PHOTOCYCLOADDITION OF CYTOSINE AND 2'-DEOXYCYTIDINES TO 2,3-DIMETHYL-2-BUTENE

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Abstract - Irradiation of far-uv light of cytosine, 2'-deoxycytidine and its *N*-acetyl-3',5'-di-*O*-acetyl derivative in the presence of 2,3-dimethyl-2-butene in acetone yielded the corresponding cyclobutane photoadducts.

There is evidence to show that cyclobutane-containing cytosines are produced as photoproducts in DNA by 280 nm irradiation. [1] Despite the biological implications of these cytosine dimers, much less has been studied about the photochemistry of cytosine and its derivatives than uracil and thymine due to the fact that they are exceedingly labile under general experimental conditions. [2,3] On the other hand, Swenton and Wexler [4~7] have studied the regioselective photoreaction of pyrimidine bases with simple olefins, and also Charlton and Lai [8] reported the photoaddition of pyrimidine nucleosides to simple olefins. However, their studies did not involve cytosine and its nucleosides as the photoreaction substrates. To our knowledge, no one have studied on photoreactions of cytosine and its nucleosides with simple alkenes.

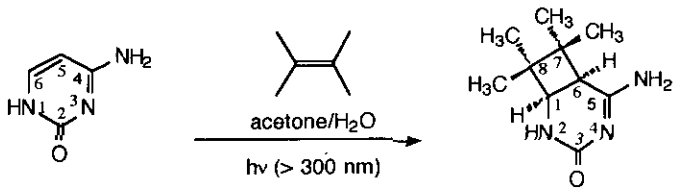
We have recently revealed that a pair of cyclobutane-type photoadducts could be isolated from acetone solution of uridines [9] and of 2'-deoxyuridines [10] containing 2,3-dimethyl-2-butene by uv-irradiation.

In this communication, we would like to report the photoadditions of cytosine (**1**), 2'-deoxycytidine (**2a**) and its *N*-acetyl-3',5'-di-*O*-acetyl derivative (**2b**) to 2,3-dimethyl-2-butene, respectively.

A degassed solution of 6 mM of **1** and 60 mM of 2,3-dimethyl-2-butene in acetone containing 5% of water was irradiated for 36 h with 400-W high pressure mercury arc through a Pyrex filter. This set-up allowed lines longer than 304 nm of the arc to be absorbed. After removal of the solvent, the residue was flash-chromato-

graphed (silica gel, eluent: chloroform-ethanol 9:4) to give the 1:1 adduct (**3**) [11] in 61% yield. The structure of **3** was confirmed by means of nmr, FAB-ms and elemental analysis.

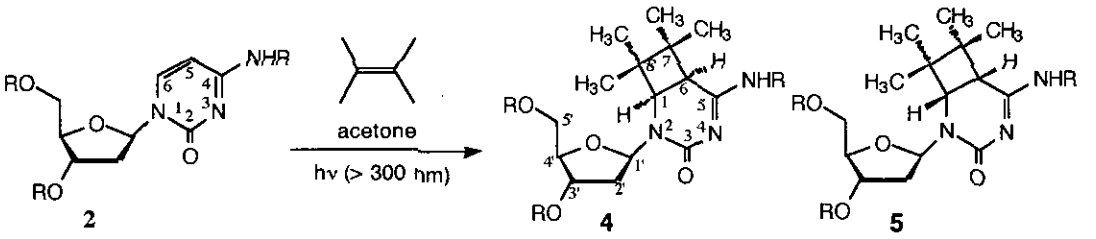
Scheme 1



1 **3**

An acetone solution of 3 mM of **2b** (R = Ac) and 30 mM of 2,3-dimethyl-2-butene was irradiated in a similar manner. One and a half hours' irradiation converted the starting material into two products. They were isolated by repeated chromatography. [12] FAB-ms showed their molecular weights as 437. Nmr spectra showed the absence of aromatic protons at positions 5 and 6 of **2b**, and instead, the presence of two aliphatic protons at higher field and four methyl groups. This unambiguously indicates that these two products are 1:1 adducts (**4b** and **5b**) of **2b** to 2,3-dimethyl-2-butene which possess a cyclobutane ring (Scheme 2).

Scheme 2



2 **4** **5**

Treatment of **4b** (R = Ac) and **5b** (R = Ac) with sodium methoxide gave **4a** (R = H) and **5a** (R = H) as colorless amorphous solids in quantitative yields. [13] Their structures were unambiguously confirmed by means of high resolution ms and nmr. Unfortunately, we could not obtain crystals of **4a** and **5a** suitable for X-ray crystallographic analysis. So, their absolute configurations at positions 1 and 6 were determined by converting them into the photoproducts (**6**) and (**7**) of 2'-deoxyuridine with 2,3-dimethyl-2-butene by treatment with aqueous NaOH, [14] whose absolute configurations have already been established (Scheme 3). [10] In this way, the upper component on tlc was turned out to be the (1*R*, 6*R*)-isomer (**4b**) and that of the lower one to be the (1*S*, 6*S*)-isomer (**5b**).

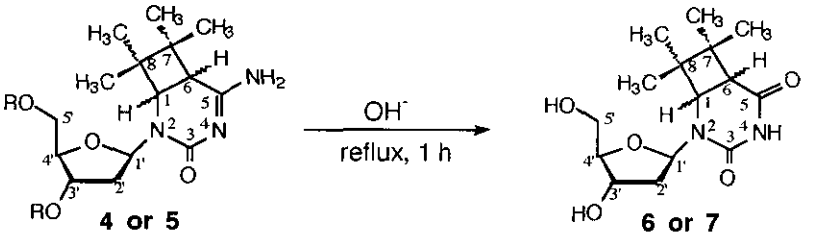
Irradiation of **2a** was performed in a similar manner, and the products (**4a**) and (**5a**) were isolated as their *N*-acetyl-3',5'-di-*O*-acetyl derivatives (**4b**) and (**5b**),[15] by treatment with acetic anhydride and pyridine (Table 1). Further work is required for the elucidation of the detailed mechanism of this reaction.

Table 1. Photoaddition of **2** to 2,3-Dimethyl-2-butene in Acetone [1)]

	R	time (h)	yield (%)	ratio of **4**:**5**
2a	H	10	75 [2)]	1.2:1.0
2b	Ac	1.5	80 [2)]	1.6:1.0

1) Substrate (1.5 mM) and 2,3-dimethyl-2-butene (10 mM) in acetone were used.
2) Isolated yield.

Scheme 3



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11. 5-Amino-7,7,8,8-tetramethyl-*cis*-2,4-diazabicyclo[4.2.0]oct-4-en-3-one (**3**): Recrystallized from methanol in colorless prisms, mp 258.5 - 259.5 °C. *Anal.* Calcd for $C_{10}H_{17}N_3O$: C 61.51; H 8.77; N 21.52. Found: C 61.22; H 8.86; N 21.52. FAB-ms (*m*-nitrobenzylalcohol) 196 (M^++1). ^{1}H-Nmr (400 MHz, Dimethylsulfoxide-$\underline{d}_6$) δ 6.8 - 8.2 (br s, 2H, 5-NH_2), 6.74 (br s, 1H, 2-NH), 3.44 (dd, 1H, *J* = 8.5, 7.2 Hz, 1-CH), 2.81 (d, *J* = 8.5 Hz, 6-CH), 1.05, 0.96, 0.88, 0.82 (each s, 3H, 7,7,8,8-CH_3's).

12. At least three chromatographic purifications (silica gel, ethyl acetate as the eluent) were required.

13. (1*R*, 6*R*)-5-Amino-7,7,8,8-tetramethyl-2-(β-D-deoxyribofuranosyl)-*cis*-2,4- diazabicyclo[4.2.0]oct-4-en-3-one (**4a**): High resolution FAB-ms (*m*-nitrobenzylalcohol, polyethylene glycol 400): M^++1 = 312.1955 (Calcd for $C_{15}H_{26}N_3O_4$ (M + 1) 312.1925). ^{1}H-Nmr (400 MHz, Dimethylsulfoxide-$\underline{d}_6$) δ 7.7 (br s, 1H, 5-NH), 7.1 (br s, 1H, 5-NH), 6.17 (dd, 1H, J = 8.0, 6.8 Hz, 1'-CH), 5.1 (br s, 1H, 3'-OH), 4.8 (br s, 1H, 5'-OH), 4.11 (dt, 1H, *J* = 4.0, 2.4 Hz, 3'-CH), 3.08 (d, 1H, *J* = 10.4 Hz, 1-CH), 3.55 (dt, 1H, *J* = 5.2, 2.4 Hz, 4'-CH), 3.42 (d, 2H, *J* = 5.2 Hz, 5',5"-CH), 2.73 (d, 1H, *J* = 10.4 Hz, 6-CH), 1.73 (ddd, 1H, *J* = 12.4, 8.0, 4.0 Hz, 2"-CH), 1.68 (ddd, 1H, *J* = 12.4, 6.8, 4.0 Hz, 2'-CH), 1.19, 0.95, 0.88, 0.77 (each s, 3H, 7,7,8,8-CH_3's). (1*S*, 6*S*)-5-Amino-7,7,8,8-tetramethyl-2-(β-D-deoxyribofuranosyl)-*cis*-2,4- diazabicyclo[4.2.0]oct-4-en-3-one (**5a**): High resolution FAB-ms (*m*-nitrobenzylalcohol, polyethylene glycol 400): M^++1 = 312.1894 (Calcd for $C_{15}H_{26}N_3O_4$ (M + 1) 312.1925). ^{1}H-Nmr (400 MHz, Dimethylsulfoxide-$\underline{d}_6$) δ 7.6 (br s, 1H, 5 -NH), 7.1 (br s, 1H, 5-NH), 6.03 (dd, 1H, *J* = 8.8, 5.6 Hz, 1'-CH), 5.0 (br s, 1H, 3'-OH), 4.7 (br s, 1H, 5'-OH), 4.00 (dt, 1H, *J* = 6.0, 2.4 Hz, 3'-CH), 3.87 (d, 1H, *J* = 9.6 Hz, 1-CH), 3.55 (dt, 1H, *J* = 10.8, 5.6 Hz, 5'-CH), 3.37 (dd, 1H, J = 10.8, 5.6 Hz, 5"-CH), 2.74 (d, 1H, *J* = 9.6 Hz, 6-CH), 1.97 (ddd, 1H, *J* = 10.8, 8.8, 6.0 Hz, 2"-CH), 1.62 (ddd, 1H, *J* = 10.8, 5.6, 2.4 Hz, 2'-CH), 1.18, 0.90, 0.87, 0.79 (each s, 3H, 7,7,8,8-CH_3's). Uv λ_{max} (MeCN) 231 nm (ε 9800).

14. As the general procedure, 45 mg of **5a** in 5 ml of 1 N NaOH was refluxed for 1 h and the product was purified by chromatography to gave **7** in 60% yield.

15. We could not clearly separate **4a** from **5a** by chromatography.

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