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THE PHOTOREACTION OF N,N-1,3-DIMETHYLURACIL WITH ETHANOL

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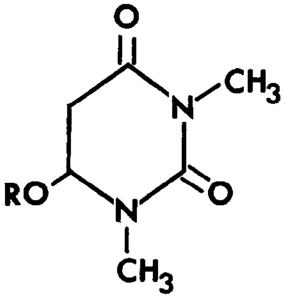
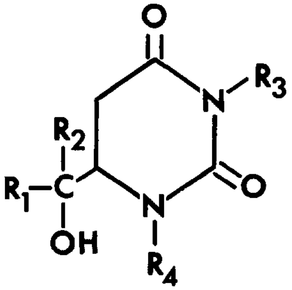
The photochemical reactions of uracil and its derivatives with hydroxylic solvents have been a subject of considerable interest among researchers interested in the photoreactivity of nucleic acids. Most of the published work has dealt with the photoreactivity of uracil and its analogs toward water; the observed reaction of uracil is addition of water across the 5,6-double bond to form an acid and heat reversible photohydrate, 6-hydroxy-5-hydrouracil. The soluble analog N,N-1,3-dimethyluracil (DMU) exhibits similar photochemical behavior in aqueous solution (1).

Considerably less work has been done on the photochemistry of uracil and its derivatives in alcoholic solution. The reported work on the behavior of uracil and DMU in primary alcohols (methanol and ethanol) indicates that ether type adducts (I) are the products of photolysis (2). On the other hand, Elad and co-workers (3) have recently shown that photolysis of DMU in isopropanol, a secondary alcohol, leads to adducts with structure IIa. Thus the published literature indicates a large difference in photochemical behavior between DMU-primary alcohol systems and DMU-secondary alcohol systems.

We report here results from studies on the DMU-ethanol system. We find at concentrations of 7 X 10^{-3} M that a product of structure IIb is produced almost exclusively. (A trace of DMU cyclobutane dimer (4) is produced, but there is no evidence for heat or acid reversible product.) Irradiations were conducted at $\lambda > 2600$ A, using a Corex filtered Hanovia 450 w medium pressure Hg lamp. (The wavelength cut off of the Corex filter was checked on a Cary 14 (5).) Compound IIb, which was first isolated as an oil, was crystallized from ethyl ether and had a melting point of 96-97°C. Mass spectrometry and CHN analysis were used to establish the molecular weight and elemental composition of IIb: MW = 186; for $C_8H_{14}N_2O_3$ calc. C (51.60) H (7.58) N (15.05), obs. C (51.60) H (7.55) N (14.89).





I

IIa R_1R_2,R_3R_4 = CH_3 IIb R_1,R_3,R_4 = CH_3,R_2 = H

IIc R_1 = C_2H_5 R_2 = H, R_3 = R_4 = CH_3

IId R_1 = $NH_2CHCOOH,R_2R_3$ = H, R_4 = Ribose

IR, NMR, and UV spectroscopy were used to establish the molecular structure of IIb. The IR spectrum showed a broad absorption centered at 3360 cm^{-1}, corresponding to the OH stretch, and amide carbonyl absorption at 1660 and 1710 cm^{-1}. The UV spectrum, run in distilled water on a Cary 14, showed a maximum at λ = 2275 (ε = 3820) and a minimum at λ = 2015 (ε = 2240). A region of weak tail absorption reached from 2600 A to 2850 A. The lack of a strong absorption peak in this region showed that the 5,6 double bond in the parent DMU had become saturated. The 60 Mc NMR spectrum, run in $CDCl_3$, indicated a doublet at δ = 1.22 (J = 6.5, 3 protons) suggesting that the alcohol had added in the manner shown in structure IIb. A considerably less intense doublet with δ = 1.21 (J = 6.5) could be resolved under very slow sweep conditions, indicating that both possible enantiomorphic pairs were formed in the photoreaction. Other NMR peaks were as follows: overlapping peaks in region between δ = 3.0 and 3.5 (7 H), an indistinct quintet centered at about δ = 4.0 (1 H) superimposed on a broad peak which disappeared when the sample was shaken with D_2O, and the AB part of an ABX pattern (a sharp peak at δ = 2.84 and a broad peak, possibly a doublet, at 2.76) (6). The point of attachment of the hydroxyethyl group to the pyrimidine ring was determined by reacting 5-d-DMU (7) with ethanol. The two proton pattern at δ = 2.84 and 2.76 was replaced by half of an AX pattern containing one proton.

Irradiation of DMU-n-propanol systems (7×10^{-3} M) leads to an adduct which has an NMR spectrum consistent with structure IIc. It is possible that reactions similar to the photoinduced reaction of DMU with ethanol may have synthetic potential in synthesizing 6-α-hydroxyalkyl-5-hydrouracils.

In view of the difference between our results and those previously reported on DMU-ethanol systems, we also irradiated solutions of DMU in ethanol and isopropanol at 10^{-4}M. In the DMU-ethanol system when 25% of the DMU was photolyzed at $\lambda > 2600$ A approximately 40% of the reacted DMU could be reconverted to DMU in 0.5 HCl, as evidenced by recovery of absorbance at the absorption maximum of DMU. When the 10^{-4}M DMU-isopropanol system was photolyzed until 40% of the inital absorbance had disappeared, about 20% of the lost absorbance could be recovered by acid treatment. Presumably the restoration of absorbance results from decomposition of compounds of structure I to form DMU and parent alcohol. Gas liquid chromatography of concentrated irradiated solution on a SE - 30 column at 230° indicated the presence of IIb as well in the 10^{-4} DMU-ethanol reaction mixture. It thus appears the type and distribution of photoproducts resulting from irradiation of DMU in alcoholic solution is strongly dependent upon the concentration of DMU.

In both the photolyzed 10^{-4} M DMU-ethanol and DMU-isopropanol systems UV absorbance also appears, upon acid treatment, in the region between 3000 A and 3500 A. Treatment of IIb with 0.5 HCl did not lead to appearance of absorbance in this region. It is possible that other products, besides the types of compounds discussed in this paper, may be produced upon photolysis of DMU in alcoholic solution at 10^{-4} M concentrations.

The mechanism of the reaction of DMU with ethanol to form IIb is probably radical in nature. Light induced decomposition ($\lambda > 3000$ A) of di-t-butyl peroxide (10 ml) in 200 ml of ethanol containing 400 mg of DMU leads to production of IIb and 2,3-butanediol. This result suggests that a radical chain mechanism, with DMU acting as a photoinitiator, may be operating in the DMU-ethanol system itself.

There is some evidence (8) that serine, for which ethanol can be considered an analog, reacts with photoexcited uracil. The results reported here indicate that reaction to form compound IId may occur. Formation of such adducts in biological systems could lead to photoinduced crosslinking between ribonucleic acid and proteins, e.g., in the observed photoinduced crosslinking between $\mathrm{tRNA}^{\mathrm{tyr}}$ and tyrosyl-tRNA synthetase (9).

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