

SYNTHETIC APPROACH TO THE ANTITUMOUR ANTIBIOTICS NEOTHRAMYCINS

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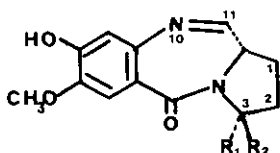
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Abstract - A new simple synthetic route to the pyrrolo[1,4]-benzodiazepines (5) related to the neothramycins (1) and (2) through the methyl imidate (7) is described.

Neothramycins A (1) and B (2) were isolated from *Streptomyces* n° MC 916-C4.¹ Their structures were established and confirmed through a synthesis by Umezawa and co-workers.² They belong to the pyrrolo[1,4]benzodiazepine group of antitumour antibiotics such as anthramycin,^{3,4} tomaymycin,⁵ sibiromycin,⁶ mazethramycin,⁷ chicamycin⁸ and are known to be less toxic than other members of this group.⁹ The characteristic structural feature of the neothramycins (1) and (2) is the presence of a hydroxyl group on the carbon 3 which can be a site of the reaction with DNA.^{10,11}

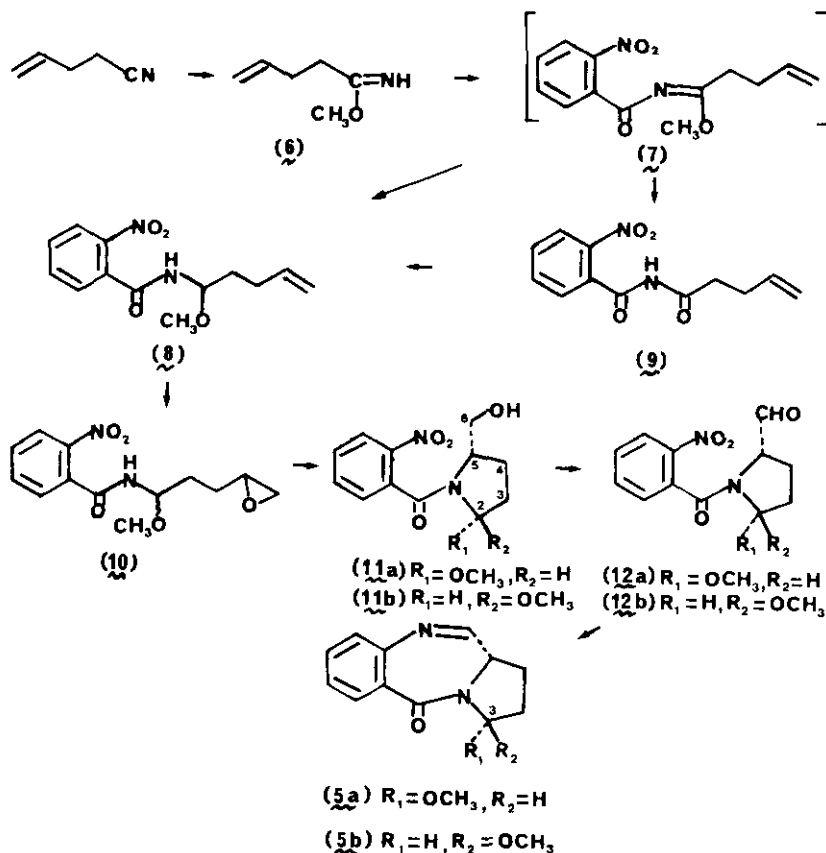


- (1) $R_1 = OH, R_2 = H$
 (2) $R_1 = H, R_2 = OH$
 (3) $R_1 = OCH_3, R_2 = H$
 (4) $R_1 = H, R_2 = OCH_3$

As the O-methylnethramycins (3) and (4) can be easily converted to the neothramycins (1) and (2) themselves,¹ at an early stage of our synthetic approach toward the model target compounds (5), we introduced a methoxy group on the carbon which should ultimately become the C3 of the pyrrolo[1,4]benzodiazepine framework.

In one of the reactions sequence explored the methyl imidate (6) prepared from

1-cyano-3-butene, led after treatment with o-nitrobenzoyl chloride to the methyl N-(o-nitrobenzoyl)imidate (7). This compound was reduced without isolation with sodium borohydride in methanol, according to the method described in the synthesis of pederine,¹² affording the N-(1-methoxy-4-pentene)-o-nitrobenzamide (8) (50-55%). The acyclic imide (9) also obtained in this reaction (ca. 15%) resulted probably from partial hydrolysis of (7) and gave (8) by treatment with sodium borohydride in the presence of methanolic hydrochloric acid following the conditions used for the reduction of cyclic imides.¹³



The elaboration of the pyrrolidine ring was achieved through epoxidation (MCPBA) of the N-(1-methoxy-4-pentene)-o-nitrobenzamide (8) to (10) (100% yield as diastereoisomeric mixture), followed by an intramolecular cyclisation reaction in the presence of sodium hydride. The two diastereoisomeric (11a) and (11b)

thus formed were isolated in 24% and 46% yield, respectively. The analysis of PMR spectra of (11) along with the appropriate decoupling experiments supported the indicated relative configurations.

The isomer (11a) was oxidized with dimethylsulfoxide and the pyridine-SO₂ complex to the aldehyde (12a) (95%) which was reduced¹⁴ to give the pyrrolo[1,4]-benzodiazepine (5a) (62%). In the same way the primary alcohol (11b) led to the diastereoisomer (5b) in 75% yield.¹⁵

Application of this pyrrolo[1,4]benzodiazepine route to a synthesis of the neothramycins (1) and (2) and other analogs is now in progress in our laboratory.

EXPERIMENTAL

Melting points were measured with a Kofler apparatus and are corrected. IR spectra (CHCl₃) were recorded on a Perkin-Elmer 257 spectrometer and uv spectra (CH₃OH) on a Jobin-Yvon Duospec 203 spectrometer. ¹H NMR spectra were obtained (if not specified in CDCl₃) on a Bruker WM400, WM200 or Varian T60 spectrometers with tetramethylsilane as the internal reference, coupling constants, J, are given in hertz; s, d, t, and m indicate singlet, doublet, triplet and multiplet, respectively. ¹³C NMR spectra (CDCl₃, δ = 0 ppm, TMS) were recorded on a Bruker WM200 spectrometer. Mass spectra were measured on a MS50.

Preparation of N-(1-methoxy-4-pentene)-o-nitrobenzamide (8). To a stirred solution of 1-cyano-3-butene (8.10 g, 0.1 mol) in dry methanol (4 ml) at 0°C was bubbled anhydrous HCl (4.75 g, 0.13 mol). The reaction medium was kept at 0°C during 72 h and then at -50°C before the filtration of the methylimidate (6) hydrochloride. The crystals (11.3 g, 75%) were washed with cold dry ether. ¹H NMR (400 MHz): 5.62 (tdd, 1H, J_{4,5a} = 18, J_{4,5b} = 11, J_{3,7} = 7, C₄-H), 4.95 (d, 1H, J = 18, C₅-H_a, trans), 4.91 (d, 1H, J = 11, C₅-H_b, cis), 4.12 (s, 3H, OCH₃), 2.72 (t, 2H, C₂-H), 2.32 (dd, 2H, C₃-H). ¹³C NMR: 179.5 (C₁), 134.2 (C₄), 117.2 (C₅), 60.5 (OCH₃), 32.1 and 29.3 (C₂ and C₃).

To a stirred solution of the methylimidate (6) hydrochloride (6.0 g, 0.04 mol) in dry dichloromethane (75 ml) was added triethylamine (15 ml). After being stirred at room temperature for 20 min was added dropwise a solution of o-nitrobenzoyl chloride (9.30 g, 0.05 mol) in dry dichloromethane (35 ml). The reaction medium was stirred at 20°C for 75 min before evaporation of the solvent under vacuum. The residue in solution in dry methanol (175 ml) was reduced with sodium borohydride (5.0 g, 0.13 mol). The mixture was stirred at 20°C for 1 h before dilution with water and extraction with dichloromethane. The usual

workup afforded the crude product (11.1 g) which was chromatographed on silica gel (hexane-ethylacetate 1-1) to obtain compounds (8) and (9).

N-(1-methoxy-4-pentene)-o-nitrobenzamide (8) (5.6 g, 53%), mp 59.60°C; ir : 3250, 3060, 2910, 1645, 1530, 1340, 1210, 1070, 905 cm^{-1} ; uv(λ_{max} nm(ϵ) : 213 (11400), 251 (5600) ; ms (m/z) : 263.1037 (M^{+} -H, $\text{C}_{13}\text{H}_{15}\text{N}_2\text{O}_4$, 0.7%), 233 (M^{+} -OCH₃, 21.5%), 209 (100%), 150. ^1H NMR (200 MHz) : 8.00 (dd, 1H, $J_o = 8$, $J_m = 2$) and 7.43 (dd, 1H, $J_o = 8$, $J_m = 2$) : C₃-H and C₆-H, 7.63 (ddd, 1H, $J = 8$, $J = 2$) and 7.53 (ddd, 1H, $J = 8$, $J = 2$) : C₄-H and C₅-H, 6.58 (broad d, 1H, $J = 10$, NH), 5.83 (tdd, 1H, $J_{4,5a} = 17$, $J_{4,5b} = 10$, $J_{3,4} = 7$, C₄-H), 5.23 (td, 1H, $J_{1,NH} = 10$, $J_{1,2} = 7$, C₁-H), 5.06 (dd, 1H, $J_{4,5a} = 17$, $J_{5a,5b} = 2$, C₅-H_a, trans), 5.00 (dd, 1H, $J_{4,5b} = 10$, $J_{5a,5b} = 2$, C₅-H_b, cis), 3.43 (s, 3H, OCH₃), 2.17 (m, 2H) and 1.73 (m, 2H) : C₂-H and C₃-H. ^{13}C NMR : 166.7 (CO), 146.4 and 132.8 (C₁ and C₂), 137.4, 133.7, 130.6, 128.6 and 124.6 (C₄, C₃, C₄, C₅ and C₆), 115.4 (C₅), 81.5 (C₁), 56.3 (OCH₃), 34.7 and 29.0 (C₂ and C₃).

N-(1-oxo-4-pentene)-o-nitrobenzamide (9) (1.39 g, 14%), mp 110°C ; ir : 3250, 3050, 2970, 1730, 1690, 1610, 1530, 1350, 1265 cm^{-1} ; uv : 209 (19500), 259 (5800) ; ms (m/z) : 248 (M^{+}), 202, 150 (100%). ^1H NMR (200 MHz) : 9.5 (broad s, 1H, NH), 8.20 (dd, 1H, $J = 8$ and $J = 2$) and 7.43 (dd, 1H, $J = 8$, $J = 2$) : C₃-H and C₆-H, 7.75 (ddd, 1H, $J = 8$, $J = 2$) and 7.60 (ddd, 1H, $J = 8$, $J = 2$) : C₄-H and C₅-H, 5.80 (tdd, 1H, $J_{4,5a} = 17$, $J_{4,5b} = 10$, $J_{3,4} = 7$, C₄-H), 5.07 (dd, 1H, $J_{4,5a} = 17$, $J_{5a,5b} = 2$, C₅-H_a), 5.00 (dd, 1H, $J_{4,5b} = 10$, $J_{5a,5b} = 2$, C₅-H_b), 2.70 (t, 2H, $J = 7$, C₂-H), 2.33 (dt, 2H, $J = 7$, C₃-H). ^{13}C NMR : 172.9 (CO), 167.6 (CO), 145.6 and 132.5 (C₁ and C₂), 136.2, 134.3, 130.7, 127.9 and 124.5 (C₄, C₃, C₄, C₅ and C₆), 116.1 (C₅), 36.5 and 28.1 (C₂ and C₃).

Reduction of N-(1-oxo-4-pentene)-o-nitrobenzamide (9). To a stirred solution of (9) (0.09 g, 0.36 mmol) in methanol (4 ml) was added sodium borohydride (0.067 g, 1.77 mmol) and small amounts of hydrochloric acid at regular intervals. Usual treatment after 2 h and extraction with dichloromethane afforded the crude product (0.088 g) which was chromatographed on silica gel (hexane-ethylacetate 9-7). The compound (8) was isolated (0.055 g, 57.4%).

Preparation of epoxide (10). To a stirred solution of N-(1-methoxy-4-pentene)-o-nitrobenzamide (8) (2.64 g, 10 mmol) in 1,2-dichloroethane (13 ml) at room temperature was added m-chloroperbenzoic acid (13.3 g, 19.1 mmol) and sodium carbonate (1.1 g, 10.4 mmol). After 30 h, the reaction mixture was extracted

with dichloromethane. The organic layers were washed with an aqueous solution of sodium carbonate (10%) and with brine. The epoxide (10) (2.8 g, 100%) was isolated after usual workup. ir : 3275, 2930, 1650, 1530, 1350 cm^{-1} ; uv : 206 (17000), 250 (6500); ms (m/z) : 249 ($\text{M}^{+}-31$), 209, 150 (100%). ^1H NMR (400 MHz) 8.05 - 7.51 (4H, aromatic), 6.59 and 6.33 (2 broad d, 1H, $J \sim 8$, NH), 5.33 (m, 1H, $\text{C}_1\text{-H}$), 3.50 and 3.49 (2s, 3H, OCH_3), 2.96 (m, 1H), 2.77 (m, 1H), 2.52 (m, 1H) and 1.85 (m) : $\text{C}_4\text{-H}$, $\text{C}_5\text{-H}$, $\text{C}_2\text{-H}$ and $\text{C}_3\text{-H}$. ^{13}C NMR : 166.8 (CO), 146.1 and 132.5 (C_1 , and C_2), 133.5, 130.4, 128.5 and 124.2 (C_3 , C_4 , C_5 , and C_6), 81.1 and 80.9 (C_1), 55.8 (OCH_3), 51.7 (C_4), 47.0 (C_5), 31.2 and 31.1, 27.5 and 27.2 (C_2 and C_3).

Preparation of N-(o-nitrobenzoyl)-5-hydroxymethyl-2-methoxypyrrolidines (11).

To a solution of epoxide (10) (2.7 g, 9.6 mmol) in dry benzene (10 ml) under argon at 20°C was added sodium hydride (0.235 g, 9.8 mmol). The reaction mixture was stirred for 2.5 h before addition of brine and extraction with dichloromethane. After usual workup, the crude product was chromatographed on silica gel (dichloromethane-methanol 97-3) to provide 11a (0.65 g, 24%) and 11b (1.23 g, 46%). (11a) mp : 100-102°C. ir : 3500, 2950, 1640 cm^{-1} . uv : 219 (10300), 257 (6800). ms (m/z) : 249, 150 (100%). ^1H NMR (400 MHz, C_6D_6) : 7.82 (d, 1H, $J = 8$) and 7.29 (d, 1H, $J = 8$) : $\text{C}_3\text{-H}$ and $\text{C}_6\text{-H}$, 7.09 (dd, 1H, $J \sim 8$) and 6.92 (dd, 1H, $J \sim 8$) : $\text{C}_4\text{-H}$ and $\text{C}_5\text{-H}$, 4.55 (m, 1H, $\text{C}_5\text{-H}$), 4.22 (1H, $\text{C}_2\text{-H}$), 4.24 (broad d, 1H, $\text{C}_6\text{-H}_a$) ; 4.05 (dd, 1H, $J_{6a,6b} = 11.5$ and $J_{5,6b} = 6.5$, $\text{C}_6\text{-H}_b$), 2.64 (s, 3H, OCH_3), 1.87 (m, 1H, $\text{C}_4\text{-H}_a$), 1.80 (m, 1H, $\text{C}_4\text{-H}_b$), 1.62 (dd, 1H, $J_{3a,3b} = 13$ and $J_{3a,4a} = 7$, $\text{C}_3\text{-H}_a$), 1.44 (m, 1H, $\text{C}_3\text{-H}_b$). ^{13}C NMR : 134.0, 130.3, 129.1 and 124.5 (C_3 , C_4 , C_5 , and C_6), 91.7 (C_2), 65.4 (C_6), 61.4 (C_5), 54.4 (OCH_3), 29.8 and 24.9 (C_3 and C_4). (11b) ir : 3500, 2950, 1640 cm^{-1} . uv : 208, 255 nm. ms (m/z) : 249, 150 (100%). ^1H NMR (400 MHz, C_6D_6) : 7.87 (d, 1H, $J = 8$) and 7.42 (d, 1H, $J = 8$) : $\text{C}_3\text{-H}$ and $\text{C}_6\text{-H}$, 7.09 (dd, 1H, $J \sim 8$) and 6.89 (dd, 1H, $J \sim 8$) : $\text{C}_4\text{-H}$ and $\text{C}_5\text{-H}$, 4.64 (m, 1H, $\text{C}_5\text{-H}$), 4.12 (1H, $\text{C}_2\text{-H}$), 4.14 (1H, $\text{C}_6\text{-H}_a$), 4.07 (dd, 1H, $J_{6a,6b} = 11$ and $J_{5,6b} = 4.5$, $\text{C}_6\text{-H}_b$), 3.62 (OH), 2.46 (s, 3H, OCH_3), 2.11 (m, 1H, $\text{C}_4\text{-H}_a$), 1.73 (m, 2H, $\text{C}_4\text{-H}_b$ and $\text{C}_3\text{-H}_a$), 1.50 (dd, 1H, $J_{3a,3b} = 11.5$ and $J_{3b,4a} = 7$, $\text{C}_3\text{-H}_b$). ^{13}C NMR : 168.7 (CO), 146.5 and 133.1 (C_1 , and C_2), 133.7, 129.9, 129.4 and 124.1 (C_3 , C_4 , C_5 , and C_6), 91.0 (C_2), 64.8 (C_6), 61.0 (C_5), 54.1 (OCH_3), 29.4 and 25.8 (C_3 and C_4).

N-(o-nitrobenzoyl)-5-carboxaldehyde-2-methoxypyrrolidines (12).

Preparation of (12a). To a solution of alcohol (11a) (280 mg, 1.0 mmol) in DMSO (4.0 ml) was added triethylamine (1.0 ml) and sulfur trioxide-pyridine (510 mg). After being stirred at room temperature until the reaction was complete, the mixture was diluted with ethylacetate and washed with an aqueous solution of sodium bicarbonate (10%) and then with water. The residue obtained after usual treatment was purified by preparative layer chromatography (ether) affording the compound (12a) (264 mg, 95%). ir : 2950, 1725, 1650 cm^{-1} . uv : 215, 260 nm. ^1H NMR (400 MHz) : 9.67 and 9.30 (2d, 1H, $J \sim 2.4$, $\text{C}_6\text{-H}$), 8.19 (d, 1H, $J = 8$, $\text{C}_3\text{-H}$ or $\text{C}_6\text{-H}$), 7.74 (dd, 1H, $\text{C}_4\text{-H}$ or $\text{C}_5\text{-H}$), 7.61 (dd + d, 2H, aromatic), 4.54 (m, 1H, $\text{C}_5\text{-H}$), 4.50 (d, 1H, $J \sim 4$, $\text{C}_2\text{-H}$), 2.89 (s, 3H, OCH_3), 2.25, 2.05 and 1.87 ($\text{C}_3\text{-H}$ and $\text{C}_4\text{-H}$).

Preparation of (12b). To a solution of (11b) (429 mg, 1.53 mmol) in DMSO (1.7 ml) and triethylamine (1.55 ml) was added a solution of sulfur trioxide-pyridine (485 mg) in DMSO (4.8 ml). The mixture was stirred at room temperature and the reaction was monitored by TLC. After completion, the reaction medium was extracted with ethylacetate and washed twice with water. The aldehyde (12b) (409 mg, 96%) was obtained after the usual workup. ir : 2920, 1720, 1640, 1520 cm^{-1} . uv : 215 (10000), 256 (6300). ms (m/z) : 249, 150 (100%). ^1H NMR (60 MHz) : 9.80 (d, 1H, $J \sim 1.5$, $\text{C}_6\text{-H}$), 8.18 (d, 1H, $J \sim 8$, $\text{C}_3\text{-H}$ or $\text{C}_6\text{-H}$), 7.64 (3H, aromatic), 4.79 (broad d, $J = 7.5$, $\text{C}_5\text{-H}$), 4.53 (d, 1H, $J = 4$, $\text{C}_2\text{-H}$), 2.84 (s, 3H, OCH_3).

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15. Notes added during the revision of the manuscript : a. The compounds (5) were also obtained by Ban and coll. following a completely different scheme (M. Mori, M. Kimura, Y. Uozumi and Y. Ban, Tetrahedron Letters, 1985, 26, 5947). b. The experimental section was included according the request of one of the referees. c. We wish to express our thanks to Professor H. Umezawa for samples of neothramycins and O-butylneothramycin A.

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