Synthesis and biological activity of (*S*)-2′-fluorodaunorubicin

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Methyl 3-amino-2,3,6-trideoxy-2-fluoro-β-L-galactopyranoside was hydrolyzed to the free sugar, (*S*)-2-fluorodaunosamine hydrochloride, which was converted into the α,β-1,4-di-*O*-acetyl-*N*-trifluoroacetyl derivative and thence into the corresponding glycosyl bromide. The latter was condensed with daunomycinone, and the product was deprotected to give the title compound. The fluoroanthracycline displayed significant cytotoxicity against a number of tumor cell lines in vitro. Antitumor activity against L1210 murine leukemia in vivo was lower than that of the parent daunorubicin, but toxicity appeared to be reduced.

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On a hydrolysé l'amino-3 tridésoxy-2,3,6 fluoro-2 β-L-galactopyrannoside de méthyle en sucre libre, le chlorhydrate de la fluoro-2-(*S*) daunosamine, que l'on a transformé en dérivé di-*O*-acétyl-α,β-1,4 *N*-trifluoroacétyle et finalement en bromure de glycosyle correspondant. On a condensé ce dernier avec la daunomycinone et on a déprotégé le produit pour obtenir le composé mentionné dans le titre. La fluoroanthracycline présente une cytotoxicité in vitro importante contre un certain nombre de lignes de cellules tumorales. L'activité antitumorale in vivo contre les cellules de murine leucémique L1210 est plus faible que celle de la daunorubicine parente; toutefois, la toxicité semble être réduite.

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Introduction

In the course of a project aimed at procuring modified anthracycline antitumor agents that are substituted by fluorine in the amino sugar moiety we have described (1) a synthesis of methyl 3-amino-2,3,6-trideoxy-2-fluoro-β-L-galactopyranoside (**1**, methyl (*S*)-2-fluoro-β-L-daunosaminide). We now wish to record the coupling of that carbohydrate component with daunomycinone (**6**), which afforded (*S*)-2′-fluorodaunorubicin hydrochloride (**11**). While this work was in progress, various derivatives of **1** were also obtained elsewhere (2–5), and a coupling reaction with **6** was described (5), similar to ours in principle but different in detail.

Results and discussion

Although hydrolysis of the 2-fluoro glycoside **1** predictably was quite difficult, it succeeded by use of 4 M hydrochloric acid (6 h, reflux) and gave (*S*)-2-fluorodaunosamine hydrochloride (**2**) in 90% yield. Trifluoroacetylation of **2**, followed by methanolysis of the crude product to effect selective *O*-deacylation, furnished the trifluoroacetamido sugar **3** in yields of ~70–90%. Acetylation of **3** gave the 1,4-diacetate **4** as a crystalline mixture of anomers (93%) from which was obtained, by fractional crystallization, the pure α-anomer and sublimable, mixed crystals of the anomers (α:β = 2:1). Treatment of **4** in dichloromethane with hydrogen bromide in acetic acid generated the bromo sugar **5**, a reactive compound that was too unstable for storage and was therefore used without delay as a glycosyl donor for the subsequent operation. However, a sample of **5** was crystallized and characterized by its ^{1}H nmr spectrum; $J_{1,2}$ = 3.9 Hz indicated α-L stereochemistry. For chemical characterization, a sample was allowed to react with methanol in the presence of silver triflate, and the product was shown by its ^{1}H nmr spectrum to be an anomeric mixture of methyl glycosides, proving the bromide to act as a glycosyl donor (see Experimental).

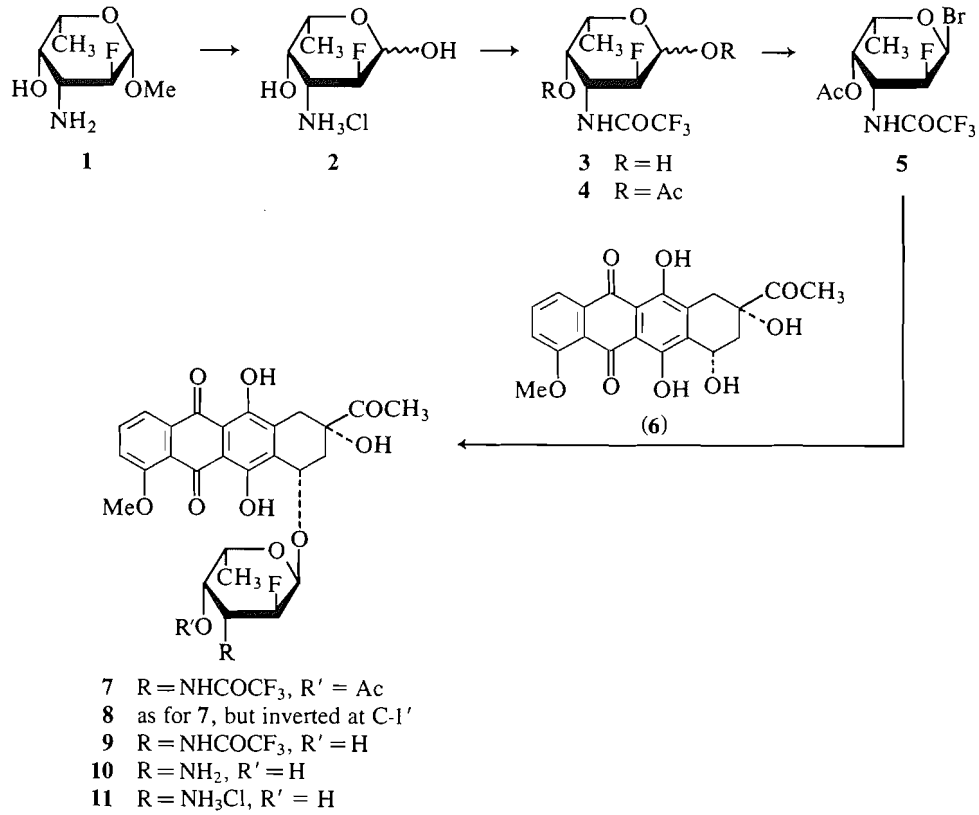
For the condensation of **5** with daunomycinone (**6**) we tried several of the procedures commonly employed in anthracycline synthesis (6), but were unable to achieve high α-selectivity. Best results were obtained when **6** and 1.2 mol equivalents of **5** were allowed to react in boiling toluene in the presence of mercuric cyanide and mercuric bromide; chromatographic isolation then gave the desired α-glycoside **7** in 41%, and its β-anomer **8** in 29% yield (based on **6**). Treatment of **5** with 1.75 mol equiv. of **6** in 1:1 dichloromethane–nitromethane solution in the presence of silver triflate proved less satisfactory; **7** and **8** could be isolated in yields (based on **5**) of 15 and 16.5% only.

Zemplén *O*-deacetylation of **7** gave crystalline (*S*)-2′-fluoro-*N*-trifluoroacetyldaunorubicin (**9**), which was then *N*-deacylated with aqueous barium hydroxide at room temperature to furnish the target compound as the free base **10**, obtained in the form of dark-red microscopic needles in 35% overall yield based on the amount of aglycon used. The hydrochloride **11** crystallized as a (nonhygroscopic) sesquihydrate showing strong dextrorotation, $[\alpha]_D$ +218° (in methanol). The product obtained by Castillon *et al.* (5), named 2′-*C*-fluoro-β-daunomycin, was isolated as a highly hygroscopic hydrochloride monohydrate that was accorded formula **11** but showed $[\alpha]_D$ −37° (in methanol).[1]

Biological activity

Tests to evaluate antitumor activity of **11** in comparison with parent daunorubicin were performed by Bristol–Myers

[1]The authors (5) prepared their compound by silver triflate-promoted condensation of aglycon **6** with the 4-*O*-benzoyl analog of **5** (compound **17** in ref. 5, erroneously depicted as the β-anomer), followed by sequential deacylation at positions 4′ and 3′. The reported (5) ^{1}H nmr data are in fair agreement with our values for **11**, but the reason for the discrepancy in optical rotation is unclear. The molecular rotation of our product **11**, $[M]_D$ +133 000°, is close to that of parent daunorubicin hydrochloride ($[M]_D$ +143 000°, from $[\alpha]_D$ +253° (7)) and to that of adriamycin hydrochloride ($[M]_D$ +144 000°, from $[\alpha]_D$ +248° (8)). In this series of aminoglycoside hydrochlorides, β-L anomers exhibit *higher* dextrorotation than their α-L counterparts. Although values for the β-anomers (9) of the daunorubicin and adriamycin hydrochlorides have apparently not been published, for the latter compound a value of $[\alpha]_D$ +472° (*c* 0.07, methanol) was communicated to us privately by Dr. S. Penco (Farmitalia Carlo Erba, Milan). Similarly, 4′-epi-daunorubicin hydrochloride and its β-anomer show $[\alpha]_D$ +314° and +357°, respectively (10), and their 6′-hydroxy analogs show $[\alpha]_D$ +388° and +412°, respectively (11).



1 **2** **3** R = H, **4** R = Ac **5** **6**

7 R = $NHCOCF_3$, R′ = Ac
8 as for **7**, but inverted at C-1′
9 R = $NHCOCF_3$, R′ = H
10 R = NH_2, R′ = H
11 R = NH_3Cl, R′ = H

Company, Wallingford, Connecticut. Although in vitro cytotoxicity tests with mouse melanoma B16-F10 and human colon tumor HCT-116, respectively, indicated a 5- to 8-fold lower potency for **11** than for daunorubicin (IC_{50} = 4.92 and 3.87 μg/mL versus 0.64 and 0.72 μg/mL), the two antibiotics showed comparable potency (IC_{50} = 17.30 vs. 21.40, 10.06 vs. 16.95, and 8.48 vs. 8.00 μg/mL) against the cell lines HCT/VP35, Moser, and SW900 (average values from four test series). Tests in vivo against L1210 murine leukemia in the dose range 0.8–16 mg/kg showed median survival time increases (T/C values) of 117–133% for **11**, whereas daunorubicin gave values of 133–167% in parallel tests with the dose range of 1–12 mg/kg. In subsequent tests compound **11** was dosed as high as 64 mg/kg; at that dose it achieved a maximum T/C of 129%. No toxicity was observed in **11** at the dose levels tested, which considerably exceeded the toxic limit for daunorubicin. Even though antitumor activity was but modest, this point encourages further study and we are currently engaged in the synthesis of similar fluoroanthracyclines for biological evaluation.

Experimental

General preparative and instrumental methods were the same as those previously employed (1). The following solvent combinations (v/v) were used for thin-layer chromatography (tlc) and column chromatography (medium pressure) on silica gel: methanol–chloroform, 1:3 (A) and 1:6 (B); methanol–acetone, 1:1 (C); acetone–dichloromethane, 1:4 (D); and ethyl acetate – hexanes in the proportions 3:1 (E), 3:2 (F), 1:1 (G), 1:2 (H), 1:3 (I), and 1:5 (J). Proton magnetic resonance spectra were taken at 300 MHz, with tetramethylsilane as internal standard. Infrared spectra were taken from Nujol mulls. Optical rotations were measured at ~25°C.

3-Amino-2,3,6-trideoxy-2-fluoro-L-galactose ((S)-2-fluoro-L-daunosamine) hydrochloride (2)

A solution of compound **1** (1) (1.08 g) in 4 M hydrochloric acid (108 mL) was boiled under reflux for 6 h. Complete conversion into the more slowly moving sugar **2** was indicated by tlc (solvent A). The acid was removed by coevaporation with added water, and the residual aqueous solution was decolorized with charcoal and brought to dryness at 35°C, to give **2** as a crystalline solid (1.10 g after drying *in vacuo* over KOH; 90%); mp ~190°C (dec.); $[\alpha]_D$ −70° (*c* 0.9, H_2O) (lit. (3) $[\alpha]_D$ −69.7°); ν_{max}: 3400 (br), 2700–2500 (several weak bands), 1600, 1580, 1530 (strong), 1300–990 (numerous sharp bands), 890, and 740 cm^{-1}. The ^1H nmr revealed the product to be the pure *β-anomer*; δ(DMSO-d_6): 8.45 (bs, 3H, exchangeable, H_3N^+), 7.18 and 5.74 (d, J = 6.7 and 5.9 Hz, exchangeable, 2 OH), 4.67 (td, collapsing to dd on D_2O exchange, $J_{1,F}$ = 3, $J_{1,2} \approx J_{1,OH} \approx$ 7.3 Hz, H-1), 4.40 and 4.23 (2 dd, 0.5H each, $J_{2,F}$ = 51.7, $J_{1,2}$ = 7.8, $J_{2,3}$ = 10.2 Hz, H-2), 3.75 (nm, H-4), 3.72 (q, $J_{5,Me}$ = 6.25, $J_{4,5}$ < 0.5 Hz, H-5), 3.52 (~td, $J_{3,4}$ = 2.4, $J_{2,3}$ = 10, $J_{3,F}$ = 12 Hz, H-3), and 1.12 (d, 3H, J = 6.25 Hz, *C*-Me).

When **2** was recovered from its aqueous solution by *freeze-drying*, it was obtained as a crystalline, 1:1 mixture of anomers; mp 182–185°C (dec.); $[\alpha]_D$ −85° (*c* 1.1, H_2O). The ir spectrum, similar to that of the pure β-anomer, was distinguished by additional, prominent peaks at 1495, 860, 807, and 755 cm^{-1}. The ^1H nmr spectrum, in addition to showing all the β-anomer signals listed above, contained the following set of signals, of equal strength, for the *α-anomer*; δ(DMSO-d_6): 7.00 and 5.69 (d, J = 3.5 and 6 Hz, exchangeable, 2 OH), 5.22 (~t, collapsing to ~d on D_2O exchange, $J_{1,2} \approx J_{1,OH} \approx$ 3.5, $J_{1,F} \approx$ 0.5 Hz, H-1), 4.76 and 4.59 (2 dd, 0.5H each, $J_{2,F}$ = 51, $J_{1,2}$ = 3.5, $J_{2,3}$ = 10.5 Hz, H-2), 4.05 (q, $J_{5,Me}$ = 6.5, $J_{4,5}$ < 0.5 Hz, H-5), 3.82 (nm, H-4), 3.58 (td, similar to, and partially overlapped by, the corresponding signal of the anomer, H-3), and 1.06 (d, 3H, J = 6.5 Hz, *C*-Me). *Anal.* calcd. for $C_6H_{13}ClFNO_3$ (201.5): C 35.76, H 6.50, Cl 17.59; found: C 35.66, H 6.50, Cl 17.71.

2,3,6-Trideoxy-2-fluoro-3-trifluoroacetamido-D-galactose (3)

A suspension of **2** (1.00 g) in dry ether (70 mL) containing triethylamine (0.7 mL) was stirred for 0.5 h, and for a further 2.5 h following the addition of trifluoroacetic anhydride (2.1 mL, 3 mol equiv.). The solution then showed a double spot, R_f ~ 0.8 (tlc, solvent D), for the α,β-ditrifluoroacetate of **3**; a small proportion of suspended

solid gave a double spot at $R_f \sim 0.25$ and proved to be α,β-**3**. (Starting **2** had R_f 0). The mixture was evaporated, with added portions of ether, to give a yellow residue that was dissolved in dry methanol (50 mL). After 2 days at room temperature, all of the diester had been methanolized to α,β-**3** (tlc). The solvent was removed and a solution of the residue in EtOAc was washed once with a small amount of ice water, dried (Na_2SO_4), and evaporated. Trituration of the material with ether gave crystalline **3** (0.84 g), mp 162–165°C, and chromatographic processing of the mother liquor augmented the yield to 1.015 g (68%); $[\alpha]_D$ −129° (*c* 0.5, acetone). In one experiment (50-mg scale), direct crystallization gave a 93% yield of **3**, mp 153–156°C; ν_{max}: 3540, 3400, 1700, 1550 cm^{-1}. Different preparations showed variable anomeric composition, according to ^1H nmr, e.g., α:β ratio ~ 10:1 and 3:2. (The latter sample was a crystalline solvate containing ½ mol of diethyl ether, not removed by drying *in vacuo*; the former sample was solvent free.) The ^1H nmr data (DMSO-d_6; low-field OH and NH resonances removed by D_2O exchange) for the *α-anomer*, δ: 5.17 (~d, $J_{1,2} = 4$, $J_{1,F} \approx 0.5$ Hz, H-1), 4.81 and 4.64 (2 dd, 0.5H each, $J_{1,2} = 4$, $J_{2,3} = 10.6$, $J_{2,F} = 50$ Hz, H-2), 4.30 (m, H-3), 4.06 (qd, $J_{4,5} = 1$, $J_{5,Me} = 7$ Hz, H-5), 3.60 (~nt, H-4), and 1.04 (d, 3H, $J = 7$ Hz, *C*-Me); for the *β-anomer*, δ: 4.64 (m, H-1), 4.50 and 4.33 (2 dd, 0.5H each, $J_{1,2} = 8$, $J_{2,3} = 10$, $J_{2,F} = 51$ Hz, H-2), 4.3 (m, H-3), 3.74 (qd, $J_{4,5} = 1$, $J_{5,Me} = 6.5$ Hz, H-5), 3.56 (~nt, H-4), and 1.09 (d, $J = 6.5$ Hz, *C*-Me). The hemietherate showed a quartet (2H, $J = 6.5$ Hz) at δ 3.37 and a matching triplet that was superposed on the sugar *C*-Me signals for a total intensity of 6H. *Anal.* calcd. for $C_8H_{11}F_4NO_4 \cdot 0.5\ C_4H_{10}O$ (298.2): C 40.27, H 5.41; found: C 40.47, H 5.64.

1,2-Di-O-acetyl-2,3,6-trideoxy-2-fluoro-3-trifluoroacetamido-L-galactose (4)

Compound **3** (1.01 g) was acetylated with acetic anhydride and pyridine (23°C, 1 h), and the crude diacetate **4** (R_f 0.4, tlc with solvent H) was isolated by exhaustive evaporation of added toluene from the mixture. Crystallization from ethyl acetate – hexane gave two crops (670 and 90 mg, mp 165–168 and 161–164°C) of **4**, with the first one representing the pure *α-anomer*, $[\alpha]_D$ −178° (*c* 0.4, $CHCl_3$); ν_{max}: 3300, 1750 (doublet), 1700, 1540 cm^{-1}; δ ($CDCl_3$): 6.41 (d, $J_{1,2} = 3.4$ Hz, H-1), 5.36 (~td, $J_{4,5} = 1.5$, $J_{3,4} \approx J_{4,F} \approx 3.5$ Hz, H-4), 4.86 and 4.69 (2 dd, 0.5H each, $J_{1,2} = 3.4$, $J_{2,3} = 11$, $J_{2,F} = 52$ Hz, H-2), 4.76 (m, H-3), 4.23 (qd, $J_{4,5} = 1.4$, $J_{5,Me} = 6.5$ Hz, H-5), 2.19 and 2.17 (s, 3H each, 2 OAc), and 1.11 (d, 3H, $J = 7$ Hz, *C*-Me).

The mother liquor of crystallization was purified by passage over a column of silica gel with solvent I, to give another crop of **4** (480 mg), for a total yield of 1.24 g (93%). This crop gave a double spot in tlc ($R_f \sim 0.6$, solvent H) and was shown to be an anomer mixture (α:β = 2:1, by ^1H nmr). On heating at 110°C in an oil-pump vacuum for 16 h it sublimed almost without residue, and the sublimate showed unchanged spectral and tlc patterns; mp 130–134°C, $[\alpha]_D$ −138° (*c* 0.6, $CHCl_3$); ^1H nmr ($CDCl_3$) signals attributable to the *β-anomer*, δ: 5.79 (dd, $J_{1,F} = 3.5$, $J_{1,2} = 7.5$ Hz, H-1), 5.30 (td, $J_{4,5} = 1.7$, $J_{3,4} \approx J_{4,F} = 3.3$ Hz, H-4), 4.59 and 4.41 (2 dd, 0.5H each, $J_{1,2} = 7.5$, $J_{2,3} = 10.7$, $J_{2,F} = 54$ Hz, H-2), 4.48 (m, H-3), 3.99 (qd, $J_{4,5} = 1.7$, $J_{5,Me} = 6.5$ Hz, H-5), 2.18 and 2.17 (2 s, 3H each, 2 OAc), and 1.18 (d, 3H, $J = 6.5$ Hz, *C*-Me). *Anal.* calcd. for $C_{12}H_{15}F_4NO_6$ (345.3): C 41.74, H 4.38, N 4.06; found: C 41.33, H 4.54, N 3.98.

4-O-Acetyl-2,3,6-trideoxy-3-trifluoroacetamido-α-L-galactopyranosyl bromide (5)

To a solution of **4** (780 mg) in CH_2Cl_2 (20 mL, distilled from CaH_2) was added 30% hydrogen bromide in acetic acid (17 mL), at room temperature. After 30 min all of the **4** was consumed (tlc), and the mixture was processed by evaporation of the CH_2Cl_2, removal of the acid by coevaporation with added toluene, decolorization of the product with activated carbon, and brief drying *in vacuo* of the syrupy, almost colorless **5** so obtained. The crude **5** was homogeneous in tlc (R_f 0.5, solvent H) except for the presence of trace impurities, and, being rather unstable, it was used without delay for the intended condensation reaction.

For characterization, a sample of the syrup was crystallized from ether–hexane (0°C, overnight), whereby the trace impurities were removed and no change in the R_f value was observed. The crystals were immediately taken to ^1H nmr analysis; δ ($CDCl_3$): 6.59 (d, $J_{1,2} = 3.9$ Hz, H-1), 6.3 (br d, NH), 5.39 (td, $J_{4,5} = 1.25$, $J_{3,4} = J_{F,4} = 3.3$ Hz, H-4), 4.82 (symm. m, 10 lines, width 32 Hz, $J_{3,4} \approx J_{3,NH} \approx 4$, $J_{2,3} \approx 11$, $J_{3,F} \approx 13$ Hz, H-3), 4.73 and 4.57 (2 dd, 0.5H each, $J_{1,2} = 3.9$, $J_{2,3} = 10.8$, $J_{2,F} = 49.9$ Hz, H-2), 4.40 (qdd, $J_{5,Me} = 6.6$ and $J_{4,5} = 1.2$ Hz, with additional splitting due to long-range coupling, H-5), 2.17 (s, 3H, OAc), and 1.17 (d, 3H, $J = 6.6$ Hz, *C*-Me).

A 10-mg sample of **5** was allowed to react with methanol (0.5 mL) in the presence of a few milligrams of silver triflate. After 1 h at 25°C, **5** (R_f 0.5) had been converted into a new product, R_f 0.4 (tlc with solvent H). The mixture was filtered through Celite, which was washed with CH_2Cl_2, and the filtrate was washed once with aqueous $NaHCO_3$ and twice with water, dried (Na_2SO_4), and evaporated. The foamy residue of *methyl glycoside* was passed through a small column of silica gel with solvent J but still failed to crystallize. The eluate contained mainly the β-anomer; δ ($CDCl_3$): 6.3 (br m, NH), 5.29 (nm, H-4), 4.52–4.36 (m, 2.5H, H-1,3, and downfield part of H-2), 4.25 (dd, 0.5H, $J_{1,2} = 7.3$, $J_{2,3} = 10.5$ Hz, H-2, upfield part), 3.85 (qd, $J_{4,5} = 1$, $J_{5,Me} = 6.5$ Hz, H-5), 3.59 (s, 3H, OMe), 2.16 (s, 3H, OAc), and 1.19 (d, 3H, $J = 6.5$ Hz, C-Me). A small *C*-Me doublet at δ 1.11 indicated the presence of some α-anomer (< 10%).

7-O-(4-O-Acetyl-2,3,6-trideoxy-2-fluoro-3-trifluoroacetamido-β-L-galactopyranosyl)-daunomycinone (7) and its β-anomer 8

Procedure A

The syrupy glycosyl bromide (**5**) obtained from 100 mg (0.29 mmol) of diacetate **4** was dissolved in dry toluene (10 mL, distilled from Na), and $HgBr_2$ (200 mg, 2 mol equiv.), $Hg(CN)_2$ (145 mg, 2 mol equiv.), and daunomycinone (**6**; 100 mg, 0.25 mmol) were added. The mixture was boiled under reflux for 5 h, with exclusion of moisture and light. Thin-layer chromatography (solvent F) indicated complete consumption of **5** (R_f 0.9) but incomplete consumption of **6** (R_f 0.2); two ill-separated spots (R_f 0.4–0.5) representing **7** and **8** were seen. The filtered solution was evaporated and the resultant residue dissolved in $CHCl_3$. The solution was washed with water (3×), dried (Na_2SO_4), and evaporated to give a red solid that still contained some inorganic material and was, therefore, triturated with $CHCl_3$ (2 mL). The filtered solution gave 180 mg of product, which was subjected to preparative tlc (triple irrigation with solvent G), furnishing slower moving **7** (71 mg, 41.4%) and faster moving **8** (49 mg, 28.5%).

Compound **7** showed mp 194–196°C (with gradual darkening from 160°C); $[\alpha]_D$ +149°C (*c* 0.6, $CHCl_3$); ν_{max}: 3500, 3300, 1745, 1700, 1615, 1570 cm^{-1}; δ ($CDCl_3$): 8.03, 7.77, and 7.38 (d, t, and d, $J \approx 8$ Hz, H-1,2,3), 6.18 (d, $J_{3',NH} = 7.9$ Hz, NH), 5.70 (d, $J_{1',2'} = 3.9$ Hz, H-1'), 5.43 (sx, $J_{4',5'} = 1.5$, $J_{3',4'} \approx J_{4',F} \approx 3$ Hz, H-4'), 5.40 (nm, H-7), 4.73 and 4.565 (2 dd, 0.5H each, $J_{1',2'} = 4$, $J_{2',3'} = 11$, $J_{2',F} = 49.5$ Hz, H-2'), 4.50 (m, H-3'), 4.39 (qd, $J = 1.5$ and 6.2 Hz, H-5'), 4.07 (s, 3H, OMe), 4.01 (s, exchangeable, OH-9), 3.26 and 3.00 (2 d, $J_{gem} = 19$ Hz, H-10e,10a), 2.41 (s, 3H, Me-14), 2.33 and 2.22 (2 dd, $J_{gem} = 16$, $J_{vic} = 1.5$ and 4 Hz, H-8e,8a), 2.16 (s, 3H, OAc), and 1.16 (d, $J_{5',Me} = 6.2$ Hz, *C*-Me). *Anal.* calcd. for $C_{31}H_{29}F_4NO_{12}$ (683.5): C 54.47, H 4.28, F 11.12; found: C 54.38, H 4.51, F 10.91.

Compound **8** showed mp 105–110°C; $[\alpha]_D$ +116° (*c* 0.6, $CHCl_3$); ν_{max}: 3500, 3300 (strong), 3090, 1740–1700, 1615 (weak), 1560 cm^{-1}; δ ($CDCl_3$): 8.03, 7.78, and 7.39 (d, t, and d, $J \sim 8$ Hz, H-1,2,3), 6.25 (d, $J_{3',NH} = 8$ Hz, NH), 5.54 (nm, width 6 Hz, H-4'), 5.22 (nm, width 6 Hz, H-7), 5.10 (dd, $J_{1',F} = 3$, $J_{1',2'} = 7.5$ Hz, H-1'), 4.50 (m, H-3'), 4.42 and 4.25 (2 dd, 0.5H each, $J_{1',2'} = 7.8$, $J_{2',3'} = 10.5$, $J_{2',F} = 51$ Hz, H-2'), 4.08 (s, 3H, OMe), 3.83 (qd, $J_{4',5'} = 1.2$, $J_{5',Me} = 6.4$ Hz, H-5'), 3.26 and 3.06 (2 d, $J_{gem} = 19$ Hz, H-10e,10a), 2.40 (s, 3H, Me-14), 2.15–2.0 (m, 2H, H-8e,8a), 2.11 (s, 3H, OAc), and 0.99 (d, $J_{5',Me} = 6.3$ Hz, *C*-Me). *Anal.* calcd. for $C_{31}H_{29}F_4NO_{12} \cdot 3.5\ H_2O$ (746.6): C 49.87, H 4.86; found: C 49.87, 49.74; H 4.80, 4.79.

Procedure B

Daunomycinone **6** (278 mg, 0.7 mmol) in dry CH_2Cl_2 (20 mL) and nitromethane (20 mL) was stirred for 1 h with powdered molecular sieve (4 Å; 1 g). Silver triflate (180 mg, 0.8 mmol) was then added,

followed after 1 h by bromide **5** (obtained from 137 mg of diacetate **4** (0.4 mmol) and dissolved in a small volume of CH_2Cl_2). The bromide was completely consumed after 1 h (tlc with solvent F). The inorganic material was removed and washed exhaustively with CH_2Cl_2, and the filtrate was shaken with concentrated aqueous NaI solution followed by water, dried, and evaporated. The crude product mixture was chromatographed, first on a column (9 g of SiO_2) eluted with solvent I followed by solvent H, which separated **7** and **8** from most of the residual **6**, and then on preparative tlc plates (double irrigation with solvent G), which furnished pure **7** (41 mg, 15%) and **8** (45 mg, 16.5%).

*7-O-(2,3,6-Trideoxy-2-fluoro-3-trifluoroacetamido-α-L-galactopyranosyl)-daunomycinone (**9**)*

A sample of **7** (35 mg) in dry methanol (2 mL) was treated at room temperature under N_2 with methanolic 1 M sodium methoxide solution (0.5 mL). Clean conversion of **7** (R_f 0.6) into **9** (R_f 0.4) occurred within 20 min (tlc with solvent E). The dark blue to violet solution was deionized with a weak-acid ion exchange resin (Amberlite IRC-50, H^+). The orange-red solution was evaporated to give crystalline **9** (31 mg, 93%), decomposing at 160–165°C after recrystallization from ethyl acetate – hexane; $[\alpha]_D$ +177.5 ± 1.5° (*c* 0.3, $CHCl_3$); ν_{max}: 3500 (bd), 1710, 1610, 1570 cm^{-1}; δ($CDCl_3$): 8.02, 7.77, and 7.37 (d, t, and d, $J \approx 8$ Hz, H-1,2,3), 6.68 (d, $J_{3',NH} = 8.1$ Hz, NH), 5.66 (d, $J_{1',2'} = 3.8$ Hz, H-1′), 5.42 (dd, $J = 2.7$, 4.0 Hz, H-7), 4.69 and 4.53 (2 dd, 0.5H each, $J_{1',2'} = 4$, $J_{2',3'} = 10.5$, $J_{2',F} = 49$ Hz, H-2′), 4.45–4.3 (m, 2H, H-3′,5′), 4.09 (nm, H-4′), 4.06 (s, 3H, OMe), 3.92 (nm, exchangeable, OH), 3.27 and 2.99 (dd and d, $J_{gem} \approx 18$ Hz, H-10e,10a), 2.40 (s, 3H, Me-14), 2.34 and 2.21 (dt and dd, $J_{gem} = 15$, $J_{vic} = 4$ Hz, H-8e,8a; small long-range coupling present in H-8e), 2.03 (d, exchangeable, $J = 6$ Hz, OH), and 1.30 (d, 3H, $J_{5',Me} = 7$ Hz, *C*-Me). *Anal.* calcd. for $C_{29}H_{27}F_4NO_{11} \cdot 0.5\ H_2O$ (650.5): C 53.54, H 4.34, F 11.68; found: C 53.34, H 4.45, F 11.49.

*7-O-(3-Amino-2,3,6-trideoxy-2-fluoro-α-L-galactopyranosyl)-daunomycinone (**10**, (S)-2′-fluorodaunorubicin) and its hydrochloride **11***

Acetate **7** (310 mg) was *O*-deacetylated in methanol (5 mL) with $NaOCH_3$ as just described, and the crude **9** so obtained was dissolved under N_2 in a saturated, aqueous $Ba(OH)_2$ solution (10 mL) at 25°C. After 20 min, tlc (solvent C) indicated absence of **9** (R_f 1.0) and the presence of **10** (R_f 0.5), and after 30 min the blue solution was diluted with some water and neutralized by addition of solid CO_2. The red solution was filtered by suction through a layer of Celite from the precipitated $BaCO_3$, and the product was extracted from the filtrate into $CHCl_3$. The extract was washed once with water, and evaporated to give **10** as dark-red microscopic needles that were dried over KOH in a desiccator; yield, 221 mg (89.5%). The product charred at 209–210°C and melted indistinctly at 212–216°C; tlc (solvent C): R_f 0.5, with trace contaminant, R_f 0.9; ν_{max}: 3500, 3400, 3330 (3 weak bands), 1715 with shoulder at 1750, 1615, 1580, 1410, 1285, 1260–1030 (several sharp, medium intensity bands), 990, 820, 767, 745 cm^{-1}; δ($CDCl_3$): 8.02, 7.72, and 7.38 (d, t, and d, $J = 7$–8 Hz, H-1,2,3), 5.63 (d, $J_{1',2'} = 3.9$ Hz, H-1′), 5.43 (nm, H-7), 4.54 and 4.38 (2 dd, 0.5H each, $J_{1',2'} = 4$, $J_{2',3'} = 10$, $J_{2',F} = 49$ Hz, H-2′), 4.17 (~q, width 20 Hz, H-5′), 4.07 (s, 3H, OMe), 3.66 (nm, H-4′), 3.3–3.2 (m, 2H, H-3′,10e), 3.00 (d, $J_{gem} = 19$ Hz, H-10a), 2.39 (s, 3H, Me-14; partially obscuring downfield part of H-8e signal), 2.35 (dd, H-8e), 2.16 (dd, $J_{7,8a} = 4$, $J_{8a,8e} = 14$ Hz, H-8a), 1.34 (d, 3H, $J_{5',Me} = 6.2$ Hz, *C*-Me).

For preparing the *hydrochloride* **11**, a solution of **10** (210 mg) in 0.1 M HCl (15 mL) was first extracted with EtOAc for removal of the aforementioned impurity (R_f 0.9), and then evaporated to dryness with additions of ethanol and benzene. The solid **11** was crystallized from ethanol–ether and dried *in vacuo* over KOH; it was not noticeably hygroscopic, and gave a single spot (R_f 0.2) in tlc (solvent B). Yield after recrystallization, 193 mg (86%); mp 224°C (with prior darkening from 217°C); $[\alpha]_D$ +218° (*c* 0.32, CH_3OH); ν_{max}: 3600–3200 (br), 1705, 1615, 1575, 1410, 1285, 1230–1030 (several medium intensity bands), 990, and 960–720 cm^{-1} (several weak bands); δ(DMSO-d_6): 7.92 (m, 2H, H-1,3), 7.65 (t, H-2), 5.72 and 5.58 (d of 5.8 Hz, and s, exchangeable, 2 OH), 5.46 (d, $J_{1',2'} = 3.7$ Hz, H-1′), 5.08 (nm, H-7), 4.80 and 4.63 (2 dd, 0.5H each, $J_{1',2'} = 3.7$, $J_{2',3'} = 10.5$, $J_{2',F} = 49.5$ Hz, H-2′), 4.33 (~q, width 20 Hz, H-5′), 3.98 (s, 3H, OMe), 3.81 (m, narrowed after D_2O exchange, H-4′), 3.40 (m, H-3′), 2.97 (s, 2H, H-10a,e), 2.27 (s, 3H, Me-14), 2.22 (dd, H-8e), 2.11 (dd, $J_{7,8a} \approx 4$, $J_{8a,8e} = 14$ Hz, H-8a), and 1.14 (d, 3H, $J_{5',Me} = 6.6$ Hz, *C*-Me). *Anal.* calcd. for $C_{27}H_{29}ClFNO_{10} \cdot 1.5\ H_2O$ (609.0): C 53.25, H 5.13, Cl 5.82, F 3.12, N 2.30; found: C 53.00, H 5.17, Cl 5.67, F 3.45, N 2.29.

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