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Solvolysis of Organic Phosphates. VI. Intramolecular Catalysis in the Hydrolysis of 2-Pyridylalkyl Phosphates[1]

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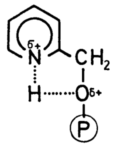
The spontaneous hydrolysis of 2-pyridylethyl, 2-pyridylpropyl, and 6-methyl-2-pyridylmethyl phosphates was investigated at 80—95°C in aqueous media with an ionic strength of 0.10. The reactions followed apparent first-order kinetics with respect to the phosphate species, and the maximum rates were observed at $-\log[H^+]$ 2—3 with a shoulder around 5—6. The participation of intramolecular catalysis played by the pyridinium group in the hydrolysis of the above pyridylalkyl phosphates as well as in that of 2-pyridylmethyl phosphate was investigated from the LFER viewpoint. For the evaluation of LFER, the acid dissociation constant was employed as the most reasonable structural parameter. As a result, the intramolecular pyridinium catalysis was found to takes place in the zwitterion hydrolysis for 2-pyridylmethyl, 2-pyridylethyl, and 6-methyl-2-pyridylmethyl phosphates. This indicates the importance of the geometry around the reaction center for the intramolecular catalytic action. The isokinetic relationship was established for the hydrolysis of pyridylalkyl phosphates with the exception of 6-methyl-2-pyridylmethyl phosphate. The anomalous behavior in the hydrolysis of the latter was attributed to the activation entropy effect which may have been caused by defreezing of solvation in the transition state.

We have investigated the extent of intramolecular catalysis in the hydrolysis of aryl and alkyl phosphates containing a hetero-aromatic ring system around the reaction center. An intramolecular general acid catalysis has been confirmed in the spontaneous hydrolysis of 2-pyridylmethyl,[2] 8-quinolyl,[3] and 3-hydroxy-2-pyridylmethyl phosphates.[4] From the results so far obtained a question arose as to whether acidity of the catalytic group or the geometry around the reaction center contributes to the intramolecular catalysis in the hydrolysis.

In this work, we intend to clarify the effect of structural environment on the intramolecular catalysis by studying the hydrolysis of 2-pyridylalkyl phosphates. If both 2-pyridylethyl and 2-pyridylpropyl phosphates secure the rate enhancement in their hydrolyses due to the intramolecular catalysis by the pyridinium group as in the case of 2-pyridylmethyl phosphate **1**, a six-membered ring **2** and a seven-membered one **3**, respectively, would be formed in their transition states.



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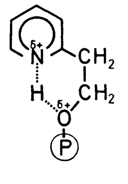

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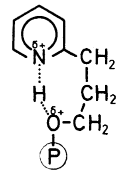

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The stability of these ring conformations yielded in the hydrolysis of the above three phosphates will be discussed in connection with the intramolecular catalysis. The electronic and steric effects due to the methyl-substituent on the intramolecular-proton transfer mechanism were investigated in the hydrolysis of 6-methyl-2-pyridylmethyl phosphate.

Experimental

Materials. 2-Pyridylethyl and 2-pyridylpropyl phosphates were prepared by the phosphorylation of the corresponding alcohols with pyrophosphoric acid.

Pyridylethanol (2-(β-hydroxyethyl)pyridine) (CP grade, Wako Pure Chemical Ind., Ltd., Osaka) was purified by vacuum distillation. Fractions distilled at 66.5°C/0.20—0.25 mmHg were combined and used for the phosphorylation. Pyridylpropanol (2-(γ-hydroxypropyl)pyridine) (Schuchard Chemische Fabrik, München) was purified by distillation, the distillates at 95°C/0.35 mmHg being used for the phos-

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[1] Contribution No. 269 from the Department of Organic Synthesis, Faculty of Engineering, Kyushu University.

[2] Y. Murakami and M. Takagi, *J. Amer. Chem. Soc.*, **91**, 5130 (1969).

[3] Y. Murakami and J. Sunamoto, This Bulletin, **44**, 1939 (1971).

[4] Y. Murakami, J. Sunamoto, and H. Ishizu, *ibid.*, **45**, 590 (1972).

phorylation. Phosphorylation of both pyridylalcohols was carried out according to the previous method.[5,6] Phosphorylated products were purified by the cation exchange column-chromatographic technique and further purified by recrystallization from aqueous ethanol.

2-Pyridylethyl Phosphate. Found: C, 40.58; H, 5.11; N, 6.78; P, 14.3%. Calcd for $C_7H_{10}NO_4P$: C, 41.34; H, 4.96; N, 6.90; P, 15.3%. IR[7] (KBr disk): ν_{O-H}, ~3200; $\nu_{P=O}$, 1254, 1156, and 1075; $\nu_{P-O-C(alk)}$, 1050, 1030, and 933 cm^{-1}. UV ($10^{-4}M$ in water): λ_{max} 262 mμ (ε=7140). Neutralization equivalence; 102.15 (calcd., 101.57).

2-Pyridylpropyl Phosphate. Found: C, 44.01; H, 5.51; N, 6.33%. Calcd for $C_8H_{12}NO_4P$: C, 44.25; H, 5.57; N, 6.45%. IR[7] (KBr disk); ν_{O-H}, ~3400; $\nu_{P=O}$, 1295, 1280, 1175, 1158, and 1080; $\nu_{P-O-C(alk)}$, 1045, 1030, and 920 cm^{-1}. UV ($10^{-4}M$ in water): λ_{max} 263 mμ (ε=7760). Neutralization equivalence; 107.72 (calcd., 108.58).

Preparation and purification of 2-pyridylmethyl[5] and 6-methyl-2-pyridylmethyl phosphates[6] were described previously.

Kinetic and Potentiometric Measurements. The apparatus and experimental procedures were essentially the same as those described previously,[2-4] except for the hydrolysis of 2-pyridylpropyl phosphate at 85, 90, and 95°C, and of 2-pyridylethyl phosphate at 95°C. For the hydrolysis of 2-pyridylpropyl and 2-pyridylethyl phosphates at higher temperatures, a 10 ml Pyrex test tube with a ground Pyrex stopper was used. An aliquot of sample solution containing a substrate, an inorganic salt to maintain the ionic strength constant, and perchloric acid to adjust the hydrogen ion concentration at an appropriate value was placed in the test tube. Several test tubes containing a sample solution of the same constituents were dipped in a water bath regulated to a given temperature. At an appropriate time interval one of them was drawn out and the sample was analyzed for inorganic phosphate liberated during the course of reaction. Since all runs using the Pyrex test tubes were carried out to cover a pH range 1—3 where the phosphate exists predominantly in a neutral zwitterion form, the pH-deviation during the course of hydrolysis reaction did not exceed ±0.02. The observed rate constant in each run was retained within an accuracy of ±2% while the reaction temperature was controlled within ±0.1—0.05°C.

Acid Dissociation Constants and Specific Rate Constants. Acid dissociation constants of the present phosphates at 80°C were obtained by the potentiometric determination[2] and/or by the dynamic (kinetic) method. Specific rate constants assigned to the reactive substrate species were calculated from the apparent first-order rate constants obtained at various pH values. The calculation procedures were described previously.[3,4]

Results

Acid Dissociation Constants. Acid dissociation constants obtained are summarized in Table 1. The acid dissociation constant for the 1-pyridinium proton is represented by K_{H_2A}, and that for the second phosphate proton by K_{HA}.

TABLE 1. ACID DISSOCIATION CONSTANTS OF THE PYRIDYLALKYL PHOSPHATES AT μ=0.10 (KNO_3) AS DETERMINED BY POTENTIOMETRIC TITRATION[a]

Phosphate	pK_{H_2A}		pK_{HA}	
	25°C	80°C	25°C	80°C
6-Methyl-2-pyridylmethyl	4.74[b]	4.50 (4.34)[f]	6.64[b]	6.36 (6.08)[f]
2-Pyridylmethyl	4.42[c]	4.15[d] (4.03)[e,f]	6.29[c]	6.54[d] (6.46)[e,f]
2-Pyridylethyl	5.31	4.84	6.83	6.79
2-Pyridylpropyl	5.60	5.09	6.90	6.75
3-Pyridylmethyl	4.86[c]	4.43[d]	6.23[c]	6.48[d]

a) Estimated error is within 0.02. Values in parentheses were evaluated by kinetic method using Eq. (1).
b) Cited from Ref. 6.
c) Cited from Ref. 5.
d) Cited from Ref. 2.
e) Cited from Ref. 3.
f) At 90°C.

Introduction of a methyl group at the 6-position of 2-pyridylmethyl phosphate resulted in an increase of both pK_{H_2A} and pK_{HA}. Similarly, the extension of the methylene-chain from 2-pyridylmethyl through 2-pyridylpropyl group brought about the lowering of acidity of both groups. Since the temperature dependency of the ionization constant varies according to the nature of the structural environment of the acid, the above trend would not necessarily be the case at a higher temperature.

Specific Rate Constants. All runs followed good first-order kinetics. The pH-rate profile for the hydrolysis of 6-methyl-2-pyridylmethyl phosphate at 90°C is illustrated in Fig. 1 as a typical example. The rate maximum in the pH-rate profile was observed at $-\log[H^+]$ 2—3. This indicates that the most reactive species is in the neutral zwitterion form followed by the monoanionic one. This holds true also for the cases of 2-pyridylethyl and 2-pyridylpropyl phosphates.

The solid line in Fig. 1 is the theoretical curve calculated from the following equation:

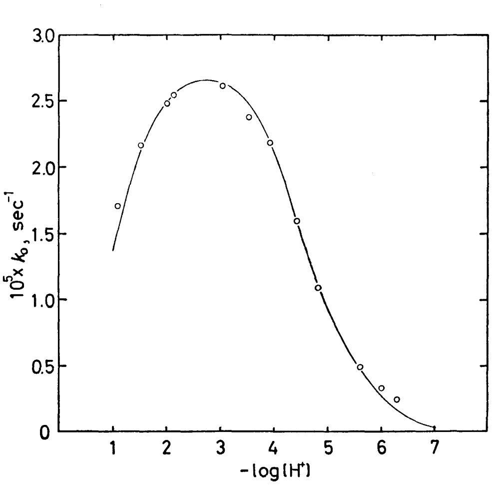

Fig. 1. pH-rate profile for the spontaneous hydrolysis of 6-methyl-2-pyridylmethyl phosphate at 90°C and μ=0.10. Solid line is a theoretical curve calculated from values listed in Tables 1 and 3 by using Eq. (1).

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5) Y. Murakami, M. Takagi, and H. Nishi, This Bulletin, **39**, 1197 (1966).

6) Y. Murakami, J. Sunamoto, H. Sadamori, H. Kondo, and M. Takagi, *ibid.*, **43**, 2518 (1970).

7) Assignments of infrared absorption bands were made by referring to: J. Nakayama, *Yuki Gosei Kagaku Kyokai Shi*, **25**, 132 (1971).

$$k_0 = k_{H_2A}X_{H_2A} + k_{HA}X_{HA} \qquad (1)$$

where X_{H_2A} and X_{HA} stand respectively for the mole fractions of neutral zwitterionic and monoanionic species, and k_0, k_{H_2A}, and k_{HA} are the overall first-order rate constant and the specific rate constants, respectively, of the above two reactive phosphate species. The calculation procedures of these parameters were the same as those described previously.[2–4] The agreement between the observed and the calculated rate constants was quite satisfactory.

TABLE 2. APPARENT FIRST-ORDER RATE CONSTANTS FOR THE SPONTANEOUS HYDROLYSIS OF PYRIDYLALKYL PHOSPHATES UNDER VARIOUS CONDITIONS[a)]

Temp °C[b)]	$-\log[H^+]$[c)]	$k_{obs}\times 10^5$, sec^{-1}[d)]	$k_{cal}\times 10^5$, sec^{-1}[e)]
6-Methyl-2-pyridylmethyl phosphate			
90.0	1.09	1.71	1.53
	1.53	2.17	2.12
	2.01	2.48	2.49
	2.15	2.54	2.55
	3.03	2.61	2.63
	3.53 (±0.03)	2.38	2.47
	3.92	2.19	2.21
	4.42 (±0.04)	1.60	1.63
	4.84	1.09	1.09
	5.60	0.481	0.529
	6.01 (±0.03)	0.337	0.341
	6.30	0.252	0.225
85.0	3.57	1.41	1.39
	3.79	1.29	1.31
	5.82	0.208	0.208
80.0	3.75	0.727	0.754
	3.48	0.784	0.802
2-Pyridylethyl phosphate			
95.0	1.91	0.986	0.997
90.0	2.01	0.582	0.582
	1.92	0.591	0.591
85.0	2.07	0.325	0.326
	1.99	0.320	0.320
2-Pyridylpropyl phosphate			
95.0	1.99	0.543	0.621
	1.98	0.535	0.619
	2.96	0.672	0.673
90.0	1.87	0.331	0.329
	1.89	0.331	0.331
	2.98	0.404	0.367
	2.99	0.413	0.367
85.0	2.96	0.205	0.210
	2.93	0.196	0.196

a) The initial concentration of the phosphates was 2.0×10^{-3} M in all runs. The ionic strength was maintained at 0.10 with $HClO_4$-$NaClO_4$ at a pH range lower than 3.5 and with KNO_3 at a range higher than 3.5.

b) Controlled within ±0.1°C.

c) Variation was maintained within ±0.02 except for the cases cited in parentheses.

d) Standard deviations in individual rate constants do not exceed ±2%.

e) Calculated from the experimental data by the aid of Eq. (1) and the p*K*-values listed in Table 1. Mole fractions of ionic species at 80—95°C were calculated with the use of the p*K*-values obtained at 80°C, except for the hydrolysis of 6-methyl-2-pyridylmethyl phosphate at 90°C where the specific rate constants and the p*K*-values were simultaneously obtained by dynamic procedures.

TABLE 3. THE SPECIFIC RATE CONSTANTS OF THE SPONTANEOUS HYDROLYSES FOR THE PYRIDYLALKYL PHOSPHATES[a)]

Temp °C	$k_{H_2A}\times 10^5$, sec^{-1}	$k_{HA}\times 10^6$, sec^{-1}
6-Methyl-2-pyridylmethyl phosphate		
90.0	2.74	5.63
85.0	1.54	2.02
80.0	0.873	0.962
2-Pyridylethyl phosphate		
95.0	1.11	——[b)]
90.0	0.640	——[b)]
85.0	0.350	——[b)]
2-Pyridylpropyl phosphate		
95.0	0.685	——[b)]
90.0	0.374	——[b)]
85.0	0.200	——[b)]

a) Calculated by Eq. (1) with the aid of the acid dissociation constants listed in Table 1; $\mu=0.10$.

b) Because of the extremely low hydrolysis rate, assignment of the rate constant was not performed.

For the hydrolysis of 6-methyl-2-pyridylmethyl, 2-pyridylethyl, and 2-pyridylpropyl phosphates, the apparent first-order rate constants and the reaction conditions are listed in Table 2. The calculated specific rate constants for the present phosphates are summarized in Table 3. We could not obtain reliable specific rate constants for the hydrolysis of 2-pyridylethyl and 2-pyridylpropyl phosphates in their monoanionic forms, for the reaction was extremely slow even at 90°C. However, participation of the intramolecular catalysis in hydrolysis can be studied for the zwitterionic species. Thus, the above data are not necessarily required for the present purpose.

Activation Parameters. Each specific rate constant satisfactorily followed the Arrhenius law. The activation parameters for the hydrolysis of the neutral zwitterion species, along with the data for 3-pyridylmethyl and 2-pyridylmethyl phosphates,[2)] are listed in Table 4.

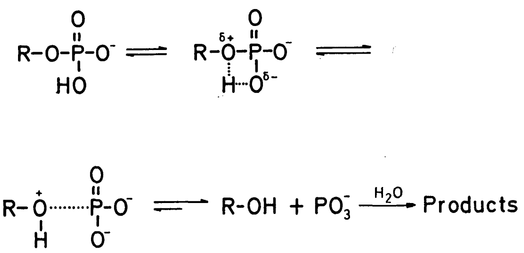
TABLE 4. ACTIVATION PARAMETERS FOR THE HYDROLYSES OF 6-METHYL-2-PYRIDYLMETHYL, 2-PYRIDYLMETHYL, 2-PYRIDYLETHYL, AND 2-PYRIDYLPROPYL PHOSPHATES IN THEIR ZWITTERION FORMS[a)]

Phosphate	E_a kcal mol^{-1}	$\Delta H^{\neq}$ kcal mol^{-1}	$\Delta S^{\neq}$ e. u.
6-Methyl-2-pyridylmethyl	29.1±0.1	28.4±0.1	−1.6±0.3
2-Pyridylmethyl	28.2±0.1	27.5±0.1	−4.3±0.3
2-Pyridylethyl	30.6±0.1	29.8±0.1	−0.5±0.3
2-Pyridylpropyl	32.0±0.1	31.3±0.1	+2.4±0.2
3-Pyridylmethyl	29.3±0.1	28.6±0.1	−3.0±0.3

a) Errors for these parameters were calculated from the standard error of rate constant and errors ascribed to the reaction conditions such as reaction temperature and pH-values.

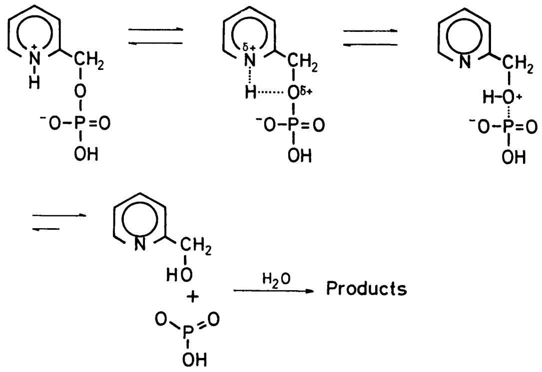
Discussion

Acid Dissociation Constants. The hydrolysis of alkyl and aryl phosphates has been provided with



Scheme 1.

many evidences for an unimolecular P–O bond cleavage upon intramolecular general acid catalysis.[8,9] The state of affairs is shown in Scheme 1 for the monoanionic phosphate species. The extent of intramolecular general acid catalysis can be evaluated by referring to the acid dissociation constant of the corresponding alcohol (the leaving group)[8] and that of the second phosphate proton.[10] The latter, represented by K_{HA}, may reflect the donability of the proton from the phosphate moiety, while the former may be related to the proton acceptability of ester oxygen at the transition state. Both values may refer to the P–O bond strength in the transition state as well.



Scheme 2.

When a nitrogen atom exists in its leaving group as in the hydrolysis of the pyridylalkyl phosphates, the most reactive species is in the neutral zwitterion form and a general acid catalysis occurs intramoleculary with participation of the 1-pyridinium proton[2] as shown in Scheme 2. We understand, therefore, that the acid dissociation constant of ionic substrate is an important structural parameter for a study of the structure-reactivity correlation in its hydrolysis.

Substitution with the electron-donating methyl group at the 6-position of 2-pyridylmethyl phosphate resulted in an increase of pK_{H_2A} and pK_{HA}. The electron donability of the methyl group seems to prevent the dissociation of both protons, on the pyridine nitrogen and on the phosphate oxygen. Similarly, the extension of the methylene-chain in the 2-pyridylalkyl phosphates may result in the increase of basicity of both pyridyl nitrogen and phosphate moiety. These trends on both pK_{H_2A} and pK_{HA} values are obvious from the data given in Table 1.

Intramolecular Catalysis. We have already proposed a mechanism for the intramolecular general acid catalysis in the hydrolysis of 2-pyridylmethyl phosphate[2] and of 8-quinolyl phosphate[3] in their zwitterion forms. The catalysis was played by 1-pyridinium and 8-quinolinium groups, respectively. In the hydrolysis of 3-hydroxy-2-pyridylmethyl phosphate in its monoanion and dianion species, a unique intramolecular general acid catalysis by the 3-hydroxy group has also been clarified.[4] In all cases, the catalytic action has certainly been dependent upon the location of the catalytic site relative to the reaction center. Thus, a functional group of catalytic ability being located sterically in the proximity of ester oxygen, the hydrolysis rate is promoted through intramolecular proton transfer. In order to know whether an intramolecular catalysis takes place also in the hydrolysis of 2-pyridylalkyl phosphates, and to evaluate the extent of catalytic effect if any, we studied LFER on the hydrolysis of pyridylalkyl phosphates in their zwitterion forms. The acid dissociation constant of the second phosphate proton pK_{HA} was considered to be the most reasonable structural parameter for the present LFER study, as obvious from the preceding discussion on this parameter.[10] The linear relationship between pK_{HA} at 80°C and the logarithm of the specific first-order rate constant for the zwitterion hydrolysis at 90°C ($\log k_{H_2A}$) is illustrated in Fig. 2.[11]

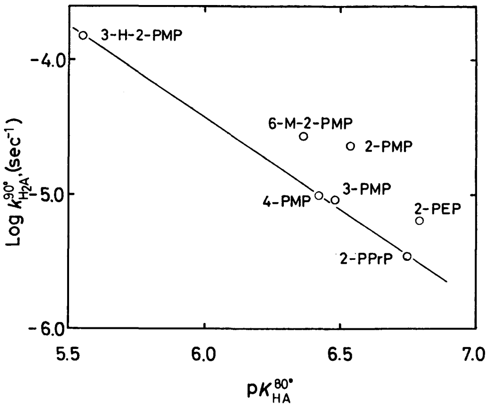


Fig. 2. Correlation between first-order rate constants at 90°C for hydrolyses of pyridylalkyl phosphates in their zwitterion forms and pK_{HA}'s of the phosphates at 80°C. Abbreviations: 3-H-2-PMP, 3-hydroxy-2-pyridylmethyl phosphate; 6-M-2-PMP, 6-methyl-2-pyridylmethyl phosphate; 2-PMP, 2-pyridylmethyl phosphate; 3-PMP, 3-pyridylmethyl phosphate; 4-PMP, 4-pyridylmethyl phosphate; 2-PEP, 2-pyridylethyl phosphate; 2-PPrP, 2-pyridylpropyl phosphate.

Judging from the location of the pyridinium group relative to the ester oxygen, the neutral zwitterions of 3-hydroxy-2-pyridylmethyl,[4] 3-pyridylmethyl, and 4-pyridylmethyl[2] phosphates can not secure an intramolecular catalysis due to the pyridinium group. Thus,

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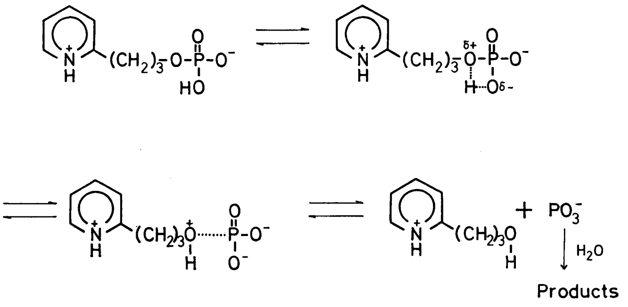
8) T. C. Bruice and S. J. Benkovic, "Bioorganic Mechanisms," Vol. 2, W. A. Benjamin Inc., New York (1966).

9) C. A. Bunton, *Accounts Chem. Res.*, **3**, 257 (1970).

10) Y. Murakami, J. Sunamoto, and N. Kanamoto, presented at the 22nd Annual Symposium on Organic Reaction Mechanisms, Nagoya, October, 1971.

11) Reliabilities of the present linear relationship and of the structural parameters used are scheduled to be discussed in a forthcoming paper along with more extensive investigations.

a straight line can be drawn in Fig. 2 through the plots for these phosphates as a reference for which the reaction proceeds without the pyridinium catalysis. 6-Methyl-2-pyridylmethyl, 2-pyridylmethyl, and 2-pyridylethyl phosphates apparently deviate upward from this line, while 2-pyridylpropyl phosphate lies on it. The significant rate enhancement of the former three phosphates, with reference to those expected from the simple LFER, may be ascribed to some additional catalysis. Thus, it can be seen that the intramolecular general acid catalysis takes place not only in the hydrolysis of 2-pyridylmethyl phosphate but also in that of 2-pyridylethyl and 6-methyl-2-pyridylmethyl phosphates in their zwitterion forms. As a result, we can conclude that formation of either a five-membered (**1**) or a six-membered ring (**2**) at the pre-equilibrium stage of the hydrolysis reaction is in favor of the intramolecular catalysis by the pyridinium group, but that formation of a seven-membered ring (**3**) is no longer favorable for such a catalysis. For the hydrolysis of 2-pyridylpropyl phosphate zwitterion, thus, we may propose a mechanistic pathway *via* an intramolecular proton transfer from the phosphate moiety, not from the pyridinium nitrogen as seen in Scheme 3, analogous to the cases of the usual alkyl phosphates.[8,9]



Scheme 3.

There exists another question as to the extent of their rate enhancement. In a series of 2-pyridylalkyl phosphates, the extents of the rate enhancement are 2.5-fold for $-CH_2-$, 2.0-fold for $-CH_2-CH_2-$, and 1.0-fold for $-CH_2-CH_2-CH_2-$ in their alkyl chains, respectively, relative to the corresponding rates expected from LFER without any additional effect as shown in Fig. 2. In reference to our previous postulate on the reaction mechanism,[2,3] the first energy barrier in this reaction pathway may exist at the stage, where the proton-transfer from the hetero-aromatic nitrogen atom or the terminal phosphate oxygen to the ester oxygen occurs, *viz.*, the pre-equilibrium protonation step. The second barrier may be located in the reaction coordinate at the stage where the P–O bond cleavage takes place to give products; this may most likely correspond to the rate-determining step, as justified by the kinetic solvent isotope effect,[3] and/or by the extent of activation parameters.[2–4,8] This phenomenon is illustrated by a typical schematic representation in Fig. 3. The fact that the linear free energy relationship between pK_{HA} and $\log k_{A_2A}$ was established apparently suggests that the rate-limiting step almost consists of an unimolecular P–O bond fission.

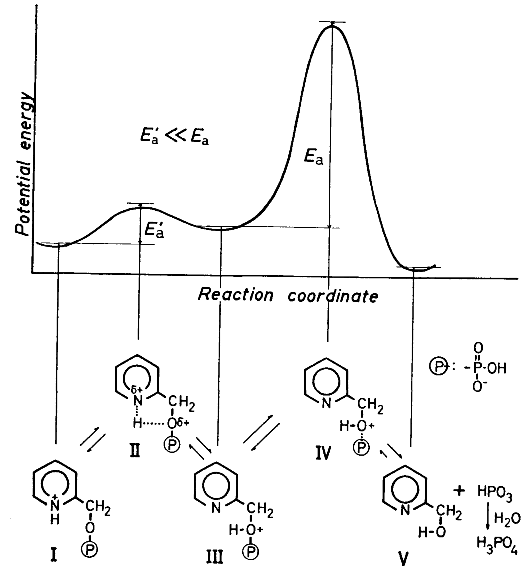


Fig. 3. Schematic representation of unimolecular mechanism in terms of potential energy *vs.* reaction coordinate as a typical intramolecular catalysis acted in the 2-pyridylmethyl phosphate hydrolysis.

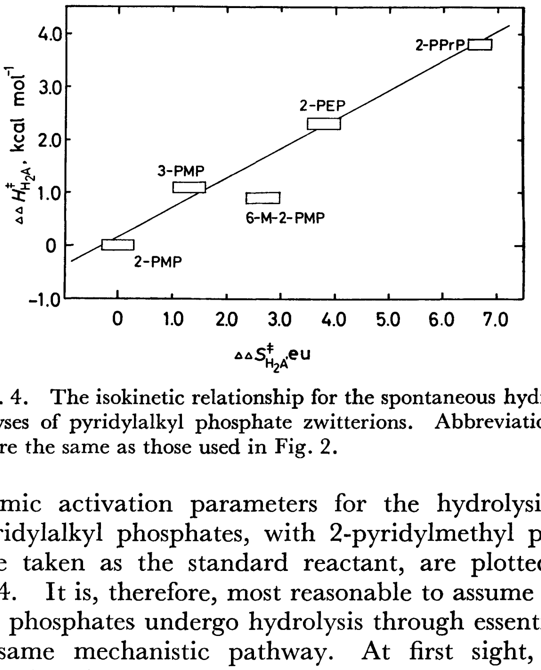


Fig. 4. The isokinetic relationship for the spontaneous hydrolyses of pyridylalkyl phosphate zwitterions. Abbreviations are the same as those used in Fig. 2.

Isokinetic Relationship. The differential thermodynamic activation parameters for the hydrolysis of 2-pyridylalkyl phosphates, with 2-pyridylmethyl phosphate taken as the standard reactant, are plotted in Fig. 4. It is, therefore, most reasonable to assume that these phosphates undergo hydrolysis through essentially the same mechanistic pathway. At first sight, the isokinetic relationship obtained does not appear to be consistent with the mechanistic criteria on the intramolecular proton-transfer path. However, entropy and enthalpy of activation obtained in the present reaction system are mostly concerned with the unimolecular P–O bond fission of the protonated species. It is not necessary to consider from where the proton was transferred at the pre-equilibrium stage. An exception was noticed for the hydrolysis of 6-methyl-2-pyridylmethyl phosphate. The presence of the 6-methyl group seems to perturb the transition state. Contrary to what would be expected, Table 3 and Fig. 2 indicate that the zwitterionic species of 6-methyl-2-pyridylmethyl phosphate is more rapidly hydrolyzed than that of 2-pyridylmethyl phosphate. This was similarly observed in the hydrolysis of monoanionic species; $k_{HA}=5.63\times10^{-3}$

sec^{-1} at 90°C for 6-methyl-2-pyridylmethyl phosphate, and 4.14×10^{-3} sec^{-1} at 90°C for 2-pyridylmethyl phosphate. A question of what is the driving force to bring about the rate enhancement must be solved. A clue can be found in Fig. 4 and Table 4. Only 6-methyl-2-pyridylmethyl phosphate showed deviation downward from the linear relationship of enthalpy-entropy compensation, being different from other pyridylalkyl phosphates. The electron-donating effect of the methyl group must be reflected in terms of activation enthalpy for the hydrolysis of 6-methyl-2-pyridylmethyl phosphate, partially in the pre-equilibrium protonation stage and mostly in the P–O bond cleavage step. The increase of activation enthalpy by 900 cal mol^{-1} was, in fact, observed as an evidence. Thus, the rate-enhancement and deviation in the isokinetic relationship caused by methyl-substitution should be attained from the activation entropy term and therefore the cause of a larger activation entropy ($\Delta\Delta S^{*}=2.7$ e.u.) should be considered. It may be most reasonable to postulate that a bulky alkyl-substituent in the neighborhood of the reaction center interferes significantly with the solvation, *viz.*, "defreezing" of solvation, in the transition state.