Improved Biomimetic Total Synthesis of d,l-Stephacidin A

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General Methods. Unless otherwise noted, all materials were obtained from commercial sources and used without purification. All reactions requiring anhydrous conditions were performed under a positive pressure of argon using flame-dried glassware. Dichloromethane, acetonitrile, toluene, and tetrahydrofuran were degassed with argon and dried through a solvent purification system (J.C. Meyer of Glass Contour). Flash chromatography was performed on standard grade silica gel (230 x 400 mesh) from Sorbent Technologies with the indicated solvent. ¹H NMR and ¹³C NMR spectra were recorded on Varian 300 or 400 MHz spectrometers as indicated. Infrared spectra were recorded on a Nicolet Avatar 320-FT IR spectrometer. Mass spectra were obtained at the Colorado State University CIF on a Fisons VG Autospec. Melting points were obtained on a Mel-Temp Laboratory Device.

Stephacidin A (1). To a solution of alcohols **13** (79 mg, 0.18 mmol) in CH₂Cl₂ (18 mL) at rt was added DEAD (166 μ L, 1.06 mmol). The mixture was stirred at rt for 5 min and PBu₃ (264 μ L, 1.06 mmol) was then added. The solution was heated to 40 °C for 20 h. The resulting mixture was concentrated. Purification by silica gel chromatography (ethyl acetate : hexane, 1 : 1, then methanol : CH₂Cl₂, 5 : 95) afforded stephacidin A (**1**) and 6-*epi*-stephacidin A as a 2.4 : 1 crude mixture of diastereomers, respectively. This mixture was triturated with methanol to afford stephacidin A (**1**) as a white solid (34.7 mg, 46%). The methanol triturate was then concentrated to afford 6-*epi*-stephacidin A (13.5 mg, 18%); All spectral data for stephacidin A (**1**) was identical to that previously reported;¹ 6-*epi*-stephacidin A: ¹H NMR (400 MHz, CDCl₃ : CD₃OD, 20 : 1) δ 1.21 (s, 3 H), 1.28 (s, 3 H), 1.41 (s, 3 H), 1.42 (s, 3 H), 1.81 (dt, J = 7.4, 14.6 Hz, 1 H), 1.88 – 2.09 (m, 4 H), 2.24 (dd, J = 4.1, 9.9 Hz, 1 H), 2.71 (m, 1 H), 2.82 (d, J = 17.9 Hz, 1 H), 3.42 – 3.52 (m, 2 H), 3.78 (d, J = 17.9 Hz, 1 H), 5.62 (d, J = 9.7 Hz, 1 H), 6.61 (d, J = 8.3 Hz, 1 H), 6.64 (d, J = 9.7 Hz, 1 H), 7.19 (d, J = 8.3 Hz, 1 H), 8.54 (s, 1 H); ¹³C NMR (100 MHz, CDCl₃ : CD₃OD, 20 : 1) δ 23.6, 24.5, 24.9, 27.3, 27.5, 28.8, 29.1, 32.7, 34.7, 44.2, 45.9, 61.6, 67.1, 75.7, 103.8, 105.3, 110.1, 117.4, 118.3, 122.1, 129.7, 133.1, 138.7, 148.7,

¹ Greshock, T. J.; Grubbs, A. W.; Tsukamoto, S.; Williams, R. M. *Angew. Chem. Int. Ed.* **2007**, *46*, 2262.

169.5, 173.3; IR (neat) 3291, 2917, 1693, 1666 cm^{-1} ; ESI/APCI-HRMS (MH^+) calcd for $\text{C}_{26}\text{H}_{30}\text{N}_3\text{O}_3$ 432.2282, found 432.2267.

Improved Stephacidin A Synthesis

***N*-Boc-3-ketoproline ethyl ester.** To a solution of LiHMDS (1.0 M in THF, 7.91 mL, 7.91 mmol) in THF (60 mL) at -78 °C was added ethyl 3-(*tert*-butoxycarbonyl(2-ethoxy-2-oxoethyl)-amino)propanoate² in THF (6 mL) dropwise over 5 min. The mixture was stirred at -78 °C for 4 h. The resulting solution was poured onto 1 M aqueous HCl and extracted 3 times with ether. The combined extracts were washed 2 times with 10% aqueous Na₂CO₃, dried (Na₂SO₄), and concentrated to afford *N*-Boc-3-ketoproline ethyl ester as a colorless oil (1.20 g, 71%); All spectral data was identical to that previously reported.²

***cis*-3-Hydroxyproline ethyl ester trifluoroacetic acid salt (15).** To a solution of *N*-Boc-3-ketoproline ethyl ester (450 mg, 1.75 mmol) in MeOH (9 mL) at 0 °C was added NaBH₄ (66 mg, 1.75 mmol) in one portion. The mixture was stirred at 0 °C for 25 min. The resulting solution was quenched with 1 M aqueous HCl and extracted with ether. The combined extracts were dried (Na₂SO₄) and concentrated to afford the crude *cis*-*N*-Boc-3-hydroxyproline ethyl ester along with a small amount of the *trans* diastereomer (424 mg, 93%) as a mixture of amide rotamers, which was carried on without further

² Williams, R. M.; Cao, J.; Tsujishima, H.; Cox, R. J. *J. Am. Chem. Soc.* **2003**, *125*, 12172.

purification. ^1H NMR (300 MHz, CDCl_3) δ 1.25 (m, 3 H), 1.37 and 1.42 (s, 9 H), 1.91 – 2.11 (m, 2 H), 2.80 (br s, 1 H), 3.42 (m, 1 H), 3.60 (m, 1 H), 4.10 – 4.39 (m, 3 H), 4.57 (m, 1 H). To a solution of the crude alcohol (276 mg, 1.06 mmol) in CH_2Cl_2 (5 mL) at 0 °C was added TFA (1 mL). The mixture was warmed to rt and stirred 3 h. The resulting solution was concentrated directly to afford the crude amine TFA salt **15**, which was used directly in the next reaction without further purification.

Peptide (16). To a solution of acid **14**³ (330 mg, 0.89 mmol) in CH_3CN (4.5 mL) was added HATU (505 mg, 1.33 mmol), $i\text{Pr}_2\text{NEt}$ (617 μL , 3.54 mmol), and *cis*-3-hydroxyproline ethyl ester **15** successively at rt. The mixture was stirred at rt for 3 h. The resulting solution was quenched with 2 M aqueous HCl and extracted with CH_2Cl_2 . The combined extracts were dried (Na_2SO_4) and concentrated. Purification by silica gel chromatography (ethyl acetate : hexane, 3 : 1) afforded amide **16** as a mixture of diastereomers and amide rotamers (440 mg, 97%); ^1H NMR (400 MHz, CDCl_3) δ 1.21 and 1.25 (t, $J = 7.1$ Hz, 3 H), 1.41 (s, 9 H), 1.58 (s, 3 H), 1.60 (s, 3 H), 2.12 – 2.21 (m, 2 H), 3.00 – 3.40 (m, 4 H), 3.59 and 3.90 (m, 1 H), 4.06 (m, 1 H), 4.14 and 4.21 (q, $J = 7.1$

³ Stocking, E. M.; Sanz-Cervera, J. F.; Williams, R. M. *J. Am. Chem. Soc.* **2000**, *122*, 1675.

Hz, 2 H), 4.61 (m, 1 H), 5.16 (d, $J = 10.6$ Hz, 1 H), 5.20 (d, $J = 17.6$ Hz, 1 H), 5.58 and 5.60 (br s, 1 H), 6.09 (dd, $J = 10.6, 17.6$ Hz, 1 H), 6.95 – 7.11 (m, 2 H), 7.21 (m, 1 H), 7.40 (m, 1 H), 8.00 and 8.12 (s, 1 H); ^{13}C NMR (100 MHz, CDCl_3) δ 27.4, 27.9, 28.3, 28.5, 30.5, 30.8, 31.1, 32.4, 33.8, 39.2, 44.0, 44.3, 45.4, 52.8, 53.4, 61.6, 62.1, 62.9, 63.9, 65.1, 70.0, 70.3, 79.7, 106.1, 110.5, 112.7, 118.7, 119.5, 121.9, 129.7, 133.8, 140.8, 145.4, 154.6, 155.1, 167.8, 169.3, 171.9; IR (neat) 3383, 2977, 1740, 1694, 1635 cm^{-1} ; ESI/APCI-HRMS (MH^+) calcd for $\text{C}_{28}\text{H}_{40}\text{N}_3\text{O}_6$ 514.2912, found 514.2899.

Alcohol (17). To a solution of amide **16** (439 mg, 0.85 mmol) in CH_2Cl_2 (17 mL) at 0 °C was added TFA (1.7 mL). The mixture was warmed to rt and stirred 2 h. The resulting solution was quenched slowly with saturated aqueous NaHCO_3 and extracted with CH_2Cl_2 . The combined extracts were dried (Na_2SO_4) and concentrated to afford the crude primary amine. To a solution of the crude primary amine in toluene (17 mL) was added 2-hydroxypyridine (16.3 mg, 0.17 mmol). The mixture was heated to reflux for 16 h and then concentrated. The crude residue was diluted with CH_2Cl_2 , washed twice with 1 M HCl, dried (Na_2SO_4) and concentrated to afford alcohol **17** as an inseparable mixture of diastereomers (292 mg, 93%); ^1H NMR (400 MHz, CDCl_3) δ 1.48 (s, 3 H), 1.52 (s, 3 H), 1.92 – 2.18 (m, 2 H), 3.21 (m, 1 H), 3.48 (m, 1 H), 3.62 – 3.95 (m, 3 H),

4.20 (m, 1 H), 4.58 (m, 1 H), 5.09 – 5.18 (m, 2 H), 5.90 and 6.40 (s, 1 H), 6.10 (m, 1 H), 7.00 – 7.35 (m, 3 H), 7.47 (m, 1 H), 8.24 and 8.33 (s, 1 H); ^{13}C NMR (100 MHz, CDCl_3) δ 27.8, 28.0, 29.8, 30.0, 30.4, 33.5, 39.0, 44.0, 44.2, 45.2, 54.9, 58.4, 61.8, 63.9, 64.6, 65.1, 69.8, 70.8, 104.4, 104.9, 110.6, 111.0, 112.0, 112.8, 117.9, 118.5, 119.7, 120.1, 121.8, 122.1, 125.4, 128.3, 129.0, 134.3, 134.5, 141.8, 145.6, 146.0, 165.8, 166.4, 167.4; IR (neat) 3358, 2969, 1742, 1655 cm^{-1} ; ESI/APCI-HRMS (MH^+) calcd for $\text{C}_{21}\text{H}_{26}\text{N}_3\text{O}_3$ 368.1969, found 368.1957.

Cycloadducts (19 and 20). To a solution of alcohols **17** (100 mg, 0.27 mmol) in CH_2Cl_2 (27 mL) at rt was added DEAD (257 μL , 1.63 mmol). The mixture was stirred at rt for 5 min and PBU_3 (408 μL , 1.63 mmol) was then added. The solution was heated to 40 $^\circ\text{C}$ for 20 h. The resulting mixture was concentrated. Purification by silica gel chromatography (ethyl acetate : hexane, 1 : 1, then methanol : CH_2Cl_2 , 5 : 95) afforded cycloadducts **19** and **20** as a 2.1 : 1 crude mixture of diastereomers, respectively. This mixture was further separated by preparative TLC (MeOH : CH_2Cl_2 , 3 : 97) to afford *syn* cycloadduct **19** as a white solid (43.6 mg, 46%) along with *anti* cycloadduct **20** as a

white solid (20.1 mg, 21%); Both cycloadducts **19**⁴ and **20**⁵ have been previously synthesized. All data was identical to that previously reported.

Enamide (18). To a solution of alcohols **17** (120 mg, 0.33 mmol) in CH₂Cl₂ (6.5 mL) at rt was added DEAD (40% wt. in toluene, 446 μ L, 0.98 mmol). The mixture was stirred at rt for 5 min and PBu₃ (245 μ L, 0.98 mmol) was then added. The solution was stirred at rt for 3 h. The resulting mixture was then concentrated. Purification by silica gel chromatography (ethyl acetate : hexane, 2 : 3 to 1 : 0) afforded enamide **18** as a white foam (98.2 mg, 86%); ¹H NMR (400 MHz, CDCl₃) δ 1.52 (s, 3 H), 1.53 (s, 3 H), 2.76 (m, 2 H), 3.22 (dd, *J* = 11.3, 14.5 Hz, 1 H), 3.70 (dd, *J* = 3.5, 14.5 Hz, 1 H), 4.05 (m, 2 H), 4.50 (br d, *J* = 10.3 Hz, 1 H), 5.12 (d, *J* = 10.5 Hz, 1 H), 5.14 (d, *J* = 17.5 Hz, 1 H), 5.73 (s, 1 H), 6.10 (dd, *J* = 10.5, 17.5 Hz, 1 H), 6.13 (t, *J* = 2.9 Hz, 1 H), 7.08 (t, *J* = 7.7 Hz, 1 H), 7.14 (t, *J* = 7.7 Hz, 1 H), 7.30 (d, *J* = 7.7 Hz, 1 H), 7.50 (d, *J* = 7.7 Hz, 1 H), 8.39 (s, 1 H); ¹³C NMR (100 MHz, CDCl₃) δ 27.8, 27.9, 28.0, 30.9, 39.1, 45.6, 57.5, 104.4, 110.9, 112.3, 118.2, 119.0, 120.0, 122.0, 128.8, 133.1, 134.4, 141.8, 145.8, 156.5, 162.6; IR

⁴ (a) Williams, R. M.; Glinka, T.; Kwast, E.; Coffman, H.; Stille, J. K. *J. Am. Chem. Soc.* **1990**, *112*, 808. (b) Jin, S.; Wessig, P.; Liebscher, J. J. *Org. Chem.* **2001**, *66*, 3984. (c) Baran, P. S.; Hafensteiner, B. D.; Ambhaikar, N. B.; Guerrero, C. A.; Gallagher, J. D. *J. Am. Chem. Soc.* **2006**, *128*, 8678.

⁵ Adams, L. A.; Valente, M. W. N.; Williams, R. M. *Tetrahedron* **2006**, *62*, 5195.

(neat) 3341, 2967, 1677, 1644 cm^{-1} ; ESI/APCI-HRMS (MH^+) calcd for $\text{C}_{21}\text{H}_{24}\text{N}_3\text{O}_2$ 350.1863, found 350.1842.

Cycloadducts (19 and 20). To a solution of enamide **18** (92 mg, 0.26 mmol) in MeOH (20 mL) at 0 °C was added 20% aqueous KOH (5 mL). The mixture was slowly warmed to rt over 1 h and stirred for 18 h. The resulting solution was quenched with saturated aqueous NH_4Cl and extracted with CH_2Cl_2 . The combined extracts were dried (Na_2SO_4) and concentrated. Purification by preparative TLC (MeOH : CH_2Cl_2 , 3 : 97) afforded *syn* cycloadduct **19** as a white solid (55.2 mg, 60%) along with *anti* cycloadduct **20** as a white solid (26.9 mg, 29%).

Brevianamide B (9). To a solution of indole **20** (10.0 mg, 0.029 mmol) in THF (1 mL) at rt was added *m*CPBA (6.4 mg, 0.037 mmol). The mixture was stirred at rt for 1.5 h. The resulting solution was quenched with two drops of dimethyl sulfide and

concentrated to dryness to afford the crude 3-hydroxyindolinene, which was used without further purification. To a solution of the crude 3-hydroxyindolinene in MeOH (1 mL) was added 0.5 M aqueous NaOH (2 mL). The mixture was stirred at rt for 16 h and then warmed to 70 °C for 2 h. The resulting solution was cooled to rt, quenched with 1 M aqueous HCl, and extracted with CH₂Cl₂. The combined extracts were dried (Na₂SO₄) and concentrated. Purification by preparative TLC (MeOH : CH₂Cl₂, 5 : 95) afforded brevianamide B (**9**) as a yellow film (4.7 mg, 45%); All spectral data was identical to the natural sample as well as those synthetic samples previously prepared in our group.⁵ See attached ¹H NMR spectrum.

Oxindole (23). To a solution of indole **20** (16 mg, 0.046 mmol) in CH₂Cl₂ (1.8 mL) at rt was added oxaziridine **22** (33 mg, 0.14 mmol). The mixture was stirred at rt for 16 h. The resulting solution was placed directly on a column of silica gel and eluted with CH₂Cl₂, followed by MeOH : CH₂Cl₂ (5 : 95) to afford oxindole **23** as a thin film (11.8 mg, 71%); ¹H NMR (400 MHz, CDCl₃) δ 1.33 (s, 3 H), 1.45 (s, 3 H), 1.84 (dt, *J* = 7.3, 13.1 Hz, 1 H), 1.96 – 2.10 (m, 5 H), 2.75 (dt, *J* = 6.7, 13.1 Hz, 1 H), 2.82 (d, *J* = 15.8 Hz, 1 H), 3.42 (t, *J* = 6.7 Hz, 2 H), 3.49 (m, 1 H), 5.30 (s, 1 H), 7.13 (s, 1 H), 7.23 (t, *J* = 7.5 Hz, 1 H), 7.37 (t, *J* = 7.5 Hz, 1 H), 7.43 (d, *J* = 7.5 Hz, 1 H), 7.53 (d, *J* = 7.5 Hz, 1 H); ¹³C

NMR (100 MHz, CDCl₃) δ 20.2, 24.5, 27.5, 29.2, 32.7, 38.0, 40.5, 44.2, 50.5, 62.1, 67.3, 82.5, 121.3, 122.4, 126.7, 130.3, 140.5, 152.1, 168.4, 172.5, 188.2; IR (neat) 3365, 2968, 1688 cm⁻¹; ESI/APCI-HRMS (MH⁺) calcd for C₂₁H₂₄N₃O₃ 366.1812, found 366.1800.

400 MHz, CDCl₃ : CD₃OD (20 : 1)

6-*epi*-stephacidin A

S13

100 MHz, CDCl₃ : CD₃OD (20 : 1) 6-*epi*-stephacidin A

400 MHz, CDCl₃

S16

400 MHz, CDCl₃

S18

400 MHz, CDCl₃

S20

170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20

S21

100 MHz, CDCl₃

400 MHz, CDCl₃

23

S22