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A Convenient Synthesis of 6-Aza-2'-deoxycytidine (1)

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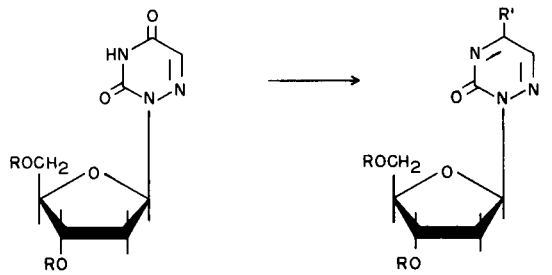
Interest in the biological properties of 6-aza-2'-deoxycytidine (IV) (2) has been aroused by the report that IV can activate the biosynthesis of deoxyribonucleic acid in Ehrlich ascites cells (3). In order to provide material for biochemical studies, we have synthesized IV, employing a modified procedure that is shorter and more convenient than the original (2).

By an adaptation (4) of the method of Nishimura and Iwai (5), 6-azauracil was converted to its bis-(trimethylsilyl) ether (6). The unpurified ether was condensed with 2-deoxy-3,5-di-*O*-*p*-toluoyl-D-ribofuranosyl chloride (V) (8) by two procedures to afford the blocked 6-aza-2'-deoxyuridine (I) (9). The first procedure involved condensation in benzene at room temperature in the presence of mercuric acetate as acid acceptor (4). The second involved fusion at 95-100° without solvent (10). Although both methods afforded equal yields, the first method was preferred because of the easier workup and purification. The yield of 25% of I equaled that obtained by Pliml, *et al.*, (9). Their longer synthesis involved extra steps in selective blocking of 6-azauracil and deblocking after the nucleoside was formed. These extra steps were necessary to prevent any nucleoside bond formation at *N*-3 of 6-azauracil.

The properties of I agreed completely with those recorded in the literature (9) indicating that the sugar was attached to the 1-position, and that it was the β-anomer. No α-anomer was isolated although the mother liquors from the crystallization of I contained additional nucleoside product. Thin layer chromatography results suggested this to be non-crystalline I and/or its α-anomer, contaminated with some unreacted sugar. These results corroborate those of the only other reported use of bis(trimethylsilyl) ethers of 6-azauracils in nucleoside synthesis (7): namely, that the sugar is attached to the 1-position of the 6-azauracil and that the β-anomer is isolated.

The blocked 6-aza-2'-deoxyuridine (I) was then converted by the literature procedure (2) to the chloro nucleoside (III) which was isolated in two crystalline forms. Both forms were suitable for conversion to the desired 6-aza-2'-deoxycytidine (IV). The properties of IV agreed completely with those reported in the literature (2).



I, R = p-toluoyl

II, R = H

III, R = p-toluoyl, R' = Cl

IV, R = H, R' = NH_2

EXPERIMENTAL (11)

1-(2'-Deoxy-3',5'-di-*O*-*p*-toluoyl-β-D-ribofuranosyl)-6-azauracil (I).

Method A.

By a literature procedure (4), 2.83 g. (25 mmoles) of 6-azauracil was treated with 0.3 ml. of trimethylchlorosilane in 15 ml. of hexamethyldisilazane at reflux temperature for 1 hour to afford, after evaporation, 10.2 g. (158%) of the bis(trimethylsilyl) ether of 6-azauracil as a yellow syrup. This ether in 20 ml. of dry benzene was added to 10.2 g. (26.2 mmoles) of freshly prepared 2-deoxy-3,5-di-*O*-*p*-toluoyl-D-ribofuranosyl chloride (V) (8) in 400 ml. of dry benzene. After 8.10 g. (25.4 mmoles) of mercuric acetate was added (4), the reaction mixture was stirred and protected from moisture for 3 days. The mixture was treated with charcoal and filtered. The filtrate was washed successively with 75 ml. of 30% potassium iodide solution, twice with 100-ml. portions of water, dried (magnesium sulfate), filtered and evaporated *in vacuo* to an orange foam. This was dissolved in 150 ml. of benzene, filtered, and the hot solution was diluted with 200 ml. of hot petroleum ether, b.p. 90-100°, to afford 3.25 g. of I. Recrystallization from methanol afforded 2.95 g. (25%) of I as white needles, m.p. 177.5-179.0°; $[\alpha]_{589}^{23}$ -65° (*c* 2.3, pyridine); ultraviolet maxima (ethanol) at 241 mμ (ϵ, 36.0 x 10^3), 270 (shoulder; ϵ, 8.1 x 10^3), homogeneous by TLC with R_f 0.6 in solvent A and R_f 0.2 in solvent B. Literature values (9); m.p. 179°; $[\alpha]_D^{22}$ -67° (*c* 2.2 in pyridine).

Substitution of *N*,*N*-dimethylformamide or dimethylsulfoxide for part of the benzene solvent in the condensation was unsuccessful. In neither case could any crystalline I be isolated from the reaction.

Some I was deacylated to afford II whose ultraviolet spectrum was determined as additional confirmation of 1-substitution in the 6-azauracil. For II, the ultraviolet maximum (ethanol) was at 265 mμ (ϵ, 4,850); literature value (9): 267 mμ (ϵ, 4,900).

Method B.

A mixture of freshly prepared bis(trimethylsilyl) ether from 1.22 g. (10.8 mmoles) of 6-azauracil and 4.44 g. (11.4 mmoles) of freshly prepared chlorosugar V was stirred and fused at 95-100° *in vacuo* (25 mm.) for 30 minutes (10). The amber melt was cooled, dissolved

in 100 ml. of chloroform and 5 ml. of ethanol, stirred overnight, filtered, and the filtrate was evaporated to dryness. Recrystallization of this residue from benzene afforded only 1.12 g. of product and it was necessary to chromatograph the mother liquor on silica gel (70 g.) with ether as the eluting agent to obtain an additional 0.28 g. of product. Recrystallization of the combined products from methanol afforded 1.26 g. (25%) of I, m.p. 178.5-179° and having the same properties as I prepared by method A.

2-(3',5'-Di-*O*-*p*-toluoyl-2'-deoxy-β-D-ribofuranosyl)-5-chloro-1,2,4-triazine-3(2*H*)-one (III).

Treatment of 2.18 g. (4.67 mmoles) of I with thionyl chloride by the literature method (2) afforded two crystalline forms of III, differing in their infrared spectra in the 7.6-12 μ region. The granular crystals (0.854 g.) had m.p. 161-166.5°. The fibrous needles (1.016 g.; total 1.87 g., 83%) had a m.p. of 154-159° (softens from 140°). Pliml and Šorm (2) reported a 91% yield of III as yellow needles, m.p. 162°. Both forms of III were suitable for conversion to IV, yielding a product (67%) whose properties agreed with those reported in the literature.

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REFERENCES

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(6) No bis(trimethyl)silyl ethers of any 6-azauracils had been prepared or used in nucleoside synthesis when this work was initiated. Very recently, the bis(trimethyl)silyl ethers of two 5-substituted 6-azauracils were prepared and used in nucleoside synthesis (7).

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(11) Melting points were taken on a Fisher-Johns apparatus and are corrected. Thin layer chromatograms (TLC) were run on silica gel HF plates with these solvents; A, ethyl acetate; B, ether. The spots were detected by ultraviolet light.

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