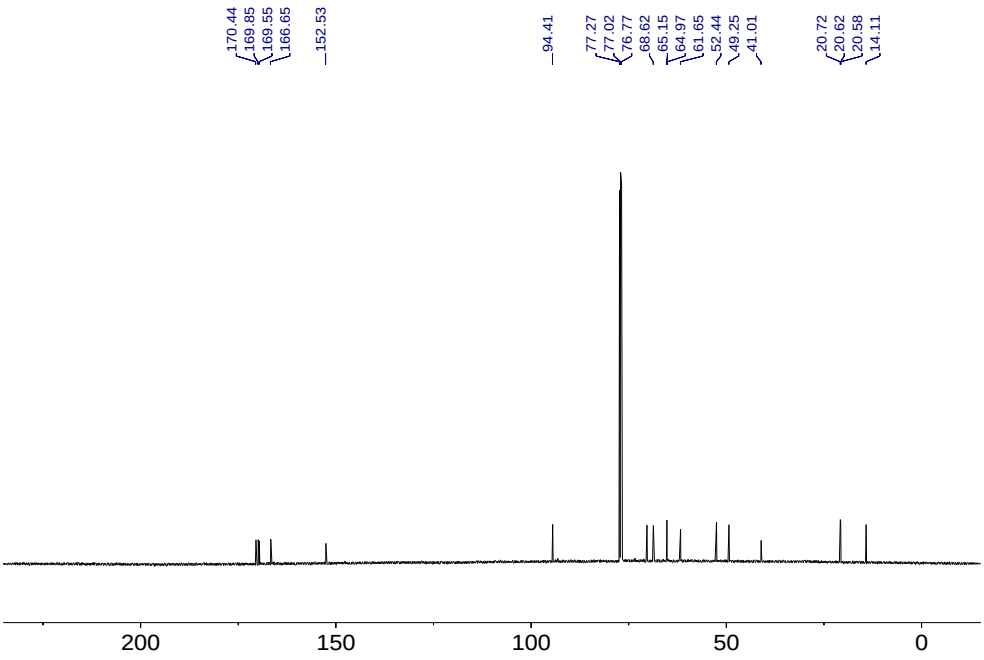


Figure S9. ^{13}C NMR spectrum of Et-O-AAM (**1**) in $CDCl_3$

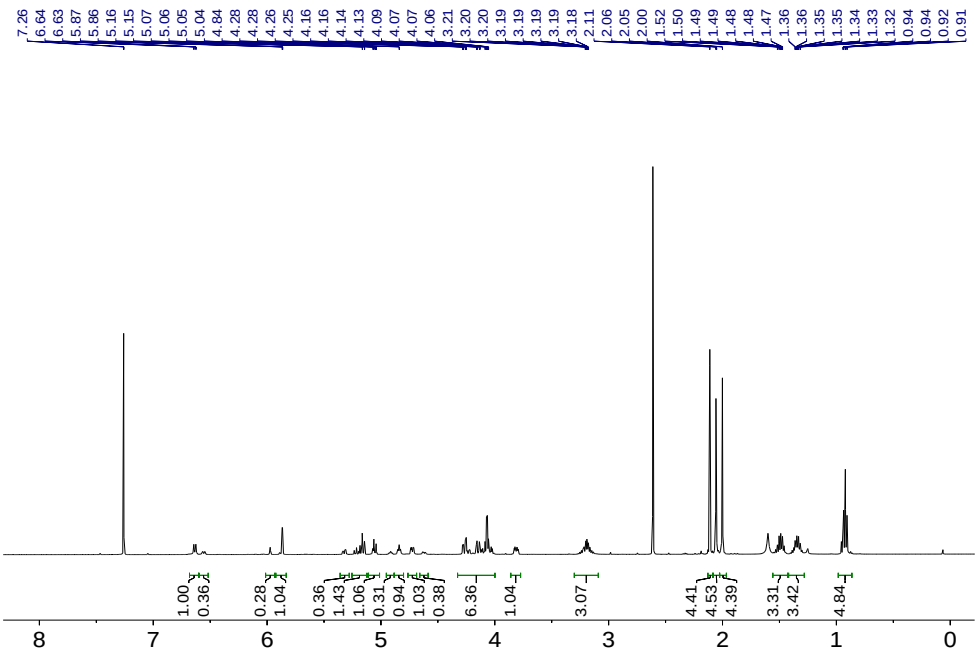


Figure S10. ^{1}H NMR spectrum of Bu-N-AAM (**2**) in $CDCl_3$

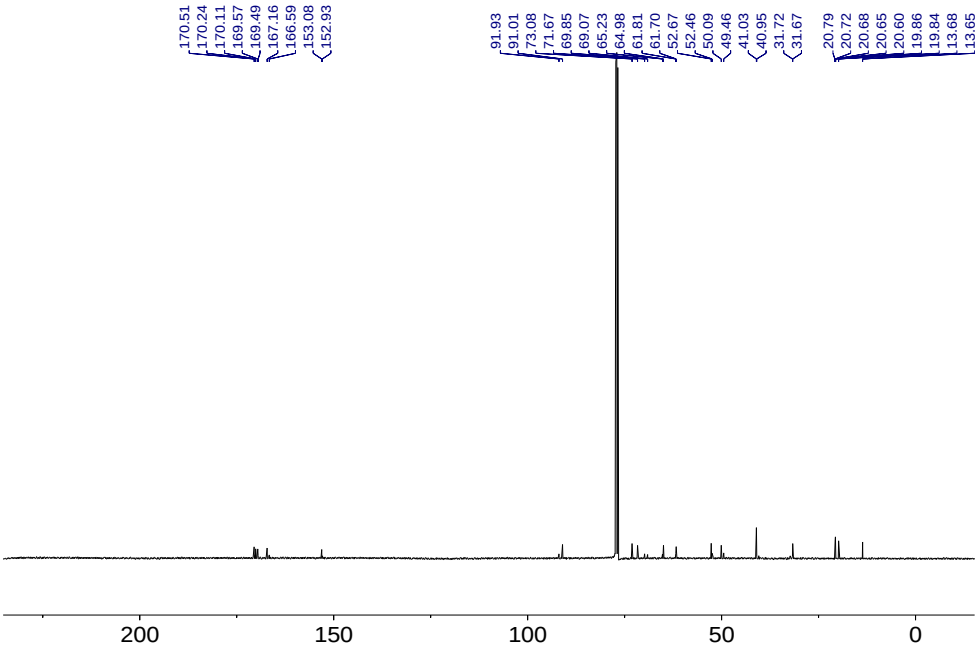


Figure S11. ^{13}C NMR spectrum of Bu-N-AAM (**2**) in $CDCl_3$

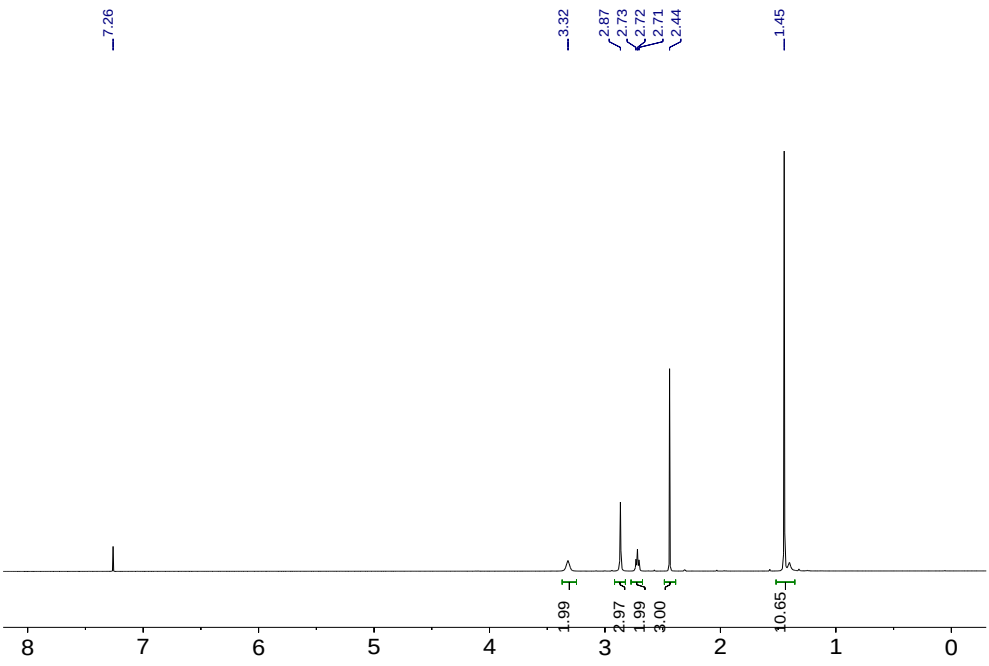


Figure S12. ^{1}H NMR spectrum of Boc-NN (**3**) in $CDCl_3$

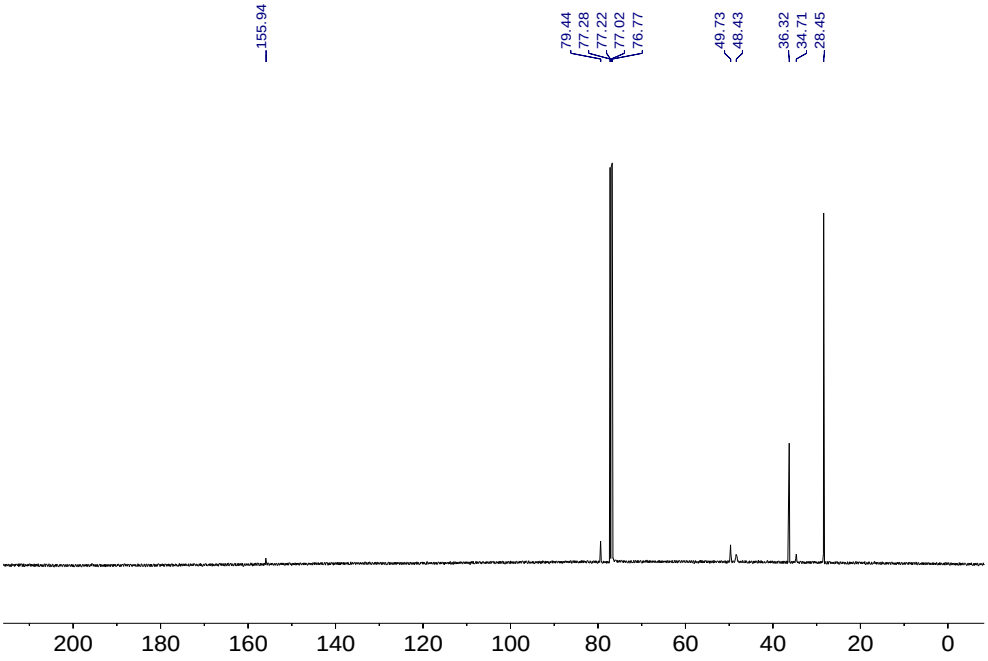


Figure S13. ^{13}C NMR spectrum of Boc-NN (**3**) in $CDCl_3$

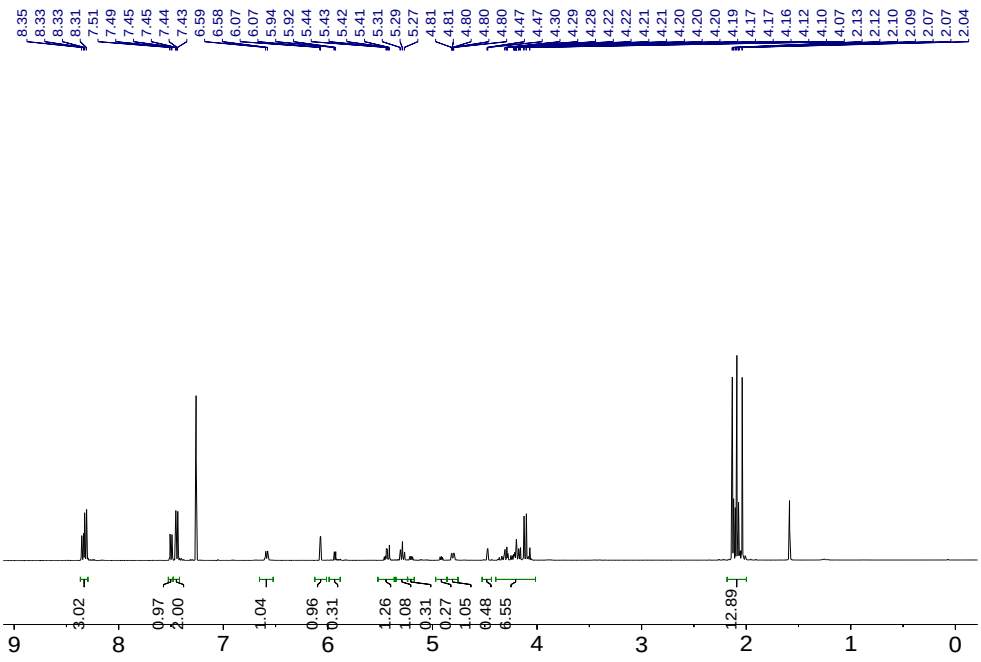


Figure S14. ^{1}H NMR spectrum of NB-O-AAM (**4**) in $CDCl_3$

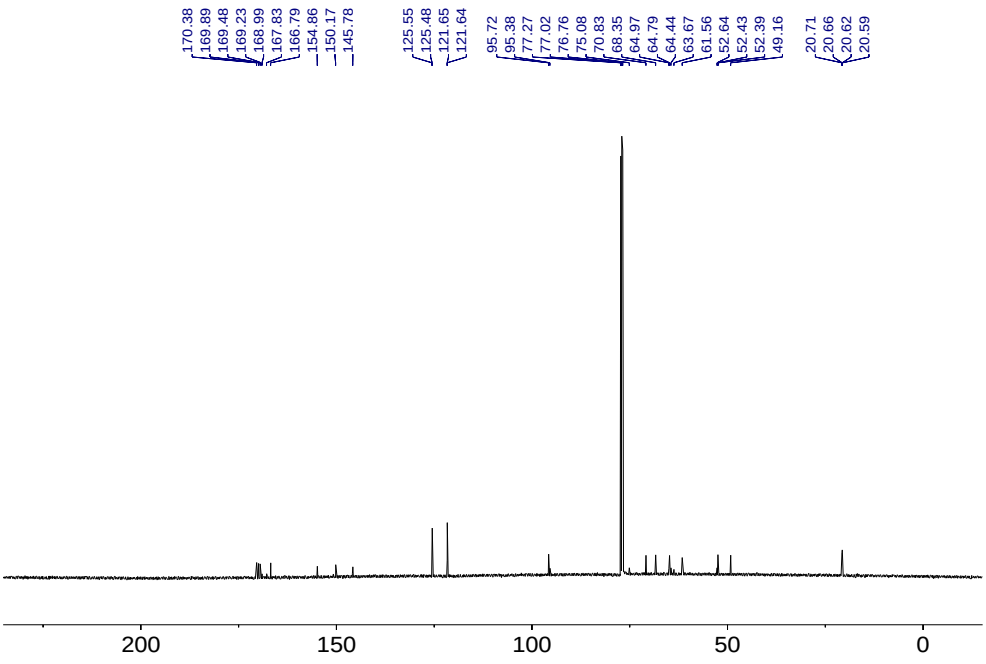


Figure S15. ^{13}C NMR spectrum of NB-O-AAM (**4**) in $CDCl_3$

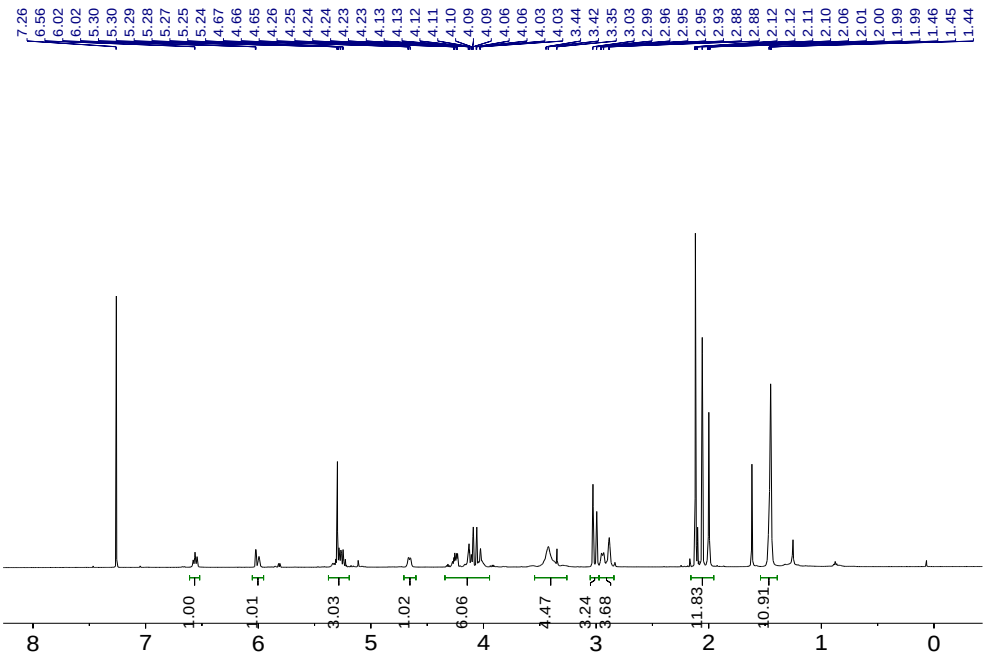


Figure S16. ^{1}H NMR spectrum of Boc-NN-AAM (**5**) in $CDCl_3$

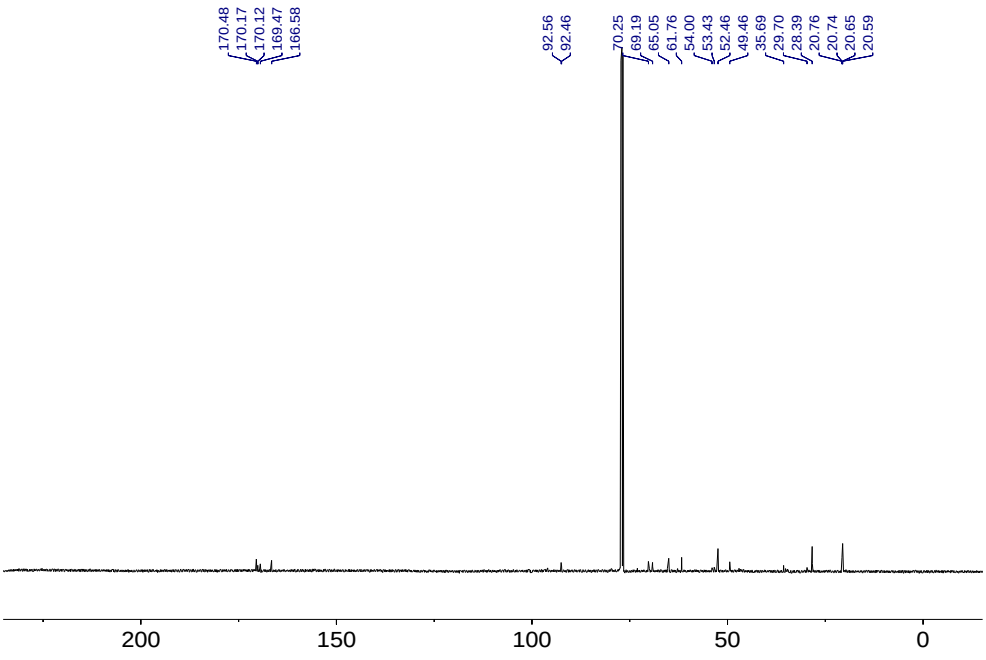


Figure S17. ^{13}C NMR spectrum of Boc-NN-AAM (**5**) in $CDCl_3$

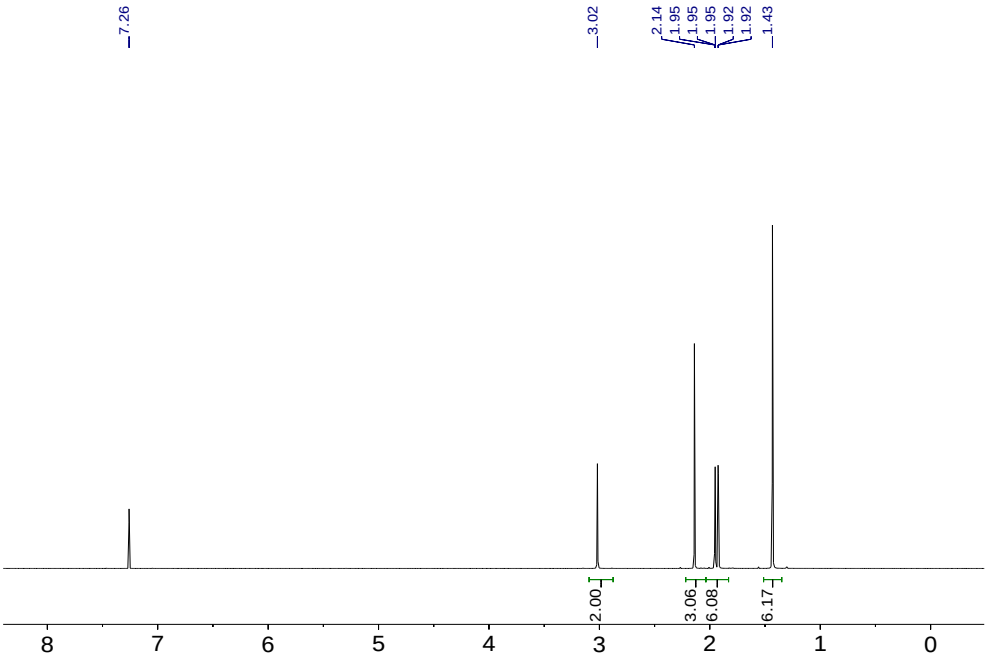


Figure S18. ^{1}H NMR spectrum of HQ-COOH (**6**) in $CDCl_3$

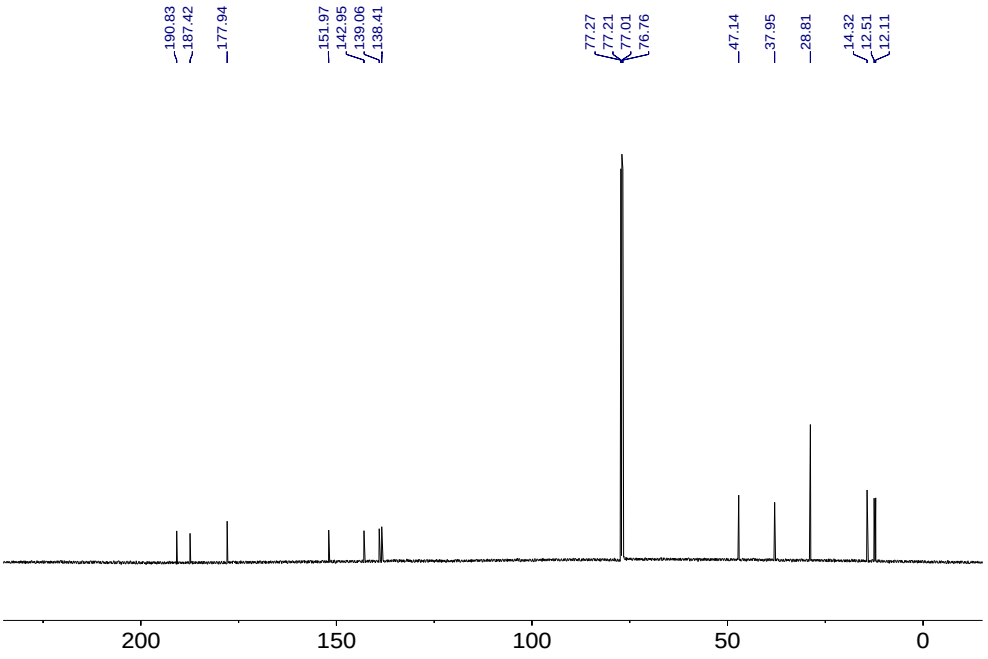


Figure S19. ^{13}C NMR spectrum of HQ-COOH (**6**) in $CDCl_3$

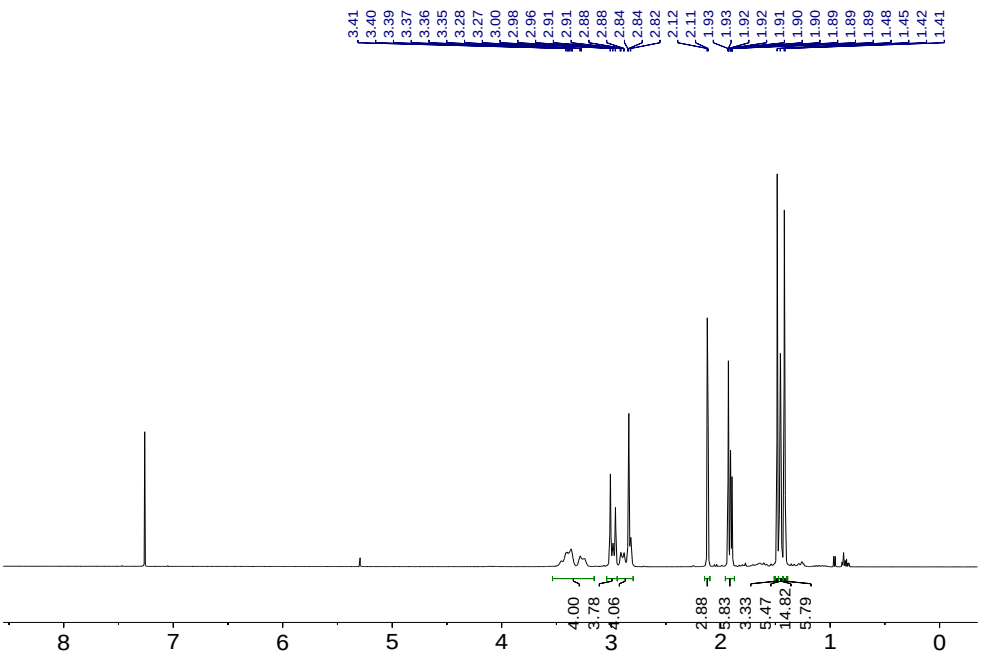


Figure S20. ^{1}H NMR spectrum of HQ-NN-Boc (**7**) in $CDCl_3$

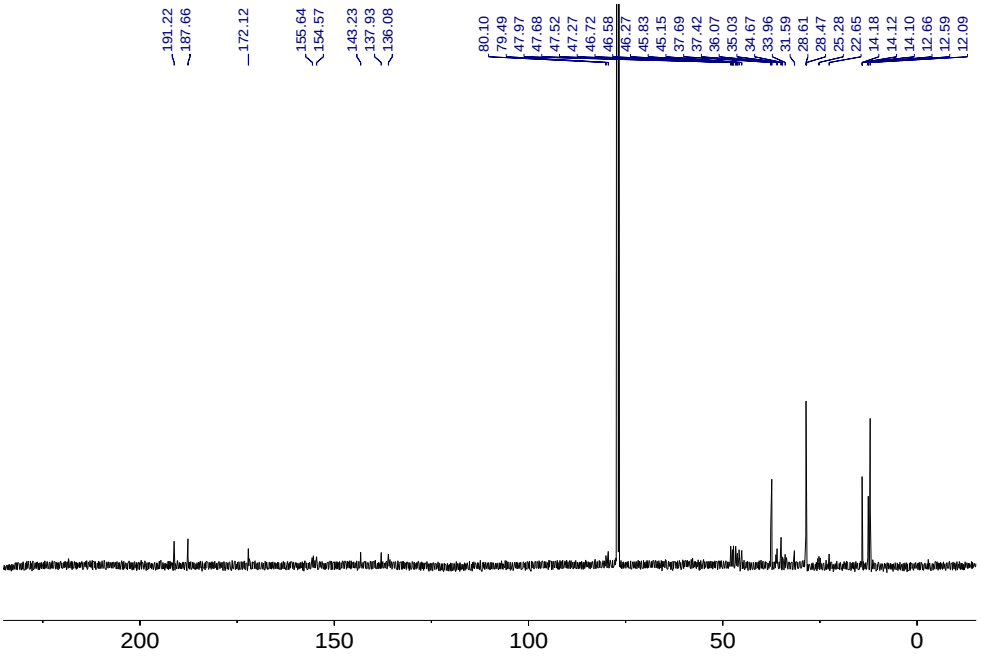


Figure S21. ^{13}C NMR spectrum of HQ-NN-Boc (**7**) in $CDCl_3$

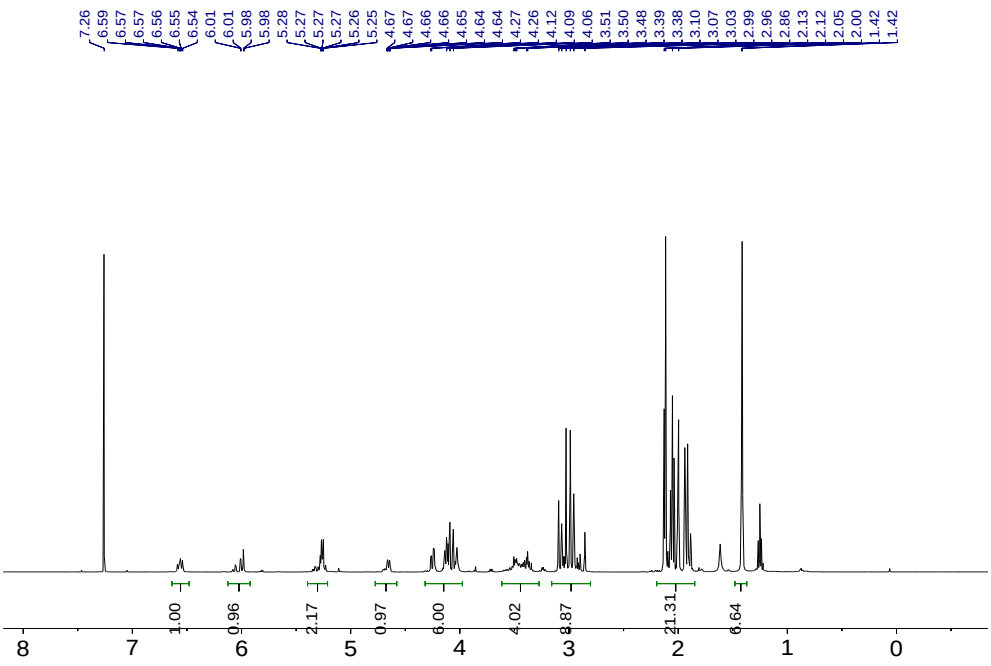


Figure S22. ^{1}H NMR spectrum of HQ-NN-AAM (**8**) in $CDCl_3$

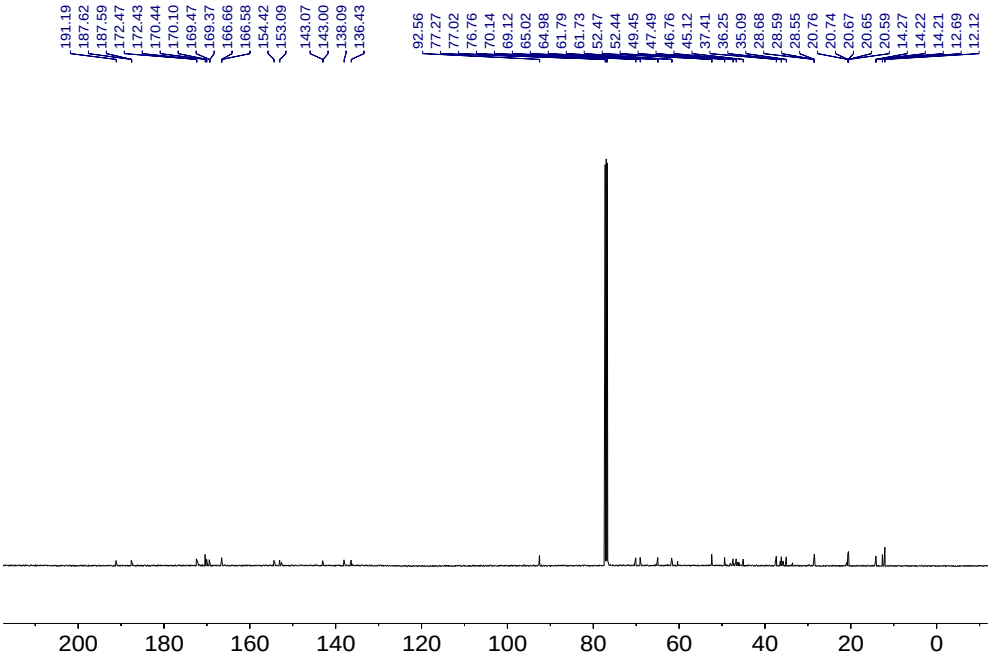


Figure S23. ^{13}C NMR spectrum of HQ-NN-AAM (**8**) in $CDCl_3$

Reference

1. R. M. Phillips, *Biochem Pharmacol*, 1999, **58**, 303-310.
2. H. Wang, R. Wang, K. Cai, H. He, Y. Liu, J. Yen, Z. Wang, M. Xu, Y. Sun, X. Zhou, Q. Yin, L. Tang, I. T. Dobrucki, L. W. Dobrucki, E. J. Chaney, S. A. Boppart, T. M. Fan, S. Lezmi, X. Chen, L. Yin and J. Cheng, *Nat. Chem. Biol.*, 2017, **13**, 415-424.
3. P. Liu, J. Xu, D. Yan, P. Zhang, F. Zeng, B. Li and S. Wu, *Chem. Commun.*, 2015, **51**, 9567-9570.