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Note

An improved synthesis of an important *S*-sialylnucleoside analogue

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The biochemistry and synthesis of sialic acid analogues, in particular influenza virus sialidase inhibitors, has been of great interest to us [1–4]. Sialic acids are considered to be important in the propagation of a number of metastatic cancers such as colon adenocarcinoma [5]. Their importance appears to be linked to the conclusion that a correlation exists between increased cell-surface sialic acid content and cancer, particularly metastatic potential [6–8]. A number of *O*- and *S*-sialylnucleosides for example, 5-fluoro-2′,3′-*O*-isopropylidene-5′-*O*-(methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-5′-uridine (**1**) and 5′-*S*-(5-acetamido-2,3,5-trideoxy-D-*glycero*-α- and -β-D-*galacto*-2-nonulopyranosylonic acid)-5′-thiocytidine (**2**) appear to be candidates as inhibitors of cell-surface sialylation and subsequent pulmonary metastasis of mouse colon adenocarcinoma by either direct or indirect inhibition of sialyltransferase activity [9,10].

Hasegawa and co-workers have reported the synthesis of *S*-sialylnucleosides via a low-temperature, selective *S*-deacetylation of methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-2-*S*-acetyl-2,3,5-trideoxy-D-*glycero*-β-D-*galacto*-2-nonulopyranosonate and further reaction with 5′-activated nucleosides [11]. Our recent interest in the synthesis of other sulfur-containing sialic acids [4,12], in particular, *S*-sialyldisaccharides [12], has provided a significantly better methodology for the preparation of a suitably protected precursor of **2** (Scheme 1).

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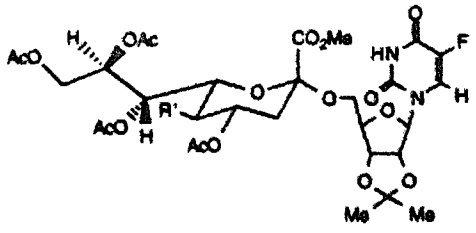
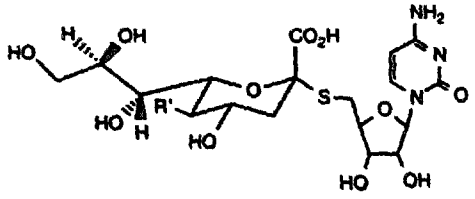


1 R' = NHAc



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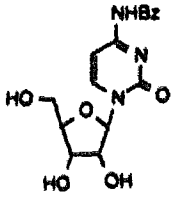
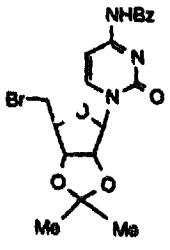


3

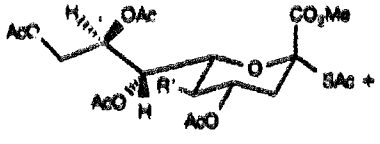
1) TPP/Br_2/THF
- 10° C → 65° C, 18 h

2)  / H^+/ THF

(87% isolated yield from 3)



4

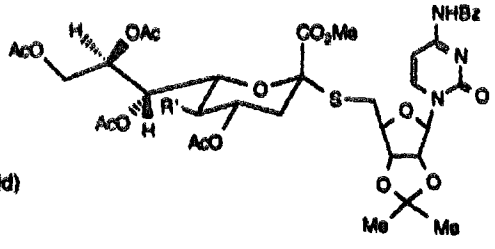


5 R' = NHAc

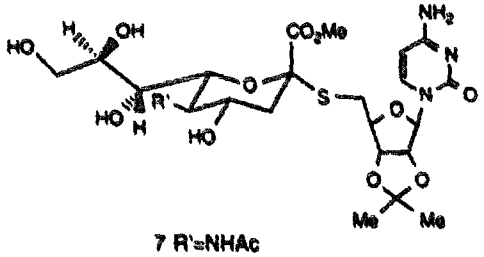
Et_3NH/Me_2NCHO

4

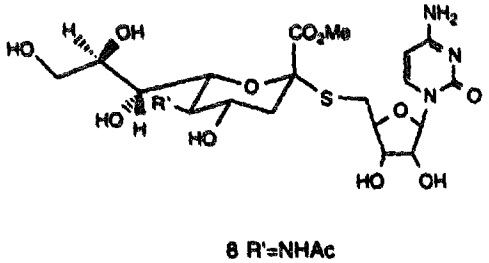
18 h, RT
(71% isolated yield)



6 R' = NHAc



7 R'=NHAc



8 R'=NHAc

Scheme 1.

We required a straightforward synthesis of the nucleosid-5'-yl moiety of **6** suitably activated for coupling to a sialosyl donor derived from **5**. Thus, we have prepared **4** by treatment of **3** [13] with triphenylphosphine:bromine (3:7) in tetrahydrofuran, followed by isopropylidenation. This method was found to be superior in both yield and ease of preparation when compared with previously reported syntheses of similar N^4-benzoyl-5'-bromo-5'-deoxycytidine analogues [11] and other 5'-bromo-5'-deoxynucleosides [14].

In one pot, the selective *S*-deacetylation of the known compound **5** [15] and subsequent coupling reaction proceeded very cleanly by treatment of **4** and **5** with diethylamine in Me_2NCHO to yield, after workup and chromatography, **6** in 71% yield. Deisopropylidenation and deesterification of **6** provided **2** in good yield (see Experimental). This method provides a simple entry into a very important class of compounds.

4. Experimental

Preparation of N^4*-benzoyl-5'-bromo-5'-deoxy-2',3'-*O*-isopropylidenecytidine* (**4**).— To a cooled (−10°C) THF solution (46 mL) of the reactive brominating agent dibromotriphenylphosphorane [16] (2.09 g, 4.95 mmol) was added **3** (1.15 g, 3.3 mmol). The mixture was warmed to room temperature and subsequently heated (65°C) for 18 h. Upon quenching with MeOH (5 mL) the mixture was concentrated under reduced pressure to a syrup, which was then taken up in THF (65 mL) and treated with 2,2-dimethoxypropane (13 mL) in the presence of a catalytic amount of Amberlyst-15 (H^+) resin for 18 h to yield **4** after the usual workup and column chromatography (1:1 EtOAc–hexane as eluting solvent) in 87% yield. Data for **4**: $[\alpha]_D^{21}$ + 15.5° (*c* 4, $CHCl_3$); ^{1}H NMR (Me_2SO-d_6, 300 MHz): δ 8.24, 7.35 (2 d, 2 H, $J_{6,5}$ 7.53 Hz, H-6, H-5), 8.01, 7.61, 7.51 (d, 2 H, t, 3 H, Ph), 5.88 (d, 1 H, $J_{1',2'}$ 1.35 Hz, H-1'), 5.18 (dd, 1 H, $J_{2',3'}$ 6.36 Hz, H-2'), 4.87 (dd, 1 H, $J_{3',4'}$ 3.42 Hz, H-3'), 4.34 (m, 1 H, $J_{4',5'a} = J_{4',5'b} \approx 6.7$ Hz, H-4), 3.82 (dd, 1 H, $J_{5'a,5'b}$ 10.12 Hz, H-5'a), 3.70 (dd, 1 H, H-5'b), 1.49, 1.30 (2 s, 6 H, Me); ^{13}C NMR (Me_2SO-d_6, 75.5 MHz): δ 167.5 (C-2), 163.60 (C=O Ph), 153.95 (C-6), 148.15, 132.99, 132.75, 128.92, 128.37 (Ph), 112.96 (C-8), 96.43 (C-5), 95.60 (C-1'), 87.44 (C-4'), 84.18 (C-2'), 83.04 (C-3'), 35.13 (C-5'), 26.77 (C-9), 24.98 (C-10); FABMS: *m/z* 449.9 (M^+), 369 (M − Br). IR (KBr) 3450 (NH), 1710 and 1230 (ester), 1530 and 1630 (amide). Anal. Calcd for $C_{19}H_{20}BrN_3O_5$: C, 50.7; H 4.5%. Found: C, 51.0; H 4.7%.

N^4*-Benzoyl-2',3'-*O*-isopropylidene-5'-*S*-(methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-2,3,5-trideoxy-*D*-glycero-α- and -β-*D*-galacto-2-nonulopyranosylonate)-5'-thiocytidine* (**6**).—To a stirred solution of **5** (47 mg, 0.086 mmol) and **4** (35 mg, 0.08 mmol) in Me_2NCHO (0.36 mL) was added diethylamine (0.18 mL) at room temperature. The mixture was let stir for 18 h, and the progress of the reaction was followed by TLC (EtOAc). After concentration under reduced pressure, the crude product was taken up in CH_2Cl_2 and washed with water, and the organic extracts were combined, dried over sodium sulfate, and finally concentrated to an oil. Upon chromatography (EtOAc), **6** was isolated in 71% yield. Data for **6**: $[\alpha]_D^{21}$ + 59.8° (*c* 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$)-Neu5Ac unit: δ 5.61 (d, 1 H, $J_{NH,5}$ 9.8 Hz, NH), 5.36 (m, 1 H, H-8), 5.34 (dd, 1 H, $J_{7,6}$ 8.8 Hz, H-7), 4.26 (dd, 1 H, $J_{9',8}$ 2.4, $J_{9',9}$ 12.5 Hz, H-9'), 4.09 (dd, 1 H, $J_{9,8}$ 4.9 Hz, H-9), 3.84

(dd, 1 H, $J_{6,5}$ 10.7 Hz, H-6), 3.76 (s, 3 H, OMe), 2.73 (dd, 1 H, $J_{3e,3a}$ 12.7, $J_{3e,4}$ 4.6 Hz, H-3e), 1.87 (s, 3 H, NAc); cytidine unit: δ 8.12 (d, 1 H, $J_{6,5}$ 7.2 Hz, H-6), 7.6 (d, 1 H, H-5), 7.47–7.94 (m, 5 H, Ph), 5.86 (d, 1 H, $J_{1',2'}$ 2 Hz, H-1'), 5.44 (br t, 1 H, $J_{2',3'}$ 5.9 Hz, H-2'), 5.26 (dd, 1 H, $J_{3',4'}$ 4.4 Hz, H-3'), 4.33 (br t, 1 H, H-4'), 3.16 (dd, 1 H, $J_{5'b,4'}$ 6.5, $J_{5'b,5'a}$ 13.7 Hz, H-5'b), 2.93 (dd, 1 H, $J_{5'a,4'}$ 5.6 Hz, H-5'a); other groups 2.14, 2.12, 2.00 (4 s, 12 H, 4 OAc), 1.56 and 1.34 (isopropylidene). FABMS: m/z 877 $(M + 1)^+$. Anal Calcd for $C_{39}H_{48}N_4O_{17}S$: C, 53.4; H, 5.5%. Found: C, 53.5; H 5.8%.

Compound **6** was deprotected to give **7** using Hasegawa's method [11] in 95% yield.

Deisopropylidenation of **7** was carried out using a literature method [17] to give **8**. Thus, compound **7** (125 mg, 0.2 mmol) was dissolved in water (1 mL), treated with Dowex-50W (H^+) (5 mg), and the mixture was heated to 60°C for 4 h. Quantitative conversion was observed by TLC, and the product **8** was isolated in 96% yield. Deesterification was achieved using standard conditions [11] to give compound **2** in 90% yield. The product was shown by spectroscopy to be identical to that previously reported [11].

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