

Reaction of 1-(2',3'-Epoxy- $\beta$ -D-lyxofuranosyl)uracil  
with Hydrogen Fluoride. The Unexpected Formation of  
1-(3'-Fluoro-3'-deoxy- $\beta$ -D-ribofuranosyl)uracil

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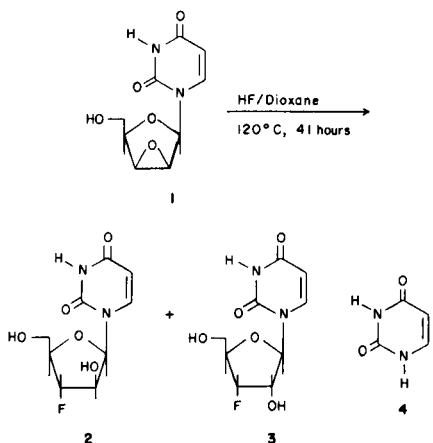
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Reaction of 1-(2',3'-epoxy- $\beta$ -D-lyxofuranosyl)uracil (**1**) with hydrogen fluoride afforded 3'-fluoro-3'-deoxy-arabinouridine (**2**, 13%) and 3'-fluoro-3'-deoxyuridine (**3**, 11%). The structure of **3** was assigned from spectroscopic data and confirmed by an unambiguous synthesis from 2',5'-di-*O*-trityl-2,3'-anhydrouridine (**5**) and hydrogen fluoride.

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Pyrimidine nucleosides having a halogen atom at the 2'-position of 2'-deoxyuridine [1-4] and the 3'-position of 3'-deoxyarabinouridine [5-7] have been investigated as potential diagnostic agents for tumor localization and as antitumor and antiviral agents [8]. The transport of 2'-halogeno-2'-deoxyuridines by the human erythrocyte nucleoside transporter has been reported [9].

3'-Fluoro-3'-deoxyarabinouridine (**2**) was required for a study to investigate the interaction of 3'-halo-3'-deoxyarabinouridines with the human erythrocyte nucleoside transporter [10]. Reaction of **1** with hydrogen fluoride in anhydrous dioxane at 120° for 41 hours using the procedure reported by Fox [11] gave a complex mixture of products. An extensive chromatographic separation of the reaction mixture using column and thin layer chromatography afforded **2** (13%), the unexpected **3** (11%) and uracil **4**, (67%).



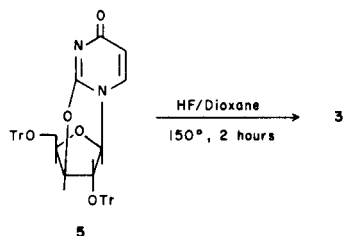
The structure of the unexpected product **3** was initially assigned on the basis of the circular dichroism (cd), ultraviolet (uv), mass (ms) and proton nuclear magnetic resonance ( $^1\text{H}$  nmr) spectra. Ulbricht and coworkers [12] have reported that the cd spectrum can be used to determine the configuration of pyrimidine nucleosides at the anomeric center. This empirical rule is based on the experimen-

tal cd curves of pyrimidine  $\beta$ -nucleosides which exhibited positive  $B_{2u}$  ( $\pi \rightarrow \pi^*$  transition) Cotton effects. The cd spectrum of **3** in aqueous solution was similar to that of uridine [13] with a positive Cotton effect at 262 nm thereby suggesting that **3** has the  $\beta$ -configuration. The uv spectrum of **3** (water) exhibited a  $\lambda$  max at 262 nm and a  $\lambda$  min at 232 nm ( $p\text{H} = 7.2$ ) and a  $\lambda$  max at 262 nm and a  $\lambda$  min at 243 nm ( $p\text{H} = 12$ ). These values are consistent with those reported earlier for pyrimidine nucleosides having the ribo configuration suggesting that the sugar moiety is attached to the N-1 position of uracil [14]. The high resolution ms of **3** exhibited a molecular ion at  $m/z$  246 ( $M^+$  Calcd. for  $C_9H_{11}N_2O_5F$ : 246.0655; Found: 246.0651) as well as a prominent peak (31.8%) at  $m/z$  135 ( $M^+$  Calcd. for  $C_5H_8O_3F$ : 135.0459; Found: 135.0457) due to the fluorinated sugar moiety.

The  $^1\text{H}$  nmr spectrum of **3** (acetone- $d_6$ -deuterium oxide) exhibited a *trans*-coupling for  $H_{1'}$  ( $J_{1',2'} = 1.5$  Hz) at 5.74  $\delta$  which would indicate that **3** must be a  $\beta$ -ribo or  $\alpha$ -arabino pyrimidine nucleoside [15]. The cd spectrum for **3** described previously was consistent with that expected for a compound having the  $\beta$ -ribofuranoside configuration [12-13]. The coupling constant  $J_{2',3'} = 4.5$  Hz suggests that the 2'- and 3'-hydrogens must be *cis* to each other ( $\phi_{H_2'H_3'} \sim 25\text{-}30^\circ$ ). It therefore follows that the 3'-fluoro substituent must be in the ribo (down) configuration. The *trans*  $J_{3',4'}$  coupling constant of 2 Hz is compatible with a 3'-ribo fluoro substituent. The assignment of  $H_{1'}$  ( $\delta$  5.74),  $H_{2'}$  ( $\delta$  4.09) and  $H_{3'}$  ( $\delta$  4.82) are consistent with the vicinal ( $J_{2',3'-F} = 4.5$  Hz,  $J_{3'-F,4'} = 32$  Hz) and geminal H-F ( $J_{3',3'-F} = 50$  Hz) coupling constants [11,16]. The large proton-fluorine coupling constants allow ready assignments of the protons by first order analysis. A long range hydrogen-fluorine coupling of 3.0 Hz between the anomeric  $H_{1'}$  proton and the C-3' fluorine was also present. The geminal proton-fluorine coupling constant ( $J_{3',3'-F} = 50$  Hz) is within the range normally associated with a  $sp^3$

carbon in a five membered furanose ring [17]. The  $^{19}\text{F}$  nmr spectrum in acetone-deuterium oxide relative to hexa-fluorobenzene appeared as a d,  $J_{3',3'}\text{F} = 50$  Hz, of d,  $J_{3',4'}\text{F} = 32$  Hz of d,  $J_{2',3'}\text{F} = 4.5$  Hz of d,  $J_{1',3'}\text{F} = 3.0$  Hz at  $\delta -42.5$ . A small  $J_{3',5'}\text{F}$  coupling was also present. The nmr data listed above is consistent with the assigned structure **3**. All assignments were confirmed by double resonance experiments.

The structure assigned to **3** was confirmed by an unambiguous alternate synthesis. Reaction of 2',5'-di-*O*-trityl-2,3'-anhydrouridine (**5**) with anhydrous hydrogen fluoride in dioxane at  $150^\circ$  for 2 hours gave **3** in 66% yield. When the same reaction was carried out at  $120^\circ$  for 21 hours the only product obtained was uracil (**4**). The spectrometric data (uv, ms,  $^1\text{H}$  nmr) for **3** prepared in this way was identical to that obtained for **3** prepared from reaction of the epoxide **1** with hydrogen fluoride in dioxane.



It was also of interest to investigate the reaction pathway by which the unexpected ribo nucleoside **3** was formed. When 3'-fluoro-3'-deoxyarabinouridine (**2**) was heated at  $120^\circ$  in anhydrous hydrogen fluoride-dioxane for 41 hours the ribo nucleoside **3** (25%), uracil **4** (22%) and unreacted **2** (45%) was obtained. On the other hand, a similar treatment of **3** with anhydrous hydrogen fluoride-dioxane afforded uracil (**4**, 95%). These results suggest that **3** arises from further reaction of **2** and uracil (**4**) arises from **3**. The mechanism by which **3** arises from **2** is not known.

## EXPERIMENTAL

Melting points were determined with a Thomas-Hoover apparatus and are uncorrected. Ultraviolet spectra were taken on a Pye Unicam SP 1800 spectrometer. The nmr spectra were recorded on Bruker WH 200, AM 300 and WH 400 spectrometers. Mass spectra were determined on an AEI MS-50 spectrometer using direct probe introduction at  $200^\circ$  and 70 eV. The cd spectrum was recorded on a ORD/UV-5 Japan Spectroscopic Co. spectrometer. Thin layer chromatography (tlc) was performed on Whatman MK6F silica gel plates ( $1 \times 3''$ ,  $200 \mu$  thickness). Preparative tlc was performed on Whatman PLK5F silica gel plates ( $20 \times 20$  cm,  $1000 \mu$  thickness). The solvent systems used were A, chloroform:methanol (10:1 v/v) and B, the separated upper phase of ethyl acetate:*n*-propanol:water (4:1:2 v/v/v). Column chromatography was performed using a 1:1 mixture of Merck Kieselgel 60 (70-230 mesh) and Mallinckrodt silicic acid (100 mesh) with chloroform:methanol:acetone (10:1:1 v/v/v) as eluant. Dioxane was dried by distillation from calcium hydride and collection under a nitrogen atmosphere prior to use. 2',5'-Di-*O*-trityl-2',3'-anhydrouridine (**5**) was prepared according to the literature procedure [18].

3'-Fluoro-3'-deoxyarabinouridine (**2**), 3'-Fluoro-3'-deoxyuridine (**3**) and Uracil (**4**).

A mixture of 1-(2',3'-epoxy- $\beta$ -D-lyxofuranosyl)uracil (**1**) (1.13 g, 5 mmoles) and liquid hydrogen fluoride (15 g, 15 ml, 0.75 mole) in dry dioxane (150 ml) was heated at  $120^\circ$  for 41 hours in a monel pressure cylinder. Water (60 ml) was added to the cooled reaction mixture which was neutralized with anhydrous sodium carbonate. The solids were removed by filtration and the solvent removed *in vacuo*. The residue was dissolved in absolute ethanol and filtered. The filtrate was concentrated *in vacuo* to give a dark brown residue which was purified on a  $2.5 \times 40$  cm silica gel column. The first eluted product was collected and purified further by preparative tlc using solvent system B. Extraction of the spot having  $R_f$  0.15 with solvent system A and recrystallization from absolute ethanol gave **2** as a white crystalline solid (0.156 g, 13%), mp  $179-180^\circ$  (lit [11] mp  $179.5-180.5^\circ$ ); high resolution ms: exact mass calcd. for  $\text{C}_9\text{H}_{11}\text{N}_2\text{O}_5\text{F}$  246.0655; found: 246.0650. The  $^1\text{H}$  nmr spectrum in acetone- $d_6$ -deuterium oxide was identical to the published spectrum and physical data [11]. Further elution gave a second compound which was purified further by preparative tlc using solvent system B. Extraction of the spot having  $R_f$  0.09 with solvent system A and recrystallization from absolute ethanol afforded **3** as a white crystalline solid (0.13 g, 11%), mp  $195-196^\circ$ ; uv (water, pH = 7.2):  $\lambda$  max 262,  $\lambda$  min 243 nm; cd (water):  $\lambda$  max 262 nm,  $\lambda$  min 232 nm; uv (pH = 12):  $\lambda$  max 262 nm, and  $\theta$  max + 13,300;  $^1\text{H}$  nmr:  $\delta$  3.75 (d,  $J_{5',5''} = 11$  Hz of d,  $J_{4',5'} = 11.5$  Hz of d,  $J_{3',5'} = 1.5$  Hz, 1H, H-5'), 3.95 (d,  $J_{5',5''} = 11$  Hz, of d,  $J_{4',5''} = 5.5$  Hz, 1H, H-5''), 4.08 (d,  $J_{2',3'} = 4.5$  Hz, of d,  $J_{2',3'}\text{F} = 4.5$  Hz of d,  $J_{1',2'} = 1.5$  Hz, 1H, H-2'), 4.12 (d,  $J_{3',4'} = 32$  Hz of d,  $J_{4',5'} = 11.5$  Hz of d,  $J_{4',5''} = 5.5$  Hz of d,  $J_{3',4'} = 2.0$  Hz, 1H, H-4'), 4.82 (d,  $J_{3',3'}\text{F} = 50$  Hz of d,  $J_{2',3'} = 4.5$  Hz of d,  $J_{3',4'} = 2$  Hz, 1H, H-3'), 5.6 (d,  $J_{5,6} = 8$  Hz, 1H, H-5), 5.73 (d,  $J_{1',3'}\text{F} = 3.0$  Hz of d,  $J_{1',2'} = 1.5$  Hz, 1H, H-1'), 7.67 (d,  $J_{5,6} = 8$  Hz, 1H, H-6); high resolution ms: exact mass calcd. for  $\text{C}_9\text{H}_{11}\text{N}_2\text{O}_5\text{F}$  246.0655; found: 246.0651.

Anal. Calcd. for  $\text{C}_9\text{H}_{11}\text{N}_2\text{O}_5\text{F}$ : C, 43.90; H, 4.50; N, 11.38. Found: C, 43.69; H, 4.69; N, 10.94.

Further elution gave uracil (**4**, 0.38 g, 67.8%) identical (tlc,  $^1\text{H}$  nmr) with an authentic sample.

### Reaction of 3'-Fluoro-3'-deoxyarabinouridine (**2**) With Hydrogen Fluoride.

A mixture of **2** (10 mg) and liquid hydrogen fluoride (0.5 ml) in dry dioxane (2 ml) was heated at  $120^\circ$  for 41 hours. The reaction was completed and the reaction mixture purified as described previously (column and tlc) to yield **3** (2.5 mg, 25%), uracil (1.0 mg, 22%) and unreacted **2** (4.5 mg, 45%).

### Reaction of 3'-Fluoro-3'-deoxyuridine (**3**) With Hydrogen Fluoride.

A mixture of **3** (1 mg) and liquid hydrogen fluoride (0.2 ml) in dry dioxane (1 ml) was heated at  $120^\circ$  for 41 hours. The reaction was completed and the reaction mixture purified as described previously to give uracil (95%) as the sole product.

### 3'-Fluoro-3'-deoxyuridine (**3**).

A mixture of 2',5'-di-*O*-trityl-2,3'-anhydrouridine (**5**) (36 mg, 0.05 mmole) and liquid hydrogen fluoride (0.2 ml) in dry dioxane (2 ml) was heated at  $150^\circ$  for 2 hours in a monel pressure cylinder. Water (1 ml) was added to the cooled solution which was neutralized with solid calcium carbonate. The solids were removed by filtration and the solvent removed *in vacuo*. The residue was dissolved in absolute ethanol and filtered. The filtrate was concentrated *in vacuo* to give a dark brown residue which was separated by column chromatography. An impure fraction was obtained which was purified on preparative tlc using solvent system B-ethyl acetate (4:1 v/v) to give **3** (8 mg, 66%). 3'-Fluoro-3'-deoxyuridine (**3**) prepared in this way was identical (mp,  $^1\text{H}$  nmr, ms) to **3** obtained from reaction of the epoxide **1** with hydrogen fluoride.

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