Photocycloaddition of Benzoylated 2′-Deoxyribonucleoside to 2,3-Dimethyl-2-butene

Shao Shun Li,[a] Xue Long Sun,[a] Haruo Ogura,*,[a] Yaeko Konda,[b] Tomomitu Sasaki,[b] Yumiko Toda,[b] Hiroaki Takayanagi,[b] and Yoshihiro Harigaya[b]

Shenyang Pharmaceutical University,[a] 103 Wenhua Road, Shenyang 110015, People's Republic of China and School of Pharmaceutical Sciences, Kitasato University,[b] 5–9–1, Shirokane, Minato-ku, Tokyo 108, Japan.

Received July 5, 1994; accepted August 16, 1994

The photochemical reactions of 3′,5′-dibenzoyl-2′-deoxyuridine and -thymidine with 2,3-dimethyl-2-butene by upon ultraviolet (UV) irradiation in acetone have been investigated. Each reactant gave a pair of diastereomers in good yield. After recrystallization, mixed crystals of diastereomers could be isolated and purified. An X-ray analysis of a mixed crystal of the thymidine derivative (3b) showed a pair of diastereomers (A and B) composing a unit cell.

Keywords deoxyribonucleoside; photocycloaddition; X-ray analysis; mixed crystal; *anti*-conformer

Photodimerization and photohydration of pyrimidine nucleosides in DNA are responsible for their high mutagenic potential.[1)] Many workers have reported photoreactions of pyrimidine and its nucleosides with simple alkenes.[2)] We considered that photochemical cycloaddition reactions of nucleosides with simple olefins might provide a route to a variety of bicyclic nucleosides, and it was anticipated that these compounds might be useful for biological studies, especially those involving interaction with repair enzymes.[3)]

Our recent work has revealed that a pair of cyclobutane derivatives could be isolated from photocycloaddition reactions of uridine and related compounds,[4)] 2′-deoxyribonucleosides[5,6)] with 2,3-dimethyl-2-butene. Interestingly, the uridine derivatives showed potent differentiation-inducing and growth-inhibitory activities towards HL-60 cells.[7)]

In the present paper, we describe the synthesis and X-ray crystal analysis of the photocycloadduct of a 2′-deoxyribonucleoside. Photoaddition was performed in acetone at room temperature by use of a 400 W high-pressure mercury lamp fitted with a Pyrex filter. The acetone-sensitized cycloaddition of 3′,5′-dibenzoyl-2′-deoxyuridine (**2a**) and -thymidine (**2b**) with 2,3-dimethyl-2-butene proceeded smoothly to form the corresponding cycloadduct (**3a**, **b**) in good yield.

The structures of these products were elucidated by NMR spectral analysis and elemental analysis. NMR data for both adducts are consistent with the cycloadduct structures (**3a**, **b**), with a diastereomer ratio of 1 : 1. The structure of 2-(2-deoxy-di-*O*-benzoyl-*β*-D-ribofuranosyl)-6,7,7,8,8-pentamethyl-*cis*-2,4-diazabicyclo[4.2.0]octane-3,5-dione (**3b**) was confirmed by NMR studies and X-ray analysis. The results of ^{1}H, ^{13}C, ^{1}H–^{1}H correlation spectroscopy (COSY), heteronuclear multiple bond connectivity (HMBC), heteronuclear multiple quantum coherence (HMQC), and nuclear Overhauser effect (NOE) experiments are summarized in Table I, and the values are consistent with the assigned 1 : 1 ratio of diastereomers (A and B).

NOEs (Fig. 1) were observed between H-1 and H_A-15, H_3-9 for both diastereomers A and B. This implied that the heterocyclic moiety should have *anti* conformation in chloroform solution. Furthermore, NOE was observed between H_3-12 and H_A-15 for A, and between H_3-12 and H_2-18 for B. Therefore the structures of the diastereomers (A and B) could be confirmed.

The structure, including stereochemistry, was definitely established by means of X-ray crystal structure analysis. A perspective drawing of **3b** is shown in Fig. 2 on the basis of the absolute configuration of D-ribose. One pair of (1*S*,6*R*)-diastereomer (A) and (1*R*,6*S*)-diastereomer (B) composed a unit cell, and A shows high-*anti* and B shows *anti* conformation[8)] as judged from the torsion angles (Table II).

In the crystal structure, the 2,4-diazabicyclo[4.2.0]-octane-3,5-dione ring moiety appears to occupy the *anti* position, as in the solution. This is consistent with data

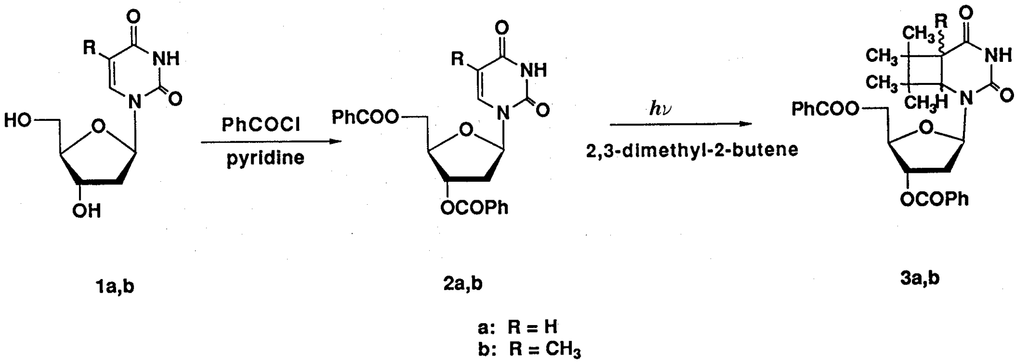
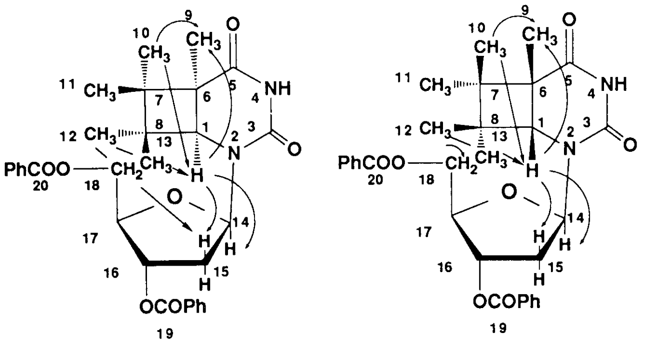


Chart 1

Table I. NMR Data for Diastereomers A[a] and B[a]

No.	δ_C	Correlated H[b] δ_H	C coupled with H[c]	H coupled with H[d]
Diastereomer A				
C-1	59.54 d	H-1 3.52 s	C-3, C-5, C-6, C-8, C-12, C-14	
N-2				
C-3	152.43 s			
N-4				
C-5	173.89 s			
C-6	47.41 s			
C-7	42.13 s			
C-8	44.39 s			
C-9	19.45 q	H_3-9 1.17 s	C-5, C-6, C-7	
C-10	21.04 q	H_3-10 0.94 s	C-6, C-7, C-8, C-11	
C-11	22.13 q	H_3-11 0.81 s	C-6, C-7, C-8, C-10	
C-12	23.86 q	H_3-12 0.89 s	C-7, C-8, C-13	
C-13	19.60 q	H_3-13 0.95 s	C-7, C-8, C-12	
C-14	83.32 d	H-14 6.38 dd (10.0, 5.0)		$H_{A,B}$-15
C-15	35.79 t	H_A-15 2.15 ddd (13.0, 10.0, 6.5)	C-14	H-16, H-14
		H_B-15 2.28 ddd (13.0, 5.0, 2.0)	C-16, C-17	H-16, H-14
C-16	74.85 d	H-16 5.58 m	C-14, C-18	$H_{A,B}$-15, H-17
C-17	80.64 d	H-17 4.35 ddd (5.5, 3.5, 2.8)	C-18	H-16, H_2-18
C-18	63.82 t	H_A-18 4.56 dd (11.5, 5.5)	C-16, C-17, C-20	H-17, H_B-18
		H_B-18 4.81 dd (11.5, 3.5)	C-16, C-17, C-20	H-17, H_A-18
C-19	166.08 s			
C-20	165.96 s or 165.93 s			
Diastereomer B				
C-1	59.85 d	H-1 3.44 s	C-3, C-5, C-6, C-8, C-12, C-14	
N-2				
C-3	151.41 s			
N-4				
C-5	173.78 s			
C-6	47.06 s			
C-7	42.21 s			
C-8	45.32 s			
C-9	19.76 q	H-9 1.13 s	C-5, C-6, C-7	
C-10	21.27 q	H_3-10 0.93 s	C-6, C-7, C-8, C-11	
C-11	22.08 q	H_3-11 0.79 s	C-6, C-7, C-8, C-12	
C-12	23.17 q	H_3-12 0.85 s	C-7, C-8, C-13	
C-13	19.30 q	H_3-13 0.99 s	C-7, C-8, C-12	
C-14	84.25 d	H-14 6.38 dd (9.0, 5.0)	C-1, C-3	$H_{A,B}$-15
C-15	35.64 t	H_A-15 2.25 ddd (13.8, 9.0, 7.0)	C-14	H-14, H-16
		H_B-15 2.45 ddd (13.8, 5.0, 1.3)	C-16, C-17	H-14
C-16	74.94 d	H-16 5.57 m	C-14, C-18, C-19	H_A-15, H-17
C-17	81.27 d	H-17 4.41 dd (6.0, 3.5)		
C-18	64.65 t	H_A-18 4.54 dd (12.0, 6.0)	C-16, C-17, C-20	H-17, H_B-18
		H_B-18 4.68 dd (12.0, 3.5)	C-16, C-17	H-17, H_A-18
C-19	166.08 s			
C-20	165.93 s or 165.96 s			

[a] Spectra were taken in $CDCl_3$. [b] HMQC. [c] HMBC. [d] ^1H–^1H COSY.



A (1*S*,6*R*)-diastereomer B (1*R*,6*S*)-diastereomer

Fig. 1

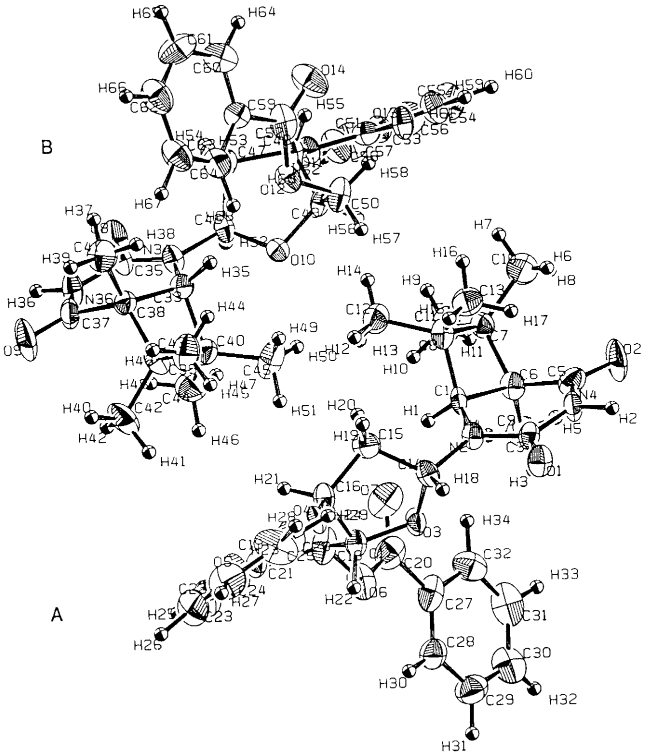


Fig. 2. A Perspective View of Diastereomers A and B in a Unit Cell

Table II. Selected Torsion Angles (°) of Diastereomers A and B

Diastereomer A	O3–C14–N2–C1	67 (1)
	O3–C14–N2–C3	−108.4 (9)
	C15–C14–N2–C1	−50 (1)
	C15–C14–N2–C3	134.5 (9)
Diastereomer B	O10–C46–N34–C33	19 (1)
	O10–C46–N34–C35	−159.0 (8)
	C47–C46–N34–C33	−99 (1)
	C47–C46–N34–C35	83 (1)

for 2-(β-D-2-deoxyribofuranosyl)-*cis*-2,4-diazabicyclo[4.2.0]octane-3,5-dione derivatives[5,6] and 5-amino-2-(β-D-2-deoxyribo-furanosyl)-*cis*-2,4-diazabicyclo[4.2.0]-octan-3-one derivatives,[5] and quite different from ribofuranosyl derivatives (*syn* conformation).[4]

Experimental

General Melting points were determined in a capillary tube and are uncorrected. NMR spectra were recorded on a Varian LX-400 spectrometer with Me_4Si (TMS) as an internal standard in $CDCl_3$. Microanalyses were performed by the members of the Shenyang group. The cell dimensions and diffraction intensities were measured on a Rigaku four-circle diffractometer, using graphite-monochromated CuK_α radiation (performed by the Kitasato group). Column chromatography was performed on Silica gel 60 (70—230 mesh, Merck).

1-(2-Deoxy-3,5-di-*O*-benzoyl-D-ribofuranosyl)uracil (2a) A mixture of 502.2 mg (2.2 mmol) of 2′-deoxyuridine (**1a**) and 846.3 mg (3 eq) of benzoyl chloride in 15 ml of pyridine was stirred at 50—55 °C for 3 d. After the reaction, it was cooled and treated with 50 ml of ice water. After refrigeration, precipitated crystals were collected by filtration. Recrystallization from ethanol–ethyl acetate gave 813.2 mg (91%) of **2a** as colorless crystals, mp 228—230 °C.

1-(2-Deoxy-3,5-di-*O*-benzoyl-*β*-D-ribofuranosyl)thymine (2b) In a similar manner to that described for **2a**, thymidine (**1b**) was benzoylated to give **2b** in 95% yield. Recrystallization from ethanol–ethyl acetate gave colorless crystals, mp 192—194 °C (reported[9]) mp 192.5—193.5 °C).

(1*R*,6*S*)- and (1*S*,6*R*)-2-(2-Deoxy-3,5-di-*O*-benzoyl-*β*-D-ribofuranosyl)-7,7,8,8-tetramethyl-*cis*-2,4-diazabicyclo[4.2.0]octane-3,5-dione (3a) A solution of **2a** (107.2 mg; 0.24 mmol) and 2,3-dimethyl-2-butene (235.3 mg; 10 eq) in acetone (500 ml) was irradiated with 400 W high-pressure mercury lamp through a Pyrex filter under a nitrogen atmosphere, until the reaction was completed (**2a** was no longer detectable by TLC). After removal of the solvent under reduced pressure, the residue was subjected to silica gel column chromatography with $CHCl_3$–MeOH (40 : 1) to afford 123.6 mg (96.7%) of **3a**. From the ^{1}H-NMR spectra, crude **3a** appears to be a 2 : 1 mixture of two cyclobutane photoproducts, which could not be further separated. Recrystallization from ethanol gave 74.6 mg of mixed crystals, mp 174—176 °C with 1 : 1 diastereomer ratio as judged from the NMR spectra. Unfortunately, a crystal suitable for X-ray diffraction study could not be obtained. *Anal.* Calcd for $C_{29}H_{32}N_2O_7$: C, 66.91; H, 6.20; N, 5.38. Found: C, 67.34; H, 6.38; N, 5.03.

(1*R*,6*S*)- and (1*S*,6*R*)-2-(2-Deoxy-3,5-di-*O*-benzoyl-*β*-D-ribofuranosyl)-6,7,7,8,8-pentamethyl-*cis*-2,4-diazabicyclo[4.2.0]octane-3,5-dione (3b) A solution of **2b** (99.0 mg; 0.22 mmol) and 2,3-dimethyl-2-butene (10 eq) in acetone was irradiated in the same manner as cited above. Purification by column chromatography afforded 107.9 mg (91.5%) of **3b**. From the ^{1}H-NMR spectrum, crude **3b** appeared to be a 5 : 1 mixture of two diastereomers, which could not be further separated. Recrystallization from methanol gave 12.1 mg of mixed crystals, mp 175—176 °C, corresponding to a 1 : 1 mixture of diastereomers from the NMR spectra (Table I and Fig. 1). *Anal.* Calcd for $C_{30}H_{34}N_2O_7$: C, 67.40; H, 6.41; N, 5.24. Found: C, 67.13; H, 6.38; N, 5.21.

X-Ray Crystallographic Analysis of 3b Data Collection: A crystal (colorless plate) of **3b** $C_{30}H_{34}N_2O_7$ having approximate dimensions of $0.10 \times 0.20 \times 0.10$ mm was used for the analysis. The cell dimensions and diffraction intensities were measured on a Rigaku AFC-5R four-circle diffractometer, with graphite monochromated CuK_α radiation ($\lambda = 1.5479$ Å). Cell constants and an orientation matrix for data collection, obtained from a least-squares refinement using the setting angles of 24 carefully centered reflections in the range of $43.58 < 2\theta < 48.44°$, corresponded to a triclinic cell with the following dimensions: $a = 11.296(1)$ Å, $b = 11.594(1)$ Å, $c = 10.7808(8)$ Å, $\alpha = 95.869(7)°$, $\beta = 92.450(8)°$, $\gamma = 104.318(8)°$, $V = 1357.5\ (2)$ Å.

For $Z = 2$ and F.W. = 534.61, the calculated density is 1.308 g/cm^3. Based on packing considerations, a statistical analysis of intensity distribution, and the successful solution and refinement of the structure, the space group was determined to be P_1.

The data were collected at a temperature of 23 ± 1 °C using the $\omega - 2\theta$ scan technique. Scans of $(0.84 + 0.30 \tan\theta)°$ were made at a speed of 8.0°/min (in omega).

Data Reduction: Of the 5227 reflections which were collected, 4946 were unique ($R_{\text{int}} = 0.095$). The intensities of three representative reflections which were measured after every 150 reflections remained constant throughout data collection, indicating crystal and electronic stability (no decay correction was applied). The linear absorption coefficient for CuK_α is 7.3 cm^{-1}. The data were corrected for Lorentz and polarization effects.

Structure Solution and Refinement: The structure was solved by direct methods.[10]) The non-hydrogen atoms were refined anisotropically. The final cycle of full-matrix least-squares refinement was based on 2585 observed reflections ($I > 3.00\sigma(I)$) and 700 variable parameters and converged with unweighted and weighted agreement factors of: $R = \sum \| F_o | - | F_c \| / \sum | F_o | = 0.052$ and $R_w = [(\sum w(| F_o | - | F_c |)^2 / \sum w F_o^2)]^{1/2} = 0.042$.

The maximum and minimum peaks on the final difference Fourier map corresponded to 0.18 and $-0.22\,e^-/\text{Å}^3$, respectively. Neutral atom scattering factors were taken from Cromer and Waber.[11]) Anomalous dispersion effects were included in F_{calc}[12]); the values for $\Delta f'$ and $\Delta f''$ were those of Cromer and Waber.[11]) All calculations were performed using the TEXSAN[13]) crystallographic software package of Molecular Structure Corporation.

References

1) S. Y. Wang, *Nature* (London), **190**, 690 (1961); A. A. Lamola, *Pure Appl. Chem.*, **34**, 281 (1973).
2) A. J. Wexler, J. S. Swenton, *J. Am. Chem. Soc.*, **98**, 1602 (1976); A. J. Wexler, J. A. Hyatt, P. W. Raynolds, C. Cottrell, J. S. Swenton, *ibid.*, **100**, 512 (1978); T. G. Savino, L. K. Chenard, J. S. Swenton, *Tetrahedron Lett.*, **24**, 4055 (1983); A. J. Wexler, R. J. Balchunis, J. S. Swenton, *J. Org. Chem.*, **49**, 2733 (1984); V. V. Kaminski, A. J. Wexler, R. J. Balchunis, J. S. Swenton, *ibid.*, **49**, 2738 (1984); C. Kaneko, N. Shimomura, *Tetrahedron Lett.*, **23**, 2571 (1982).
3) H. Kamiya, N. Murata, T. Murata, S. Iwai, A. Matsukage, C. Masutani, F. Hanaoka, E. Ohtsuka, *Nucleic Acids Res.*, **21**, 2355 (1993).
4) I. Ishikawa, T. Itoh, H. Takayanagi, J. Oshima, N. Kawahara, Y. Mizuno, H. Ogura, *Chem. Pharm. Bull.*, **39**, 1922 (1991).
5) N. Haga, I. Ishikawa, M. Kinumura, H. Takayanagi, H. Ogura, *Heterocycles*, **35**, 569 (1993); N. Haga, H. Ogura, *ibid.*, **36**, 1721 (1993).
6) N. Haga, I. Ishikawa, H. Takayanagi, H. Ogura, *Bull. Chem. Soc. Jpn.*, **67**, 728 (1994).
7) Y. Honma, T. Ikuta, T. Kasukabe, M. Hozumi, T. Itoh, H. Ogura, *Anticancer Res.*, **8**, 695 (1988).
8) M. Orozco, D. Velasco, E. I. Canela, R. Franco, *J. Am. Chem. Soc.*, **112**, 8221 (1990).
9) J. J. Fox, D. V. Praag, I. Wempen, I. L. Doerr, L. Cheong, J. E. Knoll, M. L. Eidinoff, A. Bendich, G. B. Brown, *J. Org. Chem.*, **81**, 178 (1959).
10) Structure solution methods: MITHRIL; C. J. Gilmore, *J. Appl. Cryst.*, **17**, 42—46, Univ. of Glasgow, Scotland (1984). DIRDIF; P. T. Beurskens, Technical Report 1984/1 Crystallography Laboratory, Toernooiveld, 6525 Ed Nijmegen Netherlands.
11) D. T. Cromer, J. T. Waber, "International Tables for X-Ray Crystallography," Vol. IV, The Kynoch Press, Birmingham, 1974, p. 71.
12) J. A. Ibers, W. C. Hamilton, *Acta Crystallogr.*, **17**, 781 (1964).
13) TEXSAN–TEXRAY Structure Analysis Package, Molecular Structure Corporation, 1985.