The Modular Synthesis of Pantetheine and Phosphopantetheine

Alexander L. Mandel, James J. LaClair, Michael D. Burkart

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Experimental Data

General. All reactions were carried out under argon atmosphere in dry solvents with oven dried glassware unless otherwise noted. NMR spectra were taken on Varian 300MHz or 400MHz NMR machines and standardized to the NMR solvent except for ^{31}P NMR, where signals were standardized to 85% H_3PO_4 . Chemical shifts are reported in parts per million relative to tetramethylsilane except in the aforementioned ^{31}P spectra. Silica gel chromatography was carried out with Silicycle 60 Angstrom 230-400 mesh. TLC prep plate purification was performed with EMD Silica Gel 60 F₂₅₄ pre coated plates.

Preparation of Protected Pantoic Acid (7)

[(2*R*, 4*R*)-2-(4-Methoxy-phenyl)-5,5-dimethyl-[1,3]dioxane-4-yl]methanol (6) – See literature preparation by Mukaiyama¹.

[(2*R*, 4*R*)-2-(4-Methoxy-phenyl)-5,5-dimethyl-[1,3]dioxane-4-carboxylic acid (7) – Swern oxidation was carried out on **6** (2.56 g, 18.44 mmol) as per Mukaiyama's procedure¹ for the preparation of (2*R*, 4*R*)-2-(4-Methoxy-phenyl)-5,5-dimethyl-1,3-dioxane-4-carbaldehyde and the resultant oil was purified by silica gel chromatography (6:1 to 2:1 Hexanes/EtOAc) to yield the product as a clear oil that crystallized under high vacuum (2.07 g, 81%). The product was recrystallized in hexane/pentane (1:1, 30 mL), to afford long white needles (1.51 g, 60% overall)

The product of the preceding reaction (199 mg, 0.796 mmol) was dissolved in acetone/ CH_2Cl_2 (3:1, 12 mL). A fresh solution of $\text{NaH}_2\text{PO}_4\cdot\text{H}_2\text{O}$ (1.10 g, 7.96 mmol) and NaClO_2 (360 mg, 3.98 mmol) was prepared in 3mL of water, and it was added dropwise to the stirring solution of the aldehyde. After 8 min, the volatile solvents were evaporated *in vacuo*, leaving flakes of white solid in the aqueous remnants. Solid NaHSO_3 was added to remove the bleach, and the solid was filtered out to give **7** (180mg, 85%). The product was pure by NMR, but it could be recrystallized in THF/hexanes (1:4) to give clear prisms (123mg, 85% recrystallization yield). The product should be stored cold, as it will spontaneously deprotect and reform pantolactone,

¹ Shiina, I.; Shibata, J.; Ibuka, R.; Imai, Y.; Mukaiyama, T. *Bull. Chem. Soc. Jpn.* **2001**, 74, 113-122

and the scent of anisealdehyde will become marked in the residue. ^1H NMR (CDCl_3 , 300 MHz) δ 7.41 (d, $J = 9.0$ Hz, 2H), 6.90 (d, $J = 11.6$ Hz, 2H), 5.51 (s, 1H), 4.22 (s, 1H), 3.80 (s, 3H), 3.76 (d, $J = 11.7$ Hz, 1H), 3.66 (d, $J = 11.6$ Hz, 1H), 1.19 (s, 3H), 1.09 (s, 3H). ^{13}C NMR (CDCl_3 , 100 MHz) δ 169.5, 160.2, 129.3, 127.5, 113.8, 101.6, 82.9, 78.2, 55.3, 33.1, 21.6, 19.3.

Preparation of Pantetheine (3)

2-Tritylmercapto-ethylamine (9) – To cystamine·HCl **8** (1.50 g, 14.4 mmol) and trifluoroacetic acid (3.28 g, 2.22 mL, 28.7 mol) dissolved in CH₂Cl₂ with a drying tube, trityl chloride (4.20 g, 15.1 mmol) was added. The solution immediately turned a dark yellow color. After 30 minutes the reaction was quenched with 1 M NaOH (30 mL) turning the solution back to clear. The organic layer was diluted with CH₂Cl₂ (75 mL), and additional 1 M NaOH (20 mL) was added, and the aqueous layer was separated. The organic layer was then washed with brine (20 mL), dried over sodium sulfate, and concentrated *in vacuo* to give a yellow oil. The oil was purified by flash chromatography (1:4 to 1:1 MeOH/EtOAc) to give **9** (3.14 g, 63%) as a clear oil which solidified when left under vacuum overnight. ¹H NMR (400 MHz, CDCl₃) δ 7.41 (m, 6H), 7.27 (m, 6H), 7.20 (m, 3H), 2.57 (t, *J* = 8 Hz, 2H), 2.33 (t, *J* = 8 Hz, 2H).

3-(Fmoc-Amino)-N-(2-tritylsulfanyl-ethyl)-propionamide (10) – **9** (50 mg, 0.14 mmol), Fmoc-β-Alanine (65 mg, 0.21 mmol), EDC (67 mg, 0.35 mmol), and HOBT (56 mg, 0.42 mmol) were combined and dissolved in dry THF (10 mL). DIPEA (110 mg, 0.84 mmol) was added, and the reaction was allowed to stir for 4.5 hours. The reaction was quenched with 25% NH₄Cl_(aq) and diluted with diethyl ether (20 mL). The organic layer was washed with water (5 mL), brine (5 mL), dried over anhydrous Na₂SO₄, and evaporated *in vacuo*. The resultant oil was purified by column chromatography (1:1, hexanes:EtOAc) to yield **10** (81 mg, 90%) as a sticky white solid. ¹H NMR (300 MHz, CDCl₃) δ 7.73 (d, *J* = 7.5 Hz, 2H), 7.55 (d, *J* = 7.5 Hz, 2H), 7.45-7.15 (m, 19H), 5.46 (b, 2H), 4.33 (d, *J* = 7.2 Hz, 2H), 4.17 (t, *J* = 6.6 Hz, 1H), 3.42 (m, 2H), 3.06 (q, *J* = 6.3 Hz, 2H), 2.41 (t, *J* = 6.0 Hz, 2H), 2.30 (t, 2H). ¹³C NMR (100 MHz, CDCl₃) δ 170.9, 156.3, 144.3, 143.7, 141.1, 129.3-126.7 (multiple signals), 125.0, 119.8, 66.7, 47.3, 38.2, 35.9, 31.9. *m/z* found: 635.12 amu. [M+Na]⁺ calcd. C₃₉H₃₆O₃N₂SN⁺: 635.23 amu.

2-(4-Methoxy-phenyl)-5,5-dimethyl-[1,3]dioxane-4-carboxylic acid [2-(2-tritylsulfanyl-ethylcarbamoyl)-ethyl]-amide (12) – Compound **10** (58 mg, 0.090 mmol) was dissolved in DMF (3 mL), and piperidine was added (0.75 mL). The DMF and piperidine were evaporated under reduced pressure, and to the dry residue (crude **11**) EDC (36 mg, 0.19 mmol), HOBT (31 mg, 0.23 mmol), and **7** (20 mg, 0.075 mmol) were added. The flask was evacuated and filled with argon. The contents were dissolved in THF and DIPEA (59 mg, 0.080 mL, 0.46 mmol) was added. The reaction was allowed to stir overnight and it was quenched with 25% ammonium chloride and diluted with diethyl ether (15 mL). The organic layer was separated and washed with water (5 mL), brine (10 mL), dried over anhydrous Na₂SO₄, and evaporated *in vacuo*. The product was purified by silica gel chromatography (1:1 to 1:5 Hexanes/EtOAc) to give **12** (41 mg, 85%) as a clear film. It should be noted that in this form the product will slowly deprotect in chloroform to give *S*-trityl pantetheine. ¹H NMR (CDCl₃, 400 MHz) δ 7.40-7.37 (m, 6H), 7.28-7.26 (m, 6H), 7.21-7.18 (m, 5H), 6.97 (t, *J* ~ 8 Hz, 1H), 6.89 (d, *J* = 8.8 Hz, 2H), 5.76 (t, *J* ~ 8 Hz, 1H), 5.39 (s, 1H), 4.02 (s, 1H), 3.79 (s, 3H), 3.67 (d, *J* = 12 Hz, 1H), 3.60 (d, *J* = 12 Hz, 1H), 3.48 (d, *J* = 6 Hz, 1H), 3.45 (d, *J* = 6 Hz, 1H), 3.02 (8-plet, *J* = 6.4 Hz, 1H), 2.98 (8-plet, *J* = 6.0 Hz, 1H), 2.385 (t, *J* = 6.8 Hz, 1H), 2.378 (t, *J* = 6.4 Hz, 1H), 2.31 (t, *J* = 6.0 Hz, 2H), 1.05 (s, 3H), 1.04 (s, 3H). ¹³C NMR (CDCl₃, 100 MHz) δ 170.6, 169.5, 144.6, 130.1, 129.5-126.6 (multiple signals), 113.7, 101.2, 83.8, 78.4, 66.8, 55.3, 38.2, 35.9, 34.8, 33.0, 31.7, 21.8, 19.1. ¹H-COSY couplings δ 6.97-3.46, 5.76-3.00, 3.46-2.31, 3.00-2.38.

Pantetheine (3) – Compound **12** (13 mg, 0.020 mmol) was dissolved in MeOH (2 mL), and a 0.1 M solution of iodine in MeOH (2 mL) was added. After 20 minutes, zinc metal was added to remove the iodine. The solution was filtered through celite and evaporated *in vacuo*. The remaining residue was purified twice by silica gel chromatography (1:1 MeOH/EtOAc) to remove iodine salts from the product **3** (5mg, ~85%). ¹H NMR (D₂O, 400 MHz) δ 4.00 (s, 1H), 3.51-3.54 (m, 5H), 3.40 (d, *J* = 11.2 Hz, 1H), 2.87 (t, *J* = 6.0 Hz, 2H), 2.53 (t, *J* = 6.4 Hz, 2H), 0.94 (s, 3H), 0.90 (s, 3H). *m/z* found 577.22, calcd. C₂₂H₄₂O₈N₄S₂Na⁺: 577.23

Preparation of Protected Phosphopantoic Acid (17)

(R)-3-Benzoyloxy-4,4-dimethyl-dihydro-furan-2-one (13) – Silver oxide (3.54 g, 15.3 mmol) and benzyl bromide (1.4 g, 8.4 mmol) were added to a solution of D-Pantolactone (1.0 g, 7.7 mmol) in dry DMF (25 mL) at 0°C under nitrogen. The mixture was stirred at 0°C for 2 h, then warmed to r.t. and stirred for an additional 20 h. The solution was diluted with dichloromethane (100 mL) and filtered. The filtrate was concentrated *in vacuo*, diluted with ethyl acetate, and washed with 0.5 N HCl, water, and brine. The solvent was removed *in vacuo* and then excess benzyl alcohol was removed by co-evaporation with water under reduced pressure to give a crystalline solid. The product was recrystallized from hexanes to give **13** (1.46 g, 86%) ¹H NMR (300 MHz, CDCl₃) δ 7.30-7.36 (m, 5H), 5.02 (d, *J* = 12.0 Hz, 1H), 4.73 (d, *J* = 12.3 Hz, 1H), 3.97 (d, *J* = 9.0 Hz, 1H), 3.85 (d, *J* = 8.7 Hz, 1H), 3.71 (s, 1H), 1.12 (s, 3H), 1.08 (s, 3H). ¹³C NMR (100MHz, CDCl₃) δ 175.4, 137.2, 128.4, 127.98, 127.97, 80.4, 76.4, 40.3, 23.2, 19.3. Note: this product has been synthesized previously using benzyl chloride by a different method which required base²; however, the optical purity was reduced even with the mild base Cs₂CO₃. Optical purity is preserved in this procedure and can be confirmed by the generation of only two diastereomers in the proceeding step which vary only at the anomeric carbon.

(R)-3-Benzoyloxy-4,4-dimethyl-tetrahydro-furan-2-ol (14) – To a stirred solution of **13** (3.00 g, 13.6 mmol) in dichloromethane (50 mL) at -78°C, DIBAL-H (1 M in hexanes, 16.3 mL, 16.3 mmol) was added over 30 minutes. After 2 hours, the reaction was quenched slowly at first with 60mL of a 1:1 diethyl ether/1 M H₂SO₄ mixture. The reaction was then diluted with ethyl acetate (100 mL) and the organic layer was washed with 100 mL 1 M H₂SO₄, 10 mL of NaHCO₃(sat), 10 mL of water, and twice with 20 mL of brine. The organic phase was then dried with Na₂SO₄, and concentrated *in vacuo*. The crude oil was purified by flash chromatography (2:1 Hexanes/EtOAc to EtOAc) to yield **14** (2.85 g, 94%) as a clear oil that solidified to white clumps after it was removed from the freezer and disturbed. The product turned out to be an inseparable mixture of anomers in an approximate 2:3 ratio. ¹H NMR (CDCl₃, 400 MHz) δ 7.34-7.32 (m, 5H), 5.46 (m, 3/5H), 5.36 (d, *J* = 2.8 Hz, 2/5H), 4.70 (d, *J* = 12.0 Hz, 2/5H), 4.66 (d, *J* = 11.6 Hz, 3/5H), 4.61 (d, *J* = 11.2 Hz, 3/5H), 4.57 (d, *J* = 12.0 Hz, 2/5H), 3.98 (b, 3/5H), 3.81 (d, *J* = 8.4 Hz, 2/5H), 3.71 (d, *J* = 8.0 Hz, 3/5H), 3.63 (d, *J* = 8.4 Hz, 2/5H), 3.52 (d, *J* = 2.8 Hz, 2/5H), 3.46 (d, *J* = 4.0 Hz, 3/5H), 3.41 (d, *J* = 8.4 Hz, 3/5H), 1.12 (s, 9/5H), 1.12 (s, 6/5H), 1.11 (s, 6/5H), 1.07 (s, 9/5H). ¹³C NMR (CDCl₃, 100 MHz) δ 127.5-128.7, 103.1, 97.8, 91.8, 85.6, 76.7, 79.1, 74.7, 72.7, 42.5, 26.1, 24.4, 20.8, 20.1. *m/z* found 245.07, [M + Na⁺] calcd. C₁₃H₁₈O₃Na⁺ = 245.12 amu

² Dueno, E. E.; Chu, F.; Kim, S.; Jung, K. Y. *Tetrahedron Lett.* **1999**, *40*, 1843-1846

(E,Z)-(S)-3-Benzyloxy-2,2-dimethyl-5-phenyl-pent-4-en-1-ol (15) – To a stirred solution of benzyl triphenylphosphonium bromide (1.21 g, 2.78 mmol) in THF (15 mL) at -78°C, potassium t-butoxide (1 M in THF, 2.69 mL, 2.69 mmol) was added. The solution immediately turned orange, and was allowed to stir as it turned to crimson-orange. After 30 min, a solution of **9** (206 mg, 0.928 mmol) in THF was cannulated into the stirring ylide, and the reaction was allowed to warm to room temperature. After two hours, the reaction was driven to completion by heating to reflux for 40 min. The reaction was quenched with NH₄Cl(sat) (3 mL), diluted with diethyl ether (50 mL) and the organic layer was washed with water (10 mL) and brine (10 mL) whereupon it was dried with Na₂SO₄ and concentrated *in vacuo* until a yellow oil remained. The compound was purified by flash chromatography (8:1 to 4:1 Hex:EtOAc) and concentrated to a clear yellowish oil (260 mg, 95%). The product was a mixture of regioisomers that was about 3:2 E/Z. ¹H NMR (300 MHz, CDCl₃) δ 7.45-7.20 (m, 10H^{E&Z}), 7.10-7.08 (m, 2H^{E&Z}), 6.83 (d, *J* = 12.0 Hz, 1H^Z), 6.54 (d, *J* = 15.9 Hz, 1H^E), 6.19 (dd, *J* = 16.2, 8.4 Hz, 1H^E), 5.71 (dd, *J* = 12.0, 10.8 Hz, 1H^Z), 4.64 (d, *J* = 11.7 Hz, 1H^E), 4.55 (d, *J* = 11.7 Hz, 1H^Z), 4.34 (d, *J* = 11.7 Hz, 1H^E), 4.28 (d, *J* = 11.1 Hz, 1H^Z), 4.11 (d, *J* = 11.7 Hz, 1H^Z), 3.80 (d, *J* = 8.4 Hz, 1H^E), 3.58 (d, *J* = 10.9 Hz, 1H^E), 3.54 (d, *J* = 10.9 Hz, 1H^Z), 3.40 (d, *J* = 11.1 Hz, 1H^E), 3.33 (d, *J* = 11.1 Hz, 1H^E), 0.91-0.94 (m, 6H). ¹³C NMR (100 MHz, CDCl₃) δ 137.91, 137.85, 136.6, 134.5, 126.3-128.8 (many signals), 87.67, 87.63, 71.4, 70.5, 70.0, 39.35, 39.31, 22.84, 22.79, 20.1, 19.9. *m/z* found 319.08, [M+Na]⁺ calcd. C₂₀H₂₄O₂Na⁺: 319.18 amu

(R)-2-Benzyloxy-4-(bis-benzyloxy-phosphoryloxy)-3,3-dimethyl-butyraldehyde (16) – To a stirred suspension of tetrazole (27 mg, 0.38 mmol) in CH₂Cl₂ (5 mL) at room temperature, *N,N*-diisopropyl-*O,O'*-dibenzyl phosphoramidite (131 mg, 127 μL, 0.38 mmol) was added. After 15 minutes, **15** dissolved in CH₂Cl₂ (2 mL) was cannulated into the stirring solution. After 2.5 hours, the solution was diluted with CH₂Cl₂, washed with water (5 mL), brine (5 mL), and dried over anhydrous Sodium Sulfate, and evaporated *in vacuo*. The residual oil was redissolved in a solution of CH₂Cl₂/MeOH (9:1, 5 mL), cooled to -78°C, and ozone was bubbled through the solution for 3 minutes. Dimethyl sulfide (1 mL) was added, and white vapor evolved in the flask. The flask was then removed from the -78°C bath, and the solvent was evaporated *in vacuo*. Purification followed by flash chromatography (2:1 to 1:2 Hexanes/EtOAc) to yield aldehyde **16** (113 mg, 62%) as a clear viscous oil. ¹H NMR (400 MHz, CDCl₃) δ 9.66 (d, *J* = 2.8 Hz, 1H), 7.34-7.23 (m, 15H), 5.04-4.99 (m, 4H), 4.55 (d, *J* = 11.2 Hz, 1H), 4.40 (d, *J* = 11.6 Hz, 1H), 3.87 (dd, *J* = 9.6, 4.4 Hz, 1H), 3.80 (dd, *J* = 9.6, 4.4 Hz, 1H), 3.46 (d, *J* = 2.8 Hz, 1H), 0.95 (s, 3H), 0.94 (s, 3H). ¹³C NMR (100 MHz, CDCl₃) δ 203.6, 137.0, 135.6 (d, *J* = 6.8 Hz), 127.7-128.5 (multiple signals), 86.4, 73.1, 72.1 (d, *J* = 6.1 Hz), 69.3 (d, *J* = 5.3 Hz), 39.8 (d, *J* = 8.3 Hz), 21.5, 19.8. ³¹P NMR (121.4 MHz, CDCl₃) δ -1.15 ppm.

(R)-2-Benzyloxy-4-(bis-benzyloxy-phosphoryloxy)-3,3-dimethyl-butyric acid (17) – Aldehyde **16** (74 mg, 0.15 mmol) was dissolved in MeOH/CH₂Cl₂/H₂O (6:3:2, 5 mL). NaH₂PO₄ (83 mg, 0.60 mmol) was added followed by 80% NaClO₂ (34 mg, 0.30 mmol). The solution turned green after 10 min. After 3.5 h, the reaction was complete by TLC (1:2 hexanes/EtOAc). The reaction was quenched with 1 M HCl (1 mL) and the volatile solvents were evaporated *in vacuo*. The remaining material was extracted with CH₂Cl₂ (3x30 mL), and the organic extractions were combined washed with brine (10 mL), dried over anhydrous sodium sulfate, and evaporated under reduced pressure to yield **17** as a clear oil (75 mg, 99%). The product was used and characterized without further purification. It should be noted that the NMR is pH sensitive,

and reported spectra were taken immediately after extraction. Further manipulation can cause some peaks to shift relative positions. ^1H NMR (400 MHz, CDCl_3) δ 7.33-7.24 (m, 15H), 5.00-4.97 (m, 4H), 4.58 (d, J = 11.2 Hz, 1H), 4.35 (d, J = 10.8 Hz, 1H), 3.93 (dd, J = 9.6, 4.8 Hz, 1H), 3.80 (dd, J = 10.0, 4.8 Hz, 1H), 3.80 (s, 1H), 0.99 (s, 3H), 0.95 (s, 3H). ^{13}C NMR (100 MHz, CDCl_3) δ 173.0, 136.9, 135.7, 128.9-128.0 (multiple signals), 81.6, 72.6 (d, J = 6.1 Hz), 69.6-69.3 (m), 73.2, 38.9 (d, J = 8.4 Hz), 21.3, 20.0.

Preparation of Phosphopantetheine (2)

Phosphoric acid dibenzyl ester (R)-3-benzyloxy-2,2-dimethyl-3-[2-(2-tritylsulfanyl-ethylcarbamoyl)-ethylcarbamoyl]-propyl ester (18) – 10 was deprotected by treatment with 20% piperidine in DMF (5 mL). Once the deprotection was apparent by TLC (1:2 hexanes/EtOAc), the mixture was concentrated and evaporated under vacuum until there was no remaining piperidine or DMF. The crude film of **11** was taken to the next step without further treatment.

EDC (19 mg, 0.098 mmol), and HOBt (15 mg, 0.098 mmol) were dissolved in THF (3 mL), and in separate flasks **17** (49 mg, 0.098 mmol) and **11** (78 mg, 0.187 mmol), were dissolved in THF (2 mL each). The solution of **17** was cannulated into the flask with EDC and HOBt, followed by cannulation of the solution containing **11**. DIPEA (100 μL) was then added and all of the solids within the flask dissolved. The reaction was allowed to stir for 23 hours before quenching with water. The solution was diluted with diethyl ether to 50 mL, the aqueous layer was removed, and the organic was washed with 1 M HCl (5 mL), NaHCO_3 (sat) (10 mL), and brine (10 mL). The organic layer was dried over anhydrous sodium sulfate and concentrated *in vacuo*. The resultant film was azeotroped twice with 25 mL MeOH and purified by silica gel chromatography (1:1 Hexanes/EtOAc to pure EtOAc). **18** (41 mg, 47%) was obtained as a clear film. ^1H NMR (300 MHz, CDCl_3) δ 7.40-7.15 (m, 30H), 7.00 (t, J = 6.6 Hz, 1H), 5.71 (t, J ~ 6 Hz, 1H), 4.99 (t, J = 7.8 Hz, 4H), 4.39 (d, J = 10.8 Hz, 1H), 4.27 (d, J = 10.8 Hz, 1H), 3.93 (dd, J = 9.6, 4.5 Hz, 1H), 3.72 (dd, J = 9.6, 4.5 Hz, 1H), 3.62 (s, 1H), 3.46 (q, J ~ 6 Hz, 2H), 3.00 (q, J ~ 6 Hz, 2H), 2.35 (t, J = 6.6 Hz, 2H), 2.27 (t, J = 6.9 Hz, 2H), 0.93 (s, 3H), 0.83 (s, 3H). ^1H -COSY couplings δ 7.00-3.46, 5.71-3.00, 4.39-4.27, 3.93-3.72, 3.46-2.27, 3.00-2.35. ^{13}C NMR (100 MHz, CDCl_3) δ 170.6, 170.4, 144.4, 136.7, 129.4-126.6 (multiple signals), 83.3, 73.6, 73.0 (m), 69.2 (m), 38.9 (d, J = 9.1 Hz), 38.3, 35.6, 35.0, 31.8, 21.2, 20.1. ^{31}P NMR (121 MHz, CDCl_3) δ -1.45 ppm.

Phosphopantetheine (2) – Naphthalene (271 mg, 2.10 mmol) in THF (2 mL), was added to lithium metal (15 mg, 2.2 mmol) that had been rinsed with dry hexanes. After 30 min, a dark green color evolved which turned so dark it appeared black 1 hour after addition of naphthalene. After 1.25 h, the solution was cooled to -20°C in an isopropanol/dry ice bath, and **18** (25 mg, 0.028 mmol) in THF (3 mL) was added by cannula. The solution turned from black to light red immediately. After 2 more hours water (2.5 mL) was added to the solution which removed all color. More water was added (5 mL), and the solution was washed with CH_2Cl_2 (4x, 20 mL) and with diethyl ether (15 mL). Extra solvent was evaporated, and then the aqueous layer was lyophilized. After lyophilization, a yellow solid remained, and this solid was passed through a small column of acid form AG-50W-X8 ion exchange resin, and the eluant was immediately passed through a column of Na^+ loaded AG-50W-X8 ion exchange resin. The eluant was

lyopolized to give **2** (10 mg, 90%) as a white sticky solid. ^1H NMR (400 MHz, D_2O) δ 4.12 (s, 1H), 3.75 (dd, $J = 10.8, 6.8$ Hz, 1H), 3.52 (m, 4H), 3.40 (dd, $J = 10.0, 5.2$ Hz, 1H), 2.86 (t, $J = 10.8$ Hz, 2H), 2.53 (t, $J = 10.4$ Hz, 2H), 1.00 (s, 3H), 0.84 (s, 3H). ^{31}P NMR (D_2O , 121 MHz) δ 4.50 ppm. Note that the spectra of Phosphopantetheine are pH sensitive³.

Preparation of Pantetheine & Phosphopantetheine Derivatives

General procedure for preparation of Tr-Cystamine-Amino Acid combinations (20a-f) – 9 (100 mg, 0.287 mmol, 1.0 eq), EDC (138 mg, 0.718 mmol 2.5 eq), HOBT (132 mg, 0.861 mmol, 3.0 eq), and one Fmoc protected Amino Acid of **19a-f** (0.431 mmol, 1.5 eq) were combined and dissolved in THF (5 mL). DIPEA (186 mg, 250 μL , 1.44 mmol, 5.0 eq) was added, and the solutions were allowed to stir for 4 hr. The reactions were quenched with 25% ammonium chloride (2 mL), diluted with diethyl ether (25 mL), washed with water (5 mL) and brine (5 mL), dried over Na_2SO_4 , and evaporated *in vacuo*. The residue was purified by column chromatography with conditions specific for each amino acid.

S-Trityl Cystamine-Fmoc Glycine (20a) – The residue was purified by silica gel chromatography (2:1 Hexanes/Ethyl Acetate to Ethyl Acetate) to yield **20a** (147mg, 84%) as a flaky solid. ^1H NMR (400MHz, CDCl_3) δ 7.75 (d, $J = 7.2$ Hz, 2H), 7.67 (d, $J = 7.6$ Hz, 2H), 7.41 – 7.19 (m, 19H), 5.92 (b, 1H), 5.38 (b, 1H), 4.41 (d, $J = 7.6$ Hz, 2H), 4.20 (t, $J = 7.6$ Hz, 1H), 3.77 (d, $J = 5.2$ Hz, 2H), 3.10 (q, $J = 9.0$ Hz, 2H), 2.42 (t, $J = 8.0$ Hz, 2H). ^{13}C NMR (100MHz, CDCl_3) δ 168.6, 162.6, 144.6, 143.7, 141.3, 129.5 – 126.8 (multiple signals), 125.0, 120.0, 67.2, 66.9, 47.1, 44.4, 38.2, 36.5, 31.7, 31.5. m/z found 621.07, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{38}\text{H}_{34}\text{O}_3\text{N}_2\text{SNa}^+$: 621.22 amu.

S-Trityl Cystamine-Fmoc L-Phenylalanine (20b) – The residue was purified by silica gel chromatography (2:1 to 1:1 Hexanes/Ethyl Acetate) to yield **20b** (174mg, 87%) as a clear film. ^1H NMR (400MHz, CDCl_3) δ 7.76 (d, $J = 8.0$ Hz, 2H), 7.53 (t, $J = 6.8$ Hz, 2H), 7.41 -7.15 (m, 24H), 5.58 (b, 1H), 5.35 (b, 1H), 4.41 (t, $J = 7.2$ Hz, 1H), 4.32 (b, 1H), 4.28 (m, 1H), 4.17 (t, $J = 6.8$ Hz, 1H), 3.08 (b, 1H), 2.94 (m, 3H), 2.27 (m, 2H). ^{13}C NMR (100MHz, CDCl_3) δ 170.3, 155.8, 144.5, 141.3, 129 – 126 (multiple peaks) 125.0, 120.0, 66.9, 56.2, 47.1, 38.8, 38.3, 31.4, 21.0, 14.1. m/z found 711.16, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{45}\text{H}_{40}\text{O}_3\text{N}_2\text{SNa}^+$: 711.27 amu.

S-Trityl Cystamine-Fmoc (N-Benzyl L-Histidine) (20c) – The residue was purified by silica gel chromatography (1:1 Hexanes/Ethyl Acetate to 1:4 methanol/ethyl acetate) to yield **20c** (190mg, 86%) as a clear glass. ^1H NMR (400MHz, CDCl_3) δ 7.75 (d, $J = 7.8$ Hz, 2H), 7.59 (d, $J = 7.0$ Hz, 2H), 7.39-7.18 (m, ~20H), 7.10 (s, 1H) 7.09 (s, 1H), 6.70 (b, 1H), 4.93 (s, 2H), 4.42 (q, $J = 4.4$ Hz, 1H), 4.34 (m, 2H), 4.21 (t, $J = 7.3$ Hz, 1H), 3.05 – 2.95 (m, 4H), 2.31 (t, $J = 6.2$ Hz, 2H). ^{13}C NMR (100MHz, CDCl_3) δ 170.9, 144.7, 136.8, 129.6 - 126.8 (multiple signals), 117.1, 83.2, 73.7, 69.4, 53.0, 51.0, 38.6, 32.1, 30.6, 21.7, 20.0. m/z found 769.10, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{49}\text{H}_{44}\text{O}_3\text{N}_4\text{SNa}^+$: 769.32 amu.

S-Trityl Cystamine-Fmoc D-Alanine (20d) – The residue was purified by silica gel chromatography (2:1 to 1:1 Hexanes/Ethyl Acetate) to yield **20d** (157mg, 88%) as a clear film. ^1H NMR (400MHz, CDCl_3) δ 7.76 (d, $J = 7.6$ Hz, 2H), 7.55 (d, $J = 6.8$ Hz, 2H), 7.41-7.17 (m,

³ Lee, C.; Sarma, R. H. *J. Am. Chem. Soc.* **1975**, 97, 1225-1235

19H), 6.17 (b, 1H), 5.45 (d, $J \sim 6.0$ Hz, 1H), 4.37 (d, $J \sim 5.0$ Hz, 2H), 4.19 (t, $J \sim 5.0$ Hz, 1H), 3.07 (q, $J = 6.4$ Hz, 2H), 2.40 (b, 2H), 1.34 (d, $J \sim 6.4$ Hz, 3H). ^{13}C NMR (100MHz, CDCl_3) δ 172.1, 155.9, 144.6, 143.8, 143.7, 141.3, 129.5 – 126.8 (multiple signals), 125.1, 120.0, 670, 66.8, 50.5, 47.1, 38.3, 31.8, 18.8. m/z found 635.10, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{39}\text{H}_{36}\text{O}_3\text{N}_2\text{SNa}^+$: 635.23 amu.

S-Trityl Cystamine-Fmoc (*t*-Butyl *L*-Aspartate) (20e) – The residue was purified by silica gel chromatography (2:1 to 1:1 Hexanes/Ethyl Acetate) to yield **20e** (64mg, 30%) as a clear film. ^1H NMR (400MHz, CDCl_3) δ 7.76 (t, $J = 8.0$ Hz, 2H), 7.70 (d, $J = 7.6$ Hz, 2H), 7.57 (t, $J = 6.8$ Hz, 2H), 7.40 – 7.17 (m, 17H), 6.56 (b, 1H), 5.01 (b, 1H), 4.41 (m, 2H), 4.20 (t, $J = 6.8$ Hz, 2H), 3.06 (m, 2H), 2.86 (dd, 1H), 2.57 (dd, $J = 16.8, 6.8$ Hz, 1H), 2.40 (t, $J = 6.8$ Hz, 2H), 1.43 (s, 9H). ^{13}C NMR (100MHz, CDCl_3) δ 171.1, 170.1, 155.9, 144.5, 143.7, 141.2, 140.1, 137.9, 129.4 – 126.7 (multiple signals), 124.9, 120.9, 120.0, 119.7, 107.8, 81.8, 77.2, 67.1, 66.7, 50.9, 47.1, 38.3, 37.4, 31.6, 28.0. m/z found 735.11, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{44}\text{H}_{44}\text{O}_5\text{N}_2\text{SNa}^+$: 735.29 amu.

S-Trityl Cystamine-Fmoc *L*-Valine (20f) – The residue was purified by silica gel chromatography (2:1 to 1:1 hexanes/ethyl acetate) to yield **20f** (68mg, 35%) as a clear film. ^1H NMR (300MHz, CDCl_3) δ 7.76 (d, $J = 7.5$ Hz, 2H), 7.57 (d, $J = 7.2$ Hz, 2H), 7.41 – 7.17 (m, 19H), 5.84 (b, 1H), 5.34 (d, $J = 7.8$ Hz, 1H), 4.38 (p, $J = 7.8$ Hz, 2H), 4.20 (t, $J = 6.9$ Hz, 1H), 3.87 (t, $J = 7.5$ Hz, 1H), 3.06 (m, 2H), 2.07 (m, 1H), 0.92 (d, $J = 7.2$ Hz, 3H), 0.90 (d, $J = 7.5$ Hz, 3H). ^{13}C NMR (100MHz, CDCl_3) δ 170.6, 156.1, 144.3, 143.6, 141.1, 129 – 126 (multiple signals), 124.9, 119.8, 66.9, 60.3, 47.2, 38.2, 31.9, 31.2, 19.3, 18.0. m/z found 663.15, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{41}\text{H}_{40}\text{O}_3\text{N}_2\text{SNa}^+$: 663.27 amu.

General Procedure for preparation of Amino Acid Substituted, Protected Pantetheines (21a-f) – Tr-Cystamine-AA **20a-f** (1.0eq) was dissolved in DMF (2 mL), and piperidine (1 mL) was added. After 30 minutes, the DMF and piperidine were evaporated *in vacuo* at 50°C. To the residue, **7** (1.5 eq), EDC (2.5 eq), HOBt (3.0 eq), and dry DMF (4 mL) were added. Immediately, DIPEA was added (6.0 eq). The reaction was stirred for 5 h, and then quenched with 25% ammonium chloride solution, diluted in ethyl acetate (20 mL), washed with water (5 mL), brine (5 mL), dried over Na_2SO_4 , and evaporated *in vacuo*. Each product was purified by silica gel chromatography according to its individual properties.

Protected Glycine Analog (21a) – The product was purified by prep TLC (1:1 hexanes/ethyl acetate) to afford **21a** (13mg, 52%) from **20a** (25mg, 0.040mmol) as a clear film. ^1H NMR (400MHz, CDCl_3) δ 7.43 – 7.21 (m, 17H), 7.07 (t, $J = 5.1$ Hz, 1H), 6.91 ($J = 8.6$ Hz, 2H), 6.11 (b, 1H), 5.43 (s, 1H), 4.11 (s, 1H), 3.82 (s, 3H), 3.71 (d, $J = 11.4$ Hz, 1H), 3.64 (d, $J = 11.4$ Hz, 1H), 3.05 (q, $J = 6.0$ Hz, 2H), 2.40 (t, $J = 6.4$, 2H), 1.13 (s, 3H), 1.09 (s, 3H). ^{13}C NMR (100MHz, CDCl_3) δ 170.0, 168.5, 144.7, 130 – 126 (multiple signals), 113.9, 101.6, 83.9, 78.5, 55.4, 42.8, 38.2, 33.3, 31.8, 21.9, 19.3. m/z found 647.13, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{37}\text{H}_{40}\text{O}_5\text{N}_2\text{SNa}^+$: 647.26 amu.

Protected Phenylalanine Analog (21b) – The product was purified by prep TLC (1:1 hexanes/ethyl acetate) to afford **21b** (16mg, 59%) from **20b** (29mg, 0.040mmol) as a clear film.

¹H NMR (400MHz, CDCl₃) δ 7.45 – 6.91 (m, ~25H), 5.85 (t, *J* = 5.6 Hz, 1H), 5.41 (s, 1H), 4.50 (q, *J* = 7.4 Hz, 1H), 4.02 (s, 1H), 3.84 (s, 3H), 3.71 – 3.60 (m, 3H), 3.55 (q, *J* = 5.7 Hz, 1H), 3.02 – 2.87 (m, 4H), 2.274 (t, *J* = 6.5 Hz, 1H), 2.268 (t, *J* = 6.8 Hz, 1H), 1.61 (m, 1H), 1.55 (m, 1H), 1.021 (s, 3H), 1.017 (s, 3H). ¹³C NMR (100MHz, CDCl₃) δ 170.2, 169.5, 160.3, 144.7, 136.5, 130.1 – 126.8 (multiple signals), 113.8, 102.2, 101.4, 84.0, 83.8, 79.8, 78.5, 66.9, 55.4, 53.8, 47.3, 43.5, 38.4, 37.7, 33.5, 31.5, 26.7, 25.8, 24.7, 21.8, 19.4 *m/z* found 737.14, [M+Na]⁺ calcd. C₄₄H₄₆O₅N₂SN⁺: 737.30 amu.

Protected *N*-Benzyl Histidine Analog (21c) – The product was purified by prep TLC (1:4 Methanol/Ethyl Acetate) to afford **21c** (7mg, 37%) from **20c** (19mg, 0.024mmol) as a clear film. ¹H NMR (400MHz, CDCl₃) δ 8.29 (d, *J* = 6.8 Hz, 1H), 7.53 (d, *J* = 8.5 Hz, 2H), 7.36 – 7.28 (m, ~11H), 7.07 (m, 2H), 6.99 (b, 1H), 6.88 (d, *J* = 8.7 Hz, 2H), 6.65 (s, 1H), 5.48 (s, 1H), 4.89 (s, 2H), 4.62 (q, 1H), 4.09 (s, 1H), 3.79 (s, 3H), 3.67 (s, 2H), 3.08 (dd, *J* = 14.8, 4.1 Hz, 1H), 2.96 (q, *J* = 6.3 Hz, 2H), 2.83 (dd, *J* = 14.7, 6.4 Hz, 1H), 2.29 (t, *J* = 6.6 Hz, 2H), 1.07 (s, 6H). ¹³C NMR (100MHz, CDCl₃) δ 170.9, 169.6, 144.7, 138.6, 136.7, 136.1, 130 – 126 (multiple signals), 117.4, 113.7, 101.5, 84.0, 79.0, 77.8, 67.0, 55.7, 52.8, 51.0, 38.6, 33.7, 32.1, 30.0, 22.3, 19.8 *m/z* found 795.35, [M+H]⁺ calcd. C₄₈H₅₁O₅N₄S⁺: 795.35 amu.

Protected D-Alanine Analog (21d) – The product was purified by prep TLC (1:1 Hexanes/Ethyl Acetate) to afford **21d** (24mg, 92%) from **20d** (26mg, 0.041mmol) as a clear film. ¹H NMR (400MHz, CDCl₃) δ 7.42 – 7.21 (m, 17H), 6.96 (d, *J* = 7.9 Hz, 1H), 6.92 (d, *J* = 8.7 Hz, 2H), 6.24 (t, *J* = 5.6 Hz, 1H), 5.36 (s, 1H), 4.41 (p, *J* = 7.4 Hz, 1H), 4.04 (s, 1H), 3.83 (s, 3H), 3.70 (d, *J* = 11.4 Hz, 1H), 3.60 (d, *J* = 11.4 Hz, 1H), 3.04 (q, *J* = 6.4 Hz, 2H), 2.38 (t, *J* = 7.1 Hz, 2H), 1.33 (d, *J* = 7.0 Hz, 2H), 1.12 (s, 3H), 1.09 (s, 3H). ¹³C NMR (100MHz, CDCl₃) δ 171.1, 169.5, 160.5, 144.9, 130.3 – 127.0, 114.0, 101.6, 83.9, 78.6, 77.5, 66.9, 55.6, 48.3, 38.3, 33.4, 32.0, 22.1, 19.3, 18.0. *m/z* found 661.14, [M+Na]⁺ calcd. C₃₈H₄₂O₅N₂SN⁺: 661.27 amu.

Protected *t*-Butyl L-Aspartic Acid Analog (21e) – The product was purified by prep TLC (1:1 Hexanes/Ethyl Acetate) to afford **21e** (12mg, 60%) from **20e** (20mg, 0.027mmol) as a clear film. ¹H NMR (400MHz, CDCl₃) δ 7.63 (d, *J* = 8.4 Hz, 1H), 7.40 – 7.38 (m, 6H), 7.27 – 7.25 (m, 6H), 7.20 (t, *J* = 6.1 Hz, 3H), 6.88 (d, *J* = 8.8 Hz, 2H), 6.55 (t, *J* = 5.8 Hz, 1H), 5.46 (s, 1H), 4.67 (m, 1H), 4.11 (s, 1H), 3.80 (s, 3H), 3.69 (d, *J* = 11.2 Hz, 1H), 3.65 (d, *J* = 11.2 Hz, 1H), 3.09 (6plet, *J* = 7.2 Hz, 1H), 2.95 (6plet, *J* = 6.9 Hz, 1H), 2.85 (dd, *J* = 17.0, 4.4 Hz, 1H), 2.52 (dd, *J* = 17.1, 6.4 Hz, 1H), 2.35 (t, *J* = 6.7 Hz, 2H), 1.33 (s, 9H), 1.07 (s, 3H), 1.06 (s, 3H). ¹³C NMR δ 170.9, 170.1, 169.8, 160.4, 144.8, 130.2 – 126.9 (multiple peaks), 113.9, 101.6, 83.9, 87.0, 78.7, 55.5, 48.8, 38.6, 36.6, 33.5, 31.9, 28.1, 22.0, 19.7. *m/z* found 761.16, [M+Na]⁺ calcd. C₄₃H₅₀O₇N₂SN⁺: 761.32 amu.

Protected L-Valine Analog (21f) – The product was purified by prep TLC (2:1 hexanes/ethyl acetate) to afford **21f** (17mg, 81%) from **20f** (21mg, 0.031mmol) as a clear film. ¹H NMR (400MHz, CDCl₃) δ 7.42 – 7.26 (m, 17H), 6.90 (d, *J* = 6.6 Hz, 2H), 5.99 (t, *J* = 5.7 Hz, 1H), 5.47 (s, 1H), 4.12 (s, 1H), 4.08 (dd, *J* = 8.8, 4.1 Hz, 1H), 3.82 (s, 3H), 3.70 (d, *J* = 11.4 Hz, 1H),

3.65 (d, $J = 11.5$ Hz, 1H), 3.18 – 2.98 (m, 2H), 2.50 – 2.30 (m, 2H), 2.14 (6plet, $J = 6.8$, 1H), 1.05 (s, 6H), 0.89 (d, $J = 6.8$, 2H), 0.84 (d, $J = 6.8$, 2H). ^{13}C NMR (100MHz, CDCl_3) δ 170.3, 169.3, 144.5, 130 – 126 (multiple peaks), 113.7, 101.3, 83.9, 78.5, 58.1, 55.4, 38.2, 33.5, 32.0, 30.3, 27.0, 19.7, 19.5, 18.2. m/z found 689.21, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{40}\text{H}_{46}\text{O}_5\text{N}_2\text{SNa}^+$: 689.30 amu.

General Procedure for preparation of Amino Acid Substituted Pantetheine Analogs (22a-f)

– To a stirred solution of **21a-f** in MeOH (2 mL), a solution of iodine in MeOH (0.03M) was dropped in until the solution remained dark brown (~ 1mL). After ten min, no starting material was detected by TLC (1:2 hexanes/ethyl acetate), and solid NaHSO_3 was added. The methanol solution was then diluted to 10 mL and washed 3x with hexanes (10 mL). The solution was passed through a bed of H^+ form AG50W-X8 ion exchange resin and ^-OH form Dowex AG 1-X8 resin to remove salts, and the eluant was evaporated *in vacuo*.

L-Phenylalanine Analog (22b) – Workup yielded **22b** (1 mg, 14%) as a clear film. ^1H NMR (400MHz, CDCl_3) δ 7.30 – 7.20 (m, 5H), 4.63 (dd, $J = 8.4$, 6.4 Hz, 1H), 3.80 (s, 1H), 3.48 – 3.40 (m, 4H), 3.11 (dd, $J = 13.6$, 6.4 Hz, 1H), 2.99 (dd, $J = 14.0$, 7.6 Hz, 1H), 0.90 (s, 3H), 0.84 (s, 3H). ^{13}C NMR (100MHz, CDCl_3) δ 130.3, 130.2, 129.4, 127.7, 77.1, 69.4, 55.4, 40.7, 39.6, 39.2, 38.2, 22.2, 21.2. m/z found 729.31, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{34}\text{H}_{50}\text{O}_8\text{N}_4\text{S}_2\text{Na}^+$: 729.23 amu (disulfide)

D-Alanine Analog (22d) – Workup yielded **22d** (7mg, 95%) as a clear film. ^1H NMR (400MHz, CDCl_3) δ 4.41 ppm (q, $J = 7.2$ Hz, 1H), 3.92 (s, 1H), 3.50 (dt, $J = 6.8$, 2.4 Hz, 2H), 3.48 (d, $J = 11.2$ Hz, 1H), 3.38 (d, $J = 11.2$ Hz, 1H), 2.82 (t, $J = 7.2$ Hz, 1H), 1.36 (d, $J = 6.8$ Hz, 3H), 0.94 (s, 3H), 0.93 (s, 3H). ^{31}P NMR (162MHz, CDCl_3) δ 175.2, 77.1, 70.1, 40.5, 39.6, 38.4, 21.3, 21.0, 19.0. m/z found 577.22, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{22}\text{H}_{42}\text{O}_8\text{N}_4\text{S}_2\text{Na}^+$: 577.23 amu (disulfide).

L-Aspartic Acid *t*-Butyl Ester Analog (22e) – Workup yielded **22e** (3.5 mg, 60%) as a clear film. ^1H NMR (400MHz, CDCl_3) δ 4.78 (t, $J = 5.8$ Hz, 1H), 3.88 (s, 1H), 3.64 (d, $J = 11.2$ Hz, 1H), 3.55 (t, $J = 7.2$ Hz, 2H), 3.37 (d, 1H), 2.90 – 2.70 (m, 4H), 1.52 (s, 9H), 1.09 (s, 3H), 0.99 (s, 3H).

General Procedure for preparation of Amino Acid Substituted, Protected Phosphopantetheine Analogs (23a-d) - Tr-Cystamine-AA **20a-f** (1.0eq) was dissolved in DMF (2 mL), and piperidine (1 mL) was added. After 30 min, the DMF and piperidine were evaporated *in vacuo* at 50°C. To the residue, **17** (1.0 eq), EDC (2.0 eq), HOBt (2.5 eq), and dry DMF (4 mL) were added. Immediately, DIPEA was added (5.0 eq). The reaction was stirred for 5 h, and then quenched with 25% ammonium chloride solution, diluted in ethyl acetate (20 mL), washed with water (5 mL), brine (5 mL), dried over Na_2SO_4 , and evaporated *in vacuo*. Each product was purified by silical gel chromatography according to its individual properties.

Protected Glycine Analog (23a) – **20a** (20 mg, 0.032 mmol) yielded **23a** (21mg, 75%) after purification by silica gel chromatography (2:1 hexanes/ethyl acetate to ethyl acetate). ^1H NMR (400MHz, CDCl_3) δ 7.40 – 7.19 (m, ~30H), 7.09 (t, $J = 5.6$ Hz, 1H), 5.90 (t, $J = 5.7$ Hz, 1H), 5.03 – 4.99 (m, 4H), 4.50 (d, $J = 10.9$ Hz, 1H), 4.36 (d, $J = 11.0$ Hz, 1H), 3.98 (dd, $J = 9.5$, 4.1 Hz, 1H), 3.78 – 3.75 (m, 2H), 3.72 (s, 1H), 3.06 (q, $J = 6.3$ Hz, 2H), 2.40 (t, $J = 6.4$ Hz, 2H),

0.98 (s, 3H), 0.88 (s, 3H). ^{13}C NMR (100MHz, CDCl_3) δ 171.2, 168.2, 144.4, 136.8, 129.4 – 126.8 (multiple signals), 83.2, 74.0, 73.1 (d, J = 6.1 Hz), 69.3 (d, J = 5.3 Hz), 67.0, 42.7, 39.2 (d, J = 8.3 Hz), 38.4, 31.9, 21.5, 20.0. ^{31}P NMR (162MHz, CDCl_3) δ -1.047 ppm. m/z found 879.22, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{50}\text{H}_{53}\text{O}_7\text{N}_2\text{PSNa}^+$: 879.32 amu.

Protected *L*-Penylalanine Analog (23b) – 20b (26 mg, 0.036 mmol) yielded **23b** (11mg, 31%) after purification by silica gel chromatography (2:1 hexanes/ethyl acetate to ethyl acetate). ^1H NMR (400MHz, CDCl_3) δ 7.38 – 7.08 (m, ~35H), 6.96 (d, J = 8.0 Hz, 1H), 5.88 (t, J = 5.8 Hz, 1H), 4.99 (t, J = 8.1 Hz, 1H), 4.56 (q, J = 7.0 Hz, 1H), 4.10 (d, J = 11.4 Hz, 1H), 4.07 (d, J = 11.3 Hz, 1H), 3.95 (dd, J = 11.3, 4.4 Hz, 1H), 3.68 (dd, J = 9.5, 4.6 Hz, 1H), 3.61 (s, 1H), 3.12 – 2.82 (m, 4H), 2.28 (t, J = 6.5 Hz, 2H), 0.91 (s, 3H), 0.79 (s, 3H). ^{13}C NMR δ 170.7, 170.2, 144.5, 136.8, 136.4, 129.5 – 126.7, 82.8, 73.4, 73.2, 69.5, 66.9, 53.7, 39.3, 38.5, 37.8, 31.6, 21.5, 19.9. ^{31}P NMR (162MHz, CDCl_3) δ -0.999 ppm. m/z found 969.23, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{57}\text{H}_{59}\text{O}_7\text{N}_2\text{PSNa}^+$: 969.37 amu.

Protected *N*-Benzyl *L*-Histidine Analog (23c) – 20c (27 mg, 0.034 mmol) yielded **23c** (9 mg, 26%) after purification by silica gel chromatography (2:1 hexanes/ethyl acetate to ethyl acetate). ^1H NMR (400MHz, CDCl_3) δ 7.92 (d, J = 7.3 Hz, 1H), 7.37 – 7.05 (m, ~35H), 6.64 (s, 1H), 5.02 – 4.97 (m, 4H), 4.87 (d, J = 14.6 Hz, 1H), 4.83 (d, J = 15.0 Hz, 1H), 4.62 (q, J = 5.7 Hz, 1H), 4.45 (d, J = 11.1 Hz, 1H), 4.27 (d, J = 11.2 Hz, 1H), 3.99 (dd, J = 9.3, 4.3 Hz, 1H), 3.73 (dd, J = 9.3, 4.5 Hz, 1H), 3.71 (s, 1H), 3.10 – 2.90 (m, 3H), 2.85 (dd, J = 14.9, 6.6 Hz, 1H), 2.30 (6plet, J = 7.5 Hz, 2H). ^{13}C NMR δ 170.9, 144.7, 137.4, 136.8, 129.7 – 126.8 (multiple signals), 117.1, 83.2, 73.7, 69.4, 53.0, 51.1, 38.6, 32.1, 30.6, 21.7, 20.1. ^{31}P NMR (162MHz, CDCl_3) δ -1.034 ppm. m/z found 1049.23, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{61}\text{H}_{63}\text{O}_7\text{N}_4\text{PSNa}^+$: 1049.41 amu.

Protected *D*-Alanine Analog (23d) – 20d (20 mg, 0.031 mmol) yielded **23d** (8 mg, 29%) after purification by silica gel chromatography (2:1 hexanes/ethyl acetate to ethyl acetate). ^1H NMR (400MHz, CDCl_3) δ 7.42 – 7.17 (m, ~30H), 7.06 (d, J = 7.7 Hz, 1H), 5.98 (t, J = 5.6 Hz, 1H), 5.05 – 5.00 (m, 4H), 4.47 (d, J = 10.7 Hz, 1H), 4.35 (8plet, J = 7.3 Hz, 1H), 4.28 (d, J = 10.7 Hz, 1H), 3.98 (dd, J = 9.4, 4.3 Hz, 1H), 3.77 (dd, J = 9.4, 4.5 Hz, 1H), 3.68 (s, 1H), 3.17 – 3.01 (m, 2H), 2.41 (t, J = 6.4 Hz, 2H), 1.29 (d, J = 7.0 Hz, 3H), 0.99 (s, 3H), 0.88 (s, 3H). ^{13}C NMR δ 171.4, 144.5, 136.7, 129.5 – 126.7 (multiple signals), 82.9, 74.0, 73.1, 69.4, 48.3, 39.2, 38.4, 32.0, 21.5, 19.9, 18.5. ^{31}P NMR (162MHz, CDCl_3) δ -0.984 ppm. m/z found 893.22, $[\text{M}+\text{Na}]^+$ calcd. $\text{C}_{51}\text{H}_{55}\text{O}_7\text{N}_2\text{PSNa}^+$: 893.34 amu.