and counting the difference in frequency between the observing and locking oscillators.

Spectra were calculated using the first half of LAOCOON-II.[13] Once a visual fit had been achieved, the chemical shifts and coupling constants were adjusted using LAOCN3.[9] Calculated spectra were plotted as envelopes of summed Lorentzian curves with 0.3 Hz half-height peak widths using SPCTRM-II.[18]

Cyclobutanol. Cyclobutanol was prepared by the acid-catalyzed rearrangement of cyclopropylcarbinol.[19] Cyclobutanone was prepared by the chromic acid oxidation of cyclobutanol and was obtained in a 20% yield. The lithium aluminum deuteride reduction of cyclobutanone gave cyclobutanol-1-d. Cyclopropylcarbinol-d_2 was obtained by the lithium aluminum deuteride reduction of ethyl cyclopropanecarboxylate and was treated with aqueous acid to give a mixture of cyclobutanol-2,2-d_2 and cyclobutanol-3,3-d_2.

A 1:1 mixture of cyclobutanone-2,2-d_2 and cyclobutanone-3,3-d_2 was prepared by the reaction of ketene-d_2 with diazomethane.[20] Ketene-d_2 was obtained by the pyrolysis of acetone-d_6 in a miniature ketene lamp.[21] Reduction of the ketone mixture with lithium aluminum deuteride gave a mixture of cyclobutanol-1,2,2-d_3 and cyclobutanol-1,3,3-d_3.

The exchange of the α-hydrogens of cyclobutanone with deuterium oxide was effected using sodium acetate as the catalyst.[19] Reduction with lithium aluminum hydride gave cyclobutanol-2,2,4,4-d_4. Reduction with lithium aluminum deuteride gave cyclobutanol-1,2,2,4,4-d_5. The acid-catalyzed rearrangement of cyclobutanol-2,2,4,4-d_4 using the conditions for the cyclopropylcarbinol rearrangement gave a mixture of the reactant and cyclobutanol-2,2,3,3-d_4. The acid-catalyzed rearrangement of cyclobutanol-1,2,2,4,4-d_5 gave a mixture of the reactant and cyclobutanol-1,2,2,-3,3-d_5.

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Solvolysis of Organic Phosphates. I. Pyridylmethyl Phosphates

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Abstract: Spontaneous hydrolyses of 2-, 3-, and 4-pyridylmethyl phosphates investigated at 90° and $\mu = 0.10$ over the $-\log [H^+]$ range from 1 to 8.7 followed first-order kinetics with respect to unreacted phosphate species, and the maximum rate was observed at approximately 2.5 in $-\log [H^+]$ for all the phosphates. Reactive species of each phosphate in hydrolysis were the neutral zwitterion and the monoanion, where the former was found to be more labile. A profound reactivity of the zwitterion form of 2-pyridylmethyl phosphate was attributed to the unique intramolecular proton-transfer mechanism. Among various bivalent metal ions used in hydrolysis of pyridylmethyl phosphates at 90° and $\mu = 0.10$, only the copper ion promoted the reaction rate of 2-pyridylmethyl phosphate to a considerable extent in a $-\log [H^+]$ range above 3.3. Since the copper ion did not demonstrate any meaningful catalysis for other pyridylmethyl phosphates, the particular copper complex formed with 2-pyridylmethyl phosphate in the transition state was put forward to elucidate this enhanced catalytic effect.

The considerable importance of phosphate esters in biological systems has prompted the solvolysis study of various organic phosphates in recent years, and a large amount of experimental results has been accumulated on the hydrolyses of monoalkyl phosphates. Reviews which treated the recent progress in this field have also appeared.[2-4] In spite of these active investigations hitherto carried out, the hydrolysis of organic phosphate esters, which contain heteroaromatic moieties, such as pyridine nuclei, has never been investigated to any extent.

On the other hand, the effect of metal ions on the hydrolytic reactions of phosphate monoesters is one of the interesting aspects of metal ion catalysis, and has been the subject of a number of research groups.[5] For example, the specific activities of some metal ions in the hydrolysis of salicyl phosphate have been investigated by Martell and his coworkers, and the catalytic process has been discussed in the light of the mechanistic structures of metal complexes formed with the substrate in the course of reaction.[6-8] However, previous investigations have not provided any convincing explanation of the reaction mechanism involved in these metal ion catalyses.

In the present work, the spontaneous hydrolyses of the three isomeric pyridylmethyl phosphates, *i.e.*, 2-pyridylmethyl phosphate, 3-pyridylmethyl phosphate, and 4-pyridylmethyl phosphate, were studied in aqueous media, and the catalytic effects of some bivalent metal ions on the hydrolysis reactions were investigated. In addition, the hydrolysis of phenyl phosphate was also studied in the presence of various metal ions. Since metal ions are expected to interact with phenyl phosphate at only the phosphate group in the course of reac-

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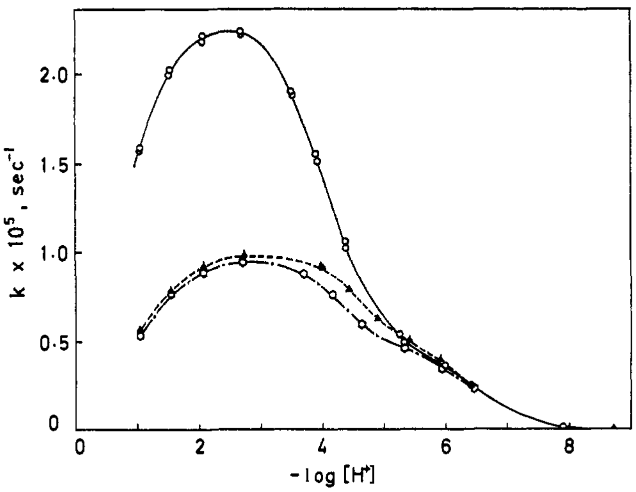

Figure 1. Rate profiles for the spontaneous hydrolysis of pyridylmethyl phosphates at 90.0 ± 0.1°, μ = 0.10: ○, 2 isomer; ○, 3 isomer; ▲, 4 isomer.

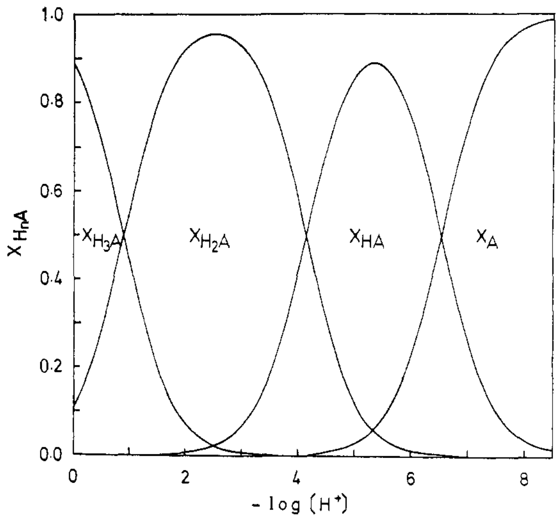

Figure 2. Distribution of ionic species of 2-pyridylmethyl phosphate as a function of $-\log [H^+]$ at 80°, μ = 0.10 ($NaClO_4$): H_3A, H_2A, HA, and A stand, respectively, for the monoprotonated, neutral, monoanionic, and dianionic species; X represents mole fraction of each species.

tion, this study may provide useful information for the catalytic hydrolyses of pyridylmethyl phosphates.

Experimental Section

Materials. Pyridylmethyl phosphates were prepared according to the procedure previously described.[9] Phenyl phosphate was prepared by hydrolyzing phenyl phosphorodichloridate, which was obtained by the reaction of phenol with phosphorus oxychloride,[10] and recrystallized from chloroform.

The stock solutions of metal ions were prepared from their nitrate salts. These solutions were standardized by the usual methods of chelatometric titration.

Apparatus. The reaction vessel used for kinetic and potentiometric measurements is a jacketed glass cell of 100-ml capacity and covered with a ground glass stopper which has several openings. The cell is equipped with electrodes for pH measurement, a delivery tip of the autoburet, and a thermometer. A set of electrodes used for pH measurements were glass electrode HG-6005, reference electrode HC-605, and temperature compensation electrode HR-105, which were designed for use at elevated temperatures by TOA Electronics, Ltd., Tokyo. The pH value of an experimental solution was maintained constant (within an accuracy of ±0.02) during each run with a TOA pH-Stat HS-1B which was fitted with an autoburet and incorporated with a TOA pH meter HM-5A.

Kinetic Measurements. The initial concentration of a substrate in an experimental solution was adjusted to 2×10^{-3} M, and the ionic strength was maintained at 0.10 with an appropriate inorganic salt, such as potassium nitrate, potassium chloride, or sodium perchlorate. The rate of hydrolysis was determined by measuring the amount of inorganic phosphate liberated in the course of reaction. The pH value of a solution was adjusted to a desired value by adding standard acid or base (0.1 N perchloric acid or 0.1 N sodium hydroxide). An experimental solution was made up to 100 ml in total volume and placed in a thermostated cell. After a desired temperature was attained, an aliquot sample (usually 2.5 ml) was taken out at an appropriate time interval and placed in a test tube. The tube was quickly cooled in cold water, and a specified volume (usually 2 ml) of the sample was pipetted into a 25-ml volumetric flask which contained a cold mixture of 60% perchloric acid (2 ml) and water (15 ml). The whole solution was then kept in a refrigerator for some time before the subsequent colorimetric analysis of inorganic phosphate.

The analytical method employed for the analysis of phosphoric acid was essentially that of Allen.[11] To a mixture which contained a 2.0-ml sample of the experimental solution, 2 ml of 60% perchloric acid, and 15 ml of water in a 25-ml volumetric flask, 2.0 ml of amidol reagent (a 100-ml aqueous solution contained 1.0 g of amidol and 20 g of sodium bisulfite) and 1.0 ml of 8.3% aqueous ammonium molybdate were added in this order. The mixture was diluted to 25 ml with water, and then placed in a constant-temperature bath at 25° for 15–25 min to develop the blue color. The cupric ion has itself absorption in the 720-mμ range, and the observed absorbance had to be corrected for the presence of this metal ion. On the other hand, the presence of chelating agents employed in this work did not show any interference with the colorimetric determination of inorganic phosphate.

pH Measurements. The pH values were established primarily by the use of standard buffers[12] over the pH range studied in the present work. The pH meter was calibrated before and after each kinetic run, and the pH variation was usually controlled within ±0.03. Then, pH values observed in accordance with the above procedure need to be converted into hydrogen ion concentrations. Thermodynamic data for weak electrolytes, which make the conversion of pH into hydrogen ion concentration possible, are not available at present. Thus, a conventional method was employed in this work; the pH values of aqueous perchloric acid of known concentrations were measured at an ionic strength of 0.10 to determine the apparent activity coefficient.

The acid dissociation constants of pyridylmethyl phosphates were evaluated at 80° by titrating 2.0×10^{-3} M solutions of the phosphates with standard base or acid. Titrations were carried out as quickly as possible to minimize any possible errors caused by the partial hydrolysis of the phosphates and checked by duplicate runs.

Results

Spontaneous Hydrolysis. Pyridylmethyl phosphates were hydrolyzed at 90° and an ionic strength of 0.10 in aqueous media. The hydrolysis reactions in the absence of metal ions followed apparent first-order kinetics with respect to the total concentration of unreacted phosphate. The specific rate constants determined experimentally are listed in Table I, together with the type of inorganic electrolyte employed to maintain the ionic strength constant. The maximum rates were observed, as is seen in Figure 1, around $-\log [H^+]$ = 2.5 and 5.5 for all the phosphates. For the assignment of specific rates to each ionic species, fractional molar concentrations were evaluated at different hydrogen ion concentrations by the aid of acid dissociation constants listed in Table II. The result for 2-pyridylmethyl phosphate is shown in Figure 2. From the comparison of

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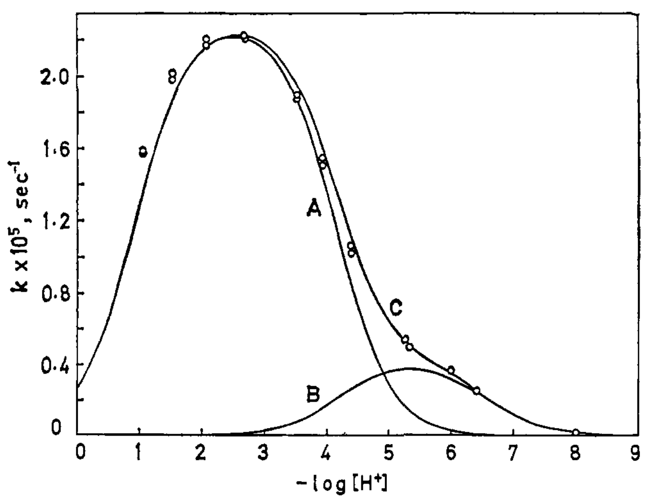

Figure 3. Contribution of each ionic species to the total rate in the spontaneous hydrolysis of 2-pyridylmethyl phosphate: A, rate profile due to the neutral species ($k_{H_2A}X_{H_2A}$); B, rate profile due to the monoionic species ($k_{HA}X_{HA}$); C, total rate profile ($k_{H_2A} \cdot X_{H_2A} + k_{HA}X_{HA}$); O, experimental point at 90°. Mole fraction of each ionic species was calculated from the pK values obtained at 80° (Table II).

Table I. Apparent First-Order Rate Constants for the Spontaneous Hydrolysis of Pyridylmethyl Phosphates at 90.0 ± 0.1° and μ = 0.10

−Log [H$^+$][a]	$k_0 \times 10^5$, sec^{-1}	Supporting electrolyte
	2-Pyridylmethyl Phosphate	
1.05	1.57	$HClO_4$
1.05	1.59	$HClO_4$
1.52	1.99	$HClO_4$–$NaClO_4$
1.53	2.02	$HClO_4$–$NaClO_4$
2.06	2.18	$HClO_4$–$NaClO_4$
2.06	2.21	$HClO_4$–$NaClO_4$
2.68	2.24	$NaClO_4$
2.69	2.21	$NaClO_4$
3.51	1.90	$NaClO_4$
3.51	1.88	$NaClO_4$
3.91	1.54	KCl
3.92	1.51	KCl
4.38	1.06	$NaClO_4$
4.39	1.02	KCl
5.25	0.533	KCl
5.32	0.486	KNO_3
6.00	0.355	KCl
6.41	0.244	KCl
7.9[b]	~0.008	KCl
	3-Pyridylmethyl Phosphate	
1.05	0.525	$HClO_4$
1.54	0.758	$HClO_4$–$NaClO_4$
2.08	0.878	$HClO_4$–$NaClO_4$
2.71	0.942	$NaClO_4$
2.72	0.942	KNO_3
3.70[c]	0.869	$NaClO_4$
3.71	0.869	KCl
4.17	0.753	KNO_3
4.65	0.589	KNO_3
5.35[d]	0.458	KNO_3
5.97	0.342	KNO_3
6.56[d]	0.231	KNO_3
(8.7)	~0.008	(Borate buffer)
	4-Pyridylmethyl Phosphate	
1.05	0.558	$HClO_4$
1.54	0.778	$HClO_4$–$NaClO_4$
2.08	0.917	$HClO_4$–$NaClO_4$
2.72	0.953	$NaClO_4$
2.73	0.975	KCl
3.99	0.908	KCl
4.02	0.900	KNO_3
4.44	0.789	KNO_3
4.91	0.625	KNO_3
5.47[d]	0.500	KNO_3
5.93	0.389	KNO_3
6.43[c]	0.247	KNO_3
(8.7)	~0.008	(Borate buffer)

[a] Variation was maintained within ±0.02 unless otherwise stated. [b] Value observed for the first hour. Deactivation of glass membrane made it difficult to measure pH value thereafter. [c] Variation was maintained within ±0.04. [d] Variation was maintained within ±0.03.

Table II. Acid Dissociation Constants of Pyridylmethyl Phosphates at 80.0 ± 0.1° and μ = 0.10 M ($NaClO_4$)

Acid	pK_{H_3A}	pK_{H_2A}	pK_{HA}
2-Pyridylmethyl phosphate	~1[a]	4.15	6.54
3-Pyridylmethyl phosphate	~1[b]	4.43	6.48
4-Pyridylmethyl phosphate	~1[b]	4.73	6.42

[a] Approximate value lies in the range 0.6–1.1. [b] Approximate value lies in the range 1.0–1.2.

Figure 1 with Figure 2, it is obvious that the reactive species are the neutral zwitterion and the monoionic form. Thus, the following equation was used to calculate the specific rate constants for these active species and the results are summarized in Table III.

$$k_0 = k_{H_2A}X_{H_2A} + k_{HA}X_{HA}$$

k_0, k_{H_2A}, and k_{HA} stand for the total rate constant and the specific rate constants of the neutral species, and of the monoionic species, respectively, and X_{H_2A} and X_{HA} refer to mole fractions of the neutral and the monoionic species, respectively. In Figure 3, the calculated pH-rate profile (curve C), as well as the calculated rate curves due to the neutral (curve A) and to the monoionic species (curve B), by the use of these k_{H_2A} and k_{HA} values is shown together with the experimental points for the hydrolysis of 2-pyridylmethyl phosphate. Likewise, satisfactory analysis of the experimental rate-profiles for the spontaneous hydrolyses of 3- and 4-pyridylmethyl phosphate was performed. The hydrolysis rates for 2- and 3-pyridylmethyl phosphates were measured at 70°: 2 isomer, $k_0 = 2.26 \times 10^{-6}$ sec^{-1} at $-\log [H^+] = 2.32$, $X_{HA} = 0.014$; 3 isomer, $k_0 = 8.64 \times 10^{-7}$ sec^{-1} at $-\log [H^+] = 2.30$, $X_{HA} = 0.007$. These data provide specific rate constants for the hydrolysis of the neutral species: 2 isomer, 2.39×10^{-6} sec^{-1}; 3 isomer, 9.11×10^{-7} sec^{-1}. Then, approximate activation energies for the hydrolysis of the neutral species of the above two phosphates were calculated from the rate data at 70 and 90°: 2 isomer, 28.2 kcal mol^{-1}; 3 isomer, 29.3 kcal mol^{-1}.

Bivalent Metal Catalysis. Equimolar concentrations of pyridylmethyl phosphate and bivalent metal ion were used to run kinetic measurements at 90° and the ionic strength of 0.10 in aqueous media. The reaction followed apparent first-order kinetics in these relatively low pH regions as shown in Table IV. However, the pH of a reaction mixture could not be adjusted at higher values well above those listed in Table IV without forming precipitates, although the magnesium ion was the exception. After all, only the copper ion demonstrated the significant effect on the hydrolysis of 2-pyridylmethyl phosphate. Thus, the catalysis of the copper ion was investigated in some detail as summarized

Table III. First-Order Rate Constants for the Neutral and the Monoionic species at 90.0 ± 0.1° and μ = 0.10

Phosphate	$k_{H_2A} \times 10^5$, sec^{-1}	$k_{HA} \times 10^5$, sec^{-1}
2-Pyridylmethyl phosphate	2.33	0.414
3-Pyridylmethyl phosphate	0.969	0.431
4-Pyridylmethyl phosphate	0.986	0.475

in Table V. The pH range was covered from 2.68, where the spontaneous hydrolysis reaches its maximum rate, up to 4.93. The copper ion catalysis becomes more effective as the $-\log [H^+]$ value of a reaction mixture increases. The copper ion catalysis was also studied in the presence of an equimolar amount of some selected chelating agents as shown in Table V. In these instances, the chelating agents tend to reduce the catalytic activity of the copper ion. It is also to be noted that the strict first-order kinetics did not hold in the $-\log [H^+]$ range above 4.15 and the deviation from this simple rate law was more marked as the pH value was raised. In these cases, the apparent first-order rate constants were obtained from the initial stage of the reaction.

Table IV. Effects of Metal Ions on the Hydrolysis of Pyridylmethyl Phosphates at 90.0 ± 0.1° and μ = 0.10[a,b]

Phosphate	Metal ion	$-\log [H^+]$	$k \times 10^5$, sec^{-1}	$(k/k_0) \times 100$[c]
2-Pyridylmethyl	Mg^{2+}	5.23 ± 0.02	0.511	97
	Co^{2+}	5.06 ± 0.02	0.586	99
	Ni^{2+}	4.97 ± 0.02	0.617	98
	Cu^{2+}	3.76 ± 0.02	2.76	165
	Zn^{2+}	3.92 ± 0.02	1.52	100
3-Pyridylmethyl	Ni^{2+}	4.52 ± 0.03	0.617	99
	Cu^{2+}	3.49 ± 0.02	0.869	96
4-Pyridylmethyl	Ni^{2+}	4.83 ± 0.03	0.633	99
	Cu^{2+}	3.59 ± 0.02	1.02	107

[a] To maintain the ionic strength constant KCl was used for 2-pyridylmethyl phosphate, and KNO_3 for both 3- and 4-pyridylmethyl phosphate. [b] $T_E = T_M = 2.00 \times 10^{-3}$ M, where T_E and T_M stand for the total molar concentration of the substrate species and of the metal ion, respectively. [c] k_0, spontaneous hydrolysis rate.

Table V. Effects of the Copper Ion and the Copper Chelates on the Hydrolysis of 2-Pyridylmethyl Phosphate at 90.0 ± 0.10° and μ = 0.10[a]

$-\log [H^+]$[b]	Ligand[c]	Supporting electrolyte	$k \times 10^4$, sec^{-1}	k/k_0
4.93[d-f]		KNO_3	1.19	18
4.72[d-f]		KNO_3	1.19	15
4.36[e,f]		KNO_3	1.19	11
4.36[f]	Tiron	KNO_3	0.314	3.0
4.15[e]		KNO_3	0.650	5.1
4.15	NTA	KNO_3	0.122	0.97
4.14	Tiron	KNO_3	0.306	2.38
3.97		KNO_3	0.422	2.85
3.95	Tiron	KNO_3	0.286	1.91
3.76		KCl	0.276	1.65
3.34		$NaClO_4$	0.233	1.16
2.68		$NaClO_4$	0.225	1.00

[a] $T_E = T_M = 2.00 \times 10^{-3}$ M unless otherwise noted. [b] Variation within ±0.02. [c] $T_M/T_L = 1$ where T_L stands for the total molar concentration of the ligand. [d] $T_E = T_M = 1.00 \times 10^{-3}$ M. [e] Precipitates were produced in the course of reaction. [f] Deviation from the first-order-rate plots was significant.

Table VI. Effects of the Copper Ion on the Hydrolysis of 2-Pyridylmethyl Phosphate at 90.0 ± 0.1° and μ = 0.10 (KNO_3)[a]

T_M/T_E	$-\log [H^+]$[b]	$k \times 10^4$, sec^{-1} [c]	k/k_0
2[d,e]	4.37	1.28	12
1.5[d,e]	4.37	0.89	8.5
1[e]	4.36	1.19	11
0.75[e]	4.36	0.833	7.9
0.5	4.38	0.528	5.1
0.25	4.36	0.283	2.7
0.1	4.37	0.172	1.64
0	4.37	0.106	1

[a] $T_E = 2.00 \times 10^{-3}$ M unless otherwise noted. [b] Variation within ±0.02. [c] First-order approximation in the initial stage. [d] $T_E = 1.00 \times 10^{-3}$ M. [e] Precipitates were produced in the course of reaction.

Table VI shows the dependence of the copper catalysis on the metal ion concentration at a constant hydrogen ion concentration with various molar ratios of substrate to metal ion. Here again, the hydrolysis did not follow the strict first-order rate law, particularly at higher copper ion concentrations. Nevertheless, the apparent rate constant increases markedly as the concentration of the copper ion was raised.

Hydrolysis of Phenyl Phosphate. The hydrolytic behavior of phenyl phosphate was studied at 80° and the ionic strength of 0.10 in aqueous media in the absence of, and in the presence of, metal ions. The observed and calculated first-order rate constants in the absence of metal ions at different $-\log [H^+]$ values are listed in Table VII, together with the assigned rate constant of each ionic form of the phosphate. The spontaneous hydrolysis of phenyl phosphate has been investigated by some workers[13-15] in some detail. Vernon and his coworkers[15] pointed out that the monoanion was the most labile and the neutral species was much less reactive at 100°. The hydrolysis of phenyl phosphate was reinvestigated here under conditions relevant to the present study, and the result was in a satisfactory agreement with those of the previous work.

Table VII. Apparent First-Order Rate Constants for the Spontaneous Hydrolysis of Phenyl Phosphate at 80.0 ± 0.1° and μ = 0.10

$-\log [H^+]$[a]	$k_0 \times 10^5$, sec^{-1}	$k_{calcd} \times 10^5$, sec^{-1} [d]	Supporting electrolyte
1.03	1.39	1.29	$HClO_4$
1.50	2.10	2.09	$HClO_4$-$NaClO_4$
2.18	2.76	2.78	$HClO_4$-$NaClO_4$
3.40	3.02	3.02	$NaClO_4$
3.70	3.02	3.02	KNO_3
4.76[b]	2.90	2.89	KNO_3
5.48[c]	2.36	2.37	KNO_3

[a] Variation of $-\log [H^+]$ during the reaction: ±0.01 unless otherwise stated; [b] ± 0.03; [c] ± 0.02. [d] Calculated rate constant by using: $k_{HA} = 3.06 \times 10^{-5}$ sec^{-1}, $pK_{H_2A} = 1.16$, $pK_{HA} = 6.02$; where k_{HA}, K_{H_2A}, and K_{HA} stand for the specific rate constant of the monoionic species and first and second acid dissociation constants of the phosphate, respectively.

The same experimental conditions as in the spontaneous hydrolysis were employed to study the metal ion catalysis, and the results are shown in Table VIII. It

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Table VIII. Effects of Metal Ions on the Hydrolysis of Phenyl Phosphate at 80.0 ± 0.1° and μ = 0.10[a,b]

Metal ion	$-$Log [H$^+$]	$k \times 10^5$, sec^{-1}	$k_0 \times 10^5$, sec^{-1}	$(k/k_0) \times 100$
Cu^{2+}	3.62 ± 0.01	2.97	3.02	98.3
Zn^{2+}	4.05 ± 0.02	2.99	3.01	99.4
Co^{2+}	4.55 ± 0.02	2.92	2.94	99.2
Ni^{2+}	4.56 ± 0.03	2.92	2.94	99.2
Mg^{2+}	4.61 ± 0.03	2.88	2.93	98.2

[a] $T_E = T_M = 2.00 \times 10^{-3}$ *M*. [b] Supporting electrolyte: below $-\log$ [H$^+$] = 2.67, $HClO_4$-$NaClO_4$; above $-\log$ [H$^+$] = 2.67, KNO_3.

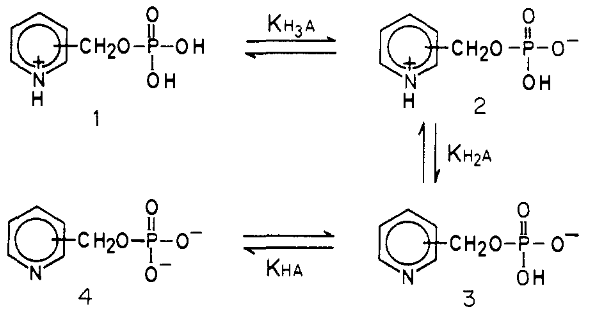
should be noted that all the bivalent metal ions used in this work tend to retard the hydrolysis rate.

Discussion

Spontaneous Hydrolysis. Previous hydrolysis studies[2-4] indicate that the monoionic form is in general the most reactive species among others of monoalkyl and of monoaryl phosphates in the intermediate pH range and the reaction proceeds through phosphorus-oxygen bond fission.

Pyridylmethyl phosphates were found to involve the dissociation process[9] shown in Scheme I. Thus, from the previous knowledge, both the neutral zwitterion form **2** and the monoionic form **3** are to be quite susceptible to hydrolysis, since both species carry the monoionic phosphate group in their structures. In fact, the total rate curve C in Figure 3 is apparently composed of two bell-shaped rate curves, each of which can be attributed to one of the above ionic forms.

Scheme I. Acid Dissociation Process for Pyridylmethyl Phosphate



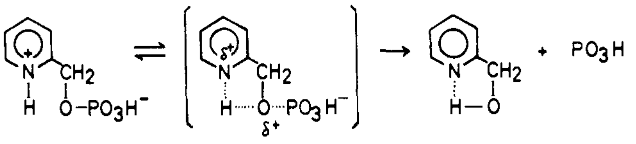
Benzyl phosphate, which is in a structural analogy to pyridylmethyl phosphates, did not demonstrate a bell-shaped rate profile and the neutral species was found to be as reactive as the monoionic species in hydrolysis reaction.[16] If this were the case in the present study, the reactivity of the protonated species **1** and of the zwitterion form would be in the same order of magnitude. The kinetic behavior of all the pyridylmethyl phosphates is quite different from what would be expected, and the reactivity of the protonated species is much less than the corresponding monoionic forms, **2** and **3**. This has been observed for an ordinary alkyl phosphate, such as methyl phosphate.[17] Relative rates at 90° and Arrhenius parameters for the hydrolysis of the monoionic species of some representative aliphatic phosphates are summarized together with the present results in Table IX. Irrespective of the phosphate species, E_a values are approximately 30 kcal mol^{-1} and A values are in the same order of magnitude with the exception of glycerol 2-phosphate. Thus, the zwitterion species of the present phosphates follow the same hydrolytic pathway as expected for the monoionic form of ordinary aliphatic phosphates. The reactivity of the zwitterion is much greater than that of the corresponding monoionic species in the present study and this is much pronounced for 2-pyridylmethyl phosphate. If the inductive effect of the pyridinium nitrogen were the single cause of this difference even for 2-pyridylmethyl phosphate, the zwitterion of this phosphate would not be more labile in hydrolysis than that of the 4 isomer; the pyridinium nitrogen appears to demonstrate its enhanced electronic effect at both 2 and 4 positions to the same extent.[18] As a result, some other hydrolytic mechanism must be operative for the 2-phosphate. The reaction pathway shown in Scheme II may provide the reasonable explanation to this anomalous reactivity of the 2-phosphate. The intramolecular catalysis in terms of proton-transfer mechanism has been proposed by Bender and Lawlor[19] for the elucidation of the profound hydrolytic tendency of the diionic species of salicyl phosphate (Scheme III). In their explanation, the last phosphate proton is transferred intramolecularly to the carboxylate group 5 and then finally to the ester oxygen. On the other hand, in the present proposal, the phosphate proton remains untransferred and the pyridinium proton is intramolecularly transferred to the ester oxygen, so that the reaction is facilitated. Although additional support to this prosposed mechanism is not provided, no other reasonable explanation for the enhanced reactivity of the zwitterion form of 2-pyridylmethyl phosphate seems to be available at present.

Table IX. Reactivity of the Monoionic Species ($ROPO_3H^-$) in Hydrolysis

R	Rel rate[a] at 90°	E_a, kcal mol^{-1}	$A \times 10^{-12}$ sec^{-1}	Ref
CH_3	1.00	30.6	6.47	17
$CH_2OHCH_2OHCH_2$	1.74	29.9	4.28	*b*
$(CH_2OH)_2CH$	3.53	30.3	15.2	*b*
$H_3N^+CH_2CH_2$	4.13	29.6	6.71	*b*
α-D-Glucose-1	1.78	30.0	5.04	*c*
2-$(C_5H_5N^+H)CH_2$	9.27	28.2	2.24	*d*
2-$(C_5H_5N)CH_2$	1.64			*d*
3-$(C_5H_5N^+H)CH_2$	3.85	29.3	4.09	*d*
3-$(C_5H_5N)CH_2$	1.71			*d*
4-$(C_5H_5N^+H)CH_2$	3.92			*d*
4-$(C_5H_5N)CH_2$	1.89			*d*

[a] Rate constants at 90° were calculated by the aid of activation energy for those cited from the literature. [b] P. A. T. Swoboda and E. M. Crook, *Biochem. J.*, **59**, xxiv (1955). [c] C. A. Bunton, D. R. Llewellyn, K. G. Oldham, and C. A. Vernon, *J. Chem. Soc.*, 3588 (1958). [d] Present work.

Scheme II



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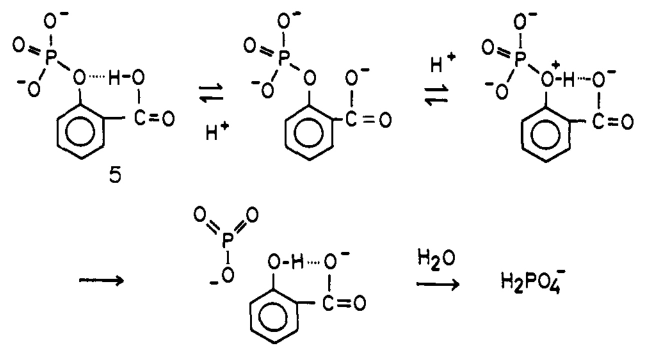
(16) J. Kumamoto and F. H. Westheimer, *J. Am. Chem. Soc.*, 77, 2515 (1955).

(17) C. A. Bunton, D. R. Llewellyn, K. G. Oldham, and C. A. Vernon, *J. Chem. Soc.*, 3374 (1958).

(18) E. Klingsberg, Ed., "Pyridine and its Derivatives," Part I, Interscience Publishers, New York, N. Y., 1960, pp 12, 16.

(19) M. L. Bender and J. M. Lawlor, *J. Am. Chem. Soc.*, **85**, 3010 (1963).

Scheme III



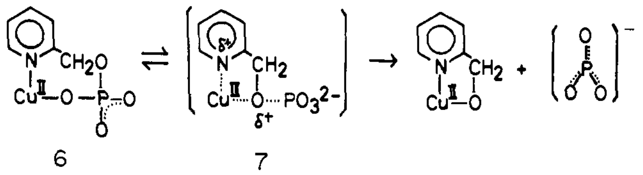
Copper Ion Catalysis. The copper ion demonstrated a profound catalytic effect on the hydrolysis of 2-pyridylmethyl phosphate, while other bivalent metal ions employed in this work were observed to be without effect on the hydrolysis rate. Since 3- and 4-pyridylmethyl phosphate did not suffer any significant catalytic effect of copper(II) on their hydrolysis, the above catalysis is most likely due to the formation of a particular metal complex of the substrate in its reaction pathway. Our previous investigation[20] on the interaction of some bivalent metal ions with 2-pyridylmethyl phosphate indicates that the copper ion forms a 1:1 complex, whose stability is much higher than the corresponding complexes of other bivalent metal ions, *i.e.*, magnesium, manganese, cobalt, nickel, and zinc, and structure **6** with a seven-membered chelate ring was presented for the copper complex. On the other hand, those other bivalent metal ions undergo interaction only with the phosphate moiety.

In the hydrolysis of phenyl phosphate, all the bivalent metals did not promote the reaction rate and even acted slightly as a negative catalyst as shown in Table VIII. In this case, the bivalent metal ions have no other choice than interact with the phosphate moiety.

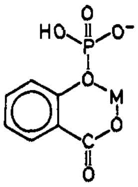
The above results suggest that bivalent metal ions do not demonstrate any catalytic effect on the hydrolysis of organic phosphates whenever they interact only with the phosphate group of substrates.

2-Pyridylmethanol, the hydrolysis product of 2-pyridylmethyl phosphate, produces very stable complexes with copper(II) upon liberation of the alcoholic proton at 1:1 and 2:1 molar ratios of ligand to metal ion,[21] while other bivalent metal ions employed in this study do not show such interaction to any noticeable extent. Thus, the copper ion has a profound affinity toward the alcoholic group of 2-pyridylmethanol. Judging from these results, it seems quite reasonable to propose the formation of activated complex **7** in the hydrolytic pathway of 2-pyridylmethyl phosphate, where the copper ion plays a role of catalyst. In conclusion, the reaction mechanism shown in Scheme IV may provide the reasonable elucidation for the marked catalytic effect of copper(II) on the hydrolysis of the phosphate of interest.

Scheme IV



The solvolysis of salicyl phosphate in the presence of metal ion may provide another interesting aspect of metal ion catalysis.[6,7] The copper, uranyl, vanadyl, zirconyl, and ferric ions demonstrated catalytic effect in this order on the hydrolysis of salicyl phosphate. Formation of copper complex **8** in the course of reaction was suggested as a possibility.[7] As a matter of fact, both iron(III) and copper(II) are known to form the corresponding stable complexes with salicylic acid.[22] Therefore, it is quite adequate to propose the formation of activated complex similar to structure **8** in the reaction pathway.[23] By referring to reaction mechanisms involved in the spontaneous hydrolysis, effective metal ions are to act as pseudo-protons in the catalytic solvolysis of both 2-pyridylmethyl phosphate and salicyl phosphate.



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[20] Y. Murakami and M. Takagi, *J. Phys. Chem.*, **72**, 116 (1968).

[21] Y. Murakami and M. Takagi, *Bull. Chem. Soc. Japan*, **38**, 828 (1965).

[22] L. G. Sillen and A. E. Martell, "Stability Constants of Metal-Ion Complexes," Special Publication No. 17, The Chemical Society, London, 1964.

[23] Professor A. E. Martell of Texas A&M University suggested this sort of hydrolysis mechanism in his plenary lecture on "Catalytic Effects of Metal Chelate Compounds," at the Tenth International Conference on Coordination Chemistry, Nikko, Japan, Sept 12-16, 1967.