DISTINCT SOLVENT-DEPENDENCE IN THE PHOTOREACTIONS OF PURINE NUCLEOSIDES WITH PYRIMIDO[5,4-*g*]PTERIDINETETRONE *N*-OXIDE: POSSIBLE GENERATION OF HYDROXYL RADICAL FROM THE EXCITED *N*-OXIDE IN ALCOHOLS †

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Abstract — Photoreaction of 2',3',5'-tri-*O*-acetyladenosine (**5**) with pyrimido[5,4-*g*]pteridinetetrone *N*-oxide (**1**) in acetonitrile gave N^6-cyanomethyl-2',3',5'-tri-*O*-acetyladenosine (**6**) as a major detectable product *via* coupling of adenosyl radical with cyanomethyl radical generated by the mediation of **1**. Under the analogous conditions, N^2-benzoyl-2',3',5'-tri-*O*-acetylguanosine (**8**) underwent oxidative degradation of the guanine skeleton by **1**. In sharp contrast, photoreactions of **5** and **8** with **1** in *tert*-butanol resulted in the formation of the corresponding 8-hydroxypurine nucleosides (**7**) and (**9**), respectively. These facts and other observations suggest that **1** could generate hydroxyl radical upon irradiation in alcohols.

In our search for the heterocyclic *N*-oxides possessing an efficient oxidation capacity under photochemical conditions, we have discovered that 1,3,6,8-tetra-*n*-butylpyrimido[5,4-*g*]pteridine-2,4,5,7(1*H*,3*H*,6*H*,8*H*)-tetrone 10-oxide (**1**) is exactly the case. The first synthesis and the structural characterization of **1** were achieved in 1972 by one of authors (Y. M.) together with

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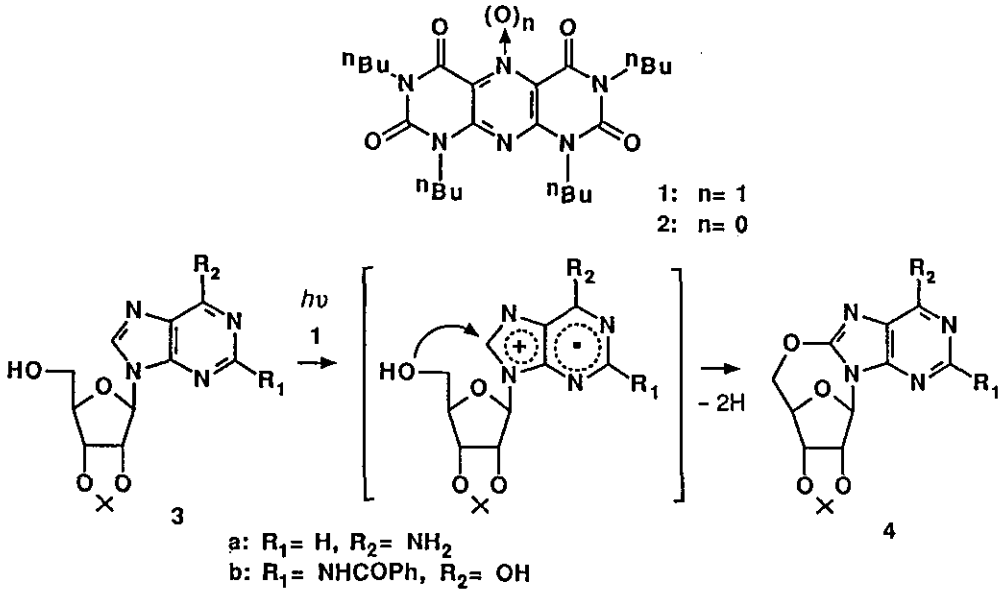
† This contribution is dedicated to Prof. Edward C. Taylor on the occasion of his 70th birthday.

Taylor and McKillop, [1] and our recent works [2,3] have proved its remarkable photochemical properties in aprotic solvents (*e.g.* acetonitrile). Thus, the *N*-oxide (**1**) functions efficiently under photochemical conditions as an electron-acceptor and as an agent for oxygenation or dehydrogenation depending upon the nature of the co-substrates. No occurrence of the appreciable intramolecular rearrangements of the *N*-oxide function during the photoreactions is particularly noticeable. [4]

Our previous work [3] has disclosed that the photoreaction of 5'-*O*-unprotected 2',3'-*O*-isopropylideneadenosine (**3a**) with **1** in acetonitrile affords the corresponding 5'-*O*,8-cycloadenosine (**4a**) possibly by an initial single-electron transfer (SET) from **3a** to **1** in the excited state followed by intramolecular trapping of transiently generated adenosine cation-radical by the 5'-hydroxyl group as illustrated in Scheme 1. This type of oxidative cyclization by **1** occurs more smoothly in the case of 2',3'-*O*-isopropylideneguanosine derivative (**3b**) which possesses a lower oxidation potential than that of **3a**. [5]

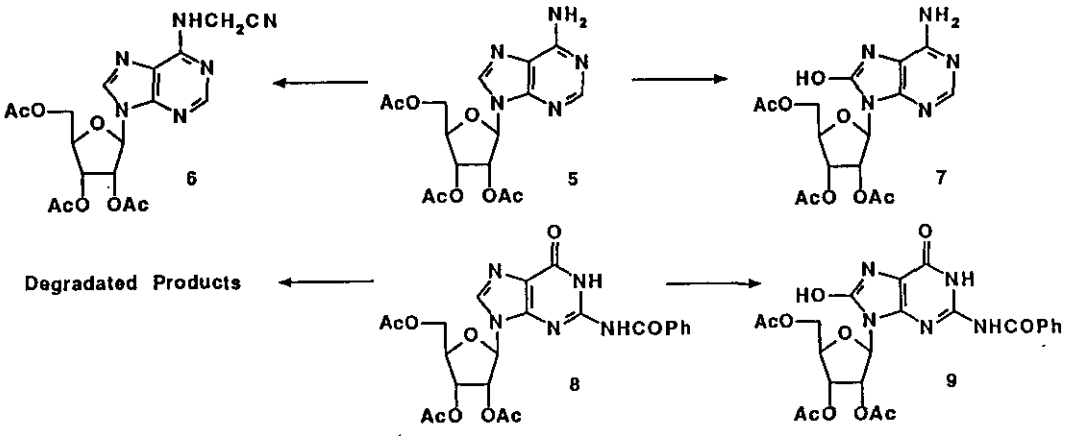


Scheme 1

Our initial objectives of the present work are based on the following expectations:

1) if 5'-*O*-protected purine nucleosides (cf. **5** and **8**) are employed in place of **3** as substrates, the corresponding 8-hydroxypurine nucleosides (cf. **7** and **9**) may be formed *via* the photochemical oxygen-atom transfer from **1**; 2) if alcohols in place of acetonitrile as a solvent are used, these

nucleosides (**5**) and (**8**) may undergo introduction of alkoxyl group at the C_8-position as a result of intermolecular trapping of the generated purinyl cation-radicals with the employed alcohols (cf. Schemes 1 and 2).



Scheme 2

Contrary to our expectations, however, we observed that the photoreaction of **5** with **1** in acetonitrile results in the formation of N^6-cyanomethyladenosine derivative (**6**) and that in *tert*-butanol gives 8-hydroxyadenosine derivative (**7**). Furthermore, it was found that the photoreaction of **8** with **1** in acetonitrile leads to the oxidative degradation of the guanine skeleton, whereas that in *tert*-butanol gives 8-hydroxyguanosine (**9**) (see Scheme 2). The photoreactions of **5** and **8** with **1** in *n*- or *sec*-butanol resulted in the oxidation of the alcohol employed rather than the oxygenation on the purine ring leading to **7** and **9**.

These facts have interesting mechanistic implications and suggest that **1** functions as an oxidant in a completely different manner depending upon the nature of solvents and generates hydroxyl radical in alcohols under the photochemical conditions.

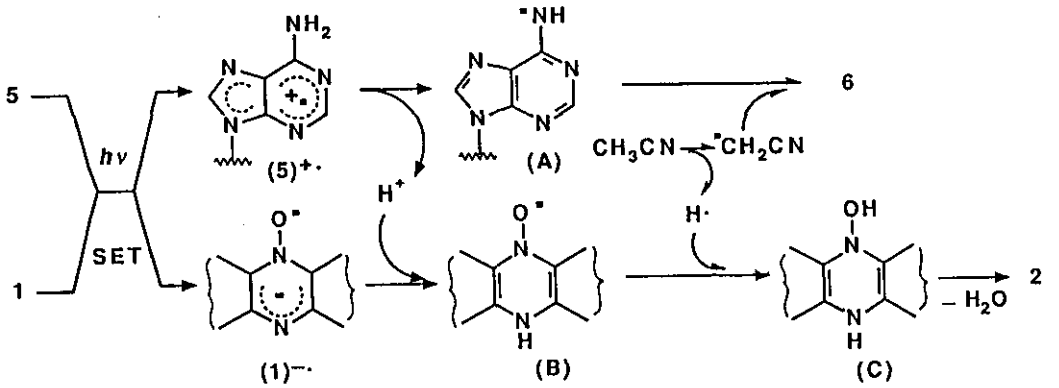
In this paper, we describe these new observations in the photochemical reactivities of **1** and their mechanistic aspects, which are unique for the photochemistry of heterocyclic *N*-oxides.

(1) Photoreactions of the 5'-*O*-Protected Adenosine and Guanosine Derivatives (5) and (8) with the *N*-Oxide (1) in Acetonitrile

A mixture of **5** [1.0 mmol] and **1** [2.0 mmol] was irradiated externally in dry acetonitrile with uv-visible light (>355 nm) at ambient temperature under Argon for 4 h. During this period, 55% of **5**

and 78% of **1** were consumed and tlc-densitometric analyses showed the formation of two products and no formation of the anticipated 8-hydroxyadenosine derivative (**7**) [6] in this reaction. Careful column chromatographic separation of the reaction mixture allowed isolation of N^6-cyanomethyl-2',3',5'-tri-*O*-acetyladenosine (**6**)[10% yield based on the employed **5**] together with deoxygenated pyrimido[5,4-*g*]pteridinetetrone (**2**). The structure of **6** was confirmed by spectral comparison with the sample prepared independently by condensation of 6-chloro-9-(2',3',5'-tri-*O*-acetylribofuranos-1'-yl)purine [7] with aminoacetonitrile.

A conceivable reaction sequence for the formation of **6** is outlined as depicted in Scheme 3. As demonstrated in previous works,[2,3] **1** effectively acts as an electron acceptor under the photochemical condition. Thus, the SET from **5** (E^{ox}_p= 1.65V *vs.* SCE) to **1** ($E^{red}_{1/2}$= - 0.97 V *vs.* SCE) in the excited state generates a pair of cation-radical $(\mathbf{5})^{+\cdot}$ and anion-radical $(\mathbf{1})^{-\cdot}$.[8] In contrast to the case of **3a**, the absence of the 5'-hydroxyl group in the jaxtaposition for intramolecular trapping of the transient $(\mathbf{5})^{+\cdot}$ causes the proton-transfer from $(\mathbf{5})^{+\cdot}$ to $(\mathbf{1})^{-\cdot}$ to give adenosyl radical (**A**) and nitroxyl radical (**B**). Without the occurrence of the anticipated radical coupling between **A** and **B**, [9] the nitroxyl radical (**B**) abstracts a hydrogen from acetonitrile employed as a solvent to generate cyanomethyl radical, [10] which possesses the ability to couple with **A** to give the ultimate product (**6**). The *N*-oxide (**1**) can be deoxygenated to produce **2** *via* dehydration of the resulting dihydro intermediate (**C**). The above results provide valuable informations on the chemical reactivities of adenosyl radical species $(\mathbf{5})^{+\cdot}$ and (**A**).



Scheme 3

Under the analogous conditions, the photoreaction of the 5'-*O*-protected guanosine derivative (**8**) with **1** gave a complex mixture of products which arises from degradation of the guanine ring as evidenced by tlc analysis, indicating that the guanosine cation-radical $(\mathbf{8})^{+\cdot}$ has chemical reactivities different from that of the adenosine cation-radical $(\mathbf{5})^{+\cdot}$ as previously demonstrated.[11] No detection of 8-hydroxy derivative (**9**) in this reaction mixture is worthwhile to note in agreement with the case of **5**.

(2) Photoreactions of the Adenosine and Guanosine Derivatives (5) and (8) with the *N*-Oxide (1) in Butanols

Figure 1 shows the stability of **1** in butanols, i.e.,*n*-, *sec*-, and *tert*-butanols, on irradiation with uv-visible light. The consumption rates of the *N*-oxide (**1**) were in the order of *sec*-butanol > *n*-butanol >> *tert*-butanol.

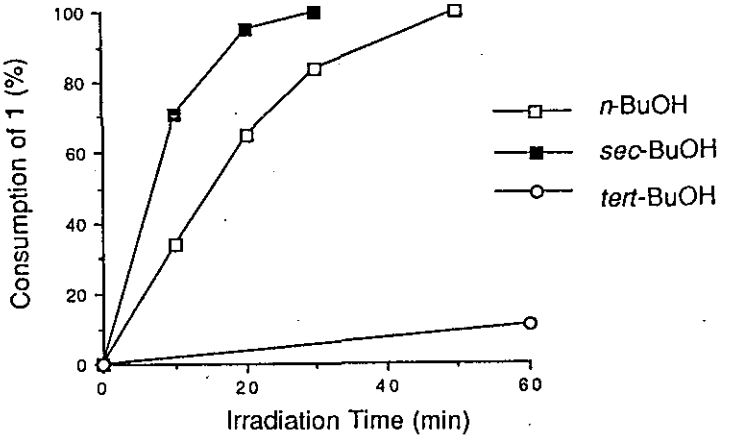


Figure 1. Stability of the *N*-Oxide (**1**) in Butanols on Irradiation

In the case of *sec*-butanol, tlc and gc analyses of the reaction mixture after irradiation for 45 min indicated the almost quantitative conversion of **1** into **2** and the formation of 2-butanone in 49% yield, indicating the occurrence of the photochemical oxidation of *sec*-butanol by **1** under the conditions employed. Analogous results were obtained in the photolysis of **1** in *n*-butanol . On the other hand, the *N*-oxide (**1**) was fairly stable under irradiation in *tert*-butanol and a prolonged irradiation time (e.g. for 5 h) caused the gradual formation of **2** and acetone which was identified as its 2,4-dinitrophenylhydrazone.

Thus, *tert*-butanol was chosen as a solvent for examination of intermolecular trapping of the generated purinyl radical species [cf. $(5)^{+\cdot}$] along our initial objectives.

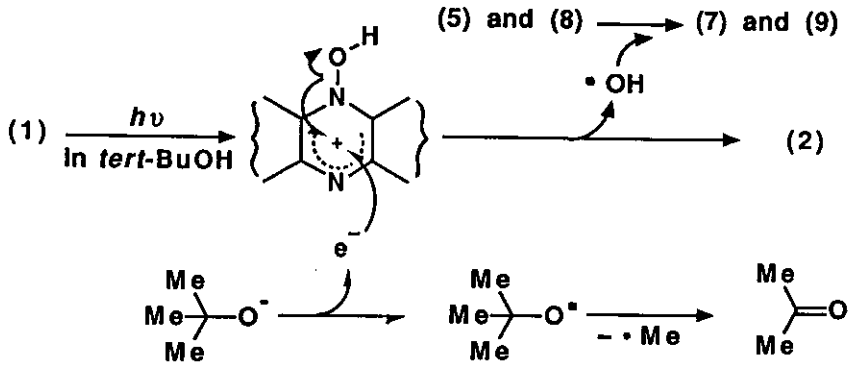
A mixture of **5** [1.0 mmol] and **1** [2.0 mmol] in freshly distilled *tert*-butanol was irradiated externally under the conditions analogous to those of the foregoing case. Tlc analysis of the reaction mixture after irradiation for 3 h showed that 23% of **5** was consumed together with the formation of **2**. Column chromatographic separation allowed isolation of the 8-hydroxyadenosine derivative (**7**) in 9% yield. The structure of **7** was fully supported by spectral comparison with the authentic sample prepared independently according to the general procedure for the C_8-hydroxylation of purine nucleosides.[12,13] Contrary to our expectation, the 8-*tert*-butoxy substituted adenosine derivative was not obtained in a detectable amount.

Analogously, the photoreaction of **8** with **1** in *tert*-butanol led to the formation of the corresponding 8-hydroxyguanosine derivative (**9**) in 14% yield.

In order to inspect the origin of the 8-hydroxyl oxygen in **7**, the reaction of **5** in *tert*-butanol was carried out upon employment of ^{18}O-labelled **1** possessing ^{18}O in the *N*-oxide function.[2] Quantitative incorporation of ^{18}O into the 8-hydroxyl group of the product (**7**) was confirmed by mass spectrometry, indicating clearly that the 8-hydroxyl oxygen in **7** originates from the *N*-oxide oxygen in **1**.

As discussed in section (1), the formation of the N^6-cyanomethyladenosine derivative (**6**) in the photoreaction of **5** with **1** provided a negative proof for the coupling of the adenosyl radical species $(5)^{+\cdot}$ or (**A**) with the nitroxyl radical species $(1)^{-\cdot}$ or (**B**) leading to **7**. Extensive studies on the reactions of adenosine and guanosine derivatives with hydroxyl radical have demonstrated the occurrence of C_8-hydroxylation of the purine ring as one of various oxidation pathways.[11,14] Crucial evidence for supporting the generation of hydroxyl radical in the photolysis of **1** in butanols was not obtained by the spin-trapping method using 5,5-dimethylpyrrolidine *N*-oxide (DMPO) and by Babbs' colorimetric method.[15] However, complete inhibition of the formation of the 8-hydroxyadenosine (**7**) in the photoreaction of **5** with **1** in *tert*-butanol by the addition of DMSO, a hydroxyl radical scavenger, is quite suggestive for the generation of hydroxyl radical during the reaction.

Thus, the present photochemical formation of **7** and **9** from **5** and **8** in *tert*-butanol is likely rationalized by a reaction sequence as depicted in Scheme 4.



Scheme 4

The generation of hydroxyl radical in *tert*-butanol involves the solvation of **1** in the excited state, a SET from *tert*-butoxy anion to the protonated form, and homolytic cleavage of the *N-O* bond in the resulting radical intermediate. The formation of a significant amount of acetone supports the intermediary formation of *tert*-butoxy radical which collapse to give acetone and methyl radical.[16] In an analogous manner, the photoreactions of **1** in *n*- and *sec*-butanols appear to cause the generation of hydroxyl radical. In these cases, however, the generated hydroxyl radical quite effectively abstracts a hydrogen from the α-position of *n*- and *sec*-butanols to give the corresponding carbonyl compounds even though **5** or **8** coexists. In fact, no formation of **7** and **9** in the photoreactions of **5** and **8** with **1** in *n*- and *sec*-butanols was observed.

In conclusion, the present work demonstrated the significant solvent dependence in the photochemical reactions of the novel *N*-oxide (**1**) with the purine nucleosides and , in particular, indicated the possible generation of hydroxyl radical in the photolysis of **1** in alcohols.

These facts show diversity of photochemical properties of **1** as an oxidant and prompt us to investigate the synthesis of water-soluble analogs of **1** and their utilization as an efficient hydroxyl radical generator by the photolysis in aqueous solution. Along this line, our studies are now in progress.

EXPERIMENTALS

All irradiations were carried out with a Riko Rotary Photochemical Reactor (400W high-pressure mercury arc lamp, Riko Kagaku Sangyo) through a $BiCl_3$ solution filter (>355 nm) under Argon at ambient temperature. The spectroscopic measurements were performed with the following instruments: Uv absorption spectra with a Shimadzu-260 spectrophotometer; ^{1}H-nmr spectra with JEOL JNX-270 spectrometer with tetramethylsilane as internal standard and J-values in Hz; mass spectra with a JEOL JMS D-300 machine operating at 70 eV. Polarographic analyses were carried out at ambient temperature under Argon with a Yanaco Polarographic Analyzer P-1100 using tetra-*n*-butylammonium perchlorate as a supporting electrolyte and dry acetonitrile as a solvent. Tlc analyses were performed on silica gel plates (Merck, art 5715) with benzene-ethyl acetate (5 : 2) for the assay of pyrimido[5,4-*g*]pteridinetetrone derivatives; methylene chloride-ethyl acetate (2 : 3) and chloroform-methanol (5 : 1) for the assay of adenosines; benzene-ethyl acetate (3 : 7) for the assay of guanosines as developer. Tlc-scanning was carried out with a Shimadzu CS-9000 dual-wavelength flying-spot scanner (detector: 360 nm for the assay of the pyrimido[5,4-*g*]pteridine derivatives; 260 nm for the assay of the nucleosides. Column chromatographic separation was accomplished on silica gel (Wakogel *C*-300).

Materials. —— 1,3,6,8-Tetra-*n*-butylpyrimido[5,4-*g*]pteridine-2,4,5,7(1*H*,3*H*,6*H*,8*H*)-tetrone 10-oxide (**1**),[1] 1,3,6,8-tetra-*n*-butylpyrimido[5,4-*g*]pteridine-2,4,5,7(1*H*,3*H*,6*H*,8*H*)-tetrone (**2**),[17] 2',3',5'-tri-*O*-acetyladenosine (**5**),[18] N^2-benzoyl-2',3',5'-tri-*O*-acetylguanosine (**8**)[19] were prepared according to the known methods, respectively.

Stability of the *N*-Oxide (1) in Butanols on Irradiation. ——

A solution of **1** (9.8 mg, 2.0×10^{-5} mol) in *n*-butanol (10 ml) was irradiated externally. The reaction mixture was sampled every 10 min for 50 min. The consumptions of **1** during the photolysis [34% (after 10 min), 65% (20 min), 84% (30 min), and 100% (50 min)] were estimated by tlc densitometry and plotted in Figure 1 as a function of irradiation time. Tlc analyses of the reaction mixtures showed almost quantitative conversion of the consumed **1** (R_f 0.27) into **2** (R_f 0.35), respectively. Treatment of the reaction mixture with 2,4-dinitrophenylhydrazine under the

conditions for trapping carbonyl compounds followed by usual work-up allowed isolation of the corresponding hydrazone and indicated the formation of *n*-butyraldehyde in 37% yield after irradiation for 1 h. The structure of the resulting hydrazone was confirmed by spectral comparison with the authentic sample.

Analogous results were obtained in the photolysis of **1** (9.8 mg, 2.0×10^{-5} mol) in *sec*-butanol (10 ml). The consumption yields of **1** were as follows: 71% (after 10 min), 95% (20 min), and 100% (30 min). The formation of 2-butanone (49% after 45 min) in this reaction was shown by gc and gc-mass analyses of the reaction mixture.

Under analogous conditions, the photolysis of **1** in *tert*-butanol was carried out. Tlc analysis of the mixture after 1 h showed only 11% conversion of **1** into **2**. The consumption of **1**, however, was significantly accelerated by the addition of an equimolar amount of the adenosine (**5**) [consumption yield of **1** after 1 h: 24%]. The formation of acetone (33% after 5 h) in this reaction was confirmed by the trapping with 2,4-dinitrophenylhydrazine.

Photochemical Reactions of the Adenosine Derivative (5) with 1. ——

(a) in Acetonitrile. A solution of **5** (78.7 mg, 2.0×10^{-4} mol) and **1** (195.4 mg, 4.0×10^{-4} mol) in dry acetonitrile (200 ml) was irradiated externally for 4 h. Tlc analyses of the reaction mixture showed consumptions of 55% of **5** (R_f 0.13; methylene chloride-ethyl acetate) and 78% of **1** and the formation of a less polar product (R_f 0.35). The consumed **1** was converted into **2** almost quantitatively. After removal of the solvent under reduced pressure, the residue was subjected to column chromatography eluted with methylene chloride-ethyl acetate (5 : 2) to isolate the product (8.6 mg, 10%) which was N^6-cyanomethyl-2',3',5'-tri-*O*-acetyladenosine (**6**) as a powder, mp 70-72 °C. The structure of the product (**6**) was assigned by microanalytical spectral data (Anal. Calcd for $C_{18}H_{20}N_6O_7$: C, 50.00; H, 4.66; N, 19.44. Found: C, 49.74; H, 4.68; N, 19.27); ms: *m/z* 432 (M^+, 2%), 373 (M^+ - OAc, 4%), 259 (29%), 175 (13%), 139 (62%), and 43 (100%); υ_{max} (KBr)/cm^{-1} 1749 (C=O) and 1619; λ_{max} (MeOH)/nm 261 and 209; δ_H ($CDCl_3$) 2.09 (3H, s, OAc), 2.12 (3H, s, OAc), 2.16 (3H, s, OAc), 4.3-4.5 (3H, m, 4'-H and 5'-H_2), 4.63 (2H, bd, collapsed to singlet by the addition of deuterium oxide, NHCH_2CN), 5.68 (1H, t, J 5.4, 3'-H), 5.95 (1H, t, J 5.4, 2'-H), 6.20 (1H, d, J 5.4, 1'-H), 7.15 (1H, bt, deuterium exchangeable N*H*CH_2CN), 8.04 (1H, s, 2- or 8-H), and 8.49 (1H, s, 8- or 2-H).

(b) in *tert*-Butanol. A mixture of **5** (19.7 mg, 5.0 x10^{-5} mol) and **1** (48.8 mg, 1.0 x 10^{-4} mol) in freshly distilled *tert*-butanol (50 ml) was irradiated under the conditions analogous to that of the forgoing case. Tlc analyses of the reaction mixture after 3 h showed the 23% consumption of **5** (R_f 0.55, chloroform-methanol) and 93% conversion of **1** into **2**, together with the formation of a polar product (R_f 0.45). Column chromatographic separation of the reaction mixture with chloroform-methanol (70 : 1) as eluent allowed isolation of the product (2.0 mg, 9%) which was 2',3',5'-tri-*O*-acetyl-8-hydroxyadenosine (**7**) [6] as a powder. The structure of the product (**7**) was assigned by microanalytical spectral data; ms: *m/z* 409 (M^+, 5%), 259 (30%), 151 (23%), 139 (46%), 97 (28%), and 43 (100%); υ_{max} (KBr)/cm^{-1} 3207 (NH), 1746 (C=O), and 1655; λ_{max} (MeOH)/nm 269, 255 (sh), and 217; δ_H ($CDCl_3$) 2.08 (3H, s, OAc), 2.11 (3H, s, OAc), 2.13 (3H, s, OAc), 4.2-4.6 (3H, m, 4'-H and 5'-H_2), 5.45 (2H, b, deuterium exchangeable NH_2), 5.86 (1H, t, J 5.8, 3'-H), 6.08 (1H, d, J 3.9, 1'-H), 6.18 (1H, dd, J 5.8 and 3.9, 2'-H), 8.19 (1H, s, 2-H), and 10.5 (1H, b, deuterium exchangeable N_7H).

The formation of **7** in this photoreaction was almost completely inhibited by the addition of DMSO at concentrations of 1 - 10% into the medium.

Photochemical Reactions of the Guanosine Derivative (8) with 1. ——

(a) in Acetonitrile. A solution of **8** (5.1 mg, 1.0 x 10^{-5} mol) and **1** (9.8 mg, 2.0 x 10^{-5} mol) in dry acetonitrile (10 ml) was irradiated externally under the conditions analogous to the case of **5**. Tlc analyses of the reaction mixture after 3 h showed the consumptions of 13% of **8** and 14% of **1** and no formation of uv-detectable products in this reaction, indicating the occurrence of the complex reactions of **8** with **1** including the oxidative degradation of guanine ring system under the conditions employed.

(b) in *tert*-Butanol. A solution of **8** (25.7 mg, 5.0 x 10^{-5} mol) and **1** (48.9 mg, 1.0 x 10^{-4} mol) in freshly distilled *tert*-butanol (50 ml) was irradiated under the analogous conditions described above. Tlc analyses of the reaction mixture showed the consumptions of 17% of **8** (R_f 0.09, benzene-ethyl acetate) and 82% of **1** and the formation of a less polar product (R_f 0.23). Column chromatographic separation [eluted with methylene chloride-ethyl acetate (1 : 1)] of the residue obtained after removal of the solvent allowed isolation of the product (4.0 mg, 14%) which was N^2-benzoyl-2',3',5'-tri-*O*-acetyl-8-hydroxyguanosine (**9**) as a powder. The structure of the

product (**9**) was assigned by microanalytical spectral data (Anal. Calcd for $C_{23}H_{23}N_5O_{10}\cdot$ 3/2 H_2O: C, 51.56; H, 5.11; N, 13.67. Found: C, 51.67; H, 4.89; N, 13.57); ms: *m/z* 528 (M^+ - H, 7%), 271 (20%), 259 (36%), 139 (63%), 105 (91%), and 43 (100%); υ_{max} (KBr)/cm^{-1} 3192 (NH), 1745 (C=O), and 1690; λ_{max} (MeOH)/ nm 317, 272, 235 (sh), and 219; δ_H ($CDCl_3$) 2.03 (3H, s, OAc), 2.12 (3H, s, OAc), 2.16 (3H, s, OAc), 4.3-4.6 (3H, m, 4'-H and 5'-H_2), 6.0-6.3 (3H, m, 1'-H, 2'-H, and 3'-H), 7.5-8.2 (5H, m, Ph-H), 9.22 (1H, b, deuterium exchangeable N_7H or N^2H), 9.48 (1H, b, deuterium exchangeable N^2H or N_7H), and 12.43 (1H, b, deuterium exchangeable N_1H).

Independent Synthesis of the N^6-Cyanomethyladenosine Derivative (6).—
A mixture of 6-chloro-9-(2',3',5'-tri-*O*-acetylribofuranos-1'-yl)purine [7] (550 mg, 1.3 x 10^{-3} mol) and aminoacetonitrile sulfate (158 mg, 7.5 x 10^{-4} mol) in dioxane (20 ml) containing 1N NaOH aqueous solution (1.5 ml) was heated at 90 °C for 2 days. After removal of the solvent under reduced pressure, the residual oil was poured into water and the solution was extracted with chloroform. The extract was washed with brine, dried over anhyd. sodium sulfate, and evaporated to dryness. The resulting residue was subjected to column chromatography eluted with chloroform-methanol (80 : 1) to isolate **6** (140 mg, 25%), together with the recovered starting material (250 mg, 45%). The product (**6**) was identical in every respects with that isolated in the photoreaction of **5** with **1**.

Independent Syntheses of the 8-Hydroxypurine Nucleosides (7) and (9).—
To a solution of the adenosine (**5**)(787 mg, 2.0 x 10^{-3} mol) in dioxane (30 ml) and 10% disodium hydrogen phosphate (30 ml), bromine (0.5 ml, 3.0 x 10^{-3} mol) was added dropwise. After stirring for 15 min at ambient temperature,[12] the reaction mixture was extracted with chloroform. The extract was washed with brine, dried over anhyd. sodium sulfate, and evaporated to dryness to give the corresponding 8-bromoadenosine derivative (930 mg, 98%) as a crude powder.
A solution of the 8-bromoadenosine (930 mg, 2.0 x 10^{-3} mol) in acetic acid (20 ml) containing sodium acetate (820 mg, 1.0 x 10^{-2} mol) was refluxed for 2.5 h.[13] After removal of the solvent under reduced pressure, the resulting residue was subjected to column chromatography with chloroform-methanol (60 : 1) as eluent to isolate the 8-hydroxyadenosine (**7**)(620 mg, 76%).
In a similar manner, the 8-hydroxyguanosine (**9**) was obtained in 31% yield starting from **8**.

These compounds (**7**) and (**9**) were identical with the photoproducts in the reactions of **5** and **8** with **1**, respectively.

Photoreaction of the Adenosine (5) with ^{18}O-Labelled *N*-Oxide (1) in *tert*-Butanol.——

A solution of **5** (39.3 mg, 1.0×10^{-4} mol) and ^{18}O-labelled *N*-oxide (**1**) [2] (97.7 mg, 2.0×10^{-4} mol) in *tert*-butanol (100 ml) was irradiated externally for 3 *h* under the conditions similar to that of the forgoing case. After column chromatographic separation of the reaction mixture, ^{18}O-content value of the oxidation product (**7**) was determined by mass spectrometry [^{18}O-content: 30% for **1** and **7**, respectively]

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REFERENCES AND NOTES

1. E.C. Taylor, Y. Maki, and A. McKillop, *J. Org. Chem.*, 1972, **37**, 1601 (cf. M. Sako, S. Ohara, K. Hirota, K. Kano, Y. Maki, and E.C. Taylor, *J. Org. Chem.*, 1991, **56**, 6302).
2. M. Sako, S. Ohara, K. Hirota, and Y. Maki, *J. Chem. Soc., Perkin Trans. I*, 1990, 3339 and preceding papers cited therein.
3. M. Sako, T. Makino, Y. Kitade, K. Hirota, and Y. Maki, *J. Chem. Soc., Perkin Trans. I*, 1992, 1801 and preceding papers cited therein.
4. Pertinent reviews for the photochemistry of heterocyclic *N*-oxides: A. Albini and M. Alpegiani, *Chem. Rev.*, **1984**, *84*, 43; A. Albini and S. Pietra, "Heterocyclic *N*-Oxides", CRC Press, Boca Raton, 1991, p. 200.
5. M. Sako, K. Shimada, K. Hirota, and Y. Maki, *J. Chem. Soc., Chem. Commun.*, 1986, 1704.
6. J. Hovinen, C. Glemarec, A. Sandstrom, C. Sund, and J. Chattopadhyaya, *Tetrahedron*, 1991, **47**, 4693.

7. J. Zemlika and J. Owens, "Nucleic Acid Chemistry" Part 2, ed. by L. B. Townsend and R. S. Tipson, John Wiley & Sons, New York, 1978, p. 612.
8. In general, any organic molecules with an $E^{ox}_{1/2}$ of 2.2V or less (*vs.* SCE) can be predicted to be susceptible to oxidation *via* a photo-induced SET process (cf. P. G. Gassman and K. J. Botorff, *J. Am. Chem. Soc.*, 1987, **109**, 7547 and references cited therein).
9. No occurrence of the radical coupling may be attributed to that both radicals, (**A**) and (**B**), are *N*- and *O*-centered radicals, respectively. (cf. For a review concerning purinyl radical species, see S. Steenken, *Chem. Rev.*, 1989, **86**, 503)
10. In the photochemical reactions using acetonitrile as a solvent, the generation of cyanomethyl radical in the presence of an oxidant is frequently observed (e.g., J.W. Bruno, T.J. Marks, and F.D. Lewis, *J. Am. Chem. Soc.*, 1981, **103**, 3608).
11. see, a reference cited in 8.
12. M. Ikehara, S. Uesugi, and M. Kaneko, *Chem. Commun.*, 1967, 17.
13. M. Ikehara, H. Tada, and K. Muneyama, *Chem. Pharm. Bull.*, 1965, **13**, 1140.
14. H. Kasai and S. Nishimura, *Gann*, 1984, **75**, 841.
15. According to Babbs' procedure (C. F. Babbs and M. J. Gale, *Anal. Biochem.* 1987, **163**, 67; M. G. Steiner and C. F. Babbs, *Arch. Biochem. Biophys.*, 1990, **278**, 478), we obtained an unidentified and unstable azosulfone derivative which shows the uv absorption band at 434 nm. The Babbs' azosulfone for the colorimetric assay of hydroxyl radical, however, exhibits the slightly different uv spectrum with λ_{max} at 420 nm. Thus, the data available at present does not provide a crucial proof for the generation of hydroxyl radical.
16. J. D. Bacha and J. K. Kochi, *J. Org. Chem.*, 1965, **30**, 3272.
17. M. Sako, S. Ohara, K. Shimada, K. Hirota, and Y. Maki, *J. Chem. Soc., Perkin Trans. I*, 1990, 863.
18. H. Brederek and A. Martini, *Chem. Ber.*, 1947, **80**, 401; P. A. Lyon and C. B. Reese, *J. Chem. Soc., Perkin Trans. I*, 1974, 2645.
19. R. J. Gregorie and T. Neilson, *Can. J. Chem.*, 1978, **56**, 487.

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