# 1143. *Nucleosides. Part III.*[1] *Synthesis of 1-Methyl-2′-deoxycytidine, and Comments on the N.m.r. Spectra of Cytosine Nucleosides*

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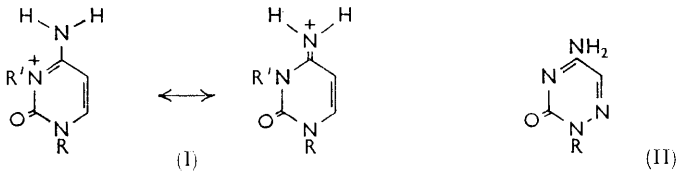
The synthesis of 1-methyl-2′-deoxycytidine, obtained by methylation of 2′-deoxycytidine with dimethyl sulphate, is described. The ultraviolet spectra at different pH values were identical with those reported for 1-methylcytidine, which had been synthesised previously in an exactly analogous manner from cytidine (P. Brooks and P. D. Lawley, *J.*, 1962, 1348) and whose structure had been unambiguously established.

In a previous publication [2] the n.m.r. spectra of cytidine, 2′-deoxycytidine, and 1-methyl-2′-deoxycytidine were compared, in order to establish the tautomeric form of 2′-deoxycytidine.[3] The fact that the n.m.r. spectra of the hydrochlorides of all three of these cytosine derivatives show two N–H singlets [2] was attributed to the amino-group, the protons of which are in a different environment from each other owing to the partial double-bond character of the $C^6$–$NH_2$ bond in the protonated species (I). N.m.r. studies by other workers [4] have shown that cytosines protonate at $N^1$ and that there is restricted rotation about the bond between $C^6$ and the exocyclic amino-group, thus confirming the above suggestion.



(I) (II)

R = ribofuranosyl or 2′-deoxyribofuranosyl; R′ = H or Me

Another nucleoside for which a tautomeric imino-structure has been proposed is the cytidine analogue, azacytidine (II). Zemlicka, Beranek, and Smrt suggested [5] that an imino-structure would explain the fact that, at neutral pH, azacytidine has only one absorption in the ultraviolet, whereas cytidine has two, and the marked difference between the ultraviolet spectra of an *N*-acetylazacytidine ($\lambda_{max.}$ 262 mμ) and an *N*-acetylcytidine ($\lambda_{max.}$ 245, 297 mμ). However, subsequent studies of the infrared spectrum of azacytidine clearly favoured the amino-structure (II).[6]

The n.m.r. spectrum of azacytidine (II; R = ribofuranosyl) in dimethyl sulphoxide has been examined. Like cytidine and 2′-deoxycytidine, and unlike 1-methyl-2′-deoxycytidine [2] (which necessarily has an imino-structure), the n.m.r. spectrum of azacytidine shows a two-proton singlet (at 2·09 τ); both the position of this peak and its gradual disappearance after addition of deuterium oxide indicate that it is due to the $NH_2$ group. This evidence supports the amino-structure (II) for azacytidine.

## Experimental

*1-Methyl-2′-deoxycytidine.*—A solution of deoxycytidine (500 mg.) in dimethylformamide (5 ml.) was left with dimethyl sulphate (2 ml.) at 37° for 40 min. The solution was diluted to 20 ml. with dry methanol, ethyl acetate added in portions, and cooled overnight, to give 580 mg.

[1] Part II, T. L. V. Ulbricht and G. T. Rogers, preceding Paper.
[2] T. L. V. Ulbricht, *Tetrahedron Letters*, 1963, 1027.
[3] See L. Gatlin and J. C. Davis, jun., *J. Amer. Chem. Soc.*, 1962, **84**, 4464; H. T. Miles, *ibid.*, 1963, **85**, 1007.
[4] A. R. Katritzky and A. J. Waring, *J.*, 1963, 3046; H. T. Miles, R. G. Bradley, and E. D. Becker, *Science*, 1963, **142**, 1569.
[5] J. Zemlicka, J. Beranek, and J. Smrt, *Coll. Czech. Chem. Comm.*, 1962, **27**, 2784.
[6] J. Pitha and J. Beranek, *Coll. Czech. Chem. Comm.*, 1963, **28**, 1507.

(75%) of 1-*methyl*-2′-*deoxycytidine methosulphate*, m. p. 146° (Found: C, 37·6; H, 5·5; N, 11·6. $C_{11}H_{19}N_3O_8S$ requires C, 37·4; H, 5·4; N, 11·9%). The free base was prepared by gradual addition of Dowex-1 hydroxide to an aqueous solution of the methosulphate, with filtration after the pH reached 4, 5, and finally 6. In this way, loss of material by absorption on large quantities of resin near neutrality was avoided. Freeze-drying left a crystalline product which was extremely hygroscopic and therefore dissolved directly in dimethyl sulphoxide for n.m.r. studies. A portion was converted into the *hydrochloride*, m. p. 160° (decomp.) (from methanol) (Found: C, 42·7; H, 6·1; N, 15·1. $C_{10}H_{16}ClN_3O_4$ requires C, 43·2; H, 5·8; N, 15·1%), $\lambda_{max.}$ (pH 4) 278, (pH 12) 266.

Nuclear magnetic resonance spectra were obtained at 60 Mc./sec. using a Varian A-60 spectrometer at Imperial College, by kind permission of Professor D. H. R. Barton, F.R.S. The azacytidine used was a chromatographically homogeneous sample (Sigma Chemical Co.); its ultraviolet spectrum was identical with that reported.[5]

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