COMMUNICATIONS

- New or improved synthetic methods
- Key intermediates
- with full experimental and analytical data

A Convenient Synthesis of 5-Methyluridine from Uridine

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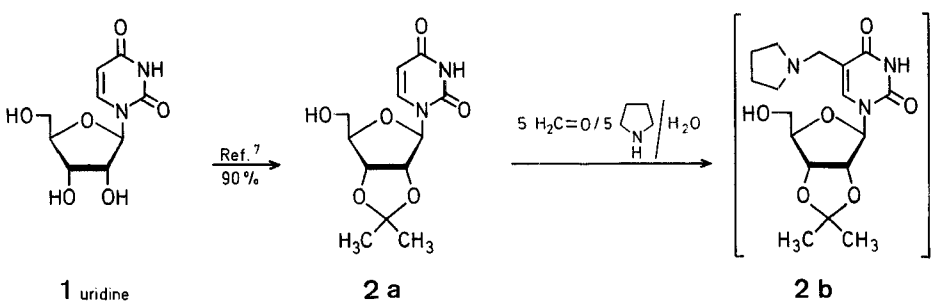
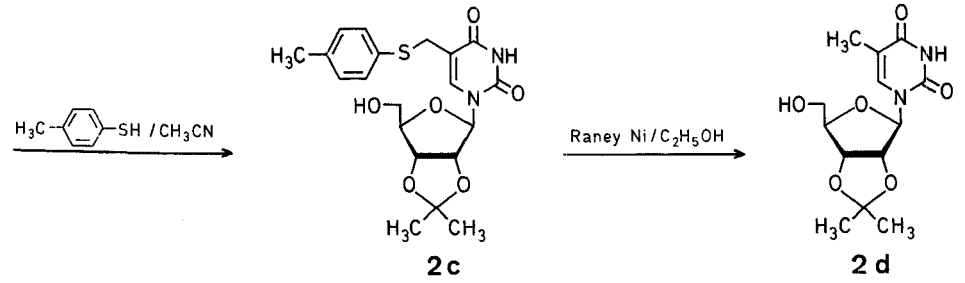
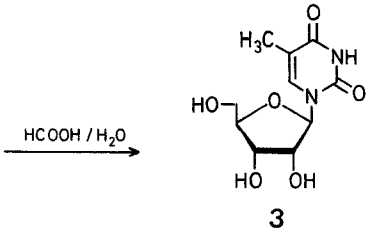
In connection with our work on the synthesis of oligoribonucleotides[1], we required a convenient source of 1-β-D-ribofuranosylthymine (5-methyluridine; **3**). The latter compound **3**, which occurs very widely as a modified nucleoside in transfer ribonucleic acids (tRNA)[2], may be prepared in two steps and in satisfactory yield from 2,3,5-tri-*O*-benzoyl-D-ribofuranosyl chloride and thymine[3] or a simple derivative of thymine[4,5]. However, in view of the fact that the 5-methyluracil residues in tRNA are derived from uracil residues by enzyme-promoted methylation[6], the development of a chemical synthesis of 5-methyluridine (**3**) from uridine (**1**) seemed to us to be an undertaking of particular interest. Furthermore, uridine (**1**) is a relatively inexpensive commercially-available starting material.





When 2′,3′-*O*-isopropylideneuridine (**2a**), which may readily be prepared from uridine (**1**) in over 90% yield[7], was heated with approximately five molar equivalents each of formaldehyde and pyrrolidine in aqueous solution, under reflux, for 1 h, it was completely consumed and a product believed to be the Mannich base (**2b**) was obtained. The crude products were not purified but were heated directly with one molar equivalent of *p*-thiocresol in dry acetonitrile solution, under reflux, for 15 min to give 2′,3′-*O*-isopropylidene-5-(*p*-tolylthiomethyl)-uridine (**2c**). The latter compound **2c** was isolated as a colourless crystalline solid, m.p. 145–146 °C, in 69% overall yield based on **2a**. When **2c** was heated, under reflux, with an excess of Raney nickel[8] in ethanol for 1.5 h, 2′,3′-*O*-isopropylidene-5-methyluridine (**2d**) was obtained and isolated as a crystalline solid in 66% yield. The desired unprotected 5-methyluridine (**3**) was readily obtained as a crystalline solid, isolated in 86% yield, following treatment of **2d** with formic acid/water (9 : 1 v/v) for 1 h at room temperature. Thus the overall yield of 5-methyluridine (**3**) for the five steps starting with uridine (**1**) was ~36% and this yield has not yet been optimized. By increasing the quantity of *p*-thiocresol from one to two molar equivalents, S. Sibanda has recently increased the yield for the conversion of **2a** into **2c** from 69 to 79%. Thus, the overall yield for the conversion of **1** into **3** has been increased to nearly 42%.



There are several previous reports in the literature relating to the conversion of uridine (**1**) into its 5-methyl derivative (**3**). The first report[9] is concerned with the platinum oxide-catalyzed hydrogenolysis of 5-hydroxymethyluridine which leads to a poor overall yield of **3**. The second[10], which perhaps is the most closely related to the present report, is concerned with the platinum oxide-catalyzed hydrogenolysis of 5-diethylaminomethyluridine leading to 5-methyluridine (**3**) in 24% overall yield. In addition, Salisbury and Brown[11] have recently reported that sodium borohydride reduction of the methiodide of the Mannich base 2′,3′-*O*-isopropylidene-5-dimethylaminomethyluridine leads to **2d**. However, in our hands, the reaction between the methiodide of **2b** and sodium borohydride leads to a mixture containing **2d** and what appears

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(by ^{1}H-N.M.R.) to be further reduction products. A third report[12] relates to a method which involves a key photochemical step and which does not appear to be practicable from a preparative point of view. We therefore feel able to conclude that the procedure reported in this paper is the procedure of choice for the conversion of uridine **(1)** into 5-methyluridine **(3)**.

2′,3′-*O*-Isopropylidene-5-(*p*-tolylthiomethyl)-uridine (2c):
Pyrrolidine (14.7 ml, 0.176 mol) and 40% aqueous formaldehyde (13.2 ml, 0.171 mol) are added to a suspension of 2′,3′-*O*-isopropylideneuridine[7] (**2a**; 10.0 g, 35 mmol) in water (180 ml). The mixture is then heated, under reflux, for 1 h when T.L.C. [Merck silica gel 60 F_{254} plates, developed in chloroform/methanol (9:1 v/v)] reveals the presence of ultraviolet absorbing material only with $R_f \sim 0$. The cooled solution is concentrated to dryness under reduced pressure and the residue obtained is evaporated with toluene, ethanol (3 times), and chloroform.

p-Thiocresol (4.37 g, 35 mmol) is added to a solution of the residual glass obtained in acetonitrile (100 ml) and the resulting solution is heated, under reflux, for 15 min. The cooled products are evaporated under reduced pressure and the oil obtained is triturated with cyclohexane. The residual glass is purified by short column chromatography[13] on silica gel (120 g). The appropriate fractions eluted from the column with chloroform/ethanol (94:6 v/v) are combined and evaporated under reduced pressure. Crystallization of the residue from aqueous ethanol gives the desired product **2c**; yield: 10.13 g (69%, based on **2a**); R_f [chloroform/methanol (9:1 v/v)]: 0.42; m.p. 145–146 °C.

$C_{20}H_{24}N_2O_6S$	calc.	C 57.13	H 5.75	N 6.66
(420.5)	found	56.8	5.9	6.7

U.V. (95% C_2H_5OH): $\lambda_{max} = 260$ ($\varepsilon = 12\,100$); $\lambda_{min} = 243$ nm ($\varepsilon = 6900$).

^{1}H-N.M.R. (250 MHz, DMSO-d_6): $\delta = 1.28$ (s, 3H); 1.47 (s, 3H); 2.28 (s, 3H); 3.5 (m, 2H); 3.68 (d, $J = 13.3$ Hz, 1H); 3.75 (d, $J = 13.3$ Hz, 1H); 4.0 (m, 1H); 4.5 (m, 1H); 4.6 (m, 1H); 5.1 (m, 1H); 5.77 (d, $J = 2.8$ Hz, 1H); 7.15 (d, $J = 7.8$ Hz, 2H); 7.24 (d, $J = 8.3$ Hz, 2H); 7.52 (s, 1H); 11.54 ppm (br s, 1H).

2′,3′-*O*-Isopropylidene-5-methyluridine (2d):
2′,3′-*O*-Isopropylidene-5-(*p*-tolylthiomethyl)-uridine (**2c**; 4.21 g, 10 mmol) and Raney nickel[8] (wet paste, ~20 g) are heated, under reflux, in ethanol (100 ml) for 1.5 h. The cooled products are filtered through Celite and the filtrate is concentrated under reduced pressure. The residual glass is, without further purification, crystallized from water (50 ml) to give the desired product **2d**; yield (in two crops): 2.03 g (66%); R_f [chloroform/methanol (9:1 v/v)]: 0.36; m.p. 86–87 °C.

$C_{13}H_{18}N_2O_6 \cdot 0.5\,H_2O$	calc.	C 50.81	H 6.23	N 9.12
(307.3)	found	50.9	6.1	9.1

U.V. (95% C_2H_5OH): $\lambda_{max} = 265$ ($\varepsilon = 9100$), $\lambda_{min} = 236$ nm ($\varepsilon = 2200$).

^{1}H-N.M.R. (250 MHz, DMSO-d_6): $\delta = 1.28$ (s, 3H); 1.48 (s, 3H); 1.76 (d, $J = 0.9$ Hz, 3H); 3.6 (m, 2H); 4.0 (m, 1H); 4.75 (dd, $J = 3.7$ and 6.4 Hz, 1H); 4.88 (dd, $J = 3.2$ and 6.4 Hz, 1H); 5.1 (m, 1H); 5.83 (d, $J = 2.8$ Hz, 1H); 7.65 (quart, $J = 0.9$ Hz, 1H); 11.2 ppm (br s, 1H).

5-Methyluridine (3):
2′,3′-*O*-Isopropylidene-5-methyluridine (**2d**; 1.752 g, 5.9 mmol) is dissolved in formic acid/water (9:1 v/v; 20 ml) and the solution is allowed to stand at room temperature. After 1 h, the products are concentrated under reduced pressure and the residue is re-evaporated from ethanol (6 times). The resulting gum is dissolved in methanol (20 ml) and the solution is slightly basified by the addition of dilute methanolic ammonia. After 1 h, the solution is concentrated under reduced pressure and the residual glass is crystallized from ethanol (25 ml) to give the desired product **3**; yield (in two crops): 1.31 g (86%); R_f [chloroform/methanol (9:1 v/v)]: 0.03; m.p. 177.5–178.5 °C (Lit.[4], m.p. 183–185 °C).

$C_{10}H_{14}N_2O_6 \cdot 0.5\,H_2O$	calc.	C 44.94	H 5.66	N 10.48
(267.2)	found	45.2	5.5	10.3

U.V. (95% C_2H_5OH): $\lambda_{max} = 267$ ($\varepsilon = 8800$), $\lambda_{min} = 238$ nm ($\varepsilon = 2100$).

^{1}H-N.M.R. (250 MHz, DMSO-d_6): $\delta = 1.77$ (d, $J = 0.9$ Hz, 3H); 3.6 (m, 2H); 3.8 (m, 1H); 3.9–4.1 (m, 2H); 5.05–5.2 (m, 2H); 5.3 (m, 1H); 5.78 (d, $J = 5.5$ Hz, 1H); 7.74 (quart, $J = 0.9$ Hz, 1H); 11.28 ppm (br s, 1H).

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