THE PREPARATION OF TRANS-PYRIMIDINE GLYCOLS BY NEAR-UV IRRADIATION*

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Summary: Near-UV irradiation of Thy, Me^{1}Thy, $Me_2^{1,3}$Thy, Ura, Me^{1}Ura, and $Me_2^{1,3}$Ura in the presence of hydrogen peroxide produced corresponding cis- and trans-glycols in ~20% yields. Similarly, Cyt, Me^{5}Cyt, and $Me_2^{4,5}$Cyt gave the corresponding glycols of Ura, Thy, and Thy, owing to facile deamination of the resultant Cyt glycols. Time sequence analysis of product-yields using ^{14}C-labeled pyrimidines revealed that the yields of a product are also contingent on its stability. Importantly, this approach provides a novel method for the direct synthesis of trans-glycols and for the preparation of certain radiation products in furthering our understanding of radiation chemistry and the biology of nucleic acids.

For quite some time, the possible biological implications have stimulated the study of the reactions of hydroxyl (HO·) and hydroperoxyl (HOO·) radicals with pyrimidine (Pyr) and purine derivatives (1). Such reactions have been regarded as particularly relevant to the understanding of the γ-radiation of nucleic acids in aerated aqueous solutions (2). Because of the complexities of radiation reactions, limitations by the volume and concentration of reaction solutions, and the instability of radiation products, the quantities of radiation products obtained are generally not sufficient to allow further research. This plight has impeded progress in the study of the radiation chemistry and biology of nucleic acids. However, we have taken advantage of the Milas reaction condition (3) to generate HO· and HOO· from hydrogen peroxide under the influence of near-UV light in order to prepare sufficient quantities of 5,6-dihydro-6-hydroperoxy-5-hydroxythymine ($ho^{5}ho_2^{6}$hThy, 6-TOOH), which has been characterized as the major radiation product of Thy (4) and possibly DNA, (5) and to synthesize cis and trans Pyr glycols. Previous preparations of Pyr glycols by $KMnO_4$ or OsO_4 oxidation of Pyr (6,7) or by the

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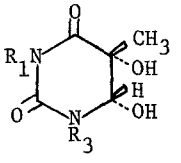
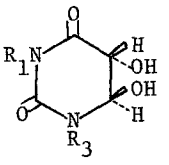
treatment of corresponding Pyr bromohydrins with Ag_2O yielded only cis isomers (8). Such a conversion has been shown (10) to be inefficient because, for example, cis Ura glycol dehydrates readily form isobarbituric acid (unpublished data). For Thy glycols, the trans isomers which can be detected from radiolysis are in negligible amounts. Therefore, a novel approach for the direct syntheses of trans Pyr glycols in reasonable yields, as reported in this communication, becomes highly desirable.

In a typical reaction, a 10 mM aqueous solution of a Pyr [Thy, $Me_2^{1,3}$Thy, Ura, Me^1Ura, or $Me_2^{1,3}$Ura] containing 40 mM of H_2O_2 was irradiated in a quartz tube with 313- or 360-nm light (11) for 24 hr. After irradiation, the excess



cis-Thy glycols



trans-Ura glycols

H_2O_2 and some organic peroxide formed were decomposed by treatment with 10% Pd-C. The solution was then concentrated at ∿35°C and applied to Whatman No. 3 paper or to Silica gel (Merck 60F-254) preparative tlc plates. After elution, the glycols were located by Finks reagent (12), extracted from the chromatograms with water, collected, and purified by rechromatography and/or crystallization from methanol-acetone as needed. The chromatographic and NMR spectral data are summarized in Table 1 and other data in Table 2.

Under similar conditions, Cyt, Me^5Cyt, and $Me_2^{4,5}$Cyt gave the corresponding cis and trans glycols of Ura, Thy, and Thy, respectively. Presumably, the formation of these glycols is the result of the deamination of the respective Cyt glycols because deamination usually occurs with 5,6-hCyt derivatives (13). However, these glycols were obtained in rather low yields, possibly due to the competitive formation of corresponding Cyt N-oxides.

Time sequence analyses of product-yields with ^{14}C-labeled compounds revealed that such yields are contingent on their stability. For instance,

Table 1. Chemical Shifts (in ppm) of NMR Spectra and R_f Values of Pyrimidine Glycols

	Uracil glycols ($ho_2^{5,6}hUra$)						Thymine glycols ($ho_2^{5,6}hThy$)					
	Ura[a]		Me^1Ura		$Me_2^{1,3}Ura$		Thy[b]		Me^1Thy		$Me_2^{1,3}Thy$	
	cis	trans	cis	trans	cis	trans	cis	trans	cis	trans	cis	trans
NMR Signal[c]												
H(5)	4.20 d,2[d]	3.66 d,2	4.27 q,5	3.70 d,2	4.32 d,5	3.86 d,5	-	-	-	-	-	-
H(6)	4.64 d,2	4.51 d,2	4.76 t,5	4.62 d,2	4.75 d,5	4.63 d,5	4.34 t,5	4.38 d,2	4.45	4.38	4.47	4.41
OH(5)	5.48	6.13 d,2	5.56 d,5	6.35 d,2	5.66 br	6.50	5.28	5.73	5.40	5.90	5.57 br	6.02 br
OH(6)	6.08	6.22 d,4	6.45 d,5	6.50 d,2	6.50 br	6.50 br	6.00 d,5	6.28 m	6.42 br	6.53 br	6.52 br	7.12 br
$CH_3(5)$	-	-	-	-	-	-	1.28	1.28	1.25	1.23	1.23	1.32
$N(1)CH_3$	-	-	2.88	2.90	2.98	2.98			2.89	2.88	2.95	2.98
$N(3)CH_3$	-	-	-	-	3.02	3.02	-	-	-	-	3.00	3.00
N(1)H	8.13	8.07	-	-	-	-	8.05 d,5	8.00 d,2	-	-	-	-
N(3)H	10.05 br	10.12	10.18 br	10.21 br			9.95	9.33	10.20	10.05	-	-
CHROMATOGRAPHY												
System[e]	paper,	A[e]	paper,	A	tlc,	B	paper,	C	paper,	A	tlc,	D
R_f	0.22	0.34	0.32	0.47	0.83	0.91	0.40	0.47	0.52	0.63	0.60	0.62

[a] *They are also the products of Cyt derivatives.*

[b] *They are also the products of Me^5Cyt derivatives.*

[c] *NMR spectra were taken with a 220 MHz spectrometer.*

[d] *Coupling constants in Hz.*

[e] *Eluents: A, 1-propanol-H_2O (10:3); B, methanol-acetone (3:1); C, 1-butanol saturated with water; D, ethyl acetate-2-propanol-water (75:16:9).*

Table 2. Vibrational Frequencies (in cm^{-1}) of IR Spectra, Mass Spectra, and Melting Points of Pyrimidine Glycols.

	Uracil glycols ($ho_2^{5,6}hUra$)						Thymine glycols ($ho_2^{5,6}hThy$)					
	Ura		Me^1Ura		$Me_2^{1,3}Ura$		Thy		Me^1Thy		$Me_2^{1,3}Thy$	
	cis	trans	cis	trans	cis	trans	cis	trans	cis	trans	cis	trans
IR Band[a]												
O-H	3356	3226sh	3300	3300	3230	3230	3425sh	3430sh	3250	3250	3300	3300
O-H	-	-	-	-	-	-	3333	3370	-	-	-	-
C=O	1156	1139	1183	1176	1130	1149	1174	1205sh	1143	1136	1136	1148
C=O	1139sh	1126	1136	1124	1170sh	1099	1114	1170	1111sh	-	1099sh	1111sh
C-O	1087	1111	1099	1075sh	1064	1064sh	1080	1130sh	-	1064sh	1075sh	1081sh
C-O	1075sh	1064	1036	1064	1053sh	1047	1058	1099	1053	1053	1053	1064
Mass[b]												
m/e	128	128	160	160	174	174	142	142	174	174	171	171
	$(M-18)^+$	$(M-18)^+$	$(M)^+$	$(M)^+$	$(M)^+$	$(M)^+$	$(M-18)^+$	$(M-18)^+$	$(M)^+$	$(M)^+$	$((M+1)-18)^+$	$((M+1)-18)^+$
M.P. ^oC	229-232[c]	>250	189-192[c]	145-148	-[d]	-[d]	191-193[e]	145-147[d]	>250	155-158	-[d]	-[d]

[a] *Spectra were taken with KBr pellets.*

[b] *Spectra were taken on CEC-21-110 mass spectrometer at 70 eV ionizing voltage and a source temperature of 120°C. (Authors thank Dr. C. Fenselau and Ms. Nancy Kan for these determinations.)*

[c] *Decomposition occured during melting.*

[d] *Product appeared gummy and possibly has an m.p. close to ambiant temperature.*

[e] *Melting points reported* (8) *are 190-191 and 195-197°C, respectively.*

the yield of the relatively stable cis Thy glycol increased with the irradiation time; however, the yields of the relatively unstable trans glycols of Thy and Ura showed no further increase after reaching ∿20% at 4 hr, and, as a matter of fact, a slight decrease was detected with continued irradiation. The yield of the unstable cis Ura glycol reached ∿20% also at 4 hr; however, its gradual decrease to ∿10% at 12 hr was observed. Thus, either in the collection of products or in the study of mechanisms, the stability and the photoreactivity leading to secondary reactions of particular compounds must be taken into consideration.

The above clearly indicate that sufficient quantities of certain radiation products may be prepared under specific photolytic conditions. This, in turn, permits further studies in radiation biology (14). In addition, because H_2O_2 and O_2 are present in biological systems, under the influence of the near-UV from sunlight, similar free-radical reactions may occur. Therefore, a detailed study of this sort has been undertaken (15) in order to gain information which may be of value to the understanding of biological near-UV effects such as aging, mutagenesis, or carcinogenesis.

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