NEW DAUNORUBICIN ANALOGS

3-AMINO-2,3,6-TRIDEOXY-α- AND β-D-*ARABINO*- AND 3,6-DIAMINO-2,3,6-TRIDEOXY-α-D-*RIBO*-HEXOPYRANOSIDES OF DAUNOMYCINONE*

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Glycosidation of 2,3,6-trideoxy-3-trifluoroacetamido-4-*O*-trifluoroacetyl-α-D-*arabino*-hexopyranosyl chloride (**19**) (or the corresponding 4-*p*-nitrobenzoate, **20**) with daunomycinone under KOENIGS-KNORR conditions afforded, after separation of the anomers and removal of the protecting groups, the individual target glycosides **8** (α anomer; major product) and **9** (β; minor) in acceptable yields. In contrast, the title diamino sugar, suitably protected with *N*-trifluoroacetyl and *O*-acetyl (or *O*-*p*-nitrobenzoyl) groups, underwent stereospecific coupling to the anthracycline aglycon by the glycal procedure to give, after deprotection, the α glycoside **12**. All three analogs were assayed *in vivo* against P388 lymphocytic leukemia. They showed little (T/C 125 for **8**; T/C 115 for **9**) or no (compound **12**) activity, but were essentially devoid of toxicity at the dose-levels tested.

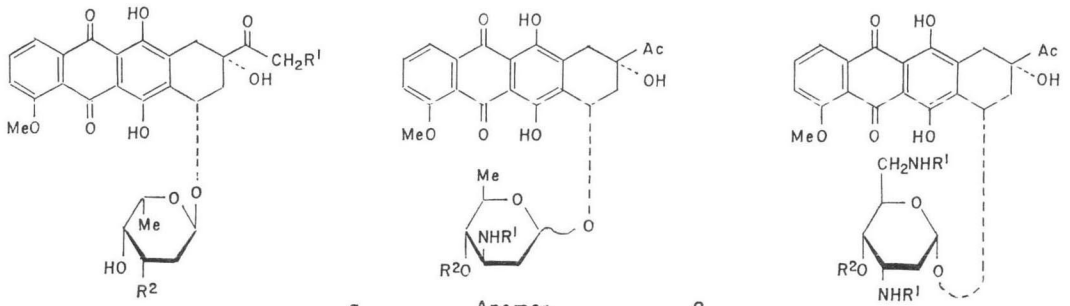
In a recent report[2], we have described the preparation of 3′-deamino-3′-hydroxydaunorubicin (**1**; NSC-284682), which displayed excellent activity against murine tumors and is currently undergoing extensive biological testing. Compound **1** is a semisynthetic analog of daunorubicin[3] (**2**; NSC-82151) in which the natural amino sugar, daunosamine[4] (3-amino-2,3,6-trideoxy-L-*lyxo*-hexose) is replaced by the corresponding 3-hydroxyl analog (2,6-dideoxy-L-*lyxo*-hexose, 2-deoxy-L-fucose). Both daunorubicin (**2**) and adriamycin[5] (doxorubicin, **3**; NSC-123127) are natural anthracycline glycosides that are potent chemotherapeutic agents[6] against a wide spectrum of human cancers. However, some deleterious side-effects, especially cardiotoxicity and mutagenicity, have limited their full scope in clinical utilization and have stimulated an extensive search for such analogs as derivatives of the natural antibiotics, and also semisynthetic and totally synthetic compounds, that might exhibit more favorable therapeutic indices than the parent drugs.

Coupling of an aglycon from the natural source with various daunosamine analogs constitutes a logical approach for semisynthetic anthracycline glycosides modified in the sugar portion. With this goal in mind, we have developed[7], in recent years, a number of preparatively useful syntheses of "fraudulent" sugars related to daunosamine, which have been subjected to coupling to daunomycinone and/or adriamycinone. The present report describes the synthesis and initial biological evaluation of three new daunomycinone glycosides [the anomeric glycosides **8** (NSC-275272) and **9** (NSC-302648) and the 3,6-diamino glycoside **12** (NSC-302048)] in which 3-amino-2,3,6-trideoxy-D-*arabino*-hexose[8] (D-acosamine) and 3,6-diamino-D-*ribo*-hexose constitute the carbohydrate components.

Chemical Synthesis

At the outset, the D-*arabino* isomer (**8**) of daunorubicin (**2**) and its β anomer **9** were obtained by

* For a preliminary presentation of part of this work, see ref. 1.



α	Anomer	β
1 $R^1 = H$; $R^2 = OH$		
2 $R^1 = H$; $R^2 = NH_2$		
3 $R^1 = OH$; $R^2 = NH_2$		
4 $R^1 = F_3CCO$; $R^2 = H$		5
6 $R^1 = F_3CCO$; $R^2 = O_2NC_6H_4CO$		7
8 $R^1 = R^2 = H$		9
		10 $R^1 = F_3CCO$; $R^2 = Ac$
		11 $R^1 = F_3CCO$; $R^2 = O_2NC_6H_4CO$
		12 $R^1 = R^2 = H$

Table 1. ^{1}H-N.m.r. spectral data of 3-amino- and 3,6-diamino-2,3,6-trideoxyhexopyranose derivatives.

Compound[a]	Chemical shifts (δ)[b] (first-order couplings, Hz, in parentheses)										
	H-1 ($J_{1,2a}$)	H-2e ($J_{1,2e}$)	H-2a ($J_{2a,3}$)	H-3 ($J_{2e,3}$)	H-4 ($J_{3,4}$)	H-5 ($J_{4,5}$)	H-6 ($J_{5,6}$)	1-OR	4-OR	NH-3 ($J_{3,NH}$)	NH-6
6[c,d]	5.52 bs[e]	←2.40—2.10 m→		~4.60 m[e]	5.01 t (10.3)	4.58 dq (10.3)	1.29 d (6.2)	[f]	8.40—7.50 m	~7.55 b[e]	—
7[c,d]	5.39 dd (8.6)	←2.40—2.10 m→ (1.8)		4.60 m	5.01 t (9.5)	4.07 dq (9.5)	1.37 d (6.2)	[f]	←8.45—7.50 m→		—
10	5.46 bs[e]	←2.20—1.80 m→		4.61 m	5.46 m	3.87 m	3.75—3.55 m	[f]	2.04 s	7.68 d (8.0)	7.05 bs
11[c]	5.49 bs	←2.30—1.85 m→		4.80 m	5.02 dd (3.1)	4.80 m (10.2)	3.75—3.50 m	[f]	8.40—8.00 m	8.20[e]	6.81 bs
14[g]	4.74 bs (W_h 7)	←2.34—1.80 m→		4.53 m	4.89 t (9.6)	3.94 dq (9.6)	1.16 d (6.2)	3.34 s	—	9.18 d (8.0)	—
17	6.38 bs (W_h 6)	←2.60—2.10 m→		4.55 m	5.00 t (9.8)	4.07 dq (9.8)	1.10 d (6.0)	—	—	9.40 d (8.0)	—

18[c,d]	6.57 m_3	←—2.60—2.40 m—→	4.90 m	5.14 t (9.4)	4.43 dq (9.4)	1.25 d (6.2)	←————8.40—8.20 m————→			—
19	6.22 bs (W_h 6)	←—2.60—2.20 m—→	←—5.10—4.60 m—→		4.37 dq (9.0)	1.14 d (6.3)	—	—	7.24 d (7.0)	—
20[e]	6.26 bd (W_h 7)	←—2.75—1.85 m—→	←—5.05—4.70 m—→		4.43 m	1.32 d (6.2)	—	8.40—8.10 m	6.56 d (7.0)	—
24[h]	4.70 dd (3.6)	2.04 ddd (1.5) 1.85 m_6 (3.6)	←—3.70—3.40 m . . . . . and 3.20—2.80 m—→ (2.7)				3.32 s	2.27 bs	2.27 bs	2.27 bs
25	4.79 bs (W_h 6)	←—2.10—1.85 m—→	4.00—3.40 m	4.43 dd[i] (4.5)	←—4.00—3.40 m—→ (9.0)		3.35 s	2.92 bs	7.82 d (7.0)	8.32 bs
26	4.90—4.55 m	←—2.30—1.80 m—→	←—4.90—4.55 m—→		3.97 ddd (10.0)	3.70—3.50 m (7.0; 3.0)	3.41 s	—	7.86 d (7.8)	6.91 bs
27	5.10—4.70 m	←—2.30—2.00 m—→	←—5.10—4.70 m—→		4.16 m	3.80—3.50 m	3.44 s	8.20—8.00 m	7.99 d (7.0)	6.94 bs
28[j]	6.23 bs (W_h 7)	←—2.40—1.90 m—→	←—4.85—4.60 m—→		4.09 m	3.70—3.40 m	2.12 s	1.99 s	7.49 d (8.0)	7.26 bs
29	6.26 bs	←—2.30—1.80 m—→	←—5.05—4.70 m—→		4.25 m	3.70—3.50 m	2.14 s	8.30—8.00 m	7.66 d (8.0)	7.36 bs
31	5.41 bs	←—2.30—2.00 m—→	←—4.85—4.60 m—→		4.23 m	3.70—3.40 m	4.85—4.60 m	2.01 s	8.24 d (8.2)	7.06 bs
32	6.26 bd (W_h 7)	←—2.30—1.90 m—→	←————5.10. . . . . 4.60 m—→			3.75—3.45 m	—	2.15 s	7.50 d (7.0)	—
33	6.52 d	4.98 dd (4.5) —	←—4.85—4.60 m—→ (10.2)		4.02 m	3.75—3.45 m	—	2.02 s	6.84 d (7.8)	6.99 bs
34[d]	6.61 d	5.36 dd (4.8) —	←—5.05—4.85 m—→ (10.0)		4.53 m	3.85—3.70 m	—	←—8.40—8.10 m—→		8.50 bs

[a] 100-MHz continuous-wave spectra in chloroform-*d*, unless otherwise stated. [b] Signal multiplicities: b, broadened; d, doublet; m, multiplet; m_x, x-line pattern; q, quartet; s, singlet; t, triplet. [c] 90-MHz FOURIER-transform spectrum. [d] In acetone-d_6. [e] Partly obscured signal. [f] Chemical shifts of the anthracycline portion are but little affected by the various sugar species; as representative data those of compound **10** are given: 14.51 and 13.12 (two s, 1 each, chelated OH), 8.01 (dd, 1, $J_{1,2}$=7.8 Hz, $J_{1,3}$=1.2 Hz, H-1), 7.79 (t, 1, $J_{2,3}$=7.8 Hz, H-2), 7.41 (dd, 1, H-3), 5.46 (narrow m, 1, H-7), 4.05 (s, 3, OMe), 3.17 (d, 1, $J_{10A,10B}$=19.0 Hz, H-10A), 2.86 (d, 1, H-10B), 2.50~2.10 (m, partly obscured by CAc signal, H-8A, H-8B). [g] In 3:1 chloroform-*d*-methyl sulfoxide-d_6. [h] $J_{2e,2a}$=14.5 Hz. [i] After exchange of the hydroxyl proton by deuterium. [j] Data taken from the n.m.r. spectrum of an almost pure (~90%) α anomer.

coupling daunomycinone with 2,3,6-trideoxy-3-trifluoroacetamido-4-*O*-trifluoroacetyl-α-D-*arabino*-hexopyranosyl chloride (**19**) under KOENIGS-KNORR conditions (HELFERICH modification) to give, after removal of the *O*-trifluoroacetyl group by methanolysis, an anomeric mixture of the partially protected glycosides **4** and **5**. Separation of these by column chromatography on silica gel and deprotection of the individual anomers by mild alkaline treatment afforded the target compounds **8** (α anomer) and **9** (β), isolated as their crystalline hydrochlorides, but obtained in disappointingly low yields (17% and 7%, respectively).

The key intermediate in this sequence of reactions, the protected chloride **19**, was prepared by a series of high-yielding steps starting from methyl 3-acetamido-2,3,6-trideoxy-α-D-*arabino*-hexopyranoside[8] (**13**). Successive treatment of **13** with aqueous barium hydroxide and trifluoroacetic anhydride afforded the hitherto unknown *N*,*O*-trifluoroacetyl derivative **14** as featherlike crystals in 94% yield. Removal of the *O*-trifluoroacetyl group with methanol furnished the previously described[8] glycoside **15**, which was hydrolyzed[8] with aqueous acetic acid to give the reducing sugar **16**. Subsequent treatment of **16** with trifluoroacetic anhydride gave the crystalline, peracylated derivative **17**, whose anomeric configuration (α) was readily established by ^{1}H-n.m.r. spectroscopy (broad singlet at δ 6.38 for H-1; $W_h \sim 6$ Hz; compare Table 1). Preparation of the L-enantiomer of **17** has been reported[9] by ARCAMONE and his coworkers by a route from a daunosamine precursor, but no data were given in their paper that would permit comparison with the D-enantiomer characterized here.

Finally, treatment of **17** with dry hydrogen chloride in ether furnished a quantitative yield of the crude glycosyl chloride **19**. Although used without further purification for glycosylation of daunomycinone, this chloride proved to be reasonably stable and it could be recrystallized from hexane to afford an analytically pure sample. The ^{1}H-n.m.r. spectral data (Table 1) for **19** corresponded fully with those reported[9] for the L-enantiomer, but no further characterization data were recorded for the latter.

Although the foregoing synthetic route was satisfactory for securing initial quantities of the desired analogs **8** and **9**, the incomplete consumption (only ~60%) of daunomycinone, even when a large excess of the chloride **19** was used, together with the tedious, low-yielding chromatographic purification required for the product-mixture, made an alternative and more-efficient route highly desirable. Such a route was ultimately found when *p*-nitrobenzoic rather than trifluoroacetic esters were used for hydroxy-group protection.

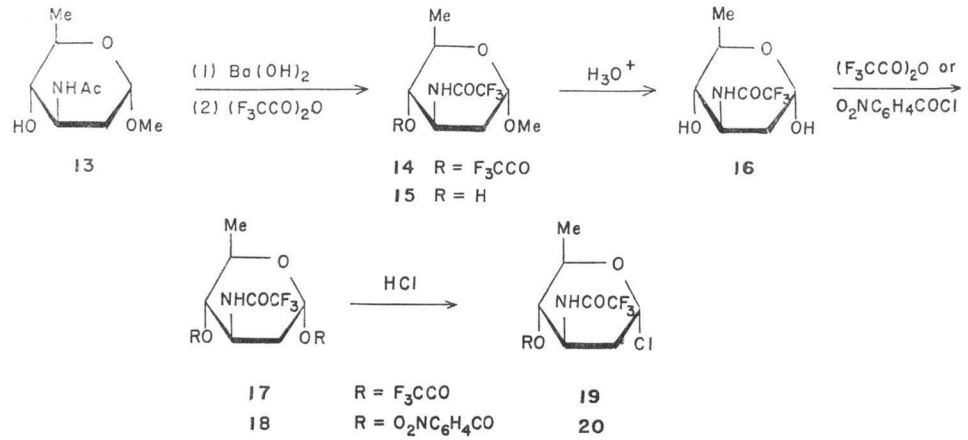
Accordingly, the reducing sugar **16** was treated with *p*-nitrobenzoyl chloride to afford in quantitative yield a 10: 1 mixture (ratio based on ^{1}H-n.m.r.-spectral analysis) of the peracylated derivative **18** and its β anomer. Although separation of the two components was not required for the subsequent conversion into the glycosyl chloride derivative **20**, the preponderant α anomer **18** could be isolated pure by fractional recrystallization.

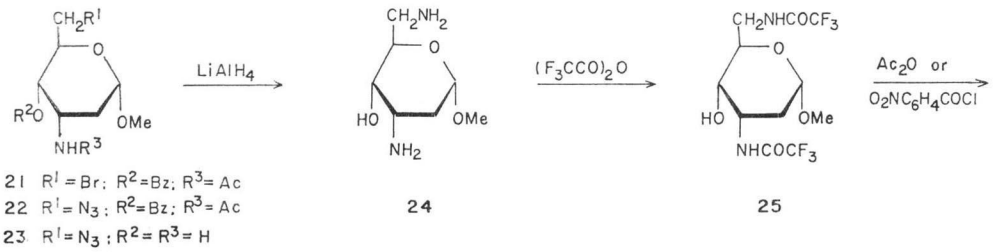
Several attempts were made, without success, to prepare from compound **18** the corresponding glycal. The latter would have been expected, by analogy with a literature precedent[10], to provide exclusively the α anomer **6** in an acid-catalyzed glycosidation reaction with daunomycinone.

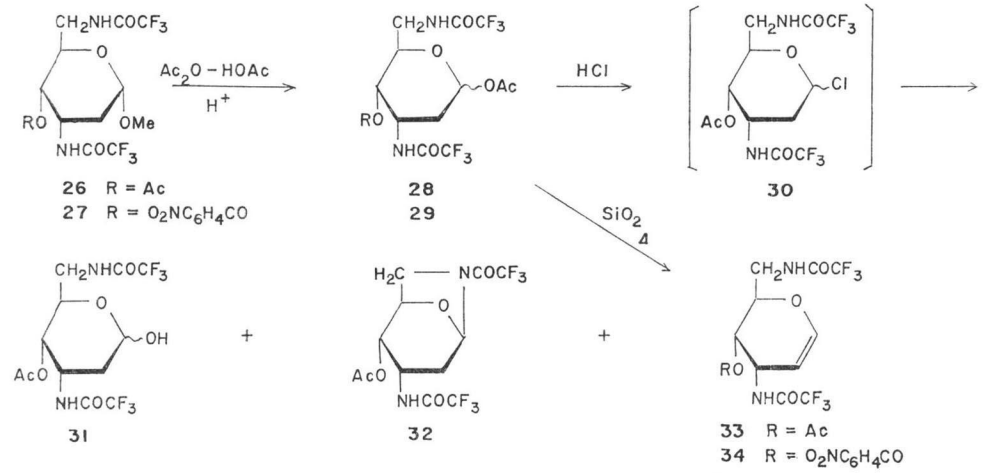
However, treatment of the mixture of crude **18** and its β anomer with dry hydrogen chloride in dichloromethane furnished, in 89% yield, the corresponding glycosyl chloride **20**, which underwent condensation with daunomycinone under KOENIGS-KNORR conditions to afford a two-component mixture (contaminated with sugar impurities) that could be readily separated by column chromato-

graphy on silica gel to give the pure, protected glycosides **6** (α anomer; 71% yield) and **7** (β anomer; 12% yield).

The individual anomers **6** and **7** were deacylated with alkali in aqueous oxolane to afford the target compounds **8** and **9**, which were isolated as their crystalline hydrochlorides in 63% and 67% yields. The overall yield for the four-step conversion from the reducing sugar **16** into the glycosides **8** and **9** totalled 40% and 7%, respectively, as compared with 14% and 6% in the first variation wherein trifluoroacetic esters had been employed for masking of the hydroxy-groups. The substantial difference in anomeric product-ratios is noteworthy and suggests a considerable measure of steric control exerted by the *p*-nitrobenzoyl group at C-4 of the coupling precursor **20**.







In a further extension of ongoing efforts in this laboratory to prepare anthracyclines having improved therapeutic properties, analogs were envisaged incorporating a natural aglycon and a daunosamine analog bearing an additional functional group (such as a carboxyl[7] or aminomethyl group) at C-5. The 3,6-diamino glycoside **24** is a most readily accessible sugar analog, and it was prepared through reduction by lithium aluminum hydride of the corresponding 6-azido precursor **23**, which, in turn, was obtained[7] by an effective preparative route involving seven steps from methyl α-D-mannopyranoside *via* the 6-bromide **21**, a key intermediate in the daunosamine synthesis[11]. Displacement of the bromide substituent in **21** by an azido group furnished[7] the fully protected derivative **22**, which was most conveniently first deprotected to give[7] the 3-aminoglycoside **23** before liberation of the second amino function (at C-6) to generate the crystalline diamino sugar **24**. The parent diamino sugar of **24**, which was further characterized as its crystalline bis(toluenesulfonate) salt, constitutes incidentally a positional isomer of nebrosamine[12] (tobrosamine), a component of the aminocyclitol antibiotic tobramycin.

The intended glycosidic coupling of the diamino sugar **24** to daunomycinone required suitable protection of the amino and hydroxyl groups prior to cleavage of the glycosidic bond and subsequent activation at C-1, in order to prevent extensive diminution in yields because of the marked tendency of 2-deoxy-D-*ribo*-hexose derivatives to form furanoid products. Therefore, after the amino groups in **24** had been masked by *N*-trifluoroacetylation to give crystalline compound **25**, the hydroxyl group at C-4 was protected in the initial approach by an *O*-acetyl group to afford the amorphous compound **26**, acetolysis of which effectively cleaved the glycosidic group. Several prior attempts to liberate the corresponding reducing sugar under acidic conditions invariably resulted in concurrent loss of protecting groups and, hence, formation of complex mixtures of tautomeric products.

Treatment of the acetolysis product **28** (a mixture of the anomeric 1-acetates) with dry hydrogen chloride in an effort to secure the 1-chloride **30** did not produce the desired glycosyl halide but gave instead a mixture of the reducing sugar **31**, the bicyclic glycosylamine **32**, and the glycal **33**. These products presumably arose from the intermediate chloride **30** by hydrolysis (**31**), by intramolecular substitution (**32**), and by elimination of hydrogen chloride (**33**). Alternatively, the reducing sugar **31** might have been the product of addition of water to the glycal **33**.

As the chloride **30** was obviously too reactive for isolation, and formation of the glycal **33** *via* the postulated intermediate (**30**) proceeded with very poor yield under the conditions described, the direct conversion of the 1-acetate **28** into **33** was investigated. The route ultimately successful required treatment of **28** with mild acid (silica gel) at an elevated temperature (140°C) to afford the desired glycal **33** as an amorphous solid in acceptable yield (60%).

Treatment of this glycal (**33**) with daunomycinone in the presence of catalytic amounts of *p*-toluenesulfonic acid proceeded satisfactorily to afford the α anomer **10** exclusively (as manifested by ^{1}H-n.m.r.-spectral analysis) in 64% yield. Attempted deprotection of the sugar moiety of **10**, however, evinced considerable difficulties, probably arising through acetyl migration from O-4 to one of the amino groups, thereby generating an extremely stable amide linkage that was resistant to all saponification reagents compatible with the rather sensitive aglycon part of the molecule. Only under stringently controlled reaction conditions, where in formation of side-products was restricted to a minimum, could the unprotected target compound **12** be obtained; it was isolated in 66% yield as its crystalline bishydrochloride.

Because of the aforementioned complications encountered with the acetyl protecting group, alternative approaches were investigated and one employing a *p*-nitrobenzoyl group for temporary masking of the 4-hydroxyl group was ultimately found to be the most successful. Consequently, the partially protected methyl glycoside **25** was converted into the *p*-nitrobenzoate **27** and carried through a similar succession of reactions (**27**→**29**→**34**) to the substituted daunomycinone glycoside **11** which, in the final step, underwent straightforward saponification without any complications of acyl migration, and afforded the desired target compound **12** in 42% overall yield from **25**.

Biological Activity

All semisynthetic anthracycline glycosides (**6**~**12**) described in this paper were evaluated by A. D. Little, Inc., under the auspices of the National Cancer Institute, for antitumor properties against transplanted P388 lymphocytic leukemia in CDF_1 mice. Generally speaking, the protected glycosides **6**, **7**, **10**, and **11** were essentially devoid of antitumor activity, but showed no significant toxicity at the dose levels tested, which ranged considerably higher ($\leq$100 mg/kg/day) than those at the toxic limit for **1** and **2**. The test results for the target compounds **8**, **9**, and **12** are listed in Table 2, together with data obtained concurrently for daunorubicin (**2**) and adriamycin (**3**) for this same experimental tumor-system. In the preliminary evaluation reported here, all three analogs (**8**, **9**, and **12**) demonstrated no marked increase in lifespan of tumor-bearing animals, although the comparatively low

Table 2. Activity[a] of daunorubicin (**2**), adriamycin (**3**), and the semisynthetic analogs **8**, **9**, and **12** on P388 lymphocytic leukemia in mice[b].

Compound	NSC No.	Dose (mg/kg)	T/C[cd] (%)	Toxic deaths	T/C[c] (%)	Toxic deaths	T/C[c] (%)	Toxic deaths
2	82151	32		5/6		6/6		6/6
		16	89	3/6	105	2/6		6/6
		8	107	1/6	125	2/6		5/6
		4	101	0/6	125	0/6		1/6
		2	109	0/6	132	0/6	105	0/6
3	123127	16	141	0/6	161	0/6		2/6
		8	151	0/6	173	0/6	94	0/6
		4	141	0/6	125	0/6	119	0/6
		2	107	2/6	144	0/6	112	0/6
		1	118	1/6	100	0/6	107	0/6
8	275272	100	125	0/6				
9	302648	50	116	0/6	110	0/3	103	1/6
		25	107 (104)	0/6	115	1/3	95	0/6
12	302048	12.5	99 (123)	0/6	110	0/3	103	0/6
		6.25	100 (102)	0/6	110	0/3	103	0/6
		3.13	98 (99)	0/6	91	2/3	100	0/6

[a] Data obtained under the auspices of National Cancer Institute, Division of Cancer Treatment, Drug Research and Development Branch.

[b] CDF_1 mice were injected ip with 10^6 P388 lymphocytic leukemia cells on day 0 and treated ip on days 5, 9, and 13 with the drug dose specified.

[c] Ratio of median survival time expressed as percent of untreated controls.

[d] Figures in parentheses from independent series of tests.

activity displayed by **1** and **2** in this particular assay is noteworthy. On the other hand, the new analogs appear to be definitely less toxic than the parent drugs and this aspect will be evaluated further.

Among the anomeric pairs of anthracycline glycosides thus far known,[9,13] all of which belong to the L-series, the α anomer has consistently displayed higher biological activity than the corresponding β anomer. In the present instance, however, the analogs **8** and **9** constitute an anomeric pair in the D-series. The equally low, albeit appreciable, activity shown by both compounds suggests that, with a more appropriate (that is, conformationally more mobile) amino sugar isomer having, for example, the D-*ribo* stereochemistry, in glycosidic conjugation with daunomycinone/adriamycinone, a β-D anomer might emerge as a worthwhile antitumor agent as the close stereochemical relationship with the parent drugs (β-D versus α-L in the appropriate conformations) may be readily visualized. Based on this hypothesis, it is unfortunate that the β anomer of the diamino glycoside **12** could not be prepared because of difficulties in securing the halide **30** as the required glycosylating agent for the (usually non-stereospecific) KOENIGS-KNORR coupling-reaction.

Experimental

T.l.c. was performed on precoated plates of Silica Gel 60 (E. Merck, Darmstadt); zones of colorless compounds were detected by spraying the plates with sulfuric acid and subsequent heating. Melting points were determined with a Thomas-Hoover apparatus and are uncorrected. Optical rotations were measured with a Perkin-Elmer 141 polarimeter. Infrared spectra were recorded with a Perkin-Elmer Model 457 grating spectrophotometer. Ultravioletvisible spectra were obtained on a Cary-14 spectrophotometer. Mass spectra were recorded with an AEI MS-9 double-focusing, high-resolution spectrometer (ionizing and accelerating potentials, 70 eV and 8 kV). N.m.r. spectra were measured at 100 MHz with a Varian HA-100 spectrometer or at 90 MHz with a Bruker HX-90 instrument; chemical shifts refer to an internal standard of tetramethylsilane ($\delta = 0.00$) and are listed, together with spin-coupling values (Hz), in Table 1. X-Ray powder diffraction data give interplanar spacings, Å, for CuKα radiation. The camera diameter was 114.59 mm. Relative intensities were estimated visually; m, moderate; s, strong; v, very; w, weak. The strongest lines are numbered (1, strongest); double numbers indicate approximately equal intensities. Analyses were performed by W. N. ROND of this Department or by Galbraith Laboratories, Inc.

Methyl 2,3,6-trideoxy-3-trifluoroacetamido-4-*O*-trifluoroacetyl-α-D-*arabino*-hexopyranoside (**14**)

A mixture of methyl 3-acetamido-2,3,6-trideoxy-α-D-*arabino*-hexopyranoside[8] (**13**; 8.0 g, 39.36 mmol) and barium hydroxide octahydrate (20 g, 63.4 mmol) in water (50 ml) was boiled for 18 hours under reflux, after which time t.l.c. (2: 3 benzene - acetone) indicated saponification to be complete. Solid carbon dioxide was added, the inorganic precipitate was filtered off, and the solution was treated with anion-exchange resin (Amberlite IRA-400, HO^-; 70 ml) and then evaporated. To the white solid residue were added dry ether (100 ml) and trifluoroacetic anhydride (20 ml, 141.9 mmol) with cooling. After 15 minutes at 0°C and 3 hours at 25°C, the clear solution was evaporated to give **14** as a white, featherlike residue which was recrystallized from hexane; yield 13.1 g (94%), m.p. 155°C, $[\alpha]_D^{22}$ +154.1° (*c* 0.9, chloroform); *m/e* (rel. intensity): 353 (0.2, M^+), 322 (3.2, M − MeO·), 208 (1.2, 322 − F_3CCO_2H), 95 (49, 208 − F_3CCONH_2), and 69 (75, F_3C^+); X-ray powder diffraction data: 8.50 m, 7.62 w, 6.12 s (1), 5.14 w, 4.81 s (3), 4.55 s (2), 4.37 w, 4.22 vw, and 4.07 m.

Anal. Calcd. for $C_{11}H_{13}F_6NO_5$ (353.22): C, 37.41; H, 3.71; N, 3.97.
Found: C, 37.64; H, 3.73; N, 4.18.

Methyl 2,3,6-trideoxy-3-trifluoroacetamido-α-D-*arabino*-hexopyranoside (**15**)

Methanolysis of the fully protected glycoside **14** (13.0 g, 36.8 mmol) in dry methanol (200 ml) for 18 hours at 25°C afforded, after recrystallization from acetone - hexane, pure **15** indistinguishable from an authentic sample[8]; yield 8.8 g (93%), m.p. 194~196°C (subl.), $[\alpha]_D^{23}$ +125.0° (*c* 0.7, chloroform).

2,3,6-Trideoxy-3-trifluoroacetamido-α-D-*arabino*-hexose (**16**)

Acid hydrolysis of the methyl glycoside **15** (8.0 g, 31.10 mmol) with aqueous acetic acid (25%, 200 ml) for 4 hours at 100°C, as described in an earlier report[8], gave the reducing sugar **16**, which was recrystallized from methanol - dichloromethane; yield 5.5 g (73%), m.p. 205°C (decomp.), $[\alpha]_D^{23}$ +35.2° (equil.; *c* 0.4, 1,4-dioxane).

2,3,6-Trideoxy-3-trifluoroacetamido-1,4-di-*O*-trifluoroacetyl-α-D-*arabino*-hexopyranose (**17**)

A solution of the reducing sugar **16** (3.0 g, 12.34 mmol) in anhydrous ether (50 ml) and trifluoroacetic anhydride (15 ml, 106.4 mmol) was kept for 18 hours at 25°C, after which time the solvent was evaporated off. Last traces of trifluoroacetic anhydride and acid were removed by evaporation of dry carbon tetrachloride from the residue. The remaining solid was recrystallized from isopropyl ether - hexane to give analytically pure **17** (which should not be dried *in vacuo* as it readily sublimes even at 18°C); yield 4.35 g (81%), m.p. 122°C, $[\alpha]_D^{22}$ +119.2° (*c* 0.9, chloroform); ν_{max} (KBr) 3320 (NH), 1810 and 1795 (ester C=O), and 1715 and 1560 cm^{-1} (amide); *m/e* (rel. intensity): 435 (1, M^+), 391 (16, M−MeCHO), 322 (12, $M-F_3CCO_2\cdot$), 207 (17, $M-2F_3CCO_2H$), 192 (55, 207−Me·), 95 (50, methylpyrylium cation), and 69 (100, F_3C^+); X-ray powder diffraction data: 9.25 s, 7.40 vw, 6.75 w, 5.53 vw, 5.21 s (3), 4.62 vs (1), 4.23 s (2), 3.95 m, and 3.67 m.

Anal. Calcd. for $C_{12}H_{10}F_9NO_6$ (435.20): C, 33.12; H, 2.32; N, 3.22.
Found: C, 33.37: H, 2.37; N, 3.26.

Although ARCAMONE and his coworkers reported[9] the preparation of the L-enantiomer of **17**, no constants were given.

2,3,6-Trideoxy-3-trifluoroacetamido-1,4-di-*O*-(*p*-nitrobenzoyl)-α-D-*arabino*-hexopyranose (**18**)

To a cold (0°C) solution of the reducing sugar **16** (4.51 g, 18.55 mmol) in pyridine (100 ml) was added *p*-nitrobenzoyl chloride (11.5 g, 62.0 mmol) and the mixture was allowed to warm to 25°C. After 4 hours sodium hydrogencarbonate (5.5 g, 65.5 mmol) was added and the mixture was poured, with mechanical stirring, into ice-water (1,000 ml). After 1 hour the precipitate that had formed was collected and dried *in vacuo* to give a 10: 1 (as judged from n.m.r. data) mixture of crude **18** and its β anomer, which could be used without further purification or separation for the following chlorination reaction; yield 10.0 g (theoretical yield). The α anomer **18** could be obtained pure by fractional recrystallization from acetone - hexane, followed by recrystallization from ethanol: m.p. 230~232°C, $[\alpha]_D^{22}$ +40.4° (*c* 0.4, acetone*); ν_{max} (KBr) 3320 (NH), 1740 and 1725 (ester C=O), 1710 and 1570 (amide), 1530 and 1355 cm^{-1} (NO_2); *m/e* (rel. intensity): 375 (4, $M-O_2NC_6H_4CO_2\cdot$), 207 (20, $M-2$ $O_2NC_6H_4CO_2H$), 192 (100, 207−Me·), 167 (18, $O_2NC_6H_4CO_2H^+$), and 150 (90, $O_2NC_6H_4CO^+$); X-ray powder diffraction data: 14.02 m, 10.64 vw, 8.11 w, 7.13 m, 6.43 w, 5.30 s (3), 4.99 vw, 4.80 m, 4.64 m, 4.47 m, 4.28 s (2), 4.11 m, 3.92 vw, and 3.65 vs (1).

Anal. Calcd. for $C_{22}H_{18}F_3N_3O_{10}$ (541.40): C, 48.81; H, 3.35; N, 7.76.
Found: C, 49.01; H, 3.05; N, 7.71.

2,3,6-Trideoxy-3-trifluoroacetamido-4-*O*-trifluoroacetyl-α-D-*arabino*-hexopyranosyl chloride (**19**)

A stream of dry hydrogen chloride was passed for 15 minutes into a cold (0°C) solution of the peracylated derivative **17** (1.74 g, 4.00 mmol) in ether (60 ml). Aftre 18 hours at +5°C, the solvent was evaporated off and dichloromethane (two 30-ml portions) was added to and evaporated from the residue to afford crude **19** (1.4 g, 98%), which could be recrystallized from hexane; yield 1.09 g (76%), m.p. 117~118°C (with sintering), $[\alpha]_D^{22}$ +146.3° (*c* 0.3, chloroform); ν_{max} (KBr) 3320 (NH), 1795 (ester C=O), 1710 and 1565 cm^{-1} (amide); *m/e* (rel. intensity): 322 (42, M−Cl·), 313 (5, M−MeCHO), 209 (55, 322−F_3CCONH_2), 192 (100, M−HCl−F_3CCO_2H−Me·), and 95 (84, methylpyrylium cation); X-ray powder diffraction data: 10.39 w, 8.38 w, 7.13 vw, 5.86 m (3,3), 5.17 m (2), 4.84 m (3,3), 4.33 s (1), 4.19 vw, and 3.89 m.

Anal. Calcd. for $C_{10}H_{10}ClF_6NO_4$ (357.64): C, 33.58; H, 2.82; N, 3.92.
Found: C, 33.45; H, 3.14; N, 3.59.

* This solvent was used because of poor solubility of **18** in chloroform.

Compound **19** should not be dried *in vacuo* as it readily sublimes, even at 18°C. The L-enantiomer of **7** has been reported[9] by ARCAMONE *et al.* who, however, did not report any constants except for ^{1}H-n.m.r. data.

2,3,6-Trideoxy-3-trifluoroacetamido-4-*O*-(*p*-nitrobenzoyl)-α-D-*arabino*-hexopyranosyl chloride (**20**)

A stream of dry hydrogen chloride was passed for 10 minutes into a cold (0°C) solution of the fully acylated derivative **18** (5.4 g, 10.0 mmol) in anhydrous dichloromethane (100 ml). The mixture was then stirred for 20 hours at 25°C with exclusion of moisture. The precipitate (*p*-nitrobenzoic acid) was filtered off and the filtrate was evaporated to give the crude, crystalline chloride **20**, which was used without delay in the subsequent coupling-step; yield 3.65 g (89%). To secure analytical data, a sample was recrystallized from dichloromethane - hexane: m.p. 150~154°C, $[\alpha]_D^{22}$ +127.0° (*c* 0.6, chloroform); ν_{max} (KBr) 3320 (NH), 1740 (ester C=O), 1710 and 1570 (amide), 1535 and 1350 cm^{-1} (NO_2); *m/e* (rel. intensity): 262 (0.2, M − Cl· − F_3CCONH_2), 260 (0.2, M − $O_2NC_6H_4CO$·), 207 (12, M − HCl − $O_2NC_6H_4CO_2H$), 192 (100, 207 − Me·), and 150 (50, $O_2NC_6H_4CO^+$); X-ray powder diffraction data: 8.93 w, 7.75 m (3,3), 6.55 w, 6.28 vw, 5.55 m, 5.21 vw, 4.99 m (3,3), 4.67 vs (1), 4.44 vw, and 4.19 s (2).

Anal. Calcd. for $C_{15}H_{14}ClF_3N_2O_6$ (410.74): C, 43.86; H, 3.44; N, 6.82.
Found: C, 44.09; H, 3.40; N, 7.05.

Methyl 3,6-diamino-2,3,6-trideoxy-α-D-*ribo*-hexopyranoside (**24**)

A mixture of the azide[7] **23** (10.1 g, 50 mmol) and lithium aluminum hydride (4 g, 105.4 mmol) in anhydrous ether (500 ml) was heated for 1 hour under reflux. After careful addition of aqueous sodium hydroxide (4%, 50 ml) to the cooled (0°C) mixture, stirring was continued for 30 minutes at 25°C. Solid potassium carbonate (50 g) and dichloromethane (500 ml) were added and the inorganic material was filtered off and thoroughly washed with dichloromethane. Evaporation of the combined filtrates afforded crude **24**, which was recrystallized from dichloromethane - hexane; yield 6.87 g (78%), m.p. 114~115°C, $[\alpha]_D^{25}$ +190.0° (*c* 1, chloroform); *m/e* (rel. intensity): 176 (0.3, M^+), 159 (16, M − HO·), and 145 (7, M − MeO·); X-ray powder diffraction data: 8.50 w, 6.88 w, 6.36 w, 5.80 s (1), 4.69 m (2), 4.21 m (3), 3.70 m, and 3.56 m.

Anal. Calcd. for $C_7H_{16}N_2O_3$ (176.22): C, 47.71; H, 9.15; N, 15.90.
Found: C, 47.64; H, 8.96; N, 15.66.

A sample of **24** (50 mg, 0.28 mmol) was dissolved in methanol (5 ml) and treated with a solution of *p*-toluenesulfonic acid monohydrate (150 mg, 0.79 mmol) in ether (25 ml). The resulting precipitate was collected and recrystallized from methanol - ether to afford the bis (toluenesulfonate) salt of **24**; yield 138 mg (95%), m.p. 203°C (decomp.), $[\alpha]_D^{25}$ +57.5° (*c* 1.5, methanol); X-ray powder diffraction data: 14.60 s, 8.66 w, 7.52 w, 7.22 w, 6.65 s, 5.96 s (3), 5.26 s (2), 4.98 w, 4.68 s (1), 4.47 m, 4.25 m, and 3.95 m.

Anal. Calcd. for $C_7H_{16}N_2O_3 \cdot 2C_7H_8O_3S$ (520.63): C, 48.44; H, 6.19; N, 5.38.
Found: C, 48.34; H, 6.18; N, 5.47.

Methyl 2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-α-D-*ribo*-hexopyranoside (**25**)

The diamine **24** (5.3 g, 30 mmol) in dichloromethane (100 ml) was treated with trifluoroacetic anhydride (20 ml, 141.9 mmol) for 4 hours at 0°C, after which time t.l.c. (1:1 beznene - acetone) indicated that all of **24** had reacted. The solution was evaporated and toluene (two 30-ml portions) was added to and evaporated from the residue, which then was dissolved in ethyl acetate (150 ml). After addition of saturated aqueous sodium hydrogencarbonate (100 ml) the mixture was vigorously stirred for 18 hours at 25°C. The aqueous layer was separated off and repeatedly extracted with ethyl acetate. The combined organic solutions were washed with water, dried (magnesium sulfate), and evaporated to afford crude **25**, which was recrystallized from ethyl acetate - hexane; yield 8.84 g (80%), m.p. 145~146°C, $[\alpha]_D^{25}$ +61.6° (*c* 1.1, acetone); ν_{max} (KBr) 3460, 3340, and 3290 (OH, NH) and 1725 and 1565 cm^{-1} (amide); *m/e* (rel. intensity): 350 (0.3, M − HO·), 337 (0.9, M − MeO·), 255 (7, M − F_3CCONH_2), 69 (39, CF_3^+), and 58 (100, $H_2C{=}CH{-}OMe^+$); X-ray powder diffraction data: 12.70 m, 6.25 w, 5.73 vs (1), 5.19 s (3), 4.47 w, 4.28 s (2), 3.97 w, 3.66 vw, 3.50 w, and 3.08 w.

Anal. Calcd. for $C_{11}H_{14}F_6N_2O_5$ (368.24): C, 35.88; H, 3.83; N, 7.61.
Found: C, 36.05; H, 4.08; N, 7.67.

Methyl 4-*O*-acetyl-2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-α-D-*ribo*-hexopyranoside (**26**)

Treatment of the hydroxyl derivative **25** (3.68 g, 10.0 mmol) with 1: 2 acetic anhydride - pyridine (30 ml) for 20 hours at 25°C afforded, after conventional processing, the title compound **26** as an amorphous solid; yield 4.0 g (98%), m.p. 60~65°C (with sintering), $[\alpha]_D^{23}$ +36.5° (*c* 1.5, chloroform); ν_{max} (film) 3375 (NH), 3100 (NH···O=C), 1760~1710 (ester C=O, Amide I), and 1550 cm^{-1} (Amide II), *m/e* (rel. intensity): 379 (9, M−MeO·), 368 (0.5, M−ketene), 350 (14, M−HOAc), 318 (19, 350−MeOH), 297 (2.5, M−F_3CCONH_2), 284 (29, M−·$CH_2NHCOCF_3$), and 224 (49, 284−HOAc).

Anal. Calcd. for $C_{13}H_{16}F_6N_2O_6$ (410.27): C, 38.06; H, 3.93; N, 6.83.
Found: C, 38.03; H, 3.82; N, 6.70.

Methyl 2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-4-*O*-(*p*-nitrobenzoyl)-α-D-*ribo*-hexopyranoside (**27**)

p-Nitrobenzoyl chloride (1.0 g, 5.4 mmol) was added to a cold (0°C) solution of the partly protected glycoside **25** (1.0 g, 2.72 mmol) in pyridine (10 ml). After 16 hours at 25°C, ethyl acetate (20 ml) and saturated, aqueous sodium hydrogencarbonate (20 ml) were added, with vigorous stirring, to the cold (0°C) solution. After 15 minutes, the organic layer was separated off, washed with water, dried (magnesium sulfate), and evaporated. Toluene (three 20-ml portions) was added to and evaporated from the residue to afford the title compound **27** as an amorphous solid; yield 1.4 g (theoretical), $[\alpha]_D^{23}$ +27.0° (*c* 0.8, chloroform); ν_{max} (film) 3380 (NH), 1735 (ester C=O and Amide I), and 1535 cm^{-1} (Amide II); *m/e* (rel. intensity): 391 (12, M−·$CH_2NHCOCF_3$), 373 (17, M−MeO·−F_3CCONH_2), 367 (8, M−$O_2NC_6H_4CO$·), and 150 (65, $O_2NC_6H_4CO^+$).

Anal. Calcd. for $C_{18}H_{17}F_6N_3O_8$ (517.34): C, 41.79; H, 3.31; N, 8.12.
Found: C, 42.15; H, 3.55; N, 8.05.

Acetolysis of **26**: 1,4-Di-*O*-acetyl-2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-D-*ribo*-hexopyranose (**28**)

A solution of the glycoside **26** (3.0 g, 7.31 mmol) in acetic anhydride (15 ml), acetic acid (10 ml), and conc. sulfuric acid (200 μl) was kept for 24 hours at 0°C, after which time t.l.c. (9: 1 chloroform - methanol) revealed the presence of two new products. Chloroform (50 ml) and sodium acetate (5 g) were added and the mixture was stirred for 30 minutes at 25°C. The solvent was evaporated off and toluene (three 20-ml portions) was added to and evaporated from the residue, which then was dissolved in chloroform (50 ml). The mixture was filtered and the filtrate evaporated to afford an anomeric mixture of the title compound **28** as a colorless foam; yield 3.04 g (95%). This material proved to be pure enough for most purposes; to secure analytical data, however, a sample was distilled *in vacuo* (bath temp. 250°C, 100 mtorr) but no attempts were undertaken to separate the two anomers; ν_{max} (film) 3425, 3330 (NH), 3100 (NH···O=C), 1760~1710 (ester C=O, Amide I), and 1550 cm^{-1} (Amide II); *m/e* (rel. intensity): 439 (0.1, M+1), 379 (1.2, M−AcO·), 318 (2.5, M−2 AcOH), and 312 (1.3, M−·$CH_2NHCOCF_3$).

Anal. Calcd. for $C_{14}H_{16}F_6N_2O_7$ (438.28): C, 38.37; H, 3.68; N, 6.39.
Found: C, 38.29; H, 3.71; N, 6.14.

1-*O*-Acetyl-2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-4-*O*-(*p*-nitrobenzoyl)-D-*ribo*-hexopyranose (**29**)

Acetolysis of the methyl glycoside **27** (1.17 g, 2.3 mmol) in acetic anhydride (36 ml), acetic acid (24 ml), and ethyl acetate (60 ml) in the presence of a catalytic amount (100 μl) of sulfuric acid for 16 hours at −18°C afforded, after conventional processing (see the preparation of **28**), the title compound **29** as a syrupy anomeric mixture that was sufficiently pure for the following elimination reaction; yield 1.20 g (97%); ν_{max} (film) 3420 and 3340 (NH), 1730 (ester C=O and Amide I), 1535 (Amide II, NO_2) and 1355 cm^{-1} (NO_2).

Anal. Calcd. for $C_{19}H_{17}F_6N_3O_9$ (545.35): C, 41.85; H, 3.14; N, 7.71.
Found: C, 42.31; H, 3.54; N, 7.53.

Attempted Chlorination of **28**

A stream of dry hydrogen chloride was passed for 20 minutes into a cold (0°C) solution of the 1-acetate **28** (1.36 g, 3.10 mmol) in dry dichloromethane (100 ml). After 18 hours at +5°C, the solvent was removed under diminished pressure (bath temp. <30°C) to afford an amorphous solid that was shown by t.l.c. (1:1 benzene - ethyl acetate) to consist of three components having Rf 0.54, 0.48, and 0.33. Column chromatography on silica gel with the t.l.c. solvent as eluant afforded the glycal **33** (360 mg, 31%; for full characterization see next paragraph).

The second, syrupy component was identified as the bicyclic derivative **32** [4-*O*-acetyl-1,6-anhydro-2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-β-D-*ribo*-hexopyranose]; yield 120 mg (10%); ν_{max} (chloroform) 3440 (NH), 1735 (ester C=O, Amide I), and 1540 cm^{-1} (Amide II); *m/e* (rel. intensity): 378 (0.5, M^+), 318 (5, M−HOAc), and 265 (2.5, $M-F_3CCONH_2$). Although the product migrated essentially as a single zone in t.l.c., an elemental analysis within acceptable limits could not be obtained.

The last compound eluted from the column was shown to be syrupy 4-*O*-acetyl-2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-D-*ribo*-hexose (**31**); yield 700 mg (57%); ν_{max} (chloroform) 3595, 3440, and 3390 (OH, NH), 1730 (ester C=O, Amide I), and 1540 cm^{-1} (Amide II), *m/e* (rel. intensity): 379 (8, M−·OH), 318 (17, $M-H_2O-HOAc$), and 266 (4, $M-\cdot OH-F_3CCONH_2$). As **31** failed to crystallize and an analysis within acceptable limits could not be obtained, an unambiguous proof of its identity was provided by conventional acetylation of **31** (420 mg, 1.06 mmol) to afford the peracetate **28**, identical (by t.l.c., n.m.r., i.r., and m.s. analysis)with an authentic sample; yield 445 mg (97%).

4-*O*-Acetyl-1,5-anhydro-2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-D-*ribo*-hex-1-enitol (**33**)

A mixture of the peracetate **28** (2.2 g, 5.0 mmol) and silica gel (40 g) in xylene (250 ml) was heated for 2 hours under reflux, after which time t.l.c. (9:1 chloroform - methanol) indicated the reaction to be complete. The silica gel was filtered off and thoroughly washed with ethyl acetate. The combined filtrates were evaporated to give pure **33** as an amorphous solid; yield 1.14 g (60%), m.p. 70~75°C (with sintering), $[\alpha]_D^{23}$ +65.0° (*c* 1.4, chloroform); ν_{max} (chloroform) 3445, 3350 (NH), 3040 (NH····O=C), 1735 (ester C=O, Amide I), 1655 (C=C), 1550 and 1530 cm^{-1} (Amide II); *m/e* (rel. intensity): 378 (0.2, M^+), 318 (0.2, M−HOAc), 205 (17, $318-F_3CCONH_2$), 192 (100, $318-\cdot CH_2NHCOCF_3$), and 168 (18, $M-F_3CCONH_2-F_3CCO\cdot$).

Anal. Calcd. for $C_{12}H_{12}F_6N_2O_5$ (378.23): C, 38.11; H, 3.20; N, 7.41.
Found: C, 38.16; H, 2.98; N, 7.13.

1,5-Anhydro-2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-4-*O*-(*p*-nitrobenzoyl)-D-*ribo*-hex-1-enitol (**34**)

A slurry of silica gel (20 g) in toluene (500 ml) was heated to boiling under reflux for 20 minutes in a flask equipped with a Dean - Stark water separator. The 1-acetate **29** (1.0 g, 1.83 mmol) then was added and heating was continued for 4 hours, after which time t.l.c. (1:1 chloroform - ether) revealed the absence of any **29** (Rf 0.14) and the formation of a new product (**34**; Rf 0.22). Processing as described for the preparation of **33** afforded a residue that was distilled *in vacuo* (bath temp. 240°C, 100 mtorr) to give **34** as a pale-yellow glass; yield 690 mg (78%), $[\alpha]_D^{25}$ +63.2° (*c* 0.5, chloroform); ν_{max} (film) 3440 (NH), 1735 (ester C=O, Amide I), 1650 (C=C), 1535 (Amide II, NO_2), and 1355 cm^{-1} (NO_2); *m/e* (rel. intensity) 318 (17, $M-O_2NC_6H_4CO_2H$), 192 (28, $318-\cdot CH_2NHCOCF_3$), and 150 (72, $O_2NC_6H_4CO^+$).

Anal. Calcd. for $C_{17}H_{13}F_6N_3O_7$ (485.30): C, 42.08; H, 2.70; N, 8.66.
Found: C, 41.97; H, 2.79; N, 8.51.

7-*O*-(Amino-2,3,6-trideoxy-α-D-*arabino*-hexopyranosyl)daunomycinone hydrochloride (**8**) and its β anomer **9**

Procedure A. Glycosidation of the Trifluoroacetylated Derivative **19**:

To a mixture of daunomycinone (398 mg, 1.0 mmol), yellow mercuric oxide (1.0 g, 4.62 mmol), mercuric bromide (280 mg, 0.78 mmol), and molecular sieve 3A (10 g) in anhydrous dichloromethane (70 ml) was added the 1-chloride **19** (1.41 g, 3.94 mmol) in four portions after 0, 14, 22, and 36 hours.

After 44 hours, methanol (50 ml) was added, the inorganic material was filtered off, and the filtrate evaporated. The resulting solid residue was dissolved in dry methanol (150 ml) and boiled for 30 minutes under reflux. Evaporation of the solvent afforded a mixture of the partially protected glycosides **4** and **5** and unreacted daunomycinone contaminated with sugar impurities, which were removed by column chromatography on silica gel with 1:1 benzene - ethyl acetate as eluant. Subsequently, the three anthracycline derivatives were separated on a second column with 200:7 chloroform - methanol to give, as fastest-migrating component, daunomycinone (145 mg, 36%). Next, the α anomer **4** (156 mg, 25%*) was isolated and finally, the β anomer **5** (60 mg, 10%*) was obtained in addition to mixed fractions (80 mg, 13%*) containing both **4** and **5**.

The partially protected glycoside **4** was treated with 0.1 M aqueous sodium hydroxide (25 ml) for 30 minutes at 25°C, whereupon t.l.c. (1:1 acetone - methanol) indicated the saponification to be complete. The pH of the solution was adjusted to 10 with 0.1 M hydrochloric acid and the product was extracted with chloroform (three 20-ml portions). The combined extracts were dried (sodium sulfate) and evaporated to give **8** as the free base. The hydrochloride salt of **8** was obtained amorphous by treating a chloroform solution of the free base with the stoichiometric amount of hydrogen chloride in dichloromethane followed by the addition of ether; yield 95 mg (17% overall yield). Recrystallization from propanol - ether gave dark-red, stout prisms of the monohydrate of **8**: m. p. 184°C (decomp.), $[\alpha]_D^{27}$ +379° (*c* 0.03, methanol); ν_{max} (KBr) 3420 (OH, NH), 1715 (C=O), and 1620 and 1580 cm^{-1} (chelated quinone); ν_{max} (ethanol) 233 nm ($\varepsilon \times 10^{-3}$ 34.9), 251 (23.4), 288 (8.3), 390 (2.8), 452 (8.5), 474 (11.4), 481 (11.5), 498 (11.7), 516 (8.3), 533 (6.6), and 583 (0.2); X-ray powder diffraction data: 11.18 vs (1), 8.46 m, 6.37 w, 6.04 m, 5.71 m, 5.40 w, 5.06 m, 4.59 m, 3.72 w, 3.51 m, 3.30 s (2), and 3.19 s (3).

Anal. Calcd. for $C_{27}H_{29}NO_{10}\cdot HCl\cdot H_2O$ (582.01): C, 55.72; H, 5.54; N, 2.41.
Found: C, 55.45; H, 5.47; N, 2.40.

In the same way as just described, the β anomer **5** was deprotected to afford, after precipitation with ether, the amorphous hydrochloride **9** (40 mg, 7.1%), which crystallized from propanol - ether as the dihydrate: m.p. 198°C (decomp.), $[\alpha]_D^{22}$ +148° (*c* 0.03, methanol); ν_{max} (KBr) 3450 (OH, NH), 1720 (C=O), 1625 and 1590 cm^{-1} (chelated quinone); ν_{max} (ethanol) 233 nm ($\varepsilon \times 10^{-3}$ 38.6), 251 (25.0), 288 (9.3), 314 (2.7), 327 (3.8), 384 (2.2), 450 (8.7), 474 (12.0), 481 (12.2), 497 (12.3), 517 (8.4), 531 (6.7), and 578 (0.4); X-ray powder diffraction data: 13.28 m (3,3), 10.84 vw, 9.98 s (1), 8.54 vw, 7.93 vw, 7.46 m, 6.34 m (2,2), 5.01 vw, 4.32 w, 3.76 m (2,2), and 3.53 m (3,3).

Anal. Calcd. for $C_{27}H_{29}NO_{10}\cdot HCl\cdot 2H_2O$ (600.03): C, 54.05; H, 5.71; N, 2.33.
Found: C, 53.61; H, 5.78; N, 2.31.

Procedure B. From the Protected Glycosides **6** and **7** by Saponification:

A solution of **6** (600 mg, 0.78 mmol) in oxolane (70 ml) and 0.1 M aqueous sodium hydroxide (70 ml) was kept for 5 hours at 0°C, after which time t.l.c. (1:1 acetone - methanol) revealed complete conversion of **6** into a single new product (Rf 0.5). The pH of the solution was adjusted to 6 with 0.1 M hydrochloric acid and oxolane was distilled off *in vacuo* (bath temp. 25°C). The remaining aqueous solution was filtered, adjusted to pH 10 with 0.1 M aqueous sodium hydroxide, and processed further as described under (A), to afford 275 mg (63%) of the hydrochloride **8**, identical in all respects with the sample already described.

Similarly, the β anomer **7** (155 mg, 0.20 mmol) was deprotected to yield 75 mg (67%) of the amorphous hydrochloride **9**, indistinguishable from the sample already described.

7-*O*-[4-*O*-(*p*-Nitrobenzoyl)-2,3,6-trideoxy-3-trifluoroacetamido-α-D-*arabino*-hexopyranosyl]daunomycinone **6** and its β Anomer **7**

To a warm (55°C) mixture of daunomycinone (996 mg, 2.50 mmol), mercuric cyanide (5.2 g, 20.6 mmol), mercuric bromide (2.4 g, 6.66 mmol), and molecular sieve 3A (12 g) in oxolane (250 ml) was added portionwise, after 1 hour, 4, 7, and 15 hours, the freshly prepared chloride **20** (a total of

* Here and in all subsequent glycosidation reactions, the yield is calculated on the basis of the original amount of daunomycinone introduced in the reaction.

3.60 g, 8.75 mmol). In the meanwhile (after 5 hours), additional mercuric cyanide (5.2 g), mercuric bromide (2.4 g), and molecular sieve (11 g) had been added. After 20 hours at 55°C, t.l.c. (4:1 benzene - acetone) revealed the absence of daunomycinone and the formation of two anthracycline glycosides having Rf 0.41 (major component) and 0.54 (minor), together with faster-migrating sugar impurities. The mixture was filtered and the filtrate evaporated. The resulting residue was dissolved in dichloromethane (100 ml), and the filtered solution was washed consecutively with 30% aqueous potassium iodide, saturated sodium hydrogencarbonate, and water, dried (magnesium sulfate), and evaporated. Column chromatography, using first 4:1 ether - petroleum ether to remove the sugar impurities and then 7:1 benzene - acetone, afforded the pure anomers. The faster migrating, minor component was recrystallized from benzene to give the pure β anomer **7**; yield 230 mg (12%), m.p. 230~232°C, $[\alpha]_D^{27}$ −81.3° (*c* 0.03, methanol); ν_{max} (KBr) 3520, 3350 (OH, NH), 1740~1710 (acetyl, Amide I), 1620, 1580 (chelated quinone, Amide II), 1530 and 1355 cm^{-1} (NO_2); λ_{max} (ethanol) 233 nm ($\varepsilon \times 10^{-3}$ 44.3), 251 (35.8), 283 (12.2), 390 (2.8), 454 (9.2), 472 (11.6), 480 (11.8), 495 (11.7), 516 (8.1), 531 (6.3), and 585 (0.3); X-ray powder diffraction data: 13.59 w, 10.71 vs (1), 8.88 vw, 8.11 s (2), 6.86 m, 6.19 m, 5.53 w, 5.27 vw, 5.05 s (3,3), and 4.75 s (3,3).

Anal. Calcd. for $C_{36}H_{31}F_3N_2O_{14}$ (772.65): C, 55.96; H, 4.04; N, 3.63.
Found: C, 55.87; H, 4.18; N, 3.56.

The more slowly-migrating, major product was recrystallized from ethyl acetate - hexane to afford the pure α anomer **6**; yield 1.37 g (71%), m.p. 254°C (with sintering), $[\alpha]_D^{27}$ +493° (*c* 0.03, methanol); ν_{max} (KB) 3460, 3340 (OH, NH), 1740~1720 (acetyl, Amide I), 1620, 1580 (chelated quinone, Amide II), 1530 and 1350 cm^{-1} (NO_2); λ_{max} (ethanol) 232 nm ($\varepsilon \times 10^{-3}$ 40.9), 250 (34.6), 286 (11.5), 388 (2.8), 452 (8.6), 473 (11.3), 480 (11.4), 498 (11.5), 517 (8.1), 533 (6.6), and 585 (0.5); X-ray powder diffraction data: 13.38 vs (1,1), 11.70 vs (1,1), 9.60 w, 8.18 m, 7.72 vw, 7.07 vw, 6.73 m, 5.90 m, 4.96 s (3,3), 4.65 s (3,3), 4.29 s (2,2), and 4.09 s (2,2).

Anal. Calcd. for $C_{36}H_{31}F_3N_2O_{14}$ (772.65): C, 55.96; H, 4.04; N, 3.63.
Found: C, 56.23; H, 4.18; N, 3.45.

7-*O*-[4-*O*-Acetyl-2,3,6-trideoxy-3,6-bis(trifluoroacetamido)-α-D-*ribo*-hexopyranosyl]daunomycinone (**10**)

To a suspension of daunomycinone (398 mg, 1.0 mmol) and *p*-toluenesulfonic acid monohydrate (10 mg) in benzene (20 ml) was added, at 55°C, a solution of the glycal **33** (1.0 g, 2.64 mmol) in dichloromethane (10 ml) in 2-ml portions after 0, 3, 6, 9, and 24 hours, while the reaction was monitored by t.l.c. (1:1 benzene - ethyl acetate). After 48 hours, the solvent was evaporated and the remaining syrup was chromatographed on silica gel with 4:1 ethyl acetate - hexane as eluant to afford, after recrystallization from ethyl acetate - hexane, analytically pure **10**; yield 495 mg (64%), m.p. 150°C (with sintering), $[\alpha]_D^{25}$ +287° (*c* 0.03, methanol); ν_{max} (chloroform) 3690, 3550 (OH), 3445, 3390 (NH), 1730 (acetyl, Amide I), 1625, 1585 (chelated quinone), and 1545 cm^{-1} (Amide II); λ_{max} (methanol) 210 nm ($\varepsilon \times 10^{-3}$ 21.7), 233 (36.6), 252 (24.4), 288 (9.0), 475 (11.6), 493 (11.2), and 526 (5.7); X-ray powder diffraction data: 13.59 s (1), 8.50 m (3), 7.43 w, 6.65 vw, and 6.04 s (2).

Anal. Calcd. for $C_{33}H_{30}F_6N_2O_{13}$ (776.60): C, 51.04; H, 3.89; N, 3.61.
Found: C, 51.03; H, 4.12; N, 3.56.

7-*O*-[2,3,6-Trideoxy-3,6-bis(trifluoroacetamido)-4-*O*-(*p*-nitrobenzoyl)-α-D-*ribo*-hexopyranosyl]daunomycinone (**11**)

By analogy with the foregoing experiment, daunomycinone (398 mg, 1.0 mmol) in benzene (20 ml) was treated, at 55°C, with the glycal **34** (1.02 g, 2.1 mmol, dissolved in 10 ml of dichloromethane) in the presence of catalytic amounts of *p*-toluenesulfonic acid monohydrate (10 mg). After chromatographic purification on silica gel with 1:1 benzene - ethyl acetate as eluant, the product was recrystallized from chloroform - hexane to give **11** as dark red, delicate crystals; yield 685 mg (78%), m.p. 158~160°C, $[\alpha]_D^{25}$ +229° (*c* 0.02, chloroform); ν_{max} (dichloromethane) 3430, 3380 (OH, NH), 1730 (acetyl, Amide I), 1630, 1580 (chelated quinone), 1530 (Amide II, NO_2), and 1350 cm^{-1} (NO_2); λ_{max} (methanol) 221 nm ($\varepsilon \times 10^{-3}$ 10.1), 236 (15.3), 252 (13.3), 284 (5.2), 450 (8.9), 476 (10.9), 493 (10.6), 526 (5.4), and 544 (1.7); X-ray powder diffraction data: 11.86 vs (1), 9.11 m, 7.34 w, 6.34 s, 6.08 m,

5.24 m, 5.08 m, 4.30 s, 4.03 s (3), 3.86 w, and 3.58 s (2).

Anal. Calcd. for $C_{38}H_{31}F_6N_3O_{15}$ (883.67): C, 51.65; H, 3.54; N, 4.76.
Found: C, 51.51; H, 3.77; N, 4.96.

7-*O*-(3,6-Diamino-2,3,6-trideoxy-α-D-*ribo*-hexopyranosyl)daunomycinone bis(hydrochloride) (**12**)

A. From the 4′-Acetate **10**.

A solution of the protected glycoside **10** (400 mg, 0.52 mmol) in methanol (100 ml) and 0.1 M aqueous sodium hydroxide (100 ml) was kept for 40 hours at +5°C, after which time t.l.c. (5:1:3 butanol - ethanol - 3% ammonium hydroxide) revealed the starting material (**10**) to be absent. Similarly, two side-products (Rf 0.33 and 0.38) that had been formed during the course of the reaction had disappeared again, and only one major component(**12**; Rf 0.24) could be detected at this point. The solution was diluted with saturated aqueous sodium hydrogencarbonate (100 ml) and extracted with chloroform (three 100-ml portions). The combined extracts were dried (potassium carbonate), filtered, and treated with a twofold stoichiometric amount of hydrogen chloride in dichloromethane. Evaporation of the solvent afforded crude **12**, which was dissolved in methanol (10 ml) and precipitated by the addition of ether to give the bis(hydrochloride) of **12** as an amorphous (diffuse X-ray powder diffraction pattern) solid; yield 210 mg (66%), m.p. 186~188°C (decomp), $[\alpha]_D^{25}$ +380° (*c* 0.0004, methanol); ν_{max} (KBr) 3400, 3250 (very broad, OH, NH), 1715 (C=O), 1620 and 1585 cm^{-1} (chelated quinone); λ_{max} (methanol) 226 nm ($\varepsilon \times 10^{-3}$ 17.4), 233 (25.9), 251 (19.4), 286 (7.9), 475 (9.2), 493 (8.9), 526 (5.2), and 572 (1.0).

Anal. Calcd. for $C_{27}H_{30}N_2O_{10} \cdot 2HCl$ (615.47): C, 52.69; H, 5.24; N, 4.55.
Found: C, 52.67; H, 5.79; N, 4.05.

B. From the 4′-*p*-Nitrobenzoate **11**.

A solution of the protected glycoside **11** (250 mg, 0.28 mmol) in methanol (100 ml) was added dropwise, at 0°C, to 0.1 M aqueous sodium hydroxide (50 ml). After 24 hours at +5°C, the mixture was processed as in the foregoing experiment to afford the bis(hydrochloride) of **12** as an amorphous powder; yield 122 mg (71%), m.p. 175~180°C.

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