Mechanism of the Rapid Alkaline Hydrolysis of a Phosphoramidothioate Ester

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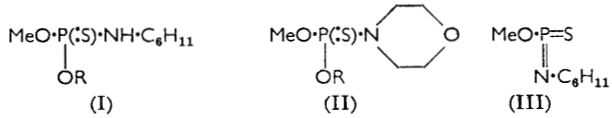
The relative rates and the stereochemistry of alkaline hydrolysis of methyl *p*-nitrophenyl *N*-cyclohexyl-phosphoramidothioate and -phosphoromorpholidothioate are reported. Despite a considerable difference in rate, both reactions show similar stereospecificity. The interpretation of these observations in relation to a metaphosphate-type intermediate in the former case is discussed.

THE intervention of monomeric metaphosphate intermediates in the reactions of phosphoric esters and related compounds has been postulated by many workers to account for the anomalously high rates of solvolysis of certain anionic species.[1-8] Attempts to trap such intermediates unambiguously have met with doubtful success and almost all the evidence for their independent existence is indirect. Since monomeric metaphosphates are expected to be planar (as is the isoelectronic SO_3), investigation of the stereochemistry of a suitable reaction should provide relatively direct evidence. We have investigated the rate and stereochemistry of the alkaline hydrolysis of methyl *p*-nitrophenyl *N*-cyclohexylphosphoramidothioate (Ia) and compared it with the behaviour of methyl *p*-nitrophenyl phosphoromorpholidothioate (IIa) under similar conditions. Whereas alkaline hydrolysis of (Ia) seemed likely (on the basis of other work [9-11]) to proceed through an intermediate (III), that of (IIa) should proceed by a normal $S_N2(P)$ displacement on phosphorus. The



a: R = C_6H_4·NO_2-*p*
b: R = H

optical rotations of the hydrolysis products given in our preliminary Communication [12] (which were determined on very small quantities of material) are incorrect, and the conclusions drawn there require complete revision.

The starting esters (Ia) and (IIa) were prepared by reaction of silver methyl *p*-nitrophenyl phosphorothioate [13] with phosphorus pentachloride to give the corresponding phosphorochloridothioate which was treated immediately with the appropriate amine. Their infrared and n.m.r. spectra were entirely consistent with the proposed structures and showed conclusively that no *S*-methyl esters were present. When the (+)-enantiomorph of the silver salt was used, the specimens of (Ia) and (IIa) were optically active but it was clear in early experiments that a considerable amount of racemisation was occuring (probably in the first stage [14]), as was evident from their wide melting range and changes in optical rotation accompanying recrystallisation. However, we were able to obtain samples which we believe contained 90—100% of one enantiomorph. Since the method of preparation was the same in both cases, the specimens of (Ia) and (IIa) possess the same configuration.

RESULTS

Rates of hydrolysis of (Ia) and (IIa) in dioxan–water (6 : 4) in the presence of a large excess of alkali were determined by following the liberation of *p*-nitrophenoxide ion spectrophotometrically. The release was quantitative (>99%) and showed strict first-order kinetic dependence. At constant ionic strength (0·100) the first-order rate constants were proportional to $[OH^-]$, the rate of neutral hydrolysis being quite negligible, and the second-order rate constants are in the Table, together with those for dimethyl *p*-nitrophenyl phosphorothioate determined under the same conditions.

Alkaline hydrolysis; 10^3k (l. mole^{-1} min.$^{-1}$)

	(Ia)	(IIa)	$(MeO)_2P(:S)\cdot C_6H_4\cdot NO_2$-*p*
At 29·0°	275	3·40	180
At 39·2°	665 (502) *	7·60 (5·65) *	395
At 49·1°	2250	17·5	
$\Delta H^\ddagger$ (kcal. mole^{-1})	15·5 ± 0·3	13·2 ± 0·3	14·1 ± 0·5
$\Delta S^\ddagger$ (e.u.) ...	−17 ± 1	−34 ± 1	25 ± 2

* At ionic strength 0·200.

Molecular rotations (±0·5° at Na D-line) of the hydrolysis products (Ib) and (IIb) (as Na salts) when the hydrolyses of (Ia) and (IIa) were conducted in 50% aqueous dioxan at 40° were: (Ia) (+61·5°) ⟶ (Ib) (+30°); (IIa) (+63°) ⟶ (IIb) (−3·5°). The pure enantiomorphs of (Ib) and (IIb) had $[\phi]_D$ +37° [15] and −6·0°, so the products possess enantiomorph compositions 9 : 1 and 8 : 2, respectively. Despite the difference in sign of $[\phi]_D$, the fact that the products have the same configuration was established by

[1] W. W. Butcher and F. H. Westheimer, *J. Amer. Chem. Soc.*, 1955, **77**, 2420.
[2] F. H. Westheimer, *Chem. Soc. Special Publ.* No. 8, 1957, p. 180.
[3] Lord Todd, *Proc. Chem. Soc.*, 1962, 199.
[4] R. F. Hudson, 'Structure and Mechanism in Organo-Phosphorus Chemistry,' Academic Press, New York, 1965, p. 250.
[5] M. Halmann, A. Lapidot, and D. Samuel, *J. Chem. Soc.*, 1960, 4762; *ibid.*, 1961, 3158.
[6] D. M. Brown and N. K. Hamer, *J. Chem. Soc.*, 1960, 1155.
[7] J. D. Chanley and E. Feageson, *J. Amer. Chem. Soc.*, 1963, **85**, 1181.
[8] A. J. Kirby and W. P. Jencks, *J. Amer. Chem. Soc.*, 1965, **87**, 3209.
[9] D. F. Heath, *J. Chem. Soc.*, 1956, 3796, 3804.
[10] E. W. Crunden and R. F. Hudson, *J. Chem. Soc.*, 1962, 3591.
[11] P. S. Traylor and F. H. Westheimer, *J. Amer. Chem. Soc.*, 1965, **87**, 553.
[12] A. F. Gerrard and N. K. Hamer, *Chem. Comm.*, 1966, 475.
[13] G. Hilgetag, G. Lehmann, A. Martini, G. Schramm, and H. Teichmann, *J. prakt. Chem.*, 1959, **8**, 207.
[14] J. Michalski, M. Micolajczak, A. Halpern, and K. Proszynska, *Tetrahedron Letters*, 1966, 1919.

utilising the reaction of the free acids with aniline described previously;[15] both (−)-(Ib) and (+)-(IIb) gave (+)-methyl hydrogen *N*-phenylphosphoramidothioate. Since this last reaction involved nucleophilic attack on the free acid forms and proceeds at approximately the same rate for both compounds, we are justified in assuming that it proceeds with the same stereochemistry in both cases.

The recoveries of the products (Ib) and (IIb) were 55—70%, being higher in the former case. The loss is mainly attributable to mechanical losses during the purification procedure necessary to obtain completely colourless solutions for the rotation determinations. However, paper chromatography of the initial hydrolysate showed that, in contrast to (Ia) which gave (Ib) exclusively, (IIa) gave 5—10% of other phosphorus-containing products in addition to (IIb). We suggest that these arise from demethylation of (IIa) by the anion of the product (a powerful dealkylating agent) competing with the hydrolysis. In the rate determinations, owing to the much smaller concentration of substrate employed, this bimolecular competition was not observed.

It was established that (IIa) was hydrolysed under the above conditions exclusively by attack on phosphorus, since, when the hydrolysis was performed in $H_2{}^{18}O$ (20% enriched) and the liberated *p*-nitrophenol examined by mass spectrometry, no significant ^{18}O incorporation was found. With sodium methoxide in methanol, however, (IIa) gave 5% of *p*-nitroanisole whereas, even under these conditions, (Ia) gave dimethyl *N*-cyclohexylphosphoramidothioate [15] and *p*-nitrophenol as the sole products.

DISCUSSION

The most striking fact which emerges from the above data is that, despite the considerable difference in rates, both (Ia) and (IIa) undergo alkaline hydrolysis to give products of the same configuration with a high degree of stereospecificity. Although the hydrolysis of (IIa) might appear to be less stereospecific than that of (Ia), this is probably not significant in view of the error in measurement of the small rotation of (IIb); moreover, we cannot be absolutely certain of the optical purity of the specimen of (IIa) but evidence is adduced (see Experimental section) to suggest that it is >90%. This stereospecificity is, of course, quite inconsistent with the intervention of a planar intermediate (III) in the hydrolysis of (Ia).

If we exclude the formation of an ester (III) as an intermediate, the difference in rates of hydrolysis of (Ia) and (IIa) requires explanation. While this difference is less spectacular (a factor of 10^2 at 40°) than the value of the order 10^7 for the hydroxide-ion catalysed hydrolysis of *NN'*-dipropylphosphorodiamidic chloride (IV) and *NNN'N'*-tetramethylphosphorodiamidic chloride (V),[11]

$(PrNH)_2P(:O)Cl$ (IV) $\qquad$ $(Me_2N)_2P(:O)Cl$ (V)

we believe that it is too large to be accounted for on the basis of electronic or steric effects on an $S_N2(P)$ displacement although these can be large.[16] For example, it is clear from the Table that (Ia) hydrolyses appreciably faster than even dimethyl *p*-nitrophenyl phosphorothioate. Moreover, in the case of the halides (IV) and (V) studied by Traylor and Westheimer,[11] nucleophilic attack by water or pyridine [under conditions where the fast base-catalysed hydrolysis of (IV) was negligible] occurred more readily on the secondary base derivative (V). The much lower value of $\Delta S^{\ddagger}$ found for (Ia) is also suggestive of a change of mechanism. Hence we believe that the difference in the rates of alkaline hydrolysis of (Ia) and (IIa) reflects a genuine difference in mechanism and that this is due to essentially similar factors as the much larger difference in hydrolysis rate of (IV) and (V).

The kinetic dependence and the stereochemical evidence are consistent with rate-determining, nucleophilic attack by a water molecule on the anion, formed from (Ia) by removal of the amide proton, *provided* that the pK_a of this proton is more than 14. Although no dissociation constants for phosphoramides of this type are yet available (for this reason the value of $\Delta S^{\ddagger}$ in the Table provides no evidence as to the nature of this transition state) the insolubility of dimethyl *N*-cyclohexylphosphoramidothioate in aqueous alkali suggests that the above condition is satisfied. The high rate of reaction of this anion with water (a weak nucleophile) suggests that the ArO–P bond is abnormally weak and that there is a distinct tendency for unimolecular elimination to occur. However, owing to the poor leaving ability of *p*-nitrophenoxide ion, solvent attack occurs before the elimination is complete. A similar explanation, based on a quite different approach, has been suggested to account for solvolysis of *p*-nitrophenylphosphate di-anions in aqueous solution.[8] Such a mechanism is consistent with the very much larger rate difference of alkaline hydrolysis of (IV) and (V) since unimolecular eliminations are much more sensitive to the nature of the leaving group than are bimolecular displacements. In the case of (IV), the elimination of chloride might be complete before the solvent attack, and a true metaphosphate-type intermediate might be formed. Studies are in progress on the synthesis of an optically active analogue of (Ia), where the *p*-nitrophenoxy-group is replaced by chlorine.

EXPERIMENTAL

Specific rotations were determined for 5—8% solutions in ethanol. Paper chromatograms were run on Whatman No. 1 paper using propan-2-ol–ammonia–water (7 : 1 : 2).

Sodium hydroxide solutions were prepared by dilution of B.D.H. concentrated volumetric solutions with carbon dioxide-free distilled water. Dioxan, purified initially according to the procedure of Vogel,[17] was refluxed with stannous chloride, dried over sodium, and stored under nitrogen in the dark.

Dimethyl *p*-nitrophenyl phosphorothioate, m. p. 35° (lit.,[18] 36°), was obtained by repeated recrystallisation of a commercial sample from methanol at −15°. Dimethyl phosphoromorpholidothioate, prepared by the reaction of

[15] N. K. Hamer, *J. Chem. Soc.*, 1965, 2731.
[16] R. F. Hudson and L. Keay, *J. Chem. Soc.*, 1960, 1859.
[17] A. I. Vogel, 'Practical Organic Chemistry,' Longmans, London, 1948, p. 175.
[18] J. A. A. Ketelaar and H. R. Gersmann, *J. Amer. Chem. Soc.*, 1950, **72**, 5777.

dimethylphosphorochloridothioate with morpholine (2 mol.) in chloroform solution, had b. p. 93°/0·3 mm. (lit.,[19] 80°/0·1 mm.).

(+)-*Silver Methyl* p-*Nitrophenyl Phosphorothioate*.—To a solution of the *N*-methostrychninium salt of (−)-methyl *p*-nitrophenyl hydrogen phosphorothioate (3·0 g.) [20] in hot methanol (210 ml.) was added a solution of silver nitrate (1·7 g.) in hot methanol (60 ml.). The mixture was maintained at reflux temperature for 20 min. while the product crystallised and was filtered hot. After washing with hot methanol (4 × 20 ml.) followed by dry ether (20 ml.), the *product* (1·3 g.) had m. p. 180—181°, $[\alpha]_D$ +62·4° (in MeCN) (Found: C, 23·7; H, 2·3; N, 3·9; P, 8·8. $C_7H_7AgNO_5PS$ requires C, 23·6; H, 2·0; N, 3·9; P, 8·7%).

Methyl p-*Nitrophenyl* N-*Cyclohexylphosphoramidothioate*.—To a stirred suspension of the racemic silver salt (0·89 g.) in dry, alcohol-free chloroform (30 ml.) at 0° was added phosphorus pentachloride (0·52 g.). The mixture was stirred for a further 10 min. at 0° and silver chloride was filtered off. To the filtrate was added dry toluene (50 ml.) and the mixture evaporated to small volume under reduced pressure at room temperature. A further similar addition of toluene, followed by evaporation, was necessary to remove the last traces of phosphoryl chloride, and a pale yellow viscous oil was obtained; without purification this was taken up in dry toluene (20 ml.), to the solution cyclohexylamine (1·0 g.) was added, and the mixture left at room temperature for 3 hr. Dilute hydrochloric acid (20 ml.) was added, and the organic layer separated, and, after washing with water (2 × 20 ml.), dried ($MgSO_4$). Removal of the solvent gave a pale yellow oil which was taken up in warm methanol. On cooling, the *product* was deposited as very pale yellow needles (320 mg.), m. p. 83—83·5° (Found: C, 47·4; H, 5·7; N, 8·5; P, 9·5. $C_{13}H_{19}N_2O_4PS$ requires C, 47·3; H, 5·8; N, 8·5; P, 9·4%).

When the (+)-silver salt (0·89 g.) was used, the same procedure produced a pale yellow oil (600 mg.), $[\alpha]_D$ +17·2°. Recrystallisation from methanol–light petroleum (b. p. 40—60°) gave crystals (370 mg.), m. p. 57·5—58·5°, $[\alpha]_D$ +18·5° (Found: N, 8·8; P, 9·7%).

Several experiments, in particular those on a larger scale, gave products with wider melting ranges and lower specific rotations, but it was possible, in most cases by repeated recrystallisation from the above solvent or methanol–water, to obtain samples identical with the above.

The n.m.r. spectrum in perdeuteriodimethyl sulphoxide showed an AB quartet at τ 1·6, 2·4 (J = 9·0 c./sec.), a quartet at 3·7 (J = 16 c./sec.), a doublet at 6·2 (J = 14 c./sec.), a broad singlet 6·8, and a complex multiplet at 7·95—8·9; the integrated intensities were in the ratio 4 : 1 : 3 : 1 : 10. Shaking with D_2O resulted in the disappearance of the quartet at τ 3·7. The infrared spectra showed the absence of P=O.

Methyl p-*Nitrophenyl Phosphoromorpholidothioate*.—The previous procedure was followed with morpholine (1·0 g.). From racemic silver salt (0·89 g.), colourless *needles* (370 mg.) were obtained, m. p. 82·5—83° (Found: C, 41·2; H, 4·5; N, 8·4; P, 9·4. $C_{11}H_{15}N_2O_5PS$ requires C, 41·5; H, 4·7; N, 8·8; P, 9·7%).

When the (+)-silver salt (0·89 g.) was used, an oil or pasty solid (700 mg.) was obtained, of very variable m. p. and $[\alpha]_D$ 11—17°. This was taken up in the minimum volume (3 ml.) of hot methanol and the solution cooled to −15°. Racemic material (m. p. 78—82°) crystallised, and removal of the solvent gave an almost colourless oil (300 mg.), $[\alpha]_D$ +20·1° (Found: N, 8·5; P, 9·6%). The n.m.r. spectrum in CCl_4 showed an AB quartet τ 1·7, 2·7 (J = 9·4 c./sec.), a doublet 6·2 (J = 14·6 c./sec.), and a multiplet 6·3—6·8 with integrated intensities in the ratio 4 : 3 : 8.

In a separate experiment the solubility of the racemate in pure methanol at −15° was determined as 20 mg./ml. but the solubility is likely to be lower in the presence of an excess of one enantiomorph, and hence the product should contain 90% of the (+)-enantiomorph.

Methyl Hydrogen Phosphoromorpholidothioate.—To a hot solution of potassium hydroxide (58 g.) in water (340 ml.) was added a solution of dimethyl phosphoromorpholidothioate (80 g.) in hot methanol (660 ml.), and the mixture heated under reflux for 18 hr. After cooling to 0°, the mixture was neutralised by cautious addition of hydrochloric acid, and the resulting solution evaporated to dryness *in vacuo* at 40°. The residual solid was extracted with warm ethanol (3 × 200 ml.) and the combined extracts were evaporated to dryness to give the crude potassium salt (76 g.) of the product. The free acid was prepared by treatment of a solution of the potassium salt (11 g.) in water (25 ml.) with hydrochloric acid (22 ml.; 3N) at 0° followed by extraction with chloroform (5 × 20 ml.). The combined extracts were dried ($MgSO_4$) and concentrated to 10 ml. *in vacuo*. Addition of light petroleum (b. p. 40—60°) gave the *product* as deliquescent plates (5·5 g.), m. p. 111—112° (Found: C, 30·8; H, 5·8; N, 6·8; P, 15·6. $C_5H_{12}NO_5PS$ requires C, 30·5; H, 6·1; N, 7·1; P, 15·7%).

Resolution of Methyl Hydrogen Phosphoromorpholidothioate.—To a stirred suspension of the racemic acid (12 g.) in acetone (120 ml.) was added quinine (*ca.* 18 g.) until a neutral solution was obtained; towards the end of the addition crystals started to separate. Water (60 ml.) was added and the mixture warmed until a homogeneous solution was obtained (filtering was sometimes necessary at this stage to remove small amounts of flocculent material); this was set aside at room temperature, and after 12 hr. the crystalline quinine salt was filtered off and recrystallised from the minimum volume of acetone–water (60 : 20), until the m. p. was constant (205° after drying). The ammonium salt was obtained, by the procedure given earlier,[15] as *needles* (from alcohol–ether), m. p. 113—113·5°, $[\alpha]_D$ = +2·9° (in EtOH) (Found: C, 28·2; H, 7·0; N, 13·2; P, 14·4. $C_5H_{14}N_2O_3PS$ requires C, 28·1; H, 7·0; N, 13·1; P, 14·5%). A sample of the quinine salt was recrystallised twice more from acetone–water, without change in m. p. or optical rotation of the ammonium salt isolated from it.

Reaction of (+)-*Methyl Hydrogen Phosphoromorpholidothioate with Aniline*.—To a solution of the (+)-ammonium salt (200 mg.) in ethanol (2 ml.) was added a solution of aniline hydrochloride (108 mg.) in ethanol (2 ml.). The precipitated ammonium chloride was filtered off and the filtrate evaporated at room temperature, to give the solid anilinium salt. This was taken up in a mixture of freshly distilled aniline (1 ml.) and dry benzene (4 ml.), and the mixture heated at 60° under nitrogen for 15 hr. Addition of water (5 ml.) followed by the removal of benzene and excess of aniline by extraction with ether gave an aqueous solution which was passed through a Dowex 50 column

[19] M. Nguyen Thanh Thuong, C. Clergue, E. Larraut, and P. Chabrier, *Bull. Soc. chim. France*, 1964, **6**, 1407.
[20] G. Hilgetag and G. Lehmann, *J. prakt. Chem.*, 1959, **8**, 224.

(NH_4^+ form). The eluate was evaporated to dryness, to give a colourless gum which was crystallised from ethanol–ether to give ammonium methyl *N*-phenylphosphoramidothioate (112 mg.), m. p. 118—120°, $[\alpha]_D$ +24°, which was identical with an authentic sample.[15]

Alkaline Hydrolysis of (+)-*Methyl* p-*Nitrophenyl* N-*Cyclohexylphosphoramidothioate*.—The sample (200—250 mg.) hydrolysed at 40° in a mixture of dioxan (50 ml.) and sodium hydroxide solution (50 ml. of 1·0M) for 5 hr. under an atmosphere of nitrogen. Solid carbon dioxide was added to reduce the pH below 8 and the solution evaporated to dryness at 40° *in vacuo*. The residue was dissolved in the minimum amount of water, applied to a Dowex AG 1 × 8 anion exchange column (15 × 3 cm.; Cl^- form) and subjected to gradient elution with sodium chloride solution. Methyl hydrogen *N*-cyclohexylphosphoramidothioate was eluted in fractions corresponding to 0·70M-NaCl, and these were combined and evaporated to dryness below 30°. The solid residue was extracted well with warm ethanol (6 × 30 ml.) and the extracts were again evaporated. The final residue was taken up in 2 ml. of dry ethanol, and filtered to remove small traces of sodium chloride, giving a colourless solution which on evaporation gave the crystalline sodium salt (90—110 mg.) (chromatographically identical with an authentic sample), $[\alpha]_D$ +12·8°.

Hydrolysis of the corresponding phosphoromorpholidothioate was conducted under similar conditions with the use of more concentrated sodium hydroxide (70 ml. of 1·5M) and was maintained at 40° for 48 hr. Elution of the product from the anion exchange column occurred at 0·2M-NaCl and, after work-up as before, there was obtained a very hygroscopic sodium salt of the product (chromatographically identical with an authentic sample), $[\alpha]_D$ −1·55°. In view of the very hygroscopic nature of this salt the concentration of the solution used in the rotation measurement was determined from phosphorus content.

Rate Measurements.—Two procedures were used; procedure A being more suitable for the faster rates of hydrolysis and B for the slower. In several cases both procedures were used and gave almost identical values for the rate constants.

Procedure A. To a mixture of dioxan (35 ml.) and water (60 ml.) in a 100-ml. standard flask in a thermostat-bath (containing sufficient NaOH and $NaClO_4$ to give a hydroxide concentration in the range 0·028—0·100M and ionic strength 0·100) was added a dioxan solution of (I) (5 ml. of 3 × 10^{-3}M) and the volume made up to 100 ml. with dioxan (0·5 ml. necessary). After rapid mixing, a sample was transferred with a preheated pipette to a cell placed in a thermostatted block of a Zeiss PMQ II spectrophotometer. The change in optical density at 400 mμ was followed up to 60—70% hydrolysis. Infinity values were obtained (and agreed to within 1% of the calculated values) by withdrawing samples from the flask after approximately eight half-lives and determining the optical density after dilution with same solvent.

Procedure B. The solution (3—5 × 10^{-4}M) was made up in the thermostatted standard flask as before, and aliquot portions (2 ml.) were withdrawn at intervals. These were made up accurately to 10 ml. (20 ml. at the later stages of the reaction) with cold dioxan–water (40 : 60), and optical density was determined immediately.

Hydrolysis of (IIa) *in* ^{18}O-*Enriched Water*.—To clean sodium wire (12·5 mg.) under dioxan (0·7 ml.) was added, slowly with cooling, water (0·6 ml.; 21% enriched in $H_2{}^{18}O$). The resulting clear solution was added to a sample of (IIa) (30 mg.), and the mixture maintained at 40° for 6 hr. Removal of the solvent *in vacuo* gave an oily residue to which was added water and the mixture extracted with chloroform (4 × 5 ml.) to remove unreacted starting material. The aqueous solution was adjusted to pH 5·5 (dilute HCl) and extracted with ether (5 × 5 ml.). The combined extracts were dried and evaporated to dryness, to give colourless crystals of *p*-nitrophenol (6 mg.) which were analysed in an A.E.I. MS9 high-resoltuion mass spectrometer. The relative intensities of the peaks at 139, 140, and 141 were 100, 8·70, and 1·20% (reproducible to ±0·05) as compared with the values 100·0, 8·65, 1·05% for an authentic sample.

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