Supporting Information

Discovery of Hypoiodite-Mediated Aminyl Radical Cyclization Lacking a Nitrogen Radical-Stabilizing Group: Application to Synthesis of an Oxazaspiroketal-containing Cephalostatin Analog

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General Methods

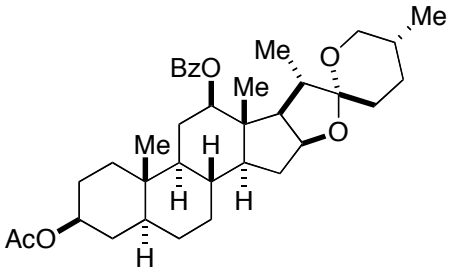
Reagents, such as triethylsilane, borontrifluoride etherate, iodobenzene diacetate, purchased from Aldrich Chemical Company Inc., were used as received. Acetonitrile, methylene chloride, pyridine, triethylamine, and *N*,*N*-diisopropylamine were distilled from calcium hydride: Methanol was distilled from magnesium turnings: THF was distilled from Na/benzoquinone. *N*-Bromosuccinimide was recrystallized from boiling water. Sodium sulfate (Na_2SO_4) was anhydrous. All chromatographic and workup solvents were distilled.

Unless otherwise indicated, all reactions were carried out under a positive pressure of argon in anhydrous solvents and the reaction flasks were fitted with rubber septa for the introduction of substrates and reagents via syringe. Progress of reactions was monitored by thin layer chromatography (TLC) in comparison with the starting materials. All TLC analyses were carried out on Merck Silica Gel 60 F254 TLC plates, thickness of 0.25 mm. The plates were visualized by ultraviolet illumination at 254 nm and immersion in visualizing solution. The two commonly employed TLC visualizing solutions were: (i) *p*-anisaldehyde solution (1350 mL absolute ethanol, 50 mL concentrated H_2SO_4, 37 mL *p*-anisaldehyde), and (ii) permanganate solution (weight percents of 1 % $KMnO_4$ and 2 % Na_2CO_3 in water).

Analytical samples were obtained from flash silica gel chromatography, using silica gel of 230-400. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker AM 500 (500 MHz). NMR spectra were determined in chloroform-d_1 ($CDCl_3$), DMSO-d_6 or

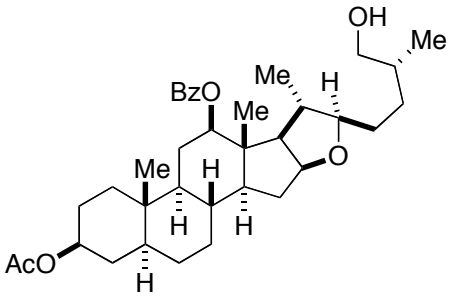
methanol-d_4 (CD_3OD) solution and are reported in parts per million (ppm) from the residual chloroform (7.24 ppm and 77.0 ppm) and benzene (7.16 ppm and 128.39 ppm) standard, respectively. Peak multiplicates in ^{1}H-NMR spectra, when reported, are abbreviated as s (singlet), d (doublet), t (triplet), m (multiplet), and/or ap (apparent) and/or br (broad). Mass spectra were all obtained on either a JEOL AX-505 or a JEOL SX-102. All UV spectra and absorbances were collected with an HP 8452 uv-vis spectrophotometer with a photo-diode array detector. HPLC analyses were carried out on an HP 1090 Liquid Chromatograph equipped with a Beckman C18 Reversed-phase column.

5α-Spirostan-3β-12β-diyl 3-acetate 12-benzoate (7)



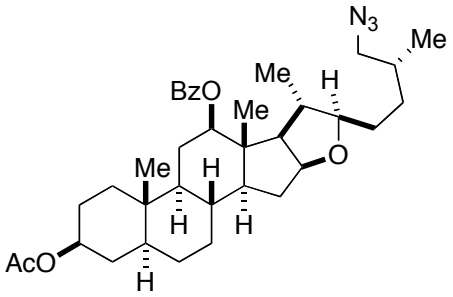
^{1}H NMR (500 MHz, $CDCl_3$) δ 8.05-7.46 (5H, m, aromatic H), 4.84 (1H, dd, *J* = 4.4, 11.0 Hz, C12-H), 4.71 (1H, m, C3-H), 4.45 (1H, dd, *J* = 7.4, 14.2 Hz, C16-H), 3.50-3.27 (2H, m, C26-H), 2.02 (3H, s, C3-OAc), 1.03 (3H, s, C18-CH_3), 0.88 (3H, s, C19-CH_3), 0.79 (3H, d, *J* = 5.9 Hz, C27-CH_3), 0.74 (3H, d, *J* = 6.7 Hz, C21-CH_3); ^{13}C NMR (75 MHz, $CDCl_3$) δ 170.5, 165.6, 132.8, 130.7, 129.4, 128.4, 128.3, 109.2, 81.9, 80.4, 73.3, 66.7, 61.5, 54.8, 52.6, 44.9, 44.4, 42.1, 36.5, 35.5, 34.1, 33.8, 31.6, 31.4, 31.1, 30.1, 28.7, 28.3, 27.2, 26.8, 21.3, 17.0, 14.0, 13.6, 12.1, 11.8.

3β-Acetoxy-12β-benzyloxy-5α-furostan-26-ol (8)



To a CH_2Cl_2 (100 mL) solution of sprioketal **7** (6.04 g, 10.4 mmol) and triethylsilane (2.15 mL, 13.5 mmol, 1.3 molar equiv), was added dropwise borontrifluoride etherate (1.90g, 13.5 mmol, 1.3 molar equiv) over 2 hours at 0 ^oC, and the resulting mixture was stirred at 0 ^oC for additional 8 hours. The reaction mixture was diluted with EtOAc, neutralized with saturated aqueous sodium bicarbonate, washed with brine, dried over anhydrous sodium sulfate, concentrated *in vacuo*, and subjected to silica gel chromatography to give a primary alcohol **8** (5.64 g, 9.32 mmol, 90%). ^1H NMR (500 MHz, $CDCl_3$) δ 8.05-7.46 (5H, m, aromatic H), 4.81 (1H, dd, J = 4.6, 14.4 Hz, C12-H), 4.67 (1H, m, C3-H), 4.29 (1H, dd, J = 7.4, 14.2 Hz, C16-H), 3.45 (2H, m, C26-H), 3.27 (1H, dt, J = 3.8, 8.4, C22-H), 1.99 (3H, s, C3-OAc), 1.01 (3H, s, C18-CH_3), 0.88 (3H, d, J = 5.9 Hz, C27-CH_3), 0.84 (3H, s, C19-CH_3), 0.72 (3H, d, J = 6.7 Hz, C21-CH_3);^{13} C NMR (75 MHz, $CDCl_3$) δ 170.5, 165.7, 132.8, 130.6, 129.3, 128.3, 90.2, 82.8, 81.9, 73.3, 67.8, 64.0, 55.2, 52.7, 45.1, 44.4, 38.7, 36.5, 35.6, 35.5, 34.2, 33.7, 31.5, 31.4, 30.3, 30.1, 28.2, 27.2, 26.7, 21.3, 17.8, 16.5, 12.0, 11.9; MS (M + H) 581; HRMS calculated $C_{36}H_{52}O_6$ 580.3764, found 580.3753.

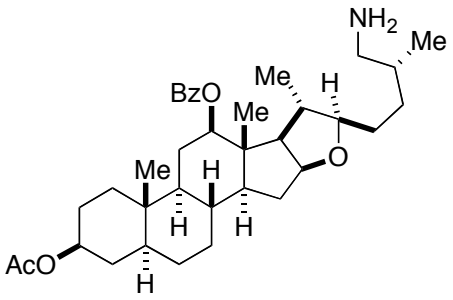
3β-Acetoxy-12β-benzyloxy-5α-furostan-26-azide (9)



To a pyridine solution of primary alcohol (5.67 g, 9.77 mmol) was added dropwise methanesulfonyl chloride (1.60 g, 14.1 mmol, 1.5 molar equiv) at 0 ^oC, and the resulting mixture was further stirred for 2 hours at 0 ^oC. After completion of the reaction, the solvent was evaporated under reduced pressure, washed with 1N HCl, dried over anhydrous sodium sulfate, and subjected to silica gel chromatography to give the corresponding mesylate (6.34 g, 9.63 mmol, 99%). To the mesylate (6.31 g, 9.58 mmol) in DMF at 60 ^oC was added excess sodium azide (3.15g, 48.4 mmol, 5 molar equiv) and the mixture was stirred for 6 hours at the same temperature. The reaction mixture was

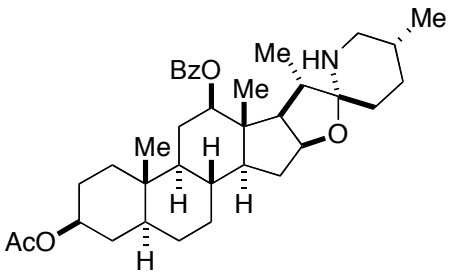
diluted with EtOAc, washed with water and brine, concentrated under reduced pressure, and subjected to silica gel chromatography to provide azide **9** (5.94 g, 91%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.05-7.46 (5H, m, aromatic H), 4.80 (1H, dd, *J* = 4.7, 11.3 Hz, C12-H), 4.67 (1H, m, C3-H), 4.29 (1H, dd, *J* = 5.2, 12.8 Hz, C16-H), 3.23 (1H, m, C22-H), 3.21 (1H, dd, *J* = 5.8, 12.0, C26-H), 3.09 (1H, dd, *J* = 6.8, 12.1, C26-H), 1.99 (3H, s, C3-OAc), 1.00 (3H, s, C18-CH_3), 0.93 (3H, d, *J* = 6.5 Hz, C27-CH_3), 0.84 (3H, s, C19-CH_3), 0.72 (3H, d, *J* = 6.7 Hz, C21-CH_3);^{13}C NMR (75 MHz, $CDCl_3$) δ 170.5, 165.7, 132.8, 130.7, 129.4, 128.3, 90.0, 82.9, 82.0, 73.8, 67.8, 64.1, 57.7, 55.2, 52.7, 45.1, 44.5, 38.8, 36.6, 35.6, 34.3, 33.8, 31.6, 31.5, 30.7, 28.3, 27.3, 26.8, 21.4, 17.9, 17.6, 12.1, 12.0; MS (M + H) 606; HRMS calculated $C_{36}H_{51}N_3O_5$ 605.3829, found 605.3810.

3β-Acetoxy-12β-benzyloxy-5α-furostan-26-amine (10)



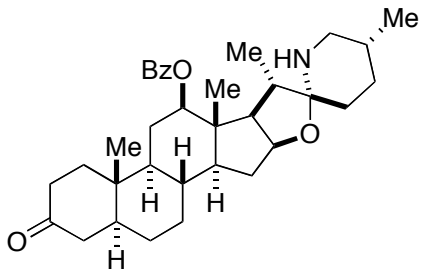
To a steroidal azide **9** (3.10 g, 5.12 mmol) in 50 mL THF, was added water (0.92 mL, 51.2 mmol, 10 molar equiv) and triphenylphosphine (4.02 g, 15.4 mmol, 3 molar equiv), and the resulting mixture was stirred for 18 hours at 25 ^{o}C. The reaction mixture was then concentrated under reduced pressure, and the residue was subjected to silica gel chromatography to give primary amine **10** (2.82 g, 4.87 mmol, 95%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.05-7.46 (5H, m, aromatic H), 4.81 (1H, dd, *J* = 4.4, 11.1 Hz, C12-H), 4.67 (1H, m, C3-H), 4.29 (1H, dd, *J* = 6.0, 11.5 Hz, C16-H), 3.26 (1H, dt, *J* = 3.8, 8.2, C22-H), 2.65-2.41 (2H, m, C26-H), 1.99 (3H, s, C3-OAc), 1.00 (3H, s, C18-CH_3), 0.87 (3H, d, *J* = 6.5 Hz, C27-CH_3), 0.84 (3H, s, C19-CH_3), 0.72 (3H, d, *J* = 6.5 Hz, C21-CH_3); ^{13}C NMR (75 MHz, $CDCl_3$) δ 170.5, 165.9, 132.8, 130.7, 129.4, 128.4, 90.3, 82.9, 82.0, 64.1, 57.7, 55.3, 52.8, 45.2, 44.5, 38.8, 36.6, 35.6, 34.3, 33.8, 31.6, 31.6, 31.2, 30.9, 28.3, 27.3, 26.8, 21.4, 17.9, 17.3, 12.0; MS (M + H) 580; HRMS calculated $C_{36}H_{53}NO_5$ 579.3924, found 579.3899.

5α,22α-Spirosolan-3β-12β-diyl 3-acetate 12-benzoate (11)



To a CH_2Cl_2 solution of iodobenzene diactate (21 mg, 0.065 mmol) and iodine (17 mg, 0.065 mmol) was added primary amine **10** (25 mg, 0.043 mmol) was added in one portion and the resulting mixture was stirred at 0 ^oC for 10 minutes. The reaction mixture was diluted with EtOAc, washed with saturated aqueous sodium thiosulfate and brine, concentrated, and subjected to silica gel chromatography to yield oxazaspiroketal **11** (22 mg, 0.038 mmol, 90%). ^1H NMR (500 MHz, $CDCl_3$) δ 8.02-7.42 (5H, m, aromatic H), 4.80 (1H, dd, *J* = 4.6, 11.1 Hz, C12-H), 4.63 (1H, m, C3-H), 4.27 (1H, dd, *J* = 6.9, 13.6 Hz, C16-H), 2.65-2.45 (2H, m, C26-H), 1.98 (3H, s, C3-OAc), 1.00 (3H, s, C18-CH_3), 0.84 (3H, s, C19-CH_3), 0.80 (3H, d, *J* = 6.2 Hz, C27-CH_3), 0.65 (3H, d, *J* = 6.5 Hz, C21-CH_3); ^{13}C NMR (75 MHz, $CDCl_3$) δ 170.6, 165.8, 132.9, 130.7, 129.5, 128.4, 98.3, 81.9, 78.5, 73.4, 62.1, 54.8, 52.7, 47.6, 45.2, 44.5, 41.8, 36.6, 35.6, 34.2, 34.1, 33.8, 31.6, 31.5, 31.3, 30.3, 28.3, 27.3, 26.9, 22.6, 21.4, 19.2, 14.3, 12.1, 12.0; MS (M + H) 580; HRMS calculated $C_{36}H_{51}NO_5$ 577.3755, found 577.3775.

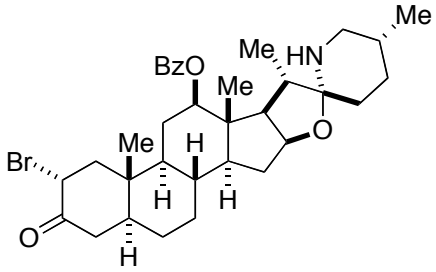
5α,22α-Spirosolan-3-oxo-12β-benzoate (17)



To a well stirred solution of steroidal acetate **11** (1.19 g, 2.06 mmol) in 5:1 MeOH/H_2O (20 mL/4 mL) at 25 ^oC was added potassium carbonate (2.84 g, 20.6 mmol). After 12h, the reaction mixture was quenched with saturated NH_4Cl and the solvent was evaporated under reduced pressure to afford crude product. Extraction with EtOAc, washing with

brine, drying over anhydrous sodium sulfate, and evaporation under reduced pressure afforded the crude product mixture, which was subjected to silica gel chromatography to give the corresponding alcohol (1.08g, 97%). To a CH_2Cl_2 solution of the secondary alcohol (26 mg) containing 4Å molecular sieves and NMO, was added TPAP in one portion. After 0.5h, the reaction mixture was quenched with saturated sodium thiosulfate and extracted with EtOAc. Concentration and silica gel chromatography provided C3 ketone **17** (24 mg, 94%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.02-7.38 (5H, m, aromatic H), 4.81 (1H, dd, *J* = 4.7, 11.1 Hz, C12-H), 4.63 (1H, m, C3-H), 4.27 (1H, dd, *J* = 4.4, 13.8 Hz, C16-H), 2.62-2.50 (2H, m, C26-H), 1.03 (3H, s, C18-CH_3), 1.01 (3H, s, C19-CH_3), 0.80 (3H, d, *J* = 5.5 Hz, C27-CH_3), 0.66 (3H, d, *J* = 6.4 Hz, C21-CH_3);^{13}C NMR (75 MHz, $CDCl_3$) δ 211.3, 165.7, 133.0, 130.6, 129.5, 128.4, 98.3, 81.8, 78.4, 62.2, 54.7, 52.3, 47.6, 46.3, 45.2, 44.5, 41.8, 38.3, 37.8, 35.7, 34.2, 34.1, 33.8, 31.5, 31.4, 31.3, 30.3, 28.7, 27.1, 19.2, 14.3, 12.0, 11.3; MS (M + H) 534; HRMS calculated $C_{34}H_{47}NO_4$ 533.3505, found 533.3505.

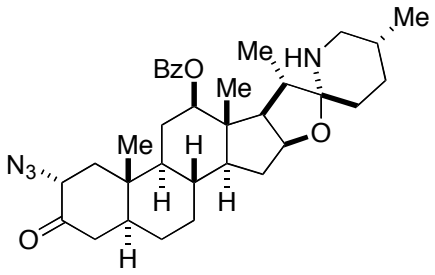
5α,22α-Spirosolan-2α-bromo-3-oxo-12β-benzoate (18)



To a solution of ketone **17** (37 mg, 0.069 mmol) in THF (7mL) at 0 ^{o}C was added HCl (100 μL, 1M solution in Et_2O) and then phenyltrimethyl ammonium tribromide (28 mg, 0.076 mmol) in one portion, and the resulting mixture was stirred at 0 ^{o}C for 1h. The reaction mixture was concentrated and subjected under silica gel chromatography to afford desired alpha-bromoketone **18** (41 mg). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.02-7.42 (5H, m, aromatic H), 4.81 (1H, dd, *J* = 4.2, 10.9 Hz, C12-H), 4.67 (1H, dd, *J* = 6.2, 13.1 Hz, C2-H), 4.29 (1H, m, C16-H), 2.65-2.45 (2H, m, C26-H), 1.09 (3H, s, C18-CH_3), 1.03 (3H, s, C19-CH_3), 0.80 (3H, d, *J* = 6.0 Hz, C27-CH_3), 0.67 (3H, d, *J* = 5.9 Hz, C21-CH_3); ^{13}C NMR (75 MHz, $CDCl_3$) δ 200.6, 165.7, 133.0, 130.4, 129.5, 128.4, 99.9, 98.3, 81.3, 78.4, 62.1, 54.4, 53.7, 52.0, 51.1, 47.5, 47.1, 45.2, 43.6, 41.8, 38.9, 34.1, 33.6, 31.4, 31.1,

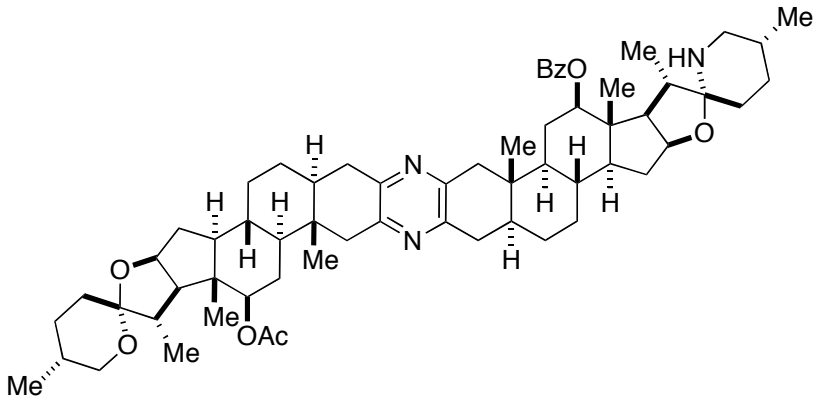
30.2, 28.1, 27.1, 19.2, 14.3, 13.1, 12.0, 11.9; MS (M + H) 612; HRMS calculated $C_{34}H_{46}BrNO_4$ 612.2688, found 612.2692.

5α,22α-Spirosolan-2α-azido-3-oxo-12β-benzoate (19)



A solution of bromoketone **18** (41 mg) in freshly dried CH_3NO_2 was cooled to 0 ^oC and $TMGN_3$ was added portionwise for 2min. The reaction was allowed to warm slowly to 25 ^oC during 6h of stirring and partitioned between EtOAc and brine, and organic layer was dried over anhydrous Na_2SO_4 and concentrated to give crude mixture, which was subjected to silica gel chromatography to give azidoketone **19**. (35 mg, 63% over two steps) ^1H NMR (500 MHz, $CDCl_3$) δ 8.02-7.42 (5H, m, aromatic H), 4.81 (1H, dd, *J* = 4.6, 11.0 Hz, C12-H), 4.29 (1H, m, C16-H), 4.67 (1H, dd, *J* = 6.3, 13.0 Hz, C2-H), 2.65-2.48 (2H, m, C26-H), 1.09 (3H, s, C18-CH_3), 1.04 (3H, s, C19-CH_3), 0.80 (3H, d, *J* = 6.1 Hz, C27-CH_3), 0.67 (3H, d, *J* = 6.3 Hz, C21-CH_3); ^{13}C NMR (75 MHz, $CDCl_3$) δ 204.6, 165.7, 133.0, 130.4, 129.5, 128.4, 99.9, 98.4, 81.3, 78.4, 63.7, 62.2, 54.4, 53.7, 52.0, 51.1, 47.5, 47.3, 45.2, 43.5, 41.8, 36.9, 34.1, 33.6, 31.4, 30.2, 28.1, 29.6, 28.2, 27.1, 19.2, 14.3, 12.5, 12.0; MS (M + H) 575; HRMS calculated $C_{34}H_{46}N_4O_4$ 575.3597, found 575.3596.

Oxazasprioketal-containing cephalostatin analog (5)



To a solution of azidoketone **19** (5 mg) and alpha-aminomethoxime **20** (4 mg) in 10 mL benzene was added dichlorodibutylstannane (cat.) and polyvinyl pyridine. The reaction flask was equipped with a Dean-Stark trap, and the mixture was heated at reflux for 3h (2mL of fresh benzene was added twice to maintain the solvent level in the reaction vessel), at which time TLC indicated no remaining azidoketone **19**. The reaction mixture was cooled and filtered, and the solids were washed with CH_2Cl_2. Evaporation of the filtrate and silica gel chromatography of the residue gave the target cephalostatin analog **5** (4.2 mg, 76%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.02-7.42 (5H, m, aromatic H), 4.81 (1H, dd, *J* = 4.3, 10.9 Hz, North C12-H), 4.54 (1H, dd, *J* = 4.6, 11.0 Hz, South C12-H), 4.41-4.25 (2H, m, South and North C16-H), 3.44-3.30 (2H, m, South C26-H), 2.65-2.45 (2H, m, North C26-H), 2.01 (3H, s, South C12-OAc), 1.04 (3H, s, C18-CH_3), 0.90 (3H, d, *J* = 4.7 Hz, CH_3), 0.85 (3H, s, CH_3), 0.80 (3H, d, *J* = 6.0 Hz, CH_3), 0.77 (3H, d, *J* = 5.9 Hz, CH_3);^{13}C NMR (75 MHz, $CDCl_3$) δ 170.2, 165.9, 148.6, 148.4, 130.1, 129.6, 129.4, 128.7, 128.5, 109.0, 98.4, 80.5, 79.1, 66.6, 62.3, 61.7, 54.5, 52.6, 47.8, 45.4, 44.5, 42.0, 34.5, 33.8, 31.0, 30.4, 29.0, 28.4, 26.8, 19.0, 16.7, 15.1, 14.0, 12.1, 11.8; MS (M + H) 998, HRMS calculated $C_{63}H_{87}N_3O_7$ 997.6592, found 997.6577.

STANDARD 1H OBSERVE

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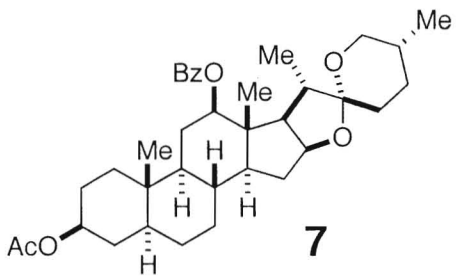

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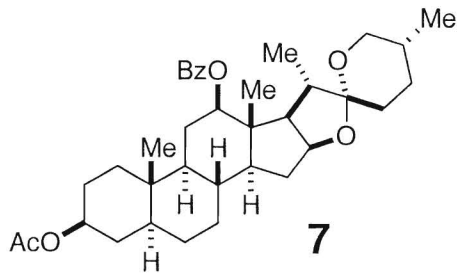

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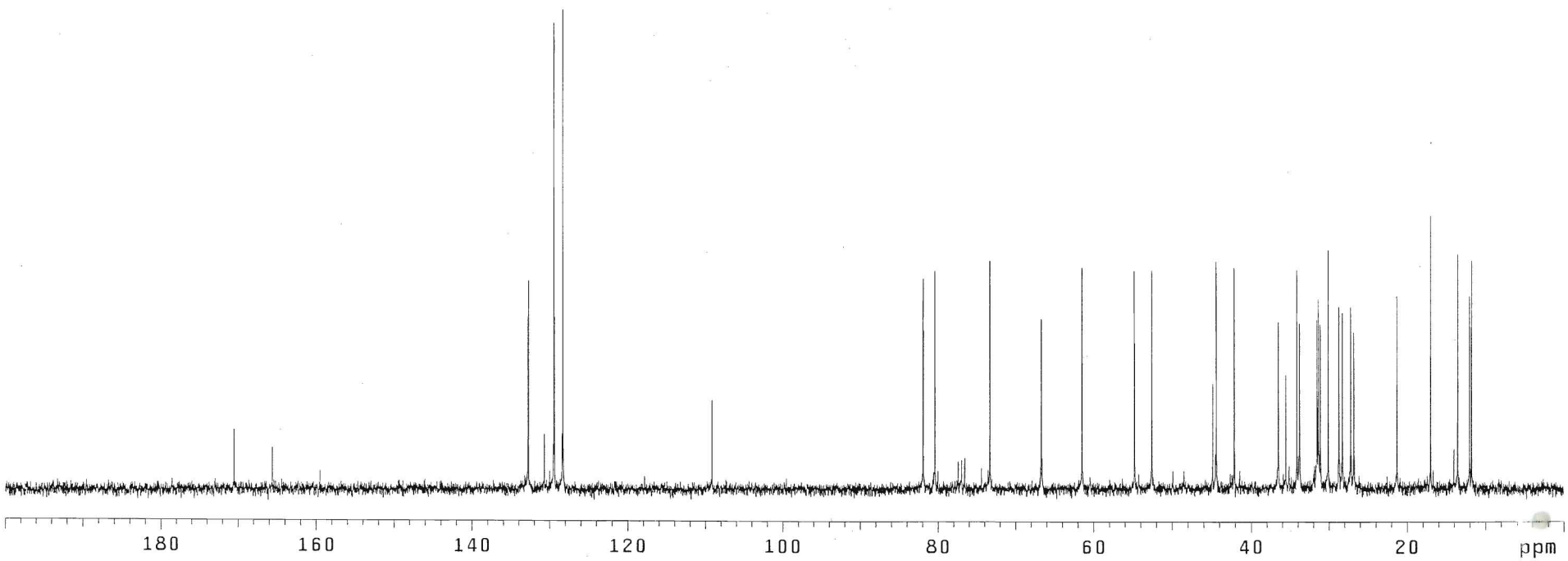

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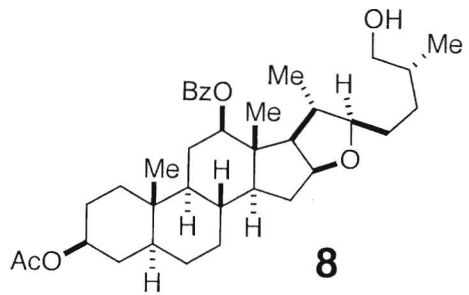

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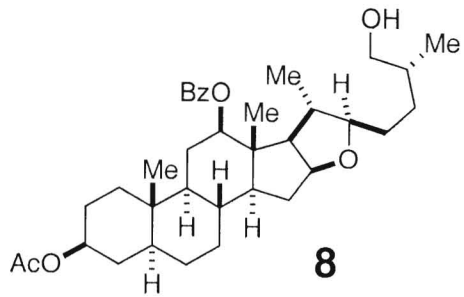

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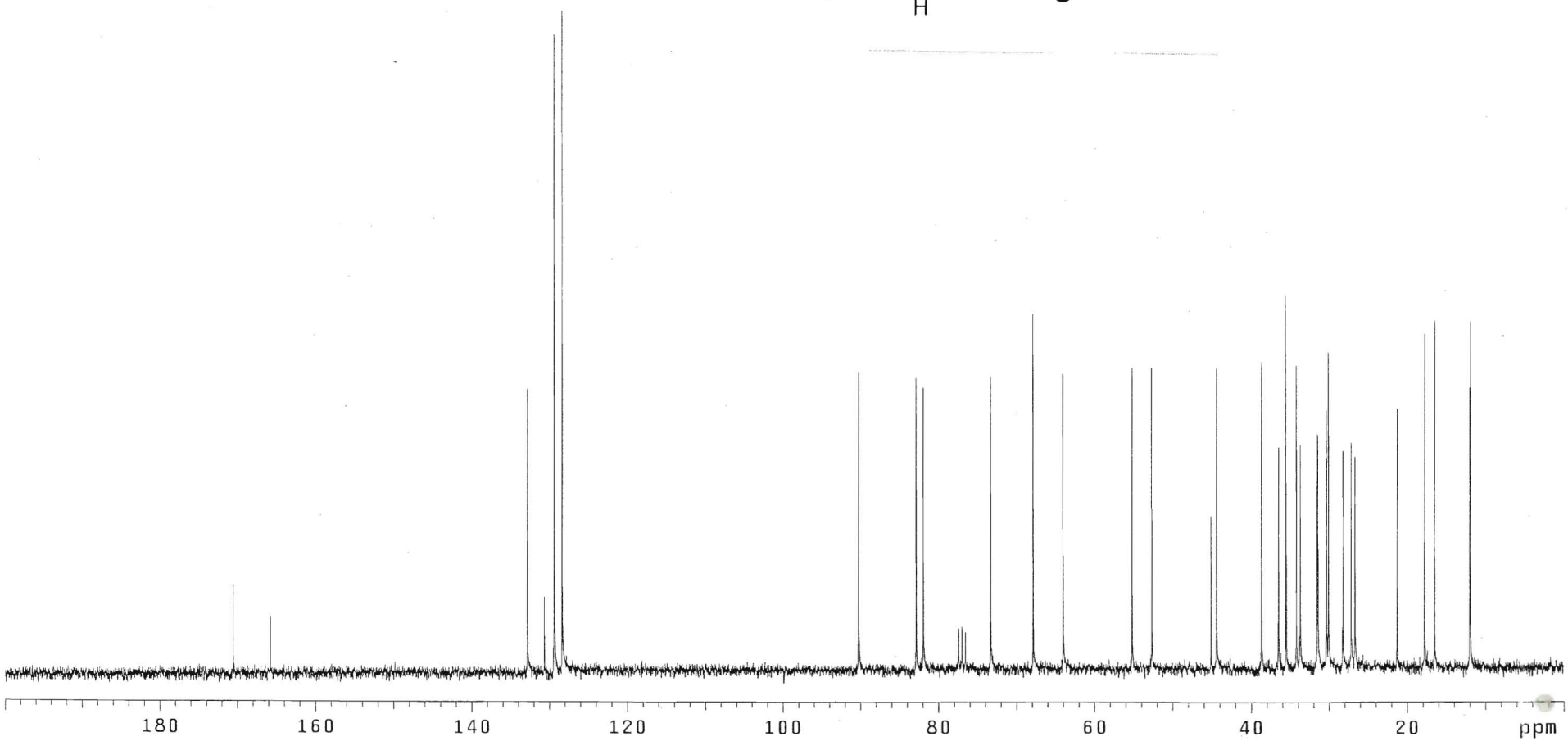

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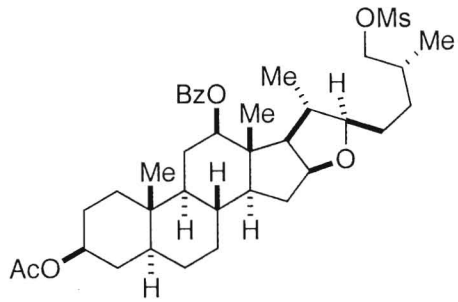
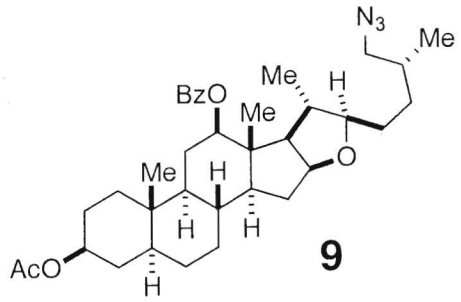


106-1-341

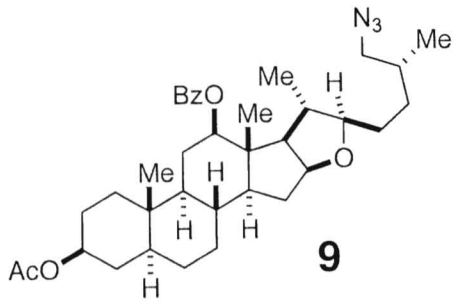
Pulse Sequence: s2pul



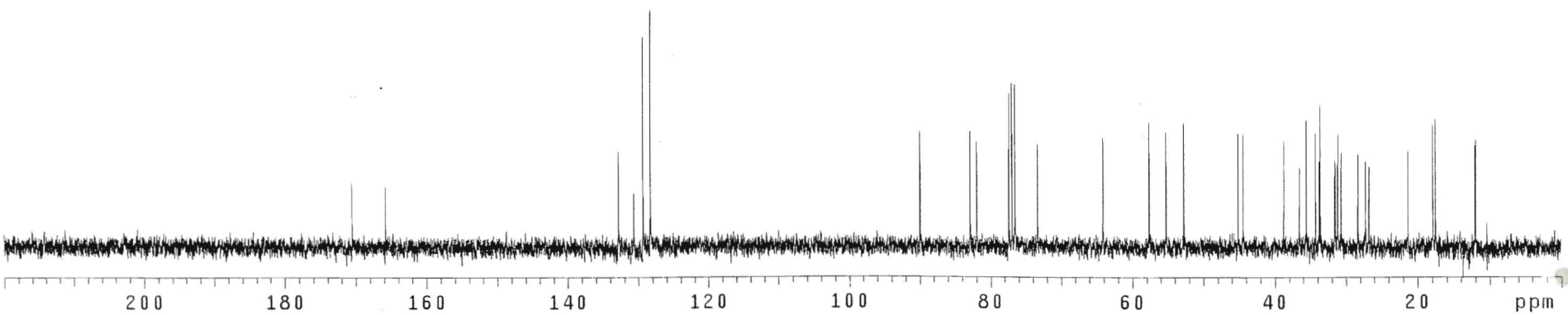
9

106-1-341-13C

Pulse Sequence: s2pul

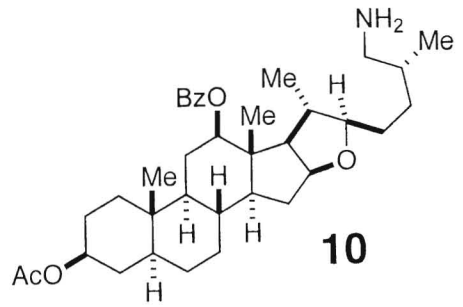


9



106-1-34

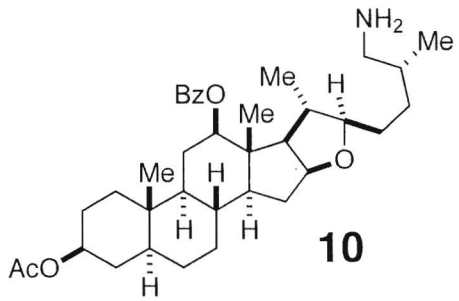
Pulse Sequence: s2pul



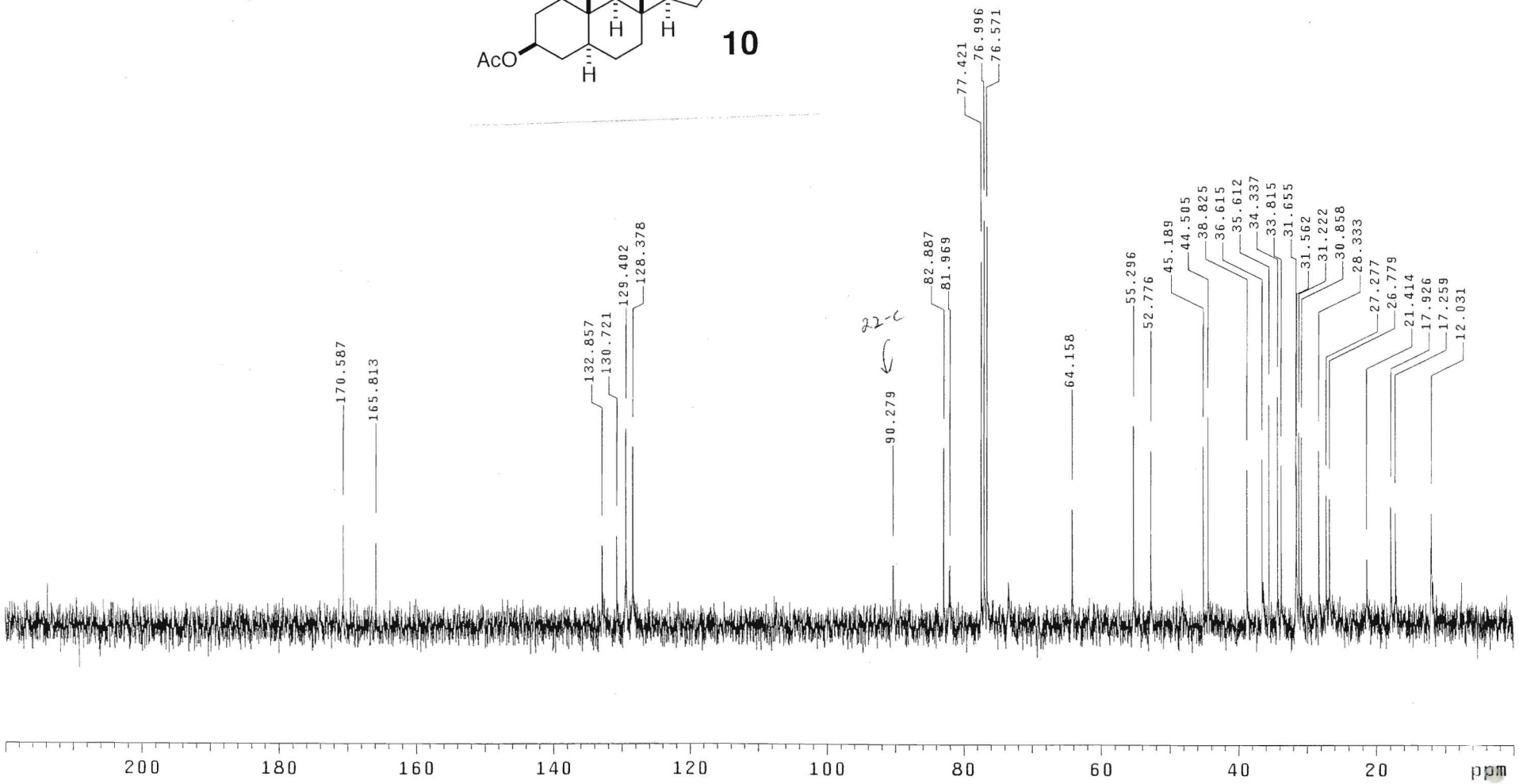
10

106-1-343-13C

Pulse Sequence: s2pul

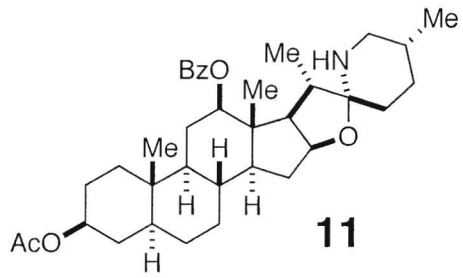


10

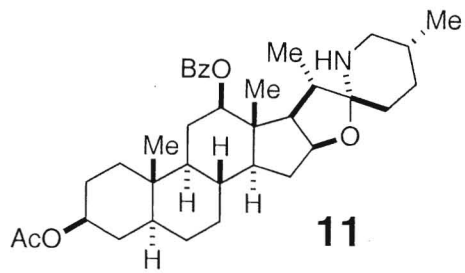


106-1-35

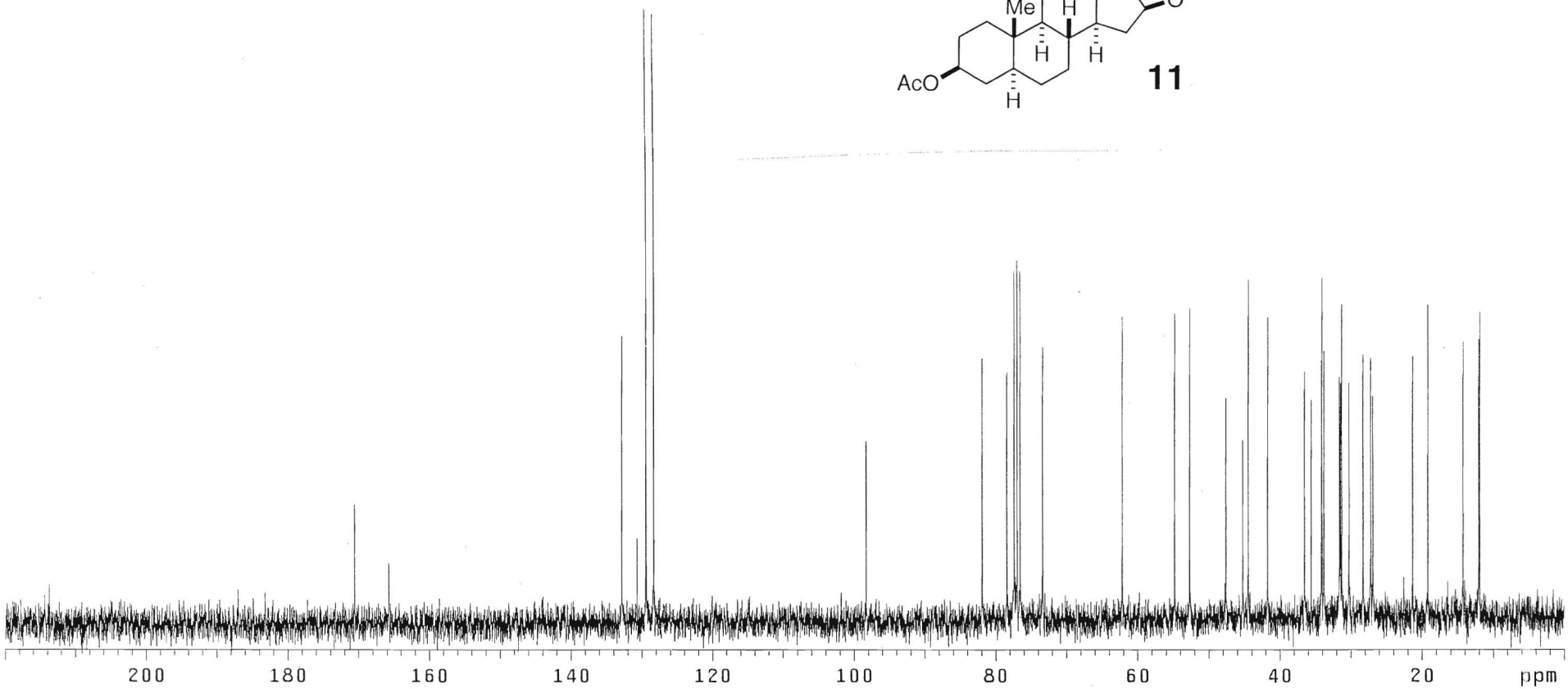
Pulse Sequence: s2pul



11

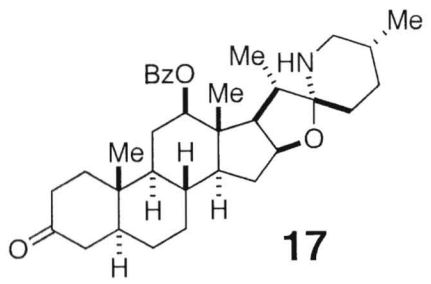

Me
Me
HN
BzO
Me
Me
H
O
H
H
AcO
H

11


200
180
160
140
120
100
80
60
40
20
ppm

106-1-383-1

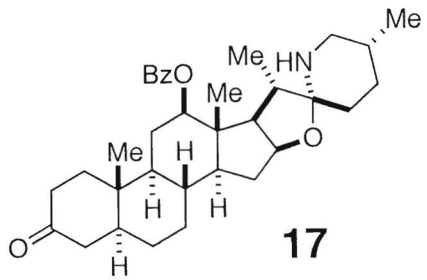
Pulse Sequence: s2pul



17

106-1-383-1-13C

Pulse Sequence: s2pul



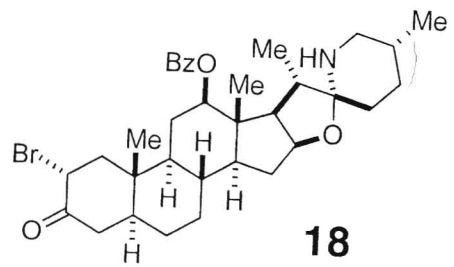
17

211.300
165.748
132.958
130.583
129.479
128.406
98.327
81.718
78.408
77.425
77.000
76.575
62.207
54.702
52.319
47.605
46.359
45.242
44.453
41.778
38.254
37.890
35.754
34.188
34.062
31.497
31.355
31.315
30.299
28.673
27.099
19.245
14.313
11.987
11.384

200 180 160 140 120 100 80 60 40 20 ppm

106-2-13-crude

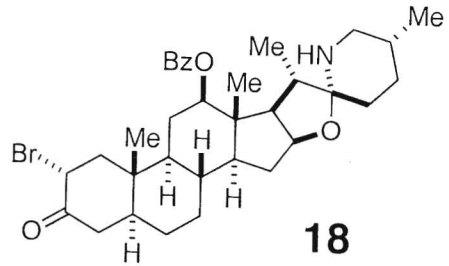
Pulse Sequence: s2pul



18

106-2-41-crude-13C

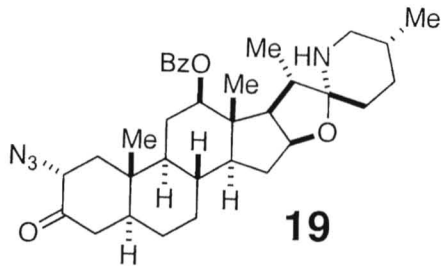
Pulse Sequence: s2pul



18

106-2-15

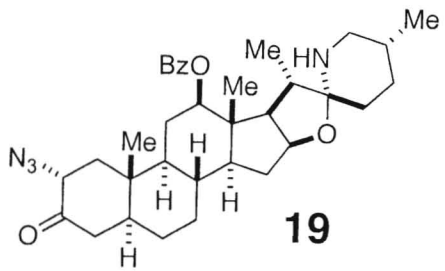
Pulse Sequence: s2pul



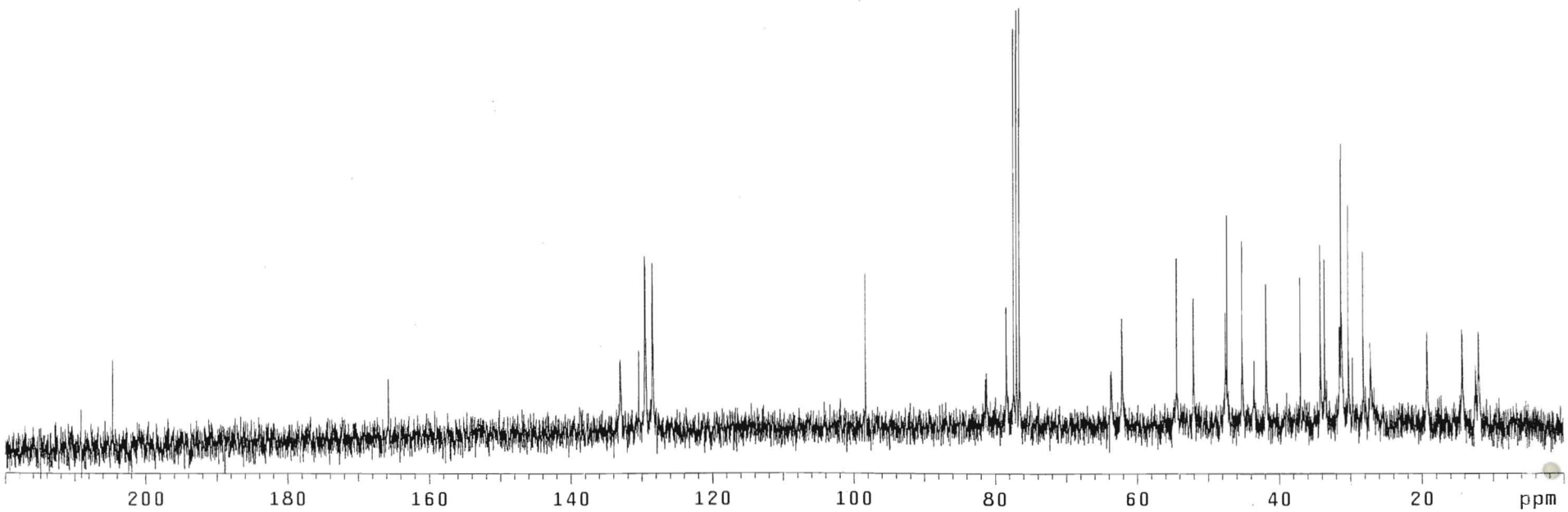
19

106-2-15-13C

Pulse Sequence: s2pul

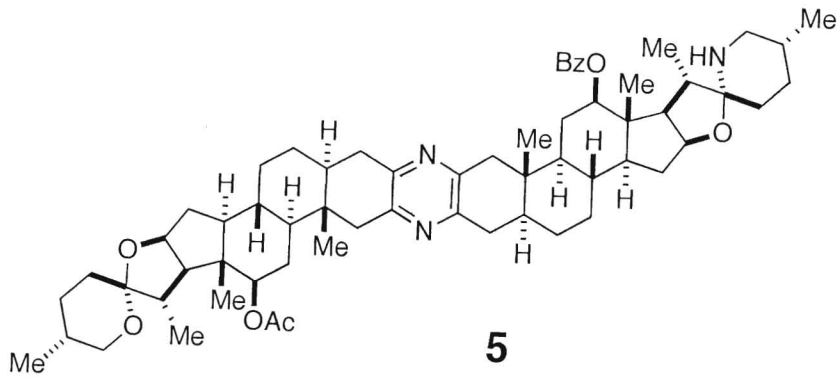


19



106-2-31-1

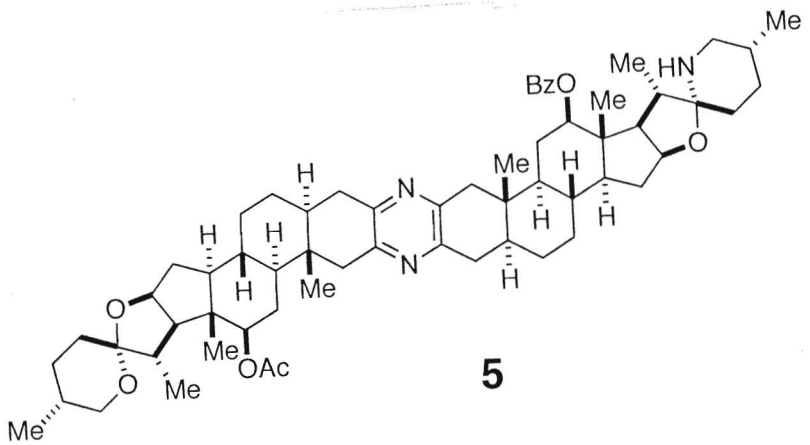
Pulse Sequence: s2pul



5

106-2-57-13C

Pulse Sequence: s2pul



5