'*O*-Phosphobiotine Analogues.' Isolation, *X*-Ray Structure, and Reactivity of the Intermediate of the Addition of Hindered Phosphoric Esters and Thioesters on the Carbodi-imide Group. O → N Phosphoryl Migration *versus* P–O–P Bond Formation

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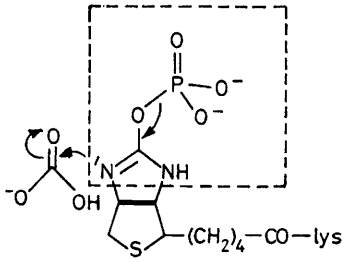
Addition reactions to carbodi-imides using oxy- and thio-phosphoric diesters crowded around phosphorus have been carried out in order to obtain a better understanding of this reaction, and to trap the intermediate, an *O*-phosphobiotin analogue. Oxy-compounds give a mixture of pyrophosphate and rearranged *N*-phosphorylated urea or react poorly if steric hindrance at phosphorus is important. The same type of addition with thiophosphoric esters has made possible the first isolation of the intermediate of this general reaction which is relevant to pyrophosphate and to nucleotide synthesis. This difference is rationalized in terms of the C–X–P bond lengths (X = O or S) in the intermediate, as shown by an *X*-ray crystallographic determination of the structure of the thio-compound. With oxy-compounds, the reaction is very sensitive to steric effects, because of the short C–O–P distances involved. Formation of the intermediate is only likely if the oxyphosphorus nucleophile does not contain bulky groups, and this leads quickly to the pyrophosphate. With thio-compounds, the analogous intermediate is formed even when bulky thiophosphorus groups are present, because of the longer C–S–P distances, and is protected against intermolecular rearrangement both by the poor overlap between the nitrogen and phosphorus orbitals and by protonation if a suitable ratio of ester to dicylohexylcarbodi-imide (DCCD) is used. Moreover this intermediate is not easily attacked by the bulky $(RO)_2P(S)S^-$ nucleophile and therefore accumulates. With a less favourable ratio of ester to DCCD, the reaction gives only the rearranged *N*-phosphorylated product. Crystals of the isolated intermediate are monoclinic, space group $P2_1/a$ with four molecules in a cell of dimensions a = 15.802(3), b = 13.409(2), c = 15.050(5) Å, β = 96.82(1)°. The structure was solved by direct methods and refined by full-matrix least-squares calculations to an R value of 0.055 for 3 144 observed reflexions. Several features of this molecular structure are discussed. Finally, a kinetic study of the rearrangement from C–S–P to N–P compounds shows that this reaction proceeds without the formation of any kinetically significant intermediate.

In our work on the chemistry of the coenzyme biotin, we became interested in the so-called '*O*-phosphobiotin model', an intermediate in which activation of the ureido-part of the molecule towards nucleophilic attack on carbonate is obtained through phosphorylation by an ATP molecule [1,2] as shown in Scheme 1. Since an intermediate close in structure has been suggested in the addition reaction of phosphoric esters with carbodi-imides, we investigated this reaction in more detail (Scheme 2).



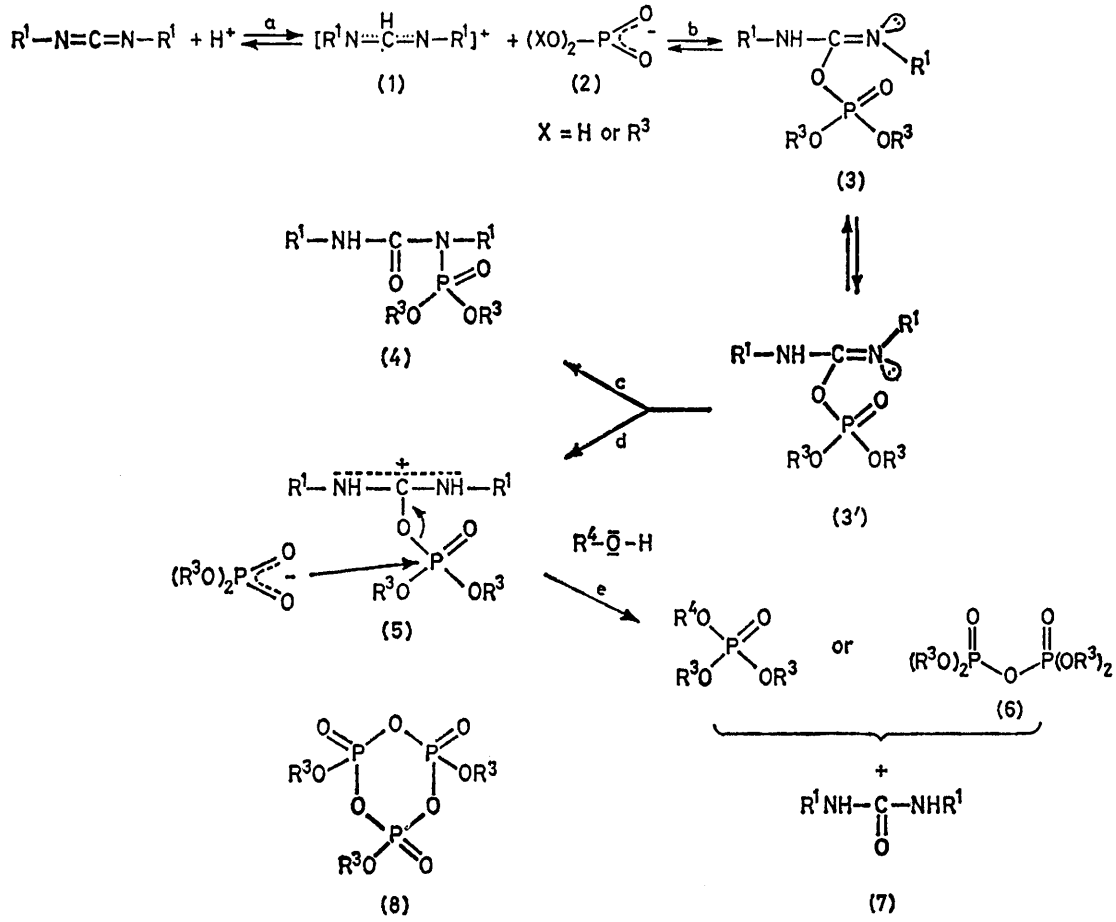
Scheme 1 *O*-Phosphobiotin carboxylation

Also, even though this reaction has been applied in many syntheses and a general scheme has been suggested,[3,4] no mechanistic study has been carried out except for the work of Glonek *et al.*,[5] which gives a thorough analysis of the successive products formed in the reaction of alcohols with phosphoric acid. For the mechanism of the reaction, the general pathway of Scheme 2 has been proposed [3] without any clear evidence, particularly concerning the intermediate (3), which for monoesters [4,6] is thought to be a trimetaphosphate (8).

The rearrangement of (3) into the *N*-substituted compound (4) which is well documented for the similar reaction of carboxylic acids with carbodi-imides [7,8] is not clearly established for phosphoric esters, although it has been suggested in phosphorylation reactions of ureas [9] and uracil.[10] We have therefore carried out a more complete mechanistic investigation of this reaction, including the structure of the key intermediate which is an *O*-phosphobiotin analogue. We have also studied the factors leading either to the pyrophosphate (reaction d, Scheme 2) or to the rearranged *N*-phosphorylated product (reaction c, Scheme 2).

The trapping or isolation of the intermediate (3) or (3') or its protonated form (5) implies that the rates of the steps c and e (Scheme 2) are slowed significantly or even that these reactions are completely retarded. Several different approaches were used to investigate these possibilities.

(a) *The Use of Different Carbodi-imides.*—Initially, we used dicyclohexylcarbodi-imide (DCCD), since this compound has been used extensively for pyrophosphate syntheses. We also employed *N*-*p*-nitrophenyl-*N'*-isopropylcarbodi-imide, in which one nitrogen atom is sufficiently basic for step a to proceed (Scheme 2) and the other is sufficiently weakly nucleophilic to avoid re-



SCHEME 2 Addition reaction of phosphoric esters to carbodi-imides

arrangement by path c. This effect has already been reported in the synthesis of esters using dissymmetric carbodi-imides.[11]

(b) *The Use of Sterically Hindered Esters.*—The reaction was carried out with esters (9) and (10). Steric

$(RO)_2P(O)OH$ (9) $(RO)_2P(S)SH$ (10)

a; R = 2,6-$(CH_3)_2C_6H_3$
b; R = 2,6-$Pr^i_2C_6H_3$

hindrance around phosphorus should first of all slow down intramolecular attack by nitrogen (reaction c, Scheme 2) and also prevent attack by the external bulky nucleophile (path e, Scheme 2) which leads to the pyrophosphate.

(c) *Comparison of Oxy- and Thio-analogues.*—Increasing the distance between phosphorus and the two nitrogen atoms in the intermediate (3) or (3′) should make rearrangement c unfavourable. The investigation was therefore extended to dithiophosphoric esters $(RO)_2P(S)SH$, where the bonds C–S and S–P are significantly longer than in the oxygen analogue. This change was made on the basis of the results reported for *O*- and *S*-acyl-isoureas and -isothioureas,[12,13] which give cyclic analogues of the intermediate (3) or (3′), where a rearrangement akin to (3′) ⟶ (4) has been studied. The corresponding *O*-acyl compounds, which rearrange too fast to be observed, have only been indirectly implicated.[14,15] Moreover, the addition of dithiophosphoric esters $(RO)_2P(S)SH$ to carbodi-imides has not yet been reported.

(d) *Molar Ratios of Reactants.*—Since the required stoicheiometry for pyrophosphate formation is 2 moles of ester per mole of carbodi-imide, the reactions in Scheme 2 should be inhibited by a deficit of ester. Therefore two molar ratios were used, namely 2 : 1 and 1 : 1.

RESULTS AND DISCUSSION

The reactions were followed by ^{31}P n.m.r. Experimental conditions and results are presented in Table 1. For the most significant cases, reactions were also run on a preparative scale and the products were isolated to enable complete characterization.

Reactivity of Phosphoric Esters.—(a) *Esters* (9a *and* b). For (9a), the spectrum of the mixture at −80 °C in dichloromethane shows only one product (δ_P −21 p.p.m.), the reactant esters (δ_P −7 p.p.m.) being partially transformed after 10 min. On warming, several changes take place in the spectrum including the disappearance of the peak at δ_P −21 p.p.m. which therefore corresponds to an

TABLE 1

Addition reactions of esters (9) and (10) to dicyclohexylcarbodi-imide in dichloromethane. Percentages calculated on phosphorus

Compound	Ratio [a]	Temp. (°C) [b]	Time (h)	Formation (%) Substrate $(RO)_2P(O)OH, (RO)_2P(O)O^-$	Intermediate [c]	*N*-Phosphorylated form [d]	Pyrophosphate form [e]
(9a)	1	−80	1/6	58	42	0	0
		+25	36	27	0	27	46
	2	+25	2	50	0	4	46
		+25	72	52	0	3	45
(9b)	1	−80	1/4	91	9	0	0
		+25	24	72	0	28	0
	2	+25	2	98	2	0	0
		+25	24	94	0	6	0
				$(RO)_2P(S)SH, (RO)_2P(S)S^-$			
(10a)	1	−80	1/6	52	48	0	0
		−45	3	16	15	69	0
		+25	1/4	0	0	100	0
	2	−80	1/6	50	50 [f]	0	0
		+25	1/4	50	50	0	0
(10b)	1	−80	1/6	50	50	0	0
		+25	24	5	5	90	0
	2	+25	1/4	50	50	0	0

[a] Molecular ratio of phosphodiester to dicyclohexylcarbodi-imide. [b] The reagents are mixed at low temperature. The solution is then allowed to warm to room temperature. [c] Structure (5*a*), Scheme 2. [d] Structure (4), Scheme 2. [e] Structure (6), Scheme 2. [f] Two isomers are observable under these conditions at δ_P 66 and 68 p.p.m.

intermediate, as two other products are formed (peaks at δ_P −4 and −25 p.p.m.). These products are identified as the rearranged compound (4) and the pyrophosphate (6), respectively. Their chemical shifts are in agreement with those given in the literature for similar compounds.[16] The formation of compound (4) is suppressed by using an excess of ester (9a) with respect to the carbodi-imide. The reaction does not go to completion, since after three days 52% of the starting material remains unchanged. The i.r. spectrum shows the carbodi-imide band at 2 120 cm^{-1} after the same reaction time. On addition of an acid (HBF_4) to the reaction mixture the yield of pyrophosphate is increased from 30 to 100% after 24 h at 25 °C.

For ester (9b), the spectrum of a sample of the reaction mixture at low temperature shows the same peak (δ_P −21 p.p.m.) as that of the intermediate, which disappears on warming the solution. Contrary to (9a) this intermediate leads only to the *N*-phosphorylated compound (4) (δ_P −4.2 p.p.m.) and not to the pyrophosphate. Thus bulky groups around phosphorus efficiently inhibit intermolecular reaction at this atom.

(b) *Esters* (10a *and* b). Different results are obtained with these esters. The n.m.r. spectrum of the reaction mixture shows that even at −80 °C, the reaction goes to completion. The ester (10a) [or (10b)] disappears immediately, and three products are formed (δ_P 103, 68, and 66 p.p.m.), the sum of intensities of the two last being equal to the intensity of the former. Moreover, depending on the ratio of reactants, the following changes are observed. With a molar ratio ester : DCCD of 1 : 1, on warming up the solution, the three peaks at δ_P 103, 68, and 66 p.p.m. disappear, and a new compound is formed (δ_P 57.5 p.p.m.). This is the only signal observed when the reactants are mixed at room temperature. With a molar ratio of 2 : 1 the peaks at δ_P 103 and 66 are observed at 25 °C, and therefore correspond to a stable compound under these conditions. (The peak at δ_P 68 p.p.m. is only observed at low temperature.) The i.r. spectrum of the solution shows an absorption at 1 640 cm^{-1}, corresponding to a C=N bond. In this second case the product with a signal at δ_P 57.5 p.p.m. is formed very slowly (a few percent at room temperature after several days).

The peaks were assigned as follows. The ^{31}P peak at δ_P 103 p.p.m. corresponds to the anion $(RO)_2P(S)S^-$. This is deduced from the n.m.r. spectrum of a mixture of (10a) with isopropylamine, which shows a resonance peak of the same chemical shift. The peak at δ_P 57.5 p.p.m. corresponds to the *N*-phosphorylated compound (4). The structure of this compound, isolated in the reaction carried out with molar ratio of 1 : 1, was established by i.r., n.m.r., and mass spectroscopy, and elemental analysis.

The two peaks at δ_P 68 and 66 p.p.m. observed with a reactant ratio of 2 : 1 correspond to an intermediate which is stable only under acidic conditions. Since the relative intensities of the two peaks at δ_P 103 and 66 p.p.m. are close to unity at all temperatures studied, the intermediate stabilized in acidic media should be the protonated form (5) (Scheme 2) with $(RO)_2P(S)S^-$ as counterion (δ_P 103 p.p.m.). The peak at δ_P 68 p.p.m., which is observed only at low temperature, could be due to an isomer of the protonated intermediate (5). This point will be discussed later. Thus, in contrast to the reactions of the phosphoric esters (9a and b), for which the intermediate is too reactive to be isolated, in the case of thiophosphoric esters (10a and b) this intermediate should be isolable since it is stable at room temperature.

Isolation and Structural Determination of the Intermediate (5) *formed in the Addition Reaction of Dithiophosphoric Ester* (10a) *with DCCD.*—Because of the general interest in this type of intermediate its isolation and structure determination were carried out by adding tetrafluoroboric acid to a mixture of (10a) and DCCD (molar ratio 1 : 1) in CH_2Cl_2 at −80 °C; a crystalline

precipitate formed. After warming to room temperature this product was isolated by filtration. Its ^{31}P n.m.r. spectrum was checked, and corresponds to that for the intermediate observed in the spectrum run *in situ*. An *X*-ray crystallographic structure determination was carried out. It agrees with that of the postulated protonated intermediate (5). Its configuration is given in

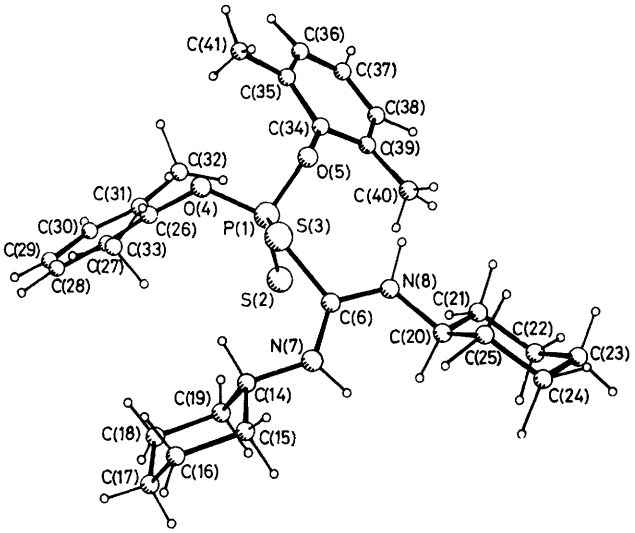


FIGURE 1 Molecular structure with numbering of intermediate (5)

Scheme 3. Thus, there is now clear evidence of the structure of the key intermediate for the addition of phosphoric esters to carbodi-imides. This reaction also appears to be a valuable route to *O*-phosphobiotin analogues (Scheme 1). This intermediate was previously proposed on the basis of n.m.r. results,[17] but no structural proof was given.

Molecular Structure of the Intermediate (5).—The structure and molecular packing in the crystal are indicated in Figures 1 and 2. Bond lengths and angles and atomic co-ordinates are given in Tables 2 and 3. Several features should be mentioned.

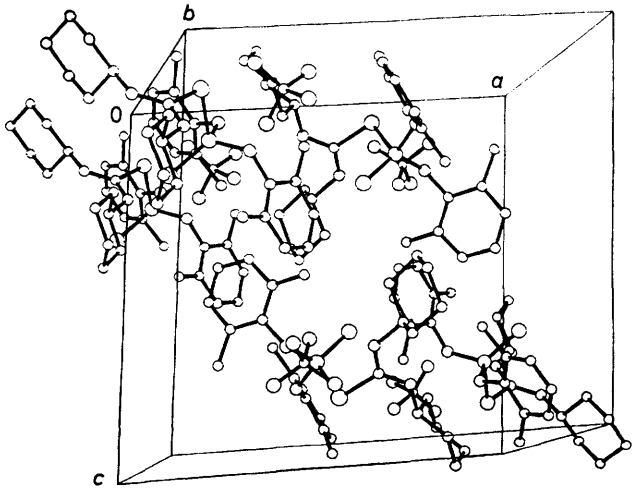


FIGURE 2 Molecular packing in the crystal of (5)

TABLE 2

Bond lengths (Å) with standard deviations in parentheses

S(2)	P(1)	1.898(2)	C(21)	C(20)	1.515(9)
S(3)	P(1)	2.128(2)	C(25)	C(20)	1.475(9)
O(4)	P(1)	1.584(4)	C(22)	C(21)	1.537(12)
O(5)	P(1)	1.578(3)	C(23)	C(22)	1.457(12)
C(6)	S(3)	1.783(5)	C(24)	C(23)	1.512(13)
C(26)	O(4)	1.438(6)	C(25)	C(24)	1.533(12)
C(34)	O(5)	1.432(6)	C(27)	C(26)	1.399(7)
N(7)	C(6)	1.319(6)	C(31)	C(26)	1.379(7)
N(8)	C(6)	1.297(7)	C(28)	C(27)	1.381(8)
			C(33)	C(27)	1.500(8)
C(14)	N(7)	1.469(7)	C(29)	C(28)	1.375(9)
C(20)	N(8)	1.489(7)	C(30)	C(29)	1.369(9)
F(10)	B(9)	1.377(9)	C(31)	C(30)	1.404(8)
F(11)	B(9)	1.354(8)	C(32)	C(31)	1.509(8)
F(12)	B(9)	1.368(9)	C(35)	C(34)	1.389(7)
F(13)	B(9)	1.362(10)	C(39)	C(34)	1.388(8)
C(15)	C(14)	1.522(8)	C(36)	C(35)	1.397(8)
C(19)	C(14)	1.511(9)	C(41)	C(35)	1.484(9)
C(16)	C(15)	1.525(9)	C(37)	C(36)	1.380(10)
C(17)	C(16)	1.529(10)	C(38)	C(37)	1.388(10)
C(18)	C(17)	1.502(9)	C(39)	C(38)	1.389(8)
C(19)	C(18)	1.517(9)	C(40)	C(39)	1.478(9)

TABLE 3

Valency angles (°) with standard deviations in parentheses

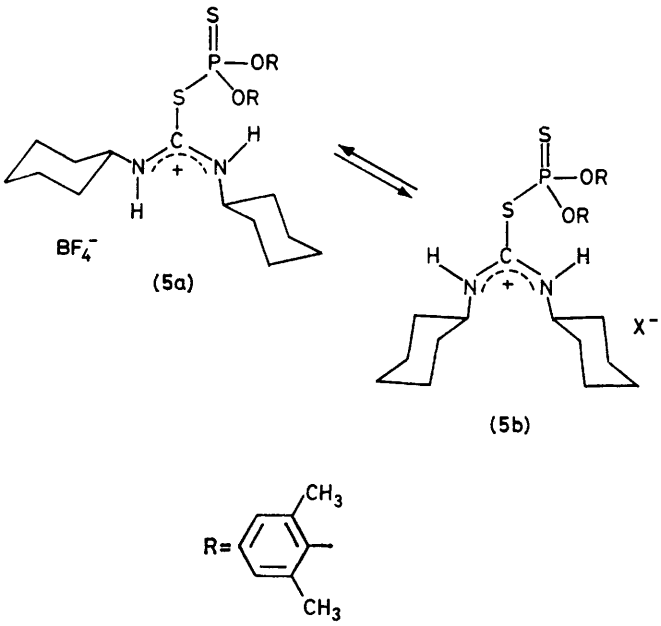
S(3)–P(1)–S(2)	116.1(1)	C(25)–C(20)–C(21)	112.7(5)
O(4)–P(1)–S(2)	117.4(2)	C(22)–C(21)–C(20)	110.3(6)
O(4)–P(1)–S(3)	99.6(1)	C(23)–C(22)–C(21)	112.4(7)
O(5)–P(1)–S(2)	120.2(2)	C(24)–C(23)–C(22)	111.9(7)
O(5)–P(1)–S(3)	97.5(1)	C(25)–C(24)–C(23)	110.9(7)
O(5)–P(1)–O(4)	102.4(2)	C(24)–C(25)–C(20)	110.7(6)
C(6)–S(3)–P(1)	101.1(2)	C(27)–C(26)–O(4)	117.9(4)
C(26)–O(4)–P(1)	125.5(3)	C(31)–C(26)–O(4)	118.3(4)
C(34)–O(5)–P(1)	124.0(3)	C(31)–C(26)–C(27)	123.3(5)
N(7)–C(6)–S(3)	119.1(4)	C(28)–C(27)–C(26)	117.3(5)
N(8)–C(6)–S(3)	116.7(4)	C(33)–C(27)–C(26)	122.6(5)
N(8)–C(6)–N(7)	124.0(5)	C(33)–C(27)–C(28)	120.0(5)
C(14)–N(7)–C(6)	127.9(5)	C(29)–C(28)–C(27)	120.5(5)
C(20)–N(8)–C(6)	126.3(4)	C(30)–C(29)–C(28)	121.3(6)
F(11)–B(9)–F(10)	111.5(6)	C(31)–C(30)–C(29)	120.4(5)
F(12)–B(9)–F(10)	109.8(6)	C(30)–C(31)–C(26)	117.0(5)
F(12)–B(9)–F(11)	111.4(6)	C(32)–C(31)–C(26)	123.1(5)
F(13)–B(9)–F(10)	111.5(6)	C(32)–C(31)–C(30)	119.8(5)
F(13)–B(9)–F(11)	110.5(6)	C(35)–C(34)–O(5)	118.3(4)
F(13)–B(9)–F(12)	101.8(6)	C(39)–C(34)–O(5)	117.9(4)
C(15)–C(14)–N(7)	109.3(5)	C(39)–C(34)–C(35)	123.6(5)
C(19)–C(14)–N(7)	110.7(5)	C(36)–C(35)–C(34)	117.6(5)
C(19)–C(14)–C(15)	110.6(5)	C(41)–C(35)–C(34)	123.8(5)
C(16)–C(15)–C(14)	110.5(5)	C(41)–C(35)–C(36)	118.5(5)
C(17)–C(16)–C(15)	110.8(5)	C(37)–C(36)–C(35)	119.7(6)
C(18)–C(17)–C(16)	112.0(5)	C(38)–C(37)–C(36)	121.0(6)
C(19)–C(18)–C(17)	111.5(6)	C(39)–C(38)–C(37)	120.7(6)
C(18)–C(19)–C(14)	111.3(5)	C(38)–C(39)–C(34)	116.9(5)
C(21)–C(20)–N(8)	108.2(5)	C(40)–C(39)–C(34)	122.6(5)
C(25)–C(20)–N(8)	109.7(5)	C(40)–C(39)–C(38)	120.2(5)

(a) The bond lengths C(6)–N(7) and C(6)–N(8) are equal. This is in agreement with the isouronium structure of (5). Stabilisation by protonation of (3) makes its isolation possible but at the same time rules out any information being obtained on the structure of the kinetic product.

(b) Atoms H(7), N(7), C(8), N(8), and H(8) are located in a plane which forms an angle of 77° with the plane C(6)–S(3)–P. This angle is responsible for the difference in distance between N(7)–P and N(8)–P (respectively, 3·896 and 3·490 Å), which is probably due to crystal packing forces.

(c) In the isolated intermediate, the bonds H(7)–N(7) and H(8)–N(8) are antiperiplanar. This configuration

corresponds to the more stable form, which is in agreement with previous n.m.r. studies on amidinium ions which are less crowded than compound (5).[18] The isomerization barrier for these amidinium lies between 7 and 12 kcal mol^{-1} and a higher value would be expected for an isouronium ion. Thus the δ_P 68 p.p.m. peak in the spectrum mentioned previously, which is very close to the peak of the isolated compound (5) and only observed at low temperature, probably corresponds to an isomer of (5a) such as (5b) (Scheme 3), since the three bulky substituents (the two cyclohexyl and thiophosphoric group) cannot easily adopt a *cis–cis* configuration. However, it is not possible to infer from this structure the stereochemistry of the addition of ester (2a) to a carbodi-imide, because the isomerisation barrier of the latter is small (*ca.* 7 kcal mol^{-1}).[19]



SCHEME 3

In fact, even though the two forms are in equilibrium, (5a) is the only stable isomer according to n.m.r. analysis, from which the rearrangement to give (4) occurs. This reaction does not require an isomerisation about a C–N bond since as pointed out earlier, the two nitrogen atoms occupy a quasi-symmetric position with regard to phosphorus.

Intramolecular Rearrangement of (5a) *to the* N-*Phosphorylated Compound* (4).—This direct reaction is confirmed by the kinetic study of (4), carried out by following changes in the n.m.r. spectrum of (10b) (at -10 ± 0.5 °C). These are caused by the decrease in the peaks due to the intermediate (5a) and its counterion $(RO)_2P(S)S^-$, and the increase in the signal due to compound (4). The reactions are first order and the observed rate constants are: disappearance of $(RO)_2P(S)S^-$ (δ_P 104 p.p.m.), k 3.13×10^{-4} s^{-1}; disappearance of (5a) (δ_P 65 p.p.m.), k 3.03×10^{-4} s^{-1}; formation of (4) (δ_P 56.5 p.p.m.), k 3.05×10^{-4} s^{-1}. These values are identical within experimental error which confirms that isomerization of (5) into (4) proceeds under these experimental conditions without any kinetically significant intermediate being formed.

Conclusions.—The present study allows the structure of the intermediate in the addition reactions of phosphoric esters to carbodi-imides to be specified. The different factors which enable isolation of this intermediate, and therefore control its conversion into either pyrophosphate or rearranged product, are the following. For dithiophosphoric esters $(RO)_2P(S)SH$, the intermediate formed in reaction with DCCD is efficiently stabilized by protonation if an excess of acid is present. Experiments carried out with less basic carbodi-imides (*N*-*p*-nitrophenyl-*N*′-isopropylcarbodi-imide and di-*p*-tolylcarbodi-imide) show this factor to be important. Despite the presence of less nucleophilic nitrogen atoms, the intermediates formed with these carbodi-imides rearrange at low temperature because they are only partially protonated. Steric hindrance at the phosphorus atom slows down intramolecular rearrangement (reaction c, Scheme 2) and also pyrophosphate formation. For instance, for phosphoric diesters $(RO)_2P(O)OH$, the yield of pyrophosphate under identical conditions decreases dramatically with the size of the groups involved:

R	$C_6H_5CH_2$ [20]	2,6-$(CH_3)_2C_6H_3$	2,6-Pr^i_2-C_6H_3
Yield (%) of pyrophosphate	90 in 0.5 h	45 in 2 h	0 after 24 h

The present results give information about the reactivity of oxy- *versus* thio-phosphoric esters in their reaction with carbodi-imides. For $(RO)_2P(O)OH$ compounds, an excess of acid slows down step c of Scheme 2, the intramolecular rearrangement, but promotes pyrophosphate formation (in the absence of steric hindrance). However, thiophosphoric esters behave differently since they do not lead to the pyrothiophosphate. The fact that in this case the intermediate can be isolated implies that the nucleophile $(RO)_2P(S)S^-$ (which is in higher concentration than its oxygen analogue since the dithioester is completely dissociated), is not reactive enough for attack on the intermediate to occur, probably due to its more delocalized charge. Formation of the pyro(oxy- or thio-)phosphate is therefore controlled by the nucleophilic reactivity of the intermediate. In addition this comparison permits an explanation of the slower intramolecular rearrangements observed with sulphur compounds than with oxygen compounds. The through-space distance between N(8) and phosphorus calculated from *X*-ray results is 3.49 Å. The same calculation for a C–O–P system with bond lengths equal to 1.43 and 1.60 Å for C–O and P–O, respectively [21] gives a through-space distance of 2.34 Å, with the same bond angles. This is close to the sum of the van der Waals radii. Therefore the faster rearrangements observed with oxygen derivatives are clearly explicable on this basis.

Finally the reactions investigated provide a model for *O*-phosphobiotin, the relevance of which as a tool for elucidation of the mechanism of action of biotin is currently under investigation in this laboratory.

EXPERIMENTAL

M.p.s (Kofler) are uncorrected. I.r. spectra were run on a Beckman IR 20A instrument, ^1H and ^{31}P n.m.r. spectra on a 90 MHz Fourier transform Brüker instrument (for phosphorus, pulse 3.5 μs, time delay 1 s, acquisition time 0.33 s, chemical shifts are relative to 85% H_3PO_4 with positive values to low field of the reference). Spectra were decoupled with hydrogen and locked on deuterium.

Reactants.—DCCD (Aldrich) was used without further purification.

N-p-Nitrophenyl-*N'*-isopropylcarbodi-imide was synthesized by dehydration of the corresponding urea according to the method of Appel *et al.*[22]

Syntheses of (9a *and* b).—These were carried out according to Scheme 4, as used for phosphoric diesters.[23,24] The

$$2\ ROH + POCl_3 + 2\ C_5H_5N \xrightarrow[(a)]{i} (RO)_2P(O)Cl + 2\ C_5H_5\overset{+}{N}HCl^-$$

$$(RO)_2P(O)Cl \xrightarrow[(b)]{ii,iii} (RO)_2P(O)OH$$

SCHEME 4 Conditions: i, room temperature 2—12 h; ii, 10% aqueous NaOH; iii, 10% aqueous HCl to neutral pH

hydrolysis step (b) is critical since this can lead to a mixture of mono- and di-ester. Therefore for (9a) the intermediate chloridate obtained in quantitative yield (3 h) was spread on a Petri box and left 5 days in a desiccator containing a saturated aqueous potassium hydroxide solution.[24] Compound (9a) was separated from unchanged $(RO)_2P(O)Cl$ by recrystallisation from 2 : 1 n-hexane–chloroform in 50% yield, m.p. 138—140 °C (lit.,[25] 141—142 °C), *m/e* 306 (M^+), 292, 276, 201, and 193, δ_P (0.33M, dichloromethane) −8.42 p.p.m. (Found: C, 62.1; H, 6.25; P, 9.8. Calc. for $C_{16}H_{19}O_4P$: C, 62.75; H, 6.2; P, 10.15%). For (9b), after 12 h reaction for step (a), hydrolysis was carried out according to step (b). The product was extracted with chloroform; evaporation of chloroform led to a single product which became crystalline on addition of hexane. Fractional crystallisation (n-hexane–chloroform 2 : 1) gives the product in 20% yield, m.p. 172—173 °C (lit.,[25] 155—159 °C), *m/e* 418 (M^+), 375, 333, 268, 161, and 134, δ_P (0.27M, chloroform) −11.2 p.p.m. (Found: C, 69.1; H, 8.3; P, 6.65. Calc. for $C_{24}H_{35}O_4P$: C, 68.9; H, 8.35; P, 7.4%).

Syntheses of (10a *and* b).—These were carried out by the method of Flechter and Hamilton.[26] Compound (10a) was obtained in 100% yield and recrystallized from n-hexane–benzene (1 : 1), m.p. 114 °C, *m/e* 338 (M^+), δ_P (0.25M, dichloromethane) 76.8 p.p.m. (Found: C, 56.7; H, 5.7; P, 9.2; O, 9.6; S, 18.7. Calc. for $C_{16}H_{19}O_2PS_2$: C, 56.8; H, 5.65; P, 9.15; O, 9.45; S, 18.95%). Compound (10b) had δ_P (0.25M, ether) 77.1 p.p.m.

Reactions with Dicyclohexylcarbodi-imide.—Samples were prepared by mixing a solution at −80 °C containing diester (0.5 mmol) in dichloromethane (1.5 ml) to a solution at −80 °C of 0.5 mmol (molar ratio 1) or 0.25 mmol (molar ratio 2) of DCCD in dichloromethane (0.5 ml). In each case the solution was homogeneous. The reaction was followed by ^{31}P n.m.r. during a temperature rise to 20 °C. The relative percentages of the products (Table 1) were calculated from the integrated intensities of the phosphorus atom signals. For diesters (10a and b) the i.r. spectrum of the solution at room temperature shows an absorption at 1 640 cm^{-1} (C=N) (only for molar ratio 2).

Synthesis of [*Bis*-(2,6-*dimethylphenyl*)*thiophosphoro*]-NN'-*dicyclohexylisothiouronium Tetrafluoroborate* (5a).—To a solution of DCCD (2.5 mmol) in ether (2.5 ml) at − 80 °C was added a solution (cooled to −80 °C) of ester (10a) (2.5 mmol) in ether (7.5 ml). The homogeneous solution was kept at this temperature for 5 min with stirring before adding tetrafluoroboric acid (excess) in ether. After 10 min the solution was slowly warmed to room temperature. The precipitate formed was separated, washed with ether, and dried (1.25 g, 79%), m.p. 195 °C (from ether), *m/e* 544 ($M^+ - BF_4^-$), 338, 305, 273, 240, 206, 138, 163, and 122, δ_P (saturated solution in CH_2Cl_2) 67 p.p.m., $\nu_{max.}$ (2%, KBr) 3 240 (NH) and 1 640 (C=N) cm^{-1} (Found: C, 55.25; H, 6.75; N, 4.25, P, 5.15; S, 9.9; B, 2.15; F, 12.1. $C_{29}H_{42}BF_4N_2O_2PS_2$ requires C, 55.05; H, 6.7; N, 4.45; P, 4.9; S, 10.15; B, 1.7; F, 12.0%.)

Synthesis of N-[*Bis*-(2,6-*dimethylphenoxy*)*thiophosphinoyl*]-NN'-*dicyclohexylurea.*—A solution of (10a) (2.5 mmol) in ether (6 ml) was added with stirring to a solution of DCCD (2.5 mmol) in ether (2.5 ml) and kept at room temperature for 30 min. A precipitate is formed, collected by filtration and washed with ether (1 g, 73%), m.p. 141 °C (from ether), *m/e* 544 (M^+), 338, 305, 240, 206, and 121, δ_P (0.25M, ether) 56.9 p.p.m. (d, $^3J_{HCNP}$ 12 Hz), $\nu_{max.}$ (2%, KBr) 3 360 (N H) and 1 120 (C=S) cm^{-1} (no bond at 1640 cm^{-1}) (Found: C, 64.15; H, 7.7; N, 5.1; O, 6.1; P, 5.7; S, 11.95. $C_{29}H_{41}N_2O_2PS_2$ requires C, 64.0; H, 7.55; N, 5.15; P, 5.7; S, 11.8%).

Kinetic Measurements.—Reactions were followed by ^{31}P n.m.r. at constant temperature (−10 ± 0.5 °C) for the addition reactions at the following reactant concentrations: (10b) 0.25M, DCCD 0.25M. Data were analysed by the VAO4A program, the best fit being obtained by an exponential equation, then by a least-squares Guggenheim program. The rate constants are given in the text. The estimated error is ±5% for the three values given.

Crystallographic Measurements for (5).—Intensities were measured by a Syntex P 2_1 automated diffractomer. Data were corrected for Lorentz and polarization factors, but not for absorption which is negligible. Of the 4 675 reflexions with 2 θ less than or equal to 47°, 3 144 having intensities ≥2.5 σ(*I*) were employed in the subsequent structure analysis and refinement. The structure was solved by direct methods using the MULTAN 80 programs.[27] All hydrogen atoms were located from a difference synthesis after initial full-matrix least-squares refinement of the C, N, O, S, B, F, and P parameters. In the final round of calculations all positional parameters except those of the hydrogen atoms were refined. The hydrogen atoms were allowed isotropic thermal parameters and the remaining atoms allowed to vibrate anisotropically. At convergence, *R* was 0.055. The weight used was $W = 2.1042/\sigma^2(F) + 0.000\ 62(F^2)$ using the SHELX programs.[28] Final co-ordinates, anisotropic thermal parameters and structure factors are given in Supplementary Publication No. SUP 23157 (18 pp.).*

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* For details of Supplementary Publications see Notice to Authors No. 7 in *J. Chem. Soc., Perkin Trans. 2*, 1980, Index Issue.

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