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Synthesis and collagenase inhibition of new glycosides of aranciamycinone: the aglycon of the naturally occurring antibiotic aranciamycin

Mikael Bols [a], Lise Binderup [b], Jytte Hansen [b] and Poul Rasmussen [b]

[a] *Department of Organic Chemistry, The Technical University of Denmark, DK-2800 Lyngby (Denmark)*
[b] *Leo Pharmaceutical Products, Industriparken 55, DK-2750 Ballerup (Denmark)*

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ABSTRACT

Glycosides of aranciamycinone were prepared by glycosylation with sugar acetates and trimethylsilyl triflate in dichloromethane. Glycosides of the following sugars were prepared: α-L-rhamnopyranose, β-D-glucopyranose, β-D-ribopyranose, β-D-xylopyranose, α-L-fucopyranose, 2-azido-2,6-dideoxy-α-L-mannopyranose, 2,6-dideoxy-α-L-*arabino*-hexopyranose, 3,6-dideoxy-α-L-*arabino*-hexopyranose, and 4,6-dideoxy-α-L-*lyxo*-hexopyranose. The new glycosides were tested for inhibition of *Clostridium histolyticum* collagenase and Yoshida Sarcoma tumor cells.

INTRODUCTION

Aranciamycin (**1**), an anthracycline antibiotic, has been isolated from *Streptomyces echinatus*[1] and *Streptomyces chromofuscus*[2]. It consists of the tetracyclic aglycon aranciamycinone[1] (**2**) and 2-*O*-methyl-L-rhamnose[3], and is a member of the steffimycin family of antibiotics[4,5].

During our screening for microbial metabolites with enzyme-inhibiting activity, we have isolated **1** from a strain of *Streptomyces griseoflavus*. While **1** is known to have borderline activity against tumor cells, we discovered it to be a specific inhibitor of *Clostridium histolyticum* collagenase. Studies were thus undertaken to investigate the structure–activity relationship in derivatives of **1**. The work now described was aimed at the synthesis of a number of glycosides of aranciamycinone (**2**), in order to study the influence of variations in the sugar on biological activity.

Correspondence to: Dr. M. Bols, Department of Organic Chemistry, The Technical University of Denmark, DK-2800 Lyngby, Denmark.

RESULTS AND DISCUSSION

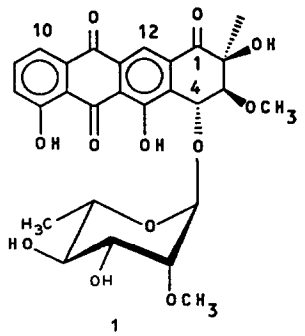
So far only a few derivatives of **1** have been reported, most notably aranciamycinone (**2**), prepared by acidic hydrolysis of **1**[1]. Aranciamycinone (**2**) was found to have a similar level of antitumor activity but did not inhibit collagenase. This indicated that the sugar played an important role in the collagenase inhibition; to study this, we decided to prepare a number of glycosides of **2**.

Glycosylation of **2** has not been reported, but coupling of the racemic 3-demethoxy analogue of **2** with bis(3-*N*-4-*O*-trifluoroacetyl)-L-daunosaminyl chloride and silver triflate has been achieved[6]. Silver triflate has been used as a glycosylation promoter with[7] or without[8] an acid acceptor; an acid acceptor is normally advantageous[9]. However, attempted glycosylation of **2** with silver triflate and either 2,3,4-tri-*O*-acetyl-α-L-rhamnosyl bromide[10] or 2,3,4,6-tetra-*O*-acetyl-α-D-glucosyl bromide in the presence of 2,4,6-collidine resulted only in orthoester formation. When collidine was omitted, glycosides were formed, together with aranciamycinone 4-acetate derived from the orthoester. As acidic conditions were necessary, we decided to try the simpler procedure of treating sugar acetates with the alcohol **2** in the presence of trimethylsilyl triflate (Me_3SiOTf)[11]. When **2** reacted with 1,2,3,4-tetra-*O*-acetyl-α-L-rhamnose and Me_3SiOTf, the α-glycoside **3a** was isolated in 55% yield. Deacetylation with sodium methoxide gave the unprotected glycoside **3** (87%). Regarding the configuration at the anomeric carbon in **3** and **3a**, the low-field chemical shift of H-1′ in the ^1H NMR spectrum showed that this proton was equatorial. As the $^4C_1$ conformation is unlikely to be present to any large extent for L-rhamnose, this was regarded as evidence for the α configuration.

Reaction of **2** with 1,2,3,4,6-penta-*O*-acetyl-D-glucose and Me_3SiOTf gave the acetylated β-glycoside **4a** (19%) together with aranciamycinone 4-acetate (36%). Deacetylation gave the β-D-glucoside **4** in 73% yield. The large value of $J_{1',2'}$ in **4** and **4a** is consistent only with the β configuration.

Similar glycosylation of **2** with tetra-*O*-acetyl-β-D-ribose, tetra-*O*-acetyl-α,β-D-xylose, and tetra-*O*-acetyl-α-L-fucose gave the β-D-riboside **5a** (67%), β-D-xyloside **6a** (27%), and α-L-fucoside **7a** (37%), respectively. The compounds were deacetylated satisfactorily in 75–93% yield to give the corresponding unprotected glycosides **5**, **6**, and **7**. The β-D configuration of **6**/**6a** was evidenced by the large proton coupling between H-1′ and H-2′ (7.1 Hz). The β-D configuration of **5**/**5a** was established by two facts: first, the low-field chemical shift of H-1′ (δ 5.5), showing this proton to be equatorial; secondly, the small couplings in **5a** between H-4′ and the H-5′ protons ($J_{4',5'b}$ 5.1 Hz, $J_{4',5'a}$ 3.1 Hz), showing that the pyranose ring was predominantly in the $^1C_4$ conformation. Similarly, the α-L configuration of **7**/**7a** was established by these facts: H-1 equatorial (δ 5.62), and the pyranose ring in the $^1C_4$ conformation ($J_{2',3'}$ 10.3 Hz).

Glycosylation with the rhamnose analogues 1,3,4-tri-*O*-acetyl-2-azido-2,6-dideoxy-L-mannopyranose, 1,3,4-tri-*O*-acetyl-2-deoxy-L-*arabino*-hexopyranose, 1,2,4-tri-*O*-acetyl-3-deoxy-L-*arabino*-hexopyranose, and 1,2,3-tri-*O*-acetyl-4-deoxy-L-*lyxo*-



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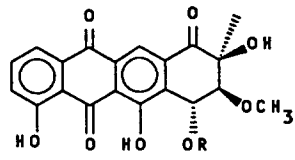

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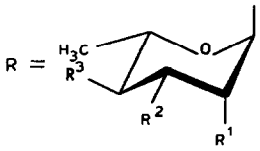
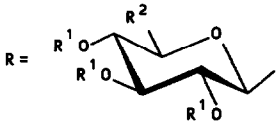




3 $R^1=R^2=R^3=OH$

3a $R^1=R^2=R^3=OAc$

8 $R^1=N_3$, $R^2=R^3=OH$

8a $R^1=N$, $R^2=R^3=OAc$

9 $R^1=H$, $R^2=R^3=OH$

9a $R^1=H$, $R^2=R^3=OAc$

10 $R^2=H$, $R^1=R^3=OH$

10a $R^2=H$, $R^1=R^3=OAc$

11 $R^3=H$, $R^1=R^2=OH$

11a $R^3=H$, $R^1=R^2=OAc$

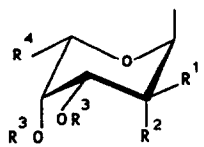
4 $R^1=H$, $R^2=CH_2OH$

4a $R^1=Ac$, $R^2=CH_2OAc$

6 $R^1=H$, $R^2=H$

6a $R^1=Ac$, $R^2=H$

R =



5 $R^1=R^3=R^4=H$, $R^2=OH$

5a $R^1=R^4=H$, $R^2=OAc$, $R^3=Ac$

7 $R^2=R^3=H$, $R^1=OH$, $R^4=CH_3$

7a $R^2=H$, $R^1=OAc$, $R^3=Ac$, $R^4=CH_3$

hexopyranose[12] also proceeded smoothly to give the expected α-glycosides **8a** (48%), **9a** (46%), **10a** (74%), and **11a** (55%), respectively. Whereas the azido analogue had a reactivity much like that of rhamnose, the deoxy analogues reacted faster, especially the 2-deoxy analogue. Also, the 2-deoxyglycoside **9a** was unstable under the reaction conditions; after 1 h at 25°, only aglycon was recovered. When run for 1 h at −15°, the reaction gave a satisfactory yield of **9a** (46%).

TABLE I

IC_{50}-values (μM) of aranciamycinone glycosides for inhibition of *Clostridium histolyticum* collagenase and Yoshida tumor cells[13]

Compound	Collagenase [a]	Tumor cell DNA synthesis [b]
Aranciamycinone (**2**)	19	0.6
Rhamnoside (**3**)	2.8	7.1
Glucoside (**4**)	8.4	16
Riboside (**5**)	6.3	2.4
Xyloside (**6**)	4.0	3.0
Fucoside (**7**)	13	4.5
2-Azidorhamnoside (**8**)	1.0	0.53
2-Deoxyrhamnoside (**9**)	3.5	2.2
3-Deoxyrhamnoside (**10**)	7.9	3.2
4-Deoxyrhamnoside (**11**)	8.9	2.8
Aranciamycin (**1**)	0.37	2.2

[a] Enzyme action on the synthetic substrate 4-phenylazobenzyloxycarbonyl-L-Pro-L-Leu-Gly-L-Pro-D-Arg for 30 min at 37° and pH 7.1. Data from spectrophotometric measurement (320 nm) of an EtOAc extract. [b] Cells incubated with test compounds for 24 h, at 37°. Data from the assessment of ^{3}H-thymidine incorporation at the end of the period.

The protected glycosides **8a**, **9a**, **10a**, and **11a** were deacetylated to give **8**, **9**, **10**, and **11**, respectively, in 71–80% yield. The α configurations of the products were proven by the low-field chemical shifts for the equatorial H-1′, which were very similar to that of the anomeric proton in **3**. Also, products **8**, **9**, and **10** had a large $J_{4',5'}$, showing that they were predominantly in the $^1C_4$ conformation.

Compounds **3**–**11** were tested for collagenase and tumor cell DNA-synthesis inhibition[13]. The results are shown in Table I. All glycosides showed higher collagenase inhibition than aglycon **2**, but lower inhibition than **1**. The 2′-analogues **3**, **8**, and **9** showed the highest inhibition. Almost all the glycosides showed lower tumor cell inhibition than both **1** and **2**. The apparent trend was that tumor cell inhibition fell with increased polarity, the glucoside **4** having the lowest activity.

In conclusion, this paper has shown that trimethylsilyl triflate promoted coupling of an anthracyclin aglycon with a wide range of different sugar acetates. The products were normally 1,2-*trans* glycosides, with the exception of the fucoside **7** where the anomerically more stable product was obtained. The advantage of the method is its simplicity and wide scope.

EXPERIMENTAL

General.—Melting points are uncorrected. NMR spectra were recorded with a Bruker AC-300 instrument with Me_4Si as internal reference. Optical rotations were measured on a Perkin–Elmer PE241 apparatus. TLC was performed on Silica Gel 60 F_{254} plates (Merck). Elemental analyses were performed by the Microanalytical Laboratory, Leo Pharmaceutical Products. For NMR data marked

with *, the resonances of the aglycon are essentially identical to those listed earlier. The assignment of spectral resonances marked with # may be reversed. Aranciamycin (**1**) was obtained by fermentation of a strain of *Streptomyces griseoflavus*[13]; aranciamycinone (**2**) was obtained by acidic hydrolysis of **1** as described by Keller-Schierlein et al.[1].

Glycosylation of aranciamycinone.—General procedure. The reaction was carried out under standard anhydrous conditions. To a solution of aranciamycinone (**2**; 100 mg, 0.26 mmol) and the peracetylated sugar (0.39 mmol, 1.5 equiv) in CH_2Cl_2 (10 mL) at −15° was added trimethylsilyl triflate (60 μL, 0.34 mmol). The mixture was stirred for the indicated time at the stated temperature. Water (50 mL) was added, and the mixture extracted with CH_2Cl_2 (3 × 10 mL). The combined organic layers were dried ($MgSO_4$) and concentrated to an orange-yellow residue. Flash chromatography in 66:33:1 EtOAc–pentane–HCOOH gave the acetylated glycoside, normally as an orange amorphous solid.

Deacetylation was carried out by dissolving the acetate in NaOMe (0.1 M, 1 mL/10 mg of acetate) and keeping it for 3 h at 25°. Aqueous AcOH (5%, 4 mL/10 mg of acetate) was added, and the mixture was extracted 6 times with equal volumes of EtOAc. The combined organic layers were dried (Na_2SO_4) and concentrated to give the desired glycoside as a crystalline or amorphous solid.

(2S,3S,4R)-4-(6-Deoxy-α-L-mannopyranosyloxy)-3,4-dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-1,6,11-(2H)naphthacenetrione (**3**).—Reaction of **2** (100 mg) with 1,2,3,4-tetra-*O*-acetyl-α-L-rhamnopyranose (121 mg, 0.36 mmol, 1.4 equiv) for 16 h at 25° gave triacetate **3a** (94 mg, 55%), $[\alpha]_D^{20}$ + 72° (*c* 0.18, $CHCl_3$). NMR data ($CDCl_3$): ^{1}H, δ 1.26 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 1.55 (s, 3 H, 2-Me), 1.88 (s, 3 H, AcO), 2.00 (s, 3 H, AcO), 2.15 (s, 3 H, AcO), 3.49 (s, 3 H, OMe), 3.70 (d, 1 H, $J_{3,4}$ 2.3 Hz, H-3), 4.04 (m, 1 H, H-5'), 5.09 (m, 1 H, H-4'), 5.12 (m, 1 H, H-3'), 5.14 (d, 1 H, H-4), 5.32 (dd, 1 H, $J_{2',3'}$ 2.8 Hz, H-2'), 5.45 (d, 1 H, $J_{1',2'}$ 1.7 Hz, H-1'), 7.23 (dd, 1 H, $J_{8,9}$ 8.4 Hz, $J_{8,10}$ 1.1 Hz, H-8), 7.65 (dd, 1 H, $J_{9,10}$ 7.4 Hz, H-9), 7.76 (dd, 1 H, H-10), 8.26 (s, 1 H, H-12), 11.74 (s, 1 H, OH), 12.59 (s, 1 H, OH).

Compound **3a** (50 mg) was deacetylated as described above, to give a residue (50 mg) from which the desired glycoside **3** (35 mg, 87%) crystallized with MeOH; mp 156–158°; $[\alpha]_D^{20}$ +194° (*c* 0.04, MeOH). NMR data [$(CD_3)_2CO$]: ^{1}H, δ 1.37 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 1.51 (s, 3 H, Me-2), 3.6 (m, 1 H, H-4'), 3.6 (s, 3 H, OMe), 3.81 (d, 1 H, $J_{3,4}$ 2.5 Hz, H-3), 3.85–4.25 (m, 3 H, H-2',3',5'), 4.59 (bs, 1 H, HO-2), 5.24 (d, 1 H, H-4), 5.53 (s, 1 H, H-1'), 7.40 (dd, 1 H, $J_{8,9}$ 8.1 Hz, $J_{8,10}$ 1.2 Hz, H-8), 7.83 (dd, 1 H, $J_{9,10}$ 7.4 Hz, H-10), 7.88 (dd, 1 H, H-9), 8.20 (s, 1 H, H-12); ^{13}C, δ 18.1 (C-6'), 23.7 (Me), 60.4 (OMe), 71.1 (C-5'#), 71.8 (C-2'#), 72.5 (C-3'#), 72.5 (C-4), 73.4 (C-4'#), 78.0 (C-2), 87.1 (C-3), 105.4 (C-1'), 116.7, 117.0, 120.0, 120.7, 125.5, 134.6, 135.0, 137.0, 138.9 (10 Ar), 163.5, 163.5 (C-5,7), 182.0 (C-11), 194.0 (C-6), 200.0 (C-1). *Anal.* Calcd for $C_{26}H_{26}O_{12} \cdot 2CH_3OH$: C, 56.56; H, 5.76. Found: C, 56.95; H, 5.73.

(2S,3S,4R)-4-(β-D-Glucopyranosyloxy)-3,4-dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-1,6,11(2H)-naphthacenetrione (**4**).—Reaction of **2** (50 mg) with 1,2,3,4,6-

penta-*O*-acetyl-β-D-glucopyranose (100 mg, 0.26 mmol, 2.0 equiv) and Me_3SiOTf (40 μL) for 18 h at 25°, as described above, gave the tetra-acetate **4a** (18 mg, 19%). Aranciamycinone 4-acetate (20 mg, 36%) was also isolated from a faster-moving fraction. NMR data ($CDCl_3$): ^1H*, δ 1.87 (s, 3 H, AcO), 2.01 (s, 3 H, AcO), 2.07 (s, 3 H, AcO), 2.11 (s, 3 H, AcO), 3.91 (ddd, 1 H, $J_{5',6'a}$ 2.4 Hz, $J_{5',6'b}$ 5.4 Hz, $J_{4',5'}$ 10.1 Hz, H-5'), 4.23 (dd, 1 H, $J_{6'b,6'a}$ 12.2 Hz, H-6'b), 4.32 (dd, 1 H, H-6'a), 5.01 (dd, 1 H, $J_{1',2'}$ 7.9 Hz, $J_{2',3'}$ 9.5 Hz, H-2'), 5.12 (t, 1 H, $J_{3',4'}$ 9.4 Hz, H-4'), 5.22 (d, 1 H, H-1'), 5.33 (t, 1 H, H-3'). ^{13}C*, δ 20.6–20.7 (4 Ac), 62.3 (C-6'), 68.6 (C-4'#). 71.5 (C-2'#), 71.6 (C-5'#), 72.2 (C-3'#), 102.7 (C-1'), 169–170.5 (4 Ac).

Compound **4a** (18 mg) was deacetylated as described above, except that, after the acidification with AcOH, extraction was omitted, and the solution was concentrated and purified by flash chromatography (99:1 EtOAc–HCOOH followed by 83:16:1 EtOAc–MeOH–HCOOH) to give amorphous **4** (10 mg, 73%), $[\alpha]_D^{20}$ +51° (*c* 0.15, MeOH). NMR data (CD_3OD): ^1H*, δ 3.2–3.7 (m, 4 H, H-2',3',4',5'), 3.79 (dd, 1 H, $J_{5',6'b}$ 5.7 Hz, $J_{6'b,6'a}$ 11.9 Hz, H-6'b), 3.98 (dd, 1 H, $J_{5',6'a}$ 1.9 Hz, H-6'a), 5.01 (d, 1 H, $J_{1',2'}$ 7.8 Hz, H-1'). *Anal.* Calcd for $C_{26}H_{26}O_{13} \cdot 4H_2O$: C, 50.49; H, 5.54. Found: C, 50.55; H, 5.31.

(2S,3S,4R)-3,4-Dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-4-(β-D-ribopyranosyloxy)-1,6,11(2H)-naphthacenetrione (**5**)—Reaction of **2** (100 mg) with 1,2,3,4-tetra-*O*-acetyl-β-D-ribopyranose (200 mg, 0.63 mmol, 2.6 equiv) and Me_3SiOTf (80 μL) for 24 h at 25°, as described above, gave the crystalline triacetate **5a** (112 mg, 67%); mp 226–230°; $[\alpha]_D^{20}$ +92° (*c* 0.1, $CHCl_3$). NMR data ($CDCl_3$): ^1H*, δ 2.09 (s, 3 H, AcO), 2.10 (s, 3 H, AcO), 2.14 (s, 3 H, AcO), 3.98 (dd, 1 H, $J_{4',5'b}$ 5.1 Hz, $J_{5'a,5'b}$ 12.5 Hz, H-5'b), 4.17 (dd, 1 H, $J_{4',5'a}$ 3.1 Hz, H-5a'), 5.1–5.2 (m, 2 H, H-2',4'), 5.35 (t, 1 H, $J_{2',3'}$ 3.5 Hz, $J_{3',4'}$ 3.5 Hz, H-3'), 5.50 (d, 1 H, $J_{1',2'}$ 4.1 Hz, H-1'); ^{13}C*, δ 20.4–20.7 (3 Ac), 61.8 (C-5'), 65.9 (C-3'#), 66.3 (C-4'#), 68.0 (C-2'#), 101.6 (C-1'), 169–170.5 (4 Ac).

Compound **5a** (14 mg) was deacetylated as described above, to give the glycoside **5** (9 mg, 80%); $[\alpha]_D^{20}$ +167° (*c* 0.02, acetone). NMR data [$(CD_3)_2CO$]: ^1H*, δ 3.70 (bs, 1 H) and 3.79 (bs, 1 H, H-2',3'), 3.85–4.0 (m, 2 H, H-4',5'b), 4.08 (dd, 1 H, $J_{4',5'a}$ 1.9 Hz, $J_{5'a,5'b}$ 13.1 Hz, H-5'a), 5.52 (d, 1 H, $J_{1',2'}$ 3.3 Hz, H-1'); ^{13}C*, δ 65.9 (C-5'), 67.5 (C-3'#), 70.4 (C-4'#), 73.0 (C-2'#), 106.0 (C-1'). *Anal.* Calcd for $C_{25}H_{24}O_{12} \cdot 3H_2O$: C, 52.63; H, 5.30. Found: C, 52.65; H, 5.28.

(2S,3S,4R)-3,4-Dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-4-(β-D-xylopyranosyloxy)-1,6,11(2H)-naphthacenetrione (**6**).—Reaction of **2** (100 mg) with 1,2,3,4-tetra-*O*-acetyl-D-xylopyranose (200 mg, 0.63 mmol, 2.6 equiv; 1:1 anomeric mixture) and Me_3SiOTf (80 μL) for 6 h at 25°, as described above, gave the triacetate **6a** (45 mg, 27%). NMR data ($CDCl_3$): ^1H*, δ 1.93 (s, 3 H, AcO), 2.01 (s, 3 H, AcO), 2.10 (s, 3 H, AcO), 3.6 (m, 1 H, H-5'a), 4.27 (dd, 1 H, $J_{4',5'b}$ 4.9 Hz, $J_{5'b,5'a}$ 11.9 Hz, H-5'b), 5.0 (m, 2 H, H-1',4'), 5.21 (t, 1 H), 5.27 (t, 1 H, H-2',3').

Compound **6a** (8 mg) was deacetylated as described above, to give the glycoside **6** (6 mg, 93%); $[\alpha]_D^{20}$ +90° (*c* 0.04, acetone). NMR data [$(CD_3)_2CO$]: ^1H*, δ 3.28 (t, 1 H, $J_{2',3'} = J_{3',4'} = 8.4$ Hz, H-3'), 3.45–3.65 (m, 2 H, H-2',5'), 4.0–4.1 (m, 2 H,

H-4',5'), 5.02 (d, 1 H, $J_{1',2'}$ 7.1 Hz, H-1'); ^{13}C*, δ 66.7 (C-5'), 70.8 (C-4'#), 75.0 (C-2'#), 77.3 (C-3'#), 107.1 (C-1'). *Anal.* Calcd for $C_{25}H_{24}O_{12} \cdot 2H_2O$: C, 54.35; H, 5.11. Found: C, 54.36; H, 5.23.

(2S,3S,4R)-4-(6-Deoxy-α-L-galactopyranosyloxy)-3,4-dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-1,6,11(2H)-naphthacenetrione (**7**).—Reaction of **2** (100 mg) with 1,2,3,4-tetra-*O*-acetyl-α-L-fucopyranose (205 mg, 0.62 mmol, 2.4 equiv) and Me_3SiOTf (80 μL) for 19 h at 25°, as described above, gave the triacetate **7a** (64 mg, 37%). Aranciamycin 4-acetate (52%) was also isolated from a faster-moving fraction. NMR data ($CDCl_3$): ^1H*, δ 1.28 (d, 3 H, $J_{5',6'}$ 6.5 Hz, H-6'), 1.92 (s, 3 H, AcO), 1.98 (s, 3 H, AcO), 2.22 (s, 3 H, AcO), 4.38 (q, 1 H, H-5'), 5.15–5.30 (m, 3 H, H-2',3',4'), 5.85 (d, 1 H, $J_{1',2'}$ 3.6 Hz, H-1').

Compound **7a** (64 mg) was deacetylated as described above, to give the glycoside **7** (38 mg, 75%); $[\alpha]_D^{20}$ +100° (*c* 0.06, MeOH). NMR data (CD_3OD): ^1H*, δ 1.35 (d, 3 H, $J_{5',6'}$ 6.5 Hz, H-6'), 3.66 (t, 1 H, $J_{2',3'} = J_{3',4'} = 3.2$ Hz, H-3'), 3.78 (m, 1 H, H-4'), 3.89 (dd, 1 H, $J_{1',2'}$ 4.1 Hz, $J_{2',3'}$ 10.3 Hz, H-2'), 4.19 (bq, 1 H, H-5'), 5.62 (d, 1 H, H-1'). *Anal.* Calcd for $C_{26}H_{26}O_{12} \cdot 5H_2O$: C, 50.32; H, 5.85. Found: C, 49.99; H, 5.17.

(2S,3S,4R)-4-(2-Azido-2,6-dideoxy-α-L-mannopyranosyloxy)-3,4-dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-1,6,11(2H)-naphthacenetrione (**8**).—Reaction of **2** (100 mg) with 1,3,4-tri-*O*-acetyl-2-azido-2,6-dideoxy-L-mannopyranose[12] (150 mg, 0.48 mmol, 1.8 equiv; α : β ratio 1 : 1) for 24 h at 25°, as described above, gave the diacetate **8a** (80 mg, 48%); $[\alpha]_D^{20}$ +99° (*c* 0.07, $CHCl_3$). NMR data ($CDCl_3$): ^1H*, δ 1.32 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 2.07 (s, 3 H, AcO), 2.09 (s, 3 H, AcO), 4.07 (m, 1 H, H-5'), 4.2 (bs, 1 H, H-2'), 5.20 (m, 2 H, H-3',4'), 5.55 (d, 1 H, $J_{1',2'}$ 1.6 Hz, H-1'); ^{13}C*, δ 17.3 (C-6'), 20.3–20.5 (2 Ac), 61.3 (C-2'), 67.9 (C-4'#), 70.3 (C-3'#), 70.6 (C-5'#), 101.4 (C-1'), 169.5–169.8 (2 Ac).

Compound **8a** (60 mg) was deacetylated as described above, to give the glycoside **8** (38 mg, 73%); $[\alpha]_D^{20}$ +62° (*c* 0.12, MeOH). NMR data (CD_3OD): ^1H*, δ 1.39 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 3.45 (t, 1 H, $J_{3',4'} = J_{4',5'} = 9.3$ Hz, H-4'), 3.85 (m, 1 H, H-5'), 3.89 (dd, 1 H, $J_{2',3'}$ 3.8 Hz, H-3'), 4.07 (dd, 1 H, $J_{1',2'}$ 1.7 Hz, H-2'), 5.49 (bs, 1 H, H-1'); ^{13}C*, δ 18.0 (C-6'), 65.9 (C-2'), 71.7 (C-4'#), 72.8 (C-3'#), 73.8 (C-5'#), 103.5 (C-1'). *Anal.* Calcd for $C_{30}H_{29}N_3O_{13} \cdot 3THF$ *: C, 58.94; H, 6.24. Found: C, 59.35; H, 5.85.

*(2S,3S,4R)-4-(2,6-Dideoxy-α-L-*arabino*-hexopyranosyloxy)-3,4-dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-1,6,11(2H)-naphthacenetrione* (**9**).—Reaction of **2** (100 mg) with 1,3,4-tri-*O*-acetyl-2-deoxy-L-*arabino*-hexopyranose[12] (100 mg, 0.36 mmol, 1.4 equiv; α : β ratio 1 : 2) for 1 h at −15°, as described above, gave the diacetate **9a** (72 mg, 46%); $[\alpha]_D^{20}$ +124° (*c* 0.16, $CHCl_3$). NMR data ($CDCl_3$): ^1H*, δ 1.29 (d, 3 H, H-6'), 1.90 (m, 1 H, H-2'), 1.99 (s, 3 H, AcO), 2.09 (s, 3 H, AcO), 2.37 (dd,

* The solvent content was verified by ^1H NMR spectroscopy. The analytical sample was prepared by concentrating a solution in tetrahydrofuran.

1 H, $J_{2'a,3'}$ 5.2 Hz, $J_{2'a,2'b}$ 12.2 Hz, H-2'a), 4.08 (m, 1 H, H-5'), 4.86 (t, 1 H, $J_{3',4'} = J_{4',5'} = 9.6$ Hz, H-4'), 5.10 (m, 1 H, H-3'), 5.66 (bd, 1 H, $J_{1',2'}$ 3.2 Hz, H-1').

Compound **9a** (21 mg) was deacetylated as described above, to give the glycoside **9** (14 mg, 78%), which crystallised from MeOH; mp 146–150°, $[\alpha]_D^{20}$ +142° (*c* 1.0, MeOH). NMR data (CD_3OD): ^1H*, δ 1.38 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 1.74 (dt, 1 H, $J_{2'b,3'}$ 4.0 Hz, $J_{2'b,2'a}$ 12.8 Hz, H-2'b), 2.21 (dd, 1 H, $J_{2'a,3'}$ 5.1 Hz, H-2'a), 3.06 (t, 1 H, $J_{3',4'} = J_{4',5'} = 9.2$ Hz, H-4'), 3.71 (m, 1 H, H-3'), 3.88 (m, 1 H, H-5'), 5.61 (d, 1 H, $J_{1',2'}$ 3.4 Hz, H-1'); ^{13}C*, δ 18.2 (C-6'), 39.2 (C-2'), 69.7 (C-3'#), 71.0 (C-4'#), 78.8 (C-5'#), 103.0 (C-1'). *Anal.* Calcd for $C_{30}H_{30}O_{13} \cdot H_2O$: C, 58.44; H, 5.23. Found: C, 58.72; H, 5.31.

*(2S,3S,4R)-4-(3,6-Dideoxy-α-L-*arabino*-hexopyranosyloxy)-3,4-dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-1,6,11(2H)-naphthacenetrione* (**10**).—Reaction of **2** (100 mg) with 1,3,4-tri-*O*-acetyl-3-deoxy-α-L-*arabino*-hexopyranose[12] (106 mg, 0.39 mmol, 1.5 equiv) for 1 h at 25°, as described above, gave the diacetate **10a** (116 mg, 74%), $[\alpha]_D^{20}$ +85° (*c* 1.0, $CHCl_3$). NMR data ($CDCl_3$): ^1H*, δ 1.31 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 1.82 (ddd, 1 H, $J_{2',3'b}$ 3.0 Hz, $J_{3'b,4'}$ 11.3 Hz, $J_{3'b,3'a}$ 13.6 Hz, H-3'b), 2.08 (s, 3 H, AcO), 2.14 (m, 1 H, H-3a'), 2.19 (s, 3 H, AcO), 4.04 (dq, 1 H, $J_{4',5'}$ 9.7 Hz, H-5'), 4.91 (ddd, 1 H, $J_{3'a,4'}$ 4.5 Hz, H-4'), 5.03 (bs, 1 H, H-2'), 5.43 (bs, 1 H, H-1'); ^{13}C*, δ 17.4 (C-6'), 20.7–20.9 (2 Ac), 29.1 (C-3'), 67.8 (C-2'#), 69.0 (C-4'#), 69.1 (C-5'#), 99.8 (C-1'), 169.7–169.8 (2 Ac).

Compound **10a** (100 mg) was deacetylated as described above, to give the glycoside **10** (61 mg, 71%), which crystallised from MeOH; mp 155–165°; $[\alpha]_D^{20}$ +146° (*c* 0.04, MeOH). NMR data (CD_3OD): ^1H*, δ 1.38 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 1.71 (ddd, 1 H, $J_{2',3'b}$ 2.9 Hz, $J_{3'b,4'}$ 10.2 Hz, $J_{3'b,3'a}$ 12.2 Hz, H-3'b), 2.01 (m, 1 H, H-3a'), 3.66 (dt, 1 H, $J_{3'a,4'}$ 4.3 Hz, $J_{4',5'}$ 10.2 Hz, H-4'), 3.84 (m, 1 H, H-5'), 3.95 (bs, 1 H, H-2'), 5.33 (bs, 1 H, H-1'); ^{13}C*, δ 18.2 (C-6'), 36.1 (C-3'), 68.1 (C-2'#), 69.2 (C-4'#), 72.6 (C-5'#), 104.6 (C-1'). *Anal.* Calcd for $C_{30}H_{30}O_{13} \cdot H_2O$: C, 58.44; H, 5.23. Found: C, 58.46; H, 5.15.

*(2S,3S,4R)-4-(4,6-Dideoxy-α-L-*lyxo*-hexopyranosyloxy)-3,4-dihydro-2,5,7-trihydroxy-3-methoxy-2-methyl-1,6,11(2H)-naphthacenetrione* (**11**).—Reaction of **2** (100 mg) with 1,3,4-tri-*O*-acetyl-4-deoxy-α-L-*lyxo*-hexopyranose[12] (100 mg, 0.36 mmol, 1.4 equiv) for 3 h at 25°, as described above, gave the diacetate **11a** (85 mg, 55%), $[\alpha]_D^{20}$ +189° (*c* 1.0, $CHCl_3$). NMR data ($CDCl_3$): ^1H*, δ 1.38 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 1.88 (m, 2 H, H-4'a,4'b), 1.98 (s, 3 H, AcO), 2.22 (s, 3 H, AcO), 4.24 (m, 1 H, H-5'), 5.12 (m, 1 H, H-3'), 5.24 (bs, 1 H, H-2'), 5.56 (d, 1 H, $J_{1',2'}$ 1.5 Hz, H-1'); ^{13}C*, δ 20.6 (C-6'), 20.7–20.8 (2 Ac), 32.9 (C-4'), 65.7 (C-5'#), 66.5 (C-3'#), 67.2 (C-2'#), 102.1 (C-1'), 169.7–169.8 (2 Ac).

Compound **11a** (74 mg) was deacetylated as described above, to give the glycoside **11** (51 mg, 80%); $[\alpha]_D^{20}$ +120° (*c* 0.23, MeOH). NMR data (CD_3OD): ^1H*, δ 1.34 (d, 3 H, $J_{5',6'}$ 6.2 Hz, H-6'), 1.75 (m, 2 H, H-4'a,4'b), 3.87 (bs, 1 H, H-2'), 3.91 (m, 1 H, H-3'), 4.18 (m, 1 H, H-5'), 5.51 (bs, 1 H, H-1'); ^{13}C*, δ 21.5 (C-6'), 36.7 (C-4'), 67.1 (C-5'#), 67.6 (C-3'#), 69.8 (C-2'#), 106.6 (C-1'). *Anal.* Calcd for $C_{30}H_{30}O_{13} \cdot H_2O$: C, 58.44; H, 5.23. Found: C, 58.22; H, 5.10.

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REFERENCES

1 W. Keller-Schierlein, J. Sauerbier, U. Vogler, and H. Zaehner, *Helv. Chim. Acta*, 53 (1970) 779–789.
2 A. Fujiwara, M. Tozoe, T. Hoshino, Y. Sekine, and M. Fujiwara, *Tennen Yuki Kagobutsu Toronkai*, 22 (1979) 448–455.
3 W. Keller-Schierlein and A. Mueller, *Experientia*, 26 (1970) 929–930.
4 M.E. Bergy and F. Reusser, *Experientia*, 23 (1967) 254–255.
5 T.F. Brodasky and F. Reusser, *J. Antibiot.*, 27 (1974) 809–813.
6 K. Krohn and E. Broser, *J. Org. Chem.*, 49 (1984) 3766–3771.
7 J. Annarp and J. Lönngren, *J. Chem. Soc., Perkin Trans. 1*, (1981) 2070–2074.
8 M. Schultz and G. Zörkler, *Liebigs Ann. Chem.*, (1989) 393–395.
9 H. Paulsen, *Angew. Chem.*, 94 (1982) 184–201; *Angew. Chem. Int. Ed. Engl.*, 21 (1982) 155–173.
10 H. Muehlemann and P.J. Lyk, *Pharm. Acta Helv.*, 45 (1970) 728–753.
11 T. Ogawa, K. Beppu, and S. Nakabayashi, *Carbohydr. Res.*, 93 (1981) c6–c9.
12 M. Bols, I. Lundt, and E.R. Ottosen, *Carbohydr. Res.*, 222 (1991) 141–149.
13 M. Bols, L. Binderup, J. Hansen, and P. Rasmussen, *J. Med. Chem.*, 35 (1992) 2768–2771.