Biomimetic Study of Intramolecular Phosphoryl Group Transfer in Thioureas under Acid/Base and Stereoelectronic Control

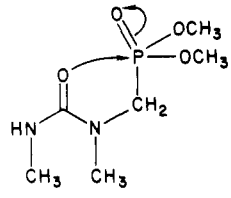
C. Blonski,[1a] M. B. Gasc,[1a] A. F. Hegarty,[1b] A. Klaebe,[1c] and J. J. Perie*[1a]

Contribution from UER-PCA-Universite Paul Sabatier, 31062 Toulouse Cedex, France, and the Department of Chemistry, University College, Belfield, Dublin 4, Ireland.
Received September 27, 1983

Abstract: Reaction of dicyclohexylcarbodiimide with sterically hindered thiophosphoric esters **3** leads at −80 °C to observable (^{31}P NMR) *S*-phosphorylisothioureas **5**. At 20 °C in methylene chloride the same reaction does not give **5** but its *N*-phosphoryl isomer **4**, together with **5′**, **2**, and the counterion **3′**. The equilibrium quantity of the *N*-phosphoryl isomer varies with the bulk of the R group (R = Ph > 2,6-$Me_2C_6H_3$ > 2.5-*i*-$Pr_2C_6H_3$). The equilibrium can also be either approached from **4** (which is isolable) or by treating the isothiouronium salt **5′** with a strong amine base (such as the proton sponge), and in each case the same observed rate constant ($=k_1 + k_{-1}$) is obtained. The equilibrium can be displaced toward **5′** in the presence of acid (HBF_4 or Et_3NH^+) or toward the elimination products **2** and **3′** in the presence of bases (tertiary amines). In the presence of excess acid the observed rate of reaction of **4** is independent of acid concentration and type, and the rate-determining step is assigned to the intramolecular *N*- → *S*-phosphoryl group transfer (**4** → **5**), governed by k_1. In the presence of excess base the rate of reaction of **4** is independent of base type and concentration, and the step which is partly rate determining in this case is consistent with the conversion of **5** to **8** (k_2) which is correctly oriented for elimination. The implications of the observation of this mobile equilibrium governed by the presence of acid or base catalysts to the question of the involvement of *O*- or *N*-phosphoryl intermediates in the mode of action of the coenzyme biotin is briefly discussed.

A two-step mechanism for CO_2 fixation and transfer for biotin dependent carboxylases has been proposed[2] and supported, inter alia, by the isolation[3] of a carboxybiotin intermediate as its methyl ester. However, the detailed mechanism of each of the steps remains unclear. In particular, the method of activation of biotin (and/or CO_2) to permit reaction at the weakly nucleophilic ureido site remains unanswered.

Recently it has been proposed that phosphorylation of biotin may play a key role in its activation,[4] (Scheme I) while phosphorylation of the carbonate group (leading to a mixed carbonic phosphoric anhydride,[5] similar to that described for carbamyl phosphate synthetase[6]) has also been invoked. Apart from the work of Kluger,[7] however, no good models for phosphoryl group transfer to and activation of a ureido group have been reported. Kluger demonstrated, using the phosphonate **1**, that hydrolysis proceeded

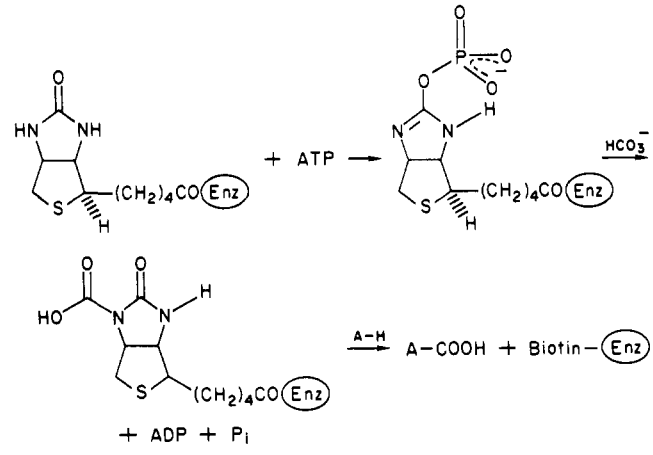


1

via participation of the ureido oxygen, leading to a transitory O-phosphorylated intermediate, a possible model for *O*-phosphobiotin.

We now report on our studies[8] of model systems which provide "*O*-phosphobiotin" analogues. Two possible routes have been investigated: direct O-phosphorylation of urea derivatives and reaction of carbodiimides with phosphoric acid derivatives (which

Scheme I



can lead initially to the same intermediates).[9] However, such adducts cannot be studied directly since two competing rapid reactions occur: intermolecular attack by a second phosphate group to give a pyrophosphate or *O*- → *N*-phosphoryl group rearrangement.

These problems can, however, be overcome by using (a) the thio analogues of phosphoric esters, (b) bulky groups about phosphorous to slow S → N phosphoryl group rearrangment, and (c) trapping of the intermediate isothiouronium form through protonation. We now report on a detailed kinetic study on the intra- and intermolecular phosphoryl group transfer from these analogues. In solution, several equilibria are set up which can be reached by three independent routes. The equilibration position is shown to depend on several factors: concentration, presence of acids and bases, and stereoelectronic control dependent on the substrate conformation.

Results

Since the isothioureas **5** react very rapidly with water, reactions were carried out in dry dichloromethane at 20 °C. Other solvents such as 1,4-dioxane or acetonitrile were also found to be unsuitable, reacting with one or other of the substrates. However, excellent and reproducible kinetics were obtained, starting from either the

(1) (a) ERA au CNRS 264, Université Paul Sabatier, Toulouse; (b) University College, Dublin; (c) ERA au CNRS 926, Université Paul Sabatier, Toulouse.
(2) Wood, H. G.; Barden, R. E.; *Annu. Rev. Biochem.* **1977**, *46*, 385. Wimmer, M. J.; Rose, I. A.; *Ibid.* **1978**, *47*, 1031.
(3) Lynen, F.; Knappe, J.; Lorch, E.; Jutting, G.; Ringelmann, *Angew. Chem.* **1959**, *71*, 481.
(4) Retey, J.; Lynen, F. *Biochem. Z.* **1965**, *342*, 256.
(5) Wimmer, M. J.; Rose, I. A.; Powers, S. G.; Meister, A. *J. Biol. Chem.* **1979**, *254*, 1854.
(6) Powers, S. B.; Meister, A. *J. Biol. Chem.* **1978**, *253*, 1258.
(7) Kluger, R.; Adawadkar, P. D. *J. Am. Chem. Soc.* **1976**, *98*, 3741.
(8) Portion of this work was presented at the 6th IUPAC Conference on Physical Organic Chemistry, Louvain, 1982 (*Bull. Chim. Soc. Belg.* **1982**, *91*, 488). See, also: Blonski, C.; Gasc, M. B.; Klaebe, A.; Perie, J. J. *Tetrahedron Lett.* **1982**, *23*, 2773.
(9) Blonski, C.; Gasc, M. B.; Klaebe, A.; Perie, J. J.; Roques, R.; Declerq, J. P.; Germain, G. *J. Chem. Soc., Perkin Trans. 2* **1982**, 7. Etemad-Moghadam, G.; Klaebe, A.; Perie, J. J. *Phosphorus Sulphur* **1981**, *12*, 61.

0002-7863/84/1506-7523$01.50/0

Table I. Observed Rate Constants for the Reaction of **4**[a]

conditions	$10^3 k_{obsd}$, s^{-1}		
	4a	**4b**	**4c**
excess acid	1.08 ± 0.03	3.18 ± 0.05	0.656 ± 0.020
excess base	0.017 ± 0.001	1.18 ± 0.05	0.554 ± 0.004
neutral		15.8 ± 0.6	2.65

[a] At 20 °C in CH_2Cl_2; initial substrate concentration = 8.87×10^{-5} M.

Table II. Calculated Percentages of Species **4**, **5′**, and **3′** Present at Equilibrium[a]

	% at equilibrium		
	4	**5′**	**3′**
for **4a**	90[b]	5[b]	5[b]
for **4b**	82	9	9
for **4c**	72	14	14

[a] At 20 °C in CH_2Cl_2; [**4**] + [**5′**] = 8.87×10^{-5} M. [b] Estimated from the reduction in optical density at 280 nm.

isothiouronium salt **5′**, the N-phosphorylated isomer **4**, or the carbodiimide **2** and the thiophosphoric ester **3** in dichloromethane.

The reactions were followed by measuring the change in the optical density at 280 nm where the *N*-phosphoryl isomer **4** has its UV maximum; ^{31}P NMR and IR were also used to estimate relative concentrations of the intermediates. Three sets of substrates were used with R = Ph (a), 2,6-$Me_2C_6H_3$ (b), and 2,6-*i*-$Pr_2C_6H_3$ (c) in order to examine the effect of the bulk of R on the inter- and intramolecular reactions.

(i) Equilibrium between S- and N-Phosphorylated Isomers (4 and 5). When the pure N-phosphorylated isomers **4b** and **4c** were introduced rapidly into dichloromethane solution, equilibration with **2**, **3′**, and **5′** occurs. A very small optical density change at 280 nm occurs for **4a**, indicating that this isomerizes to <10% in solution.

The rate constants obtained (k_{obsd}, s^{-1}) for the disappearance of **4b** and **4c** are concentration dependent and quite rapid (see Figure 2). Good first-order plots are obtained in each case, and the reactions show tight isosbestic points, indicating the absence of detectable intermediates. When the molar extinction constants for each species are known, the percentages of each species present at equilibrium (initial concentration of **4**, 8.87×10^{-5} M) can be calculated (see Table II).

The concentration dependence of the equilibrium arises from the partial dissociation to give the carbodiimide **2** and thiophosphate anion **3′**. Note that in all cases [**5′**] = [**3′**] and that the S-phosphorylated free base **5** was not detected at 25 °C. The equilibrating species are noted in eq 1. The variation in the

$$4 \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} [5] \rightleftharpoons 2 + 3 \underset{-5}{\overset{+5}{\rightleftharpoons}} 3' + 5' \qquad (1)$$

observed rate of equilibration of **4c** as a function of concentration is given in Figure 2. In general we have used the same concentration of substrate (8.87×10^{-5} M) for simplicity in the results reported.

(ii) Addition of Acid. When the N-phosphorylated substrate **4** is added to dichloromethane containing a strong acid (tetrafluoroboric acid) in excess, smooth conversion to the protonated S-phosphorylated isomer **5′** occurs. From Figure 1 it is noted that tight isosbestic points are held for this reaction; the kinetics obtained are strictly first order in **4** and independent of its initial concentration and of the concentration of (excess) acid. Moreover, the same observed rate constant is obtained in the presence of a second acid, triethylammonium tetrafluoroborate (in large excess), and is independent of the concentration of the latter (which was varied over the range 50–2000-fold excess). This independence of observed rate constants also indicates that the reaction is relatively insensitive to salt effects, an important feature since no attempt was made in this work to maintain ionic strength constant.

The observed rate constants for the conversion of **4** to **5′** in the presence of acid are summarized in Table I. The same observed rate constants were obtained when either (a) **4** was allowed to equilibrate in solution before the addition of acid or (b) it was added in solid form to CH_2Cl_2 containing the acid.

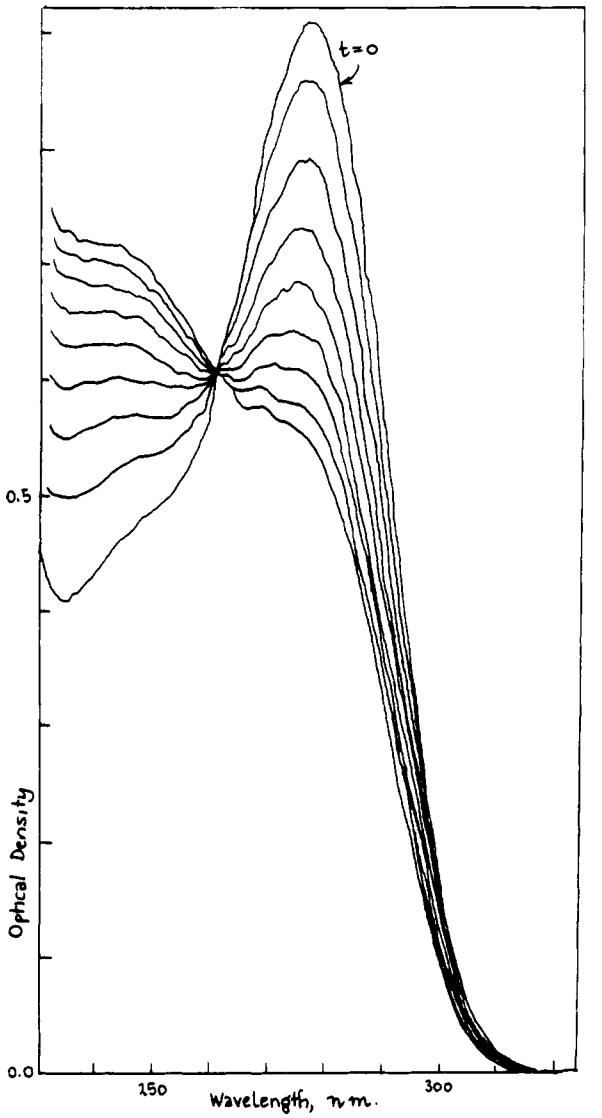


Figure 1. Repetitive scans of the ultraviolet spectrum obtained when **4b** (8.87×10^{-5} M) is treated at 20 °C in CH_2Cl_2 with excess HBF_4.

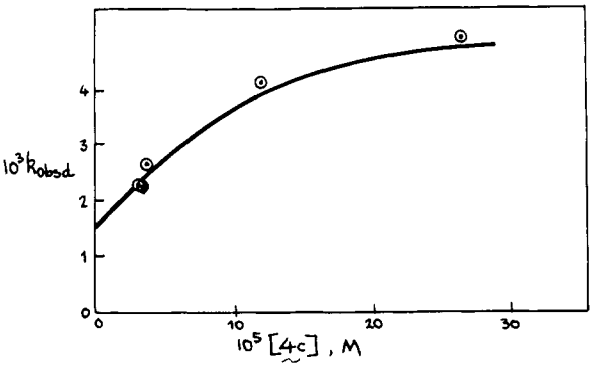


Figure 2. Plot of the observed rate constant (s^{-1}) as a function of the initial concentration of substrate **4c** (in CH_2Cl_2 at 20 °C).

With the weak acid $Et_3NH^+BF_4^-$, at lower concentrations, the rate of formation of **5b′** from **4b** decreases as the concentration of acid is increased before reaching the constant value of 3.18×10^{-3} s^{-1} (see Figure 3).

(iii) Addition of Base. When a tertiary amine base is present in equimolar quantity or in excess then the N-phosphorylated substrate **4** is converted quantitatively to carbodiimide **2** and the thiophosphate anion **3′**. Three different bases were used: tri-

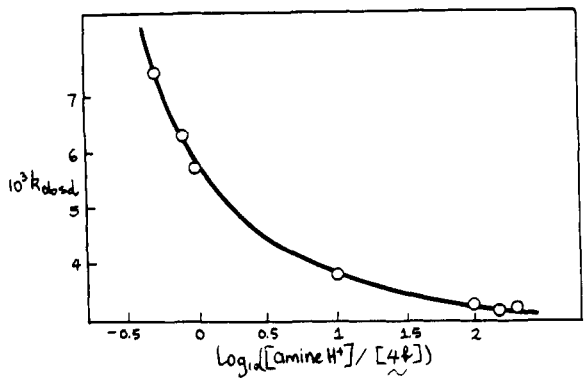


Figure 3. Variation in the observed rate constant (s^{-1}) as a function of the ratio of $[Et_3NH^+]$:[**4b**] (at 20 °C in CH_2Cl_2).

Table III. Observed Rate Constants (s^{-1}) for the Conversion of **4b** to **2** and **3′**[a] in the Presence of Triethylamine

ratio $[Et_3N]$:[**4b**]	10.15	5.1	1.13	1.01
$10^3 k_{obsd}$	1.18	1.16	1.165	1.18

[a] At 20 °C in CH_2Cl_2; initial concentration of **4b** = 8.87×10^{-5} M.

Table IV. Observed Rate Constants (s^{-1}) for the Reaction of the Isothiouronium Salt **5′b** with Amines[a]

amine	$EtNCH_2CH_2\overset{\lceil CH_2CH_2 \rceil}{O}$	Et_3N	proton sponge
pK_a	7.67	10.5	12.3
$10^3 k_{obsd}$	7.6 ± 0.3	9.6 ± 0.4	15.5 ± 0.3

[a] **4b** = 8.87×10^{-5} M in CH_2Cl_2 at 20 °C.

ethylamine, "proton sponge" (1,8-bis(dimethylamino)naphthalene), and *N*-ethylmorpholine, and in each case the same observed rate constants were observed (see Table I). These rate constants are also independent of base concentration (once the amine is in excess). Table III shows the observed rate constants for **4b** when the [**4b**]:[base] concentration is varied from 1:1 to 1:10. The same results were obtained when **4** was added in solid form to a solution containing the base or when base was added to a solution of **4** which had been allowed to come to equilibrium in CH_2Cl_2.

(iv) Reaction of Isothiouronium Salt 5′ with Base. The rates and products obtained for this reaction depend on the ratio of [base]:[substrate]. When [base]:[**5′**] is 2 or greater then the product obtained is the carbodiimide **2** and anion **3′** (in addition to the protonated amine). Under these conditions the optical density at 280 nm drops very rapidly even in dilute solution. Clearly under these conditions 1 equiv of base is used to deprotonate **5′** to **5** which then undergoes rapid elimination to **2** and **3′** catalyzed by the second molecule of base; **4** is not formed under these conditions.

With just 1 equiv of amine base then the final product is the N-phosphorylated substrate **4** (in equilibrium with a small quantity of **2**, **3′** and **5′** as formed from **4**, as shown above). There is an initial rapid decrease in optical density followed by a slower increase (which corresponds to the formation of **4**) when **5′** is introduced to CH_2Cl_2 in the presence of **1** equiv of amine. The rate of reaction depends on the base used. Interestingly with the strongest amine base (the proton sponge, pK_a = 12.3)[10] the observed first-order rate constant obtained (see Table IV) is the same as that obtained (see Table I) when **4** is used as substrate to give the same equilibrium mixture in the absence of added acid or base. As mentioned above the equilibrium for the R = Ph substrates lies almost entirely on the side of **4a** so that the rate constant of equilibration cannot be obtained starting from this side. However, we can measure a rate constant (k_{obsd} = (2.39 ± 0.09) × 10^{-2} s^{-1}) for **5a′** using a 1:1 ratio of [**5a**]:[proton sponge].

(v) Reaction of Carbodiimide 2 with the Thiophosphoric Acids 3. When the equilibrium is approached through the reaction of the acids **3** with the carbodiimide **2** (at the same initial concentration of 8.87×10^{-5} M), the first-order rate constants noted in Table V are obtained. Note that these rate constants are first order and are close to those obtained previously (see (i) and (iv) above) when the equilibrium is approached from either **4** or from **5′** with equimolar base.

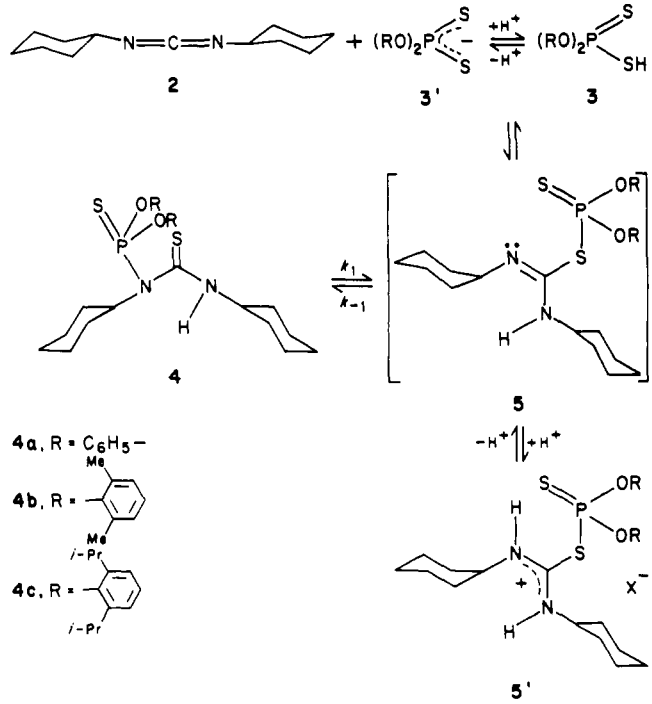
Reproducibility of the observed rate constants is, however, lower than in sections i–iv above, probably because of the instability of the thiophosphoric acid solutions. Dimerization of thiophosphoric acid esters in organic solvents is well documented.[11,12]

Table V. Observed Rate Constants for Reaction of Carbodiimide **2** with Thiophosphoric Acids **3**[a]

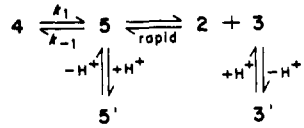
thiophosphoric acid	**3a**	**3b**	**3c**
$10^3 k_{obsd}$, s^{-1}	22.0 ± 0.5	17.2 ± 0.2	5.7[b]

[a] At 20 °C in CH_2Cl_2; [**2**] = [**3**] = 8.87×10^{-5} M; the rate-determining step is not the addition step (see text). [b] Approximate value.

Scheme II



Scheme III



Discussion

The equilibrium shown in Scheme II can be approached either (a) from **4** (which can be isolated pure but equilibrates when dissolved in CH_2Cl_2), (b) from the isothiouronium salt **5′** (isolated with BF_4^- as counterion) on the addition of ≤1 equiv of a base, or (c) from equimolar quantities of the carbodiimide **2** and acid **3**.

These reactions are shown schematically in Scheme III. Since the pK_a of the thiophosphoric ester **3** is less than that of the isothiourea **5**, conversion to the conjugate acid and base **5′** and **3′** occurs so that the concentration of **5** and **3** present at equilibrium is negligible.

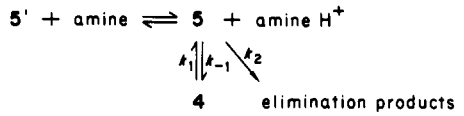
The reaction between **2** and **3** is rapid at −80 °C so that it is not unreasonable that this should be complete on mixing at 20 °C. The observed rate constants are first order, but are composite,

(10) Alder, R. W.; Bowman, P. S.; Vickery, B. L. *J. Chem. Soc., Dalton Trans.* **1972**, 395.

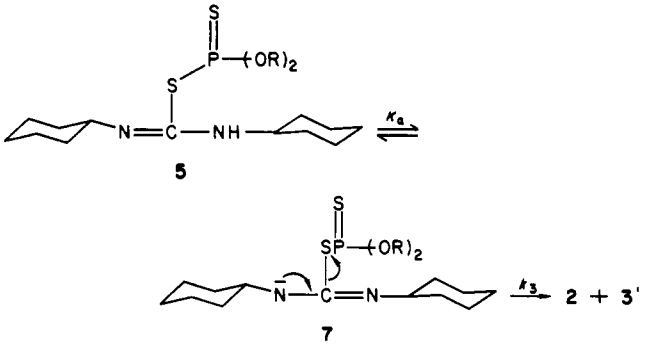
(11) Dyrssen, D.; Liem, D. H.; Ekberg, S. *Acta Chem. Scand.* **1964**, *18*, 135.

(12) Dyrssen, D. H.; Liem, D. H. *Acta Chem. Scand.* **1960**, *14*, 1091.

Scheme IV



Scheme V



and the same rate constant of equilibration ($k_{obsd} = k_1 + k_{-1}$, Scheme III) is obtained by each of the routes a–c above.

The position of the equilibrium lies completely on the side of **5'** in the presence of excess acid (k_1 is then rate determining), and complete conversion to **2** and **3'** occurs in the presence of excess base. In neutral conditions the position of the equilibrium depends on the bulk of the substituent R. With the least bulky substituent (R = Ph) the equilibrium lies almost completely on the side of the N-phosphorylated isomer **4**, but the amount of **4** at equilibrium decreases (see Table II) as R is successively 2,6-$Me_2C_6H_3$ and 2,6-*i*-$Pr_2C_6H_3$.

In the presence of a strong acid (HBF_4) the reaction of **4** (to **5'**) becomes irreversible so that k_1 can be measured directly; consistent with "trapping" of the S-phosphorylated form by protonation, the observed rate constant measured under these conditions ($k_{obsd} = k_1$) is independent of the concentration of this acid. With the weaker acid $Et_3NH^+BF_4^-$ a large excess is required to reach the same k_1 value owing to equilibrium **5** ⇄ **5'** to be shifted to right. At lower concentrations, for which this equilibrium interfers, rate constant values are higher since including a k_{-1} term (see Figure 3).

When 1 equiv of base is reacted with **5'**, then the same equilibrium mixture is obtained as that starting from **4** under neutral conditions, but only when the strongest amine base (the "proton sponge") is used. In other cases smaller rate constants are obtained which imply a preequilibrium (see Scheme IV). This is not unreasonable since the pK_a of **5'** (calculated as 9.5 in H_2O[13,14]) is in the same range as the reported aqueous pK_a's of the other amine bases used.

In the presence of excess base ([amine] ≫ 2 × [**5'**] for other amines), elimination to give **2**, **3'**, and [amine H^+] occurs. The amine "traps" the proton, rendering the reaction irreversible. The rate of formation of **2** and **3'** under these conditions is independent of the nature and concentration (once it is in large excess) of the base so that the rate-determining step under these conditions cannot be the elimination itself.

Elimination of RS^- from isothioureas has been shown to proceed via an E1cB mechanism.[15] If the elimination step (see Scheme V) were rate determining for the reaction of **4** in the presence of excess base, then this would imply that the isourea **5** was converted completely to its counterion **7** under the reaction conditions (since the reaction is observed to be independent of base at high base concentrations). This is unreasonable since the pK_a of **5** (for proton loss) can be calculated as >10,[16] whereas bases such as *N*-ethylmorpholine (pK_a in water = 7.67) give base-independent elimination. Moreover, loss of such a good leaving group as **3'** from **7** is unlikely to be rate determining. Similar arguments can be used to rule out the conjugate base of **4** reacting in the slow step, while the kinetic order rules out mechanisms involving concerted eliminations as rate determining.

This requires the inclusion of an extra step in the overall reaction profile; it is proposed that the interconversion between two isomers (invertomers about the imine nitrogen) of **5** (**5** and **8**, respectively, see Scheme VI) becomes rate determining when **4** is treated with excess base. The inclusion of the extra step is also required by the different individual rate constants obtained when the equilibrium is approached from different directions as detailed in the next paragraph.

As for the two sets of results obtained starting from **5'** with 1 or 2 mol of base, they are summarized in Scheme VII.

Table VI. Individual Rate Constants (s^{-1})[a]

rate constant	substrate 4a	4b	4c
k_1	1.08×10^{-3}	3.17×10^{-3}	6.56×10^{-4}
k_{-1}	3.28×10^{-1}	9.99×10^{-2}	7.41×10^{-4}
k_2	4.97×10^{-3}	6.13×10^{-2}	4.44×10^{-3}
k_1/k_{-1}	3.3×10^{-3}	3.2×10^{-2}	0.89

[a] In CH_2Cl_2 at 20 °C, obtained starting from **4** in the presence of excess triethylamine.

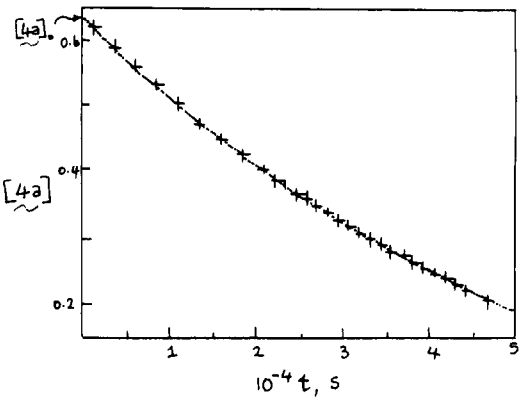


Figure 4. Typical optical density vs. time plot for the reaction of **4a** at 20 °C in CH_2Cl_2 in the presence of triethylamine; the line is experimental and the points theoretical; derived by using eq 2 and the values for the constants given in table VI.

Determination of Individual Rate Constants k_1, k_{-1}, and k_2. From Scheme VI the concentration of **4** at any time can be expressed in terms of eq 2;[17] the values of the constants in terms

$$[4] = P_1 e^{r_1 t} + Q_1 e^{r_2 t} + [4]_0 \qquad (2)$$

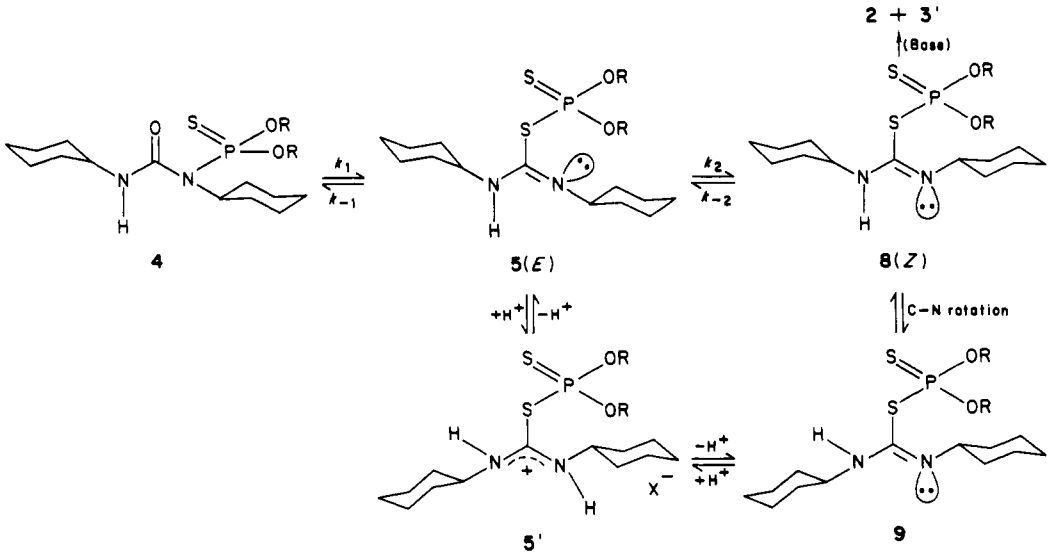
of k_1, k_{-1}, k_2 and the initial concentration of [**4**] are given in the Experimental Section. The individual rate constants were determined by using a computer fitting technique and are summarized in Table VI, together with the values of k_1 and initial concentration of **4** (which are known). An example of the fitting technique (concentration of **4** against time) is given in Figure 4. It should be noted that the trend in k_1/k_{-1} values (Tables VI) shows a decreasing fraction of the *N*-phosphoryl relative to the *S*-phosphoryl form as the bulk of R is increased. It should also be noticed that the k_{-1} term is more sensitive to steric effects than k_1; a tentative explanation is that the restricted lone pair on nitrogen is more hindered by bulky groups than the diffuse p orbital on sulfur.

When **4** is treated with excess base, **5** does not accumulate since it cannot be stabilized by protonation. The steady-state approximation can therefore be used (see eq 3).

$$k_{obsd} = \frac{k_1 k_2}{k_{-1} + k_2} \qquad (3)$$

(13) Mikolajczyk, M.; Kielbasandki, P. *Tetrahedron* **1981**, *37*, 233.
(14) Pratt, R. F.; Bruice, T. C. *Biochemistry* **1971**, *10*, 3179.
(15) Briody, T. A.; Hegarty, A. F.; Scott, F. L. *Tetrahedron* **1977**, *33*, 1469.
(16) Pratt, R. F.; Bruice, T. C. *J. Am. Chem. Soc.* **1972**, *94*, 2823.
(17) Jungers, C. J., et al. "Cinetique Chimique Appliquêe"; Ed. Institut Francais du Petrole, Paris; p 199–207.

Scheme VI



Scheme VII

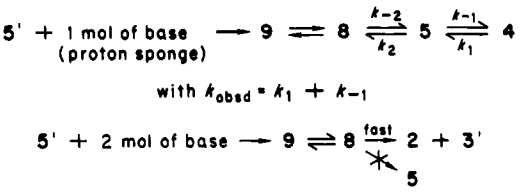


Table VII. Observed and Calculated Rate Constants (Excess Base)[a]

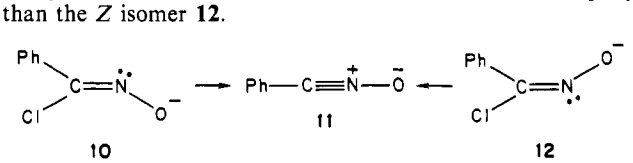
substrate	4a	4b	4c
k_{obsd}(calcd)[b]	1.61×10^{-5}	1.20×10^{-3}	5.17×10^{-4}
k_{obsd}(exptl)	1.70×10^{-5}	1.18×10^{-3}	5.54×10^{-4}
deviation	5%	2%	7%

[a] For reaction of **4** in CH_2Cl_2 at 20 °C in excess triethylamine. [b] In s^{-1}, calculated from the constants listed in Table VI.

The observed experimental values obtained under these conditions compare favorably (see Table VII) with those calculated using the k_1, k_{-1}, and k_2 values already reported (Table VI).

Kinetic Scheme. These results are consistent with the extended kinetic Scheme VI. In this an extra step (governed by the rate constant k_2) is introduced. We proposed that the extra step is the conversion of *E* (**5**) to *Z* (**8**) forms of the *S*-phosphoryl isomer.

The following observations support this assignment. Faster elimination from **8** than from **5** is expected since in **8** the lone pair on the adjacent nitrogen is trans to the leaving group. Such stereoelectronic control of eliminations is well documented. For example, Grob[18] and Deslongchamps[19] have shown that the breakdown of tetrahedral intermediates occurs in the direction predicted by an antiperiplanar arrangement of lone pairs and an adjacent leaving group. In a system related to **5** and **8** it has been shown[20] that the isomer **10** with the lone pair trans to the leaving group (Cl^-) forms the nitrilium ion **11** 6×10^5-fold more rapidly than the *Z* isomer **12**.



The X-ray structure of the isothiouronium salt **5b'** (Figure 5) shows clearly that access to the proton on N-7 is much less hindered than on N-8. More rapid deprotonation at this site gives

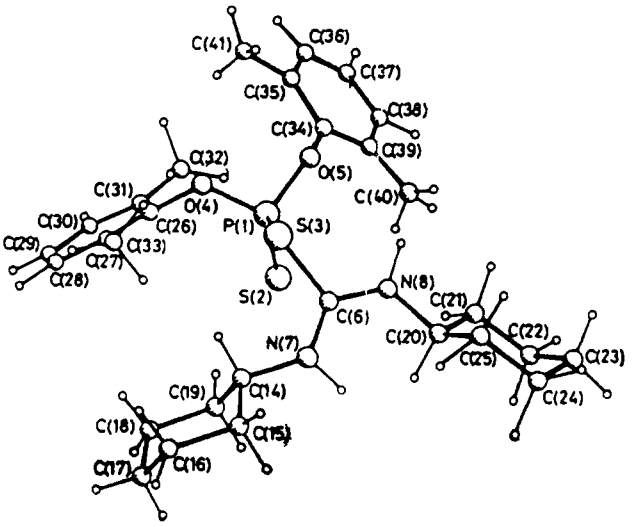


Figure 5. X-ray crystallographic structure of **5b'**.[9]

9 and then **8** which has the lone pair on the adjacent nitrogen correctly located to give direct elimination to **2** and **3**.

On the other hand *N*- → *S*-phosphoryl group transfer (**4** → **5**) necessarily gives the incorrect configuration for elimination. It has been shown[21] that acyl group transfer between O and N in *O*-acylisoureas/*N*-acylamides occurs only from the *O*-acyl isomer in which the migrating group and the lone pair on the adjacent nitrogen are cis to one another.

Although the *S*-phosphoryl compound **5** could not be observed at ambient temperature by ^{31}P NMR (equilibration to **4**, **5'**, **2**, and **3'** occurring), two isomers were observed when **5** was prepared by reacting **2** with **3** at −80 °C. The two peaks at 66.7 and 68.7 ppm could be assigned to **5** and **8**, respectively, and had the following relative intensities: 75:25 (**5a**), 85:15 (**5b**), and 100:0 (**5c**).

Because of steric hindrance in the cis form **8**, we would expect form **5** to predominate at equilibrium (i.e., $k_{-2} > k_2$). Consistent with this, the same rate constant of equilibration ($k_1 + k_{-1}$) is obtained when the isothiouronium salt **5'** is treated with 1 equiv of a strong base as when **4** is allowed to equilibrate to **4**, **5'**, **2**, and **3'** in the absence of base or acid. Moreover, in the absence of added base the reaction of **2** and **3** at 20 °C is very rapid, and the observed rate constant is again the one of equilibration between

(18) Grob, C. A. *Angew. Chem. Int. Ed. Engl.* **1969**, *8*, 535.
(19) Deslonchamps, P. *Tetrahedron* **1975**, *31*, 2463.
(20) Hegarty, A. F.; Mullane, M., unpublished results.
(21) Hegarty, A. F.; McCormack, M. T.; Brady, K.; Ferguson, G.; Brady, P. *J. Chem. Soc. Perkin Trans. 2* **1980**, 867. Pratt, R. F.; Bruice, T. C. *J. Chem. Soc. D* **1971**, 1259.

4 and **5** (i.e., $k_{obsd} = k_1 + k_{-1}$). The fact that k_2 becomes rate determining only under basic conditions may also be ascribed to acid catalysis of this imine nitrogen inversion (via the protonated form **5'**).

Conclusions

The reversible phosphoryl transfer system shown in Scheme VI can be reached by any of three routes. In general the rate-determining step is the same in each case: equilibration between **4** and **5** so that $k_{obsd} = k_1 + k_{-1}$ can be determined; the equilibrium constants k_1/k_{-1} are also accessible, and these show the key role that steric hindrance about phosphorous plays in determining the equilibrium position. The bulky nature of the groups about phosphorous also inhibits other reactions (such as intermolecular reaction to give thiophosphoric anhydrides observed with the corresponding oxygen analogues[9]), and the system is truly reversible. Neither are Arbuzov-type reactions (which would lead to a thiophosphoric ester and reactive metaphosphate and thus side products[13]) observed because of the bulk of the groups around phosphorus.

Only in the presence of base does a step other than $S \rightleftharpoons N$-phosphoryl group transfer become rate determining. E/Z isomerization about the C=N bond of amidines is strongly catalyzed by acid so that in basic solution the uncatalyzed reaction becomes rate determining. Since the actual elimination step never becomes rate determining, it is not possible to say whether elimination occurs via the conjugate base of **8** or via unimolecular ionization of **8** (the reverse of acid-acatalyzed addition to the carbodiimide).

Because of the high mobility of these systems, clearly further reaction can occur through any of the species shown in Scheme VI. Thus while the substrate can be "stored" as form **4**, reaction can occur via the nucleophilic nitrogen in **5**, displacing the equilibrium entirely through this route. Product isolation cannot be used to rationalize further reactivity in solution.

Phosphorylation of cyclic ureas[22] gives N-phosphorylated products while pyrimidines[23] and hydroxypyridines[24] give either O- or N-phosphorylation depending on the substituents present and the phosphorylating agent. Another example is the reaction of succinimide for which *N*-phospho[25] and *O*-phospho derivatives have been successively claimed.[26] Although not directly investigated, the possibility arises that the phosphoryl groups in these substrates migrate in solution. The system which we have described here also partially mimics intramolecular phosphoryl group transfer in phosphomutases where a phosphoryl group is shuttled between two histidine groups.[27] We are currently extending this study to investigate the nucleophilic properties of the *S*-phosphoryl form **5** starting from the *N*-phosphourea **4**, toward carboxylating agents—the *O*-phosphobiotin route in Scheme I—and also the phosphoryl group transfer properties of the isothiouronium salt **5'**.

Experimental Section

General. Melting points are uncorrected. Elemental analyses were carried out by Laboratoire Central de Microanalyse du CNRS. IR spectra were obtained by using a Beckman IR 20A and Perkin-Elmer 683, NMR spectra on a Brucker WH 90 MHz FT. Chemical shifts are indicated by using 85% H_3PO_4 as reference. UV spectra were obtained by using a Beckman 5260, and mass spectra were obtained on a Varian MAT 311 A using the electronic impact mode.

Substrates. Thiophosphoric esters 3 were prepared according to the method of Fletcher and Hamilton.[28] Ester **3a** (R = Ph): mp 64 °C (25% yield, from 1:1 benzene–*n*-hexane); ^{31}P NMR (0.25 M in dichloromethane) δ 79.7; MS, M^+ 282, other fragments 250, 217, 187, 171, 155, 141.

Anal. Calcd for $C_{12}H_{11}O_2PS_2$: C, 51.09; H, 3.90; P, 10.98; S, 12.70. Found: C, 50.79; H, 3.83; P, 10.55; S, 22.33.

Ester **3b**: mp 114 °C (73% yield, from 1:1 benzene–*n*-hexane); ^{31}P NMR (0.25 M in dichloromethane) δ 76.8; MS, M^+ 338, other fragments 305, 273, 216, 201, 183.

Anal. Calcd for $C_{16}H_{19}O_2PS_2$: C, 56.78; H, 5.66; P, 9.15; S, 18.95. Found: C, 56.70; H, 5.71; P, 9.22; S, 18.71.

Ester **3c** (R = 2,6-*i*-$Pr_2C_6H_3$) slowly crystallized from the oily residue on washing with *n*-hexane: mp 89 °C; ^{31}P NMR (0.25 M in dichloromethane) δ 79.2.

Anal. Calcd for $C_{24}H_{44}O_2PS_2$: C, 63.97; H, 7.83; P. 6.87; S, 14.23. Found: C, 63,92; H, 7.84; P, 6.73; S, 13.88.

Thiouronium salts 5' were prepared by adding an excess of tetrafluoroboric acid (in ether) to a solution of the corresponding N-phosphorylated thiourea **4** (in ether). The salts **5'a** and **5'c** were separated by filtration and dried in vacuo. The salt **5'b** was obtained as follows: to 2.5 mmol of **3b** in ether (7.5 mL) was added at −80 °C, dicyclohexylcarbodiimide (2.5 mmol) in ether (2.5 mL). The mixture was maintained at −80 °C for 5 min, and then an excess of HBF_4 in ether was added. The mixture was allowed to warm to room temperature, and the isothiouronium salt **5'b** was separated by filtration and dried. Physical data for the salts are as follows.

5'a: 75% yield; ^{31}P NMR (0.2 M in CH_2Cl_2) δ 71.6 (75%), 71.2 (25%); IR (2% in KBr) ν_{NH} 3260, 3260 cm^{-1}, $\nu_{C=N}$ 1640 cm^{-1}.

Anal. Calcd for $C_{25}H_{33}N_2PO_2S_2 \cdot BF_4$: C, 52.11; H, 5.77; N, 4.86; P, 5.38; S, 11.13; B, 1.83; F, 13.19. Found: C, 52.0; H, 6.05; N, 4.87; P, 5.02; S, 11.01; B, 1.43; F, 13.64.

5'b: mp 195 °C (yield, 79%); MS, $M^+ - BF_4$ 544, other fragments 338; 305; 273, 240, 206, 138, 163, 122; ^{31}P NMR (saturated solution in CH_2Cl_2) δ 67; IR (2% in KBr) ν_{NH} 3240 cm^{-1}, $\nu_{C=N}$ 1640 cm^{-1}.

Anal. Calcd for $C_{29}H_{42}N_2O_2PS_2 \cdot BF_4$: C, 55.05; H, 6.7; N, 4.45; P, 4.9; S, 10.15; B, 1.7; F, 12.0. Found: C, 55.15; H, 6.75; N, 4.25; P, 5.15; S, 9.9; B, 2.15; F, 12.1.

5'c: ^{31}P NMR (0.25 M in CH_2Cl_2) δ 67.9; IR (2% in KBr) ν_{NH} 3220, 3240 cm^{-1}, $\nu_{C=N}$ 1640 cm^{-1}.

N-Phosphorylated thioureas 4 were prepared according to the following general procedure. The corresponding thiophosphoric ester **3** (2.5 mmol) was dissolved in diethyl ether (6 mL) at ambient temperature. The precipitated thiourea which formed was filtered off (after 30 min) and washed with ether. **4a**: mp 112 °C (yield 55%, from ether): MS, M^+ 488, other fragments 405, 395, 389, 249, 240, 206 and 77; ^{31}P NMR (0.25 M in CH_2Cl_2) δ 59.6; IR (2% in KBr) ν_{NH} 3340 cm^{-1}.

Anal. Calcd for $C_{25}H_{32}N_2PO_2S_2$: C, 61.48; H, 6.6; N, 5.74; P, 6.34; S, 13.13. Found: C, 61.15; H, 6.71; N, 5.78; P, 6.32; S, 12.79.

Compound **4b**: mp 141 °C (yield 72%, from ether); MS, M^+ 544, other fragments, 338, 305, 240, 206, 121; ^{31}P NMR (0.25M in ether) δ 56.9, without decoupling $^3J_{HCNP}$ = 12 Hz; IR (2% in KBr) ν_{NH} 3360 cm^{-1}.

Anal. Calcd for $C_{29}H_{41}N_2O_2PS_2$: C, 64.0; H, 7.55; N, 5.15; P, 5.7; S, 11.80. Found: C, 64.15; H, 7.70; N, 5.10; P, 5.70; S, 11.90.

The thiourea **4c** was obtained in 90% yield: MS, M^+ 656, other fragments 575, 479, 450, 417, 240, 208; ^{31}P NMR (0.25 M in CH_2Cl_2) δ 56.6; IR (2% in KBr) ν_{NH} 3400 cm^{-1}.

Anal. Calcd for $C_{37}H_{56}N_2PO_2S_2$: C, 67.75; H, 8.61; N, 4.27; P, 4.72; S, 9.78. Found: C, 67.50; H, 8.65; N, 4.15; P, 4.46; S, 9.28.

Product Analysis. In each case, the reactions studied kinetically (at low concentration) were also studied on a preparative scale, and the products were either (a) isolated or (b) identified by comparing ^{31}P NMR and IR spectra with those of authentic samples.

Kinetics. The kinetic experiments were carried out by using a Beckman Model 5260 UV spectrophotometer in conjunction with a Hewlett-Packard 85 for data analysis. In some cases the data were also analyzed by using a Tektronix 4051 32K Computer[29] using LSG and VAO4A programs.[30] The solvent used in all cases was CH_2Cl_2 which had been dried by using standard techniques, and kinetic experiments were carried out at 20 ± 0.1 °C. Following initial repetitive scans (see Figure 1 for an example) the course of the reaction was generally followed at 280 nm (where the N-phosphorylated isomer **4** has an absorption maximum).

The reactions were prepared for study in either of two ways: (a) 10 μL of the substrate (dissolved in CH_2Cl_2 (2.65×10^{-4} M)) was introduced into the cell containing temperature equilibrated CH_2Cl_2 (3.0 mL). The reaction solution was then modified by adding 10 μL of the other reactant solution (usually acid or base). (b) In those reactions which

(22) Ning, R. Y.; Fryer, R. I.; Madan, P. B., Sluboski, B. C. *J. Org. Chem.* **1976**, *41*, 2720.

(23) (a) Reznik, V. S.; Shvetsov, Yu S.; Bakulin, V. S. *I. Sh. Bull. Acad. Sci. USSR, Ser. Chem.* **1975**, *24*, 1290. (b) Reznik, V. S.; Bakulin, V. S.; Shetsov, Yu S.; Ivanov, B. E. *Ibid.* **1975**, *24*, 1294.

(24) Guibe-Jampel, E.; Wakselman, M. *J. Chem. Soc. D* **1969**, 720.

(25) Tsolis, A. E.; McEwen, W. E.; Venderwerf, C. A. *Tetrahedron Lett.* **1964**, 3217.

(26) Mukaiyama, T.; Obata, T.; Mitsunobu, O. *Bul.. Soc. Chim. Jpn.* **1965**, *38*, 1088.

(27) Knowles, J. R. *Annu. Rev. Biochem.* **1980**, *49*, 877.

(28) Fletcher, J.; Hamilton, J. C. *J. Am. Chem. Soc.* **1950**, *72*, 2461.

(29) Detar, D. F. "Computer Programs for Chemistry"; Benjamin: New York, 1968.

(30) Powell, M. J. D. *Comput. J.* **1964**, *7*, 155.

were particularly rapid, **4** was added as a solid to the cell containing CH_2Cl_2 (3.0 mL) at 20 °C. The solid, which was finely divided, rapidly dissolved.

The initial concentration of **4** was obtained by comparison of the optical density of the equilibrium mixture with that of a reference solution. When other reactants were present, the initial concentration was obtained by using the program and extrapolation back to zero time.

Since **4b** and **4c** react in CH_2Cl_2 even in the absence of added acid or base, molar extinction coefficients were obtained by extrapolation of the optical densities at 280 nm to zero time. The following values were obtained: ϵ_{4b} 10 500 ± 100; ϵ_{4c} = 10 450 ± 50. Since **4a** does not react in neutral CH_2Cl_2 the following value was measured directly: ϵ_{4a} 10 700.

At 280 nm dicyclohexylcarbodiimide (**2**) and the thiophosphate anion **3′** do not have a significant absorption so that the optical density at equilibrium can be determined from eq 5, derived from eq 4.

$$\underset{x_0 - x}{\mathbf{4}} \rightleftharpoons \underset{y}{\mathbf{2}} + \underset{y}{\mathbf{3'}} + \underset{x - y}{\mathbf{5'}} \tag{4}$$

$$OD_{equil} = (x_0 - x)\epsilon_4 + (x - y)\epsilon_3 \tag{5}$$

The molar extinction coefficient for **5** cannot be obtained directly, but it is safe to assume that because of the relative acidity of **3** and the basicity of **5** that the concentration of free **5** present in solution will be negligible. Since it was confirmed (by IR and ^{31}P NMR) that **2** = **5′** = **3′**, eq 5 reduces to eq 6, and thus the concentration of **4** at equilibrium is given by eq 7.

$$OD_{equil} = (x_0 - x)\epsilon_4 + (x/2)\epsilon_3 \tag{6}$$

$$x_e = x_0\left[1 - \frac{\frac{OD_{equil}}{x_0} - \epsilon_4}{\frac{\epsilon_3}{2} - \epsilon_4}\right] \tag{7}$$

When **4** is treated with base its concentration varies with time according to eq 2 which is derived from eq 8. The initial value of $[\mathbf{4}]_0$ is

$$\mathbf{4} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} \mathbf{5} \xrightarrow{k_2} \text{products} \tag{8}$$

known as is k_1 and the other constants given in eq 9–11.

$$r_1 = \frac{-(k_1 + k_{-1} + k_2) + \{[(k_1 + k_{-1}) - k_2]^2 + 4k_{-1}k_2\}^{1/2}}{2} \tag{9}$$

$$r_2 = \frac{-(k_1 + k_{-1} + k_2) - \{[(k_1 + k_{-1}) - k_2]^2 + 4k_{-1}k_2\}^{1/2}}{2} \tag{10}$$

$$P_1 = \frac{[\mathbf{4}]_0(r_2 - k_1)}{r_1 - r_2};\ Q_1 = \frac{[\mathbf{4}]_0(r_1 - k_1)}{r_2 - r_1} \tag{11}$$

The other constants were determined by using a computer program with a least-squares minimization routine. An example of the resultant optical density vs. time plots (experimental and calculated are given in Figure 4).

In the presence of excess base, **3** is converted to the counterion **3′** so that the formation of **2** and **3′** is essentially irreversible. Under these conditions **5** does not accumulate in solution (tight isosbestic points are held), and the steady-state approximation for the concentration of **5** can be applied. This leads to eq 12, and using the values of k_1, k_{-1}, and k_2

$$k_{obsd} = \frac{k_1k_2}{k_2 + k_{-1}} \tag{12}$$

already determined reproduces closely the observed rate constants (see Table VII).

Registry No. **3a**, 2253-60-3; **3b**, 7355-10-4; **3c**, 81640-00-8; **3′a**, 55979-88-9; **3′b**, 83599-87-5; **3′c**, 92366-09-1; **4a**, 92366-10-4; **4b**, 83599-89-7; **4c**, 92366-11-5; **5′a**, 92366-13-7; **5′b**, 85329-00-6; **5′c**, 92366-15-9; $Et\overline{NCH_2CH_2OCH_2CH_2}$, 100-74-3; Et_3N, 121-44-8; 1,8-bis(dimethylamino)naphthalene, 20734-58-1; dicyclohexylcarbodiimide, 538-75-0.

Nondissociative Permutational Isomerization of an Octahedral Derivative of a Nonmetallic Element, a 12-Te-6 Species[1]

Ronald S. Michalak, Scott R. Wilson, and J. C. Martin*

Contribution from the Roger Adams Laboratory, University of Illinois, 1209 West California St., Urbana, Illinois 61801. *Received April 16, 1984. Revised Manuscript Received August 4, 1984*

Abstract: The reactions of bromine trifluoride with 3,3,3′,3′-tetramethyl-1,1′-spirobi[3*H*-2,1-benzoxatellurole] (**9**) and 6,6′-bis(1,1-dimethylethyl)-3,3,3′,3′-tetrakis(trifluoromethyl)-1,1′-spirobi[3*H*-2,1-benzoxatellurole] (**10**) give rise to the *all-trans*-12-Te-6 species, pertelluranes **3** [1,1-difluoro-1,1-dihydro-3,3,3′,3′-tetramethyl-1,1′-spirobi[3*H*-2,1-benzoxatellurole]-(*OC*-6-12)], and **13** [6,6′-bis(1,1-dimethylethyl)-1,1-difluoro-3,3,3′,3′-tetrakis(trifluoromethyl)-1,1′-spirobi[3*H*-2,1-benzoxatellurole]-(*OC*-6-12)]. Pertelluranes **3** and **13** have two trans fluorines, two trans oxygens, and two trans carbons at ligation sites about the central tellurium atom. The rate of isomerization from the all-trans geometry of **3** to (*OC*-6-22) stereoisomer **4**, with cis fluorines and oxygens and trans carbons, was determined in different solvents and at different concentrations at 61 °C. Failure to trap fluoride ion by added hexadimethyldisilazane provides evidence against a dissociative mechanism involving Te–F bond heterolysis to give dissociation ions. The similarity of first-order rates of reaction [2.0×10^{-5} to 2.5×10^{-5} s^{-1} at 61 °C] in solvents of differing ionizing power (Y values in the range −5.57 to −8.62, m = 0.03) provides evidence ruling out heterolytic dissociation mechanisms for the isomerization of **3** to **4**, including those with ion pair or zwitterionic intermediates. The mechanism for the permutational isomerization is concluded to be a nondissociative twist involving cleavage of none of the six bonds to tellurium. The lower activation barrier for the trigonal twist of 12-Te-6 pertellurane **3** ($\Delta G^*_{61°C}$ = 27 kcal/mol) compared to *all-trans*-12-S-6 persulfane **1** (not observed, $\Delta G^* > 50$ kcal/mol) and the relative energies of the observed stereoisomers of **1** and **3** are discussed in terms of observed ground-state trigonal distortion. The isomerization of **13** to a stereoisomer was not observed, either thermally or in the presence of Lewis acids. This could result from a higher activation barrier for the isomerization or **13** could represent the most thermodynamically stable isomer in the set of possible stereoisomers. The results of complete X-ray crystallographic structure determinations of pertellurane **4** and tellurane **5** are described.

Mechanisms of the often facile ligand rearrangements of trigonal-bipyramidal (TBP) 10-X-5[1] (X = P[2a,b], Si,[2c-g] As,[2h] Sb[2h]) compounds have been a subject of continuing ineterest, especially for 10-P-5 phosphoranes. Ligand rearrangements of 10-X-4 (X

0002-7863/84/1506-7529$01.50/0