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Selective transformations of cephalostatin analogues

Supplementary Information

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EXPERIMENTAL

Compound 4:

^{1}H NMR (400 MHz, $CDCl_3$): δ 5.54 (br s, 1H, 15'-H), 5.40 (br s, 1H, 15-H) 4.88 (dd, $J_{16'–17'}$ = 8.2 Hz, $J_{16'–15'}$ = 1.2 Hz, 1H, 16'-H), 4.71 (br s, 1H, 28b-H), 4.69 (br s, 1H, 28a-H), 4.67 (dd, $J_{16–17}$ = 7.6 Hz, $J_{16–15}$ = 2.1 Hz, 1H, 16-H), 3.40–3.56 (m, 5H, 26a/26b/26'a/ 26'b/22-H), 3.25 (dd, $J_{12'–11'a}$ = 10.6 Hz, $J_{12'–11'b}$ = 3.6 Hz, 1H, 12'-H), 2.80–3.05 (m, 4H, 1a/1'a/4a/4'a-H), 2.40–2.72 (m, 8H,1b/1'b/4b/4'b/17/17'/11a/11b-H), 2.26 (m, 1H, 8-H), 1.23 (s, 3H, 18-H), 1.13 (d, $J_{21–20}$ = 6.6 Hz, 3H, 21-H), 1.06 (d, $J_{21'–20'}$ = 6.9 Hz, 3H, 21'-H), 1.04 (s, 3H, 18'-H), 0.92 (d, $J_{27–25}$ = 6.7 Hz, 3H, 27-H),0.88 (s, 3H, 19'-H), 0.87 (s, 3H, 19-H), 0.81 (d, $J_{27'–25'}$ = 6.3 Hz, 3H, 27'-H). **^{13}C NMR** (100 MHz, $CDCl_3$): δ 160.4 (C, 14'-C), 157.2 (C, 14-C), 155.8 (C, 12-C), 148.7, 148.6, 148.4, 148.3 (all C, pyrazine–C), 119.8 (CH, 15'-C), 117.4 (CH, 15-C), 106.8 (CH_2, 28-C), 106.3 (C, 22'-C), 89.2 (CH, 22-C), 85.9 (CH, 16'-C), 84.6 (CH, 16-C), 79.0 (CH, 12'-C), 68.1 (CH_2, 26-C), 67.2 (CH_2, 26'-C), 58.5 (CH, 17-C), 56.1 (CH, 17'-C), 53.9 (C, 13-C), 52.7 (C, 13'-C), 52.1 (CH), 45.6, 45.6 (both CH_2, 1/10-C), 44.4 (CH), 41.5 (CH), 41.2 (CH), 40.1 (CH), 36.0, 35.9 (both C, 10/10'-C), 35.6, 35.5 (both CH), 35.2, 35.1 (both CH_2, 4/4'-C), 34.9 (CH),33.7 (CH), 32.4 (CH_2), 31.2 (CH_2), 30.8 (CH_2), 30.4 (CH), 30.3 (CH_2), 29.9 (CH_2), 29.4 (CH_2), 29.2 (CH_2), 28.8 (CH_2), 28.1 (CH_2), 27.8 (CH_2), 25.4 (CH_3, 18-C), 17.8 (CH_3, 21-C), 17.2 (CH_3, 27'-C), 16.6 (CH_3, 27-C), 13.9 (CH_3, 21'-C), 13.4 (CH_3, 18'-C), 11.8 (CH_3, 19'-C), 11.7 (CH_3, 19-C).

Compound 5:

Preparation of the salicylic acid-borane complex: In a dry flask flushed several times with argon, the salicylic acid solution (173 mg dissolved in 3.0 mL abs. THF at 0 °C) was treated with 1.3 mL of BH_3.THF (1.0 M solution) in slow addition. The solution was stirred for 30 min until no more gas was evolved.

To a solution of (**3**), prepared by dissolving 106 mg (126 μmole, 1 eq) in 2.0 mL abs. THF in a dry 50 mL round bottom flask at 0 °C, the salicylic acid-borane complex was slowly added and stirred for 1h at 0 °C. After conducting the reaction for four weeks at 2 °C, it was quenched with slow addition of 1.0 mL of 6.0 M NaOH solution followed by oxidation with 2.0 mL of 35% H_2O_2 solution with vigorous stirring for 2 h. Extracting the solution with chloroform twice followed by washing the organic phase with saturated solution of Na_2CO_3 and drying with $MgSO_4$. Removing the solvent and separating the crude material on silica gel column chromatography which eluted with EA/PE (1:2) yielded 36 mg of (**5**) (34%), 24 mg of the less polar methylene alcohol (in-between product) in addition to 10 mg of the starting material (**3**).

IR: ($CHCl_3/\nu_{max}/cm^{-1}$); 3617 br (O–H), 2931 s (C–H), 1649 w (C=C), 1634 w (C=C), 1458 s (C–H), 1400 s (pyrazine), 1242 s (C–O).

LC-MS (ESI/positive) 847.7230 (MH^+) base peak. Single Mass Analysis: found 846.5986 ($C_{55}H_{79}N_2O_5$), calculated 847.5989 (-0.4 PPM).

^{1}H NMR (400 MHz, $CDCl_3$): δ 5.45 (br s, 1H, 15'-H), 5.35 (br s, 1H, 15-H), 4.85 (dd, $J_{16\text{-}17}$ =8.0 Hz, $J_{16\text{-}15}$ = 1.8 Hz, 1H, 16-H), 4.83 (brs, 1H, 28a-H), 4.78 (dd, $J_{16'\text{-}17'}$ = 9.4 Hz, $J_{16'\text{-}15'}$ = 2.0 Hz, 1H, 16-H), 4.70 (brs, 1H, 28b-H), 3.42-3.57 (m, 4H, 26a/26b/ 26'a/26'b-H), 3.33 (m, 1H, 22'-H), 3.27 (m, 1H, 12'-H), 2.90-2.99 (m, 3H, 17'/1a/1'a-H), 2.81 (m, 2H, 4a/4'a-H), 2.35-2.65 (m, 6H, 1b/1'b/4b/4'b/ 11a/11b-H), 2.32 (tr, $J_{17\text{-}20}$ = $J_{17\text{-}16}$ = 8.0 Hz, 1H, 17-H), 2.27 (m, 1H, 8-H), 1.22 (s, 3H, 18-H), 1.09 (d, J21-20 = 6.8 Hz, 3H, 21-H), 1.06 (d, $J_{21'\text{-}20'}$ = 6.5 Hz, 3H, 21'-H), 1.05 (m, 1H, 9-H), 1.03 (s, 3H, 18'-H), 1.00 (m, 1H, 9'-H), 0.92 (d, $J_{27'\text{-}25'}$ = 6.9 Hz, 3H, 27'-H), 0.89 (s, 3H, 19-H), 0.87 (s, 3H, 19'-H), 0.81 (d, $J_{27\text{-}25}$ = 6.4 Hz, 27-H). **^{13}C NMR** (100 MHz, $CDCl_3$): δ 158.9 (C, 14-C), 157.7 (C, 14'-C), 155.2 (C, 12-C), 148.6, 148.5, 148.4, 148.3 (all C, pyrazine-C), 119.8 (CH, 15'-C), 117.7 (CH, 15-C), 107.4 (C, 22-C), 106.0 (CH_2, 28-C), 87.1 (CH, 22'-C), 86.0 (CH, 16'-C), 84.2 (CH, 16-C), 79.4 (CH, 12'-C), 68.2 (CH_2, 26'-C), 67.1 (CH_2, 26-C), 59.3 (CH, 17'-C), 54.7 (CH, 9-C), 53.9 (CH, 17-C), 53.4 (C, 13-C), 52.9 (C, 13'-C), 51.9 (CH, 9'-C), 45.7, 45.6 (2xCH_2, 1/1'-C), 44.1 (CH, 21-C), 41.4 (CH), 41.3 (CH), 41.2 (CH), 36.0 (C) , 35.9 (C), 35.8 (CH), 35.2, 34.6 (2xCH_2, 4/4'-C), 33.8 (CH), 32.3 (CH), 31.4 (CH_2), 30.4 (CH), 30.3 (CH_2), 30.2 (CH_2), 30.1 (CH_2), 29.7 (CH_2), 29.4 (CH_2), 29.2 (CH_2), 28.8 (CH_2), 28.0 (CH_2), 27.9

(CH_2), 23.7 (CH_3, 18-C), 17.2, 16.9, 16.6 (all CH_3, 18'/21/21'-C), 14.0, 13.9 (both CH_3, 27/27'-C), 11.8, 11.7 (both CH_3, 19/19'-C).

HMBC Experiment: (18-H, C-14), (18-H, C-12), (17-H, C-12), (21-H, C-17), (21-H, C-22), (18'-H, C-14'), (16'-H, C-14'), (17'-H, C-22'), (21'-H, C-17'), (21'-H, C-22').

Notes and spectral comparison between (**4**) and (**5**): from 1D-NMR, it was noticed that 15' and 16'-H in (**4**) are about 0.1 ppm down-fielded than their corresponding values in (**5**). However, the highest ^{13}C-chemical shift difference between (**4**) and **(5**) was recorded for C-14' which was about 2.7 ppm. On the other hand, a remarkable difference in the chemical shift between the two protons at C-28 was noticed. The $|J_{geminal}|$ between 28a and 28b-H in the closed-half of (**5**) was about 8-folds larger than that in the opened-half of (**4**). This established a quick technique to determine the opened-side in such analogues rather than 2D-NMR analysis.

Compound 7:

A solution of compound (**6**), resulted from dissolving 60 mg (68.6 μmol) in 5.0 mL abs. THF at 0°C, was treated with 3-methoxycatechol-borane complex (15 mg of the ligand dissolved in 1.0 mL of abs. THF and 0.1 mL of BH_3.THF). After stirring for one day at (0-5)°C, the substrate was further treated with 5-chlorosalicylic acid-borane complex (120 mg of the ligand (670 μmol) in 3.0 mL abs. THF and 1.0 mL BH_3.THF (1.0 M) prepared using the above-mentioned standard procedure. The reaction was conducted in refrigerator at 5°C for further 3 days before quenching it with 0.6 mL of 6.0 M NaOH solution followed by adding 0.6 mL of 35 % H_2O_2 solution and stirring for 1 h. Washing with saturated sodium carbonate solution twice to get ride of the free ligand followed by removing the solvent under low pressure, resulted in a light yellow crude material which after column chromatographic separation (silica gel, EA/PE (1:2) elution) yielded 47 mg (78 %) of the mono-opened product (**7**) in addition to 8 mg of the untransformed in-between product.

IR: ($CHCl_3/\nu_{max}/cm^{-1}$); 3610 br (O−H), 2931 s (C−H), 1728 s (C=O), 1648 w (C=C), 1602 w (C=C), 1458 m (C−H), 1400 s (pyrazine), 1241 s (C−O).

LC-MS (ESI/ positive): 979.5726 (MH^+), base peak. **FAB-MS**: (NBA-Matrix) m/z (%); 878 (M^+, 100).

^{1}H NMR (400 MHz, $CDCl_3$) δ 5.44 (br s, 1H, 15'-H), 5.34 (m, 1H, 15-H), 4.88 (dd, $J_{16'-17'}$ = 8.4 Hz, $J_{16'-15'}$ = 1.9 Hz, 1H, 16'-H), 4.76 (dd, J_{16-17} = 6.7 Hz, J_{16-15} = 2.9 Hz, 1H, 16-H), 3.74 (s, 3H, 28-H), 3.40-3.52 (m, 5H, 26a/26b/26'a/26'b/22-H), 3.24 (dd,

$J_{12'\text{-}11'a}$ = 11.2 Hz, $J_{12'\text{-}11'b}$ = 4.6 Hz, 1H, 12'-H), 2.91 (br d, $J_{1'a\text{-}1'b}$ = 16.5 Hz, 1H, 1'a-H), 2.82 (m, 2H, 4a/4'a-H), 2.34-2.69 (m, 9H, 1a/1b/1'b/4b/4'b/17/17'/11-H), 1.25 (s, 3H, 18-H), 1.06 (d, $J_{21'\text{-}20'}$ = 6.8 Hz, 3H, 21'-H), 1.03 (s, 3H, 18'-H), 0.97 (d, $J_{21\text{-}20}$ = 6.6 Hz, 3H, 21-H), 0.91 (d, $J_{27\text{-}25}$ = 6.7 Hz, 3H, 27-H), 0.88 (s, 3H, 19'-H), 0.83 (s, 3H, 19-H), 0.81 (d, $J_{27'\text{-}25'}$ = 6.2 Hz, 27'-H). **^{13}C NMR** (100 MHz, $CDCl_3$) δ 166.3 (C, 12-C), 157.2 (C, 14'-C), 148.6, 148.5, 148.4, 148.3 (all C, pyrazine-C), 135.2 (C, 14-C), 119.7 (CH, 15'-C), 113.3 (CH, 15-C), 106.7 (C, 22'-C), 91.6 (CH, 22-C), 90.2 (CH, 16-C), 84.5 (CH, 16'-C), 79.0 (CH, 12'-C), 68.1 (CH_2, 26-C), 67.2 (CH_2, 26'-C), 60.1 (CH), 58.4 (CH, 17-C), 57.4 (C, 13-C), 56.0 (CH, 17'-C), 52.7 (C, 13'-C), 52.4 (CH), 52.0 (CH_3, 28-C), 52.1 (CH, 9'-C), 45.6, 44.6 (both CH_2, 1/1'-C), 44.4 (CH), 42.7 (CH), 41.7 (CH), 41.5 (CH), 38.7 (CH, 8-C), 37.1 (C, 10-C), 36.0 (C, 10'-C), 35.8, 35.2 (both CH_2, 4/4'-C), 33.7 (CH), 32.2 (CH_2), 31.2 (CH_2), 31.0 (CH), 30.4 (CH), 29.9 (CH_2), 29.4 (CH_3, 18-C), 29.2 (CH_2), 28.9 (CH_2), 28.7 (CH_2), 28.0 (CH_2), 26.5 (CH_2), 23.4 (CH_2), 18.5 (CH_3, 27-C), 17.2 (CH_3, 27'-C), 16.6 (CH_3, 21-C), 14.1 (CH_3, 19-C), 13.9 (CH_3, 21'-C), 13.3 (CH_3, 18'-C), 11.8 (CH_3, 19'-C).

Compound 9:

A solution of (**8**) (55 mg, 60.8 μmol, dissolved in 2 mL abs. THF at 0 °C) was treated with the diaminobenzonitrile-borane complex. This complex was prepared from dissolving 80 mg of diaminobenzonitrile (600 μmol) in 3 mL abs.THF at 0 °C followed by slow addition of 0.6 mL of BH_3.THF (1M solution) then stirring for 30 min. The combined solution was stirred for 1h at 0 °C and conducted in refrigerator at 8 °C for one week which then resulted in a multi-spot products with one major (TLC follow). The reaction was worked up by adding 2 mL of 5.0 M NaOH solution and 2 mL of 35 % H_2O_2 solution and stirring vigorously for 2 h followed by extracting the resulted products with chloroform twice. Removing the solvents from the combined organic phase after drying with $MgSO_4$ and chromatographic separation of the crude material on column chromatography (silica gel, 1:2 EA:PE eluent) resulted in 20 mg of (**9**) (36 %) in addition to 12 mg of (**10**) and 7 mg of the starting material (**8**).

IR ($CHCl_3$/ν_{max} (cm^{-1})): 3612 br (O–H), 2930 s (C–H), 1725 s (C=O), 1647 w (C=C), 1458 m (C–H), 1399 s (pyrazine), 1243 (C–O).

LC-MS (ESI/positive): 923.5833 (MH^+). Single Mass Analysis: found 923.5784 ($C_{56}H_{79}N_2O_9$), calculated 923.5786 (-0.2 PPM)

^{1}H NMR: (500 MHz, $CDCl_3$) δ = 5.33 (m, 1H, 15'-H), 4.88 (dd, $J_{16'-17'}$ = 7.2 Hz, $J_{16'-15'}$ = 2.8 Hz, 1H, 16'-H), 4.39 (tr, J_{16-17} = J_{16-15} = 7.2 Hz, 1H, 16-H), 4.05 (dd, J_{15-16} = 7.2 Hz, J_{15-14} = 0.8 Hz, 1H, 15-H), 3.75 (s, 3H, 28-H), 3.72 (s, 3H, 28'-H), 3.35-3.52 (m, 4H, 26a/ 26'a/26b/26'b-H), 2.78-2.88 (m, 2H, 4'a/4'b-H), 2.32-2.70 (m, 7H, 1a/1'a/1b/ 1'b/4a/ 4b/11-H), 2.25 (m, 1H, 17-H), 1.24 (s, 3H, 18'-H), 0.97 (d, J_{21-20} = 6.7 Hz, 3H, 21-H), 0.95 (d, $J_{21'-20}$ = 6.8 Hz, 3H, 21'-H), 0.83 (s, 3H, 18-H), 0.82 (s, 3H, 19'-H), 0.80, 0.79 (2d, J_{27-25} = $J_{27'-25'}$ = 6.5 Hz, 6H, 27/27'-H), 0.78 (s, 3H, 19-H). **^{13}C NMR**: (125 MHz, $CDCl_3$) δ = 175.0 (C, 12-C), 174.6 (C, 12'-C), 164.7 (C, 14'-C), 148.5, 148.4, 148.2, 148.1 (all C, pyrazine-C), 113.9 (CH, 15'-C), 109.4 (C, 22'-C), 107.6 (C, 22-C), 87.9 (CH, 16'-C), 81.9 (CH, 16-C), 71.9 (CH, 15-C), 67.3, 66.8 (both CH_2, 26/26'-C), 61.1 (CH, 9-C), 58.8 (CH, 9'-C), 56.9 (C, 13'-C), 56.5 (CH, 17'-C), 55.8 (CH), 55.6 (CH), 54.6 (CH, 17-C), 53.3 (C, 13-C), 52.6 (CH_3, 28-C), 52.4 (CH, 11'-C), 51.9 (CH_3, 28'-C), 46.1 (CH), 44.6, 43.2 (both CH_2, 1/1'-C), 42.8 (CH, 20'-C), 41.8 (CH), 40.5 (CH), 38.5 (CH), 37.2 (CH, 8'-C), 36.1 (C, 10'-C), 35.6 (C, 10-C), 35.0 (CH_2, 4'-C), 32.0 (CH_2, 4-C), 31.8 (CH_2), 31.1 (CH_2), 31.0 (CH_2), 30.3, 30.2 (both CH), 28.9 (CH_3, 18'-C), 28.7 (CH_2), 28.6 (CH_2), 28.5 (CH_2), 27.9 (CH_2), 27.8 (CH_2), 25.5 (CH_3, 18-C), 17.2, 17.1 (both CH_3, 27/27'-C), 14.0, 13.9 (both CH_3, 21/21'-C), 12.0, 11.6 (both CH_3, 19/19'-C).