SUPPORTING INFORMATION

Classification: Biological Sciences; Immunology and Inflammation

Brief Treatment with a Highly Selective Immunoproteasome Inhibitor Promotes Long-Term Cardiac Allograft Acceptance in Mice

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Running Title: Allograft acceptance with an immunoproteasome inhibitor

Key words: allograft, dendritic cells, effector T cells, immunoproteasome, memory T cells, T cell exhaustion

Materials: Suc-LLVY-AMC, Ac-ANW-AMC, human c-20S (RBC) and i-20S (PBMC) were from Boston Biochem (Cambridge, MA). Enzymatic assays were recorded on a Molecular Devices SpectraMax M5 plate readers. Cell viability was determined with Cell-titer/glo^{TM} assay kit (PerkinElmer, Waltham, MA). Cell-based Proteasome-Glo^{TM} (G8660, Promega) was used to measure overall 20S chymotryptic-like β5 activity and its inhibition inside the cells.

IC50 Determination

We used a 96-well format for all compounds against hu i-20S and c-20S. Compounds (1 µL in DMSO and final concentration 100 – 0.0017 µM) in DMSO were spotted at the bottom of wells in black 96-well plates. Concentration of DMSO is constant at 1% in all wells. 100 µL reaction buffer containing 0.4 nM Hu i-20S and 15 µM Ac-ANW-AMC or 0.2 nM Hu c-20S and 25 µM suc-LLVY-AMC in 20 mM HEPES, 0.5 mM EDTA pH7.5 with 0.02% SDS and 0.1 mg/mL BSA was added to each well and plates were spun on a desktop plate centrifuge and then placed on an orbital shaker at room temperature for 1 minutes. The progress of reactions in each well was followed by the fluorescence of the hydrolyzed AMC at Ex 360nm and Em 460 nm for 1 - 2 hours. Z-VLR-AMC was used as β2c and β2i proteasomal activities. Ac-LLE-AMC was used as β1c substrate, and Ac-PAL-AMC as β1i substrate. PA28α was used as activator in β2 activity assays.

Table S1. IC50s of DPLG3, PKS2086 and PKS2032 against β1i, β2i, β1c and β2c of human immuno- and constitutive proteasomes

ID	IC50 (mM)			
	β1i	β1c	β2i	β2c
	Ac-PAL-AMC	Z-LLE-AMC	Z-VLR-AMC	Z-VLR-AMC
DPLG3	> 33.3	> 33.3	> 33.3	> 33.3
PKS2086	> 33.3	> 33.3	> 33.3	> 33.3
PKS2032	> 33.3	> 33.3	> 33.3	> 33.3

Cell-based Proteasome β5 Activity Assay

Karpas 1106P (80,000 cells / well) were plated and incubated with compound at indicated concentrations for 1 hr at 37ºC. The activity of the overall β5 activity including β5i and β5c in each was measured in situ after compound removal with Proteasome-Glo™ assay kit according to manufacturer's instructions. Luminescence was recorded on a SpectraMax M5 plate reader. Relative percentage of RLU was used to calculate the IC50s.

Cell Viability Assay

We used Karpas1106P B lymphoma cell line (Cat. No. 06072607, Aldrich) (1, 2), mouse bone marrow derived macrophages (3), peripheral blood mononuclear cells (4), HepG2 (2), human fibroblast cells. Cells were cultured at 37ºC in a humidified air/5%CO_2 atmosphere in medium supplemented with 10% fetal bovineserum, except for the medium for Karpas-1106P cells which contained 20% fetal bovine serum, and 100 units/ml penicillin/100 μg/ml streptomycin in RPMI

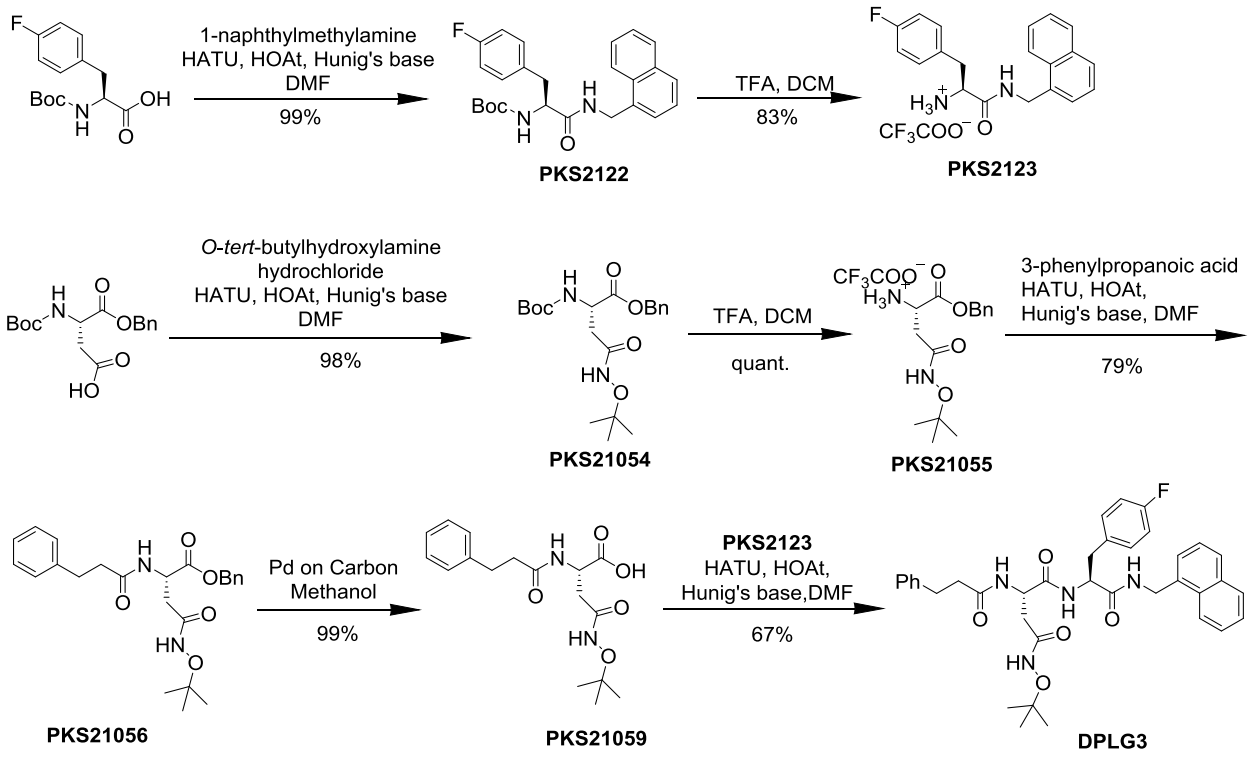
1640 medium. Karpas 1106P was used at 80,000 cells / well, mouse bone marrow cells at 100,000 cells / well, PBMCs at 100,000 cells / well, and HepG2 at 12,000 cells / well. Cells plated in a 96-well plate were treated with compounds at indicated concentrations for 72 hours at 37^{o}C in a tissue culture incubator with 5% CO_2. Viable cells were counted using Cell-titer/glo^{TM} assay kit. EC50s were calculated using PRISM (Graphpad).

Syntheses

Materials: All commercially available materials for synthesis were purchased from Bachem, Sigma, or AnaSpec and used as received, unless otherwise stated. All non-aqueous reactions were performed under Argon.

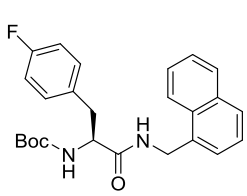
Spectroscopies: ^{1}H- and ^{13}C- NMR spectra were acquired on a Bruker DRX-500 spectrometer. Chemical shifts δ are expressed in parts per million. NMR data are reported as following: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, br = broad), coupling constant, and integration). Solvent resonance was used as an internal standard for ^{1}H and ^{13}C spectra (chloroform-*d*, ^{1}H: 7.26; ^{13}C: 77.16 ppm; DMSO-d_6, ^{1}H: 2.5 ppm; ^{13}C: 39.5 ppm). Hexafluorobenzene was used as internal standard for ^{19}F NMR. Low Resolution Mass spectra (LRMS) were obtained on a Waters Acquity UPLC-MS or a Perkin Elmer SCIEX API 100 mass spectrometer and High Resolution Mass Spectra (HRMS) were obtained on a Waters LCT Premier/XE.

We followed a reported procedure for HATU mediated amide bond formation, EDC mediated amide bond formation, boc-deprotection and O-debenzylation to synthesize compounds (DPLG-3, PKS2032, PKS2086) (2). All final compounds are > 95% pure.

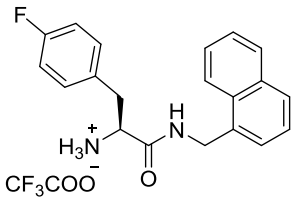


Supplementary Scheme 1. Synthetic route of DPLG3.

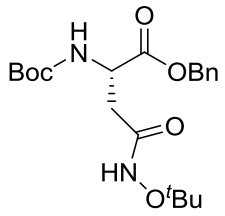
Analytic data of compounds.



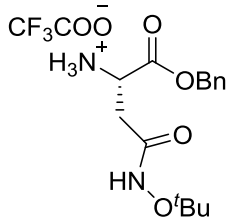
PKS2122: The title compound was prepared following the general procedure for HATU mediated coupling of Boc-4-F-Phe-OH (2.00g, 7.06 mmol) and 1-naphthylmethylamine (1.17 mL, 7.77 mmol). After completion of reaction (1 h), 100 mL water was added to the reaction mixture. A precipitate was formed. The mixture was stirred for 15 minutes and filtered. The precipitate was washed with water and dried to give 2.96 g (99%) product. The product was pure enough to use for the next step of synthesis without further purification. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.47 (t, *J* = 5.8 Hz, 1H), 8.03 (dd, *J* = 6.3, 3.4 Hz, 1H), 7.94 (dd, *J* = 6.2, 3.4 Hz, 1H), 7.84 (d, *J* = 8.1 Hz, 1H), 7.55 – 7.52 (m, 2H), 7.44 – 7.41 (m, 1H), 7.38 – 7.36 (m, 1H), 7.28 – 7.25 (m, 2H), 7.07 – 7.00 (m, 3H), 4.74 (d, *J* = 5.6 Hz, 2H), 4.25 – 4.15 (m, 1H), 2.93 (dd, *J* = 13.6, 5.1 Hz, 1H), 2.77 (dd, *J* = 13.6, 10.0 Hz, 1H), 1.30 (s, 9H).



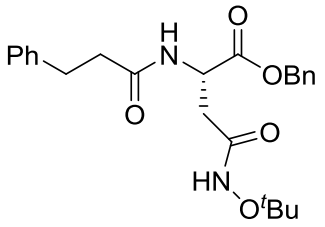
PKS2123: The title compound was prepared by following the general procedure for Boc-deprotection of PKS2122 (2.96 g, 7.00 mmol). The Crude was triturated with diethyl ether and filtered to give product as white solid (2.54 g, 83%). The product was pure enough to use for the next step of synthesis without further purification. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.91 – 8.88 (m, 1H), 8.30 (bs, 3H), 7.98 – 7.94 (m, 2H), 7.89 (d, *J* = 8.2 Hz, 1H), 7.58 – 7.55 (m, 2H), 7.44 (dd, *J* = 8.2, 7.0 Hz, 1H), 7.28 (d, *J* = 7.0 Hz, 1H), 7.22 – 7.19 (m, 2H), 7.09 – 7.06 (m, 2H), 4.81 (dd, *J* = 15.1, 5.8 Hz, 1H), 4.69 (dd, *J* = 15.1, 5.1 Hz, 1H), 4.04 – 4.01 (m, 1H), 3.06 – 2.98 (m, 2H).



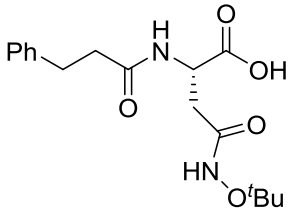
PKS21054: The title compound was synthesized by following the general protocol for HATU mediated coupling of Boc-Asp-OBn (2.00 g, 6.19 mmol) with *O-tert*-butyl hydroxylamine hydrochloride (855.2 mg, 6.81 mmol). After completion of reaction, water was added. Mixture was extracted with ethyl acetate twice. Combined organic layer was washed with saturated brine and dried over anhydrous sodium sulfate. Organic layer was evaporated to give product as colorless paste (2.40 g, 98%). The crude was used in next step without further purification. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.33 (s, 1H), 7.43 – 7.27 (m, 5H), 7.21 (d, J = 8.3 Hz, 1H), 5.11 (s, 2H), 4.43 – 4.36 (m, 1H), 2.55 (dd, J = 14.8, 5.9 Hz, 1H), 2.40 (dd, J = 14.8, 8.0 Hz, 1H), 1.36 (s, 9H), 1.13 (s, 9H).



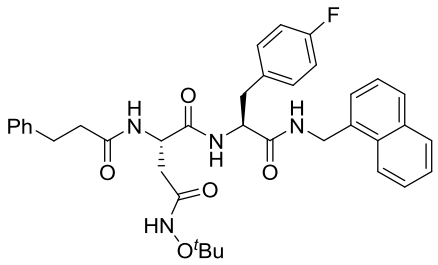
PKS21055: The title compound was synthesized by following the general procedure for boc-deprotection of PKS21054 (2.40 g, 6.08 mmol). Crude was dried under vacuum to give colorless paste (2.48 g, quant.). Product was used in next step without further purification. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.64 (s, 1H), 8.36 (bs, 3H), 7.44 – 7.34 (m, 5H), 5.23 – 5.19 (m, 2H), 4.47 – 4.39 (m, 1H), 2.71 (d, J = 5.3 Hz, 2H), 1.13 (s, 9H).



PKS21056: The title compound was synthesized by following the general procedure for HATU mediated coupling of 3-phenylpropanoic acid (991.1 mg, 6.60 mmol) with PKS21055 (2.45 g, 6.00 mmol). After completion of reaction, water was added. A white precipitate was formed. White precipitate was filtered, washed with water and dried to give product (2.02 g, 79%). Product was used in next step without further purification. Complex NMR due to presence of 90:10 rotamers. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.36 (s, 1H), 10.15 (s, 0.1H), 8.37 (d, J = 7.8 Hz, 0.9H), 8.30 (d, J = 7.7 Hz, 0.1H), 7.38 - 7.31 (m, 5H), 7.27 - 7.24 (m, 2H), 7.20 – 7.14 (m, 3H), 5.10 (s, 2H), 4.74 - 4.71 (m, 0.1H), 4.67 - 4.62 (m, 0.9H), 2.77 (t, J = 7.9 Hz, 2H), 2.57 (dd, J = 15.0, 6.2 Hz, 1H), 2.46 – 2.37 (m, 3H), 1.12 (s, 9H).

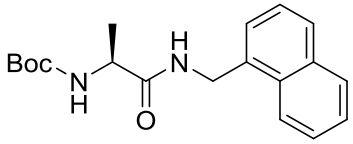


PKS21059: The title compound was synthesized by following the general procedure for *O*-debenzylation of PKS21056 (1.98 g, 4.64 mmol). Product (1.55 g, 99%) was isolated as white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 12.61 (s, 1H), 10.33 (s, 1H), 8.16 (d, J = 8.0 Hz, 1H), 7.29 – 7.23 (m, 2H), 7.22 – 7.14 (m, 3H), 4.64 – 4.46 (m, 1H), 2.81 – 2.76 (m, 2H), 2.54 – 2.46 (m, 1H), 2.43 – 2.37 (m, 2H), 2.35 (dd, J = 14.8, 7.5 Hz, 1H), 1.13 (s, 9H).

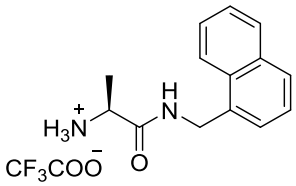


DPLG3: The title compound was prepared following the general procedure for HATU mediated coupling of PKS21059 (1.35 g, 4.00 mmol) and PKS2123 (1.92 g, 4.40 mmol). After completion of reaction, 100 mL water was added. A white precipitate was formed. Precipitate was filtered and washed with ethanol. The precipitate was triturated with methanol and filtered. Precipitate was dried to give 1.73 g (67%) pure product as white solid. Further purification for in vivo testing were carried out on a HPLC and final product was lyophilized to give white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.33 (s, 1H), 8.53 (t, J = 5.8 Hz, 1H), 8.11 – 8.04 (m, 3H), 7.97 – 7.91 (m, 1H), 7.84 (d, J = 8.1 Hz, 1H), 7.59 – 7.50 (m, 2H), 7.43 (dd, J = 8.1, 7.1 Hz, 1H), 7.36 (d, J = 7.0 Hz, 1H), 7.29 – 7.23 (m, 2H), 7.22 – 7.13 (m, 5H), 7.03 – 6.95 (m, 2H), 4.76 (dd, J = 15.3, 5.9 Hz, 1H), 4.70 (dd, J = 15.3, 5.7 Hz, 1H), 4.63 – 4.54 (m, 1H), 4.51 – 4.43 (m, 1H), 3.04 (dd, J = 13.8, 5.0 Hz, 1H), 2.82 (dd, J = 13.8, 9.2 Hz, 1H), 2.78 – 2.72 (m, 2H), 2.46 (dd, J = 14.9, 6.4 Hz, 1H), 2.39 – 2.32 (m, 2H), 2.27 (dd, J = 14.9, 7.8 Hz, 1H), 1.11 (s, 9H). ^{13}C NMR (126 MHz, DMSO-d_6) □ 171.38, 170.70, 170.45, 167.61, 160.93 (d, J = 242.0 Hz), 141.26, 134.21, 133.83 (d, J = 3.3 Hz), 133.24 , 130.98 (d, J = 8.2 Hz), 130.83, 128.48, 128.32, 128.11, 127.50, 126.22, 125.88, 125.77, 125.42, 125.37, 123.45, 114.71 (d, J = 21.1 Hz), 80.58, 54.29, 49.63, 40.23, 36.80, 36.45, 34.65,

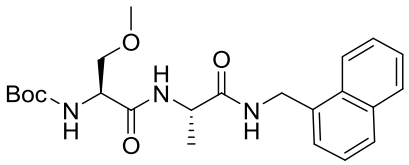
30.97, 26.26. ^{19}F NMR (471 MHz, DMSO-d_6) δ -119.24, -119.33 (m). HRMS calc. for C37H41FN4O5 $[M+H]^+$: 641.3139. Found: 641.3134.



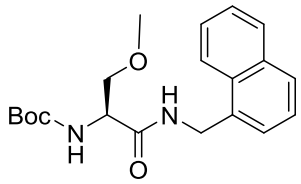
PKS3005: Boc-Ala-OSu (286 mg, 1.0 mmol) and 1-naphthylmethylamine (160 µL, 1.1 mmol) were dissolved in dichloromethane (10 mL). The solution was cooled to 0 °C and triethylamine (100 µL) was added. Reaction mixture was allowed to warm to room temperature slowly and stirred at room temperature. After completion of reaction, dichloromethane was evaporated and crude was suspended in water. Water layer was extracted twice with ethyl acetate. The combined organic layer was washed with aq. $NaHCO_3$, water, 1N HCl and brine. The organic layer was dried over anhydrous sodium sulfate and evaporated to give product (320 m, 97%), which was used in next step without further purification. ^{1}H NMR (500 MHz, Chloroform-*d*) δ 7.96 (d, *J* = 8.1 Hz, 1H), 7.87 (dd, *J* = 7.5, 1.7 Hz, 1H), 7.80 (dd, *J* = 6.9, 2.5 Hz, 1H), 7.59 – 7.48 (m, 2H), 7.44 – 7.40 (m, 2H), 6.44 (s, 1H), 4.96 – 4.88 (m, 3H), 4.18 – 4.15 (m, 1H), 1.37 (d, *J* = 7.1 Hz, 3H), 1.34 (s, 9H).



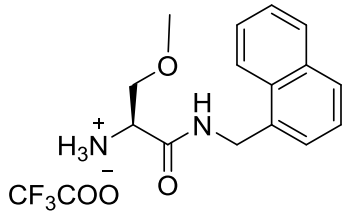
PKS2026: The title compound was synthesized by following the general procedure for boc-deprotection of PKS3005 (158 mg, 0.48 mmol). After completion of reaction (3 h), dichloromethane and excess TFA were evaporated. The crude product was dried under vacuum to give product (164 mg, quant.). ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.91 (t, *J* = 5.7 Hz, 1H), 8.15 (bs, 3H), 8.07 – 8.04 (m, 1H), 7.99 – 7.97 (m, 1H), 7.91 – 7.87 (m, 1H), 7.60 – 7.55 (m, 2H), 7.51 – 7.48 (m, 2H), 4.85 (dd, *J* = 15.2, 5.7 Hz, 1H), 4.78 (dd, *J* = 15.2, 5.5 Hz, 1H), 3.91 – 3.86 (m, 1H), 1.37 (d, *J* = 7.0 Hz, 3H).



PKS2032: PKS2026 (0.24 mmol) was dissolved in 3 mL dimethylformamide and basified with *N*-methylmorpholine. Boc-Ser(OMe)-OH (96 mg, 0.24 mmol) was added to the solution. The mixture was cooled to 0 °C and dipyrrolidino(*N*-succinimidyloxy)carbenium hexafluorophosphate (103 mg, 0.25 mmol) were added in one portion. The reaction mixture was allowed to warm to room temperature slowly and stirred at room temperature overnight. The reaction mixture was diluted with water and extracted twice with ethyl acetate. The organic layer was dried over anhydrous Na_2SO_4 and evaporated. The product was purified by silica gel column chromatography (eluent: ethylacetate and hexane) to give 95 mg (92%) of product. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.29 (t, *J* = 5.8 Hz, 1H), 8.11 (d, *J* = 7.4 Hz, 1H), 8.03 – 8.01 (m, 1H), 7.96 – 7.94 (m, 1H), 7.85 (d, *J* = 8.0 Hz, 1H), 7.57 – 7.53 (m, 2H), 7.48 – 7.45 (m, 1H), 7.42 (dd, J = 7.0, 1.4 Hz, 1H), 6.95 (d, *J* = 8.0 Hz, 1H), 4.75 (d, *J* = 5.6 Hz, 2H), 4.36 – 4.31 (m, 1H), 4.19 – 4.15 (m, 1H), 3.47 – 3.39 (m, 2H), 3.15 (s, 3H), 1.37 (s, 9H), 1.25 (d, *J* = 7.1 Hz, 3H).

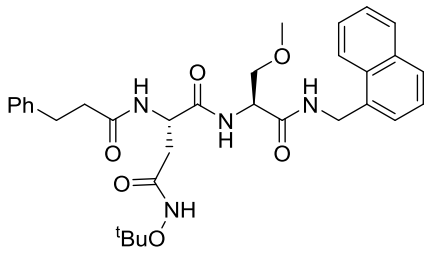


PKS2120: The title compound was prepared following the general procedure for HATU coupling. Reaction was carried using Boc-β-methoxyalanine dicyclohexylamine (1.202 g, 3.0 mmol) and 1-naphthylmethylamine (484 mL, 3.3 mmol). After completion of reaction 150 mL water was added to reaction mixture and extracted twice with ethyl acetate (2 X 150 mL). The combined organic layer was washed with brine, dried over anhydrous sodium sulfate and evaporated. The crude product was purified by silica gel column chromatography using a gradient of 20% - 40% ethyl acetate-hexane to give 1.05 g (98%) of pure product. ^{1}H NMR (500 MHz, Chloroform-*d*) δ 7.96 (d, *J* = 8.4 Hz, 1H), 7.87 – 7.85 (m, 1H), 7.79 (dd, *J* = 7.2, 2.4 Hz, 1H), 7.54 – 7.48 (m, 2H), 7.44 – 7.39 (m, 2H), 6.73 (m, 1H), 5.40 (m, 1H), 4.91 (m, 2H), 4.27 (m, 1H), 3.82 (dd, *J* = 9.0, 4.1 Hz, 1H), 3.47 (dd, *J* = 9.0, 6.2 Hz, 1H), 3.28 (s, 3H), 1.37 (s, 9H).

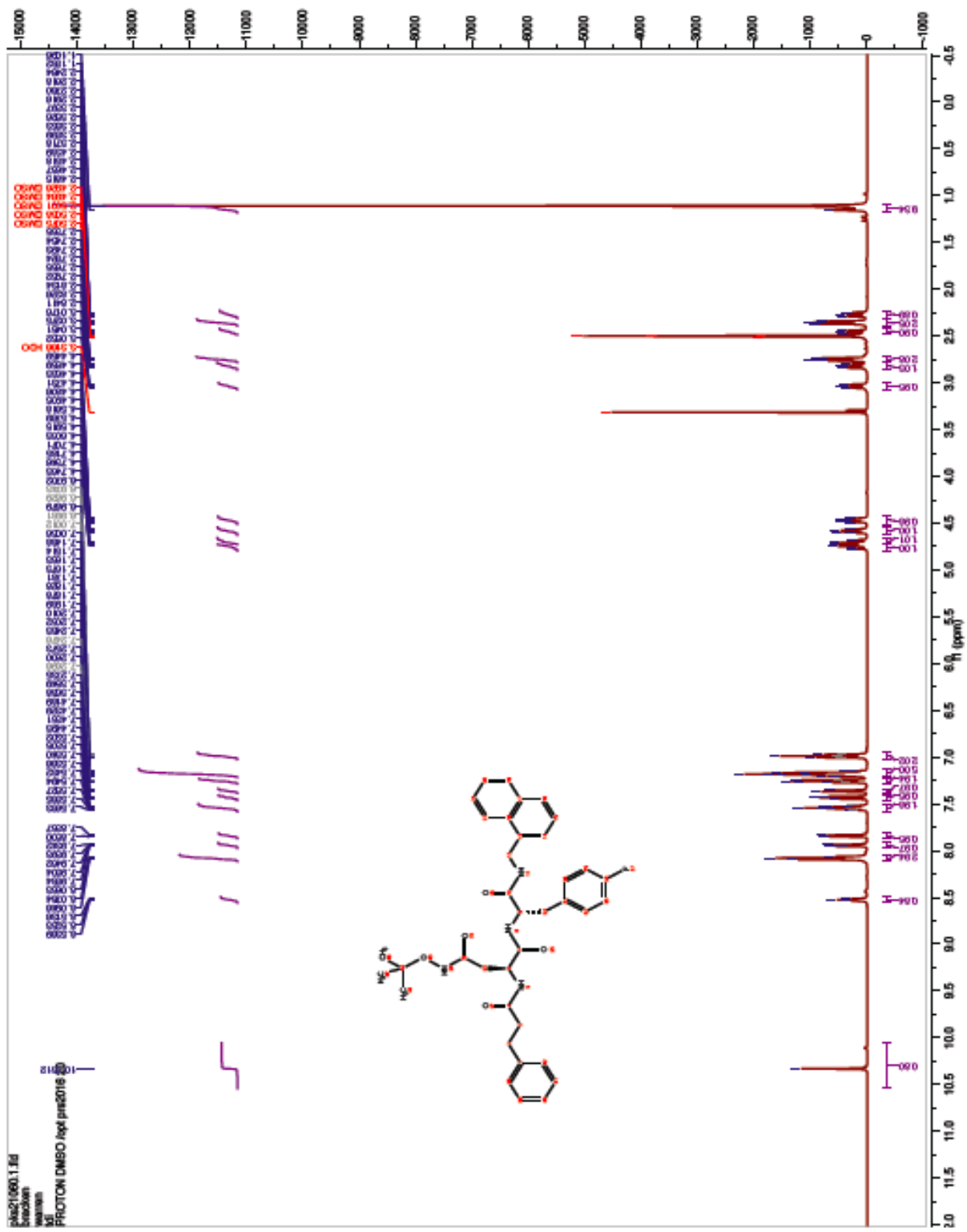


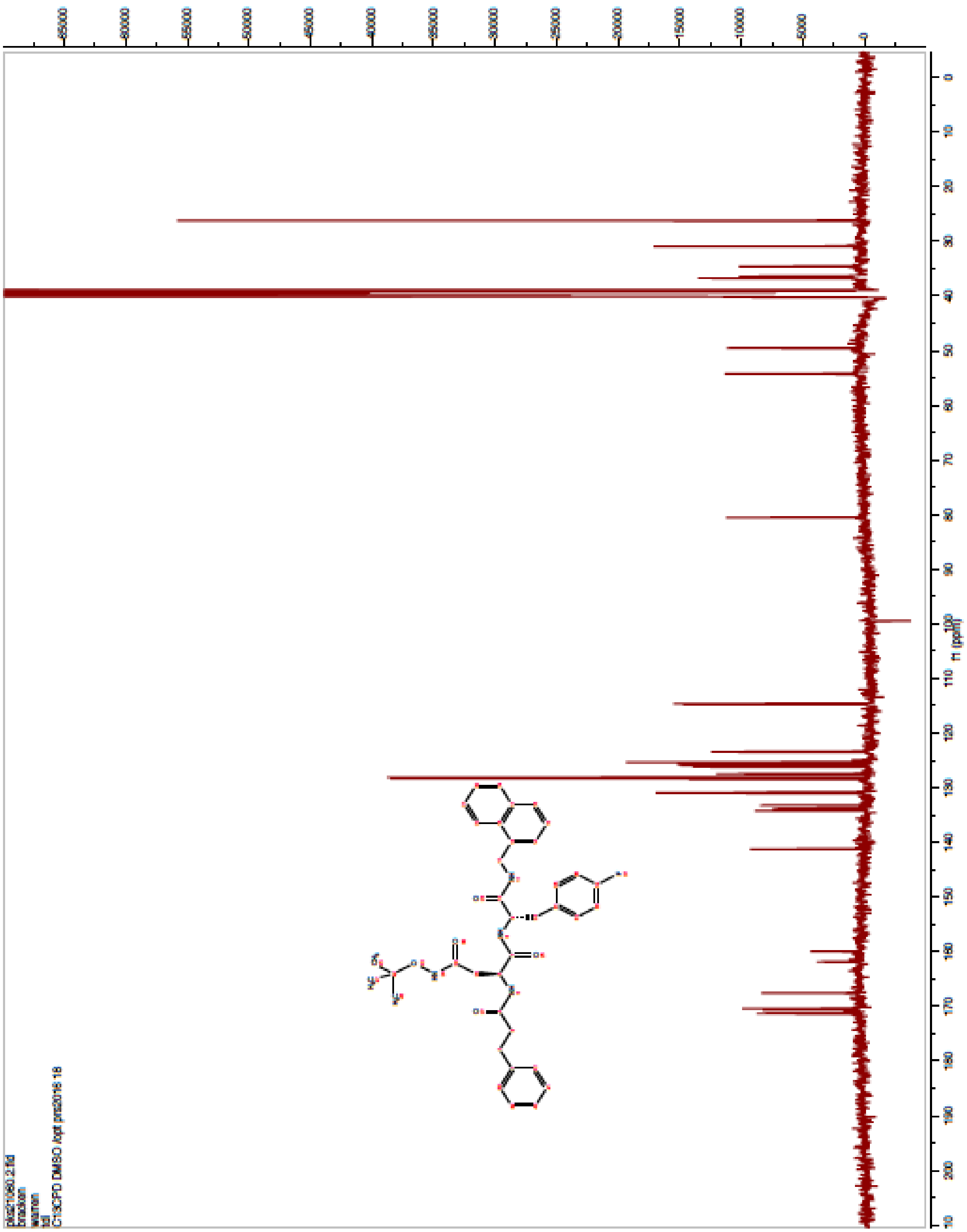
PKS2082: The title compound was synthesized by following the general procedure for boc deprotection of PKS2120 (72 mg, 0.02 mmol). After completion of reaction (4 h), dichloromethane and excess TFA were evaporated and dried under high vacuum. The paste was soluble in diethyl ether. Diethyl ether solution was extracted with water. Water layer was frozen and lyophilized to give product (67 mg, 90%). ^{1}H NMR (500 MHz, DMSO-d_6) □ 8.94 (t, *J* = 5.5 Hz, 1H), 8.20 (bs,

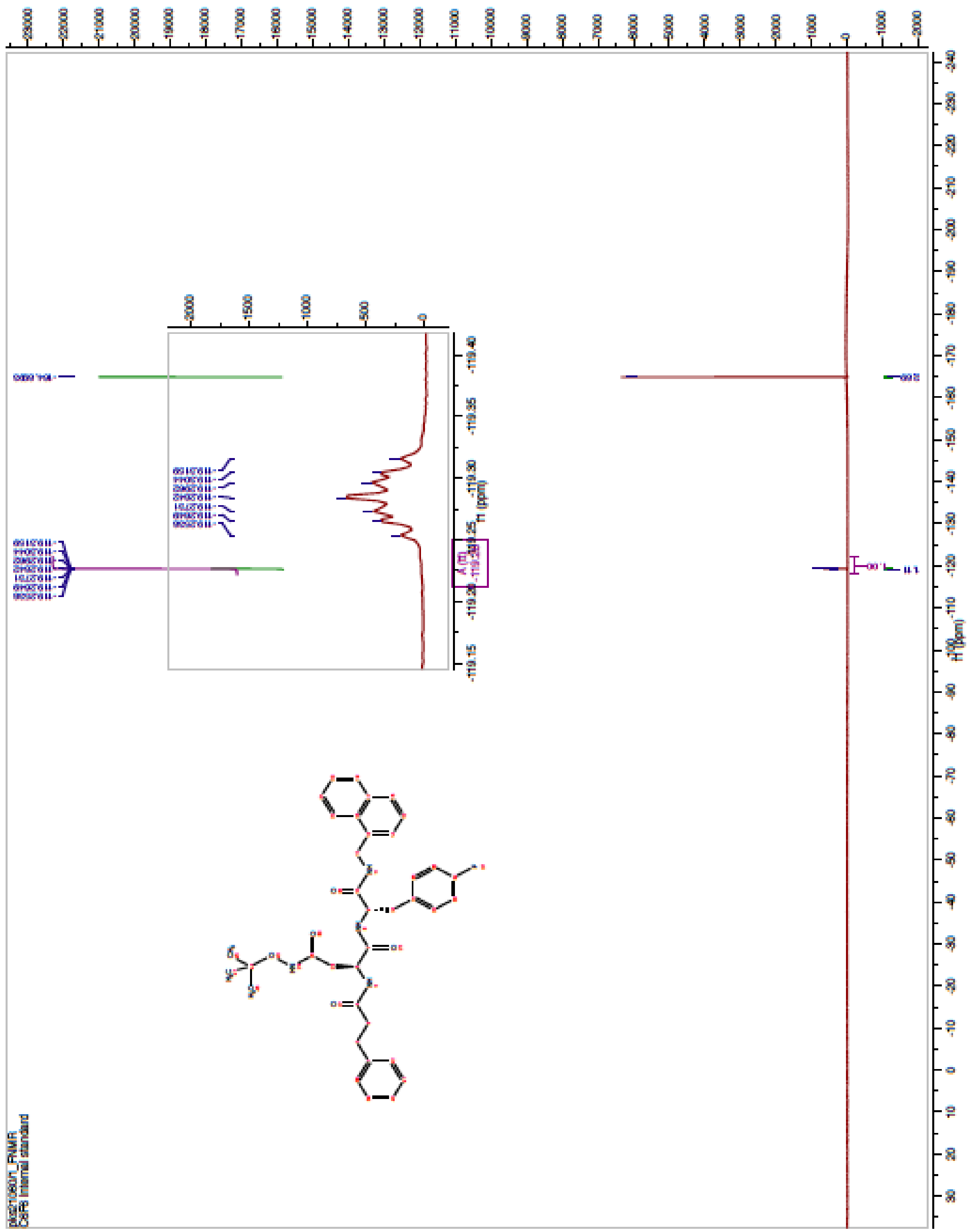
3H), 8.04 – 8.02 (m, 1H), 7.98 – 7.95 (m, 1H), 7.89 – 7.87 (m, 1H), 7.59 – 7.54 (m, 2H), 7.51 – 7.48 (m, 2H), 4.85 (dd, *J* = 15.2, 5.7 Hz, 1H), 4.77 (dd, *J* = 15.2, 5.4 Hz, 1H), 4.05 – 4.03 (m, 1H), 3.70 – 3.63 (m, 2H), 3.28 (s, 3H).



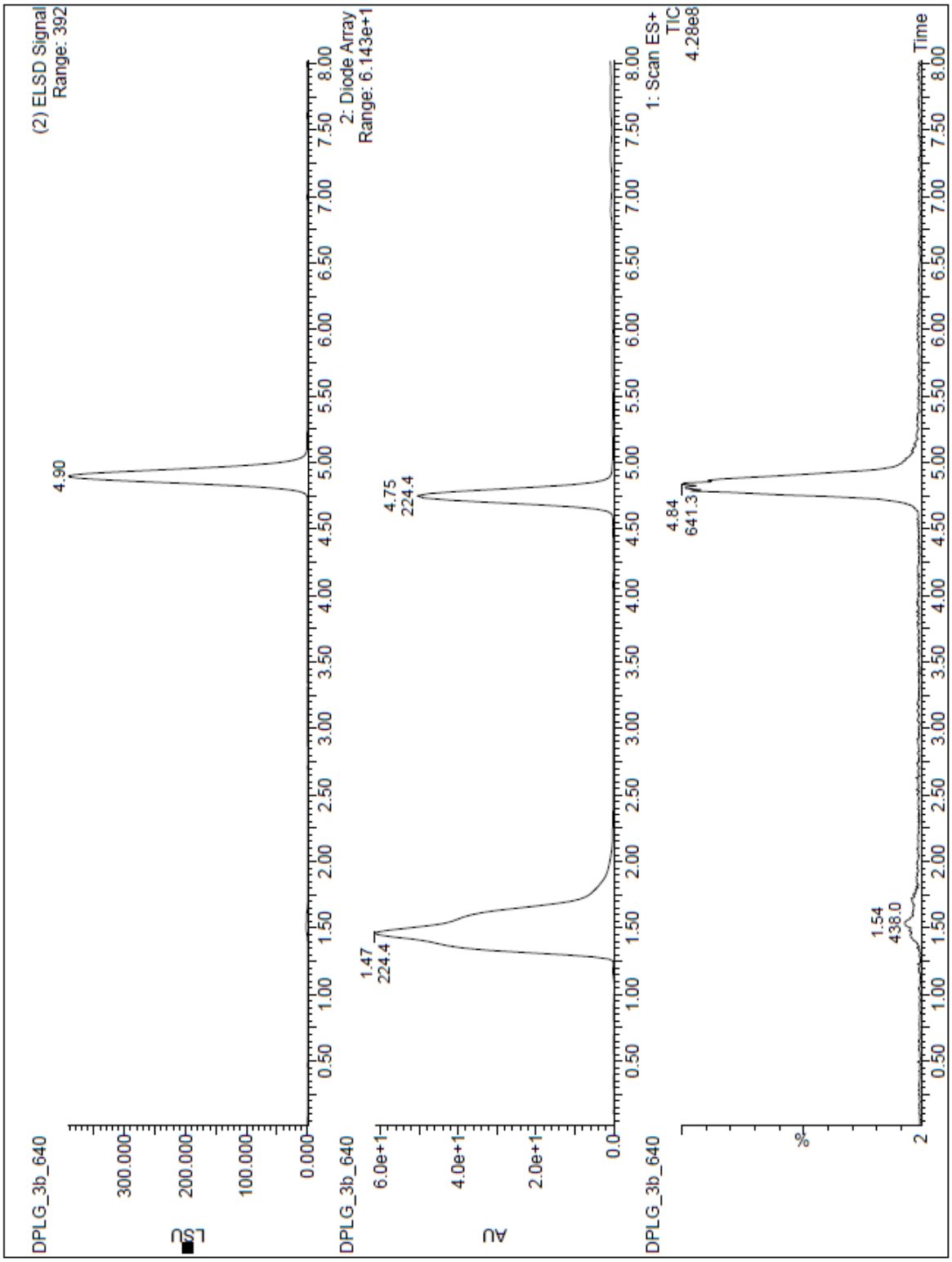
PKS2086: The title compound was prepared following the general procedure for HATU mediated coupling of PKS21059 (11.0 mg, 0.033 mmol) and PKS2082 (11.0 mg, 0.03 mmol). The product was purified by HPLC and final product was lyophilized to give white solid (yield 15.3 mg, 88%). ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.39 (s, 1H), 8.58 (t, *J* = 5.8 Hz, 1H), 8.21 (d, *J* = 8.0 Hz, 1H), 8.11 (d, *J* = 7.7 Hz, 1H), 8.09 – 8.07 (m, 1H), 7.95 – 7.93 (m, 1H), 7.84 – 7.82 (m, 1H), 7.56 – 7.52 (m, 2H), 7.48 – 7.43 (m, 2H), 7.28 – 7.25 (m, 2H), 7.20 – 7.16 (m, 3H), 4.78 (dd, *J* = 15.4, 5.9 Hz, 1H), 4.71 (*J* = 15.4, 5.7 Hz, 1H), 4.68 – 4.64 (m, 1H), 4.45 – 4.42 (m, 1H), 3.60 (dd, *J* = 9.7, 5.9 Hz, 1H), 3.51 (dd, *J* = 9.7, 4.7 Hz, 1H), 3.24 (s, 3H), 2.80 – 2.77 (m, 2H), 2.53 – 2.49 (m, 1H), 2.43 – 2.39 (m, 2H), 2.34 (dd, *J* = 14.8, 7.6 Hz, 1H), 1.11 (s, 9H). ^{13}C NMR (126 MHz, DMSO) δ 171.94, 171.38, 169.66, 168.05, 141.70, 134.69, 133.65, 131.22, 128.89, 128.76, 128.58, 127.85, 126.59, 126.32, 126.18, 125.83, 125.54, 123.87, 81.02, 72.27, 58.74, 53.69, 50.11, 40.72, 37.24, 35.30, 31.45, 26.71. HRMS calc. for C32H40N4O6 $[M+H]^+$: 577.3026. Found: 577.3005.







DPLG-3 Purity was determined by HPLC.



Supplemental figure legends

Figure S1. Docking study of DPLG3 in the active sites of the β5i and β5c. Ser27 in β5i of human and mouse immunoproteasomes is not conserved in the β5c of human and mouse constitutive proteasomes.

Figure S2. LMP7 inhibition prolonged heart allograft survival and exhibited a differential effect on effector T cells and Tregs in vivo. (A) Representative examples of cardiac allograft histology at day 7 post transplant show significantly lower grades of acute cellular rejection (H&E stain, original magnification x10) in the DPLG3 treated recipients compared to vehicle treated (n = 4 mice/group). (B) Representative dot plots of splenocytes analysis of C57BL/6 mice recipients of BALB/c hearts treated with DPLG3 (25 mg/Kg daily for 7 days) or vehicle. Data shows significant decrease in CD4 and CD8 effector T cells in DPLG3 treated compared to vehicle treated mice (n=6 mice/group). (C) Representative dot plots shows an increase in Tregs in splenocytes of DPLG3 treated recipients compared to vehicle treated. Bar graph shows a significant increase in the ratio of Tregs to Teff in the spleen of DPLG3 treated recipients compared to vehicle treated (n = 6, * $p < 0.05$). (D) Graph shows reduced proliferation of splenocytes from DPLG3 treated recipients upon stimulation with irradiated donor cells compared to vehicle treated mice ($p<0.05$).

Figure S1.

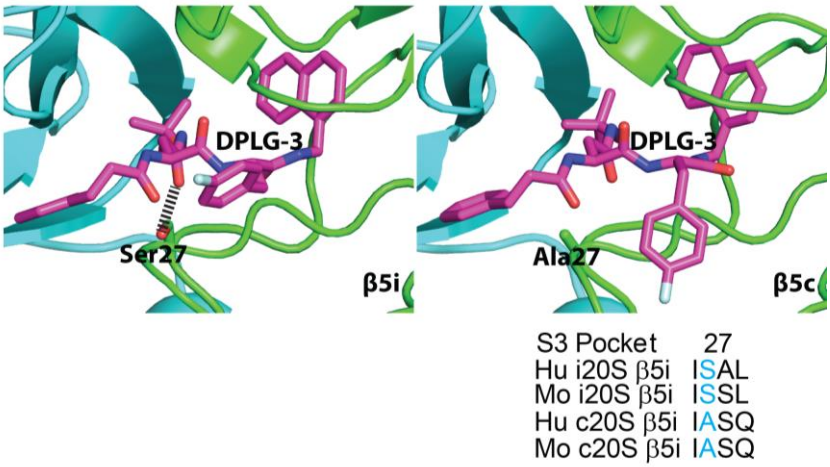
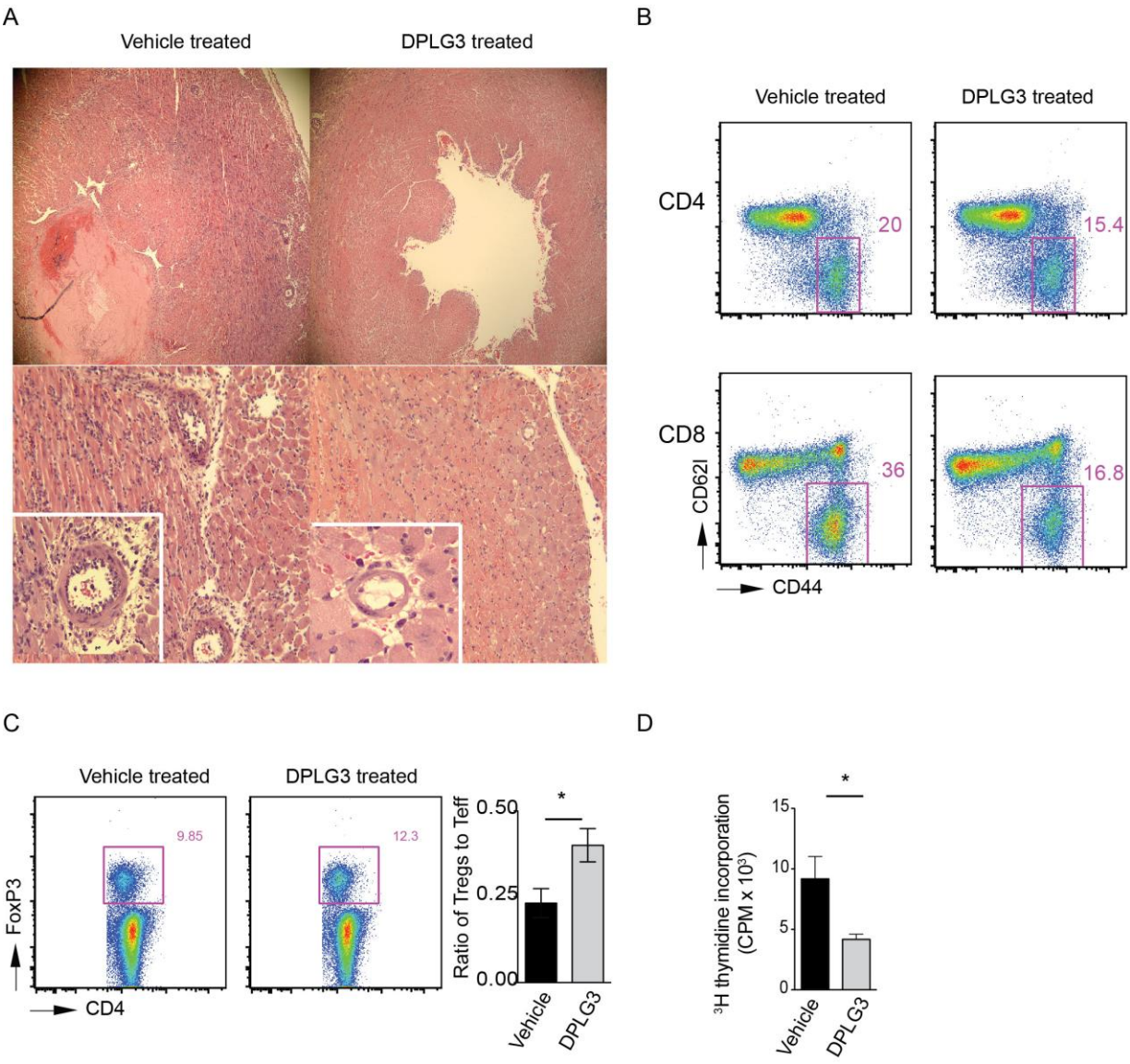

DPLG-3
Ser27
β5i
DPLG-3
Ala27
β5c
S3 Pocket 27
Hu i20S β5i ISAL
Mo i20S β5i ISSL
Hu c20S β5i IASQ
Mo c20S β5i IASQ

Figure S2.


A
Vehicle treated
DPLG3 treated
B
Vehicle treated
DPLG3 treated
CD4
20
15.4
CD8
CD62l
36
16.8
CD44
C
Vehicle treated
DPLG3 treated
FoxP3
9.85
12.3
CD4
Ratio of Tregs to Teff
0.50
0.25
0.00
*
Vehicle
DPLG3
D
^{3}H thymidine incorporation (CPM x 10^3)
15
10
5
0
*
Vehicle
DPLG3

Reference:

1. Blackburn C*, et al.* (2010) Optimization of a series of dipeptides with a P3 threonine residue as non-covalent inhibitors of the chymotrypsin-like activity of the human 20S proteasome. *Bioorg Med Chem Lett* 20(22):6581-6586.
2. Singh PK*, et al.* (2016) Immunoproteasome β5i-Selective Dipeptidomimetic Inhibitors. *ChemMedChem*:DOI:10.1002/cmdc.201600384.
3. Bryk R*, et al.* (2008) Selective killing of nonreplicating mycobacteria. *Cell Host Microbe* 3(3):137-145.
4. Vogt G & Nathan C (2011) In vitro differentiation of human macrophages with enhanced antimycobacterial activity. *J Clin Invest* 121(10):3889-3901.