Supporting Information-1

Enantioselective Total Synthesis of Largazole, a Potent Inhibitor of Histone Deacetylase

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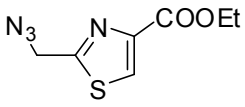
Contents

General Experimental Methods: 1H NMR and 13C NMR spectra were recorded on Bruker Avance ARX- 400 and DRX-500 spectrometers. IR spectra were recorded on a Mattason Genesis II FT-IR spectrometer. Optical rotations were recorded on a Perkin Elmer 341 polarimeter. Anhydrous solvents were obtained as follows: THF and diethyl ether by distillation from sodium and benzophenone; pyridine and dichloromethane from CaH_2. All other solvents were reagent grade. All moisture sensitive reactions were carried out in flame dried flask under argon atmosphere. Column chromatography was performed with Whatman 240-400 mesh silica gel under low pressure of 3-5 psi. TLC was carried out with E. Merck silica gel 60-F-254 plates.

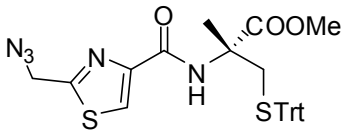


5

Ethyl 2-(azidomethyl)thiazole-4-carboxylate (5): To a stirring solution of 2-azidoacetamide (4.5 g, 44.5 mmol) in anhydrous THF (130 mL) was added Lawesson's reagent (6g, 14.7 mmol) in one portion. The resulting solution was allowed to stir at 23 °C overnight, after which the reaction mixture was concentrated in vacuo. The resulting oil was then partioned between Et_2O (100 mL) and saturated aqueous $NaHCO_3$ (200 mL), the organic layer was separated and the aqueous layer was extracted with Et_2O (3 x 100 mL). The organic layer was dried on anhydrous Na_2SO_4, concentrated *in vacuo,* and purified by column chromatography to give 2-azidoethanethioamide as a yellow oil (3.4 g, 67% yield). ^{1}H NMR (400 MHz, CD_3OD): δ 4.26 (s, 1 H). ^{13}C NMR (100 MHz, CD_3OD); δ 62.9, 204.8.

A mixture of above thioamide (3.4 g, 29.3 mmol) and ethyl bromopyruvate (5.2 mL, 18.8 mmol) in ethanol was heated to reflux for 1 h. The reaction mixture was then concentrated *in vacuo* and to this solution was added saturated aqueous $NaHCO_3$ (50 mL) and ethyl ether (50 mL). The organic layer was separated and the aqueous layer was

extracted with ethyl ether (3 x 50 mL), the combined organic layers were dried on anhydrous Na_2SO_4 and evaporated *in vacuo* to give an oil which was purified by flash chromatography (EtOAc: Hexanes 1:2) to give **5** as a colorless oil (5.10 g, 82% yield). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.35 (t, *J* = 7.1 Hz, 3H) 4.37 (q, *J* = 7 Hz, 2H), 4.73 (s, 2H), 8.15 (s, 1H); ^{13}C NMR (100 MHz, $CDCl_3$). 14.2, 51.4, 61.5, 128.2, 147.3, 160.9, 166.1. IR (film, NaCl) 3117, 2980, 2111, 1725 1484 cm^{-1}. HRMS (ESI) $[M]^+$ calcd for $C_7H_8N_4O_2S$ 212.0368, found 212.0367.

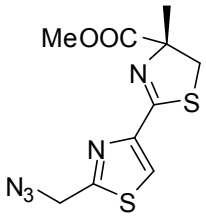


10

(*R*)-methyl-2-(2-(azidomethyl)thiazole-4-carboxamido)-2-methyl-3-(tritylthio)propanoate (10): LiOH (4.1 mL, 1 M soln in H_2O, 4.10 mmol) was added to a flask containing **5** (580 mg, 2.7 mmol) followed by a few drops of EtOH until the solution became homogeneous. This mixture was allowed to stir at 23 °C for 2 h after which the solvent was evaporated *in vacuo.* Ethyl acetate was added to the solid left behind and the mixture was acidified to pH 3 using 1N HCl. The organic layer was separated and the aqueous layer was extracted with EtOAc (3 x 10 mL), combined organic layers were then dried over anhydrous Na_2SO_4, evaporated under reduced pressure to give the acid (430 mg, 85% yield) as a white solid.

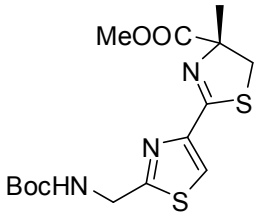
To a solution of the above acid (410 mg, 2.2 mmol) in CH_2Cl_2 (10 mL), was added EDC (556 mg, 2.9 mmol) and HOBt (392 mg, 2.9 mmol) at 0 °C. To this mixture was then added amine **6** (435 mg, 1.1 mmol) in CH_2Cl_2 (10 mL) followed by diisopropylethyl amine (3.8 mL, 22.3 mmol). The resulting solution was allowed to stir at 23 °C for 20 h, after which, the reaction mixture was concentrated under reduced pressure to remove solvents. The residue was then dissolved in EtOAc (25 mL) washed with saturated aqueous $NaHCO_3$, water, and brine successively. The organic layer was dried over anhydrous Na_2SO_4 and concentrated under reduced pressure to give an oil which was

purified by flash column chromatography (EtOAc:Hexanes 2:3) to give **10** (592 mg, 96% yield) as a white solid. $[\alpha]^{23}_{D} = -6.2^{o}$ (*c* 1, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.51 (s, 3H), 2.88 (d, *J* = 11.7 Hz, 1H), 3.07 (d, *J* = 11.7 Hz, 1H), 3.72 (s. 3H), 4.68 (s, 2H), 7.18-7.41 (m, 15H), 7.97 (s, 1H), 8.10 (s, 1H); ^{13}C NMR (100 MHz, $CDCl_3$); 23.1, 37.9, 51.1, 52.9, 59.2, 66.5, 124.3, 126.7, 127.8, 129.5, 144.4, 150.2, 159.7, 164.9, 172.9. IR (film, NaCl) 2171, 2104, 1739, 1676 cm^{-1}. HRMS (ESI) $[M+Na]^+$ calcd for $C_{29}H_{27}N_5O_3S_2Na$ 580.1453, found 580.1463



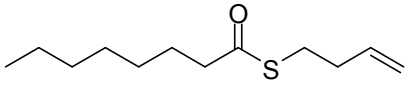
11

(*R*)-methyl-2-(2-(azidomethyl)thiazol-4-yl)-4-methyl-4,5-dihydrothiazole-4-carboxylate 11: To a ice-cooled solution of triphenylphosphine oxide (651 mg, 2.3 mmol) in CH_2Cl_2 (8 mL) was added Tf_2O (0.2 mL, 1.2 mmol) and the solution was stirred for 10 min. To this solution was then added **10** (435 mg, 0.78 mmol) at the same temperature, then the reaction was stirred for an additional 10 min. The reaction was quenched with saturated aqueous $NaHCO_3$, the organic layer was separated and the aqueous layer was extracted with CH_2Cl_2 (3 x 25 mL). The combined organic layers were dried over anhydrous $MgSO_4$, concentrated under reduced pressure and purified by flash column chromatography (EtOAc:Hexanes, 2:3) to give **11** (206 mg, 89% yield) as a colorless oil. $[\alpha]^{23}_{D} = -11.9^{o}$ (*c* 1.25, $CHCl_3$): ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.58 (s, 3 H), 3.22 (d, *J* = 11.3 Hz, 1H), 3.73 (s, 3H), 3.82 (d, *J* = 11.3 Hz, 1H), 4.67 (s, 2H), 7.97 (s, 1H). ^{13}C NMR (100 MHz, $CDCl_3$): 23.8, 41.4, 51.2, 52.8, 84.4, 122.1, 149.0, 162.6, 165.5, 173.4. IR (film, NaCl) 2368, 2299, 2104, 1727, 1599 cm^{-1}. HRMS (ESI) $[M]^+$ calcd for $C_{10}H_{11}N_5O_2S_2$ 297.0354, found 297.0353



3

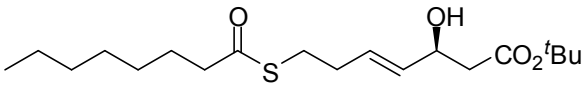
(*R*)-methyl-2-(2-((tert-butoxycarbonylamino)methyl)thiazol-4-yl)-4-methyl-4,5-dihydrothiazole-4-carboxylate (3): To a stirring solution of **11** (95 mg, 0.32 mmol) in MeOH (3.2 mL) was added PPh_3 (126 mg, 0.48 mmol) and the solution was heated to reflux for 1 h. After this time the soluiton was cooled to 23 ^oC and the solvent evaporated *in vacuo*. The resulting oil was then dissolved in CH_2Cl_2 (2 mL), cooled to 0 ^oC, and Boc_2O was added. The reaction was then allowed to warm to 23 ^oC and stirred overnight. The solvent was then evaporated and subjected to flash column chromatography (EtOAc: Hexanes 2:3) to give **4** (113 mg, 95% yield) as a colorless oil. $[\alpha]^{23}_D$ = -9.8 (*c* 1.33, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$): δ 1.42 (s, 9H), 1.61 (s, 3H), 3.23(d, *J* = 11.3 Hz, 1 H), 3.76 (s, 3H), 3.84 (d, *J* = 11.2 Hz, 1H), 4.59 (d, *J* = 5.8 Hz, 2H) 5.45 (br s, 1H), 7.91 (s, 1H). ^{13}C NMR (100 MHz, $CDCl_3$); 23.8, 28.2, 41.3, 42.2, 52.8, 80.2, 84.4, 121.7, 148.4, 155.6, 162.8, 169.6, 173.6. IR (film, NaCl) 3360, 2974, 2926, 1723, 1563 cm^{-1}. HRMS (ESI) $[M]^+$ calcd for $C_{15}H_{21}N_3O_4S_2$ 372.1052, found 372.1046.



8

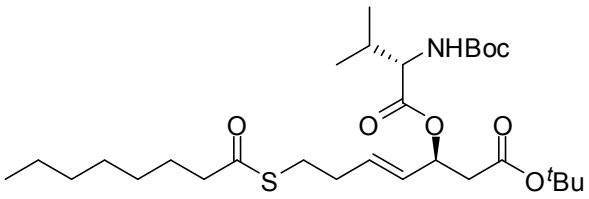
S-but-3-enyl octanethioate (8): To a stirred solution of but-3-ene-1-thiol (1.2 g, 14.3 mmol) in CH_2Cl_2 (50 mL) at 0 ^oC was added octanoyl chloride (2.9 mL, 17.2 mmol) followed by DMAP (2.3 g, 18.6 mmol). The reaction was allowed to warm to 23 ^oC and stirred for 16h. The reaction was then quenched with saturated aqueous NH_4Cl, the organic layer was separated, and the aqueous layer was extracted with EtOAc (3 x 20

mL). The combined organic layer was dried on anhydrous Na_2SO_4 and concentrated *in vacuo* and the resulting oil was subjected to column chromatography (EtOAc:Hexanes, 2:98) to give **8** (2.4g, 77%) as a colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.86 (t, *J* = 6.5 Hz, 3H), 1.25-1.28 (m, 9H), 1.60-1.68 (m, 2H), 2.31 (q, *J* = 7Hz, 2H), 2.52 (t, *J* = 7.5 Hz, 2H), 2.93 (t, *J* = 7.3 Hz, 2H), 5.02-5.09 (m, 2H), 5.72-5.82 (m, 1H); ^{13}C NMR (100 MHz, $CDCl_3$): 13.9, 22.5, 25.6, 27.9, 28.8, 31.5, 33.6, 44.0, 116.3, 136.0, 199.3. IR (film, NaCl) 2994, 2883, 1692, 1453 cm^{-1}.



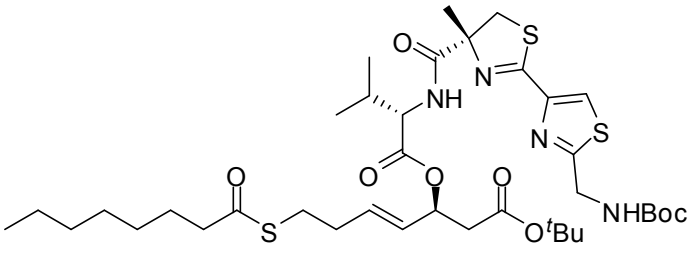
15

(*S*,*E*)-tert-butyl 3-hydroxy-7-(octanoylthio)hept-4-enoate (15): To a stirred solution of allylic alcohol **7** (700 mg, 4 mmol) and thioester **8** (1.7g, 8 mmol) in CH_2Cl_2 was added second generation Grubbs catalyst (100 mg, 0.12 mmol). The solution was heated to 40 ^{o}C for 12 h. The solvent was evaporated *in vacuo* and subjected to flash column chromatography (EtOAc: Hexanes, 1:6) to yield alcohol **3** (961 mg, 67%). $[\alpha]^{23}_D$ = -8.1 (*c* 0.97, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.87 (t, *J* = 6.5 Hz, 3H), 1.26-1.29 (m, 9H), 1.52 (s, 9H), 1.64 (t, *J* = 7 Hz, 2H), 2.30 (q, *J* = 7 Hz, 2H), 2.44 (m, 2H), 2.52 (t, *J* = 7.5 Hz, 2H), 2.90 (t, *J* = 7.3 Hz, 2H), 3.08 (d, *J* = 4 Hz, 1H), 4.44 (br s, 1H), 5.54 (dd, *J* = 6.1, 15.6 Hz, 1H), 5.66-5.73 (m, 1H). ^{13}C NMR (100 MHz, $CDCl_3$): 13.9, 22.5, 25.6, 28.0, 28.8, 31.5, 32.1, 42.2, 44.0, 68.6, 81.3, 129.2, 132.6, 171.74, 199.5. IR (film, NaCl) 2994, 2883, 1692, 1453 cm^{-1}. HRMS (ESI) $[M]^+$ calcd for $C_{19}H_{34}O_4S$ 357.2100, found 372.2093.



4

(*S*,*E*)-tert-butyl 3-((*S*)-2-(tert-butoxycarbonylamino)-3-methylbutanoyloxy)-7-(octanoylthio)hept-4-enoate (4): To a stirred solution of N-boc-valine **16** (182 mg, 0.84 mmol) in THF (4 mL) was added *i*Pr_2NEt (146 μL, 0.84 mmol) and 2,4,6-trichlorobenzyl chloride (131 μL, 0.84 mmol). After stirring the solution at 23 ^{o}C for 2 h, the solvent was removed under reduced pressure. The resulting residue was taken up in toluene (8 mL), **3** (100 mg, 0.28 mmol) and DMAP (103 mg, 0.84 mmol) were added and the reaction was stirred for another 2 h at 23 ^{o}C. The reaction was quenched with saturated aqueous NH_4Cl and extracted with Et_2O (3 x 10 mL). The combined organic layer was dried on anhydrous Na_2SO_4, concentrated *in vacuo*, and purified by column chromatography (EtOAc: Hexanes 1:6) to give **4** (141 mg, 91%) as a colorless oil. $[\alpha]^{23}_{D} = -11.6^{o}$ (*c* 1.16, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.76-0.83 (m, 6H), 0.92 (d, *J* = 6.8 Hz, 3H), 1.23-1.26 (m, 9H), 1.39 (s, 9H), 1.40 (s, 9H), 1.61 (t, *J* = 7 Hz, 2H), 2.06-2.13 (m, 1H), 2.25 (q, *J* = 7 Hz, 2H), 2.45-2.50 (m, 3H), 2.60 (dd, *J* = 7.8, 15.4 Hz, 1H), 2.84 (t, *J* = 7.2 Hz, 2H), 4.16 (dd, *J* = 4.5, 9.1 Hz, 1H), 4.99 (d, *J* = 9.1 Hz, 1H), 5.46 (dd, *J* = 7.4, 15.3 Hz, 1H), 5.59 (dd, *J* = 7.3, 13.7 Hz, 1H), 5.71-5.78 (m, 1H): ^{13}C NMR (100 MHz, $CDCl_3$): 14.0, 17.2, 18.9, 22.5, 25.5, 27.8, 27.9, 28.2, 28.8, 31.2, 31.5, 32.0, 40.7, 44.0, 58.2, 71.6, 79.5, 81.1, 128.4, 132.8, 155.5, 168.6, 171.1, 199.3. IR (film, NaCl) 2983, 2827, 1724, 1501, 1158 cm^{-1}. HRMS (ESI) $[M+Na]^+$ calcd for $C_{29}H_{51}NO_7SNa$ 580.3284, found 580.3289.



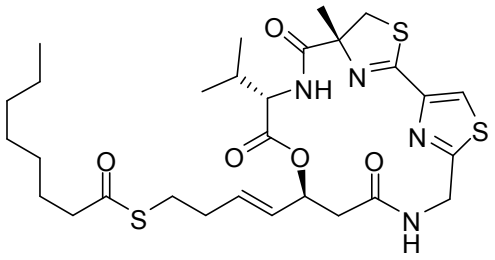
2

(*S*,*E*)-tert-butyl 3-((*S*)-2-((*R*)-2-(2-((tert-butoxycarbonylamino)methyl)thiazol-4-yl)-4-methyl-4,5-dihydrothiazole-4-carboxamido)-3-methylbutanoyloxy)-7-(octanoylthio)hept-4-enoate (2): To a stirred solution of ester **3** (45 mg, 0.12 mmol) in H_2O (0.5 mL) was added LiOH (8 mg, 0.18 mmol) and few drops of methanol at 23 ^oC and the reaction was stirred for 30 min. After this time the reaction mixture was concentrated under reduced pressure and the residue was acidified to pH 3 with 1N HCl. The aqueous layer was extracted with EtOAc (3 x 10 mL) and combined organic layer was dried on anhydrous Na_2SO_4, concentrated *in vacuo* to give the acid **18** (36 mg, 84%) as an oil.

To a stirred solution of **4** (68 mg, 0.12 mmol) in CH_2Cl_2 (1.5 mL) at 0 ^oC was added TFA (0.5 mL) and the resulting solution was stirred at 0 ^oC for 20 min. After this time the reaction mixture was carefully quenched with saturated aqueous $NaHCO_3$. The layers were separated and the aqueous layer was extracted with EtOAc (3 x 10 mL). The combined organic layer was dried on anhydrous Na_2SO_4, concentrated *in vacuo*, to give the amine **17** (52 mg, 95% yield) as an oil.

To the solution of above acid **18** (36 mg, 0.1 mmol) in CH_2Cl_2 (1 mL) was added HATU (46 mg, 0.12 mmol), HOAt (16 mg, 0.12 mmol) followed by the above amine **17** (46 mg,

0.1 mmol) in CH_2Cl_2 (1 mL) and *i*Pr_2NEt (17 μL, 0.1 mmol) and the reaction mixture was allowed to stir for 6 h. After this time the reaction mixture was concentrated *in vacuo* and subjected to flash column chromatography to give **2** (53 mg, 66%) as an oil. $[\alpha]^{23}_D$ = -38.7° (*c* 1.11, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.82 (d, *J* = 6.9 Hz, 3H),0.84-0.86 (m, 6H), 1.23-1.27 (m, 10H), 1.42 (s, 9H), 1.45 (s, 9H), 1.58 (s, 3H), 1.60-1.63 (m, 2H), 2.13-2.18 (m, 1H). 2.27 (q, *J* = 7 Hz, 2H), 2.49-2.53 (m, 3H), 2.64 (dd, *J* = 5.3, 15.5 Hz, 1H), 2.86 (t, *J* = 7.3 Hz, 2H), 3.32 (d, *J* = 11.6 Hz, 1H), 3.77 (d, *J* = 11.6 Hz, 1H), 4.49 (dd, *J* = 4.7, 9.1 Hz, 1H), 4.62 (d, *J* = 5.9 Hz, 1H) 5.28 (br s, 1H), 5.49 (dd, *J* = 7.5, 15.5 Hz, 1H), 5.62 (dd, *J* = 7.3, 13.7 Hz, 1H), 5.75-5.81 (m, 1H), 7.20 (d, *J* = 9.1 Hz, 1H), 7.96 (s, 1H). ^{13}C NMR (100 MHz, $CDCl_3$): 14.0, 17.5, 19.0, 22.5, 24.7, 25.6, 27.8, 28.2, 28.8, 31.1, 31.5, 32.1, 40.8, 41.4, 42.3, 44.0, 56.7, 71.9, 80.2, 81.2, 85.0, 121.3, 128.4, 133.0, 148.6, 155.6, 163.1, 168.6, 169.7, 170.4, 174.4, 199.3. HRMS (ESI) $[M+Na]^+$ calcd for $C_{38}H_{60}N_4O_8S_3Na$ 819.3471, found 819.3487.



1

Largazole (1): To a stirred solution of **2** (20 mg, 0.0251 mmol) in CH_2Cl_2 (2 mL) at 0 °C was added trifluoroacetic acid (1 mL) and the mixture was stirred at 23 °C for 3 h. After this time the reaction mixture was concentrated *in vacuo* and azeotropically dried

with toluene to give the crude amino acid. This amino acid was then dissolved in CH_2Cl_2 (25 mL) and HATU (19 mg, 0.0502 mmol), HOAt (7 mg, 0.0502 mmol) and *i*Pr_2NEt (17μL) were added sucessively. The reaction mixture was stirred for 4 h at 23 ^oC and then the solvent was evaporated under reduced pressue and redissolved in EtOAc. This solution was washed with H_2O and the aqueous layer was extracted with EtOAc (3 x 10 mL). The combined organic layer was dried on anhydrous Na_2SO_4 and evaporated under reduced pressure to give an oil which was subjected to column chromatography (EtOAc:CH_2Cl_2, 1:2) to give largazole **1** (6.3 mg, 40%) as a white solid. $[\alpha]^{23}_D = -24^o$ (*c* 0.13, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.51 (d, *J* = 6.8 Hz, 3H), 0.69 (d, *J* = 6.9 Hz), 0.87 (t, *J* = 6.6 Hz, 3H), 1.26-1.28 (m, 9H), 1.62-1.67 (m, 2H), 1.87 (s, 3H), 2.09-2.14 (m, 1H), 2.31 (q, *J* = 6.9 Hz, 2H), 2.53 (t, *J* = 7.5 Hz, 2H), 2.69 (dd, *J* = 2.6, 16.4 Hz, 1H), 2.86 (dd, *J* = 10.5, 16.4 Hz, 2H), 3.27 (d, *J* = 11.4 Hz, 1H), 4.04 (d, *J* = 11.4 Hz, 1H), 4.27 (dd, *J* = 2.9, 17.6 Hz, 1H), 4.61 (dd, *J* = 3.3, 9.4 Hz, 1H), 5.29 (dd, *J* = 9.5, 17.6 Hz, 1H), 5.29 (dd, *J* = 9.5, 17.6 Hz, 1H), 5.51 (dd, *J* = 6.8, 15.5 Hz, 1H), 5.65-5.68 (m, 1H), 5.82 (dt, *J* = 7.1, 14.9 Hz, 1H), 6.42 (d, *J* = 9.1 Hz, 1H), 7.16 (d, *J* = 9.5 Hz, 1H). ^{13}C NMR (100 MHz, $CDCl_3$): 14.1, 16.6, 18.9, 22.6, 24.2, 25.6, 27.9, 28.9, 31.6, 32.3, 34.2, 40.5, 41.1, 43.3,44.1, 57.7, 72.0, 84.5, 124.2, 128.4, 132.8, 147.5, 164.6, 167.9, 168.9, 169.4, 173.5, 199.4 81.1, 128.4, 132.8. 155.5, 168.6, 171.1, 199.3. HRMS (ESI) $[M+Na]^+$ calcd for $C_{29}H_{42}N_4O_5S_3Na$ 645.2215, found 645.2212.