basis of physical and spectroscopic properties. The residual tar from the reaction was not investigated.

Kinetic Study. A purified sample of *N,N*-dibromo-α-aminoisobutyronitrile (0.831 g, 0.0034 mol) was dissolved in 15 mL of methylene chloride. An aliquot (0.20 mL) was analyzed iodometrically for positive bromine with 0.008 N $Na_2S_2O_3$ solution. The solution was transferred to a 50-mL round-bottom flask (magnetic stirrer, ice bath, and drying tube). In a separate vial, recrystallized nitrosobenzene (0.367 g, 0.0034 mol) was dissolved in 10 mL of methylene chloride and chilled to 0 °C. After equilibration, the time was recorded and the solution of nitrosobenzene was added in one portion to the vigorously stirred solution of *N,N*-dibromoamine. Progress of the reaction at 0 °C was followed by observing the progressive color change: dark green, light brown, orange, and finally red. Upon development of a pronounced red color, 0.20-mL aliquots of reaction mixture were quickly withdrawn and titrated iodometrically. The process was repeated every 15–20 min until the assay for positive bromine was within 50 ± 3% of the initial value (adjusted for dilution with 10 mL of nitrosobenzene solution). By this method, reaction time was estimated to be 5.50 h. Further titration (12 h) of aliquots revealed no additional decrease in positive bromine.

According to the same procedure, the *N,N*-dibromoamine (0.410 g, 0.00169 mol) was reacted with nitrosobenzene (0.181 g) in 25 mL of methylene chloride. Estimated total reaction time was 5.25 h. Duplicate runs with reaction mixtures initially 0.00380 (25 mL) and 0.00182 mol (25 mL) in both starting materials required 6.00 and 5.50 h, respectively, for completion.

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Registry No.—**1**, 16248-71-8; **2**, 2156-72-1; **3**, 41718-25-6; **4**, 69083-98-3; **5**, 10218-84-5; **6**, 26307-01-7; **7**, 14925-83-8; **8**, 69083-99-4; **9**, 52548-05-7; **10**, 13698-16-3; **11**, 473-34-7; *N*-(isobutyronitrilo)-*N'*-phenyldiazene *N'*-oxide, 52123-68-9; *N*-*tert*-butyl-*N'*-phenyldiazene *N'*-oxide, 52123-67-8; *N*-(1-methyl-1-butylpentyl)-*N'*-phenyldiazene *N'*-oxide, 69815-23-2; *N*-*tert*-octyl-*N'*-phenyldiazene *N'*-oxide, 69815-24-3; *N*-isopropyl-*N'*-phenyldiazene *N'*-oxide, 52123-65-6; *N*-cyclohexyl-*N'*-phenyldiazene *N'*-oxide, 52123-66-7; *N*-butyl-*N'*-phenyldiazene *N'*-oxide, 52123-78-1; *N*-neopentyl-*N'*-phenyldiazene *N'*-oxide, 69815-25-4; *N*-isobutyl-*N'*-phenyldiazene *N'*-oxide, 69815-23-5; *N*-carbethoxy-*N'*-phenyldiazene *N'*-oxide, 56751-20-3; *N*-tosyl-*N'*-phenyldiazene *N'*-oxide, 60126-94-5; nitrosobenzene, 586-96-9; *N*-(isobutyronitrilo)-*N'*-*tert*-butyldiazene *N'*-oxide, 69815-27-6; 2-methyl-2-nitrosopropane, 917-95-3; *N,N*-dibromo-α-aminoisobutyronitrile, 69083-93-8; α-aminoisobutyronitrile hydrochloride, 50846-36-1; *N,N*-dibromo-*tert*-butylamine, 51655-36-8; *N,N*-dibromo-1-methyl-1-butylpentylamine, 69083-94-9; *N,N*-dibromo-*tert*-octylamine, 69083-95-0; *N,N*-dibromoisopropylamine, 55877-59-3; *N,N*-dibromocyclohexylamine, 68277-74-7; *N,N*-dibromobutylamine, 68277-73-6; *N,N*-dibromoneopentylamine, 69083-97-2; *N,N*-dibromoisobutylamine, 69083-96-1; ethyl *N,N*-dibromocarbamate, 51066-06-9; *N,N*-dibromo-*p*-toluenesulfonamide, 21849-40-1.

References and Notes

(1) Paper 30. Chemistry of *N*-Halo Compounds.
(2) Postdoctoral Fellow, 1977–1978.
(3) V. Nelson, A. Serianz, and P. Kovacic, *J. Org. Chem.*, **41**, 1751 (1976).
(4) F. R. Sullivan, E. Luck, and P. Kovacic, *J. Org. Chem.*, **39**, 2967 (1974).
(5) R. C. Zawalski and P. Kovacic, *Synth. Commun.*, **8**, 549 (1978).
(6) J. W. Mellor, "A Comprehensive Treatise on Inorganic and Theoretical Chemistry", Longmans, Green and Co., New York, 1922, p 245.
(7) J. H. Boyer, "The Chemistry of the Nitro and Nitroso Groups", Part I, H. Feuer, Ed., Interscience, New York, 1969, pp 284–285.
(8) P. Kovacic, M. K. Lowery, and K. W. Field, *Chem. Rev.*, **70**, 639 (1970).
(9) M. Anbar, S. Guttmann, and R. Rein, *J. Am. Chem. Soc.*, **81**, 1816 (1959); L. Farkas, M. Lewin, and R. Bloch, *ibid.*, **71**, 1988 (1949); M. Anbar and R. Rein, *ibid.*, **81**, 1813 (1959).
(10) P. A. S. Smith, "Open Chain Nitrogen Compounds", Vol. 2, W. A. Benjamin, New York, 1966, Chapter 13, p 373.
(11) (a) F. D. Lewis and W. H. Saunders, Jr., "Nitrenes", W. Lwowski, Ed., Interscience, New York, 1970, pp 82, 86; (b) P. A. S. Smith, *ibid.*, p 103.
(12) T. Fuchigami, T. Nonaka, and K. Iwata, *J. Chem. Soc., Chem. Commun.*, 951 (1976).
(13) W. Gottardi, *Monatsh. Chem.*, **105**, 611 (1974).
(14) D. Carr, T. P. Seden, and R. W. Turner, *Tetrahedron Lett.*, 477 (1969).
(15) (a) G. A. Russell, E. J. Geels, F. J. Smentowski, K.-Y. Chang, J. Reynolds, and G. Kaupp, *J. Am. Chem. Soc.*, **89**, 3821 (1967); (b) ref 7, pp 160 and 161.
(16) P. Kovacic and S. S. Chaudhary, *Org. Synth.*, **48**, 4 (1968).
(17) T. A. Kling, R. E. White, and P. Kovacic, *J. Am. Chem. Soc.*, **94**, 7416 (1972).

Chemical Evolution. 33. Photochemical Decarboxylation of Orotic Acid, Orotidine, and Orotidine 5'-Phosphate

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Orotic acid, orotidine, and orotidine 5'-phosphate are photochemically converted to uracil, uridine, and uridine 5'-phosphate in chemical yields of 13, 45, and 23%, respectively. The chemical yields for uracil and uridine formation are 1.6×10^{-5} and 1.7×10^{-2}, respectively. The chemical yield of uracil increases 2.5-fold when the orotic acid concentration is decreased 10-fold, indicating that bimolecular reactions limit the uracil yield. The reaction proceeds from the singlet excited state as shown by the absence of quenching by paramagnetic ions and the absence of sensitization by benzophenone and acetone. The Fe(III)- and Cu(II)-promoted photochemical formation of uracil from orotic acid proceeds in up to 17% yield. Small amounts of barbituric acid are also observed. A plausible pathway for the prebiological formation of uracil and its derivatives from HCN via orotic acid and its derivatives is discussed.

Hydrogen cyanide is considered to have been a likely starting material for the synthesis of biomolecules on the primitive Earth.[1] It is formed in a variety of primitive Earth simulation experiments, and it is present in interstellar space.[1,2] Dilute aqueous solutions of HCN condense to give oligomers which in turn undergo hydrolytic decomposition to purines, pyrimidines, and amino acids. In addition, two of the compounds formed by the hydrolysis of HCN oligomers, 4-aminoimidazole-5-carboxamide and orotic acid (**1a**), are intermediates in the contemporary biosynthesis of purine and pyrimidine nucleotides, respectively. Primitive life forms may have had enzymes for the utilization of these compounds for nucleic acid synthesis once the supply of preformed purines and pyrimidines was exhausted.[3] Probably the first enzymes were not very efficient and only enhanced the rates of chemical processes modestly over that of the rate in the absence of an enzyme. It is likely that the same chemical processes also occurred under primitive Earth conditions in the absence of enzymes. The chemical conversion of 4-aminoimidazole-5-carboxamide to purines under primitive Earth conditions has

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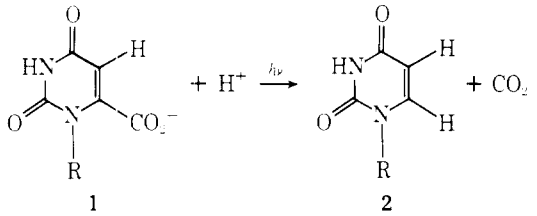
Table I. Photochemical Conversion of Orotic Acid to Uracil

irradiation time, h	concn, M	yield, %
1	6.4×10^{-4}	trace
	6.5×10^{-5}	2.0
2	6.4×10^{-4}	0.25
	6.5×10^{-5}	3.9
4	6.4×10^{-4}	0.45
	6.5×10^{-5}	7.8
6	6.4×10^{-4}	1.1
	6.5×10^{-5}	12.7
12	6.4×10^{-4}	2.2
	6.5×10^{-5}	11.5
24	6.4×10^{-4}	3.6
	6.5×10^{-5}	6.7
36	6.4×10^{-4}	4.7
48	6.4×10^{-4}	4.9
60	6.4×10^{-4}	3.3

already been described.[4] In this paper we describe the formation of uracil, uridine, and uridine 5′-phosphate from the corresponding orotate derivatives under plausible prebiological conditions.[5]

Results and Discussion

Orotic acid and its derivatives undergo a facile photochemical decarboxylation. At first it was thought that the photochemical decarboxylation of orotic acid was a new reaction, but careful scrutiny of the literature revealed that uracil formation had been suggested previously but not proved.[6] This is probably because 10^{-3} M orotic acid is converted to the dimer ($\Phi \approx 0.1$)[7] much faster than it is converted to uracil ($\Phi \approx 10^{-5}$).[5] The photodecarboxylation of orotidine and orotidine 5-phosphate has not been previously reported. The decarboxylation of orotidine to uridine constitutes a formal structure proof of orotidine.[8] The rate of formation of uridine from orotidine ($\Phi \approx 0.02$) is 1000 times faster than the rate of formation of uracil.



1 2

a, R = H
b, R = 1′-ribofuranosyl
c, R = 1′-ribofuranosyl 5′-phosphate

Bimolecular reactions decrease the yield of decarboxylated products as shown by our observation that the chemical yield of uracil goes from 5 to 13% when the orotic acid concentration is decreased from 6×10^{-4} to 6×10^{-5} M (Table I). In addition, the chemical yields of decarboxylation products from orotidine (**1b**) and orotidine 5-phosphate (**1c**), substances which do not undergo efficient photodimerization,[9] are 5–10 times greater than that of uracil from orotic acid (Tables II and III). In principle, the yield of uracil (**2a**) from orotic acid (**1a**) should be equivalent to the yield of uridine (**2b**) from orotidine (**1b**) since there is photoequilibrium between orotic acid and its photodimer.[9] The equilibrium was detected in the present work by the formation of uracil on irradiation of orotic acid photodimer. A fraction of the orotic acid present at equilibrium should be cleanly photolyzed to uracil, but the photoequilibrium is strongly to the side of the photodimer so that uracil formation requires 48 h while uridine formation requires only 4 h when identical reaction conditions are used. This prolonged irradiation time allows for the photodecomposition of uracil which lowers its yield.[10]

Table II. Photochemical Decarboxylation of Orotidine

irradiation time, h	yield of photoproducts, % uridine	uracil
1	18.6	0.7
2	37.8	1.3
3	44.9	3.9
4	37.9	5.6
6	31.6	7.3

Table III. Photochemical Decarboxylation of Orotidine 5′-Phosphate

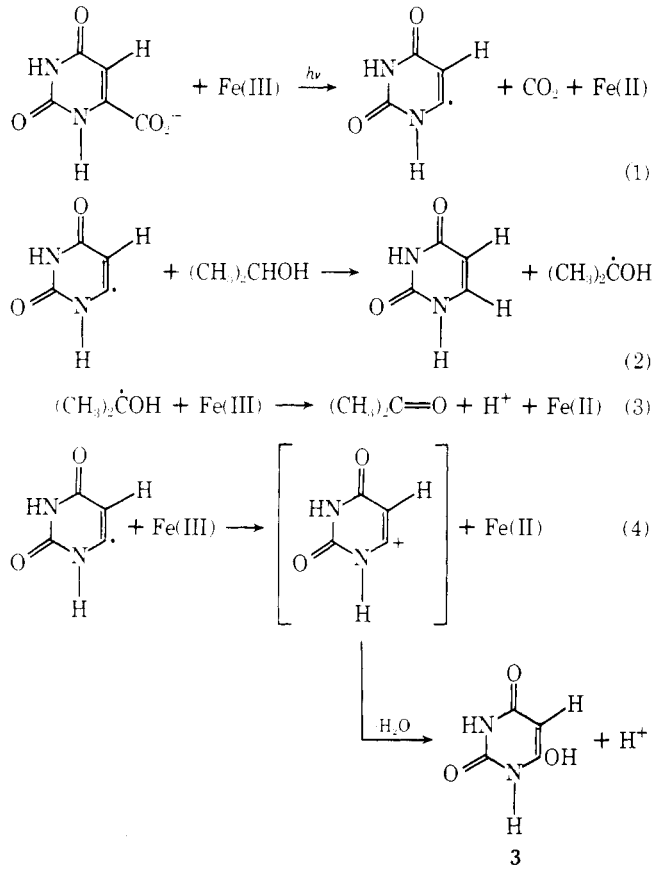
irradiation time, h	yield, % uridine 5′-phosphate	uracil	orotic acid
1	7.3	trace	2
2	14	1	1
3	22	2	<1
4	23	2.3	<1

The decreased yields of dimer formation from orotate anion, orotidine, and orotidine 5′-phosphate as compared to those of orotic acid are readily understood on the basis of the increasing steric and electrostatic repulsions. Most of the previous photochemical studies of orotic acid were performed at pH 1–3, well below pH 4.6, the pK_a of the excited state of orotic acid.[11] Dimerization is a higher energy pathway at pH 8.5, the pH used in the present research, because of the repulsions between the carboxylate anions, and hence the relative yield of uracil is enhanced. The steric bulk of the ribose moiety and the charge repulsions of the phosphate group result in a further decrease in dimer formation from orotidine and orotidine 5′-phosphate.

The photochemical decarboxylation of aryl carboxylic acids is an uncommon photoreaction and little is known about the reaction pathways or excited states involved.[12] The photodecarboxylation of benzoic acid has been shown to proceed from a vibrationally excited ground state,[13a] and the same may be true for nalidixinic acid since it undergoes a thermal decarboxylation at 150 °C as well as photodecarboxylation.[13b] The photodecarboxylation of orotic acid does not proceed from the triplet state as shown by the absence of the sensitized formation of uracil using benzophenone or acetone and the absence of quenching with Fe(II). The sensitization experiment was complicated by the concurrent photoreaction of uracil. However, a 2% yield of uracil would have survived 6 h of irradiation. The photodimer of orotic acid was the main product of the sensitization with benzophenone. Its formation proceeds with triplet sensitizers and is quenched with paramagnetic ions.[6,14-16] Our observation that decarboxylation proceeds with 254-nm but not 300-nm light suggests the reaction goes via an upper singlet excited state or a vibrationally excited ground state.[17] The vibrationally excited ground state is favored because of the literature precedents cited above[13] and because thermal decarboxylation of orotic acid and its derivatives is a known reaction pathway.[18]

The metal ion promoted decarboxylation of orotic acid is observed when orotic acid is photolyzed in the presence of Fe(III) or Cu(II) (Table IV).[19] Barbituric acid (**3**) is also formed in small amounts. The uracil yield was enhanced by the addition of 2-propanol to the raction mixture. The result is consistent with the redox pathway proposed in Scheme I. This pathway predicts that Fe(II) would not facilitate uracil

Scheme I



formation as was observed. The reducing agent which serves as the hydrogen donor in the absence of added 2-propanol (eq 2) is unknown. Water is not a likely possibility because its bond energy (119 kcal) is much greater than the C–H bond energy of most organic compounds.[20]

The previous pathways proposed for the prebiotic formation of pyrimidines required bimolecular reactions between different chemical reagents.[20] It is not always clear that sufficient quantities of both reagents would have been present at the same location on the primitive earth for reaction to take place. The formation of orotic acid and other pyrimidines from HCN requires only the presence of sufficient HCN for HCN oligomer formation to take place. The slow hydrolysis of these oligomers releases orotic acid and other biomolecules.[2] The photochemical decarboxylation of orotic acid to uracil is a plausible extension of the prebiotic synthesis of pyrimidines from HCN. The only additional reagent required, other than the HCN and the H_2O used to form the oligomers, is UV light which was the most abundant source of energy on the primitive Earth.[22] Since photochemical reactions are zero-order processes, the extent of photolysis is not limited by low concentrations of orotic acid. In fact, dilute solutions of orotic acid are a real advantage since the rate of competitive bimolecular photoprocesses is greatly diminished (Table I). The photodecarboxylation of orotidine and orotidine 5′-phosphate extends the scope of this prebiotic pyrimidine synthesis. The pyrimidine nucleosides and nucleotides may have formed on the primitive Earth either before or after the photodecarboxylation reaction took place.

The metal-ion-promoted photodecarboxylation of orotic acid is also a plausible prebiotic route to uracil. Both Fe(III) and Cu(II) form complexes with orotic acid[23] so that a bimolecular reaction between reactants present in small amounts on the primitive Earth would not have been rate limiting. The uracil yield may have been enhanced by hydrogen atom donors similar to 2-propanol in the primitive oceans. Iron and copper are among the most abundant transition metals in the Earth's crust and were undoubtedly present in significant amounts on the primitive Earth.[24] The metal ions may have initially been present in their lower oxidation states, but Fe(III) and Cu(II) oxidation states necessary for uracil formation would have formed after the atmosphere became more oxidizing.

Table IV. Metal-Ion-Promoted Photodecarboxylation of Orotic Acid[a]

metal salt	added reagent	time, h	% uracil
		6	1.1
		24	3.6
$Fe(ClO_4)_2$ (4.1×10^{-4} M)		24	3.6
$Cu(NO_3)_2$		6	7.5
$Fe(ClO_4)_3$		6	7.9
$Fe(ClO_4)_3$ (1.3×10^{-3} M)		6	7.2
$Fe(ClO_4)_3$		12	8.1
$Fe(ClO_4)_3$		24	11.9
$Fe(ClO_4)_3$		36	12.8
$Fe(ClO_4)_3$		48	14.1
$Fe(ClO_4)_3$		96	17.3
$Fe(ClO_4)_3$		120	7.5
$Fe(ClO_4)_3$	$Fe(ClO_4)_2$ (5.9×10^{-4} M)	6	6.7
$Fe(ClO_4)_3$	2-propanol (1.7×10^{-2} M)	6	7.1
$Fe(ClO_4)_3$	2-propanol	6	10.5
$Fe(ClO_4)_3$	2-propanol	24	20.3

[a] All reactants were 6.4×10^{-4} M unless noted otherwise.

The proposed prebiotic synthesis of uracil and its derivatives is similar to the contemporary biotic pathway for the conversion of orotidine 5′-phosphate to uridine 5′-phosphate; this similarity provides further support for the validity of the proposed prebiotic pathway because there would have been no discontinuities in the transition from prebiology to biology.[3] The change from a photochemical prebiological process to a thermal biological process may appear to be inconsistent with such a simple transition. However, since the photochemical reaction probably proceeds from a vibrationally excited ground state, the same thermal reaction pathway is involved in both the prebiotic and biotic reactions. The only requirement is the evolution of the appropriate enzymes to catalyze the biological process once life originated.[25]

Experimental Section

General Procedures. UV spectra were determined on a Unicam SP 800 spectrophotometer. Reaction solutions were made up in doubly distilled water in which the final distillation was performed in a glass still. Photolyses were performed using a Rayonet Photochemical Reactor equipped with lamps with their principal light output (1.2×10^{-4} einstein min^{-1}) at 254 nm. Lamps with their principal emission at 300 nm were used in the photochemical reactor as a source of longer wavelength light. Samples were irradiated in 30 × 3.5 cm quartz tubes and were purged with purified nitrogen[26] for 1 h before photolysis and during the course of the photolysis. Pyrex tubes were used when 300-nm lamps were employed. Ascending chromatography was performed on Whatman 3 mm paper using the following solvent systems: (1) 1-propanol/NH_4OH (17%) (3:1); (2) 1-butanol/acetic acid/H_2O (12:3:5); (3) ethyl acetate/formic acid/H_2O (7:2:1). Compounds were visualized with UV light and were identified by comparison of the R_f values of the eluted sample with standards in the three solvent systems. The products were further identified by comparing the UV spectrum of the eluted sample measured in acidic, neutral and basic solution with that of authentic sample. Quantitative analyses were obtained from the UV absorbances of the unknown and authentic samples. Paper electrophoresis was used to separate the photoproducts of orotidine 5′-phosphate. Electrophoresis was carried out for 2 h at 3500 V using a Savant FP-30A flat plate system. The samples were run on Whatman 3 mm paper which had been dampened with a pH 4.5 buffer made up of 9.4 mL of acetic acid, 10.3 g of sodium acetate, and 1.3 g of EDTA diluted to 3 L. The orotic acid was

obtained from Aldrich and orotidine and orotidine 5′-phosphate were purchased from Sigma. These compounds were shown to be pure by paper chromatograpy and, in the case of orotidine 5′-phosphate, paper electrophoresis. Reagent grade salts were obtained principally from Fisher Scientific Co.

Photolysis of Orotic Acid. The photolyses were performed on 6.4 $\times 10^{-4}$ M or 6.5 $\times 10^{-5}$ M solutions which had been adjusted to pH 8.5 with NaOH. After irradiation, the pH of the solutions was found to be in the 7–8 range. The uracil yield was 30–40% lower when oxygen was present. Oxygen was removed by nitrogen purging since this procedure gave yields comparable to those obtained when three freeze–pump–thaw cycles were used. No uracil was detected when 10^{-2} M orotic acid was irradiated in a Pyrex vessel using a 300-nm light source. The absorbance of a 10^{-2} M solution is greater than 1 at wavelengths below 330 nm. The results are given in Table I.

Synthesis and Photolysis of Orotic Acid Dimer. The photodimer of orotic acid was prepared by irradiating 10^{-2} M orotic acid in a Pyrex vessel using a 300-nm light source.[27] The mixture was purged with nitrogen prior to irradiation and the nitrogen was bubbled through the solution during the course of the irradiation. The dimer was eluted from a paper chromatogram which was developed with solvent system 3. The UV absorption spectrum of the eluted sample corresponded exactly with that of the published spectrum when measured in acidic, neutral, and basic media.[27]

An aqueous pH 8.5 solution of the dimer was irradiated in a quartz tube using a 254-nm light source for 6 h. Both uracil and orotic acid were identified as photoproducts by paper chromatography.

Photolysis of Orotidine. The photolyses were performed on 6.4 $\times 10^{-4}$ M solutions which had been adjusted to pH 8.5 with NaOH. The structures of the samples of uracil and uridine eluted from the paper chromatograms were assigned by UV spectroscopy. In addition, the structures of the photoproducts were assigned by the identity of the gas chromatographic retention times of their silyl derivatives on OV-1 and OV-17 with those of authetic samples.[1] Uridine was further identified by hydrolysis to uracil by heating it for 2 h at 120 °C in a sealed ampul. The uracil formed was identified by its R_f value in system 3 and by the UV absorption spectrum of the eluted sample in acidic and basic media. The yields of photoproducts are given in Table II.

The uracil obtained by the photolysis of orotidine was probably formed by the photochemical hydrolysis of uridine. Irradiation of a 4.1 $\times 10^{-4}$ M aqueous solution of uridine for 12 h resulted in an 87% loss of uridine and a 35% yield of uracil.

Quantum Yield Measurements. Quantum yields were determined using a 0.006 M ferrioxalate actinometer.[28] The ferrioxalate was irradiated in the photochemical reactor in the same tube (3.5 × 30 cm) used for the irradiation or orotic acid and its derivatives. The intensity of the 254-nm lamps was found to be 1.2 $\times 10^{-4}$ einstein min^{-1}. The quantum yield for uracil formation from orotic acid (6.4 $\times 10^{-4}$ M) was determined by measuring the uracil yield at different photolysis times. The uracil was determined by its UV absorption after purification by paper chromatography. A linear plot of uracil formed vs. time was obtained up to 12 h of irradiation. From the slope of the linear plot, it was determined that the rate of uracil formation was 1.9 $\times 10^{-9}$ mol min^{-1}. The quantum yield of uracil formation was calculated to be 1.6 $\times 10^{-5}$ from these data.[5]

The quantum yield for uridine formation from orotidine was found to be 1.7 $\times 10^{-2}$ by the same procedure. The rate of uridine formation was 2.05 $\times 10^{-6}$ mol min^{-1}.

Orotic acid exhibits a UV maximum at 278 nm (ϵ 8 $\times 10^3$)[7] and it exhibits molar extinction coefficients of 2.8 $\times 10^3$ and 3.1 $\times 10^3$ at 254 and 300 nm, respectively. A 6.4 $\times 10^{-4}$ M solution with a 3-cm path length exhibits absorbances of 5.4, 16, and 6.0 at 254, 278, and 300 nm, respectively. A 6.4 $\times 10^{-4}$ M solution of orotic acid in a 3-cm cell absorbs >90% of the light at wavelengths <310 nm. Orotidine exhibits absorbances of 5.7 $\times 10^3$, 8 $\times 10^3$, and 2.6 $\times 10^2$ at 254, 267 (UV_{max}), and 300 nm, respectively. A 6.4 $\times 10^{-4}$ M solution with a 3-cm path length exhibits absorbances of 11, 15.4, and 0.5 at 254, 267, and 300 nm, respectively. A 6.4 $\times 10^{-4}$ M solution of orotidine in a 3-cm cell absorbs >90% of the light at wavelengths <294 nm. The linearity of the plots of yield vs. time confirm that the irradiated solutions are absorbing all the 254-nm light emitted.

The actual quantum yields are about 25% greater than those calculated because the low-pressure mercury lamps used in this study emit about 25% of their light at 313 nm.[29] This longer wavelength light affects the formation of Fe(II) from ferrioxolate but does not affect the decarboxylation of orotic acid.

Photolysis of Orotidine 5′-Phosphate. The photolyses were performed on 6.4 $\times 10^{-4}$ M solutions. The initial pH of the solution was 9.1 and this decreased to about 8 during the course of the photolyses. The photoproducts were analyzed by paper chromatography in system 3 and by paper electrophoresis. The reaction products indicated in Table III were the only UV absorbing products that were detectable. The identity of the uridine 5′-phosphate was confirmed by its hydrolysis to uracil at 120 °C with 1 N HCl for 2 h. The uracil formed was identified by its paper chromatographic R_f value in system 3.

Attempted Sensitized Decarboxylation of Orotic Acid. (a) No uracil was detected by the photolysis of a 6.4 $\times 10^{-4}$ M solution of orotic acid at pH 8.5 which contained either 1.1 $\times 10^{-4}$ M benzophenone or 1.4 $\times 10^{-2}$ M acetone. The mixture was analyzed after irradiating with a 300-nm light source in a Pyrex tube. No uracil was detectable after irradiation for 24 or 96 h when benzophenone was the sensitizer and after 96 h when acetone was the sensitizer. Most of the orotic acid was converted to other products after 96 h.

(b) An aqueous solution which contained 6.4 $\times 10^{-4}$ M orotic acid and 10^{-4} M benzophenone was adjusted to pH 8.5, purged with nitrogen, and photolyzed using a 350 nm light source. Aliquots (10 mL) were taken every 2 h over the first 8 h and then after 24 h. These were concentrated on a rotary evaporator and chromatographed using solvent system 3. No uracil was detected in any of the aliquots. A 1% uracil yield (based on orotic acid) was the limit of detection. The orotic acid was converted to its dimer as shown by the end absorption at about 200 nm and a product with the R_f of orotic acid dimer (0.06) in solvent system 3. The half-life for dimer formation was 6 h, and most of the orotic acid had reacted after 24 h.

In a control experiment performed at the same time, the same mixture of orotic acid and benzophenone was irradiated after a 2% yield of uracil was added to it. The uracil could still be detected after 6 h, but none was present after 8 h and 24 h.

Metal-Ion-Promoted Decarboxylation of Orotic Acid. The photolyses were performed on a 6.4 $\times 10^{-4}$ M pH 8.5 solution of orotic acid in the presence of a 6.4 $\times 10^{-4}$ M solution of the metal salt. Uracil was identified by its R_f value in system 1 and by the UV spectrum of an eluted sample. The yields given in Table I were determined from the absorbance of the eluted sample. Barbituric acid was found to be a reaction product by irradiating orotic acid with 6.4 $\times 10^{-4}$ M $Fe(ClO_4)_3$ and 6.4 $\times 10^{-4}$ M $Cu(ClO_4)_2$ for 6 h. The reaction mixture was chromatographed using system 1, and material with the same R_f as barbituric acid was eluted and rechromatographed in system 3. The material with the same R_f as barbituric acid exhibited UV spectra in acid and base identical with those of barbituric acid. Yields of 0.27 and 1.1% were obtained using Fe(III) and Cu(II), respectively.

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Registry No.—**1a**, 65-86-1; **1b**, 314-50-1; **1c**, 2149-82-8; **2a**, 66-22-8; **2b**, 58-96-8; **2c**, 58-97-9; **3**, 67-52-7; orotic acid dimer, 6993-21-1.

Reference and Notes

(1) Sanchez, R. A.; Ferris, J. P.; Orgel, L. E. *J. Mol. Biol.* **1967,** *30,* 223–253. Ferris, J. P.; Joshi, P. C.; Lawless, J. G. *Biosystems* **1977,** *9,* 81–86. Ferris, J. P.; Joshi, P. C.; Edelson, E. H.; Lawless, J. G. *J. Mol. Evol.* **1978,** *11,* 293–311: and references therein.
(2) Horowitz, N. H. *Acc. Chem. Res.* **1976,** *9,* 1–7. Gannon, R. H. *Chem. Eng. News* **1978,** *56* (40), 21–33.
(3) Horowitz, N. H. *Proc. Natl. Acad. Sci. U.S.A.* **1945,** *31,* 153–157.
(4) Sanchez, R. A.; Ferris, J. P.; Orgel, L. E. *J. Mol. Biol.* **1968,** *38,* 121–128.
(5) A preliminary report of a portion of this work has been published: Ferris, J. P.; Joshi, P. C. *Science* **1978,** *201,* 361–362. The quantum yield for uracil formation from orotic acid was erroneously reported to be 1.6 $\times 10^{-4}$ in this preliminary paper.
(6) Charlier, M.; Helene, C. *Photochem. Photobiol.* **1967,** *6,* 501–504. Whillans, D. W.; Johns, E. H. *ibid.* **1969,** *9,* 323–330.
(7) Sztumpf, E.; Shugar, D. *Photochem. Photobiol.* **1965,** *4,* 719–733.
(8) Fox, J. J.; Wempen, I. *Adv. Carbohydr. Chem.* **1959,** *14,* 320.
(9) Sztumpf-Kulikowska, E.; Shugar, D.; Boag, J. W. *Photochem. Photobiol.* **1967,** *6,* 41–54.
(10) Varghese, A. J. *Biochemistry* **1971,** *10,* 4283–4290: and references therein.
(11) Herbert, M. A.; Johns, H. L. *Photochem. Photobiol.* **1971,** *14,* 693–704.
(12) Epling, G. A.; Ayengar, N. K. N.; Lopes, A.; Yoon, U. C. *J. Org. Chem.* **1978,** *43,* 2928–2930.
(13) (a) Chau, F.; Gibbons, C.; Barton, D. *Can. J. Chem.* **1972,** *50,* 2017–2021. (b) Detzer, N.; Huber, B. *Tetrahedron* **1975,** *31,* 1937–1941.
(14) Charlier, M.; Helene, C. *Photochem. Photobiol.* **1972,** *15,* 71–87. Greenstock, C. L.; Johns, H. E. *Biochem. Biophys. Res. Commun.* **1968,** *30,* 21–27.
(15) Beukers, R.; Berends, W. *Biochem. Biophys. Acta* **1960,** *38,* 573–575.
(16) Haug, A. *J. Am. Chem. Soc.* **1964,** *86,* 3381–3384.

(17) Turro, N. J.; Ramamurthy, V.; Cherry, W.; Farneth, W. *Chem. Rev.* **1978,** *78,* 125–145.
(18) Beak, P.; Siegel, B. *J. Am. Chem. Soc.* **1973,** *95,* 7919—7920. Beak, P.; Siegel, B. *ibid.* **1976,** *98,* 3601–3606. Atkinson, M. R.; Maguire, M. H.; Ralph, R. K.; Shaw, G.; Warren, R. N. *J. Chem. Soc.* **1957,** 2263–2268.
(19) Kimura, T.; Kamimura, J.; Takada, K.; Sugimori, A. *Chem. Lett.* **1976,** 237–240.
(20) Weast, R. C. "Handbook of Chemistry and Physics"; Chemical Rubber Publishing Co.: Cleveland, Ohio, 1972; F-189.
(21) Recent discussion of the prebiotic formation of pyrimidines may be found in: Chittenden, G. J. F.; Schwartz, A. W. *Nature, (London)* **1976,** *263,* 350–381. Schwartz, A. W.; Chittenden, G. J. F. *Biosystems* **1977,** *9,* 87–92. Choughuley, A. S. U.; Subbaraman, A. S.; Kazi, Z. A.; Chada, M. S. *ibid.* **1977,** *9,* 73–80.
(22) Miller, S. L.; Orgel, L. E. "The Origins of Life on Earth"; Prentice Hall: Englewood Cliffs, New Jersey, 1974.
(23) Tucci, E. R.; Doody, E.; Li, N. C. *J. Phys. Chem.* **1961,** *65,* 1570–1574. Tucci, E. R.; Takahashi, F.; Tucci, V. A.; Li, N. C. *J. Inorg. Nucl. Chem.* **1964,** *26,* 1263–1276. Ferris, J. P.; Joshi, P. C. unpublished.
(24) McClendon, J. H. *J. Mol. Evol.* **1976,** *8,* 175–195. Egami, F. *ibid.* **1974,** *4,* 113–120.
(25) For a recent discussion of the enzyme-catalyzed decarboxylation of orotidine 5′-phosphate see: Tax, W. J. M.; Veerkamp, J. H.; Trijbels, F. J. M.; Schretlan, E. D. A. M. *Biochem. Pharmacol.* **1976,** *25,* 2025–2032.
(26) Gordon, A. S.; Ford, R. A. "The Chemist's Companion"; Wiley: New York, 1972; p 438.
(27) Sztumpf, E.; Shugar, D. *Photochem. Photobiol.* **1965,** *4,* 719–733.
(28) Hatchard, C. G.; Parker, C. A. *Proc. R. Soc. London, Ser. A,* **1956,** *253,* 518–536. Murov, S. L. "Handbook of Photochemistry"; Marcel Dekker: New York, 1973; p 122.
(29) Technical data sheet, Southern New England Ultraviolet Co., Middletown, Conn. 06457.

Ozonation of Organic Compounds. 2. Ozonation of Phenol in Water[1]

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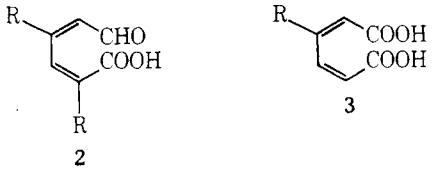
The rates and products of the ozonation of phenol were studied in water at 30 °C. In contrast to earlier works, it was found that the major product was formic acid with minor amounts of glyoxal, glyoxylic acid, oxalic acid, carbon dioxide, and hydrogen peroxide. Several other intermediates were also observed. The ozonation of several model compounds was also carried out. It was concluded that the anomalous ozonolysis of α,β-unsaturated carbonyl groups played an important role in the ozonation of phenol. The ratio of anomalous ozonolysis was measured in water for several olefins.

The ozonation of organic compounds, especially of olefins, has been studied extensively in organic solvents for many years.[2-4] As pointed out by Bailey,[5] however, ozonation in water has received less attention. Among the water-soluble organic compounds, phenol has been studied most extensively.[6] Recently, Gould and Weber[7] carried out careful analyses of the products and found hydroquinone, catechol, glyoxal, glyoxylic acid, and oxalic acid. Eisenhauer[8] also identified catechol, hydroquinone, and *o*-quinone as intermediates. Wako[9] observed, although only qualitatively, hydroquinone, *p*-quinone, muconic acid, maleic acid, and glyoxylic acid. On the other hand, Skarlatos et al.[10] reported acetic acid as one of the ozonation products from aqueous phenol. Bernatek and his collaborators[11,12] ozonized phenol in ethyl acetate and found that the products containing active oxygen gave formic acid, carbon dioxide, glyoxal, and oxalic acid by hydrolysis. They also found that the ozonation of catechol, resorcinol, and quinol gave the same products.[11,12] However, they did not find glyoxylic acid. Thus, the results hitherto reported are inconsistent, and neither the products nor the mechanism of the ozonation of phenol in water has been conclusively established.

The objectives of our present study were to measure the rates and products of the ozonation of phenol and model compounds in water quantitatively as well as qualitatively and to elucidate the ozonation mechanism. It was found that the major products from phenol were formic acid and carbon dioxide and that the anomalous ozonolysis of the double bond conjugated with carbonyl groups played an important role.

Results

Ozonation of Phenol. Figure 1 shows the typical example of the ozonation of phenol in water; 0.618 mmol of phenol was oxidized with 0.12 mmol/min of ozone in 100 mL of water at 30 °C. It shows that phenol was oxidized quite readily with ozone without any noticeable induction period and that substantially all phenol disappeared in 90 min. The major product was formic acid with smaller amounts of muconaldehyde [*cis,cis*-6-oxo-2,4-hexadienoic acid (**2**)],[13] muconic acid [*cis, cis*-2,4-hexadienedioic acid (**3**)],[13] maleinaldehyde [*cis*-4-oxo-2-butenoic acid (**5**)],[13] glyoxylic acid, glyoxal, and oxalic acid. Besides the products shown in Figure 1, hydroquinone, catechol, carbon dioxide, and hydrogen peroxide were observed. In contrast to the results of Skarlatos,[10] acetic acid was not observed. As described later, catechol and hydroquinone are more reactive toward ozone than phenol, and hence these products are accumulated only at the initial stages of ozonation. These results are in agreement with those observed by Gould and Weber.[7] **2** and **3** are also readily oxidized, and their concentrations decreased after reaching a maximum at 45–60 min. Glyoxylic acid and oxalic acid concentrations increased with reaction time, whereas formic acid and **5** concentrations leveled off.



2 **3**

R = H or OH

Table I shows the final products after a 180-min ozonation. The total acid measured by titration was 1.34 mmol, which is in excellent agreement with 1.33 mmol observed by isotachophoretic analysis. The amount of double bond formed was approximately the same as that of **5**. Therefore, the amount of mucondialdehyde [*cis,cis*-1,6-dioxo-2,4-hexadiene (**1**)][13] and maleindialdehyde [*cis*-1,4-dioxo-2-butene (**4**)][13] should be small. The concentration of glyoxal was estimated from the total carbonyl groups, 0.90 mmol, and the amounts of glyoxylic acid and **5** observed, $(0.90 - 0.166 - 0.227)/2 = 0.253$ mmol. These organic products account for 69% of initial carbon. If it is assumed that the missing carbon is converted to carbon

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