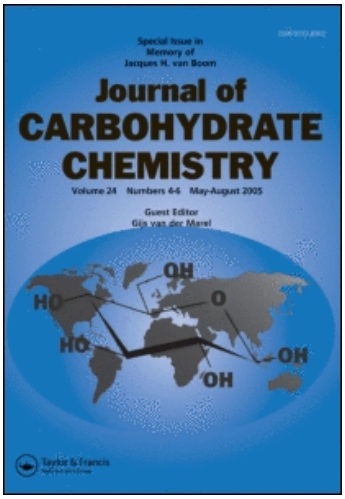


Journal of Carbohydrate Chemistry
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713617200

Semisynthetic 4-*O*-Methyl-ß-Rhodomycins: Synthesis and Structure-Activity Relationships

Cenek Kolar; Manfred Gerken; Hans-Peter Kraemer; Karsten Krohn; Haryanto Linoh

To cite this Article Kolar, Cenek , Gerken, Manfred , Kraemer, Hans-Peter , Krohn, Karsten and Linoh, Haryanto(1990) 'Semisynthetic 4-*O*-Methyl-ß-Rhodomycins: Synthesis and Structure-Activity Relationships', Journal of Carbohydrate Chemistry, 9: 2, 223 — 234
To link to this Article: DOI: 10.1080/07328309008543829
URL: http://dx.doi.org/10.1080/07328309008543829

J. CARBOHYDRATE CHEMISTRY, 9(2&3), 223-234 (1990)

SEMISYNTHETIC 4-*O*-METHYL-ß-RHODOMYCINS: SYNTHESIS AND STRUCTURE-ACTIVITY RELATIONSHIPS

Cenek Kolar*, Manfred Gerken and Hans-Peter Kraemer

Research Laboratories of Behringwerke AG, P.O.Box 1140, D-3550 Marburg, Federal Republic of Germany

and

Karsten Krohn and Haryanto Linoh

Institut für Organische Chemie, Technische Universität Braunschweig, Hagenring 30, D-3300 Braunschweig, Federal Republic of Germany

Received July 25, 1989 - Final Form December 13, 1989

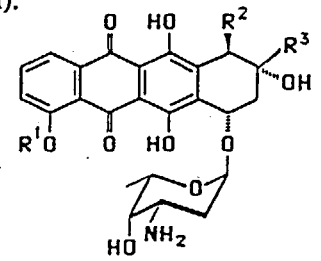
ABSTRACT

The synthesis of 7-*O*-α-L-daunosaminyl-4-*O*-methyl-β-rhodomycinone (**3**) and the determination of its cytotoxic potency compared to that of the natural 7-*O*-α-L-daunosaminyl-β-rhodomycinone (**4**) are described. Starting with natural β-rhodomycinone (**7**), trimethylsilyl protecting groups were attached to the hydroxy groups at position 7 and 10, and the 4-OH group was subsequently methylated (MeI/Cs_2CO_3), thus providing the 4-*O*-methyl-7,10-bis-*O*-trimethylsilyl-β-rhodomycinone (**10**). The two TMS groups were then deblocked to give 4-*O*-methyl-β-rhodomycinone (**12**). In a 3-stage synthesis **12** was converted into 4-*O*-methyl-10-*O*-trifluoroacetyl-ß-rhodomycinone (**15**) to which 1,4-bis-*O*-p-nitrobenzoyl-3-*N*-trifluoroacetyl-L-daunosamine **16** was selectively linked to afford the 7-*O*-α-glycoside **17**. The acyl protective groups are removed by treatment with 1N NaOH to give **3**.

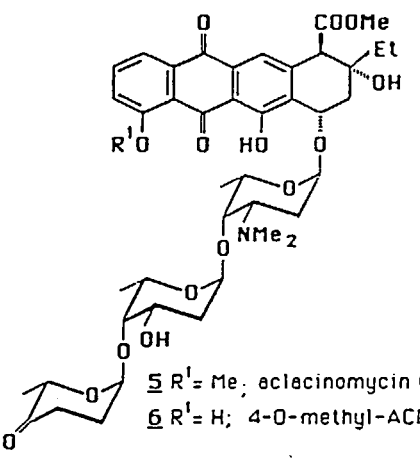
INTRODUCTION

The broad goal of our work on modified anthracycline glycosides has been the chemical synthesis of analogs of rhodomycins.[1,2] Anthracyclines with a methoxy group in position 4 exhibit pharmacological properties which differ distinctly from those of their 4-OH analogs.[3,4] Their cytotoxic effect is reduced by the presence of the 4-O-methyl group. They can therefore be administered at higher dosages, at which a wider spectrum of therapeutic action may be expected.

This important aspect of the structure-activity-relationship (L 1210 leukemia) is apparent in the anthracyclines daunorubicin **1** (IC_{50} = 38 nmol), carminomycin 2 (IC_{50} = 10 nmol) as well as aclacinomycin 5 (IC_{50} = 12 nmol) and its 4-*O*-methyl analog 6 (IC_{50} = 48 nmol).[4]



1 R^1= Me R^2= H R^3= $COCH_3$
2 R^1= H R^2= H R^3= $COCH_3$
3 R^1= Me R^2= OH R^3= Et
4 R^1= H R^2= OH R^3= Et



5 R^1= Me; aclacinomycin (ACM)
6 R^1= H; 4-O-methyl-ACM

Our aim was to examine whether the same principles could be applied to the β-rhodomycins. We therefore synthesized a 7-*O*-α-L-daunosaminyl-4-*O*-methyl-β-rhodomycinone **3** which was compared with the natural β-rhodomycin (oxaunomycin) 4[11] in an oncopharmacological series of tests.

RESULTS AND DISCUSSION

In the synthesis of 4-*O*-methyl-β-rhodomycin **3** we applied the following strategy: trimethylsilyl (TMS) protecting groups were selectively bound to the 7-OH and 10-OH groups of β-rhodomycinone **7**, temporarily directing the subsequent methylation step preferably to the 4-OH group of the aglycone.[6] After the removal of the TMS groups, a trifluoroacetate protecting group at 10-OH of 4-*O*-methyl-β-rhodomycinone permitted the selective glycosylation at C-7.

The TMS protective groups (TMS-Cl, pyridine, dichloromethane) were introduced into β-rhodomycinone 7[5] principally at the secondary hydroxy groups at C-7 and C-10. In addition to the expected disilylated product **8**, a smaller amount of the trisilylated derivative **9** was obtained.

The ^{1}H NMR spectra of compounds **8** and **9** prove the presence of unsubstituted phenolic hydroxy groups, and the signal at 4.47 ppm in the spectrum of **8** indicates the free hydroxy group at C-9.

The application of TMS protective group chemistry is of special interest with regard to the difficult separation of β-rhodomycinone and its β-iso components from microbial raw material. 7,10-Bis-*O*-TMS-β-rhodomycinone **8** can be easily separated from 7,10-bis-*O*-TMS-β-isorhodomycinone on silica gel using dichloromethane as the eluent. After cleavage of the TMS protective groups with 0.1N HCl the two aglycones can be obtained in high purity.

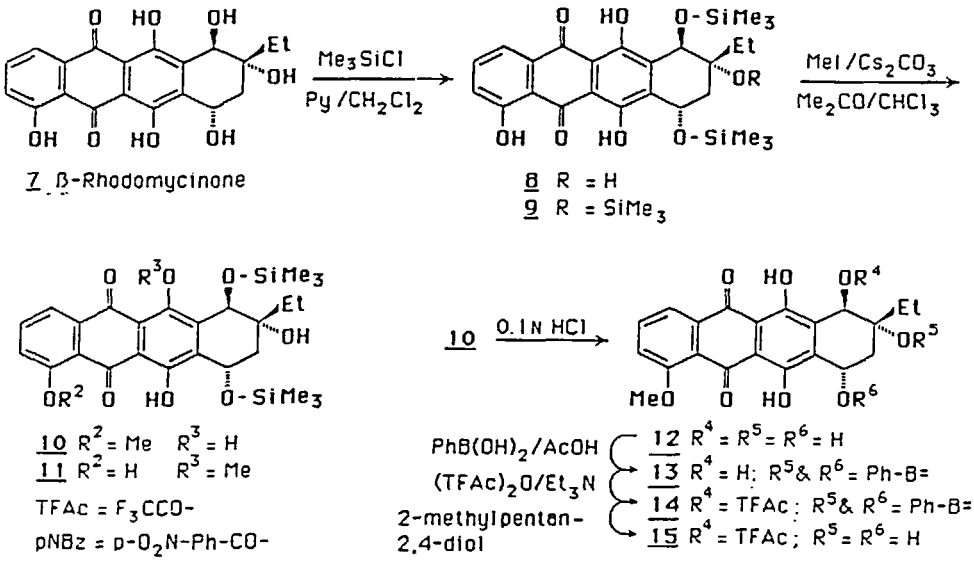
The key reaction in the synthesis of the 4-*O*-methyl-β-rhodomycinones is no doubt the methylation step.[6] The reaction of **8** with a large of excess of methyl iodide in the presence of Na_2CO_3 provides two main products: the 4-*O*-methyl derivative **10** (43%) as a red compound and the 11-*O*-methyl derivative **11** (11%) as a yellow compound. The use of Cs_2CO_3 instead of Na_2CO_3 considerably reduces the amount of MeI required but only marginally improves the selectivity of the 4-*O*-methylation step.

The red colour of **10** (similar to that of daunomycinone and adriamycinone) indicates methylation at 4-OH of the aglycone **8**. The yellow colour of the side product **11** makes methylation at 6-OH or 11-OH of **8** probable. Methylation at 4-OH in compound **10** was proved by several NMR experiments. First the ring protons and carbon atoms were assigned from the ^{1}H NMR-, ^{13}C NMR-, ^{1}H,^{1}H-COSY- and ^{1}H,^{13}C-COSY-spectra. The assignment of the 6-OH and 11-OH was possible from a COLOC-experiment that showed a long range coupling of 6-OH to C-7 and to C-8 of the aglycone. In the 4-*O*-methyl aglycone **10** the 6-OH appeared as a singlet at 13.91, the 11-OH as a singlet at 13.56 and the phenolic *O*-methyl as a singlet at 4.09 ppm. These data enabled the assignment of the phenolic hydroxyl groups in the 11-*O*-methyl compound **11** (6-OH: 13.93 ppm; 4-OH: 13.08 ppm). The data are in accordance with the published values for phenolic hydroxy groups in anthracyclines.[7]

The TMS protective groups in **10** were cleaved quantitatively with 0.1N HCl in CH_2Cl_2/methanol at 0 °C. The 4-*O*-methyl-β-rhodomycinone **12** which was obtained thus represents a new aglycone which to our knowledge is not a known constituent of microbial rhodomycins.

For the selective glycosylation of the new aglycone **12** at 7-OH, it is necessary to protect 10-OH because the reactivity of the alcoholic hydroxy group decreases from

7-OH to 10-OH to 9-OH.[2] The procedure used was as follows: in the first step **12** was converted into 7,9-*O*-phenylboronate **13** by phenylboronic acid and glacial acetic acid as a catalyst.[8] The free 10-OH group in **13** was then acylated with trifluoroacetic anhydride-triethylamine to give **14**. Esterification of the phenolic hydroxy groups was not observed in the reaction.[7a] In the next step the phenylborylene protecting group in **14** was cleaved by treatment with 2-methylpentan-2,4-diol using the method described by Broadhurst *et al.*[9] The 10-*O*-TFAc product **15** obtained was stable in the acidic pH range and was easily purified on silica gel. The yield of **15** after the three reaction steps was 62%.

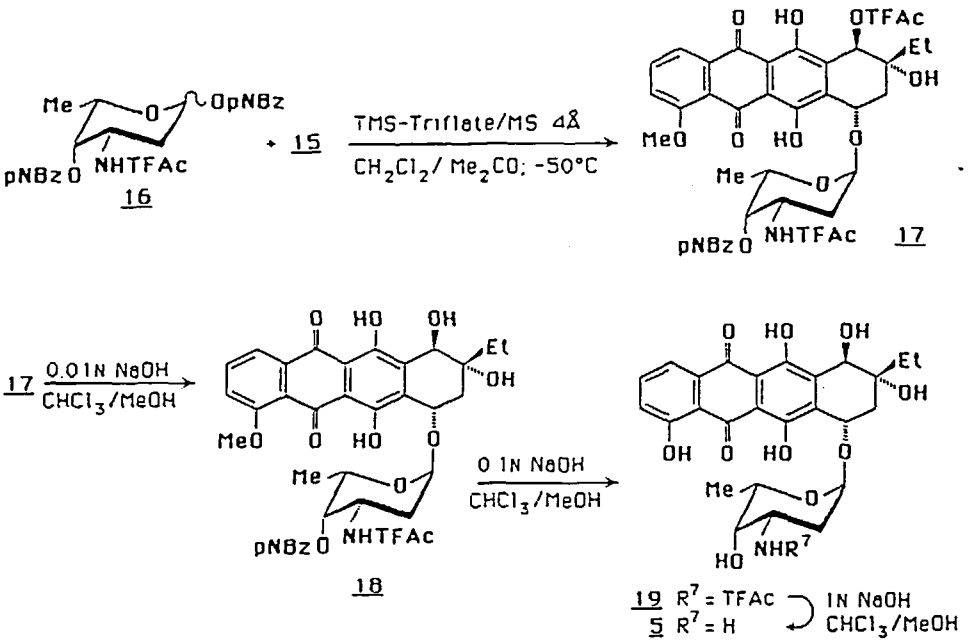


In the glycosylation of **15** we used a slight modification of Terashima's TMS triflate method.[10] The daunosamine donor **16** was reacted with **15** using 9.9 equiv of TMS triflate promoter[1] in dichloromethane-acetone at -50 °C. After completion of the reaction, excess of the promoter was neutralized with triethylamine at -50 °C in order to prevent cleavage of the acid labile sugar component in **17**. The TFAc protective group was partially cleaved in this process, and alkaline workup (0.01N NaOH) completed the deblocking to afford **18** after purification by chromatography (62% based on **15**). The ^{1}H NMR spectra for **17** and **18** provide indirect evidence that the glycosylation process occurred at the 7-OH group of the aglycone **15**. The signals at 13.84 and 13.22 ppm in the spectrum of **18** correspond to the protons of the free hydroxy groups 6-OH and 11-OH, the signal at 3.47 ppm being assigned to the proton of the 9-OH group.

Table 1

compound	IC_{50} (L 1210) [μg/mL]		LD_{50}
	1 h	7 d	(mg/kg)
3: 7-Dau-4-*O*-Me-β-RMN	0.29	0.12	1.0 (3xip)
4: 7-Dau-β-RMN	0.0055	0.0019	< 0.3 (3xip)

Evidence for the presence of the α-glycosidic linkage can be seen in the characteristic broad singlet signal at 5.69 ppm for H-1. The signal at 5.31 ppm ($J_{7,8a}$= 1.5 Hz and $J_{7,8b}$= 3.5 Hz) indicated that the configuration on the C-7 atom was unchanged. When **18** was deblocked it was observed that the p-nitrobenzoyl protecting group was cleaved by 0.1N NaOH whereas the *N*-trifluoroacetyl protecting group was only cleaved after addition of 1N NaOH. The structures of the resulting products **19** and **3** were examined by ^{1}H NMR spectroscopy, and the presence of **3** was additionally confirmed by FAB MS.



The semisynthetic rhodomycin **3** and 7-*O*-α-L-daunosaminyl-β-rhodomycinone **4**[11,12] were tested for cytostatic efficacy in an *in vitro* test system.[13] The effect of the substance on the growth of L 1210 leukemia cells over periods of 1 hour and 7 days was determined. The results IC_{50} (Table 1) show that 4-*O*-methyl-rhodomycin **3** is consider-

ably less cytotoxic than the microbial product oxaunomycin 4. The biological data obtained in the experiments for 3 and 4 were found to correlate with those determined for daunorubicin 1 and its 4-*O*-desmethyl analog carminomycin 2 and aclacinomycin 5 and its 4-*O*-methyl analog 6.[4]

EXPERIMENTAL

General Procedure. Reactions were carried out at ambient temperature unless otherwise stated. Solutions were concentrated under reduced pressure at below 40 °C (bath temperature). The phosphate or citrate buffer solutions used to wash the organic phases were prepared as follows: aqueous 0.1M potassium dihydrogen phosphate or 0.1M sodium citrate solutions were adjusted to the corresponding pH value using 0.1N NaOH or 0.1N HCl. Melting points, determined on a Büchi apparatus, are uncorrected. ^{1}H NMR spectra were recorded at 200 MHz, 300 or 400 MHz on a Bruker AC-200, a Bruker AC-300 or a Bruker AM-400 and at 400 MHz on a Jeol GX 400 NMR spectrometer, respectively. Chemical shifts for ^{1}H resonances were recorded relative to tetramethylsilane (0.0). The ^{1}H resonances were routinely assigned by ^{1}H,^{1}H-COSY experiments (^{1}H,^{13}C-COSY and COLOC experiments - see p 3), using the standard pulse sequences of the Bruker Aspect-300 software. Specific rotations were determined with a Perkin-Elmer 241 polarimeter equipped with 10 cm cuvettes. Reactions were monitored by TLC on silica gel plates 60 F 254 (Merck) and the spots were detected by ultraviolet light or by spraying with ethanolic 10% sulfuric acid solution and subsequent heating to 150 - 200 °C. The glycosylation reactions were performed under an argon or nitrogen cover. Preparative chromatography was performed on silica gel (Merck Kieselgel 60 particle size 0.015-0.040 mm) with the solvent system specified.

7,10-Bis-*O*-trimethylsilyl-β-rhodomycinone (8) and 7,9,10-Tris-*O*-trimethylsilyl-β-rhodomycinone (9). β-Rhodomycinone 7[14] (2.0 g, 5.18 mmol) was dissolved in 2:1 dichloromethane-pyridine (60 mL) and chlorotrimethylsilane (20 mL, 155.4 mmol) was added at 0 °C. After 1 h, the mixture was diluted with dichloromethane (30 mL) and washed successively with phosphate buffer (pH 7.5, 30 mL) and water (40 mL). The organic layer was concentrated *in vacuo* and coevaporated with toluene. The residue was chromatographed on a column of silica gel (100 g) with dichloromethane to give 8 (2.3 g, 84%) and 9 (0.13 g, 5%).

Compound 8: mp 185 °C; $[\alpha]_D$ +398° (c 0.06, chloroform); ^{1}H NMR (400 MHz, $CDCl_3$) δ 13.72 (s, 1H, OH-6), 12.89 (s, 1H, OH-11), 12.24 (s, 1H, OH-4), 7.89 (dd, 1H, $J_{1,2}$ = 7.5 Hz, $J_{1,3}$= 1.3 , H-1), 7.70 (dd, 1H, $J_{1,2}$, $J_{2,3}$ = 8.3 Hz, H-2), 7.31 (dd, 1H, $J_{1,3}$,

$J_{2,3}$, H-3), 5.33 (br s, 1H, H-7), 4.87 (d, 1H, $J_{8b,10}$ = 1.3 Hz, H-10), 4.47 (s, 1H, OH-9), 2.14 (dd, 1H, $J_{7,8a}$ = 1.3 Hz, J_g = 14.3 Hz, H-8a), 2.00 (dt, 1H, $J_{7,8b}$ = 1.3 Hz, J_g, $J_{8b,10}$, H-8b), 1.67 (q, 2H, $J_{13,14}$ = 7.5 Hz, H-13a and H-13b), 1.06 (t, 3H, $J_{13,14}$, H-14), 0.25 (s, 9H, $Si(Me)_3$), 0.13 (s, 9H, $Si(Me)_3$).

Anal. Calcd for $C_{26}H_{34}O_8Si_2$: C, 58.84; H, 6.46. Found: C, 58.75; H, 6.47.

Compound 9: mp 106 °C; $[\alpha]_D$ +298° (c 0.04, chloroform); ^1H NMR (200 MHz, $CDCl_3$) δ 13.85 (s, 1H, OH-6), 12.89 (s, 1H, OH-11), 12.31 (s, 1H, OH-4), 7.89 (dd, 1H, $J_{1,2}$ = 7.8 Hz, $J_{1,3}$ = 1.2 Hz, H-1), 7.68 (dd, 1H, $J_{1,2}$, $J_{2,3}$ = 8.3 Hz, H-2), 7.28 (dd, 1H, $J_{1,3}$, $J_{2,3}$, H-3), 5.13 (dd, 1H, $J_{7,8a}$ = 2.0 Hz, $J_{7,8b}$ = 4.8 Hz, H-7), 4.83 (d, 1H, $J_{8a,10}$ = 1 Hz, H-10), 2.05 (ddd, 1H, $J_{7,8a}$, J_g = 14.2 Hz, $J_{8a,10}$, H-8a), 1.98 (dd, 1H, $J_{7,8b}$, J_g, H-8b), 1.72 (m, 1H, $J_{13a,14}$ = 7.5 Hz, J_g = 15 Hz, H-13a), 1.58 (m, 1H, $J_{13b,14}$ = 7.5 Hz, J_g, H-13b), 1.02 (t, 3H, $J_{13a,14}$, $J_{13b,14}$, H-14), 0.20-0.10 (m, 27H, Si-Me).

Anal. Calcd for $C_{29}H_{42}O_8Si_3$: C, 57.77; H, 7.02. Found: C, 57.85; H, 7.04.

4-*O*-Methyl-7,10-bis-*O*-trimethylsilyl-β-rhodomycinone (10) and 11-*O*-Methyl-7,10-bis-*O*-trimethylsilyl-β-rhodomycinone (11). Compound 8 (11.3 g, 21.3 mmol) was dissolved in 100:1 dry acetone-chloroform (1000 mL) and cesium carbonate (42 g) and iodomethane (50 mL, 715 mmol) were added at 4 °C. The mixture was stirred for 3 d at room temperature and concentrated *in vacuo*. The residue was dissolved in dichloromethane (200 mL), washed successively with water (200 mL x 3), 1 N HCl (150 mL) and water. The organic layer was dried (sodium sulfate) and concentrated *in vacuo*. The residue was chromatographed on a column of silica gel (650 g) with dichloromethane to give as major products **10** (4.91 g, 43%), **11** (1.24 g, 11%), and starting material **8**.

Compound **10**: mp 202 °C; $[\alpha]_D$ +460° (c 0.05 , chloroform); ^1H NMR (300 MHz, $CDCl_3$) δ 13.91 (s, 1H, OH-6), 13.56 (s, 1H, OH-11), 8.05 (dd, 1H, $J_{1,2}$ = 7.5 Hz, $J_{1,3}$ = 1.3 , H-1), 7.78 (dd, 1H, $J_{1,2}$, $J_{2,3}$ = 8.5 Hz, H-2), 7.39 (dd, 1H, $J_{1,3}$, $J_{2,3}$, H-3), 5.36 (br s, 1H, H-7), 4.87 (d, 1H, $J_{8b,10}$ = 1.3 Hz, H-10), 4.52 (s, 1H, OH-9), 4.09 (s, 3H, O-Me), 2.13 (dd, 1H, $J_{7,8a}$ = 4 Hz, J_g = 14.5 Hz, H-8a), 1.98 (dt, 1H, $J_{7,8b}$, J_g, $J_{8b,10}$, H-8b), 1.66 (m, 2H, $J_{13,14}$ = 7.2 Hz, H-13a and H-13b), 1.05 (t, 3H, $J_{13,14}$, H-14), 0.23 (s, 9H, Si-Me), 0.13 (s, 9H, Si-Me). ^{13}C NMR (400 MHz, $CDCl_3$) δ 187.10 (CO, C-12 (C-5)), 186.47 (, C-5 (C-12)), 160.97 (C-4), 156.75 (C-11a), 156.60 (C-5a), 137.86 (C-10a), 137.11 (C-6a), 135.73 (C-12a), 135.52 (C-2), 121.06 (C-4a), 119.71 (C-1), 118.10 (C-3), 112.22 (C-6), 111.66 (C-11), 77.00 (t, $CDCl_3$), 72.37 (C-9), 67.58 (C-10), 63.28 (C-7), 56.63 (MeO-4), 33.34 (C-8), 30.17 (C-13), 6.45 (C-14), 0.63 (s, 3C, $SiMe_3$), 0.47 (s, 3C, $SiMe_3$).

Anal. Calcd for $C_{27}H_{36}O_8Si_2$: C, 59.53; H 6.66. Found: C, 59.67; H, 6.67.

Compound **11**: mp 78°C; $[\alpha]_D$ +150° (c 0.05 chloroform); ^{1}H NMR (200 MHz, $CDCl_3$) δ 13.93 (*s*, 1H, *OH-6*), 13.08 (*s*, 1H, OH-4), 7.85 (dd, 1H, $J_{1,2}$ = 7.6 Hz, $J_{1,3}$ = 1.1 , H-1), 7.67 (dd, 1H, $J_{1,2}$ = 8.3 Hz, H-2), 7.32 (dd, 1H, $J_{1,2}$, $J_{2,3}$, H-3), 5.28 (, 1H, $J_{7,8a}$ = 3.4 Hz, $J_{7,8b}$ = 2.5 , H-7), 4.87 (s, 1H, H-10), 4.44 (, 1H, OH-9), 3.95 (s, 3H, 11-OMe), 2.14 (dd, 1H, $J_{7,8a}$, J_g = 14.5 Hz, H-8a), 2.04 (dd, 1H, $J_{7,8b}$, J_g = H-8b), 1.68 (q, 2H, $J_{13,14}$ = 7.4 H-13a and H-13b), 1.06 (t, 3H, $J_{13,14}$, H-14), 0.28 (s, 9H, Si-Me_3), 0.14 (s, 9H, Si-Me_3).

Anal. Calcd for $C_{28}H_{38}O_8Si_2$: C, 60.19; H, 6.85. Found: C, 60.31; H, 6.88.

4-*O*-Methyl-β-rhodomycinone (12). 0.1N HCl (75 mL) was added to a stirred solution of **10** (2.0 g, 3.67 mmol) in 1:1 dichloromethane-methanol (300 mL) at 0 °C. The reaction mixture was stirred for 3/4 h, diluted with dichloromethane (300 mL) and successively washed with phosphate buffer (pH 7, 50 mL) and water. The organic phase was dried with sodium sulfate and concentrated *in vacuo*. The residue was crystallized from a mixture of dichloromethane - ether to give **12** (1.44 g, 98%): mp 141 °C; $[\alpha]_D$ +73° (c 0.2, chloroform); ^{1}H NMR (400 MHz, $CDCl_3$) δ 13.86 (*s*, 1H, *OH-6*), 13.38 (*s*, 1H, OH-11), 8.02 (dd, 1H, $J_{1,2}$ = 7.8 Hz, $J_{1,3}$ = 1.1 , H-1), 7.80 (dd, 1H, $J_{1,2}$, $J_{2,3}$ = 8.5 Hz, H-2), 7.40 (dd, 1H, $J_{1,3}$, $J_{2,3}$, H-3), 5.25 (br s, 1H, H-7), 4.88 (d, 1H, $J_{8a,10}$ = 2.0 Hz, $J_{10,OH}$ = 4.0 Hz, H-10), 4.10 (d, 1H, $J_{10,OH}$ = 4.0 Hz, OH-10), 4.10 (s, 3H, 4-OMe), 3.50 (d, 1H, $J_{7,OH}$ = 3.0 Hz, OH-7), 3.47 (s, 1H, OH-9), 2.21 (dd, 1H, $J_{7,8b}$ = 1 Hz, J_g = 14.2 Hz, H-8b), 2.15 (ddd, 1H, $J_{7,8a}$ = 4.8 Hz, J_g, $J_{8a,10}$, H-8a), 1.87 (m, 1H, $J_{13,14}$ = 7.2 Hz, J_g = 15.0 Hz, H-13a), 1.77 (m, 1H, $J_{13,14}$, J_g, H-13b), 1.12 (t, 3H, $J_{13,14}$, H-14).

Anal. Calcd for $C_{21}H_{20}O_8$: C, 63.00; H, 5.03. Found: C, 63.07; H, 5.04.

4-*O*-Methyl-7,9-*O*-phenylborylene-β-rhodomycinone (13). A mixture of **12** (1.6 g, 4 mmol), powdered molecular sieves 4Å (4 g), glacial acetic acid (3.2 mL) and phenylboronic acid (1.7 g) in dry toluene (140 mL) was partially concentrated to 2/3 of the original volume. A second portion of glacial acetic acid (3.2 mL) was added and the mixture was stirred for 50 h at 87 °C. The solution was filtered through Celite and the filtrate was washed successively with diluted $NaHCO_3$ solution (5%, 60 mL) and water. The organic layer was dried (sodium sulfate) and concentrated *in vacuo*. Chromatography of the residue on silica gel in 10:1:1 toluene-methanol-AcOH provided **13** (1.8 g, 92%): mp 225-230 °C; $[\alpha]_D$ +440° (c 0.02, chloroform); ^{1}H NMR (300 MHz, $CDCl_3$) δ 13.62 (s, 1H, PhOH), 13.23 (s, 1H, PhOH), 8.16, 7.71 and 7.21 (m, 5H, PhB), 7.79 (d, 1H, $J_{1,2}$= 8 Hz, (H-1), 7.58 (t, 1H, $J_{1,2}$, $J_{2,3}$ = 8 Hz, H-2), 7.28 (d, 1H, $J_{2,3}$, H-3), 5.61 (t, 1H, $J_{7,8a}$ = 2.3 Hz, $J_{7,8b}$ = 2.5 Hz, H-7), 4.88 (s, 1H, H-10), 3.97 (s, 3H, OMe), 2.24 (dd, 1H, J_g = 14 Hz, $J_{7,8a}$, H-8a), 7.21 (dd, 1H, J_g, $J_{7,8b}$, H-8b), 2.07 (m, 1H = 12 Hz, $J_{13,14}$ = 7.5 Hz, H-13a), 1.78 (m, 1H, J_g, $J_{13,14}$, H-13b), 1.17 (t, 3H, $J_{13,14}$, H-14).

Anal. Calcd for $C_{27}H_{23}BO_8$: C 66.69; H 4.77. Found: C 66.82; H 4.76.

4-*O*-Methyl-7,9-phenylborylene-10-*O*-trifluoroacetyl-β-rhodomycinone (14). Trifluoroacetic anhydride (3.4 mL) was added to a solution of compound **13** (600 mg, 1.23 mmol) in dry dichloromethane (40 mL) at 0 °C. Triethylamine (0.2 mL) was added and the mixture was stirred for 18 h at 4 °C. TLC (10:1:0.1 toluene-methanol-acetic acid) showed only the presence of **14**. After concentration *in vacuo*, the residue was successively coevaporated *in vacuo* with toluene (50 mL x 3) and chloroform (50 mL). The crude product (720 mg) was used in the subsequent steps without further purification.

4-*O*-Methyl-10-*O*-trifluoroacetyl-β-rhodomycinone (15).[1,2] To a stirred solution of compound **14** (720 mg crude product) in dichloromethane (20 mL) 2-methyl-pentan-2,4-diol (6 mL) was added. The mixture was stirred for 18 h at room temperature, diluted with dichloromethane (20 mL) and washed with 0.01N HCl (20 mL x 3). The organic layer was dried (sodium sulfate) and concentrated *in vacuo*. The residue was chromatographed on a column of silica gel with 95:5:1:0.25:0.1 chloroform-acetone-acetic acid-water-triethylamine to give **15** (445 mg, 73 %): mp 244 °C; $[\alpha]_D$ -102° (c 0.5, chloroform); ^{1}H NMR (300 MHz, $CDCl_3$) δ 13.69 (s, 1H, OH-6), 13.03 (s, 1H, OH-11), 7.94 (dd, 1H, $J_{1,2}$ = 7.5 Hz, $J_{1,3}$ = 1.5 Hz, H-1), 7.77 (dd, 1H, $J_{1,2}$, $J_{2,3}$ = 8.5 Hz, H-2), 7.38 (dd, 1H, $J_{1,3}$, $J_{2,3}$, H-3), 6.29 (d, 1H, $J_{8a,10}$ = 1.5 Hz, H-10), 5.31 (dd, 1H, $J_{7,8a}$ = 1.5 Hz, $J_{7,8b}$ = 4.7 Hz, H-7), 4.08 (s, 3H, 4-OMe), 4.00 (s, 1H, OH-9), 2.37 (ddd, 1H, $J_{7,8a}$,J_g = 15 Hz, $J_{8a,10}$, H-8a), 2.00 (dd, 1H, $J_{7,8b}$, J_g, H-8b), 1.71 (m, 1H, $J_{13,14}$ = 7.4 Hz, J_g = 13.8 Hz, H-13a), 1.53 (m, 1H, $J_{13,14}$, J_g, H-13b), 1.09 (t, 3H, $J_{13,14}$, H-14).

Anal. Calcd for $C_{23}H_{19}F_3O_9$: C, 55.65; H, 3.86; F, 11.48. Found: C, 55.74; H, 3.87; F, 11.23.

4-*O*-Methyl-7-*O*-(4-*O*-p-nitrobenzoyl-2,3,6-trideoxy-3-trifluoroacetamido-α-L-lyxohexopyranosyl)-10-*O*-tri-fluoroacetyl-β-rhodomycinone (17). To a mixture of aglycone **15** (550 mg, 1.10 mmol), daunosamine donor **16** (900 mg, 1.66 mmol) and powdered molecular sieves 4 Å (2.0 g) in 10:1 dichloromethane-acetone (60 mL) was added in three portions trimethylsilyl triflate (1.8 mL, 9.9 mmol) at -50° C. The reaction mixture was stirred for 5 h, diluted with dichloromethane (100 mL) and then triethylamine (2 mL) was added to the cooled mixture. After 5 min of stirring the mixture was filtered, and the filtrate was washed with citrate buffer (pH 4.5, 50 mL x 2), dried (sodium sulfate) and concentrated *in vacuo*. The residue (1650 mg), which contained **17** and its OH-10 derivative **18** as its major constituents, was used in the subsequent steps without further purification. A part of the crude product was purified by preparative TLC in 10:1 dichloromethane-acetone to give pure compound **17** which was used for the evaluation of analytical and NMR spectral data: mp 126-128 °C; $[\alpha]_D$ -126° (c 0.1, chloroform); ^{1}H NMR (300 MHz, $CDCl_3$) δ 13.84 (s, 1H, OH-6), 13.22 (s, 1H, OH-11),

8.34 (dt, 2H, $J_{o,m}$= 9 Hz, pNBz), 8.28 (dt, 2H, $J_{o,m}$, pNBz), 8.05 (dd, 1H, $J_{1,2}$ = 7.8 Hz, $J_{1,3}$ = 1.2 Hz, H-1), 7.83 (dd, 1H, $J_{1,2}$, $J_{2,3}$ = 8.5 Hz, H-2), 7.43 (dd, 1H, $J_{1,3}$, $J_{2,3}$, H-3), 6.39 (s, 1H, H-10), 6.29 (d, 1H, $J_{3',NH}$ = 7.0 Hz, N-H), 5.69 (s, 1H, H-1'), 5.49 (d, 1H, $J_{3',4'}$ = 2.5 Hz, H-4'), 5.31 (dd, 1H, $J_{7,8a}$ = 1. 5 Hz, $J_{7,8b}$ = 3.5 Hz, H-7), 4.45 (q, 1H, $J_{5',6'}$ = 6.5 Hz, H-5'), 4.44 (m, 1H, H-3'), 4.10 (s, 3H, 4-OMe), 2.45 (d, 1H, J_g = 15.0 Hz, H-8a), 2.11 (dd, $J_{7,8b}$, J_g, H-8b), 2.11 (m, 1H, H-2'eq), 2.08 (m, 1H, H-2'ax), 1.78 (m, 1H, $J_{13,14}$ = 7.5 Hz, J_g = 14.0 Hz, H-13a), 1.52 (m, 1H, $J_{13,14}$, J_g, H-13b), 1.27 (d, 3H, $J_{5',6'}$ = 6.5 Hz, H-6'), 1.10 (t, 3H, $J_{13,14}$ = 7.5 Hz, H-14).

FAB MS: m/z 871 ($M+H^+$).

4-*O*-Methyl-7-*O*-(p-nitrobenzoyl-2,3,6-trideoxy-3-trifluoroacetamido-α-L-lyxohexopyranosyl)-β-rhodomycinone (18). The crude product containing compounds **17** and **18** (1650 mg) was dissolved in 2:1 chloroform-methanol (30 mL), and 0.01N NaOH (5 mL) was added at room temperature. Methanol was then added to homogenize the organic and aqueous layers. After stirring for 10 min, TLC (10:1 dichloromethane-acetone) showed that the starting material had been completely converted. The mixture was neutralized with 0.01N HCl (5 mL) and concentrated *in vacuo*. The residue was dissolved in chloroform (70 mL) and washed with water (30 mL x 2). The organic layer was dried (sodium sulfate) and concentrated *in vacuo*. The resulting product was chromatographed on a column of silica gel (180 g) with 10:1 dichloromethane-acetone to give **18** (0.53 g, 62% related to aglycone): mp 196-198 °C; $[\alpha]_D$ -50° (c 0.02, chloroform).

Anal. Calcd for $C_{36}H_{33}F_3N_2O_{14}$: C, 55.82; H, 4.29; F, 7.36; N, 3.62. Found: C, 55.78; H, 4.31; F, 7.27; N, 3.54.

4-*O*-Methyl-7-*O*-(2,3,6-trideoxy-3-trifluoroacetamido-α-L-lyxohexopyranosyl)-β-rhodomycinone (19). 0.1N NaOH (2 mL, 2 mmol) was added to a stirred solution of compound **19** (50 mg, 0.064 mmol) in 2:1 chloroform-methanol (3 mL). Then methanol was added to homogenize the organic and aqueous layers. After stirring for 15 min TLC (20:1 dichloromethane-methanol) showed the complete conversion of the starting material. The mixture was neutralized with 0.1N HCl (2 mL) and concentrated under reduced pressure. The residue was dissolved in 3:1 chloroform-ethyl acetate (60 mL) and washed successively with phosphate buffer (pH 7, 20 mL) and water (30 mL). The organic layer was dried (sodium sulfate) and concentrated *in vacuo*. The resulting product was chromatographed on a column of silica gel (15 g) with 10:1 dichloromethane-acetone to give **19** (27 mg, 66%): mp 207-209 °C; $[\alpha]_D$ + 195° (c 0.02, chloroform); ^1H NMR (300 MHz, $CDCl_3$) δ 13.84 (s, 1H, Ph-OH), 13.39 (s, 1H, Ph-OH), 8.00 (dd, 1H, $J_{1,2}$ = 7.5 Hz, $J_{1,3}$ = 1.1 Hz, H-1), 7.78 (dd, 1H, $J_{1,2}$, $J_{2,3}$ = 8.9 Hz, H-2), 7.39 (dd, 1H, $J_{1,3}$, $J_{2,3}$, H-3), 6.72 (d, 1H, $J_{3',NH}$ = 8.5 Hz, N-H), 5.48 (d, 1H, $J_{1',2'}$ = 3.5

Hz, H-1'), 5.12 (dd, 1H, $J_{7,8a}$ = 1.5 Hz, $J_{7,8b}$ = 3.8 Hz, H-7), 4.92 (s, 1H, $J_{8a,10}$ = 1.0 Hz, H-10), 4.32 (q, 1H, $J_{4',5'}$ = 1.5 Hz, $J_{5',6'}$ = 6.5 Hz, H-5'), 4.18 (m, $J_{2'ax,3'}$ = 12.9 Hz, $J_{2'eq,3'}$ = 4.9 Hz, $J_{3',4'}$ = 1.5 Hz, $J_{3',NH}$ = 8.5 Hz, H-3'), 4.06 (s, 3H, MeO-4), 3.65 (dd, 1H, $J_{3',4'}$, $J_{4',5'}$, H-4'), 3.65 (s, 1H, OH-9), 2.81 (br s, 1H, OH-10), 2.20 (br d, 1H, $J_{7,8a}$, $J_{8a,10}$, J_g = 14.0 Hz, H-8a), 2.14 (dd, 1H, $J_{7,8b}$, J_g, H-8b), 1.97 (dd, 1H, $J_{1',2'eq}$ < 1.0 Hz, $J_{2'eq,3'}$ = 4.9 Hz, J_g = 13.0 Hz, H-2'eq), 1.87 (m, 1H, $J_{13a,14}$ = 7.5 Hz, J_g = 15.0 Hz, H-13a), 1.82 (ddd, 1H, $J_{1',2'ax}$, $J_{2'ax,3'}$, J_g, H-2'ax), 1.75 (m, 1H, $J_{13b,14}$ = 7.5 Hz, J_g = 15.0 Hz, H-13b), 1.33 (d, 1H, $J_{5',6'}$ = 6.5 Hz, H-6'), 1.11 (t, 3H, $J_{13,14}$ = 7.5 Hz, H-14).

Anal. Calcd for $C_{29}H_{30}F_3NO_{11}$: C, 55.68; H, 4.83. Found: C, 55.67; H, 4.81.

7-*O*-(3-Amino-2,3,6-trideoxy-α-L-lyxohexopyranosyl)-4-*O*-methyl-β-rhodomycinone (3). 1N NaOH (10 mL) was added to a stirred solution of compound **19** (500 mg, 0.64 mmol) in 3:1 methanol-chloroform (10 mL). Then methanol was added to homogenize the organic and aqueous layers. After stirring for 15 min TLC (10:5:2:1:0.5:0.05 chloroform-acetone-methanol-acetic acid-water-triethylamine) showed that the starting material had been completely converted. The mixture was neutralized with 0.1N HCl (2 mL) and concentrated *in vacuo*. The residue was chromatographed on a column of silica gel (70 g) with the eluent used for TLC and then on a column of aminated silica gel (25 g) with 10:1 methanol-chloroform to give **5** (240 mg, 71%): mp 192-194 °C; $[\alpha]_D$ +270° (c 0.01, methanol); ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.57 (d, 1H, $J_{1,2}$ = 7.0 Hz, H-1), 7.57 (dd, 1H, $J_{1,2}$, $J_{2,3}$ = 8.0 Hz, H-2), 7.28 (d, 1H, $J_{2,3}$, H-3), 4.96 (d, 1H, $J_{1',2'}$ = 2.5 Hz, H-1'), 4.65 (s, 1H, H-7), 4.37 (s, 1H, H-10), 3.76 (q, 1H, $J_{5',6'}$ = 6.5 Hz, H-5'), 3.67 (s, 3H, 4-OMe), 3.06 (s, 1H, H-4'), 2.57 (m, 1H, H-3'), 1.79 (s, 2H, H-8a and H-8b), 1.42 (ddd, 1H, $J_{1',2'ax}$ = 3.2 Hz, $J_{2'ax,3'}$ = 13.0 Hz, J_g = 13.0 Hz, H-2'ax), 1.21 (dd, 1H, J_g, $J_{2'eq,3'}$ = 4.5 Hz, H-2'eq), 1.34 (m, 2H, $J_{13,14}$ = 7.5 Hz, H-13a and H-13b), 0.85 (d, 3H, $J_{5',6'}$ = 6.5 Hz, H-6'), 0.66 (t, 3H, $J_{13,14}$ = 7.5 Hz, H-14).

Anal. Calcd for $C_{27}H_{31}NO_{10}$: C, 61.24; H, 5.90; N, 2.65. Found: C, 61.34; H, 5.92; N, 2.59.

ACKNOWLEDGEMENT

The authors would like to thank Dr. S. Berger and Dr. T. Kämpchen of the University of Marburg for measuring the NMR spectra and Dr. H.-W. Fehlhaber, Hoechst AG, Frankfurt, for performing the MS analysis.

REFERENCES AND FOOTNOTES

1. C. Kolar, K. Dehmel, U. Knödler, M. Paal, P. Hermentin and M. Gerken, *J. Carbohydr. Chem.*, **8**, 295 (1989).

2. M. Gerken, S. Blank, C. Kolar and P. Hermentin, *J. Carbohydr. Chem.*, **8**, 247 (1989).

3. F. Arcamone: *Doxorubicin*, Academic Press; New York; 289 (1981).

4. T. Oki, in *Anthracycline Antibiotics*, H. S. El Khadem, Ed. Academic Press; New York, 75 (1982).

5. H. Brockmann, K. Bauer, *Naturwissenschaften*, **37**, 492 (1950) and H. Brockmann, E. Spohler, *Naturwissenschaften*, **42**, 154 (1955).

6. H. Tanaka, T. Yoshioka, Y. Shimanchi, Y. Matsuzawa, T. Oki, T. Inui, *J. Antibiotics*, **33**, 1323 (1980).

7. A. Vigevani, M. Ballabio, E. Gandini, S. Penco, *Magn. Reson. Chem.*, **23**, 344 (1985).

7a. K. Krohn and B. Behnke, *Liebigs Ann. Chem.*, **12**, 2011 (1979).

8. R. J. Ferrier, in *Adv. Carbohydr. Chem. Biochem.*, **35**, 31 (1978).

9. M. J. Broadhurst, C. H. Hassall, G. J. Thomas, *J. Chem. Soc., Perkin Tr. I*, 2249 (1982).

10. Y. Kimura, M. Suzuki, T. Matsumoto, R. Abe, S. Terashima, *Chem. Lett.*, **4**, 501 (1984).

11. A. Yoshimoto, S. Fujii, O. Johdo, K. Kubo, T. Ishikura, H. Naganawa, T. Sawa, T. Takeuchi, H. Umezawa, *J. Antibiotics*, **39**, 902 (1986).

12. M. Gerken, M. Krause, S. Blank, C. Kolar, P. Hermentin, *Carbohydr. Symposium*, Darmstadt 1987, Abstr. Pap. A-143.

13. H.-P. Kraemer, H. H. Sedlacek, *Behring Inst. Mitt.*, **74**, 301 (1984).

14. We are indebted to H. G. Berscheid (Hoechst AG, Frankfurt) for gifts of 7,10-bis-O-(α-L-rhodosaminyl)-β-rhodomycinone.