Some 6-Substituted-9-(β-L-fucopyranosyl)purines (1)

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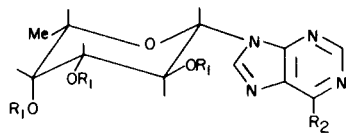
L-Fucose, an important constituent of many glycoproteins and oligosaccharides (2), was reported (3) to dramatically alter the morphology and metabolism of certain cells, particularly 3T3 mouse fibroblasts. These effects were not observed with other simple, related sugars (3). These effects suggested that some naturally occurring sugar constituents of cells such as L-fucose may play a unique role in influencing cell growth and metabolism (4). The above observations, coupled with our interest in nucleosides containing fraudulent sugars, suggested that some L-fucose nucleosides be synthesized for biological evaluation. The synthesis, and some biological and nmr results are reported here.



2, R_1 = Ac, R_2 = $NHCOC_6H_5$
3, R_1 = Ac, R_2 = OH
4, R_1 = Ac, R_2 = SH
5, R_1 = H, R_2 = NH_2
6, R_1 = H, R_2 = OH
7, R_1 = H, R_2 = SH

When either crystalline α-L-fucose tetraacetate (5) or the noncrystalline mixture of α- and β-anomers (5) was treated with hydrogen bromide in acetic acid, a syrupy bromo sugar was obtained that appeared to be α-acetobromo-L-fucose (**1**) with no other anomer detectable by nmr. These results are like those for L-acetochlorofucose, where the crystalline α-anomer has been prepared recently (6). Reaction of **1** with chloromercuri-6-benzamidopurine afforded the blocked nucleoside **2** in good yield. Deacylation with an equivalent of sodium methoxide in methanol readily afforded the adenine nucleoside **5**. Treatment with sodium nitrite in aqueous acetic acid gave the crude hypoxanthine nucleoside **6** (7) which was directly acetylated to crystalline **3**. Phosphorus pentasulfide in hot pyridine converted **3** to **4**, which on deacetylation gave the mercaptopurine fucoside **7**. Deacylation of crystalline **3** afforded the hypoxanthine fucoside **6**.

The nucleosides **2-7** were screened for antitumor activity in the mouse leukemia L 1210 system by Chemotherapy, National Cancer Institute, according to its protpcol (8). These compounds were all inactive at a dose of 400 mg./kg./day.

Some nmr date obtained at 60 MHz are given in Table I. If the bromo sugar **1** is, like the corresponding chloro sugar (6), assumed to be in the 1C conformation (9), then the coupling constant for the H-1 and H-2 protons, $J_{1,2}$ = 3.5 Hz, suggest that these protons are axial-equatorial (11) and that **1** is the α-anomer. All the nucleosides in Table I, both acetylated and unacetylated, exhibited large coupling constants, $J_{1',2'} \sim 9$ Hz, that indicated the H-1′ and H-2′ protons to be *trans*-diaxial (11). This is possible for L-fucose only for the β-anomer in the 1C conformation. Because of overlap, not all the pyranose ring protons could be distinguished in the 60 MHz spectra.

TABLE I

60 MHz Nmr Data for L-Fucopyranose Derivatives (a)

	H-1′		H-5′	H-6′		
Compound	δ	$J_{1',2'}$	δ	δ	$J_{5',6'}$	Purine
1	6.72	3.5	4.44	1.26	6.5	—
3	5.93	9	4.26	1.29	6.5	8.18, 8.52
5	5.49	9	(b)	1.21	6.5	8.27, 8.33
6	5.42	9.5	(b)	1.17	6.5	8.14, 8.27
7	5.42	9	(b)	1.15	6	8.32, 8.47

(a) These data were obtained on a Varian A-60 using deuteriochloroform as solvent with TMS internal reference for the acetylated compounds (**1** and **3**) and DMSO-d_6 with TMS external reference for the others. All signal intensities indicated the proper number of protons. The H-1′ and H-6′ signals were doublets; H-5′, quartet; and purine H-2 and H-8, singlets. The coupling constants are given to the nearest 0.5 Hz. (b) Among overlapping proton signals in the δ 3.4-4.7 region.

Two spectra were obtained at 100 MHz. The pyranose protons were all clearly resolved and confirmed the 1C conformation for the L-fucose nucleosides. See Table II. For both **2** and **4**, the large coupling constants, $J_{1',2'}$ and $J_{2',3'}$, showed that H-1′, H-2′ and H-3′ were all in the *trans*-diaxial relationship (11) expected for the 1C conformation. For **4**, the H-1′ doublet was at lowest field and is followed by H-2′, which appeared as a triplet ($J_{1',2'}$ and

TABLE II

100 MHz Data (a)

Proton		Compound 2	Compound 4
H-1′	δ	5.93 d	6.04 d
	$J_{1',2'}$	9	9
H-2′	δ	5.65 t	5.66 t
	$J_{2',3'}$	9.5	9
H-3′	δ	5.31 q	5.47 q
	$J_{3',4'}$	3	3.5
H-4′	δ	5.43 q	5.27 q
	$J_{4',5'}$	1	1
H-5′	δ	4.17 o	4.46 o
	$J_{5',6'}$	6	6.5
H-6′		1.26 d	1.13 d
Purine		8.26, 8.80	8.25, 8.42
Acetyls		1.73, 1.99, 2.25	1.78, 1.97, 2.24

(a) These were obtained on a Varian HA-100 using deuteriochloroform as solvent for **2** and DMSO-d_6 for **4** which was much less soluble. All signal intensities indicated the proper number of protons. The coupling constants are given to the nearest 0.5 Hz. Multiplicity is denoted by d = doublet, t = triplet, q = quartet and o = octet.

$J_{2',3'}$ were equal) and the H-3′ quartet. Because $J_{4',5'}$ was very small, H-4′ appeared as a pair of doublets and H-5′ as a quartet of doublets with the narrow doublets in both cases being almost singlets. The H-6′ protons appeared furthest upfield as a doublet with the coupling constant $J_{5',6'}$ being quite large. For all the L-fucose compounds examined (**1** to **7**), $J_{5',6'}$ was quite constant.

The protons of the acetoxy groups of **4** appeared as three separate singlets. The one furthest downfield is attributed to the axial acetoxy at C-4′ (11). The two equatorial acetoxy signals were upfield, but did not coincide because the C-2′ acetoxy signal is shifted further upfield by the ring current shielding effect of the equatorial purine at C-1′ (12). The results of **2** are like those for **4** except that the relative positions of the H-3′ and H-4′ protons are interchanged.

The results in Tables I and II show that all the L-fucose nucleosides examined have the 1C or *N* conformation (10) as shown in the formulas.

EXPERIMENTAL (13)

9-(2,3,4-Tri-*O*-acetyl-β-L-fucopyranosyl)-6-benzamidopurine (**2**).

A solution of 1.00 g. (3.01 mmoles) of 1,2,3,4-tetra-*O*-acetyl-α-L-fucose (**5**) in 2 ml. of 1,2-dichloroethane was treated with 3.4 ml. of 30-32% hydrogen bromide in acetic acid, allowed to stand at room temperature for 2.5 hours and worked up to give 1.00 g. (94%) of acetobromo-L-fucose (**1**) as a syrup; R_f 0.55 in solvent B, with a trace of a slower moving material. The nmr taken immediately and after 3 days storage at 4° was unchanged, as were the other physical properties.

A suspension of 2.84 g. (6.00 mmoles) of chloromercuri-6-benzamidopurine (from 4.44 g. of a 36% Celite mixture) in 60 ml. of xylene was treated with 2.0 g. (5.66 mmoles) of **1** for 2.5 hours at reflux temperature and worked up in the usual manner (14) to afford 2.19 g. (76%) of **2** as a tan foam. This was suitable for use in the next step. The product from another run was crystallized from methanol-water to give **2** (58% yield) as a white solid, m.p. 120-125°; $[\alpha]_D^{22}$ + 17.8° (c 1.00, 2-methoxyethanol); λ max (*p*H 1), 288 mμ (ε, 25,500), sh. 253 (11,200); λ max (*p*H 7), 279 (22,400), sh. 255 (12,700); λ max (*p*H 13), 302 (13,600); R_{Ad} 1.59 in solvent A.

Anal. Calcd. for $C_{24}H_{25}N_5O_8 \cdot H_2O$: C, 54.4; H, 5.14; N, 13.2. Found: C, 54.2; H, 5.13; N, 13.0.

9-(β-L-Fucopyranosyl)adenine (**5**).

A solution of 1.03 g. (1.95 mmoles) of crystalline **2** in 33 ml. of dry methanol and 105 mg. (1.94 mmoles) of sodium methoxide was stirred at room temperature for 18 hours. The white precipitate was collected and washed with methanol to afford 0.48 g. (85%) of **5**, m.p. 264-273° dec.; R_f 0.19 in solvent C. Recrystallization from water of similar material from another run gave the analytical sample. This was dried for 21 hours, first at 56°, then 110° at < 1 torr to give **5**, m.p. 271-275° dec.; $[\alpha]_D^{19}$ -11.8 (c 0.99, H_2O); λ max (*p*H 1), 256 mμ (ε, 14,900); λ max (*p*H 7), 258 (14,900); λ max (*p*H 13), 258 (14,900); R_{Ad} 0.94 in solvent A and R_f 0.19 in solvent C.

Anal. Calcd. for $C_{11}H_{15}N_5O_4$: C, 47.0; H, 5.38; N, 24.9. Found: C, 46.7; H, 5.43; N, 24.9.

Catalytic amounts of sodium methoxide in methanol rapidly removed the *O*-acetyl groups of **2**, but the *N*-benzoyl goupr was not completely removed after 20 hours at room temperature, according to tlc results.

9-(2,3,4-Tri-*O*-acetyl-β-L-fucopyranosyl)hypoxanthine (**3**).

A solution of 10.0 g. (35.6 mmoles) of the adenine nucleoside **5** and 10.0 g. (145 mmoles) of sodium nitrite in 200 ml. of water and 37 ml. of acetic acid was kept for 5 days at room temperature. After removal of the solvents, the residue was dissolved in 350 ml. of pyridine, treated with 22 ml. of acetic anhydride for 24 hours at room temperature, and worked up to give 11.08 (76%) of **3** as white crystals from ethanol, m.p. 285-287°; R_f 0.48 in solvent D. Recrystallization from ethanol of product from an earlier run afforded the analytical sample of **3**, m.p. 280-285°; $[\alpha]_D^{19}$ + 3.9° (c 0.59, 2-methoxyethanol); λ max (*p*H 1), 246 mμ (ε, 13,500); λ max (*p*H 7), 245 (13,400); λ max (*p*H 13), 252 (13,900); R_f 0.48 in solvent D.

Anal. Calcd. for $C_{17}H_{20}N_4O_8$: C, 50.0; H, 4.94; N, 13.7. Found: C, 50.2; H, 4.90; N, 13.7.

9-(β-L-Fucopyranosyl)hypoxanthine (**6**).

A solution of 3.0 g. (7.35 mmoles) of the triacetate (**3**) and 0.59 g. (11 mmoles) of sodium methoxide in 126 ml. of methanol was heated at reflux temperature for 4.5 hours to afford 1.53 g. (74%) of **6** as a white solid. Two recrystallizations from methanol gave the analytical sample of **6**, m.p. 190-197°; $[\alpha]_D^{17}$ - 19.9 (c 0.99, H_2O); λ max (*p*H 1), 248 mμ (ε, 11,900); λ max (*p*H 7), 248 (11,900); λ max (*p*H 13), 252 (13,000); R_f 0.1 in solvent D; R_{Ad} 0.63 in solvent A.

Anal. Calcd. for $C_{11}H_{14}N_4O_5 \cdot 0.75\ H_2O$: C, 44.7; H, 5.28; N, 18.9. Found: C, 44.9; H, 5.00; N, 19.0.

9-(2,3,4-Tri-*O*-acetyl-β-L-furcopyranosyl)purine-6-thiol (**4**).

A solution of 5.0 g. (12.2 mmoles) of **3** and 17.1 g. (76.8 mmoles) of phosphorus pentasulfide in 210 ml. of pyridine was heated at reflux temperature for 4.5 hours to afford 4.3 g. (83%) of **4** as a dark red solid. Recrystallization from ethanol gave 1.2 g. of **4** as a pink solid, m.p. 255-263° dec.; $[\alpha]_D^{17}$ + 46.7° (c 1.00, 2-methoxyethanol); λ max (*p*H 1), 321 mμ (ε, 25,100); λ max (*p*H 7), 317 (22,100); λ max (*p*H 13), 311 (23,900); R_f 0.59 in solvent D.

Anal. Calcd. for $C_{17}H_{20}N_4O_7S$: C, 48.1; H, 4.75; N, 13.2; S, 7.55. Found: C, 48.1; H, 4.80; N, 13.0; S, 7.68.

A second crop of 1.76 g. of **4**, m.p. 258-265° (total, 57%) was obtained from the mother liquors.

9-(β-L-Fucopyranosyl)purine-6-thiol (**7**).

A solution of 1.2 g. (2.82 mmoles) of **4** and 0.46 g. (8.46 mmoles) of sodium methoxide in 36 ml. of methanol was heated at reflux for 2 hours and kept overnight at room temperature to give 0.55 g. (66%) of **7**, m.p. 211-215° dec.; R_f 0.49 in solvent E. The product from another run was recrystallized from water to afford the analytical sample of **7**, m.p. 233-237° dec.; $[\alpha]_D^{19.5}$ -20.5° (c 0.93, H_2O); λ max (*p*H 1), 322 mμ (ε, 25,000), 224 (9450); λ max (*p*H 7), 318 (23,800), 226 (10,400); λ max (*p*H 13), 310 (23,200), 232 (15,200); R_f 0.50 in solvent E.

Anal. Calcd. for $C_{11}H_{14}N_4O_4S \cdot 0.75H_2O$: C, 42.4; H, 5.01; N, 18.0; S, 10.3. Found: C, 42.3; H, 4.89; N, 17.9; S, 10.1.

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(9) The conformation that is prevalent in all the common aldohexoses is the C1 form for the D-series and the 1C form for the L-series. It has been suggested (10) that it would be less confusing to designate the prevalent conformation as the *normal* conformation (designated by *N*), and the other as the *alternate* (*A*).

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