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STUDIES ON REACTIONS OF THE N-PHOSPHONIUM SALTS OF PYRIDINES—VII

PREPARATION OF PEPTIDES AND ACTIVE ESTERS OF AMINO ACIDS BY MEANS OF DIPHENYL AND TRIPHENYL PHOSPHITES IN THE PRESENCE OF TERTIARY AMINES

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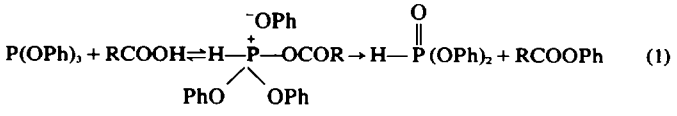
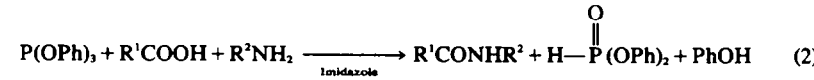
Abstract—Peptides and active esters of amino acids were produced in high yields from carboxyl and amino or hydroxyl components in pyridine with an equivalent amount of diphenyl phosphite or half an equivalent amount of triphenyl phosphite and tertiary amines. Condensation reactions competed with the reaction with a phenoxy group of the phosphite to produce the phenyl ester and were governed by the tertiary amine employed in the reaction. The reactions are assumed to proceed via the N-phosphonium salts of pyridines, similar to those obtained by the oxidation of phosphorus compounds with mercuric salts in pyridine.

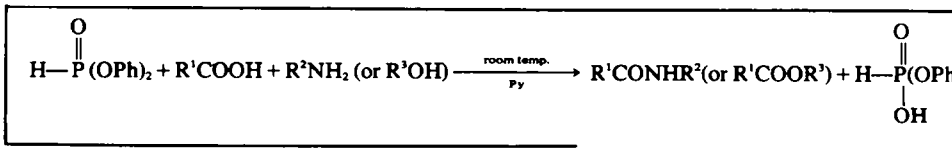
Triphenyl phosphite has been shown to react with carboxylic acids in the presence of pyridine to afford the corresponding phenyl esters,[1] and the reaction has been proposed to proceed via an acyloxy phosphonium salt (Eq 1).[2] Mitin *et al* suggest that the phosphite promotes a coupling reaction between carboxylic acids and amines in the presence of imidazole (Eq 2), and have successfully used the reaction for peptide synthesis.[3]



$$P(OPh)_3 + RCOOH \rightleftharpoons H-\overset{\overset{-OPh}{|}}{\overset{+}{P}}(PhO)(OPh)-OCOR \rightarrow H-\overset{\overset{O}{\|}}{P}(OPh)_2 + RCOOPh \quad (1)$$



$$P(OPh)_3 + R^1COOH + R^2NH_2 \xrightarrow[\text{Imidazole}]{} R^1CONHR^2 + H-\overset{\overset{O}{\|}}{P}(OPh)_2 + PhOH \quad (2)$$



$$H-\overset{\overset{O}{\|}}{P}(OPh)_2 + R^1COOH + R^2NH_2 \text{ (or } R^3OH) \xrightarrow[Py]{\text{room temp.}} R^1CONHR^2 \text{(or } R^1COOR^3) + H-\overset{\overset{O}{\|}}{\underset{\underset{OH}{|}}{P}}(OPh) + PhOH \quad (3)$$

However, these papers have referred neither to a triphenyl phosphite mediated coupling reaction between carboxylic acids and hydroxyl compounds nor to further coupling reactions with the resulting diphenyl phosphite.

In our previous reports of this series on the reactions of the N-phosphonium salts of pyridines, we have described a convenient method for the preparation of peptides via the phosphonium salts, given by the oxidation of phosphorous acid and its esters with mercuric salts and halogens in pyridines.[4] In the course of our studies on the application of an electrolytic process instead of chemical oxidation for the oxidation of the phosphorous compounds, we have found that the coupling reactions between carboxyl and amino or hydroxyl components are accomplished without oxidation, when diphenyl phosphite is used in pyridine (Eq 3).[5]

In a precise study on the rate of production of phenyl acetate in the reaction with triphenyl phosphite (Eq 1, R = CH_3) by means of gas chromatography, we found that the rate was very slow in pyridine, so that amidation proceeded preferentially in the presence of amino component, giving the corresponding amide and diphenyl phosphite in excellent yield. Hence, we expected that only half an equivalent of triphenyl phosphite might be suffi-

cient for the preparation of both peptides and active esters of amino acids in pyridine, since the diphenyl phosphite resulting from triphenyl phosphite could bring about further reactions as described above (Eq 4).

roxyl components in the place of the amino components used in peptide synthesis. These results are given in Tables 1 and 2.

The effect of tertiary amines upon the amide formation (Eq 3) was investigated in the reaction in

$$1/2\ P(OPh)_3 + R^1COOH + R^2NH_2\ (\text{or } R^3OH) \xrightarrow[Py]{} R^1CONHR^2\ (\text{or } R^1COOR^3) + 1/2\ H{-}\overset{\overset{O}{\|}}{\underset{\underset{OH}{|}}{P}}(OPh) + PhOH \quad (4)$$

This paper presents detailed results of these reactions brought about by phosphorous acid and its esters, especially diphenyl and triphenyl phosphites, in pyridine in the absence of oxidizing agents.

RESULTS AND DISCUSSION

The coupling reactions using diphenyl and triphenyl phosphites were carried out at room temperature to 40°C by simultaneously mixing the reactants in pyridine according to Eqs 3 and 4.[5] Several peptides were obtained in high yields, together with nearly quantitative yields of phenol, detected by gas chromatography. Similarly, active esters of amino acids were synthesized using hydacetonitrile using two equivalents of the tertiaryamines (Table 3). Of the amines examined, pyridine was the most effective for the reaction with diphenyl phosphite. The yield decreased with an increase in the basicity among pyridine derivatives. The lower yields obtained with 2-methyl and 2,6-dimethyl pyridines, less than a half of that by pyridine may reflect a steric effect. Although reaction in imidazole unexpectedly gave a poor yield, in the reaction with triphenyl phosphite, imidazole was significantly effective, being even more useful than pyridine and its derivatives. The different behavior of the amines in the reactions with these phosphites is difficult to understand. Triethylamine retarded both reactions.

Table 1. Peptide synthesis by means of di- and tri-phenyl phosphites in pyridine

Peptides	Diphenyl phosphite[a] Conditions Yield, %	Temp, °C	Time, h	Triphenyl[b] phosphite Yield, %	m.p. (°C)	$[\alpha]_D$
Z-Gly-Gly-OEt	91	r.t.	0·5	92(94)[c]	80	
Z-Phe-Gly-OEt(L)	65	r.t.	0·5	85	108–109	−17·7°(c = 5, EtOH)
Z-Phe-Gly-OEy(L)	90	40	6	—	—	
Z-Gly-Tyr-OEt(L)	60	r.t.	0·5	—	125–126	+19·8°(c = 5, EtOH)
Z-Gly-Tyr-OEt(L)	88	40	6	96	—	
Z-Glu(NH_2)-Gly-OEt(L)	85	40	6	—	168–170	−6·5°(c = 1, DMF)
Z-Met-Gly-OEt(DL)	91	40	6	95	72–73	

[a] The reaction was carried out using an equivalent of diphenyl phosphite.
[b] The reaction was carried out at 40°C for 12 h using half an equivalent of triphenyl phosphite.
[c] The yield obtained using an equivalent of triphenyl phosphite.

Table 2. Preparation of active esters of amino acids using di- and tri-phenyl phosphites in pyridine

Active esters	Yield, % Diphenyl phosphite	Triphenyl phosphite	m.p., °C[a]
Z-Gly-O-C_6H_4-NO_2(p)	77	83	128
Z-Gly-O-C_6H_4-NO_2(p)	69	70	121
Z-Gly-S-C_6H_5	89	64	71–72
Z-Phe-O-C_6H_4-NO_2(p)(L)	73	73	125–126
Z-Glu(NH_2)-O-C_6H_4-NO_2(p)(L)	45	48	142–143

[a] Melting point after one crystallization.

Table 3. Effect of tertiary amines on the reaction of Z-glycine with aniline using di- and tri-phenyl phosphites[a]

Tertiary amines	pKa	Yield (%) of Z-glycinanilide	
		Diphenyl phosphite	Triphenyl phosphite
Pyridine	5·23	79	65
3-Methylpyridine	5·52	59	63
4-Methylpyridine	6·02	59	65
2-Methylpyridine	5·97	38	40
2,6-Dimethylpyridine	6·99	26	37
Imidazole	7·12	38	85
Triethylamine	10·87	5	14
None	—	17	23

[a]The reaction was carried out at ambient temperature for 2 h in acetonitrile using two equivalents of the amines.

As suggested in a previous paper,[5] diphenyl phosphite and tertiary amines are involved stoichiometrically in the reactions. The effect of the amounts of pyridine and diphenyl phosphite was investigated in detail in the reaction of benzyloxycarbonyl-glycine(Z-glycine) and aniline in acetonitrile (Fig 1). The yield of Z-glycinanilide increased with increasing amounts of these components, reaching asymptotic values when the molar ratio of pyridine to the carboxyl component was 2 and that of diphenyl phosphite was 1.

A similar stoichiometric relationship between triphenyl phosphite and pyridine was observed in the reaction with the phosphite (Fig 2).

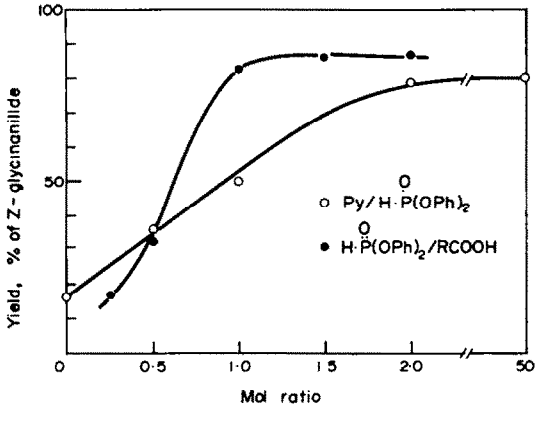


Fig. 1.

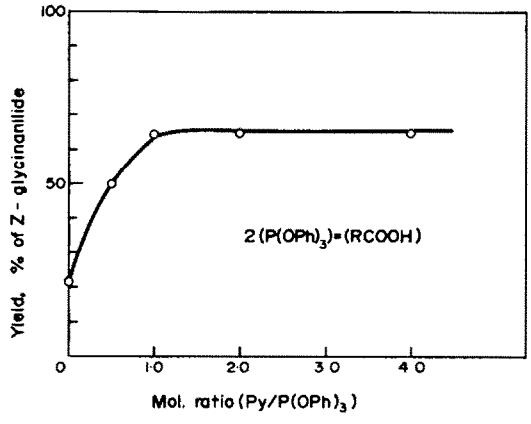
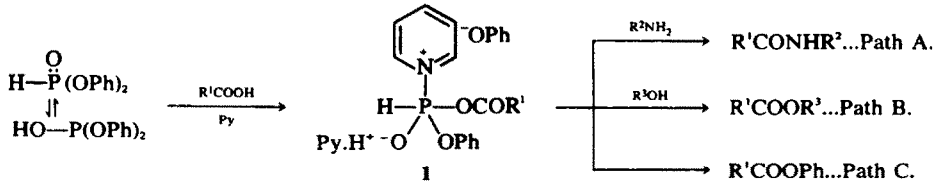
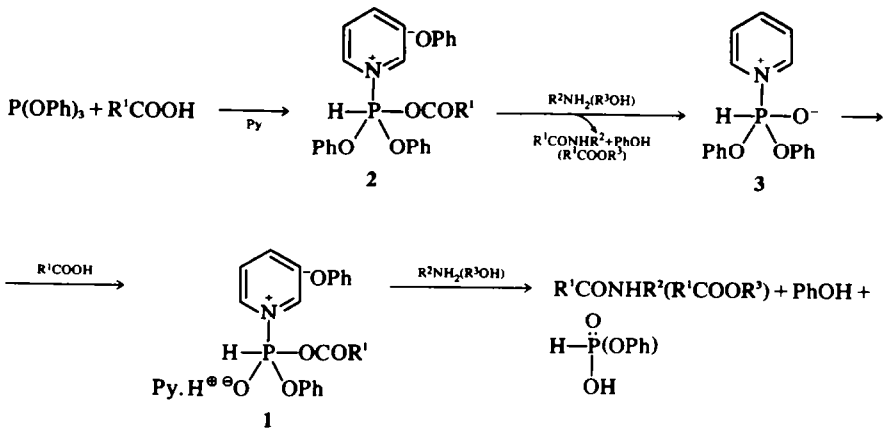


Fig. 2.

These experimental results suggest that the reactions with diphenyl phosphite proceed as follows. Acyloxy N-phosphonium salt of pyridine **1** is formed by reaction of diphenyl phosphite with a carboxylic acid, involving release of a phenolate anion from the phosphite. This salt undergoes two types of reaction, amide synthesis (Path A) or active ester synthesis (Path B) by intermolecular reaction with the incoming amino or hydroxyl components, and phenyl ester formation by intramolecular reaction with the phenolate anion released from the phosphorus atom (Path C). The two competitive reactions (Path A and C, or B and C)



SCHEME 1



SCHEME 2

are governed by the tertiary amine employed in the reaction. Actually, in a separate reaction of diphenyl phosphite with acetic acid in the absence of an amino component, phenyl acetate was produced, as it was also from triphenyl phosphite, and its rate of production was more rapid in the presence of imidazole than in the presence of pyridine.

The reactions with triphenyl phosphite in pyridine can be explained similarly to those with diphenyl phosphite (Scheme 2). Acyloxy N-phosphonium salt of pyridine **2** initially formed is converted into **3**, the amide (or the ester) and phenol by aminolysis (or alcoholysis). **3** then reacts with another carboxylic acid to yield **1**, which gives the amide (or the ester) together with phenol and the monophenyl ester of phosphorous acid as discussed in Scheme 1. Aminolysis of **2** as well as **1** may compete with the intramolecular phenyl ester formation which is affected by the tertiary amine used in the reaction, as discussed in Scheme 1.

Phosphorous acid and its diisopropyl ester were tried in the reaction of Z-glycine and ethyl glycinate in pyridine, but gave only 8 and 46% yields of ethyl Z-glycylglycinate even after refluxing for 1 h. It may be that the reaction with the diisopropyl ester does not proceed as shown in Scheme 1, since the ester cannot form an intermediate like **1** because of the low nucleophilicity of the isopropoxide group.

EXPERIMENTAL

Preparation of peptides and active esters by means of diphenyl and triphenyl phosphites in pyridine. A mixture of equimolar amounts of a benzyloxycarbonyl-amino acid (12·5 mmole), an amino acid ester hydrochloride and diphenyl phosphite in 40 ml of pyridine was kept at room temperature for 30 min or at 40°C for 12 h. The reaction mixture was worked up by the previously reported procedure[5] to give peptide. Similarly, the active ester was obtained by the reaction of diphenyl phosphite (19 mmole), hydroxyl compound (12·5 mmole) and benzyloxycarbonyl-amino acid (12·5 mmole) in 40 ml of pyridine at 40°C for 12 h. In a similar fashion, peptides and active esters of amino acids were produced using half an equivalent of triphenyl phosphite (6·25 mmole) in the place of diphenyl phosphite. The reactions were carried out varying amounts of tertiary amines and phosphites at ambient temperature for 2 h in 50 ml of acetonitrile.

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